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    Description: Figure 2 a++bone graft
    File format: image/jpeg

4): Figures 3
    Version: 1
    Description: Figure 2b cages
    File format: image/jpeg

5): Figures 4
    Version: 1
    Description: Figure 3 rigid fusion
    File format: image/jpeg

6): Figures 5
    Version: 1
    Description: Figure 4 dynamic fusion
    File format: image/jpeg

7): Figures 6
    Version: 1
    Description: Figure 4 B dynamic fusion
    File format: image/jpeg

8): Figures 7
    Version: 1
    Description: Figure 5 total disc replacement
    File format: image/jpeg

9): Figures 8
    Version: 1
    Description: Figure 6 treatment stratergies
    File format: image/jpeg

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Degeneration of the intervertebral disc with new approaches for treating low back pain.

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Abstract

This review paper discusses the process of disc degeneration and the current understanding of cellular degradation in patients who present with low back pain. The role of surgical treatment for low back pain is analysed with emphasis on the proven value of spinal fusion. The interesting and novel developments of stem cell research in the treatment of low back pain are presented with special emphasis on the importance of the cartilaginous end plate and the role of IL-1 in future treatment modalities.

Introduction;

Low back pain (LBP) affects many people posing a major public health problem worldwide[1] and in the United States back and neck pain are two of the most common reasons for visiting primary health care physicians and chiropractors[2]. Hoy *et al.,*(2010) concluded that LBP caused more global disability than any other condition and with an aging population stressed the importance for further research to gain a greater understanding into the causes of this problem[3]. LBP can present clinically as acute that is less than six weeks, sub-acute six to twelve weeks and chronic more than 12 weeks[4]. Many patients with chronic LBP have periods free from pain, followed by episodes of increasing pain and disability and can be further classified as suffering from chronic recurrent LBP. The causes of LBP are mostly unknown, however degeneration of the intervertebral disc (IVD) is believed to precede the clinical presentation which includes LBP and or lower limb pain along with paraesthesia, spinal stenosis and disc herniation[5]. Several factors have been suggested as being primary risk factors for LBP, including mechanical loading, age, obesity and smoking[6-9]. Genetics has been shown to have a major role in the development of disc degeneration[10-12] and recently the involvement of bacteria has been revisited to raise the possibility of bacterial infection as a cause of disc degeneration[13-15]. Viruses have also been suggested as causing disc degeneration by Alpantaki *et al*(2011)who isolated the herpes simplex virus type 1 (HSV-1) together with cytomegalovirus from the IVD of patients who underwent spinal surgery for disc prolapse[16].

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The Normal Intervertebral disc

The IVD is an avascular and aneural structure[17,18], composed of three morphologically distinct regions, a hyaline cartilaginous layer which separates the IVD from the adjacent vertebrae known as the cartilaginous end plate (CEP) and a highly organised outer collagenous annulus fibrosus (AF) surrounding a more gelatinous nucleus pulposus (NP).There are a small number of metabolically active cells within the IVD which has been estimated as 9 x 10^6 cell/cm^3 within the AF and 4 x 10^6 cell/cm^3 in the NP[19]. The nutrition of the disc is dependent on the transfer of solutes from the capillaries in the vertebral endplate and outer annulus fibrosus by the process of passive free diffusion dependent upon the Fick principle[20-22]. These vessels end in the CEP and are in continuity with the systemic circulation via the lumbar arteries[23]. Blood flow to the end plates of sheep has been estimated as 4ml/100gm/min which is similar to the cortical blood flow in the tibia[24]. Wallace *et al* (1994) have shown that the blood flow can be influenced by neuro humeral agents such has acetyl choline, which doubles the blood flow to the IVD[25]. IVD cells are also capable of surviving with low levels of oxygen whilst levels of glucose area much more crucial factor with low glucose levels leading to premature cells death especially in an acidic environment[26]. In addition, the activity of IVD cells can be influenced by mechanical load with physiologically normal loading regimes considered to induce anabolic responses whilst abnormal loading induces catabolic responses[27-34].

Mechanically, the IVD is capable of withstanding pressure especially as the body moves when bending and twisting but there is no evidence that the IVD acts as'shockabsorber'[27,35]. Whilst excessive loading or abnormal loading such as vibration can induce damage in the form of end plate rupture and micro-fissure formation[36].

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The Pathology of Disc Degeneration;

Yong-Hing and Kirkaldy-Willis(1983)and Boos *et al*(1994)studied the radiological and pathological changes that take place in the IVD and facet joints especially with advancing age[37,38]. Disc degeneration results in loss of disc height with subsequent altered mechanical stress on the facet joints and other spinal tissues, such as the ligamentum flavum which have been shown to undergo changes in patients with LBP[39]. Loss of disc height can also contribute further to compression of the exiting nerve root resulting in lower limb and leg and buttock pain and Olmarker and Rydevik (1992)demonstrated that long term compression on the exiting nerve root can lead to changes within the spinothalamic tract[40].

It has also been shown that LBP may be attributed to the ingrowth of nociceptive nerves and blood vessels into the inner regions of the IVD. Freemont *et al.,* (1997) identified the presence of peptide-containing sensory nerve fibres within inner regions of the degenerate IVD, and found them to be localised to microvessels in patients experiencing pain[41]. It was later discovered that these microvessels expressed the neurotrophic factor: NGF (nerve growth factor) which is the main neurotrophin responsible for the survival and growth of neurons, and these nerve fibres expressed the high affinity receptor for NGF: Trk A[42]. This led to the hypothesis that blood vessels entered the degenerate IVD first, followed by nerves. The expression of neuropeptides such as Substance P, PECAM, CGRP and NT3 further suggests that the nerves present within the IVD are able to actively transmit signals in response to painful stimuli, such as abnormal load[42-45]. There are various molecules present within the native IVD which contribute to the avascular and aneural nature of this tissue. Aggrecan (the major proteoglycan) is found in abundance within the healthy NP and provides hydration via its ability to imbibe water[19]. *In vitro* studies demonstrated that aggrecan isolated from the native IVD was

