Universitat Autònoma de Barcelona

Alternative fertilizers for urban agriculture within the circular economy framework

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Alternative Fertilizers for Urban Agriculture within the Circular Economy Framework

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Doctoral thesis

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A thesis submitted in fulfilment of the requirements for the Doctoral degree in Environmental Sciences and Technology

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Canto al bosc atent,
Sadoll de peixos, llebres, ceps.
Canto als dies magnànims,
A la brisa d'estiu, a la brisa d'hivern,
Als matins, a les vesprades,
A la pluja petita, a la pluja enfadada.
Canto a la vessant, al cim, al prat,
A les ortigues, al roser bord, a l'esbarzer.
Canto com qui fa hort,
Com qui talla una taula,
Com qui aixeca una casa,
Com qui tresca un pujol,
Com qui es menja una nou,
Com qui encén una brasa.

IRENE SOLÀ-
Canto jo i la muntanya balla

The present thesis entitled Alternative fertilizers for urban agriculture within the circular economy framework by Verónica Arcas Pilz, has been carried out at the Institute of Environmental Science and Technology (ICTA) at Universitat Autònoma de Barcelona (UAB).

Verónica Arcas Pilz

Under the supervision of Dr. Xavier Gabarrell and Dra. Gara Villalba, from the ICTA and the Department of Chemical Engineering at the UAB.
Bellaterra (Cerdanyola del Vallès), June 2022

Xavier Gabarrell Gara Villalba

Table of contents

List of abbreviations and Acronyms

1.4 DB eq – 1,4 dichlorobenzene equivalent emission
AMB- Area metropolitana de Barcelona
ATP- Adenosine triphosphate
Ca -Calcium
Ca^{2+} - Calcium ion
$Ca(NO_3)_2$ – Calcium nitrate
$CaCl_2$ – Calcium cloride
CO_2 eq – Carbon dioxide equivalent emissions
DAP- Days after planting
DAT- Days after transplanting
DNA – Deoxyribonucleic acid
DOI – Direct Object Identifyer
DWCT- Deep water culture technique
E- Egalitarian
EC- Electric conductivity
EDAR- Estació depuradora d'aigues residuals / Estación depuradora de aguas residuales
EF- Emission Factor
ET- Ecotoxicity Impact Category
EU – European Union
FDP – Fossile Depletion Impact Category
FE- Freshwater eutrophication Impact Category
FU- Functional Unit
GH- Greenhouse
GHG- Greenhouse Gases
GIS – Geographical Information System
GROOF- Greenhouses to reduce CO_2 on roofs
GW – Global Warming Impact Category
H- Hierarchical
HDPE – High density Polyethilene
I- Individualist
IC- Impact Category
ICP- Institut pleontológic de Catalunya
ICP- OES – Inductively coupled Plasma Optical Emission Spectroscopy
ICS – Ion Chromatography System
ICTA- Institut de ciencia I technologia ambiental
IPCC – Intergovernamental Panel on Climate Change
i-RTG – Integrated Rooftop Greenhouse
ISO – International Organization for Standarization

K – Potassium
K_2SO_4 – Potassium sulphate
KNO_3 – Potassium nitrate
LAI- Leaf area Index
LAU-Laboratory of Urban Agriculture in ICTA
LCA- Life Cycle Assessment
LCI – Life Cycle Inventory
LCIA – Life Cycle Impact Assessment
MAP – Magnesium ammonium phosphate
ME – Marine Eutrophication Impact Category
Mg – Magnesium
Mg^{2+} - Magnesium ion
$Mg(NO_3)_2$ – Magnesium nitrate
N - Nitrogen
N eq – Nitrate equivalent emissions
N_2O – Nitrous oxide
NE- Northeast
NFT- Nutrient film technique
NH_4^+ - ammonium ion
NO_3^- - Nitrate
NO_2^- - Nitrite
Oil eq – Oil equivalent
P - Phosphorous
P eq – Phosphorous equivalent emission
PE – Polyethilene
PO_4^{3+} - Phosphate ion
RH – Relative humidity
RNA – Ribonucleic acid
RWHS- Rainwater Harvesting System
RTG – Rooftop Greenhouse
Sostenipra – Sustainability and Environmental Prevention research group
SW- Southwest
TA – Terrestrial Acidification Impact Category
TSP- Triple Superphosphate
UA- Urban Agriculture
UAB – Universitat Autònoma se Barcelona
UNEP – United Nation Environment Programme
WOS- Web of Science
WWTP- Wastewater Treatment Plant

List of figures

List of tables

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Summary

Increasing population and demands on agriculture lead to the search for alternative food systems that can help supply food without further compromising the environment. The search for food production without further use of land and resources has made urban agriculture (UA) a good candidate to help solve this problematic. The implementation of UA can also entail other benefits that range from food security and resilience to well-being and sense of community.

Additional to the production of food UA has been regarded as a key player to increase circularity in urban areas. From a circular economy perspective, the addition of UA can serve as a sink for locally sourced materials to cover nutrient or substrate requirements, while at the same time reduce food transport and waste management.

This can be a crucial point, especially regarding the environmental footprint of nutrient synthetization and extraction, like in the case of nitrogen (N) and phosphorous (P) commonly used in hydroponic and aeroponic agriculture. The production and extraction of these nutrients are energy consuming and polluting activities increasing the footprint of these agricultural production systems. In the case of P, it is also considered a nonrenewable resource

The expansion and up scaling of UA can be beneficial for multiple reasons and on more than one level, but it is important to ensure the sustainability of the activity to avoid an impact shift.

The main objective of the present dissertation is the reduction of nonrenewable/ polluting inputs for the implementation of more sustainable UA production, within the circular economy framework.

To fulfill this main objective, the present dissemination aims to give an answer to the following research questions:

Q1 → What possibilities are available in urban areas to reduce environmental impact of fertilization in hydroponic production taking in account the circular economy framework?

Q2 → Are alternative fertilization methods effective in hydroponic production? Is it possible to maintain the commercial production?

Q3 → To what extent do we improve our production system? How much can we decrease our environmental footprint?

The materials and methodologies used in this dissertation will be explained in the following section, followed by the obtained results and the proposed further research.

Materials and methods used

The methodologies used in this research can be divided in four main sections: literature review, agronomic, environmental, and statistical which are used in different combinations throughout the dissertation.

Within these methodological approaches different materials have been used mainly outlining agricultural production, nutrient content analysis in biomass and water, N_2O emissions through gas chromatography and environmental assessment through LCA tools.

Increasing urban circularity: Nutrient recovery from urban waste

The present dissertation defines nutrient recovery technologies from urban waste found in current literature. A primary definition of the main waste sources is made dividing them into two sections: "wastewater" and "organic-, bio- food waste". From these two section a total of 18 recovery were found under the defined selection criteria, mainly focusing on the recovery of N and P. The current potential of the metropolitan area of Barcelona to recover N and P from waste is further determined specially from the wastewater flows and existing waste management and composting sites. If these recovered nutrients were to be applied to the existing and prospective UA sites results showed that the necessary P could be supplied 2,7 to 380,2 times while N 1,7 to 117,5 depending on the recovery strategy. P depletion and high energy cost for N synthesis make recovery strategies more necessary although current legislations still hider these technologies.

Phosphorous recovery in the form of struvite and its application in hydroponic agriculture.

The feasibility of using struvite precipitated from an urban wastewater treatment plant as the unique source of P fertilizer was assessed in this dissertation with green bean (*Phaseolus vulgaris*) peppers (*Capsicum annum* L.) and lettuce (*Lactuca sativa* L.).

In the case of green bean, we apply various quantities of struvite (ranging from 1 to 20 g/plant) to the substrate and assessed the production, water flows and P balances. The results obtained indicate that with more than 5g of struvite a higher yield is obtained (maximum of 181.41 g/plant) than the control (134.6 g/plant) fertilized with mineral fertilizer (KPO_4H_2). P concentrations detected in all plant organs remained lower when using struvite. Finally, different amounts of struvite remained undissolved in all treatments which indicated a great potential to grow further cropping cycles as well as a reduced concentration of P in the leachates.

Also, for pepper and lettuce crops different quantities were tested (5g, 10g and 20g per plant) and compared to a control fertilized with monopotassium phosphate. The experiment took 3 months, with 3 lettuce cycles and one long pepper cycle fertilized with the initial struvite given at the beginning of the experiment. The resulting yields obtained were competitive compared to the control, especially during the first lettuce harvest (225.5g, 249.9g, 272.6g, and 250g for 5g, 10g, 20g and control, respectively) where a greater struvite dissolution was seen. Although, the P content in the pepper biomass resulted low the productions are close or even higher than the control like in the case of 20g struvite treatment (3.6kg, 4.3kg, 7.5kg and 5.3kg for 5g, 10g, 20g and control, respectively).

The findings of these fertilization studies showed the potential and feasibility of P fertilization with struvite, a locally recovered nutrient, in hydroponic agriculture systems.

The combination of struvite and rhizobia inoculation in *Phaseolus vulgaris* production, reducing N and P needs in hydroponic fertilization.

A combination of struvite fertilization (2g and 5g treatments) and rhizobium inoculations were tested on green bean to uncover to possibilities to fully substitute N, P and Mg fertilization through fertirrigation. A control treatment was added with a full nutrient solution (N, P and Mg added in the irrigation).

The variables of plant growth, development, nutrient content, and bean production were assessed in time at three different days (35, 62 and 84). To understand the N origin in the plant the biological N_2 fixation was also determined using the 15N natural abundance method. The struvite treatments obtained lower total yields compared to the control (e.g., 59.35± 26.4 g plant-1 for 2g, 74.2±23.0 g plant-1 for 5g and 147.71± 45.3 g plant-1 for control).

Rhizobium nodulation and N_2 fixation capacities increased with increasing struvite amounts, showing Mg and P deficit in the plants over time. Although these deficiencies could explain the lower yields obtained the combination showed promising results since the N content in the struvite seamed to reinforce and not inhibit the rhizobium nodulation.

An environmental approach to struvite fertilization on hydroponic production.

The environmental approach using LCA was performed mainly in two experiments using struvite fertilization for lettuce and pepper plants (with 5g, 10g and 20g of struvite treatments) as well as beans with the combination of struvite and rhizobium inoculation (with 2g, 5g, 10g and 20g of struvite treatments).

In the case of the lettuce crops the results show a reduction of almost all impact categories (IC) when using struvite as a P fertilizer compared to the control treatment. A significant reduction was seen for the categories of freshwater eutrophication and mineral resource scarcity. For peppers a reduction in all impact categories was seen with 10g and 20g of struvite.

When a year-round production is assumed for lettuce, we can see that with an initial amount of 20g the slow dissolution of struvite can sustain competitive production for 9 crop cycles also meaning less impact in all categories except marine eutrophication.

In the case of the environmental performance of struvite and rhizobium combination in green bean production, to replace P and N fertirrigation, results indicate a yield reduction of 60% to 50% in comparison to the control which was irrigated with a full nutrient solution. This yield reduction greatly increases the environmental impact of the alternatively fertilized treatments specially experiencing an increase in the infrastructure related emissions. An estimation on the yield loss “allowed” to remain a more sustainable fertilization choice would be 10% below the control yield.

Proposed further research. What’s next?

To further dwell on the main goals proposed in this dissertation five main roads of research have been detected where further research could be made.

Firstly, the increase of urban circularity with the exploration of other urban sourced materials to be used in hydroponic agriculture as fertilizer as well as substrate.

Secondly, the increase of P availability given though struvite with the combination with bacteria, from phosphate solubilizing bacteria to further research on the combination with rhizobium. In this sense further treatments with higher struvite amounts could be tested as well as the use of different chemical compounds in the nutrient solution to avoid chemical imbalances in the rhizosphere.

Thirdly, the sustained use of struvite during longer cropping periods and different (more and less demanding) crops. As well as the use of struvite as a N fertilizer for less demanding crops.

Fourthly, the knowledge on the use of struvite and its integration in the market and “normality” in urban agriculture could be highly enriched with a social study on the public perception on the use of this crystal in agriculture.

Lastly, as a fifth proposed research topic, further work can be done on the potential emissions generated through struvite fertilization. This would entail further work on the N2O emissions and potential influence from inoculated bacteria like rhizobium as well as the comparison of environmental impact of struvite fertilization and control fertilization in long crop cycles like tomatoes.

Resum

L'augment de la població i les demandes a l'agricultura porten a la recerca de sistemes alimentaris alternatius que puguin ajudar a subministrar aliments sense comprometre encara més el medi ambient. La recerca de la producció d'aliments sense més ús de la terra i els recursos ha fet de l'agricultura urbana (UA) un bon candidat per ajudar a resoldre aquesta problemàtica. La implementació de la UA també pot comportar altres beneficis que van des de la seguretat alimentària i la resiliència fins al benestar i el sentit de comunitat.

A més de la producció d'aliments, la UA s'ha considerat com un actor clau per augmentar la circularitat a les zones urbanes. Des d'una perspectiva d'economia circular, l'addició d'UA pot servir com a destí per a materials d'origen local per cobrir les necessitats de nutrients o substrats, alhora que redueix el transport d'aliments i la gestió de residus.

Aquest pot ser un punt crucial, especialment pel que fa a la petjada ambiental de la síntesi i extracció de nutrients, com en el cas del nitrogen (N) i el fòsfor (P) utilitzats habitualment en l'agricultura hidropònica i aeropònica. La producció i extracció d'aquests nutrients són activitats consumidores d'energia i contaminants augmentant la petjada d'aquests sistemes de producció agrícola. En el cas de P, també es considera un recurs no renovable

L'expansió i l'ampliació de la UA pot ser beneficiosa per múltiples motius i en més d'un nivell, però és important garantir la sostenibilitat de l'activitat per evitar un canvi d'impacte.

L'objectiu principal de la present tesi és la reducció d'inputs no renovables/contaminants per a la implementació d'una producció d'UA més sostenible, en el marc de l'economia circular.

Per complir amb aquest objectiu principal, la present difusió pretén donar resposta a les següents preguntes de recerca:

P1 → Quines possibilitats hi ha a les zones urbanes per reduir l'impacte ambiental de la fertilització en la producció hidropònica tenint en compte el marc de l'economia circular?

P2 → Són efectius els mètodes alternatius de fertilització en la producció hidropònica? És possible mantenir la producció comercial?

P3 → Fins a quin punt millorem el nostre sistema de producció? Fins a quin punt podem reduir la nostra petjada ambiental?

Els materials i metodologies utilitzats en aquesta tesi s'explicaran a la secció següent, seguit dels resultats obtinguts i de la recerca posterior proposada.

Materials i mètodes utilitzats

Las metodologies utilitzades en aquesta investigació es poden dividir en quatre secciones principals: revisió de la literatura, agronòmica, ambiental y estadística, que s'utilitzen en diferents combinacions a la llarga de la tesi.

Dins d'aquests enfocaments metodològics s'han utilitzat diferents materials que descriuen principalment la producció agrícola, l'anàlisi del contingut de nutrients en biomassa i aigua, les emissions de N2O a través de cromatografia de gasos i l'avaluació ambiental a través d'eines LCA.

Augment de la circularitat urbana: recuperació de nutrients dels residus urbans

La present tesis defineix les tecnologies de recuperació de nutrients a partir de residus urbans trobats a la literatura actual. Es realitza una primera definició de les principals fonts de residus dividides en dos apartats: "aigües residuals" i "residus orgànics, bio- alimentaris". Aquestes dues seccions es trobaran un total de 18 recuperacions sota els criteris de selecció definits, centrant-se principalment en la recuperació de N i P. El potencial actual de l'àrea metropolitana de Barcelona per a recuperar N i P dels residus es determina a més especialment de les aigües residuals. fluxos i llocs de compostatge i gestió de residus existents. Si aquests nutrients recuperats s'apliquen als llocs d'UA existents i prospectius, els resultats mostren que el P necessari podria administrar-se de 2,7 a 380,2 vegades mentre que el N de 1,7 a 117,5 depenent de l'estratègia de recuperació. L'esgotament de P i l'alt cost energètic per a la síntesi de N fa més necessària les estratègies de recuperació, encara que la legislació actual encara no incentiva aquestes tecnologies.

Recuperació de fòsfor en forma d'estruvita i la seva aplicació en agricultura hidropònica.

En aquesta tesi doctoral es va avaluar la viabilitat d'utilitzar l'estruvita precipitada d'una planta de tractament d'aigües residuals urbanes com a única font de fertilitzant fosforat amb mongetes verdes (*Phaseolus vulgaris*), pebrots (*Capsicum annum L.*) i enciam (*Lactuca sativa L.*) .

En el cas de la mongeta verda, apliquem diverses quantitats d'estruvita (que oscil·la entre 1 i 20 g/planta) al substrat i avaluem la producció, els fluxos d'aigua i l'ús de P. Els resultats obtinguts indiquen que amb més de 5 g. d'estruvita s'assoleix un major rendiment (màxim de 181,41 g/planta) que el control (134,6 g/planta) fertilitzat amb fertilitzants mineral (KPO_4H_2). Les concentracions de P detectades en tots els òrgans de la planta es mantenen més baixes quan es fa servir l'estruvitat. Finalment, diferents quantitats d'estruvita van quedar al substrat sense dissoldre per tots els tractaments, el que va indicar un gran potencial per desenvolupar cicles de cultiu posteriors, així com una concentració reduïda de P en els lixiviats.

Tanmateix, per a cultius de pebrot i enciam s'han fet servir diferents quantitats d'estruvita (5g, 10g i 20g per planta) i s'han comparat amb un control fertilitzat amb fosfat monopotàssic. L'experiment ha durat 3 mesos, amb 3 cicles d'enciam i un cicle llarg de pebre fertilitzat amb l'estruvita inicial donada al començament de l'experiment. Els rendiments obtinguts van ser competitius en comparació amb el control, especialment durant la primera collita d'enciam (225,5 g, 249,9 g, 272,6 g i 250 g per a 5 g, 10 g, 20 g i control, respectivament) on es va observar una major dissolució de l'estruvita. Tot i que el contingut de P en la biomassa de pebrot va resultar baix, les produccions són properes o fins i tot superiors al control com en el cas del tractament amb 20g d'estruvita (3,6kg, 4,3kg, 7,5kg i 5,3kg per a 5g, 10g, 20g i control). respectivament).
Les troballes d'aquests estudis de fertilització van mostrar el potencial i la viabilitat de la fertilització amb P amb estruvita, un nutrient recuperat localment, en sistemes d'agricultura hidropònica.

La combinació de la inoculació d'estruvita i rhizobium en la producció de Phaseolus vulgaris, redueix les necessitats de N i P en la fertilització hidropònica.

Es va provar una combinació de fertilització d'estruvita (tractaments de 2 g i 5 g) i inoculacions de rizobi sobre mongeta verda per descobrir les possibilitats de substituir completament la fertilització per N, P i Mg mitjançant fertirrigació. Es va afegir un tractament de control amb una solució nutritiva completa (N, P i Mg afegits en el reg).
Les variables de creixement de la planta, desenvolupament, contingut de nutrients i producció de mongetes es van avaluar en el temps en tres dies diferents (35, 62 i 84). Per entendre l'origen de N a la planta, també es va determinar la fixació biològica de N_2 mitjançant el mètode d'abundància natural de 15N. Els tractaments amb estruvita van obtenir rendiments totals més baixos en comparació amb el control (p. ex., 59,35 ± 26,4 g de planta^{-1} per a 2 g, 74,2 ± 23,0 g de planta^{-1} per a 5 g i 147,71 ± 45,3 g de planta^{-1} per al control).
La nodulació del rizobi i les capacitats de fixació de N_2 van augmentar amb l'augment de les quantitats d'estruvita, mostrant un dèficit de Mg i P a les plantes amb el pas del temps. Tot i que aquestes deficiències podrien explicar els rendiments més baixos obtinguts, la combinació va mostrar resultats prometedors, ja que el contingut de N a l'estruvita semblava reforçar i no inhibir la nodulació del rizobi.

Un enfocament ambiental de la fertilització d'estruvita en la producció hidropònica.

L'enfocament ambiental mitjançant ACV es va realitzar principalment en dos experiments amb fertilització d'estruvita per a plantes d'enciam i pebrot (amb 5g, 10g i 20g de tractaments d'estruvita), així com mongeteres amb la combinació d'estruvita i inoculació de rizobi (amb 2g, 5g, 10g i 20 g de tractaments d'estruvita).

En el cas dels cultius d'enciam, els resultats mostren una reducció de gairebé totes les categories d'impacte (IC) quan s'utilitza l'estruvita com a fertilitzant P en comparació amb el tractament control. Es va observar una reducció significativa de les categories d'eutrofització d'aigua dolça i escassetat de recursos minerals. Per als pebrots, es va observar una reducció en totes les categories d'impacte amb 10g i 20g d'estruvita.

Quan s'assumeix una producció d'enciam durant tot l'any, podem veure que amb una quantitat inicial de 20 g la dissolució lenta de l'estruvita pot mantenir una producció competitiva durant 9 cicles de cultiu, que també significa menys impacte en totes les categories excepte en l'eutrofització marina.

En el cas del rendiment ambiental de la combinació d'estruvita i rizobi en la producció de mongetes verdes, per substituir la fertirrigació de P i N, els resultats indiquen una reducció del rendiment del 60% al 50% en comparació amb el control que es va regar amb una solució nutritiva completa. Aquesta reducció de rendiment augmenta considerablement l'impacte ambiental dels tractaments fertilitzats alternativament, especialment experimentant un augment de les emissions relacionades amb la infraestructura. Una estimació de la pèrdua de rendiment "permès" per continuar sent una opció de fertilització més sostenible seria un 10% per sota del rendiment control.

Es proposa una investigació posterior. Que segueix?

Per aprofundir més en els objectius principals proposats en aquesta tesi s'han detectat cinc vies principals d'investigació on es podrien fer més investigacions.

En primer lloc, l'augment de la circularitat urbana amb l'exploració d'altres materials d'origen urbà per ser utilitzats en l'agricultura hidropònica com a fertilitzant i substrat.

En segon lloc, l'augment de la disponibilitat de P donat a través de l'estruvita amb la combinació amb bacteris, des de bacteris solubilitzadors de fosfat fins a més investigacions sobre la combinació amb rizobi. En aquest sentit es podrien provar tractaments addicionals amb quantitats més altes d'estruvita així com l'ús de diferents compostos químics en la solució nutritiva per evitar desequilibris químics a la rizosfera.

En tercer lloc, l'ús sostingut de l'estruvita durant períodes de cultiu més llargs i conreus diferents (més i menys exigents). Així com l'ús de l'estruvita com a fertilitzant N per a cultius menys exigents.

En quart lloc, el coneixement sobre l'ús de l'estruvita i la seva integració al mercat i la “normalitat” en l'agricultura urbana es podria enriquir molt amb un estudi social sobre la percepció pública sobre l'ús d'aquest cristall en l'agricultura.

Finalment, com a cinquè tema de recerca proposat, es pot treballar més sobre les emissions potencials generades per la fertilització d'estruvita. Això implicaria un treball addicional sobre les emissions de N_2O i la influència potencial dels bacteris inoculats com el rizobi, així com la comparació de l'impacte ambiental de la fertilització amb estruvita i la fertilització de control en cicles de cultiu llargs com els tomàquets.

Resumen

El aumento de la población y las demandas sobre la agricultura llevan a la búsqueda de sistemas alimentarios alternativos que puedan ayudar a suministrar alimentos sin comprometer más el medio ambiente. La búsqueda de la producción de alimentos sin mayor uso de la tierra y los recursos ha convertido a la agricultura urbana (UA) en un buen candidato para ayudar a resolver esta problemática. La implementación de la UA también puede implicar otros beneficios que van desde la seguridad alimentaria y la resiliencia hasta el bienestar y el sentido de comunidad.

Además de la producción de alimentos, la UA se ha considerado un actor clave para aumentar la circularidad en las áreas urbanas. Desde una perspectiva de economía circular, la adición de UA puede servir como un sumidero de materiales de origen local para cubrir los requisitos de nutrientes o sustratos, mientras que al mismo tiempo reduce el transporte de alimentos y la gestión de residuos.

Este puede ser un punto crucial, especialmente con respecto a la huella ambiental de la síntesis y extracción de nutrientes, como en el caso del nitrógeno (N) y el fósforo (P) comúnmente utilizados en la agricultura hidropónica y aeropónica. La producción y extracción de estos nutrientes son actividades contaminantes y que consumen energía, lo que aumenta la huella de estos sistemas de producción agrícola. En el caso del P, también se considera un recurso no renovable

La expansión y escalamiento de la AU puede ser beneficiosa por múltiples razones y en más de un nivel, pero es importante garantizar la sostenibilidad de la actividad para evitar un cambio de impacto.

El objetivo principal de la presente tesis es la reducción de insumos no renovables/contaminantes para la implementación de una producción de UA más sostenible, en el marco de la economía circular.

Para cumplir con este objetivo principal, la presente divulgación pretende dar respuesta a las siguientes preguntas de investigación:

P1 → ¿Qué posibilidades hay en las zonas urbanas para reducir el impacto ambiental de la fertilización en la producción hidropónica teniendo en cuenta el marco de la economía circular?

P2 → ¿Son efectivos los métodos de fertilización alternativos en la producción hidropónica? ¿Es posible mantener la producción comercial?

Q3 → ¿En qué medida mejoramos nuestro sistema de producción? ¿Cuánto podemos disminuir nuestra huella ambiental?

Los materiales y metodologías utilizados en esta tesis se explicarán en la siguiente sección, seguido de los resultados obtenidos y la investigación adicional propuesta.

Materiales y métodos utilizados

Las metodologías utilizadas en esta investigación se pueden dividir en cuatro secciones principales: revisión de la literatura, agronómica, ambiental y estadística, que se utilizan en diferentes combinaciones a lo largo de la tesis.

Dentro de estos enfoques metodológicos se han utilizado diferentes materiales que describen principalmente la producción agrícola, el análisis del contenido de nutrientes en biomasa y agua, las emisiones de N2O a través de cromatografía de gases y la evaluación ambiental a través de herramientas LCA.

Aumento de la circularidad urbana: recuperación de nutrientes de los residuos urbanos

La presente tesis define las tecnologías de recuperación de nutrientes a partir de residuos urbanos encontradas en la literatura actual. Se realiza una primera definición de las principales fuentes de residuos dividiéndolas en dos apartados: "aguas residuales" y "residuos orgánicos, bio, alimentarios". De estas dos secciones se encontraron un total de 18 recuperaciones bajo los criterios de selección definidos, centrándose principalmente en la recuperación de N y P. El potencial actual del área metropolitana de Barcelona para recuperar N y P de los residuos se determina además especialmente de las aguas residuales, flujos y sitios de compostaje y gestión de residuos existentes. Si estos nutrientes recuperados se aplicaran a los sitios de UA existentes y prospectivos, los resultados mostraron que el P necesario podría suministrarse de 2,7 a 380,2 veces mientras que el N de 1,7 a 117,5 dependiendo de la estrategia de recuperación. El agotamiento de P y el alto coste energético para la síntesis de N hacen más necesarias las estrategias de recuperación, aunque la legislación actual aún no incentiva estas tecnologías.

Recuperación de fósforo en forma de estruvita y su aplicación en agricultura hidropónica.

En esta tesis doctoral se evaluó la viabilidad de utilizar estruvita precipitada de una planta de tratamiento de aguas residuales urbanas como única fuente de fertilizante de P en judías verdes (*Phaseolus vulgaris*), pimientos (*Capsicum annum L.*) y lechuga (*Lactuca sativa L.*).

En el caso de la judía verde, aplicamos varias cantidades de estruvita (que oscilan entre 1 y 20 g/planta) al sustrato y evaluamos la producción, los flujos de agua y los balances de P. Los resultados obtenidos indican que con más de 5g de estruvita se obtiene un mayor rendimiento (máximo de 181,41 g/planta) que el testigo (134,6 g/planta) fertilizado con fertilizante mineral

(KPO_4H_2). Las concentraciones de P detectadas en todos los órganos de la planta se mantuvieron más bajas cuando se usó estruvita. Finalmente, diferentes cantidades de estruvita permanecieron sin disolver en todos los tratamientos, lo que indicó un gran potencial para desarrollar ciclos de cultivo posteriores, así como una concentración reducida de P en los lixiviados.

Asimismo, para cultivos de pimiento y lechuga se ensayaron diferentes cantidades (5g, 10g y 20g por planta) y se compararon con un testigo fertilizado con fosfato monopotásico. El experimento tomó 3 meses, con 3 ciclos de lechuga y un ciclo largo de pimiento fertilizados con la estruvita inicial dada al inicio del experimento. Los rendimientos resultantes obtenidos fueron competitivos en comparación con el control, especialmente durante la primera cosecha de lechuga (225,5 g, 249,9 g, 272,6 g y 250 g para 5 g, 10 g, 20 g y control, respectivamente) donde se observó una mayor disolución de estruvita. Aunque el contenido de P en la biomasa de pimiento resultó bajo, las producciones son cercanas o incluso superiores a las del control, como en el caso del tratamiento con 20 g de estruvita (3,6 kg, 4,3 kg, 7,5 kg y 5,3 kg para 5 g, 10 g, 20 g y control, respectivamente).

Los resultados de estos estudios de fertilización mostraron el potencial y la viabilidad de la fertilización de P con estruvita, un nutriente recuperado localmente, en sistemas de agricultura hidropónica.

La combinación de inoculación de estruvita y rhizobium en la producción de *Phaseolus vulgaris*, reduciendo las necesidades de N y P en la fertilización hidropónica.

Se probó una combinación de fertilización con estruvita (tratamientos de 2 g y 5 g) e inoculaciones de rhizobium en judías verdes para descubrir las posibilidades de sustituir completamente la fertilización con N, P y Mg mediante fertirrigación. Se agregó un tratamiento de control con una solución nutritiva completa (N, P y Mg agregados en el riego).

Las variables de crecimiento, desarrollo, contenido de nutrientes y producción de frijol de las plantas se evaluaron en el tiempo en tres días diferentes (35, 62 y 84). Para comprender el origen del N en la planta, también se determinó la fijación biológica de N2 utilizando el método de abundancia natural de 15N. Los tratamientos con estruvita obtuvieron rendimientos totales más bajos en comparación con el testigo (p. ej., 59,35± 26,4 g planta-1 para 2g, 74,2±23,0 g planta-1 para 5g y 147,71± 45,3 g planta-1 para el testigo).

La nodulación de rhizobium y la capacidad de fijación de N2 aumentaron con el aumento de las cantidades de estruvita, mostrando déficit de Mg y P en las plantas a lo largo del tiempo. Aunque estas deficiencias podrían explicar los menores rendimientos obtenidos, la combinación mostró resultados prometedores ya que el contenido de N en la estruvita parecía reforzar y no inhibir la nodulación del rhizobium.

Un enfoque ambiental para la fertilización con estruvita en la producción hidropónica.

El enfoque ambiental usando LCA se realizó principalmente en dos experimentos usando fertilización con estruvita para plantas de lechuga y pimiento (con tratamientos de 5g, 10g y 20g de estruvita) así como judías con la combinación de estruvita y inoculación de rhizobium (con 2g, 5g, 10g y 20g de tratamientos de estruvita).

En el caso de los cultivos de lechuga, los resultados muestran una reducción de casi todas las categorías de impacto (IC) al usar estruvita como fertilizante P en comparación con el tratamiento de control. Se observó una reducción significativa para las categorías de eutrofización de agua dulce y escasez de recursos minerales. En el caso de los pimientos, se observó una reducción en todas las categorías de impacto con 10 g y 20 g de estruvita.

Cuando se supone una producción de lechuga durante todo el año, podemos ver que, con una cantidad inicial de 20 g, la disolución lenta de estruvita puede sostener una producción competitiva durante 9 ciclos de cultivo, lo que también significa un menor impacto en todas las categorías, excepto en la eutrofización marina.

En el caso del desempeño ambiental de la combinación de estruvita y rhizobium en la producción de judías verdes, en reemplazo de la fertirrigación con fósforo y nitrógeno, los resultados indican una reducción del rendimiento del 60 % al 50 % en comparación con el control que se regó con una solución nutritiva completa. Esta reducción del rendimiento aumenta en gran medida el impacto ambiental de los tratamientos de fertilización alternativa, especialmente experimentando un aumento en las emisiones relacionadas con la infraestructura. Una estimación de la pérdida de rendimiento "permitida" para seguir siendo una opción de fertilización más sostenible sería un 10 % inferior al rendimiento de control.

Propuesta de investigación adicional. ¿Que sigue?

Para profundizar en los principales objetivos propuestos en esta disertación, se han detectado cinco vías principales de investigación en las que se podría realizar más investigación.

En primer lugar, el aumento de la circularidad urbana con la exploración de otros materiales de origen urbano para ser utilizados en la agricultura hidropónica como fertilizante y sustrato.

En segundo lugar, el aumento de la disponibilidad de P dado a través de la estruvita con la combinación con bacterias, desde bacterias solubilizadoras de fosfato hasta futuras investigaciones sobre la combinación con rhizobium. En este sentido se podrían ensayar tratamientos adicionales con mayores cantidades de estruvita así como el uso de diferentes compuestos químicos en la solución nutritiva para evitar desequilibrios químicos en la rizosfera.

En tercer lugar, el uso sostenido de estruvita durante períodos de cultivo más largos y diferentes cultivos (más y menos exigentes). Así como el uso de estruvita como fertilizante nitrogenado para cultivos menos exigentes.

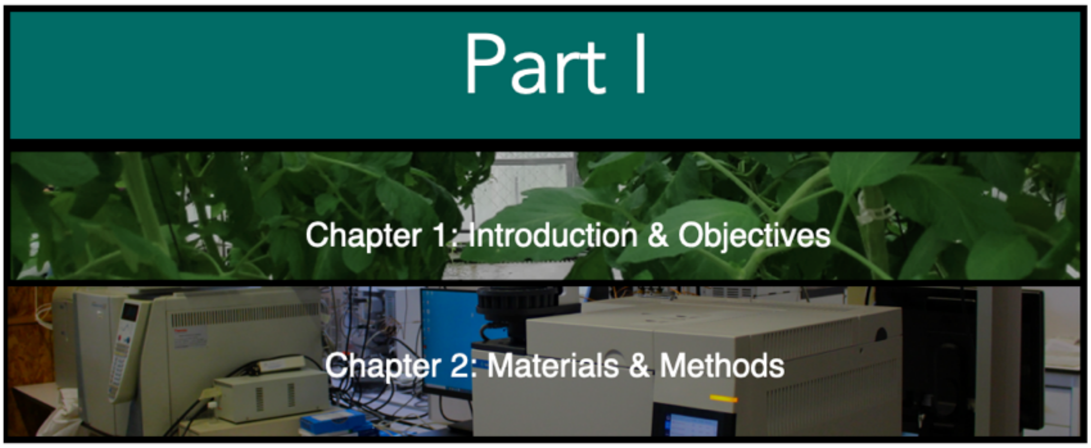
En cuarto lugar, el conocimiento sobre el uso de la estruvita y su integración en el mercado y la "normalidad" en la agricultura urbana podría verse muy enriquecido con un estudio social sobre la percepción pública sobre el uso de este cristal en la agricultura.

Por último, como quinto tema de investigación propuesto, se puede trabajar más sobre las emisiones potenciales generadas a través de la fertilización con estruvita. Esto implicaría más trabajo sobre las emisiones de N_2O y la influencia potencial de bacterias inoculadas como el rhizobium, así como la comparación del impacto ambiental de la fertilización con estruvita y la fertilización de control en ciclos de cultivo largos como los tomates.

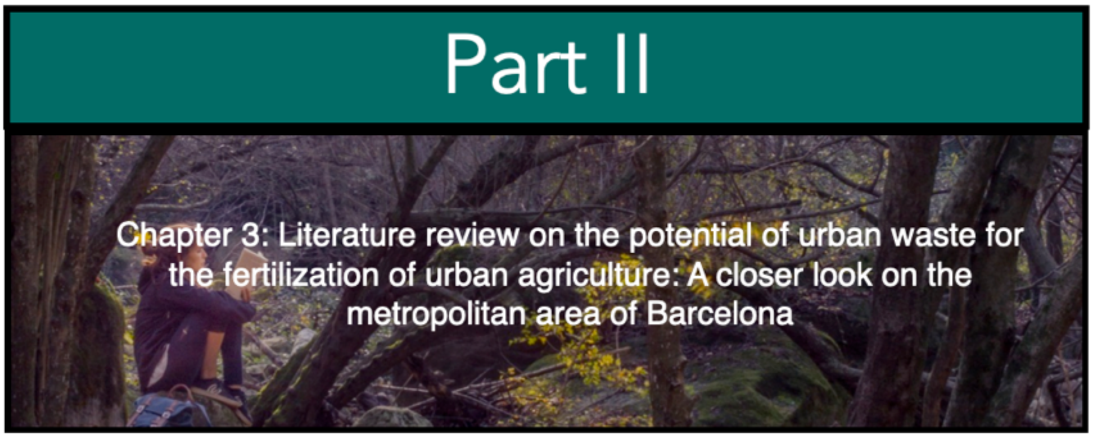
Preface

The present dissertation is divided into five main parts and an Annex.

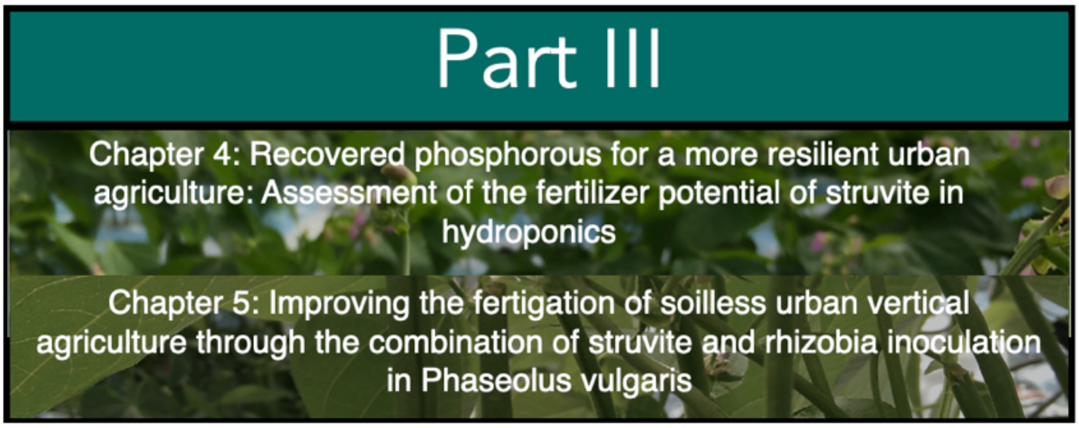
The first part is divided into the first two chapters providing an introduction to the topic, explanation of the research objective and the materials and methods used during the elaboration of this thesis.



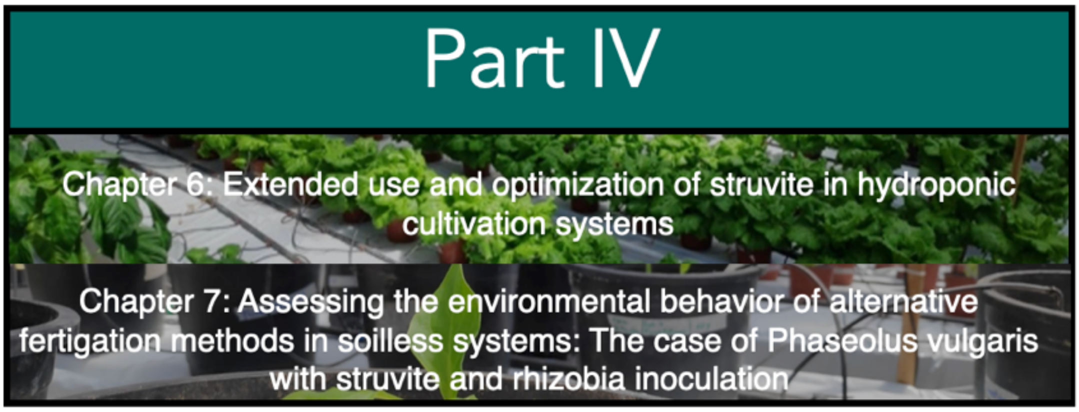
The second part contains the third chapter of the dissertation with an initial literature research on the potential of urban waste as a source of fertilizer for urban agriculture.



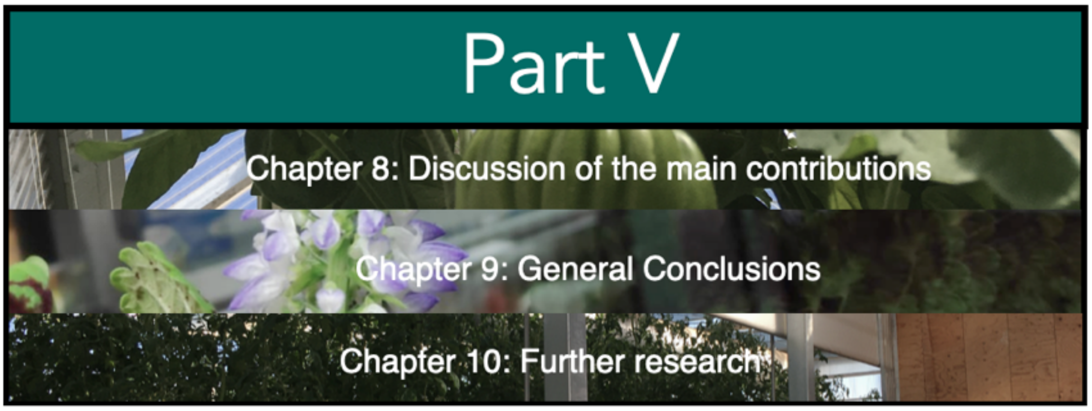
The third part focuses on the agronomic viability of the slow dissolving crystal fertilizer Struvite in hydroponic production systems. The containing chapters 4 and 5 dive into the use of struvite as an alternative P fertilizer and the potential combination with N_2 fixing bacteria rhizobium in green bean production.



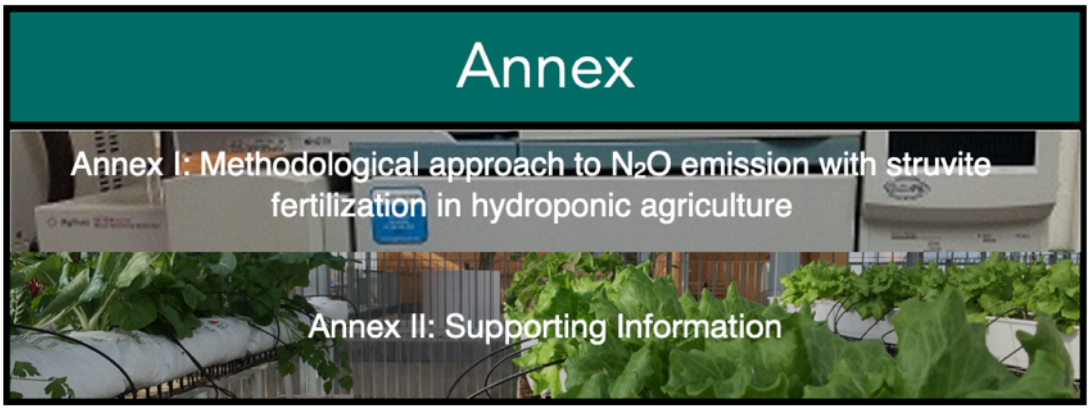
Part four of the present dissertation dives into the environmental impact of alternative fertilization methods with Struvite as well as its combination with rhizobium. The chapters 6 and 7 within this part present an LCA in their methodology added to the analytical analysis to uncover changes in the emissions generated with the use of alternative fertilizers. Additional to this part the Annex I section complements the environmental aspects of the present dissertation with an approach to N_2O emission determination from struvite in hydroponic agriculture.



The fifth and final part of this dissertation contain chapters 8, 9 and 10 with the general discussion and main contribution of the work as well as the general conclusions and proposed further research.



Additionally, the Annex provided in this dissertation contains two parts, the Annex I details further work done on the identification of N_2O emissions to air with the use of struvite fertilization, while the Annex II contains all supporting information from all previous chapters.



Dissemination and Training

Participation in scientific publications

- Recovered phosphorus for a more resilient urban agriculture : Assessment of the fertilizer potential of struvite in hydroponics', **Verónica Arcas-Pilz**, Martí Rufí-Salís , Felipe Parada, Anna Petit-Boix , Xavier Gabarrell, Gara Villalba; (2021) Science of the Total Environment, 799. doi: 10.1016/j.scitotenv.2021.149424.
- Improving the Fertigation of Soilless Urban Vertical Agriculture Through the Combination of Struvite and Rhizobia Inoculation in Phaseolus vulgaris **Verónica Arcas-Pilz**, Felipe Parada, Gara Villalba, Martí Rufí-Salis , Antoni Rosell-Melé, Xavier Gabarrell Durany; (2021) Frontiers in Plant Science. 12:649304. doi: 10.3389/fpls.2021.649304.
- Assessing the environmental behavior of alternative fertigation methods in soilless systems: The case of Phaseolus vulgaris with struvite and rhizobia inoculation, **Verónica Arcas-Pilz**, Martí Rufí-Salís , Felipe Parada , Xavier Gabarrell, Gara Villalba (2021) Science of The Total Environment, 770, p. 144744. doi: 10.1016/j.scitotenv.2020.144744.
- Extended use and optimization of struvite in hydroponic cultivation systems, **Verónica Arcas-Pilz**, Felipe Parada, Martí Rufí-Salis, Gaia Stringari, Ramiro González, Gara Villalba, Xavier Gabarrell; (2022) Resources, Conservation and Recycling, 179. doi: 10.1016/j.resconrec.2021.106130.

Participation as co-author in scientific publications

- Recirculating water and nutrients in urban agriculture: An opportunity towards environmental sustainability and water use efficiency? Rufí-Salís, Martí, Petit-Boix, Anna, Villalba, Gara, Sanjuan-Delmás, David, Parada, Felipe, Ercilla-Montserrat, Mireia, **Arcas-Pilz, Verónica**, Muñoz-Liesa, Joan, Rieradevall, Joan, Gabarrell, Xavier (2020) , Journal of Cleaner Production. doi: 10.1016/j.jclepro.2020.121213.
- Identifying eco-efficient year-round crop combinations for rooftop greenhouse agriculture, Martí Rufí-Salís, Anna Petit-Boix, Gara Villalba, Mireia Ercilla-Montserrat, David Sanjuan-Delmás, Felipe Parada, **Verónica Arcas**, Joan Muñoz-Liesa, Xavier Gabarrell (2020) International Journal of Life Cycle Assessment. The International Journal of Life Cycle Assessment. doi: 10.1007/s11367-019-01724-5.
- Closed-Loop Crop Cascade to Optimize Nutrient Flows and Grow Low-Impact Vegetables in Cities, Rufí-Salís, Martí, Parada, Felipe, Arcas-Pilz, Verónica, Petit-Boix, Anna, Villalba, Gara, Gabarrell, Xavier (2020) Frontiers in Plant Science. doi: 10.3389/fpls.2020.596550.

- Optimizing irrigation in urban agriculture for tomato crops in rooftop greenhouses, Felipe Parada, Xavier Gabarrell, Martí Rufí-Salís, Verónica Arcas-Pilz, Pere Muñoz, Gara Villalba, (2021) Science of the Total Environment, 794. doi: 10.1016/j.scitotenv.2021.148689.

Participation in Congresses/ Symposium

2nd ICTA-UAB Spring Symposium. Institut de Ciència i Tecnologia Ambientals, Universitat Autònoma de Barcelona. Bellaterra, Cerdanyola del Vallès, Barcelona, del 16 al 17 de mayo de 2019.

- Colaboración: Poster: Urban Agriculture: Estimation of the environmental impacts associated with different water sources, at the building and neighbourhood scale. Parada F., Rufí-Salís M., **Arcas V**., Villalba G., Gabarrell X.

GreenSys 2019 International Symposium on Advanced Technologies and Management for Innovative Greenhouses. Angers, Francia, del 16 al 20 de Junio de 2019.

- Poster: Different treatments for the availability of Phosphorus with Struvite for Soilless systems. **Arcas V**., Rufí-Salís M., Parada F., Petit-Boix A., Villalba G., Gabarrell X.
- Colaboración - Presentación: Substrate selection in urban agriculture, water holding capacity and resilience to water stress. Parada F., Ercilla-Montserrat, M., **Arcas V**., Rufí-Salís M., Villalba G., Gabarrell X., Muñoz P.
- Colaboración - Poster: Rooftop greenhouses for developing sustainable cities: identifying rooftop materials through hyperspectral remote sensing. Zambrano P., Parada F., Muñoz-Liesa J., **Arcas V**., Josa A., Rieradevall J., Gabarrell X.
- Colaboración - Poster: Improving urban agriculture sustainability with Life Cycle Assessment (LCA). Rufí-Salís M., **Arcas V**., Parada F., Petit-Boix A., Muñoz P.
- Colaboración – Poster: Greenhouses On Rooftop To Reduce Co2 In Urban Environment – The Groof Project. Sabre M., Schreier F., Volk D., Ancion N., Raulier P., Brulard N., Melon C., Deravet M., Wilhelm K, Morel-Chevillet G., Schott M.A., Zita N., Bini C., Dohogne C., **Arcas V**., Jijakli H.

VIII International Conference of Life Cycle Assessment in Latin America: CILCA 2019, Cartago, Costa Rica. Del 15 al 20 de Julio de 2019.

- Presentación: Comparative GHG emission analysis for the implementation of greenhouses on rooftops- the GROOF Project. **Arcas V**., Gabarrell X., Villalba G., Sabre M., Schreier F., Volk D., Ancion N., Raulier P., Brulard N., Melon C., Deravet M., Wilhelm K, Morel-Chevillet G., Schott M.A., Zita N., Bini C., Dohogne C., Jijakli H.

- Colaboración- Presentanción: Life Cycle Assessment: Reduction in water consumption through irrigation optimization strategies. Parada F., Rufí-Salís M., **Arcas V**., Muñoz-Liesa J., Petit-Boix A., Villalba G., Gabarrell X.
- Colaboración - Presentación: Intercropping, urban agriculture and circular economy. A first approach using Life Cycle Assessment to determine best annual crop combination. Rufí-Salís M, ., Ercilla-Montserrat, Sanjuan-Delmás D., Petit-Boix A., **Arcas V**., Parada F., Toboso S., Muñoz-Liesa J., Rieradevall J., Villalba G., Gabarrell X.
- Colaboración – Poster: Assessing phosphorus fertilizer potential of wastewater's struvite. Towards circular economy in urban agriculture. Rufí-Salís M., **Arcas V**., Parada F., Muñoz-Liesa J., Villalba G., Gabarrell X., Petit-Boix A.

International Society for Industrial Ecology Conference ISIE Americas 2020

- Water management strategies for a food production in circular cities. F. Parada, X. Gabarrell, M. Rufí-Salís, **V. Arcas**, P. Muñoz, G. Villalba.

12th International Conference on Life Cycle Assessment of Food 13-16 October 2020

- LCA of an alternative fertigation method with Struvite and Rhizobia inoculation in soilless hydroponic production for *Phaseolus vulgaris*. **V. Arcas-Pilz**, M. Rufí-Salis, X. Gabarrell, F.Parada, G. Villalba.

CityZen - Enhancing scalable innovations and new business models based on urban farming ecosystem values 19-20 April 2021

- Rooftop greenhouse connected to water, heat and CO2 flows of the building, Sostenipra research group **V. Arcas-Pilz,** X. Gabarrell

Participation in other Projects

During the duration of the PhD contributions and active participation was made in the following projects:

- **Fertilecity II** founded by the Spanish Ministry of Economy, Industry and Competitiveness (AEU/FEDER) [CTM2016-75772-C3-1-R]. The aim of the FertileCity II project is to deepen the research and promotion of urban agriculture through integrated greenhouses on rooftops, providing information and tools that make it possible. Fertilecity II wants to complete the development of LAU-ICTA experimental laboratories; minimize the use of resources (water and nutrients) for the production of low-carbon foods.

- **GROOF Project (2018-2022)** founded by INTERREG NWE. The GROOF (Greenhouses to reduce CO2 on Roofs) project is an innovative cross-sectoral approach to reduce CO2 emissions in the construction and agriculture sectors by combining energy sharing and local food production.
- **Pandemies** founded by Generalitat de Catalunya AGAUR 2020PANDE00021. This project aims to transform cities into resilient, sustainable and healthier spaces for the current and future pandemics, through local agriculture and recycling / waste reduction.
- **SIRAH** founded by the Spanish Ministry of Economy, Industry and Competitiveness. Promoting access to open urban agriculture from the Fertilecity lab to the city. The presented proof-of-concept project, Smart-Irrigation Resource-Agriculture- for Homes, entails de application of three innovations that stem from the Fertilecity II project. These three innovations are based on the results obtained in the former project and have been applied on different scales and can generate 4 products. The new ecodesigned products boost food production in cities in a sustainable way and will allow this activity for any citizen.

Co-Supervision Master Thesis

- Ramón Santiago (2019) – A circular economy approach for more sustainable hydroponic crop production. Thesis with experimental work with two short cycles of lettuce crop. Analisys of data for LCA using the Simapro Software. Part of the tribunal for the evaluation of this work.
- Sara Molins (2020) –Fluxes within a farmhouse with garden, wáter, waste and animal husbandry systems comparin permaculture connectivity and convential agricultura. Study based in Catalunya.
- Leonardo Pablo Parra (2020) – Comparison of policulture synergies and benefitial outcomes for different crops in urban agricultura systems. Study based in Catalunya.
- Gabriel Malaquin (2020) –Comparison of syinthetic light fixtures for indoor and rooftop crop production. Analisys of data for LCA using the Simapro Software.

Internship supervision

- Greta Casali (2019) – Master student stay from the University of Bolognia (280 horas).
- Pol Ribes (2020) – Estudiante Formación Profesional (400 horas).

Curses

- Basic level for Phytosanitary product handling and application
- obtaining the title and certificate for the handling of products(2018)
- Experimental design(2018)
- Basic statistic course with R studio (2018)
- Advanced statistic course with R studio (2019)
- Food-E Winter school (2021)
- Bioeconomy Curse (2021)

Entrepreneurial Activities

Co-founder and president of the start-up company Tectum garden, dedicated to the design, installation, maintenance, and consultancy on urban agriculture, created in 2020. Through the creation of a Spin-off company Tectum Garden the possibility of creating UA spaces in the city of Barcelona is not only increased but improved with the addition of interdisciplinary knowledge. During the duration of this dissertation dissemination activities on circularity in cities and a sustainable approach to UA have been made through the company activities. These activities range from the creation of a smart irrigation system through the SIRAH project, to the design of UA areas like the IMPD "Glories" space with the municipality of Barcelona or the maintenance and study of the social UA project "Horts al Terrat" lead by the IMPD institute and the municipality of Barcelona. Through these projects the focus has not only been the production of food goods but the integration of the circularity concept in the creation of smart products, the use of recycled material and the application of the gained knowledge throughout the thesis.

Research stay

From the 8th of November 2021 to the 28th of February 2022 a research stay was made at the university of Bologna under the supervision of Professor Francesco Orsini.

Part I

Introduction, Objectives & Methodological Framework

Chapter 1

Introduction & Objectives

Chapter 1: Introduction & Objectives

1.1 Introduction:

This introductory chapter describes the background information and the antecedents within the urban agriculture (UA) field which have led to development of this thesis. The first section is a comprehensive summary on the current problems in food supply security and how local food production (such as UA) can help reduce those risks, in addition to providing other benefits such as community cohesion and outdoor leisure. Next, the impacts associated to UA are discussed, especially how impacts can be shifted from the agricultural sector to the urban environment reflecting the importance of having a systemic approach. The third section offers a literature review of the impacts caused by mineral fertilizer production and use in modern agriculture, focusing on phosphorous and nitrogen. The next section states and justifies the motivation behind this dissertation in view of all presented above, namely, to increase urban circularity and reduce impact shifting in UA hydroponic production with the combination of locally sourced nutrients. Lastly, the research questions and objectives of this dissertation are presented.

1.1.1 Food security and local production:

Our present global food production capacity will not be able to meet the demand of the world population of 10 billion expected by 2050 (Searchinger et al. 2018). This expected population increase will require an expansion of agricultural land of 593million hectare as well as mitigation plan for the greenhouse gas (GHG) emissions expected from the additional agricultural activity (Searchinger et al. 2018; Ranganathan, Janet, Waite Richard, Searchinger 2018). The expansion of agricultural land use can reduce the soil organic carbon and organic matter in the soil, increase its salinity and erosion which can further entail the loss of nitrogen and phosphorous nutrients by 23-42Mt and 14.6-26.4Mt globally each year (IPCC 2019). On the other hand, around 21-37% of GHG emissions stem from the food system which is expected to increase by 30-40% by 2050 due to the population growth (IPCC 2019). This gap between population growth and the food system can pose a serios problematic for general food security which needs to be solved with the increase of food production without the direct increase of agricultural land and with the direct reduction of GHG emissions associated to agriculture (Searchinger et al. 2018). The pressure on agricultural performance is enhanced taking in account the increasing population density in urban areas which surpassed rural population in 2007 for the first time and is expected to increase up to 1.69% until 2030 (United Nations 2019). The loss of population density in rural areas means a reduction in the agricultural sector workforce, especially for small scale farmers, further weakening the stability of the food chain. While cities are highly demanding areas, their capacity for self-supply is almost nonexistent, being dependent on food import and production in the surrounding peri urban areas. During the COVID-19 pandemic and global lockdown the low resilience of cities was palpable, being problematic on many levels of the food value chain (Vittuari et al. 2021). Transport limitations and the closing of borders limited the access of farmers to the land for harvest which was also hampered by social distancing regulations. Overall, the production and produce transport was reduced compared to the pre COVID19 times, leaving farmers and producers with unsold fresh produce in the field or distribution facilities (Vittuari et al. 2021; Lal 2020). The social initiatives and overall actions taken during the COVID19 pandemic have led to the identification of strategies to increase the resilience in urban areas, as expressed by Vittuari *et al.*, (2021) the inclusion of local food production can reduce the vulnerability of the food chain. This idea has been expressed prior to the global pandemic in 2020, being UA a more commonly regarded concept in the past decade to increase city resilience while reducing pressure on surrounding farmlands (Grard et al. 2018).

1.1.2 Urban Agriculture typologies

UA has been encouraged in recent years, becoming a more popular activity in major cities (Appolloni et al. 2021), being one of the potential activities defined to increase their sustainability. This activity has specially grown in the last decade with an increase of rooftop farming installation accounting for around 185 cases around the world (Appolloni et al. 2021). Other activities like water harvesting and the production of clean energy on underutilized areas joint with the production of food are the baseline for the “Roof Mosaic” framework which aims to provide, food, water and energy on local scale, making buildings, neighborhoods and cities more resilient (Toboso-Chavero et al. 2019a; 2019b). The idea of UA not only regards the production of food but can excel its initial purpose, being a source for fresh food, reducing transporting distances and the need for packaging, having educational purposes or even social benefits (Specht et al. 2014).

Different types of UA can be identified, from soil based on the ground to rooftop agriculture, or even indoor farming. These typologies can also vary in their production system, being hydroponic and aeroponic systems most common for rooftop and indoor farming due to the high productivity, lower weight load and control over the plant growth and fertilization.

1.1.2.1 Placement

As expressed before UA can be performed on different locations within the urban and peri urban areas. Although agriculture on the ground is still popular, especially in peri urban areas other building-bound agriculture also known as vertical farming have been explored. In figure 1.1 we can see two main groups, soil based and vertical farming settings. While soil-based can be open air or protected with the addition of a greenhouse structure, vertical farming has a different variety of integration into the building and therefore different levels of open or protected techniques (Thomaier et al. 2015). The concept of vertical farming has been developed as an opportunity for food production without the need of additional space, profiting also from building metabolic flows (Sanyé Mengual 2015).

While little infrastructure is needed in open systems, the complexity increases with the addition of a greenhouse or even with the integration into the building structure. The same can be said about the control on climatic variables, being scarce in open air systems and gaining importance with the increasing infrastructure (Specht et al. 2014). The connection of agriculture and building can provide several benefits, one being the potential exchange of waste heat as defined by Muñoz-Liesa et al., (2021), reducing up to a 8% on building energy requirements in the ICTA institute even adding up to 45.6 kg CO_2 eq/ m^2 of carbon savings (Muñoz-Liesa et al. 2020). Another synergy drawn from this association is the CO_2 injection from building to greenhouse and recirculation of clean air back to the building as proposed by the GROOF project (Sabre et al. 2019). Lastly, positioning vegetation on building surfaces can also help reducing the urban heat island effect (K. Ackerman et al. 2014; IPCC 2019).

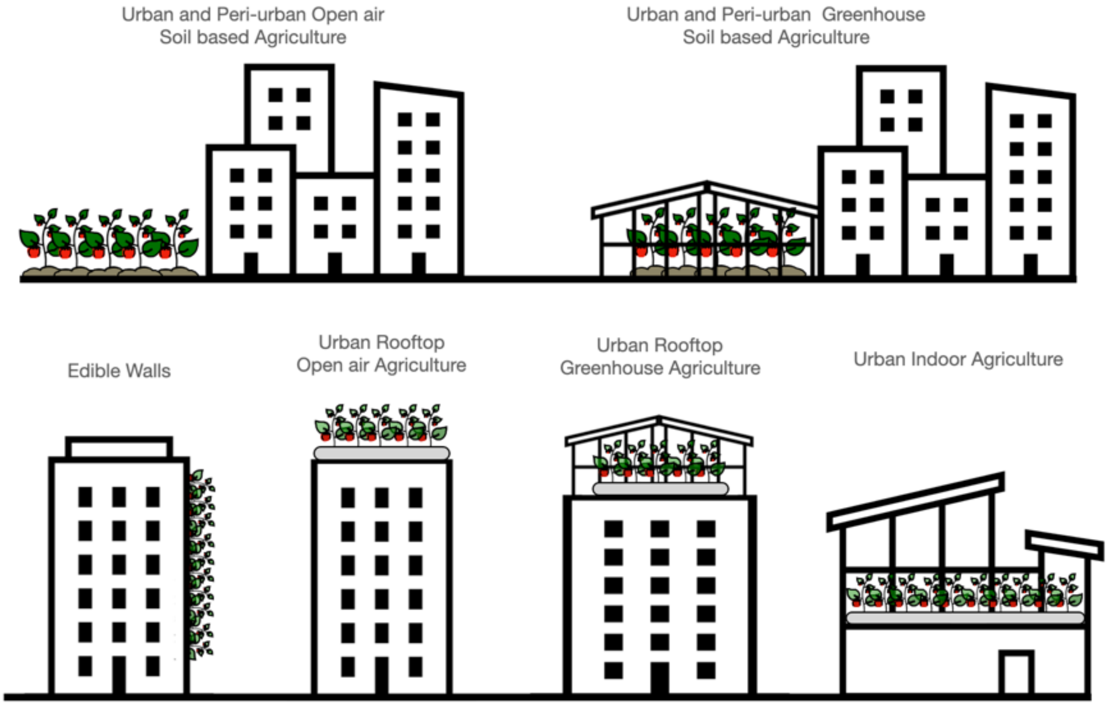


Figure 1. 1 Various forms of implementation of urban agriculture

1.1.2.2 Irrigation system

The hydroponic production systems can be performed using substrate as a way to sustain the plant in place like in figure 1.2 (A) with a drip irrigation of the nutrient rich water or without substrate using the nutrient film technique (NFT) like depicted in figure 1.2 (B). The NFT consists of the contact of the plant roots to a nutrient rich water surface holding the plants mechanically above. Aeroponic systems resemble NFT but instead of maintaining a contact between root and nutrient solution, it is diffused in the form of mist to moist the root system as shown in figure 1.2 (C). Finally figure 1.2 (D) shows the deep-water culture technique which consists of the total submersion of the plant roots into the oxygenated nutrient solution to avoid radicular death.

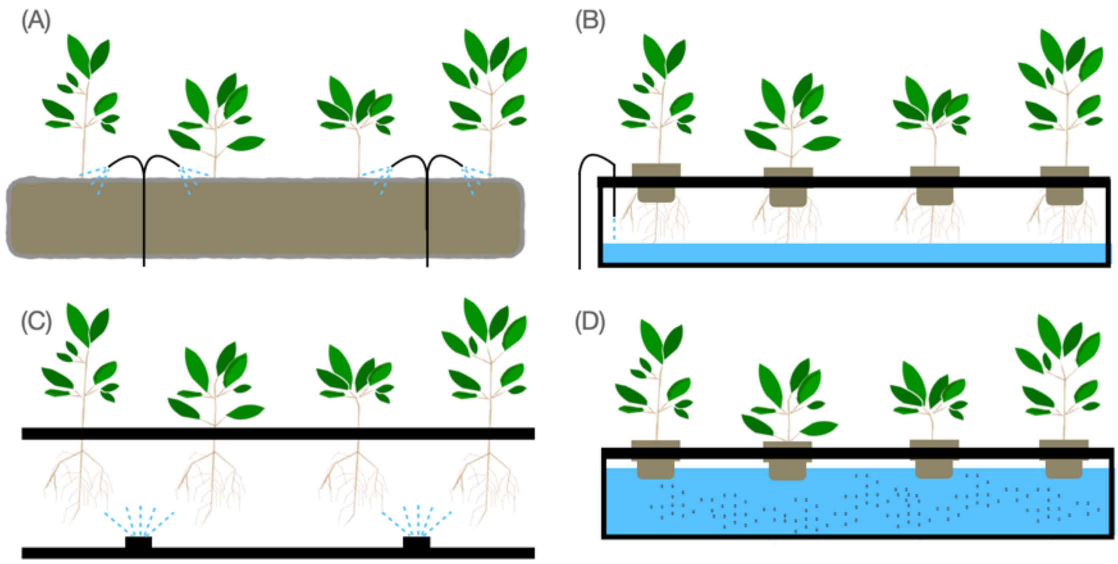


Figure 1. 2 (A) Drip Irrigation with substrate (B) Nutrient film technique (NFT) without substrate (C) Aeroponic fertilization system without substrate (D) Deep water culture technique (DWCT) without substrate.

This change has also generated a departure from traditional fertilization methods, which traditionally were given in the form of animal manure directly to the soil (Sun et al. 2018a). In substrate and non-substrate based hydroponic systems the fertilization is given through the irrigation directly with the composition of a nutrient solution with the essential macro and micronutrients necessary for plant growth and development (Sonneveld and Voogt 2009).
Production systems defined to increase the circularity of the food production are aquaponics and bioponics which reuse the wastewater from fisheries or other wastes to be used as nutrients for vegetable production (Graber and Junge 2009; Dsouza et al. 2021).

1.1.2.3 Substrate:

UA can be based on the soil on the ground but due to reduced space availability in cities and the potential heavy metal contamination of urban soils other ways of production have been developed (Ercilla-Montserrat et al. 2018), from raised beds with compost substrate to mineral or organic substrates like peat, rockwool and perlite. The choice for a good substrate to be used in drip system hydroponic agriculture depend on a set of characteristics defined by Barrett *et al.*, 2016, which can be resumed as the following in figure 1.3:

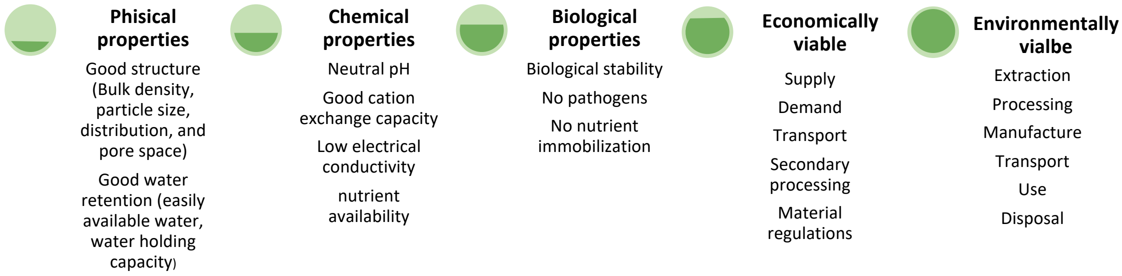


Figure 1. 3 Substrate properties as described by Barrett et al., 2016

These characteristics have helped define a great variety of substrate materials from different origins, from organic materials like fruit husks, plant fibers, composts, or peat to mineral materials like expanded clay, vermiculite, zeolite, and perlite to even synthetic materials like foams and other types of expanded plastics. Great number of studies have been focusing on the development of new substrates (Grard et al. 2018) and combinations (Parada, Ercilla-Montserrat, et al. 2021) as well as economic, environmental and social analysis of substrate materials (Vinci and Rapa 2019; Toboso-Chavero, Madrid-López, et al. 2021) to increase circularity of materials in cities making UA more sustainable and resilient.

1.1.3 Services identified from UA

In the last decades the research on UA has expanded its focus from food production to a greater variety of services this activity can offer. UA has been identified as a potential mean to increase biodiversity, boost local sense of community and purpose, reduce transport and packaging of goods as well as a sink for local resources, apart from being a source of food security in urban environments (Figure 1.4).

Figure 1. 4 Main services identified in UA

The reduction of transport and packaging are consequences of the local food production which is an approach that has already been embraced by retailers, not only to reduce outsourcing fresh produce and the transport to the supermarket but also as a marketing strategy. Examples for this approach are the Gotham greens farm (New York, USA), the LUFA farm (Montreal, Canada) and the Urban Farmer Greenhouse in top of the REWE supermarket (Wiesbaden, Germany) with a commercial rooftop greenhouse and a direct or indirect selling point in the same building (Sanyé Mengual 2015).

The increase of food security is a greatly discussed topic in literature in the context of UA and while it is not seen as a "silver bullet" to provide food for entire cities, it is regarded as a potential to increase provisions of food in cities as well as a way to approach fresh food in neighborhoods with limited access (Specht et al. 2014; Langemeyer et al. 2021).

Within these key points other specific benefits have been identified which can arise from UA. Under the umbrella of "sense of purpose and community" several roles have been recognized that can be enhanced through the practice of UA (Figure 1.5).

Figure 1. 5 Services related to "Sense of purpose and community"

Many of these points stem from a more social aspect of UA which is usually not as business-oriented as commercial gardens (Appolloni et al. 2021).

These gardens tend to focus on educational and skill building aspects, community cohesion and activism (Horst, Mcclintock, and Hoey 2017). Urban gardens can also provide a sense of purpose and identity as well as empowerment through self-sufficiency which can be regarded as social justice (K. Ackerman et al. 2014). On the other hand, the creation of commercial urban gardens can increase the creation of jobs in the area, with the development of a new production sector (Specht et al. 2014; Orsini et al. 2013).

On a different note, the potential to increase biodiversity has been identified in the creation of green spaces and green roofs in cities, generating adequate conditions for flora and fauna such as mammals, birds and insects (Walters and Stoelzle Midden 2018). Pollinators such as bees can also benefit from these green areas (Ayers and Rehan 2021) as well as other biological regulators.

Finally, UA has been deemed as very useful to further utilize local resources (figure 1.6). These local resources can be natural such as the sunlight and the rainwater or resources sourced from human activity. Urban gardens can profit from the incoming sunlight in under regarded rooftop spaces, increasing the building climatic conditions (Muñoz-Liesa et al. 2020), whilst the water uptake of the vegetation can decrease excess rainwater runoff and flooding (Mentens, Raes, and Hermy 2006; Speak et al. 2013). On the other hand, UA can also be a sink for human made waste such as, biomass (Eldridge, Yin, and Nerida 2018), wastewater (Rufí-Salís et al. 2020), and food waste (Khoshnevisan et al. 2020).

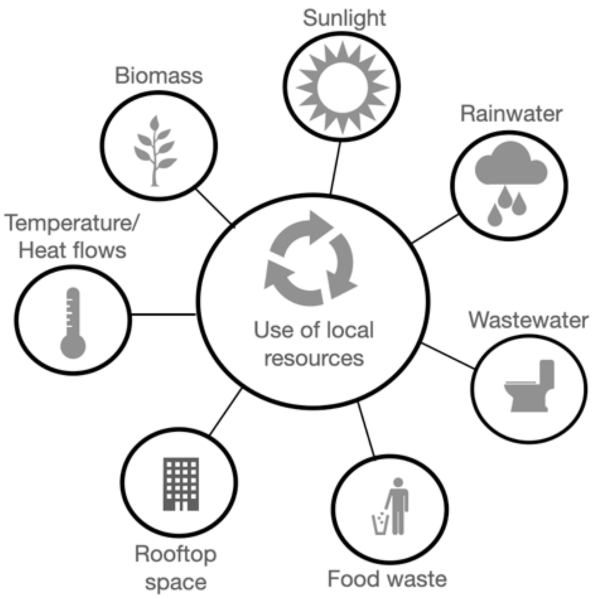


Figure 1. 6 Services related to "Use of local resources"

1.1.4 Environmental burden of UA

Although many positive impacts have been defined throughout literature, others have questioned the consequences of up-scaling UA to a city level. Great work has been done defining the environmental burden of urban farms and novel farming systems but the use of resources for the creation and maintenance of these structures may not be compensated by the reduction of food packaging and transport (Goldstein et al. 2016). This phenomenon known as "environmental burden shifting" was defined by Yu *et al.*, (2016) as:

> *"Any improvement activity taken in a phase may make the environmental impact reduce in a phase but increase in other phases. This so-called environmental burden shifting phenomenon forces analysts to collect all the related data in every time when assess the activities' environmental effects."*

Recent works have started to address this phenomenon through the life cycle analysis tool (Dorr et al. 2021), focusing on the generated environmental impacts of UA as well as its infrastructure construction (Muñoz-Liesa, Toboso-Chavero, et al. 2021). As considered by Goldstein *et al.*, (2016) the environmental burdens associated to agriculture are the use of fertilizer, pesticides, land, irrigation and fossil fuel energy and while UA can be a more sustainable method for land use, the up-scaling of this production method might imply the increase of other environmental burdens associated to agriculture in urban areas.

Hydroponic and aeroponic techniques rely on the use of mineral and synthetic fertilizers which can account for up to 25% of their environmental footprint (Sanjuan-Delmás et al. 2018). Goldstein *et al.*, (2016) and Sanjuan-Delmás *et al.*, (2018) urge for the reduction of fertilizer consumption and water leachate for a large-scale implementation of UA.

1.1.4.1 Nitrogen and Phosphorous fertilizers in UA

Nitrogen (N) and phosphorous (P) are two main nutrients indispensable for plant growth, while N is a necessary compound for amino and nucleic acid structures and therefore crucial in protein as well as DNA, RNA formation, P is part of the ATP molecule responsible for energy production (Sena and Hicks 2018). N and P and potassium (K) are considered main drivers for plant growth and while their application can vastly increase crop growth, and with it food security, the extensive use in modern agriculture has its consequences (Khan and Hanjra 2009)(figure 1.7).

- N is usually applied as NO_3^{2-} or NH_4^+ often in the form of compound fertilizer (Sonneveld and Voogt 2009). Although organic forms of N such as urea can be used the synthetization of N fertilizers from atmospheric N_2 through the Haber-Bosch process is a common practice since the early 20th century to respond to the growing food demands (Galloway et al. 2004). This production process is highly energy demanding, making N production account for more than 10 times more energy per ton than the production of P and K fertilizers (Khan and Hanjra 2009). The application of N-based fertilizers are main drivers for the emission of the greenhouse gas N_2O which contributes to stratospheric ozone depletion and has a global warming potential almost 300 times greater than CO_2 (Llorach-Massana et al. 2017; Stavi and Lal 2013; IPCC 2019). N runoff into soil and water bodies can also contribute to their acidification and water eutrophication (aquatic dead zones due to excess algal bloom)(Hamilton et al. 2018).

- P on the other hand is regarded a non-renewable resource (Cordell, Drangert, and White 2009) which is obtained through the mining of phosphate rock and given in the form of H_3PO_4 and soluble salts like $NH_4H_2PO_4$ and KH_2PO_4 (Sonneveld and Voogt 2009). It is considered that the quality of rock phosphate reserves is declining and could be depleted in the next 50-100 years causing a major crisis in the agricultural industry due to the increasing global demand (Cordell, Drangert, and White 2009). Data from 2004 showed a yearly loss of 18.5 millions of metric tons of P into the soil due to its use as fertilizer while additional 1.3 millions of metric tons were lost into the hydro- and atmosphere (Villalba et al. 2008) further adding to the excess of nutrients in water bodies and the consequential eutrophication. The distribution of rock phosphate resources can cause geopolitical tensions and increase food insecurity for small scale farmers and countries with fewer resources (Cordell, Drangert, and White 2009; Chrispim, Scholz, and Nolasco 2019).

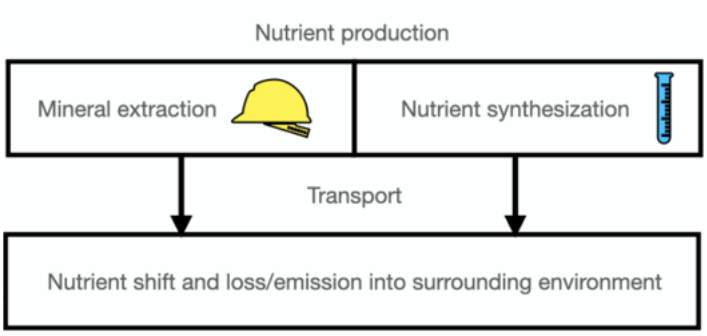


Figure 1. 7 Initial problematic with P and N production

In recent years, the search for alternatives to the synthetization, extraction and disposal of P and N have been rising in interest, suggesting, not only a reduction and better management in their use but also the recycling and recovery of nutrients from other sources (Rufí-Salís et al. 2020; Venkata Mohan, Amulya, and Annie Modestra 2020; Möller 2016; M. Ahmed et al. 2019; Weidner and Yang 2020).

1.2 Motivations of the dissertation:

While the production of food and the increase of food security is vital, the expansion of agricultural land and the use of resources are finite. And while UA can be a good alternative to further increase food production without altering the existing land use the potential consequences need to be addressed.

Since nutrient and fertilizer extraction have a major role in modern agriculture the environmental burden associated with agriculture could be also shifted into UA. On the other hand, the use of alternative nutrient sources could be a solution to make UA a sustainable practice. The need to search, test and analyze the environmental burden of alternative nutrient sources in urban areas is therefore the overall idea behind this dissertation (figure 1.8).

On one hand the nutrients sourced from urban areas can replace the burdens caused by nutrient synthetization and extraction, reducing energy needs for the Haber-Bosch process as well as reduce P extraction and insecurity over P shortage. The idea behind the local sourcing of nutrients to be used as fertilizers locally also involves the reduction of transportation and which indirectly could also infer in geopolitical disparities over nutrient availability.

On the other hand, the recovery and recycling of nutrients from urban flows can reduce their access to the surrounding environments, meaning the soil, hydrosphere, and atmosphere.

Therefore, it could be considered that UA can serve as a driver for circularity in cities rather than a threat.

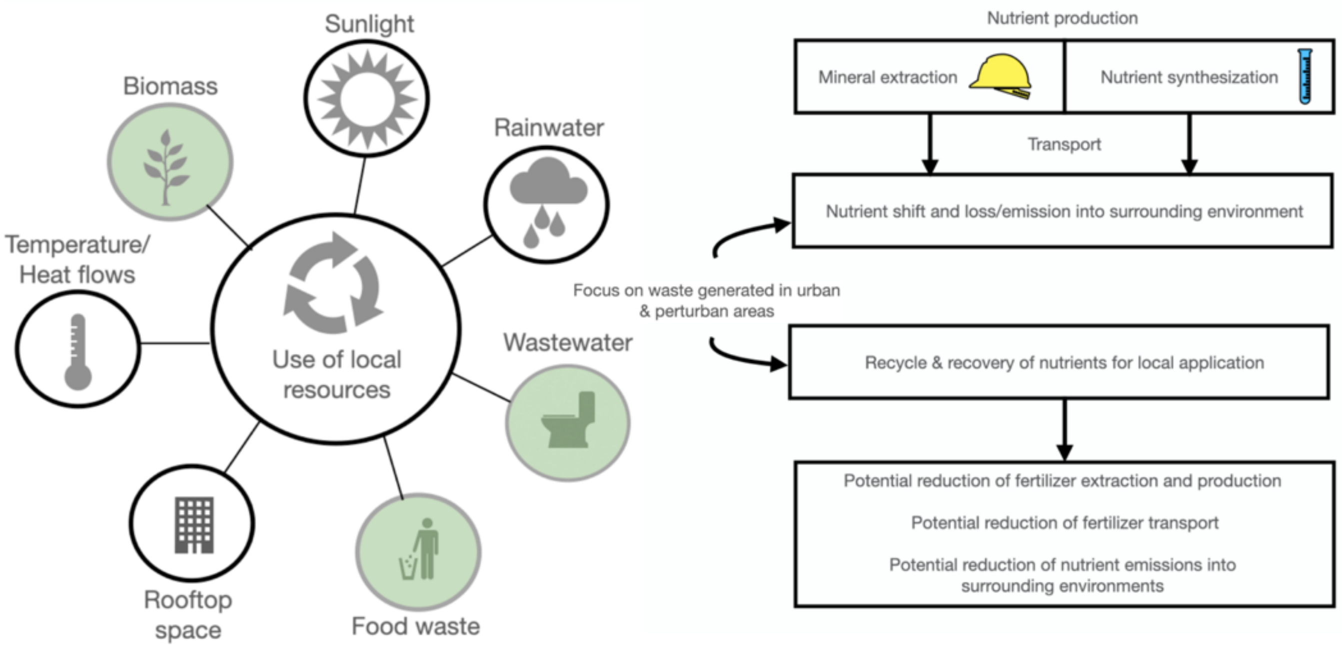


Figure 1. 8 Potential for circularity in nutrient source for UA

The source of nutrients in urban areas needs to be assessed as well as the recovery techniques available. Previous authors have detailed methodologies for nutrient recovery from urban waste, including food and biomass waste, while other authors have focused on the recovery from waste waters. A total overview of current possibilities was needed also addressing the potential to be used in UA and the potential to satisfy the production in existing or potential areas.

Not only the availability of nutrients is a key factor but also the suitability for the type of agriculture that is spreading more rapidly in vertical or rooftop agriculture. While some alternative nutrient sources have already been tested on soil-based agriculture the hydroponic and soilless production has not been contemplated. Furthermore, the reduction of both N and P use in hydroponic agriculture must also be analyzed, addressing the potential for the combination of different alternative fertilization methods. Therefore, initial steps into plant development and agricultural production need to be assessed.

