Tumor Necrosis Factor Antagonists: Different Kinetics and/or Mechanisms of Action May Explain Differences in the Risk for Developing Granulomatous Infection

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Objective Tumor necrosis factor (TNF) antagonists fall into 2 classes:etanercept (ETA) is a soluble TNF receptor, while infliximab (INF) and adalimumab (ADA) are monoclonal antibodies against TNF. All 3 drugs are effective in treating rheumatoid arthritis. However, these agents have been associated with an increased risk of granulomatous infections, such as tuberculosis and histoplasmosis. Several reports indicate that the incidence of granulomatous infections may potentially be higher in individuals treated with INF than ETA.

Methods We conducted a comprehensive literature search (1966 to 2004) to review the role of TNF in normal and disease states, and the mechanisms of action of the TNF inhibitors. Specifically, we searched for possible mechanisms for the apparent increase in granulomatous infections associated with TNF inhibitors and for reasons that there may be differences between them.

Results Infection may result from a number of differences between ETA and INF or ADA. First, binding avidities are different, with ETA binding in a 1:1 ratio and INF/ADA binding in 2 to 3:1 ratios. Second, the clearances of ADA, ETA, and INF are different, being about 13 times higher for ETA than INF or ADA, thus resulting in higher steady-state drug levels for ADA and INF. Also, the methods of administration are different, intravenously (for INF) versus subcutaneously (for ETA and ADA), which results in lower peak concentrations for ETA and ADA, potentially explaining some of the differences in effects on granuloma formation. Third, INF and ADA have somewhat different mechanisms of action from ETA: INF and ADA are associated with antibody-mediated cell lysis, while ETA is not; INF may induce apoptosis in some tissues (eg, gastrointestinal [GI] mucosa) while ETA does not—although this is controversial and may not be true at steady state in synovium, where both drugs seem to cause apoptosis; ETA binds lymphotoxin-α while INF does not (ETA may thus be more efficient at preventing granuloma formation by this mechanism than INF); finally, ADA and INF seem to inhibit IFN-γ expression (probably indirectly), while ETA does not.

Conclusions There are significant differences between the 2 classes of TNF antagonists in terms of both their kinetics and mechanisms of action. These differences may help explain the apparent differences in the incidence of granuloma-dependent infections among them.

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Keywords *tumor necrosis factor, TNFR-Fc fusion protein, antirheumatic agents, etanercept, infliximab, granuloma*

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Abbreviations	
AERS	adverse event reporting system
Fc	crystallizable fragment (of the antibody)
IBD	inflammatory bowel disease
IFN	interferon
IL	interleukin
MCP	monocyte chemoattractant protein
MIP	macrophage-inflammatory protein
RA	rheumatoid arthritis
RANTES	regulated on activation, normal T-cell expressed and secreted
TB	tuberculosis
TNF	tumor necrosis factor

Rheumatoid arthritis (RA) is a chronic systemic inflammatory disease that is characterized by the destruction of the synovial membranes and articular structures of multiple joints. The inflammation, swelling, pain, and loss of mobility caused by damage to the synovium greatly impact the lives of those affected by the disease. Traditional RA treatments, including nonsteroidal antiinflammatory agents and corticosteroids, can reduce the pain associated with RA but do not address the fundamental disease process. Methotrexate, the most common disease-modifying antirheumatic drug used to treat RA, is effective but is associated with occasional significant side effects (1). The discovery that high levels of tumor necrosis factor (TNF) may contribute to or mediate chronic inflammation and joint destruction in RA heralded a new era of targeted and highly effective biologics for RA and other chronic inflammatory diseases.

The TNF antagonists fall into 2 classes: monoclonal antibodies and soluble receptors. Etanercept (Enbrel®, Thousands Oaks, CA) is a soluble TNF receptor, while infliximab (Remicade®, Horsham, PA) and adalimumab (Humira®, Abbott Park, ILL) are monoclonal antibodies against TNF. These agents are now widely used for the treatment of RA and other inflammatory diseases. As of December 2004, more than 250,000 patients worldwide had been treated with etanercept (2) and more than 500,000 had been treated with infliximab (3). As of August 31, 2004, 10,050 patients with RA had enrolled in adalimumab clinical trials worldwide (4). TNF antagonists improve signs and symptoms, inhibit the progression of structural damage, and impact functional outcomes in patients with RA (5-10). Etanercept, in clinical trials, has these effects in patients with psoriatic arthritis, ankylosing spondylitis, and juvenile RA (7,9,10); infliximab has been approved for the treatment of ankylosing spondylitis and psoriatic arthritis, and adalimumab is being tested in these diseases (7,9,10). Infliximab has efficacy in treating Crohn's disease, while etanercept does not, at the doses used for RA (11,12).

