CHAPTER 15

Minimally Invasive PRGF Treatment for Low Back Pain and Degenerative Disc Disease

AUTHORS

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SUMMARY

Low back pain is a complex and disabling condition and its treatment is a challenge. Traditional surgery to treat lumbar spine pathology has unwanted and suboptimal results. There is now a primary medical and clinical research motivation to put into practice minimally invasive therapies, particularly in support of regenerative medicine, in the treatment of spinal pain and degenerative diseases. Infiltrations of autologous platelet rich plasma (PRP) have been widely used as an effective technological approach to tissue repair and improvements in numerous clinical conditions. Lately, PRP has been included in therapies in specific spinal structures to treat spinal lumbar pain associated with degenerative disc pathology and arthrosis. In patients with chronic low back pain, plasma rich in growth factors (PRGF) has been shown to be an efficient new procedure to reduce lumbar and joint pain after intradiscal, intra-articular facet and transforaminal epidural infiltration under fluoroscopic guidance-control in the operating theatre setting.

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1. INTRODUCTION

Low back pain (LBP) is a highly frequently occurring musculoskeletal disorder involving the muscles, nerves (Luschka plexus), ligaments (such as posterior longitudinal ligament) and bones of the spine[1], often related to a lumbar Degenerative Disc Disease (DDD) with multifactorial and multidisciplinary effects, that affects a large global population[2]. LBP is ranked among the top ten causes of worldwide morbidity and disability according to the latest Global Burden of Disease Study 2015, representing an elevated cost for the health system in general[2]. LBP is a complex, personal experience encompassing multidimensional phenomena with physiologic, sensory, affective, cognitive, behavioral, and sociocultural affects[3, 4]. Although in most patients low back pain is self-limiting, there is a subset of patients, whose symptoms will reemerge, labeling the condition as non-specific, chronic low back pain lasting longer than 3 months of evolution. The pain is primarily related to mechanical factors which mediate in muscular strain, intervertebral disc degeneration (IVDD), facet-mediated pain, and radiculopathy, though the precise mechanisms remain to be elucidated. Two fundamental spinal structures specialized in both shock-absorbing and load-bearing are intervertebral discs (IVDs) and facet joints (FJs). In response to mechanical stress, inflammatory synovitis, and degenerative arthritis, these structures undergo chronic degenerative process, which leads to chronic low back pain[5]. Aging is another factor that produces elements of spinal degenerative changes which are the predominant cause of back pain and sciatica in adult population[6, 7]. Magnetic Resonance Imaging (MRI) scan is usually employed as a valuable tool to identify anatomical abnormalities and determine structural and functional degenerative changes of the spine[8-11]. MRI findings change over time on IVDs (such as disc herniations and annular tears), vertebral subchondral bone (VSB, for example Modic changes and intravertebral herniated disc called Schmorl´s nodes) and arthrosis, which are generally used to determine structural and functional regeneration of the spine and the relationship of these pathoanatomical structures with LBP[12, 13]. The degree of lumbar disc degenerative changes can be classified by various grading systems based on changes in type of disc degeneration, signal intensity (such as discal degeneration degree-Pfirrmann Classification), and IVD (IVD height (IDH), vertebral body height (VBH), and lumbar disc pathology) or VSB (endplate degeneration degree-Modic Changes, and total endplate damage Score-TEPS) parameters from MRI scans[14, 15].

A remarkable clinical motivation for reducing pain and treating spinal degenerative disease arises from the need to return patients to their daily routines and improve their life quality. The appropriate treatment remains a daunting challenge despite advances in the management of pain, inflammation and degeneration by pharmacological and surgical procedures. Before a conventional and surgical procedure is performed, the patient should be offered an incrementally progressive treatment plan, beginning with a minimally invasive interventional therapy[16-23] for symptomatic disc herniation and degeneration, and lumbar FJ arthrosis.

2. SPINAL ANATOMY AND BIOLOGY: INTERVERTEBRAL DISC AND FACET JOINT

Intervertebral disc (IVD), vertebral subcondral bone (VSB), and facet joints (FJs) are important anatomical elements of a spinal column multivariate system affected by pain and degenerative pathology (vertebral arthrosis or osteoarthritis (OA) of the spine)[24]. Intervertebral disc is composed of distinct anatomical regions including the central gel-like nucleus pulposus (NP), the peripheral fibrous annulus fibrosus (AF), and cartilaginous endplates (CEP) at the cranial and caudal interface of one side of the disc[25-27]. IVDs provide stable support to contiguous spinal vertebrae and permit

movement to the vertebral bodies, in that way affecting plasticity of the spine[9]. Disc cells (such as NP, AF and CEP cells) take part in the production and sustaining of disc matrix macromolecules (mainly proteoglycans and collagens) that finally control the biomechanical function of the IVD[26-28]. The normal adult human IVD is mainly avascular and only innervated in the outermost layer of the annulus fibrosus[1, 26]. Nutrients are necessary to support disc cell activity, viability and function. Nutrient diffusion into the disc represents a balance between rate of nutrient supply (capillary density and transport) and rate of cellular demand (disc cell density and metabolic rate)[26, 29]. In addition, metabolic wastes from IVD cells are removed by the reverse route before they accumulate inside the disc[26].

