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## **Agglomeration of nanodiamonds during deposition**

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Science in Technology.

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### Tiivistelmä

Nanotimantilla on erinomaisen hyvät mekaaniset-, sähköiset- ja optiset ominaisuudet, joten niiden käyttöä monissa sovelluksissa on yritetty. Nanotimanttien kasautuminen paakuiksi on kuitenkin yleinen ongelma, joka vaikeuttaa nanotimanttien käyttöä esimerkiksi lääketieteen sovelluksissa, lääkeaineiden kuljetuksessa, anturi- ja implanttipinnoitteena sekä nanoelektroniikassa. Tämä diplomityö käy läpi nanotimantteihin ja niiden paakkuuntumiseen liittyvää kirjallisuutta. Nanotimantit muodostavat sitkeitä kasoja, ja yksittäisiä nanotimantteja on vaikeaa saada muodostumaan ilman mekaanista paakkujen hajottamista ennen liuotusta. Kirjallisuuden perusteella voidaan todeta, että tällä hetkellä käytetyimmät menetelmät nanotimanttipaakkujen irrottamiseksi ovat media milling- sekä beads-assisted sonic disintegration (BASD) -tekniikat. Näissä menetelmissä pienet zirkoniumoksidi kuulat liikkuvat liuoksessa ja mekaanisesti hajottavat nanotimanttipaakkuja. Näiden tekniikoiden ongelmaksi muodostuu nanotimanttien saastuminen zirkoniumilla, jota on vaikea poistaa timanteista. Tämä hankaloittaa myös nanotimanttien käyttöä sovelluksissa. Tässä tutkimuksessa tehdyissä laboratoriokokeissa havaittiin, että käytetyistä liuottimista nanotimanttien liuottamiseen parhaiten sopivat etyleeniglykoli ja dimetyylisulfoksidi (DMSO). Nanotimanttipaakkujen koko näissä liuottimissa oli selvästi pienempi kuin muissa yleisesti käytetyissä liuottimissa kuten etanoli ja DI vesi. Tämä sopii hyvin yhteen kirjallisuudesta löytyneen tiedon kanssa.

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**Avainsanat** Nanotimantti, Paakkuuntuminen, Hajottaminen, Bioyhteensopivuus, Liuotin

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### Abstract

As nanodiamond (ND) has excellent mechanical, electrical and optical properties, their use in different biomedical applications is desired. However, nanodiamond agglomeration is a common issue, that is causing problems when trying to use NDs in biomedical applications, carriers in drug delivery, coating for sensor or implant materials or in nano-electronics. This thesis goes through literature related to NDs and especially their agglomeration. NDs form resilient aggregates and single-digit NDs can't be produced without proper deagglomeration before dispersion in solvent. According to literature, it can be stated that most common methods used in mechanical deagglomeration of NDs are media milling and beads-assisted sonic disintegrating (BASD). These techniques use small zirconium oxide balls that break ND agglomerates by colliding with them in media. The problem with these methods is that they cause zirconium contamination in ND particles. Zirconium is difficult to remove from NDs and contamination makes it harder to use NDs in applications. Laboratory experiments performed in this research indicate that ethylene glycol and dimethyl sulfoxide (DMSO) were most suitable solvents for NDs. Size of ND agglomerates were smaller in these solvents than in other commonly used ND solvents tested, such as DI water and ethanol. This observation is in line with information found in ND related literature.

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**Keywords** Nanodiamond, Agglomeration, Biocompatibility, Dispersion, Solvent

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## Preface

*I would like to thank my girlfriend Lotta Pirhonen for supporting me throughout my studies. I also would like to thank my parents Juha and Sirpa, who have been a great inspiration and help for me in my life. In addition, I want to thank the Finnish education policy, that gave me the opportunity to study in university.*

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Mikael Sievänen

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## Symbols and abbreviations

BASD = Beads-assisted sonic disintegrating

CDD-ND = Carbon-dots-decorated nanodiamond

CVD = Chemical vapor deposition

DI = Deionized

DMSO = Dimethyl sulfoxide

DND = Detonation nanodiamond

FTIR = Fourier Transform Infrared spectroscopy

HPHT = High pressure high temperature

ND = Nanodiamond

OLC = Onion like carbon

SEM = Scanning electron microscopy

SYP-ND = Synthetic monocrystalline nanodiamond

TNT = Trinitrotoluene

XANES = X-ray Absorption Near-Edge Structure Spectroscopy

$\zeta$  = Zeta

# 1 Introduction

Nanodiamond is a carbon-based nanomaterial that belongs into the group of nanocarbons currently available in commercial scale (1). These small diamonds have excellent mechanical properties, however, the agglomeration of these particles into clusters (20 nm – 30  $\mu$ m) prevents their use in variety of applications that require single-digit nanodiamonds, such as biomedical applications and drug delivery. (2) (3) (4) Therefore, the potential of this promising material has not yet been fully discovered by many application manufacturers in the field of biomedical technology. (5). Even though agglomeration might be helpful in some ND applications (such as lubricants) but in biomedical applications, agglomeration of NDs is an undesired functionality. Especially when NDs are in interaction with human cells there is a specific need for them to be biocompatible and they should not form unwanted agglomerates in any circumstances.

This Master's thesis studies the reasons and consequences of nanodiamond agglomeration. Goal of the research is to find the most effective methods to minimize ND agglomeration during deposition. Majority of the work is done by studying ND related literature. In addition, there is an experimental section that covers a laboratory study of most typical ND solvents and their agglomeration after deposition on a silicon surface. The focus of literature survey is concentrated on ND agglomeration in solvents and different methods to minimize the size of aggregates. The aim of the experiments is to find out a proper solvent that could be used to deposit a smooth layer of detonation nanodiamonds (DND) on a surface of the sample with minimum agglomeration. Deposition of NDs is done with spin-coating technique. Scanning electron microscopy (SEM) imaging is used to obtain micrographs of surfaces in nanometer scale. Because nanodiamonds are dissolved in different solvents before deposition to a silicon chip, hypothesis is, that there are clear visually observable differences. This thesis tests the best practices available from literature and further validates them.

A single detonation nanodiamond has a very small particle size of 4-6 nm and a high surface area to volume ratio (1). When diamond particles are getting smaller in size the importance of their surface chemistry is increased. The modifications on a ND surface chemistry have a huge impact on the properties of the material. (6) The problem in current ND research is that studies can't be easily compared because of different parameters in ND production. In numerous studies the production parameters of NDs are not clearly presented and therefore comparison to other similar studies is difficult. It must be noted that all commercial nanodiamonds created by a different vendor are a bit different and always related to conditions they were created and purified in. Methods used in synthesis, post-synthesis and modification affect the surface chemistry of nanodiamonds, and therefore, also the applications they can be used in. (1) Because of surface versatility of the ND there is no universal model created that could describe the behavior of all different types of functionalized nanodiamonds. However, the tendency to agglomerate is common to all NDs. Agglomeration can be controlled with the help of surface modifications, fractionation and different deagglomeration methods, that will be discussed later in this thesis. Agglomeration is always closely related to the surface chemistry of NDs and due to this fact, it would be more convenient if all publications made in the field of NDs could clearly inform what type of nanodiamond was used in the study. (7)

In this study the focus is on detonation nanodiamonds because of their price and availability (8). For example, NDs produced with high-pressure, high-temperature (HPHT) method with an average particle size of 25 nm costs approximately \$75/gram. However, well purified polydispersed DND powder costs only about \$1–2/gram (with 200 nm average aggregate size) and suspensions of completely deaggregated 5 nm DND are currently priced around



\$40/gram (9). Another reason for choosing DNDs for this study is their well-known tendency to agglomerate. The DNDs used in experimental section are hydrogen terminated DNDs. ND applications have huge potential in the future, especially in the field of biomedical technology. Many biomedical applications can use nanodiamonds when their biocompatibility is fully tested and approved. However, the use of NDs in biomedical applications requires precise control of particle agglomeration. Biolabeling and bioimaging applications are possible because of NDs rich surface chemistry and fluorescence ability. There are also speculations of using NDs in biosensors, nanoelectromechanical systems (NEMS), tissue engineering and nanomedicine (8). Because of their unique mechanical properties, nanodiamonds are already used as components in lubricants, coatings and composites (10) (11) (12).

## 2 Nanodiamond fabrication

There are currently few major types of NDs commercially available that can be considered to be used in specific applications. These previously mentioned ND types are DND, high-pressure, high-temperature (HPHT) ND and chemical vapor deposition (CVD) ND. They are all created with different production methods, so they have different structures and surface chemistry, and therefore also different applications. Production methods used include detonation of carbon-containing explosives (DNDs), grinding of micron-sized diamond powders manufactured by HPHT synthesis in hydraulic presses or with CVD technique. Major differences between DNDs, HPHT grinded NDs and CVD NDs, are the sizes of primary particles, purity of the material and the state and content of nitrogen impurities in the core of the particles. (13) Advantages of DND include the few nanometer size primary particles and the ability to produce material on an industrial scale in tons quantities annually (14). Besides, due to the difference in the methods of synthesis, the NDs have distinct morphological differences. The shape and size of NDs produced by detonation can be seen in Figure 1. ND particles obtained by processing of HPHT synthetic diamond are available with the smallest average particle size around 10–20 nm. The shape of diamond nanoparticles obtained by milling of natural diamond particles or micron sized HPHT is highly irregular, these particles have spiky and elongated shapes as can be observed from Figure 2. They typically have sharp edges like seen in Figure 2 a) when compared to the basic structure of DND particles that can be seen from Figure 1. Particles are often terminated with {111} facets like in Figure 2 (b). Particles with a high aspect ratio can be found in Figure 2 (c). (13) In general, NDs created with CVD technique are composed of small diamond particles that are ca. 2–5 nm in size and have non-diamond  $sp^2$  carbon at the grain boundaries. The CVD growth is done in argon-rich, hydrogen-poor environment that may contain up to 95–98%  $sp^3$ -bonded carbon. (15)

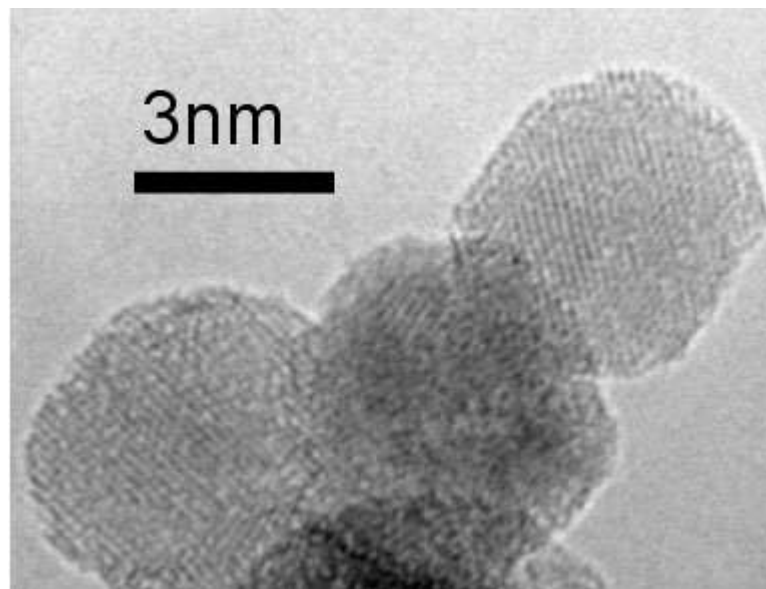


Figure 1, DNDs sized 4-5 nm taken with TEM (16)

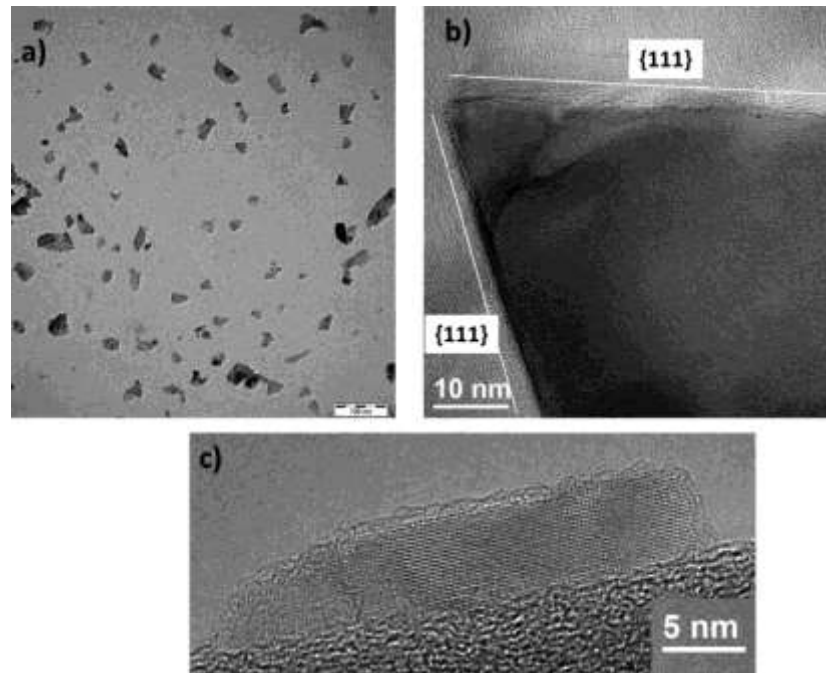


Figure 2, TEM images of diamond nanoparticles obtained by ball milling of micron sized HPHT particles. Residual nondiamond carbon can be seen on the ND particles since it was not purified after milling. (13)

HPHT NDs contain nitrogen as a natural impurity in the form of substitutional nitrogen (Ns) with a typical concentration of 100–200 ppm. ND produced by crashing of natural diamond (Ns concentration up to 3000 ppm) is also possible. (13)

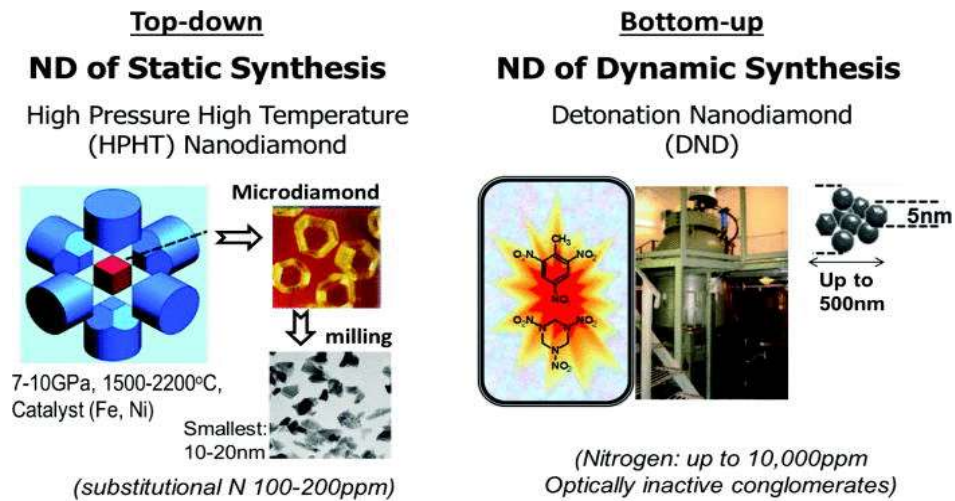


Figure 3, Shapes, sizes and nitrogen content of formed nanodiamonds in a) HPHT synthesis of ND with top-down technique and b) Detonation synthesis of ND with bottom-up technique (13)

## 2.1 DND Synthesis

DND was first synthesized in the Soviet Union in 1960s but its commercial production started in 1980s (1) (17). DNDs can also be called ultradisperse diamonds (UDD) or ultrafine diamonds (UFD) although these names are rarely used anymore. There is no specific maximum size for primary DND particles because they depend on the weight of the explosive charge. Usually most companies produce DND particles with an average size of 3.5 to 6 nm. (1) However, after DND synthesis these particles are agglomerated into agglomerates and need to be separated in post-synthesis or modification stage. These agglomerates have irregular shapes and diameters of  $\sim 100\text{--}300$  nm. (18) This agglomeration is due to the high surface energy of NDs that causes aggressive bonding of atoms on surface. This will be discussed more in chapter 3.

Detonation nanodiamonds are created by controlled explosion of carbon source in a non-oxidizing atmosphere. Explosion happens in a closed detonation chamber made of steel, that is filled with cooling medium. A typical explosive is a mixture of trinitrotoluene (TNT) and hexogen consisting of C, N, O, and H with a negative oxygen balance, so that there is excess carbon in the system. Negative oxygen balance is needed to ensure availability of free non-oxidized carbon for ND formation (13).

There are two major technical requirements for DND synthesis using explosives. First one is that the composition of the explosives needs to provide the thermodynamic conditions for formation of diamonds. This means there must be plenty of needed reagents in the detonation chamber that allow DNDs to form. The second one is that composition of gas atmosphere must provide the necessary thermal conditions to prevent diamond transformation to graphite. This means that the pressure and temperature should be high enough ( $P \geq 10$  GPa,  $T \geq 3000$  K) to enable the formation of NDs but only for a short period of time (19). The power of the explosion also provides the energy needed for the creation of nanodiamond soot. When the explosion takes place, detonation shock wave goes through the chamber. The duration of the detonation shock wave that is propagating in the reaction chamber sets the upper size limit for the growing ND particles. This is the main reason why DND soot consists of nanodiamond particles with diameter of 4-6 nm, some impurities and other allotropes of carbon. The formation of nanodiamond soot happens during these fractions of a microsecond when the pressure and temperature are high enough. After the pressure drops and when there is still elevated temperature in reaction chamber, graphitic soot is formed. (20) The diamond graphitization rate is heavily dependent on the temperature. At a certain temperature, the graphitization rate decreases to a point where the amount of carbon undergoing the transition from the diamond to the graphite phase becomes negligible. Then it is typical to say that the graphitization is 'frozen'. This temperature can also be referred to as the temperature of graphitization or diamond phase freezing. (21) Therefore, the cooling rate of the detonation soot should be no less than  $2727$  °C /min (1). If the pressure has dropped but temperature is still high enough to maintain a high mobility of carbon atoms, the diamond graphite phase transition will be more favorable than if the transition to the region of thermodynamic stability of graphite occurred at  $T < T_D$ . (9) The Debye temperature ( $T_D$ ) of nanodiamond is  $91$  °C, however, the Debye characteristic temperature for a bulk single crystal diamond particle is from  $1527$  °C to  $1969$  °C. This means that the bonding between atoms has been greatly reduced. Also the amplitude where the center for atom shifts the balance of the location is increased 2.4 times. This will lead to an increased activity of the nanodiamond. Thus, its surface is still capable of adsorbing impurities. (22)

Figure 4 shows the schematics of the synthetic detonation ND process. If a dry synthesis is wanted then the cooling medium is usually an inert gas and in the case of wet synthesis, ice

is typically used. The dry synthesis of DND results in smaller primary diamond particles and smaller average agglomerate sizes as compared to wet DND synthesis. (1)

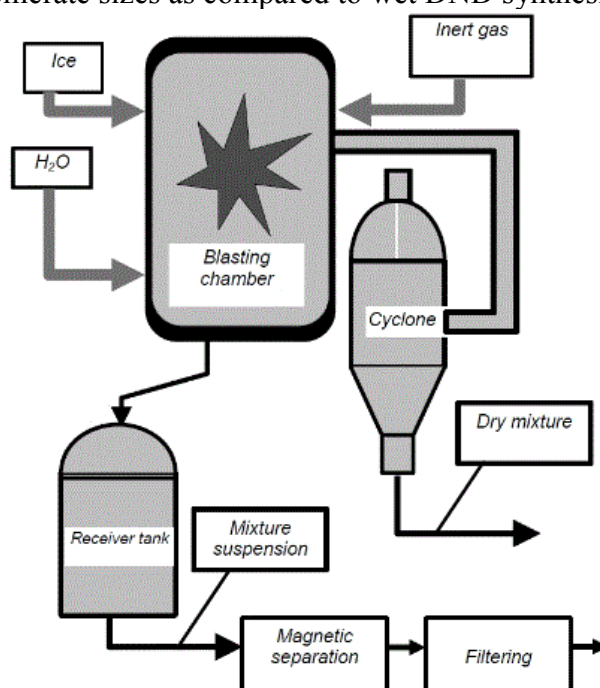


Figure 4, schematics of the detonation synthesis process (14)

There are several aspects that have effect on the yield of the process. These are: the explosion mixture, cooling media, the mass of explosive, the mass of surrounding media and the shape of the explosive charge. (1) (23) With optimal detonation conditions, the soot can contain 75 wt% of nanodiamond particles. Based on x-ray diffraction analysis, the size distribution of nanodiamond crystallites ranges between 2 and 10 nm with a sharp peak at 4–5 nm (13). The size distribution of DND particles is very narrow as the pressure in the reaction chamber remains high enough only for a really short period of time (less than a microsecond to several microseconds) (24). However, due to conditions in the blasting chamber a large-scale agglomeration of NDs happens. These conditions include high pressure and high temperature as already mentioned, and therefore, the formation of cyclohexane molecules (nuclei of the diamond structure). Collisions between these nuclei cause ND aggregation into small clusters. (19) As produced DNDs form tight aggregates of primary particles that can't be separated by sonication. Tight aggregates formed during synthesis differ from many other types of nanoparticles that agglomerate due to electrostatic or van der Waals forces when in solution or during drying from a solution. Electrostatic and van der Waals forces can cause further agglomeration of the ND aggregates, even if the aggregates are already disintegrated (13). Another factor that has a great effect on the aggregation of DNDs during synthesis is the mass of the explosive and the ratio between masses of the charge and wet cooling media used (1). The particles are not only connected by electrostatic interactions but also with covalent bonds between surface functional groups as well as by soot structures surrounding the primary particles. Therefore, it has proven to be a challenging task to produce colloidal systems of primary particles of detonation NDs (24).

## 2.2 Post-synthesis of DND

In order to separate the recently created nanodiamonds from the detonation soot, purification with liquids or gases is required. These processes aim to purify the detonation soot from metallic impurities and non-diamond carbon (1). The purification process is the most complicated and expensive stage of the ND production. (19) The impurities can be located inside the nanodiamond aggregates or attached to their outer surface. Therefore, the nanodiamond agglomerates should be first disintegrated in order to remove the trapped impurities (13). When considering elemental composition, NDs consist of 90-99% of C, H, O, N (25). Most typical ND impurities are Fe, Na, Si and Cu atoms as can be seen from Figure 5.

