The Discoid Lupus Research Project (protocol vs 4, dated 04.09.2007)

Eudract No 2006-007056-18, sponsor no 3403, REC ref 05/Q0904/75

Sponsor: The Newcastle upon Tyne Hospitals NHS Foundation Trust

End of trial study report 08/04/16

Below is a detailed synopsis summary of the above study

Trial Information: Background

Discoid Lupus Erythematosus (DLE) is an inflammatory skin disease which cause scarring and redness on visible sun-exposed parts of the body. It is encountered by dermatologists and diagnosed clinically and with a skin biopsy.

The recommended systemic therapy of choice for discoid lupus erythematosus (DLE) is the 4- aminoquinolone antimalarial hydroxychloroquine. At the time of writing our study protocol in 2007, there was limited published information on the likelihood of clinical response and, in particular, what factors influenced outcome.

Trial information: Aim of study

The overarching aim of the study was to determine response to hydroxychloroquine therapy for DLE. The central objective was to investigate the relevance of CYP 450 polymorphisms to hydroxychloroquine efficacy through a 'retrospective study' (described below).

Trial information: Summary of method

We performed a retrospective casenote study on a large cohort of patients available in 9 centres in the UK, including clinics in Newcastle and Leeds, to gain preliminary evidence on the response rate to hydroxychloroquine and the nature of the CYP genotype-drug response relationship. In these patients we classified response to hydroxychloroquine retrospectively by review of case-notes, categorising efficacy in the manner described by other smaller prior retrospective studies of DLE.

Our study protocol described a ‘retrospective study’ to gain preliminary evidence on response to hydroxychloroquine and any relationship to CYP genotype.

We conducted a multicentre retrospective casenote and pharmacogenetic study in patients with DLE who had been treated with hydroxychloroquine by their responsible dermatologist as part of their standard care. It is important to note that these patients were not commenced on hydroxychloroquine as part of the study – instead we were reviewing casenotes of patients who had already been on hydroxychloroquine as part of their standard care by their responsible dermatologist. These patients had either had absent or adequately controlled disease that was controlled on long-term hydroxychloroquine or had failed to respond to it and had moved on to other forms of systemic therapies. In all patients we intended to classify response by review of casenotes, categorizing efficacy in the manner described by other smaller retrospective studies of DLE: Patients were classified as responders by 6 months if there had been an improvement in their disease and no other oral therapies were required to achieve that improvement. Patients were deemed to be non-responders by 6 months if their disease did not change or had worsened and/or other oral therapies had been needed as a consequence. This definition of response is similar to that used in other retrospective and prospective studies assessing response to antimalarials in the context of cigarette smoking

The aim was to make a retrospective designation of clinical response to hydroxychloroquine within the first 6 months of its use and also to investigate the effects of metabolizing cytochrome P450 (CYP) polymorphisms on prior clinical outcome.

Power calculation

The sample size needed for the study was based on the number of patients needed to investigate the influence of CYP2C8 and CYP2D6 polymorphisms on the response of DLE to hydroxychloroquine. Firstly, an indication was needed as to what level of influence (or odds) on response, provided by the polymorphisms of interest, was worthy of investigation. After consultation amongst the study team, it was agreed that an odds ratio of between 2.0 to 2.5, afforded by the polymorphisms, between non-responders and responders, would be of practical relevance in a clinical setting.

A preliminary survey of all participating study collaborators indicated that collectively, approximately 350 patients with DLE and exposure to hydroxychloroquine were under follow-up. Approximately 60-70% of these patients were estimated to be responders to therapy. Taking into account the allele

frequencies of interest and the estimated ratio of non-responders to responders, power calculations indicated that with 200 patients overall, the study would have 80% power to detect an odds ratio of between 2.25 to 2.5 between non-responders and responders with respect to CYP2D6 and CYP2C8 polymorphisms (at 1% significance level).

This meant that if the true effect size of the polymorphisms was in the region of 2.25 to 2.5, the sample size of 200 patients ought to be able to detect it. Indeed, in the event that the true effect size was even higher than this, it would still be detectable. However, if the true effect size was lower than this, the study would not have enough power to show a significant association. The sample size was calculated according to the potential effects of CYP2C8 and CYP2D6 variants on disease response; these 2 enzymes are the principal enzymes metabolising hydroxychloroquine that exhibit genetic variation.

