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Review

Wnts talking with the TGF-β superfamily: WISPers about modulation of osteoarthritis

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Abstract

The Wnt signalling pathway is gaining increasing attention in the field of joint pathologies, attributable to its role in the development and homeostasis of the tissues found in the joint, including bone and cartilage. Imbalance in this pathway has been implicated in the development and progression of OA, and interference with the pathway might therefore depict an effective treatment strategy. Though offering multiple opportunities, it is yet to be decided which starting point will bring forth the most promising results. The complexity of the pathway and its interaction with other pathways (such as the TGF-β signalling pathway, which also has a central role in the maintenance of joint homeostasis) means that acting directly on proteins in this signalling cascade entails a high risk of undesired side effects. Therefore, interference with Wnt-induced proteins, such as WISP1, might be an overall more effective and safer therapeutic approach to inhibit the pathological events that take place during OA.

Key words: osteoarthritis, Wnt signalling, TGF-β, signalling, crosstalk, WISP1/CCN4, joint pathology

Rheumatology key messages

- Canonical Wnt signalling plays an active role in the development of OA pathology.
- Although providing opportunities, Wnt/TGFβ crosstalk still is too incomprehensible to serve as therapeutic target in OA.
- Wnt-induced protein WISP1 seems important in osteoarthritic processes, providing an attractive therapeutic target.

Osteoarthritis

Today, >10% of the world's population above 60 years is suffering from the impact of OA [1]. OA is a chronic degenerative joint disease, characterized by functional impairment and inflammation, leading to pain, stiffness and loss of mobility [2]. Several genetic and environmental factors have been implicated in the degeneration of the joint [3–6]. While the prevalence of OA increases with age, risk factors like abnormal joint loading and obesity additionally contribute to OA aetiopathology [7, 8]. In the past, OA was considered a disease solely leading to degeneration of articular cartilage. However, this paradigm has shifted towards the idea that changes in the bone (including sclerosis of the subchondral bone and osteophyte formation) and inflammation and fibrosis in the synovium are likely involved in the disruption of normal joint homeostasis [9].

Increasing expectancy of life and obesity will increase the incidence of OA, making it even more important to find effective treatment strategies for OA. With this in mind, various signalling pathways have gained increasing attention over time, including the Wnt and TGF-β signalling pathways. Wnt signalling is involved in both formation and turnover of cartilage and bone and is characterized as being pro-fibrotic. The active role that Wnt signalling seems to play in the aetiopathology of OA has recently been adequately reviewed by Lories *et al.* [10]. In particular, increased activation of the β-catenin–dependent canonical Wnt pathway has been implicated in OA. Studies in our lab have pointed to Wnt1-induced secreted protein-1 (WISP1), a matricellular protein that belongs to the CCN (connective tissue growth factor, Cyr61, NOV) family, as a possible central Wnt-induced protein that mediates the detrimental effects of overactive canonical Wnt signalling in the joint [11–13]. Interestingly, Wnt signalling shows

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profound crosstalk with other pathways that are involved in the development and homeostasis of joint tissues, including the TGF-β signalling pathway. TGF-β has been attributed a crucial role in maintaining joint and cartilage homeostasis, and deregulation of this signalling pathway is linked to various aspects of OA pathology [14].

This review will briefly summarize current knowledge about Wnt signalling and its possible implications in OA. Next, we will point out two aspects of Wnt signalling that can serve as possible therapeutic targets. First, we will discuss the crosstalk between Wnt and TGF-β signalling as a target for therapy. In addition, we will highlight the canonical Wnt signalling–induced molecule WISP1 as a possible target for modulating OA.

Wnt signalling

First identified in 1982 [15], Wnt proteins are today known to have crucial functions in most species [16]. Wnt signalling is involved in multiple biological processes, including embryonic development, organogenesis and tissue homeostasis. Not surprisingly, dysfunction of this signalling pathway is associated with a number of diseases [17]. The family of Wnt ligands consists of at least 19 members, characterized as glycoproteins that need to undergo post-translational modification in order to carry out their signalling activity [18–21].

Wnt proteins can signal via the β-catenin/canonical and various non-canonical pathways. The canonical pathway is characterized by the translocation of β-catenin into the nucleus, whereas β-catenin–independent cascades are collectively classified as non-canonical. Since several Wnt proteins have the capacity to signal via both pathways [22, 23], signalling is increasingly being considered dependent on the cellular context in addition to the features of individual Wnt proteins.

