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Population pharmacodynamics of romazarit

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1 The response to romazarit, a disease modifying anti-rheumatoid agent, was observed in patients with rheumatoid arthritis (RA) in a double-blind placebo controlled study.
2 Two hundred and twenty-four patients were recruited from 11 centres and treated with placebo or romazarit at a dose of 200 mg or 450 mg every 12 h for up to 24 weeks. Disease activity was measured using the Ritchie Index (RI). Plasma concentrations of romazarit were measured at each of up to eight assessments of RI.
3 The effect of romazarit was examined using analysis of variance (ANOVA) in 164 patients who contributed 61% of observations of disease activity. Observations after 12 weeks of treatment were excluded from ANOVA.
4 A population pharmacokinetic-dynamic model for the time course of disease progress and the response to placebo and romazarit was used to describe observations from all patients.
5 The population model suggested that the effect of romazarit was on the rate of progress of the disease and was describable by an E_{max} model. Concentration was a better predictor of response than dose.
6 Romazarit was significantly better than placebo in improving the RI in patients with RA. The placebo efficacy of romazarit treatment was similar to that associated with placebo treatment.
7 The population model provided a more complete description and explanation of the clinical pharmacology and therapeutic potential of romazarit than ANOVA.

Keywords romazarit rheumatoid arthritis population pharmacodynamics clinical trial design

Introduction

Romazarit is rapidly and extensively absorbed but its pharmacokinetics are non-linear in healthy volunteers [1] and patients with rheumatoid arthritis (RA) [2]. The clearance of romazarit decreases with increasing dose perhaps due to saturable renal tubular secretion of romazarit ester glucuronide. This is the main metabolite appearing in the urine and it appears to undergo reversible metabolism to the parent compound *in vivo*. This non-linearity is expected to contribute to considerable inter-individual variability in romazarit concentrations.

Romazarit has shown disease modifying activity in animal models which mimic some aspects of human RA [3]. The plasma romazarit concentrations associated with efficacy in one of these models, Type II collagen arthritis, were in the range of 50–100 mg l^{-1} (0.16–3.2 mmol l^{-1}) (Bloxham, personal communication), a range that provided a target concentration for an efficacy study in patients with RA.

A Phase II efficacy study of romazarit has been performed in patients with RA. However, further clinical development of this compound has been

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stopped. We have used this study to demonstrate how a population pharmacodynamic analysis can enhance understanding of the clinical pharmacology of a drug at an early stage of drug development. A single measure of disease activity, the Ritchie Index, was chosen to illustrate the technique. Other disease markers and further details of the results of the clinical trial are not reported in order to focus on the methodological aspects of the population analysis approach.

Methods

Patients

Eleven investigational centres recruited patients with RA to a randomized, double-blind, placebo controlled, parallel group study. Each patient received either 200 mg romazarit or 450 mg romazarit or placebo tablets 12-hourly by mouth for up to 6 months. Immediately prior to starting treatment and 2, 4, 8, 12, 16, 20 and 24 weeks afterwards the Ritchie Articular Index (RI) [4] was measured. This index measures the response of the patient to manipulation of 26 joints. The response is graded: 0 = no tenderness, 1 = felt pain, 2 = felt pain and winced, 3 = felt pain, winced and withdrew. The maximum possible value of the index is 78. A value greater than 11 at screening was an entry criterion for the study. The diagnosis of RA was established using ARA criteria [5]. There was a wash-out period in which other disease modifying antirheumatic agents were discontinued for at least 6 weeks prior to treatment with romazarit or placebo. Concomitant treatment was stabilized for at least 2 weeks prior to romazarit or placebo. Necessary comedications were restricted to prednisolone (up to 7.5 mg day^{-1}) and one of the following non-steroidal anti-inflammatory drugs: indomethacin (up to 200 mg day^{-1}), ibuprofen (up to 2400 mg day^{-1}), naproxen (up to 1000 mg day^{-1}), piroxicam (up to 20 mg day^{-1}), ketoprofen (up to 200 mg day^{-1}).

