15th ETH-Conference on Combustion Generated Nanoparticles
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Title: Combustion-derived nano particle toxicity: lessons learnt from air pollution research

Extended Abstract

Ambient particulate matter (PM) has been extensively studied in the past two decades because of the association of the level of PM and several markers for morbidity as well as premature death in the general population. Much has been done to clean up the air that we breath and in most places standards for PM are met. There are still hot spots, such as busy traffic locations for which abatement measure need to be taken to further reduce the concentrations. Since the effects are still noted below current European standards, and the cost of reducing PM further are getting higher and higher, targeted research is needed to identify the most harmful constituents and the related sources of emissions. Indeed, ultra fine or nano scale particles seem to contribute to the adverse health effects and evidence is becoming increasingly available. It is not only the respiratory tract and the lungs that are affected, evidence is emerging that the cardiovascular and central nerve system including the brain are important targets for these small, insoluble particles.

Ambient PM is nowadays regulated on the basis of mass per unit volume. However, this metric does not capture the very small particles at the nano scale, i.e. < 200 nanometer. This fraction contributes very little to the overall PM10 pr PM2.5 mass, but the particle number concentrations can be dramatically higher than that of the accumulation (>200 nm) and coarse (1-10 um) mode PM. In particular near combustion sources such as road traffic, these levels can become as high as 10^5 particles per cc. There is evidence from toxicology and to a far lesser extend from environmental epidemiology that these so called ultra fine particles are far more harmful than larger sized particles at the same mass concentrations. There are various reasons postulated to explain the difference compared to larger particles, e.g.

- These particles will penetrate deeper into the lung
- The number of hits per cells in the lung is substantially larger
- The relative surface to mass ration is large
- Surface properties may be very different (more chemically active, surface charge)
- They posses the ability to translocate easily from the lung into the blood, or from the olfactory epithelium in the nose to the brain

Various examples were presented to support these ideas. For example, very recently, nineteen healthy volunteers (mean age, 25±3 years) were exposed to filtered air and diesel exhaust in the presence or absence of a particle trap for 1 hour in a randomized, double-blind, 3-way crossover trial (Lucking et al., 2011). Bilateral forearm blood flow and plasma fibrinolytic factors were assessed with venous occlusion plethysmography and blood sampling during intra-arterial infusion of acetylcholine, bradykinin, sodium nitroprusside, and verapamil. Ex vivo thrombus formation was determined with the use of the Badimon chamber. Compared with filtered air, diesel exhaust inhalation was associated with reduced vasodilatation and increased ex vivo thrombus formation under both low- and high-shear conditions. The particle trap markedly reduced diesel exhaust particulate number (from 150 000 to 300 000/cm^3 to 30 to 300/cm^3 and mass (320±10 to 7.2±2.0 µg/m^3), and was associated with increased vasodilatation, reduced thrombus formation, and an increase in tissue-type plasminogen activator release. Exhaust particle traps are a highly efficient method of reducing particle emissions from diesel engines. With a range of surrogate measures, the use of a particle trap prevents several adverse cardiovascular effects of exhaust inhalation in men. These findings were supported by another study of the same group (Mills et al, 2011), in which it was shown that filtering particles out of engine exhaust using a HEPA filter completely

reduced to negative impact on arterial stiffness and blood clothing, whereas the exposure to elemental carbon did not affect the these parameters, suggesting that the components absorbed on the surface of carbonaceous particles may be the most harmful constituents and the solid particle only acts as carrier to transport the compounds deep into the lung. Indeed, in a study by Biswas et al (2009), it was demonstrated that in particular the semi-volatile components were responsible for most of the chemical activity. This study investigated the oxidative potential (OP) of PM from vehicles with six retrofitted technologies (vanadium and zeolite based selective catalytic reduction (V-SCRT, Z-SCRT), Continuously regenerating technology (CRT), catalyzed DPX filter, catalyzed continuously regenerating trap (CCRT), and uncatalyzed Horizon filter) in comparison to a "baseline" vehicle (without any control device). Vehicles were tested on a chassis dynamometer at three driving conditions, i.e., cruise, transient urban dynamometer driving schedule (UDDS), and idle. Significant reduction in OP (by 50-100%) was observed for thermally denuded PM from vehicles with retrofitted technologies (PM with significant semi volatile fraction), whereas particles emitted by the baseline vehicle (with insignificant semi volatile fraction) did not demonstrate any measurable changes in oxidative activity. This suggests that the semi volatile fraction of particles are far more oxidative in nature than refractory particles-a conclusion further supported by previous tunnel and ambient studies, demonstrating a decline in PM oxidative activity with increasing atmospheric dilution.
Also, in another study in human volunteers (Mills et al 2008) that were exposed to concentrated ambient PM2.5 (CAPs), not detrimental effects were noted after a single 2 hr exposure. Despite achieving marked increases in particulate matter, exposure to CAPs (low in combustion-derived particles) did not affect vasomotor or fibrinolytic function in either middle-aged healthy volunteers or patients with coronary heart disease. These findings contrast with exposures to dilute diesel exhaust (Lucking et al., 2008; Barath et al, 2010) and highlight the importance of particle composition in determining the vascular effects of particulate matter in humans. On the other hand, wearing a facemask appears to abrogate the adverse effects of air pollution on blood pressure and heart rate variability. This simple intervention has the potential to protect susceptible individuals and prevent cardiovascular events in cities with high concentrations of ambient air pollution and suggest that irrespective the nature of the particles, avoidance of exposure will result in improved health (Langrish et al, 2009). In conclusion, short term (i.e. 2 hr) exposures have to atmospheres rich in ultrafine or nano-sized particles have been demonstrated to result in adverse health effects. Surface chemistry seems to play a major role in the onset of these events.

Barath S, Mills NL, Lundbäck M, Törnqvist H, Lucking AJ, Langrish JP, Söderberg S, Boman C, Westerholm R, Löndahl J, Donaldson K, Mudway IS, Sandström T, Newby DE, Blomberg A. Impaired vascular function after exposure to diesel exhaust generated at urban transient running conditions.
Part Fibre Toxicol. 2010 Jul 23;7:19.

Biswas S, Verma V, Schauer JJ, Cassee FR, Cho AK, Sioutas C. Oxidative potential of semi-volatile and non volatile particulate matter (PM) from heavy-duty vehicles retrofitted with emission control technologies. Environ Sci Technol. 2009 May 15;43(10):3905-12

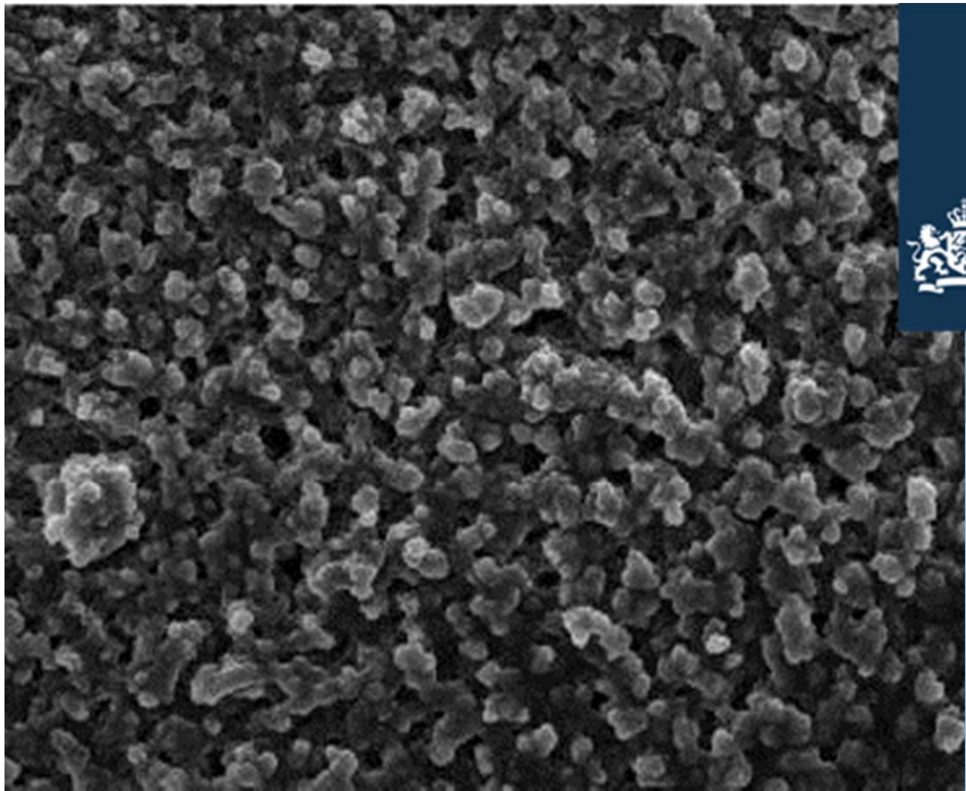
Langrish JP, Mills NL, Chan JK, Leseman DL, Aitken RJ, Fokkens PH, Cassee FR, Li J, Donaldson K, Newby DE, Jiang L. Beneficial cardiovascular effects of reducing exposure to particulate air pollution with a simple facemask. Part Fibre Toxicol. 2009 Mar 13;6:8.

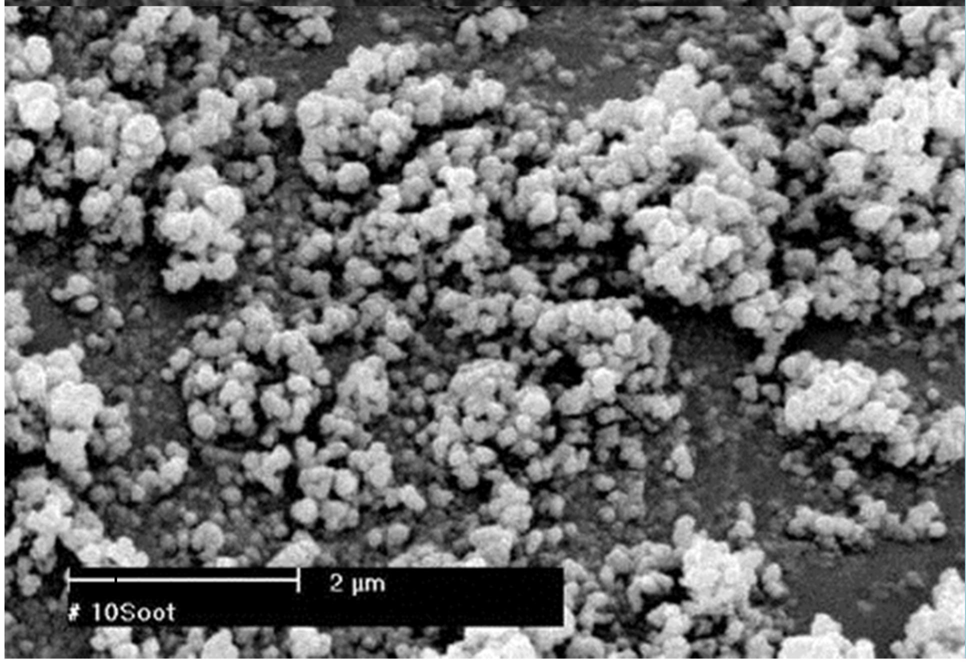
Lucking AJ, Lundback M, Mills NL, Faratian D, Barath SL, Pourazar J, Cassee FR, Donaldson K, Boon NA, Badimon JJ, Sandstrom T, Blomberg A, Newby DE. Diesel exhaust inhalation increases thrombus formation in man. Eur Heart J. 2008 Dec;29(24):3043-51.

Lucking AJ, Lundbäck M, Barath SL, Mills NL, Sidhu MK, Langrish JP, Boon NA, Pourazar J, Badimon JJ, Gerlofs-Nijland ME, Cassee FR, Boman C, Donaldson K, Sandstrom T, Newby DE, Blomberg A. Particle traps prevent adverse vascular and prothrombotic effects of diesel engine exhaust inhalation in men. Circulation. 2011 Apr 26;123(16):1721-8.

Mills NL, Miller MR, Lucking AJ, Beveridge J, Flint L, Boere AJF, Fokkens PHB, Boon NA, Sandstrom T, Blomberg A, Duffin R, Donaldson K, Hadoke PWF, Cassee FR, and Newby DE. Combustion-derived nanoparticulate induces the adverse vascular effects of diesel exhaust inhalation. Eur Heart J. 2011, in press.

