Cerium oxide nanoparticles air exposure: a comparison study using a human 3D airway model and A549 and Beas-2B cell lines

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INTRODUCTION

Human 3D airway models are fully differentiated and functional models of the respiratory epithelium (including metabolic activity, mucus production and cilia beating) and therefore may be positioned in safety evaluation of nanoparticles entering the airways. They are cultured at an air-liquid interface (ALI), allowing relevant exposure via air.
It is anticipated that these models may predict a more realistic bioavailability of inhaled compounds.

OBJECTIVE

To investigate the respiratory effects of nanoparticles, we performed air exposures of nano-sized and micro-sized cerium oxide (respectively nano-CeO_2 and micro- CeO_2) using MucilAir human 3D bronchial model and compared these to nano-CeO_2 and micro- CeO_2 exposed Beas-2B and A549 cell lines.

METHODS

Figure 1 shows the experimental set-up.

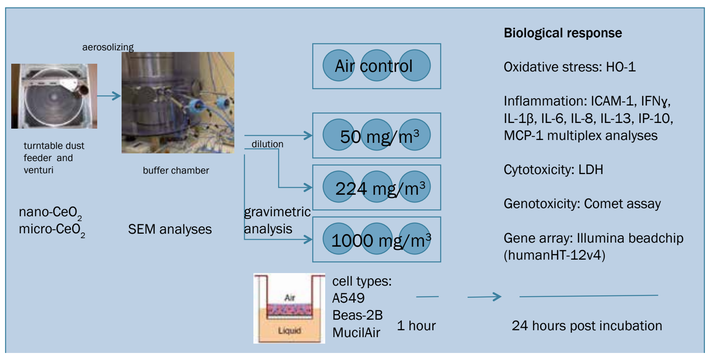

Figure 1: Experimental set-up

RESULTS

Comparison of cell models showed that MucilAir cells are less affected by the air stream compared to the A549 or BEAS-2B cells (based on IL-8 and LDH). MucilAir cells showed a slight HO-1 response for both nano-CeO_2 and micro-CeO_2 exposure, where cell lines did not (Figure 2).

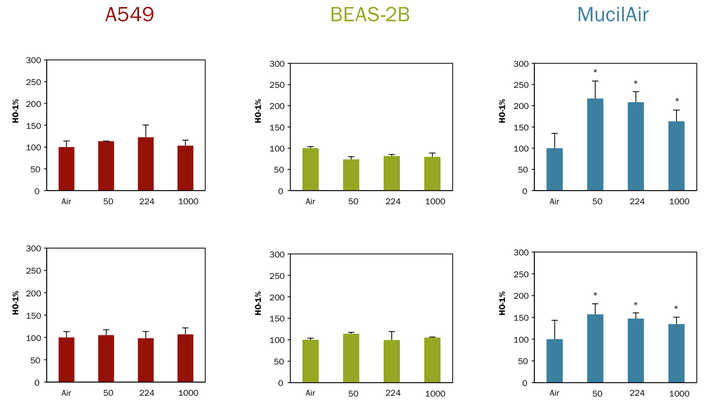

Figure 2: HO-1 response, upper panel = nano-CeO_2; lower panel = micro-CeO_2

In contrast Beas-2B cell line showed an inflammatory (IL-8) and cytotoxicity (LDH) response upon nano-CeO_2 exposure only where MucilAir does not. Both A549 and Beas-2B cell lines showed a dose response in the Comet assay upon nano-CeO_2 exposure only, in contrast to MucilAir (Figure 3).

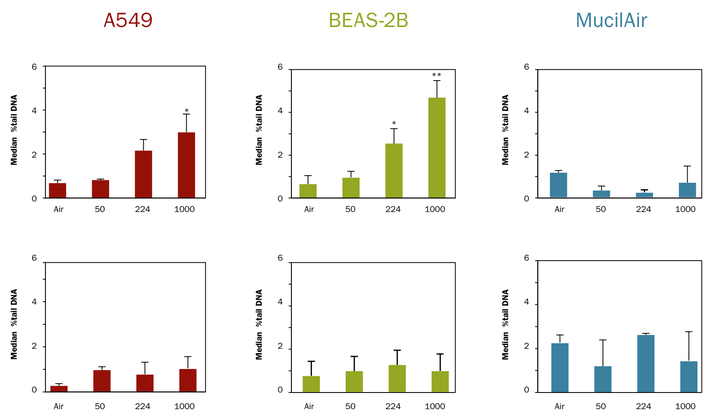

Figure 3: Comet assay response, upper panel = nano-CeO_2; lower panel = micro-CeO_2

