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Study and Identification of Environmental Nanoparticles in Governors State University (GSU's) Agricultural Fields

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**STUDY AND IDENTIFICATION OF ENVIRONMENTAL NANOPARTICLES IN
GOVERNORS STATE UNIVERSITY (GSU's) AGRICULTURAL FIELDS**



DEPARTMENT OF ANALYTICAL CHEMISTRY

GOVERNORS STATE UNIVERSITY

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ABSTRACT

Main purpose of this project is to study nanoparticles in Governors State University's agricultural field and design experimental protocol for the same. Nanoparticles have one dimension that measures between 1 - 100 nanometers or less. Nanoparticles are widely used in day to day life such as in medicines, textiles, manufacturing and cosmetics. Due to their wide use, they can enter environment through many ways such as accidental spills during handling. Waste water treatment plant disposal serves as a main source of their entrance into the environment.

Analysis of nanoparticles in environmental samples presents a number of challenges, including separation and collection of individual nanoparticles from bulk soil. Characterization of nanoparticles requires advanced analytical and spectroscopic techniques.

Literature reviews gave various detection techniques for nanoparticles which helped in identifying promising technologies for future GSU research. Literature review also focuses on soil bacteria such as Streptomyces and Rhizobiales being susceptible to titanium dioxide (TiO₂) and zinc oxide (ZnO) nanoparticles. Recent study performed on effect of ZnO and cerium oxide (CeO₂) nanoparticles on soybean plants is also included. Effect of metal based nanoparticles in soil and soil invertebrates have also been reviewed [5], [6], [31-35].

PURPOSE:

Main aim of this literature project is to design experimental protocol for study of nanoparticles in Governors State University's (GSU's) agricultural field. Selection of nanoparticles for study was made on basis of engineered nonmaterial becoming an emerging key environment contaminant. Another reason for this study is that possible health effects to humans have been proposed when coming in direct or indirect contact with nanoparticles entering environment. In this way, it could be studied under four different categories such as detection of nanoparticles in environment, measuring the amount of nanoparticles entered, life-cycle of nanoparticles in environment and toxicity of nanoparticles and their long term impact onto environment. This literature project mainly focuses on study detection and measurement techniques of nanoparticles present in environmental soil.

CONTEXT:

Due to wide use of nanoparticles in manufacturing eyeglasses, paints, wall coating and sunscreens, they can enter the environmental system through many ways such as industries discharging waste water into streams and rivers, accidental release while handling or production. Release into the environment can occur and domestic waste water discharge is also a possible source through use of nanoparticles in sun creams, cosmetics and toiletries.

Nanoparticles entering the environment may not be initially toxic to environment but may become gradually entering the chain process such as moving between water and sediments or leading to absorption of it into water.

Types of nanoparticles found in agricultural fields and their toxicological study with soil

invertebrates: Most commonly found nanoparticles in agricultural fields are:

1) Zinc Oxide:

Zinc oxide is the most studied nanoparticle for ecotoxicological study. Genetic and physiological effects were observed in earthworm *Eisenia fetida* [1].

2) Silver:

Silver nanoparticles are mainly used as bactericides; they can induce toxicity through combination effects of both dissolved ions and particles. Silver nanoparticles caused the formation of reactive oxygen species which induce toxicity, although other mechanism is also involved [2].

3) Gold:

Effects of gold nanoparticles on reproduction of earthworms were observed although no effects were found on growth or survival of them [3].

TECHNIQUES TO MEASURE AND DETECT NANOPARTICLES FROM SOIL

SAMPLES:

Soil contains many types of organic and inorganic particles among which at least one particle has nanoscale dimension range. Thus, only a small proportion of nanoparticles in soil exist as discrete entities. Organic colloids in soil are largely associated with their inorganic counterparts, due to which it is difficult to separate and collect individual nanoparticles from bulk soil and moreover, extraction yields are also low. Due to this reason, characterization of nanoparticles requires advanced analytical and spectroscopic techniques.

Here are few mentioned techniques to detect nanoparticles from soil samples. Their use includes both for determination of physical properties as well as for determination of nanoparticles identity and quantitative concentration.

1) Scanning mobility particle size (SMPS):

Scanning mobility particle sizer technique is used in measuring aerosols and can be applied to measure nanoparticles in gas phase. Therefore, SMPS is very difficult to apply for biological samples without complex sample preparation [4].

2) Field-flow fractionation (FFF), size exclusion (SEC) and hydrodynamic chromatography (HDC):

Field flow fractionation is a mild chromatography technique which does not utilize stationary phase. It separates nanoparticles based on their diffusion coefficients in a very thin open channel. This separation relies on combination of longitudinal carrier flow and applied field (magnetic or dielectric). The field acts perpendicular to separation channel and causes nanoparticles to move towards accumulation wall. This leads to formation of nanoparticles cloud whose thickness is given by ability of particles to oppose the force of the field. To monitor size distribution, FFF can be coupled with detector such as UV absorbance, ICP-MS, ESI-MS, and CI-MS [5].

In Size exclusion chromatography, the particle mixture is passed through the column filled with porous packing with distribution of pore sizes in the range of particles to be fractionated.

Particles are separated according to their hydrodynamic volume [6].

Hydrodynamic chromatography method is carried out in narrow open or wider capillaries with non-porous packing due to which it form capillary routes. Here separation is based upon hydrodynamic volume same as in SEC [7].

3) Chromatographic and Spectroscopic analyses of nanoparticles:

Techniques such as UV-Vis spectroscopy, infrared spectroscopy, nuclear magnetic resonance and mass spectrometry coupled with high performance liquid chromatography (HPLC) are commonly used.