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able to inhibit the growth and migration of nerves and blood vessels[46]. Other endogenous molecules within the IVD have been studied for their inhibitory properties towards nerve and blood vessel ingrowth such as chondromodulin[47,48] and tissue inhibitors of metalloproteinase-1 (TIMP-1) and TIMP-2[49,50]. More recently, the semaphorins have been implicated in having a role in the prevention of innervation and angiogenesis of the IVD[51] and articular cartilage[52]. During IVD degeneration however, such molecules are known to become depleted and thus could lead to the inappropriate entry of nerves and blood vessels into the vulnerable degenerate IVD. These potential regulators of nerve and blood vessel ingrowth into the IVD have recently been reviewed by Binch *et al.,* (2014). The question still remains as to what may trigger the cascade of events leading to the innervation and vascularisation of the IVD[53]. Recent studies by Binch *et al.,* (2014) and Krock *et al.,* (2014) confirmed expression of a variety of neurotrophins from NP cells and IVD organ cultures respectively[45,54], and concluded that levels of brain derived neurotropic factor (BNDF)particularly were significantly higher in degenerate IVD samples as opposed to non-degenerate samples.

IVD degeneration is characterised by a number of discrete biochemical, histological, metabolic and functional changes, **Figure 1**, leading to changes in the extracellular matrix composition resulting in dehydration which consequently alters the response to compressive loads[55]. The changes within the disc result from abnormal cell behaviour thought to be brought about by abnormal mechanical loading[30,56], altered nutritional supply[57-59] and genetic predisposition[60-63]. As degeneration progresses the extracellular matrix composition within the NP changes becoming progressively more fibrous with reduced proteoglycan synthesis and increased degradation of the matrix by degradative enzymes[44,64]. Two main families of degradative enzymes are

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involved in the degradation of the matrix during IVD degeneration, these being the matrix metalloproteinases (MMPs) and A disintegrin and metalloproteinase with thrombospondin motifs (ADAMTs) enzymes[65-69]. Both these groups of enzymes are produced particularly by the NP cells of degenerate discs and have been shown to lead to the degradation of collagens and proteoglycans within the degenerate disc [65,68,70]. These compositional changes in the matrix during IVD degeneration are driven by biochemical changes to the cells. In addition to increased cellular apoptosis[71-73] and senescence[74-76] seen during degeneration. The cells of the IVD produce a plethora of catabolic cytokines and chemokines[77-87] with highest expression seen in the NP and inner AF[77,80], expression within the outer AF is differentially expressed, with posterior AF tissue displaying higher expression of cytokines than the anterior AF[85].

There is increasing evidence supporting the role of a pivotal cytokine: interleukin (IL)-1 in the pathogenesis of IVD degeneration[45,70,77,78,80,84,88-91]. With an increase in the production of the IL-1 agonists (IL-1α and IL-1β) and their active receptor IL-1RI, without a concordant increase in the natural inhibitors: IL-1 receptor antagonist (IL-1Ra) or the decoy receptor: IL-1RII; within the cells of the NP and inner AF[80]. IL-1 has been shown to induce a plethora of catabolic events, which are linked to degradation of matrix[70,80], neuronal ingrowth and blood vessel ingrowth into the normally aneural and avascular disc[45,54,90,92]. Inhibition of IL-1 in IVD tissue *in vitro* has been shown to completely abrogate matrix degradation[70] whilst inhibition of TNF alpha had no such effect[70]. IL-1 has also been linked to induction of senescence in articular chondrocytes[93, 94], and fibroblasts[95] all of which are features associated with IVD degeneration **Figure 1**. The importance of IL-1 in the pathogenesis of disc degeneration was furthered by the observation that spontaneous IVD degeneration

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occurred in a knockout mouse model where the natural inhibitor of IL-1 (IL-1Ra) was removed[91], which is further supported by the findings that polymorphisms in the *IL-1* gene cluster increases the risk of IVD degeneration and LBP[96-101]. This improved understanding of the pathogenesis of disc degeneration holds potential to inhibiting further degeneration and creating the correct tissue niche for regenerative approaches.

New Methods for assessing the Lumbar spine;

Although MRI is now used widely for investigating and producing a qualitative image of a patients lumbar spine, new techniques have been developed such as quantitative MRI[102] which allows for measurement of solutes across the IVD over time and also for dynamic MRI using two magnetic coils so that spinal function can be evaluated[103]. Another technique attracting interest is the use of laser Doppler to image the vascular supply to the IVD[104]. These newer quantitative techniques have real potential for measuring changes that occur in patients with LBP.

Surgical treatment

In the beginning of the 20th century the spine was usually operated on for tuberculosis. The method was spinal decompression and posterior spinal fusion as described by Albee (1911)[105], Hibbs (1912)[106] and Girdlestone (1923)[107].

Anterior approach;

In the early part of the 20th century the anterior approach for treating patients with tuberculosis of the spine was considered by many surgeons to be too hazardous. In 1906 Muller reported on his experience of the transabdominal access to the spine in patients with tuberculosis[108]. Muller concluded that the transabdominal approach

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may seem too daring, but was surprised and satisfied with the relatively ease of access and visualisation achieved by this approach[108]. In 1933 Burns used the transabdominal approach on a boy aged 14 years and drilled a hole almost vertically downwards from L5 to the sacrum and inserted a cortical bone graft taken from the patient's tibia[109]. This approach was the first recorded operation of exposing the lumbar spine by the transabdominal approach for a patient with LBP. In 1934 Ito *et al.,* described an operation for the treatment of Pott's disease of the spine and also used an anterior midline approach but proceeded by a retroperitoneal approach to arrive at the lumbar vertebrae[110]. In one of the ten patients, a bone graft from the tibia was inserted into the defect at the lumbosacral level[110]. In 1936 Mercer described the transabdominal approach for treating two patients with lumbar spondylolisthesis using an lilac crest bone graft with unfortunately a fifty percent mortality[111]. Over the next few decades the anterior transabdominal approach to the spine was used mainly for the treatment of tuberculosis of the spine. Hodgson and Stock (1956), showed that the spine could be exposed by the anterior route without causing major problems to the patient[112], especially using the retroperitoneal approach, which gave confidence to surgeons in the anterior exposure of the lumbar spine. **Figure 2**. Cloward(1963) pioneered the interbody dowel technique[113] which Crock developed further using the retroperitoneal approach to the lumbar spine[23].

