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Silver nano-particles enhance the growth, yield and nutrient use efficiency of wheat

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Abstract

Application profile of nano particles is rapidly expanding even in agriculture. Silver nano particles (SNPs) are hypothesized to enhance nutrient use efficiency in plants. Present study was carried out to determine the role of SNPs for improving (NUE) in wheat. The SNPs were synthesized chemically by reducing silver nitrate with trisodium citrate and size was 10-20 nm according to X-Ray Diffraction analysis. Completely randomized design with seven graded doses of SNPs (0, 25, 50, 75, 100, 125, 150 ppm) and four replications was employed for experimental layout. Seedlings of wheat variety NARC-2009 were transplanted to pots. Pot soil was soaked with SNPs solution up to field capacity levels and distilled water was applied in control treatment. SNPs significantly enhanced most of the growth and yield attributes NPK uptake and nutrient use efficiency of wheat. Silver nanoparticles in 25ppm concentration have showed significant improvement in maximum leaf area and highest grain yield while 75ppm concentration resulted in decrease in grain yield. So silver nanoparticles have stimulatory as well as inhibitory effect on wheat growth and yield.

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Introduction

Production and quality of food can be improved by modern technologies which can meet ever increasing world food demand in environment friendly way (Wheeler, 2005). Nanotechnology seems to have potential for addressing the problem of food security (Anonymous, 2009).

Application of nano particles in agriculture and food system can reform the field by detection of diseases, resistance against diseases, targeted delivery, promoting the efficiency of plants to uptake more nutrients; endure environmental pressure and efficient system for processing and storage (Mousavi and Rezaei, 2011). It offers a new dimension for selection and dispensation of those resources that improve the eminence of product (Sharon *et al.*, 2010).

Silver nano particles (SNPs) are excellent material having antibacterial, antifungal properties and are used in food and agriculture such as food security, food packaging and pathogen detection (Quardos and Mar, 2010). It has great influence on plant growth and development such as germination, root-shoot ratio, seedling growth, root growth, root elongation, and senescence inhibition (Ma *et al.*, 2010; Shah and Belozerova, 2009). Nano-bentonite and nano active carbon coated nitrogen fertilizer increased the absorption and transportation of N, P, K to seed and yield of rice significantly (Wang *et al.*, 2011). Nano carbon as slow release fertilizer increased chlorophyll content, grain yield and nitrogen use efficiency of rice (Wu *et al.*, 2010). Application of control release fertilizer coated by nano-material increased Chinese cabbage yield and improved nutrient use efficiency significantly as compared to common straight fertilizer (Ding *et al.*, 2009).

Use of mineral fertilizers has played an important role for the survival of mankind in terms of increasing yield (Smil, 2001; Stewart *et al.*, 2005), maintaining soil productivity and fertility (Balmford *et al.*, 2005). Nutrient removal is a major cause for low crop yield in parts of the developing world. In Pakistan, 50% losses occur due to low nutrient use efficiency (Zia *et al.*, 1991). Moreover, socio-economic constraints have made it more imperative to increase nutrient use efficiency. Hence, increasing nutrient use efficiency continues to be a major challenge for world agriculture, and nano technology can potentially address this issue. Therefore, present study was carried out to explore the role of SNPs to enhance wheat growth, yield and nutrient use efficiency.

Materials and methods

Synthesis of Silver Nano Particles

Silver nano particles synthesized chemically by the reduction of silver nitrate ($AgNO_3$) with tri-sodium citrate dehydrate (Kulkarni, 2007) as below:

$$3AgNO_3 + Na_3C_6H_5O_7.2H2O \longrightarrow C_6H_5O_7 + 3NaNO_3 + CO_2 + 3Ag + 2H_2O$$

Silver nano particles were prepared by reducing 510 mg of $AgNO_3$ by 294 mg of $Na_3C_6H_5O_7.2H_2O$ through continuous stirring (6000-7000 rpm) at 70 ^oC on a magnetic stirrer, until golden color appeared. Ascorbic acid @ 1mg/L was used as stabilizing agent. This reaction ultimately produced a solution of approximately 300 ppm SNPs. Dilutions were made from stock solution for application of treatments. Size of SNPs was determined by X-Ray Diffraction (XRD) which revealed that particles size ranged between 10-20 nm (Fig. 1).