Fluorescence Spectroscopy:

Fluorescence spectroscopy is used in detection and identification of fluorescent nanoparticles. Its work was done on Cadmium telluride (CdTe) / Zinc Sulphide (ZnS) quantum dots with surface modified with amino groups. Auto fluorescence of samples can be removed by treatment with chemical mixtures [8].

Atomic emission spectroscopy (AES):

Laser – induced breakdown detection (LIBD) is a type of sampling for AES. In this type, excitation of emission spectrum of explored substance is carried out by short sharply focused laser pulse. LIBD in complex with FFF, atomic force microscopy (AFM) and mass spectrometry (MS) was applied successfully during characterization of bentonite nanoclay dispersion [9].

Raman spectroscopy:

Raman spectroscopy is based on inelastic scattering of optical radiation on molecules followed by change in frequency. Raman spectrometry permits revealing of nanoparticles in complex systems such as carbon nanotubes in organs of mice [10].

Correlated Rayleigh scattering spectroscopy (CRSS):

Correlated Rayleigh scattering spectroscopy was used for gold and silver nanoparticles characterization of sizes more than 30 nm and results obtained were confirmed with use of scanning electron microscopy (SEM) [10].

UV-Vis spectroscopy:

UV-Vis spectroscopy is an absorption spectroscopy and it uses light in visible and near region (near UV and near infrared). Absorption of light is related to concentration of chemical involved. Wavelength of absorption is specific for each type of particles [11].

Infrared Spectroscopy:

Infrared spectroscopy deals with infrared region of electromagnetic spectrum. It can analyze sample in three ways by measuring absorption, emission and reflection. Wavelength of absorbing IR radiation is specific for each type of functional groups [12].

Nuclear Magnetic Resonance (NMR):

Nuclear magnetic resonance is a research technique used to exploit magnetic properties of a molecule. It occurs when nuclei of certain atoms are immersed in static magnetic field and exposed to second oscillating magnetic field. It is applied to determine silica particles in aerosols and in water dispersion [12].

Mass Spectrometry (MS) and Inductively Coupled Plasma (ICP):

Mass spectrometry measures mass to charge ratio of a sample. It is used in determining elemental composition of a sample. It also helps in elucidating chemical structure of unknown

molecule. For biological samples, matrix assisted laser desorption / ionization (MALDI) and electrospray ionization (ESI) can be applied. Ionization of metal containing nanoparticles can be done using inductively coupled plasma (ICP). Usually for nanomaterial analysis, FFF in combination with ICP-MS is used which allows fractionation of molecules by sizes and determines chemical composition of separate fractions [12], [13].

High performance liquid chromatography (HPLC):

High performance liquid chromatography is chromatographic technique used to separate mixture of compounds with purpose of identifying, quantifying and purifying components of mixture [14].

4) Light Scattering techniques:

Light scattering methods are commonly used method to determine particle size. In this technique, electromagnetic radiation of incident photons induces an oscillating dipole in particle electron cloud. As dipole changes, radiation is scattered in all directions. Light source could be X-rays, laser light or neutron [15].

Dynamic light scattering (DLS):

In dynamic light scattering technique, fluctuations in scattered light depending on particle diffusion are utilized. These fluctuations originate from Brownian motion of particles and also depend on whether neighboring particles have constructive or destructive interference of the scattered light intensity [16].

Static light scattering (SLS):

Static light scattering is also called multi angle light scattering and it provides measurement of physical properties derived from angular dependency of light scattered by a particle [16].

Nephelometry:

Nephelometry is a particle concentration measurement which utilizes scattering of light at 90° or 180° with respect to light source. Light source could be laser or monochromatic light. Light scattering intensity depends on refractive index difference between particles and particle size [16].

5) Electron microscopy and atomic force microscopy:

Microscopic methods are usually single particle method [17], [18].

Table 1: Analytical Techniques for nanoparticles identification

Techniques	Samples	Limit of Detection	Type of nanoparticle
Inductively coupled plasma-Mass spectrometry (ICP-MS)	Ionization of sample with inductively coupled plasma; digestion in weak mineral acids may be required	ppt – ppb	Silver and gold nanoparticles [5, 6]
Electrospray Ionization – Mass spectrometry (ESI-MS)	Liquid samples of 1-20 pmol/μL required	ppb	Gold nanoparticles [5]
Transmission Electron Microscopy (TEM)	< 1μg sample in form of thin film and should be stable under electron beam and high vacuum	ppb – ppm	Gold and platinum nanoparticles [6]
Scanning Electron Microscopy (SEM)	Must be conductive or sputter coated	ppb – ppm	Gold nanoparticles [6]
Photon Correlation Spectroscopy (PCS)	Must be a very dilute suspension	1nm – 10μm	Gold nanoparticles [6]
X-Ray Diffraction (XRD)	Large crystalline sample (> 1 mg) required	Down to 1nm	Zinc oxide and silver nanoparticles [4, 19]
Aerosol Time of Flight Spectroscopy	Aerosol	100-3,000 nm	Aerosol nanoparticles like

			TiO ₂ and silicon dioxide[20]
Nanoparticle Tracking Analysis (NTA)	500 µL of suspension required	10-1,000 nm	Silver and polystyrene [5]
Atomic Force Microscopy (AFM)	Sample must adhere to substrate	ppb – ppm	Fullerenes [21]
Condensation particle counter (CPC)	Aerosol, can have high temp up to 200 °C, 0 to 100,000 particles/cm ³	2.5 to >3,000 nm	Nucleated nanoparticles like rhodium and palladium nanoparticles [22]

Critical Reviews: Here two critical literature reviews are considered:

I. Metal based nanoparticles in soil: fate, behavior and effects on soil invertebrates

Authors: Paula S. Tourinho, Cornelis A.M. Van Gestel, Stephen Lofts, Claus Svendsen, Amadeu M. V. M. Soares and Susana Loureiro.

