TNF-α is overexpressed in the synovial fluid of patients with RA. Moreover, TNF-α transgenic mice spontaneously develop arthritis. The first biologic disease modifying antirheumatic drugs (bDMARD) generated was infliximab (IFX), a chimeric monoclonal antibody (mAb) to TNF-α. Clinical trials of IFX proved that TNF blockade is highly efficacious in the treatment of RA and led to the development of other TNF inhibitors.

Another cytokine that has been targeted in the treatment of RA is IL-6, a typical cytokine featuring redundancy and pleiotropic activity that plays a key role in the development of RA.[4–6] IL-6 promotes the development of an imbalance between Th17 and regulatory T (Treg) cells and the production of autoantibodies, such as rheumatoid factor and anti-citrullinated peptide antibody. IL-6 also promotes synovial inflammation and cartilage and bone destruction and has systemic effects in cardiovascular, psychological, and skeletal disorders. The first generated bDMARD targeting IL-6 was tocilizumab (TCZ), a humanized anti-IL-6 receptor antibody. Now, other IL-6 inhibitors are also being developed and clinical trials for these agents are in progress.[6] These include fully human anti-IL-6 receptor mAb (REGN88/SAR153191 [sarilumab]), anti-IL-6 receptor nanobody (ALX-0061), anti-IL-6 Abs (CNTO136 [sirukumab], ALD518 [BMS-945429], CDP6038 [olokizumab], and MEDI5117). In this review, we highlight current data regarding the comparative efficacy and safety of TCZ and TNF inhibitors. We also discuss the positions of these agents in the treatment of RA.

## Differential pharmacology of TCZ, adalimumab (ADA), and other TNF inhibitors

Several bDMARDs are currently available for the treatment of moderate to severe active RA, including five TNF inhibitors (IFX, ADA, golimumab [GOL], certolizumab pegol [CEP], and etanercept [ETA]), an IL-6 blocker (TCZ), a T-cell stimulator blocker (abatacept), a B-cell depletory (rituximab), and an IL-1 receptor antagonist (anakinra).[4,6] The characteristic features of TCZ and five TNF inhibitors are shown in Table 1.

TCZ is a humanized IgG1 class anti-IL-6 receptor mAb that was generated by grafting the complementarity determining regions of a mouse antihuman IL-6 receptor antibody (Ab) into human IgG1.[7] TCZ blocks IL-6 mediated signal transduction by inhibiting the binding of IL-6 to both transmembrane and soluble IL-6 receptors. TCZ can be administered intravenously or subcutaneously.

IFX was the first TNF inhibitor developed and it is a chimeric immunoglobulin (Ig) composed of a murine variable region and a human constant region against TNF-α. Due to immunogenicity and response failure issues, IFX is licensed to be used with methotrexate (MTX) by intravenous injection. ADA and GOL are fully human mAbs to TNF-α and can be used subcutaneously every 2 weeks and every 4 weeks, respectively. CEP is a humanized Fab fragment conjugated to polyethylene glycol (PEG). The attachment of PEG prolongs the drug's half-life, whereas the absence of an Fc fragment prevents effector functions such as Ab-dependent cellular cytotoxicity and complement-dependent cellular cytotoxicity, as well as active transfer of CEP across the placenta during pregnancy. CEP is used subcutaneously every 2 weeks.

In contrast to these TNF inhibitors, ETA is a fusion protein consisting of two TNF receptor 2 (also known as p75TNF receptor) extracellular domains and a human Fc fragment of the IgG1 class. As TNF-α and lymphotoxin binds to TNF receptor 2, ETA neutralizes the biological activity of both cytokines. ETA is administered subcutaneously once or twice weekly.

## Comparative efficacy studies of TCZ, ADA, and other TNF inhibitors

### TCZ

The efficacy of TCZ administered alone or in combination with MTX or other synthetic disease modifying antirheumatic

**Table 1** Characteristics of tocilizumab and tumor necrosis factor inhibitors

| | Tocilizumab | Infliximab | Adalimumab | Golimumab | Certolizumab pegol | Etanercept |
|---|---|---|---|---|---|---|
| Target molecule | IL-6R | TNF-α | TNF-α | TNF-α | TNF-α | TNF-α<br>Lymphotoxin |
| Structure | Humanized Ig | Chimeric Ig | Fully human Ig | Fully human Ig | Humanized Fab-pegol | P75TNFR-Fc |
| Injection route | IV, SC | IV | SC | SC | SC | SC |
| Activity | | | | | | |
| ADCC | + | + | + | + | – | + |
| CDCC | + | + | + | + | – | ± |

**Abbreviations:** ADCC, antibody-dependent cellular cytotoxicity; CDCC, complement-dependent cellular cytotoxicity; Ig, immunoglobulin; IL-6R, interleukin-6 receptor; IV, intravenously; SC, subcutaneously; TNF-α, tumor necrosis factor-α; TNFR, TNF receptor.

drugs (sDMARDs) was verified for active RA in seven Phase III trials. The three Phase III trials AMBITION, SAMURAI, and SATORI were designed to examine the efficacy of TCZ monotherapy.[8–10] The AMBITION trial[8] involved active RA patients for whom previous treatment with MTX and TNF inhibitors had not failed. The SAMURAI trial[9] involved patients with an inadequate response to sDMARDs, and the SATORI trial[10] involved patients with an inadequate response to MTX. In all three studies, patients treated with TCZ had superior American College of Rheumatology (ACR) 20 responses and lower disease activity score (DAS) 28 at 24 weeks than controls treated with MTX or other sDMARDs.

Four Phase III trials were performed to evaluate the efficacy of TCZ combination therapy with MTX or another sDMARD. The OPTION trial was designed to evaluate the efficacy of TCZ in combination with MTX, and the results showed that combination therapy is effective for moderate to severe active RA.[11] The TOWARD trial demonstrated that TCZ combined with a sDMARD such as MTX, chloroquine, gold, sulphasalazine, azathioprine, or leflunomide is effective for reducing RA disease activity in patients with an inadequate response to monotherapy with any one of the sDMARDs.[12] The RADIATE trial proved that TCZ plus MTX is effective for achieving rapid and sustained improvements in signs and symptoms in patients whose RA is refractory to TNF inhibitors.[13] Moreover, the LITHE trial, which was designed to evaluate not only disease activity but also structural joint damage, demonstrated that TCZ plus MTX is efficacious at suppressing disease activity.[14] Radiographic evidence from the LITHE trial showed that progression of joint destruction is significantly inhibited after 52 weeks of combination treatment.[14] All of these studies enrolled patients with an inadequate response to all previous treatments, including MTX, TNF inhibitors, or other sDMARDs, and all of the studies showed that TCZ combination therapy is effective for these patient populations.

## ADA

The efficacy and safety of ADA was examined in the ARMADA trial.[15] A total of 271 patients with active RA who had an inadequate response to MTX were randomized to continue MTX in combination with either placebo or ADA (20, 40, or 80 mg subcutaneously every other week). ACR20 responses at week 24 were 47.8, 67.2, and 65.8% in the 20, 40, and 80 mg groups, respectively, whereas the response rate was 14.5% for the placebo group. Subsequently, the PREMIER study, which involved 799 patients with early and aggressive RA who had no previous MTX use, confirmed that ADA plus MTX combination therapy is vastly superior to either MTX alone or ADA alone in improving clinical signs and symptoms, inhibiting radiographic progression of joint destruction, and effecting clinical remission.[16]

## IFX

In the Phase III trial ATTRACT, 428 RA patients with active disease activity and an inadequate response to MTX were randomized to receive MTX with either placebo or IFX (3 mg/kg every 4 weeks, 3 mg/kg every 8 weeks, 10 mg/kg every 4 weeks, or 10 mg/kg every 8 weeks).[17] At week 30, patients in the IFX treated groups achieved an ACR20 response rate of 50%–58%, versus an ACR20 response rate of only 20% in the placebo group. Structural damage was also assessed with the modified van der Heijde-Sharp score at week 102.[18] Compared with the MTX only regimen, erosion and joint space narrowing scores from baseline to week 102 with early RA patients decreased significantly with each of the IFX dose regimens.

## GOL

In the Phase III trial GO-FORWARD, 444 active RA patients who had an inadequate response to MTX were randomly assigned to receive placebo subcutaneous injections plus MTX, GOL 100 mg plus placebo capsules, GOL 50 mg plus MTX, or GOL 100 mg plus MTX.[19] The proportion of patients who achieved an ACR20 response at week 14 was 33.1% in the placebo plus MTX group, 44.4% ($P$=0.059) in the GOL 100 mg plus placebo group, 55.1% ($P$=0.001) in the GOL 50 mg plus MTX group, and 56.2% ($P$<0.001) in the GOL 100 mg plus MTX group. At week 24, median Health Assessment Questionnaire Disease Index (HAQ-DI) score improvements from baseline for the placebo plus MTX, GOL 100 mg plus placebo, GOL 50 mg plus MTX, and GOL 100 mg plus MTX groups were 0.13, 0.13 ($P$=0.240), 0.38 ($P$<0.001), and 0.50 ($P$<0.001), respectively.

## CEP

In the Phase III trial Rapid-1, 982 active RA patients were randomized to receive subcutaneous CEP at an initial dose of 400 mg given at weeks 0, 2, and 4, with a subsequent dosage of 200 or 400 mg every 2 weeks plus MTX, or placebo plus MTX.[20] At week 24, the ACR20 response rates were 13.6%, 58.8%, and 60.8% for the placebo, CEP 200 mg, and CEP 400 mg groups, respectively. At week 52, mean radiographic progression from baseline was reduced in patients treated with CEP 200 mg (0.4 Sharp units) or 400 mg

(0.2 Sharp units), compared with placebo treated patients (2.8 Sharp units, $P<0.001$).

## ETA

In a Phase II study, 234 active RA patients who had an inadequate response to previous treatment regimens including MTX were randomly assigned to receive twice weekly subcutaneous injections of ETA (10 or 25 mg) or placebo for 24 weeks. At week 24, the ACR20 response rates were 51%, 59%, and 11% in the ETA 10 mg, ETA 20 mg, and placebo groups, respectively.[21] In the subsequent Phase III TEMPO trial, 682 patients with active RA were randomly allocated to treatment with ETA 25 mg (subcutaneously twice weekly), oral MTX, or the combination.[22] The numeric index of the ACR response area under the curve over the first 24 weeks was significantly greater in the combination group than the ETA alone or MTX alone groups ($P<0.0001$). Moreover, at week 52, the combination was more efficacious than ETA alone or MTX alone in protecting against joint damage (mean total Sharp score: –0.54 versus 0.52, $P=0.0006$; –0.54 versus 2.80, $P<0.0001$, respectively).

