Case Report

Autologous Chondrocyte Implantation for Cartilage Injury Treatment in Chiang Mai University Hospital: A Case Report

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Autologous chondrocyte implantation (ACI) has become one of the standard procedures for articular cartilage defect treatment. This technique provides a promising result. However, the procedural process requires an approach of several steps from multidisciplinary teams. Although the success of this procedure has been reported from Srinakharinvirot University since 2007, the application of ACI is still limited in Thailand due to the complexity of processes and stringent quality control. This report is to present the first case of the cartilage defect treatment using the first generation-ACI under Chiang Mai University's (CMU) own facility and Ethics Committee. This paper also reviews the process of biotechnology procedures, patient selection, surgical, and rehabilitation techniques. The success of the first case is an important milestone for the further development of the CMU Human Translational Research Laboratory in near future.

Keywords: *Autologous chondrocyte implantation, Cartilage defect, Treatment*

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Cartilage injury in the knee joint often results from traumatic events. The treatment has been a formidable challenge because cartilage tissue was incapable of quality repair and regeneration. Several surgical concepts have been described such as microfracture and osteochondral transplantation. Autologous chondrocyte implantation has become one of the promising surgical techniques providing repair for the hyaline cartilage that can be applied for a larger sized defect[1]. This technique has been described since 1994 by Brittberg et al[2]. The first case of ACI was reported by Chanlalit et al from Srinakharinvirot University since 2007[3], with subsequent cases being reported from the same institute[4]. The remarkable limitations are the requirement of good scientific support and the high quality control of the human laboratory. These restrictions continue to make this surgical procedure difficult to apply in Thailand.

Herein, the first case of chondrocyte implantation was performed in Chiang Mai University Hospital under the monitoring of the Ethics Committee of the Faculty of Medicine. The success of this first case is an important milestone for the further development of this surgical concept while improving the CMU Human Translational Research Laboratory.

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Case Report

A 43-year-old woman presented with right knee pain during walking and climbing up stairs. She had a knee injury from a fall two years ago. Her physical examination revealed crepitation on motion and a point of tenderness on the medial condyle of her right knee. The anterior and posterior drawer signs were negative. The radiographs of her right knee on the lateral and anteroposterior standing views revealed a normal bony alignment and normal joint space. Magnetic resonance imaging (MRI) showed an area of cartilaginous defect in the central weight bearing

area of the medial femoral condyle and a partial tear of the meniscus.

Arthroscopic examination and partial menisectomy were performed. Full thickness articular fibrillation was founded at the medial femoral condyle of 1.5x2 cm^2 in size (Fig. 1A). Debridement was performed to the fibrillated cartilage until exposure to the subchondral bone. Hyaline cartilage at the lateral edge of the intercondylar notch was harvested using a graft harvesting gouge (Fig. 1B). The fresh cartilage was then preserved in sterile transport medium and immediately transferred to the cell culture laboratory. Six weeks after the initial operation, the second operation was performed for ACI. A medial arthrotomy was performed in this case. After preparing the full thickness cartilage lesion, the proximal tibial periosteal flap was harvested and sutured securely to cover the lesion by using 6/0 vicryl, and then 0.8 mL of $1x10^6$ cell/mL chondrocytes in autologous solution were implanted. Postoperatively, a full range of motion exercise of the knee and quadriceps muscle was encouraged. The post-operative MRIs at three, and six months demonstrated an intact graft (Fig. 2). The patient was allowed to walk with toe touch weight bearing during the first three months, then protected weight bearing until the end of the sixth month. This patient will be followed until two years after transplantation before resuming normal activity.

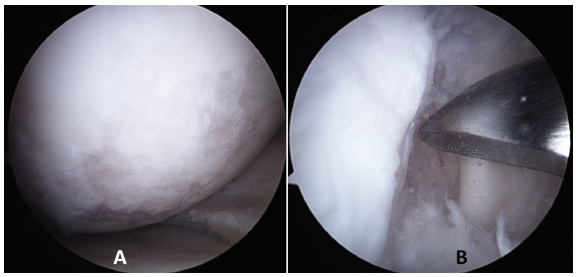

Fig. 1 Arthroscopic examination showing cartilage fibrillation (A), and full thickness lesion after debridement (B).

