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# CONSIDERATIONS FOR USING DISSOLUTION AS A FUNCTION OF SURFACE CHEMISTRY TO EVALUATE ENVIRONMENTAL BEHAVIOUR OF NANOMATERIALS IN RISK ASSESSMENTS

## A Preliminary Case Study Using Silver Nanoparticles

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# CONSIDERATIONS FOR USING DISSOLUTION AS A FUNCTION OF SURFACE CHEMISTRY TO EVALUATE ENVIRONMENTAL BEHAVIOUR OF NANOMATERIALS IN RISK ASSESSMENTS

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# FOREWORD

The OECD Joint Meeting of the Chemicals Committee and Working Party on Chemicals, Pesticides and Biotechnology (the Joint Meeting) held a Special Session on the Potential Implications of Manufactured Nanomaterials for Human Health and Environmental Safety (June 2005). This was the first opportunity for OECD member countries, together with observers and invited experts, to begin to identify human health and environmental safety related aspects of manufactured nanomaterials. The scope of this session was intended to address the chemicals sector.

As a follow-up, the Joint Meeting decided to hold a Workshop on the Safety of Manufactured Nanomaterials in December 2005, in Washington, D.C. The main objective was to determine the "state of the art" for the safety assessment of manufactured nanomaterials with a particular focus on identifying future needs for risk assessment within a regulatory context.

Based on the conclusions and recommendations of the Workshop [ENV/JM/MONO(2006)19] it was recognised as essential to ensure the efficient assessment of manufactured nanomaterials so as to avoid adverse effects from the use of these materials in the short, medium and longer term. With this in mind, the OECD Council established the OECD Working Party on Manufactured Nanomaterials (WPMN) as a subsidiary body of the OECD Chemicals Committee in September 2006. This programme concentrates on human health and environmental safety implications of manufactured nanomaterials (limited mainly to the chemicals sector), and aims to ensure that the approach to hazard, exposure and risk assessment is of a high, science-based, and internationally harmonised standard. This programme promotes international co-operation on the human health and environmental safety of manufactured nanomaterials, and involves the safety testing and risk assessment of manufactured nanomaterials.

This document is published under the responsibility of the Joint Meeting of the Chemicals Committee and Working Party on Chemicals, Pesticides and Biotechnology of the OECD.

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## 1. INTRODUCTION

1. Manufactured nanomaterials (henceforth referred to as nanomaterials) enter the environment at various life-cycle stages (e.g., from manufacturing, end-use products, recycling, etc.). To assess their safety, governments are supporting the OECD Recommendation of the Council on the Safety Testing and Assessment of Manufactured Nanomaterials [C(2013)107] and are continuing to apply existing chemical-based regulatory frameworks. These frameworks, which currently rely on endpoints developed for conventional chemicals may not be equipped to deal with the behaviour of nanomaterials. The dissolution behaviour of manufactured nanomaterials, in addition to other parameters/properties such as size (Peretyazhko et al. 2014) and surface reactivity, can affect their short and long-term fate and toxicity (including bioavailability and uptake pathways). An important challenge is therefore to distinguish if the effects are a result of the novel properties displayed by the nanomaterial, degradation of it, the release of ions or most likely a combination of all. Currently, there is no guidance on how and/or when to use dissolution information in risk assessments for nanomaterials. For example, risk assessors focusing on chemical composition and structure (the typical approach used for conventional chemicals) will assume a worst-case of the particle being transformed completely into component/secondary ions; while risk assessors focusing on the nano-properties will assume negligible dissolution (in the absence of appropriate information) and complete preservation of any unique properties (such as effects resulting from quantum confinement). These two approaches are not always appropriate to assess the safety of nanomaterials in the environment and broader nano-relevant guidance is needed to properly evaluate the fate and effects of nanomaterials in the environment.

2. Dissolution of inorganic nanomaterials in the environment generally encompasses both kinetic and thermodynamic processes including surface degradation. During dissolution, component ions and reaction products are released from the nanomaterial, and both these processes are considered for the purposes of this project due to the lack of delineation between these two processes in literature. In order to consider dissolution in environmental risk assessments of nanomaterials, this project was initiated with an aim to identify an approach to help guide current regulatory environmental risk assessments when

evaluating dissolution information using the current state-of-the-science; and extend, as far as possible using our current understanding from silver nanoparticles (the case-study) to other metal-based nanoparticles. Consistent with one of the primary objectives of the OECD Programme on Manufactured Nanomaterials i.e., to develop approaches to support ongoing risk assessments of manufactured nanomaterials, outcomes of this project will need to be continually refined and updated as relevant science and standardized test methods become available. Silver nanoparticles (AgNPs) were selected as a case study in this project since numerous studies have been published dealing with the dissolution of AgNPs with different surface modifications under different environmental conditions. The fact that AgNPs undergo an oxidative dissolution, although important in terms of understanding mechanisms, is not relevant for this project since we are interested in developing an approach on how to use dissolution within the context of an environmental risk assessment. The findings in this document are expected to apply to nanomaterials as powders and in liquid suspensions and not to releases from solid matrices. In addition, the recommendations developed herein must be considered along with all the considerations and uncertainties outlined throughout this document so that they can be used in an appropriate context.

3. Surface modification is just one parameter affecting dissolution of nanoparticles (among other factors such as temperature, solvent interactions, etc). Surface modifications are often intentionally applied by the manufacturer to stabilize nanoparticles in media and include those which interact: (i) electrostatically (charge repulsion such as citrate modifications), (ii) sterically (by spatial repulsion such as polyvinylpyrrolidone modifications), and (iii) electrosterically (a combination of electrostatic and steric modifications) with the particle and media (El Badawy et al., 2012; Misra et al., 2012). Other factors that influence dissolution of AgNPs include changes in pH, temperature, presence/absence of organic matter, oxidation potential, etc. (Kent et al, 2012). It should also be noted that literature pertaining to dissolution of AgNPs in biological media (i.e., human health relevant test data) was not considered as it is outside the scope of this project.