Mills NL, Robinson SD, Fokkens PH, Leseman DL, Miller MR, Anderson D, Freney EJ, Heal MR, Donovan RJ, Blomberg A, Sandström T, MacNee W, Boon NA, Donaldson K, Newby DE, Cassee FR. Exposure to concentrated ambient particles does not affect vascular function in patients with coronary heart disease.Environ Health Perspect. 2008 Jun;116(6):709-15.





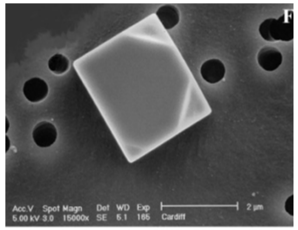
National Institute for Public Health
and the Environment
Ministry of Health, Welfare and Sport

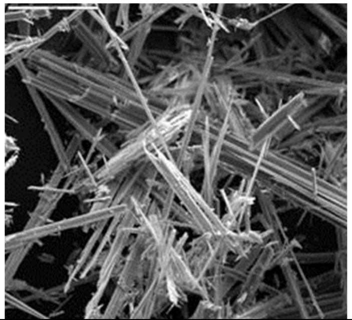
Combustion-Derived and Engineered Nanoparticle Toxicity: Lessons from Air Pollution Research

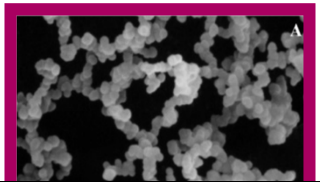
Dr Flemming R Cassee
RIVM, The Netherlands

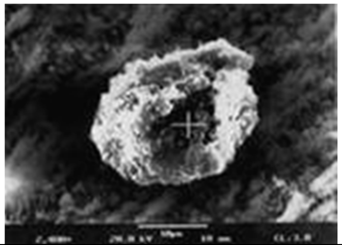
15th ETH-Conference on Combustion Generated Nanoparticles

t is unique about
oustion derived nanoparticles? Shape

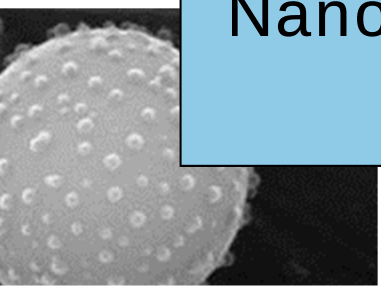


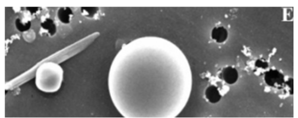




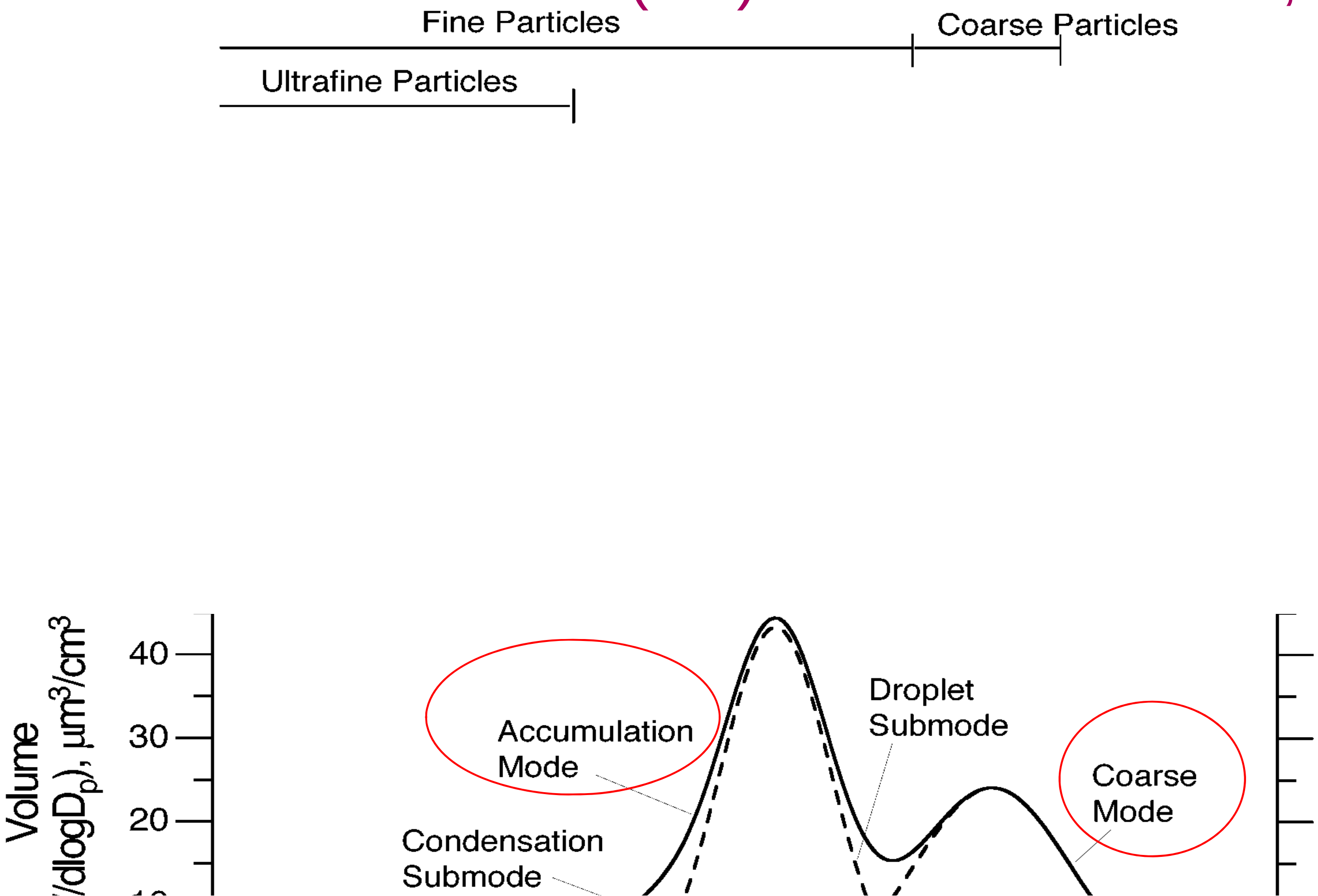


Nanoparticles aggregate, affects toxicity

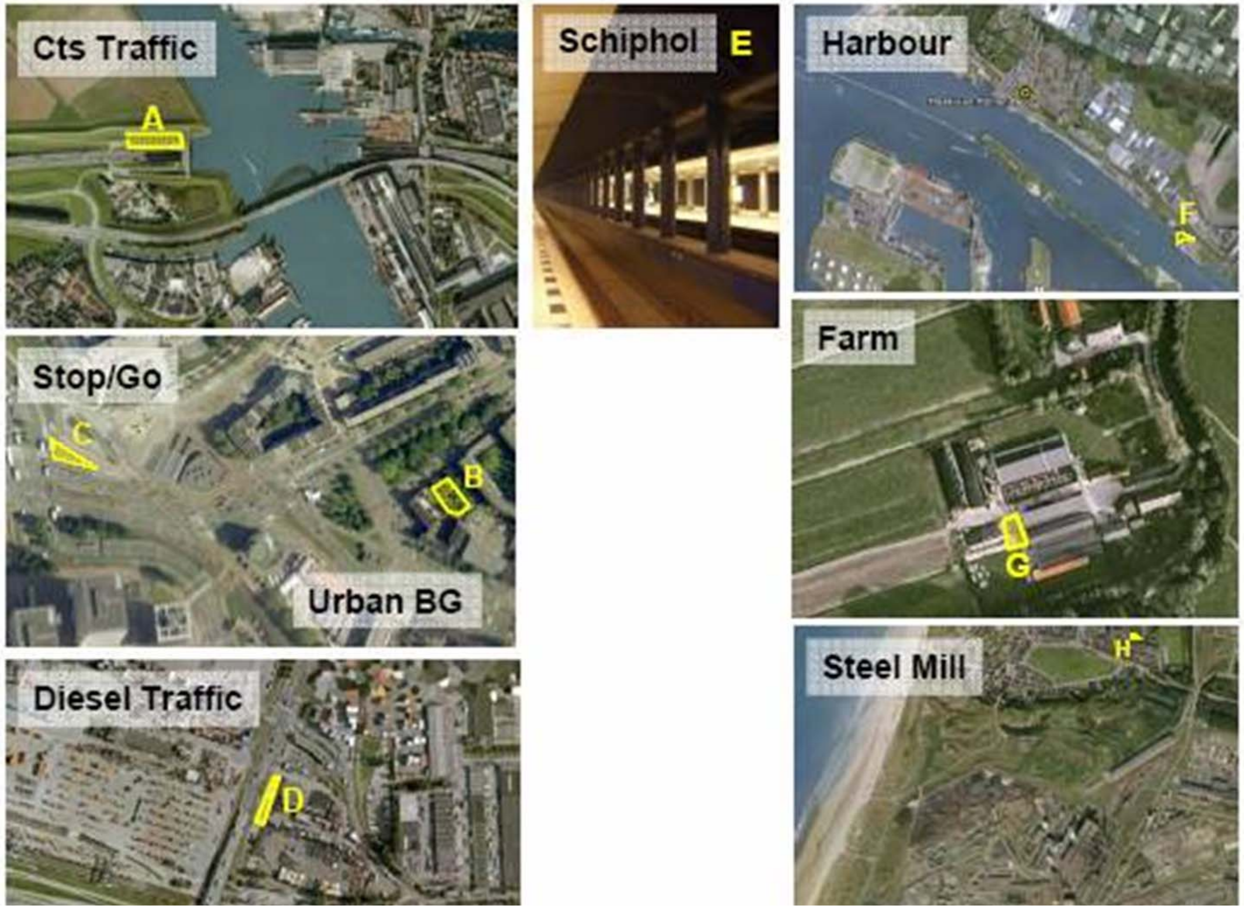




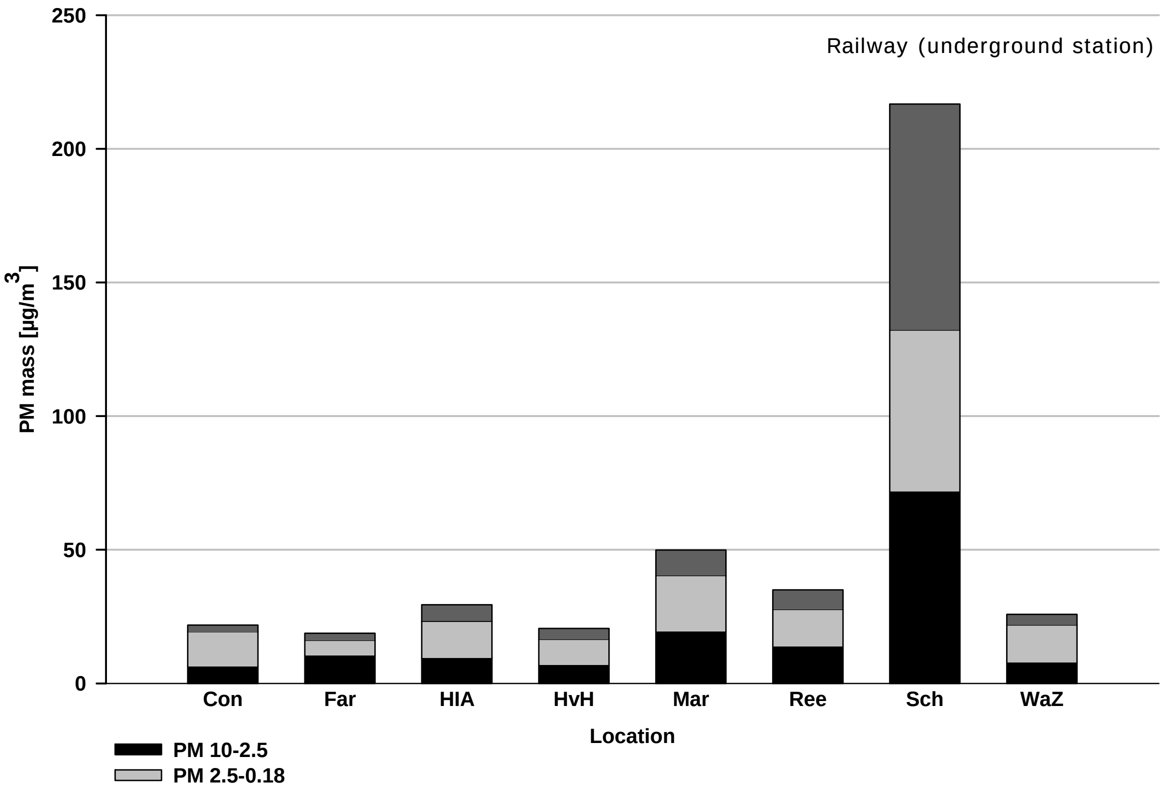
bient Particulate Matter (PM) Size Distributions;



