Osteonecrosis of the femoral head: treatment with ancillary growth factors

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Abstract Osteonecrosis (ON) of the femoral head, also known as avascular necrosis (AVN) of the femoral head, is a progressive disease that predominantly affects younger patients. During early stage of ON, decompression of the femoral head has been commonly used to improve pain. The decompression has been augmented with nonvascularized or vascularized bone grafts, mesenchymal stems cells, and growth factors. The use of adjuvant growth factors to supplement the core decompression has mainly been limited to animal models in an attempt to regenerate the necrotic lesion of ON. Factors utilized include bone morphogenetic proteins, vascular endothelial growth factors, hepatocyte growth factors, fibroblast growth factors, granulocyte colony-stimulating factors, and stem cells factors. In animal models, the use of these factors has been shown to increase bone formation and angiogenesis. Although promising, the use of these growth factors and cell-based therapies clinically remains limited.

Keywords Osteonecrosis · ON · Avascular necrosis · AVN · Growth factors · Bone morphogenic protein · BMP

Introduction

In the USA, osteonecrosis (ON) of the femoral head, also known as avascular necrosis (AVN) of the femoral head, occurs in up to 20,000 new patients every year, predominantly in the younger than 40 years age group [1–3]. ON of the femoral head occurs when the osteocytes of the trabecular and subchondral bone die, often leading to collapse of the femoral articular surface [4, 5]. Currently, it is not clear what leads to osteocyte death. However, once collapse has occurred, patients often require a total hip arthroplasty (THA) for pain relief and improvement in daily function [4, 6–8].

Staging

The treatment of ON is dependent on accurate disease staging. The classification system by Ficat is most commonly used and based on radiographs of the pelvis [5]. The presence of a “crescent sign” signifies fracture in the subchondral bone, indicating the first stages of collapse [5]. Similar to the Ficat classification, the Steinberg classification also uses the crescent sign to separate early- from late-stage ON [9]. Unlike the Ficat classification, the Steinberg classification uses magnetic resonance imaging (MRI) in addition to radiographs to increase sensitivity and specificity, making it the preferred system at our institution [10–12].

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Treatment options

Treatment modalities for ON can be used in two main categories of disease: precollapse and postcollapse. If the disease process is halted in the precollapse phase, patients can avoid THA and other salvage-type procedures. This group encompasses stages 0 to 2 of the Steinberg classification. Core decompression, with or without the addition of bone marrow or bone graft, is one treatment option for precollapse ON of the femoral head. For the purpose of this review, we will focus on the use of adjuvant growth factors, mainly in conjunction with a core decompression procedure.

Adjuvant growth factors

Bone regeneration is modulated by various cellular mediators including bone morphogenetic proteins (BMP), angiogenic growth factors, interleukins, and cytokines, all of which have been used to treat bone defects [13–16]. It is thought that through use of these agents alone or in combination with an operative procedure, healing of ON lesions can be enhanced (see Tables 1 and 2).

Bone morphogenetic proteins

Bone morphogenetic proteins (BMPs) are a family of proteins that promote osteogenesis through new bone formation, vascularization, and cartilage repair [17]. BMP-2, BMP-4, BMP-7, and BMP-9 are part of the transforming growth factor-beta (TGF- β) superfamily and act to accelerate the differentiation of mesenchymal stem cells (MSCs) into bone [18]. Furthermore, it has been shown that expression of BMP is decreased in the setting of ON of the femoral head [19]. Numerous human and animal studies have shown the osteogenic activity of BMPs through differentiation of MSCs into osteoprogenitor cells for the treatment of ON [17, 19, 20•, 21, 22, 23•, 24, 25•, 26]. Although promising, it is felt that the method for applying and delivering the BMP still needs to be modified [25•]. This is because during the healing process, supplementary BMP increases vascularity in the outer necrotic lesion; however, the central portion remains fibrous.

Animal studies investigating bone morphogenetic proteins for osteonecrosis treatment

The local administration of BMPs has been fraught with complications related to the high concentration needed to induce

