Application of Nanotechnology in Agriculture Sector – A Review

*Dr.E.N.Ganesh

*Professor ECE Dept Saveetha Engineering College

Abstract

Nanotechnology applications are currently being researched, tested and in some cases already applied across the entire spectrum of food technology, from agriculture to food processing, packaging and food supplements. Specifically in agriculture, technical innovation is of importance with regard to addressing global challenges such as population growth, climate change and the limited availability of important plant nutrients such as phosphorus and potassium. Nanotechnology applied to agricultural production could play a fundamental role for this purpose and research on agricultural applications is ongoing for largely a decade by now. This paper reviews the potential Nanotechnology Application in Agriculture like research in Nanobiotechnology in agriculture sector, Nanofabrication, DNA Sequence and Protein Analysis for Crop Improvement and come to conclusion that crop improvement is possible in extensive manner by the above techniques. We also discussed about Post harvesting Management, Food biotechnology, Nanobio engineering for agriculture products. This review will help to get awareness of using Nanotechnology in Agriculture Sector.

Keywords: Nanotechnology, Biotechnology, Post Harvesting and Nanofabrication.

1. Introduction

Nanotechnology is the creation and utilization of materials, devices and systems through the control of the properties and structure of matter at the nanometric scale. Nanobiotechnology is a new and exciting field of research, in which recent advances in nanotechnology are integrated into the biology realm, in particular into molecular biology and cell biology.[1][2] 'Nano' usually refers to a size scale between 1 nanometre (nm) and 100 nm. For comparison, the wavelength of visible light is between 400 nm and 700 nm. A leukocycte has the size of 10,000nm, a bacteria 1,000-10,000nm, virus 75-100nm, protein 5-50nm, deoxyribonucleic acid (DNA) ~2nm (width), and an atom ~0.1nm.[3][4]

All organisms, from microbes to humans, are powered by highly evolved molecular and cellular machines that operate at the nano level. Nature has been performing 'nanotechnological feats' for millions of years. Through the arrangement of atoms and molecules, biological systems combine wet chemistry and electro-chemistry in a single living system. The more we learn about these naturally occurring nano machines, the closer we come to engineering new ones with different functions. This convergence of technology with biology at the nano level is called 'nanobiotechnology'.[5][6][7]. Nanobiotechnology is a highly interdisciplinary field of research and is based on the cooperative work of chemists, physicists, biologists, medical doctors and engineers. We are just beginning to understand the nanoscale methods used in nature to create self-replicating, self-monitoring, self-controlling and self-repairing tools, materials and structures.[8][9] Figure 1 shows the explanation of Agri Products ending with Food Processing and Packaging with Suppliments.

Agriculture	Food Processing	Food Packaging	Supplements
 Single molecule detection to determine enzyme/ substrate interactions Nanocapsules for delivery of pesticides, fertilizers and other agrichemicals more efficiently Delivery of growth hormones in a controlled fashion Nanosensors for monitoring soil conditions and crop growth Nanosensors for identity preservation and tracking Nanosensors for detection of animal and plant pathogens Nanocapsules to deliver vaccines Nanoparticles to deliver DNA to plants (targeted genetic engineering) 	 Nanocapsules to improve bioavailability of neutraceuticals in standard ingredients such as cooking oils Nanoencapsulated flavor enhancers Nanotubes and nanoparticles as gelation and viscosifying agents Nanocapsule infusion of plant based steroids to replace a meat's cholesteroil Nanoparticles to selectively bind and remove chemicals or pathogens from food Nanoemulsions and -particles for better availability and dispersion of nutrients 	 Antibodies attached to fluorescent nanoparticles to detect chemicals or foodborne pathogens Biodegradable nanosensors for temperature, moisture and time monitoring Nanoclays and nanofilms as barrier materials to prevent spoilage and prevent sypoilage and prevent oxygen absorption Electrochemical nano- sensors to detect ethylene Antimicrobial and antifungal surface coatings with nanoparticles (silver, magnesium, zinc) Lighter, stronger and more heat-resistant films with silicate nanoparticles Modified permeation behavior of foils 	 Nanosize powders to increase absorption of nutrients Cellulose nanocrystal composites as drug carrier Nanoencapsulation of neutraceuticals for better absorption, better stability or targeted delivery Nanocochleates (coiled nanoparticles) to deliver nutrients more efficiently to cells without affecting color or taste of food Vitamin sprays dispersing active molecules into nanodroplets for better absorption

