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Evaluation of Clinical Characteristics and Biomarkers after Endovenous Foam Sclerotherapy for Venous Disorders

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Abstract

Objective: Endovenous foam sclerotherapy (EFS) is widely performed in the U.S, but there is a paucity of studies evaluating clinical predictors of outcomes, including biomarkers, in patients with venous disorders.

Methods: Patients undergoing EFS monotherapy for venous disorders were enrolled. Evaluation at baseline, 1 week, 12 weeks, and 26 weeks included clinical characteristics and biomarker analysis.

Results: 100 patients with venous disease were treated. At one week follow-up, 44% underwent a second injection. At 3 months, 100% of patients had obliteration of at least 80% of their affected veins, and 96% reported improved venous stasis symptoms. Adverse events were minor and deep vein thrombosis was found 4 patients at 3 months. D-dimer levels were significantly higher at week one, but returned to baseline by week 12; fibrin monomer decreased and PPL increased at one week and 3 months relative to baseline.

Conclusion: EFS monotherapy is effective in treating signs and symptoms of venous disease with few adverse effects. D-dimer levels are significantly associated with obliteration of venous segments suggesting an association between vein obliteration and activation of coagulation.

Keywords: Varicose veins; Monotherapy; Injection; Thrombosis; Microparticles

Introduction

Varicose veins are the most common manifestation of chronic venous disease affecting 25-33% of adult women and 10-20% of adult men [1]. Varicose veins pose more than just a cosmetic problem. More commonly, they produce symptoms of heaviness, fatigue, pain, swelling, restlessness, burning, and itching [2]. Varicose veins are also associated with a number of serious complications including bleeding, superficial thrombophlebitis, deep-vein thrombosis and ulceration [2].

Treatment modalities for management of varicose veins include conservative measures (compression, diet, exercise, elevation, skin hygiene, medications), endovenous/interventional therapies (sclerotherapy via laser, radiofrequency ablation, or chemical), and surgical interventions (ligation, stripping, microphlebectomy) [2]. If conservative management fails, the patient is a candidate for more invasive treatment. While surgical ligation and stripping is effective, complications such as infection, hematoma, and nerve paresis can occur [3]. The advent of endovenous sclerotherapy (EFS) has revolutionized the treatment of varicose veins. Worldwide, EFS may be the most widely used procedure for the treatment of varicose veins.

Multiple studies have demonstrated the effectiveness of endovenous sclerotherapy in immediate obliteration of varicose veins without compromise of patient safety [4]. However, despite widespread use, there is a paucity of knowledge related to the systemic and thrombotic response to EFS and few clinical predictors of success, quality of life or adverse outcome after the procedure.

D-dimer and fibrin monomer represent activation of coagulation [5]. It has been found in preliminary studies that D-dimer is increased post EFS and suggested that this activation in coagulation plays a central role in sclerotherapy efficacy, but also may be associated with thrombosis adverse effects [6].

Platelet microparticles (PPL) are released when the platelet membrane is disrupted. Microparticles possess procoagulant phospholipid activity and may be associated with the risk of thrombosis [7]. Microparticles may also indicate ongoing cell apoptosis. To date, assay technology to measure PPL remains a research tool, but PPL is being investigated as a marker of venous thrombosis especially in patients receiving chemotherapy [8].

The purpose of this prospective cohort study was to collect clinical and coagulation biomarker data related to the performance of EFS among adult patients with symptomatic venous disease from varicose veins. This information was used to assess associations between clinical characteristics and procedure outcomes.

Methods

The study was a prospective cohort study of patients who presented with symptomatic varicose veins or venous disorders that underwent EFS over a 2 year period. Patients who had a history of DVT were not

excluded. There were no deviations in usual clinical assessment and care of patients seeking varicose vein treatment related to the study. The purpose of this protocol was to collect clinical and biomarker information related to this standard treatment.

Study site

The study was performed at the University of Oklahoma Health Sciences Center campus at the OU Vascular Center. The study was approved by the OUHSC Institutional Review Board. The patients provided written informed consent and were enrolled over the period 2009 through 2011. Follow up was completed in May 2012.

