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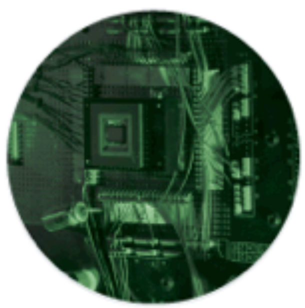
Chaos is cheap: adventures in nanomaterials synthesis

Sheryl Ehrman

Chemical and Biomolecular Engineering

University of Maryland, College Park

US Food and Drug Administration, April 16, 2015

Outline

- Chemical and Biomolecular Engineering at the University of Maryland
- Overview of aerosol synthesis
- Flame and solution CuO based photoactive nanoparticle materials
- Concerns about environmental health and safety of nanoparticle based materials
- Summary and outlook

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ChBE at the University of Maryland

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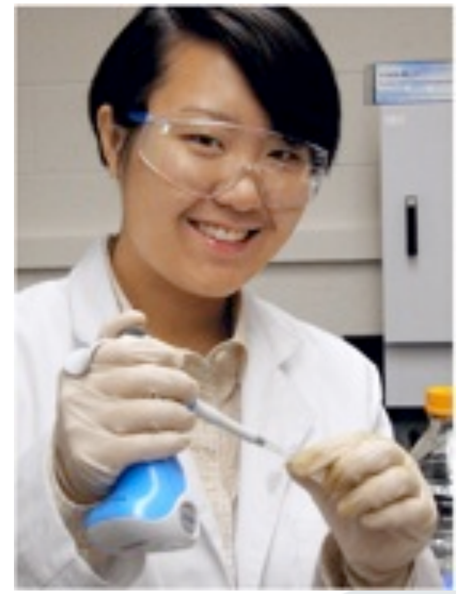
ChBE by the numbers

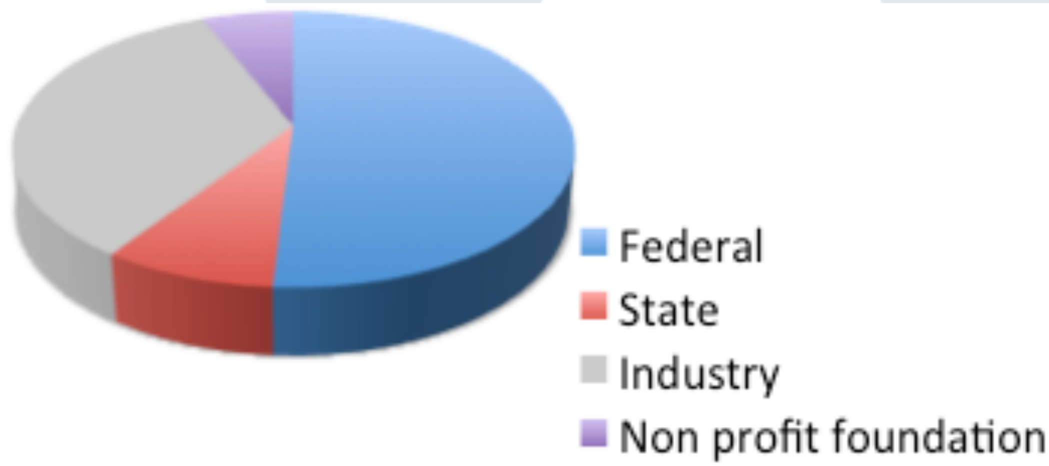


15 faculty, 2 assistant, 5 associate, 8 full professors

~ 340 undergraduate students with emphasis on undergraduate research experience

44 PhD, 18 MS students

~ 3 M external research expenditures FY 2013



CHBE Strengths: scholarly, entrepreneurial, diverse

ChBE research interests

- Transport phenomena, multiphase flow and process intensification
- Processing of advanced materials for catalysis, energy conversion and energy storage applications
- Biomolecular engineering
- Systems engineering

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Current projects in the Ehrman laboratory

- Aerosol synthesis of
  - Lithium based oxide nanoparticles for battery cathode applications, collaboration with CS Wang, UMD
  - Metal microparticles for printed electronics applications, collaboration with HD Glicksman, DuPont
- Emissions and chemical transport modeling to support regional air quality improvement, collaboration with R. Dickerson, T. Canty, J. Stehr, UMD
- Characterization of exhaled breath aerosols and respiratory protection, with applications to flu transmission, collaboration with D. Milton, UMD, Matthew Myers, Prasana Hariharan, Suvajyoti Guha, FDA

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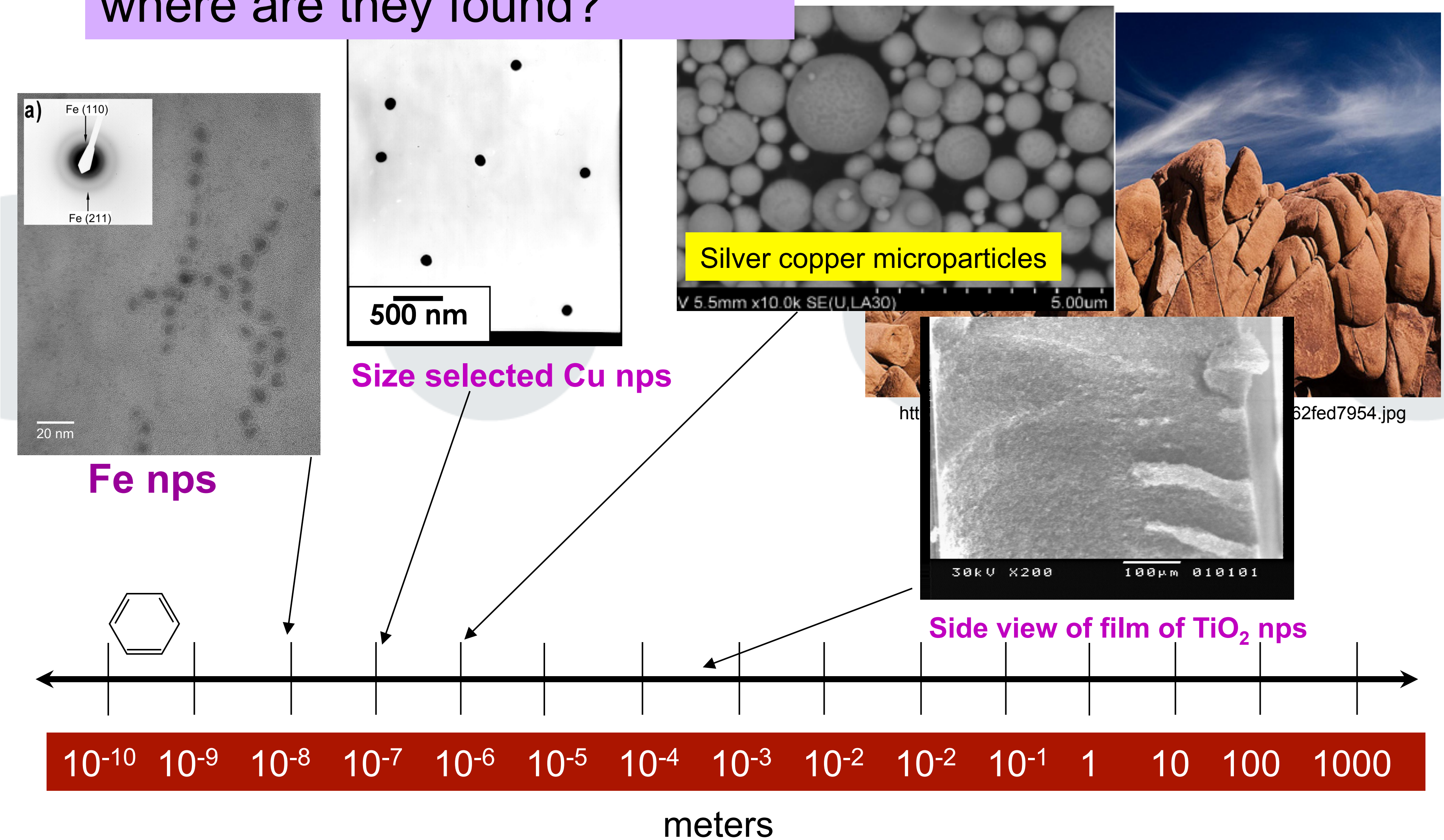
What are nanoparticles and where are they found?



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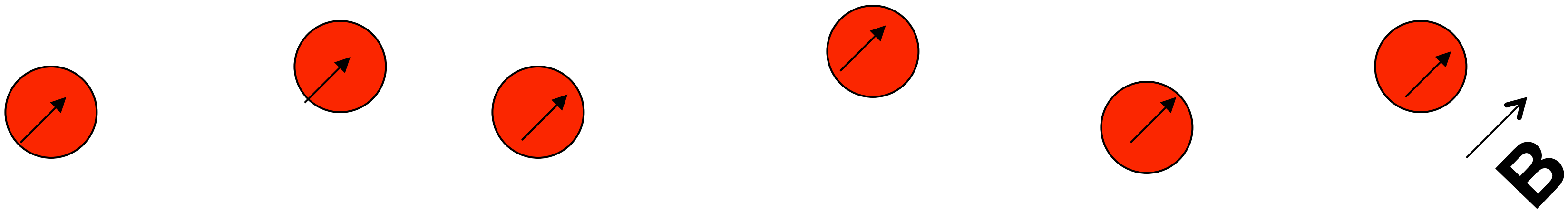
Example material: iron oxide



- Size (< 10 nm) one magnetic domain per particle, superparamagnetic nanoparticles
- MRI - positive (taken up by tissue of interest) or negative (not taken up) contrast
- Many products in clinical use
- Becomes part of normal iron load in body
- More selective active targeting via surface modification an active area of research

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Example products/future products

January 2003

300 mL | 175 µg Fe/mL

GastroMARK®
[ferumoxsil, oral suspension]

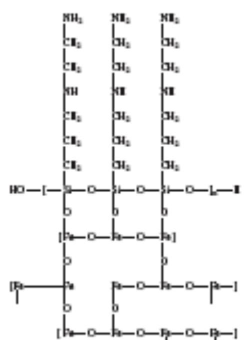
Mallinckrodt Inc.

DESCRIPTION

GastroMARK (ferumoxsil, oral suspension) is an aqueous suspension of silicone-coated, superparamagnetic iron oxide, intended for oral administration as a magnetic resonance imaging contrast media. GastroMARK is designated chemically as poly [N-(2-aminoethyl)-3-aminopropyl] siloxane coated non-stoichiometric magnetite $(FeO_x[C_5H_{13}N_2SiO_2]_y)$, which has been manufactured to obtain a small uniform particle size of approximately 0.4 microns.

GastroMARK is a turbid, slightly viscous, dark brown to orange-brown liquid formulation prepared for oral administration. The formulation contains water, sodium chloride, sorbitol, saccharin, carboxymethylcellulose, methylparaben, propylparaben, yellow dye #6, red dye #40, and flavoring. Each milliliter of GastroMARK contains 175 micrograms of iron. Each milliliter also contains 1.4 milligrams of parabens as antimicrobial agents. The osmolality of the suspension is 250 mOsm; specific gravity is 1.01 grams per milliliter. The pH is 5.5 to 9.0, adjusted with sodium hydroxide.

The product is supplied in 360 mL bottles containing 300 mL of GastroMARK.

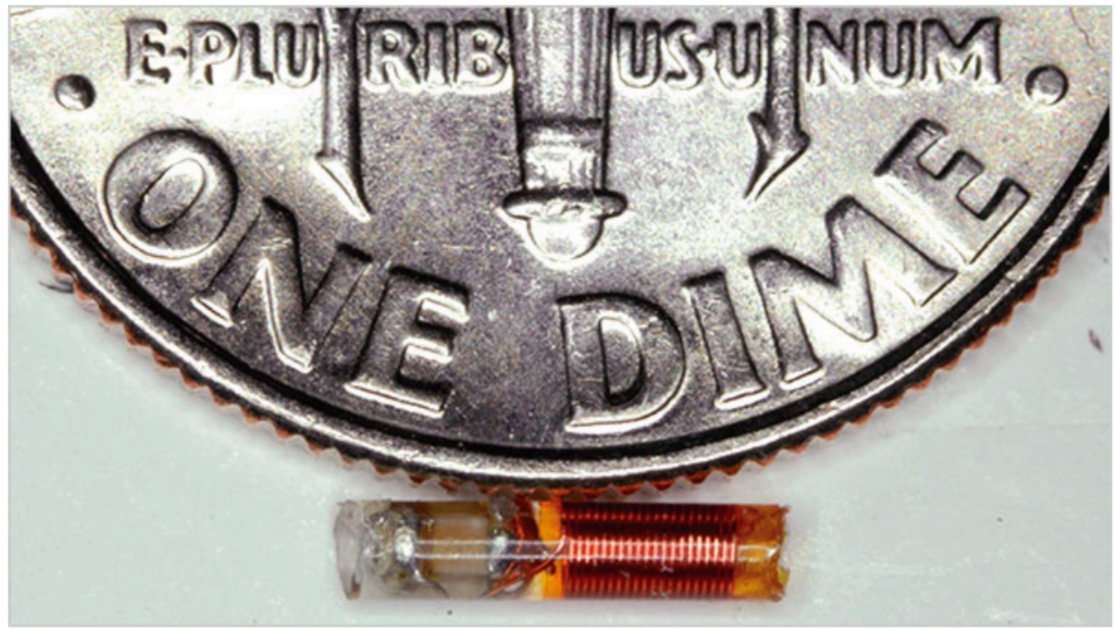


2-Dimensional Representational Formula

CLINICAL PHARMACOLOGY

General: GastroMARK is an oral aqueous suspension of a superparamagnetic magnetic resonance imaging (MRI) contrast agent. After oral administration of GastroMARK, the agent fills the stomach and small intestine by 30 to 45 minutes after ingestion. The imaging agent passes distally to the large intestine by 4 to 7 hours after ingestion. GastroMARK is primarily eliminated in the feces.

Beyond the Biopsy: A Tiny Monitor for Cancer



Christophoros Vassiliou

Michael J. Cima led a team that created an implantable device that may one day track cancer growth in humans.

