

RESEARCH ARTICLE

Tolerance to soil acidity of soybean (*Glycine max L.*) genotypes under field conditions Southwestern Ethiopia

Tolossa Ameyu Bedassa^{1,2*}, Abush Tesfaye Abebe³, Alemayehu Regassa Tolessa⁴

1 Jimma Agricultural Research Center, Jimma, Ethiopia, **2** Ethiopian Institute of Agricultural Research, Addis Ababa, Ethiopia, **3** International Institute of Tropical Agriculture, Ibadan, Nigeria, **4** Department of Natural Resource Management, Jimma University, Jimma, Ethiopia

* tolambo@gmail.com



OPEN ACCESS

Citation: Bedassa TA, Abebe AT, Tolessa AR (2022) Tolerance to soil acidity of soybean (*Glycine max L.*) genotypes under field conditions Southwestern Ethiopia. PLoS ONE 17(9): e0272924. <https://doi.org/10.1371/journal.pone.0272924>

Editor: Saddam Hussain, Department of Agronomy, University of Agriculture, Faisalabad, PAKISTAN

Received: February 15, 2021

Accepted: April 22, 2022

Published: September 15, 2022

Peer Review History: PLOS recognizes the benefits of transparency in the peer review process; therefore, we enable the publication of all of the content of peer review and author responses alongside final, published articles. The editorial history of this article is available here: <https://doi.org/10.1371/journal.pone.0272924>

Copyright: © 2022 Bedassa et al. This is an open access article distributed under the terms of the [Creative Commons Attribution License](https://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Data Availability Statement: All relevant data are within the paper and its [Supporting Information](#) files.

Abstract

Soil acidity with associated low nutrient availability is one of the major constraints to soybean production in southwestern Ethiopia. Integrated use of lime and acid-tolerant crops is believed to reduce soil acidity and improve crop production. The experiment was conducted in the field condition of Mettu, southwestern Ethiopia during the 2017/18 main cropping season. The experiment comprised fifteen soybean genotypes and two soil amendment (lime and unlimed) treatments arranged in a split-plot design with three replications. For each treatment, four rows were planted per plot; data related to growth, root, nodule, and yield of the crop were collected at a necessary stage for each. Liming and genotype interaction had significantly ($P = 0.01$) affected all parameters considered except for hundred seed weight and root volume and were affected only by the main effects of genotypes and liming. A significant reduction for most parameters was found on lime-untreated soil than treated soil. Though some genotypes showed higher performance for root, growth parameters, and yield components under unlimed soils; however, gave higher yield and yield components, when grown on lime-untreated with an average yield reduction of 13.7%, due to soil acidity. The maximum grain yield of ($1943.93 \text{ kg ha}^{-1}$) was obtained under lime treated acid soil from PI567046A genotype; while the lowest ($510.49 \text{ kg ha}^{-1}$) were recorded from SCS-1 genotype under the lime untreated acid soil. Genotype BRS268 showed higher yield ($1319.83 \text{ kg ha}^{-1}$) under lime untreated acid soil than lime treated acid soil ($1143.47 \text{ kg ha}^{-1}$) and showed less reduction percentage for a number of the nodules, root weight, and number of seeds per plant; while PI567046A showed high reduction percentage for yield, biomass, number of pod and seed per plant. A high difference was observed among the soybean genotypes for soil acidity tolerance, which might be further exploited by breeders for the genetic improvement of soybean. Genotype BRS268 had performed better than other tested genotypes under increased soil acidity. selection would be effective to improve soybean genotypes performance on acid soils and identify low Phosphorus tolerant genotype that helps smallholder farmers optimize soybean productivity on acid soils in the study area. HAWASSA-04 variety is the most tolerant among the tested materials. However, further study is required by considering additional genotypes to reach a conclusive recommendation.

Funding: The funder of this paper was only the Ethiopian Institute of Agricultural Research and Authors only. As I indicated in the first draft of the submission, the Ethiopian Institute of Agricultural Research covers daily labor to conduct this activity, providing planting materials(seed), providing land to conduct activities. However, the Ethiopian Institute of Agricultural research has no role in data collection, analysis, preparation of manuscript but those authors have.

Competing interests: The authors have declared that no competing interests exist.

Introduction

Land degradation, soil nutrient depletion and increasing soil acidity is challenging problem in south western Ethiopia. Soil acidity is one of the major problem that have profound effect on the productive potential of crops, such as soybean, because of low availability of basic cations, and excess and toxic levels of hydrogen and aluminum in exchangeable forms [1]. The major causes for soils to become acidic are high rainfall, leaching, acidic parent material, organic matter decay, and harvest of high yielding crops. Crop management practices (continuous application of acid forming fertilizers), removal of organic matter, and contact exchange between exchangeable hydrogen on root surfaces and microbial production of nitric and sulfuric acids can also contribute to soil acidity [2]. Soil acidity is often an insidious soil degradation process, developing slowly; although indicators, such as falling yields, leaf discolorations in susceptible plants, and lack of response to fertilizers might indicate that soil pH is declining to critical levels [3]. Theoretically, soil acidity is quantified based on H^+ and Al^{3+} concentrations of the soils [4]. Acidic soils limit the production potential of crops because of low availability of basic cations and excess of hydrogen (H^+) and aluminum (Al^{3+}) in exchangeable forms. It affects beneficial microorganisms, reduced root growth, which limits absorption of nutrients and water [5], consequently, leading to poor plant growth and yield of crops. However, Al^{3+} toxicity is one of the major limiting factors for crop production on acid soils by inhibiting root cell division and elongation, thereby, reducing water and nutrient uptake [6] poor nodulation or mycorrhizal infections.

Acidic soil is mostly distributed in developing countries, where there is high population growth, and food demand is ever increasing. Acid soils make up approximately 30% of the world's total land area and more than 50% of the world's potentially arable lands, particularly in the tropics and subtropics [7]. Acidic soils cover a total of 1.66 billion hectares in developing countries, while the total area affected by soil acidity is about 4 billion hectares [8]. In high rainfall areas, excessive rainfall coupled with unfavorable temperature and precipitation is high enough to leach appreciable amounts of exchangeable basic cations [9].

Soybean production and productivity have been growing rapidly in Ethiopia, in the past decade. According to the Agricultural Sample Survey of CSA (Central Statics Agency) [10], 130,022.00 private peasant holdings cultivated about 36,635.79 hectares of land and produced about 812, 34.659 tons of soybean. The average production of soybean in the country is, therefore, $2.2 t ha^{-1}$ while, that of the Mettu area is by far below ($1.3 t ha^{-1}$) the national average due to soil acidity [11].

Lime and fertilizer management practices are of primary importance for the proper management of soil acidity. Application of lime significantly increased root and shoot yields of soybean in Nigeria [12]. Nevertheless, for economic reasons, it is often not practicable for resource-poor farmers to apply high rates of lime [13]. And [14] also reported that application of lime with high rate is not practicable for resource-poor farmers, as well as, mineral fertilizers. However, previous studies revealed the existence of sufficient genotypic variability of bean germplasm for acid-tolerant [15, 16]. Hence, identification of tolerance soybean genotypes to soil acidity is an economically feasible option that might serve as an acid soil management practice [11]. Hence, the identification and use of soybean genotypes that are tolerant to acid soil conditions of Southwestern Ethiopia is a very useful approach to ensure economic stability to many subsistence farmers, who cannot afford the application of liming materials practices [11]. A preliminary field screening of soybean genotypes in southwestern Ethiopia has demonstrated the presence of genetic variability among genotypes in tolerating soil acidity stress. Studying responses of selected genotypes with contrasting tolerance to soil acidity may help in generating information that could be utilized by breeding programs aimed at developing

aluminum-tolerant cultivars for areas where soil acidity remains a key environmental constraint to crop production. Therefore, to meet the demand of soybean in Ethiopia including the study area, emphasis should be given to increase the productivity of the crops through the use of genotypes that can tolerate acid stressed soil conditions. The objective of this study was to test the hypothesis that differences exist in growth, root, yield and yield parameters among soybean genotypes selected for soil acidity tolerance when subjected to limed and unlimed acid soil.

