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Histologic and Biomechanical Evaluation of Crosslinked and Non-Crosslinked Biologic Meshes in a Porcine Model of Ventral Incisional Hernia Repair

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Abstract

BACKGROUND—The objective of this study was to evaluate the biomechanical characteristics and histologic remodeling of crosslinked (Peri-Guard, Permacol) and non-crosslinked (AlloDerm, Veritas) biologic meshes over a 12 month period using a porcine model of incisional hernia repair.

STUDY DESIGN—Bilateral incisional hernias were created in 48 Yucatan minipigs and repaired after 21 days using an underlay technique. Samples were harvested at 1, 6, and 12 months and analyzed for biomechanical and histologic properties. The same biomechanical tests were conducted with de novo (time 0) meshes as well as samples of native abdominal wall. Statistical significance ($p < 0.05$) was determined using 1-way analysis of variance with a Fisher's least significant difference post-test.

RESULTS—All repair sites demonstrated similar tensile strengths at 1, 6, and 12 months and no significant differences were observed between mesh materials ($p > 0.05$ in all cases). The strength of the native porcine abdominal wall was not augmented by the presence of the mesh at any of the time points, regardless of de novo tensile strength of the mesh. Histologically, non-crosslinked materials showed earlier cell infiltration ($p < 0.01$), extracellular matrix deposition ($p < 0.02$), scaffold degradation ($p < 0.05$), and neovascularization ($p < 0.02$) compared with crosslinked materials. However, by 12 months, crosslinked materials showed similar results compared with the non-crosslinked materials for many of the features evaluated.

CONCLUSIONS—The tensile strengths of sites repaired with biologic mesh were not impacted by very high de novo tensile strength/stiffness or mesh-specific variables such as crosslinking. Although crosslinking distinguishes biologic meshes in the short-term for histologic features, such as cellular infiltration and neovascularization, many differences diminish during longer periods of time. Characteristics other than crosslinking, such as tissue type and processing conditions, are likely responsible for these differences.

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Incisional hernias cause complications in up to 20% of patients who have had a previous laparotomy.¹⁻³ Primary suture repair is associated with unacceptably high recurrence rates, ranging from 31% to 54%.^{4,5} Repair with synthetic mesh materials has reduced recurrence rates to between 2% and 36%, and is now considered standard of care for elective ventral incisional hernia repair.⁴⁻⁷ However, any prosthetic implanted in the abdominal wall can be a source of complications. The risk of surgical site infection in patients undergoing first-time incisional hernia repair is as high as 5% to 30%.⁸ Definitive management of postoperative mesh infections sometimes requires mesh removal, but this can lead to a cycle of hernia recurrence and need for subsequent revision. Recently, there has been an exponential increase in the number of commercially available meshes with properties designed to minimize complications associated with high-risk surgical wounds. A single mesh that can be used in all scenarios has not yet been identified. However, a new class of biologic meshes has recently been developed, and these materials can be used in contaminated fields.

Acellular biologic materials derived from human or animal sources are a relatively new technology in which tissues such as dermis, intestinal submucosa, and pericardium are rendered free of cells and immunogenic moieties, leaving behind only a scaffold of extracellular matrix (ECM), in which host cells can repopulate.^{9,10} Once implanted, host collagen is deposited, and the implanted ECM is degraded as the process of remodeling commences. As such, a biologic mesh should allow remodeling to occur and still maintain enough mechanical integrity to prevent hernia recurrence. To this end, many biologic meshes are subjected to a chemical crosslinking step to render the collagen of the biologic mesh less prone to degradation in vivo. Although several authors have demonstrated the use of crosslinked and non-crosslinked materials in preclinical models,¹¹⁻¹⁷ long-term clinical outcomes data on the use of these products in humans for ventral hernia repair are not yet available.