Figure 1 Shows the Application of Nanotechnology in Agriculture – [13][14]

2. Nanobiotechnology: Molecular Biology Complementing Nanotechnology

The credit for the term "nanobiotechnology" goes to Lynn W. Jelinski, a biophysicist at Cornell University, USA. Nanobiotechnology joins the breakthroughs in nanotechnology to those in molecular biology. Molecular biologists help nanotechnologists understand and access the nanostructures and nanomachines designed by 4 billion years of natural engineering and evolution - cell machinery and biological molecules. Scientists have been taking steps to build life from the nano-scale for some time. In 1968, Indian-American chemist Har Gobind Khorana received Nobel Prize for synthesizing nucleotides (the chemical subunits - A, T, C, G that make up the DNA molecule), stringing them together into synthetic DNA. By February 1976, a California research team (that later founded Genentech) developed an automated process for synthesising DNA and constructed a fully functioning synthetic gene. Synthetic genes and synthetic DNA is now a staple of genetic engineering in medicine and agriculture. Exploiting the extraordinary properties of biological molecules and cell processes, nanotechnologists can accomplish many goals that are difficult or impossible to achieve by other means. For example, rather than build silicon scaffolding for nanostructures, DNA's ladder structure provides nanotechnologists with a natural framework for assembling nanostructures; and its highly specific bonding properties bring atoms together in a predictable pattern to create a nanostructure. Nanotechnologists also rely on the self assembling properties of biological molecules to create nanostructures, such as lipids that spontaneously form liquid crystals.[10][11]

DNA has been used not only to build nanostructures but also as an essential component of nanomachines. Most appropriately, DNA, the information storage molecule, may serve as the basis of the next generation of computers. As microprocessors and microcircuits shrink to nanoprocessors and nanocircuits, DNA molecules mounted onto silicon chips may replace microchips with electron flow-channels etched in silicon. Such biochips are DNA-based processors that use DNA's extraordinary information storage capacity. Conceptually, they are very different from the DNA chips discussed below. Biochips exploit the properties of DNA to solve computational problems; in essence, they use DNA to do math. Scientists have shown that 1,000 DNA molecules can solve in four months computational problems that require a century for a computer to solve. Other biological molecules are assisting in our continual quest to store and transmit more information in smaller places. For example, some researchers are using light-absorbing molecules, such as those found in our retinas, to increase the storage capacity of CDs a thousand-fold.[12].

Nanobiotechnology, thus, is an emerging area of opportunity that seeks to fuse nano/microfabrication and biosystems to the benefit of both. It relates to all applications of genomics including mammalian, plant and microbial. It provides the basic tools and subsequently the technology for gathering sequence information and designing innovative devices to probe questions related to the biological importance of the genomic information and the application of this knowledge in diverse fields, particularly medicine and agriculture.[13].

The impact of nanobiotechnology may be immediately felt in the following areas:

3. Nanofabricated Gel-free Systems and High Throughput DNA Sequencing

As a central process, DNA sequencing needs to be improved in terms of its throughput and accuracy. Nanofabrication technology will be critical toward this goal both in terms of improving existing methods as well as delivering novel approaches for sequence detection. The scaling down in size of the current sequencing technology allows the process to be more parallel and multiplex. Research in nanobiotechnology is advancing toward the ability to sequence DNA in nanofabricated gel-free systems, which would allow for significantly more rapid DNA sequencing. Coupled with powerful approaches such as association genetic analysis, DNA sequencing data of the crop germplasm, including the cultivated crop gene pool and the wild relatives can potentially provide highly useful information about molecular markers associated with agronomically and economically important traits. Thus, nanobiotechnology can enhance the pace of progress in molecular marker-assisted breeding for crop improvement.

4. Micro-Chips and Expression Profiling

One of the first nanobiotechnologies in the market is micro-chips for DNA or protein sequencing (bio-chips). This technology is a good example of how a technology issued from traditional microelectronic industry is combined to a recently developed biotechnology. Another technology under development concerns microfluidic bio-chips, also known as lab-on-a-chip devices. They are all based on manipulation of minute bio-objects immersed in fluids, allowing on-chip biochemical processing (sampling, mixing, amplification, separation detection and analysis). The application area of biochips and microfluidic chips is very broad, ranging from high throughput screening, cell analysis, drug discovery to portable devices for minimal-invasive therapy, precision surgery as well as drug delivery.

