IMPACT OF ENGINEERED NANOPARTICLES ON THE GROWTH OF ROOTS

By

Uday Bhagwan Gharge

Soubantika Palchoudhury
Assistant Professor of Engineering
(Chair)

Abdollah Arabshahi
Professor of Engineering
(Committee Member)

Manuel Santiago
Professor of Chemistry
(Committee Member)
IMPACT OF ENGINEERED NANOPARTICLES
ON THE GROWTH OF ROOTS

By
Uday Bhagwan Gharge

A Thesis Submitted to the Faculty of the University of Tennessee at Chattanooga in Partial Fulfillment of the Requirements of the Degree of Master of Engineering

The University of Tennessee at Chattanooga
Chattanooga, Tennessee

May 2018
ABSTRACT

There is an increasing demand for food and bioenergy crops for growing world population. Conventional fertilizers used for increasing agricultural production are required to be added to the soil in high quantity and have a slow absorption rate in plants. In comparison, engineered nanoparticles can be highly attractive as fertilizers as their small size will allow faster absorption. In particular, iron oxide-based nanoparticles will be highly promising as Fe-deficiency fertilizers because iron is required for photosynthesis in plants. We have synthesized iron oxide and hybrid iron oxide as nanoparticle fertilizers to study the significance on root growth of five different seeds: *Pisum sativum* L., *Cicer arietinum*, *Vigna radiate*, and *Phaseolus vulgaris*. We found iron oxide nanoparticles at low concentration (5.54x10^{-3} mgL^{-1} Fe) significantly increased root growth, compared to other growth solutions. The study results will be highly useful for increasing agricultural production.

Keywords: Iron oxide nanoparticles, hybrid Pt-iron oxide nanoparticles, root growth, microscopy, engineered nanoparticle fertilizers.
ACKNOWLEDGEMENTS

I would first like to thank my thesis advisor, Dr. Soubantika Palchoudhury, for continuous support of my master’s education and research work, and for her immense knowledge, patience, and encouragement. I would also like to thank the experts Dr. Abdollah Arabshahi and Dr. Manuel Santiago, who were involved as thesis committee members; without their support, guidance, and proper direction, this research work could not have been successfully conducted.

This work was performed, in part, at the Center for Integrated Nanotechnologies—an Office of Science User Facility operated for the U.S. Department of Energy (DOE) Office of Science by Los Alamos National Laboratory (Contract DE-AC52-06NA25396) and Sandia National Laboratories (Contract DE-NA-0003525).

I would like to acknowledge the assistance of The University of Tennessee at Chattanooga. Research reported in this publication was partially supported by the 2017 Center of Excellence for Applied Computational Science competition. This work was also partially supported by the Tennessee Board of Architectural and Engineering Examiners (TBAEE) Laboratory Equipment Grant for Chemical Engineering Award and Center for Integrated Nanotechnologies (CINT) User Proposal Award, #2016BU0031.

Finally, I would like to express my gratitude to my parents and to my friends for giving me endless support and encouragement throughout my study. Thank you.
# TABLE OF CONTENTS

ABSTRACT ........................................................................................................................................ iii

ACKNOWLEDGEMENTS ................................................................................................................ iv

LIST OF TABLES .......................................................................................................................... vii

LIST OF FIGURES ........................................................................................................................ viii

CHAPTER

1. INTRODUCTION .............................................................................................................................. 1

1.1 Purpose of Study ........................................................................................................................ 4

2. EXPERIMENTAL .......................................................................................................................... 6

2.1 Materials .................................................................................................................................. 6

2.2 Synthesis of Iron Oxide Nanoparticles .................................................................................... 6

2.2.1 Synthesis of Hybrid Pt – Iron Oxide Nanoparticles ............................................................... 7

2.2.2 Measurements of the Root Growth in Seeds Soaked in Nanoparticles Growth Solutions ................................................................................................................................. 8

2.2.3 Characterization of the Nanoparticles and Roots ................................................................. 9

2.2.4 Seed Plantation in the Soil .................................................................................................. 9

3. RESULTS AND DISCUSSION ..................................................................................................... 11

3.1 Dynamic Light Scattering (DLS) Characterization of the Nanoparticles ........................... 13

3.2 Chickpea Seed Plantation and Shoot Growth Measurement ............................................. 33

4. CONCLUSION AND FUTURE WORK ...................................................................................... 40

REFERENCES ................................................................................................................................... 42
APPENDIX

PICTURES OF ROOT GROWTH OF DIFFERENT TYPES OF SEEDS AND ITS MATERIAL CHARACTERIZATION AFTER SOAKING IN THE CONTROL, HIGH, AND LOW CONCENTRATION OF IRON OXIDE NANOPARTICLES .................................. 45

VITA ........................................................................................................................................ 49
LIST OF TABLES

3.1 Dynamic Light Scattering Data for Iron Oxide - PVP – 0.7, PEI – 0.......................... 14
3.2 Dynamic Light Scattering Data for Pt-coated Hybrid Iron Oxide - PVP – 0.7, PEI - 0.3 ...... 16
3.3 Quantity of the Material Used to Plant Chickpea Seeds in the Pot .................................. 34
3.4 Data Collected for Each Day by Observing Shoot Length of the Chickpeas .................... 35
LIST OF FIGURES

