The Role of Biofilm in Hyaluronic Acid Filler – An In Vitro Study

Mayuran Saththianathan BHB MBChB MS, Anita Hegde MBChB, Karen Vickery BVSc, MVSc, PhD, Anand K Deva BSc(Med), MBBS, MS.

Introduction

Soft tissue filler injections have been associated with a 0.01-0.1% risk of granuloma formation\(^1\). This granuloma may form between 2 weeks to 2 years post injection. In the past, this was considered a type IV hypersensitivity reaction\(^2\). Recent evidence, however, has suggested that this may be due to bacterial infection\(^3\).

Subclinical infection due to the formation of bacterial biofilms (a mixture of low numbers of bacteria protected in their surrounding excreted polysaccharide) has been shown to cause chronic inflammation around prosthetic material and foreign bodies. We have developed an in vitro assay to study the likelihood of biofilm formation around commonly used soft tissue fillers. In this study we compared the ability of *Staphylococcus epidermidis* and *Staphylococcus aureus* to attach to and form a bacterial biofilm on Hyaluronic acid.

Methods:

This in vitro model utilized 0.05ml of hyaluronic acid filler, which was mounted on a glass coupon and subsequently inoculated with \(10^2\) *S. epidermidis* and 100µL of 100% tryptone soy broth. The filler was incubated for 16 hours at 37°C. Samples were processed with live/dead staining and fixation with 4% Paraformaldehyde. Specimens were analysed with confocal lasar scanning microscopy (CSLM), scanning electron microscopy (SEM) and sonication with culture.

Results:

CSLM demonstrated the formation of a mature bacterial biofilm, with *S. epidermidis* embedded within the filler matrix. SEM illustrated that this bacterial biofilm with its associated exopolysaccharide integrating with the filler matrix (figure 1).

![Figure 1](image-url) The above SEM image demonstrates bacterial biofilm formation on the hyaluronic acid filler matrix, with strands of exopolysaccharide connecting the bacteria to the filler, surface and each other.
Figure 2 This image demonstrates the integration of the bacterial exopolysaccharide into the hyaluronic acid filler matrix.

**Conclusion:**

We have shown that bacterial biofilm can grow within hyaluronic acid filler and integrates into the filler matrix itself. This process occurs within 24 hours of inoculation. These data support the hypothesis that biofilm in injected fillers can produce indolent infection. Further testing is currently being performed on Poly-L-lactic acid and Polyacrylamide gel fillers, which will be included at the time of presentation.

**References:**


**Disclosure/Financial Support**

Supported by the Australian School of Advanced Medicine, Macquarie University. None of the authors has a financial interest in the products discussed in this manuscript.