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RESEARCH ARTICLE

A collagen/PLA hybrid scaffold supports tendon-derived cell growth for tendon repair and regeneration

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Abstract

A rotator cuff tendon tear is a common shoulder injury with a relatively high rate of recurrence after surgical repair. In order to reinforce the repair and reduce the risk of clinical complications, a patch scaffold is typically sutured over the tendon tear to provide post-surgical mechanical support. However, despite considerable research effort in this area, a patch scaffold that provides both superior initial mechanical properties and supports cell proliferation at the same time has not yet been achieved. In this study, we engineered a collagen/poly(lactic acid) (COL/PLA) hybrid yarn to leverage mechanical strength of PLA yarn and the bioactivity of collagen. The COL/PLA yarns were used to fabricate a tissue engineering scaffold using textile weaving technology. This hybrid scaffold had a tensile strength of 354.0 ± 36.0 N under dry conditions and 267.2 ± 15.9 N under wet conditions, which was satisfactory to maintain normal tendon function. By introducing COL yarns into the hybrid scaffold, the proliferation of tendon-derived cells was significantly improved on the scaffold. Cell coverage after 28-days of *in vitro* cell culture was noticeably higher on the COL yarns compared to the PLA yarns as a result of a larger number of cells and more spread cell morphology on collagen. Cells spread in multiple directions on COL yarns, which resembled a more natural cell attachment on extracellular matrix. On the contrary, the cells attached to the PLA filaments presented an elongated morphology along the fiber's axial direction. Combining the mechanical robustness of PLA and the biological activity of collagen, the woven COL/PLA hybrid scaffold has shown its potential to be a promising candidate for tendon repair applications.

KEYWORDS
biomaterials, biotextiles, collagen yarn, tendon regeneration, tissue engineering scaffold

1 | INTRODUCTION

The rotator cuff tendon is one of the most frequently injured tendons due to trauma or long-term overuse of the shoulder. About 27% and 37% of the general population are affected by full and partial rotator cuff tears, respectively.[1] In the United States, there are estimated 75,000 rotator cuff surgical procedures performed every year, and the number is increasing given the aging population and the demand for improved independence.[2] The management of large and massive rotator cuff tears is always a clinical challenge for the surgeon because of

tendon-retraction.[3] Tear recurrence rates after surgical repair can be as high as 34%–94%, depending on the patient's age, tear size and chronicity, muscle atrophy and degeneration, tendon quality, repair technique, and the postoperative rehabilitation protocol.[4–6] For tendon tissue of poor quality, graft augmentation is an effective strategy to reinforce the repair site and provide biomechanical support that can shield the repaired rotator cuff tendon from extreme external applied stresses.[7]

An ideal reinforcement graft for rotator cuff repair should have mechanical properties similar to native tendon tissue. At the same time, it should also promote rapid host cell in-growth and tissue healing. The extracellular matrix (ECM)-based scaffolds can promote tenden tissue healing to improve the strength of the repaired tendon and reduce the clinical rate of recurrence.[3,8–10] But opposite reports indicate that ECM-based grafts can cause inadequate initial mechanical support, loss of structural integrity, severe host immune response and high post-operative rates of tear recurrence.[11–16] On the other hand, synthetic grafts made from permanent or degradable polymers show promising clinical outcomes in terms of a stronger mechanical performance and a more durable and consistent tendon function in the long term.[5,17–19] However, synthetic grafts are associated with an adverse foreign-body reaction and poor biological healing in several animal and clinical studies leading to increased concern and risk of recurrence.[20–22] So neither of these two categories of augmentation devices possesses both sufficient mechanical properties to maintain dimensional stability and mechanical integrity, and superior biological performance for rapid healing and regeneration.

A hybrid tendon scaffold that combines biological and synthetic components is proposed to be a promising alternative candidate to provide sufficient mechanical support and promote host tissue healing at the same time. Collagen in its fibrillar form is the major component of tendon ECM. It is highly organized and aligned along the axis of the tendon, providing the load-bearing structure and accounts for approximately 60%–85% of the total dry mass of the tendon's ECM. Type I and Type III collagen account for about 90%–10% of the entire collagen content, respectively.[23] Considering its critical role in the native tendon tissue, collagen is an ideal biological material for fabricating tendon scaffolds. However, collagen as a coating material shows conflicting cellular responses, which may be due to the insufficient and inconsistent binding of collagen and the loss of collagen alignment and natural collagen configuration in the coating solution.[24–26] In this study, we used an electrochemically aligned collagen monofilament (COL), which is a continuous collagen yarn with axial collagen fibril alignment, improved mechanical properties, and an extended in vitro degradation time.[27,28] Being aligned by an electrical current, these COL yarns achieve a more densely packed collagen molecular structure and a more natural collagen fiber morphology.[29] Unlike traditional collagen coating that degrades fast and lacks bioactivity, the presence of the collagen yarn is prolonged due to slower degradation.[24,30] The mechanical properties of collagen are also improved in the COL yarns compared to traditional collagen fabrication methods. We have reported small-diameter vascular grafts fabricated from COL yarns, which show equivalent strength and compliance to the saphenous vein graft and promoted endothelial cell attachment and proliferation compared to a pure poly-L-lactic acid (PLA) graft.[31,32] The fibrous alignment of the collagen yarn is also able to guide the tenogenic differentiation, which cannot be achieved by a traditional collagen coating, film and sponge.[33] In a pilot study in a rabbit model, the manually woven COL scaffold has a positive tenogenic response with the presence of the tendon-specific marker tenomodulin, procollagen I, collagen I and collagen III.[33] However, the mechanical strength of a collagen yarn is insufficient to withstand the tension and friction during the high-speed woven manufacturing process, which hinders scale-up and commercial production of the collagen yarn-based scaffold. As a result, a combination of COL with PLA yarns is proposed in this study so as to provide adequate mechanical properties for high-speed automatic production weaving technology. PLA has been broadly used in medical devices approved by the United States Food and Drug Administration and shown to be robust and consistent in its mechanical properties.[34,35] By combining COL with PLA yarns, we anticipated achieving the benefits of both the tenogenic potential of collagen and the mechanical properties of the PLA yarn for high-speed manufacturing.

Woven structures are widely used in commercial medical devices and studied extensively in tissue engineering applications to produce scaffolds for the repair of tendons,[5,36,37] bone,[38–40] cartilage,[41–43] heart valves,[44,45] and peripheral vascular tissue.[46–48] They provide high mechanical strength and structural stability, and the properties such as thickness, porosity and strength can be easily adjusted and modified by altering the woven design pattern and the density of warp and weft yarns.[44] Its superior mechanical performance and flexibility to select a variety of different materials makes a woven patch an attractive candidate for tendon repair. In a previous study, a woven PLA scaffold show a tensile stress–strain curve similar to a human or canine infraspinatus rotator cuff tendon, which is significantly better than ECM-based scaffolds with regard to mechanical properties.[49]

Overall, in this study, we propose the combination of hybrid COL/PLA yarns and a weaving technique to fabricate a tendon repair patch scaffold with both robust mechanical properties and superior biological performance. The objective of this study is to develop an easy-to-scale-up patch scaffold that can promote rapid tendon cell proliferation for rotator cuff tendon repair. It is the first time that the continuous collagen yarn spun from extracted rat collagen is fabricated into a tissue engineering scaffold using a high-speed production weaving approach. The COL threads were combined with PLA multifilaments, with the former providing superior biological performance to improve tissue healing and ingrowth, and the latter supplying the desired mechanical properties that are necessary for high-speed manufacture of the scaffold using automated weaving technology. The collagen yarn and PLA filaments were plied together to form hybrid yarns and woven into a patch by an automated production loom. The mechanical properties and fatigue resistance of the hybrid woven patch were characterized and compared to a pure PLA woven control patch. In addition, the *in vitro* biological performance of these woven scaffolds was determined by measuring the proliferation and morphology of tendon-derived cells.

2 | MATERIALS AND METHODS

2.1 | Collagen yarns

The collagen yarn used in this study was fabricated from acid-extracted rat-tail collagen. The collagen extraction procedure and the method for spinning collagen yarns by electrochemical alignment have been published previously.[27,28] Briefly, Sprague Dawley rat tail tendons were obtained from College of Veterinary Medicine at North Carolina State University and were harvested at the time of the sacrifice of the rats which was performed in accordance with the Institutional Animal Care and Use Committee at the North Carolina State University. The tendon fibers were washed extensively in phosphate buffered saline (PBS) at pH 7.4 and then transferred to 0.15 M sodium chloride (NaCl) for a 5-minute washing followed by another 5-minute immersion in acetone to remove any fat, lipid and other impurities. The tendon fibers were then incubated in 0.5 M acetic acid and stirred at 4°C for 48 h and then precipitated in 10% w/v NaCl for 1 h. The precipitant was then re-dissolved with 0.25 M acetic acid for 24 h and then dialyzed against 17.5 mM acetic acid to obtain a final collagen solution with 3 mg ml^{-1}. The acidic collagen solution was further dialyzed with deionized (DI) water for 24 h before being introduced into the collagen electrochemical alignment device. A syringe and pump were used to introduce and maintain the flow of the solution between the anode and the cathode strips of a rotating electrode device.[27] A constant 40V electrical potential was applied to electrochemically compact the collagen molecules at the isoelectric point between the two electrodes. As a result, the collagen molecules aggregated into a thread and were drawn out and collected in a reservoir containing 80% isopropanol. The collagen thread was then dried in a fume hood at room temperature and stored in a fridge at 4°C prior to subsequent processing.

