Adaptation and Application of Cheminformatics Methods in Toxicity Assessment of Nanomaterials

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Kemijski inštitut / National Institute of Chemistry, Ljubljana, Slovenia, October 19, 2017

North Dakota

North Dakota

North Dakota

182,000 km^2

North Dakota

North Dakota

North Dakota

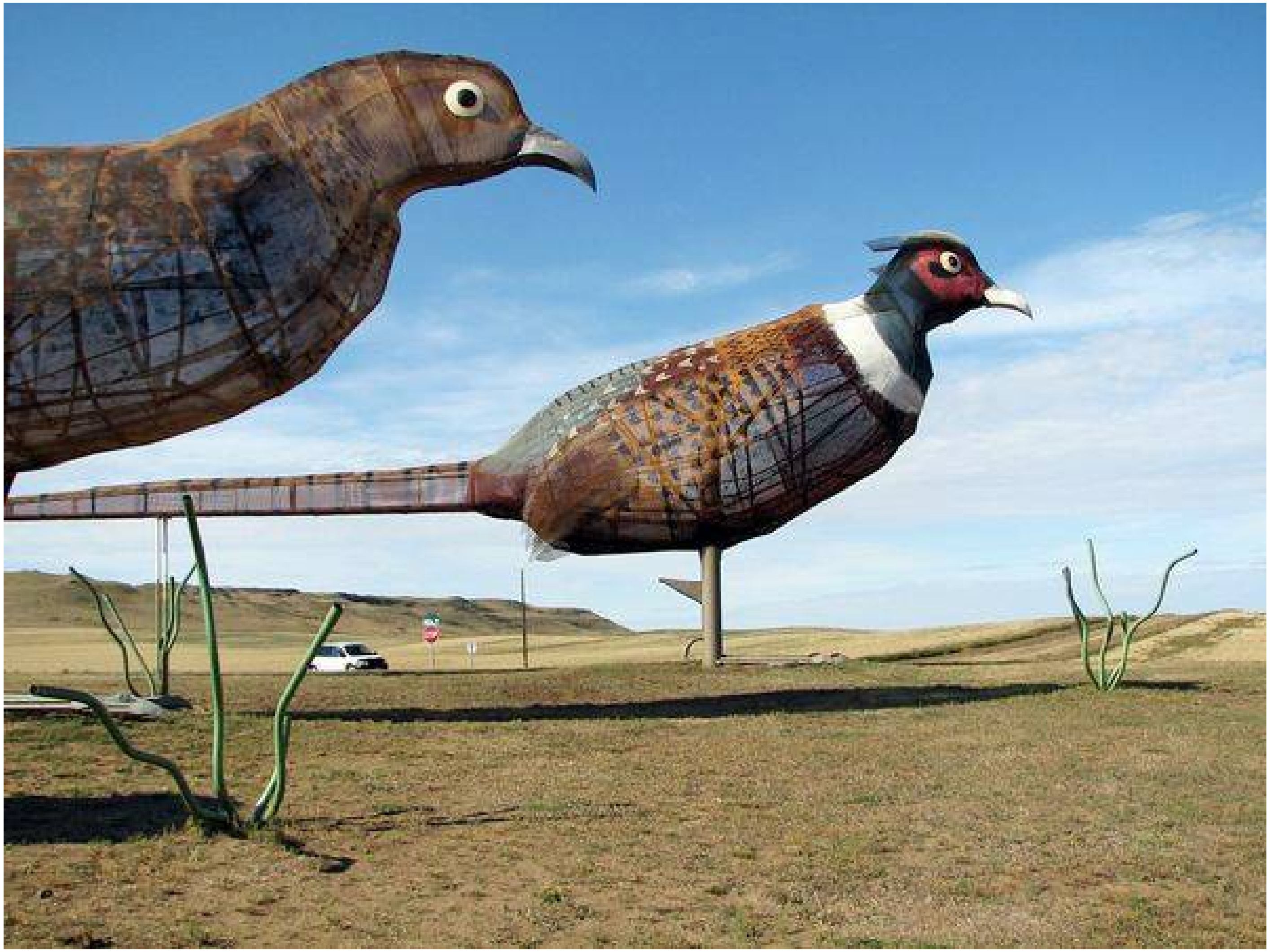
North Dakota


WORLD'S
LARGEST
BUFFALO
1959

North Dakota

North Dakota

North Dakota



Seven sensational scrap metal sculptures line the 32-mile stretch of highway in southwest North Dakota, including artist Gary Greff’s massive “Geese in Flight,” listed in the Guinness World Records as the world’s largest scrap metal sculpture.

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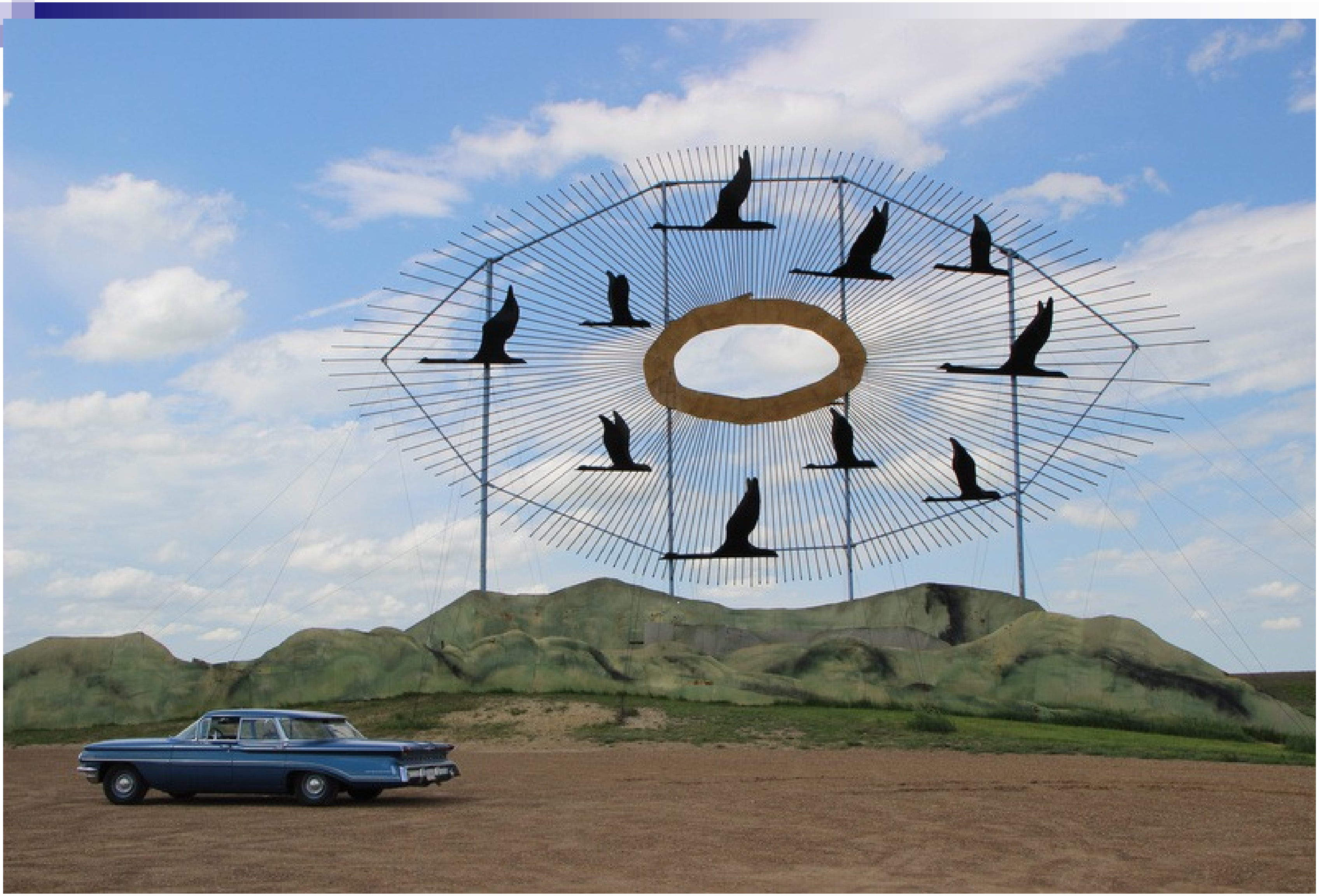

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North Dakota

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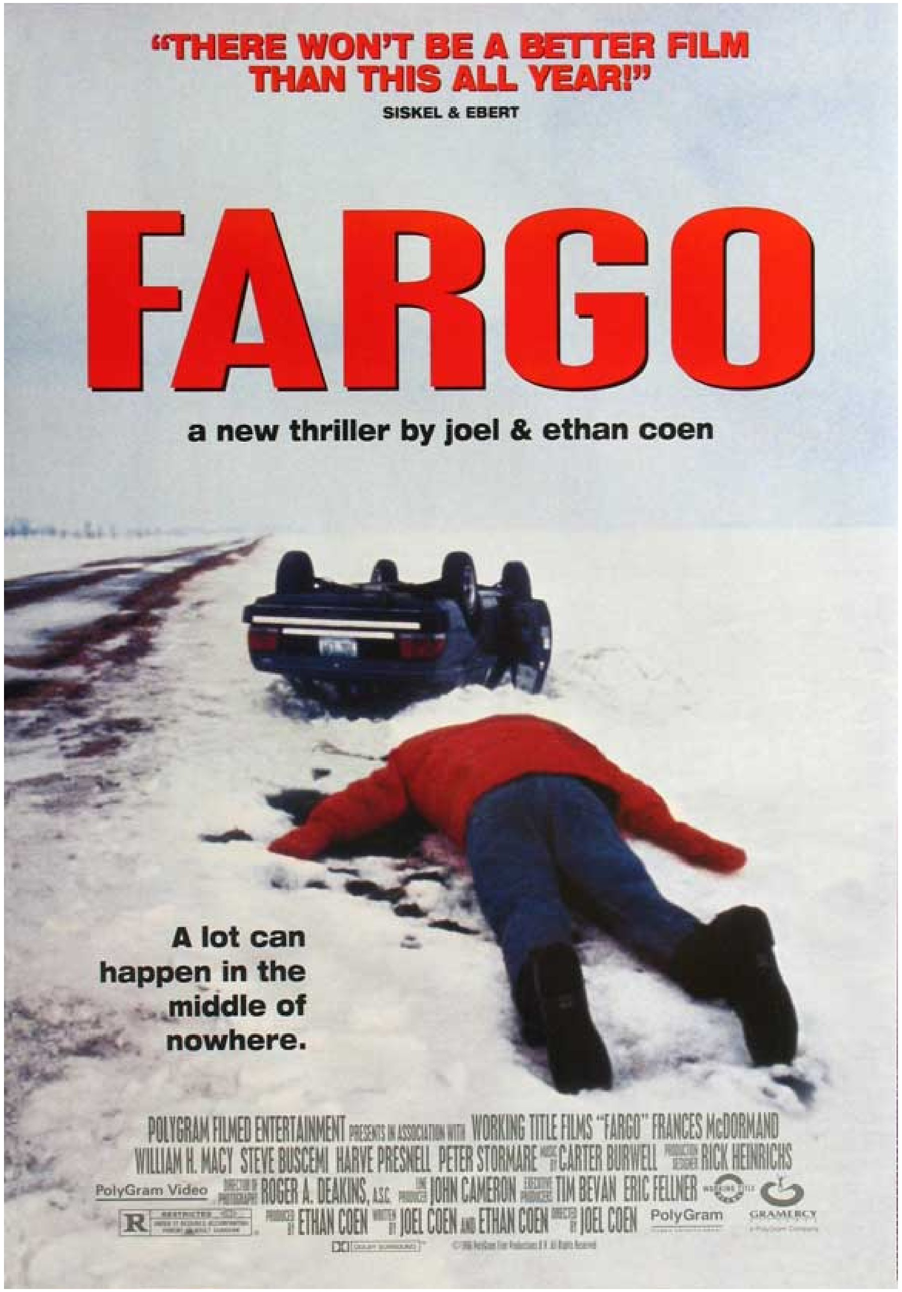
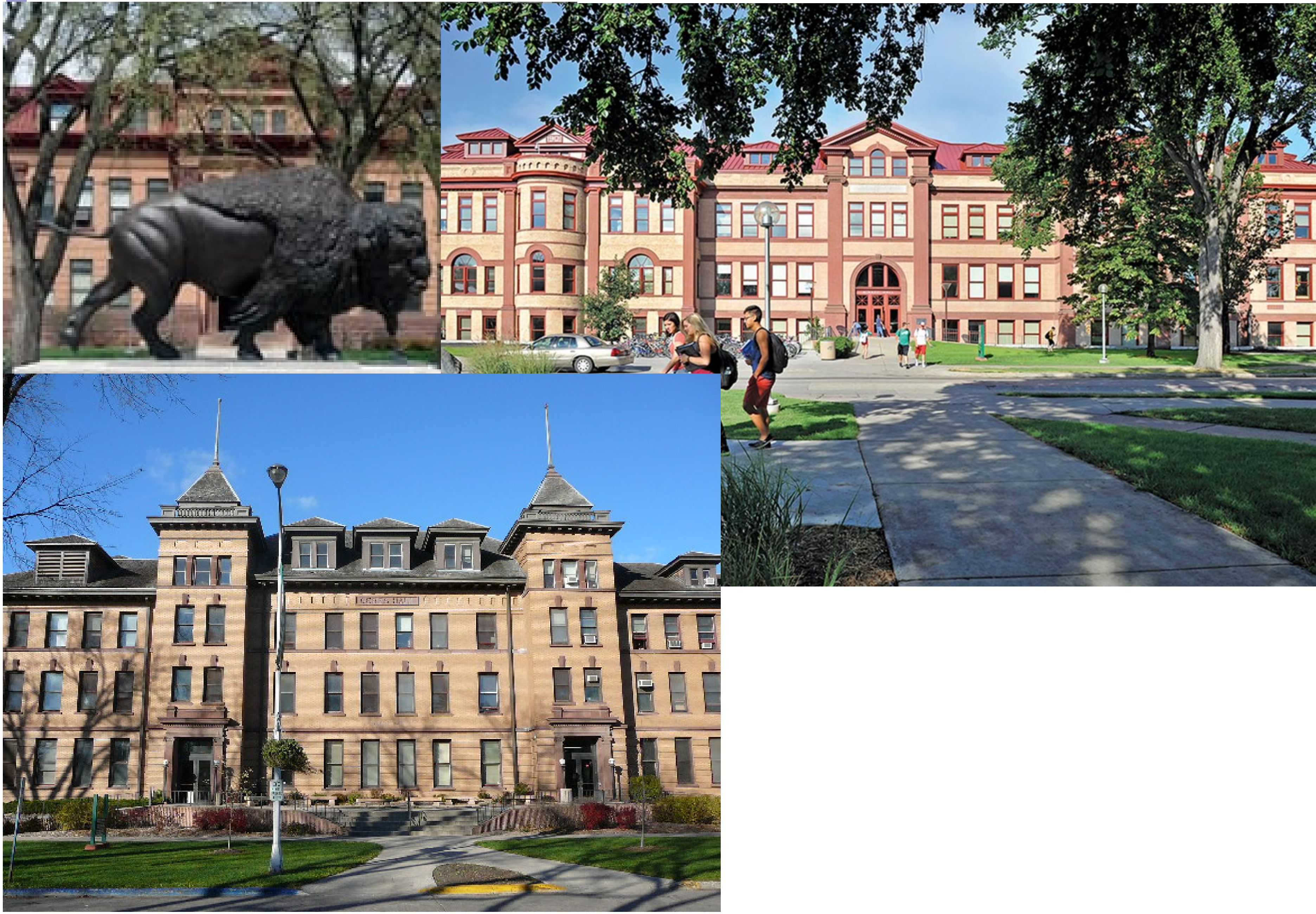
OF THE WEST
FARGO
White
UNIFORMS
4TH Ave N
THE 400


"THERE WON'T BE A BETTER FILM THAN THIS ALL YEAR!"
SISKEL & EBERT
FARGO
a new thriller by joel & ethan coen
A lot can happen in the middle of nowhere.
POLYGRAM FILMED ENTERTAINMENT PRESENTS IN ASSOCIATION WITH WORKING TITLE FILMS "FARGO" FRANCES McDORMAND
WILLIAM H. MACY STEVE BUSCEMI HARVE PRESNELL PETER STORMARE MUSIC BY CARTER BURWELL PRODUCTION DESIGNER RICK HEINRICHS
PolyGram Video
DIRECTOR OF PHOTOGRAPHY ROGER A. DEAKINS, A.S.C. LINE PRODUCER JOHN CAMERON EXECUTIVE PRODUCERS TIM BEVAN ERIC FELLNER
R
PRODUCED BY ETHAN COEN WRITTEN BY JOEL COEN AND ETHAN COEN DIRECTED BY JOEL COEN
PolyGram
GRAMERCY

FARGO


NORTH DAKOTA
STATE UNIVERSITY


NORTH DAKOTA
STATE UNIVERSITY



Department of Coatings and Polymeric Materials

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Polymers

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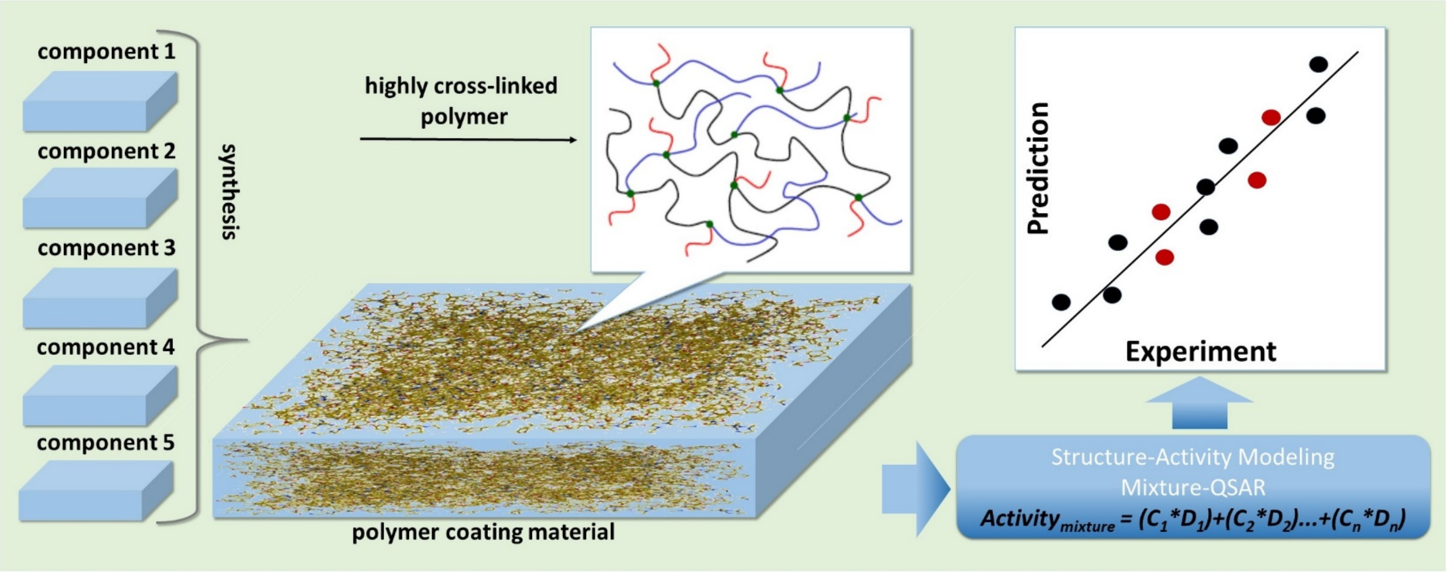
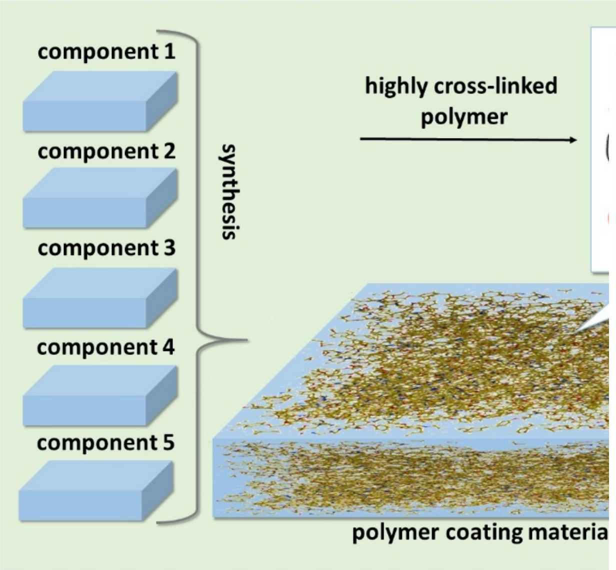
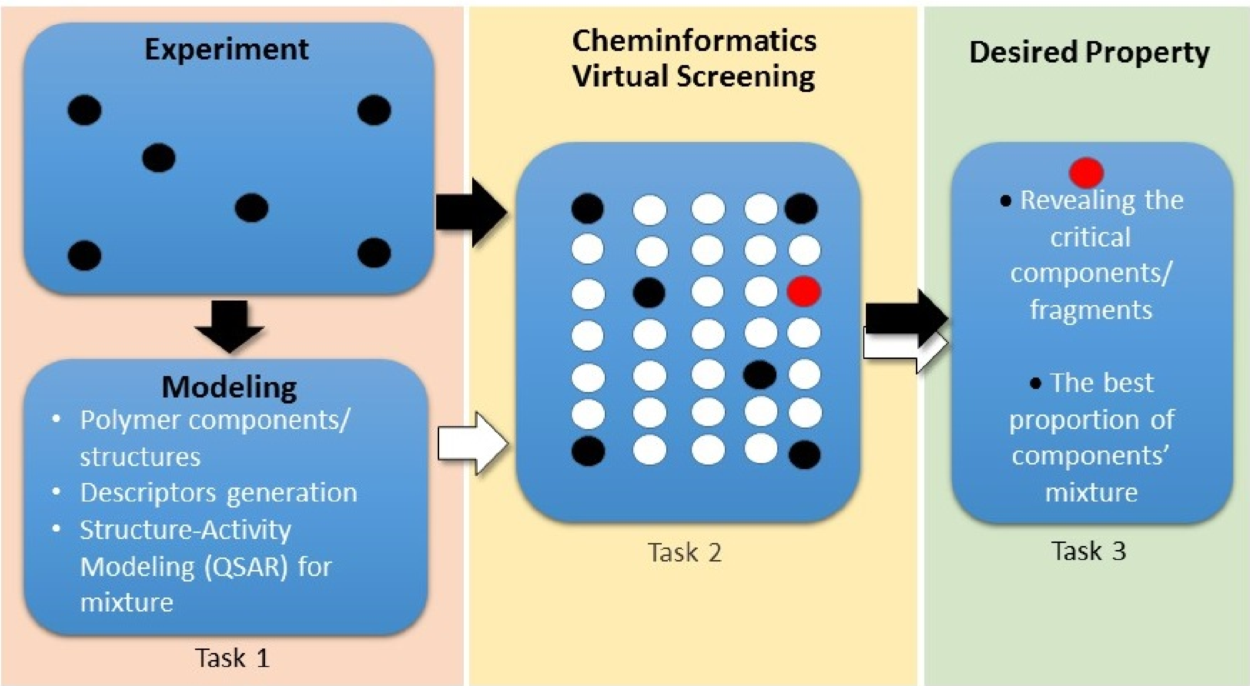


Polymers

Department of Coatings and Polymeric Materials



Polymers



Experiment

Modeling

- Polymer components/ structures
- Descriptors generation
- Structure-Activity Modeling (QSAR) for mixture

Task 1

Cheminformatics Virtual Screening

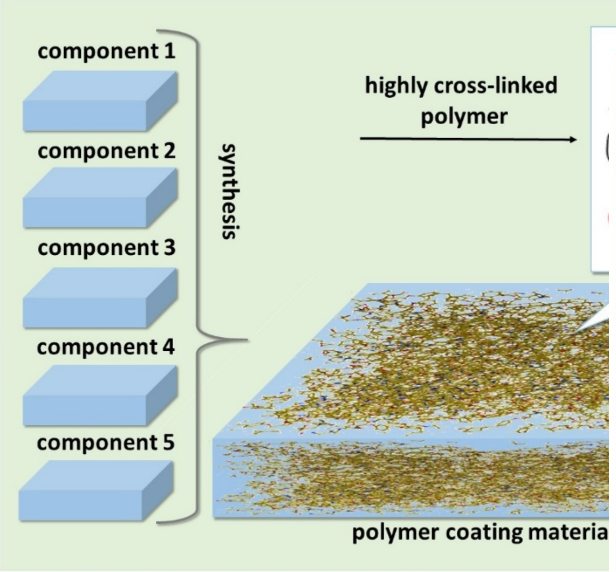
Task 2

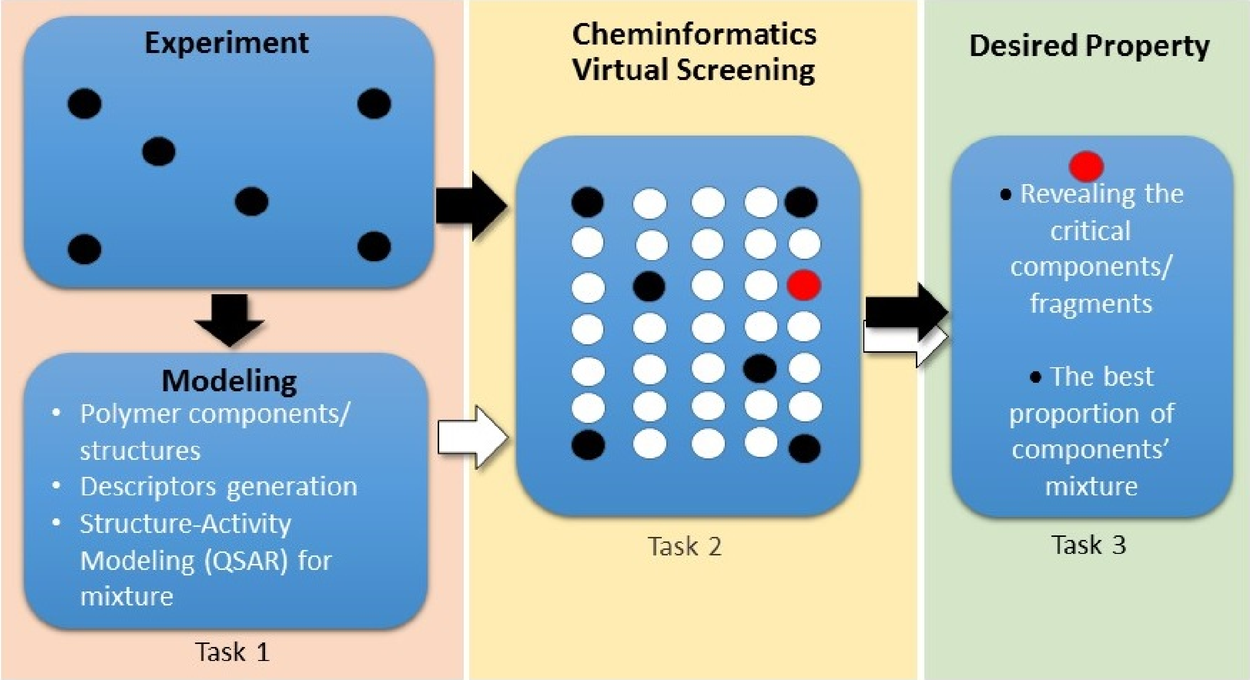
Desired Property

- Revealing the critical components/ fragments
- The best proportion of components' mixture

Task 3

Department of Coatings and Polymeric Materials

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Polymers

Nanomaterials

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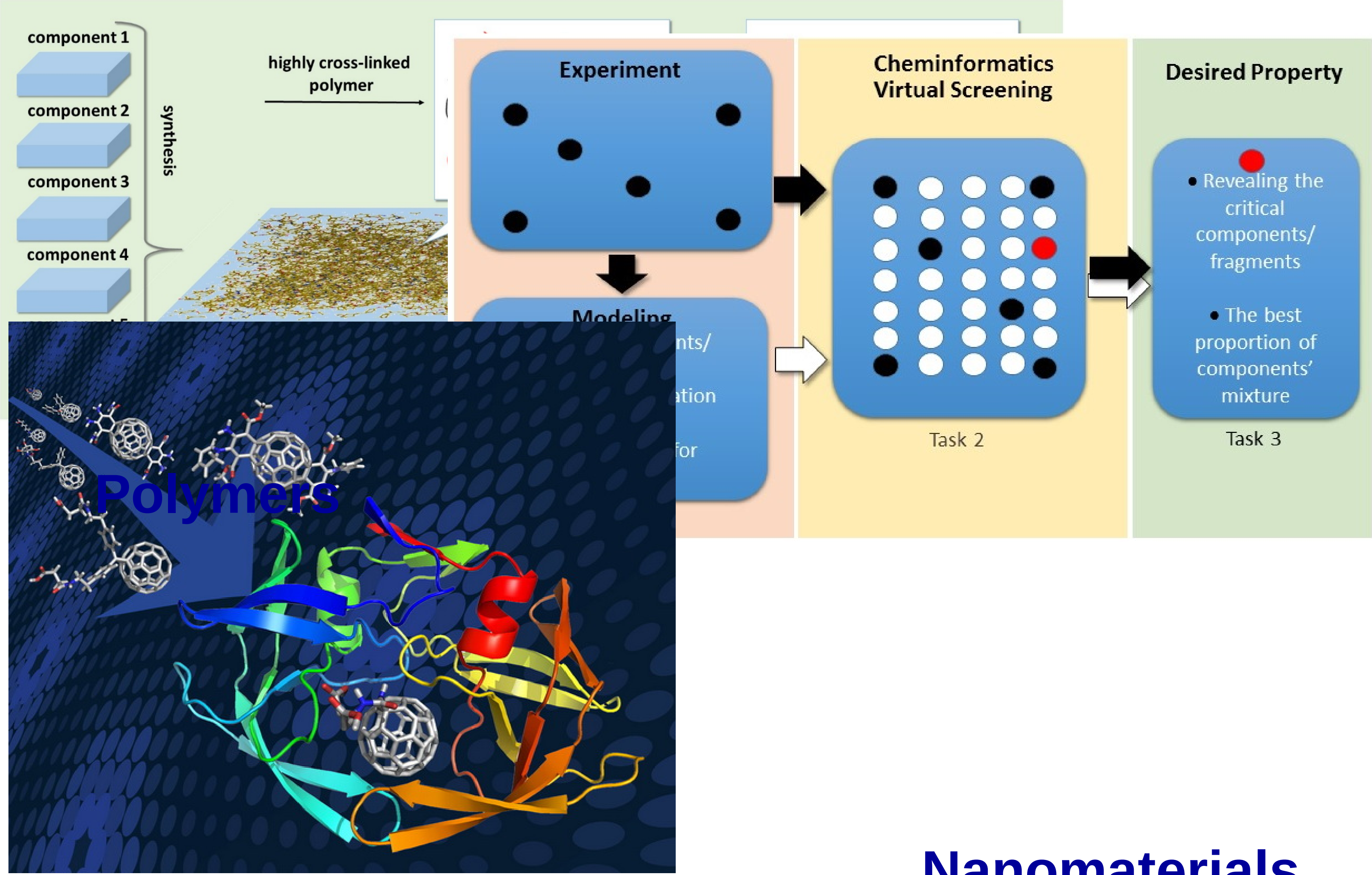
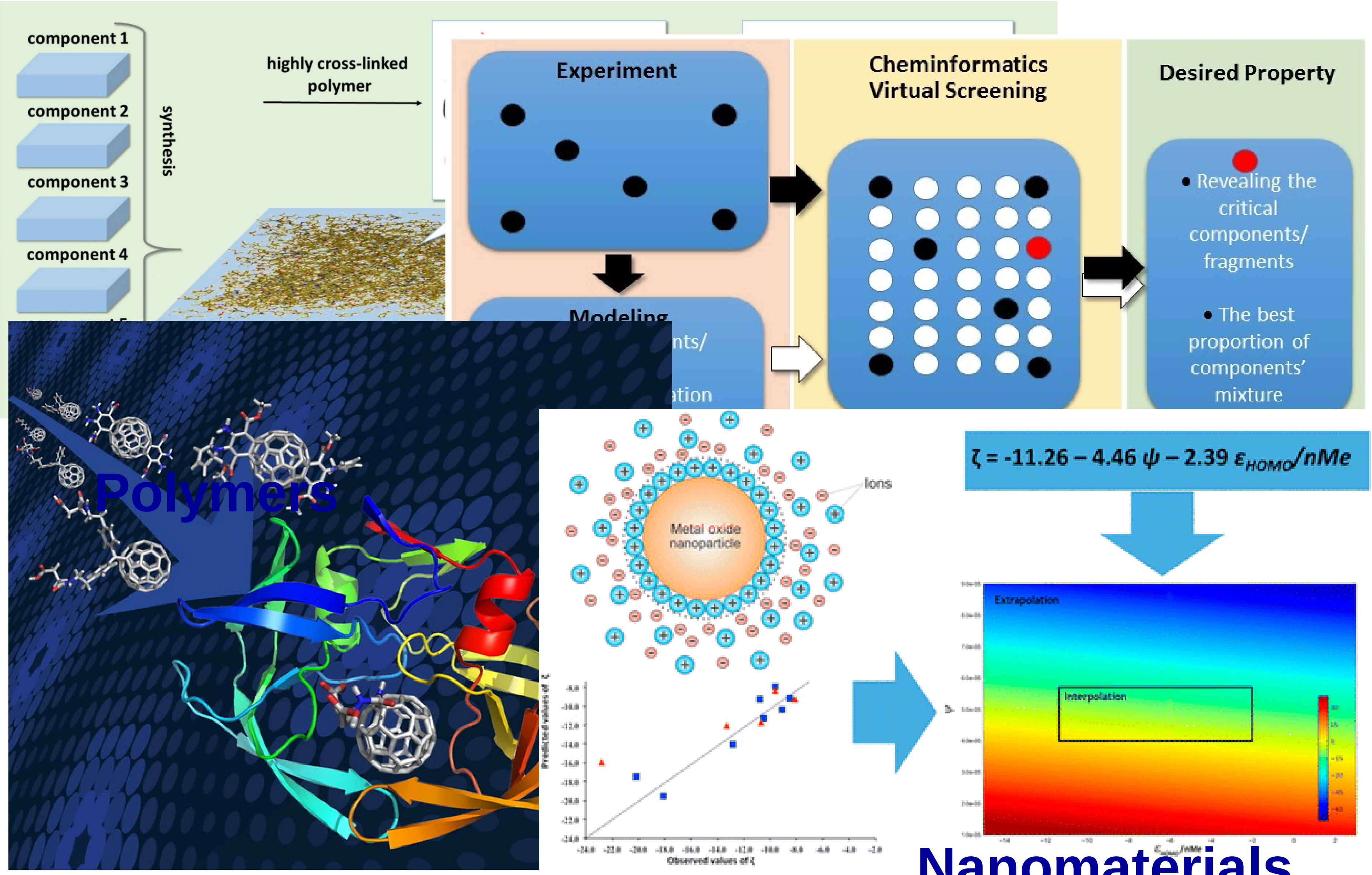


Polymers

Nanomaterials

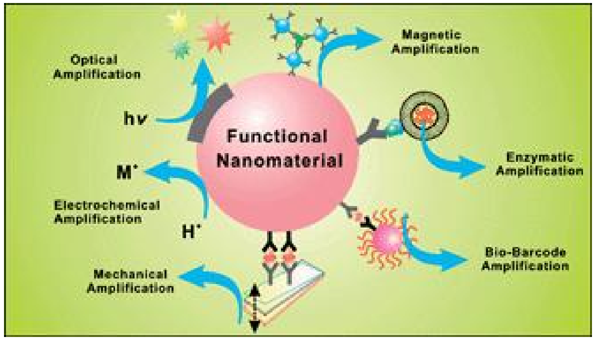
Department of Coatings and Polymeric Materials

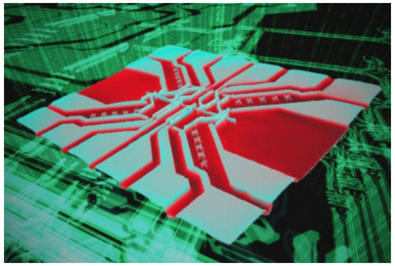


Polymers

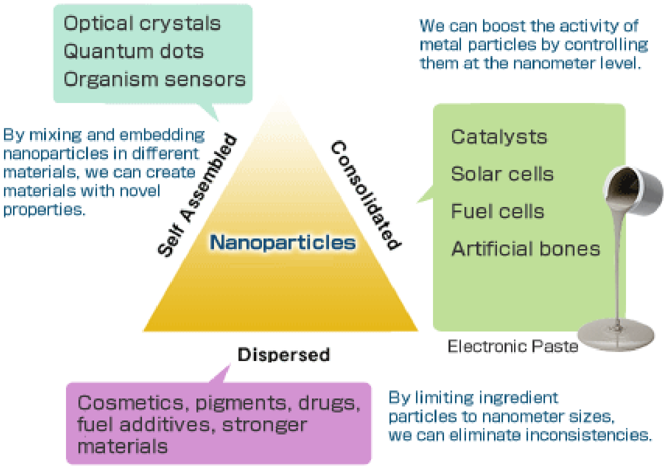
Nanomaterials

Nanomaterials Applications





■Examples of Nanomaterial Applications



Optical crystals
Quantum dots
Organism sensors

We can boost the activity of metal particles by controlling them at the nanometer level.