Drug concentrations

At the same clinic visits as the RI assessment, a blood sample was taken for measurement of romazarit concentration in plasma. The time of sampling in relation to the last dose was noted. Plasma was stored at −20° C at each centre until despatch to the Pharmacokinetics and Metabolism Department, Roche Products, where they were stored at −20° C until assayed. The chromatographic method [1] used reverse-phase h.p.l.c. with u.v. detection. It was able to separate romazarit from all concomitant non-steroidal anti-inflammatory drugs used by the patients. The minimum quantifiable concentration was 0.1 mg l^{-1}. The assay was linear from 0.1 mg l^{-1} to 200 mg l^{-1} with a bias of 3% and precision of 4% over this range.

Analysis of variance model

Analysis of variance was performed using PROC GLM [6]. The model included effects of baseline RI, centre, treatment and centre by treatment interaction. The observations for the ANOVA were restricted to those collected during the first 12 weeks of treatment. Because of the increasing number of patients who did not complete the trial it was felt that there would be insufficient power to detect any difference that might have existed at later times.

Population pharmacokinetic-dynamic model

The time course of RI scores was predicted using a pharmacokinetic-dynamic model (PKPD) [7]. The observed response can be thought of as arising from the sum of three components:

1. A function ($S(t)$) describing the disease status as a function of time, e.g. a straight line with a Y intercept $S(0)$ (disease status at the start of treatment) and slope α:

$$S(t) = S(0) + \alpha \cdot t$$

2. The active treatment effect (PD_A) producing a change determined by the concentration of active drug at its effect site, $C_{e,A}$, e.g. using a linear model for the concentration effect relationship where β_A is a parameter reflecting the potency of the active drug:

$$PD_A\,(C_{e,A}) = \beta_A \cdot C_{e,A}$$

3. The placebo treatment effect (PD_P) producing a change which can be modelled by the hypothetical concentration of a placebo substance at its effect site, $C_{e,P}$, e.g. using a model similar to that for active drug where β_P reflects the potency of the placebo:

$$PD_P\,(C_{e,P}) = \beta_P \cdot C_{e,P}$$

The time course of the response is then given by:

$$S(t) = S(0) + \alpha \cdot t + PD_A(C_{e,A}) + PD_P\,(C_{e,P})$$

This model is known as the offset model because the effect of the active drug is equivalent to producing an offset to the $S(0)$ baseline intercept. A variant of the active drug treatment model is to propose that the active drug changes the rate of disease progression, α, instead of producing an offset. This model is known as the slope model:

$$S(t) = S(0) + (\alpha + PD_A(C_{e,A})) \cdot t + PD_P(C_{e,p})$$

A key feature of the models is to predict the time course of active drug or hypothetical placebo substance in terms of a concentration at the site of action. Delays in onset of active drug or placebo effect can be described by a pharmacokinetic model linking the central compartment (e.g. plasma) and the effect compartment. These pharmacokinetic models can accept doses of active drug or placebo at any time and thus accommodate a wide variety of clinical trial designs. The active substance is not necessarily romazarit but may be a physiological mediator whose concentration is modified by romazarit. The placebo substance is unidentified but is based on a conceptual

hypothesis to account for the time course of a placebo response.

When no plasma drug concentration measurements are available the average steady state plasma drug concentration (C_{av}) can be predicted from the daily dose and a nominal value for the clearance/bioavailability ratio (CL/*F*):

$$C_{av} = \frac{\text{Daily dose}}{CL/F}$$

The concentration of active drug in the effect compartment ($C_e(t)$) is then determined by a single parameter, the effect compartment equilibration half-time, $t_{1/2,eq,A}$:

$$C_e(t) = C_{av} \cdot (1 - e^{-\frac{\ln(2)}{t_{1/2,\,eq,\,A}} \cdot t})$$

The concentration of placebo substance in the effect compartment is modelled in a similar way but is considered to arise from a single bolus input of placebo at the start of the double-blind treatment phase rather than continuous input as for active treatment. The time course of placebo concentration in plasma is assumed to disappear with a half-time, $t_{1/2,el,P}$, while the placebo substance equilibration with its effect site is determined by the placebo equilibration half-time, $t_{1/2,eq,P}$. These two half-times determine the time course of placebo concentration at its effect site:

