



CIRCULAR ECONOMY

**CiNURGi**

# Circular Nutrients for a Sustainable Baltic Sea Region (CiNURGi)

**Piloting the evaluation standards for quality control and agronomic value for recycled nutrients**

**April 2026**

Oksana Valetka, Athanasios Pantelopoulos, Helena Aronsson, Liina Edesi, Kalvi Tamm, Grete Jõgisoo, Ksawery Kuligowski, Tapio Salo, Mayka Schmitt Rahner and Katrin Kuka



**This #MadeWithInterreg project helps drive the transition to a green and resilient Baltic Sea region and is part of the EU-funded Interreg Baltic Sea Region (BSR) core project #C049, titled CiNURGi, under the 2021-2027 PROGRAMME, Priority 3: Climate-Neutral Societies, Objective 3.1: Circular Economy.**

**Organisations from the following countries cooperate together to make that happen: Sweden (LP), Denmark, Estonia, Finland, Germany, Latvia, Lithuania and Poland.**

**Project homepage: <https://interreg-baltic.eu/project/cinurgi>**

**Project LinkedIn page: <https://www.linkedin.com/showcase/cinurgi>**

**Piloting the evaluation standards for quality control and agronomic value for recycled**  
Oksana Valetska<sup>1</sup>, Athanasios Pantelopoulos<sup>1</sup>, Helena Aronsson<sup>1</sup>, Liina Edesi<sup>2</sup>, Kalvi Tamm<sup>2</sup>,  
Grete Jõgisoo<sup>2</sup>, Ksawery Kuligowski<sup>3</sup>, Tapio Salo<sup>4</sup>, Mayka Schmitt Rahner<sup>5</sup> and Katrin Kuka<sup>5</sup>

<sup>1</sup> Swedish University of Agricultural Science, Lennart Hjelm's väg 9, Uppsala, Sweden

<sup>2</sup> Centre of Estonian Rural Research and Knowledge (METK), J. Aamisepa 1, Jõgeva Township, 48309, Estonia

<sup>3</sup> Institute of Fluid-Flow Machinery Polish Academy of Sciences, Fiszerka 14 St., 80-231 Gdańsk, Poland

<sup>4</sup> Natural Resources Institute Finland (Luke), Tietotie 4, 31600 Jokioinen, Finland

<sup>5</sup> Julius Kühn-Institut (JKI), Institute for Crop and Soil Science, Bundesallee 58, 38116 Braunschweig, Germany

\*Corresponding author: Oksana Valetska

This report can be cited as follows:

Valetska, O., Pantelopoulos, A., Aronsson, H., Edesi, L., Tamm, K., Jõgisoo, G., Kuligowski, K., Salo, T., Schmitt Rahner, M., Kuka, K. 2026. Piloting the evaluation standards for quality control and agronomic value for recycled nutrients. Report from CiNURGi project, Interreg BSR #C049. DOI 10.5281/zenodo.20507855.

## Foreword

*CiNURGi (Circular Nutrients for a Sustainable Baltic Sea Region) is an Interreg BSR Core Project dedicated to advancing circular economy for nutrients within the Baltic Sea Region. By enhancing infrastructure, technology, and policy the project seeks to improve nutrient recovery from biomass and resource streams originating from agricultural, municipal, and industrial sources. This endeavor aligns with several regional and European strategies, including the HELCOM Baltic Sea Regional Nutrient Recycling Strategy, the EU's Circular Economy Action Plan under the Green Deal, and the Integrated Nutrient Management Action Plan of the Farm to Fork Strategy. The CiNURGi is ongoing from November 2023 to October 2026.*

*This report pertains to Task A2.2, focusing on piloting evaluation standards for assessing the quality, nutrient release dynamics, and agronomic value of recycled nutrient fertilizers (RNFs) across multiple evaluation centres in the Baltic Sea Region. The findings and activities detailed herein contribute directly to CiNURGi's overarching goals by providing the first coordinated validation of RNF performance, generating comparable data on nitrogen and phosphorus availability and plant response, and establishing the methodological foundation for guideline industry standards that support the wider adoption, quality assurance, and market integration of RNFs as substitutes for mineral fertilizers.*

*We acknowledge the collaborative efforts of our consortium, comprising 24 partners and 13 associated organizations from Denmark, Estonia, Finland, Germany, Poland, Latvia, Lithuania, and Sweden. Their dedication and expertise are instrumental in driving the project's success.*

*For more information about CiNURGi and its initiatives, please visit our project homepage <https://interreg-baltic.eu/project/cinurgi/>*

*April 2026*

*Erik Sindhøj & Cheryl Cordeiro, CiNURGi Project Coordinators*

*RISE – Research Institutes of Sweden*

## Contents

List of abbreviations .....	1
1. Introduction.....	1
Background.....	1
Aim of activity 2.2.....	2
Evaluation centres – contributing partners.....	2
Relation to other project activities.....	3
2. Lexicon.....	3
2.1 Plant nutrition .....	3
2.2 Agronomic terms.....	4
2.3 Technologies for producing recycled nutrient fertilisers. ....	5
2.4 Methods for the characterization of RNFs .....	6
Dry matter and volatile solids .....	6
Total nitrogen .....	7
Ammonium nitrogen .....	7
Nitrate nitrogen.....	8
P, K, Ca, Mg and trace elements.....	8
pH .....	9
3. Protocols for the assessment of the agronomic value of RNFs.....	9
3.1 Soil sampling, preparation and analysis .....	10
3.2 Common protocol for assessing RNFs C mineralization and N-P dynamics in soil incubations .....	11
3.2.1 Characterization of RNFs .....	11
3.2.2 General approach of incubations .....	12
3.3 Common protocol for determining the P fertilizing value of RNFs in greenhouse experiments .....	13
4. Evaluation of RNFs.....	14
4.1 Incubation studies .....	14
4.1.1 Evaluation centre in Sweden (SLU) .....	14
4.1.2 Evaluation centre in Finland (LUKE) .....	19
4.2 Pot trials .....	24
4.2.1 Evaluation centre in Sweden (SLU) .....	24
4.2.2 Evaluation centre in Sweden (Linnæus University).....	27
4.2.3 Evaluation centre in Finland (LUKE) .....	34
4.2.4 Evaluation centre in Estonia (METK) .....	37
4.2.5 Evaluation centre in Poland (IMP).....	44
4.2.6 Evaluation centre in Germany (JKI) .....	56
5. General conclusions and recommendations.....	60
6. Dissemination.....	62
References.....	64
Appendix.....	66

## Executive Summary

This report presents the outcomes of Activity A2.2 of the CINURGi project, which piloted harmonised evaluation standards for assessing the quality, nutrient release dynamics, and agronomic value of recycled nutrient fertilisers (RNFs) across the Baltic Sea Region (BSR). The activity established six evaluation centres in Sweden, Finland, Estonia, Poland, and Germany, each representing distinct pedoclimatic conditions and national nutrient recycling contexts. Together, these centres tested a wide range of established and emerging RNFs, including digestates, pelletised organic fertilisers, biochars, struvite, urine-based granules, and multi-waste pellets using coordinated incubation studies and greenhouse pot trials.

A central objective of A2.2 was to generate comparable, cross-country data on nitrogen (N) and phosphorus (P) release, carbon mineralisation, and plant nutrient uptake, thereby supporting the development of guideline industry standards under Objective O2.2. To achieve this, partners adopted common protocols for soil preparation, RNF characterisation, incubation procedures, and pot trial design, while allowing limited flexibility to accommodate national practices and stakeholder needs.

Across all centres, results highlighted the importance of matching RNF type to soil properties, crop nutrient demand, and the timing of nutrient release. Slow-release RNFs such as struvite and biochar-based products performed well in systems requiring sustained nutrient availability, while digestates and urine-based granules provided rapid N supply. Soil pH, texture, and compaction strongly influenced nutrient uptake, particularly for phosphorus.

The activity also fulfilled its mandate to engage target groups through cross-sectoral, bottom-up dissemination. Evaluation centres communicated results through LinkedIn updates, project website publications, scientific conferences, field demonstrations, stakeholder workshops, and integration into university teaching. These interactions created feedback loops with farmers, advisors, technology providers, and policymakers, strengthening the relevance and applicability of the emerging evaluation standards.

Overall, the findings of A2.2 demonstrate that RNFs produced from diverse organic waste streams can provide agronomic value comparable to mineral fertilisers, while contributing to circular nutrient management and reduced environmental pressure in the BSR. The harmonised protocols and multi-country dataset generated through this activity form a critical foundation for the guideline industry standards to be developed under O2.2, supporting the safe, effective, and scalable use of recycled nutrient fertilisers across the region.

**Keywords:** recycled nutrient fertilisers, nutrient release dynamics, agronomic value, incubation studies, pot trials, nitrogen availability, phosphorus availability, evaluation standards, mineral fertiliser equivalent, Baltic Sea Region

## List of abbreviations

ANR	Apparent Nitrogen Recovery
APR	Apparent Phosphorus Recovery
BSR	Baltic Sea Region
CH <sub>4</sub>	Methane
CiNURGi	Circular Nutrients for a Sustainable Baltic Sea Region project
CO	Carbon Oxide
CO <sub>2</sub>	Carbon Dioxide
DM	Dry Matter
EC	European Commission
EM	Effective Microorganisms
EU	European Union
GHG	Greenhouse Gases
MBM	Meat and Bone Meal
MFE	Mineral fertiliser equivalent
N	Nitrogen
N <sub>tot</sub>	Total Nitrogen
N <sub>inorg</sub>	Inorganic Nitrogen
N <sub>org</sub>	Organic Nitrogen
N <sub>m</sub>	Organic Nitrogen Mineralisation
N <sub>rel</sub>	Release of Total Nitrogen from RNFs
N <sub>2</sub> O	Dinitrogen Oxide (Nitrous Oxide)
NH <sub>4</sub> -N	Ammonium Nitrogen
NH <sub>3</sub> -N	Nitrate Nitrogen
NU	Nitrogen Uptake
NUE	Nitrogen Use Efficiency
P	Phosphorus
P-AL	Plant-available phosphorus
PU	Phosphorus uptake
RISE	Research Institutes of Sweden
RNFs	Recycled nutrient fertilisers
TSP	Triple Super Phosphate
TS	Total Solids
TNK	Total Kjeldahl nitrogen
WEP	Water-extractable Phosphorus
WHC	Water holding capacity

# 1. Introduction

## Background

After the first half of the 20<sup>th</sup> century, the agricultural sector achieved remarkable increases in crop production, largely due to the extensive use of mineral fertilisers. However, nitrogen (N) production remains highly energy-intensive, while phosphorus (P) sources are non-renewable and their long-term availability uncertain, placing P on the EU list of Critical Raw Materials. Currently, the EU applies approximately 8.9 Mt of N and 0.9 Mt of P across 157 million hectares of land, with projections indicating a 3% and 20% increase in N and P consumption, respectively, by 2034 (Eurostat, 2024; Fertilisers Europe, 2024). Between 2019 and 2022, mean imported quantities accounted for 35% of N and 59% of P use, highlighting the EU's dependence on third countries and vulnerability to trade disruptions.

The shift from traditional mixed farming systems, where crop and livestock production coexisted, to highly specialized systems has created mostly linear nutrient flows. In modern agriculture, products not intended for human consumption or trade are often treated as waste. Each year in the EU, 17.6 Tg of N and 1.84 Tg of P enter the agricultural sector, yet only about 20% of N and 30% of P inputs reach human diets. This reveals both significant nutrient losses and suboptimal recycling within the food system (Buckwell & Nadeu, 2016; Van Dijk et al., 2016). As a result, areas with dense livestock production or large urban populations often face nutrient surpluses in the form of “organic waste,” becoming hot spots of environmental risk.

Until recently, residual organic streams were managed mainly with low-cost, inefficient solutions that contributed to environmental degradation through nutrient emissions and underutilization of their organic matter and nutrient content. The Baltic Sea is a well-documented example of such deterioration, where agricultural nutrient inputs have led to eutrophication. Excessive mineral fertiliser uses in the region, combined with the unbalanced N:P ratio of animal manures, has contributed to phosphorus accumulation, recurrent cyanobacteria blooms, and seasonal anoxia. By contrast, effective treatment of organic nutrient resources could redistribute surplus nutrients to remote areas, reduce environmental pressures, and support a more balanced nutrient cycle.

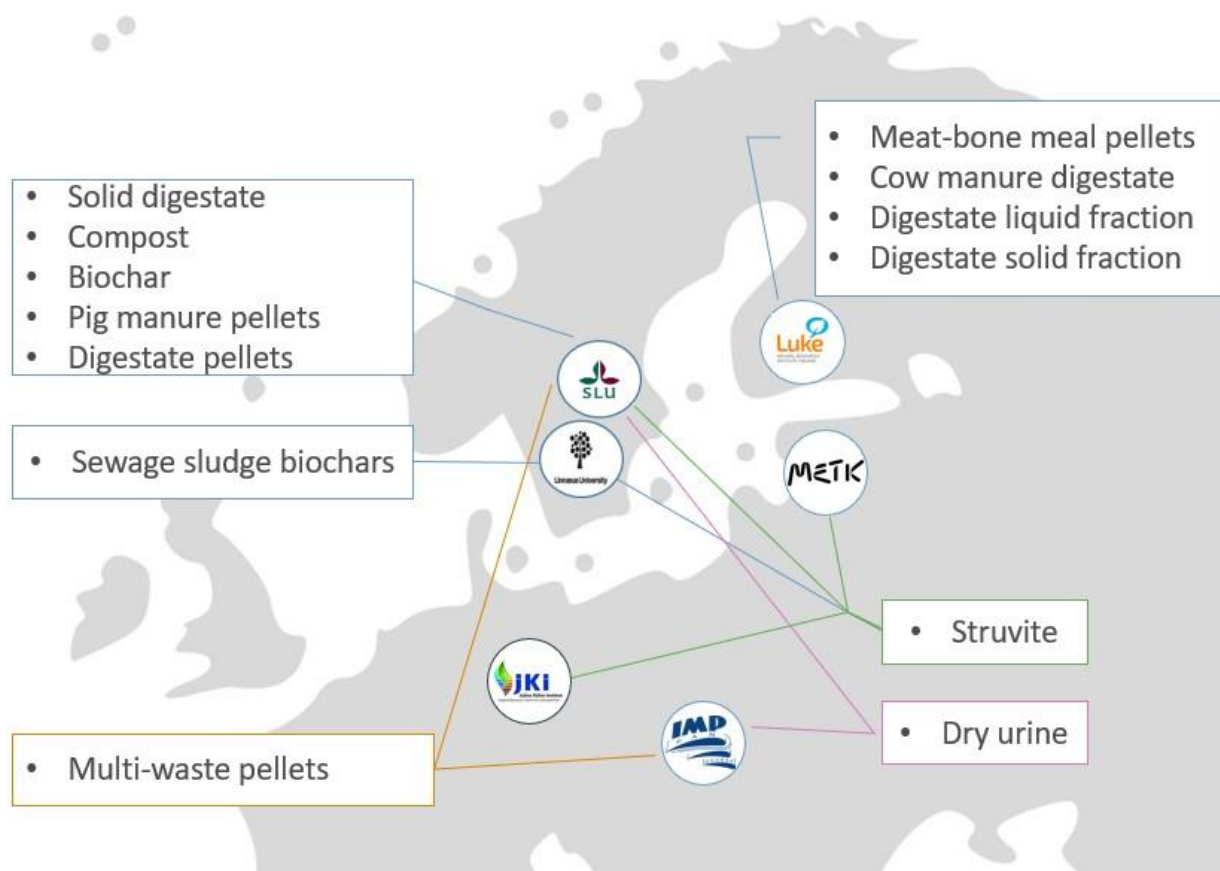
Closing nutrient loops in agriculture aligns with EU priorities under the Circular Economy framework, as well as the Green Deal and Farm-to-Fork strategies. Increasing nutrient recovery and use efficiency from organic waste streams can reduce the sector's dependence on synthetic fertilisers and mitigate environmental impacts from food production. The Interreg-funded CiNURGi project supports this transition by demonstrating sustainable solutions for managing residual organic waste streams in the BSR. By enhancing nutrient recovery from underutilized or unexploited organic resources, the project aims to improve nutrient recycling within agroecosystems, lower agriculture-induced environmental pressures, and contribute to EU circular economy objectives. Importantly, the processing of organic waste into recycled nutrient fertilisers (RNFs) alters the nutrient content and quality compared to the original feedstock, influencing their performance as potential substitutes of mineral fertilisers. Predicting the effects of RNFs on soil nutrient availability is complex and depends on both RNF type and soil characteristics.

## Aim of activity 2.2

The overall aim of the activity was to characterize and assess the agronomic value of established and emerging RNFs to support knowledge-based production and application systems. Specifically, the project aimed to establish at least four evaluation centres that would reflect the variability of pedoclimatic conditions around the Baltic Sea, the differences in the availability of organic nutrient streams, and the country-specific approaches to evaluating the agronomic value of RNFs. Moreover, common approaches and methods were adopted for the characterization of RNFs and their agronomic testing, enabling meaningful comparisons between different RNFs and more comprehensive extrapolation of results across countries.

## Evaluation centres – contributing partners

Within the project, six evaluation centres were established focusing on the assessment of the agronomic value of different RNFs in five different countries. The evaluation of the RNFs was performed at the Centre of Estonian Rural Research and Knowledge (METK), Institute of Fluid-Flow Machinery (IMP-PAN), Julius Kühn Institute (JKI), Natural Resource Institute of Finland (Luke), Linnaeus University (LnU) and Swedish University of Agricultural Sciences (SLU). The geographic distribution of the evaluation centres and the respective RNFs tested are presented in Figure 1.



**Figure 1** Recycled nutrient fertilisers (RNFs) evaluation centres established within the CiNURGi project and the respective RNFs tested.

## Relation to other project activities

Activity 2.2 naturally relates to A1.2 “Industry standards for evaluation and quality assurance of recycled nutrient fertilisers” which set the framework for the quality evaluation of RNFs. Moreover, A2.2 relied on A1.1 “Assess potential of nutrient recycling (NR) to improve national and regional nutrient balances” for the identification of most abundant and promising organic nutrient streams for production of RNFs.

## 2. Lexicon

In this chapter, some basic definitions related to plant nutrition and agronomic terms used in the report are clarified, and a brief description of the technologies leading to the production of recycled nutrient fertilisers (RNFs) is presented. Furthermore, the most common methods for the basic characterisation of RNFs are briefly presented.

**Recycled nutrient fertilisers (RNFs)** are products intended to be used in agriculture as plant fertilisers and/or amendments and derive after treatment of organic waste materials such as manure, sewage sludge, food industry by-products etc. In this report, RNFs are further distinguished into organic RNFs and mineral RNFs, according to the proportion of nutrients present in organic or mineral forms. Mineral RNFs are distinguished from the term “mineral fertilisers” as the latter derive from natural deposits or produced industrially without recycling of nutrients recovered from biomass.

### 2.1 Plant nutrition

Plants require 14 elements which are essential for their growth and development, in addition to carbon (C), hydrogen (H) and oxygen (O). Depending on their relative concentration of nutrients within a plant, elements can be classified as macro-nutrients (> 1 g kg<sup>-1</sup> plant dry matter) and micro-nutrients (< 1 g kg<sup>-1</sup> plant dry matter).

#### **Plant macro-nutrients:**

**Nitrogen (N)** is the most limiting factor for plant growth. The major sources of N taken up by plant roots are nitrate (NO<sub>3</sub><sup>-</sup>) and ammonium (NH<sub>4</sub><sup>+</sup>), the sum of which constitutes the inorganic N content (N<sub>inorg</sub>). Soil application of mineral fertilisers can directly supply these forms to plants, depending on the composition of the added fertiliser. In contrast, organic fertilisers rely on two opposing processes to determine the availability of inorganic nitrogen namely mineralization of organic nitrogen into plant-available forms, and microbial immobilization, which temporarily assimilates inorganic nitrogen into microbial biomass.

**Phosphorus (P)** is essential for the transfer and storage of energy in plants and is a constituent of cell membranes and the genome. Roots take up P as orthophosphate-P (H<sub>2</sub>PO<sub>4</sub><sup>-</sup> or HPO<sub>4</sub><sup>2-</sup>), and P concentration in plant tissues ranges between 0.1 and 0.5%. The availability of P in soil solution is regulated by processes including mineralization/immobilization, desorption or adsorption on clay and other mineral surfaces, and dissolution or precipitation. Soil pH significantly influences the rate of these processes.

**Potassium (K)** in plants influences metabolic processes related to photosynthesis and regulates the plant water homeostasis. Potassium in soil is mostly present as the positive ion in its soluble form (>95%) and K can be sufficiently supplied through mineral fertilisers or organic fertilisers.

**Sulphur (S)** is necessary for the synthesis of amino acids which are important components of plant proteins. Sulphur is taken up by roots as  $\text{SO}_4^{2-}$  but gaseous emissions can also be absorbed by plant leaves. Similar to nitrogen, sulphur is subject to microbial mediated mineralization-immobilization processes as well as adsorption/desorption on clay minerals and organic matter.

**Calcium (Ca)** acts as a structural component and regulates the translocation of carbohydrates and nutrients within the plant. Calcium concentration in plants ranges between 0.2 and 1%. Similar to K, RNFs contain Ca in an available soluble form that can replenish the Ca stock in soil.

**Magnesium (Mg)** is essential for photosynthesis, and plants contain up to 0.4%  $\text{Mg}^{2+}$ . It occurs in soil primarily in soluble form, and its availability is largely determined by the total Mg supply, soil pH, and CEC.

**Plant micro-nutrients:** This category comprises essential elements that are required by plants in concentrations < 0.1 g kg<sup>-1</sup> plant dry matter and includes chloride (Cl), boron (B), iron (Fe), manganese (Mn), zinc (Zn), copper (Cu), nickel (Ni), and molybdenum (Mo).

## 2.2 Agronomic terms

**Nutrient uptake** is the rate of nutrient acquisition from plants and is measured as the concentration of nutrients in harvest biomass. Nutrient uptake can be measured for the whole plant biomass (aerial and aboveground biomass) or for parts of the plant. It can be calculated as:

$$\text{Nutrient uptake} = \text{harvested biomass DM(g)} * \text{nutrient concentration(mg g}^{-1}\text{DM)}$$

**Nutrient uptake efficiency** expresses the plant nutrient content relatively to the respective amount of nutrient applied with fertiliser and is calculated as:

$$\text{Nutrient use efficiency} = \frac{\text{Nutrient uptake}_{\text{fertilized treatment}}}{\text{Nutrient applied}_{\text{fertilized treatment}}}$$

Values greater than 1 indicate soil nutrient mining whereas values lower than 1 indicate excessive application of nutrients. Nutrient use efficiency can be calculated for all plant nutrients, but most often respective calculations are made for N(NUE), P(PUE), and K (KUE).

**Apparent nutrient recovery (ANR)** indicates the proportion of nutrients taken up by plants to the total amount of nutrient applied to soil. In studies, the ANR from un-fertilized soil usually serves as the basis for comparison therefore, often subtracted and ANR is determined as:

$$\text{ANR} = \frac{(\text{Nutrient uptake}_{\text{fertilized treatment}} - \text{Nutrient uptake}_{\text{non-fertilized treatment}})}{\text{Nutrient applied}_{\text{fertilized treatment}}}$$

This approach is useful to estimate the relative benefits of fertilisers to nutrient uptake. High ANR values indicate low nutrient surplus in soil whereas low ANR values indicate either losses of nutrients from soil or their plant unavailability. Similarly to NUE, it can be calculated for all plant nutrients, but most often respective calculations are made for N (ANR), P (APR), and K (AKR).

Mineral fertiliser equivalent (MFE) is used to describe how efficient an RNF is to provide available nutrients to crops compared with a mineral fertiliser. Alternatively, the term is referred to as

mineral fertiliser replacement value and calculated as:

$$\text{MFE}(\%) = \frac{\text{ANR}_{\text{fertilized treatment}}}{\text{ANR}_{\text{reference}}} * 100$$

This approach uses mineral fertilisers as reference treatments applied at different rates. The crop ANR response curve to the different reference application rates is calculated either based on equal nutrient application rates between the reference and RNF treatments or based on equal ANR rates. The mineral fertiliser equivalent value can be estimated for all nutrients; however, it is often calculated for N (N-MFE) and P (P-MFE). Alternatively, MFE can be calculated from the agronomic efficiency values as:

$$\text{MFE}(\%) = \frac{\text{AE}_{\text{fertilized treatment}}}{\text{AE}_{\text{reference}}} * 100$$

Where  $\text{AE}_{\text{fertilized}}$  and  $\text{AE}_{\text{reference}}$  denote the additional yield per kg of nutrients applied in the fertilized and reference treatments, respectively.

### 2.3 Technologies for producing recycled nutrient fertilisers.

**Anaerobic digestion** refers to the controlled microbial degradation of an organic feedstock. During the process, biogas is produced which can be further used as energy source. The residual of the process is commonly referred to as digestate and can be used as an organic fertiliser and amendment due to its nutrient and organic matter content. Compared to the initial feedstock, digestate often demonstrates lower dry matter content, increased  $\text{NH}_4\text{-N}$  to  $\text{N}_{\text{tot}}$  ratio, increased pH and lower total carbon content. The higher proportion of  $\text{NH}_4\text{-N}$  to  $\text{N}_{\text{tot}}$  in the digestate may promote its N fertilizing value however, the digestate may be prone to higher ammonia ( $\text{NH}_3$ ) emissions during storage due to its high pH and  $\text{N}_{\text{inorg}}$  availability.

**Phase separation.** In most cases, the N to P ratio in organic fertilisers is lower to the respective needs of the plants and often results in an over-application of P when organic fertilisers are applied to meet crop N demands. Excessive fertilization of crops with P might jeopardize environmental quality and constitutes an irrational use of scarce P resources. Consequently, large farms often face the challenge to efficiently utilize the nutrients in the manure due to high amount of P accumulated in the farm. Phase separation of dilute organic waste produces a liquid fraction low in dry matter and rich in  $\text{NH}_4\text{-N}$  and K, which can be directly used in nearby fields. The solid fraction is relatively dry (moisture content 65–82 %), rich in organic matter, organic nitrogen and contains a large proportion of the P load of the feedstock. The concentration of P in the solid fraction, facilitates the potential exportation of excessive P and organic matter in areas with respective deficiencies.

**Composting** is an aerobic treatment of organic material aiming to increase the stability of the feedstock's organic matter, the recovery of nutrients and the sanitation of the final product called compost. For dilute feedstock, a phase separation treatment often precedes composting where the solid effluents are mixed with other materials (bulking agents) to facilitate the composting process. Traditionally composting takes place in outdoor piles with natural ventilation but closed systems with forced aeration are also available. In open systems, composting is prone to  $\text{NH}_3$ ,  $\text{CO}_2$ ,  $\text{CH}_4$  and  $\text{N}_2\text{O}$  losses nevertheless, emission mitigation measures can successfully reduce the environmental impact of the process (Bernal et al., 2017).

**Drying** of diluted organic waste aims at reducing the mass/volume of the feedstock and the concentration of nutrients in the end-product. This treatment is often adopted to decrease the transportation costs and increase the cost-efficient transportation of the dried end-product. A variety of batch and continuous drying industrial setups exists where drying temperature is often higher than 70 °C, to secure sanitation of the final product. Due to the exposure of the feedstock to high temperatures during drying, the potential plant available N form is emitted as NH<sub>3</sub>. Most large-scale dryers are equipped with wet scrubbers to reduce odour and gaseous emissions. Particularly, acid scrubbers may be used to capture the NH<sub>3</sub> emitted during drying, to reduce the environmental impact of the drying unit and profit by the utilization of the produced ammonium sulphate as mineral fertiliser.

**Pyrolysis** is called the thermo-chemical decomposition process during which biomass is heated under oxygen limited conditions at temperatures ranging between 300 °C - 1200 °C. Pyrolysis results in a carbon-rich product called biochar, emission of volatile matter which after condensation is known as bio-oil, and syngas (CO, CO<sub>2</sub>, CH<sub>4</sub> and H<sub>2</sub>) (Kambo et al., 2015). The produced syngas can be further utilized to produce methanol and dimethyl ether while bio-oil can replace diesel to produce electricity (Tayibi et al., 2021). The biochar can be used in agriculture, and studies have shown that biochar can increase the buffer capacity of soil, increase crop yields, reduce nutrient leaching and GHG emission and replenish soil organic carbon stocks. Nevertheless, similar to all organic waste treatments, the magnitude of biochar induced effects on soil properties and crop yields highly depends on the initial feedstock and the pyrolysis conditions.