ation with source of emission

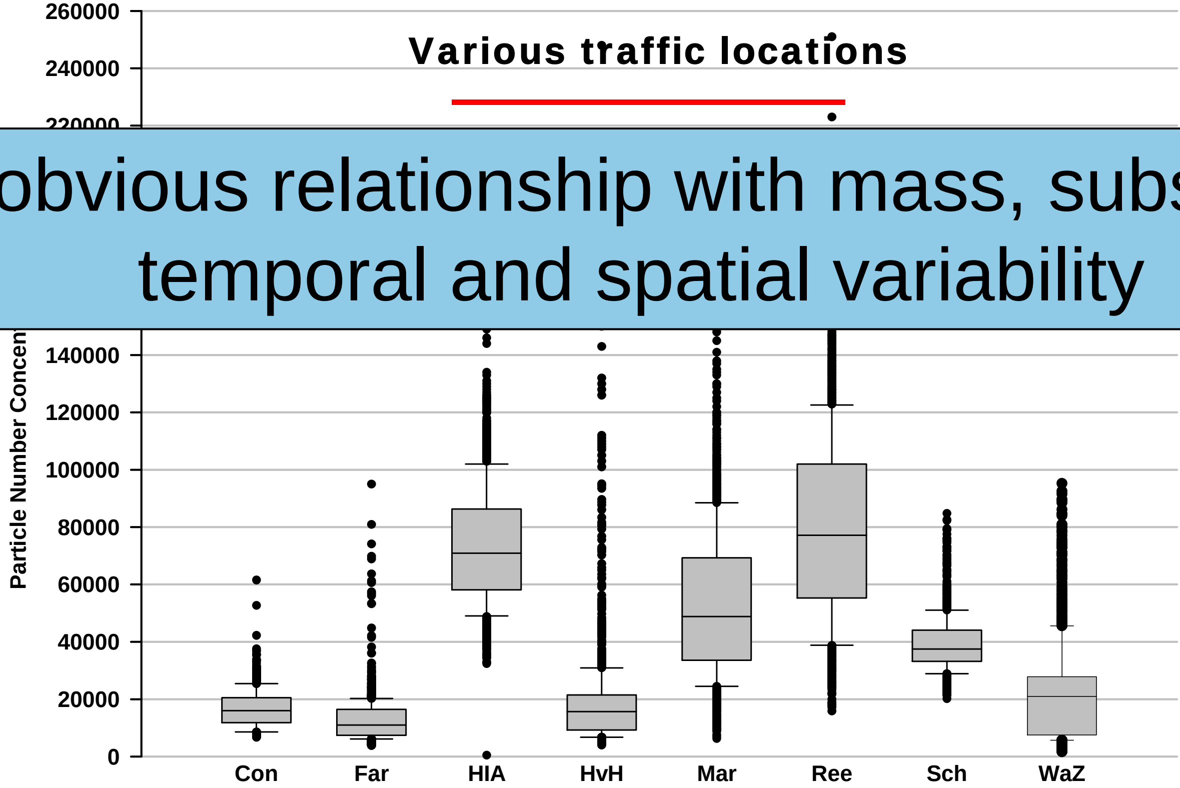


l mass



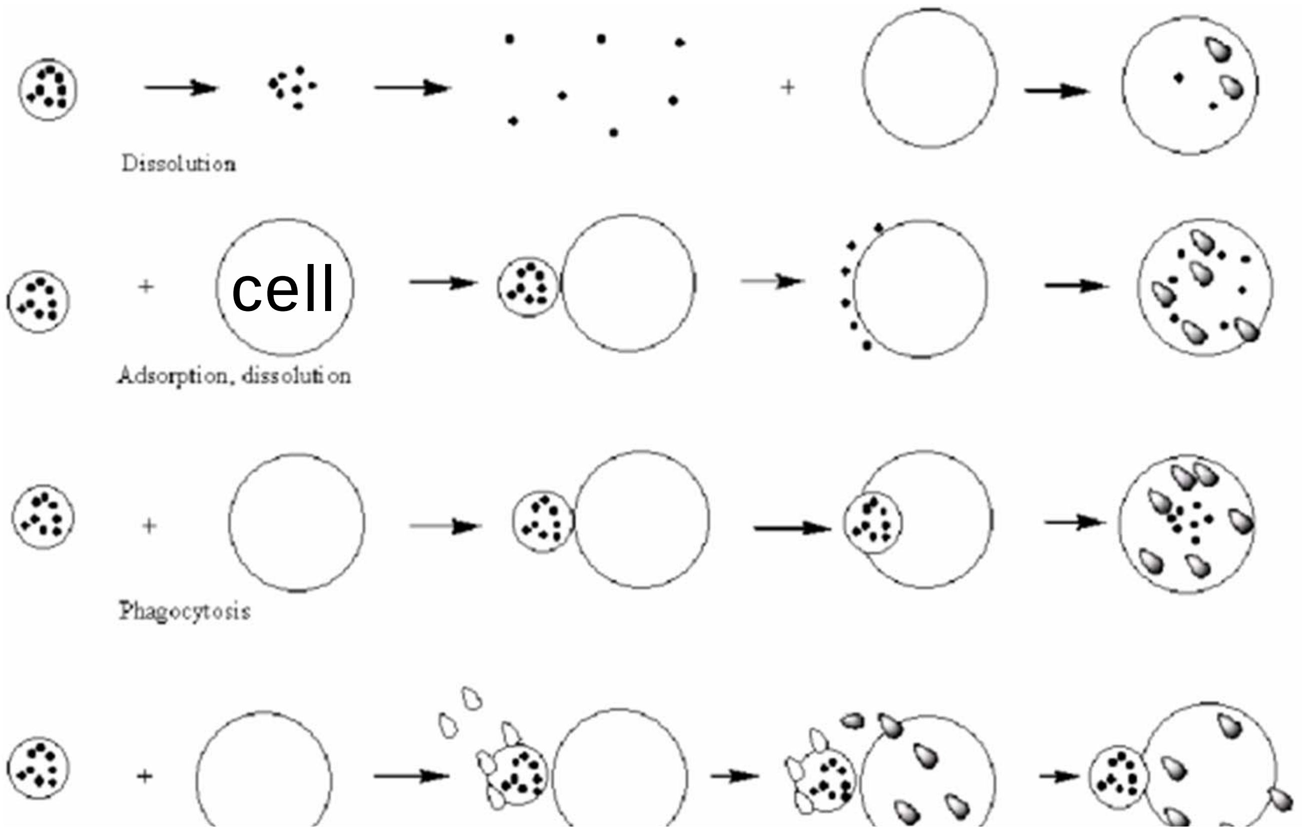
Railway (underground station)
PM mass [µg/m3]
0
50
100
150
200
250
Con
Far
HIA
HvH
Mar
Ree
Sch
WaZ
Location
PM 10-2.5
PM 2.5-0.18

