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HMGB proteins and arthritis

Noboru Taniguchi,

Department of Orthopaedic Surgery, University of Miyazaki, Miyazaki, Japan, Department of Medical Science, Tokyo Medical University, Tokyo, Japan, 5200 Kihara, Kiyotake, Miyazaki 889-1692, Japan

Yasuhiko Kawakami,

Department of Genetics, Cell Biology and Development, and Stem Cell Institute, University of Minnesota, Minneapolis, USA, 321 Church St. SE, 6-160 Jackson Hall, Minneapolis, MN 55455

Ikuro Maruyama, and

Department of Systems Biology in Thromboregulation, Kagoshima University Graduate School of Medical and Dental Science, Kagoshima 8908544, Japan

Martin Lotz

Department of Molecular and Experimental Medicine, The Scripps Research Institute, La Jolla, CA USA, 10550 N. Torrey Pines Road, MEM 161, La Jolla, CA 92037

Abstract

The high-mobility group box (HMGB) family includes four members: HMGB1, 2, 3 and 4. HMGB proteins have two functions. In the nucleus, HMGB proteins bind to DNA in a DNA structure-dependent but nucleotide sequence-independent manner to function in chromatin remodeling. Extracellularly, HMGB proteins function as alarmins, which are endogenous molecules released upon tissue damage to activate the immune system. HMGB1 acts as a late mediator of inflammation and contributes to prolonged and sustained systemic inflammation in subjects with rheumatoid arthritis. By contrast, *Hmgb2*$^{-/-}$ mice represent a relevant model of aging-related osteoarthritis (OA), which is associated with the suppression of HMGB2 expression in cartilage. *Hmgb2* mutant mice not only develop early-onset OA but also exhibit a specific phenotype in the superficial zone (SZ) of articular cartilage. Given the similar expression and activation patterns of HMGB2 and β-catenin in articular cartilage, the loss of these pathways in the SZ of articular cartilage may lead to altered gene expression, cell death and OA-like pathogenesis. Moreover, HMGB2 regulates chondrocyte hypertrophy by mediating Runt-related transcription factor 2 expression and Wnt signaling. Therefore, one possible mechanism explaining the modulation of lymphoid enhancer binding factor 1 (LEF1)-dependent transactivation by HMGB2 is that a differential interaction between HMGB2 and nuclear factors affects the transcription of genes containing LEF1-responsive elements. The multiple functions of HMGB proteins reveal the complex roles of these proteins as innate and endogenous regulators of

Corresponding author: Noboru Taniguchi, MD, PhD, Department of Orthopaedic Surgery, University of Miyazaki, 5200 Kihara, Kiyotake, Miyazaki 889-1692, Japan, Tel: +81-985-85-0986, Fax: +81-985-84-2931, nobutanigu@gmail.com.

Conflicts of interest

The authors have no conflicts of interest to declare.

inflammation in joints and their cooperative roles in cartilage hypertrophy as well as in the maintenance of joint tissue homeostasis.

Keywords

HMGB protein; rheumatoid arthritis; osteoarthritis; inflammatory mediator; Wnt signaling

HMGB protein family

The high-mobility group box (HMGB) proteins are smaller than 30 kDa in size and bind to DNA in a DNA structure-dependent but nucleotide sequence-independent manner[1]. The HMGB family includes four members: HMGB1, 2, 3 and 4. HMGB1, 2, and 3 are more than 80% identical at the amino acid sequence level and have similar biochemical properties. These proteins comprise two DNA-binding HMG domains and an acidic tail[2]. HMGB4 is a mammal-specific protein that contains two HMGB-boxes but lacks the acidic tail, and its nucleotide sequence is less conserved than the sequences of the other *Hmgb* genes[3].

Although HMGB1, 2 and 3 share high levels of amino acid sequence similarity, their expression patterns are diverse. For instance, during mouse embryonic development, both *Hmgb1* and *Hmgb2* are ubiquitously and expressed at high levels[4, 5]. By contrast, *Hmgb3* expression is more localized during embryogenesis[6], and *Hmgb4* is detected in the developing brain and pancreas at E12.5 and E14, respectively[7, 8]. In adult tissues, *Hmgb1* has been shown to exhibit ubiquitous and high levels of expression in whole tissue extract assays[9, 10]; however, other *Hmgb* genes show restricted expression. *Hmgb2* is highly expressed in the thymus and testis[5]. *Hmgb3* is highly expressed in hematopoietic stem cells in the bone marrow[11], and *Hmgb4* expression is limited to testis and neuronal cells[3, 12]. The results of these expression analyses suggest that *Hmgb1* may have broad roles in embryonic development and in adult tissues, and other *Hmgb* genes may have redundant functions depending on the developmental stage and tissue in embryos and adults.

