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Review

Is immunosuppressive therapy the anchor treatment to achieve remission in systemic sclerosis?

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Abstract

Since activation of the immune system and a perivascular infiltrate of inflammatory cells are key features of SSc, immunosuppression has long been considered to be an anchor treatment. Non-selective immunosuppression remains central to the treatment of interstitial lung disease (ILD) and skin involvement, with CYC most widely used to obtain remission. The use of MTX as a first-line agent may be considered in the presence of skin involvement without ILD. More recently, MMF has shown encouraging results in observational studies, but still needs more formal evaluation to verify if it can be considered an alternative drug to CYC or a maintenance agent such as AZA. Rituximab has provided promising results in small open-label studies and other novel therapies targeting specific molecular and cellular targets are under evaluation. Patients with rapidly progressing diffuse cutaneous SSc should be evaluated for haematopoietic stem cell transplantation.

Key words: systemic sclerosis, cyclophosphamide, methotrexate, mycophenolate mofetil, azathioprine, rituximab, hematopoietic stem cell transplantation.

REVIEW ARTICLE

Introduction

SSc is a multisystem autoimmune disease characterized by an obliterative microvasculopathy, activation of the immune system and fibrosis of the skin and internal organs. Activation of the immune system is a key process underlying SSc, influencing both fibrosis and the progression of vascular involvement. While non-selective immunosuppressants have been reported to control skin and lung inflammation, their beneficial effects are often tempered by the development of side effects leading to significant morbidity and even mortality. There is therefore a pressing need to evaluate novel therapies targeting specific cellular and/or molecular immune effectors that are thought to play crucial pathophysiological roles within this condition [1].

The ideal aim of therapy in SSc is to induce remission, thereby stopping disease progression and, if possible, reversing disease-related skin and internal organ changes. At present, CYC is the most widely used immunosuppressant to obtain remission. Immunoablative doses of CYC, followed by autologous haematopoietic stem cell transplantation (HSCT), may be successfully employed in patients with severe, rapidly progressing SSc [2] (Fig. 1).

In patients with skin involvement without interstitial lung disease (ILD), the use of MTX as a first-line treatment has been suggested [3, 4] (Fig. 1), as two randomized controlled trials (RCTs) have shown that MTX may be useful in improving skin score [5, 6]. In some cases MMF may help to achieve remission, but currently it is commonly used, as well as AZA, as a maintenance treatment [4]. The purpose of this review is to analyse the existing evidence for the use of immunosuppressants to slow progression and achieve remission in SSc and thus to be considered as an anchor treatment.

Cyclophosphamide

CYC (Table 1) is an alkylating agent that is cytotoxic to both resting and dividing lymphocytes [7]. Alkylating agents exhibit significant toxicities that must always be considered in the risk–benefit assessment.

CYC may be administered either orally or intravenously. The use of intermittent i.v. pulses reduces the toxicity [8]. At present there is no consensus about the dose of CYC (which ranges from 0.5 to 2 g/m^2 for each pulse) or the duration of therapy (generally 6–18 months).

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Fig. 1 Different immunosuppressive approaches to SSc treatment are presented: the aim is to achieve and maintain remission.

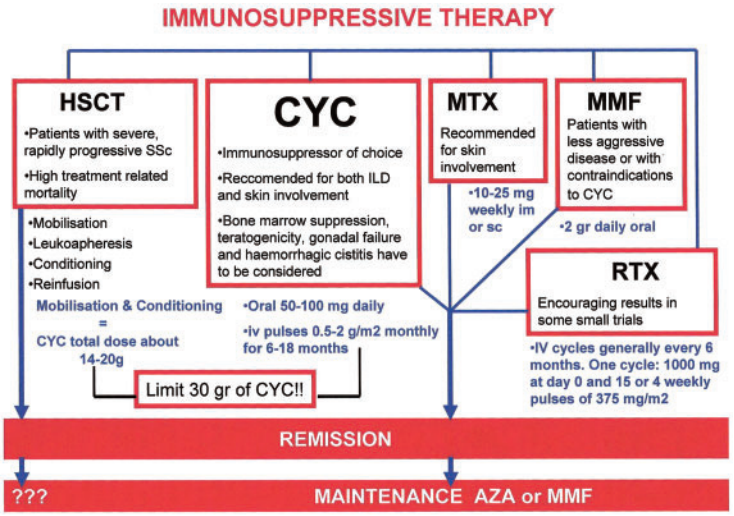

Table 1 Non-selective immunosuppressants: side effects, fertility, pregnancy and lactation

