CONCISE REPORT

A randomised, double-blind, placebo-controlled trial of a recombinant version of human α -fetoprotein (MM-093) in patients with active rheumatoid arthritis

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Background: Rheumatoid arthritis (RA) tends to remit during pregnancy, with more patients achieving remission in the third trimester, coinciding with an increase in levels of α -fetoprotein (AFP). In vitro and animal studies have shown that AFP has immunomodulatory properties. MM-093 is a non-glycosylated, recombinant version of human AFP.

Objective: To assess the safety, tolerability and clinical effects of MM-093 during a 12-week, randomised, double-blind, placebo-controlled study.

Methods: 12 patients with RA, who had active disease and were on stable doses of methotrexate, received weekly subcutaneous injections of placebo or 21 mg of MM-093. Assessments were carried out at baseline and weekly thereafter.

Results: Baseline characteristics were similar in both groups. There was one dropout in the placebo group, due to flare of disease. Treatment with MM-093 was well tolerated. No serious adverse event was observed. By day 85, MM-093 produced a significant mean improvement from baseline in Disease Activity Score 28 (DAS28; 0.913 vs 0.008, $p=0.033$) and patient's global assessment (28.9% vs –36.3%, $p=0.02$) compared with placebo.

Conclusion: This is the first randomised, controlled trial of MM-093, a recombinant version of human AFP, in patients with RA. MM-093 was well tolerated. Evidence of efficacy was observed, suggesting that MM-093 may have therapeutic potential in RA.

Persistent synovitis and subsequent joint damage in rheumatoid arthritis (RA) leads to a significant increase in morbidity and mortality. Despite the undoubted success of tumour necrosis factor inhibitors, not all patients respond to treatment, and other patients have contraindications such as infections or significant congestive cardiac failure.

Although it is well accepted that RA tends to remit during pregnancy, the mechanisms have not been established.¹ Symptomatic relief becomes more pronounced as the pregnancy progresses, with more patients achieving remission by the third trimester.² This remission coincides with the increase in maternal and fetal levels of human α -fetoprotein (AFP), which has immunomodulatory properties^{3–5}; hence, it may be a significant contributory factor. AFP is produced at low levels throughout life; however, the fetus produces much higher levels of AFP. During pregnancy, AFP reaches maximal concentrations of 300–500 ng/ml during the third trimester. After delivery, levels of AFP fall to normal levels (5–10 ng/ml).⁵ In animal models of autoimmune diseases, studies have suggested that the immunomodulatory effect of AFP may be therapeutic.^{6–9}

MM-093 (Merrimack Pharmaceuticals, Cambridge, Massachusetts, USA) is a non-glycosylated version of human AFP produced by recombinant DNA technology in a transgenic goat expression system.¹⁰ We carried out a proof-of-concept trial to assess the tolerability, safety and therapeutic potential of AFP (MM-093) in RA.

PATIENTS AND METHODS

Patients

Patients (18–80 years) were eligible provided they met the American College of Rheumatology¹¹ criteria for RA and had active disease as defined by the presence of three of the following; ≥ 6 painful joints, ≥ 3 swollen joints, early morning stiffness for at least 45 min or a C reactive protein level >20 mg/l. Patients had to have RA for at least 6 months and have been taking ≥ 10 mg of methotrexate per week for at least 2 months. Corticosteroids (≤ 10 mg) and non-steroidal anti-inflammatory drugs were permitted provided the dose was stable. Corticosteroid injections and immunosuppressants were not allowed in the 30 days prior to screening. Treatment with biological or investigational drugs was not permitted within 90 days of screening. Patients in Steinbrocker functional class IV were excluded, as were patients with an allergy to goat milk.

Study drug

MM-093 is produced in the milk of transgenic goats. To date, MM-093 has demonstrated activity equivalent to cord blood-derived human AFP in animal models and in characterisation in vitro. Pharmacokinetic data from a phase I study of MM-093 suggested that an optimum target dose in the range of 2–50 mg was needed to attain the target serum concentration of 300–500 ng/ml, which is seen in the third trimester of pregnancy. MM-093 was demonstrated to have a half-life of approximately 5 days, suggesting a once-weekly subcutaneous injection.

Study protocol

The study protocol was approved by King's College Hospital Ethics Committee. Before starting the study, patients gave written informed consent, had a medical history taken and underwent a physical examination.

