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EFFECTS OF DIFFERENT SOIL TILLAGE METHODS, PHOSPHORUS FERTILIZER DOSES AND BACTERIA INOCULATION ON YIELD AND YIELD COMPONENTS IN CHICKPEA *(Cicer arietinum* L.)

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ABSTRACT

This study was conducted to determine the effect of different soil tillage methods, phosphorus levels and bacteries on the yield and yield components in chickpea under Muğla/Turkey conditions in 2017 and 2018. Chickpea variety Inci was used as materials in this study. The study was conducted at split-split plot design with the three replication. Soil tillage methods was in main plots, phosphorus doses in subplots and bacteria application in sub-sub plots. According to finding of this study, reduced tillage system for chickpea production was the most efficient tool for obtaining the high yield performance. It is seen that phosphorus fertilizer applications cause an increase in yield in chickpea. In general, the application of rhizobia and phosphate solubilizing bacteria separately supported the yield increase in the plant. As a result, only rhizobia application, 60 kg ha^{-1} P_2O_5 and reduced tillage gave the highest yield.

Keywords: fertilization, bacterium, tillage, chickpea.

INTRODUCTION

Chickpea (*Cicer arietinum* L.) is the most produced legume plant in Turkey (Anonymous, 2020). Chickpea cultivation area is 517.785 ha and its production is 630.000 tons in Turkey (FAO, 2019). Chickpea originated in the Fertile Crescent, which borders the southeastern regions of Turkey, and spread west and south via the historically called Silk Route (Behmand et al., 2019).

Tillage regulates crop performance as it affects nutrient availability, weed growth, root growth and water use efficiency (Das et al., 2018). Tillage has some short-term benefits such as suitable soil conditions for good crop output, greater crop yield, better nutrient availability, strong seedling growth (Six et al., 1999). However, in the long term, tillage causes soil incrustation and compaction, accelerates soil erosion, increases mineralization of soil organic matter, degrades and breaks up soil aggregates, and exacerbates the loss of plant nutrients and soil organic carbon (Yadav et al., 2021). The main soil tillage system in the Turkey is the conventional plough system. Conventional intensive tillage is a widespread soil management practice that controls weeds and promotes nutrient mineralization at the expense of a degraded soil structure and soil carbon loss (Boogar et al., 2021). However, due to the costly nature of the conventional tillage system, cultivation areas using reduced tillage (RT) and no tillage (NT) systems are increasing. Reduced tillage system promotes soil structure improvement, minimizes the risk of soil erosion, increases soil organic carbon content, reduces soil temperature fluctuations, conserves soil water (Busari et al., 2015).

Food production has been increased in the world since green revolution. Consequently, the over application of chemical fertilizers (Canfield et al., 2010) contribute further to greenhouse gas emissions (Smith et al., 2013) and climate change (Richardson et al., 2012). Plant growth promoting bacteria (PGPR) are natural resources that colonize the roots of plants and directly and indirectly promote

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growth and yield (Afzal and Bano, 2008). The mechanisms by which PGPR stimulate plant growth involve the availability of nutrients originating from genetic processes, such as biological nitrogen fixation and phosphate solubilization, stress alleviation through the modulation of ACC deaminase expression, and production of phytohormones and siderophores, among several others (Souza et al., 2015).

Due to the symbiosis and the resulting biological nitrogen fixation, the application of nitrogen fertilizers has been avoided (Ferguson et al., 2010). Rhizobia, a PGPR, refers to bacterial species that can interact with the roots of legumes and induce the formation of structures called nodules (Lindstrom and Mousavi, 2019).

Plants need phosphorus especially for cell division in the early stages of growth, and for seed formation and increase in seed weight during maturity (Lafond et al., 2008). The availability of phosphate in soil is generally limited by fixation reactions that convert the monophosphate ion into various insoluble forms (Meena, 2010). Given the limited resources of P and the serious ecological and economic consequences of polluting the environment (Chardon and Withers, 2003), careful use of P is imperative. Therefore, application of bacteria's efficient of mobilizing P is considered to be a potential bio fertilizer for sustainable agriculture in a cost-effective and ecofriendly manner (Henri et al., 2008; Park et al., 2010). Phosphate solubilizing bacteries are play a important role in P solubilization by producing organic acids.

In recent years, researchers have focused on inoculation with PGPR and Rhizobia, becoming the most popular approach to improve growth in legumes. PGPR may increase the efficiency of Rhizobia inoculation in legumes through the production of antibiotics, siderophore, and certain enzymes (Iqbal et al., 2012). Tillage practice can change soil properties (Alvarez and Steinbach, 2009). The aim was to evaluate the potential of PGPR inoculation for improvement of chickpea production under different tillage systems and phosphorus doses.