Materials and methods

Description of the study site

The field experiment was conducted at Mettu Agricultural Research Sub Center. Mettu is located in south western Ethiopia at 8° 19' 0" N latitude, 35° 35' 0" E longitude, and at the altitude of 1550 meters above sea level. The average annual rainfall of the study site was 1835 mm/annum, an annual mean minimum and maximum temperatures were 12 and 27 °C respectively (Figs 1 and 2).

The soil of the study area has a pH (H₂O) value of 4.5, exchangeable acidity of 2.82 cmol kg⁻¹ soil and soil available phosphorus level of 1.16 ppm before applying the treatments. Physical and some chemical properties of the soil in the study area before sowing and after harvesting are presented (Table 1).

Soil Sampling and analysis

Prior to the field experimentation both undisturbed and disturbed samples were collected. Three undisturbed soil samples were taken by core sampler. Fresh weight and an oven dry

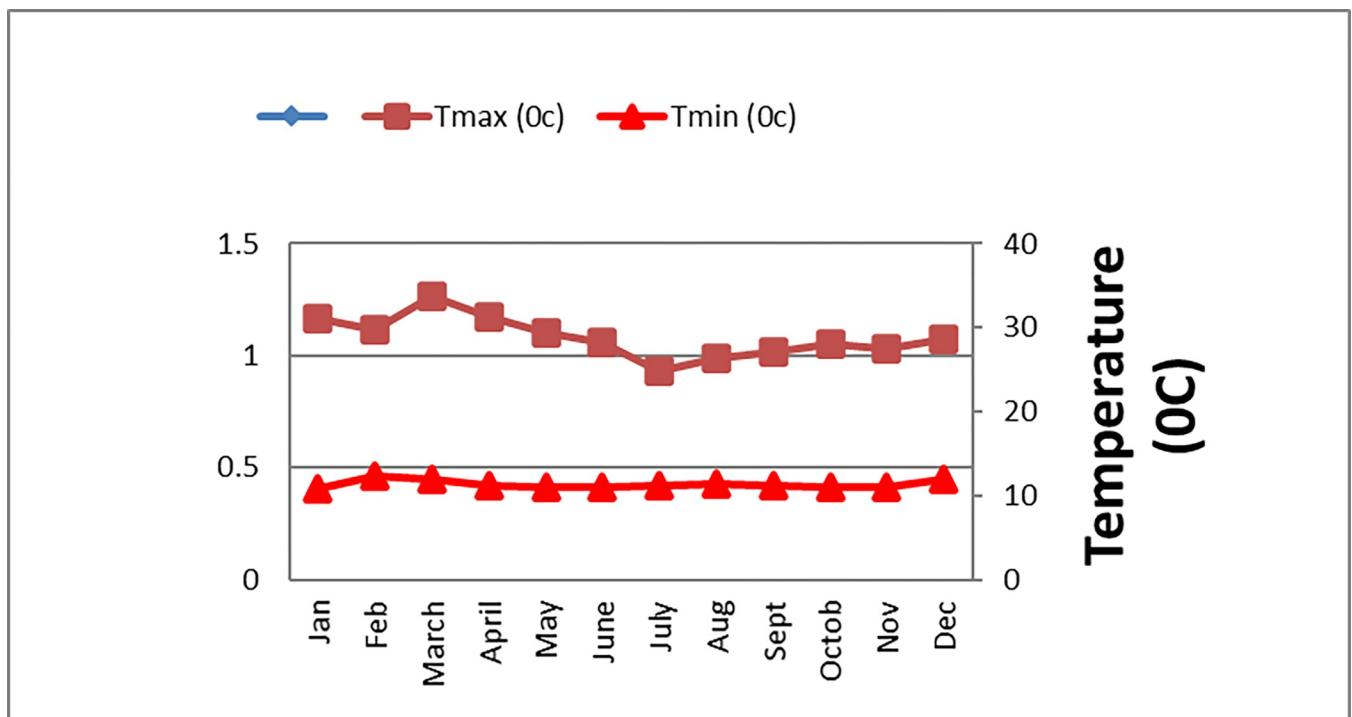


Fig 1. Mean minimum and maximum temperatures (°C) of Mettu during crop growth period in 2017.

<https://doi.org/10.1371/journal.pone.0272924.g001>

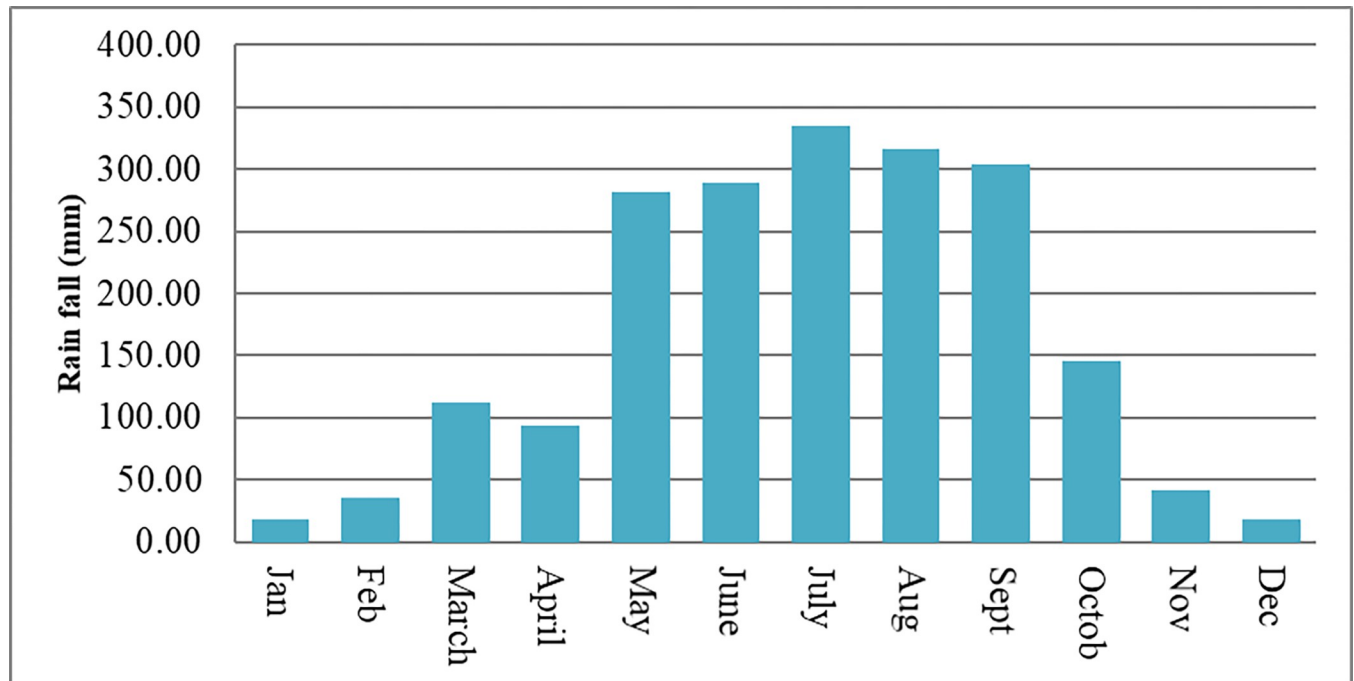


Fig 2. Monthly total rainfall (mm) of Mettu during crop growth period in 2017.

