

Towards understanding the mechanisms and the kinetics of nanoparticle penetration through protective gloves

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Abstract. Parallel to the increased use of engineered nanoparticles (ENP) in the formulation of commercial products or in medicine, numerous health & safety agencies have recommended the application of the precautionary principle to handle ENP; namely, the recommendation to use protective gloves against chemicals. However, recent studies reveal the penetration of titanium dioxide nanoparticles through nitrile rubber protective gloves in conditions simulating occupational use. This project is designed to understand the links between the penetration of gold nanoparticles (nAu) through nitrile rubber protective gloves and the mechanical and physical behaviour of the elastomer material subjected to conditions simulating occupational use (i.e., mechanical deformations (MD) and sweat). Preliminary analyses show that nAu suspensions penetrate selected glove materials after exposure to prolonged (3 hours) dynamic deformations. Significant morphological changes are observed on the outer surface of the glove sample; namely, the number and the surface of the micropores on the surface increase. Moreover, nitrile rubber protective gloves are also shown to be sensitive to the action of nAu suspension and to the action of the saline solution used to simulate sweat (swelling).

1. Introduction

Engineered nanoparticles, particularly colloidal gold nanoparticles, have attracted enormous scientific and technological interest due to their ease of synthesis, chemical stability, and unique optical properties [1-3]. These characteristics allow the use of nAu for many applications as photocatalysts [4], as Raman sensors [5], in electronics components and in medicine [6, 7]. In the field of bioscience, nAu are used in chemical sensing, biological imaging, drug delivery, and cancer treatment [8-11]. For example, Ducan et al. (2010) have shown how gold nanoparticles can be employed as platforms for drug and biomacromolecule delivery [8]. In 2004, James et al measured a significant enhancement in radiotherapy treatment in mice using gold nanorods [11]. However, a number of studies have also highlighted the toxicity of functionalized or bare nAu [12-15]. In 2009 and 2012, two studies demonstrated the effect of gold nanoparticles on human and mouse spermatogenesis, respectively [16, 17]. Wiwanitkit et al showed that when human semen was placed in contact with a suspension of gold nanoparticles, 25% of the sperm cells became non-motile. Penetration of nAu into the sperm heads and tails was observed. Moreover, a study conducted on rat skin after dermal exposure to 15 nm, 102



nm and 198 nm bare nAu shows that the nAu can be observed in the deep layer of the skin [18]. Transmission electron microscopy (TEM) analysis reveals an accumulation of larger nAu mainly in dermis and epidermis layer, whereas the smaller nAu are observed in a deeper region. Other studies indicated the penetration of ENP through intact or damaged human skin [19-22]. To handle ENP, and particularly nAu, the use of protective gloves against chemicals has been recommended by several health and safety government agencies [23, 24]. However, recent studies have shown the poor efficiency of disposable protective nitrile rubber gloves against titanium dioxide nanoparticles in suspension and in powder form [25, 26]. This paper reports some physicochemical mechanisms which facilitate the penetration of nAu through disposable nitrile rubber gloves. Moreover, it also reports correlations between the penetration of nAu suspensions and the mechanical and physical behaviour of the elastomer material used in conditions simulating occupational use.

2. Materials

2.1. Protective gloves

For this work, two models of disposable 100% nitrile rubber gloves (Nitri-care[®]3005PF and N-Dex[®]6005PF) from Showa Best Glove were selected. Both are non-latex, powder-free and silicon-free. In this paper, the gloves are identified as NBR-1 for Nitri-care[®]3005PF and NBR-2 for N-Dex[®]6005PF, and have a thickness of $73.2 \pm 3.0 \mu\text{m}$ and $117.0 \pm 6.5 \mu\text{m}$, respectively. All samples were taken from the palm section or back of the gloves.

2.2. Nanoparticles

Nanoparticle suspension in water was prepared containing 50 nm gold particles (nAu-50) coated with polyvinylpyrrolidone (Nanocomposix, San Diego, CA). The gold concentration is 0.05 mg/mL (NanoXact grade).

