Chapter

Ankylosing Spondylitis Pathogenesis and Pathophysiology

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Abstract

The pathogenesis and pathophysiology of Ankylosing Spondylitis (AS) is complex and remains only partially understood. Contributory genes including a variety of HLA-B27 subset genes and many other non-HLA genes are implicated in the literature. Novel genes and gene–gene interactions being a continuously evolving area of AS research. Dysregulation of the enteric microbiome with a corresponding aberrant immunological response is recognised in research. Certain infectious agents are thought to play a role. A variety of other influences including environmental exposures, dietary and lifestyle factors and sex hormones appear to play a role in AS pathogenesis. There is emerging evidence that that pathophysiological response in AS is an elaborate combination of both autoinflammatory and autoimmune components, however the IL-17/IL-23 pathway remains the major pathway in AS according to studies to date. The specific mechanisms that lead to characteristic clinical features of AS including sacroiliitis, spondylitis, ankylosis, uveitis and other extra articular manifestations remain occult. Further research to establish these is ongoing.

Keywords: ankylosing spondylitis, HLA-B27, pathogenesis, pathophysiology, seronegative spondyloarthropathy, microbiome dysregulation in spondylitis

1. Introduction

The pathogenesis and pathophysiology of ankylosing spondylitis (AS) involves an extremely complex interplay of factors that can be broadly categorised according to the following key areas:

- Genetic predisposition
- Environmental factors
- Altered gut microbiome and infective triggers
- Enteric wall dysfunction and altered mucosal immunity
- Aberrant systemic immune response and subsequent dysregulation

- Factors associated with the axial skeleton and its entheses
- Factors associated with peripheral entheses and joints
- Aberrations in bone metabolism

This chapter will discuss the major components that contribute to AS pathogenesis. Theories and emerging evidence in the literature are reviewed. Whilst the discussion is divided into various topics for clarity and ease of reading, in reality the factors involved interact in a elaborate matrix of multidirectional feedback all contributing to the ultimate manifestation of AS.

This chapter will review genetic associations with AS. In addition to HLA-B27, there are many novel genes thought to contribute to AS pathogenesis reported in the literature. Section 3.1 reveals the proposed theories of how HLA-B27 specifically leads to an altered immunological response resulting in inflammation.

Autoimmune vs. autoinflammatory immunological features involved in AS pathogenesis and the interplay between disturbancs of the gut microbiome, including by infection and diet leading to altered immune response. The IL-17-23 and IL-12 pathways are discussed in some details.

2. Pathogenesis

2.1 Genetic factors

2.1.1 HLAB27

Human Major Histocompatibility Complex (MHC) class I is also known as Human Leukocyte Antigen (HLA) and is one of the many surface proteins present on all nucleated cells and platelets in the human body [1]. MHC I plays a role in antigen presentation to cytotoxic T cells via the T Cell Receptor (TCR) [1].

Genetic factors contributing to development of AS have been recognised since 1961 leading to the discovery of the HLAB27 gene in 1973 [2]. There is significantly higher concordance between monozygous twins and dizygous twins with AS rates of 63% and 23–27% respectively [2, 3]. Over the years many genes have been identified as associated with AS, however the full picture of gene–gene interactions is yet to be explained.

HLA B27 belongs to the MCH class I receptor family. See the schematic representation in **Figure 1**.

There are 4 domains of this molecule as depicted in **Figure 1**. Regions α1 and α2 are located at the top of the protein where antigen binding occurs (**Figure 2**). The α3 heavy chain is located adjacent to the cell surface partially penetrates the cell membrane. The 4th domain is the β2 microglobulin which is covalently associated with the rest of the HLA B27 molecule [6].

Three main features distinguish HLA B27 from most other HLA class I molecules. Glutanine is substituted for methanine located at the 45 position. The second feature is an unpaired cysteine (Cys67) [7]. This feature enables formation of homodimers and oligomers of free heavy chains which are thought to contribute to development of AS [7] and is discussed in more detail later on in this chapter. Thirdly there is a Lys residue at position 70 that increases reactivity of the cysteine at position 67 [7].

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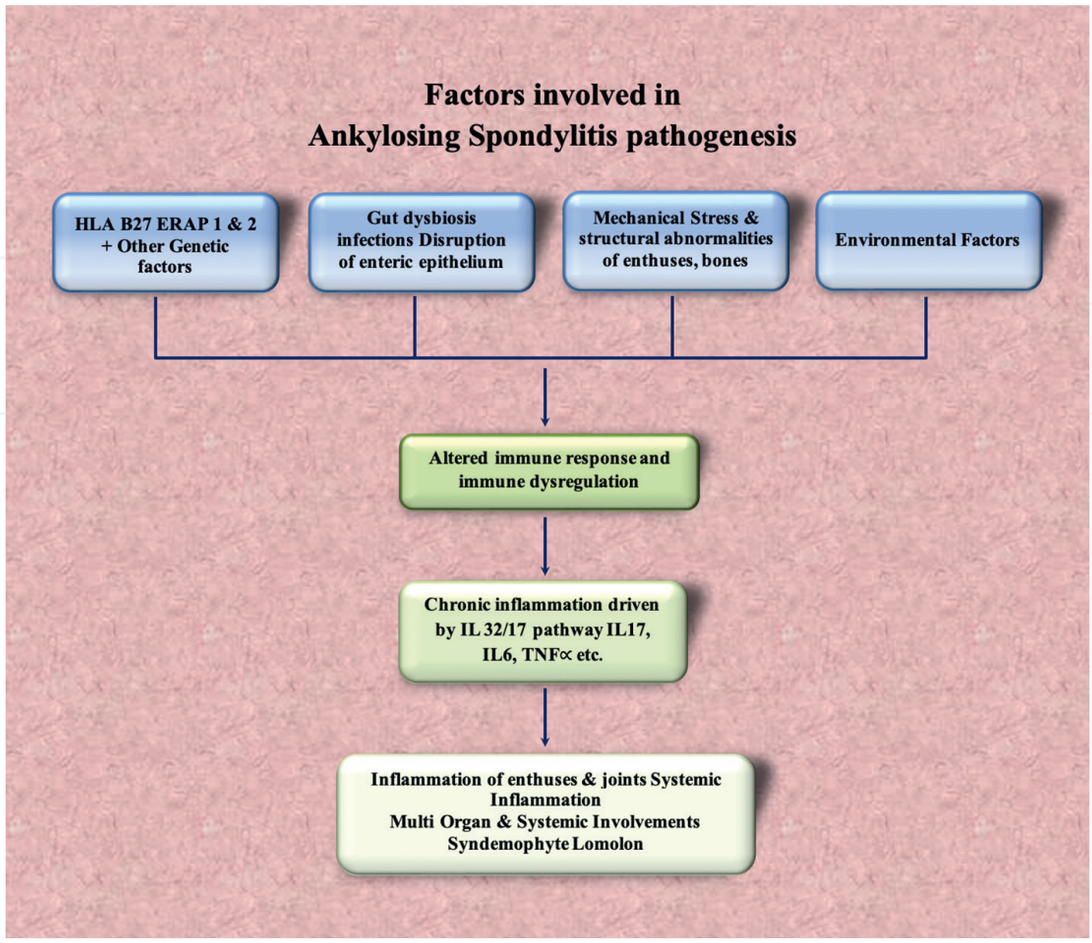

Figure 1.
Factors involved in ankylosing spondylitis pathogenesis.

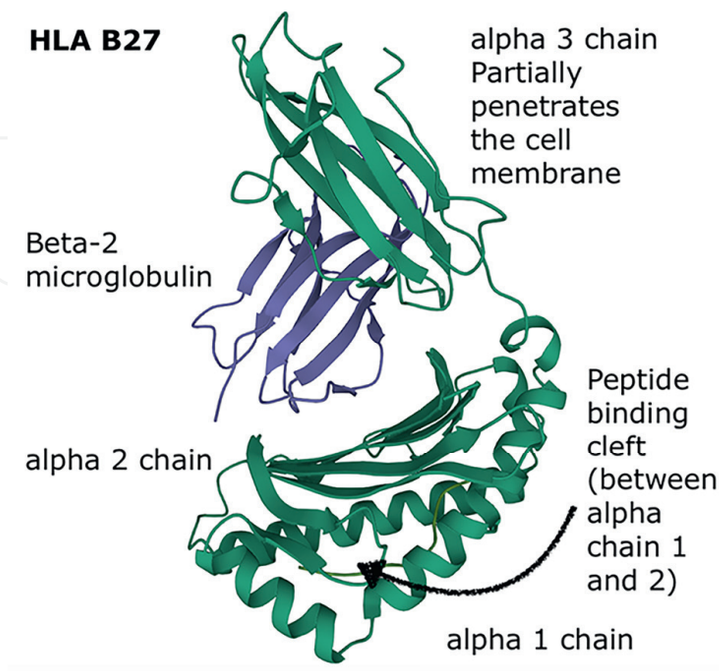

Figure 2.
HLA B27 protein structure [4, 5].