<https://doi.org/10.1371/journal.pone.0272924.g002>

weight at 105°C, of the soil samples was used to determine bulk density [17]. Five random disturbed soil samples (0-15cm depth) were collected diagonally and composite soil sample was made.

The composite sample was used for soil physiochemical analysis, and for the determination of lime requirement of the soil. The disturbed soil samples were air dried and sieved to pass through 2 mm sieve, and placed in a labeled plastic bag. Then, the samples were transported to Jimma Agricultural soil and plant tissue analysis laboratory. The soil sample were analyzed for particle size distribution(soil texture), which was done by Bouyoucos hydrometer method as [18]; while soil exchangeable acidity, exchangeable bases, soil pH, organic carbon(OC), total nitrogen(TN), available phosphorus and cation exchange capacity (CEC) for soil chemical analysis were selected. Available soil P and exchangeable acidity were determined using Bray-II method, as described by [19, 20] respectively.

After harvesting soil sample were taken from lime treated and untreated separately. The collected samples were air dried and sieved to pass through 2 mm sieve and submitted to soil laboratory for soil chemical properties analysis. Organic matter was determined using wet oxidation; Total N was determined by Kjeldahl method, as described by [21]. Cation exchange capacity (CEC) and exchangeable bases (Ca, Mg, K and Na) were determined ammonium acetate at pH 7. The potential cation exchange capacity (CEC) of the soil was determined from the NH₄⁺ saturated samples that were subsequently replaced by K⁺ using KCl solution. The excess salt was removed by washing with ethanol and the NH₄⁺ that was displaced by K⁺ was measured using the micro-Kjeldahl procedure [22], and reported as CEC. Exchangeable Ca and Mg was analyzed using Atomic Absorption spectrophotometer (AAS). Exchangeable Na and K were analyzed using flame photometer as described by [22].

Available soil P was determined using Bray-II method, as described by Bray and Kurtz (1945). The soil pH was determined in soil water suspension of 1:2.5 (soil: water ratio) using pH meter, as described by Van Reeuwijk (1992) [23]. Exchangeable acidity was determined by

Table 1. Physicochemical properties of the experimental soil prior to sowing and after harvesting.

Parameters	Before sowing	after harvesting	
		Limed	Unlimed
Particle size distribution			
Clay (%)	49.00		
Sand (%)	38.00		
Silt (%)	13.00		
Textural class	Clayey		
pH (H ₂ O)	4.400	4.73	4.48
Exchangeable acidity (cmol(+)/kg)	2.720	1.52	2.41
Exchangeable Al (cmol(+)/kg)	1.460	0.93	1.38
Organic carbon (%)	2.210	2.45	2.22
CEC (cmol (+) kg ⁻¹)	18.75	21.04	18.89
Total N (%)	0.210	0.24	0.22
Available P (BrayII) (mg kg ⁻¹)	2.950	4.39	2.98
Exchangeable K (cmol (+) kg ⁻¹)	0.330	0.67	0.40
Exchangeable Ca (cmol(+) kg ⁻¹)	3.550	5.39	3.81
Exchangeable Mg (cmol(+) kg ⁻¹)	1.380	1.59	1.40

<https://doi.org/10.1371/journal.pone.0272924.t001>

saturation of the soil samples with potassium chloride solution and titrates with sodium hydroxide as described by [13]. From the same extractant, exchangeable Al in the soil was titrated with a standard solution of 0.02M HCl.

Determination of lime requirements

The amount of lime applied was determined based on the exchangeable acidity, mass per 0.15m furrow slice and bulk density of the soil [24], considering the amount of lime needed to neutralize the acid content (Al + H) of the soil up to the permissible acid saturation level for soybean growth.

$$LR, \frac{\text{CaCO}_3 \text{kg}}{\text{ha}} = \frac{\text{Cmol} \frac{\text{EA}}{\text{Kg}} \text{ of soil} * 0.15\text{m} * 10\text{m}^2 * \text{BD} \left(\frac{\text{g}}{\text{cm}^3} \right) * 1000 * \text{crop factor}}{2000}$$

Where: BD = bulk density, EA = exchangeable acidity (exch. H⁺ + Al³⁺), LR = lime requirement, 0.15m = plough depth/depth of lime incorporation.

Planting material, treatments, experimental design and procedures

The treatments comprised of two factors namely; two soil amendments (control or no lime and limed) and fifteen different soybean genotypes (Table 2). The treatments were laid out in split plot design. The experiment has three technical replications and six biological replication in 4m by 2.4 m (9.6 m²) plot size. Per row eighty (80) and per plot/treatments 320 seeds were used. Soil amendments (lime and unlimed) applied as main plot treatments and soybean genotypes were applied to sub plot treatments. The different soybean genotypes for the trial were identified from previous advanced Multi-Location Yield Trials, including previous soil acidity tolerance screening trials.

The lime requirement (LR) of the soil for the plots was determined based on exchangeable acidity (EA) or acid saturation of the experimental soil. The lime rate was, therefore, 3457.8 kg/ha based on exchangeable acidity of the soil. Calcium carbonate (CaCO₃) was used as the source of lime and the whole doses of lime were broadcasted on limed plots manually, uniformly and mixed in the top 15 cm soil layer, a month before sowing. Reduction percentage

Table 2. Soybean genotypes used for the experiment.

Genotypes	Back ground information and Source
JM-CLK/CRFD-15-SA	Inbreed line identified from local crosses at JARC
JM-ALM/PR142-15-SC	Inbreed line identified from local crosses at JARC
JM-ALM/H3-15-SC-1	Inbreed line identified from local crosses at JARC
BRS 268	Introduced from Brazil
JM-HAR/DAV-15-SA	Inbreed line identified from local crosses at JARC
JM-CLK/G99-15-SC	Inbreed line identified from local crosses at JARC
JM-CLK/G99-15-SB	Inbreed line identified from local crosses at JARC
JM-H3/SCS-15-SG	Inbreed line identified from local crosses at JARC
PI 567046A	Introduced from USA
SCS-1	Pipe line from Pawe agricultural research Center
PI 423958	Introduced from USA
H-7	Pipe line from Mozambique ARI
HAWASSA-04	Released variety from Hawassa Agricultural research

JARC = Jimma Agricultural Research Center

<https://doi.org/10.1371/journal.pone.0272924.t002>

for grain yield, root volume, growth and nodulation parameter was calculated as the ratio in lime treated to lime untreated soil, which also showed higher differences among the tested genotypes. The seeds were sown in rows to maintain plant to plant distance of 5 cm and 60 cm between rows. The cropping was done in main season by rain fall without any germination and seedling establishment problem. Cultural practices (i.e. weeding, hoeing etc, the experimental field was weeded by hand five times during the growing period uniformly for all treatments).

Statistical analysis

The data was subjected to analysis of variance (ANOVA) using Statistical Analysis System (SAS) software version 9.3 [25] using proc GLM procedure. The difference between treatment means was separated using LSD 5% value. Correlation analysis between the traits was carried out to determine the magnitude and degree of their associations.

Results and discussion

Effects of soil acidity on yield and yield components of soybean genotypes

Genotypes reflected significant differences for number pods per plant, number of seeds per plant, grain yield, and hundred seed weight and above ground biomass in both limed and unlimed soil regimes (Table 3). The interaction of amendment*genotypes was also highly significant ($p \leq 0.01$) for all yield components and yield, however hundred seed weight was only affected by the main effect of genotypes and amendments.

