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Structure-Modifying Capacity of Anti-Tumour Necrosis Factor-α Therapy in Ankylosing Spondylitis

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Abstract

Spondylarthropathies (SpA) present mainly with spondylitis, pauciarticular peripheral arthritis and enthesopathy. Ankylosing spondylitis (AS) is the prototype disease in this concept. Other entities include reactive arthritis, arthritis in patients with inflammatory bowel disease, some forms of psoriatic arthritis and undifferentiated SpA.

NSAIDs are the classical cornerstone of medical therapy in patients with SpA. The effect of these drugs on disease progression, more specifically the ankylosis, is uncertain. Sulfasalazine can be combined with NSAIDs, particularly if peri-

pheral arthritis symptoms persist. However, this combination therapy is not effective for the spondylitis symptoms. Indeed, AS is one of the rheumatic diseases for which no real disease-modifying antirheumatic treatment is available.

Challenges in chronic autoimmune arthritis have changed dramatically, especially since biotechnological compounds became available. These compounds allow for a specific intervention in the immune cascade underlying the disease. Tumour necrosis factor (TNF)-α antagonists (monoclonal antibodies such as infliximab, or soluble receptors such as etanercept) are the first representative drugs in this category. Open-label studies have shown the efficacy of these new targeted drugs, which has been confirmed by controlled studies, at least in the short term. Improvements in several clinical parameters, function, quality of life, biological parameters, histopathological synovial characteristics and magnetic resonance imaging, have all been observed. As a result of these favourable results, anti-TNFα therapy has been approved for the treatment of AS and should be considered for patients with severe axial symptoms and elevated serological markers of inflammatory activity who have responded inadequately to conventional nonsteroidal therapy.

There is evidence that this new therapeutic approach has a disease- and even structure-modifying effect in SpA. In this context, structure modification should not only be seen as inhibition of bone and cartilage destruction but more broadly as modulation of tissue histology.

Some questions remain unanswered, such as the long-term efficacy and safety of anti-TNFα therapy, the extent of structural benefit and the cost effectiveness. However, despite these concerns, anti-TNFα therapy represents a major therapeutic advancement in the treatment of AS.

1. Spondylarthropathies (SpA) and Ankylosing Spondylitis (AS)

1.1 Epidemiological Aspects

Spondylarthropathies (SpA) are prevalent and disabling diseases, presenting mainly with spondylitis, pauciarticular peripheral arthritis and enthesopathy.[1] Ankylosing spondylitis (AS) is the prototype disease in this concept. Other entities include reactive arthritis, arthritis in patients with inflammatory bowel disease (IBD) and some forms of psoriatic arthritis. Forms of the disease that cannot be classified into one of the preceding entities are referred to as undifferentiated SpA.

SpA occur in 0.2–1% of the population, with a male : female ratio of 3 : 1.[1] Age of onset ranges from adolescence to 35 years, or to an older age in more exceptional cases. There is a strong genetic link with human lymphocyte antigen (HLA)-B27, especially for AS. The disease prevalence also parallels the frequency of this allele in different populations. AS develops in 2–6% of unrelated HLA-B27-positive individuals. The risk for the disease increases to 20% in cases of HLA-B27-positive relatives of a diseased person, whereas almost no risk exists for HLA-B27-negative relatives.[1]

1.2 Rheumatic Manifestations

Inflammatory low back pain and stiffness, with insidious onset, are typically the first symptoms of AS. Alternating buttock pain reflects the sacroiliitis. Spondylitis may affect the whole axial skeleton, with radiography revealing sacroiliitis, syndesmophytes and alterations at the zygapophysial joints, eventually leading to bamboo spine.

The arthritis mainly affects the lower limbs with an asymmetric pauciarticular pattern. In contrast to

rheumatoid arthritis, arthritis of the hands may involve the distal interphalangeal joints. Dactylitis occurs in some types of SpA, in particular if associated with Crohn's disease and psoriasis. Enthesopathy is a frequent manifestation involving the insertion of the Achilles tendon or the fascia plantaris.

Objective clinical findings in the case of spondylitis include flattening of the normal lordotic curvature and restriction of movement in all planes. Reduction of the Schober index is characteristic, and the patient is unable to touch fingers to the floor by a considerable distance. The Schober test is performed with the patient standing erect. A mark is placed on the patient's back on the imaginary line joining the posterior superior iliac spines, and a second mark is placed 10cm higher. The patient is asked to maximally bend forward. With the spine in fullest flexion, the distance between the two marks is measured again. This new distance should normally be >14cm, but is typically reduced in the case of AS. Upon involvement of the thoracic spine, chest expansion becomes restricted because of costovertebral joint fusion. Additionally, the normal kyphosis of the dorsal spine becomes accentuated. Pain and stiffness in the cervical joints are followed by a decreased ability to extend the neck fully, manifested by an increased occiput-to-wall distance. Depending on the severity of both cervical and thoracic kyphosis, the patient may evolve to standing with knees voluntarily flexed.

1.3 Radiographic Abnormalities

Sacroiliitis is usually bilateral and is the most frequent and earliest radiological manifestation of spondylitis in patients with AS. Initially, this presents as a pseudowidening of the joint with sclerosis of the joint margins, usually in the lower third. With more advanced disease, erosions appear, and finally, bony fusion across the joint occurs. Especially early changes are difficult to interpret. CT and magnetic resonance imaging (MRI) are more sensitive than conventional radiography for detecting early sacroiliitis.[2] Other pelvic abnormalities include osteitis pubis and bony erosion along the margins of ischial tuberosities.

The most characteristic radiographic changes in the lumbar, dorsal and cervical spine include squaring of the vertebral bodies due to erosions of their normally concave anterior superior and inferior surfaces. Additionally, ossification of spinal ligaments that bridge the intervertebral discs results in characteristic syndesmophytes, in some cases eventually leading to bamboo spine. Also, zygapophyseal joints may become obliterated by bony fusion. Sterile spondylodiscitis must be discriminated from an infectious complication.

Sacroiliitis and syndesmophytes are typically symmetrical in AS, but may appear asymmetrical in psoriatic or reactive arthritis. Generalised spinal osteopenia is common in AS and spinal fractions may occur after minor trauma.

Erosions in peripheral arthritis tend to be marginal, and may be accompanied by bony regeneration. In contrast to the natural evolution in rheumatoid arthritis, in SpA juxta-articular mineralisation may slowly return in affected joints that have gone into remission. Bony proliferation may also show up in the periosteum (including shafts of metacarpals, metatarsals, phalanges, distal femur) and at entheses.[3]

Psoriatic arthritis bears some characteristic radiological findings. Marginal erosions often display clear proliferative new bone formation, possibly with joint fusion. Acro-osteolysis (tufting), cupping of the proximal portion of the phalanges (pencil-in-cup) and periostitis are common in severe disease. Arthritis mutilans with osteolysis of hand or foot bones is a rare but severe and deforming type of psoriatic arthritis.

