ORIGINAL RESEARCH

Evaluation of Osteoblastic Activity of Polyether Ether Ketone Modified by Ultraviolet Radiation: An *In Vitro* Study

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Abstract

Purpose: The purpose of this study was to find the effect of ultraviolet (UV) radiation on the osteoblastic activity of polyether ether ketone (PEEK).

Materials and methods: Thirty samples of PEEK discs were made. The samples were grouped as group I ($n = 15$) PEEK with no treatment and group II ($n = 15$) PEEK modified by UV radiation. The experimental group was seeded with human osteoblastic sarcoma cells. The samples were incubated for 48 hours at 37 ± 1°C in a humid atmosphere at 5%. After 48 hours, 2.5% glutaraldehyde was applied to fix the seeded cells to the coverslips. The discs were seen under scanning electron microscopy to evaluate the colony formation and adhesion of osteoblastic cells on the PEEK discs. The observation made was tabulated and subjected to statistical analysis.

Results: Noticeable adhesion of osteoblast was found in the UV-treated PEEK samples when compared to PEEK samples with no treatment. The cells in nontreated PEEK samples were less spread and showed few colonies. PEEK modified by UV radiation showed more noticeable osteoblast cells scattered all around the sample. The adhesion of the cells was better as compared to group I. The difference between the test and control group was statistically significant when analyzed by Fisher's exact test.

Conclusion: PEEK modified with UV radiation showed more noticeable osteoblast cells scattered all around the sample. The adhesion of the cells in UV-treated samples was better as compared to no treatment.

Keywords: Dental implant, Osseointegration, Osteoblastic activity, Polyether ether ketone, Surface treatment, Ultraviolet radiation.

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Introduction

Polyether ether ketone (PEEK) is a synthetic organic polymer. It has tooth-colored appearance, so nowadays being more preferred as dental implant material.[1] It has outstanding resistance to chemicals with good mechanical and biological properties. PEEK is insoluble, has high strength, and a density of 1.32 g/cm^3. It has less modulus of elasticity.[2,3] Due to all these properties it is being preferred as an implant material along with titanium. PEEK can be used in patients hypersensitive to titanium.[4] PEEK is radiolucent, so in a patient where magnetic resonance imaging is required, it can be a better alternative to reduce the artifacts.[5,6] PEEK does not have a metallic color, so it can be a favorable material for dental implants if its properties can be modified accordingly.

PEEK has very limited immanent osteoconductive properties compared to titanium.[7] Researchers had proposed various methods to upgrade the bioactivity of PEEK such as hydroxyapatite coating of the PEEK,[8,9] increase in roughness of the surface,[10] chemical treatment,[11] and addition of particles which are bioactive in nature.[9] Increased temperature during plasma spraying and chemical modification can worsen the properties of the PEEK.[12] PEEK can flake because of their restricted bonding strength.[13]

Substantial research was already performed to increase the bioactivity of PEEK as implant materials.[14,15] The wettability property of the PEEK implant surface can be increased by UV radiation.[16] Studies had presented substantial improvement of retention, attachment, and functional avalanche of osteogenic cells obtained from humans and animals after UV treatment. UV treatment transforms hydrophobic surface of titanium to a more hydrophilic surface and separates adulterate hydrocarbons. Titanium surfaces, which are UV-treated also shows a distinctive electrostatic level and attracts cells directly. The available literature suggests that UV photofunctionalization emerged as a newer approach.[17] The hydrophilization of PEEK improved the osteoconductivity and also proves that osteoconductivity depends on the surface property and not on the material of the implant.[18] Hence the purpose of this *in vitro* study was to find the outcome of UV radiation on the osteoblastic activity of PEEK.

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Materials and Methods

A total of 30 samples of milled PEEK discs of size 15 × 2 mm were fabricated (International Organization for Standardization standard 15309:2013). The samples were grouped as—group I

(n = 15) PEEK with no treatment (Fig. 1) and group II (n = 15) PEEK modified by UV radiation (Fig. 2).