**Pelletisation** refers to the compression of biomass by mechanical pressure to enhance handling, transport, storage, and soil application of materials such as composts and powdery dried organic fertilisers and amendments. Pelletised organic fertilisers often appear to release plant nutrients at a more gradual pace compared with their non-pelletised counterparts. Occasionally, organic fertilisers are mixed with mineral fertilisers or other materials (e.g. biochar, bio-extracts) to create tailor-made products for plant growth (Sunginthara et al., 2024; Papandrea et al., 2021).

**Struvite precipitation** is a chemical method of phosphorus fixation by metal (iron or aluminium) salt from. However, several technical barriers still limit the widespread implementation of this technology. In particular, the small size of most struvite crystals makes them difficult to handle and incorporate into soil. Moreover, granulating struvite is typically far more complex than producing the crystals themselves, often requiring tighter process control and higher energy inputs (Latifian et al., 2012).

## 2.4 Methods for the characterization of RNFs

### Dry matter and volatile solids

Dry matter (DM) content, in some cases also referred to as total solids (TS) content especially in slurries, is the material remaining after water evaporation from the sample. Recommended drying temperatures for soils and RNFs is 105 °C. For accurate estimation of the DM content of a sample, the weight of the dried sample should remain constant (<0.1% DM change) after at least 4 hr additional drying time. For volatile solids determination, drying of the sample takes place in oxic conditions at 550 °C.

Calculations:

$\% \text{ Dry matter} = ((\text{weight dried sample} + \text{container}) - (\text{weight empty container})) * 100 / ((\text{weight fresh sample} + \text{container}) - (\text{weight empty container}))$

### Total nitrogen

The estimation of the total nitrogen content in RNFs is important as it will dictate the required amounts to meet the objectives of the fertilization plan. Predominantly two methods are utilized for the estimation of total N in organic fertilisers.

The Kjeldahl method relies on the conversion of total N contained in the sample to ammonium N using concentrated sulfuric acid, catalysts, and salts under elevated temperatures. Subsequently, determination of the ammonium nitrogen concentration in the Kjeldahl digest can be performed spectrophotometrically, by diffusion conductivity, or using ammonia electrodes. The results of this method are often reported as total Kjeldahl nitrogen (TKN) in laboratory results. This method involves relatively inexpensive instrumentation, and most modern systems are automated to handle large sample sizes. Moreover, the method permits the analysis of both wet and dry samples, reducing the need for sample drying. Disadvantages of the method include the need for strong acid, long digestion times, and laborious procedures. Moreover, the nitrate and nitrite content of the sample are not completely accounted for in TKN.

**Duma's method** involves combustion of samples which converts the N content of the sample to nitrous oxides and then to dinitrogen gas ( $\text{N}_2$ ). The produced  $\text{N}_2$  is then detected by thermal conductivity cell. The advantage of the method include fast analysis of the samples and complete recovery of all N forms in the sample and modern instruments can be combined with multiple element configurations to detect simultaneously carbon (C), sulphur (S), hydrogen (H). However, purchase and maintenance cost of the analytical instrumentation is costly. Moreover, many combustion instruments rely on very small sample sizes (<200 mg), which necessitates extensive homogenization to obtain a representative subsample. In addition to the challenges posed by small sample size, drying of RNFs materials can also lead to losses of  $\text{NH}_4\text{-N}$ , thereby affecting the accuracy of total nitrogen measurements. Although some equipment, such as LECO (Laboratory Equipment Corporation) is technically capable of analysing moist samples and could therefore avoid such drying-related losses, routine use was not feasible: after analysing several batches of fertiliser products, the instrument required excessive cleaning, and further analysis of such materials was subsequently prohibited.

### Ammonium nitrogen

Ammonium nitrogen is the main inorganic nitrogen form in most organic fertilisers which may dictate the immediate plant N availability of RNFs. Estimation of  $\text{NH}_4\text{-N}$  content and total-N in RNFs can give an estimation of the organic N content of samples and of the RNFs potential to provide N to plants.

**Electrode determination.** The ammonium nitrogen content in RNFs can be estimated potentiometrically using a selective  $\text{NH}_3\text{-N}$  electrode. The sample is mixed with reagent grade water, and the pH of the solution is raised with the addition of a strong base. Dissolved  $\text{NH}_4\text{-N}$  is converted to  $\text{NH}_3\text{-N}$  which diffuses through the membrane of the electrode and alters the pH of

the internal solution. Based on the calibration curve of the electrode it is possible to determine the  $\text{NH}_4\text{-N}$  of the sample. The method does not require any extraction steps, but the electrode readings often need time to stabilize, and the precision of the method is often reported lower than other methods.

**Spectrophotometry.** The  $\text{NH}_4\text{-N}$  content of RNFs is firstly extracted with 1M KCL solution at variable sample-to-extractant ratio (most commonly 1:10) and filtered extracts are analysed by either spectrophotometry, distillation/titration or diffusion-conductivity. Spectrophotometry is the most common method use is based on the alkaline phenol Berthelot reaction or on the on the sodium salicylate modified Berthelot reaction. Analytic equipment capable to perform the determination of  $\text{NH}_4\text{-N}$  include flow injection and continuous flow analysers. In the distillation/titration method the sample is distilled into a boric acid solution and subsequently titrated with sulfuric acid. The end point of the titration is detected by either a pH meter or an indicator solution. In the diffusion-conductivity method,  $\text{NH}_4\text{-N}$  is determined by the change in the conductivity measurements of a hydrophobic membrane. The extraction step of the methods is prone to ammonium contamination from filter papers, reagent and analytical equipment therefore care should be taken. Coloured samples may require clarification to avoid interference during spectrophotometric analysis.

### Nitrate nitrogen

Similar to ammonium nitrogen determination,  $\text{NO}_3\text{-N}$  requires extraction with KCL solution and filtering. The extracts can be analysed spectrophotometrically with a cadmium or hydrazine reduction method. The method does not differentiate between nitrate and nitrite content in the samples, but it is often assumed that the concentration of the latter is negligible. Often spectrophotometric analysis of  $\text{NH}_4\text{-N}$  and  $\text{NO}_3\text{-N}$  is performed simultaneously for the same KCL extracts.

### P, K, Ca, Mg and trace elements

Determining the phosphorus (P), calcium (Ca), and potassium (K) content in RNFs is important because it provides insight into the amount of nutrients that are potentially available for plant uptake. At the same time, the application of these elements to soil is often subject to regulations to prevent over-fertilization and environmental impacts. Knowing the nutrient composition of RNFs therefore allows for better management decisions, ensuring both compliance with regulations and efficient use of nutrients to support crop growth.

The principal of the specific elemental analysis of RNFs includes the destruction of any organic matter present in the sample and the subsequent solubilization of the elements. The available digestion methods vary but generally include first drying of the samples and subsequently one of the following steps i) heat samples after the addition of hydrogen peroxide and concentrated acid, ii) microwave assisted digestion of samples after the addition of concentrated acid iii) combustion of samples and dissolution of the elements after addition of concentrated acid to the resulting ashes.

Following digestion samples can be analysed by the following methods:

- **Atomic absorption spectroscopy.** The method relies on the elements atoms specificity to

absorb light at characteristic wavelengths. The sample is first converted to gaseous atoms, and light passes through the gases. The absorb light is proportional to the concentration of the target element. The method is not suitable for the determination of P and is highly specific for individual elements.

- **Inductively coupled plasma spectroscopy.** This method relies on the excitation of atoms from the ground state to an excited state. The element specific emission of light at certain wavelengths (ICP-OES) or the intensity of light emission (ICP-AES) is determined and assigned to the proportionally dedicated to the concentration of the specific element.

## pH

The solubility of nutrients in RNFs as well as NH<sub>3</sub> and other gaseous emissions are affected by pH therefore, RNFs pH regulates their potential to supply nutrients to plants and adversely impact the environment. Measurement of pH in liquid mediums is measured directly by electrode while in case of solid or semi-solid samples, pH is be measured in a 1:2 RNF/water (dry weight/volume) solution.

## 3. Protocols for the assessment of the agronomic value of RNFs

The protocols developed within the framework of A2.2 aimed to standardize the experimental approaches between partners and ensure meaningful comparison between studies and harmonization of the reporting. However, it was also the intention of the partners to allow small deviations from the commonly agreed protocol for country-specific experimental practices which would facilitate the dissemination of the results to local audiences.

To assess the agronomic performance of RNFs, both soil incubations and greenhouse pot trials were carried out within A2.2. The incubations provided information on nutrient release dynamics and carbon stability of the RNFs in soil, while the pot trials tested their effects on plant growth, yield, and nutrient uptake. Together, these approaches allowed for a comprehensive evaluation of the potential of RNFs to substitute mineral fertilisers and support sustainable crop production. Table 1 provides an overview of the RNFs tested by each partner and the type of study performed. More information about the RNFs tested is presented in Section 4 of this report.

**Table 1 List of RNFs tested by partners of the CiNURGi project in incubation and pot trials. "X" indicates the type of study utilized and the parameters assessed. C mineralisation, N and P evolution tested in incubation studies and availability of N and P in pot trials.**

Institute	RNF tested	Incubation			Pot trial	
		C	N	P	N	P
Institute of Fluid-Flow Machinery (IMP-PAN), Poland	6 kinds of the multi-waste pellets: BADSS (biochar, biomass ash, sewage sludge, fish-food waste) and SADSS (cattle shavings, biomass ash, sewage sludge, fish-food waste)				x	

Institute	RNF tested	Incubation			Pot trial	
		C	N	P	N	P
	BADSS and SADSS Pellets enriched with bakery/ dairy waste including effective microbes BADSS and SADSS Pellets enriched with a seafood waste including chitosan Dry urine granules (Sanitation 360)					
Julius Kühn Institute (JKI), Germany	Struvite					x
Natural Resource Institute of Finland (Luke), Finland	Meat and bone pellets Solid fraction of cow manure digestate Cow manure digestate Liquid fraction of digestate	x	x	x	x	
Centre of Estonian Rural Research and Knowledge (MEKT), Estonia	Struvite					x
Linnaeus University, Sweden	Sewage sludge biochar from 10 different sewage plants Struvite				x	x
Swedish University of Agricultural Sciences (SLU), Sweden	Solid fraction of digestate Compost Biochar Digested pig manure and slaughter waste pellets Digestate pellets Struvite Dry urine granules Multi-waste pellets	x	x	x		x

### 3.1 Soil sampling, preparation and analysis

- Each partner collected soil from the plough layer of a field with known fertilisation history. Efforts were made to access soil which had not received organic amendments in the recent past (more than 3 years) and had moderate to low organic matter content. Soil was sampled in sufficient amount to cover the needs of both the incubation and the pot trial.
- Soil was partially air-dried to facilitate handling and sieving. Preferably, soil should not be completely dried during preparation to avoid altering its biological/chemical characteristics.
- When not used immediately, soil should be stored relatively moist at 4 °C for minimum time (<2 months).

- Visible debris was handpicked and removed from soil before and after soil sieving through to remove gravel and large debris particles according to local practices 2 mm (incubation) / 4 mm (pot trial) sieves, depending on the gravel content of the soil.

Proposed physicochemical analyses of the soil can include: soil texture, organic C, N total/N inorganic, available P (estimation of soil's P availability can be measure based on country-specific schemes), available K, water holding capacity (WHC). Before the start of the incubation/pot trials, distilled water was added to the soil to reach 40% WHC and then the soil was pre-incubated, in the dark, for 10–14 days, at stable temperature, representing the mean temperature of the growing season in each country.

### 3.2 Common protocol for assessing RNFs C mineralization and N-P dynamics in soil incubations

The selection of RNFs tested within A2.2 relied on individual partner organization according to the relevance of each RNF for each country (feedstock availability, farmers perceptions, legislation etc.). Furthermore, it was within the scope of the CiNURGi project that evaluation centres should test the RNFs produced by the project partners. Moist RNFs were stored in a freezer (-18 °C) until further use, while dried RNFs were stored at room temperature in closed, dark containers.

#### 3.2.1 Characterization of RNFs

- Dry matter determination after drying of the samples to constant weight at 105 °C for 24h
- Volatile solids, calculated as mass loss on ignition after drying of the samples at 550 °C
- Total carbon and nitrogen of dried samples were estimated on a CN auto-analyse
- Inorganic N, NH<sub>4</sub>-N and NO<sub>3</sub>-N content of the samples was determined after extraction with 1 M KCl and filtration of extracts, by flow injection analysis.
- Total phosphorus and all micro-nutrients were determined in dry samples with ICP-OES in chemically digested samples. Nitric acid (HNO<sub>3</sub>) and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) were utilized as digestion agents to improve the dissolution of the solids
- Water extractable phosphorus, determined after water extraction (1:60, sample (DM): distilled water), shaken on a rotary shaker for 1 h and subsequent filtration through a 0.42 µm syringe filter. Inorganic orthophosphate P concentration was determined by flow injection analysis with the Ammonium Molybdate method.
- Extractable phosphorus in sludge biochars was determined using 0,5 M HCl and 10 mM oxalate extraction (Fransson 2001), after method testing, to represent different parts of the P continuum in the biochars. The sludges from different sewage plants contain variable shares of Fe and Al used to fix the P in the cleaning process and the P that is potentially available from these compounds is represented in the oxalate extraction (Campbell and Schwertman 1984). The acid extraction is a method to determine the storage P in soils (Kuo, 1996) and can be transferred to extract P from biochars.
- For mineral RNFs, basic characterization was limited to the anticipated composition.
- Unstable RNFs were stored in a freezer (-18 °C) until further use, while stable RNFs were

stored in room temperature in closed, dark containers.

- For homogeneous mixing of RNFs with small soil quantities (e.g. in incubation set ups), grinding of dry samples may be used to appropriate particle size (e.g. <2mm).

### 3.2.2 General approach of incubations

During the soil incubation studies, N and P release from RNFs was monitored over a period of five months in the absence of plants. Carbon dioxide (CO<sub>2</sub>) emissions from the soil were measured in the same experimental setup at regular intervals.

Soil portions, typically 50–100 g, were thoroughly mixed with RNFs to supply equal amounts of either N or P across all treatments, depending on the type of RNF. In some cases, RNFs were applied based on their dry matter relative to soil weight (e.g., 1% RNF: soil, equivalent to approximately 8 t ha<sup>-1</sup>). Soil/RNF mixtures were placed in individual containers with loosely attached lids to maintain aerobic conditions while minimizing moisture loss.

Each treatment was set up in triplicate and further replicated to allow non-destructive sampling throughout the incubation. Nutrient availability was measured at several time points: days 7, 14, 21, 40, 60, 90, 120, and 150 for NH<sub>4</sub>-N, NO<sub>3</sub>-N, and pH, and days 7, 60, and 150 for water extractable P.

Incubation samples were kept in the dark at stable temperatures representative of the local growing season. Soil moisture was maintained at 60% of WHC through regular addition of distilled water.

Each soil sample was mixed, and two aliquots (10 g wet weight) were removed for dry matter content (oven drying at 105 °C) and inorganic N determination. Soil was extracted with 1 M KCl for NH<sub>4</sub>-N and NO<sub>3</sub>-N determination at a 1:4 (soil dry weight/water) ratio. The mixtures were shaken using a 360° vertical shaker for 60 min at room temperature and filtered through ashless filter papers (Whatman no. 44). The extracts were kept frozen (-18°C) until they were analysed calorimetrically using flow injection analysis. Briefly½, NH<sub>4</sub>-N determination was based on the formation of NH<sub>3</sub> in alkaline media and the subsequent detection of NH<sub>3</sub>-N by gas diffusion. Determination of NO<sub>3</sub>-N was based on the formation of NO<sup>2-</sup> in a Cd reduction column and the subsequent photometrical measurement of NO<sup>2-</sup>-N. Soil pH and electrical conductivity were determined in Milli-Q water on 5 g of air-dried soil solution using a combined electrode (1:5, w/w).

#### Organic N mineralized to soil as percentage of organic N added

The amount of organic N mineralized from organic RNFs (N<sub>m</sub>) was calculated as:

$$N_m (\%) = (N_s - N_a - N_c) / N_{org} * 100$$

Where N<sub>s</sub> is the mineral N concentration in the RNF treated soil at the end of the incubation, N<sub>a</sub> is the amount of N<sub>inorg</sub> supplied in the soil in through the RNFs, N<sub>c</sub> is the N<sub>inorg</sub> concentration of the control soil and N<sub>org</sub> is the amount of organic N added to the soil by the RNFs. All units were mg N kg<sup>-1</sup>.

#### Nitrogen release to soil as percentage of total N added

Since the N<sub>inorg</sub> content of RNFs might differ significantly, presentation of organic N mineralized might not fully depict the release of N<sub>inorg</sub> to soil. Therefore, N release as a percentage of total N

added to the soil was also calculated as:

$$N_{\text{rel}} (\%) = (N_s - N_a - N_c) / N_{\text{tot}} * 100$$

where  $N_{\text{rel}}$  depicts the percentage of total N supplied in the soil solution during the incubation,  $N_s$  is the mineral N concentration in the RNF treated soil at the end of the incubation,  $N_a$  is the amount of  $N_{\text{inorg}}$  supplied in the soil in through the RNFs,  $N_c$  is the  $N_{\text{inorg}}$  concentration of the control soil and  $N_{\text{tot}}$  is the total amount of N added to the soil by the RNFs. All units were mg N kg<sup>-1</sup>.

### **Phosphorus release to soil as percentage of total P added**

Phosphorus release was presented as the proportion of water extractable- to total-P applied as:

$$P_{\text{rel}} (\%) = ((P_s - P_c) * 100) / P_{\text{tot}}$$

where  $P_{\text{rel}}$  depicts the percentage of total P supplied in the soil solution during the incubation,  $P_s$  is the water extractable P concentration in the RNF treated soil at the end of the incubation,  $P_c$  is the water extractable P concentration of the control soil and  $P_{\text{tot}}$  is the total amount of P added to the soil by the RNFs. All units were mg P kg<sup>-1</sup>.

### **Carbon mineralization as percentage of total C applied**

CO<sub>2</sub> emissions were monitored in triplicate samples according to established laboratory routines, for example using alkali traps with subsequent titration by HCl. Emissions were measured more frequently at the beginning of the incubation (days 2, 4, 6, 10, 14, 21, 30, 40) and less frequently at later stages (days 60, 80, 100, 120, 150).

## **3.3 Common protocol for determining the P fertilizing value of RNFs in greenhouse experiments**

Estimation of the P content in RNFs provides a while incubation studies provide only a snap of the situation

### **Soil Preparation and Pre-Incubation**

The previously sampled soils were homogenized and placed in individual pots according to predetermined quantities. Where necessary, soils were limed and supplemented with additional nutrients—K, Mg, S, Mn, Zn, Cu, Mo, B, and Fe—to ensure non-restrictive plant growth. Nutrients were added either as solutions or solid forms. Tentatively, the following fertilisers were used: KCl, MgSO<sub>4</sub>·H<sub>2</sub>O, MnSO<sub>4</sub>·H<sub>2</sub>O, ZnSO<sub>4</sub>·7H<sub>2</sub>O, Na<sub>2</sub>MoO<sub>4</sub>·H<sub>2</sub>O, H<sub>3</sub>BO<sub>3</sub>, and FeSO<sub>4</sub>·7H<sub>2</sub>O. Distilled water was added to adjust soil moisture to 60% of WHC, and soils were pre-incubated for at least one week.

### **RNFs and plants used**

RNFs were applied at rates adjusted to supply equal amounts of total P (approx. 50 mg P kg<sup>-1</sup> soil across all studies). To ensure homogeneous distribution, RNFs were applied in ground form except meat bone meal pellets and sludge biochars in Finland that were applied in their produced form. Mineral N was applied as ammonium-nitrate (NH<sub>4</sub>NO<sub>3</sub>) to ensure non-limiting conditions for plant growth. Ryegrass was selected as a test crop from JKI, Luke and SLU and maize was selected at Linnaeus University, due to the high nutrient demands and high growth rates of the crop. Oilseed rape was used from METK as a regionally important crop. To estimate the mineral fertiliser equivalent (MFE) of RNFs, reference treatments received sole mineral fertilisers,

establishing a plant N or P uptake dose-response curve. For instance, when RNFs supplied 150 mg N kg<sup>-1</sup> soil, reference mineral N treatments included 0%, 33%, 66%, and 133% of the total N applied with RNFs. For P-MFE, reference treatments corresponded to 25%, 50%, 100%, and 125% of the total P applied with RNFs (Table 1). Superphosphate was used as a mineral fertiliser control and non-fertilised soil as response control.

### **Pot Trial Setup and Maintenance**

After the pre-incubation period, RNFs and mineral fertilisers were applied to each pot after thorough mixing with the soil. Treatments were replicated at least four times to ensure adequate statistical power. Pots were sown with 1–2 g of perennial ryegrass seeds, covered with 300–500 g of soil, and soil moisture was adjusted to 60% WHC. Maize was pre-germinated and one sprout was transferred per pot in the sludge biochar cultivation tests.

Pots were placed in a greenhouse, randomly distributed, and regularly reshuffled to minimize positional effects. Greenhouse conditions reflected mean temperature and light conditions of the local growing season, and soil moisture was regularly checked and maintained at 60% WHC.

Ryegrass was harvested three times, with harvesting periods differing between studies. Shoots were cut 3 cm above the soil surface in the first two harvests and at soil level in the third. After each harvest, a nutrient solution containing all elements except the nutrient under investigation was applied to sustain plant growth.

Maize height growth was determined throughout the experiment, and the plants were harvested after 7–8 weeks of growth. The harvest was done at the onset of flowering to avoid nutrient retranslocation due to seed production. Two pot trials were done consecutively, first with a moderately fertile soil and then with a very poor soil.

Shoot biomass dry matter content was determined after drying samples for 72 h at 60 °C. Dried biomass was milled and analysed for total N via oxygen combustion. Total P and other elements were determined by ICP-OES after microwave digestion with nitric acid (HNO<sub>3</sub>).

### **Data Analysis**

Apparent P recovery (APR) was calculated as the difference in shoot biomass nutrient uptake between treatments and the control, divided by the P total applied. Nitrogen and phosphorus mineral fertiliser equivalence (N-MFE and P-MFE) of RNFs were determined by regressing N or P uptake from RNF-treated ryegrass against the linear response of ryegrass receiving increasing levels of mineral N and P fertilisers.

## **4. Evaluation of RNFs**

### **4.1 Incubation studies**

#### **4.1.1 Evaluation centre in Sweden (SLU)**

##### ***Soil***

For the incubation study, two soils with contrasting agronomic phosphorus (P) status were

collected from different locations in Sweden: a high-P soil from Krusenberg (59°44'28.2"N, 17°41'01.2"E) and a low-P soil from Kårbo (60°29'28.3"N, 17°29'22.5"E). The Krusenberg soil (high-P) was classified as a sandy loam with a particle size distribution of 79% sand, 12% silt, and 7.7% clay. The Kårbo soil (low-P) was sandy, with 85% sand, 10% silt, and 2.8% clay. Detailed soil characteristics are presented in Table 2.

**Table 2 Soil properties**

Soil	pH	Ctot %	NO <sub>2+3-</sub> N mg/(kg DM)	P- H <sub>2</sub> O 4	P-AL 1.8	K- AL 2.8	Mg- AL 2.9	K/Mg- AL 0.9	Ca- AL 221	Al- AL 26	Fe- AL 39	WHC %
Kårbo	6.1	3.7	9.6	4	1.8	2.8	2.9	0.9	221	26	39	40
Krusenberg	5.6	1.4	4.8	14	9.9	8.1	3.4	2.3	39	28	17	48

After collection and preparation, part of each soil was used for the pot experiment, while the remaining portion was frozen for later use in these incubation study. Following thawing, the incubation setup was established according to the developed protocol. For each treatment, 50 g of soil (dry weight) was used.

### Treatment

Organic treatments included the solid fraction of digestate, biochar derived from digestate, biothermally produced compost, struvite (magnesium ammonium phosphate), and dry human urine (Table 3) In addition, three pelletized recycled nutrient fertilisers (RNFs) were tested: pellets derived from digested pig manure, slaughterhouse by-products, and mixed feedstock. All RNFs were applied at a rate corresponding to 45 mg P kg<sup>-1</sup> soil. These treatments were compared with an unfertilized control (CR) and a mineral source of P (KH<sub>2</sub>PO<sub>4</sub> solution). Both soil types received the same treatments as in the pot experiment, except that the low-P soil received two additional RNF treatments provided by project partners.

**Table 3 Content of total nitrogen (N), phosphorus (P) and carbon (C) in the different RNFs in dry matter**

Treatment, abbreviation	Original material	Ptot, mg/g	Ntot mg/g	Ctot, mg/g	pH
Struvite (ST)	Sewage sludge	99.75	17.0	41.0	7.4
Pelletized digestate (PDA)	Digested pig manure and house slaughter waste	9.74	138.4	272.2	5.6
Digestate (CS)	Separated pig manure	0.94	2.77		
Biochar (BCS)	Digested and separated pig manure	32.5	25.1	380.0	9.39
Compost (CM)	Based on CS	11.2	26.8	367.8	6.4
Pelletized digestate (PDE)	Digested pig manure	14.48	172.8	224.6	7.5
Pellets (PLY)	Mix of waste	18.90	51.3	230.1	9.2

Treatment, abbreviation	Original material	P <sub>tot</sub> , mg/g	N <sub>tot</sub> mg/g	C <sub>tot</sub> , mg/g	pH
Urine granules (U)	Dry human urine	15.53	165.3	301.3	4.8

### Incubation set up

Incubation was carried out for 187 days at 20 °C according to the protocol. During the incubation period, soil pH, mineral nitrogen, phosphorus (P-AL and water-soluble P), and dissolved organic carbon were measured. Soil respiration was assessed by trapping CO<sub>2</sub> emissions in 10 ml of 1 M NaOH. Soil samples and traps were placed in airtight one-litre jars, and fresh oxygen was supplied during each trap replacement. The CO<sub>2</sub> concentration in the NaOH solution was later determined by measuring electrical conductivity.

### Results

Plant-available phosphorus (P-AL) dynamics differed clearly between the two soil types and among treatments over the 190-day incubation

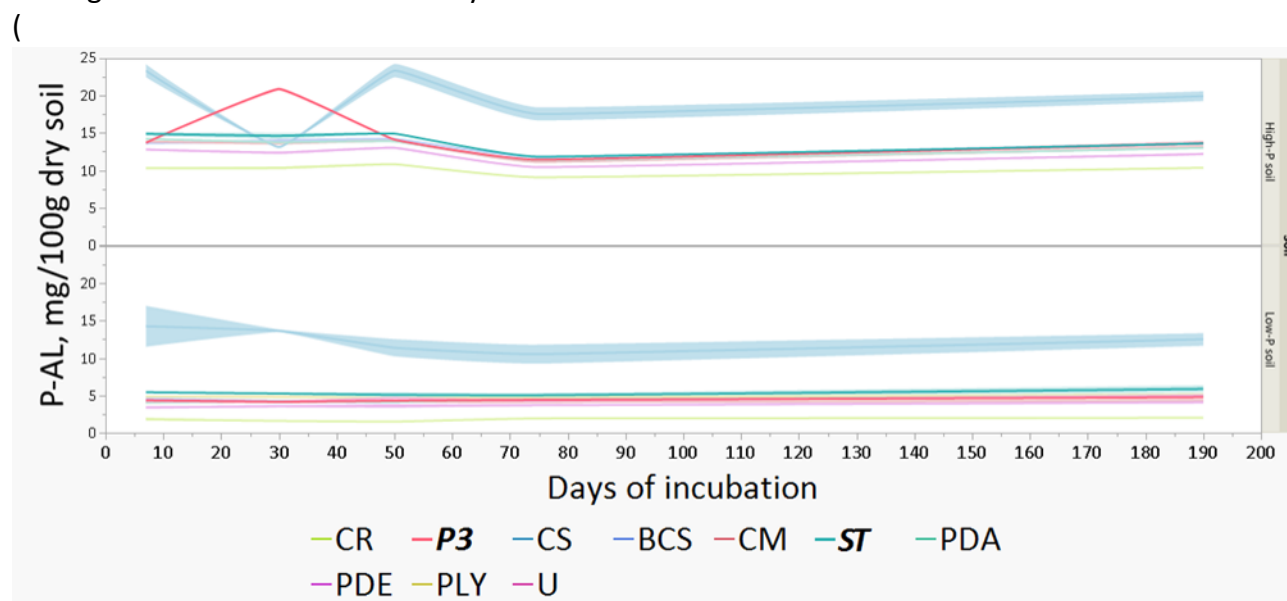
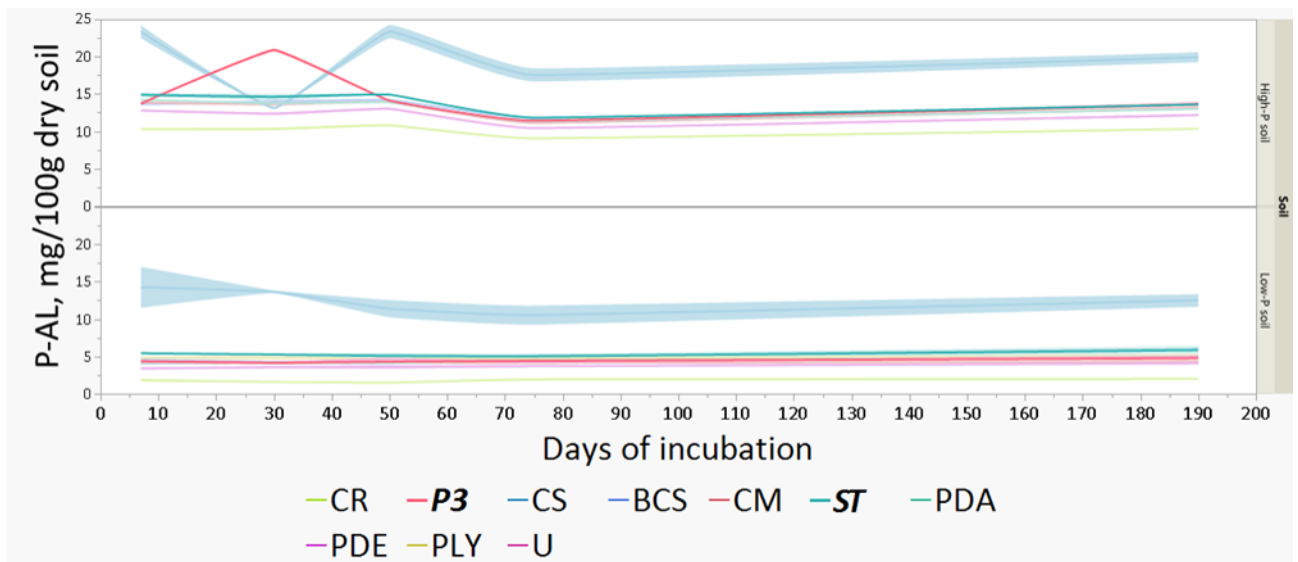


Figure 2). As expected from their initial properties, the high-P soil maintained higher P-AL values than the low-P soil, while the control (CR) remained stable in both soils (approximately 10 mg/100 g in the high-P soil and 2 mg/100 g in the low-P soil).