A recent study demonstrated that, unlike the general perception, a large proportion of patients with AS show progressive structural damage, evidenced by radiographic progression.[4] This study indicated that the progression of structural damage in AS and SpA should be considered of equal importance as rheumatoid arthritis, the alternative prototype of chronic inflammatory joint disease.

1.4 Extra-Articular Manifestations

Besides the joint-related manifestations of SpA, extra-articular manifestations mainly involving the gut, the eyes and the skin may occur.

1.4.1 Gut

Subclinical gut inflammation revealed by ileocolonoscopy in patients with different forms of SpA has been confirmed in multiple studies. In undifferentiated SpA, the prevalence of macroscopical lesions varies from 24% to 38%, while microscopic lesions were found in 24–72%.[5] A similar prevalence of gut lesions was described in AS patients.[5] In the latter population, the prevalence of gut inflammation was higher in patients with associated peripheral arthritis. Gut inflammation was also described in other diseases included in the concept of SpA: enterogenic and urogenital reactive arthritis, pauciarticular late-onset juvenile arthritis and in patients with psoriatic arthritis presenting with axial and/or pauciarticular disease.[5]

Upon follow-up of patients with SpA in whom a second ileocolonoscopy was performed, remission of joint inflammation was associated with a disappearance of the gut inflammation, whereas persistence of locomotor inflammation in most of the patients was associated with the persistence of gut inflammation, confirming the strong relationship between gut and joint inflammation.[6,7] Most of the patients with normal histology or acute intestinal lesions exhibited transient arthritis, whereas the majority of those with chronic intestinal lesions had persistent inflammatory joint symptoms. In total, 6.5% of the SpA patients who did not present with any clinical sign of gut abnormality developed IBD during the disease course.[8] All of these patients initially presented subclinical inflammatory gut lesions, and all but one had the features of chronic inflammation.

1.4.2 Eye

Uveitis is classified mainly according to its anatomical location, onset and duration. Anterior uveitis or iridocyclitis involves the iris or ciliary body; posterior uveitis or retinochoroiditis involves the choroid or retina; and intermediate uveitis involves the pars plana, peripheral retina and vitreum. The onset may be acute or insidious. Uveitis may be self-limited, recurrent or chronic. Eye inflammation, especially uveitis, is a prominent feature of SpA, with a likelihood in AS and reactive arthritis of 1 : 3 and 1 : 4, respectively.[9,10] This prevalence is lower in psoriatic arthritis and SpA associated with IBD.[10] Uveitis associated with AS and reactive arthritis is usually a unilateral acute anterior uveitis with a high tendency to recur sometimes in the contralateral eye. Uveitis associated with other types of SpA may be less characteristic in its presentation, with a higher tendency towards posterior pole involvement, bilaterality and chronicity. Acute anterior uveitis is grouped into the spectrum of HLA-B27-related disease; other genetic and environmental factors including infections by Gram-negative bacteria are supposed to play a role in its pathogenesis. The prognosis of uveitis is usually good with topical treatment, and only those patients with posterior pole involvement or a high tendency towards recurrence or chronicity might benefit from immunosuppressive therapy.

1.4.3 Skin

In some types of SpA, cutaneous manifestations related to the disease may occur. Skin psoriasis is related with SpA and a major subgroup of patients with psoriatic arthritis fulfil classification criteria for SpA. However, the exact relation between psoriatic arthropathy (PsA) and SpA remains controversial and therapeutic trials with anti-tumour necrosis factor (TNF)-α compounds in PsA have been performed in separate studies.

Specific tegumental manifestations also occur in reactive arthritis. In most cases it follows urogenital or enteric bacterial infection. It is generally considered a sterile arthritis that appears to involve immune response to bacterial organisms and genetic host susceptibility associated with the presence of HLA-B27 antigen.[11] Reactive arthritis manifests clinically as a rheumatoid factor-negative oligoarthritis associated with enthesopathy and certain mucosal and skin lesions. Balanitis circinata is a painless, erythematous lesion of variable size located at the glans penis. The lesions appear as erythema

progressing to pinpoint erosive papules and pustules with subsequent superficial ulceration around the external meatus and corona glandis. Scale crust, which may appear as psoriasiform plaques, may be prominent in the circumcised patient. Painless, erythematous, well defined lesions may also be found on the oral mucosa, including hard and soft palate, gingiva, tongue and cheeks. Typical skin lesions are referred to as keratodermia blennorrhagica: these are lesions of pustular psoriasis on the palms of the hand and/or the soles of the feet. The initial lesion is an erythematous macule evolving into a small papule that rapidly develops a vesicopustular, keratotic appearance. Individual lesions may coalesce, producing large areas of thick keratotic scale crust. These evolve into thick yellow-to-reddish-brown lesions, typically on the soles of the feet, usually developing several weeks after onset of the disease. Scaling plaques may develop on the scalp as well as over the knees and elbows, mimicking psoriasis. Subungual hyperkeratosis is a form of keratoderma that may also closely resemble psoriasis. The nails may be thickened with ridges, but pitting is usually absent. The skin lesions mostly resolve spontaneously within several weeks.

The SAPHO (synovitis, acne, pustulosis, hyperostosis, osteitis) syndrome puts together osteoarticular lesions described separately under numerous denominations, such as multifocal osteomyelitis, pustulotic arthrosteitis and acne rheumatism.[12] The locomotor involvement includes hyperostosis, aseptic osteomyelitis or arthritis localised at sites such as the anterior chest wall, sacroiliac joints or long bones. Skin disease includes acne conglobata or acne fulminans, palmoplantar pustulosis and hidradenitis suppurativa. The association of sterile inflammatory bone lesions and neutrophilic skin eruptions is the cornerstone of this syndrome, which has links with the SpA and with psoriasis.

