



# Enrichment of Chlorophyll Content of Maize (*Zea mays* L.) Hybrid Through Biodegradable Polymer Coated Urea Fertilizers

Balaganesh Balashanmugavel<sup>1,2</sup>, Murali Subramani<sup>3</sup>, Jothimani Subbiah<sup>1</sup>, Pradeesh Kumar Thangavel<sup>4</sup>, Venkatesh Vunnam<sup>5</sup>, Subash Chandra Bose Kasiviswanathan<sup>6</sup>

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## ABSTRACT

**Background:** Nitrogen is essential for chlorophyll formation which significantly influencing the growth and yield of crops. This study aimed to enhance maize leaf chlorophyll content by ensuring a steady nitrogen supply throughout the growing period. A field experiment was conducted using biodegradable polymer coated urea as a slow release nitrogenous fertilizer to achieve chlorophyll enrichment in maize.

**Methods:** There were nine treatments including uncoated urea, various biodegradable polymer-coated urea and commercially available coated urea fertilizer formulations were compared.

**Result:** In enhancing the leaf area index and chlorophyll content of maize, pine oleoresin-coated urea (POCU) and humic acid-coated urea (HACU) showed superior performance over other urea fertilizers. Leaf area index (LAI), a key indicator of canopy development, average LAI was significantly higher in POCU (2.71) and HACU (2.70) treatments which was 32.8 and 32.4% higher than uncoated urea treatment respectively. This leading to higher photosynthetic efficiency and biomass accumulation resulted, higher chlorophyll content in maize by POCU and HACU treatments. In particular, POCU showed maximum chlorophyll 'a' (1.58 mg mL<sup>-1</sup>) and 'b' (0.49 mg mL<sup>-1</sup>) content as well as total chlorophyll content (2.07 mg mL<sup>-1</sup>) at various growth stages of maize. This indicates that these slow-release formulations provided a sustained supply of nitrogen, promoting chlorophyll synthesis and enhancing photosynthetic activity. Additionally, the positive correlation between LAI and chlorophyll content in maize plants, led to improved photosynthetic efficiency, biomass accumulation and ultimately higher yields.

**Key words:** Chlorophyll, Coated urea, Leaf area index, Nitrogen, Slow release fertilizers.

## INTRODUCTION

Nitrogen (N), a pivotal element essential for plant development, is often provided through fertilizers to bolster agricultural systems (Mahboob *et al.*, 2023). Urea, boasting the highest N content, reigns as the dominant fertilizer choice for farmers worldwide. Nevertheless, concerns have been raised regarding the rapid-release nature of urea and its potential contribution to nutrient leaching, environmental contamination and uneven nutrient availability to plants (Trenkel, 2021). To address these issues, Slow-release urea fertilizers have been developed to gradually release nitrogen into the soil, thereby assisting crops in achieving higher yields. Maize requires a lot of nutrients to produce well and out of all the common nutrients these plants need nitrogen is the one they crave the most. The greenness of plants, especially in relation to nitrogen, serves as a visible indicator of nitrogen status. In maize, a deficiency of nitrogen can result in a reduction in chlorophyll content, leading to chlorosis, where leaves turn yellow due to insufficient chlorophyll levels (da Silva *et al.*, 2024). Conversely, providing maize plants with adequate nitrogen can boost chlorophyll content, promoting healthier and more robust growth (Singh *et al.*, 2022).

Nitrogen is a key component of chlorophyll molecules, so an adequate supply of nitrogen is essential for the synthesis of chlorophyll (Gao *et al.*, 2023), the pigment responsible for photosynthesis (Ahmed *et al.*, 2020).

<sup>1</sup>Department of Soil Science and Agricultural Chemistry, Tamil Nadu Agricultural University, Coimbatore-641 003, Tamil Nadu, India.

<sup>2</sup>School of Agricultural Sciences, Karunya Institute of Technology and Sciences, Coimbatore-641 111, Tamil Nadu, India.

<sup>3</sup>School of Agriculture, Vels Institute of Science, Technology and Advanced Studies, Chennai-600 001, Tamil Nadu, India.

<sup>4</sup>VIT School of Agricultural Innovations and Advanced Learning (VAIAL), Vellore-632 001, Tamil Nadu, India.

<sup>5</sup>Coromandel Fertilizers Ltd., Andhra Pradesh, India.

<sup>6</sup>Mother Teresa College of Agriculture, Pudukkottai-622 001, Tamil Nadu, India.

