Differential Effects of Four Antimicrobial Agents on the Size and Biomechanics of Tendon Grafts

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INTRODUCTION: One of the intraoperative challenges during an anterior cruciate ligament (ACL) reconstruction is intraoperative graft contamination, which can occur through graft dropping or contamination during graft harvesting [1,7], potentially making the graft a bacterial carrier [1,2]. Though the instances of this situation are rare [1,2,7], surgeons must have a plan to eliminate such contamination. Cleansing the graft is a popular option [3]. Previous studies on antiseptic solutions revealed that povidone iodine was found to be inferior to chlorhexidine as a decontaminant [4]. Another review study highlighted that soaking the graft in vancomycin could reduce the incidence of septic arthritis following ACL reconstruction [5]. Teicoplanin was shown to offer comparable efficacy to vancomycin with a lower occurrence of side effects such as nephrotoxicity in clinical use [6]. In addition to decontamination, it is also important to consider the effects of antimicrobial solutions on the biomechanical properties of the graft. In one study, both chlorhexidine and vancomycin did not compromise the tensile properties of the graft [3, 7]. However, povidone iodine, chlorhexidine, vancomycin, and teicoplanin have not been directly compared. Therefore, the objective of this study is to compare the impact of different antimicrobial solutions on the size and biomechanical properties of porcine flexor tendon allografts.

METHODS: Deep digital flexor tendons were harvested from both left and right porcine hindlimbs at age of 6 weeks. 10% povidone iodine solution, 4% chlorhexidine gluconate solution, 400mg teicoplanin / 100mL 0.9% saline solution, and 500mg vancomycin / 100 mL 0.9% saline solution were prepared. Tendons from one side underwent a 30-minute soaking in these solutions, while the other side was wrapped in gauze soaked in phosphate-buffered saline (PBS), serving as the contralateral control (n=12 for each solution). 3D models of tendons were obtained before and after soaking, along with the contralateral control, using the EinScan-SP 3D scanner (Shining 3D, Hangzhou, China). Cross-sectional area (CSA) of the middle layer was visualized, and CSA values were calculated as the average CSA of the middle 50% of each 3D tendon mode by custom MATLAB codes. Grip-to-grip tensile testing was performed by Instron Materials Testing System (Norwood, MA) with a 2 kN load cell and pneumatic clamps (6 bar). The specimens were loaded from 5N to 3% grip-to-grip strain for 10 cycles as preconditioning, followed by a tensile load to failure at a constant rate of 0.1mm/s. Paired t-test was performed to assess the statistical differences between soaked and contralateral tendons. The significance level was set at 0.05.

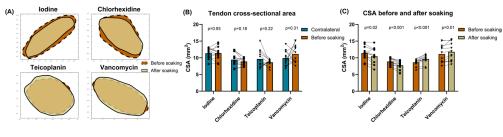
RESULTS: Differences in CSA before and after soaking were observed visually (Fig. 1A) and quantified (Fig. 1B & C). Side-to-side consistency in CSA was observed between the samples before soaking and their contralateral controls (Fig. 1B). Soaking the same samples in iodine and chlorhexidine led to a significant decrease in CSA by 8.2% and 12.8%, respectively (Fig. 1C). Conversely, teicoplanin and vancomycin increased the flexor tendon CSA by 11.8% and 6.1% after soaking (Fig. 1C). The stiffness values of samples soaked in chlorhexidine (146.2 \pm 19.8 N/mm) were significantly smaller than the contralateral controls (159.5 \pm 19.2 N/mm) (Fig. 2A), while no changes in stiffness were observed for the other three solutions. Modulus and load at failure were similar between the soaking samples and contralateral controls for all solutions (Fig. 2B & C).

DISCUSSION: Our study showed changes in size of porcine deep digital flexor tendons after soaking by four different antimicrobial solutions, but changes in tensile properties were only observed in response to chlorhexidine. While earlier research had indicated negligible effects of chlorhexidine on the structural properties of human patellar tendon allografts [1], our findings indicated that stiffness of the soaked specimens was inferior to their contralateral counterparts. This distinction may arise from the absence of paired soaked and control samples in prior work or different graft processing. Specimens in prior work underwent a standard irradiation and washing protocol before testing, while ours were freshly frozen with gauze soaked in PBS. Iodine-soaked allografts exhibited comparable tensile properties to their contralateral controls. However, CSA values decreased following the soaking process. The equivalent biomechanical properties and increased CSA values after soaking by teicoplanin and vancomycin suggested that these two solutions are recommended options when cleansing allograft for decontaminant purpose. A limitation of this study was that most tendons failed at clamp sites rather than the mid-tendon region due to the high failure load. Therefore, ultimate tensile strength values were excluded. Future work will be required to assess how allograft after soaking in different antimicrobial solutions will influence the initial joint stability after ACL reconstruction.

SIGNIFICANCE/CLINICAL RELEVANCE: Analyzing the impact of various antimicrobial solutions used for cleansing allografts can provide valuable insights to surgeons considering selections for graft decontamination, which may allow for promising graft biomechanics before surgeries.

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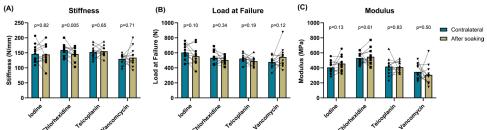


Fig. 1 Soaking tendon in iodine and chlorhexidine solutions led to a reduction in cross-sectional area (CSA) while teicoplanin and vancomycin resulted in an increase. (A) Difference in cross-sectional area before and after soaking for representative tendon samples in each solution. (B) Contralateral control vs. samples before soaking. (C) CSA of the same sample before and after soaking. Mean values shown as bars with paired samples connected. Statistical results shown in graphs.

Fig. 2 Differences in stiffness (A), load at failure (B), and modulus (C) between contralateral controls and soaking samples were quantified for iodine, chlorhexidine, teicoplanin, and vancomycin solutions. Mean values shown as bars with paired samples connected. Statistical results shown in graphs.