

## Review

# The need for prognosticators in rheumatoid arthritis. Biological and clinical markers: where are we now?

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Published: 29 May 2008

This article is online at <http://arthritis-research.com/content/10/3/208>

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*Arthritis Research & Therapy* 2008, **10**:208 (doi:10.1186/ar2418)

## Abstract

Rheumatoid arthritis is a heterogeneous disease with respect to clinical manifestations, serologic abnormalities, joint damage and functional impairment. Predicting outcome in a reliable way to allow for strategic therapeutic decision-making as well as for prediction of the response to the various therapeutic modalities available today, especially biological agents, would provide means for optimization of care. In the present article, the current information on biological and clinical markers related to disease activity and joint damage as well as for predictive purposes is reviewed. It will be shown that the relationship of many biomarkers with disease characteristics is confounded by factors unrelated to the disease, and that only few biomarkers exist with some predictive value. Moreover, clinical markers appear of equal value as biomarkers for this purpose, although they likewise have limited capacity in these regards. The analysis suggests the search for better markers to predict outcomes and therapeutic responsiveness in rheumatoid arthritis needs to be intensified.

## Introduction: setting the stage

Rheumatoid arthritis (RA) is characterized by many different phenotypes. Joint involvement, although characteristically symmetrical, can range from a monoarticular pattern to a highly polyarticular pattern, and joint damage can span from mild cartilage degradation to progressive erosive disease of juxtarticular bone [1,2]. The course of RA may be cyclic or relentlessly active [3], and extraarticular manifestations such as rheumatoid nodules or vasculitis may be present. Patients may be seronegative or may have many different autoantibodies [4]. Variable combinations of all these characteristics create a broad heterogeneity that is partly manifested by differences in disease outcomes spanning from remission to severe disability and premature mortality [5,6]. When therapeutic targets are tested in clinical trials and are prescribed in

clinical practice, however, RA is still regarded as a single disorder.

## Biomarkers and clinical markers

Disease activity, joint damage and functional impairment form the anchor points of the natural history of RA, and are characterized by a triangular interrelationship (Figure 1). It is well established that continued disease activity leads to joint damage, resulting in reduction of physical functioning – and if damage is progressive, to irreversible disability [7]. For any clinical and biological marker to be useful, therefore, it should reflect one or more of the components of the RA triad.

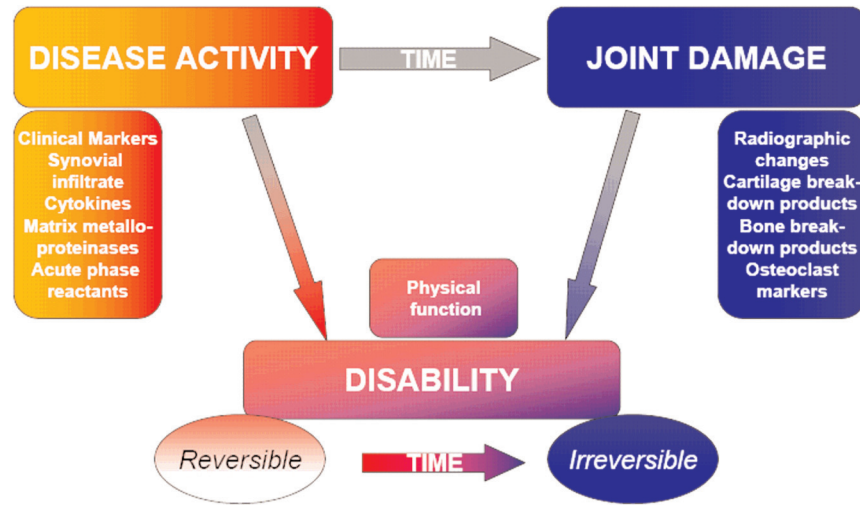
Traditionally, a marker in the present sense should constitute an indicator or a surrogate with diagnostic or prognostic utility [8] (Figure 1). A biological marker, then, would be involved in or would be a consequence of a pathological (or normal) biological process, a product of the organism that is measurable and thus bears the attribute of objectivity.

In the context of rheumatic diseases, a typical biomarker could be a gene or some product of gene expression, an autoantibody, a cytokine, an acute phase reactant, a tissue abnormality possibly visualized immunohistochemically in a synovial biopsy, or a tissue degradation product. The sources of these biomarkers could be the serum, urine, synovial fluid, tissue, cells, and so forth.

In contrast, a clinical marker would constitute a physical variable (sign or symptom), or a clinical judgment or outcome measurement, that emerges as a sequel of the underlying disease process. In rheumatology, this variable may be joint

COL2 = type II collagen; CRP = C-reactive protein; CTX = collagen C-terminal telopeptide; DMARD = disease-modifying antirheumatic drug; IL = interleukin; NTX = collagen N-terminal telopeptide; RA = rheumatoid arthritis; RANKL = receptor activator of NFκB ligand; TNF = tumor necrosis factor.

