Progressive loss of lymphatic vessels in the skin of systemic sclerosis patients

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Objective Systemic sclerosis (SSc) is a connective tissue disorder of unknown etiology characterized by microvascular and fibrotic changes in skin and internal organs. In SSc, several studies have described an irregular architecture and progressive loss of blood vessels, while the role of the lymphatic vascular system is poorly known. Our aim was to evaluate dermal lymphatic vessels in SSc patients according to different phases of skin involvement.

Methods Full-thickness skin biopsies were obtained from the clinically involved skin of the distal forearm of 25 SSc patients (11 limited and 14 diffuse SSc). Patients were classified as early (n=10) and late (n=15) SSc phase according to disease duration and histopathological stage of skin involvement. Skin samples from the same forearm region of 13 age- and sex-matched healthy donors were used as controls. Skin sections were stained with an antibody against podoplanin (D2-40), which is specifically expressed in lymphatic endothelial cells but not in blood endothelial cells. After incubation with fluorochrome-conjugated secondary antibodies, the sections were examined by confocal laser scanning microscopy. D2-40–positive lymphatic vessels were counted in at least 10 randomly chosen high-power fields (hpf) of the papillary dermis and 10 hpf of the reticular dermis per sample by 2 independent blinded observers. Data were analyzed using the Student *t* test.

Results The number of lymphatic vessels was significantly reduced in the papillary (1.03 ± 0.52/hpf vs 1.64 ± 0.44/hpf, $p<0.05$) and reticular (1.08 ± 0.35/hpf vs 1.79 ± 0.66/hpf, $p<0.005$) dermis of SSc patients as compared with healthy individuals. No differences were observed between limited and diffuse SSc. In particular, the number of lymphatic vessels in early SSc was not different from that in controls in the papillary dermis (1.42 ± 0.36/hpf), and showed a trend toward a reduction in the reticular dermis (1.21 ± 0.33/hpf). Instead, a significant reduction in lymphatic vessels in late SSc compared with controls was found (papillary dermis: 0.72 ± 0.42/hpf, $p<0.005$; reticular dermis: 0.99 ± 0.36/hpf, $p<0.01$). Moreover, the number of lymphatic vessels in the papillary dermis of late SSc was significantly lower as compared with early SSc ($p<0.005$).

Conclusions In SSc, lymphatic microangiopathy is linked to the progression and severity of skin involvement. Furthermore, our data indicate that lymphatic vessel injury starts in the reticular dermis and subsequently extends to the papillary dermis. The progressive disappearance of lymphatic vessels may have a critical pathogenetic role in the evolution of SSc from an early edematous phase to overt fibrosis.

Key words
Systemic sclerosis, lymphatic vessels, skin