1.5 Pathogenetic Concepts

The pathogenesis of SpA is linked with a genetic predisposition (essentially HLA-B27) and environmental factors (role of bacterial pathogens). Indeed, a major pathophysiological clue is provided by reactive arthritis, a subtype of SpA that is known to be triggered by infections of the gut or the urogenital tract with bacterial strains such as *Yersinia enterocolitica*, *Salmonella typhimurium* and *S. enteritides*, *Shigella flexneri*, *Campylobacter jejuni* and *Chlamydia trachomatis*. The importance of intestinal pathogens is further illustrated by the HLA-B27 transgenic rat model, in which arthritis and colitis did not develop in germ-free conditions.[13,14] In humans, there is now compelling evidence that gut inflammation and increased gut permeability play a role not only in reactive arthritis but also in other types of SpA. Correlating the bacterial hypothesis with the major histocompatibility complex class I linkage, it has been proposed that HLA-B27 is involved in the activation of cytotoxic T lymphocytes by presenting either specific bacterial peptides or arthritogenic self-peptides cross-reacting with bacterial antigens.[15] Alternatively, HLA-B27 itself could share peptide sequence homologies with bacterial antigens[16] or it could have effects that are independent of its antigen-presenting function: impairment of bacterial elimination leading to defective host defence and persistence of bacterial antigens,[17] alteration of intracellular signalling and secretion of proinflammatory cytokines,[18] misfolding during the intracellular assembly process[19,20] inducing both the activation of nuclear factor-κB[21,22] and the formation of HLA-B27 heavy chain homodimers.[23,24]

1.6 Therapy

In association with intensive physical training, NSAIDs are the classical cornerstone of medical therapy in patients with SpA. The effect of NSAIDs with potent anti-inflammatory activity, such as piroxicam, phenylbutazone or indometacin, is rapid, but temporary, with relapse of pain upon discontinuation of this treatment. The effect of these drugs on disease progression, more specifically the ankylosis, is uncertain.

A recent study looked at the effect of continuous versus discontinuous (on demand) celecoxib treatment on radiographic progression (lumbar and cervical spine) in patients with AS.[25] The progression

in the continuos group was significantly decreased versus the group with discontinuous treatment, providing evidence that continuous treatment with NSAIDs is capable of slowing down the radiographic progression of the disease.

Sulfasalazine can be combined with NSAIDs, especially if peripheral arthritis symptoms persist. However, this combination therapy is not effective for the spondylitis symptoms.[26] Indeed, AS is one of the rheumatic diseases for which no real disease-modifying antirheumatic treatment is available.

Challenges in chronic autoimmune arthritis have changed dramatically, especially since biotechnological compounds have become available. These compounds allow for a specific intervention in the immune cascade underlying the disease. TNFα antagonists (monoclonal antibodies or soluble receptors) are the first representative drugs in this category, and have shown already significant clinical efficacy in rheumatoid arthritis.[27-30] Not only have these compounds got a superior effect on the signs and symptoms of the arthritis, compared with classical disease-modifying antirheumatic drugs, they also have a structure-modifying action. This is best illustrated for infliximab in association with methotrexate in patients with rheumatoid arthritis.[28] This proof of concept of therapeutic efficacy with biotechnologically engineered compounds in arthritis has given an enormous impetus to the research in this field. Not only are the existing compounds tested in an increasing number of indications but other agents that selectively interfere with different molecules in the inflammatory cascade are also being developed. Together, this puts the rheumatologist in the forefront of a new era of molecular medicine.

2. The Rationale for Using Anti-Tumour Necrosis Factor (TNF)-α Therapy in SpA, Including AS

For use in chronic arthritis, infliximab and other TNFα antagonists were first developed for patients with rheumatoid arthritis. The reasons to consider this new therapeutic option also for patients with AS and other SpA were 2-fold: increased expression of TNFα in serum[31] and at the sacroiliac joints[32] in patients with AS, and the close immunological relationship between SpA and Crohn's disease, where infliximab has become already an established therapy.[33,34]

In both indications (rheumatoid arthritis and Crohn's disease), TNFα blockade not only suppresses clinical signs of inflammation and biological acute phase reactants, but also acts at the level of tissue structure. In patients with rheumatoid arthritis, this therapy protects against progression of damage to the joint space and the bony surface.[28] Meta-analysis of the data even suggests that, to some extent, there may exist a tendency towards reversibility in joint damage. In patients with Crohn's disease, efficacy of this therapy on the healing of fistulae best documents the capacity of this treatment to interfere with tissue remodelling.[35,36]

Gratacos et al.[31] used ELISA to measure, along with other inflammatory cytokines, serum TNFα levels in patients with AS, and found higher levels in patients with spondylitis compared with non-inflammatory back pain. However, TNFα levels did not correlate with laboratory or clinical parameters of disease activity. Furthermore, Braun et al.[32] performed CT-assisted biopsy of the sacroiliac joint in five patients with Bechterew's disease with radiographic stage 2–3 disease. *In situ* hybridisation showed a high amount of TNFα messenger RNA among the dense mononuclear infiltrates, suggesting a pathophysiological role of this proinflammatory cytokine in spondylitis.

Over the last decade, our group has brought together substantial clinical, epidemiological and immunological evidence linking SpA and IBD (table I).

Taken together, these studies in SpA and IBD link both diseases to a common pathophysiological background, and may point to the molecular changes in the gut in SpA as early immune phenomena in the immune cascade from SpA to IBD.[5-8,34,37-39]

The recognition of the immune link between AS and SpA with IBD has given a special impetus towards the development of new therapies in SpA.

Table I. Evidence linking spondylarthropathies (SpA) and inflammatory bowel disease

Evidence	References
Subclinical gut inflammation in patients with SpA	5-7
A fraction of patients with SpA and subclinical gut inflammation go on to develop overt Crohn's disease	8
Immunopathological changes (including E-cadherin expression) in the gut of patients with SpA resemble changes in Crohn's disease	34
T-cell cytokine profiling in the gut of patients with SpA resembles Crohn's disease	37,38
Anti-*Saccharomyces cerevisiae* IgA antibodies, which are a hallmark of Crohn's disease, are elevated in patients with ankylosing spondylitis	39

Indeed, given the immunological link between the gut in SpA and IBD on the one hand and between gut and joint inflammation in SpA on the other, it was an attractive hypothesis to test whether immunomodulators interfering with gut inflammation would also be of benefit for patients with SpA. Sulfasalazine was the first immunomodulator to be evaluated and found to be effective in patients with SpA. More recently, TNFα antagonists such as infliximab were successfully developed for AS and SPA. A special scientific challenge in this respect is the fact that more TNFα antagonists seem to be effective in AS than in IBD. Etanercept is an example of such a drug with discordant efficacy in both diseases.[40] The biological basis of this discrepancy is still under research, but it is believed that reasons for such a discrepancy may include differences in bioavailability and pharmacodynamics, as well as cell biological effects (induction of apoptosis) that may differ between different TNFα antagonists.

