Clinical Guidelines for using Regenalase™ Laser System (Optional: Combination with Injectable Biologics) for Treatment of Cartilage Defects in Knee Joints in an Office or Outpatient Setting

Disclaimer: In the event of any discrepancies between this Clinical Guide and the Regenalase™ Instructions for Use (IFU), the information contained in the IFU shall take precedence.

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1. Preliminary Diagnosis and Patient Selection

Indications for laser cartilage reconstruction are traumatic injuries to the hyaline cartilage of the knee joints, as well as degenerative diseases with severity of cartilage defects noted on an MRI image no higher than grade 3 according to the Outerbridge classification, and not amenable to conservative drug treatment. Example: Osteoarthritis.

2. Inclusion Criteria

1. Men and women aged 18 years and over at the time of the procedure;
2. Presence of no more than 2 zones of chondromalacia of cartilage of grades 1, 2 or 3 of the knee joints according to the Outerbridge classification (Fig. 1) and Osteoarthritis (OA) of stage 0, 1 or 2 according to the Kellgren and Lawrence (KL) classification based on MRI of the knee joint. (Figs. 2,3);
3. Chronic pain syndrome (duration of at least 3 months with Visual Analog Score (VAS) score ≥ 3). Note: VAS is a visual unidimensional scale of pain, with “no pain” equal to 0 and with “worse imaginable pain” equal to 10.

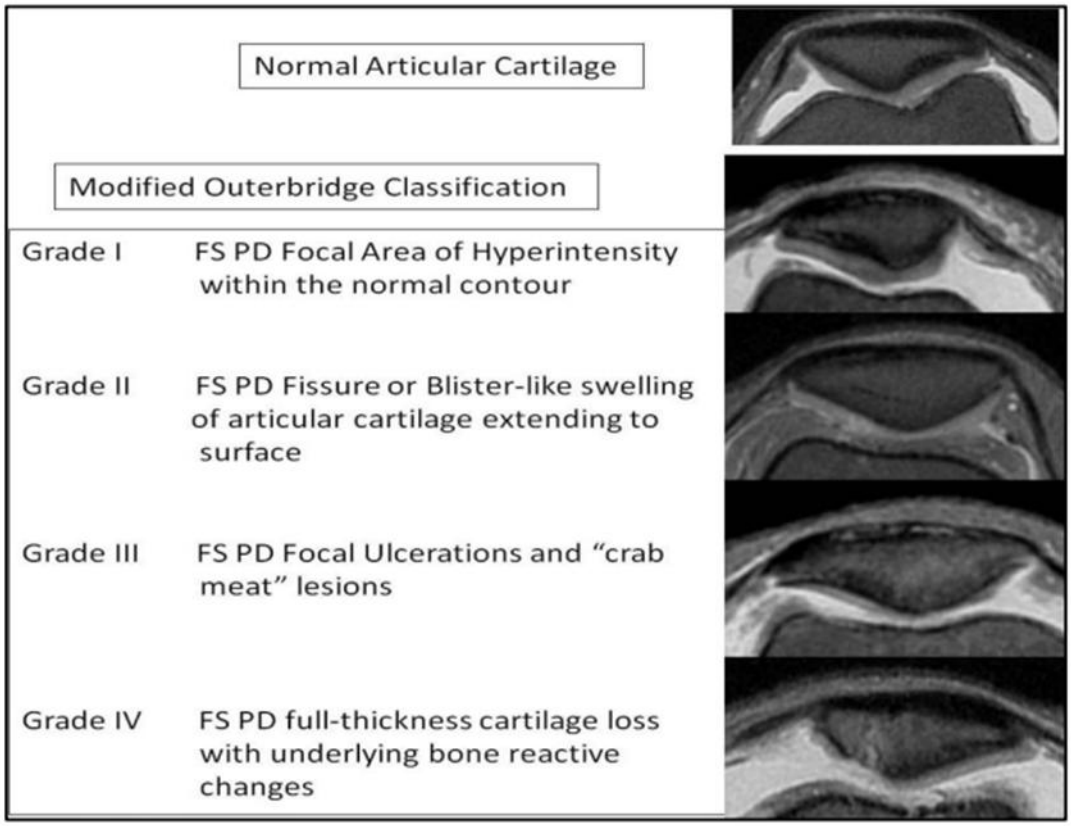
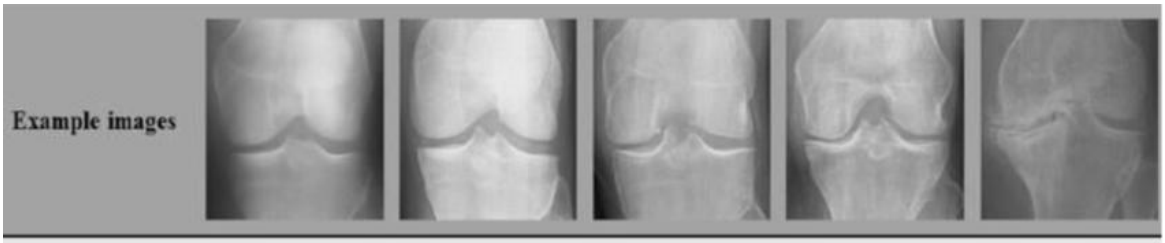


