# Quantitative characterization of transcriptional responses to quantum dots of different physico-chemical parameters

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***Short Abstract* — Recent experimental investigations have studied the relevance of factors such as surface charge, functionalization, and size to the bioimpact of various species of quantum dots on human cells. Here we analyze transcript expression data for a set of cytotoxicity-relevant genes in human lung cells exposed to CdSe quantum dots of different sizes and functionalizations. We use a network inference method based on mutual information to determine groups of genes co-regulated across various exposures. Additionally, we use principal component analysis to quantitatively characterize the transcriptional response evoked by each exposure. We find distinctly clustered responses according to the surface charge of the dot, but not according to core size or specific type of ligand.**

## I. Introduction

QUANTUM dots (QDs) are a promising class of engineered nanomaterials that have enabled technological advances in a number of areas of biological significance, including disease diagnostics, therapeutics, and imaging. However, this broad potential stands to outpace corresponding knowledge about the effect of these materials on human cells and tissues [1]. While previous studies have generally focused on core composition as a predictor of quantum dot toxicity [2-3], more recent studies have suggested a role for other factors such as surface charge, functionalization, and core size in determining toxicity [4-5].

## II. Methods

Normal human bronchial epithelial (NHBE) cells were exposed to QDs for 6 hours prior to RNA purification. The following parameters were compared using CdSe QDs: core size (3, 5, 10 nm), functionalization (MPA, MUA, AUT, cysteamine), and concentration (0.5, 20, 80 μg/mL). Differences in gene expression were assessed using the BioMark real-time PCR high throughput chip system and 96.96 dynamic arrays (Fluidigm, CA). The 96 TaqMan assays selected for this study are representative of key cellular pathways associated with apoptosis, mitochondrial function, oxidative stress, cell cycle arrest, inflammation and extracellular matrix formation. A full description of experimental methods can be found in Ref. 5.

Of the 96 measured genes, a net number of 68 genes returned usable values from the PCR procedure for all 50 exposure conditions. Log-transformed fold-change data was mean-normalized to zero for each gene prior to analysis. The context likelihood of relatedness (CLR) algorithm was used to find co-regulated genes [6]. Principal component analysis was then used to decompose the expression matrix [7].

## III. Results

The CLR algorithm identified several significant associations from the data set that were justified on a priori grounds (e.g. BRCA1/2, CYP1A1/B1), validating the methodology. Other connections suggested co-regulation that is relevant to nanotoxicity, such as a cluster centered around IL6 and CXCL5, and containing TNF and MMP9.

In projecting the gene expression changes onto the two largest principal components, we note clear separation between the clusters produced by dots with positive as compared to negative surface charge. Dots of different sizes were indistinguishable, as were the negatively-charged ligands MPA and MUA, while there was some separation between the positively-charged AUT and cysteamine. Further work will be necessary to determine if these ligands produce genuinely distinct transcriptional responses.

## IV. Conclusion

We have used quantitative analytics to examine gene expression changes in human lung cells exposed to quantum dots of different physico-chemical parameters. We find that the expression changes are strongly clustered according to surface charge, but not by specific functionalization or size. Our results support the conclusions of related work which suggests that surface charge is a predictor of QD toxicity [5].

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## References

[1] Hardman R (2006) A toxicologic review of quantum dots: toxicity depends on physicochemical and environmental factors. *Env Health Perspectives* **114**, 165-172.

[2] Li H, et al. (2011) Cytotoxicity tests of water soluble ZnS and CdS quantum dots. *J Nanosci and Nanotech* **11**, 3543-3551.

[3] Wang L, et al. (2008) Toxicity of CdSe nanoparticles in Caco-2 cell cultures. *J Nanobiotech* **6**, 11.

[4] Hoshino A, et al. (2004) Physicochemical properties and cellular toxicity of nanocrystal quantum dots depend on their surface modification. *Nano Letters* **4**, 2163-2169.

[5] Nagy A, et al. (submitted) Comprehensive analysis of the effects of CdSe quantum dot size, surface charge and functionalization on primary human lung cells. *ACS Nano*.

[6] Faith JJ, et al. (2007) Large-scale mapping and validation of *Escherichia Coli* transcriptional regulation from a compendium of expression profiles. *Plos Biology* **5**, 54-66.

[7] Abdi H, Williams LJ (2010) Principal component analysis. *Wiley Interdisciplinary Reviews-Computational Statistics* **2**, 433-459.