Actual start date of recruitment: 21/05/2007

Last patient recruited: 17/05/2010

Subject disposition: Screening details

At the time of undertaking the retrospective study and writing the protocol, there were no nationally or internationally agreed set criteria for the diagnosis of DLE, in clinical practice a combination of histological and clinical features are usually employed. For the purposes of the study, these were formalised by the study team investigators in order to ensure a high specificity for DLE. In order to be included into the study, patients needed to have:

(1) Clinical features of scarring with erythematous and/or scaly plaques.

(2) A lesional skin biopsy confirming a clinical suspicion of DLE, showing a periadnexal and/or perivascular chronic inflammatory infiltrate and/or epidermal 'lichenoid' interface change.

(3) Prior exposure to hydroxychloroquine therapy (this is a licensed drug for cutaneous lupus, with a trade name of 'plaquenil', which is administered as oral tablet form)

Patients were identified through a combination of diagnostic, histopathology and pharmacy databases. The dermatology records of all the identified potential participants were carefully reviewed to ensure patients were eligible for inclusion into the study.

Subject disposition: Recruitment process

A total of 315 patients were identified and screened for enrollment; 40 patients did not satisfy the pre-defined diagnostic criteria for DLE (for instance, there was no biopsy report available) and/or had not received hydroxychloroquine therapy and 2 patients were deceased. The 273 remaining eligible patients were invited to take part in the study, either during their routine clinic consultation and/or by a letter. 68 patients did not respond to the invitation letter and 5 patients did not wish to take part.

Protection of patients

Patients in this retrospective study only required a single visit for interview and venepuncture for CYP genotyping. As per the retrospective study nature therefore, long-term follow-up was not needed. This was a retrospective study which involved review of casenotes of patients who had been on hydroxychloroquine. Patients were also interviewed to obtain relevant demographic data (e.g. smoking status). Blood was taken for genotyping. Measures were taken to minimise harm from blood testing. In the retrospective study, participants would be undergoing blood tests. We indicated in the patient information leaflet and in the consent forms what these procedures entailed and the potential side effects. We did not come across or envisage issues regarding the interview process which was done by a single investigator for all patients. Patients were consented according to GCP. Hydroxychloroquine was being regarded as an investigational medicinal product by the MHRA in this study because we were investigating the pharmacogenetics of the drug with respect to prior recorded response in DLE. Hydroxychloroquine had been previously already prescribed in the usual manner by each participant's responsible Dermatologist as part of their standard clinical care in accordance with the terms of its licensed authorisation.

The assignment of any patient to take hydroxychloroquine had been decided as the clinician's particular therapeutic strategy for that patient and was not decided in advance from the clinical trial protocol. Therefore, the decision to prescribe hydroxychloroquine (in the best interests of the patient skin care) was clearly separated from the decision to include the patient in the study.

No placebo or randomisation was being used in this trial.

Data was reported on case report form (link-anonymised) and be kept securely by the sponsor

Baseline characteristics

A total of 200 patients with DLE were recruited (F:M = 160 : 40) and all were over the age of 18. The median age at initial diagnosis of DLE (according to the casenotes), was 40 years (range 16-81) and patients had received a median hospital follow-up of 8 years (range 0·5-37).

A total of 92 patients (46%) had DLE affecting one site, 61 patients (30.5%) had DLE in 2 sites, 11 (5.5%) in 3 sites, 19 (9.5%) in 4 sites, 15 (7.5%) in 5 sites and 2 (1%) in 6 sites. Using the pre-defined classification for disease extent, this meant that 164 patients (82%) had localised disease confined to 3 or less body sites; the remainder had disseminated disease affecting 4 or more body sites.

Autoantibody status was negative in 115 patients (57.5%). In the cohort, 41 patients (20.5%) were ANA positive; 29 patients (14.5%) had a borderline positive/positive antibody to ENA (either anti-Ro/SS-A and/or anti-La/SS-B antibody); 15 patients (7.5%) had a borderline positive/positive antibody to dsDNA. In 25 patients (12.5%), there was no evidence of autoantibody status being tested. Overall, 60 patients (30%) had either a positive ANA, and/or a positive/borderline positive antibody to ENA and/or dsDNA .

Of the 200 patients recruited, 11 patients (5.5%) had SLE; the major manifestations being: 5 glomerulonephritis including one requiring renal transplant, 8 arthritis, 2 mouth ulcers and 1 with pulmonary fibrosis. DLE predated SLE in 3 cases (after a median interval of 4 yrs, range 2-20 yrs); occurred after the diagnosis of SLE in 5 cases (after a median interval of 6.5 yrs, range 2-20 yrs) and in 3 cases the temporal relationship was equivocal. The mean weekly alcohol consumption was 6 units (range 0-48).

