



29 and predisposing risk factors, such as stratum corneum disruption due to occupational co-  
30 exposure to chemicals, and the presence of occupational skin diseases. Attention should be  
31 given to: 1) Metal NOAA, since the potential release of ions may induce local skin effects  
32 (e.g. irritation and contact dermatitis) and absorption of toxic or sensitizing metals; 2) NOAA  
33 with metal catalytic residue, since potential release of ions may also induce local skin effects  
34 and absorption of toxic metals; 3) rigid NOAA smaller than 45 nm that can penetrate and  
35 permeate the skin; 4) non rigid or flexible NOAA, where due to their flexibility liposomes  
36 and micelles can penetrate and permeate the intact skin; 5) impaired skin condition of  
37 exposed workers.

38 Furthermore, we outline possible situations where health surveillance could be appropriate  
39 where there is NOAA occupational skin exposures, e.g. when working with nanoparticles  
40 made of sensitizer metals, NOAA containing sensitizer impurities, and/or in occupations with  
41 a high prevalence of disrupted skin barrier integrity. The paper furthermore recommends a  
42 stepwise approach to evaluate risk related to NOAA to be applied in occupational exposure  
43 and risk assessment, and discusses implications related to health surveillance, labelling, and  
44 risk communication.

45

46

47        **Introduction**

48        The potential for nanoparticles, nano-objects, their agglomerates and aggregates, (NOAA,  
49 defined as having at least one dimension <100nm) to enter the body through intact skin has  
50 been a controversial issue, with some authors asserting that nanoparticles can pass through  
51 the stratum corneum, while others disputing this conclusion (Oberdoster et al., 2005, SCCP  
52 2007; Crosera et al., 2009, Labouta and Schneider 2013, Larese Filon et al., 2015),.

53        The skin is a complex organ system comprising the epidermis and dermis, with hair  
54 follicles and sweat glands providing pathways across these layers, and peripheral blood  
55 flowing into the dermis. The epidermis mainly comprises keratinocytes that migrate from the  
56 basal layer towards the skin surface forming the outer protective layer (stratum corneum).  
57 The intact stratum corneum provides an effective barrier against bacteria, viruses and most  
58 exogenous chemicals. However, the barrier is not completely impervious and it is possible for  
59 relatively small molecules, and in theory very small particles, to diffuse across the stratum  
60 corneum via cellular and/or inter-cellular pathways. If the barrier is damaged (disrupted) then  
61 permeation may be enhanced.

62        The focus of this work is to review and critically assess the evidence on the relevance and  
63 relative significance of various skin uptake pathways for NOAA, especially in occupational  
64 settings, in the context of nanotoxicology, risk assessment, occupational medicine, medical  
65 and epidemiological surveillance efforts, and in development of relevant exposure assessment  
66 strategies. Skin uptake of nanoparticles is presented in the context of local and systemic  
67 health effects, especially contact dermatitis, skin barrier integrity, physico-chemical  
68 properties of NOAA, and predisposing risk factors, such as stratum corneum disruption due  
69 to work, co- chemical exposures, and presence of occupational skin diseases. In the  
70 accompanying paper by Brouwer et al., (2016) these findings are integrated in an approach

71 for evaluating occupational dermal exposure to nanoparticles. Dermal exposure is approached  
72 both conceptually and from the perspective of evidence for exposure, by linking the use of  
73 NOAA and nano-enabled products in industrial sectors to job titles. In addition, we flagged  
74 specific job titles where there is often a high incidence rate of skin barrier disruption and skin  
75 disease. We conclude with recommendations for occupational health practitioners and risk  
76 assessors.

77 In this paper, the term nanoparticle includes both engineered and incidental nanoparticles, as  
78 well as their agglomerates and aggregates (ISO, 2011). Nanoparticles embedded in nano-  
79 enabled products, such as pastes, paints, glues, etc., are potential sources of dermal exposure  
80 to nanoparticles (Aitken et al., 2004, 2006). The term NOAA (nano-objects, and their  
81 aggregates and agglomerates) is used throughout the paper to refer inclusively to such  
82 nanoparticles. The terms penetration and permeation are used throughout the paper to mean  
83 that NOAA can reach the skin layers and pass through the skin respectively.

84

## 85 **Methods**

86 **Literature review:** An extensive literature search was conducted in major databases,  
87 including Pubmed, Thompson Reuters Web of Science (ISI), and Google Scholar using  
88 search terms “skin absorption nanoparticles” or “skin penetration nanoparticles” or “skin  
89 exposure nanoparticles”, “sensitizer and nanoparticles”, “engineered nanoparticles and skin”  
90 and similar terms. The period taken into consideration was from 1999 to 31st-12-2015. A  
91 total of 810 papers were selected and 132 analysed. The skin absorption data were presented  
92 in detail an earlier paper by the authors (Larese et al., 2015) and are summarized here for  
93 completeness.

94 The search for available studies on contact dermatitis in workers was performed on the same  
95 database using the term “occupational contact dermatitis” and epidemiology, “irritant contact  
96 dermatitis” and epidemiology. A total of 176 papers were selected and 127 were analyzed.  
97 Additional searches on these same databases and internal databases available at co-authors’  
98 institutions were performed for occupational skin disorders and occupational disease burden  
99 by industry sectors. Additional relevant information not available in the peer-reviewed  
100 literature (such as reports, white papers, personal communications) from authors’  
101 bibliographies were also analysed.

102 The abstracts of all studies were reviewed and only papers that were deemed relevant to the  
103 current objectives were analysed in detail. 132 and 127 papers were included in the final  
104 analysis.

105 **Summary data on physico-chemical (PC) properties of NOAA and impurities.** Certain  
106 metals (e.g. nickel Ni) are known to cause allergic contact dermatitis and such metals can be  
107 found as engineered nanomaterials, or as impurities in NOAA (Bello et al., 2009). For this  
108 reason, we conducted a detailed analysis for metals in NOAA. In generating summary data on  
109 PC properties of NOAA and their impurities, authors conducted summary statistical analysis  
110 using a large dataset of their own ENM (Hsieh et al., 2013). Some data on PC properties of  
111 subclasses of NOAA have been presented in earlier work in the context of exploring links  
112 between PC properties and biological oxidative damage, in vitro nanotoxicology, and  
113 exposure assessment (Bello et al., 2009; Hsieh et al., 2013). The summary analysis across all  
114 available NOAA is new, and utilizes in part a substantial subset of unpublished PC data. The  
115 methods for chemical analysis of metals (total and water soluble), organic and elemental  
116 carbon, and polycyclic aromatic hydrocarbon (PAHs) have been presented elsewhere (Bello  
117 et al., 2009) and includes sector field inductively coupled plasma mass spectrometry (SF ICP-

118 MS), thermogravimetric analysis for carbon speciation into organic and elemental (OC/EC),  
119 and gas chromatography mass spectrometry GC-MS for PAHs.

## 120 RESULTS

### 121 **Penetration of NOAA through the skin**

122 NOAAAs on the skin may penetrate stratum corneum reaching viable epidermis using  
123 different pathways, namely: a) via sweat glands and hair follicles (Lademann et al., 2009),  
124 which are probably the most efficient way for penetration and permeation of large molecules  
125 and nanoparticles; b) via the intercellular route, which is likely only possible for very small  
126 NOAAAs (<1 to 4 nm, the size of intercellular keratinocyte space) or under conditions where  
127 the skin barrier is disrupted. The intracellular pathway (Scheuplein et al., 1965, 1967) used by  
128 chemical substances and ions is not relevant for NOAAAs. Skin properties per body parts are  
129 relevant for one of the pathways mentioned above. Follicular density varies greatly between  
130 different body parts, highly in forehead and lower in calf and thigh. The surface density of  
131 hair follicles, which varies by anatomical site and ethnicity, can cover up to 13.7% of skin  
132 surface on the forehead but only 0.95% on the forearm (Otberg et al., 2004). Thickness of the  
133 skin also varies by body parts. The stratum corneum is thicker in palms and soles (up to 175  
134 and 500  $\mu\text{m}$ , respectively), and much thinner in other anatomical sites (e.g. 22.6-6.4  $\mu\text{m}$  on  
135 the abdomen with differences related to the method used; Holbrook and Odland, 1974;  
136 Egawa et al., 2007, Robertson and Rees, 2010, Huzaira et al., 2001).

137 Watkinson et al. (2013) considered that NOAAAs behave like large molecules and  
138 modelled their rate of penetration using diffusion theory. They concluded that only particles  
139 of 1 nm or less are small enough to pass through intact skin. One would further assume that  
140 in healthy, intact skin, nanoparticles larger than  $\sim 4$  nm (maximum intercellular space) cannot  
141 normally penetrate. However, there are experimental data that show that NOAAAs larger than  
142 this size can pass through disrupted skin where intercellular gaps are larger than in normal

143 skin (Labouta et al., 2013; Monteiro-Riveira & Larese Filon, 2012; Monteiro-Riveira &  
144 Riviere 2009, Larese Filon et al., 2009-2013, Poland et al. 2013).

145 The skin penetration and permeation of NOAAs is affected by many factors, including  
146 NOAA primary size, NOAA physico-chemical properties (such as rigidity/flexibility of the  
147 nanostructure, dissolution rate in water/sweat, and morphology), and skin health. Such factors  
148 have been analysed and presented in the following sections.

