

Suspending Nanoparticles (hBN and ZnO) in Non-Polar Media by Liposomal encapsulation



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Abstract: Nanoparticles have aggregation tendencies. In order to prevent aggregation and provide isolation from medium and each other, encapsulation is a solution. In this study, two selected nanoparticles, hexagonal boron nitride (hBN) and zinc oxide (ZnO) are suspended in non-polar medium by liposomal encapsulation. TEM and particle size analyses were conducted to characterize the prepared nanofluid containing liposomal nanoparticles. Results showed the stability of prepared liposomes while they stayed intact for more than three months. Prepared hBN liposomes are found to be 270 nm with 0.037 PDI and ZnO liposomes are 380 nm with 0.172 PDI value. Prepared nanofluids were introduced to SAE5W40 engine oil. Suspension stability and lubrication effects of prepared nanofluids in engine oil were examined by turbidimetry method and tribometric tests, respectively. Both hBN and ZnO nanofluids showed low TSI values; they decreased the friction coefficient of original oil by 18.83 and 19.13 percent, respectively.

Keywords: Liposome, Hexagonal Boron Nitride, Zinc Oxide, Encapsulation, Nano additives.

Polar Olmayan Ortamda Lipozom Kapsülleme ile Nanopartiküllerin (hBN ve ZnO) Süspande Edilmesi

Özet: Nanoparçacıkların topaklanma eğilimleri vardır. Agregasyonu önlemek, ortamdan ve birbirinden izole etmek için kapsülleme bir çözümdür. Bu çalışmada, seçilen iki nanoparçacık, altıgen bor nitrür (hBN) ve çinko oksit (ZnO), polar olmayan ortamda lipozom kapsülleme ile süspande edilmiştir. Hazırlanan nanoakışkan içeren lipozomal nanopartikülleri karakterize etmek için TEM ve partikül boyutu analizleri yapılmıştır. Sonuçlar, hazırlanan lipozomların üç aydan fazla bir süre bozulmadan kaldıklarını göstermiştir. Hazırlanan hBN lipozomları 0,037 PDI ile 270 nm ve ZnO lipozomları 0,172 PDI değeri ile 380 nm olarak bulunmuştur. Hazırlanan nanoakışkanları SAE5W40 motor yağına katılmıştır. Hazırlanan nanoakışkanların motor yağı içerisindeki süspansiyon kararlılığı ve yağlama etkileri sırasıyla türbidimetri yöntemi ve tribometre testleri ile incelenmiştir. Hem hBN hem de ZnO nanoakışkanları düşük TSI değerleri göstermiştir; bu nanoakışkanlar, orijinal yağın sürtünme katsayısını sırasıyla yüzde 18,83 ve yüzde 19,13 oranında azaltmıştır.

Anahtar Kelimeler: Lipozom, altıgen bor nitrür, çinko oksit, kapsülleme, nano katkı maddeleri.

ARAŞTIRMA MAKALESİ

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1.Introduction

Nanoparticles are prone to aggregate in the medium to which they are added. They can be isolated from media by different methods in order to be prevented from being aggregation. One of these methods is encapsulation of nanoparticles and by this way, forming steric or electrostatic hindrance around the nanoparticles (Pan et al., 2017; Yu & Xie, 2012). Encapsulation, by forming an outer wall to nanoparticles, does not only prevent aggregation but also makes it possible to adapt different media with different chemical properties, for example non-polar media (Sahil et al., 2011). Liposome is a convenient structure to isolate or encapsulate nanoparticles. Liposomal encapsulation can be explained as encapsulating any material with phosphatidyl choline (PC). PC is a class of phospholipids that can be found in natural substances such as eggs, soybeans, and sunflower. Unique chemical structure of it enables bilayer formation when forming liposomes. There are many examples where active materials or nanoparticles are encapsulated by liposomal structures (Al Sawaftah & Husseini, 2020; Gregoriadis & McCormack, 2005; Hong et al., 2020; Jo et al., 2020; Kakami et al., 2019; Shehzad et al., 2014).

