Nitrogen Fertilization by Encapsulating Zn-Al Layered Double Hydroxides in Alginate Beads

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Abstract: Layered double hydroxides (LDH) are anionic clays that have potential as slow-release fertilizers; however, their formulation as powders makes them difficult to apply, and their slow-release properties are impaired due to instability under acidic conditions. In the work reported, Zn-Al LDH containing interlayered $^{15}$NO$_3^-$ was synthesized for use as powder (LDH-N) or for encapsulation in alginate beads (LDH-AN), and then authenticated by X-ray diffraction, attenuated total reflectance-Fourier transform infrared spectroscopy, and elemental analyses. The two LDHs were compared to K$^{15}$NO$_3$ for evaluating their slow-release properties through (i) a kinetic study of NO$_3^-$ release in water under dynamic conditions, and (ii) a growth chamber experiment designed to estimate fertilizer N uptake efficiency (FNUE) by growing pearl millet (Pennisetum glaucum L.) on an acidic Oxisol in the absence of N losses. Both LDH materials exhibited slow-release properties in the kinetic studies, and NO$_3^-$ release was reduced for LDH-AN as compared to LDH-N. Because of these properties, FNUE measurements in the growth chamber experiment should have been lower with the LDHs than with K$^{15}$NO$_3$, but this was not the case for LDH-N, which was attributed to the structural instability of powdered LDH in the presence of soil acidity and to the exchange of NO$_3^-$ by more competitive anions such as CO$_3^{2-}$. A significant decrease in FNUE was observed for LDH-AN, demonstrating retention of slow-release behavior that most likely resulted from the presence of a physicochemical barrier having high cation-exchange and buffering capacities while limiting exposure to soil acidity and anion exchange. Alginate encapsulation expands the practical potential of LDH for slow-release NO$_3^-$ fertilization.

Keywords: layered double hydroxides; alginate; $^{15}$N; fertilizer nitrogen uptake efficiency; nitrate; slow-release fertilizer

1. Introduction

The growing demand for food and feed by a burgeoning world population leaves no alternative to modern cereal production systems with intensive use of synthetic N fertilizers. Most of these fertilizers supply NH$_4^+$, reflecting universal dependence on the Haber-Bosch process for NH$_3$ synthesis [1]. Unfortunately, there are negative consequences of applying NH$_4^+$, such as soil acidity generated by nitrification and the greater concern of gaseous N loss through NH$_3$ volatilization following application of the three major ammoniacal fertilizers: anhydrous NH$_3$, urea, and urea-NH$_4$NO$_3$ (UAN).
solutions [2–4]. A further limitation arises from the microbial preference for immobilization of NH$_4^+$ over NO$_3^-$ [5–8], which promotes heterotrophic oxidation of soil organic C with concurrent depletion of soil N [9,10] and has been implicated in lower fertilizer N uptake efficiency (FNUE) for NH$_4^+$ than NO$_3^-$ [11,12] when plant N uptake was not limited by leaching or denitrification. This difference would be favored by NO$_3^-$ storage in vacuoles [13,14], whereas NH$_4^+$ accumulation is toxic to most plant species [15].

Except for the use of NH$_4$NO$_3$ in UAN solutions and of CaNH$_4$(NO$_3$)$_2$ (CAN) as a major N source in some parts of the world, NO$_3^-$ fertilizers currently have a very limited presence in the commercial marketplace, due in part to their low N content that makes them more expensive and less practical than ammoniacal fertilizers. Owing to the inherent risk of N loss by leaching or denitrification, NO$_3^-$ applications must be synchronized with crop N demand, creating a logistical complication that becomes especially problematic for large farming operations.

A slow-release matrix could enhance the convenience of NO$_3^-$ fertilization, and has become feasible by utilizing Layered Double Hydroxides (LDHs) that have been employed in a wide variety of applications, including the removal of anionic pollutants from natural water and wastewater [16], and for the production of fertilizer materials supplying NO$_3^-$, phosphate, Zn, or borate [17–21].

