

ARTICLE

A Pilot Dose-Finding Study of Etanercept in Rheumatoid Arthritis

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A randomized, parallel-dose study assessed the pharmacokinetics (PK) and pharmacodynamics (PD) of etanercept in 61 patients with rheumatoid arthritis (RA) who received doses from 10 mg once-weekly to 50 mg twice-weekly for 4 weeks. Empiric application of a maximal-effect (E_{max}) model to pooled steady-state concentrations (C_{ss}) and PD markers provided half-maximal-effect concentration estimates of 567, 573, 465, 87, and 159 ng/mL for change from baseline in number of swollen joints, number of painful joints, erythrocyte sedimentation rate, interleukin-6, and matrix metalloproteinase-3, respectively. $C_{ss} > \sim 2,000$ ng/mL did not appear to offer additional benefit. It was concluded that the middle doses, 10 mg twice-weekly, 50 mg every 2 weeks, and 50 mg once-weekly, would provide C_{ss} in the target range of 500–2,000 ng/mL. The revised US Food and Drug Administration guideline for development of medicines for treatment of RA encourages a study design incorporating PK/PD assessment to inform later studies.

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Study Highlights

WHAT IS THE CURRENT KNOWLEDGE ON THE TOPIC?

✓ Determining the optimal dose for the treatment of rheumatoid arthritis (RA) demands several large studies conducted over at least 12 weeks of treatment or placebo control. More recently, the US guidance for drug development for RA treatments includes suggestions that pharmacokinetics/pharmacodynamics (PK/PD) studies assessing response between 2 and 8 weeks may be more informative in distinguishing doses.

WHAT QUESTION DID THIS STUDY ADDRESS?

✓ Can a small study including assessment of PK/PD over a wide range of doses over a 4-week period provide sufficient information to inform the design of subsequent studies, minimizing the number of patients and length of time

required to determine the optimal dose or a narrow range of doses suitable for further study?

WHAT THIS STUDY ADDS TO OUR KNOWLEDGE

✓ Empiric application of an E_{max} model to observations collected over a wide range of concentrations can provide insight about potentially effective dosing/concentration ranges to explore in larger phase IIb and III studies.

HOW THIS MIGHT CHANGE CLINICAL PHARMACOLOGY OR TRANSLATIONAL SCIENCE

✓ It is important to extend the dosing range as wide as possible in order to gain the most insight into a possible concentration–response range. Limiting the length of the study to a very short interval may allow doses that would not be reasonable for a longer study, due to patient safety or lack of effective treatment.

The successful registration of a new drug requires completion of at least two randomized, controlled clinical trials demonstrating safety and efficacy of the drug at a defined dose in the intended patient population. Translation of the preliminary assessment of safety and pharmacokinetics (PK) observed in first-in-human studies to patients, and of evidence of efficacy from nonclinical models to patients, are two of the most important tasks in drug development.

Rheumatoid arthritis (RA) is the third-most common type of arthritis, after osteoarthritis and gout.¹ The prevalence of RA is believed to range from 0.4–1.3% internationally, and an estimated 1.5 million (0.6%) adults aged 18 years and older in the United States were reported to have had RA in 2005.² In addition to morbidity and decreased quality of life due to pain

and joint damage, mortality is increased among those who have RA compared with those who do not.² The development of effective treatments is clearly a significant public health concern.

The first US regulatory guidance for the development of drug products for the treatment of RA was finalized in 1999,³ and a revised draft guidance was published in 2013.⁴ Key differences between the two guidance documents, which reflect the advances in clinical research and drug development of treatments of RA, are emphasis on the length of clinical trials and the role of pharmacokinetic (PK) and pharmacodynamic (PD) assessments. In the first guidance document, it is recommended that clinical trials continue for at least 6 months, due to the long duration of RA and the importance

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of demonstrating the durability of the treatment, particularly for biological agents where the development of antidrug antibodies could potentially interfere with clinical efficacy.³ In the second guidance, earlier evaluation of clinical response, i.e., between 2 and 8 weeks, prior to the likely plateau of clinical response, was suggested as likely to be more informative and more likely to distinguish between doses.⁴

A second difference between the two guidance documents is a reflection of the usefulness of PK and PD assessments of different dosage regimens in optimizing treatment. The first guidance document recommended assessment of PK primarily to understand the absorption, distribution, metabolism, and elimination (ADME) characteristics of the drug and the potential impact of immunologic response on ADME of biological therapies,³ whereas the second guidance advocates linking drug-concentration results to measures of efficacy and safety, thus providing support for the recommended dosage.⁴

Etanercept (Enbrel, Pfizer, New York, NY) is a biological disease-modifying antirheumatic drug (DMARD) that inhibits tumor necrosis factor (TNF), and thus decreases inflammation. The first clinical trials of etanercept in patients with RA began in 1993⁵ and it was approved for reducing the signs and symptoms of RA in 1998 in the United States.⁶ Subsequent studies were performed to expand the approved RA indications to include inducing major clinical response, inhibiting the progression of structural damage, and improving physical function. Development of etanercept occurred concurrently with the advancement of science, which resulted in the differences between the two guidance reports.