	Side effects	Fertility	Pregnancy	Lactation
CYC [3, 86, 87]	Leucopenia, 6–32%; thrombocytopenia, 0–4%; nausea/vomiting, 19–45%; haemorrhagic cystitis, 10–15%; increased risk of infections and malignancies (in particular bladder carcinoma)	Risk of sustained infertility	Contraindicated (potential risk of teratogenicity)	Contraindicated
MTX [87, 88]	Leucopenia, 1–4%; thrombocytopenia, 1–2%; nausea/vomiting, 10–18%; elevation of transaminases, 8–38%; fibrosis/cirrhosis, 4–20%; acute and chronic pulmonary toxicity (rare); increased risk of infections	It does not seem to increase the risk of infertility	Contraindicated (teratogenicity)	Contraindicated
MMF [89, 90]	Leucopenia, 4–25%; thrombocytopenia, 1–2%; diarrhoea, 36%; nausea/vomiting, 9–20%; elevation of transaminases at high doses; increased risk of infections	It does not seem to increase the risk of infertility	Contraindicated (no adequate studies have been performed)	Contraindicated
AZA [87, 89, 91]	Leucopenia, 4–27%; thrombocytopenia, 0–5%; nausea/vomiting, 9–23%; diarrhoea, 0–1%; elevation of transaminases, 0–5%; increased risk of infections	It does not seem to increase the risk of infertility	It can be continued during pregnancy if potential benefits outweigh the potential risk	No data; hypothetical risk of immunosuppression in the newborn outweighs benefit
CSA [87, 89, 91]	Leucopenia, 2–6%; thrombocytopenia, 0–2%; nausea/vomiting, 4–40%; diarrhoea, 2–18%; decreased renal function, 50–87%; hypertension, 33%; neurotoxicity, 10–40%; gum hyperplasia, 4–12%; hypertricosis, 7–49%; elevation of transaminases, 0–8%; increased risk of infections	It does not seem to increase the risk of infertility	It can be continued during pregnancy if potential benefits outweigh the potential risk	Contraindicated

In an Italian study, patients were treated for 6 months with CYC pulses (1000 mg/m^2), with stabilization of pulmonary function in most patients [9] (Table 2). Thereafter the use of CYC in SSc-ILD was evaluated in two RCTs *vs* placebo. In the Scleroderma Lung Study I (SLS I), patients receiving 1 year of oral CYC had a significant but modest increase of forced vital capacity (FVC) ($P < 0.03$) and a significant improvement in skin thickness ($P < 0.008$) compared with placebo [10] (Table 2). The beneficial effects of CYC persisted or increased for several months after stopping the therapy, but were no longer apparent after 12 months, suggesting that maintenance treatment after

Table 2 CYC in the treatment of SSc

Author	No. of patients	Type of study	CYC treatment	Placebo/ alternative treatment	Steroids	Follow-up, months	Outcome measures
Tashkin *et al.* SLS I [10]	158	RCT	Oral, $\leqslant$2 mg/kg/day	Placebo	None	12	FVC (difference CYC *vs* placebo): +2.53% favouring CYC ($P<0.03$) DL_{CO} (difference CYC *vs* placebo): $P=0.43$ HAQ disability scores (difference CYC *vs* placebo): −0.16 favouring CYC ($P=0.008$) mRSS (difference CYC *vs* placebo): −3.6 favouring CYC ($P=0.008$)
Hoyles *et al.* [12]	45	RCT	i.v., 600 mg/m^2 monthly for 6 months followed by AZA 2.5 mg/kg/day for 6 months	Placebo	Prednisone 20 mg alternate days	12	FVC (difference CYC + AZA *vs* placebo): +4.19% favouring CYC + AZA ($P=0.08$) DL_{CO} (difference CYC + AZA *vs* placebo): $P=0.64$ HRCT: 40% of patients with improvement in CYC + AZA group *vs* 20% in placebo group
Nadashkevich *et al.* [13]	60	RCT	Oral, 2 mg/kg/day for 12 months then 1 mg/kg/day for 6 months	AZA, 2.5 mg/kg/day for 12 months then 2 mg/kg/day for 6 months	Prednisolone <15 mg/day	18	FVC (CYC group, difference T0 − T18): $P>0.05$ DL_{CO} (CYC group, difference T0 − T18): $P>0.05$ mRSS (CYC group, difference T0 − T18): −9.5 ($P<0.001$)
Giacomelli *et al.* [9]	23	Prospective	i.v., 1000 mg/m^2 monthly		25 mg/day for 1 month then 5 mg/day	6	FVC (difference T0 − T6): improvement or stabilization in 21 out of 23 patients DL_{CO} (difference T0 − T6): improvement or stabilization in 15 out of 23 patients
Airò *et al.* [92]	13	Prospective	i.v., 750 mg/m^2 every 3 weeks		MP 125 mg every 3 weeks	18	FVC (difference T0 − T18): $P=0.005$ DL_{CO} (difference T0 − T18): $P=0.10$
Beretta *et al.* [93]	33	Prospective	Oral, 2 mg/kg/day		Prednisone 25 mg/day for 3 months then 5 mg/day for 9 months	12	FVC (difference T0 − T12): $P>0.01$ DL_{CO} (difference T0 − T12): $P<0.001$
Davas *et al.* [94]	16	Prospective	i.v., 750 mg/m^2 monthly or oral 2–2.5 mg/kg/day		Prednisone 10 mg/day	12	FVC (difference T0 − T12): $P>0.05$ DL_{CO} (difference T0 − T12): $P=0.043$ HRCT (IV group): regression of disease extent in 7 out of 8 patients
Silver *et al.* [95]	14	Prospective	Oral 1–2 mg/kg/day		Prednisone <10 mg/day	24	FVC (difference T0 − T24): $P<0.001$ DL_{CO} (difference T0 − T24): $P>0.05$
Espinosa *et al.*[96]	37	Retrospective	i.v., 600 mg/m^2 monthly for 6 months, bimonthly up to 12 months and quarterly up to 24 months		50 mg/day reduced until a dose of 5–7.5 mg/day	24	The differences between FVC and DL_{CO} performed at baseline and at 6, 12 and 18 months were <10%, which means that CYC stabilized functional parameters

