Neutral Atom Beam Technique Enhances Bioactivity of PEEK

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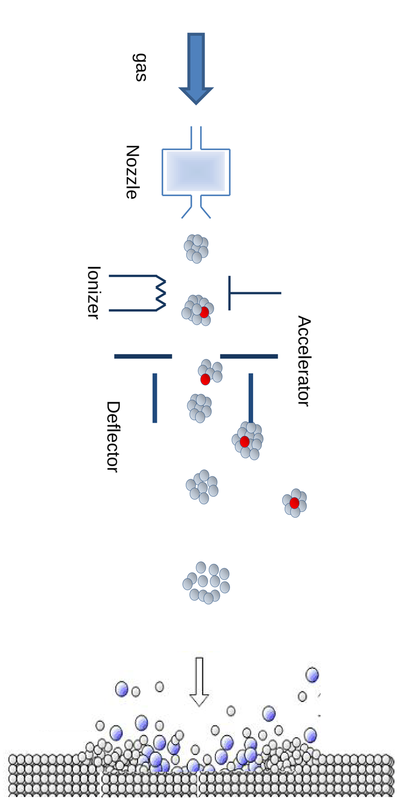
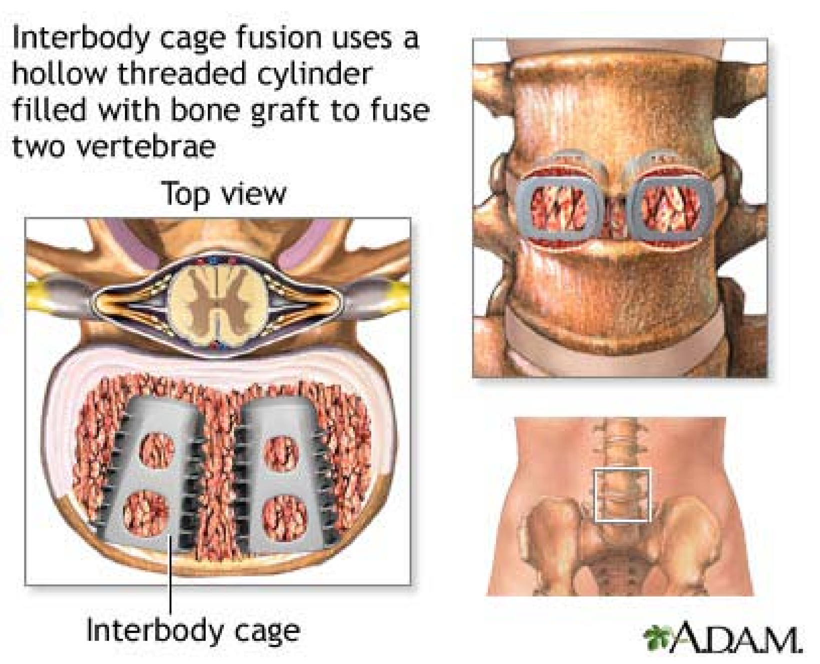
Abstract

Polyetheretherketone (PEEK) is currently gaining popularity in orthopedic and spinal applications but has potential drawbacks in use. PEEK is biocompatible, similar in elasticity to bone, and radiolucent; however, it has been shown to be inert and does not integrate well with bone. Recent efforts have focused on increasing the bioactivity of PEEK by modifying the surface to improve the bone-implant interface. We have employed a novel Neutral Atom Beam Technique (NABT) to enhance the bioactivity of PEEK. NABT employs an intense beam of cluster-like packets of accelerated unbonded neutral argon (Ar) gas atoms. These beams are created by first producing a highly energetic Gas Cluster Ion Beam (GCIB) comprised of van der Waals bonded Ar atoms, then transferring energy to the clusters so as to cause release of most of the interatomic bonds, and finally deflecting away the remaining electrically charged cluster cores of still bonded atoms. In initial studies, PEEK disks of 1cm Ø were treated with NABT at $2.5x10^{17}$ neutral Ar atoms/cm² or left as controls. Human osteoblasts (5000/cm²) were seeded onto the surface and allowed to proliferate for 4, 7, or 14 days, disks were then stained by crystal violet and visualized by light microscopy and eluted to measure cell number. An *in vivo* study using a rat calvarial critical size defect model was done to show bone growth on the surface of NABT-treated and untreated PEEK disks 3mm diameter, 1mm thick. Following 4 weeks after implantation into the calvarial defect, histology was performed to determine the amount of bone re-growth on the surface. PEEK processed by NABT resulted in increased cell attachment and proliferation at day 14 (48533±10101 cells) as compared to control (16748±10668 cells, p<0.03). The rat calvarial defect model showed that NABT-treated PEEK resulted in a bone ledge growing on top of the disk covering approximately 50% of the surface whereas the control PEEK resulted in only fibrous tissue with no bone growth at all. The controls displayed the start of bone resorption on the sides of the disks whereas good bone purchase is seen on the NABT-treated disks. NABT treatment of PEEK results in enhanced osteoblast attachment and proliferation as evidenced by cell proliferation and microscopy. Enhanced osseointegration was demonstrated in the *in vivo* study where bone tissue formation was evident only on the NABT treated PEEK. Taken together, these data suggest that NABT treatment of PEEK has the potential to enhance its bioactivity, resulting in bone formation and significantly decreasing osseointegration time of orthopedic and spinal implants.

