Toxicokinetics of Inhaled Nanoparticles

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Toxicokinetics of Inhaled Nanoparticles

Deposition – Dissolution – Translocation

- **Metal oxides: nano-TiO_2, nano-ZnO, nano-SiO_2**
- **Recent study on: Carbon black**

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BAuA-funded Project: 28-Day Inhalation Toxicity Study with 3 TiO_2 Varieties (NM-103, NM-104, NM-105)

Objectives

- To mimic an occupational exposure scenario (dry dispersion technique)
- Study on disintegration of agglomerates (TEM analysis)
- Identification of the respiratory cell types responsible for uptake of these particles

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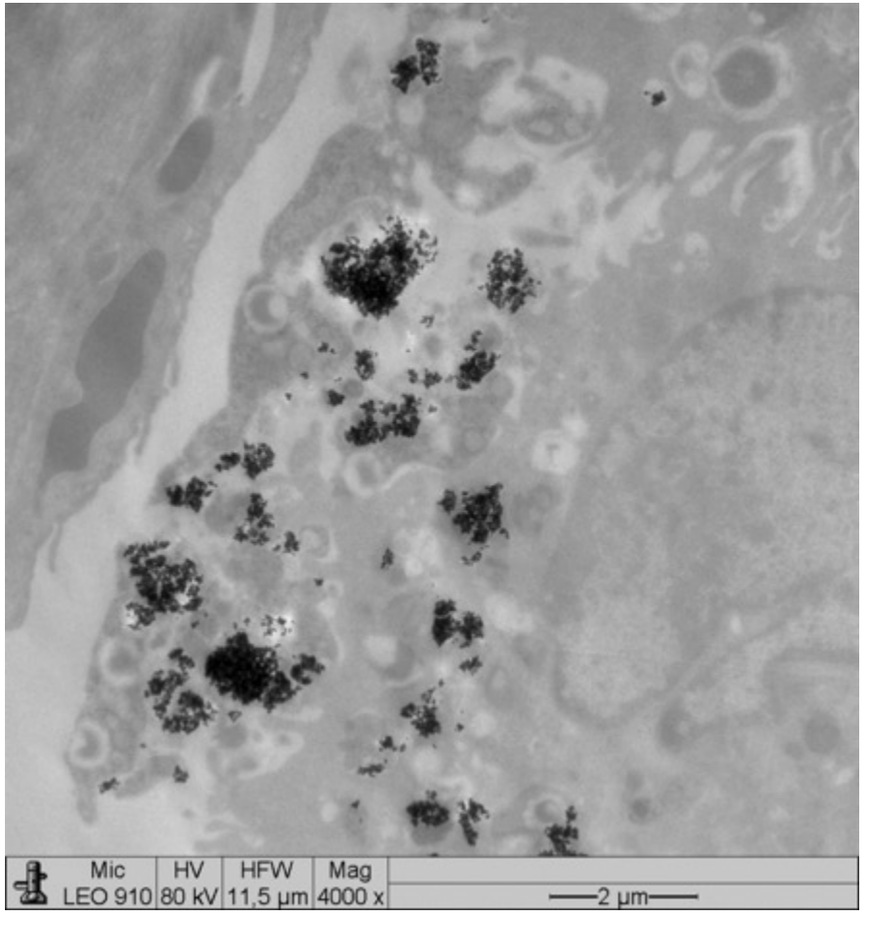
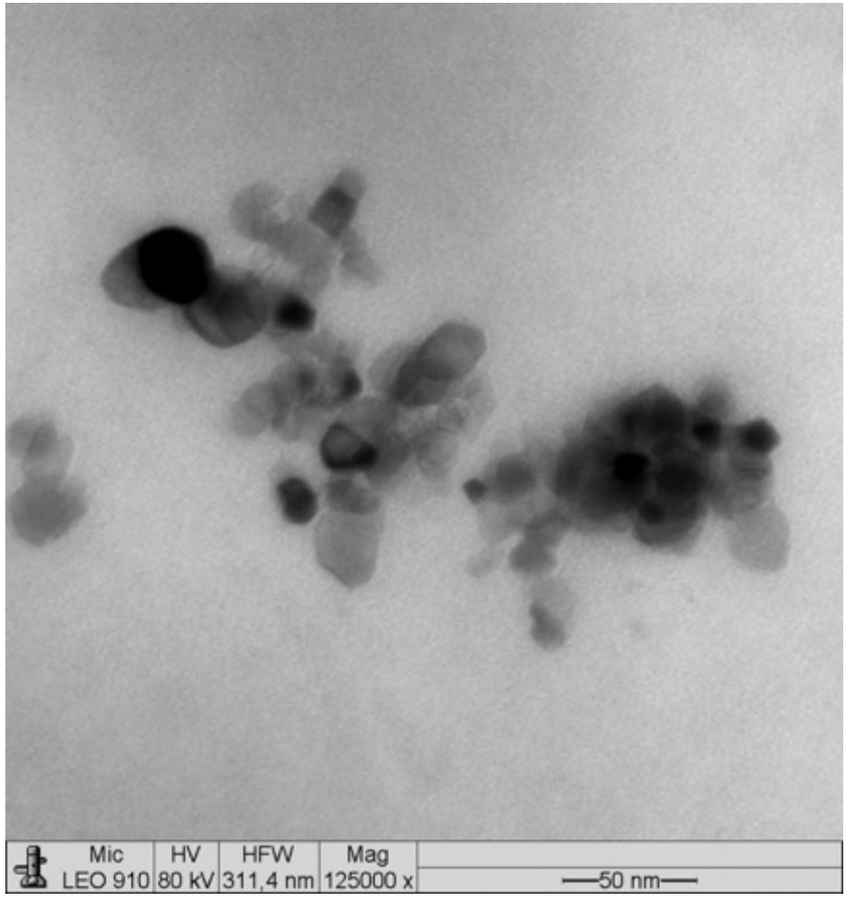
Exemplary pictures of transmission electron microscopy