Date accepted and location: 19 March 2012; Wallingford, Oxfordshire, United Kingdom.

Focus: This review article mainly focuses on the literature dealing with fate and effects of metal based nanoparticles in soil. It is based on consideration of nanoparticles released in the soil through land application of sewage sludge derived from waste water treatment. It also focuses on how nanoparticle affects environmental characteristics such as size, shape and surface charge and the soil characteristics such as pH, ionic strength, organic matter and clay content. This may lead to nanoparticle dissolution, agglomeration and aggregation.

Strategy:

1) Sources of metal based nanoparticles in soil:

Metal and metal oxide based nanoparticles are widely used in many products such as sunscreens and cosmetics (ZnO and TiO₂), printer inks (Ag and Au) and textiles (Ag). Their use also include in electronics, medicine and chemistry. Due to their diverse use, nanoparticles can enter into environmental system through different ways such as accidental spills during industrial transportation and production, through their use in soil and water remediation technologies and agriculture. Their emission to environment may lead to deposition in soils and waters from waste incineration.

However, disposal of wastewater treatment plant (WWTPs) sewage sludge is considered main source of nanoparticle disposition onto land. For example, disposal of Ag nanoparticles can be released during washing of undergarments [23] and release of titanium dioxide from paints [24]. Nanoparticles are unlikely to enter into soil in their original form due to original rich and reactive environment of WWTPs. Some authors have suggested that use of sewage sludge could contribute to input of 1 μg/Kg³ and 120 μg/Kg³ of Ag and TiO₂ nanoparticles to agricultural land per year respectively [25].

2) Behavior of metal based nanoparticles in soil:

Physical and chemical characteristics of the metal will determine the extent to which nanoparticles would undergo transformations such as agglomeration, sorption to the surface and dissolution to ionic metal. Existence of these characteristics implies characterization of nanoparticle form and presentation in the environment would be considered as a key in

understanding nanoparticle's behavior, fate and ecotoxicity. Nanoparticles may react with complex matrix present in soil.

Assessment of the form and presentation of nanoparticles in environmental matrices is hampered due to lack of appropriate procedure for their characterization.

The review suggests properties to be evaluated in ecotoxicological studies should be dispersability, agglomeration, dissolution rate, size, surface area, charge and surface chemistry.

Agglomeration and aggregation:

Nanoparticles usually have property of agglomeration and aggregation. Aggregation is association of primary particles by strong bonding and agglomeration means association through weak Vander Waals forces which results in weak bonding. Nanoparticles aggregate size in solution depends on properties such as initial size and concentration.

Using iron nanoparticles, it is found that higher concentrations (60mg/L) resulted in higher aggregation rates while with lower concentration (2mg/L) aggregate size stability has been found [26].

Zinc oxide nanoparticles being dispersed in aqueous solution resulted in formation of 10 fold larger aggregates than primary nanoparticles [21, 4].

Distribution of aggregates depends upon type of particle. For example, TiO₂ particles show uniform distribution and agglomeration while ZnO particles show wide distribution size and agglomeration [21].

Toxicity and bioaccumulation of metal based nanoparticles

Ecotoxicological tests to determine toxicity of nanoparticles to soil organisms can be conducted with different exposure media such as soil, water and food. When looking at the effects of nanoparticles and comparing them with their non-nano counterparts, different mechanism of action and effects may be expected. Due to this, it is necessary to carefully select an endpoint of the toxicity test.

The goal of toxicity testing is to help in understanding nanoparticle behavior used in standardized media. “The process of bioaccumulation of metals in soil organisms and their biomagnification is well known in literature” [27].

Bioaccumulation and biomagnification of gold (Au) nanoparticles of three different sizes were evaluated with hornworms through food consumption of tobacco leaves [28]. Authors found that nanosized Au particles caused biomagnification and not Au soluble. This process was found to be related to zeta potential rather than particle size because biomagnification was found with Au nanoparticles having higher zeta potential but with different sizes (10 and 15 nm) [28]. Some metal based nanoparticles (e.g., TiO_2) are found to accumulate in soil due to their low to negligible dissolution [29]. Kinetics experiments showed that nanoparticles tend to be more stable after forming aggregates and thus may result in occurrence of biomagnification [30].

The outcome for toxicity study in relation to soil invertebrates is to determine effective concentration (EC) and lethal concentration (LC) of nanoparticle for particular invertebrate.

Table 2: Table showing an overview of available literature data about toxicity of metal based nanoparticles (NPs) to soil invertebrates:

NPs	Initial size	Species tested	Concentration	Exposure media	Duration	End points	Outcomes	Ref.
Ag	20-30 nm	Caenorhabditis elegans	1 mg/L	K-media (Potassium ion media)	24 h and 72 h	Survival, growth and reproduction		[31]
Ag	10 nm and 30-50 nm	Eisenia fetida	10-1000 mg/Kg dry soil	Artificial and natural soil	28 d	Survival, growth and reproduction	EC50= 8.7 mg/Kg	[5]
Ag	50.3 nm	Caenorhabditis elegans	10-1000 mg/L	Agar and K-media	24 h and 48 h	Survival and reproduction	LC50=55 mg/L and EC50 = >100 mg/L in agar	[32]
Au	20 nm and 50 nm	Eisenia fetida	5-50 mg Au/Kg dry mass	Artificial soil	28 d	Survival, reproduction and gene expression		[6]
CeO ₂	15 and 45 nm	Caenorhabditis elegans	1 mg/L	K-media	24 h	Survival, reproduction and gene expression		[33]
Cu	80 nm	Enchytraeus albidus	400-1000 Cu/Kg	Field soil	48 h	Gene expression		[34]