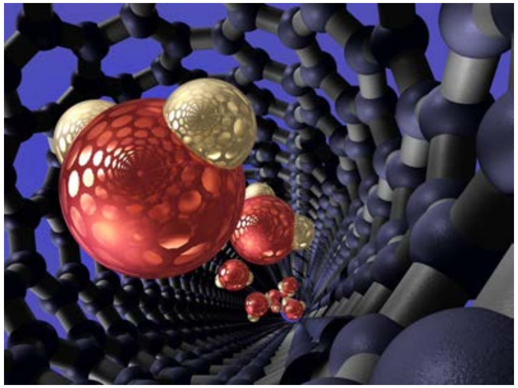
By mixing and embedding nanoparticles in different materials, we can create materials with novel properties.

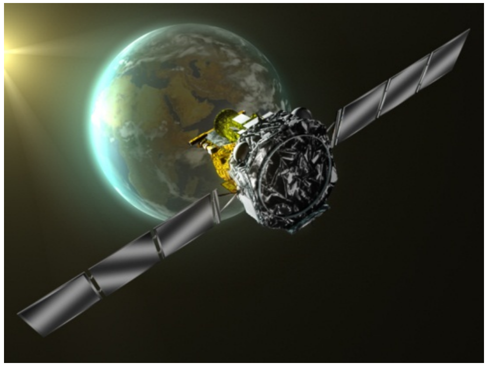
Catalysts
Solar cells
Fuel cells
Artificial bones

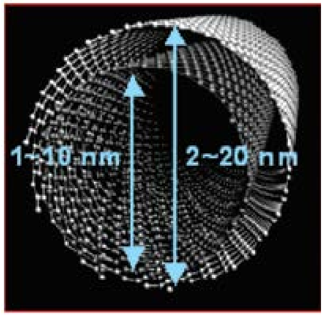
Electronic Paste

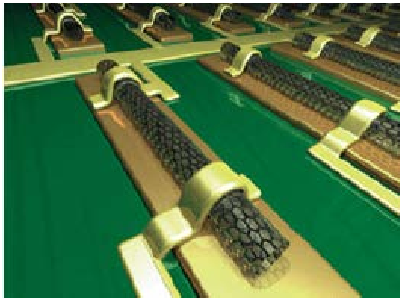
Cosmetics, pigments, drugs, fuel additives, stronger materials

By limiting ingredient particles to nanometer sizes, we can eliminate inconsistencies.

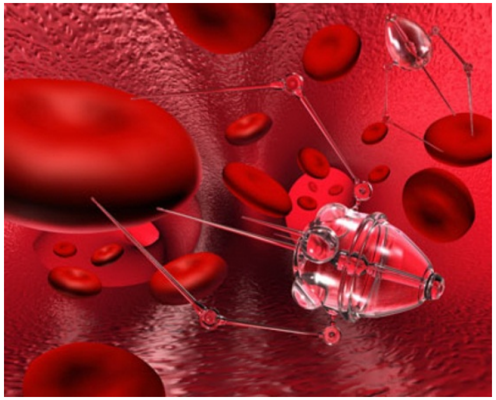




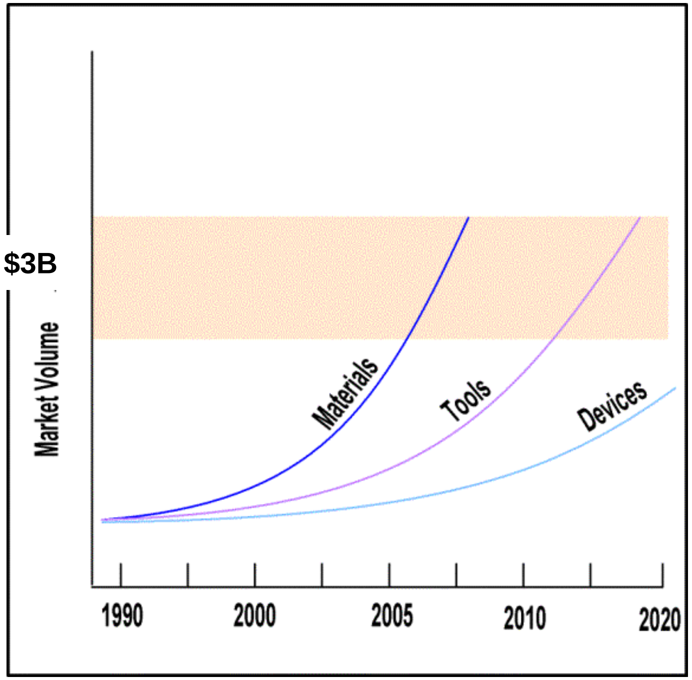


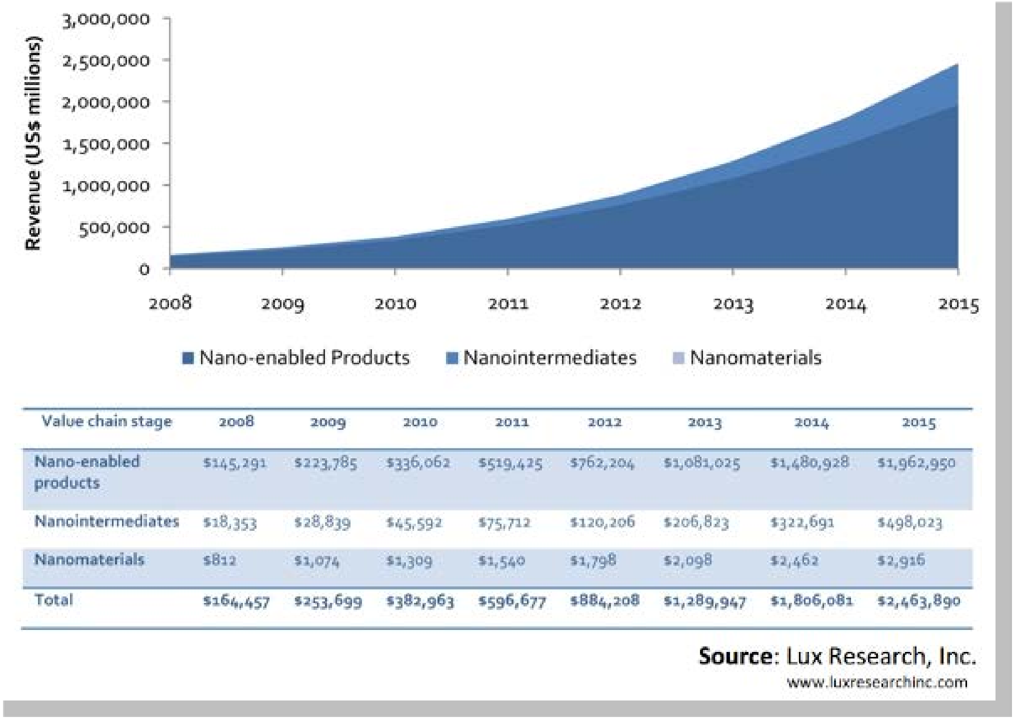


Single molecular transistors



Nanomaterials in Consumer Products





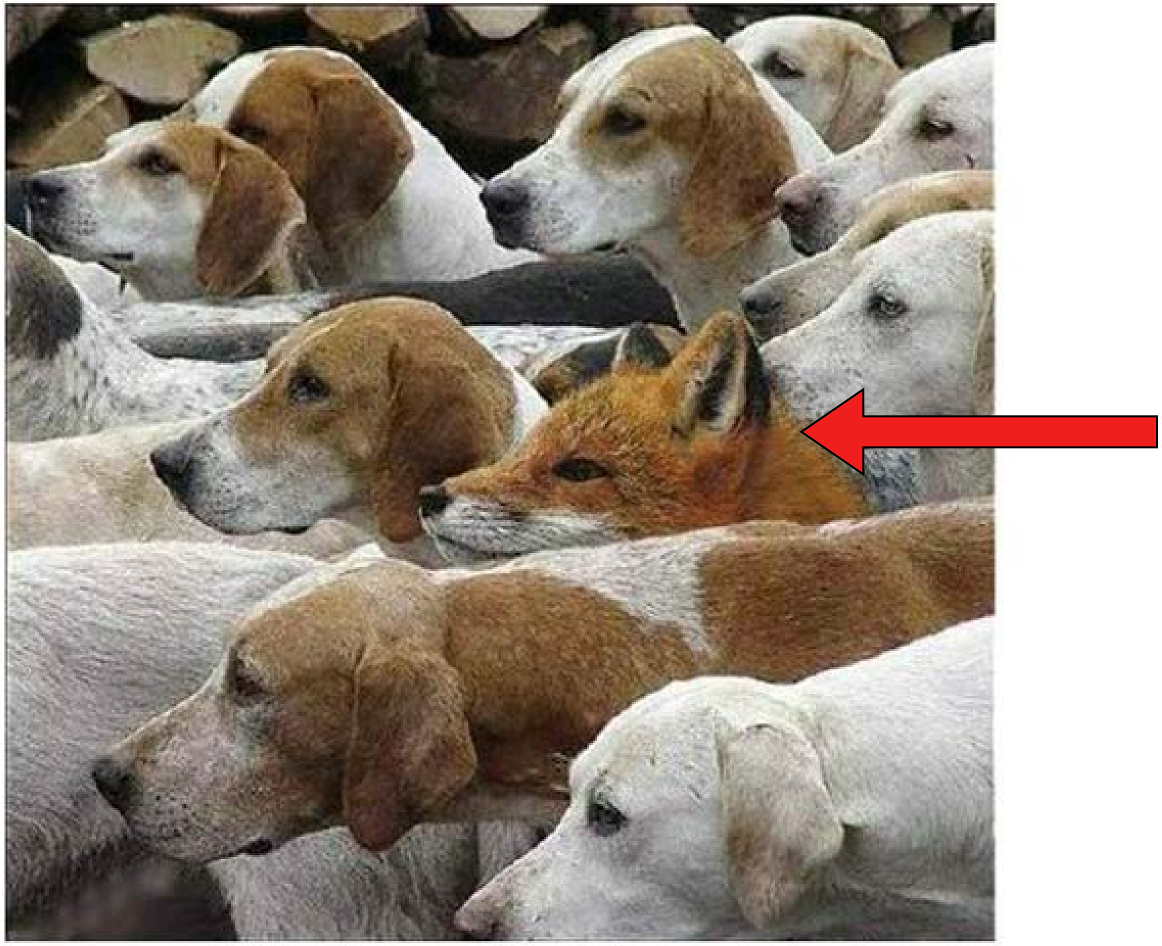
Value chain stage	2008	2009	2010	2011	2012	2013	2014	2015
Nano-enabled products	$145,291	$223,785	$336,062	$519,425	$762,204	$1,081,025	$1,480,928	$1,962,950
Nanointermediates	$18,353	$28,839	$45,592	$75,712	$120,206	$206,823	$322,691	$498,023
Nanomaterials	$812	$1,074	$1,309	$1,540	$1,798	$2,098	$2,462	$2,916
Total	$164,457	$253,699	$382,963	$596,677	$884,208	$1,289,947	$1,806,081	$2,463,890

Source: Lux Research, Inc.
www.luxresearchinc.com

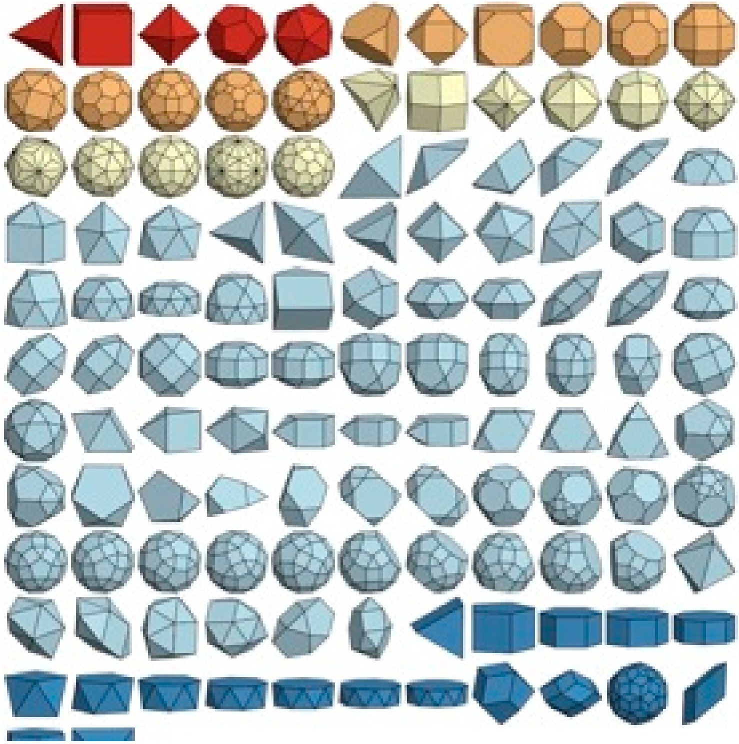
- Lux Research *Nanotechnology Report*: Projections of $4.4 trillion in global manufactured nano-products by 2018
- Wilson Center’s *Project on Emerging Nanotechnologies* 2015 Consumer Product Database: over 1600+ self-identified nano-products now on U.S. market shelves.
- Products include paints, coatings, sporting goods, sunscreens, cosmetics, personal care products, stain-resistant clothing, and light emitting diodes used in computers, cell phones, and digital cameras.

All nanomaterials are not the same

All nanomaterials are not the same

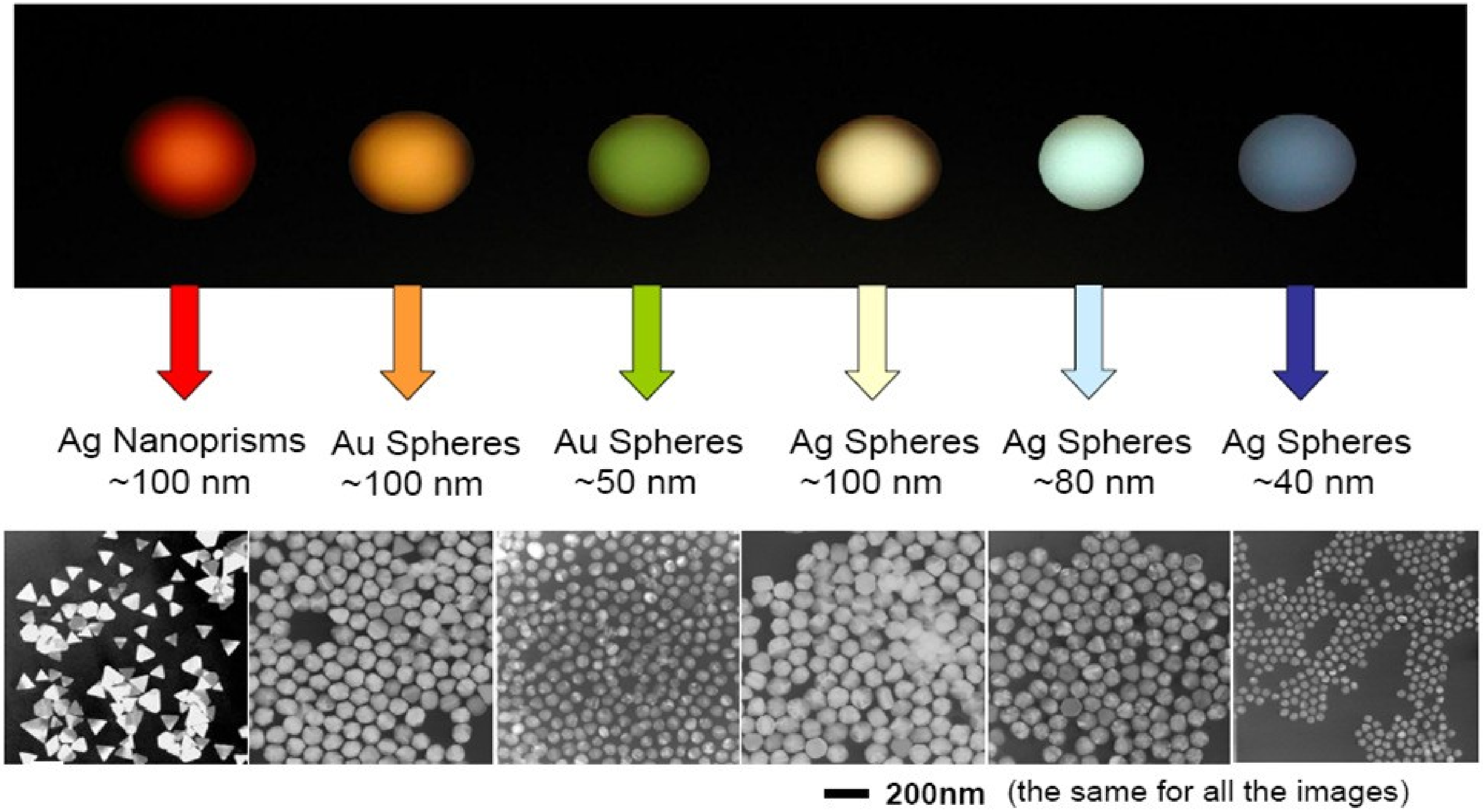


Same structure – different shapes



The properties can vary with size

Jin et al. (2001) Science 294: 1901-1903



Properties of nanoparticles

The biological activity of nanoparticles, including toxic and other environmental effects are complex phenomena, which involve the physicochemical properties based on molecular structure, atomic composition of a molecule, in addition to unusual size and surface effects.

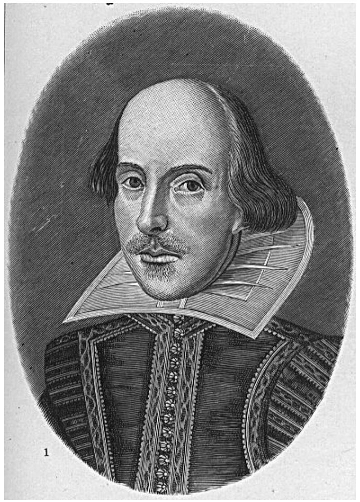
The following physicochemical properties of nanoparticles are determine the behavior in environment[a]:

- **CHEMICAL COMPOSITION** (atom composition)
- **SMALL SIZE** (particle size, size distribution)
- **LARGE SURFACE** (surface reactivity, surface coatings, surface groups)
- **CRYSTAL STRUCTURE** (crystallinity)
- **SOLUBILITY** (solubility in the relevant media)
- **SHAPE**
- **AGGREGATION** (aggregation status in the relevant media)
- **PURITY** (purity of sample)

In addition, the method of synthesis and/or preparation including postsynthetic modifications also plays important role in nanoparticle behavior.

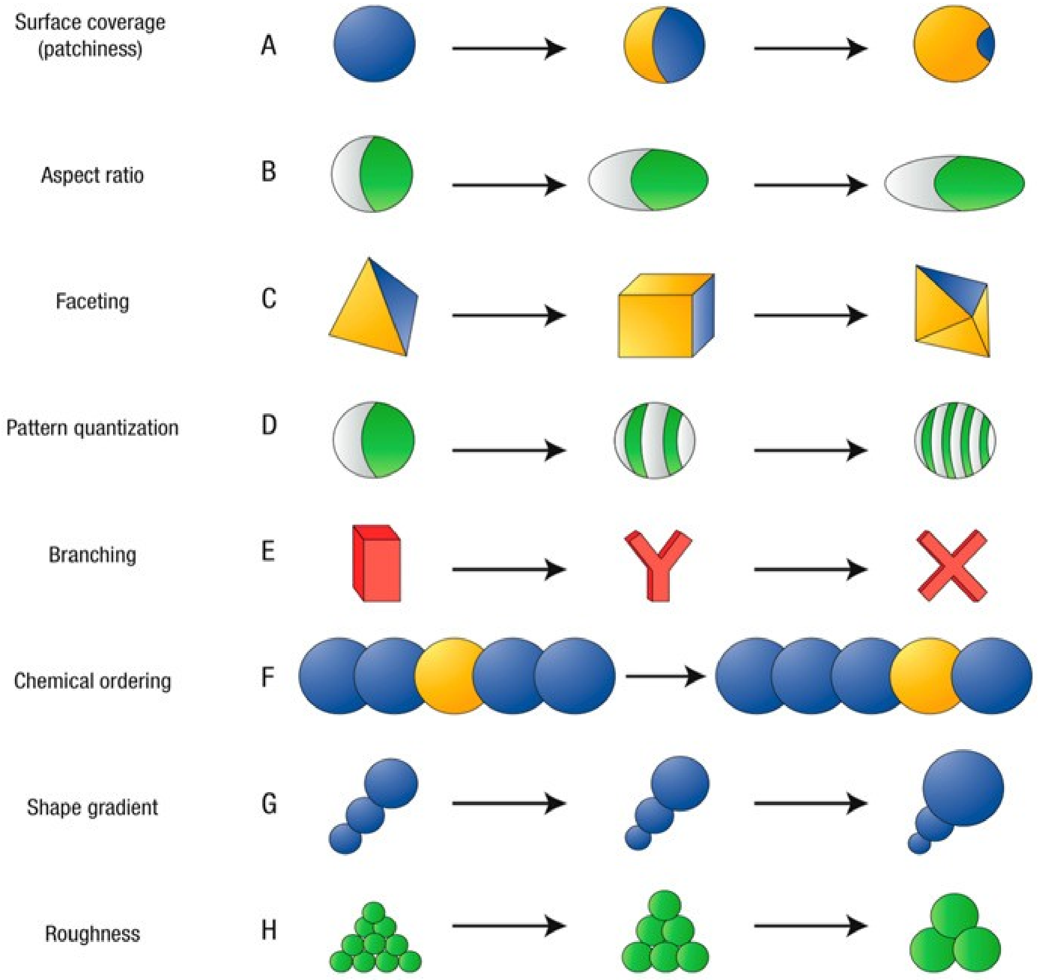
[a] Warheit D.B., *Toxicological Sciences*, 2008, 101(2), 183-185

The importance of characterization



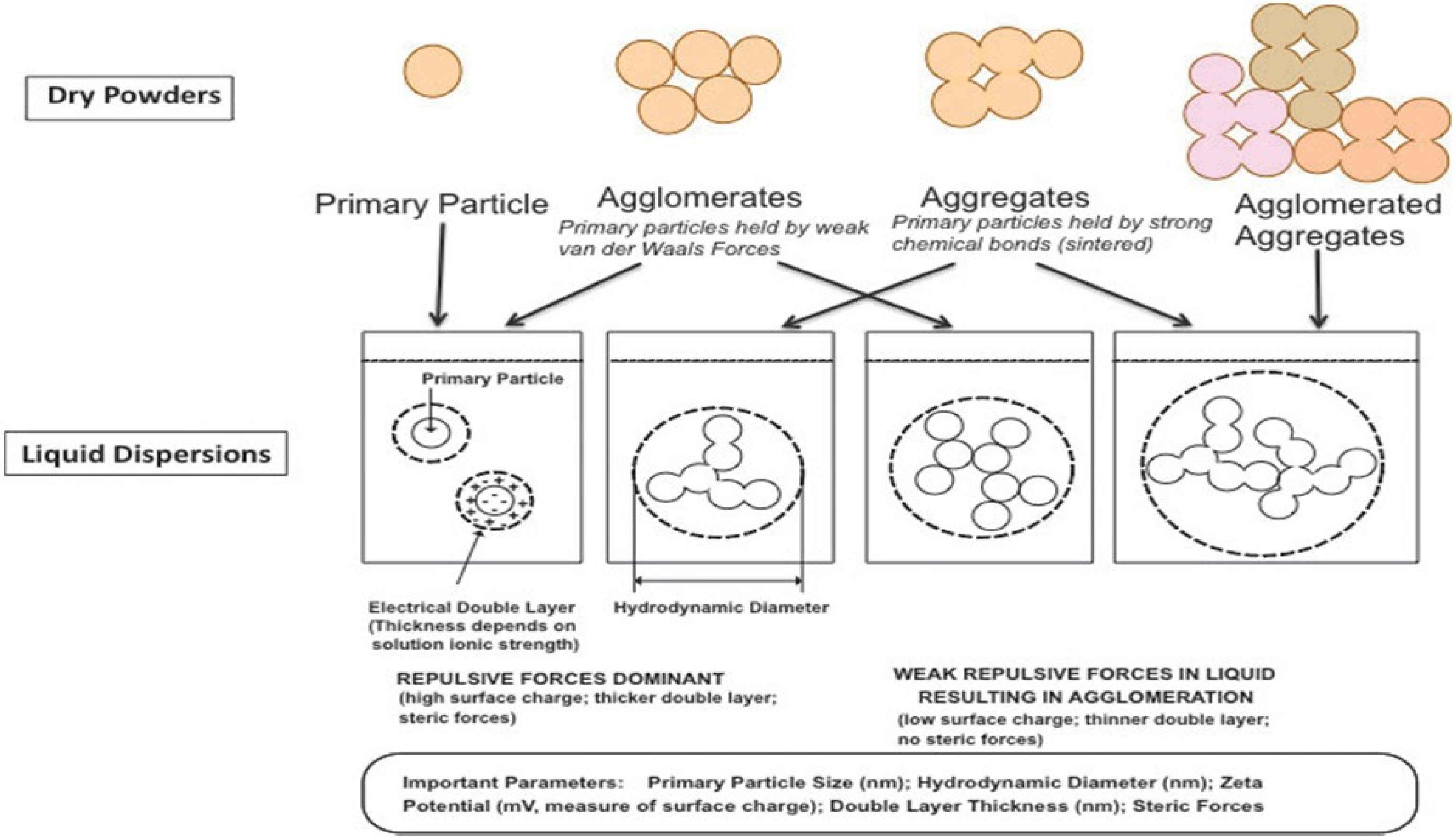
“Orthogonal dimensions” for nanoparticles

Nature Materials, 2007, 6: 557-562



Agglomeration and aggregation of nanoparticles

Jiang (2009) J. Nanopart. Res. 11: 77-89.



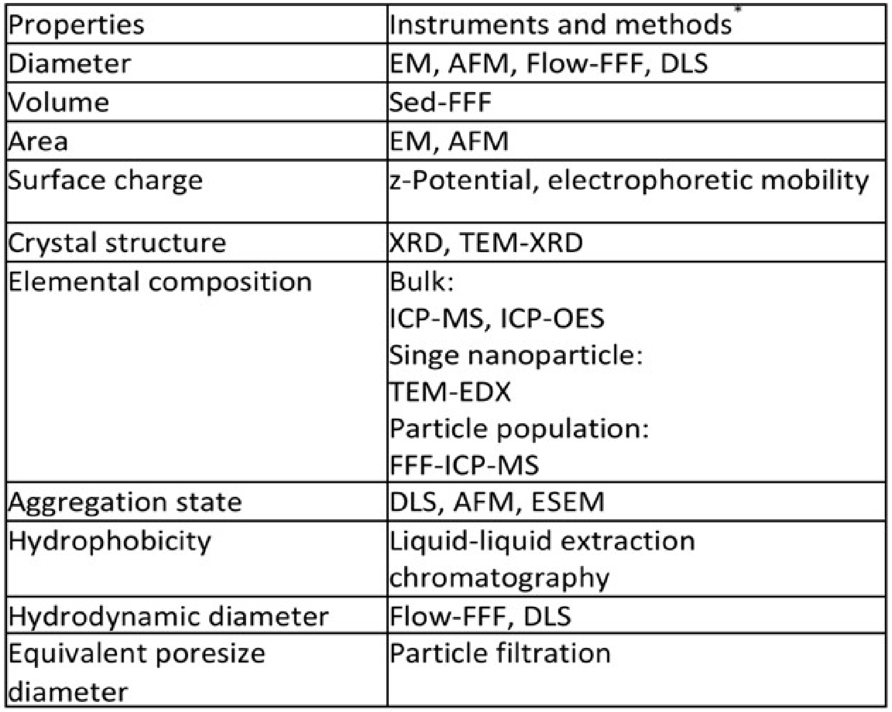
REPULSIVE FORCES DOMINANT
(high surface charge; thicker double layer; steric forces)

WEAK REPULSIVE FORCES IN LIQUID RESULTING IN AGGLOMERATION
(low surface charge; thinner double layer; no steric forces)

Important Parameters: Primary Particle Size (nm); Hydrodynamic Diameter (nm); Zeta Potential (mV, measure of surface charge); Double Layer Thickness (nm); Steric Forces

Experimental techniques that can help to get nano-properties (nano-descriptors)

Haselov et al. (2008) Ecotoxicology 17: 344-361



Properties	Instruments and methods*
Diameter	EM, AFM, Flow-FFF, DLS
Volume	Sed-FFF
Area	EM, AFM
Surface charge	z-Potential, electrophoretic mobility
Crystal structure	XRD, TEM-XRD
Elemental composition	Bulk: ICP-MS, ICP-OES Singe nanoparticle: TEM-EDX Particle population: FFF-ICP-MS
Aggregation state	DLS, AFM, ESEM
Hydrophobicity	Liquid-liquid extraction chromatography
Hydrodynamic diameter	Flow-FFF, DLS
Equivalent poresize diameter	Particle filtration

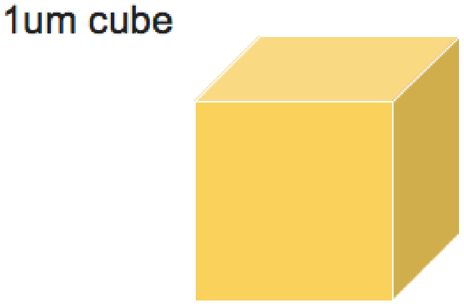
Abbreviations:

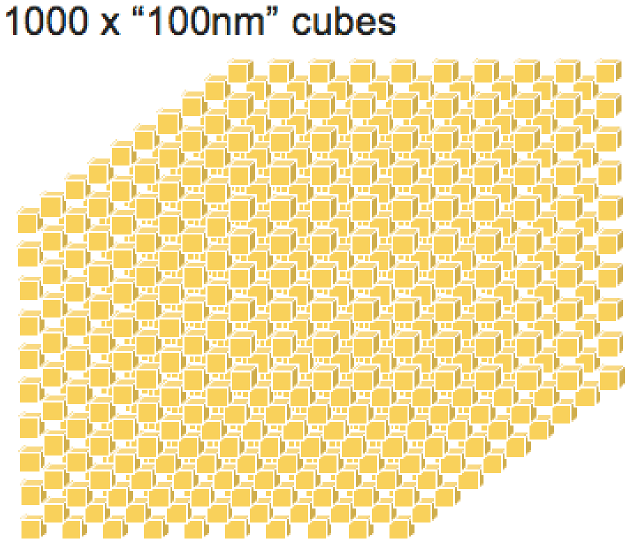
- EM- electronic microscopy,
- AFM - atomic force microscopy,
- FFF- field flow filtration,
- DLS - dynamic light scattering,
- LC- liquid chromatography,
- XRD - X-ray diffraction,
- TEM - transmission electron microscopy,
- ICP-MS - inductively coupled plasma mass spectrometry,
- ICP-OES - inductively coupled plasma emission spectroscopy,
- EDX - energy dispersive X-ray spectrometry,
- ESEM - environmental scanning electron microscopy.