In adults, at the ends of each IVD is located a endplate bilayer of cartilage (CEP) and bone (VSB)[29] that takes apart the vertebral bone from the IVD itself and prevents the vastly hydrated nucleus pulposus from bulging outward to the neighboring vertebrae[30]. In contrast to IVDs, mainly the central endplate of VSB is well innervated as is the adjacent vertebral marrow[30, 31]. It is known that the VSB plays an important role in spinal function, maintaining IVD integrity and disc nutrition supply[24].

At almost every spinal level, lumbar facets or zygapophyseal joints are three-joint complexes with biomechanical capacity as osseous stabilizers of the posterior spinal column facilitating vertebral articulation[32-34]. The three-dimensional (3D) structure is formed by the three articulations between adjacent vertebrae: one anteriorly situated IVD and a pair of small FJs that interact to form a spinal segmental movement complex[32, 35]. At each spinal level, the bilateral FJs are positioned symmetrically relative to the mid-sagittal plane in the posterolateral regions of the vertebral column[33, 35] and are the only true synovial joints between adjacent spinal levels in humans[35]. Facet is a diarthrodial synovial joint with opposing articular cartilage surfaces that provide a low friction environment and a ligamentous capsule that encloses the joint space. Together with the disc, the bilateral FJs transfers loads and guides and constrains motions in the spine as a result of their geometry and mechanical function[33]. Facet joints are anatomically and functionally distinct from the fibrocartilaginous articulation of the IVD[35]. The posterior and medial aspects of lumbar facets resist forward displacement and rotation. At L4-S1segment, angle varies from 30 to 90 degrees, creating more resistance to forward displacement of the superior articular process during flexion and extension but allowing more rotation as the lumbar spine transitions to sacrum[34]. Joint alignment and load distribution are thought to be major factors in the development and progression of FJ OA. Functionally, the three joints in each motion segment are highly interdependent, such that changes in one affect the other two and vice versa[32, 35]. In the majority of individuals, degenerative pathology begins in the disc and is followed by changes in the facet joints affecting the biomechanics of the 3D complex[35]. As a result of biomechanical changes at one level, pathological changes can occur in the motion segment at surrounding spinal levels. In support of the IVD and FJ interdependence concept, FJ OA in the lumbar spine occurs at the levels mainly affected by disc degeneration (L4–S1)[35]. With DDD, the approximation of the vertebral bodies increases the compression on the FJ and changes the relative position of the matching FJ surfaces. The resultant increase in joint compression accelerates wear of the articular facet, producing OA changes[33].

3. PRGF TREATMENT TO REDUCE LOW BACK PAIN

PRP application is a biological and technological approach to tissue repair which has been shown to be an efficient treatment to attenuate and improve several clinical conditions by reducing pain and regenerating tissue[36-40]. Over the past few years, PRP has been used in clinical studies for treatment of vertebral FJ, surrounding ligaments, nerves and IVD to treat pain related to DDD[17-22].

Since the concept of PRGF (PRGF-Endoret) has been developed and characterized[41-45] the application of PRGF-Endoret[40] has been shown to be an efficient treatment to attenuate knee and hip pain of patients with OA and to improve their clinical condition by reducing joint pain[46, 47]. Recently, it has also been reported in an observational retrospective pilot study that fluoroscopy-guided infiltrations of IVDs and vertebral FJs with PRGF-Endoret in patients with chronic LBP resulted in significant radicular and LBP reduction[18].

In light of prior promising results in basic science[48, 49], in preclinical trials, and in osteoarthritic patients whose chronic pain was significantly reduced after PRGF treatment[47, 50-53], results suggest four synergetic effects of PRGF-Endoret on synovial joint diseases, namely, a chondroprotective, anti-inflammatory, cell-phenotype modulation, and joint pain attenuation. These effects make PRGF a strong candidate for treatment of vertebral FJs and IVDD[54].

4. NEW INTRADISCAL PLUS INTRA-ARTICULAR FACET JOINT PRGF TECHNIQUE

In the surgical room of the health centre, low back PRGF protocol (Figure 1A) setup (Kirchner & Anitua, 2016) is performed by a qualified orthopaedic clinical team under international standards from the latest "World medical association declaration of Helsinki" (Brasil, 2013) revised.