Cu	80 nm	Enchytraeus albidus	130 and 230 mg Cu/Kg	Field soil	3 and 6 weeks	Energetic reserves	EC10 = 97 mg Cu NP/Kg	[35]
TiO ₂	50 nm	Caenorhabditis elegans	24.0-239.6 mg/L	Water	24 h	Survival, growth, reproduction and feeding	LC50 = 80 mg/L	[4]
ZnO	20 nm	Caenorhabditis elegans	0.4-8.1 mg/L ZnO-NPs	Water	24 h	Survival, growth, reproduction and feeding	LC50 = 2.2 mg/L	[4]
ZnO	<100 nm	Eisenia veneta	6-96 mg Zn/L DI water	Deionize (DI) water	24 h	Survival	LC50 = 1.75 mg/L	[4]

Conclusions:

- 1) Possibility of nanosized particles assimilation is indicated by presence of nanoparticles in organisms' tissue [6, 36].
- 2) Toxicity is clearly influenced by nanoparticles core elements; for example, TiO₂ was found to be less toxic than Ag [37] and ZnO nanoparticles [38].
- 3) Tests should be conducted in media close to realistic conditions to enable better understanding and have relevance understanding of nanoparticle's toxicity in the environment. For example: potassium ion media (K-media) leads to greater aggregation and is more similar to pore water conditions than agar [39].

- 4) Care should be taken while interpreting non-soil media such as liquids or agar for testing because their novel modes of action such as reactive oxygen species generation (ROS) for TiO₂, ZnO and CeO₂ [35] can lead to effects that are not of major relevance in soil organisms.

II) Quantitative Analysis of Fullerene Nanomaterials in Environmental Systems: A Critical Review

Authors: Carl W. Isaacson, Markus Kleber and Jennifer A. Field.

Date Accepted: July 14, 2009 and published at Oregon State University, Corvallis, Oregon.

Focus: This review was motivated by increased use of fullerene nanomaterials and their potential impact on human and environmental health. Fullerene nanomaterials are comprised of both fullerenes and surface-functionalized fullerenes, and are used in medical, cosmetic and optic applications. Fullerenes exhibit both physical and chemical characteristics as they transition from hydrophobic to polar (hydrophilic) forms upon exposure to water. Due to this, measuring fullerene concentration in environment is difficult. In aqueous environment, fullerenes have tendency to self-assemble into aggregates of appreciable size and hydrophobicity and acquire negative surface charge by interacting with surrounding water molecules and other chemical constituents. Fullerene nanomaterials may therefore be not limited to the application of any single analytical method [40].

This article includes following findings:

- 1) Analytical procedure needed to detect fullerenes in natural environments through use of approaches that provide information including molecular weight and number and identity of surface functional groups.
- 2) Mass spectrometry combined with modified extraction procedures leads to detection of sensitive and mass sensitive mass.
- 3) Significant improvements in analytical procedures would result from increased availability of standards, reference materials and internal standards.

The central objective of this literature review is to provide:

- 1) “State-of-the-art analytical methods” report for quantifying fullerene nanomaterials.
- 2) An exploration of specific challenges to analytical methods that must be overcome.
- 3) A discussion on how improvement in analytical methods plays an important role in understanding occurrence, fate, transport and effects of fullerene nanoparticles.

Fullerene Analysis: State-of-the-Art

For this review purpose, more focused is quantitative methods that can potentially differentiate between different molecular forms of fullerenes (for eg: C₆₀ from C₇₀, C₆₀ from C₆₀O).

Analytical methods comprise of multi-steps such as extraction, concentration, separation and detection and applied to sample in same linear fashion.

Sample Extraction:

Methods for fullerenes extraction from environmental solids including sediments and combustion-derived soots involve sonication or Soxhlet extraction with toluene. Extraction of C₆₀ from biological matrices such as plasma and homogenized tissue also utilizes toluene while

water soluble C₆₀ are not efficiently recovered from aqueous solution using toluene alone due to their polar nature. Alternative methods include use of salt along with toluene which follows evaporation to dryness process ending to solid phase extraction (Table 3).

Separation:

Table 3: Current Quantitative Methods for Fullerene Nanomaterials.

Species	Matrix	Extraction	Separation/detection and internal standard (IS)	Recovery \pm Precision	Detection/Quantification limits ($\mu\text{g/L}$)	Reference
C ₆₀ -C ₉₆	Carbon soot	Toluene extraction	LC-UV	10-162 \pm NR%	NR	[41]
Water stable C ₆₀ (from THF)	Skin permeation media (5 % bovine serum albumin-BSA) plasma	Liq:liq extraction with toluene and salt	LC-UV	94-100 \pm 15%	0.34 (aq. With BSA), 0.68 (plasma)	[42]
C ₆₀ (prepared in DMSO)	Embryonic zebrafish homogenate	Liq:liq extraction with toluene	LC-ESI-MS with ¹³ C ₆₀ as IS	90 \pm 3%	0.02 (detection) 0.04 (quantification)	[43]
C ₆₀ (prepared in toluene and injected in mice)	Blood Spleen Liver	Liq:liq extraction with toluene and 0.1 M sodium dodecyl sulfate	LC-UV	90-103 \pm 0.8-6% 100-104 \pm 3-8% 96-102 \pm 2-8 %	0.02 (detection)	[44]

NR – not reported; LC-UV – liquid chromatography-ultraviolet; LC-ESI – liquid chromatography-electrospray ionization.

