Green Synthesized Iron Oxide Nanoparticles: A Nano-Nutrient for the Growth and Enhancement of Flax (*Linum usitatissimum* L.) Plant

G. Karunakaran, M. Jagathambal, N. Van Minh, E. Kolesnikov, A. Gusev, O. V. Zakharova, E. V. Scripnikova, E. D. Vishnyakova, D. Kuznetsov

***Abstract*—**Iron oxide nanoparticles (Fe_2O_3NPs) are widely used in different applications due to its ecofriendly nature and biocompatibility. Hence, in this investigation, biosynthesized Fe_2O_3NPs influence on flax (*Linum usitatissimum* L.) plant was examined. The biosynthesized nanoparticles were found to be cubic phase which is confirmed by XRD analysis. FTIR analysis confirmed the presence of functional groups corresponding to the iron oxide nanoparticle. The elemental analysis also confirmed that the obtained nanoparticle is iron oxide nanoparticle. The scanning electron microscopy and the transmission electron microscopy confirm that the average particle size was around 56 nm. The effect of Fe_2O_3NPs on seed germination followed by biochemical analysis was carried out using standard methods. The results obtained after four days and 11 days of seed vigor studies showed that the seedling length (cm), average number of seedling with leaves, increase in root length (cm) was found to be enhanced on treatment with iron oxide nanoparticles when compared to control. A positive correlation was noticed with the dose of the nanoparticle and plant growth, which may be due to changes in metabolic activity. Hence, to evaluate the change in metabolic activity, peroxidase and catalase activities were estimated. It was clear from the observation that higher concentration of iron oxide nanoparticles (Fe_2O_3NPs 1000 mg/L) has enhanced peroxidase and catalase activities and in turn plant growth. Thus, this study clearly showed that biosynthesized iron oxide nanoparticles will be an effective nano-nutrient for agriculture applications.

***Keywords*—**Catalase, fertilizer, iron oxide nanoparticles, *Linum usitatissimum* L., nano-nutrient, peroxidase.

I. Introduction

CURRENTLY, science and technology is booming everywhere giving birth to new technologies [1] such as nanotechnology, one of the biggest achievements in science and technology. Nanotechnology covers almost every field of science and technology by its applications [2]. The recent survey has shown that nanotechnology has taken majority of contributions towards the leading world market [3]. Nanotechnology includes the synthesis and application of nanoparticles [4]. Nanoparticles are of different types such as metal based, metal oxides and composites [5]-[7]. The high surface to volume ratio has made the nanoparticles unique and efficient leading to their wide applications in fields like energy, food science, and electronics and biomedicine. However, the application of nanoparticles in agriculture is in elementary stage.

Recently, nanotechnology is focusing on the detection of plant diseases, plant disease management, and development of nano-nutrient for the enhancement of plant growth [8], [9]. In general, plant growth is promoted by applying commercially available fertilizers. However, most of the fertilizers are toxic to humans, animals, and ecosystem. In addition, application of fertilizer also faces problems like hydrolysis, decomposition, and leaching. Hence, the available fertilizers are less effective. Thus, an alternative to chemical fertilizer, which can overcome the above problems, is required now. Nanoparticle-based and nano-encapsulated fertilizers are found to be effective in enhancing plant growth by promoting precise release of nutrients to the plant [10], [11]. However, some limitations had led to its failure. In contrast, few researches have shown that some engineered nanoparticles boost the growth of plant by altering its metabolisms [12]-[16]. Recently, lot of researches are being focused on the evaluating the toxicity of nanoparticles towards microbial biomass [17], plant growth promoting bacteria [18], *Pseudomonas fluorescens* [19], cell lines [20], [21], *Chlorella pyrenoidosa* [22], maize seeds germination [23], [24], and plant phytochemicals [25]. To our surprise, only few reports are available on the beneficial effect of nanoparticles on plant growth. Hence, further research is essential to check the beneficial/toxic effects of nanoparticles on plant growth. With this insight, the present study was carried to develop a nanoparticle, which could improve the growth and yield of plant.

