

Immune response and tolerability of varicella vaccine in children and adolescents with systemic lupus erythematosus previously exposed to varicella-zoster virus

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Abstract

Objectives

The aim of the present paper is to evaluate the immune response and tolerability of varicella vaccine in children and adolescents with systemic lupus erythematosus previously exposed to varicella-zoster virus.

Methods

We performed a prospective and controlled study on a group of 54 SLE patients that were chosen at random to be or not to be vaccinated (28 were vaccinated and 26 were not). Twenty-eight healthy controls, of matching age and sex were also vaccinated. All were submitted to a questionnaire, physical evaluation and laboratory assays: lymphocyte immunophenotyping by flow cytometry, plasma varicella zoster virus (VZV) serology by ELISA and in vitro interferon gamma (IFN γ) production by T-cells after stimulus with VZV antigen. They were evaluated before vaccination and at 30, 45, 180 and 360 days afterwards.

Results

We did not observe any differences in the frequency of adverse events in both vaccinated groups. At study entry, all individuals were seropositive for VZV antibodies. The serum VZV antibody titres similarly increased after vaccination. The frequency of flares and the SLEDAI score were also similar among the patients. Thirty days after vaccination the production of IFN γ specific to VZV was lower in the SLE group compared to healthy controls. In the follow-up we observed 4 cases of herpes zoster in the SLE unvaccinated group, but no zoster in the vaccinated group.

Conclusion

The varicella vaccine was well tolerated in SLE group, who had pre-existing immunity to varicella. The varicella vaccine immunogenicity measurement by serum antibody titres was appropriate. The incidence of HZ was lower in the vaccinated lupus group.

Key words

herpes zoster, immunocompromised host, systemic lupus erythematosus, vaccine, varicella

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Introduction

Varicella zoster virus (VZV) infections as primary infection or as reactivation of VZV as herpes zoster (HZ) are often severe in patients with SLE. A number of studies have confirmed a high incidence of HZ in SLE patients (1-4) who are especially prone to recurrent zoster episodes (2, 5).

HZ in lupus patients might be prevented or attenuated by boosting VZV with live VZV vaccine as has been achieved in healthy elderly persons (6-8) or as the results of studies in HIV patients suggest (9, 10).

Although live vaccines remain contraindicated in patients taking immunosuppressant, a recent review has considered the possibility for the administration of varicella/zoster vaccination in patients with immune-mediated inflammatory diseases (11).

Due to the absence of data concerning the risk/benefit ratio for varicella vaccine in patients with SLE, the present study evaluated the immune response and tolerability of this vaccine in children and adolescents with SLE previously exposed to varicella-zoster virus.

Patients and methods

Our sample initially comprised 134 eligible patients, resulting in 54 that fulfilled the required criteria and accepted to participate in the study. These patients were compared to a group of 28 healthy controls.

Study design

We performed a prospective, blind randomised and controlled study. Two groups of SLE patients (54 patients) were randomly assigned to be vaccinated or not and a group of healthy controls (28 individuals) were also included to be vaccinated.

SLE patients who attended two Pediatric Rheumatology Outpatient Clinics from March 2007 to September 2008 were selected. Healthy children and adolescents were selected from a local school and were of matching for gender and age with the patients. This study was approved by the local Institutional Review Board and written informed consent was obtained from all participants.