To understand the environmental impact of the assessed alternative fertilization methods it is important to analyze the emissions related to the production system life cycle compared to the use of conventional fertilizers for hydroponic agriculture. This entails the analysis of nutrient production, transport, uptake, emissions to air and water as well as energy consumption and materials for the production per se.

The information gathered through the application and analysis of alternative fertilization methods in hydroponic agriculture hopes to influence the actual urban metabolism flows and point out the potential of circularity with the addition of UA.

1.3 Objectives:

1.3.1 Main Objective:

- Reduction of nonrenewable/ polluting inputs for the implementation of more sustainable urban agriculture production, within the circular economy framework.

1.3.2 Research Questions:

Q1 → What possibilities are available in urban areas to reduce environmental impact of fertilization in hydroponic production taking in account the circular economy framework?

Q2 → Are alternative fertilization methods effective in hydroponic production? Is it possible to maintain the commercial production?

Q3 → To what extent do we improve our production system? How much can we decrease our environmental footprint?

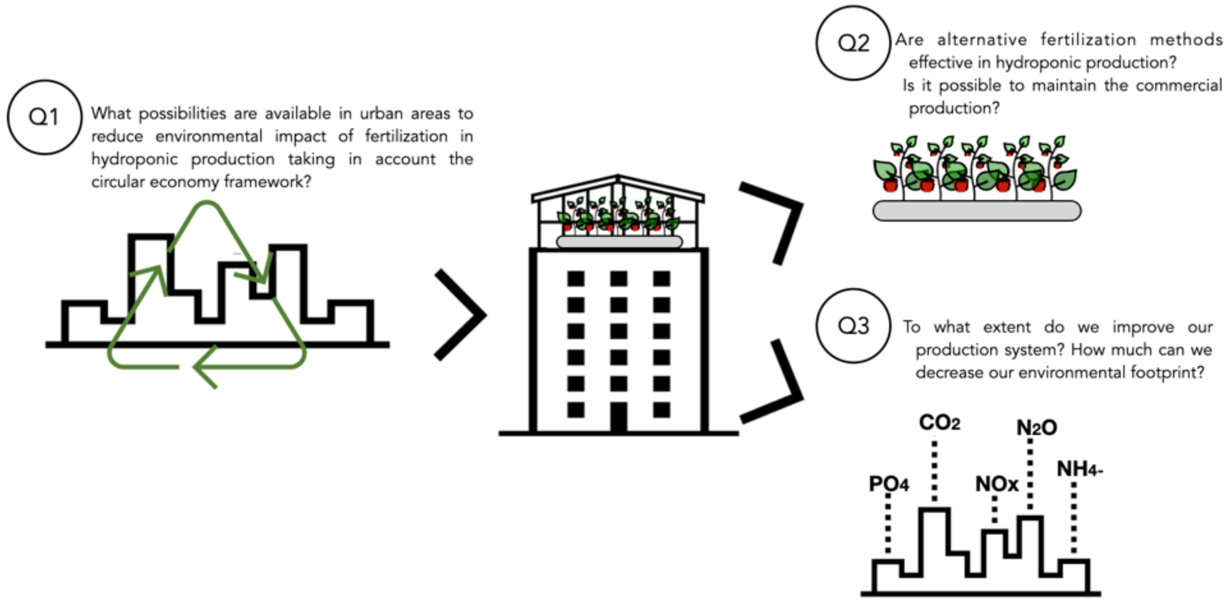


Figure 1. 9 Illustration of the research framework and research questions

1.3.3 Defined objectives:

As defined in the main objective (section 1.3.1) the end goal is the reduction of inputs for UA using the circular economy framework. Therefore, the first objective needs to address the current possibilities in urban settings to replace said inputs (figures 1.9 and 1.10):

Q1 Objective 1: Find materials and processes in urban and peri-urban areas beneficial for the reduction of the environmental impact of agricultural production within the city.

- Chapter 3
  - Literature review on the potential of urban waste for the fertilization of urban agriculture: A closer look on the metropolitan area of Barcelona

While the initial finding of potential repurposed nutrients can give an idea of the possibilities the city can bring, it is important to determine the actual use of the sourced materials and the viability in UA:

Q2 Objective 2: Determination of viability of chosen alternative fertilizers in hydroponic production in greenhouse.

- Chapter 4
  - Recovered phosphorous for a more resilient urban agriculture: assessment of the fertilizer potential of struvite in hydroponics.
- Chapter 5
  - Improving the fertigation of soilless urban agriculture through the combination of struvite and rhizobia inoculation in *Phaseolus vulgaris*.

As defined in the main objective the global purpose is the reduction of pollution in urban areas while encouraging UA. The alternative inputting materials defined must therefore be environmentally assessed:

Q3 Objective 3: Identification of the reduction of the impact of said alternative fertilizers in comparison to conventional fertilization.

- Chapter 6
  - Extended use and optimization of struvite in hydroponic cultivation Systems.
- Chapter 7
  - Assessing the environmental behavior of alternative fertigation methods in soilless systems: The case of *Phaseolus vulgaris* with struvite and Rhizobia inoculation.
- Annex I
  - Methodological approach to N_2O emission with struvite fertilization in hydroponic agriculture

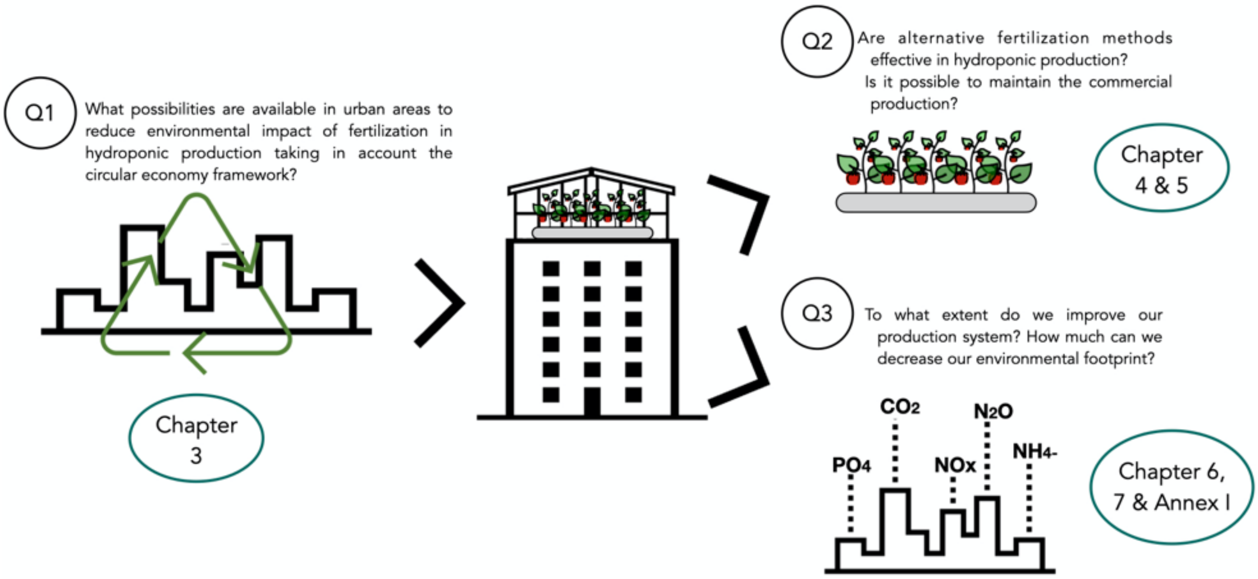


Figure 1. 10 Illustration of the research framework and research questions with the corresponding chapters of this dissertation

Materials & Methods

Chapter 2: Materials & Methods

This chapter details the methodological framework of this dissertation by defining the methods and tools employed.

2.1 Methods:

The methodological framework can be classified in four main sections: literature review, agricultural analysis, environmental analysis, and statistical analysis (Table 2.1). While the methodology of the **chapter 3** is mainly literature research all **chapters from 4 to 7** contain agronomic and chemical analysis. **Chapters 6, 7 and Annex I** also entail an environmental analysis. Finally, **chapters 4, 5 and 6** contain statistical analysis of the obtained data. These sections contain different analytical methodologies as well as materials which will be explained in the following segments.

Table 2. 1 Methodology used for each chapter

		Literature review	Agronomic analysis	Environmental analysis	Statistical analysis	Study site
Part I	Chapter 3	•				AMB
Part II	Chapter 4		•		•	ICTA- UAB Greenhouse
	Chapter 5		•		•	
Part III	Chapter 6		•	•	•	
	Chapter 7		•	•		
	Annex I		•	•		

The potential of nutrient recovery from urban areas presented in the first research question is addressed in Part I of the dissertation with **chapter 3**, a literature review on analyzed nutrient sources and recycling methodologies with a focus on the metropolitan area of Barcelona.

The Part II of this dissertation contains **chapters 4 and 5**, dwelling with the suitability of fertilization with the use of struvite (**chapter 4**) and its combination with nitrogen fixing bacteria rhizobium (**chapter 5**) in hydroponic systems with a focus on the plant development, yield and in the case of **chapter 5** the nitrogen fixation by the bacteria. Finally, Part III incorporates environmental analysis tools, understanding the potential of long and short production cycles with struvite (**chapter 6**), the environmental footprint of struvite and rhizobium combinations (**chapter 7**) and a first approach of the N_2O emission factor with struvite as source of nitrogen (**Annex I**).

2.1.1 Literature review:

A literature review serves as an information source giving a glimpse into the state-of-the-art of a specific topic, serving as a tool to understand or classify existing information as well as basis for decision-making and policies. The structure and methodology applied in a literature review can be defined depending on the goal of the resulting information (Lau and Kuziemsky 2016).
Different classifications of literature reviews define up to 9 types of methodologies (figure 2.1) (Paré et al. 2015):

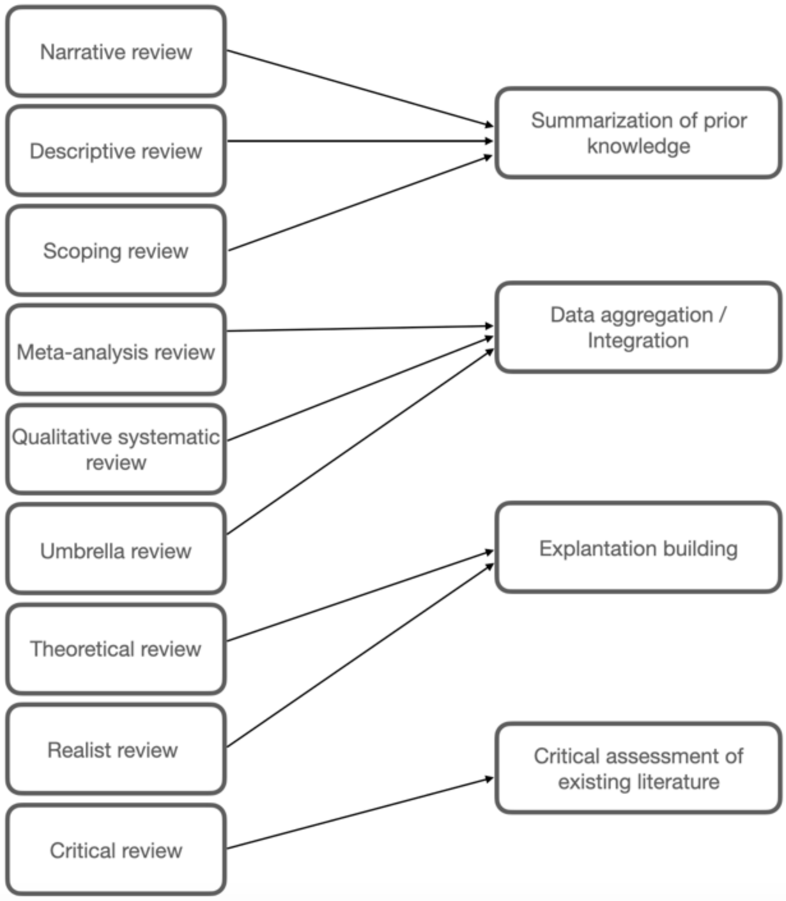


Figure 2. 1 Literature review methodologies

The first group, "summarization of prior knowledge" focuses on the synthetization of existing literature to define a state-of-the-art in a representative way like in the case of descriptive and scoping reviews (Peters et al. 2015) or without a clear reviewing process like a narrative review (Paré et al. 2015). The goal of "data aggregation or integration" uses empirical quantitative data collection from existing literature to form new patterns or findings. Theoretical and realist reviews on the other hand point out relationships and common concepts of existing conceptual and empirical literature to form new contributions and/or highlight existing knowledge gaps (Paré et al. 2015). Finally, a critical aim to highlight critical points, controversies, strengths and weaknesses of existing literature (Lau and Kuziemsky 2016).

For the literature obtention and classification of **chapter 3** several searching tools were available:

Scopus - The Elsevier abstract and citation database offers an extensive coverage of content accessible with a comprehensive searching tool. This search tool utilizes selective criteria on its coverage applied by editors. The Scopus search tool can be simplified or more advanced using different criteria to narrow down data of interest (Elsevier 2022) https://www.scopus.com

Web of Science - The web of science (WOS) website, form Clarivate is not linked to a scientific publisher, with a comprehensive database that can be accessed through a search tool. As in the case of Scopus, WOS also utilizes selective criteria on its coverage applied by experts to index their database. This search tool can be specific for documents or researchers allowing also an advanced search for further filtering and specification of the search goal (Clarivate 2022) https://www.webofscience.com

Google Scholar – The more specialized searching tool owned by Google is freely accessible in contrast to Scopus and WOS which are subscription-based databases. This search tool enables the access to abstracts and citations in all disciplines with the possibility to do an advanced search for specific research goals. Contrary to Scopus and WOS the coverage of the Google Scholar database is more inclusive with no apparent selection criteria (Google 2022).

https://scholar.google.es

2.1.2 Agronomic analysis:

2.1.2.1 Climatic variables:

During the experimental processes of **Chapters 4, 5, 6 & 7** the climatic variables temperature, radiation and relative humidity were measured on-line. These measurements were made with the following on-line sensors attached in different locations of the experimental site (Table 2.2). In the case of the **Annex I** the sensors were located within the closed chamber. To obtain the recorded data all sensors were connected to a datalogger (CR3000) from Campbell Scientific.

Table 2. 2 Sensors for climatic variables

Climatic variable	Sensor specifications
Temperature (ºC)	T107, CS215, 110PV (Campbell Scientific) 
Relative humidity	CS215 (Campbell Scientific) 
Radiation	LP02 pyranometer (Hukseflux) 

2.1.2.2 Crop production & development:

Production / yield – The specific crops used for each experiment are further described in table 2.5. The observed crops were grown including their production stages for all experiments presented in this dissertation (**chapters 4 to 7**), using the fruit or biomass production as indicator for plant development as well as functional unit for the environmental assessment. In the case of green bean production, during the productive periods a weekly harvest was made picking bean pots larger than 11 cm. In the case of lettuce harvest was determined either by the ripening of the lettuce head or the time period of the experiment. Finally, the harvest of pepper pods was made weekly during their production time to ensure a sufficient growth and ripening of the fruit.

Plant growth – The plants growth and development were recorded or determined for experiments in **chapters 4, 5, 6 & 7**. In the case of **chapters 4 & 5** the bean plants were evaluated on a weekly basis to determine the phenological stage. This entailed the recording of leaves, flower buds, opens flowers and bean pods. These non-destructive methods were performed during the total span of the experiment while punctual destructive measurements were made to obtain other values like leave weight, stem weight, leaf area index (LAI) and root weight. These last measurements were made also for **chapter 6 & 7**.

<u>Chlorophyll content</u> – The chlorophyll content measurements were made on a weekly basis with a SPAD CCM-200 plus (Opti-Sciences), for the bean crops in **chapter 5** further contributing to the information regarding nitrogen availability of green beans.

<u>Maintenance</u> – During the course of the performed experiments (**chapters 4 to Annex I**) other activities of overall maintenance had to be made. The preparation of the specific nutrient solution following the crop and experimental requirements as well as other practices like the removal of dead leaves and the cutting and clearing of biomass like in the case of green bean thinning. Additionally, to the crops used in the experiment for this dissertation other maintenance tasks were performed in tomato crop cultivations during 2018 and 2019. Such tasks involved pruning, tutoring and phytosanitary treatments.

<u>Water & nutrient solution</u> – The control of water flows in the experimental set-up is important not only to ensure plants are irrigated correctly but also to further analyze the nutrients flows through a nutrient and water balance. The irrigated volume was controlled through water flow meters in the irrigation panel. The leachates were controlled at the end of each cropping line and their volume, pH (G-PHT1, XS instruments) and electric conductivity (EC) (G-CONDT5, XS instruments) were determined daily. Water samples to further analyze nutrient content were taken 3 times a week.
Nutrient solution was prepared and filled into two separate tanks close to the irrigation panel, this separation was made to prevent the precipitation of the nutrients given (Table 2.3).

Table 2. 3 Nutrient solution component in tanks 1 & 2

Tank 1	Tank 2
K_2SO_4	Ca $(NO_3)_2$
K_2PO_4	$CaCl_2$
KNO_3	Mg $(NO_3)_2$
	Sequestrene ®
	Tradercorp ®/ Hortrilon ®

Depending on the experiment the nutrient solution was adapted, removing the K_2PO_4 content for experiments in **chapter 4, 5, 6 & 7** including **the Annex I** as well as removing KNO_3, Ca $(NO_3)_2$ and Mg $(NO_3)_2$ content in **chapters 5, 7 & the Annex I**. All experiment designs provided a control treatment with the full nutrient solution.

2.1.2.3 Nutrient content:

Water samples – were kept in cool or frozen conditions until analyzed, filtering each sample with a 0.22 µm filter (PTFG Syringe Filters) to be then placed in 1.5 mm vials. The nutrient content in the irrigation and leachate water samples was measured for anions using an Ion Chromatography devise (ICS2000, Dionex) (figure 2.2), obtaining results for NO_2^-, NO_3^-, SO_4^{2-} and Cl^- (Sanjuan Delmás 2017). To further obtain P, K, Ca, and Mg contents the samples were analyzed externally with an ICP-OES (Optima 4300DV, PerkinElmer).

Figure 2. 2 Ion Chromatography device

Biomass samples – (plant biomass as well as perlite) were dried in an oven until constant dry weight was achieved, to be consequentially shredded (figure 2.3). The samples were first digested with concentrated HNO_3 80% (v/v) in a single reaction chamber microwave, to be then externally analyzed for B, Mn, Fe, Cu, Zn, Na, Mg, K, P, S & Ca content with an ICP-OES (Agilent 5900). The elemental analysis for C, O, H, N, S was made through the sample combustion coupled gas chromatography (Thermo Scientific Flash 2000).

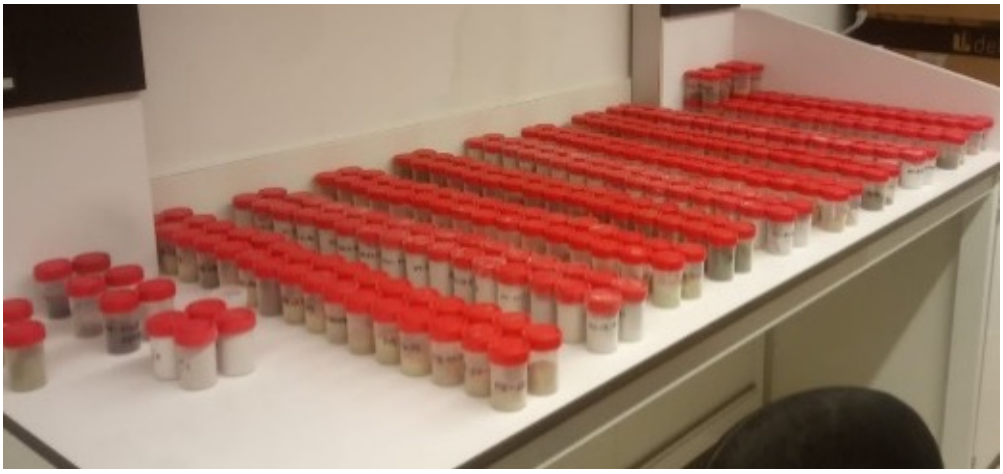


Figure 2. 3 Shredded biomass samples

2.1.2.4 Nitrogen isotopic analysis:

The nitrogen isotopic analysis made in **chapter 5** was performed to evaluate the nitrogen uptake from atmospheric fixation. The natural abundance method was used (Shearer and Kohl 1989), discerning from atmospheric nitrogen in the form of N_2 and from biological N in struvite. Biomass samples, including rhizobium fixation nodules and struvite, were dried until a constant weight was achieved and then shredded. The samples were weighted in in tin capsules and analyzed for their % δ^{15}N content using an EA-IRMS (Thermo Fisher Scientific).

For the calculation of the δ^{15}N the following equation was used (Eq1). The used standard value (0.3663) corresponds to the atmospheric ^{15}N value which gives us a result expressed in ‰. This value can be closer to the atmospheric value which usually remains close to 0 or higher depending on the other N source:

$$Eq\ 1: \qquad \delta^{15}N = \frac{Sample\ atom\ \%^{15}N - 0.3663}{0.3663} \times 1000$$

Eq 2 was then further used to obtain the percentage of nitrogen contributed by the different sources using the lowest $\delta^{15}N$ value obtained as the "B" value or fractionation, while the source 2 value was determined by the $\delta^{15}N$ value obtained from the struvite.

$$Eq\ 2: \qquad \%Ndfa = \frac{\delta^{15}N\ Source\ 2 - \ \delta^{15}N\ Sink}{\delta^{15}N\ Source\ 2 - 'B'value} \times 100$$

The obtained %Ndfa value indicates the percentage of N that comes from atmospheric fixation, in a scale between the defined value for totally biologically fixed N and other N sources. For example, if our $\delta^{15}N$ value remained close to the biologically fixed N value (close to 0) and our %Ndfa is 80% for our plant this would signify that 80% of the containing N in the analyzed tissue was obtained from N_2 fixation (figure 2.4).

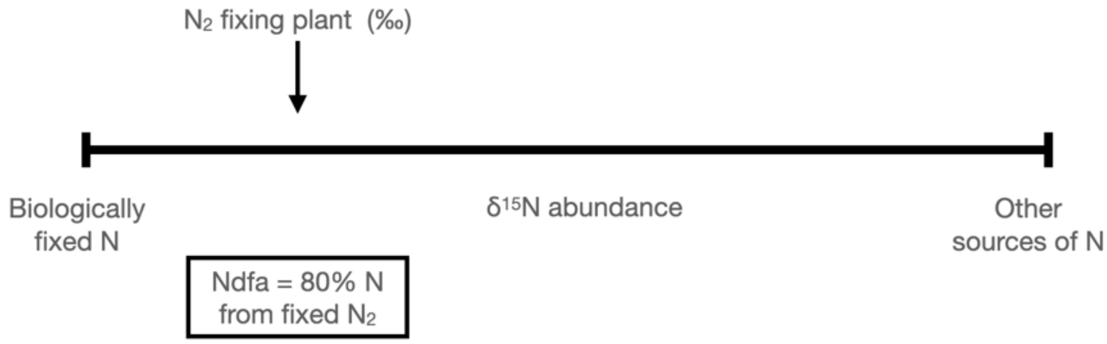


Figure 2. 4 representation of 15N Natural abundance method based on Shearer et al., 1986

2.1.3 Environmental analysis

2.1.3.1 Environmental assessment:

For **chapters 6 & 7** an environmental analysis was made with the life cycle assessment (LCA) tool. The LCA was performed following the ISO 14040 guidelines with the next four phases (ISO 2006):

Goal and scope definition – The first phase of the LCA methodology consists of the definition of a study goal which will respond for the need for the analysis and its application. The definition of the study scope on the other hand is the establishment of the key parameters in which the study will be performed, defining the system boundaries as well as the functional unit (FU) which will later be used to define the needed inventory and normalize the resulting environmental performance.

For **chapter 6** the system boundaries of the study take in consideration the fertilization of the crops and the life stages and materials bound to this operation. On the other hand,

the system boundaries for **chapter 7** are wider, taking in account the total infrastructure, equipment and operations used during the experiment time span.
The FU chosen for both chapters was based on the crop yield with 1 kg of fresh produce, resulting in two FU for **chapter 6** (1kg of fresh lettuce, 1kg of fresh peppers) and one FU for **chapter 7** (1kg of fresh green beans).

Life Cycle Inventory – Once the systems boundaries of the LCA study are defined the second step involves the collection of the necessary data to complete an inventory of all materials, inputs, energy, products, waste, and emissions encompassed within the established scope. The data can be collected through the own experimental work or collected from existing work and literature.
The on-site data obtained for the LCA assessment was obtained from:

- Cultivation system and infrastructure:
- Experimental and analytical tests
- Fieldwork

While data for struvite production and nutrient emission to air was obtained from literature and existing database.
In the case of **chapter 7** the process of allocation had to be added to the inventory definition, discerning between the flows used in the LCA and the flows serving additional purposes in the global infrastructure or operation. This allocation was made to only take into account the water harvesting of the rainwater used in the greenhouse laboratory, since the same rainwater harvesting system (RWHS) supplied water for the irrigation of the building ornamental plants.

Life Cycle Impact Assessment – The impact assessment stage entails the "translation" of the defined inventory into an impact to the environment. The generated flows in the inventory phase are associated to one or more impact category (IC) which are chosen based on the characterization method used. The characterization method defines the effect, associated to an IC, that is caused by the defined intervention (the described emission, activity, material..) (Rosenbaum et al. 2018).
In this dissertation the chosen method was ReCiPe (2016), which can be classified into 3 cultural perspectives, depending on the timespan and risk perception: Hierarchical (H), Individualist (I) and Egalitarian (E). For this thesis the hierarchical perspective was chosen due to the better application in policy related manageability (Rosenbaum et al. 2018). The IC chosen (Table 2.4) correspond to the midpoint level characterization

factors, which gives a lower uncertainty and a greater relation between de defined impact and the environmental flow (Huijbregts et al. 2017).

Table 2. 4 Chosen IC for chapters 6 & 7

Impact categories chosen from ReCiPe 2016 Midpoint (H)			
Name	**Acronym**	**Unit**	**Definition** (Huijbregts et al. 2017)
Global warming	GW	Kg CO_2-eq	Increase of integrated infrared radiative force of a greenhouse gas. Used in **chapter 6 & 7**
Terrestrial acidification	TA	Kg SO_2-eq	Fate of acidifying emissions from pollutants in the atmosphere and the soil. Used in **chapter 6 & 7**
Freshwater Eutrophication	FE	Kg P-eq	Fate of phosphorus emissions into freshwater. Used in **chapter 6 & 7**
Marine Eutrophication	ME	Kg N-eq	Fate of Nitrogen emissions into marine water. Used in **chapter 6 & 7**
Ecotoxicity	ET	Kg 1,4-DB-eq	Fate of chemical emissions (persistence, toxicity, and accumulation in human food chain) expressed as ET as the sum of freshwater ecotoxicity (FET), marine ecotoxicity (MET) and terrestrial ecotoxicity (TET). Used in **chapter 6 & 7**
Fossil Resource Scarcity	FRS	Kg oil-eq	Reduction of fossil fuel resources due to extraction. Used in **chapter 6 & 7**
Mineral Resource Scarcity	MRS	Kg Cu-eq	Decrease in ore grade due to primary extraction of a mineral resource. Used in **chapter 6**

The LCA in this dissertation was performed with the Simapro 9.1 software, which gives access to inventories from which Ecoinvent 3.7 was used for **chapters 6 & 7**.

Interpretation – The final step of the LCA methodology is the result interpretation which depends on the initial goal of the analysis. This interpretation can entail a comparative outcome and therefore help in the decision making through an environmental perspective or can be informative to understand the impact of a certain activity or component of the evaluated work.

2.1.3.2 N_2O emission assessment

To increase accuracy in environmental impact assessments it's crucial to generate knowledge of the emissions obtained from different processes which have not been assessed previously. For

the Annex I a methodology for the N_2O emission factor (EF) was defined to unveil the potential of struvite fertilization in hydroponic soilless agriculture for the reduction of greenhouse gases (GHG's).

For the determination of N_2O emissions of the lettuce crop initial experiment were made to determine if struvite could be the sole N source for the crop. After 3 validation experiments the crops were grown within a closed chamber (figure 2.5). As explained in section 2.1.2.1 the chamber was equipped with temperature, humidity, and radiation sensors. The closed chamber split in half through an inner wall will provide space for 16 plant pots with 1L capacity filled with perlite substrate on each side with a total volume of 2,64m3 (1.32m3 respectively).

Figure 2. 5 Closed chamber during N_2O emission experiment with green beans

On each side of the chamber a sampling inlet was included to extract the concentrated air after the closing of the chamber. The sampling was made with a Mini VAC-U-Chamber (SKC) filling specialized airtight 1-L air bags (SKC1 – model 1.252e01).

The air samples were then analyzed with a gas chromatograph 6890N (Agilent) (figure 2.6) and the Agilent HP-PLOT-Q 30 m, 0.53 mm, 40 mm column injected manually with an airtight Pressure-Lok® Precision Analytical Syringe (VICI).

For the calibration of the GC a calibration curve was made using known N2O concentrations from atmospheric air samples injected at different volumes from 1 ml to 0,2 ml.

Figure 2. 6 Gas chromatography device

2.1.4 Statistical analysis

The statistical analysis performed in this dissertation in **chapters 4, 5 & 6** was made using the RStudio software as well as the resulting graphics for all chapters except **chapter 3**.

The Shapiro-Wilk test was used to test normality in the resulting data while the Levene's test was further used to determine homogeneity of variance.

For parametric data the Duncan multiple range test was used to identify statistical significance in the treatments. The Duncan test has been defined as a suitable test for agronomic data even though Type 1 error could be more "liberal", due to the potential interference of "real world setting" variables where the Duncan multiple range test confers greater importance to the statistical significance. Non-parametric data on the other hand were analyzed for significance using the Kruskal-Wallis test. The statistical analyzed data was identified with alphabetical letters to differentiate statistical difference.

2.2 Case studies & Infrastructure

The experimental assessments performed in this dissertation (**Chapters 4-7**) are mainly conducted in the integrated rooftop greenhouse of the ICTA-UAB building in the Universitat Autònoma de Barcelona. For **chapter 3** the area under study for the calculations of nutrient recovery and use were made on the scale of the metropolitan area of Barcelona (AMB).

2.2.1 Metropolitan Area of Barcelona

The metropolitan area of Barcelona comprises a region of 636 km^2 with 36 municipalities (AMB 2022) being one of the biggest metropolitan areas in Europe having a population of more than 3 million people. This area has access to two rivers Besòs and Llobregat as well as to the

Mediterranean Sea and mountain being in contact with the serralada de Collserola, serralada del Garraf and serralada del Maresme. In comparison to other major metropolitan areas in Europe the AMB holds the 8th position in population density with around 5.000 inhabitants/km^2. The land use of this area contains 55% of forest, beach, parc's and unoccupied soil while 20% is residential area, and 25% is covered by industry and other land uses (AMB 2022).

These characteristics make the AMB an interesting region to study UA, being subjected to numerous previous studies on the adequacy of different areas to host UA infrastructure. The population density and the constraining geography make the AMB a good example of urban area that can and needs to innovate within the existing land use, while the Mediterranean climate and being a coastal location can be beneficial for vegetable production. On the other hand, the direct contact to the sea can also entail a greater need for waste management and nutrient removal in wastewater and nutrient management in case of UA upscaling.

2.2.2 Urban Agriculture Laboratory

Chapters 4 – 7 including **Annex I** are based on the experimental trials performed in the ICTA-UAB iRTG, specifically in the urban agriculture Laboratory 2 (LAU2). The ICTA-UAB building is situated in the university campus from the Universitat Autònoma de Barcelona in the municipality of Cerdanyola del Vallès. The building hosts the ICTA (Institute de Ciencies I Tecnologia Ambiental) and the ICP (Institute Català de Paleontología) and was constructed in 2014 with a novel design winning the LEED Gold certification (figure 2.7). The building has four areas located on the fourth floor where agricultural activity can be made, where currently two areas (LAU1 and LAU2) are actively used. The connectivity between greenhouse and building is total, using positive climatic synergies to regulate the internal temperature of the cropping areas. This enables year round production to with lower energy inputs for building and GH climatization (Muñoz-Liesa et al. 2020).

The ornamental vegetation as well as the crops are irrigated with rainwater collected from the building roof and the neighboring building EUREKA, which is then stored in two tanks underground. The yellow water generated in the building is filtered and further processed in a biofilter in front of the building.

The cropping spaces as well as the rest of the building are separated from the exterior through polycarbonate outer shell which open and close to ensure ventilation inside the building. The laboratory spaces can also be more or less exposed to the building climatic conditions through a separation sheet between the atrium (central area) and the cropping area.

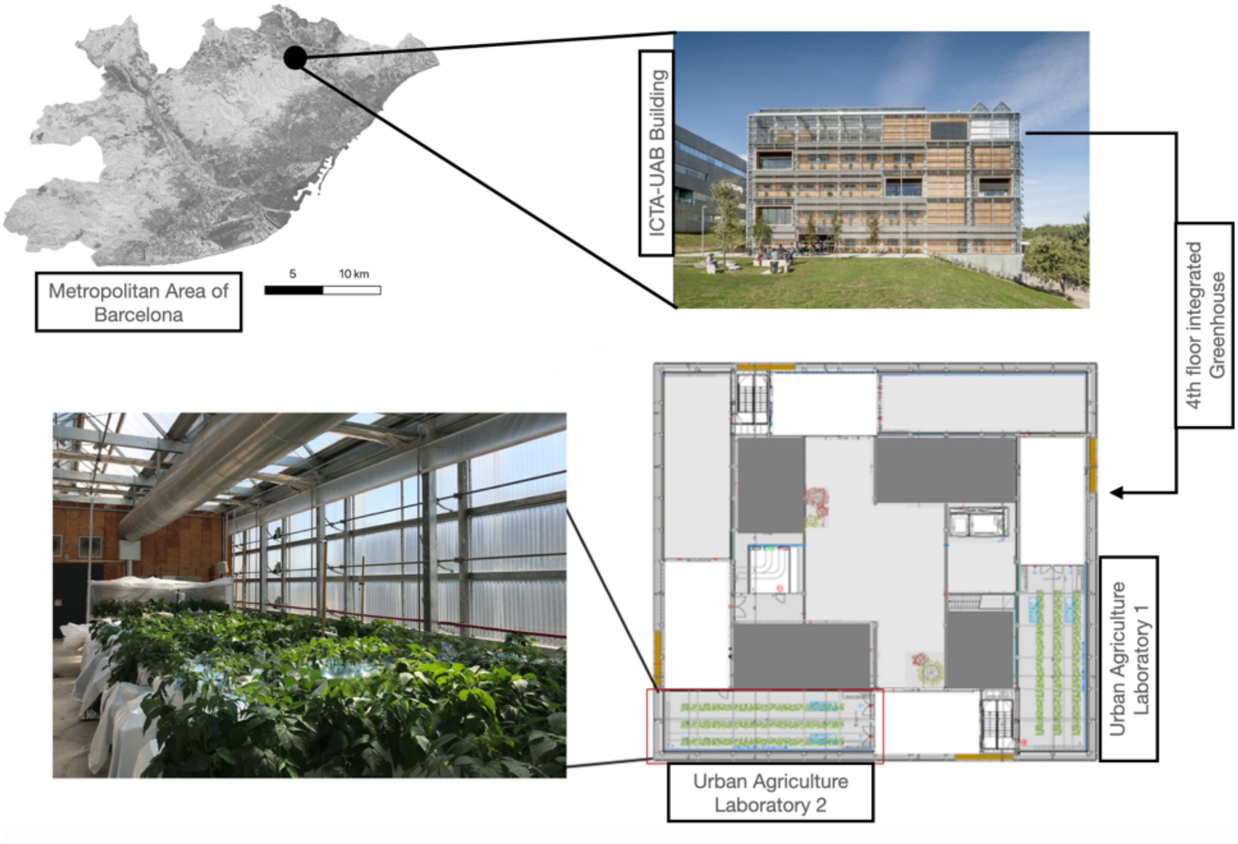


Figure 2. 7

The LAU2 cropping area is located on the southwest facing corner of the building and has a total extension of 122.8 m^2 with 84.6 m^2 of effective cropping area. Within this area 12 cropping lines are placed in 6 rows (from NE to SW) as well as an additional row (composed of two cropping lines) inside a closed chamber. The rows are 0.5 m wide and 4 m long and can hold up to 8 perlite bags (40 L). Experiments for **chapters 4, 5 & 7** were made in the LAU2 using perlite bags to grow green beans with a plantation frame of 0.125m^2. The experiment for **chapter 6 & Annex I** on the other hand were performed on pots with 1L and 5L capacity to produce lettuce and pepper respectively.

Table 2. 5 Summary on the experimental work divided by chapter

Chapter	Crop	Experiment type	Nr of plants	Duration (days)	Time
Chapter 4	Green Bean	Validation	320	78	September- December 2018
		Determination	256	72	September- November 2019
Chapter 5	Green Bean	Determination	192	84	January- April 2019
Chapter 6	Lettuce	Determination	112 x3	27 x3	June- September 2020
	Pepper	Determination	32	81	
Chapter 7	Green Bean	1st experiment	192	84	January- April 2019
		2nd experiment	192	84	February- May 2020
Annex I	Lettuce	Validation	-	30 x2	September- December 2021
		Determination	32	28	March- April 2022

2.2.3 Irrigation system

The i-RTG irrigation system used in all experimental assessments was hydroponic with the use of perlite as substrate. The perlite substrate with a neutral pH 7, a low electric conductivity of 0.009 dS·m^{-1} and granulometry range (0-6) served as an inert plant support appropriate for experimental purposes, especially with the focus on crop fertilization. The irrigation water was obtained from the rainwater harvesting system tank and pumped to the LAU2 into a 300L tank.

The automatic irrigation system was controlled with a Hunter® programmer for the activation of electrovalves to define the sector to be irrigated within the GH. The rainwater was mixed using two Dosatron® injectors connected to the fertilization tanks explained in section 2.1.2.2. For the irrigation of different treatments and different nutrient solutions a secondary irrigation incoming from the LAU1 was supplied. Additionally, a third irrigation source was installed for the experimental assessment in **chapters 6 & Annex I** with an additional nutrient solution tank and injection device.

Part II

Nutrient circularity in urban areas

Literature review on the potential of urban waste for the fertilization of urban agriculture: A closer look on the metropolitan area of Barcelona

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Chapter 3: Literature review on the potential of urban waste for the fertilization of urban agriculture: A closer look on the metropolitan area of Barcelona

Abstract:
Urban agriculture activities are increasing in popularity and importance due to the greater food demands and reduced agricultural land, also advocating for greater local food supply and security as well as the social and community cohesion perspective. This activity also has the potential to enhance the circularity of urban flows, repurposing nutrients from waste sources increasing their self-sufficiency and reducing nutrient loss into the environment.

The present work aims to define recovery technologies outlined in literature to obtain relevant nutrients like N and P from waste sources in urban areas. Through literature research tools a first definition of the waste sources was made differentiating two main groups: food-, organic-, biowaste and wastewater. Up to 7 recovery strategies were found for the food-, organic-, biowaste source while 11 were defined for the wastewater, mainly focused on the recovering of N and P which are applicable in UA in different forms.

The potential of the recovered nutrients to cover existing and prospective UA sites was further assessed for the metropolitan area of Barcelona. Nutrient recovery from current composting and anaerobic digestion of urban sourced organic matter obtained each year in the area as well as the composting of wastewater sludge, struvite precipitation and zeolite adsorption in wastewater effluent generated yearly in the existing WWTP were assessed. Results show that P requirements for the current and prospective UA in the area can be met 2,7 to 380,2 times and 1,7 to 117,5 for N depending on the recovery strategy. While the present results are promising, current perceptions and legislations don't facilitate the application of nutrient recovery strategies although a change is expected in the near future due to the pressing issue of P depletion.

3.1 Introduction

3.1.1 Urban agriculture: reducing distances and optimizing the use of space

For the past decades the increase of population in expanding urban areas has risen the demand for food (United Nations 2019), putting great pressure on the agricultural industry to supply cities within the existing agricultural land. This additional stress builds over the existing challenges on agriculture, including climatic instability and land degradation (Dsouza et al. 2021). These pressures have led to a need of highly intensified production systems which can further contribute to the land degradation, use of non-renewable resources and greenhouse gas emissions (Chojnacka, Moustakas, and Witek-Krowiak 2020; Dsouza et al. 2021). Agriculture and the total food system currently is responsible for a 21-37% of all GHG emissions, which are expected to increase by 30% in 2050 due to population growth, dietary change, income growth and consequentially land-use change (IPCC 2019). A way to alleviate these great pressures to feed cities would be to make urban centers more resilient and self-sufficient themselves. In other words, enabling the environment for food production within the city (Ercilla-Montserrat et al. 2019; Sanyé-Mengual et al. 2018).

The concept of UA has gained importance in the last years and is regarded as one plausible solution for the supply of food in urban areas as well as creating a sense of community and incrementing urban green (Lal 2020; Wielemaker et al. 2019). The potential benefits of UA also include shortening supply chains and consequently reducing transportation and food losses (Sanyé-Mengual et al. 2015; Sanyé Mengual 2015; Toboso-Chavero et al. 2019a). These benefits may change depending on the typology of UA applied, which can greatly vary between soil-based outdoor agriculture to indoor hydroponic vertical farming.

UA has different appearances and can present itself in different shapes and forms as well as motivations and functions. Urban residents are often guided by the traditional images of agriculture, therefore most family and community gardens, as well as social peri-urban farms are conceived to follow principles of organic or agroecological farming on soil. UA can still be performed on the soil on the ground although this concept has evolved in recent years and other ways of production have adapted to these ever-growing cities (Despommier 2013). However, while many cities still have plots on the soil in peri-urban or urban locations others have moved from the narrowing and highly demanded spaces on ground to the forgotten and often under regarded building rooftops (Appolloni et al. 2021).

Vegetable production on rooftops and inside buildings has been growing in interest in the past years and has been seen as a more viable alternative to a profitable production of goods, while agriculture on soil in urban areas has more vastly been regarded as a social activity rather than economically profitable.

The production systems most suitable for rooftop and indoor urban agriculture are mainly based on soilless systems with the use of alternative substrates to avoid heavy loads. The practices of hydroponic/aeroponic agriculture as well as aquaponics have shown to be great alternatives for the building based agricultural systems but can face other downsides. The productions of vegetables with these kinds of systems can entail a great investment in infrastructure and need technical specialization for their manipulation. On the other hand, soilless production systems relay on the use of mineral and synthetic fertilization given directly to the plants through the irrigation. While productivity may increase compared to soil-based agriculture, these practices imply a great environmental burden being still very linear systems (Dsouza et al. 2021; Sanjuan-Delmás et al. 2018).

Efforts have been made to reduce or minimize the emissions of UA production related to the fertilization, by closing flows within the crop system, reusing the leachate nutrients again for the same crop or as a cascade system on less demanding crops, before being discarded or further reused (Rufí-Salís et al. 2020; 2020). While this can certainly be a plausible solution to minimize fertilizer and water loss into the environment it does not entirely solve the burden of the nutrient extraction nor generation to further increase agricultural activity inside the city, like in the case of phosphate rock mining or synthetic nitrogen production (Cordell, Drangert, and White 2009; Cordell and White 2014). On the other hand, additional infrastructure is due to be installed to close the water flow as well as additional equipment to ensure the stability of the given nutrient solution as well as the control of pests and diseases making the system much more complex.

While the concept of UA and the use of building rooftops can be appealing to increase agricultural activities in cities the great technification and the greater use of mineral and synthetic fertilizers can be major constraints for its effective and sustainable application. Therefore, a change of the fertilization for soilless agriculture must be put into practice altogether.

3.1.2 Why the need to reuse fertilizers?

The extraction and synthetization of fertilizers has been an ongoing activity since the agricultural green revolution, turning into a necessity to maintain the great productions of goods to feed the current world. While earlier ways of fertilization included the use of urine, manure, human excreta and guano, the modern agricultural industry relies on the mining and synthetization of plant available nutrients (Sun et al. 2018b). This practice has greatly shifted the global nutrient pools with the export and import of these products to ensure fertilization of large land extensions (Villalba et al. 2008).

This nutrient pool shift has generated several consequences not only due to the extraction of nutrients through excessive mining, but also through the emissions generated by their transport and application elsewhere. Fertilization is also responsible of leakage of excess nutrients toward the environment, generating exosystemic problems that include acidification, eutrophication, or emissions of GHG's into the atmosphere (Cordell, Drangert, and White 2009; Yi Liu et al. 2008). The current anthropogenic sources of N_2O are mainly originated due to nitrogen over fertilization and bad management, with soil emissions of 3 MtN_2O-N each year (IPCC 2019). Today, about a 90% of the phosphate rock extracted is still used to produce agricultural fertilizer while 50% of the nitrogen demanded for agriculture is supplied through synthetic N generated through the

Haber-Bosh process (Zabaleta and Rodic 2015; Chojnacka, Moustakas, and Witek-Krowiak 2020). The application of P fertilizers is also regarded a greatly inefficient with extensive losses due to erosion and leaching, with only about 10% of the applied P reaching consumers (Chojnacka, Moustakas, and Witek-Krowiak 2020). This non-renewable resource is expected to be exhausted in only a few centuries if it's extraction continues at present rates, reaching its peak production rate between 2030 and 2040 (Möller et al. 2018).

The reduction of dependency from non-renewable nutrients and the reduction of the emission of P into the environment could largely be achieved with recovery and reuse strategies, which are currently barely carried out (Oarga-Mulec et al. 2019; Chojnacka, Moustakas, and Witek-Krowiak 2020). A large share of nutrient losses in urban areas is associated with food and human waste. Estimations of the global annual food waste indicate that 931 million tonnes were generated in 2019, around 121 kg per capita each year, where around 60% was originated from households (Forbes. H, Tom. Q, Clementine. O 2021). About 97% of the global food waste is disposed in landfills, where inappropriate management may cause nutrient leaching into the environment causing eutrophication and accumulation in soils as well as methane emission and odor (Chojnacka, Moustakas, and Witek-Krowiak 2020; Ren et al. 2020).

Through a circular economy mindset the potential of the reuse and recycle of wastes are explored and the capacity for self-sufficiency and resilience enhanced, escaping from linear production approaches that entail the need for importing and exporting resources into and out of the system (Girotto and Piazza 2021; de Kraker et al. 2019). Current urban nutrient cycles mostly don't consider a circular approach to manage and recycle nutrients, and this greatly reduces their self-sufficiency. The import of external manure, or other organic or synthetic nutrients, to pursue UA can seem redundant due to the increase of nutrient loss within the urban ecosystem as well as the already existing potential source of nutrient within urban areas (Wielemaker et al. 2019; Martin, Poulikidou, and Molin 2019).

Here is where UA can increase its benefits for a sustainable urban development, not only raising its food security with local production of goods, as well as the increase of green spaces and biodiversity, but serving as a destination for urban recovered nutrient to further close urban material flows (de Kraker et al. 2019). The increasing interest in UA and hydroponic production with the use of recycled nutrients can serve as a marker to further encourage nutrient recovery practices that can also become profitable in the near future (Martin, Poulikidou, and Molin 2019). The focus on waste disposal and nutrient recovery has been greater in current years with the impulse of new EU regulations and development goals striving for better waste management strategies with the reduction of landfilling as well as making the P recovery in wastewater treatment plants (WWTP) a requisite (Kratz, 2019).

3.2 Methodology

3.2.1 State of the Art – Nutrient recovery and reuse in urban environments

This work comprises the literature analysis of the work made on the nutrient recovery of urban waste streams for the nutrient supply for UA. For this purpose, an initial literature search was made through the search platform Scopus with the keywords "nutrient recovery" AND "Urban agriculture" OR "Urban waste".

This initial search gave a result of 17 scientific articles (Table 3.1) that could be further sorted into two main categories established depending on the waste type or source. These categories were defined as "food-bio-organic waste" and "wastewater". For the literature review only nutrients that can be sourced in urban areas have been identified, leaving out potential organic material from the agricultural industry or other imported material. It is also worth mentioning that only wastes have been regarded and no defined products made for fertilization.

From these categories a second search was elaborated with the key words "nutrient recovery" AND "Urban" AND "organic waste" AND NOT "wastewater" (11) "nutrient recovery" AND "food waste" AND NOT "wastewater" (43), "nutrient recovery" AND "bio waste" AND NOT "wastewater" (4) and "nutrient recovery" AND "urban wastewater" (27). All literature before 2017 was excluded to avoid outdated waste treatment methodologies.

Applying these criteria, the articles obtained for the search of nutrient recovery based on Food-Bio-Organic waste obtained a total of 24 results, while 28 were retrieved for wastewater sourced fertilizers. These articles were then classified into categories corresponding to the recovery technology as seen in tables 3.2 and 3.3 for food-bio-organic waste and wastewater respectively. The literature was further completed with additional research on Scopus, Web of Science and Google scholar for each nutrient recovery process and waste treatment to encourage a better description and insight.

With the initial literature search two main nutrient sources for urban nutrient recovery were identified, being wastewater- based residues as well as food wastes and other organic and bio wastes found in urban ecosystems mainly originated in households, food processing and catering industries or green areas like gardens and parks (Möller et al. 2018). All recovery technologies identified in tables 3.1, 3.2 and 3.3 have been depicted in figure 3.1 for a better understanding of the waste flows and possible combinations between methodologies within each waste type as well as different waste types combined.

Table 3. 1 Waste type, treatment and target nutrient identified with primary search on urban waste derived nutrient recovery.

	REFERENCE	WASTE TYPE /ORIGIN	RECOVERY TECHNOLOGY	TARGET NUTRIENTS	APPLICATION
1	(Weidner and Yang 2020)	Organic waste	Composting/ Insect Rearing/ Anaerobic digestion	NPK	Soil based agriculture/ Hydroponic/ Aquaponic
2	(Shrestha, Small, and Kay 2020)	Organic waste	Composting	NP	Soil based agriculture
3	(de Kraker et al. 2019)	Kitchen waste/ garden residue/ Urine	Anaerobic digestion/ Vermicomposting/ composting / struvite precipitation	NP	Urban agriculture/ Municipal Green/ Peri-Urban Agriculture
4	(Kjerstadius et al. 2017)	Centralized and source separated food waste and wastewater	Struvite precipitation/ Anaerobic biogas / Biological Nitrogen removal/ Sludge composting	NP	Agriculture
5	(Macura et al. 2019)	Agricultural residuals/ Domestic Wastewater (Blackwater)	Anaerobic digestion/ Struvite Precipitation/ Ammonia stripping	NP	Agriculture/ Food and Feed production
6	(Lohman et al. 2020)	Source separated urine	Struvite precipitation/ Urine hydrolysis/ Ion exchange	NPK	Agricultural irrigation
7	(Firmansyah et al. 2021)	Source separated domestic grey and black water/ Kitchen waste/ Centralized wastewater	Up flow anaerobic Sludge bed/ Composting/ Trickling filter	NP	Agriculture
8	(Pimentel-Rodrigues and Siva-Afonso 2019)	Source separated urine	Urine storage	P	Green roofs
9	(Möller et al. 2018)	Landscape green waste/ Urban household waste/ Catering and retailer organic waste/ Wastewater/ industrial waste	Sewage sludge/ Incineration-Thermal treatment/ Composting/ Anaerobic digestion/ Chemical precipitation	P	Agriculture
10	(Akoto-Danso et al. 2019)	Domestic Wastewater		P	Soil based Agriculture
11	(Podder, Reinhart, and Goel 2020)	Landfill leachate	Anaerobic digestate/ Struvite precipitation	NP	
12	(You, Valderrama, and Cortina 2019)	Urban Wastewater	Sorption (synthetic Zeolites Ze-CA)	NP	Food production
13	(Dsouza et al. 2021)	Urban Biowaste	Composting/ Compost tea/ Multiple parallel mineralization/ Ozonation	NPK	Hydroponic/ Bioponic
14	(Guaya et al. 2018)	Urban Wastewater	Sorption (synthetic Zeolites)	NPK	Food production
15	(Magwaza et al. 2020)	Domestic Wastewater		NP	Hydroponic
16	(Rufí-Salís et al. 2020)	Urban Wastewater	Struvite	NP	Urban Agriculture
17	(Calabria, Lens, and Yeh 2019)	Domestic Wastewater	Sorption (natural Zeolite)	N	Hydroponic

3.3 Results

3.3.1 Nutrient recovery from food and bio-waste

City landscapes and households can produce a great deal of biomass throughout the year, up to 400 to 800 g daily per person (M. Ahmed et al. 2019), which could be increased with the integration of agriculture within urban areas (Manríquez-Altamirano et al. 2020; Dsouza et al. 2021). Biomass and food waste are often underused sources of nutrients, although there is significant potential for the recovery of non-renewable and energy intensive nutrients like P and N (Zabaleta and Rodic 2015; Idowu et al. 2017). Not only the waste generated in the household is of great importance, but the over production and further disposal of food surplus is also part of the problem. Previous work has identified that up to 40% of produced food is wasted and savings from up to 25% of mined P can be made through the reduction of these food wastes (Drangert, Tonderski, and McConville 2018). The landfilling and bad management of food and biowaste can lead to great amounts of anaerobic decomposition that cause the emission of greenhouse gases and therefore needs to be avoided and processed in a controlled way (Dsouza et al. 2021). Increasing efforts to reduce the landfilling of organic waste advocate for an effective source separation and further treatment of the municipal waste to recovery and recycle nutrients from the organic fraction (Davidsson et al. 2017). This appears to be highly relevant, also considering that in 2018 only 30% of the European organic fraction is currently source separated and further recycled (Möller et al. 2018) while the rest is landfilled or incinerated (Sun et al. 2018b).

3.3.2 Nutrient recovery from wastewater

The perception on WWTP has been shifting and evolving in the last years, transitioning from waste carrying and removal technologies to resource recovery plants for nutrients as well as for energy (You, Valderrama, and Cortina 2019). Currently, wastewater and the processed sewage sludge and effluent are the main P carriers that are further recycled in agriculture in some European countries, although regulations often do not enable the environment for its direct use in conventional agriculture, and therefore is still often just being incinerated (Möller et al. 2018). A better management of the wastewater streams is endorsed in European regulations, pursuing a greater circularity in urban areas with larger water and nutrient recovery as well as the reduction of energy consumption and GHG emissions (Marinelli et al. 2021; You, Valderrama, and Cortina 2019). The proximity of WWTP to urban areas gives them a great advantage to supply nutrient for UA, avoiding great transportation or potential storage problems. The potential of this circularity has been seen in previous studies in a neighborhood in Munich, Germany to reach savings of 26% of freshwater resource while promoting UA to produce 66% and 246% of fruit and vegetables demand respectively (Marinelli et al. 2021).

Table 3. 2 Waste type and treatment identified with secondary search on Food-Bio-Organic waste derived nutrient recovery.

WASTE TYPE	TREATMENT	REFERENCE
Agro-food waste, kitchen waste, Textile sludge	Anaerobic digestion	(Oarga-Mulec et al. 2019), (Davidsson et al. 2017), (Pleissner, Lau, and Ki Lin 2017), (Ren et al. 2020), (Kumar, Samuchiwal, and Malik 2020), (Gienau et al. 2018b), (Möller et al. 2018), (W. Wang and Lee 2021), (Ravindran et al. 2021), (Reilly et al. 2021), (Weidner and Yang 2020), (Battista et al. 2020)
Food Waste	Hydrothermal Carbonization (HTC)	(Idowu et al. 2017), (W. Wang and Lee 2021), (Sarrion et al. 2021)
Food waste/Green Waste/Sewage sludge, Urban organic waste	Composting/ Co-composting/ Compost tea	(Mortula et al. 2020), (Awasthi et al. 2020), (Möller et al. 2018), (Ravindran et al. 2021), (Schröder et al. 2021), (Milinković et al. 2019), (Weidner and Yang 2020), (Shrestha, Small, and Kay 2020), (Dsouza et al. 2021)
Onion waste, Organic waste	Vermicomposting	(Pellejero et al. 2020), (Ravindran et al. 2021), (Schröder et al. 2021), (Milinković et al. 2019)
Organic Waste	BSLF/ Insect rearing	(Magee et al. 2021), (Weidner and Yang 2020)

Table 3. 3 Waste type and treatment identified with secondary search on wastewater derived nutrient recovery.

WASTE TYPE	TREATMENT	REFERENCE
Treated wastewater (primary effluent)	Zeolite sorption,	(Guaya et al. 2018), (You, Valderrama, and Cortina 2019), (Calabria, Lens, and Yeh 2019)
Treated wastewater (primary effluent)	Hydroponic agriculture	(Magwaza et al. 2020)
Source separated urine	Green roofs	(Pimentel-Rodrigues and Siva-Afonso 2019)
Untreated wastewater	Soil based agriculture	(Akoto-Danso et al. 2019)
Filtered untreated wastewater, Urine, Treated wastewater (primary effluent), Treated water (secondary effluent)	Photobioreactor	(Dalvi, Chawla, and Malik 2021), (Chatterjee et al. 2019), (Karbakhshravari et al. 2020), (González et al. 2020), (Escudero et al. 2020), (Sánchez-Zurano et al. 2021), (Robles et al. 2020a), (González-Camejo et al. 2019), (Yulistyorini 2017), (Khandan et al. 2020)
Treated wastewater (primary effluent), Source separated urine	Struvite precipitation	(Karbakhshravari et al. 2020), (Kjerstadius et al. 2017), (Rufí-Salís et al. 2020), (Rodrigues et al. 2019)
Treated water (secondary effluent)	Membrane filtration	(Vecino et al. 2019)
Wastewater sludge	Pyrolysis	(Jellali et al. 2021), (Tomasi Morgano et al. 2018),
Wastewater sludge, Urine	Anaerobic digester	(Kjerstadius et al. 2017), (Srivastava et al. 2020), (de Kraker et al. 2019), (Firmansyah et al. 2021)
Wastewater	AnMBR	(Jiménez-Benítez, F. J. Ferrer, *et al.*, 2020a), (Jiménez-Benítez, J. Ferrer, *et al.*, 2020b)
Wastewater sludge	Composting	(Oarga-Mulec et al. 2019), (Firmansyah et al. 2021)

3.3.3 Nutrient recovery from WWTP

The outline of a WWTP can incorporate more or less steps depending on the purpose of the treated water but commonly the wastewater is pretreated to remove any potential solid waste as well as oils. Then the wastewater is sent to a primary clarifier to settle the primary sludge and obtain a clearer wastewater which can then undergo a primary treatment with activated sludge. While more simple layouts may add a secondary clarifier after the primary treatment, other WWTP incorporate a secondary treatment which usually is the nitrification and denitrification process (Vilanova, Santín, and Pedret 2017; Ostace et al. 2013). Further tertiary processes can be added for further nutrient removal, where chemical removal or ultrafiltration processes are usually installed (You, Valderrama, and Cortina 2019).

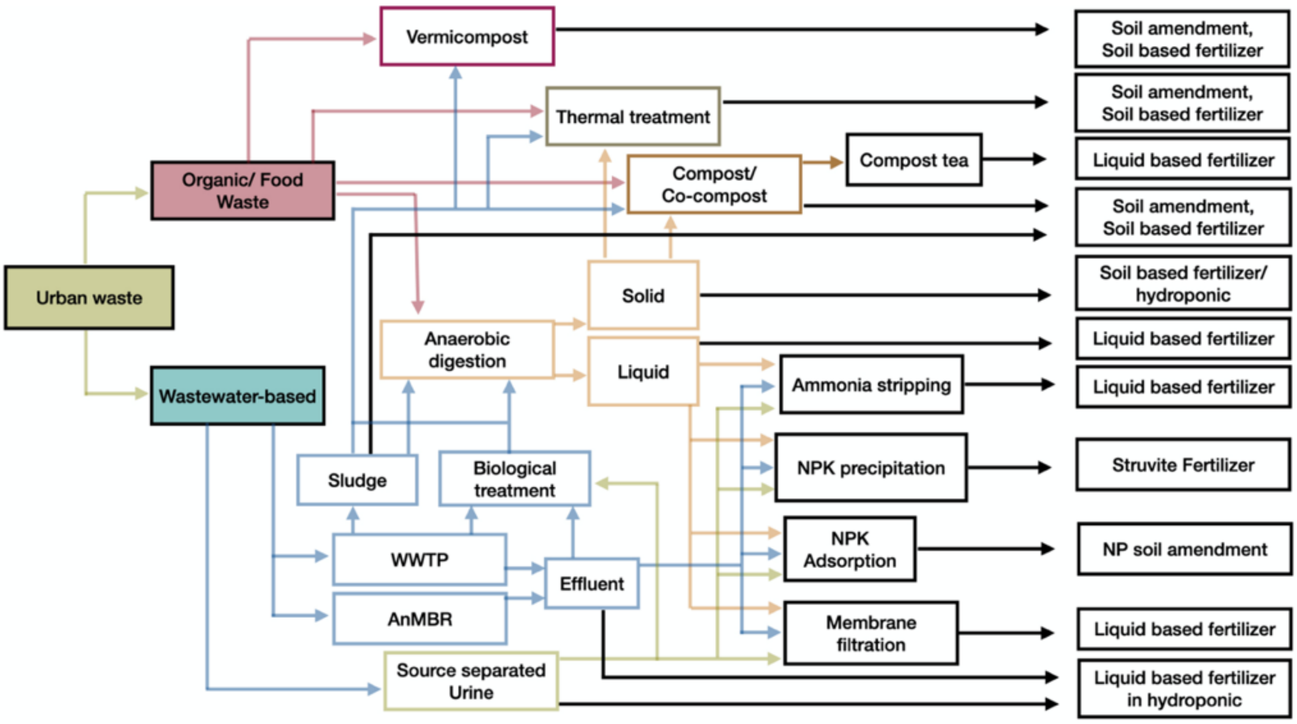

Figure 3. 1 Main flows of nutrients from urban waste to agricultural fertilizer identified from the studies compiled in tables 1, 2 and 3.

The sludge from the first, second clarifier, and nutrient removal processes is treated parallelly through mixing, dewatering, anaerobic digestion, or composting systems (Vilanova, Santín, and Pedret 2017).

WWTP's have a great potential to house nutrient recovery technologies. These installations are the main collectors of domestic wastewater in urban areas and have the sole purpose to bring the water composition below thresholds established by environmental regulations. The reduction of nutrients in the water is a requisite that usually can be implemented through biological or chemical treatments (Guisasola et al. 2019; Vilanova, Santín, and Pedret 2017). Biological treatments have been seen to be more reliable and effective methods for a great reduction of nutrients. However, as thresholds for nutrient emissions into the environment are reduced, chemical treatments are instead gaining more popularity in water treatment processes (Hospido et al. 2004).

The chemical treatment mainly consists of the reduction of soluble nutrients (mainly P) to particulate nutrients through their binding and precipitation. This can be achieved with the addition of metal salts like Ca, in the form of lime, Fe and Al. Once precipitated, nutrients can be further disposed in the sedimented sludge. The main constraint for chemical treatments is the amount of chemicals needed to effectively remove the soluble P which must be in a 1:1 relation to the present P. This causes a great investment not only for the metal salts themselves but also for the required storage infrastructure (Foley et al. 2010; Crini and Lichtfouse 2019). The excessive application of these chemicals can, on the other hand, lead to unwanted chemical reactions. For the chemical removal of N, several techniques are available, including air stripping, ion exchange or membrane filtration. In order to increase the sustainability of the process, the biological treatment has recently assumed the form of a required step for initial nutrient removal. It consists of the removal of N and P with microbial activated sludge (Vilanova, Santín, and Pedret 2017). The removal of N is based on the decomposition of the incoming organic N into ammonia (NH_4^+) through aerobic, heterotrophic bacteria, which can undergo further steps for successful ammonia removal. Most commonly the produced ammonia can be released in an aerobic environment for a nitrification process generating nitrate (NO_3^-) with the help of autotrophic bacteria. Finally, a denitrification step can be added to generate N in the form of gas (N_2) that can be exhausted into the environment (Hou et al. 2021). This is performed under anaerobic conditions by heterotrophic bacteria that require the addition of organic matter as a carbon source (Vilanova, Santín, and Pedret 2017).

The biological removal of P on the other hand is based on the addition of Phosphate accumulating organisms (PAO's), which also require anaerobic and aerobic stages. These bacteria can release P in form of phosphate (PO_4) in anaerobic conditions while P can be captured in aerobic environments (Poh et al. 2021; Hou et al. 2021; Close et al. 2021; Vilanova, Santín, and Pedret 2017; Guisasola et al. 2019). This process can be combined with the activated sludge and nitrification and denitrification stages for organic N removal (Sarvajith and Nancharaiah 2022; Hou et al. 2021), obtaining biomass that can be further disposed with the settled sludge or further re-released into an aqueous phase to be chemically precipitated (Anders et al. 2021).

Many of the technologies described in the upcoming sections can be added into the WWTP outline for further nutrient removal and most importantly recovery.

3.3.4 Nutrient recovery from anaerobic digestion

The recycling of nutrients from bio-waste through anaerobic digestion has largely been studied and considered to have a great potential in urban waste management due to the generation of methane for energy consumption as well as a high nutrient recovery in the digestate with a small fraction loss of phosphorous and nitrogen (around 0 to 10%) (Oarga-Mulec et al. 2019). Apart from macronutrients contained in the digestate other compounds like micronutrients, hormones and other organic elements can entail a positive effect on plant and soil microorganisms (Ren et al. 2020).

The process of anaerobic digestion has four stages that involve different key microbial communities in an oxygen deprived environment. During the first stage the hydrolysis and breakdown of the feed component polymers occurs, being reduced to monomers by microbial secreted hydrolases (Sikora et al. 2017; Ravindran et al. 2021). The acidogenesis is the second phase of the anaerobic digestion, where the hydrolyzed compounds undergo an acidic fermentation, followed by the acetogenesis with the formation of acetate, hydrogen and carbon dioxide, that are further transformed into methane in the methanogenesis during the last step (Ravindran et al. 2021).

The resulting digestate is mainly used in agricultural fields due to the great nutrient content, especially for nitrogen, phosphorous and potassium (Gienau et al. 2018b). The liquid and solid fraction of the remaining digestate contains mineral as well as organic forms of nitrogen greatly available for plants while P is mainly recovered in the form of phosphate (Vögeli et al. 2014; Zabaleta and Rodic 2015; W. Wang and Lee 2021). While the liquid fraction has a greater nitrogen (in the form of dissolved ammonia) and potassium content, the solid fraction has greater amounts of total nitrogen and phosphorous. Compared to sludge or dairy, food waste anaerobic digestate contains greater amounts of nitrogen in the form of ammonia and a greater N:P ratio (Dutta et al. 2021).

Further treatments can be applied to the liquid fraction to further recover nitrogen like ammonia stripping, membrane filtration, P precipitation, nutrient sorption or biomass production (Gienau et al. 2018b; Vaneeckhaute et al. 2017).

While the generated digestate proves to be a good source of nutrient recovery the generation of volatile nitrogen compounds like NH_3 can increase the impact of this waste. The composition of the digestate, ammonia release and the potential methanogenesis greatly depends on the incoming feed with suggested C/N ratios of >20. Previous work on anaerobic digestion treatments of food waste showed C/N ratios of 49, although the content of oils and spices in food wastes can be detrimental for the methanogenic activity, slowing it down (Kumar, Samuchiwal, and Malik 2020).

To obtain a digestate of the required quality for UA the organic biomass must be collected free of impurities which is difficult to achieve even through selective organic waste collection (Naroznova, Møller, and Scheutz 2016). Additional pretreatment options can be evaluated to increase the digest value and specially avoid heavy metal contamination for its use as nutrient source without risk. Previous work on biowaste pretreatment options detailed three technologies, namely “biopulp”, “screw press” and “disk screen” to reduce impurities in the digestate. Among them, the most environmentally favorable seem to be the "biopulp" technology, which allows for increasing the digestion value with greater biogas production as well as nutrient recovery (Khoshnevisan et al. 2018). Although biogas digestate is a very attractive option to provide readily available recovered nutrients for UA the need to enforce source separation or pretreatment technologies is necessary to avoid potential contamination into the food production system (Kjerstadius et al. 2017; Davidsson et al. 2017).

While solid biogas digestate can be applied as nutrient source in media-bed hydroponics, work has also been done in the application of liquid digestate in hydroponics using the nutrient film technique (Weidner and Yang 2020; Martin, Poulikidou, and Molin 2019). These studies have shown the potential of the organic fertilization based on biogas digestate, but urge for a better control of nutrients for a balanced fertilization as well as heavy metal content (Bergstrand, Asp, and Hultberg 2020; Ezziddine, Liltved, and Seljåsen 2021; W. Wang and Lee 2021).

3.3.5 Composting/ co-composting

A classic and commonly practiced nutrient recovery from household biomass is the process of composting, which has been widely used not only in larger scale with municipal green and organic waste but on smaller scales in neighborhoods and even private gardens (Shrestha, Small, and Kay 2020; Dsouza et al. 2021; Ulm et al. 2019). Not only urban green and food waste can be destined to composting sites but also anaerobic digest as well as sewage sludge can be composted, as it commonly happens with 60-90% of sewage sludge produced in UK, Ireland, Spain, France, or Luxemburg (Bastida et al. 2019).

The composting process can be divided into three stages, an initial mesophilic stage, then a thermophilic phase and finally a maturation stage. The names of the stages correspond to the temperatures reached in the compost pile and therefore the corresponding bacteria and fungi that are active in each phase (Ravindran et al. 2021). The decomposition of organic waste mostly occurs during the thermophilic phase, where oxygen is used by microorganisms and carbon dioxide and ammonia are released. This phase is also crucial for good compost quality since the high temperatures reached in this phase enable the elimination of potential pathogens (Babu, Prieto Veramendi, and Rene 2021).

In urban areas, compost has not only been regarded to recover nutrients but also a method for urban soil remediation (Heyman et al. 2019; Kranz et al. 2020; Schwartz et al. 2017). In some cities the use of compost generated in urban and peri urban areas is mostly used for landscaping inside the urban area but only a small fraction is destined for agricultural purposes (Eldridge, Yin, and Nerida 2018). Although composting might be the most common way to treat biomass, food waste and sewage sludge for nutrient recovery, it also presents some downsides. During the process of composting a loss of N can occur due to ammonia volatilization and even emission in the form of N_2O and N_2. Usually a good P mineralization can be observed generating low N:P ratios, although this also results in the leaching of P with greater compost applications (Shrestha, Small, and Kay 2020; Zabaleta and Rodic 2015; Small et al. 2019; Wielemaker et al. 2019). Further N and P losses may be also experienced when inappropriate management is provided, e.g., when the compost pile features too much moisture, high aeration, alkaline pH and low C/N ratio (Jiang et al. 2011; Tojo 2020). To achieve a good compost a long process is needed, ensuring the elimination of potential pathogens and nutrient availability. This last part might be crucial since the mineralization process of N through composting can be very slow (Zabaleta and Rodic 2015). While traditional composting can take long periods of time, in-vessel composting systems can be a good way to shorten composting periods while also having an overall better control over the composting conditions (Ravindran et al. 2021).

The closing of nutrient loops through composting has been regarded as a great potential, Dsouza *et al.*, (2021) even proposes a direct benefit from compost and plant production with CO_2 enrichment from compost exhaust, compost itself and leached nutrients. Through the addition of ammendments like bulking agents or other urban surced materials for co-composting, it is possible to reach optimal pH, particle size, moisture and C/N ratios as well as serving as biofilters for potential GHG emissions (Dsouza et al. 2021; Asquer et al. 2019; Kaudal and Weatherley 2018; Ravindran et al. 2021; Awasthi et al. 2020).

The final compost composition and quality is also highly dependant on the incoming feed and great differences can be seen between sources, and mechanical separation of organic waste from green wastes that can origin in urban settings is crucial, specially with reference to the composition and impurities content, as well as heavy metal concentrations (Smith 2009).

3.3.6 Vermicomposting

To increase and stabilize the process of composting, the use of earthworms can be encouraged. The compost derived, also called vermicompost, is the bio oxidation and stabilization of organic matter by earthworms and other microorganisms (Suthar 2007). For this process some earthworms have been identified as detritus feeder. Some examples of the earthworms most characterized in organic waste recycling are *Perionyx excavates* (Perrier), *Eisenia fetida* (Savigny) and *Eudrilus eugeniae* (Kingberg) (Suthar 2007; Gupta and Garg 2009; Biruntha et al. 2020; Pattnaik and Reddy 2010). These species have also been categorized as fast debris feeders and are capable of reducing hazardous waste material (Ahadi et al. 2020; Ravindran et al. 2021).

Although it is a very ecofriendly and mostly cost-free addition to the composting it can entail some different management skills. While the production of compost can entail 6 to 9 months the process of vermicompost can be much faster ranging between 28 to 125 days. Its end product can be more homogenous than thermophilic compost and its nutrient content also enhanced. (Schröder et al. 2021; Ravindran et al. 2021). Previous studies on the vermicomposting of sewage sludge and green waste have reposted an increase of nutrient availability and therefore greater yield production as well as the content of humic substances and plant growth promoting hormones (Tognetti et al. 2005; Biruntha et al. 2020; Hanc and Pliva 2013; Ravindran et al. 2021). On the other hand, the production of vermicompost can need greater monitoring and skill for its production, maintaining certain conditions to ensure good living conditions for the earthworms. The specifications and conditions that have to be maintained are an initial C/N range below 40:1, a temperature range of 18-67ºC, pH range of 5.9-8.3 and a around 10% of moisture content (M. Ahmed et al. 2019; Stewart-Wade 2020). To achieve these condition a previous composting phase is often encouraged (Ravindran et al. 2021). Lower temperatures than the ones achieved in the thermolysis stage in the composting process can entail a reduced effectiveness to ensure a pathogen free final product (Tognetti et al. 2005; Biruntha et al. 2020; Hanc and Pliva 2013).

3.3.7 Compost tea

Compost and Vermicompost tea, originated during the composting and vermicomposting processes or through the addition of water, have also been considered important nutrient sources and have been used in hydroponic production systems, showing promising results in the nutritional content within the plants, although yield could be compromised (Pérez et al. 2012; Preciado-Rangel et al. 2015; Santiago-López et al. 2016; García-Villela et al. 2020). The compost tea quality greatly depends on the compost composition and feed origin, being compost tea from municipal waste a great source of necessary nutrients for plant growth and applicable into hydroponic systems through the irrigation system. The increase of bacterial activity through the application of compost tea can also increase the pathogenic suppression in the plant substrate (Stewart-Wade 2020).

3.3.8 Biological treatment (Photobioreactor)

An alternative biological treatment to the one commonly seen in WWTP is the nutrient caption through algal biomass growth (Tuantet et al. 2019; Nagarajan et al. 2020). This process is regarded as a better solution for wastewater treatment than secondary activated sludge or even secondary nutrient removal technologies (Munasinghe-Arachchige et al. 2020; Mennaa, Arbib, and Perales 2019). This is due to its cost-efficiency for pathogen, BOD, N and P removal, and the generation of oxygen for organic N breakdown through photosynthesis, and therefore avoiding costly aeration mechanisms (Mennaa, Arbib, and Perales 2019). The resulting biomass can then be used in several ways, from biomass for biofuel production, for fertilization purposes and even as animal feedstock, being a more circular approach to N dissipating technologies used in nitrifying and denitrifying processes (Nagarajan et al. 2020). The algal production using wastewater as nutrient source can be performed in open air ponds with natural light conditions or in photobioreactors, with or without continuous illumination to further enhance the algal growth. Open air production can be less costly but greatly subjected to natural temperature and light conditions, while exterior or indoor photobioreactors can provide more stable environments throughout the year (Nagarajan et al. 2020). Work has been done on the combination of WWTP processes with algal production, using different wastewater stages as potential feedstock for this biomass growth, from untreated wastewater, primary clarified, anaerobically digested, tertiary treated wastewater to even source separated urine (Samorì et al. 2013; Nagarajan et al. 2020; Tuantet et al. 2019; Zhang et al. 2014). The removal rate for N and P varies depending on the growth conditions, previous work has reported recovery rates up to 52% and 38% in N and P respectively through microalgal growth (Chatterjee et al. 2019) but higher rates can be reached in photobioreactors with up to 80% of nitrogen and total P removal in urine (Tuantet et al. 2019; Zhang et al. 2014) or even higher N removal levels depending on the wastewater and algal species (Samorì et al. 2013; Nagarajan et al. 2020; Mennaa, Arbib, and Perales 2019). Even with these extensive positive traits of algal production for nutrient removal, the application of this technology as a sole large scale wastewater treatment is still not effective. Potential unsuccessful removal of toxins as well as bacterial contamination make a previous wastewater sterilization necessary, which majorly increase the treatment costs, placing photobioreactors as tertiary treatment stages. On the other hand bacterial contamination can be avoided with the use of extremophile microalgal species or with the coculture of beneficial or symbiotic bacterial cultures (Rashid, Selvaratnam, and Park 2019), making also organic matter removal possible (Robles et al. 2020b). Finally, the algal biomass sampling can also be crucial, adding an additional step and cost to the nutrient removal. This sampling can be made through centrifugation, filtration or chemical precipitation which can be expensive and energy consuming (Mennaa, Arbib, and Perales 2019; Robles et al. 2020b).

3.3.9 Thermal treatment

Waste thermal treatment like Incineration, pyrolysis, or hydrothermal carbonization (HTC) are processes that can be used with all kinds of organic residue, entailing the use of high temperature and pressure to produce ashes on incineration processes, named biochars in the case of pyrolysis or hydrochars like in the case of HTC (Möller et al. 2018). While incineration also produces ashes with inorganic P (Kirchmann et al. 2017; Hartmann et al. 2020; Fang et al. 2020), this process requires high energy inputs while generating high carbon losses. However, incineration is a common practice in waste disposal being able to recovery energy and P from this process (J. S. Li et al. 2020) like in the case of Sweden for the municipal waste (M. Ahmed et al. 2019).

The principle of biochar production is the use of pyrolysis to break down and reorder the minerals and substances in organic biomass, and while other elements disperse during the process, P remains retained. The P retention in the outcoming biochar depends on the retention time as well as temperature with greater retention at 450ºC to 600ºC (Sun et al. 2018b). Other processes to produce biochar can be made with lower temperature requirements around 440-500ºC (Low temperature pyrolysis) or even 180-250ºC like in the case of HTC (Sun et al. 2018b; Dutta et al. 2021). To generate biochars with great nutrient content it is important to give a high nutrient containing feedstock. The potential of Municipal organic waste and sewage sludge has been studied, finding high concentrations of P(Sun et al. 2018b). The high temperatures achieved during pyrolysis are favorable for the combustion of pathogens also being able to immobilize heavy metals with the right management and processes (Sun et al. 2018b; Xia et al. 2020). These characteristics make thermal combustion a suitable process to be applied in WWTP (Zheng et al. 2020; H. Wang et al. 2020), where sludges with high P content can be dried and combusted. In HTC processes on the other hand no previous drying is required being potentially more energetically efficient (Dutta et al. 2021). Food waste and food waste digestate contains great moisture, being a good candidate for HTC. The hydrochar is produced alongside a nutrient rich process water which can be further reused for fertilization purposes (Zabaniotou and Stamou 2020; H. Wang et al. 2020; Dutta et al. 2021; Zheng et al. 2020).The slow P release of the biochar and hydrochars product makes it a favorable fertilizer that could avoid further P losses into the soil and water bodies like in the case of commercial fertilizers or manure (Sun et al. 2018b; Möller et al. 2018). Further benefits from the use of biochar and hydrochar are the promotion of carbon sequestration, plant growth and the increase of microbial communities in the soil (Dutta et al. 2021; Zabaniotou and Stamou 2020; Ijaz et al. 2020).

3.3.10 Anaerobic Membrane Bioreactor - AnMBR

In recent years the waste treatment has undergone several innovations to reduce the impact of the management while obtaining cleaner water and nutrients. One of the promising solutions for wastewater management is the combination of anaerobic digestion with membrane bioreactor system. This combination is called anaerobic membrane bioreactor (AnMBR for short), and has shown promising results for the generation of high quality effluent and greater energy recovery than anaerobic treatments in WWTP (Jiménez-Benítez, J. Ferrer, *et al.*, 2020a). AnMBR has only been applied in pilot scales but previous work on this innovative treatment has shown the potential to combine wastewater as well as food and organic waste, obtaining lower environmental impacts than other anaerobic based treatments (Jiménez-Benítez, J. Ferrer, *et al.*, 2020a). The effluent quality has been reported high for the application in agriculture, since its N and P contents are elevated. While this has been formerly seen as a constraint in the AnMBR technology, it can rather be considered an opportunity for fertigation purposes (Jiménez-Benítez, F. J. Ferrer, *et al.*, 2020a; Jiménez-Benítez, J. Ferrer, *et al.*, 2020b). The study by Jiménez-Benítez, J. Ferrer, *et al.* shows the capacity of AnMBR technology in wastewater treatment to reduce the nutrient discharge into water bodies if applied in agriculture, reducing 71% and 39% of N and P mineral fertilizer application respectively.

3.3.11 Source separated urine

Human urine has been generally considered as highly suitable for fertilizer production due to its high N and P content, being the greatest nutrient contributor in wastewater (80%, 50% and 55% for N, P and K respectively) while being only 1% of the fraction found in the total wastewater (Chatterjee et al. 2019; Volpin et al. 2018).

To increase waste treatment efficiency, the separation of waste streams in domestic level has been suggested to reduce separation and nutrient recovery processes. This idea for source separation of household waste streams has been already regarded as an upcoming reality in some countries like China and Sweden, enabling the definition of new nutrient recycling regulations (Kjerstadius et al. 2017; Pimentel-Rodrigues and Siva-Afonso 2019). To date, these same countries have already established separation technologies with urine diverting toilets for a better management and fertilizer production (Pimentel-Rodrigues and Siva-Afonso 2019). Other ways to increase circularity would not only be the separation in households but the direct application of the urine in building green roofs, rooftop agriculture or green facades, directly avoiding nutrient loss due to storage and transport (Pimentel-Rodrigues and Siva-Afonso 2019).