Introduction to ~~GCIB~~ NABT

- Unique energetic ion bombardment technique.
- Clusters can be formed from a number of gases, Argon is typically used due to inert properties.
- ~~Clusters~~ Atoms that collide with surface have capability to modify material to about ~~10nm~~ <5nm below the surface.
- Surface Treatment using Gas Cluster Ion Beams :
  - Changes the surface properties without changing the surface chemistry of a material
  - Very shallow penetration – at atomic levels
  - Extremely localized process that only impacts the surface





Graphical representation of interbody cages and their placement in the spine

Benefits and Problems associated with PEEK biomaterials

Benefits:	**Problems:**
Biocompatible	Not Bioactive
Similar Elastic Modulus to Bone	Does not integrate with bone
Radiolucent	Requires addition of bone graft or BMP

Could the use of NABT surface processing result in enhanced bioactivity of PEEK resulting in device integration into bone?

Materials and Methods

Neutral Atom Beam Technique (NABT): Materials were cut to the following dimensions: PEEK film 1cm Ø, 0.2mm thick; PEEK disks 3mm Ø, 1mm thick; Titanium foil (Ti, CP1) 1cm Ø, 0.1mm thick. Materials were cleaned in 70% isopropanol for 30 minutes followed by 3 x 5 minutes washes in H_2O. All materials were prepared as controls or treated by NABT using Argon (Ar) gas on an Accelerated Particle Beam Alpha System (Exogenesis Corp) with a deflector to remove charged clusters. The effective dose of the Ar NABT was $2.5x10^{17}$ atoms per cm².

Atomic Force Microscope (AFM): AFM measurements were taken using a Park Systems XE-70 instrument in non-contact mode. Silicon tips with a resonant frequency of ~330 kHz and a force constant of 42 N/m were used (PointProbe® Plus, Nanosensors). 1µm regions of the PEEK were imaged and the arithmetical mean roughness (R_a) and ten-point mean roughness (R_z) was measured across this region.

Water Contact Angle: Contact angle was measured using the sessile drop method on a manual simplified device as described by Lamour et.al. (2010) and droplet angles were measured by ImageJ software (NIH) with the contact angle plugin.

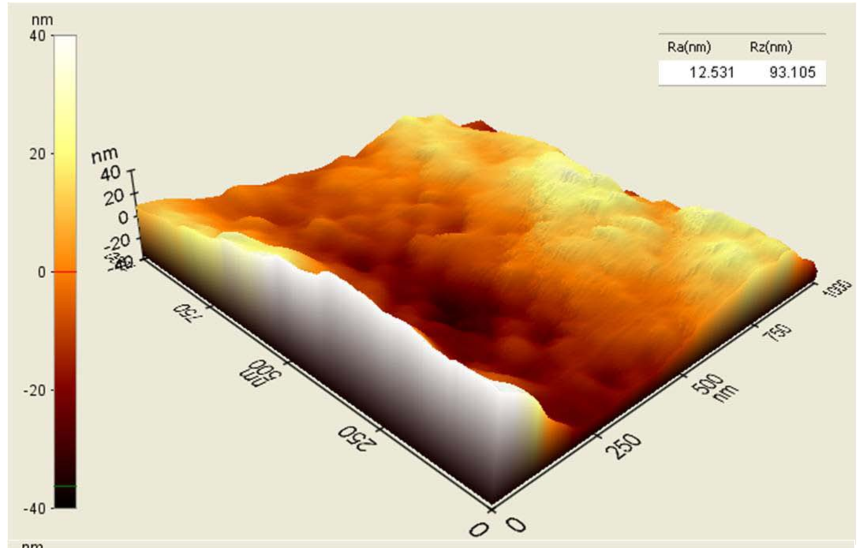
Cell Attachment and Proliferation: PEEK film (n=3 per condition and time) were placed in 24 well dishes and 5000 human fetal osteoblast cells (hFOB) were seeded on the surface in 1ml DMEM + 10% FBS and allowed to attach and proliferate for up to 14 days. In a parallel study, NABT-treated and control PEEK were compared to control Ti (which is considered bioactive) over a 10 day study.