In previous study of our team, nonanol is observed to be compatible with non-polar medium (Sahil et al., 2011). In this study, nanoparticles are encapsulated by liposomal structure in nonanol. Using nonanol as liposomal preparation medium shows two advantages. First of all, nonanol makes the liposomal structure more stable; secondly, it enables the liposomal structures formed to be suspended in non-polar media. Nonanol is used as a stabilizer in conventional liposomal preparation methods while it enhances the stability effect (Sahil et al., 2011). In addition, since nonanol has high KOW (octanol/water partition coefficient) value, high lipophilic chemical structure, outer surface of the prepared liposome becomes adaptable to non-polar media.

Two selected nanoparticles are encapsulated by liposome in this study, namely zinc oxide (ZnO) and hexagonal boron nitride (hBN). They are known to have lubrication properties (Celik et al., 2013); however, it is a challenging requirement to suspend them in nonpolar media. Researchers have tried to suspend them in non-polar media. For example. Amiruddin et al. (2015) examined the suspension stability with respect to pH value in their proceeding here they reported the dispersion stability of SAE15W40 enriched with hBN nanoparticles for 60 days. After publishing this work, they dispersed the same nanofluid with 70 nm sized hBN nanoparticles dispersed by ultrasonication in SAE15W40 with oleic acid as surfactant (Chua et al., 2016). In that study, pH effect to hBN suspension was examined where at pH 9, hBN suspension was stable for 30 days and at pH 11 and 13 the nanofluid suspended in engine oil for 60 days. pH values which is far from neutral value, increases the electrostatic forces in the solution; therefore, in such solutions, nanoparticles tend to remain suspended for longer periods of time. Nasser et al. (2020) also studied hBN to suspend it in a non-polar medium. They prepared a nanofluid of hBN and phosphonium ionic fluids and examined the synergistic effects of these nanoparticles. The nanofluid was prepared by first mixing the ionic liquid and hBN in a mortar and then they are added to PAO 32 and processed by ultrasonication. Prepared samples were characterized by sedimentation photography for suspension stability. They reported that the nanofluid stayed suspended for 60 days for one sample and for more than eight months for another sample. Prepared nanofluid decreased both the friction coefficient and wear. Although studies about hBN suspension in non-polar media were very successful, they should be diversified and other suspension methods also could be applied.

There are also several studies for suspending ZnO in various media. However, suspension stability and suspension durations of reported studies should be extended. For instance, Antolin Hernández Battez et al. (2006) used ZnO as lubrication additive. Since 2006, his team published four studies related to lubrication

effect of ZnO. First of them was to investigate the tribological effect of prepared nanofluid of ZnO (Hernández Battez et al., 2006). They used polyhydroxystearic acid as dispersing agent and they dispersed the ZnO in PAO6. They did not characterize the suspension stability of the prepared nanofluid in detail, but they measured the decreasing effect of ZnO on wear. In another study of Hernández Battez's team as lubrication additive, ZnO nanoparticles were compared with other nanoparticles, CuO and ZrO₂ (Hernández Battez et al., 2008). Nanoparticles were added to PAO6 in different ratios and they were dispersed into base oil by ultrasonication. All nanoparticle additions to base fluid were reported to have reduction on friction coefficient and wear. In another study, Bhaumik et al. (2018) searched for green alternative to commercial mineral oils. They preferred castor oil as an alternative for mineral oils and they improved lubrication properties of the castor oil by adding ZnO. ZnO nanoparticle was dispersed in castor oil by means of ultrasonication. Suspension stability of the prepared nanofluid was not given throughout the study. ZnO increased the viscosity of castor oil and boosted the formation of tribofilm on the rubbed surfaces; hence, decreases the friction coefficient. Elagouz et al. (2020) used oleic acid to disperse various amounts of ZnO nanoparticles in fully formulated 10W40 oil. They reported the preparation method of ZnO nanofluid as mechanical stirring. They characterized the suspension stability of prepared sedimentation photography and by spectrophotometry. After four days, UV absorption of prepared nanofluid began to decrease which means unstability began after four days from preparation. They also investigated the tribological behavior of the prepared nanofluid. It was stated that both the friction coefficient and wear are decreased by addition of ZnO nanoparticle into the medium. There are also recent developments about the usage of ZnO as lubrication additive. In 2020, Vardhaman et al. (2020) designed hybrid lubrication system by using both MWCNT and ZnO. In this study, ZnO and MWCNT nanoparticles were added in varying ratios to fully formulated 10W40 engine oil where oleic acid was used as dispersant. They dispersed all ingredients in oil by ultrasonication. Addition of nanoparticles decreased the friction coefficient and wear volume of original engine oil. To conclude, hBN and ZnO are known with their lubrication properties and they are studied to suspend in non-polar media. There are successful suspension studies; however, methods used and suspension durations should be diversified and extended.

