



Synergistic Effects of Urea and Slurry-Derived Alkaline Hydrolysis Solution on Soil Fertility, Growth, and Yield of Wheat

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Abstract

The integration of organic and inorganic nutrient sources is a cornerstone of sustainable agriculture. This study investigated the synergistic effects of urea and a slurry-derived alkaline hydrolysis solution (AHS) on soil nutrient availability and the agronomic performance of wheat. A pot experiment employing twelve treatments evaluated various combinations of urea (100%, 75%, 50%) and AHS. Results demonstrated that integrated treatments significantly enhanced soil nitrogen, phosphorus, and potassium levels compared to single-source applications. The combination of 100% urea with AHS (T10) proved optimal, recording the highest values for key growth parameters: plant height (cm), spike height (cm), number of spikes per pot, and shoot and root biomass (fresh and dry weight). Morphological analysis revealed that T10 also promoted the most extensive root system and shoot architecture. Treatments with reduced urea (75% and 50%) combined with AHS performed commendably, often surpassing full-dose urea alone, indicating improved nutrient use efficiency. The results are attributed to multiple mechanisms: the AHS likely enhances nitrogen retention and provides a slow-release nutrient source, improves soil physico-chemical properties for better root development, and supplies a balanced spectrum of macro- and micronutrients. We conclude that the integration of urea with slurry-derived alkaline hydrolysis solution is a highly effective strategy for optimizing wheat productivity and soil health, offering a viable pathway to reduce synthetic fertilizer dependency while promoting agricultural waste valorization within a circular economy framework.

1. Introduction

The global livestock sector is increasingly vulnerable to climate change, facing heightened risks from heatwaves, disease outbreaks, and extreme weather events. These challenges lead to significant animal mortality, disrupting the supply of essential products and threatening the economic stability and food security of rural communities in low- and middle-income countries (Thornton *et al.*, 2021). The disposal of these carcasses presents a critical biosecurity and environmental dilemma (Pandey *et al.*, 2020). Traditional methods, including landfilling, incineration, and rendering, are fraught with limitations, such as greenhouse gas

emissions, toxic air pollutants, groundwater contamination, and high energy demands (Barati *et al.*, 2020). Consequently, there is an urgent need for innovative disposal strategies that are both biosecure and environmentally sustainable.

Alkaline hydrolysis (AH) has emerged as a promising alternative for managing animal mortalities (Arias *et al.*, 2018). This process, which utilizes a strong alkali at elevated temperature and pressure within a sealed reactor, effectively hydrolyzes soft tissues into a sterile, nutrient-rich aqueous solution and a small amount of inert solid residue. A paramount advantage of AH is its proven efficacy in destroying a broad spectrum of pathogens, including

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recalcitrant prions responsible for diseases like bovine spongiform encephalopathy, thereby ensuring a high level of biosecurity (Iloba *et al.*, 2023). Furthermore, as a closed-system technology, AH minimizes the release of harmful emissions and odors, offering a cleaner operational profile compared to incineration or open composting.

Beyond safe disposal, AH aligns with the principles of a circular bioeconomy by transforming a hazardous waste stream into a potential resource. The primary liquid byproduct, alkaline hydrolysate (AH), is rich in nitrogen, phosphorus, potassium, and organic matter (Kang *et al.*, 2019). Several studies have highlighted its potential as a biofertilizer capable of improving soil fertility, stimulating microbial activity, and enhancing plant growth (Reddy *et al.*, 2016; Kiruba *et al.*, 2022; Cataldo *et al.*, 2021). This nutrient recovery pathway offers a sustainable alternative to synthetic fertilizers, whose production is energy-intensive and whose application is a primary contributor to nutrient leaching, eutrophication, and greenhouse gas emissions (Chojnacka, 2023).

Despite this promising potential, significant knowledge gaps hinder the agronomic optimization of AH (Zhou *et al.*, 2018). While its nutrient profile is beneficial, the specific impacts of AH on key physiological processes in staple crops like wheat—such as chlorophyll synthesis, photosynthetic efficiency, and hormonal regulation—remain poorly understood (Liu *et al.*, 2022). Moreover, the practical application of AH raises concerns regarding its high pH and salt content, which, if mismanaged, could induce soil alkalization and salinization, adversely affecting soil structure and nutrient bioavailability (Wang *et al.*, 2024). A comprehensive understanding of both the short-term agronomic benefits and the long-term implications for soil carbon dynamics and microbial community structure is essential for developing safe and effective application protocols.

Therefore, this study is designed to bridge this critical gap by systematically evaluating the use of alkaline hydrolysate in wheat cultivation. The overarching goal is to assess its efficacy as a sustainable soil amendment that not only supports crop productivity but also contributes to improved soil health. The specific objectives of this research are: To produce a nutrient-rich hydrolysate through the alkaline hydrolysis of animal by-products using potassium hydroxide under controlled conditions ($\text{pH} > 14$); To evaluate the effects of the resulting alkaline hydrolysate on wheat physiology, with a specific focus on growth parameters, chlorophyll content, photosynthetic activity, and ultimate yield. To assess the impact of hydrolysate application on soil chemical properties, including pH, electrical conductivity, and nutrient availability, to determine any risks of salinization or alkalization. To investigate the influence of the hydrolysate on key soil health indicators, particularly soil organic carbon and microbial activity, to gauge its potential for enhancing long-term soil fertility and carbon sequestration.