**Corresponding Author:** Balaganesh Balashanmugavel, Department of Soil Science and Agricultural Chemistry, Tamil Nadu Agricultural University, Coimbatore-641 003, Tamil Nadu, India. Email: balaagri007@gmail.com

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Chlorophyll is an important indicator for the evaluation of plant photosynthesis ability and growth status. Photosynthesis is a fundamental process in plants that converts light energy into chemical energy, allowing them

to produce glucose and oxygen from carbon dioxide and water (Sharkey, 2020). This process is crucial for plant growth and development, including crop yield. Maize can regulate the growth and development by adapting to the absorbed light profile, which is closely related to canopy growth and physiological indexes (Lacasa *et al.*, 2022; Liu *et al.*, 2023). The efficient management of nutrients in maize cultivation is integral to achieving optimal growth, yield and nutritional quality (Gezahegn, 2021). Central to these considerations is the role of chlorophyll, the pigment central to photosynthesis and indicative of a plant's photosynthetic efficiency and overall health (Meghwal *et al.*, 2024).

It's important to note that while nitrogen is essential for chlorophyll production, excessive nitrogen application can also have negative effects on maize plants, such as lodging and increased susceptibility to diseases (de Bang *et al.*, 2021; Sun *et al.*, 2020; Anas *et al.*, 2020). Therefore, it's important to carefully manage nitrogen fertilizer applications to optimize chlorophyll production and overall plant health. Responding to these challenges, the agricultural community has increasingly turned its attention to the potential benefits of slow-release nitrogen fertilizers.

The relationship between leaf area index (LAI) and chlorophyll content is intricate, as both parameters are fundamental to understanding plant growth and productivity (Chen *et al.*, 2022). LAI represents the total leaf surface area per unit ground area and is a key indicator of the plant's ability to capture light energy for photosynthesis. A higher LAI generally indicates a more extensive leaf canopy, which can potentially intercept more light and enhance photosynthetic activity (Li *et al.*, 2024; Adebayo *et al.*, 2021). An increase in LAI often correlates with a higher chlorophyll content, as a larger leaf area provides more surface area for chlorophyll-containing chloroplasts to capture light energy. This can lead to increased photosynthetic activity and, ultimately, higher plant productivity. Conversely, a decrease in LAI can result in reduced light interception and lower chlorophyll content, limiting photosynthetic efficiency and potentially reducing crop yield.

## MATERIALS AND METHODS

The field experiment was conducted in Coimbatore district of Tamil Nadu, India using maize hybrid (COH (M) 6) as test crop during 2022. The soil was classified as mixed black calcareous, fine, montmorillonitic and isohypert-hermic (Inceptisol order, Periyanaickenpalayam soil series, *Vertic ustropept*) and its physico-chemical properties were characterized and presented in Table 1.

Three biodegradable polymer coated urea were developed under laboratory using palm stearin, pine oleoresin and humic acid as coated materials. Based on the results of pot culture experiment, from each coating materials, PSCU 10%, POCU 4% and HACU 15% treatments screened for field experiment to compare the efficiency with popular coated urea fertilizers available in the market such as neem oil, sulphur, pungam oil and zinc coated urea. The treatments in the experiment were as

follows: T<sub>1</sub>- control treatment received no fertilizers, T<sub>2</sub>- uncoated urea (UCU), T<sub>3</sub>- palm stearin coated urea (PSCU), T<sub>4</sub>- pine oleoresin coated urea (POCU), T<sub>5</sub>- humic acid coated urea (HACU), T<sub>6</sub>- neem oil coated urea (NOCU), T<sub>7</sub>- sulphur coated urea (SCU), T<sub>8</sub>- pungam oil coated urea (PUOCU) and T<sub>9</sub>- zinc coated urea (ZCU). Fertilizer recommendations, tailored to the specific soil series, were N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O @ 200.9:68.1:61.3 kg ha<sup>-1</sup>. Leaf area index and chlorophyll content were measured on five randomly tagged plants at different growth stages using the following procedures.

### Leaf area index

Leaf area index is defined as the ratio of leaf area occupied by the plant to the land area and was worked out as below (Yoshida *et al.*, 1976).

$$\text{leaf area index (LAI)} = \frac{\text{leaf area plant}^1 (\text{cm}^2)}{\text{Ground area}(\text{cm}^2)}$$

### Chlorophyll 'a' and 'b' and total chlorophyll

Leaf samples were collected at vegetative, tasselling and harvest stages of maize washed with distilled water to remove the dust. The leaf sample (0.2 g) were ground and 10 mL of acetone was added and centrifuged. The supernatant was collected and the absorbance was measured at 645 and 663 nm. Chlorophyll a, b and total content were calculated using the following formulae.

$$\text{Chlorophyll 'a' (mg mL}^{-1}\text{)} = \frac{12.7 (\text{A663}) \times 2.69 (\text{A645}) \times V}{1000 \times W}$$

$$\text{Chlorophyll 'b' (mg mL}^{-1}\text{)} = \frac{22.9 (\text{A645}) \times 4.68 (\text{A663}) \times V}{1000 \times W}$$

$$\text{Total chlorophyll (mg mL}^{-1}\text{)} = \frac{20.2 (\text{A645}) \times 2.69 (\text{A663}) \times V}{1000 \times W}$$

Where,

V - Volume of purified solution (acetone).

