

Synergistic Effects of Biochar, Urea Fertilizer, and Lactic Acid Bacteria on Mitigating Soil Ammonia Volatilization

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ABSTRACT

Soil ammonia (NH₃) volatilization from urea fertilizer poses a significant challenge to nitrogen management in agriculture, resulting in considerable nitrogen losses and environmental pollution. This study investigates the combined effects of biochar, urea fertilizer, and lactic acid bacteria (LAB) on soil NH₃ volatilization and vegetable growth. Biochar improves soil structure and nutrient retention, while LAB enhances nutrient cycling and microbial activity. A series of treatments combining biochar, urea, and LAB were applied to soil, with NH₃ emissions measured using a closed dynamic air flow system during an incubation period. A field experiment with five fertilization treatments, including control, was carried out in an open field located in Perlis, Malaysia. Treatments with biochar and urea (T4), and the combination of biochar, urea, and LAB (T5), significantly reduced ammonia losses while also improving plant fresh weight and SPAD chlorophyll readings. These results suggest enhanced nutrient uptake and chlorophyll production. Further research is recommended to assess long-term effects, economic viability, and broader applicability across different crops and soil types.

Keywords: Nitrogen, Chlorophyll, Plant growth, Preservation, Shelf life.

1. INTRODUCTION

In agricultural systems, soil ammonia (NH₃) volatilization plays a crucial role in determining the efficiency of nitrogen (N) fertilizers and the sustainability of environmental practices. This process involves the transformation of ammonium ions into gaseous NH₃, leading to significant N losses and contributing to air and water pollution [1]. Understanding the key factors influencing NH₃ volatilization, such as soil pH, temperature, and microbial activity, is essential for developing effective mitigation strategies [2]. Reducing NH₃ volatilization not only improves nitrogen use efficiency but also minimizes its environmental impact, underscoring the need for in-depth research in this area.

Urea fertilizer, one of the most widely used N sources in agriculture, significantly contributes to crop yield improvement. However, its effectiveness is often limited by nitrogen losses through NH₃ volatilization, especially in alkaline soils. These losses not only reduce fertilizer efficiency but also contribute to environmental pollution. A comprehensive understanding of urea hydrolysis and volatilization mechanisms is essential for enhancing nitrogen use efficiency and reducing ecological impacts [2]. Alternatively, the Biochar has gained attention for its potential to improve crop productivity and mitigate greenhouse gas emissions [3]. Its porous structure and large surface area provide a habitat for beneficial microbes, promoting nutrient cycling and retention [4]. Biochar has also been shown to reduce nutrient leaching, offering sustainable

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solutions to soil degradation and environmental concerns [5]. Lactic acid bacteria (LAB) also play a significant role in sustainable agriculture. LAB has been associated with improved nutrient availability through N fixation, phosphorus solubilization, and the production of plant growth-promoting compounds [6]. Furthermore, LAB enhances soil microbial diversity, suppresses soil-borne pathogens, and promotes a healthy rhizosphere, all of which contribute to better plant growth and soil fertility [7].

Despite the known individual benefits of biochar, urea, and LAB, their combined effects on NH_3 volatilization are not well understood. Urea increases the risk of ammonia losses, while biochar and LAB may counterbalance this by improving soil structure and microbial activity. Exploring the synergistic interactions among these amendments could provide new insights into optimizing nutrient use and minimizing environmental losses. Therefore, this study aims to determine the combined effects of biochar, urea fertilizer, and lactic acid bacteria on soil NH_3 volatilization and vegetable growth performance. We hypothesize that the integration of these three components will significantly reduce NH_3 losses, enhance N efficiency, and improve crop growth under open-field conditions.