mber counts



No obvious relationship with mass, substantial temporal and spatial variability

any things can happen during transport to target



operties

hape including aggregates

ize

rigin

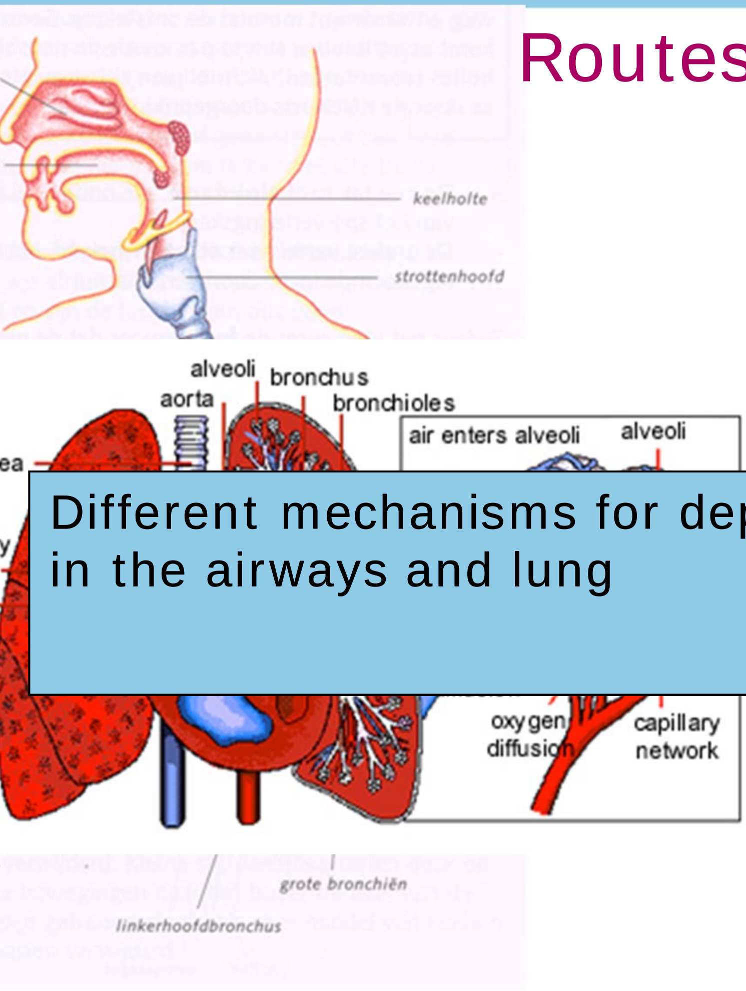
arge numbers, small mass

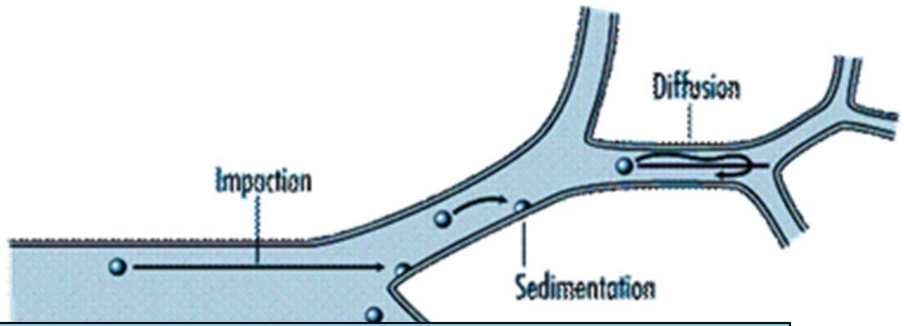
hange over time

this all affects the response of the human body upon exposure

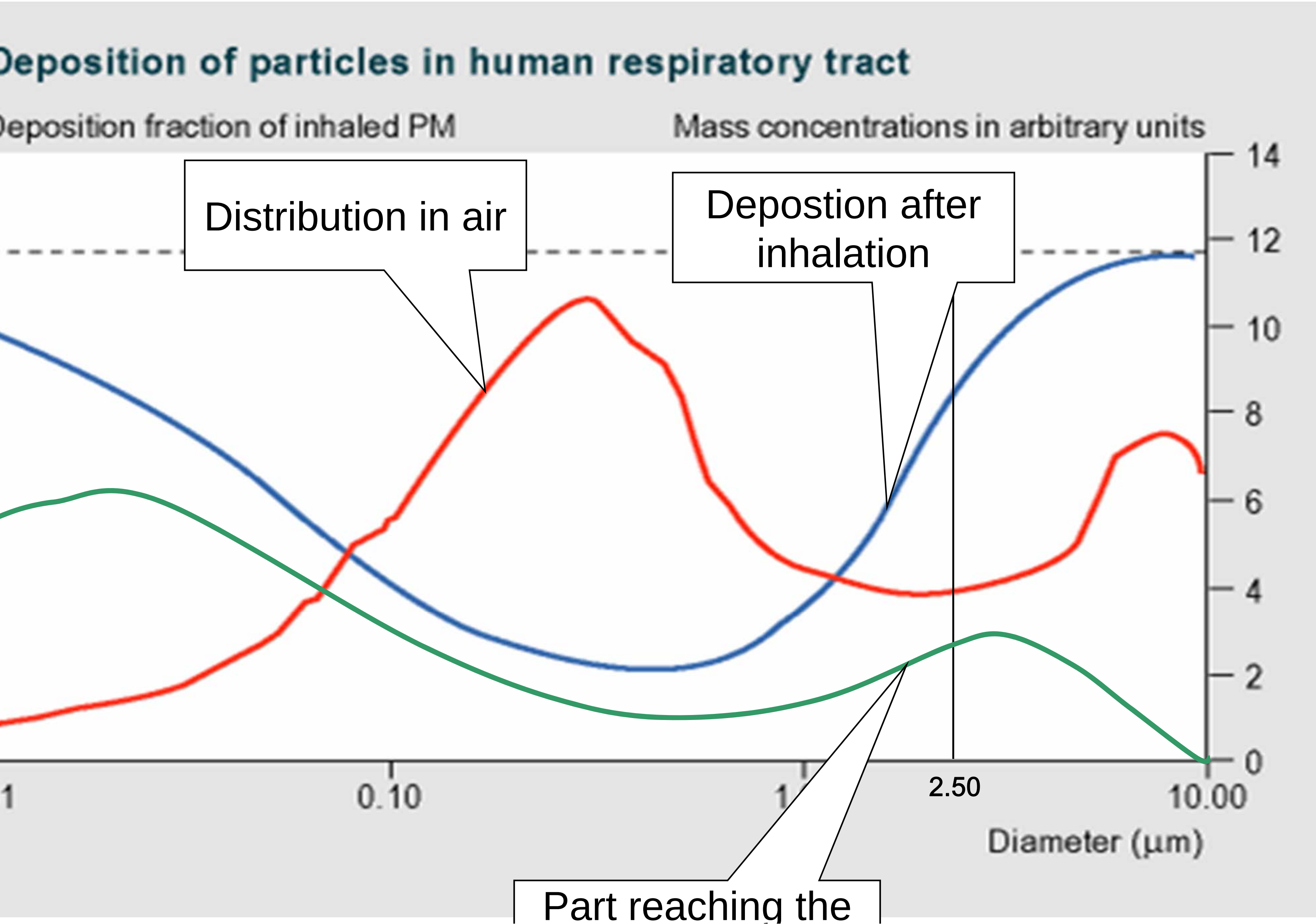
where do the go to?

Routes of exposure to particles



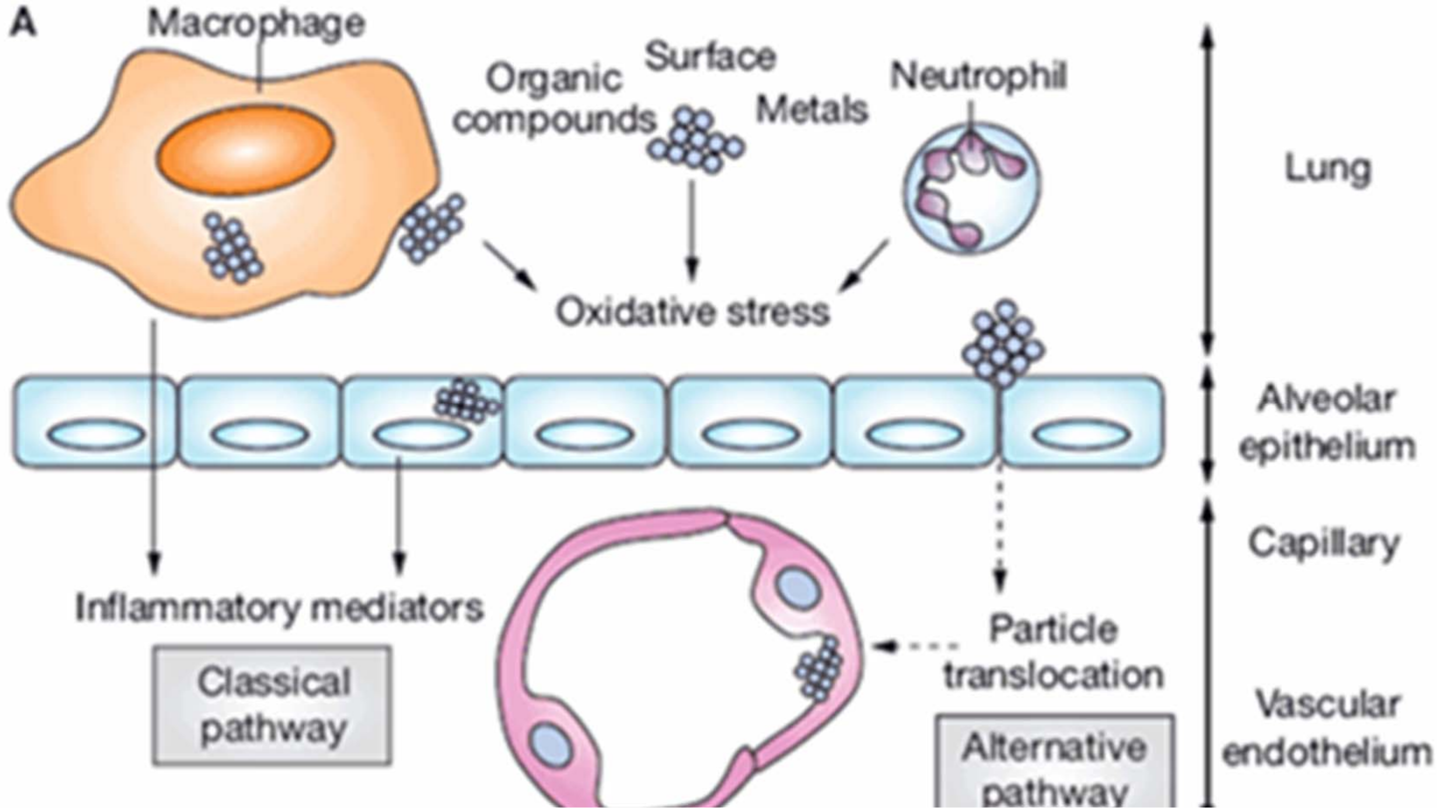


Different mechanisms for deposition for nano versus micro in the airways and lung

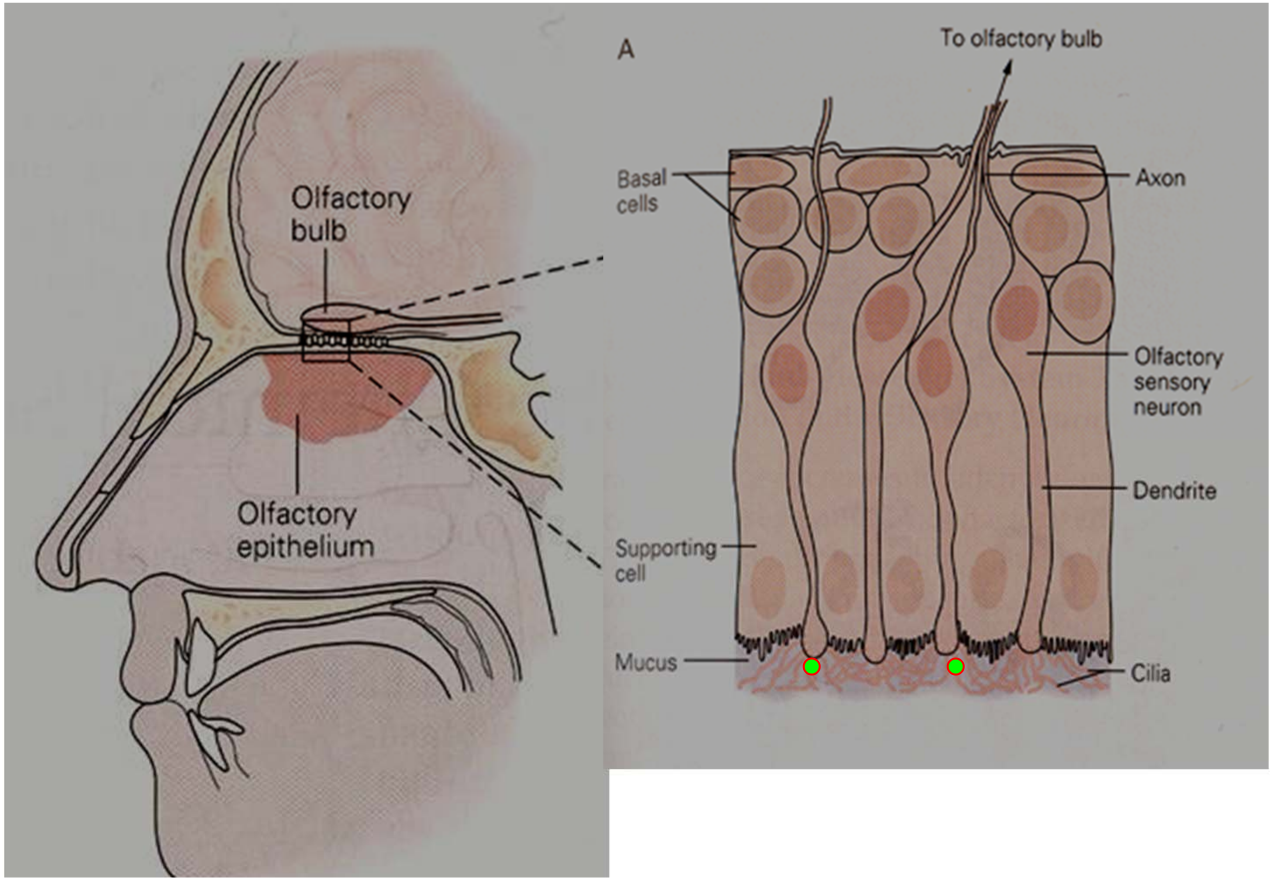


Deposition of particles in human respiratory tract
Deposition fraction of inhaled PM
Mass concentrations in arbitrary units
Distribution in air
Depostion after inhalation
Part reaching the
14
12
10
8
6
4
2
0
0.10
1
2.50
10.00
Diameter (µm)

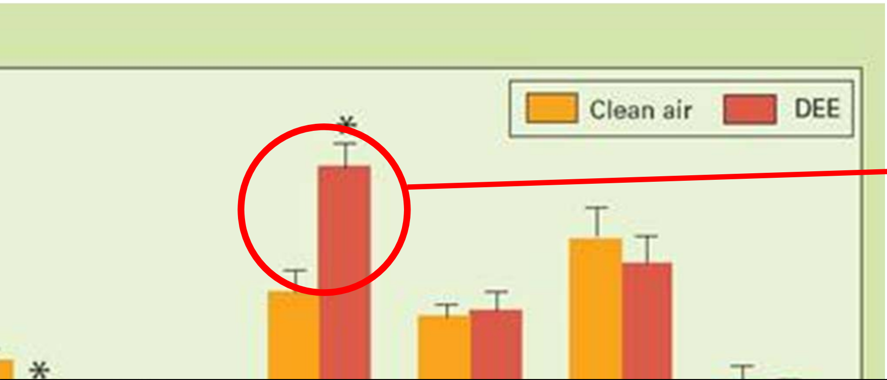
anslocation into the blood

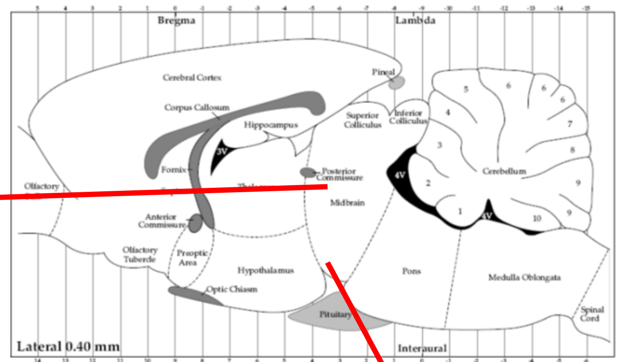


slocation into the brain



r pollution & Neurodegeneration

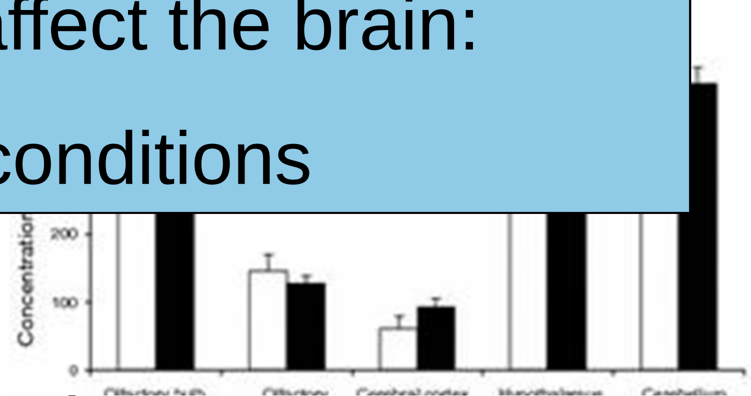




Particles can reach or affect the brain:

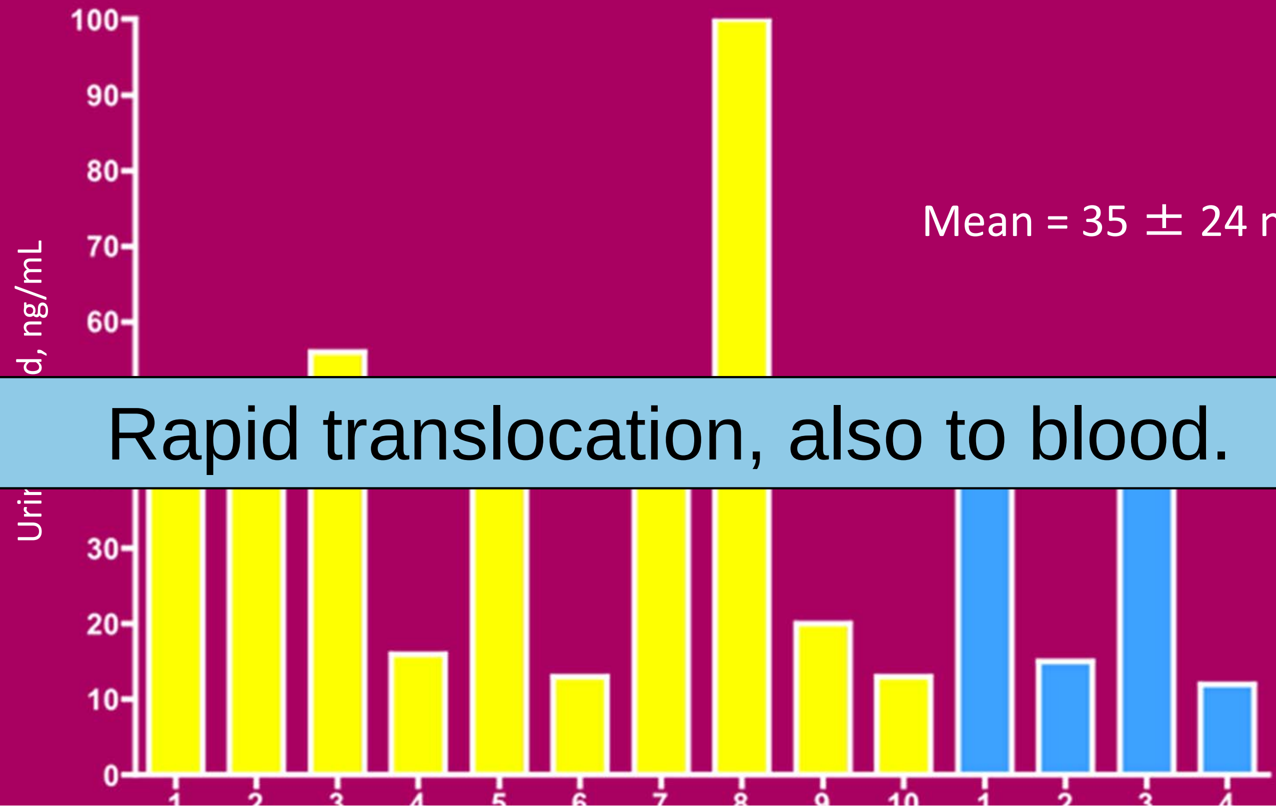
(oxidative) stress conditions

erlofs-Nijland et al, PFT 2010



o nanoparticles translocate? Nanogold 5-15 nm

e Gold Concentration (24 hr collections)



Mean = 35 ± 24 ng/mL

Rapid translocation, also to blood.

Examples of effects of nanoparticles

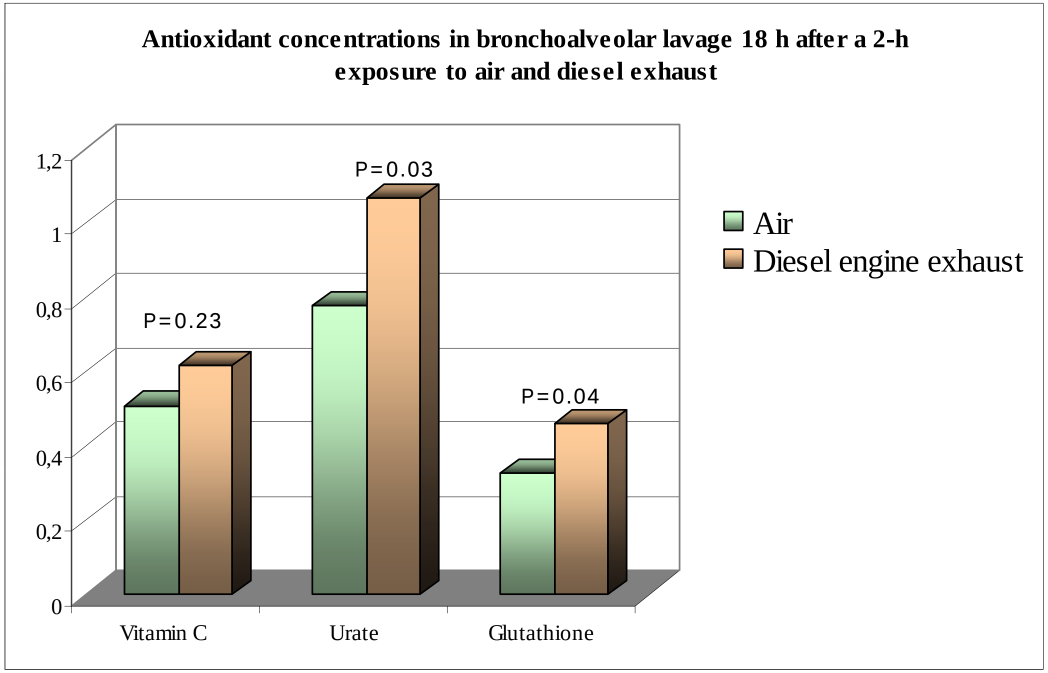
Oxidant capacity

ti-oxidants e.g. ascorbate, glutathione

: 359–365
6.06.00136904
ls Ltd 2006

/ antioxidant and inflammatory
nses to diesel exhaust exposure in
y humans

g*,√, I.S. Mudway*,¶,√, J.L. Brown#, N. Stenfors*, R. Helleday*,
n¶, S.J. Wilson#, C. Boman+, F.R. Cassee§, A.J. Frew#, F.J. Kelly¶,
m* and A. Blomberg*

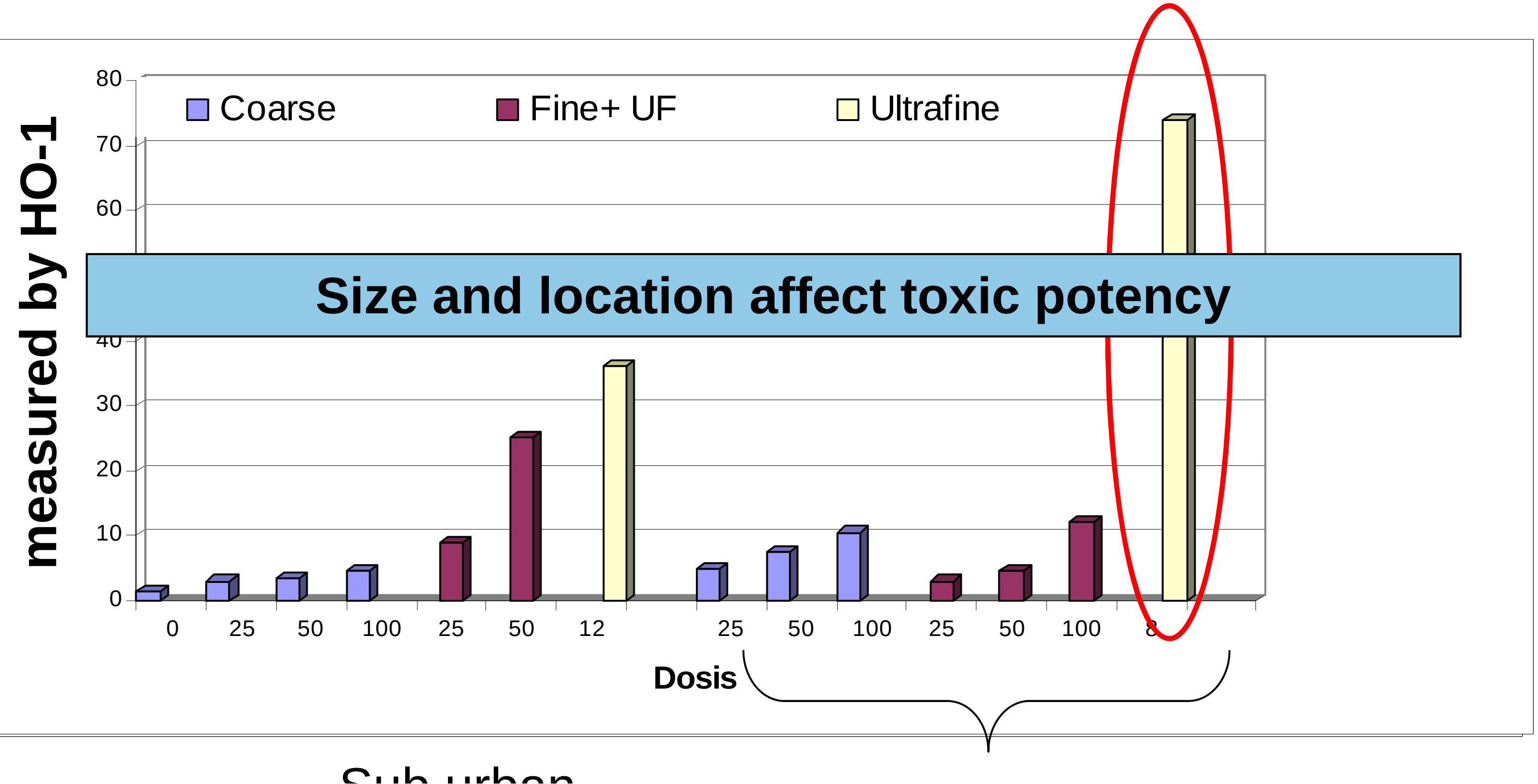


- Exposures dominated by combustion-derived nanoparticles
- Good correlation anti-oxidants and clinical symptoms