Experimental Procedure

Wheat variety NARC-2009 was grown for this experiment. Completely randomized design (CRD) with four replications and seven treatments was employed for experimental layout. Treatments included: 25, 50, 75, 100, 125 and 150 ppm of SNPs and only distilled water served as control. Sterilized seeds of wheat were placed in petri dishes containing three layers of filter papers for germination. Distilled water (15 mL) was added to soak filter papers uniformly. Pots were filled with equal amount of thoroughly mixed 15 kg soil. Soil analysis was performed before the experiment for nutrient status of soil. Recommended doses of N, P, K (90, 60 and 60 kg ha^{-1}) were applied by using urea, diammonium phosphate and potassium chloride respectively. Calculated amount of N, P and K based on recommended doses @ 0.73, 0.65 and 0.5 g/pot were applied in pots. Ten seedlings were transplanted to each clay pot. Solutions equivalent to field capacity containing different concentrations of SNPs were applied to pots. Water was applied to pots in equal amount when needed. Soil was analysed with the procedures as: soil pH (Gupta 2006), electrical conductivity (Richard, 1954), nitrate-nitrogen (Vandrell and Zupanic 1990), phosphorous (Olsen *et al.* 1954) and potassium (Helmke and Sparks 1996) in the lab. Data on the following plant parameters were collected to evaluate the effect of SNPs on nutrient use efficiency.

At flag leaf stage, fully expended three leaves were selected, leaf area was measured by using leaf area meter and average value of three leaves was recorded. From each pot one plant was taken at flag leaf stage. Shoots were separated from roots. Fresh weight (FW) was recorded by using electrical balance. Shoots were dried at 70 °C in oven. After 36 hours dry weight (DW) was recorded. Chlorophyll content determines the growth and development of the plants. It was recorded by using SPAD chlorophyll meter. From each pot three representative plants were selected and chlorophyll content was measured. Average values were used for statistical analysis. At maturity, plants were harvested and different yield parameters were recorded. Five plants from each pot were selected and data on number of grains/spike, 100 grains weight (g) and grain yield/pot (g) was recorded.

Nutrient use efficiency (NUE) for N, P and K was calculated by the following formula.

$$\text{Nitrogen Use Efficiency (\%)} = \frac{\text{Yield from treated plants} - \text{Yield from control plants}}{\text{Weight of Nitrogen Applied}} \times 100$$

$$\text{Phosphorous Use Efficiency (\%)} = \frac{\text{Yield from treated plants} - \text{Yield from control plants}}{\text{Weight of Phosphorous Applied}} \times 100$$

$$\text{Potassium Use Efficiency (\%)} = \frac{\text{Yield from treated plants} - \text{Yield from control plants}}{\text{Weight of Potassium Applied}} \times 100$$

Statistical analysis

Analysis of variance was determined and means were compared by employing Least Significant Difference (LSD) Test at 5 % level of probability. The statistical work was done using the computer based statistical package MSTATC following Steel *et al.*, (1997).

Results and discussion

Soil analysis results are presented in the (Table-1). The pH of the soil was 7.09, electrical conductivity 0.79 dSm^{-1}, nitrate nitrogen 0.035 $mgkg^{-1}$, available phosphorous 5.30 $mgkg^{-1}$ and available potassium is 80.0 mg kg^{-1}.

Table 1. Soil used for wheat cultivar NARC-2009.

