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Pharmacotherapy of Systemic Sclerosis

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Abstract

Importance of the field—Systemic-sclerosis (SSc) is an uncommon autoimmune disease with variable degrees of fibroproliferation in blood vessels and certain organs of the body. Presently, there is no cure for SSc. The purpose of this article is to review the current literature regarding pathogenesis and treatment of complications of SSc.

Areas covered in this review—All available articles regarding research related to SSc pathogenesis and treatment listed in the PubMed.gov database were searched, relevant articles were then reviewed and used as sources of information for this review.

What the reader will gain—This review attempts for the reader to highlight some current thought regarding mechanisms of SSc pathogenesis and how autoimmunity relates to vascular changes and fibrogenesis of the disease plus provide a review of results of completed clinical trials and current on-going clinical trials that address organ specific or global therapies for this disease which can aid physicians who provide medical care for patients with SSc.

Take home message—SSc is a complex autoimmune disease, the pathogenesis of which although not completely understood is under active study, and new insights into pathogenesis are continuously being discovered. Although there is no effective disease modifying treatment for patients with SSc, quality of life, morbidity and mortality can be improved by using targeted therapy directed at affecting the consequences of damage to lungs, blood vessels, kidneys and the gastrointestinal tract. Innovative approaches to treating SSc are under intense investigation.

Keywords

Systemic sclerosis; Autoimmunity; Fibrosis; Scleroderma; Platelets

1. Pathogenesis of Systemic Sclerosis

Systemic sclerosis (SSc) can be viewed as an uncommon autoimmune disease that has fibroproliferative vasculopathy, ongoing platelet activation and selective organ fibrosis as cardinal consequences of the autoimmune state. Current data suggest genetic influences and

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Declaration of interest

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exogenous triggers permit development of the disease. A major role of autoimmunity in SSc is supported by a multitude of *in vitro* and *in vivo* evidence demonstrating the capability of immune cell products and/or subsets of immune cells to mediate fibrosis and the vasculopathy characteristic of SSc (See Figure 1). For example, endothelial cell apoptosis is induced via the Fas pathway in human dermal microvascular endothelial cells by SSc natural killer (NK) cells in the presence of IL-2, and SSc patients sera contain anti-endothelial cell antibodies (1). Vdelta 1⁺/gamma/delta T cells are increased in lesional fibrotic skin, especially in early SSc and in perivascular distribution where they express HLA-DR and very late activation antigen alpha 4 (CD49d). This suggests Vdelta 1⁺ T cells home to SSc lesional skin and are expanded (2). Immune induction of fibrosis in SSc is further supported by animal models of chronic graft versus host (cGVH) disease and human cGVH disease, both of which are T cell mediated and share some features of SSc. Also, there is reversal or stabilization of SSc fibrosis and SSc vasculopathy in patients undergoing immune ablation followed by immune reconstitution with autologous CD34⁺ stem cells (3). In aggregate, there is strong evidence for an “immunocentric” mediation of the fibrogenic processes of SSc.

1.1 Role of TGF- β , IL-4 and other cytokines in medicating fibrosis in systemic sclerosis

Since its original description as a modulator of fibrosis (4), TGF- β 1 has been one of the most studied fibrogenic factors in SSc and murine models of SSc, and it is thought to play a major role in mediating the SSc fibrogenic phenotype. Studies of gene expression using DNA arrays employing skin biopsies directly or primary cultures of fibroblast derived from explants of skin from lesional and/or non-lesional skin of patients with SSc have identified differences in gene expression from similar control samples from healthy volunteers (5). One study concluded there was a TGF- β signature in a subset of dcSSc termed “diffuse proliferative” (6). None of these gene profiling studies have included disease controls of (e.g. biopsies or fibroblast cultures derived from patients with autoimmune-mediated skin disease such as systemic lupus erythematosus (SLE) or psoriasis). This is an important omission given that earlier published studies that focused on detecting TGF- β 1 and TGF- β 2 in the lesional skin of patients with SSc reached different conclusions as to its presence and specificity for fibrosis. Grushwitz et al. assessed both TGF- β 1 and TGF- β 2 mRNA by *in situ* hybridization and protein by immunohistochemistry and found TGF- β 1/ β 2 mRNA and protein were expressed in dermal and subcutaneous infiltrating cells in early and late SSc but also in other inflammatory skin diseases (7). Gabrielli found similar expression of TGF- β in endothelial cells and dermal fibroblasts in patients with SSc as in those with primary Raynaud’s without SSc (8). Patients with SLE had staining for extracellular TGF- β in all dermal layers (8). Sfikakis et al. found disposition of TGF- β 2 but not TGF- β 1 or TGF- β 3 in the extracellular space in lesional skin of patients with SSc but not in non-lesional skin or normal controls, but SLE or other inflammatory diseases were not evaluated (9).

Studies that have measured levels of TGF- β (total or active) in sera or plasma of patients with SSc have yielded divergent results (reviewed in reference (10)). Surprisingly, in a carefully performed study of 27 patients with dcSSc, 20 patients with lcSSc and 22 healthy controls (HC) there were no differences between total serum TGF- β 1 levels amongst the groups, but patients with dcSSc had significantly lower serum levels of active TGF- β 1 than lcSSc patients or HC, and in dcSSc patients serum active TGF- β 1 correlated negatively with skin score and positively with disease duration (11). The authors speculate that the low levels of active TGF- β 1 in dcSSc patients may result from its sequestration in involved SSc skin (11). Specifically targeting TGF- β 1 has thus far not proven to ameliorate SSc fibrosis as revealed in a multicenter, international, randomized, placebo-controlled Phase I/II trial in early dcSSc of three different doses of a recombinant neutralizing monoclonal antibody against TGF- β 1 (called CAT-192) given every 6weeks $\times$ 4 by intravenous infusion which did not effect significant changes in

MRSS, scleroderma HAQ, pulmonary function or other parameters after six months enrollment in the trial (12).

There are other cytokines/growth factors/chemokines that play a role in fibrogenesis in SSc as well as in animal models of the disease. These include interleukin (IL)-4, IL-6, oncostatin M, trypase, platelet-derived growth factor (PDGF), IL-1, IL-17, IL-5 and monocyte chemotactic protein (MCP)-1. IL-4 induces synthesis of types I and III collagen (CI, CIII) and fibronectin (10) and it is a potent chemoattractant for fibroblasts (13). Numerous studies show that IL-4 is expressed in patients with SSc, especially by CD8⁺ and CD4⁺ CD8⁺ T cells that preferentially infiltrate SSc lesional skin (14–16). Although the specificity of fetal-maternal microchimerism in patients with SSc is a debated topic, one study showed that male-offspring T cells that are present in the blood and skin of women with SSc compared with those from normal women have a T helper (Th)2-oriented cytokine profile with production of high levels of IL-4 (17). The connective tissue matrix in lesional skin of patients with SSc contains several “IL-4 footprints” that result from IL-4’s unique effects on matrix synthesis by fibroblasts. For example, tenascin is a large extracellular matrix glycoprotein present in lesional skin of SSc patients and it is strongly induced in cultured human dermal fibroblasts by IL-4 but not by PDGF, basic fibroblast growth factor (bFGF), TGF- β , IL-1, IL-6, interferon (IFN) γ or tumor necrosis factor (TNF) α (18). The second IL-4 footprint is hydroxylysine aldehyde derived cross-links which are abundant in SSc fibrotic tissue and which are formed by lysyl hydroxylase 2 (LH2) from fibroblasts (19). IL-4, but not IL-1 α/β or TNF α , increases synthesis of LH2 in fibroblasts grown under hypoxic conditions (19). IL-4 stimulates collagen synthesis in fibroblasts by several mechanisms including utilization of the Janus tyrosine kinase (JAK)-1, JAK-2 signaling pathways and by induction of TGF- β 1 and connective tissue growth factor (CTGF) synthesis (20–22). Skin fibrosis in the tight skin (TSK)-1 mouse model of scleroderma is inhibited by administering anti-IL-4 antibodies or deleting the IL-4 gene (23,24).