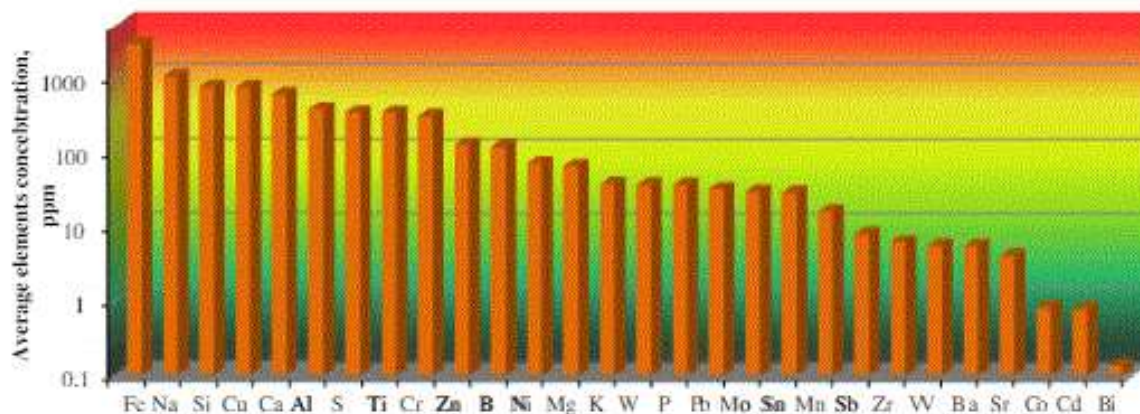


Figure 5, Average common elements content in tested NDs samples. (25)

Most vendors use wet chemistry approaches to purify ND. While these techniques are still very common, they do not always provide sufficient purity of ND. In addition, liquid-phase purification is neither cheap nor environmentally friendly process because it requires expensive corrosion-resistant equipment and costly waste disposal management (23). Usually liquid-phase purification is done by using strong liquid oxidants (such as  $\text{HNO}_3$ , a mixture of  $\text{H}_2\text{SO}_4$  and  $\text{HNO}_3$ ,  $\text{K}_2\text{Cr}_2\text{O}_7$  in  $\text{H}_2\text{SO}_4$ ,  $\text{KOH}/\text{KNO}_3$ ,  $\text{Na}_2\text{O}_2$ ,  $\text{HNO}_3/\text{H}_2\text{O}_2$  under pressure, or  $\text{HClO}_4$ ) at high temperatures and pressures.  $\text{HCl}$  is needed when trying to remove noncarbon impurities to the level of  $<0.5$  wt. %. Alternatively, a more environmentally friendly way to purify NDs is to oxidize non-diamond carbon by heating NDs in air or ozone-enriched air at elevated temperatures. (13) However, these dry chemistry approaches, including different types of oxidation methods, also require the use of either toxic and aggressive substances or supplementary catalysts which result in additional contamination or a significant loss of the diamond phase. (23)

The three major processes when creating a commercial DND product are shown in Figure 6. Currently, there are two distinct stages of alteration of ND surface groups: purification from nondiamond carbon and targeted modification (1). Purification of ND from  $\text{sp}^2$  carbon is a necessary step during production of DND; it results in enrichment of its surface with O-containing groups. After purification process, the ND surface can be additionally modified according to the intended applications. The purification also creates further aggregation, resulting in purified DND with aggregate sizes of up to a few hundred nanometers (13).

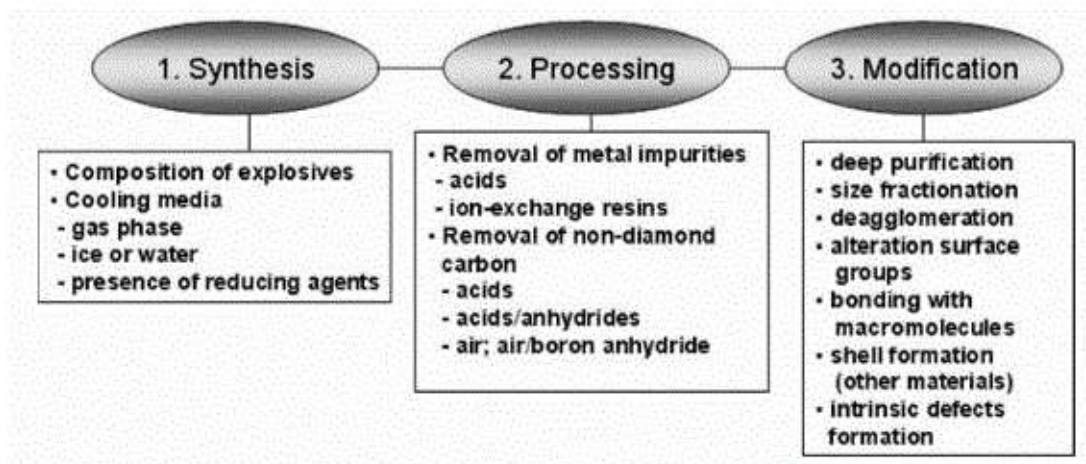


Figure 6, The basic steps of DND production (1)

Well-purified nanodiamond grains can have a close to perfect crystalline structure with almost no fractions of nondiamond carbon. Based on HRTEM observations, detonation ND particles may acquire the shape of polyhedra or a more spherical shape consisting of a diamond core made up of  $sp^3$  carbon. The core may be partially coated by a graphitic shell or amorphous carbon, and dangling bonds on the surface are terminated by functional groups. Particles with more irregular shape but still with well-defined facets are also often observed. (13)

It is important to remember that surface chemistry of nanodiamonds is always affected when in interaction with other material. Therefore, even in the purification process it must be noted that NDs tend to bond with other molecules and agglomerate very easily. The agglomeration behavior of nanodiamond of different origin varies significantly. While nanoparticles received from HPHT or CVD methods show only a moderate tendency towards agglomeration, detonation nanodiamonds usually occur in strongly bound agglomerates if no modifications are performed. (26) In the detonation soot, ND particles are covered with  $sp^2$  carbon shells. It is shown that the shell thickness is thicker when created with dry synthesis. (14) (19) (20) (22). If NDs did not contain amorphous or fullerenic carbon shells on the surface and was free of potentially toxic and undesired metal particles incorporated in these shells, nanodiamonds could be more useful in biomedical applications (23). It has been studied that oxidation removes the  $sp^2$  carbon that bridged the diamond particles into clusters (14) (23). Once  $sp^2$  carbon or other bridges in ND clusters are removed, dispersion of ND to single particles becomes possible. Dispersion of strong chemically bonded agglomerates is a major issue limiting the use of nanodiamonds, and it is determined by our ability to control the surface chemistry of ND particles (23). Figure 7 shows the soot-like structures creating nanodiamonds agglomerates.

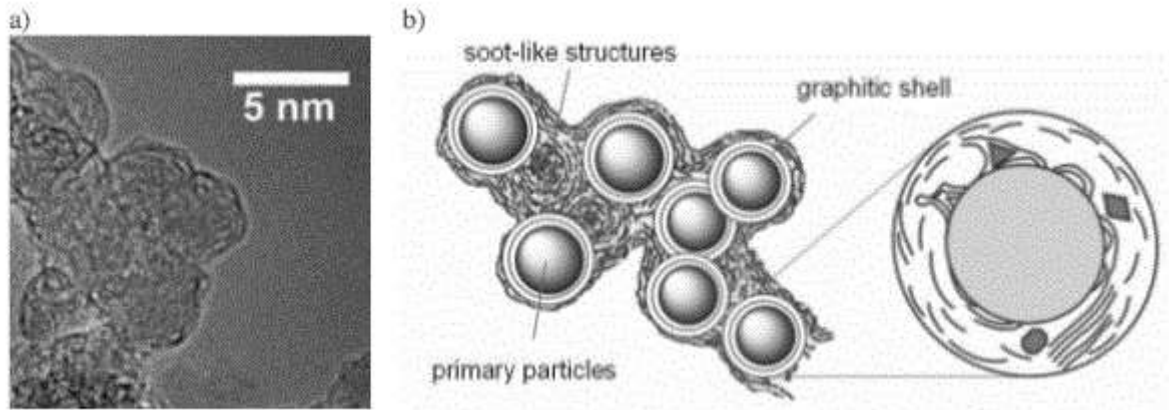


Figure 7, a) HRTEM image of pristine detonation ND. The particles are surrounded by graphitic and soot-like material; b) structure model of the ND agglomerates (24)

### 2.3 Structure

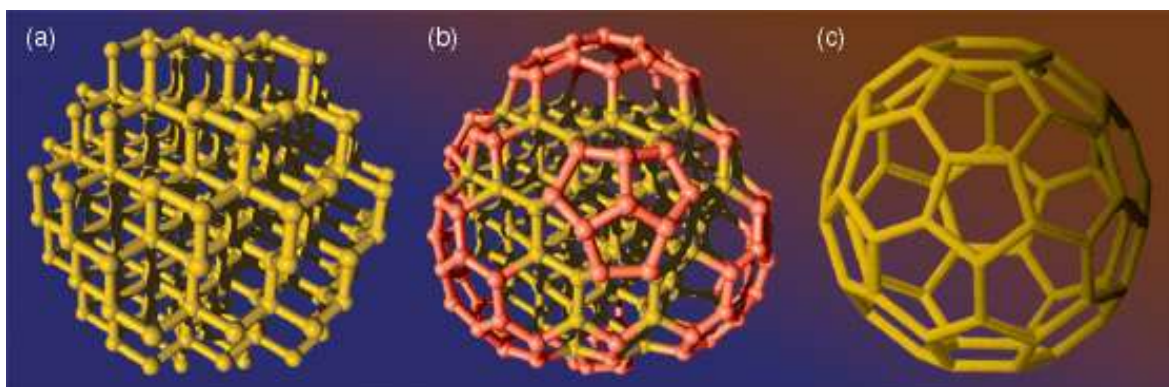
DNDs have a spherical or oval shape and they are non-abrasive so they do not have sharp edges as can be seen from Figure 1. Nanodiamonds have a cubic structure and are expected to have solid lubrication ability (22). The three major low index diamond facets found on the surfaces of natural and synthetic diamonds are (100), (110), and (111), where (111) and (100) facets are the most common (13). The surface of NDs is very versatile and can simultaneously contain hydride, hydroxyl, carboxyl, ketone, ester, lactone, and other similar groups. The existence of different functional groups on the surface of NDs gives it the ability to be modified with desired ions via electrochemical or chemical reduction from aqueous solutions. The functional groups on the surface of nanodiamonds play an important role in the properties of the material. (27) Even though nanodiamonds may have different features because of their surface chemistry, there are still some structural elements all NDs possess:

- 1) A core that is made of  $sp^3$  crystalline diamond with a diameter of approximately 4-6 nm. The core includes 70-90 % of all carbon atoms in ND. (1) (14)
- 2) A 4-10 Å thick carbon shell surrounding the core. The thickness and structure of this shell is affected by the cooling kinetics after DND synthesis. The outer layer can contain 10-30 % of carbon atoms in ND. Currently there are two different models suggested for this carbon shell structure:
  - a) The core is covered with fullerene-type shell of  $sp^2$  carbon called 'Bucky-diamond'. (28)
  - b) Outer layer of unstructured amorphous carbon with mixture of  $sp^2$  and  $sp^3$  hybridized carbon. (14)
- 3) The surface is covered by a large number of different functional groups. These groups are attached because of the highly reactive bonds of the ND surface. The mass of hydrogen, oxygen and nitrogen atoms combined may be as large as 10 – 14 % of the total mass of the particle. Oxygen is usually the main component of the surface groups. (14) (29)

It is well-known that mechanical impact breaks chemical bonds. In the case of carbon materials resulting dangling bonds may form surface  $\pi$ -bonds similar than reconstructed surfaces.



Especially for nanodiamond particles this seems to be reasonable as there are various computational studies on the stability of partially graphitized diamond, the so called bucky-diamond. (24) (28) Bucky diamonds are all carbon core-shell particles that are characterized by a crystalline diamond  $sp^3$ -bonded core, encapsulated by a single- or multi-layer  $sp^2$ -bonded fullerene-like shell that either partially or fully covers the particle surface. The fullerene-like shell is formed in the partial graphitization of diamond during detonation synthesis. (9) Figure 8 demonstrates the basic atomic structure of ND, fullerene and bucky-ball, respectively.



**Figure 8, Structures of a) nanodiamond, b) fullerene and c) bucky-ball**

Nanodiamonds have many differences when compared to macro sized bulk diamond. One major difference is the fact that the transition temperature of diamond into graphite is much lower for ND (something between 900 and 1000 °C), in contrast to bulk diamond for which the diamond–graphite transition temperature is above 3000 °C (30). (13) At room temperatures and pressures, the most stable form of carbon is graphite while diamond is metastable. The energy difference between these two phases is small, only 0.02 eV/atom. The phases are separated by a high-energy barrier ( $\sim 0.4$  eV/atom), so high temperatures and pressures and/or catalysts are needed to convert graphite to diamond. (13) However, diamond can be converted into graphite by irradiation with high energy particles or with a laser. At the nanoscale, the contribution of the surface energy to the Gibbs free energy becomes significant, and the phase equilibrium between graphite and diamond can be reversed. The research of Badziag et al. has shown that, for sizes below 3–6 nm, tetrahedral hydrocarbons are more thermodynamically stable than polyaromatics (31). Basically, that tells us that at the nanoscale, diamond nanoparticles are more stable than nanographite. Graphite-diamond-liquid has a triple point and it is approximately at 12 GPa/5000 K as can be seen from Figure 9 (31) (32) (33).

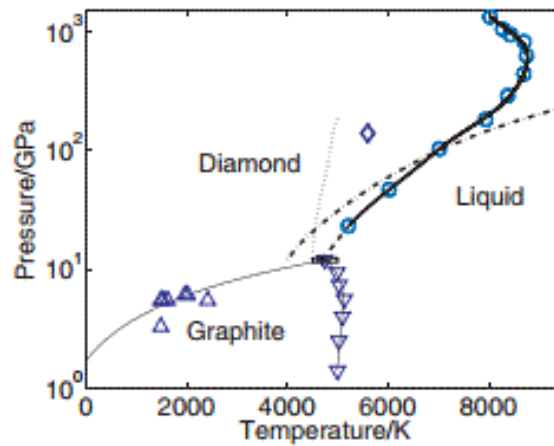


Figure 9, Carbon phase diagram, where Diamond-Graphite-Liquid triple point can be seen. (33)

As mentioned earlier, the chemical potential of the heteroatoms or surface groups depend on the environmental temperature, pressure, and chemical atmosphere (e.g.,  $O_2$ ,  $H_2$ ,  $H_2O$ , etc.). These previously mentioned attributes suggest that the functionalization can be controlled by carefully choosing suitable reaction conditions. The stability of functionalization will be significantly affected by the particle morphology, i.e., the types of facets on the ND surface, where functional groups are attached. Different groups favor different nanodiamond facets depending on the temperature and the environment of the particle, as well as the size of the nanodiamonds. A single model cannot describe all kinds of nanodiamonds. Different models should be used depending on the size, shape, and surface chemistry - parameters controlled by manufacturing and purification methods. (13)

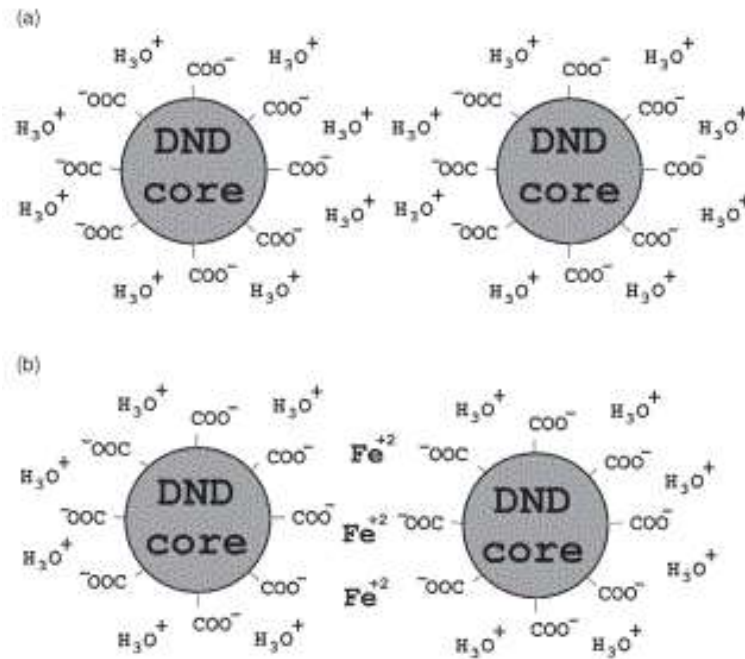
### 3 Agglomeration of DND

Agglomeration of nanodiamonds is the main reason why NDs are not used more in some commercial products yet, e.g. biomedical applications (2) (10). As discussed in chapter 2.1 the size of primary detonation ND particle is from 4 to 6 nm, the primary particles form tightly and loosely bound aggregates because of their high surface energy (14). The aggregation of nanosized particles into submicron and micron formations is actually very common phenomena, the most interesting part with ND agglomeration is the strength of the aggregates. The typical polydispersed DND powder solutions bought from vendor usually contain from 200 to 400 nm average aggregate sizes, which are unbreakable by the traditional ultrasonic treatment as mentioned in chapter 2.1. It was demonstrated by NIU et al. that the raw DNDs are aggregates of primary nanodiamonds connected with amorphous carbon and covalent bonds (34). Detonation nanodiamonds have a highly oxidized hydrophilic surface immediately after fabrication. DNDs also have a large specific surface area (300 to 400 m<sup>2</sup>/gram) and active chemical surface, which can easily cause agglomeration. (1) With recently developed deagglomeration methods such as ball milling and bead-assisted sonication (both introduced in chapter 5), it is possible to investigate the properties of the primary ND particles. As better solvents and better dispersion mechanisms are developed it is possible to find solutions where nanodiamonds do not form clusters. The issues discussed in this chapter include reasons for agglomeration, surface properties, adsorption, surface modification and different types of agglomeration.

#### 3.1 Reasons for ND agglomeration

Nanoparticles have a high surface energy and therefore the outer atoms on surface are more likely to interact with atoms near the surface. Agglomerations is spontaneous for nanodiamonds because particles tend to lower their surface energy. After chemical purification of nanodiamonds, particles about a size of few microns are formed because of agglomeration. (35) Nanodiamond particles in dry powders and suspensions can form more stable primary and less stable secondary aggregates. A. Krueger et al. have proposed a hierarchical model for the different types of ND aggregates, subdividing them into agglomerates (20-30 μm), intermediate aggregates (2-3 μm), and core aggregates (100-200 nm) (4). It should be noted that larger agglomerates and intermediate aggregates are mechanically fragile and can be disintegrated by mechanical, chemical or ultrasonic impact, but the bonds in core aggregates are very strong and cannot be broken up easily by any conventional mechanical, sonication, or surfactant-assisted techniques. (4)

For a long time, it has been considered that there are two types of bonds in the fractal DND agglomerate. The primary DND agglomerates are associated by covalent bonds, and the secondary ones are associated by van der Waals forces. It is also suggested that carboxyl groups (seen in Figure 10a) and iron ions (attached during explosion from the walls in detonation chamber) are responsible for the symmetry of electron wave functions of interactive carbon atoms. It has been estimated by Aleksenskiy, Eydelman and Vul that a single 4 nm DND particle consists of ca 10<sup>4</sup> carbon atoms and several hundred other atoms, such as carboxyl groups, that are placed on the single DND particle surface (2). The covalent bonds can explain the strength of the primary DND agglomerate trough carboxyl groups coupled by ions of metal impurities, as well as by conventional C–C–bonds. Iron increases the agglomerates size and degrades the stability of nanodiamond suspensions as seen in Figure 10b. (2)



**Figure 10, (a) Formation of a double electric layer around a DND particle in dissociation of carboxyl groups, and (b) DND particle coagulation initiated by formation of bridge bonding in the presence of iron ions. (2)**

In a recent study by Chang, Osawa and Barnard, the common theory where ND aggregation is mainly caused by van der Waals forces or by chemical bonds between the individual particles was questioned. It was realized that the ND aggregates strongly resist dispersion and a large driving force is required to disintegrate them, while X-ray diffraction (XRD) measurements show that the DND particles remain discrete (36). A speculation that the  $sp^2$  matrix can bind the core  $sp^3$  particles together was also rejected since in highly purified NDs the  $sp^2$  shell is removed while agglomeration is still observed. The current most reasonable explanation of the agglomerate formation was suggested by Barnard and Sternberg who predicted the self-assembly of the ND particles by electrostatic attraction of the oppositely charged ND surfaces (37). This mechanism has recently been confirmed by aberration-corrected electron microscopy measurements. (36) (38) Different types of ND clusters are classified (by Chang, Osawa and Barnard) in Table 1.

**Table 1, ND aggregate comparison (36)**

Name	Size, nm	Type	Nature	Configuration	Terminology
Primary particle	4.7	-	-	-	Isolated
Agglutinate	c.a. 60	Interfacial	Electrostatic	Ordered	CICI a
Agglomerate	100-200	Interfacial	Electrostatic	Random	IICI b
Powder	> 1000	Intergranular	Van der Waals	Random	VDWA c

a) Coherent interfacial coulombic interaction, b) Incoherent interfacial coulombic interaction, c) Van der Waals aggregation

Besides the van der Waals interaction, DND particles may also be linked by bridge bonding differing in nature. Significantly, it is not only the anhydride groups that can affect the particle aggregation in DND hydrosols. (9) In Figure 11 possible bridge bonding between ND is shown with different types of bridges.