## Indirect comparisons of the efficacy of TCZ and TNF inhibitors

As indicated above, the efficacy of TCZ and TNF inhibitors in treating moderate to severe RA in patients who experienced an inadequate response to MTX has been demonstrated in separate studies. Although several systematic reviews have indirectly compared the efficacy of TCZ and TNF inhibitors in treating RA, only one trial, the ADACTA, has directly compared the efficacy of these agents.[23]

Bergman et al conducted a systematic literature review of double blind, randomized, placebo-controlled trials that spanned an 18-year period and investigated the effectiveness of TCZ (three trials; OPTION, LITHE, and TOWARD) and TNF inhibitors ADA, IFX, and ETA (total 11 trials) in treating RA in patients who experienced an inadequate response to sDMARDs.[24] The effectiveness of TCZ is comparable to that of each of the TNF inhibitors with respect to ACR20 and ACR50 responses, but greater than that of the TNF inhibitors with respect to ACR70 response. Another systematic review of selected clinical trials involving combination therapy with MTX concluded that there was no difference in efficacy on the basis of ACR50 response criterion at 24/30 weeks between TNF inhibitors and TCZ.[25] Turkstra et al reported a mixed treatment comparison of the short-term efficacy of nine bDMARDs, including TNF inhibitors and TCZ in patients with established RA.[26] They found that the ACR50 response rate of TCZ at 6 months is comparable to that of ADA, ETA, GOL, and IFX. In an indirect comparison, Salliot et al found no significant difference in the efficacy of TCZ and GOL in treating RA patients who had an inadequate response to TNF inhibitors (ADA, ETA, and IFX).[27] Orme et al reported the results of a network meta-analysis of the efficacy of bDMARDs with or without sDMARDs.[28] Odds ratios (covariate analysis) of ACR20/50/70 responses for ADA plus sDMARDs and TCZ plus sDMARDs versus sDMARDs alone were 3.374/4.203/4.58 and 4.363/5.797/9.23, respectively. In contrast, odds ratios (fixed effect) of ACR20/50/70 responses for ADA and TCZ versus placebo were 4.95/4.82/11.42 and 26.17/46.94/55.54, respectively. Pierreisnard et al also reported that there were no significant differences between the various TNF inhibitors and TCZ in terms of clinical efficacy (ACR50) in patients who had an inadequate MTX response.[29] Jones et al summarized the evidence regarding radiographic damage with bDMARDs, either alone or in combination with MTX.[30] For biologic monotherapy, TCZ, ADA, and ETA were significantly better than MTX, with TCZ ranking first, whereas GOL had no significant effect (Figure 1). For a bDMARD in combination with MTX compared with MTX alone, TCZ and all TNF inhibitors were effective at slowing X-ray progression. Taken together, the evidence from these indirect comparisons indicates that the efficacy of TCZ is comparable to that of TNF inhibitors when used in combination with MTX and that TCZ monotherapy is superior to TNF inhibitor monotherapy.

## Direct comparisons of the efficacy of TCZ and ADA

The head-to-head ADACTA trial compared the efficacy of TCZ with that of ADA as monotherapy for RA.[23] A total of 325 patients were randomly assigned to receive either TCZ 8 mg/kg intravenously every 4 weeks plus placebo subcutaneously every 2 weeks or ADA 40 mg subcutaneously every 2 weeks plus placebo intravenously every 4 weeks for 24 weeks. At week 24, patients treated with TCZ had a greater decrease in DAS28 than patients treated with ADA (–3·3 versus –1·8; $P<0.0001$). The proportion of patients attaining DAS28 remission was 39.9% with TCZ and 10.5% with ADA. ACR20, ACR50, and ACR70 response rates were achieved in 65% and 49.4% ($P<0.01$), 47.2% and 27.8% ($P<0.01$), and 32.5% and 17.9% ($P<0.01$) of patients treated with TCZ and ADA, respectively. These results demonstrated the overall superiority of monotherapy with TCZ compared with monotherapy with ADA for the treatment

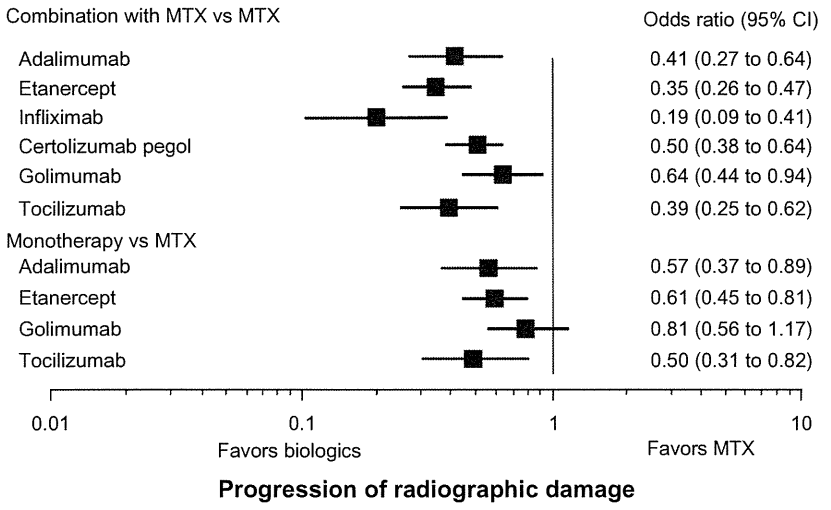


**Figure 1** Indirect comparisons of the suppressive effects of tocilizumab and tumor necrosis factor inhibitors on radiographic damage.
**Notes:** Copyright © 2012. Adapted from Dove Medical Press. Jones G, Darian-Smith E, Kwok M, Winzenberg T. Effect of biologic therapy on radiological progression in rheumatoid arthritis: what does it add to methotrexate? *Biologics*. 2012;6:155–161.[30] In combination with methotrexate (MTX) compared with MTX alone, tocilizumab and all tumor necrosis factor inhibitors are effective at slowing X-ray progression. As monotherapy, adalimumab, etanercept, and tocilizumab are significantly better than MTX, whereas golimumab had no significant effect. The x-axis shows progression of radiographic damage.
**Abbreviations:** CI, confidence interval; MTX, methotrexate.

of RA. Clinical evidence demonstrated that coadministration of TNF inhibitors and MTX is more efficacious than administration of TNF inhibitors alone in treating RA.[31] In contrast, the findings of the ACT-RAY trial comparing the efficacy of TCZ plus MTX therapy with that of TCZ monotherapy in a setting that closely resembled a real life clinical practice showed that TCZ monotherapy is not clinically inferior to TCZ combination therapy,[32] indicating that as monotherapy, TCZ appears to be more effective than TNF inhibitors at suppressing disease activity.

In a Japanese cohort, the Tsurumai Biologics Communication Registry, the proportion of patients who achieved low disease activity, clinical remission, and a moderate or good European League Against Rheumatism (EULAR) response at 24 weeks was determined following treatment with ADA or TCZ.[33] A total of 120 patients were treated with ADA (77% of patients in combination with MTX), while 99 patients were treated with TCZ (36% of patients in combination with MTX). There was no significant difference between ADA and TCZ treated patients with respect to the proportion of low disease activity and remission, but a higher proportion of patients treated with TCZ achieved a moderate or good EULAR response.

## Comparative safety and tolerability studies

The integrated safety of TCZ was evaluated in clinical trials through comparisons of adverse events (AEs) between a control population (4,199) and a TCZ treated population (4,009), and the results were reported in 2011.[34] Total exposure to TCZ was 8,580 patient-years (PYs), and the total duration of observation was 9,414 PYs. Overall AE and serious AE rates were 278.2/100 PYs and 14.4/100 PYs, respectively. AEs included serious infection (4.7/100 PYs), opportunistic infection (0.23/100 PYs), gastrointestinal perforation (0.28/100 PYs), malignancy (1.1/100 PYs), myocardial infarction (0.25/100 PYs), and stroke (0.19/100 PYs).

In another systematic review in which the total duration of observation was 12,293 PYs, infections were also the most common AE and serious AE identified, and the rate of serious infections was 4.5/100 PYs.[35] The short-term (28 weeks) safety of TCZ was monitored in a postmarketing surveillance study in Japan involving 7,901 patients.[36] The incidence of total AEs and serious AEs was 43.9% and 9.6%, respectively. Infection and infestation were the most frequent (11.1%) and serious (0.5%) AEs. Analysis of long-term clinical trial safety data showed that rates of serious AEs, serious infections, and cardiovascular events remained stable during continued exposure to TCZ. Infection was identified as the most frequent serious AE. The most common infections reported in randomized controlled trials (RCTs) were pneumonia (0.9/100 PYs) and skin or soft tissue infections (0.9/100 PYs). These results led to the conclusion that infections are the most frequent AEs associated with TCZ. A meta-analysis comparing the safety

profile of TCZ with those of TNF inhibitors (7–9/100 PYs) showed similar rates of serious infections,[37,38] although among TNF inhibitors, an increased risk of serious infection was observed with IFX.

As TNF-α plays a crucial role in the host defense against intracellular pathogens (eg, TNF-α activates macrophages and stimulates the formation and maintenance of granulomas to protect against *Mycobacterium tuberculosis* infection), TNF inhibitors increase the risk of tuberculosis reactivation, as evidenced by clinical trials showing an incidence of 0.4% with IFX.[39] Within the anti-TNF biologic cohort, IFX and ADA are associated with a 3- to 4-fold higher risk of reactivation than ETA.[40] It seems likely that the incidence of reactivation of tuberculosis is lower during TCZ treatment than during anti-TNF treatment, as there are only six reported cases in the worldwide TCZ clinical trials database, which covers >10,000 PYs of exposure.[41] Moreover, according to QuantiFERON assay data, TNF inhibitors (but not TCZ) influence tuberculosis-antigen-induced IFN-γ production,[42] suggesting that TCZ may be safer than TNF inhibitors with respect to reactivation of latent tuberculosis.

In contrast to TNF inhibitors, gastrointestinal perforation appears to be an AE specific to TCZ, with an incidence rate of 1.9–2.8/1,000 PYs.[34,43] This rate is between the 3.9/1,000 PYs for corticosteroids and 1.3/1,000 PYs for TNF inhibitors, as indicated in the United Health Care database.[43] A total of 17 of 29 (59%) reported events involved colonic diverticular perforation, suggesting that TCZ should not be used in patients with a history of diverticulitis.

Increases in mean fasting levels of plasma lipids, such as total cholesterol (TC), low-density lipoprotein (LDL), triglycerides, and high-density lipoprotein (HDL), occur in 20%–30% of patients treated with TCZ, which appeared higher in patients treated with TNF inhibitors.[34,36] A 24-week, double blind, randomized, multicenter, two part, Phase III trial followed by an 80-week open label trial (MEASURE) evaluated lipid and lipoprotein levels, HDL particle composition, markers of coagulation, and thrombosis in 132 patients with RA receiving either TCZ or placebo.[44] At week 12, median TC, LDL-cholesterol (LDL-C), and triglyceride levels increased in TCZ recipients versus placebo recipients (12.6% versus 1.7%, 28.1% versus 2.2%, 10.6% versus −1.9%, respectively; all $P<0.01$). There were no significant differences in the concentrations of mean small LDL, mean oxidized LDL, or total HDL-C, but the HDL associated serum amyloid A (SAA) content decreased in TCZ treated patients. TCZ also induced reductions (>30%) in secretory phospholipase A2-IIA, lipoprotein (a), fibrinogen, and D-dimers and an elevation in the level of paraoxonase (all $P<0.0001$ versus placebo). These data constitute detailed evidence that TCZ modulates lipoprotein particles and other surrogates of vascular risk.

Comparisons of drug survival with TNF inhibitors have been reported in some registries. In the Consortium of Rheumatology Researchers of North America registry, the 24-month persistence for biologically naive patients on the new anti-TNF treatments IFX, ETA, and ADA was 63%, 53%, and 53% respectively.[45] The Lombardy Rheumatology Network registry reported 2.5-year treatment continuation rates for IFX, ETA, and ADA of approximately 56%, 72%, and 57%, respectively.[46] The Swiss Clinical Quality Management for Rheumatoid Arthritis registry reported 2.5-year drug survival rates for IFX, ETA, and ADA of approximately 51%, 58%, and 61%, respectively.[47] An Italian study group (Gruppo Italiano di Studio sulle Early Arthritides registry) reported 2.5-year continuation rates for IFX, ETA, and ADA of approximately 52%, 65%, and 52%, respectively.[48]

There are few reports describing TCZ drug survival. The Danish Nationwide Rheumatological Database registry reported 48-, 96-, and 144-week TCZ adherence rates of 61%, 54%, and 47%, respectively.[49] In contrast, the Danish Nationwide Rheumatological Database registry reported 48-month drug survival rates for IFX, ETA, and ADA of 41%, 56%, and 52%, respectively.[50] The Japanese Osaka University Biologics for Rheumatic Diseases registry reported 1-year drug continuation rates for TCZ, IFX, ETA, and ADA of 89%, 73%, 86%, and 78%, respectively, and 2.5-year rates of 79%, 47%, 78%, and 55%, respectively.[51] In this registry, the continuation rates for TCZ and ETA are significantly higher than those for IFX and ADA. The most frequent reasons given for discontinuation are AEs for TCZ and a lack of efficacy for ADA and IFX. The Registry of Japanese Rheumatoid Arthritis Patients for Long-term Safety reported significantly lower discontinuation rates due to lack of efficacy for patients taking ETA compared with patients taking IFX or TCZ.[52] Finally, the Cohort of Arthritis Biologic Users at Kameda Institute registry reported that the drug survival and safety profiles of TCZ are similar to those of TNF inhibitors (IFX, ETA, and ADA).[53] The results regarding tolerability are summarized in Table 2. These reports indicate that tolerability of TCZ is comparable to or better than that of TNF inhibitors.