W - Weight of leaf sample.

### Data analysis

Data pertaining to leaf area index and chlorophyll content from different urea fertilizer treatments underwent statistical analysis using the randomized block design (RBD) (Panse and Sukhatme, 1954). AGRES software utilized for the analysis, with one-way analysis of variance (ANOVA) to assess the statistical significance of differences among treatments. Significant variations between treatments were further compared using critical differences at a 5% confidence level. Images were developed using Origin software version 10.1.

## RESULTS AND DISCUSSION

### Leaf area index

The leaf area index (LAI) of maize plants, a crucial indicator of canopy development, varied significantly among

treatments due to the application of different coated urea fertilizers. The LAI values increased steadily until the vegetative (0.97) and tasselling (3.34) stages, indicating vigorous plant growth during these phases. However, a decline in LAI was observed at the harvest stage (2.38). Among the fertilized treatments, the highest mean LAI was recorded in the POCU (2.71) treatment, closely followed by the HACU (2.70) treatment. In contrast, the UCU (2.04) treatment exhibited the lowest mean LAI, indicating less vigorous growth compared to the other treatments (Table 2).

Specifically, at the vegetative stage, the UCU treatment showed the maximum LAI (1.12), which was comparable to the POCU (1.04) and HACU (1.08) treatments. Conversely, the ZCU treatment exhibited the minimum LAI (0.94) at this stage. At the tasselling stage, the HACU treatment displayed the highest LAI (4.35) which was at par with POCU treatment (4.16), indicating robust canopy development in these treatments. By the harvest stage, the POCU treatment showed the maximum LAI (2.91), which was on par with PSCU treatment (2.75). These results indicate that the POCU treatment promoted greater canopy development throughout the growth stages. In contrast, the UCU treatment exhibited the lowest LAI at both the tasselling (2.91) and harvest (2.10) stages, indicating less effective growth promotion compared to the other treatments (Fig 1).

### Chlorophyll 'a' and 'b' and total chlorophyll content

The research revealed notable differences in chlorophyll content among maize plants treated with various coated urea fertilizers throughout different growth stages. Average total chlorophyll content increased steadily until the vegetative (1.57 mg mL<sup>-1</sup>) and tasselling (1.81 mg mL<sup>-1</sup>) stages, but decreased at the harvest stage (1.65 mg mL<sup>-1</sup>) (Table 3). At vegetative stage, among the fertilized treatments, the PSCU treatment showed the highest chlorophyll 'a' content (1.41 mg mL<sup>-1</sup>), was on par with HACU (1.37 mg mL<sup>-1</sup>), POCU (1.34 mg mL<sup>-1</sup>), NOCU (1.33 mg mL<sup>-1</sup>) and SCU (1.33 mg mL<sup>-1</sup>) treatments, while the lowest was recorded in the UCU treatment (1.23 mg mL<sup>-1</sup>). PSCU 14.6% and HACU 11.4% increased chlorophyll 'a' over uncoated urea. For chlorophyll 'b' content, the highest was found in the HACU treatment [0.38 mg mL<sup>-1</sup> (11.8% higher chlorophyll than UCU)], which was at par with PSCU treatment (0.36 mg mL<sup>-1</sup>), while the lowest was in the SCU treatment (0.29 mg mL<sup>-1</sup>). The maximum total chlorophyll content was recorded in the PSCU treatment (1.77 mg mL<sup>-1</sup>) which is 12.7% higher over UCU. This PSCU treatment was on par with POCU (1.67 mg mL<sup>-1</sup>), HACU (1.75 mg mL<sup>-1</sup>) and NOCU (1.65 mg mL<sup>-1</sup>) treatments, while the minimum was in the UCU treatment (1.57 mg mL<sup>-1</sup>).