Figure 1



Triad of rheumatoid arthritis and a selection of markers reflecting its respective elements. The triad of rheumatoid arthritis comprises disease activity–joint damage–disability; a selection of markers that mainly reflect the respective elements of the triad are shown. Over time, the component of disability related to joint destruction will increase and thus disability becomes progressively less reversible; in contrast, with adequate therapy, the component of disability related to disease activity will always be reversible [6,7,98].

counts, global or pain assessments and similar clinical variables, but also composite indices or functional or radiographic scores (reviewed in [9]). The sources of these clinical markers are the patients' manifestations as judged by an assessor or by the patients themselves, and they consequently carry a certain degree of subjectivity – which is less easy to standardize than many laboratory measures.

Any biological marker will therefore have to prove its value in relation to clinical markers, and not necessarily *vice versa*. Nevertheless, to be useful as true surrogate markers of disease, both types of markers have to reflect disease outcome in a broad sense; namely, a 'meaningful endpoint of how a patient feels, functions and survives' [10]. At present, therefore, none of the available biological markers or clinical markers can be employed as surrogate markers, since as such they would have to be useful as substitutes for a clinical outcome. Also, a measure that is useful as a biomarker needs to be validated objectively by demonstration that the laboratory test is accurate, reproducible and measures what it is supposed to. This is not clear for all molecules measured in body fluids that will be mentioned. With these caveats in mind, we will nevertheless use the term biomarker or biological marker in the course of the present review for the biological measurements discussed here, for the sake of simplicity and since this term has been frequently used for these measurements by many experts in the field [11,12].

The present review will address several questions. Can biologic or clinical markers eventually help to subset patients into those who may or may not respond therapeutically to a

given medication? Is there evidence that biomarkers as presumed reflections of pathogenesis are more helpful for subsetting disease and therapeutic prognostication than clinical markers? Are biomarkers more sensitive to change than clinical markers? Do targeted therapies elicit characteristic biomarker signatures in the sense of a proof of concept?

### Biological markers of joint damage

Joint damage in RA is due to changes of cartilage and bone. Radiologically, cartilage changes are reflected by joint space narrowing, while erosions signify bone destruction [13]. Different biomarkers will consequently be representative of these two components of RA joint destruction.

#### Cartilage and bone markers

Cartilage damage – regardless of its cause – will lead to changes in matrix composition. Cartilage matrix is composed of a mesh of type II collagen (COL2), the most abundant cartilage protein. Collagen consists of three chains that form a triple helix, with the exception of the nonhelical N-terminal and C-terminal ends, the telopeptides. In the extracellular matrix, the collagen molecules are linked to one another by cross-linking molecules such as pyridinoline; this cross-linking involves the telopeptides. While the nonhelical parts are degradable by many enzymes, the helical portion can only be degraded by mammalian collagenase at a specific site that yields fragments of one-quarter and three-quarter lengths, respectively. During cartilage degradation, therefore, different collagen fragments – such as crosslinked C-terminal and N-terminal telopeptides – are released (Table 1). Likewise, alpha chain fragments of collagenase degradation are set

**Table 1****Markers of degradation of cartilage and bone**

Tissue	Molecule	Marker of degradation	Designation
Cartilage	Type II collagen	Pyridinoline	PYD
		Type II collagen C-terminal and N-terminal telopeptides	CTX-II, NTX-II
		Type II collagen alpha chain fragments	Col2-3/4 long, Col2-3/4C short
	Aggrecan	Core protein fragments	
		Keratan sulfate fragments	
	Noncollagen and nonaggrecan proteins	Cartilage oligomeric matrix protein	COMP
Bone	Type I collagen	Pyridinoline, deoxypyridinolin	PYD, DPD
		Type I collagen C-terminal and N-terminal telopeptides	CTX-I, NTX-I
	Noncollagenous proteins	Bone sialoprotein	BSP
		Tartrate-resistant acid phosphatase	TRAP

free and can be measured, such as COL2 three-quarter-long collagen via the generated carboxyterminal neopeptide.

Within the COL2 network, other components of the cartilage matrix are interspersed. These components are degraded in the course of cartilage damage. Among these are aggrecan, the breakdown products of which can be detected via core protein, keratan sulfate or chondroitin sulfate epitopes, and cartilage oligomeric matrix protein.

Bone destruction is an important hallmark of RA, and therefore the ability to measure its surrogates is important. The major structural protein of bone is type I collagen, which like COL2 forms triple helical, cross-linked structures and is degraded in a similar manner as COL2. Type I collagen C-terminal telopeptide (CTX-I) and type I collagen N-terminal telopeptide (NTX-I), but also free pyridinoline crosslinks, therefore reflect type I collagen degradation. Just like cartilage, bone contains noncollagenous proteins, such as bone sialoprotein, which are released during bone damage (Table 1).