Fine + coarse = 90% of PM10

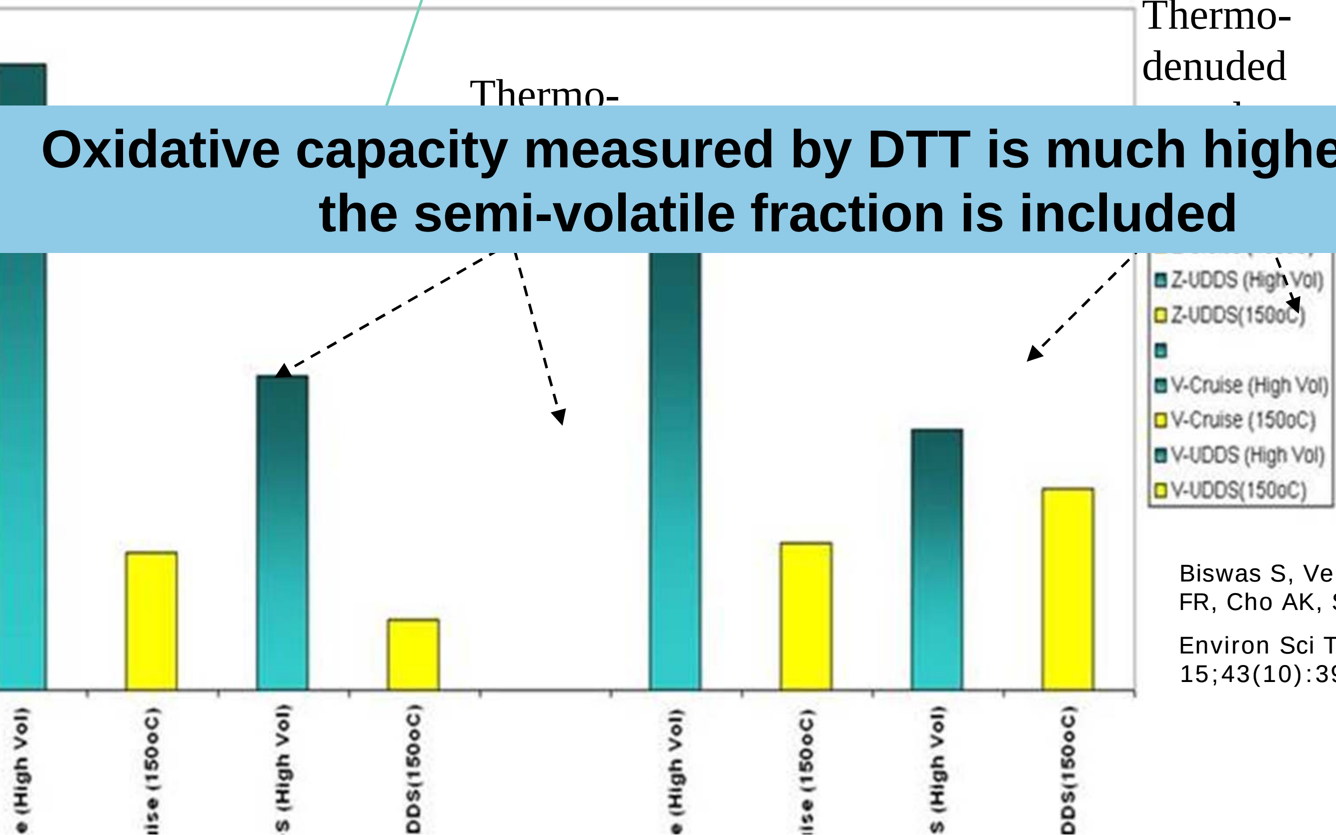
Li et al EHP, 2003

zard identification per PM size fraction



Samples including semivolatile PM

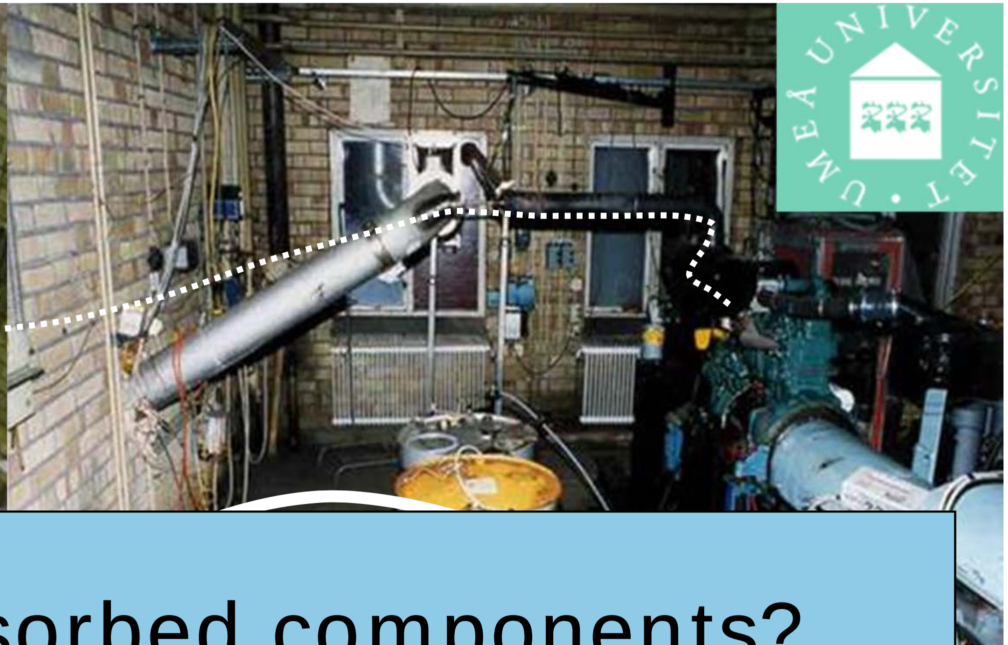
ganic component on PM



Oxidative capacity measured by DTT is much higher when the semi-volatile fraction is included

Biswas S, Verma V, Schauer JJ, Cassee FR, Cho AK, Sioutas C.

Environ Sci Technol. 2009 15;43(10):3905-12.



Carbon core or absorbed components?

Miller et al., European Heart Journal, 2011 in press

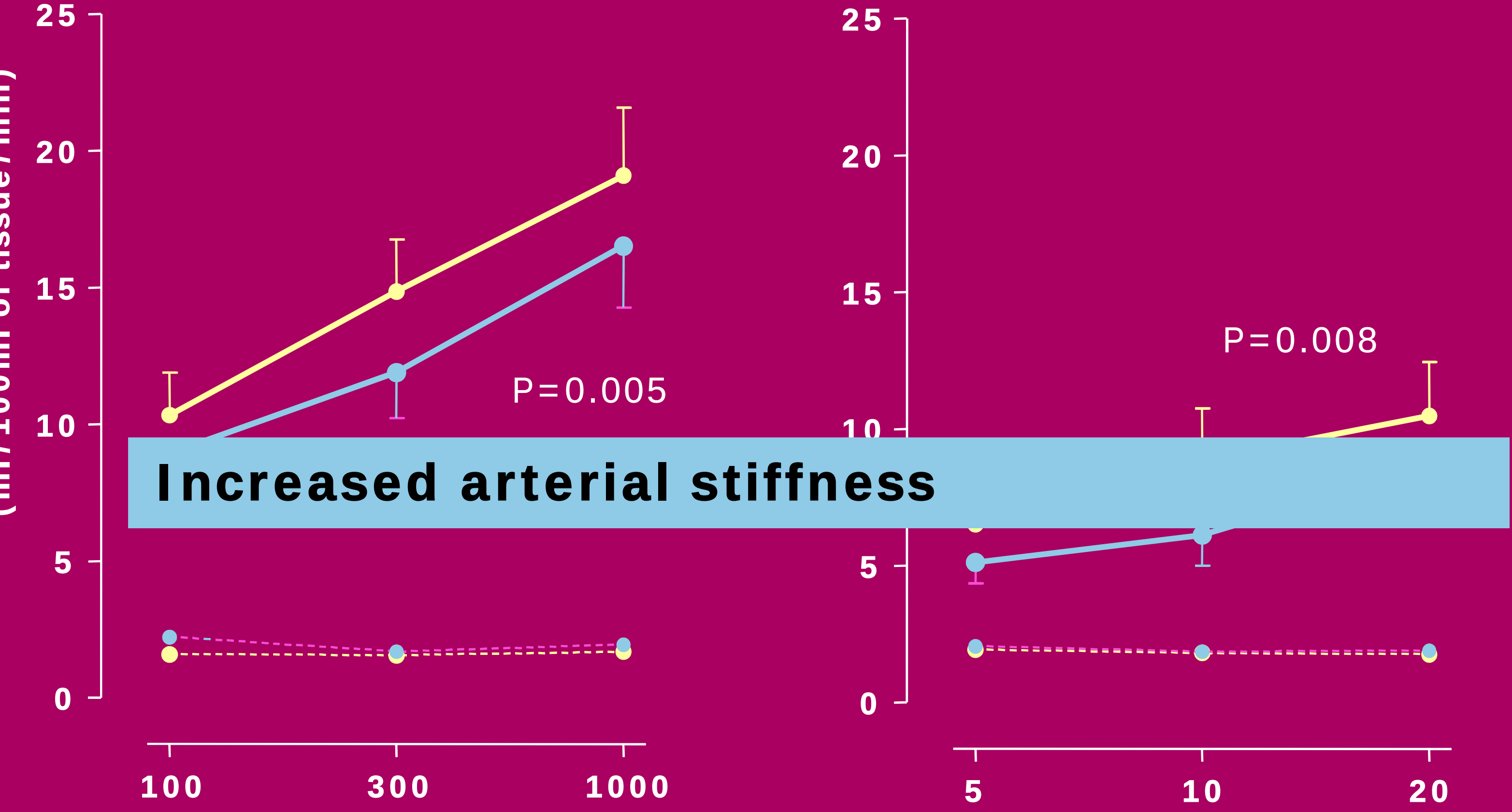
hr exposure of health volunteers

	DIESEL	FILTER	Carbon	AIR
s filter (µg/m³)	348±16	6±4	70±7	<1
cles (number/cm³)	1.2±0.1e6	1.7±0.6e3	3.9±0.1e6	2.9±0.7
cles diameter (nm)	67±1	-	37±3	-
ppm)	3.5±0.2	3.2±0.1	0.2±0.1	0.1±0.1
ppm)	0.3±0.0	0.3±0.1	0.1±0.0	0.1±0.0
(ppm)	0.2±0.0	0.2±0.0	<0.01	<0.01
(ppm)	0.6±0.0	0.6±0.0	<0.01	<0.01
perature (°C)	18.7±0.1	18.6±0.1	18.7±0.2	18.2±0.2

helium-dependent
ilatation

Miller et al., European Heart Journal, 2011 in press

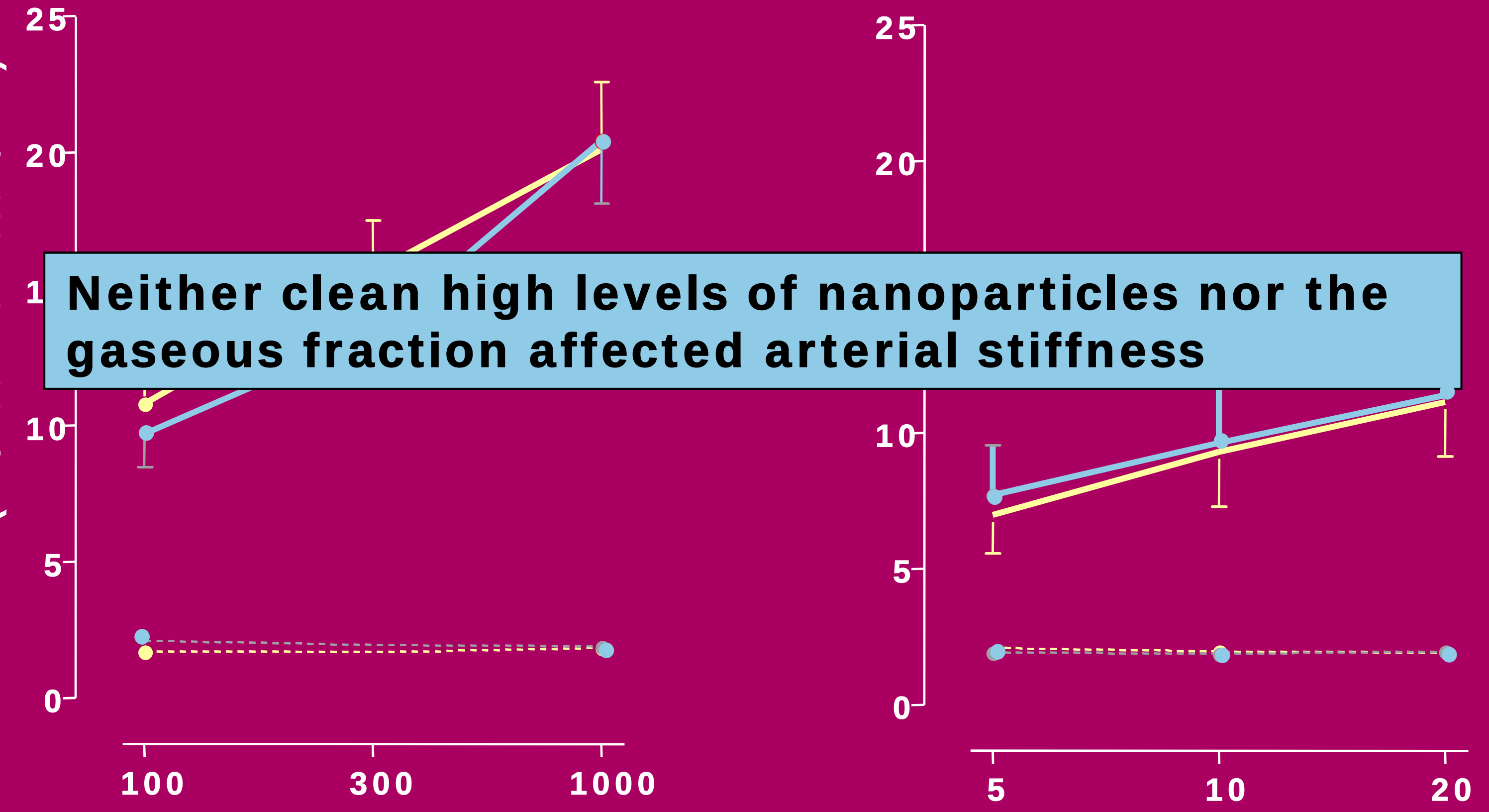
FILTERED AIR *versus* **DIESEL EXHAUST**



