Research Article *http://dx.doi.org/10.4314/mejs.v18i1.9* *Open Access*

Effect of Soil Liming on Microbial Diversity, Distribution, and Composition

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ABSTRACT

Soil acidity is a major constraint to agricultural productivity in Northwest Ethiopia, where acidic soils limit nutrient availability and reduce crop yields. Liming, a widely adopted practice to neutralize soil acidity, improves soil pH and alters physicochemical properties such as nutrient solubility and organic matter dynamics. These changes influence microbial communities that are central to nutrient cycling, soil fertility, and ecosystem health. This study aimed to evaluate the effects of liming on microbial diversity, distribution, and composition in the Debark area of the North Gondar Zone, Northwest Ethiopia. Two farmland sites, Miligebsa and Yekrar, were selected for a cross-sectional study. Five soil samples (200 g each) were randomly collected from 12 m × 12 m plots before and after liming, composited into 1 kg samples, and serially diluted for culturing. Bacteria were isolated on nutrient agar with cycloheximide, and fungi on PDA with chloramphenicol and streptomycin. Bacterial isolates were identified using microscopy, colony morphology, and biochemical tests. Data were analyzed using SPSS version 20.0. Liming significantly increased soil pH from ~5.7 to ~7.4, while moisture content remained stable. Bacterial counts rose significantly ($p < 0.05$) at both sites, from ~90 to ~136 CFU/mL, whereas fungal counts declined ($p < 0.05$), from 15.70 to 4.10 CFU/mL at Yekrar and from 12.10 to 6.00 CFU/mL at Miligebsa. *Pseudomonas* spp. dominated before liming but was replaced by *Bacillus* spp. afterwards, while *Aspergillus* spp. remained the dominant fungi, though its abundance decreased. Statistical analyses confirmed significant shifts in microbial communities, with a transition from fungal to bacterial dominance. Liming proved to be an effective soil reclamation strategy, raising pH, enhancing bacterial abundance, reducing fungal populations, and improving nutrient cycling and soil fertility. These findings highlight soil pH as a key regulator of microbial structure and demonstrate liming's role as a low-cost, practical approach for restoring degraded acidic soils and supporting sustainable agriculture in smallholder systems of Northwest Ethiopia. Future research should monitor long-term impacts of liming, optimize application strategies, and integrate liming with complementary soil management practices. Functional and crop-specific analyses, socioeconomic assessments, regional expansion, and stronger extension-policy linkages are essential to advance sustainable soil management and ensure widespread adoption among smallholder farmers.

Keywords: Acidic soil, Microbial Composition, Distribution, Liming, Microbial diversity.

1. INTRODUCTİON

Soil acidification adversely affects microbial ecosystems by altering the soil pH (Symanowicz and Toczko, 2022). Soil pH is affected by global changes through agricultural intensification, climate change, and other polluting events (e.g. acid rain) (Fatma and Ahmet, 2023; Wang et al., 2022; Wei et al., 2020). Liming has been suggested as one of the methods used to solve this

Submitted: 24th February 2025 ***Accepted: 11th February 2026*** ***Published: 2nd May 2026***

problem by significantly increasing the soil pH, resulting changes in chemical and biochemical reactions and in microbiological processes (Cha et al., 2021). Liming is a common acidic soil amendment technique that directly neutralizes soil acidity and alleviates heavy metal toxicity (Ding et al., 2023). Liming also plays an important role in improving soil structure, enhancing soil biological activity and enhancing nutrient cycling capacity (Dang et al., 2022; Ding et al., 2023). It has been found that liming can significantly reduce the content of soil toxicity and improve soil fertility by improving the activity of nitrifying microorganisms. However, excess lime in turn can significantly reduce soil fertility (Ding et al., 2023; Yusron and Susilawati, 2022). Soil pH has been shown to be controlled by biotic factors such as the microbial community composition, distribution, and diversity to a predictable level across various spatial scales (Cha et al., 2021).

Local soil characteristics and land management practices play a critical role in shaping bacterial and fungal communities in the Debark district (Zheng et al., 2019). The volcanic soils, with their loamy to clay loam texture and moderate organic matter, provide a mineral-rich environment that supports diverse microbial populations, though steep slopes and erosion risk can deplete topsoil and reduce diversity (Ewunetu et al., 2025). Rainfall of 1,126 mm annually ensures moisture that favors fungal growth, while seasonal variability influences bacterial dynamics through nutrient leaching and changes in aeration. Cultivation history also affects microbial composition, as continuous cereal and legume production selects for nitrogen-cycling bacteria, while crop residues sustain fungal decomposers (Chen et al., 2024). Practices such as crop rotation, mulching, and terracing help maintain soil fertility, reduce pathogen buildup, and preserve microbial habitats, whereas intensive cultivation without conservation measures disrupts microbial balance. The interplay of soil type, climate, and management strategies determines the abundance, diversity, and ecological functions of bacteria and fungi in these highland agricultural systems (Al-Shammary et al., 2024).

