



Qrem® Cytokine – A Lab-in-a-Box technology to obtain an autologous Cytokine Rich Serum (CRS) from the patient's blood.

General information - Fundamentals - Indications - Bibliographic review

Tecnologia Regenerativa Qrem is a technology company founded in Barcelona in 2016, with a medical device manufacturer's license number 7096-PS granted by the Spanish Agency for Drugs and Medical Devices (AEMPS).

The company's mission is to lead the implementation of autologous regenerative therapies using Lab-in-a-Box devices (a technology 100% automatic, sterile and easy to use) that allow offering these therapies to the largest number of people in a cost-effective way at the physician's office.

A new strategy in tissue regeneration

Tissue regeneration by modulating the immune system is the new way of understanding regenerative medicine.

Stem cell based strategies are still being developed in the search for broad clinical efficacy. Immune-mediated tissue repair and regeneration mechanisms could be added to existing regenerative strategies or be an alternative to the use of stem cells^{1,2}, for example: the mesenchymal cells of bone marrow or adipose tissue.

Cytokines are key in this new strategy by driving the complex process of tissue regeneration.

Cytokines

Cytokines are a family of low molecular weight proteins produced by many types of cells, including blood cells and platelets. They are responsible for regulating the immune response, inflammation, tissue remodelling, and cell differentiation.

Cytokines can be classified in different ways. Among them we can find pro-inflammatory cytokines that promote inflammation, anti-inflammatory cytokines that produce the opposite effect, and growth factors that stimulate tissue repair among many other functions.

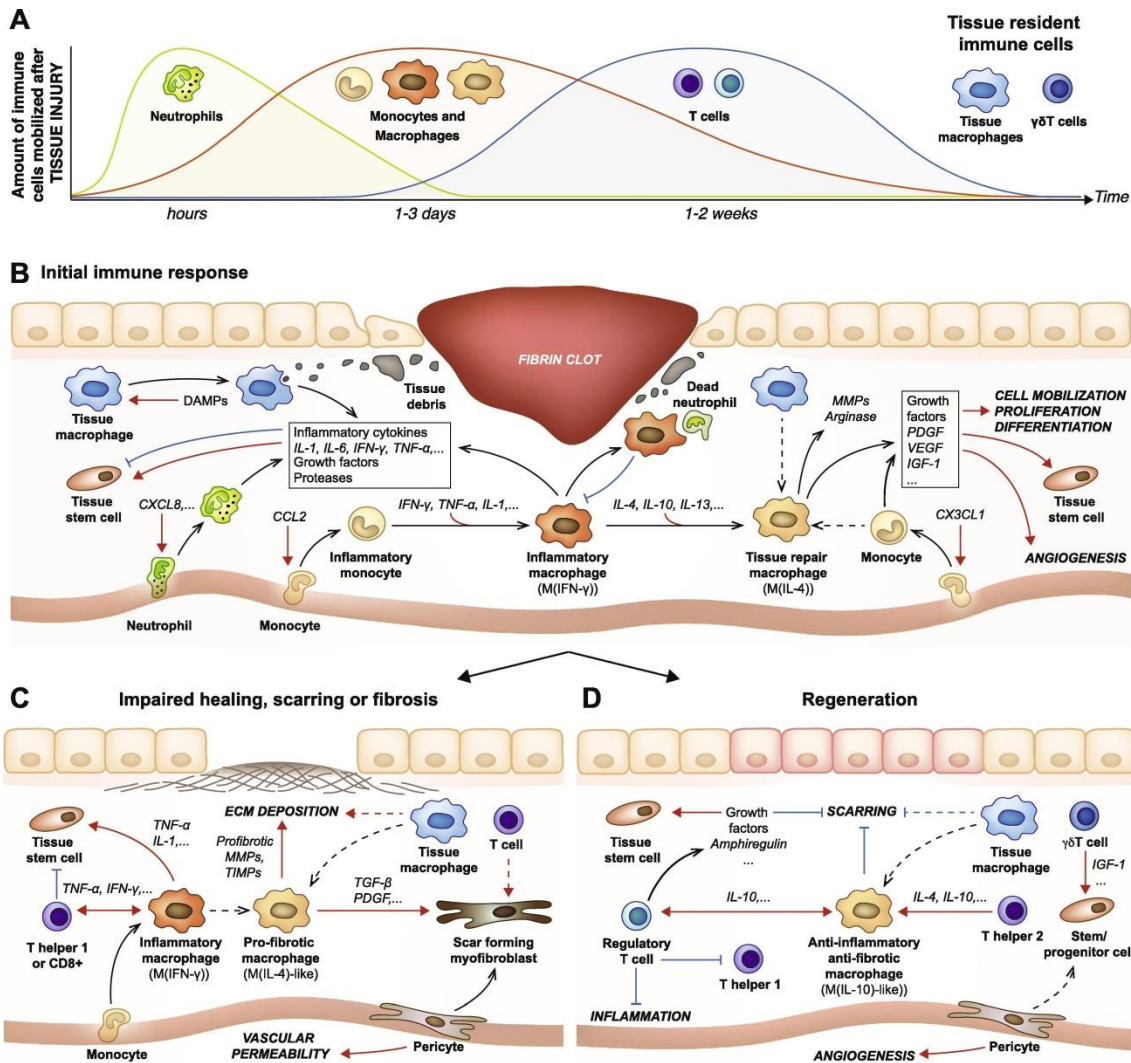
The inflammatory response produced in a damaged tissue is adequate when there is homeostasis, a balance of pro-inflammatory and anti-inflammatory cytokines and growth factors. Although inflammation is a necessary part of the healing process, it should not last