Mass-based “dose” may be inadequate

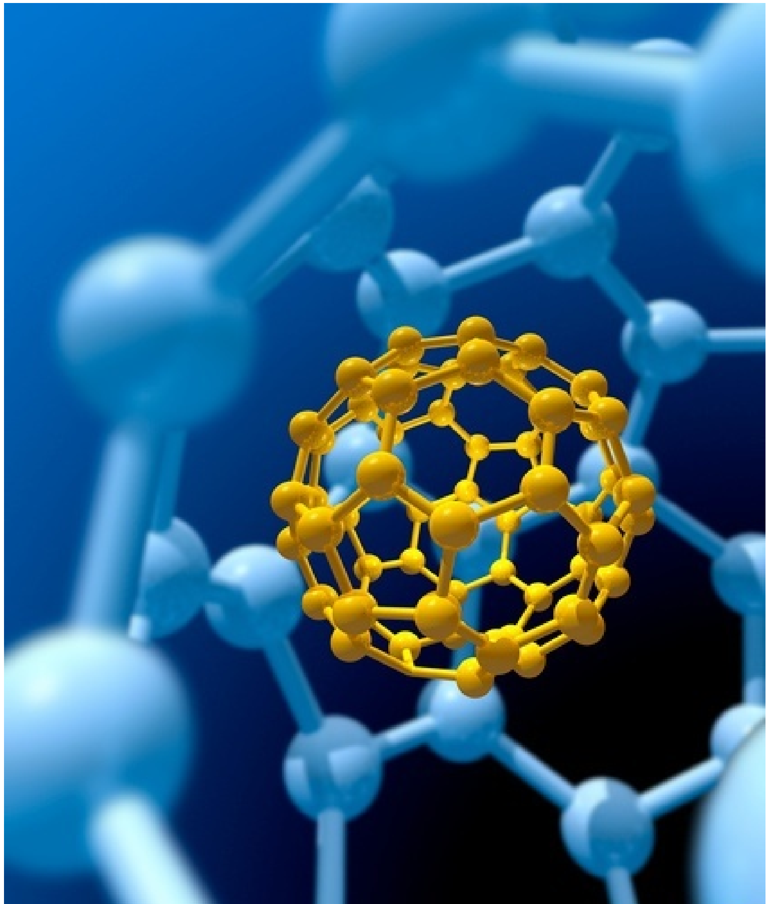
Effects may be related to surface area based “dose”

- 1um cube
  - e.g. respirable particle
  - Surface area = 6um^2
- 100nm cube
  - 1000 cubes is equivalent volume
  - Surface area = 60 um^2
- 10x more surface area for the same mass





Can we predict properties of nanomaterials?



- Physical properties of nanomaterials
- Toxicological aspects of nanomaterials
- Pharmacological properties

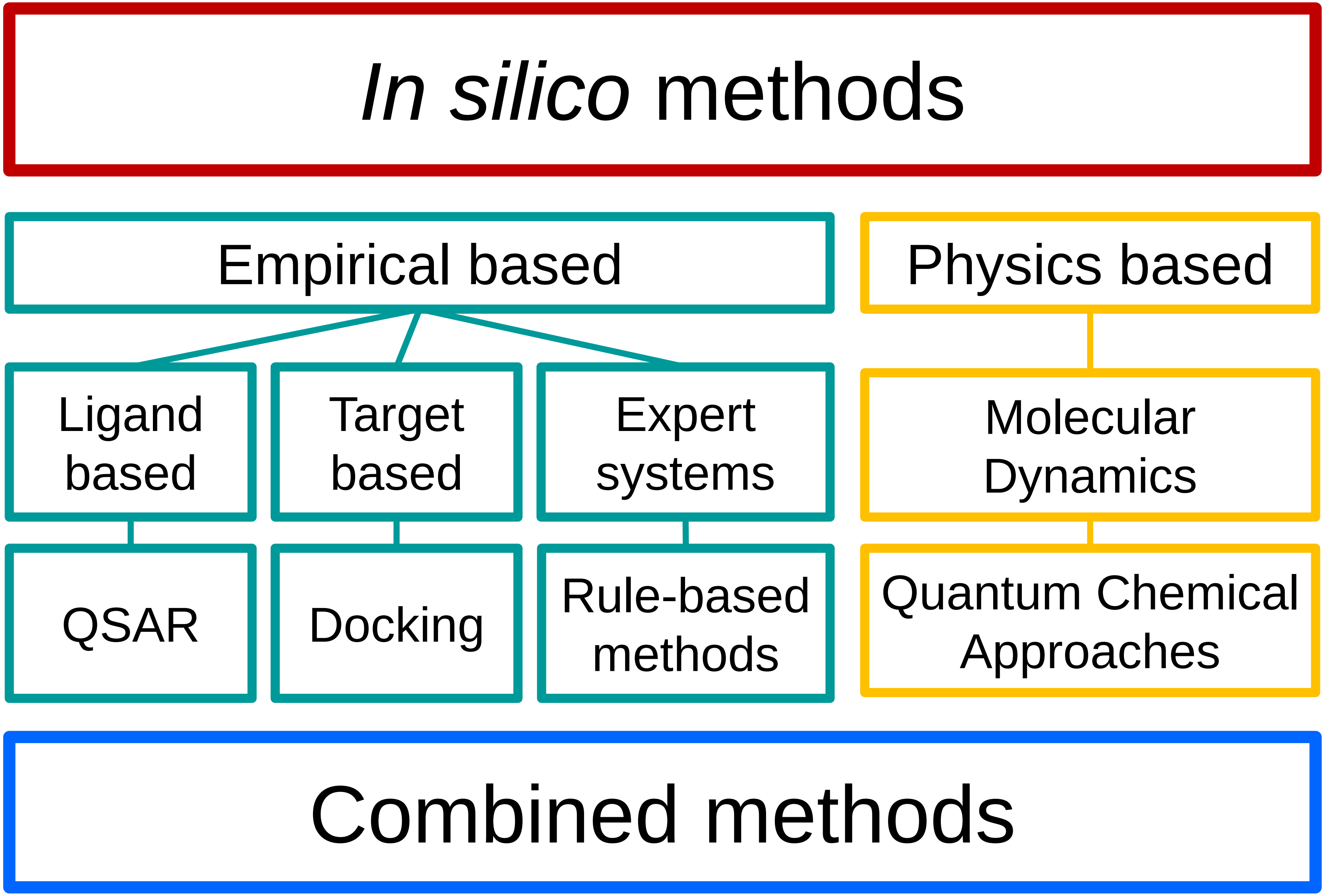
The steps towards modeling of nanoparticles properties and toxicity:

- Development of nanomaterials inventory (datasets) – collecting the data on experimental physicochemical properties, toxicity endpoints
- Identification of structural descriptors suitable for modeling nanoparticle reactivity
- QSAR modeling - exploring the relationships between structure and properties (for example, solubility), toxicity, using multivariate data analysis techniques
- Modeling the interaction of nanoparticles with biological systems - by means of computational approaches including quantum chemistry methods, molecular modeling and protein-ligand docking techniques

Two types of nanomaterials

- Metal-based nanomaterials (metal oxide NPs)
- Carbon nanostructures (fullerenes, carbon nanotubes)

Combination of computational methods to predict nano-properties



In silico methods

Empirical based

Ligand based

QSAR

Target based

Docking

Expert systems

Rule-based methods

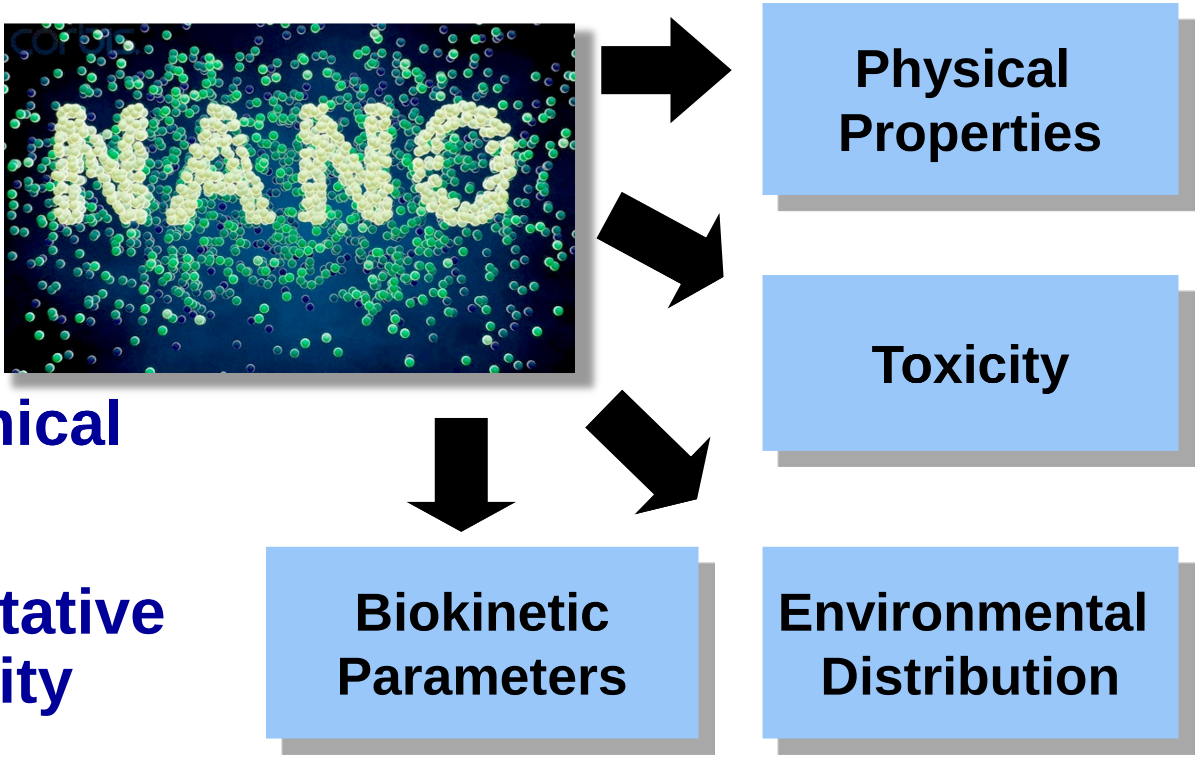
Physics based

Molecular Dynamics

Quantum Chemical Approaches

Combined methods

Computational approaches



Physical Properties

Toxicity

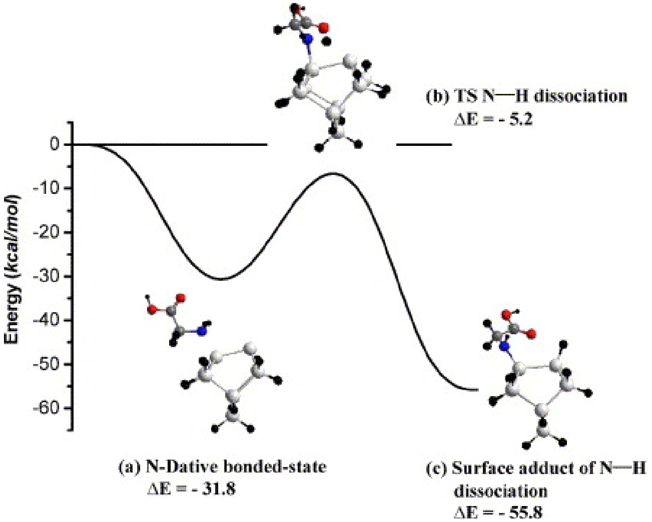
Biokinetic Parameters

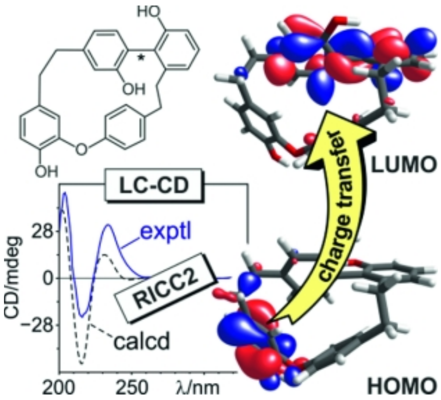
Environmental Distribution

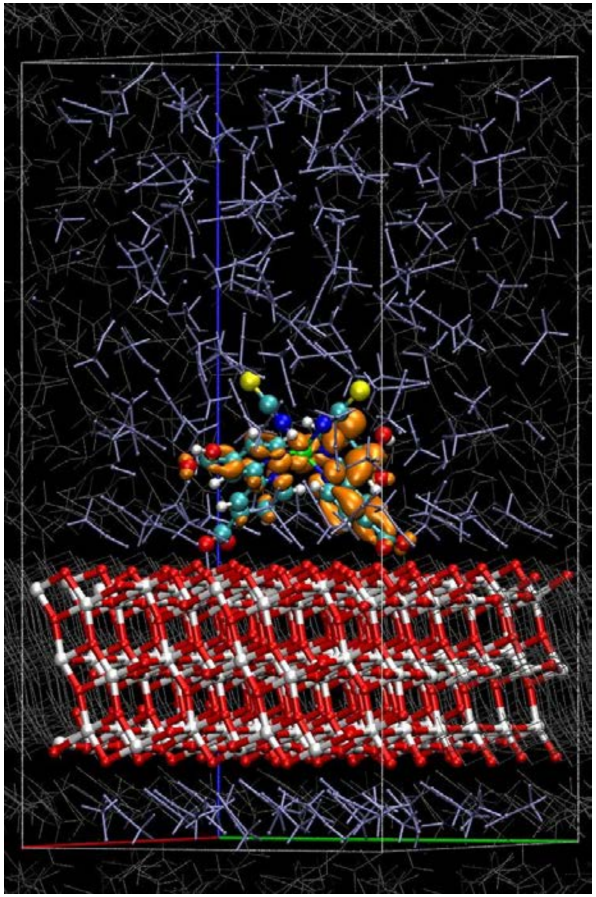
- Quantum-Chemical Approaches
- QSARs: Quantitative Structure-Activity Relationships
- Molecular modeling – Protein-Ligand Docking
- Data visualization and Pattern recognition methods

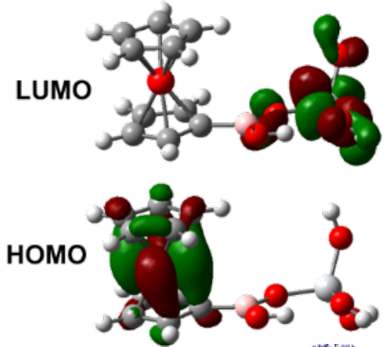
Quantum Chemistry

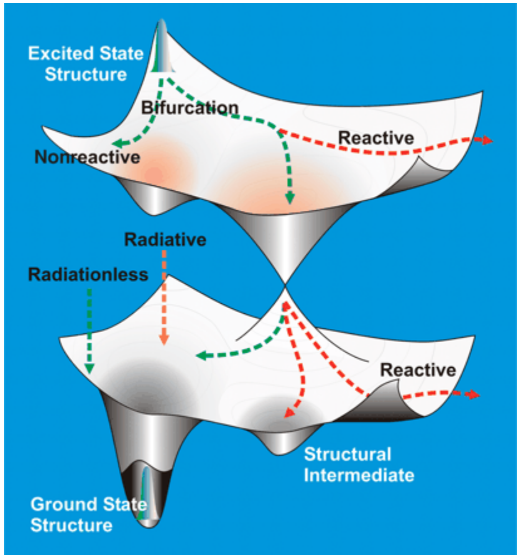
Quantum-Chemical Approaches

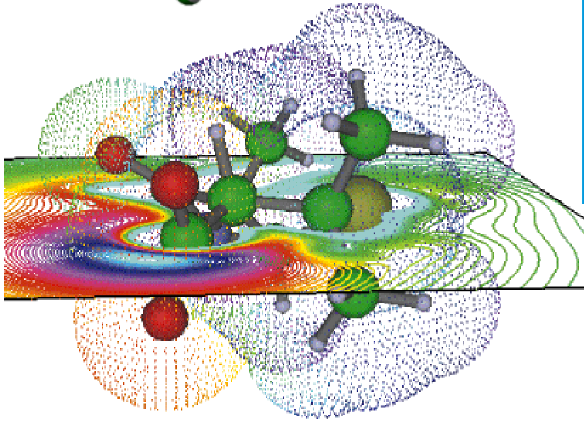

(b) TS N—H dissociation
ΔE = - 5.2
0
-10
-20
-30
-40
-50
-60
Energy (kcal/mol)
(a) N-Dative bonded-state
ΔE = - 31.8
(c) Surface adduct of N—H dissociation
ΔE = - 55.8

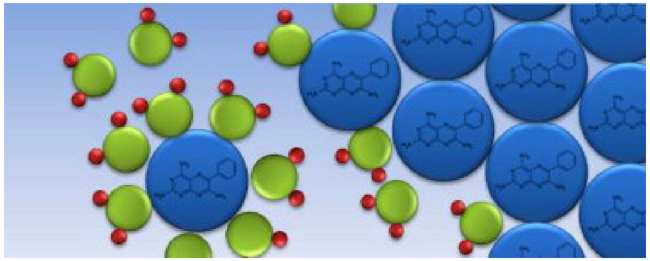

HO
*
OH
OH
LC-CD
28
0
−28
CD/mdeg
exptl
RICC2
calcd
200
250
λ/nm
charge transfer
LUMO
HOMO




LUMO
HOMO

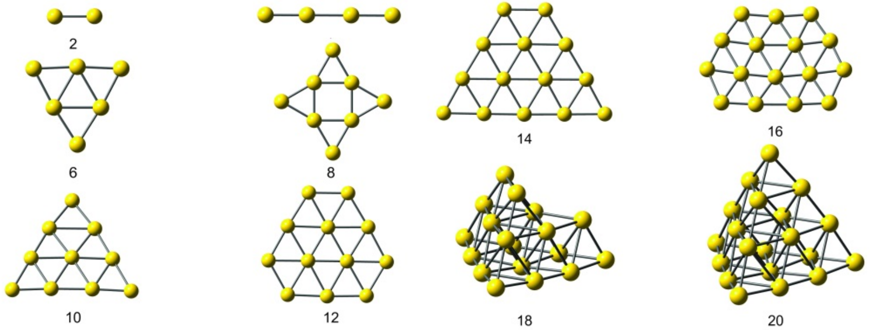

Excited State Structure
Bifurcation
Nonreactive
Reactive
Radiative
Radiationless
Reactive
Structural Intermediate
Ground State Structure

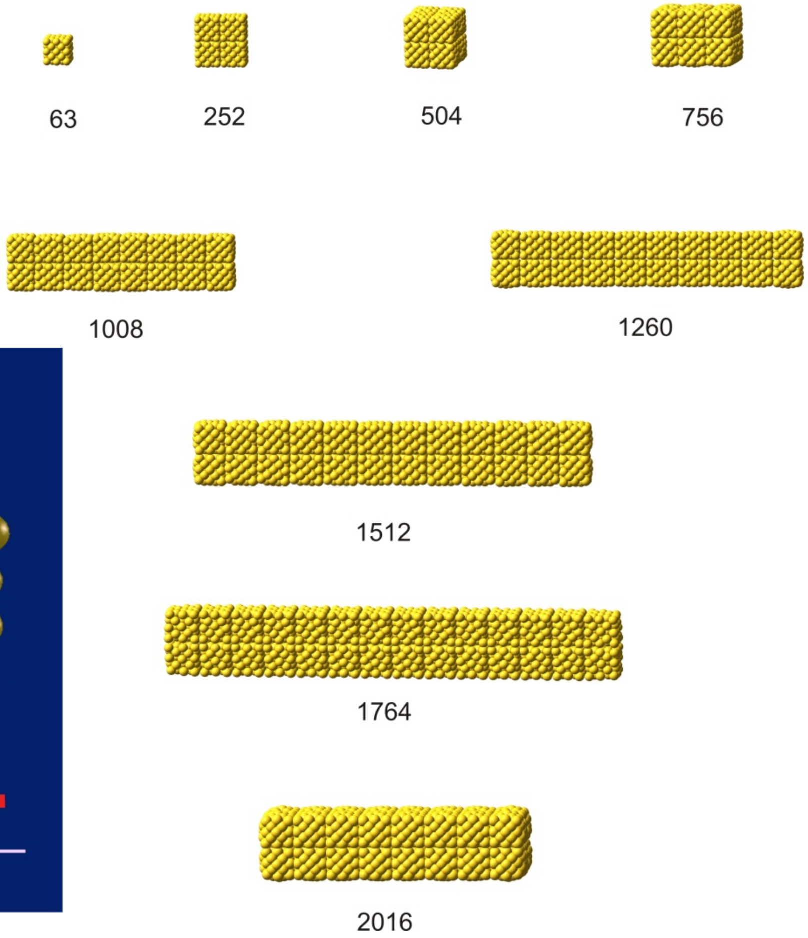


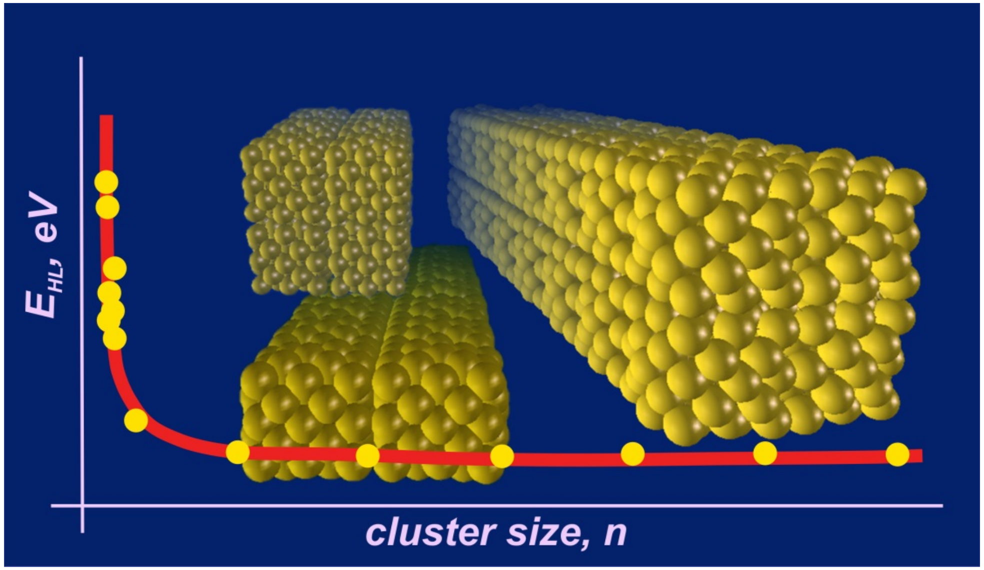


Gold nanoclusters

Small and Large gold cubic clusters, Au$_n$, n=2-2016

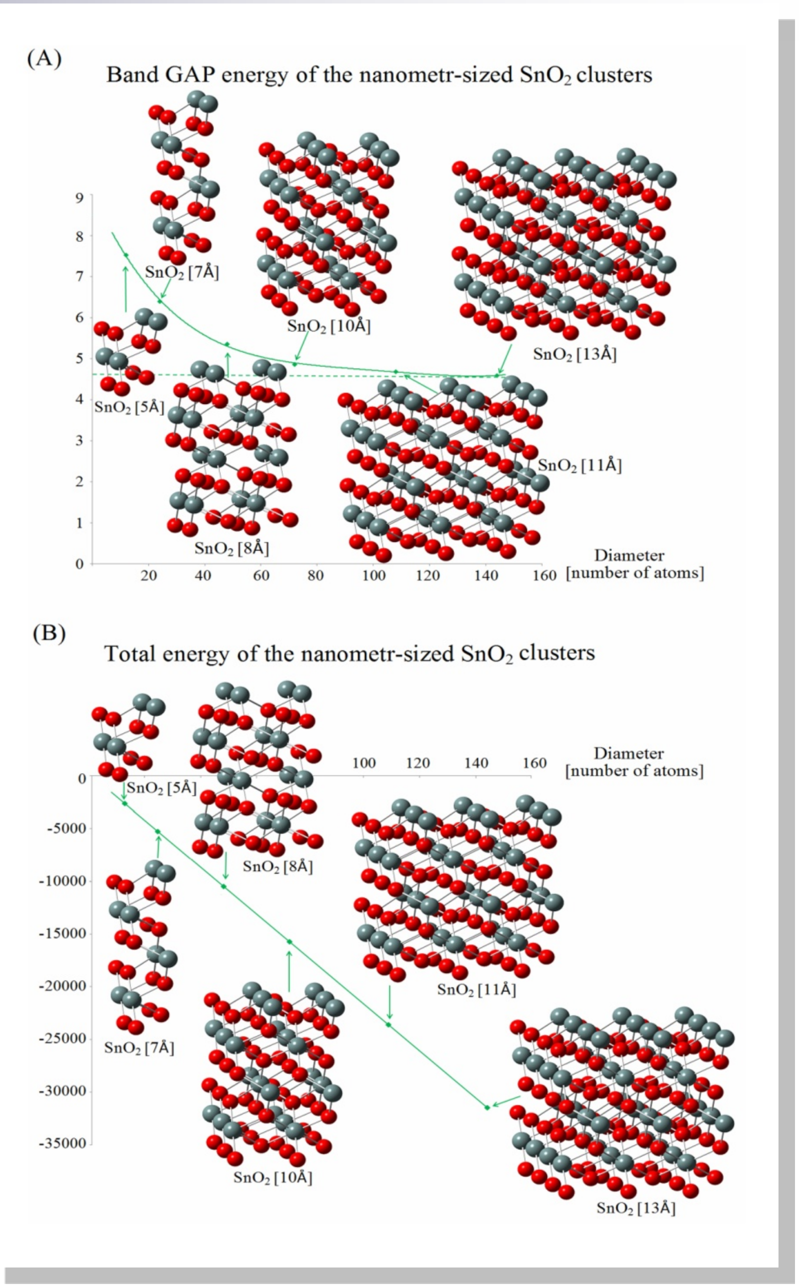






Watkins M., Rasulev B., Theodore M., Jackman J., Leszczynski J., Structures and Stabilities: Quantum-Chemical Study of Au n (n = 2-2016) Nanoclusters by Extended Huckel and DFT Approaches, *Nanosci.& Nanotech.*, (2012), 2(1), 1-12

Quantum-Mechanical Properties of Metal Oxide clusters



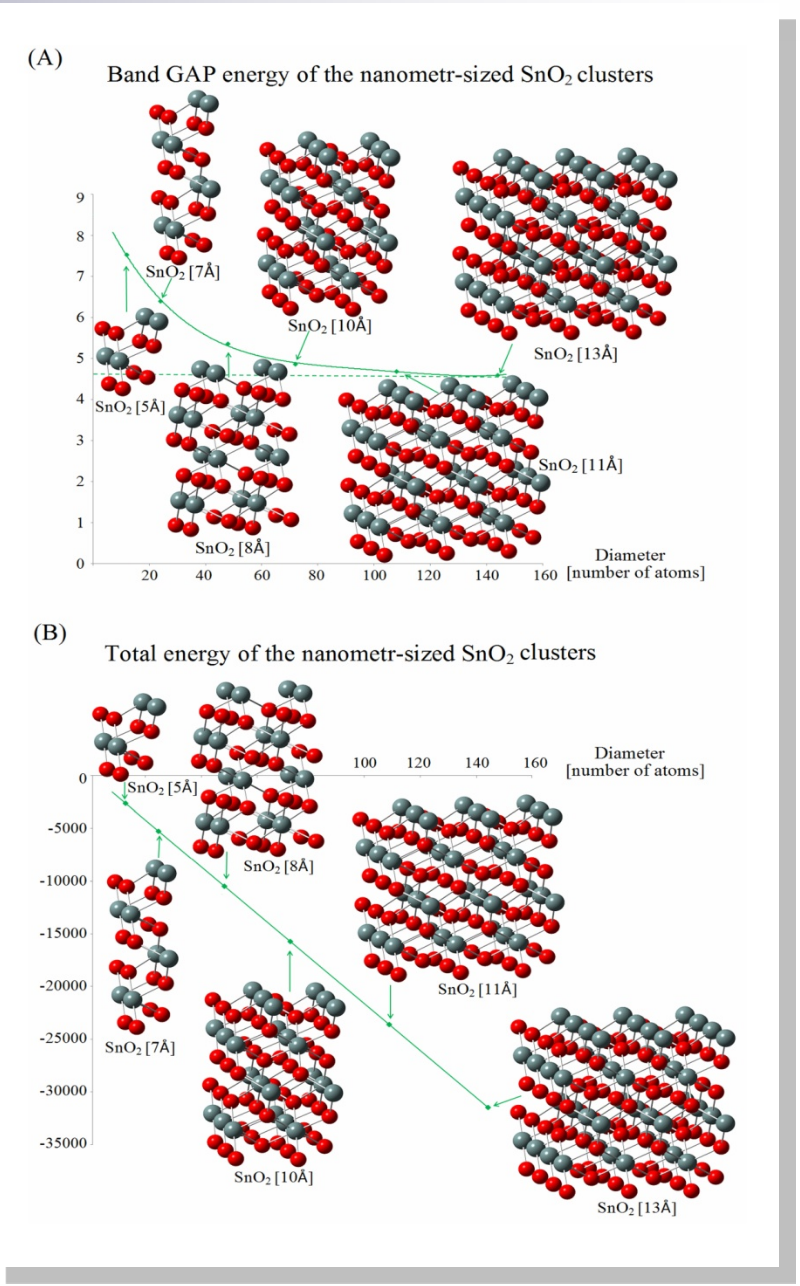
Scheme A: GAP, HOMO, LUMO, hardness, softness, electrophilicity

Scheme B: HOF, total energy, electronic energy, SAS, dipole moment

Gajewicz A., Puzyn T., Rasulev, B., Leszczynska D., Leszczynski J. Metal Oxide Nanoparticles: Size-Dependence of Quantum-Mechanical Properties, *Nanoscience & Nanotechnology*, (2011), 1, 53-58

Quantum-Mechanical Properties of Metal Oxide clusters

Non-linear



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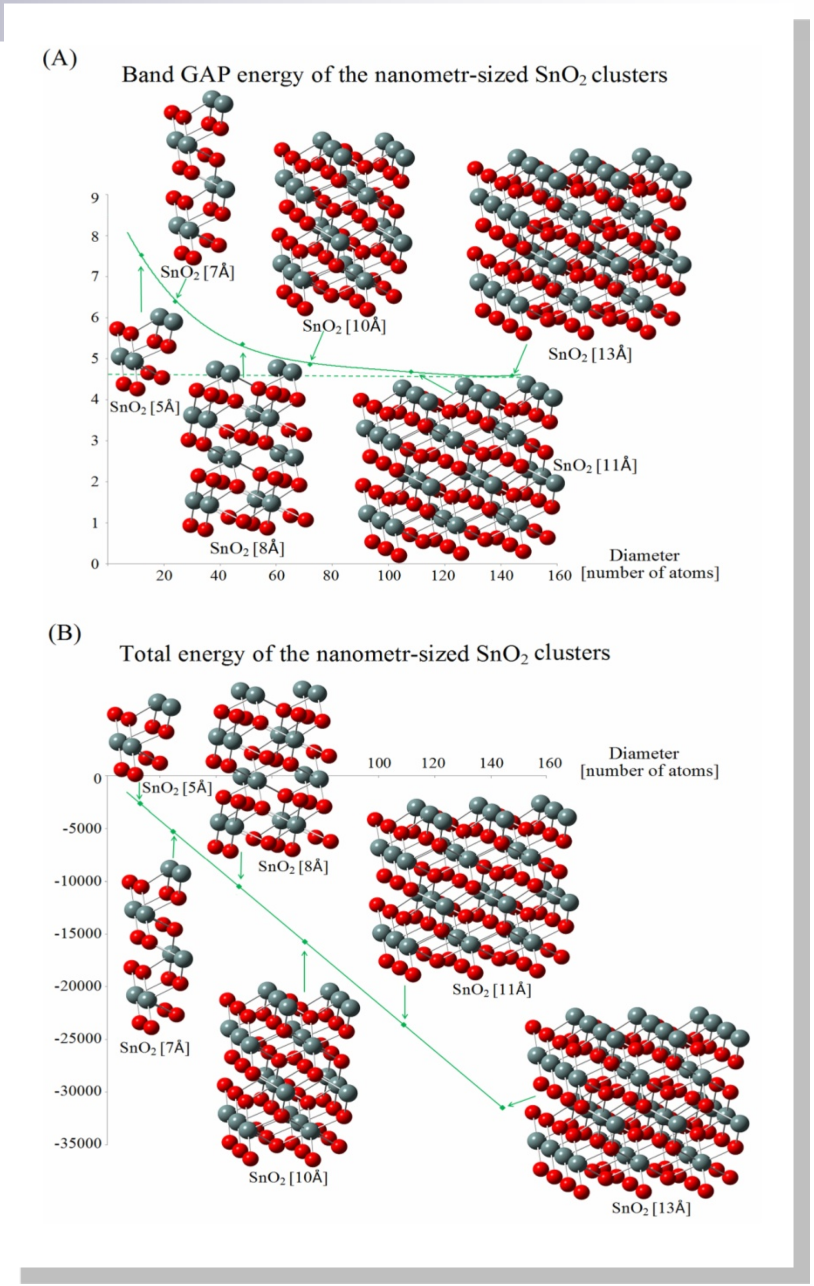
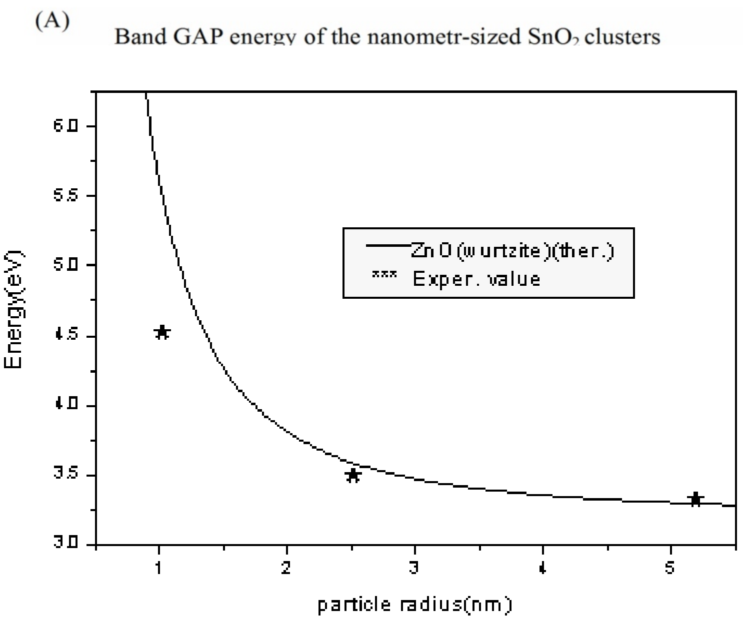
Scheme B: HOF, total energy, electronic energy, SAS, dipole moment



