Optimizing Green Bean Yield: Controlled Nitrogen Release with Nano-Urea-Modified Apatite

Muhammad A. Fathy [1], Mohamed I.D. Helal [1], Hassan A. Khater [1], Noha H. Abdelkader [1], Karima F. Abdelgawad [2]

[1] Soil Sciences Department, Faculty of Agriculture, Cairo University, Giza, Egypt
[2] Vegetable Crops Department, Faculty of Agriculture, Cairo University, Giza, Egypt

ABSTRACT

Controlled-release fertilizers (CRFs) have drawn significant attention because of their ability to improve plant nutrient uptake efficiency and mitigate environmental pollution. Current commercial CRFs need improvement to reduce synthesizing costs and to apply biodegradable materials and/or biorefinery wastes. In this study, CRFs were synthesized by treating nano-apatite with urea at different apatite:urea ratios (1:1 and 1:4). The biodegradable polymers, alginate, and lignin extracted from agricultural residue and paper manufacturing waste were used as single (alginate) and double (alginate and lignin) coating layers to form CRFs. In lab experiments, the N release behavior was studied in both water and soil. A cultivation experiment was carried out to study the efficiency of CRFs in the yield of green bean plants. The CRFs were applied with 3 N levels (75, 50, and 25% of the recommended dose). Both types of CRFs significantly increased the N release period to 24 days compared to 5 days for commercial urea. The total yield increased by 88 and 98% by applying double- and single-coated CRFs, respectively, at 75% N of the recommended dose compared with the full dose of conventional urea. In conclusion, applying CRFs at an N level of 25% of the recommended amount obtained the same yield as the full dose of conventional urea.

1. Introduction

Fertilizers are crucial for increasing crop yields and the availability of food to feed the world's population, which is expected to increase from its current 8 billion people to 9.5 billion by the year 2050 (Azeem *et al.*, 2014). Over half of this population growth will take place in Africa, according to the 2017 revision of the World Population Prospects (Dhlamini *et al.*, 2022). However, more than 20% of productive croplands in Africa have already been lost over the past four decades because of different processes of land degradation, such as water and wind erosion and chemical, physical, and biological degradation (Maria *et al.*, 2014). It will be necessary to increase global food production by 70% (Pourzahedi *et al.*, 2018). As a result, the agricultural sector will require a significant amount of traditional fertilizer. However, conventional fertilizers have numerous drawbacks, including low nutrient use efficiency (NUE) by plants, with N, P, and K NUEs ranging from 40 to 60%, 15 to 20%, and 40 to 50%, respectively (Beig *et al.*, 2022). It also causes many hazards and increases environmental pollution. It contaminates groundwater, waterways, and the air through the emission of hazardous gases; for example, nitrogen (N) fertilizers are lost by leaching from the soil by way of water-soluble nitrates and emitted as ammonia and nitrogen oxides (Kottegoda *et al.*, 2011). Therefore, it is critical to create novel fertilizer types that regulate the rate of nutrient release in smaller amounts, gradually and sustainably to boost plant uptake (Avila-Quezada *et al.*, 2022), as well as to reduce the impact of fertilizers on the environment (Alsaeedi and Alameer, 2023). In recent decades, many efforts have been made to develop controlled-release fertilizers (CRFs), such as coating conventional fertilizer with a variety of materials, such as sulfur, inorganic material, and petroleum-based material (Zhiyuan *et al.*, 2013). However, industrial production and large-scale applications of CRFs in agricultural fields are limited due to the high cost of the production process necessary to create these materials and their complicity (Ye *et al.*, 2013). Furthermore, the residue shells of these petroleum-based coating materials are non-

renewable and non-biodegradable, posing a risk to the soil environment (Briassoulis *et al.*, 2010). It is anticipated that coated nano-fertilizers will perform better than traditional slow-release fertilizers coated with polymers (DeRosa *et al.*, 2010) because of the extensive surface area of nanoparticles and their capability of holding nutrients and releasing them slowly and steadily in a more effective way that matches crop requirements (Tarafder *et al.*, 2020). There are many techniques for creating nano-fertilizers using some nanoparticles as carriers of nutrients, coating materials, or themselves as the source of nutrients (Liu and Lal, 2015). One of the most promising nanoparticles that may be used as both a carrier of N and a source of phosphorous is hydroxyapatite. This is not only because of its excellent biocompatibility and bioactivity but also because it is the main source of phosphorous (rock phosphate).

Coating as a form of encapsulation for CRFs has become a popular topic. Researchers are working to create various biodegradable, renewable coating materials, including fatty acids, starch, and cellulose. However, it is challenging to rely on these bio-based materials for large-scale manufacturing due to their hydrophilicity and high cost (Huang *et al.*, 2018; Xiao *et al.*, 2017). One of the most prevalent biopolymers in nature is lignin.

The papermaking industry produces large quantities of lignin as waste, and it is possible to recover low-cost lignin on a large scale. Green bean is one of the most important vegetable crops in Egypt. According to FAOSTAT data in 2021, Egypt's annual production of green beans grew to about 310 thousand tons. Due to its exceptionally high productivity rate, Egypt is the world's tenth-largest exporter of green beans. The current study aimed to develop controlled-release N fertilizers using renewable, biodegradable, inexpensive, and biocompatible materials as both single- and double-coated layers. Initially, nano-apatite was physically prepared by milling and then modified by urea, which is the most water-soluble and widely used N fertilizer. It was then encapsulated using bio-based materials. The inner coating layer was a hydrophobic alginate layer,

and the second was lignin. The products were characterized, and their performance in improving N release patterns was evaluated. Their efficiency in the growth and yield of green been was also tested.

2. Materials and Methods

2.1. Materials:

The analytical-grade apatite rock (18% P_2O_5) used in this study was purchased from EL-Ahram Mining Company, Cairo, Egypt. The two types of lignin used in this research, organosolv and alkaline lignin, were prepared in our lab. The black liquor was provided by the local company "Misr-Edfu Company for Paper and Printing", ,Aswan Egypt. Commercial urea $(NH_2)_2CO$ was purchased from the agro-market. Sodium alginate, calcium chloride, carboxymethyl cellulose, sodium hydroxide, and sulfuric acid were purchased from LOBA CHEMIE, Mumbai, India.

2.2. Preparation of Lignin:

The hard date palm fibrillum was used in the organosolv lignin extraction process because of its availability as an agricultural residue. The biomass was treated with a mixture of formic and acetic acids, according to the method described by Watkins *et al.* (2015). Additionally, lignin was extracted from alkaline black liquor, which is a bio-refinery residue produced during paper manufacturing. It was prepared using the Kraft method (Figueiredo *et al.*, 2018), in which black liquor was produced as a result of treating bagasse with 1.5 M sodium hydroxide at a biomass:NaOH solution ratio of 1:10 for 120 min at 170°C, treating with sulfuric acid until the pH dropped to 5 to precipitate lignin, and then filtering and washing several times before being air-dried. Fourier-transform infrared spectroscopy (FTIR) was used to characterize both types of lignin.