4. In addition to the types of surface modifications highlighted above, silver undergoes sulfidation and other surface reactions depending on environmental conditions. For clarity, it was agreed that to support the objective of this project which is broader than only AgNPs, surface reactions such as sulfidation would be considered on a case-by-case basis in risk assessments (see section on sulfidation for discussion). In addition, it is also important to highlight that for the purposes of this document AgNPs represent a class of substances that can include many combinations of silver nanomaterials and associated modifications. Additional considerations are also highlighted in the recent report by the Danish Environmental Protection Agency (Hartmann et al., 2014) which discusses key factors associated with dissolution of various nanoparticles, including silver.

5. For clarity, the data collected from literature and the OECD dossiers is organized by media type: Natural media (i.e., real environmental samples (e.g. river waters), synthetic media (including OECD media and laboratory water modified with organic matter), artificial soil media, and ultrapure water.

6. Summaries of relevant research studies are presented in Section 2 and, using information derived from these, recommendations to consider during the regulatory risk assessments of metal-based nanoparticles are discussed in Section 3.

## 2. DISCUSSION

7. Dissolution from AgNPs to silver ions is an oxidative process involving protons ($H^+$) and dissolved oxygen. Under controlled experimental conditions the dissolution is governed by Equation 1 (Liu and Hurt 2010):

$$2Ag_{(s)} + \frac{1}{2}O_{2_{(aq)}} + 2H^+_{(aq)} \Leftrightarrow 2Ag^+_{(aq)} + H_2O_{(l)} \quad (1)$$

8. There are no standardized test methods, including OECD Test Guidelines, specific for evaluating dissolution of nanomaterials. The lack of a standardized test method made it impossible to collect relevant studies which could be considered predictive of environmental behaviour. As an interim, we considered existing OECD Test Guidelines for chemicals used to evaluate solubility and dissolution only to select appropriate data from literature. While these methods may not be suitable to evaluate nano-specific behaviours (e.g., aggregation kinetics), we considered the test duration to be applicable to nanomaterials. Specifically, we consulted OECD test guidelines for chemical testing No. 105 and OECD guidance document No. 29 and the durations used ranged from 4 days (meant for the more soluble organic chemicals) to 28 days. It is understood that identifying a duration is outside the scope of this project. For the purposes of selecting data, the duration will be refined when the information is updated. Since this project focuses on AgNPs and metal-based nanoparticles, a 7 day timeframe (for short-term study) from OECD Guidance Document 29 on transformation/dissolution of metals and metal compounds in aqueous media (OECD, 2001) was considered suitable.

9. The purpose of the approach described here is to classify a given dissolution value as high, moderate, low, or negligible i.e., ions of the metallic nanoparticle released after 7 days of aquatic exposure. The term dissolution is used qualitatively in this report instead of solubility since the behaviour of nanoparticles under equilibrium conditions is still not well understood and calculating rate constants is not always feasible Descriptors such as high, moderate, low, and negligible, although qualitative, are used regularly in conventional risk assessments to organize large sets of data for the purposes of read-across and/or focusing concerns. For example, in the Canadian New Substances Program for industrial chemicals, the following conventional chemical-based solubility descriptors are used to predict environmental fate and effects: Negligible (<0.01 mg/L); Low (0.01-10 mg/L); moderate (10-1,000 mg/L); high (1,000-10,000 mg/L); very high (>10,000 mg/L). These descriptors are similar to those used by other jurisdictions, such as the US EPA Office of Pollution Prevention and Toxics which regulates new industrial chemicals. These descriptors, which are one of several used by the US EPA program, are: Negligible (<0.001 mg/L); Low (0.001-1 mg/L); Moderate (1-1000 mg/L); High >1000 mg/L). It is important, however to note that dissolution (kinetic metric) should not be used as a solubility descriptor (thermodynamic equilibrium).

10. The Expert Group for this project selected literature studies on silver nanoparticle dissolution based on the established duration of the test, as well as a set of additional criteria established by the Group. According to these criteria, to be included for review as part of this project, a study should adequately measure and report the following identity data and physical-chemical endpoints:

- Substance name, composition, overall purity, identity and amount of impurities, size information, details on surface modifications, specific surface area, aggregation/agglomeration information, pH range and variation for the duration of the study, test media, ionic strength of test media, temperature and variation, detailed method description, number of trials/replicates, analytical method, changes to the substance during test, dissolution products.

11. The literature search resulted in identification of 39 original publications of which 10 were included in the preliminary findings section (i.e. the ones that had at least 80% of the parameters listed)). While these 10 papers were found to meet the minimum quality criteria for this project, there was still a clear lack of standardization in the way the tests were conducted and endpoints reported (e.g., still many uncertainties in the success of separating ions and particles, assessing the mass balance, and calibration and validation). It should be noted that none of the publications contained all of the criteria listed above.

### Preliminary Findings on Dissolution by Media Type:

#### *Natural media:*

12. The reviewed literature indicated that dissolution of AgNPs was negligible to low in freshwater and seawater samples (freshwater and saltwater samples were both obtained from local sources in California, USA; Thio et al., 2012). Results ranged from <1% mass dissolution (of the initial concentration of nanoparticles) after 30 days for polyvinylpyrrolidone (PVP) and citrate-coated AgNPs in freshwater to 8% and 6% after 30 days for PVP and citrate-coated AgNPs in seawater, respectively with starting concentrations of 10 mg/L (Thio et al., 2012). Li and Lenhart (2012) observed a faster release of silver ions from sterically stabilized nanoparticles in suspensions in comparison to particles with other types of modifications (such as electrostatic or electrosteric) (Li and Lenhart, 2012). They concluded that sterically stabilized nanoparticles are likely to inhibit aggregation when entering water, which leads to a faster ion release rate compared to other types of surface modifications (Li and Lenhart, 2012). The initial concentration of silver nanoparticles was 1.46 mg/L, as reported by Li and Lenhart (2012). Furthermore, Angel et al. 2013 reported the 72h dissolution of a 40mg/L solution of silver nanoparticles in natural seawater was approximately 3% for both PVP coated and citrate coated AgNPs (compared to micron sized silver particles which had a dissolution of 0.3%).