The response of the observed soybean characters in acidic soil varied among genotypes. Grain yield, number of pods per plant, number of seeds per plant, hundred seed weight and above ground biomass in unlimed soil with low pH was lower than in treated soil or same with that in limed (Table 4). Even, grain yield of BRS268 genotype under unlimed acid soil were higher than in limed acid soil.

On average, the genotypes gave higher yield and yield components in lime treated soil (Table 4). PI567046A genotype gave higher grain yield, above ground biomass, number of pods and seeds per plant in lime treated acid soil; while genotype SCS-1 gave the lowest yield and genotype PI423958 gave the lowest number of pods, seeds and above ground biomass in

Table 3. Mean square of growth, root, nodulation, yield and yield components of soybean genotypes grown on limed and unlimed soil on field.

Source of variations	Gen	Lime	Lime* Gen	Error (a)	Total
Parameters					
Yield	582515.8**	341225.8**	98147.32**	4211.35	10148754.36
No of pod per plant	195.62**	239.44**	39.59**	0.41346	3558.5
No of seed per plant	855.29**	801.62**	144.54**	3.444	15005.62
Above ground biomass	5.275**	5.436**	1.3597**	0.1852	109.9
Number of nodule	206.31**	1626.47**	55.06**	1.143	5350.77
Root dry weight	0.093**	0.2722**	0.01715**	0.001716	1.9161
Plant height	810.97**	577.85****	7.4215**	2.143	12190.7
Root volume	1.0074**	4.71512**	0.2408 ^{ns}	0.227	35.624
Shoot dry weight	7.3028**	40.1735**	2.141**	0.0228	173.756
Hundred seed weight	19.486**	10.64**	0.739 ^{ns}	3.122	492.733

** = highly significant different at 1% level of significance, ns = non-significant d/t at 5% level of significance, Gen = genotypes

<https://doi.org/10.1371/journal.pone.0272924.t003>

lime untreated acid soil (Table 4). Under lime untreated soil condition, the maximum grain yield and above ground biomass was obtained from variety HAWASSA-04, and genotypes BRS268 and PI567046A gave highest number of pods and seeds per plant respectively. Under unlimed acid soil condition, the highest increasing percentage for yield and above ground biomass, was shown by BRS268 and JM-DAV/PR142-15-SA respectively, while the highest decreasing percentage was shown by PI567046A for both yield and above ground biomass.

The yield increments with lime application might be due to the probability of obtaining the available P from decomposed OM by microorganisms, when the pH value of the soil improved

Table 4. Interaction effect of genotypes and lime for yield and yield components of soybean genotypes grown under limed and unlimed soil at Mettu during 2017 main cropping season.

Genotypes	Yield (kg/ha)		NPPP		NSPP		AGB (ton/ha)	
	L	UL	L	UL	L	UL	L	UL
PI567046A	1943.93 ^a	1069.87 ^{d-i}	47.1 ^a	26.6 ^{cd}	92.33 ^a	55.87 ^c	7.02 ^a	3.23 ^{e-k}
HAWASSA-04	1576.77 ^{ab}	1553.11 ^{bc}	29.36 ^{bc}	22.93 ^{gh}	60.73 ^b	41.53 ^{e-i}	5.06 ^b	4.2 ^{bc}
JM-PR142/H3-15-SB	1328.29 ^{b-d}	1027.24 ^{e-j}	21.53 ^{h-k}	19.46 ^{l-n}	43.67 ^{ef}	37.13 ^{j-l}	4.24 ^{bc}	3.58 ^{c-g}
BRS268	1143.47 ^{d-g}	1319.83 ^{b-e}	30.33 ^b	27.6 ^{cd}	47.73 ^d	47.73 ^d	4.01 ^{cd}	3.79 ^{c-f}
JMALM/PR142-15-SC	1214.46 ^{c-f}	1121.35 ^{d-g}	20.27 ^{k-m}	21.33 ^{i-k}	44.9 ^{de}	44.67 ^{de}	4.01 ^{c-e}	3.96 ^{c-e}
JM-H3/SCS-15-SG	956.49 ^{f-k}	1096.45 ^{d-h}	20.53 ^{j-l}	20.46 ^{j-m}	44.36 ^{de}	43.53 ^{ef}	3.52 ^{c-h}	3.57 ^{c-g}
JM-CLK/G99-15-SB	1076.24 ^{d-h}	756.98 ^{j-p}	22.93 ^{gh}	21.8 ^{h-j}	38.9 ^{i-k}	39.13 ^{h-j}	2.79 ^{h-m}	2.65 ^{j-n}
JM-CLK/CRFD-15-SA	935.05 ^{f-l}	643.49 ^{m-p}	22.53 ^{hi}	18.07 ^{m-o}	40.33 ^{f-j}	33.47 ^{l-n}	3.43 ^{d-j}	2.89 ^{g-m}
JM-DAV/PR142-15-SA	934.71 ^{f-l}	915.36 ^{g-m}	24.33 ^{fg}	24.8 ^{ef}	42.76 ^{e-h}	42.26 ^{e-i}	2.99 ^{g-m}	3.07 ^{f-l}
H-7	772.48 ^{j-p}	821.79 ^{h-n}	21.33 ^{i-k}	18 ^{nop}	39.67 ^{g-j}	36.87 ^{j-l}	2.44 ^{l-n}	2.27 ^{m-o}
JM-CLK/G99-15-SC	783.83 ^{i-o}	818.21 ^{h-n}	22.53 ^{hi}	21.93 ^{h-j}	43.13 ^{e-g}	43.07 ^{e-g}	3.51 ^{c-i}	3.52 ^{c-h}
SCS-1	618.95 ^{n-p}	510.49 ^p	17.93 ^{n-p}	16.53 ^p	32.16 ^{mn}	29.8 ⁿ	2.58 ^{k-n}	2.36 ^{l-n}
JM-HAR/DAV-15-SA	737.46 ^{k-p}	690.96 ^{k-p}	19.27 ^{l-n}	17.4 ^{op}	34.33 ^{lm}	31.87 ^{mn}	2.35 ^{l-n}	2.23 ^{m-o}
JM-ALM/H3-15-SC-1	653.17 ^{m-p}	637.54 ^{n-p}	21.27 ^{i-k}	18.93 ^{m-o}	39.1 ^{ijk}	35.27 ^{k-m}	3.06 ^{f-l}	2.73 ^{h-m}
PI423958	682.82 ^{j-p}	528.21 ^o	13.33 ^q	9.76 ^f	22.13 ^o	14.33 ^p	1.87 ^{no}	1.43 ^o
Mean	1023.87	900.73	23.63	20.37	44.40	38.43	3.52	3.04
CV	6.74		2.922		4.48		13.12	

Means with the same letters are not significantly different at 5% level of significance. UL- unlimed; L- Limed; NPPP- Number of pod per plant; NSPP-Number of seed per plant; AGB- above ground biomass; CV- coefficient of variation

<https://doi.org/10.1371/journal.pone.0272924.t004>

due to liming, which might have resulted in increased grain yield. Liming also improved the ability of the plant to absorb P, when Al toxicity has been eliminated, and enhanced the vegetative growth of soybean genotypes, which resulted in increased dry biomass yield. In line with this result, [26] also reported that the highest barley grain yield was obtained under the application of 2.2 t/ha lime than unlimed acid soil. The genotypes responded to the applied lime for number of pod and seed, which might be due to lime enhanced vegetative growth and make genotypes to bear higher number of pod than lime untreated acid soil and also lime is neutralized acid soil which might increase the availability of phosphorus for plant uptake by reducing phosphorus fixation on acid soil. Lime also improved soil pH and enhanced growth and yield of soybean genotypes, as a result of increased P availability, photosynthesis intensity, flowering, seed formation and fruiting of the crops also increased (Kisinyo et al., 2016) [33]. In line with this results [13] reported that lime application increased a number of pod and seed per plant. [27] also reported that the application of lime produced the highest seeds per plant than unlimed soil. [28] reported 36.4% plant height decrease in soybean on unlimed acid soil compared with limed acid soil which has a direct relationship with yield increment under limed soil conditions.