2.3. Preparation of Nano-Apatite:

Nano-apatite was prepared in the solid phase by milling apatite rock using a desktop vibrating ball mill machine (Model PH-BML 912, Photon Scientific Company, Qalyub, Egypt). The milling process was carried out at 600 rpm for 36 h by adding apatite via four parallel cells made of stainless steel with 3 volumes of stainless steel balls under atmospheric pressure at room temperature. The product was characterized using X-ray diffraction (XRD), transmission electron microscopy (TEM), and FTIR.

2.4. Preparation of Single-Coated Nano-Urea-Modified Apatite (SCRFs):

A suspension of nano-apatite was prepared by dispersing 25 g of apatite in 100 mL of distilled water using ultrasonic mixing (30 kHz for 30 min), stirring in a saturated urea solution at two ratios (1:1 and 1:4 nano-apatite:urea) at room temperature for 12 h to let urea attached to the surfaces of nano-apatite, heating on a hot plate at 80°C before adding a 2% (w/v) Na-alginate solution, and stirring until a gel formed. After allowing the gel to stabilize for 24 h, it was carefully dropped with a syringe into a $CaCl_2$ (4% w/v) solution to form beads. These beads were left in a calcium chloride solution for 20 min to achieve cross-linking. The beads were gathered and dried at 60°C in an oven (Zhang *et al.*, 2020). Similarly, single-coated urea without apatite was used as a control. Urea-modified apatite was characterized using FTIR before alginate was added.

2.5. Preparation of Double-Coated Nano-Urea-Modified Apatite (DCRFs):

A mixture of carboxymethyl cellulose (CMC) solution (0.5% w/v) and a solution of 3.5 g lignin were dissolved in 100 mL of 1 M sodium hydroxide at a ratio of 2:8. This mixture was used for the physical coating of the previously prepared dried homogeneous SCRF beads (20 mL mixture /50 g beads). The SCRF was inserted into a 20 rpm rotating drum. A hot air stream was used to continuously dry the beads inside the rotary drum while the mixture was sprayed every 2 min at a distance of 25 cm from the spray nozzle to the rotary drum's center and continuously dried with hot air steam (60°C) (Fertahi *et al.*, 2020).

2.6. Characterization of SCRF, DCRF, and Their Constituents:

FTIR was performed in a range from 400 to 4000 cm^{-1} region using an FTIR spectrophotometer (4100 JASCO, Tokyo, Japan), with an accumulation of 16 scans while operating in the transmittance mode at a resolution of 4 cm^{-1}. For apatite nanoparticles, urea-modified apatite, and lignin, to explore the surface bond and trace the changes taking place in these bonds as a result of binding between urea and nano-apatite, the XRD patterns of nano-apatite samples were recorded using a diffractometer (Bruker D8 Discover, Berlin, Germany). The X-ray powder diffractometer used Cu Kα radiation (λ= 0.1546 nm) over a 2θ range of 5–80° with a step size of 0.01° and a step time of 1 s. A TEM (JEOL JEM 2100 electron microscope, Tokyo, Japan, operating at 200 keV) was used to measure the particle size and explore the morphology. The sample was dispersed in ethanol using ultrasonication for 15 min. The suspended nanoparticles were filtered with filter paper and then loaded on a carbon-coated copper grid (300 mesh); the sample was left to dry prior to inspection. Both imaging and selected area diffraction patterns were recorded, and scanning electron microscopy (SEM) (Zeiss Sigma 500 VP Analytical FE-SEM Carl, Zeiss Company, Oberkochen, Germany) was used to explore the particle morphology of the synthesized CRFs and coating porosity.

2.7. N Release Pattern of SCRF and DCRF Beads:

2.7.1. Solubility and N Release in Water

The solubility and release of N in water were studied in SCRFs, DCRFs, and commercial urea. Accurate weights of all studied fertilizers (0.2 g) were placed in a 50 mL Falcon tube, and 20 mL of distilled water was added to the beads and left for equilibrium periods of 24, 96, 168, 240, and 312 h. Clear solutions were collected at the end of each equilibrium period to determine the N released from the beads.

2.7.2. N Release in Soil

The release pattern of N from SCRFs, DCRFs, and commercial urea in clay loam soil was studied in a lab experiment. Soil samples weighing 30 g were placed in glass columns to a depth of 20 cm and saturated with a volume of distilled water equal to the water-holding capacity of the soil. Then, 0.5 g of the tested materials (SCRFs and DCRFs) were added to the soil surface and covered with a 2 cm layer of soil. Twenty milliliters of distilled water was added to each column, and the leachates were collected. This step was repeated several times for periods of 3, 6, 12, 18, and 24 days for each column. The total N released in the leachates was determined using the Kjeldahl method (Sáez-Plaza *et al.*, 2013).

2.8. Efficiency of SCRFs and DCRFs on the Growth and Yield of Green Beans:

The efficiency of SCRFs and DCRFs on the growth and productivity of green bean plants was evaluated in a field experiment carried out at the Experimental Station of the Faculty of Agriculture, Cairo University, Giza, Egypt. Seeds of cultivar sv1541GA (Bayer Company, Cairo, Egypt) were sown on 10th March 2022 in clay loam soil. The experimental soil was characterized by a pH of 7.58, electrical conductivity (EC) of 2.8 dSm^{-1}, organic carbon content of 1.65%,

particle size distribution of coarse sand of 4.4%, fine sand content of 31%, silt content of 27.3%, clay content of 37.3%, and clay loam texture. A randomized complete block design (RCBD) with one factor was used for this study. Two treatments of CRFs with three N levels (75, 50, and 25% of the recommended dose) and a control treatment of commercial urea (100% of the recommended dose) were added to green bean plants. Six replicates were used for each treatment. The CRFs used in this study represent both the single- and double-coated slow-release fertilizers with a nano-apatite:urea ratio of 1:4 (T1 and T2, respectively). All plants received 130 kg ha^{-1} P$_2$O$_5$ as triple superphosphate before transplanting, and potassium fertilizer was applied at a rate of 65 kg ha^{-1} K$_2$O as potassium sulfate in three portions (33 kg ha^{-1} at 15 days and 16 kg ha^{-1} each at 40 and 60 days after transplanting). Urea was added to three portions at a rate of 130 kg ha^{-1} N (recommended dose) for the control treatment. T1 (SCRF of N%=23.5%, and nano-apatite:urea ratio of 1:4) and T2 beads (DCRF of N%=23 %, and nano-apatite:urea ratio of 1:4) contained an amount of N equivalent to 75, 50, and 25% of the recommended dose (control) as SCRF and DCRF treatments. All fertilizers were applied to the surface of the soil. Using drip irrigation techniques, irrigation water with a pH of 7.25 and an EC of 0.42 dSm^{-1} was applied regularly to maintain the soil moisture at 75% of field capacity.

2.9. Vegetative Growth Characteristics:

Three randomly selected plants were sampled from each replicate 75 days after planting to determine plant length, number of branches/plant, plant fresh weight, and plant dry weight.

2.10. Yield and its Components:

The total yield per plant was determined at harvest (3 harvests), and it was expressed as g/plant. Ten pods from each replicate were used to determine the average pod weight.