2 Materials and Methods

2.1. Experimental Overview

This study was conducted to evaluate the efficacy of alkaline hydrolysis (AH) for the biosecure disposal of animal by-products and to assess the agronomic potential of the resulting hydrolysate as an organic fertilizer. The experimental work comprised two main phases: (1) the production and characterization of the alkaline hydrolysate, and (2) a pot experiment to determine its effects on wheat growth, physiology, and nutrient uptake.

2.2. Collection and Preparation of Animal By-products

Animal by-products (ABPs), including ears, hooves, and horns, were sourced from a local slaughterhouse in Faisalabad. Samples were collected at four-day intervals, with a cumulative mass of approximately 5 kg. All collection procedures followed standard microbiological safety protocols to ensure biosecurity.

2.3. Alkaline Hydrolysis Process and Hydrolysate Production

The AH process was adapted from established protocols (Shafer *et al.*, 2000; Arias *et al.*, 2018). A 4 M potassium hydroxide (KOH) solution was prepared by dissolving commercial-grade KOH in distilled water, resulting in a solution with a $\text{pH} > 14$. The solution was stirred thoroughly and allowed to equilibrate for 12 hours prior to use.

The collected ABPs were placed in a 30 L polyethylene container. The 4 M KOH solution was added at a ratio of 2:1 (solution to ABPs, v/w), ensuring the material was completely submerged. The container was sealed and maintained at ambient temperature (approximately 25–30°C). The progression of hydrolysis was monitored visually and analytically over a 20-day period, with observations recorded on days 5, 10, 15, and 20.

2.4. Neutralization and Post-Treatment of Hydrolysate

Following the 20-day hydrolysis period, the resulting hydrolysate possessed a highly alkaline pH. To render it suitable for agricultural use, a two-step neutralization process was employed. First, the hydrolysate was blended with an acidic soil conditioner ($\text{pH} 2.5$) at a 1:4 ratio (hydrolysate to conditioner). This preliminary adjustment was aimed at achieving a pH below 8. Subsequently, the mixture underwent anaerobic digestion under standard conditions to further stabilize the pH and reduce its salinity. The final treated hydrolysate was analyzed for pH, electrical conductivity (EC), and available nitrogen (N), phosphorus (P), and potassium (K) content.

2.5. Soil Collection and Pre-Sowing Analysis

The soil for the pot experiment was collected from the top 15 cm of an experimental field at the University of Agriculture, Faisalabad. The soil was air-dried, homogenized, and passed through a 2 mm sieve. A sub-sample was taken for pre-

sowing analysis. Soil pH was measured in a saturated paste using a calibrated digital pH meter, and electrical conductivity (EC) was determined in the saturated paste extract using a digital EC meter. The initial soil characteristics are presented in Table 1.

2.6. Pot Experiment Design

A pot experiment was conducted in a wire-house at the Institute of Soil and Environmental Sciences (ISES), University of Agriculture, Faisalabad. Pots with a capacity of 10 kg were each filled with 8.5 kg of the prepared soil. The wheat variety 'Dilkash 2020' was used as the test crop.

The experiment was arranged in a Completely Randomized Design (CRD) with twelve treatments, each replicated three times. The treatments were designed to evaluate the effect of the hydrolysate both with and without mineral nitrogen fertilizer, as follows:

- **T1:** Control (No fertilizer)
- **T2:** 100% Recommended N
- **T3:** 75% Recommended N
- **T4:** 50% Recommended N
- **T5:** Control + Slurry Alkaline Hydrolysis
- **T6:** 100% N + Slurry Alkaline Hydrolysis
- **T7:** 75% N + Slurry Alkaline Hydrolysis
- **T8:** 50% N + Slurry Alkaline Hydrolysis
- **T9:** Control + Alkaline Hydrolysis
- **T10:** 100% N + Alkaline Hydrolysis
- **T11:** 75% N + Alkaline Hydrolysis
- **T12:** 50% N + Alkaline Hydrolysis

Wheat seeds were sown in each pot, and tap water was used for irrigation throughout the growth period.

2.7. Data Collection

2.7.1. Agronomic Parameters

At harvest, the following growth parameters were recorded from plants in each pot. Shoot and root lengths were measured using a meter rod. For biomass determination, shoots and roots were separated, and their fresh weights were immediately recorded using a digital balance. The samples were then air-dried for two days followed by oven-drying at 60°C until a constant weight was achieved to determine shoot and root dry weights.

2.7.2. Physiological Parameters

Physiological measurements were taken on fully expanded leaves at the flowering stage. The net photosynthetic rate (A),

stomatal conductance (gs), and transpiration rate (E) were measured using an Infra-Red Gas Analyzer (IRGA). Chlorophyll content was assessed as SPAD values using a Minolta SPAD-502 meter on the third uppermost leaf, with three readings averaged per replicate.

Leaf relative water content (RWC) was determined using the method of Mayak et al. (2004). Fresh leaf discs were weighed to obtain fresh weight (FW), then soaked in distilled water for 24 hours to obtain turgid weight (TW), and finally oven-dried to determine dry weight (DW). RWC was calculated as: $RWC (\%) = [(FW - DW) / (TW - DW)] \times 100$.