3.1 Schematic overview ........................................................................................................ 12
3.2 TEM images of NPs: (a) iron oxide NPs and (b) hybrid Pt-iron oxide NPs ................. 13
3.3 Hydrodynamic diameter for iron oxide NPs .................................................................. 15
3.4 Hydrodynamic diameter for Pt-coated hybrid iron oxide .............................................. 17
3.5 Images showing seedling growth of chickpeas in two NP solutions: (a) iron oxide and (b) hybrid Pt-iron oxide. .................................................................................................................. 19
3.6 Time-dependent NP-seed interaction plots: (a) iron oxide NP-chick pea; (b) hybrid NP-chick pea, and iron oxide NP-different seeds; (c) iron oxide NPs; and (d) hybrid NPs; 95% confidence interval used ........................................................................................................ 21
3.7 Demonstrates the impact of hybrid Pt-iron oxide NPs on the growth of different seeds .... 23
3.8 Graphs showing growth rate of different embryonic roots in hybrid Pt-iron oxide NPs: (a) green pea, (b) green gram, (c) black bean, and (d) red bean ................................................................. 24
3.9 pH dependent NP-seed interaction plots for chick pea: (a) iron oxide NPs, pH 5.5; (b) iron oxide NPs, pH 8; (c) hybrid NPs, pH 5.5; and (d) hybrid NPs, pH 8 ................................................. 26
3.10 SEM and EDX images of sections of green gram roots grown in different solutions: (a) Control, DI water; (b) iron oxide NPs, low concentration (5.54x10^-3 mgL^-1 Fe), insert, section showing root hair; (c) iron oxide NPs, high concentration (27.7 mgL^-1 Fe), insert, section showing morphological changes; (d) high concentration of Pt-iron oxide NPs; (e) EDX of sample C; and (f) EDX of sample D .................................................................................................................. 28
3.11 Scanning TEM images of embryonic green gram roots, grown in different concentration of iron oxide NPs: (a) low and (b) high .................................................................................. 29
3.12 FT-IR plots for embryonic roots of green gram grown in different concentrations of iron oxide NPs: (a) low and (b) high .................................................................................. 31
3.13 Microscopic images of black beans: (a) DI water, (b) low concentration, and (c) high concentration; green gram seeds (d) DI water, (e) low concentration, and (f) high concentration ............................................................................................. 32
3.14 Chickpea shoot length of high concentration for 25 days ............................................. 36
3.15 Chickpea shoot length of low concentration for 25 days........................................37
3.16 Chickpea shoot length of both high and low concentration for 25 days..................38
3.17 Picture of chickpea shoot growth for low and high concentrations, grown in sunlight......39
CHAPTER

1. INTRODUCTION

The growing worldwide population and increasing utilization of bioenergy crops require an estimated 70% expansion in global farming generation by 2050, as per the Food and Agricultural organizations of the United Nations (FOA) ("How to feed the world in 2050," 2009). To complete this objective with current rustic workforce, new and reasonable procedure should be created, for example, high productivity fertilizers. Customary preparation of supplements are consumed at less effectiveness by plants. In correlation, nanoparticles (NPs) can bring about improved uptake or enhanced transport within the plants due its size and high surface-to-volume ratio. The United States Department of Agriculture has acknowledged the significance of nanotechnology to increment agricultural production for the developing worldwide community through its Agricultural Food and Research Initiative (AFRI) (X. Li, Yang, Gao, & Zhang, 2015; Liu & Lal, 2015).

NPs or objects with no less than one dimension < 100 nm indicate fundamentally different material properties like surface chemistry, reactivity, and attraction when contrasted with the bulk because of their high surface zone (EU-Commission, 2011). However, specific NPs like airborne particles from volcanic ejections have been a piece of the ecosystem, huge advances have now been made in the blend of man-made designed nanostructures or engineered nanoparticles (ENPs). ENPs are outlined with focused properties for particular applications. The worldwide creation and expanding
utilization of ENPs in consumer items, for example, materials, individual care things such as, sunscreens, clothing, cosmetics, sporting equipment antimicrobial operators, and nanotherapeutics is anticipated to reach over a large portion of a million tons by 2020 (EU-Commission, 2011; Maurer-Jones, Gunsolus, Murphy, & Haynes, 2013; Vance et al., 2015a).

Specifically, ENPs can be profoundly encouraging possibility for upcoming newly forming fertilizers (Kottegoda et al., 2017; Rui et al., 2016). Furthermore, ENPs have been observed to enter tomato plant roots or seed tissues without causing any adverse effect. Intake and transportation of iron oxide NPs were also noticed in watermelon plants (J. Li et al., 2013; Shankramma, Yallappa, Shivanna, & Manjanna, 2016). Iron is one of the fundamental components for plant growth and assumes an essential part in the photosynthesis process in plants. More recently, iron oxide NPs were utilized as a part of a substitute for costly Fe-fertilizers to effectively alleviate iron insufficiency in peanut plants (Rui et al., 2016).

As nanostructure could now be manufactured in different shapes, sizes combined structures, core-shell structures and surface functionalization, the most dynamic NP structures for enhanced plant growth root absorption is still to be known (J. Li et al., 2016; Ren et al., 2011; Vance et al., 2015b). Most studies have focused on toxicity analysis of NPs, but limited literature is available about the best suitable NP formulation or NP concentration to induce increased agricultural production. (Burklew, Ashlock, Winfrey, & Zhang, 2012; Rico, Majumdar, Duarte-Gardea, Peralta-Videa, & Gardea-Torresdey, 2011; Yin, Colman, McGill, Wright, & Bernhardt, 2012). Further, limited reports are accessible on the interaction of plants with iron-oxide-based NPs (Feng et al., 2013; Siddiqi & Husen, 2017). Therefore, we will specifically explore the huge potential of iron oxide NPs as fertilizers through systematic experiments in this study. It was shown in one study that
iron-oxide NPs did not cause any toxicological impacts in pumpkin plants over continued appearance period (Zhu, Han, Xiao, & Jin, 2008). Also, an examination of green gram seeds demonstrated that the 10 mgL⁻¹ iron oxide NPs encouraged an expanded physiological effect (Ren et al., 2011). Consequently, iron oxide NPs exhibit an ideal source for iron conveyance to plants inside fertilizers (Ma et al., 2010).

Soil property like pH is a vital parameter in building up of these new fertilizers since it to a great extent impacts the accessibility of nutrients (Fageria & Zimmermann, 1998). NPs experience a noteworthy change in morphology, surface structure, surface functionality and agglomeration state after collaboration with normal, natural organic matter (NOM) or nutrient in the ecosystem, independent of the type of NP (Hitchman, Smith, Ju-Nam, Sterling, & Lead, 2013; Orts-Gil, Natte, & Österle, 2013; Tejamaya, Römer, Merrifield, & Lead, 2012). Generally, these mineral supplements and biomolecules dislodge weaker restricting surface ligands on the NP surface to frame hybrid NPs; particular from the first combined condition of the ENP. However, most examinations on plants have detailed the interaction of as-synthesized iron oxide NPs without considering the impact of molecule change or pH. In this study, hybrid Pt-attached iron oxide NPs and development arrangements at various pH were created and examined to address this issue. No data is as of now accessible in the record of comparative work amongst iron-oxide and hybrid Pt-Fe₂O₃ nanostructures on plant growth (Elmer & White, 2016; Orts-Gil et al., 2013).