$$C_e(t) = \frac{1}{\frac{\ln(2)}{t_{1/2,eq,P}} - \frac{\ln(2)}{t_{1/2,el,P}}} \cdot (e^{-\frac{\ln(2)}{t_{1/2,\,eq,P}} \cdot t} - e^{-\frac{\ln(2)}{t_{1/2,\,el,P}} \cdot t})$$

Model implementation

Slope model A precise solution to the slope model would require the solution of a system of differential equations. Because of computational resource limitations we chose to approximate the solution in a piecewise fashion by extrapolating from one observation to the next using the slope predicted at the time of the earlier observation. The error arising from the piecewise approximation used with the slope model is greatest when effect site concentrations are not at steady state and the interval between observations is long compared with the equilibration half-time. In an attempt to minimize this error, which is greatest for the prediction of the first post treatment observation at 2 weeks (because this would be based on the slope predicted from a pre-treatment concentration of zero), the model used a value of plasma concentration immediately after starting treatment equal to the 2 week measured value.

Effect site concentrations The concentration of active substance at its effect site was predicted using an effect compartment model driven by the average steady state plasma concentration of romazarit. Romazarit concentrations were either predicted as being proportional to the dose or from the measured plasma concentration. The measured concentrations were treated as if they were equivalent to the average steady state concentration. The timing of the romazarit concentration measurement was scattered throughout the dosing interval so we expect that on average the measured values would be a reasonable approximation of the steady state value and potentially a better approximation than the dose alone which necessarily assumes all patients have the same clearance and bioavailability.

Pharmacodynamic models A linear pharmacodynamic model was used to describe the effect of placebo (with potency β_P) or active substance concentration (with potency β_A) on the parameters of the disease progress curve. An E_{max} model [8] was also examined to describe the effect of romazarit in terms of the parameters E_{maxA} and EC_{50A}. The placebo efficacy of romazarit (ε_A) treatment compared with placebo treatment was included in the model as a factor multiplying the placebo treatment potency parameter (β_P) when the patient was treated with romazarit.

Variability models The variability in the patient population of the PKPD model parameters was described using a proportional model. The variability parameter estimates can be considered as coefficients of variation of a log-normal distribution of the parameters in the population. The residual error was predicted by an additive error model.

Computation Parameter estimation and model building were performed using NONMEM [9] Version IV level 1.1 and NMTRAN version II level 1.1 using an HP9000/730 computer. Smoothed plots were generated using LOWESS [10] with a smoothing factor of 0.2.

Results

A total of 224 patients entered the study. Two hundred and twenty of these patients had observations suitable for use in the population PKPD analysis. Figure 1 shows the time course of the mean romazarit concentration at each visit. There was no

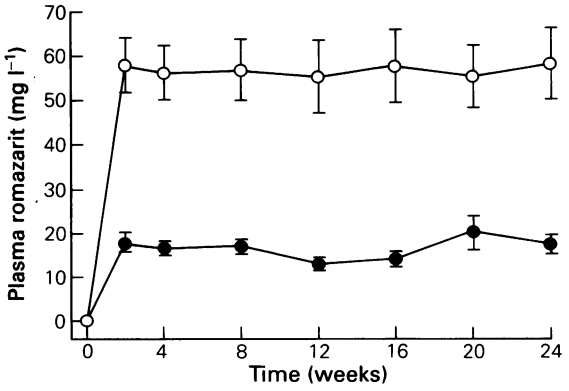

Figure 1 The time course of mean plasma romazarit concentration ± s.e. mean in all patients. ● 400 mg day^{-1}, ○ 900 mg day^{-1}.