CYC is needed [11]. In another RCT, two groups were compared—one treated with six monthly pulses of CYC followed by AZA for 6 months and one with placebo. As for FVC, a trend towards a significant difference between the two groups ($P = 0.08$), favouring patients treated with CYC followed by AZA, was observed [12] (Table 2). Nadashkevich *et al.* [13] compared oral CYC with AZA in an 18-month RCT. A trend towards FVC improvement was observed in patients treated with CYC, while patients receiving AZA showed a significant decrease in FVC ($P < 0.001$) (Tables 2 and 3). In a recent meta-analysis in which the above mentioned RCTs and six observational studies were included, the use of CYC was associated with a non-statistically significant improvement in pulmonary function [14].

It is interesting to note that the use of an immunoablative dose of i.v. CYC without stem cell rescue has been reported to significantly improve skin thickening in active diffuse cutaneous SSc (dcSSc) [15]. In view of these results, the European League Against Rheumatism (EULAR) Scleroderma Trials and Research group (EUSTAR) recommends the use of CYC for the treatment of SSc-ILD and skin involvement with a warning about its toxicity [3]. CYC may also be considered for the treatment of SSc-related myocardial involvement [2].

Methotrexate

MTX (Table 1) is an antimetabolite drug that competitively inhibits dihydrofolate reductase, leading to impaired DNA and nucleotide synthesis. Other mechanisms that are involved in the immunomodulatory action of MTX are a decrease of proinflammatory cytokine (IL-1, IL-6, TNF-α) and inhibition of antigen-induced T-cell activation [7].

In SSc, MTX may be used for skin involvement, arthritis and myositis [4]. Two RCTs have evaluated the efficacy of MTX on skin and lung involvement. In the first, patients receiving MTX showed, after 24 weeks, a trend towards a statistical improvement of the modified Rodnan skin score (mRSS) in comparison with patients receiving placebo ($P = 0.06$), while no amelioration of pulmonary function was observed [5] (Table 4). In the second, data on mRSS slightly favoured the MTX group, but also in this case statistical significance was not reached, probably due to the small number of patients ($P < 0.17$) [6] (Table 4). Considering the difficulty in recruiting a sufficient number of patients and the low power to detect important treatment effects in rare diseases, recently 71 patients with dcSSc (35 treated with MTX and 36 with placebo) have been analysed using Bayesian models. In this study, the probability that treatment with MTX results in better mean outcomes than placebo was 94% for mRSS and 96% for the University of California at Los Angeles (UCLA) skin score. These data may suggest that MTX has a high probability of beneficial effects on skin score [16].

Mycophenolate mofetil

MMF (Table 1) is an antiproliferative drug that acts through inhibition of the inosine 5′-monophosphate dehydrogenase (IMPDH), an enzyme involved in the *de novo* synthesis of purines. MMF preferentially inhibits the type II isoform of IMPDH, which is selectively expressed on activated T and B lymphocytes. Thanks to the relatively specific inhibitory effect on lymphocytes, MMF has a more favourable side-effect profile than CYC [1, 7, 17, 18].

At present there are no RCTs that evaluate MMF in the treatment of SSc. Data from observational studies suggest that MMF could be useful both for pulmonary and skin involvement [17–28] (Table 5). In a pilot study, patients treated with MMF in association with low-dose prednisone (5–10 mg/day) and after i.v. methylprednisolone (MP) pulses showed a significant improvement both in mRSS and pulmonary function [17] (Table 5). A significant decrease of mRSS after treatment with MMF was also observed in other three studies, two prospective and one retrospective [18–20] (Table 5). Nevertheless, in the study of Herrick *et al.*, and in a retrospective analysis of Nihtyanova *et al.*, there was no difference in terms of change of mRSS between patients treated with MMF or with other immunosuppressants [19, 21] (Table 5). Koutroumpas *et al.* [22] did not find any effect of MMF on mRSS (Table 5). As for pulmonary function, Liossis *et al.* [23], in a prospective open-label trial, showed that MMF was associated with a significant increase in diffusing capacity of the lung for carbon monoxide (DL_{CO}) and a trend toward an increase in FVC (Table 5). Two retrospective analyses described a significant increase of FVC and vital capacity (VC), but no significant changes in DL_{CO} [22, 24] (Table 5). In other studies, treatment with MMF resulted in the stabilization of pulmonary function without significant improvement [18, 25–27] (Table 5). Recently a prospective, open-label trial evaluated the effects of enteric-coated mycophenolate sodium (EC-MPS) on skin and pulmonary manifestations. To assess the pulmonary response the authors evaluated not only pulmonary function tests (PFTs), but also high-resolution CT (HRCT) with histography. FVC and DL_{CO} did not change significantly and HRCT showed a non-significant increase in lung density [28] (Table 5). A limitation of these studies is the small number of patients included. Therefore the role of MMF in SSc needs to be evaluated in RCTs on a larger number of patients.