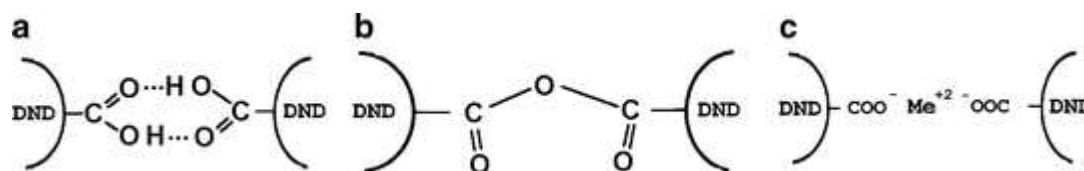


Figure 11, Possible schemes of bridging bonds linking nanodiamond particles. (9)

### 3.1.1 Adsorption

It is still not entirely clear whether total homogeneity of DND surface functional groups can be achieved. However, it is known that agglomeration is highly related to adsorption properties of ND surface. Therefore, the adsorption plays important role when studying the ND aggregation. There is a need to understand mechanisms of adsorption in order to make targeted modifications for increasing or decreasing NDs adsorption efficiency. In many applications, especially in the field of biomedicine, additional study of NDs adsorption properties is very important to solve the problem of elaboration of a multifunctional biosensor on the base of NDs. Such biosensors could be used as sorbent, drug delivery and fluorescent marker simultaneously.

A typical ND particle of 4–5 nanometers in size with a shape close to a sphere has multiple low-index facets, such as (001), (011), and (111) types, as well as high index facets exposed at its surface. The surface energies of these facets are very distinct, involving possible surface reconstructions of (111) and (001) surfaces. For different DND facets, termination with different types of surface groups might be energetically preferable, since this was the case for bulk diamond surfaces (1). At least for bulk diamond the chemisorption of atoms or molecules on the diamond surface lowers the energy on the surface. (39) The general picture of stable structures of diamond surfaces terminated with different types of surface groups is very complex. It is even more complicated for ND particles, where particle size, shape (exposed facets), presence of edges and vertexes all are structural factors influencing the strength of binding of different types of surface groups and the overall stability of a particle (13).

The predicted trend in adsorption energy strength is following:  $\text{S} < \text{Cl} < \text{OH} < \text{H} < \text{F} < \text{O}_{\text{on top}} < \text{O}_{\text{bridge}}$  (39). The C–O bonds are generally more favorable than C–O–H bonds. Oxygen and fluorine atoms tend to form very strong bonds with the carbon atoms on the diamond surfaces. Oxygen can be adsorbed in two positions: in an ether (bridge) position through covalent bonding to two adjacent carbon atoms or in a ketone position (on “top” of a carbon atom). The bridge position for oxygen on the (100) surface was predicted as the most thermodynamically favorable for high oxygen coverage. Lai and Barnard examined the stabilities of the oxygen-, hydrogen-, and hydroxyl-terminated ND particles with respect to temperature variations and different O- and H-rich environments using the density functional tight-binding simulations. (40) (41) Thermostabilities of H-, O-, and OH-passivation of NDs were found to be highly dependent of the surrounding chemical environments, as well as the ND morphology. When there is plenty of H and O around, func-

tionalization with OH groups was generally the most stable due to the formation of a hydrogen-bonding network between OH groups on the surface. This result is opposite to the results for bulk diamond termination, where coverage with O groups is the most stable. (13) Lai and Barnard also investigated in a different study the adsorption strength of a single adsorbate of COOH, OH, O, and H placed in different points on the surface of a particle. It was concluded that adsorption of hydroxyl groups is more energetically preferable than carboxyl groups at almost every point on the surface. In contrast, the adsorption strength of O and H species is much weaker than for COOH groups, and is unstable with respect to gaseous O<sub>2</sub> or H<sub>2</sub> on most of the (111) surface sites. (13) (40)

Ion adsorption properties of untreated and surface modified ND in different aqueous solutions were evaluated recently by Laptinskiy et al. (42) Adsorption efficiency of ND in the study was determined by the number of ions adsorbed on the surface unit area of ND. In Figure 12 the adsorption rate of different ions on I6 nanodiamond, I6COOH surface modified ND and charcoal are compared. In comparison with charcoal, NDs adsorption efficiency didn't show essential advantage. (42)

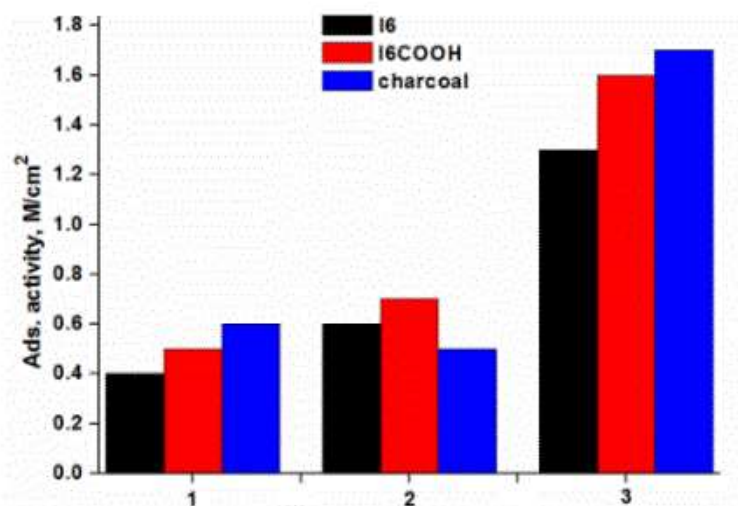


Figure 12, Adsorption activity of I6, I6COOH and charcoal: 1) Cu<sup>2+</sup> in Cu(CH<sub>3</sub>COO)<sub>2</sub>, 2) CH<sub>3</sub>COO – in Cu(CH<sub>3</sub>COO)<sub>2</sub>, 3) CH<sub>3</sub>COO – in Pb(CH<sub>3</sub>COO)<sub>2</sub> (42)

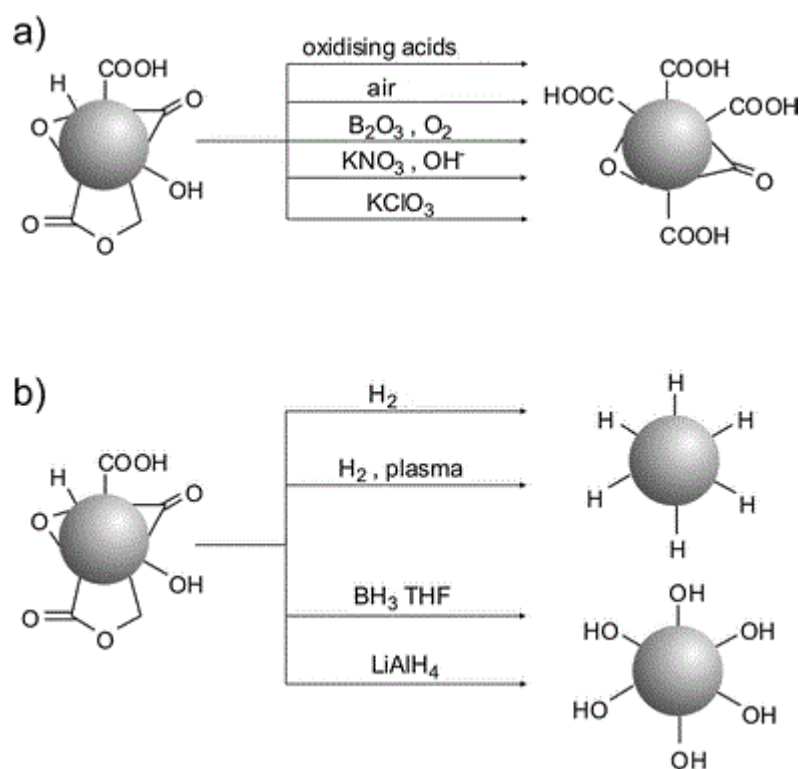
However, it was shown that Pb<sup>2+</sup> ions are adsorbed by the modified ND better than Cu<sup>2+</sup> yielding four times higher sorption efficiency of I6COOH for Pb<sup>2+</sup>. As both NDs have many oxygen containing surface functional groups, both types of nanodiamonds actively adsorb copper on their surface by hydrogen bonding. I6COOH, that has had its surface modified with oxygen containing groups, adsorbs copper more actively in comparison to normal I6 diamond. In nitrate salts solution, the adsorption efficiency of modified ND is about three times higher than that of I6 in respect to the ions. (42)

## 3.2 Surface modification

An important goal in nanodiamond research is developing more efficient methods to purify NDs from detonation soot. As mentioned earlier the detonation soot is basically a mixture of  $sp^2$  and  $sp^3$  carbon phases and some inorganic impurities (9). After purification, the content of  $sp^2$  carbon in ND should be low and the size of agglomerates small. In addition, this should be done in inexpensive, environmentally friendly way (43). As discussed earlier in chapter 2 the commercially available nanodiamonds might require additional processing or modification before they can be used as intended. This is usually because of impurities in material, amount of non-carbon is too high, average agglomerate size is too big, or surface chemistry is not suitable for planned application (1).

As mentioned earlier in chapter 2.2, different DND treatments, usually called modifications, can include additional deep purification, surface functionalization for a specific application, and deagglomeration or size fractionation. (1) The possibility of surface modification creates a lot of opportunities when considering the use of nanodiamonds. Nanodiamonds surface chemistry can be optimized to work in a certain specific application. Surface groups on ND particles play another very important role—they stabilize the diamond structure toward surface reconstructions and prevent its graphitization (13). When starting the investigation of possible ND surface modifications, there are three important things that should be inspected: the average size of DND particles and size distribution of ND clusters, the degree of chemical purification of diamond from the graphite phase - the so called  $sp^2/sp^3$  ratio, and the type of impurities on the surface of DND agglomerates (9). Useful tools for these studies are X-ray diffractometry and small-angle X-ray scattering, Raman and IR spectroscopy, high resolution transmission electron microscopy and scanning electron microscopy, energy-dispersive X-ray spectroscopy, nuclear magnetic resonance and electron spin resonance. (9)

One advantage of NDs is the ability to selectively modify their surface oxygen functional groups. This can be done by using non-covalent attachment or organic synthetic coupling chemistry. (44) Surface of ND is often functionalized with chemical reactions based on radicals, producing carboxylic acid,  $NO_2$  and fluorine surface groups (45). The majority of diamond particles have carboxylic acid groups on their surface when measured with Fourier Transform Infrared spectroscopy (FTIR). These acid groups are produced by air or ozone purification or treatment in oxidizing acids (13). (46) Modification with gases such as  $H_2$ ,  $O_2$  or air are very popular ways to alter the surface of ND. DND powders surface modification by heating in air or in a hydrogen results in oxygenated and hydrogenated form of the nanodiamond surface. In Figure 13 it is shown how ND surface can be homogenized with oxidative or reductive methods by using either gases or liquids.



**Figure 13, The surface of nanodiamond can be homogenized using (a) oxidative or (b) reductive methods. (14)**

Compared with gas treatment, wet chemistry does not require elevated temperatures and provides better selectivity because of a large number of functional group modifications already known in organic chemistry. Additionally, NDs can be modified by using the chemistry of graphitic carbon. Surface graphitization can be created by annealing the NDs in vacuum. Powerful C–C bonds can be created between the graphitic shell and the groups on the surface of diamonds. ND functional groups such as C–X bonds (where X is N, O, or S) are usually produced with techniques that rely on chemistry. There are many kinds of different options for DND surface functionalization, but still the outcome is strongly dependent on the purity and uniformity of the starting nanodiamonds. (13)

Surface properties of NDs have also important ecological implications since use and disposal of nanomaterials will lead to their release into the environment. Upon entry into the environment, depending on their surface properties, nanoparticles can form associates with other ionic or molecular chemical species in the surrounding media, aggregate, adsorb organic matter, and be accumulated at high concentration within the waste material. (13) So as can be observed, there are numerous aspects in the ND surface chemistry that need to be considered when planning ND applications. All the different methods mentioned in this chapter are aiming for the same goal: to find a cheap and easy way of minimizing the agglomeration of NDs and providing a surface chemistry with certain desired qualities. Impurities causing undesired effects and agglomeration should be removed so that nanodiamond particles are suitable for the application they were planned for.



### 3.2.1 Oxidation

Most of the currently available nanodiamond particles have an oxygenated surface (23) (26). The reason for this is the use of water or ice for the cooling in the detonation process, which results in the reaction with active hydroxyl molecules. In addition, the purification of ND is typically performed by using oxidizing mineral acids and/or air oxidation. Both treatments lead to the formation of carbonyl and carboxyl compounds on the nanodiamond surface (26). Liquid phase oxidation enjoys the widest range of applications (9). However, the air oxidation process does not require the use of toxic or aggressive chemicals, catalysts, or inhibitors (23).

Table 2 compares different oxidation methods. The important factors are the amount of non-diamond carbon (NDC) and the content of inorganic impurities. The simplest liquid approach is purification with chromic anhydride or chromates solution in concentrated sulfuric acid. This is because reaction can be done with conventional glass equipment at atmospheric pressure and at temperatures of 120–150°C (14). Oxygen of the air is the simplest and most widely used gaseous oxidizer. The temperature of the oxidation process can be lowered by adding an appropriate catalyst. The biggest advantage of this purification process is its simplicity. On the other hand, the control of oxidation is anything but a simple procedure, and the technological conditions of the process are not the same for different components of the product. The process also has a poor selectivity, because heating above 400°C results in partial oxidation of the diamond yield as well. Besides, oxidation does not remove any inorganic contaminants, and their content may become even enhanced by the presence of the catalyst. As a result, air (or any other gas) oxidation requires involving an additional stage of acid processing to remove the soluble inorganic contaminants. (9)

**Table 2, Comparison of various chemical methods of DND isolation from detonation soot and its purification. (9)**

Method of purification	Oxidizer, conditions	Degree of purification; residual content of NDC	Content of inorganic impurities
Air, catalytic oxidation	O <sub>2</sub> + catalyst; 200-300 °C	Average; 1-2%	5-0% without additional washing in acids
Air, high-temperature oxidation	O <sub>2</sub> ; 420 °C	No information	5-10%, washing of starting material with HF/HNO <sub>3</sub> mixture
Ozone oxidation	O <sub>3</sub> ; 20 °C	Very high level; <0.1%	5-10% without additional washing in acids
Chromic oxidation	CrO <sub>3</sub> in H <sub>2</sub> SO <sub>4</sub> medium; 100 °C	High; <0.2%	1-3%
Oxidation by perchloric acid	HClO <sub>4</sub> in water medium; 50-70%, 100 °C	Average; 0.5-1.0%	0.5-1.0%
Oxidation by a mixture of sulfuric and nitric acids	HNO <sub>3</sub> in H <sub>2</sub> SO <sub>4</sub> medium; 200-250 °C	Average; 0.5-2.0%	0.5-1.0%
Oxidation by nitric acid	HNO <sub>3</sub> in water medium; 50-65%; 200-250 °C	Average; 0.5-2.0%	0.5-1.0%
Oxidation by hydrogen peroxide	HNO <sub>3</sub> + H <sub>2</sub> O <sub>2</sub> in water medium	Average; 0.5-2.0%	0.5-1.0%

The initial oxidation temperature of the nanodiamond in the air is about 550 °C, which is lower than that of the bulk diamond (800°C) (22). The lower oxidation temperature is a result of smaller particle size of the nanodiamond, larger surface and the non-integrity of the crystal structure. In a study by Osswald et al. the optimal temperature range for oxidation of the ND samples was estimated to be 400–430 °C (23). In that study, it was also found that depending on the ND sample, 5-h oxidation at 425 °C increased the content of sp<sup>3</sup>-bonded carbon from 23 to 81% in starting powders to 94–96%, as determined by XANES (X-ray Absorption Near-Edge Structure Spectroscopy). This proves that oxidation in air can selectively remove amorphous and graphitic sp<sup>2</sup>-bonded carbon from nanodiamond powders. When the temperatures are higher than 760 °C, the nanodiamond is oxidized completely. (23) (22) Full decomposition of ND surface groups takes place at ~900 °C and full desorption of H occurs at ~1100 °C. Above this temperature, a few fully encircled graphitic shells are formed on the surface of large ND particle (>2 nm) followed by full transformation of detonation ND particles into OLC at temperature exceeding ~1500 °C (during 1–2 h of annealing in high vacuum). (13)

A particularly strong hydrogen-bonding interaction between oxidized ND and water has been identified, leading to the formation of a surface-confined nanophase of water. This layer

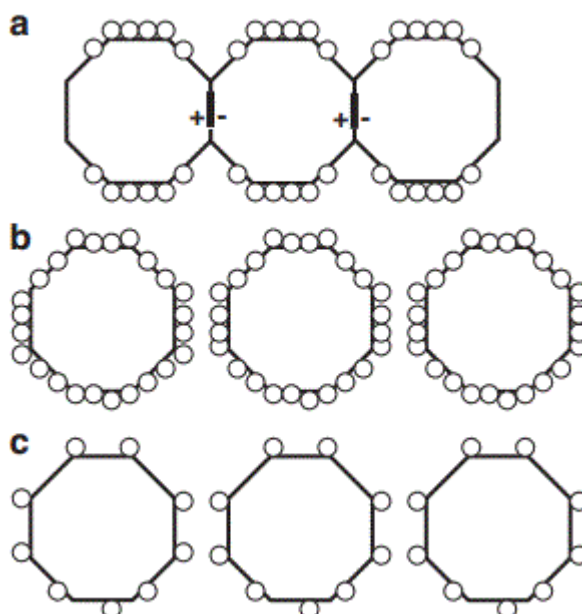
of surface-bound water is estimated to be 0.5 nm thick and requires persistent heating at temperatures of more than 200 °C to be removed. (44)

### 3.2.1.1 Ozone treatment

In studies by Shenderova et al., it was observed that NDs treated with ozone have the lowest amount of  $sp^2$  carbon, in other words, the content of its NDC is very low. The oxidizer here is ozone produced in corona discharge. This method of oxidation is performed after removing non-organic impurities from material and it stands out in its selectivity (9). Gas-phase purification used in ozone modification is an attractive alternative to the more commonly used method of detonation soot treatment with concentrated acids or other strong liquid oxidizers, which make up to 40% of the product cost. Ozone-modified nanodiamonds (NDO) were found to have a number of distinctive characteristics. The ozonated DND is composed of the polyhedrally shaped faceted primary particles, has its surface enriched with oxygen containing groups, forms stable hydrosols and shows constant zeta potential over a wide pH range (47). NDO-based suspensions possess high colloidal stability in the range of pH 2-12, exceeding a pH range of stability for other types of NDs produced from carbon-containing explosives (48). The average size of polydispersed NDO in their water suspensions is about 160-180 nm. This number is the smallest average size reported for commercially available, unfractionated, polydispersed DNDs. In addition, the content of the primary polyhedral-shaped, faceted particles with a size of 3-5 nm in the polydispersed NDO is substantially higher than that of the acid purified DNDs. This is due to the fact that, after ozone purification, necks between primary particles within aggregates are significantly etched and thinned. Then during active dispersion like sonication, aggregates can break apart resulting in large fraction of primary particles and small size of aggregates in the final material. (43) (48) However, as there are many pros in the process, there are also some downsides. These include the technically advanced equipment required, high power consumption and as a consequence, the high cost of ozone-purified DND (9).

### 3.2.2 Hydrogenation

Treatment of ND-COOH particles in a hydrogen flow or in microwave CVD plasma at temperatures over 700 °C reduces the amount of COOH groups and removes oxygen almost completely from the surface to produce hydrogenated nanodiamonds. (49) With the help of hydrogenation, the strong agglomeration can be restrained. Normally there are highly reactive oxygen groups covering the surface of NDs. Hydrogenation removes these oxygen groups and makes NDs less prone to chemical reactions and therefore, also agglomeration. Hydrogen termination is also expected to stabilize the ND structure (9). Three different models of hydrogen terminated ND particles are shown in Figure 14. In a) hydrogenated spots of limited size alternate with non-hydrogenated zones showing oppositely charged surfaces; b) diamond core completely terminated by hydrogen atoms; c) partially hydrogen-terminated diamond core with regular positioning of hydrogen atoms. Diamond cores are shown by hexagons and hydrogen atoms are shown by small open circles. (38)



**Figure 14, Three models of hydrogen-terminated nanodiamond particles. (38)**

Hydrogenation of detonation nanodiamond powders is shown to be possible via annealing in hydrogen gas. This effect is based on the reaction of the  $sp^2$  shells and bonding matrix between particles with hydrogen gas. The hydrogenation process results in a breakdown of large aggregates ( $>100$  nm) of diamond bonded in  $sp^2$  matrix into primary particles with defined surface functional groups. The resulting colloids have very high positive zeta potential over a wide range of pH and a narrow distribution of particle sizes centering on the core particle size (approx. 4 nm). The colloidal solution is stable in the long term, showing no cluster formation over extended periods of time, which is an important requirement needed in bio-applications like drug delivery and biolabeling techniques. (50)

The hydrogenation process has major effect on nanodiamonds zeta potential as can be seen from Figure 15. The untreated powder has a negative zeta potential over the entire pH range and it becomes progressively more negative as the pH is increased, which is common for acid cleaned commercial nanodiamond powders (50). The hydrogenated powder however, has a positive zeta potential over the entire pH range as seen in Figure 15. Oxygenated DND is believed to have the greatest electrochemical activity while hydrogenated DND has the smallest activity (5).