Table 1 Baseline data of treatment groups

Characteristic	*Placebo*	*Romazarit* 400 mg day^{-1}	900 mg day^{-1}
Gender	21 M/ 52 F	19 M/ 56 F	19 M/ 51 F
Age (years) Mean	54	57	57
	22–70	33–78	21–73
Weight (kg) Mean	65.2	66.9	68.8
Range	40.5–97.0	45.0–98.0	35.0–97.1
Duration of RA (years) Mean	11.1	10.0	9.6
Range	0.67–36.9	0.57–30.4	0.54–29.1
RI (units) Mean	22.3	24.1	22.2
Range	8–48	6–56	4–44

Table 2 Analysis of variance for data from 164 patients who completed the first 12 weeks of the trial

Source of variation	*Degrees of freedom*	*Sum of squares*	F *ratio*	P
Baseline	1	5381.7	91.94	<0.01
Centre	10	1051.9	1.8	0.07
Treatment	2	279.6	2.39	0.10
Centre * Treatment	20	1689.5	1.44	0.11
Error	130	7609.2		
Total	163	16102.0		

Table 3 ANOVA estimates of difference between treatment effects from 164 patients who completed the first 12 weeks of the trial

Comparison	*Estimate*	*SE*	*95% CI*	P
Placebo *vs* 400 mg day^{-1}	2.94	1.61	−0.2–6.09	0.07
Placebo *vs* 900 mg day^{-1}	4.48	1.60	1.34–7.61	0.01
400 mg day^{-1} vs 900 mg day^{-1}	1.53	1.66	−1.72–4.78	>0.10

trend in the romazarit concentrations. This suggests that a pharmacokinetic steady state had been reached by the time of the first RI observation at 2 weeks. The time of collection of the blood samples varied over a 6 h interval after the dose.

The three patient groups were comparable at the start of treatment (Table 1). One hundred and sixty-four patients (75% of total) completed at least 12 weeks of either the placebo or romazarit treatment and were eligible for ANOVA. The ANOVA revealed a significant baseline effect and there was a suggestion of a centre effect but no significant overall treatment effect (Table 2). There was, however, a significant difference between the change in RI observed in the placebo group and the 900 mg day^{-1} group (Table 3).

The 220 patients examined using the population PKPD approach contributed 1336 paired observations of RI and romazarit concentration (73 placebo patients, 75 patients on 400 mg day^{-1} mean concentration = 13.0 ± 0.9 (s.e. mean) mg l^{-1}, and 72 patients on 900 mg day^{-1} mean concentration = 44.6 ± 4.3 mg l^{-1}).

The best PKPD model was assessed using the NONMEM objective function. A difference in the objective function of 3.84 for an additional parameter in the model can be interpreted as a significant change in the fit (P = 0.05). Table 4 shows the objective function associated with some of the models that were examined. The efficacy of the placebo response associated with romazarit was not significantly different from the response following placebo treatment. The influence of romazarit was described somewhat better by an effect on the slope of the disease progress model than an offset to the baseline (difference in objective function = 4.3). Measured romazarit concentration was a better predictor than dose (difference in objective function = 20.8). An E_{max} model was better than a linear model (difference in objective function = 26.3). The effect due to placebo was best described with a delay (difference in objective function = 20.1).

Estimates of the population parameter typical values and their variability are shown in Table 5 (E_{max} plus slope model). The slope model estimated the typical starting value for RI was 22.5. The placebo treatment 'dose' was characterized by an offset of −3.99 units. Because of the delay in appearance of the placebo

Table 4 PKPD models used to describe the time course of RI response to placebo and romazarit treatment

Model description	*Objective function difference*	*Conclusion*
Romazarit placebo efficacy <> 1	−1.8	Placebo efficacy = 1.2 not different from 1
Romazarit concentration effect on slope (E_{max})	0	Best model
Romazarit concentration effect on slope (linear)	26.3	E_{max} model better
Romazarit concentration effect on offset (E_{max})	4.3	Slope model better
Romazarit dose effect on slope (E_{max})	20.8	Concentration model better
Romazarit concentration effect on slope (E_{max}) no delay for placebo effect	20.1	Placebo delay needed