Soil Depth	*Electrical Conductivity (dSm^{-1})*	*pH*	*Nitrate Nitrogen (mg kg^{-1})*	*Available Phosphorous (mg kg^{-1})*	*Available Potassium (mg kg^{-1})*
0-15cm	0.79	7.09	0.035	5.30	80

Wheat Growth Attributes

Different treatments of SNPs greatly affected the leaf area (Table-2).Maximum leaf area (19.7 cm^2) was with 25 ppm of SNPs followed by 50 ppm (18.18 cm^2) SNPs, while in control it was (15.0 cm^2). Further increase in concentration of SNPs reduced the leaf area. Liu *et al.*, (2005) reported that nano calcium carbonate increased leaf area of peanut significantly.

Shoot fresh weight and dry weight did not increase by the application of SNPs. Significantly at 5% probability level higher shoot fresh weight (4.75 g) and dry weight (0.90 g) were recorded in control where SNPs were not applied. The lowest shoot fresh weight (0.10 g) and dry weight (0.03 g) were recorded at 150 ppm of SNPs applied. Increasing concentration of SNPs significantly reduced shoot fresh weight and dry weight of wheat plant. Similar results were reported by Mirzajani *et al.*, (2013) in rice with application of SNPs.

Lin and Xing (2008) found that in the presence of ZnO nano particles rye grass biomass reduced significantly. When *Phaselous radiatus*, *Sorghum bicolor* and *Lolium multiflorum* were subjected to SNPs, reduced root growth, root length and biomass were observed (Yin *et al.*, 2011; Lee *et al.*, 2012).

Chlorophyll content of wheat plant differed significantly by application of SNPs (Table 2). Maximum chlorophyll content (51.2) was recorded at 75 ppm followed by 100 ppm (46.1) of SNPs as compare where as in control, it was (45.1). The lowest chlorophyll content (39.8) was recorded at 150 ppm of SNPs. Application of nano TiO_2 increased 45% chlorophyll formation and three times more photosynthesis rate in spinach (Zhang *et al.*, 2005). Gao *et al.*, (2006) tested that *Spinacia oleracia* treated with nano-anatse TiO_2 induced 2.67 times more activity of rubisco carboxylase than that of control. Hence, nano anatse promoted photosynthesis by molecular mechanism of carbon reaction. SNPs induced enhancement of chlorophyll content may enhance photosynthesis leading to more production of bio-mass and yield.

Table 2. Effect of silver nano particles on leaf area, shoot fresh weight, shoot dry weight and chlorophyll content of wheat cultivar NARC-2009.

Treatments	*Leaf Area(cm^{-2})± S.E*	*Shoot Fresh Weight (g)± S.E*	*Shoot Dry Weight (g)± S.E*	*Chlorophyll Content± S.E*
0 ppm	14.96 e±0.13	4.75 a±0.02	0.90 a±0.01	45.09 b±0.01
25 ppm	19.65a±0.13	4.05 b±0.01	0.82 b±0.01	45.58 b±0.01
50 ppm	18.19 b±0.46	4.00 b±0.01	0.73 c±0.01	45.85 b±0.02
75 ppm	17.05 c±0.45	4.04 b±0.01	0.67 d±0.01	51.18 a±0.72
100 ppm	16.26 d±0.18	3.21 c±0.01	0.60e±0.01	46.09 b±0.01
125 ppm	12.73 f±0.16	3.06 d±0.01	0.45 f±0.01	43.55 b±0.01
150 ppm	10.04 g±0.14	2.10e±0.01	0.32 g±0.01	39.75 c±0.02
LSD Values	0.31	0.10	0.03	2.79

Means sharing common letters in column do not differ significantly at 5% probability level.

Wheat yield components

Effect of different concentration of SNPs on number of grains/spike, 100 grains weight and yield is presented in (Table-3). SNPs applied @ 25 ppm produced significantly greater number of grains/spike (29.0) followed by 75 ppm (25.0). The lowest number of grains/spike (11.5) was recorded with 150 ppm of SNPs applied. Significant differences were observed among treatments for 100 grain weight. Maximum 100 grain weight (4.73 g) was produced with 50 ppm followed by 25 ppm (4.66 g) of SNPs against control (3.25 g). Minimum 100 grain weight (3.78 g) was recorded with 150 ppm treatment at 5% probability level. Maximum grain yield (13.3 g) was obtained with 25 ppm SNPs followed by 50 ppm (12.45 g) as compare to control (7.18 g) where no SNPs were applied. SNPs increased the yield may be due to growth, stimulating effect of silver (Sharon *et al.* 2010). Effect of SNPs on wheat yield has not been reported. Other particles like iron oxide significantly increased soybean yield (Sheikhbaglou *et al.*, 2010).