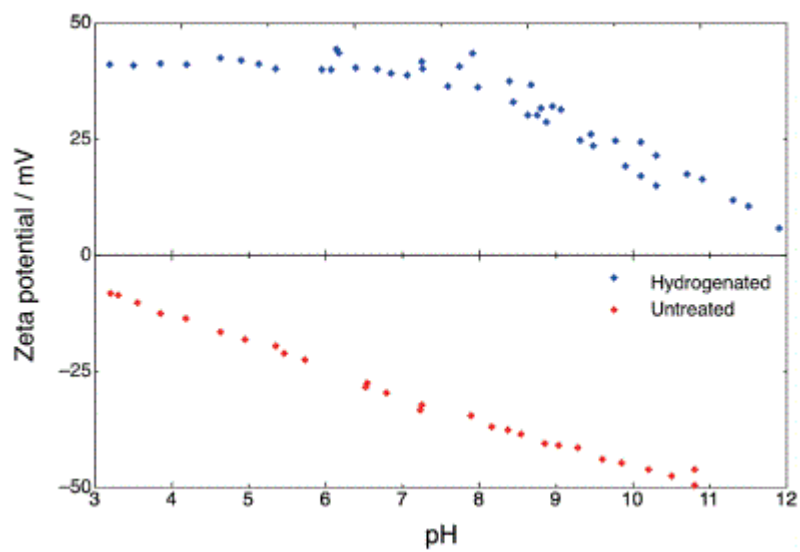


Figure 15, Zeta potential of untreated and hydrogenated nanodiamond powder as a function of pH (50)

## 4 Sensors and Applications

As discussed earlier NDs are very potential material for many applications in the field of biomedical and sensor technology. NDs have also other great properties such as: they improve the wear resistance of metallic implant coatings or medical instruments because of their adjustable roughness, high micro-hardness (3000 to 3500 kg/mm<sup>2</sup>), low coefficient of friction, lubricating quality, and fatigue durability (14). However, the large-scale agglomeration of NDs has slowed down their use in some applications in the past (2) (10). Now, by using techniques like beads milling and BASD (both methods introduced in chapter 5), a better control over the ND agglomeration is obtained and numerous applications for nanodiamonds have been planned. With current deagglomeration methods mentioned earlier ND agglomerates can be broken down to single-digit nanodiamonds that are needed in many modern applications. Also, the centrifugation technique introduced in chapter 5.1.1 allows application specific selection of ND cluster size. (5)

NDs have been considered to be an excellent candidate for nanoelectromechanical systems, biosensors, field-emission devices and other biomedical applications. Recent proposed uses of ND include as drug delivery vehicles, strengthening components of polymer composites, sorption agents in chromatography and in humidity sensors (44) (51) The reason is found in the properties of nanodiamonds that include superior hardness and Young's modulus, biocompatibility, optical properties and fluorescence, high thermal conductivity and electrical resistivity, chemical stability, and the resistance of nanodiamond to harsh environments. (8) (23) The extreme mechanical and chemical properties in ND are from the strong covalent bonds in diamond, and from diamond having the highest atomic density of any existing crystal (13). However, the unique bulk properties of diamond can be fully exploited at the nanoscale only if the interface between the nanodiamond particle core and the environment (i.e., surface of the particles) is properly engineered. (13) Therefore, as discussed in chapter 2.2, different surface groups also play an important role in the chemistry and functionality of nanodiamonds. The surface groups of the NDs needs to be modified with care to work smoothly in certain specific application. It is well-known that NDs agglomerate because of these highly reactive surface groups. Also, the high surface area-to-volume ratio of ND and the balance of sp<sup>2</sup>–sp<sup>3</sup> allow a highly active electrode material to be created. The high strength of C–C bonds and the well-researched biocompatibility makes diamond a particularly attractive substrate for biosensor applications. For example, NDs deposited on n-type silicon and functionalized with enzyme glucose oxidase have been used for detection of glucose. In the study by Villalalba et al. it was found that the response of the electrode to glucose is from 1 × 10<sup>–6</sup> M to 8 mM with response time less than 2 min (52). (53) (54)

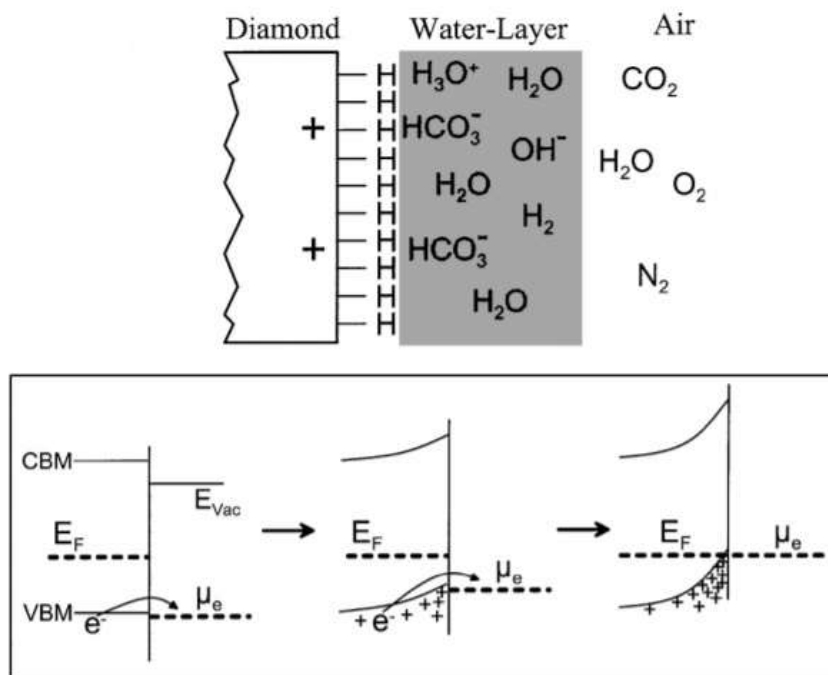
## 4.1 Conductivity and use in electronics

The electrical and thermal properties of NDs are strongly affected by surface modifications. NDs have a high thermal conductivity just like diamond, it is non-toxic (with correct surface chemistry) and can be produced in large quantities (55). Therefore, NDs are used in many materials where better thermal conductivity is needed. Yeganeh and al. made a conclusion in their study of ND thermal conductivity in Deionized (DI) water, that by increasing the dispersion quality of the nanofluids, improvements in the thermal conductivity could be made (56). With better dispersion of NDs – and therefore also lower rate of agglomerates, conductivity is indisputably better.

In addition to improving thermal conductivity of materials, it is expected that electrical conductivity of ND solution gets higher as size of agglomerates gets smaller. As discussed in chapter 2.3 there are unsaturated active bonds on the DND surface. Electron transfer is taking place between these essentially insulating particles and a redox species in solution or an underlying electrode (1). The agglomeration has effect on the redox properties of NDs. More studies to investigate how deaggregation influences the electrochemical response are necessary to determine how monodisperse the activity of the particles is or whether we are studying a collective behavior (57).

When considering electrical properties, purified ND has high electrical resistivity due to its  $sp^3$  electronic structure and the absence of  $\pi$  electrons. Annealing ND above 900 °C leads to the formation of graphitic phases and resistivity is decreased. ND is said to behave as a nearly ideal dielectric at temperatures below 350 °C, although this is sensitive to ND surface chemistry. (58) When ND is undoped and so highly resistive, it is surprising that any electrochemical response is obtained at all. In addition, despite being relatively non-conductive, NDs can promote electron transfer between a protein and an underlying electrode. The direct electrochemistry of redox proteins at electrodes is not often observed. Usually a “promoter” layer is required to correctly align the protein with respect to the electrode surface and to prevent denaturation—the ND appears to act as such a promoter. (57)

Hydrogenated diamond surfaces are known to have unique properties including hydrophobicity and, in the case of powder, typically a high positive zeta potential. Surface conductivity and negative electron affinity were also reported for hydrogenated ND. Although diamond is an insulator, it is known that hydrogen-terminated bulk diamond becomes conductive when exposed to water (59). Surface conductivity of hydrogenated nanodiamond is explained by the transfer doping mechanism: electron transfer from the valence band of the hydrogenated diamond to the redox species in the electrolyte (adsorbed water) results in holes on the ND surface. This allows two-dimensional electric conduction and a semi-conductor behavior. (59) Previously mentioned mechanism is demonstrated in Figure 16. The reaction is driven by the difference in the chemical potential of electrons in the liquid phase ( $\mu_e$ ) and in diamond Fermi level ( $E_F$ ) (60).



**Figure 16, Upper: Schematic picture of the hydrogenated diamond surface in contact with a water layer as it forms in air. Lower: Evolution of band bending during the electron transfer process at the interface between diamond and the water layer. VBM = valence band maximum (60)**

Electrodes fabricated with CVD technique are more highly active towards aqueous redox systems than conventional diamond electrodes. (61) Also, nanodiamond films possess significant electrical conductivity due to the graphitic content and high concentration of grain boundaries in the material. Since ND has a much higher surface-to-volume ratio compared to bulk diamond, the effect of surface hydrogenation of ND on its conductivity should be even more effective than for bulk diamond. Recently, Su et al. reported that hydrogenation of detonation ND by hydrogen plasma treatment increased the electrical conductivity, as estimated by impedance spectroscopy, by four orders of magnitude (62). The electric conductivity of ND is increased by almost 2 orders of magnitude when treated in hydrogen gas at 600–900 °C. Figure 17 illustrates the difference in resistivity between hydrogenated and oxidized nanodiamond with respect to temperature. The increase of the conductivity is because of the surface hydrogenation rather than graphitization. A sharp transition of ND powder was found from the relationship between the O/C ratio and the resistivity: from electrically insulating to conductive state at a critical O/C ratio of ca. 0.033. Therefore, hydrogenated ND can be considered as a surface-conductive ND, a material with many potential applications in electrochemistry, biomedical sensing, and nanocomposites. (59)



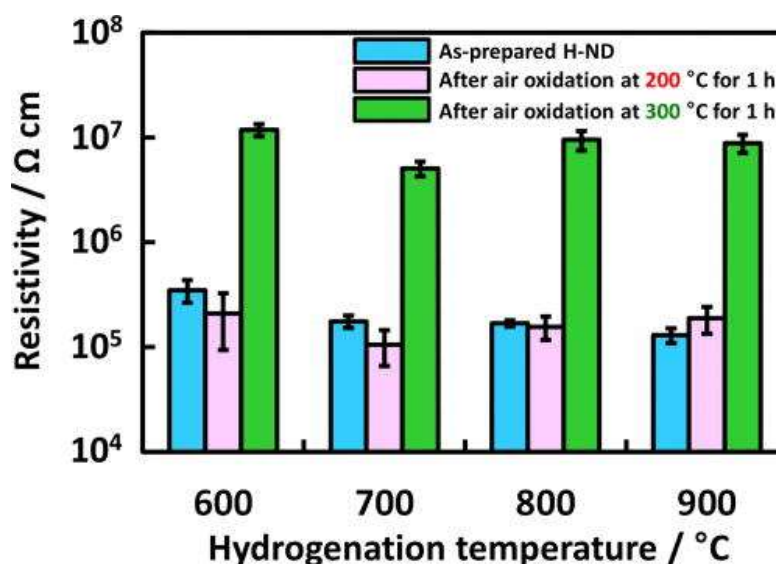


Figure 17, Resistivity of ND when hydrogenated or oxidized in certain temperature (59)

#### 4.1.1 Nitrogen doping and use in optical applications

The optical properties of NDs are exceptional due to their transparency in the visible wavelength range, which is greater than glass, and high index of refraction. Therefore, NDs are desired particles also in optical applications. Nitrogen is the most common impurity in diamond (13). The NV (nitrogen vacancy) defects and NVN (nitrogen vacancy nitrogen) color centers are the optically active defects in NDs that have received the most attention. The NV defects are shown as diamond's red or near-infrared fluorescence, while the NVN centers can be seen with bright green fluorescence (51). These impurities are demonstrated in Figure 18. The optical activity of the luminescent color centers in NDs depends on their proximity to the ND's surface and surface termination. For nanosensor applications, negatively charged NV centers are required, where the extra electron is donated by a nearby isolated substitutional nitrogen. As an indicator of the “ideal” surface for NV- centers, a slightly positive electron affinity is recommended. Surface engineering that could provide stable emission for the subsurface NV centers seems to be one of the most challenging tasks. (13) From the point of agglomeration, it needs to be remembered that NDs can't be used in some optical applications if they are agglomerated because of their size is too big. Such applications include biolabeling, biomarkers and bioimaging. The size of human cells is typically less than 10  $\mu\text{m}$ , so big ND clusters can cause multiple problems *in vivo*.

Vacancies can be produced by irradiating diamond with high-energy particles. Subsequent annealing at high temperature ( $\geq 700$  °C) causes vacancy diffusion and formation of complexes with nitrogen atoms. Currently, the primary candidates for production of luminescent ND for bioimaging applications are HPHT ND containing 100–200 ppm of N and ND derived from natural diamond containing A defects (N–N pair) with concentrations of up to 3000 ppm. DND contains up to 2–3 wt. % of nitrogen, but nitrogen clusters in DND are optically inactive. Therefore, if NDs are needed for imaging applications, the detonation NDs are not preferred candidates. (13)

Unique photoluminescent properties of crystallographic defects in the diamond lattice, such as nitrogen-vacancy and silicon-vacancy complexes, have encouraged applications of NDs as fluorescent biolabels or biomarkers (51). Optically detectable spin properties of selected color centers, particularly the NV center, stimulated developments of high-resolution mag-

netometers, atomic-size sensors for electric fields, and quantum measurements of the temperature gradient within living cells (63). Shenderova et al. produced carbon-dots decorated NDs (CDD-ND) demonstrating synergistic benefits from joining two nanostructured materials in a single synthesis approach. Nanodiamonds decorated with carbon dots have excellent potential as bioimaging probes with drug carrying capacity (64). These carbon-dot-decorated NDs demonstrated stable red/near infrared photoluminescence and were used in *in vitro* cell culture imaging. By creating carbon dots on the surface of deagglomerated 5 nm detonation ND, dye-free small NDs with bright red luminescence can be produced. (13) (64) These previously mentioned characteristics of ND allow both *in vitro* and *in vivo* imaging and detection with or without surface functionalization by multiple methods. As fluorescent labels, NDs display stable, non-photobleaching, bright internal fluorescence from nitrogen vacancy defects within the crystal lattice (65). The unique Raman spectral signal for NDs allows non-destructive detection with living cells (66). (1) . The generalized model of nanodiamond shown in Figure 18 can demonstrate only the most common and important optical features discussed in this chapter.

#### 4.1.2 Boron doping and use as semi-conductor

Nanodiamonds have a large band gap of about 5.5 eV (34). However, they have proven to be good semi-conductors when doped with boron or nitrogen. Boron is the most widely used dopant when producing conducting diamond electrodes. The doped nanodiamonds function like semi-conductors, where excess electrons carry the charge. The most striking feature of diamond electrodes is their very high overpotential for both oxygen and hydrogen evolution, which leads to one of the widest potential windows (~3.5 V) currently known for any aqueous electrolytes (1). Diamond electrodes also differ from conventional electrode materials by their very low capacitance (high signal-to-noise ratio) and by the absence of surface oxide formation and reduction reactions, which are present in conventional metal or metal oxide electrode materials between oxygen and hydrogen alteration. Because of their unique electrochemical properties, doped diamond electrodes can be used in numerous applications, including the destruction of organic and inorganic pollutants in water, water disinfection, inorganic and organic electrosynthesis, electroanalytical applications, electrochemical energy technology, bioelectrochemical applications, electroanalysis, electrochemical double-layer capacitors, and batteries. (1) (13) (67) Fabrication of boron-doped DND particles during detonation synthesis can be very beneficial due to the possibility of large scale production. Boron doped ND can be considered as one of the most ideal electrode materials for electroanalysis of many chemistry compounds (68). Figure 18 illustrates how boron atoms bring excess electrons to diamond lattice.

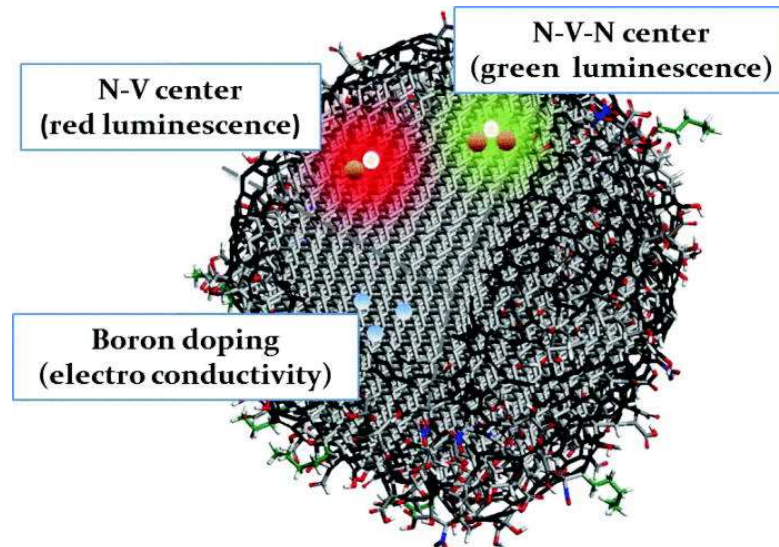


Figure 18, ND doping with nitrogen or boron. The  $sp^2$  carbon (shown in black) forms chains and graphitic patches. Some hydrocarbon chains (green) and hydrogen terminations are also seen. (Oxygen atoms are shown in red, nitrogen in blue, and hydrogen in white.) The diamond core may contain nitrogen impurities depending on the method of ND synthesis, which can be converted to luminescent color centers or form optically nonactive complexes. (13)

## 4.2 Biomedical applications

The importance of carbon nanomaterials in biological applications has been recently recognized. Examples include their potential uses in drug, gene, and protein delivery; novel imaging methods, coatings for implantable/corrosion resistant materials; biosensors/biochips; purification of proteins; medical nanorobots; and many other emerging biomedical technologies. (69) In biomedical applications of NDs the importance of small and uniform particle size is emphasized. In cellular level, no ND agglomerates are needed. Nanodiamonds themselves should be totally biocompatible and cause no stress reactions inside the body, but there is evidence that the state of oxidation of nanodiamonds can cause some toxicity (13). In a recent study by Xing et al. it was found that oxidized ND particles can activate DNA repair proteins and most likely also cause DNA damage in embryonic stem cells (70). This is an important perception, and a good advice for researchers everywhere studying ND biocompatibility *in vivo*. The positive side is that NDs surface chemistry can always be altered so that majority of all oxidic groups are removed.

It is widely thought that low concentrations of NDs are biocompatible, but unattached nanoparticles can have some toxic effects to cells when concentrations are high (e.g. between 50  $\mu\text{g/ml}$  and 200  $\mu\text{g/ml}$ ) (71) (72) (69). This is mainly because of their ability to penetrate into cells. The interaction effect depends on the type of nanoparticles, their size, concentration, time of incubation and also on cell type. When compared to other carbon nanostructures like carbon black and carbon nanotubes, NDs have a better biocompatibility when in contact with neuronal and lung cell lines (69). However, the biocompatibility of ND coatings has not been investigated extensively so further conclusions can't be done yet. (73)

The core of detonation NDs is  $\text{sp}^3$  hybridized carbon, so it shares the carbon-based composition with many biocomponents (1). But given the number of surface modifications that are possible, it is important to be certain that the functionalized nanodiamonds intended for biomedical applications remain safe. It must be noted that especially in the case of biomedical applications all the unwanted impurities on ND surface should be purified. However, as mentioned in chapter 4, the biocompatibility of NDs is well-researched subject and hopefully it will be a popular topic of study in the future. Although only few harmful effects have been found for ND materials, toxicity and biocompatibility testing of new nanodiamond-based materials should continue. (8)

One possible use for NDs could be in "hybrid" carbon based nanomaterials. These novel materials combine different carbon allotropes in order to form a unique mixture of useful properties. Hybrid materials can possibly be used to detect biomolecules because of their excellent electrochemical properties (29).

ND-mediated drug delivery has been also widely explored and their versatility as carriers for a broad array of compounds ranging from small molecules to proteins and nucleic acids demonstrated (54) (53) (74). Nanodiamonds can physically adsorb proteins and DNA oligos for biomolecular separation and preconcentration (75). A unique set of properties attractive for drug delivery applications include low cytotoxicity, large carrier capacity and a wide array of surface functional groups, which can be tailored to ensure drug binding via electrostatic attraction and environment-dependent sustainable drug release (51). However, these applications need single-digit nanoparticles or at least very small ND clusters as carriers. NDs may also be used as cellular tracer probes due to their fluorescent properties as discussed in chapter 4.1.1. Nanodiamonds are able to fluoresce in many colors without photobleaching. In addition, nanodiamonds are useful for fluorescence labeling because they are transparent in the visible wavelength range, have a large index of refraction, and have irregular surface shapes that allow for a highly dense receptor binding capacity. (76)

Nanodiamond can also be stable, biocompatible and useful for biosensing applications. NDs have been successfully used in biosensors for the detection of glucose (77). A multilayer sensor was fabricated by forming a polyelectrolyte layer on top of a gold electrode, onto where NDs were physically adsorbed. The electrochemical response of this sensor towards oxygen reduction was investigated and it was found that anodic pretreatment of the ND resulted in greatly enhanced currents for oxygen reduction. In other words, the anodic oxidation of ND increased its ability to reduce dissolved oxygen. (57) Recently, enzyme-modified diamond sensors have been realized for the detection of urea and pH of the solutions (78) The future for NDs as biomedical probes is that they will hopefully provide maximum therapeutic benefits while avoiding damage to healthy cells or tissues (1). NDs can be used as additives to intensify the action of other components. Purified NDs have already been shown to normalize blood pressure, detoxify the gastrointestinal system, and remedy cancerous conditions (14). The significant number of unpaired electrons on the surface of NDs makes each particle a powerful multi-charged radical donor, able to scavenge free radicals, which accompany many serious illnesses (14). In a study by Eidi et al. it was found out that NDs can pass even the blood-brain barrier and reach brain. Small amount of functionalized ND particles were injected into muscle of a mouse, and in all four test cases, NDs could be seen in the brain 21 days later. (79) However, NDs being redox active it must be noted that many cellular processes (e.g. respiration) have an important redox component so it is important to ensure that the redox chemistry of ND will not interfere with cell function if introduced into cells. (57)

## 5 Mechanical deagglomeration

Nanodiamonds surface modifications can affect deagglomeration, but it is not possible to produce single crystal nanodiamonds by only using functionalization. Even after additional purification, commercial DND is presently still not a powder or a hydrosol of 4-6 nm diamond particles. Therefore, several different types of mechanical deagglomeration methods have been tested to break the aggregated ND clusters back to single-digit core particles. Typically methods using strong mechanical force or high power sonication are used. These methods are extremely important for many ND applications because they can produce ND particles in the size range of less than 10 nm. In this chapter the most used deagglomeration techniques in the field of ND processing are introduced.