TNF plays a vital role in granuloma formation and maintenance. Recent studies have reported an increase in granulomatous infections, such as tuberculosis (TB) and histoplasmosis, associated with the use of TNF antagonists (4,13-25). Several of these studies have indicated the possibility of a higher incidence of granulomatous infections associated with the use of infliximab than with etanercept, although the risk of infection is elevated with etanercept use as well (13,16). This difference may be due in part to the different mechanisms of action or pharmacokinetics of monoclonal antibodies versus the soluble receptor.

In this article, we review the role of TNF in normal and disease states. We also provide an overview of the pharmacokinetic profiles and possible mechanisms of action of infliximab, adalimumab, and etanercept, paying particular attention to the relationship between these drugs and granuloma formation and maintenance.

METHODS

We conducted several comprehensive reviews of the literature, searching the PubMed database to identify English-language articles from 1966 to 2004. We searched for articles on the role of TNF in normal and disease states using the terms *tumor necrosis factor*, *infliximab*, *etanercept*, *adalimumab*, *rheumatoid arthritis*, and *Crohn's disease*. This search was supplemented using terms related to pharmacokinetic profiles and possible mechanisms of action, including *pharmacokinetics*, *avidity*, *mechanism*, *complement*, *transmembrane*, and *apoptosis*. Granulomatous disease was included using the terms *adverse effects*, *granuloma*, *granulomatous infection*, *tuberculosis*, *histoplasmosis*, *coccidioidomycosis*, and *listeriosis*. Titles and abstracts were reviewed for articles addressing the role of TNF and potential relationships between the pharmacokinetics and/or mechanism of action of TNF inhibitors and the occurrence of granulomatous infections. Hand searches of bibliographies from relevant articles and reviews, as well as consultations with expert rheumatologists, yielded additional references.

RESULTS

TNF in Normal and in Disease States

A wide variety of cells produce TNF, including macrophages, CD4+ and CD8+ T-cells, B-cells, natural killer cells, neutrophils, endothelial cells, smooth muscle cells, osteoclasts, and fibroblasts. However, the primary sources of TNF in immunity and in inflammatory diseases are cells of the monocyte/macrophage lineage, which secrete TNF in response to exogenous molecules such as lipopolysaccharide and endogenous mediators such as interleukin (IL) 1β and interferon (IFN) γ.

TNF is expressed on the cell surface and released in soluble form following cleavage of its membrane-anchoring domains (26). A potent proinflammatory cytokine, TNF stimulates release of IFNγ, IL-1β, IL-6, IL-8, and granulocyte-macrophage colony stimulating factor and

