

APPLICATION OF NANOTECHNOLOGY IN CROP DISEASE MANAGEMENT

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Abstract	<p><i>Insects and pests are the major threats to the economy of any country as they cause big losses to profit crop production. Frequent application of insecticides and pesticides has resulted in development of pest disease resistance, accumulation of residues in produce and the environmental pollution. So, there is a need for alternative strategies to control pests and pathogens. Nanotechnology offers new insights of biotechnology and agriculture. Application of nanotechnology in crop protection holds a significant promise in management of insects and pathogens, by controlled and targeted delivery of agrochemicals and also by providing diagnostic tools for early detection. Nanoparticles can serve as 'magic bullets', containing herbicides, pesticides, fertilizers, or genes, which target specific cellular organelles in the plant to release their contents. Nanoparticles are highly stable and biodegradable active compounds protected in capsules, they are degraded by external agents or the crop plant itself, and are not involuntarily dispersed into the soil, allowing the use of a reduced number of active compounds for plant treatments and consequently causing a low environmental impact.</i></p> <p><i>Further, nanoparticles linked with biomolecules with specific affinity (e.g. antibodies or aptamers) assure selectivity and specificity of targets. Along with these benefits, Nano devices for plant protection currently show also some constraints. First of all, there are not yet sufficient studies on the potential toxicity of nanomaterial (Nano silver, Nano gold, etc.) on plants, animals and the environment. Potentially, nanomaterial accumulate in vegetal and animal tissues, they can end up into the food chain. It is therefore fundamental to guarantee their safety and to correctly inform the consumers. However, the use of non-toxic materials (starch, chitin or Nano clays versus metals) eliminates such risk.</i></p>
Keywords	<p><i>Nanotechnology, disease management, nanomaterial, nanoparticles, agriculture,</i></p>

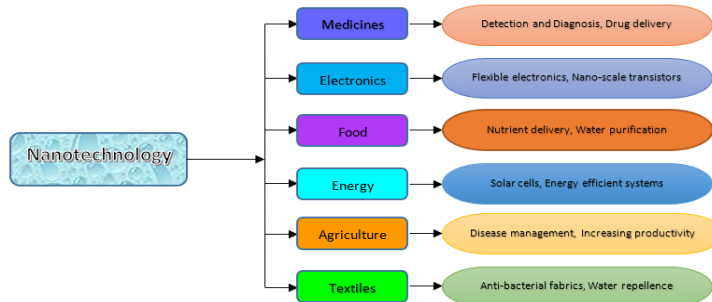
INTRODUCTION

Agriculture is the main source of income in most of the developing countries and major proportion of population depends on agriculture sector for their livelihood. Due to increasing population day by day, huge pressure is being developed on agriculture to fulfill the demand of food for all. In comparison to developed countries, a large proportion of population in developing countries faces food shortages daily as a result of poor farming practices, use of disease susceptible crop varieties and global environmental impacts including flood, drought and storms (Joseph and Morrison, 2006). Similarly, agricultural production continues to be constrained by a number of factors. For instance, diseases caused by insects, pests or other pathogens, and weeds cause considerable damage to agricultural production. Reports indicate that pests can cause up to 25% loss in rice, 5–10% in wheat, 30% in pulses, 35% in oilseeds, 20% in sugar cane and 50% in cotton (Dhaliwal *et al.*, 2010). Therefore, for developing countries like India, the crop yield can be maximized by developing drought and pest resistant crops (Joseph and Morrison, 2006).

A wide variety of bacterial and fungal pathogens are responsible to damage crops. Among them the most common bacterial agents are *Erwinia carotovora*, *Pseudomonas* spp., *Corynebacterium* and *Xanthomonas campestris* which attack most of the vegetables. Fungi commonly causing spoilage of vegetables are belonging to genera *Alternaria*, *Aspergillus*, *Cladosporium*, *Colletotrichum*, *Phomopsis*, *Fusarium*, *Penicillium*, *Phoma*, *Phytophthora*, *Pythium*, *Rhizopus* spp., *Botrytis cinerea*, *Ceratocystis fimbriata*, *Rhizoctonia solani*, *Sclerotinia sclerotiorum* etc. Some of these organisms are host specific whereas others affect a wide variety of vegetables

causing huge economic losses (Chowdappa and Gowda, 2013). Some pathogens produce toxic metabolites and adversely affect human health. Many of these agents enter the plant tissue over mechanical or chilling injuries, and cause great losses (Tournas, 2005). With the estimated doubling in global food demand in next 50 years huge challenges are expected in food production. In the year 2000 the pesticide production was about three million tons of active ingredients worldwide (Tilman *et al.*, 2002), and day by day its demand is increasing. Agricultural crops and other plants are major source of food, fibres, medicines and other livelihood resources, so, knowledge of plant diseases and their control is of vital importance to our survival.