### 5.1 Methods of deagglomeration

As almost all current applications depend on the small particle size of nanodiamonds, it is crucial that ND clusters can be deaggregated. Typically, this means that ND particles must remain well dispersed in the media relevant to the application and resist aggregation and sedimentation. A variety of strategies have been developed for creating stable colloidal suspensions of NDs in polar and nonpolar media. (13) Although ultrasonication is a powerful tool in liquid–solid chemical reactions involving deagglomeration or fragmentation, sonication alone may be insufficient in cases when interparticle bonding is too strong. Osawa made significant progress by recognizing the microstructure of DNDs agglomerates and developed techniques of wet stirred media milling and BASD to destroy the agglomerated clusters mechanically (30). As a result, the nanodiamonds were dispersed in solution. (34) Currently of all the well-documented techniques, only ZrO<sub>2</sub> microbead-assisted wet milling and BASD are capable of breaking the core ND aggregates and producing stable suspensions consisting of primary ND particles. These methods are capable of breaking agglomerates of all sizes. It is also possible to break up porous DND aggregates partially when subjected to hydrobaric processing like in centrifuge. Their distribution in size in a water medium depends on the zeta potential appearing in the medium, which also prevents their reverse agglomeration. (2)

#### 5.1.1 Centrifuging NDs

Centrifuge is an equipment that uses rotation around a certain axis to separate substances and particles with different density from each other. Heavier particles move further away from the center than the lighter ones. Normally agglomeration is a feature that is not wanted when operating with nanodiamonds but in some applications, different sized agglomerates of nanodiamonds are needed. These ND applications include heavy duty lubrication and additives to galvanization baths. (14) It is also useful to be able to fractionate DND into different, narrow distribution of sizes for different applications. For example, only DND with aggregate sizes of more than 100 nm can form such structures that diffract light in the visible region (80). Figure 19 shows possible uses for different sizes of nanodiamond agglomerates and also, which methods are able to produce these ND clusters.

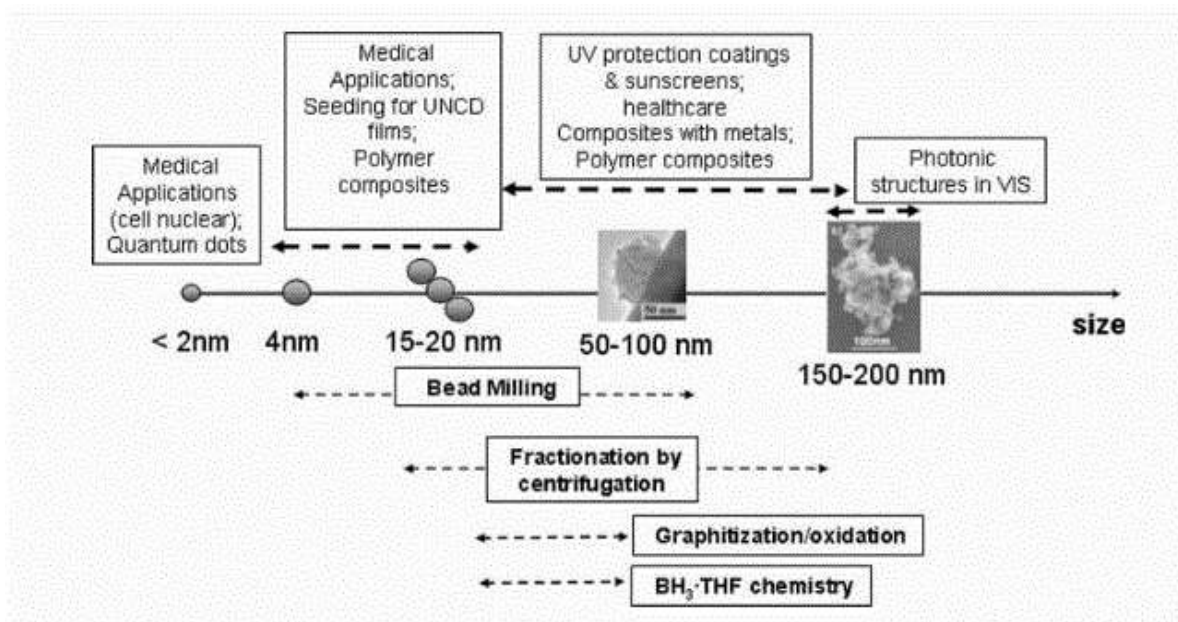


Figure 19, ND size range, possible areas of use and methods for obtaining these sizes (1)

DND solutions must possess high colloidal stability for centrifugal fractionation. It is difficult or almost impossible to fractionate an unstable suspension. DND fractionation using centrifuge has several attractive aspects. It is a totally contamination-free approach. It could be expected that the use of fractionating with ND agglomerates would be favored by at least two factors related intimately to centrifuging. The first one is that water pressure during centrifuging should provide penetration of water into the nanopores of DND agglomerates. That pressure itself should break some agglomerates. Second, penetration of water into a nanopores generates a capillary pressure as well. If water wets the pore walls, the work done by the capillary forces is close to a few electron-volts, a value comparable with the energy of chemical bonding anchoring the 4 nm agglomerate particles. However, the rotation of the centrifuge needs to be high, because at accelerations below 3000 g it is not possible to reduce the size of agglomerates to a level below 20 nm (2). Suspensions of DND with an average agglomeration size of 15 nm in water have been obtained by centrifugation. (1) Fractionation is widely used by DND manufacturers who want to provide customers diamonds within a carefully set size range.

Hydrogenated ND particles are significantly smaller than those of the untreated powder when dispersed in to water. In Figure 20a, the particle size distribution of the untreated powder before and after numerous centrifugations is shown. It can be seen that almost independently of centrifugation, the dominant particle size is over 100 nm, with no evidence of any smaller particles. The particle size distribution of the colloid made with the treated powder is shown in Figure 20b. When using centrifugation, the peak of the particle size distribution shifts to lower sizes. After centrifugation for 90 min at 5000 rpm the size distribution reduces to 28-32 nm, after 7500 rpm distribution drops to 16 nm, and then to 2-4 nm after centrifugation above 10000 rpm. The core particle size is reached after 10000 rpm centrifugation for 90 min, and the colloid can be defined as fundamentally monodisperse. Further centrifugation at higher speeds like 15000 rpm results in only a small reduction in the median and standard deviation of the particle size distribution. (50) However, as discussed later in this thesis, NDs tend to reaggregate easily. This happens mainly when NDs are dried, but there is not much research done around NDs stability in solutions.

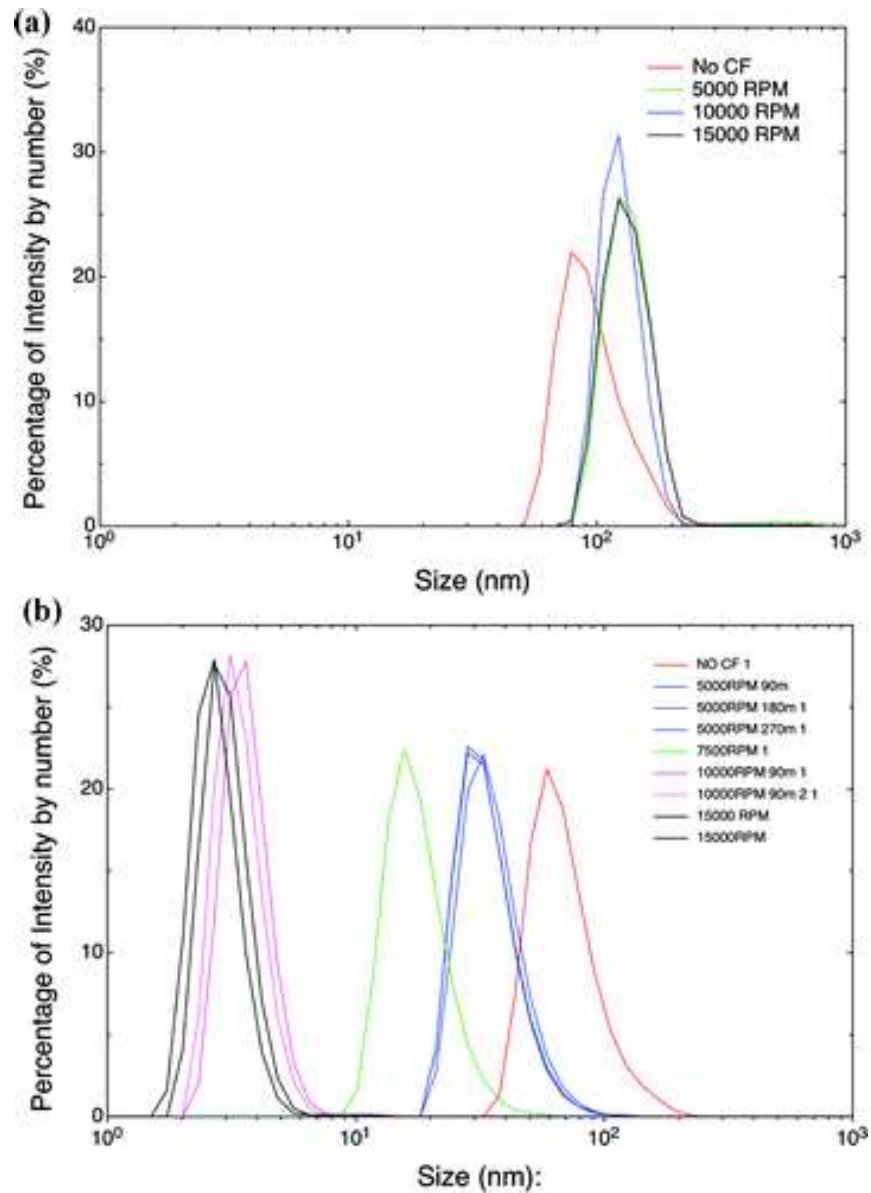


Figure 20, (a) DLS size distribution after various centrifugations of untreated powder, (b) DLS size distribution after various centrifugations of hydrogenated powder. (50)



### 5.1.2 Wet stirred media milling

Wet stirred media milling (often called also beads milling) with zirconium beads seems to be an effective method of disintegrating DND agglomerates (30). Milling technique is fast, controllable and scalable so it has been adopted to deagglomerate functionalized DNDs. (81) In stirred media milling, micrometer-sized beads are agitated to induce high-energy impact and shear forces to nanodiamond structures. The mechanism is the centrifugal force in the planetary ball milling together with milling media creates an impact force on powder particles (81). Basically, beads-milling destroys agglutinate structure including its grain boundary region by using brute force (30). During beads milling different types of collisions occur. Figure 21 demonstrates these collisions. The simplified schematics of the media milling device can be seen in Figure 22a. It is capable of breaking up to 90% of core agglomerates, with formation of a suspension consisting of single nanodiamond particles. Stirred media milling in water with the help of zirconium beads is an effective method for breaking the aggregates up and simultaneously dispersing them in water. (82) Water also removes the heat created by the process (9). Milling technique has been created for deagglomeration and functionalization purposes and therefore, DNDs to milling medium ratio has also been adjusted to get optimum results. A ratio of 1:7 of DNDs to milling media has been controlled and all experiments should be conducted at this optimized ratio (81). Therefore, usually around 70–80% of the mill-space is loaded with beads (9). Best results are gained when a high power sonication for up to 1 h is used with milling technique (4).

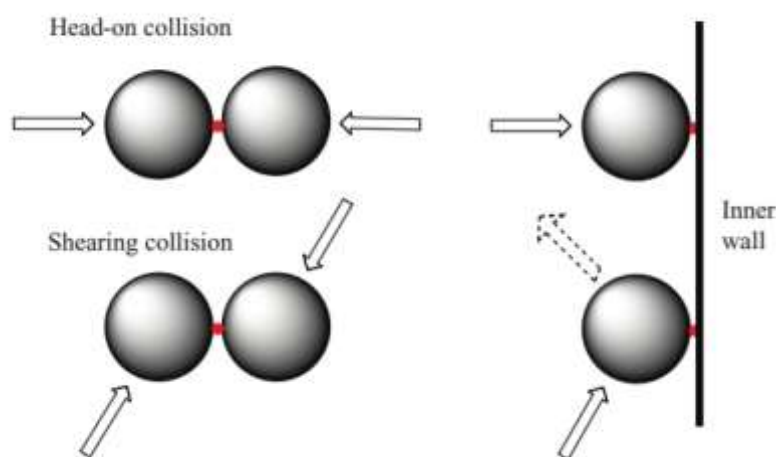


Figure 21, Types of collisions involved in beads milling (9)

Processing of NDs, specifically the high shear forces generated during bead milling, may cause additional graphitization of the sample (30). As the zirconium beads, 30  $\mu\text{m}$  in diameter, are spherical, shearing collision of beads takes place at a small contact area where large amounts of kinetic energy are released to produce high-temperature (calculated to reach 1527  $^{\circ}\text{C}$ ) and high-pressure at this point. The collisions are demonstrated in Figure 21. These conditions are severe enough to convert small portions of diamond carbon atoms on the surface into curved and partial graphitic layer by diamond–graphite phase transition. (30) Since DNDs have very pronounced sorbent properties, adsorbed fragments of  $\text{sp}^2$  carbon and hydrocarbon molecules may remain attached to their surface even after surface modification. After full deagglomeration by bead milling, ND particles must be purified from  $\text{sp}^2$  carbon formed during milling. (13) It should be noted here that the fragments of graphitic grain boundaries and zirconium particles from collision of microbeads still remain in the resulting colloid of primary particles. (30) So milling technique leads to contamination of NDs with

zirconium particles about 5 nm in diameter. The problem is that zirconium is an expensive material and it is very difficult to remove from NDs because zirconium is highly resistant to most acids/bases, affecting biomedical and other possible applications of the ND. (83) Contamination is also hard to remove by centrifugation because of nanoimpurities resulting from bead abrasion. Zirconium is chosen as a microbead material because of its high density (specific gravity 6.0 g/cm<sup>3</sup>) and therefore the ability to give beads higher momentum (9). In study by Osawa, it was discovered that concentrated alkali is expected to remove zirconium contamination and also fragmented amorphous carbons generated during beads-milling. (30) Different milling media such as NaCl and sucrose have been tested to perform deaggregation of the functionalized DNDs, A-DNDs or aminated DNDs with good future perspectives. (81) Mechanical deagglomeration of ND can be done with either wet stirred media milling or beads assisted sonic disintegration like in Figure 22 below.

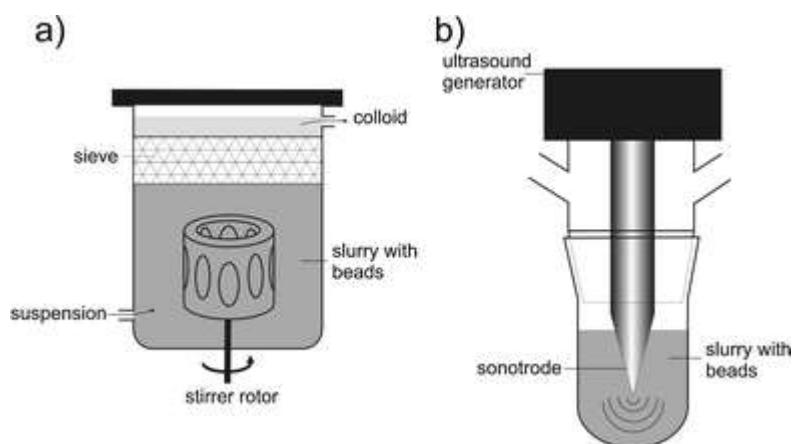


Figure 22, Schematic setup of (a) a stirred media mill and (b) a BASD apparatus (20)

### 5.1.3 Bead-assisted sonic disintegration (BASD)

As briefly mentioned in chapter 5.1, BASD technique was also created by Osawa (30). The deagglomeration characteristics in BASD resemble stirred media milling rather than beadless sonication. The mechanical stress, however, can be much greater, and the compatibility with wet chemistry processes is by far better. (7) BASD method includes high-power sonication of a ND slurry with ZrO<sub>2</sub> micrometer-sized beads (83). As the technique is simple and adaptable, the BASD setup can be easily integrated in the typical glassware systems to enable functionalization of primary nanoparticles even under conditions where particles are actually reagglomerating. (7) The BASD schematics can be seen in Figure 22b. Setup and working principle, as seen in Figure 23, is created for chemical reactions using a powerful sonicator equipped with a horn-type sonotrode. An airtight environment is maintained by a Teflon joint between the sonicator and a universal glass joint. In the reactor, microjets and shock waves generated by implosion of ultrasonic microbubbles accelerate the ceramic beads. Microjets or shock waves induced by ultrasonic cavitation can act as propelling vehicles of neighboring tiny objects. For instance, impact velocities between sonically accelerated metal particles of about 10 μm in diameter reach 500 m/s, which is far beyond the speed of stirred media milling usually operated at around 10 m/s or less (7). The presence of ceramic beads causes a high number of collision events. Breaking up of agglomerates is thought to take place owing to the impact and shear forces of the collisions between the sonically propelled beads. The higher the power density of the sonotrode, the faster the deagglomeration process. This reduces abrasion and therefore contamination from the milling

beads. Similar to beads milled samples the stability of the colloidal solution is depending on the polarity and hydrogen bond ability of the solvent. Best results are obtained with DMSO and water. (24) When the agglomerates are treated with coupling agents, freshly exposed surfaces react with the agents until all primary particles are deagglomerated. (7)

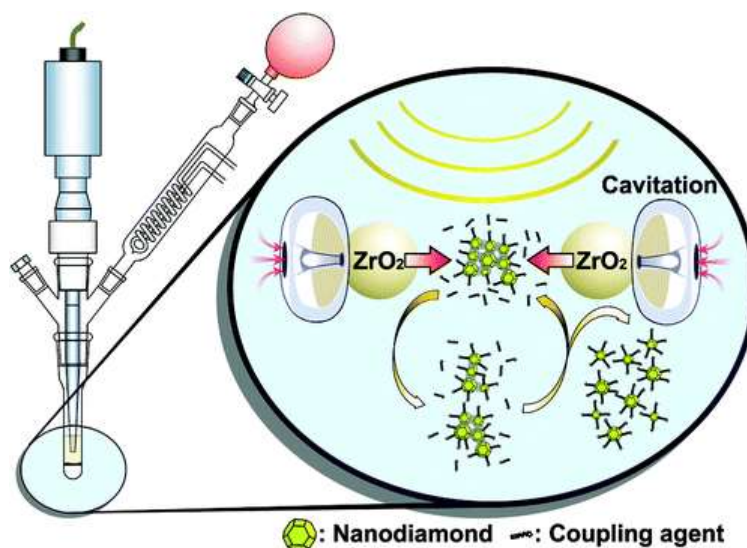
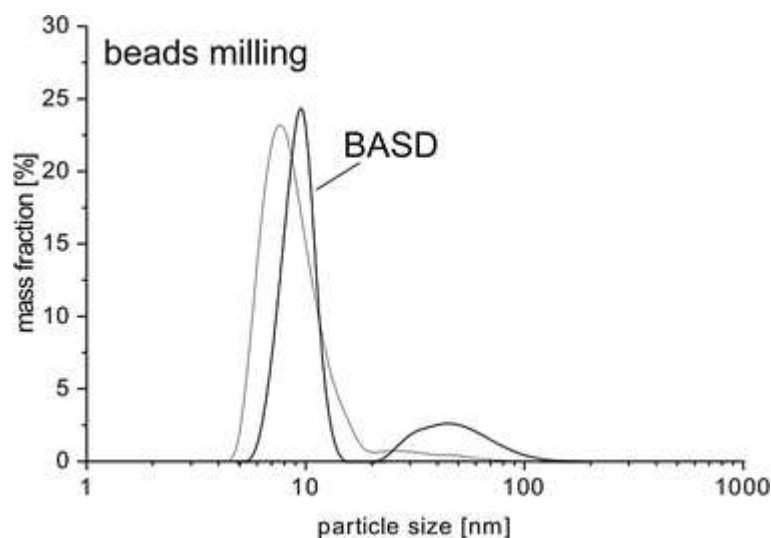


Figure 23, BASD setup (7)

The BASD method shows similar deagglomeration performance compared to stirred media milling, just by adding ceramic beads to a sonicated suspension. BASD is based on collisions of beads induced by cavitations. Therefore, in order to achieve high efficiency, a solvent with high boiling point and low viscosity has to be chosen to prevent bubble generation and to allow high-speed movement of the beads. When NDs are used in application, it must be noted that the solvent and other materials used in ND deagglomeration need to be optimal considering the planned application. Dispersibility of nanoparticles in the solvent is very important when handling BASD. A reaction under flocculating conditions gives rise to an endless cycle of deagglomeration and reagglomeration. In this case, the chosen coupling agent must reach the target particles on the exposed surface and complete the reaction before reagglomeration happens. If this reaction stabilizes the particles in the respective solvent, agglomerates will become significantly smaller. Because of the surface stabilization by the functional groups, almost no reagglomeration occurs and the primary particles are eventually liberated. BASD is a key method of deagglomeration, which is applicable not only to nanoparticles forming rather weak agglomerates such as synthetic monocrystalline nanodiamonds (SYP-ND) but also to agglomerated detonation nanodiamonds, the extreme end of tight agglomeration. BASD resulted in a remarkable decrease in the particle size: ND particles smaller than 10 nm were produced within 2 h of sonication. (7) This can be seen in Figure 24, also results for beads milling is shown to give a fair comparison of particle size distribution between these two methods.



**Figure 24, Particle size distribution of deagglomerated detonation nanodiamond. Beads milling and BASD methods results compared (20)**

Certain drawbacks of BASD should also be noted. Problems are similar than with beads milling, as were discussed in chapter 5.1.2 such as contamination with bead and sonotrode fragments and damage on nanoparticles. However, as mentioned earlier, this problem can be dealt with concentrated alkali (30). To avoid additional purification of NDs other materials for the beads can be considered, but for sonotrode Titanium alloy there are currently no substitutes. Ti-derived impurities, although usually much less important than those from beads, must be removed using corrosive reagents. However, deagglomeration with BASD is achievable by adjusting the energy input, provided the interparticle bonds are weaker than the particles themselves. Therefore, parameters such as the amplitude of sonication and type of beads should be optimized for each case. Furthermore, it should be noted that BASD might induce additional radical reactions, which might result in unexpected products (e.g., reactions with the solvent). (7)

Also, ND amorphization or graphitization might cause problems when using BASD method. However, as investigated by Niu et al. the amorphous carbon can be heated and finally destroyed at high temperature (34). Although, heating might also lead to destruction of the current surface functionalization. The fiercely exploded carbon species could further destroy the covalent bonds between the primary nanodiamonds. The carbon species react with the solution (i.e.,  $C + H_2O = CO + H_2$ ) and escape as gases. The elimination of amorphous carbon and the breaking of covalent bonds between primary nanodiamonds leads to the well-dispersed nanodiamonds in solution. (34) Despite the drawbacks, the simultaneous functionalization with BASD enables a wide variety of possible reactions on nanoparticle surfaces, including photochemistry and electrochemistry under inert conditions, due to the simple setup and procedures. (7) (83)

### 5.1.4 Salt-assisted ultrasound deaggregation (SAUD)

The technique utilizes the energy of ultrasound to break apart nanodiamond aggregates in sodium chloride aqueous slurry. System requires only aqueous slurry of sodium chloride and standard horn sonicator, still yielding highly pure well-dispersed nanodiamond colloids. SAUD is capable of producing single-digit nanodiamond particles with efficient and inexpensive way. SAUD does not require any costly materials or expensive equipment and can be used in any laboratory. The technique works with different nanodiamonds from different manufacturers, provided they are purified and air oxidized, meaning that they have a large amount of carboxyl groups on the surface. No pH adjustment is required either. It does not use any zirconium balls, so no zirconium contamination is produced. There are no toxics or difficult-to-remove impurities used and the method is therefore well-suited to produce NDs for numerous applications, including theranostics, composites, and lubrication, etc. The majority of SAUD results reported are produced with NaCl and KCl, other salts or crystalline compounds, including organic solids, can potentially be used. (84)

## 5.2 Comparison of deagglomeration methods

The ideal method for deagglomeration of NDs should be facile, simple and yield single-digit particles without impurities. Reduction in treatment time and avoidance of costly milling aids and equipment are beneficial as well. Different deagglomeration techniques were introduced in chapter 5.1 and are compared in Table 3.

