Journal of Orthopaedic Translation (2015) 3, 58—70

Available online at www.sciencedirect.com

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journal homepage: http://ees.elsevier.com/jot

REVIEW ARTICLE

Steroid-associated osteonecrosis: Epidemiology, pathophysiology, animal model, prevention, and potential treatments (an overview)

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Received 23 June 2014; received in revised form 30 November 2014; accepted 23 December 2014
Available online 13 January 2015

KEYWORDS
osteonecrosis;
pathophysiology;
prevention;
steroid;
treatment

Summary Steroid-associated osteonecrosis (SAON) is a common orthopaedic problem caused by administration of corticosteroids prescribed for many nonorthopaedic medical conditions. We summarised different pathophysiologies of SAON which have adverse effects on multiple systems such as bone marrow stem cells (BMSCs) pool, bone matrix, cell apoptosis, lipid metabolism, and angiogenesis. Different animal models were introduced to mimic the pathophysiology of SAON and for testing the efficacy of both prevention and treatment effects of various chemical drugs, biological, and physical therapies. According to the classification of SAON, several prevention and treatment methods are applied at the different stages of SAON. For the current period, Chinese herbs may also have the potential to prevent the occurrence of SAON. In the future, genetic analysis might also be helpful to effectively predict the development of ON and provide information for personalised prevention and treatment of patients with SAON.

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http://dx.doi.org/10.1016/j.jot.2014.12.002

Osteonecrosis

Osteonecrosis (ON) is a disabling clinical disease characterised by death of the osteocytes and the bone marrow, followed by resorption of the necrotic tissues and formation of new but weaker osseous tissue which leads to a progressive destruction of bone architecture, subchondral fracture, and collapse of joints, mostly occurring at the femoral head, and finally loss of joint function [1—3]. Alexander Munro first reported this medical condition in 1738 and Cruveilhier firstly attributed this disorder to an aberration of circulation in the femoral head in the mid 1800s [4]. Similar terms to ON include avascular necrosis (AVN), ischemic necrosis, subchondral AVN, aseptic necrosis of bone, or osteochondritis dissecans. AVN or ischemic necrosis is used to describe their vascular aetiology, while aseptic necrosis is used to indicate that the infection does not play an important role in ON occurrence and its development. ON occurs mostly in large weight-bearing joints, such as the hip (Fig. 1) that may subsequently develop to joint collapse and end up with joint replacement surgery.

ON can be classified into two types: one is idiopathic (primary) ON with no apparent aetiologic or risk factor(s); another is secondary ON in which the aetiology is clearly identifiable [1], including traumatic and nontraumatic ON.

Steroid-associated ON

Steroid-associated ON (SAON) is a common nontraumatic ON caused by use of steroids, which are initially prescribed for many nonorthopaedic medical conditions, including systemic lupus erythematosus (SLE), organ transplantation, asthma, rheumatoid arthritis, and severe acute respiratory syndrome (SARS). ON in large joints caused by corticosteroid administration is usually associated with the worst prognosis due to the degeneration of the bone around the prosthesis. Therefore, SAON is the focus of this review paper where we aim at facilitating multidisciplinary collaborations of medical doctors and research scientists working on SAON.

Aetiology

Of all the nontraumatic factors, corticosteroid usage is the major associated one [5]. ON development was associated with dose and duration of administrating corticosteroids [6]. Usually, the chronic administration of high-dose steroids will result in ON, but it is very difficult to predict which patient under steroid administration will finally develop ON. Dynamic magnetic resonance imaging (MRI) was a promising method for potentially better prediction in an SAON rabbit model [7] and human trials shall be conducted to confirm the value of SAON development already in Phase 0. A retrospective study showed that the interval between steroid administration and the ON onset was from 6 months to $>$ 3 years. The prospective study using MRI to detect early ON of the femoral head showed that the initial changes of necrosis were found at about 3 months after administration of steroids.

Epidemiology

Steroids are now the second most common cause of ON after trauma and the prevalence of ON varies between 3% and 38% [3]. Five to twenty-five percent of patients with