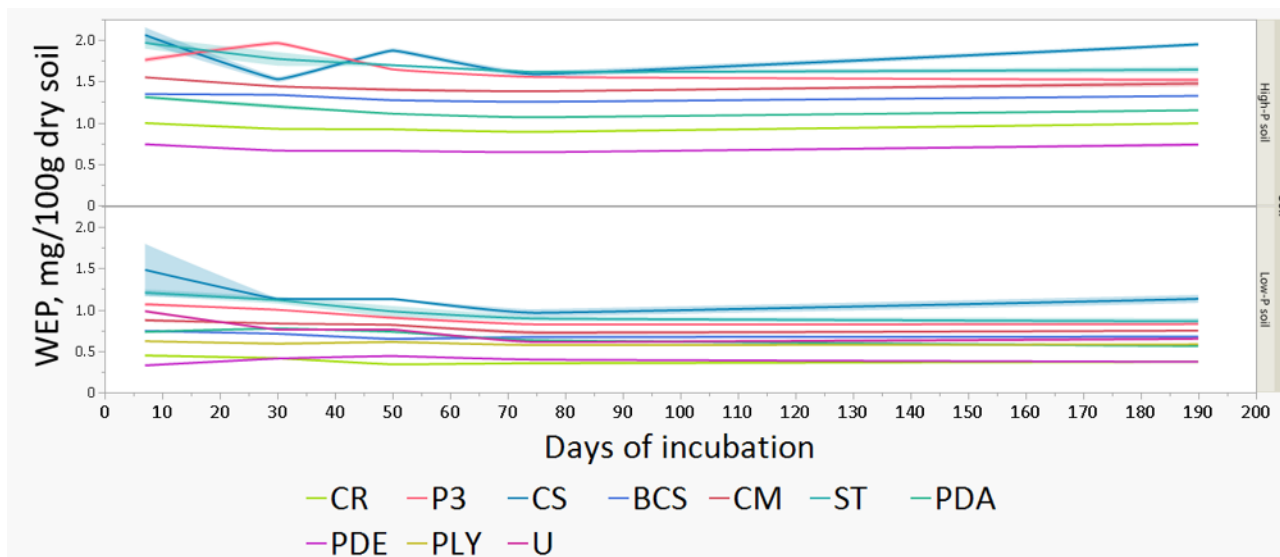


**Figure 2 Dynamics of plant-available phosphorus (P-AL, mg/100g dry soil) during 187-day incubation in high-P (Krusenberg) and low-P (Kårbo) soils amended with separated digestate (CS), biochar (BCS), and compost (CM), pelletized digestates (PDA, PDE), mixed-feedstock pellets (PLY), struvite (ST), and urine granules (U) relative to the unfertilized control (CR) and mineral P reference (P3)**

In the low-P soil, struvite (ST) and separated digestate (CS) produced the highest P-AL values, particularly at the beginning and end of the incubation. Treatments with dried urine, biochar, and PDA pellets showed P-AL dynamics similar to the mineral P reference (P3), while PLY pellets showed elevated P-AL early in the incubation but converged with the other RNFs after day 50. PDE consistently produced the lowest P-AL among the organic amendments. Overall, all RNFs in the low-P soil clustered tightly at approximately 4–6 mg/100 g, slightly above the control, and P3 did not differ markedly from the RNFs.

A similar pattern was observed in the high-P soil, although P-AL values were higher overall. Most treatments showed an increase up to day 50, a decline around day 75, and a slight rise toward the end of incubation.

Water-extractable P (WEP) also differed between soils and treatments, with consistently higher values in the high-P soil. In this soil, the control remained low and stable, while CS showed the strongest dynamics (Figure 3). WEP content after P3, BCS, and CM application declined gradually from similar starting values and converged around the same level by the end. ST showed a steady downward trend, while PDA, PLY, and PDE remained low and nearly flat, only slightly above the control. The final pattern placed CS, P3, and BCS at the upper end, CM and ST in the middle, and PDA, PLY, PDE, and CR at the lowest levels.



**Figure 3** Dynamics of water extractable phosphorus (WEP, mg/100g dry soil) during 190-day incubation in high-P (Krusenberg) and low-P (Kårbo) soils amended with separated digestate (CS), biochar (BCS), and compost (CM), pelletized digestates (PDA, PDE), mixed-feedstock pellets (PLY), struvite (ST), and urine granules (U) relative to the unfertilized control (CR) and mineral P reference (P3).

In the low-P soil, the control remained very low and unchanged. CS started highest and declined sharply before stabilising, remaining the top treatment throughout. P3 and ST began at similar levels, declined gradually, and converged by the end. BCS and CM showed small declines and remained close to each other. PDA, PLY, and PDE formed a tight cluster with minimal change, ending around the same value. U declined continuously from a relatively high starting point and finished at the control level. By the end of incubation, most treatments converged toward a narrow range, with CS clearly highest and U indistinguishable from the control.

Notably that high-P soil content mineral P application showed a rapid increase in P-AL and WEP levels at the start of incubation, peaking on day 30, followed by a subsequent decline, indicating phosphorus availability. In contrast, no such peak was observed in low-P soil which may indicate phosphorus fixation by the soil. Notably, none of RNFs showed the pronounced early peak observed with mineral form of P.

Mineral nitrogen ( $\text{N-NH}_4 + \text{N-NO}_3$ ) concentrations varied substantially among treatments, reflecting differences in organic matter content and nitrogen availability (Figure 5a). In both soils, the control (CR) and mineral P reference (P3) maintained low and similar mineral N concentrations throughout the incubation.

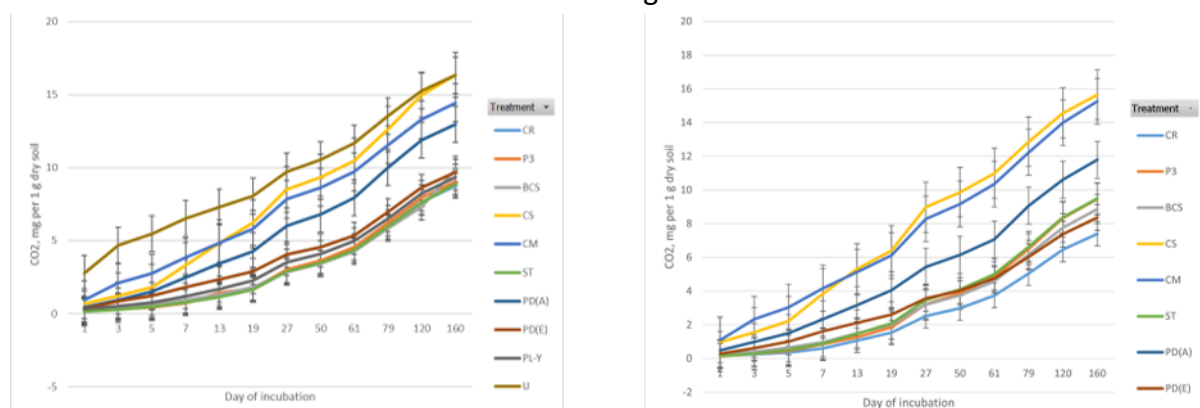
In the high P soil, mineral N clearly separated into three groups. The lowest group included CR, P3, biochar (BCS), and compost (CM), showed slow, steady increases from 5–12 mg N/kg at day 7 to 20–25 mg/kg by day 190, indicating minimal N mineralisation. The addition of mineral P (P3) had no effect on mineral N dynamics compared to the unfertilized control, confirming that P availability alone did not stimulate N mineralisation in this soil. Struvite (ST) remained slightly above this cluster, rising gradually over time. The highest group consisted of the separated digestate (CS) and the pelletized digestates (PDE and PDA). CS began around 62 mg/kg, remained stable early on, and then increased steadily toward 78 mg/kg. PDE maintained consistently high

values, rising gradually throughout the incubation. PDA showed the highest mineral N overall, with an early peak, a temporary mid incubation decline, and a recovery to the highest final concentrations.

Mineral N dynamics in the low-P soil showed a wider spread and more distinct treatment groupings. CR and P3 again formed the lowest group, increasing gradually over time. CM, ST, BCS400, and the mixed feedstock pellets (PLY) followed similar slow rising trajectories, ending slightly above the control. CS occupied an intermediate position, starting much higher than the low N group and increasing steadily after early fluctuations. PDE remained consistently high, while PDA showed a strong early peak followed by a decline and later stabilisation at values similar to PDE. Urine granules (U) were the most dynamic and highest N treatment, starting at very high levels, dipping mid incubation, and then rising sharply to the highest final mineral N concentration of all treatments. Overall, differences among treatments largely reflected the initial nitrogen content of the RNFs, and the results indicate sustained nitrogen release from the organic fertilizers across both soils.

Soil pH changed only modestly during the incubation period and remained relatively stable across both soil types and treatments. The control, mineral P reference, separated digestate, biochar, compost, multi waste pellets, and struvite all followed similar trajectories, with only minor deviations over time. Separated digestate caused a temporary increase in pH to 6.3 on day 7, after which values declined and aligned with the other treatments. In contrast, the pelletized digestates (PDA and PDE) and dried urine consistently lowered soil pH, marking them as the only treatments that induced measurable acidification.

Cumulative CO<sub>2</sub> emissions increased steadily throughout incubation in both soils. In the low-P soil, treatments with high organic C inputs (e.g., CM, BCS, PDA, PDE, PLY) produced the highest CO<sub>2</sub> emissions, indicating greater microbial activity and organic matter mineralisation (Figure 4). The control and P3 consistently showed the lowest CO<sub>2</sub> emissions, reflecting the absence of added organic substrates. In the high-P soil, overall CO<sub>2</sub> emissions were slightly lower, but treatment patterns were similar: RNFs with high C content stimulated respiration, while P3 and the control remained at the lower end of the range.



**Figure 4 Accumulated carbon emissions from RNFs during the incubation (left – low-P soil, right – high-P soil)**

## Conclusions

This 187-day incubation experiment revealed substantial differences in phosphorus availability, nitrogen mineralisation, pH dynamics, and microbial activity between a low-P soil (Kb) and a high-P soil (Kr) amended with various RNFs compared to a mineral P reference (P3, triple superphosphate as  $\text{KH}_2\text{PO}_4$ ) and an unfertilized control (CR). Most of the RNFs (BCS, CM, ST, PDA, PDE, PLY, U) provided P-AL concentrations similar to or slightly below P3, particularly in the low-P soil. WEP dynamics indicated that ST and CS provided higher readily available P than P3 in the early stages, but this advantage diminished over time. Struvite (ST) provided high initial WEP but lower P-AL, suggesting slower release kinetics. Pelletized digestates (PDA, PDE) and urine granules (U) provided substantial N but induced pronounced soil acidification, which may limit their suitability in low-pH soils. Biochar (BCS) and compost (CM) showed P availability similar to the mineral P reference but with minimal N contribution and lower microbial stimulation. These findings highlight the importance of matching RNF selection to soil type, crop N and P requirements, and soil pH management strategies.

### 4.1.2 Evaluation centre in Finland (LUKE)

#### Soil

A sandy soil with low organic matter content and a long history of use in incubation and pot studies was used. The soil had a particle-size distribution of 80.0% sand, 17.7% silt, and 6.3% clay. The organic carbon content was approximately 1.7%. The inorganic N content has varied between patches from 1 to 50 mg/kg, depending on the time of soil collection and the duration of storage. The soil pH in water extracts has varied from 4.8 to 5.8. Soil physical determinations have shown that the saturated water content is approximately 40%, the pF2 value is 25%, and the wilting point (pF4.2) is 4–7%. The soil has a relatively high plant-available phosphorus content and a low plant-available potassium content, based on Finnish soil fertility analyses using acid ammonium acetate extraction. After the soil was screened and the plant debris was removed, 100 g of dry-matter soil was weighed into each incubation pot.

#### Treatment

Organic treatments included the solid fraction of cow slurry digestate as such, same solid digestate with two rates of biochar addition (1% and 3%), meat and bone meal in pellet form, and ground meat and bone meal pellets (Table 4). All fertilisers were intended to be applied at a rate corresponding to 140 mg N  $\text{kg}^{-1}$  soil. However, because of differences in the pre-analysed N contents of the digestates, the actual application rates corresponded to only 100 mg N  $\text{kg}^{-1}$  soil. Phosphorus additions ranged from 0 to 43 mg P  $\text{kg}^{-1}$  soil. All treatments were compared with an unfertilized control and two inorganic fertilisers: calcium ammonium nitrate (27% N) and an NPK (22-5-5) compound fertiliser applied at the same rate of 140 mg N  $\text{kg}^{-1}$  soil. Due to the low pH of the test soil, 150 mg  $\text{Ca}(\text{OH})_2$  was mixed with the soil in each pot. Three replicates were prepared for each sampling time.

**Table 4 Total nitrogen (N), phosphorus (P), and carbon (C) contents of the different RNFs on a dry-matter basis**

Treatment, abbreviation	Original material	P <sub>tot</sub> , mg/g	N <sub>tot</sub> mg/g	C <sub>tot</sub> , mg/g	pH
Ground MBM (G-MBM)	Meat and bone meal (MBM)	28.9	99.2	450.3	5.9
Pelletized MBM (P-MBM)	Meat and bone meal (MBM)	28.9	99.2	450.3	5.9
Digestate (D)	Digested cow slurry	10.3	29.3	447.3	8.7
Digestate + 1% BC (1%-D)	Digested and separated solid from cow slurry with 1% biochar	8.9	25.6	518.8	8.6
Digestate + 3% BC (3%-D)	Digested and separated solid from cow slurry with 3% biochar	6.9	20.5	610.7	8.7
NPK-fertiliser (NPK)	NPK-fertiliser (22-5-5)	50	220	0	-
N-fertiliser (N)	Ammonium nitrate fertiliser (N27)	0	270	0	-

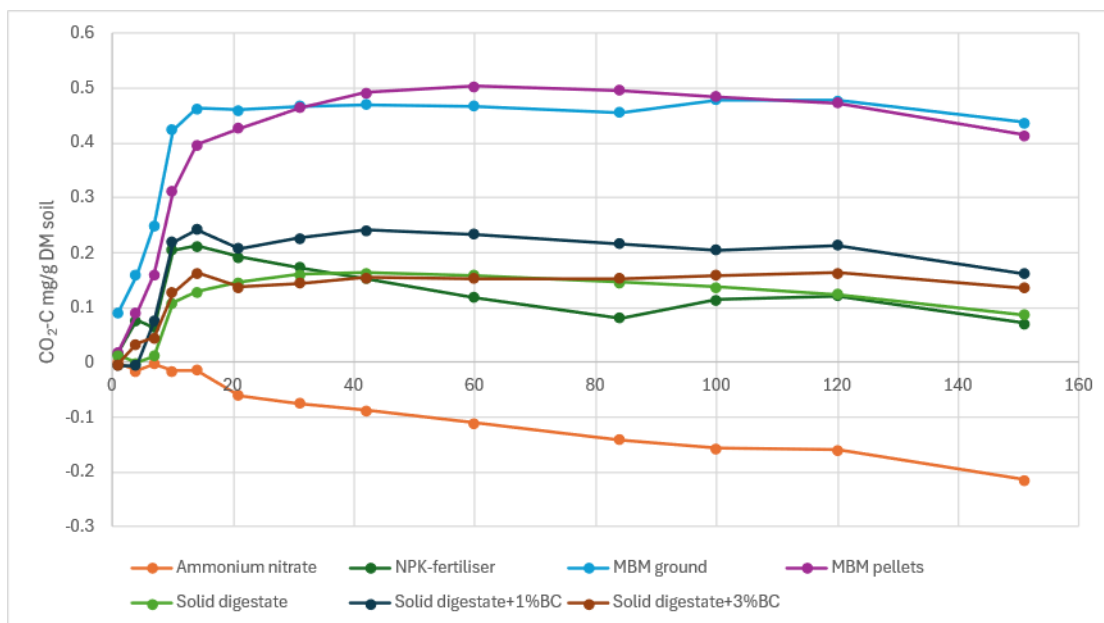
### **Incubation set up**

Incubation was carried out for 150 days at 20 °C according to the experimental protocol. Soil moisture was maintained at 50% of the water-holding capacity (WHC). During the incubation period, soil pH, mineral nitrogen, and phosphorus concentrations extractable with acid ammonium acetate were measured. Soil respiration was assessed by trapping CO<sub>2</sub> emissions in 10 mL of 1 M NaOH. Soil samples and traps were placed in airtight two-liter jars, and oxygen was replenished during each replacement of the traps. The CO<sub>2</sub> concentration in the NaOH solution was subsequently determined by titration.

### **Results**

#### **CO<sub>2</sub>-emission**

The proportion of added carbon emitted from the meat and bone meal treatments increased to 70% within 15 days. Ground meat and bone meal resulted in slightly faster carbon release than the pellets (Figure 5). However, after 30 days, the differences between the ground material and the original pellets had disappeared. In the digestate treatments, approximately 10% of the added carbon was released within 15 days, after which there was practically no further increase.

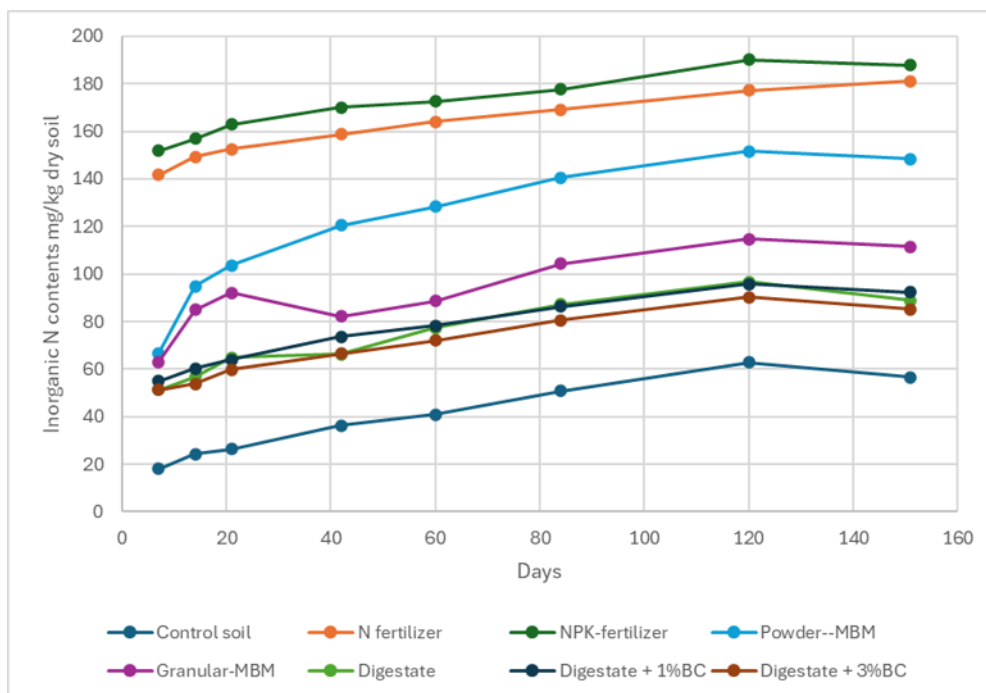


**Figure 5** Accumulated carbon emissions from different RNFs, with soil respiration of control soil deducted to illustrate the effect attributable to each RNFs.

### Inorganic N in soil

Net N mineralization in the control soil was approximately 40 mg kg<sup>-1</sup> soil during the incubation study (Figure 6). The digestates added approximately 30 mg kg<sup>-1</sup> soil of ammonium-N, which rapidly nitrified to nitrate. During the remainder of the incubation period, there appeared to be no further net mineralization. The meat and bone meal treatments initially added approximately 45 mg kg<sup>-1</sup> soil of ammonium-N. Thereafter, mineralization from the ground meat and bone meal continued slowly until the end of the incubation period. In contrast, the pelletized meat and bone meal exhibited a period of immobilization between 21 and 42 days, after which slow mineralization resumed. At the end of the incubation, more than 60% of the total N added in the ground meat and bone meal treatment had been released as inorganic soil N, whereas only 39% was released from the pellets. Inorganic fertilisers maintained constant levels of inorganic soil N, suggesting that no gaseous N losses occurred.

Table 5 shows the calculated mineralization indicators. Net organic N mineralization occurred only in the meat and bone meal treatments, and mineralization from the ground meat and bone meal was clearly higher than that from the pellets. The digestate and inorganic fertiliser treatments resulted in lower inorganic soil N contents than would have been expected based on their initial inorganic N contents. This may indicate greater N mineralization in the unfertilized control soil, which had a low inorganic N content, compared with the inorganic fertiliser treatments, or immobilization of inorganic N in the digestate treatments.



**Figure 6** Soil inorganic nitrogen (sum of ammonium-N and nitrate-N) following the application of different fertiliser products

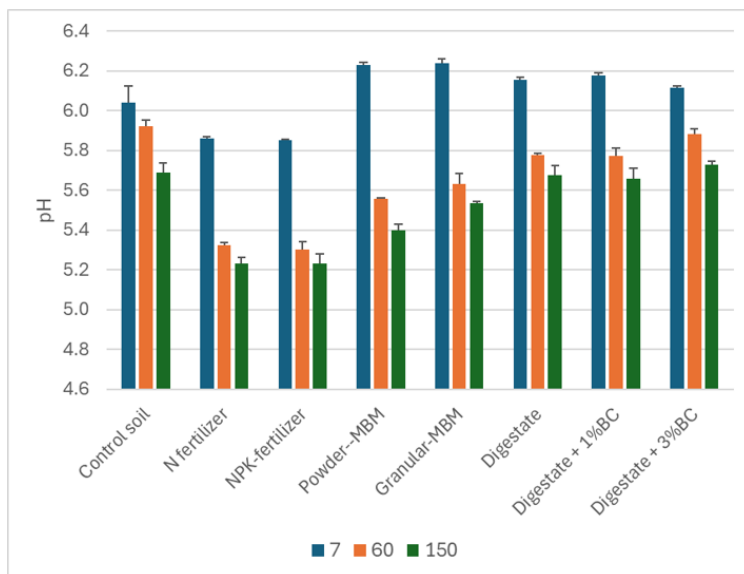
**Table 5** Amounts of inorganic N (*N<sub>inorg</sub>*), organic N (*N<sub>org</sub>*), and total N (*N<sub>tot</sub>*) applied to the soil in the pot incubation experiment, together with the estimated mineralisation of fertiliser organic N (*N<sub>m</sub>*) and the release of total fertiliser N (*N<sub>rel</sub>*).

Treatment	N inorg	Norg	Ntot	Nm	Nrel
	mg/kg			%	
N fertiliser	143	0	143		-13
NPK-fertiliser	143	0	143		-8
Ground--MBM	1	139	140	65	65
Pellet-MBM	1	139	140	39	39
Digestate	46	59	105	-23	-13
Digestate + 1%BC	44	61	105	-13	-8
Digestate + 3%BC	40	55	95	-21	-12

### Soil pH and plant available phosphorus

Soil pH decreased following the application of the inorganic N and NPK fertilisers (**Помилка! Джерело посилання не знайдено.**). At the beginning of the incubation period, meat and bone meal additions increased soil pH compared with the unamended control soil. However, after 60 days, soil pH in the meat and bone meal-treated pots decreased to levels lower than those in the control and digestate treatments. At the end of the incubation, the inorganic fertiliser and ground meat and bone meal treatments resulted in lower soil pH values than the control and digestate treatments. Overall, soil pH decreased over time, from 6.1 after 7 days to 5.5 after 150 days. Across the entire incubation period, the inorganic fertiliser treatments resulted in an average soil pH of 5.5, the ground meat and bone meal treatment in 5.7, whereas the digestate and control

treatments averaged 5.9.



**Figure 7** Soil pH following the application of different fertilizer products at 7, 60, and 150 days of incubation

Plant-available soil phosphorus, measured using acid ammonium acetate extraction, was close to 10 mg L<sup>-1</sup> in the control soil and in the treatment containing only ammonium nitrate fertiliser

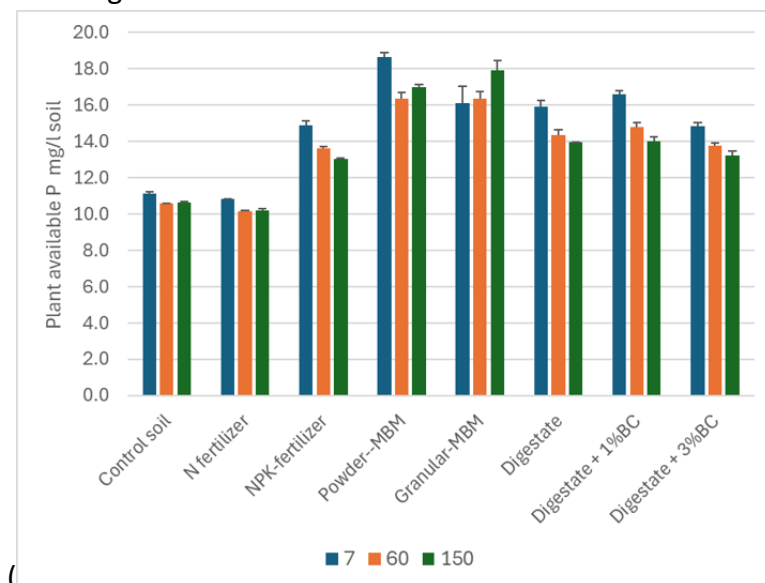
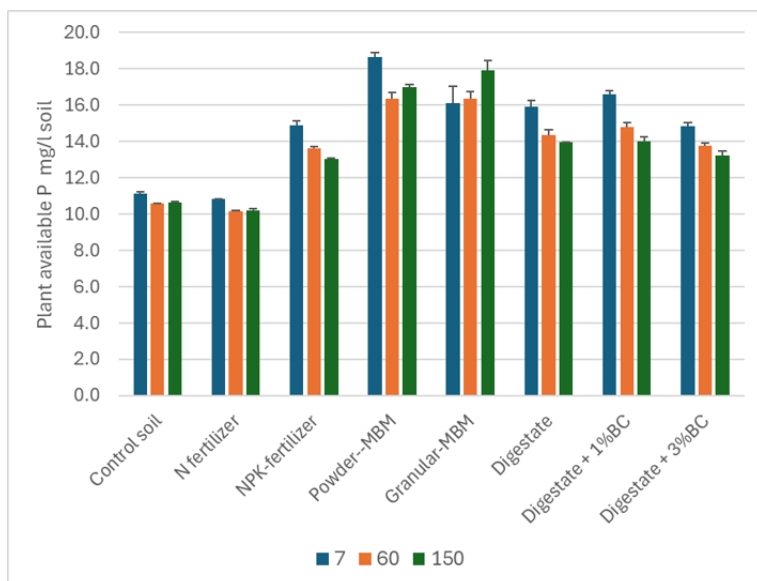


Figure 8). Application of P in the inorganic NPK fertiliser treatment (32 mg P kg<sup>-1</sup> soil) increased the soil P value to 15 mg L<sup>-1</sup>. Phosphorus application rates in the meat and bone meal treatments were 43 mg P kg<sup>-1</sup> soil, whereas those in the digestate treatments ranged from 31 to 36 mg P kg<sup>-1</sup> soil. The meat and bone meal treatments increased soil P values to up to 18 mg L<sup>-1</sup>, while the digestate treatments resulted in values of 15–16 mg L<sup>-1</sup>. In all treatments except the meat and bone meal pellets, soil P values decreased slightly during the incubation period, implying fixation of soluble P during the 150-day incubation.