Patients are placed in either prone or lateral position depending on patient circumstances. Using sterile technique, intra-articular facet, IVD, and peridural percutaneous PRGF infiltration is performed under X-ray fluoroscopy with a C-arch (BV Libra, Philips, Eindhoven, The Netherlands). Briefly, the percutaneous IVD infiltration approach is used at an oblique angle between 30 and 35 degrees along the lateral margin of the inferior ar-

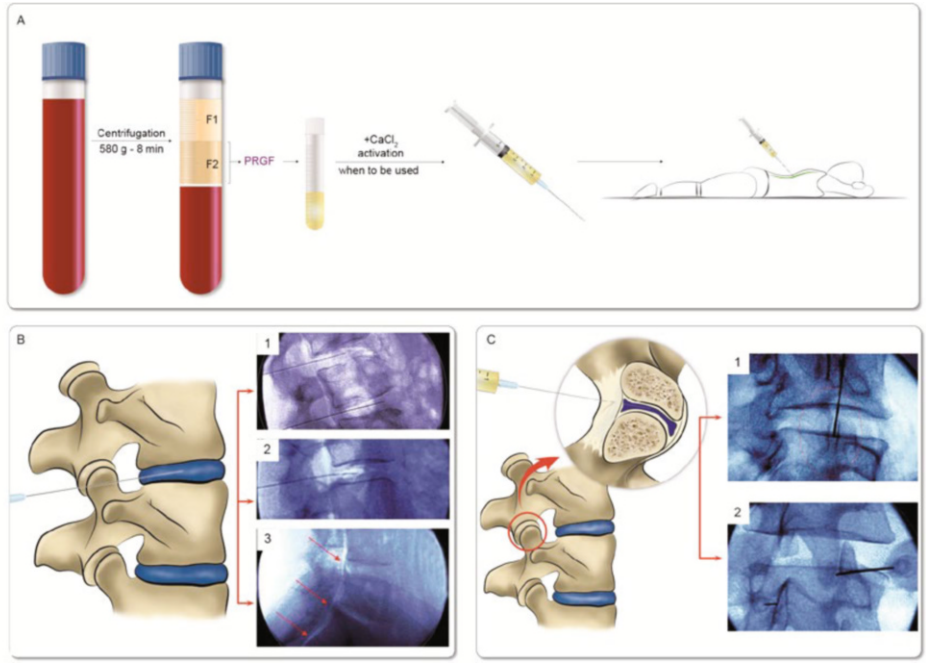

FIG. 1
PRGF preparation and infiltration of disc and lumbar facet joint. (a) Illustrative representation of PRGF obtaining process. Before the percutaneous infiltration, the fraction 2 of PRGF was activated with PRGF activator. (b) Illustration and fluoroscopic-guided lumbar intervertebral disc (insert 1 and 2) images and peridural infiltration (insert 3). (c) Drawing and fluoroscopic-guided lumbar facet joint infiltrations of PRGF showing an intradiscal (insert 1) and intra-articular (insert 2) position of the needle tip (Figure from Kirchner & Anitua, 2016. JCVJS).

ticular process of the vertebrae (Figure 1B, inserts 1 and 2) and through the neuroforamen, while preserving the nerve root. The position of the needle (Quincke needle, 22Ga:178 mm-lenght), which had a manually bent-shaped end, is guided and confirmed under antero-posterior (AP) and lateral fluoroscopic view. Once the needle end is located at the bulging or herniated disc, and checked by infiltrating a little bit of activated PRGF under fluoroscopy, 3 ml of activated PRGF is injected into the NP and the surrounding dehydrated areas while paying careful attention not to allocate more than this PRGF quantity. When the needle is removed from the disc, using the same technical procedure, a peridural infiltration is performed, injecting 1-2 ml of activated PRGF (Figure 1B, insert 3). The spinal FJ infiltration is carried out using the same procedure (Figure 1C, inserts 1 and 2). While the patient lies prone, the joints to be injected are located and marked. After cleaning and draping, the 22-Ga spinal needle is inserted until it contacts the capsule of the joint, and then with careful and refined movements the needle tip enters the joint. Once the needle position is confirmed under fluoroscopy, 1 ml of activated PRGF is injected into each of the zygapophysial or FJs.

5. RESULTS

Intradiscal and facet joint PRGF infiltrations: pain evaluation using VAS outcome

In the retrospective study performed by Kirchner & Anitua 2016[18], the pain assessment was determined using a visual analog scale (VAS) outcome where 0 score denotes "no pain" and a score of 10 denotes "pain as bad as it could be"[3, 55]. The VAS score was evaluated at the first visit before (baseline) and after the procedure at 1, 3, and 6 months. Reduction in VAS score was considered the measure of outcome of interventional therapy with PRGF. The pain reduction after the PRGF-Endoret injections showed a statistically significant drop after the treatment with respect to all the time evaluations. The analysis of the VAS over time showed that at the end point of the study (six months) 91% of patients showed an excellent score (VAS 0-3), 8.1% showed a moderate improvement (VAS 3.1-6.5), and 1.2% were in the inefficient score (VAS 6.6-10) (Figure 2).