Detection:

Analytical methods currently used that possess ability to differentiate between molecular forms of fullerenes include liquid chromatography with ultraviolet (UV) detector [45, 46] and ¹³C

NMR [45]. A mass selective detection method such as MS is found to be more sensitive than above mentioned methods. Mass spectrometry separates various forms of fullerenes based on their molecular weight and thus provides structural information which includes functional group present. Mass spectrometry is usually combine with various ionization techniques such as laser desorption ionization (LDI), matrix assisted laser desorption ionization (MALDI) [47], chemical ionization [48] and electrospray ionization [49].

Current Limitations and Research Opportunities:

This segment would include limitation associated with

- 1) Extraction
- 2) Separation
- 3) Detection of fullerenes and surface-functionalized fullerenes in complex environmental matrices.

1) Extraction:

Literature review states that “even though endogenous concentration of fullerenes from geologic samples [50, 51] and combustion derived solids [52, 42] are mentioned, only one attempt of extracting fullerene was optimized and less than 5% was recovered”[52]. No optimized methods for extraction of fullerenes from carbon soots are reported [53]. Extraction of a series of fullerenes existing in environment using single solvent is not suitable because larger fullerenes (e.g. $C_{76} - C_{94}$) can be extracted using 1,2,4-trichlorobenzene [54] and smaller fullerenes, such as C_{36} are not soluble in toluene but are rather soluble in pyridine and carbon disulfide [53].

The conventional method that was used was to increase temperature of solvent to maximize analyte extraction. Most fullerenes are extracted more efficiently at higher temperature, while

C₆₀ exhibit variable solubility from temperature 3 to 37° C depending on solvent used, however, it decreases with further increase in temperature [53].

2) **Separation:**

Current challenges faced in separating various forms of fullerenes are the development of methods having ability to differentiate between hydrophobic and polar fullerenes forms and from those having surface functional groups. The common method for chromatographic separation of fullerenes includes addition of polar solvent to toluene extract which not only dilutes the fullerenes but also affects the sensitivity. As per work done by Isaacson et al. [55], “up to 70 % of final volume consist of polar solvent to retain and separate fullerenes on C₁₈ column” [44]. Due to wide range of molecular weights and polarities, more specific stationary phases and mobile phases are required to separate individual species of fullerene nanomaterials.

Development of alternative columns [56] will reduce the amount of solvent needed for separation [57]. But minimizing the dilution of toluene extract would result in lower detection limits.

3) **Detection:**

Raman spectroscopy [58, 20] and ¹³C NMR methods are not found suitable for routine analysis. However, mass spectrometry helps in identifying fullerenes based on their molecular weight and provides structural information indicating presence or absence of functional groups. MALDI, fast atom bombardment (FAB), LDI, thermal desorption ionization (TDI) and CI ionization techniques provides molecular identity of fullerene nanomaterials, but utility of these ionization methods is limited for quantitative analysis because these ionization techniques are not compatible with separation instrumentation such as liquid chromatographs. Due to this reason,

ionization modes such as atmospheric pressure chemical ionization (APCI), atmospheric pressure ionization (APPI) and electrospray ionization (ESI) that are easily interfaced with liquid chromatographs are recommended [20].

UV is not found capable of distinguishing between fullerenes and surface-functionalized fullerenes and cannot identify peak purity for complex mixtures present in environmental matrices.

Fullerene Occurrence, Fate, Transport, Effects and the Role for Analytical Chemistry:

Literature review states “There is no data on naturally occurring fullerenes in biota or water from natural or engineered systems” [59]. The earliest work done on determining natural occurrence and concentration of fullerenes was in geological sciences. The occurrence, concentration and species of fullerenes present in combustion derived solids such as diesel exhaust and black carbon provides information on natural background levels and on human exposure to fullerenes.

Very little is known about the fate of fullerenes in natural and engineered systems. One study was conducted on fullerene decomposition by fungi [45]. Recent studies are conducted on fullerenes reaction with ozone and UV light. Reaction of water stable C₆₀ with ozone leads to reduction of parent molecule and leads to formation of transformation products which are identified by NMR, IR, MS and XPS methods [60]. Reports on increased mass of suspended C₆₀ suspended in water upon exposure to UV light [60] are documented. Techniques such as FT-IR, XPS and MS were used to document reduction in masses of fullerenes. Role of oxygen was in degradation, however, no degradation products were found, and only oxidized products were identified.

New release article:

Title and location: Effects of nanoparticles on soybean crops and research was conducted by Scientist at UC Santa Barbara's Bren School with UC center for environmental implications of nanotechnology, University of Texas, Columbia University and other international universities.

Date Published: August 20, 2012

Study Conducted:

Cosmetics containing small metal particles usually leads to passage of nanomaterials into drainage water while washing and manufacturing which eventually end up getting into agricultural soil. This was a cause of concern for scientist that conducted study on soybean plants grown in nanoparticle contaminated agricultural field. Two common nanoparticles found in agricultural soil are zinc oxide (ZnO) and cerium dioxide (CeO₂) [61].

Soybean crop was chosen for study since it is major global community crop grown in US agricultural field. Scientists found out that ZnO nanoparticles may dissolve or remain as particle after they processed through waste water treatment. At the end of waste water treatment, biosolids which is applied to soil is found. These solid material called biosolids fertilize the soil, returning nitrogen and phosphorus that are captured during waste water treatment. At this point, ZnO and CeO₂ nanoparticles enter the soil [61].

Conclusion: Literature review concluded some very important points:

Soybean, a major global community crop was amended with two high production metal oxides (nano CeO₂ and -ZnO). Following facts can be concluded [61]:

1. For nano ZnO, component metal was taken up and distributed throughout edible tissues.
2. For nano CeO₂, plant growth and yield was diminished.