Fig. 1. Scope Of Nanotechnology In Various Feilds.



DISEASE MANAGEMENT

The aim of plant disease management is to reduce the economic and aesthetic damage caused by pathogens. Traditionally, this has been called plant disease control, but in current scenario term “control” is not appropriate, so, management term looks much better. Disease management might be viewed as proactive

whereas disease control is reactive, although it is often difficult to distinguish between the two concepts, especially in the application of specific measures. Various methods have been followed to control or eradicate the pathogens, time to time, but diseases have been shown their resistance against all type of treatments, so, researchers have challegesto develop new strategies to manage these mighty pathogens.

TRADITIONAL OR CONVENTIONAL METHODS

Traditional disease management practices like crop rotation, use of healthy variety, manipulations in sowing dates, integrated pest management (IPM) are the measures undertaken by farmers to prevent and control diseases. Because these methods of disease management are economically viable, so, mostly these methods are used in low-return crops. These management methods are based on reducing the amount of initial inoculums, reducing the rate of spread of an established disease, or planting a crop at a site that is unfavorable to pathogens because of its physical environment like altitude, temperature, or humidity (Dhawan and Peshin, 2009; Peshin *et al.*, 2009; Rai and Ingle, 2012).

CHEMICAL BASED DISEASE MANAGEMENT

Synthetic chemical pesticides are widely used in conventional agriculture to control plant diseases. Environmental hazards caused by excessive use of pesticides pose health problems, so, it is not being preferred as modern society is becoming more health conscious (Kim *et al.*, 2009). As per reports, use of pesticides although began in the 1870s when arsenic and copper-based insecticides were developed. During the World War II, discovery of DDT (dichloro-diphenyl-trichloro-ethane) having pesticide properties revolutionized the pest control. DDT was effective at low concentration against almost all insect species, further; it was less expensive and supposed to be harmless to the human beings, animals and plants (Davies *et al.*, 2007). Therefore, farmers were amazed with its effectiveness and started to use it at large scale particularly during the green revolution era. But the negative impact of chemical pesticides started emerging soon and producers then turned to the much more toxic organophosphates and pyrethroid insecticides, which resulted in the development of resistant strains.

Most of the pesticides were originally based on the toxic heavy metals such as arsenic, mercury, lead and copper and that's why these are not eco-friendly (Davies *et al.*, 2007). Pesticides often kill the natural enemies along with the pests. With natural enemies eliminated, it was difficult to prevent recovered pest populations from exploding to higher and more damaging levels and often developing resistance to chemical pesticides. Initially the benefits from pest control were not huge due to use in low amount. Very soon DDT became popular and its use was increased enormously which resulted in the increase in yields, but on the other hand, their adverse effects on the environment and human health also soon became apparent. Indiscriminate, excessive and continuous use of pesticide creates a powerful selection pressure for altering the genetic make-up of the pests. This resulted in a much higher percentage pest population being resistant to pesticides (Biyela *et al.*, 2004; Levy, 2002).

BIOLOGICAL CONTROL OF DISEASES

Many biological agents have been used for the bio-control of insect pests, but only bacteria and fungi are most important. Bacteria used for biological control, infect insects via their digestive tracts. *Bacillus thuringiensis* is the most widely applied species of bacteria used for biological control of lepidopteron (moth, butterfly), coleopteran (beetle) and dipteran- true flies (Frederick and Caesar 2000). Fungi that cause disease or infection in insects are known as entomopathogenic fungi, including at least 14 species of entomophthoraceous fungi attack aphids. Species of the genus *Trichoderma* are used to manage some soil-borne plant pathogens like *Pythium*. *Beauveria bassiana* is used to manage different types of pest such as whiteflies, thrips, aphids and weevils (Thungrabeab and Tongma, 2007).

