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Original article

Treatment with cyclophosphamide i.v. pulse therapy is an option for effective treatment of skin fibrosis in patients with early systemic sclerosis

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Abstract

Objectives. SSc is a autoimmune disease characterized by fibrosis of the skin and internal organs. There is a lack of evidence for the efficacy of i.v. CYC pulse therapy on skin thickening. We aimed to examine the response of i.v. CYC pulse therapy on skin thickening in our cohort of SSc patients and analysed factors that predict this response.

Methods. We retrospectively evaluated the data for 143 patients with SSc from baseline to 12, 24 and 36 months. All patients were treated with at least 6 i.v. CYC pulses (750 mg/m^2/month). We applied the modified Rodnan Skin Score (mRSS) to assess skin thickening. A clinically relevant response was defined as a decrease in mRSS of 5 points and 25% from baseline. Different baseline variables for predicting response on month 12 were tested in logistic regression analyses.

Results. Baseline characteristics of the patients with dcSSc and lcSSc were collected. Forty-three percent ($n = 42$) of dcSSc patients had a clinically relevant response on month 12. Non-responding on month 6 predicts non-response on month 12 (odds ratio 37.1; 95% CI 4.5, 306.4).

Conclusion. We concluded that i.v. CYC pulse therapy should be considered as an effective treatment option for skin thickening in dcSSc patients, because 43% of this group of patients were found to have a clinically relevant response. Of the dcSSC patients who did not respond by month 6, only 29% had a response by month 12. This finding can help the physician and patient in shared decision making about whether or not to continue therapy.

Key words: systemic sclerosis, cyclophosphamide, skin fibrosis

CLINICAL SCIENCE

Rheumatology key messages

- Cyclophosphamide i.v. pulse therapy is an effective therapeutic option for skin fibrosis in systemic sclerosis.
- Non-response after 6 months of cyclophosphamide i.v. pulse therapy in SSc predicts non-response at month 12.
- Evaluating response after 6 months of cyclophosphamide i.v. in SSc helps in therapeutic decision making.

Introduction

SSc is a generalized autoimmune disease characterized by inflammation, vasculopathy and fibrosis clinically visible as skin thickening and in many cases internal organ involvement. Based on the extent of skin thickening, SSc is divided in two subtypes, lcSSc and dcSSc [1]. dcSSc and lcSSc are both associated with organ involvement, although dcSSc has a worse prognosis due to early and more severe internal organ involvement. Progressive skin thickening in dcSSc is a surrogate marker for disease activity, severity and mortality. In addition, improvement of skin thickening is associated with better prognosis [2, 3]. The natural course of skin thickening is characterized by a first stage of progressive skin thickening that peaks 1–3 years after disease onset, with spontaneous skin softening in later stages [4–6]. To assess skin thickening, the modified Rodnan skin score (mRSS) is a commonly used, validated tool. The mRSS is assessed clinically at 17 body sites on a 0–3 scale with a maximum of 51 and measures the extent of skin thickening [2, 4, 7–9].

There are several therapeutic options for patients with progressive skin thickening in dcSSc. MTX is efficient in

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reducing skin thickening [10]. This therapy is started if there are no signs of interstitial lung disease (ILD). Patients with ILD or progressive skin thickening not responsive to MTX are usually treated with either CYC, or MMF [11]. The effect of i.v. CYC pulse therapy on ILD in our cohort has previously been reported [12]. CYC is a cytotoxic immunosuppressive agent originally indicated for treatment of (haematological) malignancy. The main mechanisms of action are cell death, lymphocyte modulation and impairment of inflammatory responses [13].