3. Nitrogen fixation was shut down with higher concentration of CeO₂.
4. Ecotoxicological studies showed genetic and physiological effects in earthworm *Eisenia fetida* with ZnO nanoparticles.
5. Ag nanoparticles are found to induce toxicity through formation of reactive oxygen species.
6. Au nanoparticles were found to have effect on reproduction of earthworms although no effects were found on growth or survival.

CONCLUSION:

This study demonstrated characteristics of nanoparticles being dynamic in environment and thus is an important consideration for design and interpretation of nanoparticle studies.

Among methods available to detect nanoparticles, I would suggest ICP-MS as best technique for detection.

Microscopic techniques become ineffective when it comes to identification of small amount of nanoparticles and here, an analytical technique such as ICP-MS plays an important role. ICP-MS is very sensitive and quantitative tool in determining trace amount of nanoparticles.

Ionization of nanoparticles is performed by inductively coupled plasma and separation of ions is done using mass analyzer such as ion trap and time of flight analyzer. MS plays a role in differentiating ion based on their mass to charge ratio and detecting the individual fragments and providing molecular weight and structural information.

Even single particle ICP-MS can be used for nanoparticle determination as it eliminates many matrix interferences, but it does not determine particle size. Moreover, it can be used to detect

metals in nanoparticles at concentration as low as 1 part per trillion. Interpretation of ICP-MS results is easy as final outcome is of mass concentration.

Thus, ICP-MS can be used to detect various nanoparticles such as AuNPs and AgNPs which are found mostly in agricultural fields and is considered best tool for nanoparticle detection.

References:

- 1) Wang H., Wick R.L., Xing B. (2009). Toxicity of nanoparticulate and bulk ZnO, Al₂O₃ and TiO₂ to the nematode *Caenorhabditis elegans*. *Environ Pollut* 157:1171-1177.
- 2) Shoultz-Wilson W. A., Reinsch B. C., Tsyusko O. V., Bertsch P. M., Lowry G. V., Unrine J. (2011). Role of particle size and soil type in toxicity of silver nanoparticles to earthworms. *Soil Sci Soc. Am* 75:365-377.
- 3) Unrine J. M., Hunyadi S. E, Tsyusko O. V., Rao W., Shoultz-Wilson W. A., Bertsch P. M. (2010). Evidence for bioavailability of Au nanoparticles from soil and biodistribution within earthworms (*Eisenia fetida*). *Environ Sci Technol* 44:8308-8313.
- 4) Balnois E., Wilkinson K.J. (2002). Sample preparation techniques for the observation of environmental biopolymers by atomic force microscopy. *Colloids Surf Physiochem Eng Aspects* 207:229-242.
- 5) Dubascoux S., Hecho I., Hassell V. M., Kammer F., Gautier M., Lespes G. (2011). Field-flow fractionation and inductively coupled plasma mass spectrometer coupling: History, development and applications. *Journal of Analytical Atomic Spectrometry* 25:613-623.
- 6) Barth H. G., Boyes B. E. (1992). Size exclusion chromatography. *Anal Chem* 64:428R-442R.
- 7) Barth H. G., Flippen R.B. (1995). Particle size analysis. *Anal Chem* 67:257R-272R.
- 8) K. Schmid and M. Riediker, *Environ. Sci. Technol.* 42 (7), 2253. (2008).

- 9) Ebdon L., Foulkes M., Sutton K. (1997). Slurry nebulization in plasmas. *J Anal Atom Spectrom* 12:213-229.
- 10) I. V. Safenkova, S. M. Pridvorova, A. V. Zherdev, et al., Abstracts of III International Forum on Nanotechnologies (Moscow, 2010).
- 11) Andrievsky G. V., Klochkov V. K., Bordyuh A. B., Dovbeshko G. I. (2002). Comparative analysis of two aqueous-colloidal solutions of C-60 fullerene with help of FTIR reflectance and UV-vis spectroscopy. *Chem Phys Lett* 364:8-17.
- 12) Santos M. C., Nobrega J. A. (2006). Slurry nebulization in plasmas for analysis of inorganic materials. *Appl. Spectroscop Rev* 41: 427-448.
- 13) Allabashi, R., Stach, W., de la Escosura-Muniz, A., Liste-Calleja, L. and Merkoci, A. (2009). ICP-MS: a powerful technique for quantitative determination of gold nanoparticles without previous dissolving. *Journal of Nanoparticle Research* 11, 2003-2011.
- 14) Treubig J.M., Brown P.R. (2002). Analysis of C60 and C70 fullerenes using high-performance liquid chromatography-Fourier transform infrared spectroscopy, *J Chromatogr A* 960:135-142.
- 15) Schurtenberger P., Newman M.E. (1993). Characterization of biological and environmental particles using static and dynamic light scattering. In: Buffle J, van leewen HP (eds) *Environmental particles*. Lewis publishers, Boca Raton, Florida, pp 37-115.
- 16) Woo, K.S., Chen, D.R., Pui, D.Y.H., and McMurry, P.H. (2001). Measurement of Atlanta Aerosol Size Distributions: Observation of Ultrafine Particles Events, *Aerosol Science and Technology* 34: 75-87.

