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RHEUMATOID ARTHRITIS TREATED WITH TENIDAP AND PIROXICAM

Clinical Associations with Cytokine Modulation by Tenidap

BRUCE H. LITTMAN, CELENA E. DRURY, RUPERT O. ZIMMERER, CATHARINE B. STACK, and C. GORDON LAW

Objective. **To compare the effects of tenidap and piroxicam on acute-phase protein and cytokine levels in the blood of rheumatoid arthritis (RA) patients and to explore their associations with clinical disease activity.**

Methods. **A double-blind, randomized, crossover trial in 49 patients with active RA compared 6 weeks of treatment with tenidap (120 mg/day) versus 6 weeks of treatment with piroxicam (20 mg/day).**

Results. **Median values for C-reactive protein (CRP), Westergren erythrocyte sedimentation rate (ESR), serum amyloid A (SAA) protein, and interleukin-6 (IL-6) were significantly lower after tenidap treatment compared with piroxicam treatment, even in the presence of stable background treatment with prednisone, methotrexate, or prednisone plus methotrexate. The median within-patient treatment differences (after tenidap minus after piroxicam) in the CRP, ESR, SAA, and IL-6 values were −1.7 mg/dl, −10.0 mm/hour, −22.0 μg/ml, and −3.7 pg/ml, respectively, and represent −60.4%, −17.7%, −35.5%, and −26.1% of the respective baseline levels. IL-6 levels were positively correlated with CRP and SAA. Plasma IL-1β was generally below the level of detection. Tumor necrosis factor α levels were similar after tenidap and after piroxicam. Treatment differences for 4 of 7 clinical parameters favored tenidap, but did not reach statistical significance. IL-6, CRP, and ESR were significantly correlated with clinical treatment differences. Tenidap and piroxicam toleration were similar, although tenidap-treated patients exhibited a reversible increase in urinary protein excretion.**

Conclusion. **Tenidap was differentiated from piroxicam by lower levels of acute-phase proteins, ESR, and IL-6 after tenidap treatment. These treatment differences were significantly correlated with clincial parameters.**

Tenidap sodium is a new antiinflammatory/antirheumatic drug that is chemically unrelated to nonsteroidal antiinflammatory drugs (NSAIDs). In clinical trials in patients with rheumatoid arthritis (RA), treatment with tenidap resulted in a rapid and sustained reduction of blood levels of acute-phase proteins such as C-reactive protein (CRP) and serum amyloid A (SAA) protein (1,2). Tenidap treatment also led to a reduction of the erythrocyte sedimentation rate (ESR) (1,2), which is determined primarily by plasma levels of fibrinogen, another acute-phase protein (3).

Production of acute-phase proteins by the liver is regulated by cytokines, including interleukin-6 (IL-6), interleukin-1 (IL-1), and tumor necrosis factor α (TNFα) (4–6). These cytokines are produced by inflamed synovial tissues (7–10) and are present at higher levels in synovial fluid than in blood (11–15). IL-6 levels in blood are easily measured by immunoassay and bioassay, and have been shown to correlate with levels of CRP and with some clinical measures of disease activity (15–18). It has been postulated that cytokines produced in the joint, including IL-6, act as blood-borne hormones, delivering to the liver a signal that is partly responsible for the acute-phase response (19,20). This hypothesis is supported by data from RA clinical studies comparing plasma and synovial fluid cytokine levels in the same patient and by data from

Bruce H. Littman, MD, Celena E. Drury, RN, MBA, Rupert O. Zimmerer, PhD, Catharine B. Stack, MS, C. Gordon Law, PhD: Pfizer Incorporated, Groton, Connecticut.

Address reprint requests to Bruce H. Littman, MD, Central Research Division, Pfizer Inc., Eastern Point Road, Groton, CT 06340.

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studies of humans and nonhuman primates in which intravenous administration of human recombinant cytokines such as IL-1, TNFα, and IL-6 resulted in increased production of acute-phase proteins (11,12, 15,21–23). Recently, IL-6 has also been implicated as an important regulator of the stress response through interactions with the neuroendocrine system (24–26) and as a mediator of inflammatory hyperalgesic pain (27).