3. Studies with Infliximab

3.1 Open-Label Studies

On the basis of the clinical and pathogenetic link between IBD and SpA described in section 2, our group followed up four patients with Crohn's disease who were treated in an expanded access programme with infliximab at a dose of 5 mg/kg because of treatment-resistant gut inflammation.[41] All patients fulfilled European Spondylarthropathy Study Group (ESSG) criteria for SpA. Moreover, two patients fulfilled modified New York criteria for AS, peripheral arthritis was present in three patients and one patient had severe inflammatory cervical pain. In all four patients, articular signs and symptoms disappeared after infliximab treatment; however, articular disease recurred approximately 3 months after treatment in two patients. These findings suggested that refractory joint manifestations in Crohn's disease could be a potential indication for infliximab treatment, and warranted further investigation of the treatment potential of TNFα blockade in other diseases belonging to the SpA concept. As a consequence, a pilot study was set up in 21 patients with long-standing (median disease duration 17 years), active SpA according to ESSG criteria.[42] Ten patients fulfilled modified New York criteria for AS. All patients received a loading dose regimen of infliximab 5 mg/kg at weeks 0, 2 and 6. A rapid and dramatic clinical response was observed in all patients, both for axial and peripheral joint symptoms. Patient and physician assessments of disease activity, tender and swollen joint count, as well as inflammatory variables such as erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP) improved statistically significantly from day 3 onwards (all $p \leq 0.001$ compared with baseline). In patients with axial disease, after 2 weeks of treatment there was significant improvement in clinical measurements such as the Bath Ankylosing Spondylitis Metrology Index (BASMI) [$p \leq 0.05$], as well as assessments of disease activity (Bath Ankylosing Spondylitis Disease Activity Index [BASDAI]) [$p \leq 0.01$] and functional impairment (Bath Ankylosing Spondylitis Functional Index [BASFI]) [$p \leq 0.01$]. Finally, a clear amelioration of skin disease, as determined by the Psoriasis Area and Severity Index score, was seen in eight patients with psoriatic arthritis. The observed improvement was maintained over the 3-month observation period following this induction regimen. Minor adverse effects such as nausea, dizziness and headache were reported, none causing interruption or discontinuation of the treatment. No serious adverse events occurred and no peri-infusional allergic reactions were observed. Simultaneously, successful treat-

ment with infliximab was also reported in an open-label study from Germany, which treated 11 patients with AS (median disease duration 5 years) with a similar induction regimen.[43] Ten patients had substantial improvement beginning as early as 1 day after the initial infusion; one patient withdrew after the first dose because of skin rash. The median improvement of the BASDAI was 70% after 4 weeks; nine of ten patients improved >50%. The median duration of improvement before the BASDAI reached 80% of the pre-treatment value was 9.5 weeks after the third infusion (range 3–14 weeks). Subsequent open-label studies with infliximab 5 mg/kg confirmed the significant efficacy and relative safety of this drug in AS.[44,45] A Canadian study explored a lower dosage of 3 mg/kg:[46] 21 patients with AS were treated at weeks 0, 2 and 6; 17 were evaluable at week 14 (two patients withdrew because of serious adverse events, one for lack of efficacy, one lost to follow-up). After week 14, patients were retreated every 2 months; three patients required an increased dose to 5 mg/kg after 14 weeks. This study group from Berlin also reported the efficacy of infliximab in six patients with severe undifferentiated SpA. These patients received infliximab 3 or 5 mg/kg at weeks 0, 2 and 6. Over a 12-week observational period a significant efficacy was reported, with the 5 mg/kg dose having a superior effect.[47]

In total, >100 patients with AS have been treated in short-term open-label studies with infliximab; invariably, a high success rate was reported.

3.2 Controlled Studies

A double-blind comparison of infliximab with placebo in 40 patients with different subtypes of active SpA was accomplished in a monocentric study at the Ghent University Hospital (Ghent, Belgium).[48] Infusions of infliximab 5 mg/kg (n = 20) or placebo (n = 20) were given at weeks 0, 2 and 6. The primary endpoints of this study were the improvement of the patient's and the physician's assessment of global disease activity on a 100mm visual analogue scale (VAS) at week 12. Both variables improved significantly in the group treated with infliximab in comparison with the baseline value: no improvement was seen in the placebo group. At week 12, median patient global assessment (100mm VAS) dropped from 67 at baseline to 18 in the infliximab group, representing an improvement of 73% and median assessment in the placebo group deteriorated from 53.5 to 69. In the infliximab-treated group, 83% of patients experienced an improvement of their global assessment of disease activity by at least 50%, compared with 15% of patients in the placebo-treated group. The physician's global assessment showed a similar trend. There was a highly significant difference between the values for these two endpoints in the infliximab group versus the placebo group as early as week 2 ($p < 0.05$ and $p < 0.001$, respectively) and these differences were sustained up to week 12. Moreover, other disease assessments (inflammatory back pain, BASDAI, BASFI, number of peripheral tender and swollen joints, ESR and CRP) also improved significantly in the group treated with infliximab when compared with the baseline value as well as when compared with the values observed in the placebo group. In this short-term study, no significant differences (infliximab vs placebo) were detected in the metrological assessments of the spine. However, the study was not powered to detect differences in the various subgroups (AS, psoriatic arthritis and undifferentiated SpA): only 21 patients fulfilled modified New York criteria for AS (9 patients on infliximab, 12 patients on placebo).

The German group reported on a 12-week, placebo-controlled, multicentre study in patients with active AS, randomly assigning 35 patients to intravenous infliximab 5 mg/kg and 35 to placebo at weeks 0, 2 and 6.[49] The primary outcome was the number of patients experiencing a regression of disease activity (as measured by the BASDAI) of at least 50%. Fifty-three percent of patients on infliximab achieved this endpoint at week 12 compared with 9% in the placebo group, thus confirming efficacy of infliximab in the short-term treatment of patients with AS. In this study, there was also a significant improvement of function (BASFI) and metrology (BASMI).

Currently a large phase III, multicentre, 6-month, placebo-controlled study is underway.

3.3 Follow-Up Studies

One of the main concerns when using new immunomodulating biological therapies is long-term tolerability and safety, particularly the risk of serious, unusual or opportunistic infections, or the development of malignancies. In this regard, prospective follow-up of treated patient cohorts is indispensable.

In the Ghent cohort, all 21 patients who were initially included in the open-label pilot study entered into a 1-year maintenance protocol consisting of an infusion of infliximab 5 mg/kg every 14 weeks.[50] The main objective of the extension study was to analyse whether repeated infusions would effectively and safely maintain the observed effects. Nineteen of the 21 patients completed the 1-year follow-up for efficacy. Two patients changed to another treatment regimen after 12 weeks because of partial lack of efficacy; however, they were included in the safety analysis. By receiving repeated infusions, a sustained significant decrease of all disease manifestations was seen. However, the recurrence of inflammatory axial pain or peripheral arthritis in a rising number of patients before every retreatment, suggested that an interval of 14 weeks was too long to obtain continuous disease control. This led us to shorten the retreatment interval to 8 weeks. However, keeping in mind that these initial patients had longstanding disease and that, due to the described retreatment protocol, the disease control was not continuous in all patients, the significant improvement in the BASMI or other validated measures of axial mobility (such as the chest expansion) over a 1-year period was significant. Median BASMI improved from 5 (range 3–9) to 4.5 (range 2–8) at week 50 ($p \leq 0.05$). There were no major safety concerns.