Although, biological control of disease management is eco-friendly, but so far strains against all pathogens are not available. Therefore, scientists in the agricultural field started searching for alternative eco-friendly and economically viable approaches to control plant diseases. As an alternative to chemically manufactured pesticides, use of nanoparticles as antimicrobial agents has become more common as technological advances make their production more economical (Jo *et al.*, 2009; Goncalves *et al.*, 1998).

NANOTECHNOLOGY: SCOPE IN PATHOGEN CONTROL

It has been reported that very small amount approximately less than 0.1% of pesticide reaches the sites of action, due to loss of pesticide in air during application and as run-off, spray drift, off-target deposition and photo-degradation affecting both the environment and cost of application (Pimentel, 1995; Castro *et al.*, 2013). With the increasing demand of pesticide worldwide to control the pathogens and pests, there is an urgent need to minimize the excessive usage of pesticides and fertilizers by finding better alternatives.

Nanotechnology has a great scope of application in field of agriculture, medicine and food industries and if exploited properly, it can revolutionize entire society. Potential applications of nanotechnology in crop protection include controlled release of encapsulated pesticide, fertilizer and other agrochemicals in protection against pests and pathogens. It is also useful in early detection of plant disease and pollutants including pesticide residues by using Nano sensors (Ghormade *et al.*, 2011; Linet *et al.*, 2020). The applications of nanoparticles in crop protection, helps in the development of efficient and potential approaches for the management of plant diseases.

Globally, insect pests make a huge crop loss of 14% and plant pathogens cause an estimated loss up to 13% with a value of US \$2,000 billion per year (Pimentel, 2009). Nano materials are useful for efficient and safe administration of pesticides, herbicides, and fertilizers at lower doses (Kuzma and VerHage, 2006). Excessive dose of pesticides cause adverse effects on human health and on pollinating insects. So, Nano-materials are helpful in decreasing toxicity and in

increasing the efficacy of pesticides (Mousavi and Rezaei, 2011). Nano pesticide formulations increase the solubility of poorly soluble active ingredient and helps in releasing the active ingredient slowly. The absorption of poorly soluble agrochemicals can be increased through the use of additives or nanoparticles formation of agrochemicals (Kahet *al.*, 2012). Nanoparticles are loaded with pesticides and released slowly based on environmental trigger (Lauterwasser, 2005). Due to their high reactivity at Nano scale compared to their bulk counterparts, small quantity of Nano-pesticides shows better effect in crop protection (Debnath *et al.*, 2011).

Nano encapsulation of agrochemicals such as an insecticide or pesticide allows slow and efficient release to a particular host plant for insect pest control and enables chemicalsto be adsorbed properly by the plants (Scrinis and Lyons, 2007).Nanoparticles of various metals are cost effective and reliable alternative for controlling insect pests and have been successfully employed by various research workers (Stadler *et al.*,2010; Bariket *al.*, 2008; Goswami *et al.*, 2010).

Pesticides can be effectively loaded into nanoparticles and can be slowly released related to an environmental trigger, they have uniform and extremely small droplet sizes (Forgiarini *et al.*, 2001). Nanomaterials have low viscosity, high kinetic stability and optical transparency which make them smart and efficient delivery systems for many industrial applications (Lee and Tadros, 1982). The use of nanoparticles was found to be a viable and effective alternative to conventional pesticides in combating pests which have developed pesticide resistance.

NANOCAPSULES AND NANOPARTICLES

The most relevant Nano devices for plant protection are Nano capsules and nanoparticles, both at a scale ranging from 0.1 to 1,000 nm. A Nano capsule is composed by a shell loaded with an active compound of our interest, like an agrochemical product for the protection of the plant against pests or diseases. The shell can be constituted by different elements, such as polymers, lipids, viral capsids or Nano clays. Its main function is to protect the active compound until it is released, but it can also improve the solubility and the penetration of the compound into the plant tissues. Depending on the specific characteristics of the shell, the active compound can be released slowly and gradually, or completely after the shell opening is mainly triggered by certain conditions i.e. pH changes or enzymatic degradation. Nanoparticles have a solid core or a matrix that can be composed by different materials such as metals or polymers and is surrounded by active biomolecules (Fig. 2). Due to the small size, the ratio between surface area and volume is increased in the nanomaterial compared with bulk forms.