Besides occurring naturally, LDHs can be synthesized by various techniques, the most common involving coprecipitation of di- and trivalent cations with continuous pH adjustment of an alkaline medium containing the desired anion in the absence of CO$_3^{2-}$ [22]. The product obtained has the empirical formula $[M^{2+}_{(1-x)}M^{3+}_{(x)}(OH)_2]A^{m-}(x/m)\cdot nH_2O$, where $M$ represents the two metallic cations differing in valence, $A$ the interlayer anion of valence $m$, and $x$ the mole fraction of the trivalent cation [i.e., $M^{3+}/(M^{2+} + M^{3+})$]. This fraction determines the permanent, positive charge created by isomorphic substitution of $M^{3+}$ for $M^{2+}$, generating anion-exchange capacity (AEC) that typically ranges from 200 to 300 cmol$_c$·kg$^{-1}$ [16]. Anion mobility is inversely related to charge density, and is reduced as a consequence of the physical protection afforded by the layered structures of LDHs [23].

When these materials are used as fertilizers, the rate of nutrient release is enhanced by soil acidity, which reduces their structural stability [24]. The liberated hydroxyl groups become a source of alkaline buffering that retards further loss of layer integrity, thereby retaining some of the original slow-release behavior that only disappears when the buffering capacity has been completely overcome by acidity [20]. The detrimental impact of soil acidity on the fertilizer value of LDHs is exacerbated by the prevalent practice of applying these materials as powders [18,20,24], which greatly increases the surface area for exposure to acidity and precludes accurate applications under field conditions.

Both difficulties can be addressed by incorporating LDHs within microspherical capsules that limit soil contact and can easily be applied. An ideal encapsulating agent is alginate, a natural polysaccharide obtained from brown marine algae that is economical, nonhazardous, and biodegradable. Hydrogel beads are readily formed by the dropwise introduction of an alginate sol to an aqueous solution containing Ca$^{2+}$ or another suitable polyvalent cation, and can be conveniently loaded with the material of interest such as clays, polymers, or nonionic fertilizers [25–27].

The present project was undertaken with the goal of developing a slow-release NO$_3^-$ fertilizer that would be suitable for a single pre-plant application. The work involved the synthesis and characterization of powdered Zn-Al LDH containing $^{15}$NO$_3^-$ with or without alginate encapsulation, and was followed by studies comparing the two LDH materials and K$^{15}$NO$_3$ with respect to (i) the rate of NO$_3^-$ release evaluated dynamically, and (ii) FNUE determined in a growth chamber experiment designed to rigorously test the slow-release behavior of these materials by application to an acidic Oxisol.

2. Materials and Methods

2.1. Synthesis of Zn-Al LDHs Interlayered with $^{15}$NO$_3^-$

The preparation of a Zn-Al LDH interlayered with NO$_3^-$ (LDH-N) was carried out by the constant pH coprecipitation method detailed by De Roy et al. [28]. In this method, 120 mL of an aqueous
solution 0.26 mol L\(^{-1}\) in Zn(NO\(_3\))\(_2\)·6H\(_2\)O and 0.13 mol L\(^{-1}\) in Al(NO\(_3\))\(_3\)·9H\(_2\)O was dispensed using a peristaltic pump for dropwise addition (≈0.3 mL min\(^{-1}\)) into a sealed reaction chamber containing 800 mL of a 0.13 mol L\(^{-1}\) solution of K\(^{15}\)NO\(_3\) (≈1.0 atom % \(^{15}\)N) that was stirred vigorously throughout the 6-h period of cation addition. To minimize the presence of CO\(_3^{2-}\) that would compete with NO\(_3^{-}\), both solutions were prepared using ultrapure water obtained from a Milli-Q® system (MilliporeSigma, Burlington, MA, USA) that had been previously boiled under an atmosphere of N\(_2\) for removal of CO\(_2\), and a similar atmosphere was maintained throughout LDH-N synthesis. Concurrent with cation addition, the solution pH was controlled between 8.75 and 9.00 by dispensing 1.0 mol L\(^{-1}\) KOH from a potentiometric titrator (Quimis Q799; Diadema, Brazil). Following synthesis, the contents of the reaction chamber were filtered under vacuum through JP41 filter paper (Quanty, Londrina, Brazil) in a 15-cm (dia.) Büchner funnel. The solid material was scraped from the filter paper into a Pyrex® petri dish (15-cm dia.), transferred to a desiccator for drying over silica gel, and crushed to pass through a 0.15 mm screen.