3. Methods

3.1. ENP penetration experimental setup

This project used the same experimental test setup developed by Vinches et al. (Figure 1) [27]. It includes an exposure chamber to expose the outer surface of the glove samples to colloidal suspensions and a sampling chamber to collect the nAu with a sampling solution (saline solution) after their passage through the glove samples. Both chambers are separated by the sample. The test setup is also equipped with a probe (diameter 33.5mm \times length 80mm) used to apply mechanical deformations via an electronically controlled interface. The probe head used corresponds to a conical-spherical geometry which simulates MD produced by a flexing hand [26]. The time profile of the sample MD consists in 50% out-of-plane deformations applied every 10 seconds [28].

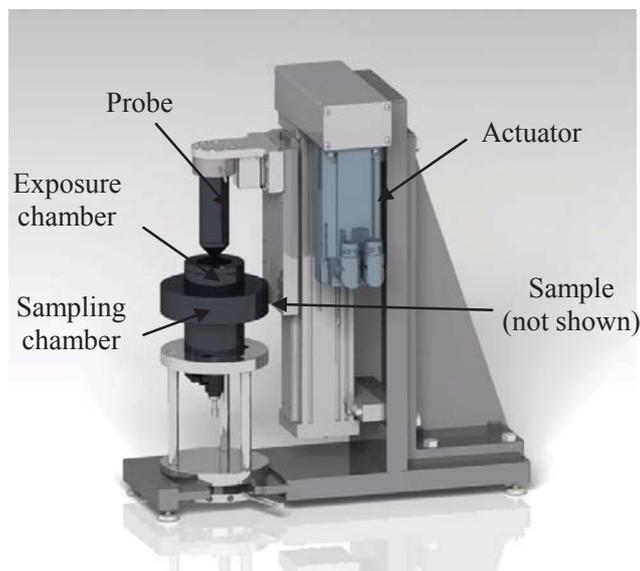


Figure 1. Isometric view of the test setup

3.2. Characterization of the nAu suspension

The size distribution of the nAu was verified by statistical analysis of more than 100 particles imaged by TEM (Hitachi JEM-2100F). The hydrodynamic diameters were also measured by dynamic light scattering (DLS) (Mobius-Wyatt). To obtain statistically significant data, triplicate measurements were performed for all analyses. Thermogravimetric analysis (TGA) (Diamond TGA/DTA Perkin Elmer) was used to evaluate the presence of additives in the liquid carrier. Gradual evaporation of the liquid carrier occurred between 25 and 150 °C at a heating rate of 5°C/min.

3.3. Morphological and chemical characterization of the protective gloves surface

The outer surface morphology of three samples for each nitrile rubber glove was analysed by scanning electron microscopy (SEM) (Hitachi S3600N – $V_{acc} = 15$ kV – magnification $\times 1000$). ImageJ image processing software was used to perform statistical analyses (more than 600 pores for NBR-1 and 700 pores for NBR-2) on the measured diameter and the surface area of the pores [28]. Energy dispersive spectroscopy (EDS) (Oxford INCA X-sight 200) analyses were conducted to determine the presence of gold in the outer surface of both glove materials.

3.4. nAu sampling protocol and detection method

For all the tests, physiological solution was used as the sampling solution. This solution is composed of 0.5 % (w/w) sodium chloride, 0.1 % (w/w) lactic acid and 0.1 % (w/w) urea [29]. The pH of the solution is adjusted to 6 with 1% ammonia solution. The sampling chamber is filled with saline solution up to the level where it makes contact with the inner surface of the glove sample. Before each test, the absence of contamination of the sampling chamber by nAu was verified with a control sample produced by rinsing the sampling chamber with the sampling solution. Gold concentrations were analysed by inductively coupled plasma mass spectrometry (ICP-MS, PerkinElmer NexION 300X). To decrease the sodium chloride concentration, all samples were diluted 10 times in MilliQ water (resistivity 18.2 M Ω ·cm and TOC < 2 μ gC·L⁻¹) before the ICP-MS analysis.

3.5. Swelling measurements

In occupational use, the outer surface of the protective glove is in contact with the ENP and the inner surface with the skin. During the wearing time of the glove, a microclimate is produced inside the glove (sweat, temperature). To evaluate the effect of the nAu suspension and the physiological solution on the material properties, length change measurements were performed on glove samples.