Although CYC is a widely used therapy, remarkably little is known about the effect on skin thickening. Recently, a post hoc analysis of the scleroderma lung studies I and II has shown an reduction in skin thickening of $\geqslant$5 points in mRSS in patients with dcSSc after oral CYC (40%) or MMF (38%) compared with placebo after 24 months of follow-up [14]. Likewise, two other smaller studies showed a positive reducing effect of oral CYC on skin thickening [15, 16]. However, evidence of i.v. CYC pulse therapy on skin thickening is still lacking. The result of i.v. CYC could be different from that of oral CYC, because the dose per administration is higher in the i.v. CYC pulse therapy. In i.v. CYC pulse therapy, the cumulative dose is lower than in oral CYC and is therefore better tolerated.

The aim of the study is to examine the response of i.v. CYC pulse therapy on skin thickening and to identify factors associated with response. The latter is relevant in deciding whether or not to continue i.v. CYC pulse therapy or to switch to another therapy.

Methods

Design

This retrospective, longitudinal, observational study was performed in the Department of Rheumatic Diseases of the Radboud University Medical Centre in Nijmegen in the Netherlands. The Nijmegen SSc database consists of 790 patients diagnosed with SSc [17].

The standard first treatment option for progressive skin thickening in dcSSc and/or SSc-related interstitial lung disease (SSc-ILD) until 2017 in our centre was i.v. CYC monthly pulse therapy. All patients who received i.v. CYC pulse therapy for progressive dcSSc and/or SSc-ILD from January 2004 until August 2016 were considered for inclusion. Due to the observational nature of this cohort, no ethical review was needed according to Dutch law and regulations.

Patients

Patients were classified as dcSSc or lcSSc according to the Leroy and Medsger criteria [1]. Disease duration was defined as the date of first manifestation of non-RP. Data for patients with progressive dcSSc and/or patients with SSc-ILD who received i.v. CYC 750 mg/m^2 at least six times with at least two measurements of mRSS was used. Follow-up data were collected until month 36 after starting CYC. In total, 184 patients were treated with i.v. CYC during the observation period and considered for analysis. Twenty-eight were excluded because there was $\leqslant$1 measurement of mRSS, 9 patients were excluded because they received $<$6 i.v. CYC pulses, and 4 patients were excluded because they only had ILD and no skin thickening. Eventually, 143 patients were included in the analysis.

Before 2013, the main follow-up treatment after i.v. CYC pulse therapy consisted of AZA, MTX or no treatment. After 2013, this consisted of MMF only. If patients had progressive disease or i.v. CYC pulse therapy failed, patients were screened for treatment with autologous stem cell transplantation (ASCT).

Assessments

Baseline demographic and clinical data were collected through chart review. Baseline data included age, gender, disease subtype, disease duration (in months), therapy indication and antibody profile. Data on mRSS was collected at baseline, 6, 12, 24 and 36 months. Data on follow-up treatment and number of completed i.v. CYC pulses were collected from month 6 onwards.

Patients are routinely seen in our outpatient clinic every 3 months, and mRSS is assessed as part of standard care by trained rheumatologists. Because of the retrospective nature of this study, mRSSs within 2 months of the nominal time point of measurement were used.

Outcomes

The primary outcome of this study was the percentage of responders after 12 months of i.v. CYC pulse therapy in the dcSSc group. A clinically relevant response was defined as an improvement of at least 5 points and 25% on the mRSS from baseline compared with month 12 [18, 19]. Because lcSSc patients are not able to fulfil the response criteria, we only used these response criteria on the dcSSc group. Patients who died during follow-up or who underwent ASCT after 6 i.v. CYC pulses were considered as non-responders in the analyses at time points after death or ASCT. Secondary outcomes were: response rates at 6, 24 and 36 months, and baseline variables that could predict response in the dcSSc group. Other secondary outcomes were, mean decrease in mRSS after 12 months and the course of mRSS during follow up in the dcSSc and lcSSc groups.

Statistical analysis

Descriptive statistics were provided as mean and S.D., median and interquartile range, or numbers with percentages (%) where appropriate. A prediction model for response at 12 months was created using logistic regression considering the baseline variables age, gender, disease duration, antibody profile, and response at 6 months as possible predictors. Predictors were included in the model when univariately associated with response at 12 months with a *P*-value smaller than a deliberately liberal threshold of 0.2.