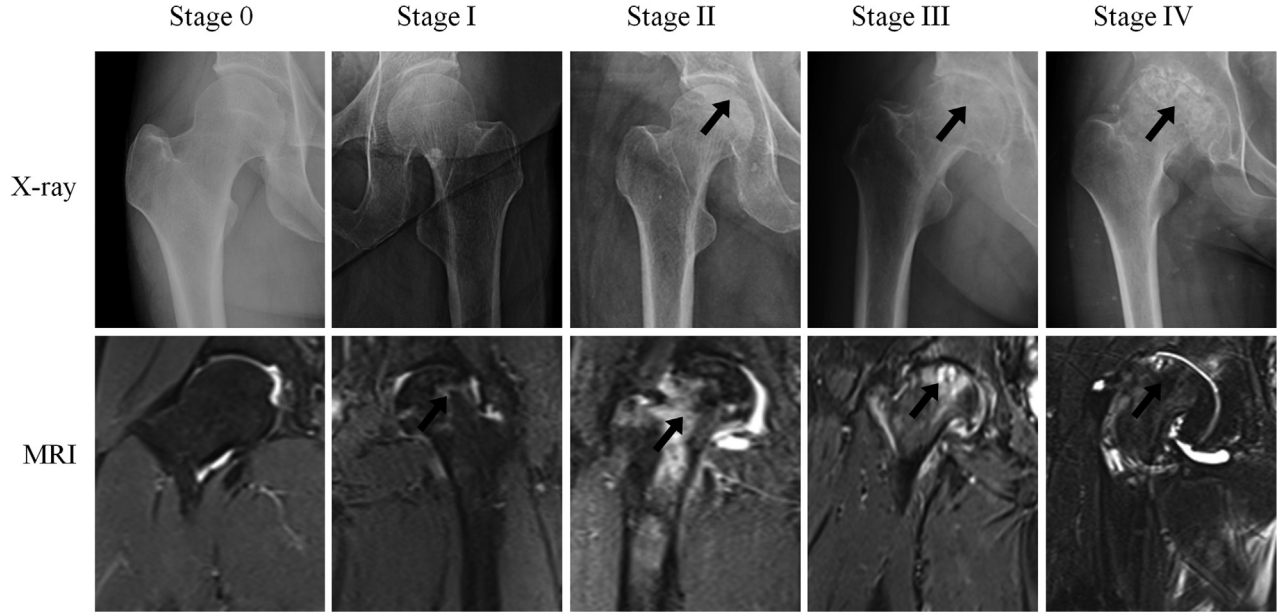

Figure 1 Five stages of osteonecrosis (ON) from X-ray and magnetic resonance imaging (MRI) according to the Association Research Circulation Osseus (ARCO) staging system on the femoral head. At Stage I, there are no visible changes on X-ray but demonstrable on MRI. At Stage II, there are some radiographic changes in the femoral head, but the joint surface maintains its integrity. As the disease progresses, the surface of the femoral head begins to collapse and finally the integrity of the joint is destroyed (Stage III and Stage IV).

atraumatic ON are a result of steroid administration and 5% of patients with a history of high-dose corticosteroid administration developed ON. In Hong Kong and the region, up to 20% ON incidence was reported following treatment of SLE patients, patients with organ transplantation, and SARS patients with steroids. The ON appearances on MRI in patients under steroid treatment were the same as those caused by other aetiologies or conditions. It was reported that at least one ON lesion was found in 31% (138/448) of SARS patients after steroid therapy from Beijing Jishuitan Hospital, China [6].

Although ON is seen in any age group, most patients are young adults, with about 75% between 30 years of age and 60 years of age and with the average age in the late 30s. The overall male-to-female ratio is about 4:1, while the prevalence of this ratio is about 7:3 in SLE patients [3].

New cases of ON at the femoral head in the United States numbered between 10,000 and 20,000 each year and about 5—10% of them ended up with total hip joint replacement [1]. In China, there are a total of 4 million patients with femoral head ON and the number of total joint replacements in ON patients is increasing.

Histopathology

Histopathologically, the ON lesion is characterised by the diffuse presence of empty lacunae or pyknotic nuclei of osteocytes in bone trabeculae, accompanied by surrounding necrotic bone marrow [8—10]. Decreased trabecular width and increased number of apoptotic osteoblasts and osteocytes were also identified in patients with glucocorticoid administration. There were two repair types in ON lesions in SAON found by others and us. Appositional bone formation with osteoblast-like cells around the necrotic lesion is classified as reparative osteogenesis, whereas granulation tissue creep linked to necrotic bone resorption is classified as destructive repair [9,11,12]. In terms of destructive repair, the necrotic bone will subsequently induce an inadequate repair process, i.e., the predominant resorption of necrotic bone exceeds bone formation that frequently leads to subchondral collapse. In terms of reconstructive repair in the femoral head, reparative bone formation starts from subchondral fractures and/or the reactive interface, that finally reduces the size of the necrotic lesion.

Pathophysiology

The pathophysiology of SAON remains controversial and different mechanisms including the abnormality of the bone marrow stem cell (BMSC) pool, hyperlipidemia, fat emboli, hypercoagulable condition, vascular endothelial dysfunction, and apoptosis of bone tissues have been proposed extensively. SAON is not caused by only one factor but multiple factors and these influence each other during the process of SAON development (Fig. 2), which subsequently results in marrow ischaemia and eventually ON.

Abnormality of BMSCs pool

The recent advance in understanding the pathophysiological mechanism of the inadequate repair at the early stage of SAON is on a decreased activity of BMSCs pool [13—15].