Fig. 1. Modified Grading of Cartilage Injuries, Outerbridge Classification.



Example images					
KL Grade	**Grade0**	**Grade1**	**Grade2**	**Grade3**	**Grade4**
Classification	**Normal**	**Doubtful**	**Mild**	**Moderate**	**Severe**
Description	No radiographic features of OA	Doubtful joint space narrowing(JSN)	Definite osteophytes Possible JSN	Multiple osteophytes Definite JSN Scierosis	Large osteophytes Marked JSN Severe scierosis

Fig. 2. Radiographic classification of OA by Kellgren and Lawrence.

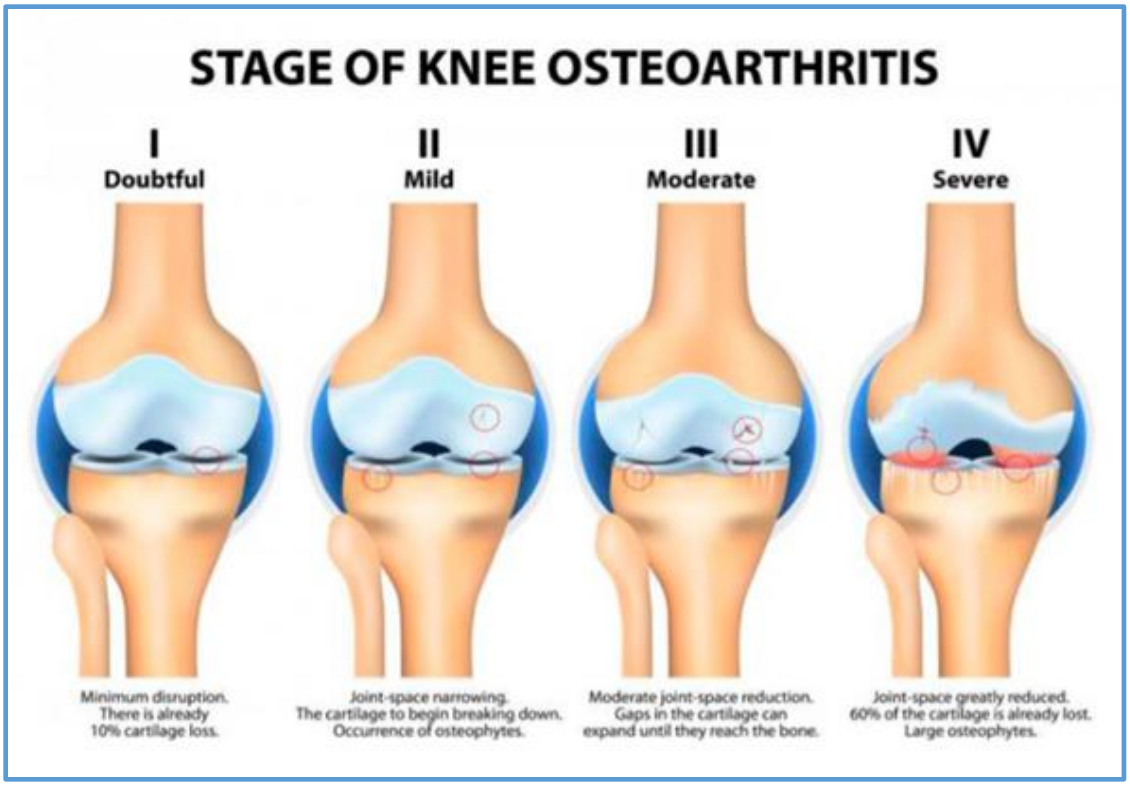


Fig. 3. Schematic representation of OA classification.