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Posterior approach;

The posterior approach to the lumbar spine for patients with LBP was championed by Wiltse (1968) who developed the posterolateral fusion technique[114] widely used today. Pedicular screw insertion has enabled rigid fixation to be achieved and enhanced the process of fusion. **Figure 3.** Although the technique of screw insertion needs to be precise and different approaches to screw insertion can lead to different

levels of security of fixation[115]. Equally so the posterior approach may lead to extensive muscle retraction which can cause posterior muscle necrosis[116]. It is also possible that fusion of the lumbar spine could also lead to an increase in the level of degeneration in the adjacent lumbar discs. Mannion *et al* (2014) undertook a long term study on patients who underwent lumbar spinal fusion and concluded that fusion was associated with loss of lower disc space height at the adjacent segment, which occurred on average at 13 years[117]. However they noted that in their study that the spinal fusion had no effect on patient self-rated outcomes. Partly to address this problem dynamic fixation using a posterior dynamic stabilisation technique was developed. **Figure 4.** Zhou *et al* 2013 undertook a systemic review of the literature on dynamic fixation and concluded that posterior dynamic stabilisation did protect the lumbar discs against adjacent segment degeneration[118].

Currently a combination of anterior and posterior fusion is now the standard operation for patients with LBP, particularly spondylolisthesis[119] and is combined with iliac crest bone graft supplemented by Bone Morphogenic Protein[120]. Strube *et al* (2012) compared the results of anterior/posterior lumbar fusion at a single level with anterior fusion alone and reported that an anterior lumbar fusion alone can lead to better clinical results then anterior posterior spinal fusion and they concluded that a standalone single level fusion was adequate[121].

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Multidisciplinary rehabilitation

An alternative to spinal fusion for LBP is an intensive back rehabilitation. Fairbank *et al* (2005) reported in a prospective study on the effective outcome of an intensive rehabilitation programme compared to spinal fusion and showed that there was no difference between two groups of patients as far as spinal function and patient satisfaction were concerned[122]. These same patients in this study were followed up

by Mannion who in 2013 reported that at 11 years there was no difference between the spinal fusion patients and those who underwent intensive rehabilitation[123]. Mannion *et al* concluded that the risks of surgery and the lack of deterioration of the non-operative patients 'outcomes, over time, suggested that lumbar fusion in chronic LBP should not be favoured particularly when multidisciplinary cognitive behavioural and exercise rehabilitation programmes were available[123]. In distinction Wood *et al*(2011)concluded that for a patient with a clearly defined spondylolisthesis there seemed to be a benefit from spinal fusion and in a comprehensive review of the literature reasoned that spinal fusion should be considered for patients with an isthmic spondylolisthesis and also for patients who had failed non operative treatment[124].

Total Disc Replacement.

Total disc replacement (TDR) is an alternative to spinal fusion with the advantage of restoring flexibility to the intervertebral joint. Most current designs of TDR are ball-and-socket joints involving articulation of ultra-high molecular weight polyethylene on metal or metal-on-metal. **Figure 5**. The technology is based on total hip replacement to achieve a low friction joint. More recently, TDR prostheses have been manufactured from flexible polymeric materials to more closely mimic the natural disc[35].

The Norwegian multi centre RCT (2014) reported that TDR was cost effective when compared with multi-disciplinary rehabilitation[125], but with only a two year follow up they stressed that their results needed to be treated with caution. A Cochrane review (2012) on the use of TDR in LBP however could not find any evidence of better outcomes in terms of LBP and patient function between those who had TDR and those who underwent spinal fusion and they also advised caution on adopting TDR

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on a large scale[126,127]. However Siepe *et al* (2014) in a prospective study reporting 5 to 10 year follow up concluded that TDR should be considered as a viable treatment alternative to lumbar fusion[128].

Other Surgical Techniques

Other surgical techniques include minimally invasive techniques such as transforaminal lumbar interbody fusion, which were reviewed by Perez-Cruet *et al*(2014)in a prospective long meta-analysis and they noted that there was a significant clinical improvement in patients with LBP due to IVD degeneration and also in patients with spondylolisthesis[129]. These authors reported very low rates of failure of interbody fusions. Other techniques such as nucleus replacement, interspinous devices and minimally invasive intradiscal thermocoagulation are too new to know of their real outcomes and the evidence of their effectiveness is open to discussion[130,131]

Future potential of Stem Cells

Current proposed treatments, driven by clinical need and scientific research, have aimed to develop a biological therapeutic strategy which addresses the underlying pathogenesis of IVD degeneration. A vast amount of research has focused on the use of cells, either alone, or together with biomaterial scaffolds, in order to repopulate the NP and simultaneously regenerate the matrix. Cell therapy has been used for the repair of articular cartilage for over two decades[132,133] but has only recently been applied to the IVD probably due to the challenges cell therapy faces in the IVD. The use of autologous NP cells would be the ideal strategy, however due to the low cellular abundance in the NP[134], the harvesting of NP cells from more than one disc or *in-vitro* expansion of cells would be required to obtain a sufficient number

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for transplantation. Furthermore autologous NP cells extracted from degenerate discs may be inappropriate for regenerative purposes due to the increased expression of catabolic factors[135], decreased synthesis of normal matrix components and increased senescence displayed by these cells[136], which could explain the limited regenerative potential of cells from herniated disc tissue in comparison to those from the non-herniated regions[137]. Concerns have also been raised regarding the needle puncture which is currently used to extract autologous NP cells, since this method has been shown to accelerate degenerative changes[138,139,140]. Other non-autologous sources of cells have been investigated such as notochordal cells[141-146], which are attractive as disc degeneration is less in animals, where they survive (such as in non-chondrodystrophoid dogs)[147]. Stem cells are an attractive cell source either from autologous or allogeneic sources since large numbers can be harvested from embryos or placenta/umbilical cords or adult tissues such as bone marrow and fat. Of these sources human mesenchymal stem cells (hMSC) are potentially the most attractive cell choice for IVD regeneration since they can be extracted from a variety of adult tissues[148], they have proliferative capacity and thus can be easily cultured and expanded *ex-vivo*, they host the ability to differentiate into multiple cell lineages[149] and finally they also avoid the ethical issues surrounding the use of embryonic stem cells. Recent studies have suggested that the source of MSCs are critical to the clinical outcome with improved expansive and chrondrogenic differentiation potential in synovial membrane and adipose tissue derived MSCs in comparison to bone marrow derived MSCs[148,150]. Increasing evidence has shown that hMSC are able to differentiate into NP like cells and produce a biologically functional NP matrix consisting of proteoglycans and collagen type II[151-154]. Studies have also shown that co-culture of hMSCs and NP cells not only enhances NP cell differentiation, but also restores normal cell function of degenerate NP cells, thus