Titanium particles (NM-103) within an intraalveolar macrophage



High magnification of titanium particles (NM-104) within a macrophage

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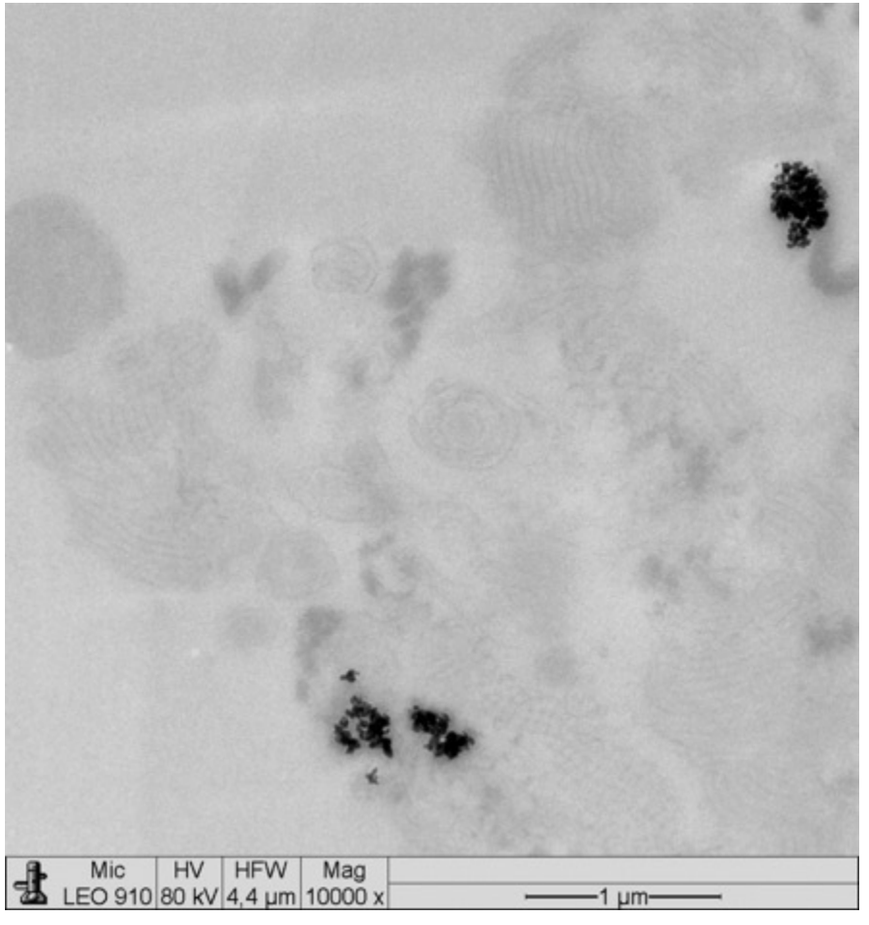
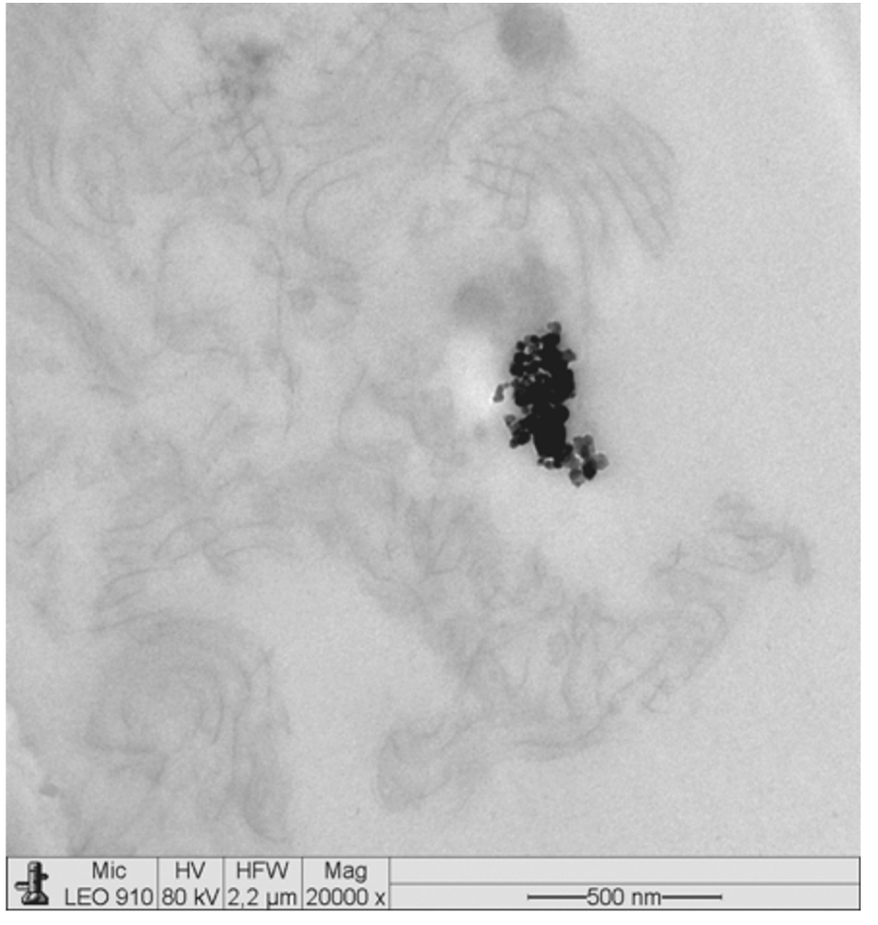
Exemplary pictures of transmission electron microscopy



Titanium particles (NM-103) attached to intraalveolar surfactant



Titanium particle (NM-105) attached to intraalveolar surfactant

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Summary

- **Retained particle mass**

  → Retention analysis data matched the values predicted by MPPD model.

  → The translocation potential from lungs was very small.

  → Solubility of test items is limited by a given maximum under the the conditions of the lung ambience (5.5%, 2.2% and 0.9% in the low/mid/high dose groups, resp.)

- **TEM**

  → Intraalveolar MPh are the most prominent compartment of particle detection (secondary: Pneumocytes type I / free particles) .