In this study, the surface treatment of PEEK was done under 20°C at a humidity of about 46%. PEEK samples were treated by UV radiation under a UV chamber for 48 hours using a 15W bactericidal lamp with an intensity of λ = 360 ± 20. The samples further were seen under the scanning electron microscope (SEM) to find the roughness of the surface of the PEEK and its topography.

To determine osteoblastic activity, osteoblast was procured and the cells were grown in cell culture lab, and cell adhesion tests were performed on PEEK discs for both groups. The test discs were seeded with human osteoblastic sarcoma cells (1 × 104 cells/cm^2 density). The samples were incubated at 37 ± 1°C in humid atmosphere of 5% carbon dioxide. After 48 hours, 2.5% glutaraldehyde was applied to fix the seeded cells to the coverslips.[19] The discs were seen under scanning electron microscopy to evaluate the colony formation and adhesion of osteoblastic cells on the PEEK discs. The observation made was tabulated and subjected to statistical analysis (Table 1). All the data were subjected to statistical analysis using the Statistical package for the Social Sciences (SPSS) (IBM Corp. released 2019. IBM SPSS for Windows, version 26.0. Armonk, New York: IBM Corp).

Results

Scanning electron microscope (SEM) image of the PEEK discs without surface treatment (group I) showed pits and cracks with few parallel lines on the surface (Fig. 3). PEEK discs treated with UV radiation (group II) showed cracks on the surface (Fig. 4).

In PEEK discs without surface treatment (group 1), SEM analysis showed osteoblastic activity with very few colonies (Fig. 5). In PEEK discs treated with UV radiation (group II)SEM analysis showed the maximum number of colonies with quality cell morphology (Fig. 6). Osteoblastic cells showed better adherence and were prominent and scattered throughout the sample as compared to group I. PEEK treated with UV radiation showed polygonal osteoblastic cells with filopodial attachment and growth under SEM (group II).

Discussion

Nowadays, PEEK has been used as an implant material in the form of abutment, implant and superstructure. PEEK is resistant to chemicals, radiolucent, and has mechanical properties comparable to human bone. It has emerged as a good alternative to a metallic implant.[20,21] PEEK has certain drawbacks as it is bioinert material and has less reaction with the neighboring tissues.

Nonmodified PEEK presents a contact angle of 80–90° with water and it's a hydrophobic value.[16,20,21] In an attempt to overcome this issue, methods have been presented, such as the incorporation of bioactive materials and surface treatment techniques. Surface coating by biomaterials such a hydroxyapatite (HA) and titanium,[22,14] nano-modified HA crystals,[23,24] and modified PEEK can enhance hydrophilicity. Increase in hydrophilicity increases the cellular proliferation with better wettability of the biomaterials and the surface of the implant and thus effects the reaction in between the implant material and the neighboring environment.[11,25]

Plasma spraying produces rough surface layer and also produces a thick appetite layer that might get split in layers and causes failure of the implant.[4] Plasma spray also coats PEEK with HA due to high temperature during the process. This temperature might destroy the PEEK structure because of its electively low melting temperature.[13] Whenever there is a coating on the surface of the implant there is a risk of coating being delaminated, which will affect the osseointegration.