Gajewicz A., Puzyn T., Rasulev, B., Leszczynska D., Leszczynski J. Metal Oxide Nanoparticles: Size-Dependence of Quantum-Mechanical Properties, *Nanoscience & Nanotechnology*, (2011), 1, 53-58

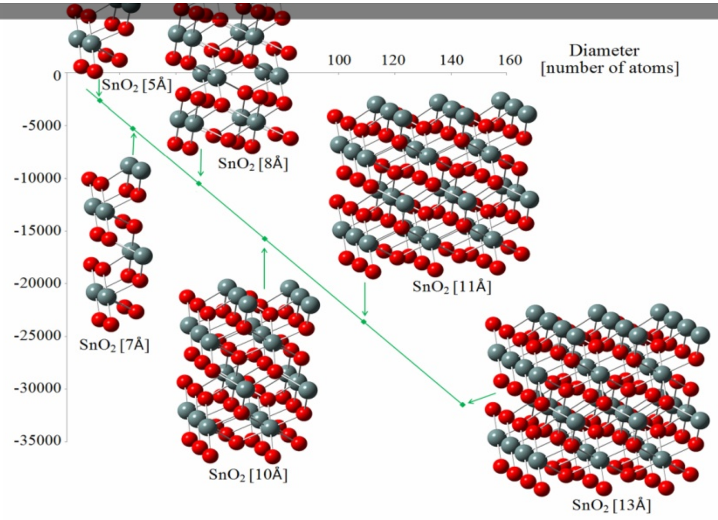
Quantum-Mechanical Propertie
of Metal Oxide clusters

Non-linear



Scheme A: GAP, HOMO, LUMO, hardness, softness, electrophilicity

Linear



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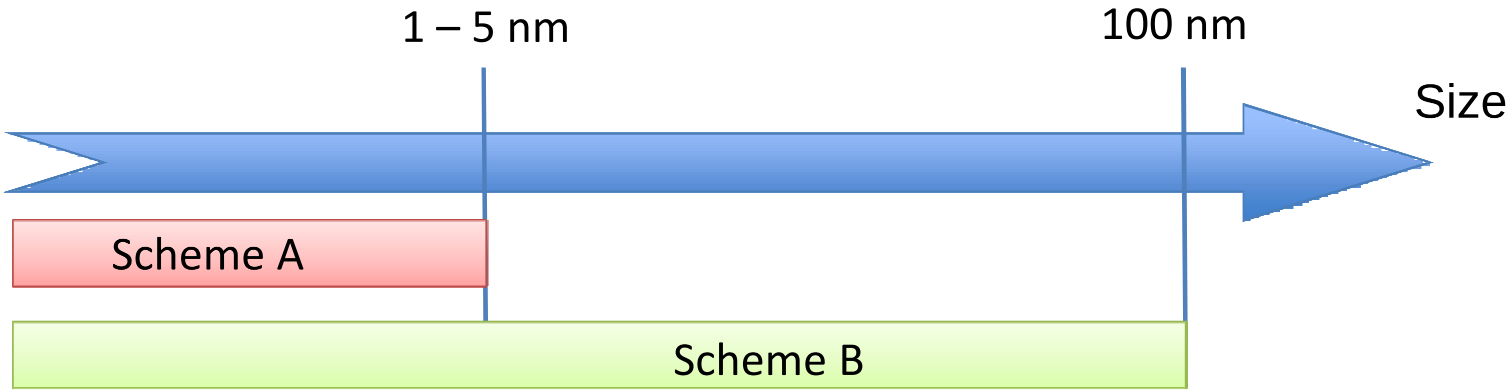
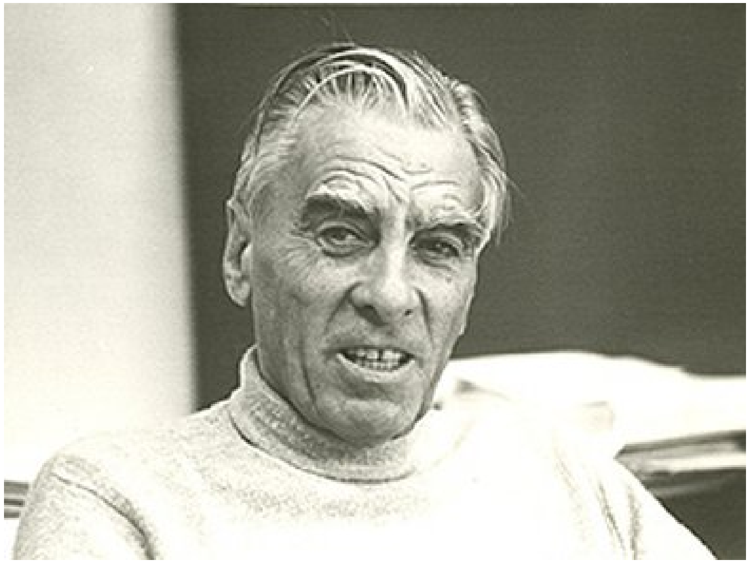
Size-Dependence of Quantum-Mechanical Properties



Gajewicz A., Puzyn T., Rasulev, B., Leszczynska D., Leszczynski J. Metal Oxide Nanoparticles: Size-Dependence of Quantum-Mechanical Properties, *Nanoscience & Nanotechnology*, (2011), 1, 53-58

QSAR

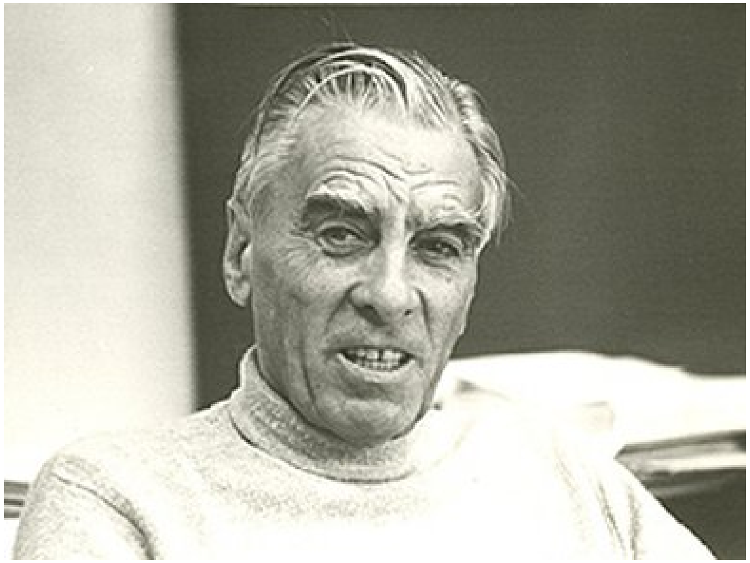
Quantitative Structure-Activity Relationship in materials research



Corwin Hansch

1918-2011

The Pioneer of QSAR



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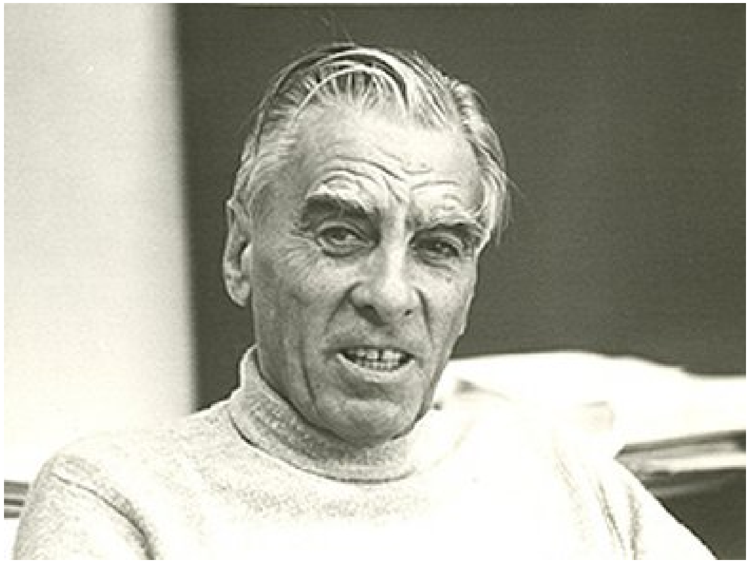
The Father of QSAR



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The Father of QSAR

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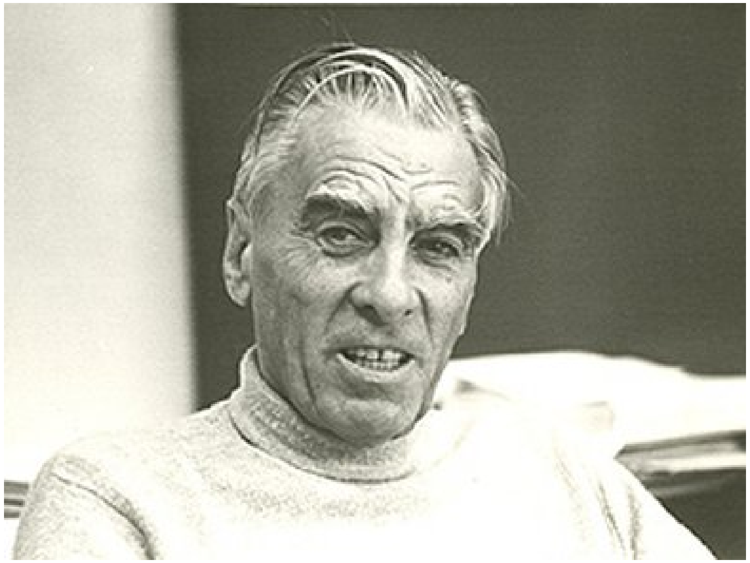

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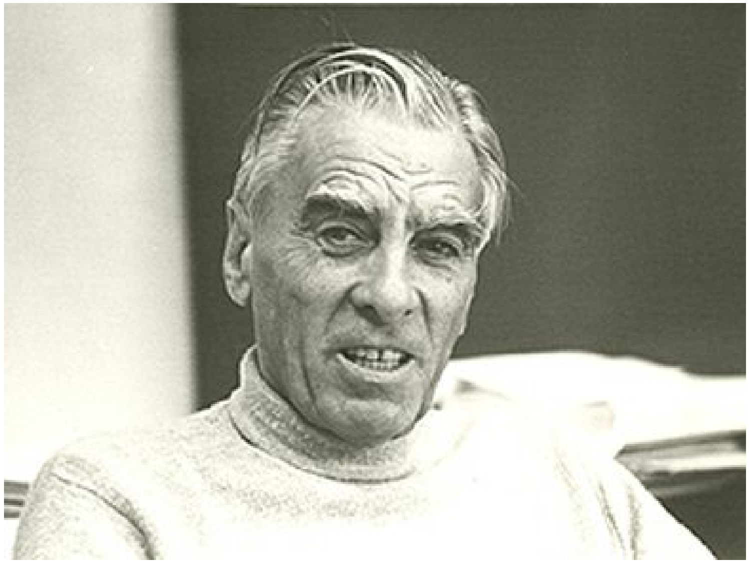

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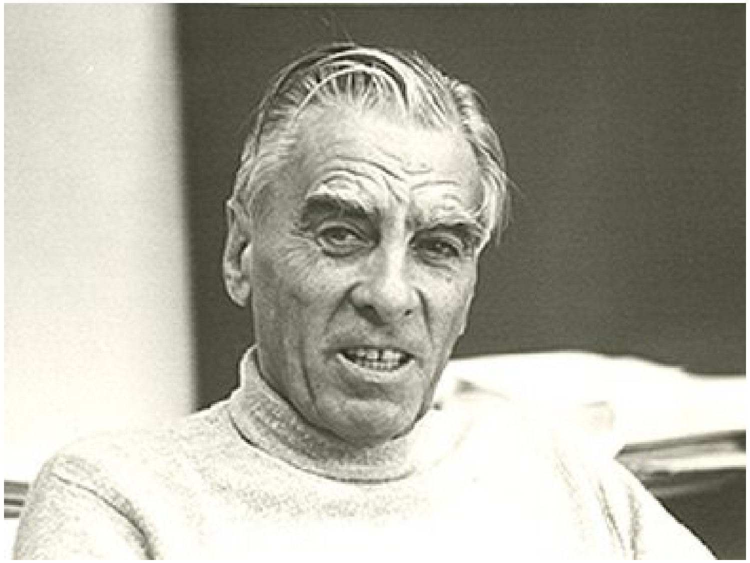

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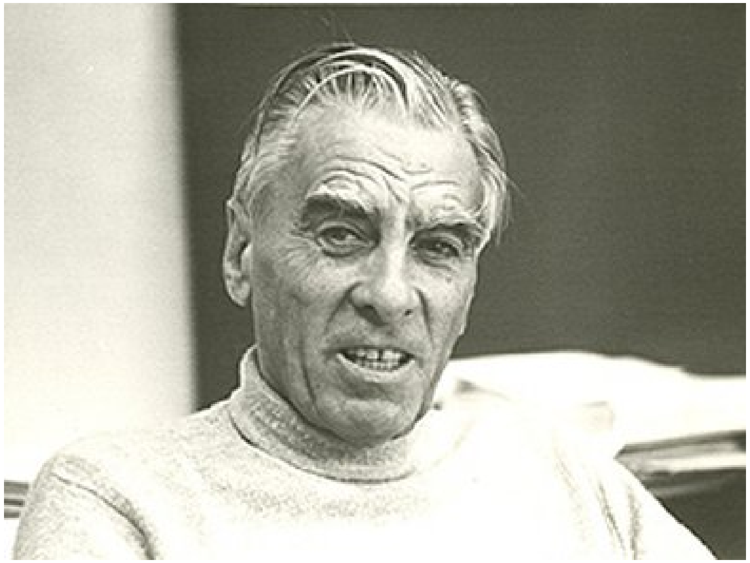
Hansch worked on the Manhattan Project at the University of Chicago and as a group leader at DuPont Nemours in Richland, Washington.



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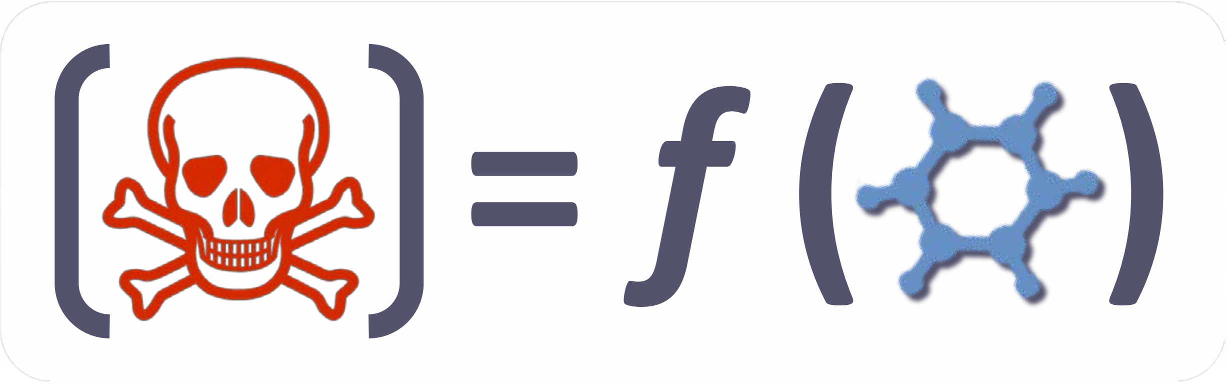
(Q)SAR

=

(Quantitative) Structure-Activity Relationship



IN SILICO

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QSAR – what is this?

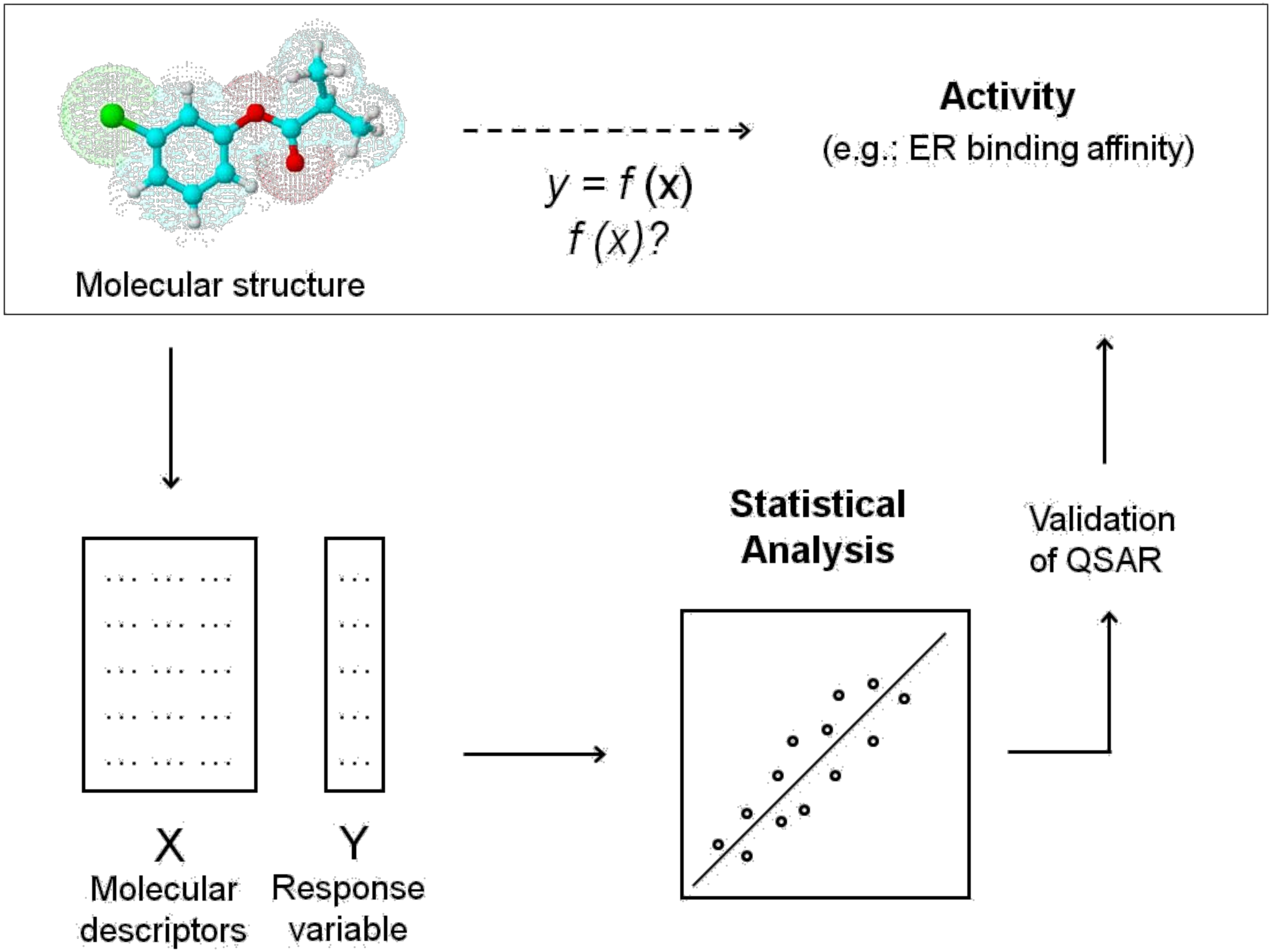
- A QSAR is a ***mathematical relationship between a biological activity of a molecular system and its geometric, chemical or physical characteristics*****.**
- QSAR attempts to find consistent relationship between biological activity and molecular properties, so that these “rules” can be used to evaluate the activity of new compounds.
- Once a valid QSAR has been determined, it should be possible to **predict** the physical property or biological activity of related compounds or drug candidates before they are put through expensive and time-consuming biological testing. In some cases, only computed values need to be known to make an assessment.

The problem of QSAR is to find coefficients $C_0, C_1, \ldots C_n$ such that:

Biological activity $= C_0+(C_1*P_1)+\ldots+(C_n*P_n)$

and the prediction error is minimized for a list of given m compounds.

QSAR methodology



Molecular structure

$y = f(\mathbf{x})$

$f(x)$?

Activity

(e.g.: ER binding affinity)

X

Molecular descriptors

Y

Response variable

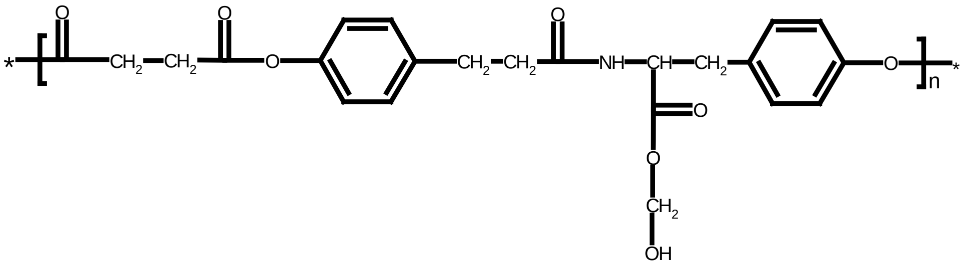
Statistical Analysis

Validation of QSAR

Types of Molecular Descriptors

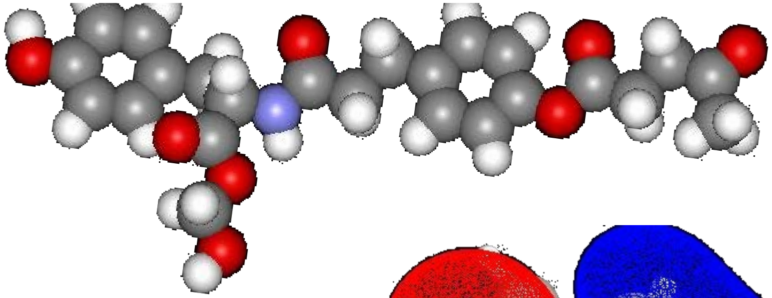
Constitutional, Topological

2-D structural formula

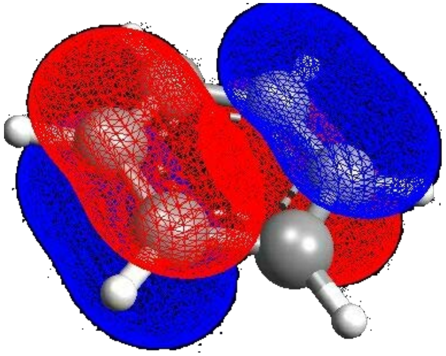


Geometrical

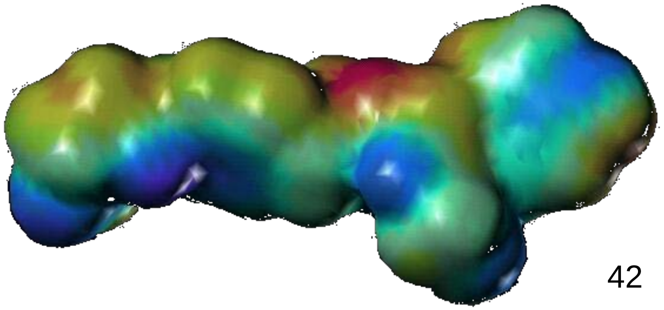
3-D shape and structure



Quantum Chemical



Electrostatic



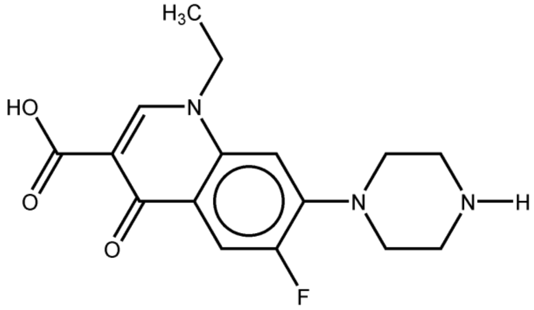
Hybrid descriptors

Examples of successful QSAR applications in industry

Norfloxacin, **antibacterial**

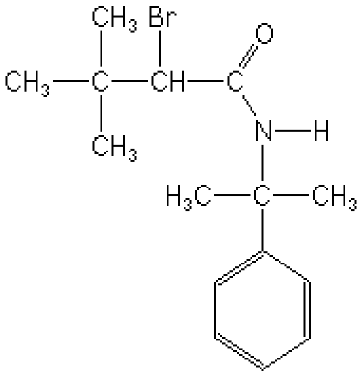
Kyorin Pharmaceutical Company, Japan

Traditional QSAR analysis of 70 compounds, up to 500 times more potent then previous analogs



Bromobutide, **herbicide**

Sumitomo chemical Company, Japan

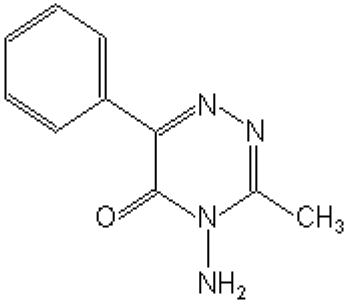


QSAR analysis of 74 compounds

Metamitron, **herbicide**

Bayer AG, Germany

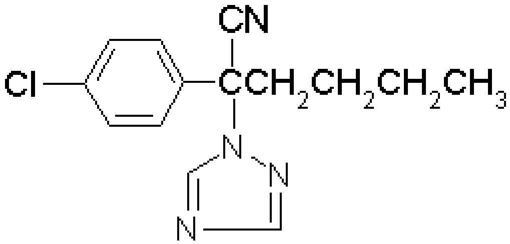
QSAR analysis of 22 compounds



Myclobutanil, **fungicide**

Rohm and Haas, USA

QSAR analysis of 67 compounds

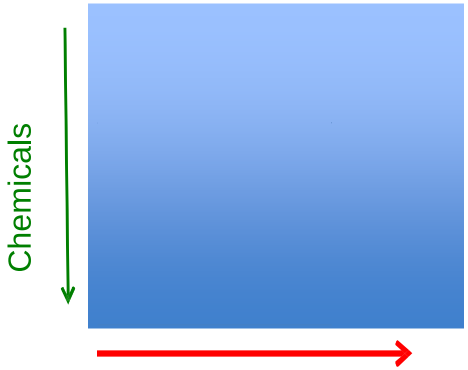


Extending QSAR to nanoparticles

There are three problems in order to extend QSAR approach to materials (nanomaterials and polymer materials):

1. QSAR mainly developed for organic compounds with diverse structure types, while nanoparticles structurally limited in diversity
2. Not enough experimental data for nanoparticles and no systematic data
3. Regular QSAR descriptors applicable for organic compounds – not applicable for nanoparticles

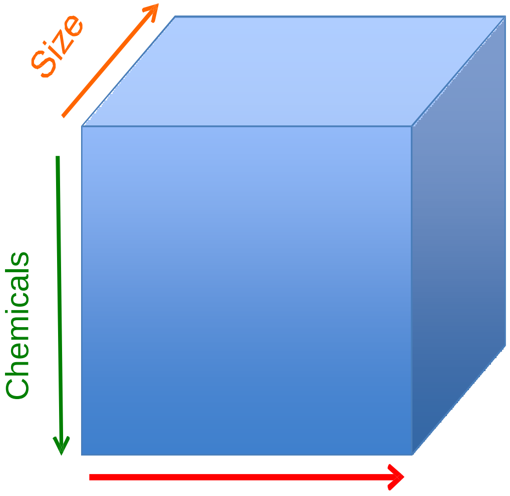
Data for “Classic” QSAR and nano-QSAR



Chemicals

Descriptors / Properties

„Classic” QSAR



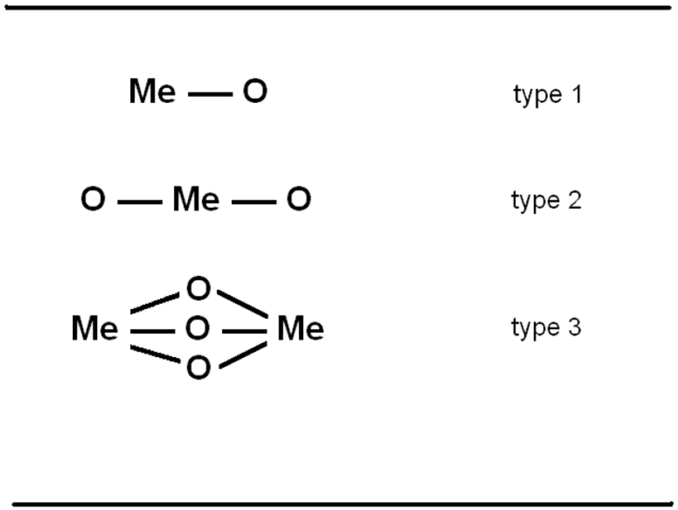
Size

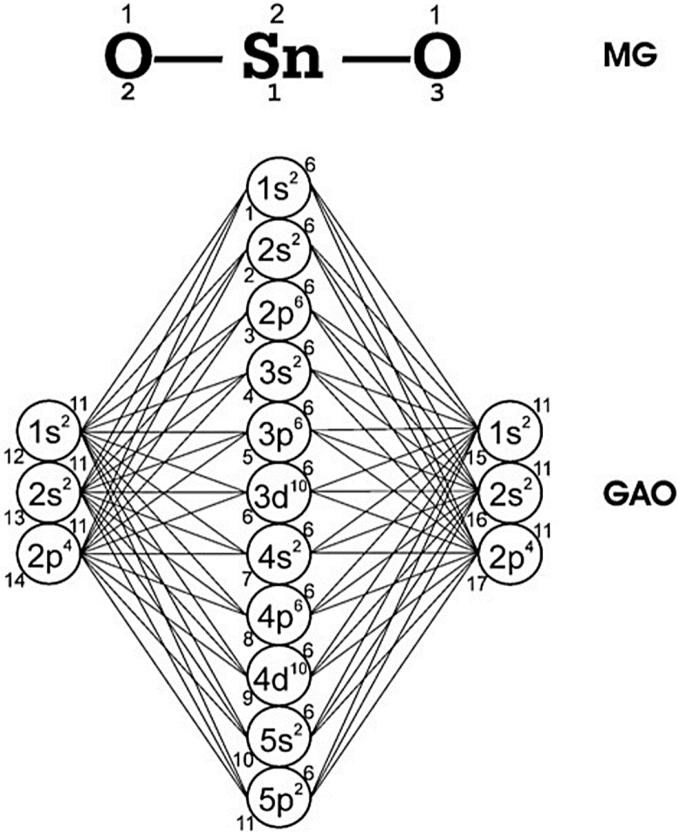
Chemicals

Descriptors / Properties

Nano-QSAR/QNTR

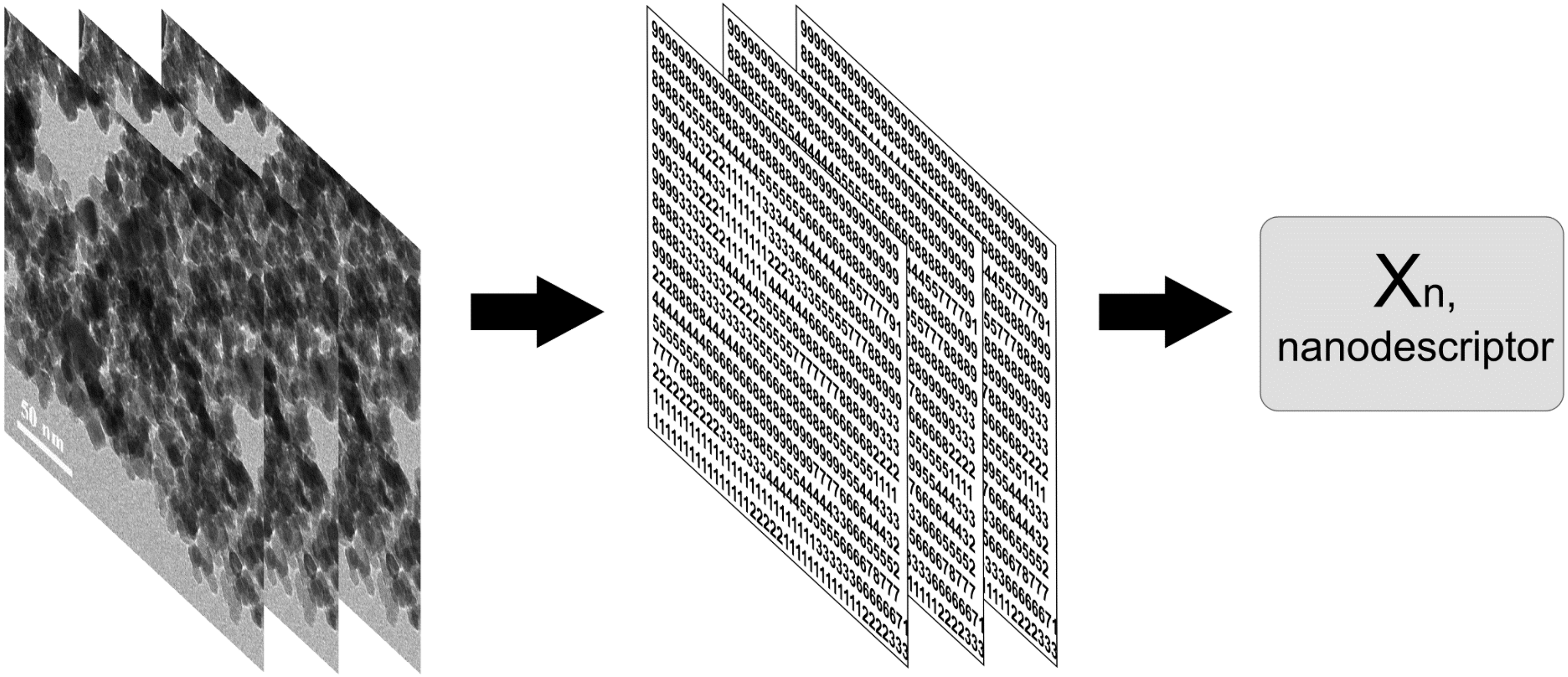
Materials' descriptors (Nano-descriptors)