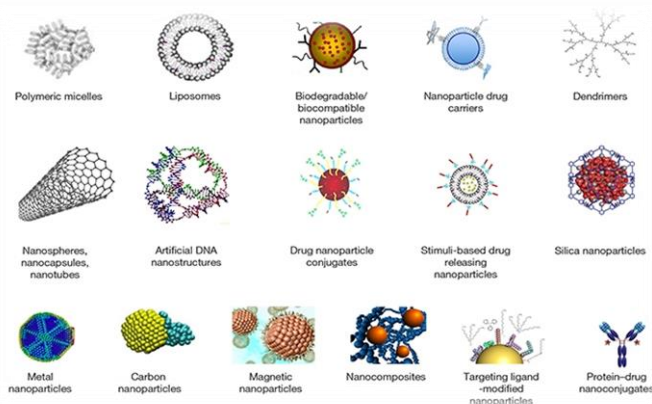


Figure 2. Types of Nanoparticles

(Source:<https://www.google.co.in/search?q=nanoparticle+types&tbm=isch&tbo=u&source=univ&sa=X&ved=0ahUKEwii-r7z1qvWAhVEsY8KHZPgCkcQsAQIbg#imgrc=YHyKhduG57V77M>.)

Nanoparticles can be synthesized chemically or biologically. Many adverse effects have been associated with chemical synthesis methods due to the presence of some toxic chemicals absorbed on the surface. Eco friendly alternatives to Chemical and physical methods are Biological synthesis nanoparticles using microorganisms (Klaus *et al.*, 1999; Konishi and Uruga, 2007) enzymes (Willner *et al.*, 2006), fungus (Vigneshwaran *et al.*, 2007), and plants or plant extracts (Shankar *et al.*, 2004; Ahmad *et al.*, 2011). The

development of these eco-friendly methods for the synthesis of nanoparticles is evolving into

an important branch of nanotechnology especially silver nanoparticles, which have many applications (Armendariz *et al.*, 2002; Kim *et al.*, 2010; Kyriacou *et al.*, 2004; Chuanxin *et al.*, 2018).

The most common methods for preparing all of these nanoparticles are wet-chemical techniques, which are generally low-cost and high-volume. However, the need for toxic solvents and the contamination from chemicals used in nanoparticle production limit their potential use in biomedical applications (Li *et al.*, 2011). Therefore a “green”, non-toxic way of synthesising metallic nanoparticles is needed in order to allow them to be used in a wider range of industries. This could potentially be achieved by using biological methods.

Many bacteria, **fungi** and **plants** have shown the ability to synthesise metallic nanoparticles and all have their own advantages and disadvantages (Suresh *et al.*, 2004; Bhainsa *et al.*, 2006; Song and Kim, 2009). Intracellular or extracellular synthesis, growth temperature, synthesis time, ease of extraction and percentage synthesised versus percentage removed from sample ratio, all play an important role in biological nanoparticle production. Finding the right biological method can depend upon a number of variables. Most importantly, the type of metal nanoparticle under investigation is of vital consideration, as in general organisms have developed resistance against a small number of metals, potentially limiting the choice of organism. However synthetic biology; a nascent field of science, is starting to address these issues in order to create more generalised chassis, able to synthesise more than one type of metallic nanoparticle using the same organism (Edmundson *et al.*, 2014).

“Natural” biogenic metallic nanoparticle synthesis can be split into two categories. The first is bioreduction in which metal ions are chemically reduced into more stable forms biologically. Many organisms have the ability to utilise dissimilatory metal reduction, in which the reduction of a metal ion is coupled with the oxidation of an enzyme (Deplanche *et al.*, 2011). This results in stable and inert metallic nanoparticles that can then be safely removed from a contaminated sample. The second category is biosorption. This involves the binding of metal ions from an aqueous or soil sample onto the organism itself, such as on the cell wall, and does not require the input of energy. Certain bacteria, fungi and plants express peptides or have a modified cell wall which binds to metal ions, and these are able to form stable complexes in the form of nanoparticles (Yong *et al.*, 2002).

Metallic nanoparticles are becoming increasingly important due to their potential application in many fields. The development of an environmentally friendly and inexpensive way of synthesising them is therefore crucial. There are numerous organisms possessing the ability to synthesise nanoparticles and which therefore have the potential to be exploited and modified to optimise them to fulfil this purpose (Table 1).

