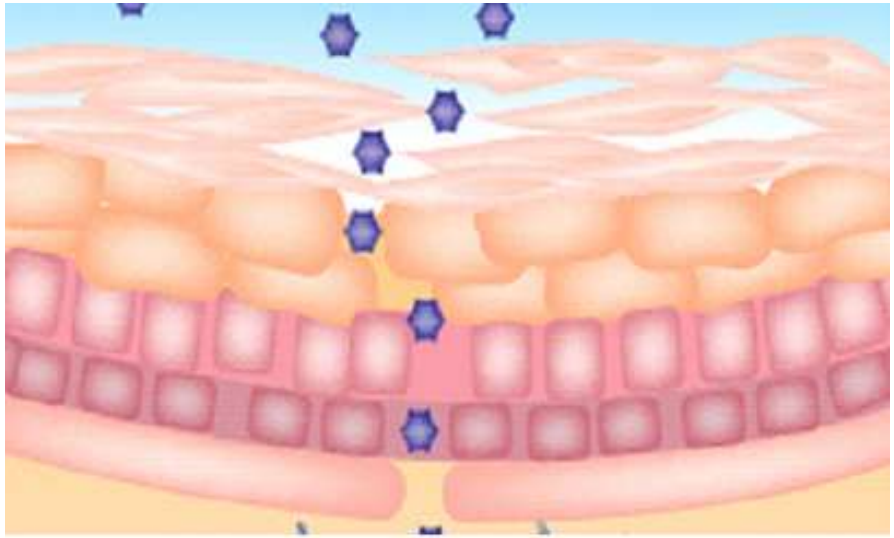
Percutaneous penetration of nanoparticles through healthy and diseased skin



Sanja Kezic
Academic Medical Centre, Amsterdam

Outline

- COST Action: SKINBAD
- Skin barrier and percutaneous penetration
- Study on percutaneous penetration of nanosilver

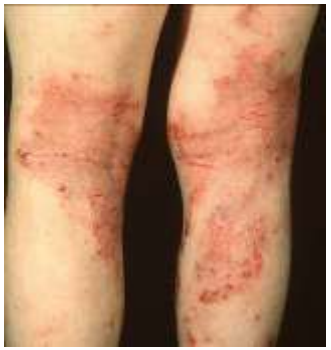
  Preliminary results and possibilities for cooperation

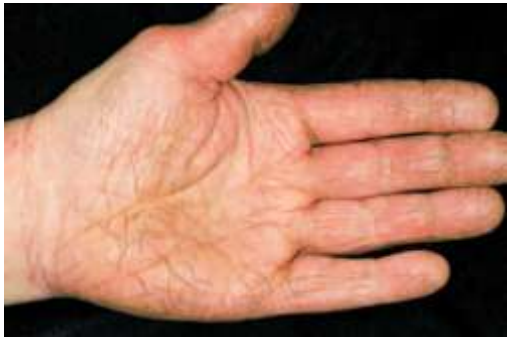
COST Action BM 0903

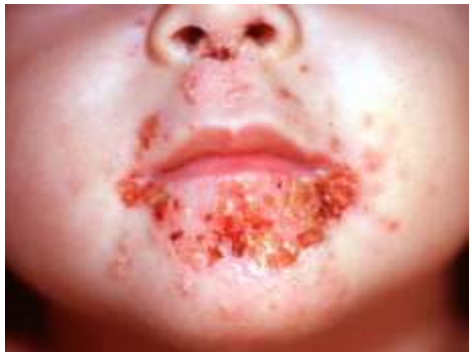
SKINBAD: Skin barrier in atopic diseases

Atopic dermatitis (atopic eczema)

Chronic inflammatory skin disease







- Inflammed, dry, itchy skin
- Reduced skin barrier even in non affected skin
- Increased susceptibility to superinfections (SA)
- Immune aberrations
- Hay fever, food allergy, asthma

Prevalence of atopic eczema

Children < 5 ys: 10 – 20 %

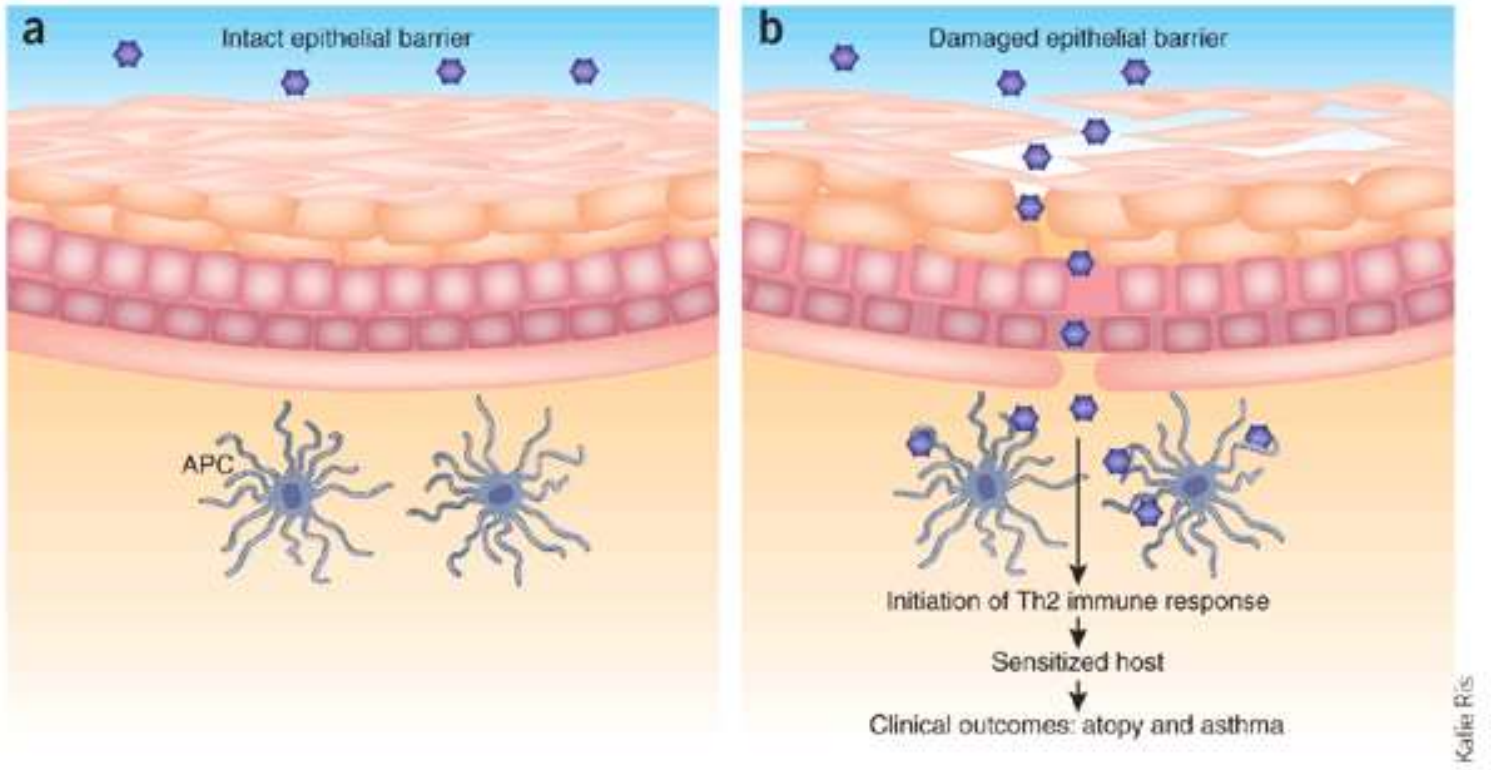
Adults: 3 – 5 %

Hanifin JM: Epidemiology of Atopic Dermatitis. Immunol Allergy Clin NA 2002; 22: 1-24