Study population

Eligible patients were aged from 5 to 18 years old, all fulfilled the 1997 revised criteria for classification of SLE (12) and had been on immunosuppressant for at least 6 months. Since live attenuated vaccines are usually contraindicated for SLE patients (13-16), we had no available guidelines to base on for inclusion and exclusion criteria. In view of that, we decided to exclude patients who had received mycophenolate, intravenous immunoglobulin, cyclophosphamide or methylprednisolone in the last 3 months. Other immunosuppressants such as cyclosporine (up to 3 mg/kg/day), azathioprine (up to 3 mg/kg/day) and methotrexate (up to 20 mg/week) were acceptable. The allowed dose of corticosteroids was below 2 mg/kg/day or up to 20 mg/day (17). Patients who were taking acetylsalicylic acid were evaluated and, in the case of the antiphospholipid antibodies being negative, the drug was discontinued 4 weeks prior to vaccination and reintroduced 6 weeks after (due to risk of Reye syndrome in individuals taking this drug who are going to be vaccinated with varicella vaccine). Leucocyte count below 700/mm³ and platelets below 100,000/mm³ on the day of vaccination were also exclusion criteria. Previous history of varicella infection and SLEDAI score (Systemic Lupus Erythematosus Activity Index) (18) were not exclusion criteria. The mean follow-up time was 35.6 months (range 24-42 months). At assessment, children and adolescents from the control group had to be free of disease and not taking any medication.

Lupus flare was defined as the need for corticosteroids introduction or increase in the baseline dosage and/or the addition of immunosuppressant during the study period (19).

Follow-up

All patients and controls were evaluated before vaccination and at days 30, 45, 180 and 360 afterwards. One year and two years after the end of the trial, the patients were called to answer about flares and HZ episodes.

At the first visit, they were all submitted to a questionnaire, physical evaluation and laboratory tests, including immu-

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nophenotyping of lymphocytes, VZV plasma antibody titres and specific varicella zoster interferon gamma (IFN γ) production by CD4⁺ and CD8⁺ T cells. The questionnaire contained questions about previous contact with VZV, such as previous infection, HZ or immunisation and about recent household contact with varicella. Their disease activity was measured by the SLEDAI (18).

VZV vaccine administration

Participants received one subcutaneous injection of varicella vaccine (Biken[®] Aventis Pasteur, ≥ 1000 plaque-forming units of virus/0.5 mL) in the deltoid region.

Evaluation of vaccine adverse events

On enrolment, subjects received a pre-formatted diary to report any local or systemic reaction for the 45 post-vaccination days, including signs and symptoms of HZ and lupus flares. Those who had any adverse reaction were instructed to immediately contact the physicians of our study group by an interactive telephone-response system, so that they could be promptly evaluated and receive acyclovir in case of the appearance of any rashes. Subjects were contacted weekly in the first weeks and thereafter at the regular consultation and at days 30, 45, 180 and 360 post-vaccination.

Blood and urine samples collection

SLE patients had a blood and urine sample collected at days 0, 30, 180 and 360. Controls had a blood sample collected on the same days and a urine sample was collected only on day 0, for pregnancy test.

Laboratory assessment

At days 0, 30, 180 and 360, routine laboratory tests were performed on the 54 patients. Antinuclear and double stranded DNA antibodies were detected by indirect immunofluorescence using HEp-2 cells as substrate and the presence of IgG or IgM anticardiolipin antibodies was assessed by ELISA (enzyme-linked immunosorbent assay). Total haemolytic complement activity (CH 100) was measured by immunohemolysis assay. A complete blood cell

count (CBC) was performed for the healthy control group at day 0, before vaccination.

Immunophenotyping of lymphocytes was performed on all subjects (SLE patients and controls) at day 0, by 4-colour flow cytometry analyses (FACSCalibur, BD Biosciences, Franklin Lakes, USA).

VZV humoral immunity

Varicella antibodies were measured by indirect ELISA, as previously described (20). Individuals with varicella antibody concentrations equal to or greater than 0.1 IU/mL were considered to be fully protected; those with antibodies equal to or greater than 0.05 and below 0.1 IU/mL were considered to have doubtful immunity; individuals with antibody levels below 0.05 IU/mL were considered to be non-immune (21).