At present there is great interest in comparing MMF with CYC as a first-line treatment. In the USA, the Scleroderma Lung Study II (SLS II), an RCT comparing MMF to CYC in the treatment of SSc-associated ILD, is ongoing. The European Scleroderma Observational Study has a similar aim, as it involves patients treated with MTX, MMF, CYC or without an immunosuppressive drug as first-line therapy in order to compare the efficacy of the commonly used immunosuppressants.

Azathioprine

AZA (Table 1) is a purine analogue that inhibits the proliferation of inflammatory cells through interference with the *de novo* synthesis of purine by its active metabolites. In two small retrospective studies, patients treated with AZA showed a stabilization of lung function [29, 30] (Table 3).

Table 3 AZA in the treatment of SSc

Author	No. of patients	Type of study	AZA treatment	Duration, months	Outcome measures
Nadashkevich *et al.* [13]	60	RCT	AZA 2.5 mg/kg/day for 12 months then 2 mg/kg/day for 6 months *vs* CYC 2 mg/kg/day for 12 months then 1 mg/kg/day for 6 months	18	FVC (difference T0 – T18, AZA group): −11.1% ($P < 0.001$) DL_{CO} (difference T0 – T18, AZA group): −11.6% ($P < 0.001$) mRSS (difference T0 – T18, AZA group): +0.2 ($P > 0.05$)
Dheda *et al.* [29]	11	Retrospective		12	FVC (difference T0 – T12): +9.1% ($P = 0.101$)
Mass *et al.* [30]	19	Retrospective	AZA 2–2.5 mg/kg/day	Mean 47 months (range 6–114 months)	No patient had worsening of lung function
Hoyles *et al.* [12]	45	RCT	AZA 2.5 mg/kg/day as maintenance treatment after 6 months of i.v. CYC	12	FVC (difference CYC + AZA *vs* placebo): +4.19% favouring CYC + AZA ($P = 0.08$) DL_{CO} (difference CYC + AZA *vs* placebo): $P = 0.64$
Paone *et al.* [31]	13	Prospective	AZA 100 mg/day	12	No outcome measures deteriorated during maintenance treatment with AZA

Table 4 MTX in the treatment of SSc

Author	No. of patients	Type of study	MTX treatment	Duration, months	Outcome measures
van den Hoogen *et al.* [5]	57	RCT (6 months) + prospective (6 months)	MTX 15–25 mg/week i.m. *vs* placebo	6 + 6	VC (difference MTX *vs* placebo): $P = 0.37$ DL_{CO} (difference MTX *vs* placebo): $P = 0.48$ Lung fibrosis (difference MTX *vs* placebo): $P = 0.50$ mRSS (difference MTX *vs* placebo): MTX −0.77, placebo +1.2 ($P = 0.06$) General Health (difference MTX *vs* placebo): $P = 0.19$ VC (difference MTX group, T0 – T12): $P = 0.42$ DL_{CO} (difference MTX group, T0 – T12): $P = 0.29$ mRSS (difference MTX group, T0 – T12): $P = 0.22$ General Health 0–100 VAS (difference MTX group, T0 – T12): +9.9 ($P = 0.08$)
Pope *et al.* [6]	71	RCT	MTX 15 mg/week, oral *vs* placebo	12	DL_{CO} (difference MTX *vs* placebo): trend in favour of MTX but no statistical difference ($P < 0.2$) mRSS (difference MTX *vs* placebo): trend in favour of MTX but no statistical difference ($P < 0.17$)
Su *et al.* [35]	18	Randomized, phase I single-blind study	MTX 20 mg/week, oral *vs* RAPA	12	FVC (difference MTX *vs* RAPA): MTX 1.2%, RAPA −10.6% ($P > 0.05$) DL_{CO} (difference MTX *vs* RAPA): MTX −5.2%, RAPA −12.7% ($P > 0.05$) mRSS (difference MTX *vs* RAPA): MTX −6.8, RAPA −5.6 ($P > 0.05$) Patient global assessment VAS 0–100 (difference MTX *vs* RAPA): MTX −11, RAPA −11.5 ($P > 0.05$)

VAS: visual analogue scale.