Table 1: **Metallic nanoparticles synthesis by different organisms with location of synthesis and method adopted.**

Name of organism	Nanoparticles Produced	Synthesis Location	Method	References
A) Bacteria				
<i>Bacillus licheniformis</i>	Ag	Intracellular	Reduction	Schlüter <i>et al.</i> 2014
<i>Bacillus sphaericus</i>	U, Cu, Pb, Al, Cd	Extracellular	Biosorption and Reduction	Das <i>et al.</i> 2014
<i>Bacillus spp.</i>	Ag	Intracellular	Reduction	Kalimuthu <i>et al.</i> 2008
<i>Delftia acidovorans</i>	Au	Extracellular	Reduction	Johnston <i>et al.</i> 2013
<i>Desulfovibrio desulfuricans</i>	Pd	Extracellular	Reduction	Cai <i>et al.</i> 2009
<i>Enterobacter cloacae</i>	Ag	Extracellular	Reduction	Kalimuthu <i>et al.</i> 2008
<i>Enterococcus faecium</i>	Ag	Extracellular	Biosorption and Reduction	Shahverdizadeh <i>et al.</i> 2007

Name of organism	Nanoparticles Produced	Synthesis Location	Method	References
<i>Escherichia coli</i>	Ag	Extracellular	Reduction	Kalimuthuet <i>al.</i> 2008
<i>Escherichia coli</i>	Pd, Pt	Extracellular	Reduction	Deplancheet <i>al.</i> 2010
<i>Klebsiellapneumoniae</i>	Ag	Extracellular	Reduction	Kalimuthuet <i>al.</i> 2008
<i>Lactobacillus spp.</i>	Ag	Extracellular	Biosorption and Reduction	Shahverdiet <i>al.</i> 2007
<i>Lactococcusgarvieae</i>	Ag	Extracellular	Biosorption and Reduction	Shahverdiet <i>al.</i> 2007
<i>Pediococcuspentosaceus</i>	Ag	Extracellular	Biosorption and Reduction	Shahverdiet <i>al.</i> 2007
<i>Pseudomonas aeruginosa</i>	Au	Extracellular	Reduction	Narayanan <i>et al.</i> 2010
<i>Rhodococcuspp.</i>	Au	Intracellular	Reduction	Park <i>et al.</i> 2011
<i>Rhodopseudomonascapsulata</i>	Au	Extracellular	Reduction	He <i>et al.</i> 2005
<i>Shewanellaspp.</i>	AsS	Extracellular	Reduction	Raveendranet <i>al.</i> 2003
<i>Shewanellaspp.</i>	Se	Extracellular	Reduction	Laudenslageret <i>al.</i> 2008
<i>Thermomonosporaspp.</i>	Au	Extracellular	Reduction	Kasthuriet <i>al.</i> 2008
B) Fungi				
<i>Aspergillusflavus</i>	Ag	Extracellular	Reduction	Sahaet <i>al.</i> 2010
<i>Aspergillusfumigatus</i>	Ag	Extracellular	Reduction	Bhainsa, D'Souza 2006
<i>Coriolusversicolor</i>	Ag	Intracellular & Extracellular	Reduction	Ahmad <i>et al.</i> 2002
<i>Fusariumoxysporum</i>	CdS	Extracellular	Enzyme Mediated	Raiet <i>al.</i> 2009
<i>Fusariumoxysporum</i>	Ag	Extracellular	Reduction	Ahmad <i>et al.</i> 2003
<i>Fusariumoxysporum</i>	Au	Intracellular	Reduction	Ma <i>et al.</i> 2005
<i>Neurosporacrassa</i>	Pt	Intracellular & Extracellular	Reduction	Sanghi, Verma 2009
<i>Verticillium sp.</i>	Au	Intracellular	Reduction	Ramanathanet <i>al.</i> 2013
C) Plants & Extracts				
<i>Acalyphaindicaleaf extract</i>	Ag	Extracellular	Reduction	Krishnarajet <i>al.</i> 2010
<i>Cymbopogonflexuosus leaf extract</i>	Au	Extracellular	Reduction	Gupta, Ganjewala 2015
<i>Jatrophacurcaslatex</i>	Ag	Extracellular	Reduction	Bar <i>et al.</i> 2009
<i>Magnolia kobus leaf broth</i>	Ag	Extracellular	Reduction	Song, Kim 2009
<i>Medicago sativa seed exudate</i>	Ag	Extracellular	Reduction	Spadaro, Gullino 2005
<i>Phyllanthusamarus</i>	Ag, Au	Extracellular	Reduction	Kasthuriet <i>al.</i> 2008
<i>Pinuseldarica bark extract</i>	Ag	Extracellular	Reduction	Iravani, Zolfaghari 2013

