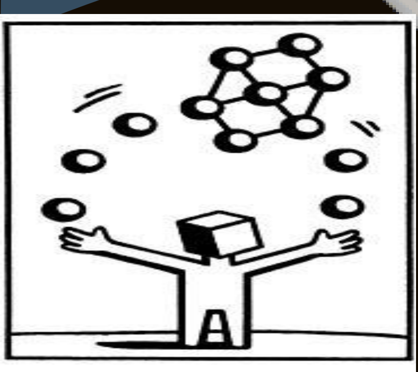
In the name of GOD

Biological synthesis of Nanoparticles



Contents

1 **Introduction**

2 **Synthesis Protocols of Nanoparticles=** نانوذرات فلزی (عنصری، سولفیدی، اکسید فلز)، نانوکپسول ها (دارورسانی ساده و هدفمند)، نانولوله های کربنی (تصویربرداری زیستی، ژن درمانی، شناسایی سلول های سرطانی)

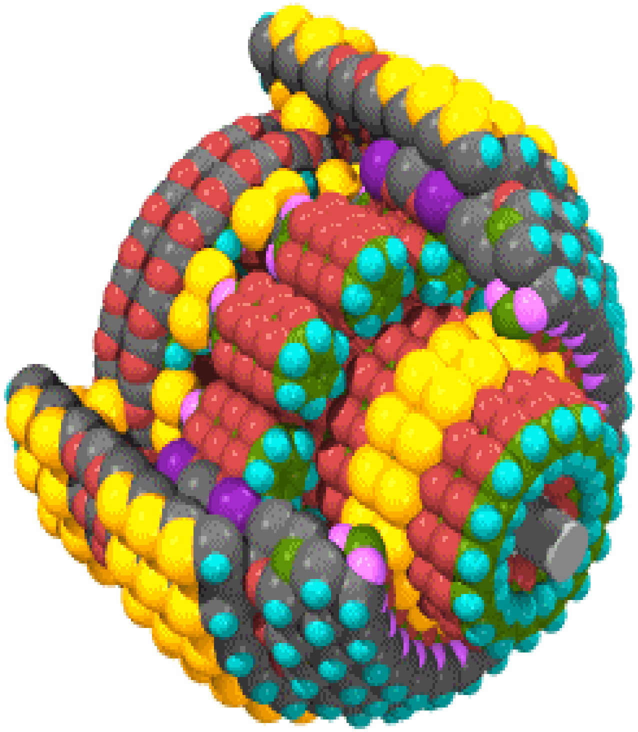
3 **The Formation Nanoparticles by Microorganisms and Their Applications**

4 **Conclusions**

- نانوذرات شامل نانوذرات فلزی، نیمه هادیها واکسیدهای فلزی هستند، بعنوان **کاتالیزورهای قدرتمند** عمل نموده که راندمان واکنش های شیمیایی را به شدت افزایش داده وهمچنین به میزان چشمگیری ازتولید مواد زاید درواکنش ها جلوگیری بعمل می آورند. بکارگیری نانوذرات درتولید مواد دیگراستحکام آنها را افزایش داده، وزن آنها راسبک کرده ، مقاومت شیمیایی وحرارتی آنها رابالا برده و واکنش آنها را دربرابر نور وتشعشعات دیگرتغییر می دهد، بنابراین درتولید **نانوکامپوزیت ها** استفاده می شوند. اخیرا درساخت شیشه های ضد آفتاب ازنانوذرات اکسید روی استفاده می شوند. استفاده ازاین ماده علاوه بر افزایش کارآیی این نوع شیشه ها ز،عمرشان را نیزافزایش می دهد. **ساخت حسگرهای زیستی**، **تجزیه آلاینده های یست محیطی**

Introduction

- History
- Nano? Why Nanotechnology?
- Nanofabrication?
- Market Capitalization

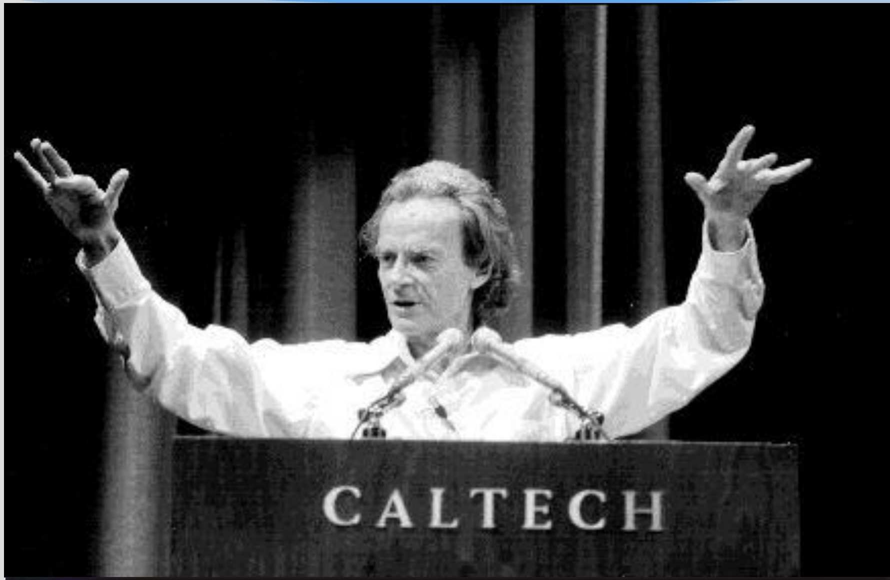


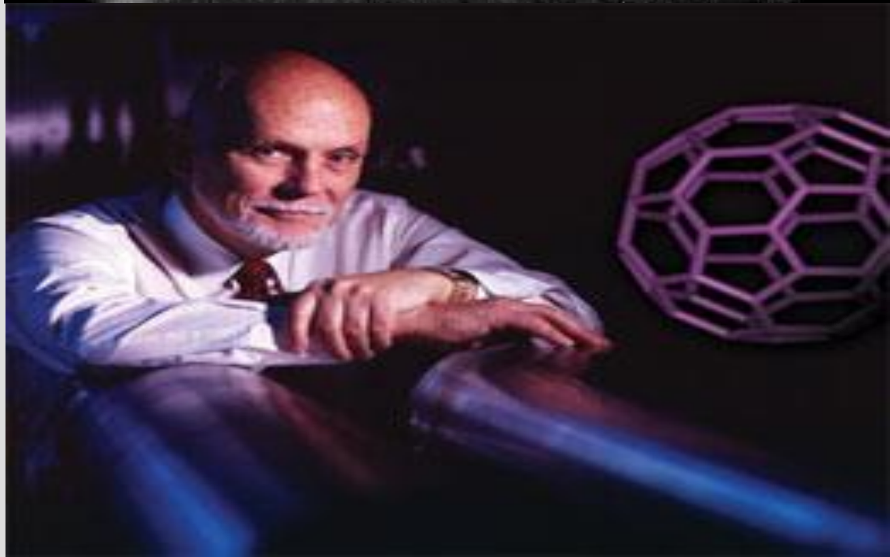
- **Richard Feynman**
- **(Nobel Prize1965)**

- **There is a plenty of room at the bottom**
- **(1960)**

- **فضای زیادی در سطوح پایین وجود دارد**

- **Richard Smalley**
- **(Nobel Prize 1966)**
- **presentation molecular nanomachines model**



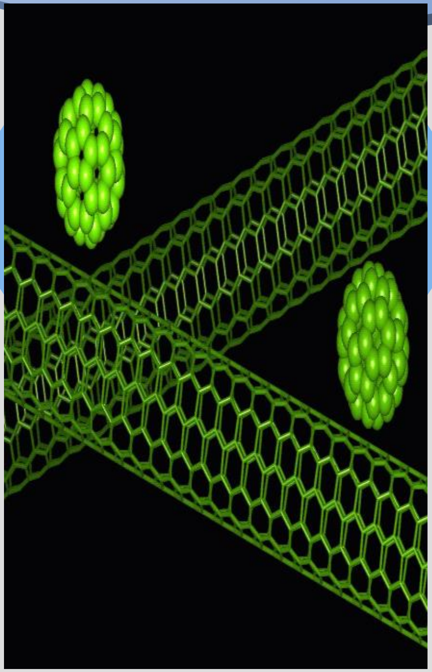


- Nano: Dwarf (10^{-9} m)
- Nanoparicles (Nanoclusters): **particles that shorter than 100 nm at least in one dimension according to a recent definition of the European Union (BSI-PAS71,2006).**

- **Particular Properties of Nanoparticles:**
- **1) anything smaller than 100 nm is no longer subject to laws of classical physics but of quantum physics.**
- **2) Increasing the ratio of S/V**

- **Conclusion: nanoparticles have unique optical, electronic, magnetic and chemical properties and it is expected nanotechnology will be developed at several levels: materials, devices and systems.**

Nano biotechnology



Top-Down:

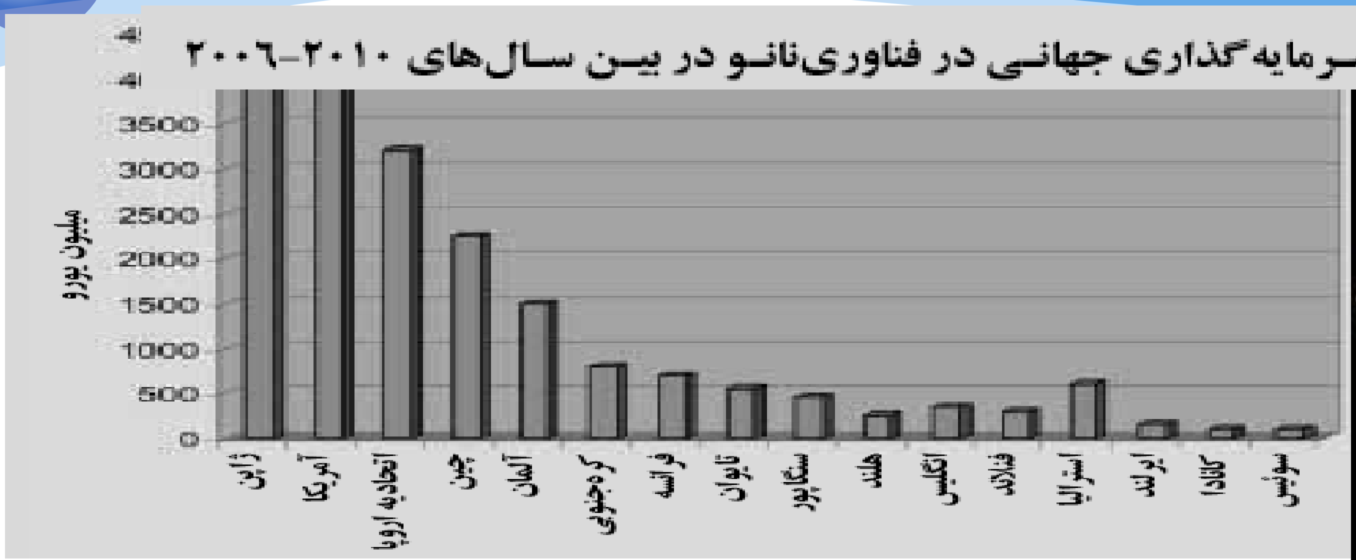
By: physical, chemical and mechanical milling techniques.

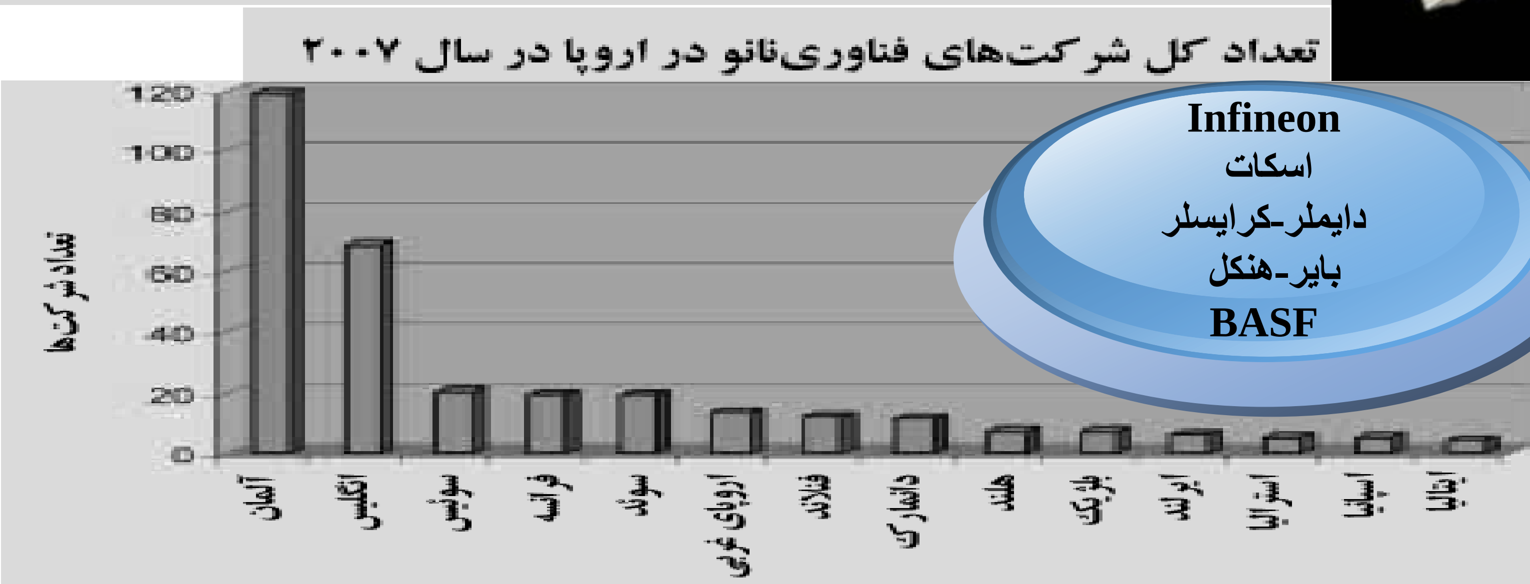
Bottom-Up

By :Scanning Probe Microscopy :

