Available online http://arthritis-research.com/content/3/1/018

Review

Anti-cytokine therapy in chronic destructive arthritis

Wim B van den Berg

Department of Rheumatology, University Medical Center St Radboud, Nijmegen, The Netherlands

Correspondence: Wim B van den Berg, PhD, Rheumatology Research Laboratory, University Medical Center St Radboud, 6500 HB Nijmegen, The Netherlands. Tel: +31 243 616540; fax: 31 243 540403; e-mail: w.vandenberg@reuma.azn.nl

Received: 8 September 2000
Accepted: 18 October 2000
Published: 10 November 2000

Arthritis Res 2001, **3**:18–26

(Print ISSN 1465-9905; Online ISSN 1465-9913)

Abstract

Tumor necrosis factor (TNF) and interleukin-1 (IL-1) are considered to be master cytokines in chronic, destructive arthritis. Therapeutic approaches in rheumatoid arthritis (RA) patients have so far focused mainly on TNF, which is a major inflammatory mediator in RA and a potent inducer of IL-1; anti-TNF therapy shows great efficacy in RA patients. However, it is not effective in all patients, nor does it fully control the arthritic process in affected joints of good responders. Directed therapy for IL-1, with IL-1 receptor antagonist, mainly reduces erosions and is marginally anti-inflammatory. It is as yet unclear whether the limited effect is akin to the RA process or linked to suboptimal blocking of IL-1. Analysis of cytokine patterns in early synovial biopsies of RA patients reveals a marked heterogeneity, with variable staining of TNF and IL-1β, indicative of TNF-independent IL-1 production in at least some patients. Evidence for this pathway emerged from experimental arthritises in rodents, and is summarized in this review. If elements of the models apply to the arthritic process in RA patients, it is necessary to block IL-1β in addition to TNF.

Keywords: animal models, cytokines, inflammation, interleukin-1, joint destruction, tumor necrosis factor

Introduction

Studies in well-defined animal models of arthritis make it clear that tumor necrosis factor (TNF) is involved in early joint swelling. However, TNF alone is neither arthritogenic nor destructive and exerts its arthritogenic potential through the induction of interleukin-1 (IL-1). Intriguingly, TNF-independent IL-1 production is found in many model situations, including pathways driven by macrophages, T cells and immune complexes. Its relevance is underlined by the great efficacy of anti-IL-1 therapy and a total lack of chronic, erosive arthritis in IL-1β-deficient mice. Osteoclast-mediated bone erosion is stimulated by IL-1 as well as by the combination of TNF and T-cell-derived IL-17. Cartilage erosion is dependent on IL-1β and is greatly enhanced in the presence of immune complexes.

Arthritogenic potency of TNF and IL-1

The synovial reaction in RA patients is characterized by the abundance of many cytokines, chemokines and growth factors. It is now generally accepted that TNF and IL-1 are master cytokines in the process of chronic joint inflammation and the concomitant erosive changes in cartilage and bone. Proinflammatory and destructive properties were first demonstrated in culture studies *in vitro* and the arthritogenic potential of TNF and IL-1 was substantiated by arthritis induction in rodents. Arthritis could be elicited by local injection of recombinant cytokines in the knee joint [1,2]; this observation was underlined by the occurrence of chronic, erosive arthritis in transgenic mice displaying general TNF overexpression [3]. Interestingly, the dominant expression of TNF-mediated pathology in

AA = adjuvant arthritis; AIA = antigen-induced arthritis; IL-1 = interleukin-1; IL-1ra = IL-1 receptor antagonist; MMP = matrix metalloproteinase; NF-κB = nuclear factor-κB; OPG-L = osteoprotegerin ligand; RA = rheumatoid arthritis; SCW = streptococcal cell wall fragments; TNF = tumor necrosis factor.

joint tissues in these transgenic mice is still largely unexplained. More recently, further proof of arthritogenicity was obtained from the induction of arthritis by local overexpression of cytokines in joint tissues by using viral vectors [4].

Intriguingly, IL-1 is much more potent than TNF in inducing cartilage destruction *in vivo*. Tiny amounts of IL-1 are sufficient to cause proteoglycan synthesis inhibition in chrondrocytes, whereas a roughly 100–1000-fold higher dose of TNF is needed to obtain the same effect [1]. Importantly, synergy between IL-1 and TNF has been demonstrated [2]. Apart from potency differences, it is clear that it is hard to measure significant TNF levels in inflamed synovial tissue or synovial fluid of RA patients and the levels are certainly not higher than those of IL-1. Most effects might be related to membrane-bound forms of cytokines, which are hard to measure. In contrast, impact on articular cartilage from synovium-derived mediators probably needs trafficking of soluble forms. The situation might be different at sites of pannus overgrowth, where close contact between synovial cells and chondrocytes does occur.

A strong argument for the limited, direct role of TNF in arthritis has emerged from elegant studies in TNF transgenic mice. Joint inflammation was completely arrested when these mice were treated with antibodies against anti-IL-1 receptor [5]. This argues that the pathology runs through the induction of IL-1, which is the real arthritogenic trigger, either alone or in synergy with TNF. TNF levels were still high after treatment with antibodies against IL-1 receptor, which implies that TNF alone is hardly arthritogenic.

TNF and IL-1 as therapeutic targets in arthritis

Both animal model studies and clinical observations have contributed greatly to the identification of TNF and IL-1 as useful therapeutic targets. Apart from the obvious demonstration that arthritis in TNF transgenic mice could be blocked with anti-TNF antibodies, it was a major breakthrough to note that collagen type II arthritis, the classical RA model in rodents, could be suppressed with anti-TNF antibodies or TNF soluble receptors [6–8]. This identified a key role of TNF in autoimmune arthritis. Further studies on this model revealed that TNF blockade was efficient when started before or shortly after the onset of arthritis, whereas anti-IL-1 treatment was at least as efficient and also arrested advanced arthritis and joint destruction [9,10]. Studies in TNF receptor knockout mice have demonstrated that the incidence and severity of collagen arthritis were less in such mice. However, once the joints were affected, full progression to erosive damage was seen in an apparently TNF-independent fashion [11].

Similar studies with neutralizing antibodies have been performed in a range of arthritis models. The relative roles of TNF and IL-1 in early joint inflammation were variable in different models, but the crucial role of IL-1 in late arthritis and erosive joint destruction was a consistent finding. This implies that overkill by other mediators might occur in the inflammatory process, and that the stimulus, type of process and probably also the stage of the arthritis are major determinants of the mediator profile. Intriguingly, IL-1 seems to be a suitable downstream target in joint erosion (see below).