Several autoantibodies to nonnuclear antigen have been detected in SSc sera that may play a role in mediating fibrosis. Antibodies to PDGF receptor have been described in sera of SSc patients and normal controls but their role in mediating fibrosis is unclear (25,26). Antibodies to fibrillin-1 from SSc sera activate fibroblasts to synthesize collagen and other matrix components and antibodies to metalloproteinases 1 and 3 in SSc sera prevent degradation of collagen and other extracellular matrix components (Reviewed in reference (27)). A pilot study using rituximab to deplete B cells in patients with dcSSc did not show efficacy in improving the MRSS (28).

1.2 Myofibroblasts in systemic sclerosis

Myofibroblasts which are characterized by expression of α smooth muscle actin, ability to contract granulation tissue in vivo and collagen gels in vitro, and expression of the ED-A splice variant of fibronectin are found in biopsies of lesional skin of patients with dcSSc in increased numbers but not in non-lesional skin or atrophic stage of dcSSc or in skin biopsies from healthy controls (29). When cultured from explants or induced with various agents (See Table I), myofibroblasts produce increased amounts of collagen and other matrix components; however, interestingly, in lesional skin of dcSSc patients, myofibroblasts did not stain for lysyloxidase, an enzyme essential for collagen synthesis (29,30). Myofibroblasts arise from many different precursor cells which could contribute to the myofibroblast population in fibrotic lesions of patients with SSc, and they are induced by different agents (See Table I).

1.3 Vasculopathy of systemic sclerosis

Frequently, the earliest symptoms experienced by patients with SSc is Raynaud’s phenomenon (RP) produced by disturbance in neural control of the peripheral vasculature (see below). There is also injury to the endothelial cells (EC) that at present is not well understood that may involve

anti-endothelial cell antibodies and oxidative stress leading to apoptosis (31,32). Von Willebrand factor is released from damaged EC in SSc patients and contributes to platelet adhesion and aggregation (32). Platelets then release pro- and anti-angiogenic growth factors such as VEGF, thrombospondin, cytokines and chemokines (32,33). Repair of the damaged EC and vasculogenesis from circulating endothelial progenitors are impaired for unknown reasons. Endothelin-1 from EC is likely a major mediator of matrix synthesis and vascular remodeling and vasoconstriction in SSc patients.

1.4 Role of platelets in systemic sclerosis

Platelets play a major role in injury and repair of normal wounds not only by promoting homeostasis but also by releasing vasoconstrictor mediators such as endothelin (ET)-1, and chemokines/cytokines/growth factors such as interleukin (IL)-1 β , PDGF-A, -B, -C, -D, transforming growth factor (TGF)- β 1 and β 2, epidermal growth factor (EGF), vascular endothelial growth factor (VEGF)-A, and -C, brain derived neurotrophic factor (BDNF), insulin-like growth factor (IGF)-1, bFGF, and CTGF which promote formation of vascularized granulation tissue and fibrous scar formation [reviewed in (33)]. Numerous studies have attested to the fact that there is ongoing platelet activation and aggregation with release of platelet contents into the circulation in patients with SSc [reviewed in (33)]. Platelet-derived microparticles are also higher in plasma of SSc patients with interstitial lung disease (ILD) (34). Platelets from patients with SSc exhibit enhanced aggregability to CI, adenosine diphosphate, 5-hydroxy tryptamine and adrenalin (33,35) and have a particular phenotype of overexpression 65 kDa CI receptor, reduced nitric oxide synthetase activity, increased phosphatidylinositol-3 (PI-3k) kinase activity, and an increase in protein kinase B (Akt) activity [reviewed in (33)]. Of interest is the observation that lymphokine-rich supernatants from CI-stimulated SSc peripheral blood mononuclear cell cultures, as well as IL-1 β and IFN γ present in these supernatants, induce a similar phenotype when added to cultures of human megakaryocytes, suggesting this SSc platelet phenotype might be induced by cytokines from autoimmune reactions (36). This is an appealing explanation for persistent fibrosis in the setting of a paucity of inflammatory and immune cell infiltrates in later stages of SSc.

1.5 Role of lysophosphatidic acid in systemic sclerosis

A recent study showed that lysophosphatidic acid (LPA) which is secreted from platelets, mast cells, macrophages and dendritic cells is higher in sera of patients with SSc than healthy controls (37). LPA can stimulate lung fibroblasts to myofibroblast differentiation and induce Rho-associated kinases (ROCK), which causes myofibroblast differentiation from SSc dermal fibroblasts and stimulate synthesis of extracellular matrix (38). These and additional biological properties with relevance to SSc pathogenesis are summarized in Figure 2. LPA might well be an heretofore significant and unrecognized player in SSc.

1.6 Role of mast cells in systemic sclerosis

Mast cells are increased in the skin of patients with SSc, especially in the early edematous phase of the disease, and the number of degranulated mast cells is higher in SSc lesional skin and in bronchoalveolar lavage fluid of patients with SSc who have alveolitis (39). Mast cells produce a wide array of mediators including IL-4, heparin, VEGF, bFGF, TNF α , IL-8, platelet activating factor (PAF), TGF- β , leukotriene (LT)B $_4$, tryptase, chymase, IL-6, histamine, stem cell factor (SCF), prostaglandin and LTA. Stem cell factor promotes the viability of mast cells and is increased in association with mast cells in lesional skin of patients with SSc (40). Tryptase secreted by mast cells stimulates fibroblast growth and collagen synthesis, and both tryptase and histamine from mast cells induce differentiation of fibroblasts to a myofibroblast phenotype (41). Activated T cells can stimulate mast cells to produce the fibrogenic factor, oncostatin M, as well as IL-8 (42,43). Mast cells via release of histamine modulate dendritic

cells to effect antigen-specific Th2 cytokine (IL-4) producing T cells (44). Evidence in mice suggest that mast cells by expressing major histocompatibility (MHC) class II molecules regulate T cell responses and preferentially activate T regs (45).

2.0 Clinical Characteristics of Systemic Sclerosis

2.1 SSc Subsets

There are several distinct subsets of SSc. The two most prevalent subsets are termed limited cutaneous (lc) SSc and diffuse cutaneous (dc) SSc. Patients with lcSSc have skin fibrosis distal to the elbow and distal to the knees with sclerosis of skin over the face, neck and upper anterior chest, have less internal organ fibrosis but are at risk for developing pulmonary arterial hypertension (PAH). Patients with dcSSc have more widespread skin fibrosis with involvement of skin over the proximal and distal portions of the extremities, the trunk, face and chest and have more extensive internal organ fibrosis, hypertensive renal crisis and higher mortality than patients with lcSSc. A less common variant termed “SSc *sine* scleroderma” has absence of skin fibrosis but involvement of internal organs with fibrosis. Undifferentiated connective tissue disease, overlap syndromes and mixed connective tissue disease often have some clinical manifestations of SSc.