T2 Biosystems, US

http://www.nytimes.com/2009/08/30/business/30novel.html?_r=1&scp=1&sq=t2%20b

http://www.amagpharma.com/documents/products/pdfs/gastromark_insert.pdf

Nanoproducts, consumer

©D. Hawxshurst/Wilson Center

Safe handling of nanotechnology, Nature 444, 267-269 (16 November 2006)

*http://www.nanotechproject.org/inventories/consumer/analysis_draft/

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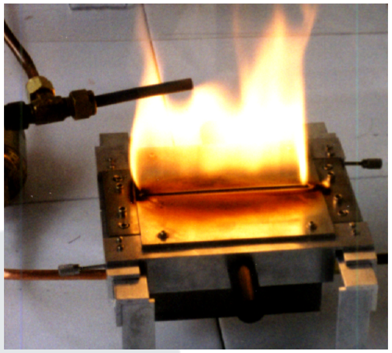
Methods of making nanoparticles

- Starting from molecular level
  - From precursor

  Aerosol
    - Combustion synthesis
    - Thermal or plasma synthesis

  Solution phase synthesis
    - Precipitation
    - Sol gel
    - Emulsion
  - Evaporation/condensation
- Starting from cluster level
  - Spray pyrolysis
  - Electrospray



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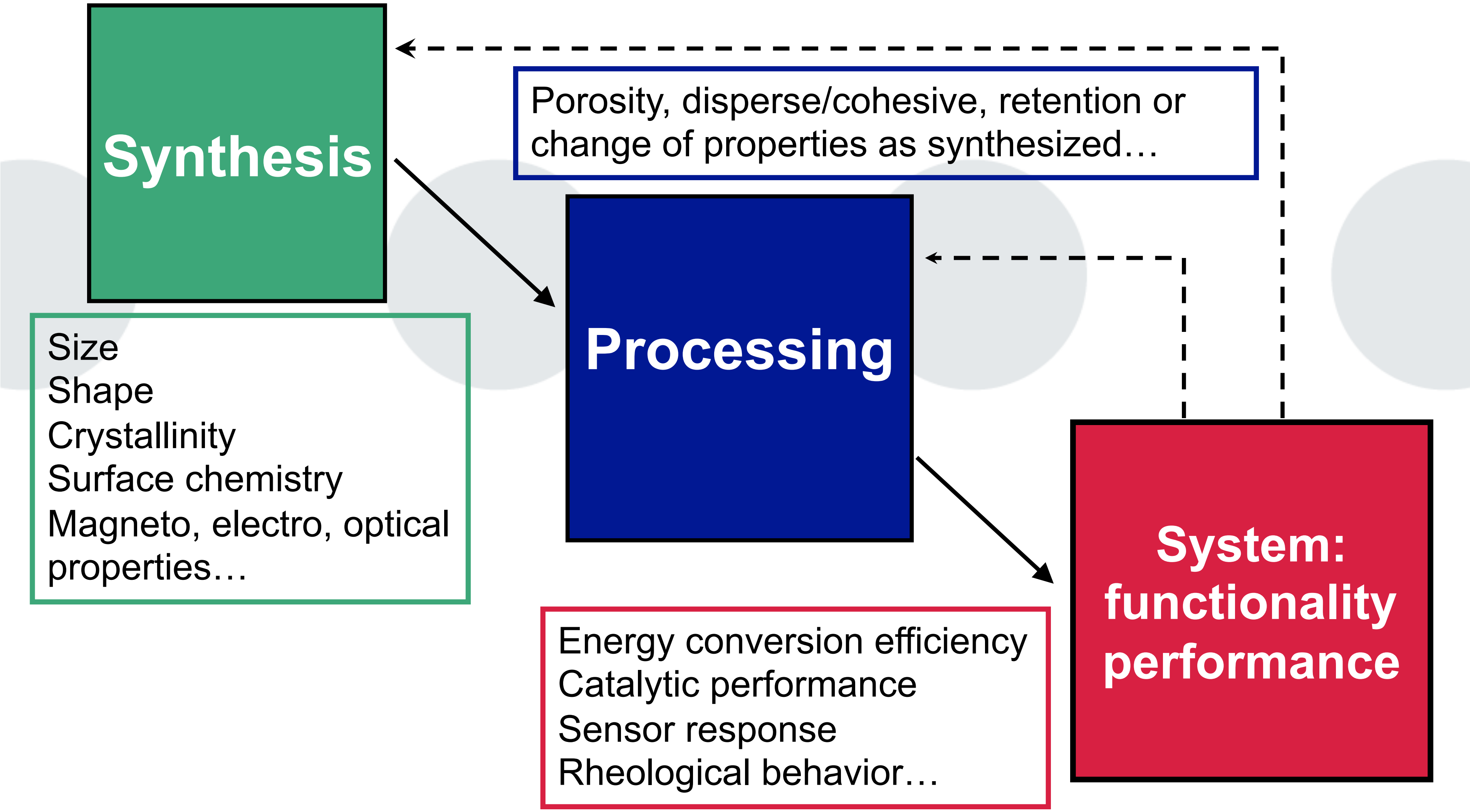
Nanoparticle engineering



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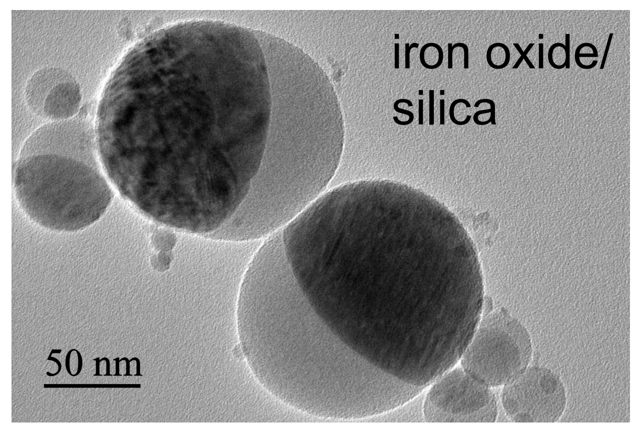
Pros and cons

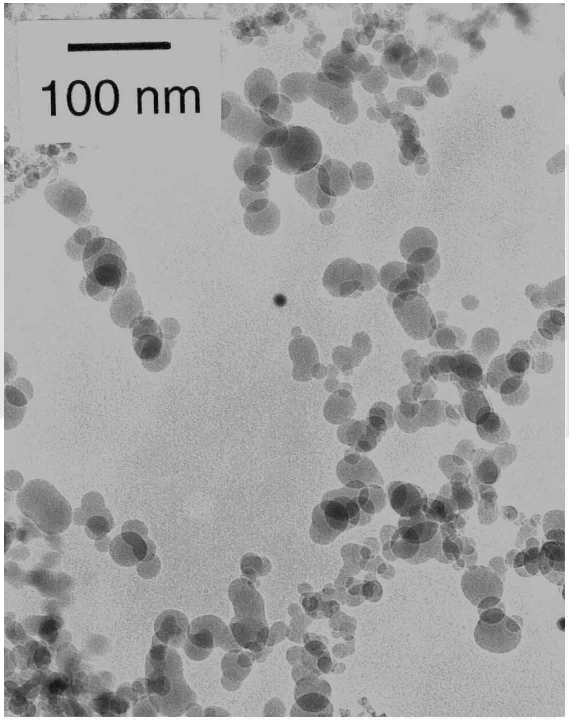
+ Rapid
+ Simple, less steps required
+ Product dry, no solvents
+ Amenable to continuous processing
+ May be scalable

– Limited size control
– Poor control of aggregation, especially gas to particle conversion
– May battle thermodynamics in mixed systems



Chaos is cheap, any proof?

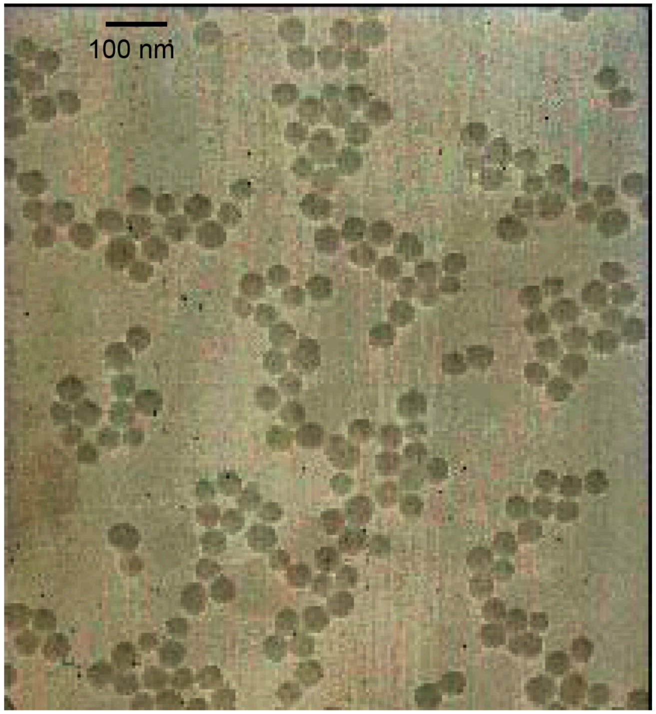
Fumed silica from our lab, not for sale



From alibaba.com

Fumed silica (dry) as low as $130/metric ton

Colloidal silica (stöber synthesis) from our lab, not for sale



Colloidal silica (30%)
$600-$1000/metric ton

Aerosol manufacturing, $$

Product	Volume, tons/yr	Market $/yr	Process
Carbon black	8 M	8 B	Flame
Titania	2 M	4 B	Flame
Fumed silica	0.2 M	2 B	Flame
Zinc oxide	0.6 M	0.7 B	Hot wall furnace
Fe, Pt, CeO_2	0.02 M	0.3 B	Hot wall furnace, spray pyrolysis

Ref: K. Wegner, S.E. Pratsinis, *Chem. Eng. Sci.* **51**, 4581 (2003)

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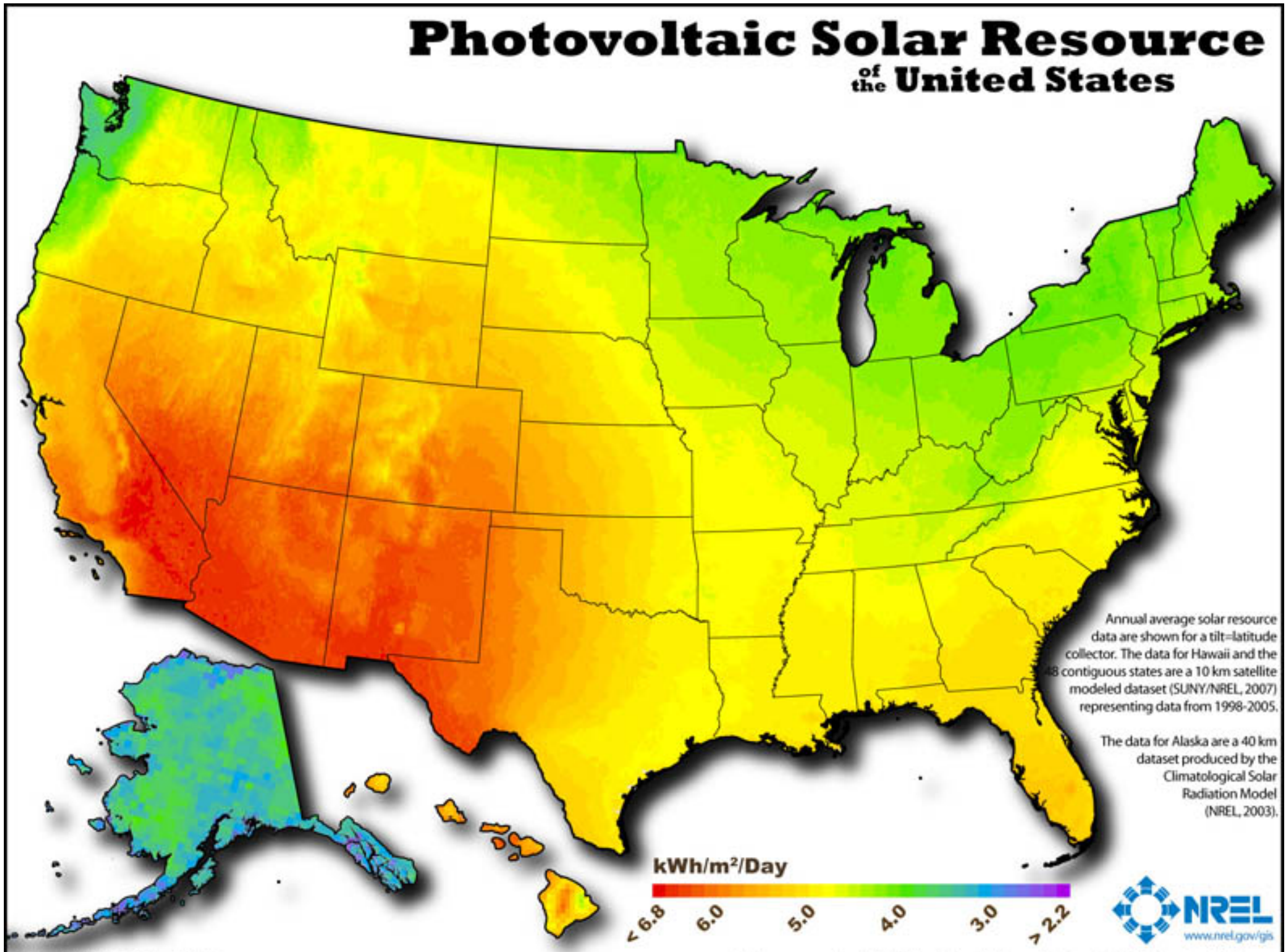
Solar energy, the potential



http://www.nrel.gov/gis/images/map_pv_national_lo-res.jpg

[redacted]

Solar energy is intermittent

- Storage of solar energy is key
- Two approaches (of many):
- Solar to electric to chemical energy

  Photovoltaic (PV) + battery, supercapacitor, or…
- Solar to chemical energy

  Photoelectrochemical cells (PEC)

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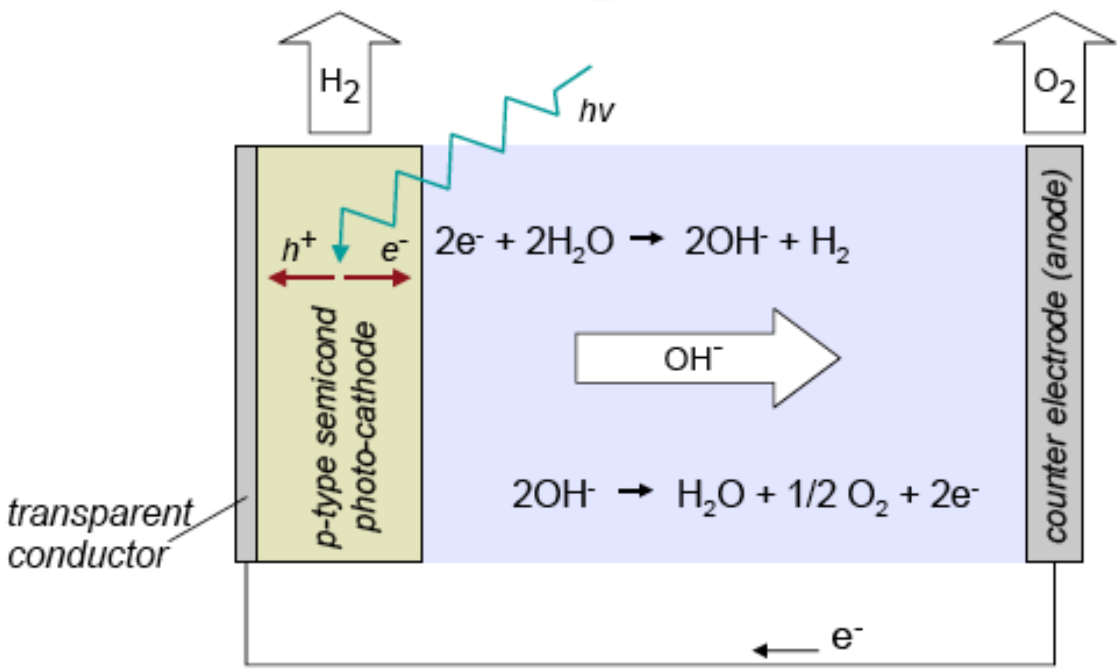
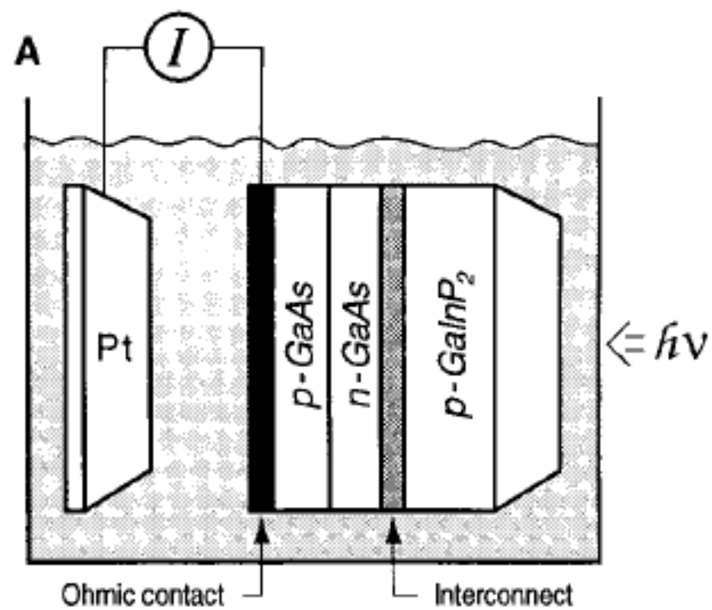
Perspective

- State of the art: solar to chemical



12.4% efficiency



Khaselev and Turner, Science **280**, 425 (1998)