This study is part of the study of suspending inorganic nanoparticles in non-polar media. Our previous work in this context was suspending nano-boric acid in non-polar media by two-step method (Tanrıseven et al., 2020). In this study, hBN and ZnO nanoparticles will be encapsulated by liposomal structure and their suspension stability and lubrication properties will be tested. TEM characterizations and particle size analysis were conducted in order to characterize the prepared liposomes. In addition, suspension stability tests conducted by turbidimetry method and lubrication effects of prepared liposomes were conducted by tribometer tests. To the best our knowledge, this study is the first that suspend nanoparticles in an engine oil by liposomal encapsulation method.

2. Materials and Methods 2.1 Preparation of liposomal hBN and

2.1 Preparation of liposomal hBN and ZnO and their addition to engine oil

Two different nanoparticles were entrapped in liposomes in this study. Hexagonal boron nitride (hBN) nanopowder was supplied from BORTEK (Istanbul, Turkey) and zinc oxide (ZnO) nanopowder was procured from EgeKimya (Izmir, Turkey). Both nanopowders are used without any further purification. As phosphatidylcholine source, P45 product of Lipoid was used. P45 is standardized soybean lecithin with 45% phosphatidylcholine from non-genetically modified plants. Liposomal preparation medium is selected to be a fatty alcohol, namely nonanol, which was supplied from Merck.

0.05 g of nanopowder was weighed and put in a glass vial and then



0.25 g of P45 was added to the nanopowder and finally 99.7 g of nonanol was added to the vial and then ultrasonic treatment was applied for 15 minutes. In this step, Hielscher UP 400St instrument was used at a constant frequency of 24 kHz for 20 minutes at 50% amplitude. The preparation of liposomes is graphically illustrated in Figure 1. After preparation of liposomes, nanoparticles that was not encapsulated in liposome were removed by means of centrifuging. For this purpose, Hettich Universal 320 centrifuge was operated at 5000 rpm for 20 minutes.

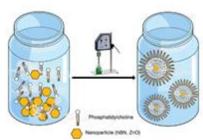


Figure 1: Graphical representation of nanoparticle-entrapped liposomal preparation.

Prepared nanofluids; i.e., liposomal hBN and ZnO, are added to SAE5W40 fully formulated engine oil. Both ZnO and hBN nanofluids were introduced by 3%.

2.2. Characterization of prepared nanofluids

Prepared liposomes were characterized by Malvern Nano ZS Zeta Sizer instrument for particle size distribution analysis and TEM for visualizing the entrapment of nanoparticles by liposomal spheres. Suspension stability measurements were conducted by means of Turbiscan Tower(R) instrument. In order to obtain TEM micrographs, a Hitachi High Tech HT7700 instrument was used. TEM characterization was conducted at DAYTAM (Erzurum, Turkey).