2.11. Pod Chemical Composition:

One gram of fresh pods was extracted with 10 mL of N,N-dimethylformamide for 48hrs, and the extract was measured with a spectrophotometer (Dlab-SP-UV1000, China) at 663, 647, and 470 nm wavelengths to calculate the total chlorophyll content as mg/g according to Moran (1982). Vitamin C was extracted from the pods using oxalic acid, and the extraction was titrated with 2,6 diclorophenonindophenol, and vitamin C was expressed as mg/100 g fresh weight according to Abdelgawad *et al.* (2022). The total soluble sugar was extracted with 80% ethanol, and 5% sulfuric acid and phenol were added to the extract after filtration. The samples were measured using a spectrophotometer at a wavelength of 490 nm according to Dubois *et al.* (1956). The methanol extract was used to measure the antioxidant activity and total phenolic compounds, according to Vernon *et al.* (1999) and Baardseth *et al.* (2010).

2.12. Analytical Procedures:

The total N content in SCRFs, DCRFs, water, soil leachates, and green bean leaves was extracted using the wet digestion method described by George *et al.* (2013). The ammonia distilling unit (Kjeldahl Distillers, DNP series DNP-1500-MP, Raypa, Spain) was used to determine the N concentration in the acid extract. Using the N concentration in the acid digestion extract, the total N content in SCRFs and DCRFs, N released in water and soil, and N uptake by green beans were calculated. Additionally, the potassium concentration in the plant leaf tissues was measured in the digestion extract using Flame Emission Spectrophotometry (Corning 4100, Halstead Essex, UK). The pH of the irrigation water and the cultivation soil (with soil:water ratio of 1:2.5) was measured using a pH meter (Accumet AR.20, Fisher Scientific, Waltham, MA, USA). The EC was measured in the soil suspension's filtrate and irrigation water using an EC meter (JENWAY, London,UK). The total organic carbon (OC) content of the experimental soil was assessed using Walkley and Black's standard method (Dewis and Freitas, 1970), based on wet oxidation by dichromate. The texture triangle was used to determine the texture class after mechanical analysis was carried out using the pipette method (Gee and Bauder, 1986).

2.13. Statistical Analysis:

MSTAT-C (version 2.1, Michigan State University, East Lansing, MI, USA) was used to conduct data analysis. Analysis of variance (ANOVA) was used to compare the different treatments. Significant differences between treatments were ascertained using mean separation tests (also known as Duncan's multiple range test). Using SPSS (version 14), a correlation between the tested parameters was conducted.

3. Results and discussion

3.1. Characteristics of SCRFs, DCRFs, and Their Constituents:

3.1.1. Mineral Composition and Surface Chemical Bonds

The XRD pattern of the nano-apatite sample used in the preparation of SCRFs and DCRFs was recorded to explore its composition. According to the XRD pattern (Figure 1), it was identified as apatite, including two predominate phases typically matched with XRD patterns of fluorapatite according to reference card COD 9013553 and hydroxyapatite (HA) with reference card COD 9010050. The fluorapatite and hydroxyapatite peaks were observed between 30 and 35 and appeared at 31.8, 32.2, 32.9, and 34.1, referring to the peaks (211), (112), (300), and (202), respectively. That is, there was an isolated peak centered at 25.9 resulting from the (002) reflection of HA, and fluorapatite showed more intensity at three peaks (26.5, 28.1, and 29.5). Other impurities, such as calcium carbonate, soluble salts, and silicate components, might exist naturally in the environment in which the apatite was formed in a relatively low amount.

Figure 1. X-ray diffraction pattern of nano-apatite.
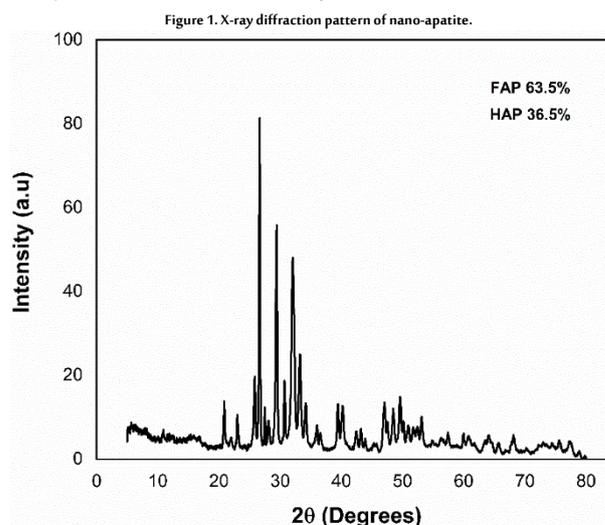
FTIR analysis was performed to explore the surface chemical bonds of nano-hydroxyfluorapatite (HF-apatite), tracing the conversion of binding due to the formation of a new product as a result of the reaction between urea and nano-HF-apatite (Figure 2A) and naming the differences in the functional groups and surface chemical bonds between organosolv lignin and alkaline lignin (Figure 2B). In both nano-HF-apatite and urea-modified apatite, the peaks related to PO$_4$$^{3-}$ appeared at 470, 572, 605, and 1040−1095 cm^{-1}, as shown in

Figure 2A (Kannan *et al.*, 2006; Liu *et al.*, 2003). Additionally, CO_3-derived bands were shown at 873 and 1424 cm^{-1} (Liu *et al.*, 2003; Okazaki *et al.*, 2005). The band observed at 3429 cm^{-1} in the HF-apatite graph represents the OH group. After treating HF-apatite with urea, four other peaks referring to NH bands were observed at 3350–3445 cm^{-1} and carbonyl group bands at 1625–1675 cm^{-1} (Madhurambal *et al.*, 2010), which confirmed the modification of HF-apatite with urea. The FTIR spectrographs of the two types of lignin (Figure 2B) showed that the lignin surface carried several chemical groups, such as O-H, C-H, C-O, C=C, and C-C stretching, corresponding to transmittance peaks of 3450, 2925, 1602, 1507, and 1466 cm^{-1}, respectively. However, the organosolv lignin graph also contained C=O bonds at 1702 cm^{-1}, which did not appear in the alkaline lignin. The carbonyl group that appeared in the lignin spectra extracted with organic acids was attributed to the esterification between the phenol and alcohol moieties of the propane chain during the pulping process (Watkins *et al.*, 2015). Although the chemical composition of lignin varied among plant families, species, plant growth stages, and extraction methods, the obtained data showed that the identified surface bonds of lignin types were almost similar. Some differences were observed between extraction methods, which demonstrated that alkaline lignin was preferred because of its inexpensive extraction method from black liquor compared with organosolv lignin extracted from plant tissue, which is laborious and requires several chemical reagents.

Figure 2. Fourier- transformer infrared spectrum of (A) nano-HF-apatite (black line), urea-modified HF-apatite (red line), (B) alkaline lignin (AL), and organosolv lignin (OL).
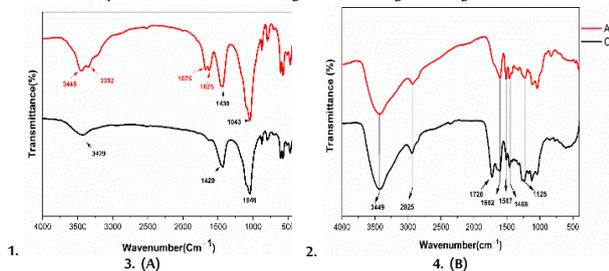
3.1.2. Size and Morphology of HF-apatite and CRFs

TEM was carried out to measure the nanoparticle size and to explore its morphology (Figure 3). The TEM images of nano-HF-apatite showed shapes attributed to the top-down preparation method (milling). The particle sizes were in the nano range, as shown in Figure 3A, with diameters ranging from 12 to 23 nm and less than 35 nm in length. The selected area of electron diffraction was evidence of the polycrystalline nature of the HF-apatite NPs, and the crystal lattice planes were perfectly aligned (Figure 3 B, C).