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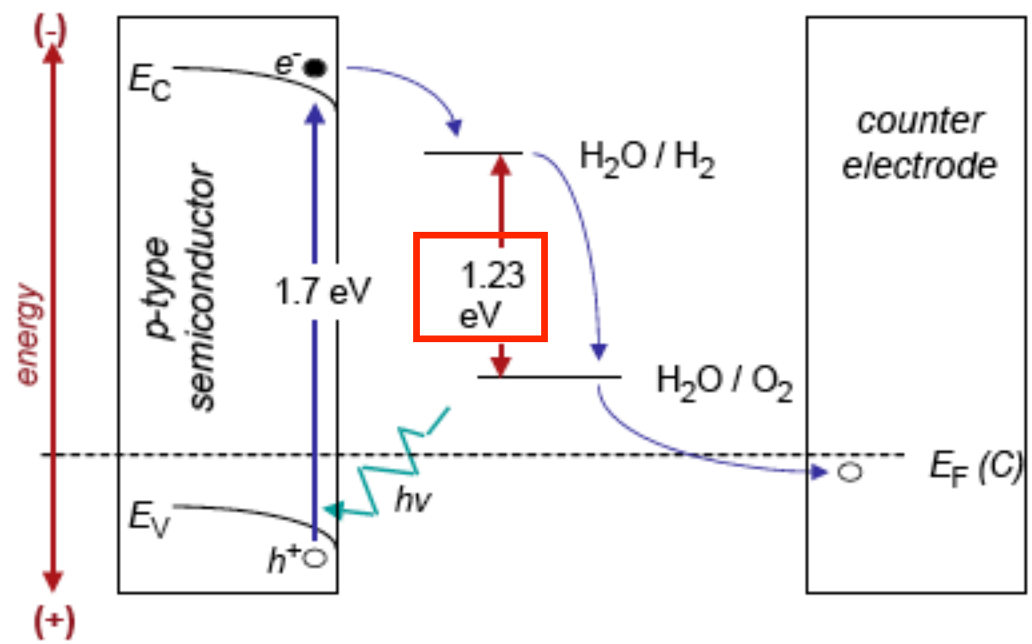
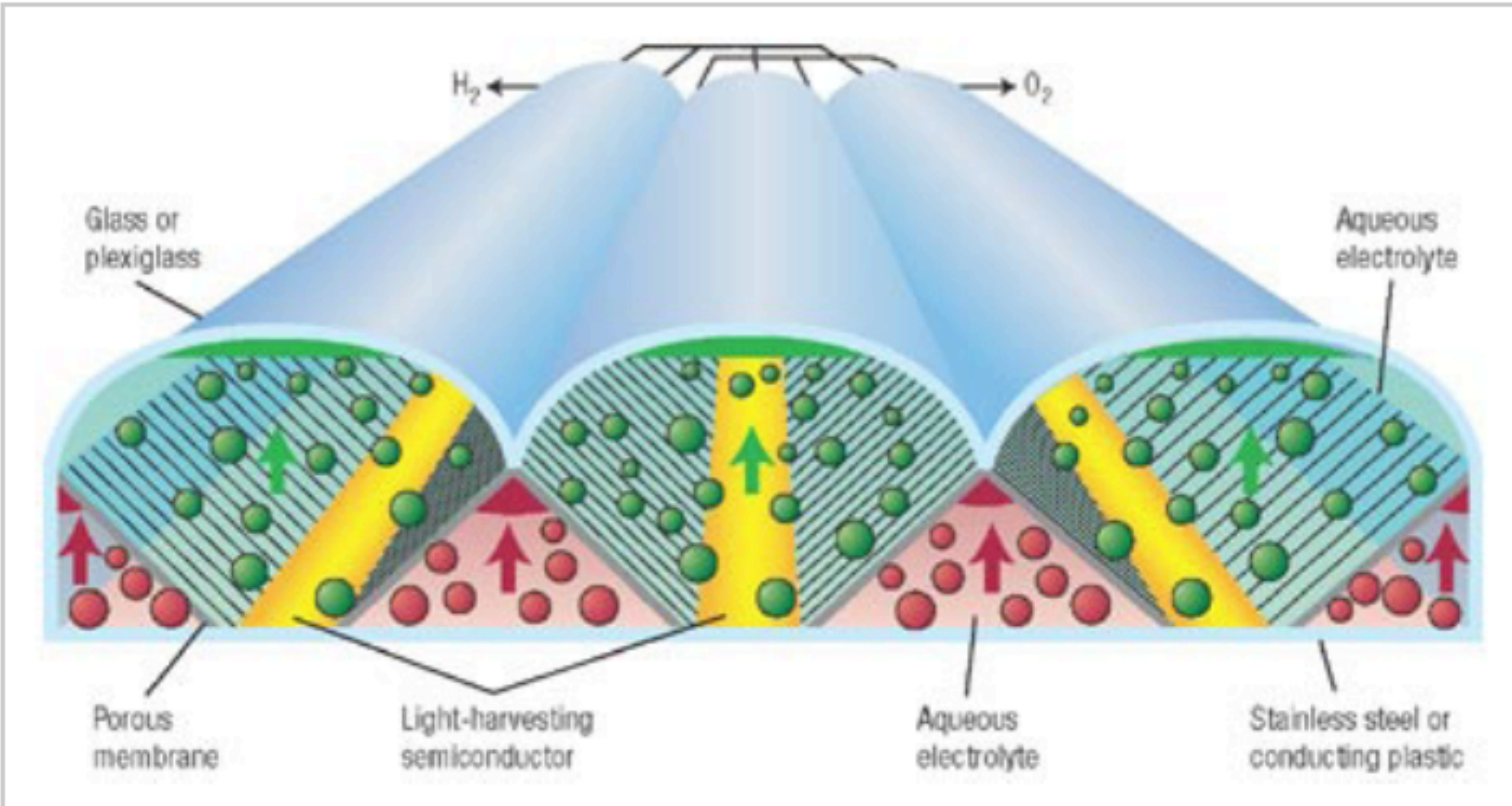
Perspective: materials constraints

- Semiconductor band gap of at least 1.4 eV required because of polarization, but should still absorb in the visible range (< 2 eV)
- Long electron-hole pair lifetimes
- Stability in electrolyte
- Efficiency and cost



On a large scale, imagine….



J. Turner, *Nature Materials*, **7**, 770-771 (2008)

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H_2 as transportation fuel

The Hindu, 2 September, 2005
Bajaj Rickshaw Prototype
http://www.hinduonnet.com/2005/09/02/stories/2005090204461400.htm

January 2012
15 H_2 powered rickshaws (Mahindra & Mahindra) deployed in New Delhi's Pragati Maidan Exposition Center
A Yee, NY Times, Oct 1, 2012

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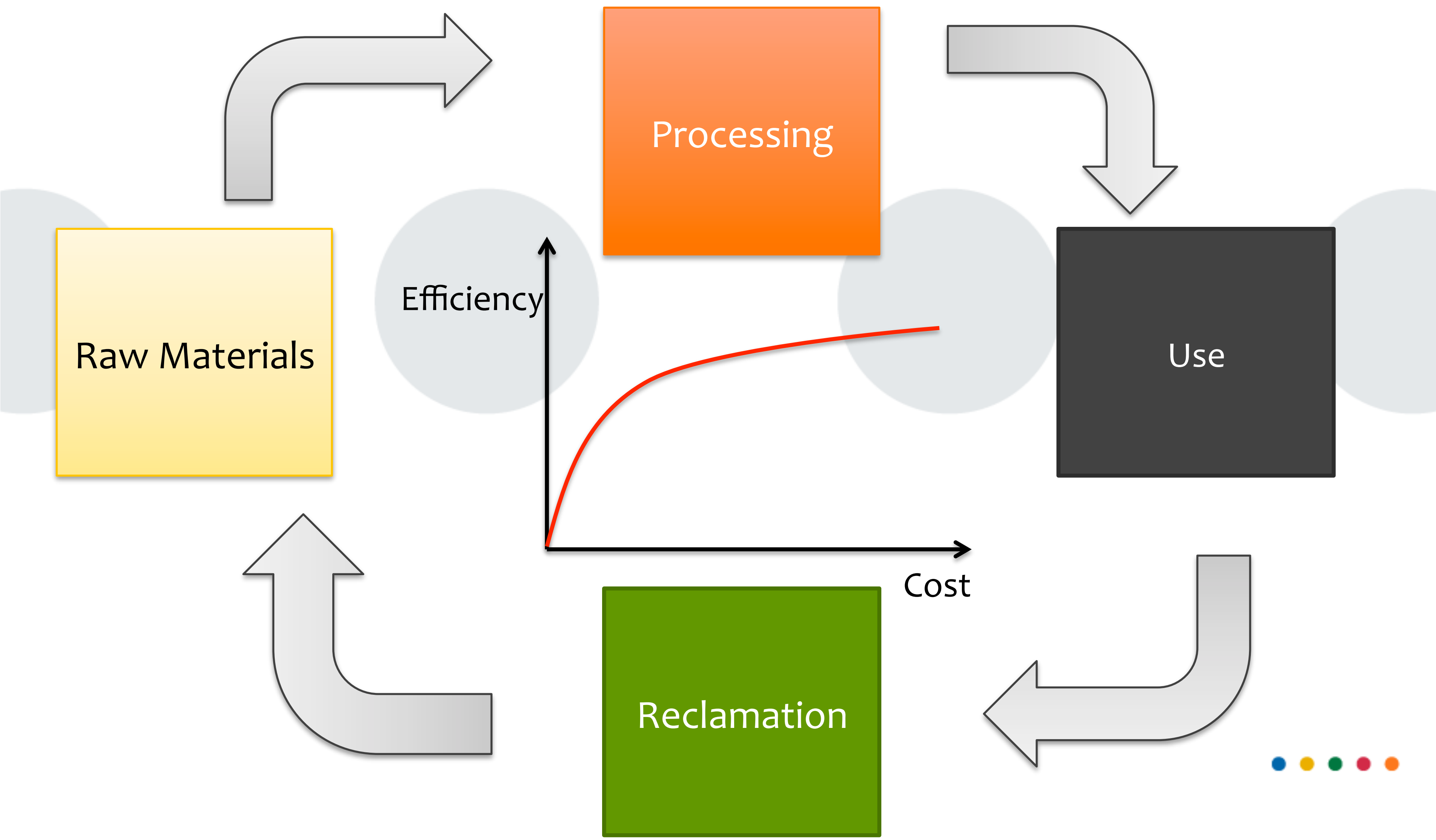
Perspective: life cycle



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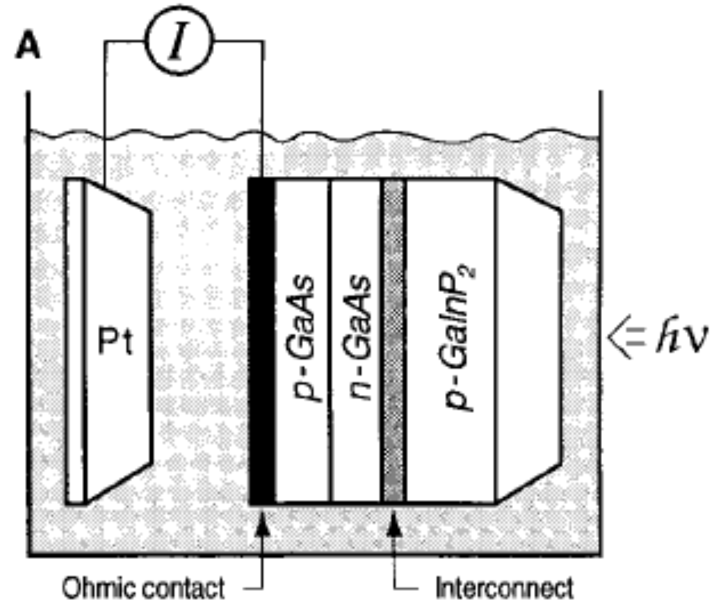
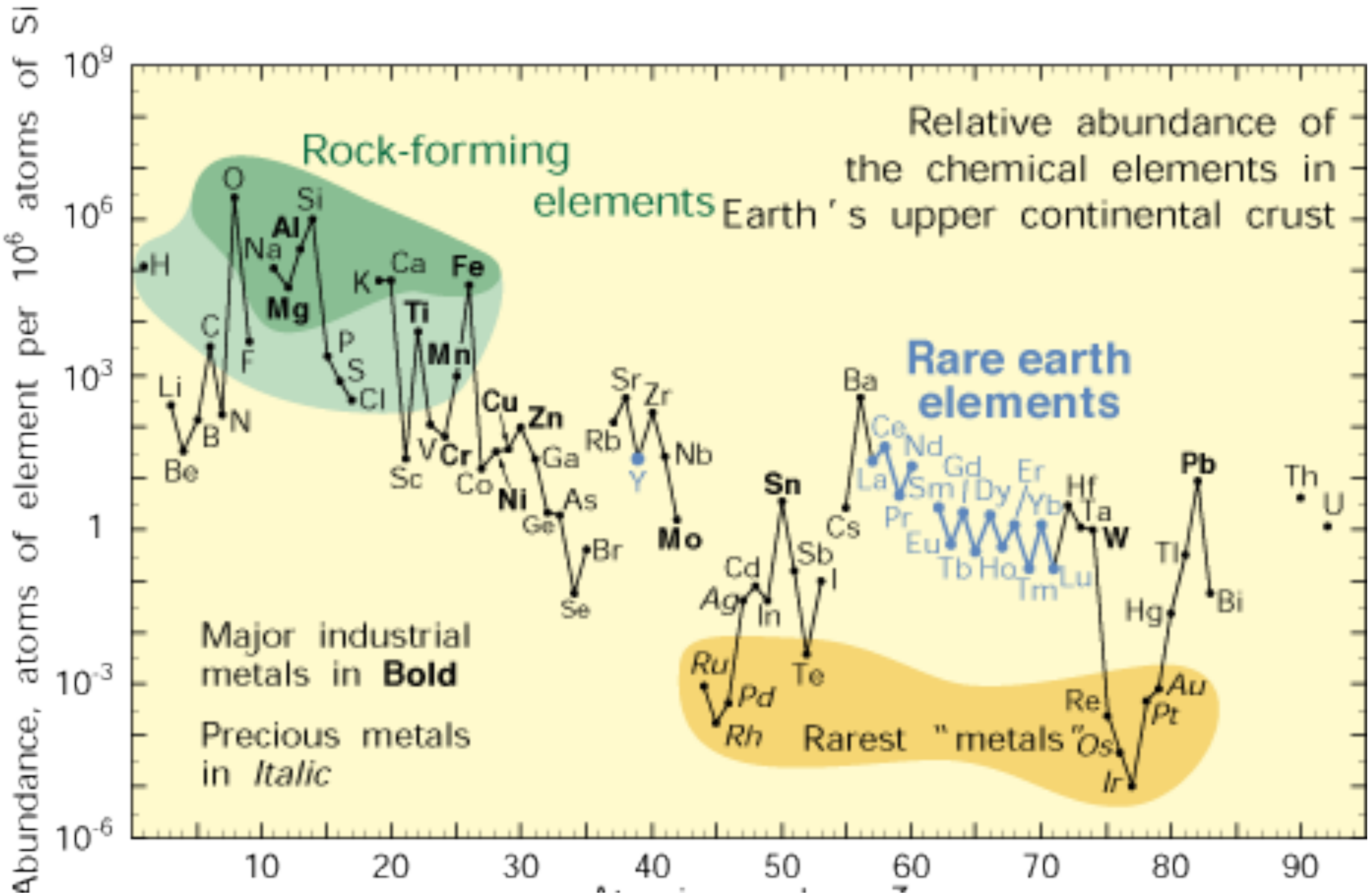
Approaches to materials optimization

- Start with highest efficiency, try to reduce costs, here, combination photovoltaic/ PEC device, NREL, eff: 12.4 %
- Or… start with inexpensive materials and try to improve efficiency
- Materials of interest include CuO, ZnO, TiO_2



Khaselev and Turner, Science **280**, 425 (1998)

