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Microscale Tribological Behavior and In Vitro Biocompatibility of Graphene Nanoplatelet Reinforced Alumina

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Abstract

Graphene nanoplatelets were added as reinforcement to alumina ceramics in order to enhance microscale tribological behavior, which would be beneficial for ceramic-on-ceramic hip implant applications. The reduction in microscale wear is critical to hip implant applications where small amounts of wear debris can be detrimental to patients and to implant performance. The addition of the GNPs lead to improvements in fracture toughness and wear (scratch) resistance of 21% and 39%, respectively. The improved wear resistance was attributed to GNP-induced toughening, which generates fine (~100 nm) microcracks on the scratch surface. In addition, active participation of GNPs was observed in the scratch subsurface of GNP-reinforced samples through focused ion beam sectioning. Friction coefficients are not significantly influenced by the addition of GNPs, and hence GNPs do not act as solid state lubricants. *In vitro* biocompatibility with human osteoblasts was assessed to evaluate any possible cytotoxic effects induced by GNPs. Osteoblast cells were observed to survive and proliferate robustly in the GNP-reinforced samples, particularly those with high (10 -15 vol.%) GNP content.

Keywords: Graphene Nanoplatelets, Ceramic Matrix Composites, Nanotribology, Microscratch, Osteoblasts, Biocompatibility

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1.0. INTRODUCTION

Advances in ceramic processing technology and the advent of ceramic matrix composites (CMCs) have led to the clinical application of ceramic knee joint replacements and ceramic-on-ceramic hip implants for total hip arthroplasty (THA).¹⁻⁴ Ceramic-on-ceramic (CoC) hip implants have proven to have superior performance over polymer, metal, and polymer-metal hip implants.^{1,2,5,6} Ceramic hip implants utilize alumina or alumina matrix composites and hence have excellent strength, scratch resistance, wettability, and biocompatibility.^{1,2,4,6} Despite these advantageous properties, issues still exist related to catastrophic fracture and abrasive wear debris.⁴⁻⁸ The low fracture toughness of alumina can cause large debris chips to form. This hard debris acts as a third-body abrasive that can induce osteolysis, which requires corrective procedures or replacement of the implant.⁵ If a large enough flaw is initiated by mechanical loading during service, catastrophic failure can occur via rapid crack propagation. There is hence a continued drive to develop biocompatible ceramic matrix composites with enhanced fracture toughness and wear resistance.⁹⁻¹¹

Over the last several years the incorporation of graphene nanoplatelets (GNPs) into ceramic matrix composites has been shown to improve mechanical damping,¹² flexural strength,¹³⁻¹⁵ fracture toughness,¹⁵⁻¹⁹ and biocompatibility.²⁰⁻²³ Several studies have also recently investigated the macroscale sliding wear behavior of GNP-reinforced ceramics.²⁴⁻³² The addition of GNPs has been seen to enhance wear resistance by up to 56%²⁵ and most studies have also reported a decrease in the coefficient of friction (CoF).^{25,27-29,32} There are two proposed mechanisms in the literature for the enhancements in wear resistance and CoF in GNP-CMCs. Belmonte et al.²⁵ were the first to propose that the beneficial effects of

GNPs on tribological performance were due to the formation of a thin GNP-rich lubricating tribofilm. The layer is believed to form due to the exfoliation of graphene layers from GNP; Scanning electron microscopy and raman spectroscopy have provided evidence for the presence of thin graphene platelets on the wear track surfaces.^{25,31} Kim et al.²⁹ achieved an order of magnitude improvement in wear resistance, yet they do not report the formation of a lubricating tribofilm. Instead they have proposed that the improvements in wear resistance and CoF are due to intrinsic lubrication provided by the GNP distributed at the grain boundaries of the alumina matrix.

In the present study, the microscale scratch (wear) behavior of GNP-reinforced alumina ceramics was investigated. The understanding of nano and microscale wear is critical to understand because most surface wear phenomena originate at small length scales. As nano and microscale asperities are formed, this leads to increased roughness of the surface, which in turn leads to higher macroscale friction. Microscale asperities can also become sites for the initiation of cracks, which can propagate rapidly in brittle ceramic systems. In addition, in a system as delicate as the human body it is critical to minimize the amount of even submicron-sized wear debris from the hip implant. Submicron debris from hip implants has been known to migrate to the liver, spleen, and lymph nodes where they induce inflammatory responses and are difficult to detect.³³ Alumina wear debris can also reduce the hip implant lifetime by quickly degrading the softer polymer and metal components of the hip implant.⁵⁻⁷

The primary objectives of the present study were: i) to investigate the intrinsic tribological behavior of Al₂O₃-GNP nanocomposites at load and length scales relevant to the initial wear stages in hip implant applications, and ii) to elucidate the intrinsic wear

mechanisms in GNP-reinforced ceramic nanocomposites. In addition, the micro- and nanoscale mechanical properties **were** evaluated in order to fully understand the wear behavior. Also, the feasibility of applying Al₂O₃-GNP nanocomposites as hip implant materials **was** evaluated by investigating their *in vitro* biocompatibility with human osteoblast cells. High volume fractions of GNPs have been utilized because some of the greatest improvements in toughness in GNP-CMCs have been attained with GNP concentrations as high as 5 - 7 vol.%.^{15,17,18,22} High GNP contents **were** also utilized in order to ensure that GNPs are present in the small volumes tested and to accentuate the effects of GNPs on biocompatibility. The Al₂O₃-GNP nanocomposites demonstrate retention of biocompatibility, and the addition of GNPs leads to the enhancement of fracture toughness and wear resistance by up to 21% and 39%, respectively. The development of biocompatible GNP-CMCs could lead to next generation hip implants with increased lifetimes and reduced instances of catastrophic failure.