**Figure 8 Plant-available soil phosphorus following the application of different fertiliser products at 7, 60, and 150 days of incubation.**

### Conclusions

The pot incubation experiment effectively described the differences among the tested soil improvers (digestates) and both inorganic and organic fertilisers. Because interfering effects were minimized, the results were primarily influenced by carbon and nitrogen mineralization and the subsequent nitrification of ammonium to nitrate. Although soil pH is not commonly measured in incubation studies, soil reaction provides useful information for understanding the effects of applied fertiliser products on pH, which may also influence mineralization and phosphorus fixation. Regarding plant-available soil phosphorus, the test soil naturally contained a satisfactory P level; therefore, it was not suitable for evaluating the phosphorus use efficiency of fertiliser products. Nevertheless, the effects of the fertiliser products on soil P status represent valuable information that can be obtained from pot incubation experiments at relatively little additional cost.

## 4.2 Pot trials

### 4.2.1 Evaluation centre in Sweden (SLU)

#### Soil

The soil described in Section 4.1.1 was used for the pot experiment, and its selection and preparation followed the established protocol.

#### Treatment

For the experiment, RNFs described in section 4.1.1 were used, except for dry urine and pellets obtained from mixed feedstock. The experiment was set up using the same doses of phosphorus with RNFs at a level of 45 mg P kg<sup>-1</sup> soil and compare with soil without P fertilization and

phosphorus mineral fertiliser application (triple superphosphate). Additional treatments for low-P included mineral phosphorus at application rates of 11.25 mg P kg<sup>-1</sup> (P1), 22.5 mg P kg<sup>-1</sup> (P2), 45 mg P kg<sup>-1</sup> (P3), 90 mg P kg<sup>-1</sup> (P4), and 135 mg P kg<sup>-1</sup> (P5), corresponding to 25%, 50%, 100%, 200%, and 300% of the phosphorus applied through RNF. A small amount of soil was first mixed with the respective fertiliser and subsequently homogenized with the entire soil mass in the pot. Before application, the RNF and TSP was ground and sieved through a 1 mm sieve.

### **Experiment set up**

The experiment used perennial ryegrass (*Lolium perenne* L.), variety Sirtaky. After fertilization, 67 seeds of perennial ryegrass seeds (rate is 1 seed per 4 cm<sup>2</sup>, with a 1000-seed weight of 1,48 g and germination 95%) was sown per pot in 3 rows and covered with a thin layer of soil (~0.5 cm, ~300 g). Grass was harvested on days 30, 50, and 75 of the growing seasons. The pots were housed in a controlled growth chamber with a 16-hour light/8-hour dark photoperiod and a temperature regime of 20-22 °C during the day and 12-14 °C at night. Plant growth was measured based on forage yield as well as nutrient content in the biomass. After harvesting mineral N, P-AL and pH was measured.

### **Results**

Dry matter (DM) yield of perennial ryegrass responded strongly to phosphorus supply and fertiliser type in both soil types (Figure 9).

In the low-P soil, ryegrass yield clearly reflected differences in phosphorus availability among the RNFs and the mineral reference treatment P3 (45 mg P kg<sup>-1</sup>). At this P level, compost (CM) and struvite (ST) achieved yields comparable to P3, indicating that these RNFs were able to supply plant-available P at a level similar to mineral fertilizer. In contrast, PDA pellets outperformed P3, forming a higher statistical group and demonstrating superior P availability under P-limiting conditions that can be connected to additional supplement of nutrients after mineralisation. Treatments with CS, PDE, and BCS produced lower cumulative yields than P3, with CS showing the lowest yield among the RNFs, suggesting restricted or delayed P release from this material.

The distribution of yield across cuts further highlights differences in P release dynamics. In CS, PDE, and BCS, the first harvest contributed the lowest share of total yield, indicating limited early P availability and a delayed plant response. By contrast, compost and struvite showed similar yield levels across harvests, suggesting a more even P supply over time. Notably, in PDE and PDA pellet treatments, the final harvest showed the highest values, pointing to a strong late-season P availability, particularly for PDE, and a combination of early and sustained release for PDA. Overall, these patterns indicate that while some RNFs (CM, ST, PDA) can match or exceed mineral P at the same dose, others (CS, PDE, BCS) provide less effective or less timely P supply in low-P soil.

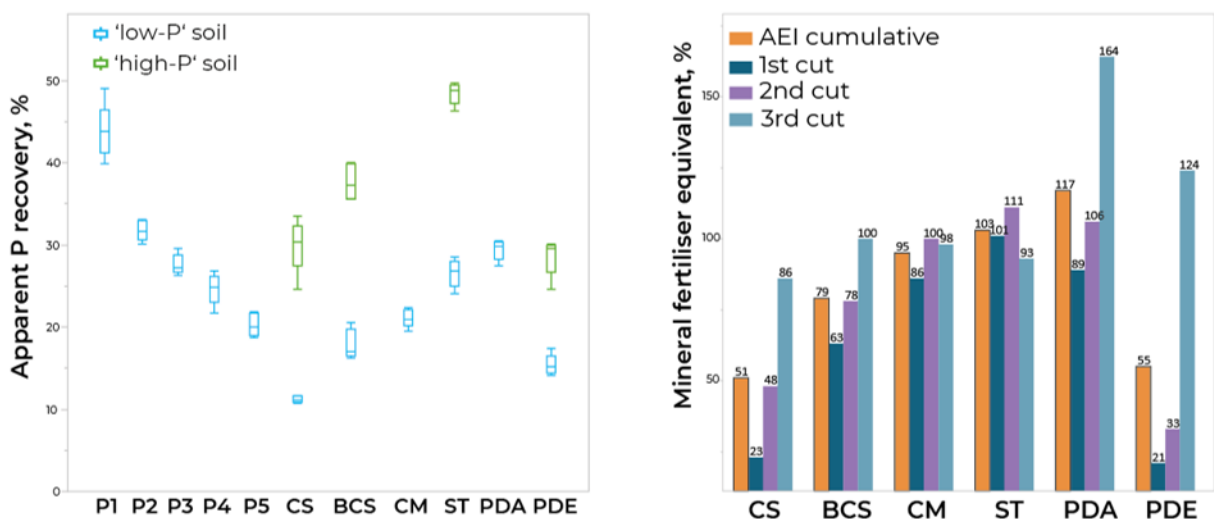
Apparent P recovery (APR) differed between soil types and among treatments (Figure 9). In the low-P soil, APR values showed a clear decline with increasing mineral P application rate. This pattern is expected under strong P limitation: when small amounts of P are applied, plants utilise a larger proportion of the added P, whereas higher doses exceed immediate plant demand and

therefore reduce the calculated recovery percentage.

At the reference level of 45 mg P kg<sup>-1</sup> (P3), APR values were statistically equivalent to those observed for PDA pellets and struvite (ST). This indicates that both RNFs supplied plant-available P at a rate comparable to mineral fertilizer under P-deficient conditions. In contrast, the lowest APR value was recorded for CS, suggesting limited P solubility or slow release from this material. Biochar-based digestate (BCS) and compost (CM) formed a statistically similar group, indicating comparable P availability from these two materials. Interestingly, BCS and PDE were also statistically indistinguishable, showing that PDE did not provide higher P recovery than BCS despite differences in material composition. These overlaps in statistical groupings highlight that several RNFs released P at similar rates, but only PDA and ST matched the mineral reference in terms of recovery efficiency.

Overall, the APR pattern in the low-P soil reflects the strong influence of fertilizer P solubility: materials with rapid or moderate P release (PDA, ST, CM, BCS) achieved higher recovery, while those with slower release (CS, PDE) showed reduced utilisation by plants.

In the high-P soil, APR values were generally higher than in the low-P soil. This may appear counterintuitive, but it reflects the fact that plants in this soil had better overall growth and root development, enabling more efficient uptake of applied P even when background P levels were elevated. Among the RNFs, struvite (ST) showed the highest APR, confirming its well-documented ability to release P in a plant-available but controlled manner. BCS ranked second, followed by CS and PDE, which showed lower but still measurable recovery. The relative ranking of RNFs in this soil suggests that the chemical form of P and the release kinetics remained important even when soil P availability was higher. The consistently strong performance of struvite across both soils highlights its suitability as a slow-release P source that aligns well with plant uptake patterns. The moderate performance of BCS suggests partial P availability, while the lower APR of CS and PDE indicates slower or less complete P solubilisation.



**Figure 9** Apparent phosphorus recovery (APR) in ryegrass shoots grown on “low-P” and “high-P” soils and amended with RNFs and TSF. Error bars represent standard error of the mean (n = 5). Mineral fertilizer equivalent of RNFs compared with TSF (P3) at equal application rates, showing crop response across three harvest cuts.

Because MFE integrates both yield response and P uptake efficiency, it effectively summarises the patterns observed in the previous two indicators (dry matter yield and APR). The MFE values therefore provide a holistic assessment of how well each RNF supplied plant-available phosphorus compared with the mineral reference (P3).

The cumulative MFE for struvite reached 103%, demonstrating that it performed as mineral fertiliser (Figure 9). Across all three harvests, ST ensured a nearly identical temporal pattern of P release compared with TSP. This confirms the suitability of ST as a slow-release P source that synchronises well with plant demand throughout the growing season.

Pellets PDA achieved the highest cumulative MFE value – 117%, outperforming all other RNFs and the mineral reference. Interestingly, the first and second cuts showed MFE values almost identical to compost, indicating moderate early-season P availability. However, the third cut displayed a substantial increase, exceeding the performance of P3. This pattern suggests that PDA releases P more slowly at the beginning of the season but becomes highly effective after approximately 50 days, when its nutrients become more available. Such delayed release may be advantageous for crops with extended growth periods.

Compost also showed high cumulative MFE (95%), placing it close to the mineral reference. Its performance across harvests was stable, indicating a steady and moderate P release. Digestate CS and pellets PDE had similar cumulative MFE values (51% and 55%, respectively), indicating lower overall effectiveness compared with other RNFs. However, their seasonal dynamics differed substantially. At the first cut, both treatments showed nearly identical MFE values (23% for CS and 21% for PDE), reflecting limited early P availability. By the end of the season, PDE showed a strong improvement, reaching 124%, whereas CS reached only 86%. This indicates that PDE pellets released P more slowly but eventually provided a much higher late-season availability than CS.

BCS achieved a cumulative MFE of 79%, placing it in the mid-range among RNFs. Its MFE increased steadily across harvests, rising from 63% to 100%. This gradual improvement suggests that biochar-based materials release P progressively, contributing more effectively as the season advances.

## **Conclusions**

This study demonstrates that recycled nutrient fertilisers (RNFs) can provide plant-available phosphorus at levels comparable to, and in some cases exceeding, mineral phosphorus fertiliser. In the low-P soil, where phosphorus limitation was pronounced, clear differences emerged among RNFs. PDA pellets, compost (CM), and struvite (ST) consistently matched or outperformed the mineral reference (P3), indicating that these materials can effectively substitute mineral P at equivalent application rates. PDA showed the strongest overall performance, with delayed but ultimately high P availability, while struvite provided a release pattern closely aligned with mineral fertiliser. Compost also performed reliably, offering steady P supply throughout the season. In contrast, CS, PDE, and BCS showed limited early P availability and lower overall effectiveness, although PDE demonstrated substantial late-season improvement.

In the high-P soil, differences among RNFs were less pronounced due to the buffering effect of

higher background P availability. Nevertheless, struvite again showed the highest APR, followed by BCS, CS, and PDE, confirming that P release kinetics remained relevant even when soil P was not limiting. The consistent performance of struvite across both soils highlights its suitability as a controlled-release P source.

Taken together, the results show that RNFs differ not only in the total amount of P they supply but also in the timing and synchrony of P release relative to crop demand. Materials such as PDA, struvite, and compost demonstrate strong potential as alternatives to mineral fertilisers, particularly in P-deficient soils. Other RNFs may require optimisation or complementary management strategies to improve their agronomic effectiveness.

#### 4.2.2 Evaluation centre in Sweden (Linnæus University)

##### Soil

Two contrasting soils were used to test nutrient uptake from sewage sludge biochar derived from a facility operated in a complementary project (testbädd Ellinge). One soil was a moderately fertile soil from Högseröd (55.80, 13.58) and was collected at the edge of an agricultural field that had been only extensively managed for the past 25 years. The other soil was collected from the Southern Swedish highlands at Puketorp (57.2521319, 14.9018076) and was a grazing field that had been extensively managed for more than 45 years. The organic matter content was similar in the two soils: 20% in the poor soil and 21% in the moderately fertile soil. The cadmium concentrations were below the detection limit (<0.4 mg/g) in both soils. The amounts of AL-extractable nutrients (Table 6) were higher in the Högseröd soil than in the Puketorp soil, except for K, which was at a similar level in both soils. The HCl-extractable K was, however, much higher in the Högseröd soil than in the Puketorp soil, confirming the difference in fertility.

**Table 6 Water holding capacity (WHC), pH, organic matter content, and total elemental concentrations in soils (Högseröd- moderately fertile soil, Puketorp- poor soil) used to test the nutrient availability (N, P and K) in sewage sludge biochars.**

Soil	pH	WHC (% of dm)	OM%	C/N ratio	Total C %	Total N %	Total elemental content (mg/g dry weight soil)				
							P	Ca	Al	Fe	K
Högseröd 1	6,4	67	21	14	43	3,1	0,9	3,3	6,1	7,2	0,8
Puketorp 2	5,8	71	20	13	39	2,9	0,7	2,7	8,5	10,4	0,4
<b>Biochar</b>											
SME	6,4		26	15	52	3,4	2,2	4,5	6,9	8,2	0,8
ÖRE			23	15	47	3,1	2,7	5,1	7,5	9,0	1,0
KLA	7,4		32	16	63	4,1	4,0	6,7	6,9	12,5	1,0
MAR	7,1		49	18	97	5,5	6,8	8,3	9,7	17,4	1,2

## Treatment

The biochars tested were from four different sewage sludge plants in Varberg, Helsingborg, Malmö, and Åkersberga, Stockholm. The sludges have different characteristics, and their composition varies (Table 7). The sludge from Helsingborg (ÖRE) is produced through biological P fixation, the sludge from SME through aluminium-based P fixation, the sludge from Malmö (KLA) through iron-based P fixation, and the sludge from Åkersberga (MAR) is undigested, as opposed to the others, which are digested. The biochars had similar surface area (BET, SME=128, ÖRE=101, KLA=110, MAR=133 m<sup>2</sup> g soil<sup>-1</sup>) and water-holding capacity. The extractable nutrient levels in undigested sludge biochar, MAR, were generally higher than in the other sludge biochars (Table 8), apart from Mg, which was slightly lower.

**Table 7 Properties of the sludge biochars (SME- Varberg sewage plant, ÖRE - Helsingborg, KLA - Malmö, and MAR- Åkersberga Stockholm) that was tested for P uptake by maize grown in pots**

Parameter		Biochar			
		SME	ÖRE	KLA	MAR
DW	%	92,6	80,1	90,8	88,5
LOI	% DW	12,7	18,1	18,2	22,1
pH (H <sub>2</sub> O)		8,2	8,1	8,2	8,4
H	mg/kg DW	0,5	0,5	0,6	1,1
C	% DW	27,2	33,8	29,0	33,9
H/C		0,018	0,015	0,021	0,032
TOC	% DW	27,1	33,7	28,9	33,8
Ash content	% av DW	72,33	66,31	74,69	63,95
N	% DW	1,34	1,97	1,56	1,85
P	mg/g soil	66	62	89	67
K	mg/g soil	5,9	11	5,6	7
S	mg/g soil	7,95	11,52	17	8,8
Mg	mg/g soil	7,7	16	14	5,2
Al	mg/g soil	92	34	15	45
Fe	mg/g soil	45	62	170	120
Ca	mg/g soil	44	52	70	49
Cd	µg/g soil	0,043	0,15	0,082	0,14

**Table 8 Extractable (AL and 0.5 M HCl) in soils (Högseröd- moderately fertile soil, Puketorp- poor soil) used to test the nutrient availability (N, P and K) in sewage sludge biochars**

Soil	AL-extraction (mg/g dw soil)				HCL extraction (mg/g dw soil)		
	P	K	Mg	Ca	P	K	Cu
Högseröd 1	0,28	0,03	0,20	1,95	1,05	0,68	0,14
Puketorp 2	0,04	0,03	0,05	0,30	0,67	0,27	0,08
Biochar							
SME	0,46	0,03	0,26	2,14	5,26	0,89	0,11
ÖRE	0,41	0,03	0,28	2,23	3,22	0,96	0,14

Soil	AL-extraction (mg/g dw soil)				HCL extraction (mg/g dw soil)		
	P	K	Mg	Ca	P	K	Cu
KLA	0,46	0,04	0,31	2,90	3,18	0,88	0,17
MAR	0,66	0,05	0,27	3,23	8,34	1,22	0,44

Soil and biochars were mixed to meet the required goal of 50 mg P kg soil<sup>-1</sup> and compensated for different densities and moisture content. This resulted in a mixture of 4 dl SME and ÖRE biochars per 4 L pot, 3,5 dL KLA biochar and 8 dL of MAR biochar per 4 L pot. One control treatment with un-fertilized soil was included, one superphosphate fertilized pot as a reference, and one struvite treatment.

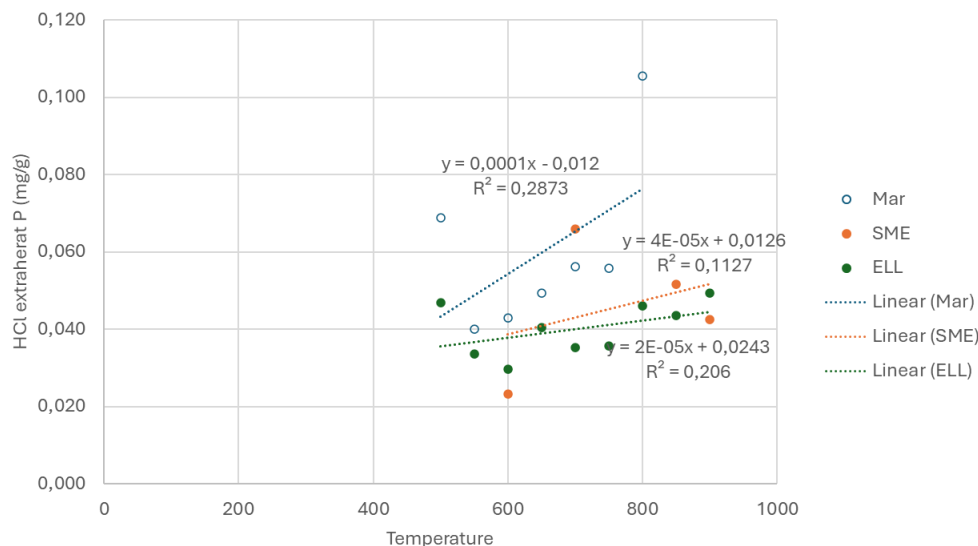
### **Experiment set up**

Maize seeds were pre-germinated for four days between moist paper and one viable seedling was planted in 4 L pots filled with soil mixtures of the different treatment. Each soil treatment was replicated in eight pots. The pots were watered to remain at approximately 25% moisture throughout the experiment regardless of soil mixture and this was calibrated once a week using a soil moisture meter and the EC was determined on one occasion (W.E.T Sensor kit, type HH2, Delta-T Devices Ltd, UK). The plants were kept in a greenhouse for 7-8 weeks, until they started producing flowers when they were harvested in the stems, leaves, and flowers apart. The plant biomass was dried at 70 C for 5 days until they were fully dried. The dry weight of the plant parts was determined gravimetrically.

### **Results**

#### **Evaluation of P extraction methods**

Methods to sample sludge biochar and methods to determine available P were modified from known soil P methods (Figure 10). The methods were evaluated, the precision was tested, and the methods were compared against each other. We could determine P in the biochars with high precision and accuracy (variation among replicas HCL 4-7%, Olsen P 18-21 %, Oxalate 8-11%). We decided to continue with the HCL and oxalate extraction methods. We found that the correlation between these methods and the Olsen P was high. The variation in extractable P was related to the temperature in different ways depending on the original plants and biochars from the different temperature batches was kept in the analysis. The variation between batches of sewage sludge from the same sewage plants was high (more than 50%).



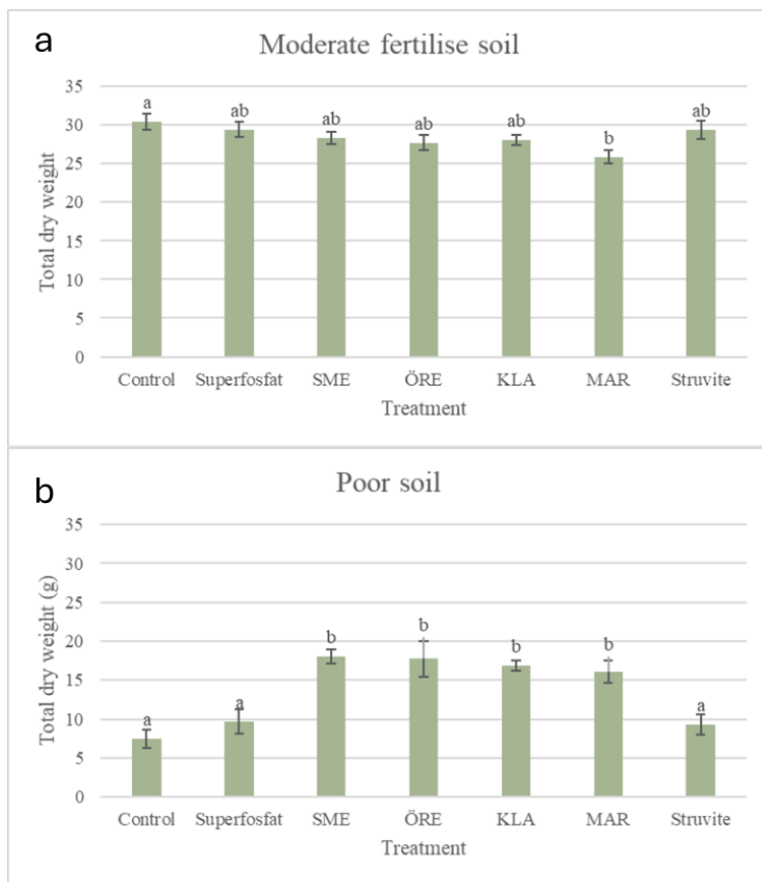
**Figure 10. The variation in extractable P related to the pyrolysis temperature and for three different sludge biochars.**

### Pot experiment

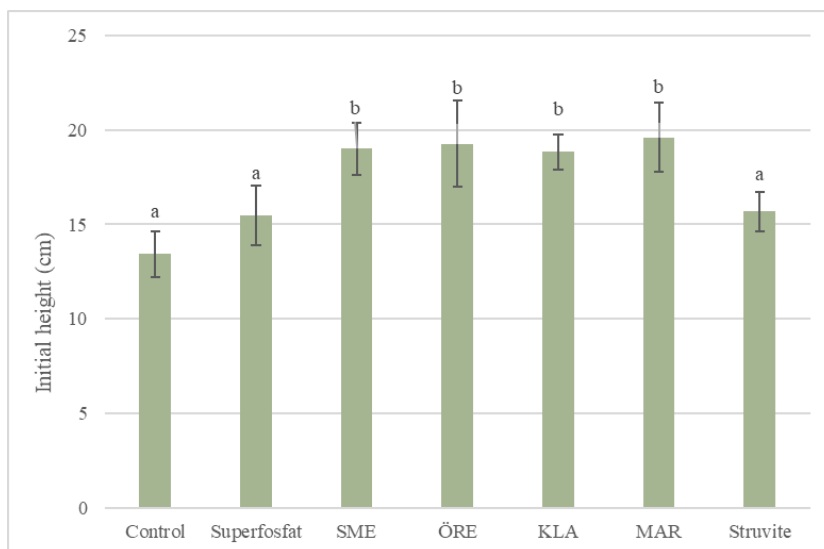
The dry weight of the plants differed substantially between the different soils. The moderately fertilise soil supported higher biomass than the poor soil. The addition of sludge biochars to the poor soil doubled the growth of the maize plants, and the onset of flowers was earlier.

The growth rate was not different among the different treatments in the moderately fertile soil, but there was a difference in plant size after two months of growth. In the fertile soil the growth of the maize plants was the same regardless of soil treatment or was reduced slightly with the MAR sludge biochar as compared to the control (Figure 11, ANOVA  $p < 0.01$ ). Struvite and superphosphate fertilised plants did not grow better than the control and the sludge biochar treated plants.

The sludge biochars had a significant positive effect on the growth of the maize in the poor soil; the biomass was almost doubled (Figure 11 b, ANOVA,  $p < 0.001$ ) at the harvest. However, the plants that received struvite or superphosphate ended up with similar biomass as the control. The effect of the sludge biochars was rapid and the differences in growth presented after 15 days of cultivation (Figure 12, GLM,  $p < 0.001$ ) and the growth rates were more rapid in the biochar treated plants.



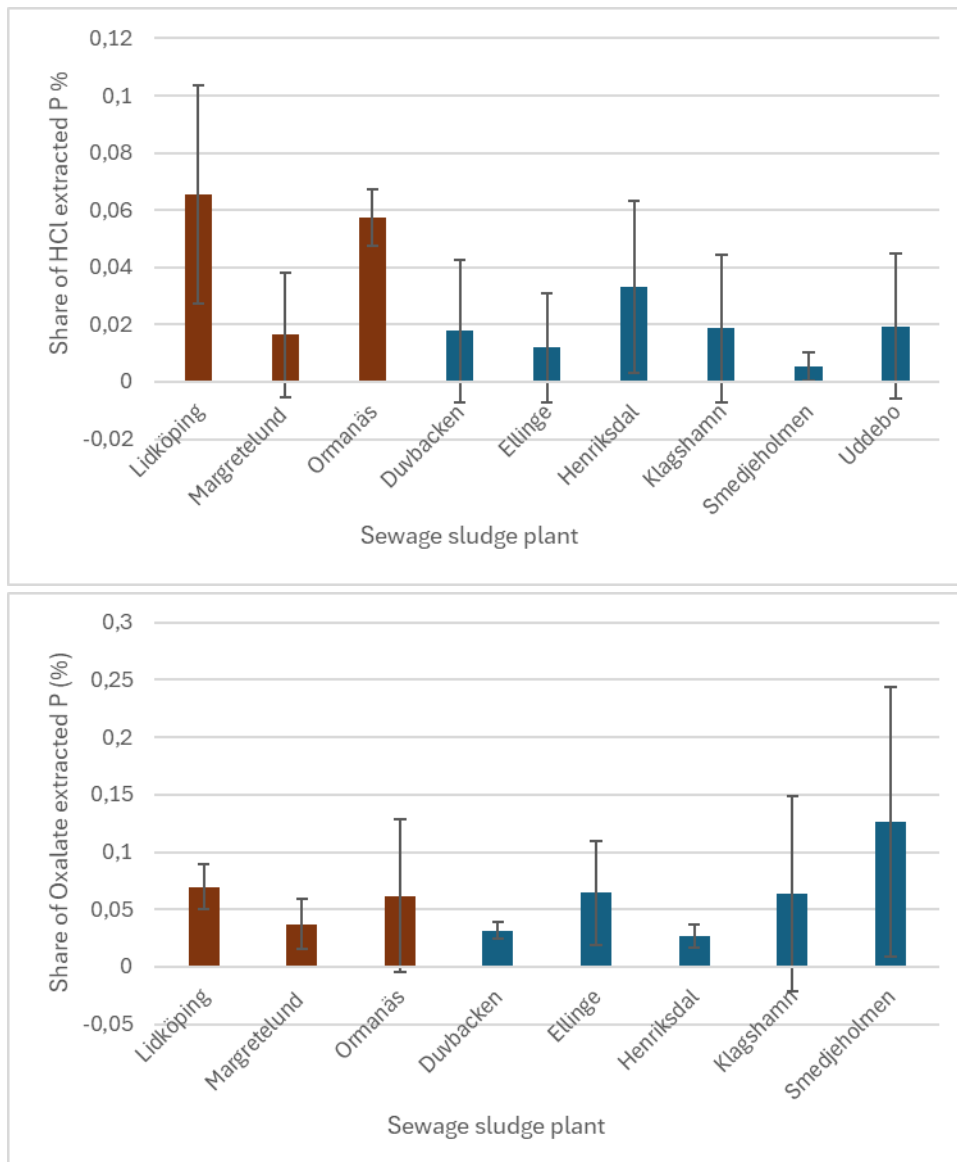
**Figure 11** The biomass of maize grown in two different soils; moderate fertile (a) and poor (b) with addition of P fertilisers (super phosphate and struvite) and sludge biochars. SME uses Al to precipitate phosphorus, ÖRE uses biological P precipitation, KLA uses Fe and MAR is an undigested sludge.