- STM
- AFM
- Heinrich Rohrer(Nobel Prize 1986)
- 2) molecular self-Assembly

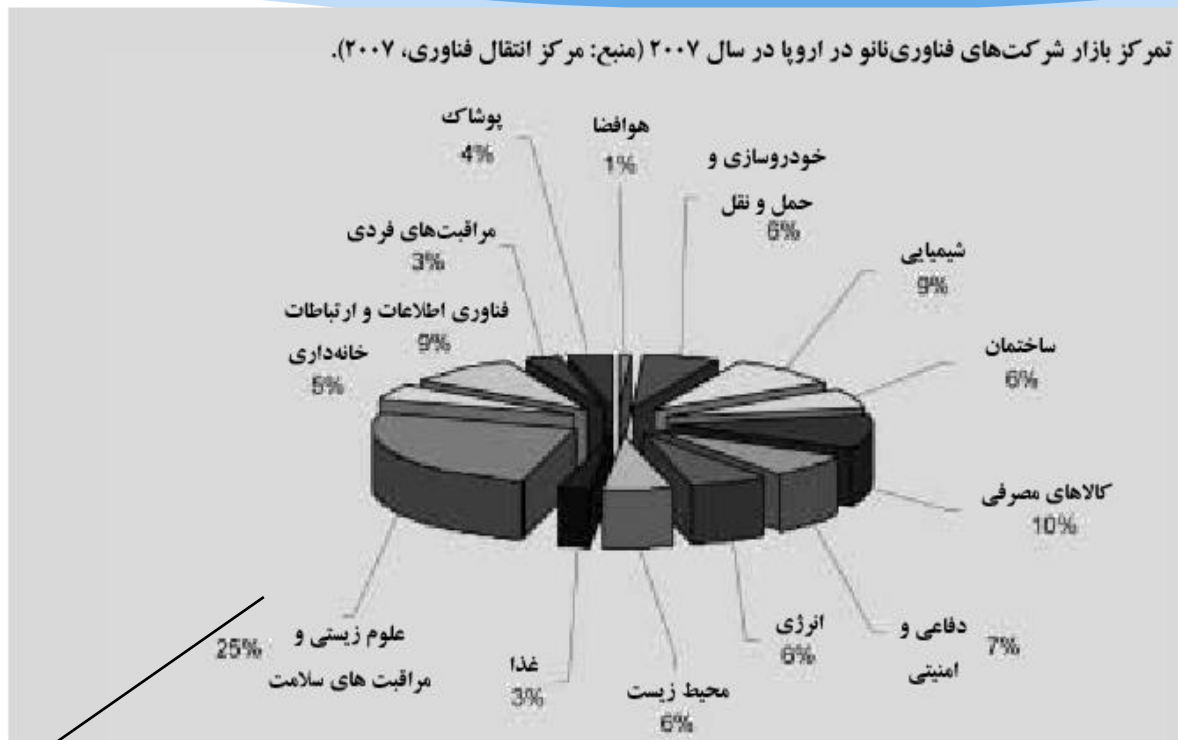
Market Capitalization



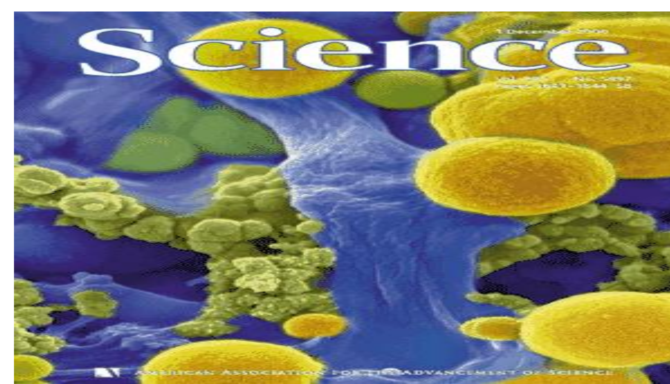


Infineon
اسکات
دایملر-کرایسلر
بایر-هنکل
BASF

- ► Drug and gene delivery
- ► Biodetection of pathogens
- ► Detection of proteins
- ► Probing of DNA structure
- ► Tissue engineering
- ► Tumour destruction
- ► Separation and purification of biological molecules and cells
- ► Antimicrobial Agents
- ► Biolabeling



Applications of nanoparticles in biology and medicine

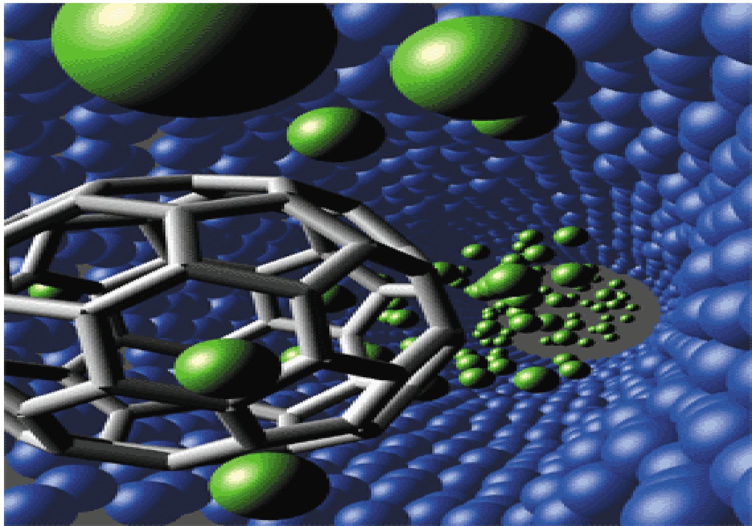


synthesis of nanoparticles

- 1) physicochemical synthesis

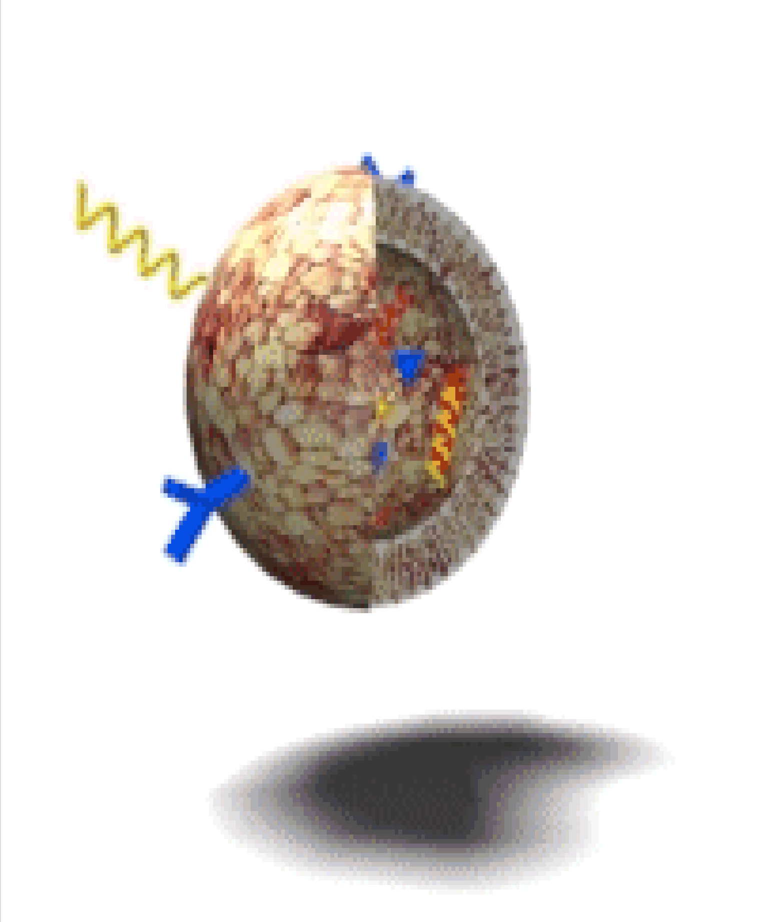
- 2) biological synthesis

- Why microbial synthesis: advantages over chemical synthesis



physicochemical synthesis

- **1) physical vapour deposition (PVD)**
- ►evaporation
- ►laser ablation
- **2) mechanical milling**
- **3) sol-gel**
- **4) electrochemical reduction**
- **5) thermal decomposition**
- **6) photolytic reduction**
- **7) spray pyrolysis**
- **8) chemical reduction**



chemical reduction

Nanoparticle formation by chemical reduction

- **Experimental**
- 1) Template (deposition onto alumina/silica) and surface pretreatment
- 2) precursor solution
- 3) Reducing agent
- 4) stabilizing agent (capping agent) =dispersing agent
- 5) Recovery of nanoparticle

Analytical Study

- **Advantage: Rapid**
- **Disadvantage:**
- **1) Low Monodispersity (15-25%)**
- **2) Low Stability**
- **3) Impurities**
- **4) high materials and energy use (expensive)**
- **5) Environmental disruption**
- **6) contamination due to side products or instability**

Why microbial synthesis?

- 1) relatively mild reaction conditions
- 2) high substrate specificity
- 3) fewer environmental problems
  - (green chemistry)
- 4) high monodispersity (40-50%)
- 5) high stability
- 6) cost-effective

Metal-microbe interactions

Metal-Microbe Interaction play an important role in several biotechnological Applications including:

► **Biomineralization**

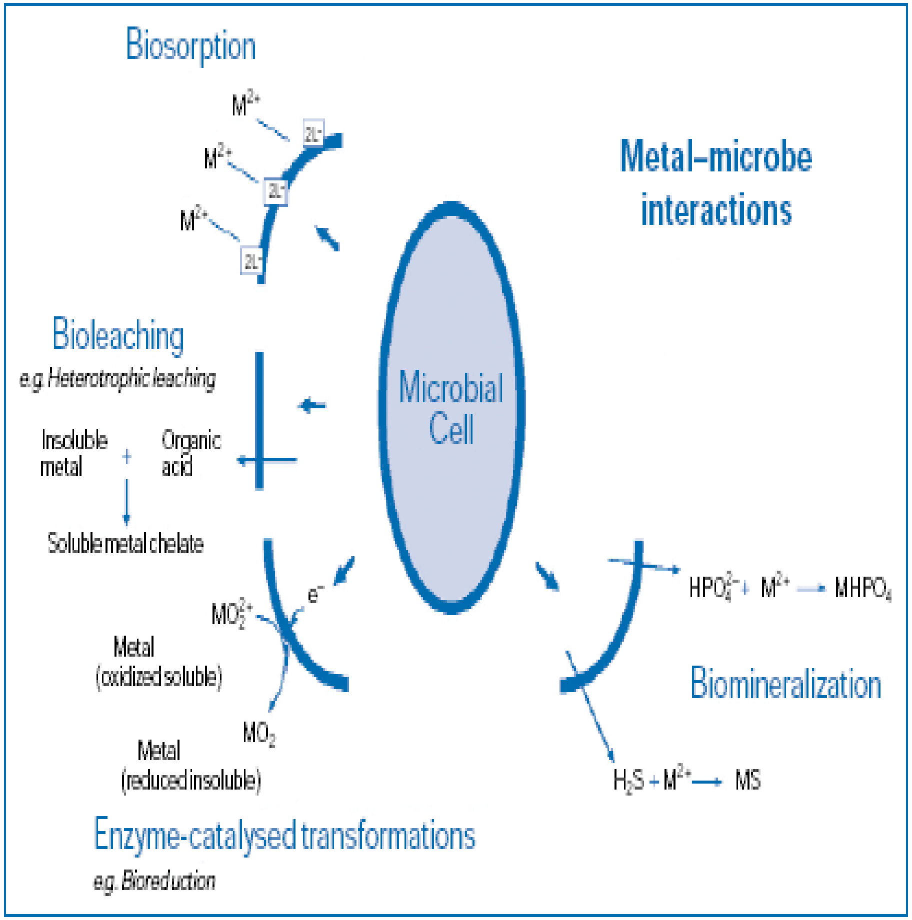
► **Bioremediation (bioreduction)**

► **Bioleaching**

► **Microbial crossion**

► **Recently: eco-friendly nanofactories**

Metal-ion: non-biodegradable and persistent in nature

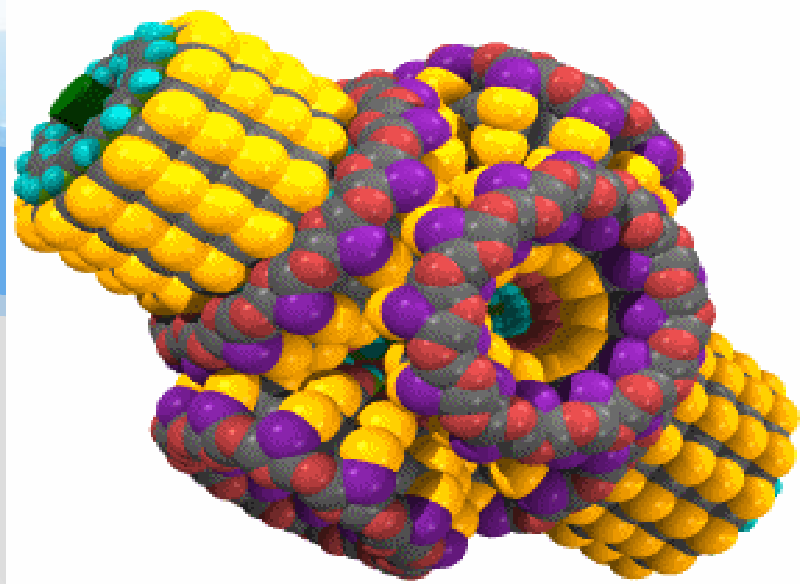


Metal-ion resistance mechanisms:

- 1) efflux systems
- 2) **alteration of solubility and toxicity via reduction and oxidation**
- 3) biosorption

Remark: genetic and/or physiological adaptation

- 4) bioaccumulation
- 5) extracellular precipitation
- 6) detoxification via metal-binding proteins
- 7) lack of specific metal transport system



Bioreduction Mechanism

Enzymatic Reduction:

► **Particular reductases and**

► **The terminal oxidases of the respiratory chain**

Non-Enzymatic Reduction:

► **Reduced forms of cellular and other reducing groups** such as aldehyde , ketone, thiol , etc.