Extensive polymorphism of HLAB 27 results from the considerable variability of the heavy chain component of the protein [7].

Studies show 90–95% of AS patients are HLA-B27 positive. The chances of developing AS for an individual with an HLA-B27 gene is 1–2%. This percentage increases to 15–20% for those who have a first degree relative with AS [2]. The relative risk of developing AS for an individual with a first degree relative who has AS is 94%, for a second degree relative 25% and for a third degree relative 4% [2].

HLA B27 is a polymorphic gene with around 223 subtypes thus far identified [6]. HLAB2705 is more prevalent in affected Caucasians, HLAB2704 is associated with affected Han Chinese whilst HLAB2702 is associated with affected people of Mediterranean ethnicity [2]. HLA B2705 and HLA B2704 are the predominant subtypes in the South Indian population [8]. Inuit and native Alaskans have the highest rates of HLA B27 In the world and correspondingly the highest prevalence of AS [9].

Even though HLA B27 is the most commonly recognised genetic factor in the development of AS, its overall contribution in the development of AS is only around 20% suggesting other environmental, genetic, immune and even anatomical phenomena are responsible for pathogenesis in AS. For example, microRNAs (miRNAs) are a class of endogenous, non-coding, RNA-modulating mRNAs and are reported to regulate AS progression by interacting with genes that are potential biomarkers for AS [10].

2.1.2 Other HLA genes

Other HLA B genes have been recognised in association with development of AS including: HLA-B730 HLA-B16, HLA-B35,31,32 HLA-B38 and HLA-B3933 [2]. These genes have been identified across a variety of ethnic groups and are associated with HLA-B27-negative AS, although the mechanism is not yet clear [2].

An HLA-C amino-acid variant in addition to HLA-B*27 confers risk for ankylosing spondylitis in the Korean population. The four amino-acid positions of HLA-B and -C account for most of the associations between AS and MHC in the Korean population. This finding updates the list of AS susceptibility loci and provides new insight into AS pathogenesis mediated by MHC class I molecules [11].

2.1.3 ERAP1 ERAP2 and NPEPPS

ERAP1 (coding for endoplasmic reticulum aminopeptidase 1 (ERAP1)), ERAP2 (coding for ERAP2) and NPEPPS (coding for puromycin-sensitive aminopeptidase) have been implicated in AS [2]. This is to do with folding of HLA B27 in the endoplasmic reticulum which is discussed in further detail later on in this chapter. Gene–gene interactions between HLAB27 and ERAP1 appear to be responsible [2].

2.1.4 IL23R

More than 90% of genetic risk single nuclear polymorphisms (SNP)s are present in non-coding regions, however this is not the case with the IL23R gene. The genetic association of IL23R loci with AS was first reported in 2007. Interestingly, the same SNP also affects the risk of developing inflammatory bowel disease. The same SNP is also associated with psoriasis, another condition closely linked to AS [12].

2.1.5 Killer immunoglobulin-like receptor (KIRLR)

Studies have demonstrated that KIR3DL2 R is up-regulated on activated CD4+ T-cells and that there are increased levels of these cells in patients with AS compared with healthy controls [13]. These cells have been found upregulated in the terminal ilium of patients with early spondyloarthropathy [14]. The theories on the role of KIRLRs in AS are many with one suggesting an imbalance between inhibitory and excitatory KIR receptors in AS patients that upregulates an NK cell response [15].

2.1.6 Other genes

Over the years several genes have been reported in the literature in association with AS with new ones emerging as further research is undertaken.

Susceptibility genes have been identified that are either directly, or indirectly involved in the IL-23–IL-17 pathway, or interact with it in AS. These include IL12B, RUNX3, EOMES, TBX21, TYK2, CARD9, IL1R1, IL1R2, IL6R, IL7R, IL12B, IL27, NKX2 and PTGER4 [16]. The interleukin (IL)-1 gene cluster is an important locus associated with susceptibility to AS. CYP 2D6 [17] and ANKH genes are also associated with AS [18].

There is discussion around the association of TLR4 genetic variants and TLR4 expression levels with AS implicates innate immunity in AS pathogenesis 27,28. The concentrations of bacterial lipopolysaccharides (LPS) are increased in AS with further suggests the association with TLR4 expression and have been noted to correlate with disease activity [16]. This supports the concept that AS has autoinflammatory components in its pathophysiology.

Many more AS-susceptibility genes are likely to be identified by ongoing research.

2.1.7 Gene–gene interactions and pleiotropy

There are significant gaps in knowledge about gene–gene interactions and the development of AS. However, certain links have been established. For example, a Taiwanese population study is suggestive of an interaction between HLA-B60 and HLA-B27 as a marker for the risk of AS susceptibility [2]. The combination of HLA B60 with HLAB 27 increases chances of developing AS by 3–6 times. HLAB27 interacting with ERAP1 gene is thought to contribute to development of AS [19].

Pleiotropy in AS has been investigated. A number of pleotrophic gene loci have been identified [13]. DNA methylation genes 3a and 3b (*DNMT3A, DNM3TB*) are recognised in genomic imprinting and X-chromosome and have been studied in cross-gene studies. Their relationship with haematopoietic stem cell development and UBE2 activation (a family of genes also known to be associated with AS) supports the hypothesis of involvement in male predominance in AS, which remains currently remains unexplained [13].

FUT2 encodes fucosyl transferase, a gene which controls secretion of blood group antigens into body fluids. This gene is known has a major effect on the gut microbiome and is thought to contribute to AS [20].

It is likely that further research will illuminate several ways in which gene–gene interactions contribute in AS pathogenesis.

2.1.8 List of genes associated with ankylosing spondylitis

A list of genes associated with Ankylosing Spondylitis can be found by searching in the National Library of Medicine gene search engine at the following link: https://www.ncbi.nlm.nih.gov/gene/?term=Ankylosing+spondylitis

2.2 Immunological factors

The complex dynamic relationship involved with inflammatory cytokines in the development of AS includes the IL17/23 axis, which is the most well studied of these pathways, but IL6, IL10, IL22 and tumour necrosis factor are also recognised as contributing to chronic inflammation. Other inflammatory cytokines have been postulated as playing a role including IL37 [21], but the facts remain that the detailed mechanisms are unknown.

The differences observed in immune cells and cytokines in AS suggest an aberrant composition of immunological factors in AS pathogenesis. In the peripheral blood of AS patients and healthy HLA-B27-positive controls, for example, the levels of T cells secreting tumour necrosis factor (TNF)- α and interferon (IFN)-γ were reportedly lower. CD8+ T cells in AS patients tended to secrete more IL10 [2].

An abnormal polarisation of macrophages induced by IL-4 was found in AS patients [20]. Reports exist showing CD163+ macrophages are the predominant cells in inflamed peripheral joints in SpA patients [22].

Dendtritic cells (DCs) play a key role in ankylosing spondylitis. AS can develop in patients who receive bone marrow from donors with AS and it appears DCs are the drivers of AS development in these cases [22, 23]. Signalling pathways of DCs are dysregulated in ankylosing spondylitis with an associated Th17 inflammatory response [22]. One study identified patients with AS demonstrate altered DC and T cell populations implying pathogenic roles for the IL-23 cytokine axis in intestinal inflammation [24].

Patients with AS have significantly higher percentages of NK cells of the subset of CD56dim CD16+ [22]. This important immunologic characteristic in AS patients might explain the relationship between the autoinflammatory and autoimmune components of AS pathophysiology.

The term seronegative implies that the disease process does not have antibodies detected in serum antibody tests that are found in autoimmune conditions such as rheumatoid arthritis, SLE, primary Sjogren's Syndrome and scleroderma spectrum disorders, however emerging research has demonstrated an association of antibodies in AS [25–28]. As discussed, whilst AS might be initiated by the innate immune system with the connecting IL-17/23 pathway being prominent in the pathophysiology, there is known B cell activation and involvement of the adaptive immune system, so the generation of antibodies makes logical sense [25, 26]. Consensus on a distinct autoantibody is lacking, however this could emerge from ongoing research in the role of B cells in AS. The molecular mimicry hypothesis as demonstrated in patients with AS who have prior klebsiella and associated circulating antibodies also supports B cell involvement in AS [29].

Synovial biopsies of AS patients have been found to contain B-cell rich follicles with some literature reporting aggregates of T-cells and B-cells arranged into structures similar in appearance to germinal centres. However, the presence of lymphocytes has been argued as potentially being secondary recruitment to an already established inflammatory processes [30]. This introduces debate in

literature on whether AS is autoinflammatory in nature rather than autoimmune, with the truth being more complex than a mere dichotomous linear continuum of possibilities.