A wide variety of nanoparticles such as calcium, magnesium, cobalt, nickel and iron oxide are available at present, among which iron oxide nanoparticles are most commonly and widely used. It is being used in different applications such as catalyst, drug delivery, environmental

Gopalu Karunakaran is with the Department of Functional Nanosystems and High-temperature Materials, National University of Science and Technology “MISiS”, Russia, Moscow, No. 4, Leninsky Prospect, Moscow, Russia and with the Department of Biotechnology, K. S. Rangasamy College of Arts and Science, Tiruchengode-637215, Tamil Nadu, India (corresponding author, phone: +7-985-663-75-69, e-mail: karunakarang5@ gmail.com).

Matheswaran Jagathambal is with the Department of Bio-chemistry/Bio-technology/Bio-informatics, Avinashilingam Institute for Home Science and Higher Education for Women Mettupalayam Road, Bharathi Park Road, Coimbatore -641 043, India.

Nguyen Van Minh, Evgeny Kolesnikov and Denis Kuznetsov are with the Department of Functional Nanosystems and High-temperature Materials, National University of Science and Technology “MISiS”, Russia, Moscow, No. 4, Leninsky Prospect, Moscow, Russia.

Alexander Gusev, Olga V. Zakharova, Elena V. Scripnikova, and Elena D. Vishnyakova are with the G .R. Derzhavin Tambov State University, 33, Internatsionalnaya Street, Tambov, 392000, Russia.

protection and sensors, because of its high activity and less toxic to the environment [26]-[28]. Iron oxide nanoparticles increased the root growth of soybean plant [29], as well as enhanced seed germination in mung bean and watermelon plants [30], [31]. However, iron oxide was found to be toxic to algae such as *Isochrysis sp.* and *Nannochloropsis sp.* [32]. Hence, the toxic effect of iron nanoparticles is not yet clearly understood. Hence, in this study, we have synthesized iron oxide nanoparticle in controlled condition, and its influence on the plant growth was determined using standard protocols.

II. Materials and Turning Tests

The precursor solution was prepared by using 0.2 M iron nitrate Fe $(NO_3)_2.6H_2O$, of AR grade with water as solvent, (Milli-Q Water, Millipore, Germany).

Flowers of *Hydrangea paniculata* were collected from the nearby Gorky Park, Russia. The flower petals were plucked and washed using sterile water to remove the adhered dust particles. The excess water was allowed to drain out at room temperature. Petals (10 g) were crushed well with mortar and pestle, and the extract was collected using Whatman No.1 filter paper. The process of filtration continued until a clear homogeneous extract is obtained.

Green synthesis was performed according to our previous study on nickel, magnesium and silver nanoparticles synthesis [33], [34]. To 50 ml of 0.2 M iron nitrate (Fe $(NO_3)_2.6H_2O$) solution on a magnetic stirrer, the flower extract was added in a drop-wise manner until the color of the solution changes. The addition of flower extract was continued until viscous colloids were obtained. The colloidal solution was centrifuged for 25 mins at 4300 rpm. The process of centrifugation was continued until a clear supernatant was obtained. This process was carried out to remove the excess of the extract, non-reactive and other unwanted residues present in the nanoparticles suspension. Once the supernatant becomes clear, the pellet was carefully collected from the bottom of the centrifuge tube followed by drying under 40-50 ^{o}C to remove the excess moisture present in it. The dried pellet was crushed into a fine powder with the help of sterile mortar and pestle.

The fine powder was subjected to various analyses, in order to confirm the formation of the targeted nanoparticle. To investigate the crystalline nature of the powder, X-ray diffraction (Difray, Saint Petersburg) technique was used using chromium (λ = 2.2909 Å) as X-ray source. To explore the different functional groups present in the nanoparticles, Fourier transform (FTIR) infrared spectrophotometer (Nicolet 380, USA) technique was used. The presence of elements in the nanoparticles was confirmed by energy dispersive (EDS) spectrum analysis (EDX SSD, JAPAN). The structural characterisation of the synthesized nanoparticles was done by scanning electron microscope (SEM-VEGA3 TESCAN, Brno, Czech Republic) analysis.