#### 149 *1. NOAA size*

150 NOAAs characteristics may change considerably when they interact with physiological  
151 media. Airborne NOAAs, which are emitted as individual nanoparticles, can subsequently  
152 agglomerate and settle on the skin and or surfaces. Therefore, the skin will come into contact  
153 mostly with agglomerates of NOAAs, especially because skin contact with contaminated  
154 surfaces and objects is a major exposure pathway. Direct contact of individual airborne  
155 NOAAs with the skin can be approached in a manner similar to gases, a process controlled by  
156 laws of diffusion (see Brouwer et al., 2016). The forces that control this deposition process  
157 depend on the primary particle size and aerodynamic behaviour of NOAAs. Once on the skin,  
158 biokinetics and transformation of NOAAs will depend on adhesion forces to the skin,  
159 interaction with sebaceous fluids and sweat, chemical stability and dissolution behaviour  
160 following such interactions. For that reason, it is critically important to characterise NOAAs  
161 behaviour in physiological media relevant to skin (i.e. sweat) to verify size modifications and  
162 rate of size change of NOAA. Changes in size towards smaller nanoparticles can enable  
163 NOAAs to pass through the skin more easily than the original NOAAs. Sonavane (2008) for  
164 example reported a greater permeation through top layers of rat skin for 15 nm AuPN  
165 compared to 102 nm particles. Rancan (2012) demonstrated that only silica NOAAs smaller  
166 than 42 nm can penetrate the skin through hair follicles and be internalized by Langerhans

167 cells (mostly) and keratinocytes in a damaged skin model. Larger NOAAs did not pass into  
168 hair follicles. Quantum dots (QD) of 37 nm were observed to permeate the mouse skin only  
169 if the skin barrier was disrupted by dermo-abrasion (Gopee et al., 2009). Smaller QD (4 nm)  
170 have been shown to penetrate intact skin (Chu et al., 2007). Some flexible NOAA (liposomes  
171 and micelles) due to their flexibility can penetrate and permeate the intact skin also at sizes  
172 >4 nm. Larese et al. published a detailed review (2015) on this topic and defined those  
173 critical sizes.

174 Therefore, it can be concluded from available data and anatomical and physiological  
175 considerations of normal intact human skin that for rigid NOAA size is perhaps one of the  
176 most, if not the most, important factor influencing skin permeation/penetration. Figure 1  
177 illustrates these concepts and table 1 summarized some relevant data from literature.

- 178 • For NOAA greater than 45 nm (primary or agglomerate size), no skin penetration and  
179 permeation is expected in healthy skin with normal barrier properties. However,  
180 penetration and permeation of NOAA > 45 nm, up to a few microns) can happen in  
181 severely damaged skin.
- 182 • For NOAA 21-45 nm, penetration and permeation can happen only in damaged skin.
- 183 • For NOAA 4-20 nm, there is possible permeation and penetration, which happens  
184 mostly through the hair follicles.
- 185 • For NOAA <4 nm: skin penetration has been demonstrated and this is consistent with  
186 expectations based on skin physiology and diffusion theory (for <1 nm) (see Larese et  
187 al., 2015 for detail).

## 188 2. NOAA Surface properties

189 NOAA surface properties, including surface charge, functional groups, Z potential, can  
190 influence penetration and permeation but their role in skin penetration is not clear and must  
191 be evaluated for each NOAAs. For some Quantum dots the surface charge as well as pH may  
192 influence penetration (Rymann-Rasmussen et al., 2006). Protein corona can play an important  
193 role in NOAA biokinetics and translocation inside the body, however the nature, role, and  
194 significance of protein corona on skin absorption of NOAA are poorly understood. Contact  
195 with solvents and oils can influence significantly skin absorption of NOAA by modifying  
196 skin permeability and/or nanoparticle diffusivity, and needs to be evaluated on a case-by-case  
197 basis. The data on factors related to impact of surface properties of nanoparticles on skin  
198 permeation/penetration is limited, yet highly relevant for occupational settings where co-  
199 exposures are common.

### 200 3. NOAA dissolution biokinetics, ions release and impurities

201 NOAA dissolution in sweat, skin-associated water and other biomolecules, is of critical  
202 importance because some metal NOAAs (such as Ni<sup>2+</sup>) are known to cause skin sensitization  
203 and allergic dermatitis. Dissolution rates of NOAAs on the skin have not been investigated  
204 experimentally, however it is expected that they have higher rates (i.e. produce a higher ionic  
205 flux) than the corresponding micron sized particles, because of their much higher  
206 surface/mass ratio. NOAAs can reach hair follicles where they can reside and release ions for  
207 a long period. That may increase the risk of allergic contact dermatitis for NOAA containing  
208 sensitizing metals such as Ni, Pd, Co (Larese et al., 2013; Journeay and Goldman, 2014).  
209 Skin pH and sweat are expected to enhance NOAA dissolution, enhancing metal release.

210 Impurities in NOAA have received considerable attention in the context of inhalation  
211 exposures and associated respiratory and systemic diseases (Donaldson et al., 2006; Hsieh et  
212 al., 2012; Guo et al., 2007) but little attention has been paid to skin exposures and associated

213 skin diseases. These impurities may include transition metals used as catalysts in the  
214 manufacture of carbon nanotubes (e.g. nickel, chromium, cobalt), organic impurities  
215 including polyaromatic hydrocarbons (PAHs) and other carbonyl compounds produced  
216 during the gas phase synthesis of several NOAA (especially CNTs), and inorganic impurities  
217 present in the raw materials used in the production of primary NOAA. These impurities can  
218 be carried through the skin by NOAA and then be released from NOAA leading to both  
219 localized and systemic adverse effects. Possible mechanistic interactions of impurities with  
220 nanoparticles in the development of skin disease have not been studied, but they may be  
221 particularly important in certain conditions, such as allergic contact dermatitis.

222

223 PAHs have been found in CNTs, carbonaceous ENMs (such as carbon black), and  
224 combustion by-products absorbed on surfaces of ENM (Plata et al., 2008). Supplemental  
225 Table S1 and S2 provide data on PAHs and organic carbon content (OC), respectively, in  
226 various classes of NOAA, collected as part of this work. OC is used as a surrogate for total  
227 organics and an index of organic impurity content. Note that carbon blacks in particular and  
228 refined fullerenes did contain several PAHs such as pyrene (~5 ppm), phenanthrene (4.7 ppm),  
229 fluoranthene, Indeno (1,2,3-cd) pyrene (up to 18 ppm), and Benzo (ghi) perylene (up to 30  
230 ppm). Several PAHs are known human carcinogens.

231

232 Table S3 summarized the total content of selected metals relevant to skin exposure, especially  
233 in the context of skin sensitization (see later section on skin disease) for different classes of  
234 NOAA. The distributions of such elements are typically right skewed, and geometric mean  
235 (GM), geometric standard deviation (GSD) and maximum values in a range of commercially  
236 relevant NOAAs are provided. The water-soluble fraction of these metals, an important

237 indicator of the likelihood of metal ions release (which are believed to be involved in  
238 sensitization), is also presented. Several observations in Table S3 are important to note:

239 i) Ni and Cr, and to some extent Co as well, were present in appreciable amounts in  
240 many commercial CNTs; GM ranging from ~10 ( $\mu\text{g/g}$ ) to 800 ( $\mu\text{g/g}$ ) and maxima  
241 as high as 1.2% (Ni); Interestingly, high concentrations of several transition  
242 metals, including Ni, Cr, Co, etc. have been found in tattoo inks, which often  
243 employ nanoscale NOAA (Hogsberg et al., 2011; Forte et al., 2009).

244 ii) Pd and As were present mostly in trace impurities in ng/g (ppb range). One notable  
245 exception was one high volume  $\text{TiO}_2$  commercial sample, which contained 50  
246  $\mu\text{g/g}$  As. Similarly, Zr was found only in certain metal oxide NOAA, notably ZnO,  
247  $\text{CeO}_2$ , and  $\text{TiO}_2$ . Zr, As and certain other metals (Fe in CB for example) are likely  
248 related to impurities in raw materials (e.g. natural ores). One zirconia sample in  
249 the dataset contained 200  $\mu\text{g/g}$  Cadmium (Cd), 5  $\mu\text{g/g}$  platinum, and 45  $\mu\text{g/g}$   
250 Yttrium (Y, added sometimes as a stabilizer). Cd and Pd are likely impurities.

251 iii) The water-soluble content of Ni, Cr, Co varied by NOAA type, with GM in the 0.001-  
252 7 ( $\mu\text{g/g}$ ) range. Water solubility varied by metal and NOAA type. The GM ratio of  
253 water soluble to total metal size distributions (i.e. GM water soluble/GM total  
254 metal) varied in the 0.2-28% range for Ni, 0.05-8% (Cr) and 0.3-80% for Co  
255 (Table 2). In CNTs, where these elements were in higher concentrations, this GM  
256 ratio was <1%; however, much higher water solubility has been observed for these  
257 metals when they appear as impurities in other NOAA (e.g.  $\text{TiO}_2$ ,  $\text{CeO}_2$  or ZnO).

258

## 259 **Effects of NOAA on the skin**

### 260 **Irritation**

261 Mechanical friction between solid objects and the skin can cause abrasion, damage to  
262 the thickness of the SC, and skin irritation. Early on Eedy (1996) reported irritant contact  
263 dermatitis in workers exposed to relatively coarse carbon fibers in micrometer range.  
264 However, more recent data shows no dermal irritation in guinea pigs exposed to carbon  
265 nanotubes (Khisore et al., 2009).

266 Experimental evidence regarding NOAA skin exposure and disease is also limited.  
267 Ema et al (2011) investigated acute skin and eye irritation and skin sensitization potential of  
268 three types of CNTs in rabbits and guinea pigs respectively and demonstrated that only one  
269 MWCNT (out of three tested) was a very weak acute irritant to the skin and eyes (Ema et al.,  
270 2011). Similarly, Park et al. (2011) demonstrated that polystyrene and titania nanoparticles  
271 did not induce phototoxicity, acute skin irritation, or skin sensitization in animals (rabbits,  
272 mice). However, subchronic skin exposures to TiO<sub>2</sub> could induce inflammation of the  
273 epidermis, leading to effects such as focal parakeratosis (flattened keratinocyte nuclei within  
274 the stratum corneum) and spongiosis (intercellular oedema between keratinocytes), (Adachi  
275 et al. 2013) whereas chronic exposures to TiO<sub>2</sub> may accelerate skin aging (Wu et al. 2009).  
276 Highly purified fullerenes were shown to be ‘minimally irritating’ to the skin and eyes, and  
277 did not present a problem with regard to skin irritation, skin sensitization, skin  
278 photosensitization or contact phototoxicity (Aoshima et al. 2009). Overall the available  
279 limited evidence suggests minimal effects of NOAA in human intact skin.