Clinical trials with anti-TNF/anti-IL-1

In addition to the evidence from studies on animal models, the cytokines TNF and IL-1 were demonstrated in increased quantities in RA synovial tissue, along with the presence of cell-associated receptors for these cytokines [12]. The remarkable anti-inflammatory activity of a first neutralizing monoclonal anti-TNF antibody in RA patients revealed the potential of anti-cytokine therapy [12] and has subsequently stimulated the development of improved anti-TNF reagents such as fully humanized antibody and engineered fusion proteins of TNF soluble receptors and Fc fragments, with reduced immunogenicity and a prolonged half-life. There is no doubt that TNF blockers provide impressive protection against pain and joint swelling in most RA patients [12–19]. It is also evident that anti-TNF therapy is not effective in all RA patients, nor does it control arthritis in all affected joints of good responders.

The initial studies targeting IL-1 were performed with soluble IL-1 type I receptor. Clinically relevant effects were not seen, which was disappointing at the time and raised questions about the relevance of IL-1 as a therapeutic target in human RA [20]. However, it is now understood that the choice of type I receptor was unfortunate because this soluble receptor has a high affinity for IL-1 receptor antagonist (IL-1ra), thus scavenging the endogenous IL-1 inhibitor. In that sense, the decoy type II receptor might make a better inhibitor, but it has the disadvantage that it has a lower affinity for IL-1. Studies are awaited with optimal, engineered IL-1 receptor fragments or potent, neutralizing anti-IL-1 antibodies.

Apart from studies with soluble receptors, clinical trials have also employed IL-1ra, the effect on joint inflammation being limited. A significant reduction of joint erosion was evident [21,22]. IL-1ra has a weak pharmacokinetic profile and it is still unclear whether the limited effect on joint inflammation is akin to the RA process or related to suboptimal blocking of IL-1. Comparisons with animal model studies teach us that continued high dosing is crucial in fully controlling IL-1. Collagen arthritis could not be controlled with a repeated daily injection of IL-1ra, but great suppressive effects were achieved with IL-1ra supplied with an osmotic minipump [9]. Similarly, local IL-1ra overexpression in the knee joint with viral vectors

showed proper efficacy in this model [23]. Until high-quality IL-1 blockers become available for clinical trials, conclusions on the relative roles of TNF and IL-1 in RA patients have to be made with great care.

Anti-TNF treatment is anti-erosive?

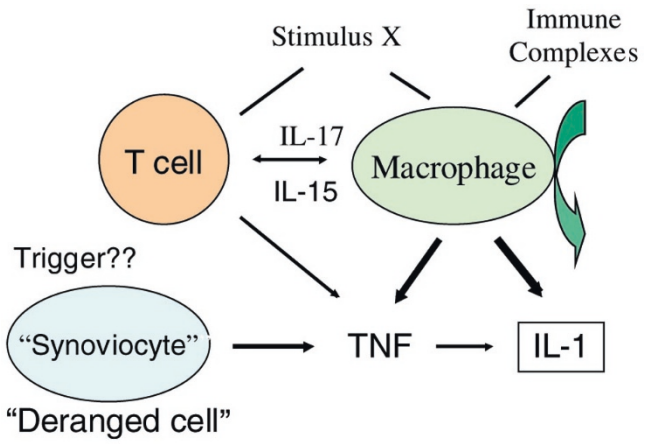
Remarkably, the recent evaluation of joint erosions after the treatment of RA patients with anti-TNF provided the first evidence for a joint protective effect, as reported at the 1999 ACR meeting. This was shown for a combination of anti-TNF antibodies with methotrexate, and also for a single treatment with antibodies as well as TNF soluble receptor. Unfortunately, the actual data have not yet been published, hampering detailed attention in this review. The finding might fit with the hypothesis that TNF overproduction in RA synovial tissue is mainly caused by deranged behaviour of synoviocytes, generating too much TNF. If this is so, TNF will drive IL-1 production and TNF blockade will be sufficient to control this TNF–IL-1 pathway (Fig. 1). Intriguingly, it is in line with the initial hallmark observation made in RA synovial cell cultures: the addition of neutralizing anti-TNF antibodies strikingly reduced the production of IL-1 [24]. Unfortunately, this observation was made with isolated cell cultures, has not been confirmed by others and awaits confirmation for intact synovial tissue.

It is still too early to accept the above observations as final proof of concept of a dominant TNF–IL-1 cascade in RA synovial tissue. Anti-TNF antibodies used in clinical studies display cytotoxic effects. This implies that one mechanism of the anti-TNF effect could be linked to binding to TNF-bearing cells and subsequent killing of these cells, potentially including TNF/IL-1-producing cells or neighbouring cells. In addition, the TNF soluble receptor used in some of the anti-TNF trials not only binds to TNF but also scavenges lymphotoxin. The latter might have an impact on T-cell-driven pathways. Significantly, TNF rather than lymphotoxin seems to be the major cytokine expressed in RA synovial tissue. A final comment to be made here is that analysis of anti-erosive efficacy in clinical trials is based mainly on bone erosions. Focal damage of cartilage is more difficult to score on X-rays. It remains to be seen whether the relative dominance of TNF and IL-1 involvement and amplifying elements by pathways mediated by T cells and immune complexes are similar or different in the destruction of cartilage and bone in RA (see below).

Heterogeneous synovial cytokine patterns in RA patients

Synovial biopsies taken from knee joints of RA patients and analysed for cytokine immunolocalization or mRNA levels identified a rather variable pattern [25–30]. Although TNF was abundantly present in some RA patients, TNF was undetectable in half of the patients. This limited presence

Figure 1



Simplified view of potential pathways of TNF overproduction. Deranged synoviocyte-mediated TNF production might initiate a TNF–IL-1 cascade. Note that general triggering of T cells or macrophages, as studied in arthritis models, gives rise to both TNF and IL-1, with considerable TNF-independent IL-1 production, and skewing to IL-1 when immune complexes are used as the stimulus.

of TNF might be interpreted as an explanation for the high efficacy of anti-TNF treatment in most patients, because the levels to be neutralized are low [27]. Alternatively, it might be seen as evidence for considerable heterogeneity of the RA process in different patients. The apparent absence of TNF in some patients and the fact that anti-TNF therapy is not efficacious in all RA patients argues for multiple pathways. Moreover, the cytokine interplay might vary in different stages of the process in the same patient; repeated sampling in large groups of RA patients is needed to shed more light on this issue.