However, the application of human urine in agriculture has been performed in the past but has shown several constraints in present times, having greater N concentration compared to other nutrients, due to the potential content of chemicals and pharmaceuticals as well as due to the general negative perception of human urine application by producers and consumers (Ikeda and Tan 1998; Simha et al. 2017; Volpin et al. 2018; Simha and Ganesapillai 2017).To avoid these constraints several technologies have been applied to enhance nutrient recovery while reducing the content of potential impurities in urine (Calabria, Lens, and Yeh 2019; Rodrigues et al. 2019; Rufí-Salís et al. 2020; Guaya et al. 2018). These same technologies can also be applied to WWTP and anaerobic digestion effluents to further remove nutrient content for a better water recovery or disposal into water bodies, as further explained in the upcoming sections. The processes that have been most explored consist of nutrient precipitation, obtaining mineral fertilizer like struvite or urine concentration to enhance nutrient removal and serve as liquid fertilizer (Yang et al. 2015). Other technologies can entail membrane filtration or reverse osmosis, ammonia stripping and adsorption through ion exchange resins or sorbents (Volpin et al. 2018). Source separated urine can also serve as feedstock for other recovery technologies like photobioreactors, being used as nutrient source for algal growth to ensure nutrient recovery.

3.3.12 Struvite precipitation

The process of P precipitation has been largely studied and considered a valuable approach to recover P, N and Mg from wastewater and human urine. The precipitation occurs when the struvite components Mg^{2+}:NH_4^+:PO_4^{3-} are present with a molar ratio of 1:1:1 and a pH value around 8.5-9.5 (de Kraker et al. 2019; Uysal et al. 2014). The amount of Mg in wastewater and urine is usually insufficient to ensure a total precipitation and therefore is usually added, although other precipitation techniques have been developed with the addition of sea water (Shaddel et al. 2020). The precipitated crystalline mineral with the composition $MgNH_4PO_4{\cdot}6H_2O$ is called Struvite or magnesium ammonia phosphate (MAP) (Simha and Ganesapillai 2017) and has been considered a valuable slow release fertilizer due to its low solubility, being also generally regarded as a pollutant and heavy metal free crystal (de Kraker et al. 2019). The precipitation of P has been endorsed in WWTP installations due to the great recovery capacity of P reaching up to 90% or even a complete recovery under the right conditions (Volpin et al. 2018; Simha and Ganesapillai 2017) while also recovering N in smaller proportions. This process can be added as a treatment for primary or secondary effluent as well as source separated urine. The addition of struvite precipitation in WWTP with enhanced biological phosphorous removal treatment in place does not entail great modifications (Rufí-Salís et al. 2020), being a further source of nutrient recovery and direct application in soil and hydroponic agriculture (Arcas-Pilz, Rufí-salís, et al. 2021; Carreras-sempere et al. 2021; Y. H. Liu et al. 2011).

3.3.13 Ammonia stripping

Ammonia stripping is an easy process that has been incorporated in wastewater treatment plants for ammonia remediation, favoring the formation of gaseous ammonia (NH_3) through an increase of pH, which is usually made with the addition of lime (Kinidi et al. 2018). While high concentrations of ammonia can be toxic for bacteria and therefore not recommended for biological treatment, ammonia stripping has a high removal success of up to 90%, being tested already in municipal waste, landfill leachate and wastewater effluent (Kinidi et al. 2018; Zangeneh et al. 2021). The recovery and further use of the stripped ammonia gas can be achieved through adsorption to acid, obtaining ammonium sulphate fertilizer (Lorick et al. 2020). Other ways of ammonia stripping have been developed through the years to avoid high energy and chemical use for this process, combining ammonia stripping processes to electrodialysis and membrane stripping (Volpin et al. 2018).

3.3.14 Ion exchange / adsorption

The processes of NH_4-N and PO_4-P caption through ion exchange or adsorption have been studied and used for nutrient removal in wastewater treatment, using mainly natural or synthetic zeolites for N and metal-loaded chelating resins, iron-based hydroxide compounds and hydrotalcites for P as captor or exchange surface (Williams 2013; Kuntke, Schaetzle, and Loos 2016). The ion exchange process is a simple exchange between the wastewater flow and an exchange material containing column, and while NH_4-N or PO_4-P is attached to the media column, other cations are released to the wastewater (Williams 2013). This method shows a great recovery (more than 95%) for both N and P, and can then be reversed with salt water which can be then precipitated as struvite to be used as fertilizer (Lohman et al. 2020; Volpin et al. 2018; Mullen et al. 2019).
The adsorption process of adsorption/desorption follows the same principle of nutrient caption using selective sorbents which can be then applied as amendment in agricultural substrates (Guaya et al. 2018; Simha and Ganesapillai 2017).

3.3.15 Membrane filtration

Membrane based separation methods for nutrient recovery entail a great variety of filtration methods like nano- micro- and ultrafiltration (NF, MF, UF) which are usually followed by reverse osmosis (RO) in treated wastewater as well as in anaerobic digestion liquid fraction (Gienau et al. 2018a). The use of MF and UF is common in WWTP and further application of NF and RO can be made to obtain non-potable clean water (Hube et al. 2020). NF and RO are pressure-driven filtration processes that can have high overall costs as well as more operational complications compared to other techniques previously explained like ammonia stripping (Volpin et al. 2018; Gienau et al. 2018a). These membrane filtration based processes like NF and RO can also lead to urea and ammonia losses that can lead to a poor N recovery (Volpin et al. 2018; Simha and Ganesapillai 2017).

3.4 Case study of nutrient recovery application: exploring the potential of NPK recovery in the AMB

Barcelona is a densely populated city in the Mediterranean area with 16’420 inhabitants km^{-2}, with a limited land availability that has prompted the inclusion of agricultural activities inside the urban area, specially focusing on rooftop agriculture systems (Zambrano-Prado, Orsini, et al. 2021; Appolloni et al. 2021). This interest has originated extensive research and educational activities from university research institutes (ICTA-UAB, UPC) and organizations (Replantem) focusing on the integration and application of agriculture in the city of Barcelona. The city council has also promoted these activities with the creation of 6 hydroponic installations on rooftop buildings for social and community integration purposes (IMPD project)(Biel 2019) and hosts a green roof contest annually to endorse projects that propose the creation of rooftop gardens (Ajuntament de Barcelona 2020). The objective is the creation of 34’100 m^2 of green roofs by 2030 (Zambrano-Prado, Orsini, et al. 2021).

The potential of the Metropolitan area of Barcelona to implement rooftop open air agriculture as well as rooftop greenhouses has already been identified for several urban and peri urban areas. Such work has been developed with the help of GIS Rooftop databases and remote sensing approaches mainly focusing on larger roof extension to host these installations, being industrial and retail parks, as well as large social housing neighborhoods the best candidates (figure 3.2) (Toboso-Chavero et al. 2019a; Zambrano-Prado, Muñoz-Liesa, et al. 2021; Nadal et al. 2017; 2018; Toboso-Chavero, Villalba, et al. 2021; Sanyé-Mengual et al. 2018; Zambrano-Prado, Pons-Gumí, et al. 2021).

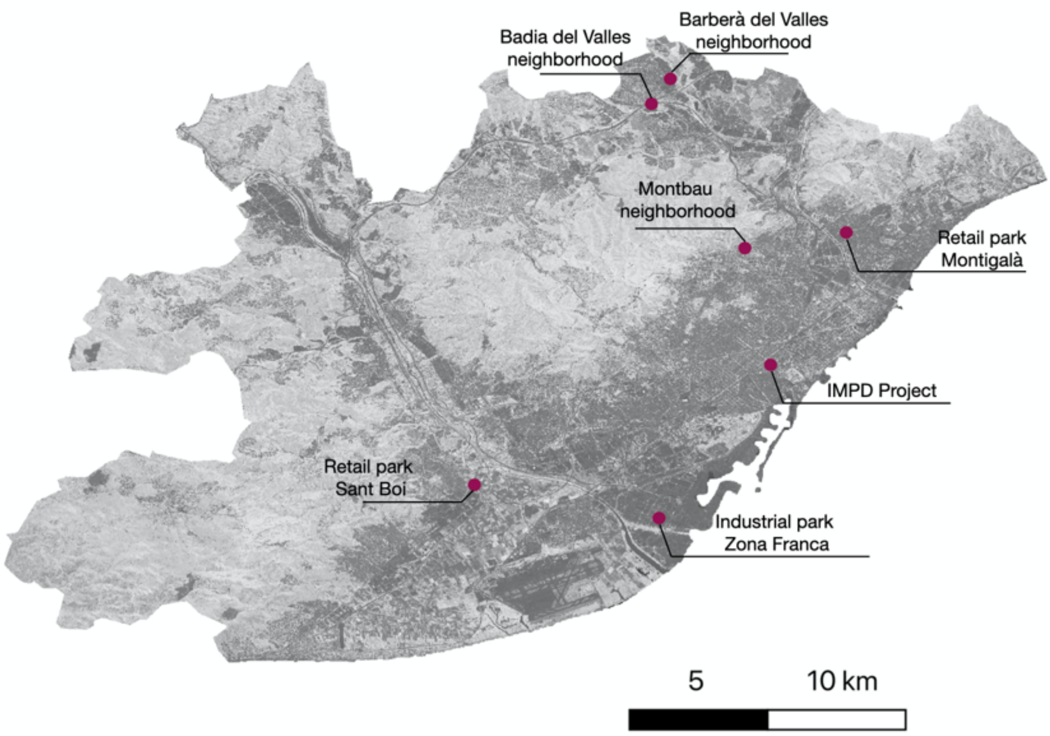

Figure 3. 2 Map of the Metropolitan Area of Barcelona with identified locations for rooftop agriculture.

While several areas have been identified as potential UA sites due to the rooftop material as well as rooftop extension, these have only been contemplated in a theoretical way. However other rooftop UA areas have been implemented, mainly through the social project “Horts al terrat” from the municipality of Barcelona and the Green roof competition. The total potential identified in literature was 44.44 ha and the area of already existing sites was 0.77 ha making a total of 45.21 ha (452’100 m^2) within the metropolitan area of Barcelona (Table 3.4). Other typologies of UA like indoor or soil based were not included.

If this area is dedicated to tomato production with an estimated productivity of up to 16.5kg m^{-2} year $^{-1}$ (Sanyé-Mengual et al. 2018) the potential production could entail up to 7459 t of tomato which equals a 13.3% of the tomato consumption within the metropolitan area (Table A3.1).

Table 3. 4 Areas identified within the metropolitan area of Barcelona for UA on rooftops.

	Area/ Project name	Building type	Area	Comment	Reference
Potential areas found in literature	Zona Franca	Industrial Park	13.06 ha	Around 14% of tomato imported (128000 people demand year^{-1})	(Sanyé Mengual 2015)
	Sant Boi	Retail Park	5.58 ha	Urban self-supply 3.8%	Sanyé-Mengual *et al.*, 2018
	Montigalà	Retail Park	5.22 ha	Urban self-supply 3.5%	Sanyé-Mengual *et al.*, 2018
	Montbau	Neighborhood	0.06ha	Up to 37% of tomato self-supply	(Toboso-Chavero et al. 2019a)
	Badia del Vallés Barberà del Vallés	Neighborhood Neighborhood	20.52 ha	Self-sufficiency for tomato 210% and lettuce 21% in the neighborhood	(Zambrano-Prado, Muñoz-Liesa, et al. 2021)
Implemented areas	IMPD Project "Hort al terrat"	Municipality Buildings	0.02 ha	3590kg year^{-1} of vegetables	(Biel 2019) (Ajuntament de Barcelona 2021)
	Green Roof Competition	Private rooftops	0.75 ha	Vegetable and urban green	(Ajuntament de Barcelona 2020)

3.4.1 Organic waste generation and treatment

While a waste recovery system is generally implemented with classified sorting bins, only around 36% of the waste is recovered separately with a greater fraction found under "rest" or "unclassified" waste. The current goal is the increase of the separated fraction up to 50% in all municipalities, which was achieved by only the 16% of all municipalities by 2018. The generation of separated organic waste in the metropolitan area can be divided in three categories: Household waste, organic waste from big producers and finally green waste. The total organic waste collected from the classified sorting bin in 2020 was 184'000 tonnes of which 78% was household waste while only 6% and 16% were originated from big producers and green waste, respectively. This differentiated waste is then transported to two specialized installations, Ecoparcs (1, 2 and 4) and composting sites. The handling of this waste fraction is similar in all Ecoparc installations, with a pretreatment to prevent impurities and the mixing of the three organic waste categories for its digestion and further production of compost. The composting sites on the other hand don't entail an anaerobic digestion step to produce compost. Although the waste is collected in separated bins, the percentage of impurities is still high depending on the municipality of origin. The Ecoparcs 1, 2 and 4 presented impurity percentage ranges of 8.7-23%, 5.4-30.3% and 1.7-36.6% respectively while the composting sites showed ranges of 2.5-19.2%.

The location of the Ecoparcs and composting sites can be seen in figure 3.3 , mostly located within the metropolitan area of Barcelona with only the exception of the Ecoparc 4. The destination of the collected organic waste from the differentiated bins in 2020 was mostly in the Ecoparc 2 with up to 44% of the generated waste while Ecoparcs 1, 4 and the composting sites received 32%, 18% and 7% of the generated waste respectively. The yearly production of compost almost reaches 30’000 tons, which can be further used in gardens and surrounding agriculture. The “rest” or “unclassified” fraction produced in 2020 was more than 800’000 tons and it is also processed in the Ecoparcs 1, 2, 3 and 4, with a distribution of 21%, 23%, 23% and 33% of the total respectively. The process undergone for this fraction is mechanic and biologic treatments and biowaste stabilization for the annual production of more than 70’000 tons of stabilized biowaste for soil amendment and landfilling.

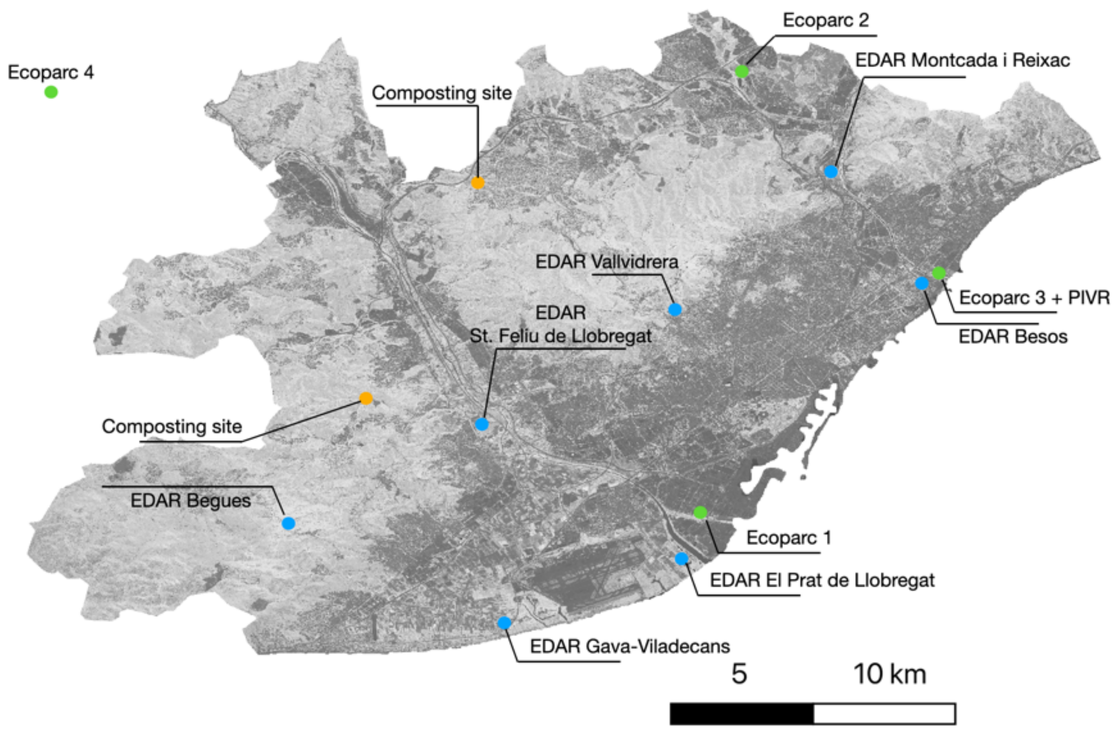

Figure 3. 3 Map of the metropolitan area of Barcelona with the location of the currently active WWTP’s (blue) and Ecoparcs (green) and composting sites (orange).

3.4.2 Wastewater generation and treatment

In the metropolitan area of Barcelona, we can find 7 WWTPs (Figure 3.3) that are responsible for the treatment of around 270’000 Mm^3 of wastewater each year. The technologies between all plants vary for both sludge and effluent treatments, between sludge anaerobic digestion and composting and sludge dewatering and field application, to secondary or tertiary water treatments (Table 3.5).

Table 3. 5 Description of the WWTP's found in the metropolitan area of Barcelona, yearly treated amount and percentage in relation to the total wastewater treated in the area. Estimation of incoming N and P based on (You, Valderrama, and Cortina 2019); CAnD – Comp

	WWTP	Incoming flow (Mm^3 y^{-1}) (2019)	% from Total	Sludge treatment	Effluent treatment	Incoming N (ton y^{-1})	Incoming P (ton y^{-1})
1	El Prat de Llobregat	92.1	36%	CAnD	Secondary and Tertiary treatment	5525 t	626 t
2	Besòs	120.4	45%	DW	Secondary treatment	7225 t	819 t
3	St. Feliu de Llobregat	18.5	7%	CAnD	Secondary and Tertiary treatment	1114 t	126 t
4	Montcada I Reixac	18.8	6%	-	Secondary treatment	1129 t	128 t
5	Gavà I Viladecans	14.8	5%	CAnD	Secondary and second decanter	892 t	101 t
6	Begues	0.3	0.7%	-	Secondary treatment	21 t	2 t
7	Vallvidrera	0.2	0.5%	DW	AnMBR	15 t	2 t

The yearly production of sludge is around 57'000 ton of dry matter (2020) which then is directly used in agriculture (24%) or composted (68%). The water treatment in the WWTP from El Prat de Llobregat follows five main steps, starting with a pretreatment for solid and oils separation, a primary treatment where the sludge is removed, a secondary treatment with nitrification and denitrification processes for nitrogen removal, tertiary treatment and denitrification processes and ultrafiltration and reverse osmosis to retrieve and regenerate water. Apart from the WWTP in Gavà I Viladecans no other WWTP's has water regenerating processes.

In conventional WWTP's about 30% of the influent wastewater nutrients are removed through active sludge separation in the primary treatment, while further nitrification and denitrification processes in the secondary water treatment can reach a removal of up to 70%. This nutrient removal is applied in 4 WWTP in the metropolitan area, namely the WWTP in el Prat de Llobregat, in St. Feliu de Llobregat, in Gavà I Viladecans and Begues. Although this can be considered a good removal rate, approximately 1200 t of N and 160 t of P are still released every year into the Mediterranean Sea only considering the WWTP in El Prat de Llobregat. The potential therefore for additional nutrient removal is great.

Work on the reduction of P and N in the wastewater effluent in El Prat de Llobregat and Besòs WWTP's has already been done, proposing struvite precipitation or zeolite adsorption. These works consider the recovery of these nutrients a success, obtaining 5000 t $year^{-1}$ of loaded zeolite with a 15% of PO_4^{3-} content and a range of 43 – 368 t $year^{-1}$ of P in the form of struvite in the El Prat de Llobregat WWTP (depending on the recovery technology).

3.4.3 Nutrient recovery potential in the Metropolitan Area of Barcelona

Taking in account all previous information collected, an estimate of the potential of nutrient recovery with the existing infrastructure of the AMB can be determined (Table 3.6, Table A3.2, Table A3.3). This estimation is again based on the assumption that all the defined area is dedicated for tomato production.

The existing generation of organic waste and the produced compost and digest in the Ecoparcs and composting sites can produce up to 550 t of N-Nitrogen and 170 t of P-Phosphorous each year, meeting around 48 and 60 times the N and P demand respectively.

The sludge compost generation in all WWTP can be a great source of nutrient, especially P, with a yearly production of more than a 1000 t. This can cover 117 and 380 times the UA requirements of N and P, respectively.

The generation of struvite can vary between recovery technologies, being defined by Rufí-Salís, Brunnhofer, *et al.*, 2020, as ranging from lowest recovery of 7% of P for incoming wastewater P in the AirPrex technology, to 60% of P recovery for incoming P in the RemNut technology. In this case only the WWTP in El Prat de Llobregat is considered due to the existing necessary installation. The generation of struvite only in this plant can meet the N demand 1.7 to 14.5 times and the P demand by 14.6 to 125.2 times.

Finally the work of You, Valderrama and Cortina, 2019 encourages the possibility of zeolite absorption processes in the El Prat de Llobregat WWTP due to the existing filtration installations. A yearly recovery of 5’000 t of loaded zeolite is defined with approximate content of 750 t of P, meeting the P demand by 255 times.

*Table 3. 6 OM= Organic Material, WWS= wastewater sludge, WWE= wastewater effluent, Min= minimum removal 7% P with AirPrex technology, Max= maximum removal 60% with RemNut Technology, * obtained from Rufí-Salís, Brunnhofer, et al., 2020, **obtained from You, Valder*

Waste type	Amount (Ton DW/year) (2020)	N content in waste (kg)	P content in waste (kg)	Times N demand is met	Times P demand is met
OM Compost	987.7	21235	7802	1.8	2.7
OM Digest	19250	525525	171325	46	58
WWS- Compost	57000	1328100	1117200	117.5	380.2
WWE - Struvite Min*	341.3	19829	43000	1.7	14.6
WWE - Struvite Max*	2920.6	169962	373000	14.5	125.2
WWE – Zeolite**	5000	-	750000	-	255.2

Although more technologies could be applied to the nutrient cycle in this area, it would be necessary to further install more infrastructure which can in turn also generate additional environmental impacts to the nutrient recovery systems (Rufí-Salís, Brunnhofer, *et al.*, 2020). On the other hand, the potential of struvite as slow-release fertilizer and the generally positive characteristics to be used in UA due to the lack of smell, low content of heavy metals and potential pathogens, begs for the question of the production that could be achieved in all WWTP (Table A3.4). If considering all WWTP had the capacity for struvite precipitation between the ranges of 7 to 60% from the incoming P. It emerges that a potential production of 126 to 1082 t of P and 52 to 489 t of N could be recovered yearly.

3.5 Constraints and obstacles to fulfilling the nutrient recovery potential

The capabilities to recycle and reuse nutrients like N and P are clear, but why are there no more advances in the application of these processes? The use of organic waste from urban areas are generally perceived as having a bad quality or containing great amounts of unwanted elements that could be toxic or polluting (Gímenez Lorang, Soliva I Torrentó, and Huerta 2005). This is also true for sewage sludge quality, which was found to contain great amounts of potentially toxic elements (PTEs) in long-term experiments that could be traced back to be generated in World War II (Möller et al. 2018). On the other hand, the quality of organic waste and sewage sludge has been increasing for the past decade making it a great nutrient resource. Still, due to public or private legislation standards most of these recovery sources are not permitted, e.g., in organic farming (Möller et al. 2018; Awasthi et al. 2020). Furthermore, the production costs of recovered P can be a constrain to implement these processes if compared to the already existing extraction chain of mined phosphate rock (Oarga-Mulec et al. 2019; Chojnacka, Moustakas, and Witek-Krowiak 2020). Therefore most of the emerging recovery technologies remain on laboratory or pilot scale with limited market uptake (Cordell, Brownlie, and Esham 2021). Although the application of recovery technologies is still not fully considered in waste treatment processes this state of mind is slowly changing and pushed towards recovery, in response to the foreseen P shortages in the coming years as well as the environmental impact of untreated waste. An increase of bio-waste derived fertilizers is expected and estimated to replace up to a 30% of the inorganic fertilizers (Chojnacka, Moustakas, and Witek-Krowiak 2020), and up to 50-60% of phosphate rock imported into Europe and used in agriculture (Möller et al. 2018).

3.6 Conclusions

The increase of nutrient circularity is a pressing matter in recent years, and while mainly associated to agriculture and rural areas the loss of nutrients is also a reality in cities. The present study has identified the literature concerning nutrient recovery technologies from urban waste flows which can be further repurposed in UA.

Three main conclusions can be drawn from this literature search.

Firstly, two main waste types were identified as most regarded in literature, namely organic-, bio-, food waste and wastewater. Under these two umbrellas 5 recovery strategies could be accounted for organic- bio- and food waste and 11 for wastewater.

Secondly, it can be concluded that the yearly production amount of both waste types can fulfill N and P requirements for UA in the metropolitan area of Barcelona in ranges of 2,7 to 380,2 and 1,7 to 117,5 times the necessary amount for P and N respectively depending on the recovery strategy.

Thirdly, the promising results for many recovery strategies are punt into a hold or left on laboratory scale due to current perceptions and legislations which don't facilitate their application and nutrient repurpose for agriculture. On the other hand, these perceptions could shift in the near future due to the pressing need for P recovery.

Part III

Agronomic approach to struvite use in hydroponic production

Chapter 4

This chapter is based on the journal paper:

Recovered phosphorous for a more resilient urban agriculture: Assessment of the fertilizer potential of struvite in hydroponics

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Chapter 4: Recovered phosphorous for a more resilient urban agriculture: Assessment of the fertilizer potential of struvite in hydroponics

Abstract:

Urban agriculture (UA) is a means for cities to become more resilient in terms of food sovereignty while shortening the distance between production and consumption. However, intensive soilless UA still depends on the use of fertilizers, which relies on depleting non-renewable resources such as phosphorous (P) and causes both local and global impact for its production and application. With the aim to reduce such impacts and encourage a more efficient use of nutrients, this study assesses the feasibility of using struvite precipitated from an urban wastewater treatment plant as the unique source of P fertilizer. To do so, we apply various quantities of struvite (ranging from 1 to 20 g/plant) to the substrate of a hydroponic Phaseolus vulgaris crop and determine the yield, water flows and P balances. The results show that treatments with more than 5g of struvite per plant produced a higher yield (maximum of 181.41 g/plant) than the control (134.6 g/plant) with mineral fertilizer (KPO_4H_2). On the other hand, P concentration in all plant organs was always lower when using struvite than when using chemical fertilizer. Finally, the fact that different amounts of struvite remained undissolved in all treatments denotes the importance to balance between a correct P supply to the plant and a decrease of P lost through the leachates, based on the amount of struvite and the irrigated water. The findings of this study show that it is feasible for UA to efficiently use locally recovered nutrients such as P to produce local food.

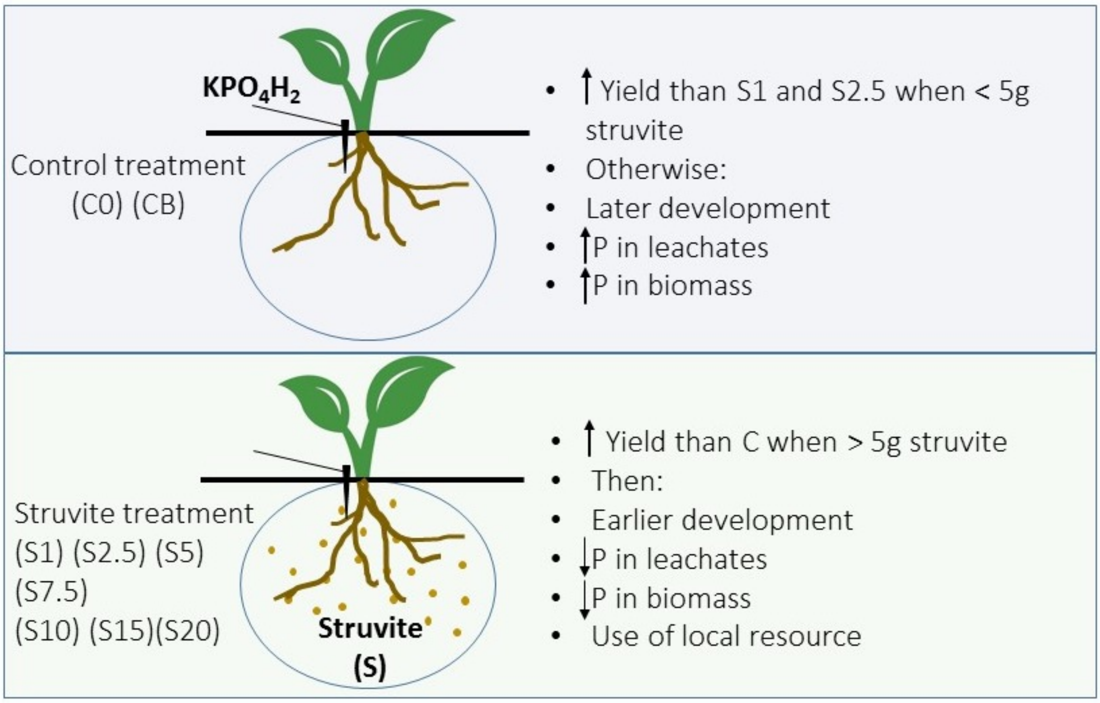
Keywords

Phosphorus, Struvite, Fertilizer substitution, Circular economy, Industrial ecology, Urban agriculture

Highlights

- Struvite is tested in hydroponic production of Phaseolus Vulgaris
- Yield, water fluxes and P balances are analysed
- More yield is produced by plants with more than 5g of struvite
- Slow release by struvite decrease the leached P
- Different factors affect the efficiency of struvite as a fertilizer

Graphical Abstract



4.1 Introduction

Meeting the food demand of the ever-growing urban population is a global challenge. Since food provision to cities is highly dependent on long and complex supply chains, the distance between production and consumption points has extensively increased. This prevents nutrient recycling, while emitting huge amounts of greenhouse gases due to long-distance transport (Thomaier et al. 2015; Rees and Wackernagel 1996). In this sense, moving towards more sustainable food systems, should be a priority in the following years (European Commission 2020). To do so, alternatives that narrow the distance between production and consumption points have already been reported, being urban agriculture one of the most prominent (Deelstra 1987). However, this implies that the resources required to produce food, mainly fertilizers and water, must now be imported to cities. In the case of water, the use of rainwater harvesting systems combined with hydroponics can help meet the irrigation requirements without compromising the yield (Astee and Kishnani 2010; Rufí-Salís, Petit-Boix, Villalba, Ercilla-Montserrat, et al. 2020). On the other hand, the use of local fertilizers is still very limited, and often reduced to the use of compost (Thomaier et al. 2015). The vertical and soilless production systems have been reported to maintain greater yields while at the same time avoiding land occupation making it a viable alternative while in some cases environmentally better than open field production (Romeo, Vea, and Thomsen 2018). On the other hand the extensive use of mineral and synthetic fertilizers is necessary, causing potential and additional environmental damage in urban ecosystems if UA continues growing without the search for alternative fertilization methods (Sanjuan-Delmás et al. 2018; Rufí-Salís, Petit-Boix, Villalba, Sanjuan-Delmás, et al. 2020b; Kwon et al. 2021).

The case of phosphorus (P) fertilizers is of great relevance, since P is primarily obtained from non-renewable phosphate rocks. Moreover, previous studies quantify that 80% of the available stock of phosphate rocks is being used in the production of fertilizers (Shu et al., 2006). Since half of the world's current economic phosphate resources will have been used up by the end of the 21st century (Steen 1998; Cordell, Drangert, and White 2009) the European Union recognizes P as a critical resource (European Comission 2014). Among its recommendations, a planned amendment of the fertilizer regulation encourages P recovery from local sources by enforcing a shift towards a more circular use of nutrients (European Comission 2016).

In this sense, urban wastewater treatment plants (WWTPs) are well-known sources of secondary P. WWTPs have already been addressed as a potential alternative to importing mineral fertilizers (e.g. de-Bashan and Bashan, 2004; Kern et al., 2008; Shu et al., 2006). Struvite, also known as magnesium ammonium phosphate (MAP with the formula $NH_4MgPO_4{\cdot}6H_2O$) is a crystalline precipitate that has been gaining popularity as a way to recover P from wastewater. To induce its precipitation a molar ratio of 1:1:1 for magnesium (Mg^{2+}), ammonium (NH_4^+) and phosphate (PO_4^{3-}) is needed, under specific pH conditions (8.5-9.5) (Le Corre et al. 2009; J. R. Buchanan, C. R. Mote, and R. B. Robinson 1994; Bouropoulos and Koutsoukos 2000). Originally the precipitation of struvite was associated to a major concern in WWTP being the cause of equipment damaging causing labor and infrastructure costs (Borgerding 1972; Doyle et al. 2003; Stratful, Scrimshaw, and Lester 2004). Struvite forced precipitation has gained attraction since the 90's, not only to avoid infrastructure damage but also as a P recovery technique (Doyle et al. 2003). This process has been studied and improved in the past years making it a more efficient precipitation process (Sena and Hicks 2018; B. Li et al. 2019; Le Corre et al. 2009). Although the production of struvite is gaining popularity, its commercial production is still scarce. The potential of P delivery of a WWTP in the form of struvite in the system where this study is located has been previously quantified by Rufí-Salís, Brunnhofer, et al., (2020), demonstrating the potential of these widespread installations to provide this ill distributed resource.

In terms of application, the properties of struvite as an effective source of nutrients (P-PO_4^{3-}, N-NH_4^+ and Mg-Mg^{2+}) for plants (X. . Li and Zhao 2003) and its low solubility in water (0.018g·100ml-1 at 25ºC) (Bridger, Salutsky, and Starostka 1961) make it a slow-releasing valuable fertilizer that can reduce economic costs in agriculture (Rahman et al. 2014). However, only limited literature has explored the application of struvite in agricultural facilities. For example, Antonini et al. (2012), Uysal et al. (2014), Gell et al. (2011) and Liu et al. (2011) assessed the maize performance of struvite with different characteristics and origins in different soils. In a review made by Li et al.

(2019) we can see that almost all struvite trials found that vegetables grown with struvite had the same -or even improved- performance compared to controls with conventional fertilizers.
Creating a closed-loop, waste-to-resource system such as that of struvite recovery within the city limits and not applying it at this scale seems contradictory within the concept of urban metabolism. In this sense, the synergy between struvite precipitation in urban WWTPs and urban agriculture seems worth exploring considering the potential of the latter to blur the lines between waste and resource within urban areas (Smit and Nasr 1992; Ferreira et al. 2018; Rufí-Salís et al. 2020). This article aims to assess the potential of struvite precipitated in a WWTP as a fertilizer within the framework of urban metabolism. Based on experimental and analytical results performed on a Phaseolus vulgaris crop grown in a hydroponic rooftop greenhouse, we determine the implications of fertilization with struvite in terms of yield, water flows and P balances and provide recommendations to further improve the performance of this waste-to-resource fertilizer.

4.2 Methodology

4.2.1 Characterization of the system

The present study was conducted in a rooftop greenhouse on the ICTA-ICP building, located in the campus of the Universitat Autònoma de Barcelona, 15km away from Barcelona. The building is equipped with a 900m^2 rainwater harvesting system that stores water in a 100m^3 tank. Most of this rainwater is used in the rooftop greenhouse (122.8m^2) to irrigate crops with a hydroponic system, i.e. mixing water with nutrients before providing the solution through a dripping system (2 L/h) to the perlite substrate bags (40L capacity). The perlite substrate has a pH of 7, an electrical conductivity of 0.09 $dS\cdot m^{-1}$, a granulometry of [0-6] mm and 4 plants can be planted in each bag.

4.2.2 Fertilization and experimental set-up

Struvite granules were obtained from Aarhusvand A/S company from Aarhus, Denmark. This company distributes fertilizer grade struvite under the name PhosphorCareTM, recovered using the PhosphogreenTM technology (Hall et al. 2020; Muys et al. 2021; Suez 2018; Chrispim, Scholz, and Nolasco 2019). This technology is based on a fluidized bed reactor that creates the specific conditions to precipitate struvite through the addition of magnesium chloride, sodium hydroxide and air. The final struvite granules have a size range of 0.5-1.5 mm.
Common bean plant (*Phaseolus vulgaris* var. Pongo) was chosen as the crop for this study, planting nursery plants (approximately 10-14 days old). To apply the struvite to the plants, we considered different possibilities. Mixing it with the nutrient solution was discarded because the

system could not benefit from the slow-release characteristics of struvite. Thus, we choose to directly apply the granules to the plant roots. Considering this option, we designed a system that consisted of mixing perlite with struvite inside a low-density polyethylene perforated bag with holes of no more than 1 mm diameter (Figure A4.1). At the same time, this system allows the interaction between struvite granules and roots and avoids the loss of undissolved struvite into the leachates due to draining through the perlite bag.

Two different experiments were carried out: the validation test and the determination test, both of them using double growing lines with 8 substrate bags each (Figures A4.2 and A4.14). The validation test served as a previous experimental set-up to determine if the proposed methodology was functional and correct possible influencing variables in the experiment, such as the use of the plastic bag to retain the struvite close to the plant rhizosphere or to scale the most suitable quantities of struvite for the determination test. On the other hand the determination test was designed with the previous experience of the validation test. For control treatments, the nutrient solution applied to the crops in milligrams per liter was KPO_4H_2 – 136, KNO_3 – 101, K_2SO_4 – 217.5, $Ca(NO_3)_2$ – 164, $CaCl_2 \cdot H_2O$ – 111, $Mg(NO_3)_2$ – 148.3, Hortilon – 10, and Sequestrene – 10. In treatments with struvite, the mineral P source, KPO_4H_2 in this case, was excluded from the initial nutrient solution. All other mineral fertilizers were maintained.

4.2.3 Phosphorus balances

To account for the P balances, Equation 1 was calculated on a plant basis for every control and struvite treatment. Figure A1 shows a diagram of the perforated bag with the elements displayed in Equation 3.

$$Eq\ 3: \qquad P_{NS} + P_{SI} = P_{LV} + P_{ST} + P_{BN} + P_{SF} + P_{LIX} + P_{AC}$$

In Equation 1, P represents mass of phosphorus. P_{NS} is the amount of mineral P supplied through the irrigation system during all the crop cycle. P_{SI} is the amount of P in the form of struvite applied at the beginning of the test. P_{LIX} is the amount of P in the leachates during all the crop cycle. P_{LV}, P_{ST}, and P_{BN}, represent P uptake by leaves, stem and beans, respectively. P_{SF} is the amount of remaining undissolved P in the form of struvite at the end of the test, plus the P adsorbed in the perlite granules. Finally, P_{AC} is the amount of dissolved P accumulated in the water retained in the substrate at the end of the crop. Three different biomass and substrate sampling dates were used in every test: 26, 54 and 78 days after planting (DAP) for the validation test and 23, 51 and 72 DAP for the determination test.

The initial nutrient concentration of the substrate was verified to be negligible at the beginning of the experiment through atomic spectroscopy and elemental analysis. Samples of the fertilizer solution were collected directly from the drippers placed in the perlite bags. Leachate samples were taken from plastic drainage buckets placed on one side of each line. To determine the P_{NS} and P_{LIX}, the respective samples were collected three times per week and externally analyzed using ICP-OES atomic spectroscopy (Optima 4300DV by Perkin-Elmer). P_{SI} was quantified summing the amount of perlite in a specific bag with the amount of struvite that was applied, considering weights obtained by drying two struvite samples and two perlite samples at 105°C in a furnace until reaching constant weight (reached after 3 days). P_{SF} was quantified differently in each test. In the validation test, all 4 samples for a specific treatment were homogenized after extracting the roots, using distilled water to separate the struvite granules from the roots. After this process, two random samples were dried at 105°C in a furnace until reaching constant weight, then grounded and digested with concentrated HNO_3 in a Single Reaction Chamber microwave and externally analyzed using ICP-OES atomic spectroscopy. On the other hand, in the determination test, roots were shredded, homogenized and integrated within every individual substrate sample. Then, a fraction of these samples was dried and analyzed using the same method as in the validation test.

P_{LV}, and P_{ST} were determined based on the nutrient content of every plant separately. Leaves and stem were separated, sorted into paper envelopes and dried in a furnace at 65°C until reaching constant weight (reached after 7 days), grounded and digested with concentrated HNO_3 in a Single Reaction Chamber microwave before analyzing externally the concentration of P through ICP-OES atomic spectroscopy. The same methodology was applied to determine the P_{BN}, with randomly chosen 500-gram bean samples being processed for every treatment. The measured P content of beans was multiplied by the measured rates of biomass production to estimate the rate of P accumulation in crop biomass.

4.2.4 Validation test set-up and justification

From September 13th until December 3rd, 2018, 10 double growing lines were used (totaling 320 plants), distributing the treatments as showed in Figure A2 of the Annex II. The aim of this experiment was to validate and keep track of different parameters of the system, like for instance, make sure that the small, perforated bag did not have negative consequences on the crop development. To do so, we split the control lines into two different treatments, VCB and VC0, using standard nutrient solution with and without the bags, respectively. Secondly, to check the correct development of bean plants with struvite in a hydroponic system, we applied different struvite amounts per plant: 5, 10, 15, 20 and 25g corresponding to the treatments tagged as V5, V10, V15, V20 and V25, respectively. Additionally, a treatment with no struvite was tagged as V0. These amounts of struvite were based on previous experiments done with the same crop species and variety in hydroponic cultivation that accounted for P uptake (Rufí-Salís, Petit-Boix, Villalba, Sanjuan-Delmás, et al. 2020b). One week after the first harvest, KPO_4H_2 was added in the nutrient solution of struvite treatments until the end of the harvest to ensure a good nutrition to the plants during the production period, which is highly demanding in P (e.g. Bender et al. 2015; Kouki et al. 2016; da Silva et al. 2019).

4.2.5 Validation test results

4.2.5.1 Production and phenological stages

The production results for the control treatments VCB and VC0 showed that the perforated bag did not have any effect on the correct crop development and yield (Figure 4.1 and Figure A4.5), as the yields from the different lines do not differ between them (VC0_2 187.54±69.35; VCB_1 186.15±84.01 g/plant). Even though treatment VC0_1 generated more yield (224.84±91.84 g/plant), it could be attributed to the fact that it was an exterior cropping line facing the border and thus received more radiation. Similarly, VCB_2 also produced more yield (195.45±88.63g/plant) than its replicate (VCB_1) although no significant differences were determined by the end of the experiment.

On the other hand, treatments with struvite (Figure A4.3 and A4.4) exerted a similar yield than the control treatments at the end of the crop. The treatment with the highest quantity of struvite (V25) had the highest production median (203.85 g/plant), while the treatment with the lowest quantity of struvite (V5) had the highest mean (216.15±93.54 g/plant). On the other hand, the treatment without struvite produced a really low yield (7.19±4.49 g/plant).

The similarities in terms of yield between all struvite treatments at the end of the cycle may be related to the addition of KPO_4H_2 fertilizer during the production phase. Moreover, we can see that struvite treatments produced more than the control in the first 3 harvests (35, 39 and 42 DAP) (Figure A4.6). This effect is similarly observed for the phenological stages (Figures 4.2 and A4.7 to A4.10). For the parameters that were quantified in different dates (number of leaves (figure. 4.2), side shoots (Fig. A4.7), open flowers (Fig. A4.8) and floral buttons (Fig. A4.9)), we can see that the treatments with struvite not only had a correct early stage development, but also develop plant organs earlier than in control treatments.

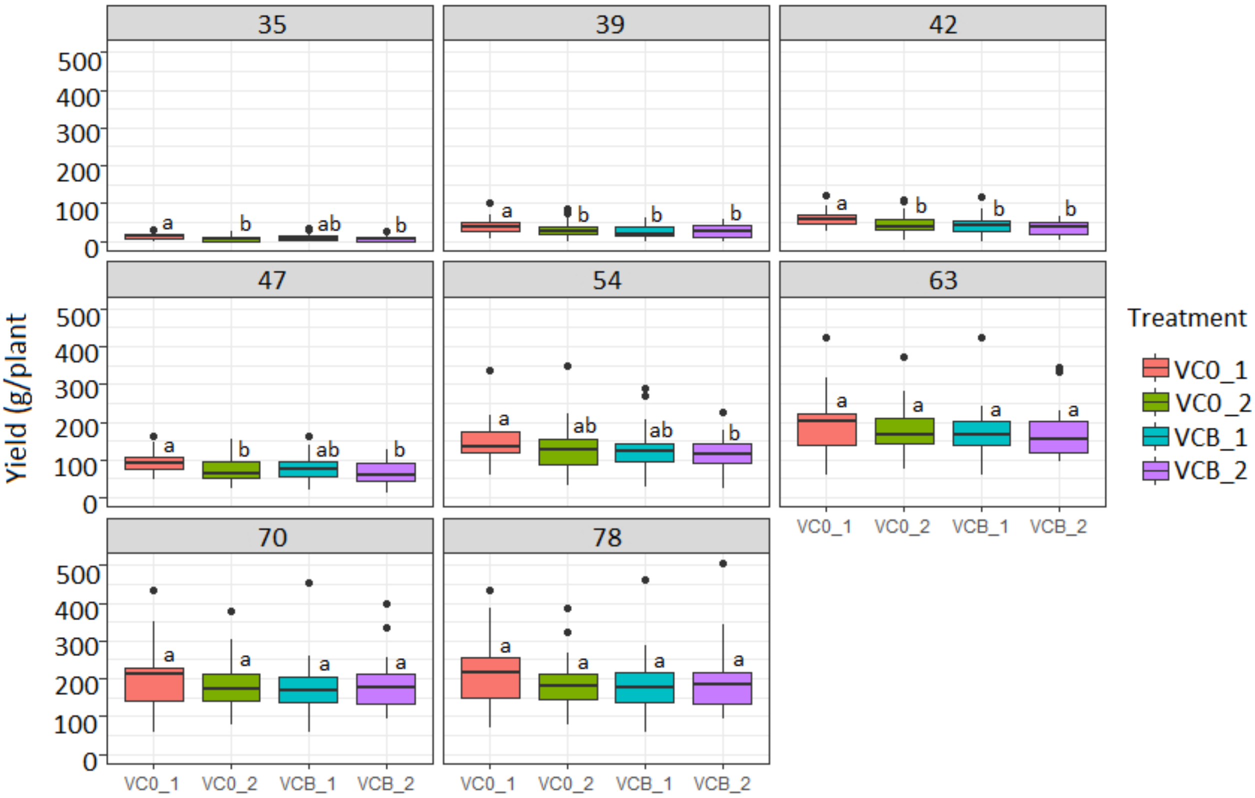


Figure 4. 1 Production (g/plant) of the control treatments in the validation test, with (VCB) and without (VC0) perforated bags for each harvest. Each panel in the figure grid represents the result for a harvesting day, with a total of 8 days (35 to 78 days after planting). Same letter (a,b) indicate no significant difference (p>0.05) between treatment for each harvest time. Sample size for harvest 1,2,3,4 and 5 (35-54 DAP) corresponds to n=28 plants, for harvests 6, 7 and 8 (63-78 DAP) n=24 plants per treatment.

4.2.5.2 Water

We applied more water in struvite treatments (125 L/plant) than to the control (94.76 L/plant) to ensure a proper dissolution of this fertilizer (Fig. A4.11). However, we can see in Figure A4.12 that if a greater amount of water flow through the perlite bag is provided through a greater irrigation, leachates emitted by the struvite treatments with higher concentrations (28.9 mg/L – V25) of this fertilizer tend to be similar to those of the control treatments. Obviously, this behaviour can only be observed before the irrigation with mineral P added during the harvesting process. Similarly, we can see that the perforated bag mechanism did not affect the P concentration in the leachates between the control treatment C0 and CB.

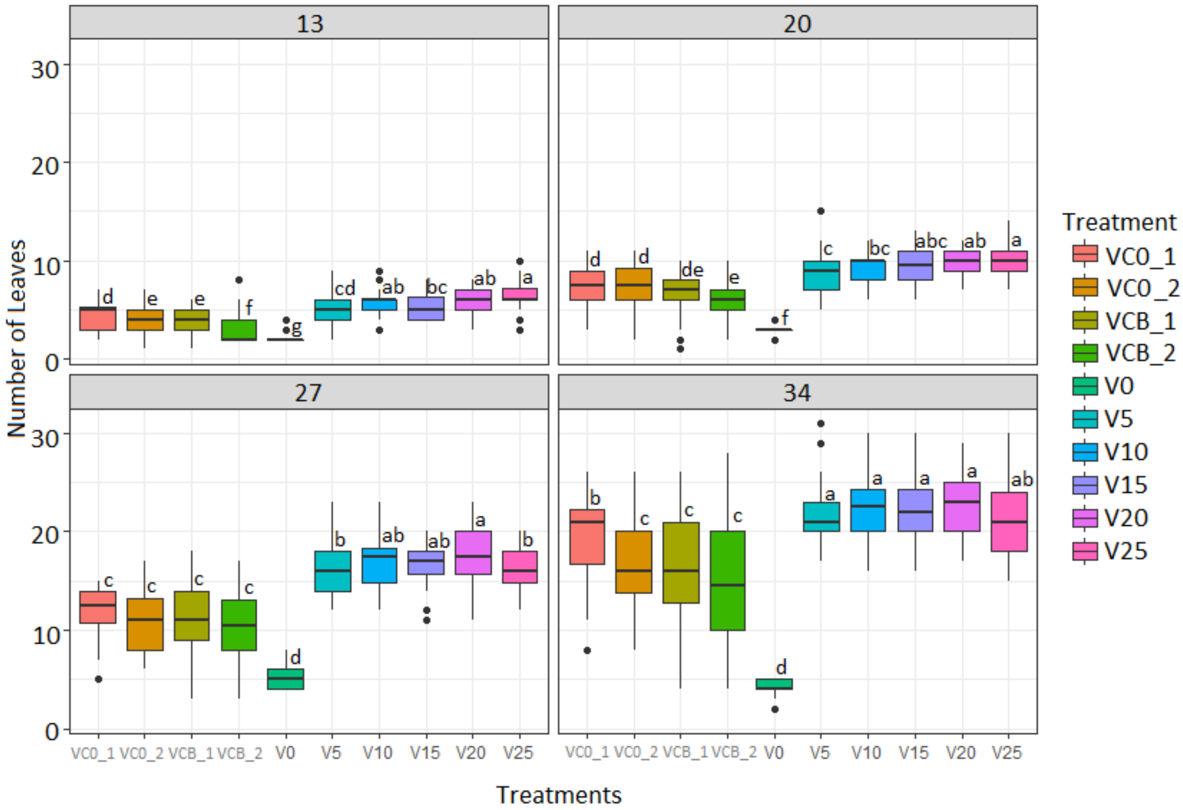


Figure 4. 2 Number of leaves per plant per treatment and Days after Transplanting (DAP) in the validation test (13, 20, 27, 34 DAP). Same letters (a, b, c, d, e, f, g) indicate no significant difference (p>0.05) between treatment for each counting time. Sample size n=32 for each treatment.

4.2.5.3 Phosphorus content

Figure A4.13 shows the total P content in the different plant organs as well as the content in the substrate, described as “undissolved”. P content in the stem show low variability along all treatments, with V25 having the highest (0.083±0.020g P) and V0 the lowest (0.008±0.002g P) at the end of the crop cycle. A great P accumulation was observed in the low production of the V0 treatment with a content of 0.107±0.005g P (54 DAP) in beans, which was even higher than the highest observed in the control for VC0 (0.094±0.013g P –54 DAP), although the greater content was found for treatment V25 with 0.172±0.023g P (54 DAP). The V0 treatment doesn’t show P results in leaves for 54 and 78 DAP because no leaves remained in the plant at the sampling time. For this same reason, there is a lack of data in beans for 78 DAP. Finally, concentration in beans for struvite treatments was similar to the one observed in the control. For all plant organs, a pattern in the accumulation of P in the plant tissue can be observed. In the first sampling all treatments show a rather low accumulation with greater content for plants with greater struvite quantities, in some cases also for the control treatments. For the second sampling, a bigger content difference can be seen with an acute increase of the V25 P content, especially for the stems and leaves. Finally, at 78 DAT, these differences between treatments even out and only treatment V0 remains significantly reduced. This last part however, does not correspond to the undissolved P in the perlite, where the P content in the substrate directly responds to the amount of struvite given, being always higher for the V25. The control treatments receive the P through irrigation making the existing content in the substrate comparably small.

4.2.6 Determination test set-up

From September 16th until November 27th, 2019, 8 double growing lines were used (totalling 256 plants), distributing the treatments as showed in Figure A4.14. The determination test was designed based on the results of the validation test. The treatment distribution was randomized throughout the Greenhouse avoiding the influence of climatic conditions. Thus, the struvite treatments were recalculated, applying per plant: 1, 2.5, 5, 7.5, 10, 15 and 20g corresponding to the treatments tagged as S1, S2.5, S5, S7.5, S10, S15 and S20, respectively. Struvite amounts below 5g were applied based on the yield and P content performance in the validation test for V5. Since we found that the perforated bag did not affect plant development, we only used one control treatment, tagged as CB, which used the same perforated bag as the struvite treatments. Moreover, considering the yield and phenological findings in the validation test, we decided not to apply mineral P fertilizer to the struvite treatments at any point, so that struvite is the only source of P to the plants.

4.2.7 Statistical analysis

The analyzed data were tested for normality using the Shapiro-Wilk test $p>0.05$. Further on, the Levene's test $p>0.05$ was used to determine homogeneity of variance. Once these parameters were validated the Duncan's multiple range test was used to assess the statistical significance of treatments. On the other hand, non-parametric data were analyzed for significance using the Kruskal-Wallis test. The significance between the treatments was marked with different letters in each plot. All statistical analyses were made with the R studio software.

4.3 Results

4.3.1 Yield

Figure 4.3 (and A15 and A16 for the final total yield) shows the results of the accumulated yield per number of harvests, being the sixth harvest (71 DAP) the final one before uprooting the plants. Only treatments S1 (78.9 g/plant) and S2.5 (128.1 g/plant) had lower yields than the control treatment (134.6 g/plant), the first being significantly lower. On the other hand, all other treatments with 5g of struvite or above produced more than the control treatment, demonstrating the potential of struvite to produce similar or even higher yields than with mineral fertilizer, as reported by Li et al. (2019).

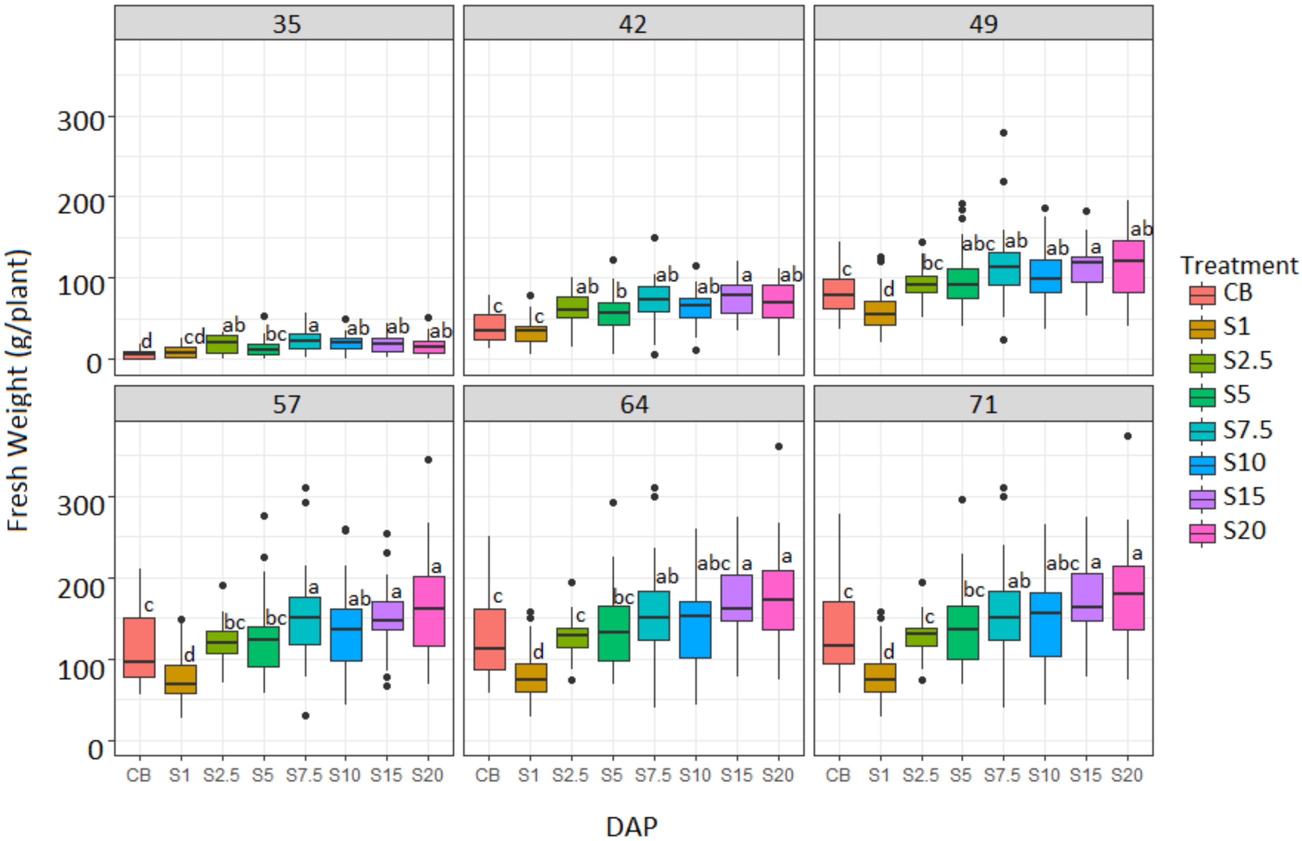


Figure 4. 3 Comparison of accumulated production of fresh bean per plant (g/plant) per treatment for each harvest time. Each panel in the figure grid represents the result for a harvesting day, with a total of 6 days (35 to 71 days after planting = DAP). Same letters (a,b,c,d) indicate no significant difference (p>0.05) between treatment for each harvest time. Sample size for all harvests is n=24 plants per treatment.

As we can see in Figure 4.3, it was not until the second harvest (42 DAP) that great differences were observed between the S1 yield and the other struvite treatments, while a decrease in S2.5 yield was observed between the 4th and 5th harvest, 57 and 64 DAP, respectively. Regarding the control treatment, the first harvest produced lower yield (6.31±5.71 g/plant) than the S5 (14.97±11.91) struvite treatment being even similar to the treatments with the lowest struvite application S1 (9.98±8.51 g/plant).

This fact reinforces the idea that the application of struvite could be beneficial for early stage plant development, as the validation test showed better behavior in struvite than in control in phenological variables. This fact could be related to the NH_4^+ supply by struvite, which could benefit the plant root balance when combined with nitrate supply (H Marschner 1995). The fact that previous literature suggests that NH_4^+ supply to common bean could be harmful for plant development (Guo et al. 2007; Chaillou et al. 1986) could be related to the amount of NH_4^+ supplied. Because struvite does not only enable a slow release of P but also of NH_4^+, reaching NH_4^+ accumulation to harmful levels seems improbable.

In terms of distribution, yields show an asymptote behaviour among treatments, where S20 produces the highest yield (g/plant) (181.41±66.16) and S1, the lowest (78.94±34.23). Figure A15 shows how treatment S10 was detected as the exception for this tendency in terms of mean production (150.50±56.10), probably related to bias parameters like shapes in the greenhouse or a non-homogenic distribution of struvite in the perlite bag. However, boxplots represented in Figure A4.16 shows how the median of the final amount of yield harvested for S10 (155.70) follows the tendency, while not presenting outliers in the distribution.

4.3.2 Water

Figure A4.17 shows that the irrigated water in the control and the struvite treatments was the same (42.5 liters per plant), while Figure 4.4 shows the accumulated P during the entire cycle in the different water streams. The quantity of P present in the control streams is much bigger than the one in the struvite streams, with the former irrigating and leaching 2.07 and 1.41 g of P per plant for the entire crop cycle, respectively. The fact that the P leachates are one order of magnitude smaller when using struvite (maximum of 0.03 g of P per plant in S20) could be related to the slow-release characteristic of struvite reported in the literature. A clear benefit of this finding is a decrease in both P depletion and freshwater eutrophication related to the leachates flow. Moreover, if the leachates of struvite treatments do not contain a large amount of P, it means that most of the struvite has been whether taken up by the plant or remains undissolved in the substrate.

When comparing Figure A4.12 and Figure A4.18, we can see that P release by struvite is highly dependent on the input water flow, represented in Figures A4.11 and A4.17 for the validation and determination test, respectively. Because the volume of irrigated water was three times less in the determination test (125.2 against 42.5 liters per plant, respectively), the P observed in the leachates is less than in the validation test, considering the period where P was not supplied through mineral fertilizer in the validation test.

Differences are observed within the struvite treatments in Figure 4.4, highly dependent on the quantity of struvite that was applied at the beginning of the crop. Treatments S1 and S2.5 stopped emitting P in the leachates just 14 DAP, which could have triggered P deficiencies. On the other hand, treatments S15 and S20 were the only struvite treatments that did not stop emitting P to the leachates flow.

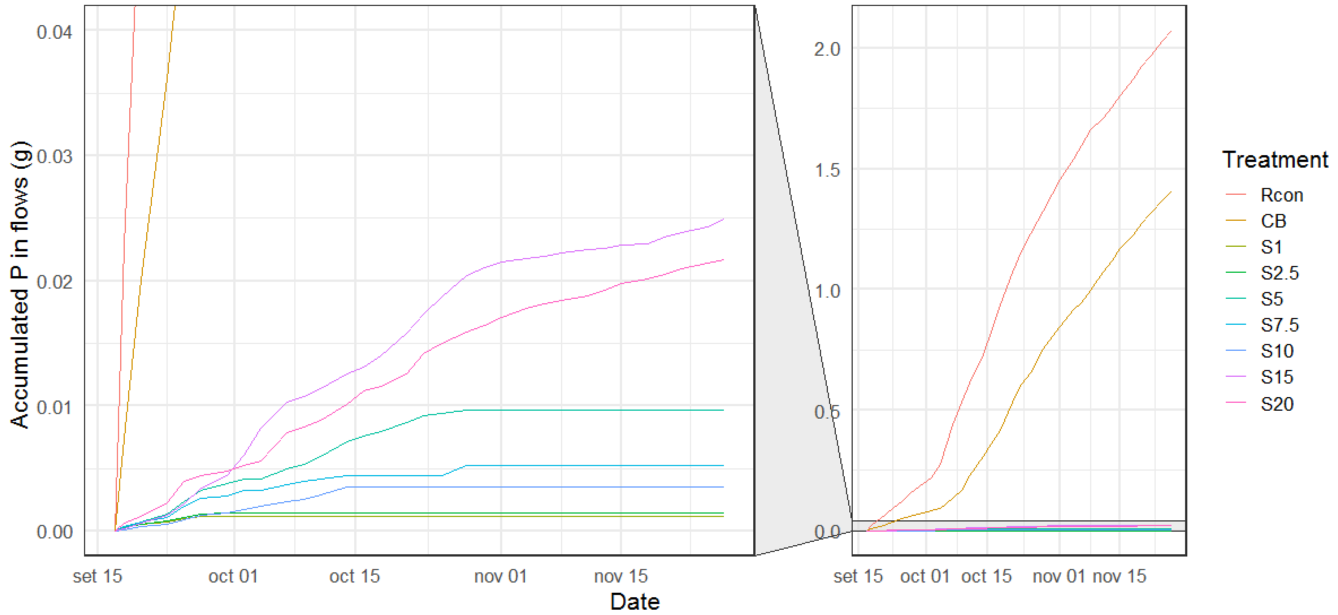


Figure 4. 4 Distribution of accumulated phosphorus in the irrigation and leachates of different treatments. Rcon: P in the control irrigation stream.

4.3.3 Substrate and undissolved struvite

Figure 4.5 shows the distribution of P among all possible input and outputs considered in the system. At the end of the crop cycle, the control treatment supplied more P (2.07 g of P per plant) than the treatment with the highest amount of struvite (S20 - 1.90 g of P per plant). Most of the P supplied in the control treatments is discharged (68%), while in the struvite treatments it still remains in the substrate.

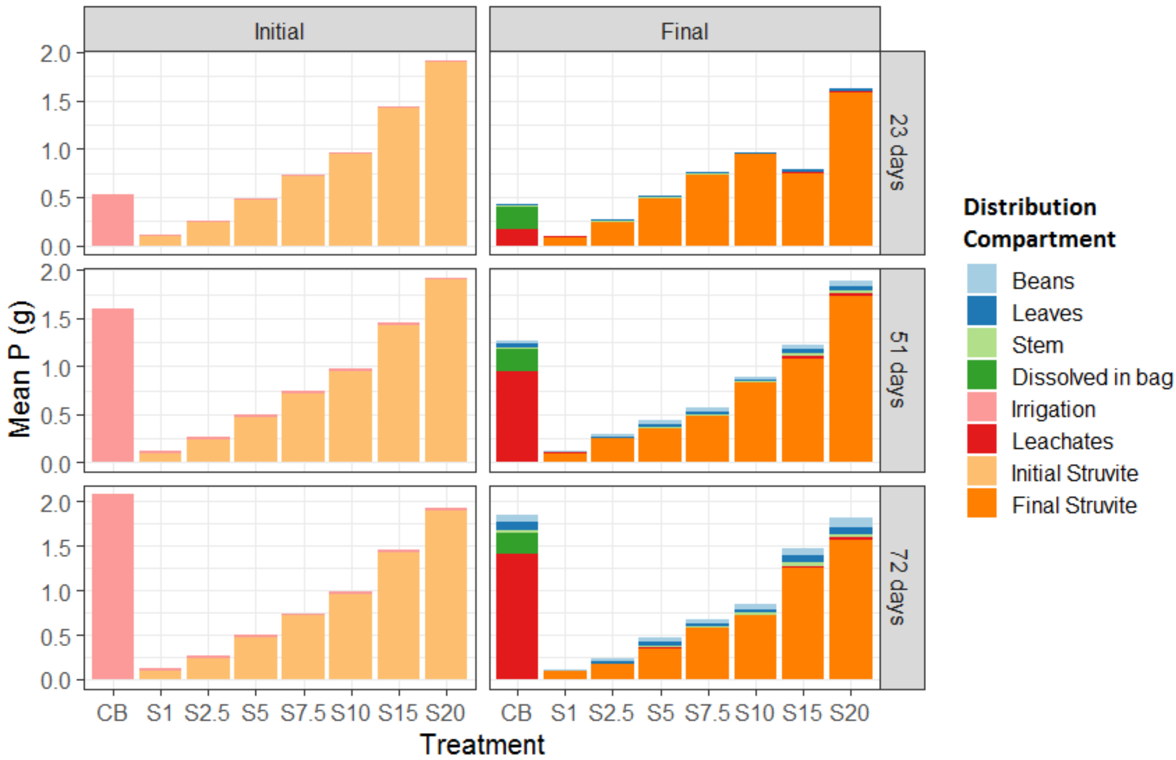


Figure 4. 5 P distribution among all water, biomass and substrate compartment flows. This amount of struvite at the end of the crop could be recovered, or the same substrate with struvite could be used for a successive crop.

4.3.4 Biomass

In terms of biomass, we can see that the concentration of P (in g per kg) (Figure 4.6) in all organs increases with the quantity of struvite applied to the treatment, having S15 and S20 similar concentrations in the leaves (7.0±1.3 and 6.7±1.8, respectively) and stem (5.0±0.9 and 4.4±1.2, respectively). However, the control treatment with mineral fertilizer presented higher concentrations of P than all struvite treatments, also in beans (7.3±0.4). This is especially relevant in the case of beans, where the P deficiency in this organ directly affects the nutritional value of the product that is going to reach the market.

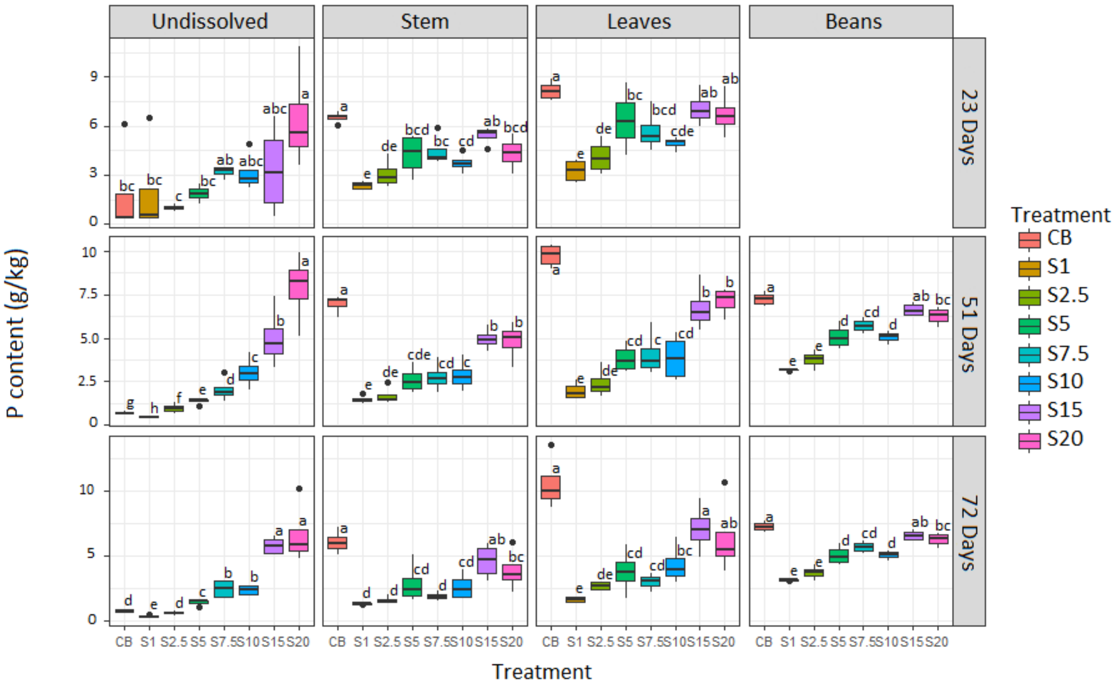


Figure 4. 6 Phosphorus concentrations (g/kg) in the different treatments, separated by plant organ and days after planting (DAP). Same letters (a, b, c, d, e, f, g, h) indicate no significant difference (p>0.05) between treatment for each harvest time. Sample size for all organs n=4. Undissolved P content n=2.

4.4 Discussion

Treatments S1 and S2.5 had lower yields than the control treatments, establishing a clear relationship between the yield and possible P deficiencies in these treatments. However, struvite remains undissolved in all treatments, even though the production and the distribution of P among plant organs was different between treatments (Figure 4.3 and Figure 4.6). The fact that we have undissolved struvite even in treatments S1 and S2.5 shows that the limitation is not only related to the quantity of struvite available, but also its dissolution (Figure 4.4 and Figure 4.6). While the struvite dissolution has been previously deemed to be due to the crystal granule size and placement (Degryse et al. 2017a; Talboys et al. 2016) previous literature fails to report the effect of the irrigated water flow. Previous experiment on the struvite dissolution in deionized water make clear that a greater dissolution can be ensured with greater temperature and stirring energy as well as an acidic pH (Ariyanto, Ang, and Sen 2017; Rahaman et al. 2006; Bhuiyan, Mavinic, and Beckie 2007; Massey et al. 2009; 2007) reaching greater dissolutions close to the commercial fertilizers. On the other hand, the volume of water flows added to the crop have not been regarded as a determining factor when granulated struvite is directly added to the substrate, especially in hydroponic production. The obtained results in the present work shed light on the effect of the incoming irrigation on the struvite dissolution as well as loss of P in the leachate.

Because the volume of irrigated water was three times lower in the determination test, the P observed in the leachates is lower than in the validation test, considering the period where P was not supplied through mineral fertilizer. Moreover, there is a significant amount of P accumulated in the substrate bag at the end of the treatment in the control test. This stored P will be depleted if a successive crop is planted, since the small nursery plants will not benefit from all of it due to the lower needs of a smaller plant. With the addition of irrigation the accumulated nutrients in the perlite bag would eventually be moved to the leachates. By applying struvite (and verified by the small amount of P in the leachates in struvite treatments) this P is not stored and thus, not lost.

Based on the findings of this study, a well-designed struvite crop cycle needs to take into account two essential parameters. First, the quantity of struvite, considering that the quantity that remains undissolved at the end of the crop can be used again for a successive cycle. Second, the irrigation management, considering that if we modify this variable to increase the dissolution of struvite granules, we would also be increasing the P in the leachates. Moreover, since previous studies highlighted the effect of the surface area of the granules on the solubility of slow-release fertilizers (Chien and Menon 1995; Gell et al. 2011; B. Li et al. 2019), the size used in our study (0.5-1.5mm) seems adequate for the balance between P supply and P lost through the leachates. Literature with higher sizes reported solubility problems that affected early plant development (Talboys et al. 2016), while studies using lower sizes or powder do not report these problems (Achat et al. 2014; Bonvin et al. 2015; Antonini et al. 2012; Gell et al. 2011). Additionally, the use of nursery plants is preferable since the struvite low dissolution has been reported to be a disadvantage when providing P to feed the transition from seeds to nursery plants (Talboys et al. 2016).

Struvite supply per plant should always be above 5g for Phaseolus vulgaris, considering that more quantity of struvite would release more P into the leachates, but ensure that P is available for plants. On the other hand, we should also account for the nutritional value of the beans, considering the ultimate function is to produce yield. In this sense, P in the biomass was a variable where the control treatment had a better performance than struvite treatments. This uptake of similar P from struvite compared to soluble fertilizers has been previously reported by Ahmed et al., 2018 determining that different crops have a greater uptake of P while other have comparable or even lower growth. While Phaseolus vulgaris was not previously observed, a study with soybean was performed compared to the P uptake with triple superphosphate (TSP). The resulting crops show a similar uptake of both P sources by the plant with different quantities of P applied (Thompson 2013). The P uptake in Phaseolus vulgaris with the use of struvite compared to monopotassium phosphate can also be seen in previous literature (Arcas-Pilz, Parada, et al. 2021) although this experiment also explores the use of rhizobium inoculation as substitute for the N fertilization, obtaining a general reduction of plant growth. It is also important to keep in mind that the quantity of applied struvite is 2g and 5g for the proposed treatments. Rech et al., 2018 also discusses the low solubility of struvite compared to TSP, also mentioning a greater uptake of P by soybean and wheat with struvite fertilization compared to the control treatment. Only S15 and S20 reach a similar P amount to the control in all plant organs. For this reason, a quantity between 15 and 20g of struvite, a responsible irrigation management and growing successive crops with the same substrate constitutes the best option to grow a well-designed struvite bean crop cycle.

Although the P uptake of the struvite fertilized treatments appears to be equal or rather smaller than the control treatment the production is greater for all treatment with more than 5g of struvite. In the literary review proposed by Ahmed et al., 2018 the increase of biomass and yield by plants fertilized with struvite can be related to the simultaneous dissolution of Mg and NH_4^+. Although the uptake of P is reported in this study the Mg and NH_4^+ concentration in the plant was not analyzed. The Mg uptake has been reported to be strongly correlated with the given Mg in the struvite and can be pointed out as a possible source reason for greater growth and production (Antonini et al. 2012; Rech et al. 2018a; N. Ahmed et al. 2018).

4.5 Conclusions

On the way towards developing more circular economies in cities, the recovery of scarce resources that can be utilized within the urban boundaries will play an important role, especially in the food vector. This study assessed the performance of the potential application of struvite recovered from WWTPs in hydroponic bean crops to diminish the need for external resources in urban agriculture. Three main conclusions could be drawn from this analysis.

First, applying struvite in hydroponics crops equals and even increases the yield compared to mineral fertilizer while diminishing P losses in the leachates, contributing to both less nutrient depletion and eutrophication potential. In this sense, a quantity above 5g/plant of struvite was observed to be enough for correct bean plant development and yield production.

Second, the input water flow was relevant in supplying enough P to the plants through dissolution using struvite. On the other hand, a correct water irrigation management is relevant to diminish P losses through over dissolution. Therefore, a balance between these two potential problems should be one of the key parameters when growing crops with struvite.

Third, a great quantity of struvite remains undissolved at the end of the crop in all treatments. In this sense, planting a successive cycle or recovering the struvite of the substrate could be alternatives to avoid losing valuable nutrients.

With the obtained information, it is adequate to say that the use of struvite in hydroponic production as a way to supply P is viable, and also serves as a way to reduce the loss of P through the water flow. The slow dissolution of this crystal also enables a single application to be effective for longer production cycles or consecutive production if the initial quantity is high enough. Our study demonstrated that no special equipment or conditions were required for the use of struvite in hydroponic production. The use of this crystal therefore is strongly recommended and it extraction and use should be pursued for further optimization of the existing P sources.

Based on the findings presented in this paper, we believe that future research should focus on three different aspects. First, the role of NH4+ supplied by struvite on plant development during the first production phase. Second, the performance of crops if successive cycles are grown using the same undissolved struvite in hydroponic systems. Third and finally, the modelling of P release by struvite based on quantity applied and input water flow.

Author contributions

All authors were responsible for the conception and design of the study. V. Arcas-Pilz, M. Rufí-Salís, A. Petit-Boix, G. Villalba and X. Gabarrell conceived the original idea for the study. M. Rufí-Salís, V. Arcas-Pilz and F. Parada set up, supervised and acquired the data for the experimental tests. M. Rufí-Salís and V. Arcas-Pilz processed and analysed the data. M. Rufí-Salís and V. Arcas-Pilz took the lead in writing the manuscript. All authors critically revised the draft for important intellectual content. All authors gave their final approval to the manuscript.

Chapter 5

This chapter is based on the journal paper:

Improving the fertigation of soilless urban vertical agriculture through the combination of struvite and rhizobia inoculation in Phaseolus vulgaris

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Chapter 5: Improving the fertigation of soilless urban vertical agriculture through the combination of struvite and rhizobia inoculation in Phaseolus vulgaris

Abstract

Soilless crop production is a viable way to promote vertical agriculture in urban areas but relies extensively on the use of mineral fertilizer. Thus, the benefits of fresher, local food and avoiding the transportation and packaging that are associated with food imports could be counteracted by an increase in nutrient-rich wastewater, which could contribute to freshwater and marine eutrophication. The present study aimed to explore the use of mineral fertilizer substitutes in soilless agriculture. Phaseolus vulgaris (common bean) was fertilized with a combination of slow-releasing fertilizer struvite (a source of N, P, and Mg), which is a byproduct of wastewater treatment plants, and inoculation with Rhizobium (a N2-fixing soil bacteria). The experiment included three bean production lines: A) 2 g/plant of struvite and rhizobial inoculation; B) 5 g/plant of struvite and rhizobial inoculation, both irrigated with a Mg-, P- and N-free nutrient solution; and C) a control treatment that consisted of irrigation with a full nutrient solution and no inoculation. Plant growth, development, yields and nutrient contents were determined at 35, 62 and 84 days after transplanting, as well as biological N2 fixation that was determined using the 15N natural abundance method. Treatments A and B resulted in lower total yields per plant than the control C treatment (e.g., 59.35± 26.4 g plant-1 for A, 74.2±23.0 g plant-1 for B and 147.71± 45.3 g plant-1 for C). For A and B, the nodulation and N2 fixation capacities appeared to increase with the amount of initially available struvite, but over time, deficient levels of Mg were reached as well as nearly deficient levels of P, which could explain the lower yields. Nevertheless, we conclude that the combination of struvite and N2-fixing bacteria covered the N needs of plants throughout the growth cycle. However, further studies are needed to determine the optimal struvite quantities for vertical agriculture systems that can meet the P and Mg requirements throughout the lifetime of the plants.

5.1 Introduction

From 1950 to 2018, the population living in urban areas grew more than fourfold to an estimated 4.2 billion people. This unprecedented population increase has greatly increased global food demand, which has exerted great pressure on natural resources (United Nations 2019). In response, new ways to efficiently produce vegetables while minimizing land use are being explored (Sanyé-Mengual et al. 2015; 2018). One of these initiatives is vertical farming with the use of soilless production systems with growing media or substrates (Sonneveld and Voogt 2009), which would reduce the transportation and packaging of foodstuffs to cities (Sanyé et al. 2012a). However, vertical agriculture relies extensively on the use of mineral fertilizer, which results in nitrates and phosphate being discharged into wastewater, which can contribute to freshwater and marine eutrophication (Anton et al. 2005; Gopalakrishnan et al. 2015; Sanjuan-Delmás et al. 2018).

This extensive use of mineral fertilizers affects not only the environment but can also be related to a high cost of production and extraction, as is the case for nitrogen fertilizers due to the Haber-Bosch process (Cherkasov, Ibhadon, and Fitzpatrick 2015) and for phosphorous due to phosphate rock extraction (Cordell and White 2013). The widepread use of these nutrients has caused vertical farming to rely entirely on them, which thus makes this agricultural practice unsustainable in the long run. The high energy cost of synthetic nitrogen production and the ever-depleting sources of phosphate rock, when added to the environmental cost of their disposal and emissions to water and air (Rufí-Salís et al. 2020; Rufí-Salís, Petit-Boix, Villalba, Sanjuan-Delmás, et al. 2020a), necessitates the search for alternatives to further implement these technologies in a sustainable way.

Many strategies have been described in recent years for the implementation of organic fertilization in vertical farming, which embrace a circular economy framework to reduce new resource inputs into cities. Some examples include fertilization that is based on gray water and urine (Ikeda and Tan 1998; Karak and Bhattacharyya 2011) and the use of biofertilizers such as Rhizobium for the cultivation of legumes (C. K. Kontopoulou et al. 2015; Savvas et al. 2018a) for the plant nitrogen supply. Other methods describe the use of sewage sludge (Frossard, Sinaj, and Dufour 1996) and sewage sludge ash (Nanzer et al. 2014) as well as struvite (Rech et al. 2018a) as alternative P sources. While these strategies may reduce the direct inputs of specific inorganic fertilizers, their use often results in lower crop yields and, in some cases, require more infrastructure for irrigation systems. These studies tend to focus on one particular nutrient alternative, not considering the combination of alternative methodologies. Therefore, innovation to provide a solution for multiple mineral fertilizers while avoiding the addition of infrastructure as well as further environmental burdens due to local nutrient sourcing, has not been widely studied.

Struvite (MgNH4PO4.6H2O), which is a crystalline byproduct of wastewater treatment plants that formes by spontaneous or induced precipitation, usually contains high N and P concentrations (Rahman et al. 2014), and is regarded as a viable slow-releasing fertilizer due to its high P, Mg and N contents, which average 12.5%, 9.9% and 5.7%, respectively (N. Ahmed et al. 2018) and are suitable for plant growth (Degryse et al. 2017a; N. Ahmed et al. 2018). Due to struvite's high nutrient concentrations, there are many ongoing efforts to optimize induced precipitation to make wastewater a valuable resource for providing a P alternative to the use of the depleting phosphate rock (Massey, M.S., Davis, J.G., Sheffield, R.E., Ippolito 2007; Cordell, Drangert, and White 2009; Talboys et al. 2016; Degryse et al. 2017a).