2.3. Effects of the nanofluids on engine oil friction coefficient

Tribometric tests were performed to observe how the hBN and ZnO nanofluids affect the friction coefficient of SAE5W40, fully formulated oil. For this purpose, a special tribometer (UTS Tribometer T30M-HT® with a reciprocating test module) was used. The tests were performed according to the ASTM G133 using ball-on-plate configuration where an 8 mm ball made of 100Cr6 and a steel plate (12 mm x 15 mm x 3 mm in dimensions) made of hardened 4140 steel with the hardness of 52 HRC were used. The surfaces of the plates used were polished carefully after machining to be able to have mirror-like surfaces. Fresh plate surface was used for each test. For each condition, at least three tests were performed to be sure of repeatable results. Friction tests were conducted with five drops of sample on the plate. The tests were made with 8 mm stroke and 2 Hz frequency for a total distance of 32 m at room temperature.

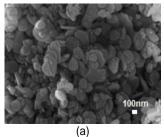
3. Results and Discussion

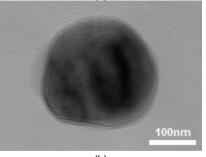
Two nanoparticles (hBN and ZnO) were passed through the steps suggested above for liposomal preparation in order to be encapsulated inside stable liposomal spheres. Prepared nanofluids and nanopowders were characterized. Afterwards, nanofluids will be added to fully formulated oil and suspension stability and lubrication effects of these nanofluids will be tested. In this part of the study, detailed results and discussions of these characterizations and tests will be shared.

3.1. Liposomal hBN nanoparticles

In this study, nanoparticles will be encapsulated into stable liposomal spheres and they will be suspended in non-polar media.

The encapsulated nanoparticles are isolated and became suitable to introduce to non-polar media. In this part of the study, the details of the successful inclusion of the hBN nanoparticle into the liposomal sphere and its characterization with the help of TEM, SEM and Zeta Sizer instrument will be discussed.





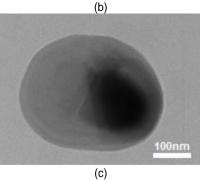


Figure 2: (a) SEM micrograph of hBN nanopowder, (b) and (c) TEM micrograph of hBN trapped liposomes.

Its morphology and particle size can be seen in the SEM micrograph of the hBN nanopowder (Figure 2). Size of hBN nanoparticles was determined as between 100 nm and 200 nm based on this micrograph. Average size of liposomal structure is determined as 270 nm from TEM micrograph. TEM micrographs shown in Figure 2 (b) and (c) were taken after more than three months from liposomal preparation. Intact liposomal structure ensures the fact that prepared liposomes are stable more than three months. TEM micrographs in Figure 2 clearly show liposomes containing hBN nanoparticle. The basic principle of TEM characterization allows us to visualize the interior of the structures in a detailed form. Taking advantage of this benefit of TEM characterization, we have viewed the content of the liposome. Figure 2 (b) and (c) show a liposomal sphere entrapping a single hBN nanoparticle. Encapsulation of hBNby stable liposomal spheres was demonstrated by the TEM micrograph. Encapsulation of hBN by liposomal structure in nonanol provides hBN nanoparticles to be easily dispersed in non-polar media. hBN nanoparticle became non-aggregated by isolation from medium and other nanoparticles by encapsulation with a stable liposome wall. This form of hBN can be easily dispersed in lipophilic media. Another important point that can be interpreted from the Figure 2 is that the 200 nm size of hBN nanopowder detected by SEM images matches the size of the nanoparticle imprisoned in the liposome in TEM images. In addition, aggregated hBN nanoparticles in the SEM micrograph are separated and isolated individually from the medium by liposomal sphere.

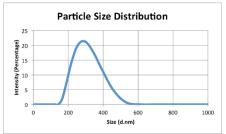


Figure 3: Particle size distribution analysis of hBN liposomal nanofluid.

