Effects of long-term cyclic iloprost therapy in systemic sclerosis with Raynaud's phenomenon. A randomized, controlled study

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Abstract

Objective

Iloprost is a stable prostacyclin analogue which has been shown to be effective in the short-term symptomatic treatment of Raynaud's phenomenon (RP) secondary to systemic sclerosis (SSc). The aim of this study was to evaluate the effects of long-term cyclic therapy with iloprost in comparison with nifedipine on the skin score, pulmonary function and Raynaud's severity score in patients with SSc and RP.

Methods

We conducted a 12-month prospective, randomised, parallel-group, blind-observer trial to compare the effects of intravenously infused iloprost (2 ng/kg/min on 5 consecutive days over a period of 8 hours/day and subsequently for 8 hours on one day every 6 weeks) with those of conventional vasodilating therapy with nifedipine (40 mg/day per os) in 46 patients with SSc and RP.

Results

At 12 months, iloprost but not nifedipine reduced the skin score (iloprost: from 13.26 ± 2.05 to 9.26 ± 1.32, p = 0.002; nifedipine: from 10.83± 2.09 to 12.17± 3.02, p = n.s.; iloprost vs nifedipine: p = 0.016) and the RP severity score (iloprost: from 2.17 ± 0.2 to 1.22 ± 0.13, p = 0.02 vs baseline; nifedipine: from 2.08 ± 0.34 to 1.33 ± 0.22, p = n.s.). Carbon monoxide diffusing capacity (DLCO), expressed as % of the predicted normal value, worsened significantly in the nifedipine group (from 69.6 ± 7.4% to 61.5 ± 6.5%, p = 0.044) and remained stable in patients treated with iloprost (from 53.2 ± 4.8 to 56.0 ± 4.6%, iloprost vs nifedipine: p = 0.026).

Conclusion

In SSc patients, cyclic intravenous iloprost infusion is able to control vasospastic disease. Our results suggest that it might also act as a disease-modifying agent, as it seems to improve the course of the disease. Further studies principally focused on organ involvement and the natural history of the disease are needed to confirm our results.

Key words

Systemic sclerosis, Raynaud's phenomenon, iloprost, controlled clinical trial.

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Introduction

Systemic sclerosis (SSc) is a multi-system disease of unknown aetiology characterised by progressive fibrotic changes of skin and internal organs and by characteristic involvement of the arterial and microvascular beds (1). There is evidence to suggest that the immune system, endothelial cells and platelets all play pivotal roles in the pathogenesis of SSc by enhancing the proliferation of fibroblasts and the synthesis of collagen and extracellular matrix (2, 3).

Severe Raynaud's phenomenon (RP) is the early onset symptom in most SSc patients. Iloprost, a stable prostacyclin analogue with vasodilating, anti-platelet, cytoprotective and immunomodulating properties (4-6), has been found to be an efficacious alternative to nifedipine for the treatment of RP in scleroderma patients (7-11). Iloprost's beneficial effects are sustained for far longer than its short half-life would predict (12, 13) and there are data showing significant and long-lasting effects at the level of cutaneous microcirculation (14) and of the resistance index of renal vessels in SSc patients (15). These observations raise the possibility that iloprost might be a disease-modifying agent for SSc. However, this hypothesis has not been extensively tested so far, since long term trials with cyclic iloprost are few and have been conducted on limited SSc series (6, 15-17). In this study we carried out a 12-month, randomised, between-patient, blind-observer study, with the aim of evaluating the effects of iloprost in comparison with nifedipine in patients with SSc and secondary RP, on cutaneous involvement (primary end-point), pulmonary function and Raynaud's symptoms (secondary end-points). Our results show that both iloprost and nifedipine are able to improve vasospastic disease. Indeed, in SSc patients, iloprost but not nifedipine seems to improve skin fibrosis and stabilize lung CO diffusion capacity.