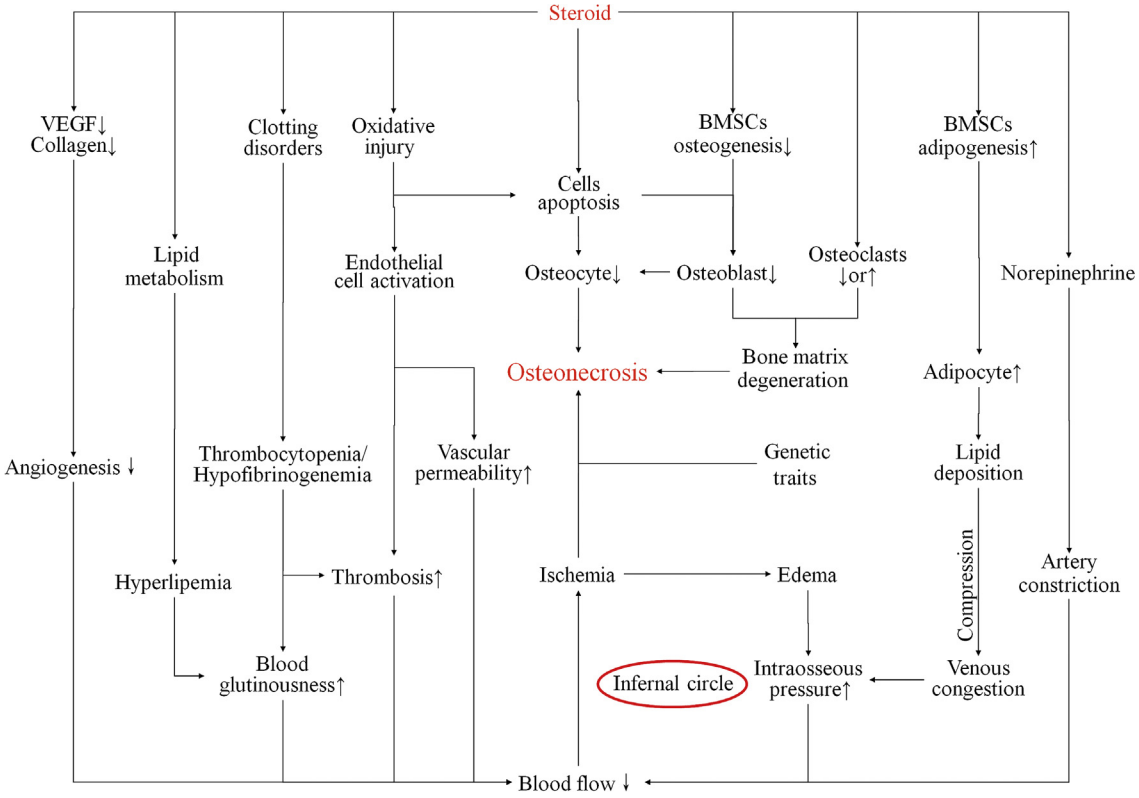

Figure 2 Pathophysiology of steroid-associated osteonecrosis (SAON) with different pathways (summarised from published work by authors of this review paper and others). BMSC = bone marrow stem cell; VEGF = vascular endothelial growth factor.

Steroids can induce differentiation of BMSCs into adipocytes linage by upregulating adipocytes transcription factor gene expression (Peroxisome proliferator-activated receptor gamma adipose-specific 422 and adipsin) and inhibit their osteogenic differentiation by downregulating osteoblast transcription factor gene expression [Cbfa1/Runx2, type I collagen, bone morphogenetic protein (BMP-2), osteocalcin, osteopontin, and osteonectin, and osteocalcin], thus resulting in increased lipid deposition including larger fat cells number and its area fraction. Clinically, Gangji and Hernigou reported decreased number and osteogenic differentiation ability of BMSCs pool in iliac or femora in patients with SAON, which resulted in the inadequate lesion repair at early stage of ON [13—15]. The authors' group also demonstrated significantly increased lipid deposition including larger fat cells number and fat deposition area at the early stage of SAON in an animal model [9,11,16,17].

Bone matrix degeneration

Decreased bone formation in SAON was regarded as the common cause of osteoporosis. Weinstein administered prednisolone to mice and observed a decrease in bone density, serum osteocalcin, and cancellous bone area along with trabecular narrowing, accompanied by diminished bone formation and turnover. Takano-Murakami et al [18] also recently found that methylprednisolone (MPSL) sodium succinate inhibited bone formation by suppressing osteoblasts proliferation and bone resorption by suppressing the recruitment of osteoclasts precursors [19]. Thus, the decreased function of osteoblasts and osteoclasts or impaired balance between the two cell types by corticosteroids would result in bone matrix degeneration, which explained the delayed bone fracture healing in animals or patients under steroid administration [12].

Articular cartilage degeneration

The cartilage layer could be found detached from the bone in severe ON under MRI or arthroscopic evaluation. Glucocorticoid receptors were present in cartilage and the steroid was revealed to cause cartilage disruption with acellular areas [20], which indicate that the steroid might result in the degeneration of articular cartilage. In most patients, subchondral collapse changes the articular surface, leading to abnormal mechanics (e.g., local stress concentration) that is believed to be associated with development of arthritis to the joint [21]. Our study revealed that the thickness of the subchondral plate of SAON emus decreased significantly and the articular cartilage showed pathological alteration with significantly decreased thickness as well as the decreased proteoglycans content located at the collapsed region [22]. Following subchondral collapse, the cartilage components might be exposed to the ongoing repair process. Cartilage constituents may thus promote the development and continuance of the disease process in ON and explain the localisation of ON to subchondral bone [23].

Cell apoptosis

As well as bone matrix degradation, SAON was also characterised by the diffuse presence of empty lacunae or pyknotic nuclei of osteocytes in bone trabeculae [12,24—26]. If the death of the bone subsequently induces an inadequate repair process, the bone underlying the joint surface will be weakened. The steroid not only decreases the function of osteoblasts and osteoclasts, but also induces the apoptosis of osteoblasts and osteocytes. The steroid-treated mice exhibited a threefold increase in osteoblast apoptosis in vertebrae and showed apoptosis in 28% of the osteocytes in metaphyseal cortical bone. Clinical research showed that nitric oxide metabolism was altered in bone cells during the development of ON at the femoral head and was accompanied by widespread apoptosis of osteoblasts and osteocytes, which suggested that the nitric oxide-mediated apoptosis was a potential mechanism for SAON [27].

A recent study demonstrated that corticosteroid treatment could induce the development of autophagy of osteocytes, suggesting a new mechanism for the effect of steroids on osteocytes and giving new insight into mechanisms responsible for bone loss in patients with SAON [26].

Abnormality of lipid metabolism and clotting disorders

Ischaemia is a very important pathomechanism of SAON (Fig. 2). Abnormality of lipid metabolism and clotting disorders of blood are the major events which lead to ischaemia. High-dose MPSL could induce multifocal ON in conjunction with thrombocytopenia, hypofibrinogenemia, and hyperlipaemia, which resulted in increased blood glutinousness followed by decreased blood flow. The altered fat metabolism could increase serum lipid levels (triglyceride and cholesterol) and a fat embolism resulted in vascular occlusion which accelerated the ischaemia. The intravascular thrombosis resulting from clotting disorders was also the key pathogenic pathway to cause ischaemia in SAON found by others and our own studies [7,10,17,28]. A study from our group showed that the inhibition of both thrombosis and lipid deposition could reduce the incidence of SAON [11,16], which proved that the abnormality of lipid metabolism and clotting disorders were indeed important mechanism for SAON. It was found that hypercoagulability of plasma happened after 24 hours of high-dose MPSL treatment, which might be the pathogenetic factors contributing to the early stage of SAON and therefore to be used as an index of early risk factor to predict later SAON occurrence.