The goal herein is to report the results of a small, previously unpublished, dose-finding study that incorporated PK/PD analyses performed during the development of etanercept, thus providing a case study of the advances in clinical trial design for RA that encompasses the differences between the two guidance documents. This may offer a helpful example of study design for dose-finding studies in other diseases, especially those with small patient populations.

METHODS

All available dose-finding studies conducted during the development of etanercept for the treatment of RA were identified. Information on the doses administered, number of patients enrolled, length of study, outcome measures (including PK assessment), and study conclusions was collated and reviewed. Five of the six studies have been published previously (Table 1).^{5,7-10} The remaining study is described below.

Patients

Eligible patients aged 18 to 75 years were enrolled from September 1997 to August 1998. All patients met the 1987 American Rheumatism Association (ARA) criteria for RA and belonged to ARA functional class I to III. Their onset of disease had to have been after 16 years of age and disease duration had to be <10 years. They had to have failed to respond to at least one previous DMARD and have had active RA, defined by the presence of \geq six swollen and \geq 12 tender joints and at least two of the following three criteria: erythro-

cyte sedimentation rate (ESR) \geq 28 mm/h, C-reactive protein (CRP) \geq 20 mg/L, and morning stiffness \geq 45 minutes. Prior treatment with an anti-TNF monoclonal antibody or with a soluble TNF receptor were exclusion criteria. The study was conducted in accordance with the Declaration of Helsinki and independent Ethics Committee approval was obtained for the protocol and all amendments. Written informed consent was obtained from all patients at the time of enrollment.

Study design

This randomized, double-blind, placebo-controlled, prospective study included a DMARD washout period, a treatment period, and a follow-up period. Etanercept was supplied in vials of sterile lyophilized powder containing 10- or 25-mg doses that were reconstituted with 1 mL of water for injection. Patients were randomly assigned in block sizes of 5 in a 2:2:1 ratio (two for each active group, one for placebo) to receive subcutaneous injections of etanercept (10 mg once weekly, 10 mg twice weekly, 50 mg once every 2 weeks, 50 mg once weekly, or 50 mg twice weekly) or matching placebo for 4 weeks. The doses and regimens were chosen to provide a large range of etanercept serum concentrations to examine potential concentration-response relationships for various markers of biologic activity. Prior to this study, doses up to 25 mg twice weekly had been evaluated. The dose of 50 mg twice weekly was added to this study as a preliminary assessment of further benefit and safety. Although the short duration of treatment would not result in optimal response, it was thought to be long enough to elicit some change in disease activity evaluations. All DMARDs had to be discontinued at least 4 weeks prior to dosing of study medication and the follow-up period after treatment was 4 weeks.

Assessments

Prestudy screening assessments included medical history, physical examination, vital signs, electrocardiogram, joint assessment, pain (visual analog scale (VAS)), duration of morning stiffness, CRP, ESR, hematology, blood chemistry, serum human chorionic gonadotropin test for women who entered menopause less than 2 years before screening visit, and urinalysis. Additional assessments taken at baseline included rheumatoid factor, etanercept concentration, interleukin (IL)-6, IL-1 receptor antagonist (IL-1Ra), matrix metalloproteinase (MMP)-3, and testing for antibodies (antinuclear, antidouble-strand DNA, anticardiolipin, anti-tanercept).

Etanercept serum concentration, number of swollen/painful joints, ESR, IL-6, and MMP-3 were assessed at baseline, on days 8, 15, 22, and 29 of treatment, and during weeks 1, 2, and 4 of follow-up. Blood samples for etanercept concentration measurements were collected at the same time as efficacy assessments. Etanercept concentrations were determined by a validated enzyme-linked immunosorbent assay with a limit of quantitation of 0.781 ng/mL based on a 1:5 minimum sample dilution. All reported adverse events were recorded.