2.7.3. Plant Nutrient Analysis

Oven-dried plant samples were ground to a fine powder for nutrient analysis. Plant tissue digestion was performed using the Wolf (1982) method. Briefly, 0.5 g of ground plant material was digested with 5 mL of concentrated H₂SO₄, followed by successive additions of 35% H₂O₂ at 350°C in a digestion block until a clear digest was obtained. The digest was cooled, filtered, and diluted to 50 mL with distilled water.

- **Nitrogen (N)** concentration was determined from the digest using the micro-Kjeldahl distillation and titration method.
- **Phosphorus (P)** concentration was measured spectrophotometrically at 410 nm using the ammonium molybdate-ascorbic acid method (Watanabe & Olsen, 1965).
- **Potassium (K)** concentration was determined using a flame photometer, calibrated with appropriate K standards (Nasim *et al.*, 2008).

2.8. Statistical Analysis

All data collected on agronomic, physiological, and nutritional parameters were subjected to analysis of variance (ANOVA) appropriate for a Completely Randomized Design. Treatment means will be compared using a suitable post-hoc test, such as Fisher's Least Significant Difference (LSD), at a 5% probability level to determine significant differences. Statistical analysis will be performed using standard statistical software (e.g., R or SAS).

3 Results

3.1 Soil Chemical attributes

The application of urea and a slurry-derived alkaline hydrolysis solution, both individually and in combination, significantly influenced the soil's nitrogen (N), phosphorus (P), and potassium (K) content. The results for each nutrient are detailed below.

Soil nitrogen levels were strongly affected by the treatment applications (Figure 1a). The untreated control (T1) showed the lowest N content, indicating a baseline depletion. Applications of urea alone (T2: 100%, T3: 75%, T4: 50%)

resulted in a dose-dependent increase in soil N, with T2 yielding the highest value among urea-only treatments. The alkaline hydrolysis solution alone (T5, T9) provided a moderate increase in N compared to the control, but its effect was substantially enhanced when combined with urea.

is the primary P source, the synergy with full-rate urea application best facilitates phosphorus retention and availability.

Similar to phosphorus, soil potassium was primarily enriched by the alkaline hydrolysis solution (Figure 1c). The untreated

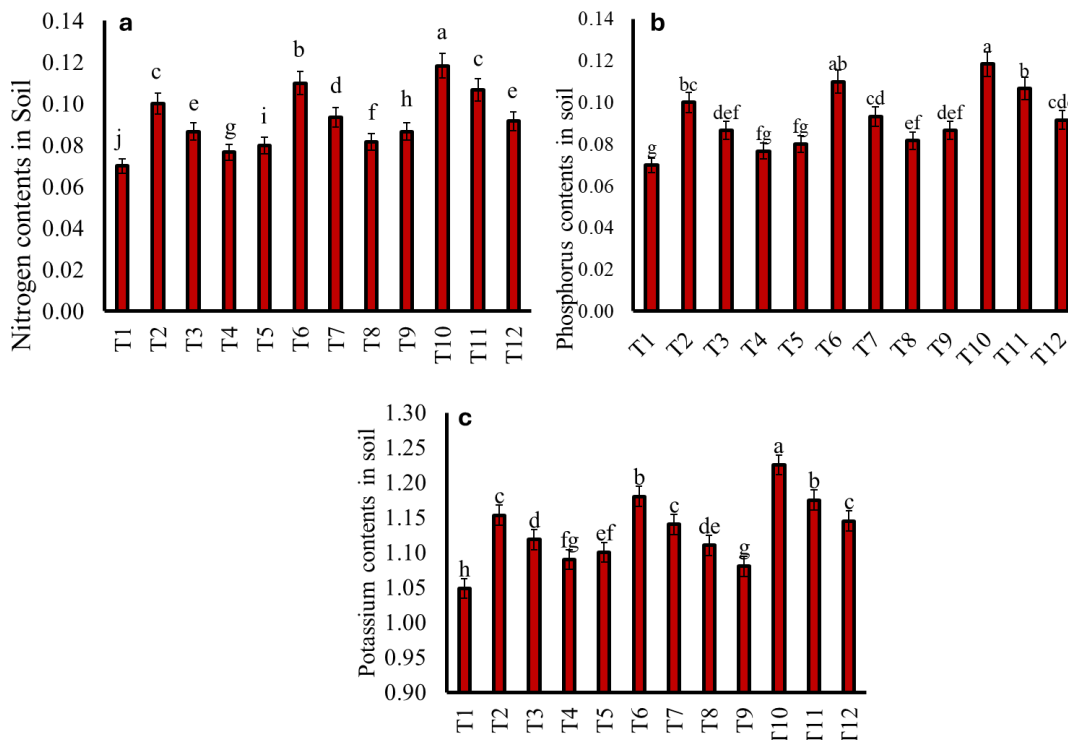


Figure 1: Soil chemical attributes under the influence of different amendments. Error bars showed the standard deviation and different letter showed level of significance ($P < 0.05$).

Integrated treatments (T6, T7, T8) consistently resulted in higher soil N than their single-source counterparts. The maximum soil nitrogen content was recorded in T10 (100% urea + alkaline hydrolysis solution), which was significantly higher than all other treatments. While combinations with reduced urea (T11, T12: 50%) still improved N levels over the controls, their values were lower than T10, confirming that a full-rate integrated application is optimal for soil nitrogen availability in wheat cultivation.