Nanoparticle Formation

(upstream processing)

Inoculum Preparation → **Nanoparticle preparation** → **Nanoparticle characterization**

Phase 1

Phase 2

Phase 3

Inoculum Preparation:

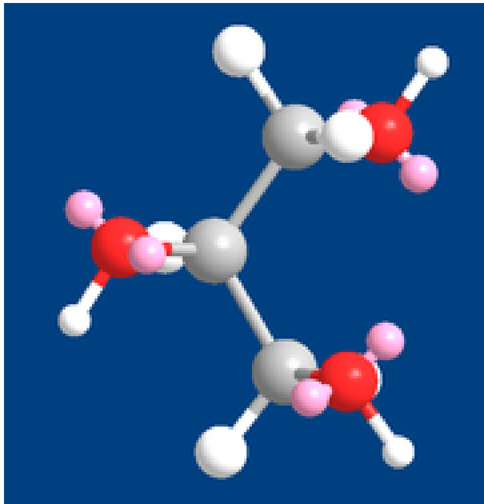
- WC: whole cells (growing/resting cells) in liquid and solid culture= wet biomass
- CS : culture supernatant
- CFE: cell extract of disrupted cells
- CD: cell debris
- Dried Biomass (60^o C for 24 h)= biosorbent and in situ bioreduction ►biosorption capacity: 50-100mg/g

❖ Nanoparticle Preparation:

- Suitable Inoculum + suitable precursor ion complex ⟶ incubation under suitable conditions.

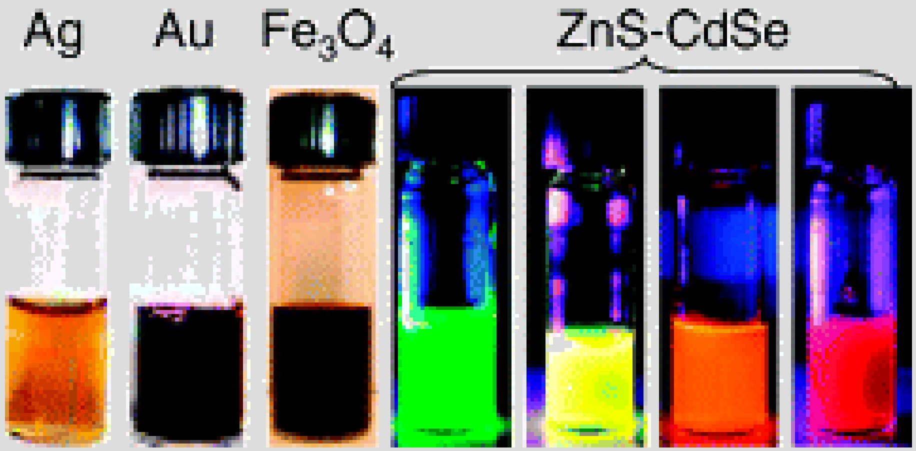
❖ Nanoparticle Characterization :

- **1) Visual Observation (change in color ?)**
- **2) Analytical Studies (Spectroscopy and Microscopy)**

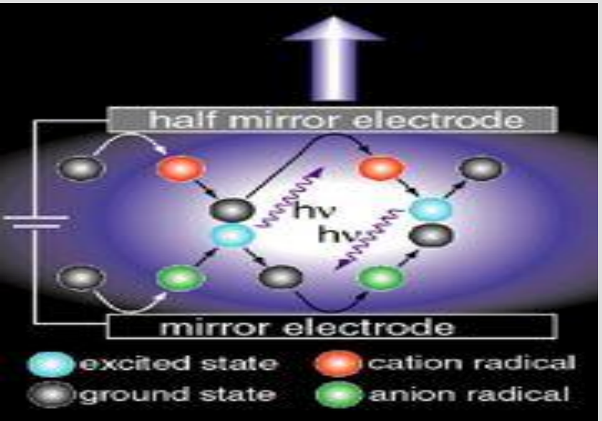


❖Visual Observation

- **Change in color (biomass/solution) :**
- 1) the sign of reduction
- 2) an indication of formation of colloidal particles , due to the excitation of surface plasmon vibrations (typically Ag, Au): confirmed by SPR band in UV-visible spectroscopy.



Nanoparticle	color	SPR band
Ag	Light yellow-brown*	425 nm
Au	Pink-purple*	545 nm
Ag/Au alloy	Blue*	537 nm
Pt	Black*	488 nm
Cu	Light brown-black*	612 nm

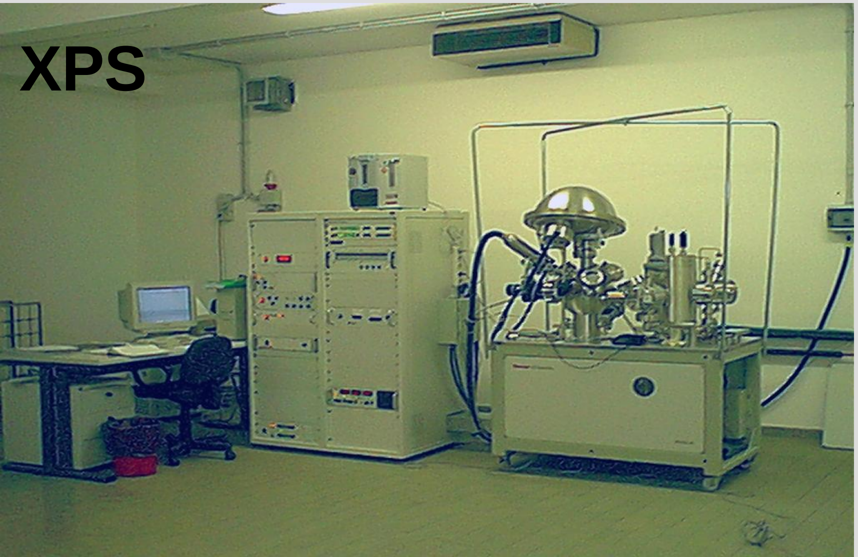


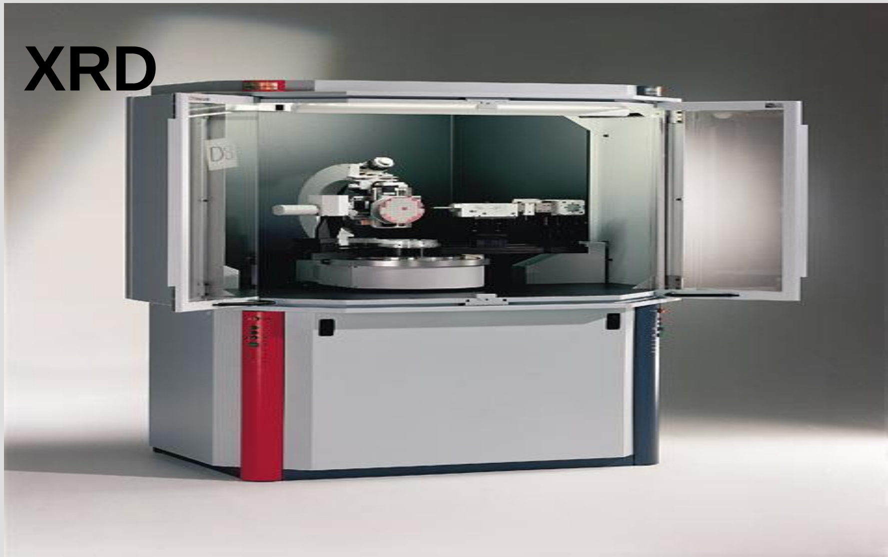
❖ TEM/EDX and SEM/EDX Analysis

- ❖ **LRTEM: Identification disperse particle from aggregated particle.**
- ❖ **HRTEM: Identification morphology, size (Average diameter), location, number of particles produced per cell.**
- ❖ EDS **(EDX=Energy dispersive X-ray spectroscopy):**
  - ❖ **Elemental Analysis**

❖ XRD/ XPS/FTIR Analysis

- **XRD (X-ray diffraction):** **1) elemental Analysis 2) chemical composition Analysis , 3) size (D=0.9λ/β.cosθ , Scherer)**
- **XPS (X-ray photoelectron spectroscopy):1) elemental Analysis 2)chemical composition Analysis , 3) Structural Analysis***





❖ FTIR /EXAFS Analysis

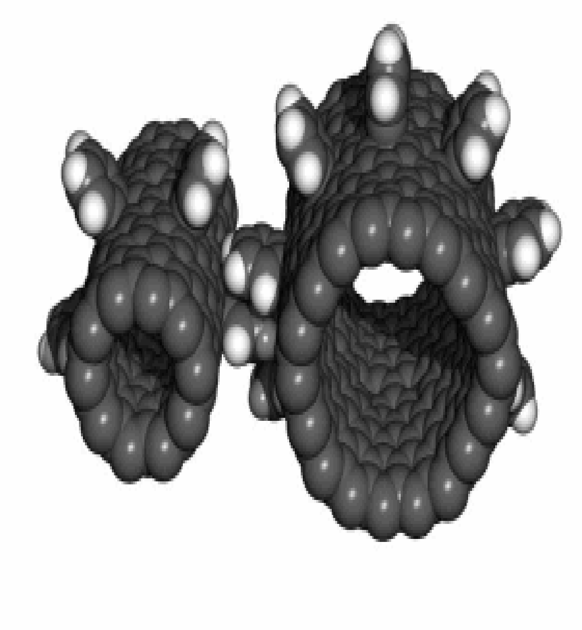
- **FTIR (Fourier Transform infrared spectroscopy):**
- **Structural Analysis (identification functional group :-OH, -C=O, -NH, -CHO, etc)**
- **<u>EXAFS*</u> (Extended X-ray Absorbtion Fine Structure) :**
- **Structural Analysis (identification functional group :-OH, -C=O, -NH, -CHO, etc)**

Quantitative Analysis

- **Uv-vis**
- **AAS**
- **ICP-MS**
- **HPLC**
- **GC-MS**

Nanoparticles Formation & Microorganisms

- Microorganisms: **Nanoparticle Formation:**
- 1) intracellular/surface reduction
- 2) Extracellular reduction
- Harvesting by ultrasonic treatment or reaction with detergents.
- Bacteria
- Yeasts
- Fungi
- Actinomycetes
- First Study: beveridge and Mourray (1980)., **Au and Ag**
- **Nanoparticle formation after 7 days under pH 7 and T=27°C.**
- **Monodispersity(5-25nm)., shape: octahedral, intracellular**.
- ***bacillus sutilis strain 168***. **(drawback)**



Types of Nanoparticles

1

Metallic nanoparticles:

Ag, Au, Ag/Au alloy, Pt, Pd, Te,Zr, Se, **Cu*, Co*,Mo*, Cr*, Fe*, Ni***

Application: as a novel catalysts.

2

Metal sulfide= semiconductors = quantum dots

ZnS, PbS, CdS, FeS

Application: as a new class of materials for biological detection and cell imaging (**quantum dot lasers)**

3

Metal oxide nanoparticles

Fe_2O_3, UO_2 , **TiO_2***, **MnO_2***, **ZrO_2***, Te/SeO_2.

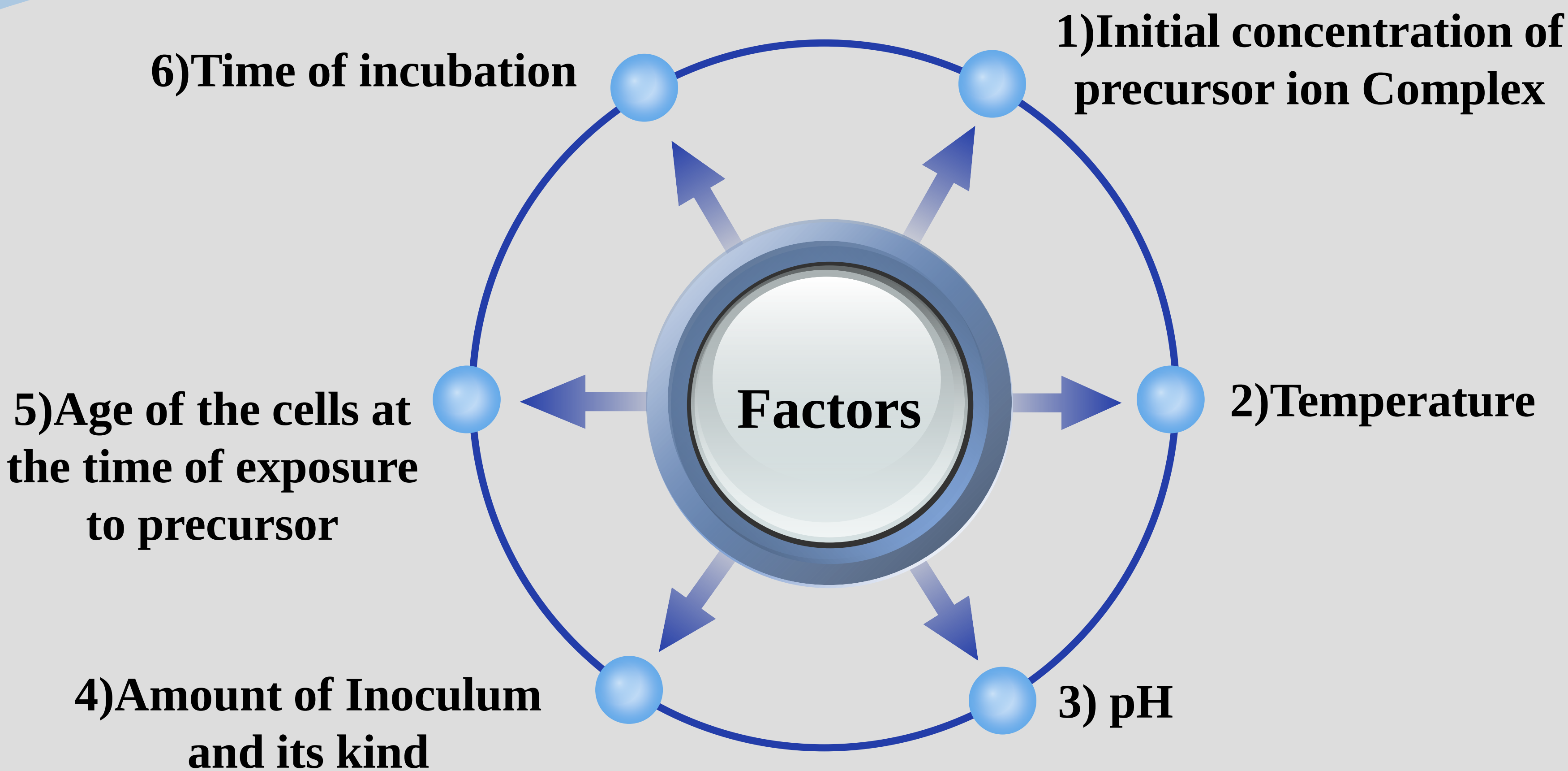
Application: as a novel raw materials for electrode manufacturing and selective catalytic oxidation catalysts.

