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### **Novel Bioactive Polymer-Apatite Nanocomposite Fibers and their Biocompatibility**

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#### **Publisher's version / Version de l'éditeur:**

*Proceedings of the International Symposium on Polymeric Materials for Regenerative Medicine 2007, 2007-04-02*

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# CYTOTOXICITY OF BIOACTIVE POLYMER-APATITE NANOCOMPOSITE FIBERS STERILIZED BY ETO AND PLASMA

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## Introduction

Polyethylene terephthalate (PET) reinforced with hydroxyapatite (HA) nanoparticles in the form of fibers is a new biomaterial developed for bone fixation surfaces for total hip replacement prosthesis. The sterilization of these novel materials with the use of accepted protocols and minimal effects on their established biocompatibility<sup>1</sup> is a fundamental requirement for their biomedical application. This work evaluated the HA nanoparticles effects on the polymeric fibers surface chemistry by XPS analysis, and compared the effects induced by ethylene oxide (EtO) and low temperature plasma (LTP) sterilization on the composites chemical composition by XPS and *in vitro* cytotoxicity.

## Experimental

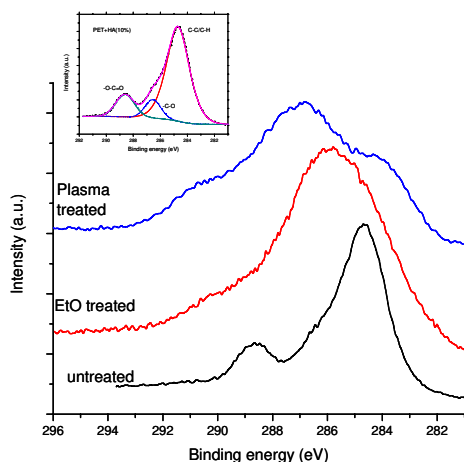
The PET fibers (made by twin screw compounding and melt blowing<sup>2</sup>) chemical composition with increasing amounts of HA nanoparticles (Plasma Biotal, UK) was analyzed by XPS and FTIR. The surface modifications of the composites, before and after LTP and EtO sterilization, were assessed by XPS. The Alamar Blue assay was performed after direct contact of the LTP- and EtO- treated fibers scaffolds with L929 fibroblasts for different time periods. The release of the inflammatory cytokine, tumor necrosis factor-alpha (TNF- $\alpha$ ) from RAW 264.7 macrophages in the presence of LTP- and EtO- treated fibers was used as a measure of the inflammatory response.

## Results and Discussion

The XPS analysis of the untreated PET/HA nanocomposite fibers revealed that HA was not exposed to the surface. However, the surface layer of the fibers increased in O concentration with increasing loading of HA (for a maximum of 12% O increase at 10% HA), mainly due to the O-C=O bond increase. XPS analysis of the fibers charged with 10% HA (PET10) before/after LTP and EtO sterilization revealed the following chemical modifications: EtO treatment induced alkylation, as seen in the increase on the C-O peak despite the constant overall O % (refer to Table 1 and Figure 1); LTP treatment induced some etching and an increase in the C-O/C-OH bonds and overall O% increase, which lead to a better biologic response as seen with the TNF- $\alpha$  release.

**Table 1** – XPS atomic composition of Oxygen and Carbon bonds

PET 10 fibers	Oxygen (%)	Carbon bonds		
		C-C/C-H	-C-O	-O-C=O
Untreated	30.8	52.5	6.9	9.8
EtO	29.8	34.2	27.1	8.9
Plasma	35.3	19	33.5	12.1



**Figure 1** – XPS analysis of the PET10 fibers composite

The *in vitro* cytotoxicity to L929 fibroblasts cells was evaluated after the LTP and EtO treatments. Despite the resulting surface modifications, the cell viability of both LTP and EtO-treated fibers remained similar. Following macrophages incubation with the fibers, a trend of higher TNF- $\alpha$  release by the EtO-treated polymer (296 pg/ml), as compared to the LTP sterilized ones (88 pg/ml), was observed. This trend suggest a higher inflammatory potential for the EtO sterilized fibers.

## Conclusion

HA loading of fibers increases the overall O content of composites. EtO treatment induces the production of C-O group as residues absorbed unto the fiber surface with no overall O increase. LTP treatment leads to an optimization of the C-O / C=O ratio through a chemical reaction (as a covalent bond), leading to better biologic response as seen by the inflammation potential decrease. Despite the acceptable performance of the EtO sterilized fibbers scaffolds, the LTP treatment appears to be a more suitable one due to its low inflammatory potential, as well as the clinical relevance.

## References

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2. A. Ajji, J Denault, M.N. Bureau, M.T. Ton-That, Trudel-Boucher and D. Côté; presented at Polymer Nanocomposites Fibers and Applications Annual Technical Conference ANTEC 2006 (SPE), May 7-11 2006 Charlotte (USA)