ANNALS OF TRANSPLANTATION

ORIGINAL PAPER

e-ISSN 2329-0358
© Ann Transplant, 2019; 24: 532-540
DOI: 10.12659/AOT.917518

Received: 2019.05.13
Accepted: 2019.07.05
Published: 2019.09.17

Allogenic Tendon-Autologous Cartilage Cells Transplantation Enhances Adhesive/Growth Ability and Promotes Chondrogenesis in a Rabbit Model of Glenoid Labrum Damage

Authors' Contribution:
Study Design A
Data Collection B
Statistical Analysis C
Data Interpretation D
Manuscript Preparation E
Literature Search F
Funds Collection G

BCDEF 1 Guofei Chen
BCDF 2 Peng Chen
BCDF 2 Tian You
BCDF 2 Xiaocheng Jiang
BCF 2 Wei Li
ADEG 2 Changqing Jiang

1 Department of Traumatic Arthrosis Orthopaedics, University of Chinese Academy of Sciences-Shenzhen Hospital Shenzhen, Shenzhen, Guangdong, P.R. China
2 Department of Sports Medicine, Peking University Shenzhen Hospital, Shenzhen, Guangdong, P.R. China

Corresponding Author: Changqing Jiang, e-mail: jcq_006@yeah.net
Source of support: This study was funded by the Shenzhen Priority Fields of Study (Grant No. 201606007) and Shenzhen Science and Technology Innovation Committee Funded Project (Grant No. JCYJ20150403091443323)

Background: Glenoid labrum injury of the shoulder commonly occurs in athletes, especially those who perform throwing motions. This study investigated the effects of the established allogenic tendon-autologous cartilage cells reconstruction approach in a rabbit model of glenoid labrum damage.

Material/Methods: The allogenic tendons were isolated and extracted using the chemical extraction method. Cartilage cells were isolated from New Zealand rabbits and identified by detecting type II collagenase. The allogenic tendon-autologous cartilage cells were transplanted to the damaged glenoid labrum. HE staining was used to observe inflammatory cells, Masson staining was used to observe muscle fibers, and scanning electron microscopy (SEM) was used to assess antigenicity of tendon tissues. PSA and AB staining were used to examine neutral protein mucopolysaccharide and acidic protein mucopolysaccharide, respectively. We assessed cartilage cell growth in autologous cartilage cells combined with allogenic tendon transplanted tissues.

Results: Allogenic tendons were well prepared using chemical extraction method due to use of HE staining, Masson staining, and SEM. TGF-β1 treatment induced cartilage cell formation and triggered expression of acidic and neutral protein mucopolysaccharides. HE staining, Masson staining, PAS staining, and AB staining methods showed that autologous cartilage cells combined with allogenic tendon transplanted tissues had better growth of cartilage cells.

Conclusions: This study establishes the allogenic tendon-autologous cartilage cells reconstruction and transplantation approach and illustrated higher adhesive ability and growth ability, and better chondrogenesis in a rabbit model of glenoid labrum damage.

MeSH Keywords: Autografts • Cartilage • Glenoid Cavity • Transplantation, Homologous

Full-text PDF: https://www.annalsoftransplantation.com/abstract/index/idArt/917518

Indexed in: [Science Citation Index Expanded] [Index Medicus/MEDLINE] [Chemical Abstracts] [Scopus]

Background

Worldwide population aging is associated with increasing incidence of health problems such as motor vehicle injuries, bone fractures, and joint injuries [1,2]. Due to the professionalization of sports, the incidence of shoulder dislocations has also increased in recent decades. Shoulder dislocations now account for an estimated 50% of all joint dislocations in humans. The re-occurrence rate after the first shoulder dislocation is high, especially for patients under 20 years old, and can be as high as 90% in that age group [3].

Glenoid labrum shoulder injuries are common in athletes, especially in athletes who perform throwing motions. Snyder et al. [4] first described and named glenoid labrum injuries. Glenoid labrum suture under arthroscopic guidance considered to be the optimal therapeutic approach for shoulder dislocation and glenoid labrum injury. In recent years, surgical repair for glenoid labrum injury has been considered to be challenging, but most of these injuries can be cured with minimally invasive endoscopic repair [5,6]. For patients undergoing repeated shoulder dislocations, the glenoid labrum always should never be sutured. Therefore, glenoid labrum reconstruction has become a promising approach to treat shoulder dislocations. Paulos et al. [7] used glenoid labrum reconstruction to repair anterior and anterior-inferior shoulder instability and achieved satisfactory outcomes. Bateman et al. [8] reported that autograft-allograft construct combining reverse shoulder arthroplasty could repair uncontained glenoid defects. Therefore, glenoid labrum reconstruction might be a promising approach for treating shoulder joint disorders.

In this study, we established a reconstructive approach for glenoid labrum injury by using 2 steps. Firstly, allogenic tendon was extracted by using Triton X-100 and sodium deoxycholate, and this was used as scaffold material for cell culture. Secondarily, autologous cartilage cells were cultured on allogenic tendon supplemented with appropriate concentrations of TGF-β1. Finally, transplanted autologous cartilage cells with allogenic tendon were used to treat glenoid labrum damage.

The established allogenic tendon-autologous cartilage cells reconstruction approach demonstrated higher adhesive ability and growth ability, and better chondrogenesis in the *in vivo* animal model. The allogenic tendon-autologous cartilage cells reconstruction method could become a clinically useful alterative to the use of extrasynovial autografts for repair of glenoid labrum damage, and the transplantation of allogenic tendon-autologous cartilage cells could be a practical approach to replace/reconstruct the diseased or injured glenoid labrum.

Material and Methods

Animals

Thirty-seven adult New Zealand white rabbits weighting 2.5–3 kg, were purchased from the Beijing Huafukang Biosci. Co. (Beijing, China). Twenty rabbits were used to isolate the allogenic tendon, 10 rabbits were used to isolate the autologous cartilage cells, and the other 15 rabbits were used to establish the glenoid labrum resection models. All rabbits used in this study were housed in commercial animal cages under the same conditions. The rabbits had free access to water, food, and bedding.