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aiding the repair process[155,156]. The use of stem cells alone for IVD repair has demonstrated some success, particularly in small animal models. Zhang *et al.,* (2005) reported increased proteoglycan and collagen type II deposition from transplanted bone marrow derived MSCs in rabbit IVD[154]. Orozco *et al.,* (2011) conducted a small human pilot study whereby ten patients with disc degeneration were injected with autologous bone marrow derived MSCs; following 12 months patients reported significant improvements in pain and disability and displayed increased water content in the NP, however disc height was not restored[153]. In contrast the transplantation of allogenic disc derived MSCs into denucleated lumbar discs in an *in-vivo* porcine model exhibited low cell survival and the formation of a collagen type I/II scar tissue following three months[157].

The success of hMSC used alone in IVD repair is varied, but may be improved by the use of an appropriate biomaterial scaffold which could provide additional mechanical support, facilitate cell survival, provide a scaffold for matrix deposition and possibly promote appropriate hMSC differentiation to an NP cell phenotype. In particular the use of thermo-reversible hydrogels which solidify at body temperature are an attractive choice for IVD repair since they provide a non-invasive cell delivery method for NP tissue regeneration[158]. The ideal load bearing, injectable biomaterial scaffold for IVD regeneration, which meets all requirements in terms of biocompatibility, bioactivity, mechanical properties and injectability, is yet to be produced, however increasing research into the development of hydrogels has produced a selection of scaffolds which have demonstrated success in terms of MSC regeneration of the NP. Richardson *et al.,* (2008) cultured MSCs for 4 weeks seeded in a temperature-sensitive hydrogel chitosan–glycerophosphate (C/Gp) hydrogel and demonstrated differentiation of MSCs to an NP cell phenotype, with production of an

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NP like matrix consisting of proteoglycans and collagens[151]. More recently Peroglio *et al.,* (2013) investigated *in-vitro* and *ex-vivo* hMSC differentiation in a hyaluronan-poly(*N*-isopropylacrylamide) (HA-pNIPAM) hydrogel under hypoxic conditions and reported successful discogenic differentiation with production of an NP like matrix[159]. The majority of studies have aimed to regenerate the NP since this is the main region which undergoes significant cellular and matrix changes during degeneration of the IVD[135,160]. Despite this, biological strategies which address the entire IVD may be required, particularly in severely degenerate cases where tears in the AF may result in extrusion of the implanted biomaterial and/or cells **Figure 6**. Consequently a variety of studies have investigated biphasic scaffolds which aim to repair the AF and NP simultaneously[161-165]. Lazenbnik *et al.,* (2011) used a biomimetic strategy by combining porcine chondrocyte cell seeded agarose gel surrounded by cell seeded electrospun polycaprolactine fibres and observed that cells were viable, well distributed and orientated themselves in the direction of the fibres[166]. Furthermore the biphasic scaffolds were found to have higher moduli than agarose gels alone, thus providing enhanced mechanical properties which may be required to restore normal IVD biomechanics in severe stages of degeneration[166]. However in IVDs where endplate calcification has occurred which could result in decreased nutritional supply and poor cell survival tissue engineering a complete IVD with all three of its components (NP; AF; CEP) may be required[167] **Figure 6**. Such a tissue engineered construct would compete with allogeneic disc transplants which have been pioneered by Ruan *et al.,*(2007), who implanted cadaveric cervical discs into patients with cervical IVD degeneration with encouraging clinical outcome and minimal loss of disc height at five year follow-up[168]. Despite increasing evidence demonstrating significant potential for MSC based regenerative strategies, there are still questions which remain to be answered, including: what is the ideal cell type and source? is

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the use of a biomaterial scaffold necessary? at what stage of degeneration is regenerative cell therapy clinically applicable? Could cells be administered as part of an intervertebral 'organ transplant'?[35].

Furthermore, assessing the regenerative capacity of MSCbased strategies to produce an appropriately biological functioning tissue is obstructed by a lack of understanding of the normal NP cell phenotype. The majority of studies, including those which have been discussed here, characterise NP cell differentiation using traditional chondrogenic genes such as aggrecan, collagen type II and sex determining region box 9(Sox-9)[169,170]. Despite this, cells and tissues of articular cartilage and NP demonstrate significant differences in terms of morphology, ECM deposition and biomechanical behaviour [171], thus elucidating a differential NP marker which can be used to inform and thoroughly assess MSC differentiation into biologically functioning NP cell is the current focus of many research studies[172,173,174].

Despite all the challenges and unknowns associated with cell therapy it has been used in the clinic in several centres. A clinical trial for treating herniated IVD following culture expansion of the resident cells reported a reduction in the patient's pain level with no loss of disc height[175,176]. Autologous MSCs have also been used to treat patients, either within a collagen sponge implanted percutaneously[177] or injected directly into the discs[137]. There remain, however, important issues to be addressed around cell based therapies for patients with discogenic LBP due to IVD degeneration. Central amongst which is whether a normal functioning CEP is required for treatment to be successful in order for IVD nutrition to be maintained[178,179]. Although recent data suggest that nutritional supply actually increases rather than decreases during disc degeneration[57,180].

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The Future

At the moment posterior interbody spinal fusion for treating patients with LBP who have failed conservative treatment for at least 3 months or who are unable to respond to a multi-disciplinary rehabilitation programme is the best option for treating the pain and improving the patients overall function. Certainly in patients with an isthmic spondylolisthesis and probably with degenerative spondylolisthesis, surgical treatment after failed conservative treatment is the appropriate treatment. Noshchnenko *et al* (2014) in a systemic meta-analysis concluded that spinal fusion of the lumbar spine is an effective treatment for patients with severe chronic LBP 181. However it is worth noting that the NHS commissioning Board for England recently stated that they would not commission spinal surgery for the treatment of chronic non-specific LBP 182. In the UK, total disc replacement has undergone a decline in use although it is interesting to note that sacroiliac joint fusion has risen, even though the long term results for this treatment are largely unknown[183].

