

Research Article

Effect of gypsum and cow manure on yield, proline content, and K/Na ratio of soybean genotypes under saline conditions

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Received 02 October 2017, Accepted 30 November 2017

Abstract: Gypsum and cow manure potential as ameliorant to increase crop production under salt stress or saline condition. This research aimed to learn the effect of gypsum and cow manure on the uptake of Na, K and the yield of soybean genotypes under saline condition. This research conducted in green house Jatikerto Experimental Farm Faculty of Agriculture, Brawijaya University, from June to September 2014. The research was arranged in a split plot design. The main plot was soybean genotypes consists of two saline susceptible varieties (G1 = Wilis and G2 = Tanggamus) and two saline tolerant genotypes (G3 = genotype IAC, 100/Bur//Malabar and G4 = genotype Argopuro//IAC, 100); sub plot was ameliorant application consists of A0 = without ameliorant; A1 = cow manure (20 t/ha); and A2 = gypsum (5 t/ha). The results of the research showed that Leaf Chlorophyll Index in susceptible varieties and tolerant genotypes were increased with ameliorant application. Accumulation of proline and K/Na ratio in susceptible varieties higher than tolerant genotypes. Ameliorant application on tolerant genotypes increased grain yield higher than susceptible varieties.

Keywords: *ameliorant, chlorophyll, K/Na ratio, proline, salinity, soybean*

To cite this article: Yamika, W.S.D., Aini, N., Setiawan, A. and Purwaningrahayu, R.D. 2018. Effect of gypsum and cow manure on yield, proline content, and K/Na ratio of soybean genotypes under saline conditions. *J. Degrade. Min. Land Manage.* 5(2): 1047-1053, DOI: 10.15243/jdmlm.2018.052.1047.

Introduction

The demand of soybean is increasing with the growing public awareness of plant-based protein foods. In 2013, soybean production was estimated at 807.57 thousand tons of dry beans, decreased by 35.58 thousand tons (4.22 percent) compared to 2012. The decline in soybean production is expected to occur due to low harvested area and a productivity (BPS, 2013). The ways to increase soybean production is done by expanding the land. Extension of this land is to utilize sub-optimal land or marginal land. In this case, the Government has programmed agricultural food extension conducted at sub-optimal land abandoned and unproductive, one saline soil. Saline soils are soils containing dissolved salts

that are large enough for plant growth, such as chloride or sulfate. The accumulation of salts in the top soil layer is usually affected by evapotranspiration or high salt content in ground water. Soil salinity has toxic effects and is able to increase the osmotic pressure that makes the roots of stunted plant growth. The high concentration of salt and exchangeable sodium is affected by the low organic matter in the soil that does not support crop growth and does not allow sustainable agriculture (Li-ping et al., 2015). Response of plant to salt stress is different for every variety or genotype (Akgun et al., 2011; Aini et al., 2012). Saline soils are often found in areas with a dry climate having rainfall of less than 500 mm per year with the electrical conductivity of > 4 mS/cm. In Indonesia, the total

area of saline land reaches 440,300 hectares, divided into 304,000 hectares of low saline and 140,300 hectares of saline (Rachman et al., 2007).

One of the strategies to overcome the shortage of domestic soybean is the use of saline soil for soybean cultivation. At present, most fertile lands are used for food crops such as rice and corn, yet saline land in Indonesia is quite extensive. Efforts to overcome the negative effects of saline land can be done by the approach of genetic and environmental modification. The Genetic approach that can be done is by the use of saline tolerant varieties. While environmental approach can be done by modification of soil environments. Environmental modification technology to decrease the negative effect of saline soil can be done by some alternatives. The first strategy is leaching the salt mineral by using good water irrigation to remove Na^+ and Cl^- from the top soil. Application of 6 t gypsum ha^{-1} in the saline soil can decrease Na-dd faster than dolomite dan Mangrove litter (Sasongko dan Warsito, 2003). The second alternative is application of soil ameliorant such as gypsum ($\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$) as a supplier of Ca^{2+} to offset Na^+ on exchange sites. The third alternative technique that has gained attention as a less expensive alternative is application of organic amendments including composts, cow manure and peat soil. They are effective in improving soil structure and aggregate stability (Roesmarkam and Yuwono, 2002; Jones, 2002). They can also improve a quality of other soil chemical and biological properties (Tejada et al., 2006).

