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BIOGENESIS OF NANOPARTICLES — A CURRENT PERSPECTIVE

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Abstract. In the last decade, synthesis of nanoparticles has been the subject of a lot of studies due to its commercial importance and applications. Their interesting characteristics and wide applications have led to numerous methods being developed for synthesis of nanoparticles of various shapes and sizes. Typically, the methods employed for their synthesis of nanoparticle include physical, mechanical and chemical methods. However, these methods are very expensive and some of them which involve hazardous chemicals. Therefore, there is emergent need to develop environmentally benign and sustainable methods for nanoparticle synthesis. Green chemistry processes led to environmental friendly method of synthesis and safe process as compared to other methods. Biological sources such as fungi, bacteria, viruses, actinomycetes, algae and plant materials, etc. can catalyze specific reactions as a part of modern and realistic biosynthetic strategies. These can be alternate to other physical, chemical methods, ultraviolet irradiation, lithography, laser ablation, ultrasonic fields, and photochemical reduction techniques. The general outline of this present work includes information of the different microbial sources, importance of physicochemical characteristics, and the applications of metal nanoparticles.
The commercial importance of "current perspectives" includes the precise and specific analysis of nanoparticles, biological systems that may support and revolutionize the art of synthesis of nanoparticles. The advances of biological route from the general, molecular, catalytic, and functional information obtained under close to optimal conditions through action of enzymatic properties may help to understand the biochemical and molecular mechanisms of nanoparticles formation.

1. INTRODUCTION

The modern technology accepts that the concept of interdisciplinary research in the areas of engineering and sciences leads to creation of environmentally acceptable "green processes", with special concern to nanoscience and nanotechnology. Nanoscience research encompasses multiple disciplines that draw knowledge from disparate science and engineering discipline sources (Fig. 1). The word *nano* has been adopted from Greek, it means "dwarf". A nanometer (nm) is one billionth of a meter, or roughly the length of three atoms side by side (Table 1). Nanotechnology provides initiative for the production, manipulation and use of materials ranging in size from less than a micron to that of individual atoms (10^{-9}). Likewise, bionanoscience and bionanotechnology is an interdisciplinary area of research that operates at the interface of chemistry, biology, materials science, engineering and medicine (Fig. 2). Nanobiotechnology is defined as an area that applies the nanoscale principles and techniques to understand and transform biosystems (living or non-living) and which uses biological principles and materials to create new devices and systems integrated from the nanoscale. The integration of nanotechnology with biotechnology, as well

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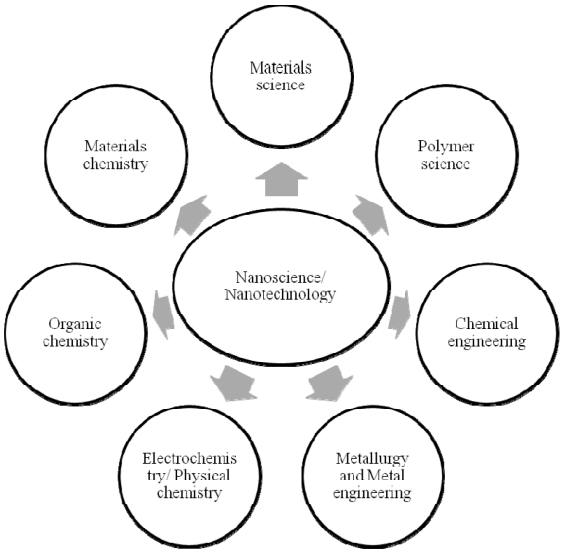

Fig. 1. Importance of nanoscience and nanotechnology in various research areas.

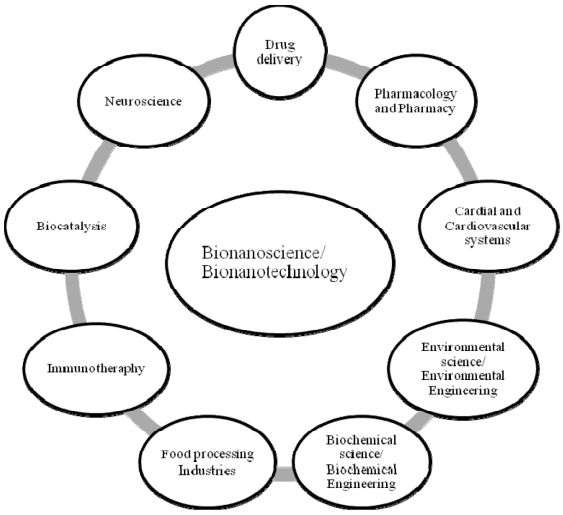

Fig. 2. Application of bionanoscience and bionanotechnology in scientific research.

as with information technology and cognitive science, is expected to accelerate in the next decade. As a result of nanotechnology, the nanoparticles have extensive applications in science and industrial engineering field. The nanoparticles have attracted interest because of their small size, exhibition of emergent properties like quantum dots, nonlinear optical, thermal, electrical, chemical properties and so on. Numerous research studies have shown the importance of nanoparticles to improve human health, electronic, magnetic and optoelectronic, biomedical, pharmaceutical, cosmetic, energy, environmental, catalytic, and material applications. In biotechnological analysis, they can be used as bacteriostatic materials, antibacterial materials, antistatic material, cryogenic superconducting materials, and biosensor materials etc. [1-6]. In medical diagnostic applications, nanoparticles can be attached to a single strand of DNA nondestructively [7]. In electronic industry, they are used to reduce the thickness of conductive films and the width of printed circuits [8]. Owing to the environment friendly nature of biosynthetic process, there is a great interest in studying the synthesis of nanoparticle by biological route. There are number of reasons for the utilization of nanoscale materials over than the microscale particles. Several studies have utilized bimetallic nanoparticles as an effective oxidant instead of granular zero-valency metal in the cleanup of environmental contaminants, mainly because (i) nanoparticles can diffuse or penetrate into a contamination zone where microparticles cannot reach, and (ii) higher reactivity to redox-amenable contaminants than that of microsized particles can be obtained. Nanoparticles can be further used to immobilize microbial cells that can degrade or biorecover specific chemicals and also be used as biocatalysts for reductive dechlorination [9].