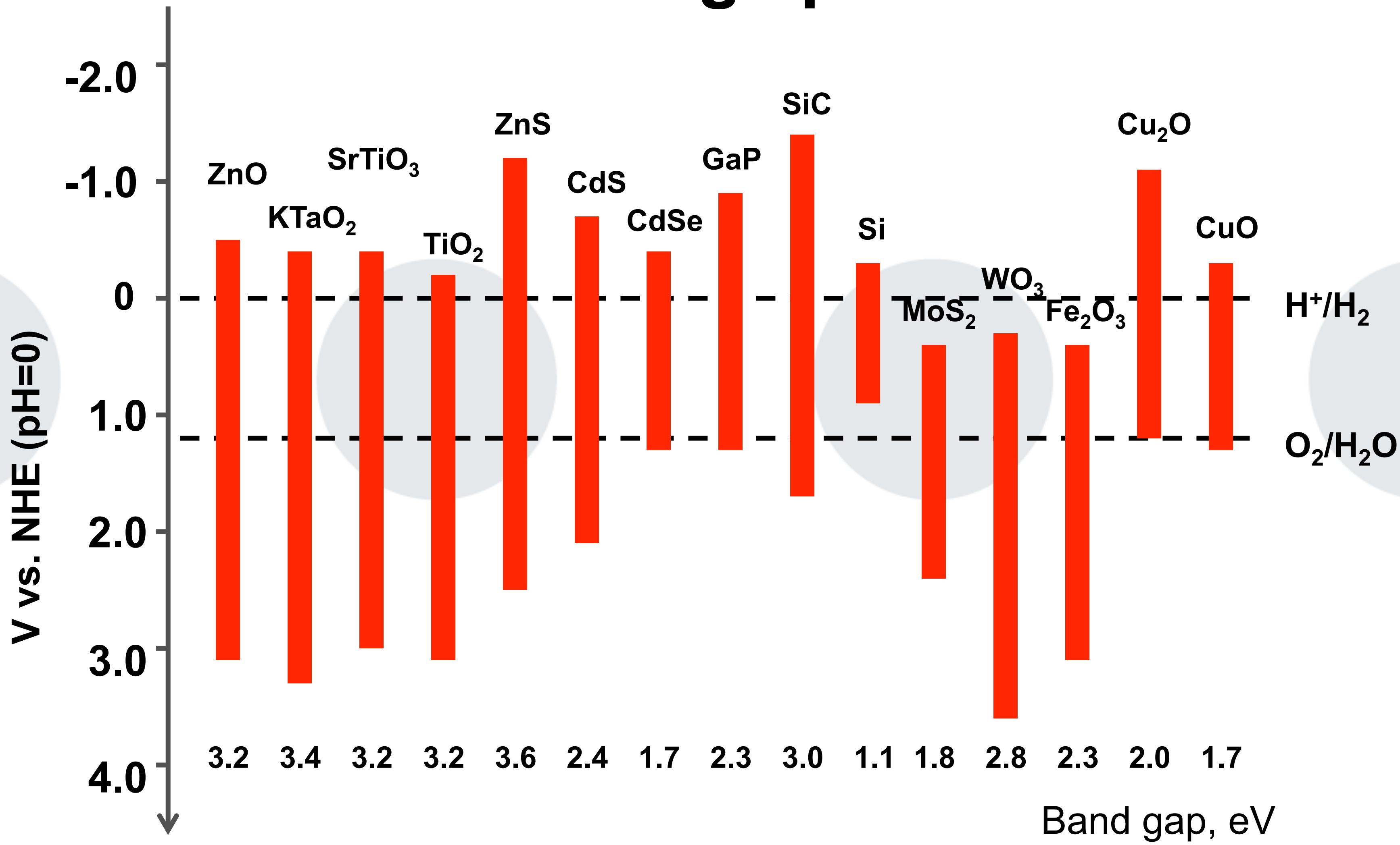
Earth abundance



Atomic number, Z

http://pubs.usgs.gov/fs/2002/fs087-02/

Favorable band edge position



ZnO to Fe2O3 adapted from Navarro Y. et al., ChemSusChem. 2009; 2:471-485
Cu2O adapted from Gerischer H., Pure Appl. Chem 1980: 52: 2649-67

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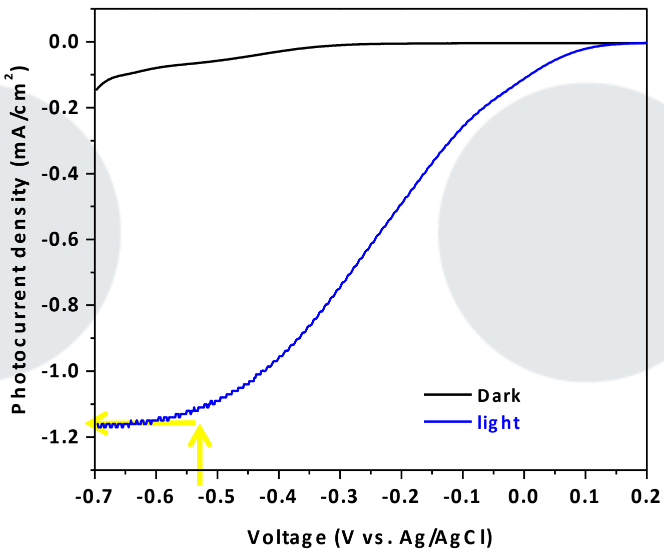
Conversion efficiency



$$\eta_c = \frac{\boxed{j_p}(E^0_{rev} - \boxed{V_{bias}})}{\boxed{I_0}} \times 100\%$$

η_c	:	Solar to hydrogen conversion efficiency (%)
I_o	:	Incident light intensity (W/m^2)
j_p	:	Photocurrent density at a certain applied voltage (mA/cm)
E^0_{rev}	:	Standard water splitting reaction potential (1.23 V vs. NHE) at pH =0.0
V_{bias}	:	Applied external potential (V vs. RHE)

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CuO systems to date, low efficiency

System	Light source	Incident power density (W/m^2)	Photocurrent density (mA/cm^2)	Solar to H_2 efficiency (%)	Reference
CuO	500W Xenon	1000*	0.08	0.06*	Nakaoka et al., 2004
CuO	150W Xenon Arc	8100**	2.2	0.20**	Chauhan et al., 2006
2% Li-CuO	150W Xenon Arc	8100	0.44	0.05	Koffyberg and Benko,1982

* Assume to be 1000 W/m^2
** Assume to be 8100 W/m^2

Photo - electro – chemical

light absorption

material with
small bandgap

electron separation

cell architecture

chemical reaction

material with
right band position

light reflection

cell architecture

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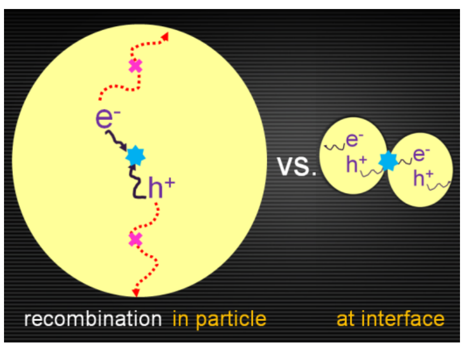
Efforts to improve performance

light absorption | **electron separation** | **chemical reaction**

light reflection

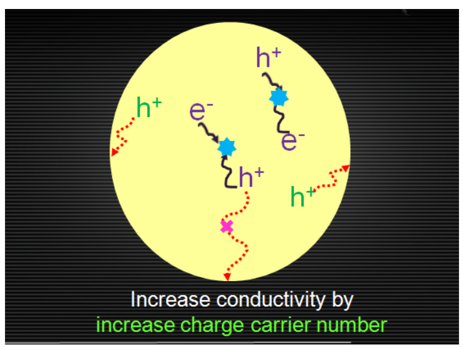
(1) Decrease particle size



Chiang et al., Int J H2 Energy, **2011**

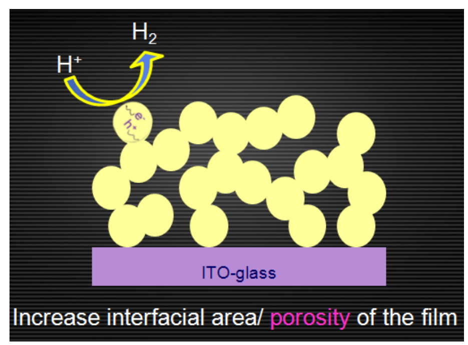
Chiang et al., Int J H2 Energy, **2012a**

(2) Increase conductivity



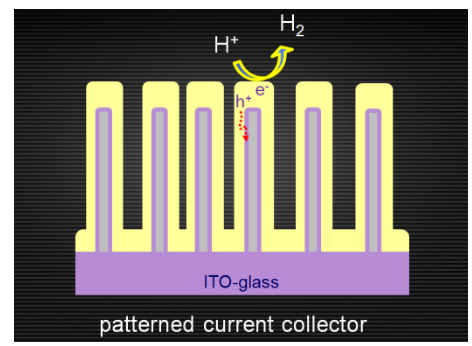
Chiang et al., J Electrochem Soc **2012**

(3) Increase porosity



Chiang et al., Int J H2 Energy **2012b**

(4) Shorten transport distance



Chiang et al., Nano Lett. 2012

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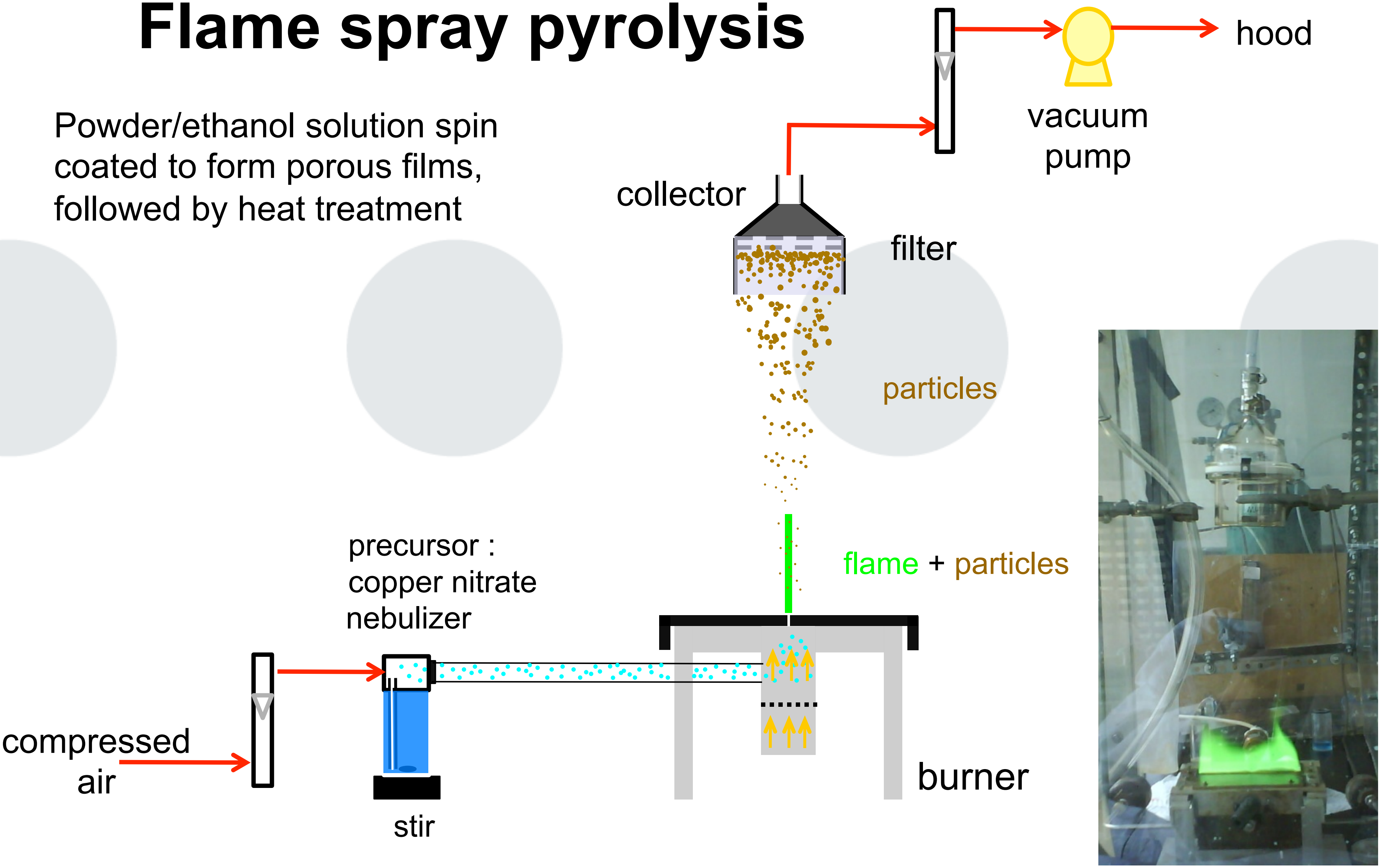
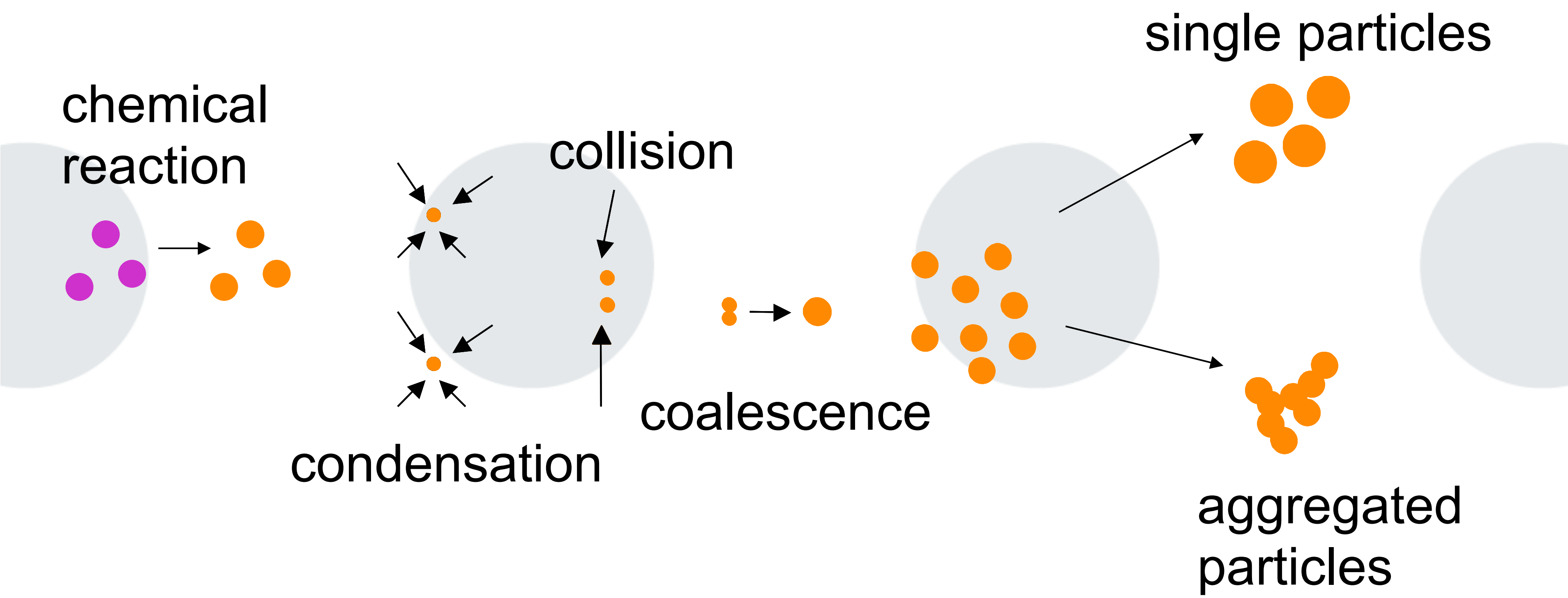
Flame spray pyrolysis