Tenidap has been shown to inhibit the production of IL-1, IL-6, and TNFα by human monocytes in vitro (28) and to inhibit the response of cells to cytokines (29,30). These in vitro activities of tenidap suggest that its ability to reduce acute-phase protein levels in RA patients could be due to inhibition of the production of cytokines at sites of inflammation and/or a reduction in the response to cytokines. The purpose of the present study was to test the hypothesis that tenidap treatment, as compared with piroxicam treatment, causes a reduction in acute-phase proteins and that this reduction is associated with a decrease in plasma levels of cytokines. Because of the large interpatient variability of these biochemical and clinical measurements, we used a crossover design to compare the intrapatient effects of 6 weeks of treatment with piroxicam with 6 weeks of treatment with tenidap. Since both tenidap and piroxicam are potent nonselective inhibitors of cyclooxygenase, this study design allowed the exploration of clinical and biochemical associations with the additional cytokine-modulating activities of tenidap (31).

PATIENTS AND METHODS

Clinical study design. A 12-week, double-blind, crossover study was conducted, comparing the within-patient biochemical and clinical effects of 6 weeks of piroxicam (20 mg/day) with 6 weeks of tenidap (120 mg/day) treatment. Patients were seen for a screening visit 4–14 days before entering the study. Patients were required to have adult-onset RA (according to the American College of Rheumatology [ACR; formerly, the American Rheumatism Association] 1987 revised criteria [32]) of at least 6 months' duration, be classified in ACR functional class I, II, or III (33), have a CRP level $\geq$1.5 mg/dl, and have active disease (defined as a minimum of 10 swollen joints and at least 2 of the following 3 findings: >12 joints tender on pressure and/or painful on motion, >1 hour of morning stiffness, or a Westergren ESR >25 mm/hour for males or >35 mm/hour for females). All patients had to be receiving an "adequate" therapeutic dose of an NSAID at the time of screening. Patients treated with piroxicam in the month prior to entry were not eligible for this study.

Patients were allowed to continue background disease-modifying antirheumatic drugs (DMARDs), including injectable gold, auranofin, D-penicillamine, methotrexate, hydroxychloroquine, or azathioprine, provided the dose and frequency of administration was stable for 3 months prior to initiation of the study drug. This therapy had to be continued unchanged during the study. In addition, patients were allowed to continue corticosteroid therapy equivalent to an average daily dose of $\leq$10 mg of prednisone, provided that the dosage had been stable for at least 1 month prior to study entry. The corticosteroid therapy could not be changed during the study. Thus, any background corticosteroid or DMARD therapy had to be identical during piroxicam and tenidap treatment periods. Patients with any other condition considered likely to influence acute-phase protein or cytokine levels were excluded. For example, patients with other inflammatory diseases, infections, burns, or major surgical operations within 1 month of study entry were excluded.

Patients were instructed to discontinue their current NSAID therapy after the last dose on the day prior to the baseline visit. At baseline, patients were randomly assigned to double-blind therapy with either tenidap or piroxicam. After treatment in the first 6-week period (weeks 1–6), patients were switched, without a washout, to treatment with the other agent for the second 6-week period (weeks 7–12). Patients were evaluated at baseline and at weeks 1, 3, 6, 7, 9, and 12. Laboratory safety studies, standard clinical measurements, and levels of ESR, CRP, and SAA were determined at each visit; plasma for cytokine determinations was obtained at all but the week-1 and week-7 visits.

Laboratory safety tests. A complete blood cell count, platelet count, urinalysis, and levels of serum electrolytes, serum creatinine, blood urea nitrogen, bicarbonate, inorganic phosphate, calcium, bilirubin, total protein (albumin and globulins), iron, glucose, cholesterol, triglycerides, and liver enzymes (alkaline phosphatase, gamma glutamyl transferase, serum glutamic oxaloacetic transaminase, and serum glutamic pyruvic transaminase) were obtained at each visit.

To specifically monitor for proteinuria, quantitative urinary protein and creatinine (to calculate protein:creatinine ratio) determinations were performed at every visit. Urinary protein excretion was quantitated because tenidap has previously been shown to increase the excretion of protein, uric acid, and phosphate, probably due to reversible inhibition of renal proximal tubule function. The crossover design of this study allowed for an evaluation of the reversibility of tenidap's effect on protein excretion and for a comparison with piroxicam.