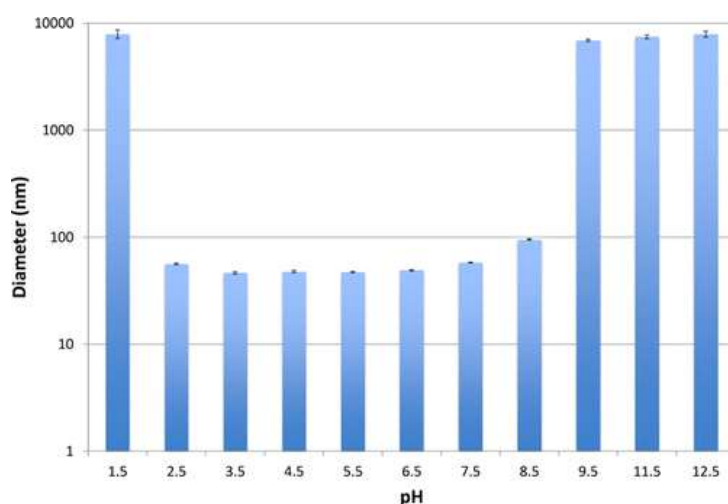
Table 3, Different techniques of ND deagglomeration (84)

Method	Additive	Ultra-sound power, W	Workup	Di-amer, nm	Comments
BASD	ZrO <sub>2</sub> and SiO <sub>2</sub> microbeads	400-500	dissolution of ZrO <sub>2</sub> contaminants in strong base or acid	4-5	difficult to remove contamination from microbeads
Beads-assisted ball milling	ZrO <sub>2</sub> microbeads	-	dissolution of ZrO <sub>2</sub> contaminants in strong base or acid	<10	difficult to remove contamination from microbeads
Salt-assisted attrition milling	NaCl crystals	-	acid treatment to remove Fe and other metals followed by pH adjustment to 11	<10	Fe contamination as result of wearing of steel balls and parts of the mill
Ultra-centrifugation	-	500	centrifugation at 18000g	4	very low yield
SAUD	NaCl crystals	150	washing/centrifugation steps (x2)	5-10	no contamination; works with different commercial NDs

As can be seen from Table 3, all methods produce good results when observing size range of NDs. Although each technique is capable of significantly reducing the size of ND agglomerates and even yielding single-digit NDs, they have crucial disadvantages that prevent their use in certain applications, especially in biology and medicine. These downsides include high price, low yield or problems with contamination. Ball milling and BASD are currently the most common methods used in ND processing, but as mentioned earlier in chapter 5.1.2, they contaminate sample with zirconium (84). However, zirconium is not considered to be toxic so it does not cause major concerns. Ultracentrifugation is contamination free approach, but it has a very low yield and can be slow when handling larger amounts of NDs. Attrition milling technique uses water-soluble, nontoxic, and noncontaminating crystalline milling media such as sodium chloride or sucrose. After the milling process is complete, the milling media can be easily removed by water. However, attritor mills are expensive, so not all research laboratories can use them. Steel jars, shafts, and balls, which are standard components of the attritor mill, provide sources of metal contaminants and are subjected to severe wear and corrosion during the milling, especially in the presence of salt. This leads to ND contamination with unwanted metal impurities, such as Fe and other components of steel. SAUD does not produce contaminants into nanodiamonds, which is an important advantage when high-purity nanodiamonds are needed, for example, in biomedical applications. SAUD is an inexpensive way to deagglomerate NDs, but it is still relatively new technique that has not been tested much yet. For future purposes ND deaggregation with SAUD method seems potential. However, currently BASD and ball milling -methods produce the most reliable results and are widely available. (84)

## 6 Dispersion in solvents

As discussed earlier in chapter 4, there are many potential applications planned for NDs. In all of the applications mentioned, an understanding of the interaction between the surface of ND particles and a solvent environment is essential to their exploitation. The types of surface groups on the ND play a key role in the dispersion of the ND into particular solvents. Modifications in detonation nanodiamond surface groups can produce a high density of chemical functionalities, as compared with nanoscale diamond powders, since almost 15 % of all atoms in the DND primary particle are located on the surface and therefore are solvent-accessible (1) (5). Depending on the surface chemistry and environment, the ND particles may interact more strongly with each other than with the solvent. This leads to particle agglomeration or possibly to the formation of non-desired molecules. (44) As well known, DNDs tend to form aggregates in solutions. These aggregates become significant when their concentration is above 0.1 wt % (85). Despite the spontaneous aggregation, the DND suspensions remain stable even though some weak cluster–cluster interactions can be observed. These clusters can even penetrate each other when solution has strong concentration and form gel-like nets. (85). Sonication and other deagglomeration methods introduced in chapter 5 are typically used to break up the agglomerates before dispersion in solvents. (86) In one recent study by Paci et al. the correlation of pH and ND agglomeration was investigated. It was found that aggregate sizes of ~50 nm remain relatively unchanged upon titration to pH values as low as ~2.5 and as high as ~8.5. The aggregate diameter implies that there were approximately 1000 particles in the average aggregate, assuming a particle diameter of 5 nm. (18) The average agglomerate size in respect to pH is shown in Figure 25.



**Figure 25, Average aggregate size versus pH. pH changes were induced using 0.01 M NaOH or 0.01 M HCl, with an initial nanodiamond concentration of 5 mg/mL. (18)**

A recent study of ND dispersion to different solvents was carried out by Khan et al. (81). The dispersion of different samples of DNDs in different solvents can be seen in Figure 26. Different colors can be observed in solutions because of Rayleigh scattering, as the particle size is well below  $\lambda/4$ . Other reason for varying colors might be formation of aromatic substructures on the particle surface during the deagglomeration procedure. The resulting aromatic structures absorb light in different regions of the spectrum and in total a dark brown coloration is obtained. (24)

Solvents used in tests were water, DMSO and ethanol and they are marked with capital letters A, B and C. Small letters (a), (b), (c) and (d) indicate differently treated NDs. In this case:

- a) as-received DNDs
- b) ammonium bicarbonate treated DNDs
- c) NaCl + ammonium bicarbonate treated DNDs
- d) sucrose + ammonium bicarbonate treated DNDs.

NaCl + ammonium bicarbonate ball milled and sucrose + ammonium bicarbonate ball milled DND samples showed better results in each solvent. Samples in which particles settle down imply poor dispersion, which may be due to small quantity of attached functional groups. The superior dispersion of NaCl assisted ball milling of DNDs is likely due to the use of NaCl grains as milling media. These grains are smaller in size providing greater area of contact to DNDs particles and during collision maximum energy is transferred from NaCl crystals to DNDs particles. Superior dispersibility has been achieved in water and DMSO for both NaCl assisted milling of DNDs and sucrose assisted milling of DNDs. (81)

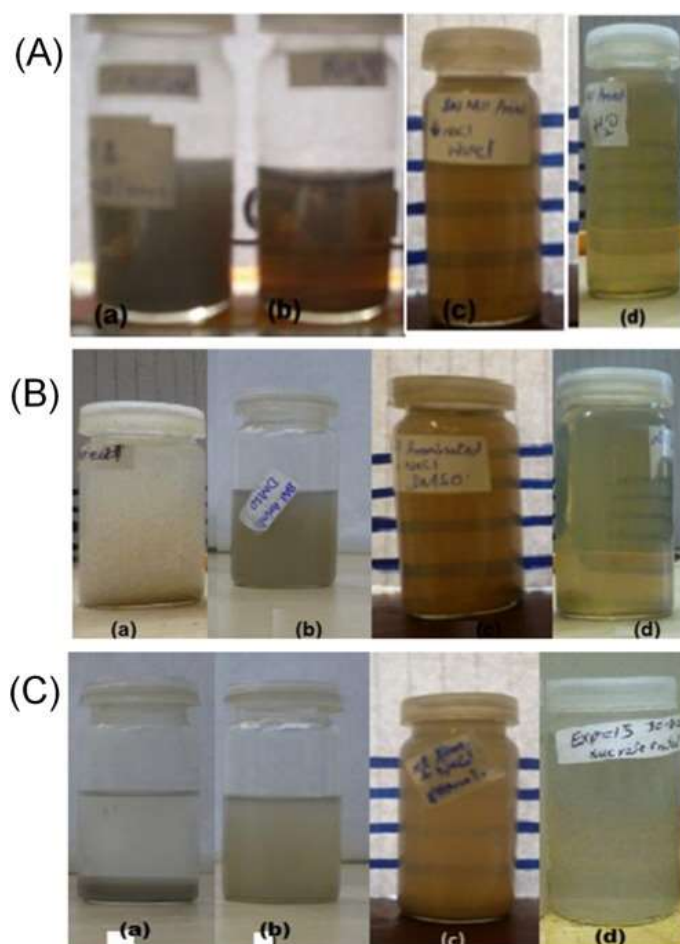


Figure 26, NDs dispersed in (A) water, (B) DMSO and (C) ethanol. Samples (a), (b), (c) and (d) indicate the type of DND used (81)



## 6.1 Zeta potential

Measurements of the zeta ( $\zeta$ ) potential can provide useful information about the charges associated with the surfaces of particles in a colloid (18). DND acquire a certain type of charge in colloids and that type, zeta potential, is important considering NDs sorption and electrophoretic properties. Zeta potential of DND suspensions is another characteristic depending on the DND surface composition and will be defined by the majority of the surface groups (basic vs. acidic species). These zeta values largely determine the colloidal stability, coagulation, and rheological characteristics of DND hydrosols. The values outside the region + 30 to - 30 mV indicate good resistance to agglomeration and sedimentation of DND in suspensions (87). There are types of commercial and modified DNDs with highly positive and highly negative zeta potential values in solutions. (1) Zeta potential values of different ND types can be seen in Figure 28. The effect of zeta potential on suspension properties is demonstrated in Figure 27.

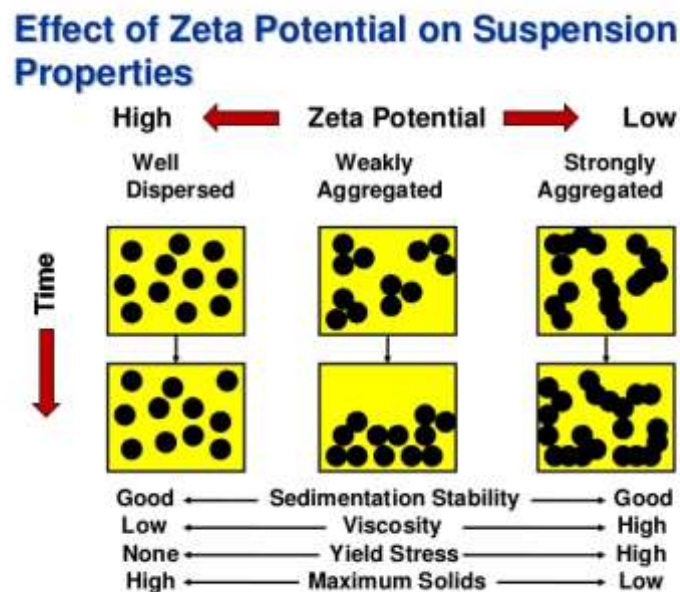


Figure 27, Effect of Zeta potential on suspension properties (88)

As determining zeta potential is very important for ND suspensions, the measurements need to be performed with care. The calculations have to be exact in order to analyze the properties of ND solutions and finding the correct use for that ND suspension. Therefore, the equation used for determining zeta potential plays an important role in the process. Zeta potential is obtained from the electrophoretic mobility of ND particles measured using the laser Doppler electrophoresis technique. After that, usually one of the two most popular theoretical expressions of electrophoresis are typically used, either Smolukhovski's or Henry's. This is done for conversion of the electrophoretic mobility into zeta potential values. (13) It was concluded in research by Petrova et al., that the use of the Miller formula (Equation 1), provides the most accurate zeta potential values for DND aggregates (89). Miller formula takes into account the electromigration fluxes of ions and electroosmotic flows of solutions in pores of dispersed particles. For monolithic ND particles (e.g. HPHT and fully deagglomerated 5 nm

DND) Ohshima's approximation developed for the arbitrary zeta potential value and the Debye length/particle radius ratio is more appropriate (90) (89).

$$u_e = \left(1 + \frac{P_a}{2}\right) \left(\frac{2\varepsilon\varepsilon_0}{3\eta} \zeta^M + \frac{\varepsilon\varepsilon_0}{3\eta} \psi F_{eo}\right) \quad \text{Equation 1,}$$

where  $P_a$  is the effective volume fraction of monolithic particles in an aggregate,  $\zeta^M$  is the zeta potential of these individual particles in a corresponding solution,  $\psi$  is the zeta potential of these particles inside a porous aggregate, and  $F_{eo}$  is a function that characterizes the intensity of an electroosmotic flow in the pores (90).

The majority of the surface groups for DND treated in air and possessing a negative zeta potential were carboxylic anhydrides, which convert into carboxylic acids in water suspensions. The DND with positive zeta potential, obtained by soot oxidation using chromic anhydride in sulfuric acid, has very low if any concentration of carboxyl groups. The level of surface oxidation of DND with positive zeta potential was much lower as compared to DND with negative zeta potential. (13) Treatments resulting in NDs with negative zeta potential seem to provide more complete oxidation with predominant carboxylic acid and acid anhydride groups on the surface. These groups are responsible for negative zeta potential in water suspensions over a wide range of pH. Functional groups on the surface of NDs with positive zeta potential are much more diverse. A recent thorough analysis by Paci et al. studied fully deagglomerated NanoAmando (particle size about 5 nm) diamonds with positive zeta potential, it was concluded that the groups are amphoteric, with high concentrations of phenols, pyrones, and sulfonic acid groups. (18) The sulfonic acid groups originate from the sulfuric acid used in the ND purification process. It was also concluded that the positive surface potential of these NDs originates from the presence of pyrones. The phenols and pyrones are closely related to the graphitic surfaces usually formed during beads milling. (13) (30) ND particles in aqueous suspensions can possess very complicated interfacial phenomena. These include protonation/deprotonation of ionogenic groups that lead to positive or negative zeta potentials of ND and valence hole formation resulting in surface conductivity and positive zeta potential of NDs. These effects occur because of different groups on ND surface and the presence of  $sp^2$  carbon structures. (13) As mentioned earlier in this chapter that different types of nanodiamonds may have either positive or negative zeta potential. This effect is demonstrated in Figure 28, with respect to agglomerate size. The chemicals or techniques used for modification can be seen in brackets after the ND name.

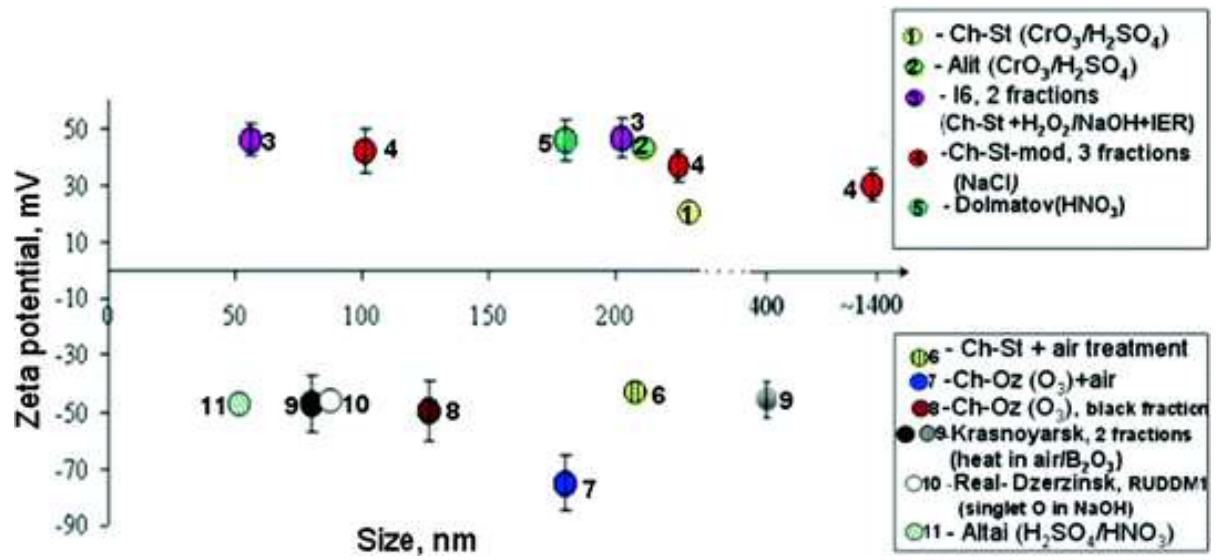


Figure 28, Zeta potential and average aggregate size for different types of commercial DND (9)

## 6.2 Solvents

Dispersion and at the same time deaggregation of DNDs can be carried out in different solvents. This process is very important for many applications where single-digit DNDs are needed. However, it has been noted that the dispersed nanodiamonds aggregated rapidly in non-polar solvents. In contrast, they dispersed well in polar solvents such as DMSO, DI water and ethanol. This dispersion behavior is linked to the polarity of the nanodiamond surface. In non-polar organic solvents, the colloidal solution is not stable and rapid precipitation occurs. This is due to the hydrophilic functional groups on the surface of the primary particles, resulting in a zeta potential of +30 mV. (20) As seen in Figure 27, high zeta potential leads to a better dispersion of ND. Therefore, the most commonly used solvents with nanodiamonds are polar solvents such as DMSO, alcohols and DI water. It is important to find solvent for ND with good chemical compatibility for better dispersion. Typically, it is necessary to use surfactants in solutions containing NDs (27). However, the excess surfactant with a high boiling point is hard to remove from a mixture. The remaining surfactant in solution would limit the practical applications of NDs. (10)

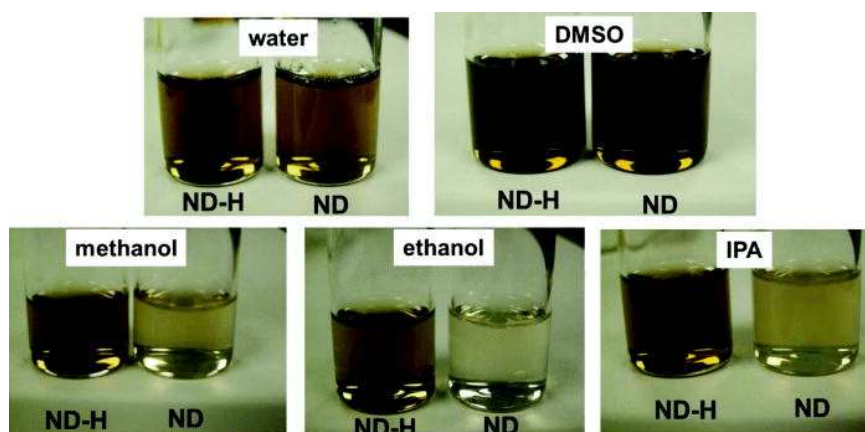


Figure 29, Results of fractionation by centrifugation in different solvents. ND has polyfunctional surface groups. ND-H was obtained from ND by treatment in a flow of  $H_2$  for a few hours at 400 °C. Centrifugation was done at 20,000 g-forces for 10 min. (13)

### 6.2.1 DI water

Deionized water is water that has had almost all of its mineral ions removed. DI water is a polar solvent that consists of hydrogen and oxygen and its chemical formula is  $H_2O$ . However, zeta potentials of ND solutions in water seem to be lower than in DMSO. (87) ND surface carboxyl groups dissociate in water suspension with formation of  $-COO^-$  ionic derivatives bonded covalently to the surface and of the  $H^+$  cations. When NDs are dispersed in DI water, dissociated acidic groups cause a negative charge on the ND surface as can be seen in Figure 10a. (1) (91) This makes DND in water suspension an ideal object for purposeful modification of the surface by various organic and inorganic atomic groups, metal ions, fluorescent organic complexes and biologically active compounds such as biomolecules (9). Usually the color of NDs in water suspensions is gray or almost black, as seen in Figure 29.