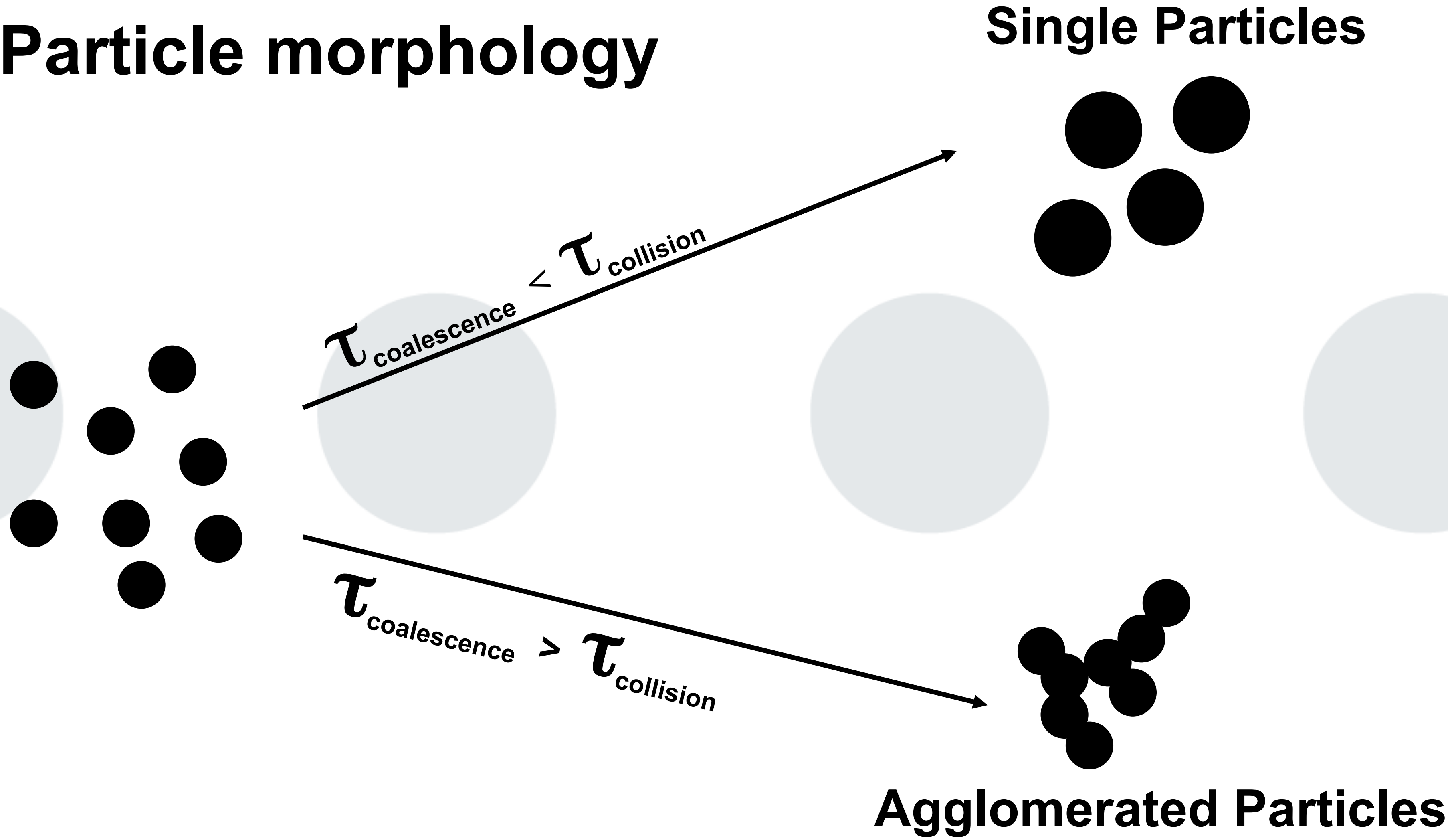
Powder/ethanol solution spin coated to form porous films, followed by heat treatment



Processes occurring



Particle morphology



Ulrich (1970’s), Helble, Sarofim (80’s), Friedlander, Koch, Windeler, Lehtinen (90’s)

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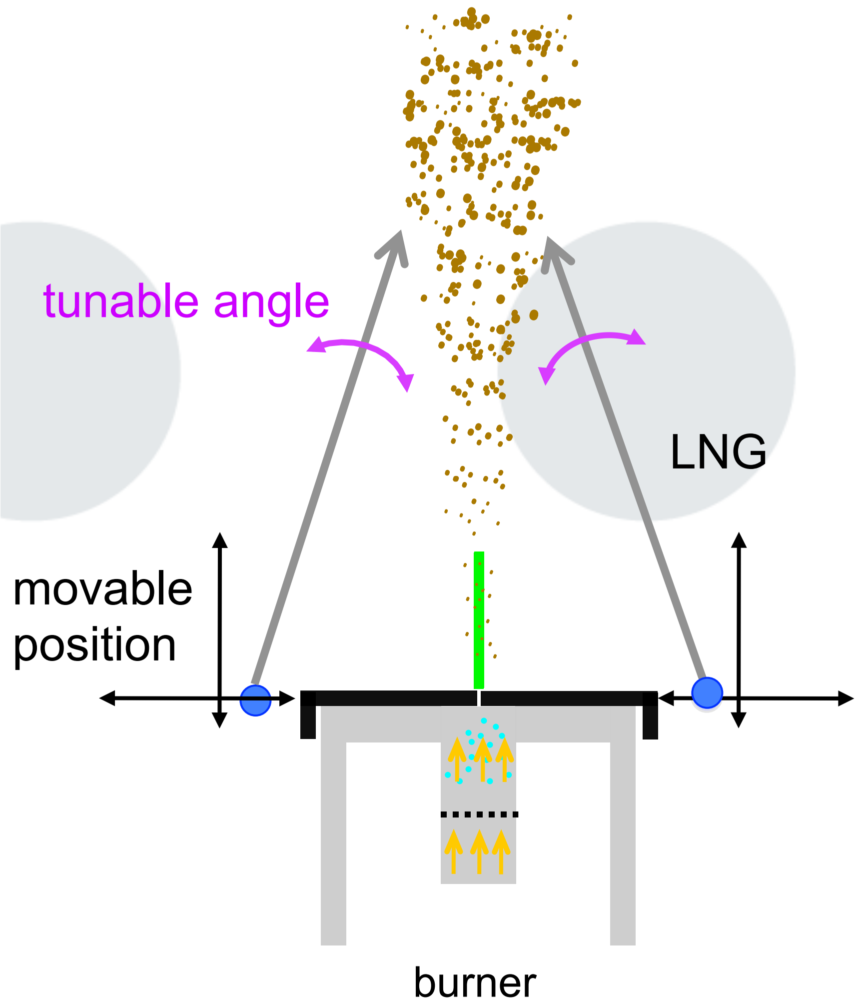
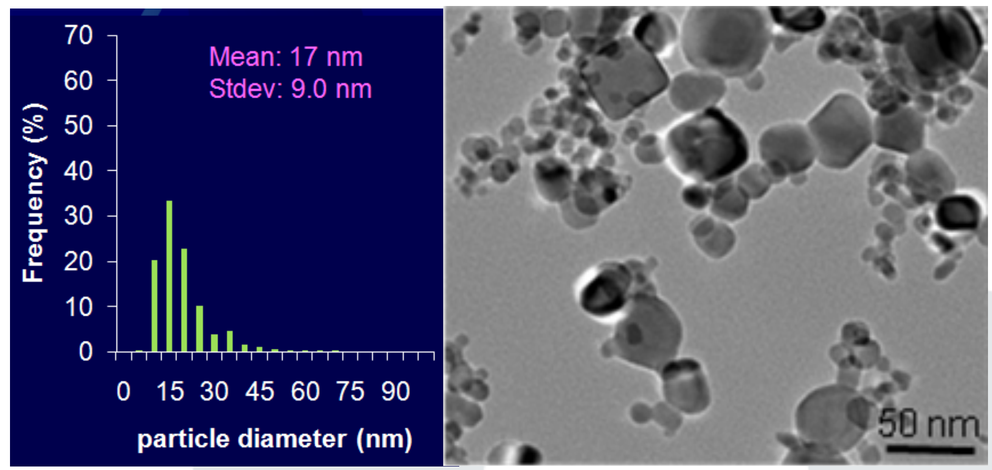
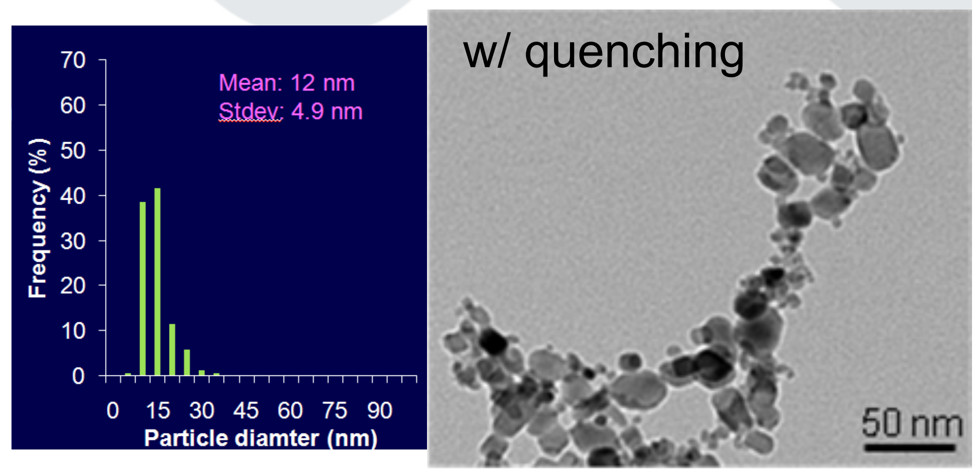
Control of particle size