1.1. Application and potential uses of biological method

Production of metal nanoparticles has a significant role in human disease control and involves the prediction of molecular structure in material science. For the synthesis of metal nanoparticles numerous techniques available including chemical and physical methods such as chemical reduction, electrochemical reduction, photochemical reduction, heat evaporation, etc. (Fig. 3). The major drawback with admission to the physical and chemical method, it

Table 1. Size distribution of nanomaterials.

Materials	Size in nm
DNA	25
Protein	50
Flu virus	100
Human blood	2000 to 5000
Human hair	10,000
Hemoglobin	5
DNA double helix span	2
Mitochondrion	Few hundred

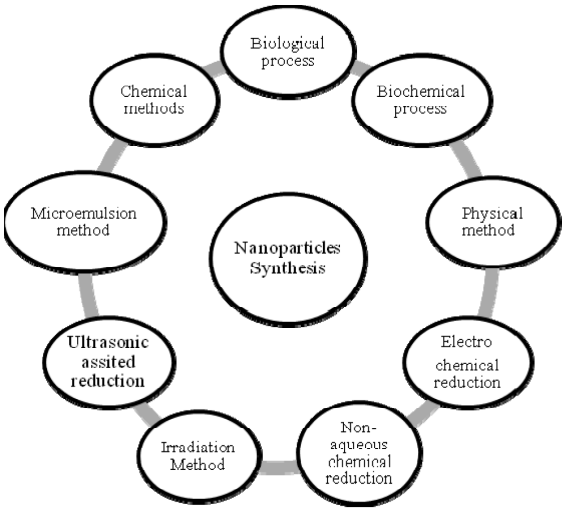

Fig. 3. Preparation of nanoparticles using various methods.

needs surface passivator reagent to predict or prevent nanoparticles from aggregation. In most cases, the organic passivators such as thiophenol, thiourea, marcapto acetate etc act as a toxic to the environment and creates the polluted nature to the environment. To overcome and make a pollution free environment there is need of new technology and/or new synthesis method for nanoparticles. Currently, there is an ever-growing need to develop environmentally benign nanoparticle synthesis processes. In recent decade attention towards the synthesis of nanoparticels using biological materials has great improvement to access the environmental friendly life. Generally metal ions inhibit the growth of microorganisms, but some microorganisms can survive and grow even at high metal ion concentration due to their resistance to the metal [10]. The applications of these metal-microbe interactions play a wide role in biotechnological processes, and in fields including bioremediation, biomineralization, alteration of solubility and toxicity by reduction or oxidation, extracellular complexation or precipitation of metals, bioleaching and microbial corrosion [11]. Biological route of synthesis of nanoparticles is mainly used because of its extensive advantages over other traditional methods. The advantages such as defined and mild reaction conditions suited to the environment, adequate range of material sources present and good nature of reduction takes place to form nanoparticles. The time for completion of the reaction, which is an obvious advantage of the biosynthetic procedures compared to the chemical methods. While the chemical and physical methods continue to be investigated in nanoparticle synthesis, the use of microorganisms and plant materials in similar nanoparticle synthesis methodologies is an exciting possibility that is relatively unexplored and under exploited.The biotechnological approach to the synthesis of nanoparticles has two different mechanisms, namely enzymatic reduction process and non-enzymatic reduction process. Similarly, the natures of synthesized nanoparticle have two different types, namely intracellular and extracellular nanoparticles. Controlling the size of nanoparticles could help in the understanding of biochemical and molecular mechanisms. Efforts have been made to predict the size of extracellular synthesized nanoparticles by altering the parameters. Furthermore, the use and application of extracellular form of nanoparticles could be beneficial over intracellular synthesized nanomaterials [12].

2. BIOGENESIS OF NANOPARTICLES

Understanding of biological processes on the nanoscale level is a strong driving force behind development of nanotechnology. Living organism ranges from prokaryotic to eukaryotic are built of cells that are typically 10 mm across. Numerous varieties of biological sources available in nature including plants and plant products, algae, fungi, yeast, bacteria, and viruses could all be employed for the synthesis of nanoparticles. However, the parts of cell are smaller than the proteins with a typical size of just 5 nm and also present in sub micron range. This simple size comparison gives an idea of using microorganisms for production of nanoparticles at the cellular machinery without introducing too much interference.

It has been well studied that a variety of microorganisms are able to produce inorganic and metal nanoparticles of different shapes and nature. Nanoparticle production and applications have been extensively studied; studies related to drug delivery, tissue engineering and bioMEMS have been undertaken for a great number of scientific publications and patents. There have been many reports about the biotechnological production of nanoparticles under different conditions by various microbial sources.

Herein, we provide an overview of various reports of biological means of nanoparticle synthesis of desired characteristics. The present work is not meant to be a review of all the current and past literature on the subject. A planned review of extensive synthesis of nanoparticles, applications of biological method, biological sources and applications

of nanoparticles of all reported works, so do check back periodically for an update.

