Relationship Between Parity and Clinical and Biological Features in Patients with Systemic Sclerosis

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ABSTRACT. ***Objective***. To assess the influence of parity on the clinical and biological features of systemic sclerosis (SSc).

Methods. We recorded the following clinical and biological data of 100 consecutive women with SSc: age, disease duration before diagnosis, cutaneous extension of sclerosis according to LeRoy's classification, pulmonary involvement, and antinuclear antibodies. We compared these features to the number and sex of children who were born before SSc onset. Date of birth of the first children was systematically recorded.

Results. Patients with limited SSc had more children before SSc onset than patients with diffuse SSc (2.4 ± 1.8 vs 1.7 ± 1.5; $p < 0.05$). The interval between first birth and SSc onset was shorter for patients with limited SSc than for patients with diffuse SSc (11.0 ± 9.9 vs 23.5 ± 14.5 yrs; $p < 0.01$). Patients with pulmonary fibrosis had more children than patients without pulmonary fibrosis (2.5 ± 1.9 vs 2.0 ± 1.6; $p < 0.05$). Age at first birth was significantly higher when the child was a girl than a boy (26.8 ± 7.5 vs 22.9 ± 5.3 yrs; $p < 0.05$). The interval between the first birth and SSc onset was shorter when the child was a girl than a boy (16.2 ± 9.6 vs 25.4 ± 13.4 yrs; $p < 0.05$).

Conclusion. Pregnancy related microchimerism could be preferentially associated with limited SSc and pulmonary fibrosis. Microchimerism may be facilitated in cases in which the fetus is female. (J Rheumatol 2001;28:509–13)

Key Indexing Terms:
SYSTEMIC SCLEROSIS PREGNANCY MICROCHIMERISM

Systemic sclerosis (SSc) is a connective tissue disease characterized by cutaneous and visceral progressive fibrosis, microvascular changes with endothelial cell damage, and perivascular inflammatory infiltration[1]. SSc has a strong predilection for women (sex ratio as high as 14:1) after child bearing years (age-specific incidence between 45 and 55 yrs)[2].

During pregnancy, bidirectional traffic of cells at the fetal-maternal interface is well known[3]. Fetal cells of various types are found in the peripheral blood of most pregnant women[4-9]. Fetal progenitor cells (CD34+CD38+ cells) can persist for decades after childbirth in some women[10]. Nelson, *et al* showed that this microchimerism of fetal origin was found more frequently and was markedly quantitatively greater in patients with SSc than in healthy patients[11]. Artlett, *et al* reported fetal cells in skin biopsies of SSc but not in controls[12]. Moreover, SSc shares clinical features with chronic graft-versus-host disease, a chimeric disorder that occurs in recipients of allogenic stem cell transplantation[13,14]. These findings led Nelson, *et al* to suggest that persisting microchimerism of fetal origin could be involved in the pathogenesis of SSc[11,15,16]. However, SSc can occur in nulligravid women or in men too, suggesting either other causes of microchimerism or the participation of other pathogenic mechanisms. Moreover, there are several clinical subsets of SSc and it is possible that pregnancy related microchimerism leads preferentially to the occurrence of a specific SSc subset.

We assessed the relationship between the characteristics of pregnancies and the clinical and biological features of SSc.

MATERIALS AND METHODS

Study population. We studied 100 consecutive women with SSc fulfilling the American College of Rheumatology criteria[17] and followed in the Department of Internal Medicine of the Claude Huriez Hospital (Lille, France) between 1990 and 1999. Date of disease onset was defined as the date of the occurrence of the first symptom of SSc [Raynaud's phenomenon (RP) in all cases], and age at diagnosis was defined as the age when the patient was first told by a physician that the diagnosis was SSc. Cutaneous extension was graded according to LeRoy's classification system[18]: limited (hands, forearms, face, or feet) or diffuse (truncal and acral). CREST syndrome (diagnosed if 3 of the 5 following items were present: calcinosis, RP, esophageal involvement, sclerodactyly, telangiectasia) was included in the limited SSc group. Pulmonary fibrosis was assessed by chest computed tomodensitometry and pulmonary function tests. Pulmonary fibrosis was defined by the presence of parenchymal micronodules, honeycombing, or linear opacities on chest computed tomodensitometry and abnormal lung

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function (lung volumes < 80% of predicted values and/or diffusing capacity < 75% of that predicted)[19-20]. Antinuclear antibodies (indirect immunofluorescence on rat liver) were either anticentromere (on HEp-2 cells), anti-Scl70 (by immunoblot analysis), or antinucleolar antibodies (on HEp-2 cells). We recorded the number, sex, and date of birth of the children in all patients.