Acidic soil is a major bottleneck for increasing crop yield and a limiting factor for acid-sensitive crop production in the Debark district, Northern Gondar zone, North-western Ethiopia. Sustainable crop production and reasonable yields require treating and correcting acidic soils through liming to a pH range suitable for optimal crop growth. Liming is practiced in the Debark area to adjust the soil pH of farmlands. Therefore, this study aimed to evaluate the effect of liming on microbial composition, distribution, and diversity in a context where the status and interrelationships between soil management practices (liming) and soil microbial activities have not received sufficient emphasis in the district's research and development strategies. Given the absence of prior studies in the Debark area, we hypothesized that liming

would significantly alter microbial community composition and distribution. To test this, we sampled two farmland sites before and after lime application. This study aimed to evaluate the effects of liming on microbial activity and to provide scientific evidence that supported agricultural extension efforts in Debark and the surrounding region. Furthermore, the findings served as a foundation for future research exploring the relationships between microbial activity, soil improvement, and crop productivity.

2. MATERIALS AND METHODS

2.1. Description of the Study Area

This study was conducted at the Miligebsa (Site 1) and Yekirar (Site 2) farmland sites in the Debark district, located 100 km north of Gondar and adjacent to the Semien Mountains National Park. Geographically, Debark lies between 13°0'08"N and 13°30'N latitude and 37°30'E and 38°15'E longitude, covering a total area of 282,105 hectares (282.105 km²) with an average annual temperature of 12.5 °C. The soils of the district are typical highland soils of Northwest Ethiopia, derived from volcanic parent materials and characterized by a loamy to clay loam texture with moderate fertility due to organic matter accumulation. Although these soils are productive, the steep slopes make them vulnerable to erosion under intensive cultivation. With an average annual rainfall of 1,126 mm and a cool climate, they are well suited for cereal and legume production, but sustainable practices such as terracing, mulching, and crop rotation are necessary to preserve long-term fertility (Hindersah et al., 2018).

2.2. Soil Sampling

Two farmlands were selected based on information provided by agricultural extension workers in the Debark district. At each site, five 12 m × 12 m plots were prepared. Before and after liming, five soil samples (200 g each) were randomly collected from each plot at a 15 cm depth and placed in sterilized polyethylene bags. The five samples from each site and time point were then combined (1 kg) and mixed homogeneously (Cha et al., 2021).

2.3. Isolation of Soil Microbes

One gram of soil was diluted in 9 mL of 0.85% saline solution and vortexed. 1 ml of this solution was serially transferred to create a final volume of 10 mL, which was then serially diluted to 10^{-1}, 10^{-2}... 10^{-10}. Subsequently, 0.1 mL aliquots from each dilution were aseptically spread onto nutrient agar and potato dextrose agar (PDA) and incubated at 25°C for 72 hours (fungi) and 37°C for 24 hours (bacteria). After incubation, plates were examined for growth, and colonies were counted (Gebreyohannes et al., 2013). Pure bacterial colonies were preserved in 5 mL of slant nutrient agar containing 20% glycerol at -20°C for further study.

Bacterial isolates were identified using selective and differential media (MacConkey, MSA, and EMB), biochemical tests (catalase, MR-VP, SIM, indole, and citrate), Gram staining, and colony morphology. Bergey's Manual of Determinative Bacteriology was used as a reference (Tassadaq et al., 2013). Fungal isolates were cultured on PDA supplemented with chloramphenicol (0.035 g/L) and incubated at 25°C for one to two weeks. Colonies were counted and sub-cultured on PDA using the streak plate method to obtain pure cultures. Fungal isolates were identified morphologically and using the cotton lactophenol staining technique (Osuntokun et al., 2018).

2.4. Data Analysis

SPSS version 20 software was used analyse data, to organize and process the collected information. Descriptive statistics were employed to summarize the findings, with percentages used to represent categorical variables and means applied to continuous variables.

3. RESULTS

Table 1 summarizes the number of microbial isolates, soil pH, and moisture content at Yekrar and Miligebsa before and after liming, showing that lime application significantly altered microbial communities at both sites. Bacterial counts increased markedly following liming, rising in Yekrar from 90.31 ± 0.23 to 136.5 ± 0.67 CFU/mL alongside a pH shift from 5.76 to 7.41, while moisture content remained stable (20.38–20.40%), and in Miligebsa from 90.10 ± 0.54 to 135.10 ± 0.54 CFU/mL with pH increasing from 5.67 to 7.35 and moisture unchanged at 36.98%. In contrast, fungal populations declined significantly, decreasing in Yekrar from 15.70 ± 1.10 to 4.10 ± 0.62 CFU/mL and in Miligebsa from 12.10 ± 1.36 to 6.00 ± 1.61 CFU/mL after liming. hese significant changes ($p \leq 0.05$) demonstrate that liming altered microbial community structure by enhancing bacterial growth and reducing fungal populations, with soil pH as the main influencing factor, while moisture content showed minimal variation.