Treatment

Patients were assigned using the SAS V.6.12 computer software package. Four patients were randomly allocated placebo, and eight patients were allocated MM-093. MM-093 or placebo was injected subcutaneously every week for 12 weeks. MM-093 was supplied as a 1.1 ml solution of sterile phosphate-buffered saline, at a concentration of 21 mg/ml. The placebo was a 1.1 ml solution of sterile phosphate-buffered saline. Patients, assessors and investigators were blinded to the treatment allocation.

Abbreviations: AFP, α -fetoprotein; RA, rheumatoid arthritis

Table 1 Demographic and baseline clinical characteristics of study patients

	MM-093 (n = 8)	Placebo (n = 4)
Age (years)	56.7 (11.1)	61.2 (6.7)
Gender (female)	4	4
Disease duration (years)	6.7 (4.4)	13.0 (11.0)
Methotrexate dose (mg/week)	16.6 (4.2)	16.3 (7.8)
Swollen joints (28 count)	13.5 (3.9)	11.0 (3.4)
Tender joints (28 count)	14.8 (5.3)	21.0 (9.5)
Pain (100 mm VAS)	61.8 (26.0)	66.5 (33.6)
Patient's global assessment (100 mm VAS)	67.6 (22.9)	58.5 (31.2)
Physician's global assessment (100 mm VAS)	68.6 (12.5)	64.5 (15.8)
HAQ	1.9 (0.68)	2.4 (0.39)
CRP (mg/l)	32.8 (31.5)	20.4 (25.5)
ESR (mm/h)	40.3 (26.7)	32.8 (20.5)
Early morning stiffness (min)	97.5 (52.6)	112.5 (125.8)
DAS28	6.49 (0.43)	6.53 (0.62)
DAS28-CRP	6.14 (0.98)	6.14 (1.28)

CRP, C reactive protein; DAS, Disease Activity Score; ESR, erythrocyte sedimentation rate; HAQ, Health Assessment Questionnaire; VAS, visual analogue scale.

All values are mean (SD).

Efficacy

Disease activity assessments were based on the World Health Organization/International League of Associations For Rheumatology/Outcome Measures in Rheumatoid Arthritis Clinical Trials and the European League Against Rheumatism core dataset, and included tender and swollen joint count (28 joints), Health Assessment Questionnaire (HAQ),¹² patient and physician global assessment of disease (visual analogue scale 0–100 mm), pain score (visual analogue scale), erythrocyte sedimentation rate and C reactive protein. These were carried out at screening, baseline and every 1–2 weeks throughout the study. Follow-up assessments were completed at 1, 2 and 4 weeks following discontinuation of the drug.

Safety and pharmacokinetics

During the study period, adverse events, clinical laboratory tests (full blood count, biochemistry and urinalysis), vital signs and local injection site tolerance were recorded weekly. 12-lead ECG was carried out every 4 weeks.

Blood samples for the assay of serum MM-093 were taken each week, including follow-up visits. Blood samples for the assay of anti-MM-093 antibodies (IgG, IgM, IgE) were taken on days 1 and 106.

Statistical analysis

The study was analysed on an intention-to-treat basis. All statistical analyses were performed using the SPSS V.II. Simple descriptive

analyses were applied to the data in both groups and unpaired t tests were used to compare the improvement in clinical variables. For all analyses, a 5% significance level was used.

This was a phase IB study to investigate the safety and tolerability of MM-093, and was not powered to demonstrate efficacy.

RESULTS

Patient demographics

Twelve patients were enrolled in the study. Table 1 lists the baseline characteristics of the patients. The only significant difference between the groups was the sex distribution. The observed difference in disease duration between the groups was skewed due to a disease duration of 29 years for one patient in the placebo group, and was not statistically significant.

In all, 11 of the 12 patients completed the study; the only withdrawal was one patient from the placebo group, due to a flare of RA.

Efficacy

MM-093 produced a significant mean improvement from baseline in DAS28 (0.913 vs 0.008, $p = 0.033$) and patient's global assessment (28.9% vs –36.3%, $p = 0.02$) compared with placebo. Four of the eight patients in the MM-093 group had a moderate response in DAS28 score at day 85 according to the European League Against Rheumatism response criteria,¹³ compared with no response in the placebo group at day 85. For individual components of the core dataset, there seemed to be a trend towards improvement in the MM-093 group compared with the placebo group, but this difference was not statistically significant (table 2).