Table 5 MMF in the treatment of SSc

Author	No. of patients	Type of study	MMF treatment	Duration, months	Outcome measures
Vanthuyne *et al.* [17]	16	Prospective	2 g/day with prednisolone (5–10 mg/day) preceded by three consecutive daily MP pulses and five additional monthly MP pulses	12	VC (difference T0 – T12): +10% ($P = 0.099$) DL_{CO} (difference T0 – T12): +13% ($P = 0.0009$) mRSS (difference T0 – T12): −7 ($P < 0.0001$) HAQ-DI (difference T0 – T12): −0.5 ($P = 0.021$)
Derk *et al.* [18]	15	Prospective	2–3 g/day	12	mRSS (difference T0 – T12): −14.1 ($P < 0.0001$) FVC (difference T0 – T12): $P > 0.05$ DL_{CO} (difference T0 – T12): $P > 0.05$
Herrick *et al.* [19]	147	Prospective	Five protocols: (i) i.v. CYC followed by MMF; (ii) ATG; (iii) MMF alone (2 g/day); (iv) no immunosuppressants; (v) other immunosuppressants	12	mRSS: significant decrease with all protocols except protocol 1 No significant differences between protocols ($P = 0.43$)
Le *et al.* [20]	68	Retrospective	2 g/day *vs* other treatments (relaxin, D-penicillamine, oral bovine collagen)	12	mRSS (MMF *vs* relaxin): $P = 0.059$ (6 months) mRSS (MMF *vs* D-penicillamine): MMF −7.59, penicillamine −2.47; $P < 0.001$ favouring MMF (12 months) mRSS (MMF *vs* collagen): MMF −7.59, collagen −3.4; $P = 0.002$ favouring MMF (12 months)
Nihtyanova *et al.* [21]	172	Retrospective	109 with MMF *vs* 63 with other immunosuppressants	60	FVC (difference MMF *vs* other immunosuppressants): $P > 0.05$ mRSS (difference MMF *vs* other immunosuppressants): $P > 0.05$
Koutroumpas *et al.* [22]	10	Retrospective	2 g/day	12	FVC (difference T0 – T12): +7.6 ($P = 0.04$) DL_{CO} (difference T0 – T12): $P = 0.66$ mRSS (difference T0 – T12): $P = 0.55$
Liossis *et al.* [23]	5	Prospective	2 g/day	4–6	FVC (difference T0 – T4/6): +10.6% ($P = 0.057$) DL_{CO} (difference T0 – T4/6): +11.2% ($P = 0.033$)
Gerbino *et al.* [24]	13	Retrospective	1–2 g/day	12	VC (difference T0 – T12):+4% ($P = 0.002$) DL_{CO} (difference T0 – T12): $P = 0.38$
Zamora *et al.* [25]	17	Retrospective	2 g/day	24	FVC (difference T0 – T24): $P = 0.57$ DL_{CO} (difference T0 – T24): $P = 0.84$
Swigris *et al.* [26]	28[a]	Retrospective	2 g/day (median dose)	371 days (median)	FVC (difference T0 – T371 days): $P = 0.77$ DL_{CO} (difference T0 – T371 days): $P = 0.41$ TLC (difference T0 – T371 days): $P = 0.15$

(continued)

Table 5 Continued

Author	No. of patients	Type of study	MMF treatment	Duration, months	Outcome measures
Simeón-Aznar *et al.* [27]	14	Prospective	MPS 1440 mg/day	12	FVC (difference T0 – T12): $P = 0.951$ DL_{CO} (difference T0 – T12): $P > 0.05$
Henes *et al.* [28]	11	Prospective	EC-MPS 1440 mg/day	12	FVC (difference T0 – T12): $P = 0.469$ DL_{CO} (difference T0 – T12): $P = 0.344$ mRSS (difference T0 – T12): $P = 0.115$ HRCT with histography: MLD (difference T0 – T12): $P = 0.4$ HAV (difference T0 – T12): $P = 0.172$

[a]Nine patients with SSc, 19 with other connective tissue diseases. TLC: total lung capacity; MLD: median lung density; HAV: high attenuation value; MPS: mycophenolate sodium.

However, these positive results have not been confirmed by an RCT in which AZA is compared with oral CYC (Tables 2 and 3). In this RCT, patients treated with AZA, unlike patients receiving CYC, showed a significant worsening of both FVC and DL_{CO} [13] (Table 3).

The use of AZA as maintenance treatment has been evaluated in a prospective, open-label study in which 13 patients treated with AZA (100 mg/day) for 12 months after i.v. CYC maintained the improvement obtained with CYC [31] (Table 3). In conclusion, AZA is not as effective as CYC as an induction treatment, but it may be an option as a maintenance drug.

Other non-selective immunosuppressants

CSA interferes with T cell production of IL-2 and inhibits T cell proliferation [1]. In SSc, CSA has been shown to improve skin disease, but with no beneficial effect on lung and cardiac involvement and a high frequency of adverse events [32–34] (Table 1). Because the overall effect of CSA seems to be modest and limited to the skin, and in view of its narrow therapeutic range and not uncommon severe side effects (i.e. renal toxicity and hypertension), we suggest that the use of CSA in SSc, if possible, should be avoided.