2.2 | Collagen/PLA hybrid yarns

In order to improve the yarn's mechanical and structural stability, two collagen threads were first plied and twisted together into a 2-ply yarn with approximately 150 turns-per-meter in an S twist direction, followed by chemically crosslinking with 1-ethyl-3-(−3-dimethylaminopropyl) carbodiimide hydrochloride (EDC) in 80% ethanol and N-hydroxysuccinimide (NHS) as the catalyst for 2 h. The molar ratio of collagen: EDC: NHS was selected to be 1:25:50. A hybrid yarn was then prepared by cabling the 2 ply crosslinked collagen yarn with a 146 denier 72 filament 2 ply PLA yarn (Xinxiang Sunshine Textile Company Ltd., Henan, China) using a twist level of 150 turns-per-meter (tpm) on a Model DirecTwist-2A (Agteks Ltd, Istanbul, Turkey) yarn plying machine. Similarly, 100% PLA warp yarns were prepared by cabling two 2 ply yarns with 150 tpm.

2.3 | Weaving

Plain woven prototype hybrid structures were fabricated on a "Muller" multi-position narrow width ribbon shuttle loom (Jakob Müller AG, Switzerland) from 28 100% PLA-cabled yarns in the warp direction and the hybrid collagen/PLA yarn in the weft direction. A 100% PLA control woven ribbon was also fabricated from pure PLA-cabled yarns in both warp and weft directions. In order to visualize the collagen protein yarns in the woven fabric, both the prototype hybrid and PLA control ribbons were dyed with Textile Identification Stain No. 3A for 10 min at 30–40 °C. After staining, the images of the two types of grafts were captured with a ZEISS Axiovert 100A inverted microscope (Carl Zeiss Microscopy LLC, NY, USA). Pore size was measured as the diagonal distance across the rectangular shaped pores at the warp and weft yarn interlacements in these woven fabrics.

2.4 | Mechanical properties of woven patches

To determine the mechanical properties of the woven patch grafts, we carried out axial tensile tests and suture retention tests on the scaffolds under dry and wet conditions. Both properties are essential for the security and performance of patch grafts for tendon repair applications. Native tendons are the major load-bearing tissue during movement and have excellent mechanical properties. In order to provide sufficient mechanical support at the wound site and prevent recurrence of the tissue tear, excellent tensile properties are necessary. On the other hand, at the anastomosis, suture security is also dependent on successful integration of the patch graft into the native tendon tissue.

The uniaxial tensile properties of the hybrid collagen/PLA woven ribbon (abbreviated as COL/PLA) and pure PLA woven ribbon control were tested. The patch samples were cut into 30 mm long specimens and tested on an Instron Model 5584 mechanical tester (Norwood, MA, USA) with a gauge length of 10 mm and a crosshead speed of 30 mm/min. The specimens were tested in both dry and wet conditions after being pretreated in 0.01 M PBS for 2 h. Each group of samples had five replicates under each condition. The maximum load, elongation at maximum load and linear stiffness were recorded. The physiologically relevant properties, such as load at 5 mm elongation and elongation at 50 N, were also reported, because 5 mm is the estimated maximum permissible retraction of a tendon after tendon repair and 50 N is the estimated physiologic load applied to an augmentation graft after tendon repair.[50]

The suture retention strength of both the COL/PLA hybrid patch and the PLA ribbon control were tested under dry conditions on the same Instron Model 5584 mechanical tester. One end of a 20 mm long rectangular patch sample was sutured with a size 2 FiberWire suture (Arthrex LLC, Naples, FL) at a distance of 5 mm from the edge of the patch. The non-sutured end of the sample was fixed by a flat clamp while the suture end was fixed by a capstan clamp. Five specimens of each type of patch were tested to failure in the warp direction at a rate of 200 mm min^{-1}.

2.5 | Biological properties of woven patches—Tendon-derived *in vitro* cell culture

To investigate the initial performance of the hybrid patch on promoting tendon cell growth and tendon healing, two different tendons,

namely the rotator cuff and Achilles tendon, were dissected under sterile conditions from adult one-month-old Sprague–Dawley rats. After rinsing with sterile 0.01 M PBS, the sheath and surrounding paratenon were removed and the tendons were minced into small pieces. The tendon pieces were then cultured in growth medium [50:50 DMEM:HAM F12, containing 10% fetal bovine serum, 1% penicillin–streptomycin solution, and 25 μg ml^{-1} ascorbic acid] under standard conditions of 37°C in a 95% air/5% CO_2 humidified atmosphere. The culture medium was changed every 3 days. When the cells in each petri-dish migrated from the tendon explants and reached confluence, they were trypsinized and subcultured in 75 ml flasks to allow proliferation under the same culture conditions. The third passage of the Achilles tendon-derived cells (ACs) and the rotator cuff tendon-derived cells (RCs) were used to evaluate the extent of cell growth on the COL/PLA patch and the 100% PLA control patch. The samples were cut into three 1×1 cm square specimens and placed in a 24-well plate, followed by ethylene oxide sterilization and pre-wetting in cell growth media. 1×10^4 cells were then seeded on each specimen and cultured in growth media under standard conditions of 37 °C in a 95% air/5% CO_2 humidified atmosphere for up to 28 days. The culture medium was changed every other day.

Cell metabolic activity for the ACs and RCs cultured on the COL/PLA and 100% PLA samples was determined qualitatively after 1, 4, 7 and 28 days. Briefly, the cell seeded patch samples were removed to new 24 well plates and the culture media were replaced with fresh media containing 10% alamarBlue® (AB) (Invitrogen) at each time point. After culturing for 4 h in the dark at 37°C, 100 μl supernatant from each specimen was transferred into a 96 well plate and the level of fluorescence was read using a Tecan Genios microplate reader (Tecan Trading AG, Switzerland).

To visualize the cell proliferation and growth, the cell-seeded COL/PLA and the 100% PLA patch samples were fixed in 10% formalin (Fisher Scientific, Loughborough, UK) for 10 min and then permeabilized by 0.5% Triton-X for10 min. Then, the cells were stained using 100 nM solution of Acti-stain 488 phalloidin (Cytoskeleton Inc., Denver, CO) and 4′,6-diamidino-2-phenylindole (DAPI) (Thermo Fisher Scientific, Waltham, MA) in 0.01 M PBS for 30 minutes in the dark at room temperature. The specimens were viewed and their images were captured at 100 X magnification using a Zeiss LSM880 laser confocal microscope (Carl Zeiss MicroImaging, NY).

Scanning electron microscopy (SEM) was also utilized to characterize the level of cell attachment to the patch graft scaffolds. The cell-seeded specimens were fixed in 10% formalin at room temperature for 30 min, followed by dehydration through a graded series of aqueous ethanol solutions, namely 30%, 50%, 70%, and 95% and finally 100% ethanol for 30 min each at 4 °C. Then, all the specimens were critical point dried for 15 min in a Samdri-795 critical point dryer (Tousimis Research Corporation, Rockville, MD) and sputter-coated with gold/palladium in a Hummer® 6.2 sputter coating system (Anatech Ltd, CA). The prepared specimens were examined in a JEOL JSM-5900LV scanning electron microscope (JEOL USA, Inc. Peabody, MA) at a 15 kV accelerating voltage and the SEM images were captured at magnifications of 100 X and 300 X.

2.6 | Statistics

Statistical analysis in this study was performed using JMP Pro 13 (JMP, Cary, NC) software. All the experimental results were reported as mean ± standard deviation (SD). Statistical differences in tensile properties under dry and wet conditions as well as cell metabolic activities among different time points were determined by two-way ANOVA, while the suture retention strength was tested by one-way ANOVA. Tukey's post hoc multiple comparison test was conducted between groups if the means were found to be significantly different by ANOVA. Significant differences were identified when $p < 0.05$.

3 | RESULTS

3.1 | Fabrication of the COL/PLA woven graft

As shown in Figure 1, the COL/PLA hybrid patch graft was woven by interlacing pure PLA yarn in the warp direction and COL/PLA hybrid yarn in the weft direction. The warp direction was used as the load bearing direction, and thus the pure PLA would be able to provide sufficient strength to the graft. In the weft direction, which is perpendicular to the warp direction, the COL/PLA hybrid yarn was used to provide cells with adhesion ligands and an ECM-like substrate to adhere to and proliferate. The pure PLA control patch was woven by using PLA yarns in both warp and weft directions.