The animal experiments were approved by the Ethics Committee of Peking University Shenzhen Hospital, Shenzhen, China. All animals were handled in accordance with the guidelines for care and use of laboratory animals of the National Institute of Health (NIH).

Preparation of allogenic tendon

Isolation of tendons was conducted according to the method described in a previous study [9]. Briefly, immediately after sacrifice, the hind paws were harvested and stored at –80°C. After thawing at room temperature, a total of 32 flexor digitorum profundus (FDP) tendons with distal phalanx were isolated from the 3rd and 4th digits (Figure 1A).

The isolated tendons were divided into chemical extraction group 1, group 2, and group 3, and a control group (3 per group). After removing the distal phalanx, tendons in the control group were lyophilized, sterilized, and stored at –80°C. The chemical extraction was performed with the following steps: 1) The tendons were soaked in 100 ml sterilized distilled water for 6 h, and washed with sterilized distilled water 3 times for 5 min each time. 2) The tendons were soaked in 100 ml 4% (Sigma-Aldrich, St. Louis, Missouri, USA), and washed 3 times with sterilized distilled water for 5 min each time. 3) The tendons were soaked in 100 ml sterilized distilled water for 6 h, and washed with sterilized distilled water 3 times for 5 min each time. 4) The tendons were soaked for 12 h in 100 ml 4% sodium deoxycholate (Sangon Biotech. Co., Shanghai, China), and washed with sterilized distilled water 3 times, and then washed with sterilized distilled water 3 times for 5 min each time. 5) The tendons were soaked in 100 ml sterilized distilled water for 12 h. Chemical extraction group 1 underwent the chemical extraction process 1 time, chemical extraction group 2 underwent the chemical extraction process 2 times, and chemical extraction group 3 underwent the chemical extraction process 3 times. Tendons in the control group did not receive any treatment.

Indexed in: [Science Citation Index Expanded] [Index Medicus/MEDLINE] [Chemical Abstracts] [Scopus]

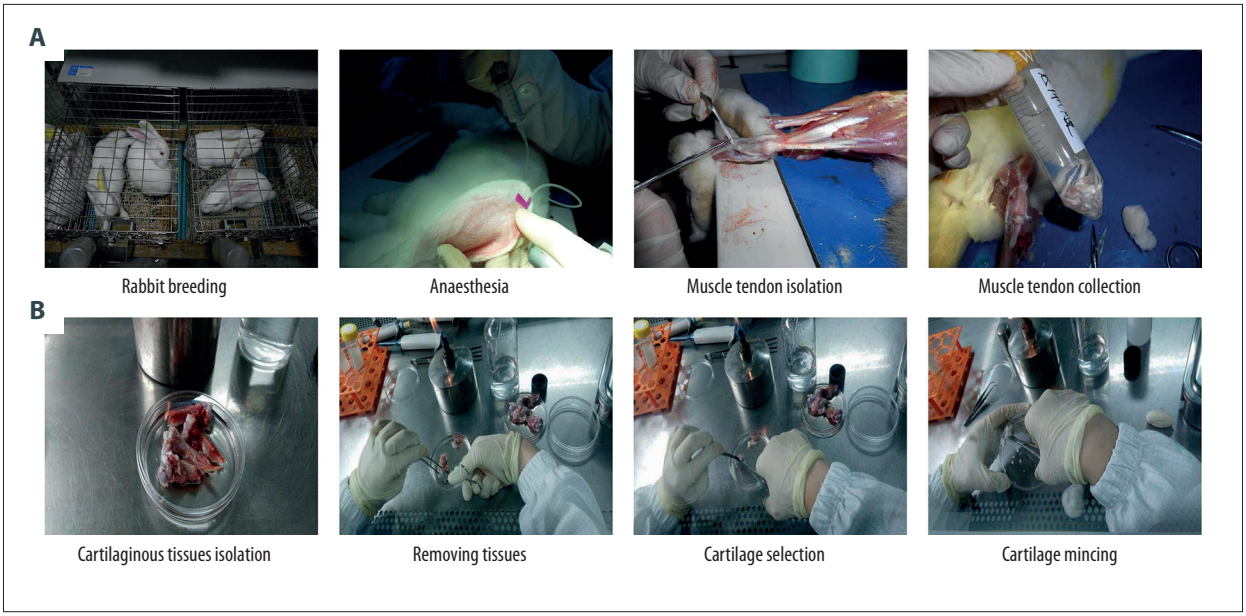

Figure 1. Preparation of allogenic tendon and cartilage cells isolated from New Zealand white rabbits. (**A**) Isolating processes of the allogenic tendons. (**B**) Isolating processes of the cartilage cells.

Preparation of cartilage cells

To establish the allogenic tendon-autologous cartilage cells reconstruction tool, the cartilage cells were isolated and cultured *in vitro* (Figure 1B). The New Zealand rabbits were anaesthetized using 7% chloral hydrate. The articular cartilage was removed under aseptic conditions and washed with PBS. Then, the cartilage cells were extracted by digesting the cartilage chips with type II collagenase (Sangon Biotech. Co., Shanghai, China) at 37°C for 12 h and with 5% CO_2 according to a previously reported method [10]. The suspension of cartilage cells were filtered with a 70-μm nylon mesh, and isolated cartilage cells were washed with PBS solution. The cartilage cells were cultured in DMEM (Gibco BRL, Grand Island, NY, USA), and cartilage cells at passage 3 were used. Cartilage cells were treated with TGF-β1 at final concentrations of 0.1 ng/ml and 0.2 ng/ml for use in further experiments.

Establishment of a rabbit model of anterior-posterior labrum shoulder joint injury

In this study, the glenoid labrum lesions were prepared according to the method described in a previous study [11]. In brief, the rabbits were anaesthetized by intraperitoneal injection of 7% chloral hydrate, and the forelimb skins were preserved and sterilized with 75% alcohol. Then, the skin at the anterior shoulder joint was cut open for 4 cm lengthways, and the glenoid labrum was exposed. A 2×2 cm piece of glenoid labrum was cut off from the shoulder joint of rabbits. Then, the rabbit model of anterior-posterior glenoid labrum was successfully established (Figure 2).