Pharmacokinetics

Serum concentrations of MM-093 in patients who received MM-093 seemed to indicate attainment of steady state by 21 days after the start of treatment, with a concentration range of approximately 1200–1500 ng/ml.

At the time of analysis, a dosing error was noted at day 64 for one patient receiving placebo, and at day 57 for one patient receiving MM-093. Serum concentration levels of MM-093 increased sharply for the patient in the placebo group on day 71, suggesting that this patient was given MM-093 on day 64. In addition, serum concentrations fell in the patient in the MM-093 group on day 64, suggesting that this patient had received placebo on day 57. Despite these dosing errors, the results were analysed using the original data, as they would only bias the results towards the negative.

Safety and tolerability

MM-093 was well tolerated. A total of 44 treatment-emergent adverse events were reported by 11 of the 12 patients. The

Table 2 Mean improvement from baseline at study end in core dataset and disease activity measures (absolute values are given in brackets)

Component	MM-093	Placebo
Reduction in DAS28	0.913	0.008*
Patient's global assessment (100 mm VAS)	28.9% (20)	–36.3%* (–9.3)
Pain (100 mm VAS)	25.3% (17.8)	–9.8% (3.5)
Swollen joint count (28 joint count)	37.4% (5)	–2.5% (0)
Tender joint count (28 joint count)	32.4% (4.4)	20.4% (3)
Physician's global assessment (100 mm VAS)	39.8% (26.38)	30.0% (18.5)
HAQ	–3.8% (–0.5)	23% (0.5)
CRP (mg/l)	4.8% (1.8)	–1.2% (–9.1)
ESR (mm/h)	–18.0% (–1.8)	–22.9% (–10.5)

CRP, C reactive protein; DAS, Disease Activity Score; ESR, erythrocyte sedimentation rate; HAQ, Health Assessment Questionnaire; VAS, visual analogue scale.

*Statistically significant ($p < 0.05$).

incidence of adverse events was similar in the MM-093 and placebo patients (88% and 100%, respectively). The most common adverse events were headaches, upper respiratory tract infections, musculoskeletal pain and injection site reactions. Only headaches and injection site reactions were considered to be related to MM-093. Injection site reactions, which occurred in two out of eight patients, were mild and self-limiting. There were no serious adverse events or deaths during the study.

There were no apparent treatment-related trends in any safety parameters. One patient in the MM-093 group had transient increased levels of liver transaminases at day 29, which returned to normal by day 43 and remained normal, despite continued administration of MM-093. No antibodies to MM-093 were detected.

DISCUSSION

This was the first randomised, controlled trial of MM-093, a recombinant version of human AFP, in RA. Although the sample size of this trial was small and the study was not powered to demonstrate efficacy, this proof-of-concept study suggests that MM-093 might have therapeutic potential in RA.

AFP suppresses autologous lymphocyte proliferation. In experimental arthritis and autoimmune thyroiditis, amelioration of disease was observed in transgenic AFP-producing mice.⁶⁻⁹ This was associated with significantly reduced numbers of CD4 and CD8 splenocytes, total thymocytes and CD4 thymocytes, suggesting that AFP modulates T cell development and immune responses.

Although tumour necrosis factor inhibitors are a major advance in the treatment of RA, they are immunosuppressive and increase the risk of infection, and are therefore contraindicated in patients with concurrent infection or those at risk of serious infections. Consequently, new treatments, which are efficacious but less immunosuppressive, are needed. In this study, administration of MM-093 was well tolerated; only headache and injection site reactions were considered likely to be treatment-related. Mechanistically, it seems unlikely that human AFP would lead to severe immunosuppression, and hence MM-093 may fill an important gap in the current treatment armamentarium in RA.

In conclusion, this pilot study of MM-093 in patients with RA demonstrated that MM-093 was well tolerated. Evidence of efficacy was observed in the study and MM-093 may have therapeutic potential in RA. This will require confirmation in larger randomised trials, and a phase II trial is currently underway in the USA.

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EHSC, MM, EJS and JM were responsible for the protocol and design of the study. LCP recruited and assessed all the patients in the study. JM and LCP carried out the statistical analysis of the study under the supervision of EHSC.

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