280 Metal (ions) of Ni, Co, Hg, and Cr (as soluble salts, e.g. sulfate or chloride), as well as  
281 antimony (Sb, as trioxide), and arsenic (as trioxide) are known skin irritants (Cohen and  
282 Moore 2007).

### 283 **Sensitization**

284 Several transition metals are known to cause sensitization and allergic contact dermatitis.  
285 There is further evidence of possible risk from exposure to metal NOAA or metal impurities

286 in NOAA. Several metals, including nickel (Ni), chromium (Cr), cobalt (Co), beryllium  
287 (Be), and palladium (Pd), are well-known skin allergens (Cohen and Moore et al., 2007; Rice  
288 & Mauro, 2008). Nickel, Cr, Co, Au, and Pd are available commercially as metallic  
289 engineered nanoparticles of various sizes. Most of these elements, except for Be, Hg and As,  
290 are commercially available as metal oxides nanoparticles, or as components of more complex  
291 nanoparticle chemistries ([http://www.nanowerk.com/phpscripts/n\\_dbsearch.php](http://www.nanowerk.com/phpscripts/n_dbsearch.php)). Q-dots,  
292 another type of engineered nanoparticle, often contain cadmium selenide (CdSe) or cadmium  
293 sulfide (CdS), sometime mixed with other metals (e.g. Zn). They can release Cd causing  
294 intoxication, as already demonstrated in animals (Chu et al., 2007; Liu et al., 2011).  
295 Nickel in jewellery is a classic example of Ni ions leaching over time and reaching the  
296 epidermis, leading to development of allergic contact dermatitis in various individuals. One  
297 case report already describes nickel NOAAs as causing asthma and skin diseases (Journey et  
298 al., 2014). NOAAs can release ions in higher amounts than bulk material due to their high  
299 surface/mass ratio. For that reason, NOAAs containing sensitizing metal/s may more easily  
300 trigger an allergic response than the corresponding microscopic bulk materials of the same  
301 composition.

302 On the other hand, it has been suggested that fullerenes may play a leading role in the  
303 inhibition of the in vitro and in vivo IgE-mediated allergic response, thus blocking histamine  
304 release or reducing nickel uptake after the application of a cream containing fullerenes  
305 (Vermula et al., 2011).

306

### 307 **Skin Diseases**

308           There is only one case report of contact dermatitis (CD) and asthma in a woman  
309 exposed to nickel NOAAs (Journey and Goldman, 2014). There are no other observational

310 data related to workplace NOAA skin exposures and skin disease, even though the authors  
311 have witnessed numerous scenarios of extensive NOAA skin exposure.

312

### 313 *Tattooing*

314 Tattooing in humans is a relevant and interesting scenario to analyse, because tattoo  
315 inks contain engineered nanoparticles, and because injected ink is delivered in the dermis  
316 (Hogsberg et al., 2011, 2013a). In a recent study among young individuals tattooed with  
317 carbon black and organic pigments, 16% complained of mostly minor symptoms, including  
318 skin itching, skin elevation/nodules, inflammation and stinging, with over half of them being  
319 sun induced (Hogsberg et al., 2013b).

320

### 321 **Factors involved in skin barrier function integrity**

322

#### 323 **Mechanical action**

324 Rouse et al. (2007) demonstrated that mechanical flexion can increase skin  
325 penetration of small fullerene (3.5 nm) that can be found in the intercellular spaces of stratum  
326 granulosum. On the contrary QDs applied to rat skin flexed for 60 min showed that larger  
327 nanoparticles QD655-COOH (18nm) and QD565-COOH (14nm) did not penetrate at 8 and  
328 24h (Zhang et al., 2008).

329

#### 330 **Skin barrier disruption**

331 Skin barrier disruption is a crucial aspect for NOAA skin penetration and permeation,  
332 so particular attention should be paid to workers who are at increased risk of irritant contact  
333 dermatitis or to atopic patients with an impaired skin barrier.

334 In certain occupations, such as construction, CD is prevalent and the disease causation  
335 in such settings is often multifactorial. The high market penetration by NOAA in this industry  
336 and potential for significant interactions of NOAA with damaged skin should be noted.  
337 Authors are not aware of any ongoing surveillance or epidemiological studies focusing on  
338 skin disease among cohorts of nanomanufacturing workers. They recommend the avoidance  
339 of skin contact with NOAA containing products and to undergo medical surveillance, with  
340 particular attention to skin conditions and skin diseases.

341 Occupational skin diseases are prevalent in most countries. More than 90% of  
342 occupational skin diseases are classified as CD (EU-OSHA, 2008). Acute irritant CD may  
343 occur as a result of exposure to strong irritants such as acids or alkalis, whereas chronic  
344 irritant CD can be caused by repeated exposure to mild irritants such as water (from wet  
345 work), soaps and detergents. Wet work is common amongst occupations such as hairdressers,  
346 food workers, cleaners and healthcare workers. Allergic CD is caused by an immunological  
347 reaction following exposure to an allergen or a sensitizer. In many cases, irritant CD can  
348 exacerbate the effects of skin sensitizers because of damage to the skin barrier (Elsner et al.  
349 1994).

350 Skin permeability may increase 4 to 100 times in atopic subjects with damaged skin (Larese  
351 et al. 2009, 2011) and it is possible for the skin barrier to be compromised, although there are  
352 no visible signs (Kezic et al., 2009).

353 Frequent, repetitive exposure to water or other irritant chemicals results in disruption  
354 of the lipid bilayers in the stratum corneum, which can lead to chapping and fissuring of the  
355 skin (Chew and Maibach, 2003). In some work situations, there may be exposure to more  
356 than one irritant, for example, in addition to wet work, healthcare workers are likely to be  
357 exposed to cleansers, detergents and disinfectants.

358 Other hazards that may influence the integrity of the skin barrier include mechanical  
359 abrasion or friction caused by dusts or powders of the skin, cuts and punctures. Further,  
360 exposure to cold, heat, and pressure may lead to skin alteration and vibration can induce  
361 sklerodermal effects (EU-OSHA, 2008). Exposure to these physical agents may affect an  
362 individual's response to other chemical agents, allowing them to penetrate the skin more  
363 easily (CCOHS, Fluhr et al. 2002, 2008).

364 The commonest causes of dermatitis are wet work, soaps and cleaners, solvents, degreasing  
365 agents, metal working fluids, dusts/friction and low humidity (HSE, 2014; Pal et al., 2009;  
366 Cahill et al., 2012, Behroozy and Keegel, 2014). For example, Cahill et al. (2012) report the  
367 most common causes in patients with a primary diagnosis of irritant CD – water and wet  
368 work (37%), soap and detergents (33%), heat and sweating (16%), oils and coolants (14%),  
369 solvents (14%), dusts and fibres (10%), acids and alkalis (4%). Wet work includes activities  
370 where there is prolonged contact for more than two hours a day, frequent or intensive hand  
371 washing and where liquid-tight protective gloves are worn for extended periods (BAuA  
372 2008). Other common agents where exposure increases the risk of dermatitis include  
373 hairdressing products, preservatives, rubber chemicals, cement, nickel, chromium and  
374 chromates, cobalt, resins and acrylics, cosmetics and fragrances, petroleum and products,  
375 disinfectants, degreasers and cutting oils and coolants (HSE, 2014; Carøe et al., 2013).

376

### 377 **Overall consideration**

378 Taking into consideration the limited penetration by NOAA through intact skin, and  
379 the easy release of metals or other impurities in nanoparticles by dissolution in the skin or  
380 skin contamination layer, it is reasonable to hypothesize that: i) skin exposure to NOAA in  
381 general may present more concerns where there is compromised skin integrity due to pre-  
382 existing disease or exposure to other factors (e.g. abrasion); ii) susceptible subpopulations

383 may be particularly at risk for allergic skin disease, especially following dermal contact with  
384 nanoparticles containing sensitizing metals, and iii) although not the primary focus of this  
385 paper, in an accompanying paper we make the argument that skin exposure should be  
386 investigated as a potentially significant pathway for ingestion of NOAA (Cherrie et al., 2006,  
387 Christopher et al., 2007; Gorman et al., 2012, 2014).

388

## 389 **RECOMMENDATIONS FOR HAZARD ASSESSMENT**

390 Taking into account the literature reviewed in the previous sections, hazard assessment  
391 should consider the following steps:

- 392 1. Evaluation of NOAA, using the diagram reported in Figures 1, 2 and 3.
- 393 2. Evaluation of skin condition of exposed workers
- 394 3. Evaluation of jobs at high risk for occupational dermatitis (irritant and allergic CD)
- 395 4. Evaluation of jobs with use of NOAA

396

- 397 1. Evaluation of NOAA

398 If applicable, assessment of dermal exposure to NOAA should be incorporated in the general  
399 cycle of risk assessment in companies to control risks in the workplace. With respect to  
400 assessment of dermal exposure to NOAA in the workplace, a stepwise approach is proposed  
401 to assess the situation in the workplace in a systematic manner that focuses on determining  
402 the potential for exposure based on a potential for release and determining the potential for  
403 skin disruption. A stepwise approach is given, of which the first step is described in this  
404 paper, and the other steps are described in the accompanying paper of Brouwer et al. (2016).  
405 After each step, a decision should be made whether the situation at the workplace is  
406 considered to be safe based on the information that is gathered during that part of the

407 assessment. If the situation is not considered to be safe, one should proceed to the following  
408 step of the assessment (Figure 3).

409 Step 1. (Primary evaluation based on the NOAA composition) consists of a primary (desk)  
410 evaluation of the occurrence of possible health risks based on the composition /  
411 characteristics of NOAA. In Figure 1 and 3 a schematic overview of this evaluation and the  
412 further course of the overall assessment is given.

413 Attention should be given to:

- 414 • Metal NOAA, since the potential release of ions may induce local skin effects (e.g.  
415 irritation and CD) and absorption of toxic or sensitizing metals;
- 416 • NOAA with metal catalytic residue, since potential release of ions may induce local skin  
417 effects (e.g. irritation and CD) and absorption of toxic metals;
- 418 • Non-rigid or flexible NOAA, since due to their flexibility liposomes and micelles can  
419 penetrate and permeate the intact skin also at sizes >4 nm;
- 420 • Co-exposure to other toxic substances present in the workplace.