Transplantation of allogenic tendon-autologous cartilage cells and reconstruction of glenoid labrum

The chemically-extracted tendons were sliced into appropriate lengths and transplanted into the shoulder joint model rabbits. The tendons were fit to the remaining glenoid labrum and the resected glenoid labrum was filled with the tendons. The edge of the glenoid labrum was sutured and fixed together with the articular capsule and soft tissues around the glenoid cavity. The sutured glenoid labrum was disinfected with H_2O_2, saline water, and iodophor. Then, the autologous cartilage cells were injected into the articular cavity of the rabbits. Transplantation was performed according to a previously described method [12].

Immunohistochemistry assay

Cartilage cells were fixed with 4% paraformaldehyde (Sangon Biotech., Shanghai, China) for 15 min and the endogenous peroxidase was inactivated with 3% hydrogen peroxide (Beyotime Biotech, Shanghai, China) at room temperature for 5 min. The cells were then blocked with 5% bovine serum albumin (BSA, Gibco BRL. Co., Grand Island, NY, USA) for 20 min and incubated with rabbit anti-rabbit collagen II polyclonal antibody (1: 3000, Cat. No. ab34712, Abcam Biotech., Cambridge, MA, USA) at 4°C overnight. Then, the cells were incubated with biotin-conjugated goat anti-rabbit IgG (1: 1000, Cat. No. ab6720, Abcam Biotech) at room temperature for 1 h. Finally, the images of stained cells were captured with an inverted fluorescence microscope (Mode: CKX 41, Olympus, Japan).

Indexed in: [Science Citation Index Expanded] [Index Medicus/MEDLINE] [Chemical Abstracts] [Scopus]

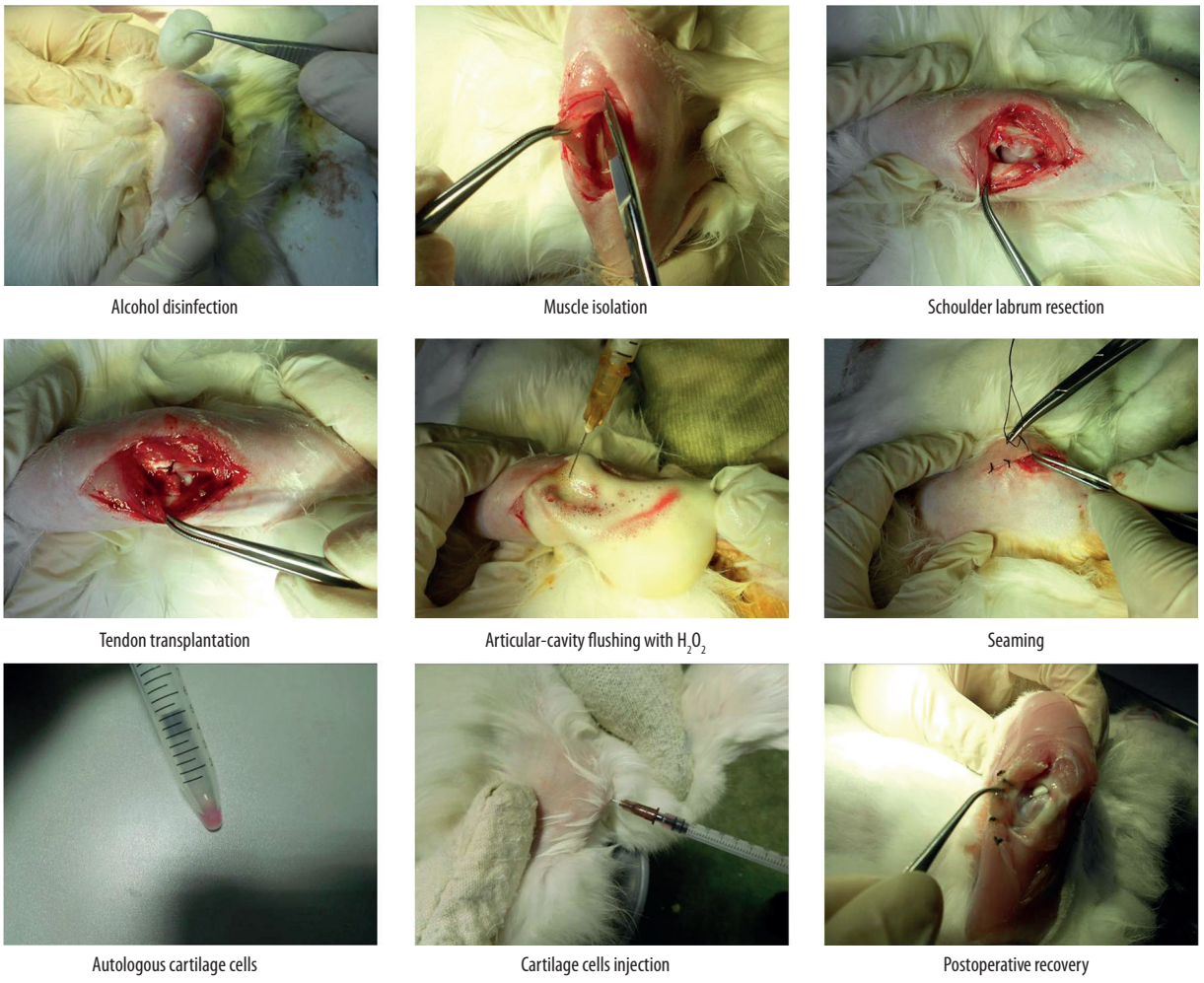

Figure 2. Images of establishment of the glenoid labrum damage rabbit model and the autologous cartilage cells combined with allogenic tendon transplantation processes.