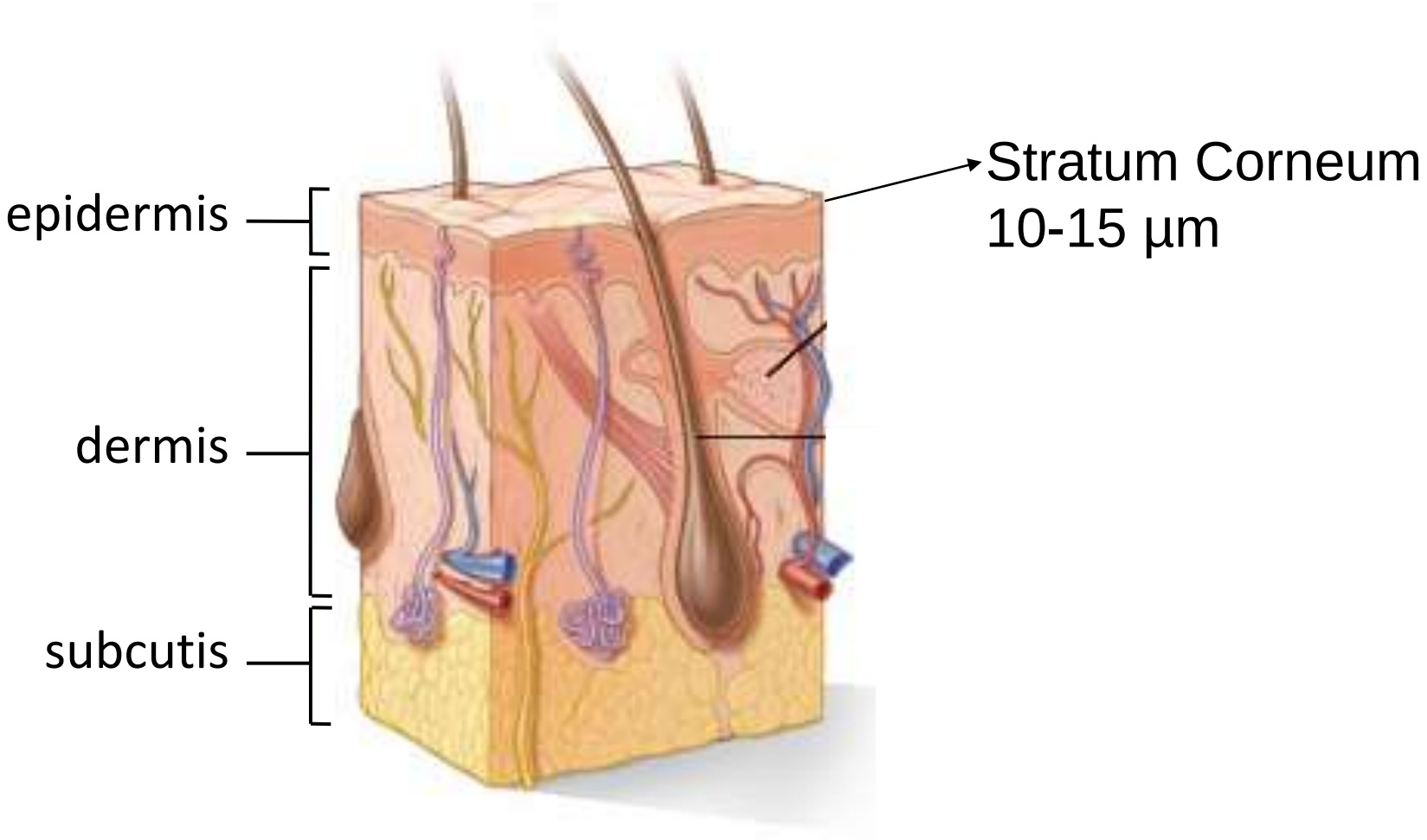
Leung DYM, Eichenfield LF, Boguniewicz M: Atopic dermatitis. In Fitzpatrick TB, Freedberg IM, Eisen AZ (eds): Fitzpatrick's Dermatology in General Medicine. McGraw-Hill Professional, New York, 2003, pp. 1180–1194

Etiology: strong genetic components
Loss-of-function mutations in gene encoding for epidermal protein filaggrin

Reduced skin barrier: primary event in development of AD



Structure of human skin



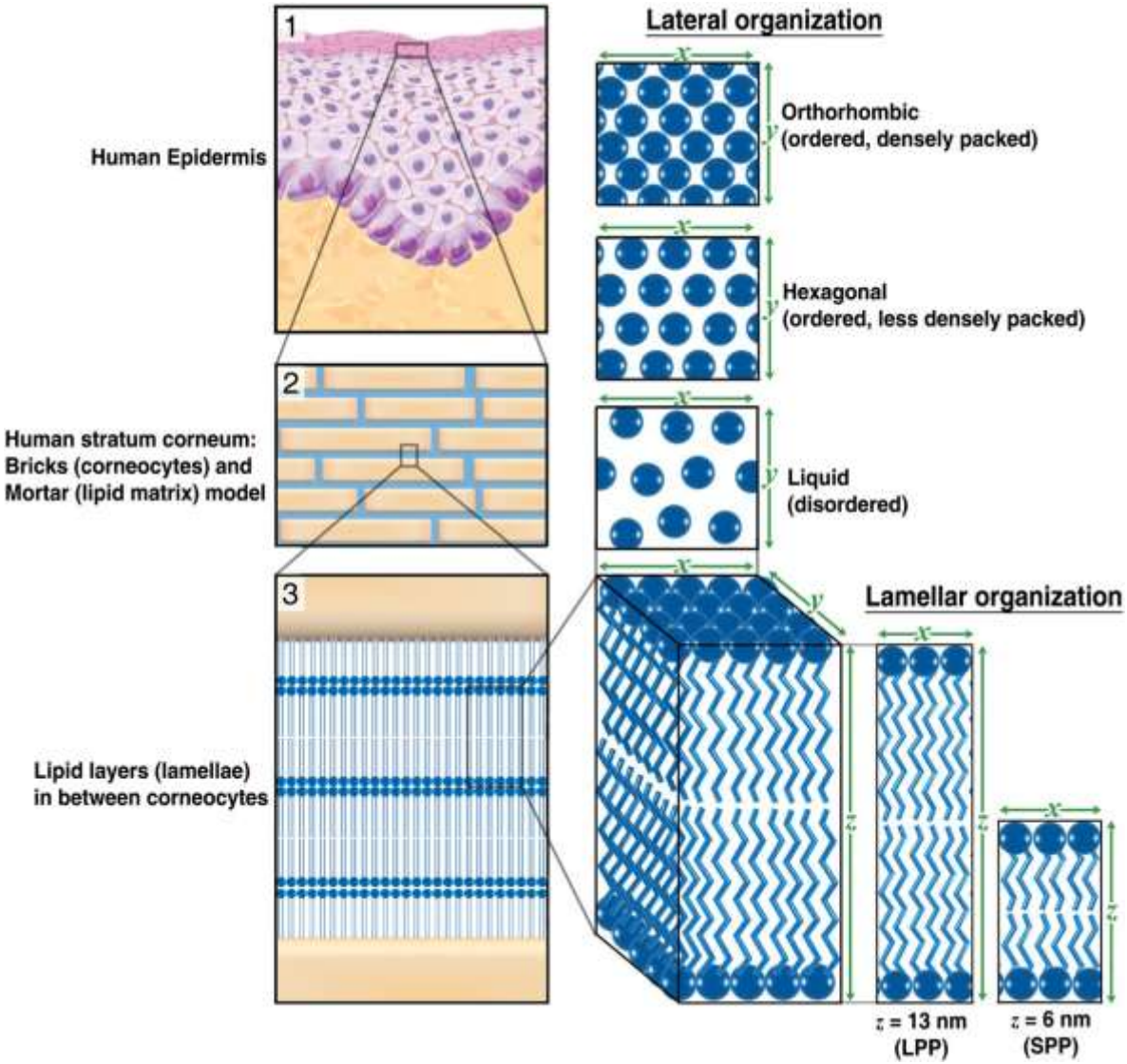
Human stratum corneum.