Data analysis

Sample size and power were not applicable and no formal hypothesis testing was done. There was no formal between-group statistical evaluation of efficacy. The last observation

Table 1 Dose-ranging studies of etanercept in patients with rheumatoid arthritis

Study	N	Dose	Duration (months)	Outcomes assessed	Conclusions
Moreland <i>et al.</i> , 1996 ⁵	22	4, 8, 16, 32 mg/m ² i.v. then 2, 4, 8, 16 mg/m ² SQ twice-weekly	1	Various timings; efficacy, safety, PK	No clear dose response; serum concentrations proportional to dose
Moreland <i>et al.</i> , 1997 ⁷	180	0.25, 2, 16 mg/m ² SQ twice-weekly vs. placebo	3	Every 2 wk for 3 mo; efficacy, safety	16 mg/m ² most efficacious, no dose-limiting toxicity
Moreland <i>et al.</i> , 1999 ⁸	234	10, 25 mg SQ twice-weekly vs. placebo	6	2 wk, 3 mo; efficacy, safety	25 mg more rapid and better response, no dose-limiting toxicity
Batho <i>et al.</i> , 2000 ⁹	632	10, 25 mg SQ twice-weekly vs. MTX	12	3, 6, 9, 12 mo; efficacy and safety	25 mg improved efficacy compared with MTX
Johnsen <i>et al.</i> , 2006 ¹⁰	77	25, 50 mg SQ twice-weekly	6	4, 8, 12, 24 wk; efficacy, safety	No difference in efficacy or safety

i.v., intravenous; MTX, methotrexate, PK, pharmacokinetics; SQ, subcutaneous.

carried forward imputation method was used for missing data.

For the PK analysis, serum concentration measurements were combined with dose administration information and fit with a one-compartment PK model with first-order absorption using WinNonlin (v. 1.5, Pharsight, Mountain View, CA) for each individual patient. Population PK methods were not utilized because of the small number of observations. Predose etanercept concentrations above the lower limit of quantification, caused by crossreactive analytes in the serum, were not subtracted from the postdose concentrations because they were very low. If a one-compartment model did not fit well for an individual, noncompartmental methods were used. The first-order elimination rate constant (λ_z) was determined using log linear regression of etanercept concentrations in the terminal portion of the elimination phase. Apparent clearance (Cl/F) was estimated by computing a ratio of dosing rate (mg/h) to concentration at steady-state (C_{ss}). Apparent volume of distribution (V/F) was calculated as a ratio of Cl/F to λ_z . Half-life was calculated as $0.693/\lambda_z$. An analysis of variance (ANOVA) was performed to test differences in PK parameters (absorption rate constant, λ_z , area under the serum concentration–time curve (AUC), Cl/F, and V/F) between dose groups. AUC values were normalized for dose prior to performance of ANOVA.

There was considerable noise in the PD observations because of a small number of sparsely timed PD observations, so a naïve pooled-estimate process was used to fit PD observations to pooled C_{ss} data as follows. Etanercept C_{ss} for each patient as measured on day 29 was used, with the exception of patients who received 50 mg once every 2 weeks, for whom the day 22 C_{ss} was used. The low concentrations of etanercept reported in patients treated with placebo were used for their day 29 C_{ss} . Next, to provide even weighting across the concentration spectrum, the range of observed C_{ss} values was broken into six intervals so that each interval of C_{ss} values contained at least five observations. The intervals were uneven in length, being shorter where there were more observations. A mean C_{ss} was calculated for each interval and the mean of the PD observations was calculated using each patient's corresponding observation. PD analysis was performed by fitting a sigmoidal maximal effect (E_{max}) model to mean C_{ss} for each interval and

mean change for each interval from baseline to 29 days in the following: number of painful joints, number of swollen joints, pain (VAS), morning stiffness, ESR, IL-6, IL-1Ra, and MMP-3. The E_{max} , half-maximal effect (EC_{50}), and sigmoidicity factors (ζ) were estimated using WinNonlin. PD assessment of adverse events was not attempted.

RESULTS

Patient disposition

Of the 61 patients enrolled, 49 received etanercept and 12 received placebo. PK parameters were determined in 43 patients. Demographic and baseline characteristics are shown in **Table 2**.

Safety

Etanercept appeared to be generally well tolerated by patients. Injection site reactions, rhinitis, and upper respiratory infections were the most common treatment-emergent adverse events for most etanercept-treated patients. Two patients in the etanercept 10 mg twice-weekly group and one patient in the placebo group withdrew from the study due to adverse events: one patient in the 10 mg twice-weekly group experienced lower extremity vascular disorder (not related to etanercept treatment); a second patient receiving 10 mg twice-weekly had facial edema and pruritus after the first injection and mild dyspnea occurred after the second injection; a patient in the placebo group had a broken tibia. These three patients were excluded from the PK analysis.