Decreased angiogenesis and elevated vasoconstriction

During the development of SAON, the destructive repair at necrotic lesions was characterised by predominant resorption of necrotic bone exceeding bone formation accompanied by decreased angiogenesis. Vascular endothelial growth factor (VEGF) is an important factor to act on the endothelial cells and induce neovascularisation during bone healing. Decreased VEGF expression after treatment with dexamethasone was both dose- and time-dependent. Clinical and animal experiments showed decreased VEGF expression at the early stage of SAON and increased VEGF in the reactive nitric oxide lesion at the late stage [Association Research Circulation Osseus (ARCO): stage IV] of the femoral head [29]. As well as the decreased VEGF expression at the early stage of ON found by others, we also found increased VEGF expression 2 weeks after ON induction, which was maintained at Week 6 during the destructive repair in SAON rabbits, whereas VEGF expression in reconstructive repair decreased from Week 2 to Week 6 [9].

Corticosteroids could also inhibit the capillary growth via suppressing VEGF and collagen synthesis. It was reported that glucocorticoids could increase $Ca2^{+}$ uptake in vascular smooth muscle cells and downregulate the expression of endothelial nitric oxide synthase, which leads to strong vessel contraction.

Oxidation injury

Oxidation injury was another pathogenesis for SAON. It was reported that oxidative injury in BMSCs was demonstrated shortly (3 days) after the administration of MPSL in a rabbit model prior to ON development [30]. The significant increased rate of ON development by using antioxidant showed that the oxidation injury was not only involved in the early stage of SAON [27]. Tissue oxidation was proven to be able to induce cells apoptosis through oxygen free radicals. Oxidative stress inhibited osteogenic differentiation of primary rabbit BMSCs and calvarial osteoblasts by stimulating extracellular signal-regulated kinases (ERKs) and nuclear factor-κB [31], implying decreased bone formation in SAON.

Genetic traits

As stated before, not all patients who received steroid administration would develop SAON, because about 11.2—40% of patients with a history of high-dose corticosteroid develop ON [32—34]. This indicated the presence of individual differences in steroid sensitivity. So, genetic traits may play an important role on predisposing the development of SAON. It was reported that inherited thrombophilia and hypofibrinolysis (high factor VIII, factor V Leiden heterozygosity, and resistance to activated protein C) were risk factors for SAON. Genetic susceptibility factors in SAON include single nucleotide polymorphism (SNP) and gene mutation. Most SNP was related with the angiogenesis, drug delivery, steroid metabolism, lipid metabolism, and drug resistance, which may influence the development of SAON. It seems that the genetic analysis showed a potential relationship between SAON and SNP, which is suggested to be useful for predicting the development of ON and of benefit to the clinician in preventing ON as early as possible.

SAON animal models

To examine the mechanism or to identify the prevention and treatment efficiency of the biomaterial or pharmacological therapy strategies for SAON, different animal models (Table 1) were built up, including rabbit [8,10,11,35—37], rat [38], mouse [39,40], pig [41], bipedal chicken [42], and emu [22]. Among all experimental models, quadrupedal rabbits are commonly used for establishing SAON, which was proven to be a good animal model for developing high ON incidence in our recent work [7,10,11,43,44] and also by others [8,35,36], yet without being able to develop to joint collapse, mainly explained by lower mechanical loading onto the weight-bearing joints.

To induce SAON in rabbits, the administration of single MPSL [37] was proven to be an efficient method, but the incidence of ON was about 33.3—43%. Two injections of high-dose lipopolysaccharide (LPS) combined with a subsequent three injections of high-dose MPSL (H-LPS × 2 + H-MPSL × 3) could improve the incidence of ON but result in high mortality [37]. In our group, we have developed a protocol by modifying the reported protocol with a single low-dose LPS, followed by three times injections of MPS which had a higher incidence of ON of up to 93%, with lower or no mortality [7,10,11,43,44]. We also established a bipedal emu model for testing and supporting Chinese herbal *Epimedium* in prevention of SAON with functions in bone formation stimulation and antiadipogenesis etc. [22].

Although a high incidence of SAON was found in different animal models, we noted that the incidence rate in humans was not so high [45], which indicated a difference between animals and human. It is benefit for us to test the prevention and treatment efficacy in animals with a high incidence of SAON.

Table 1 Steroid-associated osteonecrosis and the incidence rate of osteonecrosis in animal models.

Animal model (Ref. no.)	Induced protocol				IR	Time	Location	Mortality
	Agent	Dosage	Usage	Duration				
Rabbit [8]	MPSL	20 mg/kg	i.m.	Once	43%	4 wk	Femora, humerus	—
Rabbit [36]	MPSL	20 mg/kg	i.m.	Once	72.7%	2 wk	Femora	—
Rabbit [37]	LPS	100 μg/kg/d	i.v.	2 d	85.7%	4 wk	Femora, humerus	20%
	MPSL	100 μg/kg/d	i.m.	3 d				
Rabbit [7,10,11]	LPS	10 μg/kg	i.v.	Once	92.86—93.75%	2—6 wk	Femora	0
	MPSL	20 mg/kg/d	i.m.	3 d				
Rat [38]	Human serum	10 mL/kg/wk	i.p.	2 wk	90%	2 wk	Femora	—
	MPSL	40 mg/kg/d	i.m.	3 d				
Mouse [39]	Dexamethasone	4 mg/L	Drinking water	12 wk	40—45%	12 wk	Distal femur	17%
Pig [41]	MPSL(two steps)	30 mg/kg 5.4 mg/kg/h	i.v.	Initial bolus 23 h	Reduced blood fiow	24 h	Femora	—
Chicken [42]	MPSL	3 mg/kg/wk	i.m.	12 wk	56%	6 wk	Femoral head	—

i.m. = intramuscular; i.p. = intraperitoneal; i.v. = intravenous; IR = incidence rate; LPS = lipopolysaccharide; MPSL = methylprednisolone acetate.