Microarray-based hybridization methods allow to simultaneously measure the expression level for thousands of genes; this is referred to as 'expression profiling'. Such measurements contain information about many different aspects of gene regulation and function, and indeed this type of experiments has become a central tool in biological research. The development of

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novel formats for sequence determination and patterns of genomic expression which can have significantly higher throughput than current technologies is vital. Thousands of DNA or protein molecules are arrayed on glass slides to create DNA chips and protein chips, respectively. Recent developments in microarray technology use customized beads in place of glass slides. Overall, nanofabrication techniques can be used, for example, to pattern surface chemistry for a variety of biosensor and biomedical applications. Three areas which exemplify this are:

- Determination of new genomic sequences
- Scanning of genes for polymorphisms that might have an impact on phenotype

• Comprehensive survey of the pattern of gene(s) expression in organisms when exposed to biotic/abiotic stress.

The fundamental principle underlying the microarray technology has inspired researchers to create many types of microarrays to answer scientific questions and discover new products.

4.1 DNA Microarrays: DNA microarrays are being used to (i) detect mutations in diseaserelated genes; (ii) monitor gene activity; (iii) identify genes important to crop productivity; and (iv) improve screening for microbes used in bioremediation. Gene sequence and mapping data mean little until we determine what those genes do—which is where protein arrays come in.

4.2 *Protein Microarrays:* While going from DNA arrays to protein arrays is a logical step, it is by no means simple to accomplish. The structures and functions of proteins are much more complicated than that of DNA, and proteins are less stable than DNA. Each cell type contains thousands of different proteins, some of which are unique to that cell's job. In addition, a cell's protein profile varies with its health, age, and current and past environmental conditions.

Protein microarrays are being used to: (i) discover protein biomarkers that indicate disease stages; (ii) assess potential efficacy and toxicity of pesticides (natural and synthetics); (iii) measure differential protein production across cell types and developmental stages, and in both healthy and diseased states; (iv) study the relationship between protein structure and function; and (v) evaluate binding interactions between proteins and other molecules.

5 Crop Improvements

Nanotechnology has also shown its ability in modifying the genetic constitution of the crop plants thereby helping in further improvement of crop plants. Mutations -both natural and induced- have long since played an important role in crop improvement. Instead of using certain chemical compounds like EMS, MMS and physical mutagen like X-ray, gamma ray etc. for conventional induced mutation studies, nanotechnology has showed a new dimension in mutation research. In Thailand, Chiang Mai University's Nuclear Physics Laboratory has come up with a new white-grained rice variety from a traditional purple coloured rice variety called 'Khao Kam' through the usage of nanotechnology. The word "Kam" means deep purple, and the rice variety is known for its purple stem, leaves and grains. Using nanotechnology, the scientists changed the colour of the leaves and stems of Khao Kam from purple to green and the grain becomes whitish. The research involves drilling a nano-sized hole through the wall and membrane of a rice cell in order to insert a nitrogen atom. The hole is drilled using a particle beam (a stream of fast -moving particles, not unlike a lightning bolt) and the nitrogen atom is shot through the hole to stimulate rearrangement of the rice's DNA. This newly derived organism through the change at the atomic level is designated as 'Atomically Modified Organisms (AMOs).

6. Plant Disease Diagnostics

Diseases are one of the major factors limiting crop productivity. The problem with the disease management lies with the detection of the exact stage of prevention. Most of the times pesticides are applied as a precautionary manner leading to the residual toxicity and environmental hazards and on the other hand application of pesticides after the appearance of disease leads to some amount of crop losses. Among the different diseases, the viral diseases are the most difficult to control, as one has to stop the spread of the disease by the vectors. But, once it starts showing its symptoms, pesticide application would not be of much use. Therefore, detection of exact stage such as stage of viral DNA replication or the production of initial viral protein is the key to the success of control of diseases particularly viral diseases. Nano-based viral diagnostics, including multiplexed diagnostic kit development, have taken momentum in order to detect the exact strain of virus and stage of application of some therapeutic to stop the disease.

Detection and utilization of biomarkers that accurately indicate disease stages is also a new area of research. Measuring differential protein production in both healthy and diseased states leads to the identification of the development of several proteins during the infection cycle. These nano-based diagnostic kits not only increase the speed of detection but also increase the power of the detection.