421

422 In the case of “high hazard” NOAA, dissolution of toxic or sensitizing substances in synthetic  
423 sweat should be evaluated under physiological relevant conditions (e.g. at 32°C to mimic the  
424 temperature of the hands). If the NOAA dissolve in synthetic sweat, in addition to continuing  
425 with the assessment, it is advised to also evaluate the level of contamination of surfaces  
426 (benches, tools etc.) in the workplace and to evaluate the internal exposure to these  
427 substances by means of biological monitoring (if available, e.g. As, Cr, Co, Ni in urine) for  
428 exposed workers. Health surveillance of workers potentially exposed to such NOAA is also  
429 advisable.

430

431 2. Evaluation of skin condition of exposed workers

432 As an impaired barrier function is a crucial aspect for NOAA skin penetration and permeation  
433 is import to evaluate this risk factor.

434 Various biophysical measurement methods that reflect the deterioration of barrier function are  
435 available. Routine workplace methods to assess skin integrity must be easy to use by those  
436 who are not dermatologists and sufficiently sensitive and reproducible to detect signs of very  
437 early degradation of skin barrier function, and to identify individuals at risk of increased  
438 uptake of nanoparticles.

439           Assessment of skin condition can be made by visual examination, which may include  
440 questionnaires or scoring systems. For example, the Nordic Occupational Skin Questionnaire  
441 Group has developed the Nordic Occupational Skin Questionnaire (NOSQ-2002) for surveys  
442 on work-related skin disease on the hands and forearms in relation to exposures to  
443 environmental factors (Susitaival et al., 2003).

444           Weistenhofer et al. (2010, 2011) reviewed the skin score tools available for  
445 quantifying hand eczema. Of the many scoring systems, only three have been validated: the  
446 Hand Eczema Severity Index (HECSI), the Manuscore and the Osnabrück Hand Eczema  
447 Severity Index (OHSI). They compared these three systems and concluded that both HECSI  
448 and OHSI were relevant in practice since the risk of observer bias was low. However, in an  
449 occupational setting damage to the skin is typically minimal which makes quantification of  
450 skin condition rather than skin disease difficult.

451 We suggest a modified Hand Eczema Severity Index (HECSI) to determine skin disruption.  
452 The original questionnaire, suggested by Held et al. (2005) was modified considering only  
453 irritative aspects (fissures and scaling) and inserting ‘dryness’ as a clinical sign. Each hand is  
454 divided into five areas (fingertips, fingers (except the tips), palm of hand, back of hands,  
455 wrists. For each of these areas the intensity of the three clinical signs related to impairment of  
456 the skin (fissuring, scaling and dryness) are graded following original scale (1 - mild disease,

457 2 - Moderate, and 3 - Severe). For each locations (total of both hands) the affected area is  
458 given as score from 0 to 4 (0 = 0%, 1 = 1-25%, 2 = 26-50%, 3 = 51-75%, 4 = 76-100%). The  
459 score obtained for the extent of each location is multiplied by the total sum of the intensity of  
460 each clinical feature, and the total sum was calculated as Skin Disruption Score Index,  
461 varying from 0 to 180 (Table S4).

462 There are also a number of biophysical parameters that can be used to objectively  
463 assess skin condition. The most commonly used ones are transepidermal water loss (TEWL)  
464 from the skin surface, skin hydration and quantitative measurement of skin colour.  
465 International guidelines for the in vivo assessment of skin properties in non-clinical settings,  
466 such as the workplace, have been published (duPlessis et al., 2013; Stefaniak et al., 2013) and  
467 cover pH, TEWL and skin hydration.

468 All of these biophysical assessment methods have the advantage that they are non-  
469 invasive, simple to use, provide quantitative data and may indicate sub-clinical damage to the  
470 skin barrier. However, they can be affected by environmental factors such as humidity and  
471 temperature, which may change rapidly. Biophysical measurements of skin barrier could be  
472 used to assess the potential for uptake of nanoparticles through compromised skin, but these  
473 tools are likely only to be useful in research studies or where there is particular concern about  
474 dermal exposure to nanomaterials.

475

### 476 3 Evaluation of jobs at high risk for occupational contact dermatitis (CD)

477 Since skin absorption of NOAA is relevant in condition where skin barrier is disrupted, it is  
478 crucial to evaluate skin barrier integrity in exposed workers. Typical industries where  
479 dermatitis occurs include agriculture, food industry (including catering), chemical industry,  
480 construction, health and electronics (HSE, 2014; Cahill et al, 2012; Pal et al, 2009; Zorba et  
481 al, 2013; Behroozy and Keegel, 2014).

482 Occupations with high rates of dermatitis are hairdressers and barbers, florists, cooks,  
483 beauticians, metal working machine workers, chemical, rubber, glass and ceramic process  
484 workers, dental practitioners, dental and other nurses and podiatrists (HSE, 2014). Other high  
485 risk jobs include cleaners, mechanics and vehicle assemblers (Royal College, 2011). Nano-  
486 enabled products have penetrated extensively most, if not all, of these professions (See  
487 accompanying paper by Brouwer et al 2016) , making assessment of skin integrity essential  
488 for these professions.

489

#### 490 4. Evaluation of job title at high risk of dermatitis with use of NOAA

491 The accompanying paper by Brouwer et al (2016) links job titles with reported high incidence  
492 of skin diseases to reported use of nanomaterials or nano-enabled products or exposure to  
493 NOAA to flag potential high risk job titles with respect to dermal exposure: i.e. .nurses that  
494 can come in contact with nano drugs, dental workers that are using nanocomposites,  
495 hairdressers and beauticians using personal care products containing NOAA, construction  
496 workers using coatings, paints and mortars, cleaners using dirt repellent coating, and  
497 varnishes with NOAA.

498

#### 499 **Conclusions**

500

501 Skin contact with certain nanoparticles and nano-enabled products that may release NOAA  
502 can cause adverse effects in the skin in particular circumstances. Moreover, some NOAA can  
503 release ions that can have local or systemic effects, if they are able to cross the skin barrier  
504 and to arrive into the skin or into blood circulation. For that reason it is necessary to consider  
505 factors that can cause nanoparticles skin penetration and permeation, metal and impurities  
506 released, contact conditions (surface involved, time of contact, sweating, other chemical  
507 enhancers as soaps) and skin conditions. Nanomaterial can be transported and stored in hair

508 follicles from where they can release ions for periods of time. In conditions where skin  
509 barrier is impaired due to fissures or scaling, nanomaterial can pass directly through the  
510 stratum corneum reaching viable epidermis and derma, potentially causing adverse health  
511 effects-both locally and systemic. These concerns are most realistic for nanomaterials that are  
512 made of metal sensitizers or contain such impurities. NOAA made of sensitizer materials  
513 should be labelled for that hazard.

514 NOAA that contain them as impurities above the appropriate concentration limits, as  
515 determined in contact sensitization documents or patch testing recommendations, also should  
516 carry similar notations

517

518 Furthermore, we identify important knowledge gaps that need to be addressed  
519 experimentally, including NOAA dissolution potential, impurities released, the presence of  
520 toxic substances as well as allergic metals released, that must be considered together with  
521 skin condition for exposed workers. More data on metal release from NOAA are urgently  
522 needed for hazard assessment. The systematic stepwise approach presented here and in the  
523 accompanying paper should be linked to observations of the actual occupational use of  
524 nanoparticles and nano-enabled products to help occupational health practitioners in risk  
525 assessment and management.

526 Figure 1: Skin absorption of NOAA considered available knowledge

527 Figure 2: Overview of stepwise approach for assessment of dermal exposure to NOAA

528

529 Figure 3: Schematic overview of primary evaluation based on composition of NOAA and  
530 following steps.

531

532 Conflict of interest statement

533 The authors have no conflict of interests to disclose.

534

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540

541

542 **References**

543 Adachi K., Yamada N., Yoshida Y., Yamamoto O., 2013. Subchronic exposure of titanium  
544 dioxide nanoparticles to hairless rat skin. *Exp. Derm.* 22, 278-283.

545

546 Aitken R.J., Chaudhry M.Q., Boxall A.B.A., Hull M., 2006. Manufacture and use of  
547 nanomaterials: current status in the UK and global trends. *Occup. Med.* 56,300-306.

548

549 Aitken R.J., Creely K.S., Tran C.L., 2004. Nanoparticles: An Occupational Hygiene Review.  
550 HSE Research Report 274. London: HSE Books.

551

552 Aoshima H., Saitoh Y., Ito S., Yamana S., Miwa N., 2009. Safety evaluation of highly  
553 purified fullerenes (hpfs): Based on screening of eye and skin damage. *J. toxicol. sciences*  
554 34,555-562.

555

556 Baroli B., Ennas M.G., Loffredo F., Isola M., Pinna R., Lopez-Quintela A., 2007. Penetration  
557 of metallic nanoparticles in human full-thickness skin. *J. Invest. Dermatol.* 127, 1701–1712.

558

559 BauA, 2008. Risk resulting from skin contact – identification, assessment, measures.

560 [http://www.baua.de/en/Topics-from-A-to-Z/Hazardous-Substances/TRGS/pdf/TRGS-](http://www.baua.de/en/Topics-from-A-to-Z/Hazardous-Substances/TRGS/pdf/TRGS-401.pdf%3F_blob=publicationFile%26v=4)

561 [401.pdf%3F\\_blob=publicationFile%26v=4](http://www.baua.de/en/Topics-from-A-to-Z/Hazardous-Substances/TRGS/pdf/TRGS-401.pdf%3F_blob=publicationFile%26v=4) (accessed 15.03.16).

562

563 Behroozy A., Keegel T.G., 2014. Wet-work Exposure: A Main Risk Factor for Occupational  
564 Hand Dermatitis. *Saf. Health Work* 5,175-80.