2.2. Synthesis of Zn-Al LDHs with \(^{15}\)NO\(_3^{-}\) in Alginate Beads

Ten g of LDH-N and 1 g of sodium alginate (SA) obtained from Sigma-Aldrich (St. Louis, MO, USA) were weighed into a 100-mL beaker, 50 mL of deionized water was added, and a sol was prepared by stirring magnetically until complete homogeneity was achieved in approximately 1 h. After transfer to a 50-mL burette, the gelatinized product was dispensed by dropwise addition into a 100-mL beaker containing 50 mL of 0.2 mol L\(^{-1}\) Ca(NO\(_3\))\(_2\)·6H\(_2\)O, forming alginate beads with LDH-N (LDH-AN). A 2 mm screen was used to separate the Ca(NO\(_3\))\(_2\) solution from the beads, which were then transferred to a 15-cm (dia.) Pyrex® petri dish for drying over silica gel in a desiccator to constant mass (Figure S1).

2.3. Characterization of Materials Studied

For X-ray powder diffraction (XRD) analysis to determine crystal structure, a slightly excessive amount of SA, KNO\(_3\), LDH-N, or LDH-AN was transferred to the cavity in a sample holder, and a microscope slide was subsequently used to compress and level the sample, such that powder filled the entire cavity and was flush with the top surface of the sample holder. The XRD analysis was accomplished by using a Shimadzu XRD-6000 instrument (Shimadzu Scientific Instruments, Inc., Kyoto, Japan) equipped with a Cu cathode and a graphite monochromator producing X-ray radiation at 1.54 Å, 30 kV, and 30 mA, which was operated to provide a scanning range of 4–70° in 0.01° increments with 10 s per increment.

Attenuated total reflectance Fourier transform infrared spectroscopy (ATR-FTIR) was used to compare NO\(_3^{-}\) orientation in the four materials studied. Spectra were recorded with a Jasco FTIR 4100 spectrophotometer (Jasco Deutschland GmbH, Pfungstadt, Germany), configured to obtain 256 scans over a wavelength range of 4000 to 500 cm\(^{-1}\).

Elemental analyses of LDH-N and LDH-AN for C, H, and N were performed with a PerkinElmer 2400 Series II CHNS/O Analyzer (PerkinElmer, Waltham, MA, USA). These materials were also analyzed for Zn and Al with the use of an Agilent 240FS atomic absorption spectrometer (Agilent Technologies, Inc., Santa Clara, CA, USA), following complete dissolution by digestion with 6 mol L\(^{-1}\) HCl at 100 °C. Direct combustion mass spectrometry was employed for \(^{15}\)N analysis of the two LDH materials, and also K\(^{15}\)NO\(_3\), using an ANCA-GSL instrument (Sercon, Crewe, UK). Table 1 shows elemental analyses for the four materials studied, along with \(^{15}\)N analyses for KNO\(_3\), LDH-N, and LDH-AN.

2.4. Rate of NO\(_3^{-}\) Release from Zn-Al LDHs and KNO\(_3\)

To compare the kinetics of NO\(_3^{-}\) release from LDH-N, LDH-AN, and KNO\(_3\), a stirred-flow study was conducted by utilizing a peristaltic pump (Dionex UltiMate 3000; Thermo Fisher Scientific, Waltham, MA, USA) to supply deionized water for continuous flow (1 mL min\(^{-1}\)) through a 15 mL reactor that was stirred magnetically (500 rev min\(^{-1}\)) and connected for discharge to microcentrifuge tubes in a fraction collector. In the procedure used, water flow was initiated after introducing the
fertilizer material into the reactor and positioning a cellulose filter (0.45 µm) to retain particulates, and the outlet flow was delivered for automated collection of 40 samples in 80 min, prior to colorimetric NO$_3^-$ analyses by the method of Cataldo et al. [30].