The highest hundred seed weight was recorded for PI423958 genotype under lime treated soils and the lowest hundred seed weight was recorded for H-7 under lime untreated soil (Tables 5 and 6). In this study, the variable of tested genotypes has been observed, which indicates the presence of difference among the tested genotypes for hundred seed weight. Application of lime didn't affect hundred seed weight of genotypes (Table 5). In agreement with this result, [29] reported significant difference among soybean genotypes for hundred seed weight, in which the highest hundred seed weight was produced by BFS 39 genotype and the lowest hundred seed weight was recorded from Roba. [30] reported non-significant effect of liming on hundred seed weight of common bean. In general; lime application to the soil increased yield, number of pod and seed and above ground biomass of soybean genotypes by about 13.67, 16.06, 15.53 and 16.18% respectively, however hundred seed weight were not affected by lime.

Hundred seed weight is higher in un-limed soil, this might due to the improvement of soil pH in response to lime amendment, which enhanced growth and yield of the plant, as a result of increased availability of P that might have increased intensity of photosynthesis, flowering, seed formation and fruiting, as a result these formed fruit is competed for nutrient to fulfill the seed and the seed size become decreased which have the direct effect on seed weight.

Effect of soil acidity on root, growth, nodulation parameter of soybean genotypes

There were highly significant ($P < 0.01$) differences among genotypes for root dry weight, number of nodule, plant height and shoot dry weight in both soils regimes. The interaction of lim

Table 5. Average values of grain yield (kg/ha), NPPP, NSPP, AGB (ton/ha) and HSW (g) of soybean genotypes grown under limed and unlimed acid soil at Mettu during 2017.

Treatments	Yield (kg/ha)	NPPP	NSPP	AGB(ton/ha)	HSW(g)
Limed	1023.87 ^a	23.63 ^a	44.40 ^a	3.525 ^a	13.34 ^a
Unlimed	900.73 ^b	20.36 ^b	38.43 ^b	3.033 ^b	14.34 ^a
PR	13.67	16.06	15.53	16.22	-6.97

Means with the same letters are not significantly different at 5% level of significance. NPPP- Number of pod per plant; HSW = hundred seed weight, NSPP-Number of seed per plant; AGB- above ground biomass, PR = percent of reduction

<https://doi.org/10.1371/journal.pone.0272924.t005>

Table 6. Main effect of soybean genotypes for hundred seed weight grown under acid soil at Mettu on field during 2017 main cropping season.

Sub-plot treatments (Genotype)	HSW (g)
PI423958	17.5 ^a
JMALM/PR142-15-SC	16.43 ^{ab}
HAWASSA-04	15.08 ^{bc}
JM-H3/SCS-15-SG	15.078 ^{bc}
JM-PR142/H3-15-SB	14.68 ^{bc}
BRS268	14.46 ^{bc}
JM-CLK/CRFD-15-SA	14.228 ^c
JM-HAR/DAV-15-SA	13.83 ^c
JM-CLK/G99-15-SB	13.66 ^c
JM-CLK/G99-15-SC	13.39 ^c
JM-ALM/H3-15-SC-1	13.35 ^c
SCS-1	13.31 ^c
JM-DAV/PR142-15-SA	13.18 ^c
PI567046A	11.08 ^d
H-7	10.53 ^d
CV	12.617

Means with the same letters are not significantly different at 5% level of significance. HSW = hundred seed weight, CV = Coefficient of variation

<https://doi.org/10.1371/journal.pone.0272924.t006>

ing* genotypes was also highly significant ($p \leq 0.01$) for root dry weight, number of nodule, plant height and shoot dry weight, except for root volume only the main effect of liming and genotype s were significant. Growth, root and nodulation parameters of soybean genotypes grown under lime treated and untreated soils are indicated in (Tables 7 and 8). On average, the genotypes gave higher root, growth and nodulation parameters under lime treated acid soil (Tables 7 & 8). These results signified that application of lime increasing root, growth and nodulation parameters. JMALM/PR142-15-SC and BRS268 genotype gave higher root volume and root dry weight in both limes treated and untreated soil, and indicating these genotypes might be among acidic soil tolerant genotypes; while genotype PI423958 gave the lowest root dry weight on lime untreated acid soil (Table 8). [31] reported that among the fifteen soybean genotypes tested MLGG 0064 genotype showed the highest root dry weight under the control soil condition (pH 7), while the lowest root length was shown by genotype MLGG 0377 in Mn toxicity condition, which shows varietal difference for acid soil adaptation.

The alteration of root dry weight and root volume includes decreasing and increasing of the root length and root hair. Decrease percentage of root dry weight and root volume in acid soil stress conditions varied among the tested genotypes. Limed soil condition showed the highest root dry weight and root volume than in unlimed soil. This might be due to liming improved the P uptake capacity of plants which facilitate root growth, and then increased root diameter or root thickness of the genotypes, and root dry weight is the result of root growth and development, including root length and number of lateral roots. Alteration in root length and number of roots causes an alteration in root dry weight and root volume. Alteration also occurs on root hair length and root hair density [32] that cause an alteration in root dry weight.

There was no negative values existed for root dry weight in unlimed acid soil conditions (Table 10), but negative value existed for number of nodule. Negative value suggests an increasing variable from the optimal or relatively optimal condition to the severer condition. The highest number of nodule was obtained from JM-DAV/PR142-15-SA; while the lowest

Table 7. Interaction effect of genotypes and lime for SDW, NN and PHT of soybean genotypes grown under limed and unlimed soil at Mettu during 2017 main cropping season.

Genotypes	SDW (g/plant)		NN/plant		PHT (cm)	
	L	UL	L	UL	L	UL
PI567046A	6.32 ^{cd}	6.32 ^{cd}	32.56 ^f	20 ^m	83.73 ^a	73.267 ^b
HAWASSA-04	7.045 ^{ab}	7.045 ^{ab}	39.067 ^b	33.6 ^e	55.40 ^c	50.33 ^{cdef}
JM-PR142/H3-15-SB	5.40 ^{hij}	3.99 ^{no}	39.40 ^b	32.27 ^{fg}	54.72 ^{cd}	47.00 ^{efghi}
BRS268	5.93 ^{defg}	5.95 ^{def}	31.23 ^{ghi}	31.26 ^{ghi}	53.80 ^{cde}	48 ^{defgh}
JMALM/PR142-15-SC	6.16 ^{cde}	5.45 ^{ghij}	37.33 ^c	30.87 ^{hi}	46.27 ^{fghi}	44.2 ^{fghijk}
JM-H3/SCS-15-SG	5.58 ^{fghi}	4.46 ^{lmn}	35.20 ^d	26.2 ^j	48.93 ^{cdefg}	44.87 ^{fghij}
JM-CLK/G99-15-SB	7.56 ^a	4.46 ^{lmn}	37.33 ^c	23.33 ^{kl}	43.067 ^{ghijk}	40.67 ^{jklm}
JM-CLK/CRFD-15-SA	4.34 ^{mn}	2.92 ^{pq}	37.13 ^c	22.67 ^l	48.93 ^{cdefg}	42.73 ^{ghijk}
JM-DAV/PR142-15-SA	4.76 ^{klm}	4.78 ^{klm}	32.40 ^f	31.67 ^{fg}	37.33 ^{klmno}	34.40 ^{lmno}
H-7	5.18 ^{ijk}	4.027 ^{no}	39.00 ^b	34.13 ^{de}	32.00 ^{nop}	29.60 ^{op}
JM-CLK/G99-15-SC	5.73 ^{efgh}	4.80 ^{klm}	30.33 ⁱ	23.2 ^{kl}	48.8 ^{cdefg}	43.13 ^{ghijk}
SCS-1	3.60 ^o	1.97 ^r	24.20 ^k	15.67 ^o	44.83 ^{fghij}	39.0 ^{jklmn}
JM-HAR/DAV-15-SA	4.83 ^{kl}	4.97 ^{jk}	31.23 ^{ghi}	30.8 ^{hi}	38.6 ^{jklmn}	33.60 ^{mno}
JM-ALM/H3-15-SC-1	2.94 ^p	2.38 ^{qr}	34.36 ^{de}	17.6 ⁿ	45.87 ^{fghi}	41.50 ^{hijkl}
PI423958	6.48 ^{bc}	3.69 ^o	55.27 ^a	35.16 ^d	32.39 ^{nop}	26.20 ^p
	5.45	4.12	35.72	27.22	47.63	42.56
CV	3.15		3.39		3.245	