The main cell surface receptors for all Wnt signalling pathways are the Frizzled (Fz) receptors [24, 25]. In order to activate the canonical signalling pathway, binding of a Wnt ligand to both its Fz receptor and the co-receptor low-density lipoprotein receptor-related protein (LRP)5/6 is required. This triggers the release of β-catenin from its inhibitory complex [consisting of glycogen synthase kinase-3 (GSK3-β), caseine kinase 1-α (CK1α), adenomatous polyposis coli (APC) and Axin2], which in the off-state phosphorylates and ubiquitinates β-catenin, leading to proteosomal breakdown [26]. In the on-state, Axin is relocated to the cytoplasmic tail of LRP5/6, where it forms a complex with Dishevelled (Dvl) and GSK3-β. That results in the release of β-catenin, which accumulates and translocates into the nucleus where it binds the TCF/LEF transcription factors, converting them from repressors to transcriptional activators causing the transcription of target genes, including WISP1 [27]. Non-canonical Wnt signalling includes the Ca^{2+} and planar cell polarity pathways [28–31], which engage in the regulation of cell mobility, differentiation and communication [32–34]. While at present the majority of studies focus on the role of canonical Wnt signalling in OA, non-canonical signalling might add to the complexity of the processes involved because it can affect multiple cell types present in the joint and is known to counter-regulate canonical Wnt signalling [35, 36].

All Wnt signalling pathways are tightly regulated by a set of inhibitors, including the family of secreted Frizzled-related proteins (sFRP) 1–4 and Wnt inhibitory factor (WIF), which act as soluble scavengers of Wnt ligands. In addition, members of the Dickkopf (DKK) family and sclerostin can bind to LRP5/6, interfering with its ability to interact with Wnt-Fz, thereby specifically inhibiting the canonical Wnt signalling pathway.

The implications of Wnt proteins in OA

Balanced canonical Wnt signalling is critical for the correct development and function of the tissues that are present in the joint [37, 38] and dysfunctional Wnt signalling might disturb developmental processes and increase susceptibility to OA.

Canonical Wnt signalling has been shown to affect chondrogenic differentiation of cells at various developmental stages and to play a crucial role in skeletal development [39]. Furthermore, active β-catenin signalling is important for the formation of the secondary ossification centre and epiphysal cartilage development [40]. While canonical Wnt signalling was found to inhibit chondrogenesis during early developmental stages [41–43], it promotes hypertrophic differentiation of mature chondrocytes [44–46]. In contrast, activation of non-canonical signalling stimulates early chondrogenesis, while inhibiting terminal differentiation [47]. Finally, it has been suggested that canonical Wnt signalling stimulates osteoblast maturation [41, 46].

Various groups have stressed the requirement of Wnts for correct joint homeostasis and that imbalanced Wnt signalling is linked to the development of OA. Despite some discrepancies [48], several studies have provided strong genetic evidence for the association between single nucleotide polymorphisms (SNPs) in the gene encoding the Wnt inhibitor sFRP-3/FRZB and OA. It has been suggested that disease-associated variants show decreased binding affinity for Wnt ligands, thereby exhibiting a reduced capacity to antagonize β-catenin signalling [49]. Another genetic study confirmed the role of Wnt signalling in OA because a polymorphism in the Frizzled co-receptor LRP5 was associated with an increased risk of spinal OA [50]. Mutations in LRP5 prevent the Wnt signalling antagonists sclerostin and DKK1 from binding [51–53], thus inducing Wnt-related pathology. Accordingly, several studies have shown that LRP5 variants influence human bone density [54–56], and a homozygous mutation in LRP5 was associated with osteoporosis–pseudoglioma syndrome [55].