PK analysis

The mean etanercept concentrations by dose are shown in **Figure 1**. Exposure with the 10 mg once-weekly dose was clearly lower than the other etanercept doses and the 50 mg twice-weekly dose was clearly higher, whereas the 10 mg twice-weekly, 50 mg every 2 weeks, and 50 mg once-weekly doses provided similar exposure to each other, consistent with the respective C_{ss} values. PK parameters are shown in **Table 3**. The PK parameters for seven patients ($n = 1$ each for all dosing groups with the exception of the 50 mg once-weekly group, $n = 2$ in the 50 mg once-weekly group) were assessed using noncompartmental analysis methods, because of inadequate characterization of the first-order absorption rate constant.

Table 2 Demographic and baseline characteristics

Characteristic ^a	Etanercept					Placebo (n = 12)
	10 mg once-weekly (n = 11)	10 mg twice-weekly (n = 11)	50 mg once every 2 weeks (n = 9)	50 mg once-weekly (n = 9)	50 mg twice-weekly (n = 9)	
Age, y	51.5 ± 12.4	52.8 ± 14.7	51.6 ± 13.9	49.3 ± 12.5	55.6 ± 12.8	53.0 ± 15.3
Female, n (%)	11 (100)	10 (91)	6 (67)	7 (78)	7 (78)	11 (92)
Weight, kg	78.7 ± 11.6	64.3 ± 8.9	72.9 ± 11.4	74.9 ± 9.8	68.1 ± 12.2	68.5 ± 9.7
Disease duration, y	4.2 ± 3.1	6.5 ± 3.3	3.5 ± 2.9	4.2 ± 3.4	5.0 ± 2.6	5.0 ± 3.6
RF positive, U/L	83.3 ± 103.9 (n = 10)	48.1 ± 65.6 (n = 10)	34.7 ± 43.1	37.6 ± 22.9	79.9 ± 77.0	97.2 ± 93.2
NSAIDs, n (%)	8 (73)	7 (64)	8 (89)	7 (78)	7 (78)	8 (67)
Corticosteroids, n (%)	5 (45)	8 (73)	1 (11)	3 (33)	7 (78)	7 (58)
≥1 prior DMARD, n (%)	2 (18)	1 (9)	5 (56)	6 (67)	3 (33)	4 (33)
Pain (VAS)	67.9 ± 16.5	67.0 ± 22.6	61.6 ± 17.0	62.7 ± 34.6	58.8 ± 15.6	60.8 ± 19.6
Painful joints	29.8 ± 15.0	35.4 ± 15.9	28.6 ± 12.2	27.9 ± 13.1	26.2 ± 10.7	32.5 ± 11.8
Swollen joints	20.3 ± 12.7	28.7 ± 12.9	20.8 ± 6.6	21.7 ± 8.4	24.6 ± 9.6	23.5 ± 7.2
Morning stiffness, min	106.8 ± 75.7	177.3 ± 193.6	80.6 ± 47.2	142.5 ± 125.0 (n = 8)	142.2 ± 81.5	172.5 ± 126.7
CRP, mg/L	52.8 ± 30.1 (n = 9)	61.8 ± 32.6 (n = 10)	58.8 ± 28.8	72.2 ± 27.3	37.2 ± 27.7	47.8 ± 18.4
ESR, mm/h	31.7 ± 22.0 (n = 10)	64.7 ± 46.3 (n = 10)	42.4 ± 26.5	50.4 ± 21.6	35.9 ± 35.5	45.0 ± 34.7
IL-1Ra, pg/mL	1433 ± 690.1 (n = 10)	1165 ± 763.7 (n = 10)	1337 ± 465.3	1216 ± 832.9	1367 ± 1313	1090 ± 671.7
IL-6, pg/mL	30.6 ± 26.8 (n = 10)	46.3 ± 28.8 (n = 10)	86.9 ± 125.1	55.8 ± 35.0	30.9 ± 30.9	43.3 ± 45.7
MMP-3, ng/mL	109.3 ± 102.0 (n = 10)	192.3 ± 115.7 (n = 10)	236.3 ± 136.6	296.7 ± 154.7	128.8 ± 114.8	154.9 ± 127.0

^aValues are mean ± standard deviation unless otherwise stated.