Prevention of SAON

It is very difficult to predict which patients receiving steroids will develop ON, although prevention of SAON is essential, e.g., already at the time of steroids treatment [23]. Controlled clinical trials are lacking to show the efficiency of prevention of SAON occurrence. Most experiments on prevention of SAON were performed using laboratory animals (Table 2). It was reported from others and our group that the Chinese herb such as an *Epimedium*-derived phytoestrogenic compound or its metabolic products and Liuwei Dihuang pills [11,16,24,25,46], genistein aglycone [47], a lipid-lowering agent (lovastatin or pitavastatin) and/or an anticoagulant (warfarin or enoxaparin) [48], antioxidative substances (vitamin E) [49], lipoic acid [50], nitrate patch [51], hepatic CYP3A inducer [52], electromagnetic fields [53], or bone marrow cells [36] were able to exert partly beneficial effects on prevention of SAON in different models attributed to their osteogenic and anti-adipogenic effects, as well as inhibition of both thrombosis and lipid deposition in a dose-dependent manner.

Chinese herbs

A lower incidence of SAON happened in Hong Kong and Guang Zhou in China during explosion of severe acute respiratory syndrome (SARS) but was higher in Beijing — partially explained by use of the antiinflammatory herbs, but no clinical trials were performed, so our group established both quadrupedal and bipedal models for testing and supporting herbal *Epimedium* in the prevention of SAON, with functions in bone formation stimulation, anti-adipogenesis etc. [7,10,11,16,24,25]. Liuwei Dihuang pills and Huogu I formula can prevent SAON by improving lipid metabolism, relieving bone lesions, and protecting against cell death [46,54]. Genistein aglycone, an isoflavone acting on *de novo* protein synthesis and on amplification of the interaction between the oestrogen receptor complex and nuclear DNA in osteoblasts, could prevent complications of ON with long-term glucocorticoid treatment due to its action on trabecular bone by a mechanism involving estrogen receptor (ER)-β upregulation during the bone mineralisation phase [47].

Lipid-lowering agent

It was reported that statins usage can partly lower the risk of ON. A retrospective report from Pritchett [59] showed that the ON incidence was only 1% in 284 patients who had received high-dose corticosteroids while under concurrent statins administration, which was significantly lower than the historical value of 3—20% without adjunct therapy [55]. Ajmal et al [45] analysed the renal transplant database to prove that statins usage could reduce the incidence of SAON and identified 2881 renal transplantation patients who met the entry criteria of ON. Among patients who used statins, 4.4% developed ON, versus 7% in patients who were not on statins. The prevention of SAON in animal experiments showed that statins could prevent SAON development due to their inhibition of adipogenesis and lipid deposition, reduction of blood viscosity by suppressing PPAR(sup) expression and activating the Wnt signalling pathway [42,56,57]. Enoxaparin, one of the anticoagulants,

Table 2 Incidence rate (IR) of steroid-associated osteonecrosis in different models with different preventive agents.

Preventive agents (Ref. no.)	Usage	Model	IR without prevention	IR with prevention
Flavonoids derived from *Epimedium* [16,24,25]	Low (10 mg/kg/d) Middle (20 mg/kg/d) High (40 mg/kg/d) per os, 2 wk	Rabbit	93%	Low (56%) Middle (13%) High (6%)
Icaritin [11]	Low (5 mg/kg/d) High (10 mg/kg/d) per os, 2 wk	Rabbit	93.75%	Low (56.25%) High (6.25%)
Genistein aglycone [47]	5 mg/kg/d, i.p., 60 days	Rat	Histological score: 3	Histological score: 0.5
Liuwei Dihuang pills [46]	2 g/kg/d, per os, 8 wk	Mice	Rate of empty lacunae: 28%	Rate of empty lacunae: 8%
Statins drugs [45]	At least 1 y duration	Human	7%	4.4%
Lovastatin [48]	5 mg/kg/d, per os, 14 wk	Rabbit	68%	35%
Enoxaparin + lovastatin [48]	Enoxaparin: 1 mg/kg/d, s.c., 4 wk; lovastatin: 5 mg/kg/d, per os, 14 wk	Rabbit	68%	15%
Vitamin E [49]	50 mg/kg/d, i.v., 2 wk	Rabbit	93%	0%
Lipoic acid [50]	36 mg/kg/d, i.p., 4 wk	Rabbit	73.1%	20.8%
Nitrate patch [51]	0.675 mg/d, per os, 4 wk	Rabbit	Empty lacunae: 4/μm^2	Empty lacunae: 1.5/μm^2
Hepatic CYP3A inducer [52]	25 mg/kg/d, i.m., 6 wk	Rabbit	83%	33%
Electromagnetic fields [53]	15 Hz, 10 h/d, 5 wk	Rabbit	65%	37.5%
Autologous bone marrow cells [36]	1×10^7cells, intra-bone marrow injection, once	Rabbit	72.7%	0%

i.m. = intramuscular; i.p. = intraperitoneal; i.v. = intravenous; s.c. = subcutaneous.

has less ability to inactivate thrombin and can reduce the incidence of steroid-induced ON in rabbits when combined with lovastatin [48].

Antioxidative substances

Increased oxidative stress is considered as a crucial cause of SAON. Antioxidative substances such as vitamin E, lipoic acid, and fullerol may alleviate oxidative injury following corticosteroid administration, and thus prevent ON. Vitamin E is a fat-soluble substance in the body that significantly inhibited steroid-induced oxidative stress [49]. Lipoic acid can prevent the development of SAON by inhibiting oxidative stress and amendment of vascular endothelial dysfunction [50]. Fullerol inhibits adipogenesis and simultaneously enhances osteogenesis by marrow mesenchymal stem cells, possibly through elimination of cellular reactive oxygen species [58].

Nitrate patch

A nitrate patch, an agent which generates vasodilation in bone, had preventive effects against SAON. The substance released from the nitrate patch might replace the deficient NO and counteract the vasoconstriction [51].