Biochemical studies have primarily been performed with HMGB1 and HMGB2, and have shown that HMGB1 and HMGB2 bind to DNA without sequence specificity[13, 14]. HMGB1 and HMGB2 play a role in the formation of nucleoprotein complexes by altering chromatin structures, which promotes the binding of other factors[15, 16] and facilitates diverse DNA modifications[1, 17]. These proteins are also known to regulate various activities, such as transcription, replication, and DNA repair[13]. HMGB1 and HMBG2 both bind to HOX proteins, steroid hormone receptors[18], and Rag1 recombinase[19] and enhance the transcription and recombination activities of their partner proteins when transiently transfected in mammalian cells.

Despite their similarities in amino acid sequence, structure and biochemical characteristics, the functions of HMGB1 and HMGB2 are not completely identical[5, 20]. Functional studies of *Hmgb* genes have been performed by generating targeted mutations in mice. *Hmgb1*$^{-/-}$ mice are born without significant morphological defects but die within a day due to hypoglycemia[20], which is caused by insufficient activation of a glucocorticoid receptor. Male *Hmgb2*$^{-/-}$ mice exhibit reduced fertility, which is caused by the degeneration of Sertoli

cells and germ cells in seminiferous tubules and by immotile spermatozoa[5]. *Hmgb3*$^{-/-}$ mice exhibit erythrocythemia[21]. These studies show that *Hmgb1, 2,* and *3* are not required for embryonic development individually, with the exception that *Hmgb1*$^{-/-}$ limb long bones show delays in endochondral ossification[22]. The normal embryonic development of mutant mice suggests the functional redundancy of these proteins, particularly for *Hmgb1* and *Hmgb2*, due to their high expression levels in embryos and shared biochemical characteristics.

The role of extracellular HMGB1

Regarding an extracellular role[23], HMGB1 released by damaged cells acts as a chemoattractant and a proinflammatory cytokine[23]. The translocation of HMGB1 occurs in response to immune cell activation or cell death, and the redox state of cysteines has recently been reported to modulate the binding of HMGB1 to its receptors and its subsequent functions[24]. HMGB1 contains three cysteines: C23 and C45 form a disulfide bond, and C106 is unpaired. These cysteines are modified by redox reactions and give rise to three isoforms known as fully reduced HMGB1 for the all-thiol form, disulfide HMGB1 for the partially oxidized form, and sulfonyl HMGB1 for the terminally oxidized form[24]. Depending on the redox states of these amino acid residues, HMGB1 induces cytokine production via Toll-like receptor 4 (TLR4) or promotes chemotaxis by binding the chemokine C-X-C motif chemokine ligand 12 (CXCL12) to form a heterocomplex, which in turn binds to C-X-C chemokine receptor type 4 (CXCR4) and induces cell migration. The cysteines in HMGB1 are terminally oxidized to sulfonates; sulfonyl-HMGB1 neither is a chemoattractant nor displays cytokine-inducing activity[25]. HMGB1 also interacts with the receptor for advanced glycation end products (RAGE), a multifunctional transmembrane protein of the immunoglobulin superfamily[26], to induce the secretion of inflammatory mediators.

During the course of inflammatory disease, HMGB1 plays a dynamic role, depending on its redox state, and functions as an alarmin, which is an endogenous molecule that is released upon tissue damage to activate the immune system[27]. Cell death is an important mechanism that generates alarmins, and each major type of death (necrosis, apoptosis, pyroptosis and NETosis) leads to the release of different HMGB1 isoforms[28]. Therefore, posttranslational modifications of HMGB1 are important for determining its role during the response to tissue injury and the course of inflammatory diseases.

HMGB1 has been reported to play a role in the pathogenesis of autoimmune and inflammatory arthritis disorders, such as rheumatoid arthritis (RA), juvenile idiopathic arthritis, osteoarthritis (OA), crystal-induced arthritis, psoriatic arthritis, and spondyloarthritis[29–36] (Table 1). HMGB2 is also secreted by human THP-1 monocytic leukemia cells; extracellular HMGB2 has been detected in the blood and in other biological fluids, and it promotes the proliferation and migration of endothelial cells[37]. HMGB2 exerts these functions by engaging RAGE, as its blockade completely abrogates the cell responses described above.

HMGB1 and RA

In patients with RA, tumor necrosis factor-alpha (TNF-α) plays a clinical role in a cytokine cascade that results in joint inflammation and destruction. TNF-α acts on macrophages to enhance phagocytosis and the production of other proinflammatory cytokines and prostaglandin E2[38]. It also serves as a chemoattractant for neutrophils and induces chemokine expression on the endothelial cell lining to facilitate the transendothelial migration of neutrophils. TNF-α acts on fibroblast-like synoviocytes to induce their proliferation and pannus formation and upregulates matrix metalloproteinases (MMPs) such as MMP1 and MMP13, which participate in cartilage damage. In addition, TNF-α activates osteoclasts, which promote bone demineralization[38]. The use of anti-TNF-α therapy indicates its importance in this disease. However, many patients with RA fail to respond to this therapy or other biologics, and some patients may suffer from unexpected aggravation of joint inflammation or other autoimmune manifestations[39]. Collectively, these reports suggest that more complex interactions occur among TNF, interleukin-1 (IL-1), and other inflammatory mediators.