565

566 Bello D., Hsieh S.F., Schmidt D., Rogers E.J., 2009. Nanomaterials properties vs. biological  
567 oxidant damage: Implications for toxicity screening and exposure assessment.

568 *Nanotoxicology* 3,249–261.

569

570 Cahill J., Williams J., Matheson M., Palmer A., Burgess J., Dharmage S., Nixon R., 2012.

571 Occupational contact dermatitis: a review of 18 years of data from an occupational

572 dermatology clinic. Report for Safe Work Australia, Australia [www.safeworkaustralia.gov.au](http://www.safeworkaustralia.gov.au)

573 (accessed 15.03.16).

574

575 Carøe T.K., Ebbenhøj N.E., Wulf H.C., Agner T., 2013. Occupational skin cancer may be  
576 underreported. *Dan. Med. J.* 60,A4624.

577

578 CCOHS, <http://www.ccohs.ca/oshanswers/diseases/dermatitis.html> (accessed 12.12.15).

579

580 Chew A.L., Maibach H.I., 2003. Occupational issues of irritant contact dermatitis  
581 *Int Arch Occup Environ Health* 76,339-46.

582

583 Cherrie J.W., Semple S., Christopher Y., Saleem A., Hughson G.W., Philips A., 2006. How  
584 important is inadvertent ingestion of hazardous substances at work? *Ann. Occup. Hyg.* 50,  
585 693-704.

586

587 Christopher Y., Semple S., Hughson G.W., van Tongeren M., Cherrie J.W., 2007. Inadvertant  
588 ingestion exposure in the workplace. HSE Books (Research project R551).

589 Chu M., Wu Q., Wang J., Hou S., Miao Y., Peng J., 2007. In vitro and in vivo transdermal  
590 delivery capacity of quantum dots through mouse skin. *Nanotechnology* 18,455-460.

591

592 Cohen D.M. and Moore M.M., 2007. Occupational Skin Disease. Chapter 38, pg. 617-639.  
593 In: Rom W. and Markowitz SB editors. *Environmental and Occupational Medicine*, 4th  
594 edition. Wolters Kluwer, Lippincot Williams & Wilkins health, New York.

595

596 Crosera M., Bovenzi M., Maina G., Adami G., Zanette C., Florio C., Larese Filon F., 2009.  
597 Nanoparticle dermal absorption and toxicity: A review of the literature. *Int. Arch. Occup.*  
598 *Environ. Health* 82,1043-1055.

599

600 Donaldson K., Aitken R., Tran L., Stone V., Duffin R., Forrest G., 2006. Carbon nanotubes: A  
601 review of their properties in relation to pulmonary toxicology and workplace safety. *Toxicol.*  
602 *Sci.* 92,5–22.

603

604 Eedy D.J., 1996. Carbon-fibre-induced airborne irritant contact dermatitis. *Contact Dermatitis*  
605 35,362-363.

606

607 Egawa M., Hirao T., Takahashi M., 2007. In vivo estimation of stratum corneum thickness  
608 from water concentration profiles obtained with Raman spectroscopy. *Acta Derm. Venereol.*  
609 87,4-8.

610

611 Elsner P., 1994. Irritant dermatitis in the workplace. *Dermatol. Clin.* 12,461–467.

612

613 Ema M., Matsuda A., Kobayashi N., Naya M., Nakanishi J., 2011. Evaluation of dermal and  
614 eye irritation and skin sensitization due to carbon nanotubes. *Reg. toxicol. Pharmacol.*  
615 61,276-281.  
616

617 Ema M., Matsuda A., Kobayashi N., Naya M., Nakanishi J., 2013. Dermal and ocular  
618 irritation and skin sensitization studies of fullerene C60 nanoparticles. *Cutan. Ocul. Toxicol.*  
619 32,128-34.  
620

621 EU-OSHA European Agency for Safety and Health at Work., 2008. Occupational skin  
622 diseases and dermal exposure in the European Union (EU-25): policy and practice overview.  
623 2008. [https://osha.europa.eu/en/node/6875/file\\_view](https://osha.europa.eu/en/node/6875/file_view) (accessed 14.12.2015)  
624

625 [Fluhr J. W.](#), [Dickel H.](#), [Kuss O.](#), [Weyher I.](#), [Diepgen T.L.](#), [Berardesca E.](#), 2002. Impact of  
626 anatomical location on barrier recovery, surface ph and stratum corneum hydration after acute  
627 barrier disruption. *Br. J. Dermat.* 146,770-776.  
628

629 Fluhr JW, Darlenski R, Angelova-Fischer I, Tsankow N, Basketter D. 2008. Skin irritation  
630 and sensitization: mechanisms and new approaches for risk assessment. *Skin Pharmacol*  
631 *Physiol* 21: 124-135.  
632

633 Forte G, Petrucci F, Cristaudo A, Bocca B., 2009. Market survey on toxic metals contained  
634 in tattoo inks. *Science Total Environ.* 407,5997-6002.  
635

636 Gopee N.V., Roberts D.W., Wepp P., Cozart C.R., Sitonen P.H., Laterdresse G.R., 2009.  
637 Quantitative determination of skin penetration of PEG-coated CdSe quantum dots in  
638 dermoabraded but not intact SKH-I hairless mouse skin. *Toxicol. Sci.* 111, 37-48.  
639 Gorman Ng M., Semple S., Cherrie J.W., Christopher Y., Northage C., Tielemans E.,  
640 Veroughstraete V., van Tongeren M., 2012. The relationship between inadvertent ingestion  
641 and dermal exposure pathways: a new integrated conceptual model and a database of dermal  
642 and oral transfer efficiencies. *Ann. Occup. hygiene* 56,1000-1012.  
643

644 Gulson B., McCall M., Korsch M., Gomez L., Casey P., Oytam Y., Taylor A., McCulloch M.,  
645 Trotter J., Kinsley L., Greenoak G., 2010. Small amounts of zinc from zinc oxide particles in  
646 sunscreens applied outdoors are absorbed through human skin. *Toxicol. Sci.* 118,140-149.  
647

648 Guo L., Morris D.G., Liu X., Vaslet C., Hurt R.H., Kane A.B., 2007. Iron bioavailability and  
649 redox activity in diverse carbon nanotube samples. *Chem. Materials* 19,3472–3478.  
650

651 Held E., Skoet R., Johansen J.D., Agner T., 2005. The hand eczema severity index (HECSI):  
652 a scoring system for clinical assessment of hand eczema. A study of inter- and intraobserver  
653 reliability. *Br. J. Dermatol.* 152,302-7.  
654

655 Hogsberg T., Loeschner K., Lof D., Serup J., 2011. Tattoo inks in general usage contain  
656 nanoparticles. *British j. dermatol.* 165,1210-1218.  
657  
658 Hogsberg T., Jacobsen N.R., Clausen P.A., Serup J., 2013a. Black tattoo inks induce reactive  
659 oxygen species production correlating with aggregation of pigment nanoparticles and product  
660 brand but not with the polycyclic aromatic hydrocarbon content. *Exp. Dermatol.* 22,464-469.  
661  
662 Hogsberg T., Hutton Carlsen K., Serup J., 2013b. High prevalence of minor symptoms in  
663 tattoos among a young population tattooed with carbon black and organic pigments. *J.*  
664 *Eur.Acad..Dermat.Venereol. JEADV* 27.846-852.  
665  
666 Holbrook K.A., Odland G.F., 1974. Regional differences in the thickness (cell layers) of the  
667 human stratum corneum: an ultrastructural analysis. *J. Invest. Dermatol.* 62,415-22.  
668  
669 Honnert B. and Gryzebyk M., 2014. Manufactured Nano-objects: An Occupational Survey in  
670 Five Industries in France. *Ann Occup Hyg* 58, 121-135.  
671  
672 HSE, Health and Safety Executive, 2014. Work related skin disease in Great Britain 2014.  
673 <http://www.hse.gov.uk/statistics/causdis/dermatitis/skin.pdf> (accessed 12.12.15).  
674  
675 Hsieh S.F., Bello D., Schmidt D.F., Pal A.K., Rogers E.J., 2012. Biological oxidative damage  
676 by carbon nanotubes: fingerprint or footprint? *Nanotoxicology* 12,61-76.  
677  
678 Hsieh S.F., Bello D., Schmidt D.F., Pal A.K., Stella A., Isaacs J., Rogers E.J., 2013. Mapping  
679 the Biological Oxidative Damage of Engineered Nanomaterials. *Small* 27,1853-65.  
680  
681 Huzaira M., Rius F., Rajadhyaksha M., Anderson R.R., González S., 2001. Topographic  
682 variations in normal skin, as viewed by in vivo reflectance confocal microscopy. *J. Invest.*  
683 *Dermatol.* 116,846-52.  
684  
685 ISO/TR 14294:2011, Workplace atmospheres - Measurement of dermal exposure - Principles  
686 and methods.  
687  
688 Journeay W.S., Goldman RH., 2014. Occupational handling of nickel nanoparticles: a case  
689 report. *Am. J. Ind. Med.* 57,1073-6.  
690  
691 Kezic S., Visser M.J., Verbeek M.M., 2009. Individual susceptibility to occupational contact  
692 dermatitis. *Indust. Health* 47, 469-478.  
693

694 [Kishore A.S.](#), [Surekha P.](#), [Murthy P. B.](#), 2009. Assessment of the dermal and ocular irritation  
695 potential of multi-walled carbon nanotubes by using in vitro and in vivo methods. *Toxicol.*  
696 *Lett.* 191, 268-274.  
697

698 Labouta H. and Schneider M., 2013. Interaction of inorganic nanoparticles with the skin  
699 barrier: current status and critical review. *Nanomedicine* 9,49-54.  
700

701 Lademann J., Patzelt A., Richter H., Antoniou C., Sterry W., Knorr F., 2009. Determination of  
702 the cuticula thickness of human and porcine hairs and their potential influence on the  
703 penetration of nanoparticles into the hair follicles. *J. Biomed. Opt.* 14,021014.  
704

705 Larese Filon F., D'Agostin F., Bovenzi M., Crosera M., Adami G., Romano C., Maina G.,  
706 2009. Human skin penetration of silver nanoparticles through intact and damaged skin.  
707 *Toxicol* 255, 33-37.  
708