Rectangular samples (5 × 60 mm) were taken from the palm section or the back of the gloves. The samples were immersed in a suspension of nAu in water or in saline solution and at regular intervals, samples were removed from the liquid and their length measured using a calliper (±0.01 mm). The length change data were analyzed using Eq. (1):

$$\Delta L(t) = \frac{L_t - L_0}{L_0} \quad (1)$$

with L_t being the length at t time and L_0 the corresponding value before immersion. To obtain statistically significant data, triplicate measurements were carried out for each type of glove.

4. Results and discussion

4.1. Characterization of the nAu suspension

The size distribution of the nAu-50 was measured by TEM (Figure 2). The particle sizes measured were 47.9 ± 4.7 nm. These results confirmed the manufacturer's data. DLS analysis yielded hydrodynamic diameters for the nAu-50 of 77.7 ± 0.7 nm (Figure 3).

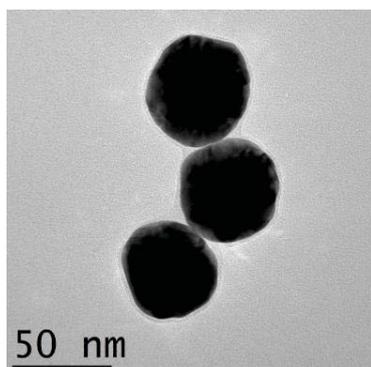


Figure 2. TEM image of the nAu-50 suspension

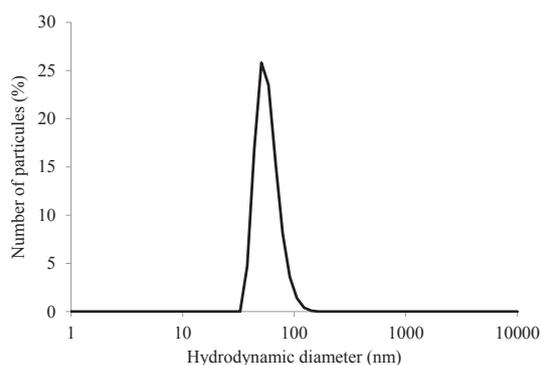


Figure 3. Distribution in number of the hydrodynamic diameter of nAu-50

Finally, thermogravimetric analyses were carried out to verify the presence of additives such as stabilizing agents in the stock colloidal suspension. Figure 4 compares the spectrum obtained with nAu-50 and that obtained with MilliQ water. A change in slope is observed near 80°C (black arrow) on the nAu-50 spectrum corresponding to the presence of one or more additives in the carrier liquid. These additives may play a significant role in the penetration of nAu through the protective gloves. Moreover, the shift towards higher temperatures is due to the presence of the nAu to which the solvent molecules adsorb, thus increasing the vapour pressure [30].

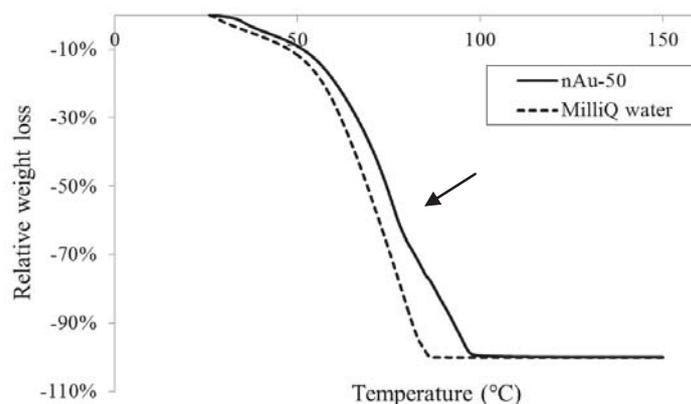


Figure 4. TGA spectra of nAu-50 in water and milliQ water

4.2. Characterization of the protective gloves surface

On the outer glove surface which will be placed in contact with the ENP and the probe, micrometer-size pores can be observed for both native nitrile rubber gloves (Figure 5).