According to an *in vivo* analysis, HMGB1 concentrations were significantly higher in the synovial fluid (SF) from patients with RA than in patients with OA. Similarly, TNF-α levels in the SF were also significantly higher in the same patients with RA than in the same patients with OA, although HMGB1 concentrations in serum were very low, with no significant difference between patients with RA and OA[40]. In RA synovial tissue, HMGB1 was detected in the cytoplasm of macrophages infiltrating the sublining layer[41]. *In vitro* studies have shown that HMGB1 stimulation of synovial fluid macrophages (SFMs) obtained from patients with RA causes the release of proinflammatory cytokines, such as TNF-α, interleukin-1β (IL-1β), and IL-6. Upon stimulation with HMGB1, the levels of TNF-α released by SFMs into the supernatant peak prior to the levels of other cytokines (IL-1β, IL-6). Since TNF-α is an upstream cytokine, it might also stimulate IL-1β and IL-6 secretion[42, 43]. Thus, HMGB1 may induce SFMs to release high levels of not only TNF-α but also IL-1 and IL-6. By contrast, TNF-α also induces HMGB1 release from SFMs, and the HMGB1-induced release of IL-1 and IL-6 from SFMs occurred much later than TNF-α release.

Based on these results, HMGB1 acts as a late mediator of inflammation and contributes to prolonged and sustained systemic inflammation[23, 44–46]; in addition, a proinflammatory loop may exist between HMGB1 and TNF-α via SFMs in patients with RA.

HMGB2 and OA

Osteoarthritis (OA) is the most prevalent joint disease with a non-immune mediated pathogenesis and primarily involves cells of mesenchymal lineage, such as chondrocytes and osteoblasts. Aging is the major risk factor for this form of arthritis, which begins with disruption of the superficial zone (SZ) of cartilage, leading to progressive cartilage erosion and bone remodeling, causing disability and reduced quality of life. A link between aging and HMGB proteins has been reported in earlier studies. Phosphorylation and ADP-ribosylation of HMGB2 in the livers of old rats decreased drastically compared with young

rats after spermine or sodium butyrate treatment[47, 48], and dexamethasone stimulated HMGB2 methylation 12-fold in the livers of young rats, whereas this change was not observed in old rats[49]. Regarding HMGB2 expression during aging, Ly et al. measured mRNA levels in actively dividing dermal fibroblasts isolated from young, middle-aged, and elderly humans as well as humans with progeria[50] and found that *Hmgb2* was one of only 9 genes that were down-regulated in both the old age and progeria groups among the 6,000 genes examined. Recently, Aird et al. sought to investigate factors regulating chromatin reorganization during senescence and identified a decrease in HMGB2 expression as one of the most significant changes associated with senescence[51].

The SZ of articular cartilage is important in many respects because it forms a fluid-tissue interface of articular cartilage in the synovial cavity. The SZ possesses 3.5-fold more cells than the radial zone[52], and a concomitant decrease in cell number along with surface fibrillation is the earliest indicator of OA development[53]. The SZ is unique because it produces lubricin, also known as proteoglycan-4/superficial zone protein (PRG4/SZP), an important joint lubricant[54–56]. Several recent studies have suggested that adult articular cartilage contains cells with functional properties and phenotypic markers of mesenchymal stem cells (MSCs), and these cells appear to be primarily located in the SZ[57–59]. HMGB2 expression is uniquely restricted to cells in the SZ in normal mature human articular cartilage[60] (Figure 1), and importantly, joint aging in humans and mice leads to the loss of HMGB2 expression. Based on results from gene expression arrays, *Hmgb1* expression increased, and *Hmgb2* expression decreased in human OA-affected cartilage compared with normal cartilage[61]. PRG4/SZP has a distribution similar to that of HMGB2 and is involved in cartilage and joint homeostasis[62, 63]. Young *Hmgb2*$^{-/-}$ mice did not exhibit an apparent reduction in PRG4/SZP expression. With advancing age, however, *Hmgb2*$^{-/-}$ mice exhibited a substantial loss, and in some cartilage regions a complete loss, of PRG4/SZP expression in the nonmeniscus-covered weight-bearing regions of the knee joint. Joint aging in humans and mice is associated with a reduction in HMGB2 expression, and this correlates with a reduction in expression of PRG4/SZP. Mice lacking *Prg4* appear normal at birth but subsequently display synovial hyperplasia, subintimal fibrosis, and an abnormal articular cartilage surface[63]. The correlation between the reduction of HMGB2 and loss of PRG4/SZP expression in the SZ of articular cartilage but not in the synovium suggests that HMGB2 plays a unique role in cartilage SZ cells[60].