¹ <https://doi.org/10.1016/j.actbio.2017.01.056>

² <https://doi.org/10.1016/j.regen.2018.03.001>

for a long time, because can permanently damage the tissues, causing the degradation of the rest of the elements that are part of the joint

Tissue regeneration is driven by an extensive network of cytokines ³ that can act synergistically as shown in the following figures.



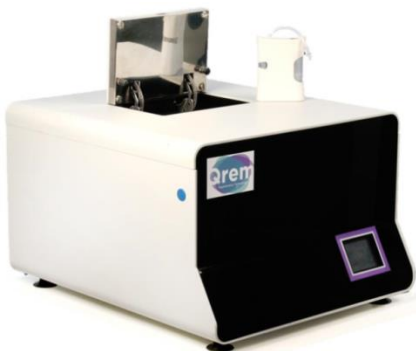
Qrem Cytokine

Qrem Cytokine is an integrated bioengineering system for obtaining the cytokines involved in tissue regeneration. An autologous Cytokine Rich Serum (CRS) is obtained from the patient's own blood through a patented biological process that emulates an ex vivo tissue damage.

Qrem Cytokine is a Class IIa medical device and it has the CE mark MED31489 granted by the Notified Body Nr. 0476.

³ <https://doi.org/10.1016/j.actbio.2017.01.056>

A Lab-in-a-Box system (closed, sterile and 100% automatic), consisting of a desktop device (Qrem Device) and a single-use sterile disposable (Kit Qrem Cytokine). It can be installed in any physician's office without the need for a special infrastructure or personnel with specific training.