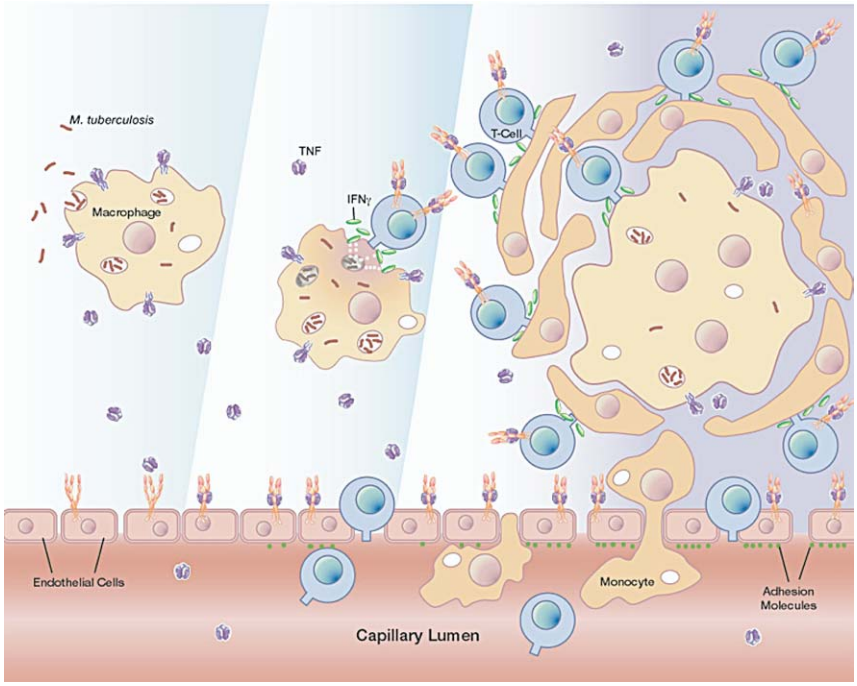

Figure 1 Granuloma formation in *M. tuberculosis* infection. Left panel: Mycobacterial components interact with toll-like receptors on macrophages triggering the production of TNF. Center panel: Secreted TNF binds to TNF receptors on endothelial cells inducing expression of adhesion molecules and chemokines leading T-cells and monocytes to leave the circulation and to migrate toward the site of infection. T-cells recognize infected cells and are stimulated with TNF leading to release of IFN-γ and the induction of the macrophage intracellular killing machinery. Right panel: Infected macrophages may fuse to form giant cells. A mature granuloma is formed with epithelioid macrophages and T-cells, effectively isolating the infection from the host. Continued secretion of TNF is required to maintain granuloma architecture.

induces production of endothelial adhesion molecules (intercellular adhesion molecule-1, vascular adhesion molecule-1, E-selectin) and chemokines (monocyte chemoattractant protein [MCP]-1, macrophage-inflammatory protein [MIP]-2, and MIP-1α), leading to transport and directed migration of leukocytes (27). In its role in resistance to infections, TNF activates neutrophils and enhances macrophage and natural killer cell killing.

In healthy humans, circulating TNF is not detectable ($<$10 fg/mL). However, in patients with inflammatory diseases such as RA (28), inflammatory bowel disease (IBD) (29), and bacterial meningitis (30), TNF can be readily detected. In septic shock, plasma TNF levels can be extremely high, in the range of 50 to 100 pg/mL or higher (30,31). Increased expression of TNF has been noted in the synovium of RA patients (32). In IBD patients, elevated TNF levels are also found in both serum and stool, and other proinflammatory cytokines have been detected in the colonic mucosa (29,33). The central pathogenic role of TNF in these and other inflammatory diseases is supported by the clinical efficacy of TNF antagonists.

TNF and the Granulomatous Response to Infection

Granulomas are cell collections composed of epithelioid macrophages and multinucleated giant cells that are encircled by lymphocytes and frequently have necrotic debris in their centers (34). They result from the protective mechanisms expressed when acute inflammatory processes cannot destroy invading agents (35). Granuloma formation is not restricted to a single type of pathogen, as the granulomatous responses to mycobacteria, fungi, protozoa, and some bacteria show important similarities. The process, which is best understood in the case of *Mycobacterium tuberculosis*, is depicted in Fig. 1. The initial cytokine response to *M. tuberculosis* (TNF, IL-12, other cytokines, and chemokines) is triggered by direct interactions of mycobacterial proteins and lipoproteins with toll-like receptors of lung macrophages (36,37). The cascade of events that follows serves first to stimulate a response comprising neutrophils and natural killer cells (38). In most individuals, however, this initial innate immune response is insufficient to control mycobacterial replication. As a result, spread of the infection to regional lymph nodes and transient hematogenous dissemination commonly occur before the infection is ultimately contained by an adaptive immune response, composed of $\gamma\delta$ T-cells, CD1-restricted cells, and later, CD4+ and CD8+ T-cells (39-43). The sequential migration of these cells to the site of infection results in the formation of a mature granuloma.

TNF plays an essential role in this process. Animal models demonstrate that TNF deficiency increases sus-

ceptibility to granulomatous infections (44-47). Neutralization of TNF decreases both the recruitment of inflammatory cells and the formation of granulomas (27,48). TNF induces chemokines including MIP-1α, MIP-1β, MIP-2, MCP-1, and "regulated on activation, normal T-cell expressed and secreted" (RANTES), and subsequent leukocyte recruitment to infected organs (27). In addition to its role in initial cellular recruitment, TNF is also required for establishing and maintaining granuloma architecture: it regulates the tight association between macrophages and lymphocytes within granulomas (27).