To assess the influence of the synthesized nanoparticle on the growth and development of plants *Linum usitatissimum L.*, viability of seeds and morphometric parameters were checked. For the plants grown in laboratories, germinating ability and vigour of germination in seeds, length of roots and vegetating parts were analyzed. To study germination, the seeds were placed in humid chamber consisting moistened filter paper with the nanoparticle solution (1 mg/L, 100 mg/L and 1000 mg/L) in a Petri dish. Each variant of the experiment was produced in four replicas. Each replica consisted of 100 seeds. Distilled water was used as control for comparison. Germination rate was assessed by the method suggested by McGuire. The procedure was followed by accounting the number of days required for completing germination studies [35]. The results were processed with Student's T-criterion at 5% significance level.

Activity of antioxidant system enzymes, such as peroxidase and catalase was analyzed in seedlings. The activity of peroxidase was determined spectrophotometrically by measuring the oxidation of benzidine (4,4'-diaminobiphenyl) in the presence of hydrogen peroxide and peroxidase [36]. The activity of catalase was determined by measuring the decomposition of hydrogen peroxide with catalase spectrophotometrically [37].

III. Results and Discussion

The phase of the green synthesized nanoparticles was analyzed by using XRD technique and is shown in Fig. 1. The obtained 2θ values were at ~27.7°, ~36.1°, ~39.4°, ~45.9°, ~54.3°, ~66.7°, ~69°, ~77°, ~84.6°, ~91.1°, ~101.8°, ~120.6°, ~128.4°, and ~131.2°. These values matched well with the ICDD standard data files (JCPDS: 96-900-5817) for iron oxide (Fe_2O_3), thus confirming that the synthesized Fe_2O_3 nanoparticles possessed face centered cubic crystal system. The functional groups present in the synthesized nanoparticle were determined by FTIR analysis. The FTIR spectrum as shown in Fig. 2 revealed different peaks at 3139, 1652, 1511, 1348, 890, and 792 cm^{-1}.

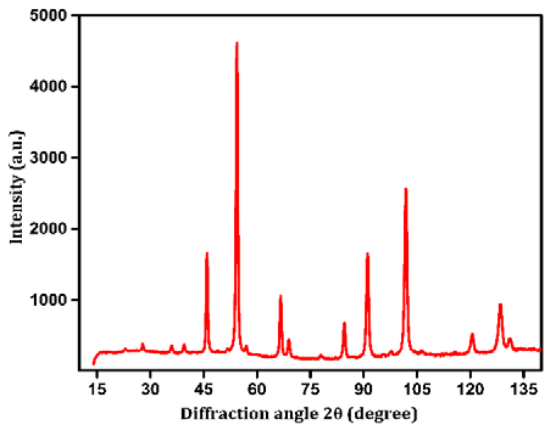

Fig. 1 XRD pattern of Iron oxide nanoparticles

Peak at 3139 indicated the presence of O-H group, which may be due to the organic molecules like esters, organic acids and phenols that participates in nanoparticles synthesis. Peaks at 1652, 1511, and 1348 cm^{-1} indicated –CH, C-H and C=O groups, whereas peaks at 890 and 792 cm^{-1} confirmed the presence of iron and its corresponding forms [38]. The organic molecules that were present in the plant extract might have attached to the nanoparticles and hence, reflected in FTIR analysis.