### 6.2.2 Dimethyl sulfoxide (DMSO)

DMSO has a low toxicity when considering interaction with humans (oral, inhalation and dermal) or environment. DMSO is one of the strongest hydrogen-bonding acceptors known, several orders of magnitude stronger than water. (82) Therefore, sizes of ND aggregates dispersed in DMSO are smaller than in suspensions of DI water. It has been found by Shenderova et al, that a mixture of DMSO/MeOH was one of the best solvent systems for fractionation since it produced small-sized nanodiamond particles (87). However, another important finding of that work was the limitation of using DMSO for dispersions and efficient fractionation of ND with only positive zeta potential. (87)

DMSO has polar resonance structure in which oxygen has a partially negative charge and sulfur has a lone electron pair and a partially positive charge. It is also suggested that the geometrical shape of DMSO affects solvation ability to some extent as well (87). The structure of the DMSO molecule is a trigonal pyramid in shape. At the apex of the pyramid there is a sulfur atom and it's directional lone pair of electrons helping to solvate molecules. (92) DMSO contains a highly polar S = O group, that forms very strong hydrogen bonds with molecules possessing acidic hydrogen atoms. These molecules include for example hydroxyl groups and protonated forms of pyrone-like structures, hypothesized to exist on the surface of NDs with positive zeta potential. (1) In addition to a highly polar group, DMSO contains two hydrophobic CH<sub>3</sub> groups. Thus, a solvation shell preventing ND particles from agglomerating is possibly formed through hydrogen bonds between surface groups of nanoparticles and S = O groups of DMSO enclosed by methyl groups at the periphery of the shell. As a result, there are no 'bridges' between ND particles formed by hydrogen bonds between solvent molecules–ND surface groups and solvent molecules themselves as they take place in water and alcohol suspensions of NDs. (87)

As mentioned earlier, the yield of small-sized ND particles in centrifuged suspensions of DMSO is significantly higher than that in water. In addition, the DMSO supernatant contained the smallest possible particle size, the 5 nm primary particles and an improved colloidal stability. NDs in DMSO also have high dispersivity and may be dried without causing reagglomeration. However, the relatively high surface tension of DMSO needs to be adjusted by adding alcohol, otherwise during drying NDs would be pulled together again. (87) For a comparison, surface tension of water is 71.8 mJ m<sup>-2</sup>, of DMSO 43 mJ m<sup>-2</sup> and of alcohols 22–23 mJ m<sup>-2</sup> (93) Zeta potentials and average particle sizes for detonation ND suspensions in both water and DMSO (1% wt/v) are illustrated in Table 4. (87) As can be seen from Table 4, not all functionalized NDs tend to disperse better into DMSO. It is mainly the compatibility of solvent and the surface chemistry of ND that makes the good dispersion.

**Table 4, Average particle sizes are provided based on intensity-based measurements for the unimodal particle size distribution. a) After sonication; sample sedimented within 2 h; b) After sonication; residue formed within hours. (87)**

Sample	Size in H <sub>2</sub> O, nm	Size in DMSO, nm	Z-potential in H <sub>2</sub> O, mV	Z-potential in DMSO, mV
1. "Alit"	260	190	31	33
2. Ch-St	250	215	17 (sedim.)	30
3. i6-NH <sub>2</sub>	144	158	42	35
4. i6	183	170	43	35
5. Ch-B	110	105	42	35
6. Ch-St-T	210	300 a	-35	-4 (sedim.)
7. Oz	200	290 a	-49	-22 (sedim.)
8. Kr-B	80	Unstable	-47	Unstable
9. RUDDM	75	138 b	-49	-22

### 6.2.3 Alcohols

For a variety of applications, suspensions of ND in alcohols are beneficial because alcohols possess low surface tension, low viscosity, and low temperature of vaporization. (87) Unlike water, alcohols like ethanol and ethylene glycol (EG) molecules have hydrophobic, non-polar alkyl groups as well as polar, hydrophilic –OH groups and so are likely to exhibit preferential orientation in how they interact with the ND surface and with each other. (44) Alcohol suspensions of typical polydispersed DND are not very well stabilized even with positive zeta potentials, as compared to the same DND suspensions in DI water and DMSO. It was also observed that the fractionation of typical DND (both with positive and negative zeta potentials) is not efficient for DND suspensions in pure alcohol solvents suspensions. An exception is hydrogenated ND, which was found to be stable in alcohols. (13)

The particle sizes and concentrations of NDs in different solvents and supernatants are summarized in Table 5. However, the stability of polydispersed ND I6 is not high in alcohols, and the fractionation of I6 in pure alcohols is not efficient, as can be seen from Table 5. (87)

**Table 5, Comparison of the particle sizes of I6 ND in different solvents as well as size and yield in a supernatant after centrifugation of 1 wt.% I6 ND at the same conditions. (87)**

<b>Solvent</b>	<b>Size of polydispersed i6 ND, nm</b>	<b>Z-potential of i6 ND, mV</b>	<b>ND size in supernatant, nm</b>	<b>%wt. Of ND in supernatant</b>
H <sub>2</sub> O	183	43	25	0.25
DMSO	170	35	30	0.5
Methanol	185	25	40	0.05
Ethanol	210	22	45	0.01
IPA	175	39	45	0.1
DMSO-H <sub>2</sub> O (1:1)	-	-	47	0.45
DMSO-Methanol (1:1)	-	-	30	0.25

## 6.2.4 Reaggregation

Powders of DND are more economical and safer to transport than suspensions and allow the flexibility in formulating different types of suspensions. However, providing DND powders is challenging because the drying of DNDs from fractionated suspensions in water causes agglomeration with an increase in the volume average size up to 100%. (87) When NDs are dried from water and other solvents, they agglomerate more aggressively. Drying NDs can lead to reaggregation due to capillary forces pulling the individual nanoparticles together. Attractive van der Waals forces also create reaggregation. (8) This makes ND functionalization more difficult because usually it involves drying of the ND from a solvent. Bondar and Puzyr developed a modification technique based upon sonication-assisted treatment of NDs in a NaCl solution (94). The result was purified NDs because of Na<sup>+</sup> ions absorbing into the ND surface. The attractive feature when treating NDs with NaCl was the possibility of drying NDs from a hydrosol to a powder without additional agglomeration. This may also explain why reaggregation does not occur when nanodiamonds are produced through NaCl-assisted milling. DND powder with average aggregate size of ~40 nm after dispersion in DI water was obtained using this method. (1) (94) ND sizes in original solvent and in resuspension are compared in Table 6.

**Table 6, Sizes of ND in initial suspensions as well as after drying and re-dispersion. a) ND powder was first dispersed in DMSO, then methanol added; b) ND was fractionated in a mixture DMSO/H<sub>2</sub>O; c) res. — Small residue presents; d) ND was fractionated in water, then DMSO added.**

Solvent	ND size in solvent, nm	ND size after drying and resuspension, nm	Solvent for resuspension
DMSO	35	35	DMSO
DMSO	35	37	H <sub>2</sub> O
DMSO	35	50	DMSO:methanol (1:9) a
H <sub>2</sub> O	30	98	H <sub>2</sub> O
DMSO/H <sub>2</sub> O (1:9) b	40	55 (res.) c	H <sub>2</sub> O
H <sub>2</sub> O+DMSO (1:1) d	30	48 (res.) c	H <sub>2</sub> O



## 7 Experimental

In this master's thesis, the idea is to compare nanodiamonds agglomeration behavior in different solutions. Nanodiamond solutions are prepared for tests with different solvents: DI water, ethanol, DMSO and ethylene glycol. Each nanodiamond solution is first sonicated (with bandelin sonorex rk 102 h) with 35 kHz frequency for 2 hours, to be sure that all the bigger agglomerates are broken down. The use of silicon as a substrate for the ND films is convenient because of their thermal expansion coefficients are very close. Nanodiamond solution is spin-coated on the silicon sample and then characterized with the help of a SEM. Before SEM, the sample needs to be made conductive with a thin layer of metal. This operation is done by sputtering a thin platinum coating on top of the sample surface. At last, the SEM micrographs of the samples are closely observed to find differences in agglomeration between solutions.

### 7.1 Materials

Nanodiamonds used in the study were hydrogen terminated DNDs produced by Carboneon (more details found in Appendix 1). The ND solutions obtained for tests were dissolved in three different solvents: 2.5 wt% ND in DI water, 4 wt% ND in ethylene glycol and 0.05 wt% ND in ethanol. Because no ND powder was available, all nanodiamond solutions were diluted to 0.05 wt% to get comparable results. Solvents used were: DMSO ( $C_2H_6OS$ ,  $\geq 99\%$  by MP Biomedicals), ethylene glycol ( $C_2H_6O_2$ , 99,8% by Aldrich), DI water and ethanol ( $C_2H_6O \geq 99.5$  ETAX Aa by Altia). After dilution there were four solutions with concentration of 0.05 wt% ND in ethanol, DI water, DMSO and ethylene glycol. DMSO suspension was diluted from NDs in ethylene glycol so there is small amount of EG in solution. These solutions are all listed in the Table 7.

Table 7, Solvents and NDs used in tests

Solvent	Chemical formula	Amount of ND in solution	Type of ND	ND crystal size
DMSO 99%	$C_2H_6OS$	0,05 wt%	Hydrogen terminated DND	$4.2 \pm 0.5$ nm
EG 99.8%	$C_2H_6O_2$	0,05 wt%	Hydrogen terminated DND	$4.2 \pm 0.5$ nm
Ethanol 99.5%	$C_2H_6O$	0,05 wt%	Hydrogen terminated DND	$4.2 \pm 0.5$ nm
DI water > 99%	$H_2O$	0,05 wt%	Hydrogen terminated DND	$4.2 \pm 0.5$ nm

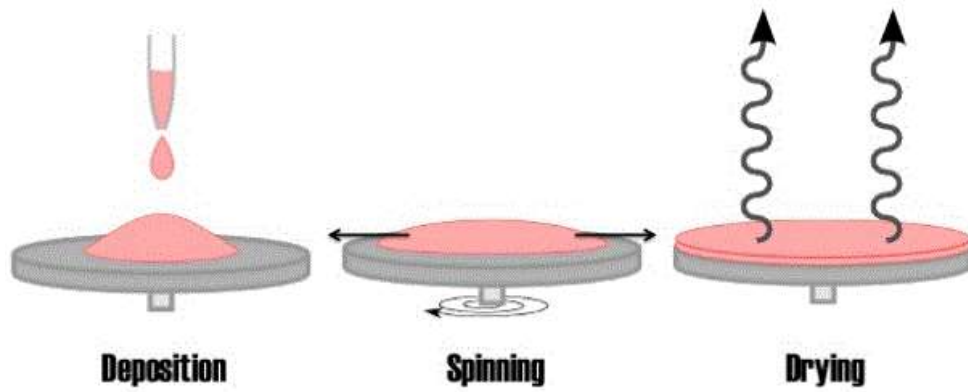
All four ND solutions after dilution are seen in Figure 30. Slightly different colors can be observed. NDs dissolved in ethanol had some coagulation on the bottom of the solution before sonication but not much other differences could be seen.



Figure 30, ND solutions used in tests

## 7.2 Spin-coat method

All 4 samples were prepared with spin-coat method. At first sample was inserted in to spin-coater, where idea is to spin the sample until the nanodiamond solution is smoothly spread on the surface. In this study the most important issue was to make sure the sample is completely covered in NDs and that the solution is evenly distributed. The silicon sample was set on (Laurell ws 650mz 23npp) spin-coater and  $25\mu\text{l}$  of ND solution was carefully dropped on it. Then the spinner was set to accelerate to 1000 rpm, then spin for 10 seconds at 1000 rpm and finally 20 seconds at 4000 rpm. Rotation of sample is continued while the solution spins off the edges of the substrate. This was done three times for all the samples to make sure the coating is evenly distributed over the sample surface. As the solvent used is volatile, it evaporates during the process. The basic principle is that the higher the angular speed of spinning, the thinner the film. The thickness of the film also depends on the viscosity and concentration of the solution and the solvent. (95) In this case, the solution needed a high speed and it was important that no liquid was left on the surface of the sample to dry. ND solutions tend to leave clear “ring” marks when they evaporate because nanodiamonds in the drop are concentrated more on the edges of the drop. Therefore, it is important that the spinning is started immediately after the drop is dropped on the sample. In this case the reaction time after dropping the solution and starting the spinning was about a second. Basic steps of spin coating are demonstrated here in Figure 31.



**Figure 31, Basics of spin coating**

After several rounds of spin coating, there should be uniform layer of nanodiamonds on top of the substrate. To inspect this hypothesis, surface is then characterized and analyzed using SEM.

### 7.3 SEM imaging of surface

As mentioned earlier in this chapter, the sputtering is needed to make the sample conductive. This was done by sputtering (with Emitech K100X) a thin platinum layer on top of the ND sample. The Pt layer was approximately 8 nm thick and it was estimated using diagram planned for Au deposition seen in Figure 32. Current used was 30 mA for 1 minute. Sputtering is performed in vacuum of  $5 \times 10^{-2}$  mbar. After the samples are made fully conductive, they can be used in SEM.

Scanning electron microscopy is a technique where a high-energy electron beam is used to interact with the target material. The SEM micrographs were taken using Zeiss Sigma VP, with acceleration voltage of 3 kV and high vacuum of  $10^{-5}$  Pa. The idea is to measure scattered and emitted electrons around the sample to form a micrograph of the target. The sample material needs to be electrically conductive. If the sample is not conductive then the material may become charged by the electron beam and cause interferences during the measurement. Therefore, samples were coated with platinum as mentioned earlier in this chapter.

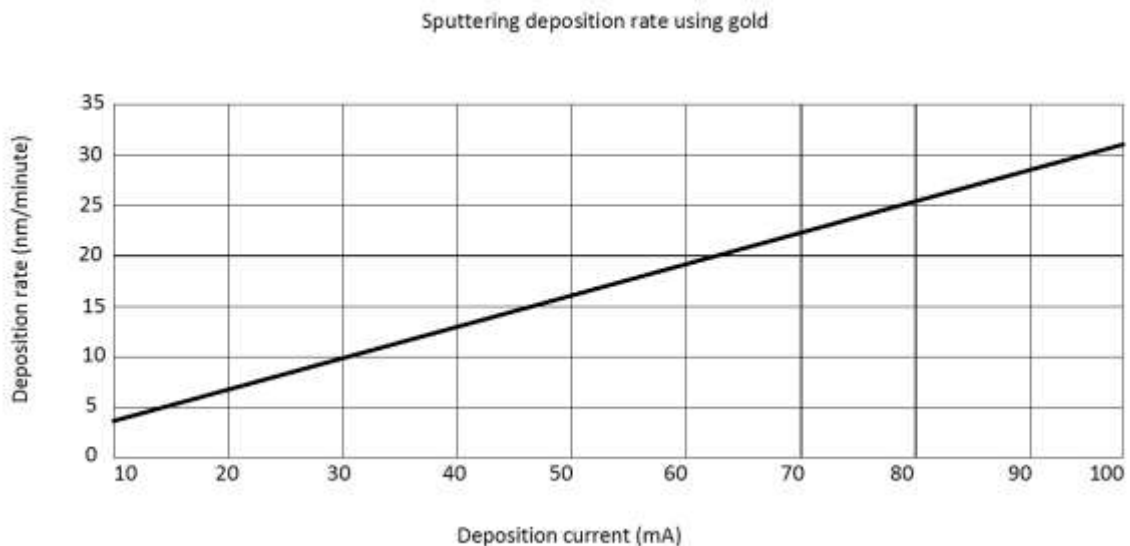


Figure 32, Sputtering diagram for Au deposition rate in respect to deposition current

## 8 Results and discussion

Comparison between different samples coated with ND solutions created in chapter 7 are discussed here. SEM micrographs show the agglomeration on the sample surface and the dispersion properties of different solvents can be analyzed.

### 8.1 Sample 1 – NDs in ethanol

As can be clearly seen from SEM micrographs in Figure 33 and Figure 34, the NDs in ethanol are still very agglomerated even after 2 hours of sonication. Big agglomeration clusters are found all around the sample surface. NDs in this solution did not disperse enough to produce even layer on top of the silicon sample. The agglomeration difference between NDs in ethanol and other samples should not be that dramatic according to literature. In this case, it might have effect on the study that NDs in ethanol had been mixed long time ago when other samples were made during the process and were instantly deposited on sample. NDs in ethanol could have precipitated to the bottom of solution and even 2-hour sonication could not separate them properly. According to related literature and

Table 5, NDs in ethanol is supposed to be the solution where the size of agglomerates is the biggest. However, as mentioned earlier, the size difference should not be as big as it is in this study. At least the agglomeration is clearly shown in comparison to other samples. When observing Figure 33 and Figure 34, it can be seen that agglomerates are very large, from several micrometers to 100 nanometers and no smaller particles can be seen. So, basically all NDs are agglomerated in this sample.

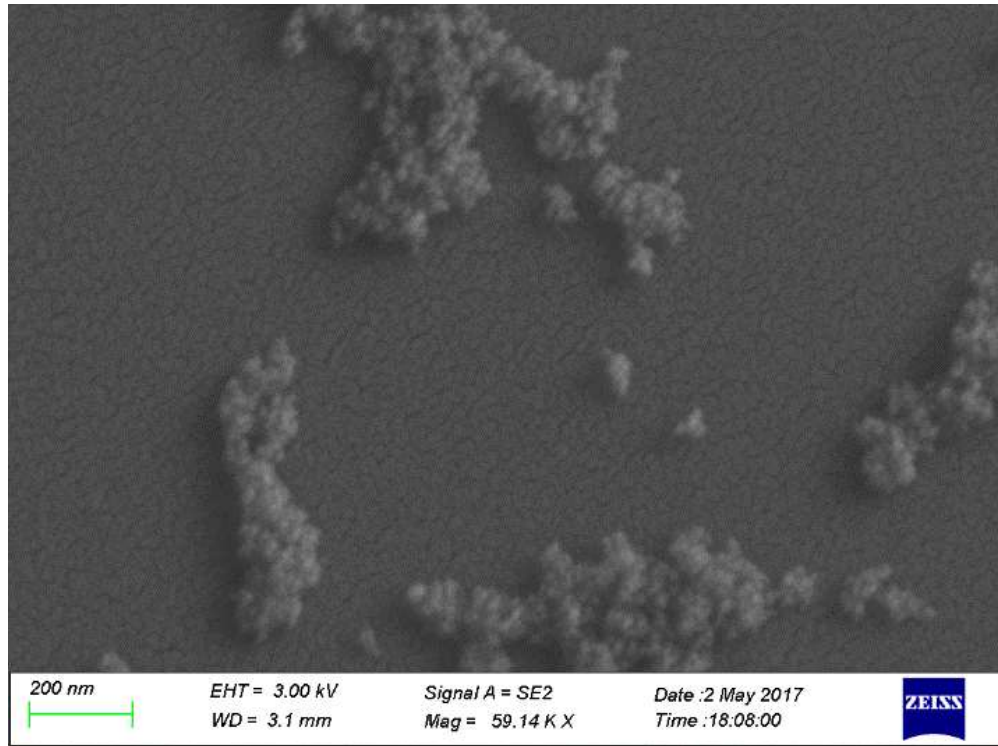
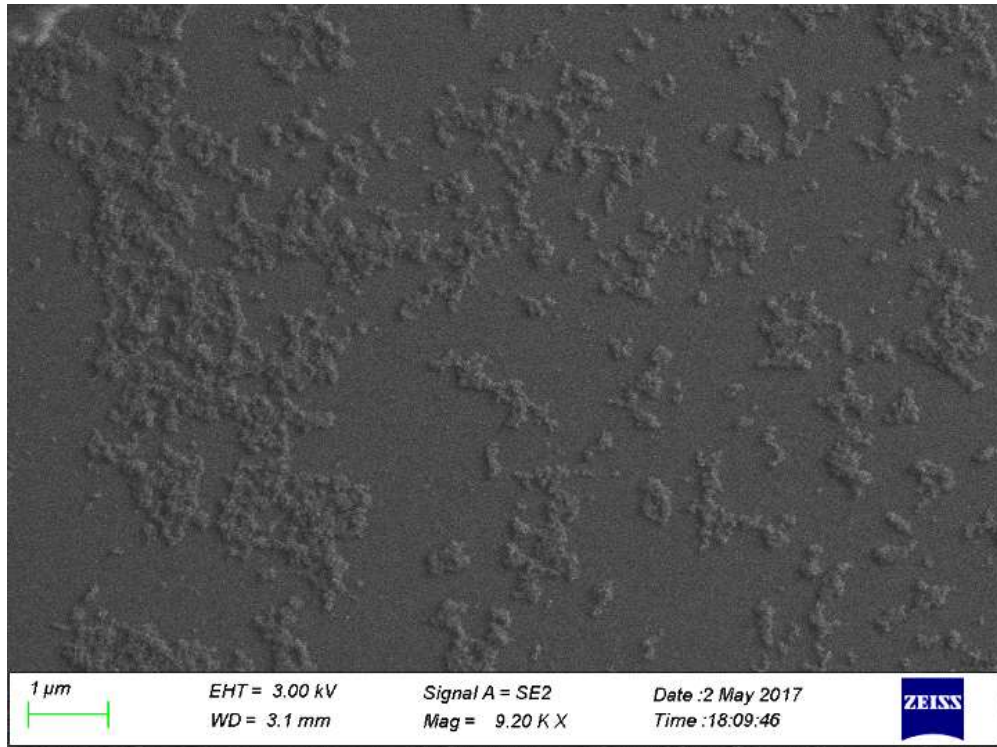


Figure 33, SEM image of NDs in ethanol solution.



**Figure 34, SEM image of NDs in ethanol solution.**

## 8.2 Sample 2 – NDs in DI water

As can be seen from the micrographs taken of NDs in DI water (Figure 35 and Figure 36), there are no large agglomerates visible on the sample and nanodiamonds seem to be more evenly distributed on the surface. However, no single nanodiamond particles can be observed either and almost all NDs are in smaller clusters. So, there is definitely agglomeration present, but not as clearly as in the first sample. Agglomerated clusters seem to be from tens of nanometers to 200 nm. It can be said that DI water is a decent solvent for NDs but smaller agglomerate size is desired in many modern biomedical applications as mentioned earlier in chapter 4.