2. MATERIAL AND METHODS

2.1 Biochar, urea, and lactic acid bacteria (LAB) solution collection and preparation

The biochar used in the study was the charred rice husks sourced from Kilang Beras Melayu Utara (M) Sdn. Bhd, Perlis, Malaysia. Urea fertilizer was obtained from Lean Vin Enterprise, while LAB solution was prepared by mixing the rice water and fresh milk in a 1:10 ratio [8]. Rice water was prepared by soaking one cup of rinsed rice in 10 cups of water for 30 minutes, then straining the milky water into a jar covered with a breathable cloth and fermenting it at room temperature (20–30°C) for 3–5 days until a slightly sour odor developed. The fermented rice water was then mixed with fresh milk, which provided lactose and proteins, and allowed to ferment for another 5 days under similar conditions. Brown sugar was added after the fermentation period to enhance LAB longevity. The LAB-rich solution, indicated by a sour aroma and tangy taste, was strained and stored in a cool place for use in improving soil health, promoting plant growth, and suppressing soil-borne pathogens.

2.2 Soil ammonia volatilization

The daily NH_3 loss was determined using a closed dynamic air-flow system. The system comprised a 500 mL conical flask serving as the exchange chamber and a 250 mL conical flask containing 75 mL of boric acid with indicator solution to trap NH_3 gas. Approximately 250 g of soil was placed in the exchange chamber and moistened with distilled water to 55% field capacity. An air pump was connected to the chamber inlet, while the outlet was linked via silicon tubing to the boric acid solution. Air was circulated through the system at a constant rate of 4 L/min, regulated using a Gilmont flow meter. The NH_3 released was absorbed in the trapping solution, which contained 75 mL of boric acid with bromocresol green and methyl red indicators, and quantified by titration with 0.1 M HCl. The incubation was conducted at room temperature, with the boric acid solution replaced every 24 hours over 10 days or until NH_3 loss dropped below 1% of the nitrogen applied. The experiment followed a completely randomized design with three replicates per treatment.

The treatments used to compare the percent of NH_3 volatilization loss in the clayey soil were as follows:

T1: Soil only

T2: 12 g Biochar only

T3: 2 g Urea fertilizer only

T4: 12 g Biochar + 2 g urea fertilizer

T5: 12 g Biochar + 2 g urea fertilizer + 6 mL LAB solution

2.3 Field experiment

A field study was conducted in an open plot located at INSAT UniMAP, Perlis, Malaysia. There were 5 fertilization treatments, including a control. The experiment was conducted using a completely randomized design with 5 replications. The test crop used in this present study was F1 hybrid amaranth green. The crops were irrigated with tap water. The amounts of urea, biochar and LAB used was based on the standard recommendation of fertilizer for the test crop.

The fertilizer applications were as follows:

T1: soil only (control)

T2: 30 g of biochar only

T3: 5 g of urea only

T4: 30 g biochar + 5g urea

T5: 30 g biochar + 5 g urea + 15 mL LAB solution

The fertilizers were surface applied at 12 and 23 days after planting (DAP). Before harvesting, the chlorophyll contents of the amaranth plants were measured using a SPAD chlorophyll meter (SPAD-502, Konica Minolta, Osaka, Japan). The crops were harvested after 32 DAP. Thereafter, they were cleaned and oven-dried at 60 °C until constant weight was attained. Afterwards, the dry weight of the amaranth green was determined using a digital weighing machine (UWA-C015, Accutec, Taipei, Taiwan).

2.4 Statistical analysis

Analysis of variance was used to test for treatment effects, and the means of treatment were compared using Tukey's test (SAS version 9.2).

3 RESULTS AND DISCUSSION

3.1 Soil ammonia volatilization

Figure 1 presents the daily NH_3 volatilization loss from soil incubation with different treatments. Urea alone exhibited the highest NH_3 loss, with a sharp peak of approximately 45% on Day 4, followed by a rapid decline over the subsequent days. This trend reflects the rapid hydrolysis of urea in the soil, which interferes with plant growth and promotes the release of NH_3 gas into the atmosphere. The large amount of N loss shows the inefficiency of urea as a fertilizer when applied in large quantities because most of its N content is lost as it volatilizes into the air instead of being absorbed by the soil and plants. This finding is consistent with previous research that reported urea to be highly prone to NH_3 volatilization, particularly under conditions of high temperature and soil alkalinity [8][9]. Consequently, this emphasizes the need for strategies to mitigate N losses and improve the efficiency of urea-based fertilizers.