Missing mRSS scores were handled using multiple imputation by chained equation to create 20 imputed datasets, which were combined using Rubin's rules, and imputed mRSSs were used for all analyses [20]. Imputations of the mRSS were made using truncated

regression based on the variables listed in Table 1. Analyses were performed using STATA 13.1. Data for patients who died or underwent ASCT after 12 months of follow-up was not imputed. These patients were classified as non-responders from the moment of death or after ASCT had taken place for the response criteria, and were excluded from the continuous mRSS analyses.

Table 1 Baseline characteristics

	lcSSc ($n = 44$)	dcSSc ($n = 99$)
Age, mean (S.D.)	58 (10)	54 (13)
Female, *n* (%)	28 (64%)	40 (40%)
Baseline mRSS, median (IQR)	3 (1–7)	17 (12–22)
Lung indication, *n* (%)	43 (98%)	51 (52%)
Disease duration in months, at start i.v. CYC median (IQR)	3.8 (0.9–12.6)	3.8 (1.5–13.5)
Antibodies, *n* (%)		
ANA positive	43 (98%)	96 (97%)
Anti-SCL70	16 (36%)	57 (58%)
Anti-centromere	6 (14%)	0 (0%)
Other	21 (48%)	39 (39%)

Disease duration from the first non-RP. Other antibodies: anti-RNP, anti-fibrilarine, anti-SSA, anti-RNA polymerase III or anti-PM-Scl70. mRSS: modified Rodnan skin score; IQR: interquartile range.

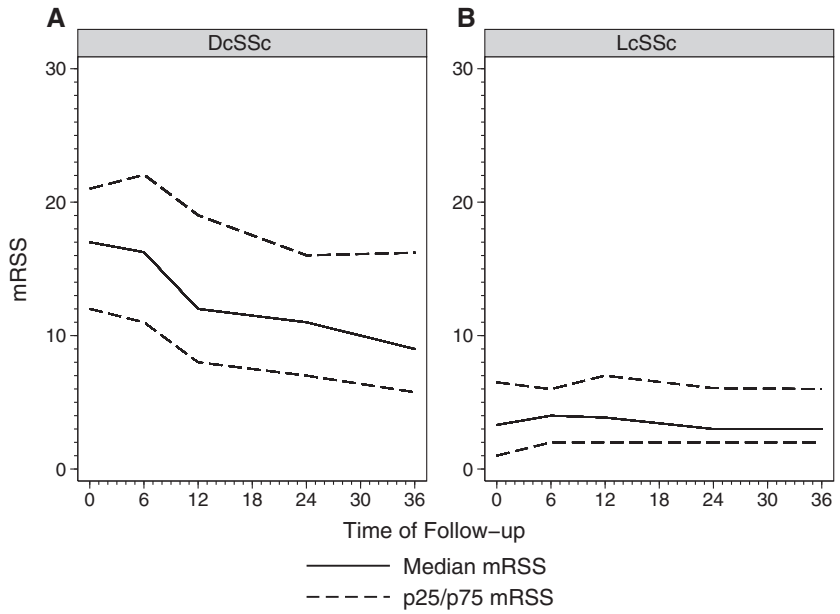
Results

Data for 143 patients were analysed for the primary end point. Table 1 summarizes baseline characteristics of the eligible patients. The mean mRSS in the dcSSc group was 17 [12–22] and the mean mRSS in the lcSSc group was 3 [1–7]. The indication for i.v. CYC pulse therapy was ILD in most patients in the lcSSc group, whereas in the dcSSc group about half of the patients had ILD as a treatment indication (Table 1). The dcSSc group consisted of more males. Both patient groups had short disease durations. Almost all patients were ANA positive, which is comparable with other SSc cohorts (Table 1).