Materials and methods

Patients

SSc with secondary RP was the main inclusion criteria for the study. The diagnosis of SSc was made according to Le Roy *et al*. (18). The diagnosis of RP was based on a history of episodic digital pallor and cyanosis and was confirmed by Doppler flowmetry and the nitroglycerin test (19). All patients were regularly followed at our outpatient clinic. Women of childbearing age were required to practice a medically acceptable method of birth control. Criteria for exclusion were: age < 18 yr, pregnancy, acute myocardial infarction or stroke in the previous 4 months, serious cardiac failure and hypertension, ischemic cardiopathy, hypotension (systolic blood pressure < 100 mmHg), hemorrhagic diathesis and/or thrombocytopenia (< 80,000/ mm^2), thrombocythemia (> 500,000/ mm^2), anticoagulant therapy, hyperviscosity syndrome, peripheral obstructive arterial disease, cryoglobulinaemia, chronic acrocyanosis (secondary to other diseases than Raynaud's phenomenon), other connective tissue diseases, systemic vasculitis, chronic hepatopathy (AST/ ALT > 30% of the upper normal limit), diabetes mellitus, renal failure, previous history of intolerance to nifedipine or iloprost. Patients with SSc sine scleroderma were also excluded. None of the patients were taking or had taken penicillamine and/or immunosuppressants.

All the patients were informed about the nature and aim of the study and gave their consent to participate. Patients were told that they were free to withdraw from the study at any time without giving a reason. The study protocol was approved by the Regional Control Authority.

Patients were required to discontinue anti-platelet drugs, calcium channel blockers, and other vasodilators at least 30 days before inclusion in the study. All patients were non-smokers.

Treatment regimens

Iloprost (Endoprost®, Italfarmaco, Milan, Italy) was given by intravenous infusion (0.1 mg in 500 ml of saline: solution of 200 ng of iloprost/ml; Terumo STC 503 infusion pump) on 5 consecutive days over a period of 8 hours/ day. On the first 3 days the treatment was started at 10 ml/h and the dose was

increased every 30 minutes by 10 ml/h up to the maximal tolerated dose (but not to exceed 2.0 ng/kg/min). The higher dose administered on the third day of infusion was then repeated on the last 2 days, and subsequently for 8 hours on one day every 6 weeks. The dose was to be maintained or reduced during the infusion, depending on the tolerance of the patients to the possible side effects usually associated with the infusion.

Oral nifedipine (Adalat AR, Bayer SpA, Milan, Italy), 20 mg, was given in a slow-release formulation twice daily, every day. Treatment duration was 12 months.

Study design

Patients were stratified according to their skin involvement (limited or diffuse cutaneous variants) and then assigned to iloprost or nifedipine treatments according to a randomisation table, in a prospective, randomised, between-patient, blind observer study, with the iloprost to nifedipine ratio 2:1. This ratio was based on ethical considerations, in light of the better results of iloprost treatment on RP reported by other investigators (11-13). Follow-up visits were scheduled every 6 weeks.

Clinical outcome variables

The primary end-point was the skin score. Secondary endpoints were pulmonary function tests and the RP severity score.

Clinical evaluation: Initial assessment (T0) was performed within 30 days before the treatments were started and consisted of a complete clinical history and physical examination with detailed evaluation of the extent and severity of skin thickening and RP as described below. A search for evidence of visceral disease was done in all patients and included chest roentgenogram, pulmonary function testing, electrocardiogram, oesophageal manometry, urinalysis and measurement of serum creatinine levels. At the follow-up visits (every 6 weeks) patients generally had a detailed clinical examination and RP evaluation. At 6 and 12 months a detailed skin examination was performed. A complete clinical examination was repeated within 30 days after the end (T12) of the study or when clinically indicated. Patients were examined by two evaluator physicians who were blinded to the therapeutic randomisation. The evaluator physicians were the same individuals throughout the study, did not have access to the study data, and had at least 3 years of experience working in our sceroderma clinic.

Skin thickening: The modified Rodnan skin score technique used (16) consisted of an evaluation of the patient's skin thickness rated by clinical palpation using a 0-3 scale (0= normal skin thickness; 1= mild skin thickness; 2= moderate skin thickness; and 3= severe skin thickness with inability to pinch the skin into a fold) for each of 17 surface anatomic areas of the body: face, anterior chest, abdomen, and (right and left separately) the fingers, forearms, upper arms, thighs, lower legs, and the dorsum of the hands and feet. These individual values were added and the sum was defined as the total skin score.

Pulmonary function: Pulmonary function was evaluated by means of chest radiographs and a pulmonary function test. Lung volumes (vital capacity inspiratory [VCIN], forced expiratory volume in 1 second [FEV1]) and carbon monoxide diffusing capacity using the single breath method (DLCO) were evaluated as a percent of the predicted normal value (17) by body plethismograph Jaeger Body Screen and Transfer Screen Jaeger, respectively. For these parameters the following definitions were applied considering percent changes in comparison with the baseline evaluation: improved if >10% increase; stable if < 10% increase or < 10% decrease; worsened if > 10% decrease.