**Association with joint damage**

Landewe and colleagues have shown a significant correlation between early changes in type II collagen C-terminal telopeptide (CTX-II) and prediction of long-term radiographic progression as a result of therapy; in contrast, changes of CTX-I were not related to joint damage [14,15]. The changes in CTX-II levels, however, were broadly overlapping with baseline values, and were therefore predictive primarily on a group level. Subanalysis of radiographic changes by joint space narrowing and erosion scores did not reveal major differences between these two components of the radiographic score in relation to these biomarkers [14]. This analysis indicates that CTX-II levels may at least partly reflect

the inflammatory response (which leads to collagen degradation via activation of matrix metalloproteinases), although there was no statistical evidence for such relationship. In contrast, MMP3 levels appear significantly associated with progression of joint damage [16,17].

Assessment of COL2 three-quarter-long collagen showed decreasing levels during treatment with methotrexate, and particularly with anti-TNF + methotrexate [16]. Moreover, baseline COL2 three-quarter-long collagen concentrations were significantly associated with cytokine levels [16].

Evaluation of cartilage oligomeric matrix protein levels revealed that these are much higher in patients whose joint damage progresses when compared with those whose radiographic changes do not progress [18]. These observations as well as the CTX-II data were recently confirmed [17], although there was again broad overlap in biomarker levels of patients with and without progression of joint damage. Yet none of these markers performed better than C-reactive protein (CRP), swollen joint counts or composite disease activity indices [17].

Another set of markers potentially useful to assess bone damage relates to osteoclast differentiation and function. Osteoclasts that derive from bone marrow progenitor cells via the monocyte lineage are the cell population responsible for erosive changes [19,20] and are pivotally dependent on the triggering of receptor activator of NFκB. The receptor activator of NFκB ligand (RANKL), a member of the TNF family, is expressed on various cell populations and also exists as a soluble molecule [21]. The ligand's action can be inhibited by osteoprotegerin, a decoy receptor. Both osteoprotegerin and RANKL can be measured in serum, and the

osteoprotegerin/RANKL ratio may be an indicator of osteoclast differentiation and activation. In fact, a low osteoprotegerin/RANKL ratio, reflecting high RANKL activity, appears to be associated with increased progression of radiographic joint damage [22]. Osteoclasts mediate their special function in degrading calcified bone by several mechanisms, including secretion of cathepsin K. Consequently, cathepsin K levels correlate with joint destruction [23] and may be reflective of effective therapy [24].

Despite some interesting, although mostly inconclusive, data in the literature, there are a number of confounding factors related to the usefulness of many of these bone and cartilage breakdown products in helping to subgroup and treat RA. First, comorbidity, such as osteoarthritis and especially generalized osteoporosis [25], may lead to collagen degradation and thus obscure the subtle changes observed. Another factor is that circadian variations in bone and cartilage marker levels have been described (that is, the timing of the measurement is critical) [26,27]. Third, physical activity may change biomarker concentrations significantly [28]. A fourth factor is that renal or hepatic disease may influence circulating and/or urine levels of biomarkers [29]; especially, urine biomarkers should always be related to creatinine levels [11]. Finally, the tissue content and/or levels of certain proteins that constitute useful biomarkers may be genetically determined, and this heritability may have to be accounted for [30]. These factors contribute to the complexity of monitoring cartilage and bone turnover in RA as they relate to predicting joint damage [31]. In an individual patient, however, short-term changes in the course of therapeutic interventions aimed at interfering with the RA process ought to reflect the modification of that process.

Another set of markers that is not derived from cartilage or bone may be predictive of joint damage: autoantibodies. It has long been recognized that radiographic progression of joint destruction is much higher in patients positive for rheumatoid factor when compared with seronegative patients [32-35]. This is particularly true in patients with high-titer rheumatoid factor; that is, rheumatoid factor  $\geq 50$  IU/ml [36]. Autoantibodies to citrullinated proteins have been shown more recently to be predictive for the occurrence of erosions [36,37]; the autoantibodies are broadly overlapping with high-titer rheumatoid factor, and the latter appears to change with effective therapy more than the autoantibodies to citrullinated proteins [35,36,38,39]. These markers of the autoimmune response characteristic of RA surpass the value of most cartilage and bone breakdown products in predicting joint damage. At present, the combination of autoantibody and acute phase protein assessment may constitute the most reliable way to predict severe erosive RA [33,40].

## Biological markers of disease activity

### Cells and cytokines

The clinical manifestations of RA are the consequences of synovial inflammation and the subsequent degradation and

destruction of cartilage and bone. These pathways comprise various cell populations characteristically involved in RA synovitis as well as cytokines and the products of their action.