Nutrient use efficiency (NUE)

Effect of SNPs on nutrient use efficiency is presented in (Table-4). The highest nitrogen use efficiency (74.3 %) was observed with 25 ppm of SNPs. Further increase in concentration of SNPs was accompanied by significantly reduction in nitrogen use efficiency. The lowest nitrogen use efficiency (36.4 %) was observed with 150 ppm of SNPs applied. It seems that lower concentration of SNPs (25 ppm) significantly enhanced NUE. Effect of SNPs on nitrogen use efficiency has not been reported. Other nano particles like TiO_2 enhanced growth of spinach by absorbing more nitrogen (Yang *et al.*, 2007). Nano blended fertilizer were also found to enhance nitrogen use efficiency. Slow and controlled release fertilizer coated and blended by nano materials significantly increased nitrogen use efficiency and yield of wheat (Zhang *et al.*, 2006).

It has been reported that sulfur nano particles and SiO_2 nano particles as nano blended fertilizer improved growth and nutrient use efficiency (Ding and Wu, 2005).

Potassium use efficiency also differed significantly in plants treated with SNPs. Significantly higher potassium use efficiency (89.0 %) was recorded with 25 ppm of SNPs followed by 50 ppm (79.3 %). Higher concentrations of SNPs from 100-125 ppm (59.4 % and 54.5 %) respectively significantly decreased potassium use efficiency in comparison to control. SNPs have never been used with the objective to enhance nutrient use efficiency. Other nano particles like nano SiO_2 and TiO_2 when applied to soybean increased absorption of water and fertilizers (Lu *et al.*, 2002). Slow release fertilizer blended with nano carbon increased grain yield and N, P, K use efficiency remarkably of super hybrid rice (Wu *et al.* 2010). Nano-bentonite and nano-active carbon coated nitrogen fertilizer increased absorption and transportation of N, P, K to seed and increase yield in rice significantly (Wang *et al.*, 2011).

Table 3. Effect of silver nano particles on number of grains/spike, 100-grains weight and yield/pot of the wheat cultivar NARC-2009.

Treatments	*No of Grains/Spike ± S.E*	*100-Grain Weight ± S.E*	*Yield/Pot (g) ± S.E*
0 ppm	18.5 c±0.18	3.35e±0.012	7.18 c±0.02
25 ppm	29.0a±0.31	4.66ab±0.02	13.25 a±0.02
50 ppm	22.0bc±0.31	4.73a±0.02	12.45 a±0.34
75 ppm	25.0b±0.54	4.40 c±0.02	10.40 b±0.10
100 ppm	22.3 b±0.27	4.43 bc±0.03	10.36 b±0.11
125 ppm	22.5 b±0.34	3.94 d±0.02	9.90b±0.08
150 ppm	11.5 d±0.34	3.78 d±0.03	9.73 b±0.06
LSD values	3.52	0.25	1.77

Means sharing common letters in column do not differ significantly at 5% probability level.

Table 4. Effect of silver nano particles on nitrogen, phosphorous and potassium use efficiency of wheat cultivar NARC-2009.

Treatments	*Nitrogen Use Efficiency (%) ± S.E*	*Potassium Use Efficiency (%) ± S.E*	*Phosphorous Use Efficiency (%) ± S.E*
0 ppm	69.75 b ±0.09	69.00 c ±0.06	68.44 b ±0.06
25 ppm	74.25 a ±0.09	89.03 a ±0.05	72.53 a ±0.06
50 ppm	55.13 c ±0.12	79.25 b ±0.04	61.15 c ±0.11
75 ppm	41.38 d ±0.07	61.06 e ±0.11	46.79 d ±0.03
100 ppm	40.88 de ±0.17	59.41 f ±0.06	47.30 d ±0.06
125 ppm	39.56 e ±0.19	54.50 g ±0.05	43.50 e ±0.08
150 ppm	36.38 f ±0.18	67.88 d ±0.05	41.28 f ±0.04
LSD Values	1.42	0.66	0.6851

Means sharing common letters in column do not differ significantly at 5% probability level.