VZV cellular immunity

Whole blood was diluted 1 in 10 in RPMI 1640 culture medium (Gibco, New York, USA), supplemented with L-glutamine and antibiotics (Gibco), and distributed into polystyrene tubes. Samples were cultured for 120h in the presence of 5 μ L varicella zoster antigen (cell supernatant of varicella infected cells) (batch numbers 03050A1 and 03051A1) (Microbix, Toronto, Canada) or control varicella zoster antigen (cell supernatant of non-infected cells) (batch numbers 29001A2 and 29003A1) (Microbix). Preliminary experiments showed that this antigen concentration and incubation period resulted in maximum response to varicella zoster antigen. Brefeldin A (Sigma, St Louis, USA), (10 μ g/mL) was added to all tubes for the last 4h of culture. After the incubation period, 2mM EDTA (Gibco) was added; cells were pelleted, the supernatant was removed and the cells were lysed for 10 minutes. After cell washing with phosphate-buffered saline, CD3-APC and CD8-PerCP conjugated monoclonal antibodies (BD Biosciences) were used for cell-surface staining. Cells were then fixed, washed, resuspended with permeabilisation buffer and incubated for 10 min at room temperature in the dark. IFN γ -FITC conjugated monoclonal antibody

was then added. Finally, the cells were washed and kept in the dark at +4°C until data acquisition. Sample acquisition was performed with FACSCalibur Cytometer (BD Biosciences) using CellQuest software (BD Biosciences). Analysis was performed using FlowJo software (Tree Star, Ashland, USA). Fifty thousand events were acquired in the lymphocyte gate based on the forward scatter and side scatter dot plot. CD3⁺T cells were selected based on the side scatter profile and CD3-APC fluorescence. CD8⁺T lymphocytes were defined as CD3⁺/CD8⁺; CD4⁺T lymphocytes were defined as CD3⁺/CD8⁻. Intracellular IFN γ production was evaluated in CD3⁺/CD8⁺ and CD3⁺/CD8⁻ cells. The final value of positive cells to each stimulus was obtained by subtracting the percentage of positive cells of the culture without stimulus (negative control) from the culture in the presence of stimulus (varicella antigen).

Statistical analysis

Numerical unrelated variables were analysed by *t*-test and numerical related variables, by paired *t*-test. For categorical variables, Chi-Square test or Fisher's exact test were used. Repeated measure ANOVA was used for analysis of data evaluated more than once, with multiple comparisons performed by Tukey test. For variables which were not normally distributed, Wilcoxon's or Mann-Whitney U-tests were used. To control the overall α level, Bonferroni adjustment was used. Level of significance was set at $p < 0.05$.

Results

Demographic and clinical data

One hundred and thirty-four patients were screened: 58 were excluded due to use of immunosuppressants; 6 due to comorbidities; one due to thrombocytopenia and 15 refused to participate. Demographic data of patients and controls at study entry are shown in Table I.

Previous VZV infection was reported by 19/28 (67.8%) patients in SLE vaccinated group, 21/26 (80.7%) in SLE unvaccinated group and 17/28 (60.7%) controls. Five SLE patients reported HZ in the past, 3 patients in the vaccinated group and 2 in the unvaccinated group;

Table I. Characteristics of the three groups: vaccinated and unvaccinated systemic lupus erythematosus (SLE) patients, and vaccinated controls at the study entry.