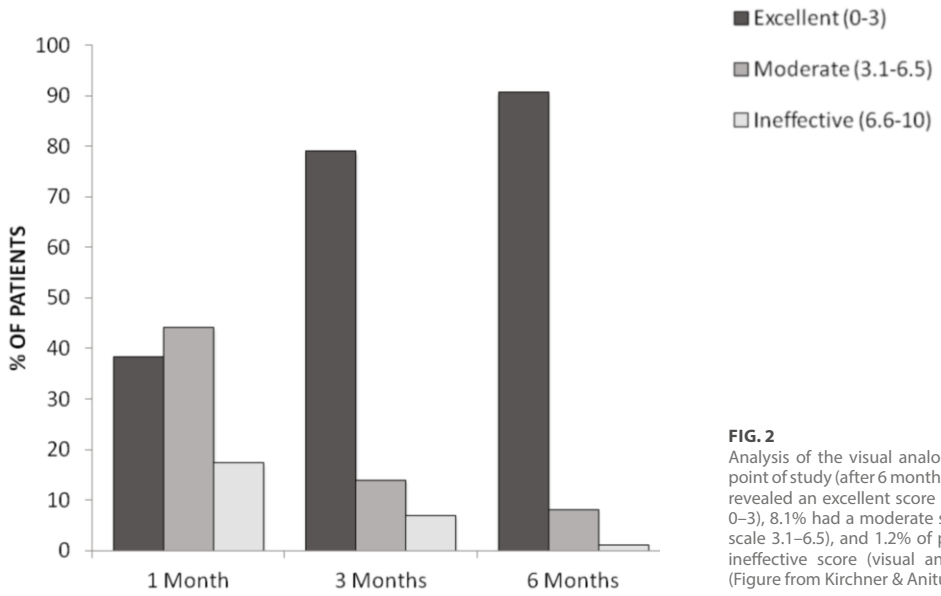

FIG. 2
Analysis of the visual analog scale. At the end point of study (after 6 months), 90.7% of patients revealed an excellent score (visual analog scale 0–3), 8.1% had a moderate score (visual analog scale 3.1–6.5), and 1.2% of patients showed an ineffective score (visual analog scale 6.6–10) (Figure from Kirchner & Anitua, 2016. JCVJS)

Taking into account the limitations and drawbacks addressed at the preliminary study just mentioned[18], and the evidence of other clinical and case reports related to PRP infiltration to treat low back pain[16, 17, 19-22], the authors are confident that infiltrations of IVDs and FJs with PRGF should be considered a valuable therapy in patients with chronic LBP.

Another important point to be considered is the fact that a standardized protocol for PRP infiltration is still not established, and variation of PRP preparation and procedures necessarily results in wide inconsistencies. In addition, the volumes of PRP infiltrations, platelet concentrations, and time differences between injections are still under discussion.

In the treatment of spine pathology PRGF-Endoret therapy, is highly recommended as an additional and specific reinforcing therapy to complete the spinal regenerative process in all patients.

Intradiscal PRGF therapy: Case Report using imaging technique MRI results.

Following an overexertion, a 33 year-old male patient presented to the Traumatology clinic in March 2014 with three months evolution of LBP and symptoms of lumbago and sciatica radiating to the right lower leg and into the foot fingers.

An initial MRI examination in January 2014 revealed decrease of T2-weighted (T2W) discal signal at L5-S1 IVD level indicative of a certain degree