The phosphorus content in the soil demonstrated the critical role of the organic amendment (Figure 1b). The control (T1) had the lowest P level. Urea applications alone (T2-T4) had a minimal impact on soil P, consistent with its composition. In contrast, the alkaline hydrolysis solution alone (T5) notably increased P availability, highlighting its value as a phosphorus source.

This effect was amplified in the integrated treatments. Combinations of urea and the alkaline solution (T6-T8) led to significant enhancements in soil P, with the highest value observed in T10 (100% urea + alkaline hydrolysis solution). Treatments with reduced urea (T11, T12) showed improved P levels compared to single-source applications but were inferior to T10. This suggests that while the organic solution

control (T1) showed minimal potassium, while urea-only treatments (T2-T4) provided only a slight, dose-dependent increase.

The alkaline hydrolysis solution alone (T5, T9) substantially improved K levels, confirming its potassium content. The most significant improvements were observed in the combined treatments (T6-T8), with T10 (100% urea + alkaline hydrolysis solution) achieving the peak soil potassium content. This indicates a synergistic effect where the organic input supplied the potassium, and the integrated nutrient management improved its retention. Treatments T11 and T12 (reduced urea + solution) were effective but less so than T10, reinforcing that the full combination is the most effective strategy for enhancing soil potassium.

3.2 Agronomic Performance of Wheat Under Different Nutrient Management Treatments

The application of urea and slurry-derived alkaline hydrolysis significantly influenced key agronomic parameters of wheat (Figure 2 a-e). The results for plant height, spike height, number of spikes per pot, fresh weight, and dry weight are presented below.

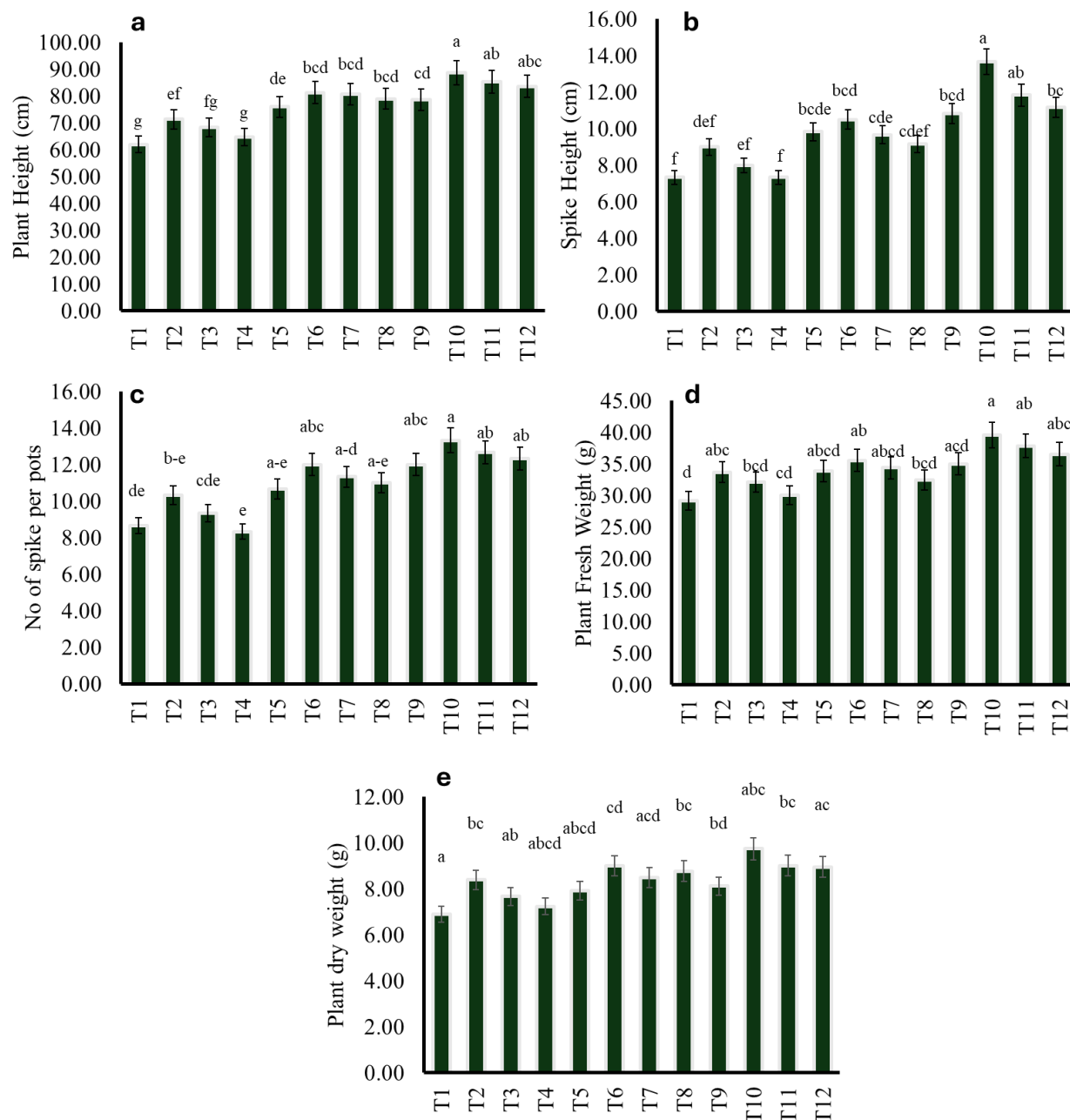


Figure 2: Agronomic Performance of Wheat Under Different Nutrient Management Treatments a-e explaining different parameters and its responses. Error bars indicated the standard deviation (n=5) and different letters showed different level of significance.

