Biomaterials 35 (2014) 9941–9950

Contents lists available at ScienceDirect

Biomaterials

journal homepage: www.elsevier.com/locate/biomaterials

High-content imaging and gene expression analysis to study cell–nanomaterial interactions: The effect of surface hydrophobicity

Bella B. Manshian [a], Daniel F. Moyano [b], Nikky Corthout [c, d], Sebastian Munck [c, d], Uwe Himmelreich [a], Vincent M. Rotello [b, **], Stefaan J. Soenen [a, e, *]

[a] MoSAIC/Biomedical NMR Unit, Department of Medicine, Catholic University of Leuven, B3000 Leuven, Belgium
[b] Department of Chemistry, University of Massachusetts, 710 North Pleasant Street, Amherst, MA 01003, United States
[c] Vlaams Instituut voor Biotechnologie (VIB), Center for the Biology of Disease, Leuven, Belgium
[d] Center for Human Genetics and Leuven Institute for Neuroscience and Disease (LIND), University of Leuven (KU Leuven), Leuven, Belgium
[e] Faculty of Pharmaceutical Sciences, Ghent University, Ghent, Belgium

A R T I C L E I N F O

Article history:
Received 5 August 2014
Accepted 20 August 2014
Available online 15 September 2014

Keywords:
High-content imaging
Nanotoxicology
Nanoparticles
Cytotoxicity

A B S T R A C T

The effects of nanoparticle (NP)-related parameters on cellular interactions are currently uncertain as analysis is complicated by the combinatorial diversity arising from the array of size, shape and surface properties. Here, we present a validated multiparametric high-content imaging method, with the utility of this approach demonstrated by in-depth analysis of the role of hydrophobicity on the interaction of Au NPs with cultured cells. In this methodology, we evaluate cell viability, membrane damage, induction of reactive oxygen species, mitochondrial health, cell area, skewness and induction of autophagy. High-content cell cycle phase studies and in-depth gene expression studies then serve to elucidate the underlying mechanisms. The data reveal a clear influence of the degree of NP surface hydrophobicity with membrane damage and autophagy induction, which is stronger than the effect of surface charge, for charges ranging between −50 and +20 mV. All labeling experiments occur in the same format, and can be further supplemented with additional parameters providing a broadly accessible format for studying cell-NP interactions under highly reproducible conditions.

© 2014 Elsevier Ltd. All rights reserved.

1. Introduction

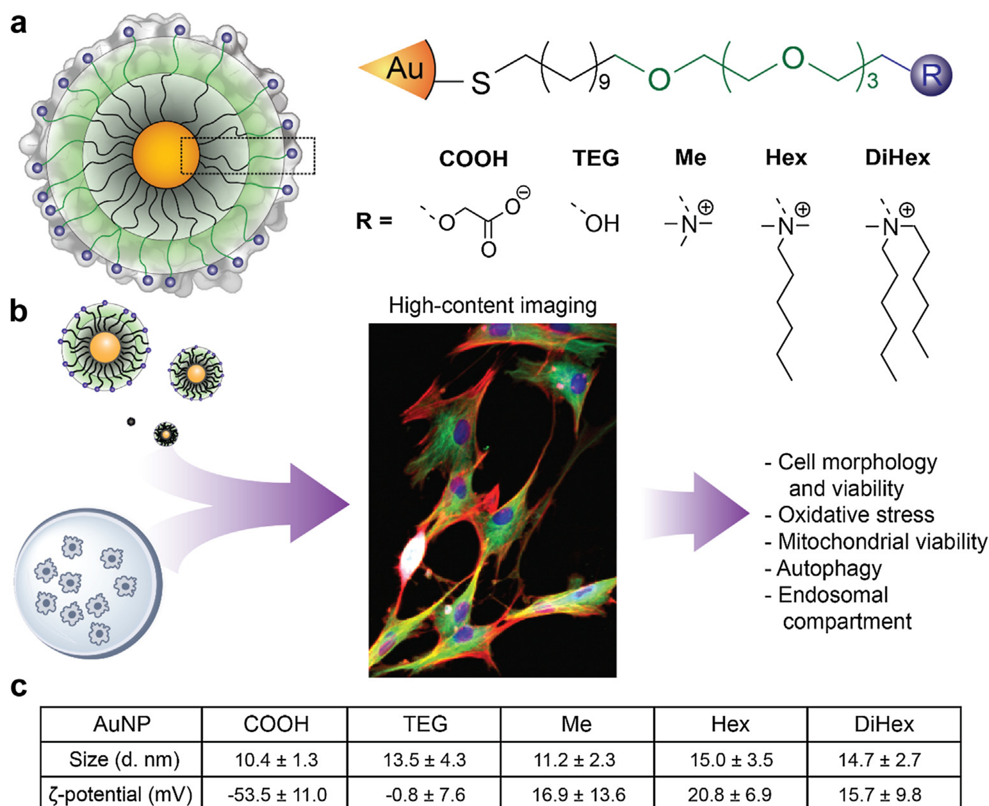
The interaction of engineered nanoparticles (NPs) with biosystems is a multi-scale challenge, with key unanswered questions from the molecular to the organismic level [1–4]. These issues have slowed the translation of NPs into clinical applications due to the potential for unwanted and unanticipated side effects. To address these uncertainties, considerable effort has been devoted to determining the precise mechanisms by which NPs interact with biosystems. This research has generated new methods to cope with the intrinsic difficulties associated with nanotoxicological research [5,6] while providing detailed characterization of cell-NP interactions [7]. Broader understanding of and hence predictive capabilities for NP-biosystem interactions has been hampered by the absence of parametric understanding of NP behavior, as well as the lack of well-defined and reproducible conditions in the literature and the need for robust and easy NP detection systems [7–9].