Statistical analysis. Number of children according to the subtypes (limited vs diffuse SSc) were compared using Student's T test and chi-square test with Yates' correction when necessary. Pulmonary fibrosis was significantly more frequent in patients with diffuse SSc and anticentromere antibodies (ACA) were more frequent in limited SSc. Therefore, we compared the number of children between patients with and without pulmonary fibrosis as well as between patients with and without ACA using a 2 way analysis of variance (ANOVA) including the subtype of SSc. Concerning the interval between first pregnancy and SSc onset according to the subtype of SSc and to the presence of ACA, we performed an analysis of covariance (ANCOVA, with age at disease onset as covariate) and a 2 way ANCOVA (including subtype of SSc), respectively. To compare the number of girls, we performed a 2 way ANOVA including the subtype of SSc and the number of children. To compare age at first birth according to the sex of the child, we performed a 2 way ANOVA including the subtype of SSc. To compare the interval between the first birth and the SSc onset according to child's sex, we performed a 2 way ANCOVA including child's sex and subtype of SSc as well as age at disease onset as covariate.

RESULTS

Characteristics of the SSc (Table 1). SSc was limited in 72 patients (72%) including 47 CREST (47%); and SSc was diffuse in 28 patients (28%). Age at diagnosis did not differ significantly between patients with limited SSc and patients with diffuse SSc (51.8 ± 13.9 vs 52.8 ± 15.9; NS). Patients with limited SSc had a longer disease duration before diagnosis than patients with diffuse SSc (15.6 ± 12.9 vs 11.6 ± 9.2 yrs; $p < 0.05$). Pulmonary involvement was more frequent in patients with diffuse SSc than in patients with limited SSc (22/28 vs 17/72; $p < 0.05$). ACA were more frequent in patients with limited SSc than in patients with diffuse SSc (47/72 vs 0/28; $p < 0.05$). Conversely, anti-Scl70 antibodies were more frequent in patients with diffuse SSc than in patients with limited SSc (15/28 vs 7/72; $p < 0.05$).

Number of children and SSc features. Eighty-three patients had had children, with a mean number of children of 2.2 ± 1.8**.** Median age at the first birth was 24 years. Patients with limited SSc had significantly more children before SSc onset than patients with diffuse SSc (2.4 ± 1.8 vs 1.7 ± 1.5; $p < 0.05$). We compared frequency of limited SSc between nulliparous patients and patients with more than 1 child [10/17 (58.8%) vs 62/83 (74.7%); $p = 0.18$], between patients with 0 or 1 child and patients with 2 or more children [23/37 (62.1%) vs 49/63 (77.7%); $p = 0.09$], and finally patients with 0, 1, or 2 children and patients with 3 or more children [44/68 (64.7%) vs 28/32 (87.5%); $p < 0.05$] (Figure 1). Patients with ACA had the same number of children as patients without ACA (2.3 1.6 vs 2.2 1.9; $p = 0.48$). Patients with pulmonary fibrosis had significantly more children before SSc onset than patients without pulmonary fibrosis (2.5 ± 1.9 vs 2.0 ± 1.6; $p < 0.05$). Among patients with limited SSc, patients with pulmonary fibrosis had more children than patients without pulmonary fibrosis (3.5 ± 2.3 vs 2.1 ± 1.6; $p < 0.05$). The difference was not statistically significant for patients with diffuse SSc (1.9 ± 1.5 and 1.3 ± 1.2; NS) (Figure 2).