When looking at arthroscopic biopsies, the composition of the cellular infiltrate is heterogeneous [41,42], and the most consistent reflection of active disease is the presence of high numbers of macrophages in the synovium [43,44]. Potent anti-inflammatory agents such as glucocorticoids lead to a reduction in synovial macrophage cellularity (but little other cellular changes) [45]. Interestingly, blocking other mediators such as the chemokine macrophage chemoattractant protein MCP-1 was not associated with changes of synovial tissue composition or with clinical benefit [39,46]. In addition, the degree of B-cell depletion in synovial tissue in the course of rituximab therapy was not significantly associated with the clinical response and B cells may be present in the synovium even if depleted in peripheral blood [39,47]. This finding also indicates that the periphery often does not reflect the events occurring in the joint, but these by themselves are also not sufficiently predictive.

With the exception of macrophage infiltrates, therefore, the composition and extent of the cellular infiltrate may not be related to clinical manifestations, in line with observations of histologic synovitis in the absence of clinical joint involvement [48]. Likewise, the immunohistochemistry of cytokines may not sufficiently reflect the disease activity of RA. The use of gene expression profiling currently does not appear to provide much additional information in this respect [49,50], although the technique may be useful to detect unforeseen changes. Serum levels of proinflammatory cytokines – including the most abundant one, IL-6 – are not highly correlated with measures of disease activity and progression [51,52]; moreover, baseline IL-6 concentrations may vary almost 100-fold between different individuals, and can increase with exercise [53].

Qualitative and/or quantitative differences exist among individual patients both on the cellular level and the cytokine level [54,55], and pathohistologic analyses have failed to reveal changes that are pathognomonic for RA. Synovial cytokine expression in RA, even at a group level, does not significantly differ qualitatively from other arthritic disorders [56-58]. Nevertheless, there appear to be differences in the quantity of cytokine expression when looking at other inflammatory joint diseases, such as psoriatic arthritis, ankylosing spondylitis or inflammatory osteoarthritis. Patients with these conditions have lower synovial TNF and IL-6 levels than RA patients [58,59]. These differences may have some important bearing given that RA is a rapidly and highly destructive joint disease, while the other disorders are usually slowly or not as destructive.

All these facets reveal the complexity we face when trying to search for good biomarkers. The complexity is further

illustrated by observations that changes found in these markers early in the course of RA may be quite different from those seen in late disease [60], regardless of the level of activity or inflammation in either early or late disease.

To better appreciate the complexity, we present four hypothetical patients in whom different cytokines or different cell populations may appear to predominate, even though in all patients many cells or soluble molecules are at least partly activated (Figure 2).

The resolution of this mosaic will be difficult: changes of biomarker levels in the course of therapy may give helpful insights, but this knowledge is insufficient to provide prediction rules for the employment of specific therapies.

### Acute phase reactants

In contrast to the markers mentioned above, the serum levels of CRP, a molecule induced by proinflammatory cytokines, especially IL-6 [61], not only reflect the extent of disease activity [62,63] but are also cumulatively associated with joint destruction [17,63,64]. Moreover, CRP levels before the start of disease-modifying therapy can predict the degree of subsequent radiographic joint damage, and greater reduction from baseline in CRP levels was associated with less progression of joint damage and higher trough serum concentrations of anti-TNF antibody [65,66]. While other acute phase reactants, such as serum amyloid A protein, also reflect disease activity, CRP determination is widely available and of low cost [67], making it the preferred biomarker of disease activity (and even joint destruction) not yet surpassed by other markers.

The erythrocyte sedimentation rate could be regarded as an alternative to measuring CRP. However, it may be influenced by various other factors not primarily related to inflammation [68]. Successful therapeutic interventions with glucocorticoids and disease-modifying antirheumatic drugs (DMARDs) including biological agents are usually associated with a fall in CRP levels, and are often also paralleled by reductions in IL-6 serum concentrations [24,69,70].

It is somewhat disappointing that levels of other marker molecules, such as various cytokines, cytokine receptors or matrix metalloproteinases, have not been shown to exhibit better correlations with actual disease manifestations or outcomes of RA than acute phase reactants. No biological marker assessed hitherto has therefore been shown to be related better to disease activity and joint damage than CRP.

### Clinical markers

Biological markers have to reflect diagnosis and/or prognosis; that is, clinical outcomes [10,71]. The extent of this association will determine the reliability and value of the marker. As revealed above, with the exception of the acute phase response and autoantibodies, no biological marker is

currently sufficiently reliable to acknowledge its usefulness as a marker of disease activity or joint damage for use in clinical practice. Markers of disease activity, however – especially composite indices using some of the core set variables, such as the Disease Activity Score employing 28 joint counts, the Simplified Disease Activity Index and the Clinical Disease Activity Index (reviewed in [9]) – have been shown useful for following disease activity and for serving as endpoints in clinical trials and observational studies [72,73]. Moreover, disease activity over time as assessed using these indices correlates significantly with progression of joint damage [63].