While polymorphism studies established a significant link between mutations in proteins of the Wnt signalling cascade and OA, these assumptions were furthermore reinforced by gene expression analyses. The association found between a mutation in the Frizzled co-receptor LRP5 and OA [50] could be supported by a study linking deficiency of LRP5 to joint pathology [57]. Absence of

FRZB resulted in increased expression of matrix metalloproteinases (MMPs) and augmented cartilage damage *in vivo* in experimental OA [58]. Cartilage loss was associated with increased β-catenin levels, suggesting that FRZB plays a regulatory role in β-catenin accumulation [59]. A link between increased β-catenin activity and OA pathology could furthermore be established, because β-catenin accumulation was found in the superficial cartilage layers in several OA models [11] and in areas of degeneration [60–63]. The pathological effect of this differential gene expression was explained as due to activation of β-catenin because it stimulated OA hallmarks, such as hypertrophy, matrix mineralization, expression of MMPs and vascular endothelial growth factor in mature cartilage cells. In a study by Zhu *et al.*, inducible overexpression of β-catenin in mature chondrocytes resulted in increased expression of the maturation markers collagen type X and osteocalcin, increased levels of MMPs and strongly increased cartilage degeneration [62]. A similar experimental setup showed that β-catenin induced articular cartilage thickening, and increased cell density and proliferation [64]. Meanwhile, while their presence in cartilage is confirmed, no (or only subtle) differential expression could be detected for Wnt proteins in cartilage of OA models [11, 65]. Dell'Accio *et al.* [65] found an upregulation of Wnt16 in human OA cartilage; however, no differential expression could be confirmed for other Wnt proteins. Interestingly, whereas we could not find differentially regulated Wnt ligands in the cartilage [11], we observed strongly increased expression of several Wnt ligands, including Wnt2b and Wnt16, in the synovium in experimental OA. These findings are of particular relevance in light of OA as a disease of the entire joint. Notably, the Wnt-induced protein WISP1 was increased in both the synovium and the cartilage, suggesting migratory capacities of Wnts produced in the synovium are enabling canonical Wnt signalling in the cartilage [11]. Synovial overexpression of Wnts resulting in canonical Wnt signalling as well as WISP1, induced OA-like cartilage lesions as early as 7 days after overexpression [13]. In addition, the Wnt signalling cascade has been linked to the induction of cytokine expression. Overexpression of Wnts in fibroblasts results in increased expression of IL-6, IL-15 and TNF-α, but also of MMPs [66–68]. Another study showed that Wnt3a enhanced the effect of IL-1β, stimulating the loss of proteoglycans from the matrix [69]. The production of pro-inflammatory cytokines and matrix-degrading enzymes in the synovium has been attributed an important role in the process of OA, as elegantly reviewed by both Berenbaum [70] and Scanzello and Goldring [71].

Less well studied is the role of Wnt signalling in other features of OA pathology, including ectopic bone formation and fibrosis. Nevertheless, canonical Wnt signalling has been described as a central pathway in bone formation [72–76]. In this context, DKK1-mediated inhibition of the canonical Wnt pathway was associated with decreased bone mass [77]. Moreover, Wnt activity orchestrates osteoblast maturation [41, 46], whereas Wnt signalling decreases osteoclast activity [74]. Hence, the balance is shifted towards anabolism, suggesting an active role of canonical Wnt signalling in osteophyte formation and subchondral plate sclerosis during OA [62, 72, 78]. Furthermore, many OA patients suffer from severe fibrosis in the synovium, which is a major contributor to joint stiffness [79]. In the past, Wnt signalling has been linked to many types of fibrosis, including pulmonary [80–83], liver [84–86] and renal fibrosis [87–89]. However, detailed discussion is beyond the scope of this paper.

Altogether, these data suggest that increased canonical Wnt signalling is not merely present in OA, but in fact plays an active role in the development of OA pathology. Despite the destructive role of excessive canonical Wnt signalling in cartilage, extreme care should be taken in inhibiting this process. Balanced Wnt signalling has been shown to inhibit chondrocyte apoptosis and has therefore been recognized as having a protective role [90–92]. In support of this, conditional knock-down of β-catenin induced chondrocyte apoptosis and cartilage destruction in mice [93]. Chondrocyte apoptosis could be detected upon inflammation-induced elevation of DKK1 expression; the associated OA phenotype underlines the necessity of Wnt signalling for cell survival [94]. Furthermore, inhibition of the Wnt signalling antagonist DKK1 showed its protective potential for cartilage and bone in a rat OA model [95]. In contrast, various other studies using overexpression of the Wnt signalling inhibitor DKK1 clearly demonstrate the protective role of DKK1 in OA [72, 78].