Innate lymphoid cells are also recognised to play an important role in AS. ILC3 cells in particular are thought to be involved in the pathogenesis of inflammation. They reside in the gut and express the alpha-4/beta-7 integrin, functions as a homing receptor. In patients with AS the ligand for this particular integrin, mucosal vascular addressin cell adhesion molecule 1 (MADCAM1), is more strongly expressed in high endothelial venules (HEV) of the intestines, the blood, synovium and also in bone marrow [31]. One theory postulates that these IL-17+ and IL-22+ ILC3 cells migrate from the gut into the systemic circulation and via integrin-ligand communication are drawn towards target tissues in the bone marrow, joints, peripheral joints, and entheses [31].

The roll of T cells in ankylosing spondylitis drives the immunological response in AS. A large range of T cells are involved. Th17 cells secrete IL-17 which is one of the main inflammatory cytokines involved in AS. A list of T cells involved with AS is provided in **Table 1**.

Strong evidence suggests a central role of IL-17 secreting γδ T cells in the pathogenesis of AS [32]. These cells have been found to be present in increased numbers in the blood of AS patients, but have also been identified as resident cells within entheses [32]. It is thought that microtrauma and mechanical loading of the entheses activates resident immune cells that trigger an aberrant immune response in AS.

T reg cells are known to be dysfunctional in AS. FOXP3+ T reg cells have been described in AS [15, 33]. FOXP3+ is a protein that is affected by hormonal fluctuations, inflammatory cytokines and danger signals and it is thought that this possibly accounts for some of the gender bias in AS presentation as well as other autoimmune disorders [33].

IL-17 positive mucosal-associated invariant T (MAIT) cells are one of the subsets of innate-like cells and are elevated in AS patients, particularly in synovial fluid, but also in circulating blood [34].

Cells that are major components in the immunological response in AS patients are summarised in **Table 1** [1, 22].

2.3 Infections and microbiome

The human enteric system contains unique microbiota with a stable ecology and dynamic equilibrium [35]. This association between host and symbiotic microbes is the result of millions of years of coevolution leading to homeostatic equilibrium between enteric flora and host health and disease [35–37]. The composition of an individuals' microbiota is generally highly resistant to change, however is influenced by a range of exogenous factors. The resilience of this complex adaptive system is impacted by major perturbations, which can lead to a 'tipping point' beyond with significant change can occur and disease might result [36]. Studies in metabolomics demonstrate small-molecule metabolites generated from microbiota can influence intestinal inflammation, and potentially result in joint inflammation [38] and the development of AS [20]. Furthermore, the enteric microbiome of patients with AS has been shown to exhibit a higher load of bacterial peptides known to be presented by HLA-B27 [20]. This suggests either HLA-B27 fails to clear these, or that these peptides drive the immune response associated with HLA-B27, a potentially important

Cell Type	Function	Alteration in AS
CD14–CD16+ Dendritic cells	Interacts with HLAB27, induces the IL23/27 pathway Secretes IL-6 and IL-1β	Increased
CD 163+ Macrophages	Secretes IL23	Increased
CD56dim CD16+ NK cells	Interacts with HLAB27 via KIR3DL1/3DS1 locus	Increased
Th1 helper cells (CD4+ subset)	Secretes (IFN-γ), IL- 2, and tumour necrosis factor alpha (TNF-α)	Increased
Th2 helper cells	Secretes IL4, IL10, IL13	Increased
Thl7 cells (CD4+ subset) (5 different types IL-17A, IL-17B, IL-17C, IL-17D, IL-17E, IL-17F)	Secretes IL17 in AS also secretes IL6, IL IL-26, IFN-γ	Increased
Thl/Th2 and Thl7/Treg	Promotes IL17 pathway	Increased ratio
T22 cells (CD4+ subset)	Secretes IL-22, IL-13, and TNF-α	Increased
TCD3+ TCD8+ cells	Differentiate into Tc1 and Tcl7 generating TNF-α, IFN-γ and IL-17 and driving inflammatory pathway as well as lysing cells via perforin/ granzyme or signalling through Fas/ FasL pathway	Increased
CD19+, CD86+,CD95+,CD27-B cells	Ectopic lymphoid structures in joints	Increased
IL-17+ Mucosal-associated invariant T (MAIT) cells	Secrete IL17, IL 22 and IFN-γ	Increased
Innate lymphoid cells	ILC1 secretes IFN-γ	Increased levels of ILC3 cells in peripheral blood
	ILC2 is the main source of IL-4, IL-5, and IL-9;	
	ILC3 secretes IL-17 and IL-22 in response to IL-23	
γδT cells (T cell subset) 50% of cell types at mucosal and epithelial sites	Produce IL17	Increased levels in peripheral blood as well as in entheses

Table 1.
Some alterations to pro-inflammatory cells in AS.

finding in the role of understanding the relationship between gut dysregulation, HLA B27 and AS [20].

The 'joint-gut' axis is a term used to describe the relationship between microbiome dysregulation inflammation and musculoskeletal manifestations in AS and other autoimmune diseases [39]. The 'leaky gut' allows bacteria derived proteins and primed immune cells into systemic circulation eliciting a systemic immune response. However, several challenges exist in establishing the role specific microorganisms play in gut dysbiosis. Firstly, the concept of an organism being pathogenic as opposed to commensal flora is largely context dependent. Secondly, establishing causation as opposed to association remains problematic [36]. Thirdly, there is the challenge of

establishing whether there exists a characteristic microbiome that is specific to AS patients (**Figure 3**) [40].

Many of these microbes were thought to be gut commensals, but new studies have shown them to be increased specifically with disease and thus are regarded as pathobiont [36].

Despite this there exists a large volume of literature demonstrating distinct changes in the microbiome of patients with AS. Experimental models of AS have attempted to describe causal microbial pathways, but this is an ongoing endeavour in research. For example, decreased numbers of Firmicutes, particularly the species *Faecalibacterium prausnitzii* and also *Clostridium leptum* have been found in spondyloathrtidites and inflammatory bowel disease (IBD) and present an important link between the seronegative spondyloarthropathies (SpA) and gut inflammation [36]. *Porphyromonas gingivalis*, a periodontal bacterium, with an ability to colonise synovial joints and exacerbate collagen-induced arthritis in mouse models [41].

Various reports estimate up to 70 percent of patients with HLA-B27-associated spondylarthritis have microscopic gut lesions [20], with around one third demonstrating overt gut inflammation [42]. First degree relatives of these patients show signs of subclinical gut inflammation and impaired gut epithelial barrier [43]. Importantly, the degree of enteric inflammation correlates with disease activity and degree of sacroiliac (SI) joint inflammation [42].

Dialister is a saccharolytic bacteria, belongs to family *Vellionelaceae*. One study demonstrated inflamed intestinal tissue contained higher levels of this bacteria compared to non-inflamed tissue and correlated with increased disease scores in AS patients [44].

Ruminococcus gnavus is a known pathobiont associated both spondyloarthritis and inflammatory bowel disease [38].

A Chinese study on AS patients showed elevated numbers of *Akkermansia muciniphila* and several *Prevotella* species, including Prevotella *melaninogenica, Prevotella copri*, and *Prevotella* spp. all of which are mucin degrading bacteria along with *Bifidobacterium* whilst reduced *Bacteroides* species were noted [45].

Increased abundance of *Mucispirillum schaedleri* is thought to possibly compromise the spatial segregation by bringing luminal microbes closer to the intestinal epithelial cells which induces and inflammatory response [36].

IgA coated *E coli* have been shown to induce IL17 inflammation and correspond with increased disease activity scores in patients with spondyloarthritis and Crohn's Disease [46].

The role of fungal agents in enteric dysbiosis is being investigated. Fungal bioproducts including β-glucan are thought to trigger AS in certain mouse models [36]. IL-17 inhibitors has demonstrated a shift in bacterial and fungal composition of the enteric microbiome in patients. Anti-*Saccharomyces cerevisiae* antibodies are associated with intestinal inflammation in patients with Axial spondyloarthritis and Chron's disease according to one study [47]. In such studies biome disturbances result in overexpression of IL-17/23 cytokines and the expansion of IL-25/17 [48].

Recent studies have examined the role of inter-kingdom fungal-bacterial interactions in AS patients revealing perturbed relations including decreased fungal to bacterial biodiversity ratios in these patients [49].