2006

تولید نانوذرات با استفاده از میکروارگانیسم‌های مختلف

میکروارگانیسم‌ها	نانو ذرات تولید شده
باکتری‌ها	
Bacillus subtilis	طلا
Shewanella algae	طلا
Pseudomonas stutzeri	نقره
Lactobacillus	طلا، نقره و آلیاژ دوتایی آنها
Clostridium thermoaceticum	سولفید کادمیوم
Klebsiella aerogenes	سولفید کادمیوم
Escherichia coli	سولفید کادمیوم
Desulfobacteriaceae	سولفید روی
Thermoanaerobacter ethanolicus	مگنتیت
Magnetospirillium magnetotacticum	مگنتیت
Thermomonospora sp	طلا
Rhodococcus	طلا
Chlorella vulgaris	طلا
Phaeodactylum tricornutum	سولفید کادمیوم
مخمرها	
Candida glabrata	سولفید کادمیوم
Torulopsis sp	سولفید سرب
Schizosaccharomyces pombe	سولفید کادمیوم
MKY3	نقره
قارچ‌ها	
Verticillium	طلا و نقره
Fusarium oxysporum	طلا، نقره و آلیاژ دوتایی آنها، سولفید کادمیوم و زیرکونیوم
Colletotrichum sp	طلا

Factors Affecting on nanoparticle formation and its stability



Chemical synthesis of silver nanoparticles

1) By using reducing chemicals: Strong ($NaBH_4$) , Weak (citrate ions and ascorbic acid) and strong + weak. Ion complex: $AgNO_3$., monodispersity: 5-25%

2) Photolytic synthesis : by UV {formation biocomposite TiO_2+Silver : increasing photocatalytic properties TiO_2. monodispersity: 5-10%., ion complex: $AgNO_3$.

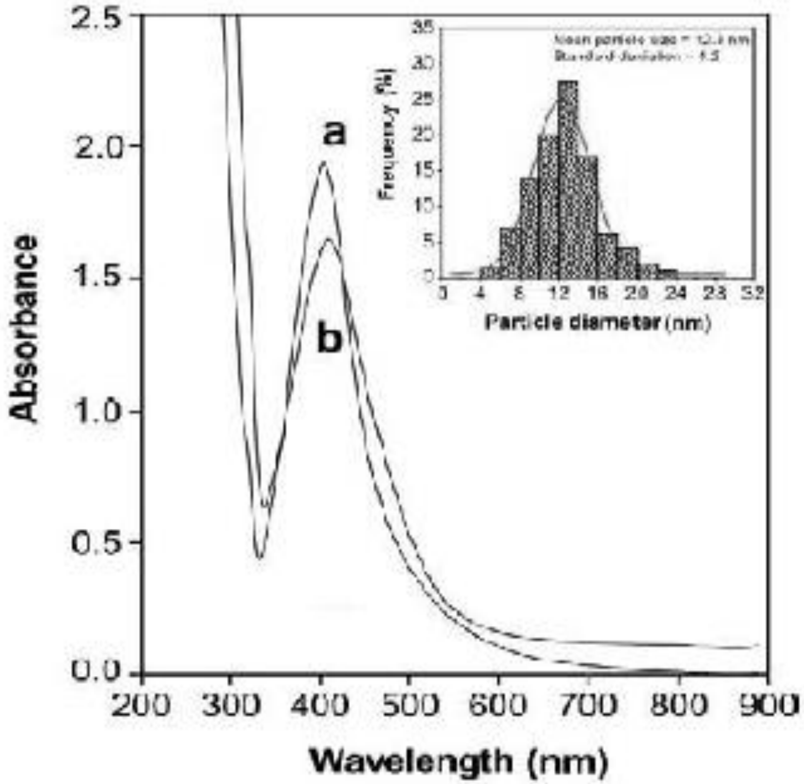
3) Reduction by H_2., ion complex: Ag_2O.

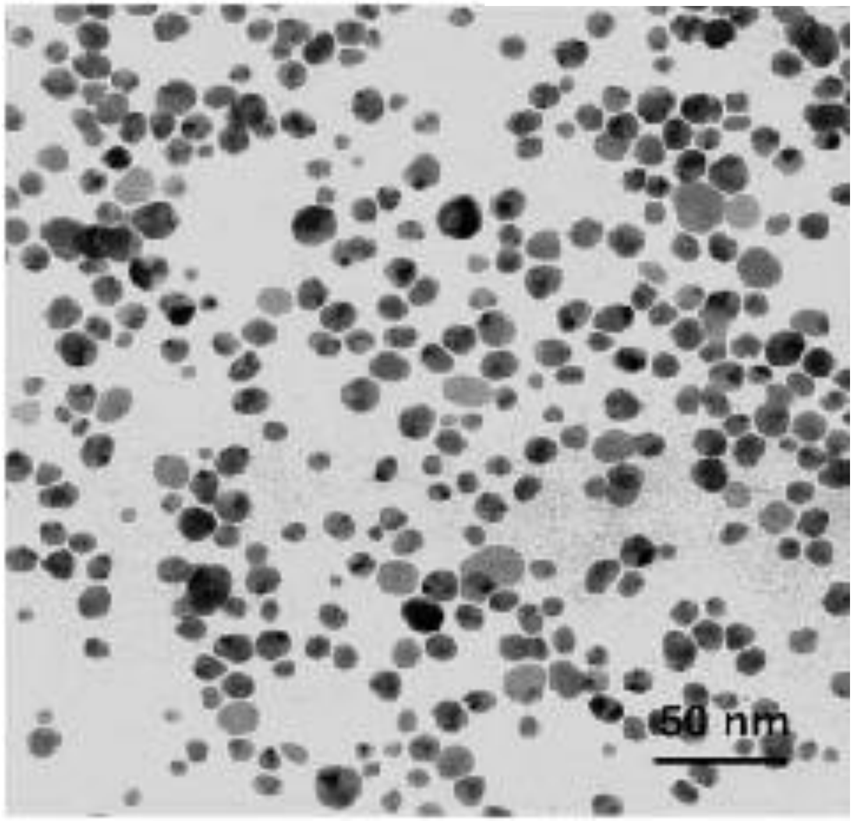
4) **Tollens process : reduction by sugars . Ion complex: $Ag(NH_3)_2^+$. Monodispersity: 20-40%

Chemical synthesis

سنتز نقره به روش تولن

تصویر TEM نانوذرات نقره

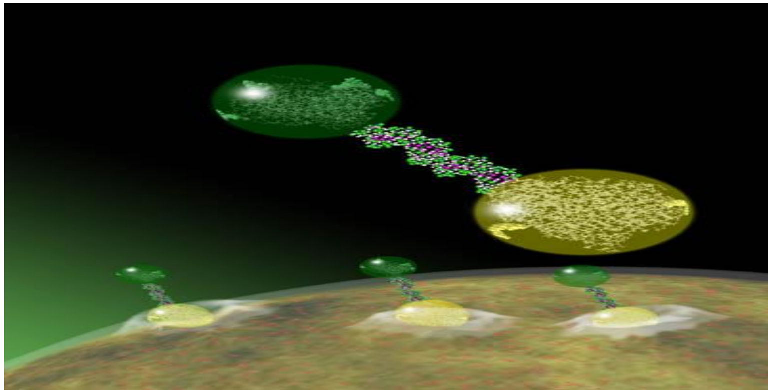


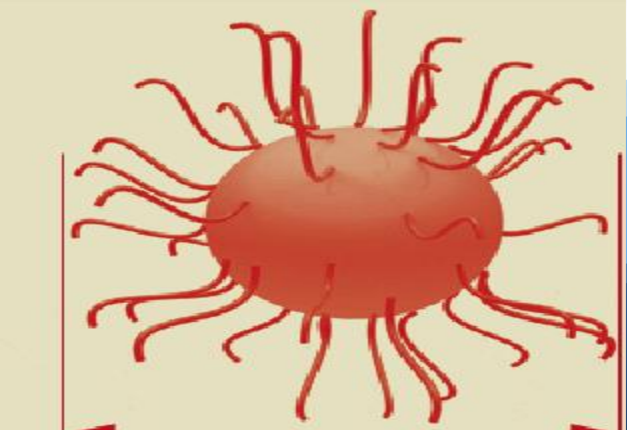


طیف هیدروسل نقره بلافاصله پس از پخش در آب (a) و پس از فریز و دوباره پخش شدن در آب (b).

Biological synthesis of silver nanoparticles

- Biological synthesis : 1) plants (alfa alfa)
- 2) microorganisms (bacteria/yeasts/fungi/actinomycetes)
- **Bcteria (2000-2008)**
- ***1) Pseudomonas stutzeri AG259****. (Tanja et al., 2001):isolated from a silver mine.*
- ***2) Aeromonas .sp. SH10*** *(Fu Mouxing et al ., 2006): isolated shanghang silver mine.*
- ***3) Enterobacteriaceae*** *(Klebsilla punemoniae, E.coli and Enterobacter cloacae) {R.Shahverdi et al 2007}: preparation from microbial collection.*





Bacteria	Inoculum	Ion complex	Reaction condition	Location	Time of Synthetic process	Dispersity/ Stability
P.stutzeri	Wet biomass 1g	AgNO3 (50 mM)	T=27 Time of incubation:72h	Periplasmic space	Slow 24 h	35-45nm +
Aeromonas. sp SH10	Dried biomass g?	10g/l Ag(NH3)2 +NaOH	T=60 Time of incubation:24h	Surface/ solution	Fast 4 h	*6.4nm +
*Klebsilla. pneumonia	Supernatant 1%v/v	AgNO3 1 mM	T=34 Time of incubation:?	extracellular	Quite fast 50 min	28-122nm ?

Biosynthesis of silver nanopraticles by fungi

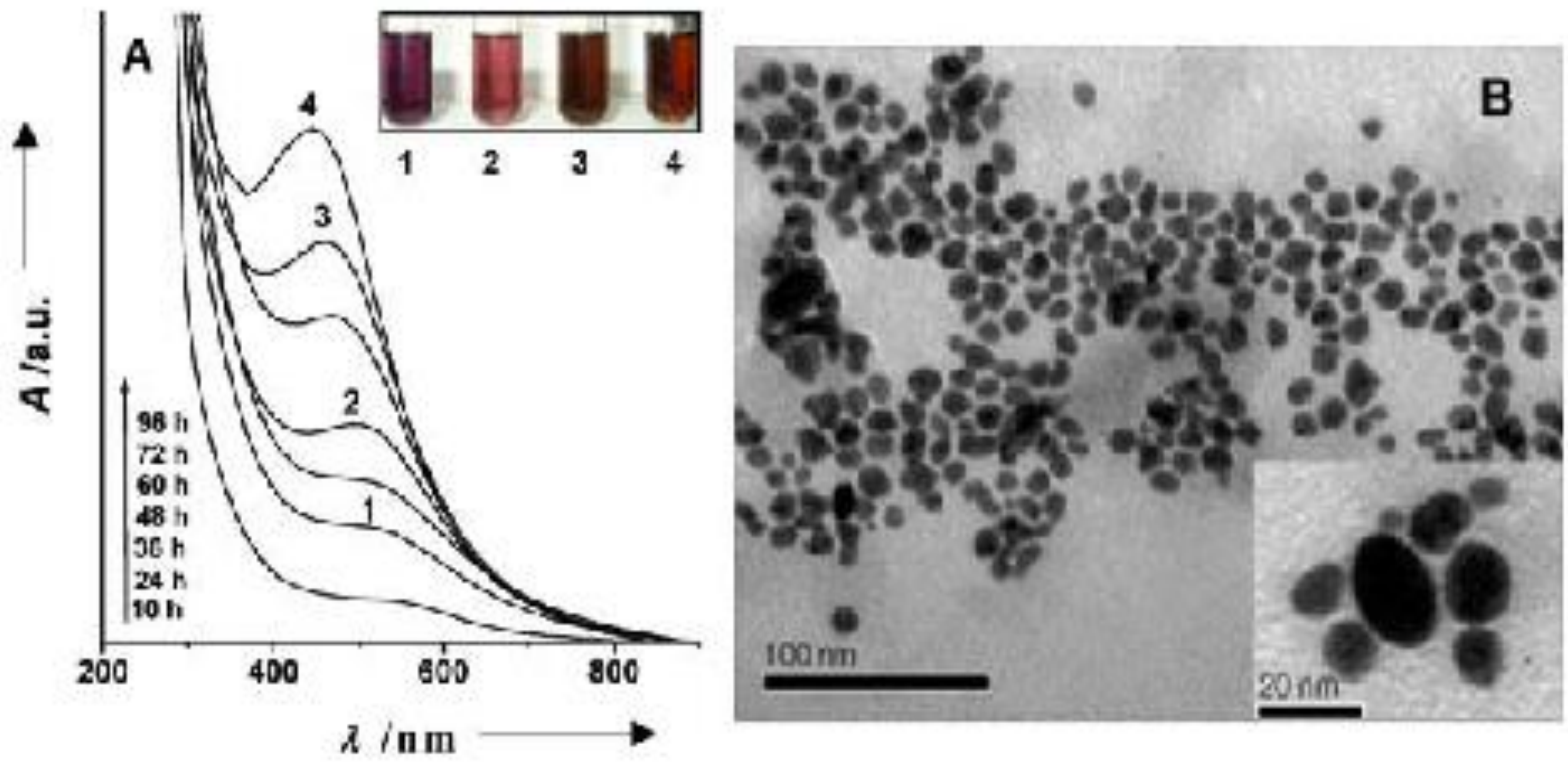
- **Yeast:** *Isolate MKY3*
- **Filamentous Fungi:**
- 1) *Verticillium SP. (Mukherjee et al ., 2001)*
- *2) Fusarium oxysporum (Nelson et al., 2005)*
- *3) Aspergillus fumiatus strain NCIM 902 (Kuer et al., 2006)*
- *4) Aspergillus flavus strain NCIM 650 (N.Vigneshwaran et al., 2007)*

- Shift from bacteria to fungi as nanofactories
- **Advantages:**
- 1) extracellular reduction
- 2) simple handling of the biomass
- 3) simple downstream processing
- 4) efficient secretors of soluble proteins (30g/l extracellular Protein) and the use of specific secreted enzymes in the synthesis of nanoparticles.