Intriguingly, IL-1β was found in most RA synovial biopsies [28]. In addition, interferon-γ levels were low but IL-17 was prominent in many RA patients [31], arguing for a reconsideration of T cell involvement. It seems obvious that future therapy will consist of combination treatment, at least involving both TNF and IL-1. It is tempting to speculate that tailor-made cytokine-directed therapy will be applicable in the near future, based on individual cytokine patterns.

Role of TNF and IL-1 in various animal models of arthritis

Further insight into the relative roles of TNF and IL-1, as well as the TNF dependence of IL-1 production under various arthritogenic conditions, has emerged from the efficacy of TNF and IL-1 blocking in a range of well-defined experimental arthritis models (Table 1). The model systems include nonimmune triggering of macrophages as well as different mixtures of arthritogenic pathways driven

Table 1

Cytokine involvement in various experimental arthritis models

				Cytokine involvement			
				Early inflammation		Erosive arthritis	
Arthritis Model	Species	References	Principal target or trigger	TNF	IL-1	TNF	IL-1
SCW-A	Mouse	[32]	Macrophages	++	–	–	++
SCW flare	Mouse	[33]	T cells/macrophages	+	+	–	++
SCW flare	Rat	[46,77]	T cells/macrophages	+	+		++
AIA	Mouse	[35,36,38]	T cells/IC	±	±		++
AIA	Rabbit	[37]	T cells/IC	+	+	±	++
AIA flare	Mouse	[38]	T cells	±	+	–	++
CIA	Mouse	[6–11]	T cells/IC	+	++	+*	++
ICA	Mouse	[40]	IC	–	++	–	++
AA	Rat	[45]	T cells	+	+	+*	+

Cytokine involvement, based on experiments with neutralizing antibodies, started at the onset of arthritis. It does not reflect potential involvement during preimmunization (CIA and AIA). SCW-A flares reflect the situation after three consecutive SCW flares with intervals of 7 days. AIA flare was induced by antigen rechallenge at day 21. CIA, collagen-induced arthritis; ICA, passive immune complex (IC) arthritis. *Efficacy of anti-TNF treatment only when started at onset of first signs of arthritis.

by T cells and immune complexes. Some crucial findings with the specific TNF and IL-1 blockers have recently been backed up with analyses of models in mice deficient in TNF and IL-1β. The latter type of approach excludes misinterpretations potentially linked to variability in the blocking quality of the different neutralizing antibodies and soluble receptors used in the experimental studies.

Macrophage-driven arthritis

The strongest TNF dependence of acute inflammation is found when arthritis is induced locally with a phlogistic trigger such as streptococcal cell wall fragments (SCWs) or yeast particles (Zymosan), as observed in rats and mice. Erosions are mild in this model but still develop after treatment with anti-TNF antibodies; this observation has been strengthened by the high degree of erosions when such models were induced in TNF-deficient mice [32,33]. Not surprisingly, IL-1 levels were still high under these conditions, identifying considerable TNF-independent IL-1 triggering. When repeat injections with SCW were given, the inflammation became partly dependent on IL-1, and erosions still developed in TNF-deficient mice and were fully absent in IL-1β-deficient mice. The repeated flare model became more dependent on T cells and arthritis seemed markedly reduced in lymphotoxin-deficient mice (WB van den Berg, unpublished data), in line with findings in encephalomyelitis [34].

Mixed T cell and immune complex pathways

Collagen-induced arthritis and antigen-induced arthritis (AIA) are models based on preimmunization with a cartilage-specific autoantigen or an exogenous protein, with the generation of T cell reactivity and antibodies. The onset of arthritis is a mixture of pathways driven by immune complexes and T cells. In AIA the onset of arthritis is vigorous and at best partly dependent on TNF and IL-1, but the cartilage erosion and propagation of inflammation are dependent on IL-1 [35–38]. In contrast, TNF is important at the onset of collagen arthritis, but IL-1 blocking is highly efficacious in this model both in the acute inflammatory state and in the advanced erosive disease. In line with this, IL-1ra-deficient DBA mice show enhanced susceptibility to collagen-induced arthritis [39]. Apparently, T cells are important mainly in the early stages, to support the production of sufficient collagen type II autoantibody, and the propagation is driven by immune complexes, showing strong dependence on IL-1.

Immune complex arthritis

A remarkable finding was the strong IL-1 dependence of the inflammatory response induced with plain immune complexes [40]. Recently, a novel autoimmune arthritis model was generated in KRN mice by the transgenic overexpression of a T cell receptor directed against MHC (major histocompatibility complex) molecules. This transgenic condition resulted in a skewed control of tolerance and was characterized by significant formation of autoantibodies. The crucial observation was that the model was transferable with purified autoantibodies, underlining the arthritogenic potential of such a pathway [41,42]. Interestingly, this model seemed to be dependent not on TNF but on IL-1 (personal communication). An intriguing discussion element is provided by the observation that small immune

complexes tend to trigger RA synovial macrophages through FcγIII receptors, resulting in TNF production and scant production of IL-1α [43]. Unfortunately, IL-1β was not analysed in this study. Comparative studies with different subsets of immune complexes and macrophages with different Fc receptor compositions are needed.

T-cell-driven arthritis

The classic model of adjuvant arthritis (AA) in rats is considered to be a pure T cell model, because the disease can be easily transferred with T cells and T cell immunomodulation greatly affects the course of AA. Recent studies have clearly identified both TNF and IL-1 as crucial elements in this model, because synergism was evident in combined treatment with TNF soluble receptors and poly(ethylene glycol)-treated IL-1ra [44,45]. Although there is no doubt that TNF is an important cytokine in T cell maturation, blocking of IL-1 has added value. This is found not only in AA but also in the T-cell-dependent exacerbations of smouldering SCW arthritis or AIA, upon rechallenge with antigen [33,38,46].