2.0. MATERIALS AND METHODS

2.1. Materials

The starting alumina powder **was** obtained from Sumitomo Chemical (AES-11 grade, Tokyo, Japan) and consisted **of** submicron (~0.45 μm) α-alumina particles with an average grain size of 0.3 μm. The starting graphene nanoplatelets (GNPs) **were** synthesized by Applied Carbon Nanotechnology (Pohang City, Republic of Korea) and **had** starting lateral dimension of ~5 μm and an average thickness of 6-8 nm. Details on the preparation of the nanocomposite powders and their consolidation can be found in our previous work.³⁴ Briefly, the GNP powder **was** ball milled in order to exfoliate platelets and relieve

agglomeration. Ball milling provides sufficient shear forces to relieve agglomeration that cannot be achieved with ultrasonication alone. The GNPs were ball milled separately from the alumina powder in order to prevent additional damage that would be induced by the presence of the hard alumina particles. The ball milled GNP powder was then ultrasonicated in an alcohol solution and subsequently the alumina powder was gradually introduced until achieving the desired composition. Compositions containing 5 vol.% (A-5G), 10 vol.% (A-10G), and 15 vol.% (A-15G) of GNPs were synthesized by this method. The alumina and nanocomposite powders were packed into 20 mm graphite dies and consolidated using a DR. SINTER SPS-825 S (Fuji Electronic Industrial Co., Kawasaki, Japan) spark plasma sintering furnace. The samples in the present study were processed using a pressure of 90 MPa, a dwell temperature of 1500 °C, and a dwell time of 5 min. A heating rate of 100 °C/min was used and the chamber was kept in a vacuum with a residual pressure of ~3-4 Pa.

2.2. Microstructural Characterization

The density of the sintered compacts was assessed using the Archimedes method. Relative densities were calculated using the manufacturer's density specification for alumina powder (3.93 g/cm³), published density values for GNP powder (1.81 g/cm³)²⁴ and the target GNP volume percent values. It is conceivable that during consolidation, functional groups on the GNPs may outgas and the density of the GNPs may approach that of graphite (2.2 g/cm³), in which case the relative density of the A-15G sample would be ~1% lower than reported here. An FEI NanoSEM scanning electron microscope (SEM) was used to observe microstructures and analyze surfaces of microscratches and microindents. A gold coating was used to ensure proper conductivity of the samples, and the SEM was operated at an accelerating voltage of 20 kV. Image J software was used to measure grain sizes and the

lengths of cracks generated during microindentation. An FEI Scios dual-beam focused ion beam – scanning electron microscope (FIB-SEM) was utilized to cross-section and characterize microindents and microscratches. Cross-sectioning was done via the FIB micro-machining capabilities; initial cuts were performed with the ion beam operating at an accelerating voltage of 30 kV and a current of 3.0 nA. Final sectioning was performed with an accelerating voltage of 30 kV and a lower current of 0.5 nA in order to avoid ion beam induced damage.

2.3. Mechanical Properties Evaluation

The sintered samples were mounted and ground using a 600 grit size SiC abrasive paper. The mounted samples were then polished using progressively finer diamond slurries, with the final polishing step utilizing a slurry with 50 nm diamond particles. Microindentation was performed on the polished samples using a Buehler Micromet 2004 microhardness tester. A standard Vickers tip was used and the loading conditions consisted of a 1000 g load with a 15 s hold. The indents generated were observed under an SEM in order to measure the lengths of the cracks generated. The crack length (c) was measured from the center of the indent to the end of the crack. The fracture toughness (K_{IC}) was then calculated using the Anstis Equation.³⁵

$$(1) \quad K_{IC} = 0.016 \left(\frac{E}{H} \right)^{\frac{1}{2}} \left(\frac{P}{c^{\frac{3}{2}}} \right)$$

The other parameters in the Anstis equation are the indentation load (P), the microhardness (H), and the elastic modulus (E). The statistical significance of the variations in indentation fracture toughness values were verified using statistical t-tests. Nanoindentation tests were conducted using an MTS Nano Indenter XP in order to measure nanohardness and elastic

modulus of the monolithic and composite samples (not individual phases). A Berkovich tip was used and the tip area calibration was done using a standard quartz sample with a known elastic modulus of 69.6 GPa. The nanoindentation load cycle consisted of a 10 s ramp to the maximum load of 12 mN, with a holding time of 3 s, followed by a 10 s unloading to 0 N. The elastic modulus was calculated from the unloading portion of the load-displacement curve using the Oliver-Pharr method.³⁶ A poisson's ratio value of 0.235 was taken for alumina during the elastic modulus calculation.^{37,38} All indentations were performed on a plane perpendicular to the pressing axis during sintering. Data reported from micro and nanoindentation tests were taken from an average of 12 indents. On average, 20 cracks were measured to calculate fracture toughness. It is emphasized that the mechanical testing techniques have been widely utilized in the field as they represent the best practice methods due to both to their accuracy and low cost. These methods are also valid on ceramic matrix composites; the samples in the present study are dense ceramics, which can be evaluated using both the Anstis equation and the Oliver-Pharr method. All error bars correspond to one standard deviation.