BIOSYNTHESIS: MECHANISM

Biosynthesis of nanoparticles by microorganisms is a green and eco-friendly technology. Diverse microorganisms, both prokaryotes and eukaryotes are used for synthesis of metallic nanoparticles viz. silver, gold, platinum, zirconium, palladium, iron, cadmium and metal oxides such as titanium oxide, zinc oxide, etc. These microorganisms include bacteria, actinomycetes, fungi and algae. The synthesis of nanoparticles may be intracellular or extracellular according to the location of nanoparticles (Hulakoti and Taranath, 2014; Mann, 2001).

INTRACELLULAR SYNTHESIS OF NANOPARTICLES BY FUNGI

This method involves transport of ions into microbial cells to form nanoparticles in the presence

of enzymes. As compared to the size of extracellularly reduced nanoparticles, the nanoparticles formed inside the organism are smaller. The size limit is probably related to the particles nucleating inside the organisms (Narayanan and Sakthivel, 2010).

EXTRACELLULAR SYNTHESIS OF NANOPARTICLES BY FUNGI

Extracellular synthesis of nanoparticles has more applications as compared to intracellular synthesis since it is void of unnecessary adjoining cellular components from the cell. Mostly, fungi are known to produce nanoparticles extracellularly because of their enormous secretory components, which are involved in the reduction and capping of nanoparticles (Narayanan and Sakthivel, 2010).

MICROBES FOR PRODUCTION OF NANOPARTICLES

Both unicellular and multicellular organisms produce inorganic materials either intra- or extracellular (Shiv Shankar *et al.*, 2004). The ability of microorganisms like bacteria and fungi to control the synthesis of metallic nanoparticles is employed in the search for new materials. Because of their tolerance and metal bioaccumulation ability, fungi have occupied the center stage of studies on biological generation of metallic nanoparticles (Sastri *et al.*, 2003).

SCOPE AND FUTURE PROSPECTIVE

The application of nanomaterials in agriculture aims in particular to reduce applications of plant protection products, minimize nutrient losses in fertilization, and increase yields through optimized nutrient management. Despite these potential advantages, the agricultural sector is still comparably marginal and has not yet made it to the market to any larger extent in comparison with other sectors of nanotechnology application. Nanotechnology devices and tools, like nanocapsules, nanoparticles and even viral capsids, are examples of uses for the detection and treatment of diseases, the enhancement of nutrients absorption by plants, the delivery of active ingredients to specific sites and water treatment processes. The use of target-specific nanoparticles can reduce the damage to non-target plant tissues and the amount of chemicals released into the environment. Nanotechnology derived devices are also explored in the field of plant breeding and genetic transformation. The potential of nanotechnology in agriculture is large, but a few issues are still to be addressed, such as increasing the scale of production processes and lowering costs, as well as risk assessment issues. In this respect, particularly attractive are nanoparticles derived from biopolymers such as proteins and carbohydrates with low impact on human health and the environment. For instance, the potential of starch-based nanoparticles as nontoxic and sustainable delivery systems for agrochemicals and bio-stimulants is being extensively investigated. Nano materials and nanostructures with unique chemical, physical, and mechanical properties (e.g. electrochemically active carbon nanotubes, Nano fibers and fullerenes) have been recently developed and applied for highly sensitive bio-chemical sensors. These Nano sensors have also relevant implications for application in agriculture, in particular for soil analysis, easy biochemical sensing and control, water management and delivery, pesticide and nutrient delivery. Nano composites based on biomaterials have beneficial properties compared to traditional micro and macro composite materials and, additionally, their production is more sustainable. Many production processes are being developed nowadays to obtain useful Nano composites from traditionally harvested materials. Commercial applications of nanotechnology in the agricultural sector from a commercial perspective, existing agro-chemical companies are investigating the potential of nanotechnologies and, in particular, whether intentionally manufactured nano-size active ingredients can give increased efficacy or greater penetration of useful components in plants. However, the nano-size so far did not demonstrate to hold key

improvements in products characteristics, especially considering the interest of large scale production and the costs involved in it. Some specific Nano-products for the agricultural sector have been put on the market by technology-oriented smaller companies, like soil-enhancer products that promote even water distribution, storage and consequently water saving.