The COL/PLA hybrid and pure PLA woven patch fabrics were fabricated in a continuous ribbon with a width approximately 17 mm as shown in Figure 1A. To visualize the collagen component, both the hybrid and the 100% PLA patch graft were stained with T.I.S Stain No. 3A (Figure 1B). The collagen yarns were dyed blue in color, while the PLA filaments were dyed a light shade of pink. When viewing the structure of the woven grafts at higher magnification under an optical microscope, the pores in the PLA control appeared to be more uniform than in the COL/PLA fabric (Figure 1C and 1D). Indeed, the size of the pores (measured under dry conditions) in the COL/PLA patch was significantly larger than those in the 100% PLA woven control ($p = 0.0073$). The thickness of the COL/PLA and the 100% PLA patch grafts were 528 ± 25 μm and 496 ± 28 μm respectively; there being no statistical difference between the two means (Figure 1E).

3.2 | The mechanical properties of the hybrid patch graft

Figure 2A shows the typical stress–strain curves of the COL/PLA patch graft and the pure PLA control sample. All of them had a relatively long toe region under both dry and wet conditions, which was likely due to the uncrimping of the woven structure under load. However, these two types of grafts had different tensile properties. The slope of the elastic region in the stress–strain curve of the PLA patch was steeper than that of the COL/PLA sample under both dry and wet conditions. Due to the incorporation of collagen, the COL/PLA

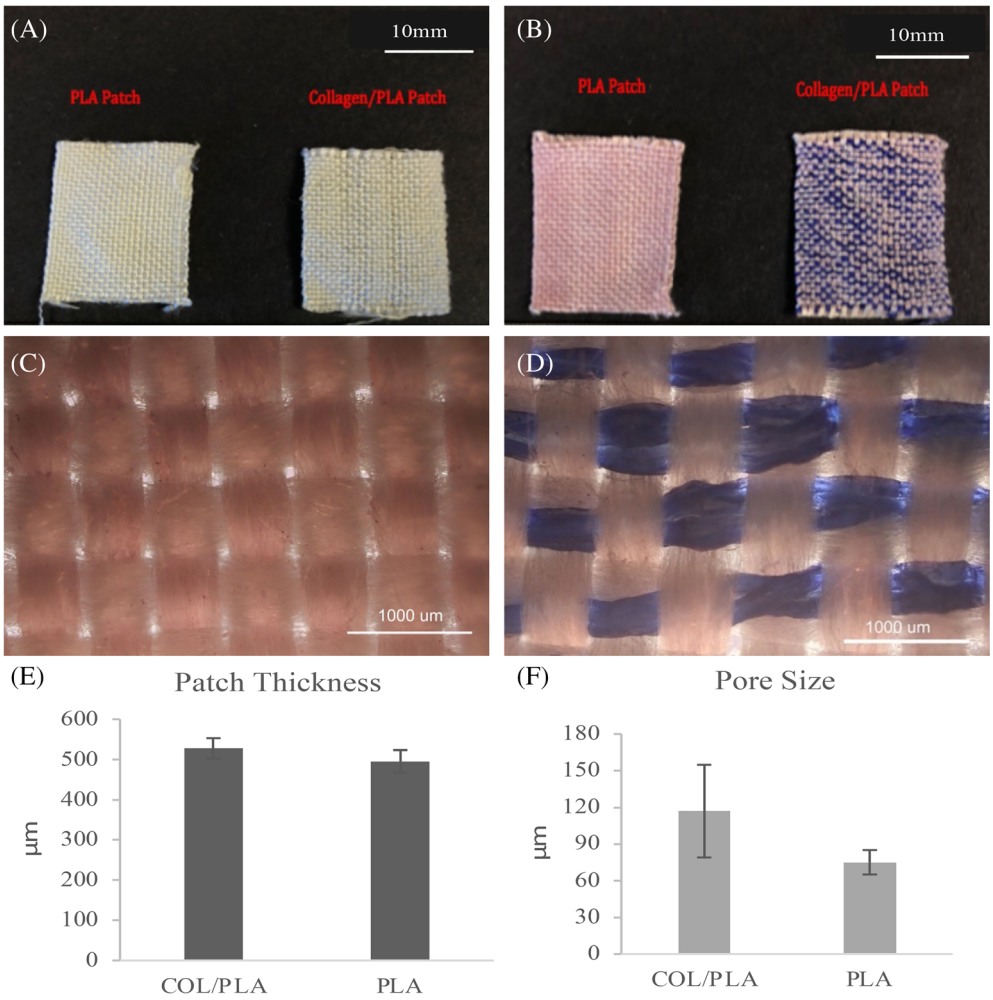

FIGURE 1 A gross view of the plain-woven COL/PLA and 100% PLA prototype patches (A) before staining and (B) after T.I.S. Stain No. 3A staining on collagen (blue). The 1/1 plain woven structures of (C) pure PLA control and (D) COL/PLA hybrid patch fabric viewed under an optical microscope at 40 X magnification. The warp direction is shown in the vertical direction. Results of patch thickness (E) and pore size (F) of COL/PLA and PLA prototype grafts.

blended patch graft had a significantly higher maximum load under dry conditions compared to the wet conditions, with values of 354 ± 36 N and 267 ± 16 N, respectively (Figure 2B). No significant difference was found in the maximum load between the COL/PLA and PLA patch grafts when tested under dry conditions.

As for the physiologically relevant properties, the PLA patch had a significantly higher load at 5 mm elongation under dry conditions compared to the COL/PLA patch ($p = 0.0341$) as shown in Figure 2C. Additionally, the difference between the PLA and the COL/PLA became more significant when the scaffolds were wet, with the PLA control sample supporting a 22.42 N higher load than the COL/PLA graft. However, the hybrid patch showed a comparable load performance at 5 mm elongation with no significant difference between dry and wet conditions.

For the elongation at maximum load, the only significant difference was found between the COL/PLA graft tested in the dry state compared to the wet state ($p = 0.0154$) (Figure 2D). The 100% PLA scaffold had a significantly lower elongation at 50 N compared to the COL/PLA scaffold both when dry ($p = 0.0188$), and when wet ($p = 0.0079$) (Figure 2E). For the linear stiffness (Figure 2F), the PLA scaffold had a significantly higher stiffness than the COL/PLA scaffold under both dry and wet conditions.

An important performance criterion of a patch graft from a clinical perspective is the suture retention strength, which is directly related to the security and stability of the repair. The results in this study showed that by adding the collagen component the suture retention strength was reduced (Figure 2G). The hybrid patch graft with collagen yarns had significantly lower suture retention strength than the pure PLA patch graft control ($p = 0.0005$).

3.3 | Effect of collagen on the *in vitro* cell-culture performance

The alamarBlue® fluorescence reading (AB reduction) was positively related to cell numbers on the scaffold. The rotator cuff tendon cells (RCs) seeded on the COL/PLA showed a significantly higher AB reduction compared to that on the PLA control grafts after *in vitro* culture for 7 days ($p < 0.0001$), 14 days ($p < 0.0001$), and 28 days ($p < 0.0001$), as presented in Figure 3A. This indicates that a more active proliferation of both the RCs as well as the Achilles tendon cells (ACs) occurred on the COL/PLA scaffold compared to the pure PLA patch graft over the 28 days of *in vitro* culture. The cell growth rate on the COL/PLA graft was much faster than that on the 100% PLA

