Investigating the molecular processes behind the cell-specific toxicity response to titanium dioxide nanobelts

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Research Article

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Abstract

Background

Whereas several engineered nanomaterials are able to incite toxicological effects, the underlying molecular processes are understudied. And the varied physicochemical properties complicate toxicological predictions. Gene expression data allow us to study the cell-specific responses of individual genes, whereas their role in biological processes is harder to interpret. An overrepresentation analysis allows us to identify enriched biological processes and link the experimental data to these, but still prompt broad results which complicates the analysis of detailed toxicological processes. We demonstrated a targeted filtering approach to compare the cell-specific effects of two concentrations of the widely used nanomaterial titanium dioxide (TiO2) -nanobelts.

Methods

We compared public gene expression data generated by Tilton et al. from colon endothelium cells (Caco2), lung endothelium cells (SAE), and monocytic like cells (THP1) after 24-hour exposure to low (10 µg/ml) and high (100 µg/ml) concentrations of TiO2 -nanobelts. We used pathway enrichment analysis of the WikiPathways collection to identify cell and concentration-specific affected pathways. Gene sets from selected Gene Ontology terms (apoptosis, inflammation, DNA damage response and oxidative stress) highlighted pathways with a clear toxicity focus. Finally, pathway-gene networks were created to show the genetic overlap between the altered toxicity-related pathways.

Results

All cell lines showed more differentially expressed genes after exposure to higher concentration, but our analysis found clear differences in affected molecular processes between the cell lines. Approximately half of the affected pathways are categorized with one of the selected toxicity-related processes. Caco2 cells show resilience to low and high concentrations. SAE cells display some cytotoxic response to the high concentration, while THP1 cells are already strongly affected at a low concentration. The networks show for up- and downregulation for the THP1 cells the most pathways. Additionally, the networks show gene overlap between almost all pathways for all conditions.

Conclusions

The approach allowed us to focus the analysis on affected cytotoxic processes and highlight cell-specific effects. The results showed that Caco2 cells are more resilient to TiO2 -nanobelts exposure compared to SAE cells, while THP1 cells were affected the most. The automated workflow can be easily adapted using other Gene Ontology terms focusing on other biological processes.

Full Text

Due to technical limitations, full-text HTML conversion of this manuscript could not be completed. However, the latest manuscript can be downloaded and accessed as a PDF.

Additional Files

Additional Files 9 and 10 are not available with this version:

Additional file 9 — Visualization of log fold change on the Toll-like Receptor Signaling pathway (wikipathways:WP75) for all six conditions.

Cell lines are depicted from left to right as Caco2, SAE and THP1. The top row depicts the low concentration whereas the bottom row depicts the high concentration. Gradient goes from blue (log fold change < -0.58) via white (log fold change = 0.0) to red (log fold change > 0.58).

Additional file 10 — List of Gene Ontology (GO) evidence annotation codes.

List of Gene Ontology (GO) evidence annotation codes which were used to remove genes related to the four GO-terms with these annotations (bottom part). Additionally, the annotation codes that were present are shown (top part).