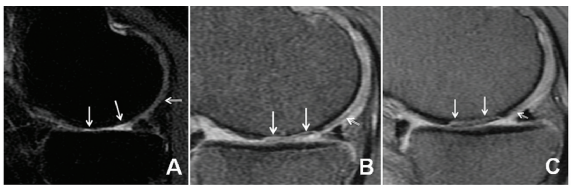

Fig. 2 Pre-operative sagittal MRI of the knee showing a cartilage defect (A), three months post-operative view of the the thin layer of graft (B), and six months after the cartilage graft which completely filled the defect resulting in the smooth articular surface between the graft and normal cartilage (C), a cartilage defect in the central weight-bearing area of medial femoral condyle (arrows) and normal cartilage (open arrow).

Patient selection for ACI

Autologous chondrocyte implantation requires a relatively unlimited size of a cartilage lesion. On the other hand, crucial concerns include determining an age with a high quality of chondrocyte, intraarticular environment and the depth of lesion, and post-operative co-operation in rehabilitation. A suitable patient meeting the recommended best selection for ACI criteria includes[5,6]

1. Age of 15 to 55 years.
2. BMI less than or equal 35 kg/m^2.
3. Persistent symptoms of disabling localized knee pain for at least six months, had have failed to respond to conservative treatment as well as established surgical intervention (microfracture, drilling, abrasion arthroplasty).
4. The condition involves a focal, full thickness, (grade III or IV) isolated defect of the knee involving the weight-bearing surface of the medial or lateral femoral condyles or trochlear region caused by acute or repetitive trauma.
5. The lesion is single and unipolar (involving only one side of the joint), and is contained with near normal surrounding articular cartilage.
6. The defect involves not more than six mm of the cartilage and the subchondral bone.
7. The total area of the cartilage defect is between 1.5 to 10 cm^2.
8. The patient is willing and able to comply with post-operative rehabilitation and weight-bearing restrictions.

Exclusion criteria for ACI

1. Active infection, inflammation, or osteoarthritis.
2. Uncorrected meniscus injury, unstable knee due to ligament injury and/or malalignment.

Patient selection begins with obtaining a careful history, which includes onset of symptoms, mechanism of injury, prior treatment, and response to previous treatment. A physical examination should carefully be obtained for a meniscus injury, the mechanical alignment of the lower extremity and a ligamentous stability of the knee joint, because

they are contraindications for ACI. A radiographic evaluation should include standing AP and lateral views, patella skyline view (Merchant). The PA 45º flexion view is made in case of a suspected posterior defect of the femoral condyle. Moreover, recent advancements in cartilage-specific MRI such as high-resolution fast spin echo sequence techniques can determine the location, size, and depth of cartilage lesions.

Surgical techniques of ACI

The first generation of ACI is a two-stage procedure and utilizes a biologic technique. The first step is an arthroscopic harvest of normal hyaline cartilage, which is cultured and expanded to obtain an adequate number of chondrocytes. The second step is an implantation beneath an autologous periosteal patch.

The first arthroscopic exam is performed in order to investigate the extent of the lesion. Grade III and IV cartilage lesions will be debrided until the exposure of the depth of the subchondral bone as well as the surrounding normal articular cartilage. Meanwhile correction of any concomitant meniscal injury and joint debridement will be performed. Two to-three hundred milligrams of normal cartilage is harvested in this step. The intercondylar notch at the superomedial/lateral femoral condylar edge is considered a suitable area for cartilage harvesting. The harvested cartilage is placed directly in sterile transport medium (Dulbecco's modified eagle medium) and transferred for the culturing process. The primary chondrocyte extraction and culture process usually will take four to six months to cultivate an adequate number of chondrocytes. Meanwhile the patient will be following a rehabilitation program with quadriceps training exercises.