CRP, C-reactive protein; DMARD, disease-modifying antirheumatic drug; ESR, erythrocyte sedimentation rate; IL-1Ra, interleukin-1 receptor antagonist; IL-6, interleukin-6; MMP-3, matrix metalloprotease-3; NSAID, nonsteroidal antiinflammatory drug; RF, rheumatoid factor; VAS, visual analog scale.

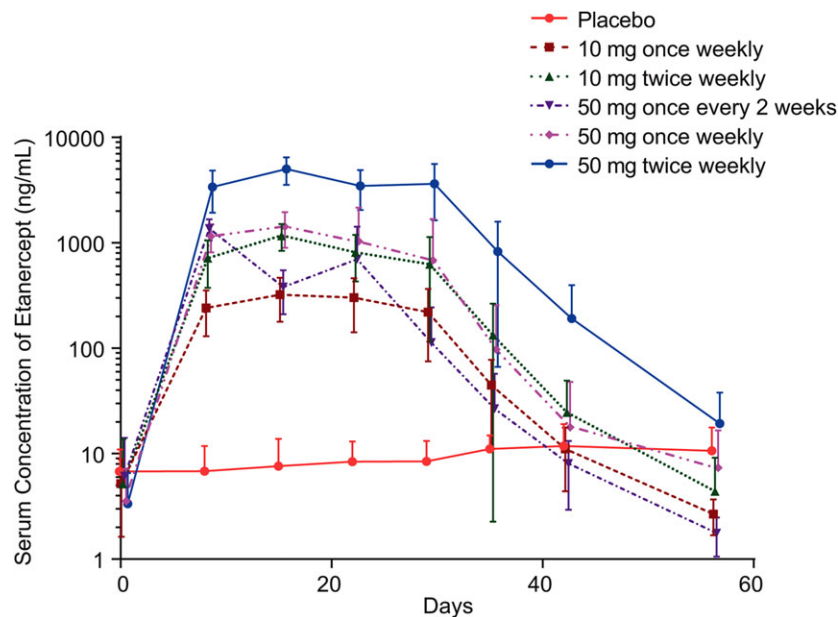


Figure 1 Mean (± SD) etanercept serum concentrations in patients with RA receiving 4 weeks of treatment.

Table 3 Steady-state etanercept PK parameters in patients with RA

Dose (n)	C _{max} (mg/L)	T _{max} (h)	T _{1/2} (h)	Cl/F (mL/h)	AUC (mg·h/L)	C _{ss} (ng/mL)	V/F (L)
10 mg once-weekly (11)	0.40 ± 0.17	65 ± 39	68.2 ± 27.4	168 ± 82	68 ± 26	300 ± 135	16 ± 11
10 mg twice-weekly (9)	0.57 ± 0.42	58 ± 27	61.0 ± 12.8	203 ± 181	74 ± 39	806 ± 424	18 ± 16
50 mg once every 2 weeks (9)	2.21 ± 1.58	43 ± 50	62.9 ± 23.1	158 ± 108	294 ± 202	915 ± 621	33 ± 55
50 mg once-weekly (5)	1.56 ± 0.73	54 ± 64	58.1 ± 19.0	234 ± 108	181 ± 132	1170 ± 660	32 ± 32
50 mg twice-weekly (9)	3.26 ± 1.66	45 ± 30	57.8 ± 26.1	143 ± 85	412 ± 162	3888 ± 1247	12 ± 7.6
Overall mean	ND	53 ± 40	62 ± 22	175 ± 116	ND	ND	21 ± 29

Values are mean ± standard deviation.

AUC, area under the concentration–time curve; C_{max}, maximum serum concentration; Cl/F, apparent total clearance; C_{ss}, concentration at steady-state; ND, not determined; T_{1/2}, elimination half-life; T_{max}, time to reach maximum concentration; V/F, volume of distribution.

Table 4 Mean concentrations and pharmacodynamic measurements (% change from baseline)

	Concentration range (ng/mL)					
	0–99 (Group 1)	100–499 (Group 2)	500–999 (Group 3)	1000–1999 (Group 4)	2000–2999 (Group 5)	3000–6080 (Group 6)
n	7	14	7	9	5	6
Concentration range, ng/mL	0.96–90.2	124–422	514–881	1,012–1,985	2,100–2,985	3,185–6,080
Mean concentration, ng/mL	20.0	296	710	1,214	2,429	4,510
Swollen joints	1.00	13.83	40.99	33.37	51.83	54.16
Painful joints	15.62	14.92	31.35	52.26	62.21	69.00
Pain (VAS)	9.38	28.73	30.95 ^a	42.27	58.78	80.04
Morning stiffness	20.24	22.44	39.88	45.31	72.92	86.11
ESR	9.38	16.67	25.82	37.25	22.35	37.24
IL-6 ^b	–124.3	25.15	22.91	32.27	–0.49	57.41
IL-1Ra ^c	5.16	–0.07	12.53	11.09	–0.24	13.98
MMP-3 ^d	–28.61	12.86	15.98	28.22	25.83	28.23 ^e

Patients receiving placebo were included in the pharmacodynamic analysis.