- 17)** Viguie J.R., Sukmanowski J., Nolting B., Royer F.X. (2007). Study of agglomeration of alumina nanoparticles by atomic force microscopy (AFM) and photon correlation spectroscopy (PCS). *Colloids Surf Physicochem Eng Aspects* 302:269-275.
- 18)** Scalf J. and West P. (2006). Part I: Introduction to nanoparticle characterization with AFM. Available from:<http://nanoparticles.pacificnanotech.com>.
- 19)** Pipan-Tkalec Z., Drobne D., Jemec A., Romih T., Zidar P., Bele M. (2010). Zinc bioaccumulation in terrestrial invertebrate fed a diet treated with particulate ZnO or ZnCl₂ solution. *Toxicology* 269:198-203.
- 20)** Woo, K.S., Chen, D.R., Pui, D.Y.H., and McMurry, P.H. (2001). Measurement of Atlanta Aerosol Size Distributions: Observation of Ultrafine Particles Events, *Aerosol Science and Technology* 34: 75-87.
- 21)** Scharff, P.; Risch, K.; Carta-Abelmann, L.; Dmytruk, I. M.; Bilyi, M. M.; Golub, O. A.; Khavryuchenko, A. V.; Prylutsky, Y. I.; Durov, S. S. (2004). Structure of C₆₀ fullerene in water: spectroscopic data. *Carbon* ,42 (5-6), 1203-1206.
- 22)** Bianchini, C.; Frediani, P.; Sernau, V. *Organometallics*. (1995).14, 5458.
- 23)** Benn T.M., Westerhoff P. (2008). Nanoparticle silver released into water from commercially available sock fabrics. *Environ Sci Technol* 42: 4133-4139.
- 24)** Kaegi R., Ulrich A., Sinnet B., Vonbank R., Wichser A., Zuleeg S., Simmler H., Brunner S., Vonmont H., Burkhardt M., Boller M. (2008). Synthetic TiO₂ nanoparticle emission from exterior facades into the aquatic environment. *Environ Pollut* 156:233-239.
- 25)** Mueller N.C., Nowack B. (2008). Exposure modeling of engineered nanoparticles in the environment. *Environ Sci Technol* 42:4447-4453.

- 26)** Phenrat T., Saleh N., Sirk K., Tilton R.D., Lowry G.V. (2006). Aggregation and sedimentation of aqueous nanoscale zerovalent iron dispersions. *Environ Sci Technol* 41:284-290.
- 27)** Unrine J., Bertsch P., Hunyadi S. (2008). Bioavailability, Trophic Transfer and toxicity of manufactured metal and metal oxide nanoparticles in Terrestrial environments. John Wiley & Sons, New York, USA.
- 28)** Judy J.D., Unrine J.M., Bertsch P.M. (2011). Evidence for biomagnification of gold nanoparticles within a terrestrial food chain. *Am Chem Soc* 45:776-781.
- 29)** Baun A., Hartmann N., Grieger K., Kusk K. (2008). Ecotoxicity of engineered nanoparticles to aquatic invertebrates. A brief review and recommendations for future toxicity testing. *Ecotoxicology* 17:387-395.
- 30)** French R.A., Jacobson A.R., Kim B., Isley S.L., Penn R.L., Baveye P.C. (2009). Influence of ionic strength, pH and cation valence on aggregation kinetics of titanium dioxide nanoparticles. *Environ Sci Technol* 43:1354-1359.
- 31)** Roh J.Y., Sim S.J., Yi J., Park K., Chung K.H., Ryu D.Y., Choi J. (2009). Ecotoxicity of silver nanoparticles on the soil nematode *Caenorhabditis elegans* using functional ecotoxicogenomics. *Environ Sci Technol* 43:3933-3940.
- 32)** Kim S.W., Nam S.H., An Y.J. (2012). Interaction of silver nanoparticles with biological surfaces of *Caenorhabditis elegans*. *Ecotoxicol Environ Safety* 77:64-70.
- 33)** Gomes S.I.L., Novais S.C., Scott-Fordsmand J.J., De Coen W., Soares M., Amorim M.J.B. (2012). Effect of Cu nanoparticles versus Cu salt in *Enchytraeus albidus* (Oligochaeta); Differential gene expression through microarray analysis. *Comp Biochem Physiol Part C: Toxicol Pharmacol* 155:219-227.

- 34)** Amorim M., Gomes S., Soares A., Scott-Fordsmand J. (2012). Energy basal levels and allocation among lipids, proteins and carbohydrates in *Enchytraeus Albidus*: Changes related to exposure to Cu salt and Cu nanoparticles. *Water, Air, Soil Pollut* 223:477-482.
- 35)** Roh J.Y., Park Y.K., Park K., Choi J. (2010). Ecotoxicological investigation of CeO₂ and TiO₂ nanoparticles on the soil nematode *Caenorhabditis elegans* using gene expression, growth, fertility and survival as endpoints. *Environ Toxicol Pharmacol* 29:167-172.
- 36)** Hooper H.L., Jurkschat K., Morgan A.J., Bailey J., Lawlor A.J., Spurgeon D.J., Svendsen C.(2011). Comparative chronic toxicity of nanoparticulate and ionic zinc to the earthworm *Eisenia veneta* in a soil matrix. *Environ Int* 37:1111-1117.
- 37)** Mcshane H., Sarrazin M., Whalen J.K., Hendershot W.H., Sunahara G.I. (2012). Reproductive and behavioral responses of earthworms exposed to nano-sized titanium dioxide in soil. *Environ Toxicol Chem.* 31:184-193.
- 38)** Canas J.E, Qi B., Li S., Maul J.D., Cox S.B., Das S., Green M.J. (2011). Acute and reproductive toxicity of nano-sized metal oxides (ZnO and TiO₂) to earthworms (*Eisenia Fetida*). *J Environ Monit* 13:3351-3357.
- 39)** Kim S.W., Nam S.H., An Y.J. (2012). Interaction of silver nanoparticles with biological surfaces of *Caenorhabditis elegans*. *Ecotoxicol Environ Safety* 77:64-70.
- 40)** Buseck, P. R.; Tsipursky, S. J.; Hettich, R. (1992). Fullerenes from the geologic environment. *Science*, 257, 215-217.
- 41)** Jinno, K.; Kohrikawa, C. (1998). Supercritical and subcritical fluid extraction of fullerenes from carbon soot. *Chimica OGGI*, 16, 9-15.