Autologous chondrocyte preparation will be performed under quality control in terms of three different properties including the number of cells, sterility test, and differentiation potential. Five to twenty million cells provide a suitable number for a 1x1 cm^2 lesion[2]. The weekly reports of culture results for bacteria, fungus and polymerase chain reaction (PCR) of mycoplasma must all be negative. The differentiation potential of the chondrocytes will be determined by RT-PCR for Col-I, Aggregan and Sox-9 gene expression. Autologous chondrocytes will be dissolved in autologous serum and transferred under sterile technique to the operating room during the second operation.

For the second operation, a midline skin incision and medial/lateral parapatellar arthrotomy will be performed depending on the site of the lesion (Fig. 3A). The fat pad and soft tissue surrounding the patellar ligament is always retained to avoid postoperative pain and stiffness. A Hohmann retractor is placed in the intercondylar notch to help displace the patella to the contralateral side for assisting the surgical exposure. A small curette and scalpel blade will be used to incise and prepare the defect until the level of the subcondral bone is exposed. Debridement is also performed until the healthy and stable vertical shoulder is presented (Fig. 3B).

The vertical incision is made on the medial aspect of the proximal tibia, just distal to the insertion of the pes-anserine tendon (Fig. 3C). The periosteum is exposed, and a periosteal patch is outlined on the basis of two mm larger than the defect allowing slight shrinkage after detachment. The patch edge is outlined with a scalpel blade and elevated with a periosteal elevator, then blunt dissection is performed (Fig. 3D and E). After the patch is detached, the outer surface should be noted to distinguish it from the inner cambium layer. A periosteal patch is placed over the defect with the cambium layer facing the defect base, and then the patch is secured onto the cartilage rim with a 6-0 absorbable suture (Fig. 4A). The suture is placed first through the patch and then through the surrounding articular cartilage, leaving an opening at the most superior corner for chondrocyte implantation. The edges of the patch are sealed with fibrin glue (Fig. 4B), and the leakage is tested by slowly injecting saline solution into the covered defect. After waterproof testing, the saline solution is aspirated to prepare the

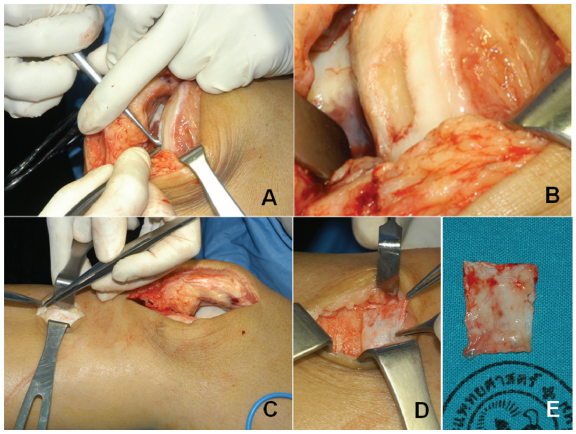

Fig. 3 Surgical technique showing cartilage defect preparation (A, B), and periosteal harvesting (C-E).

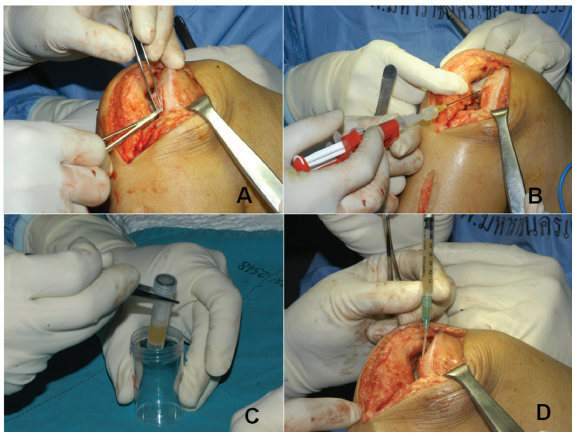

Fig. 4 Surgical technique demonstrating periosteal grafting (A, B), and the chondrocyte implantation technique (C, D).

defect for the injection of the chondrocyte suspension aspirated from the transport vials through an 18-gauge angiocatheter (Fig. 4C). The angiocatheter is placed via an opening at the top and advanced to the distal end of the defect, and the chondrocyte suspension is slowly injected while the catheter is slowly withdrawn until the defect is filled (Fig. 4D). The opening is finally closed with additional sutures and sealed with fibrin glue.