Table 5 Population parameters for the effect of romazarit (E_{max} model) on the slope of the disease progress curve using concentration as the measure of treatment intensity

Parameter	*Value*	*SE*	*CV %*
Disease			
S_0 (years)	22.5	0.58	582
α (RI/month)	−0.29	0.06	34.2
Placebo			
β_P (RI)	−3.99	1.77	306
$t_{1/2,eq,P}$ (weeks)	7.84	1.55	
$t_{1/2,el,P}$ (weeks)	7.85	1.55	
Romazarit			
$E_{max\ A,\ slope}$ (RI/week)	−0.35	0.14	342
$EC_{50A,\ slope}$ (mg l^{-1})	15.2	8.0	1640
$t_{1/2,eq,A}$ (weeks)	0.07	2.3	
Residual			
s.d.	4.9	1.6	
Objective	6391.2		

'concentration' at its effect site, defined by a half-time of 7.84 weeks and eventual decay of the placebo 'concentration' with a half-time of 7.85 weeks, the maximal placebo effect is predicted to have a magnitude of −1.5 RI units at 11.3 weeks after the start of treatment. The effect of romazarit was to add a downward component to the disease progress curve. At the average observed concentration of 13.0 mg l^{-1} in the 400 mg day^{-1} group the RI is predicted to decrease by 1.93 units in 3 months (independently of the placebo effect) and at 44.6 mg l^{-1} the RI would decrease by 3.13 units in 3 months. The onset of the romazarit effect was not importantly delayed. It could be described by a half-time of 11.8 h (Figures 2 and 3).

A confidence interval for the size of the romazarit E_{maxA} can be estimated by constructing a log likelihood profile. This was done by fixing E_{maxA} to values around the final estimate and re-estimating the other parameters. A curve was drawn through the log-likelihood values using cubic spline interpolation (Figure 4). The values of E_{maxA} associated with a 3.84 unit change in log-likelihood define the 95% confidence interval for E_{maxA} of −1.87 to −0.25 RI units/week.

Discussion

Romazarit appears to be effective as a disease modifying antirheumatic drug in patients with RA. Modification of the rheumatoid disease process has been measured using a symptomatic scale. The magnitude of the improvement in RI estimated in this study is dependent on the statistical technique used and the assumptions about how romazarit might work. The ANOVA method predicts a difference between placebo and romazarit of 4.5 units after 12