Rat Calvarial Defect Study (*in vivo*): All animal work was approved and performed at CBSET following a local ethical review. CBSET is registered with the United States Department of Agriculture (USDA) to conduct research in laboratory animals. All procedures and conditions of testing complied with the Animal Welfare Act and its amendments. Male Sprague-Dawley rats, ~250 grams (n=6 per condition) were anesthetized with Isoflurane gas and the surgical site was shaved. A small incision was made in the top of the skull, and the skin and underlying tissue was incised and reflected to expose the cranium. A drill with a 3 mm burr was used to create the defect site, using care to avoid damaging the dura mater. One implant (either a test or control device) was placed in the defect site, and the soft tissue and skin was closed appropriately in layers. Moribundity/mortality checks were performed twice daily and all animals in the study performed well. Four weeks after implantation, animals were sacrificed and the cranium was removed *in toto* and placed in 10% buffered formalin. Undecalcified, intact calvaria with implant sites were resin embedded and micro-ground and histomorphologic evaluation was used to qualitatively assess and characterize the extent of new bone deposition and relative to the surface of the implant.

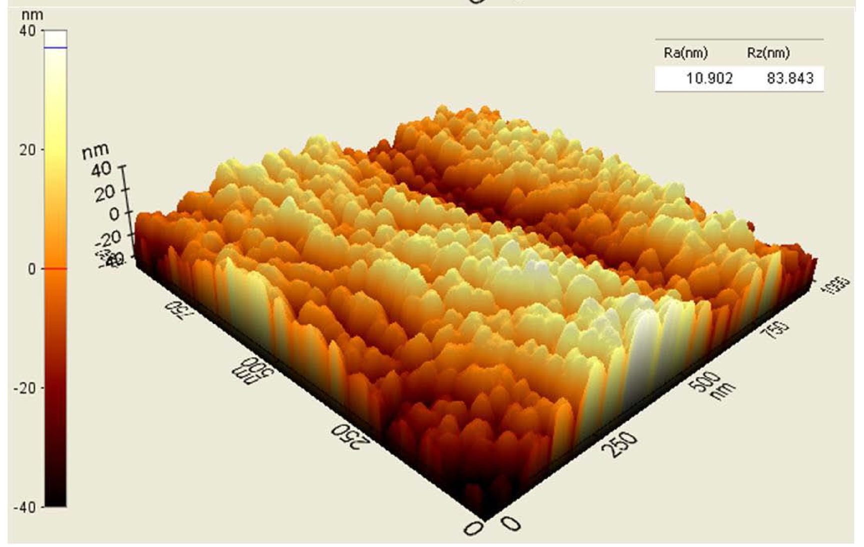
Results

NABT results in Nano-Scale Texturing of the Surface of PEEK: AFM measurements revealed a nano-scale texturing of the surface on NABT-treated PEEK film as compared to controls. This nano-texturing, by other methods, has been well established in the literature in producing surfaces favorable for cell attachment and proliferation.

Control

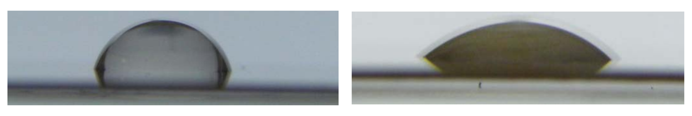


NABT-treated



Results (Continued)

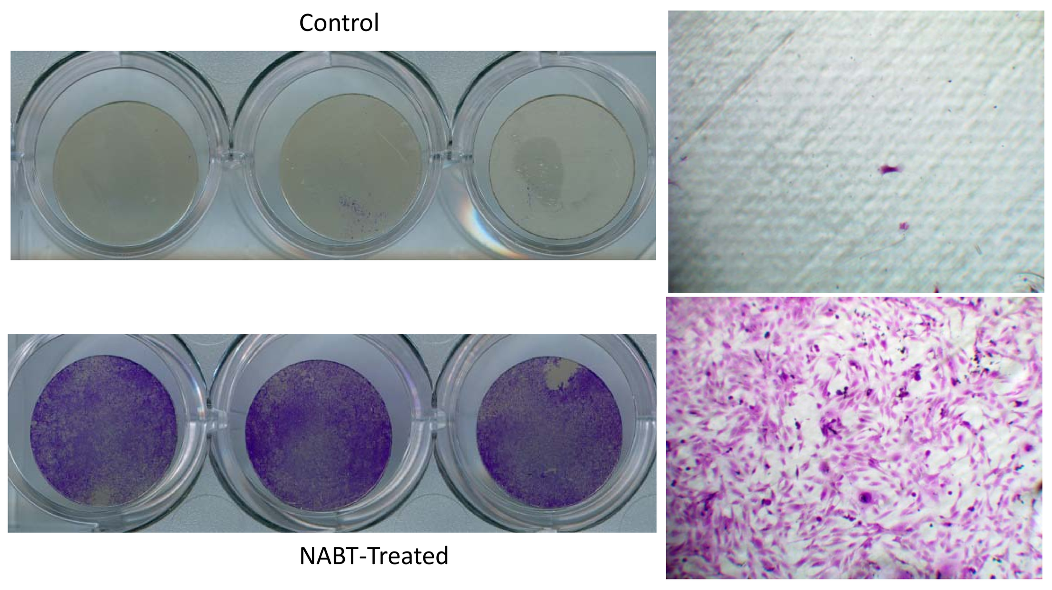
NABT results in increased PEEK Surface Hydrophilicity: By contact angle measurements, we have identified a greatly enhanced hydrophilic surface on NABT-treated PEEK (θ=36.1˚) as compared to controls (θ=76.4˚).