It would seem therefore that the original operations devised primarily for treating tuberculosis, namely spinal fusion either anteriorly or posteriorly or combined, have contributed significantly to the management of patients with chronic LBP due to disc degeneration, especially those who have failed or are unable to complete a multidisciplinary rehabilitation programme.

The future must, however, be to focus on alternative ways for treating chronic recurrent LBP which is where stem cell biotechnology may have the answers. Over the past two decades considerable research efforts have been focused on the development of biological strategies for IVD repair. In particular strategies to inhibit the pathogenesis of disc degeneration, particularly catabolic factors together with the

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use of MSCs in combination with biomaterial scaffolds have shown significant promise in the regeneration of an NP like matrix and even a whole IVD. Despite this, there are still challenges emerging which must be overcome before such strategies can be incorporated into clinical practise. Biomaterial research is critical to the continuing development of a bioactive and biocompatible material which has mechanical properties that replicates the natural disc and can be delivered via non-invasive injection prior to *in-situ* solidification. Research to define the pathological state of degeneration as well as understanding the normal NP phenotype is also critical to ensure regeneration of the correct biologically functioning NP tissue. Furthermore the safety and efficacy of regenerative therapies must be thoroughly assessed *in vivo* in the degenerate disc microenvironment. Progress so far has shown future potential for the use of MSCs, either alone or in combination with monophasic or biphasic biomaterial scaffolds offering a selection of regenerative strategies that can be chosen depending on the stage of IVD degeneration, providing a less invasive and more efficacious alternative to surgery. If the possibilities outlined in this paper can be transferred to clinical practise and if the patient outcomes show real improvement then those treating patients with LBP due to disc degeneration will have come a long way from the seminal paper by Mixter and Barr in 1934[184].

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Figure Legends;

Figure 1: Cellular and matrix components of the non-degenerate disc and their balance during normal homeostasis and the histological, biochemical, cellular changes which occur during degeneration.

Figure 2: Radiograph of the lumbar spine showing

a. cortico-cancellous bone graft as originally described by Mercer inserted between L4-5.
b. Showing two cages inserted between the L4-5 and L5-S1 levels.

Figure 3: Rigid fixation and bone graft in a patient with a degenerative L4-5 disc.

Figure 4: A radiograph showing dynamic fixation of the L4-5 level.

Figure 5: Radiographs showing a total disc replacement at the L3-4 level.

Figure 6: Potential treatment strategies for disc degeneration are likely to be tailored to the stage of degeneration dependant on the tissues which require repair. Mild degeneration treatments are likely to target purely the nucleus pulposus (NP), whilst mid degeneration where annular tears are present biphasic regeneration of the NP and annulus fibrosus (AF) are likely to be required and severe stages where the cartilaginous end plate (CEP) is compromised may require a full disc replacement.

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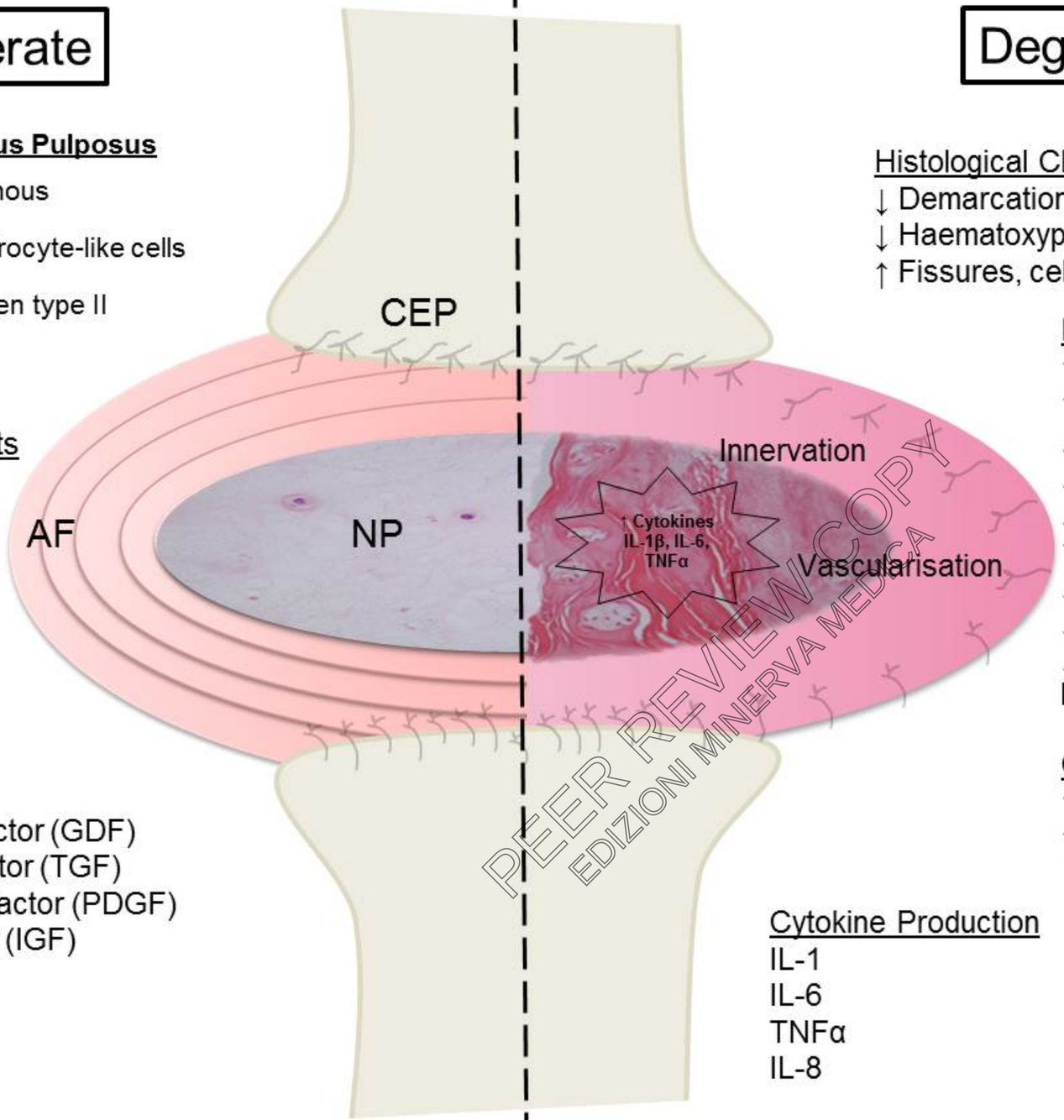
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Non-Degenerate