Hepatic cytochrome P4503A (CYP3A) inducer

Hepatic cytochrome P4503A (CYP3A) is a predominant enzyme responsible for metabolising corticosteroids, and hepatic CYP3A levels are significantly lower in patients with SAON. Hepatic CYP3A activity induced by phenobarbital was inversely correlated with the incidence of ON and extent of the necrotic area caused by the same dose of corticosteroids, suggesting prevention of SAON by reducing steroid dose in poor corticosteroid metabolisers [52].

Electromagnetic fields

Electromagnetic stimulation could prevent SAON in rabbit and rat models, and the underlying mechanisms involved decreased serum lipid levels and increased expression of transforming growth factor β1 (TGF-β1) [53]. The authors also found that this treatment can help prevent ON, but if it occurs, the treatment does not reduce its severity.

Injection of autologous bone marrow cells

Injection of autologous bone marrow cells directly into the femurs could prevent SAON in rabbits treated with high-dose steroid because of their regulation of fibrinogenolysis and coagulation in the peripheral blood [36].

Antiapoptotic agents

Prevention of glucocorticoid-induced apoptosis in osteocytes and osteoblasts might be a potential method to prevent the development of ON. Calcium-binding protein calbindin-D28k, and granulocyte colony stimulating factor and stem cell factor have been reported to prevent glucocorticoid-induced bone cell death through inhibition of glucocorticoid-induced caspase 3 activation, as well as ERKs activation [59,60]. Bisphosphonates and calcitonin promoted a rapid increase in the phosphorylated fraction of ERKs and inhibited the apoptosis of osteocytes and osteoblasts following prednisolone administration to mice. Recently, the skeletal muscle secreted factors were found to prevent glucocorticoid-induced osteocyte apoptosis through activation of β-catenin.

The prevention effects of SAON by different agents are summarised in Table 2. Each agent reported in the published studies showed positive effects on SAON prevention, but most of them were generated from animal experiments. Available human studies demonstrated that statins were able to decrease the incidence of SAON as well [45], but the prevention efficiency was lower than that generated from animal models. This might be explained by the fact that the SAON was caused not by a single but by multiple pathophysiologies (Fig. 2) and should be prevented by several measures combined together. More clinical tests and trials are desirable in future, especially with those agents with known mechanisms discussed above.

Treatment of SAON

Treatment of SAON is a big challenge, as sufficiently large prospective controlled studies are lacking. According to the different stages of ON (ARCO classification), there are several treatments available for SAON, including nonoperative and operative therapy (Table 3).

Nonoperative treatment

Nonsurgical treatments of ON are limited and are commonly indicated for patients at the early stage of ON with localised necrotic lesions. The objects of such intervention usually focus on pain relief, prevention of ON progression, and improvement of the joint function. According to the different pathophysiologies of SAON, numerous medical and biophysical treatments have been tested and have demonstrated treatment efficacy, especially for patients in early ON stages, including biological and pharmacological treatments such as statins [45,55], bisphosphonates [61], low molecular weight heparin [62], stanozolol, iloprost, hyperbaric oxygen, herbal (puerarin and *Epimedium*) [11,16] or nonpharmacological treatments including extracorporeal shock wave therapy [63,64] and electromagnetic therapy [65,66].

Operative treatment

Operative treatment alternatives for femoral head ON mainly include osteotomy, core decompression sequestrectomy, bone and/or cells grafting, and total hip arthroplasty. The age and general health of patients, ON stage particularly with collapse of the femoral head and/or acetabular involvement, and size and location of lesions are the common determining factors for considering operation options.

Table 3 Approaches for diagnosis and stage-based interventions according to Association Research Circulation Osseus (ARCO) staging system.

Stages	Diagnosis	Treatment
Stage 0	Bone biopsy, bone scan, MRI	Prevention: Pharmacotherapy Physical therapy
Stage I	Bone scan, MRI	1. Nonoperative treatment: Pharmacotherapy Physical therapy 2. Operative treatment: Core decompression Core decompression with bone grafting Core decompression with bone marrow Core decompression with cells Core decompression with growth factors Tissue-engineered approach
Stage II	Radiographs or CT, bone scan, MRI	Operative treatment: Core decompression Core decompression with bone grafting Core decompression with bone marrow Core decompression with cells Core decompression with growth factors Tissue-engineered approach Osteotomy
Stage III	Radiographs	Operative treatment: Core decompression with bone grafting Core decompression with bone marrow Core decompression with cells Core decompression with growth factors Tissue-engineered approach Osteotomy Arthrodesis Total hip arthroplasty
Stage IV	Radiographs	Operative treatment: Arthrodesis Total hip arthroplasty

CT = computed tomography; MRI = magnetic resonance imaging.

Osteotomy

Intertrochanteric rotational osteotomy or flexion osteotomy is joint-preserving therapy for SAON by which the osteonecrotic lesion is rotated away from the weight-bearing area. However, the specific complications are bony nonunion and pseudarthrosis.

Core decompression

Core decompression through creation of a surgical drilled bone tunnel is one of the least invasive surgical procedures in early ON stages when the ON lesion is still small (Fig. 3). Core decompression is usually performed in Ficat Stage II or earlier stages to prevent the subchondral collapse in SAON. Biologically, it helps to reduce intraosseous pressure and provide a conduit for angiogenesis to revascularise subchondral bone. However, concerns still do exist, including those related to incomplete reconstructive repair and its potential to weaken the trabecular bone within and next to the necrotic region when the necrotic lesion was relatively large [1,67,68]. However, once the femoral head develops a subchondral fracture, the efficacy of the core decompression will drop significantly.