Latelar organization of intercellular lipids:

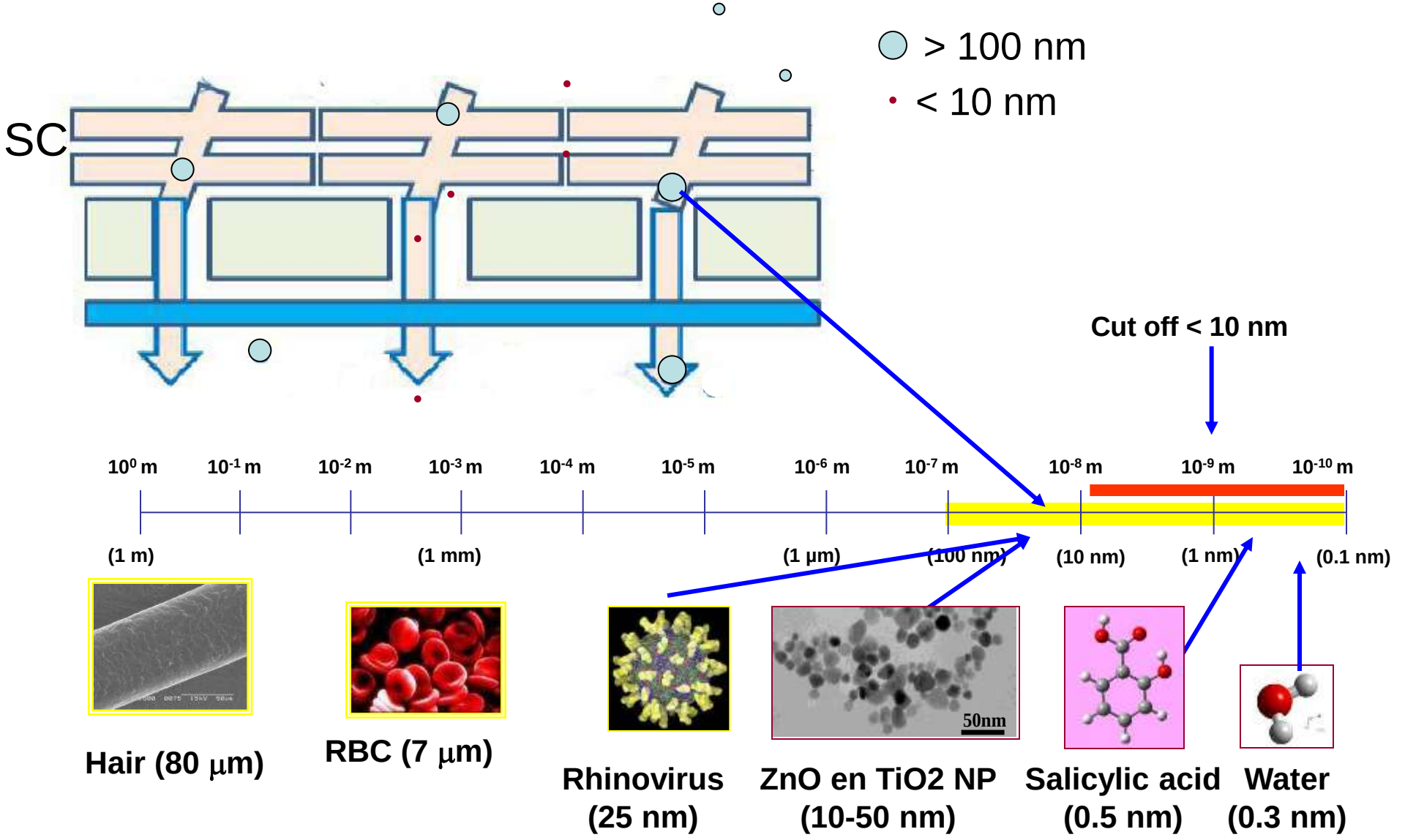
FTIR

Lamellar organization of intercellular lipids

Small angle X-ray diffraction (SAXD) European Synchrotron Radiation Facility (ESRF, Grenoble, France)

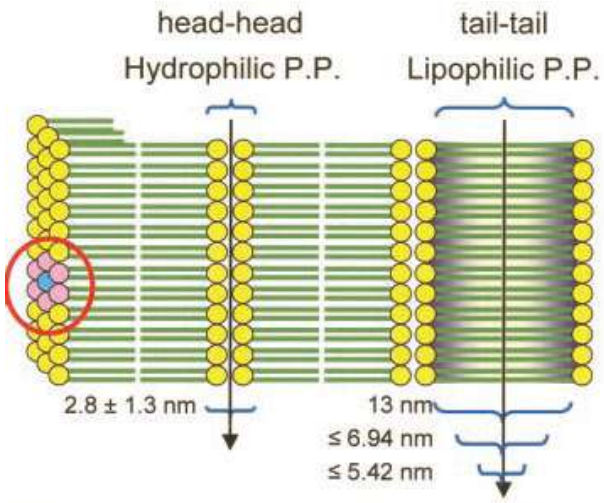


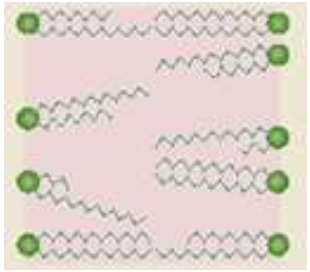
Janssens M et al. J. Lipid Res. 2012;53:2755-2766


> 100 nm
< 10 nm
SC
Cut off < 10 nm
10^0 m
10^-1 m
10^-2 m
10^-3 m
10^-4 m
10^-5 m
10^-6 m
10^-7 m
10^-8 m
10^-9 m
10^-10 m
(1 m)
(1 mm)
(1 µm)
(100 nm)
(10 nm)
(1 nm)
(0.1 nm)
50nm
Hair (80 µm)
RBC (7 µm)
Rhinovirus (25 nm)
ZnO en TiO2 NP (10-50 nm)
Salicylic acid (0.5 nm)
Water (0.3 nm)

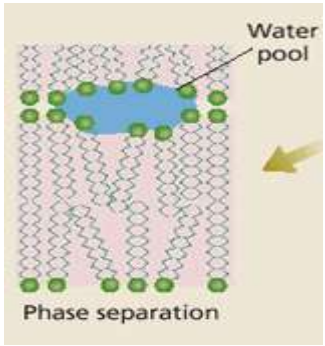

Coronel Institute

But..what about damaged skin?