2.1. Biogenesis of nanoparticles by bacteria

The application of bacterial strain for biomanufacturing processes have advantages over the other biological sources such as easy handling and short period of cultivation. *Pseudomonas stutzeri* AG259 isolated from silver mines has been shown to produce silver nanoparticles [13,14]. This strain generates well defined shapes such as pyramidal and hexagonal silver nanoparticles of up to 200 nm sizes. The produced nanoparticles were quantified by TEM, XRD to identify their different shaped crystals [14]. [15] reported that the magnetotactic bacterial strains were good candidate for nanoparticles synthesis. Magnetotactic bacteria have the ability to produce intracellular membrane bound ferromagnetic particles composed of magnetite (Fe_3O_4) or greigite (Fe_3S_4) which are covered with an intracellular phospholipid membrane; the structures thus formed are called magnetosomes. In conformity with Schuller and Frankel [15], the synthesis of magnetic nanoparticles has been reported by using magnetotactic bacteria [16]. Magnetotactic bacteria such as *Magnetospirillum magneticum* produce two types of particles; some produce magnetic (Fe_3O_4) nanoparticles in chains and some produce greigite (Fe_3S_4) nanoparticles, while some other produce both types of nanoparticles. The sulphate reducing bacterium has the ability to synthesize palladium nanoparticles using *Desulfovibrio desulfuricans* NCIMB 8307 has been shown to be [17]. [18] published that the isolated strains of *Klebsiella planticola Cd-1*, a highly cd-resistant strain from reducing salt marsh sediments. The isolated strain was found to grow different production media presence of up to 15 mM $CdCl_2$ at pH ranging from acidic or neutral. The well defined amounts of cadmium sulfide precipitated in growth medium amended with thiosulfate, as confirmed by x-ray absorption spectroscopy. The generation of Ag, Au, and Ag-Au alloy crystals of submicron dimensions upon exposure to the precursor ions by *Lactobacillus* strains is common in buttermilk. Crystal growth was observed to occur by the coalescence of clusters, and number of crystals was found within the bacterial contour. Coalescence appeared to be a route by which surface area of the crystal was reduced so that it can be effectively protected to avoid biological damage [19]. The selenium respiring bacteria has the ability to synthesize selenium nanospheres [20]. The microbial synthesis of selenium nanoparticles result in unique, complex, compact nanostructured arrangement of Se atoms. These arrangements probably reflect a diversity of enzyme involved in the assimilatory reductions that are subtly different in different microbes. More recently, supernatant of *Pseudomonas aeruginosa* was used for the reduction of gold ions resulting in extracellular biosynthesis of gold nanoparticles [12]. This finding provides a better understanding of mechanism and biochemical synthesis protocols. Cubic gold nanoparticles and octahedral gold plates produced from the filamentous cyanobacterium, *Plectonema boryanum* UTEX 485 exposed to aqueous Au $(S_2O_3)_2$ and $AuCl_4$, respectively [21]. Then, the mechanisms of gold bioaccumulation using *Plectonema boryanum* UTEX 485 to aqueous gold (III)-chloride solution, the reaction to the gold solution by *Plectonema boryanum* UTEX 485 initially promoted the precipitation of nanoparticles at the cell walls, and finally deposited metallic gold in the form of octahedral (III) platelets near cell surfaces and in solutions [22]. Lengke and Southam [23] reported that the addition to the mechanism, a sulfate-reducing bacterial enrichment was used to destabilize gold (I)-thiosulfate complex to elemental gold and proposed that this could occur by three possible mechanisms involving iron sulfide, localized reducing conditions, and metabolism. Table 2 presents some of the valuable published patents and publications regarding biotechnological production of nanoparticles.

Microbial syntheses of nanoparticles in both intracellular and extracellular form are observed efficiently from variety of bacterial strains. The variety of bacterial strains includes *Clostridium thermoaceticum* [24], *E.coli* DH5α [26], *Aeromonas sp*.SH10 [28], *Actinobacter sp*. [29], *Rhodopseudomonas capsulate* [30], *Thermomonospora sp*.[31], *Shewanella oneidensis* [25], *Morganella sp*.[32], *Bacillus megaterium* D01[32], *Lactobacillus sp*. A09[32], etc. For the complete list of biological sources, please refer to Table 2.

2.2. Biogenesis of nanoparticles by fungi

For large-scale biogenesis of nanoparticles, fungi possess unique advantages over bacteria:

1. Most fungi have a very high wall-binding capacity as well as intracellular metal uptake capacities [40].

Table 2. Microbial sources for synthesis of nanoparticles**.**

Type of sources	Microorganism	Nanomaterials/ nanoparticles	Nature of product/ Localization	Reference (s)
Bacterial sources	Pseudomonas aeruginosa	Gold	Extracellular	[12]
	Pseudomonas stutzeri AG259	Silver crystals	Intracellular	[14]
	Lactobacillus strains	Silver and gold	Intracellular	[19]
	Plectonema boryanum (Cyanobacteria)	Silver	Intracellular	[21]
	Clostridium thermoaceticum	CdS	Intracellular and extracellular	[24]
	Shewanella algae	Gold	Intracellular	[25]
	Escherichia coli DH5α	Gold	Intracellular	[26]
	Rhodococcus sp.	Gold	Intracellular	[27]
	Aeromonas sp. SH10	Silver	Intracellular	[28]
	Bacillus megaterium D01	Silver	—-	[28]
	Lactobacillus sp. A09	Silver	—-	[28]
	Actinobacter spp.	Magnetite	Extracellular	[29]
	Rhodopseudomonas capsulate	Gold	Extracellular	[30]
	Thermomonospora sp.	Gold	Extracellular	[27]
	Klebsiella pneumonia	Silver	Extracellular	[31]
	Shewanella oneidensis	Silver	Extracellular	[25]
	Morganella sp.	Silver	Extracellular	[32]
	Bacillus licheniformis	Silver	Extracellular	[33]
	Bacillus licheniformis	Silver	Extracellular	[34]
	Bacillus licheniformis	Silver	Extracellular	[35]
	Bacillus cereus	Silver	—-	[36]
	E. coli	Silver	Extracellular	[37]
	Bacillus licheniformis	Gold	Extracellular	[38]
	E. coli	CdS	Intracellular	[39]
Fungi	Verticillium	Silver	Intracellular	[50]
	Phoma sp. 3.2883	Silver	Extracellular	[51]
	Fusarium oxysporum	Gold	Extracellular	[52]
	Aspergillus fumigates	Silver	Extracellular	[53]
	Trichoderma asperellum	Silver	Extracellular	[54]
	Phaenerochaete chrysosporium	Silver	Extracellular	[55]
	Fusarium oxysporum and Verticillium sp.	Magnetite	Extracellular	[56]
	Cladosporium cladosporioides	Silver	Extracellular	[57]
	Penicillium sp.	Silver	Extracellular	[58]
	P. brevicompactum	Silver	Extracellular	[59]
	Phytophthora infestans	Silver	—-	[60]
	Fusarium oxysporumPTCC 5115	Silver	—-	[61]
Yeast	Candida glabrata	Cds	Intracellular	[62]
	Schizosaccharomyces pombe	Cds	—-	[63]
	MKY3	Silver	Extracellular	[66]
	Yarrowia lipolytica NCIM3589	Gold	—-	[69]
	Candida Sp.	Silver	—-	[70]
Plant and plant extracts	Cinnamomum camphora	Silver and gold	Extracellular	[7]
	Geranium leaves plant extract	Silver	—-	[84]
	Alfalfa sprouts	Silver	Intracellular	[85]
	Avena sativa (Oat)	Gold	Extracellular	[86]
	Lemongrass plant extract	Gold	—-	[87]