helium-dependent
ilatation

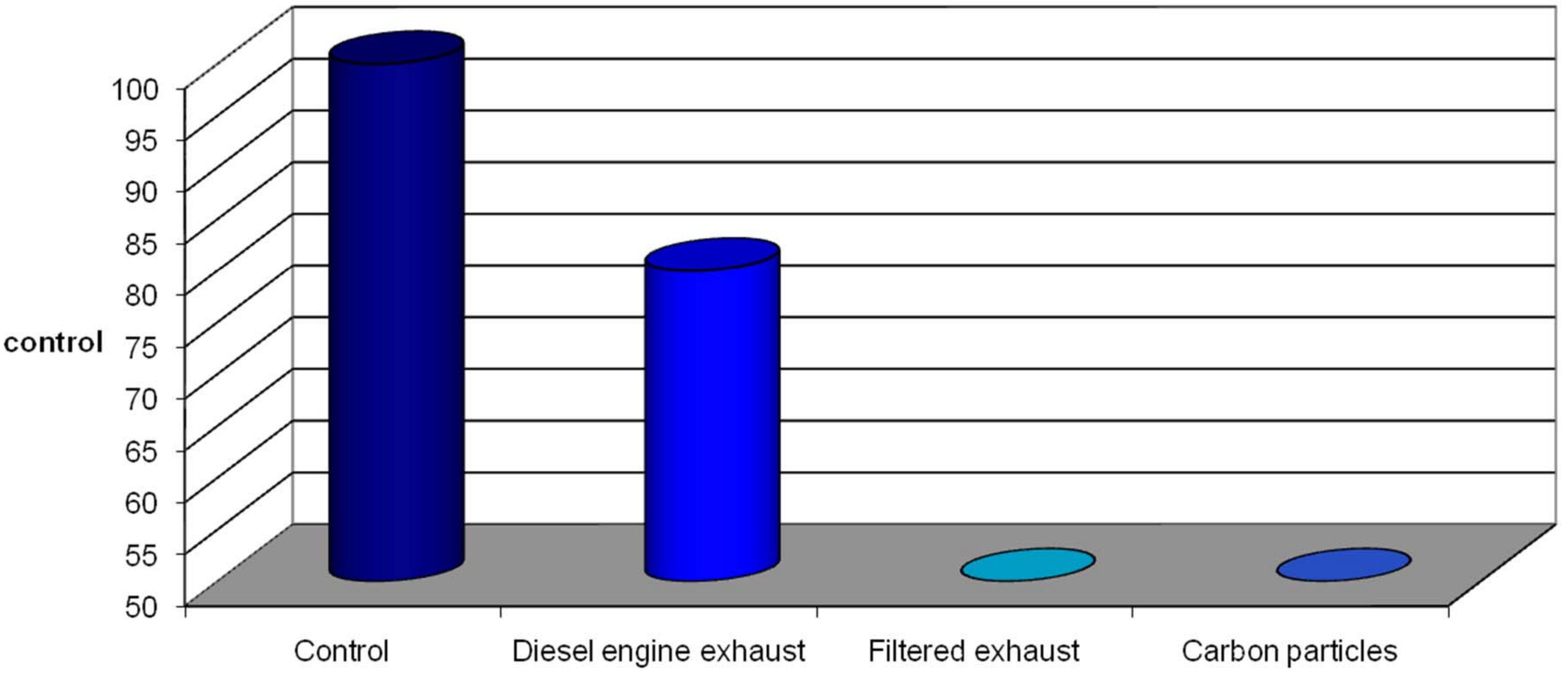
Miller et al., European Heart Journal, 2011 in press

LTERED AIR *versus* **FILTERED DIESEL (same for carbon nanoparticles)**

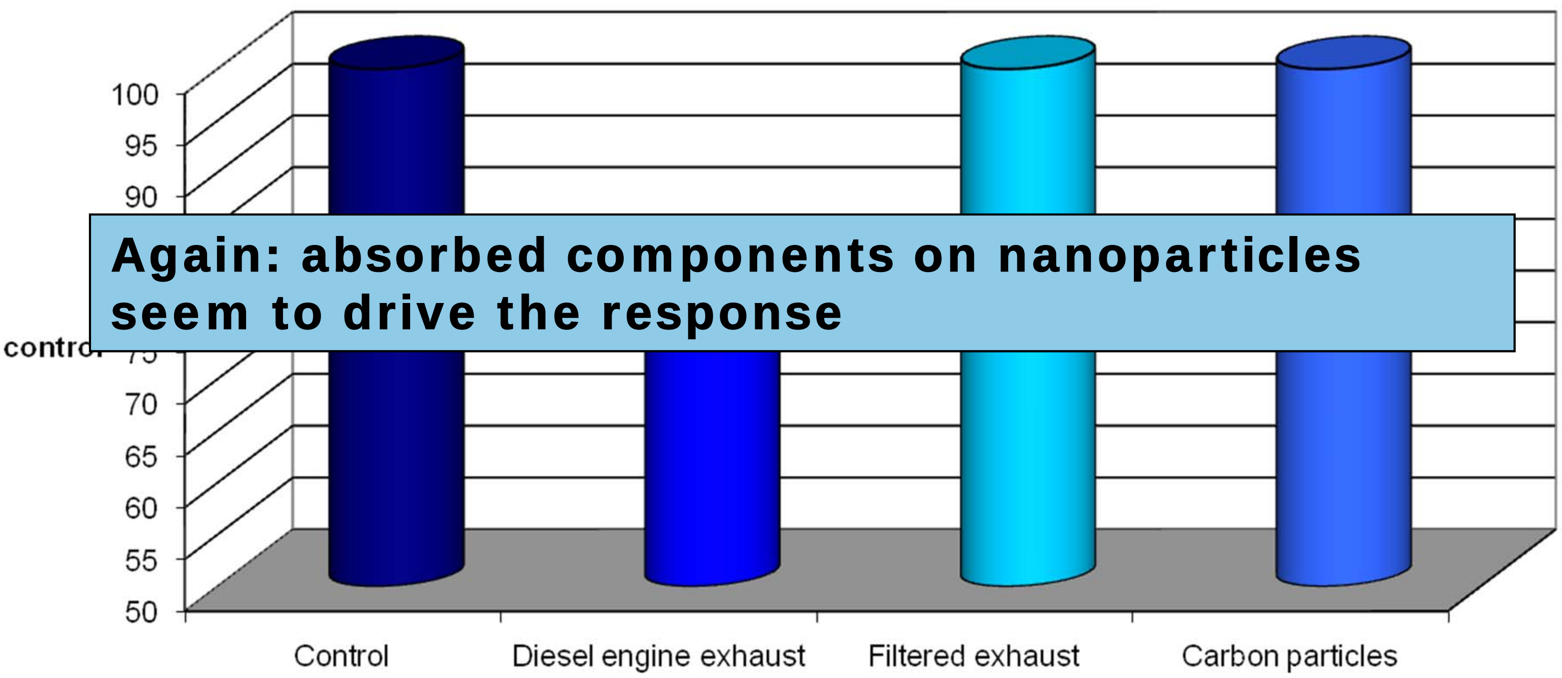


Neither clean high levels of nanoparticles nor the gaseous fraction affected arterial stiffness

Capacity to handle blood clotting



Capacity to handle blood clotting



Again: absorbed components on nanoparticles seem to drive the response

l world air pollution studies

rvention using a face mask

.angrish, Nick Mills, Dave Newby, Flemming Cassee et al
and Fibre Toxicology 2009

uring exercise

		Without Mask	With Mask
	Energy expenditure, kcal	340 (314 – 367)	364 (304 – 426)
	Systolic blood pressure, mmHg	121 (115 – 127)	114* (108 – 120)
	Diastolic blood pressure, mmHg	81 (75 – 87)	79 (74 – 83)
	Mean arterial pressure, mmHg	94 (89 – 99)	90 (86 – 94)

24-hour Analysis

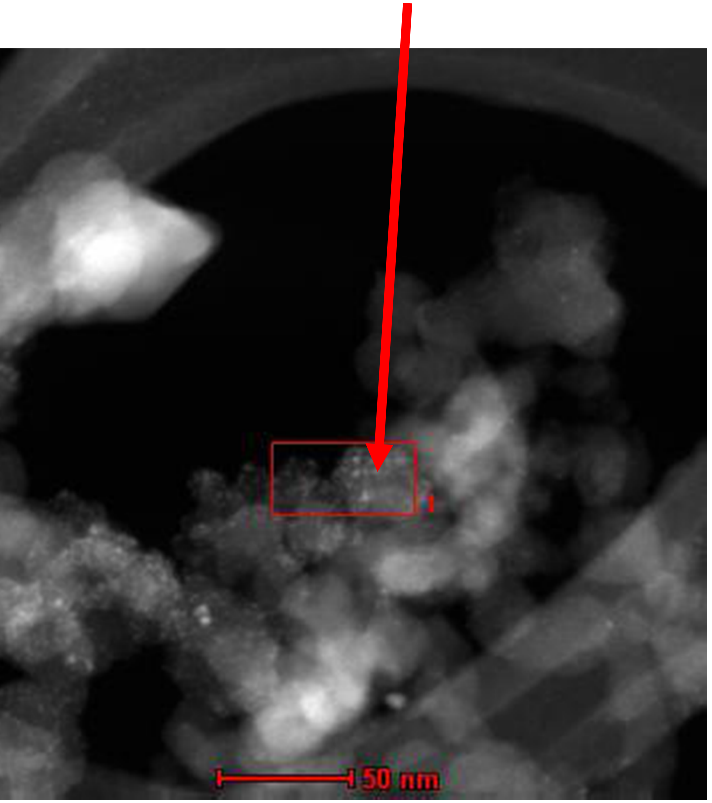
		Without Mask	With Mask
	Data validity, %	95.9	95.0
	Average NN	829	850
Heart rate variability	SDNN, ms	61.2 (54.9 – 67.5)	65.6* (59.0 – 72.2)
	Triangular Index	12.9 (11.9 – 13.9)	13.8 (13.0 – 14.5)
	LF-power, ms^2	816 (628 – 1004)	919* (717 – 1122)
	HF-power, ms^2	460 (325 – 595)	485 (400 – 569)
	LFn, ms	62.8 (56.7 – 68.9)	64.5 (60.6 – 68.4)

mmediate reduction BP and later improved heart performance

esel & nano cerium oxide

sure data: physical characteristics

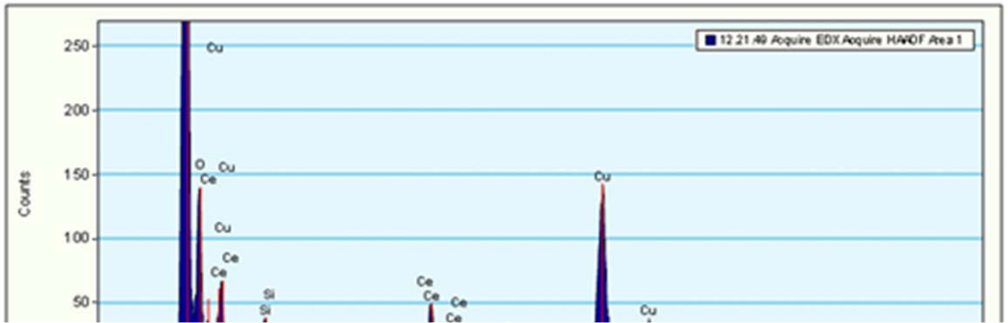
Mass (graphimetic) µg/m3	Mass (optical) µg/m3	Surface area µm2/cm3 per TB region	Number counts #/cm3 (x 10E6)	Diameter nm
1602 ± 98	1872 ± 132	4064 ± 114	5.7 ± 0.6	70 ± 1.8
1810 ± 36	1956 ± 26	4556 ± 113	5.8 ± 0.3	82 ± 1.8
1778 ± 252	1900 ± 32	4363 ± 213	4.8 ± 2.4	81 ± 1.8
1772 ± 65	1972 ± 43	3160 ± 429	4.4 ± 2.6	95 ± 1.8
1741 ± 153	1925 ± 79	4018 ± 605	5.3 ± 0.1	82 ± 1.8
1525 ± 105	1466 ± 747	3488 ± 484	3.1 ± 0.5	72 ± 1.8
1772 ± 73	1922 ± 131	4040 ± 169	3.6 ± 0.3	83 ± 1.8
1804 ± 137	1894 ± 69	3948 ± 350	3.0 ± 1.5	81 ± 1.8
1816 ± 159	1986 ± 9	3132 ± 450	2.9 ± 1.6	96 ± 1.8
1740 ± 162	1817 ± 408	3636 ± 517	3.6 ± 0.5	83 ± 1.8
100%	94%	90%	69%	101%