Interval between first pregnancy and SSc onset. The mean age at first birth was 25.2 ± 6.2 years for patients with limited SSc and 26.2 ± 7.6 years for patients with diffuse SSc (NS). Seventeen patients were nulliparous (11 limited SSc and 6 diffuse SSc). The age of onset of diffuse versus limited SSc in nulliparous women was 35.1 ± 17.4 years versus 38.0 ± 19.3 years; $p = 0.48$. Eighty-three patients were multiparous (62 limited SSc and 24 diffuse SSc). The age of onset of diffuse versus limited SSc in multiparous women was 49.4 ± 17.1 versus 35.8 ± 14.4 years; $p < 0.01$. The interval between the first birth and SSc onset was significantly shorter for patients with limited SSc than for patients with diffuse SSc (11.0 ± 9.9 vs 23.5 ± 14.5 yrs; $p < 0.01$). This interval was also shorter for patients with ACA than for patients without ACA but it did not reach statistical significance in multiple ANCOVA analysis (18.0 ± 10.5 vs 23.8 ± 13.8 yrs; $p = 0.1$).

Sex of the children and SSc features. Patients with limited SSc had the same number of girls as patients with diffuse SSc (1.3 ± 1.1 vs 1 ± 0.8; $p = 0.98$). Age at first birth was significantly higher when the child was a girl than when he was a boy (26.8 ± 7.5 vs 22.9 ± 5.3 yrs; $p < 0.05$). Interval between the first birth and SSc onset was shorter when the first child was a girl than when he was a boy (16.2 ± 9.6 vs 25.4 ± 13.4 yrs; $p < 0.05$).

Table 1. Comparison of patients with limited SSc and patients with diffuse SSc. Frequencies were compared using the chi-square test with Yates' correction. Means were compared using Student's t test.

	Limited SSc (n = 72)	Diffuse SSc (n = 28)	p
Mean age at diagnosis	51.8 ± 13.9	52.8 ± 15.9	NS
Mean age at SSc onset	36.2 ± 15.4	42.8 ± 19	< 0.01
Pulmonary involvement (%)	17 (23.6)	22 (78.57)	< 0.05
Presence of anticentromere antibodies (%)	47 (65.3)	0	< 0.001
Presence of anti-Scl70 antibodies (%)	7 (9.7)	15 (53.5)	< 0.05