2.2 Musculoskeletal Involvement in SSc

The musculoskeletal system is often involved in patients with SSc and is a major cause of morbidity and disability. This has recently been reviewed in detail (46) and major features are summarized in Table II.

3. Treatment of Systemic Sclerosis

There is no approved “disease modifying” drug for SSc. Disease modifying anti-rheumatic drugs (DMARDs) used in other rheumatic diseases including d-penicillamine, methotrexate, azathioprine, CP, mycophenolate mofetil (MMP), chlorambucil, and corticosteroids have not found widespread acceptance in treating the global manifestations of SSc. The uncommon prevalence of SSc (variously reported to be from 13.8 to 44.3 cases per 100,000) necessitates use of multicenter clinical trials. The Modified Rodnan Skin Score (MRSS) is most often used in clinical trials as an objective primary outcome variable to assess drugs that target skin fibrosis and has been shown to correlate with biopsy thickness of skin in patients with SSc (47) as well as with dermal myofibroblast content (48).

Having no definitive disease modifying treatment that addresses the basic pathogenesis of SSc, clinicians have focused on treating organ specific manifestations of SSc. This approach has lead to notable reduction in morbidity and death in patients with SSc from the two main causes of mortality, renal failure from scleroderma renal crisis (SRC) and cardiopulmonary failure from pulmonary arterial hypertension. In this review, we will highlight those treatments that have demonstrated benefit from an organ specific perspective (See Table III).

3.1 Raynaud’s Phenomenon and Digital Ulcers

Raynaud’s phenomenon which is very prevalent in patients with SSc is caused by vasospasm of small and medium sized arteries in the distal upper and lower extremities and can be precipitated by cold exposure or by emotional stress or by no recognizable triggers. Functionally, RP arises from an unbalance between vasoconstrictive and vasodilatory stimuli in involved blood vessels, and in SSc there are accompanying structural alterations of the involved blood vessels that compound the severity of RP. Although the etiology of RP is unclear, it is thought to arise from dysregulated interaction between endothelial cells, soluble mediators and neuronal stimulation. Digital ulcers (DU) occur in 30 – 50% of patients with

RP associated with SSc. The treatment of DU is compounded by associated structural narrowing of digital arteries from the vasculopathy of the disease. It is important to treat any secondary bacterial infection in the DU. Analgesics are used to manage intractable pain due to the ischemia and ulceration, and unless contraindicated anti-platelet therapy should be started. Adjunctive therapy such as warm clothing, gloves and paraffin hard baths are helpful.

Some patients respond to calcium channel blockers (CCBs), and they are usually tried, but some patients experience clinically significant hypotension or other side effects. A meta-analysis of six placebo controlled trials of CCBs in RP secondary to SSc showed a moderate reduction of 8.3 attacks over two weeks and 35% less severity (49). The healing of DU was improved from baseline by CCBs in one study, but this was not in a placebo controlled trial (50).

In placebo controlled multicenter trials, bosentan, a dual ET receptor antagonist, showed efficacy in reducing the number of new DUs but not in healing of established DUs (51,52). Although, in a mechanistic study, bosentan taken for 16 weeks in 15 patients with lcSSc did not effect improvement in microvascular structure and function (i.e. no changes in vasodilator response to acetylcholine and sodium nitroprusside, capillary permeability or density) (53). In another similar study, brachial artery ultrasound-derived flow-mediated dilatation which was decreased at baseline, significantly increased after 4 weeks of bosentan treatment in patients with SSc who had PAH and/or DU(9).

Sildenafil and related phosphodiesterase type 5 (PDE-5) inhibitors have shown benefit in reducing RP (54,55). These agents produce vasodilatation by promoting accumulation of cyclic guanosine monophosphate (cGMP) in vascular smooth muscle cells. Their efficacy in preventing DUs or promoting healing of DUs has not been adequately assessed. Statins (HMG-CoA reductase inhibitors) hold promise in treatment of RP primarily through their effect to inhibit endothelial cell activation. Atorvastatin 40 mg/day when compared to placebo effected a 50% relative risk reduction in development of new DUs (56). Both simvastatin and pravastatin improved parameters of endothelial activation in a small series of SSc patients with RP (57, 58).

Two placebo-controlled trials, one in the UK and one in the USA, of oral iloprost at 50 µg or 100 µg twice daily for 6 weeks with a 6 week post-treatment follow-up visit in patients with RP secondary to SSc were performed (59,60). The UK trial with a total of 103 patients showed in the 50 µg dose group at 6 weeks significant reduction in total daily duration of RP attacks but not in frequency or severity of attacks between the oral iloprost and placebo treated groups (59). In the larger USA trial, the oral iloprost treated patients had outcomes similar to placebo treated patients (60). In a multicenter placebo controlled study in which iloprost was administered daily via intravenous infusion over 6h at 0.5 to 2 ng/kg per minute on five consecutive days, there were significant improvements in 1) mean weekly number of Raynaud's attacks (39.1% decrease in iloprost vs 22.2% decrease in placebo), 2) in global Raynaud's severity score during the nine week follow-up period (34.8% iloprost vs 19.7% placebo), 3) physician's global assessment 54.4% iloprost vs 27.4% placebo), and at week three post treatment 14.6% more iloprost treated patients experienced a 50% or greater healing of DU than placebo treated patients (61).

3.2 Scleroderma Renal Crisis

Renal involvement with SRC is a unique syndrome and complication of SSc that occurs in 5–10% of patients (62). Patients more commonly have dcSSc with MRSS equal to or greater than 20, cardiac enlargement, contractures of large joints, prior use of prednisone and have anti-topoisomerase I and anti-RNA polymerase III antibodies (63). Associated clinical features include new onset systemic hypertension (although SRC can rarely occur in normotensive SSc

patients and in SSc patients on angiotensin converting enzyme (ACE) inhibitors for hypertension) increased plasma creatinine, microangiopathic hemolytic anemia, hyperreninemia, thrombocytopenia, proteinuria, hematuria and granular casts in the urinary sediment (62). The etiology and pathogenesis of SRC are unclear, but there are high plasma renin levels, and renal histology reveals accumulation in the arcuate and interlobular arteries of mucin deposits, mucoid intimal thickening in arterioles with fibronoid necrosis and fibrosis and platelet thrombi. After hypertension is established, there are changes compatible with hypertensive vascular damage. Increased renin production results from decreased renal perfusion with associated hyperplasia of the juxtaglomerular apparatus (62). Unfortunately, SRC can be the presenting sign of SSc. It is essential that patients with SSc have frequent monitoring of blood pressure. Other anti-hypertensive agents such as CCBs can be added to the ACE inhibitor if hypertension is not controlled. Additional complications such as pulmonary edema can be treated with beta adrenergic blockers and nitrate infusion, severe thrombotic microangiopathy with plasma exchange and renal failure with intermittent hemodialysis. ACE inhibitors should be continued even in the presence of renal failure and renal dialysis therapy, since renal function recovers in about 1/3 of patients initially requiring dialysis. However, over half of the patients may require chronic dialysis or renal transplant if dialysis is needed for longer than one year (64). Markedly elevated blood pressure must be reduced slowly to avoid inducing acute tubular necrosis and renal shutdown.