13. The use of artificial surfactants (i.e. Tween) in river water to disperse and sterically stabilize silver nanoparticles resulted in 3% silver ion release (relative to the total silver present) after 6 hours when compared to uncoated and citrate coated particles which reached 3% silver ion release only after 15 days (Li and Lenhart, 2012). It should be noted that the TWEEN coated silver nanoparticles remained at 3% dissolution for 15 days (Li and Lenhart, 2012). These findings are consistent with the work done on polyacrylate coated silver (NM-300K) by the Joint Research Centre (Klein et al., 2011).

#### *Synthetic media (water containing environmentally relevant chemicals):*

14. A study conducted by Angel et al. (2013) reports that humic acid reduced agglomeration of nanoparticles and only affected dissolution of electrostatically stabilized AgNPs (citrate coated) when compared to synthetic soft water. The results of the study using a 40mg/L initial concentration of AgNPs, indicate a 72-h dissolution of approximately 1.5% for both PVP and citrate surface modifications. The addition of humic acid to the synthetic media lowered the dissolution in citrate coated AgNPs to 0.7% after 72 hours. It was also shown in this study that AgNPs dissolution increased with chloride concentration (72-h study) (Angel et al., 2013). Speciation modeling indicated that the dissolved silver formed $AgCl_x^{y-}$ complexes with the $Cl^-$ ions, which are ubiquitous in environmental media. Further to this, it is highlighted that nanosilver dissolution is directly related to the dissolved concentration of chloride ions in solution (Angel et al., 2013). In this study, silver dissolution for both citrate and PVP coated nanoparticles was measured as a function of chloride concentration (Angel et al., 2013). After 72h at marine relevant chloride concentrations (564mM $Cl^-$) 58% and 34% of the total silver was detected as dissolved silver in the citrate and PVP coated particles respectively (Angel et al., 2013). In addition, by using OECD media to determine

stability, it was concluded by Tejamaya et al. (2012) that charge stabilized (citrate) particles were less stable than sterically stabilized (PVP) particles (21 day study) (Tejamaya et al., 2012).

***Artificial soil media:***

15. Cornelis et al. studied AgNPs dissolution in artificial soil pore water (Cornelis et al., 2012). It was found that AgNPs with a 0.1% PVP coating had a dissolution of <10% within a pH range of 4-9 after 24 h (Cornelis et al., 2012). Further to this, addition of fulvic acid or citrate to the soil system reduced AgNPs dissolution (Cornelis et al., 2012).

***Pure water:***

16. Studies performed in ultra-pure water by Kittler et al. show that PVP-coated AgNPs display proportionally higher dissolution rates relative to citrate-coated particles with increasing temperature (Kittler et al., 2010). Citrate-coated particle dissolution increased from 14% to 70% when heated from room temperature (25ºC) to 37°C, whereas PVP-coated particles increased from 9% at 5°C to 50% at 25°C and 90% at 37°C (Kittler et al., 2010) over periods ranging from 33 days to 125 days.

17. Furthermore, electrostatic stabilizers (e.g. citrate) seemed to be affected by changes in ionic strength and presence of electrolytes, while steric (PVP) and electrosteric (branched polyethylenimine, BPEI) remain largely unchanged by these conditions (El Badawy et al., 2012). In a study by Huynh and Chen, dissolution was found to be affected by ionic strength (e.g. NaCl, $CaCl_2$, $MgCl_2$) (Huynh and Chen, 2011). It was determined that PVP-coated AgNPs underwent less dissolution under these conditions than citrate coated AgNPs (Huynh and Chen, 2011). In addition, AgNPs dissolution can be minimized under anaerobic conditions (Xiu et al., 2012). While the above data is informative, it is important to note that these dissolution percentages are significantly higher than the studies in natural media (as high as 50% in the study above compared to 8% in natural media reported in the above section); however they do provide mechanistic information on dissolution.

***Other Factors Affecting Dissolution:***

***Sulfidation:***

18. Sulfidation, i.e. reaction with sulphur or a sulphur containing molecule to form $Ag_2S$ on the surface of the AgNPs, has a dramatic effect on the dissolution of AgNPs. Sulfidation can be introduced either synthetically or naturally. A buffered solution of PVP-AgNPs showed dissolution of 18 mg/L over 30 days (initial concentration of AgNPs was 100mg/L), while sulfidised PVP-AgNPs showed dissolution in the range of up to 2.5 mg/L (Levard et al., 2011). It is expected that sulfidation leads to a core-shell AgNPs structure (Dale et al. 2013). Dissolution of sulfidised AgNPs is affected by solution chemistry (e.g. concentration of sulfur ions) (Dale et al. 2013; Kaegi et al. 2011). Sulfidation also affects agglomeration kinetics, surface charge, and adsorbed mass of modification (Levard et al., 2011). It is clear that sulfidation is an environmentally relevant reaction mechanism for silver nanoparticles. However, to keep within the confines of this project it was decided that the reaction mechanism between sulfur and silver not be used to formulate the recommendations. This is not to lessen the role sulfidation plays but because the recommendations in this report need to apply to a larger set of nanomaterials and the levels of sulfidation vary drastically between metallic nanoparticles.

***Other key findings:***

19. The importance of nanomaterial dissolution has been discussed by Misra et al. (2012) and kinetically and thermodynamically evaluated by Liu and Hurt (2010). In the review by Misra et al. (2012), shape (e.g. whether the particle is a sphere, rod, cube etc. and the associated aspect ratio of that shape if

applicable), surface morphology, and radius of curvature were described as other important factors that affect dissolution (Misra et al., 2012). As well, certain crystal structures of AgNPs have different dissolution kinetics (Misra et al., 2012). Finally, media characteristics (e.g. pH, ionic strength, water hardness, presence of organic matter) play a large role in suspension stability and can lead to agglomeration/aggregation/dissolution (Misra et al., 2012). The interaction of nanoparticles with media components can also further affect their bioavailability during the process of dissolution (Misra et al. 2012).