In this manner, the point of this work is to research the impact of iron oxide NPs and hybrid Pt-iron oxide NPs particularly on the development of embryonic roots and shoot. Eatable seeds of various sizes (e.g., chickpeas, green peas, green gram, black beans, and red beans) were selected as analyzing samples in this study (Figure 1 shown in result and discussion). These seeds were chosen as representative samples due to their high worldwide consumption. Fifteen distinctive
growth solutions were tried over a time of six days for each of the five different seeds with change in NP type (functionalized iron oxide vs Pt attached iron oxide), NP concentration (0.00554 mgL$^{-1}$ and 27.7 mgL$^{-1}$), and solution pH data (5.5, 7, and 8) when contrasted with solutions without NPs. Statistical analysis of plant growth in NP soaking solutions combined with material characterization of plant samples utilizing electron microscopy, optical microscopy, and Fourier-transform infrared spectroscopy (FTIR) were conducted to understand NP-plant interactions in this study. These examinations will be useful in building up another class of iron deficiency fertilizers used for simple absorption of the nutrient and diminished utilization of the fertilizer for the safety of the ecosystem. The benefit of the NP fertilizers was two-fold. First, they were consumed more successfully by the plants because of their tiny size, in this manner limiting the general amount of fertilizers combined to the soil for a designed increment in plant growth. Second, soaking the different seeds in these fertilizers was adequate to upgrade plant yield, a prominent change over usual fertilizers that are included in expensive amounts to the soil.

1.1 Purpose of Study

The purpose of the study is to explore the chemical, physical and morphological changes in the different types of seeds. The homemade engineered NPs by using the synthesis by modified Polyol method with the help of reagents iron (III) acetylacetonate (Fe(acac)$_3$), polyvinylpyrrolidone (PVP, Mw 10 kDa, TCI, Fisher), polyethyleneimine (PEI, Mw 60 kDa, 50% aq, Alfa Aesar), triethyleneglycol (C$_6$H$_{14}$O$_4$, TREG, 99%, Acros), de-ionized water (DI, Fisher) for the iron oxide NPs and hexachloroplatinic acid (H$_2$PtCl$_6$, 10%, 3.8% Pt, EMD Millipore), sodium hydroxide (NaOH, 97%, Fisher), and hydrochloric acid (HCl, 35%, Fisher) for the hybrid NPs. These two types of NPs were used on five types of seeds, which are seeds of green pea (Pisum sativum L.),
chick pea (*Cicer arietinum*), green gram or mung bean (*Vigna radiate*), black and red beans (*Phaseolus vulgaris*). We examined how these engineered NPs will affect the seeds by soaking them in different concentrations of iron oxide NPs and hybrid NPs in addition to the control deionized water. To investigate the changes in the seeds, we measured the length of the root for the six days of the interval. We also monitored the growth of these plants in potted soil for a period of 25 days. The results from our root growth considered over a 6-day time frame were depicted in definite statistical plots for both iron oxide and hybrid Pt iron oxide NPs for every one of the five seed types. The statistical outcomes showed iron oxide NPs added to better growth solutions for plants in contrast to the hybrid Pt-decorated iron oxide at the low concentration. This outcome showed the preparatory potential for iron oxide NPs as Fe deficiency fertilizers at low concentration.
CHAPTER 2
EXPERIMENTAL

2.1 Materials

All chemicals were utilized as acquired, including iron (III) acetylacetonate (Fe(acac)_3, 99%, Alfa Aesar), polyethyleneimine (PEI, Mw 60 kDa, 50% aq, Alfa Aesar), polyvinylpyrrolidone (PVP, Mw 10 kDa, TCI, Fisher), triethyleneglycol (C_6H_{14}O_4, TREG, 99%, Acros), hexachloroplatinic acid (H_2PtCl_6, 10%, 3.8% Pt, EMD Millipore), sodium hydroxide (NaOH, 97%, Fisher), hydrochloric acid (HCl, 35%, Fisher), and de-ionized water (DI, Fisher). Seeds of green pea (Pisum sativum L.), chick pea (Cicer arientinum), green gram or mung bean (Vigna radiate), and dark and red beans (Phaseolus vulgaris) were bought from nearby grocery shops in Chattanooga, TN, USA.

2.2 Synthesis of Iron Oxide Nanoparticles

Synthesis of Iron oxide NPs were carried out by using the "modified" polyol method (Palchoudhury & Lead, 2014). In a standard synthesis, compositions of capping compounds such as a capping compound of polyethylenimine (PEI, 0.3 g), polyvinyl pyrrolidone (PVP, 0.7 g) were heated at 90 °C for 10 min with the addition of solvent, triethylene glycol (TREG, 10 mL). In addition, two mmol of the iron precursor, iron (III) acetylacetonate (Fe(acac)_3, 0.7 g), was mixed
with this solution and blended for 10 minutes. TREG assisted as lessening specialist in the chemical reaction.

The reactant arrangement was then thermally disintegrated at 260 °C for 1 h to form ultimate iron oxide NP item. This complete synthesis was carried out in air and absence of inert gas protection.

The NPs were further cleaned via centrifugation (high-speed minicentrifuge, Fisher Scientific Sovrvail Legend Micro 21 at room temperature) for 15 min at 14000 rpm to remove any remnant as the supernatant. Finally, the iron oxide NPs were dispersed in required quantity of DI water and sonicated for 15 min (Branson 1800, room temperature). To obtain the end product and desired concentrations of $5.54 \times 10^{-3} \text{mgL}^{-1}$ Fe (low concentration) and $27.7 \text{mgL}^{-1}$ Fe (high concentration). The concentration for iron oxide NPs in this experiment were affirmed by utilizing two strategies; sample test of weight and calibration plots acquired from ultra-violet visible spectroscopy (UV-Vis). These iron oxide NPs served as starting agents for synthesizing the hybrid Pt-coated iron oxide NPs. The NPs were also utilized in consequent NP-seed interaction experiments.

2.2.1 Synthesis of Hybrid Pt – Iron Oxide Nanoparticles

By definition, hybrid NPs contain two or more NP components combined into one NP assembly. Here, Pt-attached iron oxide NPs were synthesized as model hybrid NPs following a formerly reported protocol (Palchoudhury, Xu, Goodwin, & Bao, 2011). The Pt precursor ($\text{H}_2\text{PtCl}_6$) was mixed with iron oxide NPs at 10:1 Pt precursor: NP volume ratio. The solution was reduced by means of UV irradiation using hand-held UV lamp (UVL-56 Handheld UV Lamp) for 30 min to form Pt NPs on iron oxide surfaces. Similar to iron oxide NPs, the hybrid Pt-attached iron oxide
NPs were cleaned by using centrifugation, and mixed and dissolved in Di water through sonication to acquire target mass concentrations (e.g., 5.54x10^{-3} and 27.7 mgL^{-1} Fe) for NP-seed experiments.