Table 1. Effects of liming on microbial abundance, soil pH, and moisture in Yekrar and Miligebsa Farmlands.

Isolates	***Sampling Sites***	***Liming***	***CFU/mL (Mean ± SD)***	***pH***	***Moisture (%)***
Bacteria	Yekrar	Before liming	90.31 ± 0.23	5.76	20.38
		After liming	136.5 ± 0.67	7.41	20.40
	Miligebsa	Before liming	90.10 ± 0.54	5.67	36.98
		After liming	135.10 ± 0.54	7.35	36.98
Fungi	Yekrar	Before liming	15.70 ± 1.10	5.76	20.38
		After liming	4.10 ± 0.62	7.41	20.40
	Miligebsa	Before liming	12.10 ± 1.36	5.67	36.98
		After liming	6.00 ± 1.61	7.35	36.98

Note: Mean values with different superscripts before and after liming in the same site imply statistically significant difference at $p \leq 0.05$.

Microbial communities at Yekrar and Miligebsa differed significantly both before and after liming, with statistically significant differences in bacterial and fungal isolate numbers between the two sites ($p < 0.05$). Table 2 details this distribution, showing notably higher bacterial populations at Miligebsa (29.00 ± 2.95) compared to Yekrar (8.50 ± 3.03) before liming. Conversely, before liming, fungal populations were higher at Yekrar (11.60 ± 4.43) than at Miligebsa (6.10 ± 3.17).

Table 2. Distribution of Bacterial and Fungal Isolates Before and After Liming at the Yekrar and Miligebsa Farmlands.

Isolates	***Sampling sites***	***Variables***	***Mean ± SE***
Bacteria	Yekrar	Before and after liming	8.50 ± 3.03
	Miligebsa	Before and after liming	29.00 ± 2.95
Fungi	Yekrar	Before and after liming	11.60 ± 4.43
	Miligebsa	Before and after liming	6.10 ± 3.17

Note: SE: Standard Error.

Before liming, the average number of bacterial isolates was remarkably similar at both Yekrar and Miligebsa (Table 3). This similarity is confirmed by a two-sample independent t-test, which found no significant difference in bacterial distribution between the sites before liming. Table 3 further shows the results of t-tests (using both pooled and Satterthwaite variances) comparing bacterial distribution at the two sites before and after liming. Consistently, no significant difference in bacterial distribution was found between Yekrar and Miligebsa, either before or after liming, regardless of the variance assumption.

Table 3. Distribution of Bacteria Before and After Liming at the Yekrar and Miligebsa Study Sites.

Treatment	***Variable***	***Method***	***Variances***	***t- value***	***p- value***
Before liming	Bacteria	Pooled	Equal	-0.4	0.69
		Satterthwaite	Unequal	-0.4	0.69
After Liming	Bacteria	Pooled	Equal	-0.46	0.65
		Satterthwaite	Unequal	-0.46	0.65

Table 4 shows fungal distribution before and after liming at Yekrar and Miligebsa. Before liming, mean fungal distribution was 13.70 ± 22.11 CFU/mL (65 total) at Yekrar and 13.10 ± 22.15 CFU/mL (67 total) at Miligebsa. After liming, these decreased to 4.10 ± 9.62 CFU/mL (30 total) at Yekrar and 4.40 ± 10.61 CFU/mL (33 total) at Miligebsa, indicating reduced fungal abundance at both sites. Despite this reduction, fungal distribution remained remarkably similar between Yekrar and Miligebsa both before and after liming. This similarity is confirmed by statistical analysis, which showed no significant mean difference in fungal distribution between the two sites after liming ($p > 0.05$).

Table 4. Distribution of Fungi Before and After Treatment at the Two Sampling Sites.

Treatment	***Sampling sites***	***Mean ± SD***	***CFU/mL***
Before Liming	Yekrar	13.70 ± 22.11	65
	Miligebsa	13.10 ± 22.15	67
After Liming	Yekrar	4.10 ± 9.62	30
	Miligebsa	4.40 ± 10.61	33

Note*:* SD: Standard Deviation.

Statistical analysis showed no significant difference in fungal distribution between the Yekrar and Miligebsa sites, both before and after liming. This suggests that, despite potential species variations, overall fungal abundance was statistically similar at both locations throughout the study. This is supported by table 5, where the t-test results (using both pooled and Satterthwaite variance estimations) are showing no significant difference in fungal distribution between sites, regardless of variance assumptions.

Table 5. t-test results for fungal counts before and after soil liming under equal and unequal variance assumptions.