2.4. Microscale Tribological Performance

The tribological performance of the nanocomposites was assessed using the MTS Nano Indenter XP in scratch mode. Scratch tests were performed under unlubricated conditions in order to assess the intrinsic wear behavior of nanocomposite samples. Scratches were performed on the polished surfaces using a Berkovich tip with the Berkovich tip oriented so that the scratch direction was oriented along one of the edges of the tip. A hard diamond Berkovich tip was used during scratch testing in order to minimize wear of the counter surface; this enables one to assume that only the sample material being investigated

becomes worn and hence behavior observed is system independent. The scratches were performed under constant normal loads of 12 mN, 100 mN, and 400 mN with a scratch length of 50 μm and a scratch speed of 0.50 $\mu\text{m/s}$. Lateral forces were measured as the scratch was performed enabling the coefficient of friction to be obtained from the ratio of lateral to normal forces. The residual depths of the scratch grooves were profiled using a Dimension 3100 atomic force microscope (AFM) (Digital Instruments, Veeco Metrology Group, Santa Barbara, CA). The AFM profiling was done using a scan rate of 0.10 Hz and a tip velocity of 10 $\mu\text{m/s}$. The 3-D profiles obtained by the AFM were then sectioned into 2-D profiles in order to measure the cross-sectional area of the scratch groove. An average of over 20 scratch cross-sections was taken and then multiplied by the known scratch length of 50 μm to calculate the volume lost during the scratch test (i.e., wear volume). AFM profiling was also used to characterize the average surface roughness (R_a) and roughness root mean square (R_p) of the polished samples. The average of 5 scratch tests was utilized to calculate values and the error bars reported represent one standard deviation.

2.5. *In Vitro* Biocompatibility

The biocompatibility of Al_2O_3 -GNP nanocomposites was investigated by culturing osteoblast cells on them and evaluating cell viability and proliferation relative to pure Al_2O_3 samples. For each composition, cell culturing tests were performed on three samples. Human osteoblasts MG-63 ATCC CRL-1427 (American Type Culture Collection, Manassas, VA) were cultured in a 24-well plate using a cell density of 30,000 cells/mL in Dulbecco's modified eagle medium (DMEM, Invitrogen, Grand Island, NY), supplemented with 10% fetal bovine serum (FBS, ATCC, Manassas, VA), and 1% penicillin-streptomycin (ATCC, Manassas, VA). Cell viability and proliferation was evaluated using an Alamar Blue

Assay (Invitrogen, Grand Island, NY) according to the manufacturer's instructions. Briefly, 100 μ L of Alamar Blue **was** added into the cell culture medium. Cells **were** incubated for 1 hr in a standard cell incubator (~ 37 $^{\circ}$ C, 5% CO_2). After periods of 1 day, 3 days, and 5 days, 900 μ L of cell medium **was** removed from the 24-well plates and inserted into a 96-well plate, with 300 μ L of cell medium per plate. The absorbance at wavelengths of 570 nm and 600 nm was read for each plate using an Infinite 200 Pro microplate spectrophotometer (TECAN, Männedorf, Switzerland). The relative absorbance **was** converted to percentage increase over the background absorbance based on absorbance measurements from blank control samples on each plate. Cell culture samples for SEM analysis **were** prepared by removing cell medium and gently rinsing samples with phosphate buffer solution (PBS). Cells **were** fixated by applying 1 mL of 3.7% formaldehyde solution and fixating in 4 $^{\circ}$ C environment for 30 min. After fixation, samples **were** rinsed with PBS and then sequentially incubated with solutions of 50%, 75%, 90%, and 100% ethanol. Samples **were** then dried and sputter coated with gold before observation under the SEM. Statistical t-tests **were** used to evaluate whether differences in cell proliferation **were** statistically significant among the different compositions.

3.0 RESULTS AND DISCUSSION

3.1. Microstructural Characterization

The microstructure of the alumina and GNP-reinforced alumina nanocomposites is briefly described here. Further details on the microstructure of the different samples and the microstructural evolution as a function of sintering conditions can be found elsewhere.³⁴ The typical microstructure of alumina and A-5G nanocomposite are presented in Figure 1. The

inset of Figure 1b shows a kinked GNP found in the nanocomposite sample. The kinking of GNPs is one of the primary toughening mechanisms in GNP-CMCs.¹⁸ It can be seen that the A-5G sample had a much finer grain size due to the effect of GNPs suppressing grain growth. The A-10G and A-15G samples had further reductions in grain size as can be seen from the values shown in Table 1. The GNPs are also seen to be well dispersed throughout the alumina matrix and hence grain growth suppression is uniform and grain size distributions were narrower in the samples with higher GNP content.³⁴ All samples were fully densified (>99%) during the sintering process. From the data provided in Table 1 it can be seen that minor porosity was present in the A-10G and A-15G samples. This porosity is expected to be localized near the GNPs, as documented in previous studies on GNP-reinforced nanocomposites.^{18,26,27}

3.2. Mechanical Properties

3.2.1. Elastic Modulus and Hardness

The elastic modulus results, as determined by nanoindentation, are shown in Table 1. These results indicated that the stiffness of the nanocomposites decreased as the GNP content was increased. The decrease in stiffness was expected because the GNPs are a soft phase in the out-of-plane (c-axis) direction, which has the highest surface area. Nanoindentation testing is by its nature performed on small volumes, hence deformation occurs only on a few (or a single) grain. For example, for the alumina and A-5G samples the indent depth was ~150 nm, with a corresponding projected indent size of ~1 μm . GNPs that are found around or underneath alumina grains during the indentation can result in higher indentation displacement (indentation load-displacement curves are provided in

Supplementary Figure S1). The GNPs undergo a higher degree of permanent deformation and may also facilitate grain boundary sliding if the graphene layers become sheared. The higher GNP content samples also had finer grain sizes and hence there is a higher volume of grain boundaries, which also can contribute to increased permanent deformation. Decreased stiffness with increasing GNP content has been previously observed in the few studies that have investigated the elastic moduli of GNP-CMCs.^{18,20} It should be noted, however, that in one study by Zhang et al.⁵⁶ an increase in elastic modulus of up to 44% was attained with the addition of 1 wt.% GNP. The GNPs in this study were able to increase modulus because they were embedded in the ceramic matrix (instead of at grain boundaries), which prevents them from sliding and constricts their deformation in the c-axis direction.