The aging-related loss of HMGB2 expression in the cartilage surface is correlated with a reduction in cellularity, and the aging-related reduction in cellularity was more remarkable in the knee joints of *Hmgb2*$^{-/-}$ mice than in wild-type mice (Figure 2). These changes were linked to increased apoptosis in *Hmgb2*$^{-/-}$ mice, and chondrocytes from *Hmgb2*$^{-/-}$ mice studied *in vitro* exhibited increased susceptibility to anti-Fas receptor/CD95 antibody-induced apoptosis. These findings suggest that a major function of chromatin-associated factor HMGB2 is to promote SZ chondrocyte survival. A role for HMGB proteins in protecting against cell death has been suggested in neuronal cells in subjects with polyglutamine diseases[64], normal Sertoli cells[5], and certain cancer cells[65].

Based on the results from these studies, *Hmgb2*$^{-/-}$ mice represent a relevant model of aging-related OA. Not only do *Hmgb2* mutant mice develop early-onset OA, similar to many of the

previously reported mutant strains of mice[66], but also, more importantly, they feature a specific phenotype in the SZ of articular cartilage. This phenotype closely resembles human OA, which first manifests in the SZ[67] (Figure 3).

HMGB2 and β-catenin

Wnt proteins are secreted factors that regulate cell proliferation and differentiation during early stages of chondrogenesis[68, 69]. Wnt binds to Frizzled/lipoprotein receptor-related protein (LRP) receptor complexes, and signal transduction leads to the inhibition of constitutive cytoplasmic β-catenin degradation in the canonical pathway. The accumulated β-catenin translocates into the nucleus, where it acts as a co-activator for various transcription factors, such as lymphoid enhancer binding factor 1 (LEF1)[70]. Wnt/β-catenin signaling plays diverse and important roles in embryonic skeletal development[71, 72]. Recent studies have shown that increased Wnt signaling due to loss of secreted Frizzled-related protein (sFRP) function leads to the development of OA[73]. Similarly, overexpression of β-catenin in chondrocytes in postnatal mice stimulates the expression of matrix-degrading enzymes[74]. By contrast, Wnt signaling also contributes to the maintenance of normal articular cartilage function. Inhibition of β-catenin signaling by transgenic overexpression of its intracellular antagonist ICAT in articular chondrocytes causes increased cell apoptosis and articular cartilage destruction[75], suggesting that the precise temporal and spatial activation of Wnt signaling in articular cartilage determines its homeostatic versus pathogenic effects.

In the embryonic stage, Wnt/β-catenin signaling is active during joint formation[72]. Postnatally, remarkable similarities between the localization of HMGB2 and β-catenin have been observed. HMGB2 expression and β-catenin activation have been found in all zones of articular cartilage in newborn mice; however, both become more restricted to the SZ with joint maturation, and both show aging-related loss in the SZ[76]. Although HMGB2 eventually becomes completely absent in the SZ of aged mouse joints, β-catenin signaling is activated in the middle and deep zones. HMGB2 enhances the expression of Wnt/β-catenin target genes *in vitro*[60, 76].

Although transfection of HMGB2 alone did not activate LEF1/T-cell factor (TCF)/β-catenin-responsive promoters, cotransfection of HMGB2 and β-catenin resulted in synergistic activation of a reporter, and this synergy was seen in chondrogenic cell types[76]. HMGB2 does not directly bind to regulatory DNA elements, but it augments the DNA binding of LEF1, a transcription factor that associates with β-catenin and regulates Wnt/β-catenin downstream genes, and the HMG domain of HMGB2 is responsible for interaction with LEF1[76]. HMGB2 and β-catenin do not interact directly. HMGB2 does not alter the electrophoretic mobility of LEF1 that has been complexed with oligonucleotides *in vitro*, suggesting that HMGB2 dissociates from the complex after performing its architectural function[17]. The complex including HMGB2 β-catenin, LEF1 and probably other components leads to enhanced expression of genes containing LEF1 binding sites[76].

Cell survival and the expression of cyclin D1 (*Ccnd1*), GLI family zinc finger 3 (*Gli3*) and Frizzled class receptor 2 (*Fzd2*), which are representative LEF1 target genes in

cartilage[74, 77, 78], were analyzed to determine the functional consequences of the HMGB2/LEF1 interaction in a cellular context, and the expression levels of these genes were reduced by siRNA-mediated HMGB2 knockdown in chondrocytes. Moreover, the conditional deletion of β-catenin by infecting β-catenin floxed chondrocytes with Cre-expressing adenovirus increased basal and induced apoptosis *in vitro*. Given the similar expression and activation patterns of HMGB2 and β-catenin in articular cartilage, the loss of these pathways in the SZ of articular cartilage may lead to altered gene expression, cell death and OA-like pathogenesis (Figure 4).