Means with the same letters are not significantly different at 5% level of significance. UL- unlimed; L- Limed; SDW- shoot dry weight; NN–number of nodule; PHT- plant height; CV- coefficient of variation

<https://doi.org/10.1371/journal.pone.0272924.t007>

Table 8. Main effect of soybean genotypes for RV and interaction effect of lime and genotypes for RDW grown under limed and unlimed acid soils at Mettu on field during 2017 main cropping season.

Genotypes	RDW (g/plant)		V/plant in ml
	Limed	Unlimed	
PI567046A	0.75 ^{cde}	0.433 ^j	2 ^{bcde}
HAWASSA-04	0.81a-d	0.807 ^{abcd}	2.33 ^{bc}
JM-PR142/H3-15-SB	0.79 ^{bcd}	0.74 ^{cde}	2.5 ^b
BRS268	0.84a-d	0.83 ^{abcd}	2.33 ^{bc}
JMALM/PR142-15-SC	0.937 ^a	0.873 ^{abc}	3.17 ^a
JM-H3/SCS-15-SG	0.71d-g	0.637 ^{efgh}	2.167 ^{bc}
JM-CLK/G99-15-SB	0.647 ^{efgh}	0.44 ^j	1.5 ^e
JM-CLK/CRFD-15-SA	0.893 ^{ab}	0.75 ^{cde}	2.43 ^b
JM-DAV/PR142-15-SA	0.73 ^{def}	0.72 ^{def}	1.87 ^{cde}
H-7	0.637 ^{efgh}	0.58 ^{ghi}	1.833 ^{cde}
JM-CLK/G99-15-SC	0.657 ^{efgh}	0.47 ^{ij}	1.6 ^{de}
SCS-1	0.8 ^{abcd}	0.47 ^{ij}	2.1 ^{bcd}
JM-HAR/DAV-15-SA	0.55 ^{hij}	0.55 ^{hij}	2 ^{bcde}
JM-ALM/H3-15-SC-1	0.717 ^{defg}	0.657 ^{efgh}	2.033 ^{b-e}
PI423958	0.59 ^{fghi}	0.427 ^j	1.833 ^{cde}
Mean	0.735	0.625	CV = 22.55
CV		6.093	

Means with the same letters are not significantly different at 5% level of significance. RDW- root dry weight; RV–root volume; CV- coefficient of variation

<https://doi.org/10.1371/journal.pone.0272924.t008>

Table 9. Average values of SDW (g/plant), NN/plant, PHT, RDW (g/plant) and RV (g/plant) of soybean genotypes grown under limed and unlimed acid soil at Mettu.

Treatments	SDW (g/plant)	NN/plant	PHT (cm)	RDW (g/plant)	RV /plant
Limed	5.46 ^a	35.72 ^a	47.64 ^a	0.735 ^a	2.34 ^a
Unlimed	4.12 ^b	27.22 ^b	42.57 ^b	0.625 ^b	1.88 ^b
PR	32.52	31.23	11.91	17.6	24.46

Means with the same letters are not significantly different at 5% level of significance. RDW- root dry weight; RV- root volume, SDW-shoot dry weight, PHT-plant height, NN-number nodule; PR = percent of reduction; CV- coefficient of variation

<https://doi.org/10.1371/journal.pone.0272924.t009>

number of nodule was recorded from SCS1 genotype. This might be due to liming effect on nodule weight and nodule numbers. There was one genotype showing an increase in number of nodules in unlimed ed acid soil condition. In line with this finding [11] reported two soybean genotypes i.e., H3 and PR-142 [15] showed the highest number of nodules per plant at 100 kg ha⁻¹ P with lime and Essex-1 genotype showed the lowest number of nodules per plant at lime untreated plot among the other tested genotypes. The highest plant height was recorded for PI567046A genotype both under lime treated and untreated acid soils. On the other hand, the shortest plant height was recorded for PI423958 genotype under lime untreated soil. This indicated that genotypes responded to liming, which might be due to the effect of liming that neutralized soil acidity, which in turn might have improved the availability of plant nutrients, particularly phosphorus and calcium and lowered the concentration of toxic cations, mainly Al³⁺ ions. The results are similar with the results of [33] who reported that a growth of plant is increased on acid soil in response to the application of lime.

The highest shoot dry weight was obtained from JM-CLK/G99-15-SB genotype, while the lowest shoot dry weight were recorded from SCS-1 genotype (Table 7). The reduction of shoot dry weight under the control or unlimed acidic soil condition might be due to Al toxicity, and

Table 10. Decrease percentage of yield, above ground biomass, number of pod and seed per plant, number of nodules, root dry weight and plant height of some soybean genotypes under unlimed acid soil conditions compared with limed acid soil.

Genotypes	YLD	AGB	NN	PHT	RDW	NPPP	NSPP
HAWASSA-04	1.50	17.0	13.9	9.13	0.00	21.6	31.6
PI567046A	44.96	54.0	38.4	12.51	42.7	43.4	39.5
PI423958	22.64	23.4	36.5	18.8	27.1	27.4	35.26
JMALM/PR142-15-SC	7.67	1.25	17.3	4.39	7.40	-5.2	0.52
JM-HAR/DAV-15-SA	6.31	5.11	1.39	12.95	0.00	9.69	7.18
JM-PR142/H3-15-SB	22.67	15.5	18.1	14.1	6.30	9.63	14.96
H-7	-6.38	6.84	12.4	7.5	7.90	15.6	7.06
BRS268	-15.4	5.41	-0.1	10.78	1.20	9.03	0.00
JM-H3/SCS-15-SG	-14.6	-1.3	25.5	8.32	11.3	0.36	1.66
JM-CLK/CRFD-15-SA	31.18	15.7	38.9	12.67	15.7	19.8	17.02
JM-ALM/H3-15-SC-1	2.40	10.7	48.6	9.43	8.50	10.9	9.72
JM-CLK/G99-15-SC	-4.39	-0.3	23.5	11.61	27.7	2.66	0.15
SCS-1	17.53	8.41	35.2	13.02	46.3	7.81	7.07
JM-CLK/G99-15-SB	29.66	4.91	37.5	5.52	31.3	4.97	-0.51
JM-DAV/PR142-15-SA	2.06	-2.5	2.26	7.87	2.70	-1.9	1.17

Where, NPPP = number pod per plant, NSPP = number of seeds per plant, PHT = plant height, RDW = root dry weight, YLD = yield, NN = number of nodules per plant, AGB = above ground biomass

<https://doi.org/10.1371/journal.pone.0272924.t010>

low Ca, Mg and P concentrations in the shoot, which resulted in decreased photosynthetic capacity that directly affected, shoot growth and developments. This alteration was also due to the low pH inhibits root growth; reduce Ca^{2+} and Mg^{2+} in the leaf and reduce rhizobia activity to form nodule [34] as well as the Mn toxicity. Different result was reported; [35] reported that decreasing solution pH and Ca concentration decreased the shoot dry weight. However, plant height, root volume, root dry weight, number of nodule and shoot dry weight is also affected by the genotypes. Lime applications to acid soil increased plant height, root volume, root dry weight, number of nodule and shoot dry weight of soybean genotypes by about 11.91, 24.47, 17.6, 31.22 and 32.5% respectively (Tables 9 and 10).