B. Rasulev, A. Toropov, T. Puzyn, D. Leszczynska, J. Leszczynski, An Application of Graphs of Atomic Orbitals for QSAR Modeling of Toxicity of Metal Oxides, in: Federation of Analytical Chemistry and Spectroscopy Symposium (FACSS), 2007

Materials’ descriptors (Nano-descriptors)



X_n, nanodescriptor

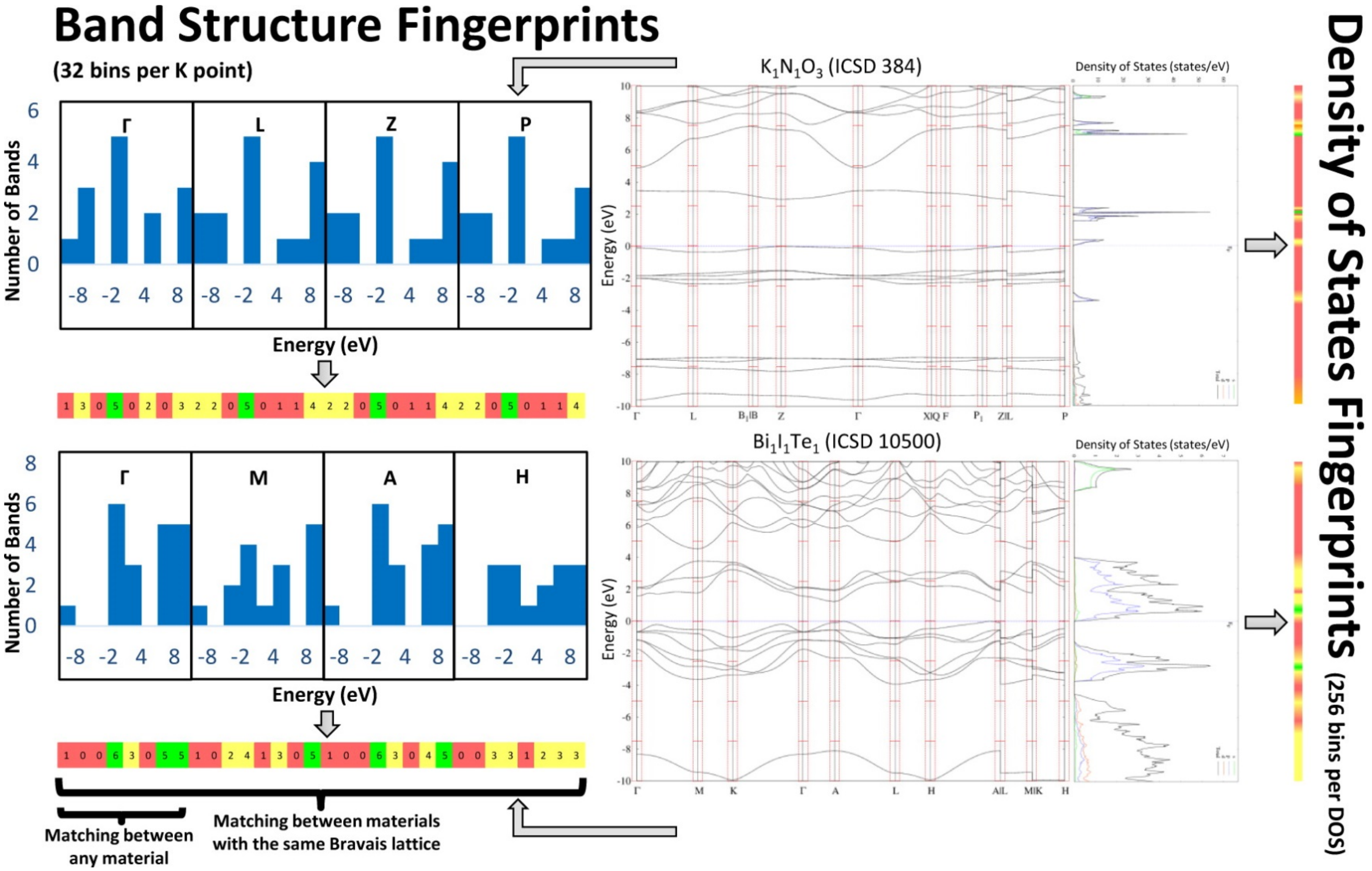
TEM images

Processed images
Numerical representation

Surface
nanodescriptor

A Gajewicz, B Rasulev, TC Dinadayalane, P Urbaszek, T Puzyn, D. Leszczynska, J. Leszczynski, Advancing risk assessment of engineered nanomaterials: application of computational approaches, *Advanced Drug Delivery Reviews*, 2012, 64 (15), 1663-1693

Fingerprint descriptors for materials



Construction of materials fingerprints from the band structure and the density of states. Copyright (Isayev et al., Materials Cartography: Representing and Mining Materials Space Using Structural and Electronic Fingerprints, *Chemistry of Materials*, 2015, 27, 735-743).

“Liquid drop” model as a nano-descriptor

[Ti] = [O] = [Ti]

⇩

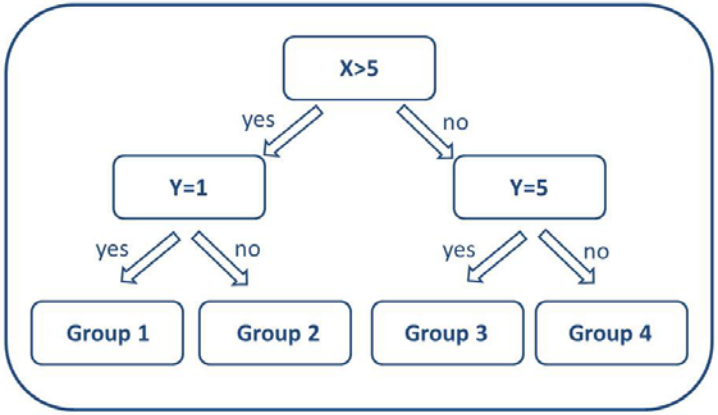
Rule for electronegativity ($A \leq 1.5 \leq B \leq 2.0 \leq C \leq 2.5 \leq D \leq 3.5 \leq F$)

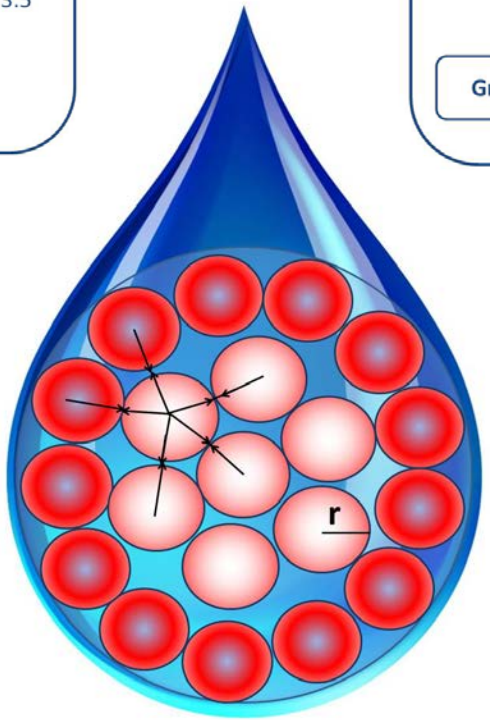
⇩

Electronegativity of [Ti] = 1.5; Electronegativity of [O] = 3.5

⇩

[A] = [D] = [A]





$$r_w = \left(\frac{3M}{4\pi\rho N_A}\right)^{1/3}$$

$$n = \left(\frac{r_0}{r_w}\right)^3$$

“Liquid Drop” nanodescriptor representation

N Sizochenko, B Rasulev, A Gajewicz, V Kuz'min, T Puzyn, J Leszczynski, From basic physics to mechanisms of toxicity: the “liquid drop” approach applied to develop predictive classification models for toxicity of metal oxide nanoparticles, *Nanoscale*, 2014, 6, 13986-993

Nano-QSAR based on SiRMS descriptors and “liquid drop” nanodescriptor

[Ti] = [O] = [Ti]

⇩

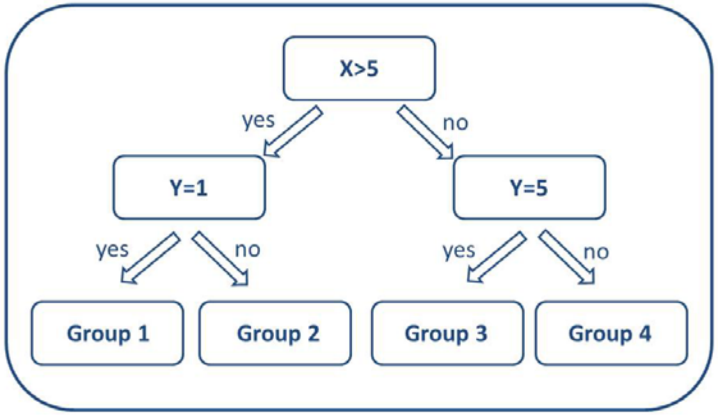
Rule for electronegativity (A≤ 1.5 ≤ B ≤ 2.0 ≤ C ≤ 2.5 ≤ D ≤ 3.5 ≤ F)

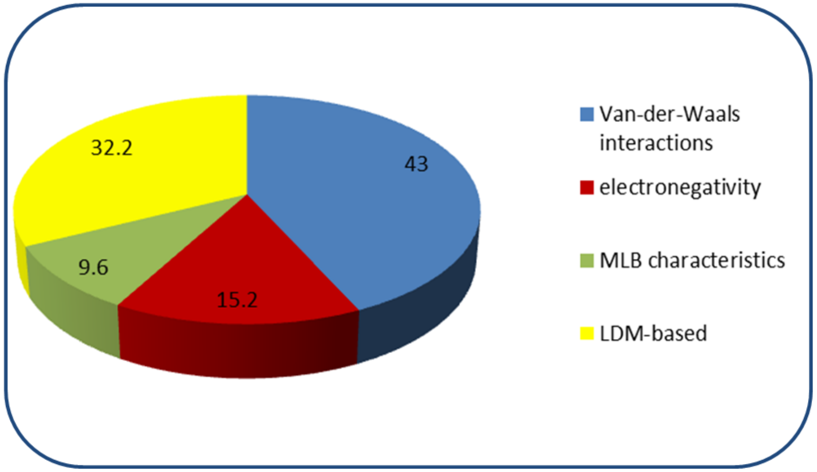
⇩

Electronegativity of [Ti] = 1.5; Electronegativity of [O] = 3.5

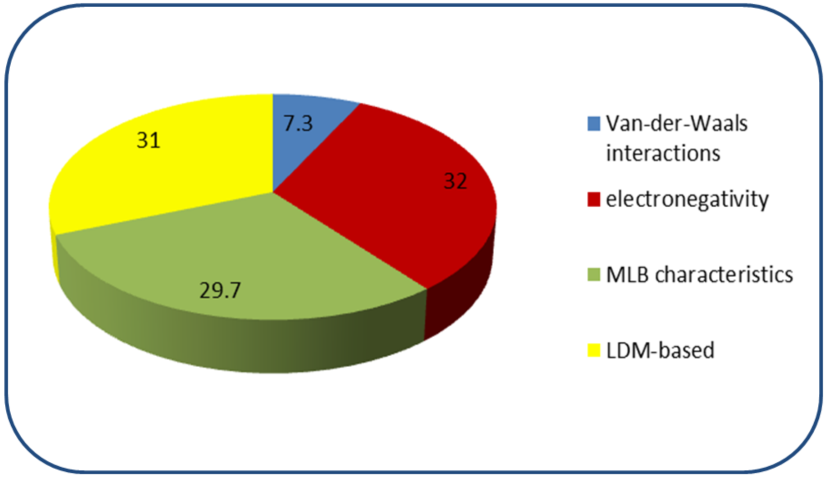
⇩

[A] = [D] = [A]





E.Coli cell toxicity

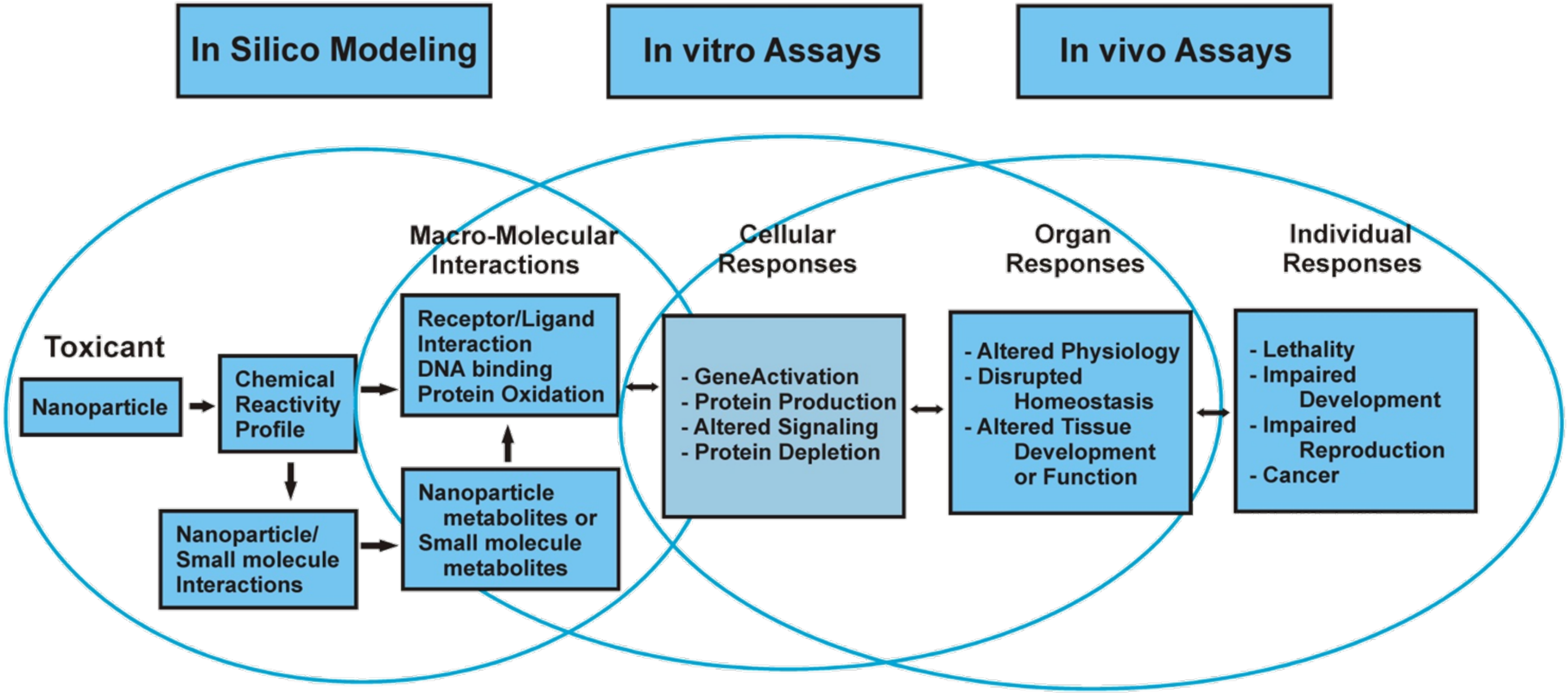


HaCaT cells toxicity

N Sizochenko, B Rasulev, A Gajewicz, V Kuz'min, T Puzyn, J Leszczynski, From basic physics to mechanisms of toxicity: the “liquid drop” approach applied to develop predictive classification models for toxicity of metal oxide nanoparticles, *Nanoscale*, 2014, 6, 13986-993

Toxicity of nanomaterials

An Example of Toxicity Pathway for Nanoparticles



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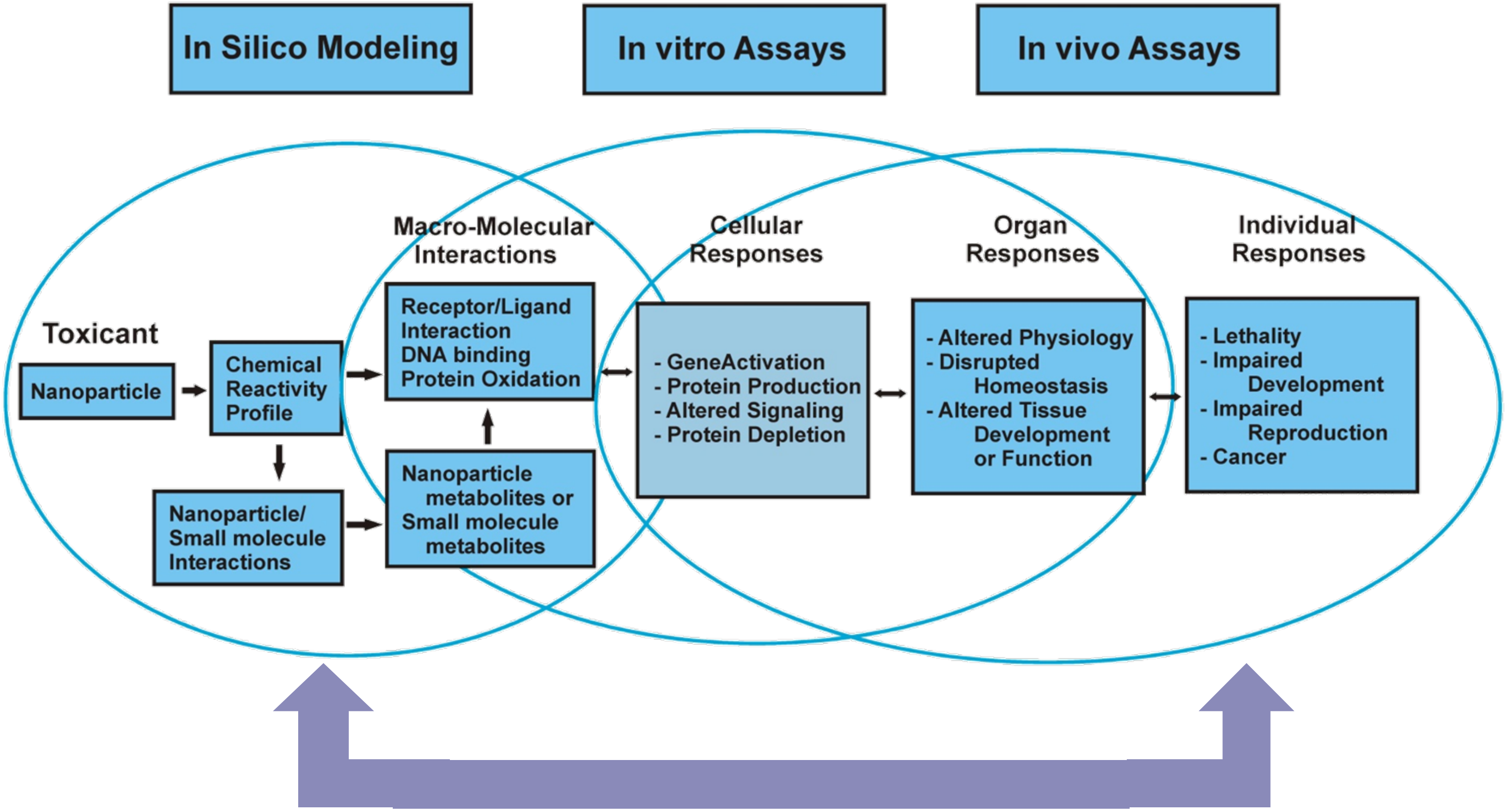


In Silico Modeling

In vitro Assays

In vivo Assays

Toxicant

Nanoparticle

Chemical Reactivity Profile

Nanoparticle/ Small molecule Interactions

Macro-Molecular Interactions

Receptor/Ligand Interaction
DNA binding
Protein Oxidation

Nanoparticle metabolites or Small molecule metabolites

Cellular Responses

- GeneActivation
- Protein Production
- Altered Signaling
- Protein Depletion

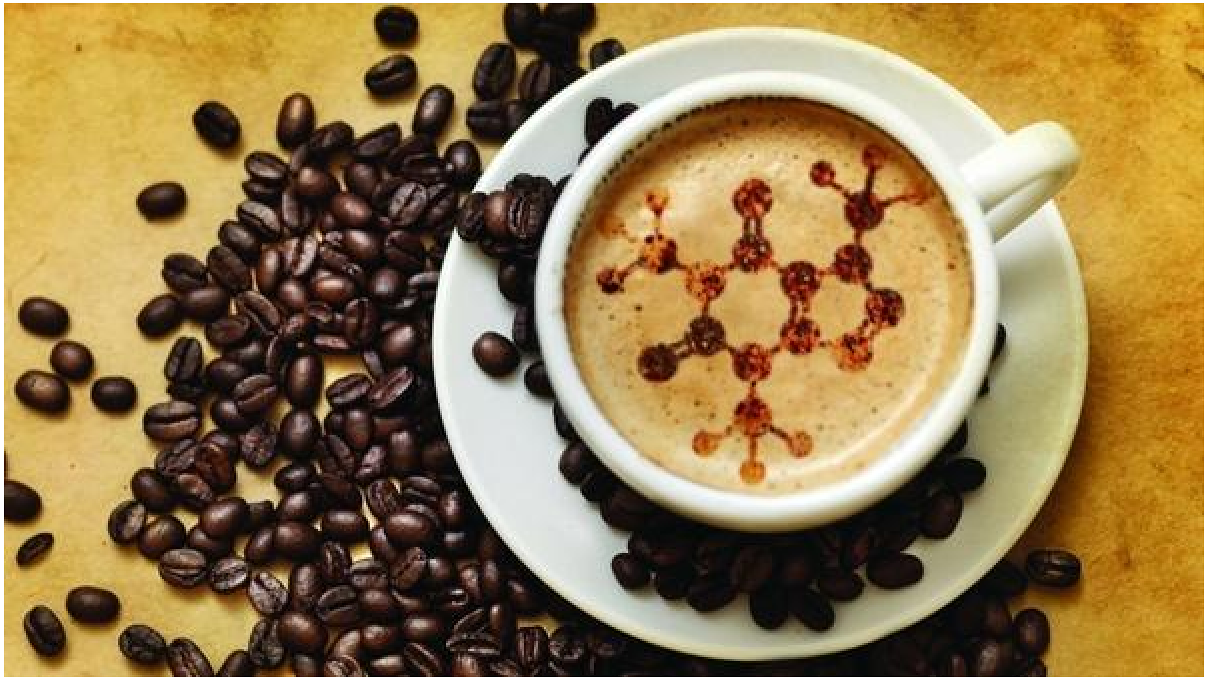
Organ Responses

- Altered Physiology
- Disrupted Homeostasis
- Altered Tissue Development or Function

Individual Responses

- Lethality
- Impaired Development
- Impaired Reproduction
- Cancer

QSAR



Do you know what you're eating?

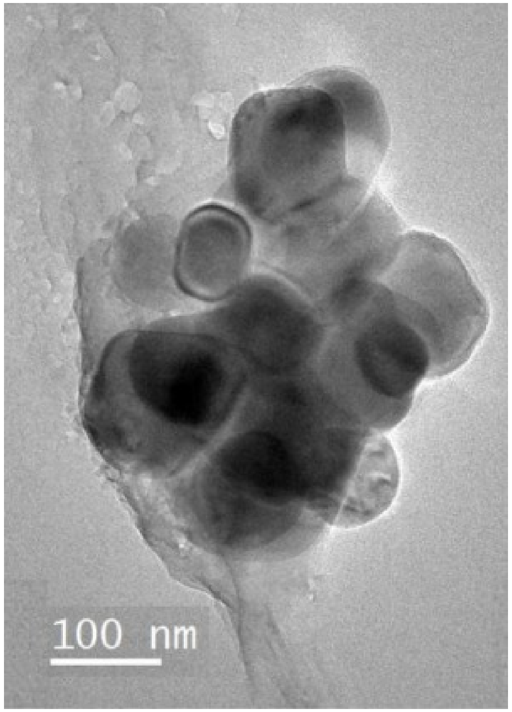
The number of American food products containing nanomaterials has increased tenfold since 2008. Nanoparticles are typically used to stretch the shelf life and improve the texture of food.

Popular lollies, sauces and dressings have been found to contain nanotechnology.

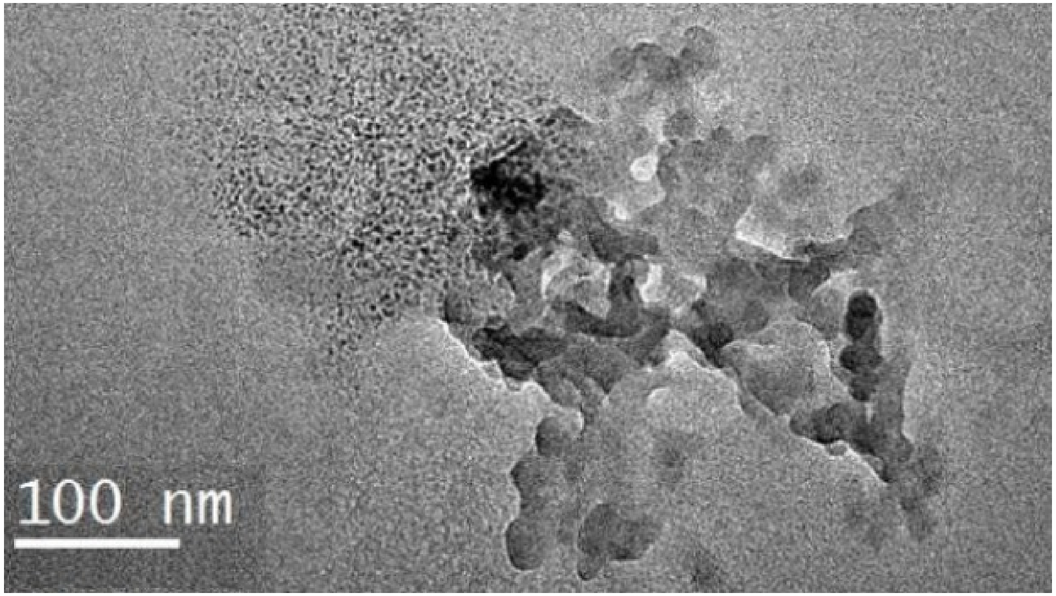
Tests that found potentially harmful nanoparticles of titanium dioxide and silica in 14 popular products, including Mars' M&Ms, Woolworths white sauce and Praise salad dressing.

The lab test of the 14 supermarket goods, which also included Eclipse chewy mints, Old El Paso taco mix, and Moccona Cappuccino, was conducted by a world-class nanotechnology research facility at Arizona State University.

The Food Standards code does not require nanoparticles to be declared on labelling. Nano-titanium dioxide (E171) can be simply described as the conventional-sized type and as "Colour (171)". Nano-silica (E551) can be listed as the conventional version and as "Anti-caking agent (551)".



Nanoparticles of titanium dioxide found in Mentos Pure Fresh Gum.
Photo: Arizona State University



Nanoparticles of silica found in Maggi's Roast Meat Gravy.
Photo: Arizona State University

Donuts

Donuts, Toxicity… and Solar cells

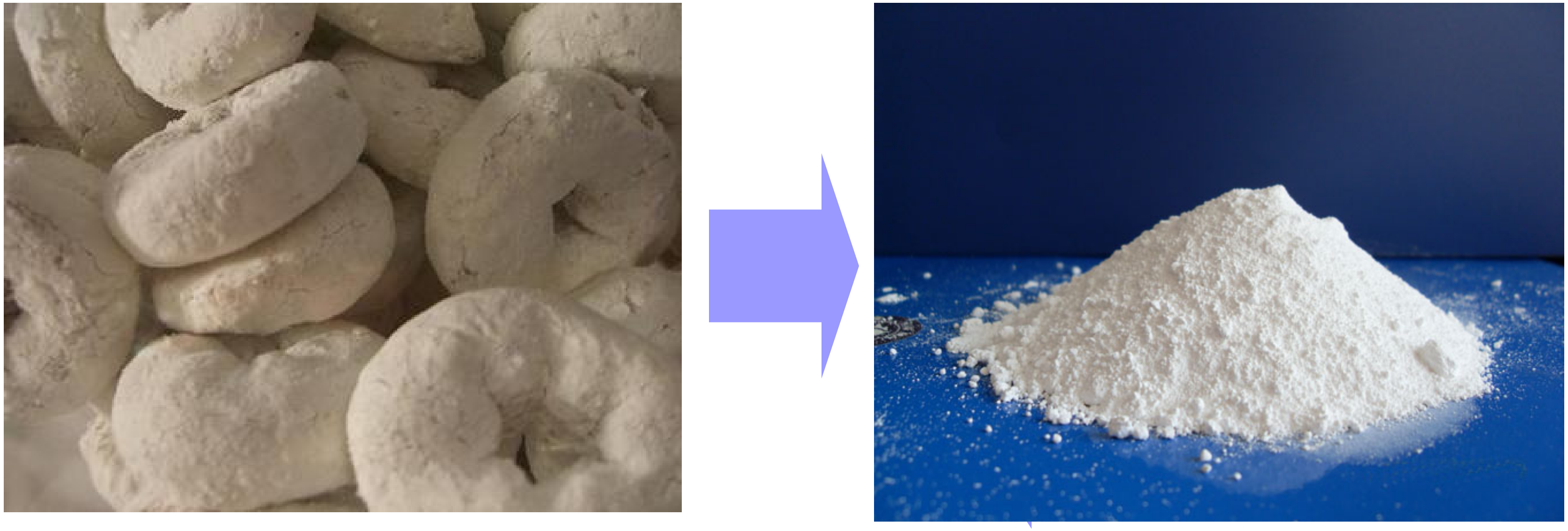
Donuts, Toxicity… and Solar cells

Sugar + TiO_2

Donuts, Toxicity… and Solar cells

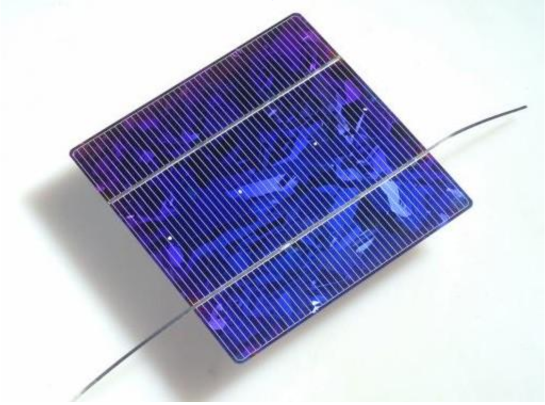
Sugar + TiO_2

Donuts, Toxicity… and Solar cells

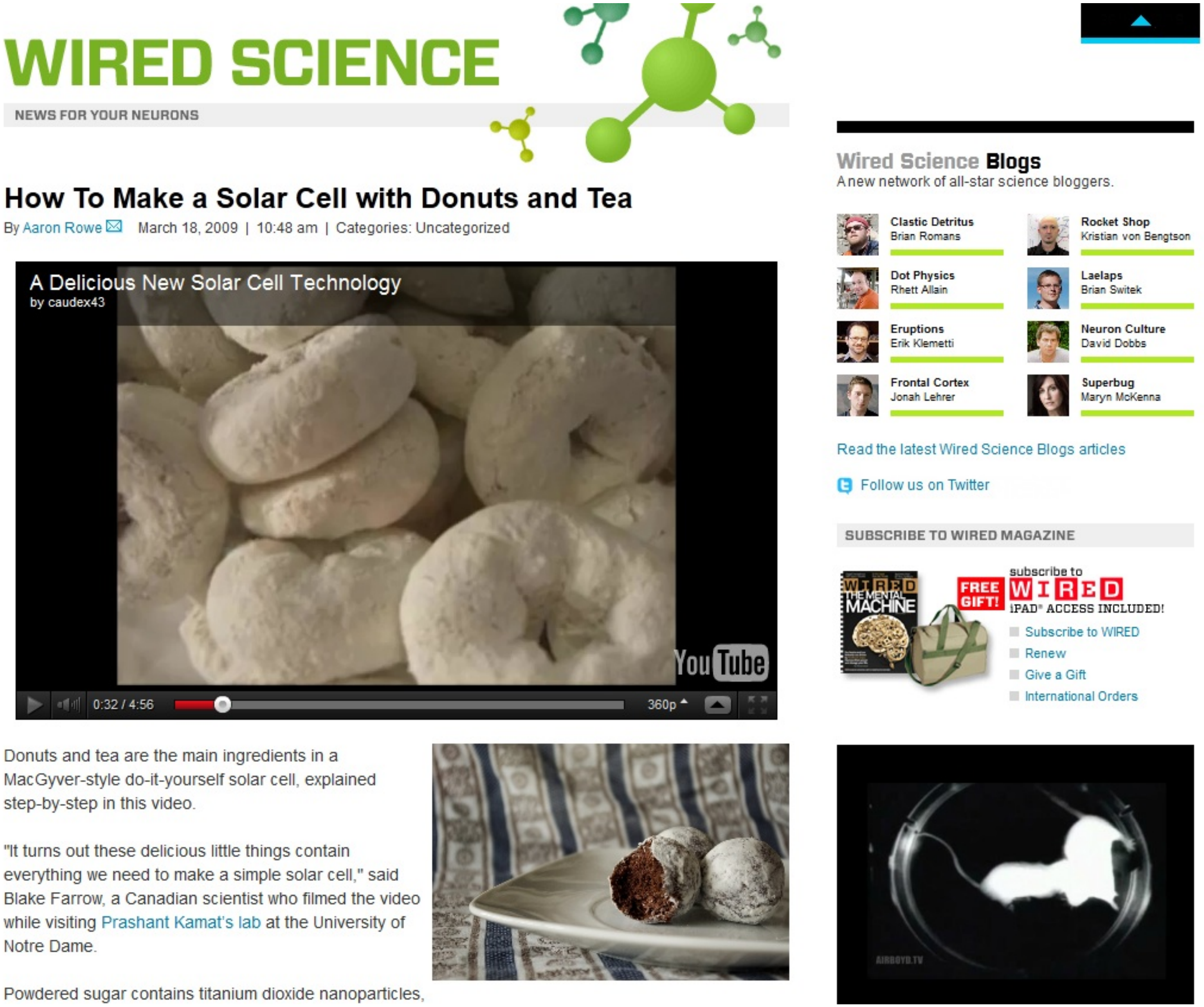


Donuts, Toxicity… and Solar cells

Donuts, Toxicity… and Solar cells



Donuts, Toxicity… and Solar cells



How To Make a Solar Cell with Donuts and Tea

By Aaron Rowe March 18, 2009 | 10:48 am | Categories: Uncategorized

Donuts and tea are the main ingredients in a MacGyver-style do-it-yourself solar cell, explained step-by-step in this video.