The SNPs also affected phosphorous use efficiency significantly. Maximum phosphorous use efficiency (72.5 %) was observed with 25 ppm of SNPs followed by control (68.43 %). Decrease in phosphorous use efficiency of wheat was observed by further increase in concentration of SNPs from 50-150 ppm. Minimum phosphorous use efficiency (41.3 %) was observed with 150 ppm SNPs applied.

Use of SNPs on phosphorous use efficiency has not been investigated earlier. Some other materials like nano blended composite fertilizer contain all macro and micro nutrients. Manure and amino acids significantly increased uptake of N, P, K and NUE by grain crops (Chinnumuthu and Boopathi, 2009). Nano blended organic iron chelated fertilizer increased 3.5 times more photosynthesis, 70% more leaf area and 20-200% production by increasing nutrient use efficiency (Moghadam *et al.*, 2012).

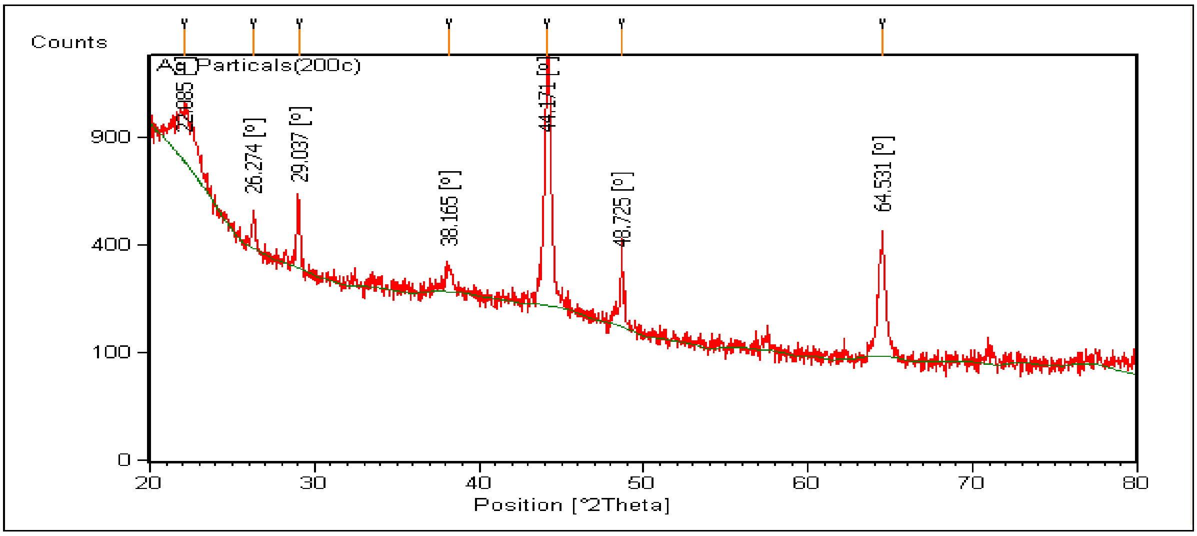

Fig. 1.X-Ray Diffraction analysis of silver nano particles.

These results revealed that application of SNPs can significantly affect nutrient use efficiency of wheat. Application of 25 ppm SNPs significantly increased N, P and K use efficiency. Further increase in SNPs caused progressive decline in nutrient use efficiency.

Conclusion

Results of present study demonstrated that by the application of silver nano particles, most of the parameters of growth and development improved. Silver nano particles @25 ppm significantly enhanced leaf area, yield and N, P and K use efficiency. However, SNPs @ 75 ppm increased chlorophyll content and SNPS @ 50 ppm produced more number of grains. Ultimately, it can be inferred that SNPs can play a crucial rule for enhancing wheat growth by improving NUE. Application of 25 ppm SNPs seems to be the most appropriate concentration for this purpose.