**Table 2** Comparative tolerability of tocilizumab with tumor necrosis factor inhibitors

| Registry name | Retention period | Drug survival rate (%) | | | |
|---|---|---|---|---|---|
| | | TCZ | IFX | ETA | ADA |
| CORRONA[45] | 24 months | | 63 | 53 | 53 |
| LOHREN[46] | 2.5 years | | 56 | 72 | 57 |
| SCQM-RA[47] | 2.5 years | | 51 | 58 | 61 |
| GISEA[48] | 2.5 years | | 52 | 65 | 52 |
| DANBIO[49,50] | 96 weeks | 54 | | | |
| | 48 months | | 41 | 56 | 52 |
| BiRD[51] | 2.5 years | 79 | 47 | 78 | 55 |

**Abbreviations:** ADA, adalimumab; BiRD, Biologics for Rheumatic Diseases; CORRONA, Consortium of Rheumatology Researchers of North America; DANBIO, Danish Nationwide Rheumatological Database; ETA, etanercept; GISEA, Gruppo Italiano di Studio sulle Early Arthritides; IFX, infliximab; LOHREN, Lombardy Rheumatology Network; SCQM-RA, Swiss Clinical Quality Management for Rheumatoid Arthritis; TCZ, tocilizumab; TNF, tumor necrosis factor.

## Comparative patient focused perspectives, such as quality of life, patient satisfaction/acceptability, adherence, and uptake

In all Phase III trials modified HAQ-DI scores significantly improved with TCZ treatment. Moreover, based on functional assessment of chronic illness therapy (FACIT), the OPTION and TOWARD studies reported that TCZ had an ameliorative effect, and the Short-Form (SF)36 Health Survey indicated both mental and physical (SF36-mental and SF-physical) effects.[11,12] In addition, the RADIATE study found that at week 24 versus placebo, TCZ treatment at 8 mg/kg was associated with significantly greater improvements in HAQ-DI, FACIT, and SF36-physical, and that TCZ treatment at 4 mg/kg was associated with greater improvements in HAQ-DI and SF36-physical.[54] Components of the Arthritis Impact Measurement Scale 2 (AIMS-2) (eg, physical score, symptom, and affect score) and those of SF36 (eg, bodily pain, general health, vitality, and mental health) improved in 39 patients in a clinical practice after 4 weeks of TCZ therapy, but there was no improvement in the social interaction component of AIMS-2 after 24 weeks of treatment.[55]

The Tocilizumab and DMARDs: Achievements in Rheumatoid Arthritis study reported improvements in diary documented fatigue, pain, and morning stiffness with TCZ treatment.[56] The mean FACIT-Fatigue score increased from 28.8±11.2 at baseline to 35.3±11.5 at week 4 and to 37.4±12.2 at week 24, and the mean HAQ-DI score decreased from 1.48±0.65 to 1.15±0.68 at week 4 and to 1.00±0.75 at week 24 or the last visit. Favorable mean changes from baseline to week 24 or the last visit were also observed in each of the domains of the SF36, especially in the physical domains. The Treatment Satisfaction Questionnaire for Medication, which was completed at the end of the study, showed a high level of patient agreement/satisfaction for each of the derived domains: "effectiveness" (69.4%), "side effects" (88.7%), "convenience" (72.4%), and "global satisfaction" (74.7%).

Fatigue represents an important symptom for patients with RA. Chauffier et al assessed the effect of biotherapies on fatigue based on data from ten RCTs involving patients with established RA.[57] Unfortunately, with respect to fatigue, they found that the overall effect size of all bDMARDs versus placebo at week 24 of treatment is small in established RA. In inadequate responders to sDMARDs, the effect size is similar for TNF inhibitors and nonanti-TNF bDMARDs including TCZ. Strand et al reported that ADA plus MTX significantly improved physical function and health-related quality of life in patients with early RA after 2 years of treatment.[58] However, no clinically meaningful differences between patients on ADA monotherapy or MTX were observed. In a recent meta-analysis, Callhoff et al studied the impact of bDMARDs including five TNF inhibitors but not TCZ on the physical function of patients with RA, as evaluated by Health Assessment Questionnaire.[59] Overall, bDMARDs produced greater improvement in physical function than sDMARDs, with an Health Assessment Questionnaire standardized mean difference of 0.44 (95% confidence interval [CI]: 0.38, 0.50). No significant differences between TNF inhibitors were observed.

Huynh et al examined patient treatment preference.[60] The most frequent reason given for choosing intravenous treatment was "safety" (62%), followed by "easy to manage" (39%). The two most frequent reasons given for choosing self-injection at home were "time constraints" and "easy to manage" (both 57%). The majority of RA patients already treated with bDMARDs in that study preferred the route of administration they were used to. The majority of the patients not currently treated with a bDMARD preferred subcutaneous treatment at home. A feeling of safety was important to patients who preferred intravenous treatment. Health professionals as a group may be biased toward the use of subcutaneous treatment. It is now possible to administer TCZ subcutaneously as well as intravenously.[61,62] Although subcutaneous injection of TCZ is disadvantageous in heavy patients, the fact that patients can now choose the administration route is a positive development.

## Comparison of the cost-effectiveness of TCZ and TNF inhibitors

Although demonstrations of the outstanding efficacy of TNF inhibitors and TCZ have led to a paradigm shift with respect

to the management of RA, the relatively high cost of these drugs imposes a large burden on both patients and society.[63] The Swedish Early Interventions In Rheumatoid Arthritis project demonstrated that drug costs increased primarily due to the introduction of biologics.[64] Sick leave decreases during the first year, but disability pensions increase, resulting in no change in indirect costs. Over the following years, disability pensions increase further and indirect costs also increase. In the 6 years after diagnosis of early RA, drug costs are partially offset by decreasing outpatient visits, but indirect costs remain unchanged and total costs increase. Therefore, the cost of bDMARDs is a significant problem. bDMARDs significantly increase the quality-adjusted life years (QALYs) gained when compared to MTX alone. QALY is a measure of disease burden affecting the quality and quantity of the life lived. In Finland, TCZ plus MTX was found to be more cost-effective than ADA plus MTX or ETA plus MTX in comparison with MTX alone.[65] A QALY gained with retail priced (wholesale priced) TCZ plus MTX costs Euro (€)18,957 (€17,057) more than MTX alone. Diamantopoulos et al reported the cost utility of TCZ in RA patients with an inadequate response to sDMARDs from a payer's perspective in Italy.[66] Replacement of TNF inhibitors (ADA, ETA, and IFX) with TCZ reduces total costs over a patient's lifetime (base-case analysis, TCZ: €141,100 versus TNF inhibitors: €143,500). Patients receiving TCZ realize more QALYs than patients receiving standard of care (9.8881 QALYs versus 9.3502 QALYs). When TCZ is added to standard of care without replacing TNF inhibitors, the incremental cost-effectiveness ratio becomes €17,100 per QALY.

In the ADACTA study, economic evaluation of the cost per response or remission of TCZ versus ADA was reported for Spain.[67] The cost per ACR20/50/70 response is lower with TCZ than with ADA (€8,105/11,162/16,211 versus €11,553/20,529/31,882). The cost of attaining DAS28 remission with TCZ and ADA is €13,204 and €54,352, respectively. Treatment with TCZ was dominant in all scenarios analyzed. Similar economic evaluation of TCZ versus ADA from the ADACTA trial was conducted in Australia.[68] TCZ monotherapy was found to result in lower total treatment costs (in Australian dollars [$]) per patient over 24 weeks compared with ADA monotherapy ($9,739 versus $10,722).

In the UK, the addition of TCZ in combination with MTX to treat severe active RA in patients with an inadequate response to sDMARDs was found to produce a gain of 1.17 QALYs per patient, at an incremental cost of UK pound (£)23,253.[69] This equates to an incremental cost-effectiveness ratio (ICER) of £19,870. The addition of TCZ in combination with MTX to the current Scottish standard of care in adult TNF inhibitor-inadequate responders with moderate to severe active RA produces a gain of 1.234 QALYs per patient, at an incremental cost of £27,465.[70] This equates to an ICER of £22,254. Tanaka et al reported the cost-effectiveness of TCZ in Japan.[71] The lifetime cumulative costs and QALYs were 35.4 million Japanese yen (¥) and 11.7, respectively, in the TCZ group and ¥23.3 million and 9.3, respectively, in the MTX group. The ICER for TCZ was ¥4.94 million, with a 66.2% probability of falling below the allowable threshold based upon probabilistic sensitivity analysis. These findings suggest that TCZ is more cost-effective than TNF inhibitors, including ADA, ETA, and IFX.

## Conclusion and place in therapy

The property of TCZ and TNF inhibitors is summarized in Figure 2. Based upon recent findings, the EULAR recommendations for the management of RA were updated in 2013.[72] In patients responding insufficiently to MTX and/or other sDMARDs, with or without glucocorticoids, use of bDMARDs should commence with MTX. First line bDMARDs include TNF inhibitors, abatacept, and TCZ, and under certain circumstances, rituximab. If biologic monotherapy must be initiated, only TCZ has supportive evidence. However, TCZ, TNF inhibitors, and other bDMARDs do not produce beneficial effects in all active RA patients. Therefore, to determine the optimal strategy for using particular bDMARDs in individual RA patients, the characteristic features of these drugs should be clarified.[73]

RA animal models have provided some clarification. The most well-known animal model of RA is collagen-induced arthritis, which involves injection of mice with type II collagen to produce an immune response directed at connective tissues. Both IL-6 and TNF-α have been shown to play a major role in the development and progression of joint destruction in the collagen-induced arthritis model. Immunization with type II collagen in this model primarily increases the frequency of Th17 cells. Treatment of immunized mice with anti-IL-6 receptor Ab during priming leads to marked suppression of both the induction of Th17 cells and arthritis development, whereas administration of soluble TNF receptor-Fc fusion protein from day 0 to 14 fails to suppress Th17 differentiation and arthritis development.[74] Anti-type II collagen Ab-induced arthritis (CAIA) is a model in which the priming phase of T-cell dependent Ab generation is skipped. Although TNF-α and IL-6 are also elevated in this model, arthritis is suppressed in TNF-α- but not in IL-6-deficient mice, indicating that TNF-α plays a more significant role than IL-6 in joint

| | Tocilizumab | | TNF inhibitors |
|---|---|---|---|
| Efficacy (with MTX) on disease activity | Excellent | = | Excellent |
| Efficacy (as monotherapy) on disease activity | Excellent | > | Good |
| Safety profile | Tolerable | | Tolerable |
| Incidence of AEs | | | |
| Overall infections | | = | |
| Reactivation of TB | | < | |
| Elevation of T-CHO | | > | |
| GI perforation | | > | |
| Tolerability | Good | > or = | Good |
| Effects on acute-phase proteins (CRP,SAA, Hepcidin) | Excellent | > | Good |
| Cost-effectiveness | | > | |

**Figure 2** Properties of tocilizumab and tumor necrosis factor inhibitors in the management of rheumatoid arthritis.
**Abbreviations:** AEs, adverse events; CRP, C-reactive protein; GI, gastrointestinal; MTX, methotrexate; SAA, serum amyloid A; TB, tuberculosis; T-CHO, total cholesterol.

inflammation in CAIA.[75] These findings suggest that IL-6 is essential for the induction of immunological abnormalities and the development of arthritis and that the pathological role of IL-6 is different from that of TNF-α, which is primarily involved in the development of joint inflammation.