Clinical measurements. Patient self-assessments included a pain score (using a visual analog scale of 0–31), global assessment of disease activity (1–5 scale), and duration of morning stiffness (in minutes) based on the preceding 3 days. At the last visit, patients were also asked to indicate their preferred treatment (first 6 weeks or second 6 weeks) and rate it on a 0–3 scale, where 0 = no difference, 1 = slightly better, 2 = moderately better, and 3 = markedly better.

Physician assessments included an evaluation of 68 joints for pain/tenderness and 66 joints (hips excluded) for swelling. The degree of pain and/or tenderness and the degree of swelling were rated on a 0–3 scale (0 = no pain on palpation; 3 = withdrawal). The results of the joint evaluations are reported as a joint count (number of joints rated

Table 1. Clinical efficacy parameters in 49 rheumatoid arthritis patients*

Efficacy parameter	Median value at baseline	Within-patient treatment difference (tenidap − piroxicam)		
		Median	% of baseline	*P*
Patient global assessment of disease activity (1–5)	3	0	0.0	0.0563
Visual analog pain scale (0–31)	20	−2	−10.0	0.1478
Duration of morning stiffness (minutes)	120	0	0.0	0.9886
Number of painful joints (0–68)	26	−3	−11.5	0.4147
Sum of joint pain scores (0–204)	38	−3	−7.9	0.1871
Number of swollen joints (0–66)	21	0	0.0	0.3474
Sum of joint swelling scores (0–198)	29	−2	−6.9	0.2508

* *P* values determined by Wilcoxon's signed rank test. See Patients and Methods for details of scoring systems.

>0) and as a joint score (sum of all joint ratings; maximum of 204 for pain/tenderness and 198 for swelling). Grip strength (measured with a hand dynamometer), physician global assessment, and functional capacity were also evaluated, but the results are not included in this report.

C-reactive protein. Serum CRP levels were determined by a central laboratory (SmithKline Bio-Science Laboratories) using a rate nephelometry assay. The lower limit of detection for the assay was 0.1 mg/dl. Values less than 0.1 mg/dl were conservatively treated as 0.09 mg/dl.

Serum amyloid A. SAA was determined by immunoassay on heparinized plasma, using a commercial kit (Hemagen, Waltham, MA) as previously described (34). The lower limit of detection was 5 μg/ml, and values over 100 μg/ml were re-assayed after dilution. Values given are the means of duplicate determinations.

Erythrocyte sedimentation rate. The ESR (Westergren) was determined locally at the time of the patient's visit.

Cytokine assays. All cytokine assay methods were validated first by confirming that cytokine readings were fully neutralized using specific antibody, that the assay could detect 100% of the cytokine spiked into RA plasma, and that all readings were in the linear portion of the standard curve. All serum and plasma samples were freshly obtained by separation in a refrigerated centrifuge (4°C) and stored frozen at −70°C until tested. All samples from a given patient were assayed together.

Plasma levels of IL-6 were determined by enzyme-linked immunosorbent assay (ELISA) using the Quantikine immunoassay kit (R&D Systems, Minneapolis, MN). Sensitivity was increased by using the ELAST ELISA Amplification System (Du Pont NEN Research Products, Boston, MA), giving an effective range of 1–190 pg/ml.

Plasma levels of IL-1β were determined by ELISA using a modification of the Quantikine immunoassay kit that also used the ELAST ELISA Amplification System. The effective range of this assay was also 1–190 pg/ml. Since RA plasma frequently had undetectable levels of IL-1β, validation work with this kit was also conducted using RA synovial fluid. Synovial fluid samples from RA patients were found to contain IL-1β, which was fully neutralized by specific antibody to IL-1β (data not shown).

Plasma levels of TNFα were determined by ELISA using the Cytoscreen immunoassay kit (N. V. Innogenetics, Antwerp, Belgium). The assay protocol was modified to increase sensitivity by substituting poly-HRP20-Streptavidin (Research Diagnostics, Flanders, NJ) for the horseradish peroxidase–conjugated streptavidin provided in the kit. The effective range of the assay was 4 to 150 pg/ml.

Statistical methods. For descriptive purposes, the median within-patient changes in CRP are presented graphically for the 2 sequence groups. However, because this was a crossover study with no washout periods, the primary analysis of clinical and biochemical parameters was a within-patient treatment difference of the final value with tenidap treatment compared with the final value with piroxicam treatment. Mathematically, this is equivalent to the change from baseline to the end of piroxicam treatment subtracted from the change from baseline to the end of tenidap treatment. This analysis was chosen in order to avoid any possible carry-over effects, and therefore assumed that any effects of the previous treatment would no longer be present 6 weeks after it was stopped. This assumption was confirmed statistically since no significant period or sequence effects were present (data not shown).