Parameters	SLE (n=54)		Vaccinated controls (n=28)	p-value
	Vaccinated (n=28)	Unvaccinated (n=26)		
Mean age in years (range) $\pm$ standard deviation	15.3 $\pm$ 2.5 (9.9–18.8)	14.0 $\pm$ 3.1 (7.6–18.3)	15.0 $\pm$ 2.5 (10.1–18.7)	0.2339 [§]
Number of female individuals (%)	21 (75.0)	22 (84.6)	21 (75.0)	0.6801 [§]
Number of individuals non Caucasians, n. (%)	20 (71.4)	14 (53.8)	21 (75.0)	0.2590 [§]
Mean time $\pm$ standard deviation in years since SLE diagnosis (range)	4.5 $\pm$ 2.6 (1.4–9.1)	4.7 $\pm$ 2.8 (0.7–10.0)	NA	0.7478 [§]
Mean SLEDAI $\pm$ standard deviation (range)	0.93 $\pm$ 1.6 (0–4)	1.62 $\pm$ 2.3 (0–8)	NA	0.1997 [†]
Number of patients on oral corticosteroids, n. (%)	18 (64.3)	18 (69.2)	NA	0.7001 [§]
Mean daily dose of corticosteroids $\pm$ standard deviation (range) (mg)	7.5 $\pm$ 3.9 (3–12.5)	9.4 $\pm$ 4.8 (2.5–15)		0.0939 [†]
Number of patients on azathioprine, n. (%)	9 (32.1)	12 (46.1)	NA	0.2913 [§]
Mean daily dose of azathioprine $\pm$ standard deviation (mg/kg/day) (range)	1.5 $\pm$ 0.5 (0.7–2.4)	1.9 $\pm$ 0.6 (1.4–3)		0.2170 [†]
Number of patients on cyclosporine, n. (%)	0	2 (7.7)	NA	0.2271 [‡]
Mean daily dose of cyclosporine $\pm$ standard deviation (mg/kg/day) (range)	NA	1.75 $\pm$ 0.3 (1.5–2.0)		
Number of patients on methotrexate, n. (%)	2 (7.1)	0	NA	0.4913 [‡]
Mean weekly dose of methotrexate (mg)	20 NA			
Number of patients on chloroquine, n. (%)	27 (96.4)	22 (84.6)	NA	0.1346 [§]
Mean daily dose of chloroquine $\pm$ standard deviation (mg/kg/day) (range)	4.6 $\pm$ 1.0 (3.2–6.6)	5.2 $\pm$ 1.0 (2.6–6.0)	NA	0.0363 ^{†*}

SLEDAI: systemic lupus erythematosus disease activity index. NA: not applicable. [§]ANOVA. [§]Chi-square test. [†]t-test. [‡]Fisher's exact test. (12 patients were on corticosteroids and azathioprine).

none of the controls had presented HZ in the past. Only one SLE patient and 2 controls had received one dose of varicella vaccine before the study entry. None of the study participants reported household exposure to varicella within the last 30 days of study entry.

Adverse events

Up to 45 days after the vaccine, 42.8% of the vaccinated individuals presented adverse events. However, only mild adverse events were reported, with no significant difference between the 3 groups ($p=0.1663$). None of the vaccinated patients presented disseminated varicella rash or HZ (Table II).

The frequency of flares was the same for both SLE groups, affecting only 4 patients in each group. Regarding SLEDAI, the 2 groups behaved similarly throughout time ($p=0.2062$). However, an increase in the index between 30 and 180 days after vaccine was observed in both SLE groups ($p=0.036$, ANOVA with repetitive measures).

Lymphocyte immunophenotyping

Total lymphocyte counts were similar in both SLE patients groups (vaccinated and unvaccinated) and lower than in controls ($p=0.005$). There was no difference observed in the number of CD3⁺, CD4⁺ and CD8⁺ T cells. In contrast, both NK and B-cell counts were lower in SLE patients than in controls

($p<0.0001$ and $p=0.009$, respectively) (Table III).

There was no statistical difference between SLE patients and controls regarding the mean percentual of maturation subsets of CD4⁺ T, CD8⁺ T cells and B lymphocytes. The co-stimulatory molecules CD28 assessed on CD4⁺T and CD8⁺T cells and CD80 and CD86 as-

Table II. Adverse events presented by vaccinated and unvaccinated systemic lupus erythematosus (SLE) patients and vaccinated controls, in the first 45 days after vaccination.

Adverse events	SLE		Vaccinated controls n=28 (%)
	Vaccinated n=28 (%)	Unvaccinated n=26 (%)	
n. (%)	12 (42.9) [§]	6 (23.1)	13 (46.4) [§]
Local reactions	2 (7.1)	0	6 (21.4)
Localised rash (less than 5 vesicles)	1 (3.6)	0	1 (3.6)
Fever ($\geq 37.8^\circ$ C)	3 (10.7)	0	4 (14.3)
Vomiting	0	1 (3.8)	1 (3.6)
Headache	10 (35.7)	5 (19.2)	6 (21.4)
Herpes zoster	0	1 (3.8)	0

[§]Chi-square test. $p=0.1663$ (between vaccinated SLE and vaccinated controls). Some individuals presented more than one adverse event.