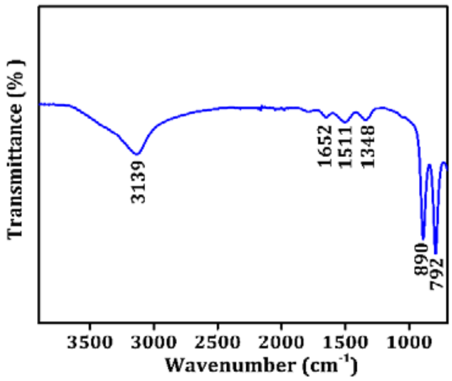

Fig. 2 FTIR spectrum of Iron oxide nanoparticles

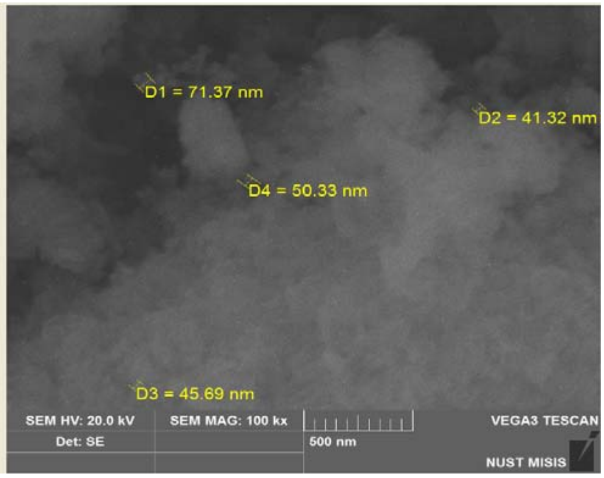

Fig. 3 SEM image of Iron oxide nanoparticles

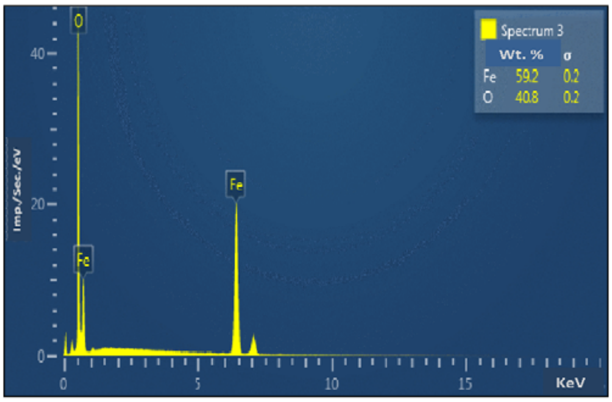

Fig. 4 Elemental analysis (EDS) of Iron oxide nanoparticles

The surface topology of the synthesized nanoparticles was analyzed by scanning electron microscopy and is given in Fig. 3. The observed result revealed the spherical shape of the nanoparticles. The elemental compositions of the synthesized nanoparticles were evaluated by EDS analysis (Fig. 4). The result indicated that the nanoparticles contained about 59.2% of iron and 40.8% of oxygen. TEM analysis (Fig. 5) revealed the size of the nanoparticles to be in the ranges 41 to 71 nm, however the average particle size was found to be 56 nm.

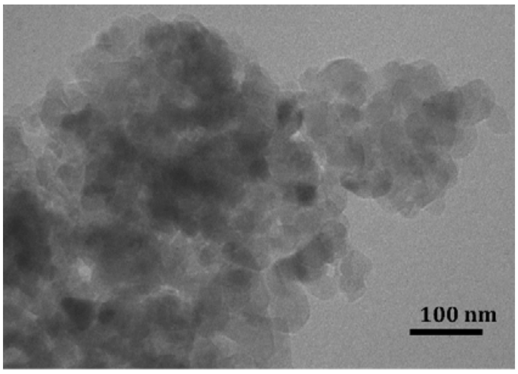

Fig. 5 Transmission electron microscopy image of Iron oxide nanoparticles

The influence of iron oxide nanoparticles on seed germination and level of antioxidant enzymes was studied employing standard protocols using different concentrations (1 mg/L, 100 mg/L and 1000 mg/L). The results obtained are shown in Tables I-III. The seedling morphometric was performed after 11 days of treatment, and the seed vigor was analyzed after four days of treatment. The seedling morphometric was found to vary with respect to nanoparticle concentration. It was noticed that the seedling length (cm) increased with an increase in the concentration of the iron oxide nanoparticles when compared to control.

The results obtained after four days of seed vigor studies showed that the average number of seedling with leaves was found to increase with iron oxide nanoparticles treatment when compared to control. The root morphometric study was performed after 11 days of the treatment. The observed difference in the root morphometric was based on the difference in the concentration of the nanoparticles used for the analysis. The higher is the concentration of the nanoparticles, the greater is the root length (cm), when compared to control.