***Organic matter:***

20. According to Liu and Hurt (2010) and Angel et al. (2013), humic and fulvic acids inhibit silver nanoparticle dissolution in a dose-dependent manner in synthetic freshwater. Liu and Hurt suggest mechanisms for this inhibition are: (a) through surface adsorption of natural organic matter (NOM) which in turn will block silver nanoparticle oxidation sites, (b) through ligand exchange with existing stabilizing agents (it has been suggested that in terms of AgNPs-stabilizing agent binding strength: PVP > Citrate > PEGylated > humic acid), (c) released silver ions reducing back to $Ag_{(s)}$ through reaction with humic/fulvic acids or (d) due to the oxidation of NOM by $H_2O_2$ produced during silver dissolution (Liu and Hurt, 2010).

## 3. RECOMMENDATIONS

21. Surface modifications are a major (but not the only) contributing factor in the dissolution behaviour and in turn the fate and effects of nanomaterials (Misra et al. 2012). As a result, the type of surface modification and how it is bound to the core particle needs to be considered in nanomaterial risk assessment.

22. In order to promote consistency with existing chemical frameworks and to ensure future linkages between nanomaterials and the corresponding larger particles, it is recommended that the duration and test conditions of any dissolution take into account the OECD Guidance Document No. 29 on *Transformation/ Dissolution of Metals and Metal Compounds in Aqueous Media* (OECD, 2001) while taking nano-specific characteristics and behaviour (e.g. agglomeration/aggregation kinetics) into consideration (until a nano-specific test guideline is available).

23. These considerations developed on the basis of this dissolution project link with two new projects being led by the US and Germany; A new test guideline on dissolution is currently being developed by the US and a guidance document on the fate decision tree on dispersion and dissolution behaviour is being led by Germany. It is expected that the new test guideline on dissolution will express results as a rate instead of a percentage loss relative to the parent nanoparticle used in this project. Once this test guideline has been validated for its use in risk assessment, this project will be updated to account for rates in terms of data selection and expression of the recommendation. The duration of 7 days discussed in this project will, if needed, be amended in a future revision of this document in order to be consistent with the new test guideline on dissolution. The duration selected should account for the dissolution of surface modified nanoparticles which may not reach equilibrium in the short-term and will likely release ions over time.

24. Finally, the analysis of the data suggests that artificial, synthetic, and pure water media may not accurately predict the complexities of real environmental media and how biotic and abiotic interactions influence bioavailability and toxicity. Ideally, a standardized artificial system which extrapolates well to environmental behaviours needs to be identified so there is consistency across the tests. At the moment however, the only surrogate that can be used to determine environmental behaviour of silver nanoparticles is by studying AgNPs in real environmental media (e.g. river/marine waters, soil leachates) and use that to extrapolate to real environmental scenarios. The scientific literature shows no clear linkage between artificial systems and environmental behaviour at this time. This is consistent with the assessment of bulk and ionic metal-based particle dissolution which apply a correction (when appropriate) to account for extrapolations from pure water to the environment (e.g., accounting for water chemistry, such as free metal speciation) (Janssen et al., 2000; Janssen et al., 2003).

25. According to the data presented above, a coated silver nanoparticle would be expected to have <10% dissolution in real environmental media, while the use of synthetic media could result in artificially high dissolution of silver nanoparticle with the same modification under the same duration. However, since the durations and starting concentrations in the studies vary quite a lot in our dataset, this conclusion needs to be carefully evaluated for AgNPs and additional studies may be needed (the data would likely be of much more value if rate constants were reported). In addition, the approach described below does not account for dissolution dynamics. It is understood that while risk assessors are more interested in understanding how nanoparticles will behave in the short to long term (i.e., under equilibrium like conditions), the test guideline being developed on dissolution will need to carefully evaluate thermodynamic factors.

26. The following approach (and its descriptors) was developed using the AgNPs case-study and information collected on other metal-based nanoparticles and may be only applied, when appropriate, to powder and liquid-suspension forms of metal based nanoparticles (see Section 4 below for discussion) being released into the environment and not to evaluate releases from solid products. In addition, the approach has two considerations. The approach described below provides guidance based on the scientific datasets available now, and it is evident that this will be modified as we have better data based on appropriate guidelines.

> **Approach:** Dissolution is considered negligible if <1% dissolution, relative to total silver content, occurs (i.e., primarily focus in the assessment may be on particles), low if between 1-10% (focus in the assessment may be on particles taking into account small amounts of dissolved species), moderate if between 10-70% (focus in the assessment may be on both particles and dissolved species and high if >70% dissolution occurs (focus may be on dissolution species). Risk assessors can use these descriptors to focus their assessments. In the case of soil; the descriptors can be used to identify which species to focus on in soil pore water after accounting for binding kinetics between soil particles and charged species of the nanoparticle. The following two considerations support this approach:

- ***Consideration 1:*** In the short-term, real-media (e.g. aquatic river samples, soil leachates, etc.) that are well characterised (since dissolution will depend on the media used) should be considered in dissolution studies so the data can be extrapolated to real environmental behaviours. Consideration may be given to other real media which is predictive of behaviour in different environmental processes, such as using wastewater media/conditions to simulate wastewater transformations/removal. Criteria are still needed to define well characterized media so that extrapolations can be made across substances and tests[1]. In the medium-term there is a need to conduct more accurate and precise studies in artificial media so these may act as better predictors of environmental behaviour. This is consistent with the approaches currently used for bulk metal-based particles.

- ***Consideration 2:*** This approach may not be unique to silver nanomaterials and may be used for other metal-based nanoparticles (as appropriate and as scientific information becomes available; see examples in Table 1). This extrapolation is largely dependent on the utilization of similar sample preparation protocols. Refinements of the applicability of this approach will be considered as science and methods becomes available.