The purpose of this study is to compare biomechanical properties and histologic remodeling profiles of crosslinked and non-crosslinked biologic meshes in a porcine model of incisional hernia repair. Due to differences in source tissue types and subsequent crosslinking, we hypothesize that there will be detectable differences in both physical properties and histologic responses to these materials at interval time points of 1, 6, and 12 months in vivo.

METHODS

This study was conducted under a protocol approved by the Washington University School of Medicine Animal Studies Committee. Forty-eight female Yucatan minipigs were used for this study. Animals were housed, fed, and handled according to the Guide for the Care and Use of Laboratory Animals and standard protocols on file with the Washington University Division of Comparative Medicine.¹⁸ Strict sterile conditions were maintained intraoperatively. Biologic meshes used in this study included Peri-Guard (crosslinked bovine pericardium; Synovis Surgical Innovations), Permacol (crosslinked porcine dermis; Covidien), Veritas (non-crosslinked bovine pericardium; Synovis Surgical Innovations), and AlloDerm (non-crosslinked human dermis, LifeCell Corporation).

A porcine model of ventral hernia repair was used by creating bilateral abdominal wall defects in Yucatan minipigs and repairing these defects with a preperitoneal technique 21 days after hernia creation. Bilateral longitudinal incisions measuring 5 cm were made through the skin, subcutaneous fat, fascia, and aponeurotic muscle layers into the preperitoneal fat, but not through the peritoneum. The abdominal wall musculature and fascia were left open. The subcutaneous fat and areolar tissue were reapproximated with interrupted 3-0 polydioxanone suture (Ethicon), and the skin was closed with interrupted subcuticular 3-0 polydioxanone suture (Ethicon). Cyanoacrylate-based dermal glue was used

to seal the incisions to provide a barrier to fluid and fecal contamination for at least 48 to 72 hours postoperatively. Postoperative antibiotic prophylaxis was provided as oral cephalexin dosed at 20 to 25 mg/kg every 12 hours for a total of 5 days.

The abdominal wall defects were allowed to mature for 21 days, followed by repair with biologic meshes (8 × 10 cm). Manufacturer's instructions for use were followed for handling and manipulation of the meshes. The repair was performed by opening the previously created abdominal wall defect and dissecting down to the underlying peritoneum. The biologic meshes were positioned bilaterally in the preperitoneal/retromuscular space because this represents a more vascularized environment than the mesothelial environment associated with intraperitoneal placement. Meshes were oriented with the long edge (10 cm) running axially and the short edge (8 cm) running transversely. They were secured with 8 circumferential transfascial interrupted #0 Prolene sutures (Ethicon) placed approximately 3 cm apart and at least 1 cm from the mesh edge. An overlap of 2 to 3 cm was provided circumferentially between the mesh-abdominal wall interface. The hernia sac was closed with interrupted #0 polydioxanone suture (Ethicon). All incisions were closed with a double layer of interrupted 3-0 polydioxanone suture (Ethicon) and sealed with cyanoacrylate-based dermal glue. Postoperative care was identical to that of the hernia creations.

After survival of 1, 6, or 12 months, animals were sedated with intramuscular telazol (4 mg/kg), xylazine (2 mg/kg), and ketamine (2 mg/kg). Euthanasia was then performed by administration of IV pentobarbital (100 mg/kg) or by administration of IV potassium chloride (>100 mg/kg IV) under anesthesia. After euthanasia, the abdomen was opened along the midline, and the mesh samples were visually inspected for in situ appearance. The abdominal wall was harvested en bloc and trimmed to one muscle layer of thickness to allow for specimens to fit into grips for tensile testing. The peritonealized transversus abdominis muscle layer was used for all specimens. As the borders of the implanted meshes were ill-defined due to their placement in the preperitoneal plane, and as a result of remodeling over time, we used the center of each repair site to ensure that biologic mesh was uniformly included in all specimens. The biologic mesh itself was not dissected out of the abdominal wall for testing. Instead, the specimens consisted of the entire repair site, including both the mesh and the associated abdominal wall tissue as one composite specimen. A 4 × 4 cm specimen of the repair site was recovered, and a 1 × 4 cm strip was reserved for histologic testing, leaving the remaining 3 × 4 cm piece of repair site composite (ie, mesh plus abdominal wall tissue) for tensile testing.