Final remarks on animal models

Major conclusions are summarized in Table 1, and details can be found in the references cited there. It is evident that IL-1 is not necessarily a dominant cytokine in the acute, inflammatory stages of most arthritis models but has a crucial role in the propagation of joint inflammation and concomitant cartilage and bone erosion in all models. IL-1α seems to act in acute stages, but IL-1β is the crucial cytokine in advanced stages [9], as underlined by the total absence of chronic erosive arthritis in IL-1β-deficient mice. The fact that the chronic, destructive stage is dependent on IL-1 and not on TNF proves indirectly that TNF-independent IL-1 production occurs under the experimental model conditions listed. This holds not only for arthritis but also for similar conditions such as encephalomyelitis [47]. If elements of the models apply to the arthritic process in subsets of RA patients, it is necessary to block IL-1β in addition to TNF.

Further support for the crucial role of IL-1 emerged from the recent demonstration of spontaneous arthritis in Balb/c mice, which are deficient in IL-1ra [48]. The occurrence of spontaneous arthritis, when the IL-1ra deficiency was backcrossed to a Balb/c genetic background, illustrates the continued arthritogenic pressure of environmental stimuli, resulting in the generation of IL-1, which is normally controlled by endogenous IL-1ra.

Remarkably, synovial cell density in chronic SCW arthritis is even more pronounced in TNF-deficient mice than in normal mice, suggesting a homeostatic role of TNF in the control of synovial cell survival. Similar observations have been made in infection models [49]. In synovial cell cultures it is clear that TNF can activate cells through the nuclear factor-κB (NF-κB) pathway but can also contribute to apotosis. This might imply that full TNF blockade should be avoided in therapeutic approaches. An elegant therapeutic alternative is to block the NF-κB pathway [50], still maintaining the TNF-induced regression of cell growth. Apart from TNF, Fas is involved in cell death and TNF interferes with Fas-mediated apoptosis [51], complicating the net outcome of TNF blockade. Recent studies identified a role of TNF-related apoptosis-inducing ligand (TRAIL) in the suppression of autoimmune inflammation [52]. It remains to be seen to what extent the various pathways are touched by anti-TNF therapy. In that sense, the inhibition of IL-1 seems safer because IL-1 is not required for normal homeostasis [53].

For further reading on animal models the review by Klareskog is recommended [54], in which elements of the possible absence of specific immune reactions in RA are discussed. It has been shown that impaired on/off regulation of TNF biosynthesis in mice induces pathologies in joints [55], but evidence that similar disregulation occurs in RA patients is still lacking. An interesting approach to obtaining further insight into possible stimuli or derangements in the RA process is the analysis of signalling pathways turned on in RA synovial cells, to see whether this fits with pathways of known stimuli [56]. However, a serious problem encountered is the fact that pathways are dependent on the maturation stage and receptor expressions of cells; the isolation and subsequent culturing of cells skews their characteristics. Differential gene expression analysis on freshly isolated pieces of synovial tissue might offer an alternative, although first studies in RA synovial tissue have already identified substantial heterogeneity.

Cartilage erosion

Destruction of articular cartilage is caused by the combination of inhibited synthetic activity of the articular chondrocytes and enzymic breakdown of the matrix. The latter can be elicited by enzymes released from chondrocytes and/or the inflamed synovial tissue, in particular at sites of so-called pannus overgrowth of the cartilage. Transfer studies in SCID (severe combined immunodeficiency) mice demonstrated the invasive and destructive character of RA synovial cells, and gene transfer studies with cytokine inhibitors identified that TNF was marginally involved in invasion, yet IL-1 was crucial in chondrocyte activation and matrix destruction [57,58].

Early cartilage damage is characterized by a loss of proteoglycans, which in principle is a reversible process. A major step in erosive tissue loss is the destruction of collagen bundles. Intriguingly, IL-1 is very potent in inducing the suppression of matrix synthesis by the chondrocytes. It also induces the release of active aggrecanase, which is the dominant enzyme responsible for proteoglycan loss. In contrast, IL-1 induces the release of latent forms of matrix metalloproteinases (MMPs), including stromelysin (MMP-3) and collagenase (MMP-13). The latter is crucial in collagen

breakdown; stromelysin seems to be pivotal in the activation of collagenase [59–61]. IL-1 alone gives limited cartilage erosions, linked to moderate autoactivation of MMPs.

Elements that might enhance the erosive character are cytokine autoinduction in the chondrocytes, with autocrine and paracrine effects on neighbouring chondrocytes, the upregulation of cytokine receptors on these cells, the presence of immune complexes in the cartilage surface, and T-cell-derived IL-17 (Fig. 2). IL-1 and TNF production by arthritic chondrocytes has been demonstrated, and TNF receptor upregulation is evident in osteoarthritic cartilage [62,63]. In the presence of immune complexes in the joint, IL-1-induced latent MMPs become broadly activated and cause major tissue erosion [64]. Fc receptor binding on leucocytes and/or chondrocytes and the release of activating enzymes are crucial elements in this activation step mediated by immune complexes. Cartilage erosion is absent in antigen-induced arthritis elicited in mice deficient in Fc receptor, despite florid joint inflammation [65]. In addition, IL-1ra treatment prevents erosions and MMP activity in this model, with limited suppression of acute joint inflammation [60]. These findings identify IL-1 as a pivotal initiating step in erosive processes and underline the role of immune complexes in the exaggeration of destruction.

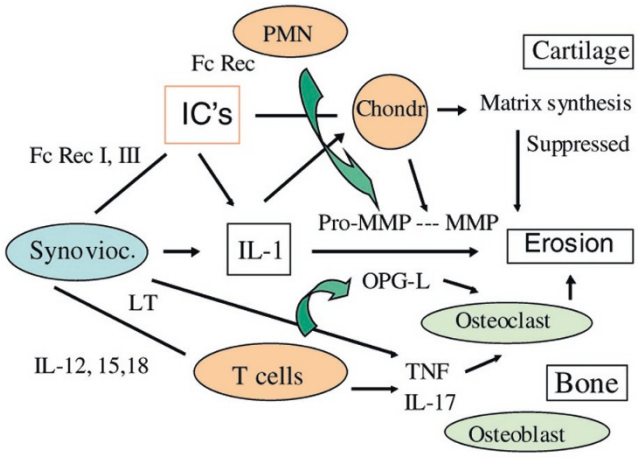
Rheumatoid factor positivity is correlated with more severe and destructive forms of RA, which might fit with the above concept. Apart from immune complexes, T-cell-derived factors might enhance cartilage damage. Interferon-γ has been shown to synergize with TNF and IL-1 in chondrocyte activation [66]; more recently, IL-17 showed direct chondrocyte-mediated cartilage damage, in common with IL-1 [67,68]. Interestingly, IL-17 levels are high in RA synovial fluid [68].