	Aloe vera	Gold	Extracellular	[90]
	Azadirachta indica(Neem)	Ag, Au, and Ag/Au bimetallic	Extracellular	[91]
	Mushroom extract	Au, Ag and Au-Ag	—-	[92]
Algae	Sargassum wightii	Gold	Extracellular	[93]
	Chlorella vulgaris	Gold	—-	[94]

2. They are easy to culture on a large scale by solid substrate fermentation, thus making a large amount of biomass available for processing.
3. Fungi can grow over the surface of inorganic substrate during culture. This leads to the metal being distributed in a more efficient way as a catalyst.
4. Fungi produce large amount of enzymes per unit biomass.

The use of specific enzymes such as reductases secreted by fungi opens up exciting possibilities of designing a rational biosynthesis strategy for metal nanoparticles of different chemical composition.

A number of different genera of fungi have been investigated for biotechnological process research and it has been shown that fungi are extremely good candidates in the synthesis of nanoparticles. Because of their acceptance and metal bioaccumulation ability, fungi are accounted as major source of biological generation of metallic nanoparticles. A variety of advantages of using fungi as source to synthesis nanoparticles, this makes the understanding and handling of downstream processing easy in their scale up using a thin solid substrate fermentation method and gives the knowledge about simple and cheap raw materials for processing, furthermore, this fungal biomass is easy to handle. In fungi as a source of nanoparticle production either intracellular or extracellular nature occurs depending upon the reduction enzymes present in it. In other words the nanoparticles were formed extracellular when the cell walls reduction enzymes were responsible for metal ions reduction as well as when the reduction enzymes secreted extracellular. While intracellular synthesis in principle may accomplish a better control over the size and shape distributions of the nanoparticles, product harvesting, and recovery are more cumbersome and expensive. The extracellular synthesis by comparison is more adaptable to the synthesis of a wider range of nanoparticles systems. Recent studies reveal that fungi sources are extremely efficient secretors of extracellular enzymes, it is thus possible to easily obtain large-scale production of enzymes. From the observation of scientific publications realize that the application of produced nanoparticles more efficient if they synthesised outside of the fungal biomass. Furthermore, the advantages of using fungal source leads to increase the economic viability of product, harvesting and handling of biomass are also easier.

The fungus *Verticillium sp*. produces the metallic nanoparticles, when exposed to gold and silver ions; metal ions reduced by biomass fairly and formed respective metallic nanoparticles [41]. The fungal biomass of *Verticillium sp*. resulted in formation of intracellular nanoparticles with 2-20 nm size dimensions, when exposed to aqueous $AgNO_3$ solution, while *Fusarium oxysporum* resulted in extracellular silver nanoparticles size range in 2-50 nm [42]. The formation of well defined dimensions and good monodispersity gold nanoparticles were produced with an experiment where bioreduction of aqueous $AuCl_4^-$ ions using *Verticillium sp*. [43]. These findings provides the idea about the extracellular synthesis of nanoparticles using fungal strain, trapping of $AuCl_4^-$ ions on surface of fungal cells could performed by electrostatic interaction combined with positively charged groups such as lysine residues in enzyme that available in the mycelia cell wall. Furthermore, the enzymes that reduce the gold ions within the cell wall this led to aggregation of metal atoms and formation of gold nanoparticles [43]. Based on the fungal/enzyme mediated synthesis of nanoparticles research, towards adopting mechanism of an in vitro approach for nanoparticle formation was admitted with strain specific NADH dependent reductase, released by *Fusarium oxysporum*, were successfully used for the reduction of $AuCl_4^-$ ions to gold nanoparticles [44]. The produced nanoparticles were stabilized using the proteins and reducing agents concealed by the fungal strain. Normally the fungal biomass has ability to produce/release minimum of four high molecular weight proteins in connection with nanoparticles. One of these was species specific NADH dependent reductase. Fluorescence spectra shows that the native form of these high molecular weight proteins present in the solution as well as bound to the surface of nanoparticles [45,46]. Importing the mechanism of

NADH dependent nitrate reductase and phytochelatin isolated from *Fusarium oxysporum* has been used for in vitro silver nanoparticle production [47]. More recently, the synthesis of highly luminescent CdSe quantum dots using *Fusarium oxysporum* when incubated with a mixture of $CdCl_2$ and $SeCl_4$ at controlled room temperature [48]. Agriculturally important and non-pathogenic fungal strain, *Trichoderma asperellum* used for green synthesis of highly stabilized nanocrystalline silver particles [49]. The demonstrations of green synthesis of nanoparticles include two steps: first the bioreduction of silver ions to produce silver nanoparticles and followed by second step stabilization and/of encapsulation of the same by using a suitable capping agent. Table 2 presents some of the valuable published patents and publications regarding production of nanoparticles using biological material as sources. For the complete list of biological sources, please refer to Table 2.