Annulus Fibrosus	Nucleus Pulposus
Fibrous	Gelatinous
Fibroblast-like cells	Chondrocyte-like cells
Collagen type I	Collagen type II

Normal Matrix Components
- Aggrecan
- Collagen II
- Highly hydrated
- ↑ Resistance to load

Growth Factors
- Growth differentiation factor (GDF)
- Transforming growth factor (TGF)
- Platelet derived growth factor (PDGF)
- Insulin-like growth factor (IGF)



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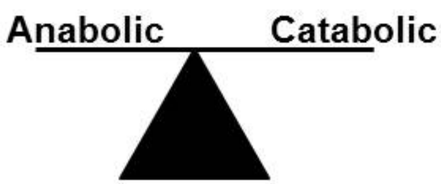
Degenerate

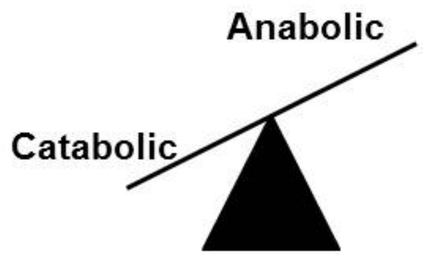
Histological Changes
- ↓ Demarcation between AF & NP
- ↓ Haematoxyphilia
- ↑ Fissures, cell clusters & infiltrating cells

Biochemical & Matrix Changes
- ↑ MMP's
- ↑ ADAMTS'
- ↓ Matrix synthesis
- ↓ Aggrecan = ↓ Hydration
- ↓ Collagen type II
- ↑ Collagen type I
- ↓ Elastin
- ↓ Chondromodulin
- ↓ Ability to withstand compressive loads