2.2.2 Measurements of the Root Growth in Seeds Soaked in Nanoparticles Growth Solutions

The impact of iron oxide and Pt-iron oxide NPs on different seeds (e.g., green pea or Pisum sativum L., chick pea or Cicer arientinum, green gram or Vigna emanate, black beans, and red beans or Phaseolus vulgaris) was evaluated particularly in root development over a period of six days.

First, the seeds were cleaned in ethanol (70%) and DI water and dried with the help of filter paper. All vials utilized for these tests are cleaned with 70% ethanol. The seeds were then soaked in vials containing NP growth solutions. In this experiment, the seeds were soaked in three types of growth solution, namely, DI water, solution containing lower NP concentration (5.54x10^{-3} mgL^{-1} Fe), and solution containing higher NP concentration (27.7 mgL^{-1} Fe). The root length from the seeds were observed every day with the use of Vernier caliper for the period of six days. The trials were run and observed at room temperature under ambient pressure. A total of five different legume seed types (e.g., green pea, chickpea, green gram, black beans, red beans) were investigated in these root growth experiments. For each seed type, this test was repeated six times with both iron oxide and Pt-iron oxide NP growth solutions for statistical review.

The pH of soil differs relying upon the soil type and minerals available in the soil. Consequently, to check whether the NPs indicated a relative impact on embryonic roots in various soils, the root growth experiments specified above was also performed in various pH arrangements (e.g., pH 5.5, 7, and 8) for each seed type. pH of the growth solutions in these experiments were adjusted using HCL and NaOH solutions.
2.2.3 Characterization of the Nanoparticles and Roots

The size and morphology of the iron oxide and Pt-coated hybrid NPs were explored on a FEI Tecnai F-20 transmission electron microscopy (TEM). Moreover, the crystal structure of the NPs was examined on a Philips Analytical X-ray diffractometer (XRD) with a Cu source. A multi-strategy material characterization method was utilized to unequivocally predict the impact of nanofertilizers on embryonic roots. Both the morphology and chemical structure of the roots were characterized utilizing a FEI Titan chemiSTEM (Thermo Fisher) with high-accumulation-point energy dispersive X-ray spectroscopy (EDX). A JEOL 7000 FE scanning electron microscope (SEM) furnished with EDX was utilized to examine the surface of the roots and its chemical structure. The roots were also observed using optical microscopy (Fisher Scientific Micromaster Microscope). To identify the surface functional group of the roots, a Bruker Alpha Fourier transform infrared spectrometer (FT-IR) was utilized to run scans on fully dried root test over 400-4000 cm\(^{-1}\) wavelength scale. The FT-IR information was utilized to identify NP-root interaction through variations in the surface ligands on the roots.

2.2.4 Seed Plantation in the Soil

The germinated seeds which were soaked in Di water, lower concentration, and higher concentration of iron oxide NP growth solutions were planted in the soil for observation of the shoot growth rate. For this purpose, we used regular garden soil (125 g), (25 g) of Garden tone fertilizer, and (25 g) of organic fertilizers mixed together to prepare the potted soil. The germinated seeds of the same type (e.g., chickpea) soaked in the three different growth solutions of DI water, low iron oxide NP concentration, and high iron oxide NP concentration were all planted in the
same pot for better comparison. All the pots were kept under the same conditions i.e., within the room near the glass window for access to sunlight and under the room temperature (~25 °C). After 4 to 5 days, the shoot came out from the soil and its length was measured each day over a period of 25 days.
In view of our analyses of five seed types, ENPs (e.g., iron oxide) can enhance the growth rate of embryonic roots by 88-366% yet are generally unexplored for agricultural use as their interaction with the organic environment is not established. Here, we examined the impact of two new model ENPs iron oxide NPs and hybrid Pt-iron oxide NPs on root and shoot development of various seeds. A combination of various material characterization of the embryonic roots and statistical investigation of the root and shoot growth rate over a period of 6-days was used to build a protocol for the first time to understand NP-plant interaction. These two NPs were interacted with five different seeds for the first time in the study. Moreover, this is the main report contrasting the growth of seeds in the iron oxide NPs, and its hybrid counterpart. Figure 3.1 demonstrates a schematic overview of our investigation.
The PVP/PEI-coated iron oxide NPs synthesized by a modified polyol technique were 16 nm in size; recommended by the TEM picture (Figure 3.2a). The Pt attached iron oxide NPs were produced using a formerly published protocol, however utilizing these new iron oxide seeds. Since Pt has a higher atomic number when compared with Fe, the Pt, NPs of size 2 nm, show up as darker spots in the TEM image (Figure 3.2b). The powdered XRD analysis confirmed a hematite (α-Fe₂O₃) crystal structure for the iron oxide NPs.
3.1 Dynamic Light Scattering (DLS) Characterization of the Nanoparticles

DLS is most typically utilized to examine NPs. Different Particle size can be measured by determining the irregular variations in the intensity of light dispersed from a suspension or chemical solution. This system is generally known as dynamic light scattering (DLS) but is called photon correlation spectroscopy (PCS) and quasi-elastic light scattering (QELS). For this work, we utilized DLS to determine the different sizes of the engineered iron oxide and Pt-coated hybrid iron oxide NPs (capping molecule: PVP 0.7g/PEI 0.3 g). Table 3.1, Table 3.2, Figure 3.3, and Figure 3.4 indicated particle size measurement for particle diameter with three different quantities: relative intensity weighted, volume weighted, and number weighted respectively. The following details were covered for the two NP samples.