2.3. Biogenesis of nanoparticles by yeast

Industrially important strain of yeast (eukaryotic microorganism) has been found to be a prominent candidate for biological synthesis of quantum semiconductors. For the first time, the biological synthesis of cadmium sulphide (CdS) quantum nanocrystals was produced by the strain of Candida glabrata. The yeast biomass produces the intracellularlly, monodispersed spherical shaped quantum nanocrystallites using cadmium salts and by neutralizing the toxicity of metal ions (metal-thiolate complex). The synthesised nanocrystals are now used in quantum semiconductor [62]. The strain of *Schizosaccharomyces pombe* were used to improve the quantity of semiconductor nanocrystals and this strain produces hexagonal lattice structured CdS nanoparticles in mid-log phase (incubated with 1 mM Cd) in the range of 1–1.5 nm, maximum nanocrystals were obtained [63]. This finding gives an idea that the greater amount of formation of Cds nanocrytals mainly depends on the nature of growth profile of yeast biomass. The mid exponential phase shows maximum production; at the same time the addition of CdS solution during stationary phase results the decreased and/or there is no formation of nanocrystals. Williams et al. [64] reported that the formation of CdS nanocrystals were found at early exponential phase of yeast growth, but this time it was affecting the cellular metabolism of the yeast and resulted in efflux of Cd from the cells. The reduction in formation of nanocrysatls in stationary phase because of a series of biochemical and metabolic reactions were generated to predict and prevent the toxic effects of metal. The mechanism involves two different steps, initially, an enzyme named phytochelatin synthase activated to synthesize phytochelatins, this reaction leads to form a low molecular weight metal-thiolate complex and eventually transport complex to across the vacuolar membrane by an ATP-binding cassette-type vacuolar membrane protein (HMT1). Ortiz et al. [65] documented that the addition of sulphide to the metal-thiolate complex in the membrane and that results in formation of high molecular weight PC-CdS^{2-} complex that allow them to sequestered into vacuole. Kowshik et al. [66] reported that the intracellular synthesis of PbS nanocrystallites quantum semiconductor by yeast, *Torulopsis sp*. with a dimension of 2–5 nm in spherical morphology when incubated with $Pb2^{+}$ exhibiting wavelength of 330 nm in UV–Vis spectrophotometer. Extracellular production of silver nanoparticles was reported using silver tolerant yeast strain MKY3, which synthesized hexagonal silver nanoparticles (2–5 nm) in log phase of growth. The proper condition for the synthesis of large scale quantities of silver nanoparticles also standardized and documented that was based on differential thawing of the samples [67]. Recently, yeast biomass has been identified for their ability to produce gold nanoparticles, whereby controlling growth and other cellular activities controlled size and shape of the nanoparticles was achieved [68]. More recently, the strain of *Yarrowia lipolytica* NCIM 3589 was found to be a good candidate for synthesis of gold nanoparticles associated with cell wall. The reduction of gold ions occurred in pH dependent manner, at pH 2.0, it produced hexagonal and triangular gold crystals due to the nucleation on the cell surfaces [69]. For the complete list of biological sources, please refer to Table 2.

2.4. Biogenesis of nanoparticles by virus

Eco-friendly microbial synthesis of nanoparticles have been received great attention and extended towards intact biological particles (viruses). The biological molecules are plays a vital role in growth of semiconductor as a template, biological molecules includes, fatty acids, amino acids, and polyphates. For example, by interchanging the ratio of different fatty acids (chain lengths), different nature of CdSe, CdS, and CdTe nanocrystals can be achieved. Similarly the variety of other biological materials are also involves in synthesis of inorganic materials. The other

important bio-factories like DNA [71-73], protein cages [74], biolipid cylinders [75,76], viroid capsules [77], bacterial rapidosomes [78], S-layers [79] and multicellular superstructures [80] are used as template-mediated production of inorganic nanomaterials and microstructured materials. Interestingly, viral scaffolds were found to be a template for the process of nucleation and assembly of inorganic materials. Certainly, cowpea chlorotic mottle virus and cowpea mosaic virus have been used as nucleation cages for the mineralization of inorganic nanomaterials. In addition to this, tobacco mosaic virus (TMV) has been used as template for the successful synthesis of iron oxides by oxidative hydrolysis, co-crystallization and mineralization of CdS and lead sulphide (PbS) crystalline nanowire, and the synthesis of SiO2 by sol–gel condensation. The process happened with the help of external groups of glutamate and aspartate on the external surface of the virus. Furthermore, peptides capable of nucleating nanocrystal growth have been identified from combinatorial screens and displayed on the surface of M13 bacteriophage. A hybrid nanowires (ZnS-CdS) are obtained with a dual peptide virus engineered to express A7 and J140 within the same viral capsid [81-83]. For the complete list of biological sources, please refer to Table 2.

2.5. Biogenesis of nanoparticles by plant source

The biological synthesis of nanoparticles using microbial sources implies success in producing the metallic and inorganic nanoparticles. Taking this idea to one step further development with the plant and plant materials mediated synthesis of nanoparticles. In recent years plant mediated biological synthesis of nanoparticles is gaining importance due to its simplicity and ecofriendliness. Plant material and plant mediated synthesis of nanoparticles involves the prediction and potential elimination of chemical derivatives and its effect to the nature. The biosynthesis of nanoparticles by using plant materials involves fairly rapidly to reduction of metallic materials. Shankar et al. [84] reported that the extracellular synthesis of silver nanoparticles by *Pelargonium graveolens* (the extract of geranium leaves) for reduction of Ag^+ ions to Ag^0 nanoparticles. This document provides detailed knowledge about the reduction of silver ions reaction much faster than the earlier studies with bacteria and fungi. Certainly, the time required for complete reduction of the metal ions using bacteria and fungi ranged from 1 to 5days; in contrast, more than 90% of the reaction using extract of geranium leaves is complete within 9 hours. Living plant and plant materials have been observed as an important candidate for synthesis of quantum dots, quantum dots have wide applications in nanobiotechnology. *Alfalfa* roots have ability of absorbing reduced silver ions (Ag^0) from production medium and transferring into shoot of the plant material in the same state of oxidation. Inside the plant tissue the reactions takes place, the accumulation of Ag atoms underwent nucleation and resulted in formation of nanoparticles with the help of Ag atoms are joining them to form larger arrangements [85].