Histological staining

Tendons and the formed allogenic tendon-autologous tissues were examined using hematoxylin-eosin (HE) staining, Masson staining, periodic Acid-Schiff (PAS) staining, and Albert (AB) staining using previously described methods [13,14]. HE staining was conducted by using the hematoxylin (Nanjing Jiancheng Biotech. Co., Nanjing, China) and eosin (Beyotime Biotech, Shanghai, China) according to the manufacturer's instructions. PAS staining was performed using a Periodic Acid-Schiff detection kit (Nanjing Jiancheng Biotech. Co., Nanjing, China). AB staining was conducted by using a commercial AB-PAS staining kit (Cat. No. DG0007, Beijing Leagene Biotech. Co., Beijing, China). The above stains were conducted by using the commercial kits (Sangon Biotech. Co., Shanghai, China).

Scanning electron microscopy (SEM) analysis

To observe the antigenic components and extracellular matrix, SEM analysis was performed routinely according to the methods described previously [15] using a Philips XL30E SEM device (Philips, Netherlands).

Results

Allogenic tendons were well prepared using chemical extraction method

The allogenic tendons had to be without inflammatory factors (as determined by HE method) and without muscle fibers (as determined by Masson staining). Therefore, the HE staining and Masson staining methods were used in this study. HE staining

Indexed in: [Science Citation Index Expanded] [Index Medicus/MEDLINE] [Chemical Abstracts] [Scopus]

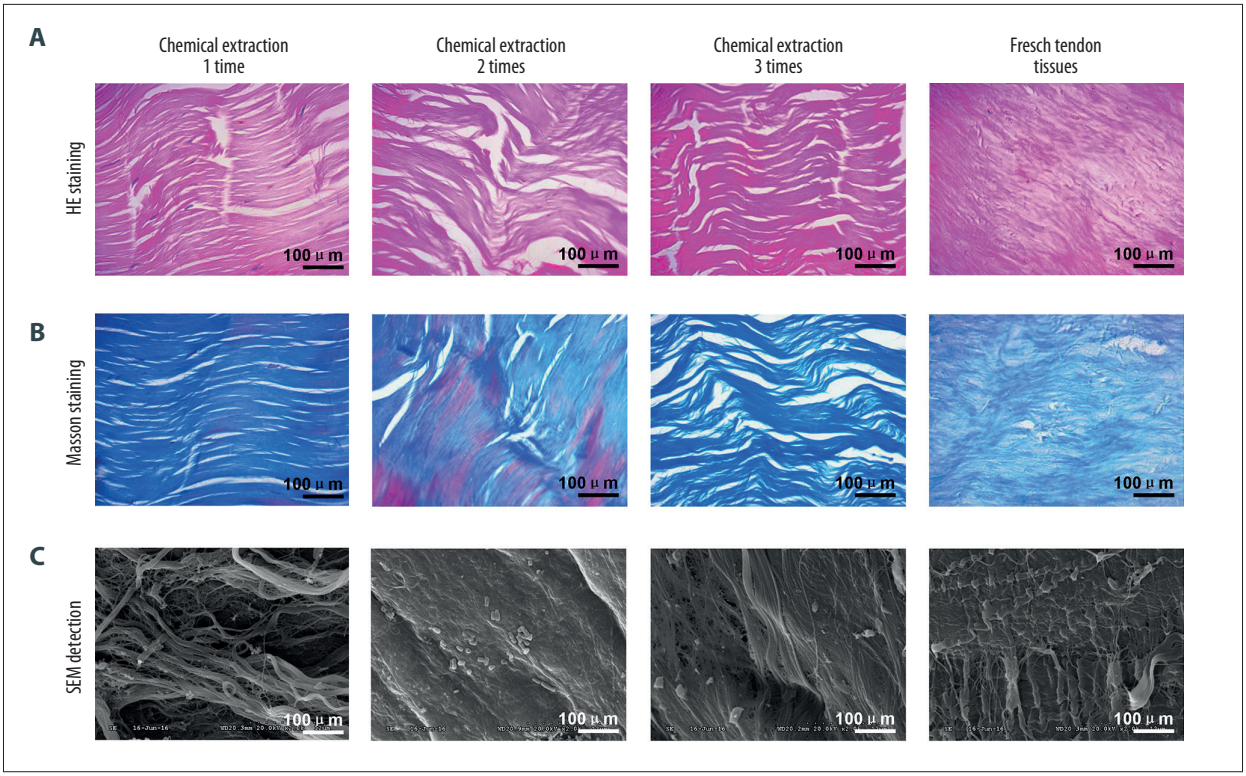

Figure 3. Identification of the allogenic tendons undergoing chemical extraction treatment by use of different staining methods. (**A**) HE staining. (**B**) Masson staining. (**C**) SEM.

showed that there were no inflammatory cells in Chemical extraction 3 group, but many inflammatory cells appeared in the fresh tendon tissues and in the Chemical extraction 1 group, and there were a few inflammatory cells in the Chemical extraction 2 group (Figure 3A). The Masson staining results indicated that there were some muscle fibers in the Chemical extraction 1, Chemical extraction 2, and fresh tendon tissues groups, but there were no muscle fibers in the Chemical extraction 3 group (Figure 3B).

SEM was also used to assess the antigenicity of the tendon tissues, showing there were many antigens in the tissues of the Chemical extraction 1, Chemical extraction 2, and fresh tendon tissues groups, and there were no antigens in the Chemical 3 extraction group except for the tendon tissues (Figure 3C).

The above results indicated the tendon tissues in the Chemical extraction 3 group were the best allogenic tendons for transplantation. Therefore, in the following experiments, the tendons in the Chemical extraction 3 group were used.

Isolation and identification of cartilage cells

According to the method described above in the Materials and Methods section, the cartilage cells were isolated. The results showed that the primary cartilage cells and passaged cells had typical cartilage cell morphology (Figure 4A). We also identified the cartilage cell-specific biomarker, collagen II, by using immunohistochemical analysis. The results indicated that collagen II was positively expressed in the isolated cartilage cell group (Figure 4B).