w/o quenching

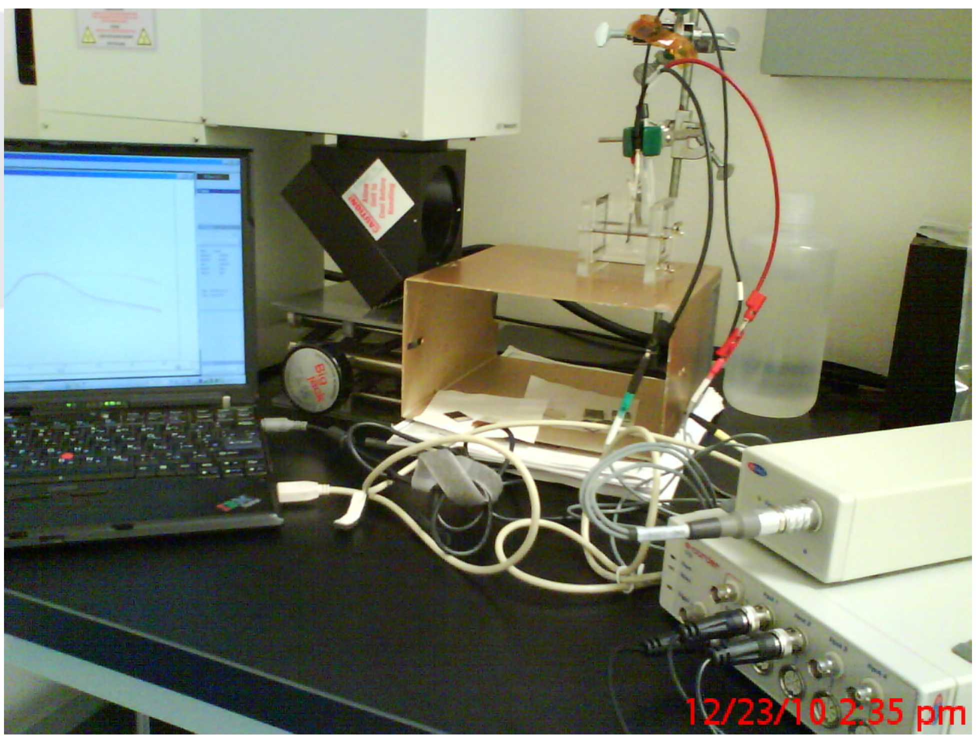


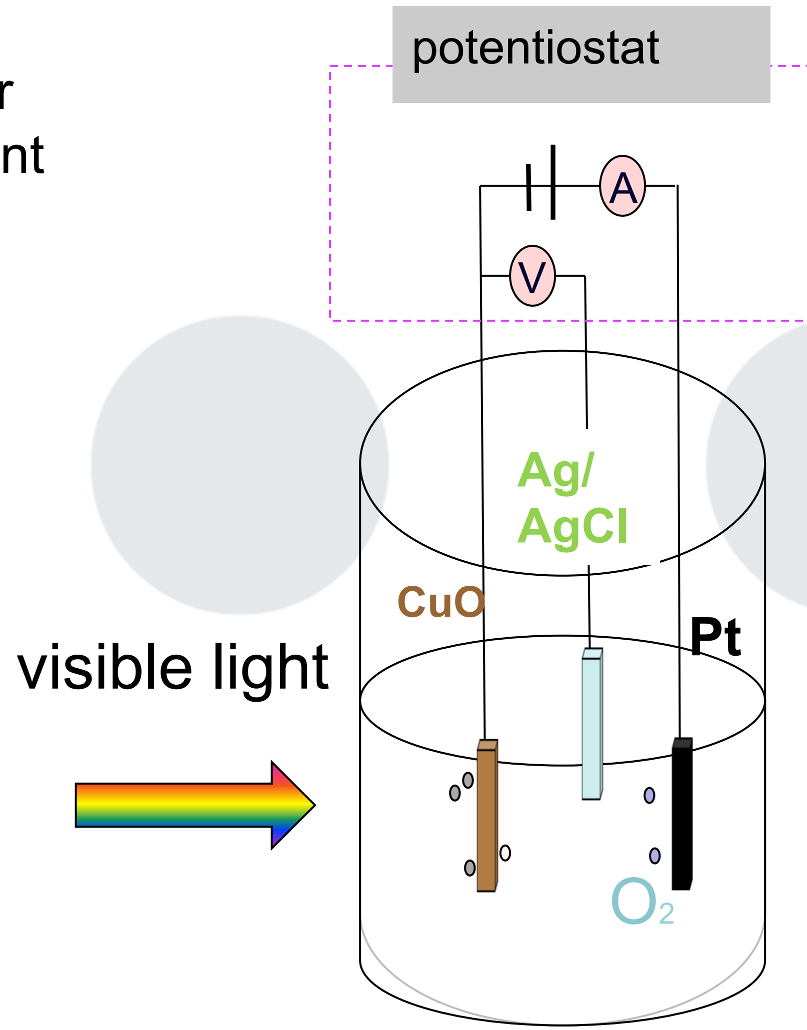


30% smaller

PEC test station

150 W Oriel solar simulator + full reflector beam turning mirror, AM1.5G filter, incident intensity = 1 sun (1000 W/m^2)

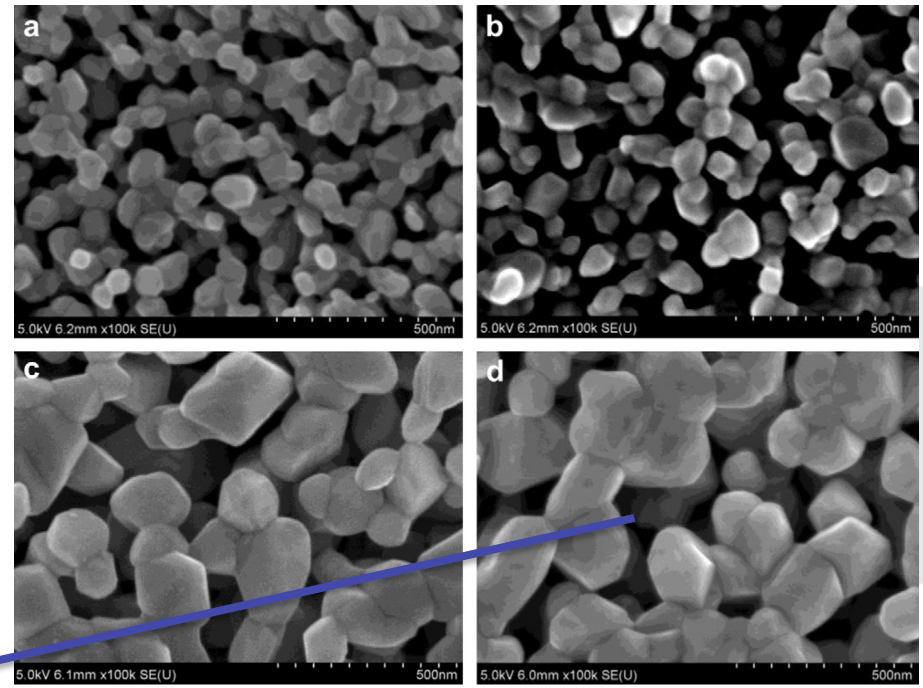


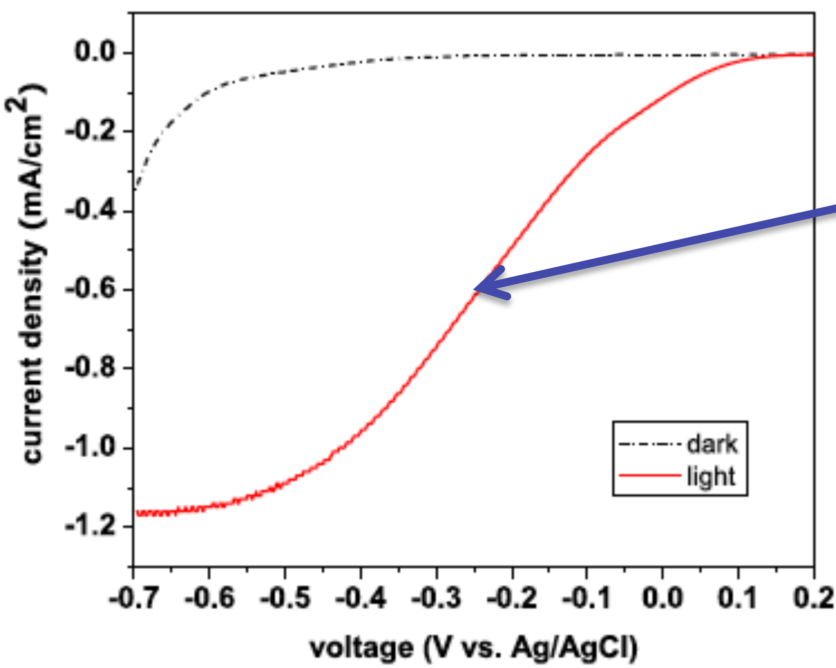


1M KOH electrolyte, pH 14

Post processing treatment optimized

Spin coated films made from powder/ethanol
Scanning electron micrographs of samples sintering at different temperature and period: (a) 450 C for 1 h, (b) 450 C for 3 h, (c) 600 C for 1 h, (d) 600 C for 3 h.





Solar to H_2 efficiency 0.3%

Chiang et al., Int J H_2 Energy, **2011**

Chiang et al., Int J H_2 Energy, **2012a**

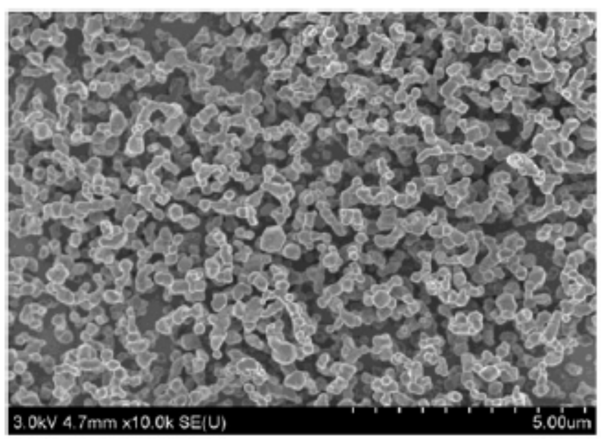
Precipitation route

copper nitrate + NaOH

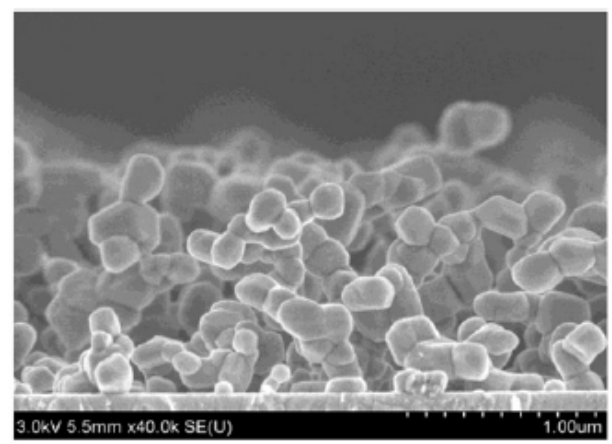
Transmission electron micrographs of samples produced at (a) 25 C for 24 h, (b) 60 C for 45 min, (c) 95 C for 30 min

+ heat treatment 600 C,1 hour

Solar to H_2 efficiency 0.7%



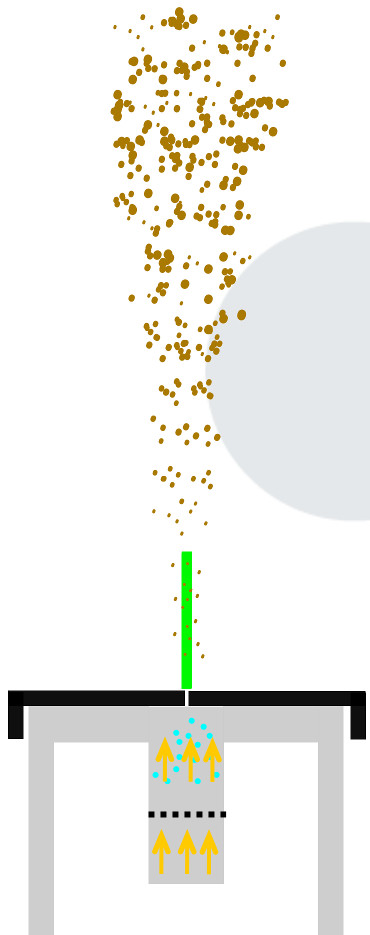
Bird-eye view

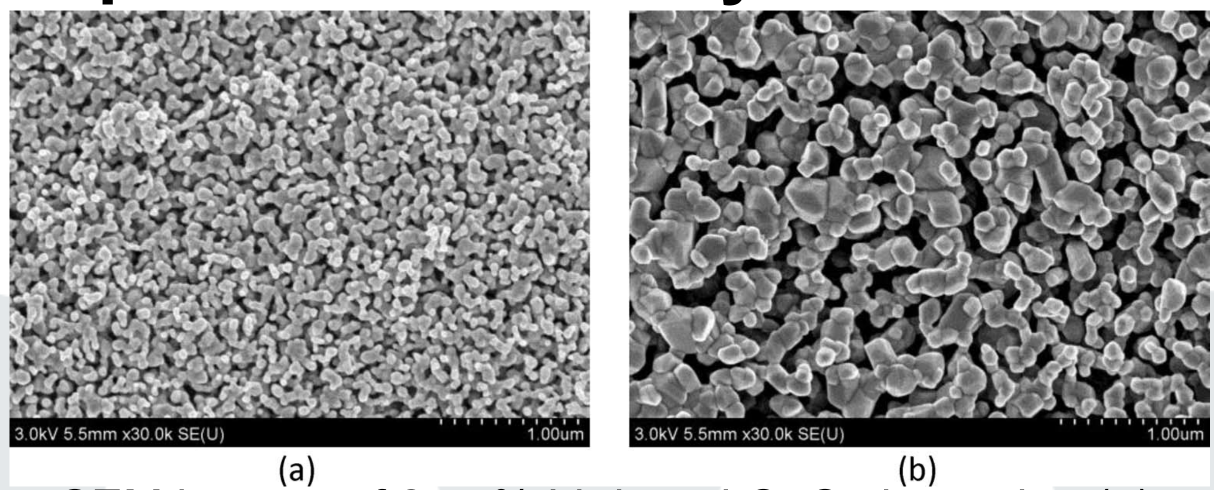


Side view

Chiang et al., Int J H_2 Energy **2012b**

Addition of Li to improve conductivity





SEM images of 2 at % Li doped CuO sintered at (a) 450◦C for 1 hr and (b) 600◦C for 3 hr.

Conductivity CuO only: 2×10^{-6} to 4×10^{-6} S/cm
with 2% Li 7×10^{-5} to 3×10^{-4} S/cm

Solar to H_2 efficiency 1.3 %

Aqueous solution of copper nitrate + lithium acetate

Chiang et al., J Electrochem Soc **2012**

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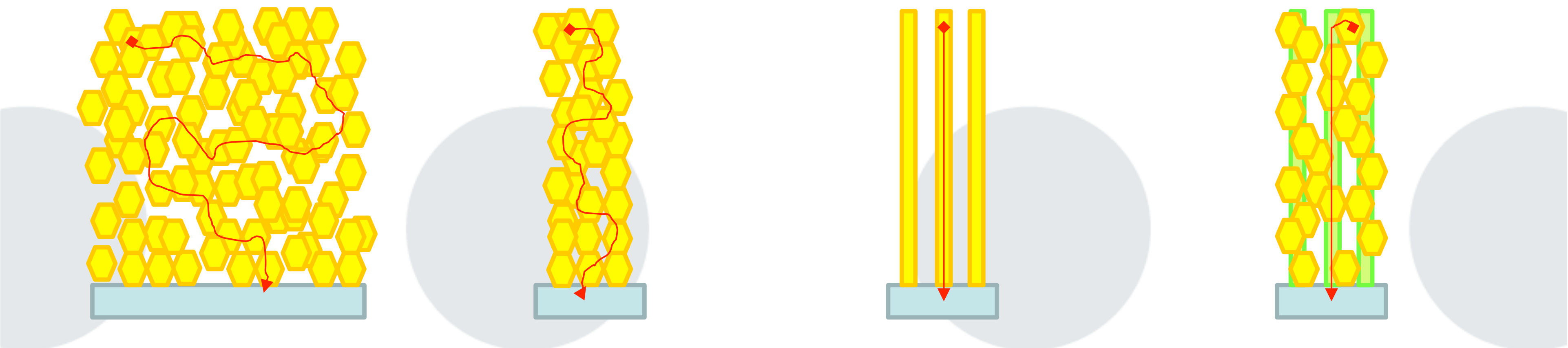
Additional structures



Polycrystalline film

Polycrystalline nanorod

Single crystalline nanowire

Current collector + polycrystalline

3.2 % efficiency

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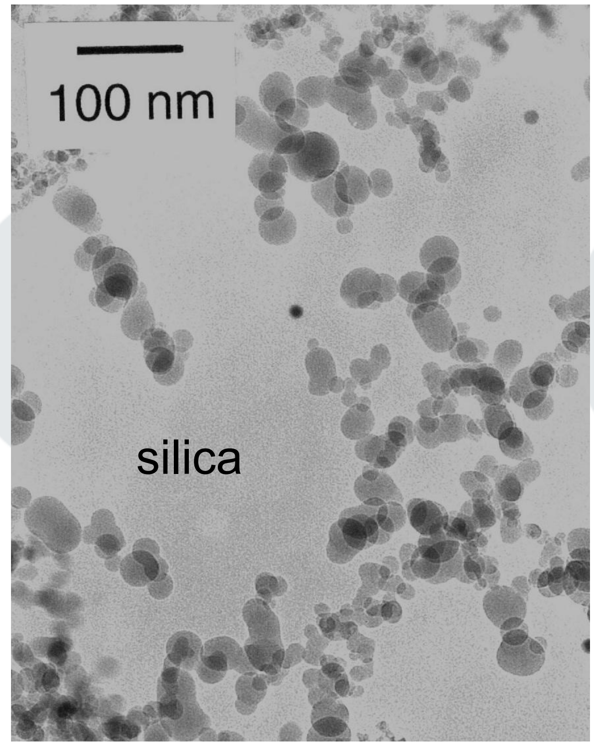
55 gallon drum of silicon tetrachloride vs fumed silica

surface area = 1.6 m^2

https://www.khlubes.com/images/55_gallon_drum.jpg



surface area = 2.4×10^7 m^2
assuming 300 m^2/g

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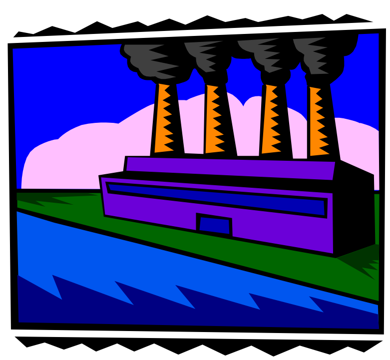
Potential risks associated with engineered nanomaterials

- Exposure opportunities over the life cycle of the nanomaterial, intended, unintended
  - Raw materials
  - Manufacturing
  - Use
  - Disposal/reclamation



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Routes for exposure, intentional or unintentional

- Dermal
- Gastrointestinal
- Inhalation

Toxicity measurements in animals
LD50 = dose required to kill 50% of a population of test animals (mg/kg)

OSHA Permissible exposure limits (PELs) in humans, 8 h time weighted average (mg/m^3)

Size of material considered in some cases, ex: respirable quartz, but no separate standards for nanomaterials

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MSDS info on titania, not nano

5 nm particles have 100 x the surface area per gram as 500 nm particles

Cas: 13463-67-7
RTECS #: XR2275000
Name: TITANIUM DIOXIDE. MOLECULAR FORMULA: TIO*2. MOLECULAR WEIGHT: 79.9.