QREM DEVICE

Desktop automatic device



KIT QREM CYTOKINE

Single use sterile disposable

How Qrem Cytokine works



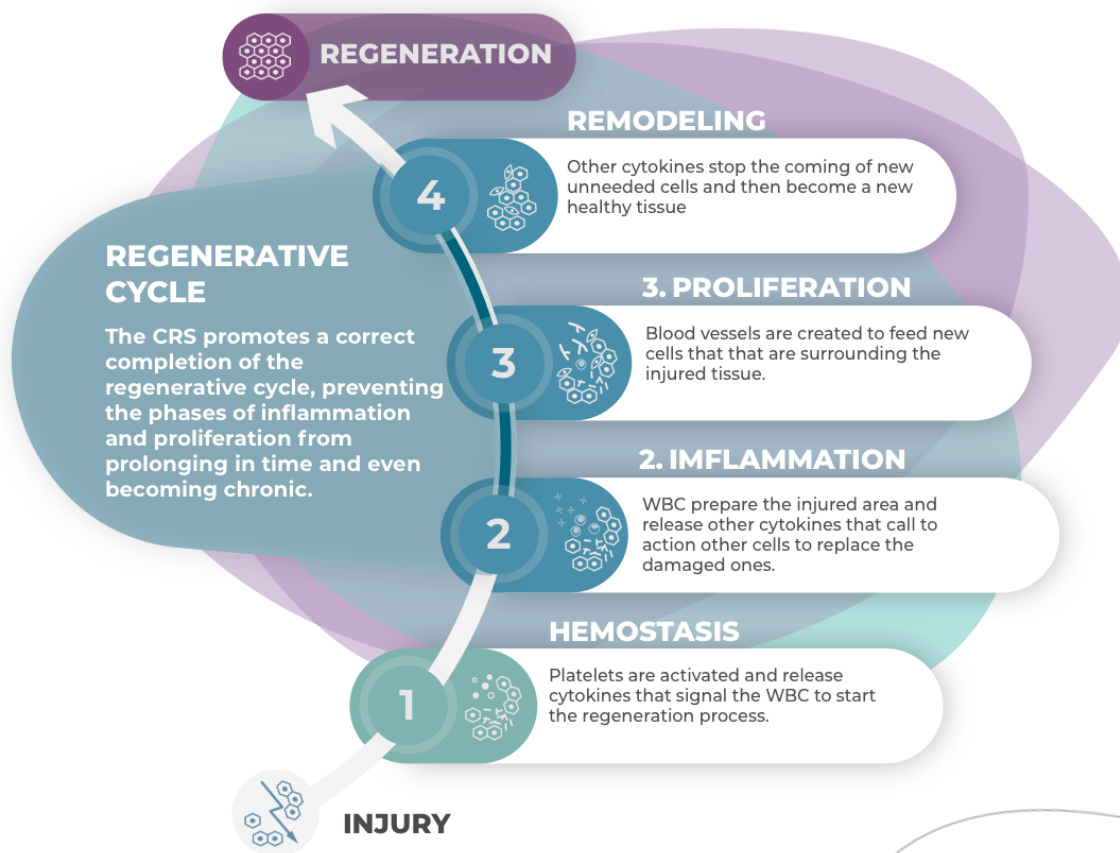
QREM DEVICE performs a multi-step process that takes approximately 35 minutes. This process emulates an ex vivo damage situation where the blood cells are activated releasing the cytokines that modulate regeneration contained in the CRS.

The KIT QREM CYTOKINE is composed of two containers (F and A) joined by a channel containing a valve that allows the fluid transfer from F to A at the appropriate time of processing. Container F is filled with 18ml of the patient's blood plus 2 ml of sodium citrate (anticoagulant). Container A is filled with 20 ml of blood. Filling is done through luer lock ports having non-return valves that ensure a closed process. The blood and the obtained CRS are confined throughout the process inside the KIT QREM CYTOKINE guaranteeing that the process is sterile and safe.

The first steps of the process allow obtaining a platelet and leukocyte concentrate in container F and a primary serum in container A. This primary serum contains autologous activating elements such as thrombin and calcium, as well as anti-inflammatory cytokines and growth factors. The next step is the opening of the valve that allows the mixing of the platelet and leukocyte concentrate with the primary serum in container A, which leads to their activation, forming a fibrin matrix. Through the last steps the matrix is broken releasing the CRS. All steps are performed automatically by the QREM DEVICE.

Cytokine Rich Serum (CRS)

CRS is an acellular orthobiological product that contains the cytokines responsible for directing tissue regeneration, such as: IL-10, TGF- β 1, PDGF and IGF-1. This product offers the possibility of carrying out regenerative strategies based on the modulation of the immune system.



CRS can be applied locally through infiltrations to treat musculoskeletal diseases or injuries. It also offers the possibility of being used in combination with other existing regenerative strategies.

While some biological products already on the market seek to obtain a single cytokine to take advantage of its specific effect, CRS is a natural serum that contains a balanced mix of cytokines that will act collaboratively in tissue regeneration⁴.

CRS uses the synergistic properties of cytokines to stimulate various signalling pathways, promoting the reduction of inflammation and activating the repairing of cartilage. The use of various cytokines and growth factors in a balanced way enhances the stimulation of the different endogenous repair pathways.

Applicable regulations

CRS is an acellular autologous biological product derived from patient's blood obtained without significant manipulation.

In Europe its use is regulated by Directive 2002/98 and the additional regulations that each Member State considers⁵. For example, in the case of Spain, it is therefore subject to the regulations established by the AEMPS (Spanish Agency for Drugs & Medical Devices) for this type of products⁶.

In any case, the Lab-in-a-Box device used to obtain the CRS is subject to the regulation of medical devices.

