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Five reasons to use bacteria when assessing manufactured nanomaterial environmental hazards and fates

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Manufactured nanomaterials (MNMs) are increasingly incorporated into everyday products and thus are entering the environment via manufacturing, product use, and waste disposal. Still, understanding MNM environmental hazards and fates lags MNM industry growth. To catch up, keep pace, and influence future MNM safe design strategies, rapid safety assessments are needed. Bacteria are important ecological nanotoxicology targets to consider when assessing MNM safety: bacteria are exposed to MNMs in water, sewage, soils, and sediments, wherein they influence MNM fates; bacteria can also be impacted — with potential health and ecosystem consequences. Routinely using bacteria for assessing MNMs would promote effective management of the environmental risks of this rapidly growing industry, but appropriate protocols and policies for this assessment need to be instituted.

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Introduction

Manufactured nanomaterials (MNMs), defined by their size as between 1 and 100 nm [1], are being incorporated into products ranging from health and personal care, agriculture, coatings, composites, electronics, and energy storage devices [2–4]. Production amounts are uncertain, but are reported as ranging from less than 1 to greater than 10 000 tons per year worldwide for the most common MNMs (e.g. TiO_2, ZnO, SiO_2, oxides of iron or cerium or aluminum, carbon nanotubes, fullerenes, silver, and quantum dots) [1]. While environmental exposures are predicted to be low [5], significant environmental concentrations for some MNMs have already been reported [6,7••]. To assess MNM environmental hazards, the starting point has been to adapt established ecotoxicity tests to the idiosyncrasies of MNMs, but those tests mostly use plants or animals [8]. However, there is mounting evidence that environmental bacteria are susceptible to MNM toxicity with impacts at subcellular, population, community, and ecosystem scales [9•,10••]. Further, bacteria can affect MNM transport and fate in the environment by binding to [11•,12•] or breaking down MNM's [7••,13].

Not only are bacteria important receptors to study in ecological nanotoxicology [9•], but they are also facile test subjects that can be used in miniaturized toxicological screening for rapid hazard identification [14,15]. The results can inform safer nanomaterial designs [16] for guiding the fast-paced MNM industry. Progressively larger and more complex experimental formats are also available to directly test MNM effects at bacterial community and ecosystem levels [9•].

This paper outlines evidence suggesting why bacteria should be foci when assessing MNM environmental fates and ecological toxicity. Five reasons are described: MNMs can reduce bacterial community diversity; MNMs can alter bacterial physiology and thus nutrient cycling; bacteria affect MNM physical characteristics and partitioning; bacteria may degrade MNMs; bacteria can initiate MNM trophic transfer to higher organisms. While some of these outcomes may have important applications, for example for new antibiotics in human health [17] or for labeling bacteria in environmental research [18], our emphasis focuses on using bacteria for assessing ecological hazards of MNMs. Ways, and needs, to move forward are suggested.

MNMs can reduce bacterial community diversity

Soil, sediment, and aquatic bacterial communities are diverse, with typically thousands of taxa present in a single gram of a given environmental sample. While understanding is incomplete, reduced bacterial community diversity in the natural environment impinges on ecosystem and human health.

MNMs, including nanoscale TiO_2 and ZnO [19••], and multiwalled [20] and single-walled [21] carbon nanotubes (MWCNTs, and SWCNTs, respectively) have been shown to reduce soil bacterial diversity. In some cases, for example for nanoscale TiO_2 [19••], soil bacterial diversity decreases because intact MNMs are bioavailable and inhibitory *in situ* [22••]. In other cases, for example for nanoscale CuO or ZnO MNMs that dissolve in the soil solution [23], or for nanoscale Ag dissolving in seawater [24], bacterial diversity is reduced as a result of well-recognized metal ion toxicity to cells and populations [25], instead of from intact nanoparticles. Regardless, as either intact particles or as toxic-ion delivery vehicles, MNMs can decrease bacterial diversity in the environment. Broadly, decreased bacterial diversity may lead to lost ecosystem functions, for example even for denitrification, which was thought to be robust because of functionally redundant bacterial taxa within complex environmental communities [26]. Therefore, as MNMs increasingly enter and accumulate in the environment, decreased bacterial diversity could impair ecosystem functions. Understanding if and how MNMs decrease bacterial diversity could serve the ecological nanotoxicological goal of predicting potential ecosystem-scale MNM impacts.