MATERIAL AND METHODS

The field experiment was conducted during the 2017 and 2018 at the fields of the Fethiye, Muğla, Turkey (36°34’ N; 29°5’ E, 146 m above sea level). Climatic datas are shown in Table 1. The total precipitation amount, which was 155.5 and 118.6 mm in the trial years, respectively, is 170.3 mm in the long term. Annual average temperature was 22.1°C in 2017 and 23.1°C in 2018. The analyzes of the soil in the trial areas are presented in Table 2.

The study was evaluated yield and yield components effects on the chickpea using two soil tillage methods (conventional and reduced), three phosphorus doses (control, 30 and 60 kg ha^{-1} P_2O_5) and four bacteria application (control, *Rhizobia*, phosphate solubilizing bacteria, *Rhizobia* + phosphate solubilizing bacteria). The experimental design was split split plot with three replicates. Soil tillage methods was in main plots, phosphorus doses in subplots and bacteria application in sub-sub plots.

Table 1. Climatic data of the research area

Months	Long Term			2017			2018		
	Temp. (°C)	Precip. (mm)	Humid. (%)	Temp. (°C)	Precip. (mm)	Humid. (%)	Temp. (°C)	Precip. (mm)	Humid. (%)
March	13.2	84.9	67.3	14.5	92.1	66.5	15.5	67.2	70.3
April	16.4	43.3	67.1	17.1	20.8	62.9	19.5	6.2	62.2
May	20.6	28.3	65.4	21.8	42.6	60.6	23.9	28.2	59.9
June	25.1	5.3	59.4	26.8	-	54.5	26.4	17.0	61.7
July	27.9	8.5	57.4	30.2	-	51.4	30.1	-	51.4
Total		170.3			155.5			118.6	
Mean	20.6		63.3	22.1		59.2	23.1		61.1

Table 2. The analyzes of the soil in the trial areas

Year	Depth	Texture	pH	Lime (%)	P (ppm)	K (me/100 g)	Organic matter (%)	Total salt	Zinc (ppm)
2017	0-30 cm	Sandy clay loam	7.4	7.6	13.9	83.9	2.6	0.11	2.0
2018	0-30 cm	Clay loam	7.9	6.53	12.2	109	2.16	0.04	0.10

Each sub-sub plot was 7.2 m^2 (4 m x 1.8 m) and chickpea was sown 30 cm row spacing and seeding rate was 48 seeds m^{-2}. İnci chickpea cultivar was used as research material. Chickpeas were sown by hand (26 March 2017 in the first year and 17 March 2018 in the second year). The first tillage was carried out in October in half of the main plots with moldboard ploughing (conventional tillage). Other half was tillaged with only rototiller (reduced tillage). Tillage depths for conventional tillage (CT) and reduced tillage (RT) were 25-30 and 8-10 cm, respectively. *Mesorhizobium ciceri* which is effective nitrogen-fixing and *Serratia odorifera* which is effective in phosphate solubilizing bacteria were used. Before sowing in rhizobia plots, seeds were inoculated into each plot using rhizobia culture prepared at a density of 10^6 cells/g (Vincent, 1970). The bacterial strain was developed as a pure culture in NB medium at 28°C and the bacterial suspension was inoculated to seeds as 10^8cfu/seed. Phosphate solubilizing bacteria were propaged in laboratory conditions and water with sugar was used for inoculation for both bacteria to provide that the bacterial culture sticked to the seeds. 21% ammonium sulphate fertilizer was applied to each plot on sowing time as 2 kg da^{-1} N. Triple super phosphate (43-45% P_2O_5) fertilizer as phosphorus fertilizer was applied on the sowing time at the above mentioned doses. Plots were hand-weeded when needed each year. The harvest time of chickpea was on 16 July 2017 and 6 July 2018 in the first and second years, respectively.

The pod number per plant, seed number per plant were evaluated on 10 randomly selected plants in each sub-sub plot. The nodule number per plant were determined in 10 plants taken from each sub-sub plot when flowering started. Each sub-sub plot was harvested, blended and biological yield (kg ha^{-1}), hundred kernel weight (g) and grain yield (kg ha^{-1}) were estimated.

The variance analysis was subjected to based on General Linear Model using the Statview package (SAS Institute, 1998). Means were compared by Tukey test.