A further positive aspect of struvite as an agricultural fertilizer substitute is its slow solubility in granular form (Talboys et al. 2016) under alkaline and neutral pH soil conditions (Bhuiyan, Mavinic, and Beckie 2007). Thus, the risks of nutrient leaching and water eutrophication are rather small under these conditions when struvite is compared to common readily soluble fertilizers (N. Ahmed et al. 2018). Furthermore, the removal of approximately 30%-40% of N and P from wastewater to produce this substance can prevent eutrophication in urban water cycles (González Ponce, López-de-Sá, and Plaza 2009; Antonini et al. 2012). The granular form of struvite also causes it to be easily manageable and could be applied in larger-scale productions by mixing it with soil or applying it to the substrates in hydroponic production systems. The use of struvite has already been tested in agriculture as a substitute for phosphate from other sources and has shown promising results with low or even no yield losses reported (Degryse et al. 2017a; J. N. Ackerman et al. 2013; González Ponce, López-de-Sá, and Plaza 2009; Cabeza et al. 2011; Y. H. Liu et al. 2011; N. Ahmed et al. 2018)

Although struvite already contains N that is available to plants, legumes have high N demands (McKey 1994). Therefore, the average N contents in struvite would not be sufficient for soilless crops to achieve commercial yields and would require a second source of N to do so. This N could be obtained from Rhizobium, which is capable of forming an endosymbiotic interaction with leguminous plants by entering root cells and forming nodules. These nodules enable atmospheric N2 fixation and ammonia (NH3) formation. Plants benefit from the bacteria that generate these compounds, while the bacteria can profit from photosynthesis-derived compounds (Long 1989). This symbiosis, on the other hand, may entail a major requirement of nutrients from the plant, such as phosphorous, to satisfy the needs of the bacteria and successful nodulation (Olivera et al. 2004). Possible N2 fixation depends on successful rhizobial root colonization, which is influenced by diverse factors, such as phosphorous fertilization, salinity, drought and initial N availability (Savvas et al. 2018a; Ntatsi et al. 2018; Araújo, Monteiro, and Carvalho 2007).

Rhizobium as a second source of N was chosen due to the lower inputs needed to achieve nitrogen intake by plants (Gopalakrishnan et al. 2015). When using the N2-fixing bacterium, Rhizobium, in hydroponic cultivation, Kontopoulou et al., (2017) described the need to apply initial N fertilization until nodulation in the root medium occurs, to further encourage nodulation and therefore N fixation, plant growth and production. Even though previous studies have reported lower production capacities for N2-fixing plants than for common beans with N fertilization (C. K. Kontopoulou et al. 2017; Olivera et al. 2004), a combination of the two N sources (e.g., struvite and N2-fixing bacteria) was used to determine the possibility of overcoming such lower yields (Savvas et al. 2018a; Pampana et al. 2017).

To determine how effective the two alternative fertilizers are in providing N to plants, the 15N natural abundance method was employed to determine the source of N throughout the experiment (Shearer and Kohl 1989). While plants with N that is acquired from symbiotic atmospheric N2 fixation show lower richness of the 15N isotope, which corresponds to the atmospheric abundance (0.3663%), plant tissues that are subjected to other N sources can exhibit greater amounts of the 15N isotope, which depend on the N fertilizer applied (Robinson 2001).

The present study aimed to add to this growing pool of knowledge on vertical urban agriculture by exploring the use of mineral fertilizer substitutes struvite and rhizobium combined in an effort to reduce emissions of simultaneously N and P to the environment in urban vertical agriculture. This combination also aims to optimize crop yields while avoiding the installation of additional infrastructure. In this study, we analyzed the growth, development and production of the common bean (Phaseolus vulgaris), which was fertilized with a combination of the slow releasing fertilizer, struvite, and the soil bacteria, Rhizobium. A combination of these alternative fertilizers can be implemented easily in terms of cost and space and promotes nutrient recycling within cities.

5.2 Materials and Methods

5.2.1 Experimental site, materials and growth conditions

This experiment was conducted in the Rooftop Greenhouse Laboratory (RTG-Lab) of the Environmental Science and Technology Building (ICTA-UAB), which is located in the Universitat Autònoma de Barcelona Campus (42°29’24” E, 45°94’36” N) (Sanjuan-Delmás et al. 2018). The bean variety used in this experiment was Phaseolus v. Pongo, which had previously been germinated in a commercial greenhouse ten days before transplanting in the RTG-Lab. The production system was soilless with a perlite substrate in 40 L bags and the use of fertigation through a 2 L/h drip irrigation system.

Bean seeds were treated with a commercial product (e.g., Nadicom GmbH©) which contained a mixture of Rhizobium phaseoli and Rhizobium giardinii strains for inoculation before planting. The inoculation procedure was an exposure of the plant seeds with the liquid commercial product before planting. Five days after the seedling was transplanted into the perlite substrate, an addition of 5ml liquid commercial mix was made to each plant therefore ensuring the presence of the bacteria in the substrate. Once the plants were inoculated, they were irrigated with an

Mg-, P- and N-free solution (Table A5.1 in the Annex II), and application of K_2SO_4 was increased to adjust for the K requirements. The control plants, on the other hand, were irrigated with a full nutrient solution. These nutrient concentrations were maintained throughout the entire experiment. The crops were irrigated 4 times a day for 3 minutes, which provided a total amount of 400 ml per day per plant.

The inoculated plants were treated with two different struvite amounts placed inside the perlite bag around the root area and surface, varying the concentration of P and N available to the plant from struvite: A) 2 g (1.02 mmol of P and 0.46 mmol of N) of granulated struvite per plant and B) 5 g (2.57 mmol of P and 1.15 mmol of N) of granulated struvite per plant. The amount of struvite that was best for growth was determined in a previous experiment conducted in the same i-RTG, in which 2.57 mmol P was deemed sufficient for common bean fertilization to reach an equivalent level of commercial production as that of mineral-fertilized beans. To ensure no struvite loss due to runoff, each plant was planted inside an additional 1 L bag containing perlite and the corresponding amount of struvite, with small holes to allow water drainage.

Each treatment was arranged randomly in four rows with 16 plants each (4 perlite bags with 4 plants per bag were planted in a frame with an area of 0.125m2) which resulted in a total of 64 plants per treatment (e.g., A, B, and Control), with 192 plants in total (Figure A5.4 of the Annex II). Due to the irrigation and leachate recovery systems, randomization could only be achieved for entire lines of 4 bags.

The plants were germinated and transplanted in duplicate and were thinned to one plant at 21 days after transplanting (DAT).

Greenhouse conditions were monitored during the entire experiment with T107 sensing devices (Campbell Scientific) that were placed along the cropping area to measure temperature, relative humidity and radiation (see Table A5.2 of the Annex II). To ensure proper plant irrigation drainage volumes, the pH and electrical conductivity levels of the leachate were recorded every day for each irrigation line.

The phenological stages of the bean plants were determined each week. This information was assessed to identify plant growth, development and productivity over time, and provided a clear view of the plant cycle, growth and production peaks that enabled accurate comparisons of plant development between treatments and the control. This was performed by counting leaves, flower buttons and open flowers. The number of ripened bean pods was also counted and weighed for each harvest. These measurements were performed for each of the eight plants that were in the two middle bags of each row (see figure A5.4) and started 14 DAT. To ensure uniform counting, leaves under 5 cm length were not considered, and only fully formed flower buttons with white coloration and fully open flowers were counted. For the bean pods, a minimum length of 11 cm was used for harvesting, while bean pods shorter than this were retained for the next harvest. The average numbers and bean pod weights per treatment were then calculated for each week. At the same time and on a weekly basis, chlorophyll content measurements were performed (with an SPAD CCM-200 plus; Opti-Sciences, Inc.) on the same eight plants in the center of each row (see figure A5.4 of the Annex II).

5.2.2 Description of plant sampling methods

To determine the changes in plant development as well as nutritional states and 15N concentrations, samples were taken during the three different crop stages. The first sampling took place 35 DAT, immediately before bean pod production started; the second sampling took place 62 DAT, during the productive phase of the plants; and the last sampling took place 84 DAT, at the end of the productive stage, which marked the last day of the experiment.

The samples consisted of eight randomly chosen plants per treatment (excluding the eight central plants of each row that were kept for phenological analysis). Each plant was washed with deionized water, excess water was dried off and each plant was separated into the four main organs: leaves, shoots, roots and nodules. These were then weighed separately to determine their fresh weights (FW). All organs were placed separately in envelopes and left to dry in an oven at 65°C until stabilized dry weights (DW) were obtained, which occurred after approximately 7-8 days. The means of the obtained values were calculated for each treatment, each organ and time. The numbers of nodules were counted prior to drying to determine the mean nodulation of each plant. In addition, fruit samples from each treatment were taken at three different times (49, 62 and 77 DAT), which closely matched the three plant harvests.

Moreover, 25% of the total sampled leaves for each plant were separated to determine their areas before the drying process. To do so, these fresh leaves were scanned with a reference pixel to obtain leaf areas using ImageJ software (Rueden et al. 2017). These leaf areas were further extrapolated to 100% of the leaf biomass of the plant. The leaf area index (LAI) was then calculated by dividing the total leaf area by the area of the planting frame of our crop (0.125m2).

5.2.3 Nitrogen isotopic (δ 15N) analysis

The goal of inoculating treatments A and B with Rhizobium was for the plants to indirectly fix N2 from the air and meet their N needs in this way. To determine how much of the N assimilated by the plants came from the atmosphere, we used the natural abundance method (Shearer and Kohl 1989) to identify the origin of the N that was obtained by the plants, which in our case, should be either struvite or atmospheric N. While treatments A and B were actively inoculated with Rhizobium strains and fertilized with struvite containing N, the control treatment was fertilized through standard N fertilization that was administered through irrigation. Additional nitrogen sources were not considered due to the laboratory conditions and production on inert perlite.

Analysis was performed with an elemental analyzer isotopic ratio mass spectrometer (EA-IRMS; Thermo Fisher Scientific). The devices used were a Flash EA 1112 analyzer and Delta V Advantage spectrometer that were coupled with a Conflo III interface. The plant and struvite samples were weighed in tin capsules and were introduced into the EA-IRMS system to obtain the δ15N values, as calculated with the following equation (Eq 4) (Robinson 2001):

$$Eq\ 4: \qquad \delta^{15}N = \frac{Sample\ atom\ \%^{15}N - 0.3663}{0.3663} \times 1000$$

Equation 4: δ15N is the natural tracer for our N sources, the sample atom %15N is the previously obtained value of our plant sample, and the value 0.3663 is a standard value that represents the percentage of 15N in the atmosphere.

δ15N values provide an indication of the N sources in plant tissues. Values close to 0 indicate that the plant N sources are mainly due to atmospheric N2 fixation, while higher values can be interpreted as indicating mixed sources or those dominated by the N obtained from struvite. The δ15N value obtained for the struvite used in the experiment was 7.1‰. To determine the relative contributions from the two sources considered, we used Eq 5, which yields an estimate of the percentage of N that was derived from N2- fixation (%Ndfa) (Shearer and Kohl 1993; Unkovich et al. 2002; Arndt et al. 2004)

$$Eq\ 5: \qquad \%Ndfa = \frac{\delta^{15}N\ Source\ 2 - \delta^{15}N\ Sink}{\delta^{15}N\ Source\ 2 - 'B'value} \times 100$$

Equation 5: %Ndfa (nitrogen derived from N2 fixation from the atmosphere), δ15N Source 2 (‰) corresponds to the δ15N value of struvite, δ15N Sink (‰) corresponds to the δ15N value from the sample, and the 'B' value corresponds to the δ15N of N2 fixation taking into account possible fractionation.

The 'B' value is the isotopic fractionation observed in N2-fixing P. vulgaris, was set to -1.16‰, which corresponded to the lowest δ15N value obtained (Shearer and Kohl 1989; Peoples, Boddey, and Herridge 2002; Kermah et al. 2018) and was similar to the values determined by Kontopoulou et al. (2017) in common bean that was fertilized without N and inoculated with Rhizobium.

The biologically fixed nitrogen (BNF) levels were further calculated with the obtained %Ndfa values as well as the obtained values for the nitrogen contents in the plants. To extrapolate to kg/ha, a theoretical plant density of 8 plants/m2 was used.

Finally, the nitrogen use efficiency (NUE) for all treatments was estimated. The methodology that was followed to perform these calculations was given by Weih, 2014, who provided a tool to successfully calculate the NUE. To accomplish this, the information provided was:

- N content at the initial stage of the plant in g/m2 (previous to the main production stage at 35 DAT)
- N content at the main productive stage in g/m2 (chosen at 84 DAT)
- N content in the harvested yield in g/m2
- Biomass yield g/m2
- Added N to the soil in g/m2 (in this case, perlite)

5.2.4 Plant nutritional analysis

Dried and ground plant organs were weighed (up to 0.25 g) and digested using a single reaction chamber microwave (Milestone Ultrawave) with concentrated HNO_3. The digested samples were then diluted with 1% HNO_3 (v/v) and were analyzed by optical spectrometry (ICP-OES) (Perkin-Elmer, Optima 4300DV). All samples were weighed, digested and analyzed in duplicate.

5.2.5 Statistical analysis

All statistical analyses in this experiment were performed with R studio software. Data normality in our values was tested with Shapiro-Wilk test $p>0.05$, to ensure homogeneity of variance the Levene test was performed $p>0.05$. When both criteria were validated Duncan's multiple range test was used to assess the statistical significance of treatments. The Kruskal-Wallis test was used for no parametric data. The significance between the treatments was tested for each harvest time separately.

5.3 Results

5.3.1 Phenology, biomass and yield

Weekly recordings of the phenological growth of the bean plants exhibited differences among all treatments (Figure 5.1). In this figure, we can see the evolution throughout the crop development of biomass production as well as flower production and finally bean pod production. The control plants (Treatment C) showed greater biomass growth and faster development in their transitions from flower buttons to open flowers and bean pod production. Although the treatment performances were similar in the earlier growth stages, once the production stage started, greater differences were observed.

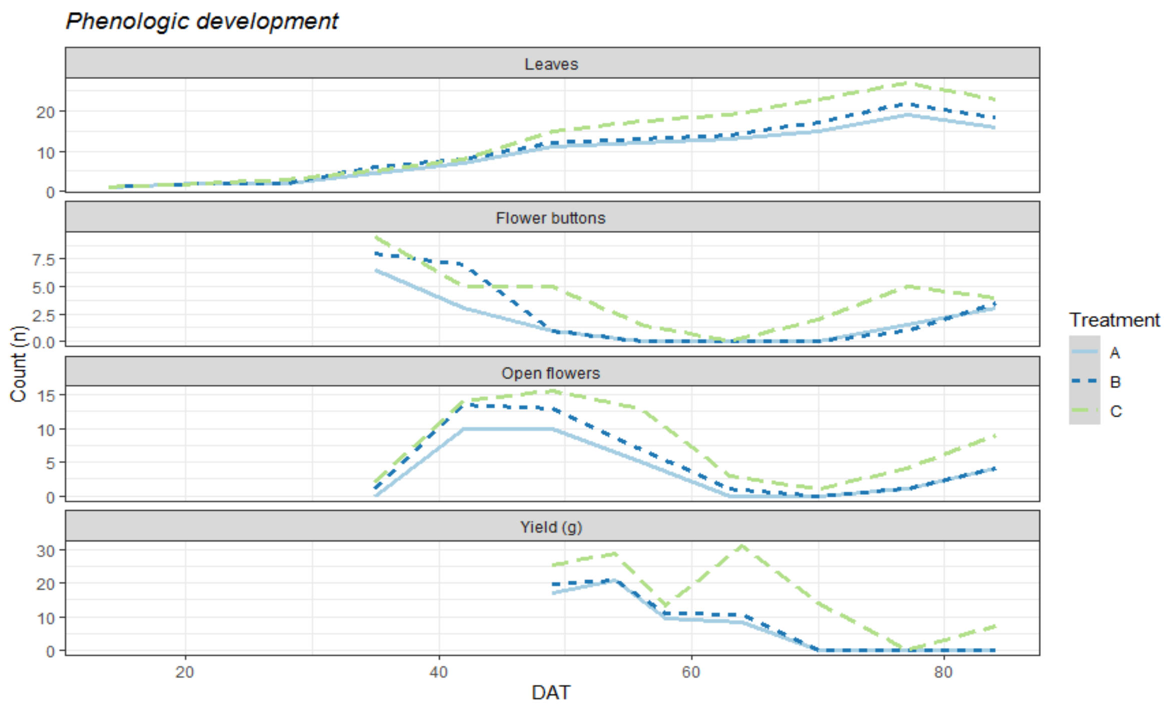

Figure 5. 1 Graphic representation of the mean numerical count per plant for each organ (Leaves, flower buttons, open flowers) and yield in g/plant on a weekly basis, DAT representing the days after transplanting inside the iRTG. The colors represent the three treatments: (A) N-free solution with Rhizobium inoculation and 2g of struvite per plant. (B) N-free solution with Rhizobium inoculation and 5g per plant. (C) Complete nutrient solution without struvite and no inoculation treatment.

At 40 DAT and 50 DAT, treatments A and B began to perform worse for leaf production as well as for the formation and opening of flower buttons than the control plants (C). Between 60 DAT and 70 DAT, a second production peak can be seen for the control treatment as well as rapid generation of flower buttons, while treatments A and B showed a declining pattern for bean pod production.

Table 5.1 shows the changes in the plant measurement results that were conducted on the sampled plants at three different developmental stages. While the first period of plant sampling, 35 DAT, showed very few significant differences among the treatments (only in the case of dry weight), the later samplings at 62 and 84 DAT showed greater differences between treatments. At this point, the leaf and shoot dry weights were greater for the control treatment, as was the measured leaf area index. The only parameter without significant differences among treatments throughout the entire experiment was the root dry weight at 62 DAT. The dry weights of the nodules exhibited persistent, significant differences for the three samplings among treatments A and B and control treatment C and reached maximum values of 0.16 g, 0.12 g and 0.05 g for treatments A, B and C, respectively. On the other hand, treatment B (with higher struvite quantities) also exhibited significantly greater numbers of nodules as well as higher weights than the other two treatments during the third sample periods.

Table 5. 1 Results for the mean values (n=8) per plant of fresh weight (FW) and dry weight (DW) of the different organs as well as the Leaf Area Index (LAI) m2 plant-1 of the three treatments (A= 2g Struvite + Rhizobium; B= 5g Struvite + Rhizobium; C = Control) in three different time periods. (35 DAT (1), 62 (2) and 84 DAT (3). Significant differences (p< 0.05) between treatments marked with different letters (a,b,c)

(1)	Leaf DW (g) per plant	Shoot DW (g) per plant	Roots DW (g) per plant	Nodules n per plant	Nodules DW per plant (g)	LAI
A	1.12a±0.22	0.46a±0.08	0.44a±0.10	132.50a±80.35	0.16b±0.07	0.57a±0.12
B	1.31a±0.46	0.56a±0.19	0.51a±0.15	156.75a±60.82	0.12b±0.06	0.62a±0.23
C	1.33a±0.57	0.58a±0.18	0.53a±0.14	148.75a±48.23	0.05a±0.02	0.65a±0.27

(2)	Leaf DW (g) per plant	Shoot DW (g) per plant	Roots DW (g) per plant	Nodules n per plant	Nodules DW per plant (g)	LAI
A	3.97a±1.25	2.02a±0.72	0.80a±0.28	127.88a±63.85	0.14b±0.09	1.28a±0.51
B	3.69a±1.53	2.24a±1.01	0.87a±0.34	172.25a±132.66	0.15b±0.14	1.29a±0.64
C	6.44b±3.09	3.85b±1.95	0.95a±0.44	82.25a±62.47	0.01a±0.01	2.64b±1.33

(3)	Leaf DW (g) per plant	Shoot DW (g) per plant	Roots DW (g) per plant	Nodules n per plant	Nodules DW per plant (g)	LAI
A	5.86a±2.96	3.09a±1.45	1.77a±0.79	136.88b±106.31	0.15b±0.13	1.74a±0.92
B	7.40a±2.17	4.53a±1.48	2.49a±0.57	186.25c±48.79	0.24b±0.11	1.80a±0.67
C	11.11b±1.51	6.91b±1.42	3.35b±0.88	39.13a±24.76	0.02a±0.02	3.72b±0.87

When examining the SPAD measurements (Figure A5.5), some differences in chlorophyll content were observed throughout the experiment. Initially we can see a significant difference between the A and B treatments and the control marking a greater chlorophyll content in the latter that is sustained until 35 DAT. From 42 DAT to 63 DAT the chlorophyll content in treatments A and B increases while treatment C remains stable. While differences towards the end of the experiment remain small, we can appreciate a greater chlorophyll content in the struvite fertilized treatments.

The final production amounts that were obtained for all three treatments were 1899.2 g, 2375.6 g and 4726.7 g of green bean pods for treatments A, B and C, respectively. Although the plants treated with struvite and rhizobium produced approximately half the yield of the mineral-fertilized plants, it is important to note that they were healthy throughout the experiment. The average yields provided per plant were 59.35± 26.4 ga plant-1 for A, 74.24±23.0 ga plant-1 for B and 147.71± 45.3 gb plant-1 for the control treatment C. These production differences can also be seen in figure 1 where the obtained yields are shown as a function of time and show greater production peaks and a more rapid capacity to develop flower buttons and open flowers after each harvest.

5.3.2 d15N, %Ndfa and Biologically fixed N

The results obtained for the δ15N values of plant tissues and bean pods (Figure 5.2 and Figure A5.6) show great variability in the enrichment of all organs except for the nodules. While treatment C showed clear enrichment over time, the pattern for treatments A and B was the opposite. For the nodules, all three treatments exhibited clear enrichment over time. Treatment B exhibited intermediate δ15N values that were between those of A and C, with decreasing δ15N values that were not as abrupt when compared to the tissues that were exposed to treatment A. It was also interesting to observe that the major decrease in δ15N values for treatment A occurred between days 35 and 62 after transplanting and remained rather constant at 84 DAT. For the plants in treatment B, the value at 62 DAT did not fall as drastically and experienced a more significant change at 84 DAT.

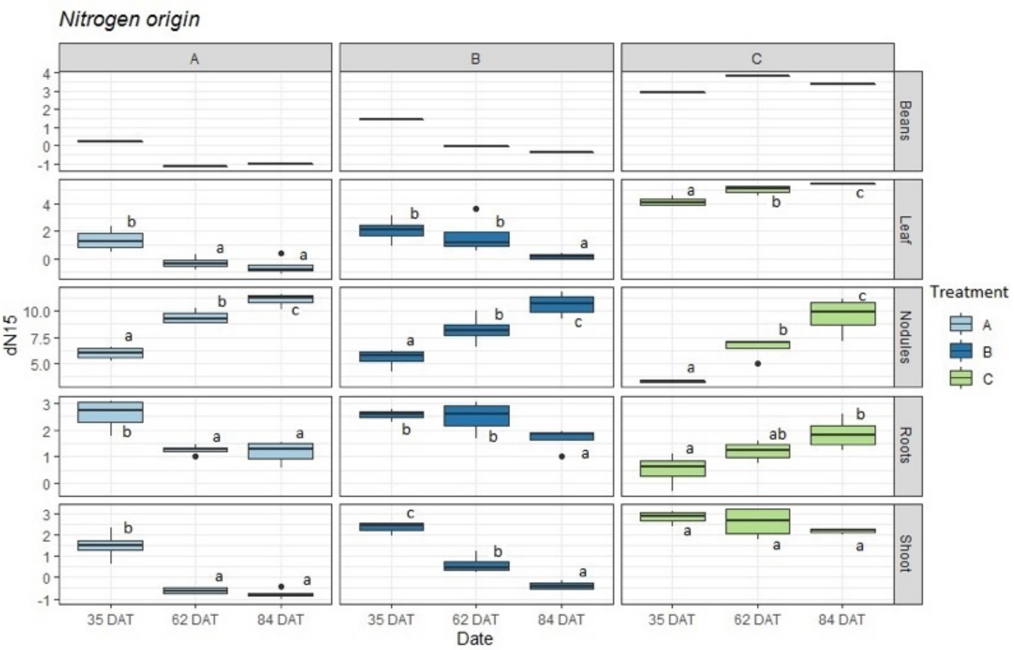

Figure 5. 2 Boxplot representing the obtained δ15N values (n=4) for treatments A) 2g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution B) 5g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution and C) standard nutrient solution – Rhizobium inoculation. These observed values are given by plant organ in three different time periods, 35 DAT, 62 DAT and 84 DAT. Significant differences (p<0.05) between dates marked with different letters (a,b,c).

When calculating the percentage of fixed atmospheric N during our three sampling periods, we obtained the values shown in Figure 5.3. This figure shows the approximate percentages of N that were derived from atmospheric fixation relative to the total N obtained by the plants.
As shown in the figure, the percentages of fixed N_2 in all three tissues were higher for the plants in treatment A, with values of 65% to 80% at 35 DAT, which reached 90% by the end of the experiment (84 DAT). On the other hand, treatment B exhibited lower values throughout the experiment, with initial values close to 50% to 60% (35 DAT), which reached final values of 80% at 84 DAT.

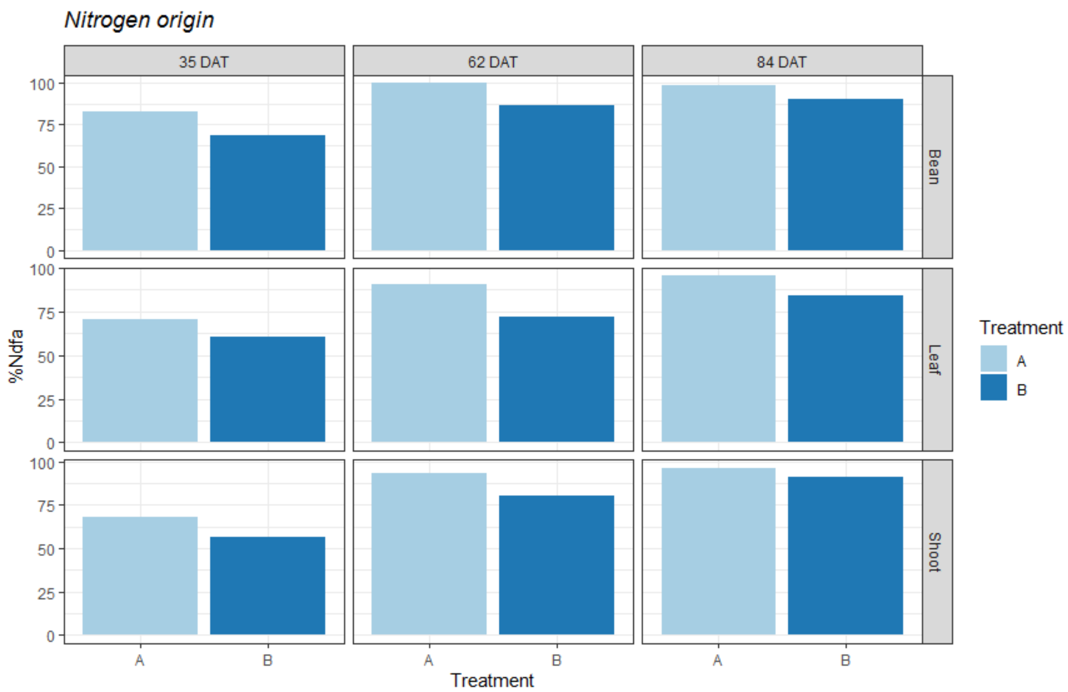

Figure 5. 3 Percentage of Nitrogen derived from atmospheric N2 fixation (%Ndfa) represented for treatments A) 2g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution B) 5g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution. These observed values are given for two plant organs (leaf; shoot) as well as the bean pods in the three different time periods, 35 DAT, 62 DAT and 84 DAT.

While the plants with less struvite in the root medium (treatment A) increased their percentages of fixed N_2 more rapidly (from 70% (35 DAT) to 90% (62 DAT) in the leaves), the plants in treatment B took longer to reach this value (from 60% (35 DAT) to 71% (62 DAT) in leaves). This corresponds to the results for the δ15N values shown in figure 5.2.
Table 5.2 shows the results of the estimations of biological fixed nitrogen (BNF) contents expressed in kg/ha. These results show the extrapolations of total N found in the plants for each treatment to kg/ha values. The N percentages that were of atmospheric origin (obtained

previously) were further used to attain the kg/ha of biologically fixed nitrogen for each treatment as well as the N from struvite that was used by the plants.

Table 5. 2 Results for percentage of Nitrogen derived from atmospheric N_2 fixation (%Ndfa) in plant, Total amount of N in plant expressed in kg/ha (Leaves+Shoot+Root+Beans) and Biologically fixed N extressed in kg/ha. Results given for three treatments (n=8 each) A) 2g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution B) 5g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution at three different time periods. 35 DAT, 62 DAT and 84 DAT. Significant differences (p<0.05) between treatments marked with different letters (a,b,c).

Date	Treatment	% Ndfa plant-1	Total N in plant kg/ha	Kg/ha Biologically fixed N	Kg/ha N from Struvite
35 DAT	A	68%	7.5±1.0a	5.4±1.0 a	2.2
	B	60%	8.6±2.2 a	5.3±1.4 a	3.3
62 DAT	A	89%	24.7±5.0 a	22.9±4.0 b	1.8
	B	73%	24.6±6.2 a	18.7±5.1 a	5.9
84 DAT	A	90%	27.3±12.8 a	25.4±13.0 a	1.9
	B	82%	35.0±9.2 a	29.2±7.8 a	5.8

Here, we can see that as the percentages of atmospheric-derived N and total N that were found in the plants increased, as well as the kg/ha values of biologically fixed N. While the plants in treatment A had higher values of biologically fixed N during the first two sampling periods at 84 DAT, the increase in the fixation percentage and total N in the plants in treatment B increased their amounts of biologically fixed N. On the other hand, the use of N from struvite increased only for treatment B and remained constant for treatment A.

5.3.3 Nutrient content

The nutrient contents in the aboveground plant organs are presented in figure 5.4 (Figure A5.8 in the Appendix II for differences between harvests). The observed concentrations of nutrients in leaves for the three treatments were at sufficient levels except for the less than optimal Mg2+ concentrations at 62 DAT for treatments A and at 84 DAT for treatments A and B and were close to deficient levels P in both treatments A and B at 62 and 84 DAT according to Hochmuth et al.(2018). In the case of N, in both leaf and shoot tissues, no deficient levels were found for any of the treatments, and no significant differences were found among treatments. On the other hand, a clear decline in P and Mg2+ over time can be seen for treatments A and B in the leaves as well as for P in the shoots. The control treatment (C), on the other hand, remained stable.

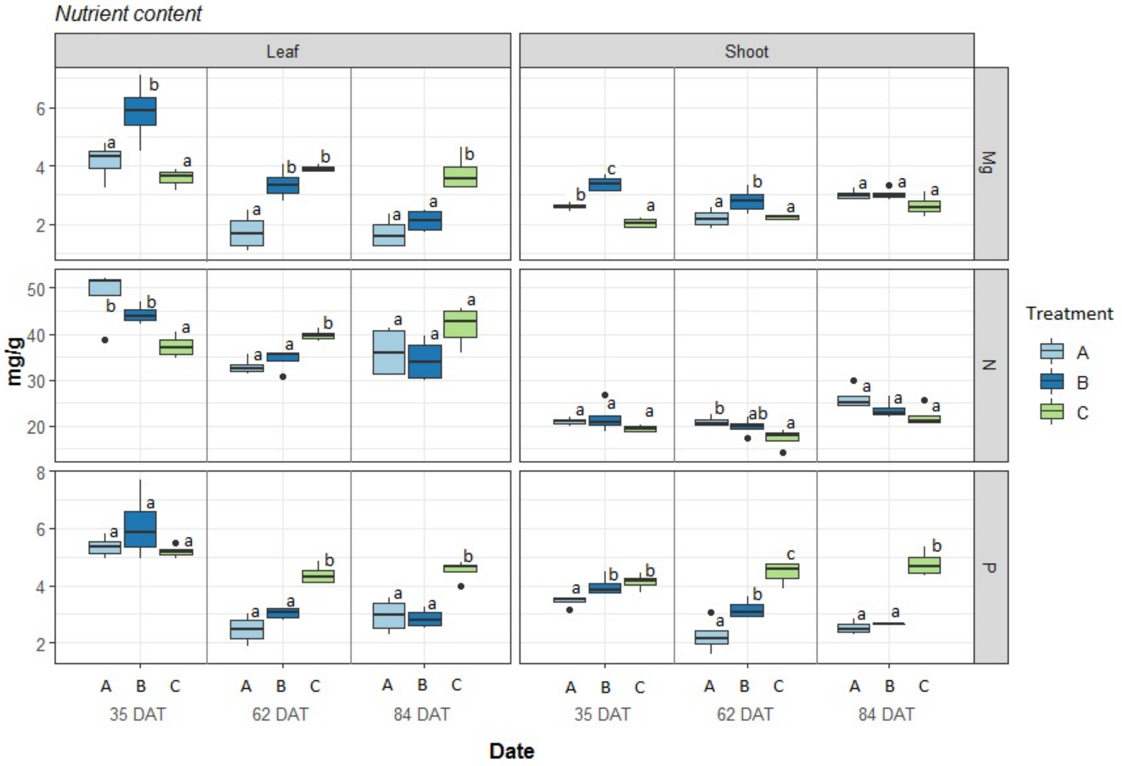

Figure 5. 4 Nutrient concentration in Phaseolus vulgaris leaves and shoots, expressed in mg/g. Boxplot (n=4) results given for three treatments A) 2g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution B) 5g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution B) 5g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution at three different time periods. 35 DAT, 62 DAT and 84 DAT. Significant differences (p<0.05) between treatments marked with different letters (a,b,c).

Figure A5.8 also indicates the total nutrient contents that are bound to the total biomass of the sampled plants. Here, it is apparent that treatment B, with more struvite, provided results that were between those of the other treatments. In the case of Mg at 35 DAT in leaves, treatment B showed levels as high as those for the control treatment, but while the latter remained constant over time, both A and B decreased. The same trend can be seen for P in both leaves and shoots. In the case of N, we can see an increase for all treatments that was faster for control C, while A and B increased in a similar fashion.

Finally, the NUEs obtained for all three treatments were 1.32 g/g, 0.55 g/g, and 0.29 g/g for A, B and C, respectively. The calculation methodology considered that N was in the soil, while the fixed nitrogen was not considered; therefore, the use efficiency can be very different for all three treatments.

5.4 Discussion

5.4.1 Plant growth and development

The results indicate that once the first production peak was reached, the control plants were more capable of continuing to produce flower buttons, while the inoculated and struvite-fertilized plants took longer. The relationship between their development and the amount of struvite given to the plants seems to be directly correlated. Generally, the biomass and bean pod production were higher in the control plants, while treatment B had a greater amount of struvite (5 g). Treatment A, with the lowest amount of struvite (2 g), was determined to be the treatment with the lowest growth and production rates. These findings agree with those presented in previous literature (Nanjareddy et al. 2014), for which lower KNO_3 availability was directly linked to a reduction in leaf and flower formation. This reduction also seemed to be related to the P and Mg availability over time due to struvite depletion, considering that the initial performance was similar in all three treatments.

By observing the SPAD measurements, the chlorophyll contents in all three treatments indicated that the N contents in the leaves were not strongly affected by the treatments but rather the LAI. Lower P availability resulted in a reduction in LAI as well as in overall plant growth, which was observed in treatments A and B. These differences were not as great as those for root weights (compared to the other plant organs), which have been reported in the previous literature to be less affected by P reductions (Rao, Fredeen, and Terry 2008; Chaudhary and Fujita 1998).

The lower nodule dry weights in the control treatment, compared to treatments A and B, have previously been reported in other studies, in which the nodule fresh and dry weights were found to be considerably reduced when inorganic NO_3^- fertilization was not restricted (Nanjareddy et al. 2014; C. K. Kontopoulou et al. 2017). On the other hand other authors report that the nodule number was not affected when exposing the crop to mineral and organic N sources but rather affected in size and weight (Pampana et al., 2017).

The increasing nodule numbers and weights throughout the experiment for the B treatment (with greater struvite), when compared to treatment A, confirm Kontopoulou's et al., (2017). findings that low initial N fertilization can restrict successful colonization. These differences, however, could also be due to the lower P amounts in treatment A compared to treatment B, since P is a limiting factor for successful nodulation (Olivera et al. 2004).

The lower bean productivities were similar to those in the study reported by Olivera et al., (2004), where bean production with lower P fertilization and Rhizobia inoculation turned out to be insufficient to reach production levels as high as those of conventionally fertilized beans. However, struvite fertilization seemed to increase the production of inoculated plants by up to 25% when treatments with 2 g and 5 g per plant were compared (59.35± 26.4 g plant-1 in treatment A and 74.23±23.0 g plant-1 in treatment B).

The effect of the struvite treatment to the increasing nodule number and dry weight indicates a successful nodulation and a greater fixation capacity with the given N. The slow release of N has presented itself as sufficient to increase the nodulation capacity as well as production capacity, without inhibiting N_2 fixation by the bacteria.

5.4.2 The effect on atmospheric N fixation capacity

The aboveground organs showed a clear pattern throughout the three measurements in terms of N assimilation. 15N enrichment levels in the A and B treatments were lower than that in the C treatment, which means that treatments A and B obtained most of their N from the atmosphere. This difference became even greater as time progressed and reflected a greater dependence on N_2 fixation in the A and B treatments. The differences between these two treatments (A and B) themselves can be due to the greater availability of struvite in the root medium and therefore a greater availability of initial N and P for treatment B than for treatment A (Olivera et al. 2004; C. K. Kontopoulou et al. 2017).

The δ15N reductions in treatments A and B over time corresponded to the availability of N provided by the struvite, assuming that it decreased over time. These reductions can be seen when the NO_3^- concentrations in the drained water were examined (see Table A5.3 of the Annex II). While initially greater amounts of N were detected in the leached water, by the end of the experiment, very low concentrations were seen. Therefore, while the δ15N values for the control treatment C remained constant over time (except in the nodules), the δ15N values for treatments A and B decreased progressively over time, which corresponded to the available N that was provided by struvite in the root medium.

This information indicates that a change in the source of N for the plants took place during the time span of 35 to 62 DAT. We can therefore assume that the availability of struvite and therefore N in the root medium was depleted mainly during that time, which forced the plants to rely on atmospheric N_2 fixation. The results obtained for %Ndfa also confirm that the levels of N_2 fixation increased over time in both treatments.

The indicated timespan of 35 to 63 DAT corresponds to the initial pod production of the plant, maximizing its nutritional needs. Therefore, a N and P source capable to uphold these needs during this stage must be contemplated. As seen in table A7 a major reduction of NO_3^- in the leachate water is found between day 35 and 49 for treatments A and B, indicating that the administered struvite was insufficient to further support a greater production.

The nodules appeared to be highly enriched with 15N during all three harvests, especially for treatments A and B. These results agree with previous literature that attributes this enrichment to the export of 15N-depleted ureides and import of 15N-enriched amino acids. Nevertheless, these values do not have a great effect on the total plant enrichment if the nodule biomass is considered (Shearer et al. 1986; Unkovich 2013; Craine et al. 2015).

The quantity of fixed nitrogen did not reach 40-50 kg/ha, which corresponds to low ranges, as reported in previous research (Farid and Navabi 2015). While treatment A, with less struvite, had higher BNF values at the first two sampling times, treatment B's BNF value had increased by the end of the experiment. These findings are in agreement with those mentioned in the literature, where BNF was found to be restricted in the presence of plant-available NO_3^-, and the BNF values increased during the mature stages of the plant with sufficient NO_3^- fertilization during early plant growth (Müller, Pereira, and Martin 1993; Hungria et al. 2006; C. K. Kontopoulou et al. 2017).

5.4.3 Plant health and Nitrogen assimilation

We conclude that all treatments had sufficient N since there were minimal differences in N concentrations in the shoots and leaves during plant growth and at the end of the experiment, as was also found by Kontopoulou et al., 2015. We consider that the lower yields were caused by the reduced uptake of K^+ and Mg^{2+} cations that was due to the electrochemical imbalance generated by the reduced presence of NO_3^- in the root medium. This idea is reinforced by the results shown in Figure A5.7 in the Annex II, where N gradually increased in all three treatments throughout the experiment, which indicated that fixation was taking place for treatments A and B. The values increased from less than 0.1 g N at 35 DAT up to 0.2 g at 84 DAT for both the A and B treatments.

The slight increase in K by the end of the experiment in the plants with less struvite (treatment A) was most likely due to the lower availability of the Mg_2^+ cation, which facilitated cation uptake (Horst Marschner 2002).

The declining N concentrations in the leachates led us to believe that the decreases in P and Mg concentrations in the aboveground organs could also be related to the depletion of struvite in the medium. This depletion occurred faster in treatment A than in treatment B, which was related to the initial amounts of struvite provided in each treatment (2 g and 5 g, respectively). It was seen that for the inoculated plants, greater amounts of P were needed to support symbiosis and nodulation, as has also been observed by other researchers (Savvas et al. 2018a; Ntatsi et al. 2018; Olivera et al. 2004). Whether the additional required P can be assimilated by adding more struvite to the substrate is worth pursuing in future studies.

These findings lead to the concept that a lack of N is not the limiting factor that is entirely responsible for the lower yields of the A and B treatments, but the limiting factor is instead the progressive loss of P and Mg in the root medium as well as the reduced cation uptake. When examining the NUEs that were obtained for all treatments, it is evident that plants with lower N inputs have greater use efficiency. This difference is very clear in treatment A with a three-times higher efficiency compared to treatment B. These differences can also be influenced by atmospheric N fixation, which was not provided as “Soil” N in the calculation tool (Weih 2014). A higher fixation capacity can therefore generate a higher NUE, which corresponds to our BNF results.

For production on larger-scale vertical farms, fertilization with struvite and Rhizobium seems possible, especially with greater struvite quantities, as in treatment B, which shows great compatibility with soil bacteria and produces larger yields than those crops fertilized with only 2 g of struvite. The initial fixation capacity of the control treatment and appearance of nodules during the first sampling stage indicate that nodulation could occur even with naturally occurring Rhizobium, which could simplify the fertilization process in soil-based agriculture. A limitation for larger-scale production could be providing precise applications of struvite in the root areas. As seen in this study, there were large production differences between the applications of 2 g and 5 g of struvite, and larger scale production in a vertical farm would mean precise weighting of the struvite amounts per plant and direct applications to each rhizosphere of each plant. As stated by Degryse et al., (2017), the location of this slow-releasing fertilizer can have a great impact on successful nutrient delivery to plants. Therefore, such applications could be highly time- and resource-consuming.

5.5 Conclusions

This work aimed to study the feasibility of using struvite and inoculation with Rhizobium bacteria as alternative Mg, N and P fertilization methods for vertical agriculture systems. For this purpose, we quantified the nitrogen sources, production, and evolution of the phenological stages of Phaseolus vulgaris with Rhizobium inoculation and different quantities of struvite and compared the results to a control treatment. Three main conclusions can be drawn from this study.

First, both alternative fertilizer treatments supplied the necessary nutrients to fulfill the plant cycle in soilless growing media. The lower yields compared to the control suggest the necessity for evaluating higher struvite quantities to fulfill plant requirements to achieve higher yields. Since previous experiments conducted with struvite suggested successful performance with 5 g/plant, its combination with the soil bacteria, Rhizobium, causes this quantity to be insufficient due to the additional nutritional requirements of the bacteria. This can be seen by the great reduction in yields of treatments A and B in comparison to the control.

Second, while nodulation seemed to not be hindered by nitrogen input through struvite in the root medium, it did not significantly improve it either, although BNF appeared to increase in the later stages for plants grown under the treatment with a greater initial quantity of struvite.

Third, the limiting factor for struvite-fertilized and rhizobia-inoculated treatments did not seem to be nitrogen, which was maintained at sufficient concentrations in the plants throughout the experiment, but rather was potassium, due to the lower uptake capacity that was caused by an electrochemical imbalance that was generated by the reduced presence of NO_3^- in the root medium as well as by magnesium and phosphorus, given that struvite depletion was reflected by the reduced plant nutrient concentrations over time.

An increase in the amount of applied struvite might be a solution for a more sustained phosphorus and magnesium supply for vertical agriculture but could also interfere with the nodulation capacity of the plants. Furthermore, we encourage the addition of nutrients in the form of anions to ensure the electrochemical balance in the root area in case NO_3^- is removed. In this sense, further studies should aim to determine the optimal struvite quantities for hydroponic bean production in combination with Rhizobium inoculation.

Part IV

Environmental approach to the use of struvite in hydroponic production

This chapter is based on the journal paper:

Extended use and optimization of struvite in hydroponic cultivation systems

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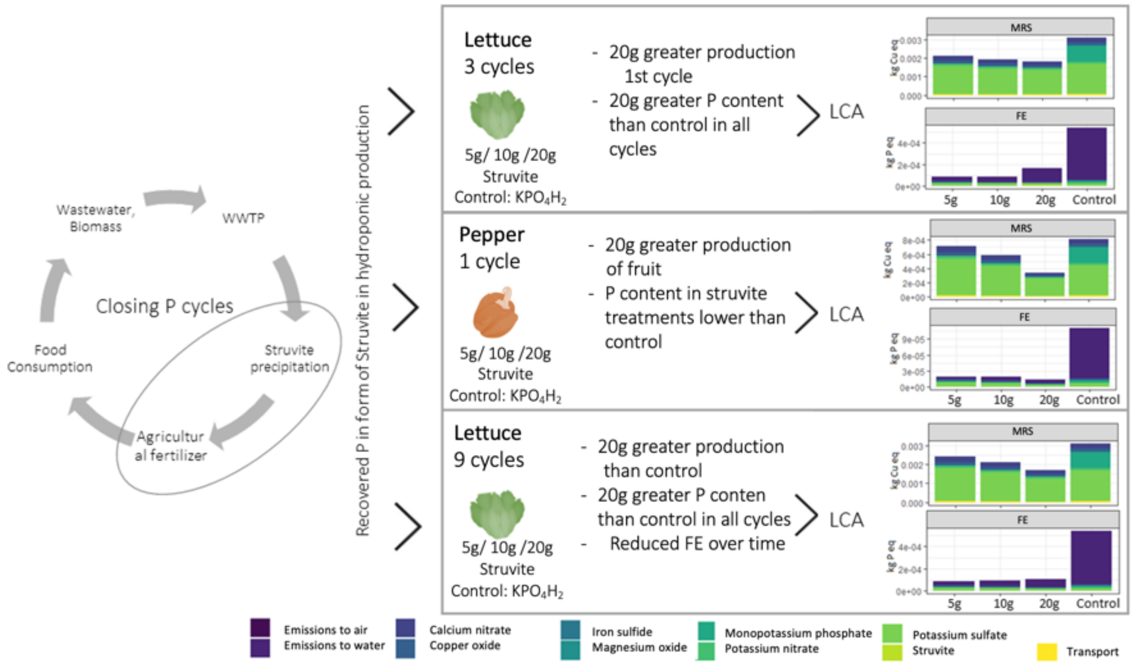
Chapter 6: Extended use and optimization of struvite in hydroponic cultivation systems

Abstract:

Hydroponic systems are an attractive form of urban agriculture due to their low weight load, inert substrate conditions, and overall better control of plant nutrition and growth. However, gaining urban food sovereignty cannot be at the cost of increasing environmental impacts, such as eutrophication and nonrenewable resource depletion, associated with phosphorus fertilizer use. Struvite, a wastewater byproduct, is a potential slow-releasing P source that can serve as a substitute for mineral P fertilizer. In this study, we explored the adequacy struvite in hydroponic systems, testing different quantities (5g, 10g and 20g per plant) compared with monopotassium phosphate for pepper and lettuce hydroponic production. The results show competitive productions for both crops with the use of struvite, especially during the first lettuce harvest (225.5g, 249.9g, 272.6g, and 250g for 5g, 10g, 20g and control, respectively) where a greater struvite dissolution was seen. Although all struvite treatments in pepper show low phosphorous content in the biomass, yields do not deviate greatly from the control (3.6kg, 4.3kg, 7.5kg and 5.3kg for 5g, 10g, 20g and control, respectively). The environmental performance of all lettuce treatments showed a reduction in all impact categories, especially freshwater eutrophication and mineral resource scarcity, except for marine eutrophication. All impact categories were reduced for all pepper treatments with 10g and 20g of struvite. When the results are extrapolated to a full year of production, we find that the slow dissolution of struvite can sustain competitive production with an initial 20g, with less impact in all categories except marine eutrophication



6.1 Introduction

Urban agriculture (UA) has the potential to significantly increase food security in cities (Toboso-Chavero *et al.*, 2019). Increasing green areas in urban landscapes have been gaining popularity, and with new technologies, greening and food production have been taken to building roofs, facades and even indoors (Despommier 2013; Specht et al. 2014; Appolloni et al. 2021). In particular, soilless agriculture is highly attractive in urban settings because of the reduced weight load on building structures, inert substrate conditions, and overall control of plant nutrition and health (Walters and Stoelzle Midden 2018; Vinci and Rapa 2019), as well as because it provides an alternative to contaminated soils. Soilless production can also be a beneficial system to improve water savings since a more controlled environment can be ensured with more accurate irrigation systems as well as water recirculation depending on the installation (Parada, Gabarrell, et al. 2021). As shown by Appolloni *et al.*, (2021) among 92 cases of urban agriculture identified from 2011 to 2019 a 46% produced with a soilless system. In addition to increasing food sovereignty, UA can promote biodiversity, CO_2 capture and pollination (Baró et al. 2014; Camps-Calvet et al. 2016; Ayers and Rehan 2021) but can also have negative effects, such as the extensive use of mineral and synthetic fertilizers (Sanjuan-Delmás et al. 2018). Soilless agriculture does not contemplate the addition of nutrients through the substrate but through a nutrient solution given with the irrigation system (El-Kazzaz 2017; Sambo et al. 2019). Previous work on life cycle assessment of hydroponic production systems shows that, while these fertilizers secure direct nutrient uptake by the plant, their production, extraction, use and disposal are known to have adverse consequences for the surrounding natural ecosystems (Sanjuan-Delmás et al. 2018; Rufí-

Salís et al. 2020; Rufí-Salís, Petit-Boix, Villalba, Sanjuan-Delmás, et al. 2020b). Alternatives for the fertilization in soilless agriculture are gaining interest being aquaponic systems most know for the efficient use of fish debris as nutrient for crop production reducing the potential impact of the production system (Graber and Junge 2009; Chen et al. 2020). However, aquaponic installations can entail a great initial investment and call for an additional production fish and therefore a greater need for maintenance and skill (Baganz et al. 2020).

The extraction of phosphate rock, the main source of phosphorus (P) for fertilizer use, has become a necessity for modern agriculture and is an indispensable nutrient for plant growth and animal feed (Rahman et al. 2014). However, phosphate rock deposits are limited due to the slow regeneration rate of their cycle compared to carbon or nitrogen, already generating supply shortages due to increasing prices and unequal distribution (Alewell et al. 2020). In recent decades, estimations have been made regarding imminent depletion if extraction continues at the present rate (Cordell, Drangert, and White 2009), which can be drastically shortened by soil erosion caused by unsustainable production practices (Alewell et al. 2020).

All the P extracted is mostly "lost" from agricultural lands and livestock management through surface and underground runoff (Rahman et al. 2014; Carpenter and Bennett 2011). This one-way nutrient flow has increased fourfold since preindustrial times (Alewell et al. 2020; Carpenter and Bennett 2011; Yi Liu et al. 2008; Villalba et al. 2008) and contributes to great ecosystem damage, such as eutrophication, especially in freshwater environments (Cordell and White 2014). While this ever-growing thread demands better management of P sources, there is possible recovery from an ongoing loss of nutrients occurring daily in our wastewater treatment plants (WWTPs) (Harder et al. 2019; Cordell and White 2013). These nutrients contained in wastewater sludge are disposed and managed mostly in complex processes due to the high content of heavy metals, pathogens and other compounds, making it a toxic residue (Panizza and Cerisola 2001; Rahman et al. 2014). While direct application of sewage sludge to the soil is practiced in several countries, it's application can entail a bad management of the soil, due to over application for P fertilization as well as the increase of pathogens and heavy metals into the soil, and the potential problematic of social acceptance of this practice due to the emitting odors (Pradel et al. 2020). Countries like Sweden have seen a reduction of the unwanted toxic metals since the 1970 and have started to regard sewage sludge as a potential nutrient provider and soil amendment, still only 20% of the sludge is applied in arable land (Kirchmann et al. 2017).

In recent decades, research has been conducted on the shift from a removal to a recovery approach in urban water cycles in terms of nutrients, not only for their further use in other production sectors but also to prevent their environmental damage in their disposal (Harder et al. 2019; Rufí-Salís et al. 2020).

One of the byproducts in these sewage treatment plants is magnesium ammonium phosphate ($MgNH_4PO_4.6H_2O$), a crystal commonly called MAP struvite or just struvite. Struvite is not a new material; its precipitation was first documented in Los Angeles (Borgerding 1972), but it was approached as a great problem since its precipitation occurs spontaneously at a 1:1:1 molar ratio of magnesium (Mg^{2+}), ammonium (NH_4^+) and phosphate (PO_4^{3-}) and under suitable pH conditions (8.5–9.5) (Le Corre et al. 2009; J. R. Buchanan, C. R. Mote, and R. B. Robinson 1994; Bouropoulos and Koutsoukos 2000). The purging of this uncontrolled struvite precipitation can be the cause of additional expenses due to damaged equipment that needs replacement or increased labor costs (Stratful, Scrimshaw, and Lester 2004). Since then, technologies aimed at struvite removal through induced precipitation in WWTPs have unveiled a product with great fertilizer potential.

The possibility of P recovery from wastewater in the form of the slow releasing fertilizer struvite has been deemed a solution not only for the supply of this nutrient in agriculture but also to avoid further phosphate rock extraction and an increase of P in wastewater streams and water cycles (Bradford-Hartke et al. 2015).

Struvite has already been tested in a variety of agricultural soils (Latifian, Liu, and Mattiassona 2012; B. Li et al. 2019), obtaining a wide range of results in crop growth and yield for a diverse range of plants, as shown by Ahmed *et al.*, (2018). Although results vary among different crops, a common perception is the slow solubility of granulated struvite; therefore, its most common application is in the form of pulverized struvite (Degryse et al. 2017a). Struvite dissolution has been tested before in continuously stirred pots and at controlled temperature, showing a lower dissolution rate than triple superphosphate (TSP) (Rech et al. 2018a; Ariyanto, Ang, and Sen 2017).

Further experiments testing struvite dissolution have demonstrated the importance of medium pH and plant root proximity (Degryse et al. 2017a; Talboys et al. 2016; Massey et al. 2009; Achat et al. 2014). This proximity can provide greater access to dissolution mechanisms from the plant that can make the P available, such as the exudation of organic acids to lower the pH of the soil or substrate (Rech et al. 2018a). This slow dissolution has been seen to hide crop development in early stages, corresponding to still early growth of the plant root.

Although information on the use of struvite as fertilizer is already available, its use in soilless agriculture is still scarce. Since hydroponic systems enable better control of plant nutrition but are designed to use chemical fertilizers, the use of struvite in exchange for the mineral phosphorous used in soilless agriculture has the potential to reduce its environmental burdens. First approaches have been made to identify the suitability of struvite in hydroponic production as well as it’s combination with biological amendments like rhizobium showing promising results (Arcas-Pilz *et al.*, 2021a). The P emissions to water seam to decrease significantly with the use of struvite compared to mineral derived P fertilizers while other studies reveal even greater productions with struvite (Arcas-pilz *et al.*, 2021b; Carreras-Sempere *et al.*, 2021). Previous work identifies the low solubility of struvite as a potential burden for plant uptake while it could also ensure reduced P emissions and longer productions over time.

With this knowledge on struvite the question arises if urban agriculture can directly profit from the nutrients generated in their immediate surroundings and strive for expansion without shifting the environmental damage to the generation of greater water and air emissions in urban settings. For this purpose, the production of crops for longer periods was proposed to understand the struvite nutrient discharge in time for short and long cycle crops.

The following experiment was performed to analyze the potential of struvite in providing P in hydroponic production systems by testing struvite solubility and uptake in granular form for two different crops: pepper plants (a highly P-demanding crop with a long growth cycle) and lettuce (shorter cycle). In addition to the dissolution and uptake analysis, this work also focuses on the nutrient discharge into water, covering the potential reduction of P loss into water bodies. To express the environmental burden of the struvite fertilizer and the mineral phosphate fertilizer, the collected information was used to perform an environmental analysis for the produced vegetables using the life cycle analysis assessment (LCA) to determine the environmental footprint during the timeframe of this experiment. The assessment was extrapolated to a one-year production period to simulate these fertilization techniques for a longer time to maximize the P available in the struvite placed in the substrate.

6.2 Materials and methods

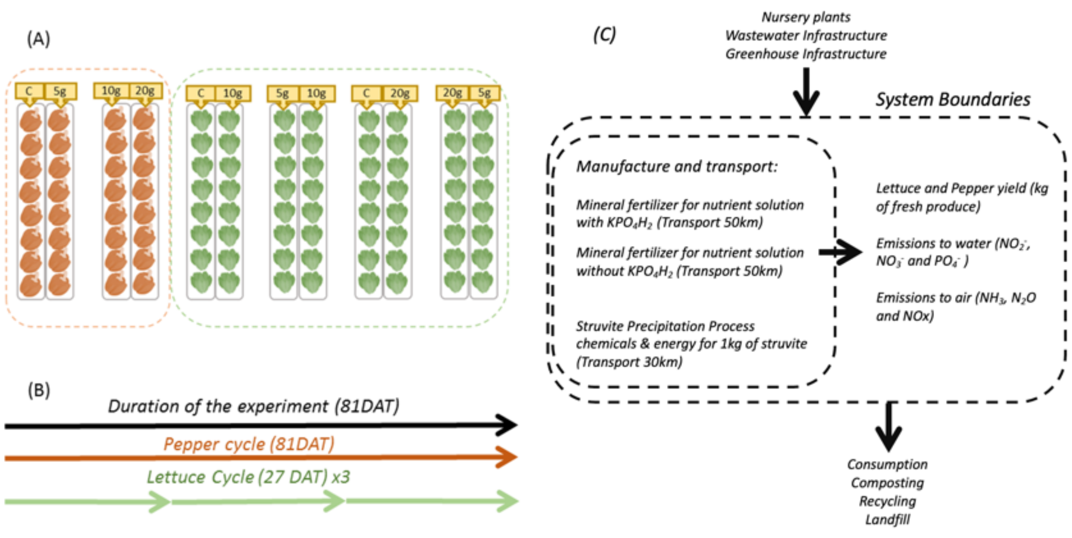
6.2.1 ICTA-UAB greenhouse

The following experiment was performed in the ICTA-UAB integrated rooftop greenhouse in the Universitat Autònoma in Barcelona from June to September (2020). The production system was hydroponic using individual pots and perlite as the substrate (see picture A 6.1 in the Annex II). Nutrients were given through fertigation, mixing concentrated nutrient solutions (NS) with harvested and filtered rainwater (RW) in a proportion of 1:100 (NS:RW) through a drip irrigation system with a 2 L h^{-1} flow. To ensure sufficient irrigation, the amount of drained water was determined daily and maintained ca. ~30% of the incoming irrigation with increasing or decreasing irrigation time. The growing frame consisted of 4 m x 0.5 m wide tables with the capacity to grow two crop lines. Between tables, a distance of 1 m was given from plant to plant.

6.2.2 Crop growth and treatments

In this experiment, the determination of struvite dissolution and uptake was carried out on two different crops, Lettuce (*Lactuca sativa* L.) and Pepper (*Capsicum annum* L.). The lettuce and pepper plants were obtained from a nearby nursery in early growth stage with the first growth of the true leaf's (about 7cm tall for lettuce and about 10cm tall for pepper), which were then planted in the greenhouse inside perlite filled pots. The perlite was previously wettened with water to ensure a better handling and provide moisture to the plants during the transplanting process. The treatments were arranged in rows to facilitate irrigation and leachate sampling as well as drainage measurement. Each row represents a treatment with a different struvite quantity ranging from 5 g (named 5LE for lettuce, 5P for pepper) and 10 g (named 10LE for lettuce, 10P for pepper) to 20 g (named 20LE for lettuce, 20P for pepper), including a control treatment (named CLE for lettuce, CP for pepper). All crops fertilized with struvite received P-deficient NS, while the control treatment was irrigated with conventional NS (NS specified in Annex II Table A6.2). To maximize the contact between struvite and the plant, the granules were placed close to the root once the seedlings were transplanted into the pots.

In the case of the lettuce crop, each treatment consisted of 28 plants arranged in two rows, making two replicates of 14 lettuce plants distributed randomly for each treatment, while for the pepper crop, eight plants were arranged in simple rows (Figure 6.1).



- - -Lettuce

- - - Pepper

Figure 6. 1 Experimental outline (A) shows the distribution of the pepper and lettuce treatments along the laboratory greenhouse (C= control treatment). The experimental timeline (B) shows the total duration of the experiment and the duration of the pepper and lettuce cycles (DAT = days after transplanting). The system boundaries for the environmental analysis (C) show the scope of the analysis within the dotted line.

During the experiment, several sensors were used to record the climatic conditions inside the greenhouse. Humidity and temperature were recorded with a CS215 Campbell Scientific, and radiation was recorded with a pyranometer (L202 by Hukseflux) (Annex II Figure A6.3).

All water flows were measured daily throughout the experiment. The irrigated water was quantified through water meters installed in the irrigation system, while the volume of drained water was measured on buckets at the end of each crop line. Samples of incoming and outgoing water were taken three times a week for each treatment. To ensure good irrigation conditions, the pH and EC for these water samples were measured immediately after collecting daily samples (Annex II Figures A6.4 and A6.5).

The short cycles of lettuce lasted a total of 27 days after transplanting (DAT). Once the plants were harvested, a new seedling was planted in the same pot (14∅ I 13 cm high for lettuce and 25∅ I 20 cm high for pepper). For each treatment, two pods were removed after each cycle to take substrate samples. Pepper plants were planted parallel to the first lettuce crop until the harvest of the third lettuce cycle (81 DAT), as shown in Figure 6.1. Pepper fruit harvests were made weekly once production started, accounting for a total of four harvests before finalizing the experiment. On the other hand, lettuce yields were weighed after each cycle, generating three harvests.

To obtain a more accurate understanding of the possible yield variations among lettuce samplings, 15 pots of each lettuce treatment were labeled with a reference letter (from A to O) maintained throughout the experiment. Relative yields obtained could then be traced back to the corresponding pot, therefore allowing precise appreciation of possible production changes.

The yield produced by the pepper plants was obtained in four harvests. The total fruit weight recorded for each treatment was obtained as the sum of all four harvests. The number of fruits produced in each harvest was also accounted for to estimate the weight per fruit.

6.2.3 Plant sampling methods

For each lettuce cycle, the fresh and dry weights of each plant were measured. After harvest, a random sample of four plants for each treatment was dried at 60 °C until a constant weight was achieved (ca. 7 days).

At the end of the experiment, all pepper plants were harvested and weighed. The pepper plants were separated into leaves, stems and roots, removing all flowers. Additionally, we quantified the leaf weight, number, area index (LAI), stem weight and length. The latter was measured accounting for the main central stem without considering ramifications. However, these ramifications were considered when weighing the stem. The LAI was obtained using the scanned images of 25% of the leaf fresh weight and further processed with a Python script (as indicated in the Annex II) (relating the number of pixels per leaf area) to give the total area of each pepper plant (Ribalta-Pizarro, Muñoz, and Munné-Bosch 2021; Garrido et al. 2020). A sample of five plants per treatment was also dried in an oven at 60 °C until reaching a constant dry weight (ca. 7 days). In the case of the fruit, a sample of pepper pods was taken from each treatment after every fruit harvesting. Fresh and dry weights were measured following the same procedure as for the plant biomass.

Before drying all plant biomass was rinsed with Elix water and dried to avoid any potential external contamination.

Once the dry weights for lettuce, pepper fruit and biomass were quantified, the samples were ground for further analysis, consisting of digestion with concentrated HNO_3 in a single reaction chamber microwave to be then analyzed for total P concentration using optical spectrometry (ICP-OES).

Substrate samples were transferred to a polypropylene sampling pot after thoroughly mixing the perlite from the pot in a clean container. For each treatment, two samples were taken at the end of each of the three production cycles. After taking the perlite samples, they were placed on aluminum trays and dried at 60 °C for 72 hours. Once dry, the samples were weighed and ground for total P determination using the method detailed before.

Irrigation and leached water samples were proportionally unified into weekly samples considering a volume ratio. These samples were filtered through a 0.22 mm filter and analyzed with ionic chromatography (ICS-200) for nitrite and nitrate contents. The Mg and P contents in the water samples were analyzed with ICP-OES (Perkin-Elmer Optima 4300DV).

To calculate the struvite dissolution rate, the amount of P found in the plant and leachate was calculated for each treatment (for lettuce, each cycle was taken separately). The obtained quantity was assumed to be the dissolved P from the struvite and divided by the liters irrigated to the crop. The dissolution rate was then plotted against the initial struvite amount given to the plant. For the second and third lettuce cycles, the initial struvite was assumed to be the remaining struvite in the perlite after the previous crop cycle.

6.2.4 Environmental Assessment

The Life Cycle Assessment (LCA) used to determine the environmental impact of the irrigation system followed the ISO norms 14040 and 14044 (ISO 2006).

6.2.4.1 Goal and Scope and Inventory

An environmental assessment of the fertilization method was made, comparing the environmental load to produce 1 kg of fresh lettuce and pepper pods considering the incoming fertilizers and outgoing emissions to water and air, as shown in Figure 6.1 (C). The life cycle assessment (LCA) tool was used to determine the environmental impact of fertilization for all treatments, which was calculated with Simapro 9.1 software, using the Ecoinvent 3.7 database to account for the background environmental information and the ReCiPe midpoint impact assessment method (Huijbregts et al. 2017). The scope of this attributional LCA was defined as cradle to grave since the production, transport, use and disposal stages for the different fertilizers were considered. On the other hand, the greenhouse infrastructure and production system were not included in the system boundaries, focusing only on the impact of the use of struvite as fertilizer.

6.2.4.2 Life Cycle inventory

The inventory for the LCA was comprised by the obtained data from the experiment. The fertilizer applied was obtained from the nutrient solution and irrigation amount controlled daily through the water meters installed in the irrigation system. The incoming irrigation as well as the leachate water was analyzed to obtain the N and P emitted to water (in the form of NO_2^-, NO_3^- and PO_4^-). From the incoming irrigation the calculation of the N emissions to air in the form of ammonia (NH_3), nitrous oxide (N_2O) and nitrogen oxides (NOx) following the emission factors established by Montero *et al.*, (2011).

The generation of struvite was accounted for in the environmental assessment based on the production of the commercial house Ostara®. For the production of 1 kg of struvite, the additional chemical input in the precipitation stage was 0.4239 kg MgCl, 0.766 kg NaOH for pH stabilization and 0.523 kWh energy applied for mixing and aeration (Amann et al. 2018). Impacts related to wastewater treatment, such as an improvement of the effluent or the additional technology required for P removal to the sludge line, were excluded from the system boundaries of this LCA. The transport for all fertilizers accounted for 50 km from the greenhouse. The struvite transport, on the other hand, was estimated to be 30 km, which corresponds to the approximate distance to the two nearest WWTPs of the city (EDAR Besós and EDAR Llobregat), although they currently are not producing or selling struvite.

The environmental assessment was made for a single plant pot, taking into account its fertilization and production. These results can then be further extrapolated to greater production. The detailed inventory and processes can be seen in the Annex II (Table A6.16)

6.2.4.3 Environmental Impact Assessment

The impact categories selected were global warming (GW), terrestrial acidification (TA), freshwater eutrophication (FE), marine eutrophication (ME) and mineral resource scarcity (MRS). These selections were based on the author's expertise and previous literature focusing on the impacts of fertilizers in soilless systems and the use of struvite (Brentrup et al. 2004; Sanjuan-Delmás et al. 2018; Rufí-Salís et al. 2020).

Global warming, expressed in kg CO_2 eq. was chosen due to the documented relevance of greenhouse gas emissions from the production of fertilizers, as well as their transport, and due to the direct emissions occurring at the plant level (Chatzisymeon, Foteinis, and Borthwick 2017; Hasler et al. 2015). This case is especially true for nitrous oxide (N_2O). Thus, we considered the additional nitrogen given in the form of ammonia through struvite. The proportional fraction of ammonia released in each treatment, determined through direct measurement in the leachates, was considered when calculating the emission factor as well as the nitrogen given in the irrigation. For the same reason, TA (kg SO_2 eq.) was also chosen to reflect the direct emissions due to the application of ammonia as well as other acidifying agents generated during transportation and manufacturing of fertilizers (Hasler et al. 2015). FE (kg N eq.) and ME (kg P eq.) have been regarded as the most relevant impact categories when analyzing fertilization methodologies, especially considering nitrogen and phosphorous (Hasler et al. 2015; Chatzisymeon, Foteinis, and Borthwick 2017; Vatsanidou et al. 2020). These impact categories are especially relevant in this study since slow struvite dissolution can provide insight into the possible reduction of P leaching into fresh and marine waterbodies, and again, the addition of N through struvite can also be reflected in the leachate quantities. FRS (kg oil eq.) was added as a relevant impact category to reflect fossil energy-related emissions that could arise due to struvite precipitation and transport compared to mineral P. Finally, MRS (kg Cu eq.) was chosen to reflect the extraction of finite mineral resources, especially focused on phosphate rock extraction *versus* the recycling and reuse of phosphorous in the form of struvite.

6.2.5 Statistical analysis

Shapiro–Wilk test $p > 0.05$ was used to test the normality of the data, while homogeneity of variance was tested with Levene's test $p > 0.05$. Duncan's multiple range test was used to assess the statistical significance of treatments when parametric criteria were validated. For nonparametric data, the Kruskal–Wallis test was used. The significance between the treatments is marked with different letters (a, b and c).

6.3 Results

6.3.1 Crop growth and yield production

The resulting productions for the three lettuce cycles can be observed in Table 6.1. Here, we can appreciate the average yields obtained for all three harvests and all treatments for their fresh and dry weights. Further information on the specific production within the marked pots (A to O) can be seen in Table A6.6 in the Annex II.

Table 6. 1 Average yield (g/ per plant) expressed as fresh weight (FW) and dry weight (DW) for all three harvests at 27 DAT, 54 DAT and 81 DAT. Significant differences (p< 0.05) between treatments marked with different letter (a,b,c)

Average Yield (g) FW	1st Harvest		2nd Harvest		3rd Harvest	
	FW	DW	FW	DW	FW	DW
5LE	$225.5^{c} \pm 43.2$	$9.1^{c} \pm 1.5$	$224.9^{b} \pm 52.1$	$9.8^{b} \pm 2.2$	$133.3^{a} \pm 28.1$	$5.5^{a} \pm 1.6$
10LE	$249.9^{b} \pm 35.2$	$10.1^{b} \pm 1.4$	$251.7^{a} \pm 56.9$	$10.9^{a} \pm 2.4$	$139.8^{a} \pm 31.2$	$5.8^{a} \pm 1.6$
20LE	$272.6^{a} \pm 32.1$	$10.9^{a} \pm 1.3$	$261.4^{a} \pm 59.2$	$11.4^{a} \pm 2.6$	$149.6^{a} \pm 56.4$	$5.8^{a} \pm 3.0$
CLE	$250.0^{b} \pm 26.6$	$10.1^{b} \pm 1.1$	$279.0^{a} \pm 33.5$	$12.1^{a} \pm 1.5$	$137.8^{a} \pm 35.7$	$5.4^{a} \pm 2.0$

We identified a general decrease in yield during the third harvest, most likely due to a remarkable decrease in the overall temperature during 54 DAT and 81 DAT in contrast to the previous crop cycles (1 DAT to 54 DAT). This variation in the climatic conditions can be observed in Figure A6.2 in the Annex II with the recordings of humidity, radiation, and temperature during all three cycles. While temperatures still ranged between 20 °C and 25 °C, the sudden change in comparison to the previous two crop cycles may have caused a delay in lettuce growth.

While no great differences can be seen in the overall yield of our lettuce cycles, the close monitoring of our pots can give us the variability of the obtained yield for the lettuces grown with the same initial struvite. This finding means that from the same pot, we can monitor the obtained yield in all three cycles. Table A6.7 in the Annex II provides us with such information showing a general decline in production, with the most acute decrease in yield in the 5LE treatment with a

-11% difference between the first harvest and the second. On the other hand, the decline for treatments 10LE and 20LE was less pronounced, with -2% for both.

In the case of pepper plant growth and production, tables 6.2 and 6.3 provide insight into the differences spotted between treatments. Table 6.2 provides the main measurements made of the pepper plants at the end of the experiment (81 DAT).

*Table 6. 2 : Mean values of pepper plant biomass measurements made in the 81 DAT. Stem weight and Leaf weight given in g for their fresh weight (FW) and dry weight (DW). *The LAI calculated in cm2. Significant differences (p< 0.05) between treatments marked with different letters (a,b,c).*

TREATMENT	Stem weight (g/plant)		Leaf weight (g/plant)		Stem length (cm)	Leaf number (nr)	LAI*
	FW	DW	FW	DW			
5P	169.3^{a}	32.2^{a}	131.9^{a}	21.2^{a}	99.1^{ab}	110.1^{a}	2.5^{a}
10P	184.0^{a}	35.2^{a}	166.7^{ab}	26.0^{ab}	107.1^{b}	127.3^{ab}	3.3^{a}
20P	204.1^{a}	40.9^{a}	195.2^{bc}	31.4^{b}	96.0^{ab}	138.9^{ab}	3.8^{a}
CP	220.1^{a}	43.9^{a}	236.2^{c}	35.5^{b}	90.0^{a}	156.3^{b}	4.1^{a}

While no significant differences were seen for the stem weight, an increase in the fresh and dry weight was observed with the increasing amount of struvite applied. The same increase was noted for the leaf weight, number, and LAI, showing significant differences in all but the latter. The control treatment generally showed greater values in all measurements apart from one, the plant stem length.

Table 6. 3 total yield obtained in four pepper fruit harvests (55 DAT, 62 DAT, 72 DAT and 81 DAT) for each treatment. The yield given in g while an average fruit weight given with g/ fruit.

Harvest	*Treatment*	*Total Fruit Weight (g)*	*Fruit number (nr)*	*Weight/number (g/ fruit)*
1ST HARVEST (55 DAT)	*5P*	1261.9	22	57.4
	10P	1402.7	21	66.8
	20P	2155.5	25	86.2
	CP	1479.0	22	67.2
2ND HARVEST (62 DAT)	*5P*	911.0	18	50.6
	10P	1240.0	19	65.3
	20P	1833.4	28	65.5
	CP	1597.0	21	76.0
3RD HARVEST (72 DAT)	*5P*	528.0	9	58.7
	10P	759.0	10	75.9
	20P	1649.2	18	91.6
	CP	1046.0	13	80.5
4TH HARVEST (81 DAT)	*5P*	860.0	20	43.0
	10P	940.0	21	44.8
	20P	1860.0	29	64.1
	CP	1225.0	19	64.5
TOTAL	*5P*	*3560.9*	*69*	*51.6*
	10P	*4341.7*	*71*	*61.2*
	20P	*7498.1*	*100*	*75.0*
	CP	*5347*	*75*	*71.3*

The yield produced by the pepper plants (table 6.3) showed a greater total weight for the 20P treatment. While the total number of fruits was also higher for the 20P treatment, the weight per fruit did not differ greatly from that of the other treatments.

6.3.2 Nutrient content in plant biomass and substrate

The results shown in Figure 6.2 depict the P content in the lettuce crop after 27 days of growing in the greenhouse for all treatments. The amount of P found in the lettuce biomass is directly related to the amount of struvite given, being lowest for the 5LE treatment followed by the 10 L and finally 20LE treatments. The amounts of P found in the 5LE and 10LE treatments decrease noticeably over time in the second and third cycles, while the 20LE treatment does not experience a great reduction during the second cycle but rather on the third cycle. It is important to point out that the results found for the second crop cycle show a much greater variability than the first and third ones.

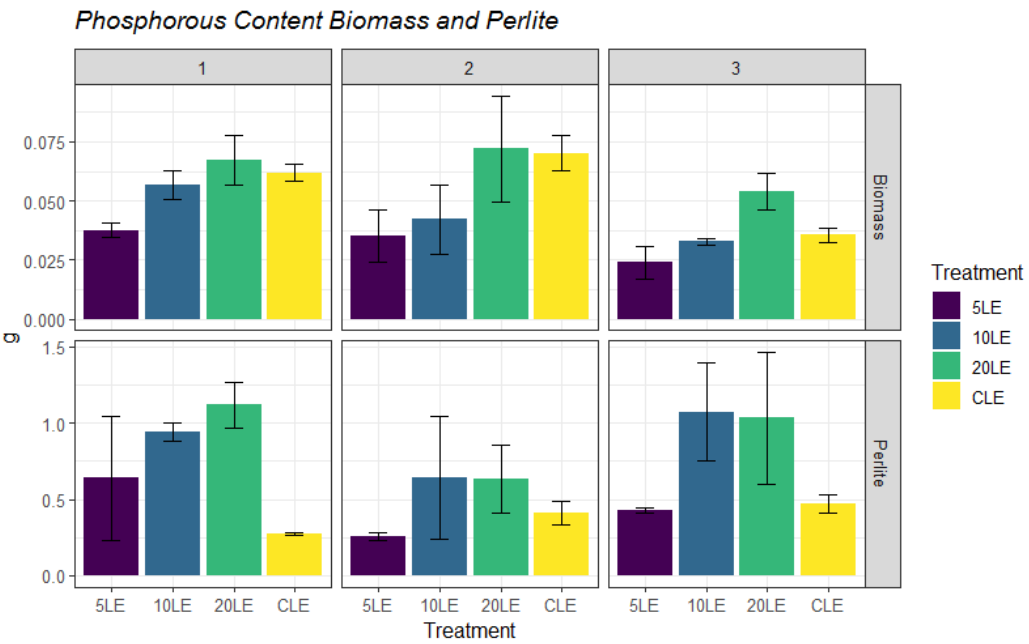


Figure 6. 2 Amount of P (g) accumulated in the lettuce biomass(above) and perlite (below) for treatments 5LE, 10LE, 20LE and CLE in all three harvests (1, 2, 3) given in total g per plant (in the case of perlite g per pot).

The remaining struvite content in the perlite and therefore P remaining in the substrate were analyzed and plotted in figure 6.2. Here, we can appreciate a great difference between the struvite fertilization treatments and the control, since the nutrient content in the perlite slowly increases over time for the latter, while the P content in the struvite treatments fluctuates and slowly decreases due to its dissolution. Here, again, a much greater variability in the results was observed for the second cycle.

Figure 6.3 depicts the P content in pepper biomass, fruit and perlite, showing great variation between struvite fertilization treatments and the control. While our treatments showed a low P content of 1.2 mg/g in leaves and 0.7–0.8 mg/g in the plant stem, giving ranges of 0.02 to 0.03 g of P in the total dry biomass, the control treatment showed values within adequate ranges of 2.1 mg/g (0.08 g of P in the total dry biomass). The amount of P in the harvested fruits reveals the differences between treatments based on the great mobility within the plant. Fruits are an ultimate sink of the phosphorous content in plants, and this result is reflected with a very clear relation to the struvite treatment. The great variability seen in these results derives from the great difference found between harvests within the same treatment, while the first pepper fruit harvest contained greater P concentrations, the third suffered a great reduction for all treatments, even the control (Annex II Figure A6.11). Finally, the amount of P found in perlite responds to the initially given struvite.

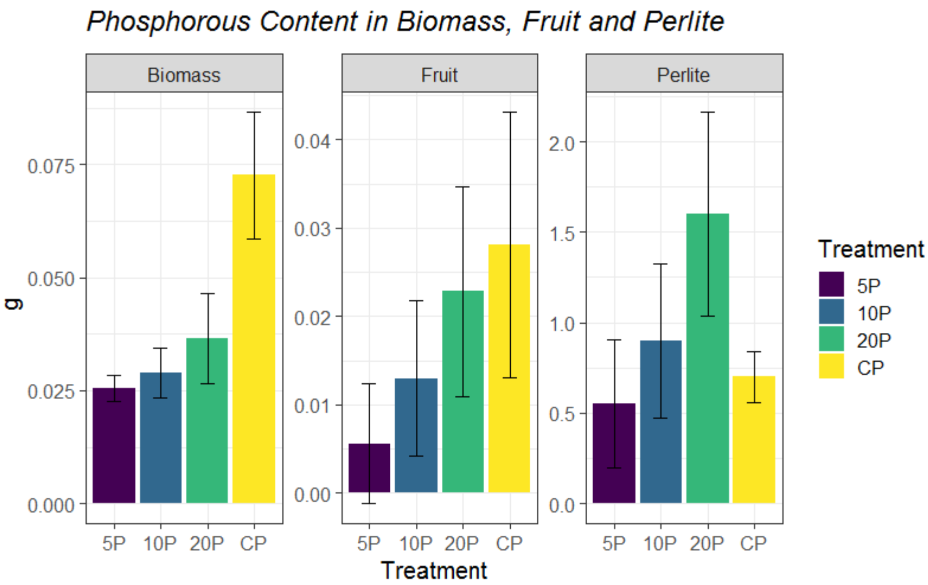


Figure 6. 3 P amount (g) in pepper biomass, fruit, and perlite for treatments 5P, 10P, 20P and CP at 81 DAT given in total g per plant.