These data suggest that clinical markers, even in the absence of any laboratory variable such as is the case with the Clinical Disease Activity Index [63], may serve the purpose of predicting joint damage at least as well as any biological marker. In fact, time-averaged disease activity using these composite indices correlated with radiographic progression better than time-averaged CRP [63].

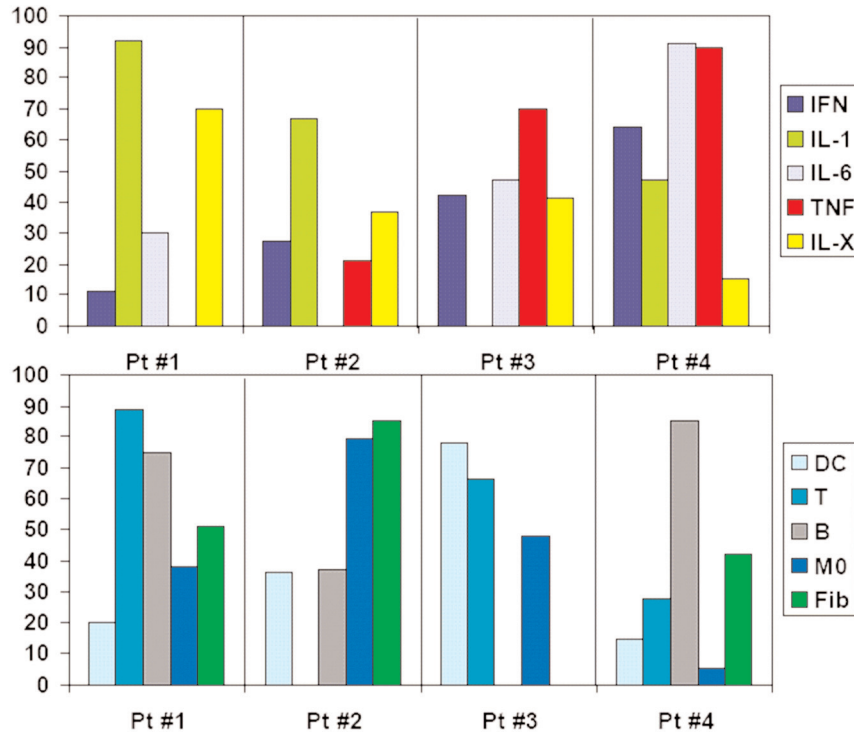
### Prediction of clinical improvement and retardation of joint damage

No biomarker currently allows one to predict the extent of clinical improvement in response to therapy, although the degree of radiographic progression on traditional disease-modifying agents can be foretold by baseline and cumulative levels of CRP, swollen joint counts or overall disease activity using respective composite scores [65].

In this context it needs to be borne in mind that TNF and IL-6 are cytokines that promote osteoclast differentiation and activation [20,21,74-77]. Importantly, under stable low concentrations of RANKL, increasing amounts of proinflammatory cytokines, such as TNF, will lead to increasing osteoclast differentiation [78]. It is therefore conceivable that this relation of TNF levels and osteoclast activation also exists *in vivo*. In support of this notion, treatment of RA patients with a TNF inhibitor plus methotrexate leads to a dissociation of the close relationship between joint damage and the inflammatory response (exemplified by disease activity measures) [79], and these data were meanwhile confirmed with another TNF-blocker [80]. These findings have led to the threshold hypothesis shown in Figure 3. According to this hypothesis, TNF will lead to joint damage especially once its levels exceed a particular threshold that lies above the threshold needed for the activation of the inflammatory response. Blocking of TNF may inhibit its bioactivity fully (Figure 3b), may reduce bioactivity to levels below the putative threshold of destruction with residual signs and symptoms but no destruction (Figure 3c), or may reduce bioactivity to levels above that threshold but still significantly retard joint damage compared with other treatment modalities, without preventing progression in full (Figure 3d). This hypothesis is in line with results from clinical trials where the use of TNF inhibitors significantly retarded or halted joint destruction despite residual active disease [81-83].



Figure 2



Depiction of potential cytokine and cellular patterns in four hypothetical patients with rheumatoid arthritis. Upper panel: hypothetical biological activities of various cytokines. Lower panel: hypothetical biological activities of various cell types. y axis, arbitrary units of activity. For example, in patient #1 TNF does not appear bioactive, while in patient #3 B cells appear uninvolved. Especially in patient #4, however, all cytokines – and in patient #1 all cell types – appear actively engaged in the disease process. In patient #2 IL-6 may not be detectable. It is unclear to what extent which of those cells and/or cytokines is contributing and if one or several targeted therapies would be efficacious. Although such relationships have not yet been elucidated, differences in synovial cellular compositions and cytokine contents have been noted in various studies [55,59,99]. DC, dendritic cells; Fib, fibroblasts.