Treatment	***Variable***	***Method***	***Variances***	***t–value***	***p–value***
Before Liming	Fungi	Pooled	Equal	-0.26	0.79
		Satterthwaite	Unequal	-0.26	0.79
After Liming	Fungi	Pooled	Equal	0.40	0.69
		Satterthwaite	Unequal	0.40	0.69

Based on morphological, Gram reaction, and biochemical tests, both Gram-positive and Gram-negative bacteria were isolated from both study areas. *E. coli* spp., *Clostridium* spp., *Pseudomonas* spp., and *Bacillus* spp. were isolated (Table 6). Before liming, *Pseudomonas* spp. was the dominant bacterium, followed by *Bacillus* spp., with mean distribution values of 63.0 ± 8.48 CFU/mL and 57.5 ± 6.36 CFU/mL, respectively. *Clostridium* spp. was more affected by the acidic soil conditions, with a mean distribution of 28.5 ± 7.78 CFU/mL. After liming, *Bacillus* spp. became the highly dominant bacterium (141.5 ± 0.78 CFU/mL). *E. coli*, which was severely affected by the initial acidic soil conditions, had a mean distribution of 75 ± 43.84 CFU/mL after liming. There was a statistically significant difference in the number of bacterial isolates before and after liming ($p < 0.05$).

Table 6. Mean Distribution of Bacterial Isolates Before and After Liming.

Treatment	***Bacterial species***	***CFU/mL (Mean ± SD)***
Before limning	*Bacillus* spp.	57.5 ± 6.36
	Clostridium spp.	28.5 ± 7.78
	E. coli spp.	52.0 ± 5.66
	Pseudomonas spp.	63.0 ± 8.48
After liming	*Bacillus* spp.	141.5 ± 0.78
	Clostridium spp.	101.5 ± 7.78
	E. coli spp.	75 ± 43.84
	Pseudomonas spp.	118.5 ± 7.78

Table 7 compares the abundance of bacterial isolates (*Bacillus*, *Pseudomonas*, *Clostridium*, *E. coli*), showing mean differences and confidence intervals. *Bacillus* and *Pseudomonas* abundance differed significantly from *E. coli*, but *Clostridium* did not. Diversity was similar between *Bacillus* and *Pseudomonas*, and between *Clostridium* and *E. coli*. *Bacillus* and *E. coli* differed significantly in diversity and distribution with a highly significant difference in CFU distribution (Dunnett's test).

Table 7. Statistical Mean Difference among Bacterial Isolates.

Comparison of Isolates	***Differences between means***		***95% confidence limits***
Bacillus spp. Vs *E. coli*	36	1.41	70.59***
Pseudomonas spp. Vs *E. coli*	27.25	-7.34	61.84
Clostridium spp. Vs *E. coli*	1.50	-33.09	36

Note: Comparisons significant at the 0.05 level are indicated by ***.

Table 8 shows the mean differences in colony-forming units (CFU/mL) of fungal isolates (*Aspergillus* spp., *Penicillium* spp., and *Rhizopus* spp.) before and after liming. Before liming, *Aspergillus* spp. had the highest mean CFU/mL (53.5 ± 10.61), followed by *Penicillium* spp. (34 ± 5.66) and *Rhizopus* spp. (14.5 ± 3.54). After liming, the mean CFU/mL decreased for all fungal isolates: *Aspergillus* spp. to 17.5 ± 2.12, *Penicillium* spp. to 9.5 ± 2.12, and *Rhizopus* spp. to 5.5 ± 2.12. These results indicate a significant decrease in the mean CFU/mL of all fungal isolates after liming, with *Aspergillus* spp. experiencing the largest reduction, followed by *Penicillium* spp. and *Rhizopus* spp. Descriptive statistical analysis showed high diversity or composition of fungal isolates before liming, and a statistically significant difference in fungal isolate diversity before and after liming. There was also a significant difference among all fungal isolates in their diversity, number, and composition. After liming, the number of fungal isolates decreased, with *Aspergillus* spp. remaining dominant and *Rhizopus* spp. being the least abundant.

Table 8. Mean difference of Fungal Isolates Before and After Liming.

Treatment	***Fungal Isolates***	***CFU/mL (Mean ± SD)***
Before liming	*Aspergillus* spp	53.5 ± 10.61
	Penicillium spp	34 ± 5.66
	Rhizopus spp	14.5 ± 3.54
After liming	*Aspergillus* spp	17.5 ± 2.12
	Penicillium spp	9.5 ± 2.12
	Rhizopus spp	5.5 ± 2.12

Table 9 presents a comparison of different fungal isolates (*Aspergillus* spp., *Penicillium* spp., and *Rhizopus* spp.) based on the differences in their mean values and 95% confidence

limits. The comparisons highlight the statistical differences between the means of each pair of fungal isolates, indicating the significance of the difference between species. For example, *Aspergillus* spp. vs. *Penicillium* spp. shows a mean difference of 13.75, with a 95% confidence interval ranging from 4.53 to 22.97, indicating a statistically significant difference. Similarly, *Aspergillus* spp. vs. *Rhizopus* spp. shows a highly significant difference, with a mean difference of 25.50 and a confidence interval of 16.27 to 34.72. *Penicillium* spp. vs. *Aspergillus* spp. has a mean difference of -13.75 and a confidence interval ranging from -22.97 to -4.54, also statistically significant. *Penicillium* spp. vs. *Rhizopus* spp. demonstrates a significant difference, with a mean difference of 11.75 and a confidence interval of 2.53 to 20.97. *Rhizopus* spp. vs. *Aspergillus* spp. shows a significant difference, with a mean difference of -25.50 and a confidence interval of -34.72 to -16.27. Finally, *Rhizopus* spp. vs. *Penicillium* spp. indicates a statistically significant difference, with a mean difference of -11.75 and a confidence interval ranging from -20.97 to -2.53.