At the tasselling stage, the highest chlorophyll 'a' content was in the POCU treatment (1.58 mg mL<sup>-1</sup>), was on

**Table 1:** Physico-chemical characteristics of the soil.

| Soil parameters                                                       | Values | Method                      |
|-----------------------------------------------------------------------|--------|-----------------------------|
| pH (1:2.5 soil/water ratio)                                           | 8.17   | Jackson (1973)              |
| Electrical conductivity (dS m <sup>-1</sup> )(1:2.5 soil/water ratio) | 0.24   | Jackson (1973)              |
| Organic carbon (%)                                                    | 0.62   | Walkley and Black (1934)    |
| Available nitrogen (kg ha <sup>-1</sup> )                             | 179.2  | Subbiah and Asija (1956)    |
| Available phosphorus (kg ha <sup>-1</sup> )                           | 16.7   | Olsen (1954)                |
| Available potassium (kg ha <sup>-1</sup> )                            | 504.1  | Stanford and English (1949) |

**Table 2:** Leaf area index (LAI) at different growth stages of maize as influenced by different coated urea fertilizers.

| Treatments               | Leaf area index (LAI) |                    |                    |      |
|--------------------------|-----------------------|--------------------|--------------------|------|
|                          | Vegetative            | Tasselling         | Harvest            | Mean |
| T <sub>1</sub> : Control | 0.58 <sup>e</sup>     | 1.92 <sup>e</sup>  | 1.17 <sup>e</sup>  | 1.23 |
| T <sub>2</sub> : UCU     | 1.12 <sup>a</sup>     | 2.91 <sup>d</sup>  | 2.10 <sup>d</sup>  | 2.04 |
| T <sub>3</sub> : PSCU    | 0.97 <sup>cd</sup>    | 3.56 <sup>c</sup>  | 2.75 <sup>ab</sup> | 2.43 |
| T <sub>4</sub> : POCU    | 1.04 <sup>abc</sup>   | 4.16 <sup>ab</sup> | 2.91 <sup>a</sup>  | 2.71 |
| T <sub>5</sub> : HACU    | 1.08 <sup>ab</sup>    | 4.35 <sup>a</sup>  | 2.68 <sup>b</sup>  | 2.70 |
| T <sub>6</sub> : NOCU    | 1.02 <sup>bcd</sup>   | 3.90 <sup>b</sup>  | 2.57 <sup>bc</sup> | 2.50 |
| T <sub>7</sub> : SCU     | 0.99 <sup>cd</sup>    | 3.10 <sup>d</sup>  | 2.43 <sup>c</sup>  | 2.17 |
| T <sub>8</sub> : PUOCU   | 1.02 <sup>bcd</sup>   | 3.07 <sup>d</sup>  | 2.39 <sup>c</sup>  | 2.16 |
| T <sub>9</sub> : ZCU     | 0.94 <sup>d</sup>     | 3.12 <sup>d</sup>  | 2.42 <sup>c</sup>  | 2.16 |
| Mean                     | 0.97                  | 3.34               | 2.38               | 2.23 |
| SEd                      | 0.038                 | 0.139              | 0.105              |      |
| CD (p=0.05)              | 0.081                 | 0.295              | 0.223              |      |

\*The same alphabet letter indicated no significant differences found between the treatment.

par with PSCU ( $1.49 \text{ mg mL}^{-1}$ ), HACU ( $1.52 \text{ mg mL}^{-1}$ ) and NOCU ( $1.48 \text{ mg mL}^{-1}$ ) treatments, with the lowest in the ZCU treatment ( $1.36 \text{ mg mL}^{-1}$ ). The maximum chlorophyll 'b' content was in the HACU treatment ( $0.51 \text{ mg mL}^{-1}$ ), which was on par with PSCU ( $0.47 \text{ mg mL}^{-1}$ ) and POCU ( $0.49 \text{ mg mL}^{-1}$ ) treatments, while the minimum was in the UCU treatment ( $0.36 \text{ mg mL}^{-1}$ ). The maximum total chlorophyll content was recorded in the POCU treatment ( $2.07 \text{ mg mL}^{-1}$ ), which was on par with HACU ( $2.03 \text{ mg mL}^{-1}$ ), PSCU ( $1.96 \text{ mg mL}^{-1}$ ) and NOCU ( $1.94 \text{ mg mL}^{-1}$ ) treatments, with the minimum in the UCU treatment ( $1.75 \text{ mg mL}^{-1}$ ).