The human gut virome has also gained attention, however has remained a particularly challenging area with limited studies. It is thought to modulate the bacteriome via bacteriophages [36, 50]. *Caudovirales* bacteriophages are reported to be increased in Crohn's Disease. In the healthy gut the viral core reportedly consists of virulent

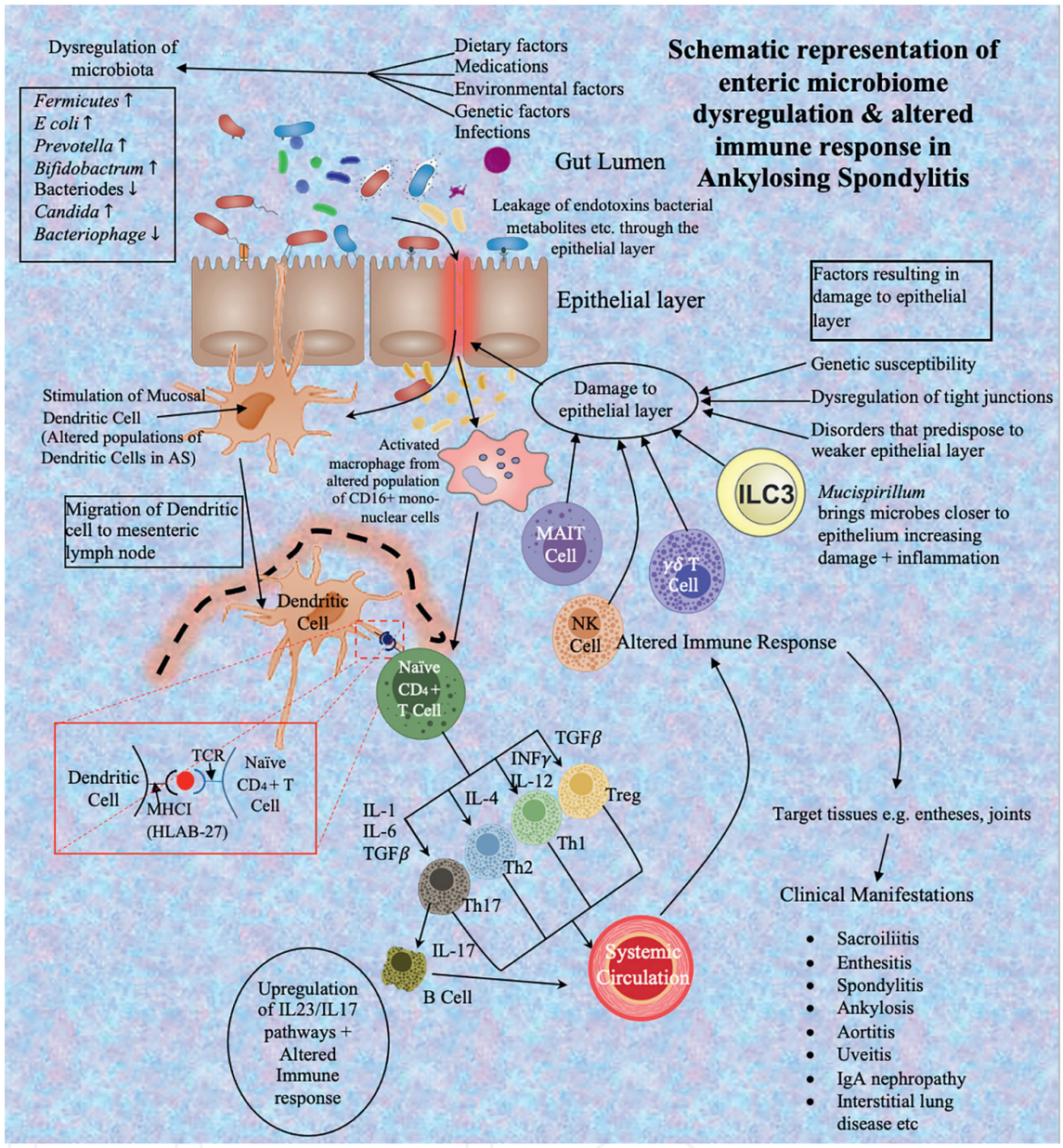

Figure 3.
Schematic representation of enteric microbiome dysregulation & altered immune response in ankylosing spondylitis.

phages. However, in patients with Crohn's Disease, the virome shifts towards a less virulent bacteriophage core that correspondingly affects the bacterial community and encourages infection [36].

The role of infection in development of spondyloarthritis and AS specifically is well documented in medical literature. Research demonstrates infections significantly increase the risks of AS [51].

Cases of reactive arthritis (ReA) following venereal infection date back to the early 1800s [52]. Approximately 12–16% of individuals who initially present with ReA go onto develop AS. Patients who are HLAB27 positive have a worse prognosis, however it is still unclear whether infection is the provoking factor in patients who develop AS in this context, or whether these patients would have developed AS anyway [53]. The pathophysiological mechanisms between ReA and development of AS have not been identified, however it is postulated that chronic infections result in altered

immunological response resulting in arthritis. Research demonstrates the presence of yersinia in the lymph nodes of those with *Yersinia* related arthritis and *Chlamydia* in the synovial fluids and tissue of those with chronic seronegative arthritis [54].

Gram negative infections in the gut are particularly associated with spondyloarthritis and reactive arthritis with chronic infections conferring a greater risk of progression to AS. (*Shigella, Salmonella, Yersinia,* are known to produce inflammatory arthritis as are *Campylobacter* species, *Clostridium difficile, Brucella,* and *Giardia* [6, 20].

The antigen-binding region of several HLAB27 genotypic subtypes, especially HLA B2705 share an amino acid sequence with nitrogenase from Klebsiella *pneumoniae*. This has led to a theory that molecular mimicry following certain infections is a possible mechanism for pathogenesis of AS in genetically susceptible hosts [29].

HLA-B27 results in a robust cytotoxic CD8+ T cell response to certain viruses, including hepatitis C, influenza and HIV, therefore it is possible that HLAB27 actually confers an evolutionary advantage in certain infectious contexts [55]. Research has demonstrated that HLA molecules can affect the severity and duration of viral infection [56]. Case reports of Sars-2-cov infection capable of triggering a reactive arthritis and uveitis in an HLA B27 positive patient exists [57, 58]. Other research suggests that Sars 2-Cov might have a predilection for people with certain HLA phenotypes [59]. The relationship between these various phenomena and the subsequent development of AS is an area which is likely to emerge in ongoing research and has important implications for both rheumatology and infectious diseases.

Prior flu infection may influence the presentation of such arthritogenic peptides especially in the presence of individuals with both ERAP1 and HLAB27 genes [3].

2.4 Metabolomics

Studies on metabolomics have gained attention in recent years. This has relevance to understanding pathogenesis, diagnosis and response to treatment in AS [60].

Studies show a range of findings in AS patients including altered pathways of tryptophan metabolism [61]. Other studies revealed significant alterations in unsaturated fatty acids (FA), linoleic acid, alpha-linolenic acid, FA degradation, and FA biosynthesis pathways [9].

One study on AS metabolomics noted alterations in several pathways including amino acid biosynthesis, glycolysis, glutaminolysis, fatty acids biosynthesis and choline metabolism developed a diagnostic panel comprising five metabolites (L-glutamate, arachidonic acid, L-phenylalanine, PC (18:1(9Z)/18:1(9Z)), 1-palmitoylglycerol). This study found that TNF inhibitors treatment could restore the equilibrium of 21 metabolites [62]. Another study revealed down regulation of the Vitamin D3 metabolite—(23S,25R)-25-hydroxyvitamin D3 26,23-peroxylactone. The ratio this vitamin D metabolite *versus* vitamin D binding protein serum levels was shown to be altered when compared with healthy controls [63].

2.5 Sex hormones

Until recently it was thought that a relationship between male gender and presence of AS existed, given males account for the majority of AS patients, however recent literature demonstrates a more homogenous sex prevalence of AS [64]. Although this is true, females with AS have different phenotypical disease due to different immunological, hormonal, and genetic responses [64].

One study examining the differences between manifestations of AS in male vs. female patients demonstrated different levels of TNF, proinflammatory cytokines IL-6, IL-17, and IL-18. These authors noted a longer diagnostic delay in female AS patients compared to males. Several studies have reported female AS patients experience a higher number of extra-articular manifestations (EAM) including inflammatory bowel disease, enthesitis, psoriasis, whereas EAMs are less common in male patients with the exception of anterior uveitis which occurs more frequently in males with AS [64]. Radiographic damage and progression of disease is worse in male patients [64].

Despite this these recognised relationships, the role of sex hormones and biologic gender in AS pathogenesis remains poorly understood [65]. This is complicated by conflicting evidence in research to date [66] with a lack of robust study design to date [67].