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[1] As science continues to develop, standardized representative media will be identified, similar to the OECD media currently used for the testing of chemicals. Since this standard media is lacking for nanomaterials, natural media which is well characterized is the only real predictor and is being suggested for use in the short to medium-term in this document.

## 4. COMPARISON TO OTHER NANOMATERIALS:

27. The thresholds found in literature varied in test duration from 6 hours to 30 days, and standardization is suggested to increase confidence in comparability in the data. In order to determine if these thresholds can be used for other metal-based nanoparticles, the percent dissolution of several nanomaterials has been compared below (Table 1).

28. For dissolution descriptors, it should be noted that the rates of dissolution are dependent on the media used and as such the most relevant media should be considered in the test which would allow appropriate extrapolation to the relevant environment.

29. The dissolution data presented in Table 1 reports the data as a fraction (expressed in terms of percent dissolved silver) of the initial concentration of particles and the concentration of that fraction (in mg/L). The data was either initially reported as a percent or converted into a percent using the model of Thio et al. (2012). In order to calculate the percentage of dissolved silver (i.e. free silver ion) the initial ion concentration measurement of the stock solution must be determined. For example, Thio et al. used Inductively Coupled Plasma-Atomic Emission Spectroscopy (ICP-AES) to deduce a particle concentration of 1100 mg/L. From there, the equation $c_1v_1=c_2v_2$ (where c is concentration and v is volume) was used to calculate the volume of the initial stock solution required to make the necessary concentration of solution (10mg/L) for an experiment. The amount of free silver ions in solution was determined by using the 10mg/L solution in the experiment (Thio et al., 2012). In this case, 0.6 mg/L and 0.8 mg/L free silver ion are measured (Thio et al., 2012). Dividing the final concentration values by the initial concentration and multiplying by 100% gives the value of free silver ions in percent (Thio et al., 2012). Since this project focuses on the impact of surface modifications on nanoparticle dissolution using a percent ion lost calculation, it does not always account for other variables affecting dissolution such as the possibility of nanoparticle recombination, and the relationship between dissolution and particle size (Peretyazhko et al. 2014 and Mitrano et al. 2014).

30. It should be noted that by viewing the data qualitatively at first glance, all these particles would be considered under the ‘low’ descriptor (with the exception of one zinc oxide measurement which would be moderate) if treated as conventional chemicals using the chemical-based descriptors mentioned in the introduction. However, using the approach developed in this project, we can see that in most cases the ions and particles need to be considered in the environmental risk assessment. In addition, the data seems to show similar behaviour across the metallic nanoparticles considered below. Thus, using the approaches in this document, risk assessors can focus efforts on more relevant environmental compartments and ensure effects studies account for relevant nanoparticulate and secondary ionic species.

**Table 1: Applying Negligible/Low/Moderate/High Criteria to Example Nanomaterials**

| **Substance**<br>*(size in nm)* | **Endpoint**<br>**%Dissolution**<br>*(Concentration)* | **Media** | **Dissolution**<br>**High** (>70% dissolution)<br>**Moderate**(10-70% dissolution)<br>**Low** (1-10% dissolution)<br>**Negligible** (<1% dissolution) | **Reference** |
|---|---|---|---|---|
| **ZnO**<br>***(104 nm by SEM)*** | 16.9%<br>*(6.77 mg/L)* | Synthetic | Moderate | OECD WPMN Dossier for ZnO NM-110 (WPMN, 2013)* |
| **ZnO**<br>***(107 nm by SEM)*** | 7.8%<br>*(3.10 mg/L)* | Synthetic | Low | OECD WPMN Dossier for ZnO NM-111 (WPMN, 2013)* |
| **ZnO**<br>***(43 nm by SEM)*** | 31%<br>*(12.4 mg/L)* | Synthetic | Moderate | OECD WPMN Dossier for ZnO NM-112 (WPMN, 2013)* |
| **ZnO**<br>***(143 nm by TEM)*** | 16.9%<br>*(6.76 mg/L)* | Synthetic | Moderate | OECD WPMN Dossier for ZnO NM-113 (WPMN, 2013)* |
| **$CeO_2$**<br>***(12 nm by TEM)*** | 0.0025%<br>*(<1 µg/L)* | Synthetic | Negligible | OECD WPMN Dossier for $CeO_2$ NM-211 (WPMN, 2013)* |
| **$CeO_2$**<br>***(28.4 nm by SEM)*** | 0.0025%<br>*(<1 µg/L)* | Synthetic | Negligible | OECD WPMN Dossier for $CeO_2$ NM-212 (WPMN, 2013)* |
| **$CeO_2$**<br>***(67 nm by TEM)*** | 0.325%<br>*(130 µg/L)* | Synthetic | Negligible | OECD WPMN Dossier for $CeO_2$ NM-213 (WPMN, 2013)* |
| **PVP-AgNPs**<br>***(50 nm by SEM)*** | 8%<br>*(0.8 mg/L)* | Seawater | Low | Thio et al. 2012 |
| **Citrate AgNPs**<br>***(40 nm by SEM)*** | 6%<br>*(0.6 mg/L)* | Seawater | Low | Thio et al. 2012 |
| **TWEEN AgNPs**<br>***(88 nm)*** | 3%<br>*(~30 µg/L)* | Freshwater | Low | Li and Lenhart 2012 |
| **Citrate AgNPs**<br>***(82 nm by DLS)*** | 3%<br>*(~30 µg/L)* | Freshwater | Low | Li and Lenhart 2012 |
| **Uncoated AgNPs**<br>***(82 nm by DLS)*** | 3%<br>*(~30 µg/L)* | Freshwater | Low | Li and Lenhart 2012 |

SEM: scanning electron microscope; TEM: transmission electron microscope; DLS: dynamic light scattering.
* Data from the WPMN dossiers is still undergoing validation.