TGF-β1 treatment induces cartilage cell formation and triggers expression of acidic and neutral protein mucopolysaccharide

Masson staining was used to examine the formation of cartilage cells, and PSA staining and AB staining were used to assess levels of neutral protein mucopolysaccharide and acidic protein mucopolysaccharide, respectively. The results indicated that there were significantly more blue-stained cartilage cells in the 0.1 ng/mL TGF-β1 treatment group compared to the untreated and 0.2 ng/mL TGF-β1 treatment groups (Figure 5A). The PAS staining results showed a high level of neutral protein mucopolysaccharide in the 0.1 ng/mL TGF-β1 treatment group, but few neutral protein mucopolysaccharides were expressed in the untreated and 0.2 ng/mL TGF-β1 treatment groups (Figure 5B). AB staining also showed higher expression of acidic protein mucopolysaccharide in the 0.1 ng/mL TGF-β1 treatment group compared to the untreated and 0.2 ng/mL TGF-β1 treatment groups (Figure 5C).

Indexed in: [Science Citation Index Expanded] [Index Medicus/MEDLINE] [Chemical Abstracts] [Scopus]

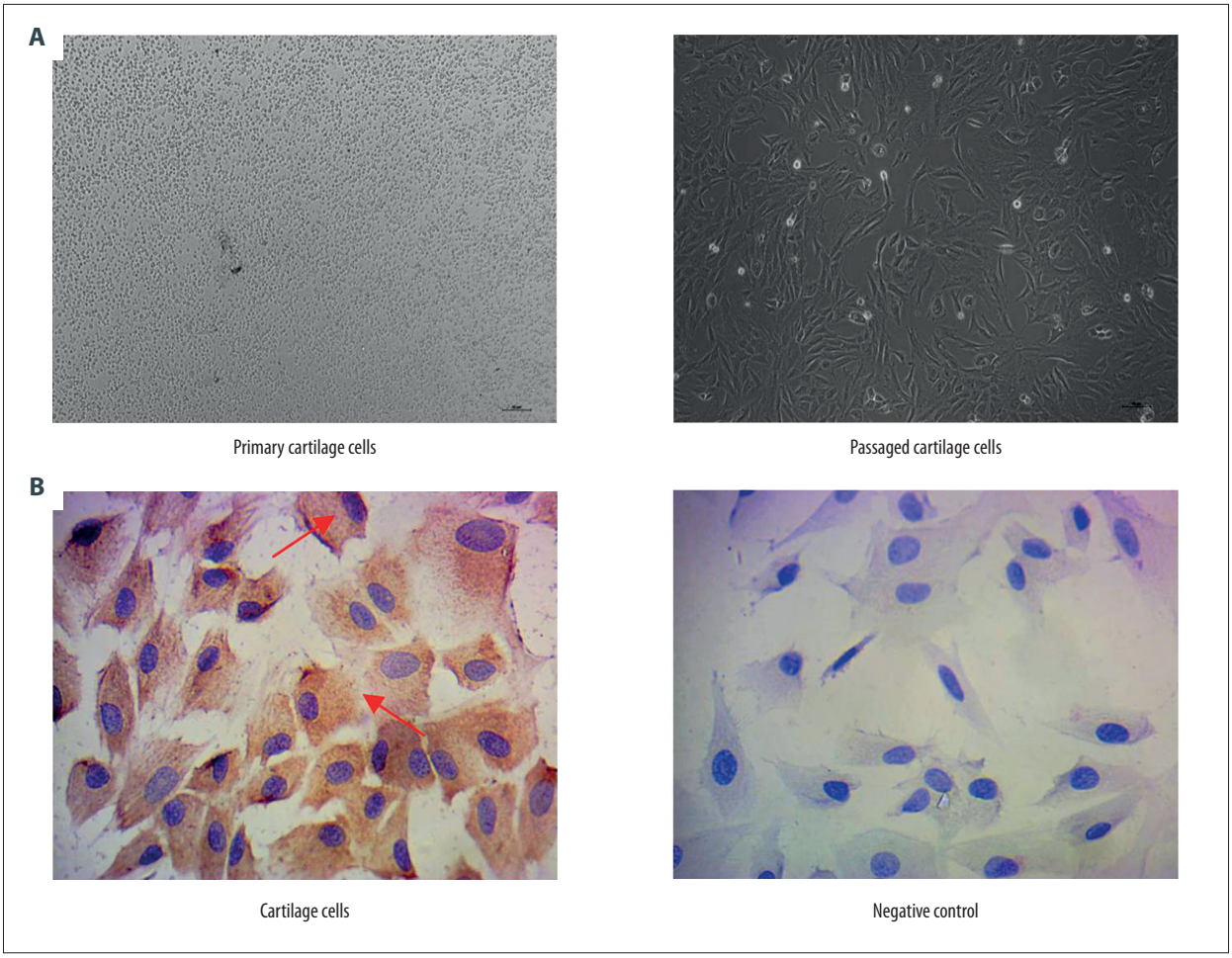

Figure 4. Observation for the cell growth and identification for cartilage cells. (**A**) Observation of cartilage cell growth. (**B**) Identification of cartilage cells by detecting the specific biomarker, collagen II.

Autologous cartilage cells combined with allogenic tendon transplanted tissues produced better growth of cartilage cells

To assess the growth conditions of the cartilage cells undergoing transplantation of autologous cartilage cells combined with allogenic tendon, HE staining, Masson staining, PAS staining, and AB staining were performed. The HE results indicated that there were no inflammatory cells in the autologous cartilage cells combined with allogenic tendon transplanted tissues, and only a few inflammatory cells in the suture only group and the simple allograft tendon transplantation group (Figure 6A). The Masson staining results showed that there were significantly more cartilage cells formed (blue stained cells) in the autologous cartilage cells combined with allogenic tendon transplantation group compared to the other groups (Figure 6B). The PAS staining results indicated there were many neutral protein mucopolysaccharides in the autologous cartilage cells combined with allogenic tendon group, but there were only a few neutral protein mucopolysaccharides in the other groups (Figure 6C). AB staining also showed higher levels of acidic protein mucopolysaccharide in the autologous cartilage cells combined with allogenic tendon transplantation group compared to the other groups (Figure 6D).