All patients included in the German placebo-controlled study who had tolerated infliximab (infliximab/infliximab group) or placebo (placebo/infliximab 12-week crossover group) therapy for 3 months entered an open-label extension trial ($n = 65$). In this study,[51] infliximab was administered at a dosage of 5 mg/kg every 6 weeks after the induction phase. The primary endpoint was a 50% improvement in the BASDAI. The primary efficacy analysis at week 54 showed that approximately 50% of patients achieved this improvement. Mean BASDAI scores improved from 6.6 to 2.4 in the infliximab/infliximab group and from 6.3 to 2.6 in the placebo/infliximab group. The dosage of NSAIDs was reduced in up to 70% of patients. Significant improvements were also noted in measures of functioning, quality of life and, notably, in the metrological assessments: mean BASMI improved from 3.8 ± 2.0 at baseline to 2.3 ± 1.9 at week 54 ($p = 0.0019$).

4. Studies with Etanercept

4.1 Open-Label Studies

In 2000, Cohen[52] reported the first successful case of the treatment of an AS patient with etanercept. This was followed by a small open-label pilot study from Leeds, UK,[53] which evaluated both clinical and MRI endpoints in ten SpA patients (seven patients with AS, two with Crohn's spondylitis, one with undifferentiated SpA). There was a statistically significant improvement in all clinical and functional parameters, as well as in quality of life ($p = 0.008$ for VAS spinal pain score and BASFI, $p = 0.005$ for the BASDAI and quality of life). Entheseal lesions, detectable on MRI in nine patients, either regressed completely or improved. Concerning axial metrology, only the Schober test for lumbar flexion was reported which improved significantly at week 24 compared with baseline ($p = 0.005$).

4.2 Controlled Studies

The safety and efficacy of etanercept in AS was evaluated in three double-blind, placebo-controlled trials. Gorman et al.[54] reported on 40 patients with active AS, randomly assigned to receive twice-weekly subcutaneous injections of etanercept 25mg or placebo for 4 months. The primary endpoint was a composite of improvements (20% in three of five measures: morning stiffness, spinal pain, function-

ing, patient's global assessment of disease activity and joint swelling). Treatment with etanercept resulted in significant and sustained improvement (treatment response in 80% of the patients in the etanercept group compared with 30% in the placebo group, $p = 0.004$). The treatment response was rapid and did not diminish over time. Of the mobility measures, only the change from baseline in chest expansion differed significantly between the two groups, with an improvement in the etanercept-treated patients at the end of the study ($p = 0.006$).

Brandt et al.[55] randomised 30 patients to receive either etanercept (n = 14) at a dosage of 25mg twice weekly or placebo (n = 16) for 6 weeks; after this initial phase, all patients were switched to etanercept for a treatment period of 12 weeks. Contrary to the previous study, no other disease-modifying drugs were allowed. The primary outcome parameter was a ≥50% improvement in the BASDAI. Fifty-seven percent of the patients in the etanercept-treated group reached this regression of disease activity at week 6 versus 6% of the placebo-treated patients ($p = 0.004$). After the placebo-treated patients switched to etanercept, 56% of patients improved. Similarly, pain, function, mobility (BASMI), quality of life (all $p < 0.05$) and mean CRP levels ($p = 0.001$) improved significantly with etanercept, but not with placebo, at week 6. Disease relapses occurred 6.2 ± 3.0 weeks after cessation of etanercept, indicating that etanercept must probably be administered continuously in most AS patients to achieve permanent inhibition of the inflammatory process.

The third study was a large, multicentre, randomised, placebo-controlled trial of adults with moderate-to-severe active AS.[56] Patients (n = 277) were treated with subcutaneous etanercept 25mg (n = 138) or placebo (n = 139) twice weekly for 24 weeks. The primary outcome measures were the percentages of patients achieving the Assessments in Ankylosing Spondylitis 20% response (ASAS20) at weeks 12 and 24. Higher ASAS responses were also evaluated. Treatment with etanercept resulted in an ASAS20 response in 59% of patients at week 12 and 57% at week 24, compared with 28% and 22% in the placebo-group, respectively ($p < 0.0001$). All individual ASAS components (global assessment, pain, functioning, inflammation), acute phase reactant levels and spinal mobility measures (chest expansion, modified Schober test, occiput-to-wall distance) were also significantly improved. The safety profile of etanercept was similar to those reported in studies of patients with rheumatoid arthritis and psoriatic arthritis.

Taken together, multiple studies (both open-label and double-blind, placebo-controlled) with infliximab and etanercept indicate a remarkable efficacy on signs and symptoms of patients with AS and SpA. Axial metrology, as measured by BASMI or other accepted measures of spinal mobility (such as modified Schober test, chest expansion and occiput-to-wall distance) showed a significant improvement, especially in the longer-term follow-up studies. Radiographic evaluation, which is still the gold standard for evaluation of structure modification, was not reported in these initial studies. MRI results are discussed in section 6, but possibly only provide data on the short-term effect on the inflammatory process, and are at present not yet validated as markers of long-term structure modification.

5. Safety of Anti-TNFα Therapy

The major impact of TNFα blockade on the immunological system raises some concerns about the safety of this approach, especially with regard to severe infections, malignancies and immune-mediated diseases. In rheumatoid arthritis and Crohn's disease, studies indicate a higher incidence of tuberculosis reactivation and the induction of antinuclear antibodies, but no major effect on the occurrence of malignancies or lupus-like syndromes. Since TNFα is an essential cytokine in the innate immune response and in host defence mechanisms, and since defective host defence plays a role in the pathogenesis of reactive arthritis and probably also other SpA subtypes, the occurrence of severe infectious complications is a major concern for TNFα blockade in SpA.

A systematic review of safety in a cohort of SpA patients on infliximab treatment (107 patients ex-

tending over 191.5 patient-years of treatment)[57] revealed the following. Severe infections were observed in eight patients, leading to interruption of infliximab therapy in five patients. Most striking was the occurrence of tuberculosis reactivation in two patients. Both patients developed fever and malaise 9 weeks after start of infliximab therapy, and presented with mediastinal lymphadenopathy and nodular lesions in the liver and spleen. Diagnosis was confirmed by histology and culture of lymph node material obtained by mediastinoscopy. Both patients recovered with tuberculostatic therapy. The presentation of these two cases in SpA was similar to other cases in rheumatoid arthritis and Crohn's disease.[58] The reported frequency of tuberculosis in association with infliximab was much higher than that of other opportunistic infections, and was higher than the available background rates. This suggests the need for appropriate screening and, when appropriate, effective antituberculosis treatment in all patients prior to initiation of anti-TNFα therapy.