## 5. ADDITIONAL NEEDS

31. Short and medium-term research should be directed to fill knowledge gaps on: (1) standardized test methods on dissolution, sample preparation, and data modeling and standardization in the form of rate constants that account for time and surface area; (2) characterization of real-media; and (3) dissolution in real soil and sediment samples. Prioritization will need to be undertaken to target research efforts in the short-term. Studies are needed to make artificial media a better predictor of real conditions (i.e., to identify a media which allows realistic extrapolation from lab-scale studies to environmental ecosystems). This is consistent with approaches used for non-nano metal-based particles. In addition, experts suggested that filter based studies could be considered as they would preclude the issue of reabsorption and these would also greatly reduce complexities in the test methodology (however, challenges such as ion sorption to filter material, etc. will need to be addressed). In addition, studies comparing the particles with and without surface modifications and the modifications alone (e.g. just citrate) are needed to identify correlations, if any, between the three systems. Consideration should be given to different categories of nanomaterials which will help prioritize research and inform on the applicability of the approach in this document to the different categories of nanomaterials. Finally, much research is still needed to explore mechanisms driving dissolution (both kinetic and thermodynamic), in both pure and environmental media along with the contribution of surface topology such as roughness.

32. The selection of real-media for dissolution testing should be a guided process, possibly a checklist or decision tree should be formulated in order to aid the choice, preparation and characterisation of appropriate test media from the environment. A set of chemical / physical descriptors for test media should be developed. In the longer run, suitable artificial media should be developed.

# REFERENCES

A comprehensive literature review was undertaken for this project. This list contains documents that were consulted during the research process. Only primary references are cited in the text.

***Primary:***

Angel, B. M.; Batley, G. E.; Jarolimek, C. V.; Rogers, N. J. The Impact of Size on the Fate and Toxicity of Nanoparticulate Silver in Aquatic Systems. *Chemosphere* **2013**, *93, 359-365.*

Cornelis, G.; Thomas, C. D. M.; McLaughlin, M. J.; Kirby, J. K.; Beak, D. G.; Chittleborough, D. Retention and Dissolution of Engineered Silver Nanoparticles in Natural Soils. *Soil Sci. Soc. Am. J.* **2012,** *76,* 891-902.

Dale, A. L.; Lowry, G. V.; Casman, E. A. Modeling Nanosilver Transformations in Freshwater Sediments. *Environ. Sci. Technol.* **2013,** *47,* 12920-12928.

El Badawy, A. M.; Scheckel, K. G.; Suidan, M.; Tolaymat, T. The Impact of Stabilization Mechanism on the Aggregation Kinetics of Silver Nanoparticles. *Science of The Total Environment* **2012,** *429,* 325-331.

Hartmann, N.B; Skjolding, L.M.; Hansen, S.F.; Kjolholt, J.; Gottschalck, F.; Baun, A.; Environmental Fate and behaviour of nanomaterials. The Danish Environmental Protection Agency: Copenhagen, 2014.

Huynh, K. A.; Chen, K. L. Aggregation Kinetics of Citrate and Polyvinylpyrrolidone Coated Silver Nanoparticles in Monovalent and Divalent Electrolyte Solutions. *Environ. Sci. Technol.* **2011,** *45,* 5564-5571.

Kaegi, R.; Voegelin, A.; Sinnet, B.; Zuleeg, S.; Hagendorfer, H.; Burkhardt, M.; Siegrist, H. Behavior of Metallic Silver Nanoparticles in a Pilot Wastewater Treatment Plant. *Environ. Sci. Technol.* **2011,** *45,* 3902-3908.

Kent, R. D.; Vikesland, P. J. Controlled Evaluation of Silver Nanoparticle Dissolution Using Atomic Force Microscopy. *Environ. Sci. Technol.* **2012**, *46*, 6977-6984.

Kittler, S.; Greulich, C.; Diendorf, J.; Koller, M.; Epple, M. Toxicity of Silver Nanoparticles Increases During Storage Because of Slow Dissolution Under Release of Silver Ions. *Chem. Mater.* **2010,** *22,* 4548-4554.

Klein, C.; Comero, S.; Stahlmecka, B. NM-Series of representative manufactured nanomaterials NM-300 silver characterization, stability, homogeneity. 2011. Joint Research Council Scientific and Technical Reports.

Levard, C.; Reinsch, B. C.; Michel, F. M.; Oumahi, C.; Lowry, G. V.; Brown, G. E. Sulfidation Processes of PVP-Coated Silver Nanoparticles in Aqueous Solution: Impact on Dissolution Rate. *Environ. Sci. Technol.* **2011,** *45,* 5260-5266.

Li, X.; Lenhart, J. J. Aggregation and Dissolution of Silver Nanoparticles in Natural Surface Water. *Environ. Sci. Technol.* **2012,** *46*, 5378-5386.

Liu, J.; Hurt, R. H. Ion Release Kinetics and Particle Persistence in Aqueous Nano-Silver Colloids. *Environ. Sci. Technol.* **2010,** *44*, 2169-2175.

Misra, S. K.; Dybowska, A.; Berhanu, D.; Luoma, S. N.; Valsami-Jones, E. The Complexity of Nanoparticle Dissolution and Its Importance in Nanotoxicological Studies. *Science of The Total Environment* **2012,** *438*, 225-232.

Mitrano, D. M., Ranville, J. F., Bednar, A., Kazor, K., Hering, A. S., and Higgins, C. P. Tracking dissolution of silver nanoparticles at environmentally relevant concentrations in laboratory, natural, and processed waters using single particle ICP-MS (splCP-MS), *Environ. Sci. : Nano*. 2014, **1**, 248-259.

OECD. OECD Series on Testing and Assessment Number 29: Guidance Document on Transformation/Dissolution of Metals and Metal Compounds in Aqueous Media. 2001.

Peretyazhko, T.S.; Zhang, Q.; Colvin, V.L. Size-Controlled Dissolution of Silver Nanoparticles at Neutral and Acidic pH Conditions: Kinetics and Size Changes. *Environ. Sci. Technol.* **2014**, *48*, 11954-11961.