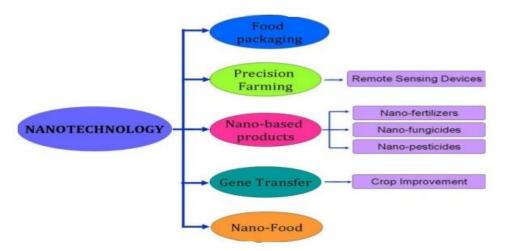


Figure 2 Nano Applications in Crop improvement and Fertilizers

Figure 2 shows how crop improvement is one of the potential applications from Nano Products.

7. Nanobiotechnology for Animal health

7.1 Nano Vaccines: Vaccination is one of the important methods of prevention of disease in advance by developing antibody against the particular pathogen. Most of the vaccines are applied as a fluid form and generally injected in to bloodstream. These vaccines require cool temperature to be stored and they also have limited life span within which they are to be utilized. These two limitations have prevented the utility of vaccines particularly in the rural area where availability of electricity, fridge and in many cases veterinarian are uncertain. Therefore, more robust and durable vaccines are the only solution for the successful eradication of particular disease.

Many organisms, particularly microorganisms, have novel and interesting structures that could be exploited, for example, the lattice-type crystalline arrays of bacterial S -layers and bacterial spore coats both of which have protective properties. In principle, the spore coat

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could be used not only as a delivery vehicle for a variety of different molecules but also as a source of new and novel self-assembling proteins. Spore coats are comprised of protein, have ordered arrays of protomeric subunits, exhibit self-assembly and have protective properties. As dormant metabolically inactive life forms, spores can survive indefinitely in a desiccated state, and indeed have been documented as surviving intact for millions of year. The spore can resist temperatures as high as 90°C as well as exposure to noxious chemicals. Most (but not all) spore forming bacteria belong to two principal genera, Bacillus and Clostridium. Clostridia spore -formers, unlike Bacillus, only differentiate under anaerobic conditions making Bacillus the most amenable genus for study. A strategy to engineer *Bacillus subtilis* spores to display heterologous antigens on the spore surface has been recently reported. A spore-based display system provides several advantages with respect to systems based on the use of conventional vaccines, these include the robustness of the bacterial spore allowing storage in the desiccated form, ease of production, safety and a technological platform supported by extensive tools for genetic manipulation. This bacterial spore-based nano-vaccines has been tested in human against tetanus with heterologous antigen inserted within it. The same kind of approach can be used successfully in animals such as cattle against deadly diseases like foot and mouth.

7.2 *Nano-apoptosis*: Like human being cattle are also prone to tumor and in some cases to cancer. This particular disorder in many times becomes the reason for death of animals. Conservation of genetically superior animal germplasm is very much critical for animal genetics and breeding studies coupled with development of the superior breeds. Although an array of systems ranging from chemotherapy to radiation are available, no remedy is fully guaranteed and in most of the cases cancerous animals die after initial improvement, this is mainly because of recurrence of cancerous cells from the remaining infectious cells. Therefore selective killing of cancerous cells are one of the feasible option to get rid of the deadly disease. Researchers at Rice University are using nanoshells injected into the animal's bloodstream with targeted agents applied to the nanoshells to seek out and attach to the surface receptors of cancer cells. Illumination of the body with infrared light raises the cell temperature to about 55°C, which 'burns' and kills the tumour.

Some research groups have been experimenting with 'smart' super-paramagnetic nanoparticles. These nanoparticles when injected in the bloodstream target tumour receptor cells. These nanoparticles are made from iron oxides that when subjected to a magnetic field enhances the ability of the nanoparticles to locate tumour cells. At the site of the tumour the nanoparticles emit an attached drug to kill the cancer cells.

Quantum dots may also be injected into the bloodstream of animals and they may detect cells that are malfunctioning. Because quantum dots respond to light it may be possible to illuminate the body with light and stimulate the quantum dot to heat up enough to kill the cancerous cell. Nucleic acid engineering-based probes and methods offer powerful new ways to deliver therapeutic or preventative treatment for particular diseases. The greatest challenge is to develop a nonviral DNA delivery system that has high levels of efficiency and specificity but low toxicity and cost. Therefore, future DNA delivery may depend on a hybrid system that combines the benefits of both viral and nonviral components. **Animal Breeding**

Efficient breeding of cattle is one of the most important factors for successful development of breeds, which mostly involve distant parents, and their oestrus is sometimes very difficult to detect and it leads to repeat breeding (failure of fertilization after repetitive insemination). The problem of repeat breeding is very common in rural areas where even artificial insemination fails to fertilize the egg as eggs are still under developed. Therefore, management of breeding is an expensive and time- consuming problem for dairy and swine farmers. One solution that is currently being studied is a nanotube implanted under the skin to provide real time measurement of changes in the level of estradiol in the blood. The nanotubes are used as a Page $\bf 6$ of $\bf 10$

means of tracking oestrus in animals because these tubes have the capacity to bind and detect the estradiol antibody at the time of oestrus by near infrared fluorescence. The signal from this sensor will be incorporated as a part of a central monitoring and control system to actuate breeding.