HMGB2 and Runt-related transcription factor 2 (RUNX2)

Recent studies show that endochondral ossification is an essential process not only for physiological skeletal growth but also for the development of osteoarthritis[79]. OA articular chondrocytes express markers of hypertrophic growth plate chondrocytes such as collagen type X alpha 1 chain (COL10A1), MMP13, and RUNX2, and RUNX2 contributes to the pathogenesis of OA through chondrocyte hypertrophy and matrix breakdown after the induction of joint instability in an experimental mouse model[80]. In *Runx2*$^{-/-}$ mice, chondrocyte maturation is disturbed[81, 82], and the overexpression of a dominant-negative form of RUNX2 in chondrocytes severely delays endochondral ossification and suppresses chondrocyte maturation[83, 84]. During mouse limb development, *Hmgb2* is expressed in proliferating and prehypertrophic zones but not in hypertrophic cartilage, where *Col10a1* is expressed robustly[85]. According to the results of an *in vitro* study, the levels of *Runx2* transcripts increase in *Hmgb2*$^{-/-}$ MSCs compared with wild-type MSCs throughout chondrogenesis. Moreover, ectopic HMGB2 expression in MSCs suppresses the expression of the chondrogenesis markers *Col2a1* and mitochondrial aspartate-glutamate carrier 1 (*Agc1)* as well as *Runx2*[85].

It has been reported that chondrocyte maturation is induced by β-catenin-mediated Wnt signaling[86–88]. The TCF/LEF consensus sequence is located downstream of an A/G-rich region of the *Runx2* promoter in a highly conserved area that contains binding sequences for NK homeobox 2 (NKX3–2), hypoxia-inducible factor 2 (HIF-2), and vitamin D-responsive elements in addition to RUNX2[89–93]. *Runx2* is autoregulated in part by negative feedback on its own promoter; overexpression of RUNX2 protein abrogates *Runx2* promoter activity, and a single RUNX2 site is sufficient for transcriptional autosuppression[89]. Based on the results from an *in vitro* physical interaction assay, HMGB2 binds to RUNX2 and LEF1[76], and RUNX2 also interacts with LEF1[94]. Moreover, HMGB2 repressed the augmentation of *Runx2* promoter activity mediated by co-transfection of β-catenin and LEF1 in chicken upper sternal chondrocytes, which differentiate into hypertrophic chondrocytes[85]. These findings support the hypothesis that the loss of HMGB2 may contribute to the pathogenesis of OA by stimulating chondrocyte hypertrophy through *Runx2* transactivation. As Wnt signaling mediates chondrocyte hypertrophy through activation of *Runx2*[86], one possible mechanism to explain the observed modulation of LEF1-dependent transactivation by HMGB2 *in vivo* is that the differential interactions between HMGB2 and nuclear factors affect the transcription of genes containing LEF1-responsive elements (Figure 4).

EFEMP1/fibulin 3 as a target for HMGB2

According to an analysis of gene expression patterns in MSCs from wild-type and *Hmgb2*$^{-/-}$ mice, EFEMP1 (EGF-containing fibulin-like extracellular matrix protein 1)/fibulin 3 levels were significantly reduced in *Hmgb2*-deficient mice[95]. The fibulin family includes 6 ECM proteins that are localized in basal membranes, stroma, and ECM fibers, mediating cell-to-cell and cell-to-matrix communication[96]. Fibulins are thought to organize and stabilize ECM structures during organogenesis and vasculogenesis[96]. *Efemp1*$^{-/-}$ mice display early aging phenotypes[97]. EFEMP1 is expressed in cartilage and bone structures in the mouse embryo[98], and EFEMP1 peptides are potential diagnostic biomarkers for OA[99].

EFEMP1 expression is uniquely restricted to cells in the SZ in normal mature human and mouse articular cartilage. The process of joint aging in humans and mice is associated with reduced EFEMP1 expression in the articular cartilage, where it is normally expressed only in the SZ[95]. However, in OA-affected cartilage, cells in the chondrocyte clusters are strongly positive for EFEMP1. PRG4/SZP displays a distribution similar to EFEMP1 in the articular cartilage and is involved in cartilage and joint homeostasis[62, 63]. Interestingly, the number of PRG4/SZP-positive cells in *Efemp1*$^{-/-}$ mice was significantly lower than that in wild-type mice. Moreover, a correlation was observed between the reduction and loss of EFEMP1 and reduction and loss of PRG4/SZP expression in the SZ of articular cartilage. The aging-related loss of EFEMP1 expression in the cartilage surface is associated with a reduction in SZ cellularity and increased apoptosis in *Efemp1*$^{-/-}$ mice. The results of *in vitro* studies showed that *Efemp1* knockdown in chondrocytes resulted in reduced cell viability[95]. *In vitro* studies of chondrogenesis revealed that levels of the EFEMP1 protein decreased as MSCs differentiated. The siRNA-mediated suppression of EFEMP1 expression in articular chondrocytes enhanced the expression of chondrogenic markers such as COL2A1, aggrecan (ACAN), and SRY-box 9 (SOX9), whereas SOX9 expression was decreased following EFEMP1 overexpression, suggesting that EFEMP1 plays a role in maintaining the immature status of cells in the SZ, similar to HMGB2[95].