- 42)** Xia, G., Monteiro-Riviere, N. A., Jr. (2006). Trace analysis of fullerenes in biological samples by simplified liquid-liquid extraction and high performance liquid chromatography. *J. Chromatogr., A*, 1129, 216-222.
- 43)** Isaacson, C.W.; Usenko, C. Y.; Tanguay, R. L.; Field, J. A. (2007). Quantification of fullerenes by LC-ESI/MS and its application to in vivo toxicity assays. *Anal. Chem.* 79 (23), 9091-9097.
- 44)** Moussa, F.; Pressac, M.; Genin, E.; Roux, S.; Trivin, F.; Rassat, A.; Ceolin, R.; Szwarc, H. (1997). Quantitative analysis of C₆₀ fullerene in blood and tissues by high-performance liquid chromatography with photodiode-array and mass spectrometric detection. *J. Chromatogr., B*, 696, 153-159.
- 45)** Fortner, J. D.; Lyon, D. Y.; Sayes, C.M.; Boyd, A. M.; Falkner, J.C.; Hotze, E.M.; Alemany, L. B.; Tao, Y. J.; Guo, W.; Ausman, K. D.; Colvin, V.L.; Hughes, J.B. (2005). C₆₀ in water: nanocrystal formation and microbial response. *Environ. Sci. Technol.*, 39 (11), 4307-4316.
- 46)** Deguchi, S.; Alargova, R.G.; Tsujii, K. (2001). Stable dispersions of fullerenes, C₆₀ and C₇₀, in water: preparation and characterization. *Langmuir*, 17 (19), 6013-6017.
- 47)** Isaacson C.; Trimpin, S.; Barofsky, D.; Remcho, V.T.; Field J. A. Method development for nanoparticle analysis in biological and environmental samples by LC ESI-MS/MS and MALDI MS. SETAC North America, 27th Annual Meeting, Montreal, Quebec, Canada, 11/5/06-11/9/06; Montreal, Quebec, Canada, (2006).
- 48)** Dunsch, L.; Kirbach, U.; Klostermann, K. (1995). Mass spectrometry-the important characterization method of fullerenes. *J. Mol. Struct.*, 348,381-384.

- 49)** Barrow, M. P.; Feng, X.; Wallace, J. I.; Boltalina, O.V.; Tayler, R.; Derrick, P.J.; Drewello, T. (2000). Characterization of fullerenes and fullerene derivatives by nanospray. *Chem. Phys. Lett.*, 330 (3-4), 267-274.
- 50)** Heymann, D.; Chibante, F.; Brooks, R.R.; Wolbach, W.S.; Smalley, R.E. (1994). Fullerenes in the cretaceous-Boundary Layer. *Science*, 265, 645-647.
- 51)** Becker, L.; Bada, J.L.; Winans, R.E.; Hunt, J.E.; Bunch, T.E.; French, B.M. (1994). Fullerenes in the 1.85-billion year old Sudbury impact structure, *Science*, 265, 642-644.
- 52)** Jinno, K.; Uemura, T.; Ohta, H.; Nagashima, H.; Itoh, K. (1993). Separation and identification of higher molecular weight fullerenes by high performance liquid chromatography with monomeric and polymeric octadecylsilica bonded phases. *Anal. Chem.*, 65 (19), 2650-2654.
- 53)** Piskoti, C.; Yarger, J.; Zettl, A. (1998). C₃₆, a new carbon solid. *Nature*, 393, 771-774.
- 54)** Jinno, K.; Matsui, H.; Ohta, H.; Saito, Y.; Nakagawa, K.; Nagashima, H.; Itoh, K. (1995). Separation and identification of higher fullerenes in soot extract by liquid chromatography-mass spectrometry. *Chromatographia*, 41, 5-6.
- 55)** Kulkarni, P. P.; Jafvert, C.T. (2008). Solubility of C₆₀ in solvent mixtures. *Environ. Sci. Technol.*, 42 (3), 845-851.
- 56)** Welch, C. J.; Pirkle, W. H. (1992). Progress in the design of selectors for buckminsterfullerene. *J. Chromatogr., A*. 609 (1-2), 89-101.
- 57)** Bouchard, D.; Ma, X. (2008). Extraction and high performance liquid chromatographic analysis of C₆₀, C₇₀ and [6, 6]-phenyl C-61 butyric acid methyl ester in synthetic and natural waters. *J. Chromatogr., A*, 1203 (2), 153-159.

- 58)** Jehlicka, J.; Frank, O.; Pokorny, J.; Rouzaud, J. (2005). Evaluation of Raman spectroscopy to detect fullerenes in geologic materials , *Spectrochem, Acta part A*, 61, 2364-2367.
- 59)** Schreiner, K.; Filley, T.; Blanchette, R.; Bowen, B. B.; Bolskar, R.; Hockaday, W.; Masiello, C.; Raebiger, J. (2009). White-rot basidiomycete- mediated decomposition of C-60 fullerol. *Environ. Sci, Technol.*, 43, 3162-3168.
- 60)** Li, Q.; Xie, B.; Hwang, Y. S.; Xu, Y. (2009). Kinetics of C₆₀ fullerene dispersion in water enhanced by natural organic matter and sunlight. *Environ. Sci. Technol.*, 43 (10), 3574-3579.
- 61)** John H. Priester, Randall E. Mielke, Allison M. Horst, Joshua P. Schimel, Lijuan Zhao, (August 20, 2012), UCSB Scientists Examine Effects of Manufactured Nanoparticles on Soybean Crops, Public Affairs and Communications, UC Santa Barbara, USA.