**Figure 12** Shoot height of maize grown with addition of different sewage sludge biochars and P fertilisers in a poor sandy silt (Puketorp) after 15 days of growth.

Several different sludge biochars have been evaluated for their P availability (Figure 13). The share of extractable P with two methods in relation to the total P in the sludge biochars was evaluated. The potentially available P in the biochars is very low, less than 0.2%. The share of HCl

extractable P availability in the sludges was higher in the undigested sludge biochars than in the biochars from digested sludge. The number of sewage sludge plants with undigested sludge is however low, and the results should be viewed as an indicator, and underlying factors may be more important. The share of oxalate extractable P in relation to the total P in the sludge biochars was more variable and no reliable differences was found.



**Figure 13** The mean share of HCl extractable P (top) and oxalate extractable P (bottom) in relation to the total amount of P in sludge biochars from different sewage plants across Sweden. Brown bars indicate biochars from undigested sludge blue digested sludge. Error bars represent standard deviation.

The P extracted with HCl and oxalate represents different fractions in the biochars. The HCl P is positively correlated to the total aluminium content of the biochars and slightly negatively correlated to the magnesium content (Помилка! Джерело посилання не знайдено.). A multiple regression of the share of HCl extractable P includes the total Al, total potassium and pyrolysis temperature in the model (forward stepwise regression,  $r^2=0.32$ ,  $p>0.001$ ). The oxalate extractable P is positively correlated to the water holding capacity and to the total potassium

content of the biochars and negatively correlated to the pH and total Fe content (Figure 14 **Помилка! Джерело посилання не знайдено.**). Stepwise multiple regression of the share of oxalate extractable P includes total potassium, and total iron in the biochar in the model (forward  $r^2=0.36$ ,  $p>0.001$ ). Neither is related to the total P content. The total P content is positively correlated to the total calcium and magnesium content and negatively correlated to the aluminium content of the biochars.

	HCLP	OX P	WHC	pH	Ash content	Ntot	Ptot	Ktot	Mgtot	Altot	Fetot	Catot		
HCLP	1	.077	.084	-.058	.193	-.122	-.251	-.218	-.356*	.451**	-.104	-.265		
OX P	.607	1	.532**	-.460**	.128	-.021	.165	.586**	.172	.362*	-.530**	.034		
WHC	<.001	.001	1	-.432**	.132	.115	-.049	.411**	-.027	.344*	-.665**	-.073		
pH	.002	.378	.443	1	.378	.443	.743	.004	.057	.018	<.001	.625		
Ash content					1	-.154	-.172	.189	-.530**	.176	-.494**	.475**		
Ntot						1	-.024	-.168	-.049	-.163	.322*	-.248		
Ptot							1	-.014	.817**	-.613**	.343*	.920**		
Ktot								1	.289*	.222	.520**	-.145		
Mgtot									1	.530**	.072	.817**		
Altot										1	-.727**	-.575**		
Fetot											1	.283		
												1		
													1	
														1

**Figure 14 Pearson correlation (r- and p-values) of extractable P concentrations (HCL P and Ox P) and various properties of the sludge biochars.**

### Conclusions

Sludge biochars have a significant positive effect on maize growth on poor soils. P fertilisation with either superphosphate or struvite did not result in similar growth effects. The effect is rapid and differences are initial but also the growth rate is higher with biochar amendments.

The four different sludge biochars affected the plant growth in a similar way. The P fixation method in the sewage plants did not seem to have any effect on the maize growth. The effect is connected to some parameter in the sludges that are similar in all the biochars like porosity, specific surface area or water holding capacity. The lower growth of the plants with the MAR sludge biochar might have been a result of the higher amount of biochar added in this treatment to reach the same P level also indicate that the effects were attributed to another factor than P content.

A very small share of the P in the sludge biochars is extractable with weak HCl and oxalate solutions. This indicates that the value of sludge biochars as a P fertiliser is limited. The two methods extract different fractions of the P, but both result in similarly low levels. The share of extractable P is related to the total potassium content of the sludge biochars, and this might be an important factor to consider. Aluminium content in the sludge was an element that improved the share of HCl-extractable P, and it seems that iron reduces the share of extractable P. However, the explanatory power is low, so there are other factors more important than the ones we studied, or the accuracy of the measurements is low.

### 4.2.3 Evaluation centre in Finland (LUKE)

#### *Soil*

The soil described in Section 4.1.2 was used in the pot experiment, and its selection and preparation followed the established protocol.

#### *Treatment*

For the pot experiment, the RNFs consisted of untreated digestate, liquid and solid fractions of digestate, and meat and bone meal either in pellet form or ground from pellets. The experiment was established using a uniform nitrogen application rate of 250 mg N kg<sup>-1</sup> soil for the RNF treatments and was compared with an unfertilized control and inorganic nitrogen fertiliser treatments (ammonium nitrate and calcium nitrate). Additional inorganic N treatments were applied at rates of 0, 83, 167, 250, and 333 mg N kg<sup>-1</sup> soil.

The actual total N additions from the RNFs were verified by analysing samples at the time of pot establishment. The largest deviation from the target application rate occurred in the digestate treatment, in which the actual N application rate was 287 mg N kg<sup>-1</sup> soil. The amounts of inorganic N applied with the RNFs were 2 mg kg<sup>-1</sup> soil for the meat and bone meal treatments, 139 mg kg<sup>-1</sup> soil for the untreated digestate, 158 mg kg<sup>-1</sup> soil for the liquid digestate fraction, and 57 mg kg<sup>-1</sup> soil for the solid digestate fraction. Lime was applied at a rate of 15 g per 6 kg of soil in each pot. Triple superphosphate was added at a rate of 1.5 g per pot (53 mg P kg<sup>-1</sup> soil), and other plant nutrients were supplied as nutrient solutions to ensure adequate nutrient availability.

The experiment was conducted with five replicates. The effect of replicate number on statistical error was evaluated by performing statistical analyses separately using combinations of three, four, or all five replicates.

#### *Experiment set up*

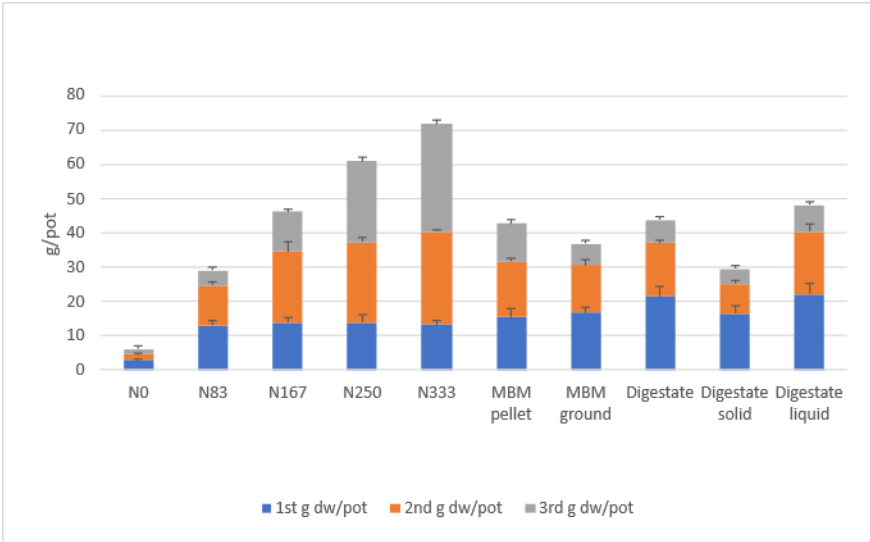
The experiment used Italian ryegrass (*Lolium multiflorum* L.), cultivar Barmultra. After fertilization, 98 ryegrass seeds were sown per pot, corresponding to a sowing density of one seed per 4 cm<sup>2</sup>. The 1,000-seed weight was 3.86 g, and the germination rate was 95%. The seeds were covered with a thin layer of soil (approximately 0.5 cm, corresponding to about 300 g of soil).

Grass was harvested on days 31, 52, and 97 of the growing period. The pots were maintained in a glasshouse with open walls, and the temperature varied according to seasonal conditions. The pots were placed on scales and watered three to four times per week, or daily during periods of maximum plant growth. Plant growth was assessed based on forage yield and nitrogen content in the biomass.

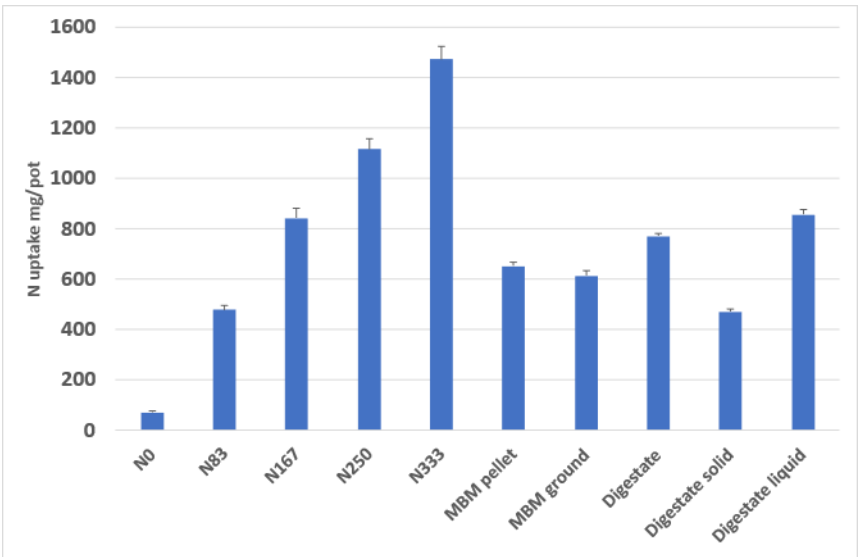
#### *Results*

Nitrogen response started to develop after the second harvest, and with three harvest the total yields separated each other (Figure 15). Solid digestate had the lowest yield of RNFs, and it corresponded to the inorganic reference N rate of 83 mg/kg soil. All RNFs yielded less than the

inorganic N control, N250. Liquid digestate produced better yield than ground meat bone meal and solid digestate.



**Figure 15** Effect of fertiliser treatments on the dry matter yield of ryegrass across three harvests. Error bars represent the standard deviation calculated from five replicates.



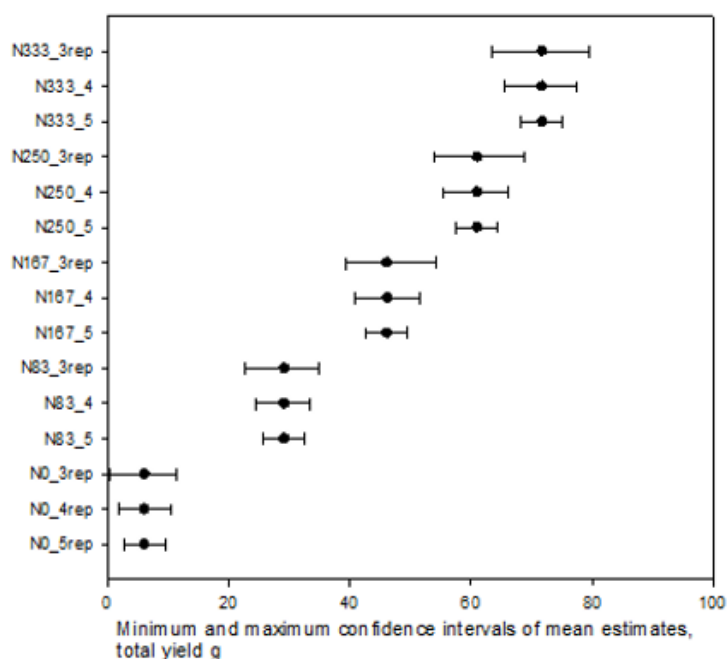
**Figure 16** Effect of fertiliser treatments on the nitrogen uptake of ryegrass. Error bars represent the standard deviation calculated from five replicates.

Nitrogen uptake in the solid digestate treatment was similar to that in the 83 mg N kg<sup>-1</sup> soil inorganic fertiliser treatment, whereas nitrogen uptake in the liquid digestate treatment was comparable to that in the 167 mg N kg<sup>-1</sup> soil treatment (Figure 16). The untreated digestate resulted in slightly higher nitrogen uptake than the meat and bone meal treatments, and the nitrogen uptake values for all three RNF treatments were intermediate between those of the N83 and N167 inorganic fertiliser treatments. Based on agronomic efficiency indicators, the liquid digestate showed the highest values, followed by the untreated digestate. The meat and bone meal treatments exhibited higher values than the solid digestate treatment.

**Table 9 Agronomic efficiency indicators, including nitrogen use efficiency (NUE), apparent nitrogen recovery (ANR), and mineral fertiliser equivalent (MFE), calculated from nitrogen uptake in the pot experiment**

Treatment, N mg/kg	N_added, mg/pot	N uptake, mg/pot	NUE	ANR	MFE_%
N0	0	71			
N83	500	479	0.96	0.82	
N167	1000	841	0.84	0.77	
N250	1500	1113	0.74	0.69	
N333	2000	1471	0.74	0.70	
MBM-pellet	1498	651	0.43	0.39	56
MBM-ground	1466	614	0.42	0.37	53
Digestate	1722	771	0.45	0.41	58
Digestate-solid	1469	470	0.32	0.27	39
Digestate-liquid	1432	856	0.60	0.55	79

The confidence limits became wider as the number of replicates decreased from five to four and three. As shown in Figure 17, the confidence limits of the two highest N application rates overlapped when three or four replicates were used, whereas with five replicates the confidence limits were separated. In contrast, the yields at the three lowest N application rates differed sufficiently such that their confidence limits did not overlap even when only three replicates were used.



**Figure 17 Effect of using three, four, or five replicates on the 95% confidence limits of treatment mean estimates for total yield calculated from three harvests.**

### Conclusions

Ryegrass proved to be a suitable test crop because of its efficient nutrient uptake capacity and

the possibility of obtaining several harvests, which helps to deplete soil nutrient reserves during the experiment. Increasing the number of replicates improved the ability to detect small differences between treatments. In particular, the variation in mean yields was relatively large when only three replicates were used.

The variability in nutrient concentrations of the RNFs highlights the importance of analysing both pre-samples and samples collected at the time of experiment establishment to verify the actual nutrient application rates. The digestate treatments produced yields corresponding largely to their inorganic N contents. The liquid digestate, which had the highest proportion of ammonium-N relative to total N, resulted in yield and N uptake comparable to those obtained with the 167 mg N kg<sup>-1</sup> soil inorganic fertiliser treatment, while the liquid digestate itself supplied 158 mg ammonium-N kg<sup>-1</sup> soil.

Meat and bone meal contained very little inorganic N, but its organic N was mineralized rapidly. Nevertheless, under the conditions of this pot experiment, the mineralization rate did not appear sufficient to support plant growth comparable to that achieved with inorganic N fertilization.

#### 4.2.4 Evaluation centre in Estonia (METK)

##### Soil

The pot experiment was conducted using arable soil collected from the topsoil layer (0–20 cm). The soil was classified as a silty loam with a low to moderate phosphorus status. Particle size analysis revealed a composition of 23.40% sand, 63.62% silt, and 12.98% clay, corresponding to a light to medium silty loam according to the national soil texture classification. Prior to the experiment, soil samples were analysed for chemical properties. Detailed soil characteristics before the trial are presented in Table 10.

**Table 10 Soil properties**

Soil	pH <sub>KCl</sub>	C <sub>org</sub> (EA), %	N <sub>tot</sub>	P*	K*	Ca*	Mg*	Cu*	B*	Mn*	Zn*	WHC
				mg/kg								%
Jõgeva	6.7	2.6	0.22	46	75	3542	203	2.69	0.72	38	1.3	33

\*Mehlich II

##### Treatment

For the experiment, struvite was used as a RNF in granular form and compared with mineral phosphorus fertilization. The experiment included a phosphorus-free control and a mineral phosphorus treatment as references, as well as four struvite treatments with increasing phosphorus application rates.

The phosphorus-free control treatment (C0) received no phosphorus but was supplied with all other essential nutrients in mineral form. The mineral phosphorus treatment (CP) received phosphorus as monocalcium phosphate (Ca(H<sub>2</sub>PO<sub>4</sub>)<sub>2</sub>) at a rate of 42 mg P kg<sup>-1</sup> dry soil, corresponding to 110 mg P per pot or 40 kg P ha<sup>-1</sup>.

Struvite was applied in granular form without grinding at four phosphorus levels: 17 mg P kg<sup>-1</sup>

dry soil (P1), 25 mg P kg<sup>-1</sup> dry soil (P2), 34 mg P kg<sup>-1</sup> dry soil (P3), and 42 mg P kg<sup>-1</sup> dry soil (P4). These application rates corresponded to 45, 65, 89, and 110 mg P per pot, or 15, 22, 30, and 40 kg P ha<sup>-1</sup>, respectively. The struvite contains 46 g N kg<sup>-1</sup>, 117 g P kg<sup>-1</sup>, 32 g C kg<sup>-1</sup>.

In all treatments, including the phosphorus-free control, nutrients other than phosphorus were supplied as mineral fertilisers in sufficient amounts to avoid limitation by other elements. This design ensured that differences in plant growth and phosphorus uptake could be attributed solely to the phosphorus source and its application rate.

### **Experiment set up**

Struvite and mineral fertilisers were applied one day before sowing according to the treatment design. Soil and fertilisers were thoroughly mixed, and all pots were watered to approximately 50% of the soil water holding capacity. Struvite was applied in granular form without grinding. Each treatment was replicated five times, and the experiment was arranged in a completely randomized design, resulting in a total of 30 pots.

Spring oilseed rape (*Brassica napus* var. *oleifera* L.), variety “Lagoon”, was used as the test crop. Seeds originated from certified seed production, with a thousand-seed weight of 6.4 g and a germination rate of 97%. Sowing was carried out at a rate of three seeds per pot at a depth of 2 cm. One week after emergence, seedlings were thinned to one plant per pot, leaving the most vigorous individual to continue growth.

The experiment was arranged in a completely randomized design with five replicates per treatment, resulting in a total of 30 pots. Plants were grown from 2 October 2024 to 3 February 2025 (approximately 18 weeks) in a controlled growth room with a 14 h light / 10 h dark photoperiod, relative humidity of 60%, and a temperature of 21–23 °C.

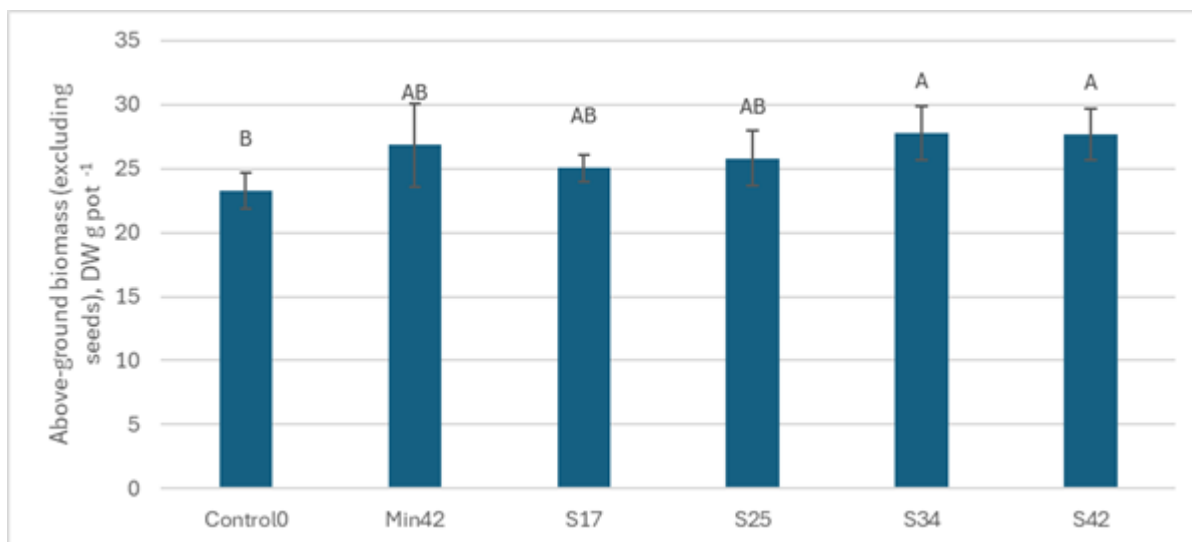
Watering was carried out manually using demineralized water at least twice per week. Pots were weighed regularly, and the required amount of water was calculated as the difference between the current pot weight and the target weight determined at sowing. To minimize spatial variability, pots were randomly rearranged in the growth room after each watering.

At the end of the experiment, plants were harvested. Fresh biomass of the aboveground parts, seeds, and roots were recorded. Plant material was then dried at 56 °C to constant weight. Dried samples were milled, and total phosphorus concentrations in aboveground biomass, seeds, and roots were determined using ICP-OES (Inductively Coupled Plasma Optical Emission Spectroscopy) after appropriate digestion.

## **Results**

### **Aboveground Biomass**

Phosphorus treatments significantly affected aboveground dry biomass (excluding seeds) ( $p = 0.024$ ). As shown in Figure 18, the highest biomass values were observed in struvite treatments 42 mg and 34 mg P kg<sup>-1</sup> dry soil. These were statistically higher than the unfertilized control.



**Figure 18 Aboveground dry biomass (mean  $\pm$  standard deviation, g DW pot<sup>-1</sup>, n=5).**

Control0 (no P added), S17, S25, S34 and S42 (struvite applied at 17, 25, 34 and 42 mg P kg<sup>-1</sup> dry soil, respectively), and Min42 (mineral phosphorus applied at 42 mg P kg<sup>-1</sup> dry soil). Different letters indicate significant differences between treatments at  $p < 0.05$  according to Tukey's HSD test.

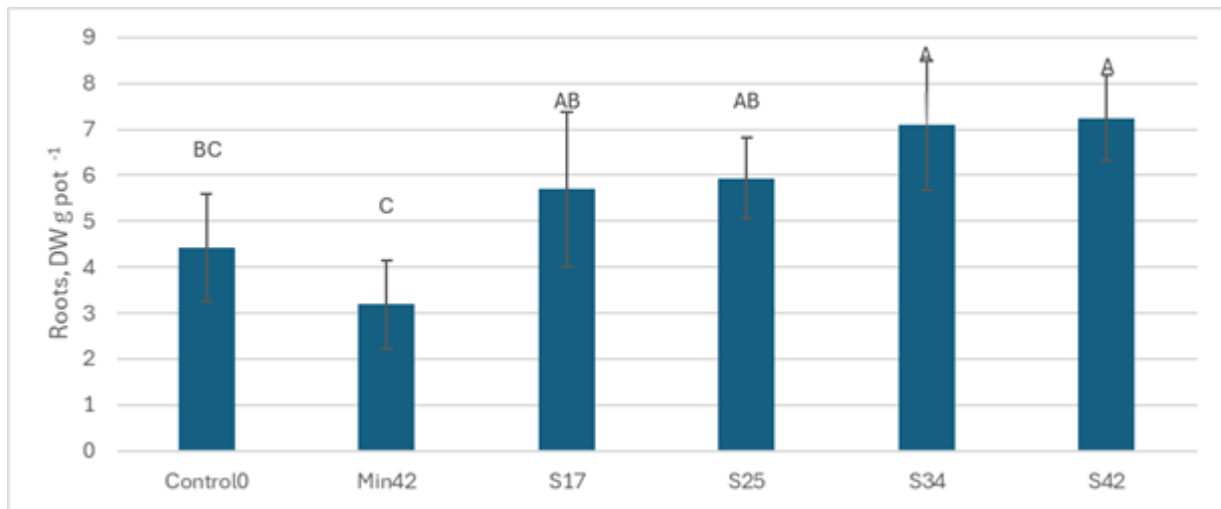
Although the results initially suggest that biomass increase was driven by phosphorus supply, the interpretation requires a broader perspective. The soil used in the pot experiment contained approximately 120 mg of plant-available P per pot (46 mg P kg<sup>-1</sup>soil), indicating that phosphorus availability per se was likely not strongly limiting. Instead, the physical properties of the soil probably played a critical role. The experimental soil had high silt and clay content (23.40% sand, 63.62% silt, and 12.98% clay) and became compacted during irrigation throughout the trial. Such compaction can reduce soil aeration, limit root penetration, and decrease effective nutrient diffusion, including phosphorus uptake. Therefore, the observed growth differences may reflect not only phosphorus rate effects but also interactions between fertiliser form and soil physical condition. The increase in biomass at higher struvite rates thus indicates that struvite is agronomically effective, but its effect may partly stem from improvements in the soil micro-environment rather than solely from increased phosphorus availability.

### Root Biomass

Root dry weight was strongly affected by treatment ( $p < 0.001$ ). As illustrated in Figure 19, root biomass increased progressively with increasing struvite rate, with the highest values recorded in treatments 42 mg and 34 mg P kg<sup>-1</sup> dry soil.

This response is particularly notable in the context of soil compaction. The clay- and silt-rich soil used in the pots compacted during the experiment, likely restricting oxygen diffusion and mechanical root penetration. In compacted agricultural soils, phosphorus uptake efficiency is often reduced despite adequate total soil P levels, because restricted root growth limits the plant's ability to explore the soil volume. The granular form of struvite likely played a key role. Granulated particles may have locally improved soil structure by creating micro-zones with slightly enhanced porosity within the dense soil matrix. These localized structural improvements may have enhanced aeration and facilitated root proliferation around fertiliser granules. Consequently, improved root growth may not have resulted solely from phosphorus availability,

but also from localized improvements in soil physical properties.



**Figure 19** Root dry biomass (mean  $\pm$  standard deviation, g DW pot<sup>-1</sup>, n=5).

Control0 (no P added), S17, S25, S34 and S42 (struvite applied at 17, 25, 34 and 42 mg P kg<sup>-1</sup> dry soil, respectively), and Min42 (mineral phosphorus applied at 42 mg P kg<sup>-1</sup> dry soil). Different letters indicate significant differences between treatments at  $p < 0.05$  according to Tukey's HSD test.

This finding suggests that in compacted field soils, phosphorus uptake can be constrained primarily by physical limitations rather than chemical P availability. Under such conditions, fertiliser form and its influence on soil structure may significantly influence plant response.