Tejamaya, M.; Romer, I.; Merrifield, R. C.; Lead, J. R. Stability of Citrate, PVP, and PEG Coated Silver Nanoparticles in Ecotoxicology Media. *Environ. Sci. Technol.* **2012,** *46*, 7011-7017.

Thio, B. J. R.; Montes, M. O.; Mahmoud, M. A.; Lee, D. W.; Zhou, D.; Keller, A. A. Mobility of Capped Silver Nanoparticles Under Environmentally Relevant Conditions. *Environ. Sci. Technol.* **2012,** *46*, 6985-6991.

WPMN. CeO2 Dossier Compiled under the Organization for Economic Cooperation and Development: Working Party on Manufactured Nanomaterials. 2013.

WPMN. ZnO Dossier Compiled under the Organization for Economic Cooperation and Development: Working Party on Manufactured Nanomaterials. 2013.

Xiu, Z. M.; Zhang, Q. B.; Puppala, H. L.; Colvin, V. L.; Alvarez, P. J. J. Negligible Particle-Specific Antibacterial Activity of Silver Nanoparticles. *Nano. Lett.* **2012,** *12*, 4271-4275.

***Secondary:***

Benn, T. M.; Westerhoff, P. Nanoparticle Silver Released into Water From Commercially Available Sock Fabrics. *Environ. Sci. Technol.* **2008,** *42*, 4133-4139.

Bone, A. J.; Colman, B. P.; Gondikas, A. P.; Newton, K. M.; Harrold, K. H.; Cory, R. M.; Unrine, J. M.; Klaine, S. J.; Matson, C. W.; Di Giulio, R. T. Biotic and Abiotic Interactions in Aquatic Microcosms Determine Fate and Toxicity of Ag Nanoparticles: Part 2 - Toxicity and Ag Speciation. *Environ. Sci. Technol.* **2012,** *46*, 6925-6933.

Borm, P.; Klaessig, F. C.; Landry, T. D.; Moudgil, B.; Pauluhn, J.; Thomas, K.; Trottier, R.; Wood, S. Research Strategies for Safety Evaluation of Nanomaterials, Part V: Role of Dissolution in Biological Fate and Effects of Nanoscale Particles. *Toxicological Sciences* **2006,** *90*, 23-32.

Chappell, M. A.; Miller, L. F.; George, A. J.; Pettway, B. A.; Price, C. L.; Porter, B. E.; Bednar, A. J.; Seiter, J. M.; Kennedy, A. J.; Steevens, J. A. Simultaneous Dispersion-Dissolution Behavior of

Concentrated Silver Nanoparticle Suspensions in the Presence of Model Organic Solutes. *Chemosphere* **2011,** *84,* 1108-1116.

Chen, S.; Zhang, H.; Lin, Q. Effect of different water conditions on dissolution of nanosilver. *Water Science and Technology* **2013**, *68, 8¸* 1745-1750.

David, C.A.;Galceran, J.; Rey-Castro, C.; Puy, J.; Companys, E.; Salvador, J.; Monné, J.; Wallace, R.; Vakourov, A.Dissolution Kinetics and Solubility of ZnO Nanoparticles Followed by AGNES. *J. Phys. Chem*. **2012**,*116, 11758–11767.*

Dobias, J.; Bernier-Latmani, R. Silver Release from Silver Nanoparticles in Natural Waters. *Environ. Sci. Technol.* **2013**, *47,* 4140-4146.

El Badawy, A. M.; Silva, R. G.; Morris, B.; Scheckel, K. G.; Suidan, M. T.; Tolaymat, T. M. Surface Charge-Dependent Toxicity of Silver Nanoparticles. *Environ. Sci. Technol.* **2010,** *45,* 283-287.

Elzey, S.; Grassian, V. Agglomeration, Isolation and Dissolution of Commercially Manufactured Silver Nanoparticles in Aqueous Environments. *J. Nanopart. Res.* **2010,** *12,* 1945-1958.

Gondikas, A. P.; Morris, A.; Reinsch, B. C.; Marinakos, S. M.; Lowry, G. V.; Hsu-Kim, H. Cysteine-Induced Modifications of Zero-Valent Silver Nanomaterials: Implications for Particle Surface Chemistry, Aggregation, Dissolution, and Silver Speciation. *Environ. Sci. Technol.* **2012,** *46,* 7037-7045.

He, D.; Dorantes-Aranda, J. J.; Waite, T. D. Silver Nanoparticle - Algae Interactions: Oxidative Dissolution, Reactive Oxygen Species Generation and Synergistic Toxic Effects. *Environ. Sci. Technol.* **2012,** *46,* 8731-8738.

Hedberg, J.; Lundin, M.; Lowe, T.; Blomberg, E.; Wold, S.; Wallinder, I. O. Interactions Between Surfactants and Silver Nanoparticles of Varying Charge. *Journal of Colloid and Interface Science* **2012,** *369,* 193-201.

Janssen, C.R.; De Schamphelaere, K.; Heijerick, D.; Muyssen, B.; Lock, K.; Bossuyt, B. Uncertainties in the environmental risk assessment of metals. *Hum. Ecol. Risk Assess.*, **2000**, *6,* 1003-1018.

Janssen, C.R.; Heijerick, D.G.; De Schamphelaere, K.A.C.;, Allen, H.E. Environmental risk assessment of metals: tools for incorporating bioavailability. *Environment International*. **2003**, *28, 8,* 793-800.

Kent, R. D.; Oser, J. G.; Vikesland, P. J. Controlled Evaluation of Silver Nanoparticle Sulfidation in a Full-Scale Wastewater Treatment Plant. *Environ. Sci. Technol.* **2014**, 48, 8564-8572.