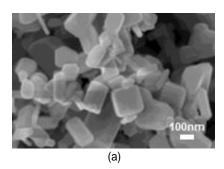
Figure 3 shows particle size distribution test result. In this result, there is one peak at 270 nm with 0.037 PDI. The particle size distribution of liposomes in Figure 3 implies that the liposomal structures prepared in nonanol are formed by a single particle size. In this wide range of particle size analysis (0-1000 nm range), only one peak observation indicates that 100% of the liposomes formed at size of 270 nm. This result also points to the same particle size as the liposomal structures in the TEM photographs seen in Figure 2(b) and (c). Thus, all the characterization results were verified by ensuring each other.

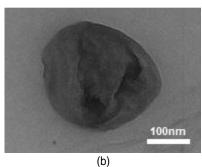
The hBN nanoparticle, which appeared in this part of the study, was successfully trapped inside the liposomal structure. hBN trapped liposome remains in its expected spherical shape for more than three months. The effect of nonanol, a selected type of fatty alcohol, on known liposomal stability was again revealed by this study (Sahil et al., 2011).

Although hBN is known as a lubrication additive, it collapses when it is added to engine oils (Chua et al., 2016); liposomal encapsulation prevents this collapse by isolating the nanoparticle from the media. In addition, outer wall of prepared liposomal hBN enables to be dispersible in oil media. When hBN nanofluid was added to fully formulated oil SAE5W40, suspension stability and the lubrication effect of prepared nanofluid were tested by turbidimetry method and tribometry, respectively.

3.2. Liposomal ZnO nanoparticles

In this part of the study, the characterizations of ZnO liposomes will be discussed in detail. ZnO liposomes were prepared by the proposed method applying the same method.





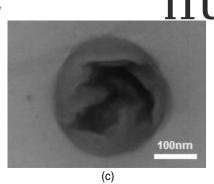


Figure 4: (a) SEM micrograph of ZnO nanopowder, (b) and (c) TEM micrograph of ZnO trapped liposomes.

SEM micrographs in Figure 4 show the structure of ZnO nanopowder and particle size is determined as approximately 100 nm on average. TEM micrographs in Figure 4 (b) and (c) show the average size of the liposomal spheres as 380 nm. Like in the case of hBN nanoparticle, TEM micrographs of ZnO were also taken after three months from preparation. Intact liposomal spheres, including ZnO nanoparticles in them, shows high stability of prepared liposomal structure. SEM and TEM micrographs (Figure 4 (b) and (c)) together reveal that several ZnO nanoparticles share one liposomal sphere. Liposomes prepared for hBN nanoparticle was 270 nm in size and ZnO nanoparticles of approximately 100 nm in size. Prepared liposomal size is capable of inclusion of more than one ZnO nanoparticles to occupy in each liposomal sphere. More than one ZnO nanoparticles placement in a liposomal structure caused a slightly larger liposomal size than hBN liposomes; where hBN liposomes are 270 nm in size, ZnO liposomes are 380 nm (Figure 2 and Figure 4). Another reason for the ZnO nanoparticle being trapped in the liposomal structure in more than one nanoparticle may be the chemical structure of the ZnO nanoparticle. The chemical structure of the ZnO nanoparticle may have caused multiple ZnO nanoparticles to enter a liposomal sphere and form liposomes of 380 nm size.

Both ZnO and hBN nanoparticles have been encapsulated by liposome successfully even though they have two different chemical structures.

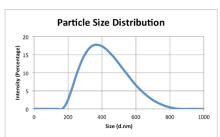


Figure 5: Particle size distribution analysis of ZnO liposome nanofluid.

Particle size distribution measurement of liposomes containing ZnO is similar to the result of hBN liposomes. Figure 5 shows that prepared liposomes are formed only in one size. 100% of the prepared liposomes are 380 nm with 0.172 PDI value.