Figures

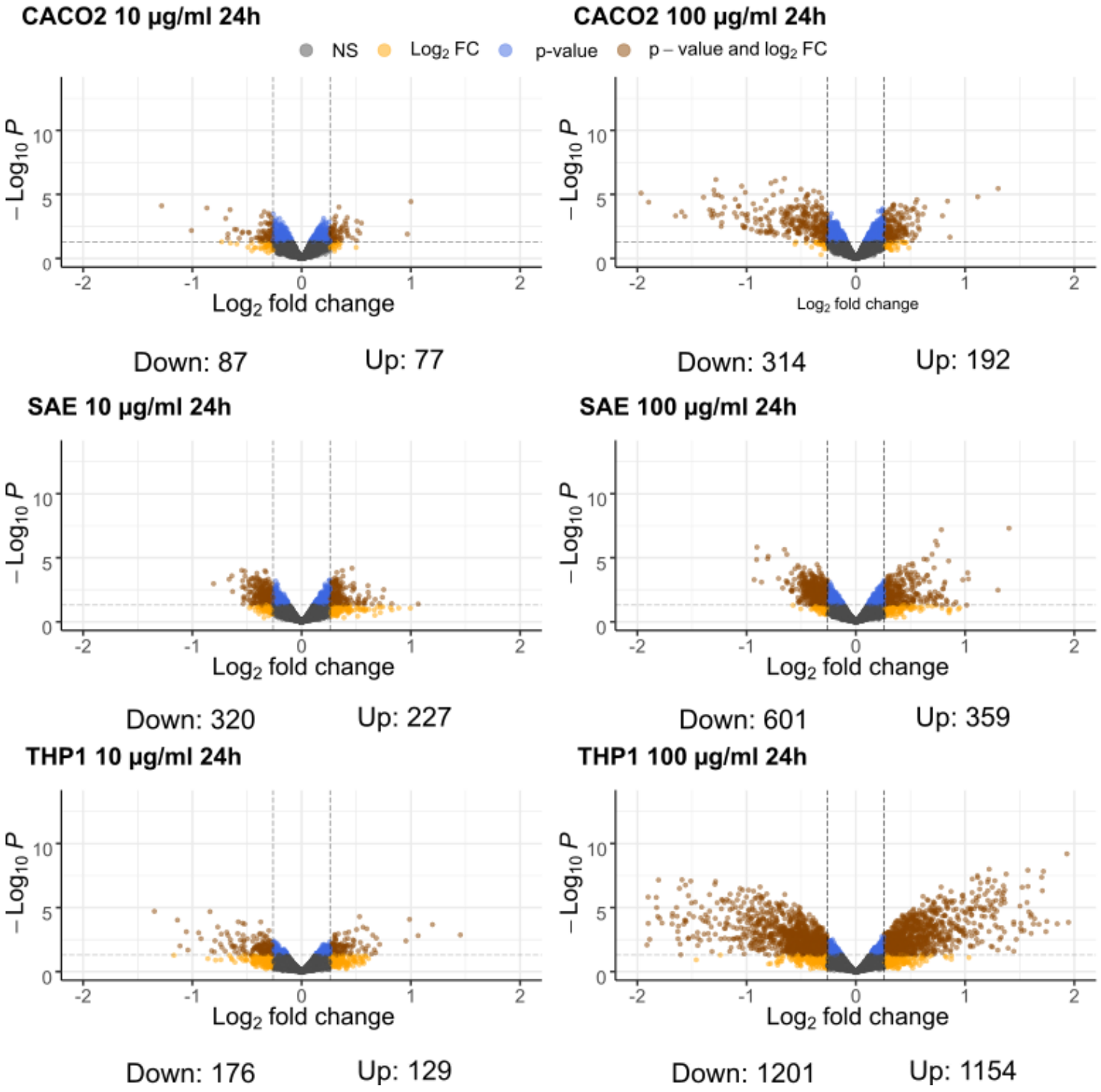

Figure 1

Gene expression volcano plots for different cell lines and TiO2-nanobelts concentrations. On the x-axis log2(fold change) is depicted whereas on the y-axis the -log10(p-value) is depicted. The dotted lines represent cut-off values for significantly changed genes (absolute log2 fold change > 0.26, p-value < 0.05). Brown color depicts that a gene meets both cut-off criteria, a blue color relates to meeting only the p-value cut-off, an orange color relates to meeting only the log2 fold change cut-off and grey color indicates that a gene does not meet any of the criteria.