Table 1. Chemical properties of sodium alginate (SA) Zn-Al layered double hydroxides with interlayered NO$_3^-$ (LDH-N) in the form of powder, or Zn-Al layered double hydroxides with interlayered NO$_3^-$ and encapsulated in alginate beads (LDH-AN). Data reported as a mean of triplicate determinations. O: determined by difference; KNO$_3$ and SA: as reported by Fisher Scientific [29]; na: natural abundance (0.366 atom% $^{15}$N).

| Material      | Elemental Analysis (g kg$^{-1}$) | Atom% $^{15}$N |
|---------------|----------------------------------|----------------|
|               | C  | H  | O  | N  | Zn | Al |
| KNO$_3$       | 0  | 0  | 475| 139| 0  | 0  | 1.844 |
| SA            | 333| 42 | 518| 0  | 0  | 0  | na    |
| LDH-N         | 4  | 30 | 499| 45 | 282| 138| 1.064 |
| LDH-AN        | 51 | 34 | 458| 48 | 277| 132| 1.022 |

2.5. Growth Chamber Experiment

2.5.1. Soil Studied

A Brazilian Oxisol (Typic Hapludox [31]), collected at a depth of 20 cm from a pasture site in the vicinity of Viçosa, Minas Gerais, was air-dried and then pulverized to <2 mm using a mortar and pestle. Table 2 shows physicochemical properties determined as detailed by Nunes et al. [32].

Table 2. Physicochemical properties of the Brazilian Oxisol used in the growth chamber experiment.

| Property                           | Value |
|------------------------------------|-------|
| pH (soil:water, 1:1)              | 5.6   |
| Organic C (g kg$^{-1}$)            | 18.4  |
| Total N (g kg$^{-1}$)              | 1.26  |
| Available P (mg kg$^{-1}$)         | 3.8   |
| Exchangeable K (mg kg$^{-1}$)      | 39    |
| Exchangeable Ca (cmol$_c$ kg$^{-1}$)| 1.46  |
| Exchangeable Mg (cmol$_c$ kg$^{-1}$)| 0.57  |
| Potential acidity (cmol$_c$ kg$^{-1}$) | 7.5   |
| Sand (%)                           | 34    |
| Silt (%)                           | 12    |
| Clay (%)                           | 54    |
| Water-holding capacity (%)         | 32    |

2.5.2. Procedure

Seeds of pearl millet (*Pennisetum glaucum* L., cultivar BR1501) were placed on sterilized paper towels moistened with deionized water, beginning a 2-day period for germination at 30 °C. After 1 day, a suitable potting medium was prepared by homogenizing 150 g of soil with 100 g of nutrient-free sand (>0.5 mm) in a polyethylene bag, and the mixture was then transferred to each of the 40 350-mL (12-oz) polypropylene storage containers that had being painted to exclude light (Figure S2). The three $^{15}$N sources (LDH-N, LDH-AN, and KNO$_3$) were applied at rates of 50, 150, and 300 mg N kg$^{-1}$, leaving four pots to serve as unlabeled controls in a completely randomized design with four replicates. Using a spatula, the fertilizers were spread uniformly across the pots and incorporated to a depth of 0.5–1 cm, and sufficient deionized water was then added to each pot so the medium would be at 70% water-holding capacity and the pots were weighed to allow subsequent watering. The following day, eight healthy seedlings of uniform size were transplanted to be evenly spaced in each pot, and the pots were transferred to a growth chamber providing 12 h of light and dark periods at a constant
temperature of 25 °C. Thinning was done 5 days after transplanting, so as to obtain a final stand of six plants per pot, and 10 mL of a nutrient solution, mostly following Novais et al. [33], was applied to supply N (10 mg kg\(^{-1}\)), P (215 mg kg\(^{-1}\)), K (200 mg kg\(^{-1}\)), Mg (24 mg kg\(^{-1}\)), S (40 mg kg\(^{-1}\)), Zn (4 mg kg\(^{-1}\)), Cu (1.3 mg kg\(^{-1}\)), Fe (1.5 mg kg\(^{-1}\)), B (0.8 mg kg\(^{-1}\)), Mn (3.6 mg kg\(^{-1}\)), and Mo (0.1 mg kg\(^{-1}\)).