(1) Nanoparticles measurement name - Sample 1 - Iron Oxide - PVP - 0.7, PEI - 0.3
    Measurement mode - Particle size
Table 3.1

Dynamic Light Scattering Data for Iron Oxide - PVP – 0.7, PEI – 0

<table>
<thead>
<tr>
<th>Particle diameter</th>
<th>Relative frequency</th>
<th>Intensity weighted</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.209</td>
<td>0.000</td>
<td></td>
</tr>
<tr>
<td>0.227</td>
<td>0.000</td>
<td></td>
</tr>
<tr>
<td>0.246</td>
<td>0.000</td>
<td></td>
</tr>
<tr>
<td>0.267</td>
<td>0.000</td>
<td></td>
</tr>
<tr>
<td>0.290</td>
<td>0.000</td>
<td></td>
</tr>
<tr>
<td>27.020</td>
<td>0.448</td>
<td></td>
</tr>
<tr>
<td>29.299</td>
<td>0.970</td>
<td></td>
</tr>
<tr>
<td>31.771</td>
<td>1.522</td>
<td></td>
</tr>
<tr>
<td>34.452</td>
<td>2.091</td>
<td></td>
</tr>
<tr>
<td>37.358</td>
<td>2.664</td>
<td></td>
</tr>
<tr>
<td>40.510</td>
<td>3.227</td>
<td></td>
</tr>
<tr>
<td>43.928</td>
<td>3.768</td>
<td></td>
</tr>
<tr>
<td>47.634</td>
<td>4.274</td>
<td></td>
</tr>
<tr>
<td>51.652</td>
<td>4.734</td>
<td></td>
</tr>
<tr>
<td>56.010</td>
<td>5.137</td>
<td></td>
</tr>
<tr>
<td>60.735</td>
<td>5.474</td>
<td></td>
</tr>
<tr>
<td>65.859</td>
<td>5.738</td>
<td></td>
</tr>
<tr>
<td>71.416</td>
<td>5.924</td>
<td></td>
</tr>
<tr>
<td>77.441</td>
<td>6.027</td>
<td></td>
</tr>
<tr>
<td>83.974</td>
<td>6.045</td>
<td></td>
</tr>
<tr>
<td>91.059</td>
<td>5.975</td>
<td></td>
</tr>
<tr>
<td>98.741</td>
<td>5.817</td>
<td></td>
</tr>
<tr>
<td>107.071</td>
<td>5.570</td>
<td></td>
</tr>
<tr>
<td>116.105</td>
<td>5.233</td>
<td></td>
</tr>
<tr>
<td>125.900</td>
<td>4.806</td>
<td></td>
</tr>
<tr>
<td>136.521</td>
<td>4.286</td>
<td></td>
</tr>
<tr>
<td>148.039</td>
<td>3.673</td>
<td></td>
</tr>
<tr>
<td>160.529</td>
<td>2.964</td>
<td></td>
</tr>
<tr>
<td>174.072</td>
<td>2.156</td>
<td></td>
</tr>
<tr>
<td>188.758</td>
<td>1.247</td>
<td></td>
</tr>
<tr>
<td>204.682</td>
<td>0.231</td>
<td></td>
</tr>
</tbody>
</table>
Figure 3.3

Hydrodynamic diameter for iron oxide NPs

(2) Nanoparticle measurement name - Sample 2- Pt Iron Oxide - PVP – 0.7, PEI - 0.3
Measurement mode - Particle size

Hydrodynamic Diameter: 83.97 nm (±2%)
Table 3.2

Dynamic Light Scattering Data for Pt-coated Hybrid Iron Oxide - PVP – 0.7, PEI - 0.3

<table>
<thead>
<tr>
<th>Particle diameter</th>
<th>Relative frequency Intensity weighted</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.209</td>
<td>0.000</td>
</tr>
<tr>
<td>0.227</td>
<td>0.000</td>
</tr>
<tr>
<td>0.246</td>
<td>0.000</td>
</tr>
<tr>
<td>0.267</td>
<td>0.000</td>
</tr>
<tr>
<td>0.290</td>
<td>0.000</td>
</tr>
<tr>
<td>56.010</td>
<td>0.943</td>
</tr>
<tr>
<td>60.735</td>
<td>2.156</td>
</tr>
<tr>
<td>65.859</td>
<td>3.365</td>
</tr>
<tr>
<td>71.416</td>
<td>4.533</td>
</tr>
<tr>
<td>77.441</td>
<td>5.623</td>
</tr>
<tr>
<td>83.974</td>
<td>6.598</td>
</tr>
<tr>
<td>91.059</td>
<td>7.423</td>
</tr>
<tr>
<td>98.741</td>
<td>8.065</td>
</tr>
<tr>
<td>107.071</td>
<td>8.497</td>
</tr>
<tr>
<td>116.105</td>
<td>8.693</td>
</tr>
<tr>
<td>125.900</td>
<td>8.633</td>
</tr>
<tr>
<td>136.521</td>
<td>8.304</td>
</tr>
<tr>
<td>148.039</td>
<td>7.695</td>
</tr>
<tr>
<td>160.529</td>
<td>6.801</td>
</tr>
<tr>
<td>174.072</td>
<td>5.623</td>
</tr>
<tr>
<td>188.758</td>
<td>4.165</td>
</tr>
<tr>
<td>204.682</td>
<td>2.436</td>
</tr>
<tr>
<td>221.950</td>
<td>0.448</td>
</tr>
</tbody>
</table>
Hydrodynamic diameter for Pt-coated hybrid iron oxide

Figure 3.4

Hydrodynamic diameter: 116.02 (±2%)
These two ENPs, iron oxide and hybrid Pt iron oxide, were interacted with five distinct kind of legume seeds over a time of six days by absorbing the seeds NP solutions for examine their impact on embryonic root growth. Each seed type selected for our investigation (e.g., green pea, green gram, chickpea, dark beans, and red beans) was of various size. For both iron oxide and hybrid Pt-iron oxide NPs, contro DI water (0 mgL⁻¹ Fe), a low (5.54x10⁻³ mgL⁻¹ Fe) and a high (27.7 mgL⁻¹ Fe) solution concentration were prepared as seed growth solutions. Moreover, all experiments related to the particular kind of seed, NP, and solution concentration was repeated six times for statistical reliability. Both iron oxide and hybrid NPs demonstrated an impact different from the control DI water on the growth of embryonic roots.