Table 1 Animal studies treating ON of the femoral head with growth factors

Growth factor	Procedure	Effect
BMP [21, 23•, 24, 27, 30–32]	<ul style="list-style-type: none"> • Trapdoor bone grafting • Core decompression • Intraarticular injection 	<ul style="list-style-type: none"> • Prevention of collapse • Increased biomechanical strength • Radiographic and histologic healing of necrotic lesions
HGF [44, 45•]	<ul style="list-style-type: none"> • Core decompression • Transfected MSCs 	<ul style="list-style-type: none"> • Prevention of collapse • New bone formation • Neovascularization • Increase vascular growth factors
VEGF [48–51]	<ul style="list-style-type: none"> • Core decompression • Transfected local cells • Plasmid/collagen • Transfected MSCs 	<ul style="list-style-type: none"> • New bone formation • Neovascularization • Increase vascular growth factors • Protection of necrotic bone from collapse
HIF-1 α [58]	<ul style="list-style-type: none"> • Core decompression • Transfected MSCs 	<ul style="list-style-type: none"> • Increased VEGF expression • New bone formation • Neovascularization
FGF [60]	<ul style="list-style-type: none"> • Plasmid/collagen 	<ul style="list-style-type: none"> • Neovascularization • New bone formation
G-CSF/SCF [61, 62•]	<ul style="list-style-type: none"> • Subcutaneous injection 	<ul style="list-style-type: none"> • Neovascularization • New bone formation • Healing of necrotic lesion • Prevention with pretreatment

BMP bone morphogenetic protein, *HGF* hepatocyte growth factor, *MSC* mesenchymal stem cells, *VEGF* vascular endothelial growth factor, *FGF* fibroblast growth factor, *G-CSF* granulocyte colony-stimulating factor, *SCF* stem cell factor

Table 2 Clinical studies treating ON of the femoral head with growth factors

Study	Growth factor	Procedure	Outcomes
Lieberman et al. [20•]	Human BMP	<ul style="list-style-type: none"> • Core decompression • Fibular allograft 	<ul style="list-style-type: none"> • Prevention of collapse in patients with smaller necrotic lesions • Progression in large lesions in the weight bearing surface
Sun et al. [25•]	rhBMP	<ul style="list-style-type: none"> • Trapdoor procedure • Synthetic bone grafting 	<ul style="list-style-type: none"> • Improved clinical outcomes with rhBMP • Improved outcomes in precollapse vs. postcollapse ON
Seyler et al. [40]	BMP-7	<ul style="list-style-type: none"> • Trapdoor procedure • Autogenous bone grafting • Bone marrow 	<ul style="list-style-type: none"> • Higher rates of collapse with large lesions • Improvement in Harris Hip Scores • 80 % survival for small lesions
Martin et al.[41•]	Human PRP	<ul style="list-style-type: none"> • Core decompression • Concentrated bone marrow 	<ul style="list-style-type: none"> • Pain relief in 86 % of patients • Progression of ON in 21 % of patients

BMP bone morphogenetic protein, *rhBMP* recombinant bone morphogenetic protein, *PRP* platelet-rich plasma

bone formation with resultant heterotopic bone formation [27–29]. In addition to using BMP alone, it has been used in conjunction with surgical modalities such as core decompression [30, 31] and “trapdoor” bone grafting [21]. Mazières showed that the administration of recombinant human BMP (rhBMP) with core decompression of the femoral head led to improvements in healing based on radiographic and histological analyses [30]. Likewise, in a study by Pan and colleagues, an improvement in the healing of necrotic lesions of rabbit femoral heads by augmenting core decompression with a poly(D,L-lactide-co-glycolide) (PLGA) and rhBMP scaffold was noted [31]. The authors affirmed that the PLGA/rhBMP scaffold could provide a synthetic bone graft for the treatment of ON of the femoral head [31]. In a sheep model, Simank and colleagues used a collagen scaffold containing BMP-2 and found improvements in healing of the ON lesion when combined with core decompression compared to decompression alone [32]. Mont and colleagues combined BMPs with bone graft following debridement of the ON lesion through a trapdoor approach in dogs [21]. The authors noted healing of the defect and preservation of the subchondral bone and biomechanically superior femoral heads compared to those without BMP at 4 months following the procedure [21].

Although BMP is an active inducer of bone formation, it is thought that the concomitant administration of anti-resorptive agents, such as a bisphosphonates, will assist in preserving the area of necrosis and prevent the collapse of the femoral head [33, 34]. Bisphosphonates have been used for the treatment of ON and have had efficacious results in clinical trials in preventing collapse. Kim and colleagues [23•] and Vandermeer and colleagues [27] have shown that the co-addition of a systemic bisphosphonate and local BMP-2 prevents collapse, however, the rate of heterotopic bone formation can be high [23•, 27]. In both of these studies, however, the authors injected the area of necrosis through the hip joint [23•, 27]. It is thought that using an approach similar to a core decompression could mitigate the formation of heterotopic ossification.