The images of two beads scanned by SEM, as illustrated in Figure 3D, E, showed the surface morphology of SCRFs and DCRFs. The appearance of the single-coating shell was smooth, compact, and homogenous with nano-HF-apatite, and only small cracks and pores were observed, indicating that alginate was an effective material for the fertilizer coating. However, lignin as a second coating layer showed more cracks, pores, and roughness compared with the alginate layer due to the preparation process used without using a crosslinker. Water molecules can penetrate this structure (lignin layer) easily if there is only a single-coating layer, causing a rapid release of N from the CRF, but the second layer is effective in reducing the reaction between water and fertilizer. This reflects the N release rate from CRFs and will help reduce the release rate. Although the alginate layer was perfect without many cracks or holes, its direct reaction with water increased the release rate compared to DCRF.

Figure 3. High-resolution transmission electron microscopy (HRTEM) image of (A) HF-apatite nanoparticles, (B) lattice fringes of HF-apatite nanoparticles, (C) electron diffraction patterns of apatite, (D) scanning electron microscopy (SEM) of SCRF, and (E) SEM of DCRF.
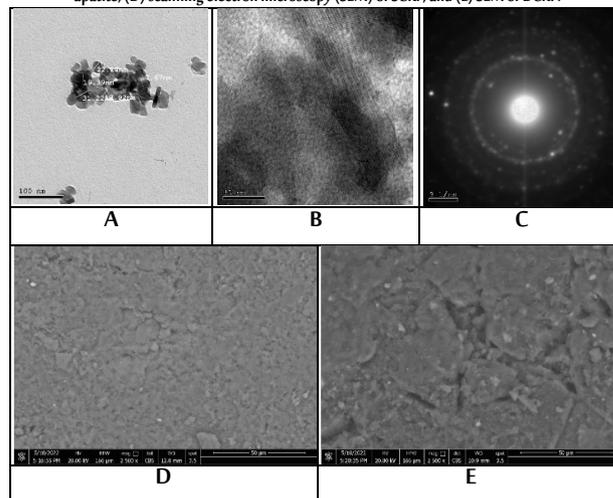
3.2. Total N Content in SCRFs and DCRFs:

The total percentages of N in all types of controlled release fertilizer ranged from 9 to 31% (Table 1). The wide range of total N could be attributed to the different ratios between HF-apatite and urea (1:1 and 1:4) as well as the coating type. The N percentages in DCRFs (lignin + alginate) were (9 and 23%) slightly lower than those (9.5 and 23.5%) of SCRFs (alginate), which could be attributed to the increase in the mass of the coating layer. The CRFs consisting of a ratio of 1:4 of HF-apatite and urea for both single- and double-coating types contained 23.5 and 23% N, respectively, which is higher than the amount of N in most nitrogenous fertilizers, except urea (46%) and ammonium nitrate (33%). However, the CRFs formed from HF-apatite and urea at a ratio of 1:1 contained a low N content (9.5 and 9%) in both SCRFs and DCRFs, respectively. This is an indication of the ability of nano-apatite to load high amounts of urea molecules because of its high surface area. It can hold the whole amount of urea added at a 1:1 ratio and more than twice that amount when treated with a higher ratio of 1:4 apatite:urea.

Table 1. Total nitrogen content (%) in single- and double-coated controlled-release fertilizers (SCRFs and DCRFs, respectively).

CRFs	Nano apatite: urea	SCRFs	DCRFs
Urea-modified apatite	1:01	9.5	9
Urea-modified apatite	1:04	23.5	23
Urea Without apatite		31	28

3.3. N Release from CRFs:

The results of the solubility and release of N from CRFs in water compared with commercial urea are given in Figure 4A. All types of CRFs showed significantly slower N release patterns compared with commercial urea, which was completely dissolved in the first portion of the added water. However, the release of N from CRFs sustains over 5 portions of added water for 13 days. Little differences were observed in the N release patterns of the 4 types of CRFs. However, the DCRFs showed slower N release rates for both apatite:urea ratios. At short periods of 1 and 4 days, the CRFs with a 1:4 apatite:urea ratio showed slower N release than those with a 1:1 ratio for both SCRFs and DCRFs. Additionally, the SCRFs with an apatite:urea ratio of 1:4 (blue line) showed a similar release pattern to the 1:1 DCRFs. During the longest period of 13 days, the slowest N release pattern was shown for DCRFs with a 1:4 ratio (purple line), followed by DCRFs with a 1:1 ratio (green line), whereas the other types (red and blue lines) showed the fastest ones. Similar previous results revealed that conventional urea dissolved immediately in water, whereas all slow-

release fertilizers (SRFs) coated with alginate and a lignin–clay nanohybrid had a slow-release capability (Zhang *et al.*, 2020). In addition, similar results were obtained for N release in water from CRF synthesized by coating urea with bio-based polymers, and the N release lasted 10 days (Zhang *et al.*, 2016).

Based on the fact that the beads with a higher ratio of nano-HF-apatite to urea (1:4) of both single and double coatings had the slowest N release pattern and the highest N content compared with the other ratios, they were used to study the release of N in soil, and the results are depicted in Figure 4B. Additionally, two types of beads of coated urea without HF-apatite were involved in this study, single-coated urea with alginate (S-urea) and double-coated urea with alginate plus lignin (D-urea), to explain the effect of the presence of nano-HF-apatite with urea in controlling N release. The N release periods of all types of CRFs in soil were much longer than in water. The release of N from commercial urea remained for 6 days, whereas its release from CRFs was sustained for 24 days, compared with 24 h and 12 days for their correspondence in water. The N release period increased plant nutrient use efficiency. The double-coated materials were highly effective in increasing release longevity. The N release patterns from coated urea (S-urea and D-urea) were significantly slower than from uncoated commercial urea. The amount of N released in 3 days from S-urea (red line) and D-urea (blue line) was 23 and 32%, respectively, compared to 75% for uncoated commercial urea. Additionally, N release from both types of coated urea remained for 12 days compared to 6 days for uncoated urea. However, the addition of nano-HF-apatite as a carrier for urea had a significant effect on the N release rate. Urea-modified HF-apatite CRFs showed the slowest N release pattern. That is, the release of N from their beads was sustained for 24 days for both SCRFs and DCRFs compared to S-urea and D-urea, which remained for only 12 days. Previous studies have mentioned that uncoated commercial urea completely dissolves after 5 days. The alginate-coated SRFs showed a slower release rate, and the SRF double coated with polyacrylic acid and an alginate/lignin–clay hybrid exhibited the slowest N release rate (Zhang *et al.*, 2020). In addition, the SRF synthesized using amorphous calcium phosphate as a carrier for urea demonstrated urea release five times slower than that of conventional urea (Carmona *et al.*, 2022).