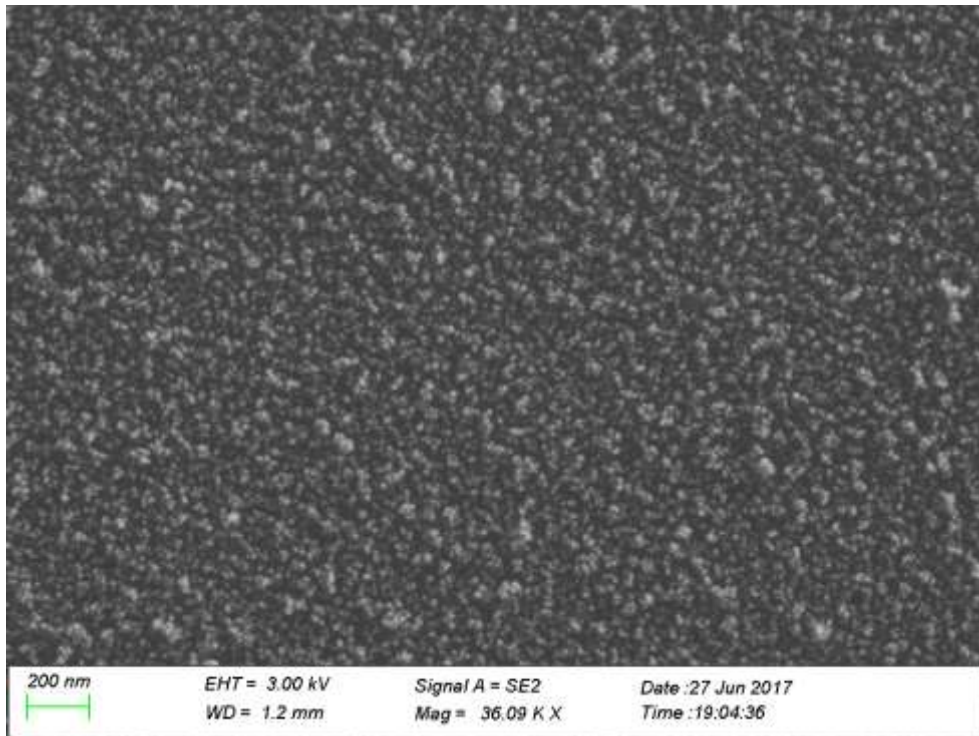
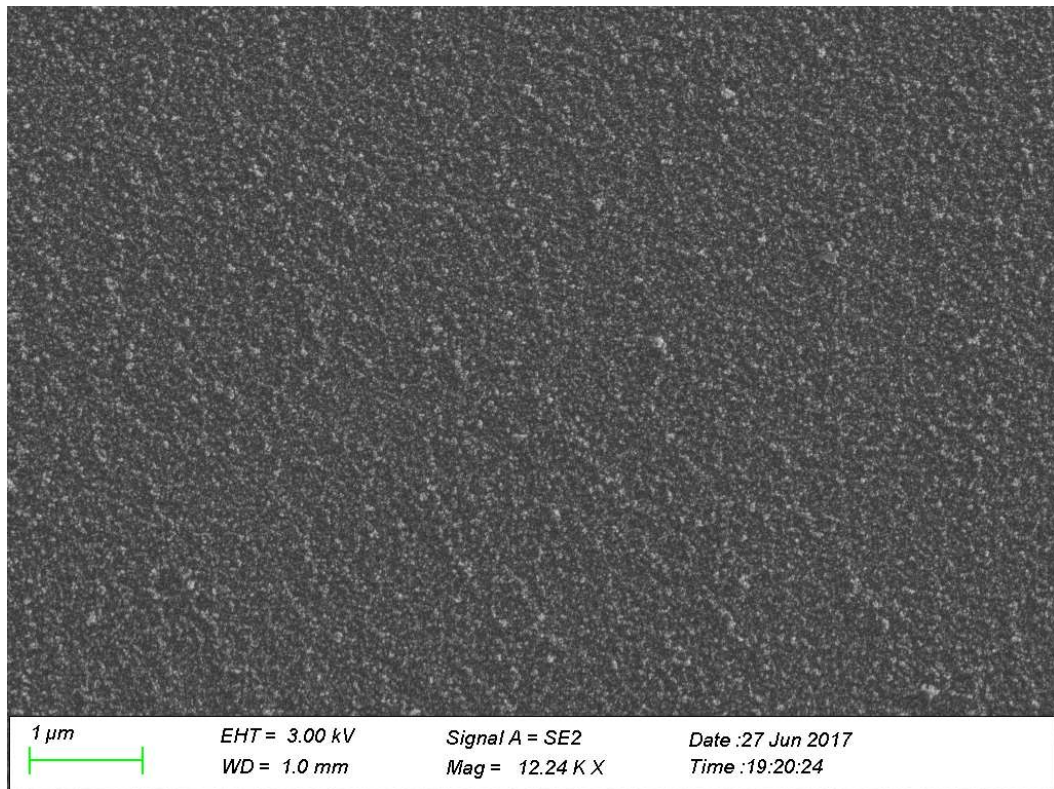


Figure 35, SEM of NDs in DI water





**Figure 36, SEM of NDs in DI water**

### 8.3 Sample 3 – NDs in DMSO

Next micrographs are from nanodiamonds in DMSO solution. The distribution is even better than in the DI water, and only few bigger ( $>100$  nm) agglomerates can be seen. Most of the particles are very small and they could be in clusters of just few or dozen nanodiamonds. Figure 37 and Figure 38 show that DMSO is indeed a good solvent for NDs as literature predicted (87).

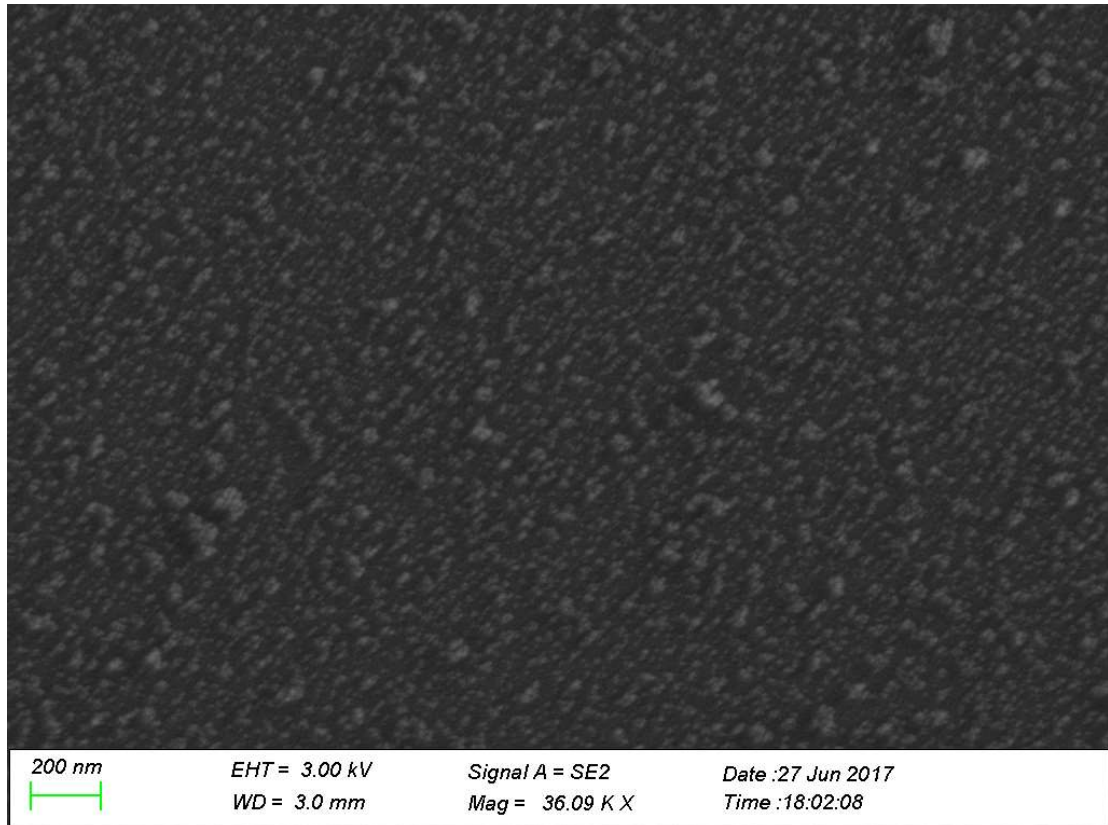
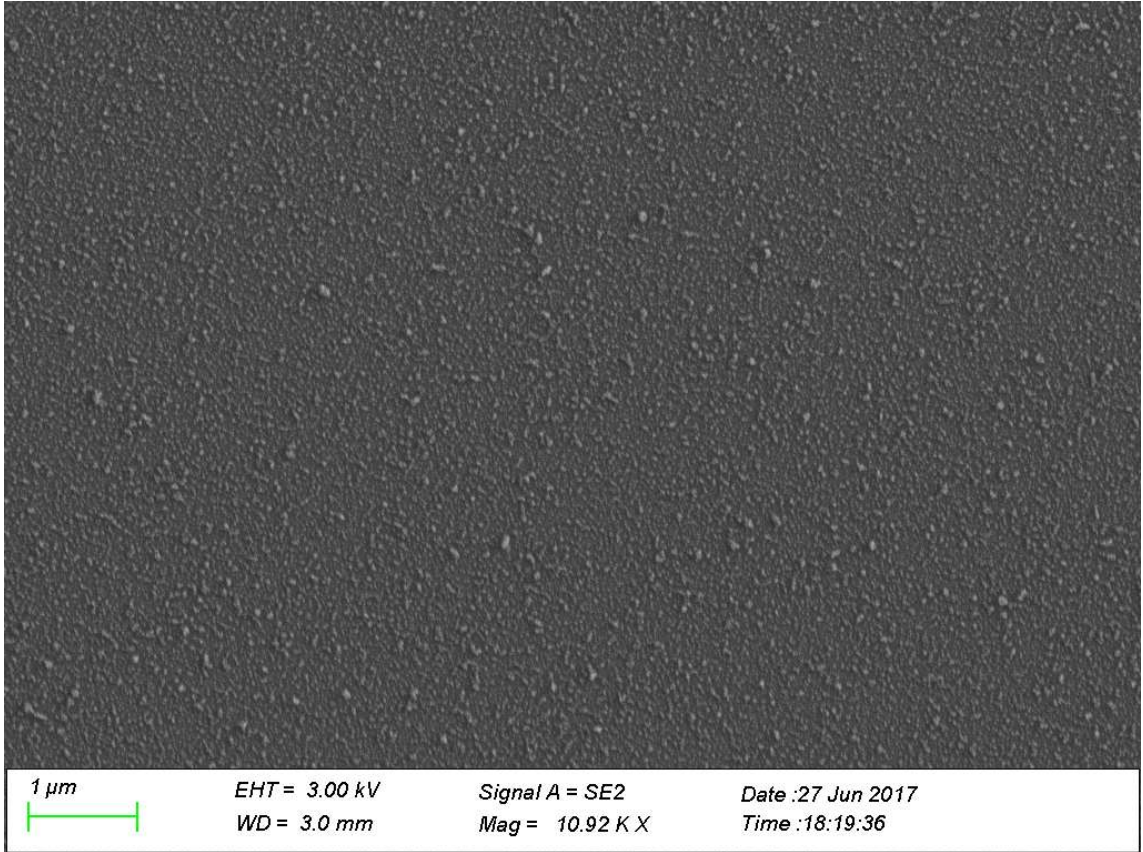


Figure 37, SEM of NDs in DMSO



**Figure 38, SEM of NDs in DMSO**

#### 8.4 Sample 4 – NDs in EG

The last sample was NDs dispersed in ethylene glycol. In this sample, the NDs are very well distributed along the surface. Also, agglomeration rate is very low and the size of agglomerates is small. It is almost possible to see single ND particles on the surface or at least just a few particle clusters can be observed. Larger agglomerates are close to 100 nm but there are not many of them present. This was clearly the best sample when observing agglomerate size and NDs distribution on the sample surface as seen in Figure 39 and Figure 40.

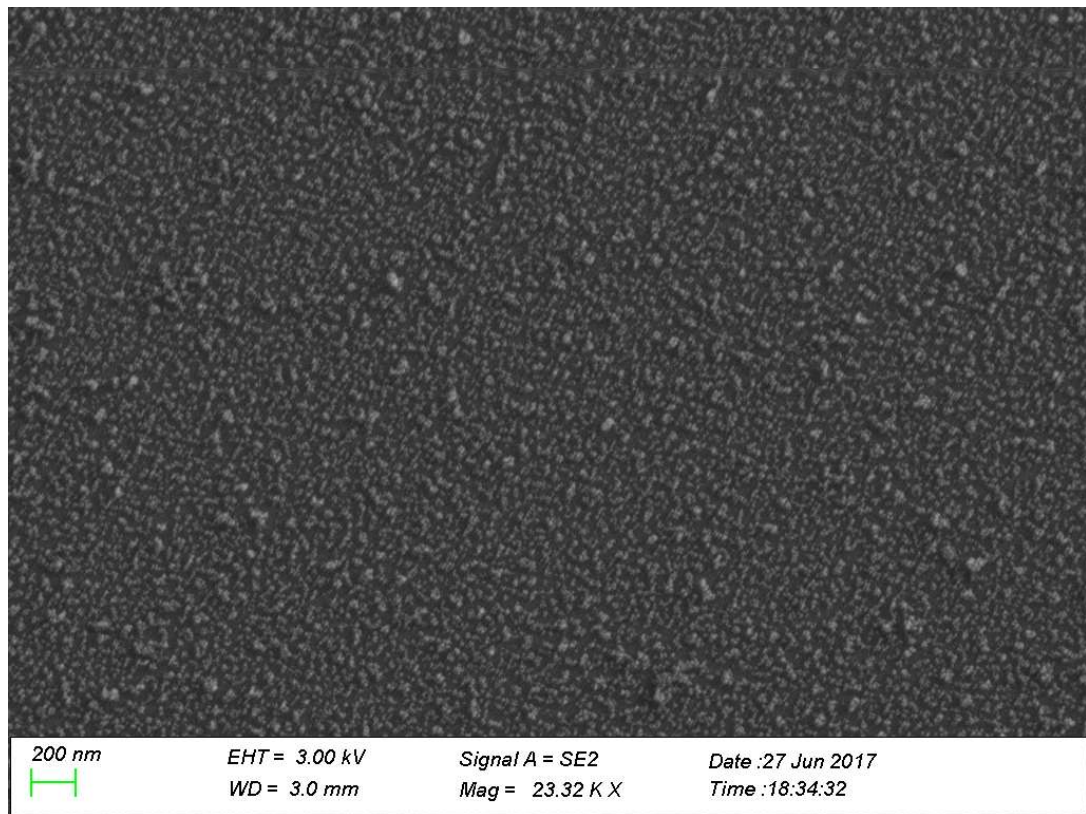


Figure 39, SEM of NDs in EG

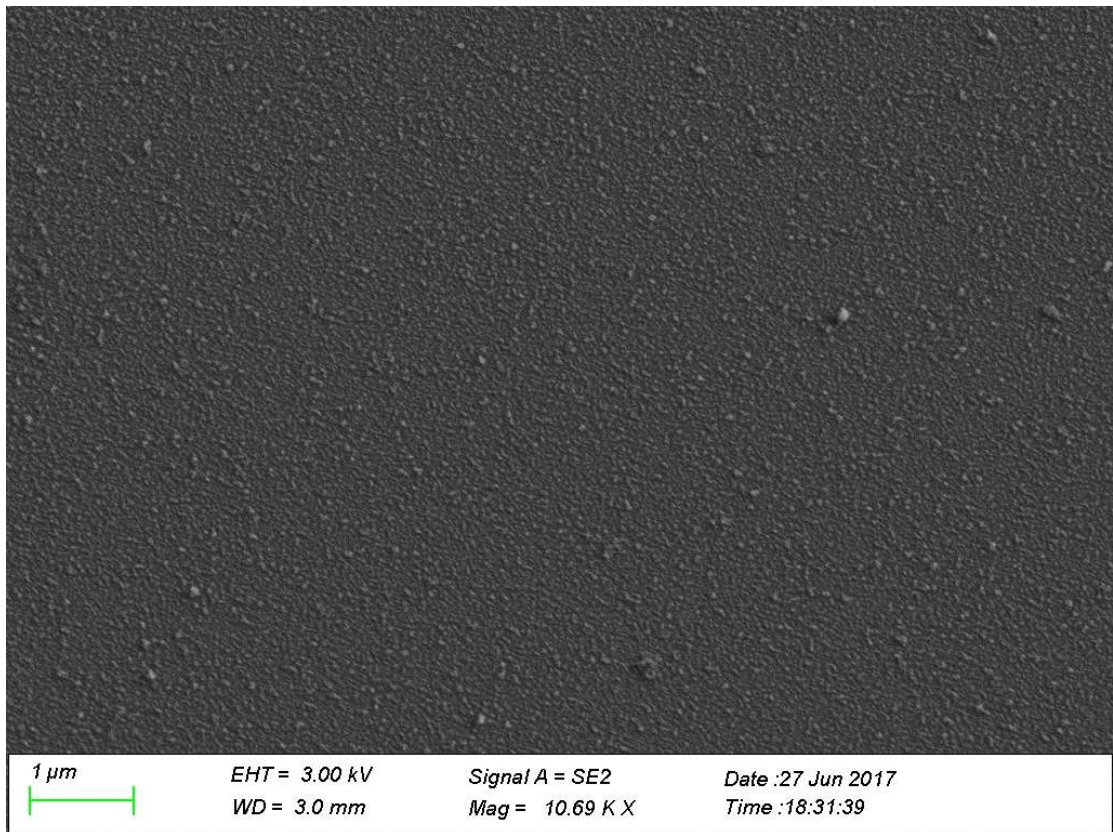


Figure 40, SEM of NDs in EG

## 8.5 Comparison

The results show that the solvent chosen for ND solution does make a big difference. According to this study the EG gave best results and DMSO was not far behind. This sounds like a reliable result when comparing to information found in literature and values seen in Table 5. The NDs dispersed in ethanol might have been sedimented to bottom of the sample and therefore, were not totally comparable to other solutions. Despite the problem with ethanol sample, results were similar than in literature. Other samples had nice smooth ND layer on top of the silicon. Diamond particles were evenly distributed on the surface and there were no spots on sample that lacked NDs completely. Still, in all micrographs agglomeration can be observed. This is because no other deagglomeration methods were available than sonication. Also, it must be noted that deposition method can affect the properties of the ND coatings (73). Spin-coating might cause some additional agglomeration of NDs on the surface, if the layer is not evenly distributed. When inspecting Table 3 in chapter 5.2, it is seen how small ND particles are after different deagglomeration methods. It can be concluded that if single-digit nanodiamonds are wanted, then proper deagglomeration method should be used before dispersing NDs to solvent. Small comparison of ND solvents used and agglomeration observed can be seen in Table 8.

**Table 8, Comparison of ND solvents used in tests**

Solvent	Deagglomeration method	Type of ND	Agglomerate size	Agglomeration rate
EG 99.8%	2h sonication	Hydrogen terminated DND	5 nm - 80 nm	Single ND can be seen
DMSO 99%	2h sonication	Hydrogen terminated DND	10 nm - 100 nm	Almost all ND agglomerated
DI water > 99%	2h sonication	Hydrogen terminated DND	20 nm - 200 nm	All agglomerated
Ethanol 99.5%	2h sonication	Hydrogen terminated DND	100 nm - 5 $\mu$ m	All agglomerated

## 9 Conclusion

In this thesis, it was shown that NDs do form tight aggregates in solution and that this agglomeration can be diminished with the help of a proper choice of solvent. The literature presumed that there are clear differences between dispersion in different solvents and those assumptions were found to be correct. The most suitable solvents for NDs found in this study were EG and DMSO. As high-power sonication was only deagglomeration method used, agglomerates could still be seen in SEM micrographs. Also deposition method can have a significant impact on the properties of the ND coating. It can be said that experimental section worked as ND related literature had predicted. Many more effective deagglomeration methods have been invented to pave the road for applications using single-digit ND particles. These methods such as BASD and beads milling are crucial in the battle against agglomeration. The agglomeration of NDs is mainly caused by surface functional groups and unwanted impurities attached to surface of diamonds. These days ND deagglomeration is mostly handled by vendors, but in many cases the NDs agglomerate again in powder. Therefore, when working with NDs it is important to know how deagglomeration and dispersion methods function. As demonstrated in experimental section in chapter 7 and in results in chapter 8.1, NDs might also form agglomerates in solution when left there for long periods of time without sonication. Therefore, it may not be wise to use NDs in applications if solution's production date is not available because diamond particles tend to sediment to the bottom of solution. However, there are not many publications discussing this issue.

Nanodiamond is definitely an attractive carbon nanomaterial and a candidate for valuable technological applications. It can be produced in bulk quantities, can be functionalized non-covalently and covalently and so far, has not shown many biohazardous effects. (20) Another property of NDs that promotes beneficial effects is its large specific surface area and high affinity for adsorbing proteins, enzymes, and other biological molecules (1). Although ND is proposed for many high impact applications there is still much to understand about the interaction of these nanoparticles with their environment and particularly the solvent. (44). There is a definite need for many of the planned ND applications so the research must be continued. It is important to remember that there are no universal "nanodiamond particles" with well-defined structure and properties as mentioned earlier in chapter 1 (13).

Another area of importance for the fields of ND applications, is the possibility of introducing desired impurity atoms into the bulk of a DND particle and development of purposeful functionalization of the surface of a DND particle (9). This is important because as mentioned earlier, ND surface chemistry defines its properties. One potential functionalized type of diamond is hydrogenated ND. It is a surface-conductive ND, a material with many potential applications in electrochemistry, biomedical sensing, and nanocomposites (59). Hydrogenated ND is also less likely to agglomerate than oxygenated ND.

In the recent years, an explosive growth of interest in the promising areas of research and application of detonation nanodiamonds has happened, an aspect that reflects a general attitude towards technology of carbon nanostructures and their future (9). One area of further research is to identify the surface chemistry that is responsible for the observed redox activity of ND. This includes the study of surface modified ND, with specific surface functionalities either removed or blocked, for example the reduction of all surface C=O groups to C-OH by chemical means. (57) The future applications for nanodiamonds could include carrier in gene therapy, drug delivery, and vaccines. Also, different types of biosensors *in vitro* or *in vivo* may be constructed of ND photonic crystals that change color when targeted molecules bind the sensor surface. (1) In addition, NDs can be used in so called "hybrid" carbon based nanomaterials as mentioned in chapter 4.2. These combination materials have even

better properties than any carbon material itself and can be potential sensor materials in the near future (29).



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## Appendix 1

### **Nanodiamonds used in experimental section.**

Carbodeon uDiamond® Andante specific characteristics of the solid material in dispersion:

Nanodiamond crystal size  $4.2 \pm 0.5$  nm

Nanodiamond content  $\geq 97$  wt. %

Oxidisable carbon content  $\leq 2.5$  wt. %

Metallic incombustible impurity content  $\leq 1.2$  wt. %

Crystal lattice constant  $0.3573 \pm 0.0005$  nm

Carbodeon uDiamond® Andante specific characteristics in the dispersion:

pH stability of dispersion 3-6

Concentration of nanodiamonds in dispersion 5 wt. %

Zeta potential: positive