RESULTS AND DISCUSSION

The effects of years, tillage and phosphorus doses were significant for all of the investigated characters except for hundred kernel weight. The effect of bacteria was significant for all of the investigated characters except for the seed number per plant and the hundred kernel weight (Table 3). While pod and seed number per plant were higher at conventional tillage, 0 kg ha^{-1} P_2O_5 and rhizobia + phosphate solubilizing plots, 30 kg ha^{-1} P_2O_5 showed the lower values same applications. Therefore tillage x phosphorus x bacteria interaction was significantly (Figure 1). Nodule number per plant showed higher performance in reduced tillage, 30 kg ha^{-1} P_2O_5 and rhizobia bacteria plots, same bacteria showed lowest performance conventional tillage and 0 kg ha^{-1} P_2O_5 plots. Therefore tillage x phosphorus x bacteria interaction was significantly (Figure 2). While the first year nodule number per plant was lower in all of the plots but the second year is higher. For this reason, year x phosphorus x bacteria interaction was significantly for nodule number per plant (Figure 2). In the second year, all bacteria applications showed high values in the reduced tillage plots, while all bacteria applications showed low values in the first year conventional tillage plots. For this reason year x tillage x bacteria interaction was significantly for nodule number per plant (Figure 3). While nodule number per plant was higher in second year at reduced tillage and 0 kg ha^{-1} P_2O_5, conventional tillage in first year showed the lower values same phosphorus doses. Therefore year x tillage x phosphorus interaction was significantly (Figure 3). In both years, all phosphorus doses showed high values in the reduced tillage plots, while they showed lower values in the conventional tillage plots. Therefore year x tillage x phosphorus interactions were significantly for biological yield and grain yield (Figure 4). While conventional tillage plots showed superior performance 60 kg ha^{-1} P_2O_5 plots in second year, same year and phosphorus doses showed lowest performance in reduced tillage plots. For this reason year x tillage x phosphorus interaction was significantly for hundred kernel weight (Figure 5).

All of the investigated characters were higher in the 2018 than the 2017 except for the hundred kernel weight (Table 3). Pod and seed number per plant, biological yield and grain yield were higher the second year than the first year. While the total precipitation was 155.5 mm in the first year, it was 118.6 mm in the second year (Figure 1). Total precipitation is higher in the first year. However, the rains in June in the second year caused the higher yield and yield component. The precipitation was less and the temperature was higher in March and April in the second year, when nodules were formed. Bacteria activity may have more effectively in this period because of precipitation is less and soil air is more suitable. Because soil temperature and weather are very important for bacteria to be effective (Gençtan, 2006). Therefore, the nodule number per plant is higher in the second year.

While the pod and seed number per plant are high in conventional tillage, nodule number per plant, biological yield and grain yield are higher in reduced tillage. Tillage methods did not affect the hundred kernel weight (Table 3). Weeds are better controlled and the soil's water holding capacity is higher in conventional tillage plots (Kasap and Dursun, 2013). Therefore, pod number per plant and seed number per plant may have been higher in conventional tillage plots. Altıkat (2013) reported that tillage methods significantly affect the pod number per plant. Nodule number per plant is higher in reduced tillage plots. Kombiok and Buah (2013) reported that tillage system and tillage depth influence the nodule number per plant in soybean plants. Reduced tillage decreased temperature fluctuations and it increased moisture. As a result, microbial activity can promote (Gençtan, 2006). While the biological yield is 5420 kg ha^{-1} in reduced tillage plots, it is 4700 kg ha^{-1} in conventional tillaged plots. While the grain yield 1930 kg ha^{-1} in reduced tillaged plots, it is 1650 kg ha^{-1} in conventional tillaged plots. High yield performances are usually are obtained due to increased water conservation and increased organic matter in reduced tillage. Guy and Cox (2002) stated that they found increases in the amount of yield in reduced tillage compared to conventional tillage. Hemmat and Eskandari (2004) reported that yield of chickpea in conservational tillage was 33% higher than conventional tillage.