The addition of gypsum and organic matter to the saline and sodic soils has a synergistic effect between the two antagonists especially for monovalent cations such as Na^+ (Mahmoodabadi et al., 2012). Mineral-organic fertilizer application consisting of a mixture of calcium sulphate, rice mill waste and humic acid to reclaiming saline soils (Ec: 8.9 dS/m) can improve the growth and yield of tomatoes. Plants fertilized with mineral-organic fertilizer had high levels of N^+ , K^+ and Ca^{2+} and low Na^+ on the leaf (Rady, 2012). The addition of cotton waste compost and chicken manure at a dose of 5 and 10 t/ha to saline soils had a positive effect on soil physical, chemical and biological properties, even after five years of ameliorant application (Tejada et al., 2006). The success of saline tolerant soybean varieties assembly is determined by the availability of resistant genes. Saline tolerant soybean varieties can be obtained through the identification of soybean germ plasma collection. Then the success of soybean planting in the saline land is determined by environmental modifications. Modification supportive environment for the

growth of soybean plants by adding ameliorant reduces levels of salt. Soil ameliorants that can be used are in the form of gypsum and organic materials such as cow manure. The aim of this study was to investigate the effect of gypsum and cow manure on the uptake of Na, K and the yield of soybean genotypes under saline condition.

Materials and Methods

This research was conducted in the green house of Jatikerto Experimental Farm belonging to the Faculty of Agriculture, Brawijaya University. The experimental farm is located at an altitude of 303 m above sea level with an average temperature of 27-29° C and rainfall of 120 mm per month. The research was carried out from June to September 2014. The research used a split plot design, the main plot was a soybean genotypes which consisted of two saline susceptible varieties (G1 = Wilis and G2 = Tanggamus) and two saline tolerant genotypes (G3 = genotype IAC,100/Bur//Malabar and G4 = genotype Argopuro//IAC,100) (Aini et al., 2014); the sub plot was ameliorant application consisting of A0 = without ameliorant; A1 = cow manure (20 t/ha); and A2 = gypsum (5 t/ha).

Planting medium used was a non-saline soil. The medium was treated with sea water mixed with groundwater to the EC up to ± 6.5 dS/m. The non-saline soil that has been air-dried and shieved was then put into a polybag as much as 5 kg. Water content of the soil was maintained at field capacity in accordance with the sea water that has been mixed with ground water. Comparison of water during the first irrigation was 250 mL sea water + 750 mL of groundwater, the second irrigation was 350 mL of sea water + 500 mL of groundwater, and the third irrigation was 250 mL of sea water + 750 mL of groundwater. After irrigation, planting media were incubated for 3 days with the intention of saline solution absorbed by the soil evenly.

Gypsum and manure were mixed evenly according to the treatment prescribed. The dose of gypsum was 10.41 g per polybag and the dose of cow manure was 41.66 g per polybag. Before planting, the grains were mixed with the insecticide active ingredient carbo sulfane (Furadan 3G) to avoid fly attack grainlines. Basic fertilizer used was NPK fertilizer (Phonska) at a dose of 300 kg/ha. NPK fertilizer was given 2 weeks after planting. Three soybean grains were planted in every polybag. Crop thinning was done 7 days after planting leaving two healthy plants per polybag. Irrigation was done two days by adjusting the conditions in the field. Irrigation was made using ground water to field capacity

conditions. Data were collected from plant growth and yield parameters. The plant growth parameter observations included Leaf chlorophyll Index using Chlorophyll meter SPAD-502 in phase reproductive initial (R1), Shoot to root proline content ratio in leaves was measured using the method of Bate (1973). Observation of parameters yield components that included grains per plant, potassium (K) and sodium (Na) content were measured by Flamephotometer Dual Chanel Model 2655-10. The data of experiment were analyzed by the F test at 5% followed by the HDS test ($\alpha = 5\%$).