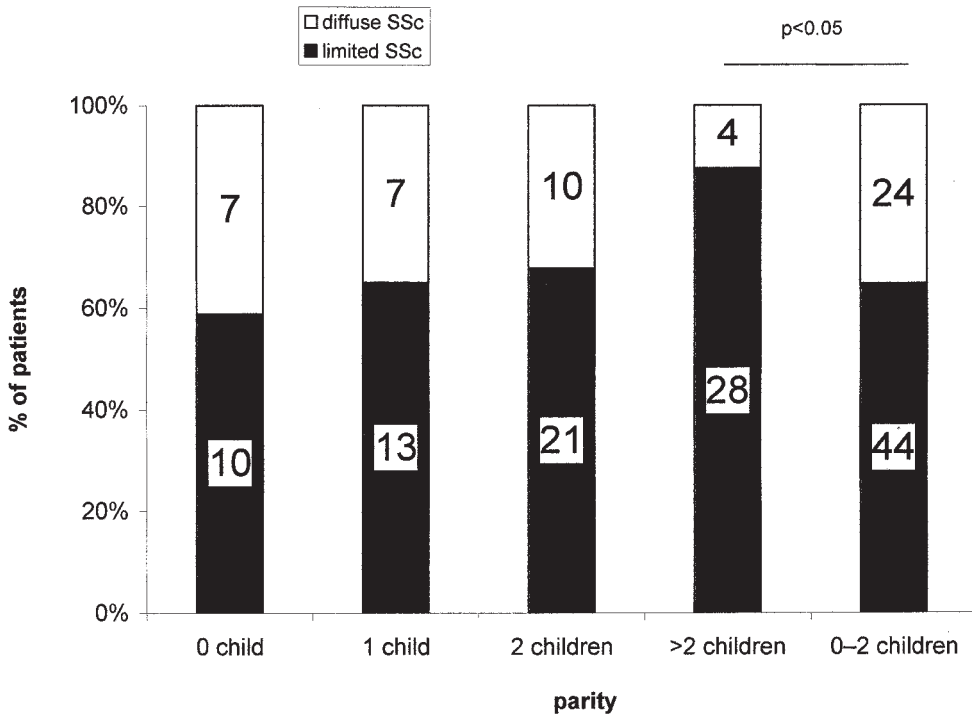

Figure 1. Parity and systemic sclerosis type. Patients who had > 2 pregnancies had significantly more frequently a limited SSc than patients who had 2 pregnancies (chi-square test with Yates' correction).

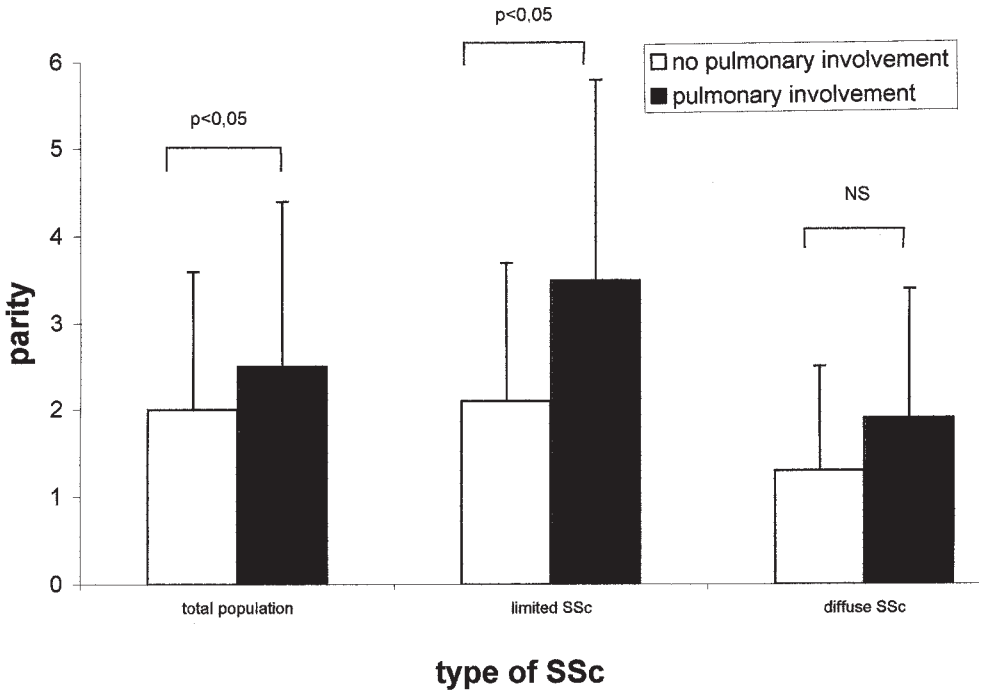

Figure 2. Parity and pulmonary involvement. Patients with pulmonary involvement had significantly more children before SSc onset than patients without pulmonary involvement.

DISCUSSION

Several studies have recently suggested the role of pregnancy related microchimerism in the pathogenesis of SSc[11,15,16,21]. However, the role of microchimerism in SSc pathogenesis remains controversial[22]: it does not address the occurrence of SSc in nulliparous women or in men, even if

an alternative source of microchimerism is proposed such as microchimerism of maternal origin[3,23], unrecognized fetal loss, engraftment of cells from an unrecognized twin, or blood transfusion[11,12,21,24]. Further, the pathogenesis of SSc probably involves other mechanisms such as exposure to toxic substances[25,26] or to other cells such as mast cells[27,28]. Therefore, several subsets of SSc may exist with distinct pathophysiological mechanisms; one of them could be microchimerism of fetal origin.

