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Platelet rich plasma enhances tissue incorporation of biologic mesh

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ABSTRACT

Background: High recurrence rates because of poor tissue incorporation limit the use of acellular dermal matrices (ADMs) in ventral hernia repair (VHR). Platelet rich plasma (PRP) is a growth factor–rich autologous blood product known to enhance tissue repair through cellular proliferation and neovascularization. We sought to study the effect of PRP on a porcine noncross-linked ADM in an *in vivo* model of VHR. We hypothesized that PRP would enhance ADM-tissue incorporation in a rat model of VHR.

Methods: Whole blood was extracted from Lewis rats followed by PRP isolation and characterization. Using a rat model of VHR, a noncross-linked ADM (Strattice) was implanted and activated PRP applied before closure. Rats were sacrificed at 2, 4, and 6 wk. Immunohistochemical staining of CD 31 on endothelial cells was used to quantify neovascularization. Hematoxylin eosin stained tissues were measured to quantify tissue deposition.

Results: Platelet concentration of PRP was standardized to 1×10^6 platelets/μL. Grossly, vessels were more evident in PRP-treated rats. Immunohistochemical analysis demonstrated neovascularization was significantly greater in the PRP-treated ADMs at all time points. This increase in neovascularization correlated with an increased thickness of tissue deposition at 4 and 6 wk.

Conclusions: PRP enhanced neovascularization and incorporation in a rat model of VHR. Enhanced neovascularization was associated with earlier and greater tissue deposition on the ADM. This suggests that PRP could be used as an adjunct to VHR in clinical scenarios where poor wound healing is anticipated and enhanced neovascularization and early tissue deposition are desired.

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1. Introduction

More than 250,000 ventral hernia repairs (VHRs) are performed in the United States annually, making incisional hernias one of our most costly complications [1,2]. Recently, the Ventral Hernia Working Group published guidelines outlining the expansion of indications for use of bioprosthetic mesh over more traditional material in VHR [3]. Traditional repair materials comprise degradable and nondegradable synthetic polymers that are associated with several complications such as infection, erosion, adhesion formation, and contraction [4–6]. This has lead to interest in developing more biological approaches to abdominal wall reconstruction. Acellular matrices can be used as biomimetic scaffolds possessing the appropriate architecture for cellular ingrowth and proliferation. Furthermore, biologic materials have greater biocompatibility and have been associated with fewer complications [7]. Bioprosthetic options include acellular extracellular matrices (ECMs) derived from human or animal sources such as dermis, intestinal submucosa, and pericardium [3,8]. These prosthetics have proved useful in the repair of abdominal incisional hernias but are themselves hindered by poor long-term mechanical strength and high cost [9]. After implantation, remodeling of the ECM by the body's natural healing processes can weaken the mechanical strength of the mesh, leading to poor integration and hernia recurrence or bulge when used to bridge unopposed fascia [10]. The recurrence necessitates further reconstruction, limiting the use of biologic meshes to act as a bridge to permanent synthetic repair [11,12]. Furthermore, if neovascularization of the biologic matrix is delayed, infection, encapsulation, and resorption are inevitable.

Platelet rich plasma (PRP), an autologous blood product rich in growth factors, is known to improve cellular migration, proliferation, ECM deposition, and neovascularization. PRP has been shown to contribute to enhanced neovascularization in tissue with poor perfusion [13]. The proregenerative effects of PRP may provide the necessary biologic instructions to speed the wound healing process in biologic mesh hernia repair, allowing neovascularization in a more rapid fashion.

We hypothesized that PRP could aid in successful biologic mesh incorporation through enhanced neovascularization and tissue deposition. We propose to investigate the effects of PRP on the noncross-linked porcine acellular dermal matrix (ADM), Strattice (LifeCell, Bridgewater, NJ). If PRP indeed speeds the rate of neovascularization of the biologic matrix, implications regarding mitigating infectious risk and speeding collagen cross-linking in VHR will be significant. This information could build a foundation of knowledge for further investigation regarding the use of both PRP in VHR with biologic materials.

2. Materials and methods

2.1. Animals

Forty-two male Lewis rats weighing approximately 300–315 g underwent VHR with noncross-linked porcine ADM (Strattice; LifeCell). Because of their inbred nature and similar genetic composition, Lewis rats were chosen. Animals were randomly assigned to receive 200-μL PRP or 200-μL saline as placebo at the time of closure. All animals were purchased from Charles River (Wilmington, MA) and housed at Houston Methodist Hospital Research Institute animal facility. The study was approved by the Institutional Animal Care and Use Committee at Houston Methodist Hospital Research Institute, and all investigators complied with the National Research Council's *Guide for the Care and Use of Laboratory Animals*.

2.2. PRP isolation and characterization

To attain sufficient amounts of blood, 10 Lewis rats were used for the isolation of PRP. Their highly inbred nature allows for pooling of donor blood. Buprenorphine (0.03 mg/kg) and carprofen (5 mg/kg) were administered for preoperative analgesia. Deep anesthesia was induced and maintained using a 2.5%–3.0% Isoflurane and/or oxygen mixture through a nonrebreather mask. Using aseptic techniques, a sternotomy was made and the heart exposed. Using a 22-gauge needle, a terminal cardiac blood draw was performed. The blood was then mixed with 10% acid citrate dextrose to prevent clotting and centrifuged at 200*g* for 15 min to remove the red cell fraction. The plasma was centrifuged at 1600*g* for 10 min to pellet the platelets. The plasma is then removed leaving 1-mL total volume. To standardize the concentration of PRP, the number of platelets was quantified using a Multisizer Coulter Counter (Beckman Coulter, Pasadena, CA) to have a final concentration of 1×10^6 platelets/μL of plasma. To confirm the final concentration, a complete blood cell count was performed on all final specimens. The PRP was then stored at −80°C for further use.

2.3. Abdominal wall defect creation

Rats were randomly allocated into treatment or placebo groups. Buprenorphine (0.03 mg/kg) and carprofen (5 mg/kg) were administered for preoperative analgesia. Anesthesia was induced and maintained using a 2.5%–3.0% isoflurane and/or oxygen mixture through a nonrebreather mask. Aseptic techniques were used for the duration of the surgery. To create the abdominal wall defect, all rats had a 3-cm midline skin incision made and a 3-cm skin flap raised using sharp dissection (Fig. 1). A full thickness abdominal midline fascia and/or muscle and/or peritoneum defect measuring 2 cm in length was then made. The skin was then approximated and stapled leaving the defect unrepaired. Fascial defects were allowed to mature over 28 d. The animals were monitored by physical examination and weight daily for 10 d and then weekly after that. Exclusion collars were applied for 5 d to ensure the animals did not disrupt the incision in the early postoperative period. After 14 d, the staples were removed.

2.4. Abdominal wall defect repair

At day 28, the animals underwent hernia repair. The protruding viscera were freed with sharp dissection, and the fascial edges defined. A 2.5 × 1.5 cm piece of Strattice was customized to the shape of the fascial defect to allow for 5 mm