A small study by Odeh *et al* [66] reported spinal syndesmophyte scores do not correlate with testosterone levels. One study suggested testosterone, in particular, attenuates inflammatory processes via a number of cellular and molecular pathways [68].

However, some reports on variations of sex hormones in AS patients compared with controls imply that increased androgen levels in both males and females possibly contributes to disease development [69]. Patients who are HLA-B27 were originally reported to have higher levels of testosterone [70]. However, a study with small sample size revealed a decreased testicular testosterone reserve, elevated luteinizing hormone level, and inversion of the normal estradiol/testosterone ratio and increased estradiol level [2]. In female studies reports show that patients with active AS have significantly lower estradiol levels in the menstrual period. Case reports of increase in incidence post pregnancy also suggest that sex hormones play a role with a proposed hypothalamic–pituitary–adrenal axis impairment [2].

One study demonstrated that SKG MICE who received oestrogen therapy showed little inflammatory infiltrates of Achilles tendon, or spinal discs compared with mice who had undergone oophorectomy [71]. One report proposed ostrogens might reduce arthritis due to their suppression of wnt signalling [72]. Another study reported that selective oestrogen receptor modulator (SERM) lasofoxifene not only suppresses the effects of joint inflammation and improves bone mineral density in SKG mice, but it also affected the composition and biodiversity of the gut microbiome, with the authors concluding further reaserch on the role of SERMS to evaluate their effects in SpA is required [73].

There is literature to suggest a relationship between women who experience excess androgen levels in conditions such as polycystic ovarian syndrome (PCOS) and development of AS [74], as well as other rheumatic diseases [75]. A Korean study questioned whether other factors besides sex hormones play a role in reduced disease progression in females noting in their study there was no significant relationship between oestrogen levels and radiographic progression [73].

This research into excess androgen levels in some AS patients has led to researchers in the past asking the question whether patients who are HLA-B27 positive would benefit from anti-androgen therapy [76]. However this remains to be addressed by research with some emerging literature demonstrating progression of ankylosis in mouse models where anti-androgen therapy has been used to treat excess androgen levels [65].

There is growing recognition of the relationship between metabolic syndrome and ankylosing spondylitis [77–79], with one Morrocon study demonstrating a prevalence of 34% and a male predominance of 67% [80] Another study demonstrated treatment with anti-TNF-α monoclonal antibody infliximab reversed excess insulin levels in non-diabetic patients [77]. It is possible to postulate that there might be a

relationship between androgen excess metabolic syndrome and the development of AS, but this relationship requires more research to establish.

Of note is the incidence among men and women is similar in non-radiographic axial spondyloarthropathy, ie in individuals meeting clinical criteria for axial spondyloarthritis without radiological evidence of sacroiliitis on x rays [2].

It should be noted that most studies conducted in this area are of small sample size only, or have been performed in animal models. It is known that immune responses due to sex differences change throughout the life of an individual and are influenced by age and reproductive status and that sex hormones impact on different immune responses between the sexes [81]. Additionally, it is now recognised that environmental factors including microbiome composition and nutrional status impact immunological response differently in males and females [81]. Therefore it appears likely that additional endocrinological, immunological, genetic and environmental factors interacting with sex hormones contribute to pathogenesis of AS. Further research is required to better understand this complex relationship.

2.6 Diet and lifestyle factors

Currently evidence on the relationship between AS and diet is extremely limited and inconclusive. This is mainly due to studies being small, single studies with moderate-to-high risk of bias, and insufficient reporting of results as reported by one systematic review [82].

No prospective cohort studies of dietary risk factors for the development of AS exist, however one report suggested that a change in dietary habit from a high protein, low-starch diet to a Westernised high-starch diet among the Inuit population of Alaska and Canada whose populations also express high percentages of HLA B-27, possibly explain an increased incidence of AS in this population [83]. Other studies have suggested adoption of a "Westernised" diet is a contributing factor to development of AS [84]. One study reported that patients with AS were breastfed less compared with healthy controls [85]. Breast feeding could potentially affect the development of AS through microbiome and other immunological factors.

Mouse models demonstrated that those with a high salt diet resulted in significantly higher Th17 in their gut lamina propria [86]. Human studies revealed males who were fed a high salt diet increased the Th17 cells in their peripheral blood and demonstrated a corresponding loss of lactobacillus species [86].

Smoking is associated with increased cumulative spinal structural damage in patients with AS [87] as well as higher disease activity, inflammatory markers and functional disability [88]. Whether smoking induces AS is unclear.

Alcohol consumption is associated with spinal structural progression in patients with axial spondyloarthritis and appears to be dose related according to one Korean study [89]. These researchers demonstrated an increase of syndesmophyte progression over a two-year period.

2.7 Environmental factors

A report demonstrated significantly higher urine concentrations of cadmium, antimony, tungsten, uranium, and trimethylarsine in patients with AS compared to healthy controls [90].

Whilst there is very limited research on the role of other environmental toxins and development of AS in the literature, there is a well-established ongoing discussion on the role of a range of environmental toxins in the development of autoimmune diseases [91, 92]. Alterations to the gut biome through interference by toxic substances is one proposed mechanism [93]. Further research is required to establish whether any chemical contributions play a role in the pathogenesis of AS. This includes examining whether endocrine disrupting chemicals, pesticides and other substances might impact the immune-endocrine axis of patients with any genetic susceptibility to developing AS.

2.8 Anatomic factors

Structural integrity of anatomical in patients could possibly predispose certain individuals to the development of AS. For example, it is known that mucosal integrity plays in important role in gut dysbiosis and factors that weaken the mucosal layer predispose to enteric inflammation and possible generation of aberrant immune pathways. It is possible to assume, therefore that disorders resulting in fragile mucosa would also potentially generate an increased risk of permeability, inflammation and associated arthritis. One report revealed circulating levels of connective tissue degradation are diagnostic and prognostic markers in AS [94]. At a microscopic level, barrier integrity of the intestinal mucosa is maintained by intestinal epithelial tight junction proteins. These include occludin, claudins, and zonula occludens. Where dysfunction of these proteins occur this could affect the tight junctions. This is proposed as a contributory mechanism by which microorganisms and their products penetrate the gut wall with associated activation of mucosal inflammation [95].

Clinical studies in patients with AS suggest there is involvement with mechanical strain and inflammation of the entheses [1] with a tendency for normal inflammatory and repair pathways to go awry in the context of AS.

Patients with conditions that potentially affect the strength and quality of these tissues might be at an increased risk of developing AS through altered responses to normal loads and associated immunological response. This means it is possible that patients who have conditions of heritable disorders of connective tissue (HDCT) to have weaker tissues that predispose them to development AS and might account for comorbid case reports in the literature, but other contributory factors including HLA-B27 might also be responsible. For example, one study reported 24% of patients with hypermobile Ehlers Danlos Syndrome (H-EDS) were positive for HLA-B27 [96]. Case reports of EDS and comorbid ankylosing spondylitis exist [97–101]. Further research is warranted to quantify this interesting connection that might provide key information on the pathophysiology in certain subsets of patients of patients with AS.

Eighty percent of tendons are comprised of collagen with type collagen 1 accounting for 60–85% of the total [51]. Disorders affecting collagen could contribute to the pathogenesis of AS. However, there might be evidence against this theory. One study revealed higher levels of both COL1A1 and RUNX2 in the AS ligament tissues than in non-AS ligament tissues [10]. Further research is required to clarify this relationship.

2.9 Racial/ethnic differences

The relationship between various ethnicities and prevalence of the HLA-B27 gene as well as development of AS, is well reported in the literature. Between 10 to 16% of Norwegians, Swedes and Icelanders are positive for HLA-B27 and between 25 to 50%

of Inuit, Yupik and Indigenous Northern Americans [102]. Ankylosing spondylitis is three times less common in American blacks than in whites. It is extremely rare in African blacks of unmixed ancestry [103]. A histocompatibility antigen HLA-B27, which does not exist in African blacks of unmixed ancestry, and is present in eight percent of white and two to four percent of the American black population, is strongly associated with ankylosing spondylitis and Reiter's disease [103]. HLA-B27 is present in more than 80 percent of white patients with ankylosing spondylitis or Reiter's disease but in less than 60 percent of American black patients [103]. HLA-B27 occurred in 62.5% of African Americans, 85.3% of White Americans, and 86.7% of Americans with Latino ethnicity ($p < 0.0001$). Higher disease activity scores have been associated with an American black ethnicity. African Americans with AS have more severe disease compared to either White Americans, or Latinos [104]. This could be partially influenced by social determinants of health.

3. Pathophysiology

Key aspects in ankylosing (AS) pathophysiology are well understood, however the overarching network of immunological dysregulation is exceptionally complex and remains elusive despite extensive research.