Figure 4. The release pattern of controlled-release fertilizer compared with conventional urea (A) in water and (B) soil.

(A)　　　　　　　　(B)
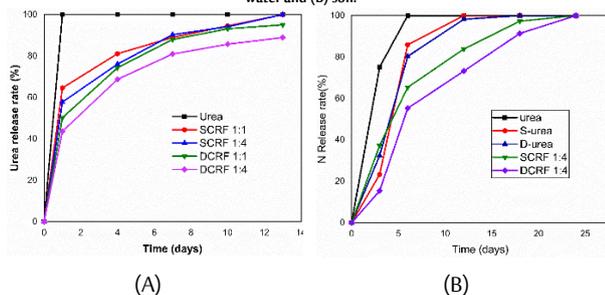
3.4. Effect of CRFs on the Growth and Production of Green Beans:

The green beans cultivation experiment showed that the two types of CRFs had a significant effect on vegetative growth parameters, N and K uptake, and total yield compared with commercial urea. The data listed in Table 2 indicate that the two types of CRF treatments at all N applied levels significantly improved all growth attributes (plant height, number of branches, plant fresh weight, and plant dry weight)

as well as N and K uptake compared with commercial urea. The 75% applied N level of the two types of CRFs showed the highest significant improvement for all growth attributes and yield of green beans, followed by the N level of 50%. The lowest applied N level (25%) for the SCRF (T1) had a positive effect on growth parameters and yield of green beans, similar to conventional urea (the control), which was added at 100% of the recommended dose. However, the DCRF (T2) applied at an N level of 25% showed a significant increase in both fresh and dry weight and N uptake compared with the 100% N level of commercial urea. The DCRF generally achieved vegetative growth parameters higher than the SCRF. This is consistent with the N release experiment, in which the DCRF had a slower N release rate than the SCRF, which led to a significant increase in N uptake, particularly at the 75% N level, as reflected in most vegetative growth parameters of green beans. N is an important macroelement for root and shoot communication. It is a key factor in plant metabolism, resource allocation, growth, development, and productivity (Yousaf *et al.*,, 2021).

Table 2. Green bean growth attributes as affected by CRF treatments.

Treatment	Level	Plant height (cm)	Number of branches	Plant Fresh weight (g)	Plant Dry weight (g)	N uptake (mg/plant)	K uptake (mg/plant)
T 1	75%	32.50 b	5.00 a	76.25 ab	21.88 b	488.26 b	166.12 b
	50%	30.50 bc	4.25 ab	71.25 b	17.13 bc	435.40 bc	143.98 bc
	25%	28.25 bc	3.25 bc	51.00 c	9.87 d	273.21 c	71.97 c
T 2	75%	37.25 a	4.50 a	89.75 a	31.13 a	853.27 a	245.72 a
	50%	31.00 bc	4.00 abc	67.75 b	17.38 bc	397.83 bc	127.78 bc
	25%	31.25 bc	4.00 abc	67.25 b	19.13 b	484.78 b	143.88 bc
Control	100%	27.25 c	3.00 c	52.75 c	11.88 cd	257.82 c	75.05 c

The two types of CRFs increased the total yield/plant as shown in Figure 5A, compared to conventional urea (control). The total yield per plant was completely improved in the two types of CRFs at the two N levels (75 and 50% of the recommended N dose) in comparison to level 25% and control treatment (urea). In the SCRF treatment, the total yield increased by 99 and 67% with 75 and 50% applied N, respectively. Additionally, the treatment with DCRF at 75 and 50% N levels increased the total yield by 88 and 61%, respectively, compared to the urea treatment. The total yield obtained at a 25% N level with both types of CRFs was equal to that of conventional urea applied at 100% of the recommended N dose. Therefore, we can achieve full green bean productivity using one-quarter of the traditional N fertilizer. Therefore, we can save 75% of the applied amount of N fertilizer, which has negative economic and environmentally significant effects. No significant differences were recorded in the total yield between SCRFs and DCRFs. Similar results were obtained by Kottegoda *et al.* (2017) using urea-modified hydroxyapatite without coating. They showed that yield increased by only 8% when applying N at the 50% level of the advised dose compared with the control (full dose of commercial urea). The average pod weight for the 25 and 50% N levels of both CRF types were similar to the full recommended dose of conventional urea, and no significant increase in the average pod weight was observed between conventional urea and the 75% N level for the SCRF. However, the DCRF at an N level of 75% significantly increased the pod weight compared to conventional urea, as shown in Figure 5B. The yield increment obtained for treatments with SCRFs and DCRFs could be attributed to the slow N release achieved by the coating techniques, which increased the soil residence time, decreased N losses, and improved the vegetative growth and total yield of green beans.

A

B

2009; Stewart *et al.*, 2001).

A

B

C

D

E
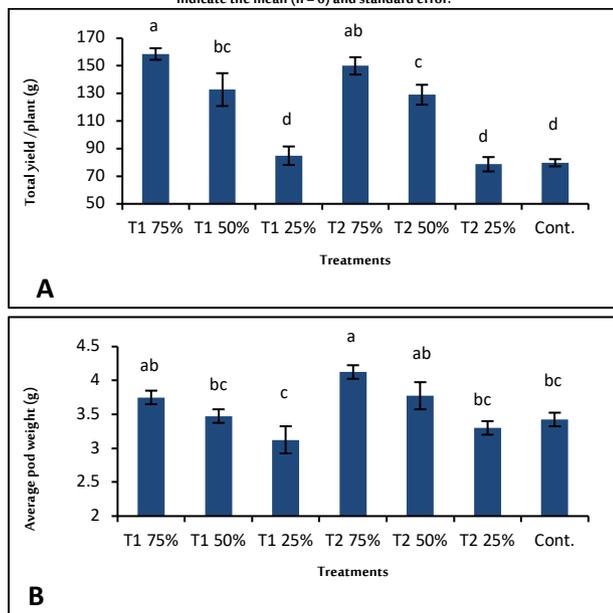
3.5. Effect of CRFs on Green Bean Pod Quality:

The pod chlorophyll content was similar for both treatments at all N levels, except for the 75% N level of both types of CRFs, which was significantly higher than the control (Figure 6A). The vitamin C content of green bean pods was affected by CRF treatments and N level, as shown in Figure 6B. The SCRF did not show a significant increase in vitamin C at 50 or 75% N levels compared to the control urea, while applying N at a level of 25% significantly decreased the vitamin C content to less than the control treatment. However, the DCRF showed a significant increase in vitamin C compared to the control treatment for all applied N levels. The highest vitamin C content for both types of CRFs was recorded at an N level of 50% of the recommended dose. Zhaoming *et al.* (2020) reported a similar outcome; they observed an initial increase in vitamin C with increasing applied N levels before declining at the highest N levels. Increasing N fertilizer application achieved green bean growth, which led to more photosynthesis and high vitamin C accumulation. Table 3 shows a positive correlation between vitamin C and plant fresh weight (0.504[**]), plant dry weight (0.510[**]), plant length (0.496[**]), and average pod weight (0.522[**]). Additionally, there was no significant difference in soluble sugar content between treatments, and no static significance was observed for any treatments except for the SCRF at the highest N level applied, as shown in Figure 6C. Antioxidant activity and phenolic compounds are responsible for vegetables' appearance, flavor, and color. Their favorable chemical characteristics also aid in the prevention of diseases such as cancer. The 75% N level in SCRFs and 50 and 25% N in DCRFs had the highest antioxidant capacity of green beans compared to all other treatments (Figure 6D). The control treatment recorded the same significance value for antioxidant capacity at N levels of 50 and 25% in SCRFs. Figure 6E illustrates that the SCRFs significantly increased the phenolic compounds at all applied N levels compared to the control, with no significant increase observed between the N levels. In contrast, the DCRF had a significant effect at the 50% N level, and the other N levels increased the phenolic compounds but not significantly. The high N content in the tissue that recorded the highest N uptake among all treatments caused a decrease in the phenolic compound content at 75% N in the DCRF. Similar results have been reported in several studies, as the high N concentration (a basic nutrient for protein synthesis) has a negative correlation with phenols (Smoleń and Sady