Table III. Mean absolute values of lymphocytes and mean percentage (range) of lymphocyte subpopulations in vaccinated and unvaccinated SLE patients and vaccinated controls on the day of vaccination.

Parameters	SLE (n=54)		Vaccinated controls (n=26)	p-value (n=28)
	Vaccinated	Unvaccinated (n=28)		
Lymphocytes (cells/mm ³)	1784.07 ± 740.35	1790.45 ± 895.98	2372.59 ± 742.06	0.005 ^{§*}
CD3+ (cells/mm ³)	1285.66 ± 582.68	1262.00 ± 622.14	1526.65 ± 514.91	0.120
CD4+ (cells/mm ³)	646.46 ± 312.47	635.45 ± 388.98	804.05 ± 295.00	0.214
Naive (%)	40.50 ± 9.59	39.33 ± 11.97	41.71 ± 12.75	0.829
Central Memory (%)	25.01 ± 7.78	24.79 ± 8.66	25.05 ± 7.44	0.995
Peripheral memory (%)	28.39 ± 9.19	29.16 ± 11.12	27.08 ± 10.99	0.702
Terminally differentiated(%)	6.09 ± 3.55	6.70 ± 5.31	6.14 ± 2.21	0.471
CD28+(%)	97.05 ± 4.14	97.98 ± 2.08	96.45 ± 4.35	0.254
CD38 (molecules/cell)	2717.61 ± 1331.12	2529.51 ± 1454.08	2356.21 ± 1249.38	0.504
CD8+	552.73 ± 291.90	528.96 ± 303.11	592.73 ± 229.50	0.661
Naive (%)	35.24 ± 12.51	37.71 ± 15.82	34.52 ± 14.04	0.701
Central Memory (%)	4.18 ± 6.78	3.12 ± 1.67	4.03 ± 2.56	0.073
Peripheral memory (%)	38.35 ± 13.97	39.63 ± 15.04	40.33 ± 14.69	0.996
Terminally differentiated (%)	22.21 ± 11.20	19.52 ± 11.32	21.10 ± 11.51	0.706
CD28+ (%)	68.42 ± 17.76	68.03 ± 16.13	67.57 ± 18.29	0.912
CD38 (molecules/cell)	2304.55 ± 1289.25	2108.04 ± 1374.41	1694.22 ± 837.19	0.378
CD19+ (cells/mm ³)	170.10 ± 117.66	227.01 ± 179.92	286.43 ± 180.07	0.009 ^{†*}
Naive (%)	75.15 ± 11.58	77.21 ± 14.12	77.83 ± 8.10	0.586
Memory (%)	24.84 ± 11.58	22.78 ± 14.12	22.16 ± 8.10	0.530
CD 80 +	27.43 ± 13.07	30.94 ± 17.15	25.30 ± 8.24	0.331
CD 86+	7.4 ± 5.64	10.17 ± 14.19	5.55 ± 4.09	0.107
CD38 (molecules/cell)	3955.60 ± 3124.77	4073.91 ± 4787.03	2966.13 ± 2640.90	0.547
NK cells (cells/mm ³)	130.54 ± 100.58	108.02 ± 111.65	313.13 ± 179.35	<0.0001 ^{†*}

*ANOVA. [†]Tukey test, both vaccinated and unvaccinated SLE patients are different from controls. [‡]SLE vaccinated patients are different from control.

essed on B-cells did not differ among the groups (Table III).

Cellular immune response

On study entry, IFN γ production by CD4 T cells ($p=0.169$) and CD8 T cells ($p=0.158$) was similar in both vaccinated groups (SLE patients and controls). Before VZV vaccine administration, SLE patients and control group had

similar CD4⁺ and CD8⁺ T specific VZV IFN γ producing cells ($p=0.783$ and $p=0.754$, respectively).