MNMs can alter bacterial physiology and thus nutrient cycling

Even when MNMs do not alter overall bacterial community diversity (i.e. taxa richness, evenness, or both) [27], they can still impact key bacterial populations and their functions. For example, many types of MNMs, including CeO_2, Ag [28], and graphene oxide [29], affect nutrient removal by bacteria in wastewater treatment. Nitrogen cycling processes can either be inhibited, for example by Ag MNMs [30], or may appear stimulated due to a bacterial stress response, for example to CdSe quantum dot nanoparticles [31].

The potential for MNMs to alter agricultural nutrient cycles is also of concern. For example, soybeans grown in CeO_2-amended soils had root nodules devoid of N_2-fixing bacteroid cells, and had limited nitrogen fixation potential as a result [32••]. Further, silver (Ag) nanoparticles entering soils via wastewater-treatment biosolids reduced microbial biomass and lowered exoenzyme (leucine amino peptidase and phosphatase) activities [33]. How MNM properties contribute to such outcomes will depend on environmental conditions, for example with redox-active ceria nanoparticles acting as either pro-oxidants or anti-oxidants depending on UV light intensities and MNM concentrations [34]. Further, MNM effects on nutrient cycling reactions can also be indirect, for example tungsten (W) nanoparticles inhibit free-living diazotroph population growth by binding to an organic metalophore that normally acquires Mo, a necessary metal in nitrogenase [35]. Regardless of the mechanisms, MNMs interfering with waste treatment and agriculture is a clear concern. Such potential impacts suggest that routinely testing MNM toxicity in select, functionally significant populations (e.g. highly specialized N_2 fixing bacteria) could usefully indicate potential ecosystem-level MNM impacts.

Bacteria affect MNM physical characteristics and partitioning

Besides MNMs impacting bacterial diversity and function, bacteria can alter MNM physicochemical states, and thereby affect MNM environmental partitioning, compartmentalization, and bioavailability. First, bacteria have large surface areas relative to their volumes, and cells strongly sorb MNMs [36••]. This enables bacteria to disperse large MNM agglomerates [11•] and to retain MNMs in wastewater treatment activated sludge [7••]. Since biosolids from activated sludge are used as an agricultural fertilizer, soils could be exposed to MNMs [33]. A corollary is that MNMs are less concentrated in treated aqueous wastewater effluent because MNMs preferentially associate with settleable biomass [7••], and thus receiving waters and sediments may be *less* exposed.

In addition, bacteria release surfactant-like macromolecules that sorb to and change MNM surface hydrophobicity or hydrophilicity [37]. In soils, this can affect bacterial bioavailability of nanoscale TiO_2 [22••]. Depending on their chemical characteristics, biomolecules can inhibit or enhance quantum dot dissolution [38]. However, such processes might be self-limiting, since bacteria can also nucleate ions from dissolved quantum dots wherein new nanoparticles precipitate [39].

Some MNMs can permeate bacterial membranes and accumulate in cells [10••]. When bacteria with internalized MNMs flow with water, they would carry their MNMs to new locations. Alternatively, bacterial biofilms on sand surfaces sorb MNMs and thereby retard MNM transport through pores [12•]. Thus, bacteria may either enhance or diminish MNM transport in the environment.

Taken together, bacterial associations with MNMs often result in sorption, uptake, or new MNM coatings that alter MNM phases, transport, and partitioning. As bacteria are prevalent in all environmental compartments, their influences on MNM transport are universally important to MNM bioavailability.

Bacteria may degrade MNMs

In addition to affecting MNM transport, bacteria may directly degrade, that is break down, MNMs. Bacteria biodegrade many conventional pollutants, and underpin industries in the bioremediation of chlorinated solvents, pesticides and petroleum hydrocarbons. Biodegradation is generally key to organic pollutant fate, since it can detoxify materials and thus protect humans and ecological receptors from exposure to contaminated water or soil. If bacteria degrade MNMs, this could decrease MNM