In the case of *M. tuberculosis*, bactericidal mechanisms, such as those involving production of nitric oxide, appear to be poorly expressed by human cells (49). Instead, human mycobacterial immunity reflects growth inhibitory mechanisms requiring direct cell contact and activation (50-52). As a result, the continuous, lifelong recruitment of antigen-specific T-cells is required to maintain granulomas and prevent progression of latent infection to active disease.

TNF Antagonists and Granulomatous Infections

A number of granulomatous infections, including those due to *M. tuberculosis* (4,13,17,21,22,24,25), *Histoplasma capsulatum* (16,18), *Cryptococcus neoformans* (19,24,25), *Coccidioides immitis* (24,25,53), *Aspergillus* (20,24,25), and *Listeria monocytogenes* (14,15,24,25, 54,55), have been reported in association with the use of TNF antagonists. Early studies done with adalimumab suggested a dose–response relationship with the occurrence of TB (8). Patients who developed active TB were receiving higher doses than the licensed dose of 40 mg every other week. Reducing the treatment dose and screening for the presence of latent TB reduced the frequency of active TB to 1 to 2 cases in the next approximately 2500 patients, although it did not eliminate the occurrence of TB completely (23).

While it is difficult to make firm conclusions due to potential biases inherent in the databases used, passive surveillance studies have indicated the possibility of a higher incidence of TB associated with the use of infliximab than with etanercept (4,13,22-25,56,57). One study analyzed all postmarketing infliximab-related TB reports received through the Food and Drug Administration's (FDA) Adverse Event Reporting System (AERS) from the licensure of infliximab in 1998 to May 29, 2001; the authors concluded that the rate of reported TB cases among infliximab-treated patients was numerically, but not statistically, higher than the available background rates (13). More recently, results from an analysis of reports from a European surveillance database supported these findings (57). A review of the AERS from November 1998 to March 2002 revealed 25 cases of TB in etanercept-treated RA patients in the US for an estimated rate of 10/100,000 patient-years of exposure (24,25). Based on patients identified from the National Data Bank for Rheumatic Diseases, Wolfe and coworkers calculated the rate for TB in patients with RA: (1) on infliximab therapy to be 53 (95% CI 14 to 130)/100,000 and (2) not on TNF antagonists to be 6.4 (95% CI 1.6 to 34)/100,000 (58). A consistent observation in all these studies was a high proportion of extrapulmonary and disseminated disease.

Other granulomatous infections have also been reported more frequently with infliximab than with etanercept (16,23-25,54). One study analyzed all reports of histoplasmosis following infliximab or etanercept therapy that were received through the AERS from licensure of the 2 drugs in 1998 to July 2001 (16). Histoplasmosis was reported in approximately 6/100,000 infliximab-treated patients and 1/100,000 etanercept-treated patients, suggesting that patients treated with infliximab may have a higher risk for developing histoplasmosis compared with patients treated with etanercept. Listeriosis was identified in 15 patients from the AERS (through December 2001) (54). Fourteen had received infliximab and 1 had received etanercept. From May 1998 through February 2003, Bergstrom and colleagues identified 13 cases of coccidioidomycosis (12 on infliximab and 1 on etanercept) from 5 clinical practices in an endemic area (53).

A comprehensive analysis of the AERS database identified 15 types of granulomatous infections associated with the use of infliximab and etanercept specifically in US patients from 1998 through the third quarter of 2002 (Table 1) (24,25). More cases of granulomatous infections were reported in infliximab-treated patients (130/100,000) than in etanercept-treated patients (60/100,000). In both groups, TB was the most frequently reported disease, occurring in 54/100,000 infliximab-treated patients and 28/100,000 etanercept-treated patients. The reported rates of TB during the first 90 days of anti-TNF treatment were 95/100,000 person-years for infliximab versus 11/100,000 for etanercept. The onset of TB in relation to duration of etanercept treatment was relatively constant, suggesting acquisition of new disease. The increased rate of infection in patients shortly after starting infliximab therapy is consistent with reactivation of disease; the decrease in the rate after 90 days indicates a shift to acquisition of new disease (24,25). Adalimumab was not yet approved during the time period of this study; however, clinical trial data also demonstrate an association with granulomatous infections. Thirteen cases of TB and 6 cases of invasive infections caused by *Histoplasma*, *Aspergillus*, and *Nocardia* species were reported in adalimumab clinical trials.