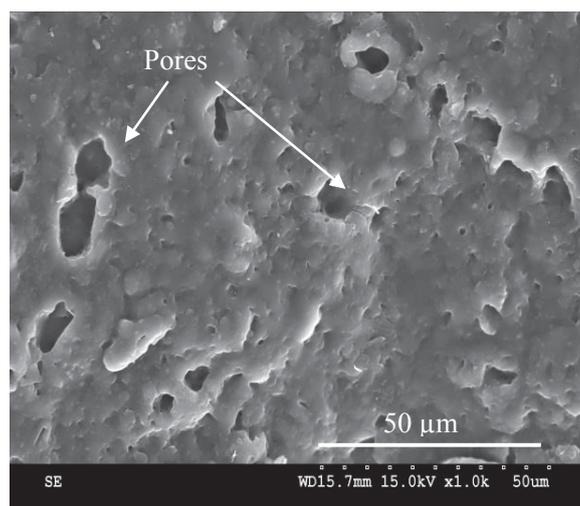


Figure 5. SEM image of the native outer surfaces of NBR-1

Statistical analysis provides three important data: (i) the number of pores as a function of pore diameter (Figure 6), (ii) the mean number of pores per unit area and, (iii) the area occupied by the pores per unit area of the glove (Table 1). Figure 6 displays that the diameter distribution of native NBR-2 pores is centered on 2 μm . In the case of native NBR-1, the distribution is also centered on 2 μm (data not shown). The number of pores per unit area is 17% greater for NBR-2 than for NBR-1. Similarly, the area occupied by the pores is 21% greater for NBR-2 (Table 1).

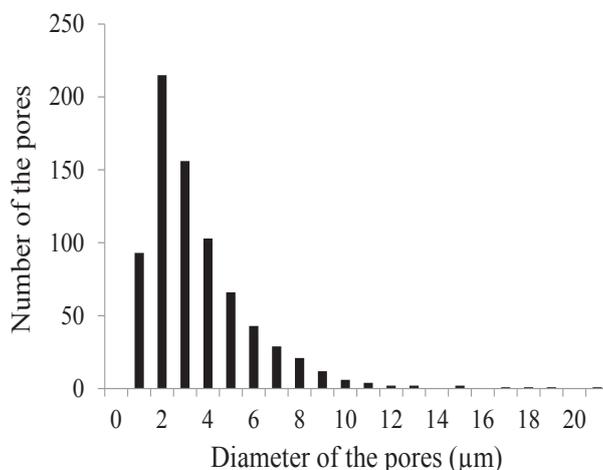


Figure 6. Statistical distribution of pore diameters for native NBR-2

Table 1. Number of pores per unit area and area occupied by the pores of both native gloves

	NBR-1	NBR-2
Number of pores/mm ²	5654	6839
Area occupied by the pores (%)	9.6 ± 2.1	12.1 ± 3.4

The chemical elements present on the outer were determined by EDS. Few differences can be noted between both glove types (data not shown), but neither contains gold.

4.3. nAu penetration: ICP-MS analyses

Glove samples were exposed to the nAu-50 in water while being simultaneously subjected to a 50%-MD every 10 seconds for up to 3h (for a total of 620 MD). The same tests were performed without nAu and in this case, the gold concentration is lower than the limit of detection (LOD) of the ICP-MS (0.048 µg·L⁻¹). For NBR-1, a clear increase in gold concentration in the saline solution can be observed after 3h of 50%-MD as a result of the presence of nAu in the sampling chamber (Figure 7).

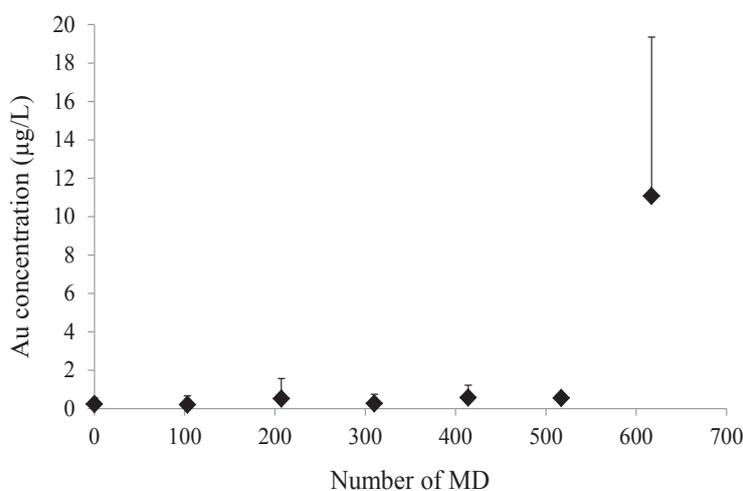


Figure 7. Variation in gold concentration as a function of test duration for NBR-1 samples exposed simultaneously to 50%-MD and nAu-50

When NBR-2 was exposed to nAu-50 in the presence of 50%-MD, no significant gold penetration was measured. This result is probably due to the greater thickness of this protective glove material.