Tensiometry was conducted using an Instron Series 5542 Universal Testing System (Instron). Each specimen was oriented vertically with each end secured inside the grips and tested to failure at a rate of 0.42 mm/s (1 in/min). The maximum load sustained by the specimen was recorded in Newtons (N), and the tensile stress was calculated by dividing the maximum load sustained by the material by the width of the specimen (N/cm). The approximate stiffness (N/mm) was calculated from the slope of the force versus displacement curve in the linear region of the curve. Eight specimens were tested for each type of implanted mesh (n = 8). Specimens of the native porcine abdominal wall away from the hernia repair sites (n = 10), as well as time 0 (de novo) specimens of each type of mesh (n = 8), were tested in an identical fashion for comparison with the biomechanical properties of the repair sites.

A 1 × 1 cm piece of mesh-tissue composite was embedded in paraffin for histologic analysis. Thin sections were stained with hematoxylin and eosin and analyzed under light microscopy at 40×, 100×, and 200× magnification by a veterinary pathologist. Five to 10 non-overlapping fields per specimen were evaluated at 100× magnification and graded for cellular infiltration, cell types present, ECM deposition, scaffold degradation, fibrous

encapsulation, and neovascularization according to a scale adapted from Valentin and colleagues¹⁹ and presented previously.²⁰ Higher scores on this scale represent more favorable outcomes with respect to graft remodeling, greater cellular infiltration, deposition of ECM, scaffold degradation, and neovascularization with low levels of inflammation and fibrous encapsulation conferring the highest scores. A composite histologic score was also calculated for each sample by taking the average of the scores in each of the 6 subcategories mentioned here.

Statistical analysis

Data were analyzed by a 1-way analysis of variance, followed by a Fisher's least significant difference post-test to determine whether significant differences were observed. A *p* value <0.05 was considered statistically significant. Data are presented as mean $\pm$ standard error of mean.

RESULTS

De novo biologic meshes subjected to uniaxial tensile testing sustained maximum loads as follows: Veritas, 89.6 ± 9.7 N; AlloDerm, 253.0 ± 15.2 N; Permacol, 317.2 ± 23.6 N; and Peri-Guard, 169.5 ± 18.3 N (Fig. 1A). Tensile stress was calculated by dividing the maximum load sustained by the width of the specimen, neglecting thickness. A wide range of values were observed: Veritas, 29.9 ± 3.2 N/cm; AlloDerm, 84.3 ± 5.1 N/cm; Permacol, 105.7 ± 7.9 N/cm; and Peri-Guard, 56.5 ± 6.1 N/cm (Fig. 1B). De novo Permacol was significantly stronger than all other meshes; de novo AlloDerm was significantly stronger than de novo Peri-Guard and de novo Veritas; and de novo Peri-Guard was significantly stronger than de novo Veritas (*p* < 0.01).

The stiffness of all 4 de novo biologic meshes was also calculated from the slope of the linear region of the force versus displacement curve. De novo meshes exhibited stiffnesses of: Veritas, 10.0 ± 1.3 N/mm; AlloDerm, 18.2 ± 1.3 N/mm; Permacol, 58.3 ± 4.0 N/mm; and Peri-Guard, 34.8 ± 2.7 N/mm (Fig. 1C). Again, significant differences were observed between the 4 meshes (*p* < 0.05 for all comparisons). De novo Permacol was significantly stiffer than all other meshes; de novo Peri-Guard was significantly stiffer than de novo AlloDerm and de novo Veritas; and de novo AlloDerm was significantly stiffer than de novo Veritas (*p* < 0.01).