Table 9. Statistical Mean Difference among Fungi Isolates.

Comparison of Fungal Isolates	***Difference between means***		***95% confidence limits***
Aspergillus spp. Vs *Penicillium* spp	13.75	4.53	22.97***
Aspergilus spp Vs *Rhizopus* spp	25.50	16.27	34.72***
Penicillium spp Vs *Aspergilus* spp	-13.75	-22.97	-4.54***
Penicillium spp *Vs Rhizopus* spp	11.75	2.53	20.97***
Rhizopus spp Vs *Rhizopus* spp	-25.50	-34.72	-16.27***
Rhizopus spp Vs *Penicillium* spp	-11.75	-20.97	-2.53***

Note: Comparisons significant at the 0.05 level are indicated by ***.

4. DİSCUSSİON

Soil pH strongly influences microbial community structure and function. Microbes exhibit varying pH preferences, impacting their survival and activity. Soil pH affects key microbial processes like enzyme function, nutrient uptake, and cell stability, ultimately determining which microbial groups thrive. This has profound implications for soil processes such as nutrient cycling and decomposition, affecting plant health. Understanding this pH-microbe relationship is crucial for soil management, predicting environmental impacts, and ensuring ecosystem sustainability, given pH's cascading effects on soil fertility and plant growth (Zhang et al., 2022). The present study's results, showing a shift from acidic to neutral soil pH after liming, align with previous study demonstrating that lime application to agricultural fields effectively increases soil pH, thereby maintaining soil quality and favoring the growth of diverse microbial populations (Swer et al., 2011). Liming increases soil pH toward neutrality,

enhancing nutrient cycling, fertility, and crop yield by promoting beneficial microbes and restoring diversity. As a low-cost, sustainable practice, it reduces dependence on chemical inputs, improves long-term soil quality, and requires regular monitoring for site-specific management, especially in erosion-prone areas like Debark.

In the present study, the shift in soil pH from acidic to near-neutral significantly altered the distribution and composition of the bacterial community. Prior to liming, the acidic pH likely limited bacterial communities due to selective pressure. However, after liming, the more neutral pH provided a selective advantage, resulting in a significant increase in the number of all bacterial groups studied. This observation supports previous findings that demonstrated a strong correlation between soil pH and bacterial community composition across a wide range of pH values (Ding et al., 2023). Adjusting soil pH through liming enhances microbial activity, nutrient cycling, and soil fertility by promoting beneficial bacteria that improve crop productivity and alleviate acid stress. It restores microbial balance and diversity, strengthens ecosystem resilience, and offers a low-cost, sustainable approach that reduces dependence on chemical inputs. The strong relationship between soil pH and bacterial composition underscores the need for continuous monitoring and site-specific lime application, particularly in erosion-prone highland areas like Debark, where soil variability can affect long-term soil quality and management outcomes.

Soil pH directly influences bacterial community composition due to the narrow pH growth tolerances of bacterial taxa. Each species thrives within a specific pH range; outside this range, growth is limited or impossible. The pH acts as a strong selective pressure, favouring adapted taxa and shaping distinct bacterial communities in varying pH soils. These narrow tolerances are likely due to pH's impact on enzyme activity, cell membrane stability, and nutrient availability, all crucial for bacterial growth. Thus, bacterial distribution and abundance are directly linked to soil pH (Jiang and Zeng, 2022; Peng et al., 2022). Soil pH strongly shapes bacterial communities, making its regulation through practices like liming essential for enhancing nutrient cycling, soil fertility, and crop productivity. Because microbial diversity is highly sensitive to pH shifts, maintaining optimal levels supports ecosystem stability, pathogen suppression, and sustainable yields. Proper pH management reduces reliance on chemical inputs and promotes environmental sustainability, while continuous monitoring of soil pH and microbial responses is vital for guiding land management and site-specific interventions in regions such as Debark.

Among the bacterial isolates, *Bacillus spp.* was the most dominant Gram-positive bacterium. This dominance is likely due to their versatile enzyme production and broad

substrate degradation capabilities. Their diverse extracellular enzymes (amylases, proteases, cellulases, lipases, and xylanases) break down complex organic molecules into usable nutrients, allowing them to thrive in diverse environments. This ability to degrade numerous substrates, including plant cell walls and polysaccharides, provides a competitive advantage, especially in complex soil environments. This metabolic flexibility, along with the production of beneficial compounds like antimicrobials, further enhances their survival and contributes to their dominance (Danilova and Sharipova, 2020). Other studies report that Gram-positive bacteria are often more abundant in limed soils due to their association with plant-derived carbon and their ability to utilize recalcitrant compounds. Their diverse enzymatic systems enable efficient degradation of complex polymers such as lignin and tannins, giving them a competitive advantage in plant-rich environments. Liming further enhances organic matter breakdown, supporting their proliferation. Thus, access to plant carbon sources and the capacity to degrade complex substrates largely explain their increased abundance in limed soils (Goupil et al., 2015; Mencel et al., 2022; Narendrula-Kotha and Nkongolo, 2017).