4.4. Behaviour of the protective glove outer surface submitted to MD

Statistical analysis was performed to determine the effects of the MD on the outer surface of both protective nitrile rubber gloves. Firstly, the pore size distribution for NBR-1 native gloves and the same model after 620 50%-MD (3 hours of testing) is compared (Figure 8). After 620 50%-MD, the pore size distribution is centered on 3 μm . This represents a shift of 1 μm compared to the NBR-1 native glove. Thus, mechanical deformations increase the pore diameters but their number by mm^2 and the occupied area are smaller after 3h 50%-MD (Table 2). Interestingly, for NBR-2, all pores are completely missing after exposure to 3h of 50%-MD (Figure 9). This phenomenon is due to abrasion induced by the contact between the probe and the glove sample [28]. For NBR-2, abrasion likely removes the first layer of the glove material.

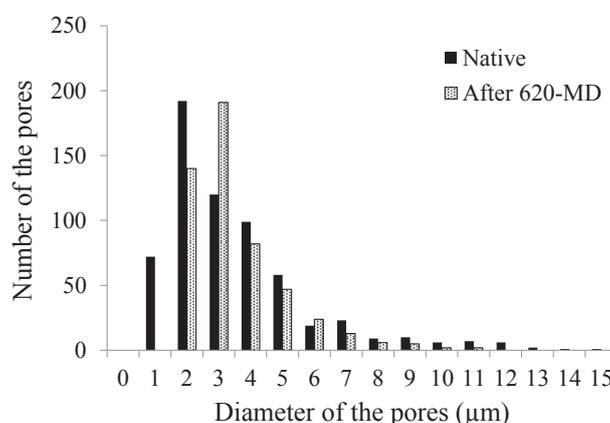


Figure 8. Statistical distribution of pore diameters for native NBR-1 after 620 50%-MD

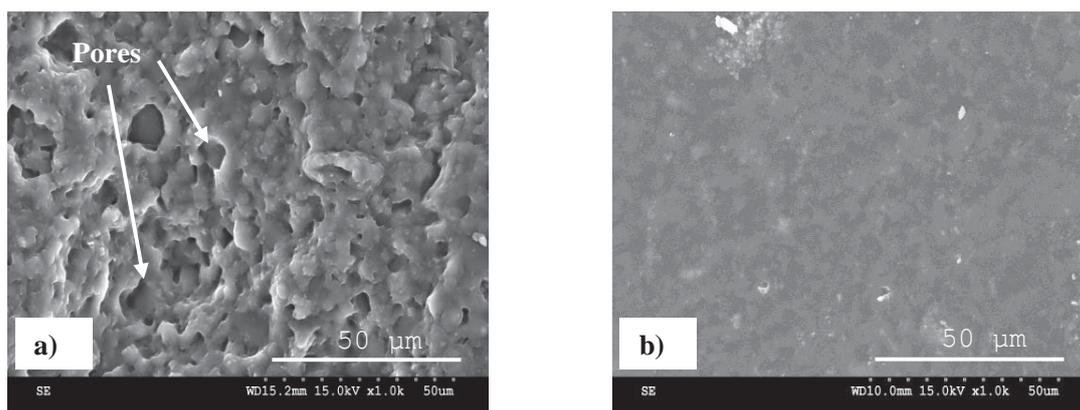


Figure 9. SEM images of the outer surfaces of NBR-2 a) native and b) after 620 50%-MD

Table 2. Number of pores per unit area and area occupied by the pores after 620 50%-MD

	NBR-1	NBR-2
Number of pores/mm^2	4632	N/D
Area occupied by the pores (%)	6.7 ± 1.6	N/D