Control NABT-Treated

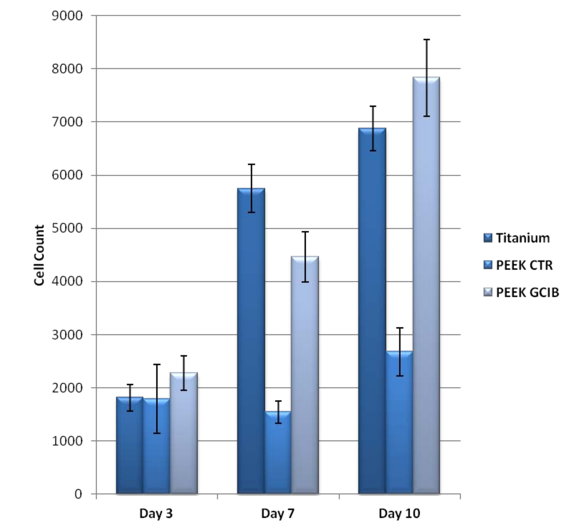
NABT results in significantly increased cell proliferation on PEEK: hFOB cells seeded onto control PEEK results in very little initial cell attachment and subsequently fewer cells over time, however, NABT-treated PEEK results in very good initial cell attachment and subsequently increased cell proliferation over time.

Control



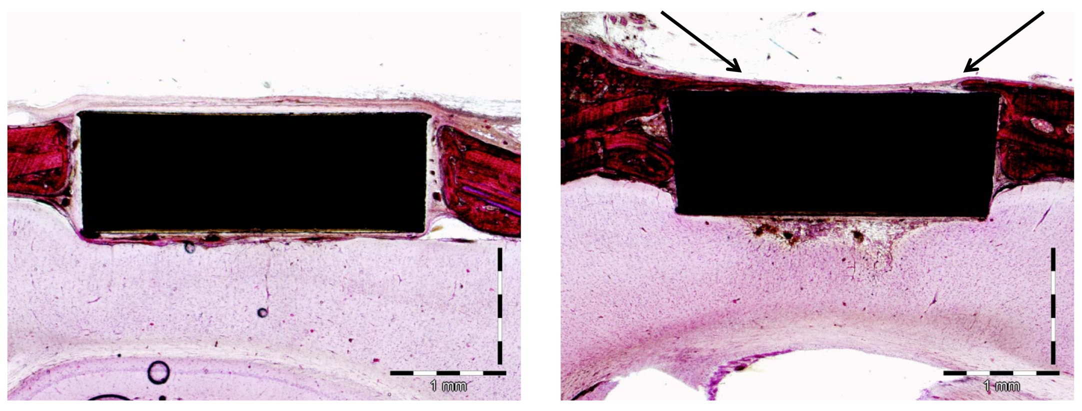
NABT-Treated

NABT-treatment of PEEK results in Comparable cell Proliferation as Bioactive Titanium: hFOB cells growing on surfaces of NABT-treated PEEK display rapid proliferation as compared to control surfaces. NABT results in increasing the bioactivity of PEEK, comparable cell attachment and proliferation is seen on bioactive Ti.



NABT Enhances Bioactivity of PEEK and results in Bone Integration *in vivo*: Rat calvarial defect study revealed NABT-treated PEEK resulted in a bone ledge growing on top of the disk covering approximately 50% of the surface, whereas the control PEEK resulted in only fibrous tissue with no bone growth at all. Further, the controls displayed bone resorption on the sides of the disk whereas good bone purchase is seen on the NABT-treated disks.

CTR NABT



Conclusions

We have created a novel beam processing by utilizing accelerated neutral atom beam. NABT results in a nano-scale texturing and improved hydrophilicity on otherwise chemically resistant PEEK. The resulting processing of PEEK by NABT significantly enhances osteoblast cell attachment and proliferation. These cellular events, therefore, enhance medical device integration into bone. NABT processing would allow for this integration to occur without the use of additional proteins or coatings. NABT results in creating a bioactive PEEK.

Questions or Further Information
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