Nano Pollution: A Pedagogical Strategy to Raise Awareness among Students

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Abstract— The introduction of nanosized pollutants in the natural environment is termed as Nanopluution or Nanocontamiation. Parallel to the growth of nanotechnology in every sphere of life, Nano contamination has also increased. It affects every aspect of our life especially plants and animals. It is an interdisciplinary field and it is very important to tackle this problem. For that we need trained personnel in this field. Unfortunately, there is a real shortage of any significant teaching strategies in the curriculum that can deal with the multidisciplinary character of this subject. This module is a small step in this direction. There has been very little study of effect of nanoparticles on plants. Keeping this in mind, the students were trained in preparation of silver nanoparticles, preparation of MS (Murashige and Skoog's) nutrient medium, incorporation of silver NPs (Nanoparticles) into the nutrient medium, sterilization of seeds, culture conditions, in vitro studies, transfer of plantlets to soil and in silico studies.

Key words: Nano pollution, Vigna radiata (L.), silver nanoparticles, tissue culture

I. INTRODUCTION

Nanotechnology is progressing leaps and bounds and its impact on society, industry and science in general is unprecedented. It includes a variety of subjects such as chemical sciences, material science, physical sciences, engineering, biological sciences, and medicine. This ever evolving area is rapidly growing in industry and research. This has led to a need of fostering it at undergraduate and postgraduate level. The need for teaching strategies for the integration of nanotechnology into the curriculum has been recognized in engineering and science education due to its multidisciplinary nature [1a]. Nano pollution is a generic name for waste generated by nanodevices or during the nanomaterials manufacturing process. Ecotoxicological impacts of nanoparticles and the potential for bioaccumulation in plants and microorganisms is a subject of current research, as nanoparticles are considered to present novel environmental impacts [1b].

The capacity for nanoparticles to function as a transport mechanism also raises concern about the transport of heavy metals and other environmental contaminants. Two areas of concern can be identified. First, in their free form nanoparticles can be released into the air or water during production, or production accidents, or as waste by-product of production, and ultimately accumulate in the soil, water, or plant life. Second, in fixed form, where they are part of a manufactured substance or product, they will ultimately have to be recycled or disposed of as waste. There is an urgent need to create awareness about this and for that we need trained personnel. The authors have recognized this need and the development of this module is a step in this direction [2].The first and foremost step in preparing a module is to plan a strategy that will take care of the interdisciplinary character in an innovative way.

We decided to incorporate the training of students in in vitro, in vivo and in silico techniques. Keeping this in mind, the problem selected was 'Study of effect of nanoparticles on plants using Tissue Culture Technique'.

II. MATERIAL AND METHODS

We proceeded with a mixed bag of students from Chemical Sciences, Plant Sciences and Life Sciences. As mentioned earlier, students were trained in in vitro, in vivo and in silico techniques to give them a thorough understanding of the various tools and techniques available to study the effect of nanoparticles on plants. The problem selected was the Study of effect of nanoparticles on plants using Tissue Culture [3, 4, 5]. To be specific, the effect of silver nanoparticles on the percent seed germination and seedling growth of Vigna radiata (L.) R. Wilczek (Fabaceae), an economically important plant was studied. Students were trained in the following techniques.

1) Preparation of Silver (Ag) Nanoparticles

Out of physical and chemical methods to prepare Ag Nanoparticles we have preferred to use chemical reduction. First of all, 100 ml of standard solution of silver nitrate (10mg in 100ml water) was prepared. After that 50 ml solution of sodium citrate (0.5g in 50ml distilled water) was prepared. Silver nitrate solution was poured in the beaker and the beaker was put on magnetic stirrer for half an hour.4ml of sodium citrate was slowly added to 100ml of silver nitrate solution, using pipette. The color change was observed after half an hour. After the color changed to grey, the stirring was stopped. Tween-20 was added as a stabilizing agent.

2) Preparation of MS Medium

The nutrient medium used was Murashige and Skoog's semi-solid medium [6] at a strength of 1/20. Sucrose at 3% concentration was added as carbohydrate source to the nutrient medium. The medium was solidified by adding 0.8% agar. The pH of the medium was adjusted to 5.8.

3) Addition of Silver NPs (Nanoparticles) Into the Nutrient Medium

Nanoparticles cannot be autoclaved as they are thermolabile. Therefore, silver NPs were filter sterilized by passing the solution, through sterilized Millipore filtration system with a pore size of 0.3 µm attached with clinical syringe. Then equal

amount of the nutrient medium supplemented with NPs was dispensed into culture tubes. All these steps were carried out in a Laminar Air Flow Chamber.

4) Sterilization of Seeds

The seeds of V. radiata were procured from a local nursery. They were first washed with teepol for 10 minutes. For surface sterilization, seeds were treated with 0.5% mercuric chloride solution for 8 minutes. They were finally washed with autoclaved double distilled water 3-4 times in a Laminar Air Flow Chamber.

5) Culture Conditions

The cultures of V. radiata were maintained in a culture room at a temperature of 26±20C and illumination of 80 µ mol sec-1 m-2 of 8 hours light and 16 hours dark.

6) In Vitro Studies

Two seeds per culture tube were inoculated for raising the cultures of V. radiata. The data was recorded on the following parameters: percent seed germination, shoot length, day of emergence of leaves, number of leaves, leaf characteristics, day of appearance of the radical, root length and number of root laterals.

7) Transfer of In Vitro Raised Plantlets to Soil

The axenically raised plants of V. radiata were transferred to pots containing vermiculite where they thrived well.

8) In Silico Studies

For studying the structural aspects, the DFT calculations were carried out in DMol3 [7] code. The generalized gradient approximation (GGA) in the revised Perdew–Burke–Ernzerhof (RPBE) form was adopted in the calculations. Students measured physiochemical properties such as particle size and distribution, shape, crystal structure, chemical composition and surface area.

III. RESULTS AND DISCUSSION

Since the module was specifically designed to deal with the interdisciplinary character of the subject, the learning experience was very comprehensive and unique. Students learnt a range of skills and techniques. It raised awareness about Nano pollution among the students. They learnt the techniques of plant tissue culture like medium preparation, inoculation and recording of the observations. They also received hands on experience for techniques such as UV Spectrophotometry, Fluorescence, Scanning and Transmission Electron Microscopy. Besides, the students also learnt the preparation of nanoparticle suspensions, knowledge of molecular modelling techniques that are the need of the hour for any student pursuing sciences. This module also helped students learn how to acquire and collate information, plan and design the experiment innovatively, the art of scientific writing besides honing their logistic and analytical skills. They have acquired general skills both in wet-lab and visual lab i.e. in silico techniques [8] and have also analyzed and synthesized the information collected. They have understood the implications of using technology at a rapid pace without proper checks and balances. Also, this module helped students understand the environmental impact of nanotechnology by themselves. No amount of lecturing or tutoring can raise this kind of awareness in which students themselves determined the effect nanoparticles have on plants. The module was a success with the students and a very satisfying experience for the teachers.

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