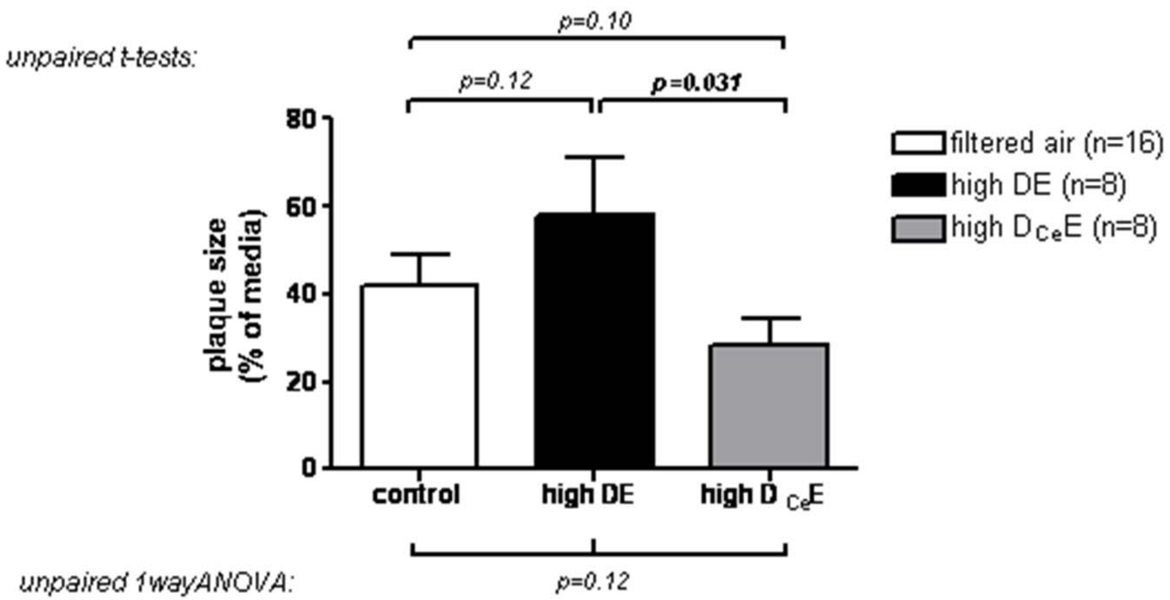
The small white dots are cerium- containing particles.

Typical size of the nano-particles: 1-2 nm.

Up to 6 ug/mg particle mass is cerium



of diesel exhaust on the size of atherosclerotic
es in the brachiocephalic artery

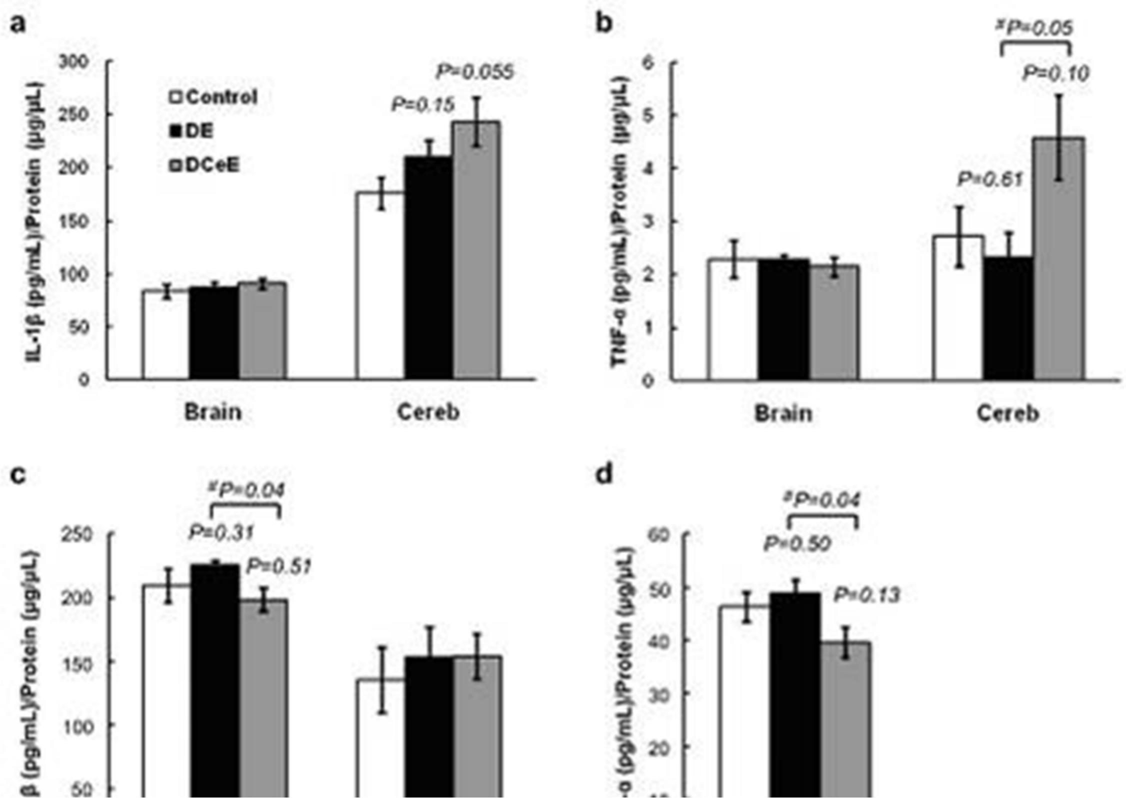


Mean±S.E.M. (n=8-16), p-values shown are results from
the corresponding statistical test indicated to the left of the

creased 'stress; in the brain due to nano-cerium

ain

ver

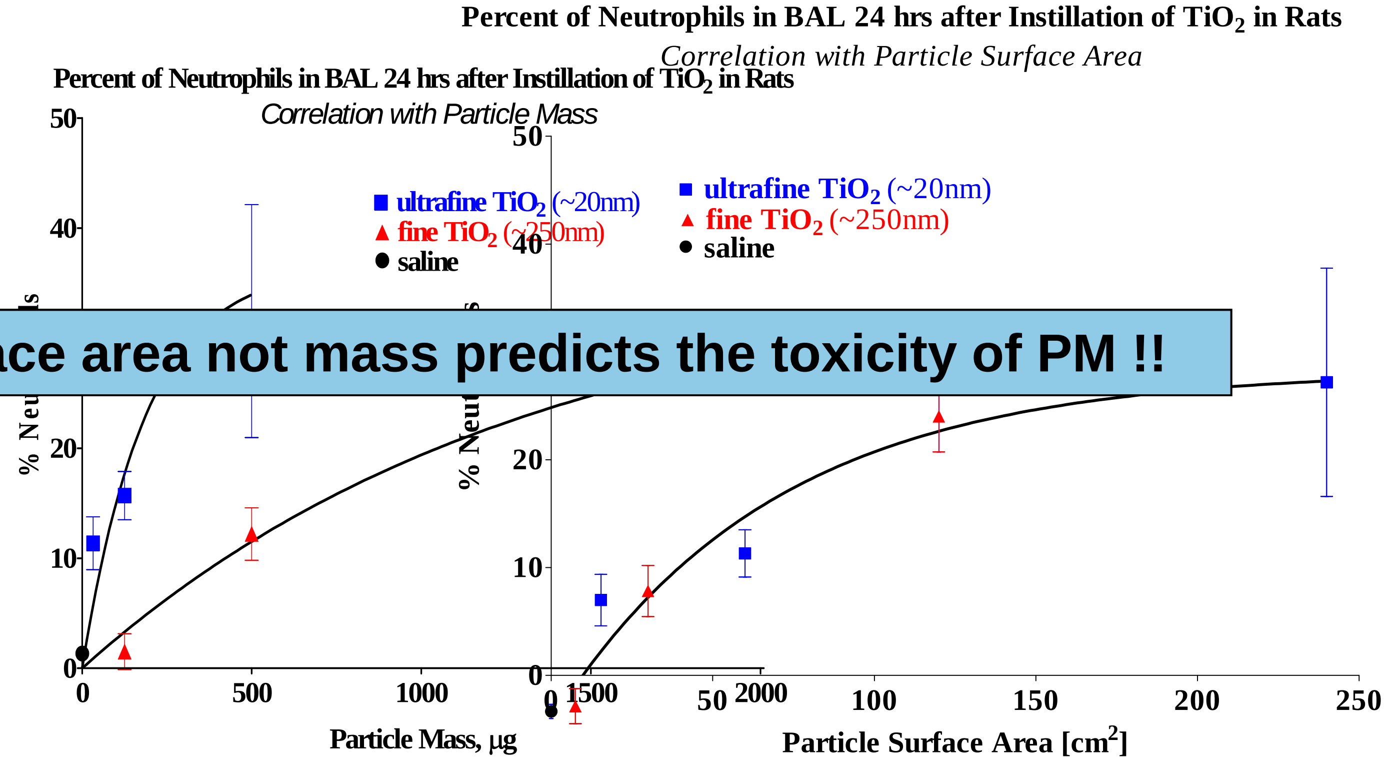


Paracelsus

"All things are poison and nothing is without poison. Solely the dose determines that a thing is not a poison."

Question in case of nanoparticles: what is the metric for ‘dose’?

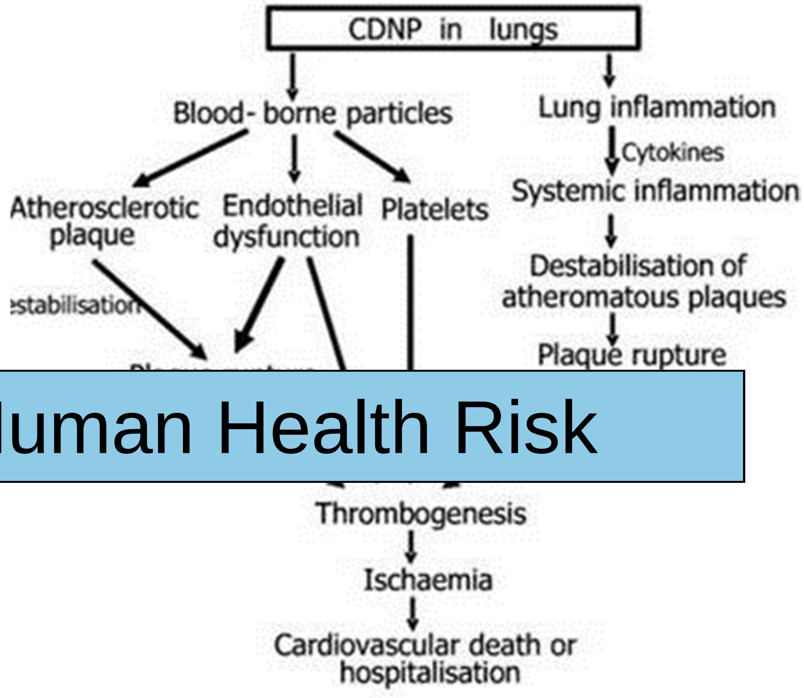
Inflammation after 24 hrs in lungs of rats



Surface area not mass predicts the toxicity of PM !!

nclusions

- ombustion derived anoparticles can be a hazard t current levels
- M mass not per se the best



Donaldson et al, Part Fibre Toxicol. 2005; 2: 10.

Hazard x Exposure = Human Health Risk

- ikely that components on urface of nanoparticles metals or organics) contribute o health effects
- essons for better indicators in pidemiology, e.g. Number,

knowledgement

riam Gerlofs-Nijland, Ilse
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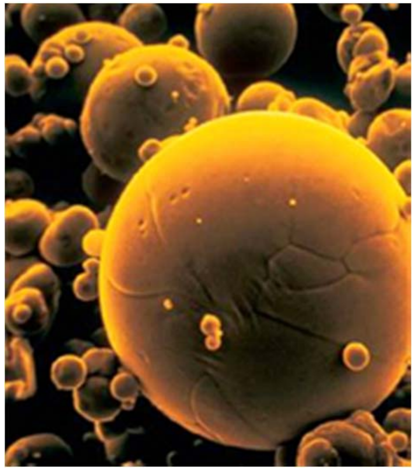

hank you for your attention