In a mechanistic study, Armendariz et al. [86] have reported that synthesis of gold nanoparticles by *Avena sativa* (Oat) biomass and subsequent formation of gold nanoparticles of variable sizes. In this study, the synthesis of gold nanoparticles highly dependent on the pH outside and rod shaped nanoparticles were formed. The variation in size and shape of nanoparticles depends on pH and interaction to the binding site. The Au (III) is normally act as an anion in aqueous medium and at low pH values, biomass might carry more positive functional groups that allow the Au(III) ions to get more closure to binding site. At low pH 2, larger nanoparticles (25-85 nm) were found in small quantities. Surprisingly, smaller nanoparticles (5-20 nm) in larger quantities were formed at pH 3 and 4. The low pH favours the aggregation process for formation of larger nanoparticles over the nucleation to form new nanoparticles. The increase in pH 3 and 4 results in reduction of size, because number of functional groups is increased for gold binding, thus a higher number of Au (III) complexes bind to the biomass at the same time. This leads to the subsequent formation of larger quantities of nanoparticles with smaller diameters. Hence the size and shape of the nanoparticles can be controlled by changing the outside pH of the reaction conditions. Synthesis of triangular gold nanoprisms by the plant extract of lemongrass (*Cymbopogon flexuosus*) has been reported [87]. The extracellular synthesis of highly stable silver and gold nanoparticles has been achieved with the use of *Emblica officinalis* fruit extract acting as a reducing agent [88].The authors extended their work for rapid synthesis of stable ans size controlled gold nanotriangles at high concentration using tamarind leaf extract acting as a reducing reagent [89]. Most recently, potential production of gold and silver nanoparticles has been achieved from *cinnamomum camphora* leaf extract. Mainly the polyol components and water soluble heterocyclic components were found to be respon-

sible for the reduction of silver ions or chloroaurate ions and the stabilization of the nanoparticles, respectively [7]. For the complete list of biological sources, please refer to Table 2.

2.6. Biogenesis of nanoparticles by algae

Review of literature revealed that the synthesis of nanoparticles using algae as source has been unexplored and underexploited. More recently, there are few, reported that algae being used as a biofactory for synthesis of metallic nanoparticles. In an isolated report, Singaravelu et al. [93] implemented an efficient approach for synthesis of stable gold nanoparticles by the reduction of aqueous $AuCl_4^-$ by using *Sargassum wightii*. Interestingly, this the first report for synthesis of stable metallic nanoparticles by the extract of marine algae, it results relatively very short period of incubation time requires compared with other biological materials. The reduction of the gold ions resulted in the formation of high density, extremely stable gold nanoparticles in the size ranging from 8 to 12 nm with average size of 11 nm. Certainly, 95% of the bioreduction of $AuCl_4^-$ ions take place within 12 h at agitation condition. The researchers prolonged their work to the formation of palladium and platinum nanoparticles starting with their corresponding metallic chloride containing salt solution [93]. For the complete list of biological sources, please refer to Table 2.

3. IMPORTANCE OF PHYSICOCHEMICAL AND BIOMECHANICAL PROPERTIES

Various factors of physicochemical and biomechanical properties influence the bioreduction of metal ions into metallic nanoparticles in biological systems like temperature, pH and agitation speed etc. Produced metal nanoparticles size and stability have strong dependence on physicochemical and biomechanical properties, which allows nanoparticles as a physically powerful candidate for various applications such as biocatalysis, biosensing, recording media, and optics [95-102]. The physical property like temperature has an immense effect on the formation of nanoparticles. Review of literatures shows that the various temperature conditions represents different kinetic and structural characteristics. At mild reaction temperature condition aids in the formation of stable and increased quantities of production with defined dimensional size distributions. At the same time the increased temperature shows the decreased particle size and narrower size distribution. It is normally attributed to the increased reaction rates at higher temperatures. Experimental demonstration of gold nanotriagnles and spherical nanoparticles showed that the temperature plays a major role for controlling the ratio size and relative amount of production. By changing the temperature from lower to higher, the reaction conditions, the shape, the size, the amount and optical properties of the anisotropic nanoparticles can be finely tuned [103]. Kinetically controlled and highly stable gold nanoparticles and gold triangles observed at the low temperature range [104]. In addition to this, pH of the production medium influences a lot to the size and stability of synthesized nanoparticles. Formation of gold nanoparticles from plant extract of *Avena sativa,* it was observed that size of the gold nanoparticles can be controlled by altering the pH (lower to higher) of the medium [105]. Other factor like chloride, bromide and iodide ions also affects the nanoparticles formation in plants. Presence of chloride ions regulates and induces the growth of nanotriangles during synthesis, whereas presence of iodide ions distorts the nanotriangles morphology and induces the formation of aggregated spherical nanoparticles [103]. The controlling particles morphology at the different pH levels would play an important role during optimization of a process. Brown et al. [106] demonstrated the role of repeating polypeptides in controlling gold crystal growth, the variety of changes in the shape of crystals with different pH. The outcome of this demonstration aids in the control of gold crystal morphology was attributed to pH control (regulation of proton concentration, rather than to general acid catalysis).