^aOne patient was removed from the analysis due to an outlier reading of percentage change from baseline of –933%.

^bValues were scaled prior to analysis by adding 125 to score to avoid negative numbers.

^cValues were scaled prior to analysis by adding 1 to score to avoid negative numbers.

^dValues were scaled prior to analysis by adding 30 to score to avoid negative numbers.

^eOne patient was removed from the analysis due to an outlier reading of percentage change from baseline of –248%.

ESR, erythrocyte sedimentation rate; IL-6, interleukin-6; IL-1Ra, interleukin-1 receptor antagonist; MMP-3, matrix metalloproteinase-3; VAS, visual analog scale.

PD analysis

There were between five and 14 concentration–effect pairs in each of the six C_{ss} ranges. The means of the pooled etanercept concentrations and PD observations used for the modeling procedure are shown in **Table 4**. Attempts to fit a model to pain (VAS), duration of morning stiffness, and IL1-Ra were not successful. The model did not fit or was inappropriate for various reasons, including failure to demonstrate a relationship and/or lack of a clear maximum effect. However, PD measures for swollen joints, painful joints, ESR, IL-6, and MMP-3 and corresponding mean estimates of C_{ss} were successfully fit with a sigmoidal E_{max} model. The estimates for PD model parameters are shown in **Table 5**. The plots

Table 5 PD parameters from E_{max} models for clinical and biological efficacy variables after 1 month of etanercept treatment in patients with RA

	E _{max} (%)	EC ₅₀ (ng/mL)	Sigmoidicity factor (ϕ)
Swollen joints	56.97	567	1.33
Painful joints	66.82	573	1.67
ESR	46.00	465	0.48
IL-6	28.11	87	3.13
MMP-3	27.88	159	1.39

ESR, erythrocyte sedimentation rate; IL-6, interleukin-6; MMP-3, matrix metalloproteinase-3.

of the predicted vs. observed effect for swollen joints and painful joints, representative of those for other PD measures, are shown in **Figure 2a,b**, respectively. These figures both show a plateau at ~2,000 ng/mL, with no increase in percent change from baseline in either swollen joints (**Figure 2a**) or painful joints (**Figure 2b**), despite increased etanercept concentrations.

DISCUSSION

The goal of this article was to present this previously unpublished study that includes two important changes in the guidelines for the development of medications to treat RA: i) make early assessments of clinical response; ii) to use PK/PD assessments to plan further studies. The analyses conducted may not reflect all of the advances that have occurred in the 20 years since this study was conducted, but they are the ones that were used at the time and were used to inform subsequent work.

Although conditions were not optimal for assessment of PK parameters, due to the small number of observations and the approximate timing of collection with respect to dose, a reasonable approximation was made. The PK observations in this study were similar to those reported elsewhere.¹¹ Exposure was proportional to dose received. In this study,