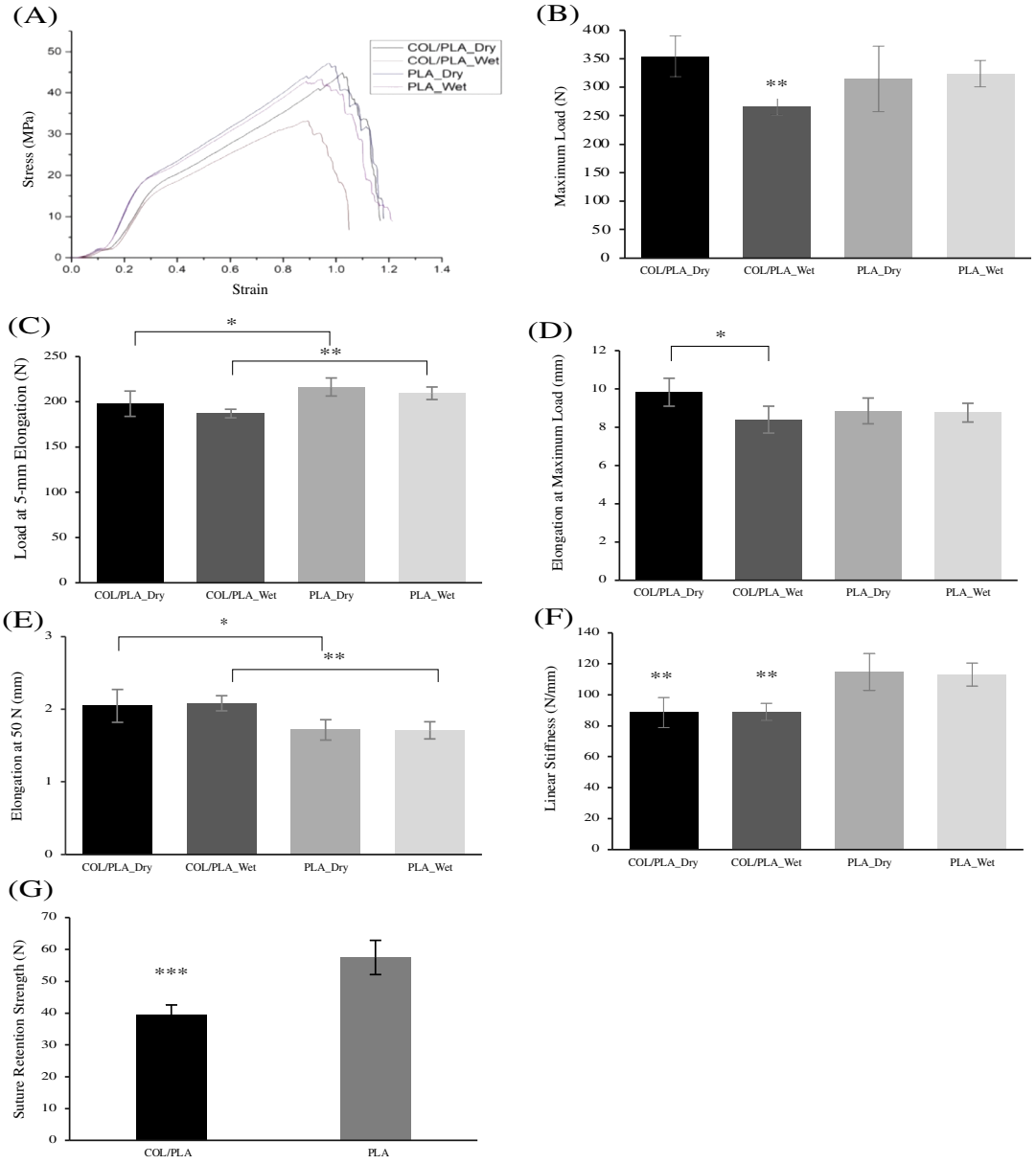

FIGURE 2 (A) Typical stress–strain curves for the COL/PLA and PLA patch grafts under dry and wet conditions. The axial tensile properties of the COL/PLA and PLA woven patch grafts under dry and wet conditions; (B) Maximum tensile load; (C) Load at 5 mm elongation; (D) Elongation at maximum load; (E) Elongation at 50 N, (F) Linear stiffness, (G) Suture retention strength. (*p < 0.05; **p < 0.01; ***p < 0.001)

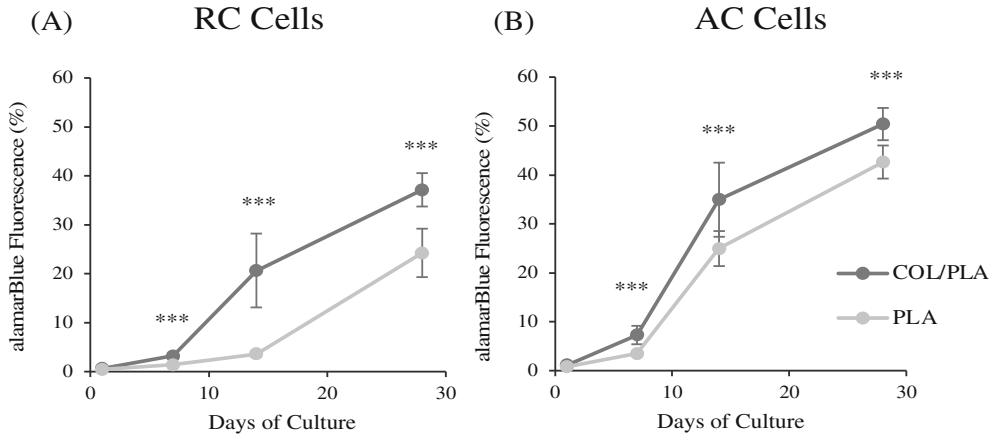

FIGURE 3 Percent reduction in alamarBlue® fluorescence for (A) rotator cuff tendon cells (RCs) and (B) Achilles tendon cells (ACs) during different periods of culture on COL/PLA and pure PLA patch grafts (***p < 0.001)

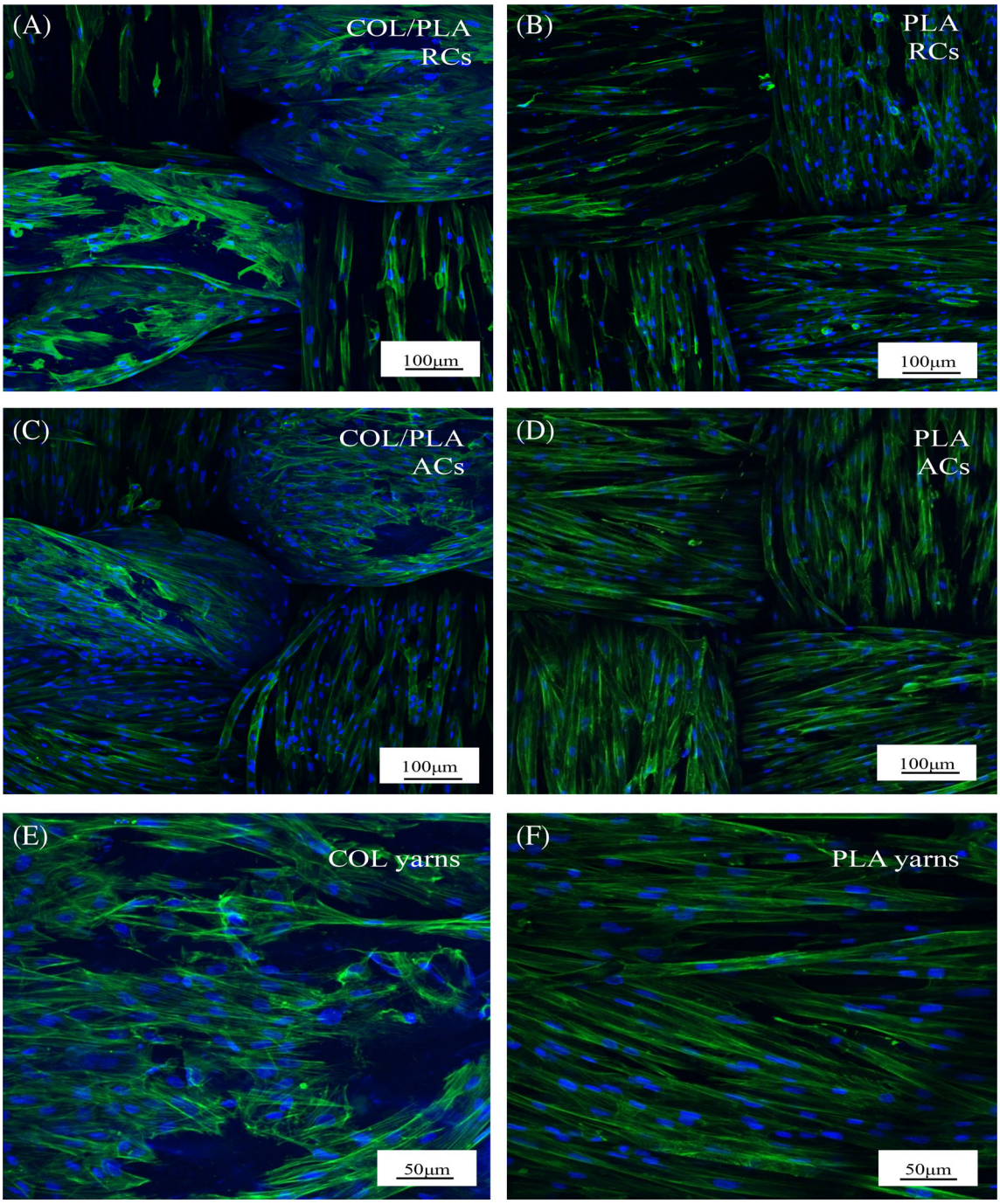

FIGURE 4 Confocal microscopic images of the COL/PLA patch graft after 28 days of culture with (A) rotator cuff tendon cells (RCs) and (C) Achilles tendon cells (ACs), and of the 100% PLA control graft after 28 days of culture with (B) RCs and (D) ACs. View of the ACs on COL yarns (E) and PLA multifilament yarns (F), at high magnification. Blue: DAPI stained cell nuclei, Green: Phalloidin stained Actin filaments.

graft from Day 7 to Day 14. Similarly, there was a significantly higher number of AC cells on the COL/PLA patch graft compared to the 100% PLA graft after 7 days ($p = 0.0002$), 14 days ($p = 0.0008$), and 28 days ($p < 0.0001$) of *in vitro* cell culture, as shown in Figure 3B.