Growth conditions play an important role throughout the production of nanomaterials while using the biological culture. The fungal strain of *Trichothecium sp*. produces nanoparticles in both forms extracellular and intracellular. The strain produces the extracellular gold nanoparticles under stationary conditions. The remarkable reason for this is the presence of enzymes and proteins led to the formation of extracellular nanoparticles. The bioreduction enzymes and proteins were released into the medium under stationary conditions and not in shaking conditions. The same strain results the formation of intracellular gold nanoparticles with shaking conditions, because of absence of enzymes and proteins [107]. Extracellular protein mediated hydrolysis of the anionic complexes reports in the facile room temperature synthesis of crystalline ti-

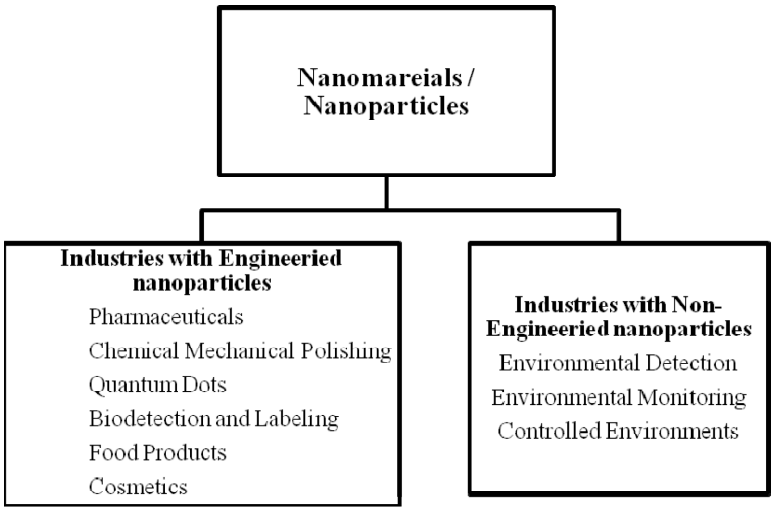

Fig. 4. Examples of industries with engineered and non-engineered nanoparticles.

tania particles while calcinations required at 300 °C for crystallization of silica [108]. The synthesis of magnetic nanoparticles using two fungal strains, *Fusarium oxysporum* and *Verticillium sp*. at controlled room temperature reported [56]. These strain secreted proteins which were capable of hydrolyzing iron precursors extracellularly to form iron oxides predominantly in the magnetite phase.

4. APPLICATIONS AND POTENTIAL USES OF NANOPARTICLES

Nanomaterials are at the leading edge of the rapidly developing filed of nanotechnology. Nanoparticles are important in interdisciplinary research, in general these can be classified in to two broad groups:
(1). Engineered nanoparticles.
(2). Non-engineered nanoparticles.

Engineered nanoparticles are produced and designed to attain the specific applications with their physicochemical properties. Non-engineered nanoparticles are unintentionally generated products, such as atmospheric nanoparticles produced during combustion. Either way, the particle's physical properties are extremely important to their performance and the performance of any product into which they are ultimately incorporated. The applications of engineered and non-engineered nanoparticles used in major industries for manufacturing goods, examples of industries shown in Fig. 4.

In last two decades, biosynthesized nanoparticles has wide range of applications in drug delivery systems, acts as strong antimicrobial sources, chemical sensing, biochemical catalysis, and biosensors etc., [109-112]. Applications of produced nanoparticles in biology and biotechnology fields are listed in Table 3. Nanotechnologies acts prospective role in stem cell biology research includes (1) tracking of stem cell surface molecules and detailed examination of molecular motion without photobleaching, (2) noninvasive tracking of stem cells and progenitor cells transplanted in vivo, (3) stem cell delivery systems that enhance the survival of transplanted cells by releasing prosurvival biomolecules, (4) nanopatterned substrates that present covalently tethered biologically active molecules (adhesion sites, growth factors, and synthetic

Table 3. List of application of nanomaterials in biology and biotechnology.

Applications of nanomaterials in biology/ biotechnology	References
Fluorescent biological labels	[145-147]
Drug and gene delivery	[148, 149]
Bio detection of pathogens	[150]
Detection of proteins	[151]
Probing of DNA structure	[152]
Tissue engineering	[153, 154]
Tumour destruction via heating (hyperthermia)	[155]
Separation and purification of biological molecules and Cells	[156]
MRI contrast enhancement	[157]
Phagokinetic studies	[158]

peptides) for stem cell differentiation and transplantation, and (5) intracellular delivery of DNA, RNAi, proteins, peptides, and small drugs for stem cell differentiation [113,114].

As mentioned earlier, only some microorganisms have resistance to the metal ions, when microorganisms exposed to metal ions it reduces the ionic strength and produces the corresponding metallic nanoparticles. Metallic nanoparticles, includes siver, CdS and gold have resistance to microbial disease. To our knowledge gold nanoparticles can act as slow acting drug in rhematology and silver nanoparticles is highly toxic to the some microbial cells and it can be act as a antimicrobial agent of biocide. The special attention towards the silver nanoparticles because of their nature of strong antimicrobial activity either in metallic nature and nanoparticles form also, so it found that silver nanoparticles has different applications to the environment and human,