Course over time

The mean change in mRSS in the dcSSc group was −3.9 (95% CI −5.4, −2.5) at 12 months. Patients in the lcSSc group remained stable: the mean change was 0.3 (95% CI −0.7, 1.3) at month 12, as shown in Fig. 1 and Table 2. Data from patients who either died ($n = 6$) or underwent ASCT ($n = 5$) after 12 months' follow-up was excluded from the moment of either event for the continuous mRSS analyses and regarded as non-response for the dichotomous outcome. Forty-three percent of the dcSSc patients achieved a response according to our criteria at month 12. After 24 and 36 months, response rates still increased slightly (Table 3); however, it should be noted that at 24 and 36 months, the data of >20% mRSSs was missing.

Fig. 1 Course of mean Modified Rodnan Skin Score (mRSS) over 36 months



A. dcSSc. B. lcSSc. Mean improvement in mRSS in the dcSSc group after 12 months −3.9 (95% CI −5.4, 2.5); mean improvement in mRSS in the lcSSc group after 12 months 0.3 (95% CI −0.7, 1.3).

Table 2 Mean change in mRSS compared with baseline

Time (months)	6	12	24	36
lcSSc ($n = 44$)	0.2 (−0.8 to 1.2)	0.3 (−0.7 to 1.3)	−0.4 (−1.6 to 0.8)	−0.3 (−2.0 to 1.4)
dcSSc($n = 99$)	−0.5 (−1.8 to 0.9)	−3.9 (−5.4 to −2.5)	−5.2 (−6.7 to −3.7)	−6.6 (−8.6 to −4.6)

Negative value is a decrease in mRSS, positive value is an increase in mRSS. mRSS: modified Rodnan skin score.

Table 3 Proportion of responders over time

Time (months)	6	12	24	36
dcSSc ($n = 99$)	19 (20%; 95% CI 11%, 27%)	42 (43%; 95% CI 33%, 53%)	47 (47%; 95% CI 36%, 58%)	49(50%; 95% CI 39%, 61%)

Data are shown as frequencies and percentages (%). lcSSc patients were excluded as they were unable to meet the response criterion due to a low baseline mRSS.

Table 4 Variables tested in univariate analysis

Variable	OR	*P* value
Age	1.00 (0.98, 1.04)	0.613
Female gender	0.62 (0.26, 1.45)	0.265
Disease duration	1.01 (0.89, 1.14)	0.933
Baseline mRSS	0.94 (0.89, 0.99)	0.020
All 12 infusions completed	0.26 (0.07, 0.93)	0.037
ILD at baseline	0.54 (0.25, 1.32)	0.190
ANA positivity	2.78 (0.24, 33.33)	0.412
Anti-SCL70	0.89 (0.39, 2.04)	0.785
Other antibodies	1.28 (0.55, 2.94)	0.570
Non-response at 6 months	41.11 (5.15, 327.91)	<0.001

OR: odds ratio (95% CI); mRSS: modified Rodnan skin score; ILD: interstitial lung disease.

Table 5 Multivariate logistic regression

Variable	Odds ratio (95% CI)	*P* value
Baseline mRSS	0.92 (0.91, 1.01)	0.114
All 12 infusions completed	0.26 (0.05, 1.39)	0.117
ILD at baseline	0.71 (0.25, 1.09)	0.520
Non-response at 6 months	36.26 (4.31, 304.78)	0.001

Results of multivariate logistic regression with non-response on skin thickening as dependent variable, and variables univariately associated with $P < 0.2$ as independent variables. OR: odds ratio; mRSS: modified Rodnan skin score: ILD: interstitial lung disease.

Prediction model for dcSSc patients

In univariate analyses, baseline mRSS, non-response at 6 months and not completing 12 i.v. CYC pulses were significant predictors of non-response at month 12 (Table 4). For the latter variable, it should be mentioned that patients ($n = 19$) who did not achieve a response at month 6 did not continue i.v. CYC for that reason. Table 4 displays all variables that were univariately tested. In the multivariate model including all variables with a univariate $P < 0.2$, only non-response at month 6 was a significant predictor of non-response at month 12 (Table 5).