We have recently developed a microscopy-based multiparametric methodology [8] for studying cell-NP interactions that provides quantitative description of the effects of NPs on cellular parameters, and hence provide a better understanding of the mechanisms related to the toxic effects of the NPs. The utility of this methodology has been demonstrated in different studies, with non-toxic levels of the NPs defined and linked to their final functionality, e.g. magnetic resonance imaging contrast (iron oxide particles) or fluorescence brightness [10–12]. This methodology is slow, however, and the data generated using microscopy-based techniques remains statistically limited, as the number of cells that are typically analyzed remain low (hundreds).

High-content (HC) imaging provides a promising tool for determining NP-cell interactions, generating large datasets that include thousands of cells per condition. Coupled with precise and automated image analysis, multiple parameters can be extracted for every condition, providing vital information for bioinformatics-based modeling

* Corresponding author. Department of Medicine, Catholic University of Leuven, Leuven, Belgium
** Corresponding author. Department of Chemistry, University of Massachusetts, MA 01003, United States
E-mail addresses: rotello@umass.chem.edu (V.M. Rotello), Stefaan.Soenen@med.kuleuven.be, Stefaan.Soenen@UGent.be (S.J. Soenen).

http://dx.doi.org/10.1016/j.biomaterials.2014.08.031
0142-9612/© 2014 Elsevier Ltd. All rights reserved.



AuNP	COOH	TEG	Me	Hex	DiHex
Size (d. nm)	10.4 ± 1.3	13.5 ± 4.3	11.2 ± 2.3	15.0 ± 3.5	14.7 ± 2.7
ζ-potential (mV)	-53.5 ± 11.0	-0.8 ± 7.6	16.9 ± 13.6	20.8 ± 6.9	15.7 ± 9.8

Fig. 1. a) Chemical structure of the gold nanoparticles, featuring the ethylene glycol spacer for colloidal stability and biocompatibility, and the ligand headgroup to control charge and hydrophobicity. b) Schematic representation of the high-content imaging technique employed to study the outcomes of nanoparticle–cell interactions. c) Size and zeta potential of the gold nanoparticles (synthesis described in the ESI).

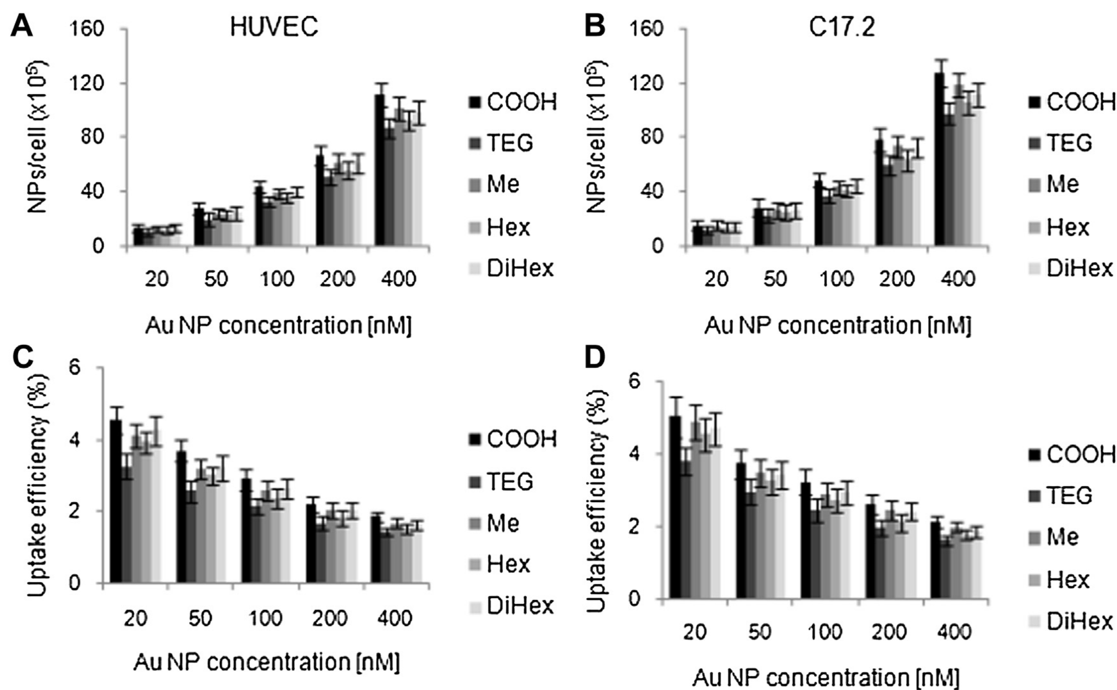


Fig. 2. Cellular uptake of Au NPs determined by ICP-MS for A) HUVEC and B) C17.2 cells exposed to various concentrations of the different Au NPs used in this study expressed as the mean number of nanoparticles per cell. C,D) Cellular uptake efficiency for C) HUVEC and D) C17.2 cells exposed to various concentrations of the different Au NPs determined by taking the ratio of the number of cell-associated Au NPs and the total number of Au NPs initially present in the incubation medium given to the cells. Data are represented as mean ± SEM ($n = 3$).

Download English Version:

https://daneshyari.com/en/article/5957

Download Persian Version:

https://daneshyari.com/article/5957

Daneshyari.com