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Biokinetics of nano-ZnO (NM-111) and nano-SiO_2 (NM-200) after Inhalation in Rats

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90-Day Study: Absolute Zn content in organs, blood, urine and feces

NM-111: ZnO coated with triethoxycaprylylsilane (2%)

Day 1 post-exposure

	Clean air		Z-Cote® HP1		Z-Cote® HP1		**Z-Cote® HP1**		microscaled ZnO	
	control		0.3 mg/m³		1.5 mg/m³		**4.5 mg/m³**		4.5 mg/m³	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Day 1 postexposure	(µg/Organ)		(µg/Organ)		(µg/Organ)		(µg/Organ)		(µg/Organ)	
LALN	2.60	1.55	2.86	0.78	2.01	0.40	2.53	0.70	2.98	1.68
MSLN	2.62	0.77	3.52	0.60	2.72	1.79	2.20	0.52	4.43	2.74
Brain	23.2	0.9	22.9	0.7	21.4	1.3	21.4	1.4	*20.8	1.4
Kidneys	58.1	3.5	56.2	5.4	*49.4	3.4	*49.5	4.1	51.7	6.7
Liver	276	19	320	29	287	27	287	20	302	80
Lung	19.9	0.2	20.4	0.6	22.2	2.0	**35.8	1.7	22.4	1.9
Blood	327	37	278	15	291	17	368	15	355	61
	(µg/16h)		(µg/16h)		(µg/16h)		(µg/16h)		(µg/16h)	
Urine	2.90	1.91	5.68	5.04	3.63	1.92	2.40	0.72	4.46	1.84
Feces	565	185	472	111	466	259	673	104	475	210

90-Day Inhalation Test + 1-mth recovery

Name NM	Mass balance / Recovery Bioavailability	Biodistribution Tissue levels	C,t-curve(s) T1/2, ke	Variation
NM-111	Deposited test items were eliminated within 1-mth post-exposure period	**Lungs:** approx. 2% of deposited mass after end of exposure; Tissue levels not increased	**$T_{1/2}$ < 1 wk**	-
µ-ZnO		Lung and tissue levels not increased	**$T_{1/2}$ <1 day**	-

NM-111: Coated triethoxycaprylylsilane (2%)

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14- and 90-Day Study: Summary ZnO

Zn chemical analysis:

Detectable only at day 1 post-exposure;
statistically significant in lungs for NM-111

Not longer increased at day 14 or day 29 post-exposure

ZnO particles in tissues not detectable by TEM

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14-Day Study: Summary SiO_2 (NM-200)

→ precipitated synthetic amorphous silica

Toxicokinetics

- Si analysis: Detectable only in lungs - Day 1 and 14 post-exposure
- SiO_2 particles detectable in lungs/LALN up to 14 day post exposure

  – not detectable in remote organs by TEM

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90-Day Study: Summary SiO_2 (NM-200)

→ **precipitated synthetic amorphous silica**

Toxicokinetics

- Si analysis: Detectable only in lungs - Day 1, 29 and 91 post-exposure
- SiO_2 particles detectable in lungs/LALN up to 91 day post exposure

  – not detectable in remote organs by TEM
  (nasal epithelium, trachea, larynx, liver, spleen, kidney and mesenteric lymph node)

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Toxicokinetic Study on Carbon black

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Objectives

- **To perform a study for regulatory purposes**
- **To compare two high-volume grades of carbon black, without and with surface functionalization (oxidized)**
- **To assess the translocation of carbon black nanoparticles beyond the target organs lung (and GI tract/oral gavage).**

Test items

	^{7}Be-tagged Monarch® 1000	^{7}Be-tagged Printex® 90
Name	Carbon black, amorphous	Carbon black, amorphous
Trade name	Monarch® 1000 (Cabot)	Printex® 90 (Orion)
Purity	100%	100%
Specific surface (m^2/g)	340	230
CAS number	1333-86-4	1333-86-4
Surface functionalization	Yes; oxidized	No

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Preparation of ^{7}Be-tagged carbon black

- Direct irradiation of carbon black (Abbas et al., 2013; Bäcker et al., 2019)
- Proton irradiation → ^{7}Be radioisotope is produced directly in the crystal lattice of carbon

  Nuclear reaction: natC(p,x)^{7}Be, mainly via ^{12}C(p,3p3n)^{7}Be channels;
  Proton energy range 24-38 MeV;
  ZAG Zyklotron, Karlsruhe, Germany

- **Spallation process: ^{7}Be → γ tracer; half-time = 53 days**
- Approx. 20 mg of carbon black were filled into an aluminium capsule and the cavity sealed with a screwed cap.