The significance of the tenidap–piroxicam comparison was determined using Wilcoxon's signed rank test on the difference in the final results for each patient. Medians were determined and are presented in this report (rather than the means) because the data for these parameters were not clearly normally distributed. In addition, to give some perspective to the quantitative difference between the effects of piroxicam and tenidap, the median values for the differences in the final results for each patient are presented as a percentage of the median baseline value.

It was of interest to explore correlations between clinical treatment differences and treatment differences for acute-phase proteins, ESR, and IL-6 levels. A nonparametric method was used for the reasons indicated above. Spear-

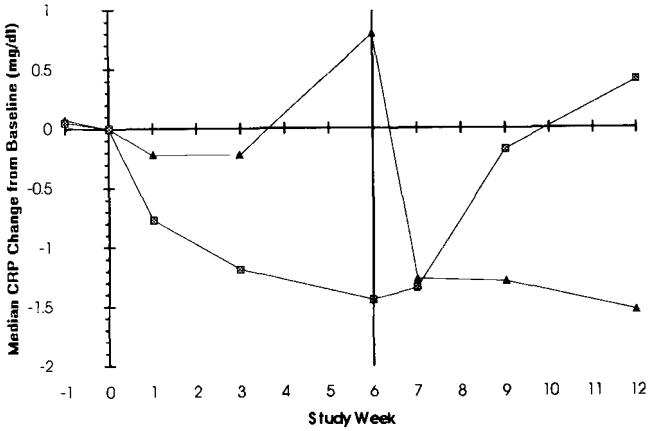

Figure 1. Median within-patient changes in C-reactive protein (CRP) over time, in 27 rheumatoid arthritis (RA) patients who received tenidap during weeks 1–6 and piroxicam during weeks 7–12 (squares) and in 22 RA patients who received piroxicam during weeks 1–6 and tenidap during weeks 7–12 (triangles). Week −1 is the screening visit, and week 0 is the baseline visit.

man's correlation coefficients and their significance were calculated using the within-patient treatment difference data.

RESULTS

Baseline characteristics of the study patients. Seventy patients were randomized for treatment, but since this was a crossover study, only the results from the 49 patients who completed both treatments are presented. The reasons for discontinuation included lack of efficacy (3 during tenidap and 3 during piroxicam), gastrointestinal side effects (1 during tenidap and 2 during piroxicam), laboratory abnormalities (1 during tenidap and 1 during piroxicam), intercurrent illnesses, and administrative reasons.

Of the 49 patients who completed the study, 22 received piroxicam first and then tenidap, and 27 received tenidap first and then piroxicam. The median duration of RA in these 49 patients was 12.5 years, and their median age was 56.8 years. The group was predominantly seropositive (77.6%) and female (69.4%). Thirty patients were receiving stable background treatment with either prednisone, methotrexate, or both. Table 1 includes the median baseline measures of clinical activity.

Treatment differences for clinical efficacy parameters. Tenidap was quantitatively superior to piroxicam for 3 pain-related measures and for the sum of joint swelling scores; however, none of these treatment differences reached statistical significance (Table 1). There were no quantitative differences between