All 4 meshes were significantly stronger and stiffer at time 0 compared with their corresponding repair sites (mesh-abdominal wall tissue composites) after 1, 6, or 12 months (*p* < 0.01 for all comparisons). In addition, although significant differences were observed between the strengths and stiffnesses of the 4 meshes at time 0, no significant differences were detected between mesh-repaired sites at 1, 6, or 12 months due to the type of mesh used to repair the defect (*p* > 0.05 in all cases). In addition, no significant differences in strength or stiffness of the repair site were detected over time for any of the meshes used (*p* > 0.05 in all cases).

As demonstrated in Figure 1, mesh-repaired sites sustained maximum loads of approximately 20 to 30 N for Veritas, 14 to 37 N for AlloDerm, 23 to 35 N for Permacol, and 29 to 40 N for Peri-Guard during the course of the study. Tensile stresses were approximately 7 to 10 N/cm for Veritas, 5 to 13 N/cm for AlloDerm, 7 to 12 N/cm for Permacol, and 9 to 14 N/cm for Peri-Guard, and stiffnesses were approximately 0.9 to 2.6 N/mm for Veritas, 0.6 to 3.6 N/mm for AlloDerm, 1.1 to 5.8 N/mm for Permacol, and 1.9 to 5.8 N/mm for Peri-Guard. Similarly, the native porcine abdominal wall sustained a maximum load of approximately 10 to 17 N, tensile stress of approximately 3.4 to 5.6 N/cm, and stiffness of approximately 0.8 to 1.1 N/mm. No significant differences were observed

between the mesh-repaired sites and the native porcine abdominal wall for any of the meshes at any of the time points ($p > 0.05$). Greater preimplantation strength/stiffness did not correlate with augmented strength/stiffness at the repair site for any of the meshes used at any of the time points ($p > 0.05$).

Histologic evaluation revealed differences between materials with respect to crosslinking and tissue type (ie, dermis versus pericardium). Six histologic features of remodeling, including cellular infiltration, cell types present, ECM deposition, scaffold degradation, fibrous encapsulation, and neovascularization were scored individually for each specimen. These 6 subcategory scores were then averaged to produce an overall composite score. This composite score for both non-crosslinked meshes (Veritas and AlloDerm) revealed more favorable remodeling characteristics at 1 month (Fig. 2) compared with both crosslinked meshes (Permacol and Peri-Guard) ($p < 0.04$). In addition, Veritas scored higher than AlloDerm ($p < 0.001$) at 1 month. At 6 months postimplantation, both non-crosslinked meshes again demonstrated greater overall remodeling compared with both crosslinked meshes ($p < 0.001$). None of the biologic meshes examined remained static in situ, however. Over time, all 4 meshes showed substantial improvement in composite scores compared with their respective 1-month scores ($p < 0.04$). At all time points, Veritas demonstrated the highest degree of remodeling even when compared with the other non-crosslinked mesh, AlloDerm.

Additional examination of histologic features by subcategory revealed that non-crosslinked meshes initially permitted more cellular infiltration than the crosslinked meshes ($p < 0.008$). By 6 and 12 months this trend disappeared and all meshes achieved equivalent cellular infiltration (Fig. 3A). Additionally, both non-crosslinked meshes achieved peak levels of cellular infiltration within the first month, and subsequent levels at 6 and 12 months showed no additional increase. However, cellular infiltration into both crosslinked meshes increased significantly over time ($p < 0.001$), with cells typically reaching the center of the crosslinked scaffolds by 12 months.

For cell types, fewer inflammatory cells and more fibroblasts were detected at 1 and 6 months in Peri-Guard compared with AlloDerm ($p < 0.02$), and all other comparisons at these time points were not significant. Over time, more fibroblasts and fewer inflammatory cells were observed for all 4 mesh types ($p < 0.04$). However, by 12 months, Veritas exhibited the greatest content of fibroblasts and least amount of inflammatory infiltrate compared with the other 3 materials ($p < 0.04$) (Fig. 3B).