Plant height was strongly influenced by nutrient treatments. The untreated control (T1) resulted in the shortest plants, while applications of urea alone (T2-T4) produced only a moderate, dose-dependent increase in height. The alkaline hydrolysis solution by itself (T5) also showed a limited effect. In contrast, integrated treatments combining urea and the alkaline solution (T6-T8) led to a significant enhancement in plant stature, indicating a synergistic relationship. The maximum plant height was achieved with T10 (100% urea + alkaline hydrolysis solution). Treatments with reduced urea

levels (T11, T12) also promoted substantial growth but were inferior to T10, demonstrating that the full combination of nutrients is optimal for maximizing plant height. The treatments had a pronounced effect on the reproductive architecture, as measured by spike height. The control (T1) exhibited the poorest development. Urea-only treatments (T2-T4) showed a clear, linear reduction in spike height as the nitrogen dose decreased. A notable improvement was observed when urea was combined with the alkaline hydrolysis solution (T6-T8), with these combinations

consistently outperforming their single-component counterparts. The tallest spikes were recorded in T10, confirming the superior efficacy of the full integrated application. While T11 and T12 also benefited from the combination, their performance was lower than T10, underscoring the advantage of using both inputs at full strength.

Spike count, a key indicator of reproductive yield potential, was significantly affected by the nutrient management strategy. The control (T1) produced the fewest spikes. Urea application alone (T2-T4) increased spike numbers in a dose-dependent manner but failed to reach optimal levels. The most substantial increases were observed in the combined treatments (T6-T8), which significantly outperformed both the urea-only and organic-only (T5, T9) treatments. The highest number of spikes per pot was unequivocally recorded in T10. Treatments T11 and T12 showed commendable results but did not match the output of T10, reinforcing that the full integrated treatment is most effective for promoting tillering and spike initiation.

The fresh biomass of wheat plants responded markedly to the nutrient sources. The lowest fresh weight was found in the control (T1). Urea-only treatments (T2-T4) showed a positive correlation between nitrogen dose and biomass, though the gains were limited. A significant boost in fresh weight was observed in the integrated treatments (T6-T8), highlighting a synergistic effect that enhanced overall plant growth and water-rich biomass accumulation. Treatment T10 yielded the highest fresh weight, establishing it as the most effective strategy. The reduced urea combinations (T11, T12) also improved biomass over the controls but were less effective than the full combination.

The dry weight data, reflecting structural biomass accumulation, further confirmed the superiority of integrated nutrient management. The control (T1) registered the lowest dry matter. Urea application alone (T2-T4) resulted in a progressive decrease in dry weight with reducing nitrogen levels. The combination of urea and the alkaline hydrolysis solution (T6-T8) led to a significant increase in dry matter production compared to all single-source applications. Treatment T10 achieved the maximum dry weight, solidifying its status as the optimal treatment for biomass accumulation. The results from T11 and T12, though positive, were suboptimal compared to T10, demonstrating that a full-rate integration of organic and inorganic nutrients is crucial for maximizing dry matter production in wheat.

3.3 Biomass Partitioning and Morphological Development of Wheat as Influenced by Integrated Nutrient Management

The application of urea and a slurry-derived alkaline hydrolysis solution significantly modulated both aboveground and belowground growth parameters in wheat. The synergistic effects of combining inorganic and organic nutrient sources were evident across all measured traits of biomass accumulation and morphological development

(Figure 3 a-f). The analysis of shoot biomass revealed critical insights into nutrient utilization and plant productivity. The untreated control (T1) established a baseline, yielding the lowest shoot fresh (SFW) and dry (SDW) weights, indicative of severe nutrient limitation.

Urea alone (T2-T4) resulted in a positive but suboptimal response. T2 (100% urea) produced the highest SFW and SDW among urea-only treatments, with T3 (75%) and T4 (50%) showing a progressive decline, confirming a direct correlation between nitrogen availability and biomass production. However, even the full urea dose failed to maximize growth potential. Organic amendment alone (T5, T9) demonstrated a limited capacity to enhance shoot biomass, with performances similar to the lower-tier urea treatments, suggesting that the alkaline hydrolysis solution provides beneficial compounds but insufficient readily available nitrogen. The synergistic effect of combined applications (T6-T8) was pronounced. T6 (100% urea + solution) showed a substantial increase, nearly matching the performance of the top treatment. This indicates that the organic amendment improves the efficiency of urea nitrogen, potentially through improved soil microbiology or nutrient retention. The optimal treatment, T10 (100% urea + alkaline hydrolysis solution), achieved the maximum SFW and SDW, significantly outperforming all other treatments. This underscores a powerful synergy where the organic component likely enhanced nitrogen use efficiency and provided secondary benefits, leading to maximal carbon assimilation and biomass accumulation. Reduced urea combinations (T11, T12) performed commendably, surpassing all single-source applications, but their values were consistently lower than T10. This confirms that while integration is beneficial at reduced nitrogen levels, the full agronomic potential is only realized with a complete, integrated nutrient package.