It is important to note that the observed decrease in elastic modulus with increasing GNP content observed in the current study would be beneficial for hip implant applications, because ideally the entire implant would have an elastic modulus that is equivalent to that of bone (17.4 GPa³⁹). Current Al₂O₃ and zirconia toughened alumina (ZTA) ball and cups have elastic moduli that are significantly higher (360 – 400 GPa⁴⁰). This is accepted as a design tradeoff because no material with the elastic modulus of bone has the necessary combination of strength, toughness, and wear resistance. Decreasing the elastic modulus of the ball-and-cup components would alleviate uneven loadings and thereby reduce damage that occurs to surrounding bone^{4,5}.

Hardness was evaluated at both nano- and microscales, as shown in the results tabulated in Table 1. The soft GNP phase was responsible for the observed loss in hardness, however, the reduced grain sizes partially mitigated GNP-induced softening, and hence decreases in hardness were less pronounced than decreases in elastic modulus.

Microhardness values displayed the general trend of decreased hardness with increased GNP content, however the values were consistently lower than the nanohardness values as the volume of material tested was significantly larger. The higher volume means there is a higher volume of defects, GNPs, and localized porosity present in the material tested, which lowers the hardness as their ability to resist permanent deformation is lower than that of the hard ceramic matrix. It was seen that the smallest decreases in hardness at both the nano- and microscale occur for samples with the highest GNP contents. The high volume fraction of GNPs results in a relatively high volume of GNPs being present even in the small volumes tested during nanoindentation, thereby exhibiting similar behavior at both the nano- and microscale.

3.2.2. Fracture Toughness

Indentation fracture toughness results are provided in Figure 2; it can be seen that the addition of GNPs enhanced the toughness of A-5G and A-10G samples relative to pure alumina by 21% and 19%, respectively. The A-15G sample, however, was less tough than the pure alumina sample. This indicates that even with a uniform distribution of nanofiller, there is a limit to the amount of GNPs that are beneficial to toughening. The fact that toughness plateaus at 10 vol.% indicates that the percolation limit for toughening occurs between 5 – 10 vol.%. As the GNP content was increased further it became increasingly likely that GNPs would be stacked onto one another. Thick stacks of GNPs induce porosity and can act as stress concentrators that facilitate crack nucleation and propagation. The presence of stacked GNPs and localized porosity was likely greater in the A-15G sample as the sample surpassed the percolation limit and was likely the cause for the higher standard deviation in this sample.

In addition to the kinked GNP structures seen in the microstructure, the inset in Figure 2 provides direct evidence of GNP energy dissipating mechanisms. Figure 2 shows a kinked GNP bridging a crack generated during the microindentation; energy is dissipated during the kinking and pull-out of the GNP. It is noteworthy that despite the high content and uniform dispersion of GNPs, the improvement in toughness **was** modest relative to other studies on GNP-CMCs where toughness **was** enhanced by up to ~135% with much lower (≤ 6 vol.%) GNP content.^{17,18} The effectiveness of GNP reinforcements can be understood in terms of micromechanical models such as the Cox model⁴¹ which state that there is a critical length for fiber reinforcements to provide effective reinforcement. Zhang et al.⁴² showed **ed** that for a typical GNP consisting of up to 40 layers of graphene the critical length for effective reinforcement is ~700 nm. The GNPs in the present study **were** ~1 μm long and hence **served** as effective reinforcements. Zhang et al.⁴¹ and Ramirez et al.⁴³ have also shown that the energy dissipated during GNP bridging and pull-out is maximized with higher GNP dimensions as the interfacial shear stress transfer will be greater with higher surface area. Studies on GNP-CMCs with the greatest improvements in toughness typically utilize GNPs with lateral dimensions of ~5 μm .¹⁶⁻¹⁸ The modest improvements in toughness in the present study **were** the result of the reduction in GNP lateral dimensions (from 5 μm to 1 μm) during the ball milling step in the nanocomposite powder preparation. The higher content of GNPs **was** hence offset by their reduced effectiveness in dissipating energy through interfacial shear stress transfer due to their reduced lateral dimensions.

3.3. Microscale Tribological Performance

3.3.1. Wear Resistance

Figure 3a shows the average scratch penetration depths under various normal loads for the Al_2O_3 and Al_2O_3 -GNP nanocomposites. The scratch penetration depth is a measure of wear resistance prior to elastic recovery. This parameter is important in loading environments where there are high frequency or continuous loadings. During locomotion a hip implant will experience such an environment, because a normal walking gait will lead to continuous contact between the ceramic head and cup. It can be seen that at the lowest load of 12 mN the performance of all the samples was similar. Under this small load, the scratch penetration depth is ~100 nm and hence the volume scratched was not sufficient for the GNP to play a role in the wear response. Differences in scratch depth became salient when the normal load was increased to 100 mN. At this load, the Al_2O_3 and A-5G samples had equivalent performance, while the A-10G and A-15G had steadily increasing penetration depths. The scratch depths at these loads are on the order of 500 – 900 nm and were largely controlled by the hardness of the samples. The scratch depths at 100 mN loads do in fact mirror the trends seen for nanohardness and can be explained by the same mechanisms. At a loading of 400 mN the penetration depth increased to 1 – 2 μm and the occurrence of fracture became an important aspect of the wear response. This penetration depth was on the order of the grain size and hence the GNPs are expected to play a larger role in the wear response as GNPs are typically found at grain boundaries. It can be seen that the A-5G and A-10G samples had significantly reduced penetration depths as compared to the unreinforced alumina, while that of A-15G was significantly higher.