At the harvest stage, the highest chlorophyll 'a' content was in the POCU treatment ( $1.48 \text{ mg mL}^{-1}$ ), which was on par with HACU ( $1.42 \text{ mg mL}^{-1}$ ) and NOCU ( $1.37 \text{ mg mL}^{-1}$ ) treatments, with the lowest in the UCU treatment ( $1.17 \text{ mg}$

$\text{mL}^{-1}$ ). The maximum chlorophyll 'b' content was in the POCU treatment ( $0.50 \text{ mg mL}^{-1}$ ), was on par with HACU ( $0.47 \text{ mg mL}^{-1}$ ) treatment, while the minimum was in the UCU treatment ( $0.31 \text{ mg mL}^{-1}$ ). The maximum total chlorophyll content was recorded in the POCU treatment ( $1.98 \text{ mg mL}^{-1}$ ), which was on par with HACU ( $1.89 \text{ mg mL}^{-1}$ ) treatment, with the minimum in the UCU treatment ( $1.48 \text{ mg mL}^{-1}$ ). Based on the results, the coated urea fertilizers had varying effects on chlorophyll content in maize plants across different growth stages. The POCU treatment consistently performed well, showing high chlorophyll 'a' and 'b' content as well as total chlorophyll content at various stages. The HACU treatment also showed promising results, especially in terms of chlorophyll 'b' content. The PSCU, NOCU and SCU treatments also performed

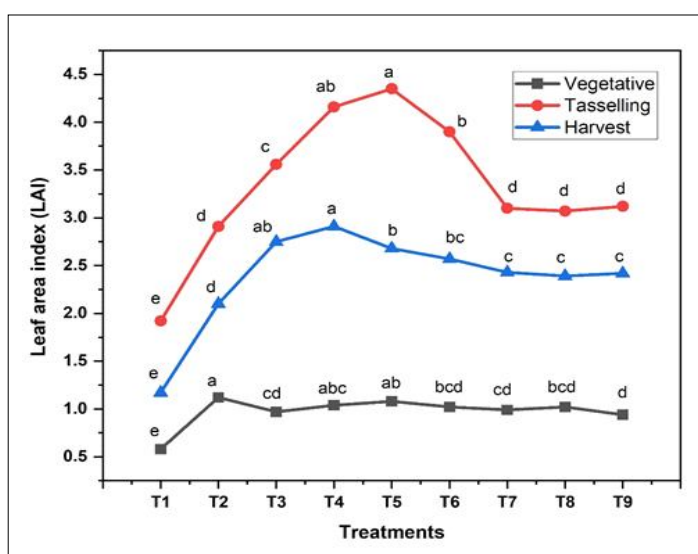


Fig 1: Effect of different urea fertilizers on leaf area index of maize at different growth stages.

Table 3: Chlorophyll a and b ( $\text{mg mL}^{-1}$ ) content at different growth stages of maize as influenced by different coated urea fertilizers.