The total length of lateral roots also increased on treatment with iron oxide nanoparticles. Plants respond to stress and injury by changing their metabolic activity (such as stress hormone, jasmonic acid, and auxin), which in turn leads to a change in their morphology and growth rate. Such a phenomenon is known as thigmomorphogenesis. Hence, to evaluate the change in metabolic activity due to nanoparticle treatment, peroxidase and catalase activities were estimated [39].

It was clear from the observed results that the order of peroxidase and catalase activity was found to be Fe_2O_3NPs 1000 mg/L > Fe_2O_3NPs 100 mg/L > Fe_2O_3NPs 1 mg/L. This clearly showed that nanoparticles have decreased oxidative stress in plant by triggering the enzyme activity, and hence reduced the formation of reactive oxygen species (ROS) [40]. The reduction of ROS has promoted the plant growth as reflected from seedling and root lengths. Thus, iron oxide nanoparticles could efficiently trigger plant growth and hence, could be used instead of harmful fertilizers for agriculture applications.

TABLE I
SEEDLINGS MORPHOMETRIC (11 DAYS) AND SEED VIGOR (4 DAYS) STUDIES

Treated Sample names	Seedlings length (cm)	Average Number of seedlings with leaves (n)	Average Number of seedlings with roots (n)
Control	5.1	4	6
Fe_2O_3NPs 1 mg/L	5.9	4	10
Fe_2O_3NPs 100 mg/L	6.2	3	10
Fe_2O_3NPs 1000 mg/L	6.6	5	23

TABLE II
ROOTS MORPHOMETRIC STUDIES (11 DAYS)

Treated Sample names	Length of main root (cm)	Total length of lateral roots (cm)	Number of lateral roots, pieces
Control	4.1	3.5	3.1
Fe_2O_3NPs 1 mg/L	4.8	3.0	3.1
Fe_2O_3NPs 100 mg/L	5.1	3.1	3.3
Fe_2O_3NPs 1000 mg/L	5.7	3.8	4.0

TABLE III
PEROXIDASE AND CATALASE ACTIVITY IN SEEDLINGS

Treated Sample names	Peroxidase activity, mU/mg	Catalase activity, mU/mg
Control	4.568	24.1
Fe_2O_3NPs 1 mg/L	5.981	28.32
Fe_2O_3NPs 100 mg/L	6.551	29.67
Fe_2O_3NPs 1000 mg/L	6.751	31.58

IV. CONCLUSION

In this study, iron oxide nanoparticles were synthesized by biosynthesis method and its influence on the growth and yield of Flax (*Linum usitatissimum* L.) plant was analyzed by using standard protocols. The synthesized iron oxide nanoparticles were characterized by different characterization techniques to confirm its physicochemical properties. The obtained result revealed that the nanoparticle exhibited cubic phase and possessed spherical shaped with size ranged between 41 to 71 nm. The average particle size was 56 nm. The influence of different nanoparticles on seed germination and antioxidant enzymes (peroxidase and catalase) was analyzed using standard protocols. It was observed that the seedling length (cm), average number of seedling with leaves and root length (cm) were increased with an increase in the concentration of the nanoparticles when compared to control. Hence, to evaluate the change in metabolic activity due to nanoparticle treatment, peroxidase and catalase activities were measured. The result showed that higher concentration of iron oxide nanoparticles (Fe_2O_3NPs 1000 mg/L) has enhanced the activity of both the enzymes indicating the inhibition of ROS generation and hence promoted plant growth. Thus, it is concluded that iron oxide nanoparticles could be efficiently applied in agriculture for the better growth of plants in place of harmful chemical fertilizers.

ACKNOWLEDGMENT

The work was carried out with financial support from the Ministry of Education and Science of the Russian Federation in the framework of Increase Competitiveness Program of NUST «MISiS» (№ К4-2015-017), *implemented by a governmental decree dated 16th of March 2013, N 211.*

REFERENCES

[1] A. Sankaranarayanan, G. Munivel, G. Karunakaran, S. Kadaikunnan, N. S. Alharbi, J. M. Khaled and D. Kuznetsov, “Green Synthesis of Silver Nanoparticles Using *Arachis hypogaea* (Ground Nut) Root Extract for Antibacterial and Clinical Applications,” Journal of Cluster Science, 2016, pp. 1-14, doi:10.1007/s10876-016-1084-x.