Broadly speaking the pathophysiological processes within AS can be considered as musculoskeletal (articular, osteo and entheses-related) and extra articular (ocular, enteric, renal, pulmonary, vascular and dermatological).

Within the musculoskeletal aspects of AS, three major features of the disease exist. Inflammation of the joint and entheses, significant bone demineralisation and ossification of characteristic joints and entheses.

The extra-articular pathophysiological processes involved in AS are poorly understood and are not the major focus of this chapter.

The chronic inflammation in AS results in fibrosis and ossification and ultimately a fused spine with the characteristic bamboo spine seen on xray imaging. Inflammatory responses involved include CD4+ and CD8+ T lymphocytes and macrophages, cytokines, particularly tumour necrosis factor-α (TNF-α) and transforming growth factor-β (TGF-β). The IL-23/IL-17 axis is recognised as the principle driving immunological force responsible for perpetuation of the chronic inflammation with Th17 cells and the production of the proinflammatory cytokine IL-17 playing a central role in the process [105].

There has been debate in the literature on whether AS is an autoinflammatory condition, or an autoimmune disease [16]. Autoinflammatory diseases are primarily driven by the innate immune system and, whilst autoimmune diseases are driven by the adaptive immune system and primarily present as a B cell response. Literature examining the role of the innate immune system communicating with the adaptive immune system has led some researchers to recommend that autoinflammation and autoimmunity should not be considered as opposing mechanisms, but rather as extreme opposites of a continuum, with different combinations and permutations resulting in specific clinical phenotypes [15, 16]. Whilst conceptually it is possible to separate aspects of immunological responses for the purposes of discussion and research, *in vivo* particularly with regards to AS, the situation is a complex symphony of enmeshed networks between adaptive and innate pathways that remain to be fully understood. These processes should be considered interlinked non-linear matrices rather than 2 systems on a dichotomous spectrum.

Genetic studies support the role of auto inflammation in AS pathophysiology. Research on Turkish and Iranian individuals reported a significant association between variants of MEFV and the risk of developing AS. MEFV is a gene that encodes for pyrin which is a recognised promoter of autoinflammation. Comorbidities of spondyloarthritides are reported in association with familial Mediterranean Fever [16], whilst there is discussion in the literature on the relationship between Behcet's Disease and AS although there are also reports of a relation. A theory has emerged on the concept of an overarching 'MCH-1-opathy' that accounts for the possible shared pathophysiology between the seronegative spondyloarthropathies and Behcet's Disease [23].

In the 1980s there was literature published discussing an apparent increase in responsiveness to complement activators in diseases associated with HLA-B27 [106]. There is now an emerging body of literature examining the role of compliment in AS [107–109]. The complement system is a cascade of protein cleaving resulting in a powerful pro inflammatory response. Complement is capable of destroying invading pathogens, however when there is dysregulation of complement including uncontrolled activation of the cascade combined with insufficient regulation, the destructive capabilities against invading pathogens turn against host cells. There are several studies in the literature demonstrating elevated complement products C3, C4 and C3d IgA, IgG, C-reactive protein (CRP), serum amyloid A, apolipoprotein A in AS patients [110, 111]. Circulating immune complexes have been reportedly higher in AS [112]. Additionally, reports that tumour necrosis factor inhibitors appear to reduce complement levels and reduction in arthrogenesis [108] suggest the complement cascade might be a contributing factor in the pathophysiology of AS. It is possible through this mechanism extra articular manifestations of AS including amyloidosis and IgA vasculitis might be triggered although there are reports that the manitose binding lectin levels are decreased in patients with AS which goes against this theory [113]. Infection with Klebsiella is recognised as being a significant complement cascade activator via cross-reactive antibodies against autoantigens in the infective process [107].

3.1 The IL17/23 pathway and HLA B27

There are several theories on the involvement of HLA-B27s involvement in the IL17/23 pathway. Despite this, the exact mechanisms and relationship between inflammation and resulting clinical signs of ankylosis are not fully elucidated and there is acknowledgment of knowledge gaps that remain problematic in much published research.

HLA-B27's functional role is in peptide presentation to cytotoxic T cells and NK cells. Within the endoplasmic reticulum (ER) of the antigen presenting cell, HLA-B27 binds short peptides which are then trafficked to the cell surface via the golgi apparatus and displayed ready for presentation to other immunological cells [54]. In the case of AS it is thought that HLA-B27s role in triggering the immune system might occur through different pathways. Several theories exist around how HLA-B27 results in an aberrant immunological response in the context of AS.

Firstly, protein misfolding in the ER accumulate generating ER stress that generates upregulation of IL-17 and other proinflammatory cytokines. HLA-B27 must fold and bind with protein β2m in the ER. This process is slow in HLA-B27 especially in the case of misfolding resulting in increased oxidation and formation of disulphide linked homodimers [114]. ER stress results from an inability of the ER to remove misfolded proteins resulting in what is known as the "unfolded protein response" (UPR) essentially a gain of function response. Degradation of abnormal proteins is regulated by ERAD pathway. This can result in a loss of function [7].

In extreme instances the accumulation of aberrant HLA-B27 results in apoptosis. It is theorised that the UPR results in upregulation of IL-12/via the transcription factor CHOP 23 [115]. In rat models UPR produces an up regulation of TNF alpha and ILS which results in an overall osteoclastogenesis response [115] and accounts for bone demineralisation seen in AS.

The ER stress response is also thought to be the reason behind the association of ERAP1 gene in AS pathogenesis both in HLA-B27 positive and negative patients. Whilst ERAP1 is primarily found in the ER it is also secreted by macrophages and activated by INF-gamma and lipopolysaccharides. ERAP1 is responsible for protein trimming in the ER and altered genes can contribute to the production of aberrant proteins in the HLA-B27 production pathway [115].

Although there is conflicting evidence in current research, some literature proposes quantitative changes in the peptide composition in AS and that an 'arthritogenic peptide repertoire' could contribute to AS pathogenesis [54]. NK cell receptors can recognise MHC class I molecules in addition to TCR recognition. This includes HLA-B27, and the reciprocal receptor for HLA-B27 heterotrimers KIR3DL1, has been shown to be sensitive to certain properties of peptides bound to HLA-B27 [116, 117]. Therefore, it is possible that alterations in these peptides could affect immune responses via KIR signalling [54].

Changes in this peptide repertoire could potentially affect misfolding and free heavy chain, or homodimer expression and contribute to this aspect of pathogenesis in AS [54]. A Trimolecular complex comprised of a B27 heavy chain, α2 microglobulin and a third peptide of either HLA-B27, a free heavy chain, or homodimers of HLA-B27, can possibly be recognized as neoantigens by the T cell receptor on CD4+ T cells leading to an autoimmune response [118–120].

The third theory around aberrant HLA-B27 is that misfolded HLA-B27 heavy chains are expressed on the cell surface as homodimers are responsible for an increased Th17 response via stimulation of the killer immunoglobulin receptor on natural killer (NK cells) and T cells which then drive an IL17 inflammatory response [54].

An additional theory with evidence is that dendritic cells exhibiting aberrant HLA-B27 result in loss of immune tolerance and development of autoimmunity [54].

Finally, it is postulated that certain microorganisms and cross-reactive epitopes might alter HLA27, but also that HLA-B27 itself exerts an effect on the composition of gut flora (**Figure 4**) [121].

IL-23 and IL-12 are cytokines that act as a bridge between the innate and adaptive branches of the immune system. Their critical and central role in AS is possible evidence that AS is both an autoinflammatory and autoimmune disease. The intestines are the major site of production of IL-23 [39]. HLA-B27 is thought to play a major role in IL23 and IL-12 production via the theories discussed in the previous section and the dendric cell is the primary cell that secretes these in the context of AS.

Studies on the role of IL-23 in ankylosing spondylitis demonstrates the IL-23/IL-17 axis is a non-linear matrix of complex pathways displaying overlapping yet distinct pathobiology [122]. Despite this, some research reveals that blocking the IL-17 pathway does not prevent the progression of ankylosing spondylitis [122].