Results and Discussion

Effect of ameliorant on leaf chlorophyll index

A mechanism of salt stress on growth of soybean is that a high concentration of ion Na^+ in growth media will replaceseon K^+ causing the effect of accumulation of Na^+ in plant tissues. Potassium has a function in the biochemical process i.e. maintaining an integrity of a photosynthetic structure, CO_2 fixation, photosynthate transport, regulating of chlorophyll content and adjusting of turgor pressure (Alikhani et al., 2011). The accumulation of Na ion causes chlorosis in leaf and dry out of leaf. It can be seen with a lower

leaf chlorophyll index on susceptible varieties. A Leaf chlorophyll index can be affected by photosynthesis rate. Chlorophyll content index (CCI) is a measure of photosynthesis activity especially in salt stressed plant. It has been used as a good indicator of salt tolerance in some species of soybean (Golezani and Noori, 2011). Increase in CCI at higher level of potassium can be due to decrease in activity of chlorophyllase.

Leaf chlorophyll index of soybean was significantly affected by the interaction between genotype and kind of ameliorant (Figure 1.). In general, application of cow manure and gypsum significantly increased leaf chlorophyll index. This was probably due to the role of organic matter in improving availability of soil nutrients that can be absorbed by soybean. Gypsum significantly increased leaf chlorophyll index instead of cow manure both of tolerance and susceptible varieties. This result was in accordance with that of Frazen et al. (2006) that application of gypsum on saline soil increased soil aggregation and percolation, and decreased soil pH. Ca^{2+} ions can replace Na^+ ions on saline soil, therefore no Na accumulation in plant tissue and roots. Ca^{2+} ion reduced absorption of Na^+ ion and increased absorption of K^+ (Hanafiah, 2007).

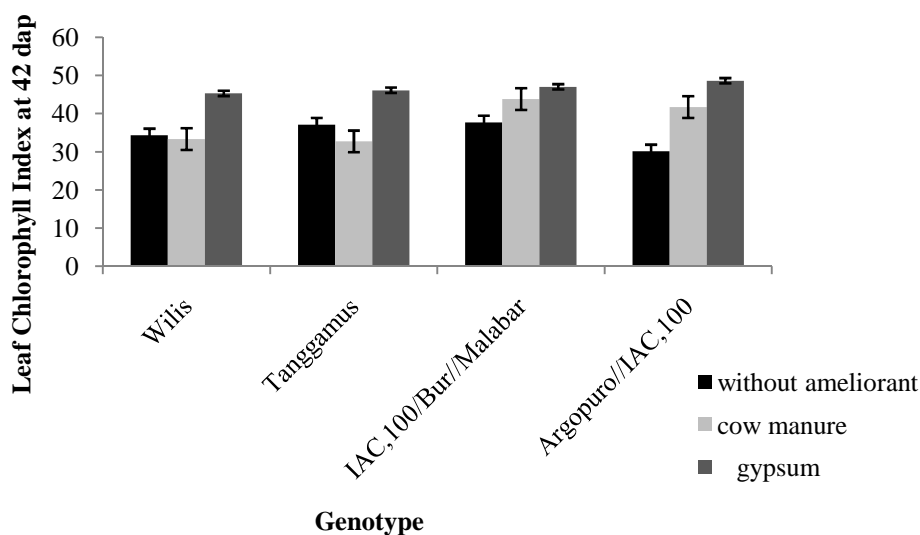


Figure 1. Leaf chlorophyll index of soybean genotype at 42 dap with application of ameliorant