| Treatments               | Chlorophyll content ( $\text{mg mL}^{-1}$ ) |                    |                    |                     |                     |                    |                     |                    |                    |
|--------------------------|---------------------------------------------|--------------------|--------------------|---------------------|---------------------|--------------------|---------------------|--------------------|--------------------|
|                          | Vegetative                                  |                    |                    | Tasselling          |                     |                    | Harvest             |                    |                    |
|                          | Chlorophyll 'a'                             | Chlorophyll 'b'    | Total chlorophyll  | Chlorophyll 'a'     | Chlorophyll 'b'     | Total chlorophyll  | Chlorophyll 'a'     | Chlorophyll 'b'    | Total chlorophyll  |
| T <sub>1</sub> : Control | 0.75 <sup>d</sup>                           | 0.22 <sup>f</sup>  | 0.97 <sup>c</sup>  | 0.83 <sup>e</sup>   | 0.26 <sup>g</sup>   | 1.09 <sup>d</sup>  | 0.68 <sup>e</sup>   | 0.19 <sup>f</sup>  | 0.87 <sup>f</sup>  |
| T <sub>2</sub> : UCU     | 1.23 <sup>c</sup>                           | 0.34 <sup>bc</sup> | 1.57 <sup>b</sup>  | 1.39 <sup>cd</sup>  | 0.36 <sup>f</sup>   | 1.75 <sup>c</sup>  | 1.17 <sup>d</sup>   | 0.31 <sup>e</sup>  | 1.48 <sup>e</sup>  |
| T <sub>3</sub> : PSCU    | 1.41 <sup>a</sup>                           | 0.36 <sup>ab</sup> | 1.77 <sup>a</sup>  | 1.49 <sup>abc</sup> | 0.47 <sup>abc</sup> | 1.96 <sup>ab</sup> | 1.35 <sup>bc</sup>  | 0.45 <sup>b</sup>  | 1.80 <sup>bc</sup> |
| T <sub>4</sub> : POCU    | 1.34 <sup>abc</sup>                         | 0.33 <sup>c</sup>  | 1.67 <sup>ab</sup> | 1.58 <sup>a</sup>   | 0.49 <sup>ab</sup>  | 2.07 <sup>a</sup>  | 1.48 <sup>a</sup>   | 0.50 <sup>a</sup>  | 1.98 <sup>a</sup>  |
| T <sub>5</sub> : HACU    | 1.37 <sup>ab</sup>                          | 0.38 <sup>a</sup>  | 1.75 <sup>a</sup>  | 1.52 <sup>ab</sup>  | 0.51 <sup>a</sup>   | 2.03 <sup>a</sup>  | 1.42 <sup>ab</sup>  | 0.47 <sup>ab</sup> | 1.89 <sup>ab</sup> |
| T <sub>6</sub> : NOCU    | 1.33 <sup>abc</sup>                         | 0.32 <sup>cd</sup> | 1.65 <sup>ab</sup> | 1.48 <sup>abc</sup> | 0.46 <sup>bc</sup>  | 1.94 <sup>ab</sup> | 1.37 <sup>abc</sup> | 0.45 <sup>b</sup>  | 1.82 <sup>bc</sup> |
| T <sub>7</sub> : SCU     | 1.33 <sup>abc</sup>                         | 0.29 <sup>e</sup>  | 1.62 <sup>b</sup>  | 1.46 <sup>bcd</sup> | 0.41 <sup>de</sup>  | 1.87 <sup>bc</sup> | 1.33 <sup>bc</sup>  | 0.39 <sup>c</sup>  | 1.72 <sup>cd</sup> |
| T <sub>8</sub> : PUOCU   | 1.28 <sup>bc</sup>                          | 0.30 <sup>de</sup> | 1.58 <sup>b</sup>  | 1.39 <sup>cd</sup>  | 0.38 <sup>ef</sup>  | 1.77 <sup>c</sup>  | 1.26 <sup>cd</sup>  | 0.35 <sup>d</sup>  | 1.61 <sup>de</sup> |
| T <sub>9</sub> : ZCU     | 1.29 <sup>bc</sup>                          | 0.30 <sup>de</sup> | 1.59 <sup>b</sup>  | 1.36 <sup>d</sup>   | 0.43 <sup>cd</sup>  | 1.79 <sup>c</sup>  | 1.34 <sup>bc</sup>  | 0.37 <sup>cd</sup> | 1.71 <sup>cd</sup> |
| Mean                     | 1.26                                        | 0.32               | 1.57               | 1.39                | 0.42                | 1.81               | 1.27                | 0.39               | 1.65               |
| SEd                      | 0.05                                        | 0.01               | 0.06               | 0.05                | 0.02                | 0.07               | 0.06                | 0.02               | 0.07               |
| CD (p=0.05)              | 0.11                                        | 0.02               | 0.13               | 0.11                | 0.04                | 0.14               | 0.12                | 0.03               | 0.14               |

\*The same alphabet letter indicated no significant differences found between the treatment.

relatively well in maintaining chlorophyll levels. Finally, POCU and HACU treatments appear to be the most effective coated urea fertilizers for enhancing chlorophyll content in maize plants, followed by PCU, NOCU and SCU treatments (Fig 2).

Uncoated urea registered the highest Leaf Area Index (LAI) up to the vegetative stage due to its early dissolution. However, this early availability of urea might not have been sustained in subsequent stages of maize growth, leading to a reduction in LAI as the growth progressed. This reduction indicates a potential decline in photosynthetic activity. In contrast, coated urea fertilizers exhibited a lower LAI during the vegetative stage because their slow-release properties might have inhibited vegetative growth. This resulted in smaller leaves, shorter internodes and ultimately (ulti *et al.*, 2012). However, the LAI steadily increased until the tasselling stage and then gradually decreased towards maturity.

Among the treatments, pine oleoresin coated urea (POCU) and humic acid-coated urea (HACU) were more effective in providing a steady supply of nitrogen during the post-silking stage. Their specific nutrient release patterns sustained a balanced nutrient supply, promoted higher LAI and supported better canopy development. This pattern can be attributed to the slow release of nitrogen, which

supported maize growth even after the tasselling stage. According to Ma *et al.* (2021), slow-release fertilizers not only enhanced leaf vitality and photosynthetic capacity but also delayed leaf senescence and increased nitrogen uptake after silking. As the crop absorbed sufficient nitrogen, this nutrient might have been redirected towards canopy expansion and grain filling.