Discussion

The findings in this study support our experimental design that the autologous cartilage cells combined with allogenic tendon, when applied as an adjunct in the reconstruction for glenoid labrum or repair, significantly enhanced the levels of collagen organization, which was not observed previously by utilizing protein intervention, gene treatment, or traditional transplantation [16]. Although transplantation of tendons has been associated with adverse effects [17], there were no obvious evidence of this in our study. Tendon-to-bone treatment is the most challenging method for reconstruction of cartilaginous tissues or bone healing [9]. A previous study [18] indicated that tendon–bone integration of allograft tendons was not

Indexed in: [Science Citation Index Expanded] [Index Medicus/MEDLINE] [Chemical Abstracts] [Scopus]

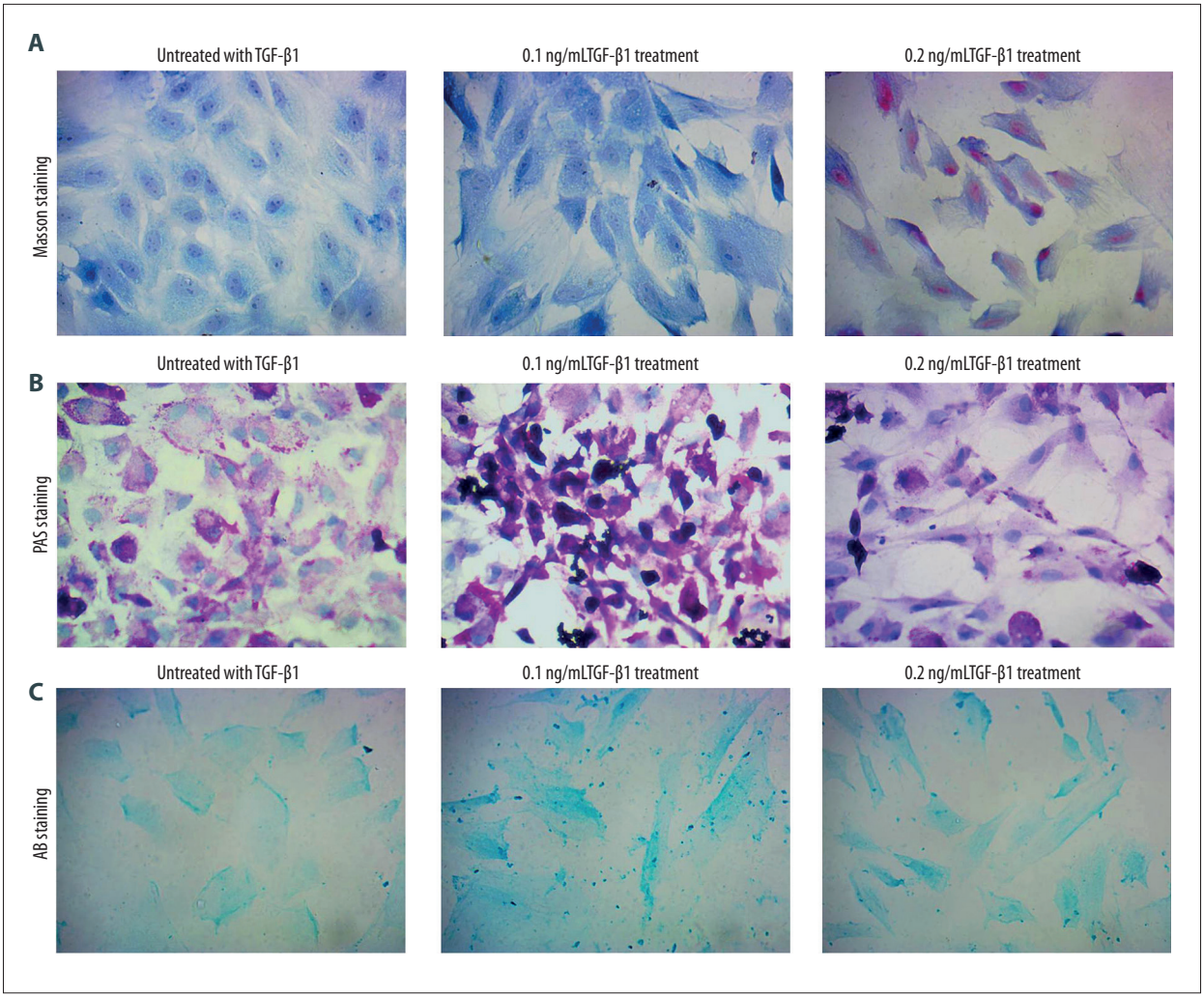

Figure 5. Evaluation of the effect of TGF-β1 treatment on inducing cartilage cell formation and acidic and neutral protein mucopolysaccharide expression. (**A**) Cartilage cell formation observation using Masson staining. (**B**) Acidic protein mucopolysaccharide observation using PAS staining. (**C**) Neutral protein mucopolysaccharide observation using AB staining.

achieved even 2 years after reconstruction. The improvement of the damaged glenoid labrum in this study suggests either the unique cellular qualities of the employed cartilage cells or a promising function of the improved allogenic tendon scaffold.

Chemical extraction has been widely used to obtain tendons in previous studies [19,20]. In this study, we used the chemical extraction method to extract the tendon tissues, and HE staining showed that there were no inflammatory cells in the tendons extracted from the Chemical extraction 3 group. The Masson staining results indicated that no muscle fibers were present in the Chemical extraction 3 group. SEM showed there were no antigens in the Chemical 3 extraction group except for the tendon tissues. Therefore, in the following experiments, we selected the Chemical 3 extraction group as the experimental tendons.

Cartilage cells have been used in glenoid labrum reconstruction in several studies [21,22], but the growth of cartilage cells was poor. In this study, to enhance cartilage cell growth, TGF-β1 was used to treat the cartilage cells. The results showed that the cartilage cells triggered cartilage growth. Previous studies [23,24] reported that acidic and neutral protein mucopolysaccharide expression reflects better growth potential; therefore, we examined the acidic and neutral protein mucopolysaccharide levels in TGF-β1-treated cartilage cells. The results indicated that 0.1 ng/mL TGF-β1-treated cartilage cells exhibited higher acidic and neutral protein mucopolysaccharide expression. Therefore, we applied the 0.1 ng/mL TGF-β1 to treat the cartilage cells in the following experiments.