Differences between CRS and PRP (Platelet Rich Plasma)

The main difference is the use of leukocytes during the CRS preparation process, which allows obtaining anti-inflammatory cytokines^{7,8}.

CRS does not contain platelets, fibrinogen or any blood cells. It directly contains anti-inflammatory cytokines and growth factors that are the active principles involved in regeneration.

PRP is a concentrate of mediating platelets activated with the addition of CaCl₂ to promote the release of the granules, while CRS is obtained through a totally biological process, without additives.

The success of the clinical experience with PRP, mainly in patients with moderate pathologies, has led to the exploration of new orthobiological products such as Qrem Cytokine to improve efficacy and broaden the range of patients who may benefit^{9,10}.

⁴ <https://www.sciencedirect.com/science/article/abs/pii/S2468498817300215?via%3Dihub>

⁵ https://ec.europa.eu/health/sites/health/files/blood_tissues_organs/docs/swd_2019_376_en.pdf

⁶ <https://www.aemps.gob.es/medicamentosUsoHumano/medSituacionesEspeciales/docs/PRP-AEMPS-DEF-mayo13.pdf>

⁷ <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4747972/>

⁸ <http://www.elizavetakon.it/images/2017/10/10/ZB%20EMEA%20Kneed%20To%20Know%20Newsletter%208-18-17.pdf>

⁹ <https://orthogen.com/en/home-2/>

¹⁰ <https://www.zimmerbiomet.com/emea/nSTRIDEtrial>

Clinical results so far

There are studies of similar products such as nSTRIDE from ZimmerBiomet¹¹ and Orthokine from Orthogen¹² that clinically validate the use of autologous blood-derived cytokines, specifically in knee osteoarthritis.

A pilot clinical study in knee osteoarthritis was conducted with Qrem Cytokine, the results of this study are summarized below.

Eight patients diagnosed with knee osteoarthritis participated in the study:

- 5 women and 3 men
- Average age 69.8 years (range: 50-80 years)
- Kellgren-Lawrence grade: Gr. II (4 patients), Gr. III (1 patient) and Gr. IV (3 patients)
- One grade-IV and two grade-II patients also had meniscus tears.

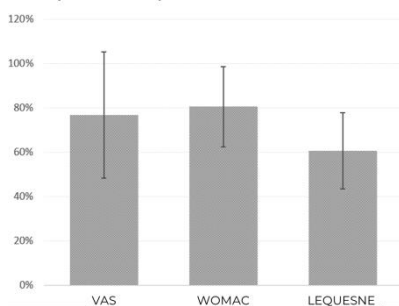
All of them treated with 3 intra-articular CRS infiltrations every 5/7 days in the physician's office. Before each of the first 2 infiltrations, the synovial fluid is extracted from the cavity to be analysed.

After the infiltration, the patients were able to go home immediately without requiring assistance and lead a normal life. Some of them reported local pain in the area of the infiltration and slight local inflammation for a few days.

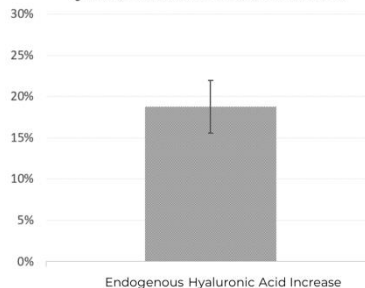
The main conclusions of the study that can be seen graphically at the end of this explanation were:

- The 8 patients treated presented a significant clinical improvement at 12 months of treatment, which is shown in the evident reduction of the VAS, WOMAC and Lequesne indexes. The average improvement after one year of treatment is at least 60%.
- An increase in the concentration of endogenous hyaluronic acid in the synovial fluid was observed after 2 weeks
- A decrease in the pro-inflammatory cytokine IL-6 was also observed in synovial fluid after 2 weeks.