Higher levels of ECM deposition were observed at 1 month in non-crosslinked versus crosslinked materials ($p < 0.02$), with Veritas exhibiting the highest degree of ECM deposition, even compared with AlloDerm ($p < 0.03$). At 6 months, Veritas, AlloDerm, and Permacol all demonstrated greater ECM deposition than Peri-Guard ($p < 0.02$), but by 12 months, the only remaining difference was that Veritas exhibited the highest level of ECM deposition compared with Peri-Guard and AlloDerm ($p < 0.03$). It is important to note that, except for Veritas, which reached a maximum by 1 month's time, deposition of ECM increased steadily between 1 and 6 months for all other meshes, regardless of source tissue type or crosslinking (Fig. 3C).

Analysis of scaffold degradation scores showed that non-crosslinked meshes were more markedly degraded at 1 and 6 months compared with their crosslinked counterparts ($p < 0.03$). Veritas also exhibited significantly more scaffold degradation than the other non-crosslinked mesh, AlloDerm, at both 1 and 6 months ($p = 0.010$ and $p = 0.002$, respectively). At 12 months, Veritas remained significantly more degraded than the others ($p < 0.001$). At all time points, Veritas demonstrated significantly greater scaffold degradation compared

with the other non-crosslinked mesh (AlloDerm), as well as to both crosslinked meshes ($p < 0.05$, Fig. 3D). Within each mesh type, only Permacol exhibited significant degradation over time, with greater degradation at 12 months than the degradation observed at both 1 and 6 months ($p < 0.04$).

Examination of fibrous encapsulation revealed that Veritas was significantly less encapsulated (higher score means less encapsulation) than all other meshes at 1 month ($p < 0.01$). By 6 months, AlloDerm and Veritas scored similarly, and both were significantly less encapsulated than the crosslinked meshes ($p < 0.001$). However, at 12 months, crosslinked meshes showed decreasing levels of encapsulation, suggesting that this process might be reversible. By 12 months, only Peri-Guard was significantly more encapsulated than Veritas ($p = 0.016$, Fig. 3E).

Lastly, significantly greater neovascularization was observed in both non-crosslinked meshes at 1 and 6 months compared with both crosslinked meshes ($p < 0.05$). By 12 months, however, Veritas and AlloDerm reached significance in comparison with Peri-Guard, but not Permacol ($p < 0.01$). Permacol exhibited a slight increase in neovascularization between 6 and 12 months, such that by 12 months it achieved levels similar to the non-crosslinked meshes ($p > 0.05$) (Fig. 3F). All 4 meshes exhibited increased neovascularization over time, with greater neovascularization observed at both 6 and 12 months compared with 1 month ($p < 0.02$). Representative photographs of each mesh at each time point are depicted in Figure 4.

DISCUSSION

Although there are currently at least 14 different types of biologic mesh materials on the market, the hernia repair literature lacks long-term studies directly comparing these materials. This study represents the first of its kind to directly compare the biomechanical characteristics and histologic features of 4 commonly used biologic meshes in a long-term animal model of ventral hernia repair. It should be noted that although biologic mesh materials are typically used in contaminated settings, this study was limited to clean settings. The performance of these materials in the presence of bacteria was not examined.

One could hypothesize that the strength of the abdominal wall is augmented by the presence of a biologic mesh and that a biologic mesh with greater de novo tensile strength would augment the strength of the repair compared with a mesh with lower de novo strength. Surprisingly, our biomechanical testing results showed that none of the repair sites achieved strengths substantially greater than the native porcine abdominal wall, indicating that the use of biologic mesh did not augment the native strength of the abdominal wall. However, the strength of a hernia defect without mesh placement was not evaluated. It is possible that biologic mesh augments the strength of a healed defect site, but this was not evaluated. In addition, even though the 4 biologic meshes exhibited a wide range of de novo tensile strengths, all repair sites demonstrated similar strengths at 1, 6, and 12 months postrepair regardless of the type of mesh used to perform the repair. Overall, there does not appear to be a relationship between the de novo strength of the biologic mesh chosen to repair the defect and the strength of the repair for the short- or long-term.