6.3.3 Phosphorous content in the leachate

The resulting phosphorous concentrations found in the leachates were calculated for the total outgoing water weekly per plant, generating the patterns found in figure 6.4. The accumulation of P in the leachates for the lettuce and pepper crops can be seen in the Annex II Figure A6.10

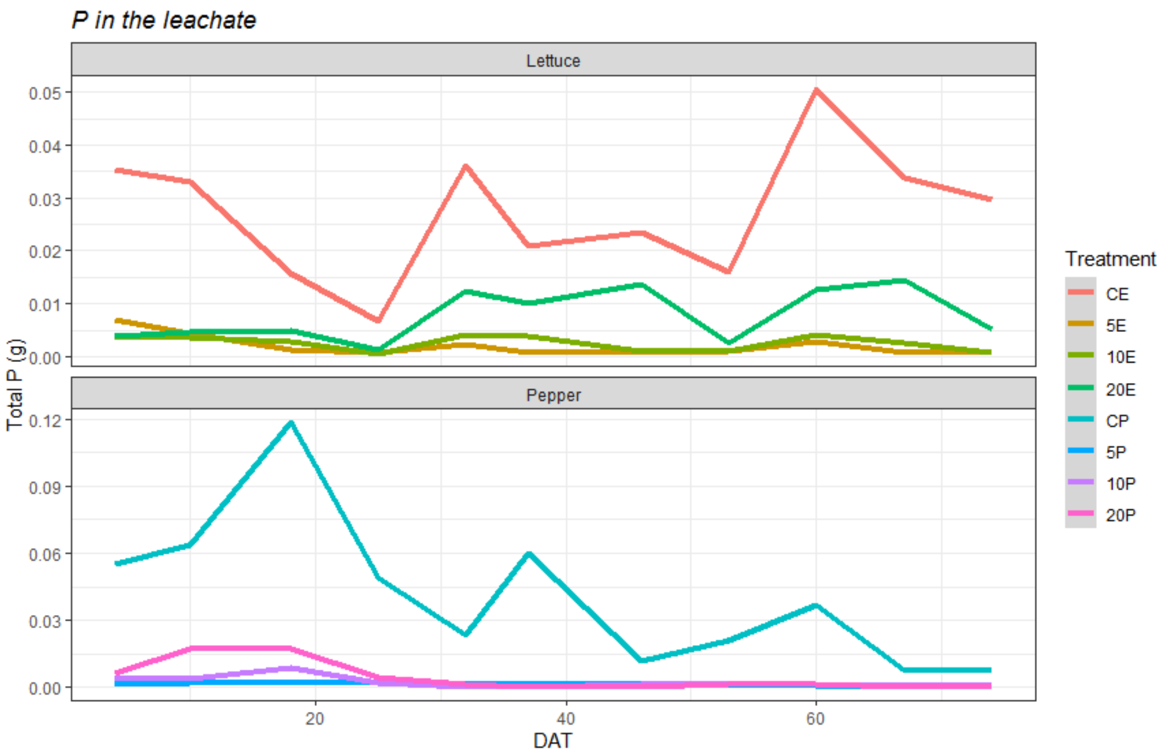


Figure 6. 4 Total phosphorous (Total g) found in the leachate water from 4 DAT to 74 DAT in Lettuce and Pepper crops for treatments with 5g, 10g and 20g of struvite as well as Control treatments (CP and CLE) irrigated with KPO_4H_2.

The results for the lettuce crop show the discharge of phosphorous during all three cycles, recognizing a clear pattern before and after each harvest. This pattern was highly noticeable for the CLE treatment, where the phosphorous content in the leachates decreased with the growth of the plant and rose once the plant was harvested and replaced with a seedling. This same pattern can be observed for all struvite fertilization treatments for lettuce, finding greater amounts for 20LE and less for 10LE and 5LE.

The phosphorous content in water, on the other hand, differs greatly when observing the CP and the 5P, 10P and 20P treatments. The biomass growth, climatic conditions and subsequent irrigation amount define the loss of phosphorous in the CP treatments, showing an overall decrease in the concentration with a peak at approximately Day 37 after transplanting. All treatments with struvite showed very low concentrations in the leachates, especially after 20 DAT.

6.3.4 Phosphorous balance

The results obtained in the previous sections enable us to generate the nutrient balance for P during these cycles for all treatments. This understanding helps us estimate the P flows into the plant, substrate and water. These nutrient balances were calculated for the P flows in the lettuce and pepper crops (Table 6.4) and averaged to obtain data for one plant (Figures A6.8 and A6.9 in the Annex II). In addition, the water balances per plant are given in Figures A6.14 and A6.15 in the Annex II. The nutrient balance is subjected to potential inaccuracies given through the sampling of substrate, water and biomass and the generation of mean values for all samples generating approximate values close to 100%.

Table 6. 4 Phosphorous balance per plant for the lettuce and pepper crop for treatments 5LE, 10LE, 20LE, CLE, 5P, 10P, 20P and CP. Initial P given through struvite and NS. Biomass 1, 2 and 3 corresponding to the 3 Lettuce cycles at 27 DAT, 54 DAT and 81 DAT. Pepper total biomass corresponds to total P found in Fruit Production and Biomass. For the biomass the amount of P from the root was also included with the root DW and root phosphorous content obtained from literature* (Xu et al. 2004; Pereira-Dias et al. 2018; Erel et al. 2019).

TREATMENT	INITIAL P	BIOMASS 1*		BIOMASS 2*		BIOMASS 3*		TOTAL BIOMASS		PERLITE		LEACHATES		BALANCE
LETTUCE	Struvite /NS g	g	%	g	%	g	%	g	%	g	%	g	%	%
5LE	0.625	0.047	8	0.047	8	0.035	6	0.130	21	0.443	71	0.022	4	95
10LE	1.25	0.064	5	0.050	4	0.042	3	0.157	13	0.885	71	0.028	2	86
20LE	2.5	0.077	3	0.079	3	0.064	3	0.221	9	1.196	48	0.085	3	60
CLE	1.049	0.071	7	0.081	8	0.044	4	0.196	19	0.384	37	0.318	30	86
	INITIAL P	PRODUCTION		BIOMASS*				TOTAL BIOMASS		PERLITE		LEACHATES		BALANCE
PEPPER	Struvite /NS g	g	%	g		%		g	%	g	%	g	%	%
5P	0.625	0.029	5	0.069		8		0.099	12	0.561	90	0.014	2	104
10P	1.25	0.055	4	0.076		4		0.130	9	0.904	72	0.026	2	83
20P	2.5	0.086	3	0.092		3		0.178	6	1.602	64	0.051	2	72
CP	2.595	0.106	4	0.189		7		0.295	11	0.709	27	0.501	19	57

The balance for lettuce gives an overall picture of the obtained results of the phosphorous flows into the plant biomass as well as leachates. Compared to the control treatment, the phosphorous flow into the outgoing water was approximately 10 to 14 times lower for the 10LE and 5LE treatments, respectively, while the flow into the plant biomass remained similar. The remaining phosphorous in perlite remained high in the 5LE and 10LE treatments, while more than half was reduced in the 20LE treatment. We also appreciate an accumulation of P in the perlite of the CLE treatment.

For pepper, the biomass flows were divided between the fruits produced and the generated biomass on the day the plants were cut and weighed. Here, we can appreciate the great quantity of phosphorous found in the pepper fruits, which equals the total phosphorous found in the plant leaves and stem. The total biomass showed a great difference between the CP treatment and the struvite fertilization treatments, revealing a much greater P content in the control. Due to the greater irrigation needs of pepper plants compared to lettuce plants, the CP treatment received an overall greater amount of P through irrigation compared to the CLE treatments. Therefore, although the P in the perlite and leachates is lower in CP than in the CLE treatments in terms of percentage, the absolute amounts are greater. In the case of the pepper plants fertilized with struvite, the P in the leached water was similar and even smaller than the amount found in the lettuce crop. The outgoing P in the leachates of the pepper plants was 10, 19 and even 35 times lower than that in the control treatment (CP) for the 20P, 10P and 5P treatments, respectively.

The calculated dissolution rates for the applied struvite in lettuce and pepper are shown in figure 6.5. The struvite dissolution was estimated by the P contained in the water leachates as well as P in the plant biomass. This dissolution has a direct impact on the P uptake by the plant that was estimated as the P contained in the P biomass. The results for lettuce show greater dissolution with a greater initial amount of struvite. The dissolution of the struvite was also higher during the first lettuce cycle (marked with number 1 in the figure), showing smaller differences between the second and third cycles (marked with 2 and 3, respectively). The dissolution rate found in the pepper crop was smaller than that in the lettuce crop but followed the same pattern as seen before, with greater dissolution with higher amounts of struvite.

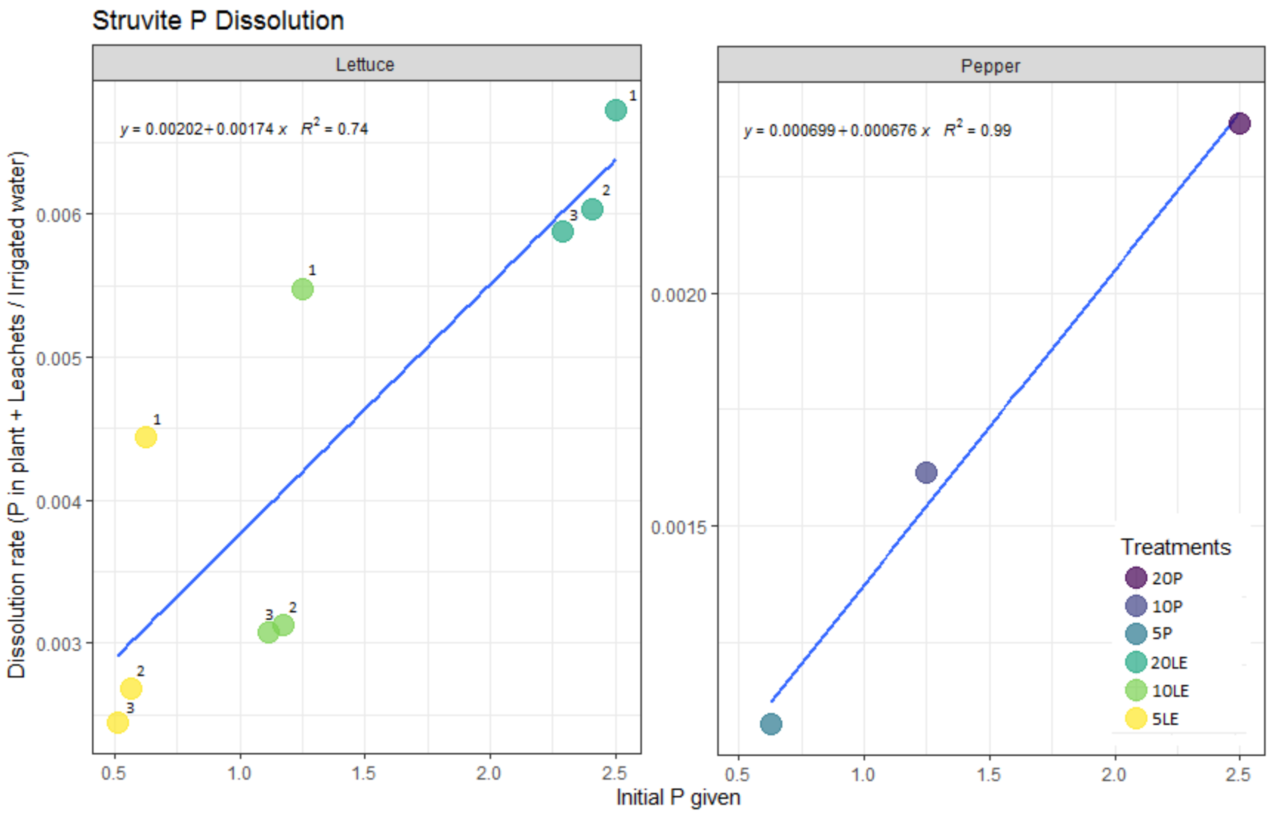


Figure 6. 5 Correlation of the dissolution rate of the struvite P and initial P given in Lettuce and Pepper for treatments 5LE, 10LE, 20LE, 5P, 10P and 20P. For lettuce all three cycles are taken in account, marked as 1, 2 and 3 respectively.

6.3.5 Life Cycle Assessment

Figure 6.6 shows the results for lettuce, and figure 6.7 shows the results for the environmental assessment of the fertilization treatments. Since only the fertilization of the crops was considered for the analysis (figure 6.1), all differences will be related to the use of struvite instead of monopotassium phosphate (MKP) in the form of KPO_4H_2, leaving out the laboratory infrastructure and auxiliary equipment, as well as the end-of-life processes.

The obtained results for six out of seven impact categories show that fertilization with struvite has lower impacts than the control, and for the cases of ET, MRS, FRS and GW, impacts are also reduced as we increase the amount of struvite applied. In terms of eutrophication, FE, which is directly related to the emissions to water, had the greatest impact on the control irrigated with mineral P, followed by 20LE, which was the treatment with the highest quantity of struvite per plant.

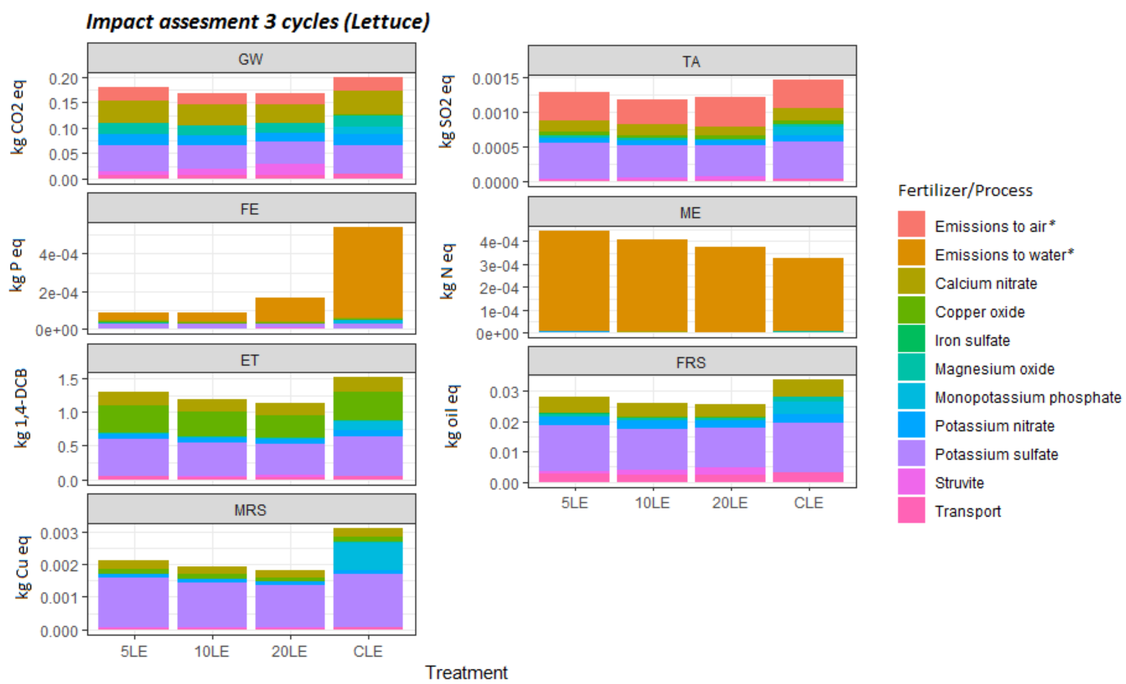


*Figure 6. 6 Impact assosciated to the system fertilization for 3 consecutive lettuce productions for 81 days. The obtained emissions have been calculated in relation to the resulting yield as FU. GW (global warming), TA (Terrestrial acidification), FE (Freshwater eutrophication), ME (Marinewater eutrophication), ET (Ecotoxicity), FRS (Fossile resource scarcity), MRS (Mineral resource sarcity). *Emissions to air were based on the emission factors of Ammonia, N2O and NOx obtained for the applied nitrogen; the emissions to water were directly obtained from the nitrogen and phosphorous detecte in the water leachate.*

ME, although related to the emissions to water, also does not decrease substantially for the struvite-treated crops due to its relation to nitrogen emissions, which are sustained for all treatments. Furthermore, we can observe that although a reduction of the impacts is occurring for the 20LE treatment, this reduction is most likely not a consequence of a reduced N emission to water but due to greater yields obtained; on the other hand, treatment 5LE is overshadowed by the lower yields generated and a proportionally greater N emission due to the smaller plant growth.

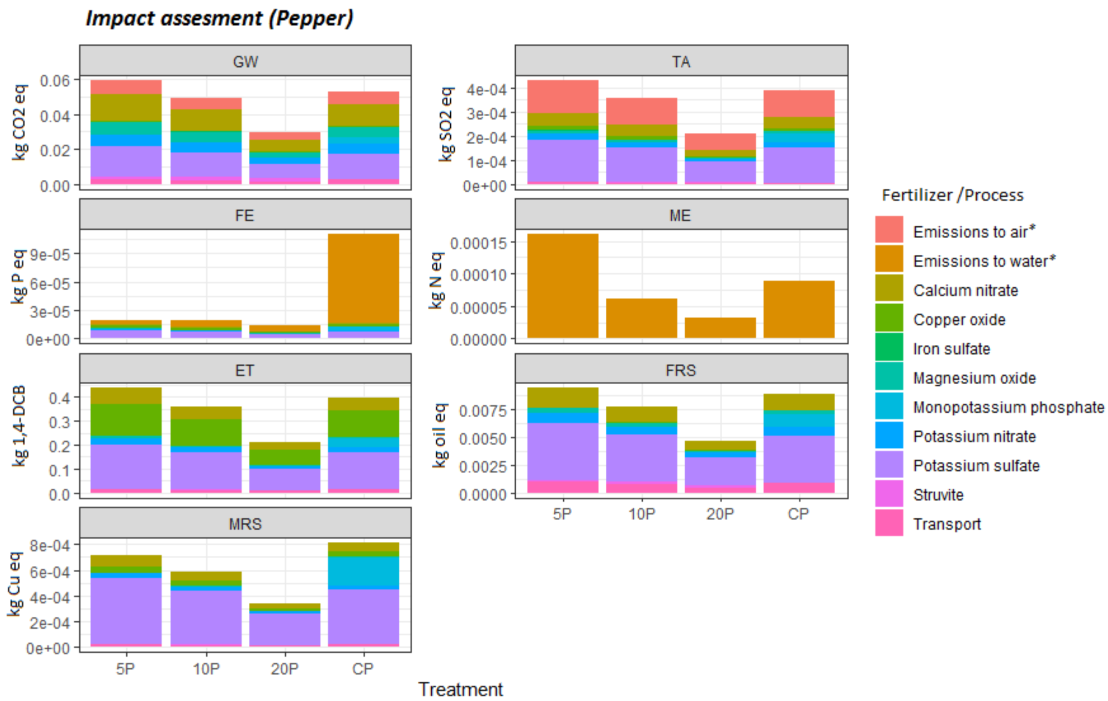


*Figure 6. 7 Impact assosciated to the system fertilization for pepper production during 81 days. The obtained emissions have been calculated in relation to the resulting yield as FU. GW (global warming), TA (Terrestrial acidification), FE (Freshwater eutrophication), ME (Marinewater eutrophication), ET (Ecotoxicity), FRS (Fossile resource scarcity), MRS (Mineral resource sarcity). *Emissions to air were based on the emission factors of Ammonia, N2O and NOx obtained for the applied nitrogen; the emissions to water were directly obtained from the nitrogen and phosphorous detecte in the water leachate.*

The results obtained for the pepper crop indicate a considerably abrupt decrease in the emissions in all impact categories for treatments 10P and 20P in comparison to the CP treatment. In comparison to the lettuce crop, the ME was severely reduced for these two treatments. The 5P treatment with lower production rates and therefore lower FU experiences much greater values for all impact categories except FE and MRS, which are slightly below the control treatment CP in the latter.

Overall emissions for pepper production were lower than those found for the three lettuce cycles combined. This finding can be explained by the greater weight obtained with pepper production, making a direct comparison between crops difficult with a functional unit only accounting for the obtained yield.

6.4 Discussion

6.4.1 Is struvite a good fertilizer for hydroponic production?

The results show that the long cycle of pepper and short cycles of lettuce fertilized with struvite did not differ greatly from each other in the uptake and use of P. We identified that the amounts accumulated in the plant biomass between treatments with the same struvite quantity (5LE and 5P, 10LE and 10P, 20LE and 20P) did not change substantially. This information reveals that little to no effect on struvite uptake can be attributed to the crop cycle duration or needs. This second idea is reinforced by the level of P found in the pepper biomass, corresponding to low concentrations and mirrored in the fruit P content (Hochmuth et al. 2018). Although a clear P deficiency is shown in the plant biomass nutrient content, no such deficiency can be traced in the plant physiology or production capacity (Zelia et al. 2017). Pepper fruit production increases with the given struvite, as well as leaf production and growth, showing significant differences that indicate the relevance of the given struvite amount to the plant. Related to the findings of Talboys *et al.* 2016 in 90-day experiments with struvite-fertilized crops, the amount of P taken by the plant is lower in the case of struvite but does not affect the final yield, being very similar to the more soluble triple superphosphate (TSP). This finding has been attributed to the struvite residual value in the substrate in comparison to TSP, enabling P uptake by the plant during a sustained timespan.

The leachate P for lettuce and pepper plants was also shown to be a great indicator of the slow solubility of the fertilizer and increased with greater water flow when lettuce was harvested. The higher water demands of the pepper plants could therefore have been a defining factor contributing to low struvite dissolution, as seen in figure 6.5. Although the plants had sufficient irrigation indicated by the daily water drainage, the leaching of phosphorous into the drained water only increased during the early stages of plant growth until 20 DAT. Once temperatures start to rise and drainage is reduced, the emissions of P into the drainage are also reduced. Although greater temperatures have been seen to increase struvite solubility (Rahaman et al. 2006; Ariyanto, Ang, and Sen 2017), its use as a fertilizer unveils that irrigation plays a major role in plant phosphorus uptake (Silber et al. 2005; Turner 1985; Lunt, Kofranek, and Clark 1964).

The greater variability obtained in the second lettuce cycle can also be attributed to the increasing temperatures enabling a greater dissolution of struvite in the perlite substrate as well as the slight reduction of the pH from the nutrient solution increasing the struvite solubility (Ariyanto, Ang, and Sen 2017). The uptake and use of the plant could have been affected by the delay in the irrigation adjustment to meet the plant needs.

The capacity of struvite dissolution, which has been attributed to different factors in previous literature, like the plant rhizosphere exudation (Dakora and Phillips, 2002; Kamilova *et al*., 2006; Khademi *et al*., 2010; Talboys *et al*., 2016), plant growth stage (Degryse *et al*., 2017) and plant needs. These factors for greater struvite dissolution have not been reflected in these results, indicating a reduced uptake from the pepper plant compared to the lettuce crop. The idea of plant rhizosphere exudation being important for struvite dissolution was also questioned by Rech *et al*., 2018, who demonstrated the inefficiency of low-concentration root exudates to solubilize granular struvite.

Overall, the quantity of P in the plant biomass (9–21% and 6–12% of the applied P for lettuce and pepper, respectively) as well as the P leachate (2–4% and 2% of the applied P for lettuce and pepper, respectively) in both crops indicate that the amount of dissolved P is very small. This information is reinforced by the analysis of the perlite substrate, indicating that a large amount of struvite remains undissolved in the substrate. This effect was also seen in previous literature with other crops, such as soybean and wheat (Rech et al. 2018a) and common bean (Arcas-Pilz, Rufí-Salís, et al. 2021). These low dissolution percentages coincide with dissolutions in media with pH values ranging from 7.5 to 8 (Talboys et al. 2016; Rech et al. 2018a), which were mainly found in the present study. While the pepper plants did not reach adequate ranges of P in the biomass with struvite fertilization, the lettuce crops did not differ greatly from the control treatment, especially for 10LE and 20LE. This information reinforces the idea of further reusing the given struvite for consecutive cycles within the same substrate with short cycle crops, such as lettuce.

On the other hand, the dissolution rate seems to be greater during the first plant cycle in all lettuce treatments. The struvite crystal composition and available P could be more prone to dissolve earlier, progressively reducing the dissolution rate with consecutive plant cycles. This same dissolution trend was seen by Rech *et al.* (2018) when observing the P concentration in the soil solution of wheat and soybean crops with the fertilization of three different struvite types. Concentrations of P were recorded for 40 days, showing a decrease and stability by the end of the experiment. The close dissolution rate of the second and third cycles could indicate this point of stability.

6.4.2 Does the use of struvite reduce the environmental burden of hydroponic production?

The environmental analysis showed that the 5LE and 5P treatments had the highest impacts since they had the lowest yields. On the other hand, the greater use of struvite can also generate a greater discharge of P into the water system compared to treatments with less applied struvite.

This finding is reflected in the case of the lettuce crops for the ME and FE impact categories. While greater yields were achieved for the 20LE treatment, greater P and N leachates were generated, increasing the environmental footprint in comparison to the other struvite treatments. Smaller crop growth in the case of 5LE and 5P can also increase the amount of leachates and discharge of N to the environment. This finding has been observed both for lettuce and pepper, where smaller crop growth leads to greater water and nutrient discharge. However, the P discharge in the struvite treatments was always lower than that in the control and thus impacted freshwater eutrophication.

The impact of the struvite production compared to the monopotassium phosphate seem significantly smaller, being most noticeable in GW and FRS. The production of monopotassium phosphate on the other hand has a large impact on the MRS as predicted, due to the extraction of the finite phosphate rock. The impact of monopotassium phosphate is also noticeable in the ET, TA and FRS categories, responding to the emissions of chemical agents into the environment for the extraction and transport to site. The overall impacts seem to be more dominated by the production emissions associated to potassium sulfate, being present in almost all IC due to its major role in the nutrient solution.

Takin in account the influence of the struvite slow solubility to reduce the emissions of P to water as well as the reduction of the impacts associated to the production of monopotassium phosphate, a great reduction of the impacts of fertilization can be seen.

While the pepper crop shows a clear reduction in emissions related to fertilization with the use of 10 g and 20 g of struvite, sustained production is unclear due to the low content of P in plants. While the production of pepper continues and demands on P can increase, its dissolution and uptake might not be sufficient in time. On the other hand, the lettuce needs were covered for all three cycles for all treatments, showing a P content similar to that of the control treatment. The idea of sustained production for longer periods of time corresponds to the findings of Bonvin *et al.*, 2015 and Rech *et al.*, 2018 urging for the definition of the residual value of the remaining struvite after the initial crop production.

To understand the environmental impact of one year of lettuce cycles, several assumptions were made. To generate the nine-cycle scenario (Annex II Table A6.13) that would correspond to yearly production, the three initial cycles for our three treatments were taken as references to generate correlations for the P uptake in biomass from the initial P given, as well as the potential yield produced with the P content in the plant biomass (Annex II Figure A6.12).

Further on, the error detected in this last correlation was subjected to a sensitivity analysis (Annex II Figure A6.18) adding a standard deviation of a total 46% to the yield production for a 9 cycle

production of lettuce in all treatments. The control was also given a standard deviation of 10%, taking in account that the P fertilization was consistent over time (Annex II Table A6.17).
The P loss through the leachates was estimated from the average obtained in all treatments due to its direct relation with irrigation. With the following prediction, the total biomass content of yearly production as well as the resulting yield and emissions to water were obtained to further extend the environmental outcome (figure 6.8). The control treatment was estimated with the generated yields and emissions from the three initial cycles. All other fertilizers for all treatments were based on the NS used for the three initial cycles extended for nine production cycles. The obtained emissions were then divided by the obtained total yields.

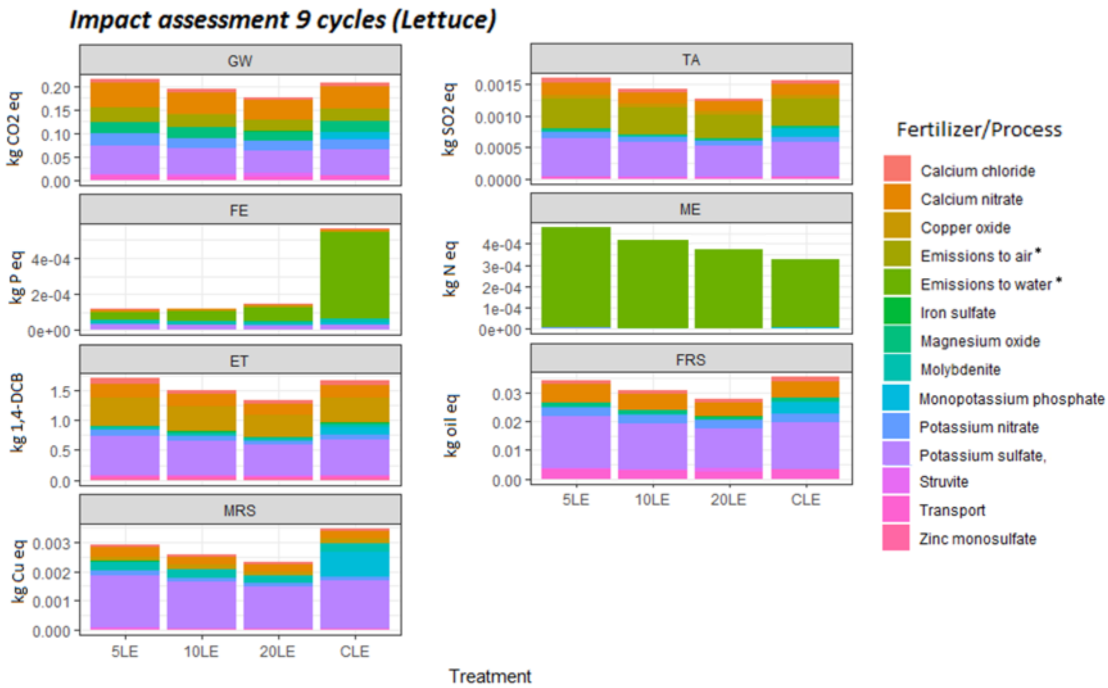


*Figure 6. 8 : Impact associated to the system fertilization for 9 consecutive lettuce productions. The obtained emissions have been calculated in relation to the prospective yield as FU. GW (global warming), TA (Terrestrial acidification), FE (Freshwater eutrophic eutrophication), ME (Marine water eutrophication), ET (Ecotoxicity), FRS (Fossil resource scarcity), MRS (Mineral resource scarcity). *Emissions to air were based on the emission factors of Ammonia, N2O and NOx obtained for the applied nitrogen; the emissions to water were directly obtained from the nitrogen and phosphorous detected in the water leachate.*

The LCA for the year's production with the same initial struvite shows a slight emission increase for all ICs, especially for the 5LE and 10LE treatments. The changes observed indicate that the productions obtained for the 5LE and 10LE treatments decrease to a point where the FU is reduced and consequently emissions are increased. On the other hand, control treatment yields were sustained in time and maintained close to identical emissions of the three lettuce cycles. The prospective production obtained for the 20LE treatment was similar to that of the control, obtaining results that reduced the environmental emissions for all impact categories compared to the control treatment except ME.

The 20LE treatment maintains the capacity for competitive production in time compared to the other struvite treatments which can also be seen in the sensitivity analysis in figure A6.18 in the Annex II, staying below the control emissions in lower production scenarios, especially for FE and MRS. This, however, implies a potential greater emission to water, as reflected in the FE and ME impact categories generated by the leaching of the struvite containing N and P. The use of the discharged water for less demanding crops (Rufí-Salís, Calvo, *et al.*, 2020) can further reduce nutrient leaching into the urban water cycle as well as a reduction and adjustment of the nutrient solution N content with the addition of struvite.

Further loss into the environment can be assessed with a specific analysis of the struvite nitrogen emission factor to the air in the form of ammonia, N_2O and NOx in soilless systems, which is strongly encouraged to determine the GW impact more accurately. This result has been viewed as both interesting and necessary research to understand whether slow dissolution can discourage emission to air or if the composition of N struvite in the form of ammonia will further induce processes of nitrification and denitrification in the substrate.

The findings in this work point out that the successful reuse of struvite in hydroponic production is possible and is been growing in importance, even being used in the fertirrigation for other crops achieving equal results to conventional fertilizers (Carreras-Sempere *et al.*, 2021). Similar work has been made with source separated urine, integrated into hydroponic production as nutrient source, and also using phytoremediation systems for yellow water treatment (Yang et al. 2015; Simha and Ganesapillai 2017; Volpin et al. 2018; Ikeda and Tan 1998). These works have found promising results on the reuse of urine although its application can be considered controversial (Simha et al. 2017).

This new way to find circularity in urban ecosystems is deemed as necessary and imposed specially in the waste treatment sector. The capacity to find an added value to the outcome of urban waste can help achieve new environmental goals like the compulsory recovery of P in certain regions of the EU (Kratz, Vogel, and Adam 2019). The local P recuperation and local administration can increase the local resilience to P pricing and distribution; therefore, the P precipitation and struvite production should be encouraged in WWTP.

6.5 Conclusions

The main conclusions drawn from the present experiment can be divided into two main aspects, one regarding the production and uptake by the plants and the second regarding the environmental benefits when compared to the use of mineral fertilizer. We found that the three cycles of lettuce treated with 20 g of struvite had the highest and most sustained overall yield, although such a high struvite concentration resulted in very slow dissolution. In this case, 50% to 70% of the struvite remains undissolved in the substrate after three lettuce cycles, indicating great potential for further consecutive production cycles. Estimations of a year's worth of lettuce production with the same initial struvite indicate sustained production similar to the control, while production for all struvite treatments apart from 20LE would be reduced. While no signs of P deficiency can be seen in the pepper plants, even when obtaining a greater production, the P content was regarded as very low due to the slow struvite solubility. Pepper production was successful in the three-month experiment, although longer production cycles were not tested.

The environmental outcome of the experiment shows a general reduction in the environmental impacts, especially regarding the use and emission of P for freshwater eutrophication and mineral resource scarcity. The production of 20LE is sustained over time, therefore reducing the emissions below the control treatment except for ME. The greater N emissions to water associated with the ME can be reduced by adjusting the nutrient solution, considering the N delivered by the struvite. The findings of this study further encourage the use of struvite in hydroponic production due to the capacity of sustained production of shorter and longer cycle crops as well as the reduction of the environmental impacts compared to mineral fertilizer, such as MKP.

Author contributions:

All authors were responsible for the conception and design of the study. V. Arcas-Pilz, M. Rufí-Salís, F. Parada, G. Villalba and X. Gabarrell conceived the original idea for the study. V. Arcas-Pilz, G. Stringari, R. Gonzalez and F. Parada set up, supervised and acquired the data for the experimental tests. V. Arcas-Pilz processed and analysed the data. V. Arcas-Pilz took the lead in writing the manuscript. All authors critically revised the draft for important intellectual content. All authors gave their final approval to the manuscript.

Chapter 7

This chapter is based on the journal paper:

Assessing the environmental behavior of alternative fertigation methods in soilless systems: The case of Phaseolus vulgaris with struvite and rhizobia inoculation

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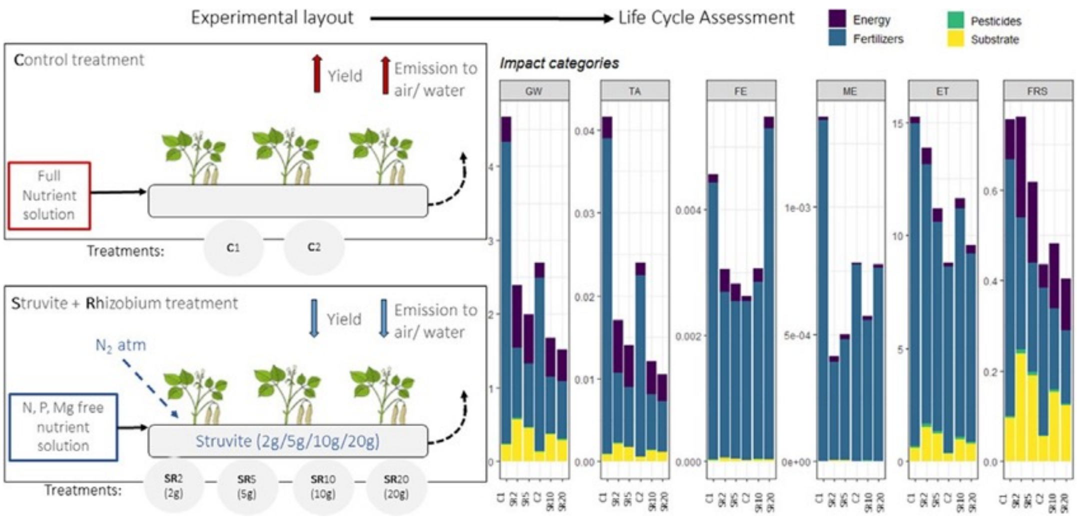
Chapter 7: Assessing the environmental behavior of alternative fertigation methods in soilless systems: The case of Phaseolus vulgaris with struvite and rhizobia inoculation

Abstract

Urban agriculture, while being a promising solution to increase food sovereignty in cities, can lead to an unprecedented discharge of nutrient and fertilizer-related emissions into the urban environment. Especially relevant are nitrogen (N) and phosphorus (P), due to their contribution to marine and freshwater eutrophication. Therefore, alternative methods of fertilization need to be put into practice to avoid such impacts to the surrounding environment. Struvite, has been studied as a potential slow releasing fertilizer due to its high P content, while the bacteria rhizobium has been used to fix N directly from the atmosphere. Legumes, like the common bean are N-demanding crops capable of symbiosis with the bacteria rhizobium and have previously shown positive responses to fertilization with struvite. This study aims to analyze the environmental performance of plant production in hydroponic systems combining rhizobium inoculation and struvite (2g, 5g, 10g, 20g) irrigated with a N and P deficient nutrient solution, using life cycle analysis (LCA). The nutrient content of in- and out-going irrigation was analyzed as well as in plants and beans. The functional unit for the LCA was 1kg of fresh beans. The results obtained indicate a yield reduction of 60% to 50% in comparison to the control which was irrigated with a full nutrient solution. The impacts from operational stage are less in all impact categories, where most significant reductions up to 69% and 59% are seen in marine-eutrophication and global warming respectively. Although the infrastructure does not change between treatments, its impacts increase due to lower the yields. We determine that below a 10% of the control yield, the alternative systems have more impact than the use of conventional mineral fertilizers in almost all impact categories, thus pointing to the importance of infrastructure to truly reduce environmental impacts for urban agriculture.



7.1 Introduction

Urban Agriculture (UA) has the potential to replace traditional food supply chains to some degree, thereby reducing transportation, packaging and food losses while increasing food sovereignty of cities (Sanyé et al. 2012b; Sanjuan-Delmás et al. 2018; Tornaghi 2017; Siegner, Acey, and Sowerwine 2020). However, the additional need of inorganic chemical fertilizers inevitably results in greater discharge of these chemicals into the environment as well as an increase of the resource depletion potential (Rufí-Salís et al. 2020). This is especially relevant considering the emission of nitrogen and phosphorus species, substantially contributing to marine and freshwater eutrophication, causing oxygen deprivation in aquatic environments. Specifically, urban water cycles and runoff are a great concern with their high implication in water eutrophication damaging ecosystems close to cities as well as close to intensely fertilized agricultural sites (John H. Ryther and Dunstan 1971; Lewis, Wurtsbaugh, and Paerl 2011). The integration of agriculture within city boundaries could therefore further increase the potential of emissions into the urban water cycles.

It is important to find ways for UA to be highly resource-efficient so that urban areas are able to expand these production practices without incurring significant environmental impacts associated with the additional water, energy, and nutrient requirements. To mitigate these environmental impacts, alternative and more environmentally friendly fertilizers have to be applied to attain competitive yields without causing great impacts to the surrounding environment (Lewis, Wurtsbaugh, and Paerl 2011) as well as avoiding further extraction of phosphorous for agricultural purposes (Linderholm, Tillman, and Mattsson 2012).

Recent work has been focused on the recovery of nutrients from wastewater treatment plants (Lam, Zlatanović, and van der Hoek 2020; Harder et al. 2019; Shaddel et al. 2020), showing a great range of possible alternatives for fertilization generated in urban areas as well as their constraints.

One of the available options showing great potential for its use in agriculture, is struvite. The struvite crystal is formed by a spontaneous precipitation in wastewater treatment plants and is regarded as a slow releasing fertilizer due to its low dissolution and high content of phosphorous (12.5%), magnesium (9.9%) and nitrogen (5.7%) (Talboys et al. 2016; Rahman et al. 2014; Degryse et al. 2017b). It has been reported that the formation of struvite can recover up to 90% of Phosphate in wastewater sludge, reaching even higher percentages depending on the precipitation process and source (Kataki et al. 2016).

Studies on the use of struvite in agriculture (Massey, M.S., Davis, J.G., Sheffield, R.E., Ippolito 2007; Degryse et al. 2017a; J. N. Ackerman et al. 2013; Talboys et al. 2016) point out that the use of these recovered nutrients can reduce mineral fertilizer requirements while implying little to no cost for farmers (Karak and Bhattacharyya 2011). The use of struvite has been shown to successfully substitute the use of mineral phosphorous fertilizers, while reducing nutrient losses to the environment due to its slow dissolution rate (N. Ahmed et al. 2018). Agricultural production with struvite as the main source of P has been tested on a variety of crops rainging from ryegrass (*Lolium perenne*) and broad beans (*Vicia faba*), which experience an increase of fresh yiels of 76% and 54% respectively, to canola (Brassica napus) and wheat (*Triticum aestivum L*.), that suffer a reduction of the nutrient uptake and therefore a reduction of the plant yield (N. Ahmed et al. 2018). Other crops, however, have been seen to experience no significant changes with the use of struvite like the case of maize (Uysal et al. 2014) and corn (Thompson 2013).

The environmental performance of the struvite extraction as P fertilizer has been previously studied (Ishii and Boyer, 2015) and while its benefits in comparison to virgin phosphorous have been identified in the reduction of nutrient emissions and offsets commercial fertilizer production, its total environmental performance depends on the chemical inputs used for the struvite precipitation as well as the infrastructure and the recovery accounted in the life cycle inventory (Linderholm, Tillman and Mattsson, 2012; Ishii and Boyer, 2015; Lam, Zlatanović and van der Hoek, 2020).

Whereas these studies have mainly substituted phosphorous based fertilizers, the use of nitrogen in the form of ammonium nitrate, urea and monoammonium is still given to the crops. As previously established, the emissions of nitrogen in the environment are greatly damaging and especially crucial for high N demanding crops like legumes.

The inoculation of legume crops with the bacteria rhizobium has been explored as a way for the plant to fix its nitrogen directly from the air without boosting its environmental footprint (Olivera et al., 2004; Gopalakrishnan et al., 2015; Kontopoulou et al., 2015, 2017; Savvas et al., 2018; Araujo, Urbano and González-Andrés, 2020; Sammauria et al., 2020; Sanyal, Osorno and Chatterjee, 2020). This bacteria forms an endosymbiotic interaction with the plant, profiting from compounds generated through photosynthesis while fixing atmospheric N2 that is then given to the plant in form of ammonia (NH_3) (Long 1989; Fisher and Long 1992). As a result of these previous studies, it has been seen that in terms of the obtained yields, rhizobium tends to diminish the crop production in comparison to synthetic nitrogen fertilizers (Savvas et al. 2018a; Sanyal, Osorno, and Chatterjee 2020; C. K. Kontopoulou et al. 2015; 2017) while its use on soil for common bean reduces about 19% per ha of the environmental burden when mineral N fertilization is replaced (Araujo, Urbano, and González-Andrés 2020).

To summarize the state of the art in nutrient recovery for mineral fertilizer substitution, the above-mentioned studies have shown that struvite can reduce and substitute a significant amount of P fertilizer while recovering great amounts of phosphate nutrients from wastewater treatment plants (WWTP). On the other hand, rhizobium can reduce the need for nitrogen mineral fertilizers, only partially reducing the environmental impact associated to the use of fossil-dependent mineral fertilizers. However, no study has attempted to use both struvite and rhizobium to completely avoid the application of nitrogen and phosphorous fertilizer, thereby reducing environmental impacts even further.

This paper aims to fill this gap by exploring the feasibility and environmental impact of applying struvite combined with rhizobium inoculation as alternative fertilizers of a UA system. To do so, we use the Life Cycle Assessment (LCA) to quantify the environmental impacts of a common bean crop in which the seeds were inoculated with soil bacteria rhizobium and different quantities of struvite were applied, compared to conventionally fertilized bean plants. The bush bean Phaseolus vulgaris "Pongo" was used in the experiments due to previous tryouts showing a good production in the perlite substrate as well as for being a highly consumed leguminous crop in Spain. The objective is to show the benefits and costs of this fertilizer alternative when compared to mineral fertilizer and provide knowledge towards reducing resource extraction for Urban Agriculture (UA) as well as avoiding possible emissions into natural ecosystems.

7.2 Materials & Methods

7.2.1 Description of integrated Rooftop Greenhouse (i-RTG)

The experiments were conducted in the greenhouse laboratories for UA located on the integrated Rooftop Greenhouse Laboratory (i-RTG-Lab) of the Environmental Science and technology building (ICTA-UAB) located in the Universitat Autònoma de Barcelona campus (UTM: 42º29'24" E, 45º94'36" N). The irrigation system is hydroponic on substrate with primary the use of rainwater. The 900m2 rainwater harvesting system (RWHS) is included in the building structure as well as a 100m3 storage tank located underground from which the water for irrigation is pumped to the cropping sites. The building structure and its year-round production have been previously analyzed to identify the environmental impact reduction due to the connectivity and synergy between the greenhouse and the building (Sanjuan-Delmás et al. 2018). This building has two greenhouse laboratories for UA on the fourth floor, where this experiment was conducted. The beans were planted on the South- West facing laboratory (Urban Agriculture Laboratory 2) with a total area of 122.8m2 as can be seen on the plant layout shown in figure 7.1.

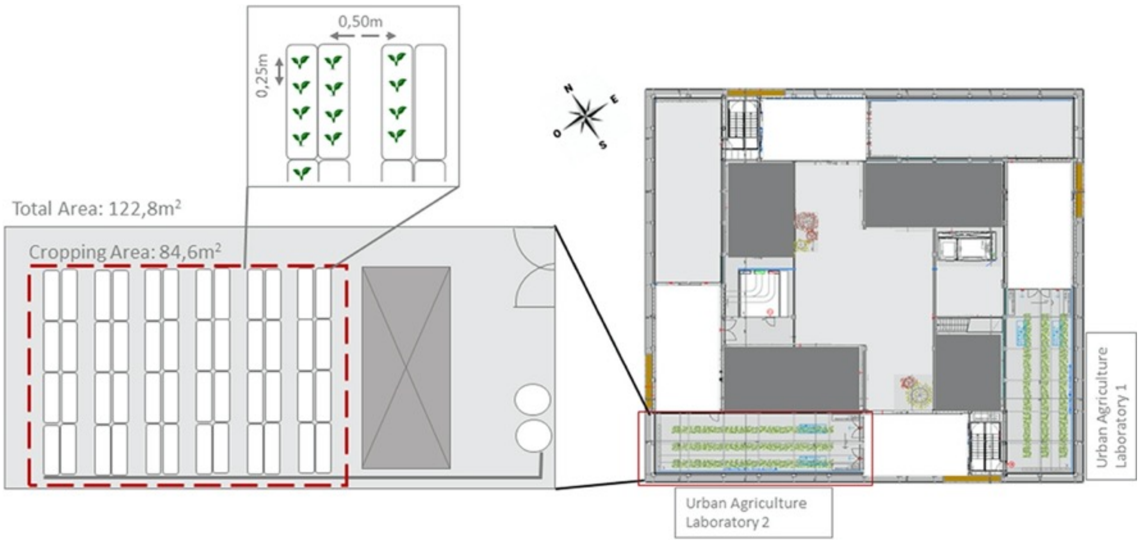


Figure 7. 1 Experimental layout of the experiment in the i-RTG

Several sensors were used to monitor temperature (T107 Campbell Scientific) and relative humidity of the i-RTG cropping areas (Table A7.1 in the Annex II). Irrigation water, water drainage, electric conductivity and pH for each irrigation line were measured three times a week.

7.2.2 Plant materials and growth conditions

The seedlings were obtained from a nursery, where the seeds were inoculated with the rhizobium mix and transported to the i-RTG 10 days after planting. The production system is soilless with perlite substrate and nutrient solution given through the 2L/h drip irrigation system. The cropping area was arranged in twelve rows with four 40L perlite bags each (figure 7.1). Four bean plants were planted in each 1m long perlite bag, making a total of 192 plants, divided in three treatments (64 per treatment). The plantation frame was 0.125m2 within a total cropping area of 84.6m2. The irrigation was set 4 times a day for 3 minutes giving a total amount of 400ml per day for each plant.

Two experiments were performed. The first experiment took place in 2019, starting on the 16th of January and ending on the 10th of April. The second experiment took place in 2020, and the plants were transplanted on the 13th of February and the experiment was finalized on the 7th of

May, lasting each one a total of 84 days. We used different concentrations of struvite during the two experiments to determine the N and P assimilation rates and how the yield was affected. In 2019, the bean plants were treated with 2g of struvite (0.25g of P; 0.114g of N) per plant (SR2) and 5g of struvite (0.625g of P; 0.285g of N) per plant (SR5). In 2020, we incremented the amount of struvite to 10g (1.25g of P; 0.57g of N) per plant (SR10), and 20g of struvite (2.5g of P; 1.14 of N) per plant (SR20). The inoculation was made prior to their sowing, embedding the bean seeds in the commercial liquid rhizobium mixture before planting. We performed a control experiment both years parallel to the treatments under the same temporal and climatic conditions but fertilized with a full nutrient solution, with zero struvite and without inoculation. These control treatments were also made with the same crop and during the same time, lasting 84 days in the rooftop greenhouse as well.

The struvite granules were placed close to the root area to ensure a better absorption by the plant. To avoid possible runoff of struvite granules a 1L bag with small holes for water drainage, was placed around the root area to retain the crystalline granules close to the plant. The granulated urine derived struvite was given directly to the plants rhizosphere after transplanting them into the integrated greenhouse.

Commercial inorganic fertilizer

Two nutrient solutions (Table 7.1) were made for both campaigns, one standard full nutrient solution (NS) with nitrogen, phosphorous and magnesium and a second solution deficient in nitrogen, phosphorous and magnesium with a higher content in K_2SO_4 to avoid potassium as a limiting factor. All nutrients were mixed into a concentrated solution stored in 50L tanks, further diluted with rainwater when irrigated in a ratio of 1:100 (NS:Rainwater).

Table 7. 1 Nutrient Solution composition

Nutrients applied	Control NS	Mg, P, N-FREE NS
KPO4H2	136 mg/L	–
KNO3	101 mg/L	–
K2SO4	304.5 mg/L	435 mg/L
Ca(NO3)2	410 mg/	–
CaCl2	111 mg/L	111 mg/L
Mg(NO3)2	148.3 mg/L	–
Hortrilon*	0.1 mg/L	0.1 mg/L
Sequestrene**	0.1 mg/L	0.1 mg/L

7.2.3 Commercial Rhizobium inoculant

The inoculant used for this experiment was obtained through a company based in Karlsruhe, Germany, nadicom GmbH. This 1L liquid product contained a mixture of two rhizobia strains, Rhizobium phaseoli and Rhizobium giardinii, that were directly applied on the bean seeds (except for the Control) before planting and again 5 days after transplanting to the ICTA RTG- Lab. The manufacture and transport of this commercial product was not included in the LCA as an input for our alternatively fertilized crops, since the production impact has been considered minimal.

7.2.4 Commercial struvite

The struvite used for the experiment was urine derived, obtained from a wastewater treatment plant (WWTP) in Denmark. The plant recovers struvite from the digestate flow through the addition of reagents to reach stoichiometric levels that trigger struvite precipitation. The obtained struvite (Mg(NH4)PO4·6H2O) has a composition conformed by 12.5% w/w phosphorous; 5.7% w/w nitrogen and 9.9% w/w magnesium and a granule size of 1 to 3mm. The heavy metal content in struvite from different origins and production systems has been analyzed and set at levels under the European threshold, also ranging far below the amount of possible impurities that can be found with the production of phosphate rock as well as untreated sewage sludge from WWTP (Bastida et al. 2019).

7.2.5 Experimental analyses and nutrient balances

Water samples were taken from each irrigation system as well as the drained water 3 times a week. Production of the bean plants was counted and weighted. The amount of drained leachates were measured daily and sampled three times a week. The concentrations of Cl^-, NO_2^-, NO_3^-, PO_4^{3-}, SO_4^{2-}, Ca^{2+}, K^+ and Mg^{2+} were measured using ionic chromatography. Additionally, the pH and EC were measured daily for both the nutrient solutions and leachate water. To reduce the possible error generated through the irrigation and sampling the generated data was adjusted to a curve. The incoming and outgoing nutrients were quantified as well as the nutrients found in the plant biomass and beans. The plant biomass was collected at the end of the experiment with a sample number of 8 plants per treatment. These samples were dried and weighted before being digested with a Single Reaction Chamber microwave with concentrated HNO_3. The digested samples where then analyzed using Optical Spectrometry (ICP-OES). The same procedure was applied to the obtained production of beans, sampled throughout the experiment. The final balance per plant was assessed with the following equation:

$$Eq\ 6: \qquad Fns + Fs + Ffix = Fp + Fb + Fpl + Fl$$

Equation 6: Fns= g nutrients in nutrient solution, Fs= g in struvite , Ffix= g nutrients, Fp= g nutrients in production, Fb= g nutrients in biomass, Fpl= g nutrients in perlite, Fl= g nutrients in leachates

To calculate the fraction needed per plant to close the balance, the following equation was used (Eq7):

$$Eq\ 7: \quad \text{Balance}\ \% = 100 * \frac{Fp}{(Fns + Fs + Ffix)} + \frac{Fb}{(Fns + Fs + Ffix)} + \frac{\text{Fpl}}{(Fns + Fs + Ffix)} + \frac{\text{Fl}}{(Fns + Fs + Ffix)}$$

Equation 7: Fp= g nutrients in production, Fb= g nutrients in biomass, Fpl= g nutrients in perlite, Fl= g nutrients in leachates, Fns= g nutrients in nutrient solution, Fs= g in struvite, Ffix= g nutrients obtained through N2 fixation.

The following results depict the data collected in 2019 for plant biomass, irrigation and leachate nutrient content as well as yield production and nutrient content. The 2020 study was included to provide further information on the effect of greater struvite quantities to increase the yield. Therefore, the LCA results for 2020 only defer from the 2019 inventory in the amount of struvite used as well as the yield.

Additionally an analysis to calculate the fraction of N in the biomass obtained from N_2 fixation was made, using an elemental analyzer- isotopic ratio mass spectrometer (EA-IRMS; Thermo Fisher Scientific), attaining the δ15N values (in ‰) for our treatments SR2, SR5 and control as well as our alternative fertilizer struvite which was set in 7.1‰. Contributions from each source (atmospheric or struvite) were then calculated with the following equation (Shearer and Kohl 1993; Unkovich et al. 2002; Arndt et al. 2004), using the lowest δ15N value obtained as our 'B' value (-1.16‰) (Shearer and Kohl 1989; Peoples, Boddey, and Herridge 2002; Kermah et al. 2018):

$$Eq\ 8: \qquad \%Ndfa = \frac{\delta^{15}N\ Source\ 2 - \delta^{15}N\ Sink}{\delta^{15}N\ Source\ 2 - 'B'value} \times 100$$

Equation 8: %Ndfa (Nitrogen derived from N2 fixation from the atmosphere), δ15N Source 2 (‰) corresponds to the δ15N value of struvite, δ15N Sink (‰) corresponds to the δ15N value from the sample, 'B' value set at -1.16‰

7.2.6 Life Cycle Assessment (LCA)

The LCA is a tool with a standardized methodology (ISO 2006) used to determine the environmental performance of goods in all stages of their life cycle of the four proposed treatments (SR2, SR5, SR10 and SR20) and control. The scope of the LCA study is cradle to gate of the bean production system. The functional unit (FU) chosen is 1kg of fresh beans at the collection point. The cut-off method in the Simapro software was applied which allocates the benefit of the recycled materials to the recycled products. To calculate the life cycle environmental impacts of the treatment, we used the Simapro software and the EcoInvent 3.5 attributional database. The following impact categories (IC) were selected, all from the ReCiPe (H) Midpoint method: Global warming (GW), Terrestrial acidification (TA), Freshwater eutrophication (FE), Marine Eutrophication (ME), Fossil Resource Scarcity (FRS) and Ecotoxicity (ET), which is the sum of Freshwater, Marine and Terrestrial ecotoxicities.

The system definition is illustrated by figure 7.2, which differentiates between the subsystems infrastructure, operation and end of life. For infrastructure, we considered the production and end of life of the greenhouse, the rainwater harvesting system and the auxiliary equipment such as pumps and fertirrigation installed and all the transportation required. All steps shown in figure 7.2 for raw material extraction, processing, transport to construction site, construction/ maintenance, as well as the transport to the landfill or recycling site were considered. On the operational side, the study includes the production, use and end of life, including transport, of all the resources required for the duration of the experiments (perlite substrate, fertilizers, struvite, pesticides, water, and energy). Exceptions to this are the production of nursery plants and the composting of the residual biomass as well as the rhizobium production. For the production of struvite the additional inputs for controlled precipitation were accounted, in this case the chemical inputs can be seen in the LCA inventory, which consisted in MgCl, Energy and NaOH for 1kg of struvite as described by the technology developed by Ostara® (Amann et al. 2018). The used wastewater for the struvite precipitation was not considered within the system boundaries for this study.

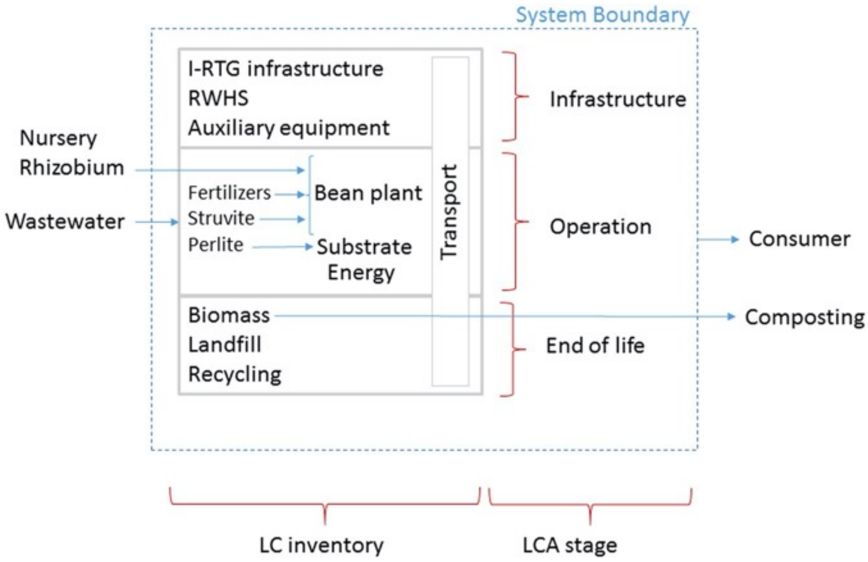


Figure 7. 2 Representation of the LCA system boundaries of the present experiment for fresh bean production. Division between the operation al system, the infrastructure and the end of life subsystems. LCA inventory represented on the left and the LCA stages on the right. The system boundary defined by the dotted line delimiting within the accounted materials and stages in this study.

For the end of life subsystem of our production several assumptions were made. The remaining biomass generated in the greenhouse goes to composting as well as the used substrate after 5 years of use. The composting of the residual biomass was not considered within the system boundaries. The leachate water was discharged into the urban water cycle entering the wastewater treatment plant. All phosphates and nitrates discharged into the water are therefore considered direct emissions to water, in the case of the treatments fertilized with struvite, also as direct emissions to the air. For the system infrastructure it was considered that the RWHS as well as the Auxiliary equipment were assumed to be disposed of into the landfill. The distance to the landfill and recycling site were assumed to be 30 km from the greenhouse.

The inventory data for the infrastructure and auxiliary equipment was compiled from Sanyé et al., 2012; Sanyé Mengual, 2015; Sanjuan-Delmás et al., 2018; Rufí-Salís, Petit-Boix, et al., 2020. For both the rainwater harvesting system (RWHS) and the i-RTG System a lifespan of 50 years was considered while the auxiliary equipment was set at 10 years, taking into account previous work by Sanjuan-Delmás et al., 2018 and Rufí-Salís, Petit-Boix, et al., 2020. Emission factors for N to air were calculated according to Llorach-Massana et al., 2017, while N and P in water were directly measured. The emissions to air in struvite where calculated taking into account the emission factor of the total nitrogen in the applied quantity of struvite, even when not all struvite was dissolved.

For the transportation of all materials average values for the transport to markets were given. The transport to the i-RTG was then added with a distance of 50km for all pesticides, fertilizers and auxiliary equipment as well as the struvite and rhizobium applied. Transport distance of

850km was applied for the substrate bags following the methodology of Sanjuan-Delmás et al., 2018. No transport of the horticultural production was considered since one of the benefits of urban agriculture is the on-site selling of the products, therefore the product procurement by the consumer is located outside our system boundaries.

The data for the operation was collected during the experiment, including the amount of fertilizers, the substrate used as well as the energy used to work the irrigation system. The energy used during the campaign was estimated by the water pumps and amount of water pumped to the greenhouse and crops.

The full inventory is available in supporting information (Tables A7.4 to A7.8 from the Annex II).

7.3 Results

7.3.1 Yields and nutrient balance

The total and average-per-plant production for both campaigns (2019 and 2020) can be seen in Table 7.2 for each treatment. The results show that as struvite concentration is increased from SR2 to SR20, production also increases from 1899.2g to 4821.5g indicating a significant assimilation rate of P by the plants. It is crucial to point out that the two controls differ greatly as well from one campaign to the other. Since the campaign of 2020 began in February and the 2019 campaign in January, we can consider the climatic conditions as an explanation for greater productions, taking into account that minimal temperatures were higher in 2020 (by more than 6ºC), as well as the average temperature throughout the experiment (Table A7.1 Annex II). These conditions would enable a greater and faster production of flowers and an earlier bean growth. The first bean pod harvest made in 2019 was 49 days after transplanting the bean plants into the greenhouse while in 2020 the bean pod harvest began 39 days after transplanting (Figure A7.2 in the Annex II). Due to the different climatic conditions and resulting productions the control treatment for both campaigns has to be considered as a reference. The observed productions do not reach more than 60% of the achieved production in the control treatment, staying between 40 to 60%, in both campaigns.

*Table 7. 2 Production for campaigns 2019 and 2020 for all treatments. *Difference to control = per- centage in relation to control as 100%.*

Production	2019			2020		
Treatment	SR2	SR5	Control 1	SR10	SR20	Control2
Total	1899.2 g	2375.6 g	4726.7 g	3542.2 g	4821.5 g	8198.4 g
Average per plant	59.3 g	74.2 g	147.7 g	110.7 g	150.6 g	256.2 g
Dif to control*	40.2%	50.3%	100%	43.2%	58.8%	100%

7.3.2 Nutrient fluxes

The obtained nutrient balance can be seen in table 7.3. Here the nutrient content for Nitrogen, Phosphorous, Magnesium, Potassium, Sulfate and Calcium in the incoming nutrient solution can be observed, as well as the outgoing fluxes of production (bean pods), biomass (leaves, stems, roots), perlite and leachates. In the case of nitrogen, the fixated N_2 is also taken into account as seen in figure 7.3. Table A7.3 in the Annex II also provides some incoming (struvite input) and out coming flows (leachates) for the SR10 and SR20 treatments for P, N and Mg.

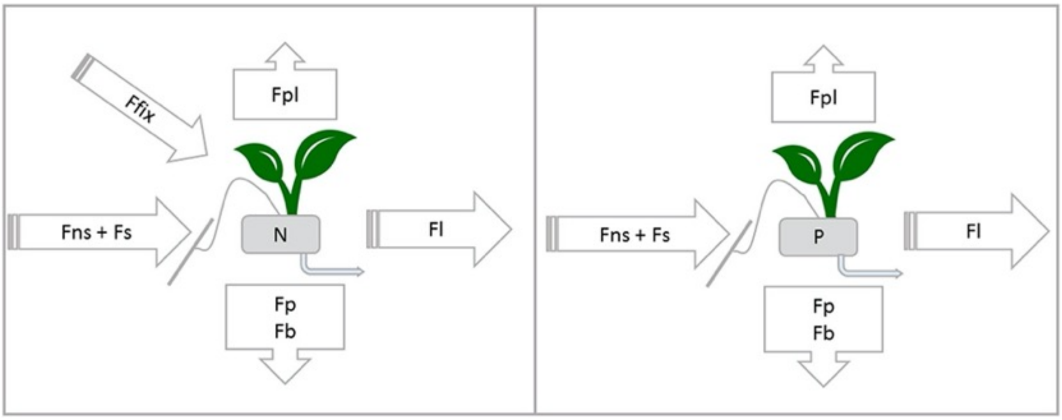


Figure 7. 3 Nutrient flow representation for N (left) and P (right). Fp = g nutrients in production, Fb = g nutrients in biomass, Fpl = g nutrients in perlite, Fl = g nutrients in leachates, Fns = g nutrients in nutrient solution, Fs = g in struvite, Ffix = g nutrients obtained through N_2 fixation.

The obtained results for the fixed g of N were achieved thanks to previous studies with isotopic N15 analyses where the percentages of fixation for SR2, SR5 and the 2019 control where obtained. The average percentage of fixed nitrogen for SR2 was 82% while for SR5 it was 72% and 16% for the control of the total N found in production and biomass. This fixed Nitrogen was further given as an additional inflow.

*Table 7. 3 Nutrient balance per plant for N, P, Mg, K, S and Ca Fp = g nutrients in production, Fb = g nutrients in biomass, Fpl = g nutrients in perlite, Fl = g nutrients in leachates, Fns = g nutrients in nutrient solution, Fs = g in struvite, Ffix = g nutrients obtained through N_2 fixation *perlite was obtained from Sanjuan-Delmas.*

Nutrient	Treatment	Nutrient solution+ struvite (Fns + Fs)	AtmN_2 Fix (Ffix)	Production (Fp)		Biomass (Fb)		Perlite* (Fpl)	Leachates (Fl)		Balance
		g	g	g	%	g	%	%	g	%	%
N	SR2	0.114	0.293	0.152	37%	0.204	50%	0%	0.031	8%	95%
	SR5	0.285	0.301	0.159	27%	0.262	45%	0%	0.052	9%	81%
	Control	1.271	0.123	0.376	27%	0.375	27%	6%	0.323	23%	83%
P	SR2	0.25	0	0.021	8%	0.036	14%	0%	0.041	17%	40%
	SR5	0.625	0	0.030	5%	0.057	9%	0%	0.055	9%	23%
	Control	0.740	0	0.081	11%	0.109	15%	6%	0.274	37%	69%
Mg	SR2	0.198	0	0.011	6%	0.020	10%	0%	0.096	49%	64%
	SR5	0.495	0	0.016	3%	0.050	10%	0%	0.133	27%	40%
	Control	0.461	0	0.032	7%	0.060	13%	0%	0.292	63%	83%
K	SR2	6.357	0	0.173	3%	0.270	4%	0%	4.249	67%	74%
	SR5	6.357	0	0.206	3%	0.357	6%	0%	4.128	65%	74%
	Control	4.774	0	0.576	12%	0.739	15%	0%	3.664	77%	104%
S	SR2	1.626	0	0.009	1%	0.030	2%	0%	1.420	87%	90%
	SR5	1.626	0	0.011	1%	0.040	2%	0%	1.405	86%	90%
	Control	1.611	0	0.030	2%	0.060	4%	0%	1.040	65%	70%
Ca	SR2	1.713	0	0.021	1%	0.150	9%	3%	1.126	66%	79%
	SR5	1.713	0	0.025	1%	0.195	11%	3%	1.374	80%	96%
	Control	1.719	0	0.061	4%	0.475	28%	3%	1.127	66%	100%

The balance for nutrients N, H, K, and Ca is close to 100%, indicating that the inflows can be traced almost entirely in the different outflows. On the other hand, there are losses of P and Mg in the SR2 and SR5 treatments, which are unaccounted for in the mass balance showing percentages under 50% reaching values as low as 23% for the P balance in SR5. We consider that the reason for such low accounting of the balance for P and Mg is the possibility of them remaining undissolved in the perlite bag. While Sanjuan-Delmas provides information of the amounts of these nutrients in fertigation that can be found in the perlite, the remaining P, Mg and N from struvite left in the perlite bag was not determined. This factor can generate uncertainty in our nutrient balance which has to be taken into account. When observing the percentages of out coming P and Mg in treatments SR10 and SR20 (Table A7.3 in the Annex II) we can observe a trend in the leachate amount being around 8 to 11% for SR10 and SR20 respectively. These leachate percentages close to the observed in the SR2 and SR5 treatments further support the idea of an uncompleted dissolution in the perlite bag. On the other hand the N balance seems to fit the incoming and out coming flows, while the amount of the given N with the struvite is respectively lower to Mg and P and the atmospheric N_2 fixation also amounts to additional nitrogen in plant biomass and production. When regarding the percentage of discarded P and N into the leachates we can observe a reduction of almost threefold in N when comparing the control (23%) to both treatments (SR2 with 8% and SR5 with 9%) and double (SR2 with 17%) or even fourfold (SR5 with 9%) in P compared to the control (37%). When regarding the quantity of P in the leachate water of the control treatment and the SR20 treatment we can see that it is almost identical, pointing out that the given quantity of struvite has achieved flows to wastewater similar to the control treatment. On the other hand, when considering the percentages in regard to the total amount given to the plant the control treatment has leached more than threefold the added P (37%) compared to the SR20 treatment (11%). When observing the N incoming flows the input given for the control treatment in the irrigation is similar to the quantity given, in form of struvite, in the SR20 treatment. We must bear in mind that for the treatment SR20 the quantity of N gained from atmospheric fixation is not known. On the other hand the leached outflow for the SR20 accounts for about a 13%, almost half of the leached amount in the control (23%). The lower quantities of P and N measured in water, although similar or greater N and P quantities were given to the control treatment, indicate a much slower dissolution of this fertilizer, reinforcing the idea of a remaining undissolved amount of struvite in the perlite bag.

7.3.3 Environmental performance of the treatments

The LCA impacts per functional unit (FU) are disaggregated into the life cycle stages that resulted in the highest impacts for all four treatments and controls, as shown in figure 7.4. Since the controls resulted in higher yields, consequently the impacts are reduced considerably in all categories.

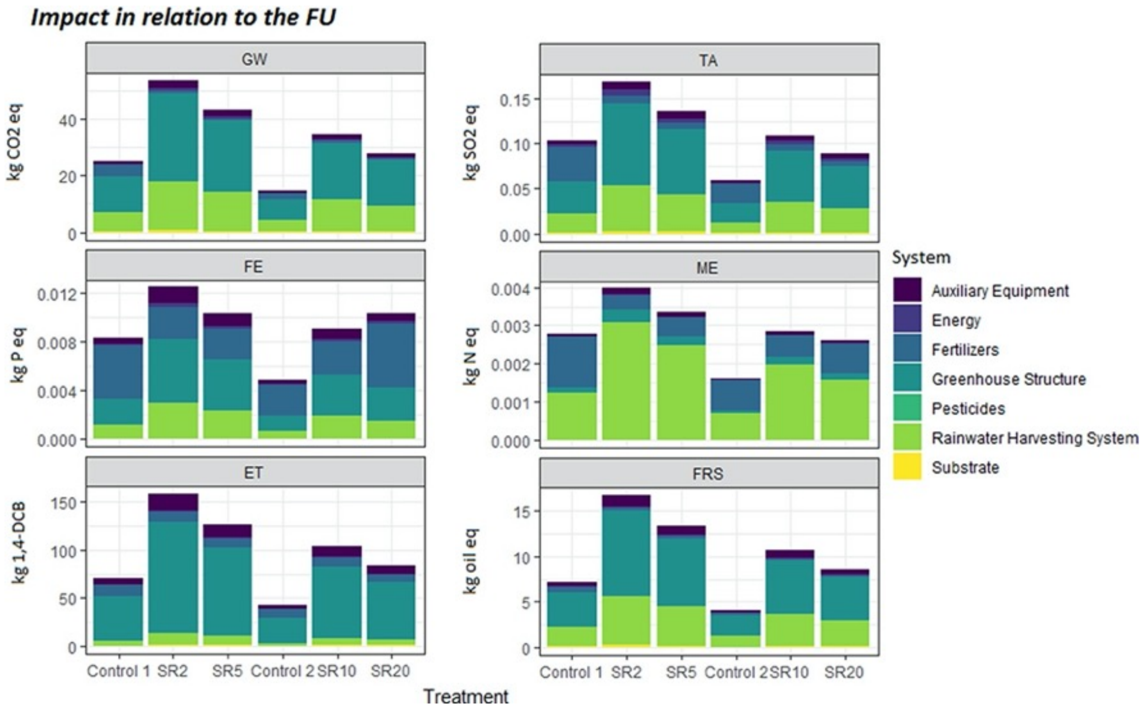


Figure 7. 4 Total System and operation impacts in relation to the functional unit.

Within each impact category we can clearly state that the greenhouse structure and the rainwater harvesting system account for most of the generated impact especially in GW, TA, ME and FRS. This can be due to the large transport distances, the processing and construction of larger amounts of materials like aluminum and steel.

While the auxiliary equipment and fertilizers seem to have lower impacts in most categories, the implication of the latter in the ME, FE and TA categories is of great importance for the control treatments where a full nutrient solution was used. Even when the emissions to air and water of struvite were taken into account, the reduction in these categories for treatments SR2 and SR5 is especially clear.

While the higher production in the control treatment reduces impacts of the RTG- infrastructure and RWHS, the impact generated by the fertilizers is still greater in all IC for the control despite the higher yields, only being surpassed in the treatment SR20 for FE. The percentage contribution of the accounted system stages can be further observed in figure 7.5. The reduction of the impact generated by the alternative fertilizer can be seen when comparing the smaller percentages for fertilization in the treatments SR2, SR5 SR10 and SR20 to the control treatments 1 and 2. For the treatment SR20 we can again observe an increase of the impact in the FE category.

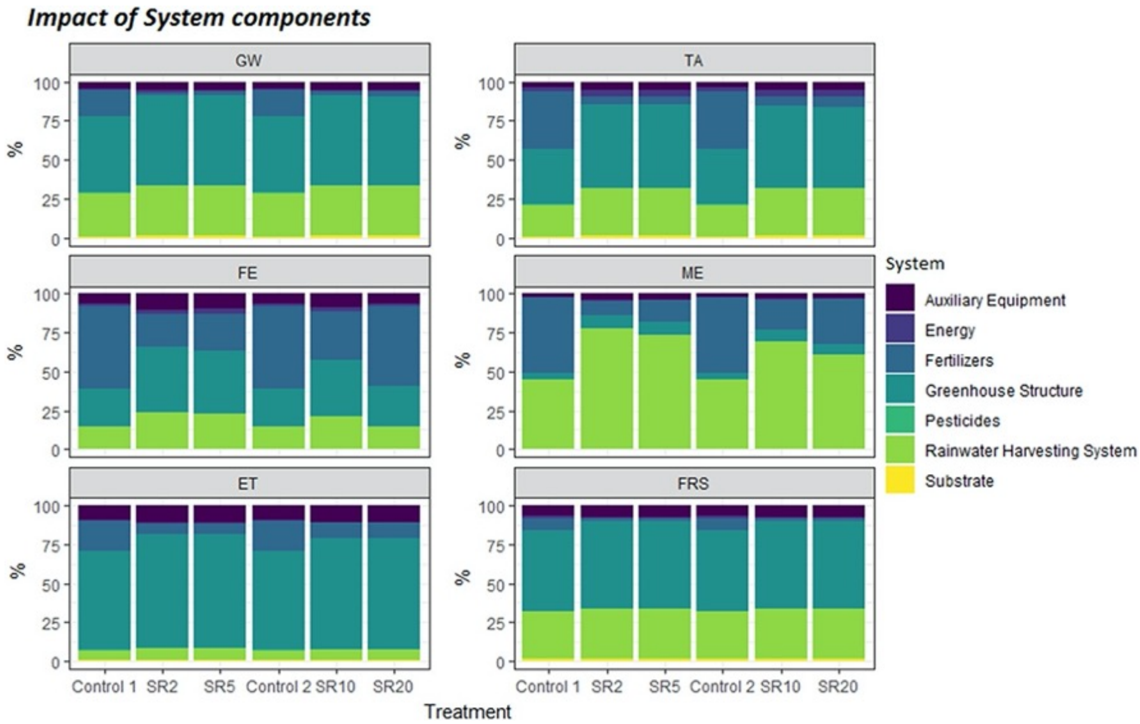


Figure 7. 5 Impact of system components (%).

When adding the percentages corresponding to the infrastructure (RTG-structure, RWHS, auxiliary equipment) and operation (energy, fertilizers, pesticides, and substrate) stages of production for each impact category, a shift of the weight of impact contribution with the alternative fertilization can be seen (table 7.4).

Table 7. 4 Emission origin in our experiment from Infrastructure or Operation of the System in each impact category (IC).

IC	SR2		SR5		Control 1	
	Infrastructure	Operation	Infrastructure	Operation	Infrastructure	Operation
GW	96%	4%	95%	5%	82%	18%
TA	90%	10%	90%	10%	59%	41%
FE	76%	24%	73%	27%	45%	55%
ME	90%	10%	85%	15%	51%	49%
ET	91%	9%	91%	9%	79%	21%
FRS	95%	5%	95%	5%	89%	11%
IC	SR10		SR20		Control 2	
	Infrastructure	Operation	Infrastructure	Operation	Infrastructure	Operation
GW	95%	5%	95%	5%	82%	18%
TA	89%	11%	88%	12%	59%	41%
FE	66%	34%	47%	53%	45%	55%
ME	80%	20%	70%	30%	51%	49%
ET	89%	11%	89%	11%	79%	21%
FRS	95%	5%	95%	5%	89%	11%

The change in the fertilization mainly generates a shift in the eutrophication impact categories (FE and ME) which reach up to more than 55% of the total impact of the operation in the control treatment whilst staying under 30% in both treatment SR2 and SR5. It is also worth mentioning that overall, the change in the fertilization has an effect on all categories, shifting the weight of the impact from the operational phase to the infrastructure when comparing the control to all other treatments.

Due to the great percentage taken up by the greenhouse structure and rainwater harvesting system, the contribution of the operational side of the bean production is overshadowed. Since the infrastructure remains the same for all treatments and is highly specific to this particular site, it was excluded from consideration in figure 6 for a better exploration of the effects of the substituting fertilizer (Figure A7.9 in the Annex II for Environmental performance of the operation system in percentage per IC).

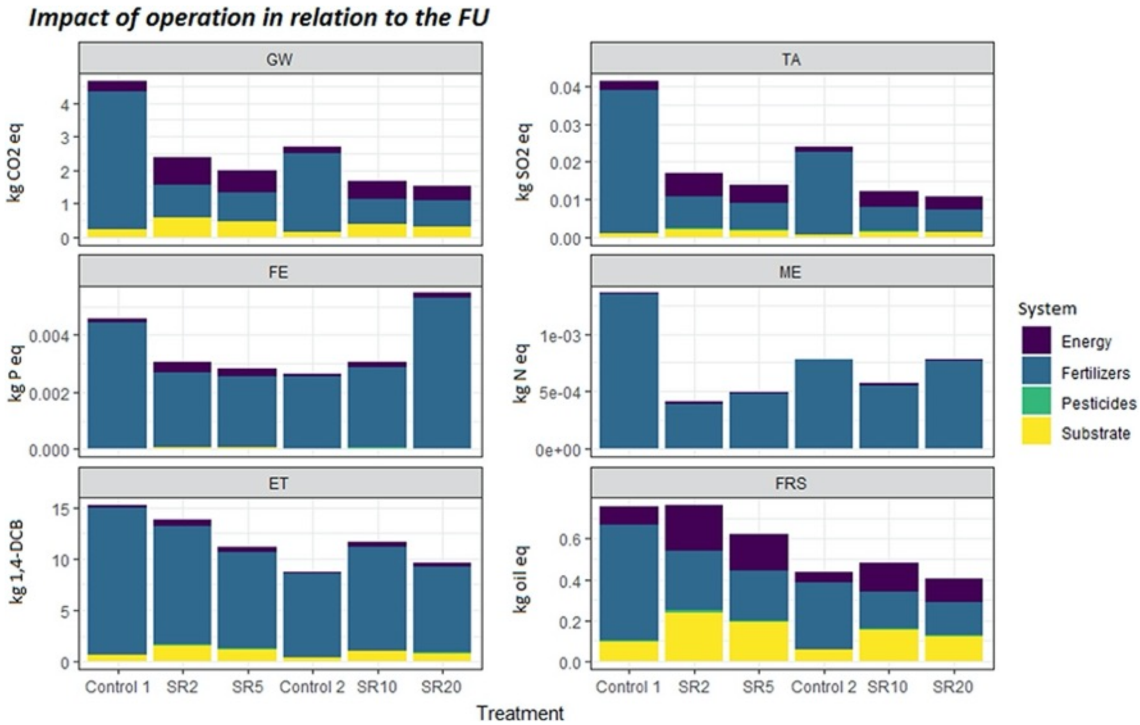


Figure 7. 6 Impact of the production operation in relation to the functional unit.

When observing figure 7.6 the applied fertilization appears to be the main cause for emissions in all IC in the operation subsystem. While yield is smaller in all four struvite and rhizobium treatments, emissions remain lower than controls 1 and 2 in categories GW and AT. In the case of FE, ME ad ET emissions from SR2 and SR5 remain below the control 1 treatment while being higher for the treatments SR10 and SR20. In the case of fossil resource scarcity, it is worth mentioning that the reason for a higher emission in these two categories is not bound to fertilization but due to an increase of the weight of substrate and energy in the operation impact. While emissions are mostly lower in the four alternative fertilizer treatments, especially in SR20 we can observe an increase with greater amounts of struvite in both categories FE and ME. To further understand the changes in emissions bound to fertilization, figure 7.7 depicts all accounted factors considered in the LCA for the fertilization.