3.3. Gastrointestinal Manifestations of SSc

All portions of the gastrointestinal (GI) tract can be affected by SSc, often causing significant morbidity and symptoms (up to 90% of SSc patients) and even mortality (6–12%) (65). The general functions of the various components of the GI tract are often affected by SSc, leading to impairment of motility, digestion, absorption and excretion. Although smooth muscle atrophy and fibrosis occur later, often there is a paucity of morphologic changes. This raises the question whether circulating functional autoantibodies such as antimyenteric neuronal antibodies and/or anti-muscarinic receptor 3 antibodies might be contributing to GI dysmotility in SSc (66,67). Difficulty swallowing (resulting from dysmotility of the lower 2/3 of the esophagus and decreased lower esophageal sphincter tone) is often an early complaint of patients with SSc before other GI tract symptoms appear (65). Omeprazole twice daily is routinely used to treat esophageal dysmotility symptoms in patients with SSc as well as other proton pump inhibitors (65). Gastroesophageal reflux is often present with esophageal dysmotility which can manifest as peptic esophagitis, erosive esophagitis, bleeding or esophageal ulceration that can lead to stricture, fistulae formation and an achalasia-like syndrome that may contribute to risk of developing Barrett's esophagus (65).

Barrett's esophagus occurs in 6.8–12.7% of SSc patients (68) and can progress to cancer (Barrett's cancer), but whether SSc patients are at greater risk of development of Barrett's cancer is unresolved (65). SSc may be at greater risk of developing esophageal cancer in general as well as oropharyngeal cancer compared to the general population (69). Regular surveillance endoscopies should be performed at intervals dictated by the stage of Barrett's esophagus (65).

The most common gastric abnormality in patients with SSc is delayed gastric emptying secondary to electrophysiological dysfunction which then leads to food intolerance and reduced gastro-esophageal emptying (65). Pro-motility drugs like metoclopramide and domperidone can be employed to increase tone and amplitude of gastric contractions plus relax the pyloric sphincter. Low-dose erythromycin which has motilin-like properties can be used in prokinetic-resistant patients (65,70). Patients whose gastric motility impairment can not be managed by pharmacotherapy may require additional interventions such as gastrostomy tube placement for intermittent drainage and artificial nutritional support (65). Another serious gastric

complication of SSc of unknown etiology is gastric antral vascular ectasia (GAVE), also known as “watermelon stomach”, which derives from the appearance at endoscopy of multiple, parallel longitudinal columns of red vessels within the antrum that radiate to the pylorus that have the appearance of watermelon stripes (65). Endoscopic laser ablation (or safer argon plasma coagulation) is often effective in treating the bleeding from GAVE (71). Refractory patients may need antrectomy. The blood loss is treated with blood transfusion and an oral iron supplement (65).

Involvement of the small intestine has a spectrum of severity ranging from blind loops that foster bacteria overgrowth, malabsorption, pseudo-obstruction from dysmotility, and at the extreme marked intestinal failure requiring intravenous nutrition. It is desirable to evaluate small bowel function to document malabsorption and the nature of dysmotility (with small bowel manometry) to select patients who may respond to octreotide (72). Bacteria overgrowth is treated with antibiotics which are usually only temporarily effective.

Like other portions of the GI tract, the colon usually exhibits some degree of reduced motility. The gastrocolic response is often absent. In early disease motility can be restored with neostigmine or metoclopramide, implicating a neural defect (73,74). Barium enemas reveal morphologic radiologic changes in up to 50% of patients with SSc (75) which include wide-mouth diverticuli, increased luminal fluid, postevacuation residual, dilatation and loss of haustrations (74). These morphologic changes do not always correlate with symptoms. Functional studies show delayed whole gut transit time, which correlates with constipation, in 23% of patients with SSc (76,77). A complication of reduced intestinal motility and propulsion is pseudo-obstruction. Clinically, this resembles clinically mechanical bowel obstruction and is accompanied by diarrhea or constipation which are, respectively, related to predominance of bacterial overgrowth in the small bowel or to colonic inertia (74). Pseudo-obstruction is treated first by decompression via nasogastric tube, and by keeping the patient NPO. Prokinetic drugs such as erythromycin, cisapride, prucalopride or octreotide are useful to improve bowel colonic motility acutely (74). Sustained increase in bowel motility has been effected by combining erythromycin with octreotide (less than 100 µg octreotide at night so effect of eating on motor and pancreatic function will not be affected) (74). It is important to treat chronic constipation and pseudo-obstruction to avoid complications such as volvulus or stercoral ulcers (which can perforate) (78,79).

Pneumatosis cystoides intestinalis (PCI), also called “pneumatosis coli” is caused by leakage of air from the bowel lumen into the intestinal wall due to defects in the mucosa resulting from mucosal atrophy, ischemic damage or bacterial overgrowth (80). PCI can be treated with conservative measures including oxygen and antibiotics (74). Occasionally, pneumoperitoneum occurs as a result of perforations in diverticuli or ruptured cysts of PCI and can persist without peritonitis, in which case it should be managed conservatively without surgery (81).

Fecal incontinence is common in patients with SSc, occurring in up to 38% (82). This is due to abnormalities in the internal anal sphincter from disrupted neurogenic and myogenic components that progress to thinning of the muscularis propria due to atrophy and later by fibrosis of circular and longitudinal muscles accompanied by ischemia from sclerosis of small arteries and arterioles (74).

Rectal prolapse in patients with SSc is associated with impairment of the rectoanal inhibitory reflex (83). Fecal incontinence can be treated by solidifying liquid stools, biofeedback or sacral nerve stimulation (84). Rectal prolapse can be managed by surgery.

3.4 Cardiac Complications of SSc

Cardiac involvement is common in SSc and can be primary, which is the direct consequence of SSc pathogenesis directed toward cardiac structures or secondary, which results from coexisting systemic hypertension or pulmonary arterial hypertension (PAH). Delayed enhanced magnetic resonance imaging performed on 41 consecutive patients with SSc, showed changes in 66% compatible with myocardial fibrosis. The distribution of fibrosis was predominantly in the left ventricular midwall involving basal and midcavity segments (85). There was no difference in fibrosis in lcSSc vs dcSSc, but patients with RP ≥ 15 years duration and patients with abnormal Holter monitoring had a greater number of enhancing segments (85). There is no association of myocardial fibrosis with vascular areas of the epicardial coronary arteries, and there is sparing of fibrosis in the subendocardial layers of the myocardium, suggesting that the fibrosis is not due to spasm of small coronary arteries and compromised cardiac perfusion (85). Autopsy data show that myocardial fibrosis is present in 50–80% of patients with SSc (86). Primary cardiac involvement in SSc is usually asymptomatic. Myocardial fibrosis is associated with anti-topoisomerase antibody and rapid or intermediate progressive increase in MRSS (87) and is responsible for most of the heart related mortality in SSc, causing conductive system abnormalities, arrhythmias, heart failure and sudden death (85). An implantable cardioverter defibrillator device may be needed to protect against ventricular tachycardia or fibrillation.