Application of gypsum as a chemical ameliorant has been reported to improve physical and chemical properties of saline soil. Other properties such as soil aggregate structure, soil permeability, bulk density and alkalization can be

improved significantly with the addition of Gypsum (Clark and Baligar, 2003). In addition, applying cow manure could effectively reduce salt content, improve soil fertility, enhance an activity of soil microbes and increase the enzyme

activity in soil, so it could promote crop growth (Njoku et al., 2008). In the other result, the application of ameliorant reduced Na accumulation in stem and roots. Gypsum and cow manure were associated with the most significant decrease in Na accumulation, but cow manure more significantly decreased Na accumulation in roots than gypsum (Li-ping et al., 2015). This means that cow manure is associated with the higher increase K content in stem and root than gypsum.

Effect of ameliorant on proline content of leaf

Synthesis of proline in the plant grown under stress condition is higher than that grown under

normal conditions. Proline accumulation can be used as an indication that the plant is susceptible or tolerant to salinity conditions. Proline content of leaf soybean was significantly affected by an interaction between genotype and kind of ameliorant. The results showed that susceptible varieties accumulated proline higher than the tolerant genotypes (Figure 2). These results might be consistent with the findings of other studies which imply that the plants in salt stress will synthesize proline higher than the plants without salt stress (Cha-um and Kirdmanee, 2009; Nazarbeygi et al., 2011 ; Shafi et al., 2011; Akgun et al., 2011).

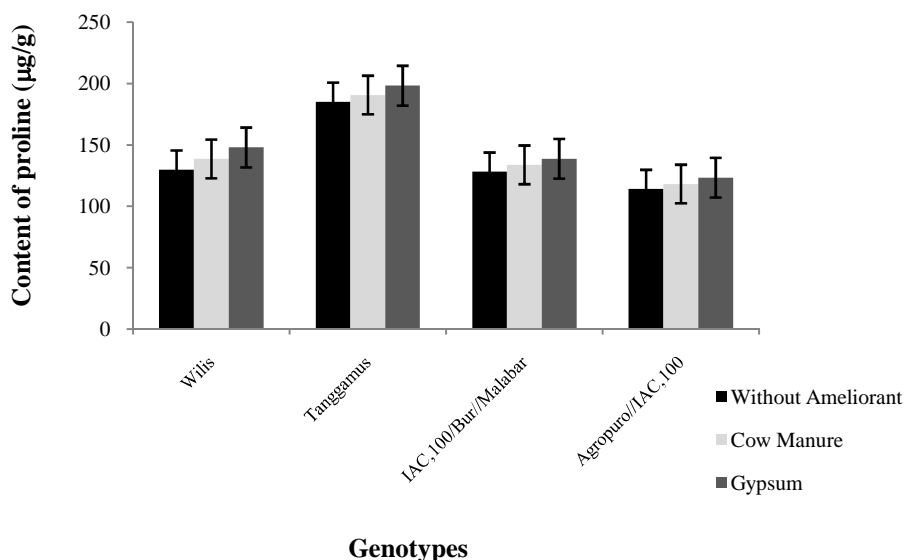


Figure 2. Content of proline of soybean genotype with application of ameliorant

Proline accumulation increases as soil salinity increases. Proline is accumulated in larger amounts than other amino acids in salt stress plants. Proline mitigates the effect of NaCl on cell membrane disruption. Proline accumulation was observed in *Jatropha curcas* L. under saline condition. Saline condition increased proline concentration, particularly in plants inoculated with PGPR. The results showed that vermicompost ameliorant had lower proline concentration than PGPR *Pseudomonas pseudoalcaligenes* (Patel and Meenu, 2012).