After transplantation of autologous cartilage cells combined with allogenic tendon, the cartilage cells illustrated better growth, few inflammatory cells, and many neutral protein

Indexed in: [Science Citation Index Expanded] [Index Medicus/MEDLINE] [Chemical Abstracts] [Scopus]

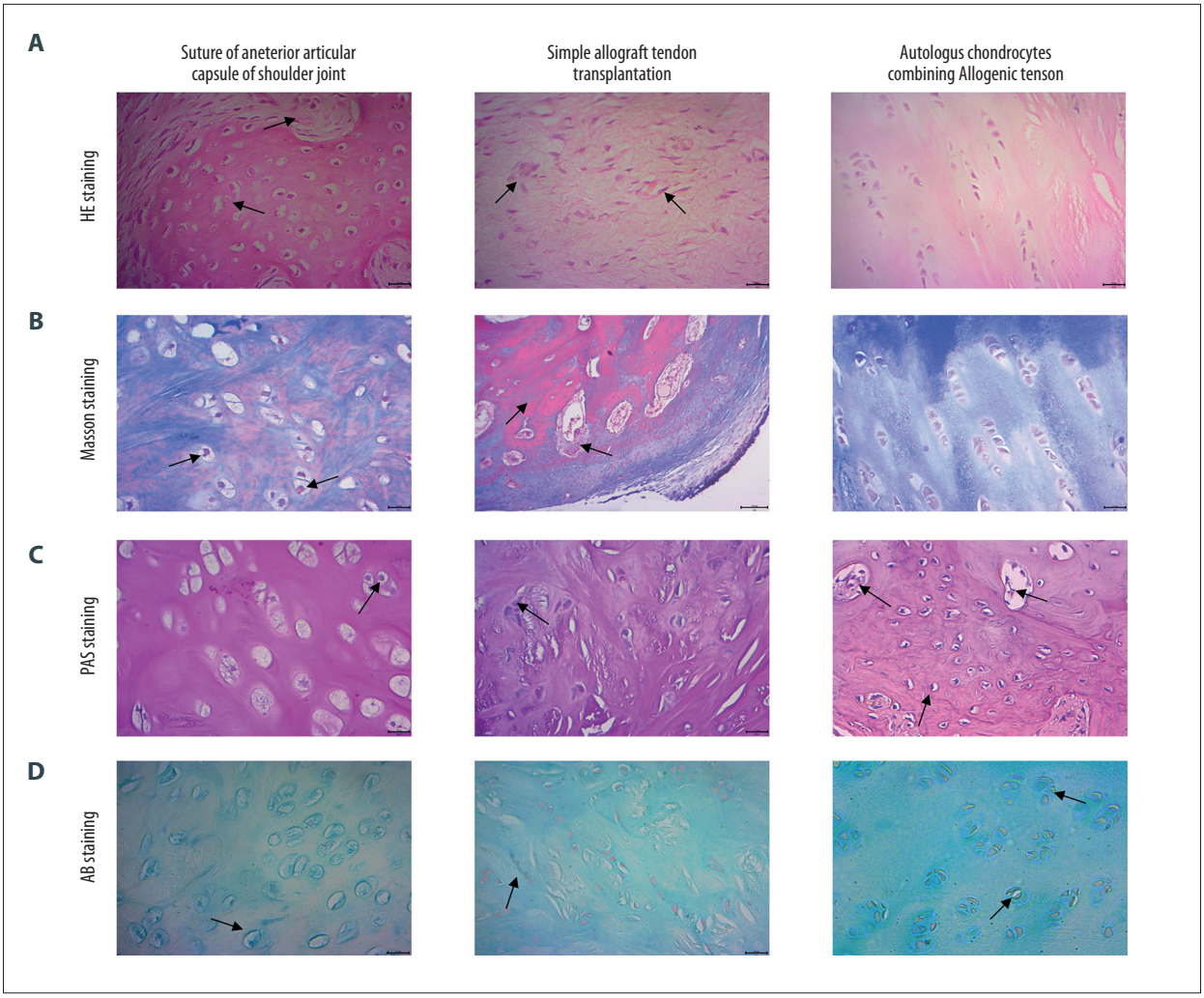

Figure 6. Cartilage cell growth in the autologous cartilage cells combined with allogenic tendon transplanted tissues. (**A**) HE staining to detect inflammatory cells. (**B**) Masson staining of muscle fibers. (**C**) Acidic protein mucopolysaccharide observation using PAS staining. (**D**) Neutral protein mucopolysaccharide observation using AB staining. The black arrows illustrate cells staining positive. The black arrows in **A** represent the inflammatory cells. The black arrows in **B** represent the cells staining Masson-positive. The black arrows in **C** represents the PAS staining positive cells. The black arrows in **D** represent the cells staining AB-positive.

mucopolysaccharides and acidic protein mucopolysaccharides, showing that the cartilage cells grew better in the transplanted glenoid labrum tissues.

Although previous studies [25–27] have used the cartilage tissue analogue to treat cartilage injury, the present study is the first to combine autologous cartilage cells and allogenic tendons.

The present study was only a preliminary investigation of allogenic tendon-autologous cartilage cell reconstruction, and we have not conducted more experiments for the specific diseases that can be induced in rabbits, which is a limitation of this study. We also did not perform quantitative detection for investigating effects of allogenic tendon-autologous cartilage cell reconstruction on glenoid labrum damage, which is another limitation. In the subsequent studies, we plan to perform quantitative detection and analysis to assess the efficacy of allogenic tendon with autologous cartilage cell reconstruction.

Conclusions

This is the first report on the effects of autologous cartilage cells combined with allogenic tendon on damaged glenoid labrum. This study established the allogenic tendon-autologous cartilage cells reconstruction approach and illustrated higher adhesive ability and growth ability, and better chondrogenesis in the glenoid labrum-damaged animal model.