The plants were harvested 25 days after transplanting (Figure S3), by cutting the stem to within 0.5 cm of the soil surface using scissors that were cleaned with ethanol between samples to prevent isotopic cross-contamination. After inverting the pot to collect the medium by hand, the roots were separated by gentle washing with deionized water. Shoot and root samples were collected in paper bags, followed by drying for at least 72 h in a forced-air oven at 65 °C. The samples were subsequently weighed for dry matter determination, and then segmented to <3 mm using scissors that were cleaned between samples as previously described. After samples were thoroughly mixed, a subsample of each was transferred to fill approximately half the volume of a 2-mL microcentrifuge tube, a 3-mm stainless steel ball was dropped into the tube, and the tube was sealed for grinding to <150 µm using a Retsch MM 400 mixer mill (Retsch GmbH, Haan, Germany). Direct combustion mass spectrometry was employed for total N and \(^{15}\)N analyses.

2.6. Data Analyses

Data generated by XRD and ATR-FTIR were plotted using Origin version 7.0 software (OriginLab Corp., Northampton, MA, USA). Identification of diffraction peaks was carried out by a comparison with the hydrotalcite diffractogram from the diffraction equipment database [34] and with a series of Zn-Al LDHs described in the literature [35,36]. Interlayer distances were calculated from 2θ values by using Bragg’s equation, \(n\lambda = 2d_{hkl}\sin\theta\), where \(n\) is the diffraction order, \(\lambda\) is the wavelength of the X-ray, \(d_{hkl}\) is the interlayer distance for the hkl peak, and \(\theta\) is Bragg’s angle as determined by the diffraction peak. ATR-FTIR spectra were analyzed by comparison with LDH-N described in the literature [37,38].

From dry matter, total N, and isotopic data collected in the growth chamber experiment, the percentage of FNUE was calculated for shoots and roots as \(\frac{N_t}{N_f} \times \frac{(SR - B)}{(F - B)} \times 100\), where \(N_t\) (mg pot\(^{-1}\)) is the total N determined for the plant sample analyzed, \(N_f\) (mg pot\(^{-1}\)) is the amount of fertilizer \(^{15}\)N applied, and the remaining variables represent isotopic enrichments (atom %, \(^{15}\)N) measured for the shoot or root sample analyzed (SR), the fertilizer N applied (F), and the corresponding plant sample (shoot or root) collected from the unlabeled control pots (B).

Microsoft® Excel was used to generate descriptive statistics from replicate data collected in the growth chamber study. Past version 3.22 [39] was utilized for analysis of variance (ANOVA) and for mean comparisons involving Tukey’s procedure (\(p < 0.05\) to 0.001).

3. Results and Discussion

3.1. Characterization

X-ray diffractograms are presented in Figure 1 for KNO\(_3\), alginate, LDH-N, and LDH-AN. The XRD pattern in Figure 1a exhibits well-defined peaks typical of a pure salt with high crystallinity. In contrast, Figure 1b shows a single weak broad diffraction peak at 13.32° but nothing else, indicating the same amorphous character documented by diffractograms previously obtained for alginate [40,41]. The X-ray diffraction pattern of the pristine LDH (Figure 1c) exhibits the presence of basal peaks (00\(n\) Reflex)[(003), (006)] related to the stacked layers characteristic of the LDH structure. The basal spacing distance (\(d\)) calculated from the position of the (003) and (006) peaks was 7.54 Å, coinciding with the value observed for NO\(_3\)\(^{-}\) intercalation in Zn-Al LDHs [35,36]. The residual presence of KNO\(_3\) is indicated by the four peaks labeled as (*), which coincide with the corresponding peaks obtained for KNO\(_3\) (Figure 1a) but are absent from the diffractogram obtained for LDH-AN (Figure 1d). In all other respects Figure 1c,d show the same pattern, verifying that pristine LDH-N was successfully incorporated into alginate beads.
Figure 1. Diffractograms of (a) KNO3, (b) alginate, (c) Zn-Al layered double hydroxides with interlayered NO3− (LDH-N) in the form of powder, and (d) Zn-Al layered double hydroxides with interlayered NO3− and encapsulated in alginate beads (LDH-AN). (*) KNO3 phase.