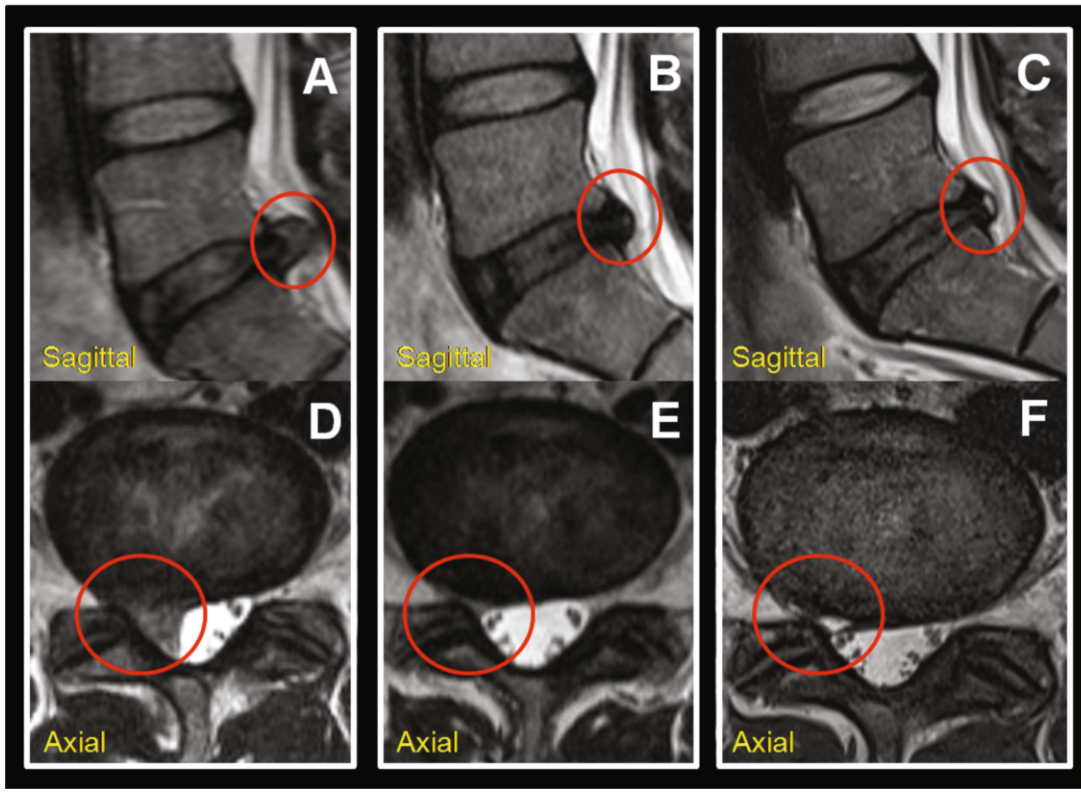

FIG. 3
MRI scan at the lumbar spinal level L5-S1 showing pre and post-PRGF intradiscal treatment T2-weighted (T2W) axial and sagittal section. Red circle delineate extrusion size. (2014) Pre-treatment sagittal (A) and axial (D) MRI scan sections. (2016) Post-treatment MRI sagittal (B) and axial (E) sections. (2017) Final MRI sections at sagittal (C) and axial (F) levels.

of degeneration (Pfirrman degree degeneration grade III), and a significant right-sided postero-paramedial (PP) and preforaminal spinal herniated disc (extrusion) (Figure 3A, 3D) at the L5-S1 IVD space causing compression of thecal sac and right S1 traversing nerve root.

The patient agreed to receive a minimally invasive treatment and underwent three bilateral PRGF applications during his complete therapy. A first, fluoroscopic-guided intradiscal L5-S1, peridural and root percutaneous disc infiltration approach was performed in a surgical suite setting in April 2014. Later, second and third PRGF therapies were completed in an adapted outpatient operating room. The second PRGF infiltration was carried out 20 days after the first PRGF application at L4-L5 and L5-S1 levels. One year after the first PRGF application, the patient received his last PRGF therapy at the same levels as the previous outpatient PRGF infiltrations.

Table 1 reviews the measurements (Figure 4) and findings from MRI scan analysis of the patient before and after PRGF treatment.

A second MRI scan performed approximately two years later (Figure 3B, 3E) revealed a Pfirrmann grade III of degeneration at L5-S1 disc space that was unchanged from the previous posterior lumbar spine MRI examination. The height of L5-S1 disc (Figure 4A) space (IDH) remained decreased compared to the other levels and was slightly variable from the earlier and later lumbar spine MRI