#### Seed yield and plant height

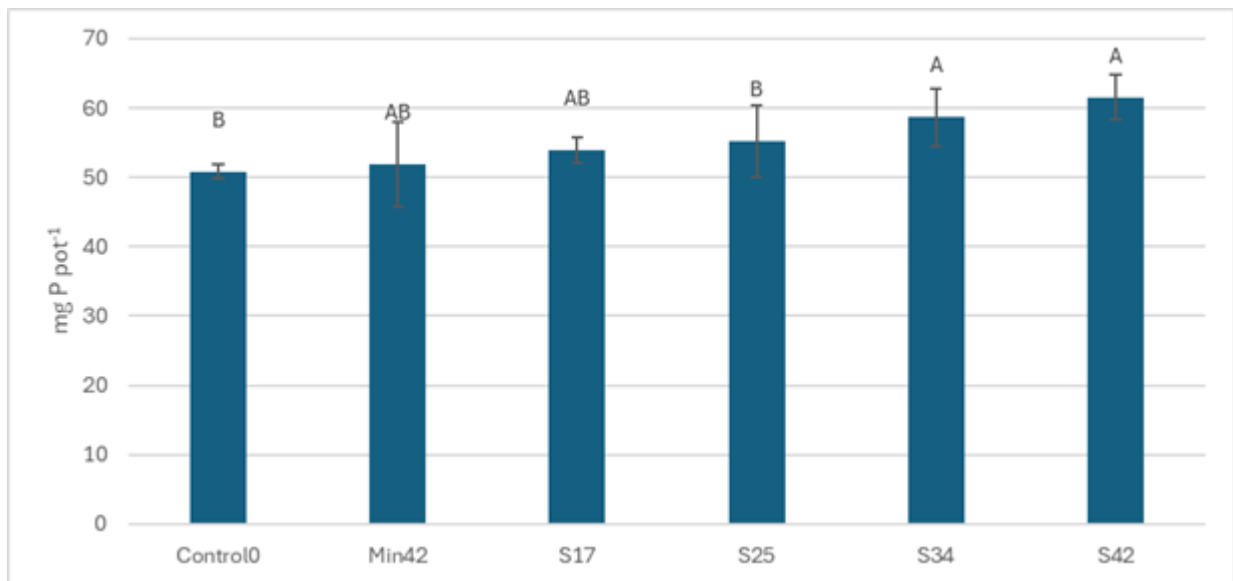
Plant height ranged from 102.5 to 116.6 cm across treatments. The tallest plants were observed in the Min42 treatment (116.6  $\pm$  28.92 cm), while the shortest were in S17 (102.5  $\pm$  6.56 cm). Variability was highest in Min42 and lowest in S17. There were no statistically significant differences in plant height between treatments (ANOVA,  $p > 0.05$ ).

Seed dry weight per pot varied between 4.60 and 5.43 g pot<sup>-1</sup>. The highest average seed weight was recorded in S25 (5.43  $\pm$  0.15 g pot<sup>-1</sup>), and the lowest in S42 (4.60  $\pm$  0.54 g pot<sup>-1</sup>). Differences among treatments were not statistically significant (ANOVA,  $p > 0.05$ ).

Overall, treatments did not significantly affect plant height or seed dry weight under the conditions of this experiment.

#### Total phosphorus uptake

Total phosphorus (P) uptake differed significantly among treatments ( $p = 0.0028$ ). As shown in Figure 20, the highest total P uptake was observed at the struvite application rate of 42 mg P kg<sup>-1</sup> dry soil, followed by the 34 mg P kg<sup>-1</sup> treatment. Nevertheless, the magnitude of the differences among treatments remained moderate.



**Figure 20 Total phosphorus uptake (mean  $\pm$  standard deviation, mg P pot<sup>-1</sup>, n=5).**

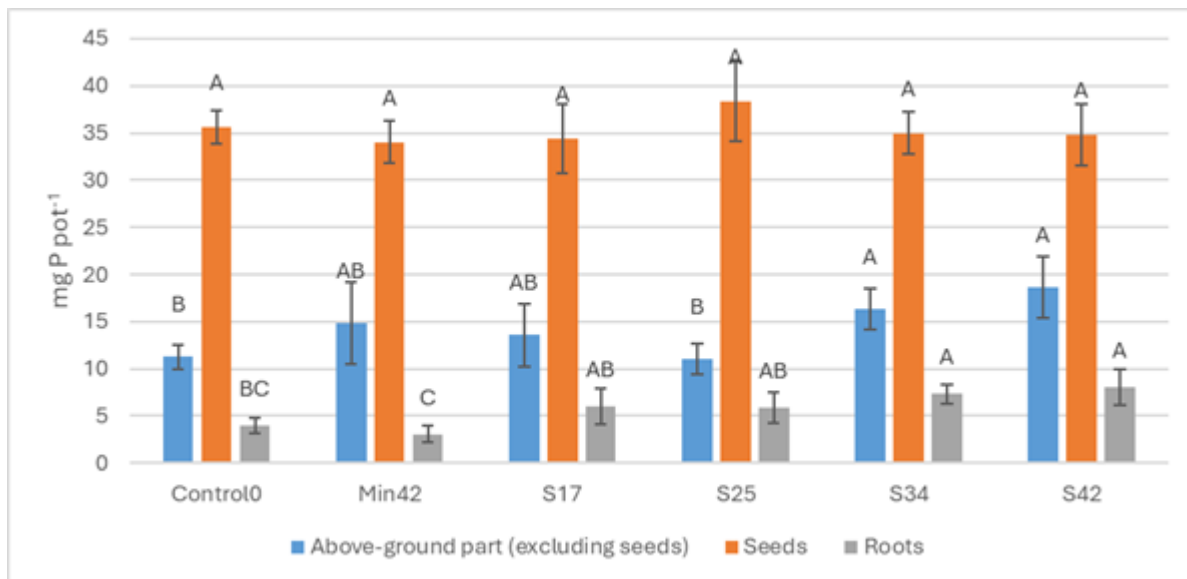
Control0 (no P added), S17, S25, S34 and S42 (struvite applied at 17, 25, 34 and 42 mg P kg<sup>-1</sup> dry soil, respectively), and Min42 (mineral phosphorus applied at 42 mg P kg<sup>-1</sup> dry soil). Different letters indicate significant differences between treatments at  $p < 0.05$  according to Tukey's HSD test.

Despite substantial variation in applied phosphorus rates, total P uptake across treatments remained within a relatively narrow range. This pattern suggests that the experimental soil likely contained sufficient plant-available phosphorus to satisfy a large proportion of crop demand even in the absence of additional P inputs. Consequently, increasing the P supply did not proportionally enhance plant uptake.

Moreover, soil physical constraints appear to have played a critical role in limiting nutrient acquisition. Soil compaction likely restricted root growth and reduced effective root surface area, thereby constraining phosphorus uptake efficiency. Under such conditions, phosphorus availability per se may not have been the primary limiting factor for plant growth. Instead, impaired root exploration in compacted soil likely reduced the plant's capacity to access both native and applied phosphorus. As a result, even when higher amounts of P were supplied, the crop's ability to utilize the additional nutrient input remained limited.

#### **Phosphorus uptake partitioning between aboveground biomass, roots, and seeds**

Phosphorus partitioning among plant organs varied significantly in response to phosphorus application, as illustrated in Figure 21. Root phosphorus content increased markedly with increasing struvite rates ( $p < 0.001$ ), demonstrating a clear positive response to enhanced P supply. Similarly, phosphorus content in the aboveground biomass increased significantly across treatments ( $p = 0.002$ ). In contrast, seed phosphorus content did not differ significantly among treatments, indicating that reproductive P allocation remained relatively stable regardless of fertiliser rate.



**Figure 21 Phosphorus uptake partitioning between aboveground biomass, roots, and seeds (mean  $\pm$  standard deviation, mg P pot<sup>-1</sup>, n=5).**

Control0 (no P added), S17, S25, S34 and S42 (struvite applied at 17, 25, 34 and 42 mg P kg<sup>-1</sup> dry soil, respectively), and Min42 (mineral phosphorus applied at 42 mg P kg<sup>-1</sup> dry soil). Different letters indicate significant differences between treatments at  $p < 0.05$  according to Tukey's HSD test.

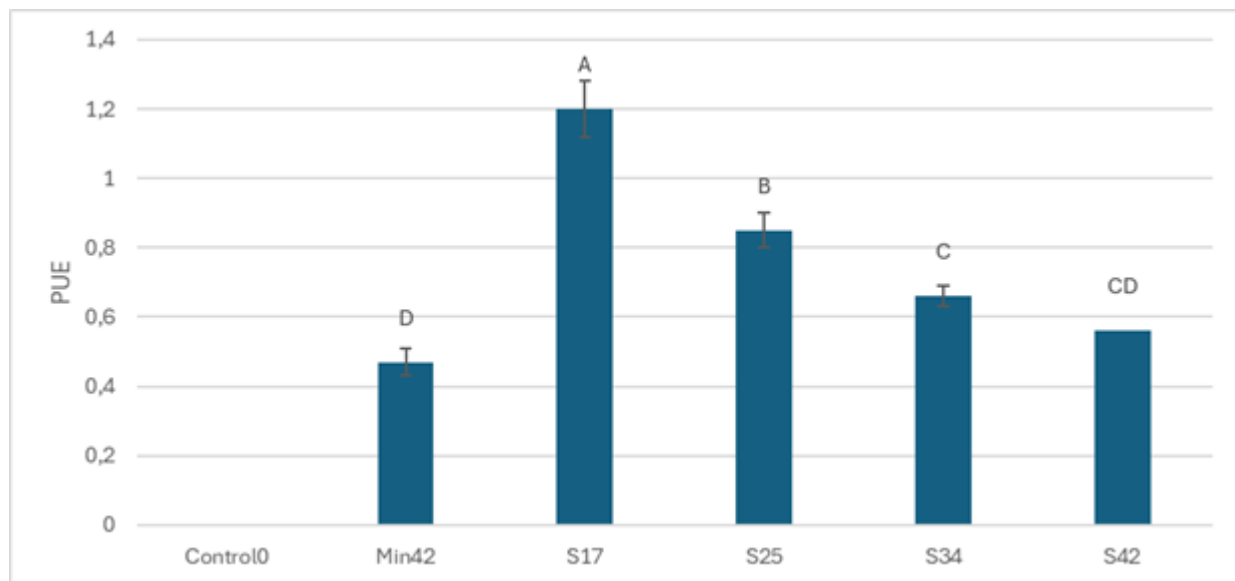
The distribution pattern of phosphorus within the plant indicates that additional P was preferentially allocated to vegetative tissues rather than to seeds. This shift suggests that improved phosphorus availability primarily stimulated vegetative growth and root development, while reproductive allocation remained comparatively conservative. The pronounced increase in root P accumulation further supports the hypothesis that localized changes in soil conditions surrounding struvite granules enhanced root activity and nutrient uptake. In compacted soil, such microsite improvements may have facilitated more effective nutrient acquisition, even though overall soil physical constraints continued to limit root expansion at the macro scale.

### Phosphorus use efficiency (PUE)

Phosphorus use efficiency (PUE) declined progressively with increasing phosphorus application rate, as illustrated in Figure 22. The highest PUE value (1.20) was recorded at the lowest struvite rate of 17 mg P kg<sup>-1</sup> dry soil, indicating the most efficient conversion of applied phosphorus into plant uptake under limited input conditions. In contrast, the mineral fertiliser treatment at 42 mg P kg<sup>-1</sup> dry soil exhibited the lowest PUE (0.47). Although higher struvite rates (34 and 42 mg P kg<sup>-1</sup> dry soil) resulted in greater total phosphorus uptake, their proportional efficiency declined relative to the lower application rates.

The observed inverse relationship between phosphorus application rate and PUE reflects diminishing marginal returns under conditions where baseline soil phosphorus availability was already relatively adequate. When soil P supply approaches or exceeds crop demand, additional fertiliser contributes proportionally less to plant uptake. Furthermore, in compacted soils, physical limitations on root growth and nutrient diffusion constrain the plant's capacity to access applied phosphorus efficiently. Consequently, increasing fertiliser rates does not translate into proportional increases in nutrient uptake, resulting in reduced nutrient use efficiency at higher

application levels.



**Figure 22 Nutrient use efficiency (PUE, mean ± standard deviation, n=5)**

Control0 (no P added), S17, S25, S34 and S42 (struvite applied at 17, 25, 34 and 42 mg P kg<sup>-1</sup> dry soil, respectively), and Min42 (mineral phosphorus applied at 42 mg P kg<sup>-1</sup> dry soil). Different letters indicate significant differences between treatments at  $p < 0.05$  according to Tukey's HSD test.

### Conclusions

The results demonstrate that struvite functioned as an agronomically effective phosphorus source under the controlled pot conditions of this experiment. Higher struvite application rates (34 and 42 mg P kg<sup>-1</sup> dry soil) significantly enhanced aboveground biomass and, even more markedly, root biomass production. The stimulation of root growth was particularly pronounced and consistent, indicating a strong belowground response to increasing struvite supply.

However, the experimental soil contained approximately 46 mg plant-available P kg<sup>-1</sup> soil ( $\approx 120$  mg P per pot), suggesting that baseline phosphorus availability was already moderate. Consequently, the observed plant responses cannot be attributed solely to correction of phosphorus deficiency. Instead, the results point toward an interaction between fertiliser form and soil physical conditions.

The soil used in the experiment was rich in silt and clay and became compacted during irrigation. Under such conditions, physical constraints-reduced aeration, limited mechanical root penetration, and restricted nutrient diffusion-likely constrained phosphorus uptake efficiency. The granular structure of struvite may have created localized microsites with improved porosity and aeration, thereby facilitating root proliferation around fertiliser particles. The significant increase in root biomass under higher struvite rates supports this interpretation.

Total phosphorus uptake increased with increasing struvite rate, but the magnitude of the increase remained moderate relative to the applied P gradient. This indicates diminishing marginal returns in a soil that was not strongly P-deficient and where physical constraints limited full exploitation of the soil volume. The decline in phosphorus use efficiency (PUE) with increasing application rate further supports this interpretation: the highest efficiency was achieved at the

lowest struvite rate, while the mineral fertiliser treatment exhibited the lowest PUE.

Importantly, seed yield and plant height were not significantly affected by phosphorus treatments. This indicates that reproductive development was relatively stable across treatments and that the primary treatment effects were expressed in vegetative growth and root development rather than in reproductive allocation.

Overall, the findings suggest that in compacted, fine-textured soils, phosphorus uptake may be limited more by physical soil constraints than by chemical phosphorus availability. Under such conditions, fertiliser form and its localized effects on the soil micro-environment may play a critical role in determining plant response. Moderate struvite application rates may therefore represent an agronomically efficient strategy in soils with adequate baseline phosphorus levels.

#### 4.2.5 Evaluation centre in Poland (IMP)

##### *Soil*

The sandy soil was sieved to <2 mm (Table 11) and mixed with peat at a sand-to-peat ratio of 5:1 (w/w), corresponding to a volumetric ratio of 1:1.5. The resulting substrate contained 89% total solids (TS), 9.88% volatile solids (VS), 1.043 g N kg<sup>-1</sup> TS, 486.57 mg P kg<sup>-1</sup> TS, 19.04 mg Olsen-P kg<sup>-1</sup> TS, and 610 mg K kg<sup>-1</sup> TS, with a pH of 8.29. Approximately 1.85 kg of the prepared soil–peat mixture was transferred into pots with an internal diameter of 14.5 cm (surface area: 0.0165 m<sup>2</sup>). BADSS, SADSS, BADSS+EM, SADSS+EM, BADSS+Ch, SADSS+Ch, UBG and CMG were applied at seven rates equivalent to 20–370 kg N ha<sup>-1</sup> in order to establish the full nitrogen response curve of the forage grass mixture and to include elevated fertilization scenarios.

**Table 11 Initial physicochemical characteristics of the soil used in the experiment**

Soil parameters (sand + peat)	Unit	Value	SD
Total Solids	% fresh matter	89.90	0.699
Organic matter, o.m.	% fresh matter	9.880	0.462
Total Nitrogen, TN	gN/ kg d.m.	1.043	0.088
Orto-Phosphate, PO <sub>4</sub> <sup>3-</sup>	mgP/ kg d.m.	158.77	1.68
Total Phosphorus	mgP/ kg d.m.	486.567	4.58
P-Olsen	mgP/ kg d.m.	19.04	0.68
Total Potassium, TK	mgK/ kg d.m.	610.00	26.48
K-Olsen	mgK/ kg d.m.	64.94	19.22
pH		8.287	NA
Redox potential	mV	-63.8	NA
EC	μS/cm	159.7	NA

##### *Treatment*

The effect of pelleted waste-based fertilizers and their enrichments on the growth of ryegrass was studied in a pot experiments carried out in a glasshouse – Gdynia Wiczlino, Pomerania,

Northern Poland during a four-month period (mid-April to mid-August 2025). Recycled nutrient fertilizers were produced by Rendben Ltd. (Figure 23).

BADSS is a multi-component pelleted fertilizer derived from organic and mineral waste, containing 17.5–22.5% fishcake processing waste, 17.5–22.5% digested sewage sludge, 32.5–37.5% biomass ash, and 22.5–27.5% biochar (Table 12). Its production process involves mixing, pelleting, and drying under controlled conditions, ensuring a fertilizer with high nutrient content, low heavy metal content, and good physical stability. The relevant patent application has been submitted to the Polish Patent Office on Nov. 12th 2025: [WIPO ST 10/C PL453720].

SADSS is a multi-component pelleted fertilizer derived from organic and mineral waste, containing 17.5–22.5% fishcake processing waste, 17.5–22.5% digested sewage sludge, 32.5–37.5% biomass ash, and 22.5–27.5% chromium-free cattle shavings from a leather car upholstery plant (Table 12). Its production process involves mixing, pelleting, and drying under controlled conditions, ensuring a fertilizer with a high nutrient content, low heavy metal content, and good physical stability. The relevant patent application has been submitted to the Polish Patent Office on Nov. 12th 2025: [WIPO ST 10/C PL453721].

BADSS+EM includes BADSS enriched with an additive based on bakery and dairy products or waste and dosing it before and during plant growth. The relevant patent application has been submitted to the Polish Patent Office on Nov. 12th 2025: [WIPO ST 10/C PL453722]. SADSS+EM contains the same enrichment but applied to SADSS. The relevant patent application has been submitted to the Polish Patent Office on Nov. 12th 2025: [WIPO ST 10/C PL453723]. BADSS+Ch includes BADSS enriched with a chitosan-based enrichment additive from seafood waste and dosing it before and during plant growth. The relevant patent application has been submitted to the Polish Patent Office on Nov. 12th 2025: [WIPO ST 10/C PL453724]. SADSS+Ch contains the same enrichment but applied to SADSS. The relevant patent application has been submitted to the Polish Patent Office on Nov. 12th 2025: [WIPO ST 10/C PL453725].

Additionally, UBG Urine Based Granules with NPK content of 15-2-4 (%) supplied from Sanitation360 (Project Partner). According to the information found on their website, urine is treated directly in the collection tank of each urinal or urine-diverting toilet. This process lowers the pH, preserving over 90 % of the nitrogen and 100 % of the phosphorus and potassium while preventing ammonia formation and with it, reducing unpleasant odours. In the next stage, convective air-drying removes about 95 % of the liquid volume, eliminating the need for bulky piping or large storage tanks.

**Table 12 Basic characteristics of fertiliser materials applied and their photos (below)**

Pellet	T.S. (%)	N (%)	P (%)	K (%)	P2O5 (%)	K2O (%)	Mg (%)	Enterobacteriaceae (units/ g)
SADSS	88,2	4,93	1,64	9,2	3,76	11,09	1,26	< 1,0 x 10
BADSS	78,1	0,62	1,44	7,2	3,30	8,68	1,13	< 1,0 x 10
Pellet	Cu (mg/kg)	Zn (mg/kg)	Cr (mg/kg)	Cd (mg/kg)	Ni (mg/kg)	Pb (mg/kg)	Hg (mg/kg)	C org (%)
SADSS	84,4	1030	32	6,6	13,2	21,6	0,261	19,6
BADSS	80,5	1000	24,4	6,7	14,4	21,7	0,265	20,3



**BADSS pellet**



**SADSS pellet**

**Figure 23 Pellets used in the pot experiment**

In total, seven waste streams were investigated in a glasshouse experiment and compared with the commercially available organic NPK fertilizer (cow manure granulated, CMG). Experiment also includes control treatment. Six pots without organic any fertilizer addition served as the control treatment.

The analysed recycled nutrient fertilizers SADSS and BADSS contain substantial macronutrients, organic carbon near ~20 % (19.6 % and 20.3 %) and very low enterobacteriaceae counts ( $<1.0 \times 10$  units/g), which is favourable for safety. The total concentrations of heavy metals (Table 12), fall within many nationally adopted or EU-influenced heavy metal guidance values for soil and sludge application and are well below the more stringent cadmium threshold of 60 mg Cd/kg  $P_2O_5$  established under the EU Fertilising Products Regulation 2019/1009 for phosphate fertilisers marketed in the internal market as CE-fertilising products (where applicable). While Regulation (EU) 2019/1009 harmonises maximum contaminant limits (including heavy metals such as Cd, Pb, Hg and Ni) and safety criteria for both mineral and recycled/organic fertilisers, forming the basis for environmental and health protection in the EU, specific limit values for all heavy metals (beyond Cd for phosphate fertilisers) may be defined at the product function category (PFC) or member state level, and producers must demonstrate compliance through testing and documentation before placing products on the EU market under CE marking. Based on available EU regulatory benchmarks, both SADSS and BADSS heavy metal concentrations would generally meet the current EU contaminant safety expectations, suggesting suitability of these recycled fertilizers for EU markets, though product-specific conformity assessment and labelling in accordance with Regulation (EU) 2019/1009 is still required.

The study also used enrichments including (1) self-made EM containing silage from bakery/ diary waste for testing the effects of lower pH and bacillus in increasing nutrients bioavailability and (2) suspension containing seafood waste (shrimp shells) with chitosan possibly reducing plants

stress and enhancing their growths. Photos of these boosters are presented below (Figure 24).



**Figure 24** *Enrichment boosters used in the experiment: Silage from bakery/ dairy with EM (left) and seafood-waste suspension with chitosan (right)*

The 60-day fermentation of the bakery and dairy waste mixture generated the occurrence of effective microorganisms, presumably lactic acid bacteria (EM) resulted in a nutrient-rich substrate characterized by high organic content and moderate mineral availability (Table 1 and 2, Appendix). The solid fraction exhibited a dry matter content of  $11.08 \pm 0.47\%$  (fresh matter basis) and an exceptionally high organic matter proportion of  $93.89 \pm 0.47\%$ , indicating efficient stabilization of biodegradable components during fermentation. Total nitrogen reached  $27.14 \pm 0.03 \text{ g N kg}^{-1} \text{ d.m.}$ , confirming substantial nitrogen enrichment, while total phosphorus concentration was  $1021.17 \pm 18.87 \text{ mg P L}^{-1}$ , with orthophosphate accounting for  $330.37 \pm 6.99 \text{ mg P L}^{-1}$ , suggesting good phosphorus bioavailability. The liquid fraction contained  $0.810 \pm 0.086 \text{ g N L}^{-1}$  and  $449.40 \pm 1.15 \text{ mg P L}^{-1}$ , demonstrating effective nutrient solubilization. The measured pH of 4.69 reflects acidic conditions typical of organic fermentation processes, while the redox potential (138.10 mV) indicates moderately reducing conditions, favourable for microbial activity. Electrical conductivity remained low ( $6.09 \mu\text{S cm}^{-1}$ ), suggesting limited salinity. Overall, the results confirm that 60 days of fermentation produced a stabilized, nutrient-rich bio-booster, possibly with EM with high organic matter content and readily available nitrogen and phosphorus, highlighting its potential suitability for agricultural or biotechnological applications. The enrichments were added to each pot 3 times during the first month of growth according to the following steps: 100 g of solid material once prior to seeding, 12 days later 45 ml of liquid material (supernatant) and 24 days later 30 ml of liquid material again. This resulted in introducing additional 0.3612 g N (originated from the enrichment) to each pot, possibly boosting the growths.

Many papers report chitin (the natural polymer in shells) as the fraction you measure on a dry-weight basis; chitosan is made from that chitin by chemical/enzymatic de-acetylation and so its final yield is necessarily  $\leq$  the chitin fraction and depends on process losses and deacetylation efficiency. Because (a) chitin in shrimp shells is commonly reported between 15–30% dry weight, and (b) chitosan extraction yields reported in the literature commonly fall in the  $\sim 10\text{--}25\%$  band

(with examples at ~18% chitin and ~23% chitosan), a conservative working average for chitosan content (as extracted product per dry shrimp shell mass) is ~20% ( $\pm$  ~7–10%) (Ahmed et al., 2025; Antonino et al., 2017; Hu et al., 2020; Verardi et al., 2023). The enrichments in a form of 1:1 water solution containing blended shrimp shells fresh mass were added to each pot 3 times: 5.43 g (27.04.2025), 1.39 g (08.05.2025) introducing in total 6.82 g/ pot as referred to shrimp shells fresh mass. This resulted in introducing additional 0,092 g N (originated from the enrichment) to each pot, possibly also boosting the growth.

Assuming a positive response of the plant to nitrogen-rich organic waste, fertilizer doses were applied at rates from 20 up to 370 kg N/ha to reach the plateau of the N response curve. Table 13 outlines the experimental design, showing the assumed fertilizer dose and the corresponding (calculated) nitrogen and fertilizer amount per pot.

**Table 13 Amounts of RNFs and reference treatment, added to the soil in the glasshouse experiment based on N content**

Application rate		RNFs, g fertiliser/ pot							
Nr	kg N/ha	CMG	UBG	BADSS	SADSS	BADSS+EM	SADSS+EM	BADSS+Ch	SADSS+Ch
1*	20	1,179	0,413	6,817	0,759	6,817	0,759	6,817	0,759
2	70	4,126	1,444	23,859	2,657	23,859	2,657	23,859	2,657
3	120	7,073	2,476	40,902	4,555	40,902	4,555	40,902	4,555
4**	170	10,021	3,507	57,944	6,453	57,944	6,453	57,944	6,453
5	220	12,968	4,539	74,987	8,351	74,987	8,351	74,987	8,351
6	270	15,915	5,570	92,029	10,248	92,029	10,248	92,029	10,248
7	370	21,810	7,633	126,114	14,044	126,114	14,044	126,114	14,044

\* 1- normal, \*\* - max application rate in Poland, Cow Manure Granulated (CMG), Urine Based Granules (UBG), N application rate corresponds to N introduced with pellets only excluding booster N.

Table 13 illustrates how increasing nitrogen (N) application rates resulted in markedly different fertilizer masses added to the pots, depending on the N concentration of each product. For cow manure granulate (CMG), which has a relatively low N content, progressively higher application loads (from 20 to 370 kg N ha<sup>-1</sup>) required moderate increases in fertilizer mass, ranging from 1.18 g pot<sup>-1</sup> at the lowest dose to 21.81 g pot<sup>-1</sup> at the highest dose. In contrast, BADSS, characterized by very low N concentration, demanded substantially larger fertilizer quantities to achieve the same N targets, increasing sharply from 6.82 g pot<sup>-1</sup> at 20 kg N ha<sup>-1</sup> to as much as 126.11 g pot<sup>-1</sup> at 370 kg N ha<sup>-1</sup>. SADSS, with higher N content than BADSS, required considerably smaller amounts, rising from only 0.76 g pot<sup>-1</sup> to 14.04 g pot<sup>-1</sup> across the same N range. The addition of effective microorganisms from the bakery/ diary waste silage (B+EM and S+EM) in fact introduced additional N into each pot but the data presented above denote only masses based on N contained in pellets only. Whereas these enrichments act as boosting agents, therefore the additional content of N is not considered while calculating the fertilizer masses. Overall, the results clearly demonstrate that fertilizers with lower intrinsic N content require disproportionately higher application rates to meet plant N demand, while more N-rich materials (such as SADSS) allow precise nutrient delivery with much smaller dosages, which has important

implications for handling, cost efficiency, and potential soil loading effects.