Indexed in: [Science Citation Index Expanded] [Index Medicus/MEDLINE] [Chemical Abstracts] [Scopus]

Conflict of interests

None.

References:

1. Khosravi Shadmani F, Mansori K, Karami M et al: Avoidable burden of risk factors for serious road traffic crashes in Iran: A modeling study. J Prev Med Public Health, 2017; 50: 83–90
2. Shi Y, Hu Y, Lv G et al: Effects of reactive oxygen species on differentiation of bone marrow mesenchymal stem cells. Ann Transplant, 2016; 21: 695–700
3. Kane P, Bifano SM, Dodson CC et al: Approach to the treatment of primary anterior shoulder dislocation: A review. Phys Sportsmed, 2015; 43: 54–64
4. Snyder SJ, Karzel RP, Del Pizzo W et al: SLAP lesions of the shoulder. Arthroscopy. 1990; 6: 274–79
5. Provencher MT, Ghodadra N, Romeo AA: Arthroscopic management of anterior instability: Pearls, pitfalls, and lessons learned. Orthop Clin North Am, 2010; 41: 325–37
6. Mazzocca AD, Cote MP, Solovyova O et al: Traumatic shoulder instability involving anterior, inferior, and posterior labral injury: A prospective clinical evaluation of arthroscopic repair of 270 degrees labral tears. Am J Sports Med, 2011; 39: 1687–96
7. Paulos LE, Evans IK, Pinkowski JL: Anterior and anterior-inferior shoulder instability: Treatment by glenoid labrum reconstruction and a modified capsular shift procedure. J Shoulder Elbow Surg, 1993; 2: 305–13
8. Bateman E, Donald SM: Reconstruction of massive uncontained glenoid defects using a combined autograft-allograft construct with reverse shoulder arthroplasty: Preliminary results. J Shoulder Elbow Surg, 2012; 21: 925–34
9. Qu J, van Alphen NA, Thoreson AR et al: Effects of trypsinization and mineralization on intrasynovial tendon allograft healing to bone. J Orthop Res, 2015; 33: 468–74
10. Ding Q, Zhong H, Qi Y et al: Anti-arthritic effects of crocin in interleukin-1beta-treated articular chondrocytes and cartilage in a rabbit osteoarthritic model. Inflamm Res, 2013; 62: 17–25
11. Abe H, Itoi E, Yamamoto N et al: Healing processes of the glenoid labral lesion in a rabbit model of shoulder dislocation. Tohoku J Exp Med, 2012; 228: 103–8
12. Ozeki N, Muneta T, Koga H et al: Transplantation of Achilles tendon treated with bone morphogenetic protein 7 promotes meniscus regeneration in a rat model of massive meniscal defect. Arthritis Rheum, 2013; 65: 2876–86
13. Wang R, Xu B, Xu HG: Up-Regulation of TGF-beta promotes tendon-to-bone healing after anterior cruciate ligament reconstruction using bone marrow-derived mesenchymal stem cells through the TGF-beta/MAPK signaling pathway in a New Zealand white rabbit model. Cell Physiol Biochem, 2017; 41: 213–26
14. Sun HB, Schaniel C, Leong DJ et al: Biology and mechano-response of tendon cells: Progress overview and perspectives. J Orthop Res, 2015; 33: 785–92
15. Horn MM, Martins VC, Plepis AM: Influence of collagen addition on the thermal and morphological properties of chitosan/xanthan hydrogels. Int J Biol Macromol, 2015; 80: 225–30
16. Guadilla J, Fiz N, Andia I et al: Arthroscopic management and platelet-rich plasma therapy for avascular necrosis of the hip. Knee Surg Sports Traumatol Arthrosc, 2012; 20: 393–98
17. Llombart Blanco R, Valenti A, Diaz de Rada P et al: Reconstruction of the extensor mechanism with fresh-frozen tendon allograft in total knee arthroplasty. Knee Surg Sports Traumatol Arthrosc, 2014; 22: 2771–75
18. Malinin TI, Levitt RL, Bashore C et al: A study of retrieved allografts used to replace anterior cruciate ligaments. Arthroscopy, 2002; 18: 163–70
19. Cartmell JS, Dunn MG: Effect of chemical treatments on tendon cellularity and mechanical properties. J Biomed Mater Res, 2000; 49: 134–40
20. Mohamad NA, Mustafa S, El Sheikha AF et al: Modification of gelatin-DNA interaction for optimised DNA extraction from gelatin and gelatin capsule. J Sci Food Agric, 2016; 96: 2344–51
21. Elser F, Braun S, Dewing CB et al: Glenohumeral joint preservation: Current options for managing articular cartilage lesions in young, active patients. Arthroscopy, 2010; 26: 685–96
22. Thomas SJ, Reuther KE, Tucker JJ et al: Biceps detachment decreases joint damage in a rotator cuff tear rat model. Clin Orthop Relat Res, 2014; 472: 2404–12
23. Harada Y, Garenaux E, Nagatsuka T et al: Interaction of 70-kDa heat shock protein with glycosaminoglycans and acidic glycopolymers. Biochem Biophys Res Commun, 2014; 453: 229–34
24. Battaglia MJ, Cordasco FA, Hannafin JA et al: Results of revision anterior cruciate ligament surgery. Am J Sports Med, 2007; 35: 2057–66
25. Lee SU, Lee JY, Joo SY et al: Transplantation of a scaffold-free cartilage tissue analogue for the treatment of physeal cartilage injury of the proximal tibia in rabbits. Yonsei Med J, 2016; 57: 441–48
26. Chiang ER, Ma HL, Wang JP et al: Allogeneic mesenchymal stem cells in combination with hyaluronic acid for the treatment of osteoarthritis in rabbits. PLoS One, 2016; 11: e0149835
27. Cheng F, Yang Z, Zeng J et al: Anesthesia management of modified *ex vivo* liver resection and autotransplantation. Ann Transplant, 2018; 23: 274–84

Indexed in: [Science Citation Index Expanded] [Index Medicus/MEDLINE] [Chemical Abstracts] [Scopus]