On the other hand, the use of biochar and combined with urea and lactic acid bacteria (LAB) can significantly reduce NH_3 volatilization. Biochar alone showed consistently low soil NH_3 loss. It also shows its ability to absorb ammonia and increase N retention in the soil [10]. When biochar is combined with urea, NH_3 loss is further reduced compared to urea alone, with a peak of less than 15%. This shows the ability of biochar to slow the hydrolysis of urea. The most effective treatment was the combination of biochar, urea, and LAB, which showed the lowest NH_3 loss throughout the experimental period, with a peak of less than 5%. LAB likely contributes to this reduction by increasing microbial nitrogen immobilization and stabilizing NH_3 in the soil. This

finding is in line with the study by Park et al. (2023) [11] and Chen et al. (2022) [12], who highlighted the synergistic effect of biochar and microbial inoculants in minimizing NH_3 emissions. This demonstrates that integrating biochar and LAB with urea fertilizers can effectively mitigate N losses, improve N use efficiency, and support sustainable agricultural practices.

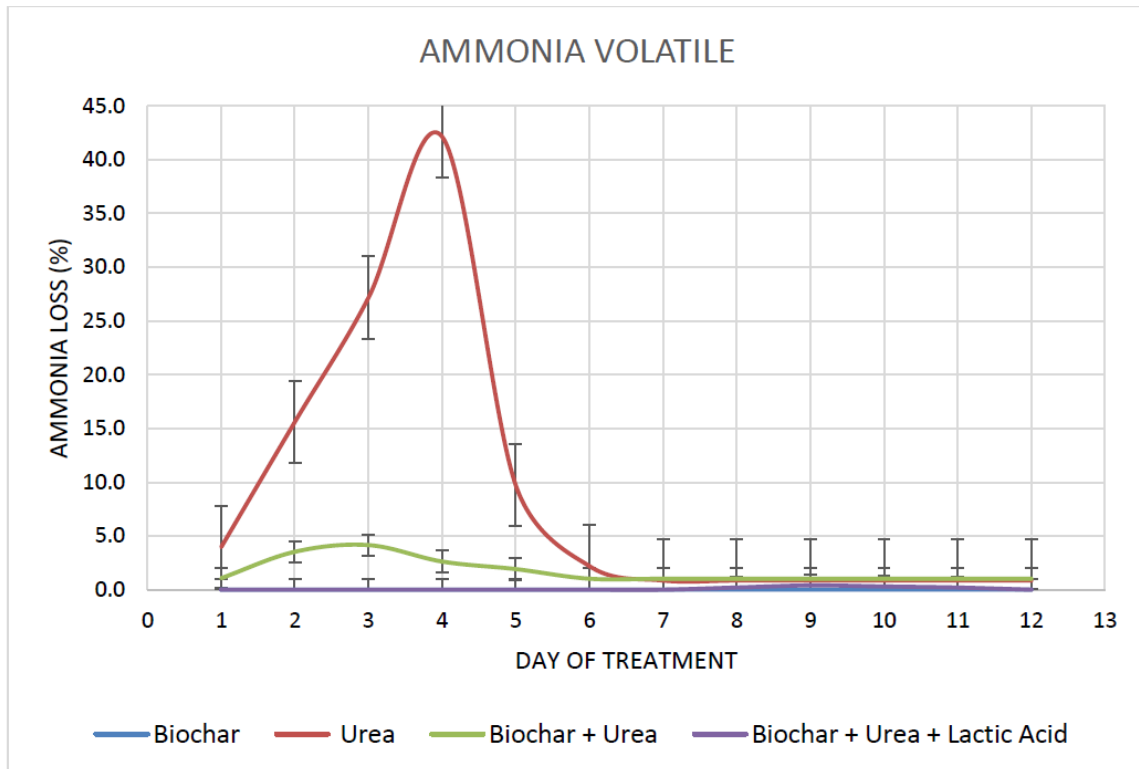


Figure 1: Daily ammonia volatilization loss from soil incubation with different treatments.