RP assessment: The parameter considered for efficacy was the RP severity score (13) which was expressed as a mean score considering pain, numbness, burning, throbbing and impaired hand function (each evaluated on a 4-point scale, with 0 = none, 1 = mild, 2 = moderate, 3 = severe, 4 = incapacitating). Data were recorded by patients on diary cards which were collected at each scheduled visit.

Digital ischemic lesions were also counted at entry, during the follow-up visits and at the end of the study.

Statistical analysis

All of the results are expressed as the mean ± standard error of the mean (x ± SEM). Non-parametric tests (Wilcoxon test for paired data and Mann-Whitney U test) were used to evaluate the RP severity score. All the other data were analysed statistically by means of "split-plot" ANOVA with correction for repeated measurements, the factors being the patients, treatments and times. The Fisher's exact test was used as appropriate. A p value < 0.05 was considered to indicate statistical significance.

Results

Forty-six patients were enrolled in the study: 29 were assigned randomly to take iloprost and 17 to take nifedipine. Table I shows the demographic and clinical characteristics of the two groups.

Eleven patients withdrew from the study: 6 on iloprost and 5 on nifedipine. In the iloprost group, the reasons for withdrawal were: lack of compliance (3 pts), scleroderma renal crisis (1 pt), interstitial pneumonia (1 pt), and acute myocardial infarction (1 pt); in the 3 latter cases, the complications were not related in time to the drug infusion and the patient who suffered a renal crisis had been given only the basal cycle. In the nifedipine group, the withdrawals were due to intolerance.

Skin involvement

As shown in Figure 1, iloprost but not nifedipine significantly reduced the skin score values. The reduction was statistically significant after the first 6 months of therapy (iloprost: from 14.15 ± 2.34 to 10.81 ± 1.9, p < 0.; nifedipine: from 10.69 ± 1.93 to 11.00 ± 2.43; between treatments: p = 0.009) and was maintained after 12 months (iloprost: 13.26 ± 2.05 vs 9.26 ± 1.32, p = 0.002 vs. baseline; nifedipine: 10.83 ± 2.09 vs 12.17 ± 3.02; between treatments p = 0.016). In both groups no correlation was observed between the last skin score value and the degree of skin softening.

Pulmonary involvement

No changes from baseline chest roent-

Table I. Demographic and clinical features of patients.

	Iloprost (n = 29)	Nifedipine (n = 17)	p
Female (%)	76	100	< 0.04
Age (yrs)*	51 (25-75)	56 (32-74)	n.s.
Disease duration (yrs)*	5 (1-19)	6 (1-29)	n.s.
RP^ duration (yrs)*	9.5 (1-26)	11 (1-30)	n.s.
dSSc† (%)	48	35	n.s.
Skin score*	14 (1-50)	9 (1-24)	n.s.
Esophageal involvement (%)	93	94	n.s.
Restrictive pattern @ (%)	41	24	n.s.
Pericarditis (%)	79	65	n.s.
Anti-topo I (%)	66	41	n.s.
Anti-centromere (%)	21	29	n.s.

^Raynaud's phenomenon; *median value (range); @VCIN < 80% of the predicted value with normal FEV1%/VCIN%; † diffuse cutaneous systemic sclerosis.

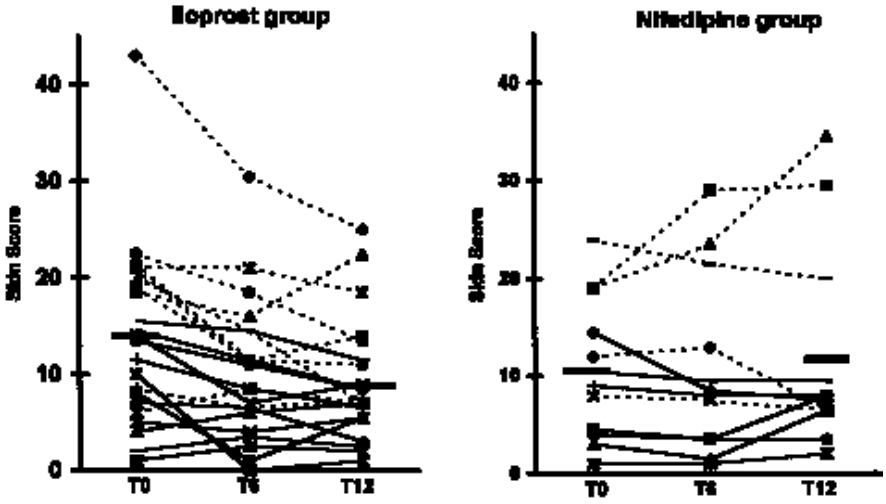

Fig. 1. Individual skin scores in SSc patients, before (T0) and after 6 (T6) or 12 (T12) 6 months of treatment with iloprost or nifedipine.