8. Post-Harvest Management and Food Biotechnology

8.1 Nano Bar Codes and Identity Preservation: Usage of bar code is the essential characteristics for selling almost all commodities both in the international market as well as in the national market. This bar code is essentially a sticker having a number of black and white bars with certain digits written at the bottom. The bar code is nothing but an electronic data depicting several parameters such as date of production, place of packaging, prices etc. and reading this code requires an electronic data reader. With the advent of nano technology, nano based bar codes are also available which can do the same function as that of conventional bar codes, thereby helping in tracking and controlling the quality of food product and give all relevant details in minute.

Each day a huge amount of shipments of livestock and other agricultural products are moved all over the world and it is becoming increasingly difficult to keep a track on critical control points of the production, shipment and storage processes. Lack of finances also limits the number of inspectors that can be employed at these critical control points. An identity preservation (IP) system can be installed that creates increased value by providing consumers with information about the practices and activities used to produce an agricultural product and it is possible to provide stakeholders and consumers with access to information, records and supplier protocols regarding the farm of origin, environmental practices used in production, food safety and security, and information regarding animal welfare issues.

Nano-based identity preservation has the potential to revolutionize the entire agri-based industry as it can continuously track and record the history of a particular agricultural product. The nanoscale monitors linked to recording and tracking devices can improve the IP of food and agricultural products. The keys are biodegradable sensors for temperature and other stored data containing the history of stored food for both physical and biological parameters. The future of the meat industry may well depend on an ability to track all stages in the life of the product, including the birth of the animal, its medical history, and its movements between the ranch, the slaughterhouse and the meat-packing plant, right through to the consumer's table *Monitoring Quality of Agricultural Products:* Nanotechnology also has applications in the agri-food sector. Many vitamins and their precursors, such as carotinoids, are insoluble in water. However, when formulated as nanoparticles, these substances can easily be mixed with cold water, and their bioavailability in the human body also increases.

Many lemonades and fruit juices contain these specially formulated additives, which often also provide an attractive colour. The world market potential of such micronized compounds is estimated at \$ 1 billion. In the future bio and gas sensors could gain importance. These sensors could be integrated into packaging materials to monitor the freshness of the food. Spoiling of the food could be indicated by a colour change of the sensor. Several concepts have already been developed for such applications based e.g. on silicon or polymer thin film sensors.

Bioselective surfaces are the new innovation of nano science technology with a principle that surfaces are the environment and location on which most chemical and biological interactions occur. A bioselective surface has either an enhanced or reduced ability to bind or hold specific organisms or molecules. With this bioselective surfaces minute amount of chemicals and even presence of bacteria and viruses can be detected with ease. These surfaces are important to

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the development of biosensors, detectors, catalysts and the ability to separate or purify mixtures of biomolecules.

8.2 Enzymatic nano bioengineering: Nanotechnology also has applications in the agri-food sector. Many vitamins and their precursors, such as carotinoids, are insoluble in water. However, when formulated as nanoparticles, these substances can easily be mixed with cold water, and their bioavailability in the human body also increases. Many lemonades and fruit juices contain these specially formulated additives, which often also provide an attractive colour. The world market potential of such micronized compounds is estimated at \$1 billion. In the future bio and gas sensors could gain importance. These sensors could be integrated into packaging materials to monitor the freshness of the food. Spoiling of the food could be indicated by a colour change of the sensor. Several concepts have already been developed for such applications based e.g. on silicon or polymer thin film sensors.

A huge amount of agricultural products and foods are wasted starting from the harvest at the field, their transportation, storage and further processing. These wastes are either rotten or damaged by rats leading to the huge crop loss. On the other hand it is very difficult to reduce the amount of wastage incurred due to either shortage of skilled manpower or lack of mechanization. In this scenario enzymatic nano bioengineering can be a right answer where more efficient enzymes are made through nano science technology to further process in order to make energy. Basically certain atoms of certain amino acids or the amino acids itself are engineered at nano scale to make the enzymes more efficient and rapid in degradation. This new approach is being explored in the USA, Israel and Japan.