Thirty days after vaccination, SLE patients showed a lower percentage of CD4⁺ and CD8⁺ T specific VZV IFN γ producing cells than the control group ($p=0.001$ and $p=0.0004$ respectively). At later dates (180 and 360 days), mean percentage of CD4⁺ and CD8⁺

T specific VZV IFN γ producing cells were similar between SLE and control groups (Figures 1 and 2).

No correlation was found between mean absolute values of CD4 T cells, CD8 T cells before vaccination, use of immunosuppressants or lymphocytes subsets and production of IFN γ 30 days after vaccination.

Humoral immune response

At study entry, all individuals presented protective plasma VZV antibody levels equal to or greater than 0.1 IU/ml. Mean antibody levels were also comparable among the groups ($p=0.191$). Individuals from SLE and control groups did not differ with respect to mean VZV antibodies at each time point analysed. Both vaccinated SLE patients and controls had a significant increase in antibody levels between days 0 and 30 ($p<0.001$ for both analyses) (Fig. 3).

Herpes zoster during the study

Over the mean 35.6 months of follow-up period, we reported 4 cases of HZ in the unvaccinated SLE group, whereas

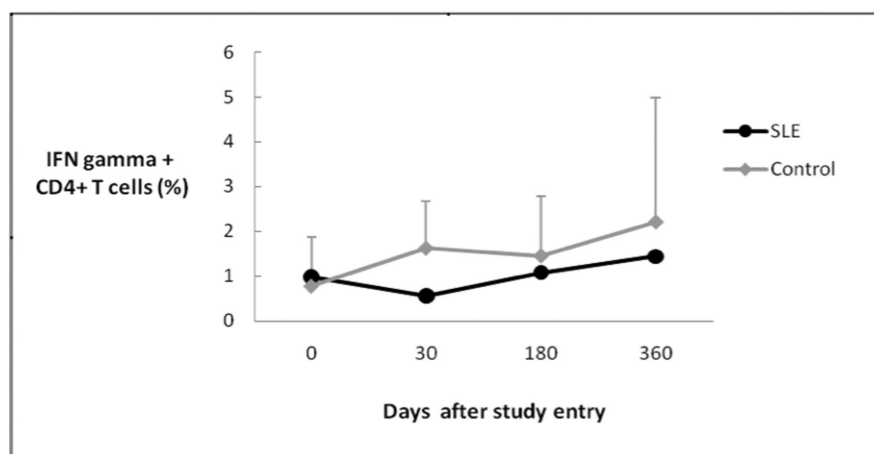


Fig. 1. Percentage of IFN γ producing CD4⁺ T cells after in vitro stimulation with VZV antigen at days 0, 30, 180 and 360 after varicella vaccination in SLE patients and in controls.

in the vaccinated group no case was observed.

It is worth noting, none of those episodes occurred after an increase in immunosuppressive drugs. The relative risk of HZ could not be calculated due to the small number of patients with HZ.

Discussion

To our knowledge, this is the first study that has evaluated the immune response and tolerability of varicella vaccine in SLE patients.

All our patients and healthy controls presented antibodies, although not all of them had reported previous VZV disease. This is probably due to previous oligosymptomatic disease (22). Since most individuals acquire immunity through natural infection in Brazil, VZV seroprevalence among Brazilian adolescents is estimated to be 95.5% (22). VZV vaccine is only available in the Brazilian public health system for susceptible individuals with increased risk of severe disease and for their household contacts.

Although the response to humoral immunity has been present in all patients and controls, it is well known that some conditions can hamper the specific VZV cellular immunity and predispose to zoster (1-5, 23-28).

Following a single dose of varicella vaccine, SLE patients showed an increase in specific VZV antibody levels, similar to healthy controls. We observed a decrease in specific cell-mediated immunity to VZV in SLE patients.