OSHA PEL: 15 MG/M3

~ 275 x EPA 24 h standard for 2.5 micron diameter or less particulate air pollution

*PEL 8 hour time weighted permissible exposure limit

- https://fscimage.fishersci.com/msds/23510.htm

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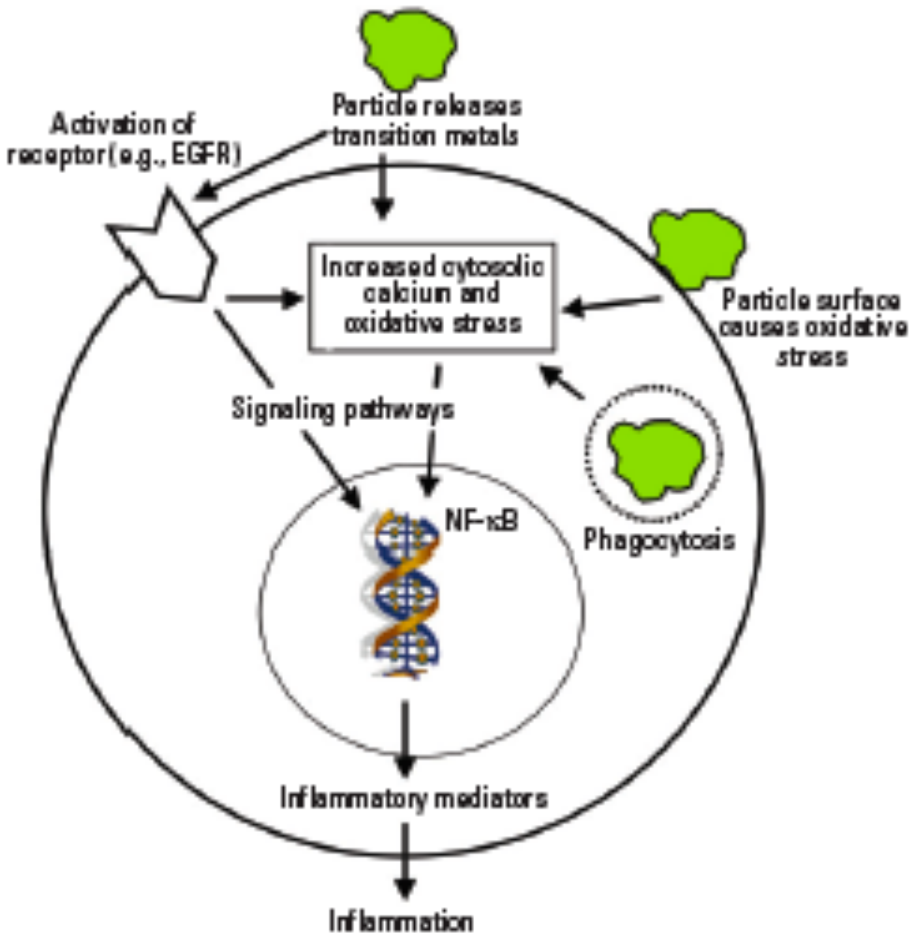
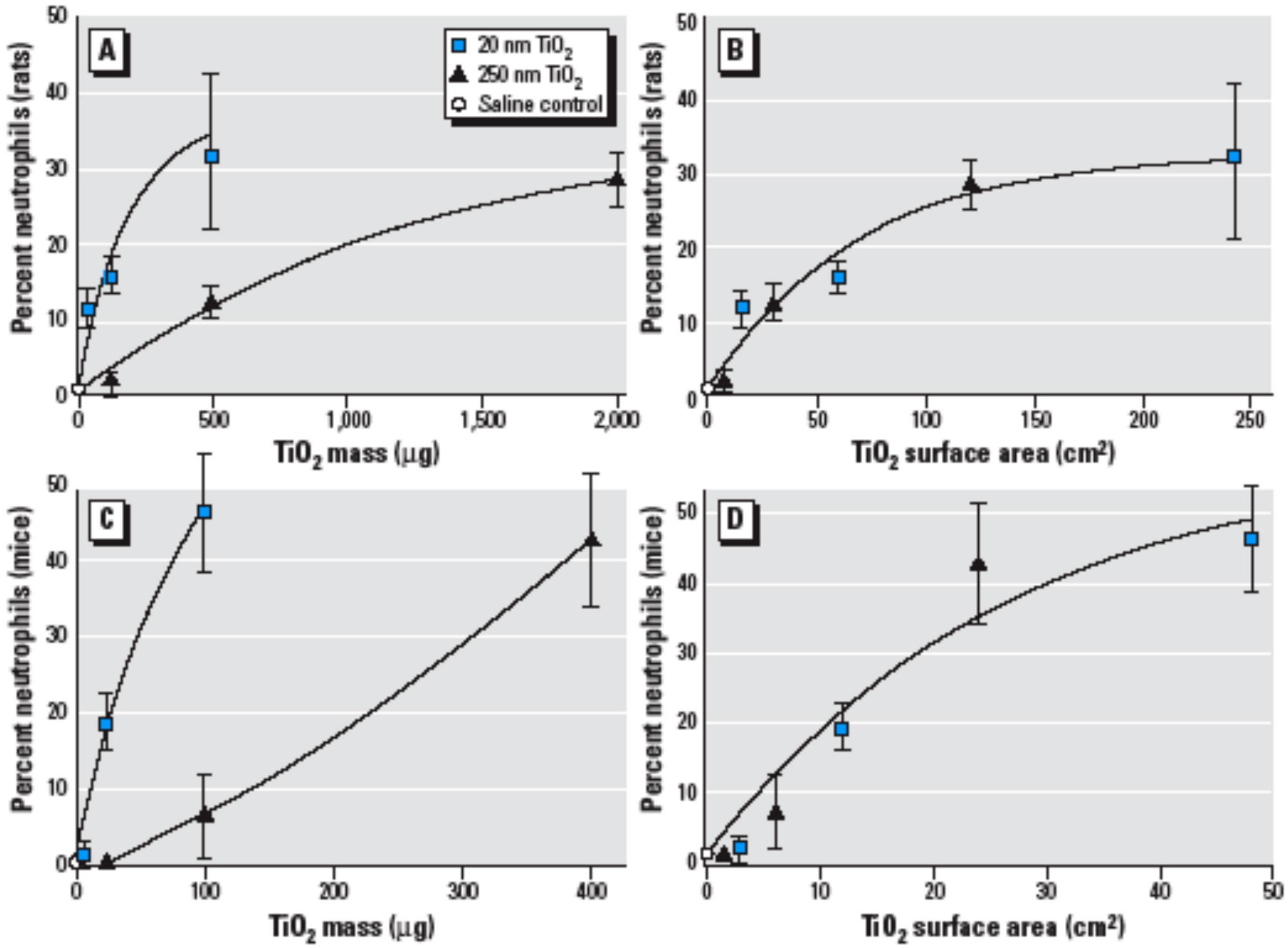
Primary pathways hypothesized at the cellular level



- Particle surface/cell interactions cause oxidative stress, increased intercellular calcium and gene activation
- Transition metals from NPs cause same
- Cell surface receptors are activated by transition metals from NPS leading to gene activation
- NP/mitochondria interactions cause oxidative stress

Figure in Oberdoerster et al., Environmental Health Perspectives, 113, 823-838 (2005)
Adapted from Donaldson and Tran Inhal. Toxicol. 14 5-27 (2002)

Response in rats and mice correlates well with surface area, for inhaled engineered NPs



Oberdörster G, Philos Trans R Soc Lond A 358(2000) 2719–2740
Oberdörster et al., Environ. Health Perspectives, 113 (2005) 823-839

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Not just surface area

- Free silanol group intensity

Pandurangi et al., *Environ Health Perspect* **86**: 327-36 (1990)

- Ability to catalyze reactive oxygen species formation *in vitro*

Limbach et al. *Environ Sci Tech* **41**: 4158-4163 (2007)

.....

- No generalized trends have emerged across all material systems

What about combinations of nanoparticles with other substances?

Real exposure scenarios - may not be single component

- Other components physi- or chemisorbed to the engineered nanomaterial surface
  - Nanoparticle based targeted therapeutics
  - Consumer product formulations
  - Accidental release of nanoparticles together with condensable or reactive species
  - Nanoparticles modified by environment between release and exposure

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Our testable hypothesis

- Initially non-toxic nanoparticles may serve as carriers of toxic compounds into biological systems
- Adverse effects may correlate with nanoparticle specific surface area

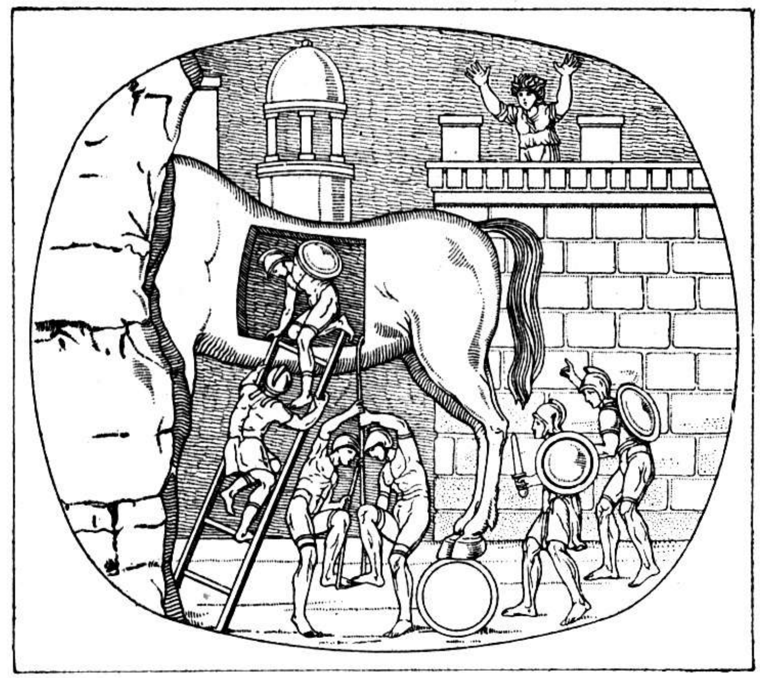

Image: http://www.php4windows.com/Articles/trojan_horse_in_business_home_software.jpg

Testing this hypothesis, system components

- Relatively non-toxic ubiquitous nanoparticles
  - Fumed silica, Aerosil
  - Evonik, ~ 50 through 350 m^2/g specific surface area
- Low molecular weight water soluble (~ 2.4 g/L) toxic molecule, cisplatin
- Positive control, quartz, ~ 1 micron mass median diameter
- Cell line, bovine chondrocytes

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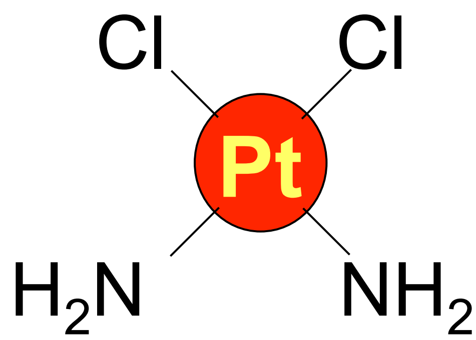
About cisplatin



- Potent antitumor agent used to treat different type of cancers
- Cytotoxic mode of action
  - Interacts with DNA
  - Forms intra-strand cross-linkages
  - Leads to activation of apoptosis
- Cisplatin and some other chemo agents adversely effect skeletal system of children, thus effects on chondrocytes previously studied

Experimental approach

- Attachment of toxin (cisplatin) to non-toxic (fumed silica) surface
- Characterization of the surface modified silica particles
- *in vitro* tests on chondrocytes
  - Membrane integrity via fluorescence microscopy using the Live/Dead (calcein/ ethedium bromide) stain
  - Metabolic function via the MTT (methylthiazol tetrazolium) assay
  - Electron microscopy

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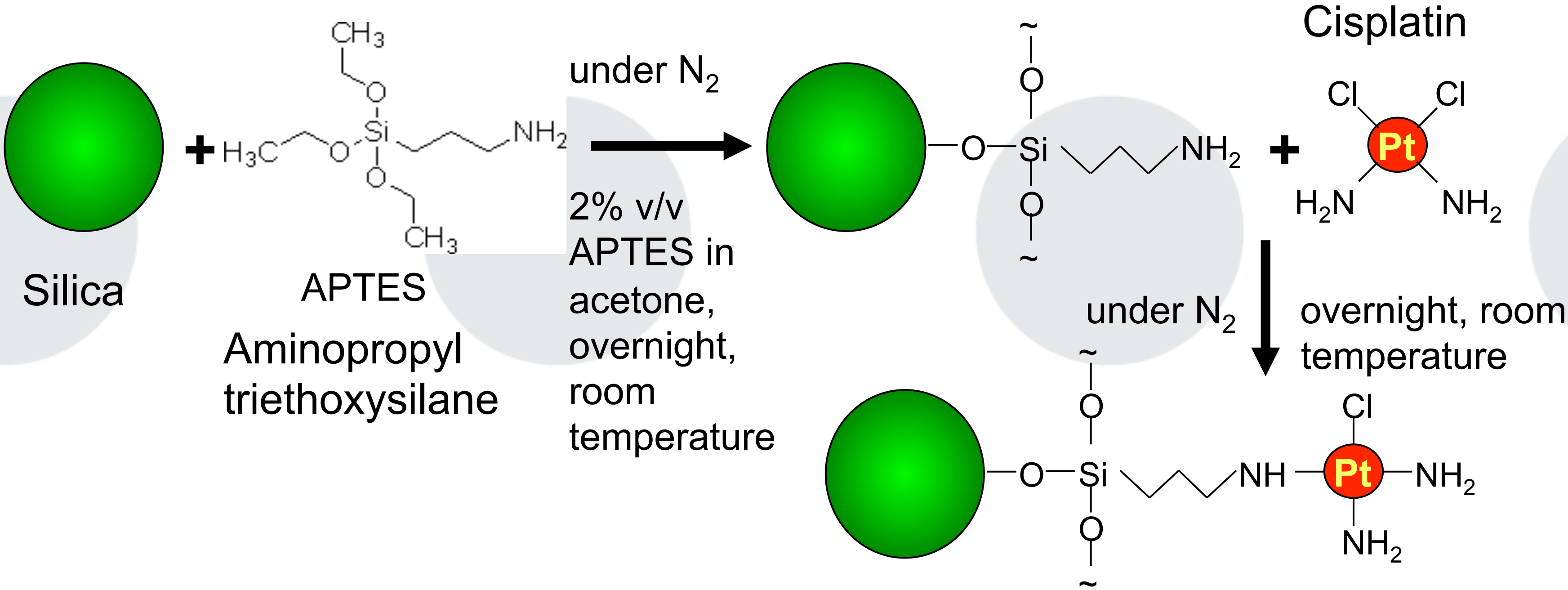
Attachment of cisplatin to silica



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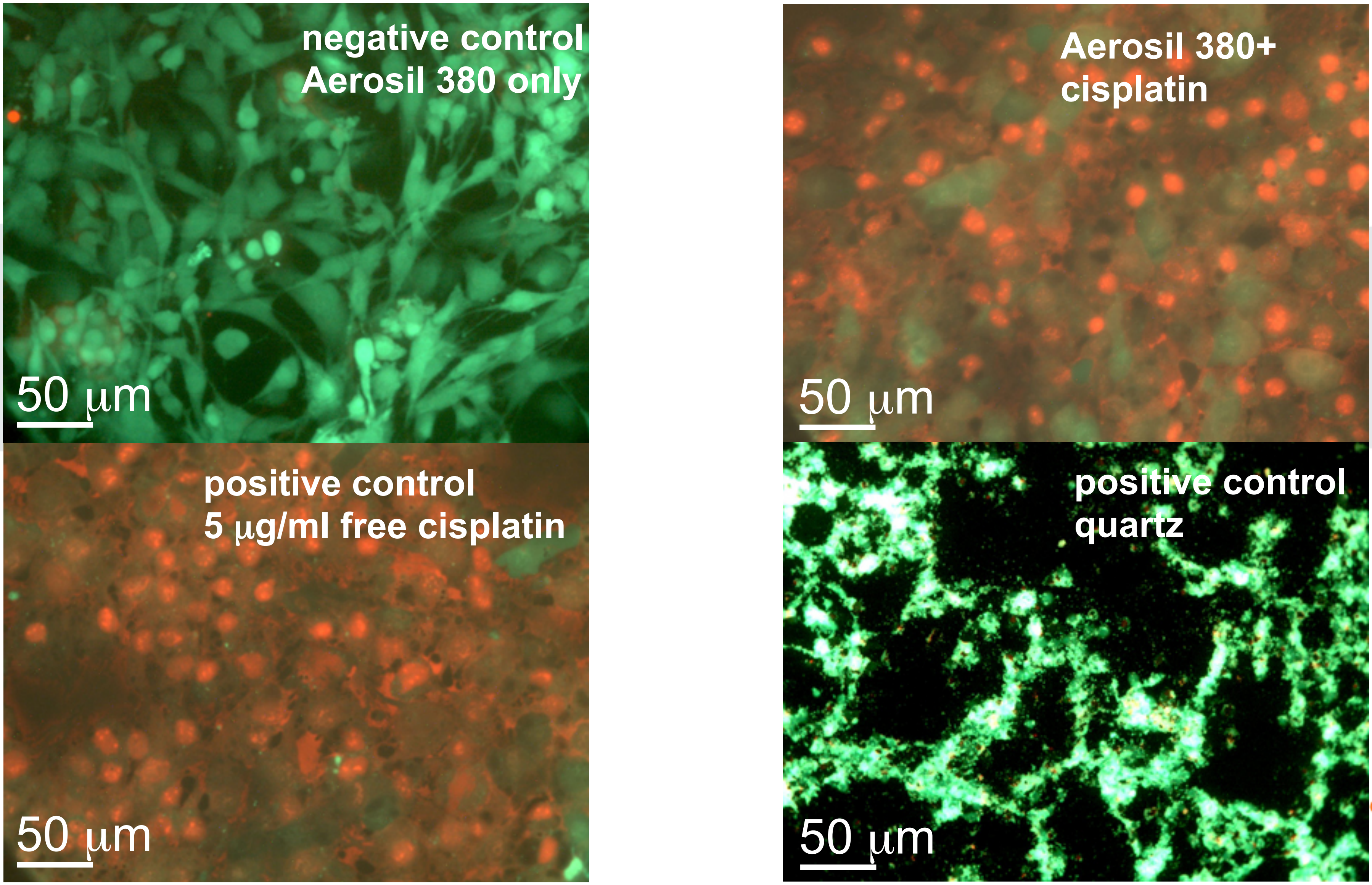
24 hrs exposure