The types of bacterial species obtained after liming showed slight differences from those in the unlimed soil samples. Liming altered the bacterial species composition, likely due to the lime-induced pH change. This pH shift acted as a selective pressure, favouring bacteria adapted to near-neutral conditions and potentially disadvantaging acidophiles. Different bacterial species have varying pH tolerances and optima, influencing their survival and proliferation. Species may also employ physiological or genetic adaptations to cope with the changing pH. This shift in community structure aligns with other studies showing increased microbial activity and diversity after liming, as it creates a more hospitable environment for a broader range of microorganisms (Madegwa and Uchida, 2021; Yin et al., 2021a).

After liming, the number of bacterial isolates increased while fungal isolates decreased. This shift in the microbial community is primarily due to the pH change from acidic to near-neutral, which favours bacterial growth. Bacteria generally have wider pH tolerances than fungi, and the near-neutral pH after liming is more conducive to bacterial proliferation. Conversely, many fungi prefer acidic conditions, and the pH shift may hinder their growth. Liming acts as a selective pressure, favouring bacteria adapted to the altered pH and potentially hindering acidophilic fungi (Kamrun Nahar et al., 2022). The greater number of fungal isolates before liming correlated directly with the physiological nature of fungi, as they can withstand and grow in acidic environments better than bacteria. The distribution of fungi after liming significantly decreased due to the change in soil pH to near-neutral (resulting from the decrease in soil acidity).

In corroboration with our findings, previous studies have shown that fungi often dominate in low-pH environments due to their physiological adaptation to acidic conditions, enabling them to outcompete bacteria, which generally prefer neutral or slightly alkaline soils. Acidic conditions not only promote fungal growth and proliferation but also suppress bacterial activity, thereby reinforcing fungal dominance (Janowski and Leski, 2022; Nguyen, 2023). This imbalance can reduce nutrient cycling and soil fertility, ultimately constraining crop productivity. Since many beneficial bacteria thrive under neutral pH, regulating soil acidity through practices such as liming is essential to restore microbial balance, enhance bacterial diversity, and build resilient soil ecosystems that support sustainable land management.

Other studies also showed a decreased abundance of fungal species in limed soil samples, suggesting that fungal activity was limited beyond acidic environments. Different fungal species react differently to liming. As soil pH shifts from acidic to alkaline conditions, nutrients like calcium and magnesium become more readily available to plants and microbial populations (Li et al., 2022; Yin et al., 2021b). The observed decrease in fungal abundance in limed soils, coupled with species-specific responses to liming and increased availability of nutrients like calcium and magnesium at higher pH, has significant implications for soil ecology. These findings suggest that soil pH is a key driver of fungal community structure and activity, with liming practices potentially altering fungal diversity and function. Such shifts in fungal communities can impact crucial processes like nutrient cycling and decomposition, ultimately affecting plant health and overall ecosystem function, highlighting the need for careful consideration of liming strategies in agricultural and ecological management.

5. CONCLUSION

This study demonstrated that liming significantly improved soil properties and reshaped microbial communities in the Debark district by raising soil pH from acidic to near-neutral at both Yekrar and Miligebsa, which increased bacterial abundance (from ~90 to ~136 CFU/mL, $p < 0.05$) while reducing fungal counts. *Bacillus* spp. became the dominant bacteria after treatment, replacing *Pseudomonas* spp., whereas *E. coli* was more sensitive to acidic conditions; *Aspergillus* spp. remained the leading fungus, though overall fungal diversity declined. These shifts, particularly the rise of beneficial bacteria, suggest enhanced nutrient cycling and soil fertility with potential benefits for crop productivity. The findings highlight liming as a practical solution to agricultural challenges in Debark, where acidic soils and erosion constrain productivity, as it fosters beneficial bacteria, suppresses acid-tolerant fungi, improves soil health, stabilizes microbial communities, and supports cereal and legume

production. When combined with conservation practices such as terracing, mulching, and crop rotation, liming helps restore soil fertility, reduce erosion, and strengthen food security in highland farmlands. Farmers in acidic soils should adopt liming to improve pH and microbial balance. Promoting *Bacillus* spp. as biofertilizers and monitoring fungal diversity are important for ecological stability. Site-specific strategies, integration with crop studies, long-term monitoring, and combining liming with organic amendments are recommended for sustainable soil health. Further research is needed to understand interactions between liming, microbial activity, and crop yields. Future studies should emphasize long-term monitoring, optimize lime application methods, and conduct functional microbial and crop-specific analyses to clarify ecosystem impacts and agronomic benefits.