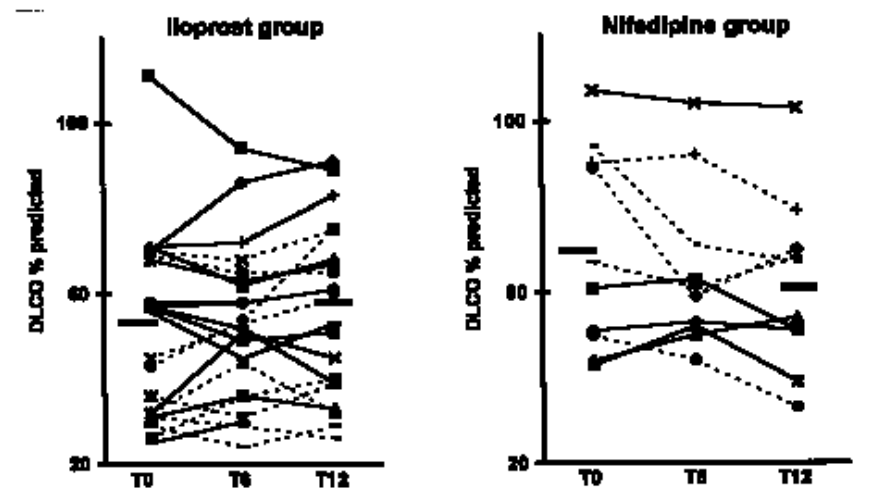

Fig. 2. sbDLCO (as percent of predicted value) in SSc patients before (T0) and after 6 (T6) or 12 (T12) 6 months of treatment with iloprost or nifedipine.

genograms was observed at the end of the study in any of the patients. After 12 months VCIN and FEV1 values were not modified by either treatment (VCIN: iloprost [n = 23]: from 81.2 ± 5.2% to 82.1 ± 4.9%; nifedipine [n = 11]: from 94.6 ± 5.4% to 95.7 ± 5.8%. FEV1: iloprost [n = 22]: from 82.4 ± 3.9% to 82.7 ± 4.3%; nifedipine [n = 11]: from 96.1 ± 5.9% to 95.9 ± 5.02%). DLCO significantly worsened in the nifedipine group ([n = 10]: from 69.6 ± 7.4% to 61.5 ± 6.5%, p = 0.044), on the contrary and remained stable in iloprost-treated patients ([n = 20] from 53.2 ± 4.8% to 56.0 ± 4.6%, p = 0.026 vs. nifedipine) (Fig. 2). Individually, 18 of 20 patients treated with iloprost and 5 of 10 patients treated with nifedipine showed an improvement or stability of DLCO (p < 0.05, Fisher's exact test).

Raynaud's phenomenon

Treatment with iloprost significantly decreased the RP severity score with nifedipine obtaining a trend close to statistical significance (iloprost: from 2.17 ± 0.2 to 1.22 ± 0.13, p < 0.02 vs. baseline; nifedipine: from 2.08 ± 0.34 to 1.33 ± 0.22, p = 0.054 vs. baseline). At entry into the study, 14 patients in the iloprost group and 3 in the nifedipine group had digital ischemic lesions; all of them took part in the entire study. Among these, all the patients treated with nifedipine (3/3) and 12 (out of 14) of those treated with iloprost had improved, after 12 months. No statistical analysis was performed, because of the small number of the subjects.

Skin and pulmonary involvement in patients with recent onset disease

In this study, 5 patients in the iloprost group and 3 patients in the nifedipine group had been affected by SSc for no more than 2 years. In this subgroup of patients as well, iloprost but not nifedipine significantly reduced skin score values and was shown to stabilise the CO lung diffusion capacity (Table II).