Laser treatment of cartilage is likely ineffective for patients with any of the following conditions: osteoarthritis stage III or IV (Kellgren and Lawrence classification), chondromalacia stage IV (Outerbridge classification), or severe valgus and varus deformities of the lower extremities.

3. Exclusion Criteria

1. Fibrous ankylosis, presence of contracture of the knee joint;
2. Presence of a local or systemic inflammatory process;
3. Purulent-inflammatory processes of the skin in the area of surgical intervention;
4. Acute period of knee joint injury;
5. Diseases of the blood coagulation system;
6. Women pregnant or lactating at the time of the procedure.

Please consult the Regenalase™ Instructions for Use (IFU) for a complete list of contraindications.

4. Description of Procedures

[Formulation of treatment plan]

This Clinical Guide describes surgical procedures in a physician's office or on an outpatient care basis.

The surgical laser procedure may be followed by noninvasive irradiation of the affected area with the 980 nm wavelength laser as an adjunctive treatment. This radiation is applied topically through the skin.

The procedure may also be followed by injections of biologics. Physicians can choose their preferred method to improve the biologics' performance by pre-treatment with the Regenalase™ system. For example, plasma rich platelets (PRP) may be irradiated by the 980 nm wavelength laser before the injection.

[Additional diagnostic steps]

1. Sample Testing (blood and urine)
   - general clinical blood and urine tests;
   - determination of blood type and Rh factor;
   - coagulogram;
   - biochemical blood tests;
   - screening for latent infections.
2. Magnetic resonance imaging (MRI) with a power of at least 1.5 T;
3. X-ray (2 projections);
4. Lower Extremity Topograms;
5. Ultrasound examination.

5. Treatment Procedure

[Acceptable visualization modalities]

The procedure is performed using either a NanoScope (or similar imager) or an ultrasound imager for visualization of the treatment area.

[Acceptable anesthesia/antibiotic prophylaxis methods]

Laser treatment of knee joint cartilage may be performed under local anesthesia. Antibiotics are administered for patients at risk of infection. For most percutaneous procedures, antibiotic prophylaxis is not needed but may be given orally the day of and the day after the procedure.

[Laser safety]

Personnel, patients, and all those in areas with a possibility of exposure to laser radiation from a Class 4 laser device must wear eyeglasses that are appropriate for the laser wavelength and output power. Consult the IFU for details.

[Patient preparation]

The patient is typically supine with the knee flex at 90 to 100 degrees depending on lesion location. They may also sit up leaning against the back of a table or wall if they wish to observe the procedure. If the patella or trochlear groove is targeted then full extension with a lateral firing needle/fiber is recommended. The affected knee is preliminarily prepared with alcohol and chlorhexidine.

[Laser and Fiber-Needle Preparation, Standard Case]

A fiber with 400 μm diameter and a sterile 18 gauge needle should be selected for the procedure. The assistant outside the sterile field powers on the Regenalase™ system, enters the passcode per the IFU, selects ‘Surgery’ and then ‘Standard’ on the Regenalase™ screens (Figure 4.1). The assistant then selects the ‘Low Energy’ preset followed by ‘Straight 400 mm fiber’ and enters the fiber’s alphanumeric serial number shown on the sterile package (Figure 4.2).

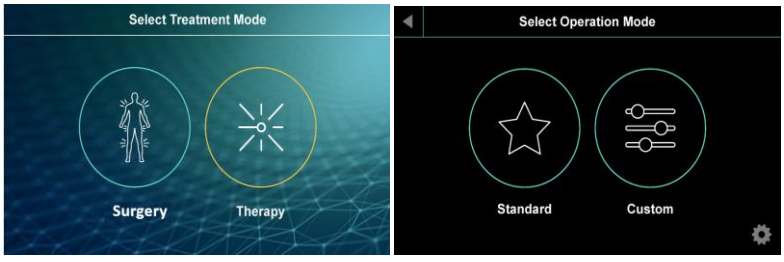


Fig. 4.1. Treatment and Operation Mode Screens.