Cell viability
- ↑ Apoptosis
- ↑ Senescence

Cytokine Production	Chemokine Production
IL-1	CCL2
IL-6	CCL7
TNFα	IL-16
IL-8	

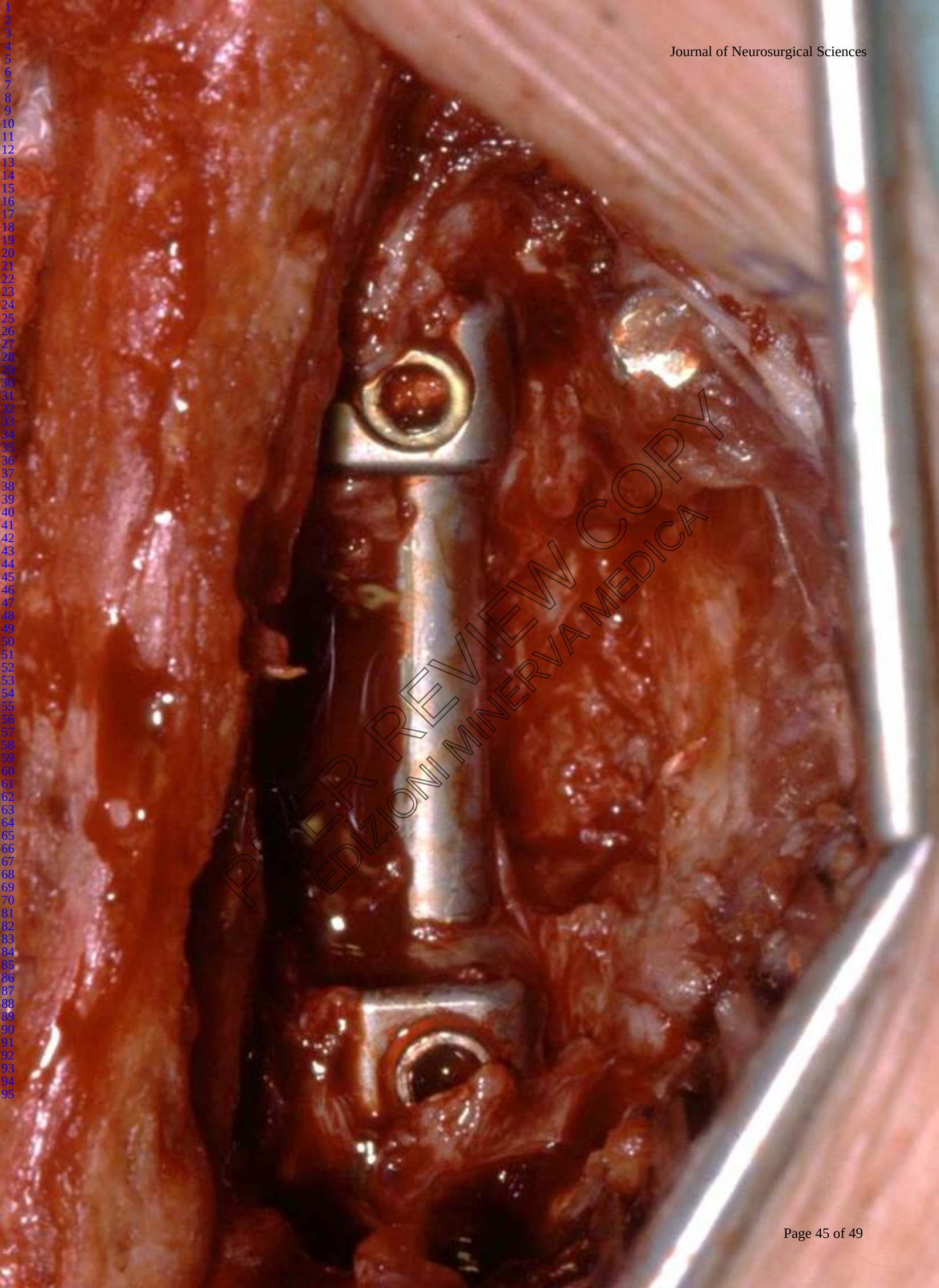






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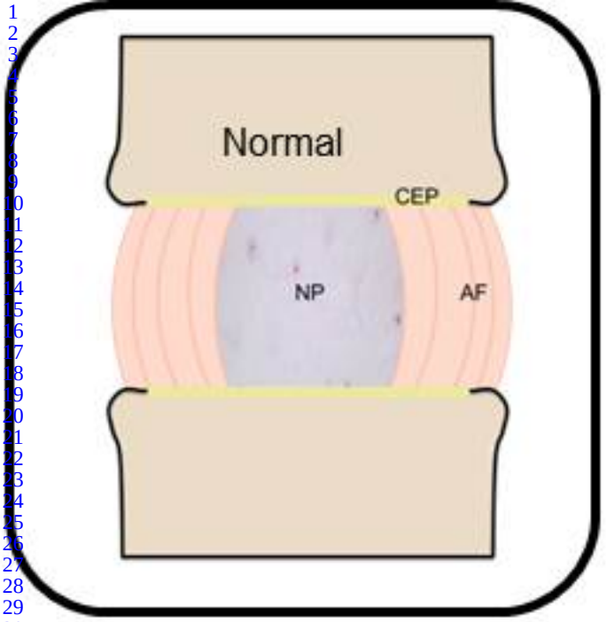


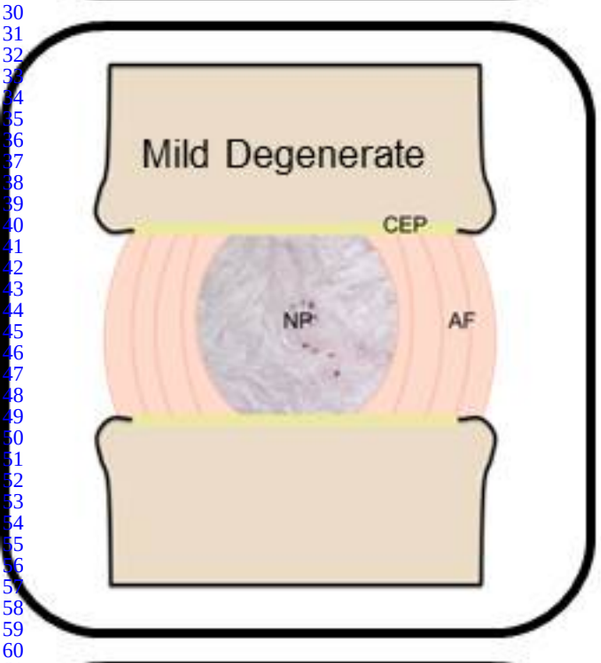
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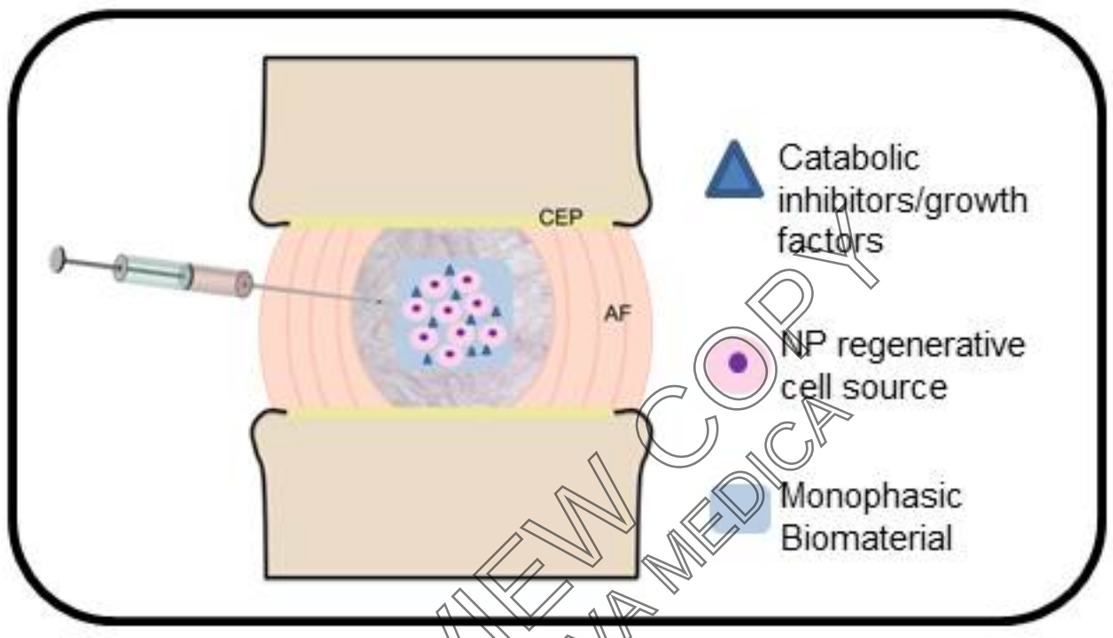
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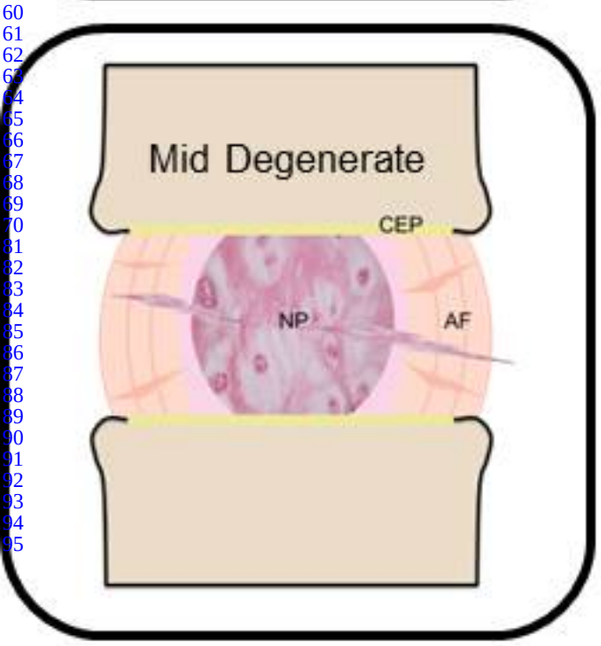
Treatment strategy for mild degeneration: NP repair