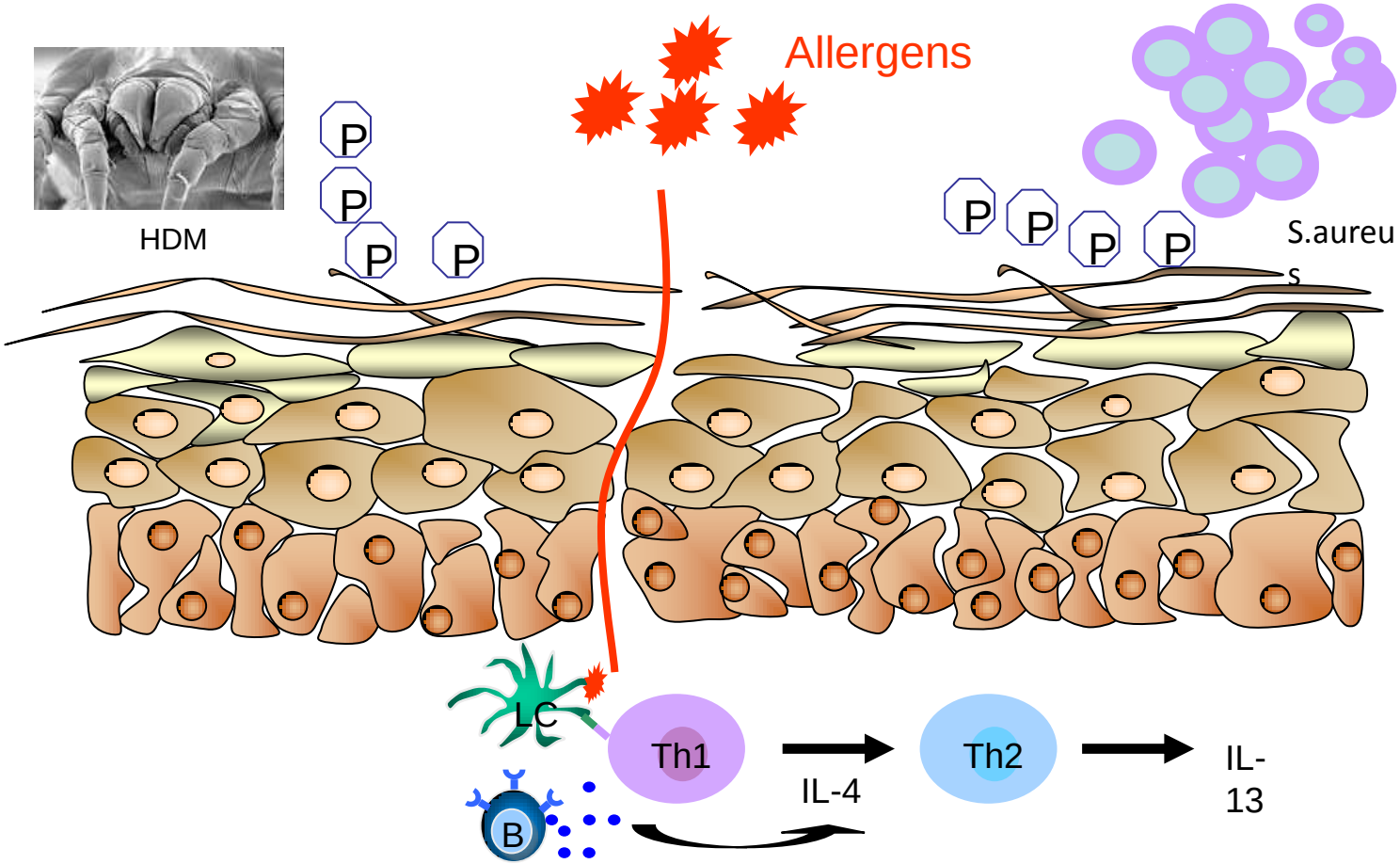
Organic solvents



Excessive hydration

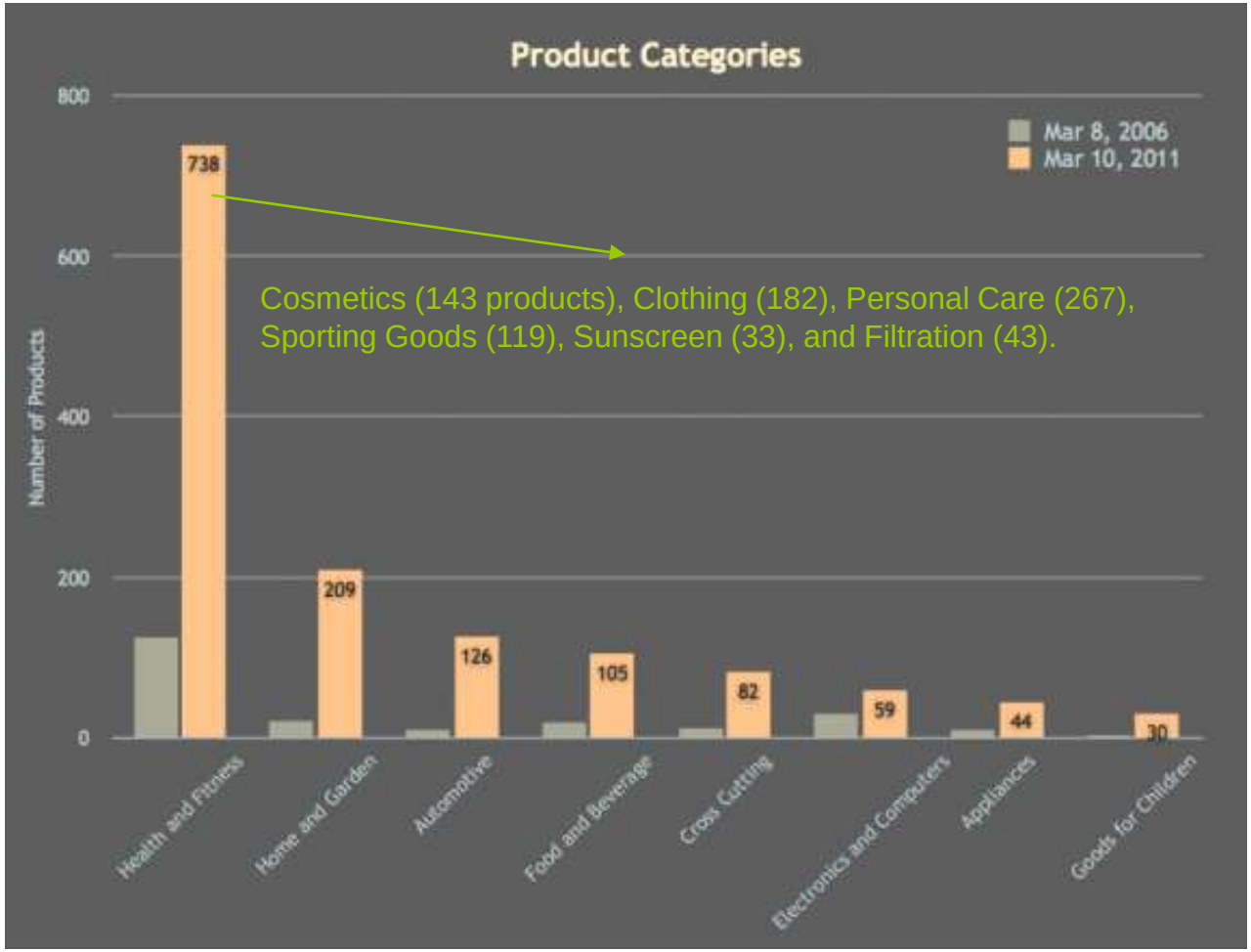
..UV radiation, diseased skin……

Atopic skin: Reduced skin barrier even in non affected sites