Bone erosion

Apart from cartilage damage, chronic arthritis is characterized by erosions of the underlying bone. The recently identified osteoprotegerin ligand (OPG-L) seems to be a crucial mediator in this process [69–71], because bone loss was absent in OPG-L deficient mice. OPG-L is the pivotal mediator of osteoclast differentiation and activation; it triggers the cells through RANK (receptor activator of NF-κB). Its production can be stimulated by a range of cytokines. IL-1 is the most potent activator and is also a dominant factor in osteoclast differentiation. TNF and IL-17 are less potent but display considerable synergy in osteoclastic bone resorption [72], suggesting that TNF is a crucial factor, along with IL-17, at sites of activated T cells. It remains to be identified whether OPG-L is a good target for the prevention of bone erosion or whether it is safer to target the inducing and modulating cytokines.

An obvious side effect of the direct blocking of OPG-L is unwanted interference with normal turnover of bone. Interestingly, it is claimed that TNF can stimulate osteoclast differentiation by a mechanism independent of RANK [73], which might fit with a direct anti-erosive effect of anti-TNF treatment in RA patients. Apart from effects on osteoclasts, it is evident that the cytokines IL-1, TNF and IL-17 have an effect on osteoblasts, potentially complicating the net effect of anti-cytokine treatment on bone production and resorption.

Figure 2



Amplifying elements in erosive processes. Immune complexes (IC's) generate high levels of IL-1 and, through Fc interaction, also provide additional mediators to activate pro-metalloproteinases (MMPs). T cells might be involved in enhanced bone erosion through TNF, IL-17 and the direct production of osteoprotegerin ligand (OPG-L). T cells come close to the bone at erosion sites. IL-17 also promotes cartilage erosion; a role for OPG-L in this remains to be determined. Fc Rec, Fc receptor; LT, lymphotoxin; PMN, polymorphonuclear cell.

Conclusions

This review has focused on TNF and IL-1, in line with a key role for these cytokines and the therapeutic applicability of specific inhibitors. It is necessary to block IL-1β in addition to TNF. Apart from suppression of arthritogenic mediators, an alternative approach is to apply modulatory cytokines such as IL-4 and IL-10. In addition, IL-6, IL-12 and probably also IL-15 and IL-18 [74–76] might prove to be valuable targets in chronic destructive arthritis. A detailed discussion is beyond the scope of this review.

References

1. Van de Loo AAJ, van den Berg WB: **Effects of murine recombinant IL-1 on synovial joints in mice: measurement of patellar cartilage metabolism and joint inflammation.** *Ann Rheum Dis* 1990, **49**:238–245.
2. Henderson B, Pettipher ER: **Arthritogenic actions of recombinant IL-1 and TNF in the rabbit: evidence for synergistic interactions between cytokines in vivo.** *Clin Exp Immunol* 1989, **75**: 306–310.
3. Keffer J, Probert L, Cazlaris H, Georgopoulos S, Kaslaris E, Kioussis D, Kollias G: **Transgenic mice expressing human tumor necrosis factor: a predictive genetic model of arthritis.** *EMBO J* 1991, **13**:4025–4031.