3.2 SPAD value

The SPAD value is commonly used as an indirect indicator of a plant's N status. Table 1 presents the SPAD chlorophyll meter readings of amaranth plants taken just before harvest, at 32 days after planting (DAP). Treatment T5 recorded the highest SPAD value of 30.60, followed by T4 with a value of 29.63. This suggests that the combination of biochar, urea, and LAB solution enhanced N retention, thereby increasing chlorophyll content in the plants. Treatments T1, T2, and T3 exhibited lower SPAD readings (Table 1). The absence of additional fertilizer in T1 and T2 likely contributed to the reduced chlorophyll content, as reflected by the lower SPAD values. Similarly, the sole application of urea in T3 resulted in comparatively low SPAD readings, suggesting that nitrogen alone may not be sufficient to support optimal chlorophyll development.

Table 1: SPAD values of amaranth plants measured at 32 days after planting DAP, just before harvest.

Treatment	SPAD Chlorophyll Meter Reading
T1	21.80 ^b ± 0.91
T2	22.83 ^b ± 0.49
T3	24.00 ^b ± 0.35
T4	29.63 ^a ± 1.71
T5	30.60 ^a ± 0.50

T1 – soil only (control); T2 – 30 g of biochar only; T3 – 5 g of urea only; T4 - 30 g biochar + 5g urea; T5 - 30 g biochar + 5 g urea + 15 mL LAB solution. All values are calculated as mean ± standard error with n=5. Different letters indicate significant differences among treatments, as determined by ANOVA at P < 0.05.

3.3 Plant yield

Table 2 presents the fresh weight of amaranth plants at 32 days after planting. Our findings revealed that treatment T4 and T5 recorded the highest fresh weight of amaranth plants. This is consistent with Bolan et al. (2023) [13] reported that incorporating biochar with microbial inoculants and fertilizer can enhance the crop productivity. The absence of fertilizer in T1 and T2 resulted in the lowest fresh weights. In contrast, T3, which received only urea, showed higher plant weight than T1 and T2 but was still lower compared to treatments with the co-application of urea, biochar, and LAB. This indicates that combining biochar with urea generally promotes more beneficial plant growth than using urea alone. A similar finding was reported by Shi et al. (2020) [14], who observed that blending biochar with urea enhanced biomass production by improving nitrogen retention through surface adsorption.

Table 2: Fresh weight of amaranth plants at 32 days after planting.

Treatment	Plant fresh weight
T1	5.5 ^c ± 0.55
T2	7.6 ^c ± 0.26
T3	12.97 ^b ± 0.80
T4	24.83 ^a ± 1.30
T5	31.63 ^a ± 3.90

T1 – soil only (control); T2 – 30 g of biochar only; T3 – 5 g of urea only; T4 - 30 g biochar + 5g urea; T5 - 30 g biochar + 5 g urea + 15 mL LAB solution. All values are calculated as mean ± standard error with n=5. Different letters indicate significant differences among treatments, as determined by ANOVA at P < 0.05.

4. CONCLUSION

In conclusion, this study demonstrates that the combined application of biochar, urea, and LAB solution resulted in the lowest ammonia volatilization rates, the highest SPAD values, and the greatest fresh weight. These findings offer a promising foundation for developing effective soil management strategies that enhance crop productivity while mitigating environmental impacts associated with nitrogen losses. Future research should investigate the long-term effects of these amendments on soil health, microbial communities, and crop yields across diverse agroecosystems to confirm their broader applicability in sustainable agriculture.

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