GUIDED TISSUE REGENERATION: IN VITRO BIOCOMPATIBILITY OF A NOVEL MEMBRANE OF THE COMPOSITE POLY(VINYLIDENE-TRIFLUOROETHYLENE)/BARIUM TITANATE

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INTRODUCTION: Guided tissue regeneration (GTR) is a technique that uses a barrier membrane, which can allow the repopulation of local sites by their own cells. This technique has been applied in clinical dentistry to various cases including dental implant therapy. The most popular membrane used for GTR is an expanded polytetrafluoroethylene (e-PTFE) membrane and some investigators have demonstrated successful bone regeneration by using this material [1-2]. Despite the advantages of this membrane, alternative materials could be employed in GTR. This study aimed at investigating the in vitro biocompatibility of a novel membrane of the composite Poly(vinylidene-trifluoroethytele)/Barium titanate (PVLTrFE).

METHODS: Human alveolar bone fragments (explants) were obtained from healthy donors, under approved research protocols of Committee of Ethics in Research. Osteoblastic cells were obtained from these explants by enzymatic digestion and cultured in α-MEM supplemented with 10% fetal bovine serum, 50 μg/ml gentamicin, 0.3 μg/ml fungizone, 5 μg/ml ascorbic acid, 7 mM β-glycerophosphate, and 10^{-7} M dexamethasone until subconfluence. Cells from first passage were subcultured ($2x10^4$ cells/well) on PVLTrFE and e-PTFE (control) membranes in 24-well culture plates. During all the culture period, cells were maintained at 37ºC, 5% CO_2 and 95% air, and the medium were changed every 3 or 4 days. For attachment evaluation, cells were incubated for 24 hours, enzimatically released and counted. Data were expressed as percentage of adherent cells. For proliferation, cells were cultured for 1 and 10 days, enzimatically released and counted. Data were expressed as doubling time in hours [3]. At 14 days, ALP activity was measured using a commercial kit (Labtest) and expressed as μmol thymolphtalein/h/mg protein. Bone-like nodule formation was stained by alizarin red at 21 days and examined by epiluminescence under a conventional fluorescence microscope. Data were compared by Student t-test.

RESULTS: Data are shown in Table 1. Cell attachment (p=0.001) and ALP activity (p=0.0001) were greater on PVLTrFE membrane. Doubling time was greater on PVLTrFE membrane (p=0.03), which indicates a decreased proliferation rate. Bone-like nodule formation occurred only on PVLTrFE membrane (Fig. 1).

Table 1. Attachment, proliferation, and ALP activity of osteoblastic cells cultured on PVLTrFE and e-PTFE membranes. Data were expressed as mean ± standard deviation (n=5).

	PVLTrFE	e-PTFE
Attachment	62.5±11.4	25.0±8.8
Doubling time	69.2±8.0	55.5±7.8
ALP activity	23.8±3.8	4.32±1.6

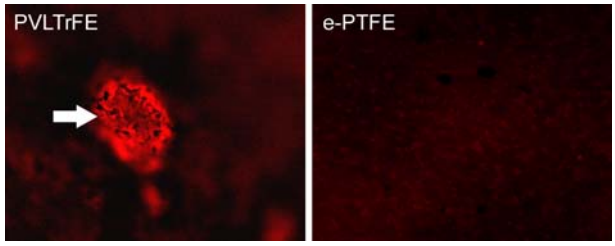

Fig. 1. Photomicrograph of membranes stained by alizarin red. Bone-like nodule formation was observed only in PVLTrFE membrane (arrow). Objective 10X.

DISCUSSION & CONCLUSIONS: The present results showed that both membranes are biocompatible. However, PVLTrFE presented a better in vitro biocompatibility when compared with e-PTFE. Also, PVLTrFE allowed bone-like nodule formation, the final event of in vitro osteogenesis. Therefore, PVLTrFE membrane could be an alternative to be used in GTR.

REFERENCES: [1] R.K. Shenck, et al (1994) *Int J Oral Max Impl* **9**: 13–29. [2] C.H.F. Hämmerle, et al (1998) *Int J Oral Max Impl* **13**: 522–30. [3] M.K. Patterson Jr (1979) *Methods Enzymol* **58**: 141-52.

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