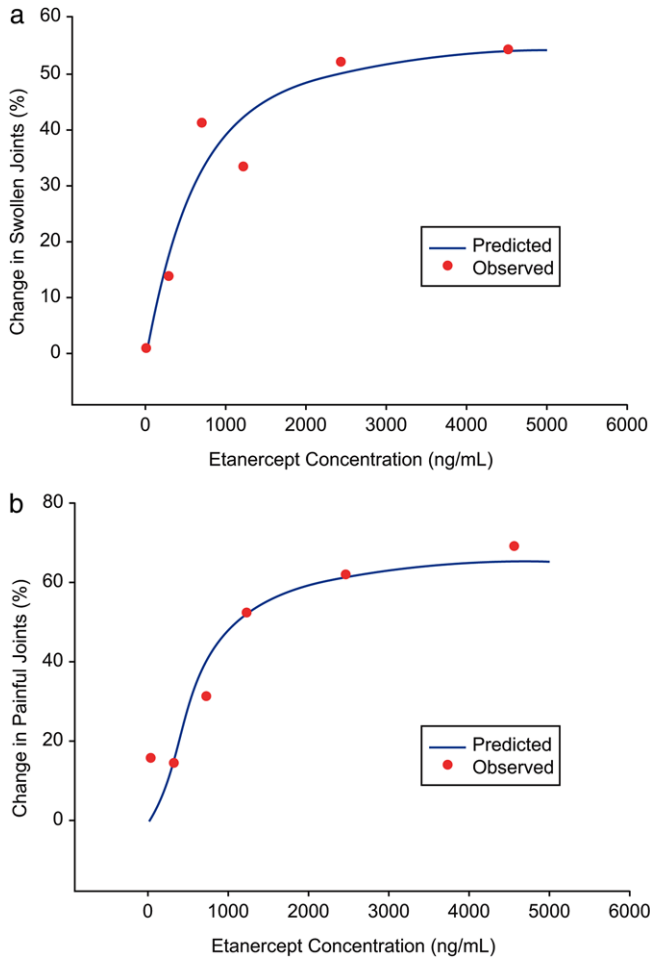


Figure 2 (a) Mean change from baseline in swollen joints vs. mean etanercept concentration; (b) Mean change from baseline in painful joints vs. mean etanercept concentration.

the mean C_{ss} was 1,170 ng/mL in patients receiving 50 mg once-weekly, which is consistent with the AUC calculation of 143,600 ng·h/mL after a 25-mg dose (~965 ng/mL) that was reported in a study of 25 patients with RA who received 25 mg twice-weekly for 6 months and had PK assessed at the beginning and end of the study.¹¹

In an early study of patients with RA treated with etanercept by both intravenous (i.v.) and subcutaneous (s.c.) administration, Moreland *et al.*⁵ reported they were unable to discriminate between the doses administered. Their study was similar in design to the current study; however, it included only 22 patients, of whom 18 received active drug. The administration of i.v. doses prior to starting s.c. twice weekly doses may have obscured the difference between dose groups. Additionally, the range of doses was lower than in the current study, with only the highest dose group (16 mg/m²/week (~50 mg/week)) in the range of what has been shown to be effective. Although etanercept concentrations were collected during that study, they were not used for PK/PD analysis.

The current study utilized all of the available data from all patients, even those receiving placebo. Lacking enough

data to support a population PK/PD analysis, an E_{max} model was empirically applied to the broad range of observations collected in the small group of patients studied. An effort to weight values somewhat evenly across the range was made by permitting uneven interval lengths that included similar numbers of observations. This study showed that after 4 weeks, further improvement in PD measures was not observed, as concentrations increased above ~2,000 ng/mL. Studies of longer duration might be expected to have different results. However, after 1 month of treatment, the EC_{50} values for swollen and painful joints and ESR were similar (range 465–573 ng/mL), suggesting that doses must achieve at least these concentrations. Thus the 10 mg once-weekly dose, which resulted in a C_{ss} of 300 ± 135 ng/mL (Table 3), would not be effective in most patients, and a 50 mg twice-weekly dose would be excessive; observations that were confirmed in the longer studies by Moreland *et al.*^{7,8} and Johnsen *et al.*¹⁰ The approved dosage for the treatment of RA is 50 mg per week administered in a single 50-mg dose or as divided doses of 25 mg twice-weekly; therefore, the results of the optimal concentration range in this dose-finding study are consistent with what was ultimately shown to be the effective dose.

C_{ss} was the PK parameter used in this PK/PD analysis. Other parameters, such as C_{max} or AUC, were not explored and may represent a limitation. The limitation, particularly in a short study such as the current study, is mitigated by the observation that etanercept exhibits linear PK and so C_{max} and AUC would be expected to vary in direct relation with C_{ss} .¹¹ An exposure–effect relationship conducted by Lee *et al.*, which explored the probability of achieving ACR20 after 6 months of treatment in 182 patients with RA, used cumulative AUC, calculated from the first dose, as the measure of etanercept exposure.¹² These investigators suggested that cumulative AUC was a better measure of exposure than C_{ss} because it better reflected the entire 6-month drug experience rather than the more limited C_{ss} observed at 6 months. However, trough concentrations have successfully been used by other investigators to evaluate clinical response, as will be discussed further below.