An enormous increment in root length was observed for the different seeds in iron oxide NP solutions when compared with the hybrid Pt-decorated iron oxide NPs. The most outstanding improvement of root growth was found in green gram and chickpeas seeds; because of their necessity for iron. Figure 3.5a-b demonstrate the growth of embryonic roots in green gram seeds-soaked iron oxide and hybrid Pt-decorated iron oxide NP solutions, respectively. The low concentration iron oxide NP growth solution supported the most noteworthy increment in embryonic root growth in green gram, as observed in Figure 3.5a. This was additionally seen in different seeds. Therefore, the low concentration iron oxide NP growth solution showed higher root growth of 88-366% for the different seed types when compared with the control DI water growth solutions.
Results from our root growth experiments over six days were represented in statistical plots for both iron oxide and hybrid Pt iron oxide. The measurable outcome passed on to findings; iron oxide NPs showed better root growth when differentiated with the hybrid Pt-decorated iron oxide NPs, and the low concentration iron oxide growth solution was best appropriate soak for extended root growth. These outcomes exhibited the preparatory potential for iron oxide NPs as Fe...
deficiency fertilizers at low concentration. Figure 3.6a-b indicated time-dependent plots of the average root length of chickpea seeds in iron oxide and hybrid NPs. The control, low, and high NP concentrations are set apart as water, low NP, and high NP, independently, in the plots for both iron oxide and hybrid NPs (Figure 3.6). At low concentration ~ 5.54x10⁻³ mgL⁻¹ Fe, iron oxide NPs fundamentally upgraded root growth, as recommended from the measured seedling length over than six days. However, a reduced growth was seen at high NP concentration (~ 27.7 mgL⁻¹ Fe), (Figure 3.6a). Since iron oxide NP growth solution performed superior to the Pt decorated iron oxide NPs, we explored the comparative impact of iron oxide NPs at both low and both concentration of the five distinct legumes in a statistical plot. Figure 3.6c-d demonstrated the bar plot for average root length following six days for each seed type under various iron oxide NP growth solutions. When we analyzed the average root length over six days for chickpeas for various treatments, we noticed that there was a notable increment in growth of the root length were utilizing low NP in compared to high NP and water.
Time-dependent NP-seed interaction plots:
(a) iron oxide NP-chick pea; (b) hybrid NP-chick pea, and iron oxide NP-different seeds;
(c) iron oxide NPs; and (d) hybrid NPs; 95% confidence interval used.

Figure 3.6
Specifically, the root length of chickpea seeds in low iron oxide NPs extended by 88% and 184%, individually when differentiated with the chickpea seeds in water and high iron oxide NP growth solutions. Moreover, we noted that low iron oxide NP treatment intensified the growth of green pea and green gram by 160% and 366%, respectively, (Figure 3.6c-d). The impact of NPs relied upon the size and type of seeds in addition to the concentration and morphology of the NPs, on basis of these plots in Figure 3.6. Among the seeds analyzed, green gram seedlings soaked in low concentration iron oxide NP growth solution demonstrated the most increment in growth rate by ~366%, when compared with controlled DI water growth solutions (Figure 3.5 and Figure 3.6c-d). In contrast with chickpea, green pea, and green gram, the growth of black and red bean seedlings were not affected by iron oxide NPs (Figure 3.6c-d). This was acceptable because the beans are richer in iron substance as compared to the other legume seeds. The beans were less influenced by iron-deficiency fertilizers such as iron oxide NPs as the average essential iron concentration in beans (55 µgg⁻¹) is high in contrast with alternate crops (Petry, Boy, Wirth, & Hurrell, 2015).

Effects of iron oxide NPs and hybrid iron oxide NPs on different types of seeds demostrated in Figure 3.7 and Figure 3.8 respectively. Low concentration of NPs were showed significant morphological change on green pea and green gram seeds. For black bean and red bean beacause of higher iron content in the seed itself, the effect of iron oxide NPs were not much higher comapared to the green pea and green gram seeds. On the other hand, adverse effect of hybrid Pt- iron oxide NPs were observed on different types of seeds and its growth rate.
Figure 3.7

Demonstrates the impact of hybrid Pt-iron oxide NPs on the growth of different seeds
Graphs showing growth rate of different embryonic roots in hybrid Pt-iron oxide NPs: (a) green pea, (b) green gram, (c) black bean, and (d) red bean.

We observed the seedling growth in NP solutions at various pH 5.5 and 8 to understand if these trends in NP fertilizers-plant interaction were pH-dependent (Figure 3.9). For the iron oxide NPs, seedlings indicated same reaction trend at all pH, however, a slower growth rate of embryonic roots was observed at pH 5.5 or 8, when compared with neutral pH (Figure 3.9a-b). On the other
hand, the NP concentration growth trend of seedlings in hybrid NPs were more susceptible to pH changes (Figure 3.9c-d). However, embryonic roots were found in all examples, showing no severe toxic impact of these NPs at concentrations <27.7 mgL\(^{-1}\) Fe.
The root surfaces were examined by SEM and EDX following a 6-day growth period to research any variations in physiology and chemical composition after interacting with the NPs (Figure 3.10). Figure 3.10a demonstrates the SEM image of embryonic green gram roots growth in DI water after Au sputtering. Complete epidermis is seen in this control test root. The epidermis is
undamaged in roots developed in a low concentration of iron oxide NPs, as recommended by the SEM image (Figure 3.10b). Moreover, these roots have noticeable root hairs to encourage expanded absorption of supplement as shown by Figure 3.10b, insert. Conversely, discernable change in morphology of the epidermis is observed in embryonic root grown in higher concentration of iron oxide NPs, recommending conceivable adverse effects at these high NP concentrations (Figure 3.10c and 3.10e; Wang, Chen, Chen, & Ma, 2012). Roots developed in Pt iron oxide hybrid NPs indicated NP aggregates on the surface, which probably induced the reduced growth (Figure 3.10d and 3.10f).
SEM and EDX images of sections of green gram roots grown in different solutions:
(a) Control, DI water; (b) iron oxide NPs, low concentration (5.54×10^{-3} mgL^{-1} Fe), insert, section showing root hair; (c) iron oxide NPs, high concentration (27.7 mgL^{-1} Fe), insert, section showing morphological changes; (d) high concentration of Pt-iron oxide NPs; (e) EDX of sample C; and (f) EDX of sample D

In the P, K, Ca, O and S originate from the lipids in roots while C and Al is from the TEM holder and tape. However, in roots developed in low concentration iron oxide NPs, the Fe is possibly taken up or assimilated by the roots (Shankramma et al., 2016). Interestingly, Fe and O were recognized in roots grown in high concentration hybrid NPs, however, Pt was not seen at the surface. This could be attributed to the smaller sized Pt-NPs being consumed through porous in the plant walls, inducing the adverse effect on root growth (Wang et al., 2012).

Iron oxide NPs, especially at the low concentration, prompted fertilizers-like effects on embryonic root growth, on the basis of our understanding on various seedlings. So, green gram sample root developed in the low and high concentration of iron oxide NPs were investigated by means of
TEM to understand the uptake of these NPs (Figure 3.11). No individual iron oxide NP detected in roots grown with low concentration iron oxide NP growth solutions. Some NP aggregates were likely detected. On the other hand, for the roots grown in high concentration iron oxide NPs, lot of individual iron oxide NPs were likely detected within the roots. This could explain the difference in root growth under the two different concentrations. The TEM characterization of the roots gives an initial understanding on the uptake of iron oxide NPs. Complete characterization of NP transport and uptake is as of now being explored by our group through elemental chemical analysis and hyper spectral imaging analysis.