In addition, Keane and coworkers noted a higher frequency of extrapulmonary TB associated with infliximab-treated patients than with nontreated patients (13); such forms of TB are commonly observed in cases of immunosuppression (59). Disseminated forms of TB are associ-

Table 1 Incidence of Granulomatous Infections in US Patients Treated with Infliximab and Etanercept*[24,25]

Infection	Infliximab†	Etanercept‡
	N (*N*/100,000 treated patients)	
Tuberculosis§	106 (54)	32 (28)
Histoplasmosis§	37 (19)	3 (2.7)
Atypical mycobacteriosis	22 (11)	7 (6.2)
Candidiasis	20 (10)	6 (5.3)
Aspergillosis	17 (8.6)	7 (6.2)
Listeriosis§	17 (8.6)	1 (0.88)
Cryptococcosis	10 (5.1)	8 (7.1)
Coccidioidomycosis§	11 (5.6)	1 (0.88)
Nocardiosis	7 (3.6)	1 (0.88)
Toxoplasmosis	4 (2.0)	0 (0)
Total§	255 (130)	68 (60)

*As of September 2002. Rates were calculated based on 197,000 and 113,000 treated patients for infliximab and etanercept, respectively.
†In addition, 1 case each of bartonellosis, legionellosis, leprosy, and pneumocystosis were identified and these are included in the column total.
‡In addition, 2 cases of salmonellosis were identified and are included in the column total.
§$P \leq 0.01$.

ated with higher mortality and morbidity; examples of disseminated TB cases observed include lymph-node disease, peritoneal disease, bone disease, brain abscess, and central nervous TB such as tuberculous meningitis (13,59). The atypical presentation of these TB cases result in their underdiagnosis or late diagnosis, further increasing morbidity and mortality rates (59,60).

Mechanisms of Action of TNF Antagonists and Their Potential Role in Granulomatous Infections

Although treatment with any of the 3 TNF antagonists results in modulation or neutralization of TNF-induced and regulated biological responses, differences among the TNF blockers with respect to pharmacokinetics and mechanism of action may explain the potential risk differences for developing granulomatous infections.

Binding Avidity

Etanercept is a dimeric fusion protein that consists of 2 molecules of the extracellular ligand-binding portion of human TNF receptor 2 attached to the Fc domain of human IgG1 (61). Infliximab is a chimeric IgG1 monoclonal antibody composed of human constant and murine variable regions (62), whereas adalimumab is a recombinant human IgG1 monoclonal antibody (63). Each infliximab or adalimumab molecule can bind up to 2 TNF molecules; a single TNF homotrimer can bind up to 3 molecules of infliximab or adalimumab (64,65). In contrast, etanercept binds to the interface of 2 TNF subunits in a 1:1 ratio (64). One possible ramification of these differences in avidity is that it may be easier to titrate TNF suppression to inhibit inflammation without reducing resistance to infusion by drugs that bind less avidly or less efficiently (in this case, etanercept). Another ramification is that the monoclonal antibodies may be more potent, and effective inhibitors of TNF.

Pharmacokinetics

It is possible that some of the apparent differences in the incidence of activation of latent TB are due to pharmacokinetic differences among the 3 TNF blocking agents. Infliximab and adalimumab have similar half-lives, and longer serum half-lives than etanercept (Table 2) (61). For example, infliximab can still be detected in the serum after 8 weeks following a single 3 mg/kg dose (62), and detectable concentrations of free infliximab have been observed for up to 28 weeks (66). Based on the pharmacokinetic profile of infliximab compared with etanercept, TNF suppression may be greater and more prolonged during infliximab treatment than during etanercept treatment. Prolonged inhibition of TNF may produce a "functional knockout" of macrophage function, with increased susceptibility to infection (conversely, it may also improve efficacy).

