Novel Tissue Processing Technique Conserves Native Soluble Proteins

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Introduction:

The concept of regenerative therapy requires a scaffold, soluble factors, and cellular constituents. This is a well-developed and broadly accepted approach for the recovery of chronic injuries across an array of medical practices[1-3]. Often, synthetic or non-human scaffolds are employed, which by nature lack appropriate human-derived soluble factors including, growth factors, chemokines, and cytokines[4]. Generally, the use of resident cells in the tissue of interest is valuable in infiltrating and functioning in the appropriate scaffold embedded with soluble factors[5]. Several commercially available scaffold products claim the presence of key soluble factors, though have been refuted by others[6]. While native tissue is known to contain many of these factors natively, harsh processing methods including chemical treatments, dilutive washes, freezing, and dehydration nullify their function[7-9]. As a result, final tissue scaffold products often lack the necessary soluble factors to achieve optimal results compared to the "fresh" tissue counterpart. Here, we seek to characterize the difference in total soluble protein available in a placental soft tissue compared to that processed via the "Nativus" method.

Method:

Freshly recovered placental connective tissue was compared to the equivalent tissue from the matched donor processed via Nativus. In this assay, fresh, rinsed (2x saline washes) and Nativus processed tissue of the equivalent weight (per mg) was placed in 1mL sterile water and allowed to elute soluble protein over an established time course, spanning 30, 90 and 270 minutes. At each interval, a 90µl aliquot was collected from the liquid was collected and incubated with Coomasie (Thermo Scientific, Cat# 23236) per manufacturer's instructions. Total protein was quantified via Molecular Devices EMAX Plus plate reader at 595nm with the recommended background subtraction wavelength.

Results:

Compared to the equivalent fresh placental connective tissue from a matched donor, the Nativus processed tissue displayed an accelerated rate of protein elution over the established time course reaching equilibrium by 90 minutes, demonstrating the retention and saturation of soluble constituents in the processing method. Importantly, rinsed tissue showed reduced elution of total soluble factors and at a slower rate compared to the fresh and Nativus-processed counterparts, indicating the respective levels of bioavailable soluble factors (Figure 1). Statistical significance is not represented due to the nature of the report, which represents an n of 1.

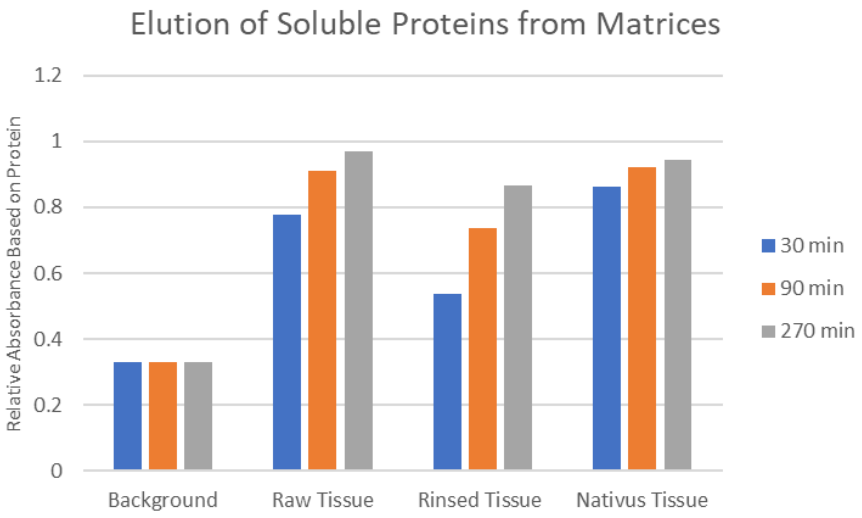
Discussion:

This brief pilot demonstrates that the Nativus process preserves soluble factors from the native environment. This rapid elution of factors, which are suspected to include cytokines and chemokines, are superior to conventional, highly processed commercial tissue products.

Significance:

Ongoing IRB approved studies are underway, comparing Nativus-processed tissue products to the historical performance of highly processed alternative commercial products. Initial clinical data, paired with this analysis, illustrates the novelty of Nativus processed tissues and provides insight as to how these products can recover chronic wounds at an unprecedented rate.

Figure 1:



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