Functional abnormalities of the heart in otherwise asymptomatic patients with no PAH have been detected with newer methods. Pulsed tissue-Doppler echocardiogram has revealed patients with SSc have a wider mean left atrial diameter, reduced left ventricular relaxation, higher pulmonary artery pressure, reduced right ventricular contractility and a trend toward reduced left ventricular ejection fraction compared to matched healthy controls (88). Of potential relevance, is the finding that myocardial contractility is reduced by LPA via an anti-adrenergic mechanism that involves $G\alpha_i$ protein and signaling through LPA_1 and LPA_3 receptors (89). As noted above, LPA is elevated in sera of patients with SSc (37) (Figure 2).

Cardiac vessels, like those in peripheral sites have abnormal vasoreactivity, arterial concentric intimal hypertrophy in patients with SSc, and myocardial infarction occurs in SSc patients with patent epicardial coronary arteries (90). Myocardial perfusion can be assessed via use of cardiovascular MRI and is useful in detecting and monitoring sub-endocardial defects, fibrotic myocardium and myocarditis associated with polymyositis (PM) overlap (91). These imaging techniques can guide selection of therapies to treat myositis (immunosuppressive drugs and high dose corticosteroids) and vascular ischemia (CCBs and ACE inhibitors). Myocarditis is uncommon in patients with SSc, and other causes such as viral myocarditis should be excluded. Sclerodermatous myocarditis maybe confirmed by endocardial biopsy and requires treatment with immunosuppressive agents (92). Autologous stem cell transplant induced resolution of sclerodermatous myocarditis in one reported case (93).

Pericardial effusion has been noted in 33–72% of autopsied patients with SSc and, although present, is usually asymptomatic, however, large pericardial effusions causing cardiac tamponade are usually a predictor of poor clinical outcome (94). Pericardial effusions are often present in SSc patients with PAH or congestive heart failure (95). The type of treatment for pericardial effusions depends on the condition with which it is associated. A symptomatic inflammatory pericardial fluid not due to viral, bacteria, mycobacterial or fungal etiology can be treated with immunosuppressive drugs such as azathioprine and prednisone (94). Persistent hemodynamically significant pericardial effusions require surgical drainage.

Although atherosclerotic coronary artery disease does not show increased prevalence, there is a greater likelihood of coronary vasospasm in SSc patients with coronary disease (94). Response of abnormalities in cardiac perfusion to CCBs, ACE inhibitors or intravenous

dipyridamole as assessed by Thallium-201 single proton emission computed tomography or MRI can be used to select which will be useful in treating a given SSc patient to improve myocardial perfusion (96).

3.5 Interstitial Lung Disease (Pulmonary Fibrosis)

The lung is a major target organ for development of fibrosis, being found at autopsy in most patients with SSc (86). Mortality in patients with SSc attributable to interstitial lung disease (ILD) is about 16% (97). High resolution computed tomography (HRCT) has virtually replaced lung biopsy for establishing the presence of ILD in patients with SSc. In patients with SSc, the ILD more often has changes on HRCT that resemble patients with idiopathic nonspecific interstitial pneumonia (i.e. “ground glass” appearance) rather than that seen in patients with idiopathic pulmonary fibrosis (98).

Therapeutic options are limited in treatment of ILD in SSc patients since strong immunosuppression for 1 year with daily CP is the only regimen shown in a multicenter large placebo-controlled trial to date to stabilize SSc ILD (99). Unfortunately in this same study, after discontinuing CP, lung function was stable for six additional months but then deteriorated (99,100). There is interest in using agents such as MMF (inhibitor of purine synthesis) or imatinib (tyrosine kinase inhibitor) alone or in combination with CP, but results of large controlled trials with these agents have yet to be reported.

3.6 Pulmonary Arterial Hypertension

Elevated mean pulmonary artery pressure (PAP) ≥ 25 mmHg in patients with SSc can develop from 1 or more of 4 causes. These are 1) primary pulmonary arterial hypertension (PAH), believed to arise from excessive vasoconstrictive stimuli such as from thromboxane A₂ (from perturbed endothelial cells or aggregated/activated platelets) and ET-1 and reduced generation of the main vasodilators, nitric oxide and prostacyclin synthase which converts arachidonic acid to prostacyclin; 2) elevated pulmonary venous pressure which can be caused by systolic and/or diastolic dysfunction or valvular disease; 3) chronic hypoxia from intrinsic lung disease (e.g. ILD); and 4) chronic thromboembolic pulmonary hypertension. It is important to establish which or if one or more of these 4 causes of pulmonary hypertension are present in a given SSc patient. The diagnosis of PAH requires in addition to elevated mean PAP of ≥ 25 mmHg at rest or ≥ 30 mm Hg with exercise, a pulmonary capillary wedge pressure of < 15 mm Hg (101). If untreated or uncontrolled by therapeutic regimens, PAH carries a high mortality from progressive intractable right ventricular overload and failure. Dyspnea on exertion in a patient with SSc should definitely trigger assessments including Doppler echocardiogram (ECHO) that includes assessment of velocity of tricuspid regurgitation and pulmonary function tests (PFTs) with carbon monoxide diffusion capacity (DLCO). There are difficulties in diagnosing PAH. Asymptomatic patients without dyspnea should have yearly ECHO since PAH can be asymptomatic in its early stages. In SSc patients with dyspnea in which PFTs reveal a forced vital capacity (FVC)/DLCO ratio of ≥ 1.6 , PAH should be suspected and confirmed with ECHO. The ECHO can give false positive and false negative results with regard to presence of PAH. To confirm PAH, right heart catheterization should be performed if ECHO and/or DLCO suggest PAH. Although in need of further study, pro- β -natriuretic peptide may be a useful biomarker to follow response to treatment of PAH.

Several vasodilators such as nitrates and CCBs should be given a trial in SSc patients with PAH before going to more targeted therapeutics. Target therapies address the 3 basic pathogenic pathways largely responsible for the condition. To target deficiency in the prostacyclin pathway, prostacyclin analogs have been used (epoprostenol, treprostinil and iloprost). Epoprostenol delivered continuously intravenously via pump and treprostinil delivered subcutaneously improve exercise capacity and hemodynamics in patients with

idiopathic PAH and PAH in patients with connective tissue diseases, including SSc (101–104). Inhaled iloprost improved exercise capacity in patients with idiopathic or connective tissue diseases related PAH (105).

The contribution of excessive ET-1 production in PAH has been targeted by development of ETB and/ or ETA receptor antagonists. These agents, (bosentan which blocks ETA and ETB, and sitaxsentan and ambrisentan which block the ETA receptor) have the convenience of being able to be administered orally. Bosentan has been shown to improve exercise capacity, lengthen the time to clinical worsening and survival in patients with idiopathic or connective tissue diseases (including SSc) related PAH (106). Sitaxsentan has shown improvement in exercise capacity in patients with idiopathic and connective tissue diseases related PAH (107). Ambrisentan in patients with PAH significantly improved exercise capacity and lengthened the time to clinical worsening (108).

Diminished nitric oxide levels in PAH has been addressed by using PDE-5 inhibitors such as sildenafil which inhibit degradation of cGMP, thereby providing more cGMP for nitric oxide mediated vasodilatation. Sildenafil is approved at a dose of 20 mg three times daily for treating PAH and has shown efficacy in improving exercise capacity, hemodynamics, and New York Heart Association functional class in patients with either idiopathic or connective tissue diseases associated PAH (109). Recently, similar results were observed with another PDE-5 inhibitor, tadalafil (110).