We assessed the possible influence of parity on the features of SSc. First, we found that most of the patients with SSc (83%) had children before SSc onset, with a mean number of 2.2 ± 1.8. We then found that multiparity was mainly associated with limited SSc and pulmonary fibrosis. Moreover, limited SSc occurred more quickly after the first birth than diffuse SSc. Conversely, nulliparous women with limited SSc did not have an earlier onset of disease than in nulliparous women with diffuse SSc. This could be an additional reason to think that the shorter interval between the first birth and disease onset in limited SSc versus diffuse SSc has a biological significance. Taken together, these results may suggest that multiparity may be involved. However, an alternative explanation to the observed difference of parity between limited and diffuse SSc could be that fertility and/or pregnancy outcome may be different between these 2 subsets of SSc. Discrepancies are found in the literature concerning fertility and pregnancy outcome in patients with SSc. Silman, *et al* and Englert, *et al* reported an increased incidence of spontaneous abortion and infertility in women with SSc before disease onset[29,30]. However, no information about the subtype of SSc (limited or diffuse SSc) was available in these studies. Moreover, a recent study reports that infertility and miscarriage were not more frequent in patients with SSc than in controls[31]. This study involved patients with diffuse SSc (45%). The only reported difference between patients with diffuse versus limited SSc is that patients who chose not to have children were more likely to have diffuse disease than the normal control women. This could explain why, in our study, patients with diffuse SSc with an earlier onset of disease were nulliparous. However, although we cannot totally rule out that infertility and/or miscarriages are more frequent in patients with diffuse SSc than patients with limited SSc, the literature does not provide firm evidence for this hypothesis. Therefore, we have also suggested that limited SSc and pulmonary fibrosis may be a subtype of SSc in which pregnancy related microchimerism is involved. We think that subtypes of SSc should be distinguished in future studies about microchimerism.

The factors that facilitate microchimerism are poorly understood. Tolerance of women to fetal cells is probably of great importance to prevent the clearance of these cells by the mother's immune system[32]. Risk of chronic graft-versus-host disease after stem cell transplantation or blood transfusion is greater if donor and recipient have HLA compatibility[33]. We found that the interval between the first birth and the SSc onset was shorter when the first child was a girl. Bonney, *et al* showed that pregnancy can be associated with priming for a cellular immune response against H-Y Ag, which could facilitate clearing of the migrating male fetal cells and account for greater microchimerism in pregnancies with a female fetus[34]. However, this is difficult to prove, because technical methods to detect microchimerism can only detect microchimerism from a male fetus. This possible greater microchimerism in the female fetus may account for the shorter interval observed between the first birth and the disease onset in this case.

We found no correlation between diffuse SSc, anti-Scl70 antibodies, and parity. Pathogenesis of diffuse SSc may involve other mechanisms such as exposure to toxic agents[25,26]. Nietert, *et al* reported that anti-Scl70 antibodies were more frequent in patients exposed to solvents[26]. Non-immune cells may also be involved in the pathogenesis of SSc, especially diffuse SSc. In previous studies performed on labial salivary glands of patients with SSc we suggested a role for tissue mast cells in SSc pathogenesis[27,28].

We also found that patients with pulmonary fibrosis had significantly more children before disease onset, and this was mainly observed in patients with limited SSc. The significance of this correlation remains unclear, but it could represent new evidence for the participation of T lymphocytes in the pathogenesis of pulmonary fibrosis in SSc. In a recent study, Atamas, *et al* found a correlation between cytokine production by CD8+ lung cells and decline in pulmonary function in patients with SSc[35]. In another study, Wells *et al* found an increase in memory T cells in lung interstitium of patients with SSc[36]. One can suggest that some of these CD8+ and memory T cells are of fetal origin. However, we did not compare these results with a control group of patients with non-SSc related pulmonary fibrosis, which would have provided important information regarding the disease specificity of this finding.

To conclude, our study suggests that multiparity could be preferentially associated with limited SSc and pulmonary fibrosis. Limited SSc may therefore be the subset of SSc in which pregnancy related microchimerism is involved. Our study suggests that limited and diffuse SSc may represent distinct disorders, each with its own causes and pathogenic mechanisms. Moreover, we think that subtypes of systemic sclerosis should be investigated in future studies about microchimerism.

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