709 Larese Filon F., Crosera M., Adami G., Bovenzi M., Rossi F., Maina G., 2011. Human skin  
710 penetration of gold nanoparticles through intact and damaged skin. *Nanotoxicology* 5,493-  
711 501.  
712

713 Larese Filon F., Crosera M., Timeus E., Adami G., Bovenzi M., Ponti J., Maina G., 2013.  
714 Human skin penetration of cobalt nanoparticles through intact and damaged skin. *Toxicol In*  
715 *Vitro* 27,121-7.  
716

717 Larese Filon F., Mauro M., Adami G., Bovenzi M., Crosera M., 2015. Nanoparticles skin  
718 absorption: New aspects for a safety profile evaluation. *Reg. Toxicol. Pharmacol.* 72,310-22.  
719

720 Lee S.E., Choi K.J., Menon G.K., Kim H.J., Choi E.H., Ahn S.K., Lee S.H., 2010.  
721 Penetration pathways induced by low-frequency sonophoresis with physical and chemical  
722 enhancers: iron oxide nanoparticles versus lanthanum nitrates. *J. Invest. Dermatol* 130, 1063–  
723 1072.  
724

725 Liu W., Zhang S., Wang L., Qu C., Zhang C., Hong L., Yuan L., Huang Z., Wang Z., Liu S.,  
726 Jiang G., 2011. CdSe quantum dot (QD)-induced morphological and functional impairments  
727 to liver in mice. *PLoS One* 6,e24406.  
728

729 Mauro M., Crosera C., Bianco C., Adami G., Montini T., Fornasiero P., Bovenzi M., Larese  
730 F., 2015 Human skin penetration of platinum and rhodium nanoparticles through intact and  
731 damaged skin. *J Nanoparticles Res.* 17, 253-262.

732

733 Monteiro-Riviere N. and Larese Filon F., 2012. Nanomaterial interaction with skin in  
734 Adverse effects of engineered nanomaterials. Fadell, Pietroiusti, Shvedova ed. Elsevier  
735 London 185-208.

736

737 Monteiro-Riviere N.A. and Riviere J.E., 2009. Interaction of nanomaterials with skin:  
738 Aspects of absorption and biodistribution. *Nanotoxicology* 3:188-193.

739

740 Oberdörster G., Oberdörster E., Oberdörster J., 2005. Nanotoxicology: An Emerging  
741 Discipline Evolving from Studies of Ultrafine Particles. *Environ. Health. Perspect.* 113,823-  
742 839.

743

744 Otberg N., Richter H., Schaefer H., Blume-Peytavi U., Sterry W., Lademann J., 2004.  
745 Variations of hair follicle size and distribution in different body sites. *J. Invest. Dermatol.*  
746 122,14-9.

747

748 Ovissipour M., Sablani S.S., Rasco B., 2013. Engineered nanoparticle adhesion and removal  
749 from tomato surfaces. *J. Agric. Food Chemistry* 61,10183-10190.

750

751 Pal T.M., de Wilde N.S., van Beurden M.M., Coenraads P.J., Bruynzeel D.P.. 2009.  
752 Notification of occupational skin diseases by dermatologists in the Netherlands. *Occup. Med.*  
753 59, 38-43.

754

755 Park Y.H., Jeong S.H., Yi S.M., Choi B.H., Kim Y.R., Kim I.K., 2011. Analysis for the  
756 potential of polystyrene and TiO<sub>2</sub> nanoparticles to induce skin irritation, phototoxicity, and  
757 sensitization. *Toxicol.in Vitro* 25,1863-1869.

758

759 Plata D.L., Gschwend P.M., Reddy C.M., 2008. Industrially synthesized single-walled carbon  
760 nanotubes: Compositional data for users, environmental risk assessments, and source  
761 apportionment. *Nanotechnology* 19,185706.

762

763 Plessis J.D., Stefaniak A., Eloff F., John S., Agner T., Chou T.C., Nixon R., Steiner M.,  
764 Franken A., Kudla I., Holness L., 2013. International guidelines for the in vivo assessment of  
765 skin properties in non-clinical settings: Part 2. Transepidermal water loss and skin hydration.  
766 *Skin Research Technol.* 19, 265–278.

767

768 Poland C.A., Read S.A.K., Varet J., Carse G., Christensen F.M., Hankin S.M., 2013. Dermal  
769 Absorption of Nanomaterials Part of the "Better control of nano" initiative 2012-2015. The  
770 Danish Environmental Protection Agency.  
771

772 Rancan F., Gao Q., Graf C., Troppens S., Hadam S., Vogt A., 2012. Skin penetration and  
773 cellular uptake of amorphous silica nanoparticles with variable size, surface functionalization  
774 and colloidal stability. *ACS Nano* 8, 6829-6842.  
775

776 Rice R.H. & Mauro T.M., 2008. Toxic responses of the skin. Chapter 19, pg. 741-759. In:  
777 Curtis D. Klaassen editor, Casarett & Doull's Toxicology, The basic science of poison, 7th  
778 edition. 2008. McGraw-Hill Medical Publishing Division, New York.  
779

780 Robertson K., Rees J.L., 2010. Variation in epidermal morphology in human skin at different  
781 body sites as measured by reflectance confocal microscopy. *Acta Derm. Venereol.* 90,368-73.  
782

783 Rouse J.G., Yang J., Ryman-Rasmussen J.P., Barron A.R., Monteiro-Riviere N.A., 2007.  
784 Effects of mechanical flexion on the penetration of fullerene amino acid-derivatized peptide  
785 nanoparticles through skin. *Nano Lett.* 7, 155-160.  
786

787 Royal College of Physicians. 2011. Concise guidance: diagnosis, management and prevention  
788 of contact dermatitis.  
789

790 Ryman-Rasmussen J.P., Riviere J.E., Monteiro-Riviere N.A., 2006. Penetration of intact skin  
791 by quantum dots with diverse physicochemical properties. *Toxicol Sci* 91,159-165.  
792

793 SCCP – Scientific Committee on Consumer Products, 2007. Preliminary opinion on safety of  
794 nanomaterials in cosmetic products. European Commission, Brussels, Belgium.  
795 [http://ec.europa.eu/health/archive/ph\\_risk/committees/04\\_sccp/docs/sccp\\_o\\_123.pdf](http://ec.europa.eu/health/archive/ph_risk/committees/04_sccp/docs/sccp_o_123.pdf) (accessed  
796 12.12.2015).  
797

798 Scheuplein R.J., 1965. Mechanism of percutaneous absorption. I. Routes of penetration and  
799 the influence of solubility. *J. Invest. Dermatol.* 45, 334-46.  
800

801 Scheuplein R.J., 1967. Mechanism of percutaneous absorption. II. Transient diffusion and the  
802 relative importance of various routes of skin penetration. *J. Invest. Dermatol.* 48,79-88.  
803

804 Sonavane G., Tomoda K., Sano A., Ohshima H., Terada H., Makino K., 2008. In vitro  
805 permeation of gold nanoparticles through rat skin and rat intestine: Effect of particle size.  
806 *Colloids Surf. B.* 65,1-10.  
807

808 Stefaniak A.B., Plessis J., John S.M., Eloff F., Agner T., Chou T.C., Nixon R., Steiner M.F.  
809 C., Kudla I., Holness L.D., 2013. International guidelines for the in vivo assessment of skin  
810 properties in non-clinical settings: part 1. pH. *Skin Res.Technol.* 19, 59–68.

811

812 Susitaival P., Flyvholm M.A., Meding B., Kanerva L., Lindberg M., Svesson A., Olafsson  
813 J.H., 2003. Nordic Occupational Skin Questionnaire (NOSQ-2002): a new tool for surveying  
814 occupational skin diseases and exposure. *Contact Dermatitis* 49, 7-76.

815

816 Vermula P.K., Anderson R.R., Karp J.M., 2011. Nanoparticles reduce nickel allergy by  
817 capturing metal ions. *Nat Nanotechnol* 5,291-5.

818

819 Watkinson A.C., Bunge A.L., Hadgraft J., Lane M.E., 2013. Nanoparticles do not penetrate  
820 human skin--a theoretical perspective. *Pharm. Res.* 30,1943-6.

821

822 Weistenhöfer W., Baumeister T., Drexler H., Kütting B., 2011. How to quantify skin  
823 impairment in primary and secondary prevention? HEROS: a proposal of a hand eczema  
824 score for occupational screenings. *Brit. J. Dermatol.* 164, 807–813.

825

826 Weistenhöfer W., Baumeister T., Drexler H., Kütting B., 2010. An overview of skin scores  
827 used for quantifying hand eczema: a critical update according to the criteria of evidence-  
828 based medicine. *Brit. J. Dermatol.* 162, 239-250.

829

830 Wu J., Liu W., Xue C., Zhou S., Lan F., Bi L., 2009. Toxicity and penetration of tio2  
831 nanoparticles in hairless mice and porcine skin after subchronic dermal exposure. *Toxicol.*  
832 *letters* 191,1-8.

833

834 Zorba E., Karpouzis A., Zorba A., Bazas T., Zorbas S., Alexopoulos E., Zorbas I., Louskoukis  
835 K., Konstandinidis T., 2013.Occupational dermatoses by type of work in Greece. *Safety and*  
836 *Health at Work* 4, 142-148.