Fungi & silver nanoparticles

1 mM $AgNO_3$	inoculum	location	Dispersity/ stability	Time of synthetic process Mechanism
Verticillium SP.	biomass	Surface/ intracellular	25-30 +	72 h ?
F.oxysporum	Biomass/ *supernatant	extracellular	20-50 +	2 h Nitrate-dependent reductase and shuttle quinone
A.flavus	biomass	Surface/ intracellular	4-14 +	48 h ?
A.fumigatus	Biomass/ *supernatant	extracellular	5-25 +	10 min ?

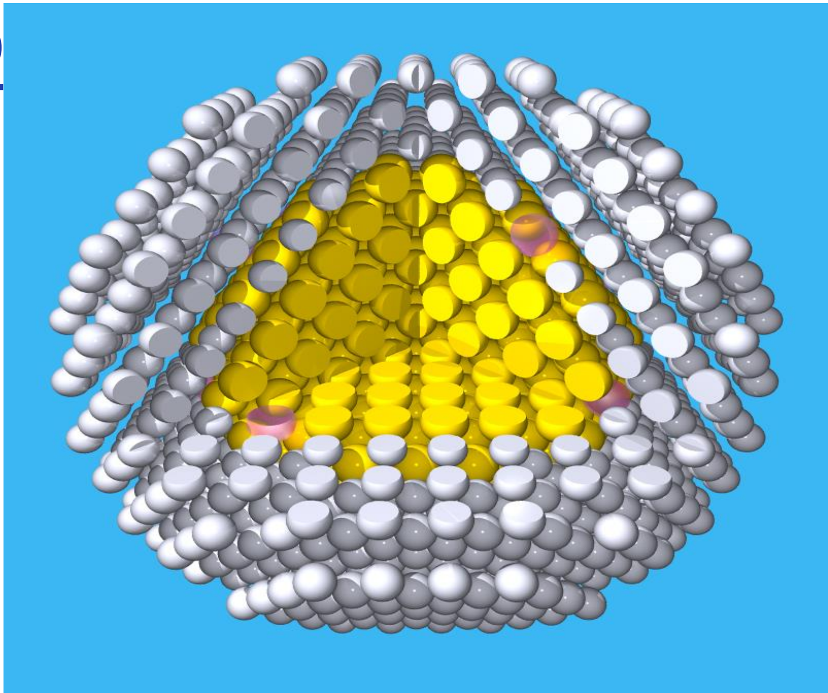
a UV–Vis spectra of and b TEM analysis Au–Ag alloy nanoparticles, after the reaction of a mixture of a solution containing 1 mm HAuCl4 and 1 mm AgNO3 with 60 g F. oxysporum wet biomass for 96 h.



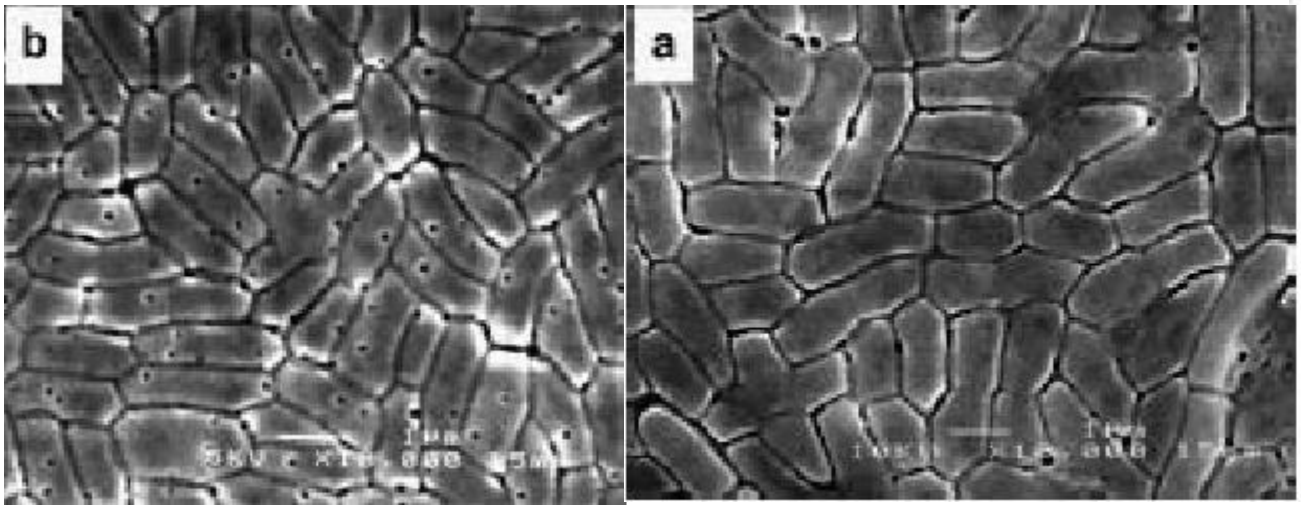
Silver and Gold nanoparticles application

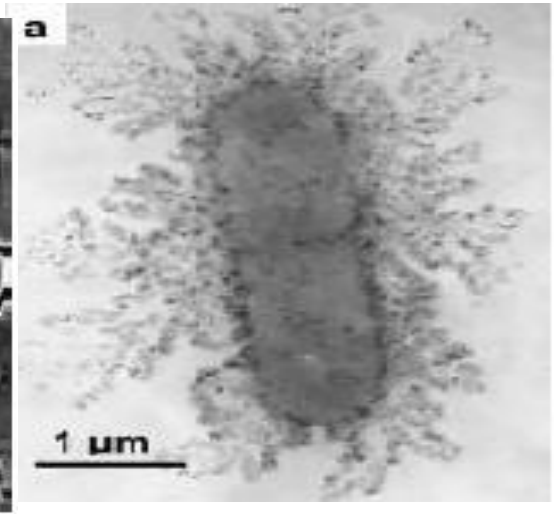
silver

- 1) selective coating for solar energy absorption
- 2) intercalation material for electrical batteries
- and optical receptors
- 3) catalysts in chemical reaction
- **4) antimicrobial agent (1g/100m^{2})**
- Gold
- 1)rapid detection of cancers
- 2) catalysts in chemical reactions



- مکانیزم عملکرد نقره در برابر باکتری‌ها





استفاده از خاصیت ضد میکروبی نقره

صنايع غذايي
صنايع نساجي
صنايع كاغذ
استفاده در صنعت لوازم منزل
استفاده در مواد شوينده وبهداشتي

Synthesis of quantum dots and its application

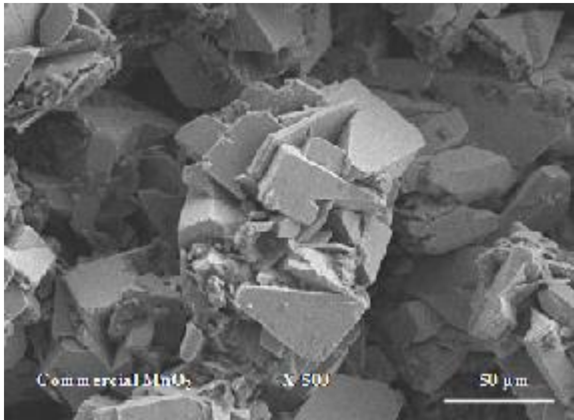
- Semiconductors: ZnS/PbS/CdS/FeS
- **Bacteria**
- 1) *Clostridium thermoaceticum*：CdS (cell surface/medium)., 50-100 nm., $CdCl_2$+ cysteine hydrochloride.
- 2) *Klebsilla* : CdS (cell surface) , 20-100 nm, $CdCl_2$ +buffer components
- 3) *E.coli* : CdS (intacellular) , 20-40 nm., $CdCl_2$+ Na_2S .
- 4) *SRB (desulfobacteriacea):* ZnS , FeS ., 2-5 nm., Zn/$FeSO_4$.
- **Yeasts (intracellular)**
- **1) *candida glabrata* (CdS , 2-5 nm)** **2) Torulopsis SP. (PbS, 2-5 nm)**
- **3) *Schizosaccharomyces pombe* (CdS, 1-1.5 nm).**
- **Fungi: *Fusarium oxysporum* ., CdS ($CdSO_4$), cell surface/medium .**
-

Application

- **Properties of quantum dots**: 1) ability to tune the optical absorption/emission properties 2) high flexibility 3) high adsorption

- **Application:**

1) For making of quantum lasers
2) For biological detection
3) For cell imaging =تصویر برداری زیستی دقیق
4) Because of high adsorption , as a suitable matrix for the long-term safe of a number of vital ions to the nuclear industry . (400-500 m^2/g).

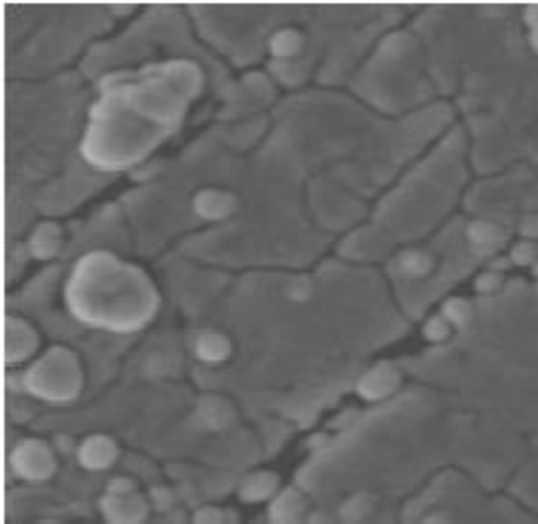


Formation of Te and Se nanoparticles

- Tellurium is a toxic metalloid present as a trace element (0.002 ppm) in the earth crust.
- Te oxidation states: 6^{+},4^{+},0, 2^{-}.

- Bacterial resistance mechanisms: 1) bioreduction 2) volatilization

- Chemical synthesis of the Te nanoparticles:
- 1) laser ablation (T=500^{o} C)
- 2) sonication
- 3) auto-oxidation of highly reactive chemicals
- (e.g. NaHTe)



Biological synthesis of Te /Se nanoparticles

1)Bacillus selenitrireducens

2) Sulfurospirillum barnesii

(Dissimilatory reduction in presence lactate)

Dispersity: *in (1)* ***10-100*** *and in (2)* ***10-50 nm****)*

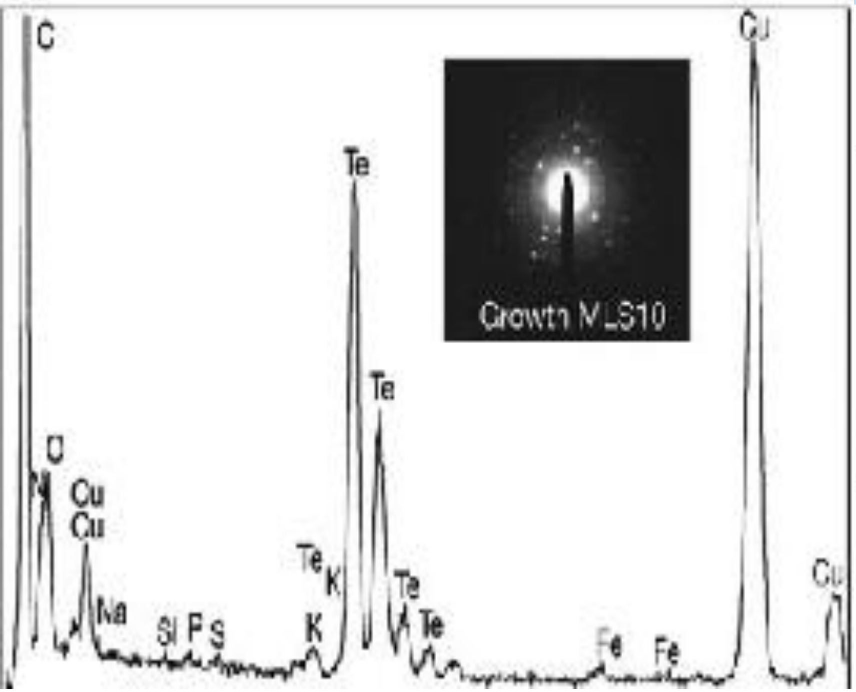
Location: medium/surface/cytoplasm !