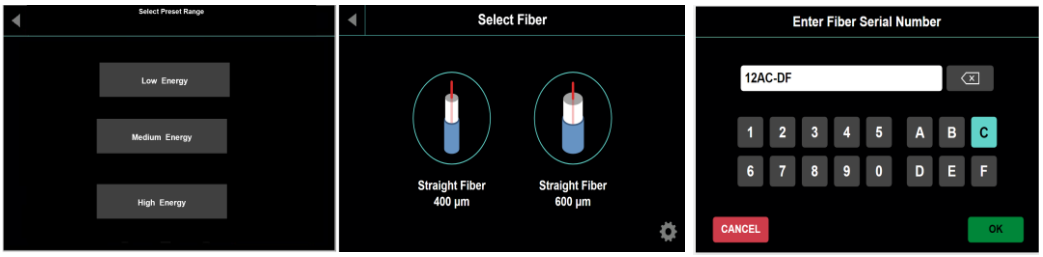


Fig. 4.2. Preset, Fiber Selection and Fiber Serial Number Screens.

The assistant then opens the package containing the sterile fiber and presents it to the doctor in the sterile field who extracts the inner sterile package. The doctor opens the inner sterile package, carefully removes the fiber from the retaining cardboard backing and gives the connector end back to the assistant. While the doctor performs the following steps, the assistant connects the fiber to Port B of the system and then waits for the doctor to be ready for fiber calibration before pressing 'OK' on the Fiber Calibration screen (Figure 5.1).

Fig. 5.1 Fiber Calibration Screen.

The doctor must feed the fiber distal end into the 18 ga needle ensuring that the fiber tip extends approximately 1 mm beyond the tip of the needle after tightening the Luer lock to secure the fiber to the puncture needle. To ensure the fiber tip is not damaged, the doctor points the fiber tip to a nonreflecting, white surface at a distance of at least 2 cm and should see a well-defined, round green spot when the calibration procedure begins. An irregularly, dispersed shaped spot indicates fiber is damaged and should be discarded (Fig. 5.2). The assistant selects 'OK' on the Fiber Calibration screen to begin the calibration procedure.

Well-defined circle indicates high quality fiber	Dispersed pattern indicates lower quality or broken fiber
	

Fig. 5.2 Laser B beam shape evaluation

After the calibration passes the Standby screen (left side Fig. 6) opens. The assistant ensures that 'High' Power and 'Pulse Train' are both highlighted and awaits the doctor's request to begin the procedure before pressing 'Start Procedure' to enter Ready Mode (right side Fig. 6).

Fig.6. Standby Mode screen (left) and Ready Mode screen (right).

The preset parameters for each spot treatment are as follows:

- Peak pulse power 1 W, pulse width 0.1 sec, interval between pulses 0.9 sec
- Pulse repetition rate 1 Hz (one per second)
- Pulse series consists of 10 pulses (= 10 secs duration)
- Interval between Pulse series is 10 secs (no pulses occur)
- Total time for one spot laser treatment is 50 sec (3 Pulse series + 2 intervals)

[Establishing laser fiber access to the joint]

Ultrasound Visualization Tool:

Prior MRI images are useful to diagnose and determine the area of the chondromalacia and should be displayed in the treatment room to reference during the procedure. After verification of the area of chondromalacia, a 25 gauge, 1.5 inch needle or a 25 gauge, 2 inch needle may be used to administer local anesthesia and for localization of the lesion. Once the needle tip is in place over the lesion, a number 11 scalpel blade may be used for a co-located stab incision oriented to allow for easier maneuverability of the 18 gauge needle during treatment of larger lesions. Remove the 25 gauge needle and insert the 18 gauge needle with the laser fiber into the established access port.

Note that only a single access port is needed for the ultrasound-guided laser procedure. Advance the 18 gauge introducer puncture needle onto the target area of the cartilage and proceed with laser treatment as described below.

NanoScope Visualization Tool:

A NanoScope field of vision is displayed in Fig. 7 showing the chondromalacia and the distal end tip of the cannula containing the fiber at the target spot to be treated. Note that the laser system's green aiming light is on.