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Figure 1  
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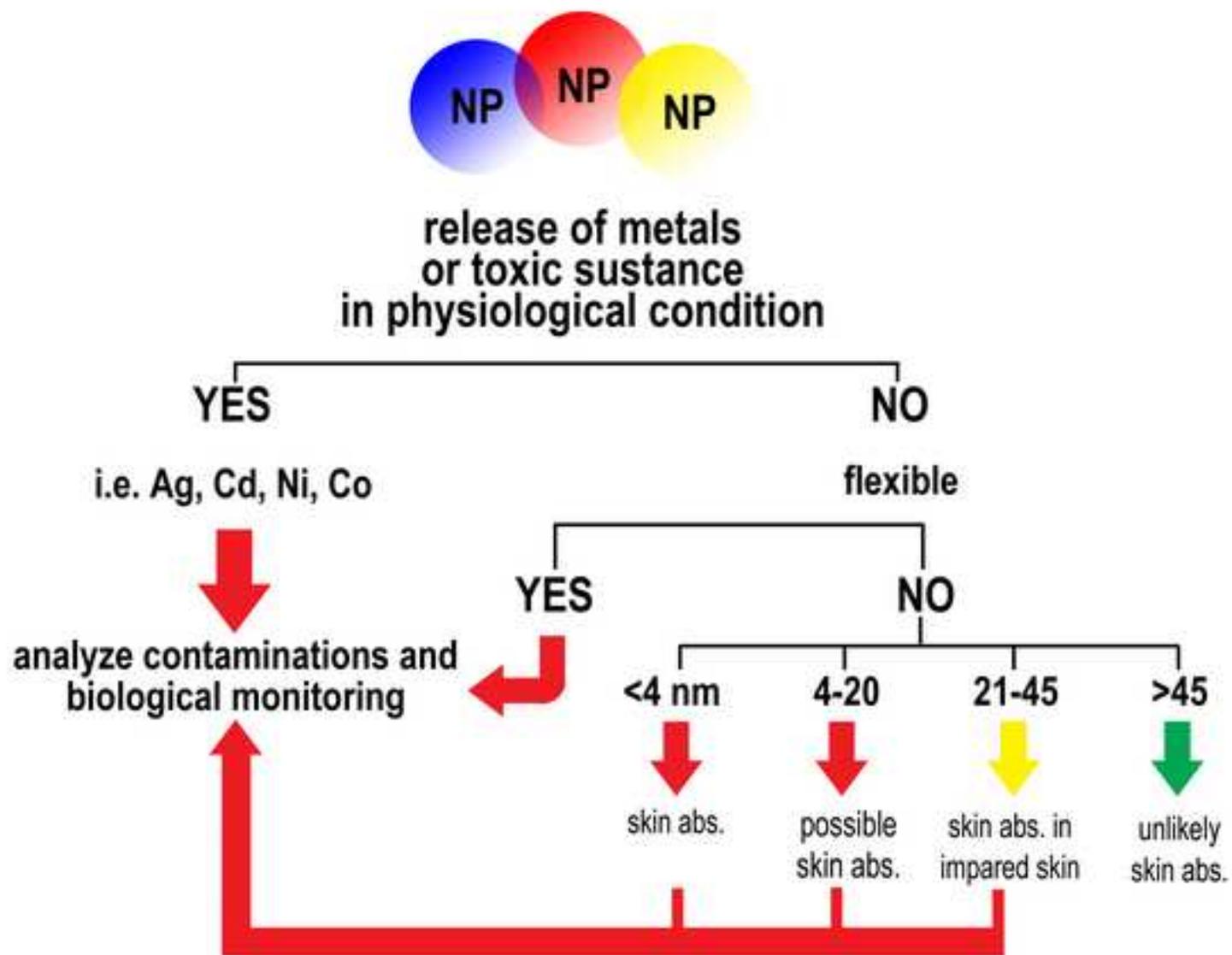
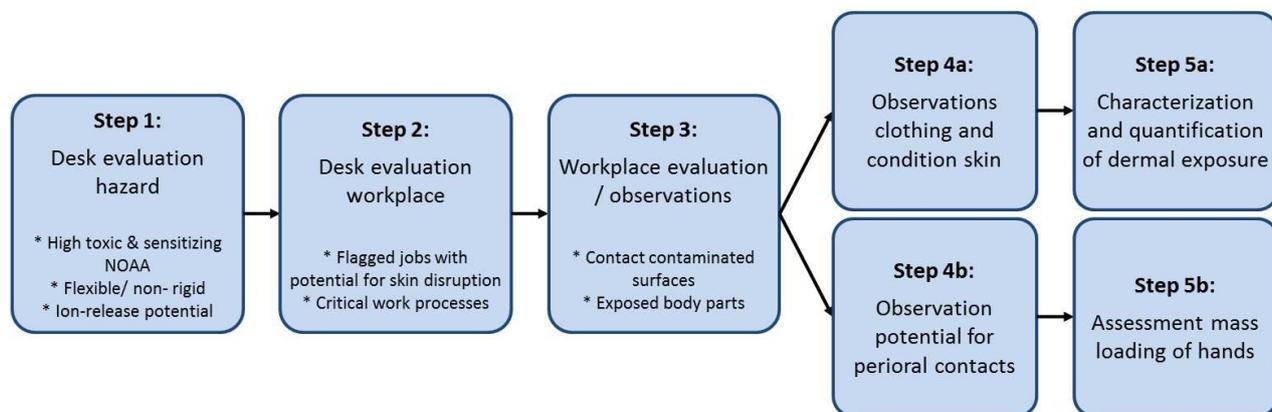


Figure 2: Overview of stepwise approach for assessment of dermal exposure to NOAA



**Figure 3:** Schematic overview of primary evaluation based on composition of NOAA and following steps.

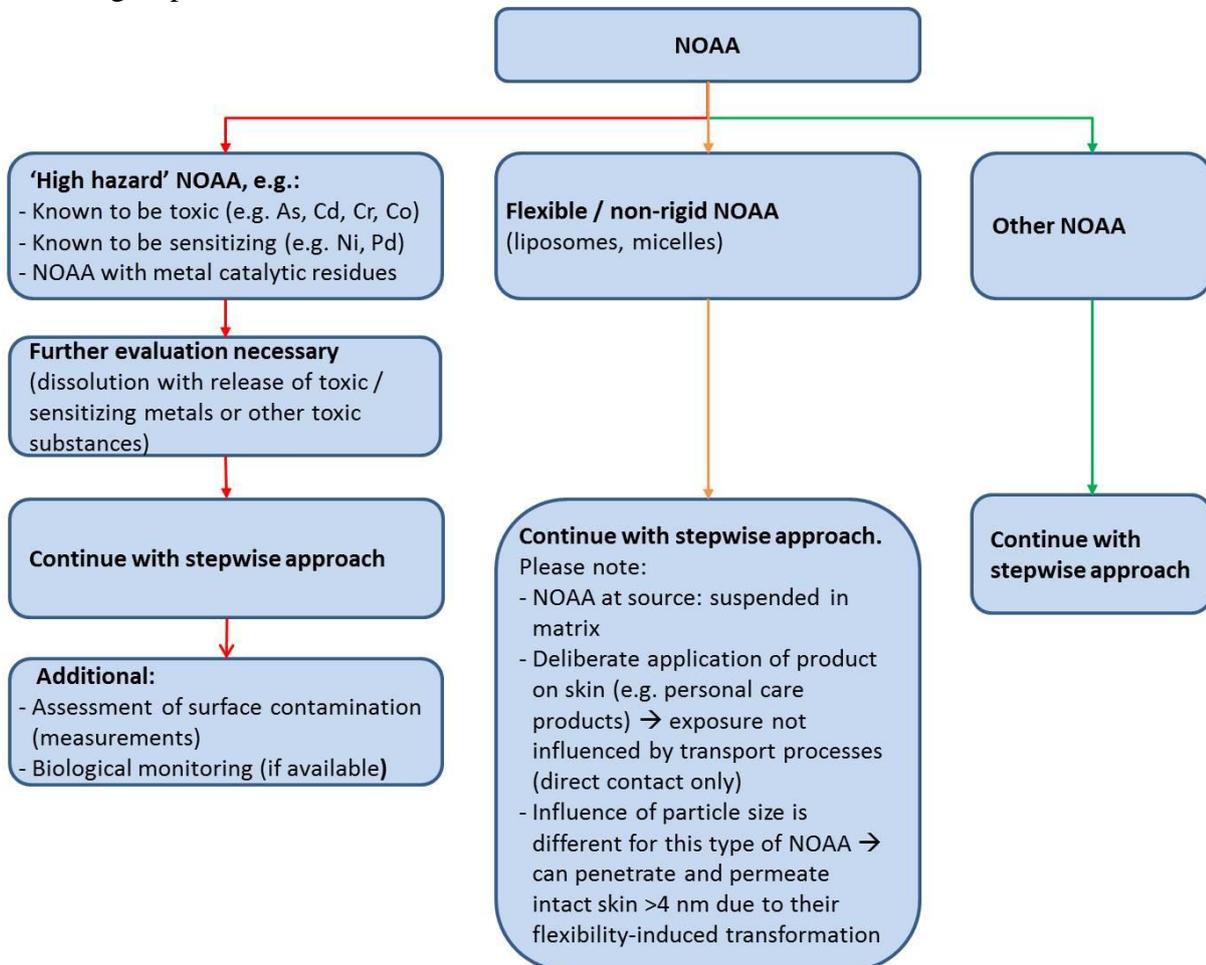


Table 1: Some examples of relevant data on effect and penetration/permeation of NOAA

Nanomaterials	Examples	Critical size (nm)	Comment	Ref.
Carbon nanotubes		Not specified in the paper	Possible only irritation effects	Eedy 1996
Non-metal NPs	Fullerene	3.5	Penetration and permeation in flexures	Rouse 2007
	Silica	42	Penetration and permeation possible in damaged skin through follicles	Rancan 2012
Quantum dots	CdSe	4-12	Penetration and penetration possible and ions release	Chu 2007
Metal-oxides	TiO <sub>2</sub> ZnO	-	No penetration or permeation in vitro. One paper reports systemic absorption in vivo for ZnO containing cream (Gulson 2010)	Labouta 2011 (review)
	Fe <sub>3</sub> O	6-10	Possible permeation with blade incision (10 nm) – Penetration in intact skin (6 nm)	Lee 2010 Baroli 2007
Metal NPs	Fe, Ag, Co, Ni, Pd	12-25	They can release ions so permeation can be related to dissolution. They can cause sensitization (except for Fe)	Baroli 2007, Larese 2009-2015
	Au, Rh, Pt	12	They can't release ions in physiological conditions. Possible penetration.	Sonovane 2008, Larese 2011 Mauro 2015