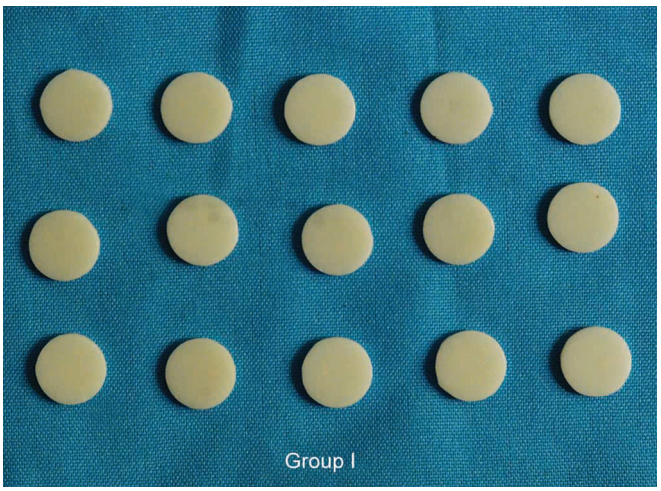

Fig. 1: Group I—PEEK discs without any modification

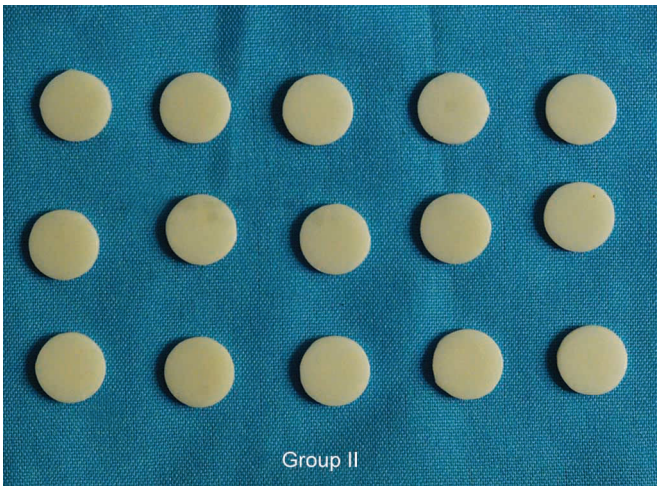

Fig. 2: Group II—PEEK discs modified with UV radiation

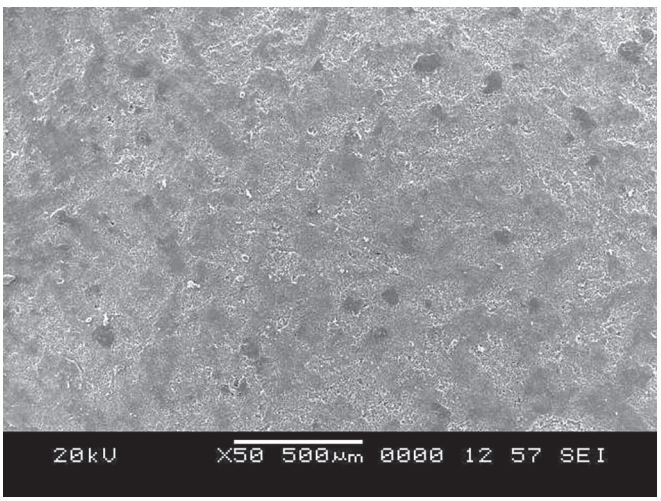

Fig. 3: Group I—few parallel lines, pits and cracks on the surface

Table 1: Statistical analysis using Fisher's exact test

		Osteoblastic cells			
		Less spread of osteoblastic cells	*Increased spread of osteoblastic cells*	*Total*	*Fisher's exact test*
Group II (modified by UV radiation)	N	0	15	15	0.001*
	%	0.0%	100.0%	100.0%	
Group I (no treatment)	N	15	0	15	
	%	100.0%	0.0%	100.0%	
Total	N	15	15	30	
	%	50.0%	50.0%	100.0%	