Figure 2 shows the functional group composition of the four materials studied when characterized by ATR-FTIR spectroscopy. The two bands noted in the spectrum obtained for KNO3 (Figure 2a) are attributable to the vibrational modes of NO3− at 1350 (n3) and 820 cm−1 (n2) [42,43]. In the alginate spectrum (Figure 2b), the presence of the broad band in the region between 3000 and 3700 cm−1 is associated with OH stretching vibration; the bands at 1600 and 1420 cm−1 are due to symmetric and asymmetric stretching vibrations of carboxylate (COO−) groups; and the band at 1030 cm−1 corresponds to the stretching vibration of C-O-C groups [44]. For LDH-N (Figure 2c), the spectrum shows a very broad band in the region between 3600 and 3000 cm−1 assigned to OH stretching of the H2O molecules and hydroxyl groups present in the inorganic layers. The band related to the bending mode of water molecules is observed at 1640 cm−1, while the bands at 1350 and 820 cm−1 confirm the presence of interlayered NO3− and the band at 560 cm−1 is assigned to the Al/Zn-OH translation mode of LDH layers [45,46]. The spectrum of LDH-AN presented in Figure 2d shows bands related to the presence of both alginate and LDH-N as evidenced by bands at 1600, 1420, and 1030 cm−1 due to vibration stretching of alginate functional groups, and by those at 1350 and 820 cm−1 related to modes of vibration of interlayered NO3− present in LDH.

3.2. Kinetic Study

Based on previous work demonstrating slow-release properties for LDHs [17,19,21,47], the same behavior was expected in the present study. Confirmation is provided by Figure 3, which compares KNO3, LDH-N, and LDH-AN in terms of NO3− release in a continuous stirred flow system using deionized water as the liquid phase. The results show a very different pattern for the two LDH materials than for KNO3, in that NO3− release occurred much more gradually and was far from complete. In both respects, the decrease was larger for LDH-AN than for LDH-N, as would be expected from the much lower surface area of alginate spheres as compared to powdered LDH.
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3.3. Growth Chamber Experiment

To more realistically evaluate the fertilizer potential of the two Zn-Al LDH materials under acidic conditions that would differentiate their slow-release properties, a growth chamber experiment was conducted that compared FNUE when 15NO$_3^-$ was supplied to a tropical Oxisol by applying 50–300 mg N kg$^{-1}$ as KNO$_3$, LDH-N, or LDH-AN (Figure S2). The efficiency of 15N uptake was enhanced by (i) densely planting a forage crop with high N demand (pearl millet) in a soil-sand mixture with very limited N supply, (ii) using pots without drainage holes so that leaching losses were prevented, and (iii) taking care to ensure that soil moisture content never exceeded 70% WHC, thereby controlling denitrification. In keeping with soils typical of the tropics, the Oxisol studied was used without previous pH adjustment, and soil acidity would have been exacerbated by the high planting density (6 plants per 100 cm$^2$).

Figure 4 compares dry matter production with and without the three N sources studied. Regardless of the N rate applied, dry matter was significantly lower with LDH-AN than with LDH-N or KNO$_3$ (Figure S4), which is consistent with the slow-release pattern documented by Figure 3 that was more pronounced for LDH-AN than for LDH-N.
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![Figure 4](image_url)  
**Figure 4.** Dry matter determined for pearl millet (*Pennisetum glaucum* L.) grown 25 days after transplanting in a growth chamber study with an Oxisol involving application of 50, 150, or 300 mg $^{15}$N kg$^{-1}$ as KNO$_3$, Zn-Al layered double hydroxides with intercalated NO$_3^-$ (LDH-N) in the form of powder, or Zn-Al layered double hydroxides with interlayered NO$_3^-$ and encapsulated in alginate beads (LDH-AN). Data shown for shoots plus roots as a mean from four replicate pots, with standard error bars. At each N rate, treatments having the same lowercase letter do not differ significantly ($p < 0.05$) by Tukey’s test.