6. ACKNOWLEDGEMENTS

The authors acknowledge the University of Gondar for financial support and thank the reviewers for their valuable contributions in improving the quality of the paper.

7. CONFLICT OF INTERESTS

There is no conflict of interest.

8. REFERENCE

Al-Shammary, A. A. G., Al-Shihmani, L. S. S., Fernández-Gálvez, J & Caballero-Calvo, A. 2024. Optimizing sustainable agriculture: A comprehensive review of agronomic practices and their impacts on soil attributes. *Journal of Environmental Management*, **364**: 121487, https://doi.org/10.1016/j.jenvman.2024.121487.

Cha, S., Kim, Y. S., Lee, A. L & Lee, D. 2021. Liming Alters the Soil Microbial Community and Extracellular Enzymatic Activities in Temperate Coniferous Forests. *Forests*, **12**: 1–16.

Chen, Q., Song, Y., An, Y., Lu, Y & Zhong, G. 2024. Soil Microorganisms: Their Role in Enhancing Crop Nutrition and Health. *Diversity*, **16(12)**: 1–26, https://doi.org/10.3390/d16120734.

Dang, L. Van, Ngoc, N. P & Hung, N. N. 2022. Effects of Biochar, Lime, and Compost Applications on Soil Physicochemical Properties and Yield of Pomelo (Citrus grandis Osbeck) in Alluvial Soil of the Mekong Delta. *Applied and Environmental Soil Science*, **2022**: 1–10, https://doi.org/10.1155/2022/5747699.

Danilova, I & Sharipova, M. 2020. The Practical Potential of Bacilli and Their Enzymes for

Industrial Production. *Frontiers in Immunology*, **11:** 1–7, https://doi.org/10.3389/fmicb.2020.01782.

Ding, Z., Ren, B., Chen, Y., Yang, Q & Zhang, M. 2023. Chemical and Biological Response of Four Soil Types to Lime Application: An Incubation Study. *Agronomy*, **13**: 1–16, https://doi.org/10.3390/agronomy13020504.

Ewunetu, T., Selassie, Y. G., Molla, E., Admase, H & Gezahegn, A. 2025. Soil properties under different land uses and slope gradients: Implications for sustainable land management in the Tach Karnuary watershed, Northwestern Ethiopia. *Frontiers in Environmental Science*, **13**: 1–13, https://doi.org/10.3389/fenvs.2025.1518068.

Fatma, K & Ahmet, Ö. 2023. Negative effects of acid rains on agricultural areas. *Annals of Environmental Science and Toxicology*, **7(1)**: 013–016, https://doi.org/10.17352/aest.000065.

Gebreyohannes, G., Moges, F., Sahile, S & Raja, N. 2013. Isolation and characterization of potential antibiotic producing actinomycetes from water and sediments of Lake Tana, Ethiopia. *Asian Pacific Journal of Tropical Biomedicine*, **3(6)**: 426–435, https://doi.org/10.1016/S2221-1691(13)60092-1.

Goupil, K., Nkongolo, K. K & Nasserulla, S. 2015. Characterization of fungal communities in limed and unlimed lands contaminated with metals: Phospholipid fatty acid (PLFA) analysis and soil respiration. *American Journal of Biochemistry and Biotechnology*, **11(2)**: 45–56, https://doi.org/10.3844/ajbbsp.2015.45.56.

Hindersah, R., Handyman, Z., Indriani, F. N., Suryatmana, P & Nurlaeny, N. 2018 Vulnerability of mountain communities to climate change and natural resources scarcity in Northwest Ethiopia: the case of Debark Woreda. *Journal of Degraded and Mining Lands Management*, **6(1)**: 1467–1482, https://doi.org/10.15243/jdmlm.2018.061.1467.

Janowski, D & Leski, T. 2022. Factors in the Distribution of Mycorrhizal and Soil Fungi. *Diversity*, **14(12)**: 1–15, https://doi.org/10.3390/d14121122.

Jiang, C & Zeng, H. 2022. Comparison of soil microbial community structure and function for karst tiankeng with different degrees of degradation. *Ecology and Evolution*, **12(12)**: 1–11, https://doi.org/10.1002/ece3.9615.

Kamrun Nahar, M., Nazmul Haque, M., Kumar Paul, G., Islam, S., Abu Saleh, M., Salah Uddin, M & Zaman, S. 2022. Bacteria isolated from cultivated soil after liming promote seed germination and seedling growth of crop plants. *Current Research in Biotechnology*, **4(2021)**: 21–31, https://doi.org/10.1016/j.crbiot.2021.12.001.

Li, S., Liu, J., Yao, Q., Yu, Z., Li, Y., Jin, J., Liu, X & Wang, G. 2022. Short-term lime

application impacts microbial community composition and potential function in an acid black soil. *Plant and Soil*, **470**: 35–50, https://doi.org/10.1007/s11104-021-04913-0.

Madegwa, Y. M & Uchida, Y. 2021. Liming improves the stability of soil microbial community structures against the application of digestate made from dairy wastes. *J. Environmental Management*, **297**: 113356, https://doi.org/10.1016/j.jenvman.2021.113356.