At the time of recruitment only 40 patients (20%) had never smoked with the remainder being either ex-smokers (63 patients; 31.5%) or current regular smokers (97patients; 48.5%). For the purposes of the study, it was deemed important to know whether patients had smoked or not during the initial 6 months of hydroxychloroquine use when response was being assigned: 125 patients (62.5%) were smokers during that period and the remaining 75 (37.5%) were not.

Hydroxychloroquine had been commenced after a median disease duration of 6.5 months (range 1 – 276); at a 200mg twice-daily dose in 144 patients (72%), at a 200mg once-daily dose in 49 patients (24.5%) and at a 200mg thrice-daily dose in 7 patients - providing a cohort mean starting dose of 5.3mg/kg body weight (range: 1.6 - 14.5 mg/kg).

End points: Reporting of overall response rate and statistical analysis

We reported the overall response rate for the 200 patients recruited for the retrospective study. This was a countable measure based on the retrospective casenote review.

We found that although the majority (60%, n=120) of patients responded to hydroxychloroquine within the first 6 months of use, 39% (n=78) failed to respond and/or withdrew from therapy within that timeframe. In 1% (n=2), response was equivocal.

Univariate logistic regression analysis was performed (using STATA) to determine if the CYP genotypes were significantly associated with response. For each of the CYP genes of interest (2C8 and 2D6), response of wild type (*1/*1) individuals was compared with heterozygotes and homozygous mutants combined using chi-square tests. Multivariate logistic regression analysis was undertaken putting all significant baseline variables from the initial univariate analysis into a model and retaining them if they remained significantly associated with response (after adjustment for other factors) using a forward stepwise approach.

CYP genotype did not have any significant influence on response to hydroxychloroquine. The Odds Ratio was 1.55 (95% CI 0.82-2.96) for response associated with CYP2C8 variants (p value 0.18). The Odds Ratio was 1.20 (95% CI 0.67 -2.18) for response associated with CYP2D6 variants (p value 0.54).

Multivariate analysis indicated that disseminated disease (odds ratio (OR): 0.21; 95% confidence interval (CI): 0.08-0.52; P<0.001) and concomitant systemic lupus erythematosus (SLE; OR: 0.06; 95% CI: 0.01-0.49; P = 0.009) were significantly associated with lack of response to hydroxychloroquine. These findings suggest that baseline lupus severity and SLE could be predictors of response to hydroxychloroquine.

Separate pharmacokinetic study in 6 patients

In our study protocol, we also described a pharmacokinetic substudy to assess blood levels of hydroxychloroquine in patients who were already on hydroxychloroquine to provide information on time-to peak concentrations and variability in concentrations between patients. Six patients were recruited with cutaneous lupus erythematosus who had each been on hydroxychloroquine 200mg twice daily for at least 6 months, so that they were at steady-state. Each patient was fasted overnight and had standardised meals and dosing schedule. Whole blood was sampled at 7 timepoints over 24 hours. Whole blood hydroxychloroquine levels were measured with High Performance Liquid Chromatography using gradient elution, fluoremetric detection and chloroquine as an internal standard. The assay had a mean inter and intra-day coefficient of variation of 10% and 5% respectively and a limit of detection of 5ng/ml. Hydroxychloroquine levels appeared to follow a biphasic pattern over the sampling period. Maximum levels were noted after a median of 4 hours (range 2-6) after ingestion. Median intra-patient variation between trough and peak levels (Cmax) was 27% (range 8-150%). This study demonstrates that whole blood Hydroxychloroquine levels exhibit marked variation within an individual over a 12 hour period. This degree of variation might represent a potential confounding factor in the previous studies suggesting a link between clinical response and Hydroxychloroquine blood concentrations. We suggest that future studies investigating this relationship should use the trough level as a single standardised timepoint for sampling patients.

Adverse events

The retrospective casenote study was solely based on patients who had received the licensed drug hydroxychloroquine as part of their standard care by their dermatologist previously. As part of this assessment we reviewed the case notes of response and any adverse events that were noted in the casenotes were recorded. As such this was a non-systematic retrospective assessment and was based on data entry in case notes only, as per the retrospective design of the study.

There were no SAEs or SUSARs. There were no deaths.

There were 48 non-serious adverse events (all recorded from the retrospective study). There were 17 individuals with gastrointestinal (GI) events (nausea, diarrhoea or abdominal pain), 13 with rashes, 5 with transient visual disturbances (including blurred vision or 'floaters' but not retinopathy), 5 with headaches, 4 with malaise, 2 with dizziness. In 2 patients, an asymptomatic transient transaminitis was noted on monitoring blood tests.