Due to the short half-life and heterotopic ossification seen with locally administered BMP, studies have examined the use of gene therapy to increase BMP production at the site of bone defects and spinal fusions [15, 35–37]. In a goat model for ON, Tang and colleagues were able to show the formation of lamellar bone using a β -tricalcium phosphate (β -TCP) scaffold loaded with BMP-2 gene-modified MSCs [22]. The authors noted new bone formation in the pores of the β -TCP scaffold, with no collapse of the femoral head. Furthermore, the resultant compressive strength approached that of a normal femoral head in the goats treated with scaffold [22].

Along the same lines, various carriers for BMP have been proposed in order to provide a graft that is both osteoconductive and osteoinductive. This would provide a scaffold allowing for ingrowth of bone and also blood vessels, while recruiting MSCs to the area to form bone [24]. Various scaffolds composed of PLGA, collagen hydroxyapatite, and β -TCP have been used to “carry” BMP-2 and release it over a sustained period of time [22, 24, 31, 32, 38, 39]. In a study by Wang and colleagues, an improvement in healing of necrotic segments of bone with controlled release of BMP was noted [24]. The authors were able to determine that a continued release of BMP over a 1-week time period led to replacement of necrotic bone with new bone through creeping substitution and neovascularization of the necrotic segment [24].

Clinical studies investigating bone morphogenetic proteins for osteonecrosis

Similar to the animal studies previously listed, BMP has also been used in human clinical studies, typically in combination with other surgical modalities [20•, 25•, 40]. Lieberman and colleagues first examined the use of BMP in conjunction with core decompression and cortical strut allografts [20•]. In this study of 15 patients, 3 failed treatment and eventually required definitive management with THA [20•]. The success of this procedure was based on the amount of necrotic head involvement of the weight-bearing surface of the femoral head. No patient required THA if the necrotic lesion was less than one

third the weight-bearing surface, and only one patient failed if the lesion was between one third and two thirds of the weight-bearing surface [20•].

Seyler and colleagues combined BMP-7 with synthetic bone graft and decellularized bone matrix to treat 21 hips with ON through a trapdoor procedure [40]. In this study, however, the authors only noted a survival of 67 % [40]. Similar to the Lieberman study, the authors noted a worse survival for larger ON lesions and those on the weight-bearing surface, with seven of nine hips with large lesions progressing to collapse [40]. No further surgery was required for 25 of 30 small- and medium-sized lesions (80 %). The authors of this study concluded that the addition of BMP-7 did not improve the success of nonvascularized bone grafting [40]. In a similar study looking at the use of a trapdoor procedure, bone grafting, and rhBMP-2, Sun and colleagues noted greater survival of the femoral head of 82 %, with improved survival over control patients where rhBMP-2 was not used [25•].

Angiogenic factors

The characteristic feature of ON is the avascular nature of the lesion. Numerous growth factors have been investigated in order to increase lesion vascularity through stimulation of angiogenesis. A majority of these studies have focused on gene therapy and are limited to animal models. However, the addition of platelet-rich plasma (PRP) to bone marrow concentrate has been recently described [41•]. The PRP adds additional growth factors such as vascular endothelial growth factor (VEGF), PDGF, and fibroblast growth factors (FGF) which may increase the rate of healing of these lesions [41•]. Although PRP supplied these additional factors, 21 % of the patients eventually required THA [41•].

Hepatocyte growth factor

Hepatocyte growth factor (HGF) is an endothelium-specific growth factor which increases angiogenesis and is the most potent stimulator of endothelial cellular growth [42–44]. Regenerating femoral head vascularity is thought to be essential for healing osteonecrotic lesions. Thus, the use of a potent angiogenic agent would be ideal for stimulating this process. Based off this idea, Wen and colleagues used gene therapy to induce HGF protein production in MSCs and then transferred these cells into an area of necrosis in rabbit femoral heads through core decompression and fibrin glue [44]. The authors found that in rabbits with early stages of ON, they were able to recover the femoral head and prevent collapse and disease progression [44]. New bone formation and neovascularization was noted only in rabbits where the HGF MSCs were injected into the necrotic lesion [44]. Wen and colleagues followed up this study investigating HGF-transfected MSCs for healing traumatic ON [45•]. The authors found that through

transduction of HGF, areas of traumatic necrosis were able to recover and increase the expression of vascular growth factors [45•].

Vascular endothelial growth factor

Although not as potent as HGF, VEGF regulates cellular events which lead to angiogenesis. As such, it has also been investigated for the treatment of ON of the femoral head where avascularity predominates [46]. Likewise, it has been previously shown that ON of the femoral can be induced by blocking the VEGF receptor with an antibody [47]. Yang and colleagues were able to transfect cells in the avascular lesion with a VEGF gene in a rabbit model [48]. They noted a significant increase in angiogenesis and bone formation compared to lesions that did not receive gene therapy [48]. The authors also showed similar results when mixing the VEGF plasmid with collagen and implanting the mix into necrotic segments [49].