The improved wear performance of the A-5G and A-10G samples can begin to be understood by analyzing the scratch groove morphology after the 400 mN normal load scratches, which is provided in the SEM images in Figure 3b-d. It can be seen from Figure

3b that at this loading the alumina sample showed typical signs of a brittle wear response. Cracks and fractured regions can be seen on the alumina scratch groove. It is also seen that cracks were initiated at the scratch boundary and propagated for several microns into the undeformed regions. This compromises the structural integrity and would be expected to lead to a more rapid deterioration in wear behavior during subsequent loadings. Figure 3c shows a typical 400 mN scratch for A-5G, which is also representative of the A-10G sample. It can be seen that features of a brittle wear response were greatly reduced. Smooth regions without any cracking were commonly found and crack propagation into the surrounding material was absent. The wear debris in A-5G also consisted of finer particles, typically ≤ 1 μm in diameter as compared to the wear debris in the alumina sample in which 2 - 3 μm diameter particles were seen. The finer debris indicates that fracture was less severe. The inset of Figure 3c shows a pulled-out GNP arresting a crack, which provides evidence that GNP toughening mechanisms can actively participate in the wear response at these higher loadings. In sharp contrast to the other samples, the scratch grooves of sample A-15G, shown in Figure 3d, indicate that fracture occurred throughout the scratch. The fracturing of the scratch grooves indicates a much lower resistance to wear and hence the higher penetration depths. The fracture within the groove compromised the structural integrity of the material and hence deteriorated the material's ability to withstand further scratch loadings. Furthermore, the large amount of debris formed would be detrimental in subsequent scratch loadings as the debris would act as third body abrasives. The inhibition of brittle wear responses is critical to hip implant applications. The formation and accumulation of wear debris would serve as third body abrasives, which would greatly deteriorate the lubricious properties of the synovial fluid. The wear debris would be carried

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5 to other parts of the joint via the synovial fluid and thereby accelerate the wear of the hip
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7 implant. The reduction in brittle wear responses induced by GNP in the A-5G and A-10G
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9 samples makes these materials less prone to generating undesirable wear debris and hence
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11 would be beneficial by inhibiting the wear of hip implants.
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15 The wear resistance was further characterized by calculating the wear volume
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17 because this is a direct measure of the material lost and takes into account the elastic
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19 recovery. Profiles of typical 400 mN scratches are provided in Figure 4a from which the
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21 wear volumes in Figure 4b were calculated. The track profiles of Al₂O₃, A-5G, and A-10G
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23 were similar, with the primary differences being higher residual scratch depth and increased
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25 pile-up in the alumina sample. It was seen that the addition of 5 vol.% and 10 vol.% of
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27 GNP enhanced wear resistance by 39% and 30%, respectively, when a 400 mN load was
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29 applied. Comparison of the scratch penetration depth in Figure 3 and residual scratch depth
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31 in Figure 4 shows elastic recovery was greatest in pure alumina. Elastic recovery was
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33 expected to be lower in the nanocomposite samples because the GNP deform plastically
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35 when they undergo kinking, bending, or sliding. However, the high elastic recovery of
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37 alumina was not sufficient to compensate for the greater scratch penetration and material
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39 removal during the scratch process. The A-15G sample deviates significantly from the above
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41 trends as the wear volume was ~313% larger than alumina despite the penetration depth only
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43 being about 34% greater. This large difference is due to the A-15G sample severely
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45 fracturing, which results in minimal elastic recovery. The severe fracturing in sample A-15G
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47 was also seen from the lack of pile-up material in the scratch profile. This indicated that the
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49 material instead collapses into the groove as the scratching process caused fracture. The
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51 inhibition of microscale wear is critical to hip implants as microscale wear will lead to fine
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5 scratches and grooves on the implant surface which increase roughness. Increased roughness
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7 leads to higher frictional forces which will exacerbate implant wear and consequently
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9 adversely affect implant lifetime and performance. Hence the enhanced microscale wear
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11 resistance of samples A-5G and A-10G relative to unreinforced Al_2O_3 is beneficial for hip
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13 implants because it will inhibit the formation of fine scratches which ultimately have
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15 adverse effects on implant performance and lifetimes.
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20 The variations in wear volume were not as significant at lower loads, because the
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22 role of the GNPs was diminished at smaller scratch depths. At 100 mN loading the Al_2O_3 ,
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24 A-5G, and A-10G samples had equivalent wear volumes. The A-15G wear volume was still
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26 substantially greater than the other samples under 100 mN loads, in fact wear volume for A-
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28 15G at 100 mN was comparable to wear volumes of the other samples at 400 mN loading.
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30 The addition of 15 vol.% GNP would hence have adverse effects on hip implant
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32 performance at 100 mN loadings due to increased wear. The wear volumes for 12 mN load
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34 scratches were substantially smaller in all samples as no fracture was seen to occur at these
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36 smaller loads (scratch morphologies for 12 mN and 100 mN load scratches are characterized
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38 via SEM in Supplementary Figure S2). The diminished role of GNPs and increased elastic
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40 recovery of alumina resulted in the smallest wear volume at this load. The wear volume
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42 increased with higher GNP content due to decreased hardness. At these very low loadings
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44 (12 mN), the A-5G and A-10G samples had equivalent wear responses to Al_2O_3 , however,
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46 the addition of higher GNP contents (15 vol.%) lead to higher nanoscale wear which would
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48 have an adverse effect on the wear of hip implants.
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3.3.2. Coefficient of Friction

The coefficient of friction (CoF) values derived from scratch tests under the various normal loads are presented in Figure 5. The general trend **was** that the addition of GNPs **did** not impact CoF. This contradicted **ed** the expected result based on Kim et al.'s²⁹ proposed mechanism of intrinsic GNP lubrication. If GNPs on the surface of grains serve as solid state lubricants to facilitate sliding of the counter surface, then CoF should decrease even at the lowest loadings. Similarly, if GNPs provided **ed** lubrication between grains, then the CoF should have decreased **d** with increasing GNP content at the largest loads where the scratched volume encompassed **d** many grains. GNPs located on grain boundaries **were** constrained by the surrounding grains in a dense structure and hence GNP induced grain boundary sliding **was** unlikely. Our results therefore indicated **d** that the presence of GNPs in grain boundaries **was** not the source of GNP-induced lubrication seen in previous works^{25,27-29,32} on the macroscale wear behavior of GNP-CMCs.