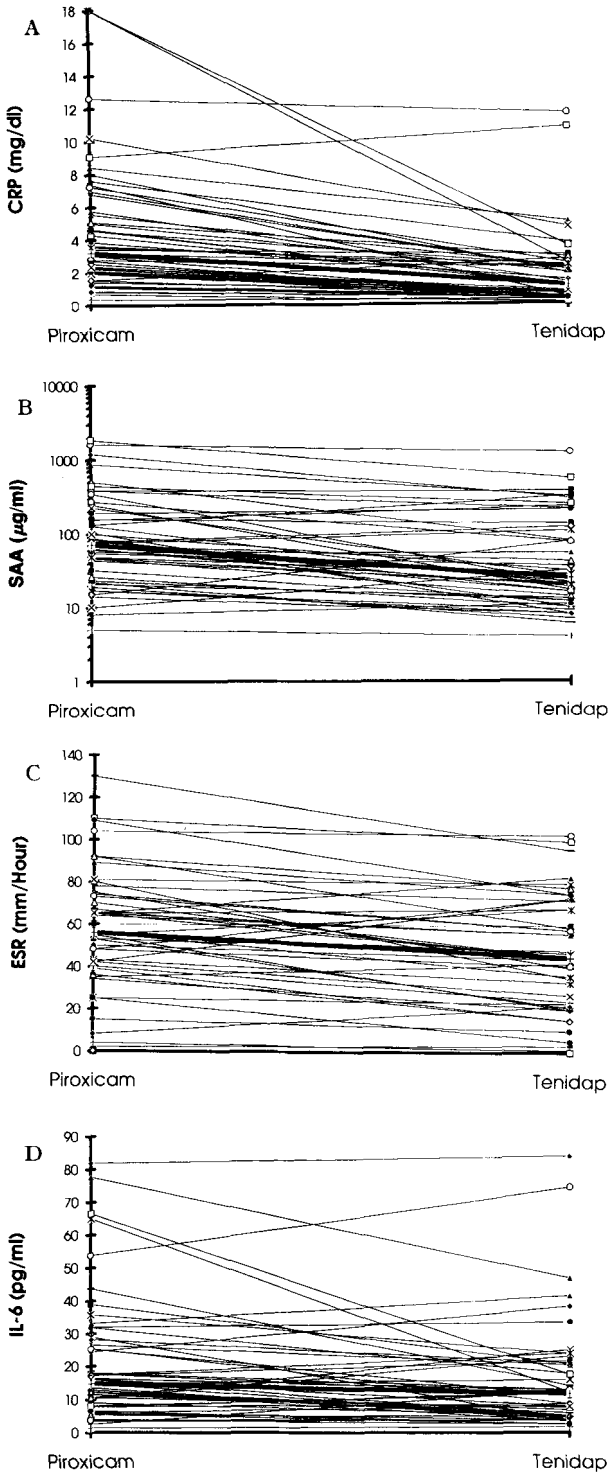

Figure 2. Values for **A**, C-reactive protein (CRP), **B**, serum amyloid A (SAA) protein, **C**, erythrocyte sedimentation rate (ESR), and **D**, interleukin-6 (IL-6) in individual rheumatoid arthritis patients after receiving 6 weeks of piroxicam treatment and after 6 weeks of tenidap treatment. Heavy bars show the median.

Table 2. Treatment differences for acute-phase proteins, ESR, and IL-6 values, according to background treatment*

	No. of patients	Median value at baseline	Within-patient treatment difference (tenidap − piroxicam)		
			Median	% of baseline	*P*
All patients					
CRP (mg/dl)	49	2.8	−1.7	−60.4	0.0001
ESR (mm/hour)	44	56.5	−10.0	−17.7	0.0001
SAA (μg/ml)	49	62.0	−22.0	−35.5	0.0001
IL-6 (pg/ml)	49	14.2	−3.7	−26.1	0.0078
Background treatment					
Prednisone					
CRP (mg/dl)	23	2.7	−1.8	−64.6	0.0001
ESR (mm/hour)	21	52.0	−20.0	−38.5	0.0003
SAA (μg/ml)	23	126.0	−32.0	−25.4	0.0026
IL-6 (pg/ml)	23	14.0	−7.1	−50.7	0.0092
Methotrexate					
CRP (mg/dl)	15	3.6	−1.8	−49.4	0.0001
ESR (mm/hour)	14	55.0	−17.0	−30.9	0.0188
SAA (μg/ml)	15	66.0	−20.0	−30.3	0.0125
IL-6 (pg/ml)	15	15.1	−4.9	−32.5	0.0833
Prednisone + methotrexate					
CRP (mg/dl)	8	3.1	−1.7	−55.5	0.0078
ESR (mm/hour)	8	58.0	−21.0	−36.2	0.0078
SAA (μg/ml)	8	216.5	−26.0	−12.0	0.1094
IL-6 (pg/ml)	8	14.9	−6.2	−41.4	0.0391

* *P* values determined by Wilcoxon's signed rank test. ESR = erythrocyte sedimentation rate (Westergren); IL-6 = interleukin-6; CRP = C-reactive protein; SAA = serum amyloid A (protein).

tenidap and piroxicam for the number of swollen joints, patient's global assessment of disease activity, or duration of morning stiffness. Interestingly, when these 49 patients were asked to rate the relative effectiveness of the 2 treatments, 24 preferred tenidap, 17 preferred piroxicam, and 8 had no preference. Among those who reported marked improvement, 13 patients noted this during tenidap and 7 during piroxicam treatment.