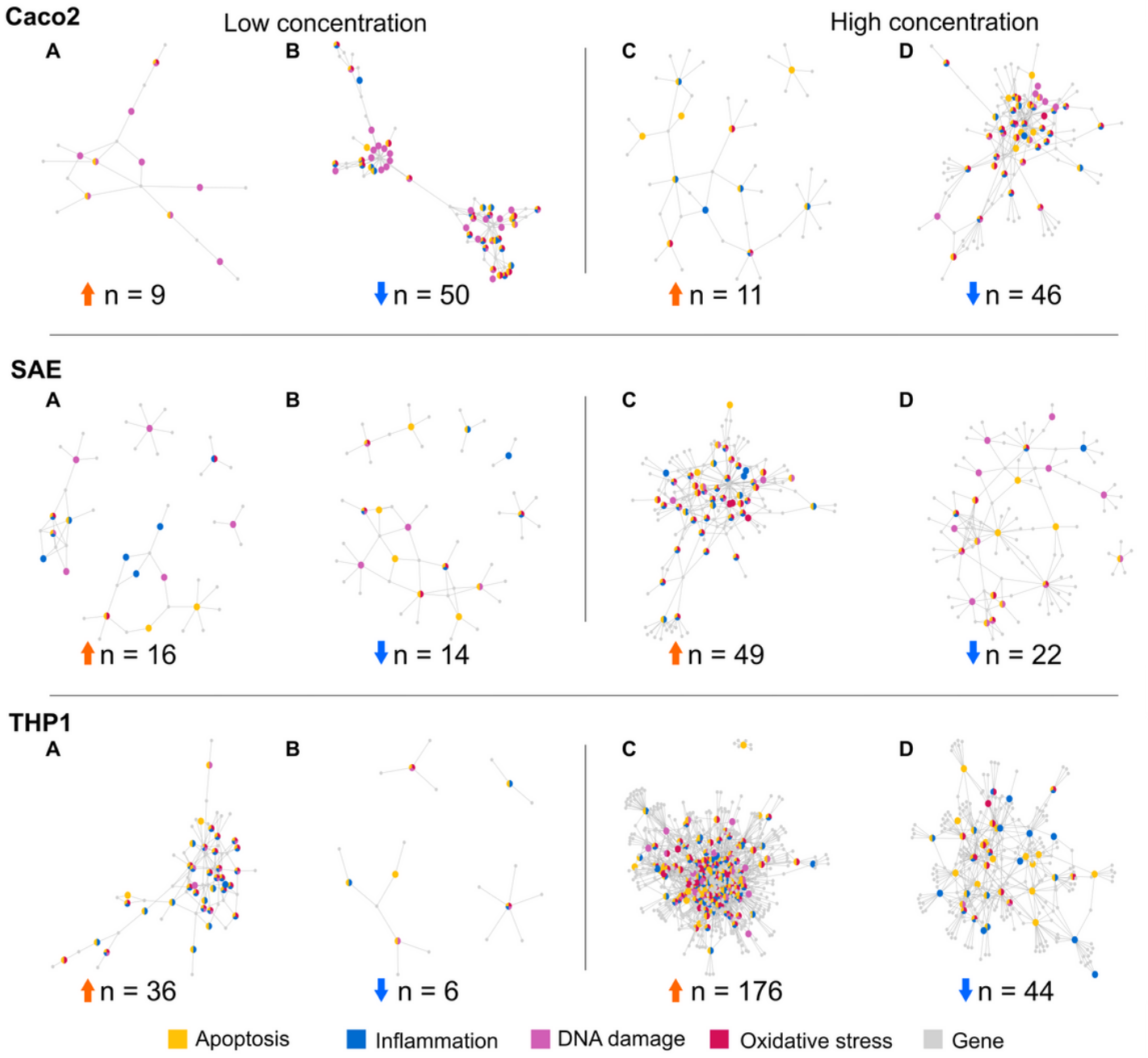

Figure 2

Pathway-gene networks of altered toxicity-related pathways. Color of the nodes depict to which GO-term the pathway is affiliated. Orange depicts apoptosis, blue depicts inflammation, pink depicts DNA damage and bordeaux depicts oxidative stress. Gray nodes represent genes. Number (n) represents the number of significantly overrepresented pathways that are depicted in the networks. A and B depict up- and downregulated of the low concentration of TiO2-nanobelts, respectively, while C and D depict up- and downregulated of the high concentration of TiO2-nanobelts, respectively.