Thus, EFEMP1 is specifically expressed in the SZ of mature human and mouse articular cartilage, and its expression is profoundly reduced with increasing age. EFEMP1 has a role in maintaining the immature status of cells in the cartilage SZ. The loss of EFEMP1 is linked to the loss of SZ chondrocytes and compromises the integrity of the cartilage surface. Thus, EFEMP1 may be an HMGB2 target protein, and strategies preventing the loss of EFEMP1 expression in SZ chondrocytes or restoring the normal level of expression might have therapeutic potential in the management of patients with OA.

Conclusions

HMGB1 and 2 are involved in many aspects of joint diseases, such as RA and OA, as well as normal skeletal development. Given the roles of both HMGB2 and HMGB1 in controlling inflammatory responses, studies dissecting their respective contributions to inflammation and investigating whether their combined targeting has an additive or synergistic effect would be interesting. The multiple functions of HMGB proteins in tissue injury in other systems reveal the complex roles of these proteins as innate and endogenous regulators of

inflammation in cartilage and joints as well as their collaborative roles in cartilage hypertrophy and the maintenance of joint homeostasis.

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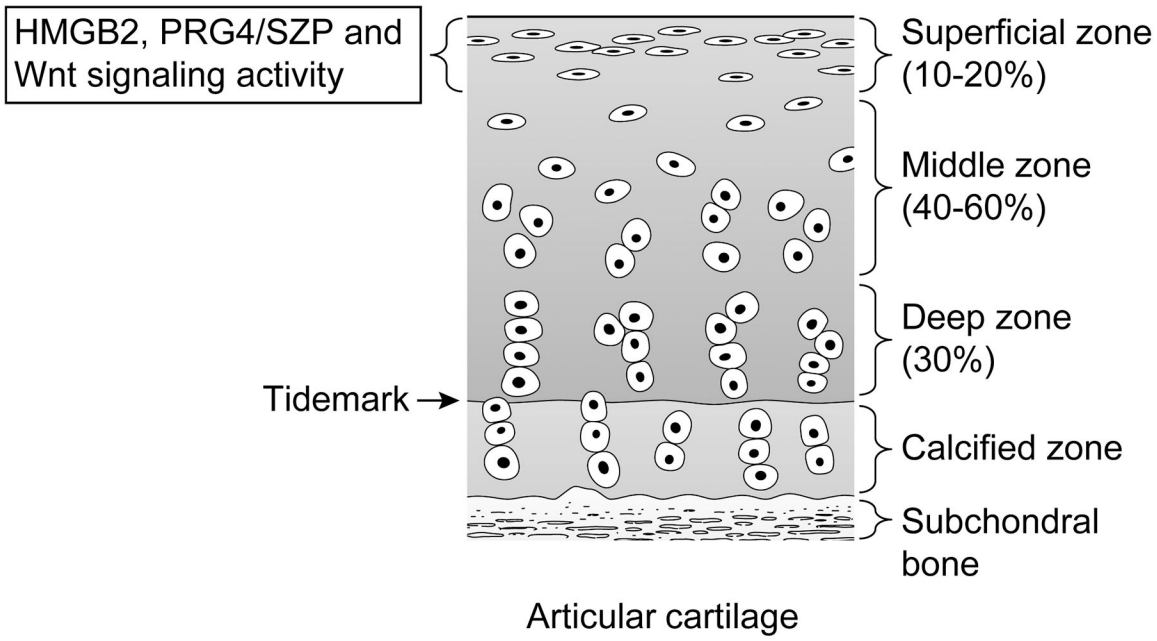

Figure 1.
Expression of HMGB2, PRG4/SZP and Wnt signaling activity in articular cartilage.