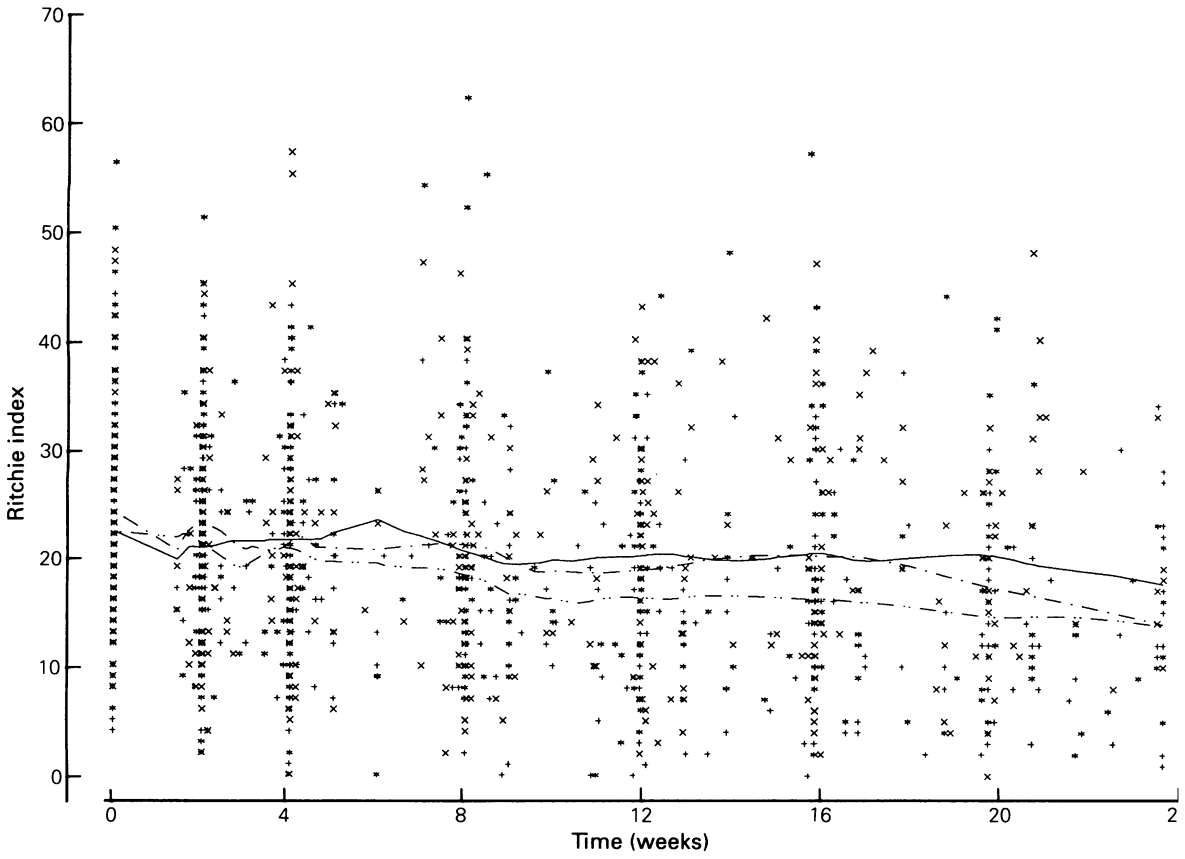

Figure 2 Scatterplot of all RI observations and smoothed curves derived from the observations. Separate curves are shown for the mean placebo (—), 400 mg day^{-1} (– • – •) and 900 mg day^{-1} (– •• –) data.