Using a confocal microscope, we further confirmed the supportive role of collagen yarn towards AC and RC growth and level of activity. In Figure 4A-D, the collagen yarn was in the horizontal direction, which could be easily recognized because of its thicker dimension compared to the PLA multifilament yarn (Figure 4A and 4C). On Day 28, more RCs and ACs were attached to the collagen yarns in the horizontal direction than to the PLA filaments in the vertical direction, as observed in Figure 4A and 4C. The observation of such non-uniform cellular distribution indicates that both the RCs and ACs preferred to attach and proliferate on the collagen yarns. This difference of cell distribution in the vertical and horizontal directions was not observed on the pure PLA patch graft. As shown on the PLA graft

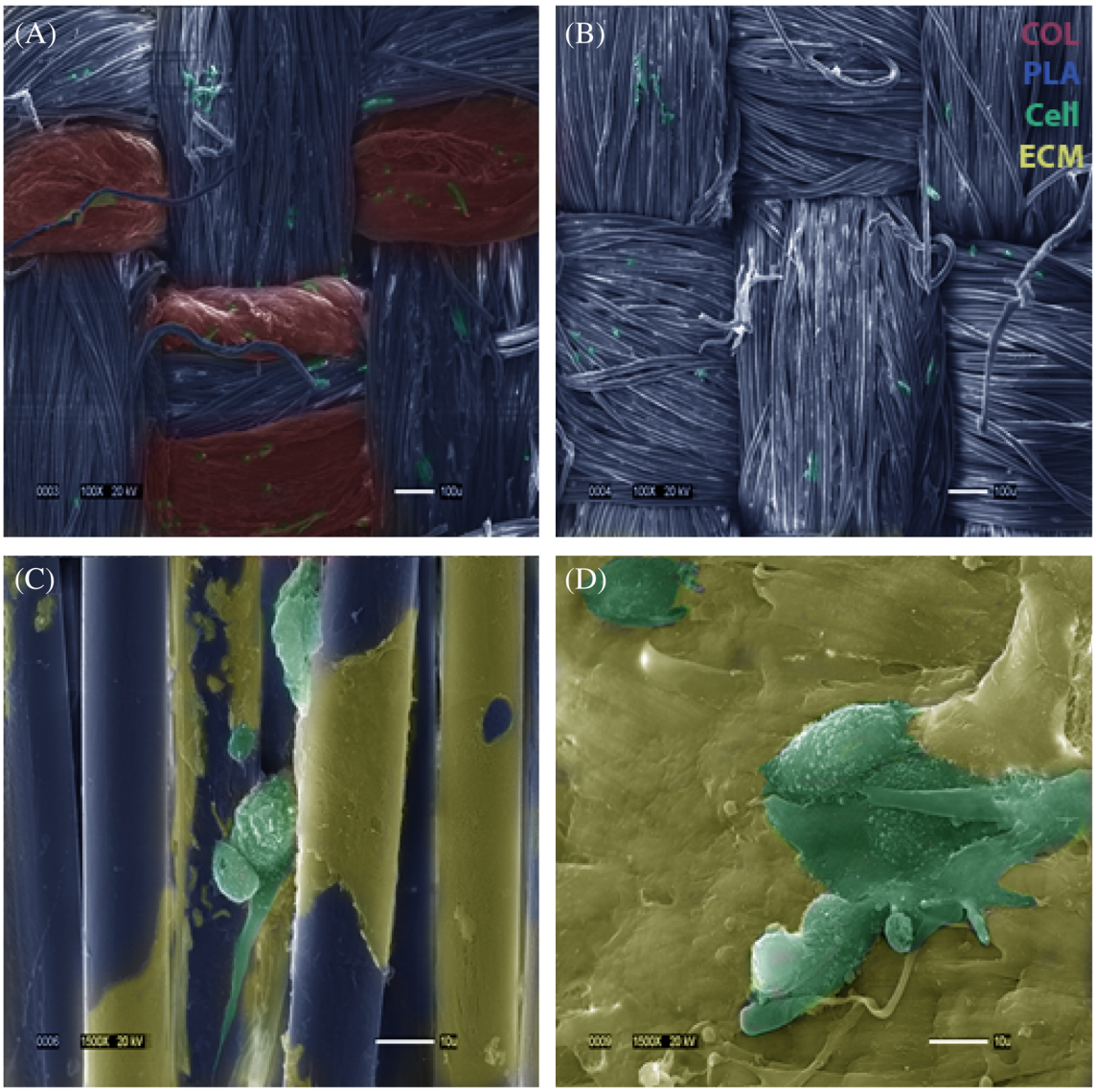

FIGURE 5 SEM images of RCs seeded for 28 days on (A) the COL/PLA patch graft and (B) the 100% PLA control graft at a magnification of 100 X, scale bar: 100 μm. Single RC cells attached to (C) a PLA filament and (D) the collagen yarn at a magnification of 1500 X. Scale bars: 10 μm. Pseudocolor: red: collagen yarns; blue: PLA yarns, gold: cell secreted extra cellular matrix; green: RCs

(Figures 4B and 4D) 2,5 cell distributions on the warp yarns (vertical direction) were equivalent to those on the weft yarns (horizontal direction).

By analyzing the confocal and SEM images of the surfaces of the COL/PLA and PLA patch grafts after 28 days of cell culture in Figures 4E, 4F and 5, it was clear that there were significant differences in cell morphology and coverage between these two grafts. The cells seeded on the collagen yarns spread more extensively with a greater cell size, indicating closer attachment (Figure 5D). On the contrary, the cells on the PLA filaments maintained their spherical morphology, indicating limited attachment (Figure 5C). The surface of the collagen yarns appeared to be rougher compared to that on the PLA yarns, which might provide evidence of the newly deposited extracellular matrix (ECM) (Figure 5C and 5D).

4 | DISCUSSION

In recent years, continuous collagen yarns have been produced by various advanced extrusion and spinning technologies.[28,35,51] Continuous collagen yarn is an attractive material for tissue engineering scaffold fabrication due to its ECM-like anisotropic morphology.[27] In previous studies, a textile scaffold was fabricated by manually weaving the collagen yarns into a plain woven structure.[27] The collagen scaffold promoted tenogenic differentiation for rotator cuff tendon repair. However, the manual weaving approach limits the ability to scale-up manufacture of collagen yarn-based scaffolds. Advanced textile technology and machinery can facilitate the scale-up of scaffold production. However, the use of textile techniques for scaffold production and tissue repair requires high mechanical strength of the

yarn materials.[5,52] In order to acquire sufficient mechanical strength, synthetic materials need to be incorporated to reinforce the collagen yarn. In this study, the collagen yarn only had an initial tensile strength of 1.3 ± 0.2 N. After reinforced by the PLA yarn, the initial strength significantly increased to 8.7 ± 0.2 N.

Not only does the scaffold need initial strength, it also needs to maintain its integrity and strength for a period of time to allow tissue healing. By using the hybrid collagen/PLA yarn to fabricate the hybrid woven graft, we aim at a secure and effective use of the scaffold for tendon repair applications over a relatively longer period of time. Given that both PLA and collagen yarns are biodegradable, we measured the degradation of the collagen yarn and collagen/PLA hybrid yarn in our previous work.[28,32] The mechanical properties of the pure collagen yarn, including maximum load, extension and stiffness, did not change significantly during the first 4 weeks of an *in vitro* degradation assay. However, the maximum tensile strength of the collagen yarn decreased from the initial 1.3 ± 0.2 N to 0.3 ± 0.1 N after 8 weeks. When further reinforced by plying with a PLA multifilament, the maximum load of the collagen/PLA hybrid yarn decreased from the initial 8.7 ± 0.2 N to 5.3 ± 1.0 N after 8 weeks of *in vitro* degradation.[32] To simplify the experimental design, we only selected the simplest woven structure, the 1/1 plain weave, with the purpose of highlighting the effect of materials on the mechanical and biological properties of the woven patches. When viewing the structure of the woven patches more closely under an optical microscope, the woven structure of the COL/PLA patch was less uniform compared to the 100% PLA patch due to the non-uniform thickness of the collagen yarns.