Bone grafting

In order to reinforce the surgically-induced bone defect resulting from core decompression sequestrectomy and delay the need for arthroplasty, several types of bone grafts have been used to provide mechanical support for the affected joint in Ficat Stage III or in even earlier stages (Fig. 4). These include autogenous or allogenous cortical bone grafts of ilium, fibula, or tibia alone, or alternatively, these procedures may be combined with core decompression [1,3]. A vascularised bone graft was used that showed some superior results as compared with most other procedures designed to preserve the femoral head collapse [69,70]. The autologous bone graft is used as a good substitute of the necrotic site, but limitations include insufficient supply and variation in the osteogenic potential of the graft material. The harvesting of host bone often results in donor site morbidity. Fresh osteochondral allografting is a reasonable salvage option for ON of the femoral condyles, thus total knee arthroplasty (TKA) was avoided in 27 of the 28 of knees at last follow-up [71]. Tantalum rod implantation is a safe "buy-time" technique, especially when other joint salvage procedures are not an option [72]. Tantalum

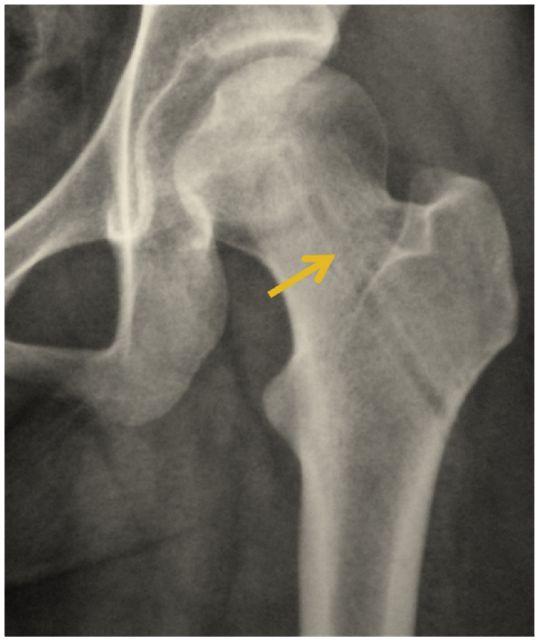

Figure 3 Core decompression procedure (arrow) of femoral head with steroid-associated osteonecrosis (SAON).

rod implantation (Fig. 5) combined with bone grafting is an effective joint-preserving method for the treatment of intermediate-stage ON of the femoral head [73,74]. Recently, the PLGA/tricalcium phosphate (TCP)/icaritin scaffold could be used to treat bone defects in rabbits with SAON [75].

Bone marrow or BMSCs transplantation

Due to the decreased activity of MSCs pool within or around ON lesions in SAON, additional MSCs or bone marrow

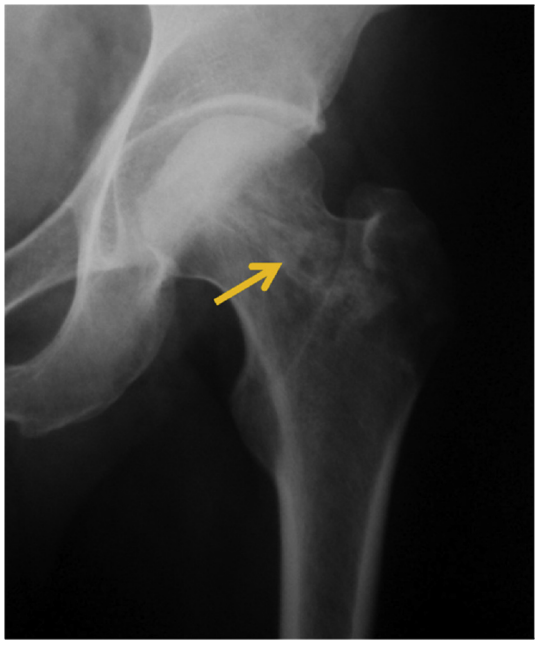

Figure 4 Femoral head with osteonecrosis (ON) treated with allogenous bone graft (arrow).

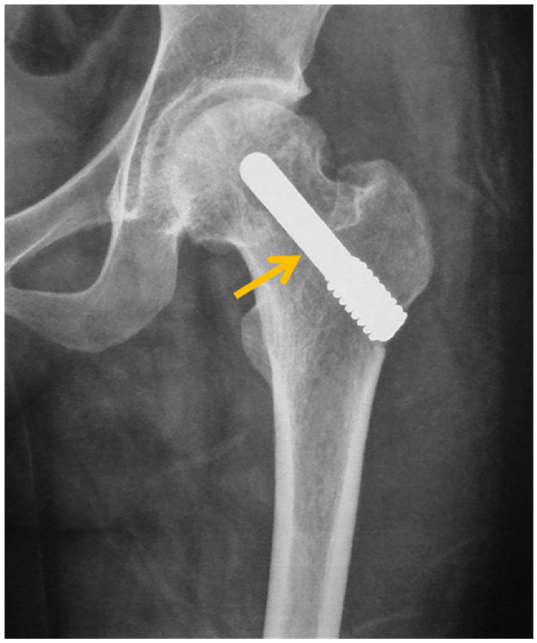

Figure 5 Femoral head with osteonecrosis (ON) treated with tantalum rod implantation (arrow).

grafting are promising treatment options for SAON attributed to their differentiation properties, easy accessibility, and proliferative capacity [76—78]. Clinically, good results were generously reported from treatment of SAON using autologous bone marrow grafts combined with core decompression and those who had the greater number of progenitor cells transplanted in ON lesions had better outcomes [13,14,79]. Most of the bone marrow was collected from the iliac crest, but it was reported that the bone marrow near the site of ON could also be used [80]. Their surgical procedure was less invasive, but it might not be effective for large ON lesions because of larger marrow spacing created after core decompression sequestrectomy. In addition, the animal studies also showed that bone marrow mononuclear cells were beneficial for vascularisation and bone regeneration in SAON [35,78]. Transplantation of autologous endothelial progenitor cells was also beneficial for SAON at the femoral head in rabbits [81]. Although the supplement of enough cells could enhance the repair of ON lesions, we also realise that the implanted cells might lose their ability to repair, as it was found that the pluripotential marrow cells produced adipocytes when they were transplanted into steroid-treated mice [82].