"It turns out these delicious little things contain everything we need to make a simple solar cell," said Blake Farrow, a Canadian scientist who filmed the video while visiting Prashant Kamat's lab at the University of Notre Dame.

Powdered sugar contains titanium dioxide nanoparticles,

Wired Science Blogs

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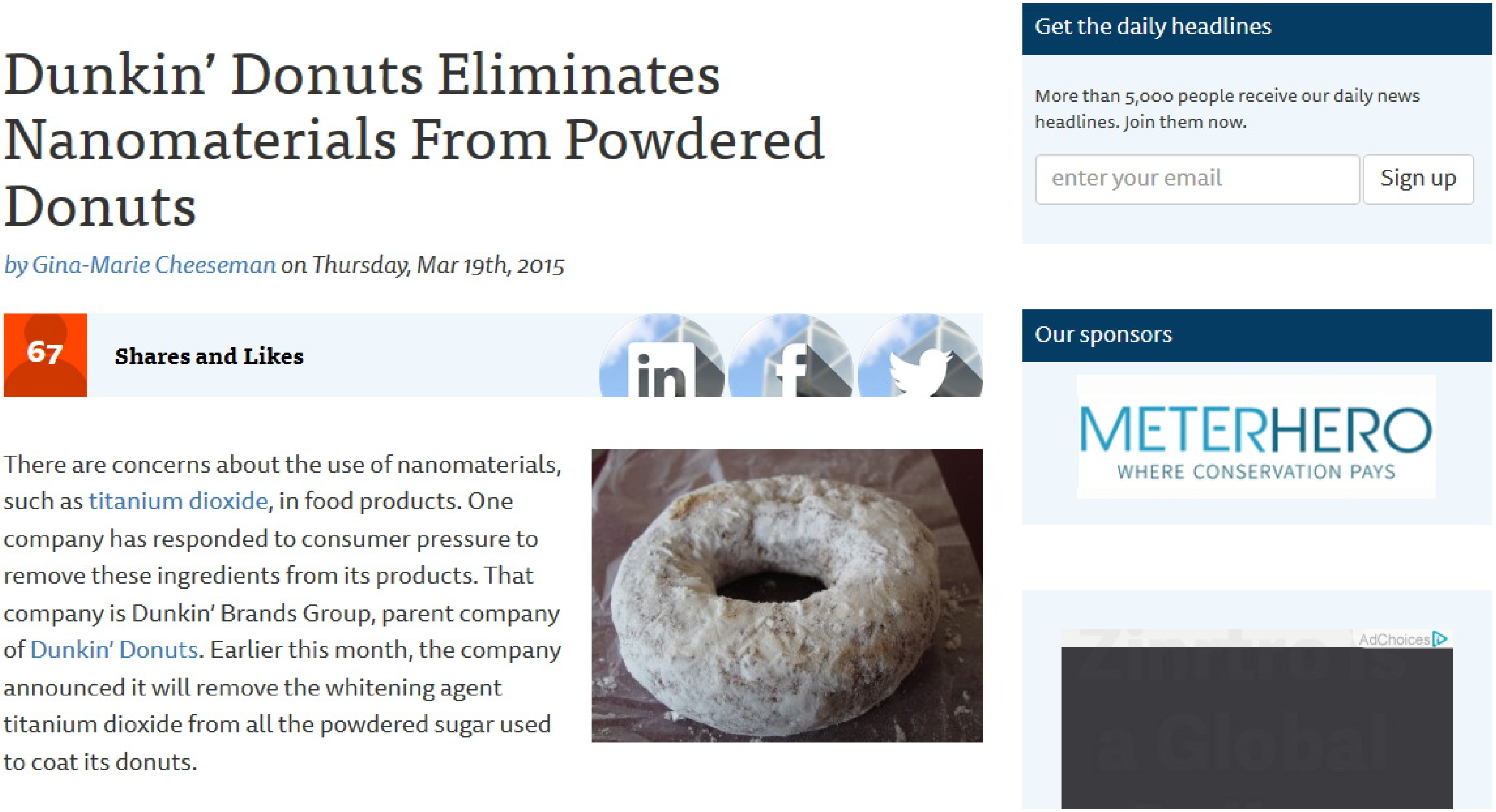

Dunkin' Donuts Eliminates Nanomaterials From Powdered Donuts

by Gina-Marie Cheeseman on Thursday, Mar 19th, 2015

67 **Shares and Likes**

There are concerns about the use of nanomaterials, such as titanium dioxide, in food products. One company has responded to consumer pressure to remove these ingredients from its products. That company is Dunkin' Brands Group, parent company of Dunkin' Donuts. Earlier this month, the company announced it will remove the whitening agent titanium dioxide from all the powdered sugar used to coat its donuts.

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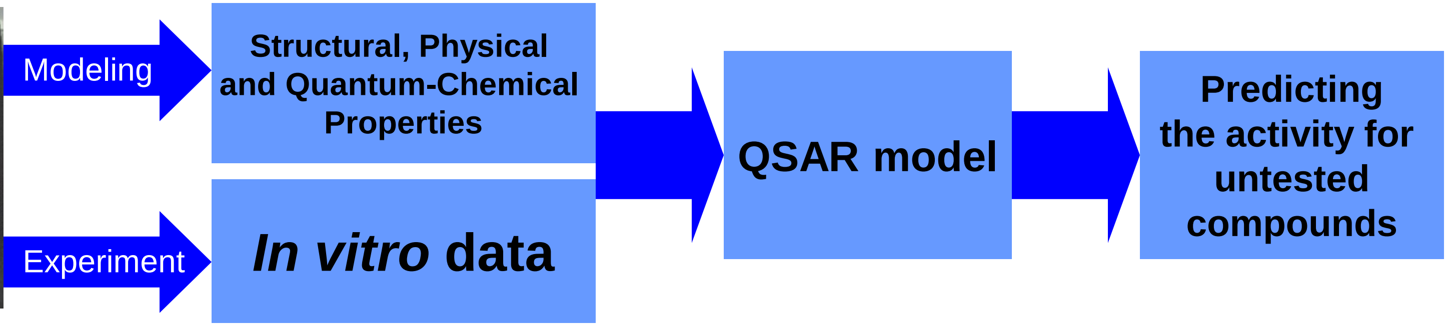
Nano-QSAR
for metal oxide nanoparticles
(Toxicity to *E.coli bacteria*)

Main strategy



Modeling

Experiment

Structural, Physical and Quantum-Chemical Properties

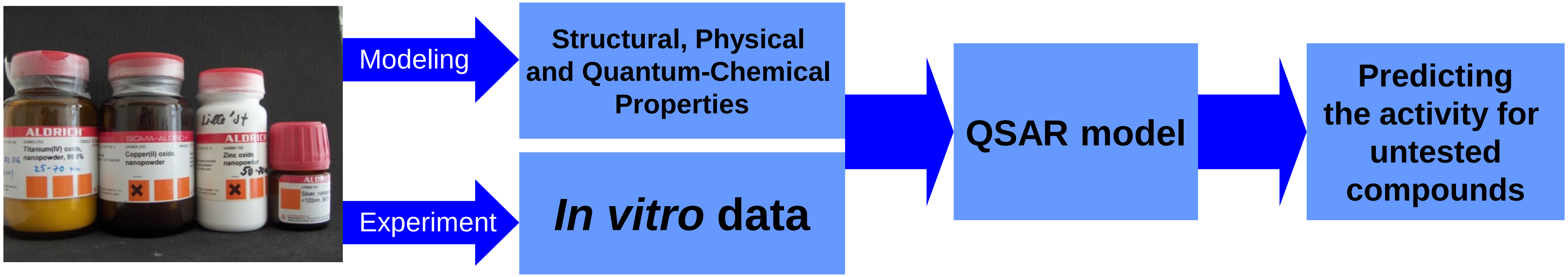
***In vitro* data**

QSAR model

Predicting the activity for untested compounds

Puzyn T., Rasulev B., Gajewicz A., Hu X., Dasari T.P., Michalkova A., Hwang H-M., Toropov A., Leszczynska D. and Leszczynski J., Using nano-QSAR to predict the cytotoxicity of metal oxide nanoparticles, *Nature Nanotechnology*, 2011, 175-178

Main strategy



Modeling → **Structural, Physical and Quantum-Chemical Properties**

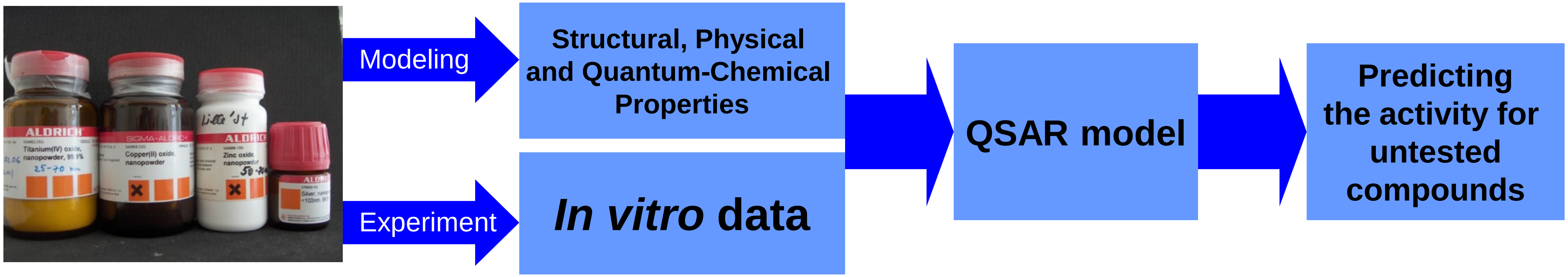
Experiment → ***In vitro* data**

→ **QSAR model** → **Predicting the activity for untested compounds**

We were wondering – why up to date (2009-2010) no studies regarding a series of nanoparticles at the same experiment (same lab, same conditions)?

Puzyn T., Rasulev B., Gajewicz A., Hu X., Dasari T.P., Michalkova A., Hwang H-M., Toropov A., Leszczynska D. and Leszczynski J., Using nano-QSAR to predict the cytotoxicity of metal oxide nanoparticles, *Nature Nanotechnology*, 2011, 175-178

Main strategy



Modeling

Structural, Physical and Quantum-Chemical Properties

Experiment

***In vitro* data**

QSAR model

Predicting the activity for untested compounds

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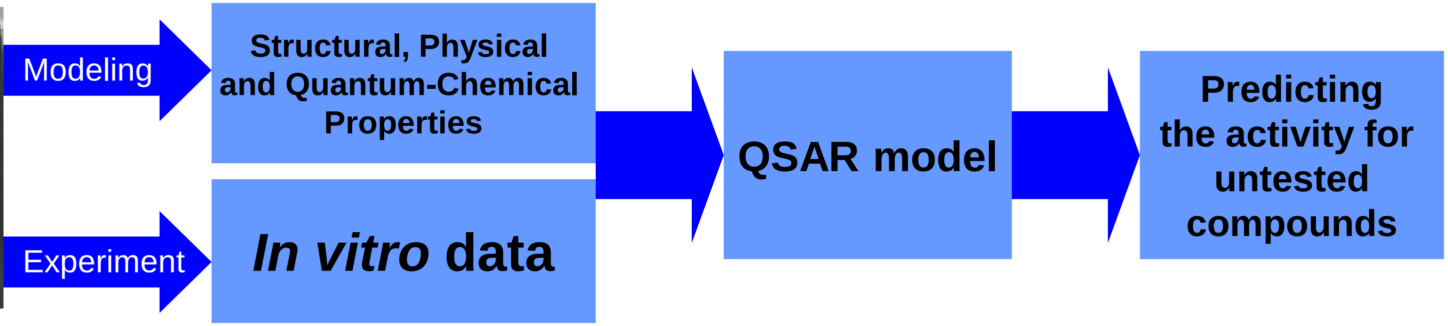
Main strategy



Modeling

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Structural, Physical and Quantum-Chemical Properties

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Modeling → **Structural, Physical and Quantum-Chemical Properties**

Experiment → ***In vitro* data**

→ **QSAR model** → **Predicting the activity for untested compounds**

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Finally, we had a chance to build a **QSAR model** !!!

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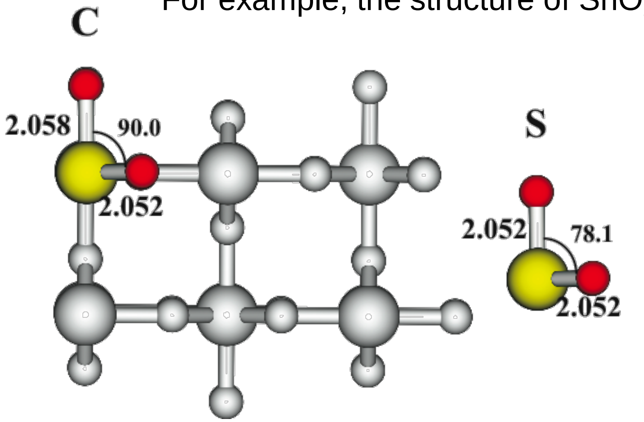
QSAR model of toxicity towards *E.coli* bacteria for nanosized oxides – Quantum-Chemical method + QSAR.

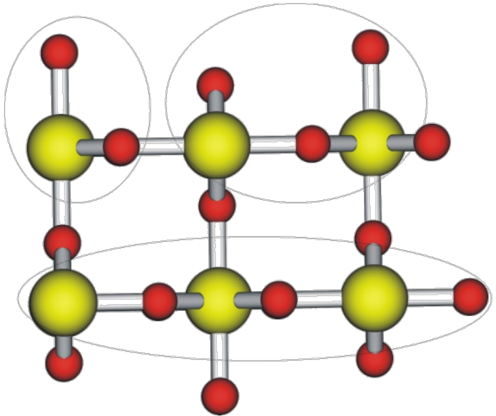
The following metal oxides in nanosized form were selected: ZnO, TiO_2, SnO_2, La_2O_3, Fe_2O_3, CuO, Al_2O_3, Sb_2O_3, V_2O_3, Y_2O_3, In_2O_3, Bi_2O_3, SiO_2 , CoO, NiO, Cr_2O_3, and ZrO_2.

All of these nanosized metal oxides are widely used in many products that present around us. All of them are quite toxic to some extent.

The quantum-chemical methods were applied to find parameters that could be responsible for the toxicity properties for nanosized metal oxides. 12 electronic descriptors were calculated.
As source structures we have used the crystal structures data obtained by X-Ray analysis.

For example, the structure of SnO_2





Ionization potentials (IP1, IP2, IP3) and electron affinities (EA1, EA2, EA3) of, respectively, single (i.e., SnO_2), double (i.e., Sn_2O_4) and triple (Sn_3O_6) stoichiometric fragments cut from the crystal structure:

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Final model with only one parameter.

Nano-QSAR equation, utilizing only one descriptor to predict the cytotoxicity of the metal oxide nanoparticles:

$$\log(1/EC_{50}) = 2.59\ (\pm 0.07) - 0.50\ (\pm 0.07) \cdot \Delta H_{Me+}$$

(n=10, n_{test}=7, **R^2=0.85**, F=45.4, p<0.001, **Q^2_{CVLOO}=0.77**, the externally validated regression coefficient **Q^2_{Ext}=0.83,** RMSEC = 0.20, RMSECV = 0.24, RMSEP = 0.19)

where the descriptor ΔH_{Me+} represents the enthalpy of formation of a gaseous cation having the same oxidation state as that in the metal oxide structure.

$$Me\ (s) \rightarrow Me^{n+}\ (g) + n \cdot \bar{e} \rightarrow \Delta H_{Me+}$$

The descriptors were calculated using quantum-chemical methods. Since from a quantum-mechanical point of view, the calculations of nanoparticles of 15-90 nm size (those used in the experiments) were not feasible (too large systems) it was necessary to maximally simplify the structural models utilized to calculate the descriptors.

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Splitting a dataset

Training Set	Validation set 1	Validation Set 2
ZnO CuO Y_2O_3 Bi_2O_3 In_2O_3 Al_2O_3 Fe_2O_3 SiO_2 SnO_2 TiO_2	V_2O_3 Sb_2O_3 ZrO_2	CoO NiO Cr_2O_3 La_2O_3

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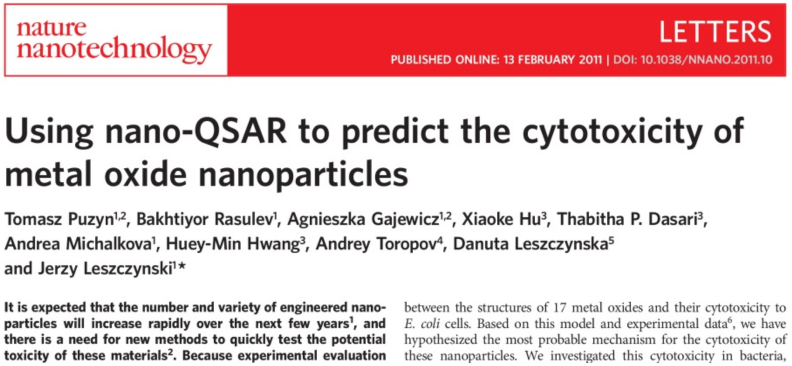
Cytotoxicity nano-QSAR model for MeOx nanomaterials

Nano-QSAR model, which successfully predicted the cytotoxicity of the metal oxide nanoparticles

$$\log(1/EC_{50}) = 2.59 - 0.50\cdot\Delta H_{Me+}$$



nature nanotechnology

LETTERS

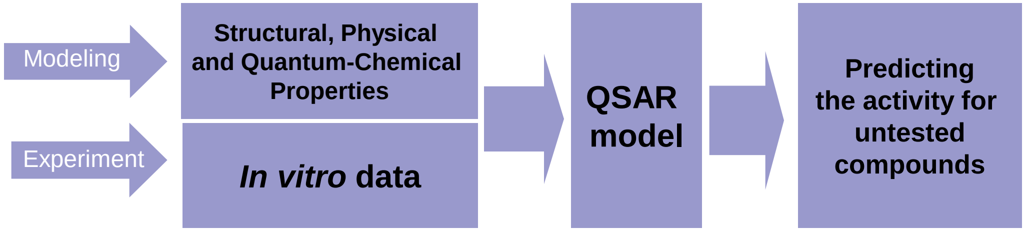
PUBLISHED ONLINE: 13 FEBRUARY 2011 | DOI: 10.1038/NNANO.2011.10

Using nano-QSAR to predict the cytotoxicity of metal oxide nanoparticles

Tomasz Puzyn[1,2], Bakhtiyor Rasulev[1], Agnieszka Gajewicz[1,2], Xiaoke Hu[3], Thabitha P. Dasari[3], Andrea Michalkova[1], Huey-Min Hwang[3], Andrey Toropov[4], Danuta Leszczynska[5] and Jerzy Leszczynski[1]*

It is expected that the number and variety of engineered nanoparticles will increase rapidly over the next few years[1], and there is a need for new methods to quickly test the potential toxicity of these materials[2]. Because experimental evaluation

between the structures of 17 metal oxides and their cytotoxicity to *E. coli* cells. Based on this model and experimental data[6], we have hypothesized the most probable mechanism for the cytotoxicity of these nanoparticles. We investigated this cytotoxicity in bacteria,



Modeling

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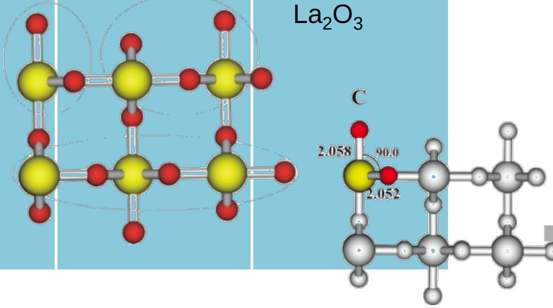
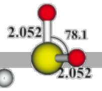
***In vitro* data**

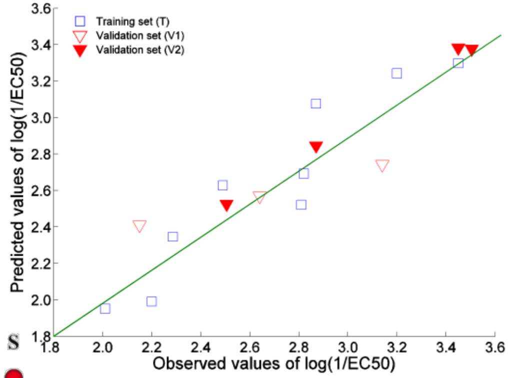
QSAR model

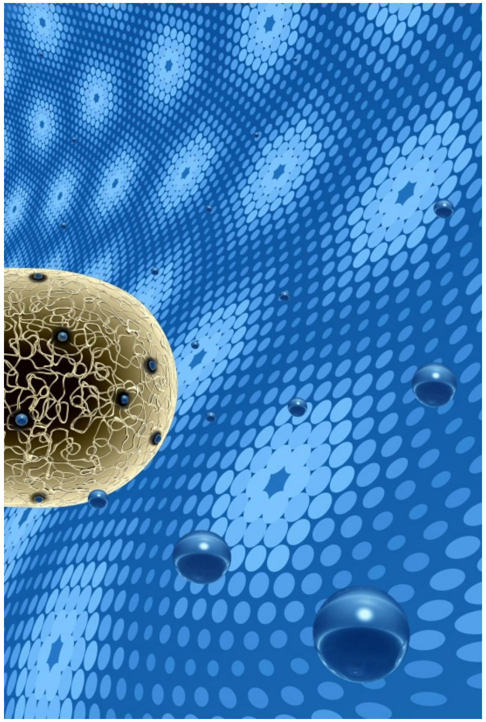
Predicting the activity for untested compounds

Training Set	Validation set 1	Validation Set 2
ZnO	V_2O_3	CoO
CuO	Sb_2O_3	NiO
Y_2O_3	ZrO_2	Cr_2O_3
Bi_2O_3		La_2O_3
In_2O_3		
Al_2O_3		
Fe_2O_3		
SiO_2		
SnO_2		
TiO_2		









E. coli **and nanoparticle surface**

Results – Cytotoxicity trend

$$\log\left(1/LC_{50}\right) = 2.59(\pm 0.07) - 0.50(\pm 0.07)\cdot \Delta H_{Me+}$$

Me^{2+}

$$Me_{(s)} \rightarrow Me^{n+}_{(g)} + n\cdot e$$

$$\Delta H_{Me+} = \Delta H_{S} + \sum_{i=1}^{n} IP_i$$

Me^{3+}

$$a\cdot Me^{n+}(g) + b\cdot O^{2-}(g) \rightarrow Me_aO_b(s)$$

Me^{4+}

$$e + O_2 \longrightarrow O_2^-$$

$$O_2^- + 2H^+ + e \longrightarrow H_2O_2$$

$$H_2O_2 + O_2^- \longrightarrow OH^\bullet + OH^- + O_2$$

Puzyn T., Rasulev B., Gajewicz A., Hu X., Dasari T.P., Michalkova A., Hwang H-M., Toropov A., Leszczynska D. and Leszczynski J., Using nano-QSAR to predict the cytotoxicity of metal oxide nanoparticles, *Nature Nanotechnology*, 2011, 175-178

The way to cover prediction for cytotoxicity for all existing nano-sized metal oxides by using neural network method

The counter propagation artificial neural network (CP ANN) models for prediction of cytotoxicity of MeOx NPs for data sets of 17, 36 and 72 metal oxides were employed in the study..

The following metal oxides in nanosized form were selected to train the model: ZnO, TiO_2, SnO_2, La_2O_3, Fe_2O_3, CuO, Al_2O_3, Sb_2O_3, V_2O_3, Y_2O_3, In_2O_3, Bi_2O_3, SiO_2 , CoO, NiO, Cr_2O_3, and ZrO_2.

The cytotoxicity model for studied metal oxide NPs was taking into account:

(i) *χ-metal electronegativity (EN) by Pauling scale,* and composition of metal oxides characterized by

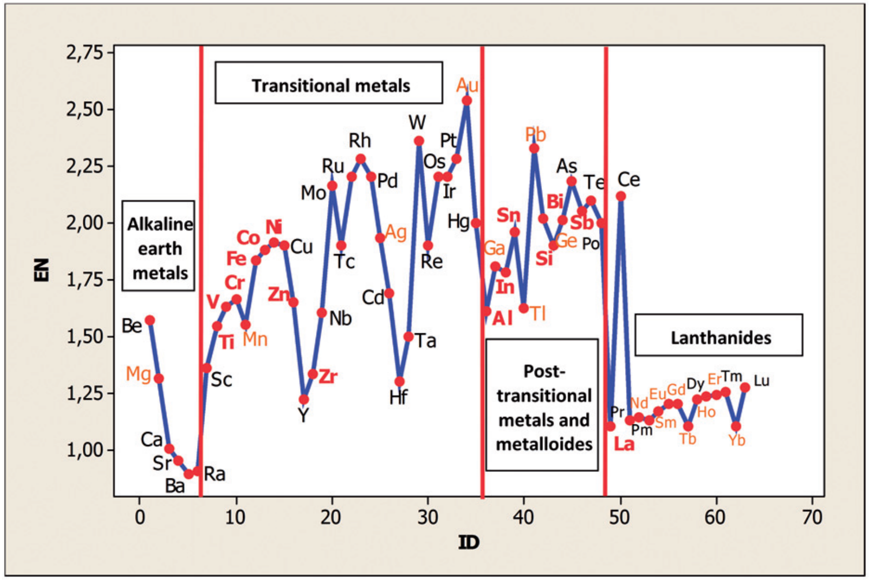
(ii) *number of metal atoms in oxide,*

(iii) *number of oxygen atoms in oxide,*

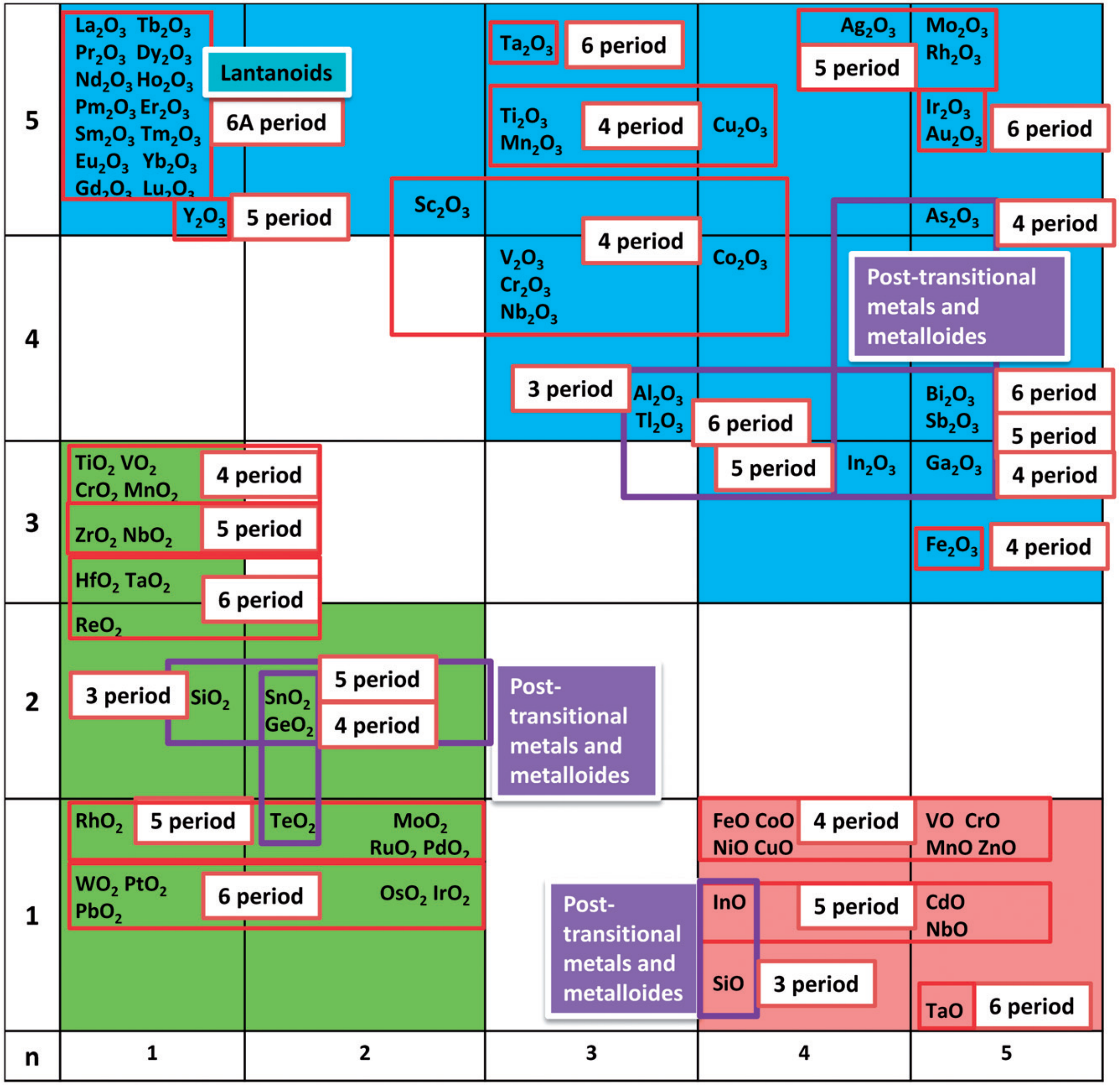
(iv) *charge of metal cation in oxide.*

Quantitative CP ANN models showed a good prediction power of models with the leave one out Q^2 in the range of 0.83–0.92. The categorical CP ANN models were capable to predict class of cytotoxicity with accuracy equal to 1.

The methodology is expected to be useful for potential hazard assessment of MeO_x NPs and prioritization for further testing and risk assessment.