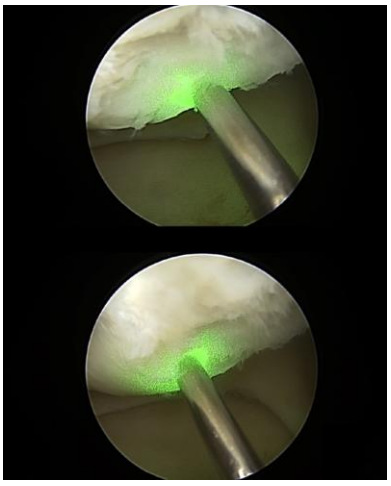


Fig. 7. Demonstration of laser irradiation of the chondromalacia area

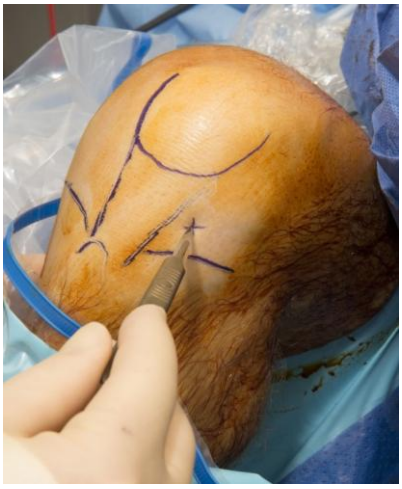


Fig. 8. Formation of the access ports (accesses) for NanoScope.

The desired portal site (standard anterolateral or anteromedial portal) is identified (Fig. 8), and a 25-gauge needle is used to inject approximately 7.5 cc of a mixture of local anesthetic, including 1% lidocaine with epinephrine or 0.5% Marcaine without epinephrine at the capsule only, creating a wheel. The epinephrine as a vasoconstrictor is helpful in improving hemostasis at the portal site(s). Both portal sites can be anesthetized if the surgeon anticipates using both portals during the procedure (i.e., one for viewing and the other for treatment). However, if using for diagnostic purposes only, it is preferable to anesthetize only one portal site.

A sterile field adjacent to the examination table with either a Mayo stand or a side table is prepared with sterile, disposable drapes. The skin over the anterior aspect of the knee, from the superior pole of the patella to the region of the distal patellar tendon is prepped with chlorhexidine, and drapes are placed along the superior and inferior borders of this sterile field, creating a working space centered over the patella. Next, using an 18- or 20-gauge needle, ~20 cc of 0.25% Marcaine without epinephrine is injected into the joint. After a few minutes, the 2.2-mm inflow probe, either NanoScope or needle sheath, with sharp trocar is inserted through the anterolateral (or anteromedial) portal into the knee joint, the trocar is withdrawn, the one way stop valve is attached, and the 1.9-mm NanoScope or 18-gauge needle is inserted. When using the NanoScope, a 30-cc syringe of sterile 0.9% normal saline is then connected to the device via a one-way stop valve. The valve is opened, and the saline is used to insufflate the knee. This is followed by injection of additional saline, up to 90-100 mL total, injected as needed to insufflate the joint and improve visualization. The diagnostic

arthroscopy of the knee is then performed based on the patient's clinical history, physical examination, and any previously obtained diagnostic imaging. The superior recess of the joint, the patellofemoral joint, the lateral and medial pockets, the medial part of the joint, the intercondylar notch, and the lateral part of the joint are examined. The condition of the synovial membrane of the joint, the integrity and condition of the articular surfaces of the knee joint, and the presence of chondromatous (foreign) bodies are assessed. Next, an arthroscopic probe (feeler) is inserted through the anteromedial port to assess the presence of chondromalacia by pressing on the articular cartilage. Intra-articular structures such as the medial and lateral meniscus, anterior and posterior cruciate ligaments are also assessed.

Using the opposite portal, a NanoScope may be used to evaluate the pathology; it is especially useful for sizing chondral defects and/or for probing the meniscus. If necessary, the scope may be alternated between the anterolateral and anteromedial portal sites, depending on required visualization, although it is recommended that unnecessary withdrawal and subsequent reintroduction of the scope be avoided to maintain a high efficiency of the procedure.

[Laser Treatment]

Under ultrasound or NanoScope guidance, insert the fiber with introducer cannula and adjust the fiber tip to be proximal to the zone of chondromalacia as diagnosed by the MRI. The fiber tip position is determined tactilly by contact with the target cartilage surface at the desired treatment spot followed by withdrawal to the appropriate fiber-cartilage 'quasi-contact' gap (0.1 – 0.3 mm). Optimally, the fiber should be perpendicular to the cartilage surface (90 degrees) but no less than 30 degrees.
The laser's "Start Procedure' button must be pressed to activate the footswitch for firing the laser. When ready, press and hold the foot pedal continuously for approximately 50 seconds while maintaining fiber in quasi-contact with the cartilage. Release the foot pedal when the single-spot treatment is complete. Treatment progress is shown by the advancing green bar below the pulse train schematic (Fig. 9).