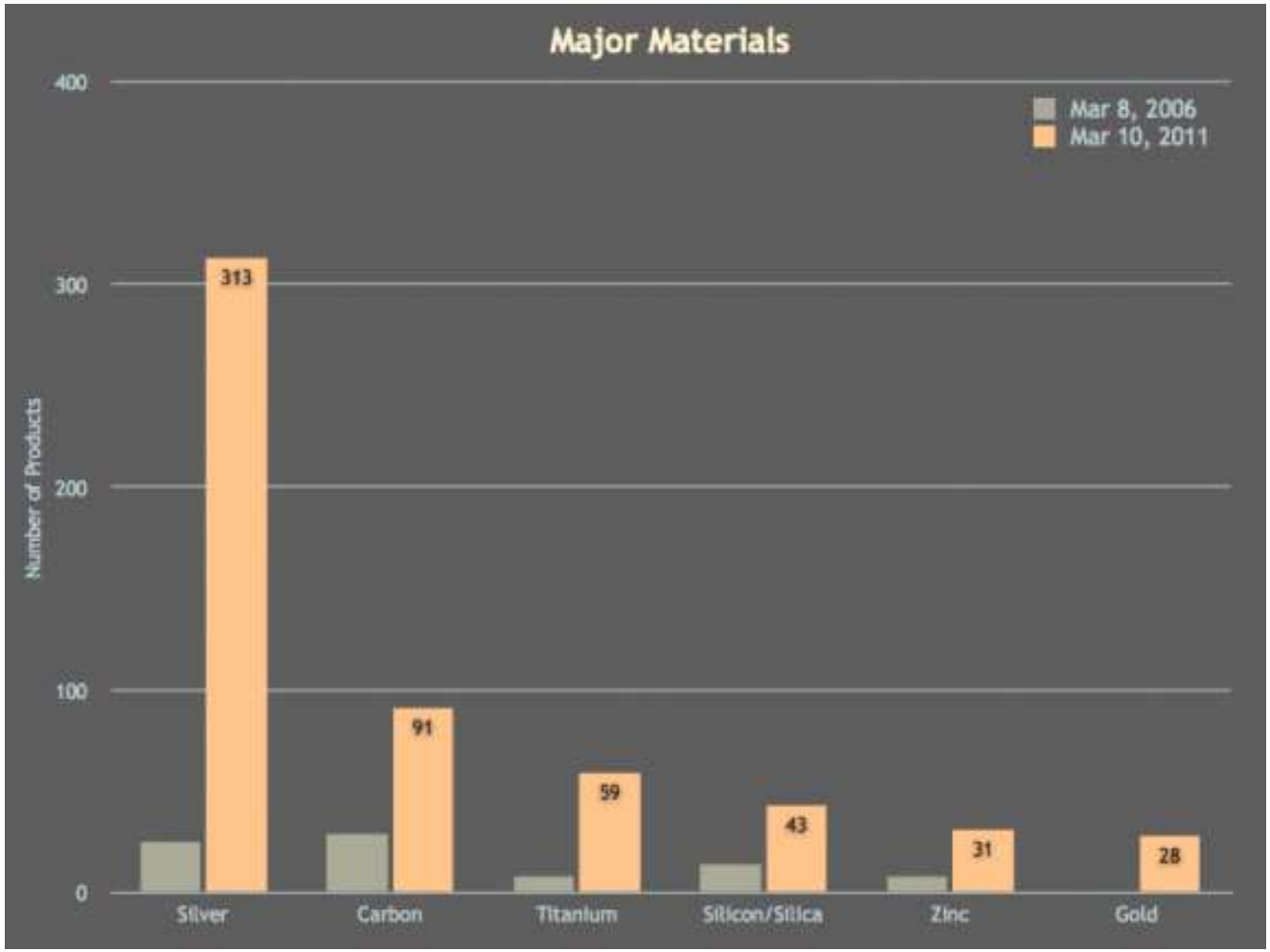


Why nanoparticles?

Explosive increase of nanoparticles in consumer products



Which products?



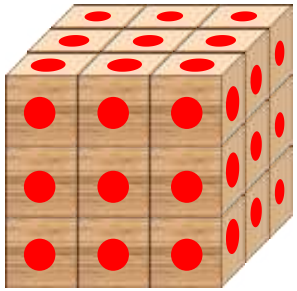
http://www.nanotechproject.org/inventories/consumer/analysis_draft/