Recently, a number of reviews on the use of vaccines in patients with rheumatic diseases have been published (13-16), but no study on SLE patients and varicella vaccine is available in English literature. In light of the risks, a strict set of inclusion criteria was required for this study. Although SLEDAI score was not an exclusion criterion, all patients presented low SLEDAI scores reflecting the low activity of the disease and the limited use of immunosuppressants. After varicella vaccine administration, only mild adverse reactions were observed, most of them probably not associated with the vaccine. Although the frequency of adverse events was higher in our study population than described

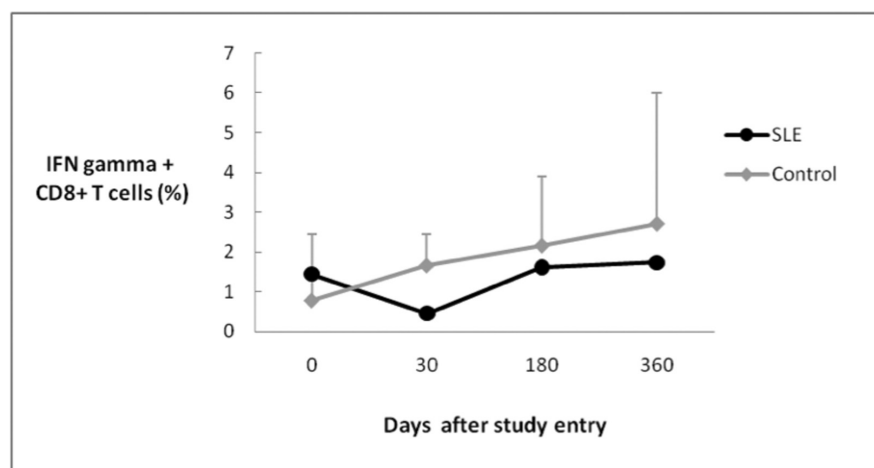


Fig. 2. Percentage of IFN γ producing CD8 $^{+}$ T cells after *in vitro* stimulation with VZV antigen at days 0, 30, 180 and 360 after varicella vaccination in SLE patients and in controls.

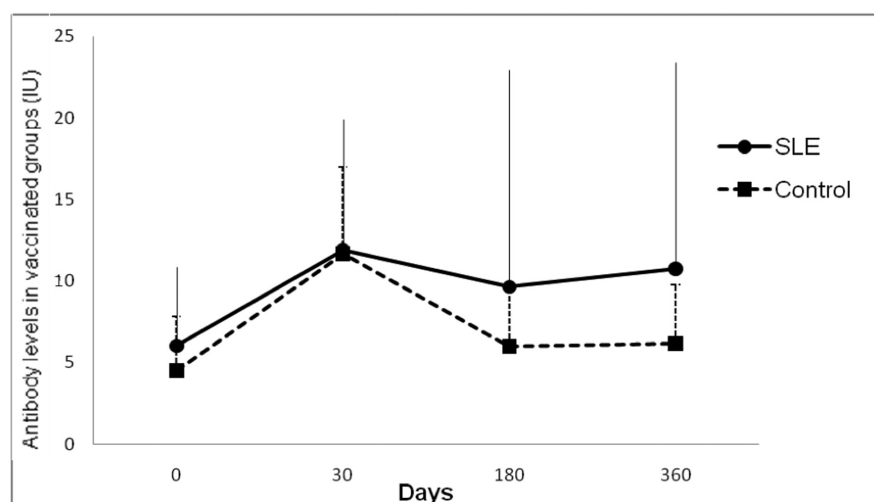


Fig. 3. Mean plasma varicella antibody levels in vaccinated groups (SLE and controls) at days 0, 30, 180 and 360.

in the literature, none were severe (29) and they were equally distributed in the lupus group and in the control group. Moreover, we did not observe lupus flare in the first 30 days after administration. Further flares were not time-related to vaccination and were similar in both SLE groups (vaccinated and unvaccinated), suggesting that the vaccination was not a trigger for new flares. Regarding the immunophenotyping profile, noteworthy distinctions between SLE patients and controls lay on the total lymphocyte, B-cell and NK-cell counts that were lower in SLE patients than in controls. Lupus patients are known to have a low number of NK cells (30-32). In our study, contrary to the literature, the CD4 T cell counts were not lower than in the controls

(33), which could be explained by the low disease activity of our patients. Also, the absence of major CD4/CD8 T alterations suggests that our patients were not at risk to receive a live attenuated vaccine and might be able to respond to this antigen challenge.