Median changes in CRP levels. To illustrate how the biochemical changes seen in this study mirrored the crossover design, the median within-patient change in CRP from the pretreatment baseline value was plotted for both sequence groups (Figure 1). Week −1 is the screening visit, week 0 is the baseline visit, and subsequent time points are the weeks after starting study drug.

For patients randomized to receive tenidap in the first 6 weeks followed by piroxicam in the second 6 weeks (n = 27), there was a rapid initial reduction in the CRP. After week 6, when the treatment was switched to piroxicam, the CRP levels rapidly increased, such that by week 12 they had returned to the levels found at study entry. For patients who received piroxicam first (n = 22), there was little change in the CRP value until the tenidap treatment was begun. The CRP levels then decreased markedly during the second 6 weeks of the study. A similar pattern of response was seen for SAA levels, the ESR, and IL-6 levels.

Correlation of CRP, SAA, and ESR with IL-6. In an effort to determine whether IL-6 values were correlated with acute-phase proteins and the ESR, a correlation coefficient (r) for these values was calculated for each patient using the baseline and week 3, 6, 9, and 12 values (5 time points). Wilcoxon's signed rank test was then performed on the list of r values for each correlation. IL-6 was significantly correlated with CRP (median r = 0.44, $P = 0.0001$) and with SAA (median r = 0.39, $P = 0.0001$). IL-6 was not significantly correlated with ESR (median r = 0.07, $P = 0.3681$).

Treatment differences for acute-phase proteins, ESR, and IL-6. The posttreatment values for CRP, SAA, ESR, and IL-6 for each patient and the median values for these parameters are illustrated in Figure 2. Note that for each of these, the level after 6 weeks of piroxicam was generally higher than the level after 6 weeks of tenidap. Statistical analyses of these treatment differences are presented in Table 2.

For each patient the value at the end of piroxicam treatment was subtracted from the value at the

Table 3. Correlation analysis of clinical and biochemical treatment differences*

	CRP	ESR	SAA	IL-6
Patient global assessment				
r	0.19	0.22	−0.10	0.28
P	0.181	0.151	0.480	0.048
Visual analog pain scale				
r	0.27	0.33	0.01	0.31
P	0.058	0.028	0.961	0.028
Duration of morning stiffness				
r	0.09	0.30	−0.15	0.27
P	0.557	0.045	0.307	0.063
Number of painful joints				
r	0.23	0.13	−0.09	0.24
P	0.115	0.410	0.532	0.094
Sum of painful joint scores				
r	0.24	0.09	−0.10	0.24
P	0.095	0.543	0.490	0.096
Number of swollen joints				
r	0.24	0.12	−0.23	0.02
P	0.090	0.454	0.112	0.902
Sum of swollen joint scores				
r	0.30	0.20	−0.17	0.00
P	0.035	0.198	0.255	0.987

* Spearman correlation coefficients and significance are shown. CRP = C-reactive protein; ESR = erythrocyte sedimentation rate (Westergren); SAA = serum amyloid A (protein); IL-6 = interleukin-6.

end of tenidap treatment. Using CRP as an example, this median treatment difference for all 49 patients was −1.7 mg/dl, and this difference was significantly different from 0 ($P = 0.0001$). The values for SAA, ESR, and IL-6 are similarly presented. Tenidap treatment also resulted in significantly lower levels of SAA ($P = 0.0001$), ESR ($P = 0.0001$), and IL-6 ($P = 0.0078$) than piroxicam treatment.

To put the treatment difference into perspective, we have provided in Table 2 the median baseline and the percentage of baseline that the treatment difference represents. Using CRP as an example, for all 49 patients, the −1.7 mg/dl median treatment difference was equivalent to −60.4% of the median baseline level of 2.8 mg/dl.

Table 2 also shows the values for each subset of patients who were taking stable background treatment (prednisone, methotrexate, or prednisone plus methotrexate). The median treatment differences for all parameters were quantitatively similar for all background treatment subsets. In addition, these differences remained significant for all parameters in patients taking prednisone (the largest subset), and for 3 of 4 of these parameters in the smaller subsets of patients taking methotrexate and taking methotrexate plus prednisone. Therefore, the observed ability of tenidap treatment to decrease levels of acute-phase proteins, ESR, and IL-6 in all 49 patients who completed the study was present in the subsets of patients taking background prednisone, methotrexate, and the combination of methotrexate and prednisone.