Adverse reactions

Table III shows the frequency of adverse effects in the two groups. Hypotension, nausea or vomiting required a temporary dose reduction in individual

Table II. Variation of skin scores and of %DLCO in SSc patient with a disease duration (2 years.

		T0 x ± s.e.	T6 x ± s.e.	T12 x ± s.e.
Total skin score*	iloprost (n=5)	19.2 ± 8.1	14.6 ± 7.2	7.0 ± 0.6
	nifedipine (n=3)	17.5 ± 1.5	20.3 ± 6.1	23.8 ± 8.3
DLCO % **	iloprost (n=5)	50.0 ± 6.9	49.0 ± 5.6	47.2 ± 7.7
	nifedipine (n=3)	80 ± 13.5	66.0 ± 5.0	67 ± 0.5

Iloprost vs nifedipine *p < 0.002; **p < 0.05.

Table III. Adverse reactions to study treatments.

Iloprost (n = 29)	%	Nifedipine (n = 17)	%
Headache	100	Hypotension	29
Nausea-vomiting	83	Headache	18
Jaw pain	69	Headache	6
Myalgia	34	Tachycardia	6
Diarrhoea	28		
Chills	17		
Hypotension	14		
Arrhythima	7		
Hyperkinesia	3		

cases during iloprost infusion and/ or adequate pre-medication. Side effects disappeared immediately after the end of the infusion.

In the nifedipine group 3 patients withdrew because of headache and hypotension, one because of hypotension and one for ankle oedema.

Discussion

This was a long term, randomised controlled study of iloprost versus nifedipine in systemic sclerosis and secondary Raynaud's phenomenon carried out with the aim of evaluating the possible effects of treatments on the natural history of the disease. Both iloprost and nifedipine were able to significantly reduce RP symptoms, with no differences between treatments. It has been suggested that the symptomatic relief of RP in iloprost-treated patients could be mainly due to long lasting improvement of the skin nutritional blood flow, probably through normalization of the dysfunctional arterio-venular anastomoses as demonstrated by computer-assisted video capillaroscopy (14). However, no data have been provided regarding the possible correlation between the significant improvement in nutritional skin flow and possible changes in the skin score.

Our data show that in iloprost-treated patients there was dramatic and highly significant skin softening, as demonstrated by the impressive improvement in the skin score. In most patients, this change had appeared by the end of the third or fourth iloprost infusion cycle and was already statistically significant within 6 months of the start of treatment. The natural history of skin involvement in SSc has been described as occurring in 3 clinical phases (22): an initial period of progressive thickening (phase I), followed by a variable period of minimal change or plateau (phase II), and then by a period of softening or thinning (phase III). The patients included in this study cover a large range of ages. Therefore it is possible that the improvement we observed in the iloprost-treated patients could have been due to the natural evolution of the disease. We believe that this was not the case, however, since the same marked improvement in skin fibrosis is observed in the subgroup of patients with disease lasting ≤ 2 years, such patients being an optimal group in which to test the effectiveness of a disease-modifying treatment (23). Furthermore, a worsening of skin thickness rather than skin softening was observed in the nifedipine-treated patients.

Recently Fillaci *et al.* reported that the combination of cyclosporinA plus iloprost, but not iloprost alone, resulted in a significant improvement in skin fibrosis in SSc patients (16). We used a different technique to evaluate the total skin score, but do not believe that this difference is sufficient to explain the conflicting results, since both the plicometer skin test (24) and the modified Rodnan skin score (25) are reliable measures of skin fibrosis. Indeed, a noticeable, if not significant, improvement in the skin scores was observed by Filaci *et al.* in their iloprost-treated patients.

It has previously been reported that nifedipine (19), but not iloprost (20),is effective in increasing the diffusing capacity for carbon monoxide in SSc patients. This improvement was suggested to be mainly due to a sustained increase in the lung capillary volume induced by the chronic administration of nifedipine (19), but not by intermittent iloprost infusion. In our study, DLCO significantly worsened in the nifedipine group, whereas the majority of patients treated with iloprost had a stable or increased DLCO as evidenced by a mean trend toward mild improvement. We feel that an increase in lung capillary volume does not account for our results. The different trends observed between the two treatments might be better explained by a substantial slowing and reduction in lung fibrosis in the iloprost group. Whether prolonged or more intensive treatment might result in more pronounced improvement remains to be investigated. The lack of concomitant results in VCIN can be explained by the lower sensitivity of VCIN than DLCO in the detection of early modifications of pulmonary function. In their study Filaci *et al.* (16) were not able to demonstrate any improvement in lung function by either iloprost alone or iloprost coupled to CyA. However, in considering lung parameters they scored the DLCO and

VCIN data together, probably reducing the sensitivity of the analysis.