Fathy, M.A., Helal, M.I.D., Khater, H.A., Abdelkader, N.H. and Abdelgawad, K.F. (2024). Optimizing green bean yield: Controlled nitrogen release with nano urea-modified apatite. *Scientific Journal of King Faisal University: Basic and Applied Sciences*, 25(1), 13–20. DOI: 10.37575/b/agr/230070
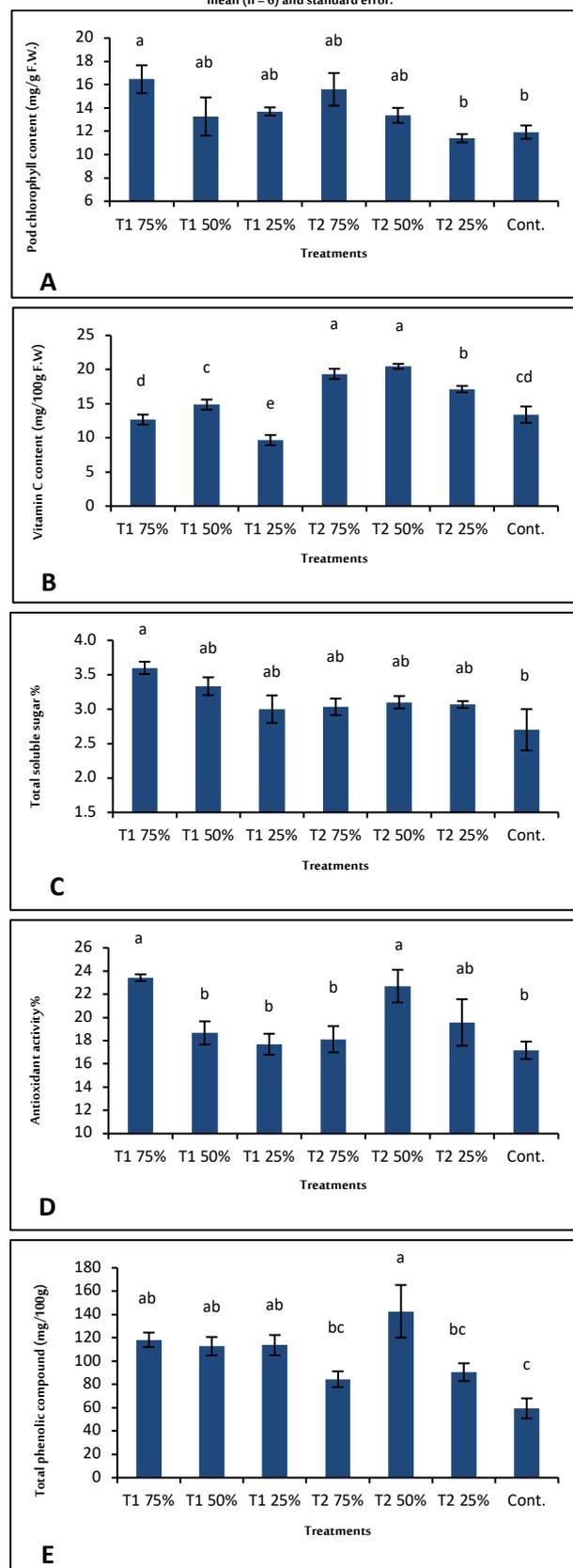
Table 3. Correlation coefficients (matrix) among green bean parameters.

	Total yield	Fresh weight	Dry weight	Number of branches	Plant length	K uptake	N uptake	Pod weight
F.W	0.670**							
D.W	0.557**	0.732**						
Number of branches	0.644**	0.639**	0.613**					
Plant length	0.566**	0.834**	0.778**	0.577**				
K uptake	0.550*	0.791**	0.956**	0.669**	0.815**			
N uptake	0.445*	0.733**	0.957**	0.522**	0.775**	0.935**		
Pod weight	0.565**	0.643**	0.642**	0.152	0.571**	0.592**	0.599**	
V.C	0.308	0.504**	0.510**	0.239	0.496**	0.466	0.475*	0.522**

**Correlation is significant at the 0.01 level; *Correlation is significant at the 0.05 level

4. Conclusions

In this study, we developed single- and double-coated nano-urea-modified HF-apatite CRFs to increase the period of N release in soil, which increased plant N uptake and agriculture production and reduced environmental pollution. The FTIR analysis revealed a similarity in the chemical functional groups for the two types of lignin; hence, it would be better to use alkaline lignin due to its low-cost extraction method and its abundance as a waste of the paper manufacturing process. Additionally, SCRFs and DCRFs had a significant effect on improving plant growth and enhancing green bean yield compared with commercial urea fertilizer. Despite the single-coating technique conferring a total yield higher than the double-coating technique, the DCRF had a slower N release rate, which means that its effect will continue to the next crop. Finally, we recommend using DCRFs at an N application level of 25% of the recommended dose to reduce fertilizer consumption without any reduction in green bean yield or application at the 75% N level to double productivity while reducing fertilizer consumption by 25%.

Biographies

Muhammad A. Fathy

Soil Sciences Department, Faculty of Agriculture, Cairo University, Giza, Egypt, 00201118252962, mohamed.fathy@agr.cu.edu.eg

Muhammad Fathy, Egypt, is a teaching assistant in the Soil Science Department; he graduated from the Faculty of Agriculture at Cairo University in 2017. After graduation, he worked as a research assistant on an international project. Two research groups were involved in this project; one of them was Egyptian and the other was an American group from Florida University. Recently, Muhammad was hired as the Vice Director of the Desert Land Reclamation and Development Central Laboratory, located at Cairo University Research Park. ORCID: 0000-0002-1973-0105

Mohamed I.D. Helal

Soil Sciences Department, Faculty of Agriculture, Cairo University, Giza, Egypt, 00201119540356, mohamed.dosoky@agr.cu.edu.eg

Prof. Mohamed Helal is an Egyptian professor. He received his Ph.D. from the Faculty of Agriculture at Cairo University. He held several positions, including Vice President of the Egyptian National Committee of Soil Sciences, Academy of Scientific Research and Technology, and Ministry of Scientific Research, Egypt, from 2014 to 2017. In addition, he was the supervisor of the Community and Environment Service Laboratory, Soil Science and Water Department, College of Agriculture and Veterinary Medicine, Al-Qassim University, Saudi Arabia, from 1998 to 2008. ORCID: 0000-0001-5900-6659