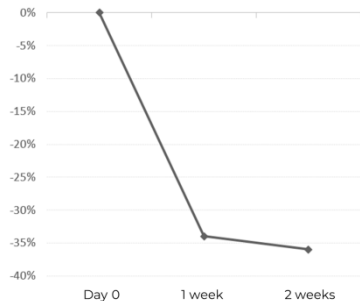
Improvement in pain and function at 12 months



Increase in endogenous Hyaluronic Acid in synovial fluid 2 weeks after treatment



Decreased IL-6 in synovial fluid from the start of treatment

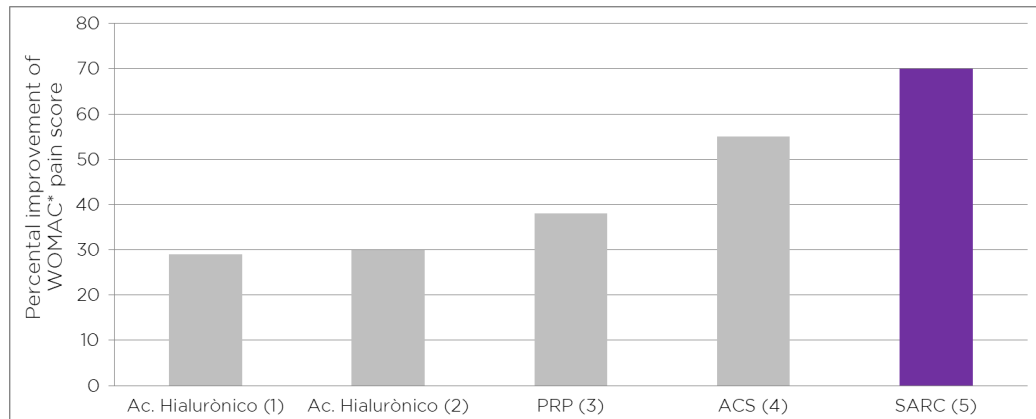


¹¹ <https://www.zimmerbiomet.com/emea/nSTRIDEtrial>

¹² <https://orthogen.com/en/home-2/>

Qrem Cytokine value proposition

- A treatment for osteoarthritis and other musculoskeletal diseases with proven efficacy. A higher pain reduction and a better recovery of mobility and quality of life.



(1) Petrella et al., *Effects of Hyaluronate Sodium on Pain and Physical Functioning in Osteoarthritis of the Knee. A Randomized, Double-blind, Placebo-Controlled Clinical Trial.* Arch Intern Med 2002; 162: 292-298. (2) Huang et al., *Intra-articular injections of sodium hyaluronate (Hyalgan) in Osteo-arthritis of the knee. A randomized, controlled, double-blind, multicenter trial in the Asian population.* BMC, Musculoskeletal Disorders 2011, 12:221. (3) Sanchez et al., *A Randomized Clinical Trial Evaluating Plasma Rich in Growth Factors (PRGF-Endoret) Versus Hyaluronic Acid in the Short-Term Treatment of Symptomatic Knee Osteoarthritis.* Arthroscopy: The Journal of Arthroscopic and Related Surgery 2012, Vol 28, No 8 (August), pp 1070-1078. (4) Baltzer et al., *Autologous conditioned serum (Orthokine®) is an effective treatment for knee osteoarthritis.* Osteoarthritis and Cartilage 2009, 17, 152-160. (5) Data on file, Qrem

- A new regenerative strategy based on the modulation of the immune system.
- A novel and pioneering technology, Lab-in-a-Box
- A closed, sterile and automatic technology that guarantees the sterility of CRS (Cytokine Rich Serum) and allows it to be used in the physician's office.
- Obtaining of CRS is very fast, the physician has the autonomy to administer the CRS during the first visit avoiding the risks that the conservation and preservation of this type of biological products may entail.
- An innovative system very easy to use

Indications to be treated with cytokines

The CRS must always be used under medical prescription and it must be a medical doctor who, having made a previous accurate diagnosis, decides the suitability of providing this treatment to the patient.

The dosage depends on the indication, the severity of the diagnosed pathology and the clinical evolution of the patient. This dosage in the initial treatment is usually 2 or 3 injections, each of them separated between 7 and 15 days.

The CRS is being used for the treatment of different musculoskeletal pathologies:

- Articular osteoarthritis: knee, hip, ankle, shoulder, ...
- Tendinopathies: epicondylitis, supraspinatus tendon, shoulder rotator cuff
- Muscle injuries

Great benefits have also been observed in other indications such as:

- Treatment of local pain
- Scars and ulcers healing
- Dermal regeneration

Regenerative Medicine treatments in musculoskeletal pathologies

While treatments with mesenchymal stem cells from bone marrow and adipose tissue require an operating room, anaesthesia and, in many cases, a specific authorization from Local Health Authorities, blood derivatives such as CRS allow outpatient treatments from 40-60ml blood.