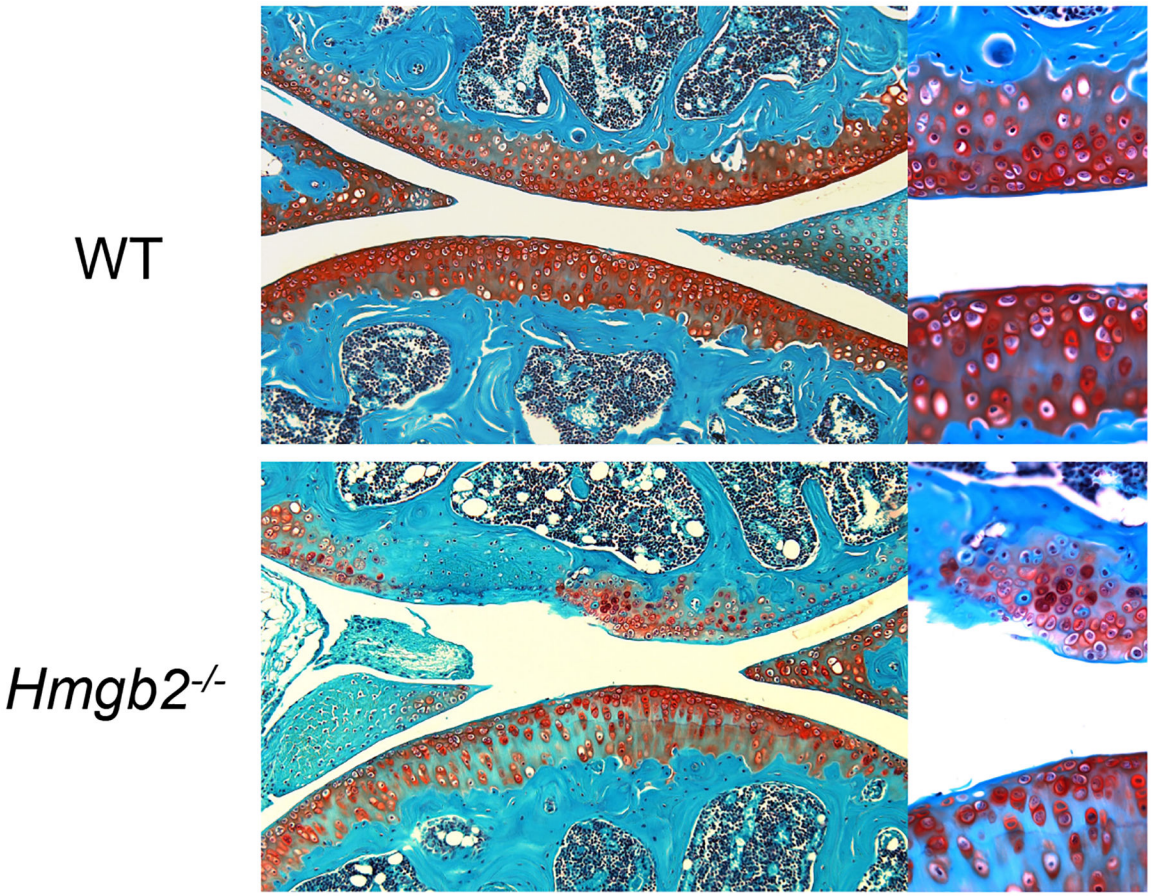

Figure 2.
Safranin O staining of the articular cartilage of knee joints from 9-month-old wild-type mice and *Hmgb2*$^{-/-}$ mice. *Hmgb2*$^{-/-}$ mice show structural cartilage defects.