Fig 9. Emission Mode screen.

At the end of each single spot treatment, the assistant should select 'Next Spot' to continue the treatment for an additional spot or 'Finish' to end the procedure (Figs. 10a and 10b).

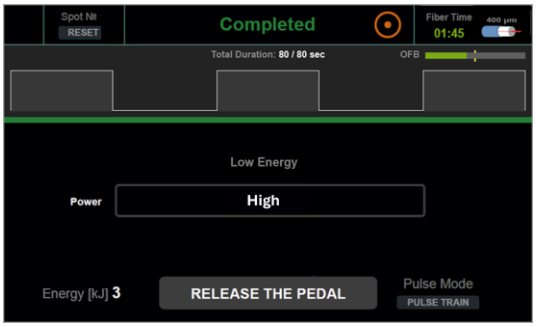


Figure 10a. Completed Single-Spot Treatment

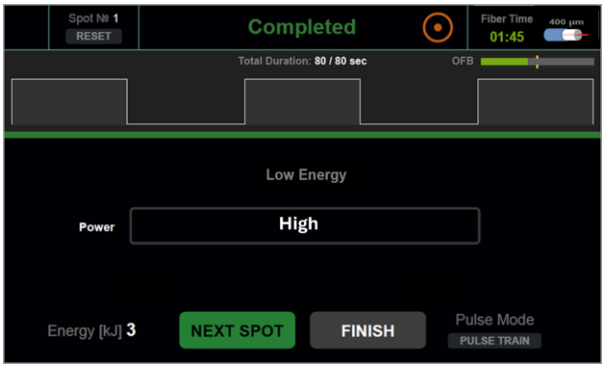


Figure 10b. Completed Single-Spot Treatment & Finish Procedure

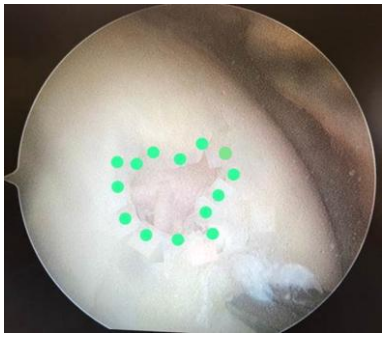


Fig. 11. Suggested placement of treatment spots around cartilaginous lesions.

After treatment of a spot, retract the puncture needle approximately 2 mm and relocate the fiber tip to treat a new spot in quasi-contact. Maintain no less than 1.5 to 2 mm distance (i.e., pitch) between spots. Repeat this pattern of retracting the puncture needle and advancing/treating until you cover a circumferential distance around the defect approximately equal to three times the diameter of the defect (Fig. 11). The approximate number of treated spots for adequate coverage around a lesion of a given diameter is shown in Table 1.

Table 1 Suggested range of number of treatment spots depending on lesion dimension

Lesion Diameter (mm)	Min # Spots (Ave pitch 2 mm)	Max # Spots (Ave pitch 1.5 mm)
10	9	16
15	12	24
20	16	31
25	20	39
30	24	46

After completing circumferential treatment of the cartilage lesion, the fiber instrument together with the puncture needle is removed from the knee joint cavity, after which the joint cavity is washed with a physiological solution, the shaft is transferred to the upper recess of the knee joint, and all the fluid is evacuated from the joint cavity. Skin sutures and an aseptic bandage are applied to the access wound(s).

The assistant may then record the procedure's energy dose (in kiloJoules, kJ, lower left on the display), if desired, and turn off the power switch. To maintain optical cleanliness, the cap should be kept on the laser B port when a fiber is not installed.

6. Post-treatment Procedures

The post-procedural plan includes permitted weight-bearing and range of motion, as tolerated, and the use of acetaminophen and/or NSAIDs for pain control. No narcotics are prescribed post-procedure, and no sedatives are used before or during the procedure. In summary:

- Use oral or over-the-counter analgesics.
- Early stress on the limb post procedure should be about five days post Tx.
- Begin physical therapy the same week.

Post treatment procedure NSAID's, especially if using PRP, bone marrow or fat, can be an issue and three days of Ultracet or hydrocodone with acetaminophen is probably preferred.