Root development mirrored the aboveground response, highlighting the systemic nature of nutrient-mediated growth enhancement. The control (T1) exhibited the least root fresh (RFW) and dry (RDW) weight, reflecting constrained resource allocation to root growth under nutrient deficiency. Urea-only treatments (T2-T4) stimulated a modest, dose-dependent increase in root biomass. This suggests that nitrogen availability directly influences root growth, but the effect is limited without complementary soil amendments. The organic amendment alone (T5, T9) provided a mild stimulatory effect on root development, likely by improving soil structure and providing a slow-release nutrient source. The combined treatments (T6-T8) generated a significant boost in root biomass. The enhancement in both RFW and RDW was markedly greater than the arithmetic sum of the individual effects, indicating a true synergy. This improved root system under T6-T8 suggests better nutrient and water foraging capacity. Treatment T10 again proved superior, producing the most extensive and robust root system, as measured by both RFW and RDW. A well-developed root system is critical for sustained nutrient uptake and overall

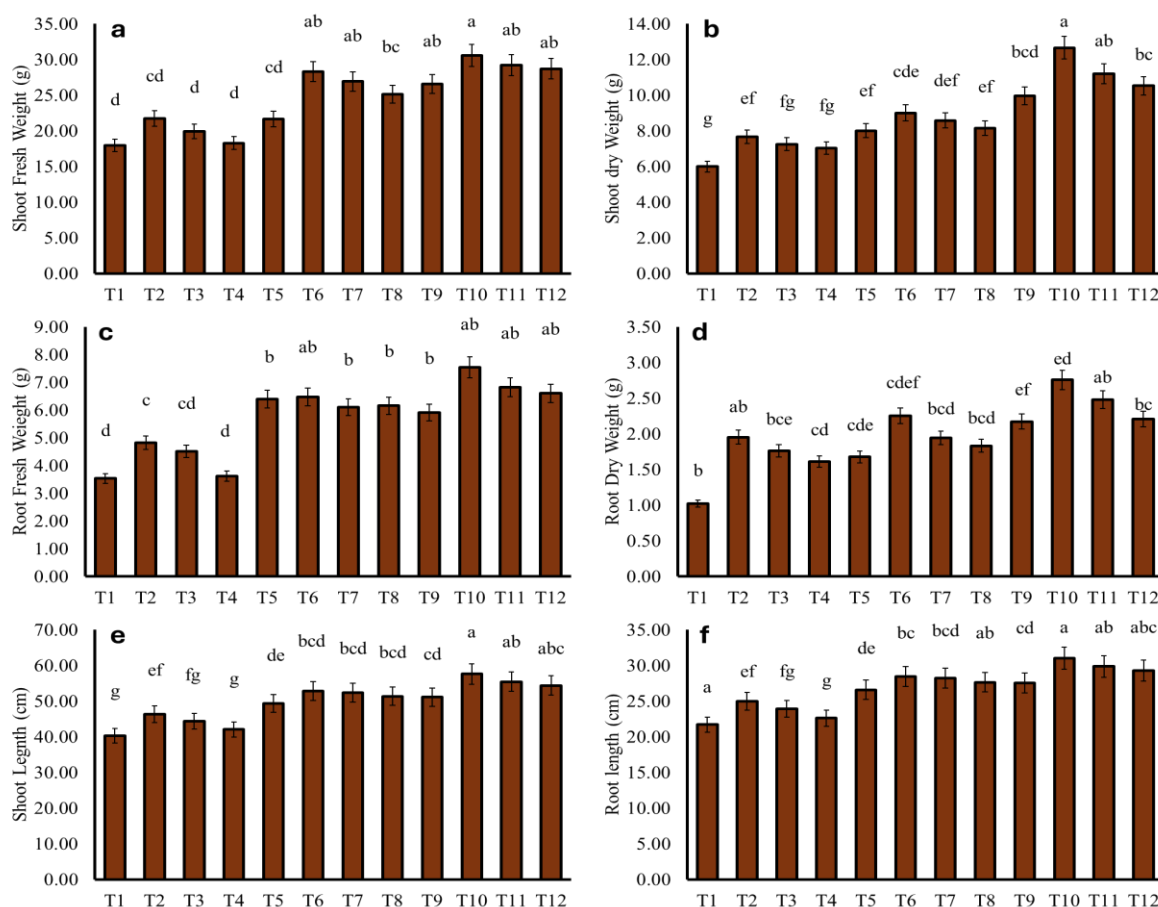


Figure 3a-f: Biomass Partitioning and Morphological Development of Wheat as Influenced by Integrated Nutrient Management. Error bars indicated the standard deviation (n=5) and different letters showed different levels of significance ($p<0.05$).

plant health, contributing to the superior shoot performance observed in T10. Treatments T11 and T12 fostered healthy root development but could not match the extensive system developed under T10, emphasizing that root investment is maximized under full nutrient integration.

The morphological traits of shoot and root length further detailed the plant's architectural response to nutrient management. The control (T1) resulted in the most stunted growth, with the shortest shoot and root lengths. For urea-only treatments (T2-T4), a clear gradient was observed: shoot and root length decreased progressively with reducing urea concentration. This demonstrates nitrogen's role as a key driver of cell elongation and division. The alkaline hydrolysis solution alone (T5, T9) promoted a slight but significant improvement in elongation over the control, hinting at the presence of growth-promoting substances or improved rhizosphere conditions. The most significant morphological enhancements were recorded in the integrated treatments (T6-T8). The combination led to noticeably taller shoots and longer roots compared to any single nutrient source. This improved architecture under combined application facilitates better light interception for shoots and a greater soil volume

exploration for roots. Treatment T10 consistently produced the tallest shoots and the most elongated root systems. This optimal architecture is a direct result of the synergistic relationship, likely involving enhanced hormonal signaling, nutrient availability, and soil physical properties. While T11 and T12 positively influenced plant morphology, their effects on length were intermediate, falling between the single applications and T10. This graded response solidifies the conclusion that the full combination of urea and alkaline hydrolysis solution is imperative for achieving the most favorable growth architecture in wheat.