In a similar study using rabbits, Cao and colleagues implanted a bone matrix loaded with a recombinant plasmid for VEGF through a core decompression in necrotic lesions of the femoral head [50]. The authors found that animals with the transfected VEGF had increased collagen type I expression, bone formation, and capillary formation [50]. Similarly, Liu and colleagues used an adenovirus vector to increase the expression of VEGF in a rabbit model of ON [51]. They noted increased blood vessel and bone formation in the rabbits transfected with the viral vector compared to animals without the increased VEGF expression [51]. The new bone formation was found to protect the areas of necrotic bone, and in areas where new bone formation did not occur, the necrotic bone was readily resorbed [51]. Hang and colleagues used gene therapy to increase VEGF expression in bone marrow-derived MSCs and found that, in a dog model, the animals with the increased VEGF expression had increased capillary and new bone formation compared to animals treated with stem cells or core decompression alone [52].

Hypoxia-inducible factor-1 α

Hypoxia-inducible factor-1 α is a mediator to the cellular response to hypoxia and has been found to enhance the capacity for stem cells to produce bone and increases angiogenesis in vitro [53, 54•, 55]. It is known that hypoxia-inducible factor-1 α (HIF-1 α) is upregulated and increases the expression of other pro-angiogenic factors in the setting of ON, and as such, it is thought that the increased production of HIF-1 α could lead to healing of the necrotic lesions [56, 57]. In a rabbit model for ON, Ding and colleagues used a viral vector to increase the production of HIF-1 α by bone marrow-derived MSCs implanted at the time of a core decompression [58]. The authors noted that through the increased production of

HIF-1 α , there was an increase in the expression of VEGF and increased osteogenesis [58]. This translated into the rabbit model, where the increased HIF-1 α led to increased new blood vessel volume and bone formation in the necrotic lesion [58].

Fibroblast growth factors

FGF increase the vascular endothelial cells and stimulate angiogenesis even when given in small amounts [59]. Similar to studies examining the use of plasmids to increase the expression of VEGF, recombinant plasmids increasing the expression of FGF have been used to treat areas of ON of the femoral head [60]. Yang and colleagues noted an increase in angiogenesis as well as an increase in new bone formation, accelerating repair of the necrotic lesion [60].

Granulocyte colony-stimulating factor and stem cell factor

Granulocyte colony-stimulating factor (G-CSF) and stem cell factor (SCF) have been used in combination in rabbit models of steroid-induced ON of the femoral head [61, 62•]. G-CSF and SCF both increase proliferation and survival of MSCs and have been shown to “hone” MSCs into an area of necrosis to increase angiogenesis [61, 62•, 63]. An initial study by Wu and colleagues showed increased angiogenesis and blood vessel volume in the region of ON with subcutaneous injection of G-CSF and SCF [61]. The increased vascularity subsequently led to new bone formation and healing of the necrotic lesion [61]. In a follow-up study, Wu and colleagues noted that pretreatment of G-CSF and SCF could prevent the formation of corticosteroid-induced ON of the femoral head [62•]. The authors noted a significant decrease in apoptosis through the inhibition of caspase-3 [62•].

Limitations of ancillary factors

The use of growth factors, especially the BMPs, has been extensively investigated, and clinical trials have shown improvement in rates of spinal fusion and long bone fracture healing [29, 64, 65]. One of the major drawbacks of using recombinant factors has been the need to implant high doses of the recombinant factor to achieve the desired results, at a very high financial cost. The off-label use of recombinant BMP has led to complications including local edema, resorption of surrounding bone, and ectopic bone formation, as seen in multiple animal models in the setting of ON of the femoral head [23•, 27]. Likewise, studies have shown that cell-mediated gene therapy approaches have been found to be more efficient at delivering growth factors [36, 37, 66]. Although vector-mediated cell-based therapy has shown promise in animal models, the transition to clinical trials is potentially limited in a nonlife-threatening disease such as ON due

to the complications previously seen with their use, including death and lymphoproliferative disorders [67–69].

Conclusion

ON of the femoral head is a complex, multifactorial medical condition. The exact pathophysiology behind ON remains to be elucidated. Currently, the use of ancillary growth factors for the treatment of this disease process attempts to treat ON at a pathophysiological level. Although promise has been observed in several animal models, the clinical use of adjuvant growth factors remains investigational.

Compliance with Ethics Guidelines

Conflict of Interest Matthew T. Houdek and Cody C. Wyles declare that they have no conflict of interest.

Rafael J. Sierra reports personal fees from Biomet.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

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