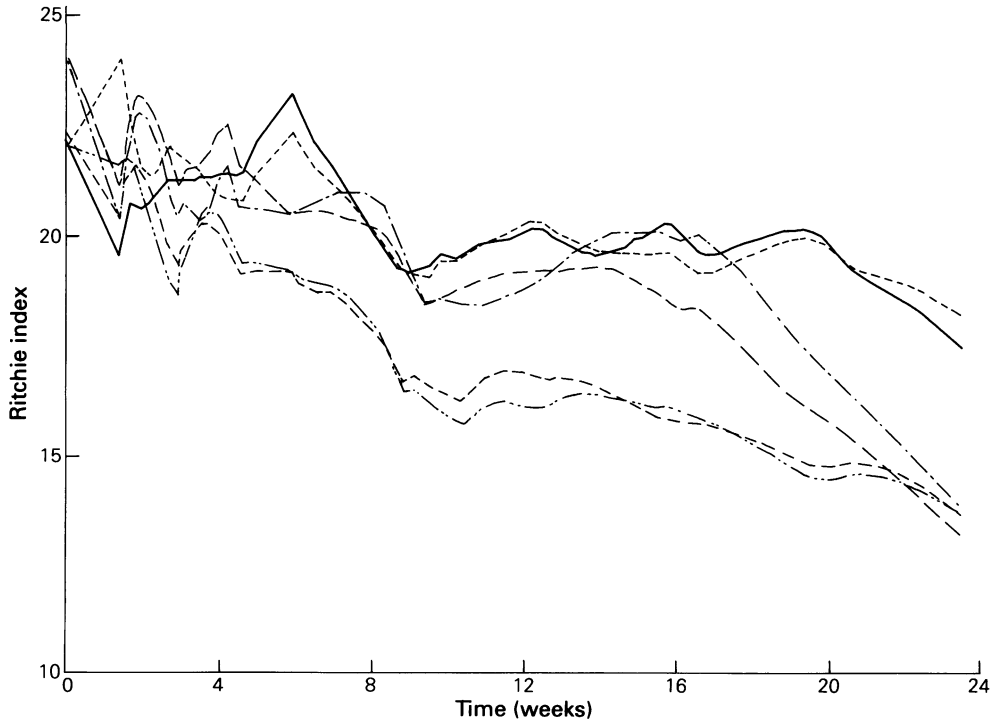

Figure 3 Similar to Figure 2 but with the observation points removed and the vertical scale expanded to show the time course of the observations and predictions more clearly. The upper line in the key for each treatment is a smooth through the observations while the lower line is a smooth through the corresponding predictions using the E_{max} slope model. – – – placebo, – • – 400 mg day^{-1}, – • • – 900 mg day^{-1}.

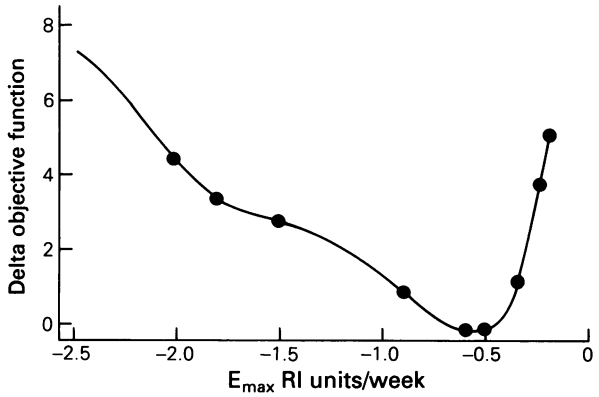

Figure 4 The log-likelihood profile for the romazarit maximum effect parameter $E_{maxA,slope}$.

weeks of treatment with 900 mg day^{-1} of romazarit. The PKPD model predicts a change in the rate of disease progression which would manifest as a difference of 3.13 RI units after 12 weeks with 900 mg day^{-1}.

The relationship between dose and effect (ANOVA) and concentration and effect (population model) suggests a diminishing effect at higher intensities of treatment. The ANOVA estimated a 52% extra response with a 125% addition to the dose (Table 3). The PKPD model predicts a 62% extra effect for the 143% extra concentration associated with the higher dose rate.

The demonstration of effectiveness of romazarit in this trial is supported by both the ANOVA and population PKPD approaches. This is as far as the ANOVA analysis could go in helping to understand the clinical pharmacology of romazarit. ANOVA is limited to answering questions of the type 'Is there a significant difference and how big is it?' If the analysis of this trial relied only on ANOVA then very little would have been learned about this new drug and almost nothing to guide future development and how to enhance its effectiveness. The population approach allows a more complete picture to be painted by using models of the disease and treatment effects as a framework for interpreting the observations.

A linear model for disease progress can be expected to be a reasonable approximation of any pattern provided the general rate of change is relatively slow in relation to the period covered by the model. The disease progress rate, α, was estimated to be negative implying an improvement in disease activity over the period of the trial. This could be a real phenomenon, e.g. due to seasonal variation in disease activity, or an artefact arising from model misspecification, e.g. due to confounding with the empirical placebo effect model.

The concept of a placebo dose at the start of treatment which gives rise to a gradually increasing then decreasing concentration of an active placebo substance is obviously a hypothetical construction in physical terms but it seems reasonable that the placebo response might follow a time course that rises and falls and this is what this placebo 'pharmacokinetic' model achieves. A similar model has been used to describe the placebo response to treatment with tacrine, a potential treatment for Alzheimer's disease, and has been able to distinguish cultural differences in its magnitude and time course [11, 12]. The estimate of the placebo efficacy, ε_A, for romazarit treatment was not different from 1 which means that the placebo response from placebo and romazarit treatments was indistinguishable.

The ANOVA results were based on a subset of patients enrolled in the trial and covers a more limited period. The population approach allowed all observations to be used and may therefore be more representative of the true response but the description of romazarit effect is dependent on assumptions that have to be made about the evolution of RI over time, the time course of the placebo response and the nature of the effect of romazarit. We believe that the assumptions we have made are reasonable in terms of a simple description of the behaviour that might occur.