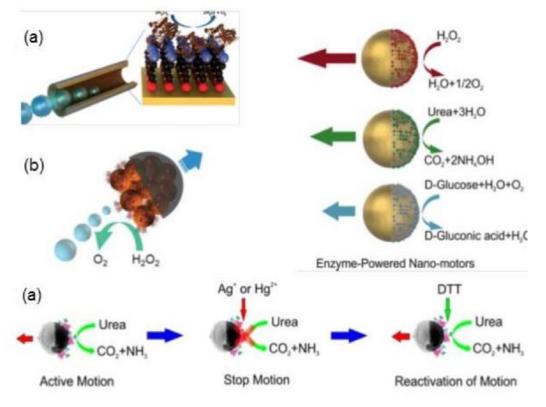


Figure 3 Enzyme Powered Smart Nano bio devices for Agri products [16]

India, undoubtedly, has the required talent in the individual fields of computational science, biology, physics and engineering. However, one can find hardly find an institute in the country that can claim to have a strong research and educational programmes linking all these vital

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components required for launching an effective nanobiotechnology programmes. This important rate-limiting factor can possibly be addressed by enhancing the capacity of the country in relation to high-throughput experimental technologies, and making necessary institutional adjustments for faculty hiring and orientation.

Some Universities in the USA have launched nanobiotechnology research and education programmes; notable among these are Cornell, Rice, Caltech, MIT, NC State and Harvard. Cornell University is at present leader among these institutions. Cornell's educational programme in nanobiotechnology requires the graduate students to be exposed to Applied Mathematics, Electronics. Drafting, Art, Embedded Systems, Hardware/Software Development, Genetics, Artificial Intelligence, Number Theory, Game Theory, Welding, Computer Graphics & Animation, Chaos Theory and Cosmology! Most importantly, this programme is strongly supported by faculty from diverse disciplines belonging to a Consortium of Institutions (including private sector) at the Cornell University's Nanobiotechnology Center. Fivure 3 shows the Smart Nano dievices using CNT and Cantilever with bio enzymes for crop Fertilizations.

9. Nano Applications in Agriculture:

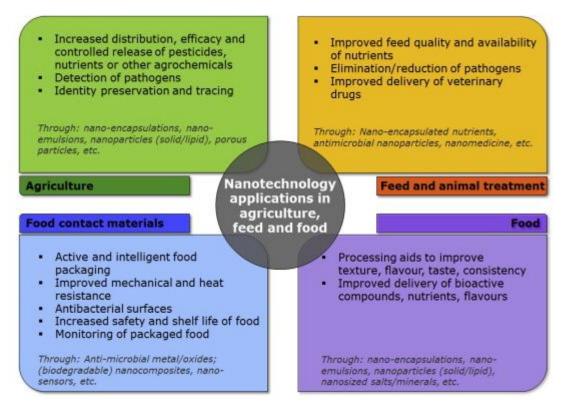


Figure 4 The Complete Nano Applications in Agriculture Sector [15]

Figure 4 shows the overall picture of Nanotechnology in Agriculture Sector. First Quadrant talks about Improved Feed Quality and nutrients availability and also veterinary drugs. Second Quadrant discuss about the distribution, control and detection of Pathogens. Third Quadrant explains about Food packaging, Anti bacterial properties and monitoring of food from agricultural products. Final Quadrants addresses bioactive compounds on agriculture products. Final Overall common Applications where the agriculture stands (as in Second Quadrant) will give us an idea that this nanotechnology (Encapsulations, Emulsions and

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Porous Particles) plays important role in overall crop Improvement and efficiency.

10. Conclusion:

This paper discussed about potential application of Nanotechnology in agriculture sector. We discussed molecular biology and DNA Sequencing and come to conclusion that Genomics plays important role through which fusing Nano fabrication and Bio technology possible. Protein, DNA and other Profile arrays shows the dealing disease of crops, Pesticides and relation with the molecules. This study will be helpful for crop improvement. We also explained the nature of usage of this technology in plant disease, crop improvement and Nano biotechnology. Finally complete figure of Nano applications in Agriculture is discussed in detail. We conclude that Nanotechnology plays vital role in crop improvement in yielding and gives us much profit.

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