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Purification of ^{7}Be-tagged carbon black

- Purification of the test item - liberation from soluble or loosely attached moieties
- Triplicate of solvents: EtOH/H_2O 1/1 v/v – 0.01 N HCl – Artificial lysosomal fluid - ALF) (*LeFevre & Joel, 1986;* Abbas et al., 2013)
- An aqueous suspension of approx. 0.3 mg of the ^{7}Be-carbon black sample in a syringe was suspended in water and the carbon black separated by pressing through a filter (pore size: 0.4 µm; Swinnex® Filtration System CAT # SX0001300, EMD Millipore).

Intratracheal instillation and oral gavage

Lungs

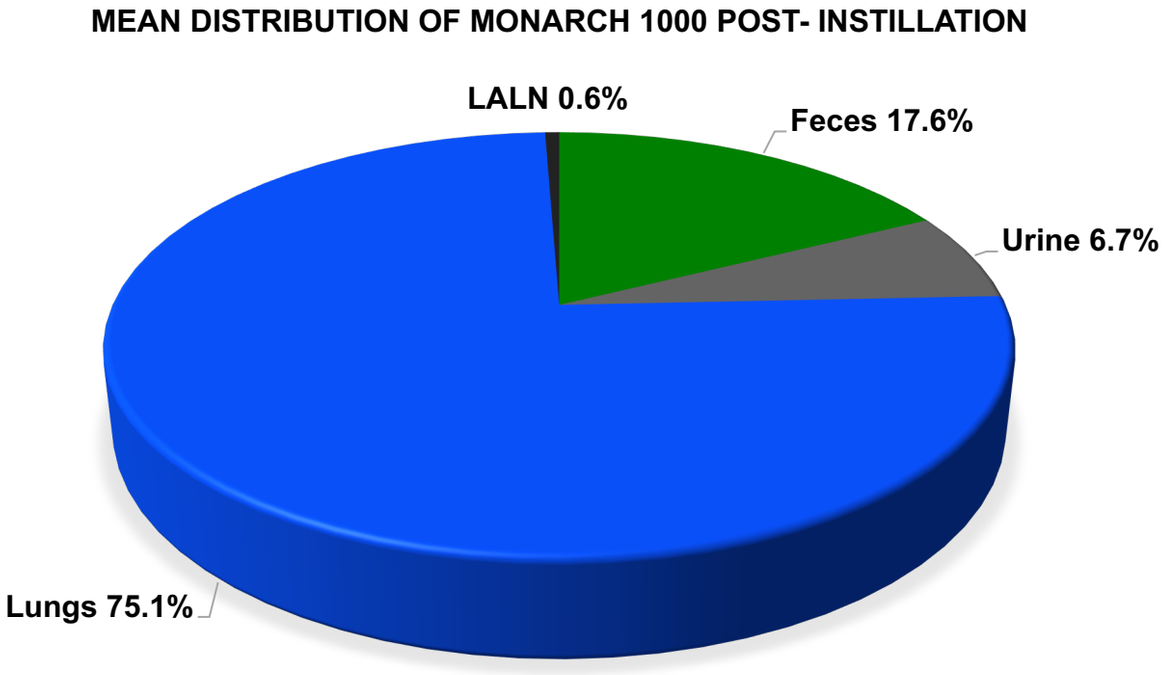
- Single intratracheal instillation of approx. 0.3 mg carbon black suspended in 0.3 ml of sterile isotonic saline
- Dose below lung overload levels does not compromise the physiological ADME capability
- Instillation was preferred to inhalation as only limited amounts of radiolabeled test items were available