Sirolimus (rapamycin, RAPA) blocks T and B lymphocyte response to cytokines, including IL-2 and other activation stimuli [1]. Recently a single-blind, randomized, phase I study compared the safety and efficacy of RAPA *vs* oral MTX (Table 4). RAPA demonstrated a reasonable safety profile. As for efficacy, both in patients treated with RAPA and MTX, a statistically significant improvement of mRSS was observed while pulmonary function declined in patients treated with RAPA but not in the MTX group [35] (Table 4). In view of some reports of lung toxicity in patients receiving RAPA [36, 37] after organ transplantation, the favourable safety profile of RAPA and its efficacy in skin involvement obtained in this phase I RCT, need to be confirmed in larger trials.

Antithymocyte globulin (ATG) therapy acts through T cell depletion, but the therapeutic efficacy also relies on its ability to interfere with lymphocyte transmigration and to cause B cell depletion [1]. In a small open-label trial, administration of a single course of ATG (10 mg/kg for 5 days) to 10 SSc patients did not result in any improvement in skin and pulmonary involvement [38]. In another study, 13 patients were treated with ATG as induction treatment followed by MMF. The mean skin score decreased significantly ($P < 0.01$) during the study, while no significant change in FVC was observed [39]. Since one patient died from scleroderma renal crisis (SRC) after ATG and five experienced a serum sickness reaction, concerns have been raised about the safety of ATG.

Corticosteroids

Corticosteroids are used in SSc for their anti-inflammatory and immunosuppressive properties. They may also have an anti-fibrosing effect by reducing the synthesis of

mucopolysaccharides necessary for collagen formation [40]. The use of MP pulses in association with CYC pulses has been suggested as induction treatment of SSc-ILD, with favourable results [41, 42]. Data have also highlighted the need for early treatment, since patients with established severe functional impairment have responded less well to treatment [42].

Oral corticosteroids at a low to medium dose may be used to control arthritis and myositis in SSc patients [4]. Nevertheless, four retrospective studies suggest that the use of steroids, in particular at medium to high doses, may be associated with a higher risk of SRC [43–46]. Steen and Medsger [43] observed that for many SRC patients, a recent history of corticosteroid use (e.g. prednisone or equivalent at $\geqslant$15 mg/day) preceded an SRC diagnosis. Nevertheless those SSc patients that are most often treated with corticosteroids are those with early aggressive dcSSc that are at highest risk for SRC [47].

Biologic immunotherapies

Rituximab

Rituximab (RTX) is a chimeric monoclonal antibody directed against CD20, an antigen expressed on early pre-B and mature B cells [1]. B cell abnormalities, characterized by autoantibody production, hypergammaglobulinaemia and polyclonal B cell hyperactivity are important features of SSc [48]. For this reason, targeting B cell activation in SSc has been employed in some small open-label studies. In eight patients treated with a single cycle of RTX (1000 mg administered at baseline and day 15), a significant improvement in skin score was observed at 24 weeks (−10.5, $P < 0.001$) [49]. However, another open-label study in which 15 patients received the same cycle of RTX did not show any significant change either in mRSS or in FVC over the same period [50]. A recent pilot study randomized 14 dcSSc patients with ILD to receive either RTX plus standard therapy (eight patients) or standard therapy alone (six patients). Patients in the treatment arm received two cycles of RTX at baseline and 24 weeks (each cycle consisting of four weekly RTX infusions, 375 mg/m^2/week). After 1 year of follow-up, patients treated with RTX, but not controls, showed a significant improvement in lung function (FVC +7.5%, $P = 0.0018$; DL_{CO} +9.75%, $P = 0.017$) and skin score (mRSS −5.13, $P < 0.001$) [51]. The eight patients treated with RTX then received another two cycles at an interval of 6 months. After a follow-up of 2 years, a significant improvement in pulmonary function (FVC +9%, $P < 0.0001$; DL_{CO} +10.88%, $P < 0.001$) and skin involvement was observed (mRSS −8.63, $P < 0.0001$) [52]. Although these results are encouraging, the number of patients included is small and formal recommendations for the use of RTX await validation from RCTs.

Other biologic immunotherapies

Abatacept is a recombinant fusion protein that binds to CD80/CD86 on the surface of antigen-presenting cells (APCs), thus preventing their interaction with CD28 on T cells. Blockade of CD28 binding prevents the so-called second signal of T cell activation [53]. A recent observational study from EUSTAR showed that abatacept is useful to control joint involvement in SSc [54].

Alefacept is a recombinant fusion protein with a similar mechanism of action to abatacept, as it inhibits the co-stimulatory interaction between APC and CD2 memory T effector cells. In a small pilot study, eight patients with SSc-ILD were treated with alefacept. The drug was well tolerated and a stabilization of lung function was observed at 26 weeks after the first infusion [55].

