THE EFFECTS OF DNA EXTRACTS FROM UROLOGICAL TISSUE MATRICES

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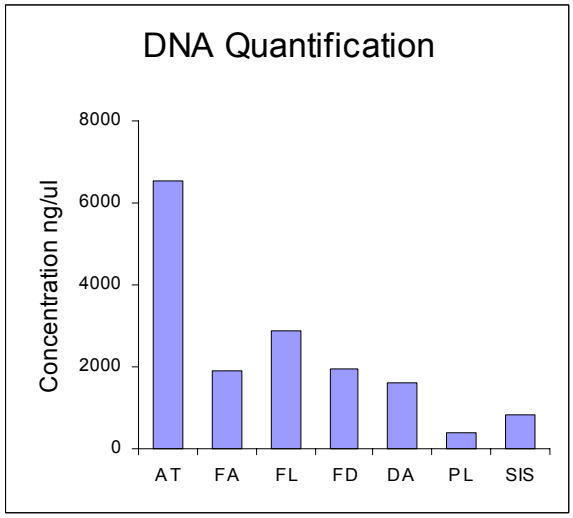
Statement of Purpose: Allografts and xenografts are widely used in urological surgeries to treat incontinence and in pubovaginal sling surgeries. Autografts are a good choice as there is a smaller chance of graft rejection, but postoperative pain and morbidity in some cases that are associated with autografts make it a less attractive option. Allografts are obtained from cadavers after careful screening of medical and social history. Allografts are mostly harvested from the dermis or the fascia. Xenografts are obtained from pig, harvested from the small intestinal submucosa (SIS) or from dermis. There are several commercial sources which procure these tissues. All of these procured tissues have to maintain the standards lineated by the American Association of Tissue Banks (AATB)(1). Allografts and xenografts are processed and sterilized using different patented techniques, which try and eliminate the cellular content and inactivate infection/ disease causing agents like bacteria,virus and prions. Inspite of extensive efforts to reduce risk of disease transmission, there have been several reports of HIV transmission via solid organ and tissue transplantation(2). There is some literature available on the risk of disease transmission in bone allografts but not much documented evidence of disease in soft tissue allografts. There have been limited studies to isolate genetic material in processed tissues (1,3). In this experiment, we have determined the presence of DNA and conducted cell proliferation assay in seven tissue samples being clinically used as implants for urological reconstruction..

Methods: We evaluated seven different tissue samples from 4 different companies. They are 1.Small intestinal submucosa (SIS) (Cookbiotech), 2.Tutoplast Fascia lata(FL) (Mentor Corp.) and 3.Tutoplast fascia dermis (FD) (Mentor Corp.) 4. Acellular tissue graft (AT) (LifeCell Corp.) 5. Pelvicol (PL) (C. R. Bard),6. Dermal allograft (DA) (C. R. Bard) and 7. Faslata allograft (FA) from (C. R. Bard). Each of these samples were processed using a standard phenol chloroform DNA extraction method to isolate the DNA. The genetic material obtained was quantified using the spectrophotometer and the DNA purity was assessed through the agarose gel electrophoresis. Cell response to the extracted DNA was examined using the MTT assay . Fibroblast cells and the isolated genetic DNA were incubated for 48 hours to gauge the cell response.

Result: Of the seven samples of tissues DNA was isolated from all of them (100%). The concentration of DNA in AT was significantly higher then all the other six samples (Figure). This maximum value was followed by FL,FD, FA,DA, SIS and PL. The MTT cell proliferation assay determined that there was a variation in proliferation rate of the fibroblasts with varying concentrations of DNA. Fibroblast stimulate index did increase with specific DNA concentrations and cell proliferation declined considerably at DNA concentrations of 5 ng/µl.



Discussion: The quantity of the DNA extracted varied depending on the processing and sterilization techniques of the tissues. Through processing, tissues are claimed to be devoid of cells. However the genetic material can still be present and this can represent an inherent risk of disease transmission. We displayed that the DNA extracted from these tissue implants caused a significant proliferation of fibroblasts. These cells are vital in the production of scar tissue and foreign body reactions. The scope of research will be extended to study the response of various other cells and proteins present in the immediate surrounding of the urological implant in the body. Introduction of genetic material within the implanted tissues may be partly responsible for early failure and rejection of the tissue by the host, as well as transmission of pathogenic DNA.

Conclusion: There is presence of intact genetic material in the commercially available allografts and xenografts that causes proliferation of surrounding cells.

References:

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