Effect of ameliorant on plant growth and yield

Shoot to root ratio shows the ratio between shoot dry weight and root dry weight. If the shoot to root ratio is high, the shoot dry weight is greater

than the root dry weight. If shoot to root ratio is low the shoot dry weight is smaller than the root dry weight. Shoot to root ratio on tolerant genotype was higher than that of susceptible varieties (Figure 3). On susceptible varieties (Wilis and Tanggamus), ameliorant application did not give significant responses compared to that without ameliorant. On tolerant genotypes (IAC,100/Bur//Malabar), application of cow manure gave shoot to root ratio higher than gypsum, while on Argopuro//IAC,100 genotype, application of gypsum gave shoot to root ratio higher than cow manure. Application of ameliorant on tolerant genotypes increased grain yield of soybean. Application of cow manure and gypsum on genotype IAC,100/Bur//Malabar and Argopuro//IAC,100 gave a significant increase of grain yield compared without ameliorant.

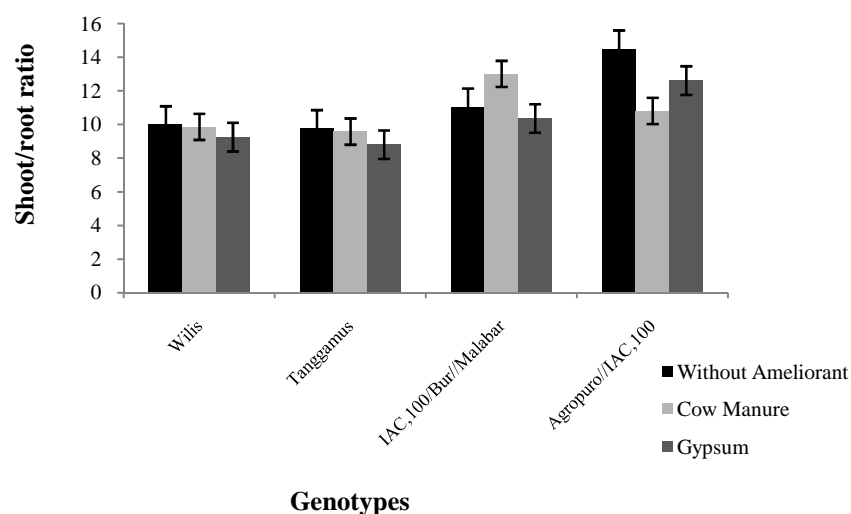


Figure 3. Shoot root ratio of soybean genotype with application of ameliorant

Application of gypsum on genotype Argopuro//IAC,100 had higher grain yield than application of cow manure. The increases of grain yield caused by application of ameliorant on IAC,100/Bur//Malabar were 43,56% by cow manure and 30,68% by gypsum, while on Argopuro//IAC,100, application of cow manure increased grain yield by 108,27% and application of gypsum increased 202,75%. Application of gypsum on varieties of Wilis and Tanggamus also increased grain yield by 6,45% and 18,44%, respectively, but application of cow manure in both susceptible varieties could not increase grain yield compared to without ameliorant (Table 1). Grain yield is the effect of plant growth, which in

general, crop yield is effected by plant growth. Plant growth hampered by an internal factors or external factors will have decreased crop yield. Salinity is an external factor and genotypes is an internal factor which effects crop yield. Application of ameliorant such as organic matter affects soil chemical and physical properties. Soil properties influenced by organic matter includesoil structure; aggregate stability, moisture holding capacity, stabilizer soil temperature and cation exchange capacity, that are beneficial and harmful to crop production and nutrient availability (Simanungkalit et al., 2006; Bot and Benites, 2005).

Table 1. Grain yield of soybean genotypes with application of ameliorant

Genotypes	Dry weight of grain yield (g/plant)		
	Without Ameliorant	Cow manure	Gypsum
Wilis	2.67 b	1.25 a	3.11 b
Tanggamus	3.20 b	1.17 a	3.79 bc
IAC,100/Bur//Malabar	2.64 b	3.79 bc	3.45 bc
Argopuro//IAC,100	1.45 a	3.02 b	4.39 c
LSD 5%		1.15	

Remarks: Mean values within the same column followed by the same letter do not differ significantly (0.05 level) according to the LSD test.