The mechanism of action of romazarit is not well defined. Observations of the time course of its effect in the rat collagen arthritis model shows a delay of about a week before reaching a parallel dose related shift in the response curve [3]. The effective concentrations observed in this animal model proved to be valid predictors of the concentrations associated with a therapeutic effect in humans.

We do not feel confident in distinguishing between the mechanisms of effect of romazarit either on the slope or as an offset to the disease progress curve because of the short duration of the study. An effect on the slope appeared to be somewhat better than on offset.

We conclude that romazarit has significant effects on the Ritchie Index. The use of population PKPD models has challenged us to describe the important components of the observed response. By identifying and quantifying the influence of each of these we can not only draw conclusions about the effect of a potential medicine but also predict the time course of the treatment response over the period of the trial. Prediction of treatment responses over a period greater than 6 months is not feasible in the absence of a better understanding of the mechanism of action of romazarit. We have identified a clear hypothesis about the general nature of the effect, i.e. whether it is on the slope of the disease progress curve or an offset to it. This hypothesis could be tested by a longer study based upon the insights derived from the present analysis.

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References

1 Williams PEO, Muirhead GJ, Worth E, Zimmer R, Luecker P. Pharmacokinetics and tolerance of romazarit after oral administration of ascending single doses to healthy volunteers. *Eur J Drug Metab Pharmacokin* 1990; **15**: 317–322

2 Williams PEO, Bird HA, Minty S, Helliwell PS, Muirhead GJ, Bentley J, York A. A pharmacokinetic and tolerance study of romazarit in patients with rheumatoid arthritis. *Biopharm Drug Disp* 1992; **13**: 119–129.

3 Bloxham DP, Bradshaw D, Cashin CH, Dodge BB, Lewis EH, Westmacott D, Self CR. Biologic properties of romazarit (Ro 31-3948), a disease-modifying antirheumatic drug. *J Pharmac exp Pharmac* 1990; **252**: 1331–1340.

4 Ritchie DN, Boyles RA, McInnes, JM, Jasini MK, Dalakos TG, Grieveson P, Buchanan WW. Clinical studies with an articular index for the assessment of joint tenderness in patients with rheumatoid arthritis. *Quart J Med* 1968; **147**: 393–406.

5 Arnett FC, Edsworthy SM, Bloch DA, McShane DJ, Fries JF, Cooper NS, Healey LA, Kaplan SR, Liang MH, Luthra HS, Medsger TA, Mitchell DM, Neustadt DH, Pinals RS, Schalter JG, Sharp JT, Wilder RL, Hunder GG. The American Rheumatism Association 1987 revised criteria for the classification of rheumatoid arthritis. *Arthritis & Rheumatism* 1988; **31**: 315–324.

6 SAS/STAT, Version 6, SAS Institute Inc., Cary, NC, USA

7 Holford NHG. Parametric models for the time course of drug action—the population approach. In *New strategies in drug development and clinical evaluation: The population approach.* eds Rowland M, Aarons L, EUR 13775 Commission of the European Communities, Brussels, 1992.

8 Holford NHG, Sheiner LB. Understanding the dose-effect relationship: clinical application of pharmacokinetic-pharmacodynamic models. *Clin Pharmacokin* 1981; **6**: 429–453.

9 Beal SL, Sheiner LB. The NONMEM system. *Am Statist* 1980; **34**: 118–119.

10 Cleveland WS. Robust locally weighted regression and smoothing scatterplots. *J Am stat Ass* 1979; **74**: 829–836

11 Holford NHG, Peace K. Methodologic aspects of a population pharmacodynamic model for cognitive effects in Alzheimer patients treated with tacrine. *Proc Natl Acad Sci USA* 1992; **89**: 11466–11470.

12 Holford NHG, Peace K. Results and validation of a population pharmacodynamic model for cognitive effects in Alzheimer patients treated with tacrine. *Proc Natl Acad Sci USA* 1992; **89**: 11471–11475.

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