The inset of Figure 5 shows the typical real-time CoF data for the various normal loads utilized. For a given load, real-time COF data **were** similar across all samples; the real-time COF data are presented in order to emphasize differences observed as the normal load **was** increased. The primary difference **was** the substantially lower CoF under a 12 mN normal load. Lower CoF during nanoscale wear **was** expected since the mechanical response at those length scales **was** mostly elastic and the wear process proceeds largely through a micro-ploughing mechanism.⁴⁴⁻⁴⁶ It can be seen however that the A-15G had **d** a noticeably larger CoF than the other samples at the 12 mN loading. This **was** due to the increased surface roughness ($R_a \approx 17$ nm) as compared to the other samples ($R_a \leq 7$ nm); data for surface roughness is provided in Supplementary Figure S3. The increased roughness only

impacts the wear behavior at lower loadings as the roughness value is ~10% of the penetration depth, while at the higher loadings it became substantially smaller (< 1% at 400 mN). This explains why the level of CoF oscillation decreased substantially at the higher loadings. Previous studies^{25,28,30} have demonstrated increased roughness with increasing GNP content despite identical surface preparation procedures. This has been attributed to the irregular shape of GNPs and localized porosity. GNPs are inherently a rougher phase due to the wrinkled nature of graphene and the kinking and bending present in GNPs. This is in contrast to the smooth faceted grains of the alumina ceramic matrix. The larger increase in roughness for the A-15G sample may be due to its slightly higher porosity and the onset of agglomeration, as the percolation threshold was surpassed in this sample.

3.3.3. GNP Induced Wear Mechanisms

Previous studies on the wear behavior of GNP-CMCs have attributed improvements in wear resistance to lower CoF induced by GNPs. In the present study the GNPs do not affect CoF and hence the improved wear resistance is independent of CoF and requires further elaboration. In this section GNP induced wear mechanisms at different length scales are discussed based on both current results and those seen in previous studies. Our results indicated that at the nanoscale (e.g., for scratch penetration depths below 500 nm), the GNPs do not have significant influence on the wear response and hence improved wear behavior of GNP-CMCs is not expected at the nanoscale.

The 100 mN and 400 mN load scratches in the present study were representative of microscale behavior, because penetration depths exceeded 1 μm . In this regime, wear (scratch) resistance was enhanced by up to 39%. As alluded to above, the primary mechanisms responsible for enhanced wear resistance in A-5G and A-10G were GNP

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5 induced toughening. Direct signs of GNP induced energy dissipation were seen via pulled-
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7 out GNPs in the wear track as well as crack bridging during microindentation.
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9 Characterization of the scratch groove surface further revealed that extensive micro-cracking
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11 occurs in A-5G and A-10G, as shown in Figure 6a. The micro-cracks generated were
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13 typically less than 1 μm in length, and secondary nanoscale cracks of ~ 100 nm were seen
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15 emanating from the micro-cracks. These micro- and nano-cracks were not large enough to
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17 propagate rapidly and hence had negligible effects on the structural integrity. Instead, these
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19 micro-cracks are highly beneficial as they dissipate elastic strain energy that would
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21 otherwise be consumed by the propagation of larger cracks, which can cause catastrophic
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23 (fast) fracture. The formation of micro-cracking is due to the finer grain size and GNP
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25 toughening mechanisms present in A-5G and A-10G. It is emphasized that no previous
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27 study has investigated the nanoscale or microscale wear behavior of GNP-CMCs.
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34 At the macroscale it is believed that GNPs enhance wear behavior by both the
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36 toughening mechanisms present at the microscale in the present study, as well as by the
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38 formation of a thin GNP rich lubricating tribofilm seen in several studies.^{25,27,32} The
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40 formation of a tribofilm explains why the CoF is reduced in macroscale testing but not in the
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42 current study. The reduction in CoF is hence not an intrinsic property of GNP- Al_2O_3 ; instead
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44 the GNPs lead to the formation of a tribofilm during extended wear. No such film formation
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46 was possible in the current study as testing consisted of a single scratch. However, the
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48 present study supports the theory that a graphene lubricating film can quickly form during
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50 extended macroscale testing. SEM analysis of the wear debris indicated that GNPs are
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52 readily found in debris of GNP-reinforced samples, as shown in Figure 6b. Two salient
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54 features of the debris are: i) the GNPs were electron transparent due to their ultrathin nature
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and ii) GNPs were detached from the Al₂O₃ matrix. These features indicate that after only a single scratch, GNPs can be sheared and detached from the matrix, which indicates the subsequent tribofilm formation could rapidly occur. It is emphasized that the source of detached GNPs is the worn surface, hence the detachment of GNPs would not adversely affect toughening as the material being toughened has already been removed during the wear process.