The yield reduction of some soybean under saline condition is probably caused by the decrease of soil potential and increase of uptake Na and Cl that make water deficiency and inhibit nutrient absorption. Application of soil amendment like gypsum and organic amendments may have positive effects on soil functions and plant

growth. Application of organic ameliorant can also act as a fertilizer by saving nutrients or increasing the fertilizer use efficiency of plant.

Effect of ameliorant on K/Na ratio

K/Na ratio can be used as an indicator of plant performance under salinity stress. This research

results showed that K/Na ratio was significantly affected by genotype and ameliorant (Figure 4). K/Na ratio on susceptible varieties was higher than tolerance genotypes. According to Asch et al. (2000), high K/Na ratio indicates high potassium uptake. In contrast, low K/Na ratios indicates high sodium concentration in the leaf blade. This result

was slightly different from the results of Aini et al. (2014) that the concentration of K in the leaves in susceptible varieties to saline conditions (Willis and Tanggamus varieties), was lower than that of salinity tolerant genotypes (IAC,100/Bur//Malabar and Argopuro//IAC,100).

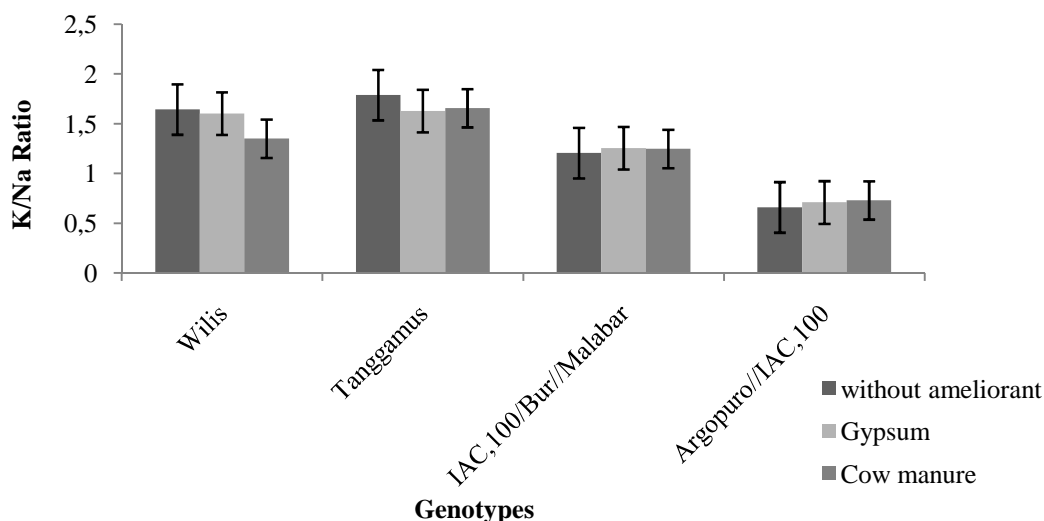


Figure 4. Effect of ameliorants on K/Na ratio of soybean genotypes

Conclusion

The application of gypsum and cow manure increased leaf Chlorophyll Index in a susceptible varieties and tolerance genotypes. Accumulation of proline and K/Na ratio of susceptible varieties were higher than tolerant genotypes. In tolerance genotypes (Argopuro//IAC,100), grain yield could increase more than 100% but in susceptible varieties application gypsum could increase grain yield in the range of range 6 to 18%.

Acknowledgements

This research was funded by Directorate of Higher Education through the Competitive Research Grant Program for Decentralization Research Brawijaya University with contract number: 410 113/UN10.21/PG/2014. We also thank Nur Cholid Susianto, a graduate of Faculty of Agriculture, Brawijaya University.

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