Since the values of CD4 T cells are an important parameter to vaccinate other immunosuppressed patients (*e.g.* HIV infected) (34), we tested the possibility of a relationship between the individual absolute values of CD4 and CD8 T cells and the cellular specific response to varicella vaccine, but we did not find any direct relationship.

T cell dysfunction has been described at the cellular and molecular level in both humans and animal models of lupus (35, 36). Little is known on T-cell

response to VZV epitopes, though CD4 T cells may play a greater role in control of VZV infection than they do in some other viral infections (2, 37). Park *et al.* analysed the association of reduced *in vitro* CD4 T cell responses specific to VZV in SLE patients and found that percentage of IFN γ positive CD4 T cells was significantly lower in patients with SLE than in healthy controls (38). Immunologic studies show that HZ high incidence in patients with SLE is probably due to cellular immunity dysfunction rather than to immunosuppressants (2, 5).

All patients and controls had protective VZV antibody levels at study entry. After vaccination, antibody levels increased and remained increased in both vaccinated groups, showing the patients' ability to respond to the vaccine in terms of humoral immunity.

We observed a decrease in the IFN γ production specific to varicella, which is still unclear from our point of view. Impaired *in vitro* T-cell proliferation and altered cytokine production in response to foreign antigens, alloantigens, and mitogens were observed by others (36). It is possible that the response had occurred before the 30-day evaluation and so remaining undetectable to us. It is worth mentioning that IFN γ is intrinsically related to the pathogenesis of SLE and therefore it may interfere with specific responses to VZV. We cannot rule out the possibility that a less expressive cell-mediated immune response that occurred in our study was due to low antigenic stimuli. One study indicated that higher potencies are required to elicit a significant increase in cell-mediated immunity to VZV (7).

Although *in vitro* IFN γ production in response to antigen exposure is considered a marker of cellular function, a remarkable clinical response in terms of the absence of zoster episodes among vaccinated SLE patients was observed. None of the SLE patients from the vaccinated group manifested HZ, whereas 4 patients from the unvaccinated group presented the infection. So it is possible that the varicella vaccine protected the vaccinated individuals. The same finding was described by Gershon *et al.*, while administering varicella vaccine

to 46 HIV-infected children who had previously had varicella (10). For individuals with previous VZV infection, the administration of a single dose of varicella vaccine or even herpes-zoster vaccine would act as a booster. Re-exposure to a pathogen could generate more T-cells that can localise in peripheral tissues (39, 40).

We assume that the high incidence of HZ in SLE patients cannot be explained merely by a low frequency of VZV-specific IFN γ CD4⁺ T cells; perhaps more complex genetic and environmental factors related to the pathogenesis of SLE or host immunomodulation from interfering drugs might help to explain the high incidence rate.

It is a fact that our sample size was small, which could possibly have interfered with the statistical significance. However, the rare occurrence of the disease and the limited number of enrolled patients due to the strict inclusion criteria, are reasons to justify such a small number of individuals. Also, one could suggest the use of a more immunogenic vaccine. As we were unaware of the risks of vaccinating our patients with a varicella vaccine containing 1000 PFU, we could not for safety reasons vaccinate them with a higher virus dosage. Interestingly, although the zoster vaccine is a live vaccine with a higher potency than varicella vaccine, some are considering the risk:benefit ratio for this vaccine for patients on a low dose of immunosuppressant, especially since rescue therapy with acyclovir is possible (39).

Our results allow us to conclude that in this group of SLE patients with pre-existing immunity to varicella, with low disease activity and taking low doses of immunosuppressants, the varicella vaccine was safe.

Although more studies reproducing our results are necessary, administering varicella booster vaccines to SLE patients might be a safe strategy to help to reduce the incidence of HZ among this very special group of individuals.

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