The reactivation of tuberculosis does not come totally unexpected since TNFα has a special role in the organisation of granulomata, which is of relevance in the defence against intracellular microorganisms such as *Mycobacterium tuberculosis*.[59,60] Detailed analysis of the histology of biopsied lymph nodes in the case of tuberculosis reactivation indicates the presence of granuloma-like structures with multinucleated giant cells and areas of necrosis, but with complete absence of a palisading layer of histiocytes. Although this supports the hypothesis that TNFα blockade can influence the structural integrity of tuberculous granulomas and can, thus, contribute to spreading of the disease, the exact mechanisms involved are not well defined. Recent evidence in animal models indicates that TNFα is a critical regulator of type 1 immune activation during intracellular bacterial infection and that suppression of TNFα in mycobacteria-infected mice leads to apoptosis and necrosis of granuloma lung structures mediated by T helper (Th)-1 cells.[61,62] Of interest, infliximab does not appear to affect other types of granulomata such as rheumatoid nodules.[63]

Other infections in the studied SpA cohort included three patients who developed retropharyngeal abscesses (in one of these patients, blood cultures were positive for *Streptococcus pyogenes*) and three patients who developed palmoplantar pustulosis, which has been linked to tonsillitis and focal nasopharyngeal infections with streptococci. These data suggest that blocking TNFα might impair the cellular defence against streptococci and, thereby, contribute to the development of both retropharyngeal abscesses and palmoplantar pustulosis. In rheumatoid arthritis, occurrence of serious bacterial infections has been linked to downregulation of toll-like receptors, a hypothesis which should be further evaluated in SpA.[64] The three other major infections were one case of sepsis and two of procedure-related infections. There were no cases of malignancies or lupus-like syndromes, although induction of antinuclear antibodies and anti-double strand (ds)DNA antibodies was frequent (see section 7).

Other studies with infliximab in SpA showed similar safety profiles. In a double-blind, placebo-controlled trial in AS, 3 of the 34 infliximab-treated patients had to stop treatment because of a serious adverse event, including one case of systemic tuberculosis.[49] In the 1-year open-label extension of this study, an additional 4 of 30 patients had serious adverse events, including development of peripheral arthritis and liver function abnormalities.[51] Thus, both the Belgian and the German patient cohort indicate the occurrence of serious adverse events in 10–20% of patients.

Although clinical trials with etanercept in AS and SpA cannot be compared directly with infliximab since they were performed in other patient cohorts (including a reduced risk of tuberculosis reactivation in the US than in Europe), etanercept treatment in SpA was associated with an increased risk of upper respiratory tract infections, but severe adverse events such as tuberculosis or opportunistic infections were not reported.[49,55,56]

The potential risk of anti-TNFα therapy for development of malignancy, and lymphoma in particular, has been suggested by data from different sources. Brown et al.[65] identified 26 cases of lym-

phoproliferative disorders following treatment with etanercept (18 cases) or infliximab (8 cases), after a median time period of 8 weeks. In two cases, lymphoma regression was observed following discontinuation of anti-TNFα treatment, in the absence of specific cytotoxic therapy directed toward the lymphoma.

In a prospective study involving 18 572 patients with rheumatoid arthritis, the overall standardised incidence ratio for lymphoma was 1.9.[66] The same ratio was 1.7 for methotrexate users, 2.6 for infliximab and 3.8 for etanercept, while it was 1.0 for those not receiving methotrexate or biologicals. Lymphoma was associated with increasing age, male sex and education. The investigators suggested that the increased lymphoma rates observed with anti-TNFα therapy may reflect channeling bias, whereby patients with the highest risk of lymphoma preferentially receive anti-TNFα therapy. Recently, the US FDA reviewed safety data with particular focus on the incidence of neoplasia and lymphoma in patients receiving TNFα antagonists. In the combined clinical trial population with infliximab for rheumatoid arthritis and Crohn's disease, there was approximately 6-fold higher incidence rate for lymphoma, compared with the general population. However, patients with chronic arthritis and Crohn's disease with highly active disease and chronic exposure to immunosuppressive drugs, may be at higher risk than the general population for the development of this complication. Yet, as a result of the recent US FDA evaluation, a warning concerning malignancy has been added to the labelling for all therapeutic agents that block TNFα.

6. Imaging Studies During Anti-TNFα Therapy in AS and SpA

In rheumatoid arthritis, the rheumatic indication for anti-TNFα therapy that has been most extensively studied, both infliximab and etanercept therapies have shown to interfere significantly with the progression of structural damage. Lipsky et al.[28] treated 428 patients who had active rheumatoid arthritis despite methotrexate therapy with placebo or infliximab at intravenous dosages of 3 or 10 mg/kg every 4 or 8 weeks in combination with oral methotrexate for 54 weeks, and assessed clinical responses as well as the effect on joint damage. The combination of infliximab and methotrexate resulted in a sustained reduction in the signs and symptoms of rheumatoid arthritis. Also, radiographic evidence of joint damage increased in the group given placebo and methotrexate, but not in the group given infliximab and methotrexate. Radiographic evidence of progression of joint damage was absent in infliximab-treated patients whether or not they had a clinical response. For etanercept in rheumatoid arthritis, Genovese et al.[67] compared the clinical and radiographic outcomes in patients with rheumatoid arthritis who received monotherapy with either etanercept or methotrexate. 632 patients with early, active rheumatoid arthritis were randomised to receive either twice-weekly subcutaneous etanercept (10 or 25mg) or weekly oral methotrexate for at least 1 year in a double-blind manner. Following the blinded phase of the trial, 512 patients continued to receive the therapy to which they had been randomised for up to an additional year, in an open-label manner. At 24 months, more etanercept 25mg than methotrexate patients had no increase in total score and erosion scores on the Sharp scale. The mean changes in Sharp total and erosion scores in the etanercept 25mg group were significantly lower than in the methotrexate group.

For AS and SpA, anti-TNFα therapy is more recently introduced, and data on the effect of this new therapy on the structural level, as evaluated by imaging, are more scarce. Validated imaging scores in this disease are also introduced more recently than in the case of rheumatoid arthritis, and it appears that, in relation to anti-TNFα therapy, limited imaging data – mostly MRI – are available. To date, such imaging has provided more information on the inflammatory process occurring in the bone tissue and at the site of the enthesis than on the loss or reparation of the structural tissue.