Discussion

This study shows that 43% of patients with early dcSSc who received i.v. CYC pulse therapy had a clinically significant response to this treatment according our criteria after 12 months of therapy. The mRSS decreased −3.9 points at 12 months in dcSSc group, whereas the lcSSc group remained stable. Until now, there was little evidence for the efficacy of i.v. CYC pulse therapy on skin thickening, despite its frequent use, and this study adds to the knowledge about the effect of i.v. CYC pulse therapy on skin thickening in SSc. In addition, we found that if dcSSc patients had not responded by month 6 they had a 29% chance of responding by month 12. These findings could help physicians and patients to decide whether or not to continue i.v. CYC pulse therapy after month 6.

Our findings are comparable with those of a recent study on the effect of oral CYC compared with MMF on skin thickening. In that study, a response to treatment was found in 40% of the CYC group after 12 months [14]. In a European observational study of early dcSSc patients, the ESOS study, no treatment resulted in a decrease in mRSS of 2.2 after 12 months, and i.v. CYC pulse therapy resulted in a decrease of 3.3 after 12 months, using a lower dosage of CYC [19]. We found in our study a comparable

decrease in mRSS of 3.9 after 12 months. The mean improvement in mRSS in the i.v. CYC pulse therapy arm of the ASTIS trial was substantially higher than that shown in our results (−8.8 at 1 year) [21]. An explanation for this could be that patients included in that trial had a higher baseline mRSS compared with the patients in our study, leaving more room for improvement and making a regression to the mean effect more likely.

This study has several strengths. We analysed data from a large group of patients followed by the same rheumatologists according to a standardized protocol with a long follow-up time, including repeated measurements of the mRSS. In addition, we used data from clinical practice, which improves generalizability of the results.

There are also limitations in our study. Most importantly, the study lacked a control group, and we cannot therefore exclude the possibility that the improvement in skin thickening is due to factors other than efficacy of i.v. CYC pulse therapy, such as the natural course of disease and/or regression to the mean, or the placebo effect in the assessor or patient. In addition, as a result of the retrospective nature of this study, we did not have a mRSS for every patient at all time points, so we used multiple imputation to reduce the impact of the missing data. However, the proportion of missing mRSSs at the 24 and 36 month time points was significant, reducing the reliability of the estimates at these times [18–20].

It is known from previous studies that the mRSS can decline even if patients are not treated, but studies on the course of the mRSS in dcSSc patients show that those who spontaneously improve have longer disease duration and a higher mean mRSS at baseline than patients in our study [3]. Studies that examine patients that worsen show that dcSSc patients with shorter disease duration and lower mRSS at baseline are more likely to worsen, which applies to our patients, as our patients had a mean disease duration of 3.8 months at inclusion [18, 19]. Therefore, we believe the reduction in the mRSS seen in this study is at least partly, but probably for the main part, a treatment effect rather than reflecting the natural course of disease.

There is no international consensus on mRSS response criteria. The measured effect of therapy is partly determined by the criteria that are chosen for response. We used an improvement of $\geqslant$5 units and 25% from baseline, because these criteria are both considered clinically significant and are most widely used in literature [18, 19]. Recently, it has been debated whether the mRSS is the most appropriate outcome measure in SSc placebo-controlled trials. The value of composite outcome measurements, such as the combined response index for SSc (CRISS), mostly including the mRSS, have recently been examined [10, 22]. However, in daily clinical practice the mRSS remains important for the assessment of progression of dcSSc.

In conclusion, treatment with i.v. CYC pulse therapy should be considered as a treatment option for dcSSc after either MTX has failed or as first-line therapy for patients. A clinical meaningful improvement in mRSS was reached in 43% of the patients after 12 months. Our data show that after 6 months of therapy, a decision can be made according to the response to therapy regarding whether to continue i.v. CYC pulse therapy or to choose another therapy (such as MMF or ASCT) if patients are eligible.

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