In summary, increased β-catenin activity has been shown to induce cartilage destruction by affecting the chondrocyte phenotype and stimulating the expression of pro-inflammatory cytokines and proteases. However, blocking of excessive signalling might trigger equally detrimental effects, as has been suggested in chondrocyte viability studies. Thus, a correct balance of active Wnt signalling is ultimately required to ensure healthy joint tissue, making targeting of upstream Wnt signalling complicated. Possible leads for therapeutic intervention might be found in the profound crosstalk that Wnt signalling has with other pathways, such as the TGF-β signalling pathway.

Crosstalk between the Wnt and TGF-β signalling pathway

Whereas the importance of balanced Wnt signalling for maintenance of the tissues in the joint has been described in the previous section, similar importance in maintaining joint homeostasis has been attributed to the TGF-β signalling pathway. Interestingly, while canonical Wnt signalling enhances the rate of chondrocyte maturation, TGF-β signalling via ALK5, resulting in Smad2/3 phosphorylation, potently blocks chondrocyte hypertrophy. Additionally, TGF-β has been shown to bind to the ALK1 receptor, which results in Smad1/5/8 phosphorylation, a pathway that is known to mediate BMP signalling and

which is associated with chondrocyte hypertrophy and hallmarks of cartilage destruction [96–99].

Several studies have highlighted a profound interplay between Wnt signalling and TGF-β signalling and have shown that this interaction orchestrates the processes in which both signalling pathways are involved [100, 101]. However, the molecular mechanisms and impact of the communication between the Wnt and TGF-β pathways are largely unknown. In this section we present up-to-date knowledge about the complex interplay between the pathways at the extracellular, cytoplasmic and nuclear level, and this is schematically summarized in Fig. 1.

Extracellular Wnt/TGF-β crosstalk

TGF-β and Wnt ligands have already been shown to regulate each other's expression during early development. Wnt8c induces the expression of the TGF-β family member Nodal [102], whereas a study in *Xenopus* found Wnt8 expression to be regulated by TGF-β family members [103] (Fig. 1, extracellular compartment, where modulators of extracellular signalling are shown in yellow). On the other hand, BMP-2 enhanced chondrogenesis by downregulating Wnt7 and β-catenin expression [104]. An additional study revealed the requirement of β-catenin for BMP-4 expression in human cancer cells [105]. Several extracellular proteins, including sclerostin, CTGF and sFRPs, modulate and are modulated by receptors or ligands of both the Wnt and TGF-β pathways [106–110]. Among others, sclerostin was shown to antagonize canonical Wnt signalling by binding to the receptor LRP5/6. Notably, inhibition via sclerostin was, in turn, shown to be modulated by BMPs [106, 107]. Like sclerostin, CTGF modulates Wnt signalling [109] and exhibits binding capacity for both BMP-4 and TGF-β. Interestingly, whereas binding of CTGF to BMP receptors reduces the activity of BMP-4, TGF-β activity is enhanced upon binding to CTGF [108]. In contrast, CTGF expression has been shown to be tightly regulated by Wnt3A and BMP-9, which have been suggested to be crucial in normal osteoblast differentiation [111].