Braun et al.[68] evaluated an MRI scoring system for the assessment of spinal inflammation in patients with AS who participated in a placebo-controlled trial of infliximab, and examined whether this anti-

TNFα compound is also effective for the reduction of MRI-proven spinal inflammation. Twenty patients with AS were examined at baseline and after 3 months. Nine patients received infusions with infliximab 5 mg/kg at weeks 0, 2 and 6, and 11 patients received placebo. Chronic or active lesions were evaluated by specific MRI sequences. Active spinal lesions were detected in 15 of 20 patients (75%). Depending on the score used, active lesions improved in the infliximab group by 40–60% compared with a small improvement or even deterioration in the placebo group. Chronic lesions improved by 7% in the infliximab group but worsened by 35% in the placebo group. The active MRI changes correlated with improvement in the BASDAI score. Thus, the investigators were able to demonstrate significant regression of spinal inflammation in the infliximab-treated group using the MRI activity scores.

Maksymowych et al.[46] included MRI in six patients in a prospective, observational cohort of patients with NSAID-refractory AS, in whom infliximab therapy (3 mg/kg) was given. MRI evaluation included dynamic MRI with gadolinium augmentation of affected joints. Maximal rate of augmentation was determined at baseline and 84 days. MRI-defined gadolinium augmentation was significantly decreased ($p = 0.04$). Also, Stone et al.[44] reported improvement of MRI imaging of inflammatory changes in nine AS patients treated with infliximab.

Marzo-Ortega et al.[53] studied the effect of etanercept 25mg subcutaneously twice weekly on the clinical manifestations of resistant SpA and on axial and peripheral entheseal lesions using MRI (scans of sacroiliac joints, the lumbar spine and affected peripheral joints with a 1.5T scanner employing T1-weighted, T2-weighted fat-suppressed and T1-weighted fat-suppressed postgadolinium sequences) at baseline and at 6 months. Enthesitis and associated osteitis were scored semiquantitatively in pre- and post-treatment scans. Nine patients had a total of 44 MRI-detectable entheseal lesions. Overall, 86% of MRI-detected entheseal lesions either regressed completely or improved. No new lesions developed. Thus, the clinical effect of etanercept was shown to be associated with marked improvement of enthesitis and associated osteitis pathology, as determined by MRI.

Osteoporosis is frequently associated with AS. The group at Cochin Hospital (Paris, France) evaluated the changes in bone mineral density (BMD) in 29 patients with SpA treated with infliximab.[69,70] Twenty-five patients were treated with infliximab 5 mg/kg and four with 3 mg/kg at weeks 0, 2 and 6, and then received either no infusion ($n = 3$) or additional infusion of infliximab every other month ($n = 6$), and the remainder received one infusion only in the case of a relapse. Lumbar and femoral BMD was measured by dual energy x-ray absorptiometry at baseline and 6 months later. Serum osteocalcin and urinary deoxypyridinoline were measured in 19 patients at weeks 0, 2 and 24, and in 13 patients at all visits. In 6 months there was a significant increase in BMD at the spine (3.6%), total hip (2.2%) and trochanter (2.3%). There was an increase in osteocalcin between baseline and week 6 (third infusion).

7. Biological Studies Suggesting Structure-Modifying Capacities of Anti-TNFα Therapy in AS and SpA

Whereas clinical measurements might be relatively insensitive for assessing structure-modifying capacities and radiological measurements require longer term follow-up, it is tempting to hypothesise that evaluation of the biological immunomodulation by TNFα blockade might provide additional evidence for a disease- or even structure-modifying effect in SpA. In this context, structure modification should not only be seen as inhibition of bone and cartilage destruction but more broadly as modulation of tissue histology rather than just downregulation of inflammation.

The first evidence that TNFα blockade in SpA had not only an anti-inflammatory effect but also a profound impact on the immune alterations underlying the disease came from the study of Th1/Th2 profiles during infliximab treatment. Patients with SpA have an impaired Th1/Th2 balance, with decreased T-cell production of interferon (IFN)-γ, TNFα and interleukin (IL)-2, and increased IL-10

synthesis.[71,72] The impaired Th1 cytokine production is highly compatible with the clinical efficacy of TNFα blockade, since high TNFα levels can suppress T-cell activation *in vitro*.[73] Treatment with three infusions of infliximab in SpA patients resulted in a rapid and sustained increase of Th1 cytokines (IFNγ and IL-2) to levels comparable with those in healthy controls.[71] A reduction of IL-10-positive T cells was observed in those patients with high baseline values. However, this effect was only observed in the first 4 weeks. No effect was seen on IL-4 production. These findings were globally confirmed by other studies with both infliximab and etanercept.[74,75] Recently, one study suggested a decrease rather than an increase of the Th1 response in AS patients treated with infliximab,[76] but these data should be interpreted with care since they might have been biased because of technical issues.[77] Together, these data support the view that TNFα blockade essentially reverses the state of anergy of Th1 cells, while no significant effect is observed on Th2 cells, a concept which fits with the recent data on TNFα as a negative regulator of type I immune responses.[61]

Considering the impressive clinical efficacy of infliximab in SpA, further studies were undertaken to analyse the effect on immunopathological and structural features of SpA. The major drawback for histopathological investigations is that most tissues targeted by the disease are not easily accessible for biopsy sampling (uvea, axial skeleton, sacroiliacal joints, enthesis) and can, thus, only be assessed indirectly by new imaging techniques such as MRI. Regarding peripheral joint inflammation, the availability of needle arthroscopy allowing repeated synovial biopsy sampling has led to two studies. In the first study, the synovial histopathology was studied in eight patients (three AS, one undifferentiated SpA and four psoriatic arthritis patients). These patients were treated with infliximab 5 mg/kg at week 0, 2 and 6 and synovial biopsies were obtained at baseline, week 2 and week 12.[78] At baseline, the synovial tissue samples depicted the typical characteristics of SpA synovitis: a moderate lining hyperplasia, a strong hypervascularity with endothelial activation, and a moderate and diffuse inflammatory infiltration with macrophages as well as lymphocytes and polymorphonuclear cells. Evaluation at week 2 indicated a significant reduction in numbers of macrophages and polymorphonuclear cells in the sublining layer as well as an impaired expression of vascular cell adhesion molecule (VCAM)-1, suggesting that infliximab acts on SpA synovitis by reducing the influx of inflammatory cells. The effect on macrophages, neutrophils and VCAM-1 was maintained at week 12, with an additional trend for reduction of CD4+ lymphocytes. Interestingly, two other effects were observed at week 12. First, there was a decrease of the hypervascularity and a trend to reduction of the synovial lining hyperplasia, indicating that infliximab modulates not only inflammation as such but also the structural synovial characteristics of the disease. Secondly, there was a significant increase in the number of B cells and plasma cells in the synovium of the infliximab-treated patients suggesting that, at least in SpA, infliximab does not affect B-cell homing and/or maturation.