4. Ghivazzani SC, Kang R, Georgescu HI, Lechman ER, Jaffurs D, Engle JM, Watkins SC, Tindal MH, Suchanek MK, McKenzie LR, Evans CH, Robbins PD: **Constitutive intraarticular expression of human IL-1β following gene transfer to rabbit synovium produces all major pathologies of human RA.** *J Immunol* 1997, **159**:3604–3612.
5. Probert L, Plows D, Kontogeorgos G, Kollias G: **The type I IL-1 receptor acts in series with TNF1 to induce arthritis in TNF1 transgenic mice.** *Eur J Immunol* 1995, **25**:1794–1797.
6. Williams RO, Feldmann M, Maini RN: **Anti-TNF ameliorates joint disease in murine collagen-induced arthritis.** *Proc Nat Acad Sci USA* 1992, **89**:9784–9788.
7. Wooley PH, Dutcher J, Widmer MB, Gillis S: **Influence of recombinant human soluble TNF receptor FC fusion protein type II collagen-induced arthritis in mice.** *J Immunol* 1993, **151**:6602–6607.
8. Van den Berg WB, Joosten LAB, Helsen MMA, van de Loo AAJ: **Amelioration of established murine collagen induced arthritis with anti-IL-1 treatment.** *Clin Exp Immunol* 1994, **95**:237–243.
9. Joosten LAB, Helsen MMA, van de Loo FAJ, van den Berg WB: **Anticytokine treatment of established type II collagen-induced arthritis in DBA/1 mice: a comparative study using anti-TNFα, anti-IL-1Ɪ/β, and IL-1ra.** *Arthritis Rheum* 1996, **39**:797–809.
10. Joosten LAB, Helsen MMA, Saxne T, van de Loo FAJ, Heinegard D, van den Berg WB: **IL-1Ɪβ blockade prevents cartilage and bone destruction in murine type II collagen-induced arthritis, whereas TNF-α blockade only ameliorates joint inflammation.** *J Immunol* 1999, **163**:5049–5055.
11. Mori L, Iselin S, De Libero G, Lesslauer W: **Attenuation of collagen induced arthritis in 55 kDa TNF receptor type I (TNFR)-IgG1 treated and TNFR1 deficient mice.** *J Immunol* 1996, **157**:3178–3182.
12. Maini RN, Taylor PC: **Anti-cytokine therapy for rheumatoid arthritis.** *Annu Rev Med* 2000, **51**:207–229.
13. Charles P, Elliott MJ, Davis D, Potter A, Kalden JR, Antoni C, Breedveld FC, Smolen JS, Eberl G, de Woody K, Feldmann M, Maini RN: **Regulation of cytokines, cytokine inhibitors, and acute-phase proteins following anti TNFα therapy in RA.** *J Immunol* 1999, **163**:1521–1528.
14. Edwards CK III: **PEGylated recombinant human soluble TNF receptor type I: novel high affinity TNF receptor designed for chronic inflammatory diseases.** *Ann Rheum Dis* 1999, **58(suppl 1)**:173–181.
15. Fox DA: **Cytokine blockade as a new strategy to treat rheumatoid arthritis: inhibition of TNF.** *Arch Intern Med* 2000, **160**: 437–444.
16. Garison L, McDonnell ND: **Etanercept: therapeutic use in patients with rheumatoid arthritis.** *Ann Rheum Dis* 1999, **58 (suppl 1)**:165–169.
17. Kavanaugh A, St Clair EW, McCune WJ, Braakman T, Lipsky P: **Chimeric anti-TNF-α monoclonal antibody treatment of patients with rheumatoid arthritis receiving methotrexate therapy.** *J Rheumatol* 2000, **27**:841–850.
18. Lorenz HM, Grunke M, Hieronymus T, Antoni C, Nusslein H, Schaible TF, Manger B, Kalden JR: **In vivo blockade of TNFα in patients with rheumatoid arthritis: longterm effects after repeated infusion of chimeric monoclonal antibody cA2.** *J Rheumatol* 2000, **27**:304–310.
19. Moreland LW, McCabe DP, Caldwell JR, Sack M, Weisman M, Henry G, Seely JE, Martin SW, Yee CL, Bendele AM, Frazier JL, Kohno T, Cosenza ME, Lyons SA, Dayer JM, Cohen AM, Edwards CK III: **Phase I/II trial of recombinant methionyl human necrosis factor binding protein PEGylated dimer in patients with active refractory rheumatoid arthritis.** *J Rheumatol* 2000, **27**:601–609.
20. Drevlov BE, Lovis R, Haag MA, Sinacore JM, Jacobs C, Blosche C, Landay A, Moreland LW, Pope RM: **Recombinant human IL-1 receptor type I in the treatment of patients with active RA.** *Arthritis Rheum.* 1996, **39**:257–265.
21. Bresnihan B, Alvaro-Garcia JM, Cobby M, Doherty M, Domljan Z, Emery P, Nuki G, Pavelka K, Rau R, Rozman B, Watt I, Williams B, Aitchison R, McCabe D, Musikic P: **Treatment of rheumatoid arthritis with recombinant human IL-1ra.** *Arthritis Rheum* 1998, **41**:2196–2204.
22. Van den Berg WB, Bresnihan B: **Pathogenesis of joint damage in RA: evidence of a dominant role for IL-1.** *Bailliere's Clin Rheumatol* 1999, **13**:577–597.
23. Bakker AC, Joosten LAB, Arntz OJ, Helsen MMA, Bendele A, van de Loo FAJ, van den Berg WB: **Prevention of murine collagen-induced arthritis in the knee and ipsilateral paw by local expression of human IL-1ra protein in the knee.** *Arthritis Rheum* 1997, **40**:893–900.
24. Brennan FM, Chantry D, Jackson A, Maini RN, Feldmann M: **Inhibitory effect of TNF1 antibodies on synovial cell IL-1 production in rheumatoid arthritis.** *Lancet* 1989, **ii**:244–247.
25. Deleuran B: **Cytokines in RA: localization in arthritic joint tissue and regulation in vitro.** *Scand J Rheumatol* 1996, **25(S104)**:1–38.
26. Kirkham B, Portek I, Lee CS, Stavros B, Lenarczyk A, Lassere M, Edmonds J: **Intraarticular variability of synovial membrane histology, immunohistology, and cytokine mRNA expression in patients with rheumatoid arthritis.** *J Rheumatol* 1999, **25**:777–784.
27. Ulfgren AK, Grondal L, Lindblad S, Khademi M, Johnell O, Klareskog L, Andersson U: **Interindividual and intra-articular variation of proinflammatory cytokines in patients with rheumatoid arthritis: potential implications for treatment.** *Ann Rheum Dis* 2000, **59**:439–447.
28. Barrera P, Joosten LAB, den Broeder A, van de Putte LBA, van Riel PLCM, van den Berg WB: **Effect of therapy with a fully human anti-TNF monoclonal antibody on the local and systemic homeostasis of IL-1 and TNF in patients with rheumatoid arthritis.** *Ann Rheum Dis* 2000, in press.
29. Alsalameh S, Winter K, Al-ward R, Wendler J, Kalden JR, Kinne RW: **Distribution of TNFα, TNF-R55 and TNF-R75 in the Rheumatoid synovial membrane: TNF receptors are localized preferentially in the lining layer; TNFα is distributed mainly in the vicinity of TNF receptors in the deeper layers.** *Scand J Immunol* 1999, **49**:278–285.
30. Dolhain RJ, Tak PP, Dijkmans BA, de Kuiper P, Breedveld FC, Miltenburg AM: **MTX treatment reduces inflammatory cell numbers and expression of monokines and adhesion molecules in synovial tissue of patients with RA.** *Br J Rheumatol* 1998, **37**:502–508.
31. Chabaud M, Durand JM, Buchs N, Miossec P: **Human Il-17 A T cell derived proinflammatory cytokine produced by the rheumatoid synovium.** *Arthritis Rheum* 1999, **42**:963–970.
32. Kuiper S, Joosten LAB, Bendele AM, Edwards CK III, Arntz OJ, Helsen MMA, van de Loo FAJ, van den Berg WB: **Different roles of TNFα and IL-1 in murine streptococcal cell wall arthritis.** *Cytokine* 1998, **10**:690–702.
33. Van den Berg WB, Joosten LAB, Kollias G, van de Loo FAJ: **Role of TNFα in experimental arthritis: separate activity of IL-1β in chronicity and cartilage destruction.** *Ann Rheum Dis* 1999, **58 (Suppl I)**:S140–S148.
34. Suen WE, Bergman CM, Hjelstrom P, Ruddle NH: **A critical role for lymphotoxin in experimental allergic encephalomyelitis.** *J Exp Med* 1997, **186**:1233–1240.
35. Van de Loo AAJ, Joosten LAB, van Lent PLEM, Arntz OJ, van den Berg WB: **Role of interleukin-1, tumor necrosis factor Ɪ and interleukin-6 in cartilage proteoglycan metabolism and destruction. Effect of *in situ* cytokine blocking in murine antigen- and zymosan-induced arthritis.** *Arthritis Rheum* 1995, **38**:164–172.
36. Wooley PH, Whalen JD, Chapman DL, Berger AE, Richard RA, Aspar DG, Staite ND: **The effect of IL-1ra protein on type II collagen-induced arthritis and antigen-induced arthritis in mice.** *Arthritis Rheum* 1993, **36**:1305–1314.
37. Ghivazzani SC, Lechman ER, Kang R, Tio C, Kolls J, Evans CH, Robbins PD: **Direct adenovirus-mediated gene transfer of IL-1 and TNFα soluble receptors to rabbit knees with experimental arthritis has local and distal anti-arthritic effects.** *Proc Natl Acad Sci USA* 1998, **95**:4613–4618.
38. Van de Loo AAJ, Arntz OJ, Bakker AC, van Lent PLEM, MJM Jacobs, van den Berg WB: **Role of interleukin-1 in antigen-induced exacerbations of murine arthritis.** *Am J Pathol* 1995, **146**:239–249.
39. Ma YS, Thornton GP, Boivin D, Hirsch R, Hirsch E: **Altered susceptibility to collagen induced arthritis in transgenic mice with aberrant expression of IL-1ra.** *Arthritis Rheum* 1998, **41**:1798–1805.
40. Van Lent PLEM, van de Loo FAJ, Holthuysen AEM, van den Bersselaar LAM, Vermeer H, van den Berg WB: **Major role for IL-1 but not for TNF in early cartilage damage in immune complex arthritis in mice.** *Rheumatol* 1995, **22**:2250–2258.