Differences between MucilAir and the cell lines are confirmed by the gene expression analyses (PCA analyses, Figure 4). For Beas-2B a higher and significant gene regulation is observed in nano- then in micro-CeO_2 exposed cells. In addition, a dose related decrease is found in the number of genes and induced pathways for nano-CeO_2, whereas a dose related increase is found for the micro-CeO_2 exposed cells. For MucilAir a dose related decrease was observed upon exposure to micro-CeO_2.

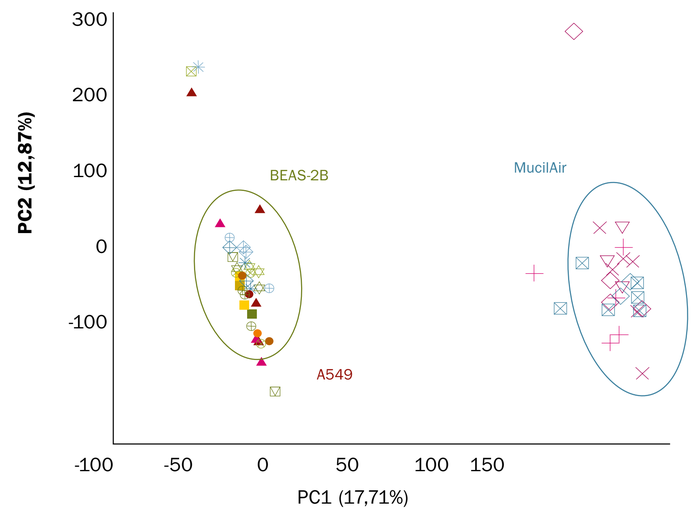

Figure 4: Gene expression PCA

SUMMARY

Table 1. Summary of the results

	Parameter	A549	Beas-2B	MucilAir
Nano	Oxidative stress: HO1	-	-	All doses
	Inflammation: IL8	-	High dose	-
	Cytotoxicity: LDH	-	High dose	-
	Genotoxicity: Comet	Dose response	Dose response	-
	Gene expression		Dose related decrease	Not determined
Micro	Oxidative stress: HO1	-	-	All doses
	Inflammation: IL8	-	-	-
	Cytotoxicity: LDH	-	-	-
	Genotoxicity: Comet	-	-	-
	Gene expression	-	Dose related increase	Dose related decrease

CONCLUSION

We conclude that MucilAir human 3D airway model is more resistant to air stream and nano-CeO_2 compared to the cell lines, most likely due to its in vivo relevant and protective morphology (cilia, mucus layer etc.). Results suggest that MucilAir reacts in Tier1, where the cell line Beas-2B represents a Tier2/3 situation upon exposure (Figure 5).
Overall the use of human 3D airway models might predict a more realistic response where cell lines might overestimate the effect of nanoparticles.

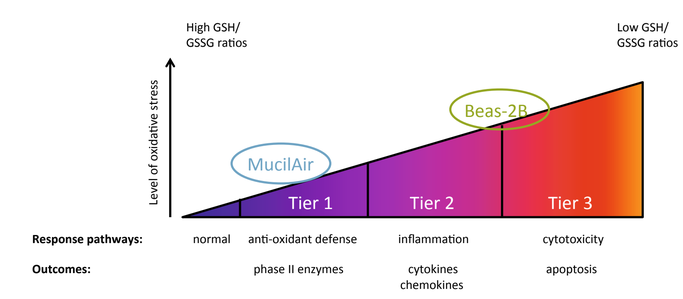

Figure 5: Hierarchical oxidative stress model (after Li et al.2003)

Reference

N. Li et al. Clinical Immunology 109 (2003) 250–265

This work was performed in close collaboration with TNO Triskelon.

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