We performed an extension of this study in a second cohort of 15 patients,[79] in which 12 of the patients were treated with infliximab 5 mg/kg at weeks 0, 2 and 6, and three patients with placebo. In the infliximab-treated group, evaluation at week 12 confirmed the reduction in lining layer hyperplasia, VCAM-1 expression in the lining and hypervascularity. This was paralleled by a significant decrease in serum levels of soluble vascular endothelial growth factor (VEGF), E-selectin and intercellular adhesion molecule (ICAM)-1 in these patients. However, there was no decrease in serum levels of soluble VCAM-1. There was a decrease in overall cell infiltration and number of macrophages, polymorphonuclear cells, CD3+, CD4+ and CD8+ T lymphocytes but no decrease at all of B cells and plasma cells. Whereas these data confirm the previous observations in an independent patient cohort, more recent studies extended this concept and demonstrated clearly that infliximab has a major impact on the presence and influx of specific inflammatory macrophage subsets in the synovial membrane, including the pathogenic CD163-positive macro-

phages and the myeloid-related protein (MRP)-8/MRP14-positive infiltrating monocytes.[80-82]

Of particular interest is the differential effect of infliximab on T and B cells. As indicated, the infiltration of the synovial membrane with B lymphocytes and plasma cells was not decreased, but increased after infliximab treatment.[78] However, no follicular organisation or germinal centre formation was observed. This is compatible with findings in animal models indicating that TNFα deficiency is associated with disturbed follicle formation, follicular dendritic cell networks and germinal centre formation.[83,84] Moreover, follicular exclusion has an important impact on B-cell maturation and differentiation, including isotype switch.[85,86] Whereas data on the effect of TNFα blockade on humoral immunity are still scarce, it is well known that infliximab treatment in rheumatoid arthritis and Crohn's disease may induce autoantibodies such as antinuclear antibodies and antibodies to dsDNA, which my be related to lupus. We described the induction of antinuclear, anti-dsDNA and other fine antinuclear autoreactivities in a well defined cohort of 35 SpA patients, treated in the context of a placebo-controlled trial, where the placebo group received active treatment with infliximab after 12 weeks.[87] At baseline, 6 of 35 (17.1%) patients were positive for antinuclear antibodies, whereas after infliximab treatment 31 of 35 (88.6%) patients tested positive at week 34 ($p < 0.001$ vs baseline). An increase in fluorescence intensity for antinuclear antibody testing of two or more steps (on a 0–5+ scale) following infliximab therapy was observed in 26 of 35 (74.3%) patients. Initial baseline screening with indirect immunofluorescence on *Crithidia luciliae* and ELISA revealed that none of the 35 SpA patients were positive for anti-dsDNA antibodies in either assay. At week 34, 6 of 35 (17.1%) SpA patients were positive for anti-dsDNA antibodies ($p = 0.031$ vs baseline). The induced anti-dsDNA antibodies were further isotyped. None of these six SpA patients had anti-dsDNA antibodies of the IgG class at week 34. In three of six SpA patients, the anti-dsDNA antibodies were of both the IgM and IgA class and two of six patients were positive for anti-dsDNA antibodies of the IgM class. However, no lupus-like symptoms were observed during the investigational period. Longer-term follow-up of these patients is needed to counter the concern of clinically relevant lupus induction completely. Taken together, these data support the concept that TNFα blockade has a strong influence on humoral immunity, which could be partially mediated by an effect on follicular exclusion. Further studies are currently being performed to investigate specific mechanisms underlying this process, to assess if this is restricted to autoantibodies or also extends to other humoral responses, and to study if this is a class effect of TNFα antagonists or only specific for infliximab.

The effect of infliximab on vascularity and lining layer hyperplasia suggests that TNFα blockade not only influences inflammation as such, but also has an effect on the synovial architecture. More recent studies have investigated in further detail some of these aspects and have started investigating the mechanism underlying a structure modification by infliximab. One study[88] focused on the synovial membrane of patients with psoriatic arthritis subtypes, and confirmed the pronounced effect of infliximab on the vascularity. The study data indicated both a decrease of the neovascularisation marker αVβ3 and a downregulation of VEGF and its receptor VEGFR2, and also an increase in angiotensin II. Taken together, these data suggest that infliximab treatment in SpA contributes to an inhibition of neovascularisation as well as to vascular regression. Of particular interest, the integrin αVβ3 has not only been implicated in neovascularisation but also in aggressive behaviour of synovial fibroblasts and in bone resorption by osteoclasts. How far these processes are also influenced by TNFα blockade *in vivo* remains to be investigated.

Another study[89] investigated extensively the matrix metalloproteinases (MMPs) in SpA synovitis. First, the investigators indicated that MMPs and their inhibitors (tissue inhibitors of MMPs [TIMPs]) are expressed equally in SpA and rheumatoid arthritis synovitis. Secondly, they demonstrated that serum levels of MMP3 reflect the local production in

the joint of this particular MMP well and, in contrast with CRP and ESR, might be a useful biomarker for peripheral joint disease in SpA. Finally, they showed a highly significant and rapid downregulation of synovial MMPs and TIMPs by infliximab treatment. Since MMPs are involved in neovascularisation, matrix degradation, and cartilage and bone destruction, this biological effect may support the concept that TNFα blockade could influence structural damage in the long term. Other cellular and molecular mechanisms involved in bone and cartilage destruction, such as osteoclastogenesis and osteoclast-mediated bone resorption in psoriatic arthritis, have been shown to be TNFα-dependent *in vitro*[90] but how far they are influenced by TNFα blockade *in vivo* in SpA is yet to be investigated. Molecular and cellular mechanisms of tissue remodelling in SpA certainly deserves further attention as they could represent unique short-term biomarkers to predict the long-term effect of new treatment modalities on the structural damage in SpA.

8. Conclusion

AS is a disease frequently leading to significant functional impairment. Anti-TNFα therapy has brought new perspectives in this condition. Two agents are currently used in the treatment of AS: infliximab and etanercept. Open-label studies have shown the efficacy of these drugs and these findings have been confirmed by controlled studies, at least in the short term. Improvements in several clinical parameters, function, quality of life, biological parameters, histopathological synovial characteristics and MRI have been observed. Because of these favourable results, anti-TNFα therapy has been approved for the treatment of AS and should be considered for patients with severe axial symptoms and elevated serological markers of inflammatory activity who have responded inadequately to conventional nonsteroidal therapy. Some questions remain unanswered, such as the long-term efficacy and safety of anti-TNFα therapy, the extent of structural benefit and its cost effectiveness. Despite these concerns, anti-TNF therapy represents a major therapeutic advancement in the treatment of AS.

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