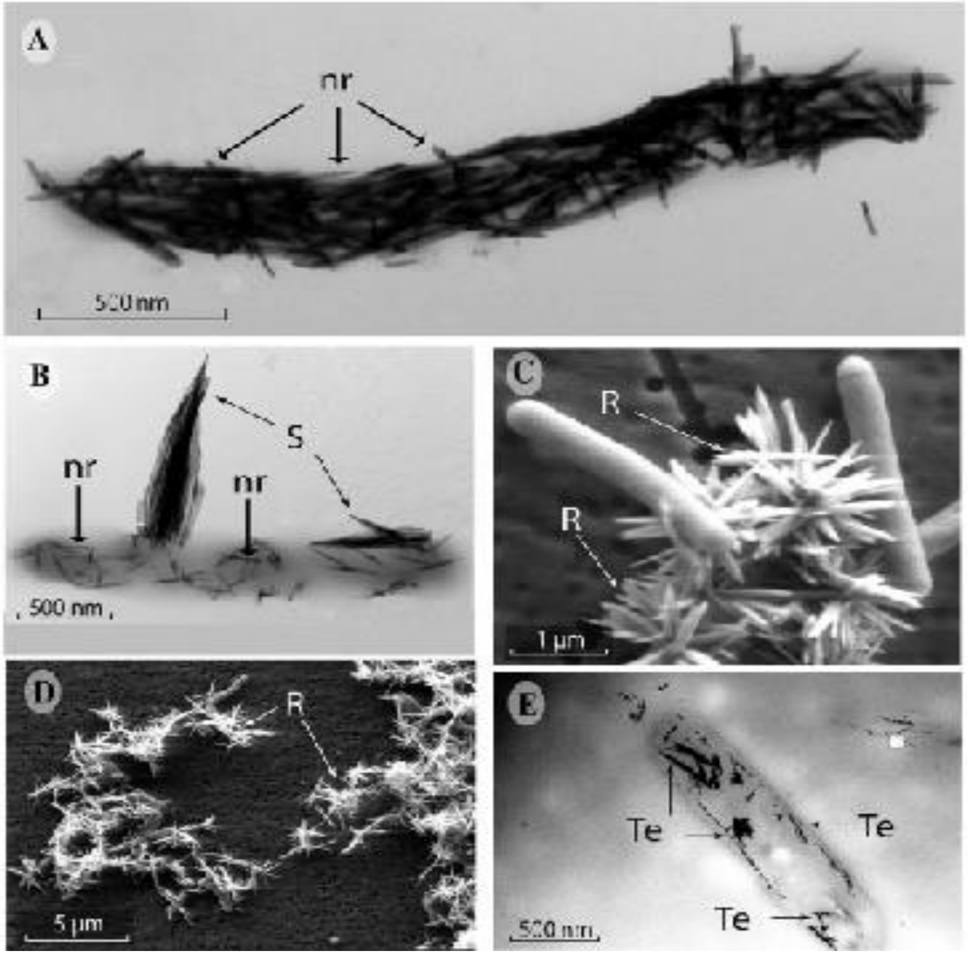
Application: solar cell (CdTe)

$$\text{Lactate}^{-} + \text{TeO}_3^{2-} + \text{H}^{+} \rightarrow \text{Acetate}^{-1} + \text{Te(0)} + \text{HCO}_3^{-} + \text{H}_2\text{O}$$

$$\Delta G_r^{\circ} = -227 \text{ kJ mole lactate}^{-1} \text{ or } -61 \text{ kJ mole-eq electrons}^{-1}$$

Formation of irregular /Se/Te) "nanorods" by salinicoccus iranensis. TEM and SEM/EDX analysis



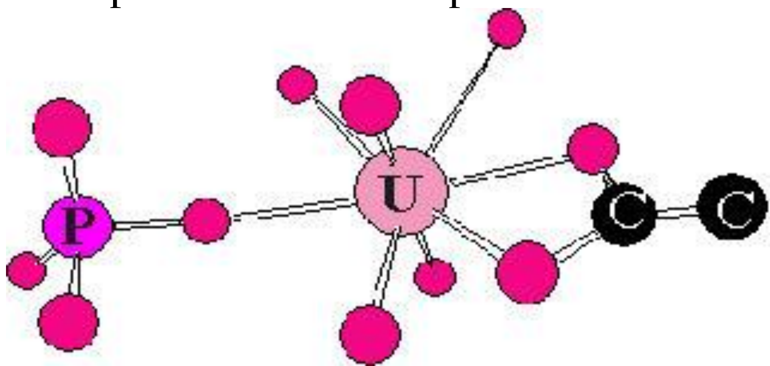


Formation of uraninite (UO2) nanoparticle

- ►Uranium in the environment occurs primarily as 3 of its 17 known isotopes, 238 U (99.27%), 235 U (0.72%), and 234 U (0.005%). All are radioactive; however, it is the chemical toxicity that is of greatest ecological risk .

►Unfortunately, the anthropogenic use of uranium for nuclear research, fuel production, and weapons manufacturing has resulted in widespread environmental contamination. Additional contamination has resulted from trace amounts of uranium being released from the combustion of coal .

►Seeking to achieve this goal of remediation quickly, with reasonable expense and a minimum of environmental disruption, the DOE has been the driving force for the examination of microbial processes that can be exploited for cleanup.



- ►In oxic surface waters, UO_2^{2+} is present and form stable, soluble complexes with **carbonate**, phosphate, and humic substances . The formation of these complexes is governed by pH, Eh (reduction potential), temperature, and ligand concentration.

Mirobial Uranium Reduction

- **First report : 1962 *(Micrococcus lactilyticus (veillonella alcalescens)***
- ***Procees abiotic reduction: sulfide , Fe(II) or hydrogen***
- ***Procees biotic reduction: DMRB (in particular the Geobacter and Shewanella gebera***
- ***Because of respiratory versatility.***

Bacteria shown to reduce U(VI) to U(IV)

Bacterium	Reference(s)	Bacterium	Reference(s)
Anaeromyxobacter dehalogenans strain 2CP-C	78	*Desulfovibrio sulfodismutans* DSM 3696	51
Cellulomonas flaigena ATCC 482[a]	82	*Desulfovibrio vulgaris* Hildenborough ATCC 29579	51
Cellulomonas sp. WS01	82	*Geobacter metallireducens* GS-15	50
Cellulomonas sp. WS18	82	*Geobacter sulfurreducens*	38
Cellulomonas sp. ES5	82	*Pseudomonas putida*	4
Clostridium sp.	22	*Pseudomonas* sp.	4
Clostridium sphenoides ATCC 19403	21	*Pseudomonas* sp. CRB5	57
Deinococcus radiodurans R1	24	*Pyrobaculum islandicum*	39
Desulfomicrobium norvegicum (formerly *Desulfovibrio baculatus*) DSM 1741	51	*Salmonella subterranea* sp. nov. strain FRC1	87
Desulfotomaculum reducens	92	*Shewanella alga* BrY	14, 93
Desulfosporosinus orientis DSM 765	91	*Shewanella oneidensis* MR-1 (formerly *Alteromonas putrefaciens*, then *Shewanella putrefaciens* MR-1)	50
Desulfosporosinus spp. P3	91	*Shewanella putrefaciens* strain 200	11
Desulfovibrio baarsii DSM 2075	51	*Veillonella alcalescens* (formerly *Micrococcus lactilyticus*)	99
Desulfovibrio desulfuricans ATCC 29577	49	*Thermoanaerobacter* sp.	77
Desulfovibrio desulfuricans strain G20 (to be renamed *Desulfovibrio alaskensis*)	68	*Thermus scotoductus*	43
Desulfovibrio sp. UFZ B 490	72, 73	*Thermoterrabacterium ferrireducens*	42

C-type cytochrome-dependent formation uraninite nanoparticle

Resting cells

0.2mM Uranyl acetate

10mM Lactate

24 h

Shewanella oneidensis

1-5 nm

UO_2-EPS

Extracellular /periplasmic

High monodisperisity
High stability

Laser photolysis

$^{235}UF_6 \rightarrow {}^{235}UF_5 + F.$???

Formation of PGM nanoparticles

►PGM metals (Pt/Pd/Ra) are used worldwide on an increasing scale in catalysis and electronics, due to conductivity and catalytic activity.

So, very interesting: 1) recovery of these precious metals 2) clean-up of PGM contaminated waste waters and 3) production of PGM nanoclusters .

Applications of PGM nanoparticles

As environmental catalysts in fuel cells	As metal catalysts in Selective hydrogenation and hydroformylation reactions	Ability to catalyse various Chemical reductions such as dehalgenations Of polychlorinated biphenyls Or reduction of Cr(VI)

H_2PtCl_6: pale-yellow
►Pt nanoparticle: gray-*black

S.algae

Resting cells of *S.algae* were able to reduce (90%) 1 mM H_2PtCl_6 into elemental Pt nanoparticles at room temperature and natural pH within 50 min, when sodium lactate was provided as the electron donor. Dispersity: 5-10 nm., Location: periplasmic space .

S.oneidensis

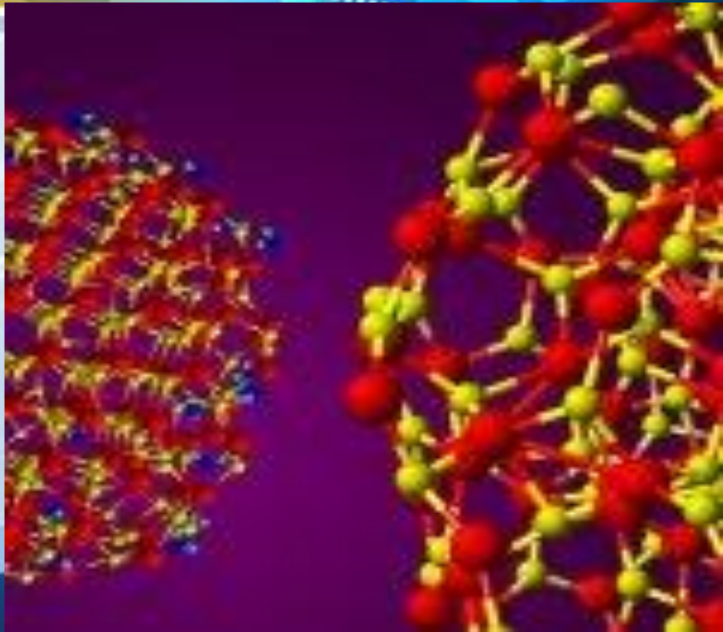
Resting cells of *S.oneidensis* were able to reduce 5 mM H_2PdCl_4 into elemental Pd nanoparticles at room temperature and natural pH within 1 h, when sodium lactate was provided as the electron donor. Dispersity: 10-15 nm., Location: *cytoplasm/periplasmic space .

B.spaericus

S-layer proteins of B.sphaericus arehighly interesting for clean-up,recovery and production of nanoparticles.

Solution precursor:Na_2PdCl_4 ., location: s-layer ., dispersity: 0.8-1 nm(19-43 atoms) ., mechanism of reduction: 1)biosorption by phosphate and carboxyl groups on the proteins of S-layer (confirmed by EXAFS method) 2) reduction by the addition of H_2 as an electron donor.

Conclusion



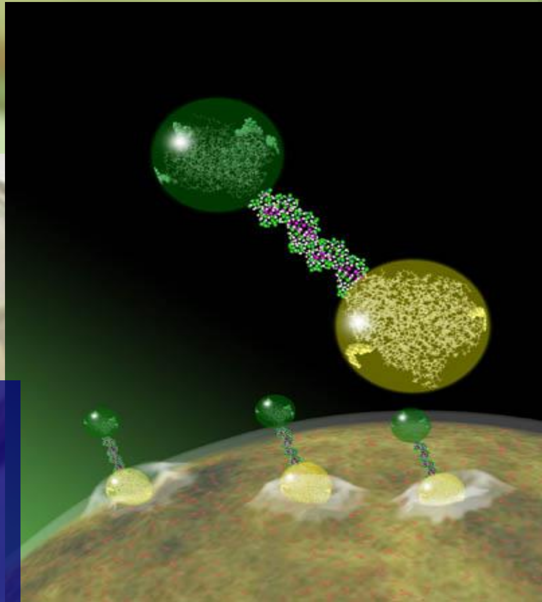
Microbial nanoparticle production

- **Microorganisms** (bacteria, yeasts, actinomycetes and fungi) as **nanofactories.**

- For **compete with existing physical and chemical synthesis** protocol , points of view require to be addressed:

- **1) Microbiology** :the elucidation of **biochemical/molecular pathways** leading to metal ion reduction that lead to the possibility of genetically engineering microbes to overexpress specific reducing molecules and capping agents, for controlling size and sharp.

- **2) Nanotechnology** : high reactivity and high stability.



The rational use of constrained environments within cells such as the periplasmic space and cytoplasmic vesicular compartments (e.g., magnetosome) to modulate nanoparticle size and shape is an exciting possibility.