![Scanning TEM images of embryonic green gram roots, grown in different concentration of iron oxide NPs: (a) low and (b) high](image)

**Figure 3.11**

Scanning TEM images of embryonic green gram roots, grown in different concentration of iron oxide NPs: (a) low and (b) high

To additionally examine the improvements in chemical composition of embryonic roots interacted with NPs, the surface functional groups of well dried root samples were examined utilizing FT-IR spectroscopy. The FT-IR range of roots developed in low concentration of iron oxide NPs presented the characteristic N-H extending of protonated fundamental amine at 3254 cm$^{-1}$, similar
to PEI-covered iron oxide NPs, demonstrating either binding or uptake of NPs by the roots. The peak at 1773 cm\(^{-1}\), attributed to carbonyl stretch of lipids shown in the root analyses was not noticed in powdered iron oxide NP samples. The groups between 1558-1507 cm\(^{-1}\) could be allocated to N-H vibrations in the root contrasted with the range of roots in low NP concentration. The N-H band appeared to 3274 cm\(^{-1}\) for roots developed in high NP concentration, demonstrating distinctive hydrogen bonding. This distinction in chemical composition could clarify the lower growth rate of roots examined in high NP concentrations. It is likely that the high concentrations of amine-covered NPs for this situation encouraged strong hydrogen bonding with the peptide groups in the roots.
Figure 3.12

FT-IR plots for embryonic roots of green gram grown in different concentrations of iron oxide NPs: (a) low and (b) high

After germinating the seed roots in the different concentrations of the NPs roots of the black beans and green gram seeds investigated under the Micromaster Microscope at the University of Tennessee at Chattanooga chemical engineering laboratory. Figure 3.13 shows the two different types of legume seeds, black beans, and green gram soaked in the DI water, low and high
concentration of NPs. Thus, observing the difference between DI, low and high concentration, we can clearly see the low and high concentration NPs deposited on the root of the seed.

![Microscopic images of black beans](image)

**Figure 3.13**

Microscopic images of black beans: (a) DI water, (b) low concentration, and (c) high concentration; green gram seeds (d) DI water, (e) low concentration, and (f) high concentration

The combined approach of estimating the embryonic root length and multi-strategy material portrayal of the roots by means of SEM, TEM, microscopy, and FT-IR can be utilized as a robust strategy to study new nanofertilizers. The outcomes from these diverse characterization methods gave dependable and important insights into the notable effect of iron oxide and Pt-iron oxide NPs on seedling growth in this investigation. Seedling growth in hybrid NPs indicates patterns, particularly unique in relation to those in the iron oxide NPs, it indicated a notable effect of the
hybrid morphology on embryonic roots. Moreover, the two NPs actuate a slower development rate in seedlings at the high concentration (~ 27.7 mgL\(^{-1}\) Fe). Iron oxide NPs are known to be naturally nontoxic, however, an important finding of our investigations is the noteworthy improvement in the development of embryonic roots by the iron oxide NPs at the lower concentration range (~ 5.54x10\(^{-3}\) mgL\(^{-1}\) Fe). Since the expansion in root development with iron oxide, NPs were observed for some seed types, and dependably predicts the immense capability of iron oxide NPs as nanofertilizers. This coordinated, measurable, and material portrayal approach shows in as a lab-scale evidence for the utilization of iron oxide NPs in agricultural applications. The point-by-point system of NP take-up and distribution inside the plant will be fundamental in growing new iron oxide NP-based fertilizers and is now under investigation.

3.2 Chickpea Seed Plantation and Shoot Growth Measurement

Engineered NPs used to examine the morphological changes with the different types of seeds. The following data is showing one type of the iron oxide NPs concentration with the chickpeas seeds and quantity of the material used to plant chickpeas seeds in soil pot.

Nanoparticles concentration:

- PVP - 0.7 gm, PEI - 0.3 gm, TEG - 10ml, Fe(acac)\(_3\) - 0.7 gm (Pure NPs)
- DI water - 4ml
- High concentration NPs - 1ml (1ml of Pure NPs added in 10ml of DI water.)
- Low concentration NPs – 0.1ml (0.01ml of pure NPs added in 5ml of DI water)
Table 3.3

Quantity of the Material Used to Plant Chickpea Seeds in the Pot

<table>
<thead>
<tr>
<th>No.</th>
<th>Material</th>
<th>Weight/quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Garden soil</td>
<td>125 gm</td>
</tr>
<tr>
<td>2.</td>
<td>Fertilizer</td>
<td>25 gm</td>
</tr>
<tr>
<td>3.</td>
<td>Garden tone organic fertilizer</td>
<td>25 gm</td>
</tr>
<tr>
<td>4.</td>
<td>Magic grow</td>
<td>2 drops Twice/week</td>
</tr>
<tr>
<td>5.</td>
<td>DI/Tap water</td>
<td>10-15ml/day</td>
</tr>
</tbody>
</table>
Table 3.4