Fluorescence microscopy, Live/Dead stain



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MTT assay

- Cell culture wells washed, then 300 µl of MTT solution (0.5 mg/ml) added
- Plates incubated at 37 ^{0}C for 2 hrs allowing for reduction of tetrazolium salt to a formazan (blue) dye by active mitochondria of the cells
- 300 µl of solubilizing buffer (10% Triton X 100 with 0.1N HCl in anhydrous isopropanol) added to dissolve the formazan
- Absorbance was measured at 570 nm using microplate reader

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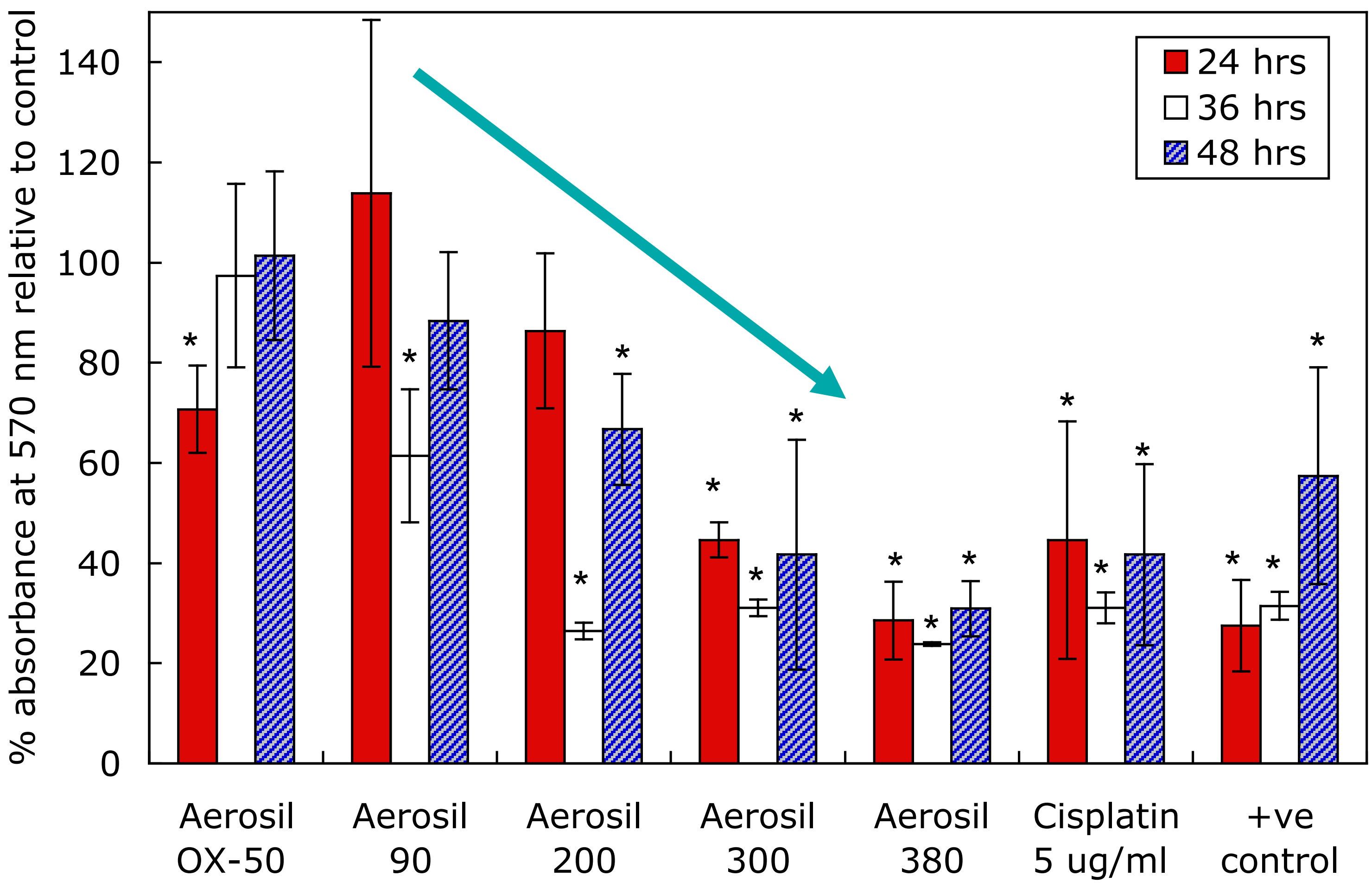
MTT results, silica/cisplatin



Error bars represent standard deviation of four replicates
Asterisks denote statistically significant ($p < 0.05$) differences between experiment and control

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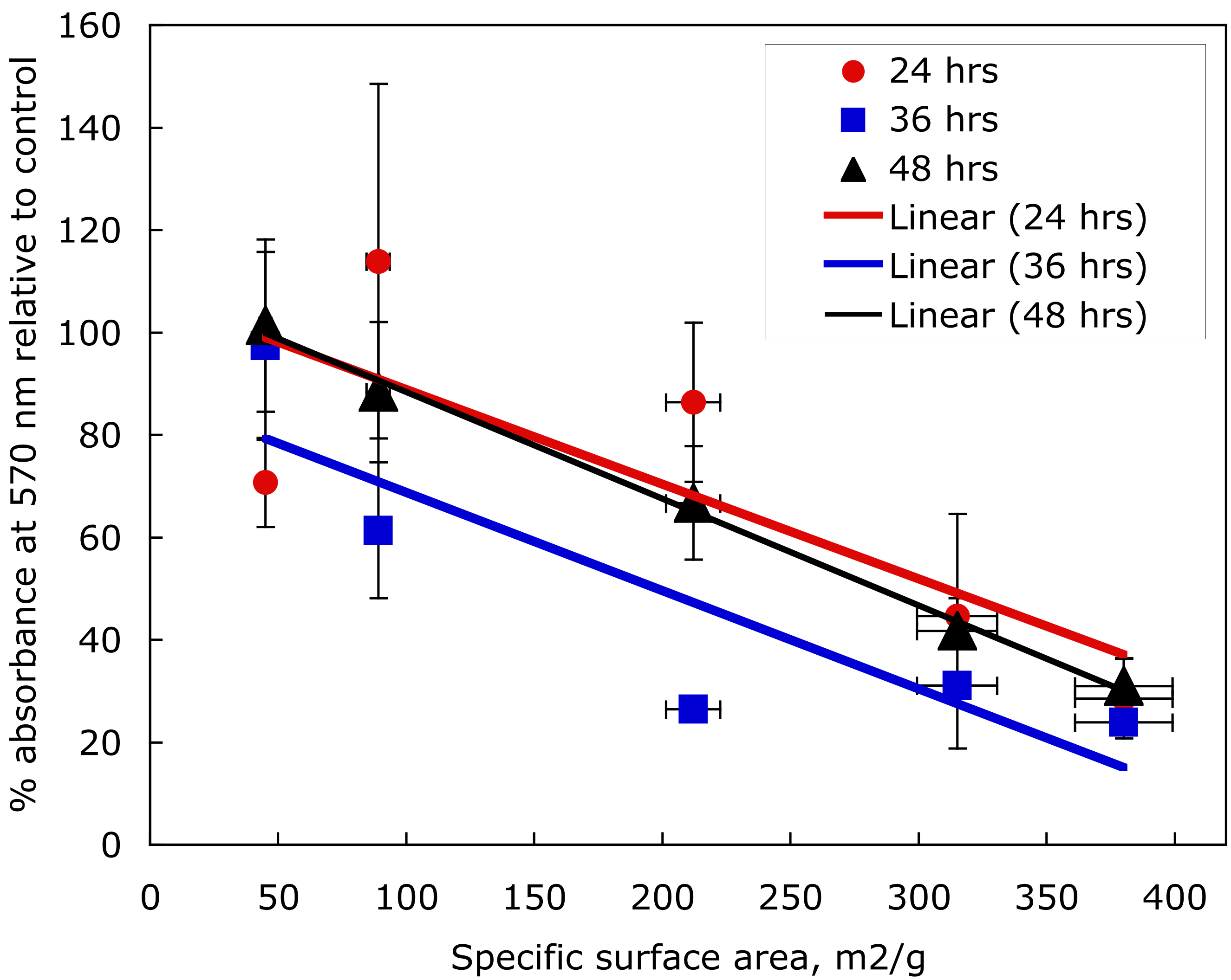
Correlation



R^2 = 0.61
R^2 = 0.76
R^2 = 1.0

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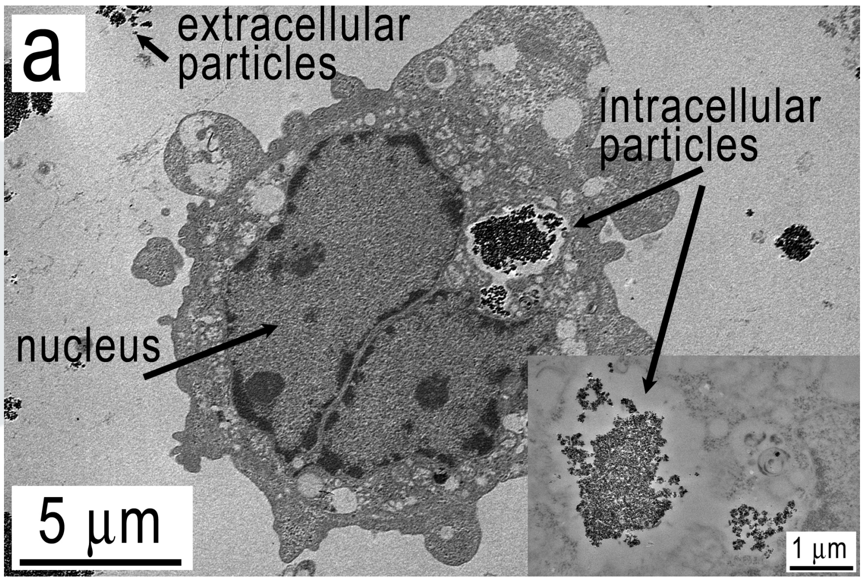
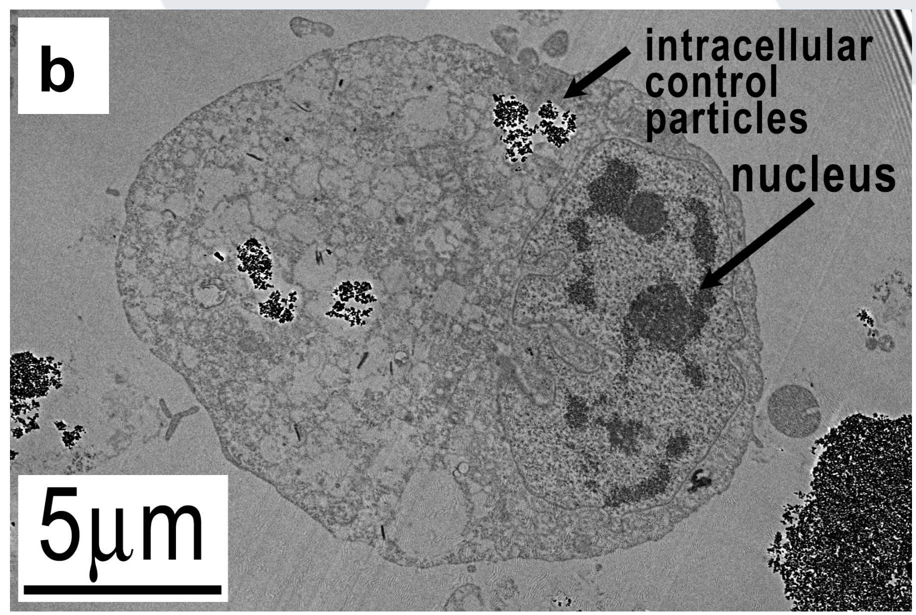
Transmission electron microscopy comparison to control…





Qualitative observations
- intracellular particles observed in both
- nuclei more compact in control sample

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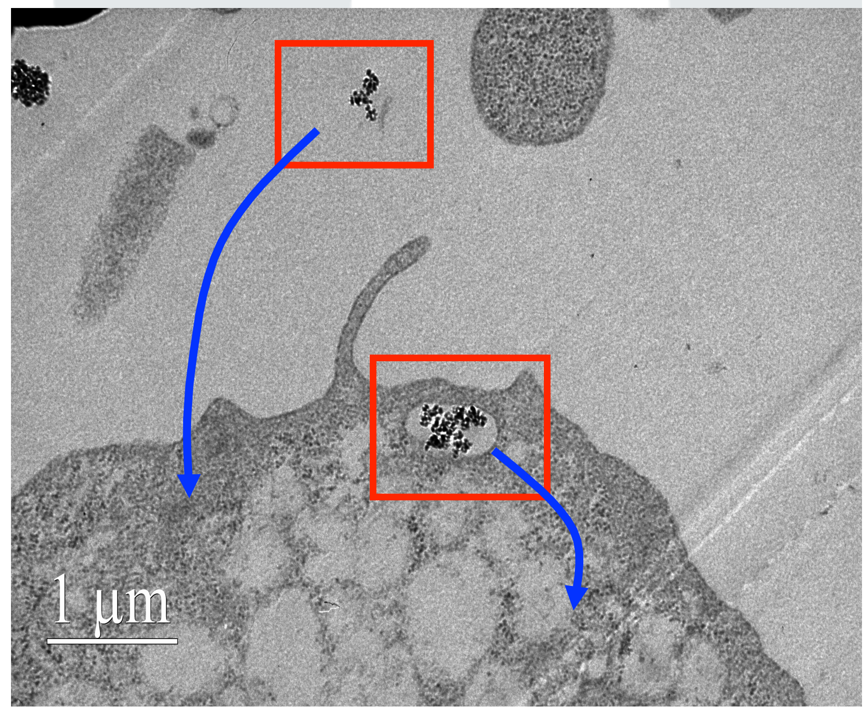
Discussion

- Free cisplatin can enter cells by active and passive transport
- Adverse effects on cell function may result from
  - cells internalizing particles/cisplatin (true trojan horse)
  - particles releasing cisplatin near the cell membranes



Discussion

- Metabolic function correlated negatively with surface area
- Fluorescence imaging indicates significant adverse effects on cell membranes and DNA
- Effects comparable to free cisplatin at 5 μg/ml
- No synergistic adverse effects observed, nor did silica mitigate effects of cisplatin on cells
- TEM images show the particles are inside the cells
- Adverse effects on cell function may result from transport of cisplatin near cells or directly into cells by particles

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Take home message

- May not be enough to consider nanoparticles by themselves when examining effects of engineered nanoparticles on biological systems
- Must evaluate potential for co-release with other compounds, reactive or condensable, that may be hazardous
- Must evaluate fate of nanoparticles in the complex environment after release

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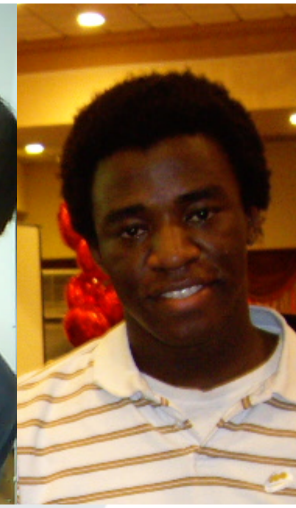

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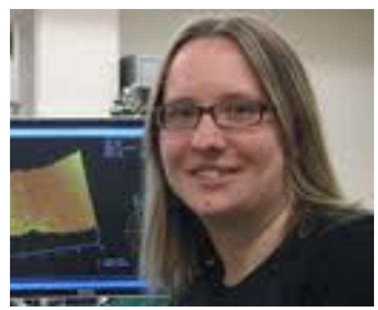

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