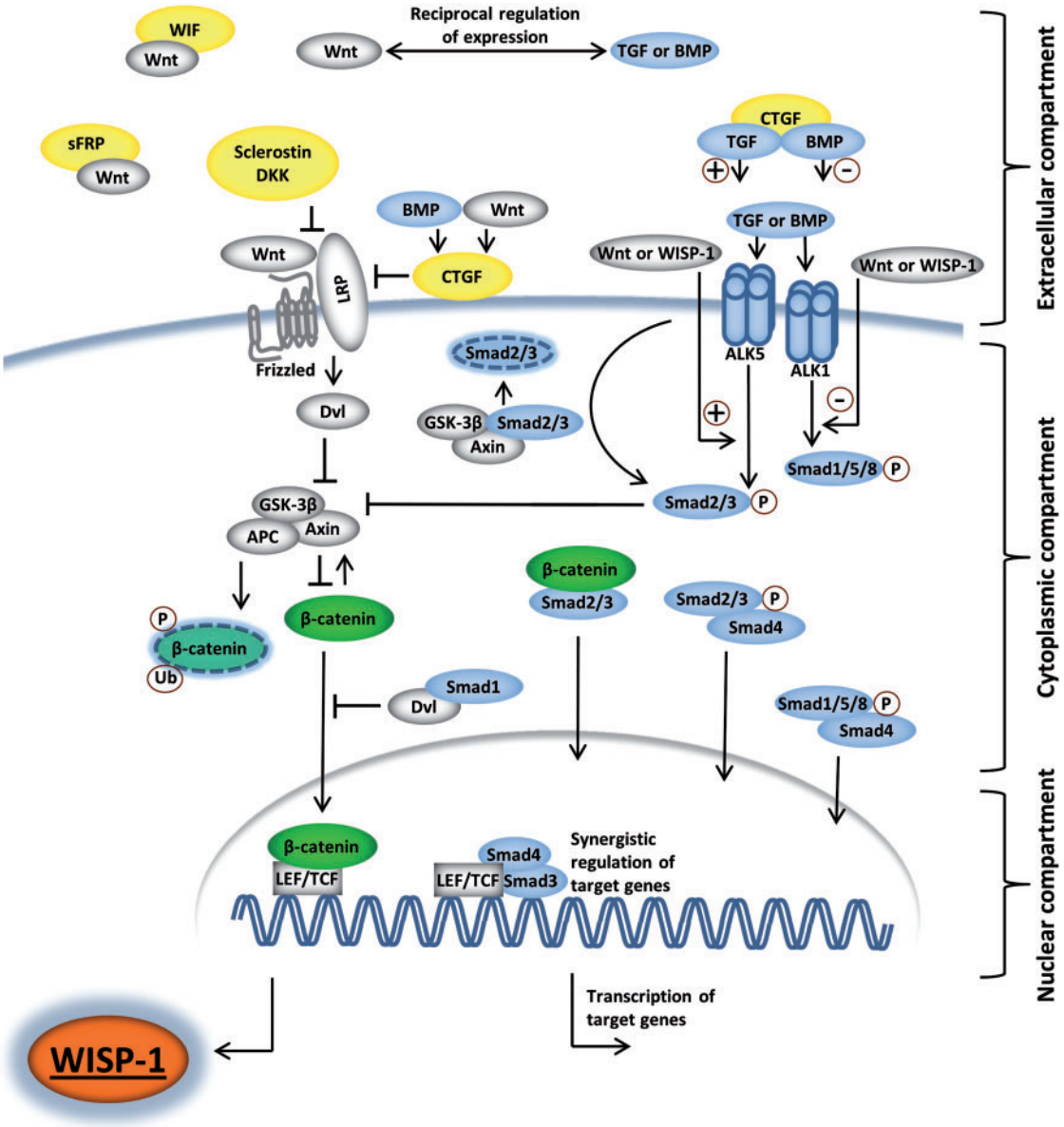
Communication in the cytoplasm

In addition to the active crosstalk between the extracellular compartments of Wnt and TGF-β signalling, complexity of interaction between both pathways is further enhanced on the intracellular level. (An overview of the interactions described in this section can be found in Fig. 1, cytoplasmic compartment.) In the cytoplasm of adult human mesenchymal stem cells (MSC), TGF-β induces nuclear translocation of β-catenin in a Smad3-dependent manner, thereby increasing activation of canonical Wnt signalling. β-catenin thus stimulates the proliferation of MSCs and inhibits their osteogenic differentiation via TGF-β [112]. Synergistic signalling was found to stimulate chondrocyte differentiation and to inhibit adipocyte gene expression [113]. The effect of the Wnt pathway on chondrogenesis was further investigated by Im and Quan [114], who demonstrated enhancement of early chondrogenesis upon application of Wnt inhibitors. The effect of the Wnt inhibitor sFRP-1 on chondrogenic expression levels was, however, obscured by TGF-β during long-term culture, suggesting that TGF-β overrides the effect of β-catenin during later chondrogenesis. In murine MSCs, BMP-7 was introduced to regulate chondrogenic and osteogenic differentiation. Simultaneously, BMP-7 inhibits Wnt11 and BMP-4 expression, whereas Wnt5b expression was found to be upregulated during chondrogenic differentiation [115]. Therefore, even during the early developmental stages, a tight regulation of the crosstalk between Wnt and TGF seems essential for accomplishing the normal course of bone and joint development. In contrast, BMP-2 was found to inhibit Wnt signalling and osteoblast differentiation in mouse MSCs. In this context, an interaction between Smad1 and Dvl, occurring in the presence of BMP-2, prevented nuclear translocation of β-catenin [116]. It has been suggested that signalling between the pathways is further modulated via the Axins and GSK3-β. Active canonical Wnt signalling increases the expression of Axin. In contrast, TGF-β–induced Smad signalling decreases its expression [117], and in doing so it releases a brake on canonical Wnt signalling. Both proteins were found to be essential for Smad3 phosphorylation [118]. Another study showed that binding of non-phosphorylated Smad3 to Axin and GSK3-β results in its basal degradation [119]. In line with the β-catenin–dependent inhibition of Smad2/3 phosphorylation via the Axins, we recently found that canonical Wnt3a decreased levels of phosphorylated Smad2/3 [12]. In addition, Wnt3a and WISP1 increased Smad1/5/8 phosphorylation, thus possibly inducing chondrocyte hypertrophy. In line with this, other studies showed that WISP1 was able to inhibit Smad2 phosphorylation in human bone marrow stromal cells [120], while inducing Smad1/5/8 signalling [121], which can affect the chondrocyte phenotype. (These interactions are shown at the right side of the cytoplasmic compartment in Fig. 1.)