4 Discussion

The results of this study demonstrate unequivocally that the integrated application of urea and a slurry-derived alkaline hydrolysis solution (AHS) synergistically enhance the growth, biomass, and yield-related parameters of wheat, surpassing the effect of either input applied alone. The superior performance of treatment T10 (100% urea + AHS) across all measured variables from root and shoot biomass to spike characteristics points to a complex interplay of improved nutrient supply, enhanced soil health, and

physiological efficiency. This discussion elucidates the potential mechanisms underpinning these observed synergies.

4.1. Synergistic Nutrient Release and Enhanced Nitrogen Use Efficiency

The most pronounced effect of the integrated treatment was on nitrogen-dependent growth parameters, such as plant height, shoot biomass, and spike development. While urea alone (T2-T4) provided a baseline level of nitrogen, its efficiency is often hampered by rapid hydrolysis, NH_3 volatilization, and nitrification, leading to significant losses (Oviedo Ventura, 2023). The presence of AHS likely mitigates these losses through multiple mechanisms.

First, the organic carbon and other compounds in the AHS can act as a carbon source for soil microorganisms. This microbial activity can facilitate immobilization, temporarily locking up mineral nitrogen in microbial biomass, and subsequently mineralizing it in a more synchronized manner with plant demand (Dong *et al.*, 2020). This "synchrony concept" reduces the asynchrony between N availability from urea and plant uptake, thereby enhancing Nitrogen Use Efficiency (NUE) (Govindasamy *et al.*, 2023).

Second, the alkaline nature of the hydrolysis solution might influence the urease enzyme activity and the subsequent fate of ammonium ions. While high pH can favor NH_3 volatilization, the complex organic matrix in the AHS could form transient complexes with urea or its hydrolysis products, slowing down the release rate and providing a more sustained nitrogen supply. This controlled release mechanism would explain the consistent superiority of the combined treatments (T6-T12) over the abrupt, and potentially wasteful, nutrient pulse from urea alone.

4.2. Improvement in Soil Physico-chemical and Biological Properties

The significant improvements in root growth (length, fresh, and dry weight) under integrated treatments, particularly T10, point to benefits beyond mere NPK nutrition. The application of organic amendments like AHS is known to improve soil structure by promoting aggregation, which enhances porosity, aeration, and water infiltration (Pereira *et al.*, 2018). An improved physical environment reduces soil mechanical resistance, allowing for better root penetration and proliferation, as observed in our root length and biomass data.

Furthermore, organic amendments serve as a substrate for soil microbes, increasing microbial biomass and diversity (Liu *et al.*, 2023). A vibrant microbial community contributes to nutrient cycling and produces plant growth-promoting substances, such as auxins and gibberellins, which can directly stimulate root and shoot elongation (Sun *et al.*, 2022). The moderate improvement in growth with AHS alone (T5, T9) supports this, indicating a direct bio-stimulatory effect. When combined with urea, this effect is amplified, as the abundant energy (carbon from AHS) and nutrient (N from

urea) sources create an ideal environment for microbial activity, fostering a positive plant-soil feedback loop.

4.3. Beyond Nitrogen: The Role of Secondary Nutrients and Rhizosphere Modulation

While urea primarily supplies nitrogen, the AHS is likely a source of other essential nutrients, including phosphorus (P), potassium (K), and a range of micronutrients, which were not measured in the shoot but are critical for overall plant development. The enhanced spike number and height in combined treatments suggest improved P availability, as phosphorus is vital for energy transfer and reproductive development (Zhang *et al.*, 2023). Organic acids and other compounds in the AHS can chelate cations, solubilize fixed P in the soil, and increase its bioavailability (Sindhu *et al.*, 2022).

Moreover, the integration of organic and inorganic inputs creates a more balanced nutrient environment. The "microbial bridge" hypothesis suggests that microbes mineralize organic nutrients from the AHS while simultaneously accessing inorganic N from urea, leading to a more balanced nutrient release profile (Scott and Prater, 2018). This balanced nutrition is critical for optimizing physiological processes, leading to the recorded increases in both fresh and dry biomass. Dry matter accumulation, in particular, is a function of photosynthetic efficiency and optimal resource partitioning, which is maximized when multiple nutrient constraints are alleviated.

4.4. Agronomic and Environmental Implications

The finding that T11 (75% urea + AHS) and T12 (50% urea + AHS) performed remarkably well, though slightly below T10, has significant agronomic and environmental implications. It suggests that integrating organic amendments with reduced chemical fertilizer rates can sustain high crop productivity while minimizing environmental footprints. This practice aligns with the principles of sustainable intensification, reducing the risk of groundwater contamination with nitrates and lowering greenhouse gas emissions associated with synthetic fertilizer production and use (Srivastav *et al.*, 2024).

AHS, being a derivative of agricultural slurry, also represents a viable pathway for nutrient recycling and waste valorization. By converting waste into a valuable agricultural input, this approach supports a circular economy in agriculture, reducing dependency on synthetic fertilizers and mitigating pollution from untreated waste.