N/D: Not detectable

4.5. Swelling measurements

Swelling measurements were carried out by recording the length of glove samples after immersion in nAu-50 suspension, in physiological solution or in MilliQ water. Figure 10 compares the length change ratio (LCR) recorded for NBR-1 samples immersed in the three different solutions. For each case, a steady increase in sample length is observed until a plateau is reached. The LCR after 3h of immersion reaches 3.8 % in the three liquids. Thus, the swelling behaviour is the same for an immersion time lower than 3h (maximum duration of penetration tests). However, following longer immersion times, differences are observed in the LCR for the different soak solutions. After 175 hours of immersion, the maximum LCR is similar for nAu suspension and MilliQ water (between 13.6% and 14.6% for MilliQ water and nAu-50, respectively), but it is considerably lower for the saline solution (8.5%). This difference in swelling behavior can be attributed to differences in the solvents' ability to penetrate (diffuse) into the elastomer. According to these results, gold nanoparticles (in nAu suspension) do not have a significant effect on the swelling of the glove material when compared to MilliQ water, the liquid carrier for the nanoparticles. On the other hand, the chemical composition of the physiological solution seems to decrease the swelling phenomenon compared to MilliQ water. Ongoing studies are underway to confirm this hypothesis.

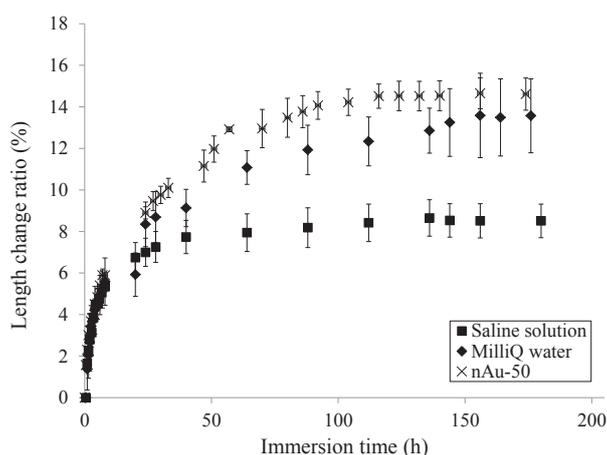


Figure 10. Length change ratio for NBR-1 as a function of immersion time in nAu suspension in water, saline solution or MilliQ water

Table 3 summarizes the LCR after 3h of immersion and the maximum LCR (value reached at the plateau) for both protective nitrile rubbers. Unlike NBR-1, a difference can be observed for LCR after 3 hours of immersion between nAu-50 suspension (2.2%) and the other solutions (3.7% and 3.6% in milliQ water and in saline solution respectively). The maxima of LCR are increasingly important for NBR-2 than NBR-1. These results are consistent with those obtained by Vinches et al. [28].

Table 3. LCR after 3h of immersion and the maximum LCR for NBR-1 and NBR-2 as a function of immersion time in nAu-50 suspension in water, in saline solution and in MilliQ water

	<i>NBR-1</i>		<i>NBR-2</i>	
	LCR after 3 hours (%)	Maximum of LCR (%)	LCR after 3 hours (%)	Maximum of LCR (%)
MilliQ water	3.7 ± 1.0	13.6 ± 1.7	3.7 ± 0.5	19.0 ± 0.8
Saline solution	3.9 ± 0.5	8.5 ± 0.8	3.6 ± 0.2	13.0 ± 0.6

nAu-50 3.8 ± 0.2 14.6 ± 0.7 2.2 ± 0.4 13.0 ± 1.1

5. Conclusion

This study has confirmed the penetration of ENP – in this case nAu – through nitrile protective gloves after repeated deformations simulating occupational use. Nanoparticle penetration has been recorded after 620 50%-MD corresponding to 3h of wearing gloves. Secondly, mechanical and physicochemical phenomena facilitating ENP penetration have been quantified. Significant changes have been observed on the outer surface of the glove material. Indeed, the number and the area occupied by the pores decrease after 3h of 50%-MD and the first layer of glove material were torn from the surface. Moreover, the swelling of the elastomeric membrane has been measured in the presence of nAu-50 as well as in physiological solution simulating the sweat produced inside the glove during their use. To conclude, the disappearance of the first layer of glove materials after repeated deformations, the swelling of elastomeric membrane in contact with the colloidal suspension, exposure to sweat and the measured decrease of the material resistance due to glove use may lead to the weakening of the efficiency of protective gloves used to handle ENP. Ongoing studies are being carried out to further identify the mechanisms facilitating nanoparticle penetration through glove materials.

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