Growth factors

Growth factors such as BMPs and VEGF are osteogenic or angiogenic to promote the bone or vessel formation. BMPs are osteoinductive growth factors that induce osteogenic cell differentiation *in vitro* and osteogenesis in bone healing *in vivo*. The growth factors can be used for ON treatment directly or combined with bone graft after core decompression [83]. The growth factor genes could also be transfected into BMSCs and were capable of treating the ON in its early stage(s), e.g., demonstrated at the femoral head in rabbit or goat models and the promoted osteogenesis and angiogenesis were observed [84].

Tissue-engineered approach

Although core decompression with supplementation of BMSCs could be used to treat ON, it might not be effective for large ON lesions or post collapse cases. Due to the apoptosis of osteocytes and degeneration of trabecular bone matrix in the femora, the bone harvested showed decreased viability in SAON. Kawate used tissue-engineered approach for treating SAON in the femoral head, where cultured BMSCs/b-TCP composite granules were implanted into the cavity that remained after curettage of necrotic bone, together with a subsequent free vascularised tibia grafting [85]. BMSCs/biphasic calcium phosphate ceramic scaffolds and porous β-TCP loaded with BMP-2-gene-transduced BMSCs performed in animal models also provided effective treatment results [84,86].

Total hip arthroplasty

Total hip arthroplasty (Fig. 6) is usually used primarily for ON Stages III and IV, but occasionally also used for Stages I and II. The prognosis of joint replacement is poor in patients with idiopathic or traumatic ON, but still better as compared with SAON patients. The aetiology of ON did not normally affect the final outcome, but less favourable long-term results were found in SAON patients. Patients with femoral head ON are generally young and total hip arthroplasty is often an unfavourable choice. Joint preservation is too difficult due to large pre collapse lesions and post collapse disease, so surgical alternatives for these patients may also include limited femoral resurfacing and bipolar hemiarthroplasty. However, the most useful and common treatment is total hip arthroplasty.

Summary and further translational research

In conclusion, ON is a challenging orthopaedic condition which resulted from unwanted adverse effects of corticosteroids administration. Normally, no single factor is involved in the pathophysiology of SAON initiation and its development. Evaluation of approaches developed for prevention and treatment of ON in both animal and human studies indicated that a certain pathophysiology resulted in ON, including abnormality of BMSCs pool, bone matrix and cartilage degeneration cell apoptosis, abnormality of lipid metabolism and clotting disorders, decreased angiogenesis and elevated vasoconstriction, and oxidation injury. Orthopaedic surgery is important for the treatment of SAON, yet its prognosis is rather poor. This implies the importance of prevention in two aspects, including: (1) to prevent the happening of SAON after the administration of steroid; and (2) to prevent the development of SAON from early stage to late stage.

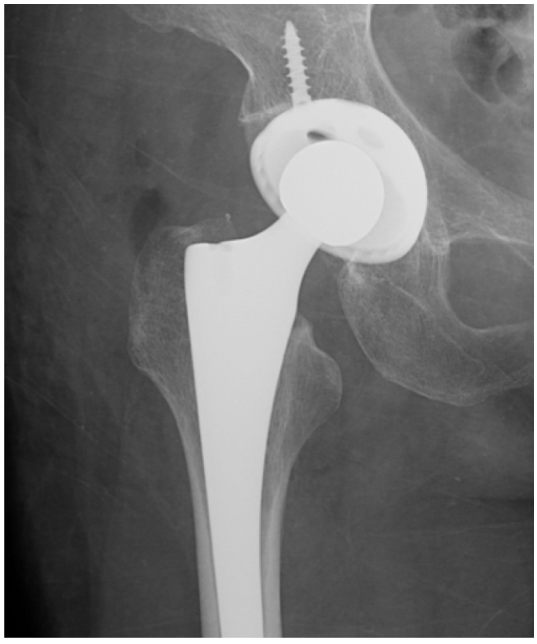

Figure 6 Femoral head with osteonecrosis (ON) treated with total hip arthroplasty.

Based on the possible pathophysiology and the effective prevention or therapy performed in animals and humans with steroid administration, much more bench to clinic translational work shall be done in the future to promote efficiency of early prevention, diagnosis, and treatment of SAON. Good prevention requires good predictive diagnosis. Gene analysis or hypercoagulability of plasma might be earlier methods to predict ON development, although dynamic perfusion MRI is another advanced and promising bioimaging approach. Chinese herb and statins treatment produced significantly lower incidences of SAON both in humans and animal models, implying that they might be promising alternatives for prevention and eventually treatment of SAON. The apoptosis of osteoblasts, osteoclasts, and osteocytes suggested a new mechanism for the effect of steroids, giving a new insight into mechanisms responsible for bone loss in patients with SAON. Core decompression combined with bone grafting, bone marrow or cells transplantation, and growth factors is the best documented therapy for early SAON stage(s). The newly tested osteopromotive porous composite scaffolds were also promising for enhancing repair of ON lesions after core decompression [87,88]. Gene therapy was proven to be effective in animal models, but its safety and ethical issues are still controversial. Despite all treatments, joint replacement is still advocated for patients at the late ON stage(s). How to improve postoperative prognosis after joint replacement therapy remains a challenging topic for not only orthopaedic surgeons, but also biomedical engineers and biomaterial scientists. We hope that with all collective efforts, more scientifically confirmed approaches will be available for clinical applications and will benefit patients suffering from ON, especially SAON.

Conflicts of interest

All contributing authors declare no conflicts of interest.

Acknowledgements

The work was supported by Hong Kong General Research Fund (GRF CUHK-473710/473011) and a general financial grant from the China Postdoctoral Science Foundation (2012M511800).

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