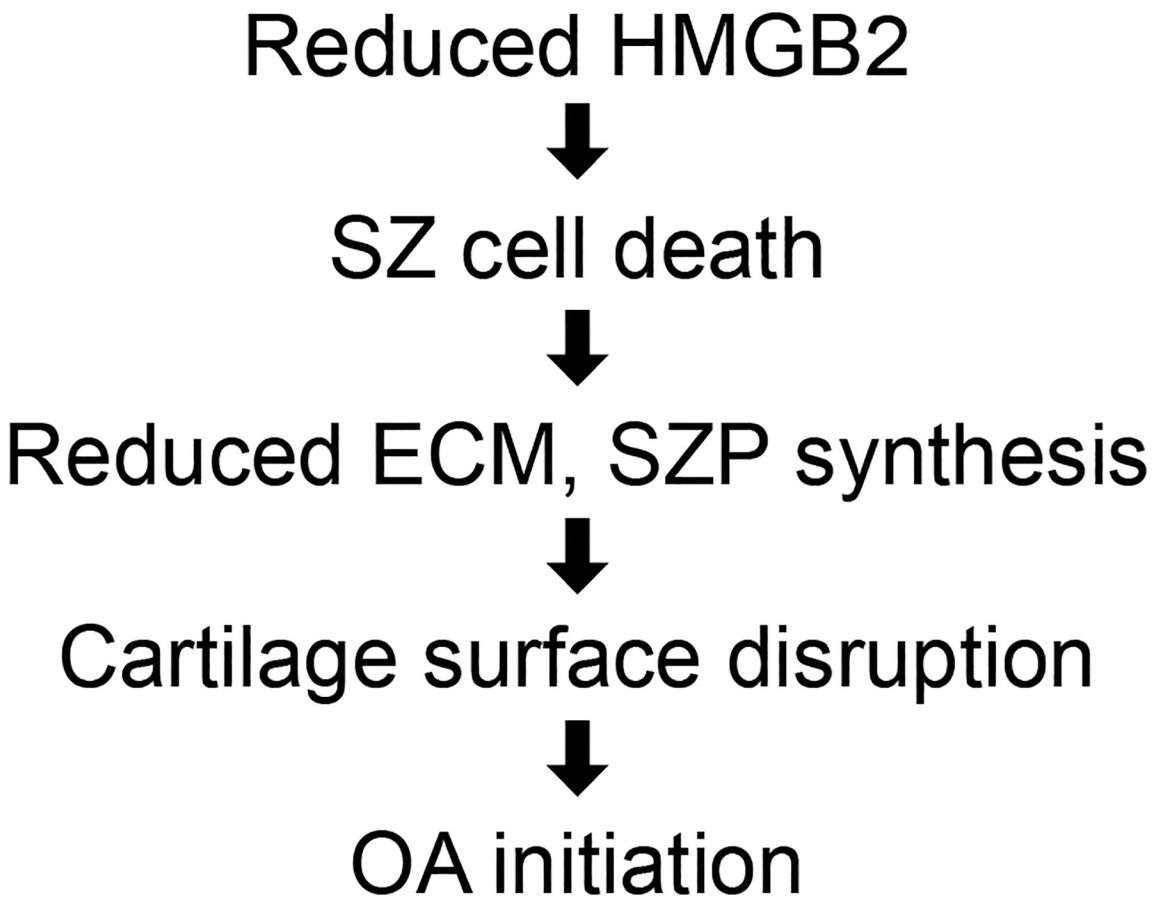

Figure 3.
The proposed mechanism of osteoarthritis associated with aging-related loss of HMGB2.

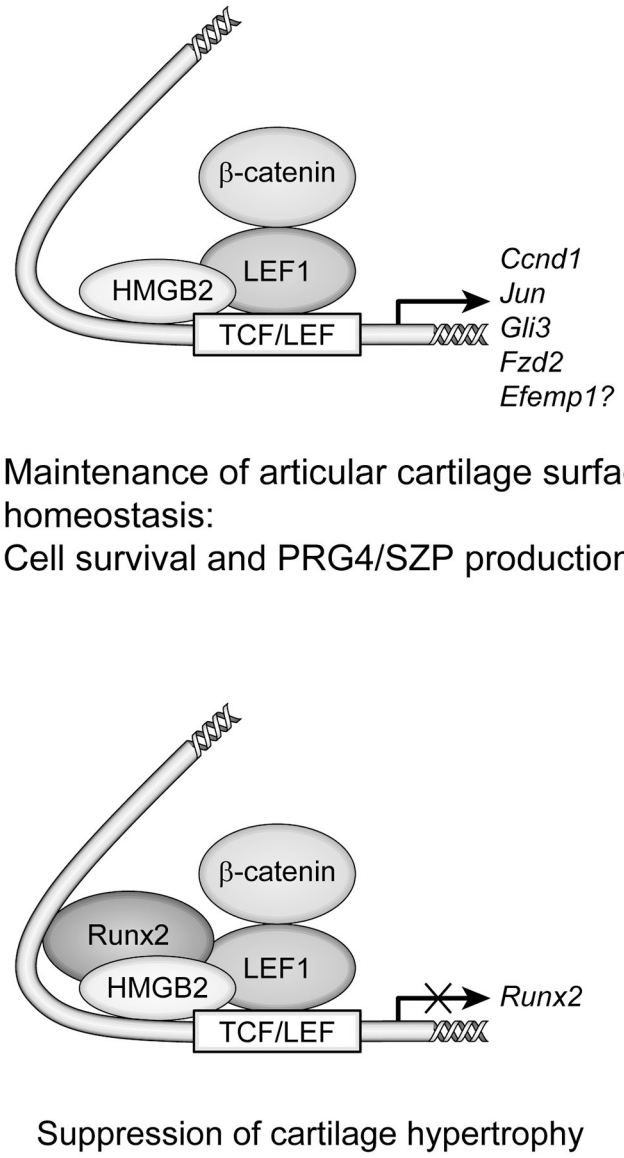

Figure 4.

Functional and physical interactions between HMGB2, RUNX2, and LEF1 on a promoter containing a TCF/ LEF motif.

Table 1

The role and function of HMGB proteins in arthritis.

HMGB protein	Disease	Location	Function	Mechanism	References
HMGB1	rheumatoid arthritis	extracellular	synovial inflammation	binds to RAGE, TLR2, and TLR4 to activate the NF-κB	34, 40, 41
	osteoarthritis		cartilage and bone destruction	pathway and signal transduction through JNK and p38	29, 30
	juvenile idiopathic arthritis		induction of proinflammatory cytokine and chemokine expression		31, 32, 33
	crystal-induced arthritis				34
	psoriatic arthritis				35
	spondyloarthritis				36
HMGB2	osteoarthritis	nuclear	chondrocyte survival and homeostasis	binds to LEF1 to enhance Wnt signaling	76
				maintains SZ homeostasis via EFEMP1	95
			suppression of cartilage hypertrophy	binds to LEF1 and RUNX2 to suppress RUNX2 activity	85