The FNUE calculated from N and $^{15}$N data obtained for shoots plus roots are reported by Figure 5. Regardless of the N rate applied, FNUE was significantly ($p < 0.001$) lower for LDH-AN than for KNO$_3$ or LDH-N. The data in Figure 5 show that, when applied to an acidic soil, LDH-AN was highly effective for slowing the release of NO$_3^-$, whereas LDH-N was not effective.
physicochemical protection, arising not only from a limitation in LDH contact with soil, but also powder as aginate beads, since recent work by Everaert et al. [19] showed that P uptake efficiency was intensified by the broadcast application that was made with subsequent incorporation to a dense stand of pearl millet. The effect of soil acidity on degrading the powdered LDH-N would have been intensified by the broadcast application that was made with subsequent incorporation to maximize soil exposure of this material. This exposure would have promoted the exchange of NO$_3^-$ by anion exchange. Owing to the presence of structural hydroxyl groups, LDHs are inherently reactive toward H$_3$O$^+$ ions and are solubilized in the proximity of an alkalized LDH, owing to root and microbial respiration promoted by the densely planted stand of pearl millet. The effect of soil acidity on degrading the powdered LDH-N would have been intensified by the broadcast application that was made with subsequent incorporation to maximize soil exposure of this material. This exposure would have promoted the exchange of NO$_3^-$ by other anions that are much more competitive for adsorption due to higher valence and charge density [16,49]. The most effective competitor would have been CO$_3^{2-}$, which is retained more tightly than other monovalent or divalent anions [16,23] and would have been formed in abundance in the proximity of an alkalized LDH, owing to root and microbial respiration promoted by the densely planted stand of millet.

The complete absence of slow-release behavior with LDH-N can be attributed to (i) soil acidity that would have degraded the Zn-Al LDH structure, (ii) the surface area associated with a broadcast application of powder, and (iii) weak NO$_3^-$ retention by anion exchange. Owing to the presence of structural hydroxyl groups, LDHs are inherently reactive toward H$_3$O$^+$ ions and are solubilized in the presence of acidity that exceeds their alkalizing capacity [16,20,24,48]. In the present study, excess acidity would have existed when LDH-N was applied at a rate of 50 or 150 mg N kg$^{-1}$, but probably not with 300 mg N kg$^{-1}$ unless there was substantial acidification during the uptake of basic cations by a dense stand of pearl millet. The fertilizer materials were each applied at three different N rates to ascertain whether a higher rate would enhance the slow-release properties of LDH materials by prolonging their presence, unless there was substantial acidification during the uptake of basic cations by a dense stand of pearl millet. The fertilizer materials were each applied at three different N rates to ascertain whether a higher rate would enhance the slow-release properties of LDH materials by prolonging their presence, unless there was substantial acidification during the uptake of basic cations by a dense stand of pearl millet. The fertilizer materials were each applied at three different N rates to ascertain whether a higher rate would enhance the slow-release properties of LDH materials by prolonging their presence,
which would decrease FNUE. Such a decrease was indeed observed with either LDH-N or LDH-AN (Figure 5), but also occurred with KNO$_3$ and thus is not indicative of slower N release. The most plausible explanation for this decline is the inverse relationship that normally exists between fertilizer N rate and FNUE.

4. Summary and Conclusions

For potential use as a slow-release NO$_3^-$ fertilizer, an anionic clay was prepared by synthesizing Zn-Al LDH containing interlayered $^{15}$NO$_3^-$ and formulated as either powder (LDH-N) or alginate beads (LDH-AN). The latter formulation was superior to the former when their slow-release properties were evaluated relative to K$^{15}$NO$_3$ in a kinetic experiment using water as the liquid phase. Moreover, LDH-AN was effective for minimizing the loss of these properties in a growth chamber study with an acidic Oxisol, as documented by significantly lower FNUE than for LDH-N or K$^{15}$NO$_3$. Alginate encapsulation expands the practical potential of LDH for slow-release NO$_3^-$ fertilization.

Supplementary Materials: The following are available online at http://www.mdpi.com/2504-3129/1/2/11/s1, Figure S1: Zn-Al-$^{15}$NO$_3$ layered double hydroxides encapsulated in alginate beads (LDH-AN), Figures S2–S4: growth chamber experiment with pearl millet (Pennisetum glaucum L).

Author Contributions: J.T., F.G.P. and V.L.N.N. synthesized and characterized the LDHs utilized; R.B.C. and V.L.N.N. performed the stirred-flow and greenhouse experiments; V.L.N.N. and J.T. analyzed the data; R.L.M. and V.L.N.N. wrote the paper. All authors have read and agreed to the published version of the manuscript.

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