Stomach

- 0.3 mg carbon black in 1.5 ml tap water

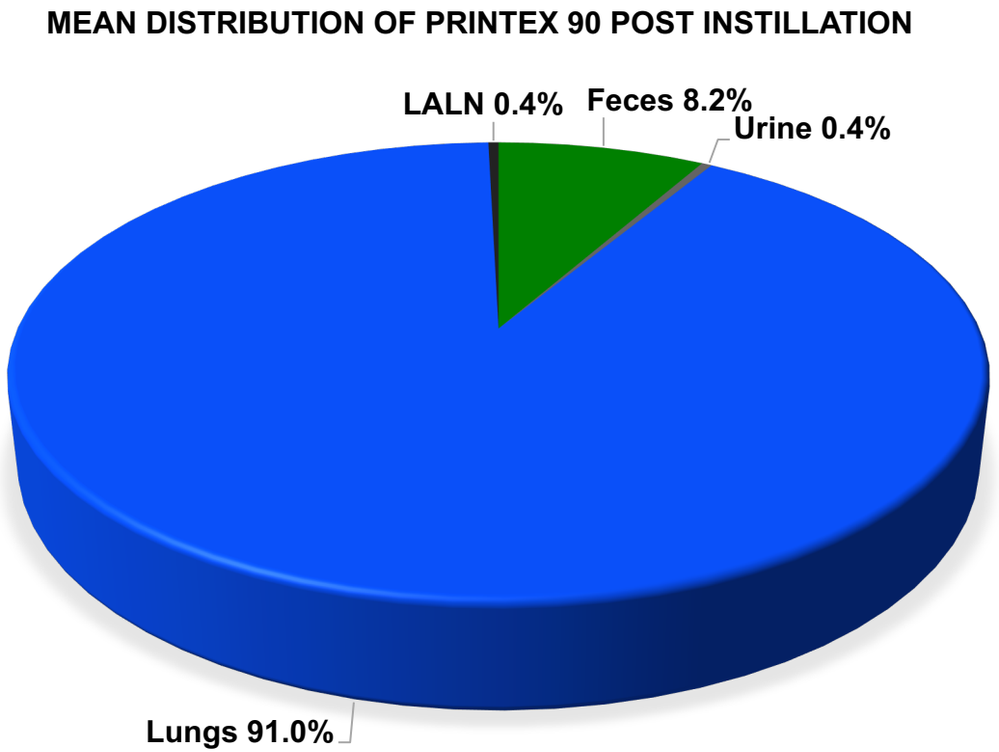
Toxicokinetic Study on Carbon black

Monarch® 1000 – following IT instillation



Toxicokinetic Study on Carbon black

Printex® 90 – following IT instillation



Conclusion

Fate of carbon black agglomerates after deposition

- Carbon black test items act as insoluble microscaled agglomerates (not as individual nanoparticles)

- No evidence for translocation of the test items beyond the lung or the GI tract into the blood or other body compartments (very small amounts only in LALN)