Process Biochemistry xxx (xxxx) xxx-xxx

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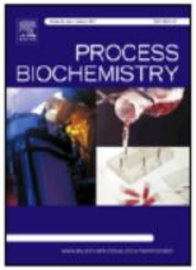

Development and evaluation of different strategies for the clean synthesis of silver nanoparticles using *Yarrowia lipolytica* and their antibacterial activity

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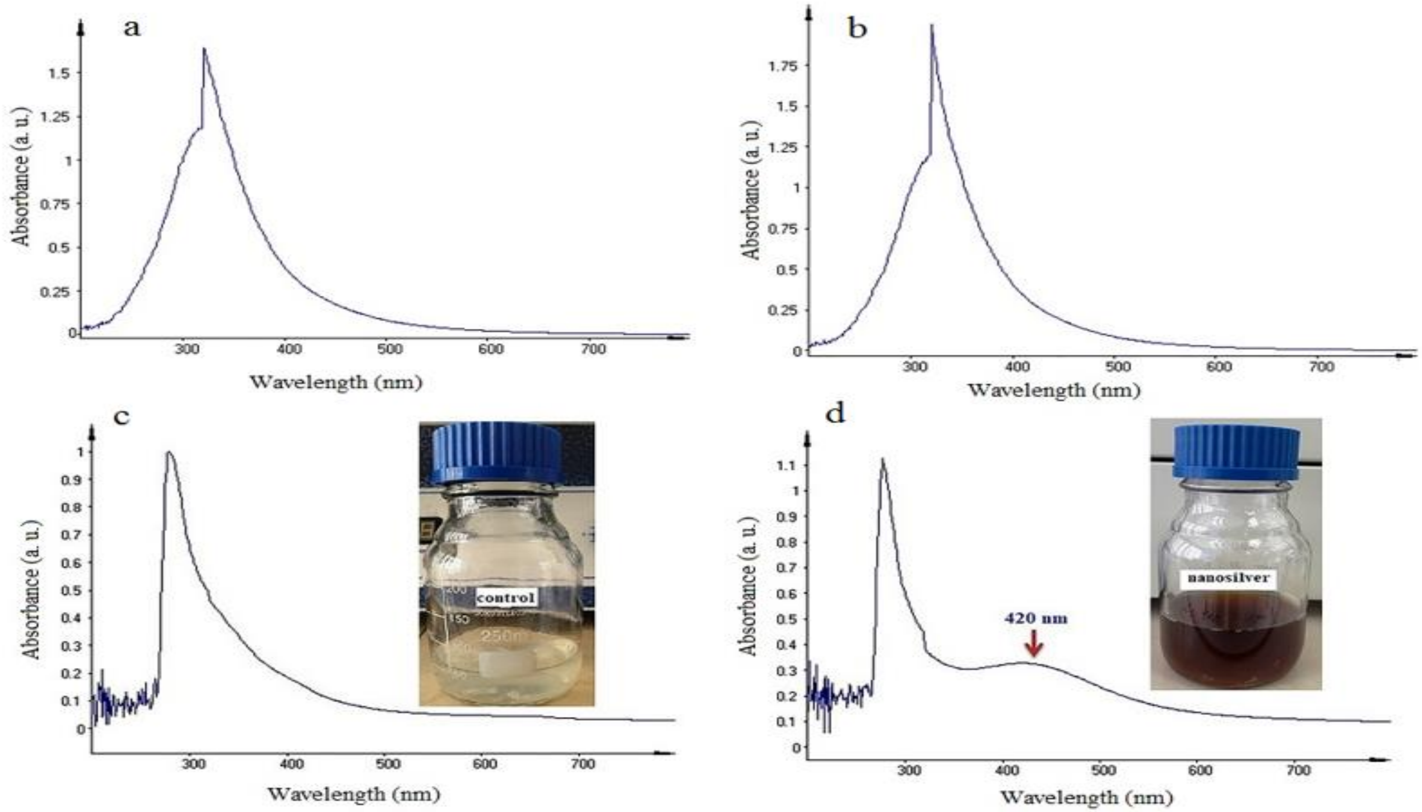
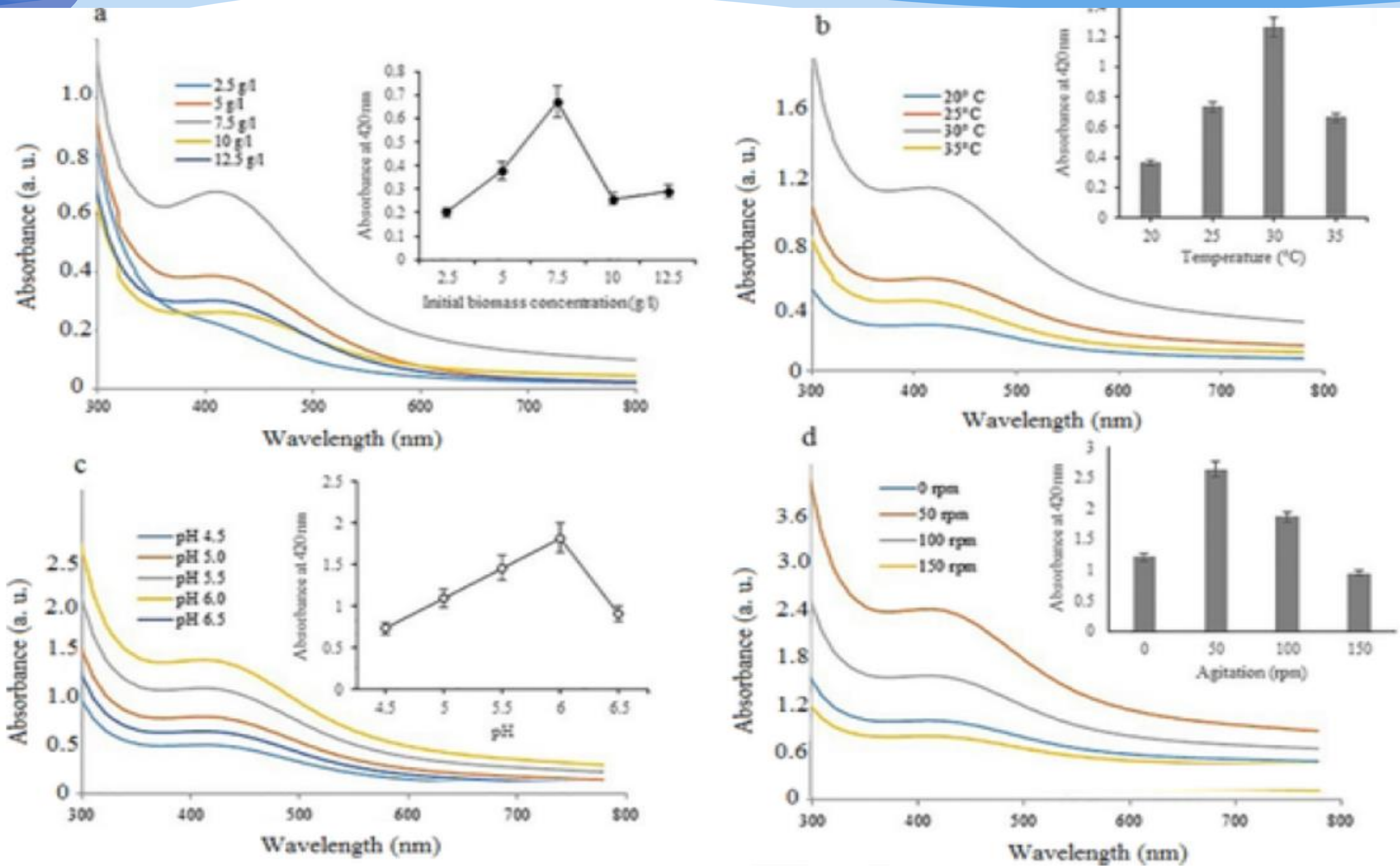
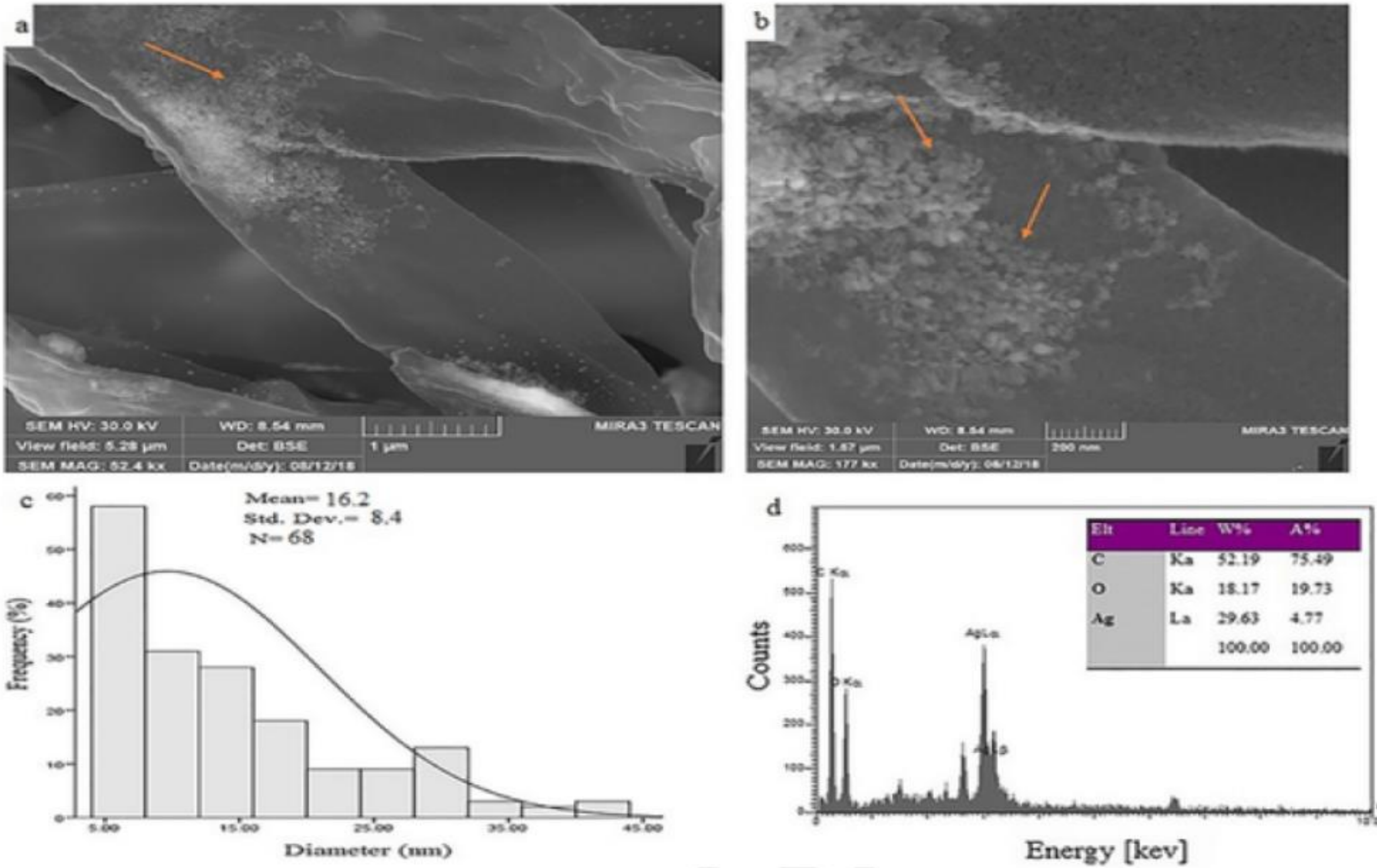


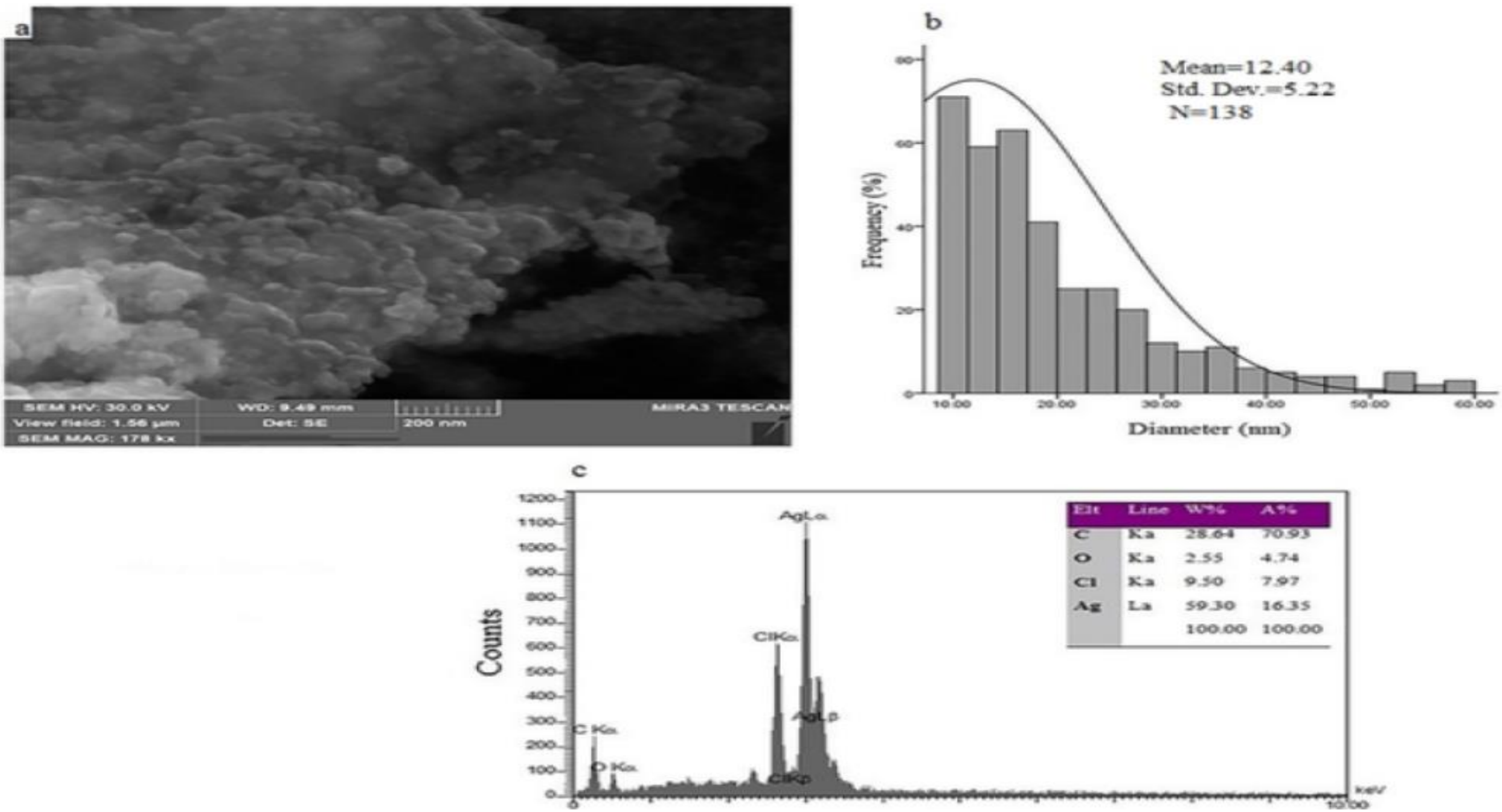
Fig. S2. Absorbance vs wavelength graph of UV-Vis spectroscopy confirms biosynthesis of AgNPs at 420 nm by *Y. lipolytica* DSM 3286. (a) YPD medium treated with silver nitrate at the beginning of reaction and (b) after 48 h of reaction; (c) YPD inoculated with yeast and silver nitrate at the beginning of reaction and (d) after 48 h of reaction.