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References

Anonymous.2009.Nanotechnology and nano science applications: Revolution in India and beyond. Strategic Applications Integrating Nano Science.www.sainsce.com

Balmford A, Green R, Scharlemann JPW. 2005.Sparing land for nature: Exploring the potential impact of changes in agricultural yield on the area needed for crop production. Global Change Biology **11,** 1594-1605.
http://dx.doi.org/10.1111/j.1365-2486.2005

Chinnumuthu CR, Boopathi PM.2009.Nanotechnology and Agro ecosystem.The Madras Agricultural Journal **96(1-6),**17-31.

Ding H, Zhang Y, Zhignuol Z, Mingjhan Y. 2009. Effect of controlled release fertilizer on the yield and quality of Chinese cabbage and nutrient use efficiency. Journal of Changjiang Vegetables**12,**1-6.

Ding Y, Wu Q. 2005. Preparation of seed coating agent containing nano particles with low toxicity and high efficiency. Chinese Patent.
http://dx.doi.org/pdfplus/10.1021/cr400366s

EPA. 2007. Nanotechnology White Paper Science Policy Council. U.S. Environmental Protection Agency, Washington, DC. EPA 100/B-07/001.

Gao F, Hong F, Liu C, Zheng L, Su M, Wu X, Yang F, Wu C, Yang O. 2006. Mechanism of Nano-anataseTiO_2 on promoting photosynthetic carbon reaction of spinach.Biological Trace Element Research,**111**,239-253.

Gupta PK. 2006. Soil, plant, water and fertilizer analysis.Agrobios.India, 84 p.

Helmke PA, Sparks DL. 1996.Lithium, Sodium, Potassium, Rubidium and Cesium.In: A. L. Page & SparksD. L. (3rd Edition) Methods of Soil Analysis. Part 3. Soil Sci. Soc. America. No. 5. Madison, WI, USA. 551-601 p.

Kulkarni SK. 2007. Nano technology: Principles and Practices. Capital Pub. Co. New Delhi, India.

Lee WM, Kwak JI, An YJ. 2012. Effect of Silver nano particles in crop plants *Phaseolus radiates* and *Sorghum bicolor*; Media effect on phtotoxicity.Chemosphere **86**,492-499.

Lin A, Xing B. 2008. Root uptake and phytotoxicity of ZnO nano particles. Environmental Science and Technology,**42**,5580-5585. http://dx.doi.org/abs/10.1021/es800422x

Liu XM, Zhang FD, Zhang SQ, He XS, Feng R, Wang W. 2005. Response of peanut to nano-calcium carbonate. Plant Nutrition and Fertilizer Science **11**,3-9.

Lu CM, Zhang CY, Wen JQ, Wu GR, Tao MX. 2002.Research of the effect of nanometer materials on germination and growth enhancement of *Glycine max* and its mechanism. Soybean Science**21**,168. http://dx.doi.org/10.1002/etc.58/references

Ma Y, Kuang L, He X, Bai W, Ding Y, Zhang Z, Zhao Y, Chai Z. 2010. Effect of rare earth oxide nanoparticles on root elongation of plants. Chemosphere,**78**,273-279. http://dx.doi.org/10.1002/jpln.201200016/references

Mirzajani F, Askari H, Hamzelou S, Farzaneh M, GhassempourA. 2013.Effects of silver nano particles on *Oryza Sativa* L. and its rhizosphere bacteria. Ecotoxicology and Environmental Safety,**88**,48-54.

Mousavi SR, Rezaei M. 2011.Nano technology in agriculture and food production. Journal of Applied Environmental and Biological Sciences **1(10)**,414-419.