IL-12 induces INF-γ which drives inflammation in AS. δγT cells and innate lymphoid cells exhibit the IL-23 receptor demonstrating their first line response to IL-23 [39]. However, Th0 cells and CD4+ cells do not exhibit an IL-23 receptor and require prior stimulation with a range of proinflammatory cytokines to become responsive to IL-23. These include IL1β and a range of other proinflammatory cytokines. Once activated via IL-23, αβT cells, δγT cells and innate lymphoid cells secrete

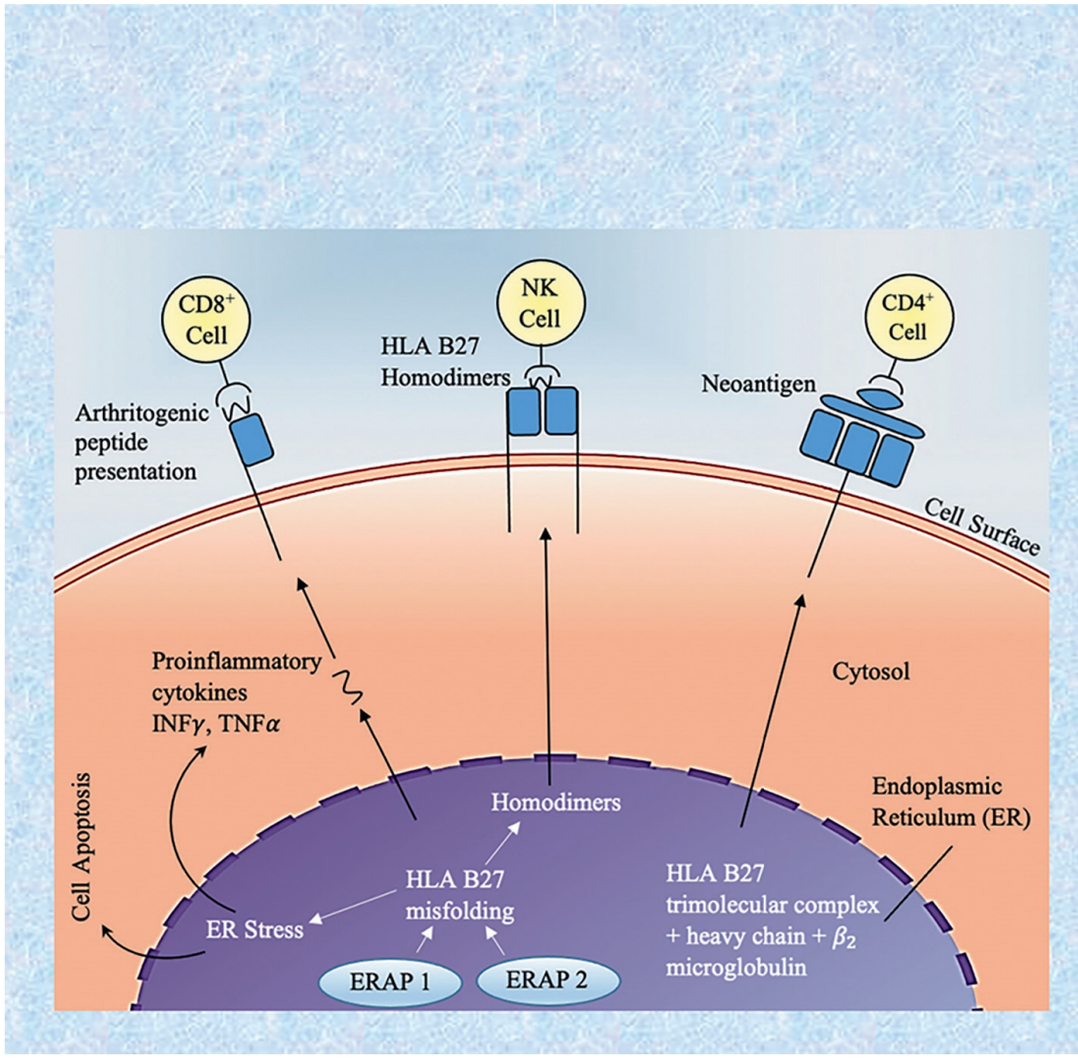

Figure 4.
Proposed theories on the role of HLA B27 & ERAP genes in pathogenesis of ankylosing spondylitis.

a range of proinflammatory cytokines including IL-17, IL-22, TNF-α and moreover αβT cells become unresponsive to the suppressive activity of T reg cells [39]. δ γT cells and innate lymphoid cells play a major role in the pathophysiology of AS [32]. Dysfunctional regulatory T cell behaviour has been identified in AS and has been associated with a loss of Tim-3, a member of the novel Tim (T cell immunoglobulin and mucin domain) family [123, 124].

T cells without IL-23 receptors exhibit IL-12Rβ1 receptors that are responsive to IL-12. Intracellular pathways involving Janus Kinase 2 (JAK2) and Tyrosine Kinase 2 (TYK2) produce STAT 4 and STAT 3 phosphorylation respectively resulting in a product ophan receptor gamma tau (RORγτ) which is essential for development of Th17 cells that ultimately drive the IL-17 aspect of this pathway (**Figure 5**) [125].

IL-17 is produced via a variety of mechanisms in AS. The IL-23 pathway discussed above leads to activation of T CD8+ cells, NK T cells and δγT cells all of which secrete IL-17, however in AS the major secreters of IL-17 come from CD4+ Th17 cells [39]. IL-17 plays a major role in the recruitment of neutrophils via IL-6 [1]. IL-17 also activates osteoclasts and directly stimulates B cells which are thought to form germinal centres in AS [26].

Pro-inflammatory cytokines critical drivers of the chronic inflammation in AS. IL-17F has recently been proposed to contribute to the pathobiology of both

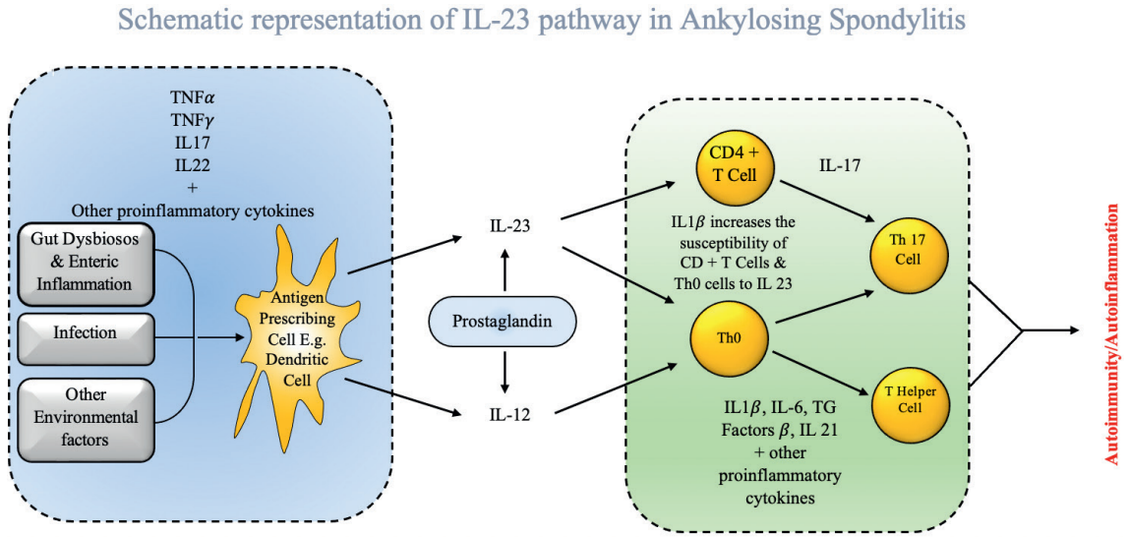

Figure 5.
Schematic representation of IL 23 pathway in ankylosing spondylitis.

inflammation and new bone formation in spondyloarthritis. Other inflammatory cytokines are also thought to contribute to chronic inflammation including granulocyte colony-stimulating factor (G-CSF) raising the possibility of research into targeting upstream activators of Th17 cells rather than IL-17 itself (**Figure 6**) [122].

3.2 Bone Remodelling in AS

Molecular mechanisms underlying the extra-articular bone formation in patients with AS remain poorly understood. Mechanical factors including repetition and overload are regarded as contributing factors.

Dickkopf-related protein 1 (Dkk-1) and sclerostin levels, with a corresponding increase in the wnt pathway have been reported in AS [87]. TGFβ signalling pathway, Hedgehog signalling pathway, hypoxic cell signalling pathway are all reported to contribute to ossification [51]. Studies indicate the Wnt pathway appears to be a major factor in the dual relationship between new bone formation and bone loss in found in AS. Observed decreased serum levels of both Dkk-1 and sclerostin, suggest a link between excessive Wnt exposure and the new focal bone formation. There has been a negative association between Dkk-1, spinal BMD, and vertebral fractures reported in one study [126]. One report noted conflicting evidence in the literature on the role of Dkk-1 in AS offering the opinion this could partly be explained by variations in PTH and vitamin D and the fact vitamin D metabolism is impaired in inflammatory disease [72].

Evidence exists supporting theories on activation of bone morphogenetic protein signalling and a decrease in bone remodelling in AS [127].

The link between inflammation of the entheses and ossification, particularly within the annulus fibrosis with characteristic syndesmophyte formation in ankylosing spondylitis remains contentious. It is a non-linear and complex process. Some research suggests new bone formation may progress independently of the inflammatory process, and may even be accelerated by the resolution of inflammation [128].