There were also no differences in the strengths of the repair sites over time. This might indicate that new tissue is deposited at the repair site as the scaffold is degraded, preventing the site from weakening over time.

In addition to the de novo tensile strengths of the biologic mesh chosen for a ventral hernia repair, the de novo stiffness could also potentially influence the repair. Stiffness can be loosely described as the load that a material withstands as it is deformed (ie, stretched). For

hernia repair applications, materials must resist deformation to prevent bulging and hernia recurrence. However, these materials must not possess supraphysiological stiffness that could potentially hinder movement and lead to discomfort during respiration, bending, etc. Our biomechanical testing results showed that the stiffness of the repair sites was not substantially greater than that measured for the native porcine abdominal wall. Even though the 4 biologic meshes exhibited a wide range of de novo stiffnesses, all of the repair sites demonstrated similar stiffness at 1, 6, and 12 months postrepair, regardless of the type of mesh used to perform the repair. There were also no differences in the stiffness of the repair sites over time. Overall, there does not appear to be a relationship between the de novo stiffness of the biologic mesh chosen to repair the defect and the stiffness of the repair for the short- or long-term, even in the case of a very stiff material such as Permacol. Similarly, we did not observe any visible bulging or evidence of reherniation for materials such as AlloDerm and Veritas, which exhibited the lowest de novo stiffnesses.

It should be noted, however, that biologic meshes are collagenous materials, the stiffness of which depends on the strain at which those values are measured. Strain is the change in length (ie, the amount that the material is stretched) divided by the original length. At low strain, collagenous materials exhibit different stiffness values than at high strain. The high strain region of the curve is generally near the peak strength and can be interpreted as representing more intense physical activities that stretch the abdominal wall to a greater extent. In this study, de novo AlloDerm was shown to have greater stiffness than Veritas ($p = 0.0353$). However, the stiffness measurements obtained in this study were recorded in the high strain region of the curve. Many less-intense activities would likely fall in the low strain region of the curve, and it is likely that materials with greater intrinsic elasticity (ie, dermis) would exhibit less stiffness in this region compared with a material derived from pericardium.

With regard to overall remodeling characteristics, our histologic analyses revealed that both non-crosslinked materials exhibited more favorable remodeling characteristics, as evidenced by higher composite scores, compared with either of the crosslinked materials at all time points evaluated. Of the non-crosslinked materials, Veritas scored considerably higher than AlloDerm in terms of overall remodeling, as well as many of the individual subcategory scores. This composite score was designed to encompass 6 important histologic features of remodeling that determine how well the material performs in vivo. It is important to associate remodeling with all 6 of these characteristics rather than 1 in isolation. For example, there must be a balance between degradation of the scaffold, infiltration of cells, and deposition of new tissue and blood vessels. When the scaffold can no longer be detected visually, it is considered fully degraded. Without evidence of the other features of remodeling, scaffold degradation alone does not constitute true remodeling. The balance between ECM deposition and scaffold degradation is a critical aspect of remodeling. It is still unknown what effect the ratio of matrix deposition to implant degradation has on the longevity of the hernia repair.

Histologic analyses also revealed that Veritas tended to reach the highest scores by 1 month, with little change during the course of 6 or 12 months, except for cell types and neovascularization scores, which both increased over time. AlloDerm tended to follow the same trends as Veritas, with similar high scores early on and no substantial increases at later time points. Differences between these non-crosslinked materials were apparent in the ECM deposition and scaffold degradation scores with Veritas scoring higher than AlloDerm. These differences suggest that characteristics of these materials, such as tissue type (ie, pericardium versus dermis) or processing/sterilization conditions, ultimately influence the extent of ECM deposition and scaffold degradation in both the short- and long-term in a porcine model of ventral hernia repair. However, other features such as cellular infiltration,

cell types present at the repair site, and extent of neovascularization do not appear to be affected by these differences.