[2] R. F. Service, “Is nanotechnology dangerous?” Science, 2000, vol. 290, pp. 1526-1527.

[3] G. Karunakaran, M. Jagathambal, A. Gusev, E. Kolesnikov, A. R. Mandal and D. Kuznetsov, “Allamanda cathartica flower's aqueous extract-mediated green synthesis of silver nanoparticles with excellent antioxidant and antibacterial potential for biomedical application,” MRS Communications, vol. 6. 2016, pp. 41-46.

[4] G. Karunakaran, Matheswaran Jagathambal, Alexander Gusev, Nguyen Van Minh, Evgeny Kolesnikov, Arup Ratan Mandal and Denis Kuznetsov, “*Nitrobacter* sp. extract mediated biosynthesis of Ag_2O NPs with excellent antioxidant and antibacterial potential for biomedical application,” IET Nanobiotechnology, vol. 10, 2016, pp. 425 – 430.

[5] G. Karunakaran, M. Jagathambal, A. Gusev, J.A.L. Torres, E. Kolesnikov, and D. Kuznetsov, “Rapid Biosynthesis of AgNPs Using Soil Bacterium *Azotobacter vinelandii* With Promising Antioxidant and Antibacterial Activities for Biomedical Applications,” JOM, 2016, pp. 1-7, doi:10.1007/s11837-016-2175-8.

[6] N. R. Dhineshbabu, G. Karunakaran, R. Suriyaprabha, P. Manivasakan, P. Prabu, and V. Rajendran, “Electrospun MgO/Nylon 6 Hybrid Nanofibers for Protective Clothing,” Nano-Micro Letters vol. 6, 2014, pp. 46-54.

[7] G. Karunakaran, Andrey Grigorjevich Yudin, Matheswaran Jagathambal, Arup Ratan Mandal, Nguyen Van Minh, Alexander Gusev, Evgeny Kolesnikov, and Denis Kuznetsov, “Synthesis of five metal based nanocomposite via ultrasonic high temperature spray pyrolysis with excellent antioxidant and antibacterial activity,” RSC Advances, vol. 6, 2016, pp. 37628-37632.

[8] G. Karunakaran, R. Suriyaprabha, P. Manivasakan, R. Yuvakkumar, V. Rajendran, and N. Kannan, “Effect of nanosilica and silicon sources on plant growth promoting rhizobacteria, soil nutrients and maize seed germination,” IET-Nanobiotechnology, vol.7, 2013, pp. 70-77.

[9] G. Karunakaran, R. Suriyaprabha, P. Manivasakan, R. Yuvakkumar, V. Rajendran, and N. Kannan, “Influence of nano and bulk SiO_2 and Al_2O_3 particles on plant growth promoting rhizobacteria and soil nutrient contents,” Current nanoscience, vol. 10, 2014, pp. 604-612.

[10] M.C. DeRosa, C. Monreal, M. Schnitzer, R. Walsh and Y. Sultan, “Nanotechnology in fertilizers,” Nature Nanotechnology, vol. 5, 2010, pp. 91. doi:10.1038/nnano.2010.2.

[11] R. Nair, S.H. Varghese, B.G. Nair, T. Maekawa, Y. Yoshida and D.S. Kumar, “Nanoparticulate material delivery to plants,” Plant Science, vol. 179, 2010, pp. 154–163.

[12] J.P. Giraldo, M.P. Landry, S.M. Faltermeier, T.P. McNicholas, N.M. Iverson, A.A. Boghossian, N.F. Reuel, A.J. Hilmer, F. Sen, J.A. Brew and M.S. Strano, “Plant nanobionics approach to augment photosynthesis and biochemical sensing,” Nature Materials, 2014, doi:10.1038/nmat3890.