Rehabilitation

Post-operative rehabilitation is the important factor that contributes to the success of neocartilage formation and maturation. The proliferation of chondrocytes progenies lasts up to six weeks. The tissue usually has firmed up by three to six months and the neocartilage is putty-like within six to nine months. The maturation process will take two to three years. The goal of rehabilitation is composed of three main steps including progressive weight bearing, restoration of range of motion (ROM) and enhancement of muscle strengthening. A good rehabilitation program will protect the soft gelatinous-like tissue during the initial period and enhance the maturation process in the later steps[7].

For the first months, continuous passive motion of the affected joint is encouraged. An active range of motion or bicycle without resistance is recommended in the second and third months. Toe touch weight bearing during ambulation is recommended in the first three months. Increasing active ROM exercise with minimal resistance in the unrepaired zone and progressive weight bearing ambulation are recommended during the three to six month period. Progressive resistive exercise in full ROM and unrestricted activities of daily living are encouraged after six months. Light sports activity will be encouraged after one year, and unrestricted contact sports activity will be allowed after two years.

Acknowledgement

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Potential conflicts of interest

None.

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การปลูกถ่ายเซลล์กระดูกอ่อนเพื่อรักษาภาวะกระดูกอ่อนบาดเจ็บ: รายงานผู้ป่วย

ประสิทธิ์ วงศ์ตรีรัตนชัย, ดำเนินสันต์ พฤกษากร, พีรพรรณ โปธาเจริญ, ภูวพงศ์ นิ่มกิ่งรัตน์, นัทธยา ปัทมภาสพงษ์, ชนะการ พรพัฒน์กุล, ศศิวริยา เศรษฐสิทธากุล, ลัดดา ฟองสถิตย์กุล, สุรีพร พรหมแพทย์

การปลูกถ่ายเซลล์กระดูกอ่อนเป็นวิธีผ่าตัดเพื่อรักษาภาวะกระดูกอ่อนบาดเจ็บ โดยเป็นที่ยอมรับในระดับสากลว่าให้ผลการรักษาที่ดี แต่ในขณะเดียวกันวิธีการเตรียมเซลล์และขั้นตอนการผ่าตัดต้องอาศัยวิธีการที่ซับซ้อนและอาศัยบุคลากรจากสหสาขาวิชา สำหรับประเทศไทยนั้นเคยมีรายงานการผ่าตัดด้วยวิธีดังกล่าวครั้งแรกในปี พ.ศ. 2550 โดยคณะแพทย์จากมหาวิทยาลัยศรีนครินทรวิโรฒ แต่การผ่าตัดด้วยวิธีดังกล่าวยังคงไม่สามารถทำได้อย่างแพร่หลายในประเทศไทยเพราะความซับซ้อนของวิธีการและขั้นตอนการควบคุมคุณภาพ ดังนั้นรายงานนี้จึงเป็นการรายงานผู้ป่วยรายแรกที่ทำการรักษาด้วยวิธีการปลูกถ่ายเซลล์กระดูกอ่อนภายใต้ศักยภาพ และบุคลากรของคณะแพทยศาสตร์ มหาวิทยาลัยเชียงใหม่ และภายใต้การควบคุมดูแลของคณะกรรมการจริยธรรมการวิจัยของมหาวิทยาลัย ทั้งนี้ในรายงานยังบรรยายขั้นตอนของวิธีการในห้องเพาะเลี้ยงเซลล์ การคัดเลือกผู้ป่วย วิธีการผ่าตัด และการทำกายภาพบำบัด ความสำเร็จของการผ่าตัดด้วยวิธีดังกล่าวถือว่าเป็นก้าวที่สำคัญสำหรับการถ่ายทอดเทคโนโลยีชีวภาพสู่การใช้ประโยชน์ทางคลินิก ซึ่งจะยังคงต้องพัฒนาให้ดีขึ้นอีกในอนาคตอันใกล้