However, the commercial market application of these products is so far only achieved at small scale, due to the high costs involved in their development. These costs are normally compensated by higher returns in the medical or pharmaceutical sectors, but so far there are no such returns in the agricultural sector. Research continues in the commercial agro-chemical sector to evaluate potential future advantages. Companies are also facing challenges derived from the definition of nanomaterials that is adopted by the EU. One crucial point related to the EU definition is the possibility that non-active substances already used for many decades in commercial products formulations will fall within the scope of the nanodefinition, although not intentionally developed as nanoparticles or having specific nano-scale properties. Nanoscaleformulants (e.g. clay, silica, polymers, pigments, macromolecules) have been used for many decades and are also ubiquitous in many daily household products. The concern is that the need for labelling of products that are already on the market since decades results in a scenario, in which the technology is stigmatized, preventing further and innovative applications of nanotechnology in agriculture.

DRAWBACKS

Despite these potential advantages, nanotechnology applications in the agricultural sector are still comparably marginal and have not yet made it to the market to any large extent in comparison with other industrial sectors. The wave of research discoveries seems to be mainly claimed by the academic sector or small enterprises, while big industries reveal a large patent ownership. The trends of patent applications (mainly from agro-chemical companies) are continuously growing, but no new nano-based products for the agricultural sector have reached the market. This suggests that applicants are actively patenting and keeping broad patent claims in order to assure future freedom to operate and to guarantee future exploitation in case of promising commercial developments.

Large companies are investigating the potential that nanotech solutions offer in the agricultural field. However, according to industry experts, agricultural nanotechnologies so far do not demonstrate a sufficiently high economic interest. Nanotech products require high initial investments that can be counterbalanced only by large-scale field uses, which is not currently the case. Among the reasons for the difficulties of agricultural nanotechnology developments at field level, industrial organisations cite regulatory issues and public opinion.

One of the most important aspects of regulating nano-materials is the achievement of a definition agreed among the involved parties and, possibly, harmonized at international level. The definition of nano-materials seems not to be straightforward and is not just a matter of size. The nano-scale can be applied to one or more dimensions and the form of the particles can be in aggregate, agglomerates or nanostructured materials. Moreover, since nanotechnology is applied in different industrial sectors, different regulatory bodies and regulations are involved in its safety assessment.

CONCLUSION

Application of nanotechnology in crop protection holds a significant promise in management of insects and pathogens, by controlled and targeted delivery of agrochemicals and also by providing diagnostic tools for early detection. Nanoparticles can serve as 'magic bullets', containing herbicides, nano-pesticides, fertilizers, or genes, which target specific cellular organelles in the plant to release their content. Nanoparticles are highly stable and

biodegradable active compounds protected in capsules, they are not degraded by external agents or the crop plant itself, and are not involuntarily dispersed into the soil, allowing the use of a reduced amount of active compounds for plant treatments and consequently causing a lower environmental impact. But in spite of many advantages, agro-nanotech innovative products are experiencing difficulties in reaching the market, making agriculture still a marginal sector for nanotechnology. This is due in particular to the high production costs of nanotech products, which are required in high volumes in the agricultural sector, unclear technical benefits and legislative uncertainties, as well as public opinion. Nevertheless, the R&D landscape is very promising and the possibilities offered by nanotechnology in several agricultural applications are being actively explored. Additionally, nanotechnology is progressing at rapid pace in other fields. The knowledge gained in other emerging sectors, such as energy and packaging, may over time be transferred, or may provide spill-overs, to agricultural applications as well. For instance, improved fuel additives and lubricants could also improve the performance and the carbon footprint of agricultural machinery and improved packaging measures could benefit farmers by reducing the degradation of products before consumption. Meanwhile progress in environmental monitoring and drug delivery techniques could positively affect the agricultural and livestock sector indirectly.

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