Activity

- Initially, limit the weight/stress on the treated limb. After 5 days, the patient may put as much weight as comfortable on their leg.
- Patient may bend and straighten the knee as much as they like.
- Patient should not engage in prolonged periods of standing or walking the first day after surgery.
- For two weeks after the procedure, patient should avoid long periods of sitting or travel involving long periods of continuous sitting.

Dressings and Incisions

- During the first 2 days after surgery, patients can expect a small amount of red-tinged drainage on their dressings. This is normal.
- Keep the dressing clean and dry; if going to shower/bathe, the dressing should be protected. You may not soak in a pool, lake, hot tub, or the ocean for 2 weeks.

- Patient may remove the dressing 4 days after surgery.
- After 4 days plaster dressings may be applied to the portals or they may be left open to air.
- Do not use bactericidal ointments on the portals.

Pain and Inflammation

- Ice: ice bags wrapped in a dry towel may be applied for pain relief and to lower inflammation.
- Compression: use compression elastic bandage as needed to decrease swelling, but this is not required.
- Elevation: keep foot elevated above heart level if increased swelling or discomfort occurs.
- Pain Medication: 500 mg to 650 mg of acetaminophen may be taken every 4 to 6 hours as needed and as directed per the package insert. Do not take more than 3 grams or 3,000 mg in 24 hours.
- Anti-inflammatory medications (Ibuprofen, etc.) may be taken as needed and as directed per the package insert.

Emergencies

- Someone should remain with the patient for the first 24 hours after surgery.
- Patient should call the clinic or the orthopedist on call if:
  - Drainage from the incision soaks the dressings and continues to drain after a bandage change,
  - A fever (>38.5˚) or chills develop,
  - Leg or calf pain, leg swelling, or difficulty breathing occurs.

[Follow-up scheduling]

Schedule a follow-up visit with the patient to review the surgery 10 to 14 days postoperatively. A remote video or phone follow up instead is at your discretion.

The procedure may be followed by irradiation of the treatment area with the 980 nm wavelength laser during follow up office visits. This irradiation is applied topically (noninvasive) and may be performed as frequently as practical (See Regenalase™ IFU for 980 nm laser settings).

7. Combination with Biologics

[List of compatible biologics]

(1) Platelet-rich plasma injections (PRPs) – Recommended

Platelet-rich plasma is an autologous concentration of platelets containing a variety of growth factors and cytokines postulated to aid in cellular anabolism and tissue healing. Formulation is based on centrifugation of whole blood products into component fractions, which are subsequently re-centrifuged to prepare a final product. In general, however, four main categories of PRP final products exist, classified based on the presence of fibrin network and/or leukocytes: 1) pure platelet-rich plasma (P-PRP); 2) leucocyte- and platelet-rich plasma (L-PRP); 3) pure platelet-rich fibrin (P-PRF); and 4) leucocyte- and platelet-rich fibrin (L-PRF). However, pure platelet-rich plasma (LP-PRP) (leukocyte-poor; with low density fibrin architecture) is the most commonly utilized for the treatment of cartilage degeneration, as it has shown the most promising clinical benefit, whereas leukocyte rich- and platelet-rich plasma (LR-PRP) may actually potentiate osteoarthritis.

The PRP substance also may be irradiated by the laser with the wavelength at the 980 nm. A number of studies have shown that platelets act as cell elements and are capable of participating in tissue regeneration because of their high content of proteins, especially growth factors, and that they can enhance the regeneration process. In-vitro release studies have shown that photoactivated PRP via PAC (a plasma arc light source) releases significantly more prolonged and higher amounts of PDGF, bFGF, and TGFß than PRP activated with CaCl2. The release of growth factors from PRP activated by PAC was primarily mediated through the diffusion process.

(2) Bone marrow-derived mesenchymal stem cells (BMSCs)

BMAC formulation was prepared by harvesting bone marrow from three sites on each patient's iliac crest, centrifuged down to obtain marrow cell concentrate (median 34,400 MSCs), and combined with platelet-poor bone marrow plasma prior to injection. Autologous MSCs potentially eliminate the risks for a post-injection immune response, which theoretically allows for increased efficacy relative to their allogeneic counterparts. However, autologous stem cell harvesting can cause donor site morbidity and, if the MSCs are cultured prior to injection, also entail a waiting period.