3.3.4. Scratch sub-surface damage

The present microscale study can also be used to provide preliminary insight into the expected behavior at larger scales by evaluating the level of damage induced by a single scratch on the scratch sub-surface. The damage zone on the sub-surface can be estimated by calculating the region of elastoplastic deformation during a single indentation. The radius (r_{ep}) of the elastoplastic deformation zone during indentation by a Berkovich tip can be estimated by Eqn. (2),^{47,48}

$$(2) \quad r_{ep} = \sqrt{\frac{0.3F_{MAX}}{\sigma_Y}}$$

where F_{MAX} is the maximum normal force and σ_y is the yield strength. The yield strength of alumina ceramics can be estimated from hardness measurements by $H/\sigma_y \approx 1.8$.^{49,50} The calculated elastoplastic deformation zone radii for Al₂O₃, A-5G, and A-10G under a 400 mN normal load were calculated to be 2.58 μ m, 2.59 μ m, and 3.05 μ m, respectively. The Al₂O₃ and A-5G samples had similar elastoplastic zones due to their similar nanohardness, while that of A-10G was ~ 17% larger. Elastoplastic calculations do not apply to A-15G as fracturing was the dominant wear response. The structure of the elastoplastic deformation zones in Al₂O₃ and A-5G were investigated by FIB sectioning the 400 mN scratches; sub-

surface microstructures are presented in Figure 7. It can be seen that in the Al_2O_3 sample a large crack initiated in the scratch surface is seen to propagate into the subsurface, through and beyond the calculated 2.6 μm radius elastoplastic deformation zone. Sub-surface cracking undermines the structural integrity and hence the wear response during subsequent scratches would be aggravated by cracks formed during previous scratches. Large cracks can lead to fracture of large segments, which results in large pieces of debris that become third body abrasives. In contrast, the A-5G sample had no such cracking, instead thin sheet-like structures can be seen throughout the structure. The inhibition of cracking **was** attributed to GNP energy dissipating structures seen in the sub-surface and highlighted in the inset of Figure 7b. Several kinks and bends **were** seen near the center where the contact stresses **were** highest. The presence of these kinks is direct evidence of the ability of GNPs to limit damage to the sub-surface structure through energy dissipating mechanisms. **The limiting of sub-surface damage is especially critical to the wear of hip implants as large cracks generated would likely remain undetected. The introduction of large cracks during the sliding process could lead to eventual fast fracture of the implant, which would cause implant failure, potential injury to the patient, and require surgery to replace the implant. The addition of GNPs is hence beneficial to hip implant applications as the energy dissipating mechanisms of GNPs inhibit sub-surface cracking which can go undetected and lead to fast fracture (failure) of the hip implant.**

3.4. *In Vitro* Biocompatibility

The cytotoxicity of the graphene reinforcement **was** assessed by investigating the viability of human osteoblast cells grown on the surface of Al_2O_3 and Al_2O_3 -GNP

nanocomposites. The *in vitro* assessment of the biocompatibility of these Al₂O₃-GNP nanocomposites with human osteoblasts has never been investigated before, and is utilized here as a preliminary evaluation into whether the incorporation of GNPs could have any adverse effects on the biocompatibility of Al₂O₃ ceramic implants. It is noted that further studies on wear debris and inflammatory responses would be needed to fully verify the applicability for actual biomedical applications. Cell viability data obtained from Alamar Blue assay testing is presented in Figure 8; pure alumina is known to be biocompatible and therefore data for nanocomposites was normalized relative to pure alumina. It can be seen that cell proliferation is robust in samples with high contents of GNP (A-10G and A-15G) throughout the entire period investigated. Cell counts in these samples were typically 1.5 – 2.0 times higher than in the pure alumina sample. Statistical analysis indicates that differences between the control sample, Al₂O₃, and both A-10G and A-15G nanocomposite samples was statistically significant ($p < 0.02$ for all tests except for Day 5 test for Al₂O₃ and A-15G, which had a statistical significance of $p < 0.08$). Also, except for Day 1, Al₂O₃ and A-5G sample had no statistically significant differences in cell viability. SEM micrographs in the inset of Figure 8 show a higher amount of cells present on the surface of sample A-15G after incubation of 1 day, as compared to the pure Al₂O₃ sample. Cells grown on sample A-5G exhibited a lower viability than the sample after 1 day of incubation, however cell growth recovered in days 3 and 5 and was equivalent to that of Al₂O₃. GNPs therefore do not induce any cytotoxic effects in Al₂O₃-GNP nanocomposites, instead higher concentrations of GNPs lead to robust proliferation of cells.

The biocompatibility of graphene has been seen previously in the literature for several types of cells including human bone marrow derived mesenchymal stem cells,⁵¹⁻⁵³

neural stem cells,^{54,55} and chondrocytes.⁵¹ However studies on the biocompatibility of GNPs in ceramics matrix composites are limited to hydroxyapatite systems.^{56,57} The addition of GNP reinforcement in hydroxyapatite nanocomposites has been shown to enhance the biocompatibility with human osteoblasts, primarily by inducing mineralization of apatite due to GNPs serving as nucleation sites.^{56,57} and the reduction in grain size that enhances calcium ion dissolution.⁵⁶ Neither of these mechanisms would be present in the Al₂O₃ system and hence the primary feature of the GNPs responsible for enhanced biocompatibility was their nanometric dimensions. Nanometric materials have been shown to enhance biocompatibility due to their nanoscale ‘roughness’⁵⁸ which in the case of GNPs consist largely of wrinkles and kinks in their structure.^{24,59} Increased roughness enhanced cell adhesion by increasing the available surface area. Surface roughness measurements in Supplementary Figure S3 confirm increased surface roughness with increased GNP content. TEM analysis by Liu et al.⁵⁷ confirms that GNPs (like other nano-textured surfaces) enhance absorption of vital cell proteins such as fibronectin and vitronectin.⁶⁰ Enhanced adhesion of cells and absorption of cells were therefore believed to be the reasons for robust proliferation of cells in samples A-10G and A-15G.