Data Collected for Each Day by Observing Shoot Length on the Chickpeas

<table>
<thead>
<tr>
<th>Day</th>
<th>Date</th>
<th>High Conc. Length of the shoot in (mm)</th>
<th>Low Conc. Length of the shoot in (mm)</th>
<th>Water added per day (ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>11/02/2017</td>
<td>7</td>
<td>9</td>
<td>5-10</td>
</tr>
<tr>
<td>2.</td>
<td>11/03/2017</td>
<td>7.5</td>
<td>10</td>
<td>5-10</td>
</tr>
<tr>
<td>3.</td>
<td>11/04/2017</td>
<td>10.5</td>
<td>14</td>
<td>5-10</td>
</tr>
<tr>
<td>4.</td>
<td>11/05/2017</td>
<td>12.5</td>
<td>17.5</td>
<td>5-10</td>
</tr>
<tr>
<td>5.</td>
<td>11/06/2017</td>
<td>32</td>
<td>40</td>
<td>5-10</td>
</tr>
<tr>
<td>6.</td>
<td>11/07/2017</td>
<td>44</td>
<td>72</td>
<td>5-10</td>
</tr>
<tr>
<td>7.</td>
<td>11/08/2017</td>
<td>58</td>
<td>88</td>
<td>5-10</td>
</tr>
<tr>
<td>8.</td>
<td>11/09/2017</td>
<td>63.5</td>
<td>95.25</td>
<td>5-10</td>
</tr>
<tr>
<td>9.</td>
<td>11/10/2017</td>
<td>69</td>
<td>99.75</td>
<td>5-10</td>
</tr>
<tr>
<td>10.</td>
<td>11/11/2017</td>
<td>76</td>
<td>103</td>
<td>5-10</td>
</tr>
<tr>
<td>11.</td>
<td>11/12/2017</td>
<td>81</td>
<td>111</td>
<td>5-10</td>
</tr>
<tr>
<td>12.</td>
<td>11/13/2017</td>
<td>88.5</td>
<td>119.25</td>
<td>10-15</td>
</tr>
<tr>
<td>13.</td>
<td>11/14/2017</td>
<td>92.71</td>
<td>130</td>
<td>10-15</td>
</tr>
<tr>
<td>14.</td>
<td>11/15/2017</td>
<td>95.25</td>
<td>139</td>
<td>10-15</td>
</tr>
<tr>
<td>15.</td>
<td>11/16/2017</td>
<td>101.6</td>
<td>148.75</td>
<td>10-15</td>
</tr>
<tr>
<td>16.</td>
<td>11/17/2017</td>
<td>105.41</td>
<td>152.4</td>
<td>10-15</td>
</tr>
<tr>
<td>17.</td>
<td>11/18/2017</td>
<td>113.5</td>
<td>165</td>
<td>10-15</td>
</tr>
<tr>
<td>18.</td>
<td>11/19/2017</td>
<td>118</td>
<td>171.4</td>
<td>10-15</td>
</tr>
<tr>
<td>19.</td>
<td>11/20/2017</td>
<td>124.75</td>
<td>184.15</td>
<td>10-15</td>
</tr>
<tr>
<td>20.</td>
<td>11/21/2017</td>
<td>129</td>
<td>186</td>
<td>10-15</td>
</tr>
<tr>
<td>21.</td>
<td>11/22/2017</td>
<td>134.25</td>
<td>195</td>
<td>10-15</td>
</tr>
<tr>
<td>22.</td>
<td>11/23/2017</td>
<td>138</td>
<td>202.75</td>
<td>10-15</td>
</tr>
<tr>
<td>23.</td>
<td>11/24/2017</td>
<td>146</td>
<td>209.25</td>
<td>10-15</td>
</tr>
<tr>
<td>24.</td>
<td>11/25/2017</td>
<td>151.75</td>
<td>214</td>
<td>10-15</td>
</tr>
<tr>
<td>25.</td>
<td>11/26/2017</td>
<td>156</td>
<td>219.75</td>
<td>10-15</td>
</tr>
</tbody>
</table>

The following figures show chickpea shoot growth in the soil mixed with the organic fertilizers for high and low concentration of NPs.
Figure 3.14

Chickpeas shoot length of high concentration for 25 days

\[ R^2 = 0.9784 \]
Figure 3.15

Chickpea shoot length of low concentration for 25 days

$R^2 = 0.9748$
Chickpea shoot length of both high and low concentration for 25 days

Figure 3.16
Figure 3.17 shows low- and high-concentration chickpeas’ seed plantation and seed growth after 12 days in the presence of the sunlight.

Comparing above data from Figure 3.17, we can say that low concentration nanoparticles chickpeas seed is growing faster than the high concentration.
In conclusion, utilizing diverse material characterization, a simplistic technique was designed to survey the effect of two distinctive iron oxide-based nanostructures with respect to seedling growth. The concentration, morphology, and nanostructure of the NPs and the class of seed was observed to significantly effect root growth, considering the outcomes from five types of consumed seeds. The best development in seedlings is seen with iron oxide NPs at low concentrations (~ $5.54 \times 10^{-3} \text{mgL}^{-1} \text{Fe}$). The investigation follows in as a proof-of-the-idea for the promising capability of iron oxide NPs as Fe-rich fertilizers. Also, seeds in both iron oxide and hybrid NP arrangements demonstrate root growth, showing no toxic effect of the NPs. The electron microscopy and FT-IR examination of the roots affirmed NP uptake and gave key insight to the concentration dependent changes in NP-plant exposure. This strategy will be attractive in making new nanofertilizer materials to improve agricultural production, while keeping a careful evaluation of the nanotoxicity associated with the product.

Though, additional examination of plant growth in the soil will be needed to understand growth patterns and association with NP fertilizers on a more extensive type of plants. Moreover, particular mechanism of NP absorption and circulation inside the plant will be significant in growing new iron oxide NP-based fertilizers and is as of now under inspection. At present, studies are being directed on hyperspectral imaging and TEM investigation of the shoot and leaf tests from plants developed with NP-soaked seeds to accomplish this objective. Hyperspectral imaging to
detect NP accumulation in the root, shoot, and leaves, elemental analysis of the plant samples, and enzymatic assay on the NP-interacted plant samples will be conducted to understand the role of NPs in plant growth. A comparative cost analysis will be performed for the NP fertilizer developed and commercially available fertilizers to project the estimated profit in using NP fertilizers to enhance agricultural production.
REFERENCES


APPENDIX

PICTURES OF ROOT GROWTH OF DIFFERENT TYPES OF SEEDS AND ITS MATERIAL CHARACTERIZATION AFTER SOAKING IN THE CONTROL, HIGH, AND LOW CONCENTRATION OF IRON OXIDE NANOPARTICLES
Root growth chickpeas, green peas and red beans after soaking in the control, high, and low concentration of iron oxide nanoparticles.
Root growth black beans and green gram seeds after soaking in the control, high, and low concentration of iron oxide nanoparticles.
The following pictures indicated the material characterizations of the Black bean root under the observation of Micromaster Microscope. For DI water pH 5.5, 8 and Low concentration NPs pH 5.5, 8.

Material characterization of the black bean root images under the Micromaster Microscope:

A. DI water pH 5
B. Low concentration pH 5
C. DI water pH 8
D. Low concentration pH 8.
VITA

Uday Gharge was born in Maharashtra, India, to the parents Bhagwan and Ujwala Gharge. Uday attended Shivaji University in Kolhapur, India, where he studied for a Bachelor of Engineering in Chemical Engineering. After graduating from university in India, Uday moved to Chattanooga, Tennessee in 2016 to pursue a Master of Science degree in Chemical Engineering at The University of Tennessee at Chattanooga. After joining this University, he accepted a Research Assistantship at The University of Tennessee at Chattanooga in August 2016. There, he worked with Dr. Soubantika Palchoudhury on nanomaterials and investigated their toxicological effects on environment.