While data are not yet definitive, it may be that there is a lower incidence of latent TB activation after adalimumab (at doses used in the clinic) than infliximab. Because the terminal half-lives, volumes of distribution, and clearances of these 2 compounds are approximately equivalent, simple steady-state concentrations would not account for the difference in latent TB activation as steady-state concentrations are probably quite similar for these 2 compounds. Beyond obvious differences in bioavailability, what pharmacokinetic factor, then, might account for the difference? Since infliximab is given intravenously and adalimumab is given subcutaneously, an important difference might be the difference in peak concentrations. For example, a higher peak concentration may temporarily saturate protein binding or, by simple mass action, result

Table 2 Pharmacokinetics of the TNF Antagonists[61–63,66,83,84]

	Etanercept*	Infliximab	Adalimumab
C_{max} (μg/mL)	1.1 ± 0.6	118†	4.7 ± 1.6§
Time to C_{max} (h)	69 ± 34	NA	131 ± 56§
Bioavailability	76%	100%	64%§
Clearance (mL/h)	160 ± 80	11†	12¶
Volume of distribution (L)	10.4	3.0	4.7–6.0¶
$t_{1/2}$ (days)	4.25 ± 1.25	8–10‡	10–20¶

NA: information not available.
*25 mg s.c.
†5 mg/kg i.v.
‡3 to 20 mg/kg i.v.
§40 mg s.c.
¶0.25 to 10 mg/kg i.v.

in higher concentrations of infliximab than adalimumab in deeper tissues (eg, within granulomata). If on the other hand there is also a difference between adalimumab and etanercept with respect to the incidence of latent TB activation (ie, lower incidence with etanercept), then factors such as clearance, serum half-life, and/or dosing regimens may play a role rather than peak concentration.

At present the specific role of pharmacokinetic differences in the activation of latent TB remains speculative. Ongoing studies may elucidate the role of kinetics and intermittent TNF suppression on monocytes/macrophage activity.

Antibody-Mediated Cell Lysis

Cells coated with antibody isotypes that fix complement and bind Fc receptors (such as human IgG1) can activate complement-dependent cytolysis and antibody-dependent cell-mediated cytotoxicity. Infliximab induces complement-dependent cytolysis and antibody-dependent cell-mediated cytotoxicity in a murine myeloma cell line expressing membrane-associated TNF (67). Macrophages and monocytes are among the cells that express membrane-associated TNF. The monocytopenia observed in patients following treatment with infliximab that can persist for weeks following infusion (68) may reflect direct killing of cells expressing membrane-associated TNF by infliximab. This has clinical implications because monocytes are an essential component of granulomas; monocyte elimination might lead to susceptibility to granulomatous diseases. Adalimumab may have similar activity because its effector portion is identical to that of infliximab (IgG1).

Etanercept contains the Fc portion of IgG1, but reportedly does not fix complement (69), perhaps because steric hindrance prevents C1q binding, which initiates the classical complement cascade. Furthermore, because etanercept binds only single molecules of TNF, it is unlikely to form aggregates that can activate complement-dependent cytolysis and antibody-dependent cell-mediated cytotoxicity.

Induced Apoptosis

Some data suggest a role for infliximab in inducing apoptosis in activated monocytes and T-cells (70). In IBD patients, infliximab is associated with an unexpectedly sustained clinical response (11). Assays of intestinal biopsies from Crohn's disease patients taken 24 hours after infliximab infusion found an increase in apoptotic CD3+ cells (71). The same authors showed that infliximab induced apoptosis in activated but not resting T-cells in vitro. These results suggest that infliximab may exert its sustained therapeutic effects in IBD by causing apoptosis of T-lymphocytes since uncontrolled T-cell activation plays a central role in IBD pathogenesis. Apoptosis also has been observed in circulating monocytes from Crohn's disease patients following infliximab infusion (72). Recently, Catrina and coworkers demonstrated that both etanercept and infliximab can induce apoptosis in monocytes and macrophages from patients with RA (73). This effect was more prominent in cells from the synovial fluid than from peripheral blood; no such effect was observed in lymphocytes.