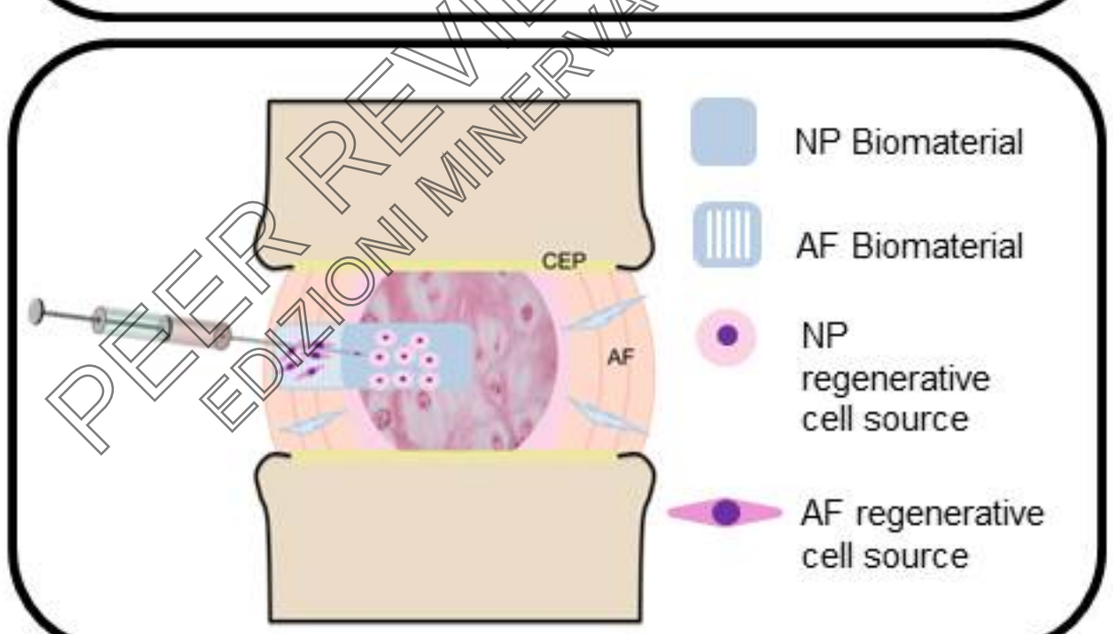
Inhibitors of degeneration

Growth Factors

Regenerative cell source.

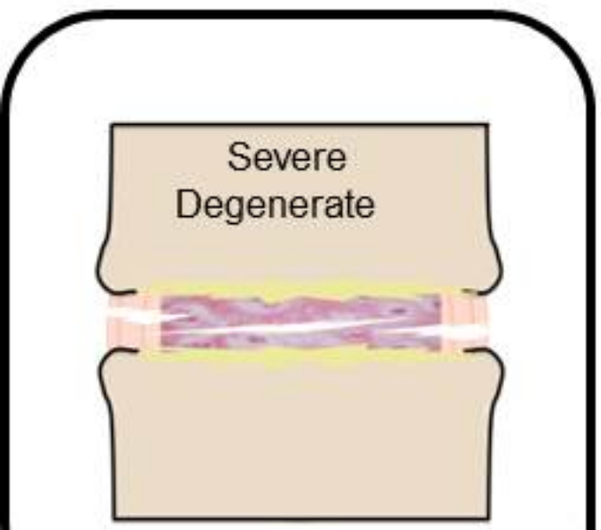
Monophasic biomaterial

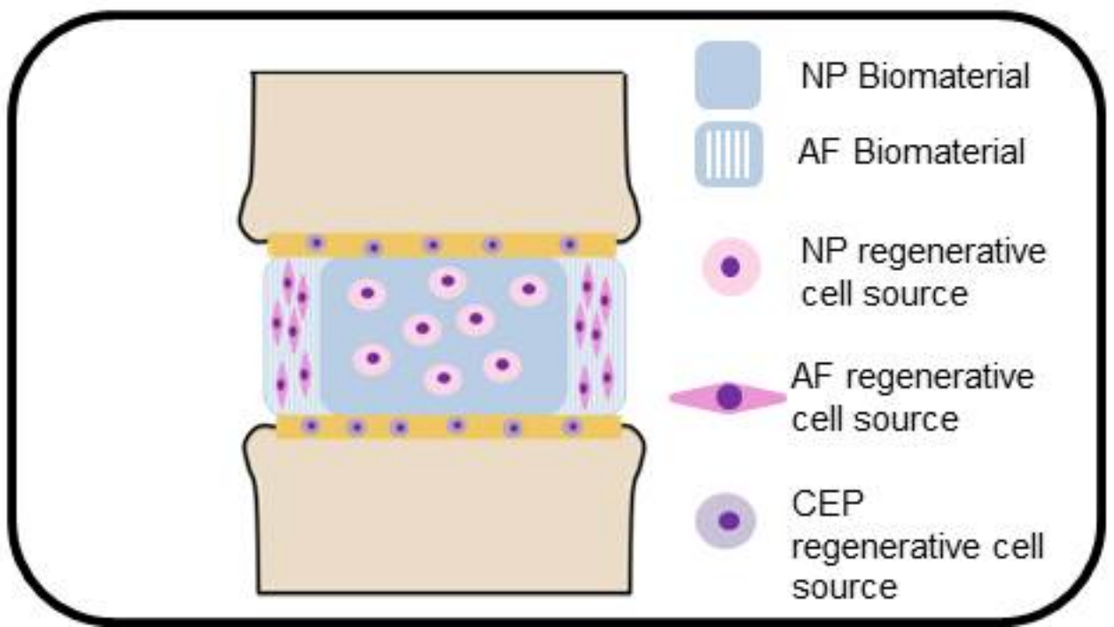




Treatment strategy for mid degeneration: NP and AF repair

Biphasic biomaterial scaffolds with regenerative cell source:





Treatment strategy for severe degeneration: NP, AF, CEP

Acellular or cellular tissue engineered whole disc replacement.

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