Analyses of various markers during biologic treatment are also helpful to clarify the characteristics of bDMARDs. Both TNF inhibitors and TCZ lead to improvements in serological and urinary markers related to bone and cartilage metabolism. Several immunological studies have sought to clarify the mechanisms underlying the effects of TCZ. Of particular importance is to determine whether TCZ can correct the Th17/Treg imbalance, which is thought to be a fundamental immunological abnormality in RA.[76] The results of preliminary studies suggest that inhibition of IL-6 function by TCZ corrects the imbalance between Th17 and Treg cells in the peripheral CD4-positive T-cell population.[77,78] In contrast, TNF-α suppresses Treg function by dephosphorylating serine 418 in the C-terminal DNA-binding domain of the forkhead box P3, whereas anti-TNF therapy can restore Treg cell function.[79] Moreover, a study involving eight patients with RA demonstrated that 6 months of treatment with TCZ causes a selective decrease in IL-21 production by memory/activated T-cells.[80] IL-21 is known to promote plasma cell differentiation and induce IgG4 production, and TCZ treatment leads to a reduction in the serum levels of IgG4-specific anticitrullinated peptide antibody, indicating the presence of a pathway involving IL-6, IL-21, and IgG4 autoantibodies in RA. In another study, Roll et al examined the in vivo effect of TCZ on the B-cell compartment in 16 RA patients and found that TCZ induces a significant reduction in peripheral preswitch and postswitch memory B-cells.[81] In addition, TCZ (but not ETA) significantly reduces somatic hypermutation in immunoglobulin gene rearrangements in preswitch memory B-cells,[82] suggesting modulation of memory B-cells as a possible mechanism for TCZ. Further evaluation is required to clarify the effects of bDMARDs in treating the immunological abnormalities associated with RA.

IL-6, which was found to be identical to hepatocyte-stimulating factor, induces the expression of various acute phase proteins, such as C-reactive protein, hepcidin, SAA, and fibrinogen, indicating that IL-6 plays a role in the development of systemic inflammatory symptoms, signs, and complications. TCZ treatment is expected to

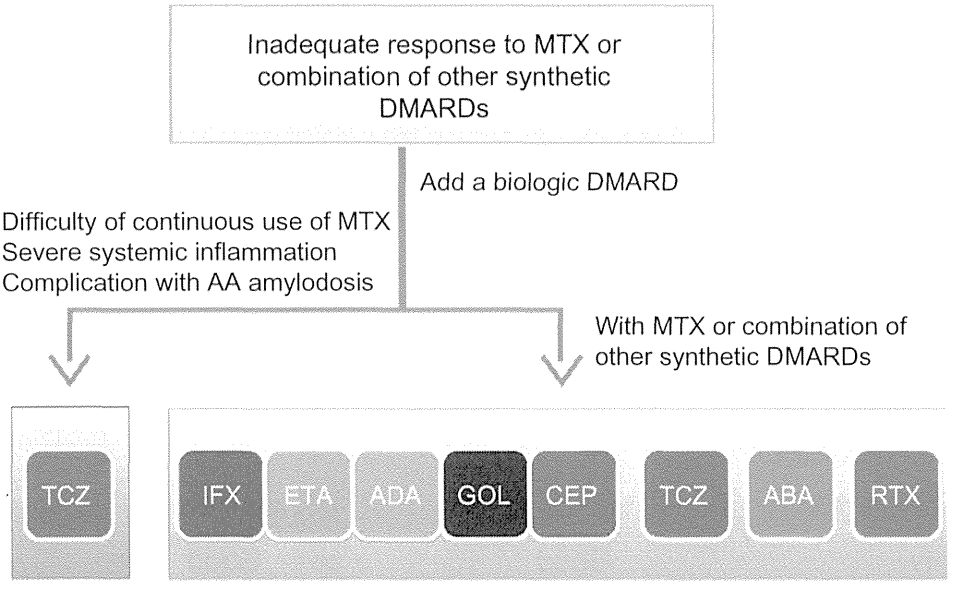


**Figure 3** Selection of biologic disease modifying antirheumatic drugs.

**Notes:** Rheumatoid arthritis patients who fail to respond to methotrexate (MTX) alone or in combination with other synthetic disease modifying antirheumatic drugs (DMARDs) need to be treated with a biologic DMARD. For patients who can continue to receive MTX, any of the seven biologic DMARDs should be selected. These include five tumor necrosis factor inhibitors (infliximab, etanercept, adalimumab, golimumab, and certolizumab pegol), the IL-6 receptor blocker tocilizumab, the T-cell stimulation blocker abatacept, and the B-cell depletory rituximab. Rituximab is recommended to be used for patients who have certain contraindications for other agents such as a recent history of lymphoma, latent tuberculosis with contraindications to the use of chemoprophylaxis, live in a tuberculosis endemic region, or a previous history of demyelinating disease. Tocilizumab may be selected for patients who 1) cannot continue treatment with MTX or other synthetic DMARDs, 2) present with severe inflammatory findings, and 3) have or who are at high risk of developing amyloid A amyloidosis.

**Abbreviations:** ABA, abatacept; ADA, adalimumab; CEP, certolizumab pegol; DMARDs, disease modifying antirheumatic drugs; ETA, etanercept; GOL, golimumab; IFX, infliximab; MTX, methotrexate; RTX, rituximab; TCZ, tocilizumab.

ameliorate the inflammatory effects and inhibit the development of complications. Increased production of hepcidin predominantly induced by IL-6 leads to anemia associated with chronic disorders.[83] A comparative evaluation of the effects of TCZ and TNF inhibitors on serum hepcidin and anemia found that significant improvement in anemia and reduction in serum hepcidin levels are more pronounced in the TCZ treated patients than in TNF inhibitor treated patients.[84] Amyloid A amyloidosis is a serious complication of RA, as amyloid fibril deposition causes progressive deterioration in various organs,[85] although due to a marked progression of antirheumatic treatment, the incidence of amyloid A amyloidosis has recently decreased.[86,87] SAA is an amyloid fibril precursor protein. Because the synthesis of SAA depends primarily on IL-6, TCZ injection promptly reduces the serum concentration of SAA, just as in the case of C-reactive protein, and the suppressive activity of TCZ on the serum SAA level is more powerful than that of TNF inhibitors.[88,89] Case reports and series studies published to date have demonstrated the marked ameliorative effect of TCZ on gastrointestinal symptoms and renal dysfunction caused by amyloid A amyloidosis.[90–92]

On the basis of these findings, we suggest that TCZ can be selected as the first line biologic for patients who 1) cannot continue treatment with MTX or other sDMARDs, 2) present with severe inflammatory findings, and 3) have or who are at high risk of developing amyloid A amyloidosis (Figure 3). Moreover, medication adherence and cost-effectiveness appears to favor TCZ in comparison with TNF inhibitors. However, further evaluation and clarification of the characteristic features of bDMARDs are essential to determine the optimal treatment for individual RA patients.

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## References

1. Mclnnes IB, Schett G. The pathogenesis of rheumatoid arthritis. *N Engl J Med.* 2011;365(23):2205–2219.
2. Smolen JS, Aletaha D, Koeller M, Weisman MH, Emery P. New therapies for treatment of rheumatoid arthritis. *Lancet.* 2007;370(9602):1861–1874.
3. Thalayasingam N, Isaacs JD. Anti-TNF therapy. *Best Pract Res Clin Rheumatol.* 2011;25(4):549–567.