Pathological bone remodelling in AS thought to be fundamentally different from physiological bone turnover [127].

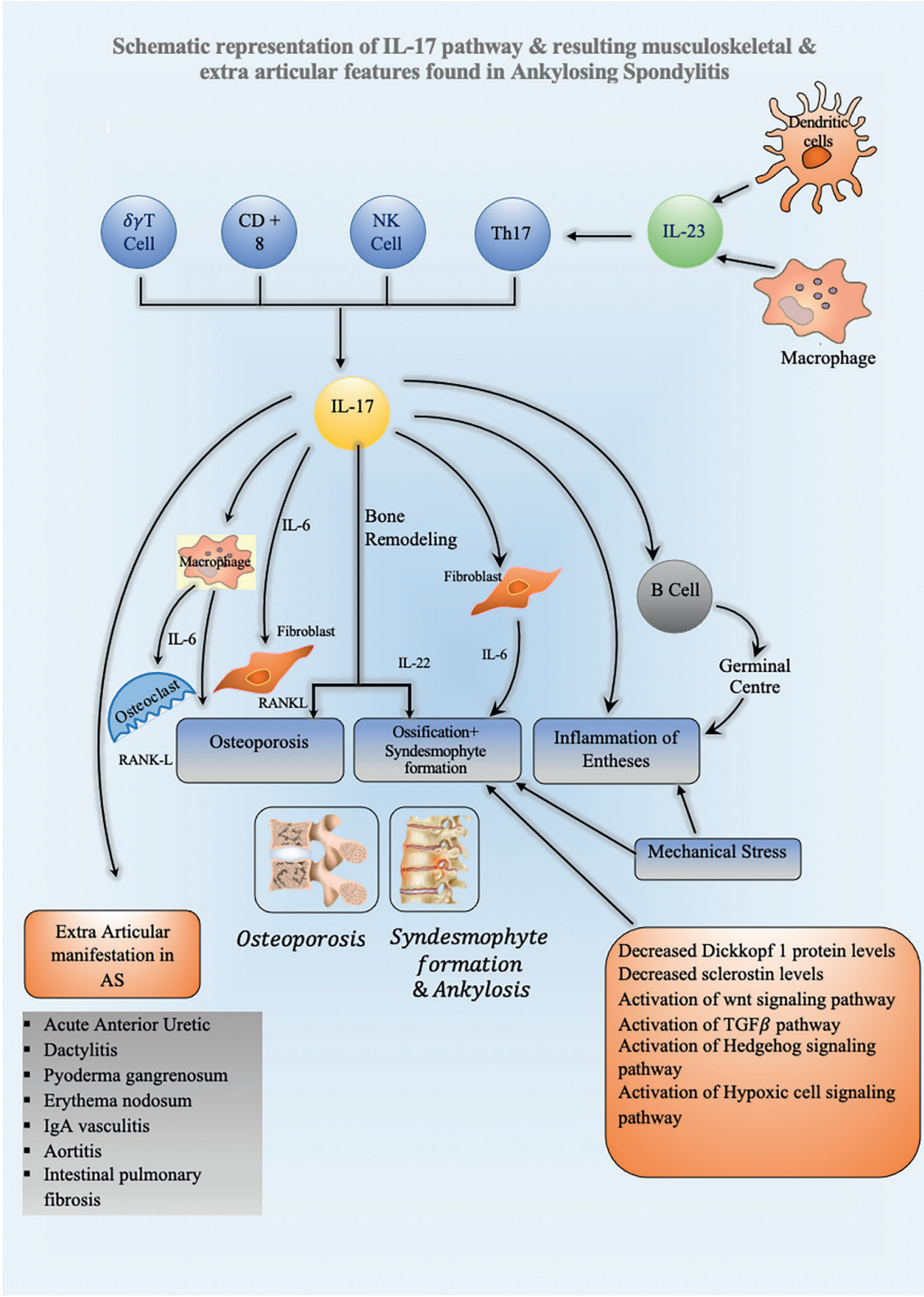

Figure 6.
Schematic representation of IL-17 pathway & resulting musculoskeletal & extra articular features found in ankylosing spondylitis.

This aberrant osteogenesis/proliferation leading to ankylosis occurs adjacent to and in connection to existing bone but extends beyond normal physiological boundaries, often along adjacent entheses [127].

There is MRI data in some patients demonstrating ossification where no previous inflammation has occurred in support of the theory that ossification occurs independently to chronic inflammation in AS [127].

MRI studies suggest active inflammatory lesions might evolve into 'remodelling' lesions characterised by mesenchymal tissue responses, ultimately leading to ossification [127]. Emerging literature suggests a range of processes resulting in heterotopic ossification of the tendons and ligaments (HOTL) [51]. HOTL is described as a dynamic process [51], similar to fracture repair, involving stages and a variety of signally pathways that start with inflammation, injury and trauma, followed by mesenchymal stromal cell recruitment, chondrocyte differentiation and finally ossification. More research is required to establish the role of HOTL in AS.

The rates and severity of bone formation vary significantly among AS patients [128].

Some studies have demonstrated patients with high markers of tissue turnover biomarkers experience more structural damage [127].

There are several pro inflammatory mediators involved in bone remodelling including tumour necrosis factor-α (TNF-α), insulin-like growth factor II (IGF-II), insulin-like growth factor-binding protein 5 (IGFBP5), prostaglandin reductase 1 (Ptgr1), latent-transforming growth factor beta-binding protein 3 (LTBP3), transforming growth factor beta-1 (TGF-β1), neutrophil elastase (NE), serum amyloid A-4 protein (SAA4), protein S100-A9 and prostaglandin-H2 D-isomerase [51].

Some researchers suggest cellular pyrophosphate exportation contributes to the pathological ossification during AS progression, which is regulated by pyrophosphate transfer-related genes, such as ANKH [128].

IL-17 impacts bone metabolism by activating the production of matrix metalloproteinases from macrophages [1]. IL-17 stimulates TNF-α production and can mediate osteoclast activation via shifting receptor activator of nuclear factor κB ligand (RANKL)/osteoprotegerin (OPG) balance towards RANKL. As OPG functions as a soluble receptor for RANKL, serving to neutralise RANKL and osteoclast formation, it is not surprising that low serum levels of OPG have been associated with osteopenia in AS [127].

IL-17A blockade may be more effective than TNF-α inhibition in halting pathological new bone formation [122, 129]. In fact, TNF-α inhibitors have been shown to accelerate ossification in some AS patients [122].

3.3 Extra-articular/musculoskeletal disease in AS

As with other areas in AS pathophysiology the exact mechanisms that are associated with the various systemic and organ specific manifestations of the disease remain unclear although HLA-B27 has been shown to be associated with acute anterior uveitis (AAU) [130]. However, one study reported that HLA-B27 negativity was associated with an increase in peripheral arthritis, dactylitis, and extra-articular manifestations including AAU [131].

One study demonstrated a relationship between HLA-B27 positivity in AS, aberrant IL-17 production can cause aortitis, acute anterior uveitis, and interstitial lung fibrosis [118]. Pathogen-associated molecular pattern (PAMP), damage-associated molecular pattern (DAMPS), natural cytotoxicity triggering receptor 2 (NPC2) and CD336 have been identified in extra-articular features of AS although more research is required to better qualifythe pathophysiology of these phenomena [118].

4. Conclusion

The pathogenesis and pathophysiology of Ankylosing Spondylitis (AS) is complex involving a variety of factors and interactions that remains to be fully understood. Genetic factors including a variety of HLA-B27 subset genes, ERAP 1, ERAP2, IL-23R and other genes are well documented in the literature, with novel genes and gene–gene interaction continuously identified in AS research. Dysregulation of the enteric microbiome with characteristic changes to certain bacterial, viral, fungal and interactions between microbiota are recognised in AS patients. There exists a corresponding aberrant immunological response with altered behaviour in DCs, macrophages, T cells, B cells that appear to be drive and be driven by the IL-17/IL-23 pathway. As such there is evidence that the pathophysiological response in AS is a combination of both autoinflammatory and autoimmune components, but further research is required to establish the intricate mechanisms involved. There remains controversy in the literature about mechanisms giving rise to the characteristic clinicical features of AS including sacroiliitis, spondylitis, ankylosis and whether these occur as a direct result of inflammation, or whether these arise independently as a result of altered mechanisms to bone metabolism in AS specifically.

The complete pathophyisiological pathways resulting in uveitis and other extra articular manifestations remain occult. Further research to establish complete understanding of factors involved in pathogenesis and pathophysiology of AS is required.

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Conflict of interest

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