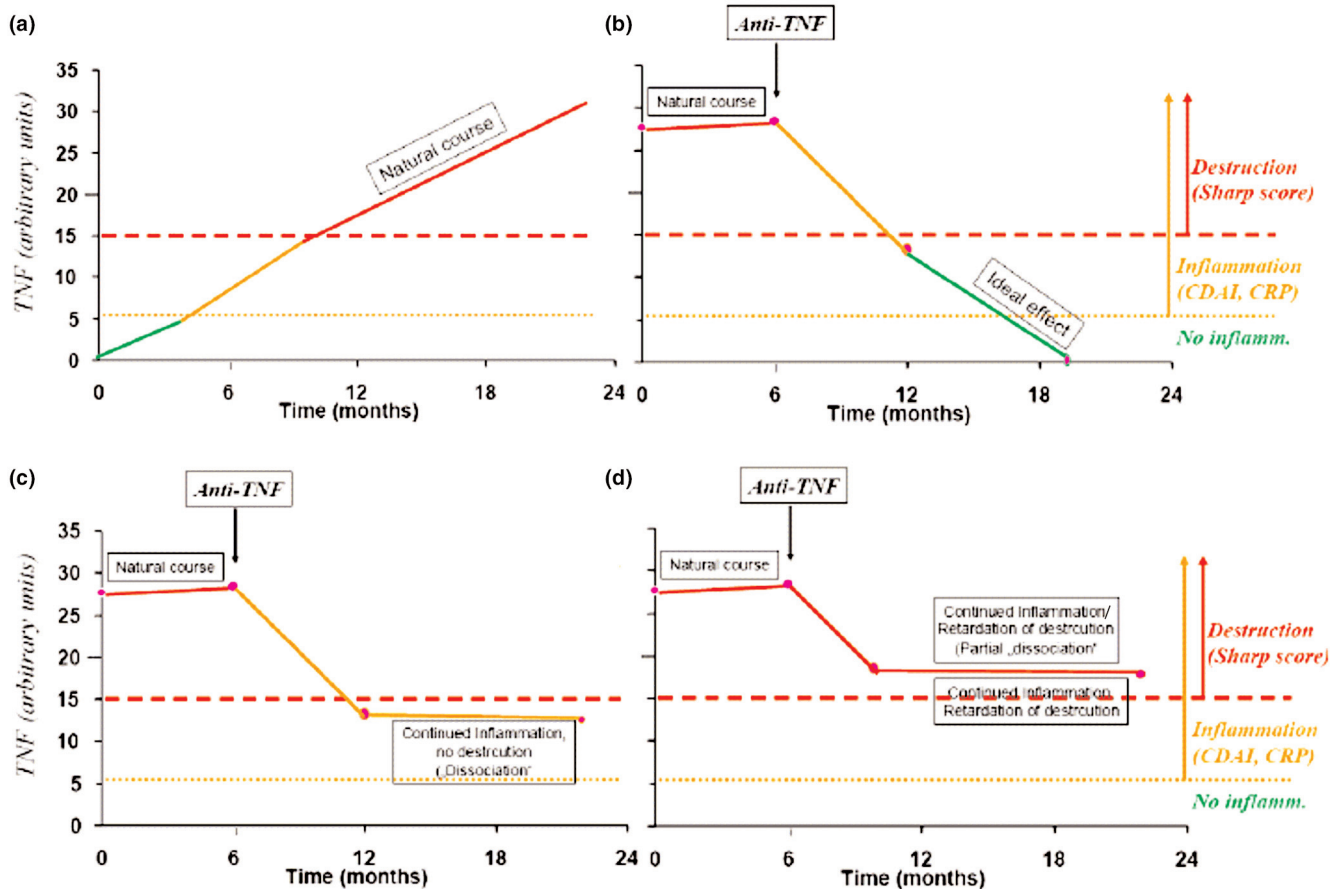
Whilst the focus of attention with respect to joint damage has primarily been the osteoclast, it should be borne in mind that bone repair mechanisms may be deficient in TNF-mediated arthritis and can be induced by osteoblast activation [84]. Biomarkers of osteoblast function, including proteins involved in the Wnt signaling pathways [85], may therefore constitute further interesting markers for the future.

Importantly, however, achieving low disease activity or remission with traditional DMARDs will lead to highly effective reduction of progression of joint damage [86]. Another important aspect in our attempts to predict outcomes is therefore the estimation of the clinical response to treatment.