4. Tanaka T, Ogata A, Narazaki M. Tocilizumab for the treatment of rheumatoid arthritis. *Expert Rev Clin Immunol*. 2010;6(6):843–854.
5. Tanaka T, Narazaki M, Kishimoto T. Therapeutic targeting of the interleukin-6 receptor. *Annu Rev Pharmacol Toxicol*. 2012;52: 199–219.
6. Tanaka T, Ogata A, Kishimoto T. Targeting of interleukin-6 for the treatment of rheumatoid arthritis: a review and update. *Rheumatol Curr Res*. 2013;S4:002.
7. Sato K, Tsuchiya M, Saldanha J, et al. Reshaping a human antibody to inhibit the interleukin-6 dependent tumor cell growth. *Cancer Res*. 1993;53(4):851–856.
8. Jones G, Sebba A, Gu J, et al. Comparison of tocilizumab monotherapy versus methotrexate monotherapy in patients with moderate to severe rheumatoid arthritis: the AMBITION study. *Ann Rheum Dis*. 2010;69(1):88–96.
9. Nishimoto N, Hashimoto J, Miyasaka N, et al. Study of active controlled monotherapy used for rheumatoid arthritis, an IL-6 inhibitor (SAMURAI): evidence of clinical and radiographic benefit from an x-ray reader-blinded randomized controlled trial of tocilizumab. *Ann Rheum Dis*. 2007;66(9):1162–1167.
10. Nishimoto N, Miyasaka N, Yamamoto K, et al. Study of active controlled tocilizumab monotherapy for rheumatoid arthritis patients with an inadequate response to methotrexate (SATORI): significant reduction in disease activity and serum vascular endothelial growth factor by IL-6 receptor inhibition therapy. *Mod Rheumatol*. 2009;19(1):12–19.
11. Smolen JS, Beaulieu A, Rubbert-Roth A, et al; OPTION Investigators. Effect of interleukin-6 receptor inhibition with tocilizumab in patients with rheumatoid arthritis (OPTION study): a double-blind, placebo-controlled, randomized trial. *Lancet*. 2008;371(9617):987–997.
12. Genovese MC, McKay JD, Nasonov EL, et al. Interleukin-6 receptor inhibition with tocilizumab reduces disease activity in rheumatoid arthritis with inadequate response to disease-modifying antirheumatic drugs: the tocilizumab in combination with traditional disease-modifying antirheumatic drug therapy study. *Arthritis Rheum*. 2008;58(10):2968–2980.
13. Emery P, Keystone E, Tony HP, et al. IL-6 receptor inhibition with tocilizumab improves treatment outcomes in patients with rheumatoid arthritis refractory to anti-tumor necrosis factor biologicals: results from a 24-week multicentre randomized placebo controlled trial. *Ann Rheum Dis*. 2008;67(11):1516–1523.
14. Kremer JM, Blanco R, Brzosko M, et al. Tocilizumab inhibits structural joint damage in rheumatoid arthritis patients with inadequate responses to methotrexate: results from the double-blind treatment phase of a randomized placebo-controlled trial of tocilizumab safety and prevention of structural joint damage at one year. *Arthritis Rheum*. 2011;63(3): 609–621.
15. Weinblatt ME, Keystone EC, Furst DE, et al. Adalimumab, a fully human anti-tumor necrosis factor alpha monoclonal antibody, for the treatment of rheumatoid arthritis in patients taking concomitant methotrexate: the ARMADA trial. *Arthritis Rheum*. 2003;48(1):35–45.
16. Breedveld FC, Weisman MH, Kavanaugh AF, et al. The PREMIER study: A multicenter, randomized, double-blind clinical trial of combination therapy with adalimumab plus methotrexate versus methotrexate alone or adalimumab alone in patients with early, aggressive rheumatoid arthritis who had not had previous methotrexate treatment. *Arthritis Rheum*. 2006;54(1):26–37.
17. Maini R, St Clari EW, Breedveld F, et al. Infliximab (chimeric anti-tumour necrosis factor alpha monoclonal antibody) versus placebo in rheumatoid arthritis patients receiving concomitant methotrexate: a randomised phase III trial. ATTRACT Study Group. *Lancet*. 1999;354(9194):1932–1939.
18. Breedveld FC, Emery P, Keystone E, et al. Infliximab in active early rheumatoid arthritis. *Ann Rheum Dis*. 2004;63(2):149–155.
19. Keystone EC, Genovese MC, Klareskog L, et al. Golimumab, a human antibody to tumour necrosis factor alpha given by monthly subcutaneous injections, in active rheumatoid arthritis despite methotrexate therapy: the GO-FORWARD Study. *Ann Rheum Dis*. 2009;68(6):789–796.
20. Keystone E, van der Heijde D, Mason D Jr, et al. Certolizumab pegol plus methotrexate is significantly more effective than placebo plus methotrexate in active rheumatoid arthritis: findings of a fifty-two-week, phase III, multicenter, randomized, double-blind, placebo-controlled, parallel-group study. *Arthritis Rheum*. 2008;58(11):3319–3329.
21. Moreland LW, Schiff MH, Baumgartner SW, et al. Etanercept therapy in rheumatoid arthritis. A randomized, controlled trial. *Ann Intern Med*. 1999;130(6):478–486.
22. Klareskog L, van der Heijde D, de Jager JP, et al; TEMPO (Trial of Etanercept and Methotrexate with Radiographic Patient Outcomes) study investigators. Therapeutic effect of the combination of etanercept and methotrexate compared with each treatment alone in patients with rheumatoid arthritis: double-blind randomised controlled trial. *Lancet*. 2004;363(9410):675–681.
23. Gabay C, Emery P, van Vollenhoven R, et al; ADACTA Study Investigators. Tocilizumab monotherapy versus adalimumab monotherapy for treatment of rheumatoid arthritis (ADACTA): a randomised, double-blind, controlled phase 4 trial. *Lancet*. 2013;381(9877):1541–1550.
24. Bergman GJ, Hochberg MC, Boers M, Wintfeld N, Kielhorn A, Jansen JP. Indirect comparison of tocilizumab and other biologic agents in patients with rheumatoid arthritis and inadequate response to disease-modifying antirheumatic drugs. *Semin Arthritis Rheum*. 2010;39(6): 425–441.
25. Gallego-Galisteo M, Villa-Rubio A, Alegre-del Rey E, Marquez-Fernandez E, Ramos-Baez JJ. Indirect comparison of biological treatments in refractory rheumatoid arthritis. *J Clin Pharm Ther*. 2012;37(3): 301–307.
26. Turkstra E, Ng SK, Scuffham PA. A mixed treatment comparison of the short-term efficacy of biologic disease modifying anti-rheumatic drugs in established rheumatoid arthritis. *Curr Med Res Opin*. 2011;27(10): 1885–1897.
27. Salliot C, Finckh A, Katchamart W, et al. Indirect comparisons of the efficacy of biological antirheumatic agents in rheumatoid arthritis in patients with an inadequate response to conventional disease-modifying antirheumatic drugs or to an anti-tumour necrosis factor agent: a meta-analysis. *Ann Rheum Dis*. 2011;70(2):266–271.
28. Orme ME, Macgilchrist KS, Mitchell S, Spurden D, Bird A. Systematic review and network meta-analysis of combination and monotherapy treatments in disease-modifying antirheumatic drug-experienced patients with rheumatoid arthritis: analysis of American College of Rheumatology criteria scores 20, 50, and 70. *Biologics*. 2012;6: 429–464.
29. Pierreisnard A, Issa N, Barnetche T, Richez C, Schaeverbeke T. Meta-analysis of clinical and radiological efficacy of biologics in rheumatoid arthritis patients naive or inadequately responsive to methotrexate. *Joint Bone Spine*. 2013;80(4):386–392.
30. Jones G, Darian-Smith E, Kwok M, Winzenberg T. Effect of biologic therapy on radiological progression in rheumatoid arthritis: what does it add to methotrexate? *Biologics*. 2012;6:155–161.
31. Emery P, Sebba A, Huizinga TW. Biologic and oral disease-modifying antirheumatic drug monotherapy in rheumatoid arthritis. *Ann Rheum Dis*. 2013;72(12):1897–1904.
32. Dougados M, Kissel K, Sheeran T, et al. Adding tocilizumab or switching to tocilizumab monotherapy in methotrexate inadequate responders: 24-week symptomatic and structural results of a 2-year randomised controlled strategy trial in rheumatoid arthritis (ACT-RAY). *Ann Rheum Dis*. 2013;72(1):43–50.
33. Takahashi N, Kojima T, Kaneko A, et al. Clinical efficacy of abatacept compared to adalimumab and tocilizumab in rheumatoid arthritis patients with high disease activity. *Clin Rheumatol*. 2014;33(1):39–47.
34. Schiff MH, Kremer JM, Jahreis A, Vernon E, Isaacs JD, van Vollenhoven RF. Integrated safety in tocilizumab clinical trials. *Arthritis Res Ther*. 2011;13(5):R141.
35. Genovese MC, Rubbert-Roth A, Smolen JS, et al. Longterm safety and efficacy of tocilizumab in patients with rheumatoid arthritis: a cumulative analysis of up to 4.6 years of exposure. *J Rheumatol*. 2013;40(6): 768–780.

36. Koike T, Harigai M, Inokuma S, et al. Effectiveness and safety of tocilizumab: postmarketing surveillance of 7901 patients with rheumatoid arthritis in Japan. *J Rheumatol*. 2014;41(1):15–23.
37. Singh JA, Wells GA, Christensen R, et al. Adverse effects of biologics: a network meta-analysis and Cochrane overview. *Cochrane Database Syst Rev*. 2011;16(2):CD008794.
38. Jain A, Singh JA. Harms of tumor necrosis factor inhibitions in rheumatic diseases: a focused systematic review of the literature. *Immunotherapy*. 2013;5(3):265–299.
39. Gómez-Reino JJ, Carmona L, Valverde VR, Mola EM, Montero MD; BIOBADASER Group. Treatment of rheumatoid arthritis with tumor necrosis factor inhibitors may predispose to significant increase in tuberculosis risk: a multicenter active-surveillance report. *Arthritis Rheum*. 2003;48(8):2122–2127.
40. Tubach F, Salmon D, Ravaud P, et al; Research Axed on Tolerance of Biotherapies Group. Risk of tuberculosis is higher with anti-tumor necrosis factor monoclonal antibody therapy than with soluble tumor necrosis factor receptor therapy: the three-year prospective French Research Axed on Tolerance of Biotherapies registry. *Arthritis Rheum*. 2009;60(7):1884–1894.
41. van Vollenhoven RF, Nishimoto N, Yamanaka H. Experience with Mycobacterium tuberculosis infection reported in the tocilizumab worldwide RA safety database. *Ann Rheum Dis*. 2009;68(Suppl 3):567.
42. Ogata A, Mori M, Hashimoto S, et al. Minimal influence of tocilizumab on IFN-gamma synthesis by tuberculosis antigens. *Mod Rheumatol*. 2010;20(2):130–133.
43. Gout T, Ostör AJ, Nisar MK. Lower gastrointestinal perforation in rheumatoid arthritis patients treated with conventional DMARDs or tocilizumab: a systematic literature review. *Clin Rheumatol*. 2011;30(11):1471–1474.
44. McInnes IB, Thompson L, Giles JT, et al. Effect of interleukin-6 receptor blockade on surrogates of vascular risk in rheumatoid arthritis: MEASURE, a randomised, placebo-controlled study. *Ann Rheum Dis*. Epub December 24, 2013.
45. Greenberg JD, Reed G, Decktor D, et al; CORRONA Investigators. A comparative effectiveness study of adalimumab, etanercept and infliximab in biologically naive and switched rheumatoid arthritis patients: results from the US CORRONA registry. *Ann Rheum Dis*. 2012;71(7):1134–1142.
46. Marchesoni A, Zaccara E, Gorla R, et al. TNF-alpha antagonist survival rate in a cohort of rheumatoid arthritis patients observed under conditions of standard clinical practice. *Ann N Y Acad Sci*. 2009;1173:837–846.
47. Du Pan SM, Dehler S, Ciurea A, et al; Swiss Clinical Quality Management Physicians. Comparison of drug retention rates and causes of drug discontinuation between anti-tumor necrosis factor agents in rheumatoid arthritis. *Arthritis Rheum*. 2009;61(5):560–568.
48. Iannone F, Ferraccioli G, Gremese E, Lapadula G; for the Group Italiano di Studio sulle Early Arthritis (GISEA). Drug survival on TNF inhibitors: 2003–2004 data from Italian National Register (GISEA Registry). *Ann Rheum Dis*. Epub December 24, 2013.
49. Leffers HCB, Østergaard M, Glintborg B, et al. Three-year drug survival and effectiveness of Abatacept and Tocilizumab in patients with rheumatoid arthritis treated in routine care. Results from the Nationwide Danish Danbio Registry. *Arthritis Rheum*. 2013;65(Suppl 10):S612.
50. Hetland ML, Christensen IJ, Tarp U, et al; All Departments of Rheumatology in Denmark. Direct comparison of treatment responses, remission rates, and drug adherence in patients with rheumatoid arthritis treated with adalimumab, etanercept, or infliximab: results from eight years of surveillance of clinical practice in the nationwide Danish DANBIO registry. *Arthritis Rheum*. 2010;62(1):22–32.
51. Hishitani Y, Ogata A, Shima Y, et al. Retention of tocilizumab and anti-tumour necrosis factor drugs in the treatment of rheumatoid arthritis. *Scand J Rheumatol*. 2013;42(4):253–259.
52. Sakai R, Tanaka M, Nanki T, et al; REAL Study Group. Drug retention rates and relevant risk factors for drug discontinuation due to adverse events in rheumatoid arthritis patients receiving anticytokine therapy with different target molecules. *Ann Rheum Dis*. 2012;71(11):1820–1826.
53. Yoshida K, Tokuda Y, Oshikawa H, et al. An observational study of tocilizumab and TNF-α inhibitor use in a Japanese community hospital: different remission rates, similar drug survival and safety. *Rheumatology (Oxford)*. 2011;50(11):2093–2099.
54. Strand V, Burmester GR, Ogale S, Devenport J, John A, Emery P. Improvements in health-related quality of life after treatment with tocilizumab in patients with rheumatoid arthritis refractory to tumour necrosis factor inhibitors: results from the 24-week randomized controlled RADIATE study. *Rheumatology (Oxford)*. 2012;51(10):1860–1869.
55. Fusama M, Nakahara H, Hamano Y, et al. Improvement of health status evaluated by Arthritis Impact Measurement Scale (AIMS-2) and Short Form-36 (SF-36) in patients with rheumatoid arthritis treated with tocilizumab. *Mod Rheumatol*. 2013;23(2):276–283.
56. Feist E, Rubbert-Roth A, Kaufmann J, et al. Effectiveness after 4 and 24 weeks and safety of the novel interleukin-6 receptor inhibitors tocilizumab (TCZ) in patients with active rheumatoid arthritis (RA)–analysis of the patient-reported outcomes (PRO) in the TAMARA study. *Ann Rheum Dis*. 2010;69(Suppl 3):544.
57. Chauffier K, Salliot C, Berenbaum F, et al. Effect of biotherapies on fatigue in rheumatoid arthritis: a systematic review of the literature and meta-analysis. *Rheumatology (Oxford)*. 2012;51(1):60–68.
58. Strand V, Rentz AM, Cifaldi MA, Chen N, Roy S, Revicki D. Health-related quality of life outcomes of adalimumab for patients with early rheumatoid arthritis: results from a randomized multicenter study. *J Rheumatol*. 2012;39(1):63–72.
59. Callhoff J, Weiß A, Zink A, Listing J. Impact of biologic therapy on functional status in patients with rheumatoid arthritis – a meta-analysis. *Rheumatology (Oxford)*. 2013;52(12):2127–2135.
60. Huynh T, Østergaard A, Egsmose C, et al. Patients and health professional preferences to route of administration of biologics agents for RA treatment. *Ann Rheum Dis*. 2013;72(Suppl 3):431.
61. Burmester GR, Rubbert-Roth A, Cantagrel A, et al. A randomised, double-blind, parallel-group study of the safety and efficacy of subcutaneous tocilizumab versus intravenous tocilizumab in combination with traditional disease-modifying antirheumatic drugs in patients with moderate to severe rheumatoid arthritis (SUMMACTA study). *Ann Rheum Dis*. 2014;73(1):69–74.
62. Ogata A, Tanimura K, Sugimoto T, et al. A phase 3 study of the efficacy and safety of subcutaneous versus intravenous tocilizumab monotherapy in patients with rheumatoid arthritis (MUSASHI). *Arthritis Care Res (Hoboken)*. Epub August 27, 2013.
63. Tsao NW, Bansback NJ, Shojania K, Marra CA. The issue of comparators in economic evaluations of biologic response modifiers in rheumatoid arthritis. *Best Pract Res Clin Rheumatol*. 2012;26(5):659–676.
64. Hallert E, Husberg M, Kalkan A, Skogh T, Bernfort L. Early rheumatoid arthritis 6 years after diagnosis is still associated with high direct costs and increasing loss of productivity: the Swedish TIRA project. *Scand J Rheumatol*. Epub December 19, 2013.
65. Soini EJ, Hallinen TA, Puolakka K, Vihervaara V, Kauppi MJ. Cost-effectiveness of adalimumab, etanercept, and tocilizumab as first-line treatments for moderate-to-severe rheumatoid arthritis. *J Med Econ*. 2012;15(2):340–351.
66. Diamantopoulos A, Benucci M, Capri S, et al. Economic evaluation of tocilizumab combination in the treatment of moderate-to-severe rheumatoid arthritis in Italy. *J Med Econ*. 2012;15(3):576–585.
67. Navarro Sarabia F, Blanco FJ, Alvaro Gracia JM, et al. [Economic evaluation of rheumatoid arthritis monotherapy with tocilizumab and adalimumab]. *Rev Esp Salud Publica*. 2013;87(4):343–350. Spanish.
68. Ngo P, Sorathia RC, Todd C, Jones G, Nash P. Australian economic evaluation of tocilizumab (TCZ) monotherapy vs adalimumab (ADA) monotherapy in patients with rheumatoid arthritis (RA) based on the ADACTA trial. *Internal Medicine Journal*. 2013;43(12):12.
69. Papadakis K, McNamara S, Wintfeld N, et al. A UK cost-effectiveness analysis on the use of tocilizumab (Roactemra®) with methotrexate in the treatment of adults with moderate to severe active rheumatoid arthritis who have failed to respond adequate to one or more DMARDs. *Ann Rheum Dis*. 2010;69(Suppl 3):389.