It is reported that the post-operative recurrence of a repaired rotator cuff tendon has a high incidence in the range of 20%–68%[52] due to (i) anastomotic failure from suture pull-out at the suture–tendon interface, (ii) tissue degeneration,[53] (iii) insufficient healing and (iv) tension overload,[54,55] particularly for large and massive tears. Augmentation scaffolds should have the potential to provide mechanical support to the repaired tissue and protect the suture site from being mechanically overloaded. However, the ECM-based tendon repair scaffolds typically show inferior mechanical properties that are neither comparable with native tendons nor with synthetic scaffolds. Commercial ECM-based scaffolds are made from decellularized animal or human tissues, such as the Zimmer Collagen device (porcine dermis), the Restore device (porcine small intestine submucosa) and the GraftJacket device (human dermis). They are reported to have an ultimate tensile load less than 50 N.[56] In comparison, the COL/PLA patch graft in this study had an ultimate tensile load of 267 N under the physiologically relevant condition, which is comparable to the commercial X-repair woven PLLA patch graft[57] and superior to previously reported ECM-based patch grafts.[56]

The addition of an augmentation graft during rotator cuff repair shares an estimated 20–35% of the load applied to the repaired tendon construct.[55,58] The supraspinatus tendon typically supports a maximum load in the range of 88–411 N.[59] The maximum load and load at 5 mm elongation of COL/PLA and PLA prototype grafts reported in this study fell within this required range under both dry and wet conditions. When compared to the elongation values at maximum load reported in the literature, the Zimmer Collagen and Restore devices have strains around 0.35–0.36, and the GraftJacket device has strains in the range of 0.7–0.8.[56] Native rotator cuff tendons have an even lower elongation of less than 0.1.[49] However, none of the experimentally fabricated COL/PLA and pure PLA prototype grafts in this study had an average breaking strain close to native tendons regardless of whether they were tested under dry or wet conditions. This is probably due to the fact that the textile structure experiences an uncrimping period under initial loading prior to the stress being applied directly to each fiber in the textile construct. Therefore, a modification on the patch structure is necessary in future studies so as to match the strain at break to native tendons.

The linear stiffness or elastic modulus is a key measurement to determine if the artificial graft has an equivalent mechanical performance to the native rotator cuff tendon. The native rotator cuff tendon may have a linear stiffness as high as 210 N mm^{-1} and an elastic modulus of over 600 MPa, depending on the type of tissue, location, position and direction of movement.[1,59–61] The X-Repair scaffold has a similar mechanical performance to native tendons. However, in this study, the COL/PLA and PLA scaffolds had a stiffness of about 89 N mm^{-1} and 115 N mm^{-1}, respectively, lower than the X-Repair device which is around 195 N mm^{-1}.[5] This is probably due to the difference in thickness between the experimental patch grafts in this study and the X-repair device, which is a double-layer woven graft with a thickness of 0.8 mm compared to the single layer woven PLA scaffolds with a thickness of approximately 0.5 mm.

For rotator cuff tendon repair, the patch scaffold has to survive not only tendon retraction, but also the movement of the shoulder at any time post-operatively. To ensure the effectiveness of a repaired tendon, the combined suture retention strength should be at least 30% of the tissue failure load, which is estimated to be between 100 and 200 N per suture.[49] Both the COL/PLA and PLA grafts had a suture retention strength (39.5 ± 3.0 N and 57.4 ± 3.4 N respectively) higher than that of the Zimmer Collagen commercial device (about 30.2 ± 7.9 N),[56] but lower than the commercial woven X-repair patch (in a range of 220–400 N) and the human rotator cuff tendon (approximately 250 N).[50] The lower suture retention strength of the COL/PLA and the PLA patch grafts in this study is most likely caused by raveling of the woven structure along the cut edge, which is likely to reduce the stress resistance of the weft yarns close to the edge. And the significant decrease in the suture retention strength of the COL/PLA woven construct is due to the inferior mechanical properties of the collagen yarn itself.[28]

An important objective of incorporating collagen yarn in the patch graft was to improve the biological performance and promote healing of torn and repaired tendons. As a key component of ECM, collagen is well recognized for its capacity to bind to cell receptors and promote intercellular communication.[62] As we expected, by incorporating collagen yarns, the number of tendon derived cells on the COL/PLA patch graft was significantly higher than on the 100% PLA patch at the end of the 28-day *in vitro* cell culture period. The presence of integrin binding sites on collagen might have contributed to the improvement of cell

attachment and growth.[63] The hydrophilicity of collagen might also improve its cell adhesion[64] compared to hydrophobic synthetic polymers, such as PLA. This probably explains why the patch graft with collagen yarns had significantly improved initial cell adhesion and early cell growth within the first 2 weeks of *in vitro* cell culture. At the same time, PLA may inhibit the growth of tendon cells and osteoblasts due to the release of lactic acid and other degradation byproducts,[51,65] which motivates the strategy to replace PLA with collagen.

Although PLA is not as biologically attractive as collagen, PLA has demonstrated its safety *in vivo* as an extensively used resorbable polymer for medical applications. The *in vitro* cell response to various rotator cuff repair scaffolds, such as the X-repair PLA woven patch, shows that synthetic materials have similar long-term biocompatibility in terms of cell growth to most biological scaffolds made from decellularized porcine or human tissues.[24] Intriguingly, in our study, the pure PLA patch achieved a similar growth rate to that of the collagen hybrid patch after 28 days, even though the initial cell growth rate was not comparable to the hybrid patch from Day 1 to Day 14. It is possible that the PLA multifilaments have a larger surface area, which can provide cells with more spaces to attach and proliferate.[24,66] Therefore, the different cell growth rate between collagen monofilmant and PLA multifilaments may not solely depend on different surface chemistries. The dimensional and morphological differences between them may also contribute to their different performances. Future studies should compare COL/PLA and PLA scaffolds that share a similar dimension and surface morphology.

Although the number of cells attached to the PLA patch graft increased significantly during the period of cell culture, the morphology of the cells attached to the PLA filaments was observed to be different compared with those on the collagen yarns. The morphology of tenocytes on various commercial synthetic scaffolds is oriented and aligned along the synthetic fiber direction with an extended cell morphology, while those on ECM-based scaffolds spread out randomly in numerous different directions.[24] The fluorescence confocal microscopic images and the SEM images in this study were consistent with this finding; namely the hybrid patch with its collagen component gave a more favorable biological response than the pure PLA patch in terms of cell growth and cell morphology. On the collagen yarns, the attached cells were flatter, more spread out with extended pseudopods. In contrast, the cell morphology on the PLA filaments appeared to be in more of a spherical form, aligned along the axis of the PLA filaments and wrapping around individual filaments.

There are some limitations in this pilot study that need to be addressed in the future. In this study, we focused on the feasibility of fabricating the hybrid scaffold using an industrial weaving technology to facilitate the scale-up manufacture of the COL/PLA hybrid patch. Upon successful fabrication of the patches with automated technology, we further measured their initial mechanical properties. However, given both the PLA and collagen are biodegradable materials, any changes in mechanical properties over time are also important to monitor. Although the degradation of both the collagen and the hybrid COL/PLA yarn have been evaluated in other studies,[28,32] the impact of degradation on the mechanical properties of the resulting hybrid patch has not yet been measured. A long-term *in vivo* degradation study is recommended in the future to further determine if the hybrid patch can provide mechanical support until the repaired tendon is sufficiently healed. In addition, in the case when cutting is needed during surgery to adjust the size of the patch for the specific patient, yarns raveling out the edge of the patch may reduce the suture retention strength. Such concern will need to be considered and addressed in the future study by improving the structural design and fixing or reinforcing the edges of the patch. In this study, we evaluated the cytocompatibility of the hybrid patch by measuring the morphology and proliferation of the seeded tendon-derived cells. It would be necessary to conduct further in-depth *in vitro* studies to determine the tenogenic effect of the patch based on gene expression and protein production and to perform *in vivo* studies to validate the tenogenic potention and the immune host response to the hybrid patch in clinically relevant animal models.

5 | CONCLUSIONS

In this preliminary study, using a high-speed industry-scale weaving technology, a collagen/PLA hybrid textile patch was successfully designed and fabricated. It provided adequate mechanical support for use as an augmentation or bridging patch graft for tendon repair, and promoted the proliferation of tendon-derived cells. To the best of our knowledge, this is the first time that hybrid COL/PLA yarns were woven into a ribbon scaffold using an industrial-scale automated weaving technology. Although the tensile properties of the hybrid patch containing collagen were inferior to the pure PLA control patch graft, particularly when it was wet, its tensile properties were still comparable with a native rotator cuff tendon. Additionally, the presence of the collagen in the hybrid scaffold promoted cell proliferation and an extended cell morphology compared to those cells attached to the pure PLA scaffold. It was also observed that the cells grew preferentially on certain regions of the collagen yarns, while the cells on the PLA scaffold were observed at a lower density. Based on this preliminary study, we believe that this hybrid textile scaffold containing collagen yarns is a promising candidate for long-term mechanical support and tendon tissue regeneration in various orthopedic applications. In addition, this study has demonstrated that scale-up of this technology is feasible, thereby facilitating commercialization of the device and enabling translation to the clinic.

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DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from author (YX) upon reasonable request.