(3) Adipose-derived mesenchymal stem cells (ADSCs)

Adipose-derived mesenchymal stem cells (ADSCs) have emerged as another promising treatment for knee cartilage lesions and osteoarthritis (OA). Unlike bone marrow-derived MSC harvesting, which is associated with significant donor site morbidity and pain, adipose derived MSCs can be acquired via a minimally invasive technique with few or no complications. In addition, BMSCs require a long expansion time in vivo due to low stem cell yield per sample, whereas ADSCs are associated with both elevated stem cell yield from lipo-aspirates and faster cell proliferation.

[Recommendations on timing of biologics insertion after laser treatment]

The laser treatment should be performed first. After the laser irradiation, the chosen biologic compound may be injected through the same 18 gauge access puncture needle used to insert the fiber with the aid of the same visualization device and protocol used to guide the laser treatment (i.e., Ultrasound or NanoScope). The injection of the biologic compound may be repeated as needed during follow up visits. Commonly, the second injection is administered 1.5 – 2 months after the laser treatment and first injection.

Safety precautions

(1) Complications caused by anesthesia may occur as a result of patient's intolerance to medications, the patient's condition, or anesthesiologist's error.

(2) Hemarthrosis is a massive hemorrhage into the joint cavity. It is extremely rare and almost never seen after arthroscopy.

(3) Thromboembolic complications are rare with adequate precautions and compression garments. In exceptional cases, patients may develop thrombosis of the veins of the lower limb or pulmonary embolism (PE).

(4) With excessively rapid resumption of physical activity, synovial fluid can leak out of the joint cavity and into the peri-articular tissues. To avoid this, early stress on the limb should be avoided.

(5) Damage to nerve branches during surgery can cause paresthesia in the knee joint. In this case, the patient may feel a "crawling sensation" after arthroscopy.

(6) Stretching of the medial collateral ligament. This is rare but can occur if the surgeon performs intensive manipulations, increasing the distance between the femur and tibia for better access to the meniscus.

(7) Deforming osteoarthrosis may develop late after the procedure as a result of progressive damage to the articular cartilage. It is difficult to treat and over time can lead to impaired function and mobility of the knee.

8. Assessment of Clinical Improvement

Recommended follow-up times are 1 mo, 3 mo, 6 mo, and 12 months post treatment. The following tests are suggested to assess clinical improvement:

8.1 The Knee and Osteoarthritis Outcome Score (KOOS)

The Knee and Osteoarthritis Outcome Score (KOOS) can be used to evaluate short-term and long-term symptoms and function in subjects with knee injury and osteoarthritis. The KOOS holds five separately scored subscales: Pain, other Symptoms, Function in daily living (ADL), Function in Sport and Recreation (Sport/Rec), and knee-related Quality of Life (QOL).

8.2 The 36-Item Short Form Survey (SF-36) Bodily Pain Scale

The SF-36 Bodily Pain Scale is based on two questions (a. how much pain during past 4 weeks and b. how much did the pain interfere with your normal work?). A 6-point rating scale that spans from “none” (0) to “very severe” (6) is used for the first question. A 5-point scale from “not at all” (0) to “extremely” (5) is used for the second question.

8.3 Visual Analog Scale (VAS) for pain

The VAS pain score is a scale ranging from 0 (“no pain”) to 10 (“Worst imaginable pain).

8.4 MRI Evaluation

MRI imaging at follow ups are compared with images at baseline.

8.5 Naughton Protocol

Treadmill Test measures pain during a 12 minute walk at a constant speed of 2.0 mph with a gradual increase in incline as per protocol. The patient rates pain from 0-10 every two minutes and at the end of walk. The test is stopped if the patient’s pain increases 2 to 3 levels from baseline or if the patient can no longer continue due to poor cardiovascular conditioning.

8.6 Wall Squat Test

The Wall Squat test assesses lower body strength and mobility The patient performs a 60 degree (partial) squat with a Swiss Ball. The exercise is first demonstrated and then the patient is allowed three trials, focusing on proper form and controls. The patient performs as many squats through the instructed range of motion as possible in one minute. The number is recorded and compared with baseline.

Addendum A. Typical list of supplies

1. Syringe with needle for anesthesia
2. 18-gauge needle with introducer and Luer lock for fiber
3. Alcohol, povidone-iodine, or chlorhexidine swabs
4. Gauze pads
5. Bandage
6. Ethyl chloride spray
7. Pillow or wedge for patient comfort