We would point out that all of these events are consistent with hydroxychloroquine's known side effects, as reported in its SmPC.

Other notes re interruption to study

Recruitment for the retrospective part of the discoid lupus research project commenced in 21st May 2007 and then was halted on 13th August 2007, after it became clear that an ethical re-submission on the correct CTIMP form was required. Upto this point 15 patients had been recruited towards the retrospective study. Recruitment since recommenced following approval of this CTIMP study on 25/10/2007.

Limitations to study and caveats applicable to summary of results

Although there are limitations in undertaking a retrospective assessment of response, the study design attempted to minimise the potential for bias with the following measures: a single investigator recruited and interviewed all patients, reviewed their case notes according to a pre-defined case report proforma and importantly a single investigator made the judgement on response. Despite this, some data obtained on clinical characteristics of patients (i.e. disease extent, body sites affected, disease duration) was clearly reliant on case note entries and clinic letters by dermatologists. The effect of missing or incorrectly interpreted data cannot be ignored and is a problem with retrospective casenote studies.

Recruitment sites encompassed 9 centres including both teaching and district general hospitals. This breadth of recruitment sites was important for both completing the study and limiting the effect of selection bias. One centre received tertiary referrals for management of DLE and 17 of the 40 patients recruited from that particular site were tertiary referrals. Even after excluding these 17 patients from the overall analysis, 35% of the remaining 183 patients still failed to respond adequately to hydroxychloroquine and furthermore, disseminated disease and concomitant SLE remained significantly associated with poor outcome (i.e. non-response).

Publications and dissemination of results

We have published our study findings and disseminated our results nationally and internationally at relevant scientific meetings. Our findings have also been presented regionally to departments in the North East and Scotland. Co-investigators have been informed of all the findings. The consultants can use this information (which they have co-authored; JID paper 2011) to help guide treatment in their patients.

Relevant publications

Wahie S, Meggitt SJ. Long-term outcomes to hydroxychloroquine in patients with Discoid Lupus Erythematosus. Br J Dermatol. 2013; 169: 653-9.

Wahie S, Daly AK, Cordell HJ, Goodfield MJ, Jones SK, Lovell CR, Carmichael AJ, Carr MM, Drummond A, Natarajan S, Smith CH, Reynolds NJ, Meggitt SJ. Clinical and pharmacogenetic influences on response to hydroxychloroquine in discoid lupus erythematosus: a retrospective cohort study. J Invest Dermatol. 2011; 131: 1981-6.

Wahie S, McColl E, Reynolds NJ, Meggitt SJ. Measuring disease activity and damage in cutaneous lupus erythematosus.Br J Dermatol. 2011; 164: 221-2.

Wahie S, Reynolds NJ, Meggitt SJ. Lack of association between cigarette smoking and response to hydroxychloroquine in 200 patients with Discoid Lupus Erythematosus. J Invest Dermatol 2010; 130 (Suppl 2): 58.

Wahie S Goodfield MJ, Carmichael AJ, Carr MM, Drummond A, Jones SK, Lovell CR, Reynolds NJ, Meggitt SJ. Response to hydroxychloroquine in patients with

discoid lupus erythematosus Br J Dermatol 2010; 163 (Suppl 1): 9.

Wahie S, Reynolds NJ, Meggitt SJ. Assessment of disease activity and damage in discoid lupus erythematosus. J Invest Dermatol 2009; 129 (suppl 2): 18.

Wahie S, McColl E, Reynolds NJ, Meggitt SJ. Measuring disease activity and damage in discoid lupus erythematosus. Br J Dermatol 2010; 162: 1030-7.

Relevant National and International Presentations

Steady-state pharmacokinetics of hydroxychloroquine in patients with cutaneous lupus erythematosus.

Presented at: British Association of Dermatologists 87th Annual Meeting, London

7/7/11

Response to hydroxychloroquine in patients with discoid lupus erythematosus

Awarded best registrar oral presentation prize

Presented at: British Association of Dermatologists 86th Annual Meeting, Manchester

07/07/10

Lack of association between cigarette smoking and response to hydroxychloroquine in 200 patients with Discoid Lupus Erythematosus

Presented at: European Society for Dermatological Research, Barcelona 7/09/10

Measuring disease severity and damage in discoid lupus erythematosus

Presented at:

European Society for Dermatological Research, Budapest 10/09/09

British Association of Dermatologists 89th Annual Meeting, Glasgow 07/07/09