4.0. CONCLUSIONS

The mechanical properties, microscale tribological behavior, and *in vitro* biocompatibility of Al₂O₃ samples reinforced with 5- 15 vol.% GNPs were investigated. The introduction of the soft GNP phases lead to decreased hardness and elastic modulus. Fracture toughness and microscale wear resistance were enhanced with addition of 5 – 10 vol.% GNPs, with sample A-5G displaying the greatest improvements in toughness (21%)

and wear resistance (39%). Coefficients of friction were not significantly impacted by the addition of GNPs under high normal load (100 – 200 mN) scratch tests and hence GNP induced lubrication was not responsible for improved wear resistance. Enhanced wear resistance was attributed to GNP induced toughening, which lead to the formation of fine (~100 nm) microcracks that dissipated strain energy. SEM analysis of scratch grooves revealed a largely smooth surface in A-5G and A-10G, in contrast to the Al₂O₃ sample where fracture and crack propagation were observed. In addition, sub-surface analysis conducted via FIB sectioning revealed enhanced damage tolerance in the structure immediately below the scratch in GNP reinforced samples. GNPs in the subsurface of sample A-5G were observed to kink and thereby dissipate energy, which inhibited cracking during the scratch loading. In contrast, large cracks extending beyond the calculated elastoplastic zone were observed in the pure alumina sample. Biocompatibility with human osteoblasts was evaluated, and it was seen that no cytotoxic effects are induced by GNPs. Cell proliferation became more robust in samples reinforced with higher GNP contents due to the surface properties of GNPs and the finer grained microstructure induced by the presence of GNPs. The toughened Al₂O₃-GNP nanocomposites with enhanced microscale wear resistance and biocompatibility are hence feasible material systems for biomechanical applications such as wear resistant hip implant ball and socket components.

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Tables

Table 1: Microstructure and Mechanical Properties of Al_2O_3 -GNP Composites

Sample	Relative Density (%)	Grain Size (μm)	Elastic Modulus (GPa)	Micro-hardness (GPa)	Nano-hardness (GPa)	Fracture Toughness ($\text{MPa m}^{1/2}$)
Al_2O_3	100	2.5 ± 1.2	430 ± 14	19.6 ± 0.9	32.2 ± 1.9	2.8 ± 0.4
A-5G	$100^\dagger$	$1.3 \pm 0.5^\dagger$	356 ± 26	$17.7 \pm 1.0^\dagger$	32.1 ± 3.6	3.4 ± 0.3
A-10G	$99.8^\dagger$	$0.9 \pm 0.3^\dagger$	291 ± 26	$15.1 \pm 1.1^\dagger$	23.2 ± 2.9	3.4 ± 0.5
A-15G	$99.4^\dagger$	$0.6 \pm 0.2^\dagger$	186 ± 16	$13.6 \pm 1.1^\dagger$	14.3 ± 1.2	2.1 ± 0.6

[†] Data published previously in Ref [34]

Fig 1: Scanning electron micrographs of a) alumina microstructure, and b) A-5G microstructure. GNP reinforced samples have finer grain sizes and typically have a uniform distribution of GNPs, Inset: Kinked GNPs found in alumina-GNP samples. Yellow arrows denotes GNPs visible in the SEM, thinner (< 10 nm) GNPs may not be easily discernible in an SEM.

Fig. 2: Indentation fracture toughness (IF) of alumina-GNP samples; Inset: Scanning electron micrograph of kinked GNP bridging crack in GNP reinforced sample. Yellow arrows denote kinks on GNP. A-5G and A-10G samples are shown to be statistically greater ($p < 0.001$) than pure alumina and A-15G samples.

Fig. 3: a) Scratch penetration depth as a function of GNP content, b) scanning electron micrograph (SEM) of characteristic scratch groove of pure alumina after 400 mN load scratch, c) SEM of A-5G scratch groove after 400 mN load scratch, typical of grooves seen for A-5G and A-10G, Inset: Pulled-out GNP pinning a propagating crack in the scratch groove, d) scratch groove of A-15G shows typical fracture-surface like morphology after 400 mN load scratch. Scratch direction in all SEM images is left to right.

Fig. 4: a) Scratch groove profiles of alumina and alumina-GNP composites used to calculate wear volume for 400 mN load scratches , b) wear volume as a function of GNP content for scratches performed under varying normal 400 mN and 100 mN loads, Inset: Wear volume of scratches with 12 mN normal loads

Fig. 5: Coefficient of friction as a function of GNP content for scratches under various normal loads; Inset: Typical real time CoF data at varying normal loads

Fig. 6: Typical features found on scratch grooves of A-5G and A-10G scratch grooves, a) scanning electron micrograph of typical micro and nano cracks which inhibit formation of larger cracks, b) scanning electron micrograph of thin electron transparent GNP debris

Fig. 7: Cross-sections of 400 mN scratch grooves, dotted yellow line denotes edge between scratch top surface and cross-section, a) alumina scratch cross section, where red arrows denote path of crack initiated at scratch surface which is seen to propagate several microns into the sub-surface, b) A-5G scratch cross section, Inset: Higher magnification image showing kinked GNP features in the sub-surface. Green arrows denote kinks

Fig. 8: Relative cell viability of osteoblasts on Al_2O_3 -GNP composites cultured for different incubation periods relative to those grown on unreinforced Al_2O_3 . Insets show scanning electron micrographs of cells on Al_2O_3 and A-15G samples after a 1 day incubation period