Levels of TNFα. TNFα was assayed in plasma samples from 43 of the 49 patients who completed the study. Only 4 of the 43 patients did not have detectable levels of TNFα at baseline. The median baseline level of TNFα was 12.8 pg/ml, the median level after tenidap treatment was 10.2 pg/ml, and the median level after piroxicam treatment was 9.8 pg/ml. The median within-patient difference in TNFα (after tenidap minus after piroxicam) was 0.21 pg/ml, and was not significant. Thus, tenidap and piroxicam treatment resulted in similar plasma levels of TNFα.

Levels of IL-1β. IL-1β was assayed in plasma samples from all patients but was above the lower limit of detection in only a few. As a result, the effects of tenidap and piroxicam on plasma levels of this cytokine could not be determined.

Correlation of clinical and biochemical treatment differences. Results of the correlation analysis of clinical with biochemical treatment differences are presented in Table 3. In general, correlations were noted for CRP with both pain and joint swelling parameters. The ESR was significantly correlated with pain and morning stiffness. IL-6 was significantly correlated with pain-related parameters and not at all correlated with joint swelling. SAA was not correlated with any of the clinical treatment differences.

Urine protein excretion. Urinary excretion of protein increased during tenidap treatment. For the 27 patients who received tenidap and then piroxicam, the median ratio of protein to creatinine (P:C) increased from 0.07 to 0.15 at 6 weeks. This increase was rapidly reversible. By 3 weeks after switching to piroxicam, this ratio decreased to 0.08. For the 22 patients who received piroxicam and then tenidap, the P:C ratio increased slightly with piroxicam, from 0.07 to 0.11, and increased further with tenidap, to 0.15. This P:C value of 0.15 translates into an estimated 24-hour protein excretion of about 127 mg. Reversible increases in protein excretion after long-term treatment with tenidap have previously been reported (35). This is thought to be due to reversible inhibition of renal proximal tubule function, which includes reabsorption of small filtered plasma proteins.

DISCUSSION

Acute-phase protein production by the liver is known to be regulated by cytokines such as IL-6, TNFα, and IL-1 (4–6), and blood levels of these cytokines, which are produced by inflamed synovial

tissues (7–10), are reported to correlate with clinical disease activity (13,15–18). Radiographic progression of RA has also been correlated with chronically elevated CRP (36–38). We reasoned that patients with both active disease and elevated levels of acute-phase proteins would therefore be more likely to have measurable levels of these cytokines in the blood. Patients with active RA and elevated CRP (≥1.5 mg/dl), despite their current therapy, were thus selected for study. We tested the hypothesis that the effects of tenidap on acute-phase proteins occurred in conjunction with similar effects on cytokines and that this activity would not be shared by piroxicam, an NSAID. In addition, we studied treatment differences in clinical efficacy so that we could evaluate possible relationships with treatment differences in acute-phase protein and cytokine levels. Since we studied each treatment for only 6 weeks, we did not expect to find any significant clinical treatment differences between these 2 potent cyclooxygenase-inhibiting agents. Tenidap's superior efficacy at 6 months of treatment (compared with naproxen and piroxicam) has previously been shown (35,39).

The results reported here confirm the ability of tenidap treatment to reduce levels of acute-phase proteins (1,2) and demonstrate that this activity is not shared by piroxicam. The ability of antirheumatic drugs to lower levels of acute-phase proteins has been studied by other investigators. CRP levels have been examined in the most detail. Those studies have shown that NSAIDs generally do not affect levels of CRP (40–42). In one study, however, when only the data for the clinically responsive patients were considered, there was an apparent association of NSAID treatment with reduced levels of CRP (43). This is not surprising since many investigators have demonstrated that changes in CRP levels reflect clinical changes in RA (42–44). Unlike NSAIDs, treatment with corticosteroids and with some DMARDs, including gold salts, D-penicillamine, antimalarials, methotrexate, and sulfasalazine, has been associated with reduced levels of acute-phase proteins (40–42,44–49).