5 Mm
7.5 Mm
2.5 Mm
10 Mm
1Mm
12.5 Mm
Conrtol
15 Mm

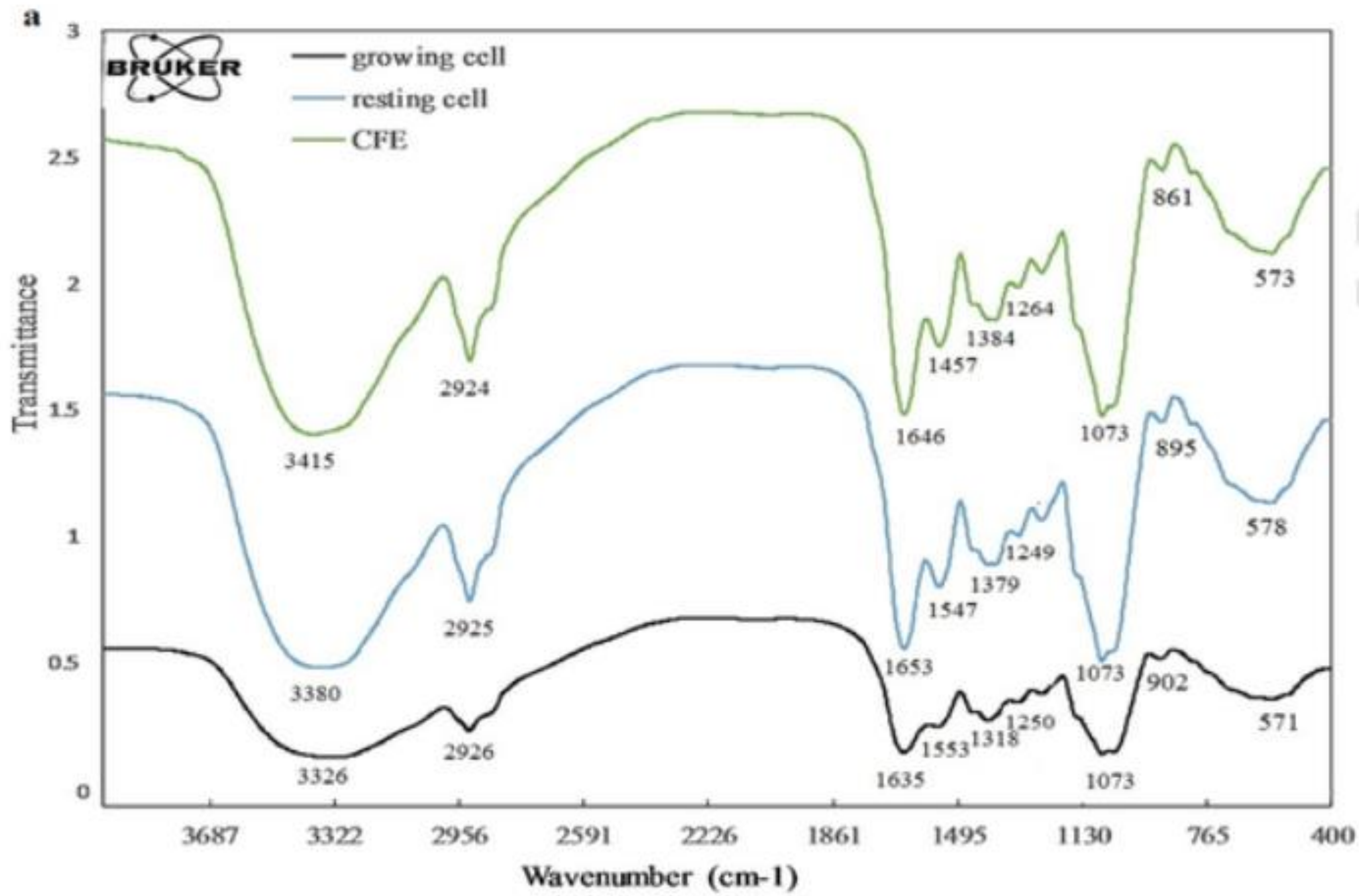


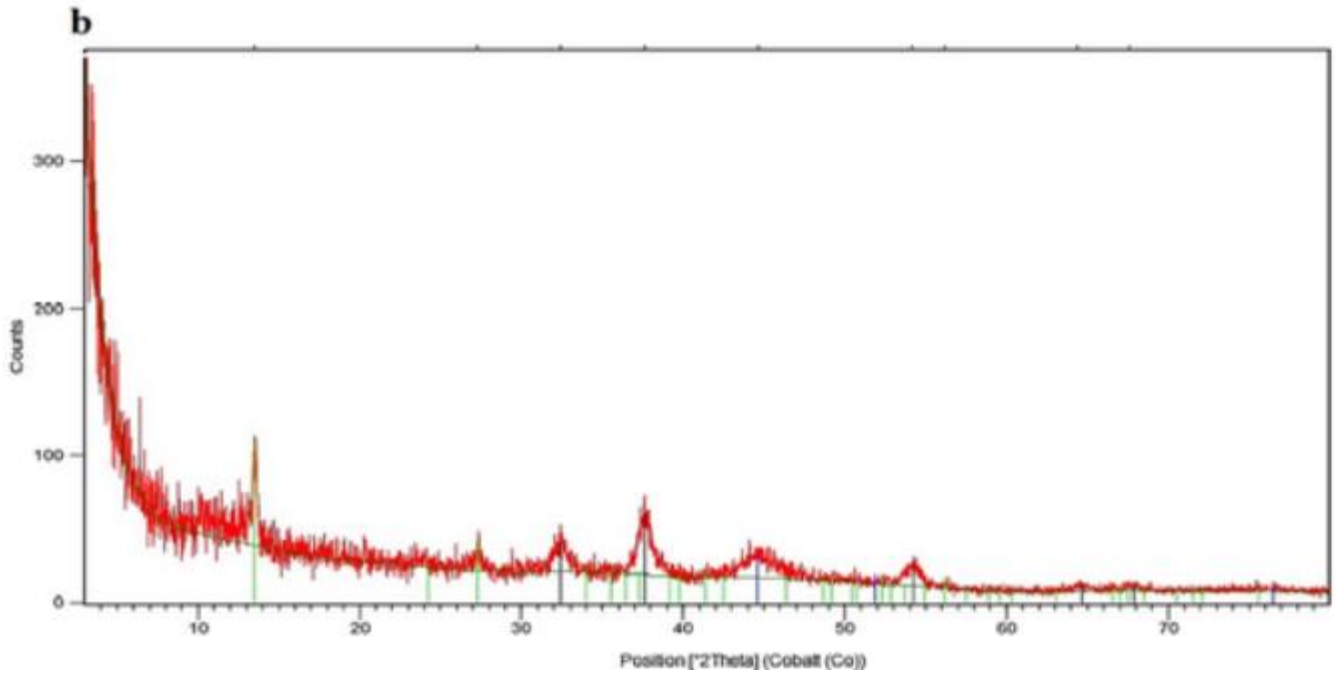
a
Absorbance (a. u.)
Wavelength (nm)
2.5 g/l
5 g/l
7.5 g/l
10 g/l
12.5 g/l
Absorbance at 420 nm
Initial biomass concentration(g/l)
b
Absorbance (a. u.)
Wavelength (nm)
20° C
25° C
30° C
35°C
Absorbance at 420 nm
Temperature (°C)
c
Absorbance (a. u.)
Wavelength (nm)
pH 4.5
pH 5.0
pH 5.5
pH 6.0
pH 6.5
Absorbance at 420 nm
pH
d
Absorbance (a. u.)
Wavelength (nm)
0 rpm
50 rpm
100 rpm
150 rpm
Absorbance at 420 nm
Agitation (rpm)

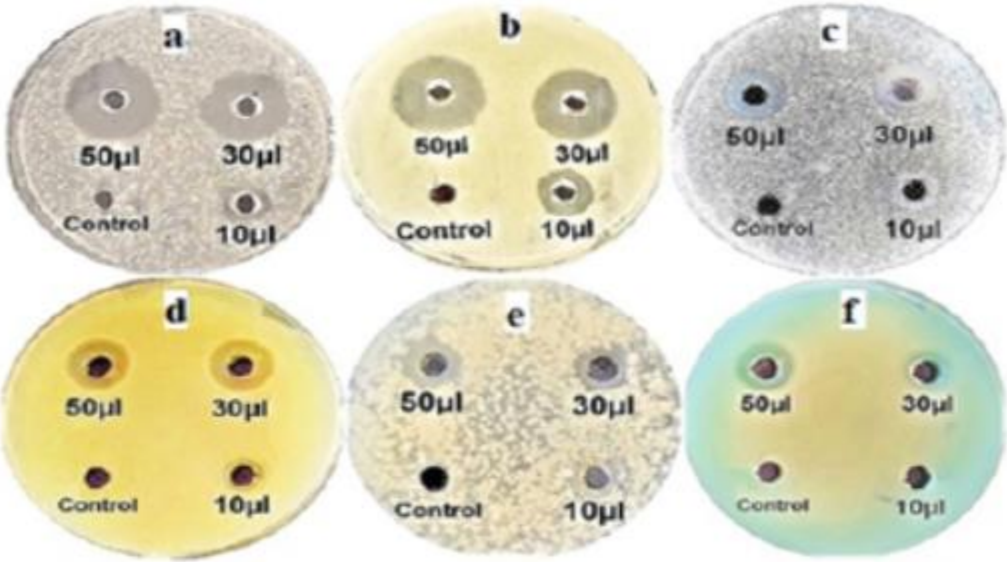

a
SEM HV: 30.0 kV
WD: 8.54 mm
MIRA3 TESCAN
Det: BSE
1 μm
b
200 nm
c
Mean= 16.2
Std. Dev.= 8.4
N= 68
Frequency (%)
Diameter (nm)
5.00
15.00
25.00
35.00
45.00
d
Counts
Energy [kev]
C Ka
O Ka
AgLa
Elt Line W% A%
C Ka 52.19 75.49
O Ka 18.17 19.73
Ag La 29.63 4.77
100.00 100.00



a
SEM HV: 30.0 kV
WD: 9.49 mm
MIRA3 TESCAN
View field: 1.56 µm
Det: SE
200 nm
SEM MAG: 178 kx
b
Mean=12.40
Std. Dev.=5.22
N=138
Frequency (%)
Diameter (nm)
c
Counts
AgLα
ClKα
AgLβ
C Kα
O Kα
ClKβ
keV
Elt Line W% A%
C Ka 28.64 70.93
O Ka 2.55 4.74
Cl Ka 9.50 7.97
Ag La 59.30 16.35
100.00 100.00

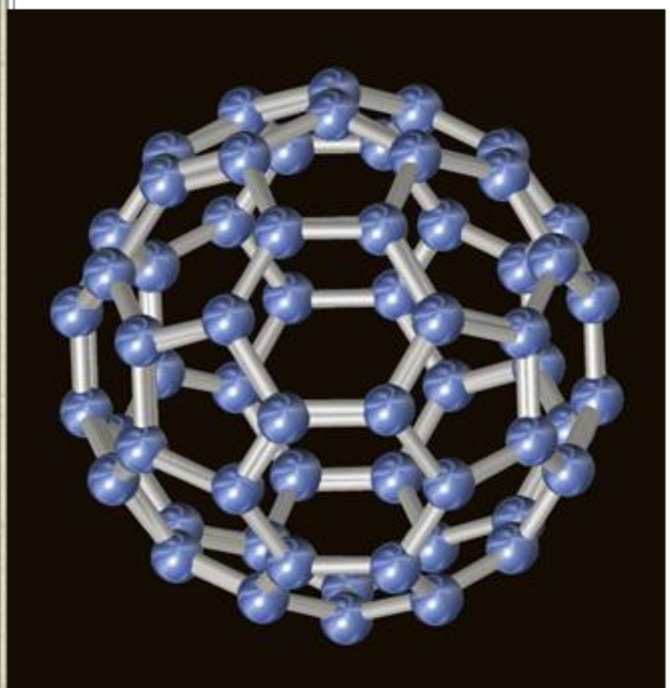

a
BRUKER
growing cell
resting cell
CFE
Transmittance
Wavenumber (cm-1)
3
2.5
2
1.5
1
0.5
0
3687
3322
2956
2591
2226
1861
1495
1130
765
400
3415
2924
1646
1457
1384
1264
1073
861
573
3380
2925
1653
1547
1379
1249
1073
895
578
3326
2926
1635
1553
1318
1250
1073
902
571


b
Counts
300
200
100
0
10
20
30
40
50
60
70
Position [°2Theta] (Cobalt (Co))



	Bacterial strains	Zone of inhibition (mm in diameter)		
		10µl	30µl	50µl
a	*Staphylococcus aureus* ATCC33591	8	20	23
b	*Escherichia coli* ATCC25922	10	20	21
c	*Enterococcus faecalis* ATCC29212	7	13	15
d	*Proteus vulgaris* ATCC49132	6	12	14
e	*Streptococcus pyogenes* ATCC19615	5	9	11
f	*Pseudomonas aeruginosa* ATCC27853	0	7	9

Thanks For Your Attention!



براي انسانهاي بزرگ بن بستي وجود ندارد

چون براين باورند

يا راهي خواهم يافت

يا راهي خواهم ساخت