Since the 3 pathways discussed above may be involved in PAH in a given patient, combination therapy using prostacyclin analogs, PDE-5 inhibitors, and/or ET-1 receptor blockers are an option to consider when monotherapy is not giving the desired improvement. Clinical trials of combination therapies are currently being performed. Other agents such as adrenomedullin, vasoactive intestinal peptide, or inhibitors of serotonin transporters or PDGF signaling have potential as additional therapeutics for PAH. Adjunctive therapy includes anticoagulation (unless contraindicated) and continuous oxygen supply for patients with PAH.

4. Novel Therapies and Clinical Trials for Systemic Sclerosis

The literature is filled with anecdotal treatments or small Phase I clinical trials which have to be interpreted with caution since some manifestations such as skin fibrosis and RP naturally wax and wane. Below, we review some ongoing clinical trials in SSc with new therapeutic agents which are registered in the USA (<http://clinicaltrials.gov>).

4.1 Anti-fibrotic and disease arresting therapies

The lack of animal models that recapitulate all pathogenetic features of SSc, has hampered development of effective anti-fibrotic and disease arresting therapeutics. In the two most studied models (Tsk-1 and bleomycin skin or lung fibrosis models), TGF- β 1 and CTGF play major roles in effecting fibrosis. Agents that target these growth factors are very effective in preventing fibrosis in these models. The extrapolation of therapeutics found to be effective in these TGF- β dependent models to human scleroderma may fail, since as illustrated in Figure 1, multiple cytokine/growth factors/chemokines and possibly fibrogenic antibodies are operative in SSc which would not be affected by anti-TGF- β or anti-CTGF based therapies.

Betaglycan peptide—A currently enrolling phase II study of the efficacy and safety of topical application of P144 in the treatment of skin fibrosis in patients with SSc. Peptide 144 (P144) is the acetic salt of a 14mer peptide from human TGF- β 1 type III receptor (betaglycan). P144 TGF- β 1-inhibitor has been specifically designed to block the interaction between TGF- β 1 and TGF- β 1 type III receptor, thus blocking its biological effects. P144 has shown

significant antifibrotic activity in mice receiving repeated subcutaneous injections of bleomycin (111).

Imatinib—Imatinib and related compounds antagonize specific tyrosine kinases that mediate fibrotic pathways involved in the pathogenesis of SSc, including c-Abl, a downstream mediator of transforming growth factor (TGF)-beta, and platelet derived growth factor (PDGF) receptors. There are several ongoing phase II studies with imatinib in SSc (Table IV).

Immunosuppression—A sizeable body of anecdotal and non-controlled study literature points to benefits of broad-spectrum immunosuppression in SSc. Mycophenolate mofetil is an antimetabolite that blocks the rate-limiting enzymatic formation of guanosine nucleotides, specifically it blocks the type II isoform found in activated lymphocytes more potently than the type I isoform found in both T- and B-lymphocytes. In SSc, MMF has been used after anti-thymocyte globulin (ATG) in one small open label study that showed a significant improvement in skin scores. Additional Phase I studies are enrolling patients (Table IV).

Cyclophosphamide pulse therapy is effective in suppressing active alveolitis. A large, multicenter randomized clinical trial comparing MMP to oral CP in scleroderma ILD (Scleroderma Lung Study II) is enrolling patients now (Table II). A current ongoing phase III study of SSc patients randomized to high-dose intravenous immunoglobulin infusion or placebo will be informative (Table IV). A safety and tolerability study (phase II) is also currently enrolling SSc patients to receive a hyperimmune goat serum product (AIMSPRO) (Table IV). Methotrexate was shown to effect statistically-significant improvement in skin scores of two small independent placebo-controlled trials (112), but the clinical significance of the improvement is less certain (113). Methotrexate is being compared to rapamycin in a phase I/II clinical trial (Table IV).

Autologous stem cell transplant—With careful selection of patients healthy enough to withstand the preparative regimen, the overall treatment-related mortality of autologous stem cell transplant (SCT) has improved greatly. Based on the encouraging results of a phase II pilot study (3), the scleroderma cyclophosphamide or transplantation (SCOT) trial has ongoing enrollment for patients with dcSSc. The SCOT trial is comparing two ways of treating severe SSc: 1) high-dose immunosuppressive therapy (total body irradiation, CP (120 mg/kg), and equine ATG followed by autologous CD34⁺ SCT and 2) high-dose pulse IV CP. An ongoing phase II randomized study is comparing pulse CP to high dose CP and rATG rescued with autologous peripheral blood SCT (Table IV). Milder immunosuppression is being assessed in study participants that are not healthy enough to undergo SCT in another currently enrolling phase II study of the toxicity of CP and rATG as salvage therapy in patients with dcSSc (Table IV). There are multiple phase I and II studies of various intensity preparations; the largest is an attempt to further characterize the effects of GVHD in non-myeloablative preparations and subsequent SCT response followed by allogeneic peripheral blood SCT (maximized GVHD) versus Umbilical Cord Blood Transplant (minimal GVHD) (Table IV).

Other clinical trials—Success in murine models (114) has led to renewed interest in modulating B cell immunity, and there are two small studies using anti-CD20 mAb to deplete circulating B cells in dcSSc (Table II). Levels of the cytokine B-cell activating factor correlate with extent of disease in SSc, and blockade of this cytokine is another potential therapeutic approach (115).

A placebo controlled study with 64 participants receiving ultraviolet (UV)A1 or sham photopheresis had encouraging results, but wasn't powered sufficiently to show statistical benefit over a strong placebo effect (116). A currently enrolling phase I/II trial is using a randomized intra-individual half body irradiation protocol investigating whether treatment

with UVA1 is effective against skin fibrosis in SSc (Table IV). A fully humanized monoclonal antibody directed against subunit 1 of the type 1 interferon receptor (MEDI-546) is being evaluated in several clinical trials (Table IV).

In an attempt to attain specific T cell tolerance, oral CI (CI is an autoantigen in SSc) treatment was studied in a large phase II trial of patients with dcSSc consisting of patients with Early Phase $\leq$ 3 years duration and Late Phase $>$ 3 – 10 years duration. Patients received placebo or 500 μ g bovine CI daily for 12 months and returned at month 15 after 3 months off study drug for assessment. The MRSS did not improve in the total population or in Early Phase patients at months 12 or 15 in study completers. However, at month 15, Late Phase completers treated with oral CI had a statistically greater reduction in MRSS than Late Phase placebo treated patients (117).

Another strategy that merits further investigation is targeted depletion of activated T cells with the anti-CD25 monoclonal antibody, basiliximab (118). And a currently enrolling phase II protocol in dcSSc patients is investigating abatacept, a recombinant fusion protein that blocks T cell activation.