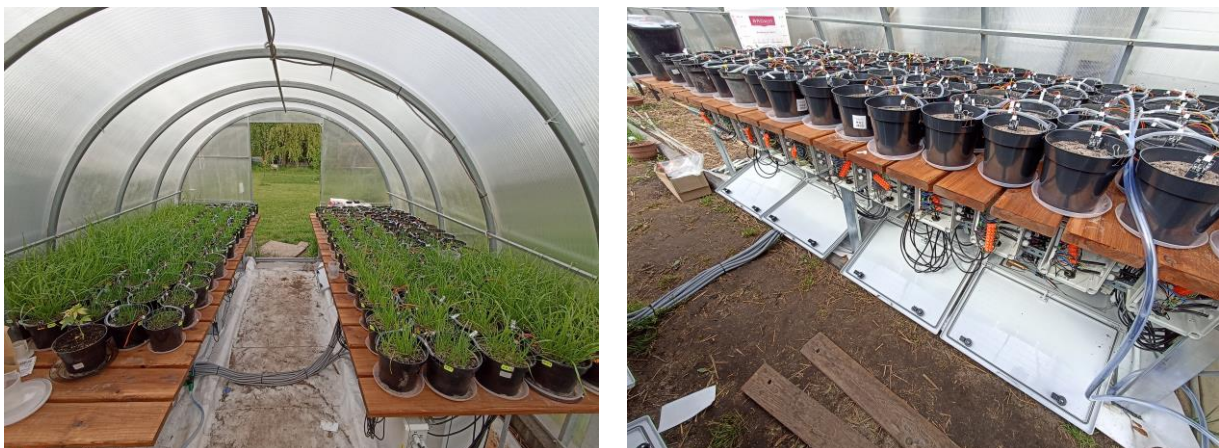
### **Experiment set up**

Plants were cultivated in the <2 mm sieved fraction of a sandy soil mixed with peat in a weight ratio of sand/peat = 5/1, equivalent to a volume ratio of 1:1.5. Approximately 1.85 kg of this prepared soil was placed in pots with an internal diameter of 14.5 cm (surface area: 0.0165 m<sup>2</sup>). Each pot received supplementary nutrient solutions (except nitrogen), according to the recipe: 12 ml/pot and 6 ml/pot of K<sub>2</sub>SO<sub>4</sub> (42 g/L); CaCl<sub>2</sub>·2H<sub>2</sub>O (90 g/L); MgSO<sub>4</sub>·7H<sub>2</sub>O (24 g/L); MnSO<sub>4</sub>·H<sub>2</sub>O (6 g/L); ZnSO<sub>4</sub>·7H<sub>2</sub>O (5.4 g/L); CuSO<sub>4</sub>·5H<sub>2</sub>O (1.2 g/L); H<sub>3</sub>BO<sub>3</sub> (0.42 g/L); CoSO<sub>4</sub>·7H<sub>2</sub>O (0.16 g/L); Na<sub>2</sub>Mo<sub>4</sub>·2H<sub>2</sub>O (0.12 g/L).

The soil in each pot was pre-watered with 120 ml of tap water, then thoroughly mixed with nutrients in the top 5 cm layer. Eighty grains of annual ryegrass—0.5 g in total (containing *lolium perenne* 40%, *lolium multiflorum-estanzuela* 20%, *festuca rubra* 25%, and *lolium hybridum* 15%)—were sown on the soil surface and covered with an additional 80 g of soil. Experiments were conducted in triplicate, with pots re-randomized every 7 days to equalize light exposure and maintained at a constant moisture at field capacity (ca. 65%) thanks to automatic irrigation system. Harvests took place monthly over a 4-month period, cutting plants about 1 cm above the soil. The harvested material was then placed in paper bags and dried at 105 °C until reaching a constant weight (Figure 25).

### **Plant and residual soil analyses**

After each of the four harvests, ryegrass tops were dried, ground, and analysed for Total Kjeldahl Nitrogen (TKN). Samples were digested in concentrated H<sub>2</sub>SO<sub>4</sub> with a titanium-based catalyst using a SpeedDigester K-436 (Büchi Labortechnik AG), then steam-distilled (K-355 distillation unit, Büchi Labortechnik AG) into boric acid solution with a Tashiro indicator before titration with HCl to determine ammonia content. After the finalization of the experiment, soil samples were analysed for pH and electrical conductivity EC (1:5 H<sub>2</sub>O), along with total soil nitrogen. Below are the photos of the greenhouse used: general view (left) and close to the automatic irrigation system.

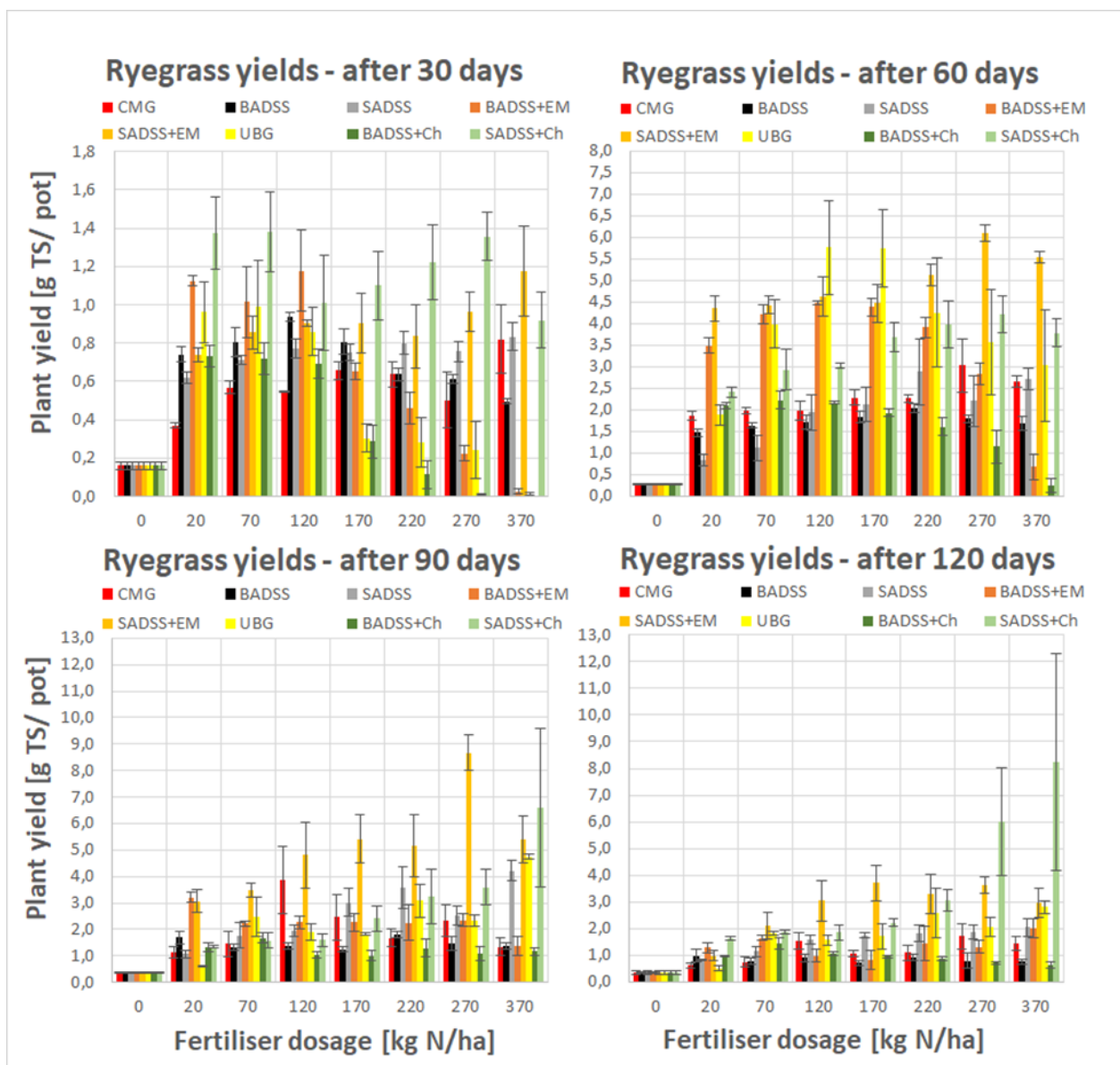


**Figure 25** Pot experiment conducted in Evaluation Centre

## Results

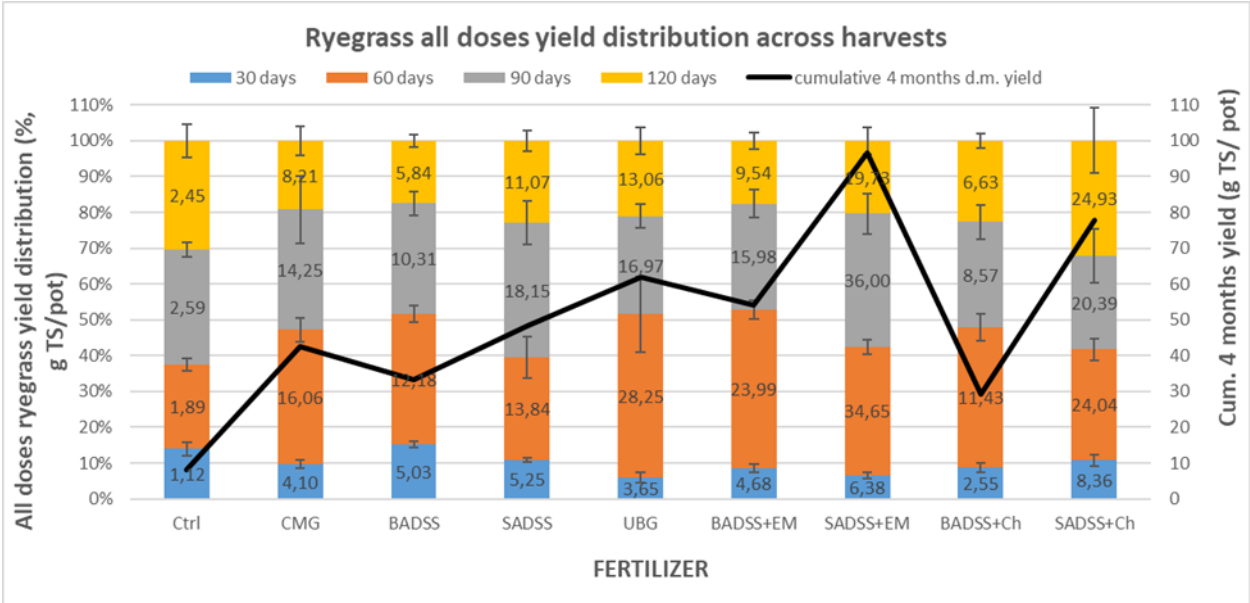
### Response of the forage grass mixture biomass yield to pelleted waste-based fertilizers

Ryegrass dry matter (DM) yields showed clear differences across fertilizer type, N application rate, and harvest time, reflecting both nutrient availability and amendment-specific effects (Figure 26). At early growth (30 days), yield differences were modest, with all treatments producing relatively low biomass. However, SADSS+Ch and BADSS+Ch already showed a tendency toward higher yields at moderate N rates (70–120 kg N ha<sup>-1</sup>), suggesting early stimulation of growth. CMG and BADSS remained among the lowest-performing treatments, indicating slower nutrient release. At 60 days, treatment effects became more pronounced.



**Figure 26** Forage grass mixture biomass yields response to recycled nutrient fertilizers compared to the commercial organic fertilizer (CMG) for harvests after 30, 60, 90 and 120 days. Standard deviations included, insignificant if not visible

Treatments UBG and SADSS+EM achieved the highest yields, particularly at  $\geq 120$  kg N ha<sup>-1</sup>, indicating rapid N availability and strong plant response. BADSS+EM showed variable performance, often lower than SADSS+EM, confirming weaker synergy between EM and the BADSS matrix. By 90 and 120 days, late-season dominance shifted toward SADSS+Ch and UBG, especially at high N rates (220–370 kg N ha<sup>-1</sup>). SADSS+Ch reached the highest cumulative yields, indicating sustained nutrient release and prolonged growth stimulation. In contrast, BADSS+Ch declined at higher N rates, and BADSS and CMG consistently remained among the lowest-yielding treatments.



**Figure 27 Ryegrass growth (as total dry matter yield) dynamics across four harvests for 7 RNFs, cow manure granulated and a control: percentage of total dry matter in each harvest (left axis) and cumulative 4 months dry matter yield (bold line – right axis). Standard deviations included, insignificant if not visible.**

Across all treatments, yields generally increased from 20 to 120–170 kg N ha<sup>-1</sup>, followed by diminishing or inconsistent gains at higher rates. High N inputs ( $\geq 220$  kg N ha<sup>-1</sup>) often led to yield plateaus or declines (unlike SADSS+Ch), particularly in BADSS-based systems, reflecting reduced efficiency and possible stress or nutrient imbalance.

Yield accumulation increased strongly over time, with the largest differences between treatments emerging at 90 and 120 days, highlighting the importance of nutrient release dynamics. Treatments with controlled or prolonged N release (SADSS+Ch, SADSS+EM) performed best in later harvests.

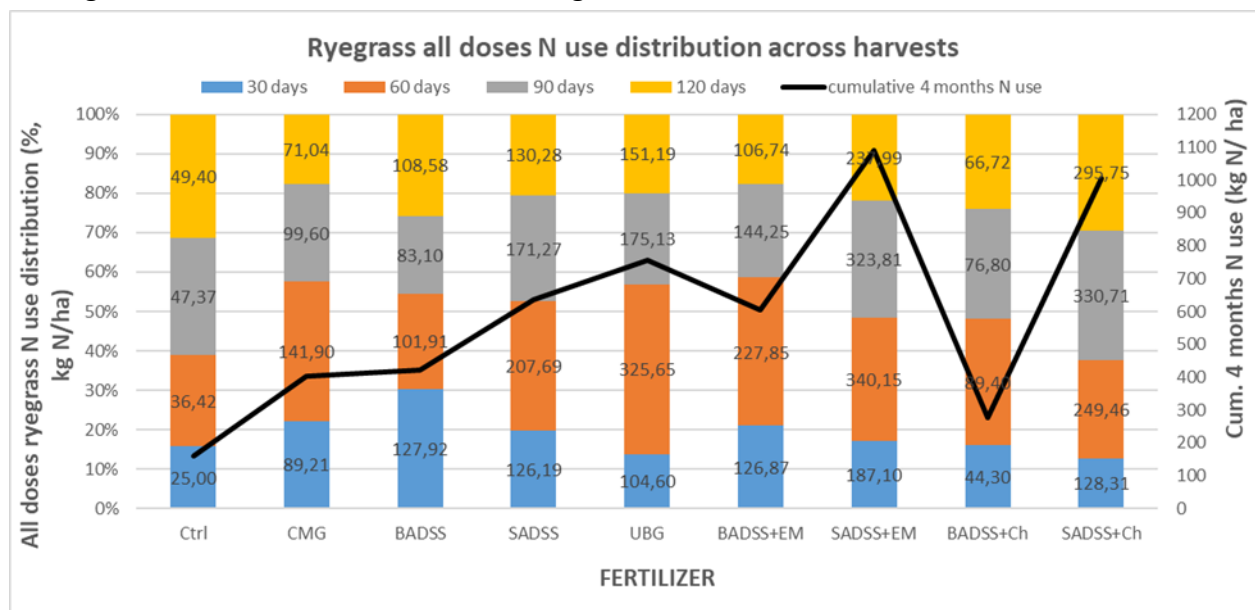
Standard errors were generally small at low N rates and early harvests, indicating consistent responses. However, SE increased markedly at higher N rates and later harvests, particularly for UBG and SADSS+Ch, reflecting greater variability in plant response under high nutrient supply. BADSS+EM also showed high variability, confirming unstable performance. In contrast, CMG and BADSS exhibited relatively low SE, indicating more consistent but lower yields.

Overall, SADSS-based systems (especially with Ch or EM) and UBG delivered superior yields,

particularly at later growth stages, while BADSS-based treatments were less efficient and more variable, especially at high N loads.

### Integrated discussion of yield and N use dynamics across harvests

The combined analysis of biomass yield and nitrogen (N) use dynamics reveals clear temporal shifts in nutrient utilization efficiency and treatment performance across the four harvests (Figure 27). Across all treatments, early growth (30 days) contributed only a small fraction to both total yield and cumulative N use (Помилка! Джерело посилання не знайдено.). This stage was characterized by limited biomass accumulation (typically <10–15%) and relatively low N uptake, indicating that initial plant establishment was not strongly driven by fertilizer type. Differences among treatments were minimal at this stage.



**Figure 28 Nitrogen use dynamics across four harvests for 7 recycled nutrient fertilizers, cow manure granulated and a control. percentage of total N use in each harvest (left axis) and cumulative 4 months N uptake (bold line – right axis). Standard deviations included, insignificant if not visible.**

At 60 days, a substantial increase in both yield contribution and N use was observed, marking the transition to active growth. For most treatments, this harvest accounted for the largest share of N uptake, often exceeding 30–40% of total N use. Notably, treatments such as SADSS+EM and UBG exhibited particularly high N uptake at this stage, indicating rapid nutrient availability and strong early responsiveness. However, yield contributions, although increased, remained more evenly distributed across subsequent harvests.

By 90 days, divergence between treatments became more pronounced. Yield contributions increased significantly, especially for SADSS, UBG, and SADSS+EM, reflecting sustained nutrient supply. In contrast, N use remained high but began to stabilize or redistribute, suggesting improved synchronization between nutrient release and plant demand. Treatments like BADSS+EM showed relatively high N use without proportional yield gains, indicating reduced efficiency.

Dose		NUE for CMG (%)				NUE for UBG (%)			
kg N/ ha	30 days	60 days	90 days	120 days	30 days	60 days	90 days	120 days	
20	36,33	80,79	39,56	35,56	104,67	63,16	33,31	28,20	
70	13,86	25,86	15,33	8,72	42,49	39,88	36,67	37,47	
120	9,08	18,18	20,77	10,67	21,14	30,68	11,94	16,31	
170	7,66	11,68	10,88	5,69	5,64	38,35	7,93	11,33	
220	6,03	8,20	5,24	4,03	4,69	36,27	13,05	14,32	
270	5,06	9,53	6,04	5,21	3,01	23,65	9,79	7,77	
370	5,78	6,00	2,62	3,36	0,14	10,66	16,17	7,57	
Dose		NUE for BADSS (%)				NUE for SADSS (%)			
kg N/ ha	30 days	60 days	90 days	120 days	30 days	60 days	90 days	120 days	
20	77,61	59,90	83,38	83,83	72,00	64,31	64,85	56,00	
70	22,56	20,00	11,79	20,45	24,24	24,21	21,47	19,64	
120	19,32	10,89	8,22	9,76	14,98	21,20	17,13	17,96	
170	12,36	8,79	6,19	7,89	10,25	17,12	14,05	11,65	
220	8,83	7,27	6,44	7,91	8,92	21,10	13,27	8,89	
270	6,68	5,84	5,10	6,36	6,90	12,78	8,93	7,65	
370	4,03	4,37	2,66	4,81	5,72	11,47	12,31	6,42	
Dose		NUE for BADSS+EM (%)				NUE for SADSS+EM (%)			
kg N/ ha	30 days	60 days	90 days	120 days	30 days	60 days	90 days	120 days	
20	133,96	130,18	150,59	58,36	115,69	217,59	129,72	53,41	
70	36,96	59,70	22,13	22,91	38,09	76,92	47,76	37,55	
120	27,13	33,20	14,91	9,30	21,36	35,15	36,09	29,74	
170	12,18	30,67	13,10	6,31	15,13	27,88	39,01	32,43	
220	6,15	16,60	9,73	8,43	11,12	21,60	26,36	21,21	
270	2,52	9,86	7,42	5,74	10,41	20,32	23,36	14,81	
370	0,17	1,33	4,60	6,24	9,02	13,74	9,12	6,38	
Dose		NUE for BADSS+EM (%) corrected*				NUE for SADSS+EM (%) corrected*			
kg N/ ha	30 days	60 days	90 days	120 days	30 days	60 days	90 days	120 days	
239	11,21	10,90	12,61	4,88	9,68	18,21	10,86	4,47	
289	8,95	14,46	5,36	5,55	9,23	18,64	11,57	9,10	
339	9,60	11,75	5,28	3,29	7,56	12,45	12,78	10,53	
389	5,32	13,41	5,73	2,76	6,61	12,19	17,05	14,17	
439	3,08	8,32	4,88	4,23	5,57	10,83	13,21	10,63	
489	1,39	5,44	4,10	3,17	5,75	11,22	12,90	8,18	
589	0,11	0,84	2,89	3,92	5,67	8,63	5,73	4,01	
Dose		NUE for BADSS+Ch (%)				NUE for SADSS+Ch (%)			
kg N/ ha	30 days	60 days	90 days	120 days	30 days	60 days	90 days	120 days	
20	54,23	81,28	54,94	46,82	90,93	94,51	65,27	71,05	
70	15,21	26,73	19,15	18,64	24,91	32,11	23,70	25,20	
120	11,07	11,03	5,87	7,18	11,01	21,32	15,86	16,70	
170	4,06	6,51	5,65	6,23	8,49	25,12	17,44	16,14	
220	1,09	6,43	5,74	4,59	9,29	20,77	20,66	19,63	
270	0,08	4,45	3,70	2,82	8,01	16,83	19,31	28,79	
370	0,00	1,07	3,55	1,99	6,21	13,15	41,84	25,82	
Dose		NUE for BADSS+Ch (%) corrected*				NUE for SADSS+Ch (%) corrected*			
kg N/ ha	30 days	60 days	90 days	120 days	30 days	60 days	90 days	120 days	
76	14,32	21,46	14,51	12,36	24,01	24,95	17,23	18,76	
84	12,64	22,21	15,91	15,49	20,69	26,68	19,69	20,93	
184	7,21	7,18	3,82	4,67	7,17	13,89	10,33	10,88	
284	2,43	3,90	3,38	3,73	5,08	15,02	10,43	9,65	
384	0,62	3,68	3,29	2,63	5,32	11,89	11,83	11,24	
484	0,05	2,48	2,06	1,57	4,47	9,38	10,77	16,05	
684	0,00	0,58	1,92	1,08	3,36	7,11	22,62	13,96	

**Figure 29 . Nitrogen Use Efficiency calculated for each dose at each scenario and harvest time. Note that NUE for BADSS+EM, SADSS+EM, BADSS+Ch and SADSS+Ch was displayed as a function of pellets N content applied and \* as a function of pellets + EM/ Ch enrichment (booster) N content applied.**

At 120 days, the final harvest contributed the largest share of total yield in several treatments,

particularly SADSS+Ch and UBG, confirming strong late-season productivity. However, N use at this stage was generally lower than at 60–90 days, indicating that plants were able to produce biomass with comparatively lower additional N uptake. This reflects improved internal N utilization and recycling within the plant system.

The cumulative trends further highlight key differences. SADSS+Ch and SADSS+EM achieved the highest total yields, but their N use patterns differed: SADSS+EM showed very high cumulative N uptake, while SADSS+Ch combined high yield with slightly lower N use, indicating better efficiency. In contrast, BADSS+EM exhibited high N consumption without proportional yield gains, confirming inefficiencies in this system.

Overall, the results demonstrate that high yield does not necessarily correspond to proportional N use, and that treatment performance depends on the temporal alignment between N release and plant demand. SADSS-based systems, particularly with chitosan enrichment, achieved the most balanced yield–N use relationship, while EM enrichment tended to increase N uptake intensity, sometimes at the expense of efficiency.

### **Effect of N dose**

Across all fertilizers, NUE declined sharply with increasing N application rate, confirming diminishing returns under surplus N supply. The highest NUE values were consistently observed at 20 kg N ha<sup>-1</sup>, while values at ≥220–370 kg N ha<sup>-1</sup> dropped to very low levels, often below 10% (Figure 29). This decline was particularly pronounced for BADSS-based systems and for EM-enriched treatments when corrected for total N input, indicating strong sensitivity to over-fertilisation.

### **Effect of fertilizer type**

Among non-enriched fertilizers, BADSS and SADSS exhibited higher NUE than CMG, particularly at low N rates, reflecting more efficient nutrient recovery from recycled matrices. UBG showed very high NUE at low N (up to ~105%), but declined rapidly with increasing dose, indicating fast N availability but poor efficiency under high inputs.

EM-enriched treatments (BADSS+EM, SADSS+EM) displayed extremely high apparent NUE at low doses (often >100%), especially at early harvests. However, once corrected for booster-derived N, NUE values decreased substantially (typically to 5–20%), revealing that much of the apparent efficiency was driven by both additional N inputs and improved nutrient use.

A similar pattern was observed for chitosan-enriched treatments. While SADSS+Ch maintained relatively high NUE across growth stages, particularly at low to moderate N rates, BADSS+Ch showed rapid NUE decline with increasing N. After correction, SADSS+Ch remained more stable, whereas BADSS+Ch dropped to very low values at high N loads.

### **Effect of growth stage**

NUE was generally highest at early growth stages (30–60 days) and declined over time, particularly at high N rates. However, some treatments (e.g., BADSS at low N and SADSS+Ch) maintained relatively stable NUE across harvests, indicating sustained nutrient release and uptake synchrony.

### **Corrected vs. uncorrected NUE**

Corrected NUE values clearly demonstrate that accounting for all N inputs is essential. Both EM

and chitosan treatments show reduced efficiency after correction, but SADSS-based systems retain higher NUE stability, whereas BADSS-based systems exhibit strong efficiency collapse.

Overall, SADSS and SADSS+Ch provided the most balanced NUE across doses and time, while EM treatments inflated apparent efficiency unless corrected, and high N inputs consistently reduced NUE across all systems.

### **Conclusions**

The present study demonstrated that pelleted waste-based fertilizers can effectively support forage ryegrass production, although their agronomic performance differed substantially depending on fertilizer composition, enrichment strategy, nitrogen (N) dose, and harvest stage. Overall, SADSS-based fertilizers, particularly those enriched with chitosan (SADSS+Ch), consistently produced the highest biomass yields and the most balanced nitrogen use patterns across the four harvests.

Biomass production generally increased with increasing N application rate up to moderate levels (approximately 120–170 kg N ha<sup>-1</sup>), after which yield responses plateaued or declined in several treatments, especially in BADSS-based systems. The superior performance of SADSS+Ch and, to a lesser extent, SADSS+EM and UBG, was particularly evident during later harvests (90–120 days), indicating sustained nutrient release and improved synchronization between nutrient availability and plant demand. In contrast, BADSS-based fertilizers exhibited lower and more variable productivity, especially at high N inputs, suggesting lower nutrient recovery efficiency and potential nutrient imbalance under excessive fertilization.

The analysis of biomass and nitrogen uptake dynamics further revealed that fertilizer performance depended strongly on temporal nutrient release characteristics. Early growth stages contributed relatively little to total yield and N uptake, while the greatest nutrient demand occurred during the second and third harvests. Treatments enriched with effective microorganisms (EM) stimulated rapid early N uptake, particularly in SADSS+EM and BADSS+EM, but this did not always translate into proportional biomass gains. By contrast, SADSS+Ch combined high cumulative yields with comparatively lower N uptake during later stages, indicating improved internal nitrogen utilization and greater nutrient use efficiency.

Nitrogen Use Efficiency (NUE) declined markedly with increasing N application rates across all treatments, confirming diminishing returns under surplus N supply. The highest NUE values occurred at low N rates, while excessive fertilization substantially reduced nutrient recovery efficiency. Corrected NUE calculations highlighted the importance of accounting for all N inputs associated with enrichment additives. Although EM-enriched treatments initially appeared highly efficient, much of this effect was attributable to additional N introduced through the enrichment process. After correction, SADSS+Ch remained one of the most stable and efficient systems, whereas BADSS-based treatments showed pronounced efficiency losses at high N loads.

Taken together, the results indicate that RNFs derived from waste streams can successfully replace or supplement conventional organic fertilizers when appropriately formulated. Among the tested systems, SADSS+Ch demonstrated the most favourable combination of sustained biomass production, balanced N uptake, and stable NUE across harvests and N doses. These

findings highlight the potential of optimized waste-derived pelleted fertilizers to contribute to circular nutrient management and more sustainable forage production systems, while also emphasizing the importance of matching nutrient release dynamics with crop demand to maximize agronomic efficiency and minimize nutrient losses.

#### 4.2.6 Evaluation centre in Germany (JKI)

##### *Soil*

For the greenhouse pot study, a soil substrate with low phosphorus (P) content, developed at JKI-PB Germany was selected. The soil substrate consists of 88% coarse quartz sand (0.7 – 1.2 mm), 10% fine calcium bentonite, and 2% bark humus (0 – 6 mm). Before starting the pot trial, the soil substrate was homogenized and placed in individual pots. Each pot was filled with 2000 g of soil substrate (dry weight).

##### *Treatment*

For the treatments, the RNF granulated struvite (STR, SF-SoepenberGmbH) was used, also the triple superphosphate (TSP) conventional mineral fertiliser and a P-zero (P0) variant. The experiment was set up using the same doses of phosphorous with the STR and TSP fertilisers at a level of 42 mg P pot<sup>-1</sup>. To reach this target amount, approximately 370 mg struvite and 200 mg TSP were applied. Distilled water was added to the soil substrate to reach 70% maximum water holding capacity. A small quantity of soil was initially mixed with the designated fertiliser, followed by homogenization with the total soil mass within the pot. The sampled soil substrates containing the treatments were then incubated for two weeks at a temperature of 17 ± 5 °C, being the pots covered with a plastic film.