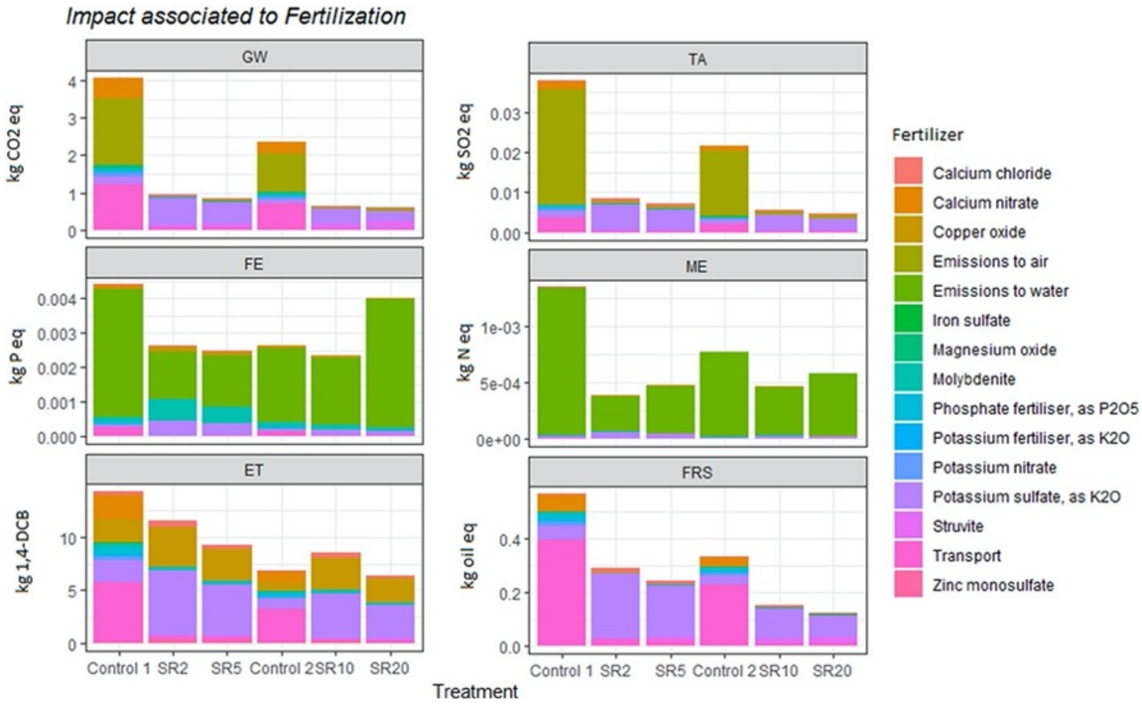


Figure 7. 7 Impact associated to the treatment fertilization in relation to the functional unit.

When observing the fertilization emission in figure 7.7, great impact reductions are made due to the reduced emissions to air and water (as seen in TA, GW, FE and ME) and transport of fertilizers (as seen in ET, FRS and again GW). Here we can appreciate that the increase of impact seen for treatment SR20 achieved in categories FE and ME is due to water emissions increase due to greater struvite quantities.

7.4 Discussion

The life cycle assessment performed on the bean production experiments in soilless substrate fertilized with struvite and rhizobium has shown that there are significant benefits in terms of eutrophication. These findings confirm studies of other authors such as (Rufí-Salís et al. 2020; Sanjuan-Delmás et al. 2018) in which fertilization has been deemed as a major contributor to the environmental footprint of urban agriculture. However, because the yield is lower than that of the conventional mineral fertilizer, the impacts associated to the infrastructure required for the fertilizer substitution increase. Fertilization has shown to be of great importance in the impacts regarding our bean production. The sole removal of the nitrogen, phosphorous and magnesium from the fertigation has shifted the weight of the emissions from the operational part to the infrastructure in a drastic manner. These emission reductions not only affected the expected IC (ME and FE) but all due to their transport (for GW, ET and FRS) and the emissions to Air (for TA) as seen in figure 7.7. While this information depicts great flaws in the implementation of such production systems it also gives a great chance for improvement. While the application of struvite has shown to fulfill the entire cycle of the crop with some yield reduction its production and transport do not affect the given IC to a greater extent except for the treatment SR20. The accumulation of all three fertilizers (P, N and Mg) in one and the possibility of its local generation and application can considerably improve the operational footprint of our agricultural systems. While the struvite recovery technology developed by Ostara® and used for this experiment requires inputs of MgCl, energy and NaOH, has been seen to have lower environmental impacts compared to other processes (Rufí-Salís et al. 2020) further advancements are being made on the use of saltwater, to further reduce the use of chemical resources and potentially lowering its environmental footprint (Amann et al. 2018; Hasler et al. 2015; Martínez-Blanco et al. 2014).

On the other hand, as we have seen in the nutrient fluxes for the SR2 and SR5 treatments, the percentages of the balances for P and Mg remain low as well as the SR10 and SR20 leachate fluxes. The previously described slow dissolution of the struvite fertilizer (Bhuiyan, Mavinic, and Beckie 2007; Degryse et al. 2017b) has been identified as the reason for the lower balance percentages, leaving struvite in the bag that has still not been diluted. This dissolution could be remedied with a lower pH in the irrigation as well as an increase of the irrigation points inside the bag. The location of the struvite itself with regard to the root area has also been regarded as relevant for its plant uptake (Degryse et al. 2017a). On the other hand the remains of struvite inside the bag can favor the reuse and recycling of the perlite bag for a less P and Mg demanding crop in the case of treatments SR2 and even a second production of beans like in the case of SR5. A second bean production without the addition of struvite would even further reduce the needed inputs and its operational footprint.

The use of alternative fertilizers like struvite avoids the consumption of mineral or synthetic fertilizers (Lam, Zlatanović, and van der Hoek 2020) described as fertilizer offset accounting. The environmental benefit of the use of struvite should not only be accounted in the moment of its use (emissions to air and water) and transport but in the avoided production of N, P, and Mg fertilizer. Even further, the environmental benefit of the removal of these nutrients from urban waterbodies should be taken into account as well. As described before, the generation of struvite has requirements but removes potential water and air emissions from WWTP (Igos et al. 2017; Ishii and Boyer 2015a; Lam, Zlatanović, and van der Hoek 2020). While this last benefit has not been taken into account for this study, the further use of struvite as fertilizer and the consequent fertilization offset accounting have, and can be well observed in these results with the emission reductions in almost all IC.

The yield reduction in all treatments compared to the respective controls has a great impact on the production footprint. While a higher production has been reported with a greater application of struvite, a limitation to reach greater yields still remains. Plausible explanations for these losses are the reduced struvite dissolution (Degryse et al. 2017a), the higher P requirement due to the rhizobia symbiosis(Long 1989; Olivera et al. 2004) or a possible electrochemical imbalance causing a reduced uptake of cations in the root area as described by Kontopoulou et al., 2015.

While the yield reduction remains unclear, its impact on the environmental performance is quick to be identified in the obtained results. The loss of production increases the environmental footprint of production, reaching higher emissions than the control treatments (especially when infrastructure is considered), even with lower total emissions of P an N to water in all treatments (Table A7.3 in the Annex II).

A higher yield without the additional use of these fertilizers would decrease the impact of our production, which begs the question as up to what yield loss percentage we can afford and still remain more sustainable than the control treatment.

To answer this question the control (2019) yield was regarded as our hypothetical 100% yield and therefore a scenario with 0% yield reduction. From here on the emissions for all IC were calculated with a yield loss of 10% to 60%. The values used for the 60% and 50% yield loss where directly taken from the SR2 and SR5 treatments respectively, since the obtained yields corresponded to the simulated yield losses. The emissions were also derived from the SR2 and SR5 treatments, since the emissions to water from treatments SR10 and SR20 were deemed as too similar to the control with no major yield increase (in comparison to the respective control treatment).

Figures 7.8 and 7.9 depict these scenarios for the yield reduction impact in infrastructure and operation and only operation respectively. The control treatment line in both figures 7.8 and 7.9 is where the baseline from the control treatment was set. Above this control line the emissions are increased with regards to the control (in %), while below this line the impacts are decreased. When considering the infrastructure and operation (figure 7.8) we can observe that no yield can be lost in order to bring all IC under the control treatment line.

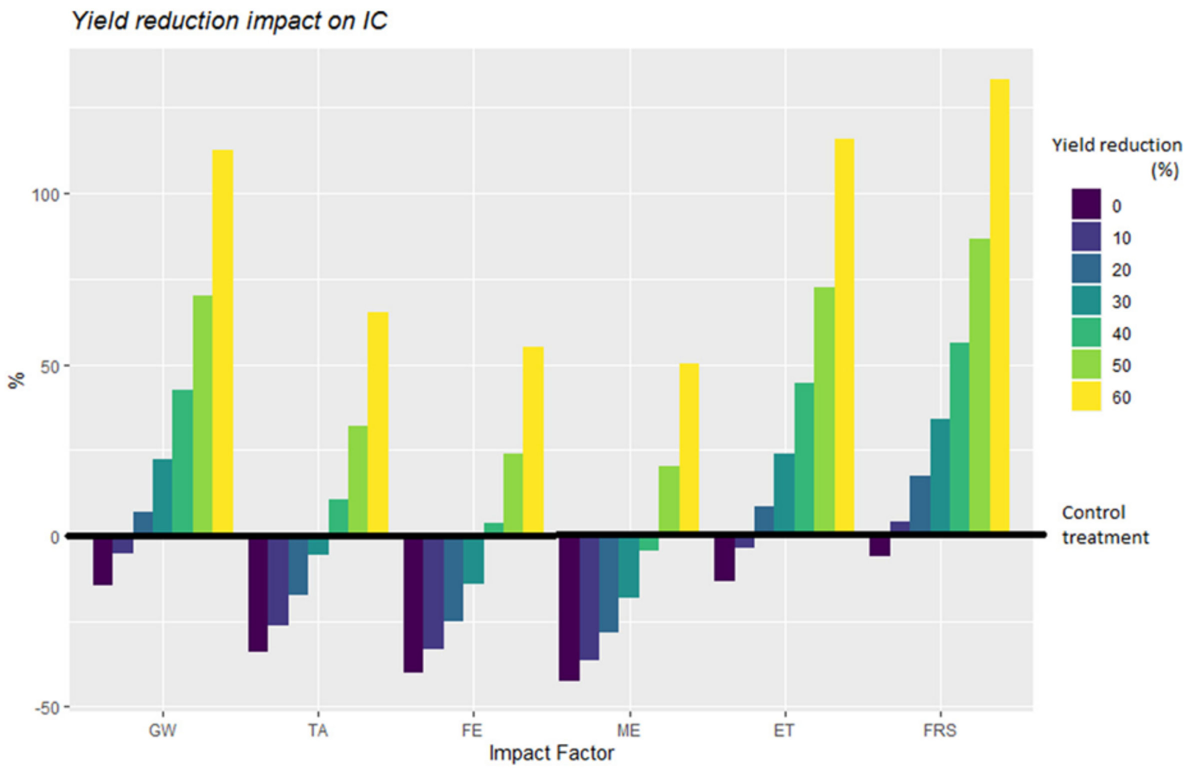


Figure 7. 8 Yield reduction (in %) impact on IC (in %) for infrastructure and operation systems.

While TA, FE and ME are well below the baseline with a yield reduction of 30% other IC like GW and ET start to decrease at 10% yield loss and below. In the case of figure 9, when infrastructure is not considered, a 50% yield loss can occur and still decrease all emissions in all IC.

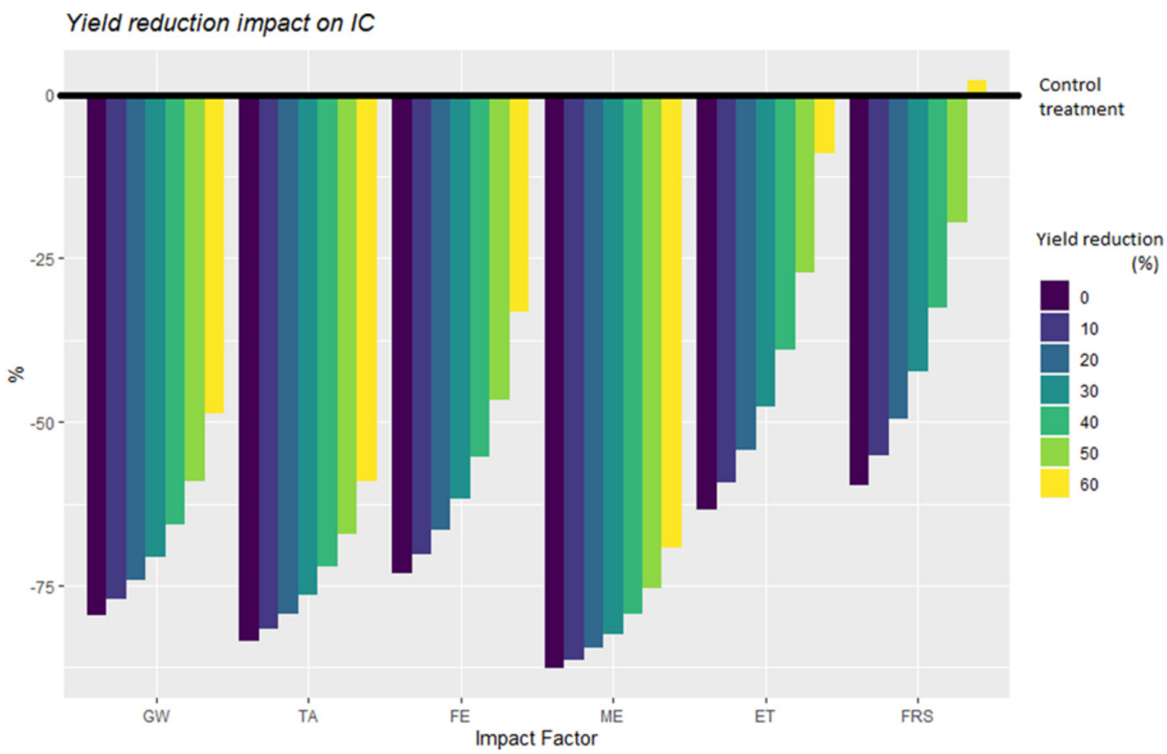


Figure 7. 9 Yield reduction (in %) impact on IC (in %) for operation system.

The importance of reducing mineral or synthetic fertilizer to avoid emission in all IC has been regarded throughout the experiment, although the importance of maintaining production levels while using alternative fertilization methods has been laid out clearly. While impact categories like FE and ME are greatly decreased, the capacity to affect other categories in a significant way can only be achieved with low yield reductions. Especially when considering urban agriculture, the production system might entail more complex infrastructure (rooftop greenhouse, indoor agriculture), leaving reduced margins of yield loss.

The slow dissolution of struvite and the feasibility of its use in soilless agriculture make this fertilizer a good candidate to avoid further P extraction and loss. Due to the uncertainty of the estimation of available mineral P in the next centuries, new ways of nutrient recovery need to be considered in our immediate future (Alewell et al. 2020). This work has demonstrated that the emissions to water, especially for P can be reduced in comparison to conventional fertilization methods. When talking about the extraction of these nutrients to produce struvite further efforts should be made to make this process possible in wastewater treatments plants, reducing transport emissions of agricultural fertilizers to surrounding urban and agricultural areas. Emissions related to transportation of said minerals can be ultimately reduced as well as avoiding emission and loss of nutrients into urban water streams in WWTP (Carpenter and Bennett 2011; Harder et al. 2019). Ultimately, the capacity to recover P nutrients in local scales can reduce agricultural pressures to obtain said fertilizer in due to its distribution or market instability and increasing prices (Alewell et al. 2020; Kataki et al. 2016).

7.5 Conclusions

The present work mainly aimed to analyze the environmental impact of a bean production with the use of alternative fertilizing methods of struvite and the inoculation of rhizobium bacteria. It also aimed to study the feasibility of production with the mentioned change in fertilization, exploring different potential yield losses to gain a broader view of the possibilities this methodology presents. To this purpose, two experiments with different struvite quantities were made and a quantification of the environmental impact using life cycle assessment as our tool. Three main conclusions can be drawn from this study:

Firstly, the total reduction of nitrogen and phosphorous mineral and synthetic fertilizers for vegetable production has been shown to be viable with the use of the recycled slow-releasing fertilizer struvite and the bacterial inoculation with rhizobium strains. Although a yield reduction in all cases was observed compromising its efficiency to reduce the environmental impact in all IC except FE and ME.

Secondly, the use of struvite and rhizobium inoculation reduced emissions in all IC mainly due to transport and emissions to air and water due to a slower dissolution in the substrate. The struvite, being available in all WWTP installations can be obtained with no great environmental cost in its operation while reducing transport and extraction of three separate minerals (N, P and Mg).

Thirdly, the complexity of the infrastructure and operational inputs will increase the environmental impact in all IC, as well as the yield loss. Only the reduction of yield loss up to 0% can equal the environmental impact of the control treatment in all selected categories when the infrastructure is considered. Without the infrastructure the margins for yield loss can range up to 50% staying below the control treatment. Therefore, we consider crucial to reduce infrastructure complexity in the prospect of urban agriculture as well as the reduction of mineral and synthetic fertilizers to truly reduce potential environmental impacts.

Authors contribution:

Verónica Arcas-Pilz: Conceptualization, Methodology, Formal analysis, Data curation, Writing-Original draft; Martí Rufí-Salís.: Methodology, Data curation, Writing - review & editing; Felipe Parada: Methodology, Writing - review & editing; Xavier Gabarrell: Conceptualization, Methodology, Writing - review & editing, Supervision; Gara Villalba: Conceptualization, Methodology, Writing - review & editing, Supervision, Funding acquisition.

Part V

Discussion of the main contributions, General Conclusions, Further research

Chapter 8

Discussion of the main contributions

Chapter 8: Discussion of the main contributions

This chapter discusses the main contributions of this dissertation and the potential implications to improve urban agriculture. This dissertation emphasizes three main dimensions from which to understand and evaluate urban agriculture: Agronomic, Environmental, Circularity as shown in figure 8.1.

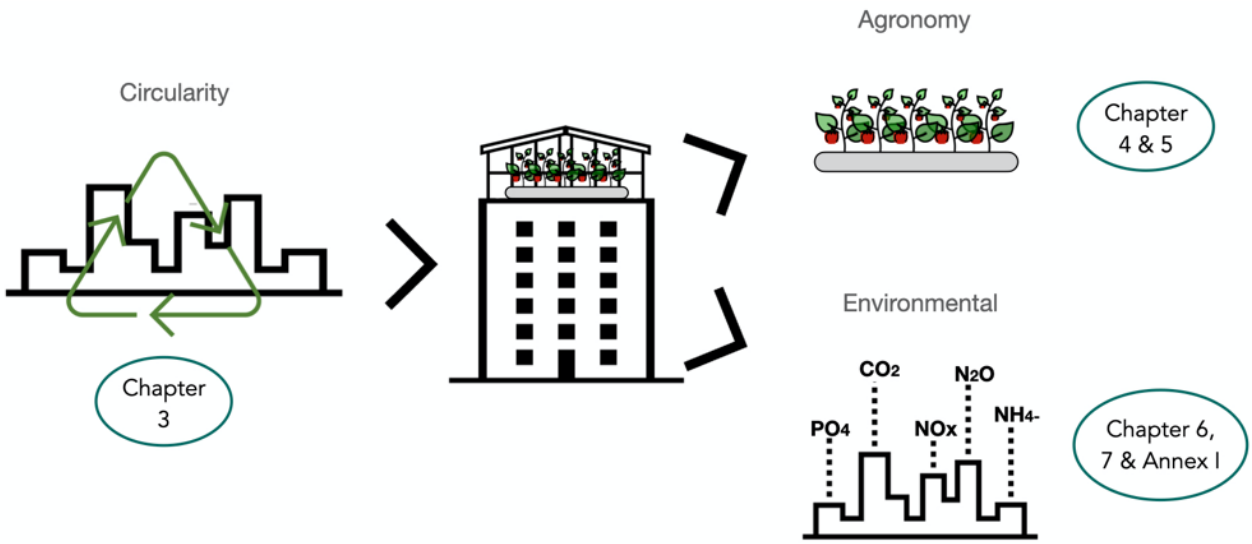


Figure 8. 1 Overall scheme of the 3 focus points of the dissertation: Circularity, Agronomy & Environment (based on Figure 1.10)

8.1 Circularity

Circular economy has been considered a tool to improve the sustainability of urban environments, being the principal objective the recovery of locally sourced resources to be further used in the same area. In this sense UA has been deemed as a useful activity to increase urban circularity (figure 9.2). This circularity not only contemplates the potential of local food production like expressed by Sanyé Mengual (2015), avoiding the importing of goods, but also the use of urban resources as compiled in **chapter 3**. Previous literature on the recovery of resources in urban areas deals with different materials to be used as substrate (Grard et al. 2018) as well as technologies to recover nutrients from different sources. The work made in **chapter 3** compiles and summarizes the state of the art of nutrient recovery technologies using the AMB as a case study to discuss the possibility of providing sufficient locally sourced P and N nutrients for the existing or studied urban agriculture sites. This work gives a glimpse of the potential of urban settlements to supply nutrients for local production by the sheer magnitude of bio, organic and food waste as well as wastewater produced each year. This work also discusses the potential of UA to help reduce the impact of waste in the environment by decreasing the nutrient load for crop growth. **Chapters 4 to the Annex I** on the other hand, provide experimental work on the use of struvite in hydroponic agriculture, being struvite a byproduct of WWTP. All chapters also dwell on the benefits of UA and city connexon evaluating emission reduction to the environment with the use of struvite, especially **chapters 3, 6, 7 and Annex I**.

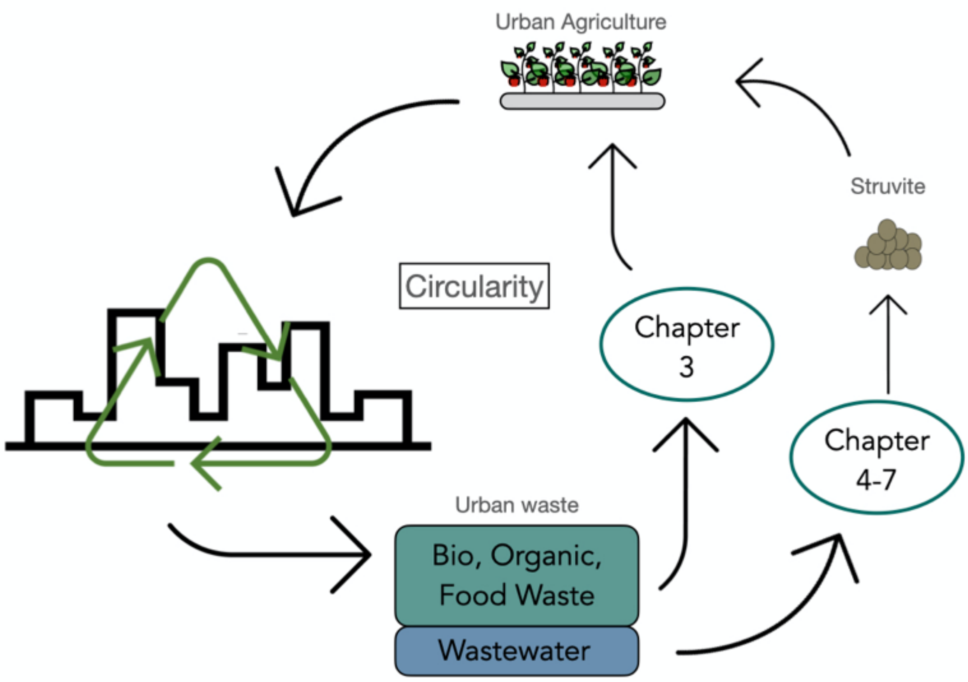


Figure 8. 2 Contributions of the present dissertation on the focus point circularity.

8.2 Agronomic contributions

The increasing population has put great pressure on global resources being further stressed with the loss of arable land and the need to reduce impacts associated to agriculture (IPCC 2019). The present situation calls for alternatives in the agricultural production system, remaining highly efficient and productive at the same time to respond to the increasing food demand. This last point is a crucial theme of this dissertation relying heavily on experimental work across **chapters 4 to the Annex I** that pretends to understand the potential of struvite as an alternative to commercial fertilizer observing the plant development and production for different crops, with different amounts and different production times (figure 9.3). The use of struvite in hydroponic production with perlite substrate was novel in the experiment performed for **chapter 4** providing a first idea of the usability, P uptake and yield production of green bean plants with granulated struvite. The use of different amounts of granulated struvite and its placement are valuable information to further determine struvite usability in hydroponic agriculture. While previous work had determined field applications of struvite in soil-based agriculture, using different dissolution strategies the **chapters 4 to the Annex I** chapters show an adequate dissolution of granular struvite in perlite under neutral pH conditions and commercial irrigation strategies. Early struvite experiments with the replacement of compound P fertilizers showed an increase of N emissions to water due to the N content in the struvite crystal. Through **chapters 5 and 7** the combination of struvite and rhizobium inoculation was tested to further replace synthetic nitrogen application as well as P. This combination was regarded suitable due to recent studies that observed a better rhizobium nodulation with an initial supply of N. The combination with the struvite was observed to determine if the N content of the crystal could increase the rhizobium nodulation while being small enough to avoid its inhibition. This combination was also a novelty for alternative fertilization methods in hydroponic agriculture, showing an increasing nodulation with greater struvite applications related to the available P and N.

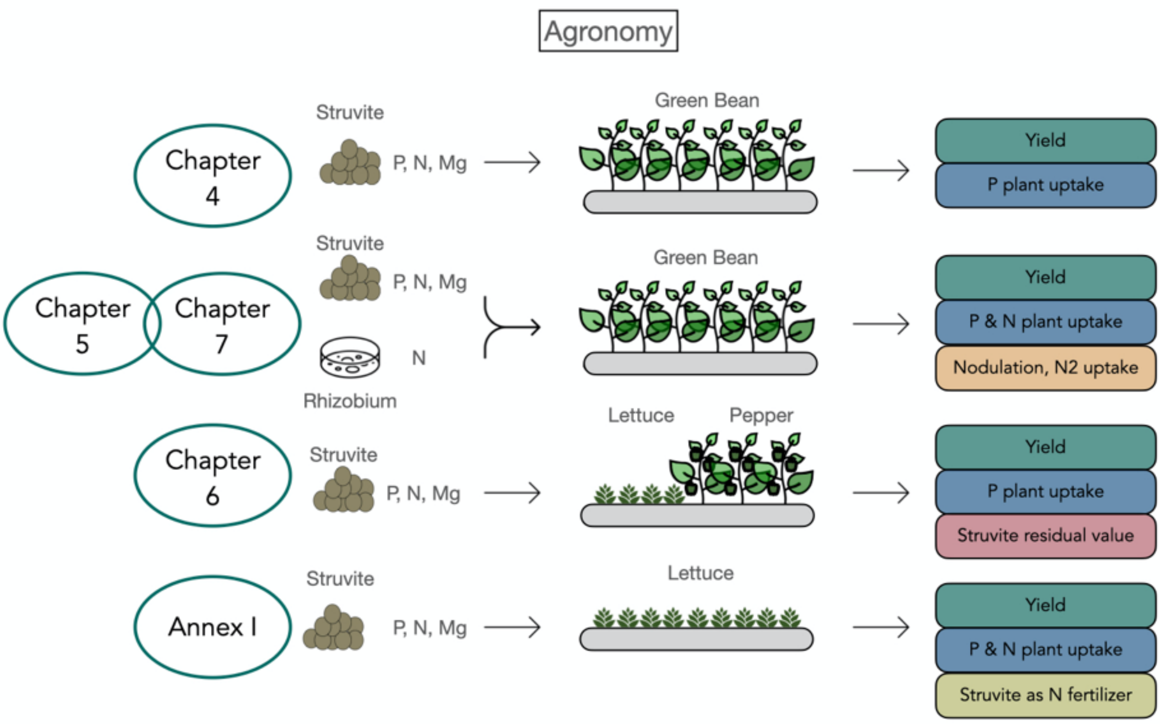


Figure 8. 3 Contributions of the present dissertation on the focus point Agronomy.

The use of struvite in **chapters 4, 5 and 7** gave further understanding of the crystal dissolution and plant uptake, showing a great potential of the struvite residual value (Rech et al. 2018a). This residual value had been expressed before and was further explored in **chapter 6** using of long and short cycle crops as well as consecutive cropping with the same initial struvite fertilization. **Chapter 6** conveys information of struvite fertilization on leafy crop lettuce and fruit production of pepper crop, adding to the pool of knowledge of struvite uses and P uptake in different crops. Through this chapter an estimation of annual lettuce production was made concluding that 20g of initial struvite can sustain 9 consecutive cycles of lettuce with similar estimated yields to the compound P fertilized crops.

Finally, the **Annex I** chapter explores the potential of the contained N in the struvite crystal in leafy crops, like lettuce in the performed experiment. The synthetic N was entirely substituted by the struvite N to identify the growth of crops without additional N and P. This experiment further enabled the exploration of struvite N emissions to the environment as N_2O being the sole N source of the crop.

8.3 Environmental contributions

The use of compound soluble fertilizers in hydroponic production while being a common practice has been regarded as one of the main practices increasing the overall system footprint in previous environmental assessments, especially ME, FE and ET indicators (Sanjuan-Delmás et al. 2018). This increase is mainly due to the fertilizer production and extraction in the case of N and P respectively as well as their emission to the environment into freshwater and marine water ecosystems. The use of fertilizers is responsible for the emission of derived GHG like the case of N_2O through N fertilization (Beltran et al. 2022) which have increased drastically in the last three decades (IPCC 2014).

The use of struvite, a locally recovered slow-releasing fertilizer, opens new possibilities to reduce the environmental impact of hydroponic agricultural systems (figure 8.4). First approaches to P water emissions with the use of struvite are provided in **chapter 4**, where a notable reduction of P concentration in the leachates can be observed. Providing a P mass balance, this study also gives a good understanding of the P flows in a green bean crop production, showing the P concentration in water runoff, plant uptake and most importantly the remaining undissolved struvite in the substrate.

The life cycle assessment performed in **chapter 7,** based on experimental data from **chapter 5,** analyzes the environmental impact of using struvite combined with N_2 fixing bacteria Rhizobium as fertilization for N, P and Mg. This study shows significant benefits in terms of eutrophication with the removal of P, N and Mg from the nutrient solution providing these nutrients solely with struvite and rhizobium inoculation. This further confirms the claims that fertilization can be a major environmental constrain for the sustainability of hydroponic production (Rufí-Salís et al. 2020; Sanjuan-Delmás et al. 2018). The removal of N, P and Mg from the nutrient solution generates a shift from emission weight from operation to infrastructure drastically, reducing emissions in ME, FE, GW, ET, FRS as well as emissions to air in TA for the operation. The obtained yield for the treatments in **chapter 5 and 7** suffered a reduction for all treatments compared to the control. Estimations made in **chapter 7** show that no yield loss compared to control can be suffered to reduce emissions in all IC if infrastructure is taken in account, while up to 50% of yield loss can be assimilated and still reduce emissions in all IC if infrastructure is not considered.

The LCA performed in **chapter 6**, focusing on the use of different struvite quantities in lettuce and pepper crops, gives further information on the environmental potential of this slow releasing fertilizer, especially its residual value for short and long cycle crops. Environmental emissions for all IC are reduced for the pepper crop when 10g and 20g of struvite are given, while lettuce crops experience an increasing emission of N and therefore higher impacts for ME for all treatments. The lettuces fertilized with 20g of struvite showed sustained yields when making a 1-year prediction showing reduced impacts in all IC except ME. The work made in **chapter 6** gives environmental significance to the struvite residual value showing that an initial fertilization with 20g can reduce impacts in the crop fertilization for most IC.

Finally, the **Annex I** chapter discusses the possibility of emission reduction with struvite as sole input of N and P in lettuce production, observing reduced emissions to water for both nutrients. The emissions to air caused by N fertilization in hydroponic production systems had been assessed previously, determining a reduced N_2O emission in perlite substrate with lettuce crops (Llorach-Massana et al. 2017). On the other hand, the N_2O emission factor of struvite had not been determined to date for hydroponic production with perlite substrate.

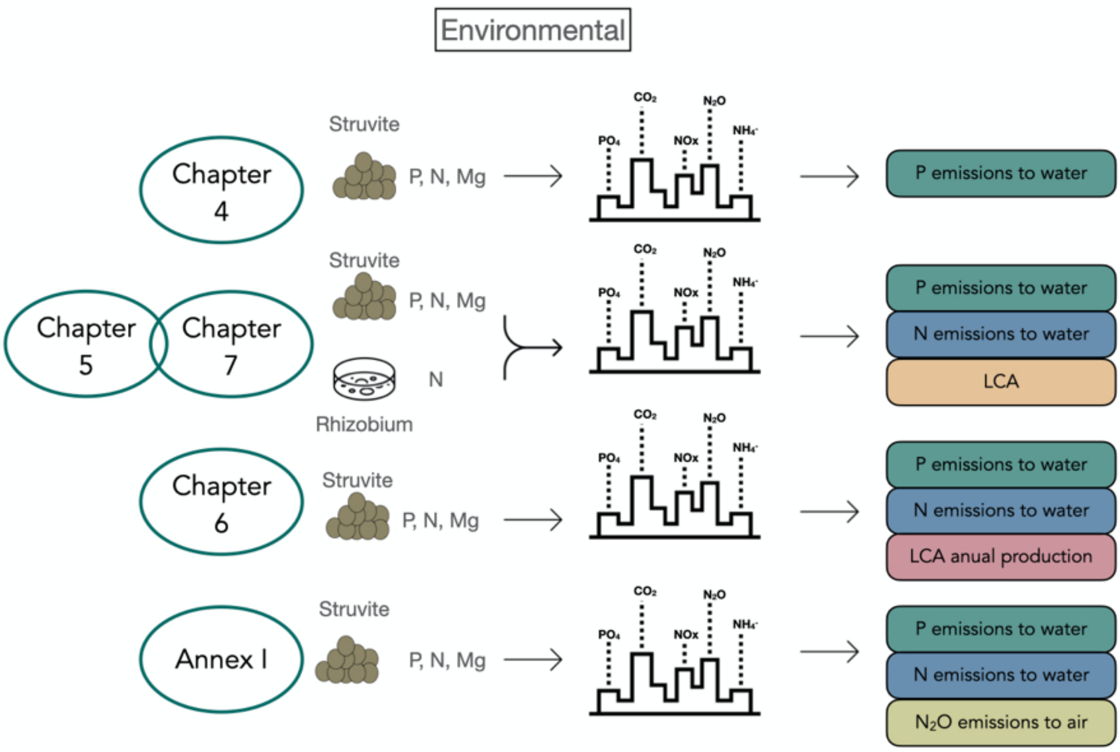


Figure 8. 4 Contributions of the present dissertations on the focus point Environment.

8.4 Global contributions and UA dissemination

The three focus points of this dissertation can be considered separately but increase the value of the obtained contributions when aggregated. The present dissertation aims to assess the possibility of locally resourced fertilizers like struvite in UA from various angles, granting a better understanding of their potential advantages and disadvantages.

While **chapter 3** gathers the potential of nutrient recovery and supply for UA **chapters 4, 5 & 6** determine the actual agronomic feasibility which is further evaluated in **chapters 6, 7 & Annex I** for the actual environmental benefit this change in fertilization could suppose.

Chapter 9

General conclusions

Chapter 9: General Conclusions

This chapter presents the general conclusions of the present dissertation providing comprehensive responses to the questions raised in Chapter 1.

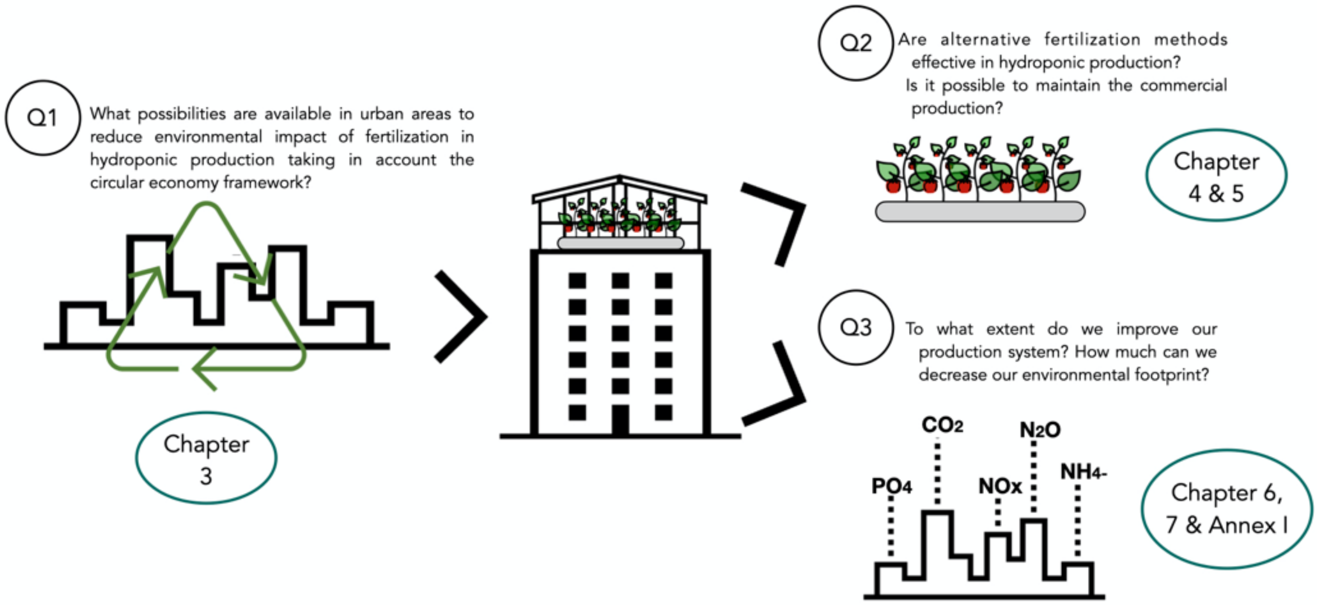


Figure 1. 10 Illustration of the research framework and research questions with the corresponding chapters of this dissertation

> Q1 → What possibilities are available in urban areas to reduce environmental impact of fertilization in hydroponic production taking in account the circular economy framework?

Due to the impacts associated to fertilization production like in the case of synthetic nitrogen or the mining of P as well as in the nutrient disposal and impact in the environment a reduction of the impact should be assessed from a circular point of view. This would entail the avoidance of nutrient loss into the environment and its subsequent use in agriculture avoiding further nutrient production. Additionally, this circularity enables the local provision of nutrients avoiding additional transport and nutrient shifts.

To answer the research, question this dissertation includes a literature review approach (**chapter 3**) to understand the current technologies available to recover and recycle nutrients in urban areas. As proposed in the question raised the circular economy framework invites us to focus on the existing waste flows, mainly food, bio, organic waste, and wastewater. The specific search under these two categories resulted in 5 recovery strategies for organic- bio- and food waste and 11 for wastewater. With the current organic waste and wastewater recovery scenarios in the

AMB it can be concluded that the amount of N and P that can be repurposed into UA can cover the current and potential needs of new installations.

Q2 → Are alternative fertilization methods effective in hydroponic production? Is it possible to maintain the commercial production?

This dissertation has focused on the use of struvite as P fertilizer in hydroponic production, providing a great range of experimental outcomes for crop yields on different crops during different production systems as well as in combination with other fertilization methods. The application of the WWTP byproduct showed to be simple and adequate for substrate-based hydroponic systems.

The performed studies with struvite as the sole P fertilizer for green bean (**chapter 4**) showed successful productions with yields equal or higher than the control treatment when more than 5g of struvite was given per plant. This first approach showed a successful application of struvite as a form to supply P for green bean plants. Yields with the greatest amount of 20g tested exceeded the control productions by +37%, while still showing reduced concentrations of P in the leachate streams.

This dissertation further indicates the potential of struvite to fertilize consecutive production cycles due to its residual value (**chapter 6**), obtaining competitive yields for 3 consecutive lettuce cycles with initial quantities of 5, 10 and 20g of struvite. Showing comparable productions to the control treatment specially for the 20g fertilized crops with fluctuating productions of -7% to +9% compared to the control. While the lowest production was given with 5g of struvite with a -20% yield reduction compared to the control. One 3-month long pepper production cycle showed exceeding productions of +40% above the control treatment for crops fertilized with 20g of struvite. After three experimental cycles the remaining struvite still entailed 50-70% of the initially applied slow-release fertilizer, therefore considering an increase of the consecutive production cycles to a year's worth production (9 cycles). The estimated production for 9 lettuce cycles showed a similar, even higher production (+9%) for lettuces fertilized with 20g of struvite while the lowest production occurred for 5g the treatment with a -21% yield reduction.

The production of green bean with struvite combined with N fixing bacteria Rhizobium (**chapters 5 & 7**) to provide both N and P where considered the only experiments where a yield reduction was detected in all treatments. The yield production was related to the initial struvite provided showing -60% losses for plants with 5g and -42% losses for plants fertilized with 20g compared to the control productions. It is although worth mentioning that all plants showed sufficient concentrations of nitrogen, being potassium the limiting factor in the plant nutrient content. Nodulation was found in all treatments, indicating good compatibility between struvite and rhizobium, even showing greater nodulation with greater struvite amounts. Further experimentation with higher struvite amounts and a more precise control of the electrochemical composition of the rhizosphere is therefore encouraged for achieving commercial yields.

Which the obtained information it can be concluded that struvite is a suitable and effective fertilizer in hydroponic production systems, with equal or even higher productions than commercial fertilization methods, especially the application of 20g of struvite in green bean crops, lettuces and peppers has been promising.

Q3 → To what extent do we improve our production system? How much can we decrease our environmental footprint?

While the production of a crop might be affected using a certain fertilization method, this research question asks for a bigger picture of the system and not only demands an evaluation of the crop yield but the actual environmental footprint of said production.

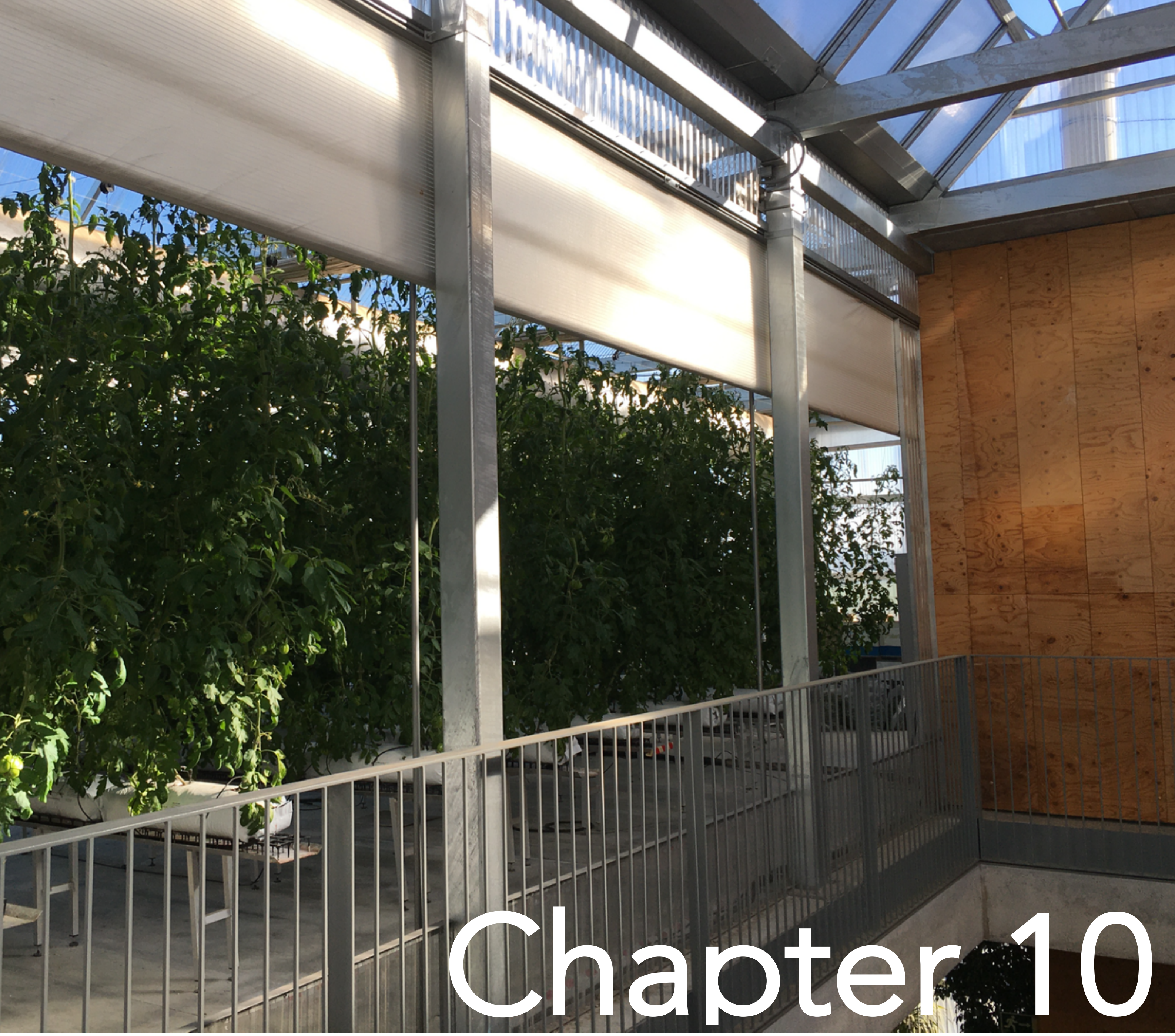
This dissertation provides an answer to this question on three different levels.

On a fist level we have the analytical proof of the direct use of struvite as a fertilizer. This analytical proof was provided during the experimental phase, recording the struvite dissolution and nutrient concentration in the leaching water. When focusing solely on P losses into the water (**chapter 4**), the slow-releasing nature of struvite has shown useful remaining mostly in the growing substrate, while providing sufficient P to the plant. In comparison, the green bean crop experiment showed that 68% of the total P fertilizer given in the control treatment was discharged into the leachates after being applied. This can again be seen in longer production cycles for pepper crops where a very reduced P nutrient discharge was detected during the 3-month recording of leachate compared to the control treatment. Consecutive lettuce cycles showed increasing emissions of P into the discharged water during early plant stages and after plant sowing gut still remain at least 3 to 5 times lower than the concentrations for the control.

Secondly, this dissertation provides environmental assessments with the use of the Life Cycle Analysis tool. While the analytical results of the water flows are also considered in this assessment all impacts of the struvite fertilization in the hydroponic system are considered. The obtained results in the LCA assessments performed in **chapter 6** for 3 and 9 consecutive lettuce cycles show not only a reduction of the environmental impact related to the emission of P to freshwater (FE), but also a reduction of emissions related to mineral extraction (MRS). Due to the local obtention of the P nutrient a reduction in the impacts related to CO_2 (GW) emissions and fossil resource consumption (FRS) are also reduced with the avoided long transportation. The additional emissions of chemical agents into the environment (TA, ET) related to the nutrient extraction are again further reduced when considering struvite precipitation in comparison to phosphate rock extraction. However, the low production of lettuce fertilized with 5g of struvite had consequently an emission increase in all IC indicating that a fertilization of at least 10g of struvite or higher is needed to reduce the production footprint. The production of peppers showed an even greater emission reduction in all impact categories, even for ME related to N emissions into water, again with crops fertilized with at least 10g and 20g of initial struvite.

The results obtained in the LCA for **chapter 7** indicate that even with a yield loss of 50% and 60% impacts were reduced due to the use of struvite and Rhizobium as P and N fertilizers. Emissions related to P and N experienced and increase at greater struvite applications mainly affecting FE and ET. The combination with the Rhizobium bacteria enables the reduction of emissions related to N fertilizer production, transport and emissions to air and water while at the same time gravely affecting the yield. The application of 2, 5 and 10g were seen as correct to reduce overall emissions.

The third level in which the environmental impact was assessed consisted of the generation of a methodology for the quantification of GHG N_2O emission with the use of struvite compared to a fertirriation with NO_3^-. The low level of emission from this fertilization gives further information and specification of the N related emissions with the use of this slow releasing fertilizer. Although no concrete conclusions can be drawn from the low detection of N_2O emissions in the **Annex I** chapter, this low detection in both fertilization methods confers that there is no great difference between their emissions.



Chapter 10

Further research

Chapter 10: Further research

In this chapter new lines of research are proposed in line with the present dissertation and which results could further complement these findings.

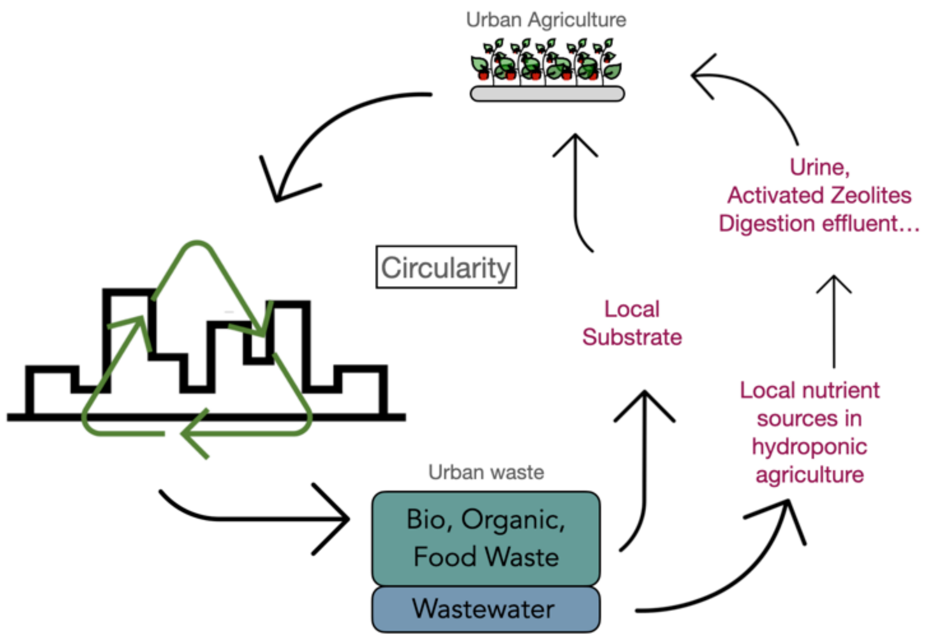
Continuing with the previously proposed structure in chapter 8 the further proposed research is presented in the three focus points: Circularity (figure 10.1), Agronomy (figure 10.2), Environment (figure 10.3).

- Local substrate from urban derived materials

In the case of circularity, it is considered that local waste materials can also conform good local substrate alternatives. By products from urban and peri urban business factories, building and construction business as well as waste from various sources can be further analyzed to determine characteristics and suitability as substrates for urban agriculture. An example of these wastes could be the use of coffee husk waste from coffee roasters, as well as the use of cork waste from wine cork production.

- Local nutrient sources in hydroponic agriculture

While this dissertation mainly focuses on the use of struvite, other nutrient recovery techniques and products have yet to be tested in hydroponic production. The direct use of yellow water from the ICTA-UAB building into the hydroponic system could be a great way to increase circularity of the building while opening new research paths both in the agronomic and environmental aspect.

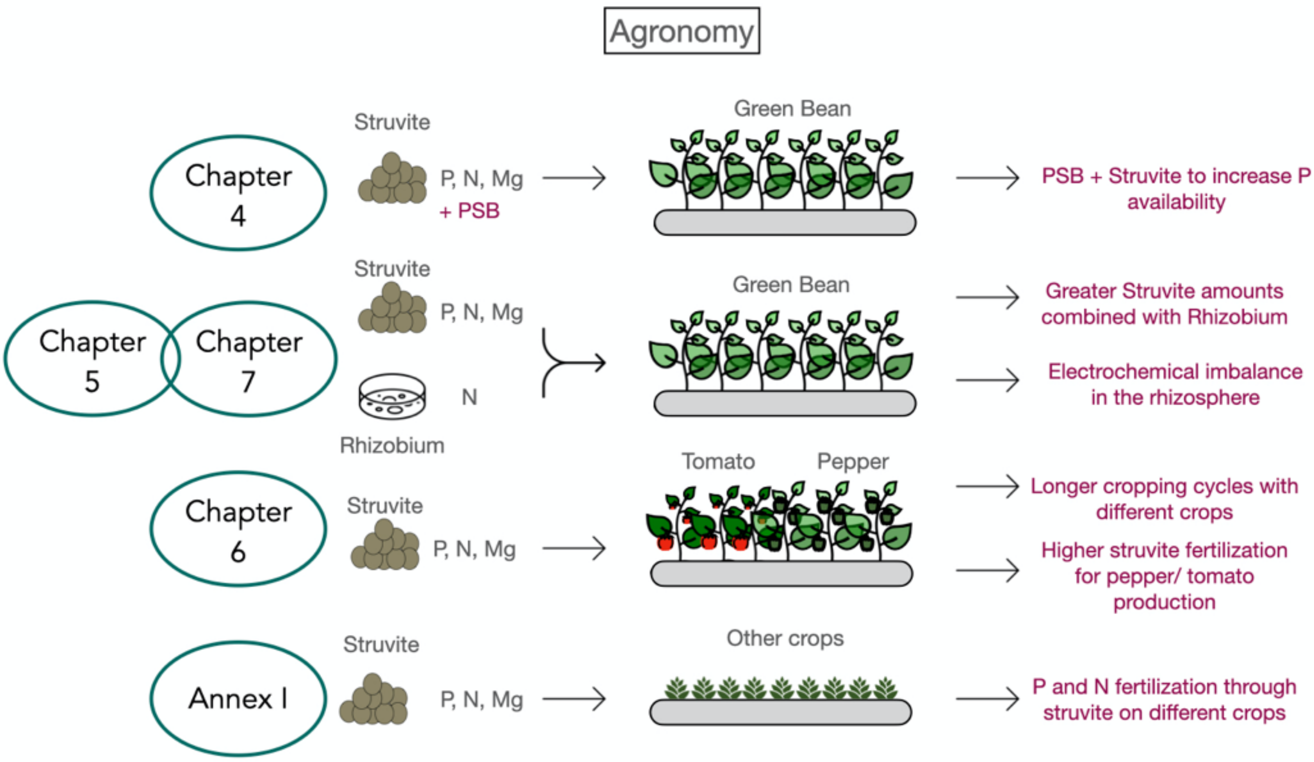


- Phosphate solubilizing bacteria (PSB) + Struvite to increase P availability

The slow dissolution of struvite has been reported to be problematic in early plant growth stages and could be further problematic for highly P demanding crops. While this could be partially solved with an increased addition of struvite the addition of PSB could further increase P solubility without the need of increasing struvite quantities.

- Greater struvite amounts combined with Rhizobium

As seen in **chapter 5 and 7** greater fertilization amounts of struvite resulted in greater nodulation (**chapter 5**) and greater overall yields (**chapter 7**). Still yields remained at least 40% lower than the control treatment. To understand when yield increase can occur while nodulation is not inhibited further experiments with greater struvite amounts can be made.



- Electrochemical imbalance in the rhizosphere

The results in **chapter 5 and 7** pointed out a limiting K and Mg plant uptake in struvite + rhizobium fertilized plants. The deficient K^+ and Mg^{2+} cation uptake was attributed to a electrochemical imbalance in the rhizosphere due to the reduced presence of NO_3^- in the root medium. This can further be assessed with the addition of nutrients in the form of anions to reestablish an electrochemical balance.

- Longer cropping cycles with different crops

While longer cropping and consecutive cropping cycles have been tested for pepper and lettuce respectively other crops such as leafy greens or aromatic plants could be further assessed for longer or consecutive production cycles.

- Higher struvite fertilization for pepper / tomato production

Greatly demanding crops like pepper and tomato could benefit from greater amounts of struvite. This would further add information to the experiments conducted in **chapter 6** with pepper production observing P uptake with greater amounts and potentially producing for longer periods like in the case of tomatoes with cropping cycles of up to 6 months.

- P and N fertilization through struvite on different crops

While results from **Annex I** have shown that N content in the struvite crystal can be sufficient for lettuce production this can be further assessed with other crops with similar N requirements and with higher N requirements to determine the necessary quantity to be given.

- Social perception on struvite fertilization

Current perception and legislation still see nutrient recovery from wastewater in a bad light and while policies might change in the near future the public perception of struvite use in food production can still be a controversial topic. The social assessment of struvite use might be an interesting window into the public opinion to further advance in nutrient recovery strategies or further acknowledge the existing gaps in modern society.

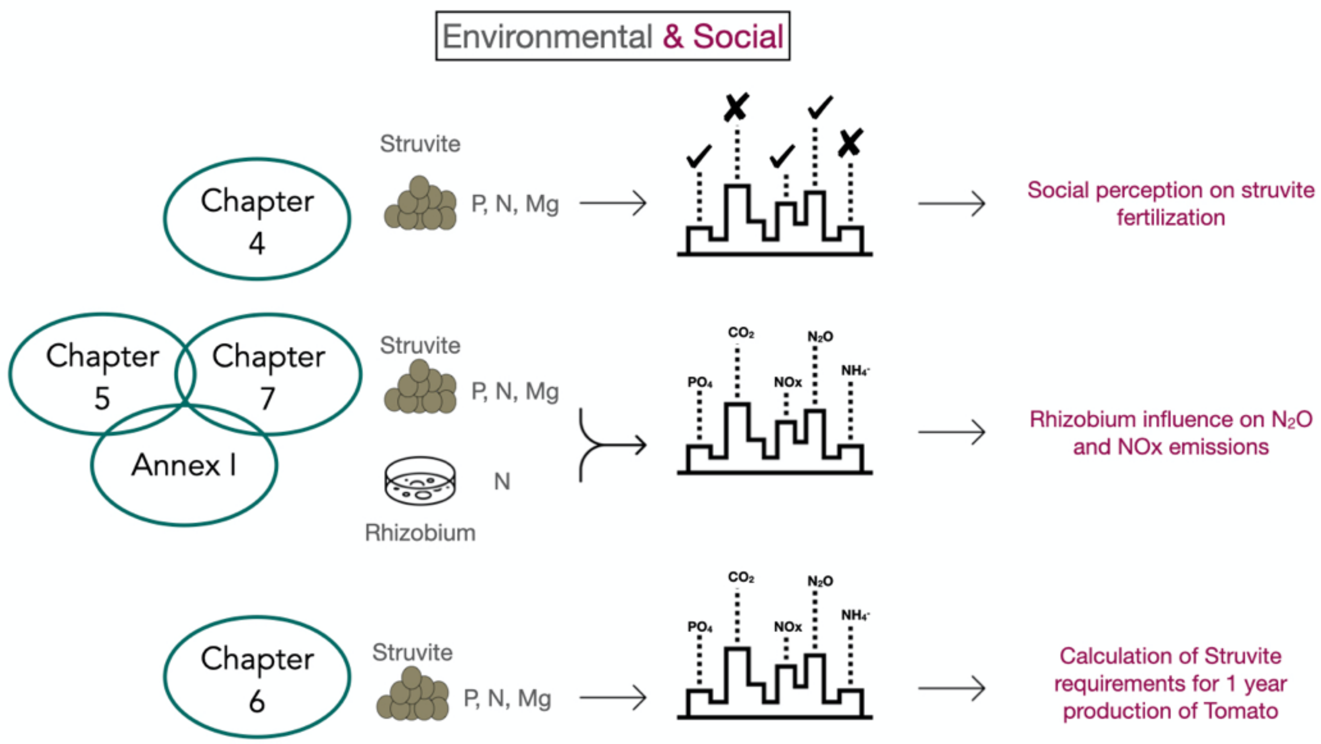
- Rhizobium influence on N_2O and NOx emission with struvite fertilization

While N_2 fixation through rhizobium inoculation has been determined in combination with struvite (**chapter 5**) as well as the N_2O emissions related to struvite fertilization (**Annex I**) no assessment of N_2O emissions was made combining struvite and rhizobium fertilization.

- Calculation of Struvite requirements for 1 year production of Tomato

As performed in chapter 6 a year worth production of lettuce crop was made concluding that 10g and 20g of struvite could be sufficient to sustain commercial productions for 9 consecutive cycles.

The production of tomato is commercially very relevant in Spain as well as a greatly demanding crop when it comes to fertilization needs. The use of struvite in tomato production could reduce environmental impacts of the overall production although struvite requirements for a 1-year worth production should be assessed.



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Annex

Annex I: Methodological approach to N_2O emission with struvite fertilization in hydroponic agriculture

Annex I: Methodological approach to N_2O emission with struvite fertilization in hydroponic agriculture

A1 Introduction

The concept of urban agriculture (UA) has been gaining popularity in the past decades (Appolloni et al. 2021), being regarded as one potential solution to increase food resilience in cities (Zambrano-Prado, Orsini, et al. 2021). The use of urban space like indoor vertical farming and rooftop farming can also help confront the ever-growing food demand without increasing farmland and reducing the transport and production chains from field to consumer (Specht et al. 2014). The use of different production systems like hydroponic, aeroponic and aquaponic are popular among urban farmers due to the greater production density and precision as well as a replacement of the soil with growing substrate or direct contact to a nutrient solution. Avoiding the use of soil, these production technologies can be installed on building rooftops and facades due to their lower weight load (Thomaier et al. 2015).

Hydroponic and aeroponic growing systems rely on the use of synthetic and chemical nutrients which are directly added to the irrigation water. The use of nutrient compounds with NH4+ and NO3- is a common and the origin of these N based nutrients is mostly synthetization through the resource demanding Haber-Bosch process (Chojnacka, Moustakas, and Witek-Krowiak 2020; Galloway et al. 2004; Woods et al. 2010).

Not only the generation of these N based nutrients can be concerning but also their fate when used in conventional and urban agriculture (Hasler et al. 2015; Beltran et al. 2022). N fertilization and over fertilization is commonly associated to eutrophication in water bodies being its cause in 60% of cases in Europe due to its leaching and runoff (UNEP 2011; Halbert-Howard et al. 2021). The GHG N_2O emissions have also increased steadily for the last three decades with a yearly increase of 0.73ppb usually associated to N based fertilization (IPCC 2014).

This GHG has a lifetime of 121 years and a global warming potential (GWP) of about 265 for a cumulative forcing of 100 years. This GWP is 265 times higher than for CO_2 and almost 10 times higher than for CH_4 (IPCC 2014).

The generation of N_2O induced by N based fertilization stems from chemical processes in the soil carried out by the existing microbial communities. In anaerobic conditions and with N availability in the soil in the form of NO_3- or NO_2- microbial denitrification reduces the nitrate and nitrite into N_2, N_2O and NO (Henault et al. 1998). Additionally the process of nitrification can add to the emission of N_2O with N availability in the form of NH4+, forming nitrate which then can further reduced to nitrite and atmospheric nitrogen (Beltran et al. 2022; Dsouza et al. 2021)

When considering the further expansion of UA it is important to contemplate the potential emissions that could be rising from it and could further impact the urban ecosystem.
The use of the crystalline fertilizer Struvite has been gaining interest not only due to its content of P, N and Mg but also due to its slow dissolution and its origin as a byproduct from WWTP (Degryse et al. 2017; Rech et al. 2018).
Previous work has shown promising results in the use of struvite fertilizer in hydroponic agriculture on perlite substrate, also indicating remarkably lower P and N emissions to water compared to commercial soluble fertilizers (Arcas-Pilz et al. 2022; Arcas-Pilz, Rufí-salís, et al. 2021).
While struvite has been used extensively as an alternative P fertilizer it also contains N in the form of NH_4, which could sustain less demanding crops.
While N emissions to water can be reduced with the use of struvite little work has been done on the analysis of potential emissions to air in the form of N_2O. As expressed by Halbert-Howard et al. (2021), the reduced uptake of the struvite by the plant, remaining for long periods of time in the substrate could make it mor prone to be used in microbial processes.
The combination of struvite and another recycled fertilizer vinasse showed increase N_2O emissions compared to urine-based fertilizers "Aurin" and "Crop" as well as the NPK synthetic mineral fertilizer in hydroponic tomato production. This was explained through the addition of organic-C by the vinasse which increases the anaerobic denitrification processes due to microbial respiration (Halbert-Howard et al. 2021).
On the other hand, the N_2O emission when conducting hydroponic agriculture on substrate like perlite and rockwool have been shown to decrease due to greater inert conditions and
The present work aims to add to the pool of knowledge on the N_2O emission factor of struvite in hydroponic production compared to synthetic NO_3^- fertilizer.

A2 Materials and methods

The present experiment was carried out in the rooftop agriculture laboratories in the ICTA-UAB building. These laboratories are located inside the institute building acting as itegrated rooftop greenhouses (i-RTG), with a hydroponic irrigation system for the production of crops.
For the determination of the GHG N_2O the chosen methodology was of a closed chamber build inside the rooftop laboratory in the ICTA-UAB. The closed chamber split in half through an inner wall provides space for 16 pelite filled plant pots (1L) on each side with a total volume of 2,64m^3 (1.32m^3 respectively) as seen in figures X1 and X2. Each side of the closed chamber was equipped

with Cambell sensors for temperature (107 probe; CS215) , radiation (LP02 TR) and humidity (CS215) (CR3000 ; CR1000x data-logger) as well as a sampling inlet.

Annex X. 1 Closed chamber outside view

Annex X. 2 Closed chamber inside view (left side).

A2.1 Irrigation treatments:

The compared treatments for the N_2O emission determination were a struvite fertilized lettuce crop with a N,P,Mg free nutrient solution and a lettuce crop fertilized with a N,P,Mg full solution. The nutrient solution composition can be found in Table 8.1, showing the composition of the control treatment with the nutrient rich solution and the struvite treatment with the amount of struvite given.

Annex X. 3 Nutrient solution table for treatments: Contol and P,N,Mg free nutrient solution with struvite fertilization.

NUTRIENT SOURCE	COMPONENT	g/L	P	N	Mg
CONTROL NUTRIENT SOLUTION	KPO_4H_2	13.625			
	KNO3	10.125			
	K2SO4	30.45			
	Ca(NO3)2	16.375			
	CaCl2*2H2O	11.1			
	Mg(NO3)2	14.83			
	Hortrilon	1			
	Sequestrene	1			
P, N, Mg-FREE NUTRIENT SOLUTION	KPO_4H_2	-			
	KNO3	-			
	K2SO4	43.5			
	Ca(NO3)2	-			
	CaCl2*2H2O	16.6			
	Mg(NO3)2	-			
	Hortrilon	1			
	Sequestrene	1			
STRUVITE	20g		2.5g	1.14g	1.98g

A2.2 Validation experiments:

The lettuce crop was chosen due to its short growth cycle providing the opportunity to generate previous validation experiments on the capability of struvite to act as the sole source of N for the lettuce plant. The validation experiments took place outside and inside the closed chamber and privided further information on the influence of irrigation in the struvite dissolution. Table X.4 contains a summary on the validation and determination experiments made, as well as their location and analysis.

Annex X. 4 Summary on the validation and determination experiments performed. Location is referred as the experiment location in relation to the closed chamber.

Number	*Crop*	*Time*	*Treatment*	*Location*	*Analysis*
Validation experiment 1	Lettuce	27 days	20g struvite + ½ N Normal irrigation Duble irrigation	Outside	Water Biomass
Validation experiment 2	Lettuce	34 days	20g struvite + ½ N Normal irrigation Duble irrigation	Outside & Inside	Water Biomass
Validation experiment 3	Lettuce	34 days	20g struvite Normal irrigation	Inside	Water Biomass Perlite
Determination experiment	Lettuce	34 days	20g struvite Normal irrigation	Inside	Water Air

Different treatments were made to decide the irrigation pattern for the struvite treatment, ensuring sufficient nutrient uptake by the plants. While the control treatment (C) was irrigated with a full nutrient solution validation experiments 1 and 2 had two other treatments fertilized with struvite and ½ N. These two treatments differ on their irrigation being RN a normally irrigated treatment and RD a treatment with duble irrigation.

Validation test 1 and 2 were made outside the closed chamber while validation test 2 and 3 were also performed inside. The treatments inside the chamber were control (Chamber C) and struvite fertilized with 20g and normal irrigation (Chamber St).

These validation experiments were important to identify the capability of struvite to sustain a lettuce crop without additional N from the irrigation.

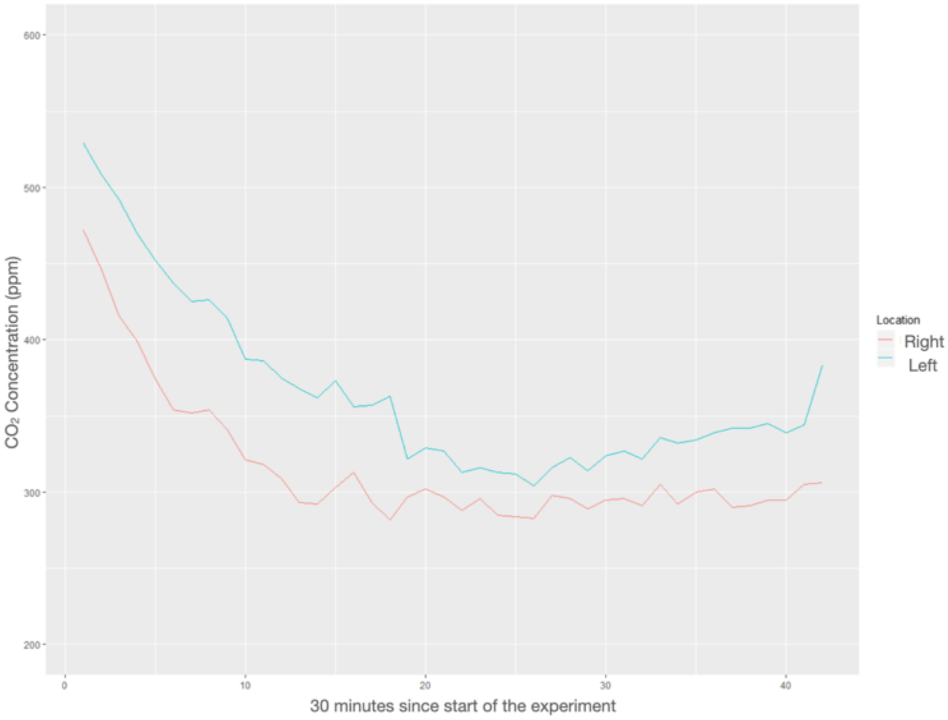
A 2.3 Gas sampling:

Once the lettuce plants were transplanted inside the closed chamber air samples were collected 3 times a week twice a day. This samples were collected through a Vac-U-sampler and an air pump and stored in specialized air tight 1-L air bags (SKC1 – model 1.252e01) (figure X.5).

Annex X. 5 Vac-U-sampler used with a SKC standard air pump

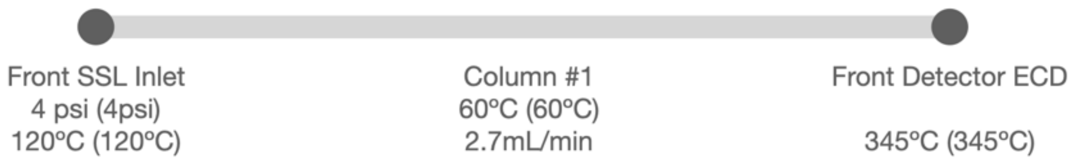
A sampling of each side of the chamber was done, as well as the exterior in the building atrium outside of the cropping area, before closing the chamber.

The chamber tighness was ensured with previus CO_2 measurments in 30 minute intervals with the Ultramat 23 (SIEMENS) multivalve. This tighness experiment was performed leaving the chamber closed with plants inside and masuring the CO_2 concentration during 24hours. The CO_2 was seen to decline in both sides of the chamber stadily with measurements taken every 15 minutes. The CO_2 measurements in the thightness experiment can be seen in figure X.6 with a marked decrease until 10 hourse after closing the chamber.



Annex X. 6 Tightness experiment result for both sides of the closed chamber during 24 hour CO_2 analysis.

A second sampling was done (for each side of the chamber and the exterior) after a predetermined period of time, followed by the opening of the chamber.
The timespan of the closed chamber was chosen to be 1, 3 and 5 hours to procure greater gas concentratios while avoiding givin stress to the crops.
The obtained air samples are analyzed with the Agilent chromatograph 6890N and the Agilent HP-PLOT-Q 30 m, 0.53 mm, 40 mm column injected manually with an air tight Pressure-Lok® Precision Analytical Syringe (VICI).
The chosen method for the gas analysis was the following temperature settings shown in figure X.7, using N2 as carrier gas and He as makeup flow.



Annex X. 7 GC settings used in the present study.

A 2.4 Water sampling:

Water samples were also taken 3 times a week from the incoming irrigation as well as the leachates for both sides of the closed chamber.
The P and N concentrations of the collected water were analyzed in a chromatograph DIONEX-ICS 1000 Ion System with a Dionex Ion Pac AS9-HC RFIC Analytical 4 _ 250 mm column. The volume of incoming and outgoing water was measured daily.

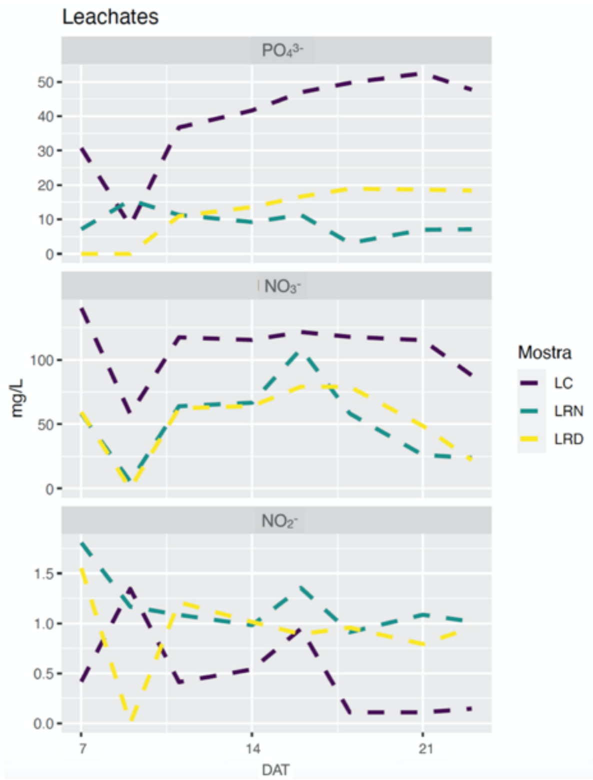
A 2.5 Yield determination and nutrient content:

To perform the nutritional analysis, the plants were harvested, dried and grinded and consequently weighted up to 0.25g with an analytical balance (XPE205DR Mettler Toledo). The digestion was performed in a MARS Xpress (CEM) microwave digestor adding concentrated HNO3. The digested samples were then diluted with HNO3 1% (v/v). All samples were weighted, digested and analyzed per duplicate. The total P content of the lettuce biomass at the end of the crop was determined by an external laboratory, although previouse digestions of said samples were done in the ICTA laboratory.

A3 Results and Discussion

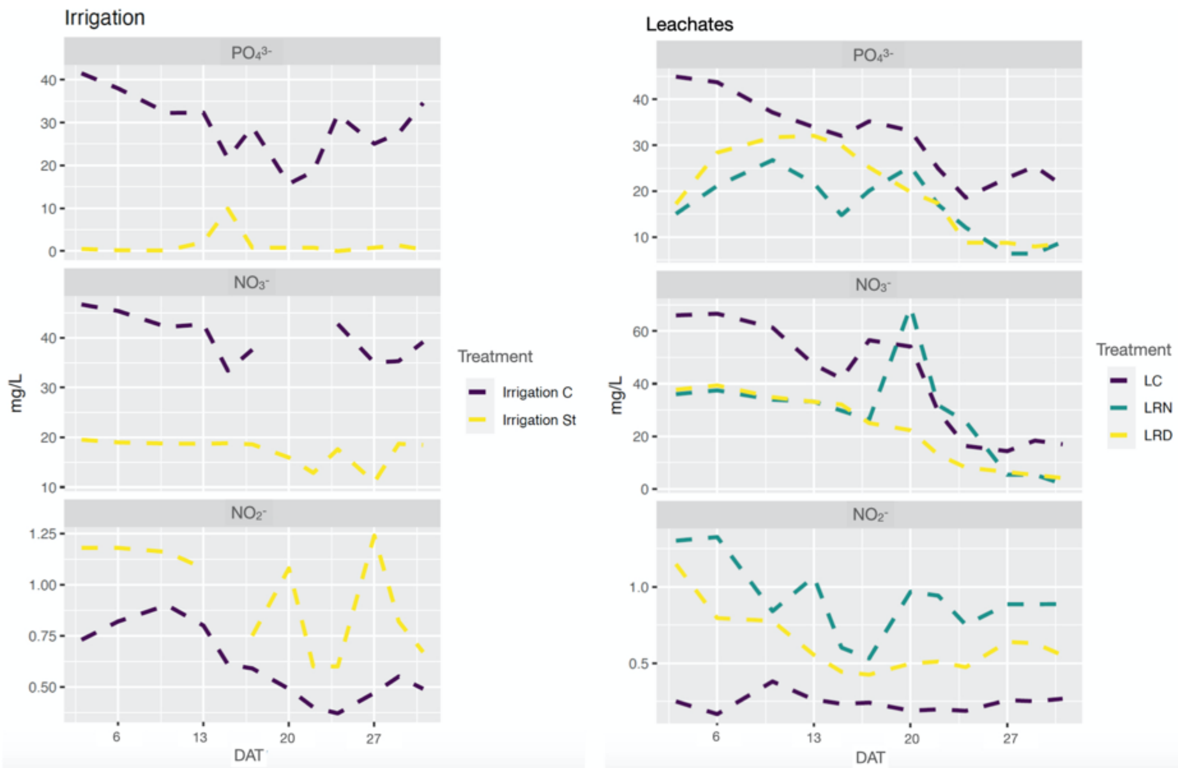
A3.1 Validation experiments:

Validation experiments 1 and 2 still included struvite treatment with ½ the amount of N in the nutrient solution tank compared to the control. This can be seen in the irrigation and leachate water in figures X.8 and X.9, where the control treatment still leachates greater amounts of P but not as clearly for N. The validation test 2 which includes a treatment with double irrigation shows an initial greater amount of P than the normally irrigated treatment. This however is not clearly seen for the N in both nitrate and nitrite.



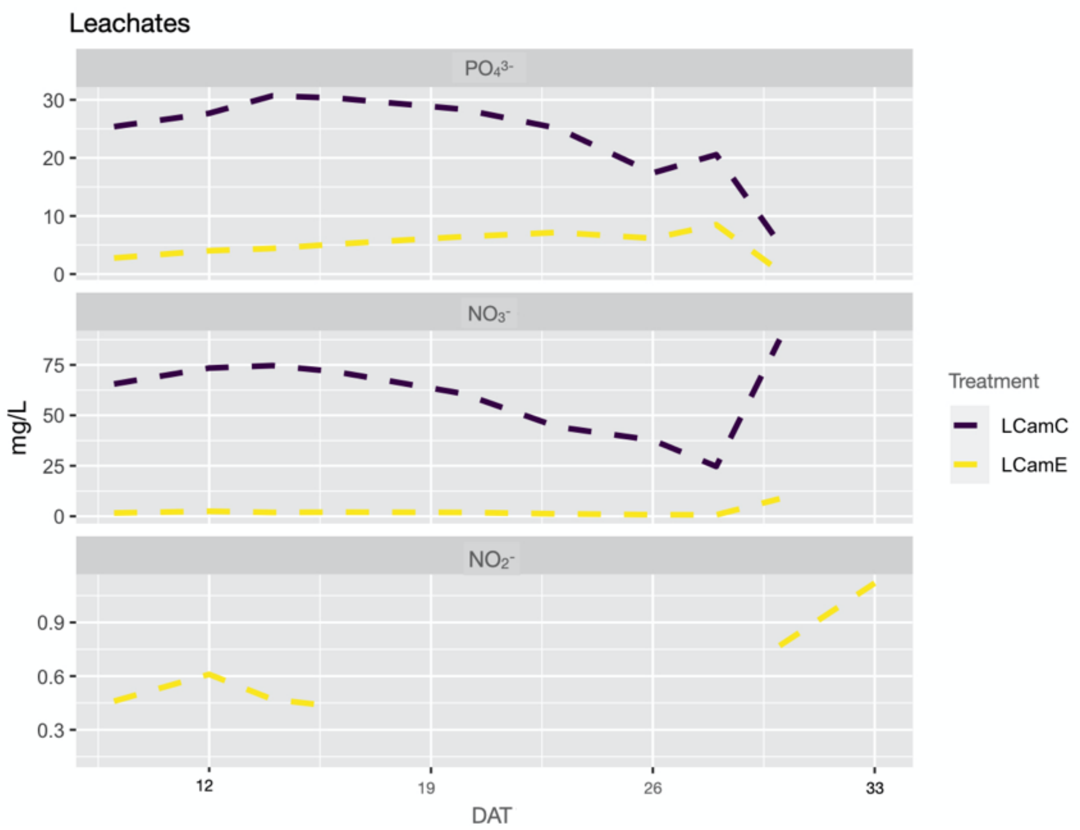
Annex X. 8 Leachate analysis for PO_4^{3-}, NO_3^- and NO_2^- for the validation experiment 1 for the control treatment (LC), normal irrigation treatment (LRN) and double irrigation treatment (LRD).

Figure X.10 shows the P and N (in the form of nitrate and nitrite) content in the leachates for the validation experiment 3 performed inside the closed chamber. This validation experiment was performed with a control treatment (Chamber C) and with a struvite fertilized treatment with an incoming irrigation without P, N and Mg (Chamber St). The low concentrations of N and P in the leachates for the Chamber St treatment compared to the control indicate that a total omission of these nutrient in the irrigation can highly reduce their emission to water, only relying on struvite in the perlite substrate.



Annex X. 9 Irrigation and leachate analysis for PO_4^{3-}, NO_3^- and NO_2^- for the validation experiment 2 for the irrigation with full nutrient solution (Irrigation C), irrigation with 1/2N and P free nutrient solution (Irrigation St.), control treatment (LC), normal irrigation treatment (LRN) and double irrigation treatment (LRD).

The production and nutrient uptake for both validation tests 1 and 2 can be seen in figures X.11 and X.12. The yield from treatment with double irrigation (RD) appear to be greater in both validation experiments outside the chamber. The productions within the chamber in the validation test 2 are highly similar to the treatments control and normal irrigation outside the chamber, giving further information that the chamber infrastructure does not greatly influence the yield.