Moghadam A, Vattani H, Baghvaei N, Keshavarz N. 2012.Effect of different levels of fertilizer nano-Iron chelates on growth and yield characteristics of two varieties of spinach. Research Journal of Applied Sciences, Engineering and Technology **4(12)**,4813-4818.

Olsen SR, Cole CV, Watanabe FS, Dean LA. 1954. Estimation of available phosphorus in soils by extraction with sodium bicarbonate. U.S. Dep. of Agric. Circ. 939.

Quardos ME, Mar LC. 2010. Environmental and human health risks of aerosolized silver nano particles. Journal of the Air & Waste Management Association. **60**,770-781.

Richards LA.1954. Diagnosis and improvement of saline and alkali soils. USDA Agric. Handbook 60. Washington, D.C. http://dx.doi.org/10.1029/GM108p0197/references

Shah V, Belozerova I. 2009. Influence of metal nanoparticles on the soil microbial community and germination of lettuce seeds.Water, Air and Soil Pollution, **197**,143-148.

Sharon M, Choudhary A, Kumar R. 2010.Nanotechnology in Agriculture disease and food safety. Journal of Phytology **2(4)**,83-92. http://dx.doi.org/10.1.1.310.174

Sheykhbaglou R, Sedghi M, Shishevan MT,Sharifi RF. 2010.Effect of nano-iron oxide particles on agronomic traits of soybean.NotulaeScientiaBiologicae**2(2),** 112-113.

Smil V. 2001.Enriching the earth: Fritz Haber, Carl Bosch, and the transformation of world food production.The MIT Press, Cambridge, MA, USA.

Steel RGD, Torrie JH, Dickey DA. 1997. Principles and Procedure of Statistics; A Biological Approach.3rd Edition. McGraw Hill Book Inc., New York. USA. http://dx.doi.org/10.1002/0471476471.refs/pdf

Stewart WM, Dibb DW, Johnston AE, Smyth TJ. 2005. The contribution of commercial fertilizer nutrients to food production. Agronomy Journal, 97: 1-6.

Vendrell PF, Zupancic J.1990. Determination of soil nitrate by transnitration of salicylic acid. Communications in Soil Science and Plant Analysis, **21,**1705-1713. http://dx.doi.org/10.1002/hyp.1046/pdf

Wang X, Hai-xing S, Qiang L, Xiang-min R, Jian-wei P, Gui-xian X, Zhen-hua Z ,Shu-juan W. 2011. Effects of nano-preparation coated nitrogen fertilizer on nutrient absorption and yield of early-rice. Hunan Agricultural Sciences**11,**1-4.

Wheeler S. 2005. Factors influencing agricultural professional's attitudes toward organic agriculture and biotechnology. Center for Regulation and Market Analysis, University of South Australia.

Wu M, Ruo-chao H, Xiao-hai T, Xiao-ling W, Guo-hui M, Hai-tao T. 2010.Effect of Adding Nano-carbon in slow release fertilizer on grain yield and nitrogen use efficiency of super hybrid rice. Hybrid Rice **4,**1-5.

Yang F, Liu C, Gao F, Su M, Wu X, Zheng L, Hang F, Yang P. 2007. The improvement of Spinach Growth by Nanoanatase TiO_2 treatment is related to Nitrogen Photoreduction. Biological Trace Element Research **119,**77-88.

Yin L, Cheng Y, Colman BP, Auffan M, Wiesner M, Rose J, Liu J, Bernhardt ES. 2011. More than the ions: The effect of silver nano particles on *Loliummultiflorum*. Environmental Science and Technology **45,**2360-2367.

Zhang F, Wang R, Xiao Q, Wang Y, Zhang J. 2006. Effect of slow control release fertilizer cemented and coated by nano-materials on plants. Nanoscience **11(1),**18-26.

Zheng L, Liu D, Lu FS,Liu C. 2005.Effect of Nano TiO_2 on strength of naturally aged seed and growth of spinach. Biological Trace Element Research, 104:83-91.

Zia MS, Gill MA, Aslam M, Hussain MF. 1991. Fertilizer use efficiency in Pakistan. Progressive Farming **11,**35-38.