5. Conclusion

The morbidity and mortality of patients with SSc is largely the result of excessive accumulation of extracellular connective tissue matrix in the vasculature and vital organs. A unified hypothesis can attribute these changes to a unique dysregulated autoimmune response that leads to fibrosis. This hypothesis is supported by many *in vivo* and *in vitro* observations. Many different fibrogenic antibodies, cytokines/growth factors/chemokines and LPA elevated in lesional tissue or the circulation in patients with SSc are derived from immune cells and platelets. These are capable of effecting the pathophysiological changes of SSc. Although organ directed therapy has improved survival and quality of life in SSc patients, true disease modifying therapy designed to correct this dysregulated autoimmune response rather than global immunosuppression should be a major focus of future research initiatives.

6. Expert Opinion

Thus far research has allowed identification of patients that fit into subsets of SSc so that by performing laboratory and clinical assessments of the disease physicians know what complications to expect so organ specific treatments can be maximized before irreversible damage occurs. While survival from scleroderma renal crisis and PAH have been greatly improved, there has been thus far little progress in halting or reversing fibrosis of organs and vasculature. We still don't know what triggers the onset of SSc and what degree ongoing autoimmunity is responsible for the vasculopathy and fibrosis.

Recent success in resetting the "immunostat" by use of autologous CD34+ stem cell transplantation, which leads to reversing or stabilizing SSc, plays a significant role for autoimmunity in SSc pathogenesis. There are many molecular fibrogenic agents besides TGF- β that may be targets of therapy in SSc, although such targeting may do little to stop the autoimmune engine driving the disease. Research on SSc is challenging due to the uncommon occurrence of the disease and lack of animal models that recapitulate all aspects of the human disease.

Several ongoing clinical trials are targeting TGF- β and PDGF pathways, and results of these studies will provide insight as to whether this will be a successful strategy or not to pursue in the future. Results of autologous stem cell transplant trials scheduled to be completed in the next few years will likewise reveal whether this is an appropriate approach to arrest and/or reverse SSc disease manifestations. Recent studies underscore the potential of platelets through

release of LPA and other mediators that are capable of effecting many of the recognized perturbations of the immune system, inflammatory cells, endothelium, and vasculature and fibroblasts/myofibroblasts that effect the pathology of SSc. More focus on the role of platelets in SSc may produce new advances and insights into understanding and treating SSc.

Article Highlights

- SSc is a complex, uncommon disease with increased mortality characterized by interrelated autoimmunity, vasculopathy and fibrosis.
- Although TGF- β has a major role as a fibroproliferative agent of SSc, there are other identified cytokines, chemokines and auto-antibodies capable of effecting fibroproliferative changes similar to TGF- β , suggesting mechanisms driving fibrosis are likely redundant in this disease.
- The role of platelets in SSc pathogenesis deserves intense study since recent evidence highlights their potential importance in this disease.
- Scleroderma renal crisis, pulmonary arterial hypertension and to a lesser extent Raynauds' phenomenon and digital ulcers can be managed reasonably well with organ specific pharmaceuticals.
- Major clinical trials are underway to explore effectiveness of targeting TGF- β and PDGF signaling pathways and resetting the immunostat with autologous stem cell transplantation, the results of which may point the way to future management of SSc.

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Figure 1. The Profibrotic, Vasculopathy and Platelet SSc Phenotype Signals from Immune Cells
 On the left-hand side of the figure are listed cells of the immune system with arrows to targets labeled with cytokines and other products from these immune cells that affect endothelium, megakaryocytes and fibroblasts inducing in the SSc phenotype: LPA=lysophosphatidic acid; ADCC=antibody dependent cellular cytotoxicity; AECA=anti-endothelial cell antibody; Anti-PDGFR AB=anti-platelet-derived growth factor antibody; AB=antibody; CTGF=connective tissue growth factor; TGF=transforming growth factor; MCP=monocyte chemotactic protein; IL=interleukin; FGF=fibroblast growth factor; ET=endothelin; IGF=Insulin-like growth factor; VEGF= vascular endothelial growth factor; CI=type I collagen; type III collagen; CVI=type VI collagen; GAGs=glycosaminoglycans; IFN=interferon.

Figure 2. Biological Properties of Lysophosphatidic Acid Relative to SSc Pathogenic Processes: The LPA Footprint?

Lysophosphatidic acid (LPA) is one of several phospholipid growth factors that interact with one or more G protein-coupled seven transmembrane receptors (GPCRs) on many different cell types. GPCRs recognized by LPA are designated LPA₁₋₈. Studies in murine models have shown that LPA mediates a variety of biologic effects via specific receptors. For example, LPA₁ is involved in brain development, neuropathic pain and renal and lung fibrosis, whereas LPA₂ protects the intestine from radiation injury. LPA is normally present in serum/plasma in high amounts (up to 100 μM) where it is generated from lysophospholipids by the plasma

enzyme, autotaxin, but may also be released from unactivated or activated platelets, dendritic cells, mast cells and macrophages.

LPA has many effects on target cells or tissues that are involved in SSc pathogenesis. These include a likely role of LPA to induce hypertension via increasing intracellular Ca^{++} and proliferation of vascular smooth muscle cells (Can J Cardiol 2003, 19:1525–1536). LPA decreases myocardial contractility by an anti-adrenergic effect (J Mol Cell Cardiol 2003; 35:71–80). Platelets release LPA especially when activated, and LPA in turn causes changes in platelet shape, aggregation and formation of platelet-monocyte aggregates (Blood. 2004, 103:2585–2592). $CD4^{+}$ T cells increase IL-13 production when submaximally stimulated in the presence of physiological levels of LPA, and LPA reduces production of IL-2 (FASEB J. 2000, 14:2387–2389; Am J Physiol Lung Cell Mol Physiol. 2006, 290:L66–L74). Migration of T cells is modulated in a concentration dependent manner by LPA (Biochim Biophys Acta, 2002, 1582:161–167). Dendritic cells recognize LPA as a chemoattractant, and their cytokine profile is changed to diminish Th1 and increase Th2 differentiation of T cells (J Immunol, 2002, 169:4129–4135; Immunol, 2008, 125:289–301). B cell growth and immunoglobulin synthesis are increased by LPA (Am J Physiol, 1998, 274:C1573–C1582). The effect of LPA on the endothelium is to increase leakiness, tube formation and lymphocyte adhesion plus upregulation of synthesis of VEGF-C, Prox-1, LYVE-1 and prodopamine (J Cell Biochem, 2006, 99:1216–1232; Cell Mol Life Sci, 2008, 65:2740–2751). Fibroblasts are modulated by LPA in a manner that promotes synthesis of new matrix including reduction in apoptosis, and increases in chemotaxis, collagen synthesis (at 1–10 μ M LPA stimulates cardiac fibroblast type I collagen synthesis), growth, synthesis of CTGF and fibroblast to myofibroblast transition (Biochim Biophys Acta, 2008, 1781:582–587; Biochem J, 200, 352:135–143; J Periodontol, 2004, 75:297–305; FEBS Letters, 2006, 580:4737–4745; AJP-Cell Physiol, 2007, 294:575–82). Natural killer cells chemotax to LPA, and $IFN\gamma$ synthesis is increased by LPA (Eur J Immunol, 2003, 33:2083–2089).