The duration of this study was short compared with most PK/PD studies. However, on the basis of better dose discrimination, the revised US guideline encourages assessment of treatment in the 2- to 8-week window after treatment initiation, as early responses point to more effective therapy.⁴ At the end of 4 weeks of treatment, it was reasonable to assume that steady-state etanercept concentrations had been achieved and that patients would have achieved some measure of relief of their symptoms. Patients in other dose-finding studies reported improvement in their symptoms as early as 2 weeks after starting twice-weekly treatment.^{7,8} The range of doses and concentrations for studies of shorter duration, such as the current study, can be broader than is possible for many conventional clinical studies because patients receiving inadequate treatment in those studies will tend to discontinue the study drug and/or safety concerns may be higher for studies of longer duration.

This study and discussion focused primarily on the translation of initial PK/PD information into a dosing range that would be reasonable to evaluate in larger phase III clinical

trials. It was performed across a wide range of doses, in patients who were treated for only a short period of time, and was able to obtain PD assessments for painful and swollen joints, ESR, IL-6, and MMP-3. Thus, this study design may offer useful information prior to the larger and longer studies that are necessary, given the relatively large number of patients suffering from the disease and the duration of their expected treatment.

The dose-finding studies conducted during etanercept development as well as the PK/PD analysis conducted in the current study supported the use of 50 mg weekly and concluded that further dose escalation was unlikely to be helpful; however, patients differ in both their PK parameters and their response to treatment. Early dose-finding studies were largely based on clinical findings, but further investigation of PK may be warranted. Two more recent studies in TNF-inhibitor-naïve patients with RA treated with etanercept 50 mg weekly (as either a single dose or two divided doses) reported that higher serum concentrations are associated with better clinical outcomes. In a study of 19 patients with active RA reported by Daien *et al.*, 3-month etanercept concentrations were found to be lower in six nonresponders (median 1,750 ng/mL) than in 13 responders (median 3,700 ng/mL, $P = 0.03$) after 6 months of treatment and correlated significantly with change in disease activity score in 28 joints between baseline and 6 months ($r = -0.62$, $P = 0.006$).¹³ Although all of the patients in the study were treated with 50 mg weekly and there was no change in the dose administered, the authors suggested that the absence of an optimal clinical response to etanercept may have been due to low etanercept concentrations and they postulated that patients with low etanercept levels may benefit from either an increased treatment dose or earlier change of treatment. The concentrations observed by these investigators are higher on average than those observed in the current study, and may reflect differences in administration technique, compliance, sampling time, and assay. There may be other differences between the patients that could explain their differences in response, such as differing from each other in their disease activity, but considering increasing doses rather than changing medications may be a reasonable choice for some patients.

Results from a study by Jamnitski *et al.* in 292 patients with RA also demonstrated that lower etanercept levels were associated with a lack of response to therapy.¹⁴ Among patients who were treated with 50 mg weekly, administered in either one or two doses, etanercept levels were significantly higher ($P < 0.05$) in patients with good European League Against Rheumatism (EULAR) responses (median 3,780 ng/mL; interquartile range (IQR) 2,530–5,170 ng/mL) than in those with moderate EULAR responses (median 3,100 ng/mL; IQR 2,120–4,470 ng/mL) or nonresponders (median 2,800 ng/mL; IQR 1,270–3,930 ng/mL) following 6 months of therapy. The concentration ranges overlap and were higher than observed for the same dose in the current study. The authors suggested that therapeutic drug monitoring and adjusted dosing regimens might be needed in certain groups of patients to obtain optimal response.

Extrapolating from the results of the Daien *et al.*¹³ and Jamnitski *et al.*¹⁴ studies, the relationship between etanercept

concentrations and effect needs additional exploration. A study demonstrating improvement in clinical response with changes in dosing and etanercept concentration would fill a gap in our current knowledge, as would better understanding of individual patient characteristics associated with less than optimal response. The target range of etanercept concentrations may indeed be higher than anticipated from the current study for some populations of patients.

Although an exhaustive review of the many studies on drugs in development for the treatment of RA is outside the scope of this study; there are other examples of small (fewer than 100 patients), short (no longer than 1 month) studies with a wide range of doses (greater than fivefold) that include PK assessment to permit further exploration of response information collected.¹⁵ This approach, a small, parallel-dose study of patients with active disease of very short duration, may also be appropriate in other disease areas.

In conclusion, sigmoidal E_{\max} models were successfully applied to mean PD observations and steady-state etanercept concentration data collected from a small number of patients with RA administered doses that ranged 10-fold, from 10 mg once-weekly to 50 mg twice-weekly. The EC_{50} values suggest that doses ranging between 10 mg twice-weekly and 50 mg once-weekly would provide concentrations associated with clinical response; however, doses outside the range would either be ineffective or would add no additional benefit.

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