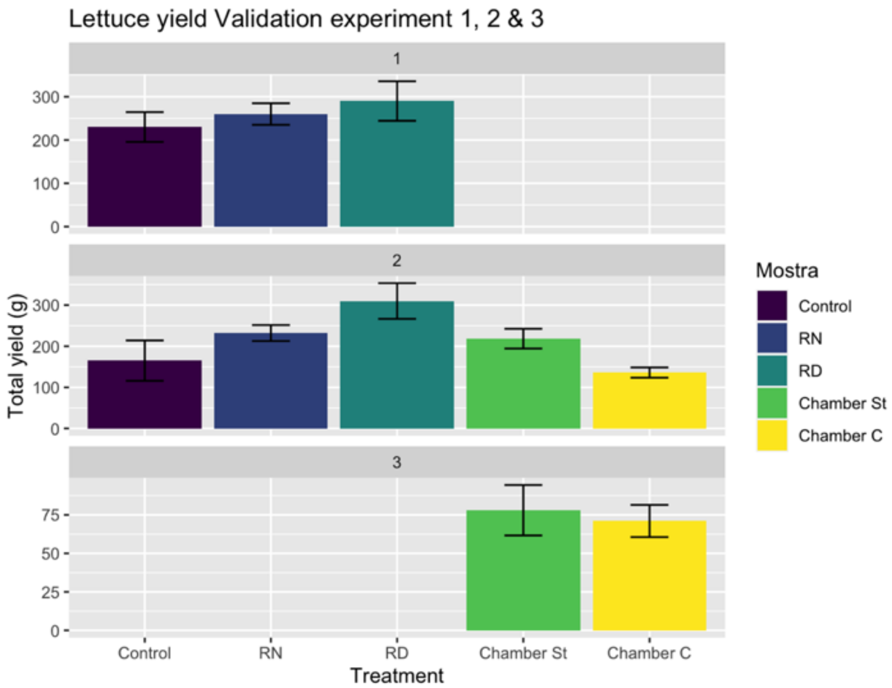


Annex X. 10 leachate analysis for PO_4^{3-}, NO_3^- and NO_2^- for the validation experiment 3 for the) control treatment inside the chamber irrigated with a full nutrient solution (LCamC), and the struvite fertilized (20g) and N, P, Mg free nutrient solution irrigated treatment (LCam E).

On the other hand, the P content in the lettuce produced inside the chamber seems to be influenced as we can see in figure X.12 displaying not only considerably less amount of P in the plant biomass but also a great difference between both treatments.

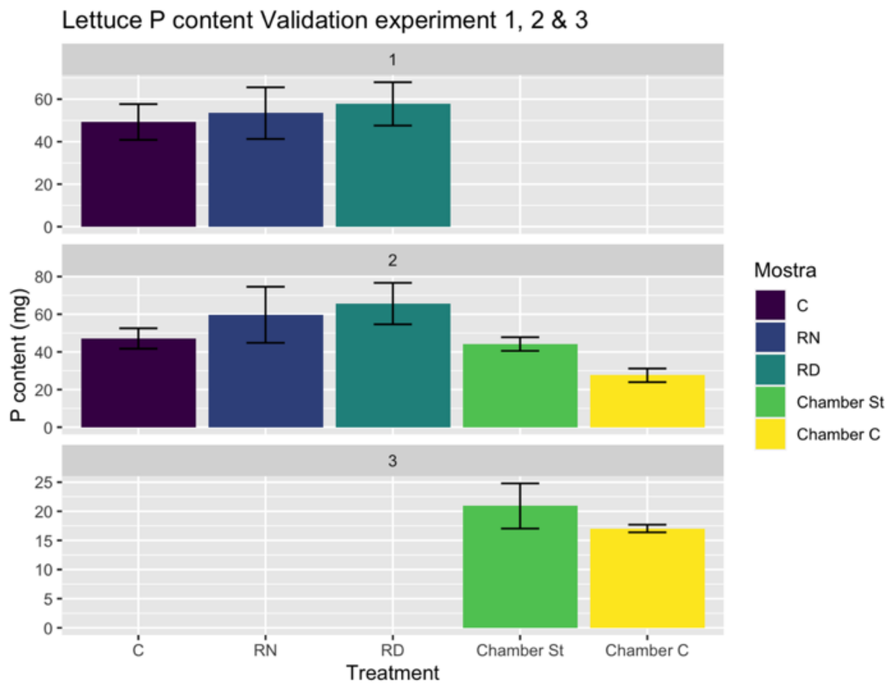
The validation experiment 2 and 3 present productions made inside the closed chamber with different irrigation treatments. While the irrigation for Chamber St in the validation experiment 2 contained ½ of the control added N the treatment Chamber St for the validation experiment 3 does not contain additional N in its nutrient solution relying solely on struvite for P, N and Mg.

While yields in the chamber for the validation experiment 2 are similar to the treatments Control and RN outside the chamber, with less variation within treatments the validation experiment 3 yields seem greatly reduced. This yield reduction can be explained due to the climatic condition during this experimental period. Still productions were higher for the Chamber St. treatment than the Chamber C although differences are not considered significant.



Annex X. 11 Total Lettuce production (g) during validation experiments 1, 2 and 3 for the control treatment as well as treatments with normal irrigation, double irrigation and productions inside the chamber (Chamber C & Chamber St.)

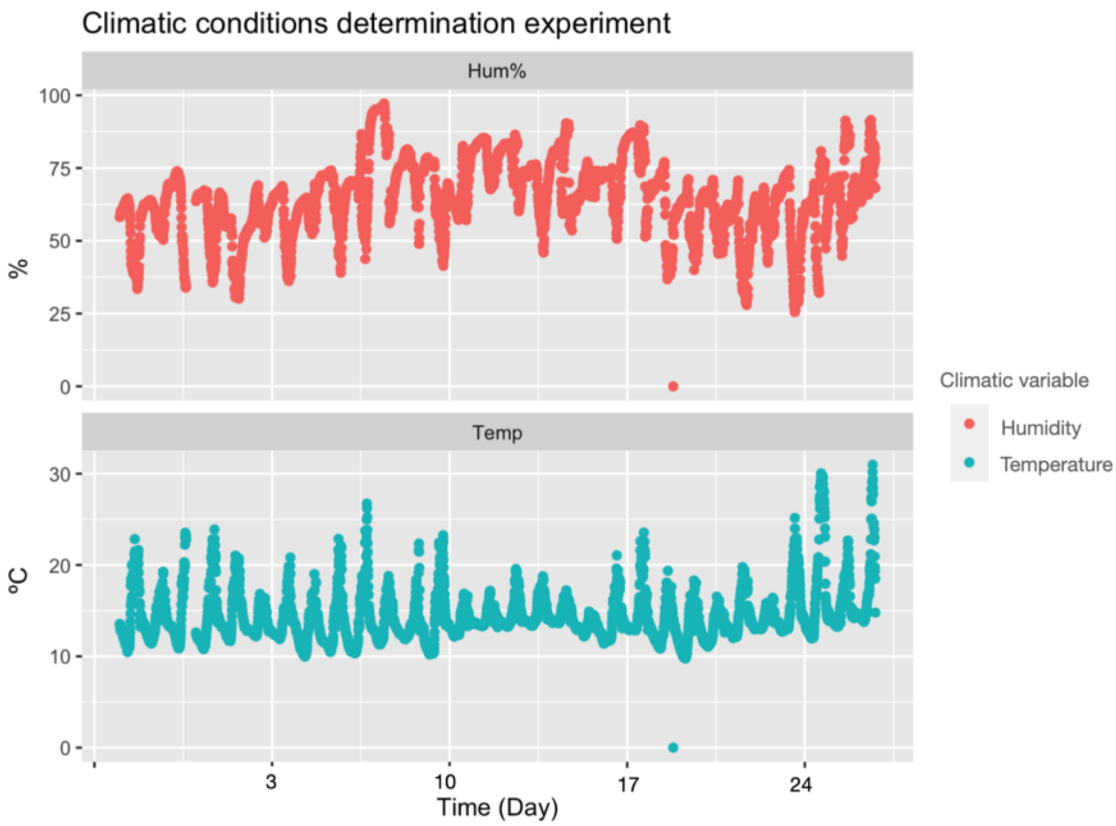
Finally, the results from the validation experiment 3 for the P content in the plant biomass indicates greater content for the Chamber St treatment as seen before in the validation experiment 2. This information, added to the greater yields indicate that the lettuce crop can be sustained with 20g of struvite with a normal irrigation pattern and a completely N,P and Mg deficient nutrient solution.



Annex X. 12 P content (mg) in lettuce biomass during validation experiments 1, 2 and 3 for the control treatment as well as treatments with normal irrigation, double irrigation and productions inside the chamber (Chamber C & Chamber St.)

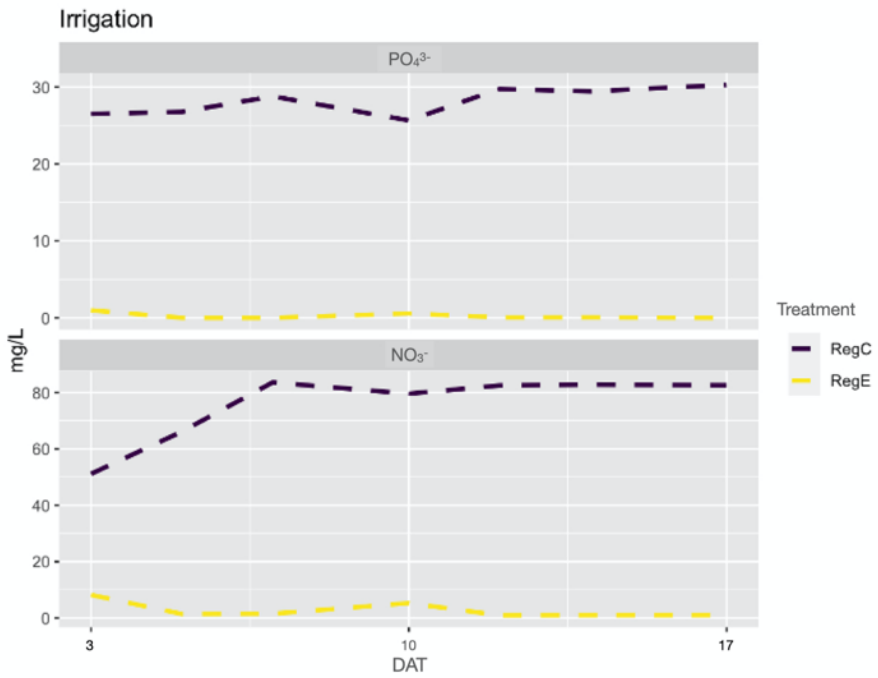
A3.2 Determination experiment:

The climatic conditions recorded for the humidity and temperature of the determination experiment show great variations reaching high levels of humidity close to 100% at certain points (figure X.13. These higher humidity points also coincide with temperature peaks which could mainly appear when the chamber is closed. The chamber increases its humidity and temperature while closed and can sustain these conditions during longer periods if the chamber has been closed for a longer time. While higher temperature has been seen to potentially increase N_2O emissions favoring microbial metabolism the greater humidity can be a factor that could difficult the gas sampling and analysis due to its dilution (Rapson and Dacres 2014). While no humidity filter was used during this experiment it is greatly encouraged in future sampling methodologies.



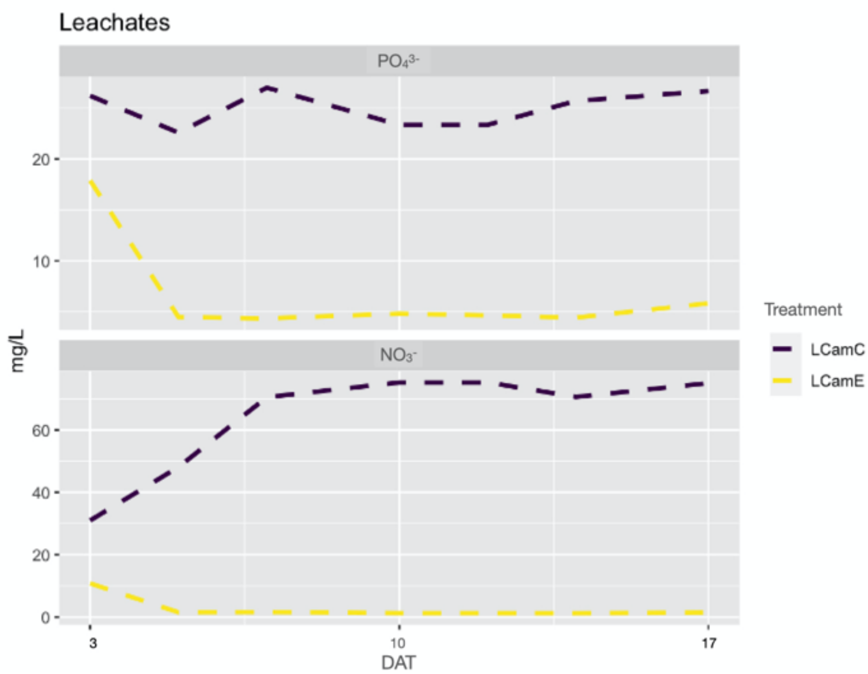
Annex X. 13 Relative humidity (%) and temperature (ºC) measurements inside the closed chamber during the determination experiment.

The determination experiment performed in the chamber showed very low concentrations of PO_4^{3-} and NO_3^-, with even less detection of NO_2^-, in the incoming irrigation for the struvite fertilized treatment without N no P and Mg (figure X.14). This confirms the treatment were irrigated correctly ensuring the treatment and nutrient availability for each treatment is correct.



Annex X. 14 Irrigation water analysis for PO_4^{3-}, NO_3^{-} for the determination experiment for the control treatment inside the chamber irrigated with a full nutrient solution (RegC), and the struvite fertilized (20g) and N, P, Mg free nutrient solution irrigated treatment (Reg E).

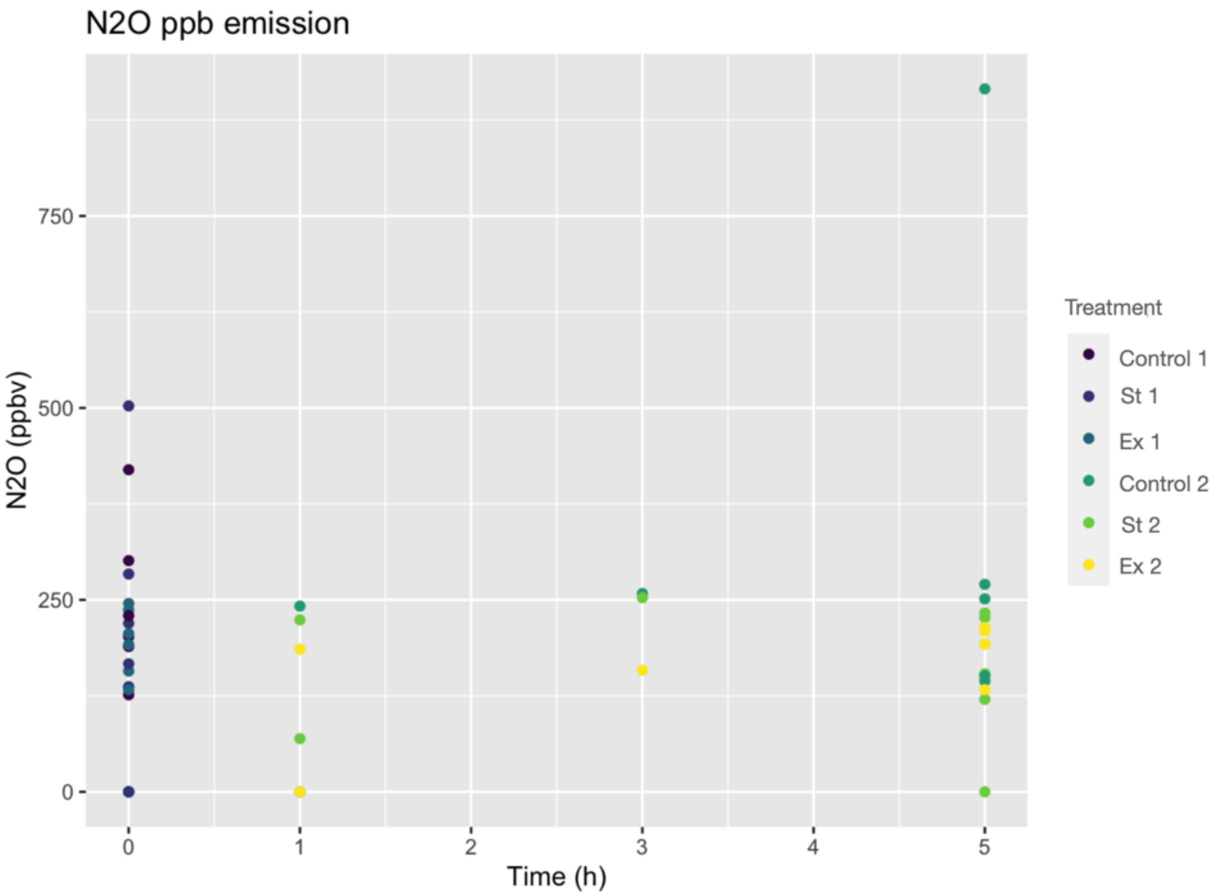
The leachate water was analyzed and displayed in figure X.15, showing a clear difference between treatments and their emissions of P and N compounds into water. This experiment has shown a very low dissolution of the struvite with concentrations close to 0 for NO_3^- as well as highly reduced in the case of PO_4^{3-} compared to the control treatment.



Annex X. 15 leachate analysis for PO_4^{3-}, NO_3^{-} for the determination experiment for the control treatment inside the chamber irrigated with a full nutrient solution (LCamC), and the struvite fertilized (20g) and N, P, Mg free nutrient solution irrigated treatment (LCam E).

When observing the analyzed N_2O emissions to air figure X.16 displays the amount of the GHG detected at the different times the chamber was left closed. While the initial samples taken from each side of the chamber once it was closed reveal amounts between 0 and 500ppbv with a greater cluster between 175 and 250ppbv.

While the samples taken at 1, 3 and 5h do not greatly differ from one another the samples taken from the building atrium outside the GH are slightly below the samples from the control and struvite treatments on both sides of the closed chamber.



Annex X. 16 N2O in ppbv detected over time (h) for treatments control, struvite fertilized crops (St) and exterior sampling site (Ex) before closing the chamber at time 0 (1) and after closing the chamber at times 1, 3 or 5 (2).

No great differences could be identified from the treatments control and struvite, while further analysis needs to be made. Previous work on the N_2O emission determination for struvite fertilization indicates that a greater emission was seen with the addition of the organic fertilizer vinasse, while other mineral recycled fertilizers showed no emission or rather low emission during the 80 day trial (Halbert-Howard et al. 2021). The same low emission was also identified for the control NPK fertilizer. This low emission of the mineral fertilizer struvite can be seen here with overall similar amounts reached after the closed chamber than in the initial sapling.

Overall, the methodology needs further revision, not only with the addition of a humidity filter but with the method for the gas chromatography. While the carrier gas N_2 has been seen to be a good option for greater precision in the N_2O detection other works recommend the use N_2 and of 5% of CH_4 (Rapson and Dacres 2014).

A4 Conclusions

The present work reflects the work done on the N_2O emission detection in lettuces fertilized with struvite as main source for N, P and Mg.

The validation experiments 1, 2 and 3 provide an understanding on the possible influence of the chamber in the lettuce growth and development as well as an understanding of the crop development and emissions to water with a full nutrient solution, ½ N irrigation and finally no N irrigation with N obtained from struvite.

The experiments show that while no effect on the plant growth was identified due to the closed chamber infrastructure the P content in the biomass was reduced compared to the crops outside. On the other hand, a yield increase could be seen in crops with struvite application and ½N nutrient solution in the validation experiments 1 and 2 with a slight increase of P emission to water for the double irrigation treatment.

For the validation experiment 3 a greater yield was obtained for the plants fertilized with struvite and a P, N and Mg deficient nutrient solution.

This last experiment made in the closed chamber let to the conclusion that 20g of struvite could sustain a lettuce production with yields similar or greater than the control.

Finally, N_2O emission analysis made in the determination experiment within the chamber showed similar amounts to the initial sample before closing the chamber. This result gives an initial idea that no great emissions occur during the closing periods although the methodology has room for improvement with the addition of a sample pre-treatment due to the great humidity in the chamber, and the changing of the gas carrier in the GC method.

Annex II

Supplementary Information

List of Tables & Figures Annex II

Annex II

*Annex A3. 1 Potential tomato production in the selected UA areas, and self-sufficiency in the metropolitan area of Barcelona (AMB) *value obtained from (Sanyé-Mengual et al. 2018)*

Crop yield	kg/m^2	16,5*
	kg/ha	165000
UA Area	ha	45,21
Total crop yield	kg	7459650
	Ton	7459
Tomato consumption	kg/person/year	17,2*
Population AMB	Nº	3239337
Consumption	kg/year	55716596,4
	Ton/year	55716
Self -supply	%	13,3

*Annex A3. 2 Nutrient content in urban waste. OM= Organic material, DW= dry weight, Hum%= humidity content, * value obtained from range 20,9-31 ** value obtained from range 6,1-12.*

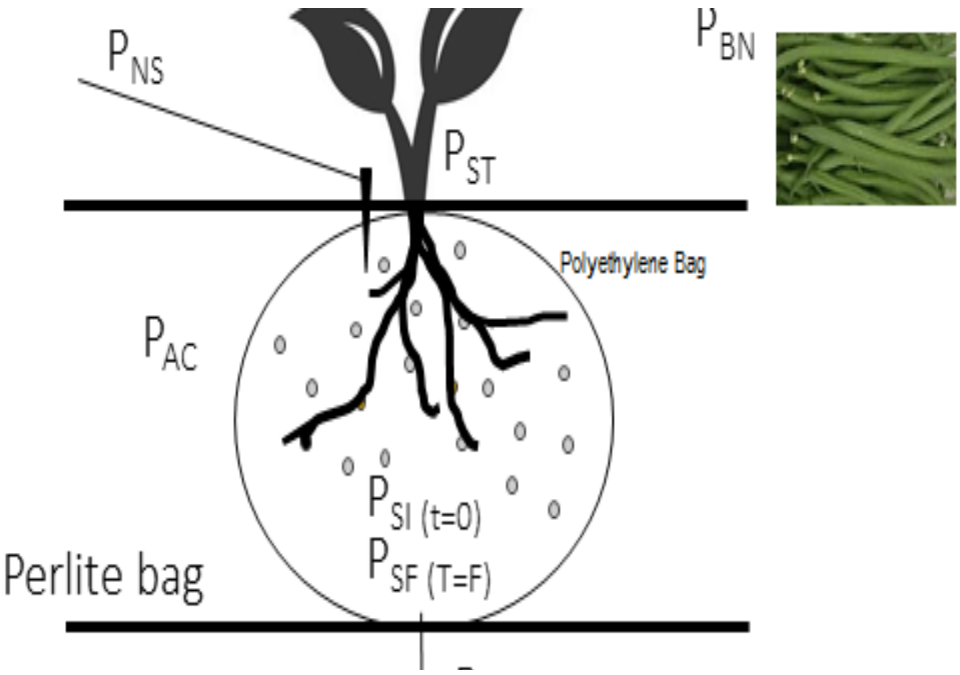
Waste Type	N g/kg (DW)	P g/kg (DW)	Hum %	Source
OM Compost	21,5	7,9	30%	(Gímenez Lorang, Soliva I Torrentó, and Huerta 2005)
OM Digest	27,3*	8,9**	30%	(Carabassa i Closa 2020)
Sludge EDAR	23,3	19,6	26,8%	(Gímenez Lorang, Soliva I Torrentó, and Huerta 2005)
Struvite	57	126		(N. Ahmed et al. 2018)

*Annex A3. 3 Tomato crop N and P demand and upscale for the selected UA area *values obtained from (Marinelli et al. 2021)*

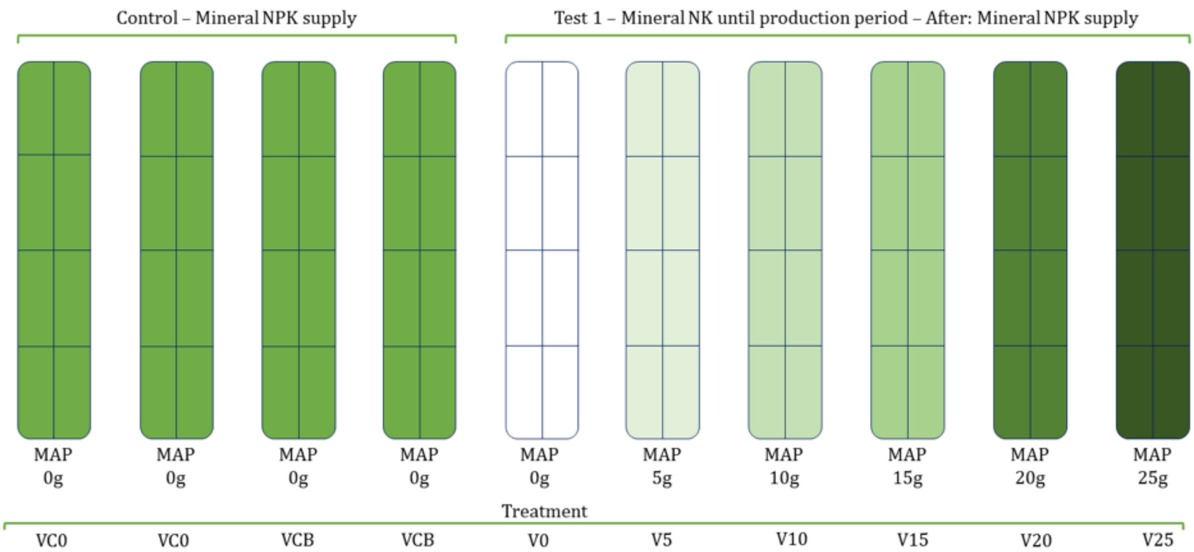
Tomato Crop		
N- demand	kg/ha	250*
P- demand	kg/ha	65*
UA Area		
Total area	ha	45,21
N- demand	kg	11303
P- demand	kg	2939

Annex A3. 4 P and N recovery potential through struvite precipitation in all WWTP of the AMB with data form 2020. Recovery potentials based on Rufí-Salís, Brunnhofer, et al., 2020, with minimum removal 7% P with AirPrex technology and maximum removal 60% with RemNut Technology.

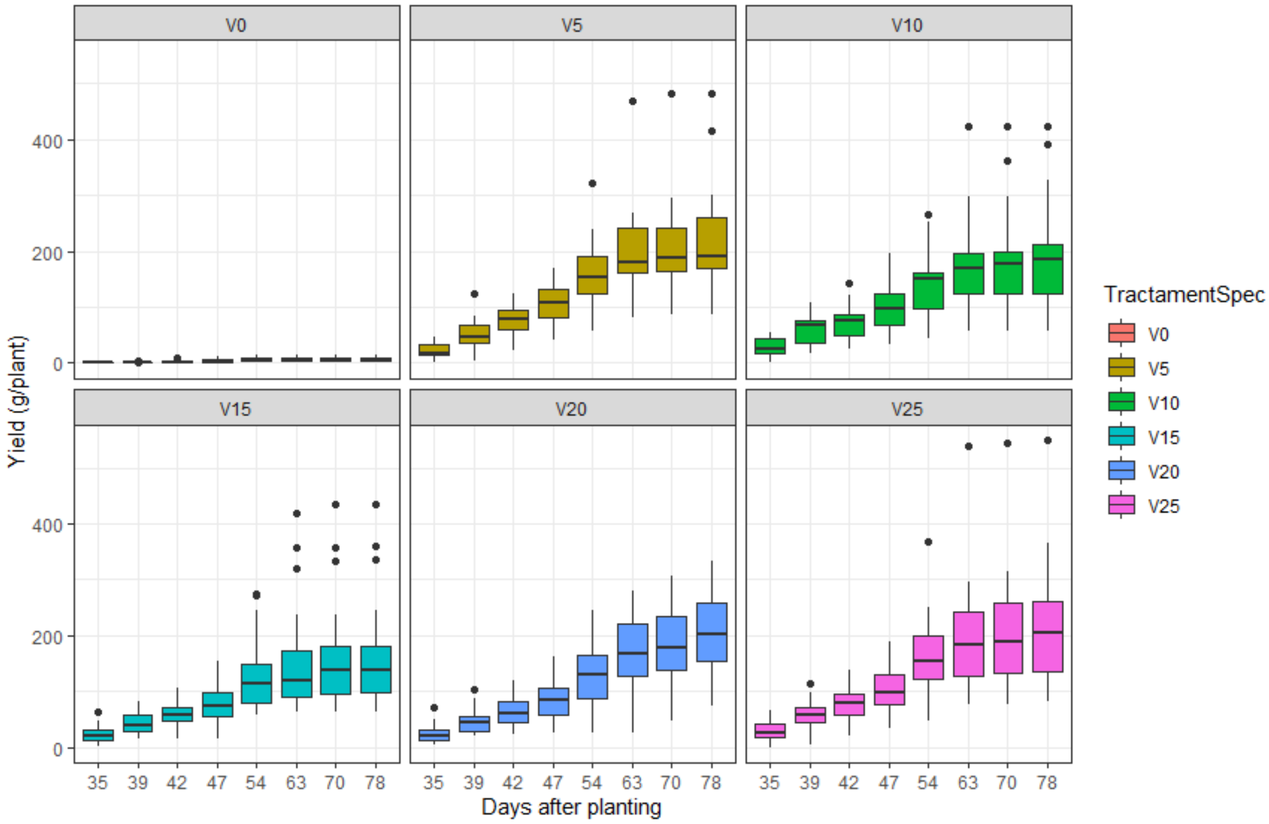
WWTP	Struvite recovery (Ton)			
	P (7%)	P (60%)	N (7%)	N (60%)
Besos	57,3151053	491,272331	25,9282619	222,242245
El Prat de Llobregat	43,8325366	375,707457	19,8290047	169,962897
St. Feliu de Llobregat	8,83685956	75,7445105	3,99762694	34,2653738
Montcada I Reixac	8,95509224	76,7579335	4,05111316	34,7238271
Gava I Viladecans	7,07318102	60,6272659	3,19977237	27,4266203
Begues	0,16604118	1,42321008	0,07511387	0,64383313
Vallvidrera	0,12085116	1,03586712	0,05467076	0,46860655
Sum	126,299667	1082,56857	57,1355637	489,733403



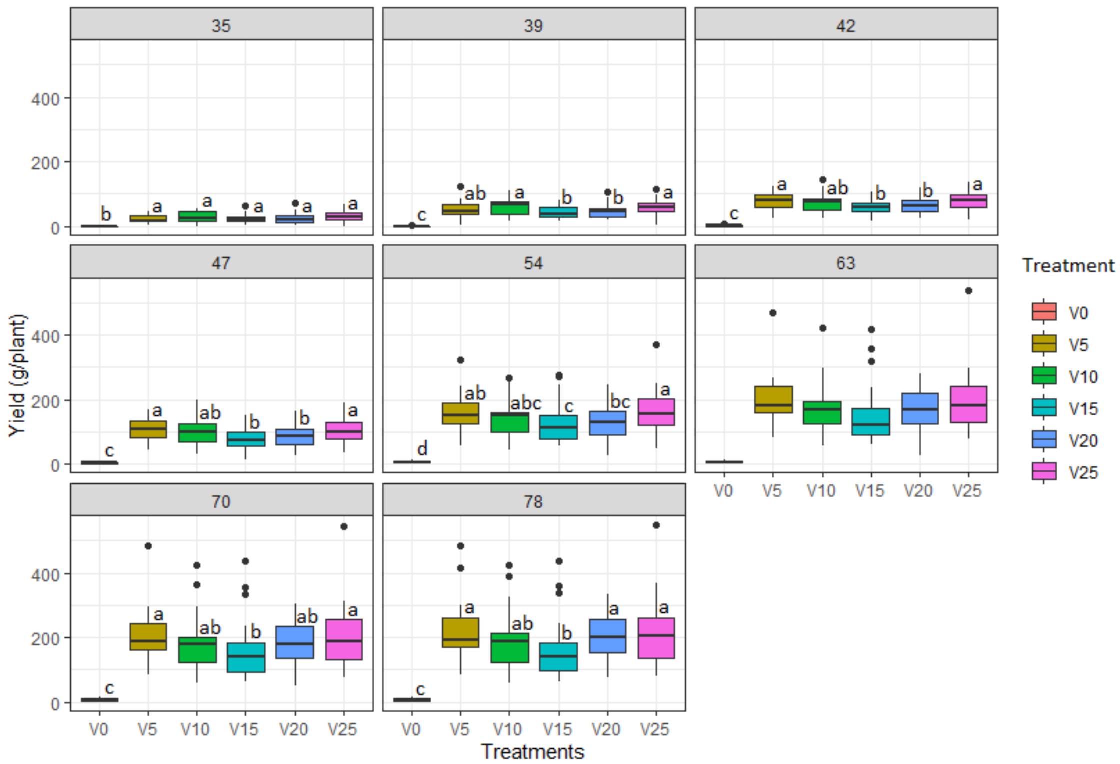
Annex A4. 1 Diagram of the perforated bag. PNS = mineral P in the nutrient solution; PSI = P in initial struvite; PLIX = P in leachates; PLV, PST, and PBN,= P in leaves, stem and beans, respectively; PSF = undissolved P (final struvite); PAC = dissolved P accumulated in the water retained in the substrate at the end of the crop.



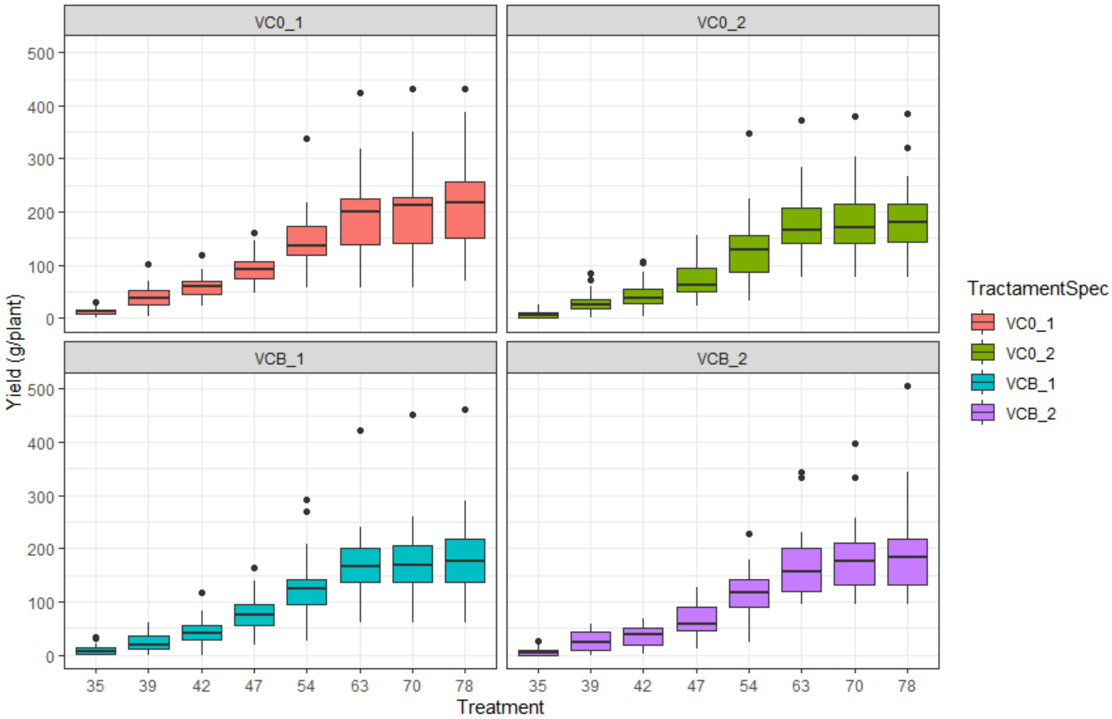
Annex A4. 2 Distribution of growing lines in the validation test. Each growing table consisting on two irrigation lines, with four perlite bags each (eight perlite bags per growing table). From left to right: first four growing tables supplied with mineral P fertilizer as control treatment, VC0= without the perforated bag, VCB= with the perforated bag. Lines five to ten irrigated without mineral P and with the addition of 0, 5, 10, 15, 20 and 25g of struvite.



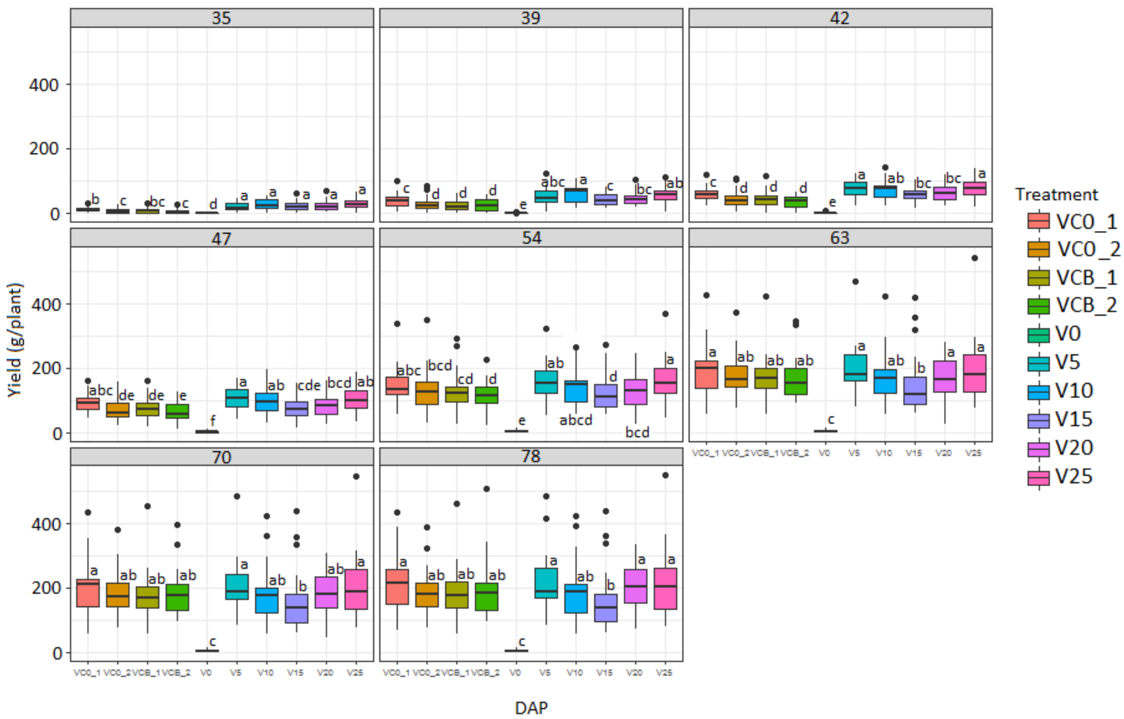
Annex A4. 3 Production (g/plant) of the struvite treatments in the validation test, with different amounts of struvite ranging from 0g to 25g (V0, V5, V10, V15, V20, V25) for each harvest. Sample size for harvests 1, 2, 3, 4 and 5 (35- 54 DAT) corresponds to n=28 plants, for harvests 6, 7 and 8 (63-78 DAT) n= 24 plants per treatment.



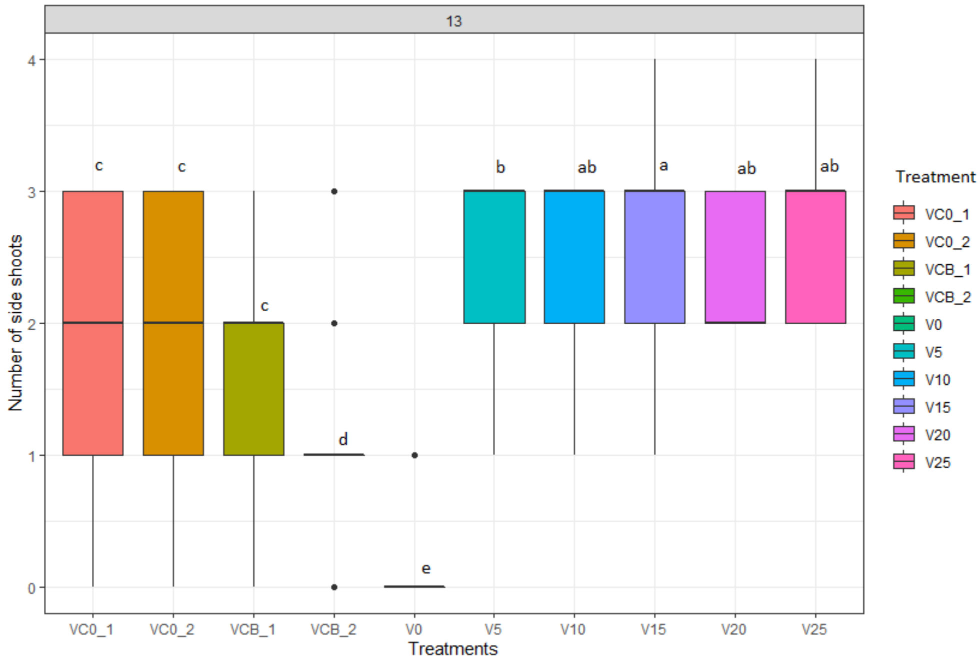
Annex A4. 4 Production (g/plant) of the struvite treatments in the validation test, with different amounts of struvite ranging from 0g to 25g (V0, V5, V10, V15, V20, V25) for each harvest. Same letters (a,b, c) indicate no significant difference (p>0.05) between treatment for each harvest time. Sample size for harvests 1, 2, 3, 4 and 5 (35- 54 DAT) corresponds to n=28 plants, for harvests 6, 7 and 8 (63-78 DAT) n= 24 plants per treatment.



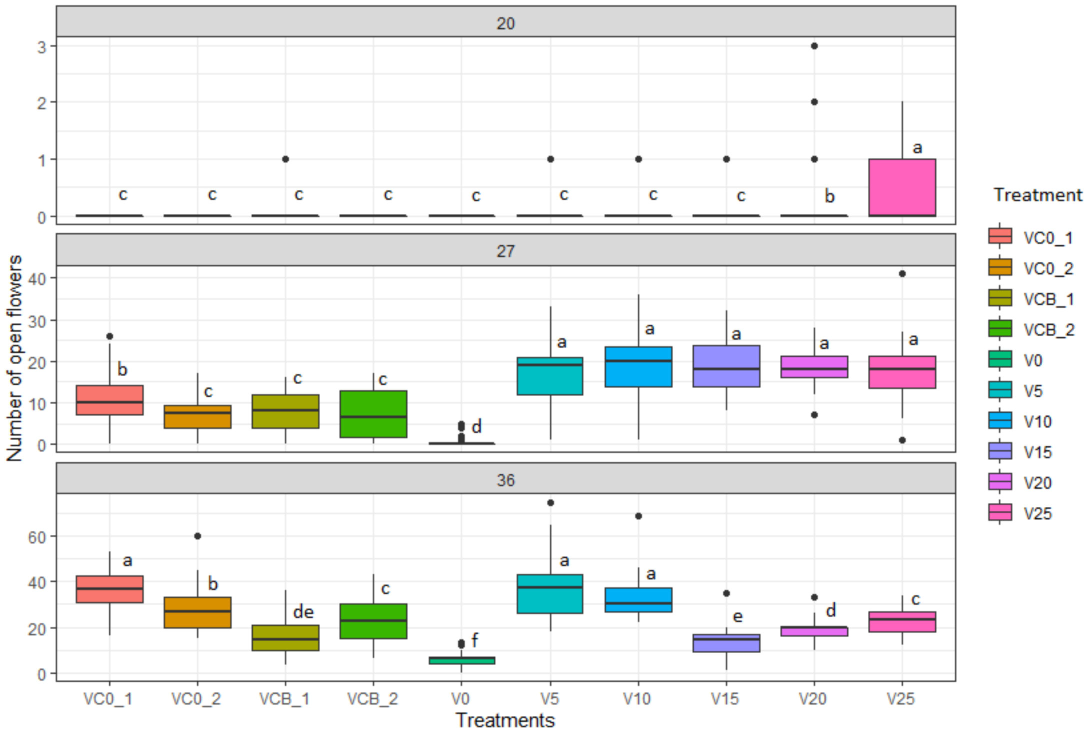
Annex A4. 5 Production (g/plant) of the control treatments in the validation test, with (VCB) and without (VC0) perforated bags for each harvest. Sample size for harvests 1, 2, 3, 4 and 5 (35- 54 DAP) corresponds to n=28 plants, for harvests 6, 7 and 8 (63-78 DAP) corresponds to n=28 plants, for harvests 6, 7 and 8 (63-78 DAP) n=24 plants per treatment.



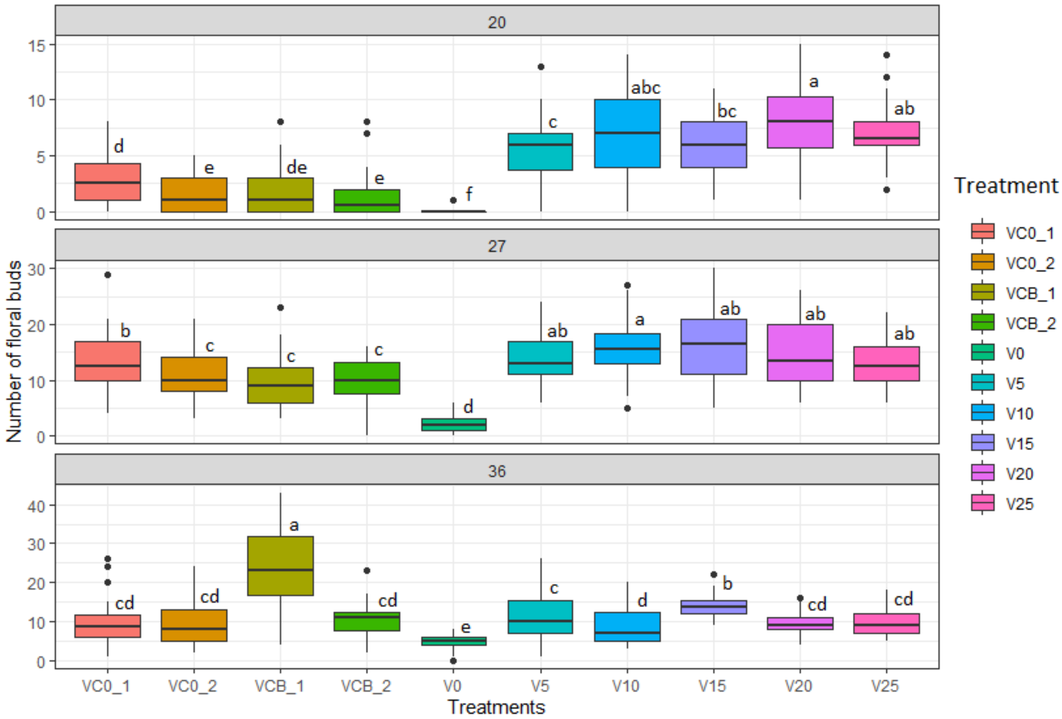
Annex A4. 6 Production (g/plant) in the validation test of the control treatments, with (VCB) and without (VC0) perforated bags and the struvite treatments, with different amounts of struvite ranging from 0g to 25g (V0, V5, V10, V15, V20, V25) for each harvest. Same letters (a, b, c, d, e, f) indicate no significant difference (p>0.05) between treatment for each harvest time. Sample size for harvests 1, 2, 3, 4 and 5 (35- 54 DAT) corresponds to n=28 plants, for harvests 6, 7 and 8 (63-78 DAT) n=24 plants per treatment.



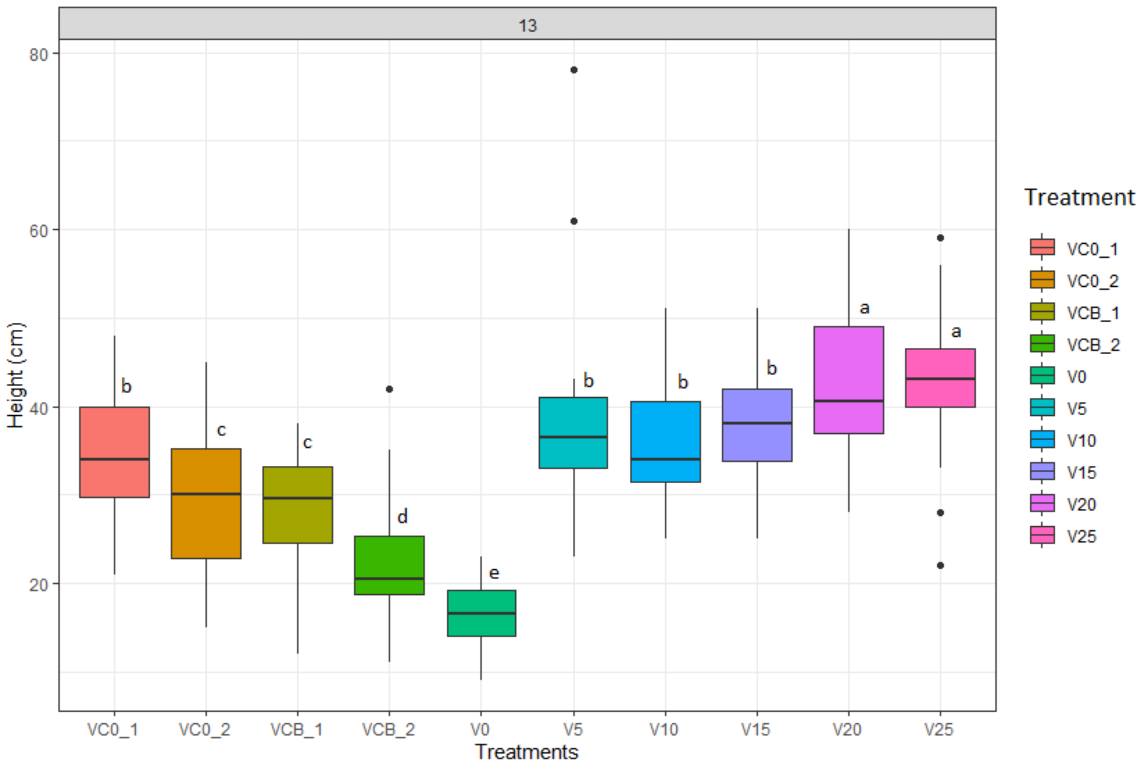
Annex A4. 7 Number of side shoots per plant per treatment and Days after Transplanting (DAP) in the validation test (13 DAP). Same letters (a, b, c, d, e) indicate no significant difference (p>0.05) between treatment for each counting time. Sample size n=32 for each treatment.



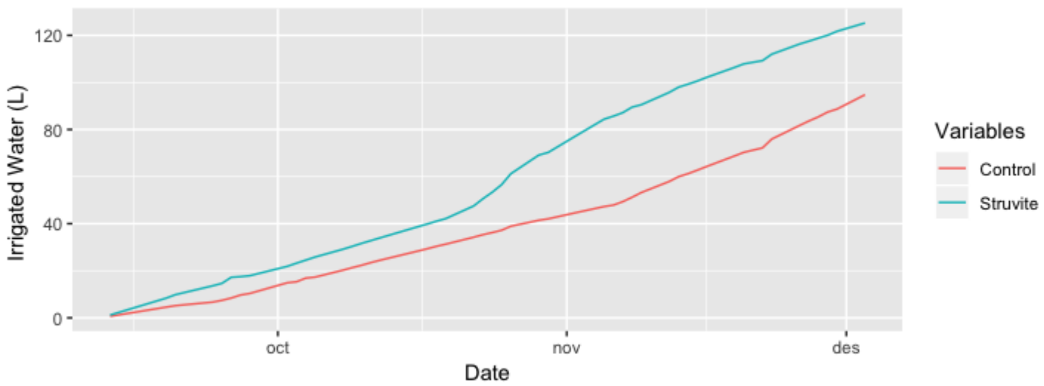
Annex A4. 8 Number of open flowers per plant per treatment and Days after Transplanting (DAP) in the validation test (20, 27, 36 DAP). Same letters (a, b, c, d, e, f) indicate no significant difference (p>0.05) between treatment for each counting time. Sample size n=32 for each treatment.



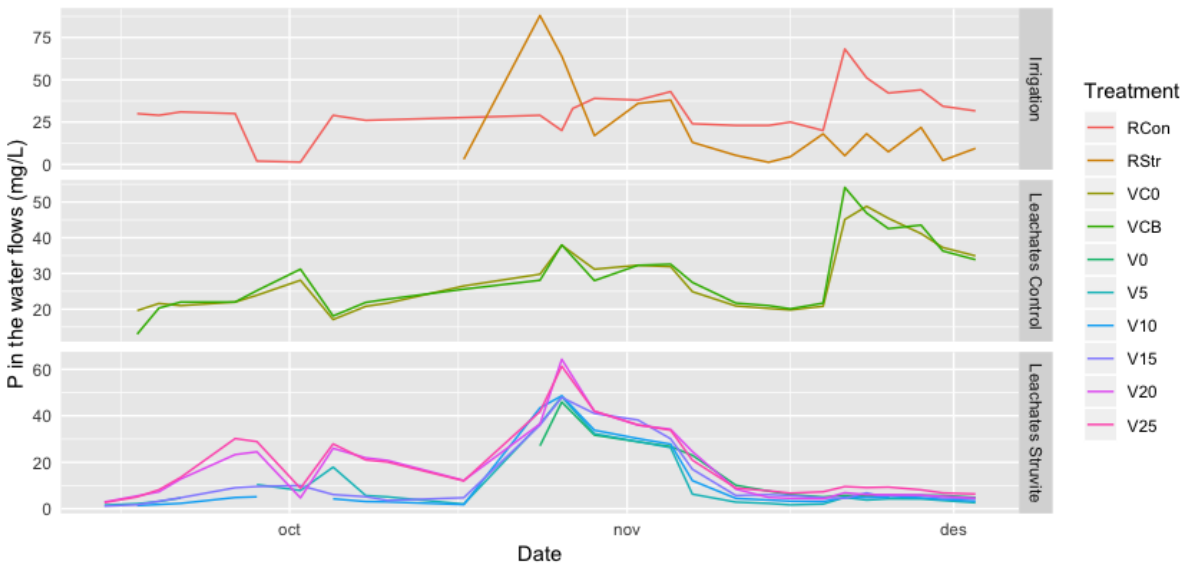
Annex A4. 9 Number of floral buttons per plant per treatment and Days after Transplanting (DAP) in the validation test (20, 27, 36 DAP). Same letters (a, b, c, d, e, f) indicate no significant difference (p>0.05) between treatment for each counting time. Sample size n=32 for each treatment.



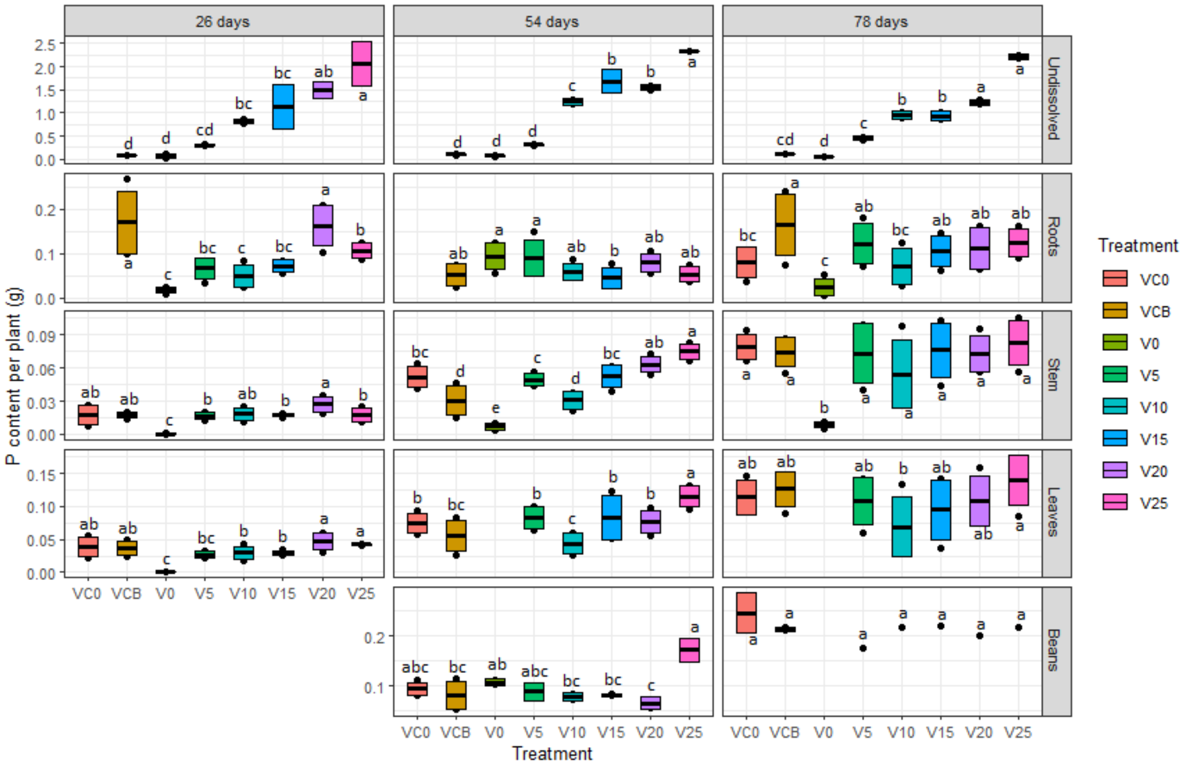
Annex A4. 10 Height (cm) plant per treatment and Days after Transplanting (DAP) in the validation test (13 DAP). Same letters (a, b, c, d, e) indicate no significant difference (p>0.05) between treatment for each counting time. Sample size n=32 for each treatment.



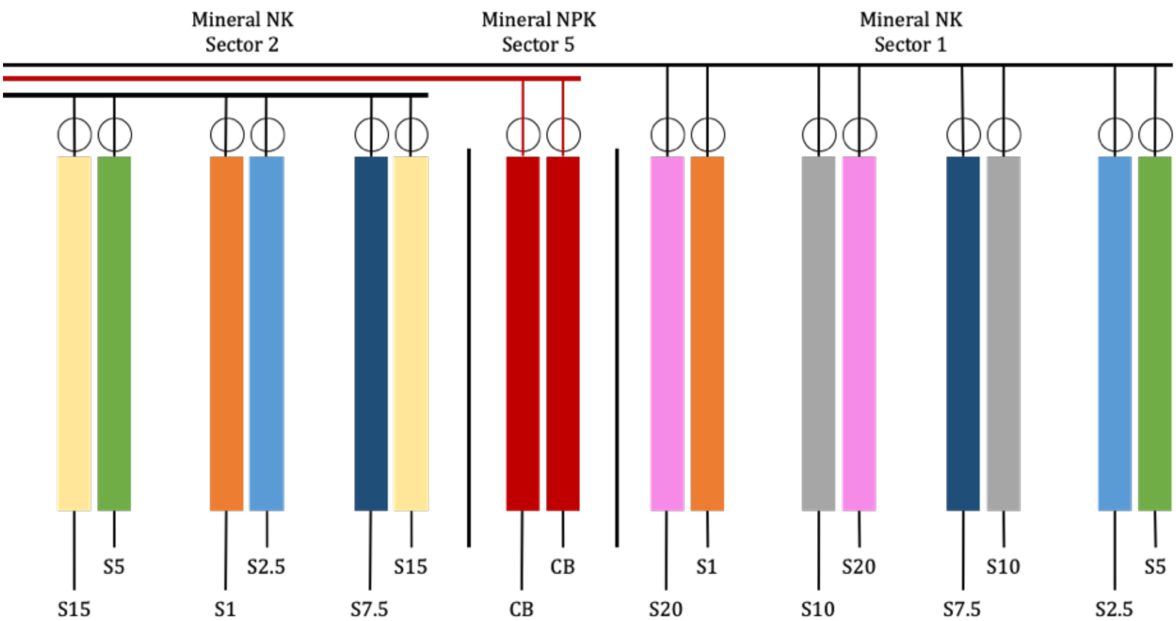
Annex A4. 11 Water irrigated (L) per plant in the validation test.



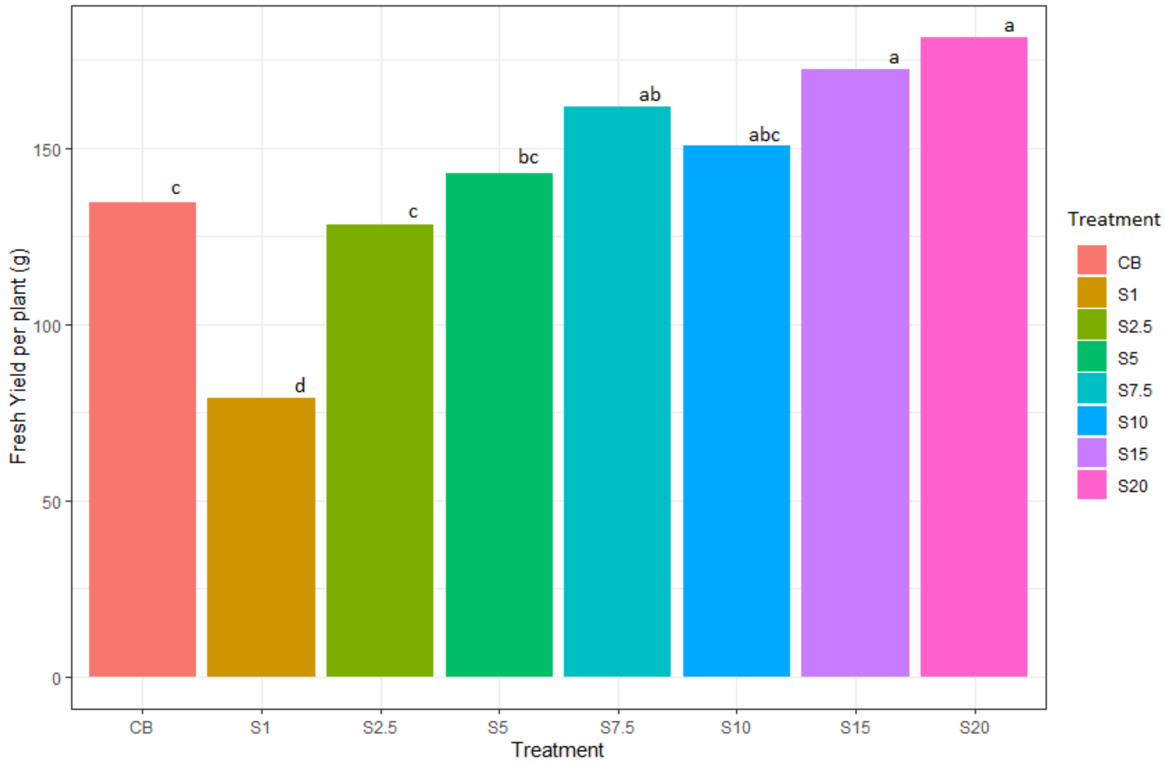
Annex A4. 12 Phosphorus concentrations in the multiple water streams in the validation test.



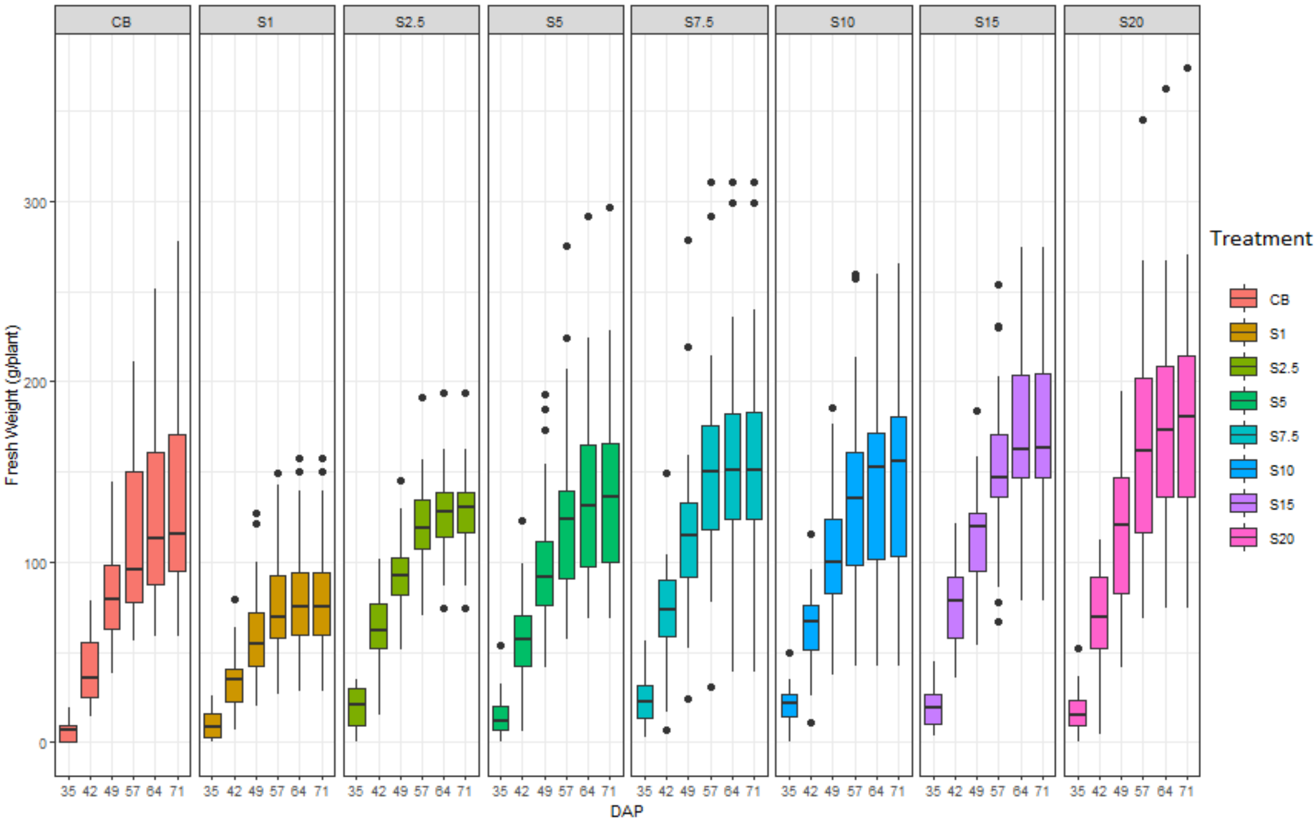
Annex A4. 13 content per plant (g) of the struvite treatment in the validation test. Same letters (a, b, c, d, e) indicate no significant difference (p>0.05) between treatment for each harvest time. Sample size for all organs n=4. Undissolved P content n=2



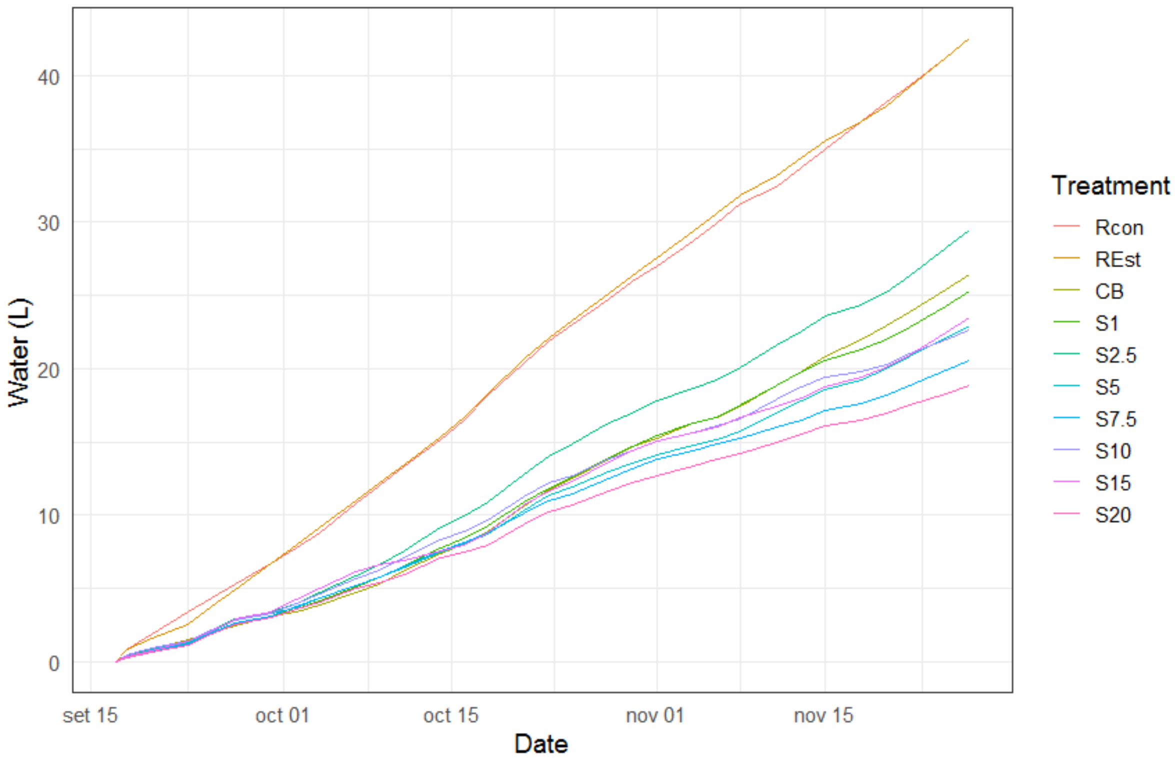
Annex A4. 14 Distribution of growing lines in the validation test



Annex A4. 15 Mean aggregated production per plant per treatment in the determination test



Annex A4. 16 Distribution of accumulated production per plant per harvest (DAP = days after plantin) of different treatments. Sample size for all harvests is n=24 plants per treatment.



Annex A4. 17 Water irrigated and drained per plant in the different treatments in the determination test

Annex A4. 18 Phosphorus concentrations in the multiple water streams in the determination test

Annex A5. 1 Applied nutrient solutions (NS) for the control treatment and the Mg, P, N-free nutrient solution for treatments inoculated treatments additionally fertilized with struvite.

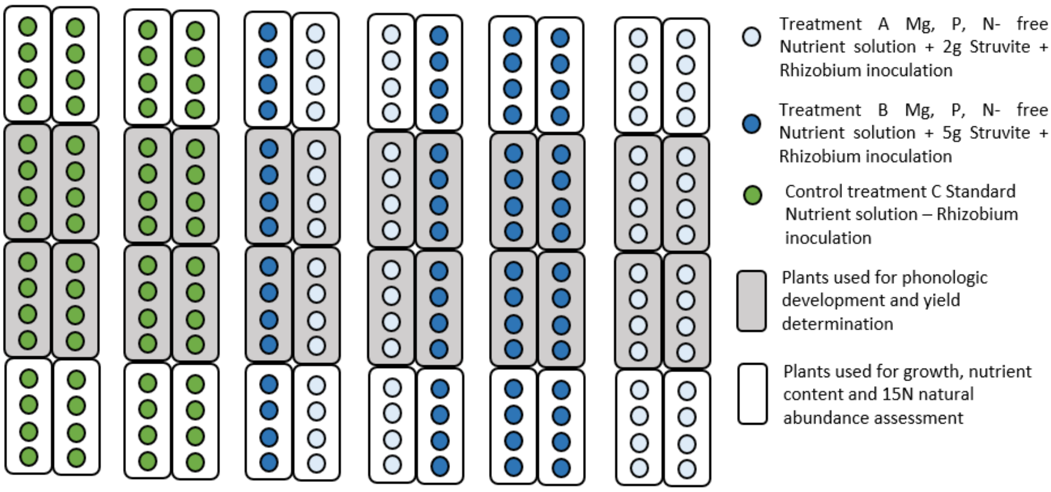
Nutrients applied	*Control NS*	*Mg, P, N-free NS*
KPO_4H_2	136 mg/L	---
KNO_3	101 mg/L	---
K_2SO_4	217 mg/L	435 mg/L
$Ca(NO_3)_2$	164 mg/	---
$CaCl_2$	111 mg/L	111 mg/L
$Mg(NO_3)_2$	148.3 mg/L	---
Hortilon	0.1 mg/L	0.1 mg/L
Sequestrene	0.1 mg/L	0.1 mg/L

Annex A5. 2 Climatic conditions inside the RTG Lab.

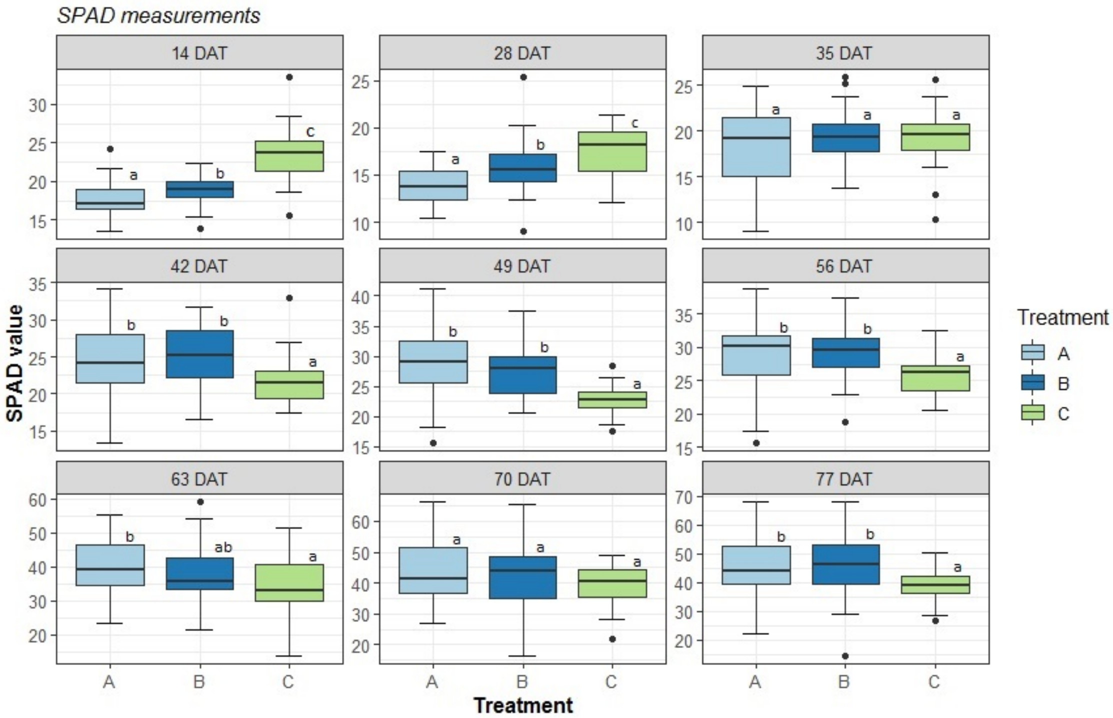
TEMPERATURE	
AVERAGE T ºC	*18.9*
MINIMUM T ºC	*4.5*
MAXIMUM T ºC	*29.9*
STANDARD DEVIATION	*2.1*
REALATIVE HUMIDITY	
AVERAGE (RH)	*38.1*
MINIMUM (RH)	*5.7*
MAXIMUM (RH)	*77.4*
STANDARD DEVIATION	*5.8*

Annex A5. 3 Leachate NO3- content (mg/L) from results given for three treatments A) 2g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution B) 5g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution and C) standard nutrient solution and C) standard nutrient solution - Rhizobium inoculation at five time periods from the 14 DAT until 77 DAT.

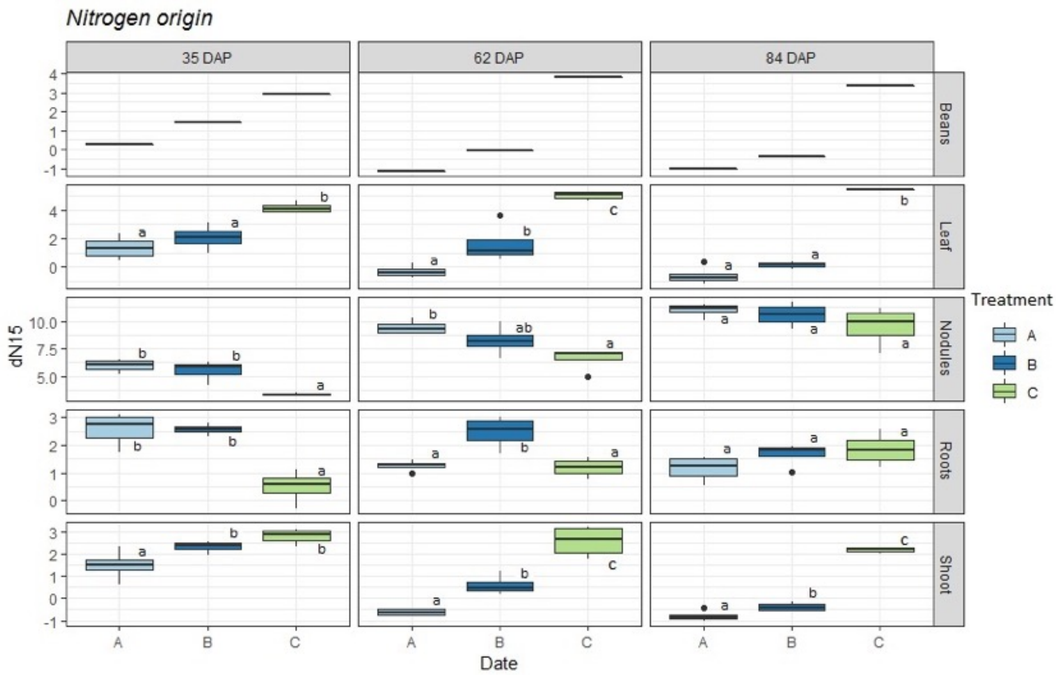
DATE	*A*	*B*	*C*
14 DAT	*7.71*	*10.57*	*8.54*
35 DAT	*3.41*	*4.89*	*32.89*
49 DAT	*0.92*	*0.91*	*47.87*
63 DAT	*0.03*	*0.36*	*51,97*
77 DAT	*0.32*	*N.A.*	*55.93*



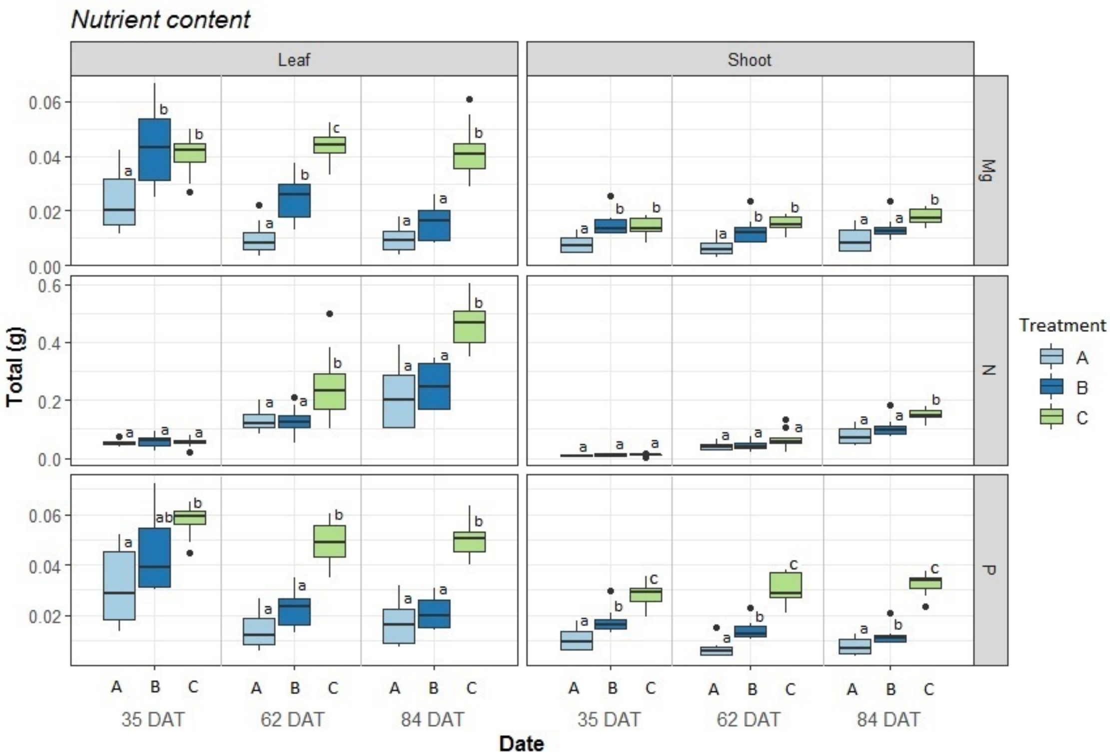
Annex A5. 4 Image of the Experimental layout in the RTG Lab.



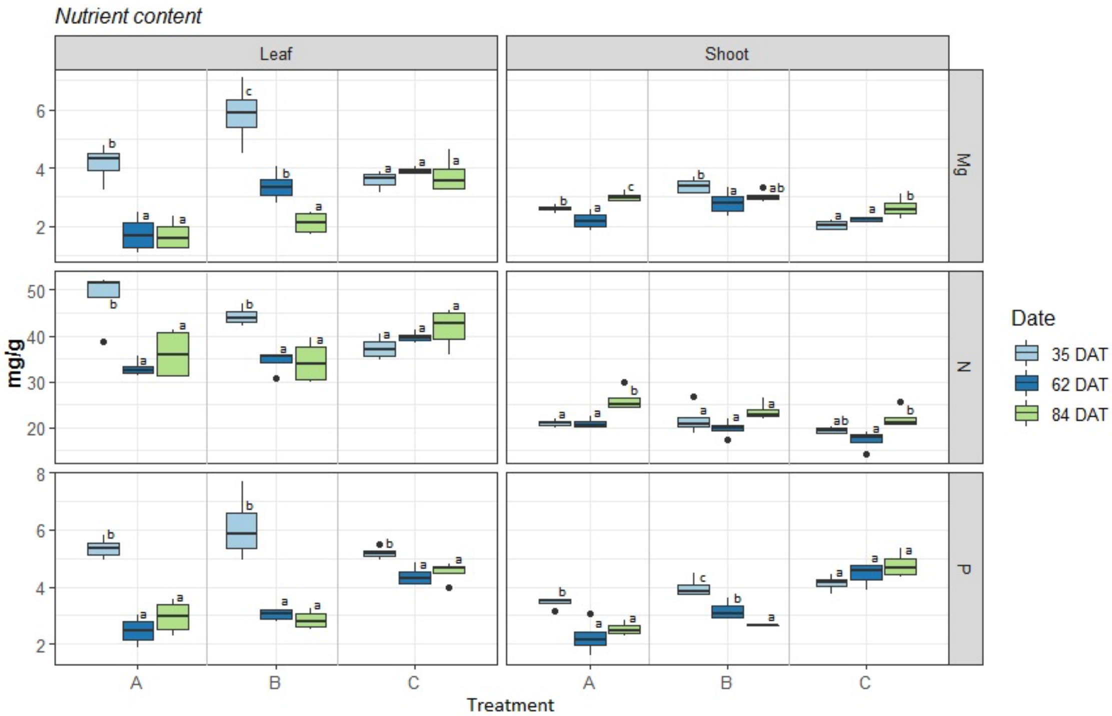
Annex A5. 5 Chlorophyll content measurements (SPAD) in Phaseolus vulgaris leaves. Boxplot (n=32) results given for three treatments A) 2g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution B) 5g of struvite + Rhizobium inoculation + P, Mg, N- free nutrient solution and C) standard nutrient solution - Rhizobium inoculation measured at 9 time periods throughout the crop cycle. Significant differences (p< 0.05) between treatments marked with different letter (a,b,c).