Correlation analysis

Grain yield was significantly ($P \leq 0.01$) and positively correlated with all root parameters viz., root dry weight and root volume and with all growth parameters viz., plant height and shoot dry weight and also with number of nodule at both limed and unlimed soil (Table 11). The significant and positive correlations of grain yield with the rooting parameters viz., root volume and root dry, under acid soil condition or under unlimed acid soil (hydrogen and aluminum toxicity) indicates the importance of the root parameters for acid soil tolerance. This also implies that selection for acid soil tolerance should consider these important root parameters. Similar to this finding [11] also reported the significant and positive associations of soybean grain yield with its root characters like root volume, root dry and fresh weight.

Grain yield is the product of its yield components, such as number of pods per plant, number of seeds per plant and above ground dry biomass were highly significant and positively correlated with its grain yield at both lime treated and untreated acid soil (Table 11). However, grain yield was strongly correlated with above ground biomass ($r = 0.90$), followed by number of seeds ($r = 0.87$) and number of pods per plant ($r = 0.82$) at limed soil among yield parameters, respectively. Other authors, such as [36, 37] reported that the significant associations of barley grain yield with its yield components. Results obtained in this study on soil treated with lime clearly showed that the remarkable increase in number of pods and seeds per plant, and greatly contributed to increase in grain yield of soybean. The negative correlation of number of nodules with number of seed and pod under unlimed soil (Table 11) indicates the competitiveness of these traits.

Conclusion

For the conclusion, the observed characters showed a different response in acid soil toxicity. The fifteen genotypes responded differently to acid soil. A preliminary field screening of soybean genotypes in south western Ethiopia has demonstrated the presence of genetic variability among genotypes in tolerating soil acidity stress. The observed characters of the sensitive genotypes decreased, while the tolerant genotypes could remain stable or increased. Root dry weight, root volume, number of nodule, plant height, shoot dry weight, grain yield, biomass, number of pod per plant, number of seed per plant, and hundred seed weight under unlimed soil were lower than in limed soil condition. However, not all these characters always decrease in unlimed soil condition. Their increments of grain yield in unlimed soil were found for BRS268 genotype. Genotype of BRS268 was the tolerant genotypes based on the reduction percentage of selected parameters. These results also give a clear indication that the grain yield was very closely associated with number of pods per plant, seed per plant, root dry weight, root volume, and shoot dry weight and number of nodule in both unlimed and limed soil. It seems that these parameters are useful characters to select for high yield in soybean breeding programs for soil acidity tolerance. Studying responses of selected genotypes with contrasting

Table 11. Pearson correlation analysis for growth, root, nodulation, yield and yield components of soybean genotypes grown under lime treated (1st) and lime untreated (2nd) soil on field at Mettu.

	YLD	PHT	NSPP	NPPP	AGB	SDW	RV	RDW	NN
YLD	1								
PHT	0.82**	1							
NSPP	0.87**	0.90**	1						
NPPP	0.82**	0.87**	0.95**	1					
AGB	0.90**	0.91**	0.92**	0.86**	1				
SDW	0.56**	0.21ns	0.31*	0.28 ^{ns}	0.31*	1			
RV	0.15 ^{ns}	0.15ns	0.04 ^{ns}	-0.07 ^{ns}	0.22 ^{ns}	-0.08 ^{ns}	1		
RDW	0.37*	0.38*	0.27 ^{ns}	0.23 ^{ns}	0.43**	-0.07 ^{ns}	0.55**	1	
NN	0.01 ^{ns}	-0.29 ^{ns}	-0.25 ^{ns}	-0.29 ^{ns}	-0.19 ^{ns}	0.43**	0.07 ^{ns}	-0.18 ^{ns}	1
	YLD	PHT	NSPP	NPPP	AGB	SDW	RV	RDW	NN
YLD	1								
PHT	0.52**	1							
NSPP	0.66**	0.75**	1						
NPPP	0.68**	0.67**	0.92**	1					
AGB	0.76**	0.55**	0.70**	0.67**	1				
SDW	0.63**	-0.13ns	0.24ns	0.35*	0.45**	1			
RV	0.48**	0.19ns	0.21ns	0.23ns	0.51**	0.34*	1		
RDW	0.59**	0.09ns	0.29ns	0.36*	0.61**	0.51**	1.0**	1	
NN	0.39**	-0.36*	-0.15ns	-0.08ns	0.07ns	0.70**	0.27ns	-0.18ns	1

Where, NPPP = number pod per plant, NSPP = number of seeds per plant, PHT = plant, Height, SDW = shoot dry weight, RDW = root dry weight, YLD = yield, NN = number of nodules per plant, AGB = above ground biomass

<https://doi.org/10.1371/journal.pone.0272924.t011>

tolerance to soil acidity may help in generating information that could be utilized by breeding programs aimed at developing aluminum-tolerant cultivars for areas where soil acidity remains a key environmental constraint to crop production. In conclusion identification and use of soybean genotypes that are tolerant to acid soil conditions of Southwestern Ethiopia is a very useful approach to ensure economic stability to many subsistence farmers, who cannot afford the application of liming materials practices.

Supporting information

S1 Table. Supporting information" files" raw data for each parameter.
(DOC)

Acknowledgments

The authors are grateful to Jimma agricultural research center for facilitating the research work and also acknowledge the staff members of Mettu research sub center for their technical assistance during the time of conducting the field experiment and data collection.

Author Contributions

Conceptualization: Tolossa Ameyu Bedassa, Abush Tesfaye Abebe, Alemayehu Regassa Tolessa.

Formal analysis: Tolossa Ameyu Bedassa.

Funding acquisition: Abush Tesfaye Abebe, Alemayehu Regassa Tolessa.

Investigation: Tolossa Ameyu Bedassa, Abush Tesfaye Abebe, Alemayehu Regassa Tolessa.

Methodology: Tolossa Ameyu Bedassa, Abush Tesfaye Abebe, Alemayehu Regassa Tolessa.

Project administration: Tolossa Ameyu Bedassa, Abush Tesfaye Abebe.

Software: Tolossa Ameyu Bedassa.

Supervision: Tolossa Ameyu Bedassa, Abush Tesfaye Abebe, Alemayehu Regassa Tolessa.

Validation: Tolossa Ameyu Bedassa, Abush Tesfaye Abebe.

Visualization: Tolossa Ameyu Bedassa, Abush Tesfaye Abebe, Alemayehu Regassa Tolessa.

Writing – original draft: Tolossa Ameyu Bedassa.

Writing – review & editing: Tolossa Ameyu Bedassa.