Whether and how iloprost exerts a global anti-fibrotic action in SSc patients remains to be confirmed. In agreement with previously suggested hypotheses, this effect might be at least partially due to a cytoprotective effect on endothelial cells, thereby breaking the vicious cycle of vascular injury, platelet aggregation and release of soluble factors that stimulate fibroblast proliferation and synthetic activity (28, 29). Indeed, it has been recently demonstrated that iloprost inhibits the *in vitro* production of the fibrogenetic lymphokines IL-1 and IL-6 by *in vitro*-activated human peripheral blood cells (5). Furthermore, long-term cyclic iloprost infusion but not continuous nifedipine treatment was able to decrease the *ex vivo* production of both IL-1 and TNF-alpha by patients' peripheral blood mononuclear cells. This decrease seemed to show a significant correlation with skin score improvement (6).

The adverse reactions encountered in our study are similar to those reported in other studies of iloprost (9). The side effects to the pharmacological action of iloprost were limited to the period of drug infusion and tended to be reduced immediately after dose reduction. The low prevalence of side effects to nifedipine could be partially due to the exclusion from the study of patients intolerant to this drug, widely and routinely used for SSc.

We feel that the main limitation of this study was the lack of blindness in its design. However, on the one hand it would have been almost impossible, from both the ethical and practical point of views, to achieve blindness for such a long period of time. On the other hand, the use of the same blinded observers should have reduced or eliminated this possible bias.

Furthermore, the conservative statistical approach with correction for multiple testing and the reduced number of study end-points should have reduced the possibility of false positive conclusions. On the contrary, the possibility of false negative results cannot be excluded. To this extent our results with regard to some of the lung function parameters probably deserve further specific investigation. The dose of nifedipine used in our study might also be open to some criticism. However, RP symptom control was almost equivalent in the two study groups, thus supporting the validity of the nifedipine dose used in our study.