Recent analyses in a large cohort of clinical trial patients have revealed that baseline disease activity is already somewhat related to disease activity at 1 year of therapy, especially with methotrexate treatment [87]. Irrespective of the type of therapy or disease duration, however, 3 months after initiation of treatment the disease activity – as assessed by the Simplified Disease Activity Index, the Clinical Disease Activity Index or

the Disease Activity Score employing 28 joint counts – was highly correlated with disease activity at the end of the observation period [87] (Figure 4). These data were further validated by studying an observational cohort of RA patients [88]. The probability to attain remission or low disease activity was more than 75% for patients achieving low disease activity by the Simplified Disease Activity Index already after 3 months of treatment, while it amounted to only 25% for patients having high disease activity at that point in time [87].

Needless to say, achieving lower disease activity after 3 months of therapy will also result in better outcomes in the other components of the triad – physical function and joint damage. Moreover, starting effective treatment early, and especially before initial damage has occurred, constitutes the optimal strategy (Figure 5). This optimization, however, requires early referral and early diagnosis [89,90]. Moreover, there is sufficient evidence to allow stating that DMARDs will interfere with the disease process at any point in time and will prevent progression of disease regardless of patient age or disease duration – it is the past damage that has to be carried on.

**Figure 3**

Threshold hypothesis of osteoclast activation. **(a)** Osteoclast activation is assumed to occur only after passing a putative threshold. **(b)** Anti-TNF therapy may ideally lead to total inhibition of bioactive TNF. **(c)** In other patients, anti-TNF therapy may reduce TNF activity below the threshold of osteoclast activation; these patients may continue having signs and symptoms of rheumatoid arthritis. **(d)** In yet another group of patients, the TNF activity may be reduced, but not to a level that goes below the threshold of osteoclast activation; in these patients, there will be more inflammation than in (c) and some residual destruction – in relation to other therapies, such as methotrexate, the destruction will be significantly less at a similar level of inflammatory signs and symptoms of rheumatoid arthritis [79]. CDAI, Clinical Disease Activity Index; CRP, C-reactive protein.

### Complexity of rheumatoid arthritis

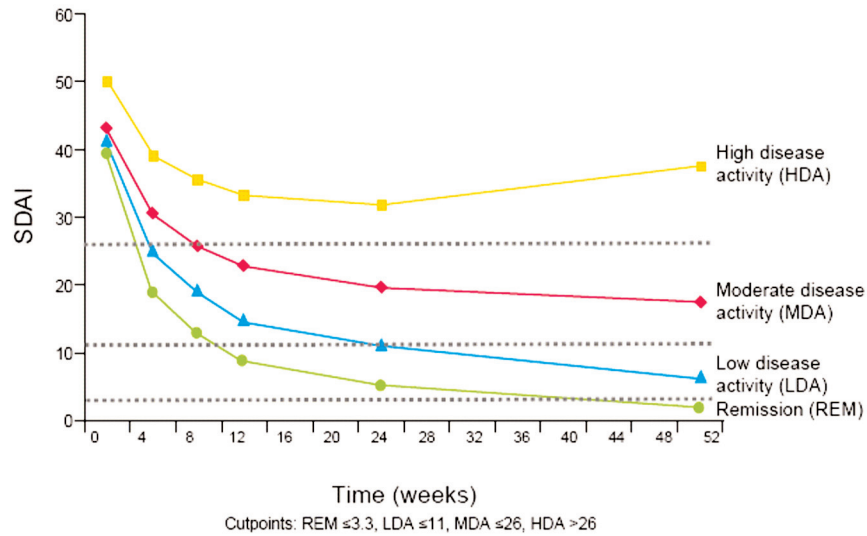
The ability to utilize biologic markers and or clinical markers to predict disease outcomes as well as therapeutic response early in the course of the disease, so as to achieve remission, is our ultimate goal in RA. We have indicated that predicting disease outcome and therapeutic response may be difficult due to the heterogeneity of the disease regarding both its clinical manifestations as well as putative pathogenic characteristics. Contributing to this has been the observed variability of biomarker levels. Data presently reveal that different therapeutic agents, including targeted therapies directed against different molecules, lead to similar therapeutic responses. To which degrees these responsive populations overlap, however, is currently unclear: do most of the patients achieving an American College of Rheumatology 70% response (or remission) to one drug have different characteristics than patients responding to a similar extent to

another agent, or are they overlapping? Answers to these questions are mandatory for institution of proper treatment to gain control of disease and to induce remission.