##### *Experiment set up*

After the two weeks of soil incubation in the greenhouse, *Lolium* perennial ryegrass was seeded (22 seed per pot) for the target rate of 1200 seed m<sup>2</sup> (Figure 30). Following this process, the treated and sowed soil substrate was enriched with the macronutrients Mg (30 mg pot<sup>-1</sup>), N (300 mg pot<sup>-1</sup>), S (30 mg pot<sup>-1</sup>), and K (292 mg pot<sup>-1</sup>). Also, the other micronutrients Fe (10 mg pot<sup>-1</sup>), Mn, Zn (1 mg pot<sup>-1</sup>), B (0.5 mg pot<sup>-1</sup>), Cu (0.2 mg pot<sup>-1</sup>), and Mo (0.1 mg pot<sup>-1</sup>) were added to supply the plants. The pots were placed in a randomized block setup to minimize experimental errors. To ensure reliability and reproducibility, each treatment (STR, TSP and P0) was carried out in five replications. Watering was conducted on a regular basis, exclusively with distilled water. Two harvests were applied, each of approximately one month of growing. Figure 30 shows the pots in the greenhouse after seeding (a) and before the 2nd harvest.

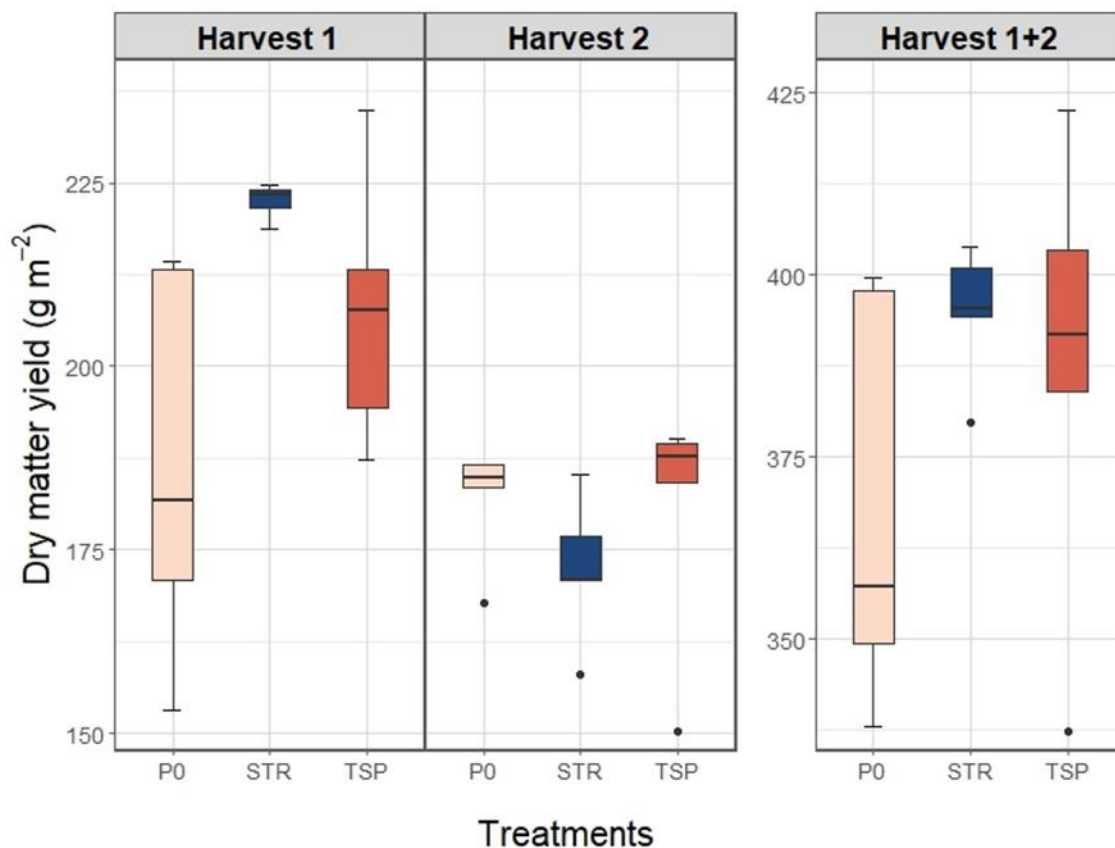
The ryegrass plant material from each harvest was weighed before and after drying at 105 °C to determine plant growth based on forage yield. Furthermore, X-ray micro-computed tomography (μCT) was employed to quantify root development following the two harvests. This allowed the volume and thickness of the roots to be evaluated between the treatments in order to investigate the effect of the RNF struvite underground.



**Figure 30** Pot trial arrangement in the greenhouse just after seeding (a) and before harvest 2 (b).

### Results

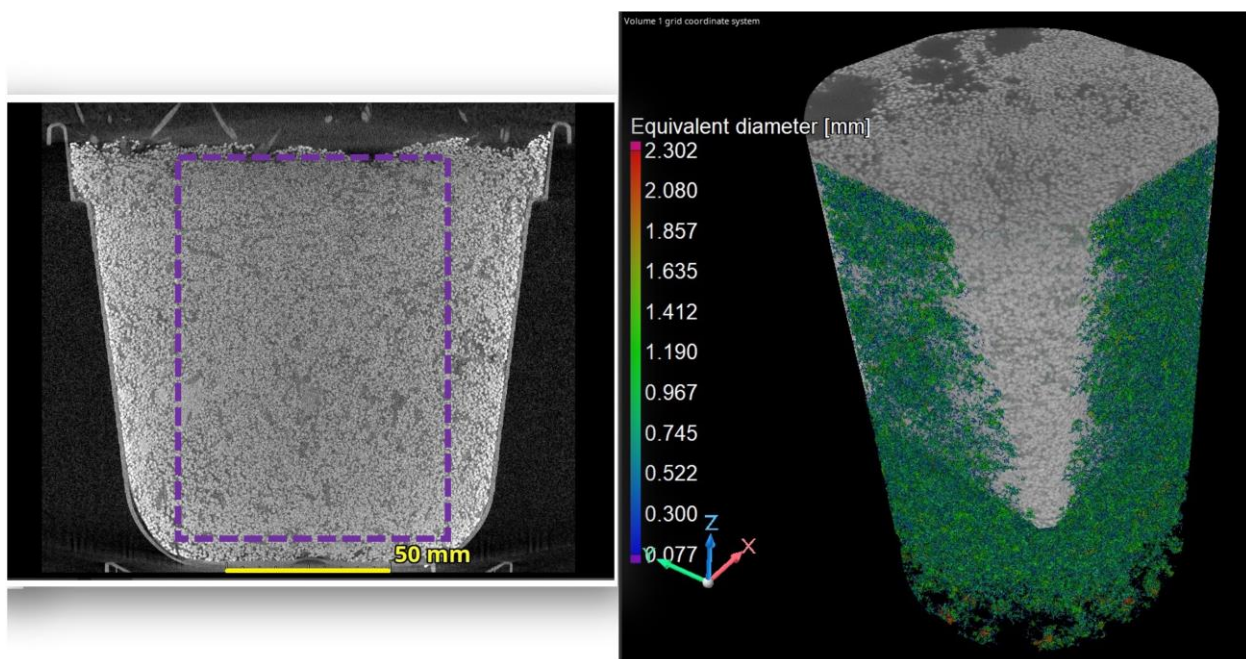
Figure 31 shows the dry matter yield differences between the treatments across the two harvests. In the first harvest, the RNF struvite treatment showed the highest yields, while the unfertilized control produced the lowest median biomass. However, the unfertilized samples exhibited a particularly large variability, with yields ranging widely among replicates. The conventional mineral treatment showed intermediate yields but also a relatively broad spread of values.



**Figure 31** Effect of treatments on the singular and total dry matter yield (g m<sup>-2</sup>) of ryegrass in the greenhouse experiment.

In the second harvest, overall yields were lower than in the first harvest. Differences between treatments were less pronounced. The slightly values for the RNF struvite in the second harvest, suggests a potential reduction in regrowth following high initial biomass production. However, when considering the cumulative yield across both harvests the RNF struvite and the conventional mineral fertilisers are comparable, showing higher overall biomass production than the control. Nevertheless, variability remained substantial, particularly in the control and mineral fertiliser treatment, indicating considerable differences between pots. The cumulative yields therefore reflect both the higher initial productivity of the fertilized treatments and the variability observed among replicates across harvests.

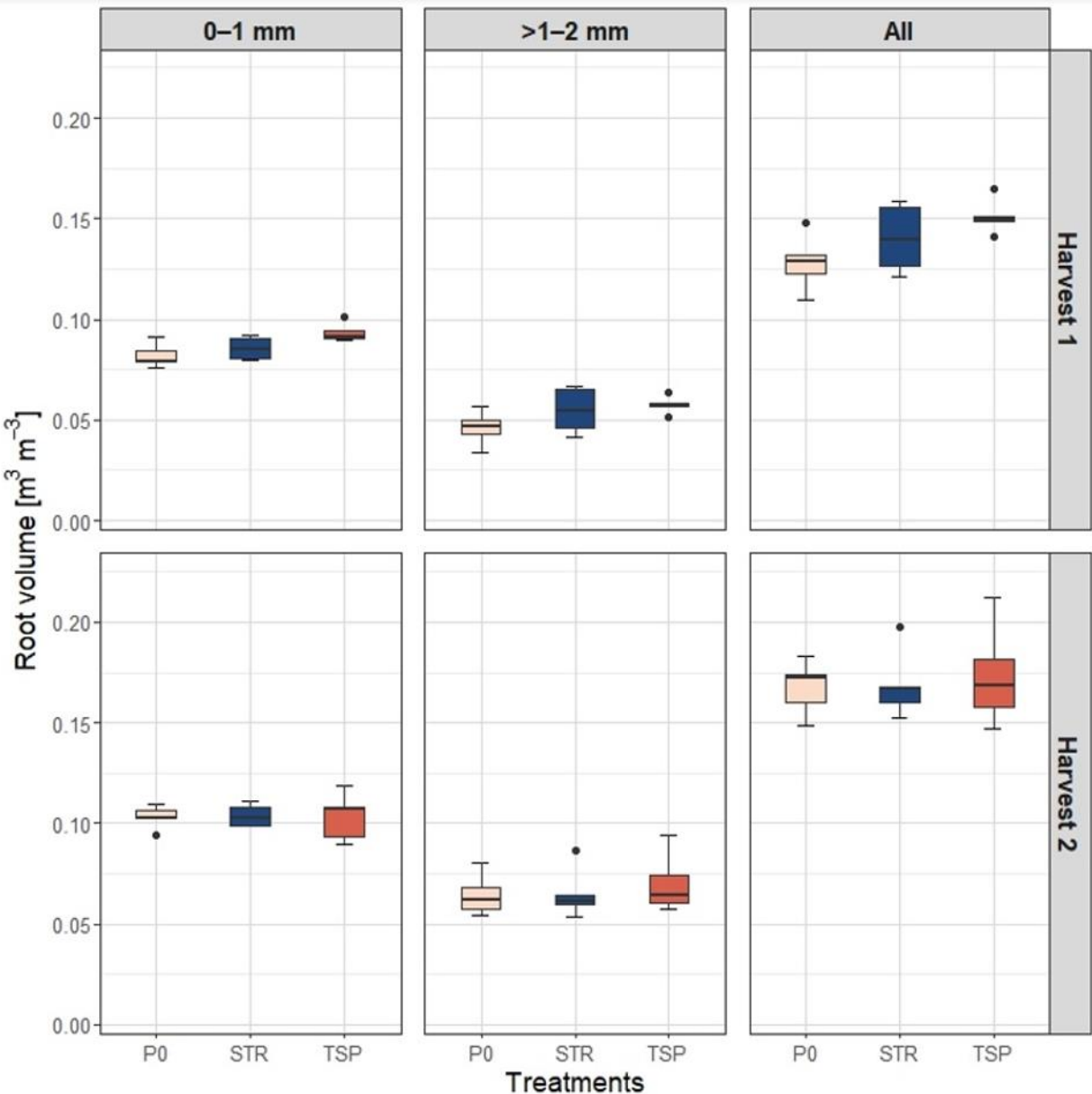
The root development in response to phosphorus treatments was studied by micro-computed tomography ( $\mu$ CT), for that the reconstructed 3D volume data acquired by the  $\mu$ CT technique were imported into the VGStudio Max software for visualization, segmentation, and quantification of root system (Figure 32). The calculation of the roots volume and their equivalent diameter was performed utilizing the watershed algorithm integrated within the software. Figure 32 presents the 2D and 3D image views of a RNF struvite sample and the volume selected for the  $\mu$ CT analyses (region inside the purple lines). The same procedure was than applied to all 15 samples (control, RNF struvite and mineral the triple superphosphate, 5 replicas each).



**Figure 32** A schematic view of a pot plant with the RNF fertiliser is shown, with the  $\mu$ CT scanned image presented. The volume analysed is indicated by the dotted purple lines. On the right side are the segmented roots and their quantified diameters, as determined by the watershed algorithm.

As shown in Figure 33, root volume varied among treatments, diameter classes, and harvest times. Overall, root volume increased from the first to the second harvest across all diameter classes, indicating a continued development of the root system over time. At the first harvest, the finest root fraction (0–1 mm) showed slightly higher volumes in the fertilized treatments compared with the control. The mineral fertiliser displayed the highest values, followed by the

RNF struvite, whereas the control showed the lowest root volumes. Variability within treatments was moderate, although some spread of values among replicates was visible. In the intermediate diameter class (>1–2 mm), root volumes at the first harvest were relatively similar among treatments. The RNF struvite tended to show somewhat higher values than the control and the mineral fertiliser, while the overall spread of observations indicated moderate variability between replicates. When considering the total root volume across all diameter classes, the fertilized treatments again showed higher values than the control at the first harvest. Both the RNF and the mineral fertiliser tended to produce larger overall root volumes than the control. At the second harvest, the differences were less pronounced, however root volume increased in all treatments and diameter classes compared with the first harvest.



**Figure 33** Root volume and their thickness between the treatments after first and second harvest, measured by X-ray  $\mu$ CT.

## Conclusion

The greenhouse pot experiment demonstrated that phosphorus fertilization influenced both biomass and roots plant development of *Lolium perenne*:

- In the first harvest the recycled nutrient fertiliser struvite showed that the highest and less variable dry matter yield values. Although the values after the second harvest were slightly lower for the RNF struvite compared with the other treatments, the cumulative yield values remained comparable to the conventional mineral fertiliser and much higher than the unfertilized control.
- Root analyses using  $\mu$ CT showed that root volume increased over the two harvests in all treatments, reflecting ongoing root system development. Fertilized treatments generally exhibited larger root volumes than the control during the first harvest, while differences between treatments became less pronounced at the second harvest.
- Overall, we observed that the RNF struvite can support plant growth and root development at a level comparable to the conventional mineral phosphorus fertiliser. Despite some variability between replicates, the agronomic performance of the analysed RNF struvite in granules was consistently higher than the unfertilized control and comparable to conventional mineral fertilization.

## 5. General conclusions and recommendations

The results obtained in Evaluation centres established in different countries BSR confirm that the agronomic value of the tested RNFs depends strongly on nutrient form, release dynamics and soil conditions, but several products showed clear potential to substitute mineral fertiliser. Struvite, tested in Germany, Sweden and Estonia, behaved consistently as a slow-release phosphorus fertiliser. In incubations it released P more gradually than soluble mineral P, but in greenhouse studies it still supported substantial plant P uptake and biomass production, especially where soil conditions favoured P acquisition. Across countries, struvite therefore showed a high P fertiliser value but not always immediate equivalence to superphosphate, meaning its performance was generally closer to a medium-to-high P-mineral fertiliser equivalent (P-MFE) than a full one-to-one replacement under all soils. The Swedish and Estonian results particularly support its role as a reliable recovered P source, while the German results strengthen this conclusion under an additional pedoclimatic context.

Dry urine, tested in Poland and Sweden, showed a different pattern. Because its N is more readily plant-available, it behaved more like a fast-acting N fertiliser, with rapid N supply and correspondingly good crop response. The results indicate that dry urine had comparatively high N-mineral fertiliser equivalent (N-MFE), in some cases approaching mineral fertiliser performance more closely than the slower-release P-oriented products. At the same time, its P value was less central than its N value. This makes dry urine particularly relevant where quick early N supply is needed, though storage and handling remain important because drying can affect ammonium conservation.

For multi-waste pellets, tested in Sweden and Poland, the validation is more nuanced but still

positive. These pellets provided agronomic value in both countries, yet their response was more variable than for struvite or dry urine because nutrient release depends on pellet composition, degree of processing, and the balance between organic and mineral nutrient forms. In general, the pellets acted as intermediate-release fertilisers: less immediately available than mineral fertiliser or dry urine, but able to support plant growth and nutrient uptake over time. Their mineral fertiliser equivalent was therefore moderate and product-specific, rather than uniformly high. The Swedish and Polish datasets together show that these materials can substitute part of mineral fertiliser demand, but product standardisation is critical.

The partners who tested other RNFs also confirmed agronomic value across a broader group of recycled products. Digestate fractions and urine-based granules tended to provide rapid N availability, while composts, biochars and some pellets contributed more through slower nutrient release and soil-improving effects than through immediate crop response. This means agronomic value should not be judged only by short-term yield: some RNFs function mainly as quick nutrient suppliers, whereas others combine nutrient recycling with improved carbon retention, buffering capacity or residual nutrient effects. Overall, the multicountry evidence supports that RNFs are not a single category but a spectrum of fertiliser products with different substitution niches.

In the Polish comparison with organic fertiliser as reference, the results suggest that the tested RNFs were generally more predictable and in several cases more agronomically efficient than conventional organic fertiliser. This is mainly because nutrient content in RNFs was more concentrated and their nutrient release was better aligned with plant demand especially when RNFs were improved by different additives (EM and chitosan). Organic fertiliser still contributed useful nutrients and organic matter, but its effect was usually less direct and more dependent on mineralisation conditions. The Polish study therefore supports the view that well-processed RNFs can bridge the gap between traditional organic fertilisers and mineral fertilisers.

Based on the incubation studies, carbon stability differed markedly among RNFs and was closely linked to the degree of processing and the share of easily degradable organic matter. Materials containing more labile organic compounds, such as some digestate-derived and mixed organic products, stimulated stronger early CO<sub>2</sub> emissions after soil application, indicating faster microbial decomposition and therefore lower carbon stability in soil. In contrast, more processed materials such as biochar-based products and, to a lesser extent, struvite, contributed little to microbial respiration because they contained either highly recalcitrant carbon or almost no carbon at all; these products were therefore associated with high carbon stability but limited contribution to short-term microbial turnover. Pelletised and dried RNFs generally showed intermediate behaviour, with carbon mineralisation proceeding more gradually depending on feedstock composition and pellet structure. Overall, the incubation results show that RNFs cannot be treated as a single class with respect to soil carbon dynamics: some function mainly as short-term nutrient and microbial substrates, while others behave as more stable carriers of carbon or nutrients, which is important when assessing their role not only as fertilisers but also as contributors to longer-term soil organic matter management.

The developed incubation and greenhouse protocols were sufficiently robust to assure relevant comparison across locations, even though some local adaptation remained necessary. Common

rules for soil preparation, RNF characterisation, equal nutrient application, moisture control, incubation timing and pot trial design created a sound basis for comparing nutrient release, plant uptake and MFE values between countries. The fact that similar functional patterns emerged across sites for struvite, urine-based products and pellets indicates that the methodology worked. However, differences in soil pH, texture, crop choice and local greenhouse conditions still influenced absolute values, so the protocol is best seen as enabling comparable interpretation, not perfect numerical uniformity.

More broadly, the tested RNFs have strong potential to substitute mineral fertilisers because they can deliver plant-available N and P while recycling nutrients from waste streams. Their main strengths are circularity, reduced dependence on mined or industrial fertilisers, and in some cases improved handling through drying, granulation or pelletisation. Their limitations are also clear: nutrient availability may be slower or more variable than in mineral fertilisers; contaminants and hygienic safety must be controlled; and usability depends on moisture content, storage stability and compatibility with spreading equipment. Products such as struvite stand out for clean, stable, P-focused recovery, whereas urine-derived and multi-waste products are promising but require tighter quality assurance.

In relation to the quality standards outlined in the report *“Draft industry standards for quality assurance of recycled nutrient fertilizers”*, the RNFs tested in this study span several relevant component material and product function categories, including recovered mineral fertilisers such as struvite, processed organic fertilisers like composts and digestates, and organo-mineral-type pellet products. Stability was partly addressed through incubations and indirectly through moisture content: dry and pelletised products are generally more storable, while wetter or biologically active materials need more careful preservation. Hygienization is particularly relevant for urine- and waste-derived products and drying or thermal processing can support this. Heavy metals remain an essential criterion, especially for sewage-sludge-based materials and mixed waste pellets, meaning agronomic validation must always be accompanied by contaminant compliance. Storability is closely linked to moisture content, as low-moisture products are easier to store, transport and apply. Finally, application technique is a practical determinant of success: granules and pellets are better suited to conventional spreaders, while nutrient availability must be judged both directly from plant response and, for N, indirectly from incubation mineralisation and release data.

## 6. Dissemination

Dissemination activities within A2.2 combined online communication with extensive offline engagement, reflecting the task’s objective to not only share research findings but also to establish an interactive, cross-sectoral network around RNF development. All evaluation centres contributed to online dissemination through LinkedIn posts and project website updates, ensuring continuous visibility of the pot and incubation trials and enabling broad access to preliminary results. These posts introduced diverse audiences to the aims of A2.2, highlighted ongoing experimental work, and encouraged dialogue on nutrient recycling solutions. Partners also used their institutional channels to share national updates, such as the Estonian communication on the METK struvite trial, thereby strengthening the project’s digital outreach

across the BSR.

Offline dissemination was closely aligned with the activity's requirement to engage target groups through a bottom-up approach and to create feedback loops that inform the development of guideline industry standards. In Sweden, the results of the pot experiment were presented at the EUROSOIL 2025 Conference, including a dedicated student session, fostering exchange with both researchers and future agricultural professionals. Further interactions with farmers, advisors, and industry representatives took place during Borgeby Fältdagar (Field days) and a field demonstration day at SLU's Lana research station, where discussions centred on practical challenges and opportunities related to RNF use under different crop and rotation systems. The findings were also integrated into SLU's teaching programmes, embedding RNF knowledge in long-term educational pathways.

In Finland, the A2.2 results were shared within Luke's internal expert networks and incorporated into broader national dissemination activities, contributing to ongoing dialogue on nutrient recycling and circular bioeconomy practices. Estonia supported cross-sectoral engagement by communicating the METK trial results to professional audiences, helping to raise awareness of RNF performance under local conditions.

Polish partners undertook extensive dissemination through international and national events, presenting greenhouse results at the ICDTSA 2026 conference, where the contribution received awards and sharing insights at the Bioeconomy 2030+ event and a local conference on digestate management. Study visits to research institutes in Italy and Spain further expanded the project's interactive network, enabling exchange with experts working on circular economy and biowaste valorisation. These engagements provided valuable feedback on RNF applicability across different agricultural and climatic contexts.

In Germany, dissemination was embedded in the ongoing research and demonstration activities at JKI-PB, where the pot trial contributed to broader discussions on evaluating P-rich fertilisers and sustainable nutrient management. The results were shared within the institute's research community, informing joint evaluation strategies and methodological development. Regular agronomic demonstrations at JKI-PB facilitated direct interaction with farmers, advisors, students, and policymakers, supporting the project's aim to build a cross-sectoral network of interest. The work also led to the preparation of two scientific papers, soon to be submitted to peer-reviewed journals, further strengthening scientific dissemination.

## References

- Ahmed, H.A., El-Maradny, Y.A., Shalaby, M.A., El-Menshawy, H., Abd EL-Wahab, A.E., 2025. *Isolation and characterization of Chitosan from shrimp shell waste and the sustainable preparation of salicylic acid-loaded Chitosan nanoparticles for antibiofilm applications*. *Sci. Rep.* 15, 1–17. <https://doi.org/10.1038/s41598-025-03355-3>
- Antonino, R.S.C.M.D.Q., Fook, B.R.P.L., Lima, V.A.D.O., Rached, R.Í.D.F., Lima, E.P.N., Lima, R.J.D.S., Covas, C.A.P., Fook, M.V.L., 2017. *Preparation and characterization of chitosan obtained from shells of shrimp (Litopenaeus vannamei Boone)*. *Mar. Drugs* 15, 1–12. <https://doi.org/10.3390/md15050141>
- Bernal, P.M., Sommer, S.G., Chadwick, D., Qing, C., Guoxue, L., Michel, F.C., 2017. *Advances in Agronomy*. *sciencedirect* 144, 143–233. <https://doi.org/10.1016/bs.agron.2017.03.002>
- Buckwell, A. Nadeu, E. 2016. *Nutrient Recovery and Reuse (NRR) in European agriculture. A review of the issues, opportunities, and actions*. RISE Foundation, Brussels.
- European Commission, Eurostat., 2024. *Consumption of inorganic fertilisers in agriculture*. [https://doi.org/10.2908/AEI\\_FM\\_USEFERT](https://doi.org/10.2908/AEI_FM_USEFERT)
- Fertilizers Europe. (2024). *Forecast of food, farming & fertilizer use in the European Union 2024–2034*. Brussels: Fertilizers Europe
- Dijk, K.C. van, Lesschen, J.P., Oenema, O., 2016. *Phosphorus flows and balances of the European Union Member States*. *Science of the Total Environment* 542, 1078–1093. <https://doi.org/10.1016/j.scitotenv.2015.08.048>
- Draft industry standards for quality assurance of recycled nutrient fertilizers. Report CiNURGi project <https://interreg-baltic.eu/project/cinurgi/#output-5>
- Hu, X., Tian, Z., Li, X., Wang, S., Pei, H., Sun, H., Zhang, Z., 2020. Green, Simple, and Effective Process for the Comprehensive Utilization of Shrimp Shell Waste. *ACS Omega* 5, 19227–19235. <https://doi.org/10.1021/acsomega.0c02705>
- Kambo, H.S., Dutta, A., 2015. A comparative review of biochar and hydrochar in terms of production, physico-chemical properties and applications. *Renew. Sustain. Energy Rev.* 45, 359–378. <https://doi.org/10.1016/j.rser.2015.01.050>
- Papandrea, S.F., Cataldo, M.F., Palma, A., Gallucci, F., Zimbalatti, G., Proto, A.R., 2021. Pelletization of Compost from Different Mixtures with the Addition of Exhausted Extinguishing Powders. *Agronomy* 11, 1357. <https://doi.org/10.3390/agronomy11071357>
- Sung-inthara, T., Juntahum, S., Senawong, K., Katekaew, S., Laloon, K., 2024. Pelletization of soil amendment: Optimizing the production and quality of soil amendment pellets from compost with water and biochar mixtures and their impact on soil properties. *Environ. Technol. Innov.* 33, 103505. <https://doi.org/10.1016/j.eti.2023.103505>
- Tayibi, S., Monlau, F., Bargaz, A., Jimenez, R., Barakat, A., 2021. Synergy of anaerobic digestion and pyrolysis processes for sustainable waste management: A critical review and future perspectives. *Renew. Sustain. Energy Rev.* 152, 111603. <https://doi.org/10.1016/j.rser.2021.111603>

Verardi, A., Sangiorgio, P., Moliterni, S., Errico, S., Spagnoletta, A., Dimatteo, S., 2023. Advanced technologies for chitin recovery from crustacean waste. *Clean Technol. Recycl.* 3, 4–43. <https://doi.org/10.3934/ctr.2023002>

## Appendix

Table 1. Basic characteristics of EM boosters/ enrichments applied to both BADSS and SADSS.

Bakery/ Dairy Waste + EM	Unit	Value	SD
<b>SOLID</b>			
Dry matter, d.m.	% fresh matter	11,08	0,47
Organic matter, o.m.	% fresh matter	93,891	0,467
Total Nitrogen, TN	g N/ kg d.m.	27,14	0,03
Orto-Phosphate, PO43-	mg P/ L	330,37	6,99
Total Phosphorus	mg P/ L	1021,17	18,87
<b>LIQUID</b>			
Total Nitrogen, TN	g N/ L	0,810	0,086
Total Phosphorus, TP	mg P/ L	449,40	1,1547
pH		4,69	NA
Redox potential	mV	138,10	NA

Table 2. Basic characteristics of chitosan enrichments applied to both BADSS and SADSS.

Fresh seafood Waste (shrimp shell)	Unit	Value	SD
Dry matter, d.m.	% fresh matter	14,840	0,003
Organic matter, o.m.	% fresh matter	94,218	0,959
Fresh matter (booster addition)	g/ pot	6,82	
Dry matter (booster addition)	g/ pot	1,01	
Chitozan from booster addition*	g/ pot	0,20	0,10
Total Nitrogen, TN	g N/ kg TS	90,932	0,780
Orto-Phosphate, PO43-	mgP/ L	ND	NA
Total Phosphorus	mgP/ L	ND	NA

\* based on the literature values