References

1. SILVERRM: Clinical aspects of systemic sclerosis (scleroderma). *Ann Rheum Dis* 1991; 50: 854-61.
2. FURST DE: The endothelium in the pathogenesis of systemic sclerosis:is it primary or secondary? *J Malad Vasc* 1999; 24: 95-8.
3. STREHLOW D, KORN JH: Biology of the scleroderma fibroblast. *Curr Opin Rheumatol* 1998; 10: 572-8.
4. BERGMAN G, KIFF PS, ATKINSON L, KERKEZ S, JEWITT DE: Dissociation of platelet aggregation and vasodilatation with iloprost: a stable orally active, prostacyclin derivative. *Circulation* 1983; 68: 398.
5. DELLA BELLA S, MOLTENI M, COMPASSO S, ZULIAN C, VANOLI M, SCORZA R: Differential effects of cyclo-oxygenase pathway metabolites on cytokine production by T lymphocytes. *Prostaglandins Leukot Essent Fatty Acids* 1997; 56:177-84.
6. DELLA BELLA S, MOLTENI M, MASCAGNI B, ZULIAN C, COMPASSO S, SCORZA R: Cytokine production in sclerodermia patients: effects of therapy with either iloprost or nifedipine. *Clin Exp Rheumatol* 1997; 15:135-41.
7. RADEMAKER M, COOKE ED, ALMOST NE *et al.*: Comparison of intravenous infusion of iloprost and oral nifedipine in treatment of Raynaud's phenomenon in patients with systemic sclerosis: a double blind randomized study. *Br Med J* 1989; 298: 564-7.
8. WATSON HR, BELCHER G: Retrospective comparison of iloprost with other treatments for secondary Raynaud's phenomenon. *Ann Rheum Dis* 1991; 50: 359-61.
9. WIGLEY FM, SEIBOLD JR, WISE RA, MCCLOSKEY DA,DOLE WP: Intravenous iloprost treatment of Raynaud's phenomenon and ischemic ulcers secondary to systemic sclerosis. *J Rheumatol* 1992; 19: 1407-14.
10. TORLEY HI, MADHOK R, CAPELL HA *et al.*: A double blind, randomised,multicentre, comparison of two doses of intravenous iloprost in the treatment of Rynaud's phenomenon secondary to connective tissue diseases. *Ann Rheum Dis* 1991; 50: 800-4.
11. COOKE ED, KIRBY JDT, MANT TGK: Double-blind comparison of three days infusion of iloprost and continuous oral nifedipine on symptomatology and digital blood flow in patients with Raynaud's phenomenon secondary to systemic sclerosis. *Angiologie* 1988: 555-7.
12. YARDUMIAN DA, ISEMBERG DA, RUSTIN M *et al.*:Successful treatment of Raynaud's syndrome with iloprost, a chemically stable prostacyclin analogue. *Br J Rheumatol* 1988; 27: 220-6.
13. RADEMAKER M, THOMAS RHM, PROVOST G, BEACHAM JA, COOKE ED, KIRBY JDT: Prolonged increase in digital blood flow following iloprost infusion in patients with systemic sclerosis. *Postgrad Med J* 1987; 63: 617-20.
14. ARPAIA G, CIMMINIELLO C, SARDINA M *et al.*: Microcirculatory effects of Iloprost in patients with suspected secondary Raynaud phenomenon. *Vasc Surg* 1995; 29: 37-42.
15. SCORZA R, RIVOLTA R, MASCAGNI B *et al.*: Effect of Iloprost infusion on the resistance index of renal vessels of patients with systemic sclerosis. *J Rheumatol* 1997; 24: 1944-8.
16. FILLACI G, CUTOLO M, SCUDETTI M *et al.*: Cyclosporin A and iloprost treatment of systemic sclerosis: clinical results and interleukin-6 serum chenges after 12 months of therapy. *Rheumatology* 1999; 38: 992-6.
17. DE LA MATA J, GOMEZ-SANCHEZ MA, ARANZANA M, GOMEZ-REINO JJ: Long-term iloprost infusion therapy for severe pulmonary hypertension in patients with connective tissue diseases. *Arthritis Rheum* 1994; 37: 1528-33.
18. LE ROY EC, BLACK C, FLEISCHMAJER R *et al.*: Scleroderma (systemic sclerosis): classification, subsets and pathogenesis. *J Rheumatol* 1988; 15: 202-5
19. MONTORSI W, ANNONI F, TAJANA A: La velocimetria doppler nella diagnostica arteriosa dell'arto superiore. *Min Chir* 1982; 37: 1771-4.
20. BRENNAN P, SILMAN A,BLACK C *et al.*:Reliability of skin involvement measures in scleroderma. *Br J Rheumatol* 1992; 31: 457-60.
21. QUANJER PH: Standardized lung function testing: Report from working party on "standardisation of lung function test", European Community for Coal and Steel. *Bull Europ Physiopathol Repir* 1983; 19: 7-10.
22. STEEN VD, MEDSGER JR TA: Epidemiology and natural history of systemic sclerosis. *Rheum Dis Clin North Am* 1990; 16: 1-10.
23. WHITE B, BAUER EA,GOLDSMITH LA *et al.*: Guidelines for clinical trials in systemic sclerosis (scleroderma). I. Disease-modifying interventions. *Arthritis Rheum* 1995; 38: 351-60.
24. PARODI MN, CASTAGNETO C, FILACI G *et al.*: Plicometer skin test:A new technique for the evaluation of cutaneous involvement in systemic sclerosis. *Br J Rheumatol* 1997; 36: 244-50.
25. FURST DE, CLEMENTS PJ, STEEN VD *et al.*: The modified Rodnan skin score is an accurate reflection of skin biopsy thickness in systemic sclerosis. *J Rheumatol* 1998; 25:184-8.
26. SFIKAKIS PP, KYRIAKIDIS M, VERGOS C *et al.*: Diffusing capacity of the lung and nifedipine in systemic sclerosis. *Arthritis Rheum* 1990; 33: 1634-9.
27. THURM CA, WIGLEY FM, DOLE WP, WISE RA: Failure of vasodilator infusion to alter pulmonary diffusing capacity in systemic sclerosis. *Am J Med* 1991; 90: 547-52.
28. GOODWIN JS, CEUPPENS J: Regulation of the immune response by prostaglandins. *J Clin Immunol* 1983; 3: 295-315.
29. GOODWIN JS: Immunomodulation by eicosanoids and anti-inflammatory drugs. *Curr Op Immunol* 1989; 2: 264-8.