### Conclusion

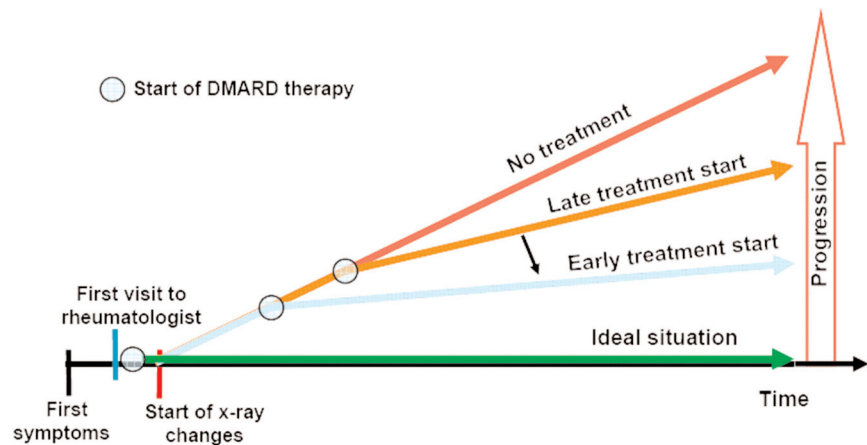
Many biological markers reflect the ongoing disease process of RA. The markers' correlation to the typical manifestations of RA, the signs and symptoms of active disease, the destruction of joints or the impairment of physical function, however, has been poor for most of them. This may be due to a variety of reasons. First, what we measure in various body fluids may not reflect sufficiently well what is ongoing in the microenvironment of the joint, and the leakage of various molecules into the body fluids may differ among, and within, patients. Second, where biomarkers have important pathophysiological functions, the concentrations measured may not reflect their functional fraction. Third, the pathogenesis of RA

Figure 4



Time course of disease activity in patients attaining particular disease activity states after 1 year of therapy. Patients who achieved low disease activity or remission at 1 year attained a low disease activity state within 3 to 6 months from the onset of treatment. SDAI, Simplified Disease Activity Index. Reproduced with permission from [87].

Figure 5



Effect of disease-modifying antirheumatic drug therapy. Disease-modifying antirheumatic drugs (DMARDs) will interfere with the disease process at any time point, and will lead to a deflection of the slope of progression from its natural course. The ideal situation would be to diagnose and treat rheumatoid arthritis early; at best, before damage has occurred.

may be highly heterogeneous, with different markers being preponderate in different patients. Fourth, pathogenetic mechanisms may even vary within a patient in the course of the disease. Finally, diurnal and genetic variations may change biomarker levels and confound our ability to interpret them – with relatively low levels in one patient being highly pathogenic, while a high level in another patient may mean little for that particular patient’s disease.

Importantly, we have focused here on markers that are frequently used to evaluate disease activity, cartilage and bone damage. There is more going on in relation to the immunopathogenic events, however, than mere production of cytokines and the consequences of their activity. For example, certain T-cell subpopulations have shown changes in active RA [91,92]. The application of such markers in practice, however, is limited by the lack of widespread availability of the



respective detection techniques. Also, some imaging techniques, such as magnetic resonance imaging and ultrasound, may allow new insights and may contribute interesting information on disease activity or even outcome [93-95].

More information is presently needed, and the search for the best set of biomarkers for assessment and prediction of disease activity, damage and response to therapy as well as efforts to better standardize biomarker assessment must, and will, go on. Appropriate cohorts of patients and appropriate validation procedures will be needed to this end. For the time being it appears too early to recommend the use of these markers in routine practice or as major outcomes in clinical trials. System biologic approaches may provide better insights in the not too distant future [96], but their applicability in routine settings will constitute yet another challenge. Likewise, proteomic approaches employing various methodological means may prove helpful, but at present merely confirm the complexity of the biological interplay we are dealing with in RA [97].

There are two exceptions, however, which make the above summary much less disappointing: acute phase reactants, especially CRP, are highly reliable markers of disease activity and, in the long term, radiographic outcomes; and autoantibodies, especially rheumatoid factor and autoantibodies to citrullinated proteins, have diagnostic and prognostic value. That these old markers surpass the value of new ones and that some of the old techniques employed for marker determination may be more reliable than new ones sounds inadequate, but nevertheless it is satisfying that such usefulness and validity does exist for at least few molecules.

Equally important, research activities of the past decade have allowed one to obtain clinical assessment tools – the composite disease activity indices, which are not only reliable for assessing the wellbeing of patients with RA during follow up, but are also highly associated with functional and radiological disease outcome. A combination of such tools with a novel approach to biomarker evaluation may therefore allow for optimized understanding and prediction of the fate of the individual RA patient. There is recognition that clinical assessment (measuring and noting the change in index and the disease activity state attained) early in the course of therapy (3 to 6 months after initiation of therapy) allows one to judge longer-term treatment effects and to make rapid changes of therapeutic modalities in the individual patients in whom low disease activity is not achieved. Such a strategy will consequently be associated with lesser costs by avoiding prolonged use of ineffective therapies and will also lead to a better outcome of RA.

### Competing interests

The authors have received honoraria and/or grant support from Abbott, Amgen, BMS, Centocor, Roche, Sanofi-Aventis, Schering-Plough, UCB and Wyeth. There has been no financing of the current manuscript by any of the above.

### Acknowledgements

The authors would like to acknowledge the critical reading and suggestions by Dr Joseph Markenson. This paper was supported by the Center of Musculoskeletal Disorders, Medical University of Vienna.

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