Interactions in the nucleus

In addition to their cytoplasmic interaction, the Wnt and TGF signalling pathways communicate within the nucleus (Fig. 1, nuclear compartment), wherein they synergistically regulate target genes. Co-regulation by these pathways has been established for several genes involved in developmental processes [122–125] as well as during tumour development and progression [126, 127]. A study in mouse gastric cancer cells shows that the mouse gastrin promoter is synergistically regulated by the Wnt and TGF pathway. Smad3/Smad4 and Lef/Tcf can, thereby, mutually act as co-factors while their interaction is stabilized by the p300 co-activator protein. Notably, either the Smad or Lef/Tcf binding side is sufficient to recruit the transcriptional activation complex [128]. Synergistic activation of LEF1/TCF by the TGF-β and Wnt signalling pathways was confirmed by Letamendia *et al.* [129]. Furthermore, Smads and Wnts regulate their expression interdependently, because β-catenin was found to inhibit TGF-β signalling in chondrocytes [117]. In turn, Li *et al.* [130] demonstrated that Smad3 signalling could induce

FIG. 1 Profound crosstalk between TGF-β and canonical Wnt signalling is present on various levels in the cell



A schematic overview of the many interactions that TGF-β and canonical Wnt signalling have, both outside and inside the cell. Proteins involved in the Wnt signalling pathway are depicted in grey, with β-catenin as a central protein in canonical Wnt signalling depicted in green. Furthermore, proteins involved in TGF-β signalling are indicated in blue. Additionally, extracellular modulators of cell signalling are shown in yellow. Finally, WISP1 is a Wnt/β-catenin–induced protein that plays a central role and is depicted in orange.

β-catenin signalling in murine neonatal sternal primary chondrocytes.

Clear crosstalk between canonical Wnt signalling and TGF-β signalling that affects the disease outcome has been demonstrated in fibrotic disorders, and this might similarly apply to OA pathology. While TGF-β is a well-known pro-fibrotic factor, several studies have shown Wnt activation in TGF-β–induced fibrogenesis [131, 132]. Akhmetshina *et al.* [132] demonstrated that TGF-β stimulates canonical Wnt signalling in cultured fibroblasts. TGF-β–mediated activity of β-catenin has previously been shown to be induced via Smad3 [131]. Furthermore, inhibition of the canonical Wnt signalling pathway effectively blocked TGF-β receptor 1–driven fibrosis [133], while Wnt signalling was activated downstream of TGF-β in fibroblasts during wound repair [134]. Furthermore, TGF-β induced WISP1 expression in pulmonary fibrosis and liver fibrosis [135, 136]; TGF-β itself was found to be regulated by the microRNA miR-92a [136]. Furthermore, WISP1 showed upregulated gene

expression in TGF-β–induced myofibroblast differentiation [137], strengthening the purported role of crosstalk between TGF and Wnt in fibrotic processes.