70. McNamara SK, Papadakis KN, Wintfeld N, et al. An economic evaluation on the use of tocilizumab (Roactemra®) with methotrexate in the treatment of adults with moderate to severe active rheumatoid arthritis who have failed to respond adequately to an anti-TNF drugs in Scotland. *Ann Rheum Dis*. 2010;69(Suppl 3):389.
71. Tanaka E, Inoue E, Hoshi D, et al. Cost-effectiveness of a humanized anti-interereleukin-6 (IL-6) receptor monoclonal antibody, tocilizumab, in rheumatoid arthritis using IORRA cohort database. *Ann Rheum Dis*. 2012;71(Suppl 3):668.
72. Smolen JS, Landewe R, Breedveld FC, et al. EULAR recommendations for the management of rheumatoid arthritis with synthetic and biological disease-modifying antirheumatic drugs: 2013 update. *Ann Rheum Dis*. Epub October 25, 2013.
73. Ogata A, Hirano T, Hishitani Y, Tanaka T. Safety and efficacy of tocilizumab for the treatment of rheumatoid arthritis. *Clin Med Insights Arthritis Musculoskelet Disord*. 2012;5:27–42.
74. Fujimoto M, Serada S, Mihara M, et al. Interleukin-6 blockade suppresses autoimmune arthritis in mice by the inhibition of inflammatory Th17 responses. *Arthritis Rheum*. 2008;58(12):3710–3719.
75. Kagari T, Doi H, Shimozato T. The importance of IL-1 beta and TNF-alpha, and the noninvolvement of IL-6, in the development of monoclonal antibody-induced arthritis. *J Immunol*. 2002;169(3):1459–1466.
76. Tanaka T. Can IL-6 blockade rectify imbalance between Tregs and Th17 cells? *Immunotherapy*. 2013;5(7):695–697.
77. Samson M, Audia S, Janikashvili N, et al. Brief report: inhibition of interleukin-6 function corrects Th17/Treg cell imbalance in rheumatoid arthritis patients. *Arthritis Rheum*. 2012;64(8):2499–2503.
78. Pesce B, Soto L, Sabugo F, et al. Effect of interleukin-6 receptor blockade on the balance between regulatory T cells and T helper type 17 cells in rheumatoid arthritis patients. *Clin Exp Immunol*. 2013;171(3):237–242.
79. Nie H, Zheng Y, Li R, et al. Phosphorylation of FOXP3 controls regulatory T cell function and is inhibited by TNF-a in rheumatoid arthritis. *Nat Med*. 2013;19(3):322–328.
80. Carbone G, Wilson A, Diehl SA, Bunn J, Cooper SM, Rincon M. Interleukin-6 receptor blockade selectively reduces IL-21 production by CD4 T cells and IgG4 autoantibodies in rheumatoid arthritis. *Int J Biol Sci*. 2013;9(3):279–288.
81. Roll P, Muhammad K, Schumann M, et al. In vivo effects of the anti-interleukin-6 receptor inhibitor tocilizumab on the B cell compartment. *Arthritis Rheum*. 2011;63(5):1255–1264.
82. Muhammad K, Roll P, Seibold T, et al. Impact of IL-6 receptor inhibition on human memory B cells in vivo: impaired somatic hypermutation in preswitch memory B cells and mutational targeting in memory B cells. *Ann Rheum Dis*. 2011;70(8):1507–1510.
83. Nemeth E, Rivera S, Gabayan V, et al. IL-6 mediates hypoferremia of inflammation by inducing the synthesis of the iron regulatory hormone hepcidin. *J Clin Invest*. 2004;113(9):1271–1276.
84. Song SN, Iwahashi M, Tomosugi N, et al. Comparative evaluation of the effects of treatment with tocilizumab and TNF-α inhibitors on serum hepcidin, anemia response and disease activity in rheumatoid arthritis patients. *Arthritis Res Ther*. 2013;15(5):R141.
85. Obici L, Merlini G. AA amyloidosis: basic knowledge, unmet needs and future treatments. *Swiss Med Wkly*. 2012;142:w13580.
86. Laiho K, Tiitinen S, Kaarela K, Helin H, Isomaki H. Secondary amyloidosis has decreased in patients with inflammatory joint disease in Finland. *Clin Rheumatol*. 1999;18(2):122–123.
87. Immonen K, Finne P, Gronhagen-Riska C, et al. A marked decline in the incidence of renal replacement therapy for amyloidosis associated with inflammatory rheumatic diseases – data from nationwide registries in Finland. *Amyloid*. 2011;18(1):25–28.
88. Tanaka T, Hagihara K, Hishitani Y, Ogata A. Tocilizumab for the treatment of AA amyloidosis. In: Guvenc IA, editor. *Amyloidosis – An Insight to Disease of Systems and Novel Therapies*. Croatia: INTECH Open Access Publisher; 2011:155–170.
89. Okuda Y, Ohnishi M, Matoba K, et al. Comparison of the clinical utility of tocilizumab and anti-TNF therapy in AA amyloidosis complicating rheumatic diseases. *Mod Rheumatol*. 2014;24(1):137–143.
90. Nishida S, Hagihara K, Shima Y, et al. Rapid improvement of AA amyloidosis with humanised anti-interleukin 6 receptor antibody treatment. *Ann Rheum Dis*. 2009;68(7):1235–1236.
91. Hakala M, Immonen K, Korpela M, Vasala M, Kauppi MJ. Good medium-term efficacy of tocilizumab in DMARD and anti-TNF-α therapy resistant reactive amyloidosis. *Ann Rheum Dis*. 2013;72(3):464–465.
92. Miyagawa I, Nakayamada S, Saito K, et al. Study on the safety and efficacy of tocilizumab, an anti-IL-6 receptor antibody, in patients with rheumatoid arthritis complicated with AA amyloidosis. *Mod Rheumatol*. Epub October 21, 2013.

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# IL-6 in Inflammation, Immunity, and Disease

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# IL-6 in Inflammation, Immunity, and Disease

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Interleukin 6 (IL-6), promptly and transiently produced in response to infections and tissue injuries, contributes to host defense through the stimulation of acute phase responses, hematopoiesis, and immune reactions. Although its expression is strictly controlled by transcriptional and posttranscriptional mechanisms, dysregulated continual synthesis of IL-6 plays a pathological effect on chronic inflammation and autoimmunity. For this reason, tocilizumab, a humanized anti-IL-6 receptor antibody was developed. Various clinical trials have since shown the exceptional efficacy of tocilizumab, which resulted in its approval for the treatment of rheumatoid arthritis and juvenile idiopathic arthritis. Moreover, tocilizumab is expected to be effective for other intractable immune-mediated diseases. In this context, the mechanism for the continual synthesis of IL-6 needs to be elucidated to facilitate the development of more specific therapeutic approaches and analysis of the pathogenesis of specific diseases.

IL-6 is a soluble mediator with a pleiotropic effect on inflammation, immune response, and hematopoiesis. At first, distinct functions of IL-6 were studied and given distinct names based on their biological activity. For example, the name B-cell stimulatory factor 2 (BSF-2) was based on the ability to induce differentiation of activated B cells into antibody (Ab)-producing cells (Kishimoto 1985), the name hepatocyte-stimulating factor (HSF) on the effect of acute phase protein synthesis on hepatocytes, the name hybridoma growth factor (HGF) on the enhancement of growth of fusion cells between plasma cells and myeloma cells, or the name interferon (IFN)-β2 owing to its IFN antiviral activity. When the BSF-2 cDNA was successfully cloned in 1986 (Hirano et al. 1986), however, it was found that the molecules with different names studied by various groups were in fact identical, resulting in the single name IL-6 (Kishimoto 1989). Human IL-6 is made up of 212 amino acids, including a 28-amino-acid

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signal peptide, and its gene has been mapped to chromosome 7p21. Although the core protein is ~20 kDa, glycosylation accounts for the size of 21–26 kDa of natural IL-6.

## BIOLOGICAL EFFECT OF IL-6 ON INFLAMMATION AND IMMUNITY

After IL-6 is synthesized in a local lesion in the initial stage of inflammation, it moves to the liver through the bloodstream, followed by the rapid induction of an extensive range of acute phase proteins such as C-reactive protein (CRP), serum amyloid A (SAA), fibrinogen, haptoglobin, and α1-antichymotrypsin (Fig. 1) (Heinrich et al. 1990). On the other hand, IL-6 reduces the production of fibronectin, albumin, and transferrin. These biological effects on hepatocytes were at first studied as belonging to HSF. When high-level concentrations of SAA persist for a long time, it leads to a serious complication of several chronic inflammatory diseases through the generation of amyloid A amyloidosis (Gillmore et al. 2001). This results in amyloid fibril deposition, which causes progressive deterioration in various organs. IL-6 is also involved in the regulation of serum iron and zinc levels via control of their transporters. As for serum iron, IL-6 induces hepcidin production, which blocks the action of iron transporter ferroportin 1 on gut and, thus, reduces serum iron levels (Nemeth et al. 2004). This means that the IL-6-hepcidin axis is responsible for hypoferremia and anemia associated with chronic inflammation. IL-6 also enhances zinc importer ZIP14 expression on hepatocytes and so induces hypozincemia seen in inflammation (Liuzzi et al. 2005). When IL-6 reaches the bone marrow, it promotes megakaryocyte maturation, thus leading to the release of platelets (Ishibashi et al. 1989). These changes in acute phase protein levels and red blood cell and platelet counts are used for the evaluation of inflammatory severity in routine clinical laboratory examinations.