[13] D.W. Galbraith, “Nanobiotechnology: silica breaks through in plants,” Nature Nanotechnology, vol. 2, 2007, pp. 272–273.

[14] F. Torney, B.G. Trewyn, V.S.-Y. Lin and K. Wang, “Mesoporous silica nanoparticles deliver DNA and chemicals into plants,” Nature Nanotechnology, vol. 2, 2007, pp. 295–300.

[15] M.H. Lahiani, E. Dervishi, J. Chen, Z. Nima, A. Gaume, A.S. Biris and M.V. Khodakovskaya, “Impact of carbon nanotube exposure to seeds of valuable crops,” ACS Applied Material Interfaces, vol. 5, 2013, pp. 7965–7973.

[16] M.H. Siddiqui and M.H. Al-Whaibi “Role of nano-SiO_2 in germination

of tomato (*Lycopersicum esculentum* seeds Mill.),” Saudi Biological Science, vol. 21, 2014, pp. 13–17.

[17] R. Suriyaprabha, G. Karunakaran, R. Yuvakkumar, P. Prabu, V. Rajendran, and N. Kannan, “Effect of silica nanoparticles on microbial biomass and silica availability in maize rhizosphere,” Biotechnology and Applied Biochemistry, vol. 61, 2014, pp. 668–675.

[18] G. Karunakaran, R. Suriyaprabha, P. Manivasakan, R. Yuvakkumar, V. Rajendran, and N. Kannan, “Impact of Nano and Bulk ZrO2, TiO2 Particles on Soil Nutrient Contents and PGPR,” Journal of Nanoscience and Nanotechnology, vol. 13, 2013, pp. 678-685.

[19] R. Suriyaprabha, G. Karunakaran, R. Yuvakkumar, P. Prabu, V. Rajendran, and N. Kannan, “Augmented biocontrol action of silica nanoparticles and *Pseudomonas fluorescens* bioformulant in maize (Zea mays L.),” RSC Advances, vol. 4, 2014, pp. 8461,

[20] G. Karunakaran, R. Suriyaprabha, P. Manivasakan, R. Yuvakkumar, V. Rajendran, and N. Kannan, “Screening of in vitro cytotoxicity, antioxidant potential and bioactivity of nano and micro ZrO_2 and TiO_2 particles,” Ecotoxicology and Environmental Safety, vol. 93, 2013, pp. 191-197.

[21] G. Karunakaran, R. Suriyaprabha, P. Manivasakan, V. Rajendran, and N. Kannan, Effect of contact angle, zeta potential and particles size on in vitro behaviour of Al_2O_3 and SiO_2 nanoparticles, IET-Nanobiotechnology, vol. 9, 2015, pp. 27 – 34.

[22] G. Karunakaran, M. Jagathambal, A. Gusev, E. Kolesnikov, and D. Kuznetsov, “Assessment of FeO and MnO Nanoparticles Toxicity on Chlorella pyrenoidosa,” Journal of Nanoscience and Nanotechnology, vol. 17, 2017, pp. 1712-1720.

[23] G. Karunakaran, R. Suriyaprabha, V. Rajendran, and N. Kannan, “Influence of ZrO_2, TiO_2, SiO_2 and Al_2O_3 nanoparticles on maize seed germination under different growth conditions,” vol. 10, 2016, 171-177.

[24] R. Suriyaprabha, G. Karunakaran, P. Manivasakan, R. Yuvakkumar, V. Rajendran, and N. Kannan, “Application of silica nanoparticles in maize (Zea mays. L) to enhance fungal resistance,” IET-Nanobiotechnology, vol. 8, 2014, pp. 133–137.

[25] R. Suriyaprabha, G. Karunakaran, R. Yuvakkumar, V. Rajendran, and N. Kannan, “Foliar application of silica nanoparticles on the phytochemical responses of maize (Zea mays L.) and its toxicological behaviour,” Synthesis and Reactivity in Inorganic, Metal-Organic, and Nano-Metal Chemistry, vol. 44, 2014, pp.1128–1131,

[26] S. He, Y. Feng, H. Ren, Y. Zhang, N. Gu and X. Lin, “The impact of iron oxide magnetic nanoparticles on the soil bacterial community,” Journal of Soils Sediments, vol. 11, 2011, pp. 1408-1417.