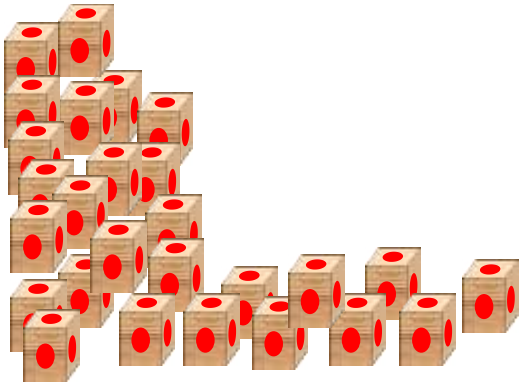
Nanosilver

Antimicrobial properties: Ag^+

Nanosilver: improved antimicrobial properties

Elevated emission of ions, higher active surface, more penetration into the cell

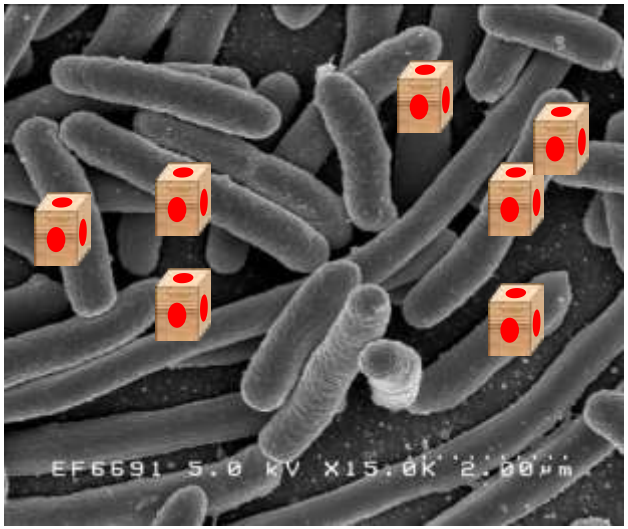




1 cm = 1 = 6 cm2

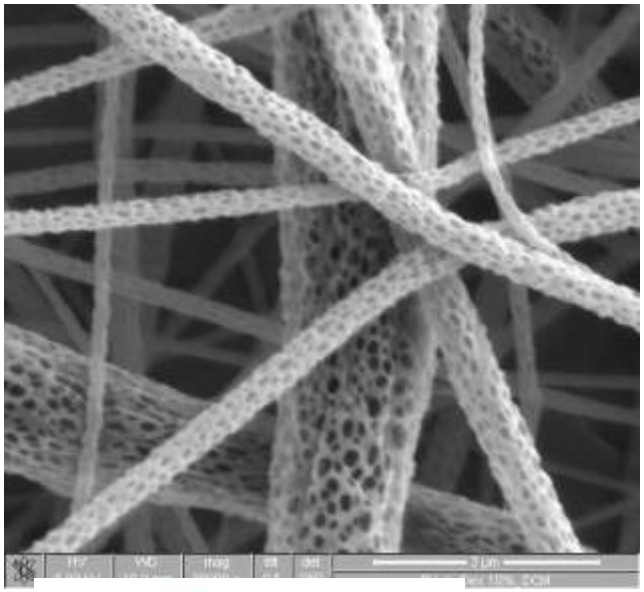
1 mm = 1000 = 60 cm^2

1 nm = 1x 10^{21} = 60 000 000 cm^2



E coli

Nanosilver: use



Wound healing

Health risk assessment: knowledge gaps

Very few data on percutaneous penetration, local and systemic toxicity,
➔most of data from *in vitro studies*

No data on damaged skin under ‘’in use’’ exposure scenarios

Objectives

1) Percutaneous absorption of NP *in vitro* and *in vivo* in intact and damaged skin

2) Local effects of NP (e.g. inflammation, OS, antimicrobial properties)

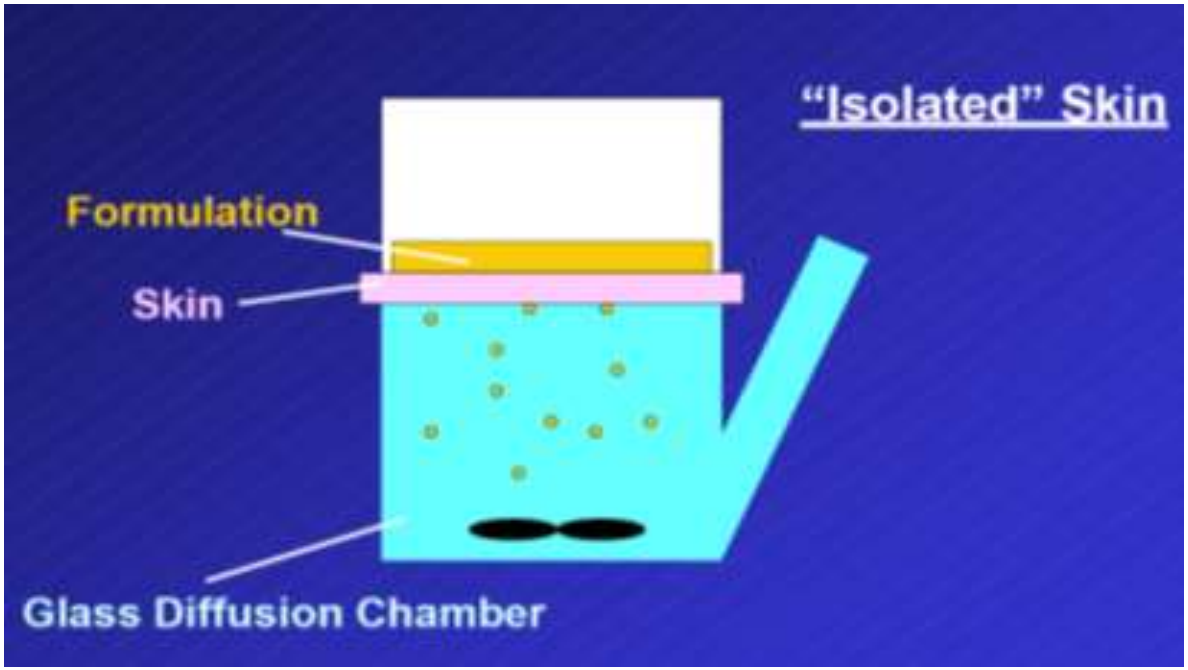
NanoNextNL: Human Health Risks

	Name (affiliation)
Project 1 Risk assessment	H. Marvin (WUR-RIKILT)
	A. Sips (RIVM)
	D. Kroese (TNO)
	-
	-
Project 2 Detection	A. Schmidt-Ott (TUD)
	D. Brouwer (TNO)
	-
	-
Project 3 Exposure	E. Tielemans and D. Brouwer (TNO)
	J. van Engelen (RIVM)
	H. Goosens (Philips Aerasense BV)
	P. van de Broekhuizen (IVAM)
	M. van Wijk (BECO)
	P. Borm (Hogeschool Zuyd)
	R. Vermeulen (UU)
Project 4 Bioavailability	H. Bouwmeester (WUR-RIKILT)
	W. de Jong (RIVM)
	S. Bellman, M. Rennen (TNO)
	P. Krystek (MiPlaza)
	S. Kezic (UvA)
Project 5 Toxicity	I. Rietjens (WUR-TOX)
	W. de Jong (RIVM)
	F. Kuper (TNO)
	-
	S. LeGac (UT)