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REFERENCES

1. Lomas AJ, Ryan CNM, Sorushanova A, et al. The past, present and future in scaffold-based tendon treatments. *Adv Drug Deliv Rev*. 2015; 84:257-277. doi:10.1016/j.addr.2014.11.022
2. Thangarajah T, Pendegrass CJ, Shahbazi S, Lambert S, Alexander S, Blunn GW. Augmentation of rotator cuff repair with soft tissue scaffolds. *Orthop J Sports Med*. 2015;3:1-8. doi:10.1177/2325967115587495
3. Mori D, Funakoshi N, Yamashita F. Arthroscopic surgery of irreparable large or massive rotator cuff tears with low-grade fatty degeneration of the infraspinatus: patch autograft procedure versus partial repair procedure, arthroscopy. *J Arthroscopic Related Surg*. 2013;29: 1911-1921. doi:10.1016/j.arthro.2013.08.032
4. Neri BR, Chan KW, Kwon YW. Management of massive and irreparable rotator cuff tears. *J Shoulder Elbow Surg*. 2009;18:808-818. doi:10.1016/j.jse.2009.03.013
5. Derwin KA, Codsi MJ, Milks RA, Baker AR, McCarron JA, Iannotti JP. Rotator cuff repair augmentation in a canine model with use of a woven poly-L-lactide device. *J Bone Joint Surg Am*. 2009;91:1159-1171. doi:10.2106/JBJS.H.00775
6. van der Meijden OA, Wijdicks CA, Gaskill TR, Jansson KS, Millett PJ. Biomechanical analysis of two tendon posterosuperior rotator cuff tear repairs: extended linked repairs and augmented repairs. *Art Ther*. 2013;29:37-45. doi:10.1016/j.arthro.2012.07.012
7. Clark RR, Dierckman BD, Bahk MS, Ghodadra NS, Snyder SJ, Burns JP. Patch augmentation for rotator cuff repair: indications, techniques, and outcomes. *Oper Tech Sports Med*. 2012;20:224-232. doi:10.1053/j.otsm.2012.07.001
8. Gupta AK, Hug K, Boggess B, Gavigan M, Toth AP. Massive or 2-tendon rotator cuff tears in active patients with minimal glenohumeral arthritis: clinical and radiographic outcomes of reconstruction using dermal tissue matrix xenograft. *Am J Sports Med*. 2013;41:872-879. doi:10.1177/0363546512475204
9. Gupta AK, Hug K, Berkoff DJ, et al. Dermal tissue allograft for the repair of massive irreparable rotator cuff tears. *Am J Sports Med*. 2012;40:141-147. doi:10.1177/0363546511422795
10. Gilot GJ, Alvarez-Pinzon AM, Barcksdale L, Westerdahl D, Krill M, Peck E. Outcome of large to massive rotator cuff tears repaired with and without extracellular matrix augmentation: a prospective comparative study. *Arthroscopy J Arthroscopic Related Surg*. 2015;31:1459-1465. doi:10.1016/j.arthro.2015.02.032
11. Soler JA, Gidwani S, Curtis MJ. Early complications from the use of porcine dermal collagen implants (permacol™) as bridging constructs in the repair of massive rotator cuff tears: a report of 4 cases. *Acta Orthop Belg*. 2007;2007:432-436.
12. Walton JR, Bowman NK, Khatib Y, Linklater J, Murrell GA. Restore orthobiologic implant: not recommended for augmentation of rotator cuff repairs. *J Bone Joint Surg Am*. 2007;89:786-791.
13. Malcarney HL, Bonar F, Murrell GA. Early inflammatory reaction after rotator cuff repair with a porcine small intestine submucosal implant: a report of 4 cases. *Am J Sports Med*. 2005;339:907-911. doi:10.1177/0363546504271500
14. Zheng M, Chen J, Kirilak Y, Willers C, Xu J, Wood D. Porcine small intestine submucosa (SIS) is not an acellular collagenous matrix and contains porcine DNA: possible implications in human implantation. *J Biomed Mater Res B Appl Biomater*. 2005;73:61-67.
15. Sclamberg SG, Tibone JE, Itamura JM, Kasraeian S. Six-month magnetic resonance imaging follow-up of large and massive rotator cuff repairs reinforced with porcine small intestinal submucosa. *J Shoulder Elbow Surg*. 2004;13:538-541. doi:10.1016/j.jse.2004.03.005
16. Moore DR, Cain EL, Schwartz ML, Clancy WG. Allograft reconstruction for massive, irreparable rotator cuff tears. *Am J Sports Med*. 2006;34:392-396. doi:10.1177/0363546505281237
17. Encalada-Diaz I, Cole BJ, MacGillivray JD, et al. Rotator cuff repair augmentation using a novel polycarbonate polyurethane patch: preliminary results at 12 months' follow-up. *J Shoulder Elbow Surg*. 2011; 20:788-794. doi:10.1016/j.jse.2010.08.013
18. Koh JL, Szomor Z, Murrell GA, Warren RF. Supplementation of rotator cuff repair with a bioresorbable scaffold. *Am J Sports Med*. 2002; 30:410-413. doi:10.1177/03635465020300031701
19. Proctor CS. Long-term successful arthroscopic repair of large and massive rotator cuff tears with a functional and degradable reinforcement device. *J Shoulder Elbow Surg*. 2014;23:1508-1513. doi:10.1016/j.jse.2014.01.010
20. Kimura A, Aoki M, Fukushima S, Ishii S, Yamakoshi K. Reconstruction of a defect of the rotator cuff with polytetrafluoroethylene felt graft: recovery of tensile strength and histocompatibility in an animal model. *J Bone Joint Surg Br*. 2003;85:282-287. doi:10.1302/0301-620X.85B2.12823
21. Lenart BA, Martens KA, Kearns KA, Gillespie RJ, Zoga AC, Williams GR. Treatment of massive and recurrent rotator cuff tears augmented with a poly-l-lactide graft, a preliminary study. *J Shoulder Elbow Surg*. 2015;24: 915-921. doi:10.1016/j.jse.2014.09.044
22. Inui A, Kokubu T, Fujioka H, et al. Application of layered poly (L-lactic acid) cell free scaffold in a rabbit rotator cuff defect model. *Sports Med Arthrosc Rehabil Ther Technol*. 2011;3:1-7. doi:10.1186/1758-2555-3-29
23. Thorpe CT, Birch HL, Clegg PD, Screen HR. Tendon physiology and mechanical behavior: structure–function relationships. In: Gomes ME, Reis RL, Rodrigues MT, eds. *Tendon Regeneration*. Elsevier Inc.; 2015:3-39.
24. Smith RD, Carr A, Dakin SG, Snelling SJ, Yapp C, Hakimi O. The response of tenocytes to commercial scaffolds used for rotator cuff repair. *Euro Cells Mater J*. 2016;31(2016):107-118.
25. Röhlecke C, Witt M, Kasper M, Schulze E, Wolf C, Hofer A, Funk RW. Synergistic effect of titanium alloy and collagen type I on cell adhesion, proliferation and differentiation of osteoblast-like cells. *Cells Tissues Organs* 168(2001):178-187. doi:10.1159/000047833
26. Dolder Van den, Bancroft GN, Sikavitsas VI, Spauwen PH, Mikos AG, Jansen JA. Effect of fibronectin-and collagen I-coated titanium fiber mesh on proliferation and differentiation of osteogenic cells. *Tissue Eng*. 9(2003):505-515. doi:10.1089/107632703322066688
27. Younesi M, Islam A, Kishore V, Anderson JM, Akkus O. Tenogenic induction of human MSCs by anisotropically aligned collagen biotextiles. *Adv. Funct. Mater*. 24(2014):5762-5770. doi:10.1002/adfm.201400828
28. Xie Y, Chen J, Celik H, Akkus O, King MW. Evaluation of an electrochemically aligned collagen yarn for textile scaffold fabrication. *Biomed. Mater*. 16(2021):025001. doi:10.1088/1748-605X/abdf9e
29. Uquillas JA, Kishore V, Akkus O. Genipin crosslinking elevates the strength of electrochemically aligned collagen to the level of tendons. *J Mech Behav Biomed Mater*. 2012;15:176-189. doi:10.1016/j.jmbbm.2012.06.012
30. Zhang F, King MW. Biodegradable polymers as the pivotal player in the design of tissue engineering scaffolds. *Adv Healthc Mater*. 2020; 9 (13): e1901358. doi:10.1002/adhm.201901358
31. Zhang F, Xie Y, Celik H, Akkus O, Bernacki SH, King MW. Engineering small-caliber vascular grafts from collagen filaments and nanofibers with comparable mechanical properties to native vessels. *Biofabrication*. 2019;11:1-17. doi:10.1088/1758-5090/ab15ce
32. Zhang F, Bambharoliya T, Xie Y, et al. A hybrid vascular graft harnessing the superior mechanical properties of synthetic fibers and the biological performance of collagen filaments. *Mater Sci Eng C: Mater Biol Appl*. 2021;118:111418. doi:10.1016/j.msec.2020.111418
33. Learn GD, McClellan PE, Knapik DM, et al. Woven collagen biotextiles enable mechanically functional rotator cuff tendon regeneration during repair of segmental tendon defects in vivo. *J Biomed Mater Res B*. 2019;107:1864-1876.