Our findings have also clearly demonstrated that tenidap treatment results in significantly lower levels of plasma IL-6 than does piroxicam treatment. Others have reported a reduction in IL-6 levels in RA patients treated with some DMARDs, including azathioprine (46), methotrexate (46), injectable gold salts (47), auranofin (49), and sufasalazine (48,49). The finding that tenidap treatment results in reduced levels of plasma IL-6 in patients with RA may reflect the cytokine-modulating activity of tenidap in synovial tissues. This could be of clinical importance since cytokines are known to be produced at the cartilage–pannus junction (10) and to regulate the production of collagenase, stromelysin, and other factors important in the pathogenesis of joint tissue injury (19).

The relationship between IL-6 levels and levels of the acute-phase proteins is of interest since IL-6 is known to regulate hepatic production of many acute-phase proteins (19,20). We found that within-patient levels of CRP and SAA were significantly correlated with those of IL-6 (regardless of treatment sequence) during the 12 weeks of this study. Although IL-6 has been shown to regulate hepatic production of fibrinogen (6), the plasma protein most directly related to the ESR (3), ESR values did not correlate with IL-6 levels. The plasma half-life of fibrinogen is 4 days, whereas the half-lives of CRP and SAA are approximately 19 hours and 24 hours, respectively. Thus, changes in plasma levels of fibrinogen as a result of changes in IL-6 may lag behind IL-6–induced changes in CRP and SAA, and may account for our finding. Alternatively, other factors influencing the ESR may overshadow the IL-6 changes.

We also found that the effects of tenidap on acute-phase proteins and IL-6 occurred even if patients were receiving background treatment with prednisone, methotrexate, or both. While treatment with either of these 2 drugs can reduce acute-phase protein levels (45,46), and methotrexate can reduce IL-6 (46), we selected study patients with active disease despite these background treatments. Presumably, this selected for patients with continued cytokine production. Since tenidap reduced IL-6 and acute-phase proteins in patients also receiving methotrexate and prednisone, our findings suggest that the mechanism for tenidap's cytokine-modulating activity may be different from that of these agents.

The clinical significance of these biochemical effects of tenidap was evaluated by determining whether clinical treatment differences between tenidap and piroxicam were statistically related to acute-phase protein, ESR, and IL-6 treatment differences. Clinical disease activity has previously been correlated with CRP and ESR (41–44). We also found clinical correlations with the CRP and ESR treatment differences, thus confirming the clinical relevance of these measures of the acute-phase response. CRP was generally correlated with joint swelling and pain, and the ESR was correlated with pain and morning stiffness.

Although other investigators have shown a correlation between IL-6 and some clinical findings, less is known about the clinical significance of an elevated

IL-6 level in RA patients (15–18). We found that the IL-6 treatment difference was significantly correlated with clinical efficacy treatment differences that are related to pain. This finding linking reduced IL-6 with reduced pain is interesting because of recent data establishing a role of IL-6 (and other cytokines) in the neuroendocrine stress response (24–26) and in inflammatory hyperalgesia (27). An interesting example is a study comparing the degree of pain, corticosteroid response, CRP level, and cytokine level in patients undergoing cholecystectomy by either laparotomy or laparoscopy. In that study, IL-6 levels also correlated with pain scores and CRP levels (50). Alternatively, lower IL-6 levels during tenidap treatment and lower levels of joint pain may simply reflect less synovial inflammation during tenidap treatment and less cytokine stimulation of hyperalgesic pain. This hypothesis is supported by animal studies, in which hyperalgesic pain could be blocked by antibodies to cytokines such as TNFα and IL-6 (27).

Tenidap and piroxicam were similarly tolerated among our patients. Clinical laboratory evaluation showed that only tenidap's ability to induce low-level proteinuria distinguished it from piroxicam. Quantitating proteinuria by measuring the urine protein:creatinine ratio enabled us to demonstrate this effect of tenidap and to document its reversibility during piroxicam treatment. Proteinuria was not clinically significant, and was usually manifested by trace or 1+ protein levels on dipstick analysis.

In summary, we used treatment differences to compare the status of individual patients after 6 weeks of treatment with tenidap and 6 weeks with piroxicam. Since both of these study drugs are potent cyclooxygenase inhibitors, the treatment differences presumably reflected other activities that are not shared by these drugs. Tenidap was clearly differentiated from piroxicam on the basis of differences in acute-phase proteins, ESR, and blood levels of IL-6. These biochemical differences were significantly correlated with differences in measures of clinical disease activity.

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