Crosslinked materials (Peri-Guard and Permacol) tended to score lower than the non-crosslinked materials for many of the histologic subcategory scores, particularly at 1 month postrepair. However, by later time points, many scores were improved to levels similar to the non-crosslinked materials. For example, Peri-Guard and Permacol both demonstrated increased cellular infiltration between 1 and 6 months, with scores equivalent to the non-crosslinked materials at 6 months, and maintenance of these scores at 12 months. These trends suggest that cross-linking can hinder cellular infiltration in the short-term (ie, 1 month postrepair). However, because all 4 meshes showed equivalent cellular infiltration at later time points, it is possible that crosslinking might not influence cellular infiltration substantially in the long-term.

In general, the crosslinked materials also demonstrated increased scores by 12 months for cell types, ECM deposition, scaffold degradation, fibrous encapsulation, and neovascularization compared with their scores at 1 month. In many cases, the 12-month scores for the crosslinked materials were similar to those for AlloDerm at 12 months. However, Veritas tended to continue to score considerably higher than all other materials at 12 months. Again, these differences suggest that factors beyond crosslinking, such as tissue type or processing conditions, affect long-term outcomes in this animal model.

For scaffold degradation, it is important to note that greater scaffold degradation was observed at both 1 and 6 months for the non-crosslinked materials, compared with the crosslinked materials ($p < 0.05$). These results support the idea that crosslinking alters the structure of the collagen and prevents rapid degradation in the short-term. However, Veritas also exhibited substantially greater scaffold degradation compared with AlloDerm, which indicates that some characteristic other than crosslinking also influences the rate of scaffold degradation. It should also be noted that Veritas, AlloDerm, and Peri-Guard all reached their maximum scaffold degradation at 1 month post-repair. Only Permacol demonstrated increasing scaffold degradation during the course of the study, indicating that different crosslinking methods can also influence the rate of scaffold degradation.

Similarly, less fibrous encapsulation was observed for the non-crosslinked materials at 1 and 6 months post-repair compared with both of the crosslinked materials ($p < 0.05$ in all cases), indicating that crosslinking can lead to more foreign-body response in the short-term. Of all 4 materials, Veritas exhibited the least fibrous encapsulation and did not show any differences in the long-term. All 3 of the other materials exhibited some fibrous encapsulation early on, with a significant reduction in encapsulation over time ($p < 0.02$). By 12 months, only Peri-Guard exhibited significantly more fibrous encapsulation than Veritas ($p = 0.016$). AlloDerm and Permacol were similar to Veritas by 12 months. These results indicate that fibrous encapsulation can be a reversible process. Although crosslinked materials caused greater fibrous encapsulation in the first 6 months, Permacol materials exhibited a substantial decrease in encapsulation between 6 and 12 months, making it similar to the non-crosslinked materials by 12 months. However, Peri-Guard materials maintained comparable levels of fibrous encapsulation throughout 1, 6, and 12 months, indicating that some characteristic other than crosslinking leads to a greater foreign-body response to these materials.

Overall, the data suggest that although crosslinking differentiates biologic meshes in the short-term with regard to many histologic features such as cellular infiltration and neovascularization, many of the histologic features are not impacted substantially in the long-term by crosslinking alone. It is possible that other variables, such as source/tissue

type, and processing conditions, such as decellularization and sterilization procedures, affect biocompatibility considerably more in the long-term compared with the short-term effects of crosslinking. Although crosslinking affected scaffold degradation scores substantially, the strength and stiffness of the repair sites did not appear to be impacted by crosslinking alone.

CONCLUSIONS

Although many differences were identified between the 4 biologic meshes examined in this study, it is difficult to speculate whether any of these biologic meshes possess superior biocompatibility for ventral hernia repair because there are such wide variations in both the clinical scenarios encountered and the biological responses of individual patients. Future research should concentrate on differences between materials based on the type/source of tissue and processing conditions beyond crosslinking, including the decellularization and sterilization processes.