Mencel, J., Mocek-Płóciniak, A & Kryszak, A. 2022. Soil Microbial Community and Enzymatic Activity of Grasslands under Different Use Practices: A Review. *Agronomy*, **12(5)**: 1–16, https://doi.org/10.3390/agronomy12051136.

Narendrula-Kotha, R & Nkongolo, K. K. 2017. Microbial response to soil liming of damaged ecosystems revealed by pyrosequencingand phospholipid fatty acid analyses. *PLoS ONE*, **12(1)**: 1–22. https://doi.org/10.1371/journal.pone.0168497.

Nguyen, N. H. 2023. Fungal Hyphosphere Microbiomes Are Distinct from Surrounding Substrates and Show Consistent Association Patterns. *Microbiology Spectrum*, **11(2)**: 1–13, https://doi.org/10.1128/spectrum.04708-22.

Osuntokun, O., Thonda, O. A & Ige, O. O. 2018. Use of the Biomerieux ID 32C Fungal Identification System for Fungal Isolated from Mother & Child Hospital Dump Site in Akure, Ondo State Nigeria. *Journal of Bacteriology & Mycology*, **6(1)**: 12–17, https://doi.org/10.15406/jbmoa.2018.06.00168.

Peng, Z., Liang, C., Gao, M., Qiu, Y., Pan, Y., Gao, H., Liu, Y., Li, X., Wei, G & Jiao, S. 2022. The neglected role of micronutrients in predicting soil microbial structure. *Npj Biofilms and Microbiomes*, **8(1)**: 1–10. https://doi.org/10.1038/s41522-022-00363-3.

Swer, H., Dkhar, M. S & Kayang, H. 2011. Fungal Population and Diversity in Organically Amended Agricultural Soils of Meghalaya, India. *Journal of Organic System*, **6(2)**: 1–10.

Symanowicz, B & Toczko, R. 2022. Enzymatic Activity of Soil after Applying Mineral Fertilizers and Waste Lignite to Maize Grown for Silage. *Agriculture*, **12**: 1–14, https://doi.org/10.3390/agriculture12122146.

Tassadaq, H., Aneela, R., Shehzad, M., Iftikhar, A., Jafar, K., Veronique, E. H., Kil, Y. K & Muhammad, A. 2013. Biochemical characterization and identification of bacterial strains isolated from drinking water sources of Kohat, Pakistan. *African Journal of Microbiology Research*, **7(16)**: 1579–1590, https://doi.org/10.5897/ajmr12.2204.

Wang, S., Szeles, B., Krammer, C., Schmaltz, E., Song, K., Li, Y., Zhang, Z., Blöschl, G & Strauss, P. 2022. Agricultural intensification vs. climate change: What drives long-term changes in sediment load? *Hydrology and Earth System Sciences*, **26(12)**: 3021–3036,

https://doi.org/10.5194/hess-26-3021-2022.

Wei, H., Liu, Y., Xiang, H., Zhang, J., Li, S & Yang, J. 2020. Soil ph responses to simulated acid rain leaching in three agricultural soils. *Sustainability (Switzerland)*, **12(1)**: 1–12, https://doi.org/10.3390/su12010280.

Yin, C., Schlatter, D. C., Kroese, D. R., Paulitz, T. C & Hagerty, C. H. 2021a. Impacts of lime application on soil bacterial microbiome in dryland wheat soil in the Pacific Northwest. *Applied Soil Ecology*, **168(2021)**: 104113, https://doi.org/10.1016/j.apsoil.2021.104113.

Yin, C., Schlatter, D. C., Kroese, D. R., Paulitz, T. C & Hagerty, C. H. 2021b. Responses of Soil Fungal Communities to Lime Application in Wheat Fields in the Pacific Northwest. *Frontiers in Microbiology*, **12**: 1–13, https://doi.org/10.3389/fmicb.2021.576763.

Yusron, M. H, M & Susilawati, P. N. 2022. Application of local microorganism solution and lime to improve soil fertility and maize yield Application of local microorganism solution and lime to improve soil fertility and maize yield. *2nd International Conference on Environmental Ecology of Food Security*, 1–8, https://doi.org/10.1088/1755-1315/1107/1/012003.

Zhang, N., Ma, Z., Li, D., Ni, H., Sun, B & Liang, Y. 2022. Soil pH Filters the Association Patterns of Aluminum-Tolerant Microorganisms in Rice Paddies. *MSystems*, **7(1)**: 1–15, https://doi.org/10.1128/msystems.01022-21.

Zheng, Q., Hu, Y., Zhang, S., Noll, L., Böckle, T., Dietrich, M., Herbold, C. W., Eichorst, S. A., Woebken, D., Richter, A & Wanek, W. 2019. Soil multifunctionality is affected by the soil environment and by microbial community composition and diversity. *Soil Biology and Biochemistry*, **136**: 107521, https://doi.org/10.1016/j.soilbio.2019.107521.