Table 3. Effects of different tillage, phosphorus doses and bacteria on some traits of chickpea

	Pod number per plant	Seed number per plant	Nodule number per plant	Biological yield (kg ha^{-1})	Grain yield (kg ha^{-1})	Hundred kernel weight
2017	25,1 b	26,1 b	3,7 B	4990 b	1710 b	36.0
2018	30,0 a	29,3 a	7,1 A	5140 a	1870 a	35.7
Mean	27,5	27,7	5.4	5060	1790	35.9
Conventional	29,8 a	30,2 a	4.8 b	4700 B	1650 B	36.0
Reduced	25,3 b	25,1 b	6.0 a	5420 A	1930 A	35.7
Mean	27,5	27,7	5.4	5060	1790	35.9
0 kg ha^{-1} P_2O_5	30,0 A	29,9 A	5.8 A	4950 C	1760 b	35.9
30 kg ha^{-1} P_2O_5	24,2 B	24,3 B	5.4 B	5080 B	1790 b	35.9
60 kg ha^{-1} P_2O_5	28,3 A	28,7 A	5,0 C	5160 A	1830 a	35.8
Mean	27,5	27,7	5.4	5060	1790	35.9
Without inoc.	29,1 a	28,4	5.9 A	4980 C	1740 C	35.7
Rhizobia	26,5 b	27,3	4.9 C	5160 A	1860 A	35.6
Phosphate sol.	27,8 ab	27,9	5,3 B	5070 B	1810 B	35.8
Rhiz. + Pho. sol.	26,7 ab	26,9	5.5 B	5030 BC	1770 BC	36.4
Mean	27,5	27,7	5.4	5060	1790	35.9
General mean	27,5	27,7	5.4	5060	1790	35.9
Year	*	*	**	*	*	ns
Tillage	*	*	*	**	**	ns
Phosphorus do.	**	**	**	**	*	ns
Bacteria	*	ns	**	**	**	ns
Year x tillage	ns	ns	*	ns	ns	ns
Year x phosp.	ns	ns	**	ns	ns	ns
Tillage x phosp.	ns	ns	*	*	**	*
Ye. x til. x phosp.	ns	ns	**	*	*	*
Year x bacteria	ns	ns	**	ns	ns	*
Tillage x bacteria	ns	*	**	ns	*	ns
Phosp. x bacteria	**	**	**	ns	*	ns
Year x til. x bac.	ns	ns	**	ns	ns	ns
Ye. x pho. x bac.	ns	ns	**	ns	ns	ns
Til. x pho. x bac.	**	**	**	ns	ns	ns
Y. x til. x pho. x bac.	ns	ns	**	ns	ns	ns

ns: non-significant, *: $p \leq 0.05$, **: $p \leq 0.01$.

Pod, seed and nodule number per plant were higher in 0 kg ha^{-1} P_2O_5 plots. Yılmaz (2010) reported that he obtained the highest pods number per plant from the 0 kg ha^{-1} P_2O_5 plots. The researcher stated that climatic and soil conditions were effective in such results. The amount of phosphorus in the soil is sufficient in both years (Table 2). Pod, seed and nodule number per plant may not have responded to phosphorus fertilization due to sufficient phosphorus in the soil. Biological yield and grain yield were higher 60 kg ha^{-1} P_2O_5 plots. Phosphorus fertilization had a positive effect on biological yield and grain yield. Yagmur and Engin (2005) and Sumer and Erten (2022) reported that the biological yield and grain yield increased with increasing phosphorus doses. Phosphorus fertilization increases root development, and therefore nutrient intake increases. Thus, biological yield and grain yield improvement (Kacar and Katkat, 1999).

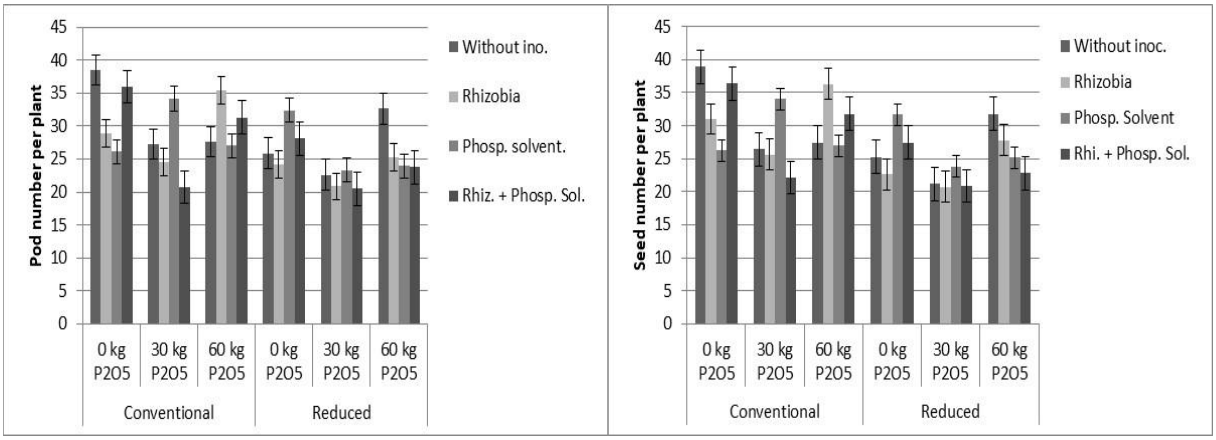

Figure 1. Pod number per plant and seed number per plant interactions of chickpea