41. Korganov AS, Ji H, Mangialaio S, Duchatelle V, Pelanda R, Martin T, Degott C, Kikutani H, Rajewsky K, Pasquali JL, Benoist C, Mathis D: **From systemic T cell self-reactivity to organ-specific autoimmune disease via immunoglobulins.** *Immunity* 1999, **10**:451–461.
42. Matsumoto I, Staub A, Benoist C, Mathis D: **Arthritis evoked by linked T and B cell recognition of a glycolytic enzyme.** *Science* 1999, **286**:1732–1735.
43. Abrahams VM, Cambridge G, Lydyard PM, Edwards JCW: **Induction of TNF-α production by adhered human monocytes.** *Arthritis Rheum* 2000, **43**:608–616.
44. Bendele AM, McComb J, Gould T, Frazier J, Chlipala E, Seely J, Kieft G, Edwards CK III: **Effects of PEGylated soluble TNF receptor type I alone and in combination with methotrexate in adjuvant arthritic rats.** *Clin Exp Rheumatol* 1999, **17**:553–560.
45. Bendele AM, Chlipala ES, Rich WR, Edwards CK III: **Combination benefit of treatment with recombinant human IL-1ra and pegylated rec human soluble TNF R type I in animal models of RA.** *Arthritis Rheum* 1999, **42**: S171
46. Schwab JH, Anderle SK, Brown RR, Dalldorf FG, Thompson RC: **Pro- and anti-inflammatory roles of IL-1 in recurrence of bacterial cell wall-induced arthritis in rats.** *Infect Immun* 1991, **59**: 4436–4442.
47. Schiffenbauer J, Streit WJ, Butfiloski E, LaBow M, Edwards III C, Moldawer LL: **The induction of EAE is only partially dependent on TNF receptor signaling but requires the IL-1 type I receptor.** *Clin Immunol* 2000, **2**:117–123.
48. Horai R, Saijo S, Tanioka H, Nakae S, Sudo K, Okahara A, Ikuse T, Asano M, Iwakura Y: **Development of chronic inflammatory arthropathy resembling rheumatoid arthritis in IL-1 receptor antagonist-deficient mice.** *J Exp Med* 2000, **191**:313–320.
49. Marino MW, Dunn A, Grail D, Inglese M, Noguchi Y, Richards E, Jungbluth A, Wada H, Moore M, Williamson B, Basu S, Old L: **Characterization of TNF-deficient mice.** *Proc Natl Acad Sci USA* 1997, **94**:8093–8098.
50. Zhang HG, Huang N, Liu D, Bilbao L, Zhang X, Yang P, Zhou T, Curiel DT, Mountz JD: **Gene therapy that inhibits nuclear translocation of nuclear factor κB results in TNF-α-induced apoptosis of human synovial fibroblasts.** *Arthritis Rheum* 2000, **43**:1094–1105.
51. Ohshima S, Mima T, Sasai M, Nishioka K, Shimizu M, Murata N, Yoshikawa H, Nakanishi K, Suemura M, McCloskey RV, Kishimoto T, Saeki Y: **TNFα interferes with Fas-mediated apoptotic cell death on rheumatoid arthritis synovial cells: a positive mechanism of rheumatoid synovial hyperplasia and a clinical benefit of anti-TNFα therapy for RA.** *Cytokine* 2000, **12**:281–288.
52. Song K, Chen Y, Goke R, Wilmen A, Seidel C, Goke A, Hilliard B: **TNF-related apoptosis-inducing ligand (TRAIL) is an inhibitor of autoimmune inflammation and cell cycle progression.** *J Exp Med* 2000, **191**:1095–1104.
53. Labow M, Shuster D, Zetterstrom M, Nunes P, Terry R, Cullinan EB, Bartfai T, Solorzano C, Moldawer LL, Chizzonite R, McIntyre KW: **Absence of IL-1 signaling and reduced inflammatory response in IL-1 type I receptor-deficient mice.** *J Immunol* 1997, **159**:2452–2461.
54. Klareskog L, McDevitt H: **Rheumatoid arthritis and its animal models: the role of TNFα and the possible absence of specific immune reactions.** *Curr Opin Immunol* 1999, **11**:657–662.
55. Kontoyiannis D, Pasparakis M, Pizarro TT, Cominelli F, Kollias G: **Impaired on/off regulation of TNF biosynthesis in mice lacking TNF AU-rich elements: implications for joint and gut-associated immunopathologies.** *Immunity* 1999, **10**:387–398.
56. Bondeson J, Browne KA, Brennan FM, Foxwell BM, Feldmann M: **Selective regulation of cytokine induction by adenoviral gene transfer of IκBα into human macrophages: lipopolysaccharide-induced, but not zymosan-induced, proinflammatory cytokines are inhibited, but Il-10 is nuclear factor-κB independent.** *J Immunol* 1999, **162**:2939–2945.
57. Müller-Ladner U, Evans CH, Franklin BN, Roberts CR, Gay RE, Robbins PD, Gay S: **Gene transfer of cytokine inhibitors into human synovial fibroblasts in the SCID mouse model.** *Arthritis Rheum* 1999, **42**:490–497.
58. Muller-Ladner U, Roberts CR, Franklin BN, Gay RE, Robbins PD, Evans CH, Gay S: **Human IL-1ra gene transfer into human synovial fibroblasts is chondroprotective.** *J Immunol* 1997, **158**: 3492–3498.
59. Van Meurs JBJ, van Lent PLEM, Stoop R, Holthuysen AEM, Singer II, Bayne EK, Mudgett JS, Poole R, Billinghurst C, van der Kraan PM, Buma P, van den Berg WB: **Cleavage of aggrecan at Asn341-Phe342 site coincides with the initiation of collagen damage in murine antigen-induced arthritis: a pivotal role for stromelysin-1 in MMP activity.** *Arthritis Rheum* 1999, **10**:2074–2084.