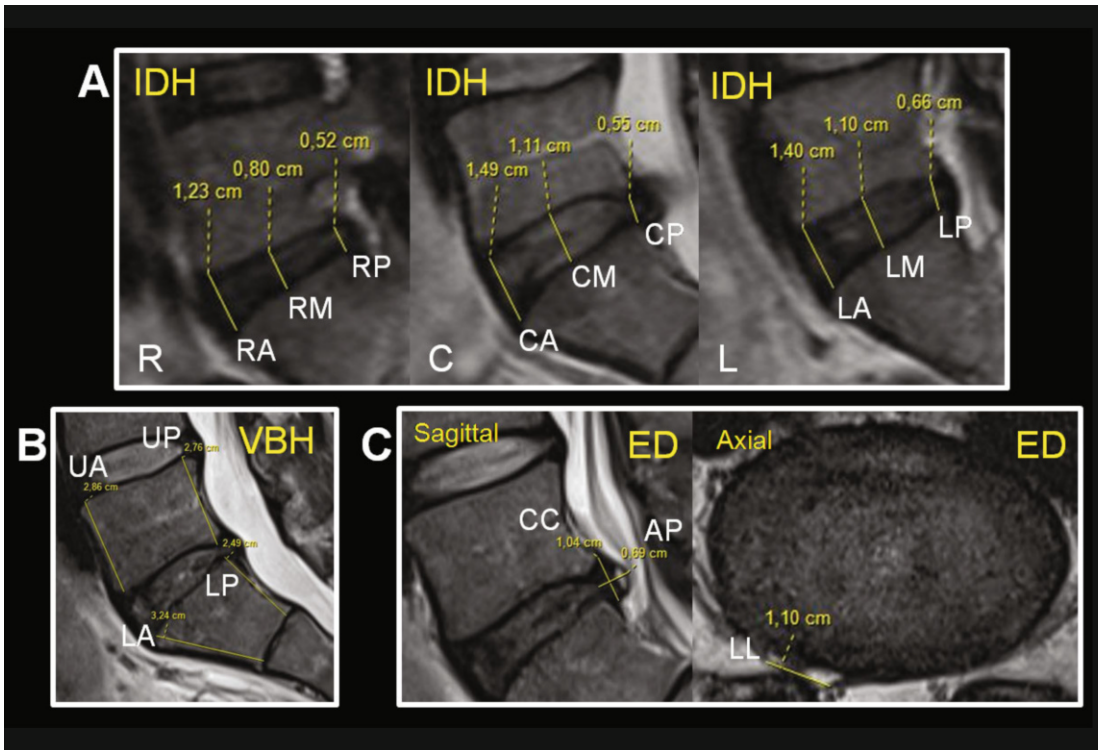

FIG. 4
T2-weighted (T2W) axial and sagittal sections at the lumbar spinal level L5-S1 showing MRI measurements (in mm) using medical imaging remote Workstation AW 4.3 (GE Healthcare). (A) Sagittal measurements of Intervertebral Disc Height (IDH) at Right Anterior (RA), Right Middle (RM), Right Posterior (RP), Centre Anterior (CA), Centre Middle (CM), Centre Posterior (CP), Left Anterior (LP), Left Middle (LM) and Left Posterior (LP) sections; (B) Sagittal measurements of Vertebral Body Height (VBH) at L5-Upper Anterior (UA), L5-Upper Posterior (UP), S1-Lower Anterior (LA), S1-Lower Posterior (LP) sections; and (C) Sagittal and Axial dimension of lumbar disc extrusion diameter at Craneo-caudal (CC), Latero-lateral (LL), and Antero-posterior (AP) sections.

examination. However, an important extrusion resorption (Table 1) was found showing a smaller right-sided PP extrusion diameter (ED) (Figure 4C) at the L5-S1 IVD level evidencing a 42% decrease at latero-lateral (LL) axial section and 22.5% reduction at sagittal AP section without changes at sagittal craneo-caudal (CC) section. No evidence of compression of thecal sac and nerve root on T2W images (T2WI) were observed. Parameters analysed over time for VBH (Figure 4B) showed minimum measurement variation (Table 1). A last, MRI study carried out in January 2017, showed little change with respect to the 2016 MRI report (Table 1, Figure 3C, and 3F). In particular the ED remained diminished and almost the same size as the MRI scans of 2016.

6. DISCUSSION

Kirchner & Anitua[18] showed that a minimally invasive therapy consisting of PRGF-Endoret infiltrations of IVD, FJs and peridural space in patients with chronic LBP resulted in significant pain reduction assessed by VAS outcome. PRGF-Endoret is a treatment without any side effects and no complications were experienced by the patient, so the therapy could be carried out as many times as necessary within the period up to completing the treatment.

At the end of sixth month follow-up period, 91% of patients showed a pain value of 0-3 in the VAS score, which is considered "excellent" in the litera-

MRI parameters: Spine Level L5-S1	Pre-MRI 2014	Post-MRI 2016	Post-MRI 2017
Intervertebral Disc Height (IDH) mm			
Right Anterior (RA)	1.23	1.35	1.42
Right Middle (RM)	0.80	0.95	0.95
Right Posterior (RP)	0.52	0.61	0.52
Centre Anterior (CA)	1.49	1.45	1.51
Centre Middle (CM)	1.11	1.14	1.02
Centre Posterior (CP)	0.55	0.65	0.49
Left Anterior (LP)	1.40	1.19	1.32
Left Middle (LM)	1.10	0.89	0.94
Left Posterior (LP)	0.66	0.66	0.54
Vertebral Body Height (VBH) mm			
L5-Upper Anterior (UA)	2.95	2.91	2.86
L5-Upper Posterior (UP)	2.93	2.72	2.76
S1-Lower Anterior (LA)	3.29	3.20	3.24
S1-Lower Posterior (LP)	2.74	2.47	2.49
Lumbar Disc Pathology	**Hernia-Extrusion**	**Hernia-Extrusion**	**Hernia-Extrusion**
Extrusion diameter (ED) mm			
Craneo-caudal (CC)	1.06	1.06	1.04
Latero-lateral (LL)	1.74	1.01	1.10
Antero-posterior (AP)	1.02	0.79	0.69
Discal Degeneration Degree			
Pfirrmann Classification	Grade III	Grade III	Grade III