References

1. Fageria K, Baligar C. Ameliorating soil acidity of tropical Oxisols by liming for sustainable crop production. *Advances in agronomy*, 2008. 99: 345–399.
2. Brady C, Weil R., et al. The nature and properties of soils, 13th. *Pearson education (Singapore) Pte. Ltd. Indian Branch*, 2002. 482: 621–624.
3. Takala, B. Soil Acidity and Its Management Options in Western Ethiopia, 2019.
4. Moody W and Cong P., 2008. Soil constraints and management practice: guidelines for Sustainable management for tropical upland soils. Australian Center for Agricultural Research (ACIAR), Australia 86p
5. Delhaize E., Gruber D. and Ryan R., et al. The roles of organic anion permeases in aluminium resistance and mineral nutrition. *Febs Letters*, 2007. 581: 2255–2262. <https://doi.org/10.1016/j.febslet.2007.03.057> PMID: 17418140
6. Wang P., Raman H., Zhang P., Mendham N. and Zhou X., et al. Aluminium tolerance in barley (*Hordeum vulgare L.*): physiological mechanisms, genetics and screening methods. *Journal of Zhejiang University SCIENCE B*, 2006. 7:769–787.
7. Kochian V., Hoekenga A. and Pineros A., 2004. How do crop plants tolerate acid soils? Mechanisms of aluminum tolerance and phosphorous efficiency. *Annu. Rev. Plant Biol.*, 55: 459–493 <https://doi.org/10.1146/annurev.arplant.55.031903.141655> PMID: 15377228
8. Von Uexkull R, Mutert E., et al. Global extent, development and economic impact of acid soils. *Plant and soil*, 1995. 171:1–15.
9. Mesfin A. Nature and management of acid soils in Ethiopia. Addis Ababa, Ethiopia, 2007. 99p.
10. Shahid Q., Saleem F., Khan Z. and Anjum A., et al. Performance of soybean (*Glycine max L.*) under different phosphorus levels and inoculation. *Pakistan Journal of Agricultural Sciences*, 2009. 46: 237–241.
11. Abush T., Githiri M., Derera J. and Tolessa D., et al. Genetic variability in soybean (*Glycine max L.*) for low soil phosphorus tolerance. *Ethiopian Journal of Agricultural Sciences*, 2017. 27:1–15.
12. Anetor O. and Akinrinde E., et al. Response of soybean [*Glycine max (L.) Merrill*] to lime and phosphorus fertilizer treatments on an acidic alfisol of Nigeria. *Pakistan Journal of Nutrition*, 2006. 5: 286–293.
13. Okpara D, Muoneke C., et al. Influence of lime on the performance of high-yielding soybean varieties in southern Nigeria. *Agro-Science journal*. 2007. ISSN: pp. 1119–7455
14. Uguru M. I., Oyiga B. C., & Jandong E. A. (2012). Responses of some soybean genotypes to different soil pH regimes in two planting seasons. *The African Journal of Plant Science and Biotechnology*, 6, 26–37.
15. Rao I.M., 2001. Role of physiology in improving crop adaptation to a biotic stresses in the tropics: The case of common bean and tropical forages. *Handbook of plant and crop physiology*, pp.583–613.
16. Rengel Z. and Zhang W.H., 2003. Role of dynamics of intracellular calcium in aluminium-toxicity syndrome. *New Phytologist*, 159(2), pp.295–314 <https://doi.org/10.1046/j.1469-8137.2003.00821.x> PMID: 33873357
17. Baruah C, Barthakur P. et al. *A Textbook of Soil analysis*, Vikas Publishing House, New Delhi, India, 1997.

18. Bouyoucos J. Hydrometer Method Improved for Making Particle Size Analyses of Soils1. *Agronomy journal*, 1962. 54: 464–465.
19. Bray H, Kurtz T, et al. Determination of total, organic, and available forms of phosphorus in soils. *Soil science*, 1945. 59:39–46.
20. McLean O. Aluminum. In: Black C.A.(ed.), *Methods of Soil Analysis*. Agronomy, No.9. Part II. American Society Agronomy, Madison, Wisconsin. USA, 1965. pp 978–998.
21. Black A. *Methods of soil analysis*. Part I, American Society of Agronomy. Madison, Wisconsin, USA. 1965. 1572p.
22. Chapman HD. Cation exchange capacity by ammonium saturation. In: Black CA, editor. *Methods of soil analysis*. Agronomy Part II, No. 9. Madison: American Society of Agronomy; 1965.
23. Van Reeuwijk P., 1992. *Procedures for soil analysis*, (3rd ed.), International soil reference and information center (ISRIC), Wageningen, and the Netherlands. 34p.
24. Shoemaker E., Mclean O. and Pratt F., et al. Buffer methods for determining lime requirement of soils with appreciable amounts of extractable aluminum 1. *Soil Science Society American*. Proceeding, 1961. 25: 274–277.
25. SAS (Statistical Analysis System) soft ware, 2012. Version9.3, SAS institute, Cary, NC, USA
26. Temasgen D., Alemu G., Adella A. and Tolessa D., et al. Effect of lime and phosphorus fertilizer on Acid soils and barley (*Hordeum vulgare L.*) performance in the central highlands of Ethiopia. *Experimental Agriculture*, 2017. 53: 432–444.
27. Workneh B and Asfaw H., 2012. Influence of Inoculation Methods and Phosphorus Levels on Nitrogen Fixation Attributes and Yield of Soybean (*Glycine max L.*) at Haru, Western Ethiopia. *American Journal of Plant Nutrition and Fertilization Technology*, 2: 45–55.
28. Tigist A., 2017. Soybean (*Glycine max L.*) Response to Lime and Vermicompost Amelioration of Acidic Nitisols of Assossa, North Western Ethiopia. MSc Thesis Haramaya University
29. Habtamu A., 2017. Genotype x Environment x Management Interaction of Common Bean (*Phaseolus vulgaris L.*) on Acidic Soils of Western Ethiopia. An MSc Thesis Presented to the School of Graduate studies of Haramaya University.
30. Bambara S, Ndakidemi A., et al. Phaseolus vulgaris response to Rhizobium inoculation, lime and molybdenum in selected low pH soil in Western Cape, South Africa. *African Journal of Agricultural Research*, 2010. 5: 1804–1811.
31. Kuswanto H. Tolerance of fifteen soybean germplasm to low pH condition. *Intl J Plant Breed Genetics*, 2015. 9: 189–197.
32. Haling E., Simpson J., Culvenor A., Lambers H. and Richardson E., et al. Effect of soil acidity, soil strength and macropores on root growth and morphology of perennial grass species differing in acid-soil resistance. *Plant, Cell & Environment*, 2011. 34: 444–456. <https://doi.org/10.1111/j.1365-3040.2010.02254.x> PMID: 21062319
33. Kisinyo O. Long term effects of lime and phosphorus application on maize productivity in an acid soil of Uasin Gishu County, Kenya. *Sky Journal of Agricultural Research*, 2016. 5: 48–55.
34. Liang C., Pinero's A., Tian J., Yao Z., Sun L., Liu J., et al. Low pH, aluminum, and phosphorus coordinately regulate malate exudation through GmALMT1 to improve soybean adaptation to acid soils. *Plant Physiology*, 2013. 161: 1347–1361. <https://doi.org/10.1104/pp.112.208934> PMID: 23341359
35. Murata R, Hammes S., Zharare E., et al. Effect of solution pH and calcium concentration on germination and early growth of groundnut. *Journal of plant nutrition*, 2003. 26: 1247–1262.
36. Ortiz-Monasterio P., H. Pfeiffer, H. Hede, et al. Phosphorus use efficiency, grain yield and quality of triticale and durum wheat under irrigated conditions In: *Proceedings of the 5th International Symposium*, Annex, Radzikow, Poland, 2002.
37. Abeledo G., Calderini F. and Slafer A., et al. Genetic improvement of barley yield potential and its physiological determinants in Argentina (1944–1998). *Euphytica*, 2003. 130:325–334.