Table 1

Origins of Myofibroblasts *

Cell of Origin	Inducing Agent
Resident Fibroblasts	TGF- β 1 (Potentiated by ET-1 and CTGF) TGF- β 3 GM-CSF IL-6 IL-4 Thrombin Bradykinin Histamine (From Mast Cells) Tryptase (From Mast Cells)
Epithelial Cells	Oncostatin M TGF- β 1
Endothelial Cells	TNF α
Pericytes	TGF- β 1
Circulating Fibrocytes	TGF- β 1 IL-4 IL-13 PDGF-B ET-1

* Resident fibroblasts differentiate into myofibroblasts when grown in the presence of several agents including TGF- β 1, TGF- β 3 (J Pediatr Surg 2000, 35:183–187), IL-4 (Annals Rheum Dis 1997, 56:426–431), GM-CSF (Thromb Haemost 2004, 92:262–274), bradykinin (J. Allergy Clin Immunol 2005, 116:1242–1248), IL-6 (J Invest. Dermatol 2006, 126:561–568), thrombin (Clin Exp Rheumatol 2004, 22 (suppl 33):S38–S46), and histamine and tryptase from mast cells (J Invest Dermatol 2001, 117:1113–1119). Epithelial cells from different tissues transition into myofibroblasts in the presence of TGF- β (Neoplasia 2004, 6:603–610) or oncostatin M (J Am Soc Nephrol 2004, 15:21–32). Microvascular endothelial cells transition into myofibroblasts when exposed to TNF- α or IL-1 β (Medical Hypothesis 2007, 68:650–655). Pericytes differentiate into myofibroblasts when exposed to TGF- β 1 (Arterioscler Thromb Vase Biol 2008, 28:541–547). Circulating fibrocytes differentiate into myofibroblasts when exposed to TGF- β 1, ET-1, IL-4, IL-13 or PDGF-B (Histochem Cell Biol 1998, 109:349–357; J Leukocyte Biol 2009, 86:1111–1118).

Table 2**Musculoskeletal Involvement in SSc***

Site	Manifestations
Joints	Ranges from symmetrical polyarthralgias to polyarthritis (Similar distribution as in rheumatoid arthritis). Radiographic findings include juxta-articular osteoporosis, erosions, joint space narrowing, pencil in cup deformity and/or subchondral sclerosis.
Bone	Distal phalangeal resorption in hands>feet, osteoporosis, resorption of bone (carpal bones, distal radius and ulna, clavicles, ribs, spine, mandible) and thickening of periodontal membrane.
Tendons	Inflammatory proliferative tenosynovitis, tendon rupture. Fibrous deposits within tendon sheaths → tendon friction rubs in wrists, elbows, knees and shoulder are associated with more severe disease. Contracture of flexor tendons.
Muscle	Myopathy is present to some degree in most patients. In dsSSc, proximal muscle weakness in upper and lower extremities can have overlap with polymyositis or have piecemeal infarction from SSc vasculopathy, fibrous myopathy or ill defined myopathies. Skeletal myopathy is often related to myocardial disease in SSc.

* Adapted from reference ³⁶

Table 3

Pharmacotherapy of SSc Disease Manifestations

Disease Manifestation	Pharmacotherapy
Raynaud's Phenomenon	Calcium Channel Blockers, Sildenafil and related phosphodiesterase type-5 inhibitors, intravenous iloprost
Digital Ulcers	Bosentan, statins, intravenous iloprost, aspirin
Scleroderma Renal Crisis	ACE inhibitors plus calcium channel blockers and addition of beta adrenergic blockers or nitrate infusions as warranted to control hypertension
Esophageal Dysmotility	Proton pump inhibitors
Delayed Gastric Emptying	Pro-motility drugs (metoclopramide domperidone, erythromycin)
Small Bowel Dysmotility (Blind Loops)	Octerotide, Antibiotics
Colonic Dysmotility	Erythromycin, prucalopride, octreotide
Coronary Artery Vasospasm	Calcium channel blockers, ACE inhibitors, dipyridamole
Progressive Pulmonary Fibrosis	Cyclophosphamide, ? Mycophenolate mofetil
Pulmonary Arterial Hypertension	Prostacyclin analogs (epoprostenol, treprostinil, iloprost), bosentan, sitaxsentan, ambrisentan, phosphodiesterase type-5 inhibitors
Arthritis	DMARDs (hydroxychloroquine, methotrexate, sulfasalazine)
Osteoporosis	Calcium, Vitamin D, intravenous bisphosphonates

Table 4
Registered Clinical Trials That Are Recruiting and/or On-going Utilizing Patients with SSC*

Drug/Intervention	Clinical Trial Identifier	Country	Enrollment		Study Type	Status
			Number			
Imatinib	NCT00555581	USA	30		Phase IIa (No Placebo)	Recruiting
Imatinib	NCT00506831	USA	15		Phase I/II (No Placebo)	Recruiting
Imatinib	NCT00573326 (For ILD)	Italy	30		Phase II (No Placebo)	Recruiting
Imatinib	NCT00479934	France	34		Phase II (Placebo)	Recruiting
Imatinib	NCT00613171	Multinational	27		Phase II (No Placebo)	Ongoing (Not recruiting)
Imatinib	NCT00512902	USA	20		Open Label Pilot	Recruiting
Dasatinib	NCT00764309 (For ILD)	Multinational	98		Phase II (Placebo)	Recruiting
P144 β -glycan Peptide (Topical)	NCT00781053	Multinational			Open Label Extension of NCT00574613	Recruiting
Mycophenylate Mofetil	NCT00433186	USA	30		Phase I	Recruiting
Rapamycin vs Methotrexate	NCT00241189	USA	18		Phase I/II	Ongoing Not Recruiting
Cyclophosphamide IV 4 days plus GCSF	NCT00501995	USA	30		Phase III	Ongoing Not Recruiting
Mycophenylate vs Cyclophosphamide (oral)	NCT00883129 (For ILD)	USA	150		Phase II	Recruiting
MEDI-546	NCT00930683	USA	29		Phase I (No Placebo)	Recruiting
MEDI-546	NCT00946699	USA	29		Phase I (Placebo)	Not Yet Open
Anti-CD20	NCT00936546	Belgium	10		Controlled Phase	Recruiting
Anti-CD20 plus methylprednisolone	NCT00379431	Belgium	8		Phase II	Ongoing Not Recruiting
Abatacept	NCT00442611	USA	12		Phase I/II Placebo Control	Recruiting

Drug/Intervention	Clinical Trial Identifier	Country	Enrollment		Study Type	Status
			Number			
Transplant/Allogeneic Hematopoietic Stem Cell	NCI00282425	USA	10		Phase I	Recruiting
Transplant/Mesenchymal Stem Cell	NCI00962923	China	20		Phase II/III	Recruiting
Transplant/Antologous CD34+ Stem Cell vs Cyclophosphamide	NCI00114530	USA	226		Phase II/III	Recruiting
Transplant/Hematopoietic Stem Cell Support, Cyclophosphamide plus rabbit anti-thymocyte globulin	NCT00278525	USA	60		Phase II	Recruiting
UV A1-Irradiation	NCT00628797	Germany	60		Phase I/II	Recruiting
AIMSPRO (Hyperimmune Caprine Serum)	NCT00769028	United Kingdom	20		Phase II (Placebo Control)	Recruiting
GB-0998 IVIG	NCT00348296	Japan	60		Phase III (Placebo Control)	Ongoing Not Recruiting
Platelet Gel	NCT00463125 (Digital Ulcers)	Italy	40		Phase II/III (Placebo Control)	Recruiting

* As listed on <http://clinicaltrials.gov>