N Fjodorova, M Novic, A Gajewicz, B Rasulev, The way to cover prediction for cytotoxicity for all existing nano-sized metal oxides by using neural network method, *Nanotoxicology,* 2017, 11(4), 475-483



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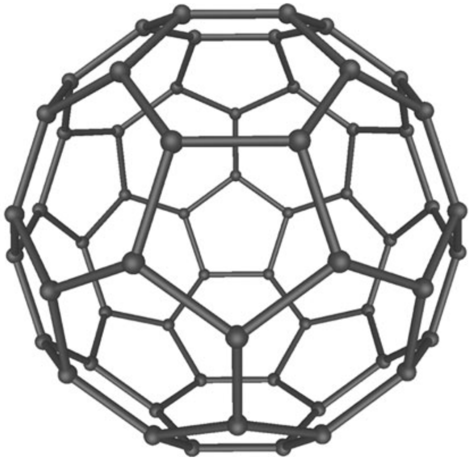
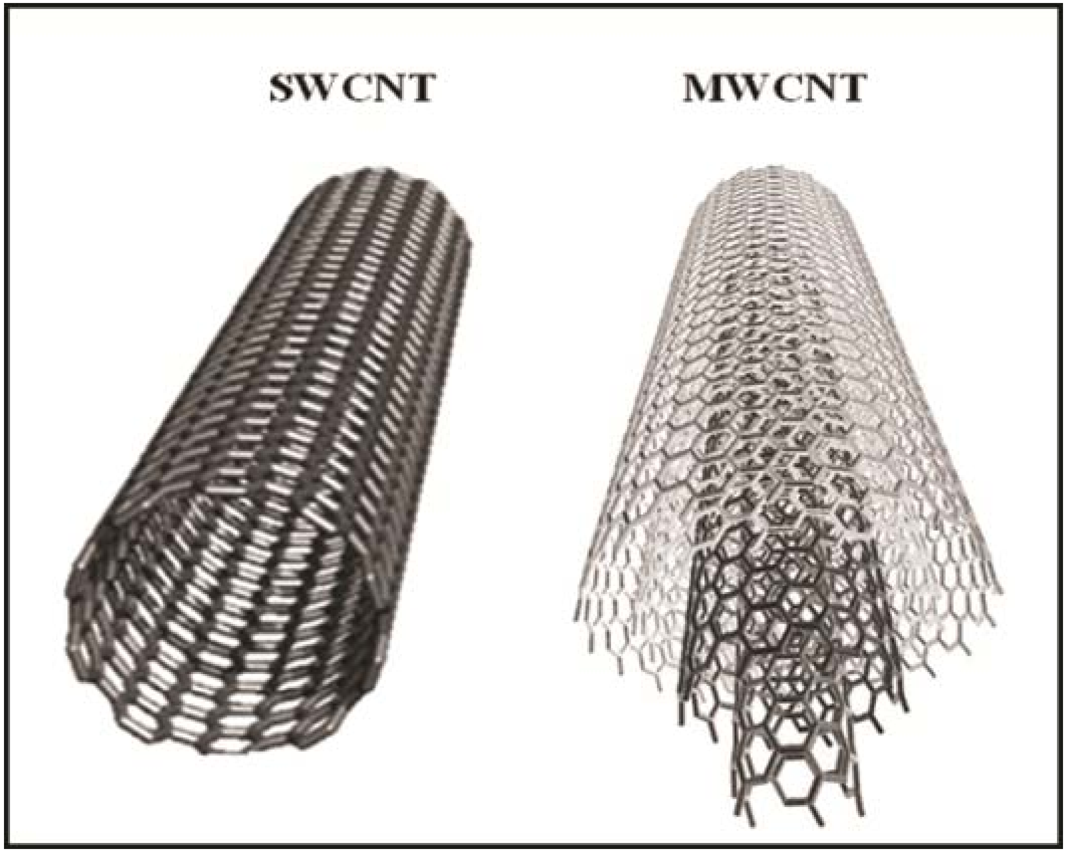
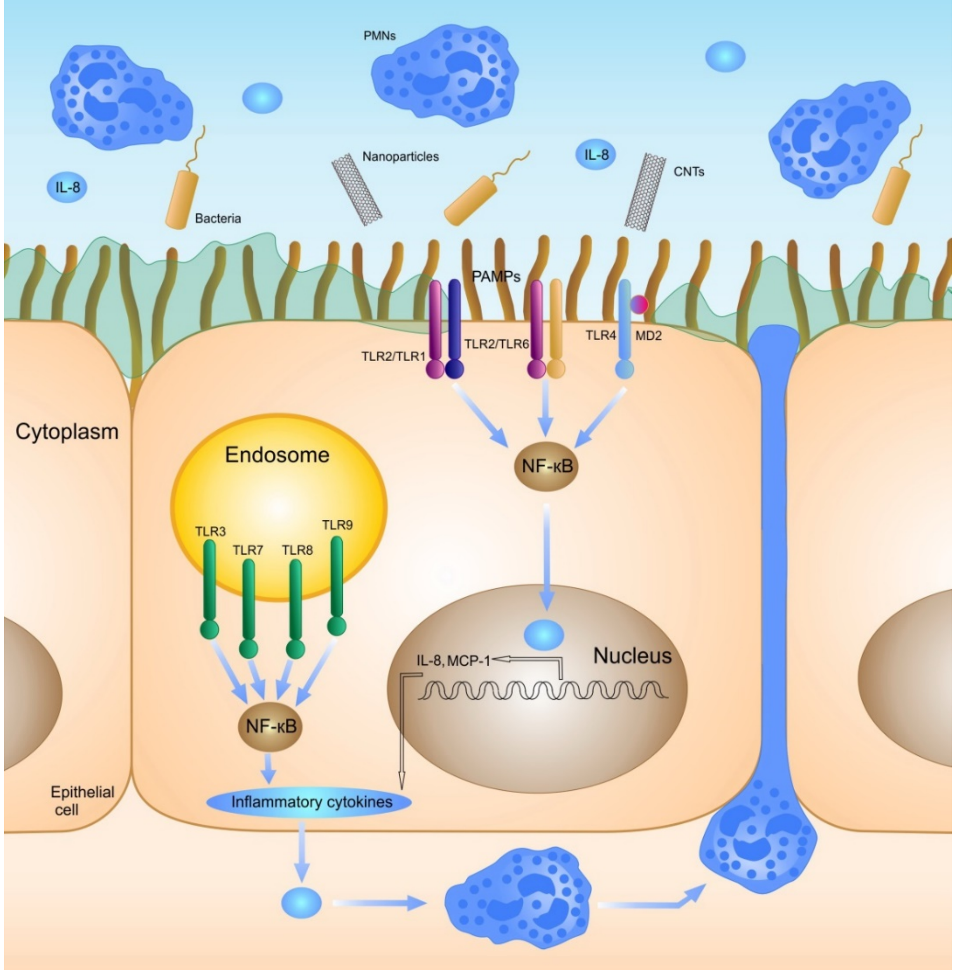
Carbon nanostructures fullerene C60 and carbon nanotubes (CNTs)



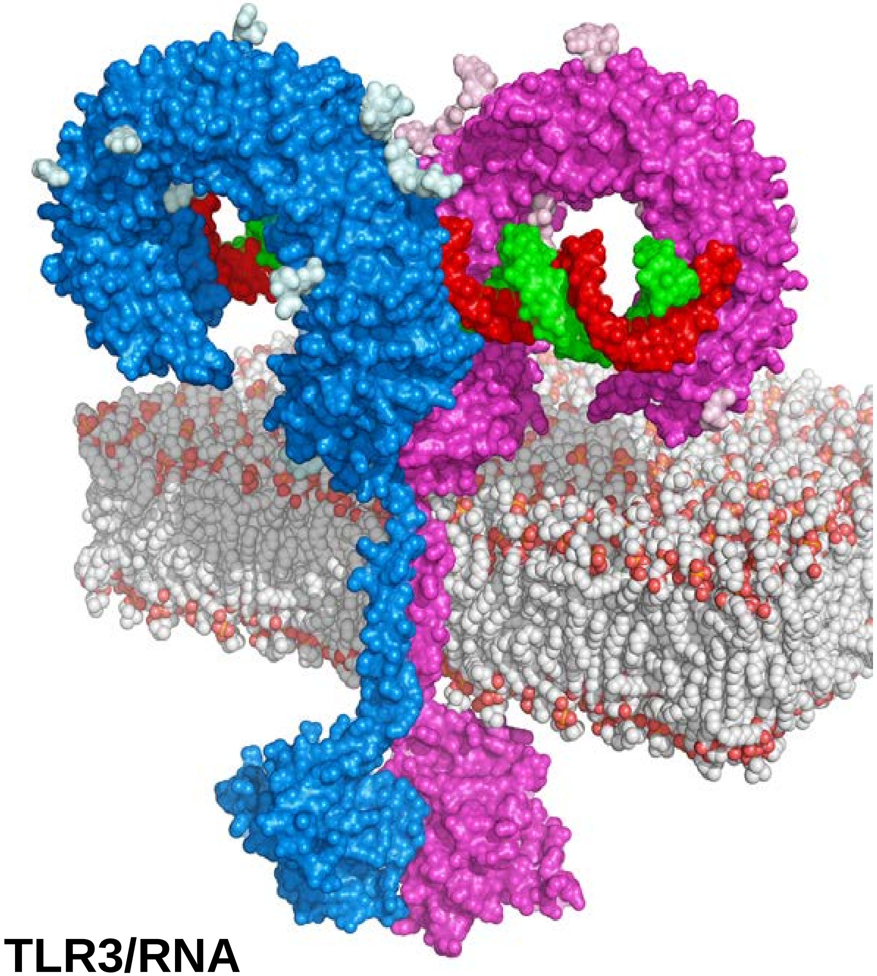


Immunotoxicity of nanoparticles: CNTs and fullerenes might be recognized as pathogens by Toll-like receptors



M. Turabekova, B. Rasulev, M. Theodor, J. Jacksman, D. Leszczynska, J. Leszczynski, *Nanoscale*, 2014

Pattern Recognition Receptors signaling pathway



TLR3/RNA

- ✓ Macrophages play a vital role in the immune system.
- ✓ and have pattern recognition receptors (PRRs) to identify pathogens.
- ✓ PRRs are represented by membrane-associated Toll-like receptors (TLRs) and cytoplasmic Node-like receptors (NLRs).
- ✓ Each TLR and NLR recognize specific, conserved pathogen-associated molecular patterns (PAMPs) present in microbial proteins, nucleic acids, lipids, and carbohydrates.
- ✓ These PAMP-containing molecules act as ligands to trigger PRR-dependent intracellular signaling pathways that ultimately induce the expression of pro-inflammatory and antiviral cytokines.

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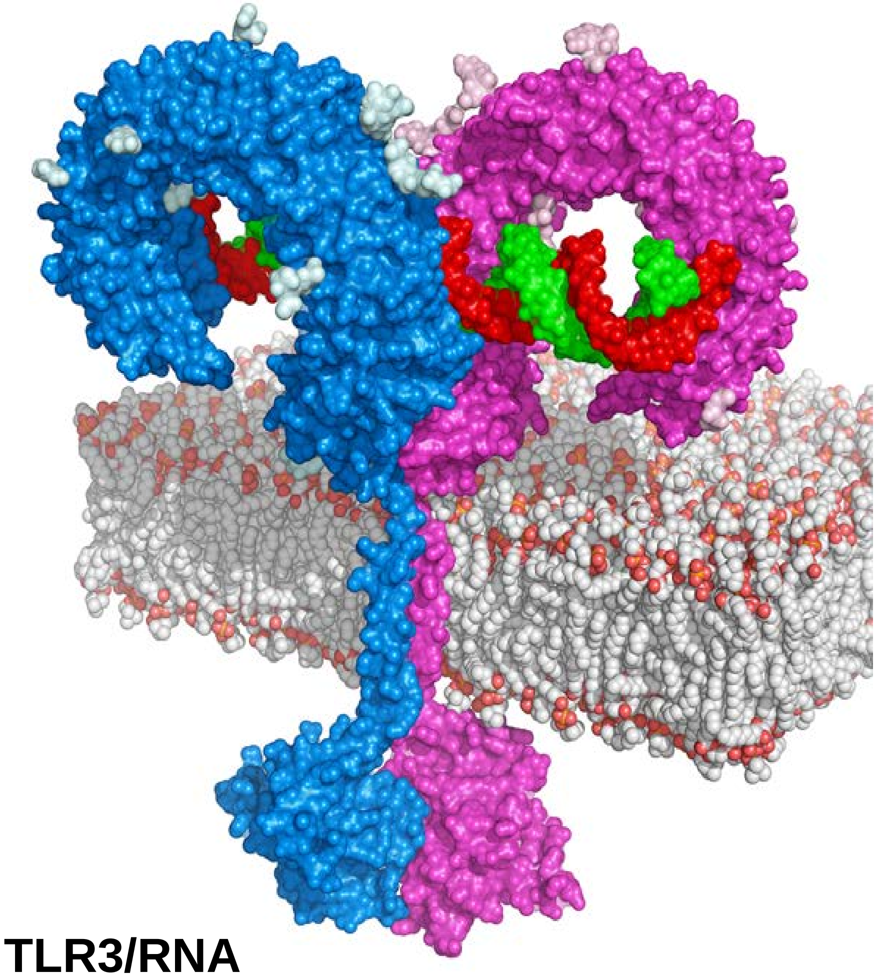

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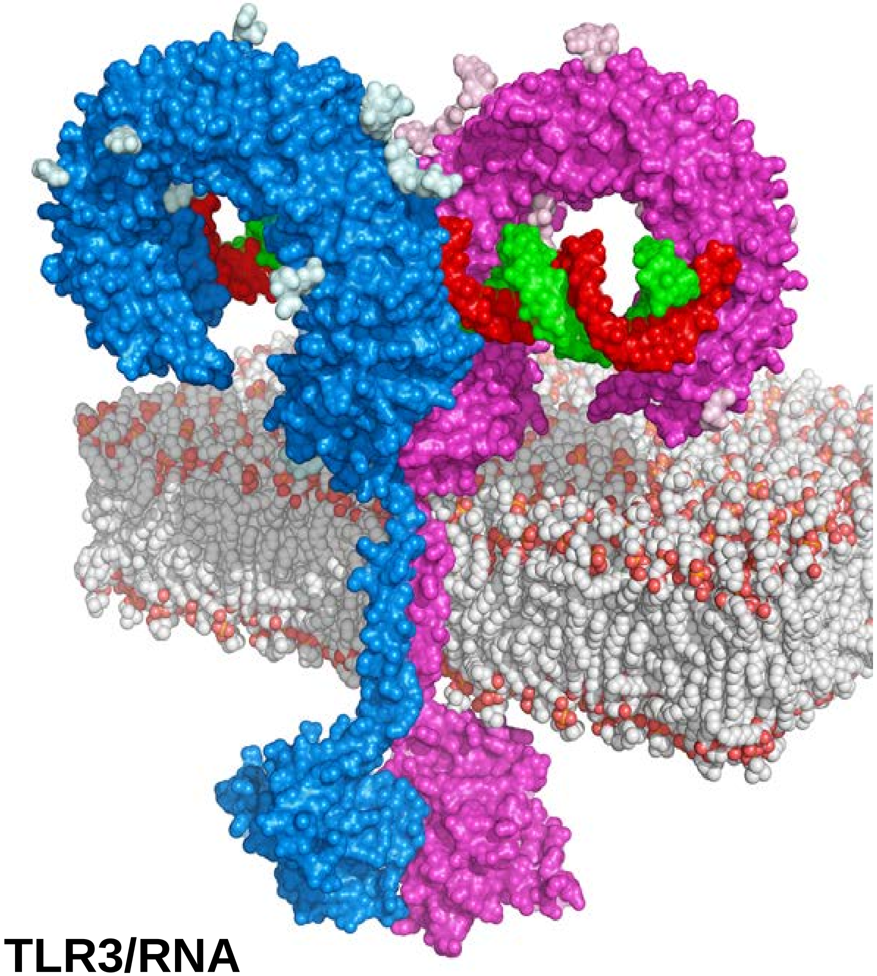

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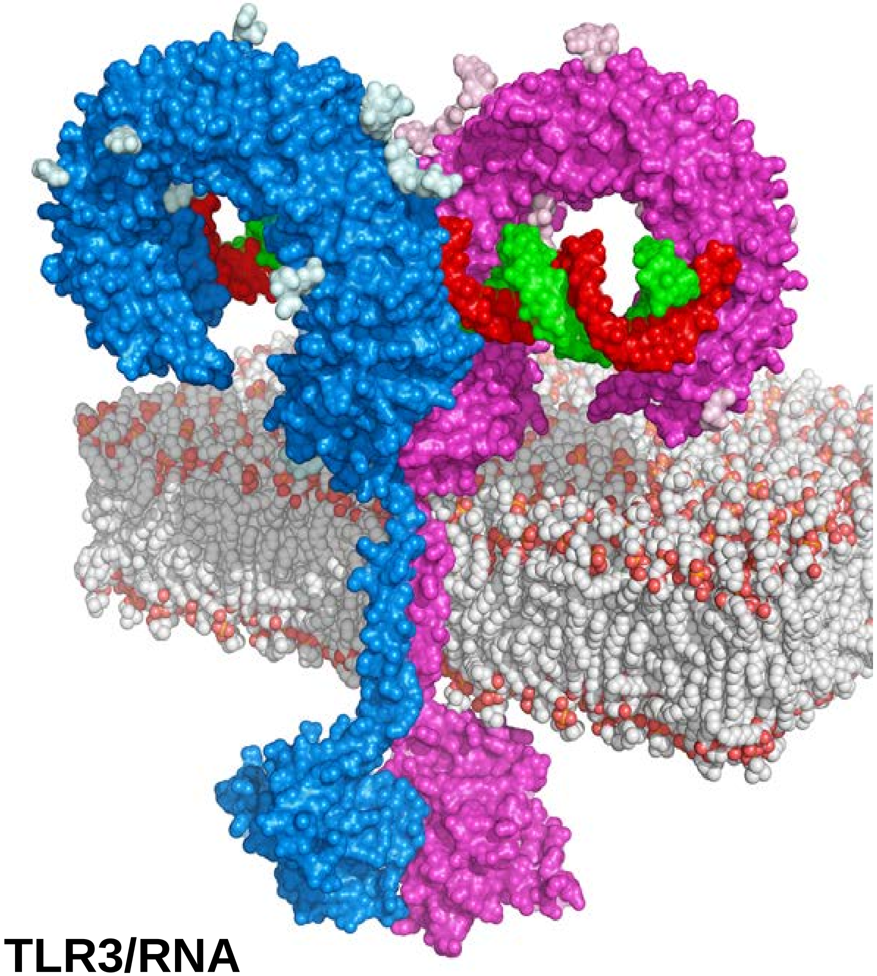
↓

TLRs act as the forefront PAMPs (carbon nanoparticles) recognizers in macrophages.

Pattern Recognition Receptors signaling pathway



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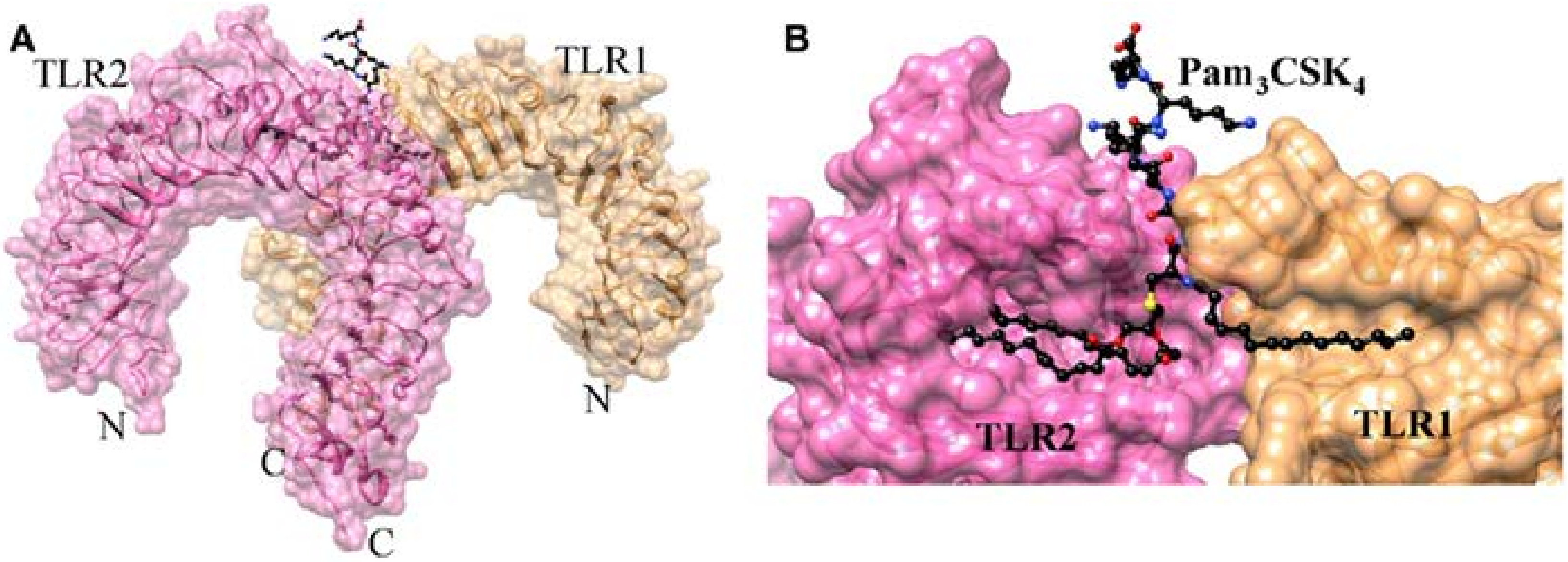
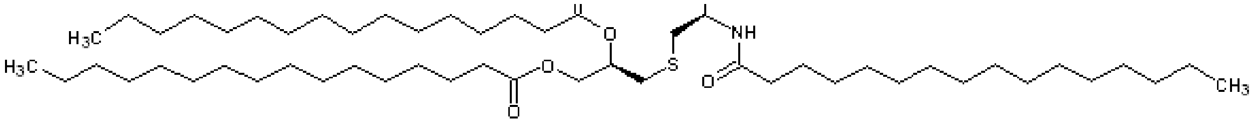
TLRs act as the forefront PAMPs (carbon nanoparticles) recognizers in macrophages.

Toll-like Receptors: TLR1/TLR2

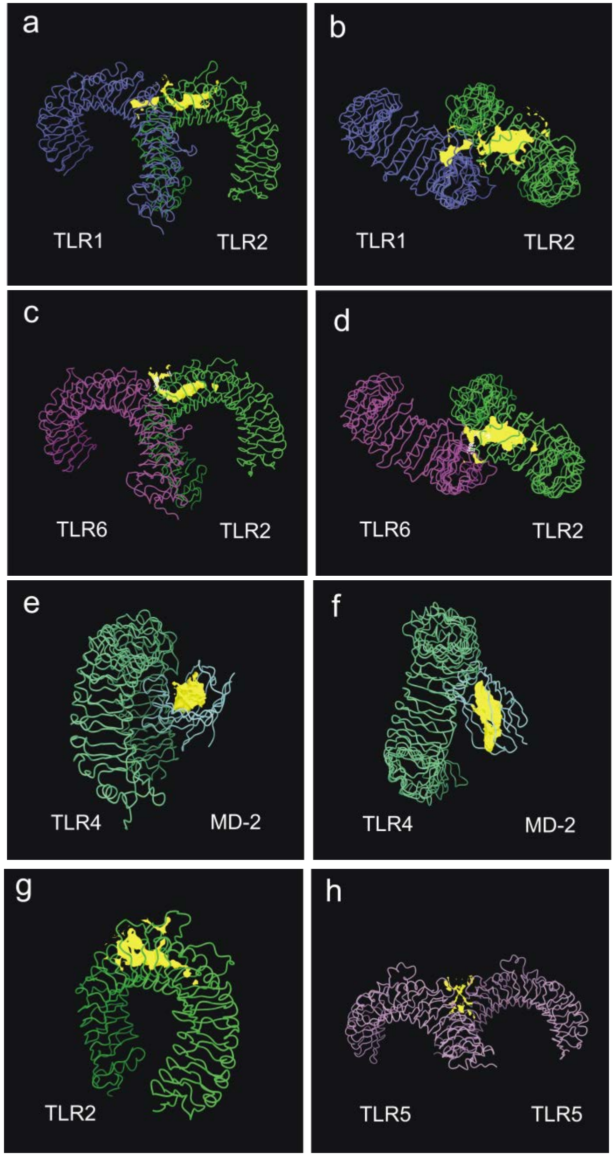
TLRs have evolved to recognize PAMPs expressed by the broad classes of pathogens (e.g. viruses, bacteria, and fungi).
High specificity of TLRs helps them to recognize well-conserved features in pathogens, including bacterial cell-surface **LIPOPEPTIDES**





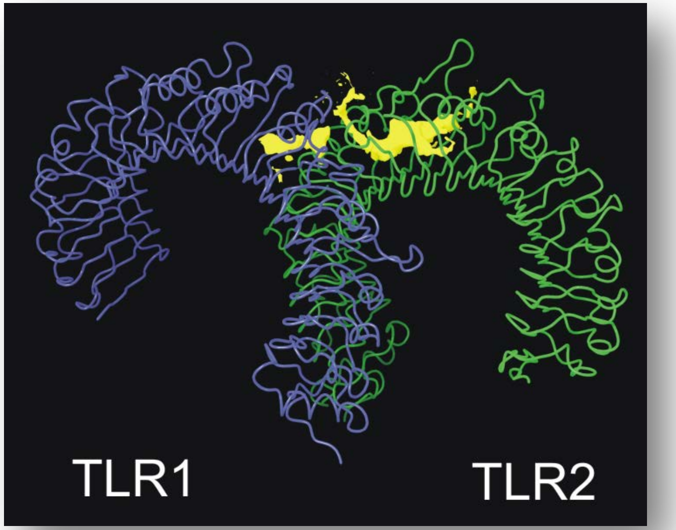
M.S. Jin et all., Cell, 130 (2007) 1071-1082.

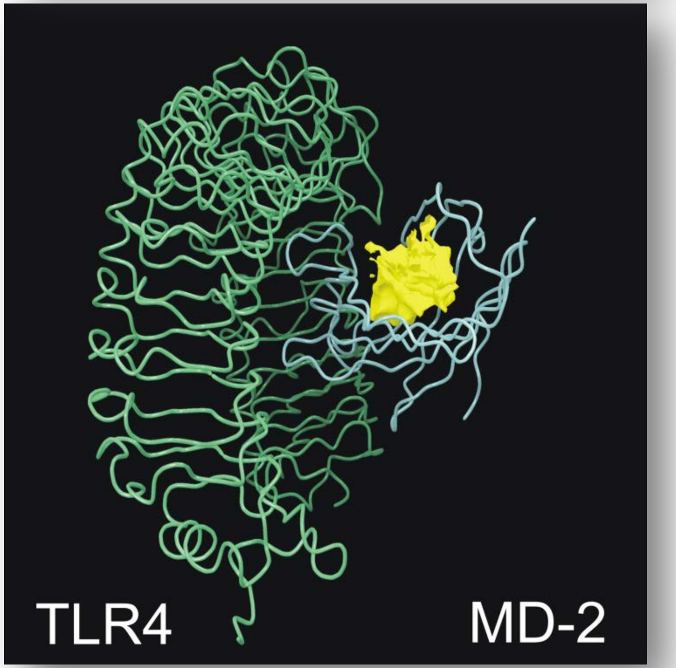
Identification of Hydrophobic Binding Sites



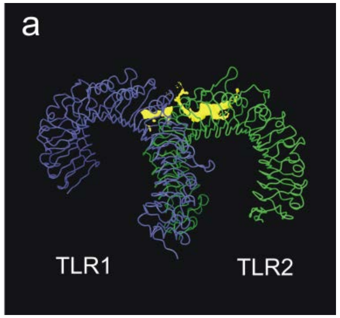
Site Volume, Å
1035.86
637.98
596.82
1182.66*

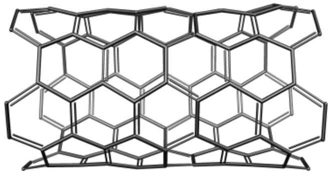
* TLR2



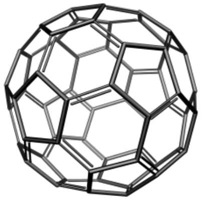


Glide XP docking: TLR1/TLR2





-15.460



-8.747

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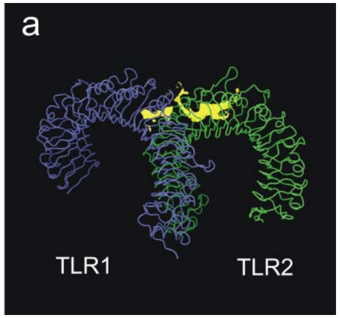

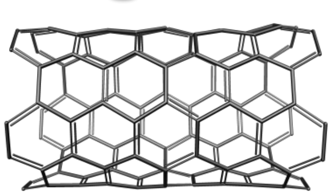

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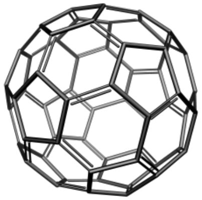

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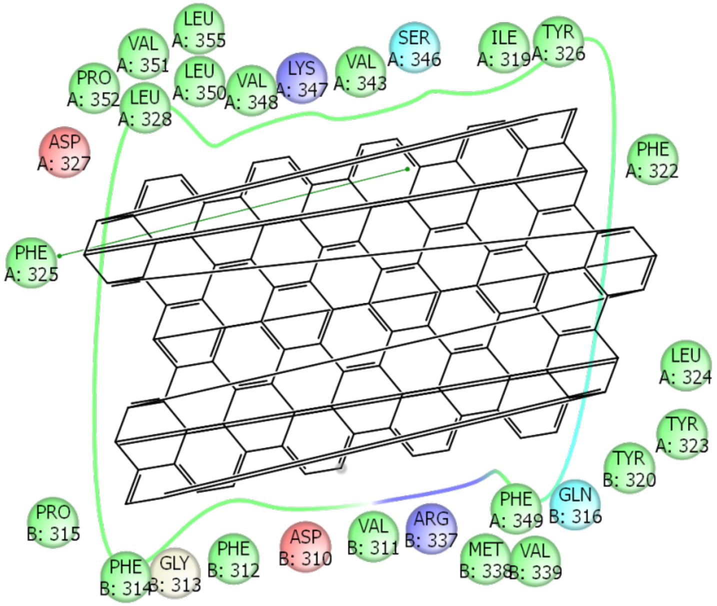
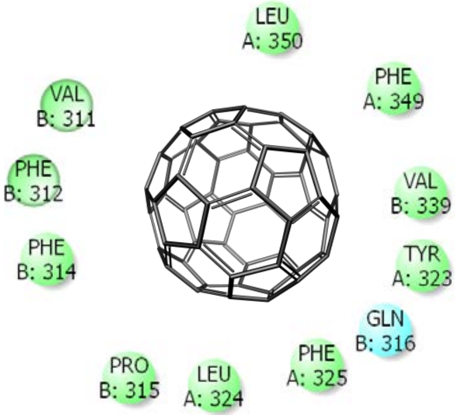
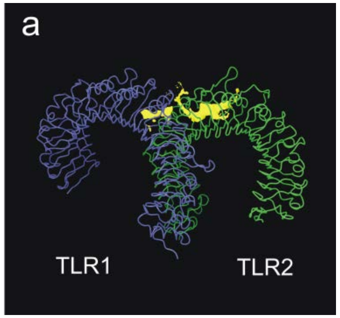
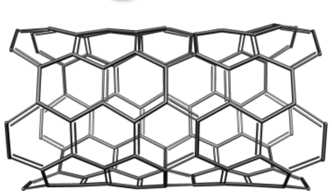




- ● Charged (negative)
- ● Charged (positive)
- ● Polar
- ● Hydrophobic
- ● Glycine
- ● Metal
- H₂O Water
- ● Hydration site
- ☒ Displaced hydration site
- ↔ π-π stacking
- → π-cation
- → H-bond (backbone)
- → H-bond (side chain)
- → Metal coordination
- ○ Solvent exposure

Glide XP docking: TLR1/TLR2





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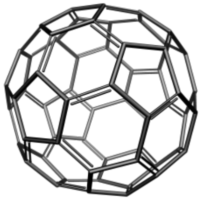

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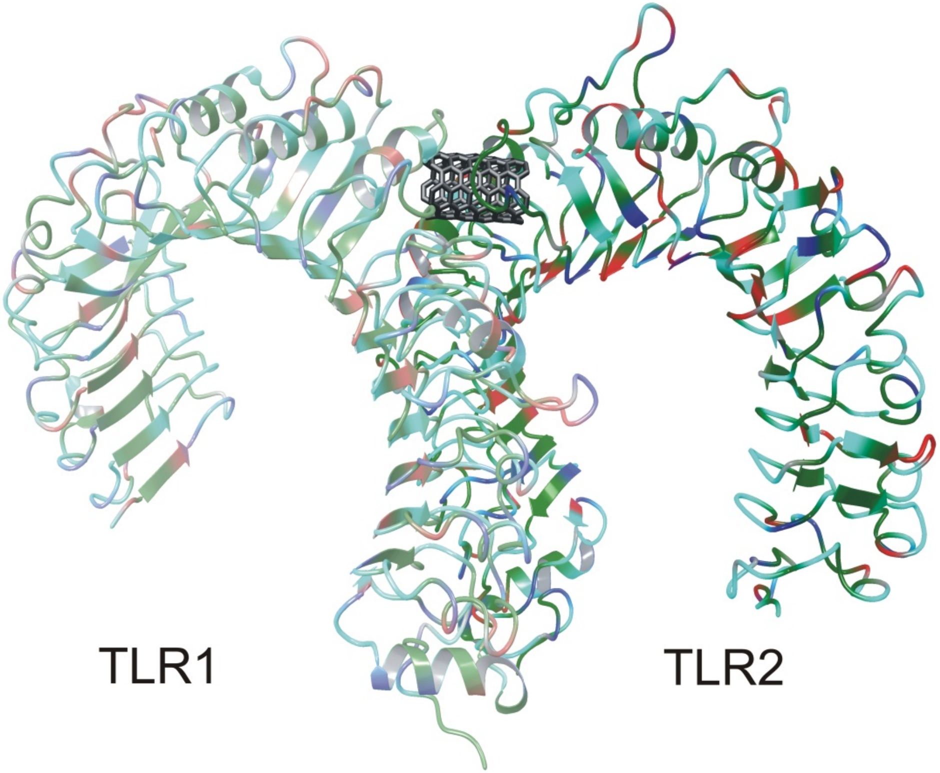
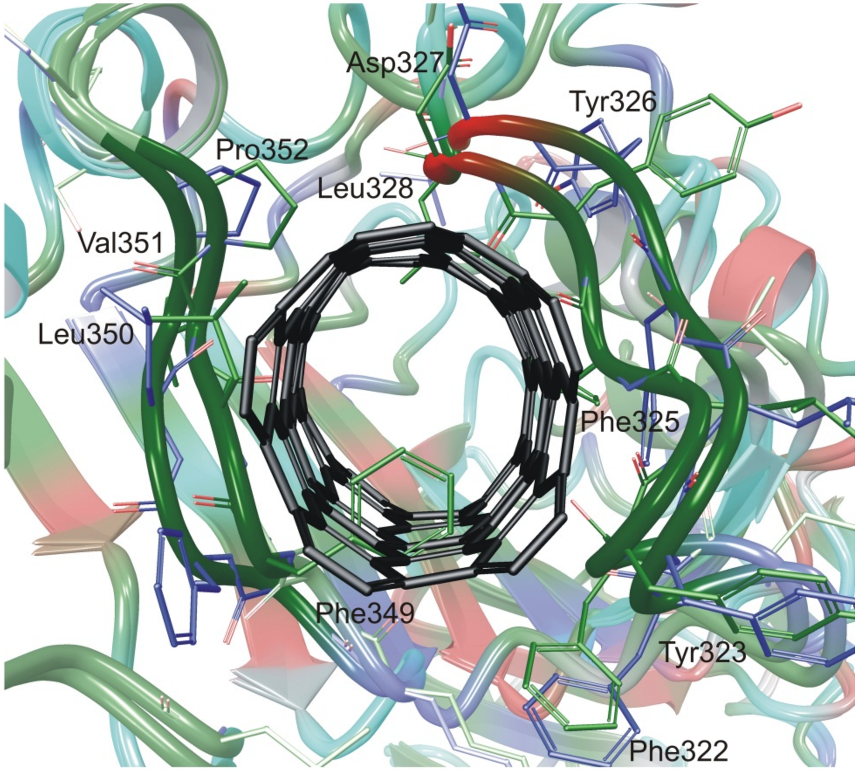
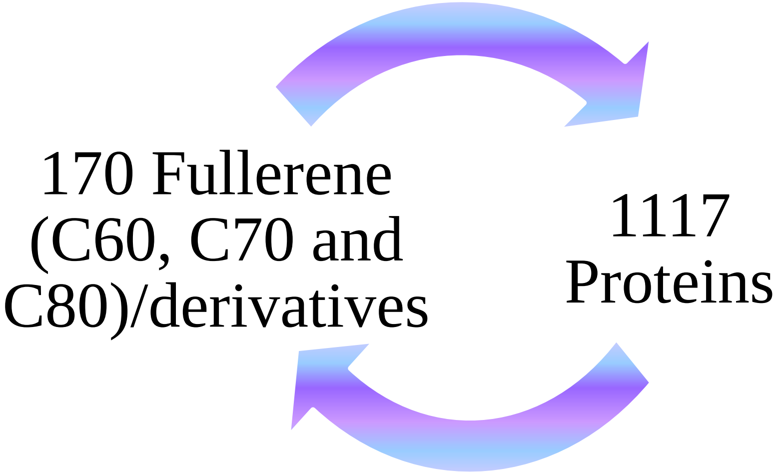