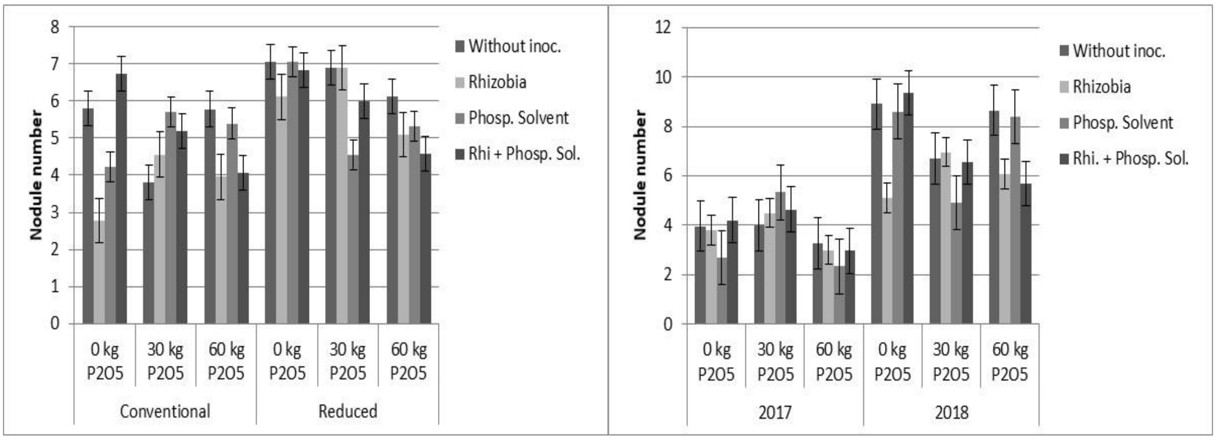

Figure 2. Nodule number per plant interactions of chickpea

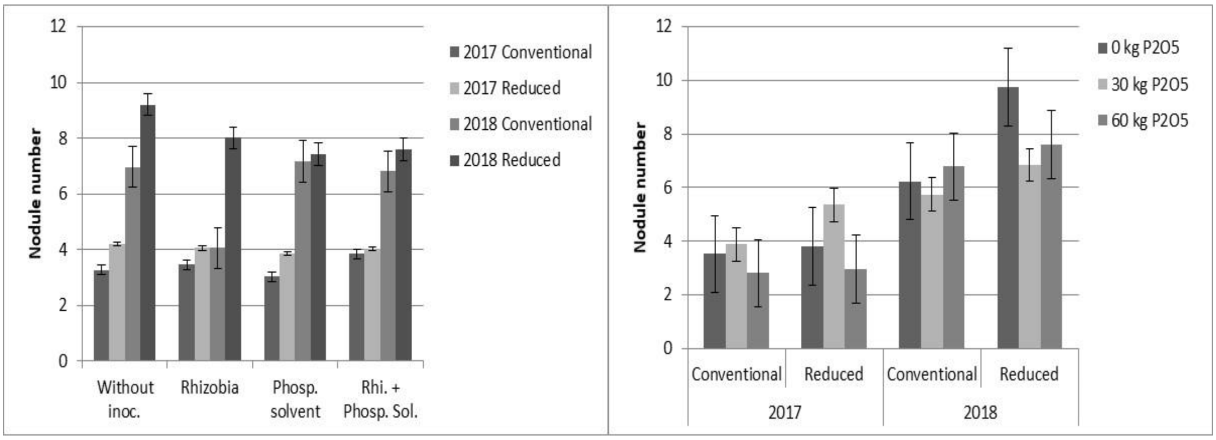

Figure 3. Nodule number per plant interactions of chickpea

Pod and nodule number per plant were higher without inoculation plots. Bacteria applications did not positively affect the pod and nodule number per plant. Sowing was done in March in both of year. The month of March, when nodules were formed, coincided with rainy period in both years (Table 1). Excess moisture negatively affects the formation of nodules and the functioning of bacteria. In excess moisture, bacteria are deprived of oxygen and die (Adak, 2021). In addition, if there is enough bacteria in the soil, there may be no reaction to bacteria inoculation. Consequently inoculation become ineffective. Özturan Akman (2017) reported that the effects of bacteria and nitrogen application on the nodule number per plant are insignificant. Biological yield and grain yield were higher rhizobia plots. Rhizobia plots were followed by phosphate solubilizing plots. The lowest biological yield and grain yield were determined in the control plots without bacteria. Kumar and Chandra (2008) reported that phosphate solubilizing bacteria convert the insoluble phosphorus in rock phosphate and soil into a form that plants can take. With the organic acids secreted by phosphate solubilizing