Removal of activated monocytes, macrophages, and CD4+ T-cells might be desirable in chronic inflammation. However, because these cell types play an essential role in granuloma formation and maintenance, infliximab-induced apoptosis may lead to more potent and long-lasting inhibition of the granulomatous response than expected from TNF neutralization alone.

Lymphotoxin-α Binding

Etanercept differs from the monoclonal antibodies in that it binds lymphotoxin-α. Lymphotoxin-α binds to TNF receptors but in contrast to TNF its expression is primarily limited to lymphocytes. Little is known about the biological role of lymphotoxin-α. Roach and colleagues have shown that lymphotoxin-α, acting independently of TNF, plays an essential role in cellular recruitment and organization of granulomas in *M. tuberculosis* infection (74). This evidence suggests that etanercept could exacerbate granulomatous diseases more potently than agents that merely neutralize TNF.

Inhibition of IFNγ Production

IFNγ is produced by activated T-cells. Like TNF, IFNγ is required for host defenses against mycobacterial infection (75). Zou and coworkers have examined the effects of TNF blockers on the frequency of IFNγ-producing T-cells. IFNγ expression was decreased by infliximab, but not by etanercept (76,77). A recent report by Wallis and coworkers examined the effects of therapeutic concentrations of TNF blockers on antigen-induced IFNγ production using whole blood culture. Peak and trough concentrations of infliximab and adalimumab caused significant inhibition of IFNγ, whereas no significant effect was observed with etanercept even at supratherapeutic concentrations (78). The mechanism of this effect is apparently indirect, as neither monoclonal is capable of directly binding IFNγ. The possible excess TB risk posed by infliximab may therefore reflect its ability to inhibit both TNF and IFNγ.

DISCUSSION

Monoclonal antibodies and soluble receptors that target TNF are effective in treating RA and have dramatically improved the lives of people suffering from this disease. However, there are significant differences between these 2 classes of TNF inhibitors. Pharmacokinetic differences are profound, with the monoclonal antibodies (infliximab and adalimumab) demonstrating lower clearances, greater volumes of distribution, and longer half-lives than the soluble

receptor (etanercept). Furthermore, the intravenous form of monoclonal antibody (infliximab) has greater bioavailability and shows much higher peak concentrations. This probably results in more constant and also higher peak tissue concentrations for the monoclonal antibodies, despite the differing doses and dosing regimens among the 3 drugs. These differences, in turn, may result in different effects on TNF concentrations and different effects on effector cells. Monoclonal antibodies may also eliminate activated T-cells and monocyte/macrophages directly either by cell lysis or by inducing apoptosis. These differences may explain the greater efficacy of infliximab in Crohn's disease on the one hand, and the seeming trend toward greater susceptibility to granulomatous infections such as TB with infliximab on the other. Improved data about tissue kinetics and about the relative risks of different TNF agents would be valuable, as would information on relative risks in patients with different disease states.

To address concerns about increased susceptibility to TB reactivation or infection, screening for TB exposure with tuberculin skin testing or newer interferon-based serum tests (which are available, although not fully validated) (79) should be performed before beginning therapy with anti-TNF agents. Anergy is known to occur in patients with RA or Crohn's disease, and the possibility of false-negative skin tests must be taken into consideration. More sensitive and specific tests for TB are being developed. Activation of latent TB may be especially difficult to identify in patients treated with systemic steroids (80-82). Furthermore, screening tests are not routinely available for latent infection with the other agents listed in Table 1. Therefore, a high index of suspicion for granulomatous infections should be maintained in patients treated with TNF antagonists. For example, prolonged unexplained malaise, fevers, and weight loss need to be followed up, despite negative skin or serum tests or chest radiographs. Development of active granulomatous infection should prompt immediate discontinuation of anti-TNF antagonists and aggressive diagnostic and therapeutic maneuvers to ascertain the extent and virulence of an infection. While this article seeks to clarify and explain differences among TNF blocking agents with respect to the most common granulomatous infection, TB, many questions clearly remain. We have already mentioned the continuing need to understand the pharmacokinetic effects of these drugs and the many pharmacodynamic effects of TNF blocking agents. Additionally, other host factors (eg, ethnicity, concomitant diseases, cytokine modulation of responses, other drug interactions) and environmental factors (eg, dose of and virulence of infecting organisms, host nutrition, socioeconomic factors) will also need to be elucidated.

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