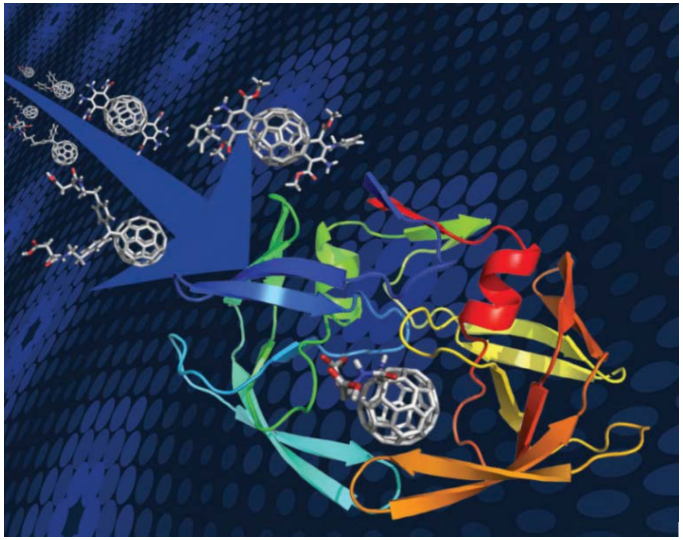


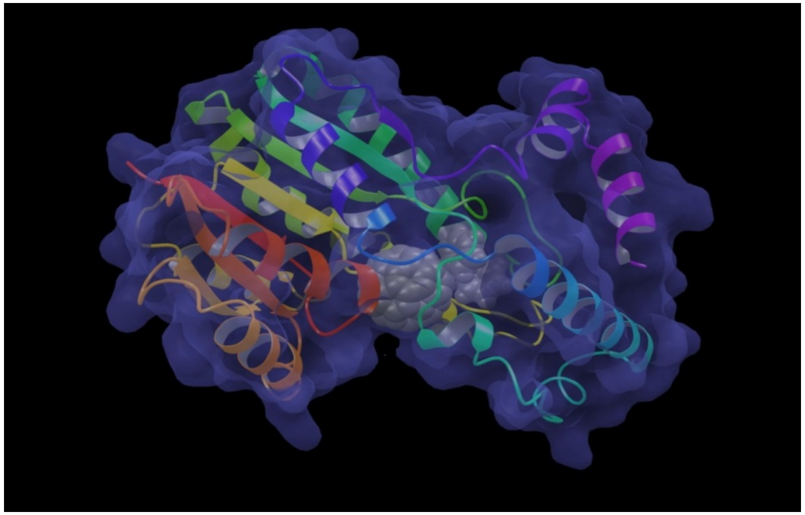
Inhibitors or toxins? Large library target-specific screening of fullerene-based nanoparticles

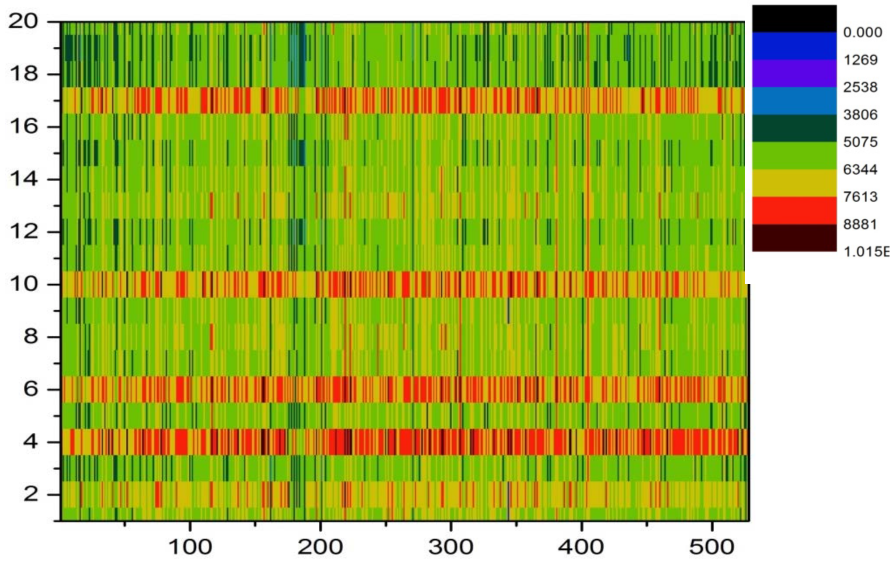


170 Fullerene (C60, C70 and C80)/derivatives

1117 Proteins







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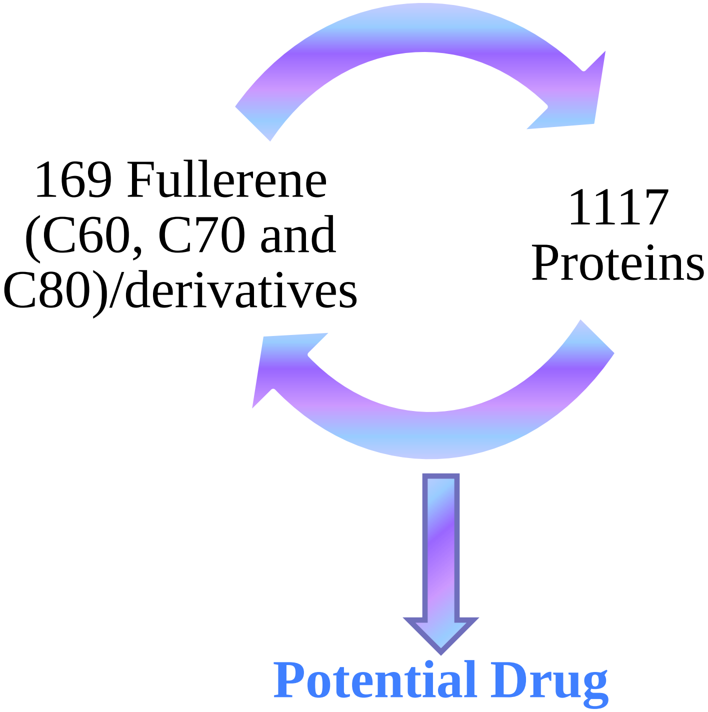
Ligand-Protein Inverse Docking

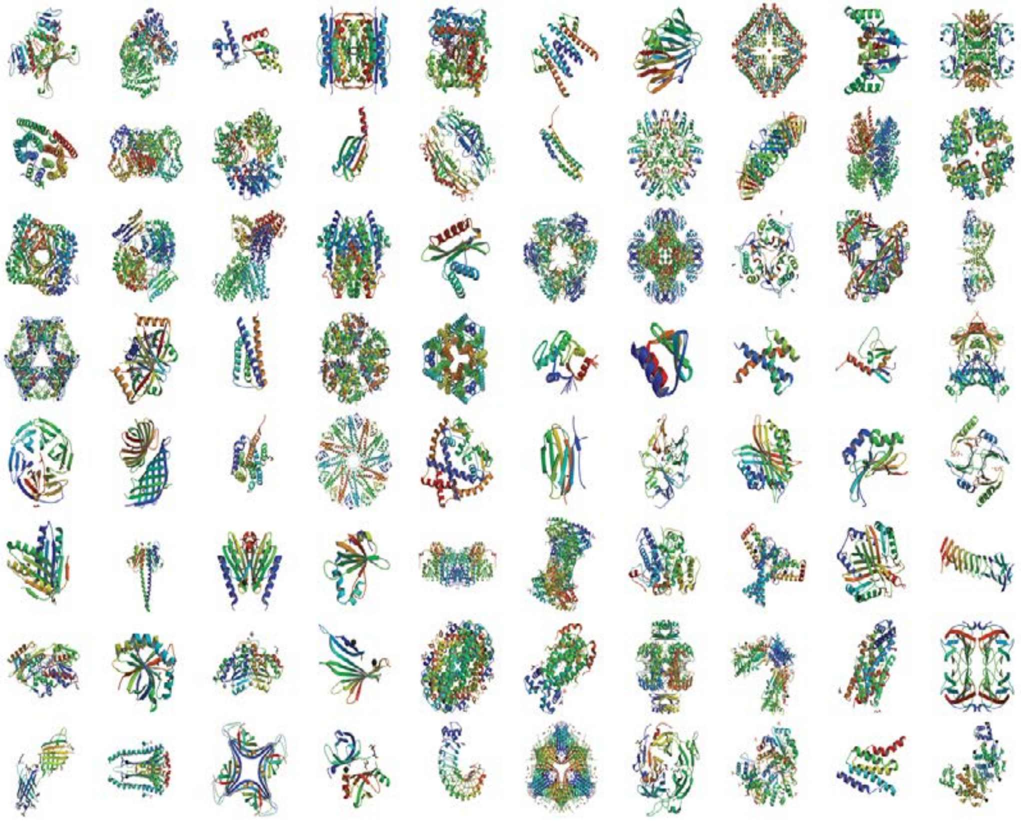
We’ve selected existing fullerene derivatives and decided to dock all possible proteins related to diseases.



169 Fullerene (C60, C70 and C80)/derivatives

1117 Proteins

Potential Drug



Series of Proteins

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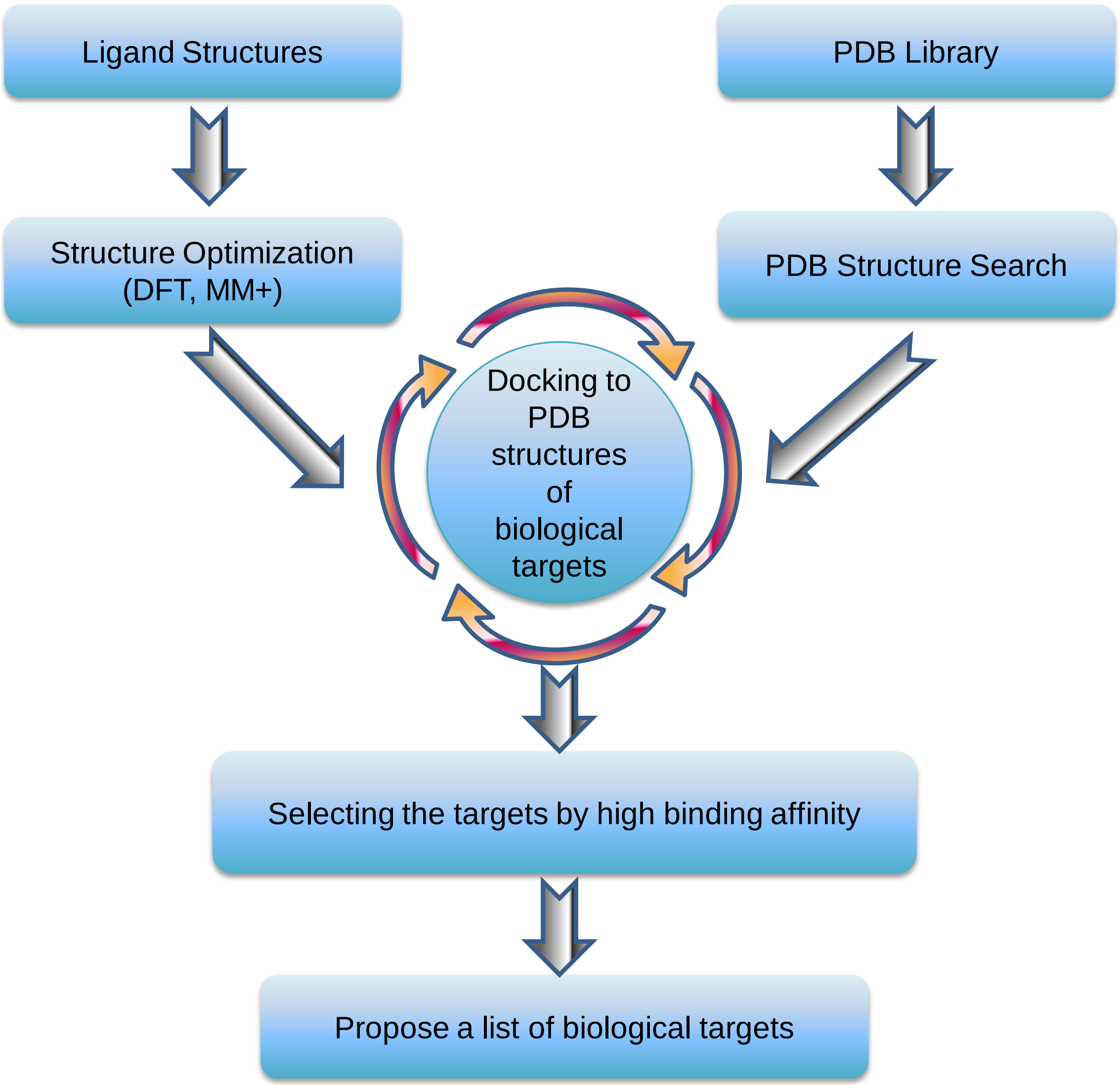
Overall Schematic Diagram of the Study



Ligand Structures

Structure Optimization
(DFT, MM+)

PDB Library

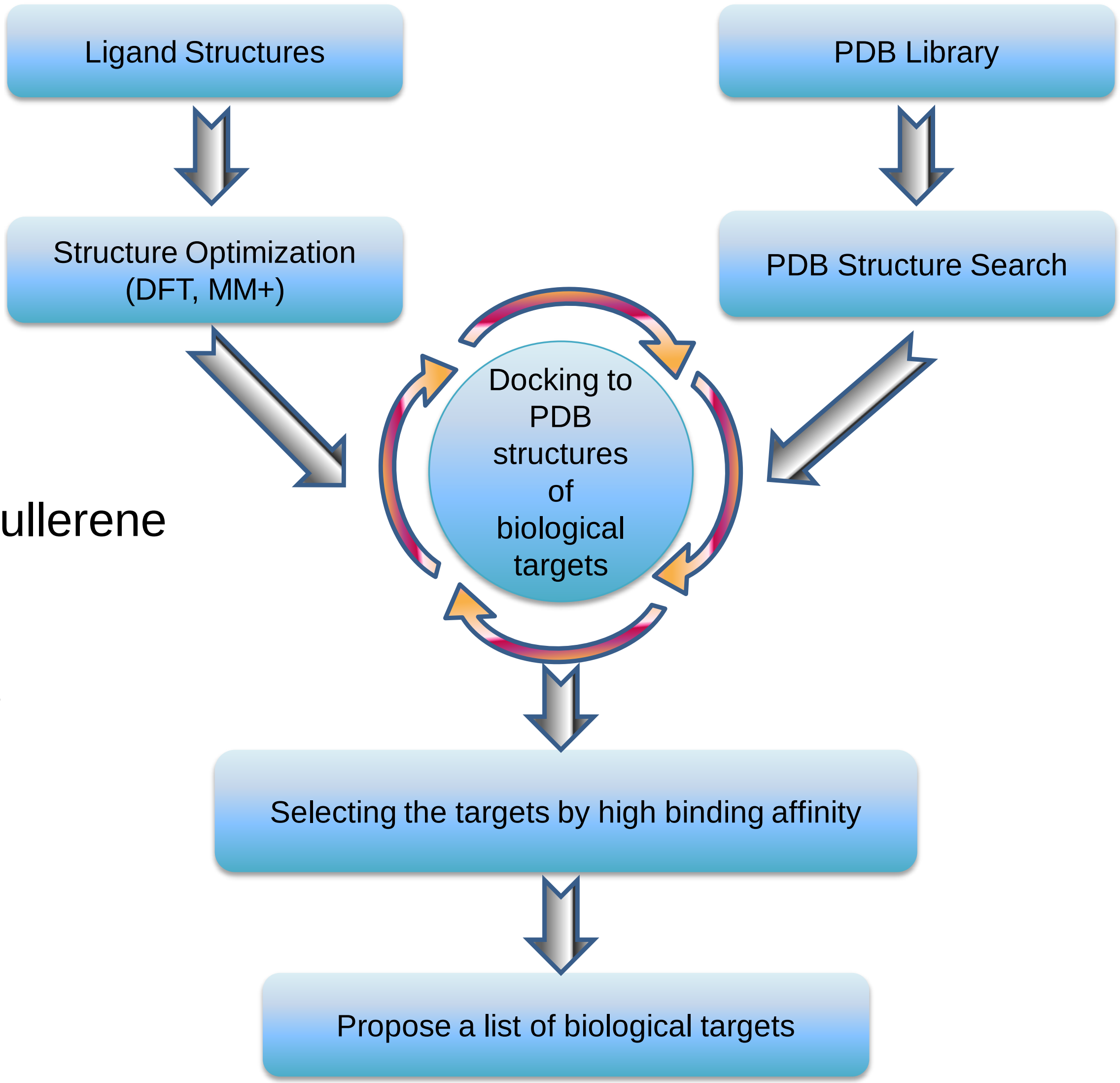
PDB Structure Search

Docking to PDB structures of biological targets

Selecting the targets by high binding affinity

Propose a list of biological targets

Overall Schematic Diagram of the Study



Ligand Structures

PDB Library

Structure Optimization
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PDB Structure Search

Docking to
PDB
structures
of
biological
targets

Selecting the targets by high binding affinity

Propose a list of biological targets

200 000
pairs protein-fullerene

≈ 1 000 000
docked poses

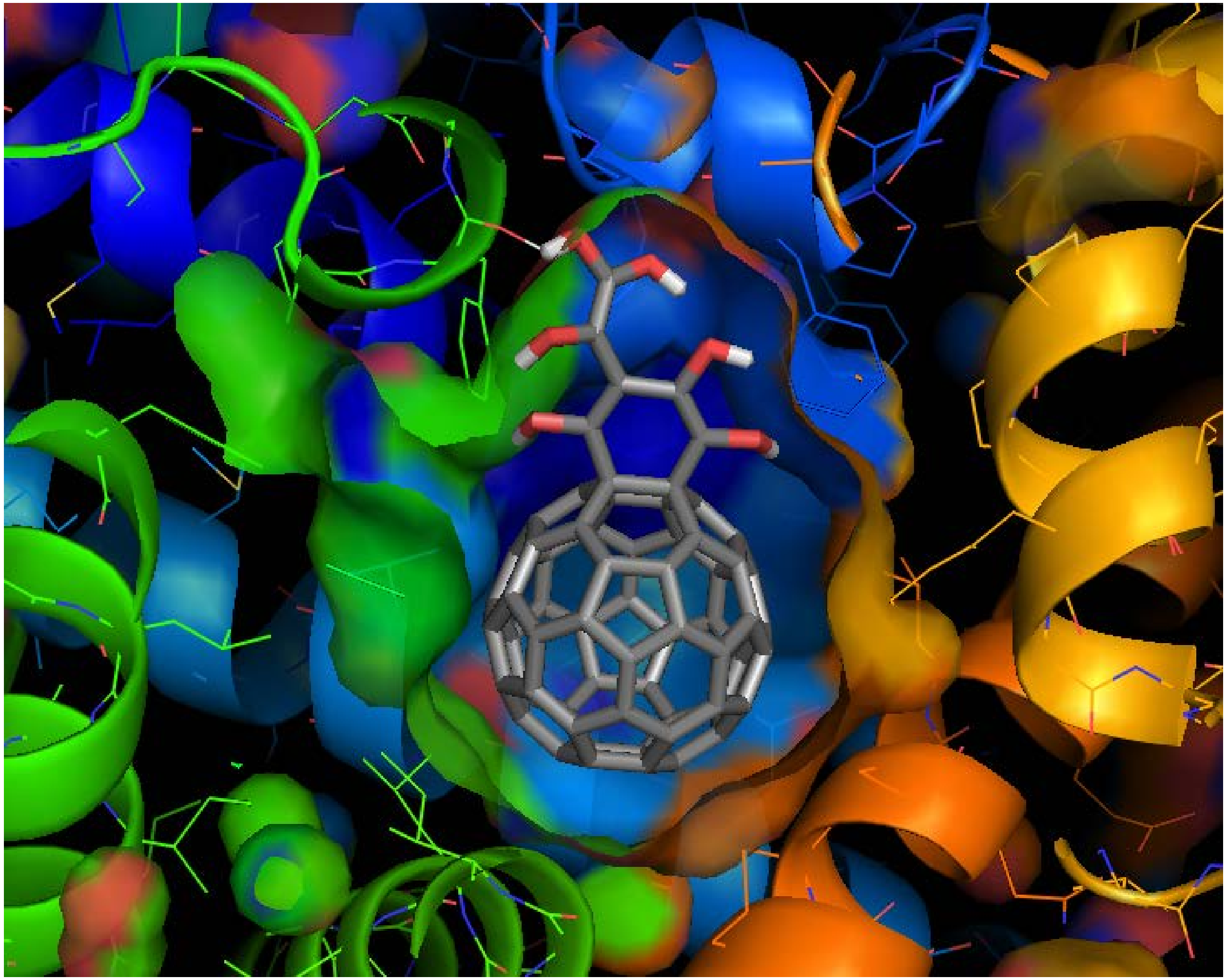

Figure: Glutamate transporters (Glt$_{ph}$, PDB ID: 1XFH) has a homotrimeric subunit with a large central water-filled cavity that restricts ligand diffusion to the exterior bulk medium. Fullerene derivative trapped in the cavity.

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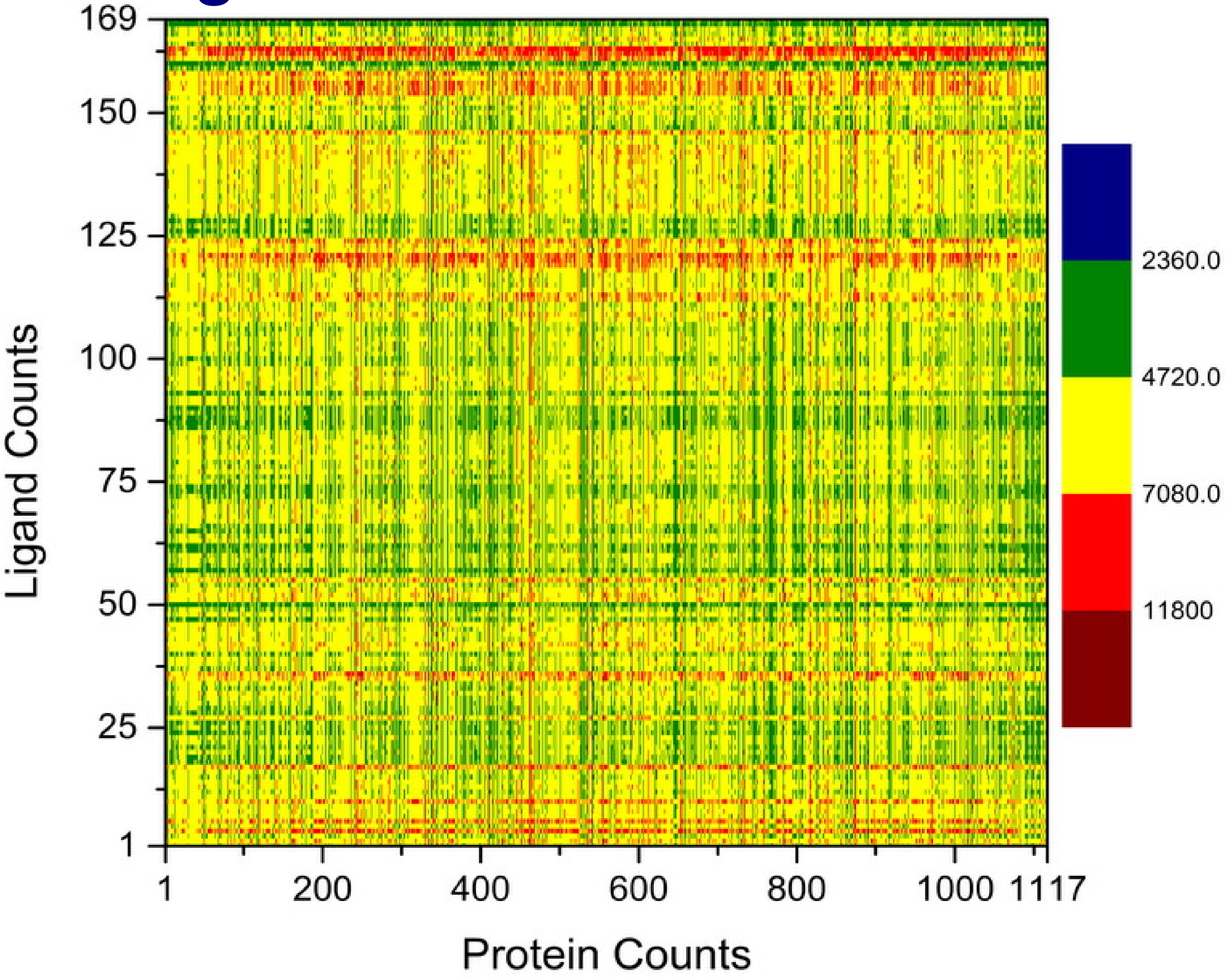
Binding Score



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Top target proteins

Rank	PDB_ID	Biochemical Type	Therapeutic Area	Target Details
1	1RTD	Enzyme	Viral infections	DNA Polymerase/reverse Transcriptase, HIV-1 Reverse Transcriptase
2	1HKB		Hormones and hormone antagonists	D-Glucose 6-Phosphotransferase
3	2BU5	Enzyme		Pyruvate dehydrogenase kinase-2
4	1CVI	Enzyme		Prostatic acid phosphatase
5	1OVM	Enzyme	Vitamins	Indole-3-Pyruvate Decarboxylase
6	8CAT	Enzyme		Oxidoreductase
7	1H9U	Nuclear Receptor	Vitamins	Retinoid X Receptor, Beta
8	2VAA	Monoclonal Antibodies		Murine MHC class I H-2Kb
9	1KAE	Enzyme	Synaptic and neuroeffector junctional sites and central nervous system	Histidinol Dehydrogenase
10	1IG0	Enzyme	Vitamins	Thiamin Pyrophosphokinase
11	2BWN	Enzyme		5-Aminolevulinate synthase
12	2F9Q	Enzyme		CYP2D6
13	6COX	Enzyme	Inflammation	Cyclooxygenase 1,2(COX-1,COX-2)
14	1HNI	Enzyme	Viral infections	HIV-1 Reverse Transcriptase
15	1IYH	Enzyme	Blood and blood-forming organs	Hematopoietic Prostagladin D Synthase

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Descriptors + Data Mining = New Materials Design Risk Assessment

Nano-Descriptors Generation and Calculation

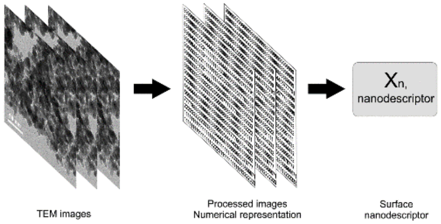


Figure. A representation of theoretical descriptor generation based on experimental TEM images for NMs (Copyright - Gajewicz A, Rasulev B. et al, Advanced Drug Delivery Reviews, 2012).

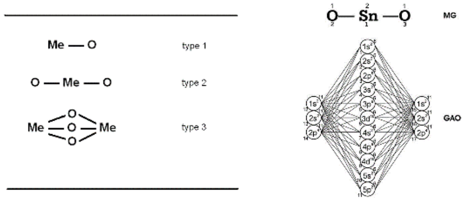


Figure. A representation of graph-atomic orbitals descriptors for encoding of metal oxide NMs (Copyright – Rasulev B. et al, FACSS conference proceedings, 2007).

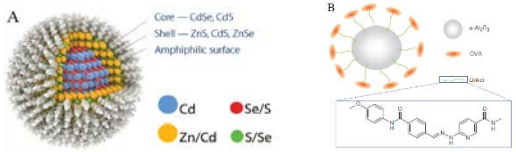


Figure. The structure of CdSe/ZnS quantum dot with ligand coating (a) and Al2O3 nanoparticle with OVA and linker (b). Copyright – (a)en.rusnano.com, (b)-nature.com (Li et al., 2011).

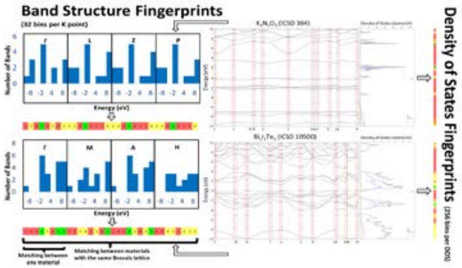
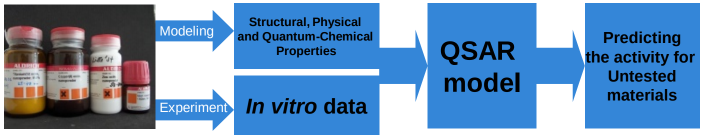


Figure. Construction of materials fingerprints from the band structure and the density of states. Copyright (Isayev et al., 2015).

Modeling of Nanomaterials Structure-Property Relationship



Modeling → Structural, Physical and Quantum-Chemical Properties

Experiment → *In vitro* data

QSAR model → Predicting the activity for Untested materials

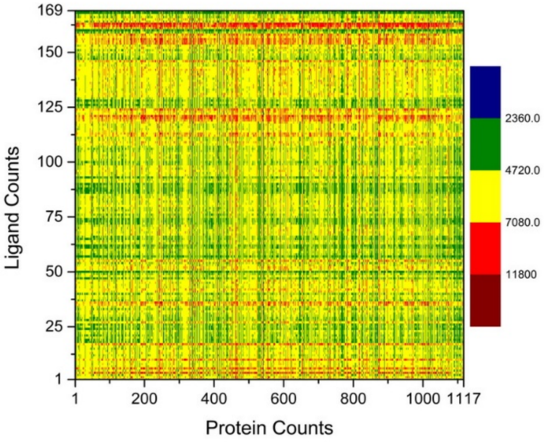
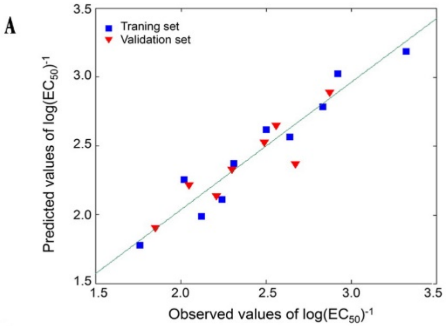
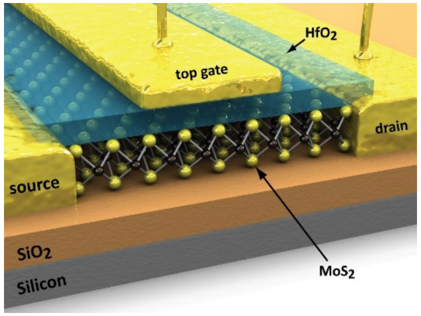
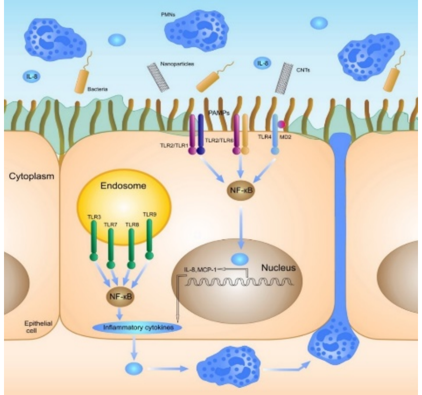


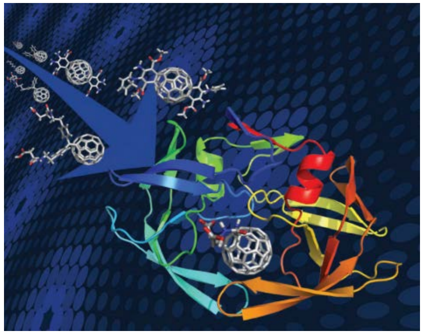
Figure. A representation of data mining for protein-ligand docking studies of 1200 proteins and 169 fullerene nanoparticles (*Nature Nanotechnology.* Copyright - Ahmed L, Rasulev B. et al,, 2015, under review).



Development of new Nanomaterial with improved properties or predicting Toxicity







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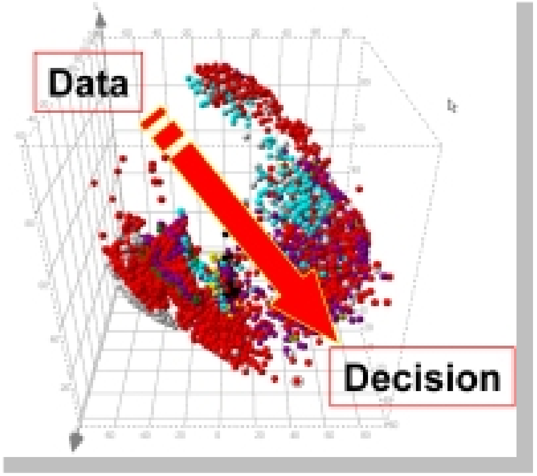

Thanks
for your attention!