Altogether, these findings indicate that communication between TGF and Wnt signalling proteins plays an important role in processes that act in the regulation of joint tissue homeostasis and are therefore likely to be important in the development of OA pathology. Crosstalk was found not only in cartilage, influencing chondrogenesis and chondrocyte hypertrophy, but was furthermore found in fibrogenesis and osteogenesis. Affecting all tissues involved in the whole-joint disease OA, crosstalk between TGF and Wnt signalling may have the exciting potential for therapeutic purposes. However, due to the complex nature of both the separate pathways and the profound crosstalk that takes place on many levels, it is difficult to determine the net outcome of therapeutic interventions. Therefore, targeting this crosstalk is not feasible until enough is known about the consequences of interference. In addition, since Wnt signalling is extremely complex and tightly regulated and can affect many biological processes, upstream targeting of Wnt signalling is likely to produce undesired side effects. Thus, a downstream target might be a more appropriate therapeutic target, less likely to produce side effects. In this context, WISP1 has drawn increasing attention as a feasible target for OA therapy.

WISP1 as central mediator of Wnt signalling in OA pathology?

WISP1 (CCN4) is one of six members of the cysteine-rich CCN family, which are characterized by four distinct functional domains: next to an insulin-like growth factor-binding protein-like module, all members except CCN5 consist of a von-Willebrand factor type C repeat, a cysteine-rich C-terminal module and a thrombospondin type 1 repeat (TSP1). The CCN protein family has been assigned to the group of matricellular proteins, characterized by their function in extracellular signal modulation and coordination [138]. Various biologic processes have been indicated to be modulated by CCN proteins: among others, tumourigenesis, chondrogenesis, osteogenesis, angiogenesis, apoptosis and haematopoiesis [139]. Given the many processes that CCN proteins are implicated in, it is evident that they are furthermore involved in numerous pathologies, including various cancers and fibrotic disorders [140–143].

The importance of WISP1 in the context of skeletogenesis has been demonstrated, since a particularly high expression of WISP1 was found at sites of new bone formation and in healing fracture calluses. Additionally, WISP1 was shown to promote BMP-2-mediated osteoblast differentiation *in vitro*, while repressing chondrocytic differentiation [144].

Recently, WISP1 has gained attention as a promising target for OA therapy research because a SNP in WISP1 has been associated with spinal OA, and in particular with the severity of endplate sclerosis [145]. Additionally, an earlier study of ours [11] demonstrated that WISP1 was significantly upregulated in experimental human OA cartilage and synovium. Since no upregulation of Wnt proteins could be detected in OA cartilage [11, 65], it was suggested that WISP1 was produced as a response to Wnts that have diffused into the cartilage after expression in other tissues such as synovium.

Linking WISP1 expression to cartilage destruction, WISP1 was shown to trigger the release of matrix-modulating enzymes such as MMPs and aggrecanases from macrophages and chondrocytes, independent of IL-1 [11]. In line with this, we [12] showed that WISP1 can skew TGF-β–induced Smad signalling towards dominant signalling via Smad1/5/8, which can result in increased hypertrophic differentiation of chondrocytes. Previously, WISP1 has been shown to regulate the TGF-β pathway to control osteoblast function in human bone marrow stromal cells [120]. Additionally, a study by Ono *et al.* [121] shows that WISP1 can potentiate BMP-2 effects on osteogenesis by increasing Smad1/5/8 phosphorylation. The same study demonstrated that WISP1 potentiated osteogenesis in transgenic mice.