TABLE 1
A pre and post-treatment Nuclear Magnetic Resonance (NMR) analysis performed at the L5-S1 discal space and corresponding vertebrae level. According to T2-weighted axial and sagittal sections of the lumbar spine, the networked medical imaging remote Workstation AW 4.3 (GE Healthcare) was used to import, interpret and process DICOM images from MRI scans to categorize and obtain measurements of the following parameters: Intervertebral Disc Height (IDH); Vertebral Body Height (VBH); Lumbar disc pathology; and discal degeneration degree (Pfirrmann Classification).

ture[23]. This result is consistent with the values published by Sanchez et. al.[47] for the treatment of OA of the hip with PRGF infiltration, and comparable to the values reported by Becker and colleagues[56] using epidural perineural injections of autologous conditioned serum (ACS) on patients with lumbar radicular compression, and substantially better than the pain relief shown by infiltrating metylprednisolone in the lumbar FJs[57], triamcinolone in epidural perineural injections[56] or betametasone in periganglionar infiltration[23] in patients with LBP. Nevertheless, the course of treatment reported by Chatuverdi and coauthors[57] reached a peak of 93% of responders at fourth week of treatment and then declined to 62.5% at third month of treatment, an outcome which is not consistent with our results.

The report of Kirchner & Anitua is one of the few in the clinical literatures regarding the use of PRP injections of IVD and FJs as a minimally invasive therapy to treat LBP and IVDD.

Until now, the large number of PRP studies performed on the spine to study pain management and disc pathology have been focused using in vitro (cell cultures) or in vivo (animal model) systems which have produced promising positive results[58-62]. In contrast, so far only two prospective clinical trials have been reported[16, 20, 21] and three papers exist in the literature describing case reports[17, 19, 22] showing positive statistical and clinical improvements based on pain and functional outcomes.

Focusing on clinical studies, at the beginning of 2016, Tuakli-Wosurnu and colleagues reported for the first time pain, function and participant satisfaction outcomes for patients subjected to lumbar intradiscal PRP (Harvest PRP kit, Plymounth, MA, U.S) injection for discogenic LBP[20]. Related to pain outcome, these authors used a Numeric Rating Scale (NRS) to describe pain intensity. After two months follow-up, they reached the maximum mean NRS-pain reduction with statistically significant improvements showing for the participant who received the PRP therapy. Using an approximate comparison of VAS and NRS as two different systems of pain evaluation, higher pain reduction values were registered for Kirchner & Anitua although the maximum mean-VAS score was obtained at six months[18, 20]. However, Levi et. al.[21] using a PRP Harvest Smartprep kit (Harvest, Plymounth, MA, U.S) similar to that used by Tuakli-Wosurnu and coauthors, achieved maximum VAS improvement only in 47% of patients after 6 months with an even lower percentage of patient improvement and VAS score values than Kirchner & Anitua. Levi and colleagues reported as categorical success at least 50% decrease in VAS sore after PRP treatment.

In offering a serviceable comparison, it will be helpful to have three considerations in mind in interpreting the effective differences reported by the above authors: (1) the authors have used two different PRP preparation kits which include a red blood cell-rich and leukocyte PRP preparation (Harvest) and a PRGF formulation without leukocytes (PRGF-Endoret), (2) distinct amounts of PRP volumes to the infiltrated disc were used, and (3) different protocols of PRP total volume infiltration, including the injection of other compounds at the same IVD level, were also performed.

Also notable are the case reports such as Aufiero and colleagues[22], Monfett et. al.[19] and Mascarinas and coauthors[17] which report high VAS and NRS values of pain relieve at different times post- autologous PRP intradiscal procedure.

To date, limited scientific literature exists regarding clinical information on the effect of PRP treatment follow-up and the structural spinal regenerative process. In some cases, a well-known phenomenon for spontaneous regression of herniated lumbar disc material has been documented[63, 64] but the exact mechanism responsible for the regression of herniated IVD is still controversial. Mascarinas and coauthors[17] are the first to have mentioned for a case report of a MRI improvement in disc regeneration after 1 year of intradiscal PRP injection that correlates with improvement in LBP. On the same basis that MRI provides more detailed information about disc herniations, a case report is presented in this chapter showing considerable MRI

improvements in IVD resorption after 2 years post PRGF treatment. As biological strategy developed for regeneration of the IVD, PRP may contribute to understanding of the mechanisms underlying the regression of protruded disc herniation. This case description could illustrate the need to achieve results using imaging techniques for lumbar spine such as conventional radiology, MRI or Computerized Tomography-CT[65, 66] in order to correlate improvements of pain and functional outcomes with vertebral related-structure regeneration in the field of spinal regenerative medicine.