Hassan A. Khater

Soil Sciences Department, Faculty of Agriculture, Cairo University, Giza, Egypt, 00201000736300, hakhater2000@agr.cu.edu.eg

Prof. Hassan Khater is an Egyptian professor. He specializes in soil chemistry and organic farming. He received his Ph.D. from Cairo University. He supervised many research projects and worked as an associate professor in Libya. In addition, he supervised many master's and Ph.D. students. He was previously the supervisor of the Soil Reclamation and Development Centre, Faculty of Agriculture, Cairo University (2012—2015). He is a member of the International Union of Soil Science. He has published 31 publications in international and local scientific journals. ORCID: 0009-0008-2401-6353

Noha H. Abdelkader

Soil Sciences Department, Faculty of Agriculture, Cairo University, Giza, Egypt, 00201148247038, noha.abdulkader@agr.cu.edu.eg

Noha Hamdy is an Egyptian doctor with a Ph.D. in soil sciences from the Faculty of Agriculture, Cairo University. She has also supervised many master's students. She worked as a researcher for 3 years on a research project funded by the Science, Technology, and Innovation Funding Authority (STDF). Her minor specialization is soil reclamation, and she has published many articles in this field. Her research has been published in international and local scientific journals. She is also a reviewer for the Egyptian Journal of Soil Science. ORCID: not available

Karima F. Abdelgawad

Vegetable Crops Department, Faculty of Agriculture, Cairo University, Giza, Egypt, 00201223008531, karima.abdelgawad@agr.cu.edu.eg

Karima Abdelgawad is an Egyptian associate professor in the Faculty of Agriculture at Cairo University. She received her Ph.D. from Cairo University. She has published many articles in high-impact factor journals in the field of post-harvest treatments and vegetable crops. She has 18 international publications in the Scopus database in the world's largest publishers, including Elsevier, Springer, Taylor and Francis, and MDPI, and her h-index is nine. She has worked as a researcher on several local and international projects and supervised many master's and Ph.D. students. ORCID: 0000-0002-6960-2945

Acknowledgments

References

Abdelgawad, K.F., Awad, A.H., Ali, M.R., Ludlow, R.A., Chen, T. and El-Mogy, M.M. (2022). Increasing the storability of fresh-cut Green beans by using chitosan as a carrier for tea tree and peppermint essential oils and ascorbic acid. *Plants*, **11**(6), 783. DOI: 10.3390/plants11060783

AlSaeedi, A.H and Alameer, S.J (2023). The role of nanosilica in ameliorating the deleterious effect of salinity shock on cucumber growth. *Scientific Journal of King Faisal University: Basic and Applied Sciences*, **24**(1), 21—9. DOI: 10.37575/b/agr/230026

Avila-Quezada, G.D., Ingle, A.P., Golińska, P. and Rai, M. (2022). Strategic applications of nano-fertilizers for sustainable agriculture: Benefits and bottlenecks. *Nanotechnology Reviews*, **11**(1), 2123—40. DOI: 10.1515/ntrev-2022—0126

Azeem, B., KuShaari, K., Man, Z.B., Basit, A. and Thanh, T.H. (2014). Review on materials & methods to produce controlled release coated urea fertilizer. *Journal of Controlled Release*, **181**(1), 11—21. DOI: 10.1016/j.jconrel.2014.02.020

Baardseth, P., Bjerke, F., Martinsen, B.K. and Skrede, G. (2010). Vitamin C, total phenolics and antioxidative activity in tip‑cut green beans (Phaseolus vulgaris) and swede rods (Brassica napus var. napobrassica) processed by methods used in catering. *Journal of the Science of Food and Agriculture*, **90**(7), 1245—55. DOI: 10.1002/jsfa.3967

Beig, B., Niazi, M.B.K., Sher, F., Jahan, Z., Malik, U.S., Khan, M.D. and Vo, D.V.N. (2022). Nanotechnology-based controlled release of sustainable fertilizers. A review. *Environmental Chemistry Letters*, **20**(4), 2709–26. DOI: 10.1007/s10311-022-01409-w

Briassoulis, D. and Dejean, C. (2010). Critical review of norms and standards for biodegradable agricultural plastics part 1. Biodegradation in soil. *Journal of Polymers and the Environment*, **18**(3), 384–400. DOI: 10.1007/s10924-010-0168-1

Carmona, F.J., Guagliardi, A. and Masciocchi, N. (2022). Nanosized calcium phosphates as novel macronutrient nano-fertilizers. *Nanomaterials*, **12**(15), 2709. DOI: 10.3390/nano12152709

DeRosa, M.C., Monreal, C., Schnitzer, M., Walsh, R. and Sultan, Y. (2010). Nanotechnology in fertilizers. *Nature Nanotechnology*, **5**(2), 91. DOI: 10.1038/nnano.2010.2

Dewis, J. and Freitas, F. (1970). Physical and chemical methods of soil and water analysis. *FAO Soils Bulletin*, **n/a**(10), 275.

Dhlamini, B., Paumo, H.K., Kamdem, B.P., Katata-Seru, L. and Bahadur, I. (2022). Nano-engineering metal-based fertilizers using biopolymers: An innovative strategy for a more sustainable agriculture. *Journal of Environmental Chemical Engineering*, **10**(3), 107729. DOI: 10.1016/j.jece.2022.107729

DuBois, M., Gilles, K.A., Hamilton, J.K., Rebers, P.T. and Smith, F. (1956). Colorimetric method for determination of sugars and related substances. *Analytical Chemistry*, **28**(3), 350–6.

FAOSTAT (2021). *Food and Agricultural Organization of the United Nations*. Available at: https://www.fao.org/faostat/en/#data/QCL (accessed on 5/12/2023).

Fertahi, S., Bertrand, I., Ilsouk, M., Oukarroum, A., Zeroual, Y. and Barakat, A. (2020). New generation of controlled release phosphorus fertilizers based on biological macromolecules: Effect of formulation properties on phosphorus release. *International Journal of Biological Macromolecules*, **143**(n/a), 153–62. DOI: 10.1016/j.ijbiomac.2019.12.005

Figueiredo, P., Lintinen, K., Hirvonen, J.T., Kostiainen, M.A. and Santos, H.A. (2018). Properties and chemical modifications of lignin: Towards lignin-based nanomaterials for biomedical applications. *Progress in Materials Science*, **93**(51), 233–69. DOI: 10.1016/j.pmatsci.2017.12.001

Gee, G.W. and Bauder, J.W. (1986). Particle-size Analysis. In A. Klute (ed.) *Methods of Soil Analysis Part 1*. United State: Soil Science Society of America Book Series.

George, E., Rolf, S. and John, R. (2013). *Methods of Soil, Plant, and Water Analysis: A Manual for the West Asia and North Africa Region*. 3rd edition. Beirut, Lebanon: International Center for Agricultural Research in the Dry Areas (ICARDA).