34. Ceonzo K, Gaynor A, Shaffer L, Kojima K, Vacanti CA, Stahl GL. Polyglycolic acid-induced inflammation: role of hydrolysis and resulting complement activation. *Tissue Eng*. 2006;12:301-308. doi:10.1089/ten.2006.12.ft-21
35. Wu D, Chen X, Chen T, Ding C, Wu W, Li J. Substrate-anchored and degradation-sensitive anti-inflammatory coatings for implant materials. *Sci Rep*. 2015;5:1-12. doi:10.1038/srep11105
36. Hakimi O, Mouthuy PA, Zargar N, Lostis E, Morrey M, Carr A. A layered electrospun and woven surgical scaffold to enhance endogenous tendon repair. *Acta Biomater*. 2015;26:124-135. doi:10.1016/j.actbio.2015.08.007
37. Wu S, Wang Y, Streubel PN, Duan B. Living nanofiber yarn-based woven biotextiles for tendon tissue engineering using cell tri-culture and mechanical stimulation. *Acta Biomater*. 2017;62:102-115. doi:10.1016/j.actbio.2017.08.043
38. Gao Y, Shao W, Qian W, et al. Biomineralized poly (l-lactic-co-glycolic acid)-tussah silk fibroin nanofiber fabric with hierarchical architecture as a scaffold for bone tissue engineering. *Mater Sci Eng C*. 2018;84:195-207. doi:10.1016/j.msec.2017.11.047
39. Gilmore J, Burg T, Groff RE, Burg KJ. Design and optimization of a novel bio-loom to weave melt-spun absorbable polymers for bone tissue engineering. *J Biomed Mater Res B Appl Biomater*. 2017;105:1342-1351. doi:10.1002/jbm.b.33700
40. Persson M, Lehenkari PP, Berglin L, et al. Osteogenic differentiation of human mesenchymal stem cells in a 3D woven scaffold. *Sci Rep*. 2018;8:10457. doi:10.1038/s41598-018-28699-x
41. Huynh NP, Brunger JM, Gloss CC, Moutos FT, Gersbach CA, Guilak F. Genetic engineering of mesenchymal stem cells for differential matrix deposition on 3D woven scaffold. *Tissue Eng*. 2018;24:1531-1544. doi:10.1089/ten.TEA.2017.0510
42. Friedman JM, Sennett ML, Bonadio MB, et al. Comparison of fixation techniques of 3D-woven poly (ϵ-Caprolactone) scaffolds for cartilage repair in a weightbearing porcine large animal model. *Cartilage*. 2017;9:428-437. doi:10.1177/1947603517700953
43. Moffat KL, Goon K, Moutos FT, et al. Hybrid cellularized structures created from an interpenetrating polymer network hydrogel reinforced by a 3D woven scaffold. *Macromol Biosci*. 2018;18:1800140. doi:10.1002/mabi.201800140
44. Liberski A, Ayad N, Wojciechowska D, et al. Weaving for heart valve tissue engineering. *Biotech Adv*. 2017;35:633-656. doi:10.1016/j.biotechadv.2017.07.012
45. Wu S, Duan B, Qin X, Butcher JT. Living nano-micro fibrous woven fabric/hydrogel hybrid scaffolds for heart valve engineering. *Acta Biomater*. 2017;51:89-100. doi:10.1016/j.actbio.2017.01.051
46. Li G, Liu Y, Lan P, Li Y, Li Y. A prospective bifurcated biomedical stent with seamless woven structure. *J Text*. 2013;I(104):1017-1023. doi:10.1080/00405000.2013.767429
47. Yokota T, Ichikawa H, Matsumiya G, et al. In situ tissue regeneration using a novel tissue-engineered, small-caliber vascular graft without cell seeding. *J Thorac Cardiovasc Surg*. 2008;136:900-907. doi:10.1016/j.jtcvs.2008.02.058
48. Iwai S, Sawa Y, Taketani S, Torikai K, Hirakawa K, Matsuda H. Novel tissue-engineered biodegradable material for reconstruction of vascular wall. *Ann Thorac Surg*. 2005;80:1821-1827. doi:10.1016/j.athoracsur.2005.03.139
49. Ratcliffe A, Butler DL, Dyment NA, et al. Scaffolds for tendon and ligament repair and regeneration. *Ann Biomed Eng*. 2015;43:819-831. doi:10.1007/s10439-015-1263-1
50. Aurora A, Mesiha M, Tan CD, et al. Mechanical characterization and biocompatibility of a novel reinforced fascia patch for rotator cuff repair. *J Biomed Mater Res A*. 2011;99:221-230. doi:10.1002/jbm.a.33179
51. Yaari A, Schilt Y, Tamburu C, Raviv U, Shoseyov O. Wet spinning and drawing of human recombinant collagen. *ACS Biomater Sci Eng*. 2016;2:349-360. doi:10.1021/acsbiomaterials.5b00461
52. Ciampi P, Scotti C, Nonis A, et al. The benefit of synthetic versus biological patch augmentation in the repair of posterosuperior massive rotator cuff tears: a 3-year follow-up study. *Am J Sports Med*. 2014;42:1169-1175. doi:10.1177/0363546514525592
53. Hakimi O, Mouthuy PA, Carr A. Synthetic and degradable patches: an emerging solution for rotator cuff repair. *Int J Exp Pathol*. 2013;94:287-292. doi:10.1111/iep.12030
54. Cummins CA, Murrell GA. Mode of failure for rotator cuff repair with suture anchors identified at revision surgery. *J Shoulder Elbow Surg*. 2003;12:128-133. doi:10.1067/mse.2003.21
55. McCarron JA, Milks RA, Mesiha M, et al. Reinforced fascia patch limits cyclic gapping of rotator cuff repairs in a human cadaveric model. *J Shoulder Elbow Surg*. 2012;21:1680-1686. doi:10.1016/j.jse.2011.11.039
56. Chaudhury S, Holland C, Thompson MS, Vollrath F, Carr AJ. Tensile and shear mechanical properties of rotator cuff repair patches. *J Shoulder Elbow Surg*. 2012;21:1168-1176. doi:10.1016/j.jse.2011.08.045
57. Smith RD, Zargar N, Brown CP, et al. Characterizing the macro and micro mechanical properties of scaffolds for rotator cuff repair. *J Shoulder Elbow Surg*. 2017;26:2038-2046. doi:10.1016/j.jse.2017.06.035
58. Shea KP, Obopilwe E, Sperling JW, Iannotti JP. A biomechanical analysis of gap formation and failure mechanics of a xenograft-reinforced rotator cuff repair in a cadaveric model. *J Shoulder Elbow Surg*. 2012;21:1072-1079. doi:10.1016/j.jse.2011.07.024
59. Itoi E, Berglund LJ, Grabowski JJ, et al. Tensile properties of the supraspinatus tendon. *J Orthop Res*. 1995;13:578-584. doi:10.1002/jor.1100130413
60. Halder A, Zobitz M, Schultz F, An K. Mechanical properties of the posterior rotator cuff. *Clin Biomech*. 2000;15:456-462. doi:10.1016/S0268-0033(99)00095-9
61. Halder A, Zobitz M, Schultz F, An K. Structural properties of the subscapularis tendon. *J Orthop Res*. 2000;18:829-834. doi:10.1002/jor.1100180522
62. Whelan MC, Senger D. Collagen I initiates endothelial cell morphogenesis by inducing Actin polymerization through suppression of cyclic AMP and protein kinase a. *J Biol Chem*. 2003;278:327-334. doi:10.1074/jbc.M207554200
63. Chang HI, Wang Y. Cell responses to surface and architecture of tissue engineering scaffolds. In: Eberli D, ed. *Regenerative Medicine and Tissue Engineering-Cells and Biomaterials*. InTechOpen Ltd.; 2011:569-588.
64. Liu X, Zheng C, Luo X, Wang X. Recent advances of collagen-based biomaterials: multi-hierarchical structure, modification and biomedical applications. *Mater Sci Eng C*. 2019;99:1509-1522. doi:10.1016/j.msec.2019.02.070
65. Meyer F, Wardale J, Best S, Cameron R, Rushton N, Brooks R. Effects of lactic acid and glycolic acid on human osteoblasts: a way to understand PLGA involvement in PLGA/calcium phosphate hybrid failure. *J Orthop Res*. 2012;30:864-871. doi:10.1002/jor.22019
66. Murphy CM, Haugh MG, O'Brien FJ. The effect of mean pore size on cell attachment, proliferation and migration in collagen-glycosaminoglycan scaffolds for bone tissue engineering. *Biomaterials*. 2010;31:461-466. doi:10.1016/j.biomaterials.2009.09.063

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