Kroll, A.; Hahn, D.; Wohlleben, W.; Schulze Isfort, C.; Göbbert, C.; Voetz, M.; Hardinghaus, F.; Schnekenburger, J. Cytotoxicity screening of 23 engineered nanomaterials using a test matrix of 10 cell lines and 3 assays. *Particle and Fibre Toxicology* **2011,** *8, 9.*

Lazarus, L. L.; Riche, C. T.; Malmstadt, N.; Brutchey, R. L. Effect of Ionic Liquid Impurities on the Synthesis of Silver Nanoparticles. *Langmuir* **2012,** *28,* 15987-15993.

Levard, C.; Hotze, E. M.; Lowry, G. V.; Brown, G. E. Environmental Transformations of Silver Nanoparticles: Impact on Stability and Toxicity. *Environ. Sci. Technol.* **2012,** *46,* 6900-6914.

Levard, C.; Mitra, S.; Yang, T.; Jew, A.D.; Badireddy, A.R.; Lowry, G.V.; Brown, G.E. Effect of Chloride on the Dissolution Rate of Silver Nanoparticles and Toxicity to E. *coli*. *Environ. Sci. Technol.* **2013,** *47,* 5738-5745.

Li, X.; Lenhart, J. J.; Walker, H. W. Dissolution-Accompanied Aggregation Kinetics of Silver Nanoparticles. *Langmuir* **2010,** *26,* 16690-16698.

Li, X.; Lenhart, J. J.; Walker, H. W. Aggregation Kinetics and Dissolution of Coated Silver Nanoparticles. *Langmuir* **2012***, 28, 1095-1104.*

Lowry, G. V.; Espinasse, B. P.; Badireddy, A. R.; Richardson, C. J.; Reinsch, B. C.; Bryant, L. D.; Bone, A. J.; Deonarine, A.; Chae, S.; Therezien, M.; Colman, B. P.; Hsu-Kim, H.; Bernhardt, E. S.; Matson, C. W.; Wiesner, M. R. Long-Term Transformation and Fate of Manufactured Ag Nanoparticles in a Simulated Large Scale Freshwater Emergent Wetland. *Environ. Sci. Technol.* **2012,** *46,* 7027-7036.

Ma, R.; Levard, C.; Marinakos, S. M.; Cheng, Y.; Liu, J.; Michel, F. M.; Brown, G. E.; Lowry, G. V. Size-Controlled Dissolution of Organic-Coated Silver Nanoparticles. *Environ. Sci. Technol.* **2011,** *46,* 752-759.

MERCER, T. T. 1967.On the role of particle size in the dissolution of lung burdens. *Health Physics,* **1967***, 13***,** *1211-21.*

Mittelman, A.M.; Taghavy, A.; Wang, Y.; Abriola, L.M.; Pennell, K.D.; Influence of dissolved oxygen on silver nanoparticle mobility and dissolution in water-saturated quartz sand. *J. Nanopart. Res.* **2013***, 15,* 1765-1778.

Nel, A.; Xia, T.; Madler, L.; Li, N. Toxic Potential of Materials at the Nanolevel. *Science* **2006,** *311,* 622-627.

Ostermeyer, A.; Mumuper, C.K.; Semprini, L.; Radniecki, T. Influence of Bovine Serum Albumin and Alginate on Silver Nanoparticle Dissolution and Toxicity to *Nitrosomonas europaea. Environ. Sci. Technol.* **2013**, *47*, 14403-14410.

Prathna, T. C.; Chandrasekaran, N.; Mukherjee, A. Studies on Aggregation Behaviour of Silver Nanoparticles in Aqueous Matrices: Effect of Surface Functionalization and Matrix Composition. *Colloids and Surfaces A: Physicochemical and Engineering Aspects* **2011,** *390,* 216-224.

Pokhrel, L.R.; Dubey, B.; Schueurman, P.R. Impacts of Select Organic Ligands on the Colloidal Stability, Dissolution Dynamics, and Toxicity of Silver Nanoparticles. *Environ. Sci. Technol.* **2013**, *47,* 12877-12885.

Reinsch, B. C.; Levard, C.; Li, Z.; Ma, R.; Wise, A.; Gregory, K. B.; Brown, G. E.; Lowry, G. V. Sulfidation of Silver Nanoparticles Decreases Escherichia Coli Growth Inhibition. *Environ. Sci. Technol.* **2012,** *46,* 6992-7000.

Schlich, K.; Klawonn, T.; Terytze, K.; Hund-Rinke, K. Effects of Silver Nanoparticles and Silver Nitrate in the Earthworm Reproduction Test. *Environmental Toxicology and Chemistry* **2013,** *32,* 181-188.

Unrine, J. M.; Colman, B. P.; Bone, A. J.; Gondikas, A. P.; Matson, C. W. Biotic and Abiotic Interactions in Aquatic Microcosms Determine Fate and Toxicity of Ag Nanoparticles. Part 1. Aggregation and Dissolution. *Environ. Sci. Technol.* **2012,** *46,* 6915-6924.

Zhang, W.; Yao, Y.; Li, K.; Huang, Y.; Chen, Y. Influence of Dissolved Oxygen on Aggregation Kinetics of Citrate-Coated Silver Nanoparticles. *Environmental Pollution* **2011,** *159,* 3757-3762.

Zhao, C.-M.; Wang, W.-X. Importance of Surface Coatings and Soluble Silver in Silver Nanoparticles Toxicity to Daphnia Magna. *Nanotoxicology* **2011,** *6,* 361-370.

Zook, J. M.; Halter, M. D.; Cleveland, D.; Long, S. E. Disentangling the Effects of Polymer Coatings on Silver Nanoparticle Agglomeration, Dissolution, and Toxicity to Determine Mechanisms of Nanotoxicity. *J. Nanopart. Res.* **2012,** *14,* 1-9.

Zook, J. M.; Long, S. E.; Cleveland, D.; Geronimo, C. L. A.; MacCuspie, R. I. Measuring Silver Nanoparticle Dissolution in Complex Biological and Environmental Matrices Using UV-Visible Absorbance. *Anal. Bioanal. Chem.* **2011,** *401,* 1993-2002.