Table 1S

Table S1. Polycyclic aromatic hydrocarbon (PAH) analysis of 18 diverse ENM using the US EPA Method 3546-microwave extraction coupled with method 8270 GC-MS SVOC analysis. The '< x' symbol indicates that the analyte was not positively identified in the sample, and the amount represents the maximum estimated concentration of particular analyte. All results are expressed in ng/g (ppb). The more abundant analytes (>0.5 µg/g) are highlighted in bold for easier observation. CB, carbon black

Sample <sup>1</sup>	Fluorene	Phenanthrene	Anthracene	Fluoranthene	Pyrene	Benzo (a) anthracene	Chrysene	Benzo (b) fluoanthene	Benzo (k) fluoranthene	Benzo (a) pyrene	Indeno (1,2,3-cd) pyrene	Dibenz (a,h) anthracene	Benzo (ghi) perylene
CB N110	2	21	< 17	4	22	< 17	< 17	< 43	< 17	< 17	< 58	< 72	< 29
CB, N550	< 13	<b>846</b>	<13	<b>834</b>	<b>5659</b>	< 13	< 13	< 33	< 13	< 13	< 44	< 55	46
CB, N990	< 56	323	< 56	<b>1447</b>	<b>4765</b>	119	141	<b>1269</b>	<b>727</b>	<b>5252</b>	<b>13843</b>	< 234	<b>29531</b>
Fullerene, soot	2	17	< 24	< 24	< 24	< 24	< 24	< 60	< 24	< 24	< 80	< 100	< 40
Fullerene, refined	<b>4133</b>	<b>4693</b>	< 34	457	<b>743</b>	49	39	< 86	< 34	5	< 115	< 143	< 57
Fullerene, purified	39	73	< 78	18	46	< 78	< 78	< 195	< 78	< 78	< 260	< 325	< 130
SWCNT_L	95	189	< 37	< 37	< 37	< 37	< 37	< 93	< 37	< 37	< 124	< 155	< 62
SWCNT_S	21	88	< 25	< 25	< 25	< 25	< 25	< 63	< 25	< 25	< 84	< 105	< 42
MWCNT_S	20	59	< 13	< 13	< 13	< 13	< 13	< 32	< 13	< 13	< 43	< 53	< 21
MWCNT_I	61	<b>677</b>	< 33	95	147	< 33	< 33	< 84	< 33	< 33	< 111	< 139	< 56
MWCNT_L	34	136	< 37	19	22	< 38	< 38	< 94	< 38	< 38	< 125	< 156	< 63
SWCNH-ox	17	83	< 37	< 37	< 37	< 37	< 37	< 93	< 37	< 37	< 123	< 154	< 62
nTiO <sub>2</sub> , Anatase	< 48	1	< 48	< 48	< 48	< 48	< 48	< 121	< 48	< 48	< 161	< 202	< 81
nAl <sub>2</sub> O <sub>3</sub>	< 33	14	< 33	< 32	< 33	< 33	< 33	< 81	< 33	< 33	< 108	< 136	< 54

<sup>1</sup>The source and physicochemical properties of this set of ENM has been described in detail in an earlier publication by Bello et al 2009. This set of PAH data has not been published previously. [Bello D, Hsieh SF, Schmidt D, Rogers EJ. 2009. Nanomaterials properties vs. biological oxidant damage: Implications for toxicity screening and exposure assessment. *Nanotoxicology* 3:249–261.] CB, carbon black; N110 (15 nm), N550 (44 nm), N990 (>200 nm). SW- (Single wall) and MW- (multi wall) CNT (carbon nanotubes), L long, S, short; I, industrial grade; nTiO<sub>2</sub>, nano titanium dioxide, nAl<sub>2</sub>O<sub>3</sub>, nano alumina.

Table S2. Organic carbon content for select ENM stratified by ENM type. Note the unit is  $\mu\text{g}/\text{mg}$  (parts per thousand). The OC% represents percent of the total carbon.

ENM Class	N	OC ( $\mu\text{g}/\text{mg}$ )				OC%
		<i>GM</i>	<i>GSD</i>	<i>Min</i>	<i>Max</i>	<i>Range</i>
<b>MWCNT</b>	19	192	2.1	26.4	510	2.2 - 54.3
<b>SWCNT</b>	6	192	1.3	111	258	9.2 - 26.8
<b>Graphenes</b>	5	138	1.3	103	189	10.1 - 32.6
<b>CB</b>	7	62	1.3	38	90	3.4 - 8.4
<b>TiO<sub>2</sub></b>	2	9.0	2.1	5.4	15.1	99.7 - 100
<b>CeO<sub>2</sub></b>	1	6.4	-	6.4	-	100
<b>ZnO</b>	1	3.3	-	-	-	100
<b>ZrO<sub>2</sub></b>	1	1.3	-	-	-	100

Table 3. Summary of total and water-soluble metal impurities for select contact sensitizer and irritants by ENM type. Analysis was conducted by ICP-MC (microwave assisted acid digestion for total metal) and extraction with D.I. water (for 90 min at 37 °C). Units are in ( $\mu\text{g/g}$ , ppm). The minimum is the method limit of detection for each element (routinely in ng/g, ppb). All samples were above the limit of detection for Ni, Cr, Co, Pd, and As (except for ZnO). Zr was not detected in any of the carbonaceous samples.

ENM Class <sup>1</sup>	N	Type	Ni		Cr		Co		Pd		Zr		As	
			GM (GSD)	Max	GM (GSD)	Max	GM (GSD)	Max	GM (GSD)	Max	GM (GSD)	Max	GM (GSD)	Max
MWCNT	21	Total	816 (15)	11757	19 (3.7)	138	67 (14.8)	5656	0.05 (17)	4.8	-	-	0.1 (2.2)	0.7
		WS	1.3 (18)	208	0.04 (6.6)	2	0.2 (8.3)	11	0.001 (5)	0.1	-	-	0.008 (2.5)	0.03
SWCNT	7	Total	80 (1.8)	111	759 (1.6)	1931	2238 (1.3)	3225	0.08 (10)	2.5	-	-	0.18 (2.4)	0.7
		WS	0.6 (6.9)	3.4	0.4 (3.0)	3.1	7.0 (2.8)	28	0.001 (5.7)	0.01	-	-	0.006 (4)	0.03
Graphenes	6	Total	14 (14)	230	6.9 (20.7)	249	5.2 (02.8)	24	0.04 (4.4)	0.8	-	-	0.12 (1.9)	0.4
		WS	0.7 (99)	37.9	0.14 (50)	23	0.08 (27)	3	0.001 (4.1)	0.004	-	-	0.013 (2.5)	0.05
CB	8	Total	2 (1.3)	3	0.7 (2.4)	4.5	0.08 (2.0)	0.3	0.04 (2.8)	0.01	-	-	0.1 (1.8)	0.3
		WS	0.04 (2.2)	0.14	0.005 (2.5)	0.03	0.004 (4.0)	0.03	0.000 (3.1)	0.001	-	-	0.01 (1.8)	0.02
TiO <sub>2</sub>	7	Total	0.7 (3.5)	3	0.3 (10)	4	0.09 (2.3)	0.2	0.06 (4.2)	0.2	21 (8.8)	166	4.3 (6.7)	51
		WS	0.09 (4.2)	0.4	0.7 (3.8)	1.8	0.008 (3.2)	0.04	0.002 (8)	0.01	0.1 (5.9)	3.4	0.6 (94)	14
CeO <sub>2</sub>	2	Total	0.9 (1.1)	1	0.9 (2.3)	1.5	10.04 (1.9)	0.06	0.12 (1.2)	0.15	1664 (8.6)	5866	0.4 (1.3)	0.5
		WS	0.04 (4.3)	0.1	0.02 (1.7)	0.02	0.005 (-)	0.1	0.007 (1.3)	0.01	0.03 (2.0)	0.05	0.03 (1.1)	0.04
ZnO	2	Total	0.4 (35)	5	0.7 (3.8)	1.8	0.04 (5.1)	0.1	0.06 (1.5)	0.1	90 (59)	1600	1.2 (-)	1.2
		WS	0.1 (2.9)	0.2	0.01 (2.1)	0.02	0.03 (2.7)	0.06	0.001 (4.1)	0.04	0.067 (1.1)	0.072	-	-

1. The source, physicochemical properties and other descriptions have been published in a previous publication [Hsieh et al 2013 Small, 27;9(9-10):1853-65. doi: 10.1002/sml.201201995]. MWCNT, multi wall carbon nanotubes; SWCNT, single wall carbon nanotubes; CB, carbon blacks of different primary particle sizes (15 nm to >200nm), TiO<sub>2</sub>, nanoscale titania of different phases and primary sizes. CeO<sub>2</sub>, cerium oxide; ZnO, zinc oxide.

**OCCUPATIONAL DERMAL EXPOSURE TO NANOOBJECTS, AND THEIR  
AGGLOMERATES AND AGGREGATES: Part I - Factors affecting skin absorption**

**Supplemental material**

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Table S4. Scoring of skin condition to obtain skin disruption score index (SDSI), showing a worked example for a worker with severe fissures on the whole back of both hands and moderate scaling and dryness on the whole palm of the right hand (resulting in a SDSI of 20 out of 180).

Clinica signs	Fingertips	Finger (except tips)	Palm of hands	Back of hands	Wristes
Fissures (F)	0	0	0	3	0
Scaling (S)	0	0	2	0	0
Dryness (D)	0	0	2	0	0
SUM (F+S+D)	0	0	4	3	0
Extent (Ex)	0	0	2	4	0
Area Score (AS) (SUM*Ex)	$AS_{\text{fingertips}} = 0$	$AS_{\text{fingers}} = 0$	$AS_{\text{palms}} = 8$	$AS_{\text{backs}} = 12$	$AS_{\text{wristes}} = 0$
Total Disruption Score	$AS_{\text{fingertips}} + AS_{\text{fingers}} + AS_{\text{palms}} + AS_{\text{backs}} + AS_{\text{wristes}} = 0 + 0 + 8 + 12 + 0 = 20$				