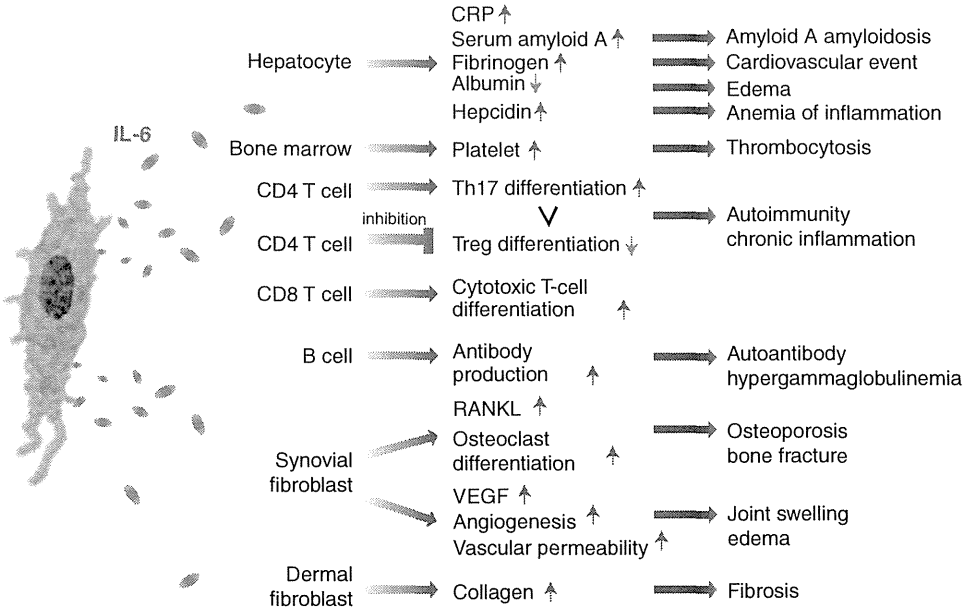


Figure 1. IL-6 in inflammation, immunity, and disease. IL-6 is a cytokine featuring pleiotropic activity; it induces synthesis of acute phase proteins such as CRP, serum amyloid A, fibrinogen, and hepcidin in hepatocytes, whereas it inhibits production of albumin. IL-6 also plays an important role on acquired immune response by stimulation of antibody production and of effector T-cell development. Moreover, IL-6 can promote differentiation or proliferation of several nonimmune cells. Because of the pleiotropic activity, dysregulated continual production of IL-6 leads to the onset or development of various diseases. Treg, regulatory T cell; RANKL, receptor activator of nuclear factor κB (NF-κB) ligand; VEGF, vascular endothelial growth factor.

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Furthermore, IL-6 promotes specific differentiation of naïve $CD4^+$ T cells, thus performing an important function in the linking of innate to acquired immune response. It has been shown that IL-6, in combination with transforming growth factor (TGF)-β, is indispensable for Th17 differentiation from naïve $CD4^+$ T cells (Korn et al. 2009), but that IL-6 also inhibits TGF-β-induced Treg differentiation (Bettelli et al. 2006). Up-regulation of the Th17/Treg balance is considered to be responsible for the disruption of immunological tolerance, and is thus pathologically involved in the development of autoimmune and chronic inflammatory diseases (Kimura and Kishimoto 2010). It has been further shown that IL-6 also promotes T-follicular helper-cell differentiation as well as production of IL-21 (Ma et al. 2012), which regulates immunoglobulin (Ig) synthesis and IgG4 production in particular. IL-6 also induces the differentiation of $CD8^+$ T cells into cytotoxic T cells (Okada et al. 1988). Under one of its previous names, BSF-2, IL-6 was found to be able to induce the differentiation of activated B cells into Ab-producing plasma cells, so that continuous oversynthesis of IL-6 results in hypergammaglobulinemia and autoantibody production.

IL-6 exerts various effects other than those on hepatocytes and lymphocytes and these are frequently detected in chronic inflammatory diseases (Kishimoto 1989; Hirano et al. 1990; Akira et al. 1993). One of these effects is that, when IL-6 is generated in bone marrow stromal cells, it stimulates the RANKL (Hashizume et al. 2008), which is indispensable for the differentiation and activation of osteoclasts (Kotake et al. 1996), and this leads to bone resorption and osteoporosis (Poli et al. 1994). IL-6 also induces excess production of VEGF, leading to enhanced angiogenesis and increased vascular permeability, which are pathological features of inflammatory lesions and are seen in, for example, synovial tissues of rheumatoid arthritis (RA) or edema of remitting seronegative symmetrical synovitis with pitting edema (RS3PE) syndrome (Nakahara et al. 2003; Hashizume et al. 2009). Finally, it has been reported that IL-6 aids keratinocyte proliferation (Grossman et al. 1989) or the generation of collagen in dermal fibroblasts that may account for changes in the skin of patients with systemic sclerosis (Duncan and Berman 1991).

## REGULATION OF IL-6 SYNTHESIS

IL-6 functions as a mediator for notification of the occurrence of some emergent event. IL-6 is generated in an infectious lesion and sends out a warning signal to the entire body. The signature of exogenous pathogens, known as pathogen-associated molecular patterns, is recognized in the infected lesion by pathogen-recognition receptors (PRRs) of immune cells such as monocytes and macrophages (Kumar et al. 2011). These PRRs comprise Toll-like receptors (TLRs), retinoic acid-inducible gene-1-like receptors, nucleotide-binding oligomerization domain-like receptors, and DNA receptors. They stimulate a range of signaling pathways including NF-κB, and enhance the transcription of the mRNA of inflammatory cytokines such as IL-6, tumor necrosis factor (TNF)-α, and IL-1β. TNF-α and IL-1β also activate transcription factors to produce IL-6.

IL-6 also issues a warning signal in the event of tissue damage. Damage-associated molecular patterns (DAMPs), which are released from damaged or dying cells in noninfectious inflammations such as burn or trauma, directly or indirectly promote inflammation. During sterile surgical operations, an increase in serum IL-6 levels precedes elevation of body temperature and serum acute phase protein concentration (Nishimoto et al. 1989). DAMPs from injured cells contain a variety of molecules such as mitochondrial (mt) DNA, high mobility group box 1 (HMGB1), and S100 proteins (Bianchi 2007). Serum mtDNA levels in trauma patients are thousands of times higher than in controls and this elevation leads to TLR9 stimulation and NF-κB activation (Zhang et al. 2010), whereas binding of HMGB1 to TLR2, TLR4, and the receptor of advanced glycation end products (RAGE) can promote inflammation. The S100 family of proteins comprises more than 25 members, some of which also interact with RAGE to evoke sterile inflammation (Sims et al. 2010).

In addition to immune-mediated cells, mesenchymal cells, endothelial cells, fibroblasts, and many other cells are involved in the production of IL-6 in response to various stimuli (Akira et al. 1993). The fact that IL-6 issues a warning signal to indicate occurrence of an emergency accounts for the strict regulation of IL-6 synthesis both gene transcriptionally and posttranscriptionally. A number of transcription factors have been shown to regulate the IL-6 gene transcription (Fig. 2). The functional *cis*-regulatory elements in the human IL-6 gene 5′ flanking region are found binding sites for NF-κB, specificity protein 1 (SP1), nuclear factor IL-6 (NF-IL-6) (also known as CAAT/enhancer-binding protein β), activator protein 1 (AP-1), and interferon regulatory factor 1 (Libermann and Baltimore 1990; Akira and Kishimoto 1992; Matsusaka et al. 1993). Activation of *cis*-regulatory elements by stimulation with IL-1, TNF, TLR-mediated signal, and forskolin lead to activation of the IL-6 promoter.

A polymorphism at position -174 of the IL-6 promoter region is reportedly associated with systemic onset juvenile idiopathic arthritis (Fishman et al. 1998) and susceptibility to RA in Europeans (Lee et al. 2012). Stimulation with lipopolysaccharide (LPS) and IL-1 did not evoke any response in a reporter assay using -174 C construct. A -174 G construct, on the other hand, was found to promote transcription of the reporter gene, suggesting that a genetic back-

| | Protein | MicroRNA |
|---|---|---|
| Promotion | NF-κB, SP-1<br>NF-IL-6, AP-1<br>IRF1, Tax<br>TAT, HBVX<br>Mutant p53 | |
| Repression | Ahr,GR, ER<br>p53, Rb<br>PPARα | miR-155 (targeting NF-IL-6)<br>miR-146a/b (targeting IRAK1)<br>miR-223 (targeting STAT3) |

**Transcriptional regulation**

| | Protein | MicroRNA |
|---|---|---|
| Stabilization | Arid5a<br>P38α<br>ORF57 | |
| Degradation | Regnase-1<br>TTP<br>BRF1<br>BRF2 | miR-365<br>miR-608 |

**Posttranscriptional regulation**

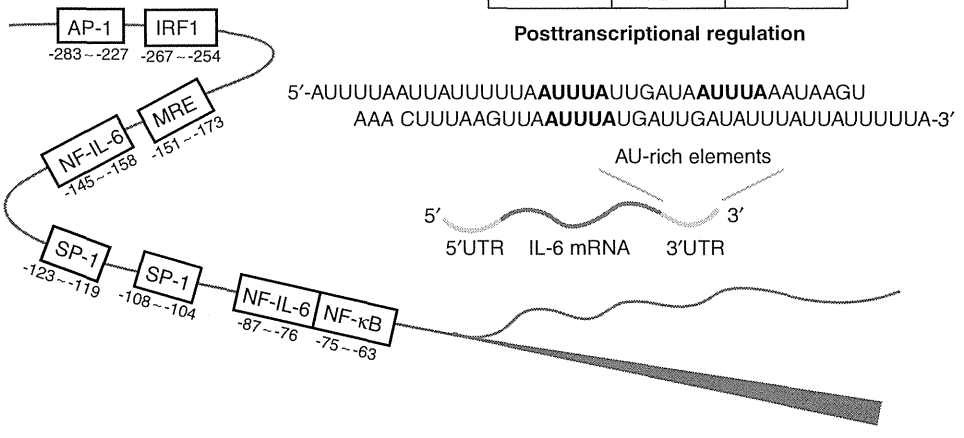


Figure 2. Transcriptional and posttranscriptional regulation of IL-6 gene. The expression and degradation of IL-6 mRNA is regulated transcriptionally and posttranscriptionally by several proteins and microRNAs. Activation of these proteins and microRNAs determines the fate of IL-6 mRNA. NF-IL-6, nuclear factor of IL-6; Tax, transactivator protein; TAT, transactivator of the transcription; HBVX, hepatitis B virus X protein; Ahr, aryl hydrocarbon receptor; GR, glucocorticoid receptor; ER, estrogen receptor; Rb, retinoblastoma; PPARα, peroxisome proliferator–activated receptor α; miR, microRNA; IRAK1, IL-1 receptor–associated kinase 1; STAT3, signal transducer and activator of transcription 3; ORF, open reading frame; TTP, tristetraprolin; BRF1, butyrate response factor 1.

ground of excess IL-6 production constitutes a risk factor for juvenile idiopathic arthritis and RA.

An interesting finding is that some viral products enhance the DNA-binding activity of NF-κB and NF-IL-6, resulting in an increase in IL-6 mRNA transcription. An instance of this phenomenon is that interaction with NF-κB of the Tax derived from the human T lymphotropic virus 1 enhances IL-6 production (Ballard et al. 1988; Leung and Nabel 1988). Another example is the enhancement of both NF-κB and NF-IL-6 DNA-binding activity by the transactivator of the TAT protein of the human immunodeficiency virus 1 (Scala et al. 1994; Ambrosino et al. 1997). Moreover, it has been shown that DNA binding of NF-IL-6 can be enhanced by the human hepatitis B virus X protein (Mahe et al. 1991; Ohno et al. 1999).