1. Trace amount of silver are particularly good as decontaminating agent in water and have been found useful to prevent biofilm formation in food-contact surfaces [115].
2. The antimicrobial nature of silver ions plays a promising role in food packaging systems. Silver ions are preventing the growth of fouling contaminants on processing and improve the shelf life period of food. Different silver technologies are approved by the Food and Drug Administration (FDA) and the European Food Safety Authority (EFSA) and are being employed in aseptic surfaces. Silver exchanged zeolites are also being incorporated into food contact polymers, as polyethylene, polypropylene or nylon [116]. Silver based antimicrobial fillers in polymer matrices base their antimicrobial activity on a sustained release of silver ions [117].
3. Silver ions involves in the ATP production, it inhibiting the expression of enzymes in the progressive action with ribosomes. The mechanism of production of ATP, silver ions interact with cytoplasmic components and nucleic acids, and to interfere with the membrane permeability [118]. Additionally silver ions probably interacts with the respiratory chain of Escherichia coli cells [119].
4. Silver nanoparticles have antibacterial properties mediated by silver ions, which are probably chemisorbed in the partially oxidized nanoparticles [120].
5. Silver sulfadazine shows slow and steady reaction with serum and other body fluids, it acts better healing of burn wounds [121].
6. Silica gel micro-spheres mixed with silica thiosulfate are used for long lasting antibacterial activity [122].
7. It acts a good preservative in food and various food related products [122].
8. Treatment of burns and various infections [123].
9. In medicine and medicinal devices like surgical masks and implantable devices show significant antimicrobial efficacy [124].
10. Silver nanoparticles can be used for water filtration [125].
11. The silver nanoparticles are reported to show better wound healing capacity, better cosmetic appearance and scarless healing when tested using an animal model [126].
12. The nanocrystalline silver dressings, creams, gel effectively reduce bacterial infections in chronic wounds [127-129].
13. The Fe_3O_4 attached Ag nanoparticles can be used for the treatment of water and easily removed using magnetic field to avoid contamination of the environment [130].
14. The silver nanoparticle containing poly vinyl nanofibres also show efficient antibacterial property as wound dressing [131].
15. Environmental-friendly antimicrobial nanopaint can be developed [132].

Nanotechnology applied to make nanoscale impound environments channels or post arrays for long polymers such as DNA [133]. The DNA molecule could be used as a 'conveyor-belt' for attached molecules, and also used in the manufacture of automated nanochip sensors, therapeutic, diagnostic and tissue engineering devices to regulate biological processes [133]. A vast array of new signal transduction technologies have been implemented to produce biosensors, bioprobes, biochips, biotransducer and other biological systems using nanomaterials synthesized from living organisms [134]. Biosignal transducer are similar to semiconductors except that instead of having electronic circuits, they have biological material, DNA, RNA or protein, attached to the surface of a "chip", which can be glass, plastic or silicon. For biosensors, VoDinh et al. [135] provide a simple definition, a device consisting of a bioreceptor, the biorecognition system, and a transducer. The bioreceptor produces a physical effect that is converted to a measurable signal by transducer. Common biorecptor systems utilize the interactions of antibodies and antigens, nucleic acids, enzymes, cells, or synthetic biomimetic materials. With the advances made in micro and nanofabrication, miniature microelectro-

mechanical systems (MEMS) show great promise as potentially catheter based and/or implantable biosensors. The categories of nanobiosensors include optical based, nanoparticles and nanowire etc.

The gold nanoparticles have great importance in area of research due to their unique and tunable applications in biomedical science, tissue engineering and MEMS including drug delivery, tissue/tumour imaging, delivery of biomolecules, for protein delivery, photothermal therapy and immunochromatographic identification of pathogens in clinical specimens [136]. Mainly the gold nanoparticles provide non-toxic environment carriers for drug and gene delivery applications. Recently, gold nanoparticles have been identified as an attractive candidate for delivery of various payloads into their targets. The payloads could be small drug molecules or large biomolecules, like proteins, DNA, or RNA [137,138]. And also gold nanoparticles applied for delivering large biomolecules too, without restricting themselves as carriers of only small molecular drugs. They have shown the success in delivery of peptides, proteins, or nucleic acids like DNA or RNA. Gold nanoparticles have emerged as a promising scaffold for drug and gene delivery that provide a useful complement to more traditional delivery vehicles [139]. The gold nanotriangles (highly anisotropic planer shape) are find application in photonics, optoelectronics, and optical sensing. Furthermore, the gold nanotriangles biologically synthesized using tamarind leaf extract used as chemical sensors in vapour sensing [89].

The optical properties and surface plasmon properties of bacterial synthesized nanomaterials are used in coating process, the magnetite (Fe_3O_4)/ greigite (Fe_3S_4) and siliceous material produced from magnetotactic bacteria and diatoms, respectively, are approved and applied as optical coatings for solar energy production unit and used in electrical batteries as ion insertion materials [140]. The magnetosome particles isolated and synthesized from megnetotectic bacteria has been used as carrier for immobilization of bioactive substances such as enzymes, antibodies, DNA and RNA [141]. Recently, nanoparticles find applications in bioremediation of radioactive wastes such as uranium (a long lived radionuclide hazardous for both plant and animals) resulted from nuclear power plants and nuclear weapon production. Cells and S-layer proteins of Bacillus sphaericus JG-A12 have special capabilities for the clean-up of uranium contaminated waste waters [142]. Nanoparticles activates biomineralization, is the process where organisms form minerals. Mineralized skeletons of animals and plant derive their exceptional material properties from the extremely small size of the crystals comprising these nanostructures. Hybrid bionanomaterials can also be applied to build novel electronic, optoelectronics and memory devices [143,144].

5. CONCLUSIONS

Bionanotechnology has emerged up as integration between biotechnology and nanotechnology for developing bioactive, biosynthetic and ecofriendly technology for synthesis of nanomaterials. The present attention towards synthesis of metallic nanoparticles is based on their extensive potential applications in catalysis, microelectronics, biomedicine, and nanobiotechnology. The interaction at cell level increases the scope of investigating and regulating the synthesis of nanoparticles by synthetic and biological materials. Biological synthesis of nanoparticle is mainly developed for processing of clean, toxic free, effective and ecofriendly technologies. Use of biological materials is rapidly gaining significance owing to its growing success, cost effective process and simplicity. Biological materials ranging from simple prokaryotes to complex eukaryotic organisms including higher angiospermic plants and viruses are used for the nanoparticles synthesis. Current perspective of biological synthesis process should focus towards the use of highly structured physical and biosynthetic activities of microbial cells to achieve controlled manipulation of the size and shape of the particles. Further, more effect is needed in order to develop more productive process for metallic nanoparticle production. In addition, improvements on biogenesis process are needed for the development of cheaper processes.

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