1. Intestinal
2. Lung
3. Skin
4. Placenta

Study design: 1. *in vitro*

Ag-NM in artificial sweat

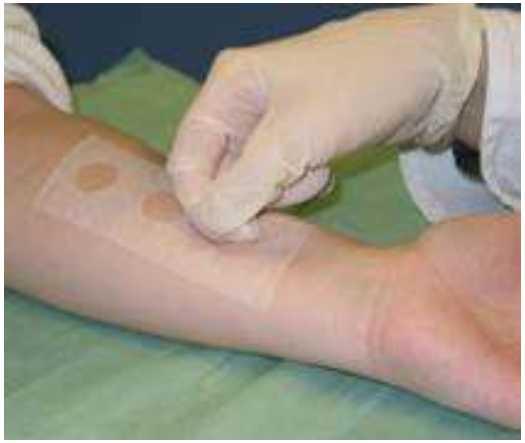


1. Ag-total: donor, receptor, all skin strata
2. Particle characterisation

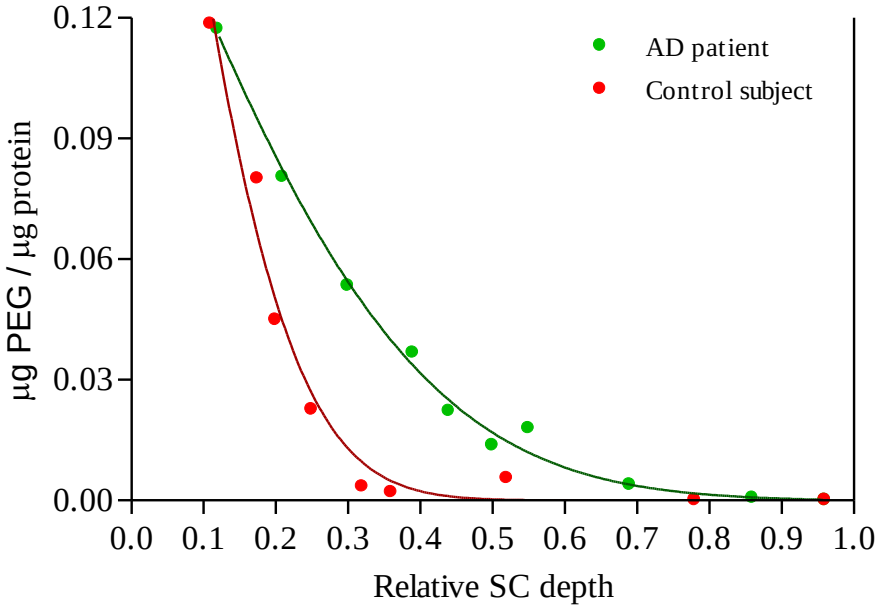
Study design: 2. *in vivo*

- Healthy volunteers + AD patients
- Exposure: 2-3 weeks, 8 hrs/day

Sampling: 1. penetration



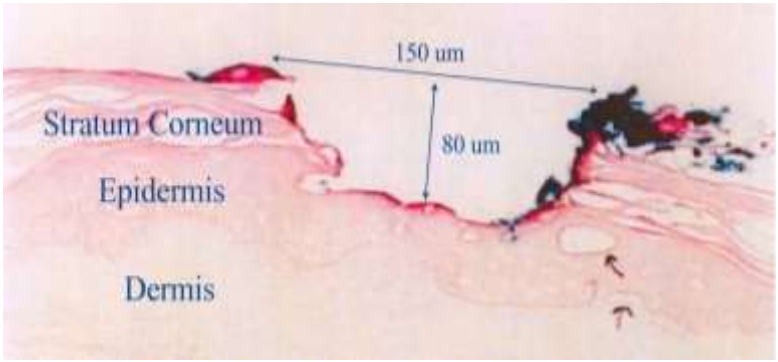
Tape stripping

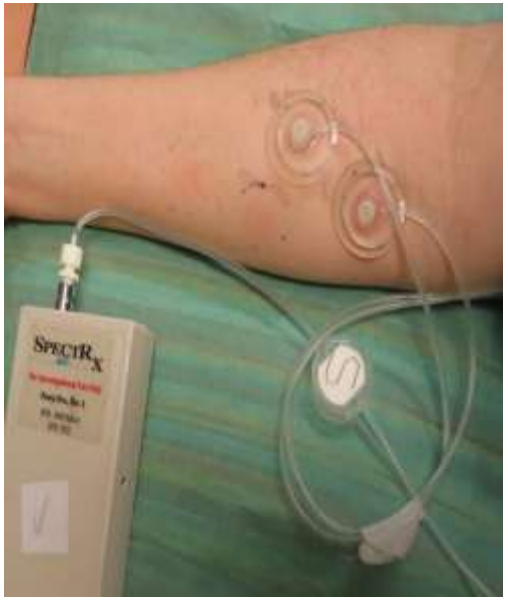


Biopsy + urine (systemic uptake)

Staphylococcus aureus (swabs, TS)

2. Transdermal fluid





Inflammatory mediators, Oxidative stress parameters

Which NP?

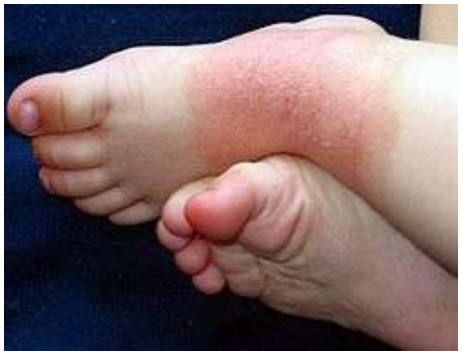
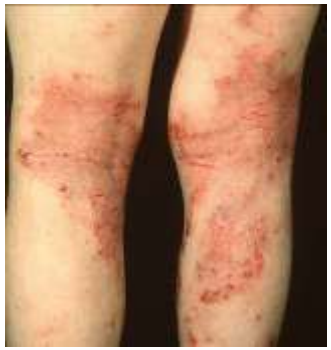
Nanosilver from commercial products: garments for AD, wound healing medicinal products, sport socks; T-shirts

Later: defined particle size

Model of damaged skin?

Atopic skin: reduced skin barrier even in non affected skin

- Reduced skin barrier even in non-affected skin
- Increased susceptibility to superinfections (SA)
- Silver garments to cure infections