Basiliximab is a monoclonal antibody that blocks the α chain of the IL-2 receptor (CD25) expressed on T cells, leading to the inhibition of their activation and proliferation. In an open label study, 10 dcSSc patients treated with basiliximab showed a significant improvement of mRSS (−15, $P = 0.015$) and a trend toward an increase in FVC (6.3%, $P = 0.073$) [56].

Since the role of TNF-α in fibrogenesis is unclear, the potential use of anti-TNF-α agents in SSc has been a matter of debate. The majority of *in vitro* studies have shown antifibrotic effects of TNF-α, as it suppresses the production of collagen [57–60]. In contrast, *in vivo* animal studies have demonstrated an antifibrotic function of anti-TNF-α [61, 62].

In a retrospective analysis, 15 of 18 SSc patients with joint involvement treated with etanercept showed a significant decrease in signs of inflammation, while skin and pulmonary function were not influenced by the therapy [63]. In a pilot study on nine dcSSc patients treated with etanercept, positive effects on both skin and joint involvement were observed [64].

An open-label study in which 16 patients received five infusions of infliximab (5 mg/kg) reported no significant change in skin score but a significant decrease in two laboratory markers of collagen synthesis [65]. Anti-infliximab antibodies were found in ~30% of patients (5/16) and were significantly associated with suspected infusion reactions. In another study, five patients received infliximab, administered at weeks 0, 2, 6, 14, with MTX followed by etanercept (without MTX). All the patients showed an improvement of mRSS and HAQ Disability Index (HAQ-DI) after 6 months [66].

An investigation carried out among EUSTAR centres detected that most of the experts did not recommend the routine use of anti-TNF-α in SSc and that the improvement obtained with anti-TNF-α is mainly seen in patients with arthritis, whereas the effects on fibrosis remain unclear [67, 68]. A recent analysis showed that the use of anti-TNF-α in SSc is potentially associated with an increased risk of malignancy [69].

Tocilizumab is a monoclonal antibody directed against the IL-6 receptor. IL-6 is a proinflammatory cytokine whose expression is reported to be high in both skin and serum of SSc patients [70]. Fibroblasts isolated and cultured from the lesional skin of SSc patients constitutively produce higher levels of IL-6 than non-lesional or healthy donor fibroblasts [71, 72]. These data justify further explorations of IL-6 as a therapeutic target in SSc.

Skin softening has been reported in two SSc patients treated with tocilizumab [73]. A recent observational study from EUSTAR showed that SSc patients with arthritis receiving tocilizumab had a significant improvement in joint involvement (significant reduction of 28-joint count DAS) [54]. A phase II/III, multicentre, randomized, double-blind, placebo-controlled trial to asses the efficacy and safety of tocilizumab *vs* placebo in patients with SSc is now under way.

Alemtuzumab (CAMPTH-1H) is a monoclonal antibody targeting CD52, a protein expressed on the surface of mature T and B cells. A rapid and substantial improvement in skin score has been reported in a patient with SSc secondary to polyvinyl chloride treated with alemtuzumab [74].

Haematopoietic stem cell transplantation

HSCT preceded by high-dose immunosuppressive therapy (HDIT) has been used over the last 15 years for the treatment of severe autoimmune diseases refractory to approved therapies. The underlying hypothesis is that intensive immunosuppression ablates the immune response driving disease activity and infused haematopoietic progenitors allow outgrowth of a non-autoreactive immune system [75].

HSCT involves four steps: mobilization, leucoapheresis, conditioning and reinfusion. The aim of mobilization is to increase the number of stem cells in the peripheral blood in order to harvest sufficient cells during leucoapheresis. Mobilization is performed using CYC (2–4 g/m^2) followed by growth factors. Once mobilized, stem cells are harvested via leucoapheresis and then cryopreserved. Conditioning consists of intensive immunosuppression in order to ablate the immune response driving disease activity. It may be performed using a non-myeloablative (CYC with or without ATG) or a myeloablative regimen (CYC with total body irradiation or busulfan). Both the efficacy and toxicity of HSCT depend on conditioning. Stem cells are re-infused 24–48 h after conditioning [76, 77].

Data collected from the European Group for Blood and Marrow Transplantation and the European League Against Rheumatism (EBMT/EULAR) Working Party on Autoimmune Diseases database reported remarkable improvements of skin involvement after HSCT (improvement of mRSS >25% in 69% of patients). However, lung function did not change significantly. In the first patients treated, a high mortality rate related to the transplantation was reported (17%) [78]. The subsequent report from the EBMT/EULAR registry confirmed the beneficial effects on skin involvement with a lower mortality rate (8.7%), obtained through strict exclusion criteria and changes to the conditioning protocols (i.e. avoidance of total body irradiation) [79].