[27] J.M. Perez, T. Oloughin, F.J. Simeone, R. Weissleder and L. Josephson, “DNA based magnetic nanoparticle assembly acts as a magnetic relaxation nanoswitch allowing screening of DNA cleaving agents,” Journal of the American Chemical Society, vol. 124, 2002, pp. 2856-2857.

[28] Y.Q. Wang, J. Hu, Z.Y. Dai, J.L. Li and J. Huang, “In vitro assessment of physiological changes of watermelon (Citrullus lanatus) upon iron oxide nanoparticles exposure,” Plant Physiology and biochemistry, vol. 108, 2016, pp. 353-360.

[29] D. Alidoust and A. Isoda, “Effect of Fe_2O_3 nanoparticles on photosynthetic characteristic of soybean (Glycine max (L.) Merr.): foliar spray versus soil amendment,” Acta Physiologiae Plantarum, vol. 35, 2013, pp. 3365-3375.

[30] H. Ren, L. Liu, C. Liu, S. He, J. Huang, J. Li, Y. Zhang, X. Huang and N. Gu, “Physiological investigation of magnetic iron oxide nanoparticles towards chinese mung bean,” Journal of Biomedical Nanotechnology, vol. 7, 2011, pp. 677-684.

[31] J. Li, P. Chang, J. Huang, Y. Wang, H. Yuan and H. Ren, “Physiological effects of magnetic iron oxide nanoparticles towards watermelon,” Journal of Nanoscience and Nanotechnology, vol. 13, 2013, pp. 5561-5567.

[32] V. Demir, M. Ates, Z. Arslan, M. Camas, F. Celik, C. Bogatu, S. S. Can, “Influence of alpha and gamma-iron oxide nanoparticles on marine microalgae species,” Bulletin of Environmental Contamination and Toxicology, vol. 95, 2015, pp. 752-757.

[33] M. Kundu, G. Karunakaran and D. Kuznetsov, “Green synthesis of NiO nanostructured materials using *Hydrangea paniculata* flower extracts and their efficient application as supercapacitor electrodes,” Powder Technology, In Press, 2017, doi:10.1016/j.powtec.2017.01.085.

[34] G. Karunakaran, M. Jagathambal, M. Venkatesh, G.S. Kumar, E. Kolesnikov and D. Kuznetsov, “*Hydrangea paniculata* flower extract-mediated green synthesis of MgNPs and AgNPs for health care applications,” Powder Technology, vol. 305, 2017, pp. 488-494.

[35] J.D. McGuire, “Speed of germination-aid selection and evaluation for seedling emergence and vigor,” Crop Sciences, 1962, pp. 176–177.

[36] F. Van Assche, C. Cardinaels, H. Clijsters, “Induction of enzyme capacity in plants as a result of heavy metal toxicity: Dose-response relations in Phaseolus vulgaris L., treated with zinc and cadmium,” Environmental Pollution, vol. 52, 1988, pp. 103–115.

[37] H. Aebi, “Catalase in vitro,” 1984, pp. 121–126.

[38] M Kundu, G Karunakaran, N Van Minh, D Kuznetsov, Improved Electrochemical Performance of Nanostructured Fe_2O_3 Anode Synthesized by Chemical Precipitation Method for Lithium-ion Batteries, Journal of Cluster Science, (2016). doi:10.1007/s10876-016-1140-6.

[39] E.W. Chehab, E. Eich and J. Braam, “Thigmomorphogenesis: a complex plant response to mechano-stimulation,” Journal of Experimental Botany, vol. 60, 2009, pp. 43–56.

[40] C. Krishnaraj, E.G. Jagan, R. Ramachandran, S.M. Abirami, N. Mohan and P.T. Kalaichelvan, “Effect of biologically synthesized silver nanoparticles on Bacopa monnieri (Linn.) Wettst. plant growth metabolism,” Process biochemistry, vol. 47, 2012, pp. 651-658.