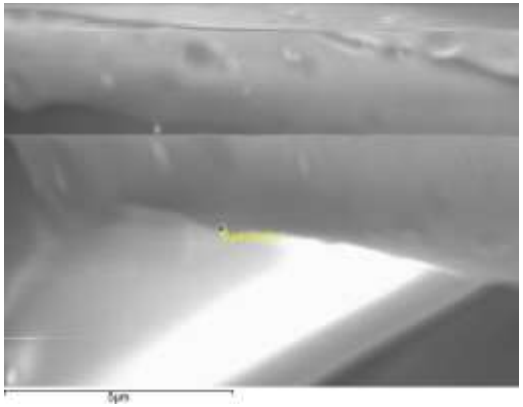



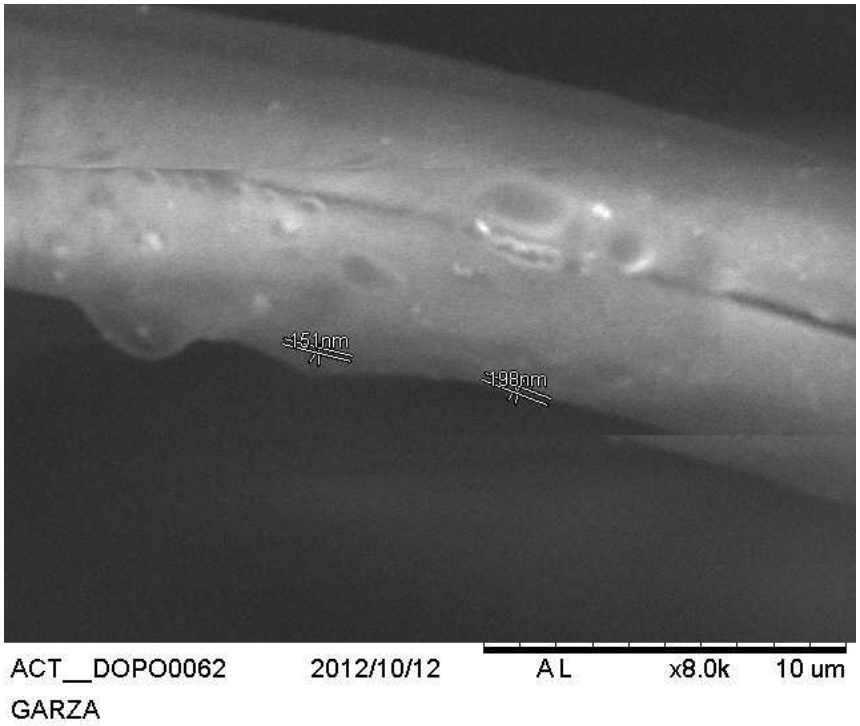
Planning

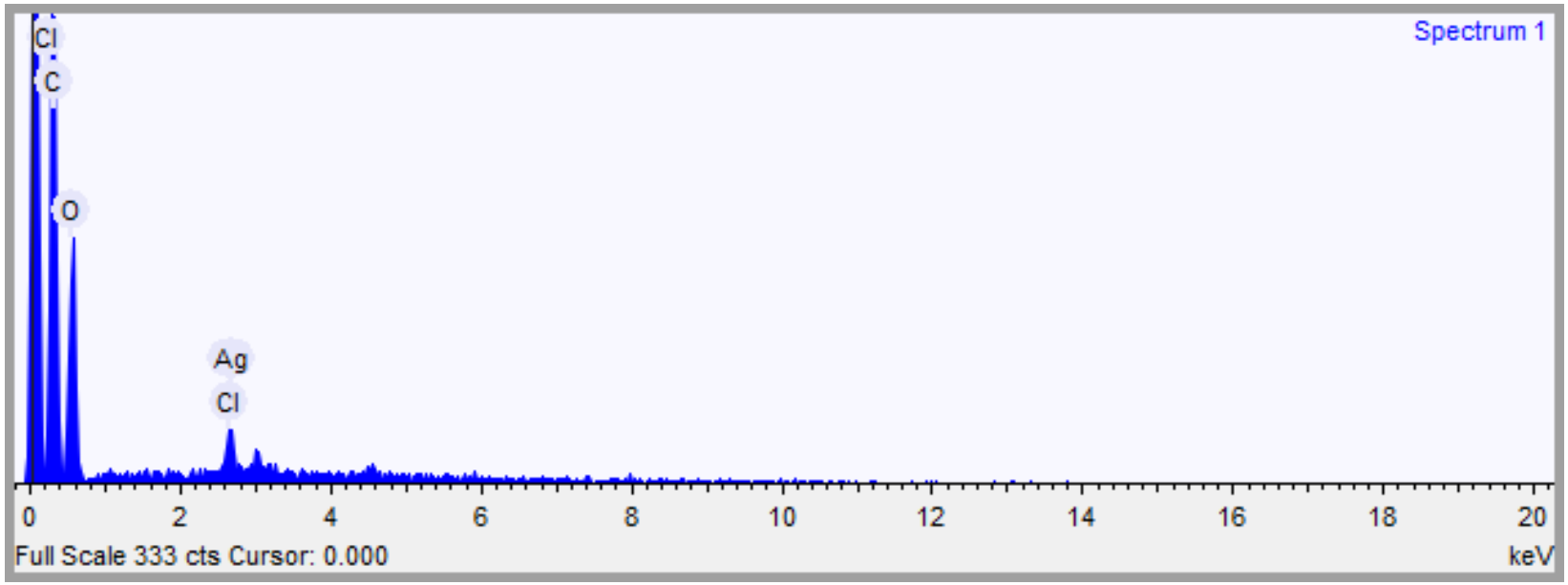
October	Release tests
November	*In vitro*
February- July 2013	*In vivo*

Release: SEM

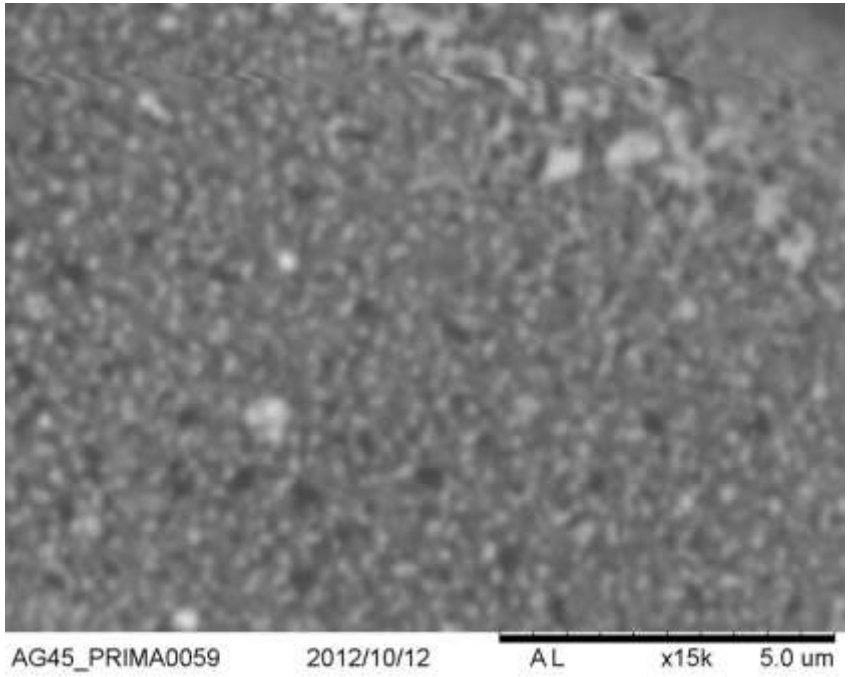
SEM analysis after 24 hour of soaking into synthetic sweat.
Fig. 1: Morphologic analysis of the gauze reveals silver nanoparticles released by the silver grid.
Fig. 2: Electron image of silver nanoparticles on the gauze surface shows Silver secondary peak absorption (L). Silver Chloride is present on the surface as expected.






Spectrum 1
Cl
C
O
Ag
Cl
0
2
4
6
8
10
12
14
16
18
20
Full Scale 333 cts Cursor: 0.000
keV

Product 2: before release test



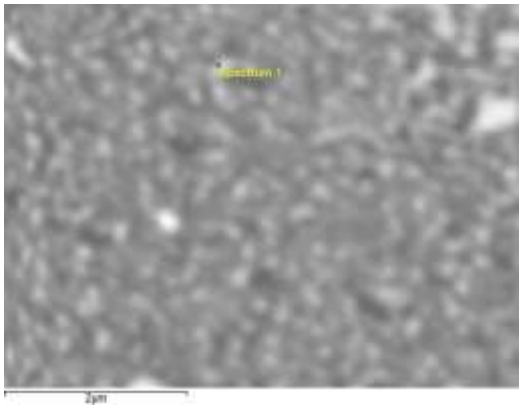


Fig. 1: Morphologic analysis of Silverlon Ag 45 reveals silver nanoparticles on the fabric surface (50-190 nm).
Fig. 2: Electron image of Silverlon Ag 45 shows Silver secondary peak absorption (L).

After release