In the USA, a pilot phase II single-arm study with HDIT and HSCT was performed in 34 patients with dcSSc. A significant decrease in skin score was observed (mRSS −22.08, $P < 0.001$) while lung, heart and kidney function, in general, remained clinically stable. Treatment-related mortality was very high (23%) [80]. Based on these preliminary findings, two multicentre, prospective RCTs of CYC *vs* HSCT are now under way: Autologous Stem Cell Transplantation in Scleroderma (ASTIS) in Europe and Scleroderma: Cyclophosphamide or Transplant (SCOT) in the USA. Initial results of the ASTIS study show fewer deaths in the transplant arm [20.2% (16/79)] than in the controls [31.2% (24/77)]. While in the transplant group eight deaths were treatment related, compatible with a 100-day treatment-related mortality of 10% (8/79), in the control group none died from treatment-related causes and most deaths were due to progressive disease [81].

At present HSCT seems to be the most effective treatment for skin involvement [78–80] in SSc and could be considered as a treatment option for patients with poor prognosis dcSSc. However, before deciding for transplantation, the high treatment-related risk of death should always be considered and discussed with the patient because no other treatment has such a high risk. For a more extensive use of this treatment procedure, it is necessary to reduce the treatment-related toxicity.

Conclusions

Since SSc is such a heterogeneous disease, it is of paramount importance to identify all predictors of disease progression. First, a physician should be able to distinguish the two major subsets depending on the extent of skin sclerosis: limited cutaneous SSc (lcSSc), when only skin distal to the elbows and knees is affected (with or without face involvement) and dcSSc, when skin thickening also includes the trunk, upper arms and thighs [2]. lcSSc patients have a better prognosis and, if they do not have ILD, may usually be treated symptomatically. Conversely, the use of immunosuppression is generally recommended in patients with dcSSc. When choosing an immunosuppressant drug, patient preference, cost, toxicity and contraindications should be taken into account. Treatment should be started early in the course of the disease in order to possibly block or significantly slow disease progression before damage occurs. The aim of immunosuppressive therapy is to induce remission, or at least to achieve low disease activity. In dcSSc the challenge is to identify patients who need more aggressive treatment because of a higher risk of developing complications. Poor prognostic factors are extension and rapid worsening of skin and lung disease, myocardial involvement, pericardial effusion, renal involvement, skin pigmentation disturbances, tendon friction rubs and positivity for anti-topoisomerase or RNA polymerase III autoantibodies. Moreover, the early disease phases are characterized by the highest risk of worsening [2, 47, 82–84]. In dcSSc patients at risk of progression, an aggressive treatment with high-dose i.v. CYC or HSCT should be considered to obtain remission. At present there is no consensus about the regimen of CYC administration and a trial to compare different CYC regimens in terms of efficacy and toxicity is warranted. It is worth noting that while some beneficial effects on ILD have been described with CYC [9–13], preliminary results from HSCT have not reported lung improvement, despite the high dose of CYC used for conditioning [78, 80].

CYC is the first-line agent that has shown more favourable results during the early phases of the disease. After CYC, administered at a maximum dose of 30–50 g, as patients receiving ⩾80 g were shown to develop malignancy [85], patients should promptly receive a maintenance treatment with MMF (2 g/day) or AZA (2–3 mg/kg/day) in order to preserve the benefits obtained with CYC. In fact, it has been shown that the effects of CYC dissipated 6–12 months after the end of the therapy [11]. Since MMF has shown encouraging results in observational studies, this drug may be considered as an alternative to CYC in young women who would like to have a child; however, MMF still needs to be evaluated in RCTs. MTX, at a dose of 10–25 mg/week, has to be considered as a first-line therapy in patients with skin involvement without ILD [3–6].

Since the disease usually has waves of activity, patients should be closely monitored to identify complications in real time and provide timely treatment. Indeed, it is not uncommon that a patient treated with MTX for skin thickening is switched to CYC because ground-glass opacities become evident on HRCT. A clear algorithm on the management of immunosuppressants in SSc is still lacking. Therefore the results of ongoing and future trials are of paramount importance to achieve standardization of treatment.

In the recent years, new treatments directed towards specific molecular and cellular effectors have been proposed. Although some of them, such as RTX, seem to be promising, the value of the current results is limited by the small number of patients included in the studies. Ignorance about the key factor orchestrating the high number of downstream effectors in SSc pathogenesis still remains a significant limitation, in particular in developing drugs directed towards specific effectors.

Despite the fact that mortality due to SRC is reduced, overall survival has not changed significantly over the past 40 years [82]. SSc remains the connective tissue disease with the worst prognosis and in some cases all treatments at our disposal are completely ineffective.

At present, non-selective immunosuppressants may be considered the anchor drugs to achieve remission, or at least to slow disease progression, even if in some cases all treatments at our disposal are ineffective. Larger trials are warranted in order to prevent damage to internal organs and preserve quality of life in a growing number of SSc patients.

Rheumatology key messages

- Although there are recently proposed therapies against specific effectors, immunosuppressants are still the main treatment for dcSSc.
- SSc patients, at risk of progression, should receive cyclophosphamide first, then MMF or AZA.

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