The LAI and chlorophyll content in maize are closely related as they both reflected the physiological status and growth of plants. A higher LAI indicates a larger leaf canopy, which contributed to higher photosynthetic rates and chlorophyll production. Our study found that the POCU treatment had 15.4% higher chlorophyll content than the UCU treatment, commanded to higher nitrogen uptake and utilized the nitrogen properly for the chlorophyll synthesis. This result was in line with Singh and Sharma (2020). Our study further demonstrated, compared to UCU, the addition of nitrogen through POCU significantly increased 6.4, 18.3 and 33.8% of chlorophyll 'a' and 'b' and total chlorophyll at the vegetative, tasselling and harvest stages, respectively (Fig 3). Consistent release of nitrogen from POCU were readily available at the photosynthesis site, promoting chlorophyll content ultimately led to yield development. This might be because of enhanced functional leaf area and delayed leaf senescence, facilitated by phytohormones

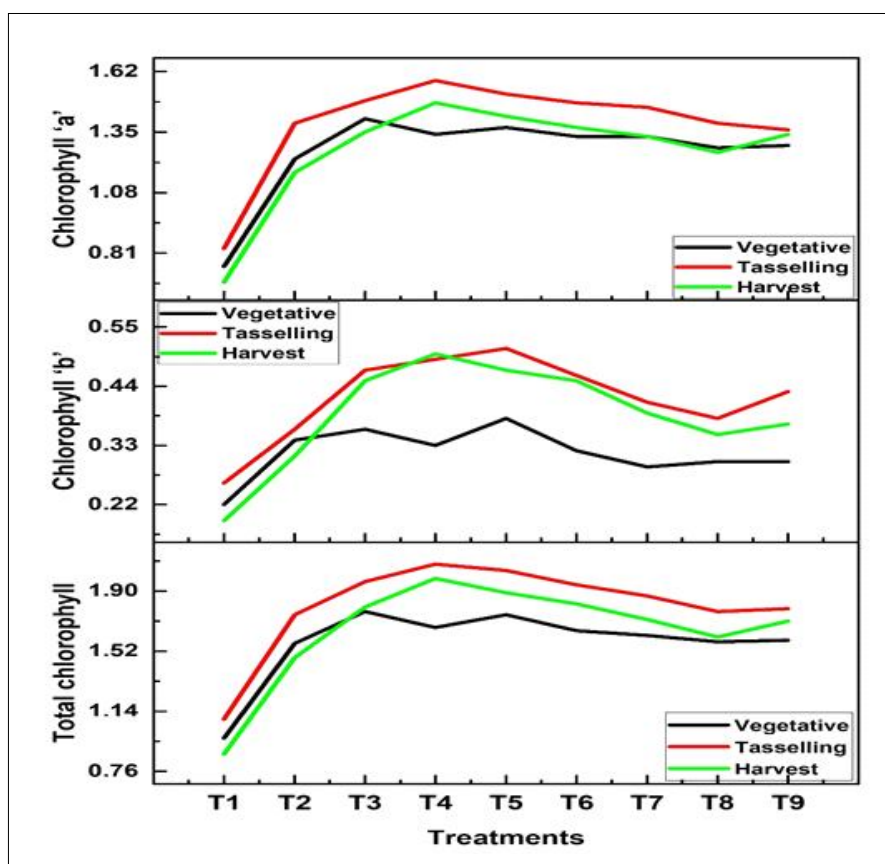


Fig 2: Effect of different urea fertilizers on chlorophyll content (mg mL<sup>-1</sup>) of maize at different growth stages.

promoting cell division and elongation due to continuous uptake of nitrogen from slow release fertilizers. Overall, the slow release N fertilizers produced significant leaf greenness level than UCU fertilizers. This finding was consistent with Gil-Ortiz *et al.* (2020) and Ashraf *et al.* (2019).

Outperformed treatments such as POCU and HACU treatments in terms of LAI and chlorophyll enrichment due to a great compatibility between coating material and urea. POCU provided a barrier for rapid release and the sustained release of nitrogen synchronized with crop growth stages. Additionally, pine oleoresin containing urease inhibitor facilitating the slow transformation of nitrogen oxidation process of  $\text{NH}_4^+\text{-N}$  to  $\text{NO}_3^-\text{-N}$ . Hence, prolonged nitrogen availability could be achieved in the soil by restricting different losses. A concomitant result was observed from Balaganesh *et al.* (2021). Similarly, HACU

started lower, consistently increased LAI and chlorophyll content, surpassing uncoated urea possibly due to gradual nutrient release and humic acid's growth-promoting effects. In addition, humic acid functional groups such as phenolic and carboxylic groups inhibit urease activity to reduce the rate of urea hydrolysis. This keeps the soil  $\text{NH}_4^+\text{-N}$  content at a low level, which reduces the risk of  $\text{NH}_3$  volatilization and potential nitrification, leading to lower N losses. This result was aligned with the finding of Kong *et al.* (2022).