Annex A5. 6 Nutrient concentration in Phaseolus vulgaris leaves and shoots, expressed in mg/g. Boxplot (n=4) results given for three treatments A) 2g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution B) 5g of struvite + Rhizobium inoculation + P Mg, N-free nutrient solution and C) standard nutrient solution - Rhizobium inoculation at three different time periods. 35 days after transplanting, *62 days after* transplanting *and 84 days after* transplanting*.* Significant differences (p< 0.05) between treatments marked with different letter (a,b,c).



Annex A5. 7 Nutrient content in Phaseolus vulgaris leaves and shoots, expressed in g. Boxplot (n=4) results given for three treatments A) 2g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution B) 5g of struvite + Rhizobium inoculation + P, Mg, N- free nutrient solution and C) standard nutrient solution - Rhizobium inoculation at three different time periods. 35 days after transplanting*, 62 days after* transplanting *and 84 days after* transplanting*.* Significant differences (p< 0.05) between treatments marked with different letter (a,b,c).



Annex A5. 8 Nutrient content in Phaseolus vulgaris leaves and shoots, expressed in mg/g. Boxplot (n=4) results given for three treatments A) 2g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution B) 5g of struvite + Rhizobium inoculation + P, Mg, N-free nutrient solution and C) standard nutrient solution - Rhizobium inoculation at three different time periods. 35 days after transplanting, *62 days after* transplanting *and 84 days after* transplanting*.* Significant differences (p< 0.05) between dates marked with different letter (a,b,c).

Annex A6. 1 Hydroponic installation in the ICTA-UAB rooftop greenhouse.

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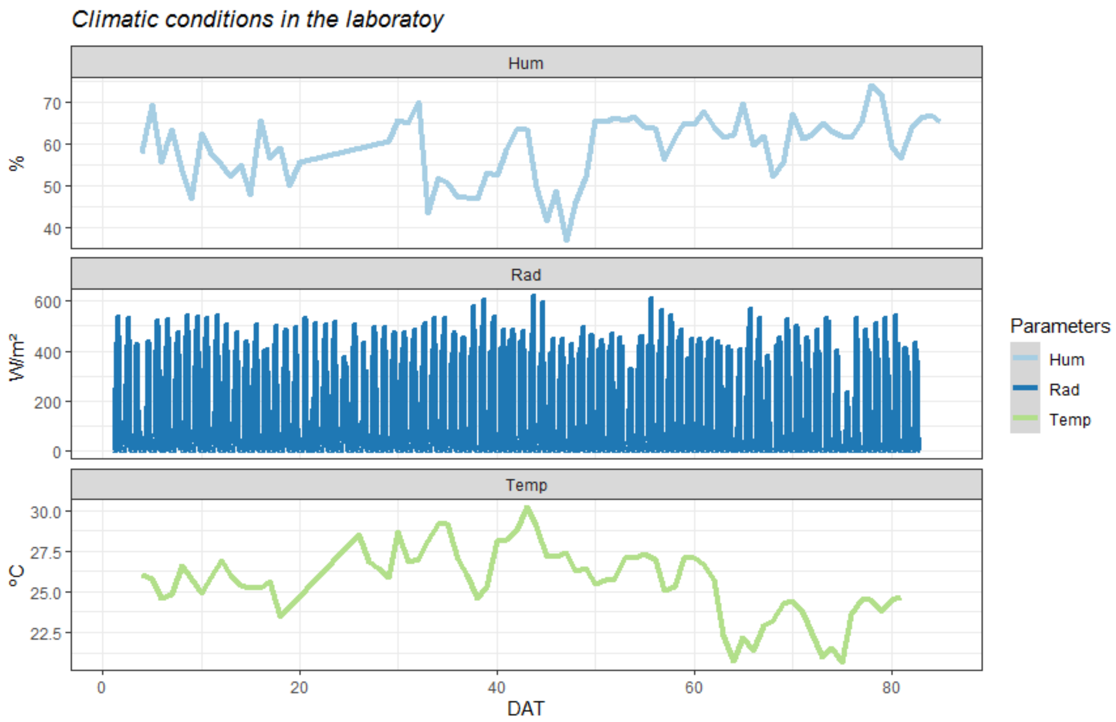
Annex A6. 2 Nutrients applied to struvite fertilized treatments and control treatment. P, N and Mg content in struvite treatments.

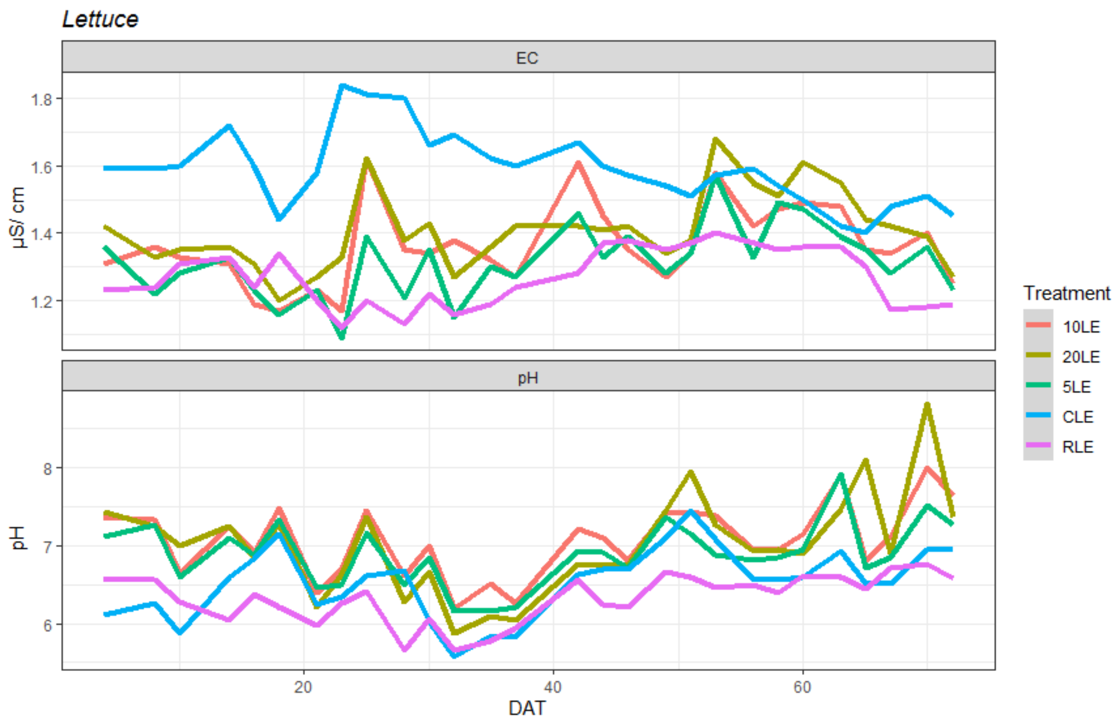
		g/L	P	N	Mg
CONTROL NUTRIENT SOLUTION	KPO_4H_2	13.625			
	KNO3	10.125			
	K2SO4	43.5			
	Ca(NO3)2	16.375			
	CaCl2*2H2O	14.75			
	Mg(NO3)2	22.25			
	Hortrilon	1			
	Sequestrene	1			
PHOSPHOROUS FREE NUTRIENT SOLUTION	KNO3	10.125			
	K2SO4	43.5			
	Ca(NO3)2	16.375			
	CaCl2*2H2O	14.75			
	Mg(NO3)2	22.25			
	Hortrilon	1			
	Sequestrene	1			
STRUVITE	5g		0.625g	0.285g	0.495g
	10g		1.25g	0.57g	0.99g
	20g		2.5g	1.14g	1.98g

Phython script:

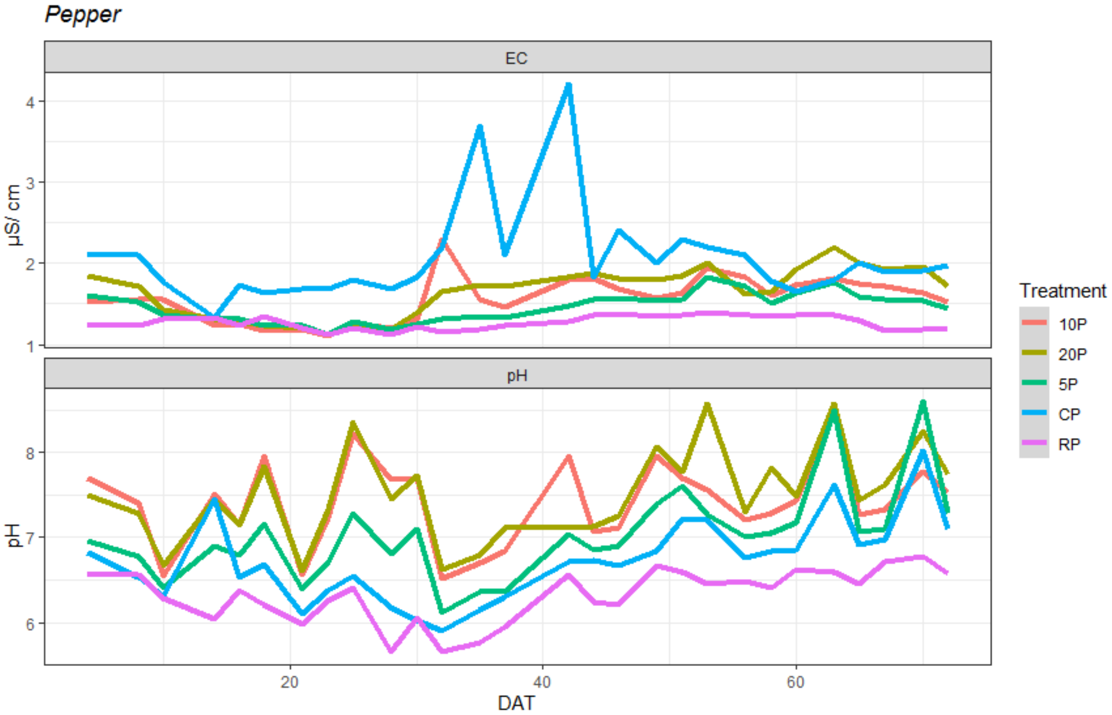
```
import cv2
from os import listdir
import numpy as np
from os.path import isfile, join

if __name__ == '__main__':
    carpetaImatges = 'PATH' + '/'
    fitxersImatges = listdir(carpetaImatges)
    for iImatge in range(0, len(fitxersImatges)):
        if fitxersImatges[iImatge][-4:] == '.jpg' or fitxersImatges[iImatge][-4:] == '.png':
            imatge = cv2.imread(carpetaImatges+fitxersImatges[iImatge])
            maskNegre = 255*np.ones((imatge.shape[0],imatge.shape[1]))
            maskNegre[405:730,1014:1549] = 0
            imatge_G = imatge[:,:,1]
            imatge_G = imatge_G < 200

            imatge_G = imatge_G*maskNegre
            cv2.imwrite(carpetaImatges + fitxersImatges[iImatge][:-4] + 'Bin' + '.jpg',imatge_G)
            print('Image: ' + fitxersImatges[iImatge])
            print('Leaves Area: ' + str(np.sum(imatge_G>0)) + ' (' + str(np.sum(imatge_G>0)/(imatge.shape[0]*imatge.shape[1])) + '% of the image)')
```



Annex A6. 3 Climatic conditions recorded in the greenhouse laboratory DAT= days after transplanting, Hum= relative humidity (%), Rad = incoming radiation recorded inside the building (W/m2), Temp= Recorded temperature inside the greenhouse (ºC).



Annex A6. 4 Irrigation and leachate parameters recorded for lettuce production. DAT= days after transplanting, EC= electric conductivity (µS/cm), pH= pH value. Treatments 5LE= leachate from crops fertilized with 5g of struvite, 10LE= leachate from crops fertilized with 10g of struvite, leachate from crops fertilized with 20LE= 20g of struvite, CLE= leachate from control treatment, RLE= irrigated nutrient solution).



Annex A6. 6 Irrigation and leachate parameters recorded for pepper production. DAT= days after transplanting, EC= electric conductivity (µS/cm), pH= pH value. Treatments 5P= leachate from crops fertilized with 5g of struvite, 10P= leachate from crops fertilized with 10g of struvite, leachate from crops fertilized with 20P= 20g of struvite, CP= leachate from control treatment, RP= irrigated nutrient solution).

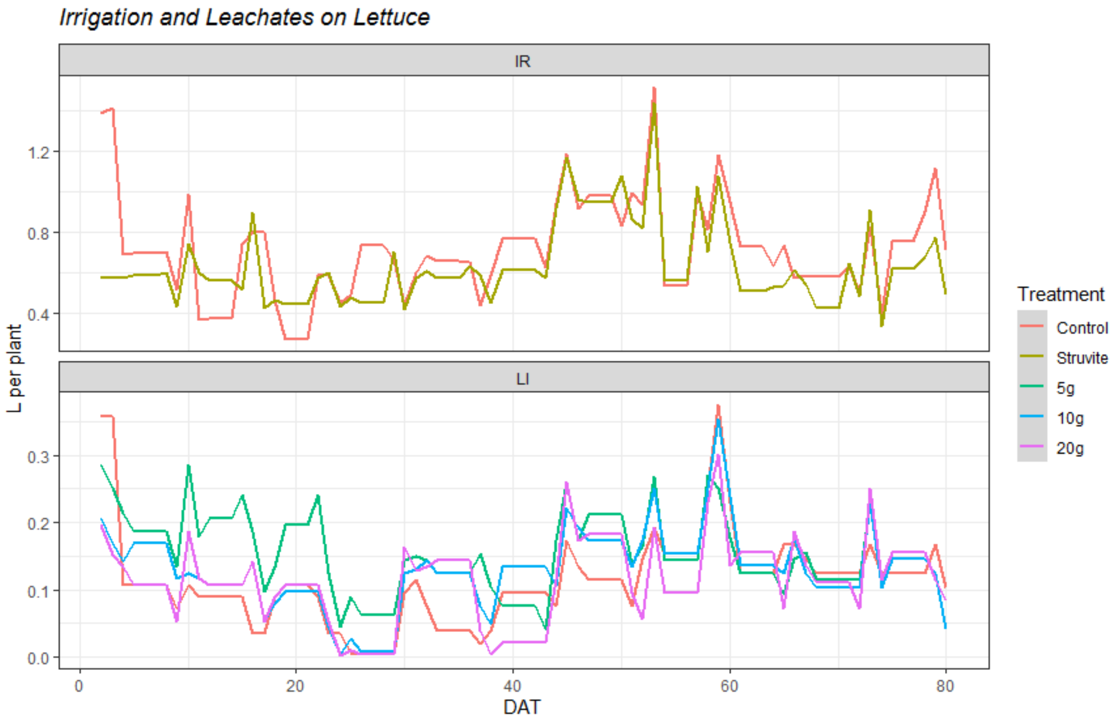
Annex A6. 5 Lettuce yield recorded from labeled pots (A to O) for all harvests (Sampling 1, 2 and 3), for treatment 5LE (5g), 10LE (10g) and 20LE (20g). Yield differences between samplings 1 (S1) and sampling 2 (S2) as well as sampling 2 and sampling 3 (S3) within the same growing pot given in %.

	5g				10g				20g		
	1st Sampling	2nd Sampling	3rd Sampling		1st Sampling	2nd Sampling	3rd Sampling		1st Sampling	2nd Sampling	3rd Sampling
A	259.7	228.9	130	A	271	305.1	165	A	254	237.1	210
B	221.8	270.9	140	B	265.2	222	155	B	207.1	243.9	230
C	295.2	290.5	155	C	282.7	280.5	180	C	315.3	309.4	160
D	236.8	120	180	D	257.8	253.7	115	D	280.6	269.6	40
E	261.6	223.2	130	E	268.4	243.3	130	E	295.3	224.9	205
F	228.5	211.3	155	F	298	234.3	160	F	317.2	294.9	175
G	226.9	245.4	100	G	286.8	287.2	135	G	307.5	312.9	225
H	279.4	198.3	110	H	256.1	295.1	165	H	260.1	234.8	145
I	222.6	226	120	I	283.1	246.2	145	I	255.3	270.1	130
J	267.3	232.8	95	J	302.6	293.3	150	J	342.8	241.2	150
K	235.3	180.8	135	K	307.4	324.5	170	K	313.4	317.9	200
L	232.8	181.8	115	L	269.1	261.9	120	L	253.4	292.4	170
M	313.2	203.5	155	M	263.4	300.6	125	M	267.6	303.6	160
N	106.7	262.7	115	N	274.5	240.6	180	N	263.4	299.4	155
O	183.1	227.6	140	O	226.6	225.6	135	O	283.3	250.6	190

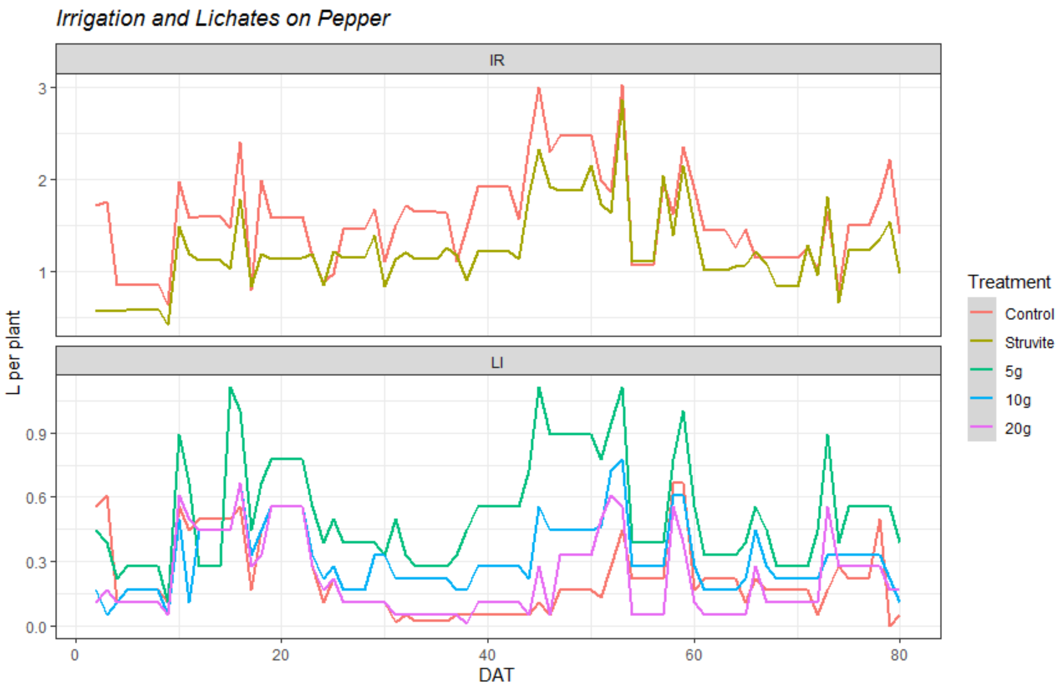
YIELD DIF	5g	10g	20g
DIF S1-S2	-11%	-2%	-2%
DIF S2-S3	-36%	-44%	-34%

Annex A6. 7 Lettuce average production for all harvests (given in g) for treatments 5LE, 10LE, 20LE and CLE. Difference to control calculated for each harvest (given in %).

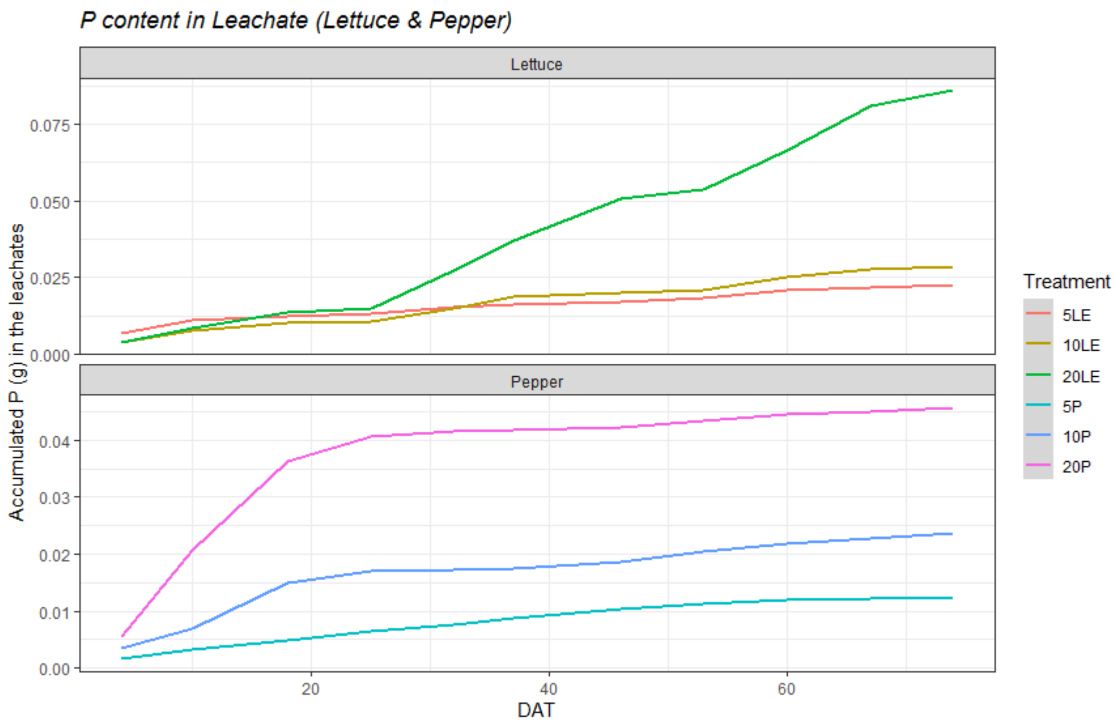
	5LE	10LE	20LE	CONTROL
1ST HARVEST	225.5	249.8	272.6	250
DIF TO CONTROL	-11%	0%	8%	
2ND HARVEST	224.8	251.6	261.3	279
DIF TO CONTROL	-24%	-11%	-7%	
3RD HARVEST	135.5	143.9	164.7	140.2
DIF TO CONTROL	-3%	3%	15%	



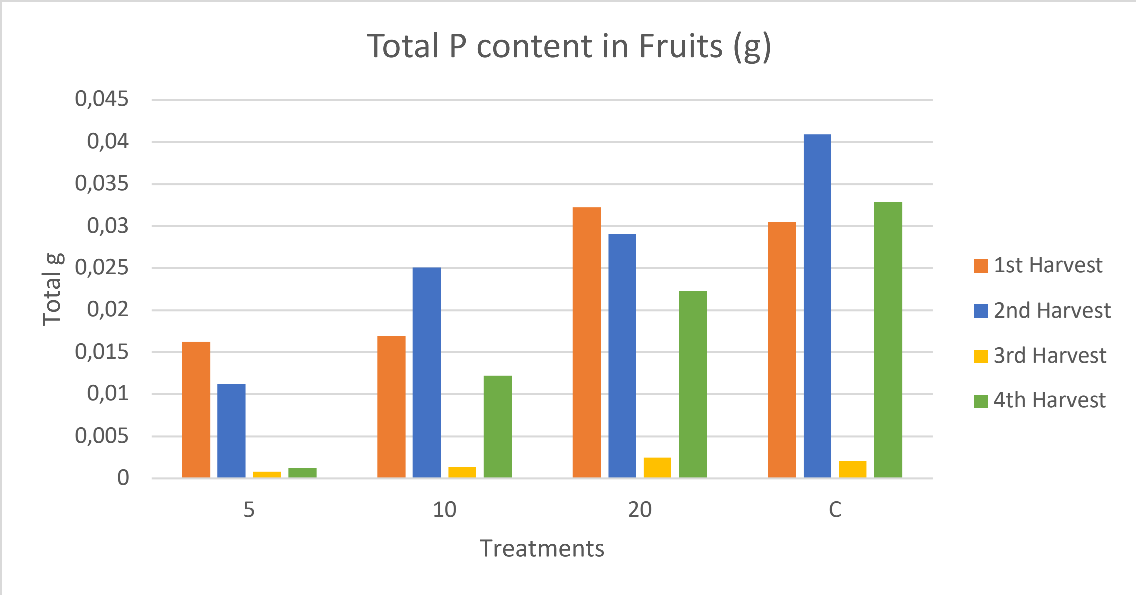
Annex A6. 8 Water flows recorded for the lettuce production. DAT= days after transplanting, IR= irrigated water (L/plant), LI= leachate water (L/plant), 5g= leachates generated from plants fertilized with 5g struvite, 10g= leachates generated from plants fertilized with 10g struvite, 20g= leachates generated from plants fertilized with 20g struvite, Control= leachates generated from control plants and water irrigated to control plants, Struvite= water irrigated to struvite fertilized treatments.



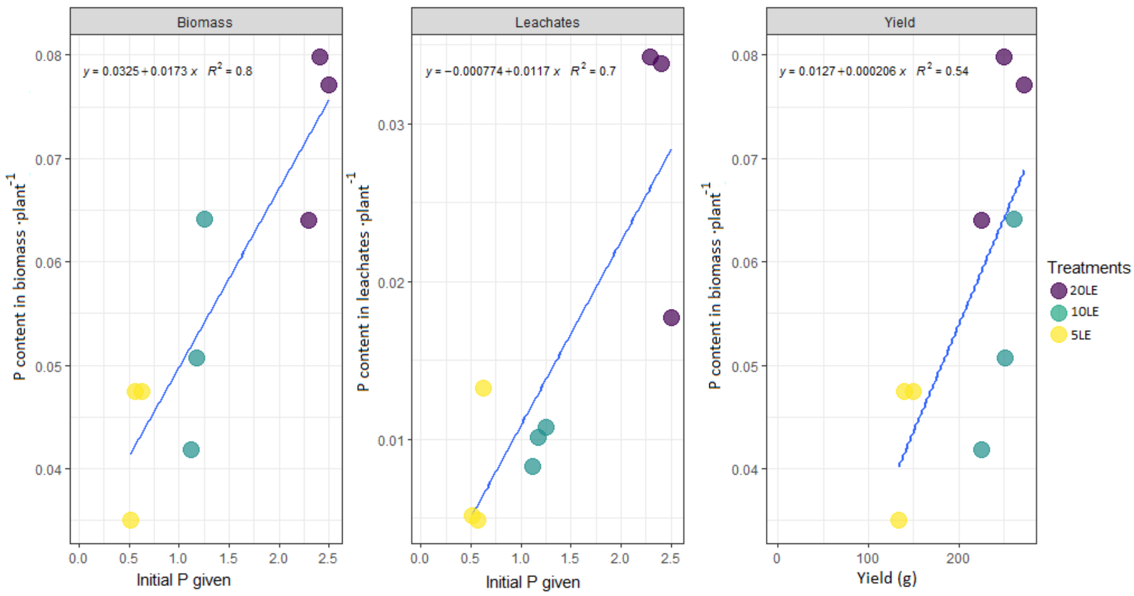
Annex A6. 9 *Water flows recorded for the pepper production. DAT= days after transplanting, IR= irrigated water (L/plant), LI= leachate water (L/plant), 5g= leachates generated from plants fertilized with 5g struvite, 10g= leachates generated from plants fertilized with 10g struvite, 20g= leachates generated from plants fertilized with 20g struvite, Control= leachates generated from control plants and water irrigated to control plants, Struvite= water irrigated to struvite fertilized treatments..*



Annex A6. 10 Accumulated P (g) recorded in the leachates generated per lettuce plant. DAT= days after transplanting, 5g= plants fertilized with 5g of struvite, 10g= plants fertilized with 10g of struvite, 20g= plants fertilized with 20g of struvite. Accumulated P (g) *recorded in the leachates generated per pepper plant. DAT= days after transplanting, 5g= plants fertilized with 5g of struvite, 10g= plants fertilized with 10g of struvite, 20g= plants fertilized with 20g of struvite.*



Annex A6. 11 P content (g) in pepper fruit harvests for treatments 5 = plants fertilized with 5g of struvite, 10 = plants fertilized with 10g of struvite, 20 = plants fertilized with 20g of struvite and C= control treatment. Values given for all harvests (1=55 DAT, , 2= 62 DAT, 3= 72 DAT and 4= 81 DAT).

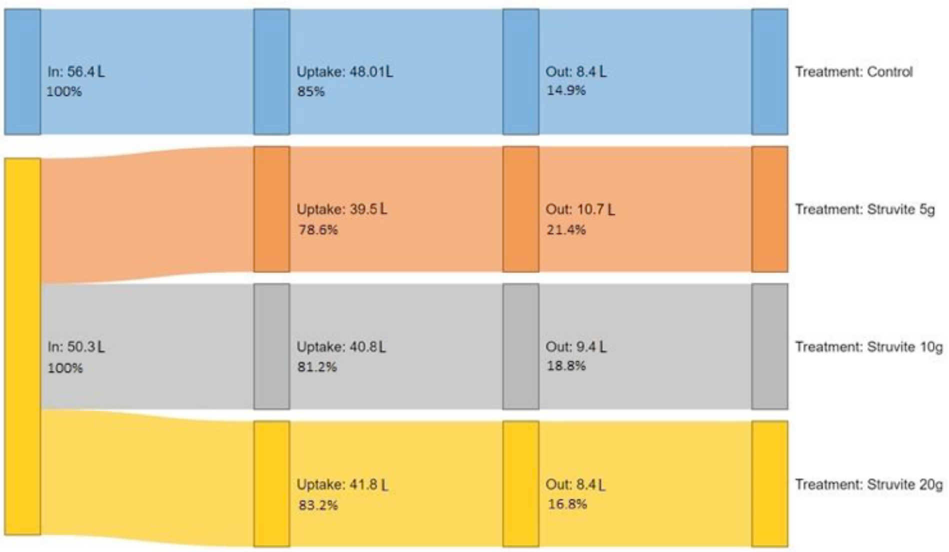


Annex A6. 12 Correlation between the initially given P to the plant (g) and the total P (g) found in the lettuce biomass.

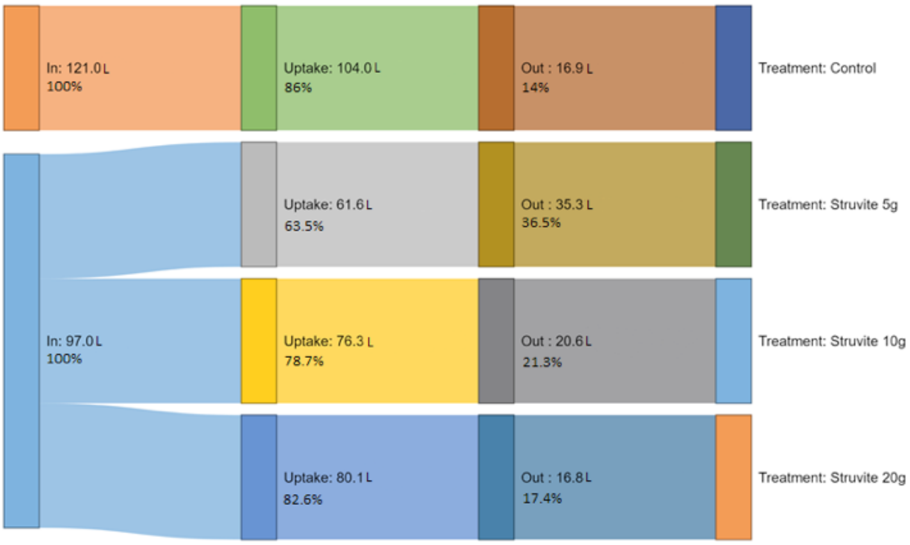
Annex A6. 13 Prospection of 9 lettuce crop cycles and balance flows for each cycle for lettuces with initial 20g, 10g and 5g of struvite. Key values labeled as L= P lost in leachates, B = P in lettuce biomass, Y = Prospective yield.

	1st Cycle				2nd Cycle				3rd Cycle				4th Cycle				5th Cycle		
	5g	10g	20g		5g	10g	20g		5g	10g	20g		5g	10g	20g		5g	10g	20g
L:	0.01	0.01	0.02	L:	0.00	0.01	0.03	L:	0.01	0.01	0.03	L:	0.01	0.01	0.03	L:	0.01	0.01	0.02
B:	0.05	0.06	0.07	B:	0.05	0.05	0.08	B:	0.03	0.04	0.06	B:	0.04	0.05	0.07	B:	0.04	0.05	0.07
Y:	225	249	273	Y:	224	252	261	Y:	133	140	150	Y:	171	198	249	Y:	169	195	244

	6th Cycle				7th Cycle				8th Cycle				9th Cycle				Total		
	5g	10g	20g		5g	10g	20g		5g	10g	20g		5g	10g	20g		5g	10g	20g
L:	0.01	0.01	0.02	L:	0.00	0.01	0.02	L:	0.00	0.01	0.02	L:	0.00	0.01	0.02	L:	0.05	0.10	0.23
B:	0.04	0.05	0.07	B:	0.04	0.05	0.07	B:	0.04	0.05	0.06	B:	0.04	0.05	0.06	B:	0.36	0.45	0.62
Y:	167	193	240	Y:	165	190	236	Y:	163	187	232	Y:	161	185	228	Y:	1580	1790	2113



Annex A6. 14 Water balance per Lettuce crop. In = incoming irrigation water, Uptake = evapotranspired and evaporated



Annex A6. 15 Water balance per Pepper crop. In = incoming irrigation water, Uptake = evapotranspired and evaporated water, Out = collected leachates a the end of the line.

Annex A6. 16 LCA Inventory for treatments 5LE/10LE/20LE/CLE/5P/10P/20P/CP

5LE			
<u>Fertilizers</u>	Materials/assemblies		
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0	kg
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0.1350	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.5801	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0.2183	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.1967	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0.2967	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0066	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0031	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.0006	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0025	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.0003	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0133	kg
MgCL2/ NaOH	Struvite	0.14	p
Processes Fertilizer Transport	Transport, van <3.5t/RER S	0.15	tkm
Emissions to Air	Ammonia	0.0034	kg
	Dinitrogen monoxide	0.0094	kg
	Nitrogen oxides	0.000 9	kg
Emissions to Water	Nitrogen	0.0222	kg

	Phosphorous	0.0006	kg
Struvite 1kg	Materials/assemblies		
MgO	Magnesium oxide {GLO}| market for | Cut-off, S	0.4239	kg
pH	HCl	0.766	kg
	Sodium hydroxide, without water, in 50% solution state {GLO}| market for | Cut-off, S	0.039	kg
	Tap water, at user/RER S	0.189	kg
	Processes		
Elictricity	Electricity, medium voltage {ES}| market for | Cut-off, S	0.523	kWh
Transport	Transport, van <3.5t/RER S	0.003	tkm

10LE			
Fertilizers	Materials/assemblies		
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0	kg
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.1350	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0.5801	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.2183	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0.1967	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.2967	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0066	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.0031	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0006	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.0025	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0003	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0.0133	kg
MgCL2/ NaOH	Struvite	0.28	p
Processes Fertilizer Transport	Transport, van <3.5t/RER S	0.15	tkm
Emissions to Air	Ammonia	0.0034	kg
	Dinitrogen monoxide	0.0174	kg
	Nitrogen oxides	0.0017	kg
Emissions to Water	Nitrogen	0.0125	kg
	Phosphorous	0.0008	kg
Struvite 1kg	Materials/assemblies		
MgO	Magnesium oxide {GLO}| market for | Cut-off, S	0.4239	kg
pH	HCl	0.766	kg
	Sodium hydroxide, without water, in 50% solution state {GLO}| market for | Cut-off, S	0.039	kg
	Tap water, at user/RER S	0.189	kg
	Processes		
Elictricity	Electricity, medium voltage {ES}| market for | Cut-off, S	0.523	kWh

Transport	Transport, van <3.5t/RER S	0.003	tkm

20LE			
Fertilizers	Materials/assemblies		
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0	kg
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0.1350	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.5801	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0.2183	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.1967	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0.2967	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0066	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0031	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.0006	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0025	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.0003	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0133	kg
MgCL2/ NaOH	Struvite	0.56	p
Processes Fertilizer Transport	Transport, van <3.5t/RER S	0.15	tkm
Emissions to Air	Ammonia	0.0034	kg
	Dinitrogen monoxide	0.0333	kg
	Nitrogen oxides	0.0033	kg
Emissions to Water	Nitrogen	0.0147	kg
	Phosphorous	0.0024	kg
Struvite 1kg	Materials/assemblies		
MgO	Magnesium oxide {GLO}| market for | Cut-off, S	0.4239	kg
pH	HCl	0.766	kg
	Sodium hydroxide, without water, in 50% solution state {GLO}| market for | Cut-off, S	0.039	kg
	Tap water, at user/RER S	0.189	kg
	Processes		
Elictricity	Electricity, medium voltage {ES}| market for | Cut-off, S	0.523	kWh
Transport	Transport, van <3.5t/RER S	0.003	tkm

Control LE			
Fertilizers	Materials/assemblies		
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0.1186	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0.0781	kg
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0.1461	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.6277	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0.2362	kg

CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.2128	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0.3210	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0073	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0034	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.0006	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0027	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.00028	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0144	kg
	50,7		
Processes			
Fertilizer Transport	Transport, van <3.5t/RER S	0.18	tkm
Emissions to Air	Ammonia	0.0036	kg
	Dinitrogen monoxide	0.0015	kg
	Nitrogen oxides	0.0002	kg
Emissions to Water	Nitrogen	0.0142	kg
	Phosphorous	0.0084	kg

5P			
<u>Fertilizers</u>	Materials/assemblies		
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0	kg
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0.0786	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.3376	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0.1270	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.1145	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0.1726	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0039	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0018	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.0004	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0015	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.0002	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0078	kg
MgCL2/ NaOH	Struvite	0.14	p
Processes			
Fertilizer Transport	Transport, van <3.5t/RER S	0.08	tkm
Emissions to Air	Ammonia	0.0019	kg
	Dinitrogen monoxide	0.0088	kg
	Nitrogen oxides	0.0008	kg
Emissions to Water	Nitrogen	0.0153	kg
	Phosphorous	0.0017	kg
<u>Struvite 1kg</u>	Materials/assemblies		

MgO	Magnesium oxide {GLO}| market for | Cut-off, S	0.4239	kg
pH	HCl	0.766	kg
	Sodium hydroxide, without water, in 50% solution state {GLO}| market for | Cut-off, S	0.039	kg
	Tap water, at user/RER S	0.189	kg
	Processes		
Elictricity	Electricity, medium voltage {ES}| market for | Cut-off, S	0.523	kW h
Transport	Transport, van <3.5t/RER S	0.003	tkm

10P			
<u>**Fertilizers**</u>	Materials/assemblies		
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0	kg
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.0786	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0.3376	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.1270	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0.1145	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.1726	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0039	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.0018	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0004	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.0015	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0002	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0.0078	kg
MgCL2/ NaOH	Struvite	0.28	p
Processes Fertilizer Transport	Transport, van <3.5t/RER S	0.08	tkm
Emissions to Air	Ammonia	0.0019	kg
	Dinitrogen monoxide	0.017	kg
	Nitrogen oxides	0.002	kg
Emissions to Water	Nitrogen	0.0069	kg
	Phosphorous	0.0002	kg
<u>**Struvite 1kg**</u>	Materials/assemblies		
MgO	Magnesium oxide {GLO}| market for | Cut-off, S	0.4239	kg
pH	HCl	0.766	kg
	Sodium hydroxide, without water, in 50% solution state {GLO}| market for | Cut-off, S	0.039	kg
	Tap water, at user/RER S	0.189	kg
	Processes		
Elictricity	Electricity, medium voltage {ES}| market for | Cut-off, S	0.523	kW h
Transport	Transport, van <3.5t/RER S	0.003	tkm

20P			
Fertilizers	Materials/assemblies		
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0	kg
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0.0786	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.3376	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0.1270	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.1145	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0.1726	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0039	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0018	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.0004	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0015	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.0002	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0078	kg
MgCL2/ NaOH	Struvite	0.56	p
Processes Fertilizer Transport	Transport, van <3.5t/RER S	0.08	tkm
Emissions to Air	Ammonia	0.0019	kg
	Dinitrogen monoxide	0.0333	kg
	Nitrogen oxides	0.0033	kg
Emissions to Water	Nitrogen	0.0062	kg
	Phosphorous	0.0004	kg
Struvite 1kg	Materials/assemblies		
MgO	Magnesium oxide {GLO}| market for | Cut-off, S	0.4239	kg
pH	HCl	0.766	kg
	Sodium hydroxide, without water, in 50% solution state {GLO}| market for | Cut-off, S	0.039	kg
	Tap water, at user/RER S	0.189	kg
	Processes		
Elictricity	Electricity, medium voltage {ES}| market for | Cut-off, S	0.523	kWh
Transport	Transport, van <3.5t/RER S	0.003	tkm

Control P			
Fertilizers	Materials/assemblies		
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0.0595	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0.0392	kg
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0.0733	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.3149	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0.1185	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.1068	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0.1611	kg

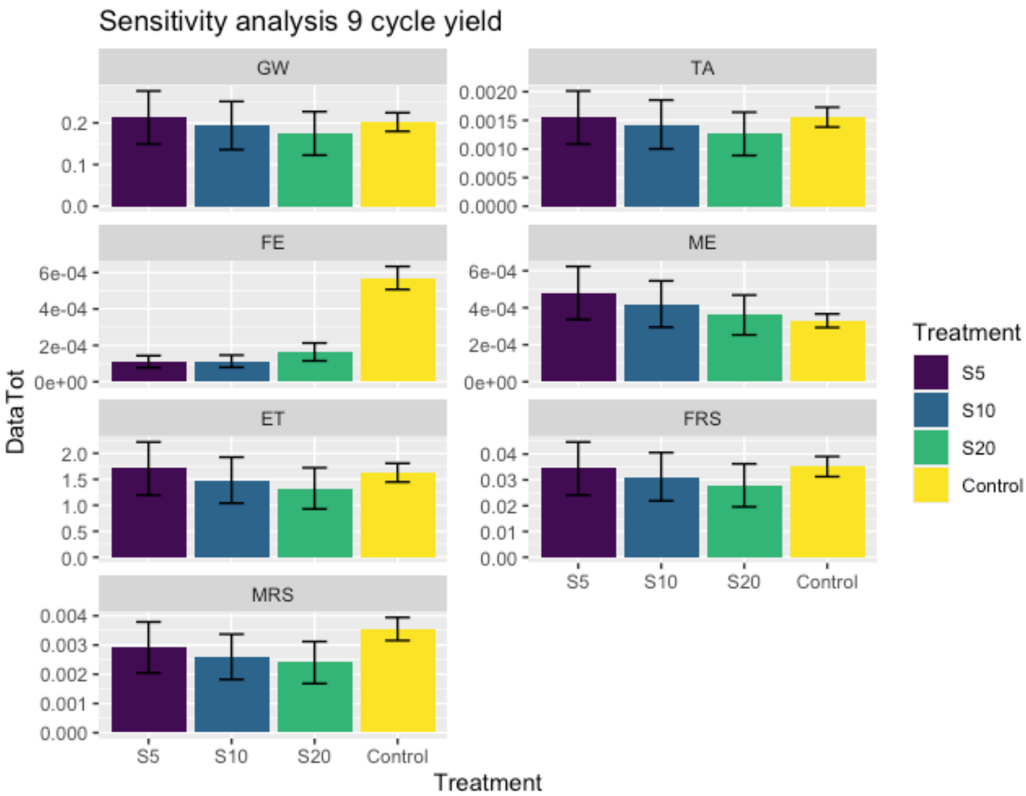
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0037	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0017	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.0003	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0014	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.0002	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.00724	kg
Processes			
Fertilizer Transport	Transport, van <3.5t/RER S	0.18	tkm
Emissions to Air	Ammonia	0.00182	kg
	Dinitrogen monoxide	0.0008	kg
	Nitrogen oxides	0.0001	kg
Emissions to Water	Nitrogen	0.0126	kg
	Phosphorous	0.0040	kg

Annex A6. 17 Sensitivity Analysis 9 cycle lettuce crop

Origin of uncertainty		
Basis of uncertainty →	Yield production based on r^2:0.56 correlation →	46% error in yield
Calculation of SD based on a total error of 46% SD for Control treatment yield set at 10%		

Treatment	IC	SD	Total DataFU9
S5	ET	0,51019171	1,70698721
S5	FE	3,3003E-05	0,00011042
S5	FRS	0,01028621	0,03441535
S5	GW	0,06361367	0,21283708
S5	ME	0,00014341	0,00047981
S5	MRS	0,00087233	0,00291861
S5	AT	0,00046307	0,00154934
S10	ET	0,44274616	1,48183647
S10	FE	3,3622E-05	0,00011253
S10	FRS	0,00932998	0,03122671
S10	GW	0,05783644	0,19357399
S10	ME	0,0001255	0,00042003
S10	MRS	0,00077547	0,00259543
S10	AT	0,00042637	0,00142703
S20	ET	0,39614857	1,325442
S20	FE	4,89E-05	0,00016361
S20	FRS	0,00833395	0,02788391
S20	GW	0,05219116	0,17462227
S20	ME	0,00010786	0,00036088
S20	MRS	0,00071788	0,0024019
S20	AT	0,00037777	0,00126394

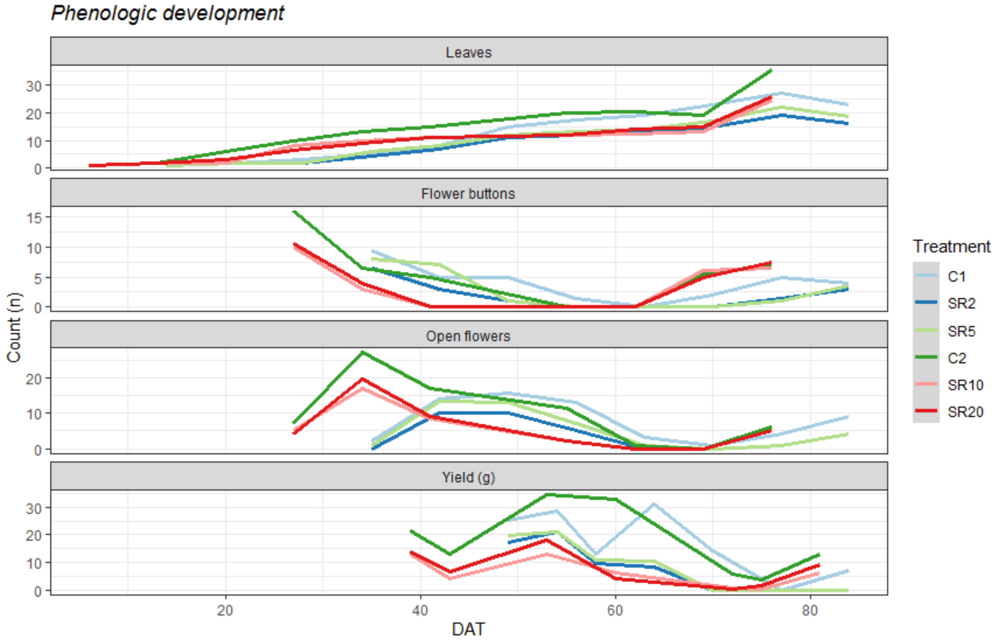
Control	ET	0,18078535	1,62706814
Control	FE	6,3327E-05	0,00056994
Control	FRS	0,00390856	0,03517707
Control	GW	0,02243944	0,201955
Control	ME	3,6652E-05	0,00032986
Control	MRS	0,00039427	0,00354847
Control	AT	0,00017285	0,00155561



Annex A6. 18 Sensitivity analysis for total impact

Annex A7. 1 Climatic conditions in the laboratory during the experiments

Temperature	*2019*	*2020*
Average T ºC	*18.94*	*20.48*
Minimum T ºC	*4.48*	*11.23*
Maximum T ºC	*29.89*	*29.48*
Standard Deviation	*2.09*	*4.68*
Realative Humidity	***2019***	***2020***
Average (RH)	*38.11*	*38.78*
Minimum (RH)	*5.65*	*7.51*
Maximum (RH)	*77.37*	*59.78*
Standard Deviation	*5.83*	*11.25*



Annex A7. 2 Phenologic development for all treatments during the experimental time.

*Annex A7. 3 Leachate flows from SR10 and SR20 treatments for N,P and Mg and percentage in regards to the applied in struvite. *Indicating an estimated percentage of the leachate nitrogen due to unknown assimilation of atmospheric N_2.*

Nitrogen	*Struvite (g)*	*Leachates (g)*	*(%)*
SR10	*0.57*	*0.079*	*11**
SR20	*1.14*	*0.1479*	*13**
Phosphorous	*Struvite (g)*	*Leachates (g)*	*(%)*
SR10	*1.25*	*0.106*	*8.5*
SR20	*2.5*	*0.277*	*11*
Magnesium	*Struvite (g)*	*Leachates (g)*	*(%)*
SR10	*0.99*	*0.180*	*18*
SR20	*1.98*	*0.303*	*15*

Annex A7. 4 LCA Inventary for treatment SR2

SR2			
Auxilary Equippment	Materials/assemblies	Amoun t	
Iron Pump + PS	Cast iron {GLO}| market for | Cut-off, S	0.2019	kg
Steel Pump + PS	Steel, low-alloyed {GLO}| market for | Cut-off, S	0.0238	kg
HDPE Pump + PS	Polyethylene, HDPE, granulate, at plant/RER S	0.0119	kg

HDPE Digital timer	Polyethylene, HDPE, granulate, at plant/RER S	0.0118	kg
Electronics Digital timer	Electronics, for control units {GLO}| market for | Cut-off, S	0.0006	kg
Dosatron	Polypropylene, granulate {GLO}| market for | Cut-off, S	0.0366	kg
Pipe 32 mm	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0113	kg
Pipe 25 mm	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0558	kg
Joints	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.1386	kg
Nutrient tank	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.1871	kg
Water Tank	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.3462	kg
Iron Flow Meter	Cast iron {GLO}| market for | Cut-off, S	0.1093	kg
HDPE Flow Meter	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0058	kg
Primary pipe	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	0.0564	kg
Secondary pipe	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	0.0697	kg
Joint	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0028	kg
Joint insertion	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0028	kg
Stopper	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0022	kg
Drip tube	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0236	kg
Drip	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0039	kg
Gripping piece	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0079	kg
Manometers	Cast iron {GLO}| market for | Cut-off, S	0.0069	kg
Manometers	Bronze {GLO}| market for | Cut-off, S	0.001	kg
Manometers	Glycerine {Europe without Switzerland}| esterification of rape oil | Cut-off, S	0.0087	kg
Processes			
Transport	Transport, van <3.5t/RER S	0.06633	tkm
Transport to end of Life	Transport, van <3.5t/RER S	0.06633	tkm
Injetion moulding	Injection moulding/RER S	0.9915	kg
Metal working	Metal working, average for metal product manufacturing {RER}| processing | Cut-off, S	0.3428	kg
Energy	Processes		
Water pump New Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Water pump Leachates Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Waterproof pump Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Fertilizers	Materials/assemblies		
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0	kg
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.93	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.23	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0111	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0051	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.00103	kg

18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0041	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.00044	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.022	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0	kg
MgCL2/ NaOH	Struvite	0.128	p
Processes			
Fertilizer Transport	Transport, van <3.5t/RER S	0.06658	tkm
Emissions to Air	Ammonia	0.00005925	kg
	Dinitrogen monoxide	2.46875E-05	kg
	Nitrogen oxides	0.0001975	kg
Emissions to Water	Nitrogen	0.001975	kg
	Phosphorous	0.002609	kg
<u>**Greenhouse Structure**</u>	Materials/assemblies		
Concrete	Concrete roof tile {GLO}| market for | Cut-off, S	6.09	kg
LDPE	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	2.24	kg
Polycarbonate	Polycarbonate {GLO}| market for | Cut-off, S	4.6	kg
Polyester	Glass fibre reinforced plastic, polyester resin, hand lay-up {GLO}| market for | Cut-off, S	0.224	kg
Aluminium	Aluminium, primary, ingot {IAI Area, Russia & RER w/o EU27 & EFTA}| aluminium production, primary, ingot | Cut-off, S	0.224	kg
	Steel, low-alloyed {RER}| steel production, electric, low-alloyed | Cut-off, S	24.04	kg
	Processes		
Transport	Transport, lorry >32t, EURO5/RER S	1.8709	tkm
Transoceanic freight ship	Transport, freight, sea, transoceanic ship {GLO}| market for | Cut-off, S	3.71	tkm
Machinery use	Energy, from diesel burned in machinery/RER Energy	0.0115	kWh
End of Life	Transport, lorry 16-32t, EURO5/RER S	1.323	tkm
<u>**Pesticides**</u>	Materials/assemblies		
Spintor	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
20% Potassium soap	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0,04*0,2	kg
80% Potassium soap	Water, deionised, from tap water, at user {Europe without Switzerland}| market for water, deionised, from tap water, at user | Cut-off, S	0,04*0,8	kg
Costar	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
Wettable sulphur	Sulfur {GLO}| market for | Cut-off, S	0	kg
MeemAzal	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
	Processes		
Pesticide Transport	Transport, van <3.5t/RER S	0.02	tkm
<u>**Rainwater Harvesting System**</u>	Materials/assemblies		
Water tank	Glass fibre reinforced plastic, polyamide, injection moulded {GLO}| market for | Cut-off, S	3.01	kg
Pipes	Polyethylene, HDPE, granulate, at plant/RER S	0.23	kg

Iron Pump	Cast iron {GLO}| market for | Cut-off, S	0.05	kg
Steel Pump	Steel, chromium steel 18/8 {GLO}| market for | Cut-off, S	0.01	kg
	Processes		
Transport GFRP	Transport, lorry 7.5-16t, EURO3/RER S	0.36	tkm
Transport pipes and pump	Transport, van <3.5t/RER S	0.03	tkm
Excavation	Excavation, hydraulic digger {GLO}| market for | Cut-off, S	0.07	m3
Transport End of Life	Transport, lorry 3.5-7.5t, EURO5/RER S	0.2	tkm
Injection moulding	Injection moulding/RER S	3.23	kg
Metal working	Metal working, average for metal product manufacturing {RER}| processing | Cut-off, S	0.05	kg
<u>Struvite 1kg</u>	Materials/assemblies		
MgO	Magnesium oxide {GLO}| market for | Cut-off, S	0.4239	kg
pH	HCl	0.766	kg
	Sodium hydroxide, without water, in 50% solution state {GLO}| market for | Cut-off, S	0.039	kg
	Tap water, at user/RER S	0.189	kg
	Processes		
Elictricity	Electricity, medium voltage {ES}| market for | Cut-off, S	0.523	kWh
Transport	Transport, van <3.5t/RER S	0.003	tkm
<u>Substrate</u>	Materials/assemblies		
HDPE Substrate	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.12	kg
Perlite	Perlite {GLO}| market for | Cut-off, S	4.39	kg
	Processes		
Transport	Transport, lorry 16-32t, EURO5/RER S	3.544	tkm

Annex A7. 5 LCA Inventary for treatment SR5

SR5			
<u>Auxilary Equippment</u>	Materials/assemblies	Amount	
Iron Pump + PS	Cast iron {GLO}| market for | Cut-off, S	0.2019	kg
Steel Pump + PS	Steel, low-alloyed {GLO}| market for | Cut-off, S	0.0238	kg
HDPE Pump + PS	Polyethylene, HDPE, granulate, at plant/RER S	0.0119	kg
HDPE Digital timer	Polyethylene, HDPE, granulate, at plant/RER S	0.0118	kg
Electronics Digital timer	Electronics, for control units {GLO}| market for | Cut-off, S	0.0006	kg
Dosatron	Polypropylene, granulate {GLO}| market for | Cut-off, S	0.0366	kg
Pipe 32 mm	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0113	kg
Pipe 25 mm	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0558	kg
Joints	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.1386	kg
Nutrient tank	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.1871	kg
Water Tank	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.3462	kg
Iron Flow Meter	Cast iron {GLO}| market for | Cut-off, S	0.1093	kg
HDPE Flow Meter	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0058	kg

Primary pipe	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	0.0564	kg
Secondary pipe	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	0.0697	kg
Joint	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0028	kg
Joint insertion	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0028	kg
Stopper	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0022	kg
Drip tube	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0236	kg
Drip	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0039	kg
Gripping piece	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0079	kg
Manometers	Cast iron {GLO}| market for | Cut-off, S	0.0069	kg
Manometers	Bronze {GLO}| market for | Cut-off, S	0.001	kg
Manometers	Glycerine {Europe without Switzerland}| esterification of rape oil | Cut-off, S	0.0087	kg
	Processes		
Transport	Transport, van <3.5t/RER S	0.06633	tkm
Transport to end of Life	Transport, van <3.5t/RER S	0.06633	tkm
Injetion moulding	Injection moulding/RER S	0.9915	kg
Metal working	Metal working, average for metal product manufacturing {RER}| processing | Cut-off, S	0.3428	kg
<u>Energy</u>	Processes		
Water pump New Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Water pump Leachates Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Waterproof pump Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
<u>Fertilizers</u>	Materials/assemblies		
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0	kg
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.93	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.23	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0111	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0051	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.00103	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0041	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.00044	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.022	kg
MgCL2/ NaOH	Struvite	0.32	p
Processes			
Fertilizer Transport	Transport, van <3.5t/RER S	0.0761885	tkm
Emissions to Air	Ammonia	0.0000669	kg
	Dinitrogen monoxide	0.000027875	kg
	Nitrogen oxides	0.000223	kg

Emissions to Water	Nitrogen	0.00223	kg
	Phosphorous	0.0035	kg
Greenhouse Structure	Materials/assemblies		
Concrete	Concrete roof tile {GLO}| market for | Cut-off, S	6.09	kg
LDPE	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	2.24	kg
Polycarbonate	Polycarbonate {GLO}| market for | Cut-off, S	4.6	kg
Polyester	Glass fibre reinforced plastic, polyester resin, hand lay-up {GLO}| market for | Cut-off, S	0.224	kg
Aluminium	Aluminium, primary, ingot {IAI Area, Russia & RER w/o EU27 & EFTA}| aluminium production, primary, ingot | Cut-off, S	0.224	kg
	Steel, low-alloyed {RER}| steel production, electric, low-alloyed | Cut-off, S	24.04	kg
	Processes		
Transport	Transport, lorry >32t, EURO5/RER S	1.8709	tkm
Transoceanic freight ship	Transport, freight, sea, transoceanic ship {GLO}| market for | Cut-off, S	3.71	tkm
Machinery use	Energy, from diesel burned in machinery/RER Energy	0.0115	kWh
End of Life	Transport, lorry 16-32t, EURO5/RER S	1.323	tkm
Pesticides	Materials/assemblies		
Spintor	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
20% Potassium soap	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0,04*0,2	kg
80% Potassium soap	Water, deionised, from tap water, at user {Europe without Switzerland}| market for water, deionised, from tap water, at user | Cut-off, S	0,04*0,8	kg
Costar	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
Wettable sulphur	Sulfur {GLO}| market for | Cut-off, S	0	kg
MeemAzal	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
	Processes		
Pesticide Transport	Transport, van <3.5t/RER S	0.02	tkm
Rainwater Harvesting System	Materials/assemblies		
Water tank	Glass fibre reinforced plastic, polyamide, injection moulded {GLO}| market for | Cut-off, S	3.01	kg
Pipes	Polyethylene, HDPE, granulate, at plant/RER S	0.23	kg
Iron Pump	Cast iron {GLO}| market for | Cut-off, S	0.05	kg
Steel Pump	Steel, chromium steel 18/8 {GLO}| market for | Cut-off, S	0.01	kg
	Processes		
Transport GFRP	Transport, lorry 7.5-16t, EURO3/RER S	0.36	tkm
Transport pipes and pump	Transport, van <3.5t/RER S	0.03	tkm
Excavation	Excavation, hydraulic digger {GLO}| market for | Cut-off, S	0.07	m3
Transport End of Life	Transport, lorry 3.5-7.5t, EURO5/RER S	0.2	tkm
Injection moulding	Injection moulding/RER S	3.23	kg
Metal working	Metal working, average for metal product manufacturing {RER}| processing | Cut-off, S	0.05	kg
Struvite 1kg	Materials/assemblies		

MgO	Magnesium oxide {GLO}| market for | Cut-off, S	0.4239	kg
pH	HCl	0.766	kg
	Sodium hydroxide, without water, in 50% solution state {GLO}| market for | Cut-off, S	0.039	kg
	Tap water, at user/RER S	0.189	kg
	Processes		
Elictricity	Electricity, medium voltage {ES}| market for | Cut-off, S	0.523	kWh
Transport	Transport, van <3.5t/RER S	0.003	tkm
Substrate	Materials/assemblies		
HDPE Substrate	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.12	kg
Perlite	Perlite {GLO}| market for | Cut-off, S	4.39	kg
	Processes		
Transport	Transport, lorry 16-32t, EURO5/RER S	3.544	tkm

Annex A7. 6 LCA Inventary for treatment SR10

SR10			
Auxilary Equippment	Materials/assemblies	Amount	
Iron Pump + PS	Cast iron {GLO}| market for | Cut-off, S	0.2019	kg
Steel Pump + PS	Steel, low-alloyed {GLO}| market for | Cut-off, S	0.0238	kg
HDPE Pump + PS	Polyethylene, HDPE, granulate, at plant/RER S	0.0119	kg
HDPE Digital timer	Polyethylene, HDPE, granulate, at plant/RER S	0.0118	kg
Electronics Digital timer	Electronics, for control units {GLO}| market for | Cut-off, S	0.0006	kg
Dosatron	Polypropylene, granulate {GLO}| market for | Cut-off, S	0.0366	kg
Pipe 32 mm	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0113	kg
Pipe 25 mm	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0558	kg
Joints	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.1386	kg
Nutrient tank	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.1871	kg
Water Tank	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.3462	kg
Iron Flow Meter	Cast iron {GLO}| market for | Cut-off, S	0.1093	kg
HDPE Flow Meter	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0058	kg
Primary pipe	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	0.0564	kg
Secondary pipe	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	0.0697	kg
Joint	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0028	kg
Joint insertion	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0028	kg
Stopper	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0022	kg
Drip tube	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0236	kg
Drip	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0039	kg
Gripping piece	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0079	kg

Manometers	Cast iron {GLO}| market for | Cut-off, S	0.0069	kg
Manometers	Bronze {GLO}| market for | Cut-off, S	0.001	kg
Manometers	Glycerine {Europe without Switzerland}| esterification of rape oil | Cut-off, S	0.0087	kg
Processes			
Transport	Transport, van <3.5t/RER S	0.06633	tkm
Transport to end of Life	Transport, van <3.5t/RER S	0.06633	tkm
Injetion moulding	Injection moulding/RER S	0.9915	kg
Metal working	Metal working, average for metal product manufacturing {RER}| processing | Cut-off, S	0.3428	kg
Energy	Processes		
Water pump New Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Water pump Leachates Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Waterproof pump Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Fertilizers	Materials/assemblies		
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0	kg
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.93	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.23	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0111	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0051	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.00103	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0041	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.00044	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.022	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0	kg
MgCL2/ NaOH	Struvite	0.64	p
Processes			
Fertilizer Transport	Transport, van <3.5t/RER S	0.092	tkm
Emissions to Air	Ammonia	0.00010944	kg
	Dinitrogen monoxide	0.000456	kg
	Nitrogen oxides	0.003648	kg
Emissions to Water	Nitrogen	0.0051	kg
	Phosphorous	0.0068	kg
Greenhouse Structure	Materials/assemblies		
Concrete	Concrete roof tile {GLO}| market for | Cut-off, S	6.09	kg
LDPE	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	2.24	kg
Polycarbonate	Polycarbonate {GLO}| market for | Cut-off, S	4.6	kg
Polyester	Glass fibre reinforced plastic, polyester resin, hand lay-up {GLO}| market for | Cut-off, S	0.224	kg

Aluminium	Aluminium, primary, ingot {IAI Area, Russia & RER w/o EU27 & EFTA}| aluminium production, primary, ingot | Cut-off, S	0.224	kg
	Steel, low-alloyed {RER}| steel production, electric, low-alloyed | Cut-off, S	24.04	kg
	Processes		
Transport	Transport, lorry >32t, EURO5/RER S	1.8709	tkm
Transoceanic freight ship	Transport, freight, sea, transoceanic ship {GLO}| market for | Cut-off, S	3.71	tkm
Machinery use	Energy, from diesel burned in machinery/RER Energy	0.0115	kWh
End of Life	Transport, lorry 16-32t, EURO5/RER S	1.323	tkm
Pesticides	Materials/assemblies		
Spintor	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
20% Potassium soap	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0,04*0,2	kg
80% Potassium soap	Water, deionised, from tap water, at user {Europe without Switzerland}| market for water, deionised, from tap water, at user | Cut-off, S	0,04*0,8	kg
Costar	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
Wettable sulphur	Sulfur {GLO}| market for | Cut-off, S	0	kg
MeemAzal	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
	Processes		
Pesticide Transport	Transport, van <3.5t/RER S	0.02	tkm
Rainwater Harvesting System	Materials/assemblies		
Water tank	Glass fibre reinforced plastic, polyamide, injection moulded {GLO}| market for | Cut-off, S	3.01	kg
Pipes	Polyethylene, HDPE, granulate, at plant/RER S	0.23	kg
Iron Pump	Cast iron {GLO}| market for | Cut-off, S	0.05	kg
Steel Pump	Steel, chromium steel 18/8 {GLO}| market for | Cut-off, S	0.01	kg
	Processes		
Transport GFRP	Transport, lorry 7.5-16t, EURO3/RER S	0.36	tkm
Transport pipes and pump	Transport, van <3.5t/RER S	0.03	tkm
Excavation	Excavation, hydraulic digger {GLO}| market for | Cut-off, S	0.07	m3
Transport End of Life	Transport, lorry 3.5-7.5t, EURO5/RER S	0.2	tkm
Injection moulding	Injection moulding/RER S	3.23	kg
Metal working	Metal working, average for metal product manufacturing {RER}| processing | Cut-off, S	0.05	kg
Struvite 1kg	Materials/assemblies		
MgO	Magnesium oxide {GLO}| market for | Cut-off, S	0.4239	kg
pH	HCl	0.766	kg
	Sodium hydroxide, without water, in 50% solution state {GLO}| market for | Cut-off, S	0.039	kg
	Tap water, at user/RER S	0.189	kg
	Processes		
Elictricity	Electricity, medium voltage {ES}| market for | Cut-off, S	0.523	kWh
Transport	Transport, van <3.5t/RER S	0.003	tkm

Substrate	Materials/assemblies		
HDPE Substrate	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.12	kg
Perlite	Perlite {GLO}| market for | Cut-off, S	4.39	kg
	Processes		
Transport	Transport, lorry 16-32t, EURO5/RER S	3.544	tkm

Annex A7. 7 LCA Inventary for treatment SR20

SR20			
Auxilary Equippment	Materials/assemblies	Amount	
Iron Pump + PS	Cast iron {GLO}| market for | Cut-off, S	0.2019	kg
Steel Pump + PS	Steel, low-alloyed {GLO}| market for | Cut-off, S	0.0238	kg
HDPE Pump + PS	Polyethylene, HDPE, granulate, at plant/RER S	0.0119	kg
HDPE Digital timer	Polyethylene, HDPE, granulate, at plant/RER S	0.0118	kg
Electronics Digital timer	Electronics, for control units {GLO}| market for | Cut-off, S	0.0006	kg
Dosatron	Polypropylene, granulate {GLO}| market for | Cut-off, S	0.0366	kg
Pipe 32 mm	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0113	kg
Pipe 25 mm	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0558	kg
Joints	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.1386	kg
Nutrient tank	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.1871	kg
Water Tank	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.3462	kg
Iron Flow Meter	Cast iron {GLO}| market for | Cut-off, S	0.1093	kg
HDPE Flow Meter	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0058	kg
Primary pipe	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	0.0564	kg
Secondary pipe	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	0.0697	kg
Joint	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0028	kg
Joint insertion	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0028	kg
Stopper	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0022	kg
Drip tube	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0236	kg
Drip	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0039	kg
Gripping piece	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0079	kg
Manometers	Cast iron {GLO}| market for | Cut-off, S	0.0069	kg
Manometers	Bronze {GLO}| market for | Cut-off, S	0.001	kg
Manometers	Glycerine {Europe without Switzerland}| esterification of rape oil | Cut-off, S	0.0087	kg
	Processes		
Transport	Transport, van <3.5t/RER S	0.06633	tkm
Transport to end of Life	Transport, van <3.5t/RER S	0.06633	tkm
Injetion moulding	Injection moulding/RER S	0.9915	kg
Metal working	Metal working, average for metal product manufacturing {RER}| processing | Cut-off, S	0.3428	kg
Energy	Processes		
Water pump New Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh

Water pump Leachates Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Waterproof pump Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
<u>Fertilizers</u>	Materials/assemblies		
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0	kg
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.93	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.23	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0111	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.0051	kg
4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.00103	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.0041	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.00044	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.022	kg
MgCL2/ NaOH	Struvite	1.28	p
Processes			
Fertilizer Transport	Transport, van <3.5t/RER S	0.12415	tkm
Emissions to Air	Ammonia	0.00021888	kg
	Dinitrogen monoxide	0.000912	kg
	Nitrogen oxides	0.007296	kg
Emissions to Water	Nitrogen	0.0095	kg
	Phosphorous	0.0177	kg
<u>Greenhouse Structure</u>	Materials/assemblies		
Concrete	Concrete roof tile {GLO}| market for | Cut-off, S	6.09	kg
LDPE	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	2.24	kg
Polycarbonate	Polycarbonate {GLO}| market for | Cut-off, S	4.6	kg
Polyester	Glass fibre reinforced plastic, polyester resin, hand lay-up {GLO}| market for | Cut-off, S	0.224	kg
Aluminium	Aluminium, primary, ingot {IAI Area, Russia & RER w/o EU27 & EFTA}| aluminium production, primary, ingot | Cut-off, S	0.224	kg
	Steel, low-alloyed {RER}| steel production, electric, low-alloyed | Cut-off, S	24.04	kg
	Processes		
Transport	Transport, lorry >32t, EURO5/RER S	1.8709	tkm
Transoceanic freight ship	Transport, freight, sea, transoceanic ship {GLO}| market for | Cut-off, S	3.71	tkm
Machinery use	Energy, from diesel burned in machinery/RER Energy	0.0115	kWh
End of Life	Transport, lorry 16-32t, EURO5/RER S	1.323	tkm
<u>Pesticides</u>	Materials/assemblies		
Spintor	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
20% Potassium soap	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0,04*0,2	kg

80% Potassium soap	Water, deionised, from tap water, at user {Europe without Switzerland}| market for water, deionised, from tap water, at user | Cut-off, S	0,04*0,8	kg
Costar	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
Wettable sulphur	Sulfur {GLO}| market for | Cut-off, S	0	kg
MeemAzal	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
	Processes		
Pesticide Transport	Transport, van <3.5t/RER S	0.02	tkm
<u>Rainwater Harvesting System</u>	Materials/assemblies		
Water tank	Glass fibre reinforced plastic, polyamide, injection moulded {GLO}| market for | Cut-off, S	3.01	kg
Pipes	Polyethylene, HDPE, granulate, at plant/RER S	0.23	kg
Iron Pump	Cast iron {GLO}| market for | Cut-off, S	0.05	kg
Steel Pump	Steel, chromium steel 18/8 {GLO}| market for | Cut-off, S	0.01	kg
	Processes		
Transport GFRP	Transport, lorry 7.5-16t, EURO3/RER S	0.36	tkm
Transport pipes and pump	Transport, van <3.5t/RER S	0.03	tkm
Excavation	Excavation, hydraulic digger {GLO}| market for | Cut-off, S	0.07	m3
Transport End of Life	Transport, lorry 3.5-7.5t, EURO5/RER S	0.2	tkm
Injection moulding	Injection moulding/RER S	3.23	kg
Metal working	Metal working, average for metal product manufacturing {RER}| processing | Cut-off, S	0.05	kg
<u>Struvite 1kg</u>	Materials/assemblies		
MgO	Magnesium oxide {GLO}| market for | Cut-off, S	0.4239	kg
pH	HCl	0.766	kg
	Sodium hydroxide, without water, in 50% solution state {GLO}| market for | Cut-off, S	0.039	kg
	Tap water, at user/RER S	0.189	kg
	Processes		
Elictricity	Electricity, medium voltage {ES}| market for | Cut-off, S	0.523	kWh
Transport	Transport, van <3.5t/RER S	0.003	tkm
<u>Substrate</u>	Materials/assemblies		
HDPE Substrate	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.12	kg
Perlite	Perlite {GLO}| market for | Cut-off, S	4.39	kg
	Processes		
Transport	Transport, lorry 16-32t, EURO5/RER S	3.544	tkm

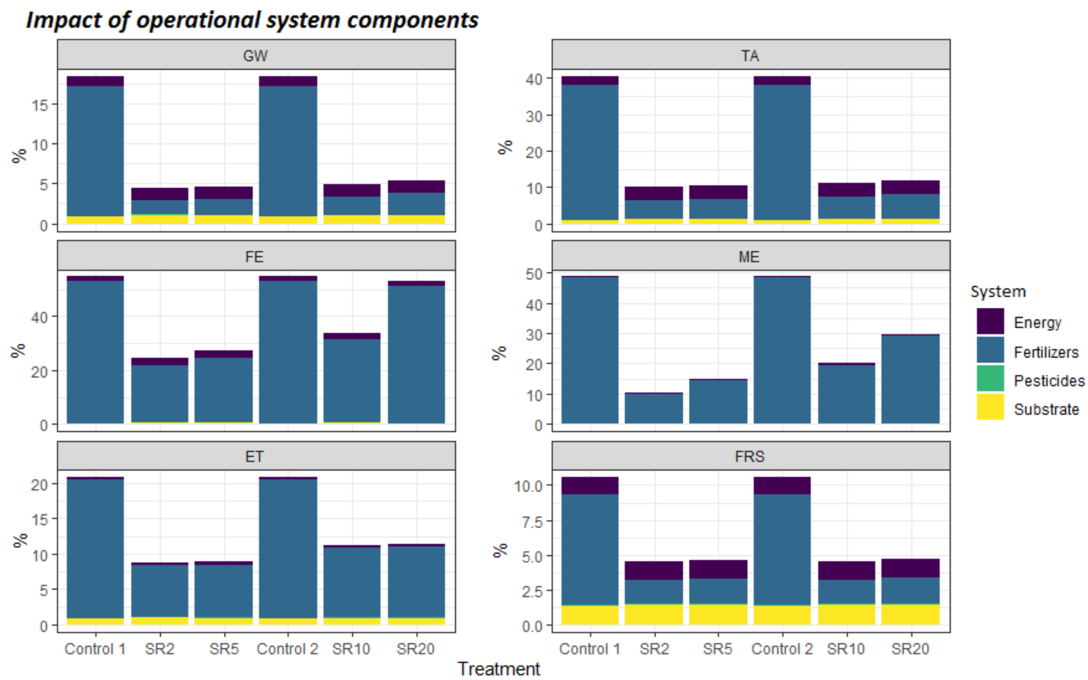
Annex A7. 8 LCA Inventary for the Control treatment

Control			
<u>Auxilary Equippment</u>	Materials/assemblies	Amount	
Iron Pump + PS	Cast iron {GLO}| market for | Cut-off, S	0.2019	kg
Steel Pump + PS	Steel, low-alloyed {GLO}| market for | Cut-off, S	0.0238	kg

HDPE Pump + PS	Polyethylene, HDPE, granulate, at plant/RER S	0.0119	kg
HDPE Digital timer	Polyethylene, HDPE, granulate, at plant/RER S	0.0118	kg
Electronics Digital timer	Electronics, for control units {GLO}| market for | Cut-off, S	0.0006	kg
Dosatron	Polypropylene, granulate {GLO}| market for | Cut-off, S	0.0366	kg
Pipe 32 mm	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0113	kg
Pipe 25 mm	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0558	kg
Joints	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.1386	kg
Nutrient tank	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.1871	kg
Water Tank	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.3462	kg
Iron Flow Meter	Cast iron {GLO}| market for | Cut-off, S	0.1093	kg
HDPE Flow Meter	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0058	kg
Primary pipe	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	0.0564	kg
Secondary pipe	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	0.0697	kg
Joint	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0028	kg
Joint insertion	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0028	kg
Stopper	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.0022	kg
Drip tube	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0236	kg
Drip	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0039	kg
Gripping piece	Polyvinylchloride, bulk polymerised {GLO}| market for | Cut-off, S	0.0079	kg
Manometers	Cast iron {GLO}| market for | Cut-off, S	0.0069	kg
Manometers	Bronze {GLO}| market for | Cut-off, S	0.001	kg
Manometers	Glycerine {Europe without Switzerland}| esterification of rape oil | Cut-off, S	0.0087	kg
Processes			
Transport	Transport, van <3.5t/RER S	0.06633	tkm
Transport to end of Life	Transport, van <3.5t/RER S	0.06633	tkm
Injetion moulding	Injection moulding/RER S	0.9915	kg
Metal working	Metal working, average for metal product manufacturing {RER}| processing | Cut-off, S	0.3428	kg
Energy	Processes		
Water pump New Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Water pump Leachates Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Waterproof pump Water RTG	Electricity, high voltage, production ES, at grid/ES S	1.01	kWh
Fertilizers	Materials/assemblies		
60,3% KPO4H2	Phosphate fertiliser, as P2O5 {GLO}| market for | Cut-off, S	0.221	kg
39,7% KPO4H2	Potassium fertiliser, as K2O {GLO}| market for | Cut-off, S	0.145	kg
KNO3	Potassium nitrate {GLO}| market for | Cut-off, S	0.21	kg
K2SO4	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0.59	kg
CaNO32	Calcium nitrate {GLO}| market for | Cut-off, S	0.74	kg
CaCl2	Calcium chloride {RER}| market for calcium chloride | Cut-off, S	0.23	kg
MgNO32	Magnesium oxide {GLO}| market for | Cut-off, S	0.382	kg
50,7% Hortilon	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.0111	kg
23,6% Hortilon	Magnesium oxide {GLO}| market for | Cut-off, S	0.005	kg

4,7% Hortilon	Zinc monosulfate {RER}| market for zinc monosulfate | Cut-off, S	0.001	kg
18,9% Hortilon	Copper oxide {GLO}| market for | Cut-off, S	0.004	kg
2% Hortilon	Molybdenite {GLO}| market for | Cut-off, S	0.00044	kg
Sequestrene	Iron sulfate {RER}| market for iron sulfate | Cut-off, S	0.022	kg
Processes			
Fertilizer Transport	Transport, van <3.5t/RER S	3.053572	tkm
Emissions to Air	Ammonia	0.0686	kg
	Dinitrogen monoxide	0.02859	kg
	Nitrogen oxides	0.00287	kg
Emissions to Water	Nitrogen	0.0206	kg
	Phosphorous	0.0175	kg
Greenhouse Structure	Materials/assemblies		
Concrete	Concrete roof tile {GLO}| market for | Cut-off, S	6.09	kg
LDPE	Polyethylene, low density, granulate {GLO}| market for | Cut-off, S	2.24	kg
Polycarbonate	Polycarbonate {GLO}| market for | Cut-off, S	4.6	kg
Polyester	Glass fibre reinforced plastic, polyester resin, hand lay-up {GLO}| market for | Cut-off, S	0.224	kg
Aluminium	Aluminium, primary, ingot {IAI Area, Russia & RER w/o EU27 & EFTA}| aluminium production, primary, ingot | Cut-off, S	0.224	kg
	Steel, low-alloyed {RER}| steel production, electric, low-alloyed | Cut-off, S	24.04	kg
	Processes		
Transport	Transport, lorry >32t, EURO5/RER S	1.8709	tkm
Transoceanic freight ship	Transport, freight, sea, transoceanic ship {GLO}| market for | Cut-off, S	3.71	tkm
Machinery use	Energy, from diesel burned in machinery/RER Energy	0.0115	kWh
End of Life	Transport, lorry 16-32t, EURO5/RER S	1.323	tkm
Pesticides	Materials/assemblies		
Spintor	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
20% Potassium soap	Potassium sulfate, as K2O {GLO}| market for | Cut-off, S	0,04*0,2	kg
80% Potassium soap	Water, deionised, from tap water, at user {Europe without Switzerland}| market for water, deionised, from tap water, at user | Cut-off, S	0,04*0,8	kg
Costar	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
Wettable sulphur	Sulfur {GLO}| market for | Cut-off, S	0	kg
MeemAzal	Pesticide, unspecified {GLO}| market for | Cut-off, S	0	kg
	Processes		
Pesticide Transport	Transport, van <3.5t/RER S	0.02	tkm
Rainwater Harvesting System	Materials/assemblies		
Water tank	Glass fibre reinforced plastic, polyamide, injection moulded {GLO}| market for | Cut-off, S	3.01	kg
Pipes	Polyethylene, HDPE, granulate, at plant/RER S	0.23	kg
Iron Pump	Cast iron {GLO}| market for | Cut-off, S	0.05	kg
Steel Pump	Steel, chromium steel 18/8 {GLO}| market for | Cut-off, S	0.01	kg

	Processes		
Transport GFRP	Transport, lorry 7.5-16t, EURO3/RER S	0.36	tkm
Transport pipes and pump	Transport, van <3.5t/RER S	0.03	tkm
Excavation	Excavation, hydraulic digger {GLO}| market for | Cut-off, S	0.07	m3
Transport End of Life	Transport, lorry 3.5-7.5t, EURO5/RER S	0.2	tkm
Injection moulding	Injection moulding/RER S	3.23	kg
Metal working	Metal working, average for metal product manufacturing {RER}| processing | Cut-off, S	0.05	kg
<u>Substrate</u>	Materials/assemblies		
HDPE Substrate	Polyethylene, high density, granulate {GLO}| market for | Cut-off, S	0.12	kg
Perlite	Perlite {GLO}| market for | Cut-off, S	4.39	kg
	Processes		
Transport	Transport, lorry 16-32t, EURO5/RER S	3.544	tkm



Annex A7. 9 Environmental performance of the operation System in % per IC.