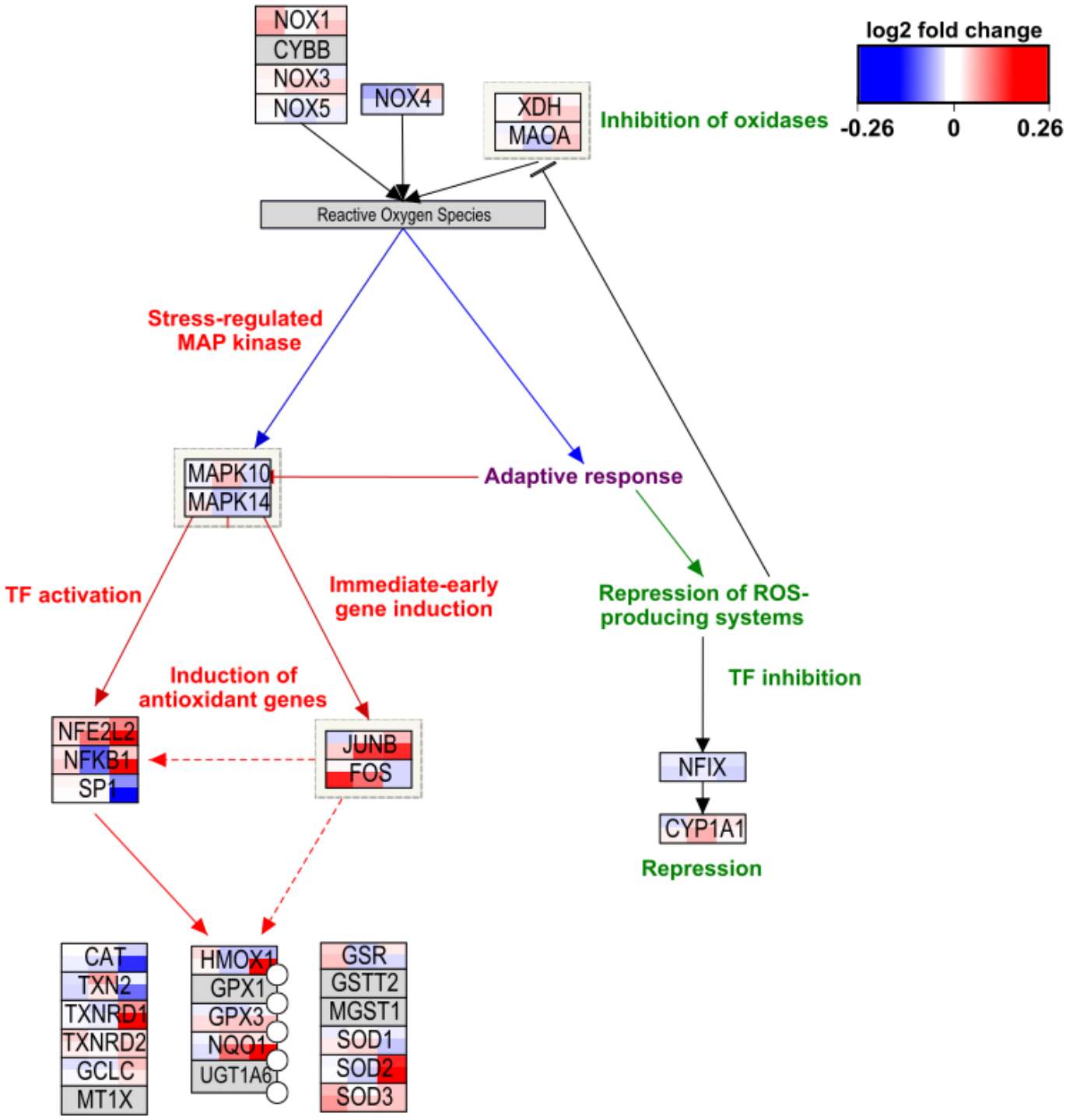

Figure 3

Visualization of log fold change on the Oxidative Stress pathway (wikipathways:WP408) for all six conditions. Cell lines are depicted from left to right as Caco2, SAE and THP1. The top row depicts the low concentration whereas the bottom row depicts the high concentration. Gradient goes from blue (log fold change < -0.58) via white (log fold change = 0.0) to red (log fold change > 0.58).

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- figS1.png
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