60. Van Meurs JBJ, van Lent PLEM, Singer II, Bayne EK, van de Loo FAJ, van den Berg WB: **IL-1ra prevents expression of the metalloproteinase-generated neoepitope VDIPEN in antigen-induced arthritis.** *Arthritis Rheum* 1998, **41**:647–656.
61. Van Meurs JBJ, van Lent PLEM, Holthuysen AEM, Singer II, Bayne EK, van den Berg WB: **Kinetics of aggrecanase and metalloproteinase induced neoepitopes in various stages of cartilage destruction in murine arthritis.** *Arthritis Rheum* 1999, **42**: 1128–1139.
62. Alsalameh S, Mattka B, Al-Ward R, Lorenz HM, Manger B, Pfizenmaier K, Grell M, Kalden JR: **Preferential expression of tumor necrosis factor receptor 55 (TNF-R55) on human articular chondrocytes: selective transcriptional upregulation of TNF-R75 by proinflammatory cytokines interleukin 1β, tumor necrosis factor-α, and fibroblast growth factor.** *J Rheumatol* 1999, **26**:645–653.
63. Li J, Sarosi I, Yan XQ, Morony S, Capparelli C, Tan HL, McCabe S, Elliott R, Scully S, Van G, Kaufman S, Juan SC, Sun Y, Tarpley J, Martin L, Christensen K, McCabe J, Kostenuik P, Westacott CI, Barakat AF, Wood L, Perry MJ, Neison P, Bisbinas I, Armstrong L, Millar AB, Elson CJ: **TNF-α can contribute to focal loss of cartilage in osteoarthritis.** *Osteoarthritis Cartilage* 2000, **8**:213–221.
64. Van Meurs J, van Lent P, Holthuysen A, Lambrou D, Bayne E, Singer I, van den Berg WB: **Active matrix metalloproteinases are present in cartilage during immune complex-mediated arthritis: a pivotal role for stromelysin-1 in cartilage destruction.** *J Immunol* 1999, **163**:5633–5639.
65. Van Lent PLEM, van Vuuren AJ, Blom AB, Holthuysen AEM, van de Putte LBA, van de Winkel JGJ, van den Berg WB: **Role of Fc receptor K chain in inflammation and cartilage damage during experimental antigen-induced arthritis.** *Arthritis Rheum* 2000, **43**:740–752.
66. Dodge GR, Diaz A, Sanz-Rodriguez C, Reginato AM, Jimenez SA: **Effects of IFNγ and TNFα on the expression of the genes encoding aggrecan, biglycan, and decorin core proteins in cultured human chondrocytes.** *Arthritis Rheum* 1998, **41**:274–283
67. Lubberts E, Joosten LAB, van de Loo FAJ, van de Bersselaar LAM, van den Berg WB: **Reduction of IL-17 induced inhibition of chondrocyte proteoglycan synthesis in intact murine articular cartilage by IL-4.** *Arthritis Rheum* 2000, **43**:1300–1306.
68. Ziolkowska M, Koc A, Luszczykiewicz G, Ksiezopolska-Pietrzak K, Klimczak E, Chwalinska-Sadowska H, Maslinski W: **High levels of IL-17 in rheumatoid patients: IL-15 triggers in vitro IL-17 production via cyclosporin A-sensitive mechanism.** *J Immunol* 2000, **164**:2832–2888.
69. Hsu H, Lacey DL, Dunstan CR, Solovyev I, Colombero A, Timms E, Tan HL, Elliott G, Kelley MJ, Sarosi I, Wang L, Xia XZ, Elliott R, Chiu L, Black S, Capparelli C, Morony S, Shimamoto G, Bass MB, Boyle WJ: **Tumor necrosis factor receptor family member RANK mediates osteoclast differentiation and activation induced by osteoprotegerin ligand.** *Proc Nat Acad Sci USA* 1999, **96**:3540–3545.
70. Suda T, Takahashi N, Udagawa N, Jimi E, Gillespie T, Martin TJ: **Modulation of osteoclast differentiation and function by the new members of the TNF receptor and ligand families.** *Endocrine Rev* 1999, **20**:345–357.
71. Hsu H, Fletcher F, Dunstan CR, Lacey DL, Boyle WJ: **RANK is the intrinsic hematopoietic cell surface receptor that controls osteoclastogenesis and regulation of bone mass and calcium metabolism.** *Proc Natl Acad Sci USA* 2000, **97**:1566–1571.
72. Van Bezooijen RL, Farih-Sips HCM, Papapoulos SE, Löwik CWGM: **Interleukin-17: a new bone acting cytokine in vitro.** *J Bone Miner Res* 1999, **14**:1513–1521.
73. Kobayashi K, Takahashi N, Jimi E, Udagawa N, Takami M, Kotake S, Nakagawa N, Kinosaki M, Yamaguchi K, Shima N, Yasuda H, Morinaga T, Higashio K, Martin TJ, Suda T: **TNFα stimulates osteoclast differentiation by a mechanism independent of the ODF/RANKL–RANK interaction.** *J Exp Med* 2000, **2**:275–285.
74. Van den Berg WB: **Joint inflammation and cartilage destruction may occur uncoupled.** *Springer Semin Immunopathol* 1998, **20**:149–164.

75. McInnes IB, Leung BP, Sturrock RD, Field M, Liew FY: **Interleukin-15 mediates T cell dependent regulation of TNFα production in rheumatoid arthritis.** *Nat Med* 1997, **3**:189–195.
76. Gracie JA, Forsey RJ, Chan WL, Gilmour A, Leung BP, Greer MR, Kennedy K, Carter R, Wei XQ, Xu D, Field M, Foulis A, Liew FY, McInnes IB: **A proinflammatory role for IL-18 in rheumatoid arthritis.** *J Clin Invest* 1999, **104**:1393–1401.