Successful preparation of ZnO liposomes can be used in many applications. One of these areas is nanolubrication. Although ZnO nanoparticles have lubrication property, when they are alone nanoparticles are collapsed in engine oil medium (Wu et al., 2016). By means of proposed mechanism, ZnO liposomes were dispersed in SAE5W40 fully formulated oil and the lubrication effects of nanofluids were tested by tribometer.

3.3. Suspension stability characterization of nanofluid-added engine oil

Prepared nanofluids were added to SAE5W40 engine oil by 3%.





After addition, suspensions were characterized by turbidimetry method, which was explained in detail in previous study of our team (Tanrıseven et al., 2020).

TSI values related to these analyses were given in the Table 1. Low TSI values show the fact that liposomal encapsulation of hBN and ZnO provide stable suspension of these nanoparticles in non-polar media.

Table 1: Turbiscan Stability Index (TSI) values of engine oil with nanofluids.

| Sample Name | Turbiscan Stability Index (TSI) |
|-----------------------------------|---------------------------------|
| hBN nanofluid added engine oil | 1.2 |
| ZnO nanofluid added engine oil | 4.3 |

3.4. Tribometer tests of nanofluid added engine

hBN and ZnO nanoparticles are well-known lubrication additives. They are studied to suspend in non-polar media like engine oils (Wu et al., 2016). To the best our knowledge, this study is the first to make liposomes of nanoparticles to suspend them in engine oil and determine their lubrication properties. In fact, liposomal encapsulation methods in the literature are very expensive and contain several high-cost methods; therefore, encapsulation of a nanoparticle by liposome to use it as engine oil additive may be very expensive. However, proposed method in this study suggests a single step and relatively low-cost method to prepare nanofluids containing liposomal nanoparticles that are suitable to suspend in non-polar media.

Prepared nanofluids are also tested for their effect on friction coefficient.

Figure 6 and Table 2 shows the effect of prepared nanofluids on tribological properties. They affected friction coefficient in a very good manner. Liposomes have three lubrication properties on one compound. Liposomal nanoparticles are spherical, they are surrounded by surfactant and they contain nanoparticles that are known with their lubrication effects. These three lubrication effects of prepared nanoparticles showed significant decrease on friction coefficient value of fully formulated engine oil.

Table 2 and Figure 6 shows the friction coefficients of nanofluids added and non-added nanoparticles. Blue lines belong to non-added engine oil and red lines belong to nanofluids added to fully formulated engine oil. Figure 6 clearly shows the fact that adding nanofluids decrease the friction coefficient of original oil. Table 1 shows the numeric values of the graphs. hBN and ZnO nanofluids decreased friction coefficient by 18.83 and 19.16%, respectively. Spherical shapes of liposomes and intrinsic lubrication effects of hBN and ZnO can explain these huge decreases.

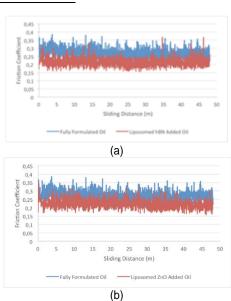


Figure 6: Friction coefficient value graphs of engine oil alone and engine oil with addition of (a) hBN and (b) ZnO nanofluids with respect to sliding distance.





Table 2 Friction coefficient values of the fully formulated engine oil with and without hBN and ZnO nanofluid additions. The reduction values with nanofluid additions were also inserted in the table.

| Sample Name | Friction Coefficient Value | Percent Reduction on Friction Coefficient Values by | |
|-------------------------------------|-------------------------------|---|---------------------|
| | | Average | Nanofluid Additions |
| Fully Formulated SAE5W40 Engine Oil | 0.2761 | | |
| | 0.2755 | 0.2761 | - |
| | 0.2768 | | |
| Engine oil, with hBN nanofluid | 0.2237 | | |
| | 0.2251 | 0.2241 | -18.83 |
| | 0.2236 | | |
| Engine oil, with ZnO nanofluid | 0.2235 | | |
| | 0.2231 | 0.2232 | -19.16 |
| | 0.2231 | | |



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