Huang, C., Ragauskas, A.J., Wu, X., Huang, Y., Zhou, X., He, J. and Yong, Q. (2018). Co-production of bio-ethanol, xylonic acid and slow-release nitrogen fertilizer from low-cost straw pulping solid residue. *Bioresource Technology*, **250**(n/a), 365–73. DOI: 10.1016/j.biortech.2017.11.060

Kannan, S., Lemos, A.F. and Ferreira, J.M.F. (2006). Synthesis and mechanical performance of biological-like hydroxyapatites. *Chemistry of Materials*, **18**(8), 2181–6. DOI: 10.1021/cm052567q

Kottegoda, N., Munaweera, I., Madusanka, N. and Karunaratne, V. (2011). A green slow-release fertilizer composition based on urea-modified hydroxyapatite nanoparticles encapsulated wood. *Current science*, **101**(1), 73–8.

Kottegoda, N., Sandaruwan, C., Priyadarshana, G., Siriwardhana, A., Rathnayake, U.A., Berugoda Arachchige, D.M. and Amaratunga, G.A. (2017). Urea-hydroxyapatite nanohybrids for slow release of nitrogen. *ACS Nano*, **11**(2), 1214–21. DOI: 10.1021/acsnano.6b07781

Liu, J., Ye, X., Wang, H., Zhu, M., Wang, B. and Yan, H. (2003). The influence of pH and temperature on the morphology of hydroxyapatite synthesized by hydrothermal method. *Ceramics International*, **29**(6), 629–33. DOI: 10.1016/S0272-8842(02)00210-9

Liu, R. and Lal, R. (2015). Potentials of engineered nanoparticles as fertilizers for increasing agronomic productions. *Science of the Total Environment*, **514**(n/a), 131–9. DOI: 10.1016/j.scitotenv.2015.01.104

Madhurambal, G., Mariappan, M. and Mojumdar, S. (2010). Thermal, UV and FTIR spectral studies of urea–thiourea zinc chloride single crystal. *Journal of Thermal Analysis and Calorimetry*, **100**(3), 763–8. DOI: 10.1007/s10973-010-0758-0

Maria, P.A., Juan, C.S., Florin, I., Jose, L.G., I.V., Ioan, V.A., Henn, K., Gálos, B.

(2014). *Climate Change and Restoration of Degraded Land*. Madrid, Spain: Colegio de Ingenieros de Montes.

Moran, R. (1982). Formulae for determination of chlorophyllous pigments extracted with N, N-dimethylformamide. *Plant Physiology*, **69**(6), 1376–81. DOI: 10.1104/pp.69.6.1376

Okazaki, M., Hirata, I., Matsumoto, T. and Takahashi, J. (2005). Advantages of TOF-SIMS analysis of hydroxyapatite and fluorapatite in comparison with XRD, HR-TEM and FT-IR. *Dental Materials Journal*, **24**(4), 508–14. DOI: 10.4012/dmj.24.508

Pourzahedi, L., Pandorf, M., Ravikumar, D., Zimmerman, J.B., Seager, T.P., Theis, T.L. and Lowry, G.V. (2018). Life cycle considerations of nano-enabled agrochemicals: are today's tools up to the task?. *Environmental Science: Nano*, **5**(5), 1057–69. DOI: 10.1039/C7EN01166K

Sáez-Plaza, P., Michałowski, T., Navas, M.J., Asuero, A.G. and Wybraniec, S. (2013). An overview of the Kjeldahl method of nitrogen determination. Part I. Early history, chemistry of the procedure, and titrimetric finish. *Critical Reviews in Analytical Chemistry*, **43**(4), 178–223. DOI: 10.1080/10408347.2012.751786

Smoleń, S. and Sady, W. (2008). Effect of various nitrogen fertilisation and foliar nutrition regimes on carrot (Daucus carota L.) yield. *The Journal of Horticultural Science and Biotechnology*, **83**(4), 427–34. DOI: 10.1080/14620316.2008.11512402

Stewart, A.J., Chapman, W., Jenkins, G.I., Graham, I., Martin, T. and Crozier, A. (2001). The effect of nitrogen and phosphorus deficiency on flavonol accumulation in plant tissues. *Plant, Cell and Environment*, **24**(11), 1189–97. DOI: 10.1046/j.1365-3040.2001.00768.x

Tarafder, C., Daizy, M., Alam, M.M., Ali, M.R., Islam, M.J., Islam, R. and Khan, M.Z.H. (2020). Formulation of a hybrid nanofertilizer for slow and sustainable release of micronutrients. *ACS omega*, **5**(37), 23960–6. DOI: 10.1021/acsomega.0c03233

Vernon, L.S., Rudolf, O. and Rosa, M.L. (1999). Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. *Methods in Enzymology*, **299**(n/a), 152–78. DOI: 10.1016/S0076-6879(99)99017-1

Watkins, D., Nuruddin, M., Hosur, M., Tcherbi-Narteh, A. and Jeelani, S. (2015). Extraction and characterization of lignin from different biomass resources. *Journal of Materials Research and Technology*, **4**(1), 26–32. DOI: 10.1016/j.jmrt.2014.10.009

Xiao, X., Yu, L., Xie, F., Bao, X., Liu, H., Ji, Z. and Chen, L. (2017). One-step method to prepare starch-based superabsorbent polymer for slow release of fertilizer. *Chemical Engineering Journal*, **309**(n/a), 607–16. DOI: 10.1016/j.cej.2016.10.101

Ye, Y., Liang, X., Chen, Y., Liu, J., Gu, J., Guo, R. and Li, L. (2013). Alternate wetting and drying irrigation and controlled-release nitrogen fertilizer in late-season rice. Effects on dry matter accumulation, yield, water and nitrogen use. *Field Crops Research*, **144**(n/a), 212–24. DOI: 10.1016/j.fcr.2012.12.003

Yousaf, M., Bashir, S., Raza, H., Shah, A.N., Iqbal, J., Arif, M. and Hu, C. (2021). Role of nitrogen and magnesium for growth, yield and nutritional quality of radish. *Saudi Journal of Biological Sciences*, **28**(5), 3021–30. DOI: 10.1016/j.sjbs.2021.02.043

Zhang, S., Fu, X., Tong, Z., Liu, G., Meng, S., Yang, Y. and Li, Y.C. (2020). Lignin–clay nanohybrid biocomposite-based double-layer coating materials for controllable-release fertilizer. *ACS Sustainable Chemistry and Engineering*, **8**(51), 18957–65. DOI: 10.1021/acssuschemeng.0c06472

Zhang, S., Yang, Y., Gao, B., Wan, Y., Li, Y.C. and Zhao, C. (2016). Bio-based interpenetrating network polymer composites from locust sawdust as coating material for environmentally friendly controlled-release urea fertilizers. *Journal of Agricultural and Food Chemistry*, **64**(28), 5692–700. DOI: 10.1021/acs.jafc.6b01688

Zhiyuan, M., Xin, J., Jiamei, H., Zhiyong, L., Heyun, W. and Feng, Z. (2013). Mussel-Inspired thermosensitive polydopamine-graft-poly (N-isopropylacrylamide) Coating for controlled-release fertilizer. *Journal of Agricultural and Food Chemistry*, **61**(50), 12232–37. DOI: 10.1021/jf4038826