On the other hand, some transcription factors suppress IL-6 expression. Peroxisome proliferator–activated receptors (PPARs) are ligand-activated transcription factors consisting of three subtypes: α, β, and γ. Among three PPARs, fibrates-activated PPARα interacts with c-Jun and p65 NF-κB subunits, which negatively regulate IL-6 transcription (Delerive et al. 1999). In addition, some hormone receptors have been identified as repressors of IL-6 expression. The increase in serum IL-6 after menopause or ovarectomy is reportedly associated with suppression of IL-6 expression by estrogen receptors (Jilka et al. 1992), whereas activation of the glucocorticoid receptor can repress IL-6 expression, and this is thought to be one of mechanisms responsible for the anti-inflammatory effects of corticosteroids (Ray and Prefontaine 1994). It has further been shown that retinoblastoma protein and p53 repress the IL-6 gene promoter, whereas it is up-regulated by mutant p53 (Santhanam et al. 1991).

In addition, some microRNAs directly or indirectly regulate transcription activity. Interaction of microRNA-155 with the 3′ untranslated regions (UTR) of NF-IL-6 results in suppression of NF-IL-6 expression (He et al. 2009), whereas microRNA-146a/b and -223 indirectly suppress transcription of IL-6 by respectively targeting IL-1 receptor–associated kinase 1 and STAT3 (Chen et al. 2012; Zilahi et al. 2012).

## PRODUCTION AND FUNCTION OF IL-6 AND ARYL HYDROCARBON RECEPTOR

Aryl hydrocarbon receptor (Ahr) not only affects IL-6 transcription, but also regulates innate and acquired immune response. Ahr, also known as the dioxin receptor, is a ligand-activated transcription factor that belongs to the basic helix-loop-helix PER-ARNT-SIM family (Burbach et al. 1992; Ema et al. 1992). Ahr is present in the cytoplasm, where it forms a complex with Ahr-interacting protein (Bell and Poland 2000). On binding with a ligand, Ahr moves to the nucleus and dimerizes with the Ahr nuclear translocator (Arnt). Within the nucleus, the Ahr/Arnt heterodimer then binds to the xenobiotic response element (XRE), which leads to various toxicological effects (Fujii-Kuriyama et al. 1994; Dragan and Schrenk 2000; Ohtake et al. 2003; Puga et al. 2005). Although the physiological ligands for Ahr are not well known, indoleamine 2,3-dioxygenase (IDO), which catalyzes tryptophan into kynurenine, is induced by Ahr signaling and kynurenine is one of the ligands of Ahr (Vogel et al. 2008; Jux et al. 2009).

An animal model of RA was used to show the essential role of Ahr in the induction of Th17 cells and Th17-dependent collagen-induced arthritis (CIA) (Kimura et al. 2008; Nakahama et al. 2011). Stimulation of naïve T cells with IL-6 plus TGF-β (Th17 cell–inducing condition) induced Ahr expression and deletion of the Ahr gene nullified the induction of Th17 cells.

Ahr interacted with and inhibited the activities of STAT1 or STAT5, which mediate the anti-inflammatory signals of IL-27 and IFN-γ, or IL-2, respectively (Harrington et al. 2005; Stumhofer et al. 2006; Laurence et al. 2007; Kimura et al. 2008), thus suppressing the inhibitory signals for the induction of Th17 cells. Retinoid-related orphan receptors (ROR) γ and α, which are activated by STAT3, are essential transcription factors for Th17-cell induction (Ivanov et al. 2006; Yang et al. 2008) and Ahr was

found not to affect the ROR-γ and -α expression. In the Ahr gene–deficient mice, no arthritis developed in CIA. Moreover, with T-cell-specific deletion of the Ahr gene, no development of CIA was observed (Nakahama et al. 2011). These results clearly show that CIA is a T-cell-dependent disease and the presence of Ahr is essential for its development. In these Ahr-deficient mice, the number of Th17 cells decreased and that of Th1 cells increased but no significant changes were observed in Foxp3-expressing Treg cells.

Ahr also regulates Th17-cell induction through regulation of microRNAs. In our study, Ahr-induced microRNA-132/212 cluster under Th17 cell–inducing conditions and transfection of microRNA-212 into naïve T cells under these circumstances augmented the expression of IL-17-related genes such as IL-17A, IL-22, and IL-23R (Nakahama et al. 2013). One of the target genes of this microRNA is B-cell lymphoma 6, which is known as an inhibitor of Th17-cell induction (Yu et al. 2009). All of these findings show that Ahr accelerates inflammation through the enhancement of Th17-cell induction by several mechanisms.

Interestingly, Ahr showed a negative regulatory effect on peritoneal macrophages and bone marrow–derived dendritic cells (BMDC) (Nguyen et al. 2010). In the absence of Ahr, LPS-induced production of inflammatory cytokines such as IL-6, TNF, and IL-12 showed major increases in macrophages, indicating that Ahr negatively regulates inflammatory cytokine production. Ahr interacts with STAT1 and NF-κB and the resultant complex of Ahr/STAT1 and NF-κB leads to inhibition of the promoter activity of IL-6 and other inflammatory cytokines (Kimura et al. 2009). In BMDC, Ahr is required for the activation of IDO leading to kynurenine production because the deletion of Ahr in BMDC leads to loss of IL-10 and kynurenine production. Coculture of naïve T cells with Ahr-deficient BMDC in the presence of LPS resulted in reduction of Treg-cell induction, whereas addition of kynurenine rescued the induction of Treg cells by BMDC (Nguyen et al. 2010). These findings indicate that Ahr is required for the regulatory BMDC cells through the induction of IDO.

## STABILIZATION AND DEGRADATION OF IL-6 MRNA (ARID5A AND REGNASE-1)

As for posttranscriptional regulation of cytokine expression, cytokine mRNA is controlled through both the 5′ and 3′UTR (Chen and Shyu 1995; Anderson 2008). Initiation of mRNA translation is determined by the 5′UTR, and the stability of mRNA by the 3′UTR. IL-6 mRNA is regulated by modulation of AU-rich elements located in the 3′UTR region, whereas a number of RNA-binding proteins and microRNAs bind to the 3′UTRs and regulate the stability of IL-6 mRNA (Fig. 2). For example, IL-6 mRNA stabilization is promoted by mitogen-activated protein kinase (MAPK) p38α via 3′UTRs of IL-6 (Zhao et al. 2008), and the stabilization of both viral and human IL-6 mRNA by the Kaposi's sarcoma–associated herpesvirus (KSHV) ORF-57 by competing with the binding of microRNA-1293 to the viral or of microRNA-608 to the human IL-6 mRNA (Kang et al. 2011). RNA-binding proteins, such as TTP and BRF1 and 2, on the other hand, promote IL-6 mRNA degradation (Palanisamy et al. 2012), whereas IL-6 mRNA levels are reduced by microRNAs such as microRNA-365 and -608 through direct interaction with IL-6 3′UTR (Kang et al. 2011; Xu et al. 2011).

It was recently found that a nuclease known as regulatory RNase-1 (regnase-1) (also known as Zc3h12a) plays a part in the destabilization of IL-6 mRNA, and that the relevant knockout mice spontaneously develop autoimmune diseases accompanied by splenomegaly and lymphadenopathy (Matsushita et al. 2009). The inhibitor of NF-κB (IκB) kinase (IKK) complex controls IL-6 mRNA stability by phosphorylating regnase-1 in response to IL-1R/TLR stimulation (Iwasaki et al. 2011). Phosphorylated regnase-1 underwent ubiquitination and degradation. Regnase-1 re-expressed in IL-1R/TLR-activated cells was found to feature delayed kinetics, and regnase-1 mRNA to be negatively regulated by regnase-1 itself via a stem-loop region present in the regnase-1 3′UTR. These findings show that IKK complex phosphorylates not only IκBα, activating transcription, but also regnase-1, releasing the brake on IL-6 mRNA ex-

pression. Regnase-1 also regulates the mRNAs of a set of genes, including c-Rel, Ox40, and IL-2 through cleavage of their 3′UTRs in T cells. T-cell receptor engagement then leads to cleavage of regnase-1, which frees T cells from regnase-mediated suppression, thus indicating that regnase-1 may play a crucial role in T-cell activation (Uehata et al. 2013).

We have recently identified a novel RNA-binding protein, AT-rich interactive domain-containing protein 5a (Arid5a), which binds to the 3′UTR of IL-6 mRNA, resulting in the selective stabilization of IL-6 but not of TNF-α or IL-12 mRNA (Masuda et al. 2013). Arid5a expression was found to be enhanced in macrophages in response to LPS, IL-1β, and IL-6, and also to be induced under Th17-polarizing conditions in T cells. We also found that Arid5a gene deficiency inhibited elevation of IL-6 levels in LPS-injected mice and preferential Th17-cell development in experimental autoimmune encephalomyelitis. Moreover, Arid5a counteracted the destabilizing function of regnase-1 on IL-6 mRNA (Fig. 3), indicating that the balance between Arid5a and regnase-1 plays an important role in IL-6 mRNA stability. All of these results suggest that posttranscriptional regulation of IL-6 mRNA by Arid5a and regnase-1 may play an important role in the expression of IL-6 and that the predominance of Arid5a over regnase-1 promotes inflammatory processes and possibly induces the development of autoimmune inflammatory diseases.

During the so-called "cytokine storm," a potentially fatal immune reaction induced by hyperactivation of T cells, a major boost in IL-6 production is observed but without comparable production of other inflammatory cytokines. A recent study showed that the cytokine storm induced by cancer immunotherapy using T-cell transfection was counteracted by the anti-IL-6 receptor antibody, tocilizumab (Grupp et al. 2013). Experimentally, inhalation by mice of peroxidized phospholipids induced a cytokine storm resulting from a greatly marked increase in the production of IL-6 but not TNF (Imai et al. 2008).

These results showing the IL-6-specific elevation without any effect on the other inflammatory cytokines strongly suggest the impor-

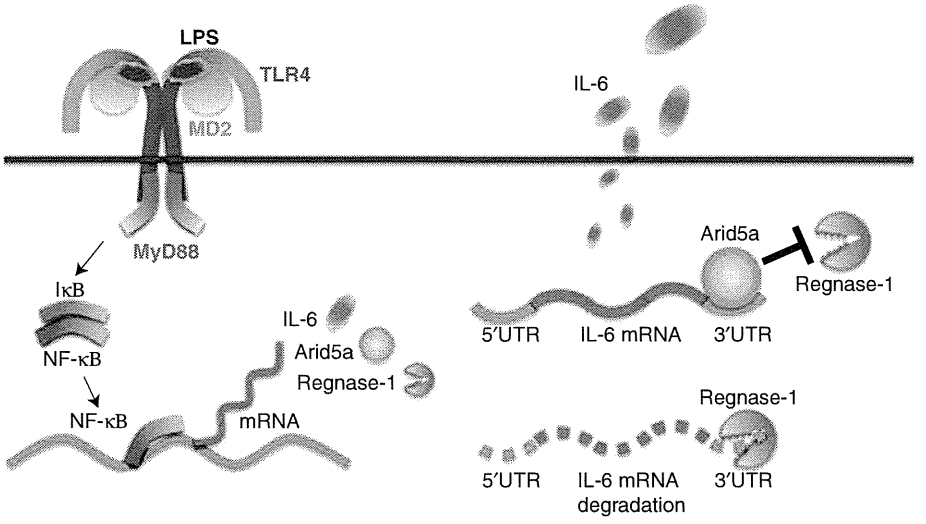


Figure 3. IL-6 synthesis and regulation of IL-6 mRNA stability by Arid5a. Pathogen-associated molecular patterns are recognized by pathogen-recognition receptors to induce proinflammatory cytokines; in this figure, TLR4 recognizes LPS and induces IL-6 mRNA via activation of the NF-κB signaling pathway. Regnase-1 promotes IL-6 mRNA degradation, whereas Arid5a inhibits destabilizing effects of regnase-1. The balance between Arid5a and regnase-1 is important for the regulation of IL-6 mRNA. MD2, myeloid differentiation protein 2; MyD88, myeloid differentiation primary response 88; IκB, inhibitor of NF-κB.