The Proadhesive Phenotype of Systemic Sclerosis Skin Promotes Myeloid Cell Adhesion

Bradley J. Rabquer1, Yong Hou1, Francesco Del Galdo2, G. Kenneth Haines III3, Michele L. Jaffe1, Sergio A. Jimenez2, James R. Seibold1, and Alisa E. Koch4,1

ABSTRACT

Systemic sclerosis (SSc) is characterized by microvascular abnormalities and leukocyte infiltration. Previous studies have suggested a proadhesive phenotype in SSc skin, with several adhesion molecules overexpressed in SSc skin and on SSc dermal fibroblasts including VCAM-1, ICAM-1, and CD99. However, the role of these molecules in leukocyte adhesion to SSc skin is unknown. The aim of this study was to determine the expression and role of selected adhesion molecules on normal (NL), proximal SSc, and distal SSc skin fibroblasts.

Methods: The expression of ICAM-1, JAM-B, JAM-C, CD44, and CD99 and VCAM-1 on SSc fibroblasts was determined using flow cytometry. Immunohistological analysis of the skin of SSc patients and controls was performed using standard procedures and double labeling fluorescent techniques. Adhesion assays were performed using U937 cells and SSc fibroblasts. The percent of maximal binding was determined using U937 cells and SSc fibroblasts in the presence or absence of antibodies against ICAM-1, VCAM-1, or both antibodies, or IgG control.

Results: ICAM-1, JAM-B, and JAM-C were expressed on SSc fibroblasts. Immunohistological analysis showed that CD99 was expressed in the stratum spinosum of normal and SSc skin. Overexpression of select adhesion molecules was found in SSc skin fibroblasts. The percent of maximal binding was significantly different in the presence or absence of antibodies against ICAM-1, VCAM-1, or both antibodies, or IgG control.

Conclusions: These results suggest that ICAM-1 and VCAM-1 functionally mediate myeloid cell adhesion to SSc skin fibroblasts. However, future studies are needed to determine the role of these molecules in the pathogenesis of SSc skin.

INTRODUCTION

Systemic sclerosis (scleroderma, SSc) is a connective tissue disorder characterized by Raynaud’s phenomenon, vascular abnormalities, and fibrosis of the skin and internal organs. Patients with SSc have increased numbers of circulating inflammatory cells, which may contribute to the microvascular abnormalities, fibrosis, and immune-mediated skin destruction.

MATERIALS AND METHODS

Patient samples: Peripheral samples from SSc patients with diffuse SSc (DSSC) and limited cutaneous SSc (LSSC) were obtained. Burned and blanched skin, one from the proximal (less clinically involved) and the other from the distal (more clinically involved) forearm of SSc patients were collected from the Scleroderma Research Center of the University of Michigan.

Immunohistochemical analysis: Frozen skin samples were subjected to immunohistology using standard procedures. Immunohistological analysis was performed using standard protocols and double labeling fluorescent techniques. Immunohistological analysis was performed using standard protocols and double labeling fluorescent techniques. Immunohistological analysis was performed using standard protocols and double labeling fluorescent techniques. Immunohistological analysis was performed using standard protocols and double labeling fluorescent techniques.

Adhesion assays: Adhesion assays were performed using U937 cells and SSc fibroblasts. The percent of maximal binding was determined as the number of cells adhering to the fibroblasts in the presence of the test antibody divided by the number of adherent cells on the control antibody.

ACKNOWLEDGMENT

This work was supported by the National Institutes of Health (grants AI-49976 and AR-48277), the Office of Scientific Development and the Developmental Research Program of the Veterans Affairs, the Scleroderma Research Foundation, and the University of Michigan Center for Translational Science Activities (award U54 RR024988, and by funding from the Scleroderma Center of the University of Michigan.)