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Figure 1.

Biomechanical characteristics of mesh-repaired sites over time compared with de novo strength and native porcine abdominal wall. (A) Maximum load (Newton [N]), (B) tensile strength (N/cm), (C) stiffness (N/mm). All 4 meshes were significantly stronger and stiffer at time 0 compared with their corresponding repair sites (mesh-abdominal wall tissue composites) after 1, 6, or 12 months ($p < 0.01$ for all comparisons). Although significant differences were observed between the strengths and stiffnesses of the 4 meshes at time 0, no significant differences were detected between mesh-repaired sites at 1, 6, or 12 months due to the type of mesh used to repair the defect ($p > 0.05$ in all cases). In addition, no significant differences in strength or stiffness of the repair site were detected over time for any of the meshes used ($p > 0.05$ in all cases). Abd, abdominal.

Figure 2.

Composite scores for both non-crosslinked meshes (Veritas and AlloDerm) were significantly higher than the composite scores for both crosslinked meshes (Permacol and Peri-Guard), $p < 0.05$ and Veritas scored higher than AlloDerm; $p < 0.05$.

Figure 3.

Histologic scores, separated by mesh type and length of time in vivo. (A) Cellular infiltration scores demonstrated that the non-crosslinked meshes (Veritas and AlloDerm) initially permitted more cellular infiltration than the crosslinked meshes (Permacol and PeriGuard); $p < 0.008$. By 6 and 12 months, this trend disappeared and all meshes achieved equivalent cellular infiltration ($p > 0.05$). (B) Cell types scores revealed that fewer inflammatory cells and more fibroblasts were detected at 1 and 6 months in PeriGuard compared with AlloDerm ($p < 0.02$), and all other comparisons at these time points were not significant ($p > 0.05$). By 12 months, Veritas exhibited the greatest number of fibroblasts and least inflammatory infiltrate compared with the other 3 materials ($p < 0.04$). (C) Extracellular matrix (ECM) deposition scores at 1 month demonstrated that the non-crosslinked meshes (Veritas and AlloDerm) initially permitted more cellular infiltration than the crosslinked meshes (Permacol and PeriGuard); $p < 0.02$. At 6 months, Veritas, AlloDerm, and Permacol all demonstrated greater ECM deposition than PeriGuard ($p < 0.02$), and by 12 months, the only remaining difference was that Veritas exhibited the highest level of ECM deposition compared with PeriGuard and AlloDerm ($p < 0.03$). (D) Scaffold degradation scores showed that non-crosslinked meshes (Veritas and AlloDerm) were markedly more degraded at 1 and 6 months compared with the crosslinked meshes (Permacol and PeriGuard); $p < 0.03$. Veritas also exhibited significantly more scaffold degradation than the other non-crosslinked mesh, AlloDerm, at both 1 and 6 months ($p = 0.010$ and $p = 0.002$, respectively). (E) Fibrous encapsulation scores revealed that Veritas was significantly less encapsulated (higher score means less encapsulation) than all other

meshes at 1 month ($p < 0.01$). By 6 months, both non-crosslinked materials (AlloDerm and Veritas) scored similarly, and both were significantly less encapsulated than the crosslinked meshes ($p < 0.001$). However, at 12 months, the crosslinked meshes showed decreasing levels of encapsulation, suggesting that this process might be reversible. (F) Neovascularization scores were significantly higher for both non-crosslinked meshes (Veritas and AlloDerm) at 1 and 6 months compared with both crosslinked meshes (Permacol and Peri-Guard); $p < 0.05$. By 12 months, however, Veritas and AlloDerm reached significance only in comparison with Peri-Guard, but not Permacol ($p < 0.01$).

Figure 4. Photographs of hematoxylin and eosin–stained specimens of each mesh-repaired site after 1, 6, and 12 months in vivo (100× magnification).