Moreover, next to POCU and HACU, other coated urea fertilizers such as NOCU and SCU were also found to improve maize growth characteristics compared to UCU treatment. This improvement could be attributed to the inhibitor present in NOCU and the slow-release nature of SCU, which released nitrogen slowly and enhanced the plant growth characteristics by synchronizing with plant requirements. Overall, slow-release fertilizers have a

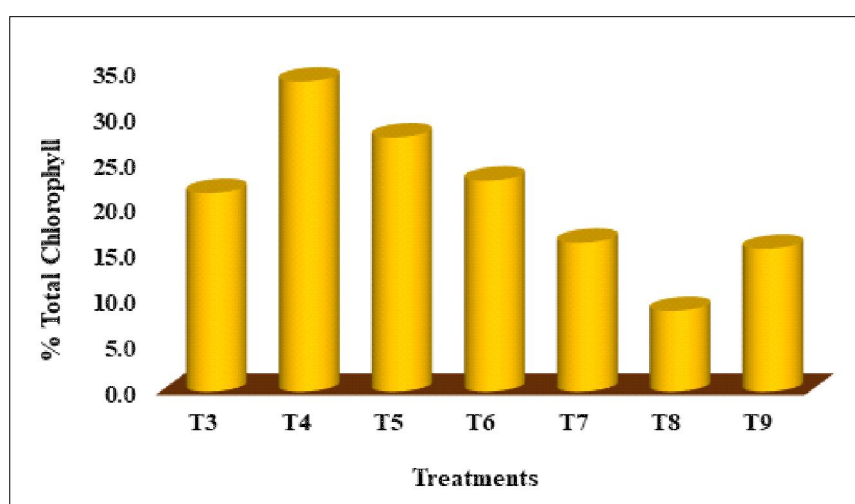


Fig 3: Effect of different coated urea fertilizers on percent increment of total chlorophyll over uncoated urea at harvest stage of maize.

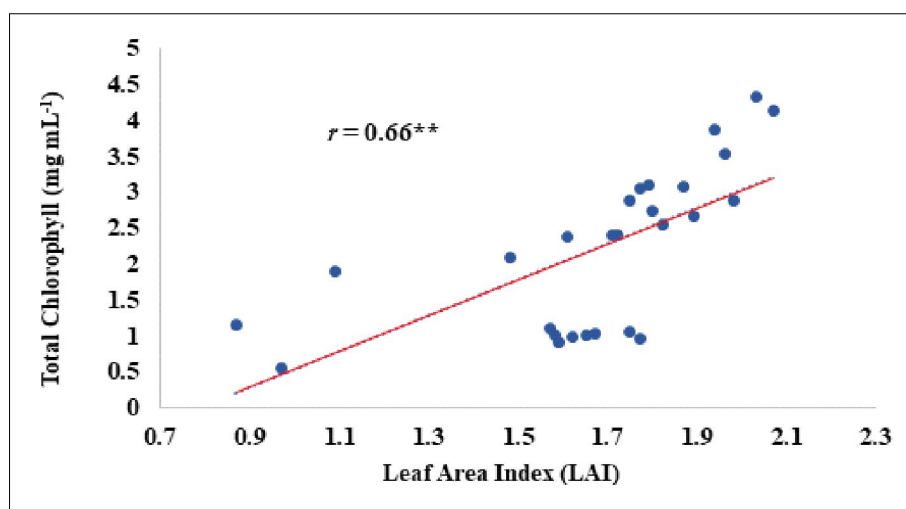


Fig 4: Correlation between leaf area index and chlorophyll content of maize.

positive and enhanced effect on the growth and yield of crops (Ghafoor *et al.*, 2022) by assimilation of chlorophyll transmitting captured light energy to PSII. This process is influenced by electron transfer and photochemical activity (Guo *et al.*, 2022).

The correlation between LAI and chlorophyll content could be attributed to the fact that a larger leaf area allows for more photosynthetic activity, which in turn leads to higher chlorophyll production. The positive correlation between LAI and total chlorophyll content ( $r=0.66$ ) indicated higher LAI conceded higher chlorophyll content (Fig 4). Therefore, treatments that promote higher LAI, such as the POCU and HACU treatments, are likely to also resulted in higher chlorophyll content. This result was in agreement with the findings of Singh and Sharma (2020); Bijay and Singh (2017); Zhang *et al.* (2019).

## CONCLUSION

In summary, POCU and HACU as the most effective coated urea fertilizers for enhancing maize growth and chlorophyll content, demonstrating their potential for improving maize canopy development and photosynthetic efficiency. These findings suggest that POCU could be promising option for optimizing maize production, warranting further investigation and potential adoption in agricultural practices. Future research may be focused on long-term field studies to assess the sustained effects of coated urea fertilizers on maize growth and yield. Additionally, investigating the underlying mechanisms and evaluating the environmental impact and economic viability of these fertilizers would be beneficial.

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## Conflict of interest

All authors declared that there is no conflict of interest.

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