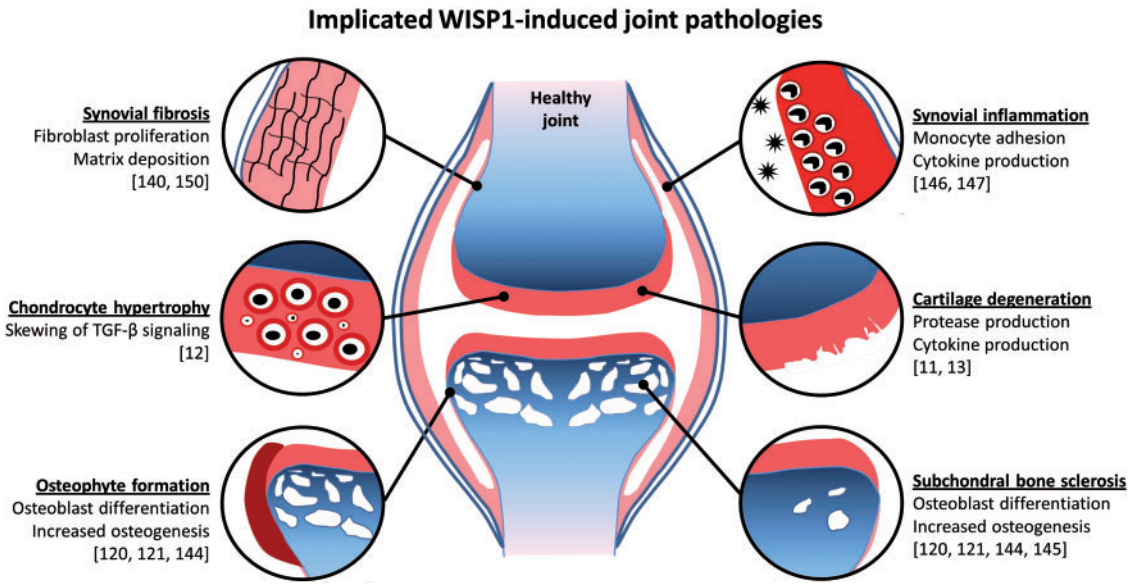
Additional studies suggest a critical role of WISP1 in synovial tissue. WISP1 was found to stimulate a pro-inflammatory response in macrophages of human adipose tissue and to increase the expression of IL-6 in human synovial fibroblasts, via activation of PI3K, Akt and NFκB [146]. Furthermore, WISP1 induced the expression of vascular cell adhesion molecule-1 in human OA synovial fibroblasts [147], promoting monocyte adhesion. This could promote synovitis, because mononuclear cell migration has previously been identified as an important modulator in synovial inflammation [148, 149]. Furthermore, WISP1 plays a role in fibrotic processes, in which it is shown to stimulate fibroblast proliferation along with matrix protein expression in cardiac fibroblasts [150]. An additional study revealed WISP1 as a potential therapeutic target in pulmonary fibrosis, modulating the expression of genes known to be involved in fibrosis towards an attenuation of lung fibrosis [140].

These results show that WISP1 might be an appealing downstream target of canonical Wnt signalling to study in the field of OA. The studies described in this section make it likely that WISP1 is involved in the induction of OA-associated processes like cartilage damage and osteophyte formation, changes in the subchondral bone, and fibrogenesis and inflammation of the synovium. These implications about how WISP1 might be involved in the induction of OA pathology are shown in Fig. 2. However, the exact pathway by which WISP1 modulates the tissues in the joint is still largely unknown. Progress in understanding the underlying mechanism of WISP1-induced joint destruction might reveal an attractive alternative therapeutic approach.

Conclusion

Imbalance of Wnt signalling results in the development of OA pathology. Canonical Wnt signalling, therefore, has

Fig. 2 Proposed implications for WISP1 in the osteoarthritic joint



The Wnt/β-catenin–induced protein WISP1 is strongly overexpressed in both the synovium and cartilage during experimental and human OA. Based on the literature and our own findings, we propose that WISP1 is likely to play a role in many processes seen during OA. These include the skewing of TGF-β signalling towards dominant signalling via Smad1/5/8, which can lead to chondrocyte hypertrophy and cartilage degeneration (the latter via increased production of proteases that can break down the cartilage matrix). In addition, WISP1 has been shown to induce fibrogenesis and inflammation by both monocyte adhesion and cytokine production. Finally, WISP1 is a well-known inducer of osteoblast differentiation and a potent inducer of osteogenesis, which can result in increased osteophyte formation and sclerosis of the subchondral bone in the joint.

been proposed as a possible tool for the re-establishment of joint homeostasis. However, due to the complexity of the pathway, extreme care should be taken when interfering with proteins in the upstream Wnt signalling cascade, which could be prone to affecting other biological processes or triggering undesired side effects. Thus, though that may be a possible future therapeutic approach, intervention in the profound crosstalk between Wnt and TGF signalling pathways is not feasible until knowledge has reached such a level that negative effects can unerringly be excluded. Here, we would like to put forward WISP1 (a Wnt/β-catenin–induced protein) as a promising therapeutic target, because WISP1 is thought to be involved in the processes that are present during the course of OA. In addition, because WISP1 is a more downstream target in Wnt signalling, targeting this protein is expected to give fewer undesired side effects. Because the pathway that WISP1 uses to affect the joint tissues has not yet been elucidated, targeting WISP1 with blocking antibodies currently seems to be the most feasible option.

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