bacteria, soil pH decreases and accordingly the availability of phosphorus increases (Çakmakcı, 2005). Kaçar et al. (2004) and Bulut (2013) reported that inoculation with rhizobia bacteria increased grain yield. Some researchers have reported that phosphate solubilizing bacteria increase grain yield (Khan et al., 2003; Şahin et al., 2004; Öztürk et al., 2003).

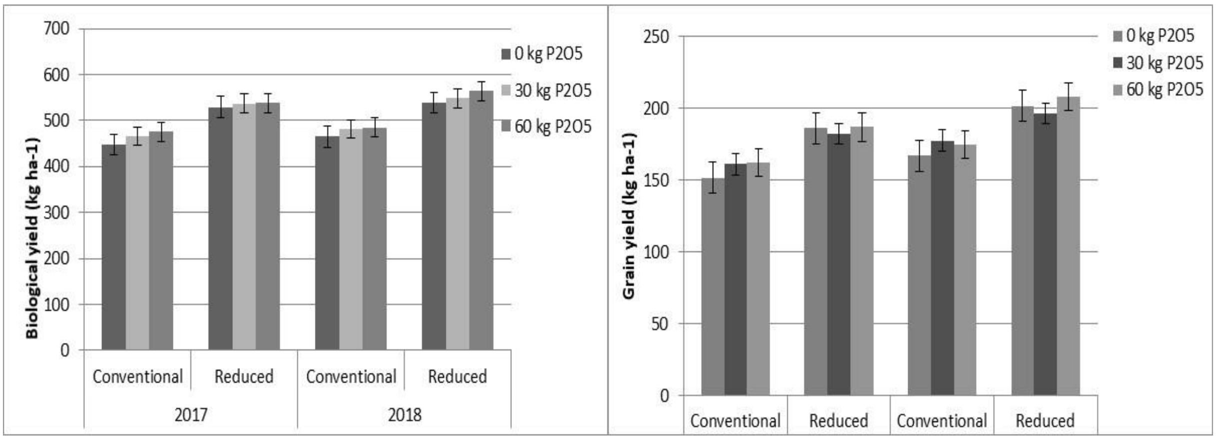

Figure 4. Biological yield and grain yield interactions of chickpea

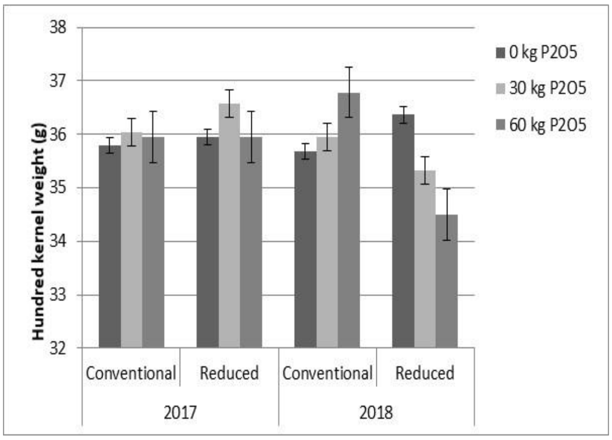

Figure 5. Hundred kernel weight interactions of chickpea

CONCLUSIONS

According to finding of this study, reduced tillage system for chickpea production was the most efficient tool for obtaining the high yield performance in Mediterranean zone. It is seen that phosphorus fertilizer applications cause an increase in yield in chickpea. In general, the application of rhizobia and phosphate solubilizing bacteria separately supported the yield increase in the plant. In the region where the research was conducted, chickpea cultivation has increased in recent years. Studies on chickpea plants in the region should be encouraged in order to prevent unnecessary fertilizer use and to get the highest yield. As a result, only rhizobia application, 60 kg ha^{-1} P_2O_5 and reduced tillage gave the highest yield in Mediterranean zone.

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