*$p < 0.05$ is significant

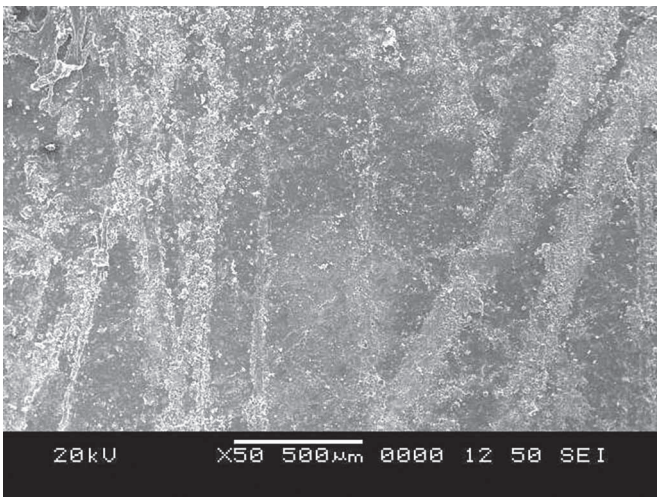

Fig. 4: Group II—showed lines, pits and cracks on the surface

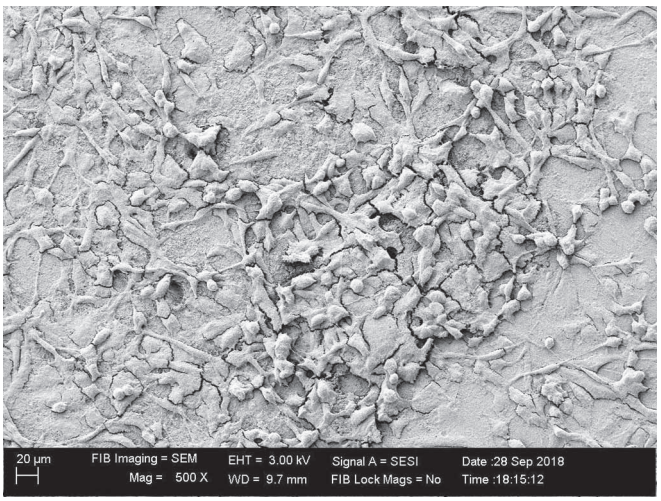

Fig. 6: Group II—osteoblastic cell was very prominent and scattered throughout the sample

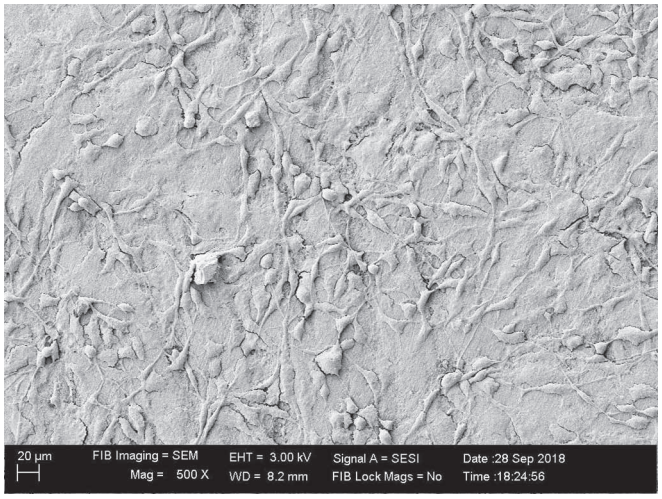

Fig. 5: Group I—osteoblastic activity was seen with very few colonies

Huang et al.,[26] Neiman et al.,[27] and Qahtani et al.[16] demonstrated that unmodified PEEK is bio inert and shows a contact of 80–90°, which is a hydrophobic value. Matheison and Bradley[28] used UV treatment to modify the energy of the PEEK. The results presented increased surface wettability of the treated PEEK by UV. In the present study also, increased osteoblastic activity of PEEK after UV treatment was found, which was similar to the study done by Al Qahtani et al.[16] who reported that the PEEK surface is hydrophilized after UV radiation. Modification of PEEK increases the hydrophilic property of the PEEK. This causes an increase in the proliferations of the cells with better wettability and thus effects the association between the material and the neighboring environment.[16]

Limitations of the study, being this *in vitro* study cannot fully extrapolate into *in vivo* conditions, mainly cell adhesion was studied, cell proliferation and maturation can be evaluated the effect of UV radiation on the osteoconductive potential of PEEK *in vivo* set up should be explored. Osteogenic potential with alkaline phosphatase can be focused along with the evaluation of cytotoxicity. Various other methods available for osteogenic potential evaluation should also be used. Further animal studies can be carried out. *In vivo* conditions, tissue response can be further evaluated.

The main strategies to improve the bioactivity of PEEK should provide an effective way to obtain both mechanical and biological benefits. Further research and clinical trials are required to explore the surface treatment modification that is required to improve osseointegration.

Conclusion

Within the limitations of the study, the following conclusions were drawn:

- The cell adhesion in PEEK without treatment showed less spread of osteoblastic cells and had fewer osteoblastic cell colonies.
- PEEK modified by UV radiation showed more prominent osteoblastic cells that were scattered throughout the samples and showed better adhesion of osteoblast compared to group I.

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