5 Environmental Implications of Integrated Nutrient Management

The adoption of integrated nutrient management (INM), specifically the combination of urea with a slurry-derived alkaline hydrolysis solution (AHS), presents significant positive environmental implications that address several critical challenges in modern agriculture. The findings of this study demonstrate that this approach can contribute

substantially to sustainable agricultural systems by mitigating pollution, enhancing resource efficiency, and promoting soil health.

A primary environmental benefit lies in the potential for reduced nitrogen pollution. Conventional urea application is notoriously inefficient, with significant portions lost to the environment through ammonia volatilization, nitrate leaching, and denitrification as nitrous oxide (N_2O), a potent greenhouse gas. The organic compounds in the AHS likely act as a carbon source for soil microbes, fostering a temporary immobilization of mineral nitrogen. This process creates a "slow-release" effect, synchronizing nitrogen availability with plant uptake and thereby minimizing losses. This enhanced Nitrogen Use Efficiency (NUE) directly translates to lower fertilizer requirements, as evidenced by the strong performance of treatments with only 75% urea combined with AHS. Reducing the application rate of synthetic nitrogen fertilizers without compromising yield is a key strategy for shrinking agriculture's nitrogen footprint and protecting water bodies from eutrophication.

The diagram outlines the multi-faceted pathways through which the combination of chemical and organic nutrient sources enhances plant growth and soil health. Inputs (yellow nodes) include Urea, which provides readily available nitrogen (N), and AHS, an organic amendment derived from slurry. Key Processes (blue nodes) begin with the hydrolysis of urea into ammonium (NH_4^+). Concurrently, the AHS provides a carbon source that stimulates microbial activity, leading to the temporary immobilization of mineral N. This is followed by slow mineralization, creating a synchronized N release that matches plant demand. Additional processes include the direct release of phosphorus (P), potassium (K), and micronutrients from the AHS, improvement of soil physical structure, and activation of beneficial soil microbes. Core Mechanisms (green nodes) emerge from these processes, including enhanced Nitrogen Use Efficiency (NUE) via reduced losses (volatilization, leaching), improved root architecture due to better soil porosity, and a more balanced plant nutrition. Dashed and dotted lines highlight critical feedback loops; for example, improved root growth further enhances nutrient uptake efficiency. These synergistic mechanisms culminate in Final Outcomes (coral nodes): significantly improved soil health (increased organic matter, microbial biomass, and better structure), enhanced plant growth (increased biomass, height, and root development), optimized wheat yield, and notable environmental benefits through reduced N pollution, lower greenhouse gas emissions, and waste valorization within a circular economy framework.

Furthermore, this strategy embodies the principles of a circular economy. The AHS is a valorized product from agricultural slurry, a waste stream that otherwise poses a disposal challenge and is a known source of methane emissions and nutrient runoff. By processing this waste into a stable, nutrient-rich amendment, the system closes the

nutrient loop on the farm. This reduces dependency on synthetic fertilizers, whose production is energy-intensive and relies on non-renewable resources, thereby lowering the overall carbon footprint of crop production. It also provides a sustainable alternative to the direct, unmanaged application of raw manure, which can lead to phosphorus buildup in soils and contamination of water resources.

Moreover, the long-term application of such organic-inorganic combinations builds soil organic carbon stocks and improves soil structure. Healthy soils with high organic matter have greater water-holding capacity, reducing the need for irrigation and enhancing resilience to drought. Improved soil structure also reduces erosion, preventing the loss of topsoil and the associated transport of nutrients and pesticides into waterways. The stimulation of a diverse soil microbiome, as promoted by the AHS, supports robust nutrient cycling and soil health, creating a self-sustaining system that requires fewer external inputs over time.

In conclusion, integrating urea with a slurry-derived alkaline hydrolysis solution is not merely an agronomic practice for yield enhancement; it is a multifaceted environmental strategy. It directly addresses the pressing issues of nitrogen pollution, agricultural waste management, and greenhouse gas emissions, while simultaneously fostering healthier, more resilient soils. Widespread adoption of such INM practices is crucial for developing productive agricultural systems that operate in harmony with, rather than to the detriment of, the surrounding environment.

6 Conclusion

In conclusion, the superior performance of the integrated urea and alkaline hydrolysis solution treatment is attributed to a confluence of mechanisms: (1) enhanced nitrogen use efficiency via synchronized mineralization-immobilization, (2) improved soil physical properties and microbial activity leading to better root development, and (3) a more balanced supply of macro and micronutrients. This study strongly advocates for the adoption of integrated nutrient management systems that combine organic amendments like AHS with chemical fertilizers. Such a strategy not only maximizes wheat productivity but also promotes long-term soil health and environmental sustainability. Future research should focus on quantifying NUE gains, characterizing the microbial community shifts, and conducting life-cycle assessments to fully evaluate the environmental benefits of this approach.

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9 Authors Contribution:

Moazam Batool and Muhammad Awais: Writing Original Draft; **Mohsin Ashfaq and Muhammad Junaid Nazir:** Supervision, Writing reviews and corrections.

10 Conflict of Interest:

Authors declare no conflict of interest.

11 Sustainable development goals:

This study directly supports SDG 2 (Zero Hunger) and SDG 12 (Responsible Consumption and Production).

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