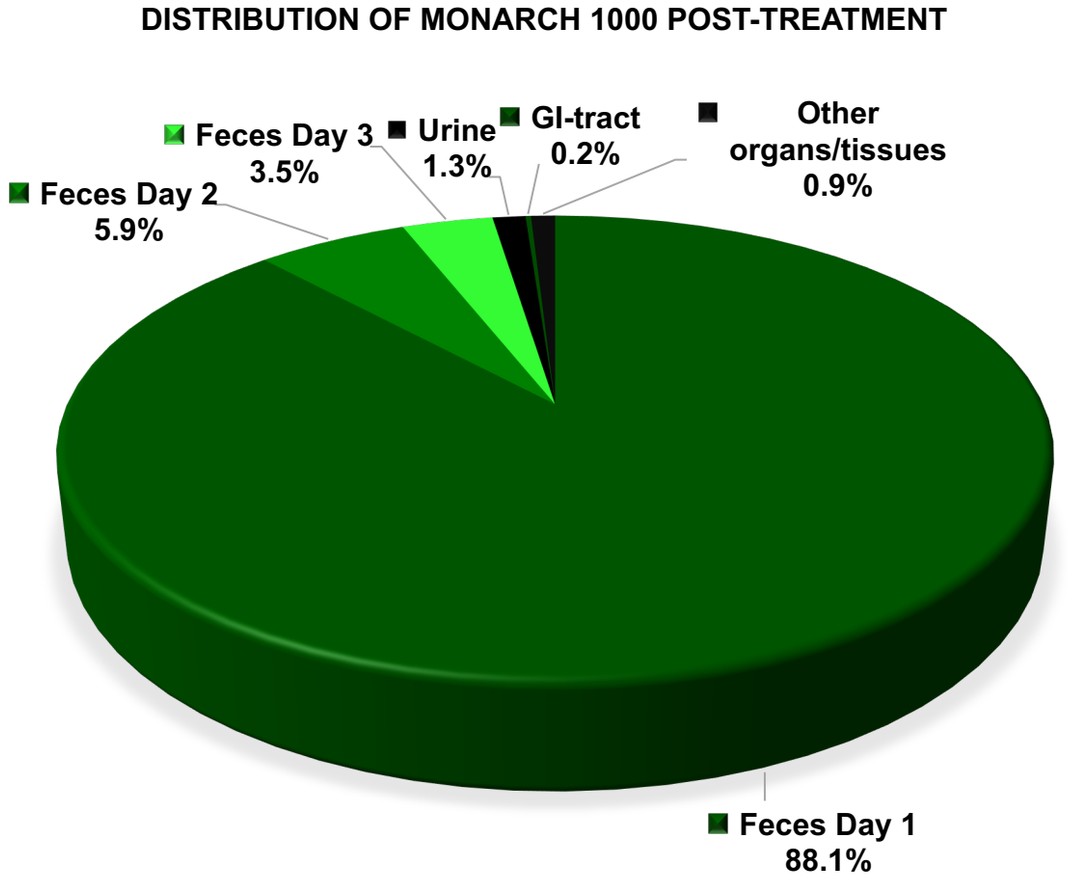
Conclusion

Fate of carbon black agglomerates after deposition

- Following an exhaustive bronchoalveolar lavage (BAL) and centrifugation separate γ activity analysis:
  - Total of radioactivity detected in leukocyte sediment
  - Cell-free supernatant showed no radioactivity

Toxicokinetic Study on Carbon black

Monarch® 1000 – following oral gavage



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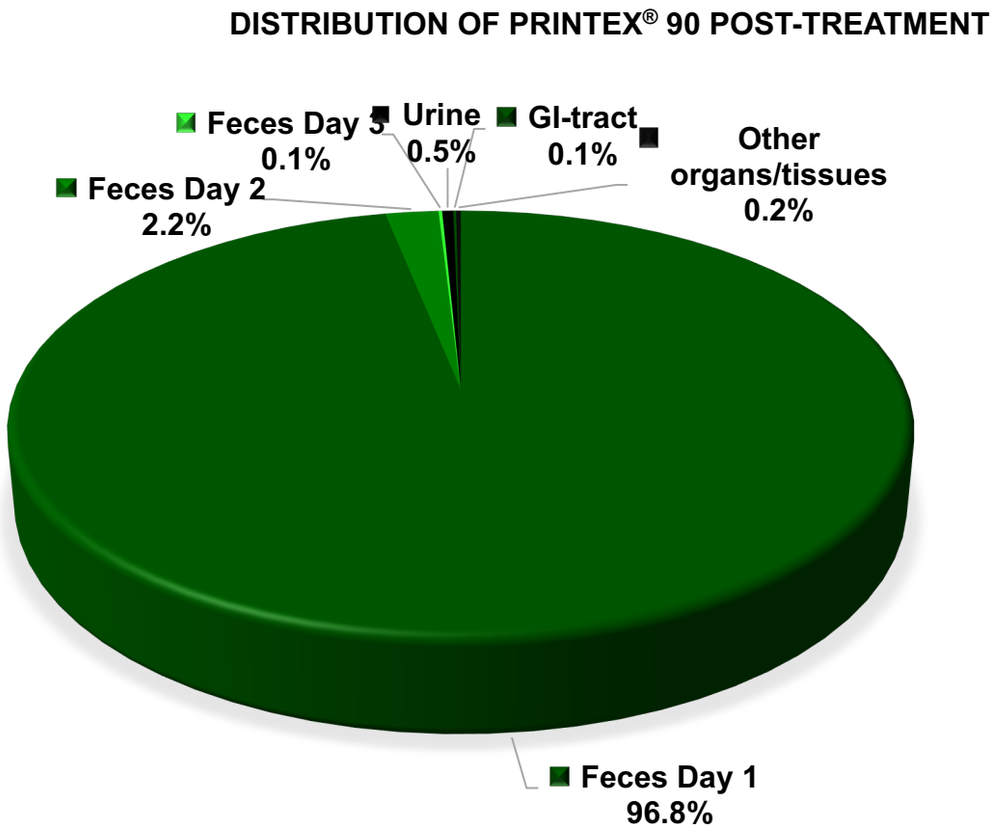
Toxicokinetic Study on Carbon black

Printex® 90 – following oral gavage



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Summary

In occupational inhalation exposure scenarios:

- **Nanoparticles form agglomerates**
  → Deposition and translocation behaviour as observed for microscaled particles

- **Particles do not translocate to remote organs**
  → Dissolved materials are the active agent in remote organs

Acknowledgement
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Thank you for your attention

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