Although the relative contribution of various structures in chronic LBP is varied, the pain source is commonly attributed to inflammatory response of FJs and/or IVDD and herniation of cells due to mechanical stress stimuli[57]. FJs and IVDs undergo chronic degenerative process in response to non-physiological mechanical stresses, bringing about collagen and aggrecan cleavage, a decrease of collagen and proteoglycan synthesis, an increase of proteases and cell death which parallels articular cartilage degeneration in OA and whose clinical hallmark is joint pain[35].

There are several potential mechanisms by which PRGF-Endoret infiltrations might either suppress or slow down the progress of IVD and facet degeneration. PRGF- Endoret is an autologous product that conveys a mimetic biomaterial, namely fibrin, which is embedded with a pool of growth factors (fibroblast growth factor (FGF), platelet-derived growth factor (PDGF), insulin-like growth factor-1 (IGF-1), vascular endothelial growth factor (VEGF), transforming growth factor-β (TGF-β), hepatocyte growth factor (HGF), connective tissue growth factor (CTGF), and nerve growth factor (NGF)) stemmed from activated platelets and plasma, and acting as a biological scaffold from which a sustained and a gradual delivery of GFs is released at the dysfunctional and degenerate sites instead of as a bolus delivery modality. Furthermore, a proteomic characterization study of PRGF fibrin matrix[44] reported a significant representation of acute-phase proteins, a strong network of interconnected proteins linked to the nuclear factor kB (NF-kB) pathway. This pathway is in part responsible for the inflammatory response of stressed cells such as chondrocytes, tenocytes, fibroblasts and macrophages. In addition, both, IVD and FJs share many developmental, functional, and biological features with articular cartilage and synovial joints[67].

In vitro, several of the GFs present in PRP such as TGF-β, IGF-1, and CTGF have been shown to exert a powerful effect on extracellular matrix (ECM) synthesis and proliferation in IVD[68, 69]. Furthermore, PDGF and IGF1 have been shown to exert a cell survival action on IVD cells[70]. In addition, through TGF-β or as a PRP whole product, PRP has been shown to promote the synthesis of ECM components such as proteoglycans and collagen in human NP cell cultures[67, 71]. More importantly, PRP administration using biodegradable gelatin hydrogel microspheres into degenerated IVD animal model resulted in a preservation of water and IDH, suppression of the IVD degeneration progression, and a synthesis of proteoclycans 8 weeks after the treatment, highlighting the importance of both delivering GFs in a sustained and gradual manner and the effectiveness for early IVDD intervention[72, 62]. Similar regenerative effects have been reported applying PRP to DDD in animal models[61, 73, 74].

Inflammation is a term that encompasses clinical, physiological, cellular, and molecular phenomena, with pain being the hallmark or the tip of the iceberg underlying pro-inflammatory cytokine release, ECM catabolism, and cell death. Thus, pain and inflammation are flip sides of the same coin, namely, tissue damage. Data coming from animal studies strongly suggest that proinflammatory cytokines such as tumor necrosis factor-α (TNF-α), interleukin-1β (IL-1β), and interleukin-6 (IL-6) are pivotal for the onset and maintenance of pain mainly stemmed from the damaged peripheral tissues[75]. In contrast, anti-inflammatory cytokines such as interleukin-4 (IL-4) and interleukin-10 (IL-10) have analgesic properties[75]. Some components present in PRGF-Endoret (HGF, lipoxin A4 (LXA4), platelet factor 4 (PF4), IGF-1, PDGF, and TGF-β)[48, 76-79] inhibit the NF-kB signaling pathway in several cell lineages including macrophages, chondrocytes, and fibroblasts. NF-kB plays an im-

portant role in mediating the gene expression of proinflammatory cytokines such as TNF-α, IL-1β, prostaglandin E2 (PGE2) and cyclooxygenase 1 (COX-1) and cyclooxygenase 2 (COX-2)[80].

As a consequence, it is reasonable to consider that PRGF-Endoret could exert an antiapoptotic, ECM-protective, antiinflammatory and pain reduction effect in a similar manner as in knee and hip synovial joints[52, 53, 81]. In macrophages, the inhibition of the NF-kB might contribute to the polarization from M1 to M2 phenotype, thereby favoring the resolution of inflammation and generating a switch in the ECM from a proinflammatory and algesic milieu to an anti-inflammatory and analgesic context. Furthermore, the in situ generated fibrin matrix would be gradually removed as a result of local activation of the tissue plasminogen activator/plasminogen system. This fibrinolytic remodeling process overlaps with the homing of survival fibrochondrocytes and migratory mesenchymal stem cell (MSCs), which might have been attracted by chemoattractans such as SDF-1, HGF, IGF-1, TGFβ or bFGF sequestered within the fibrin matrix and gradually released during the fibrinolytic-remodelling process[82].

Further case studies and clinical trials using standardized methods are necessary if we are to understand the efficacy of PRGF as a minimally invasive regenerative treatment for spinal pathologies where improvements are reliably measured with respect to pain reduction and recovery of structure and function.

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