Baseline assessment

Each patient had the following baseline information collected: demographic characteristics, indication for endovenous sclerotherapy, medical history, varicose vein mapping study, reflux testing using the Valsalva and calf compression maneuvers, assessment of severity of venous disease using the Villalta score [9] and quality of life (QOL) questionnaire. Two QOL questionnaires were administered: the general short-form SF-36 questionnaire evaluating general quality of life and the VEINES questionnaire [10] specific to patients with venous disease. CEAP classification was also performed in most patients.

Procedure information

Endovenous sclerotherapy was performed as per usual clinical protocol. Procedural characteristics were documented including the volume of sclerosant used, and total number of injections required. Both truncal and axial veins were elgible to be treated with foam including the great saphenous vein. Patients were observed for 20 to 30 minutes post procedure and any immediate adverse events were recorded during this time. Sodium tetradecyl sulfate 1% or 3% was the sclerosant used in all patients and only one injection was performed per session for the majority of patients. Foam was created at the bedside using the Tessari method using a 1:4 sclerosant to room air ratio. After injection, all patients were placed in a 30-40 mmHg thigh length compression hose and instructed to wear this 24 hours a day for 5 days and then daily until the follow-up visit. These instructions were repeated after each subsequent injection.

Blood collection and analysis

A blood sample (4 ml in blue top citrated tube) was taken immediately prior to the first sclerosant injection and before each subsequent injection if additional injections were required. At each follow-up visit, a blood sample was obtained for measurement of D-dimer, fibrin monomer, and total microparticles. The blood samples were processed, frozen and stored and then analyzed at the completion of the 6 month follow-up period. D-dimer fibrin monomer were analyzed per company instructions (Diagnostica Stago, Inc. STA-Liatest D-di and STA-Liatest FM). Total microparticles (PPL) were also analyzed according to company instructions (Diagnostica Stago, Inc. Procoag-PPL) as a clot-based assay that has good correlation compared to phospholipid-positiive microparticle events assessed by annexin binding/flow cytometry.

Follow-up

Patients were routinely assessed at 1 week post sclerotherapy procedure, 12 weeks post procedure, and 26 weeks post procedure. At each visit, the patient was assessed clinically for improvement in signs and symptoms of varicose veins, and underwent a bedside ultrasound evaluation to document completeness of varicose vein sclerosis and presence of any thrombosis as per routine care. Obliteration of the vein was classified subjectively as complete (all of affected vein closed), >80%, 50-79%, or minimal (<50%) estimated by closure of the linear length of the treated vein. Any adverse effects the patient experienced were documented as well as interval medical history. In addition, patients had a blood sample obtained and completed the two quality of life questionnaires as outlined above.

Outcome measures

The primary outcome measures were the frequency of obliteration (total or partial) of injected varicosity, the number of patients with improvement in venous stasis symptoms, the change in quality of life prior to and at 6 months post sclerotherapy and any associations between clinical outcomes and biomarkers. The secondary outcome measures were the number of patients with recurrence of varicosities and adverse events reported following the procedure.

Statistical analysis

Descriptive statistics and plots were used to summarize the subject demographic and disease characteristics at baseline, and vein sclerosis characteristics on follow-up. Changes in continuous measures (general and vein-specific quality of life) over time (at 1, 12 and 26 weeks) relative to baseline were descriptively summarized. Trends in biomarkers of coagulation at 1, 12 and 26 weeks were analyzed using generalized estimating equations to fit linear models, accounting for the correlation among longitudinal measures made on the sample patient. Median biomarker measures were compared between independent groups defined by degree of varicose vein sclerosis and development of adverse events using a two-sample Wilcoxon rank sum test.

Results

One hundred patients who underwent EFS were enrolled including 29 men and 71 women. Ninety percent were Caucasian. The mean age was 59 years (range 24-85). Among these, 74 had foam sclerotherapy on a single leg (n=42 left and n=32 right) and 26 had both legs treated for a total of 126 total initial EFS sessions. Demographic information is given in Table 1.

Characteristic	N	%
Patient-level Summaries (n=100)		
Male	29	29%
Age	60 median	24-85 range
Body Mass Index (kg/m^2)	29.4 median	18-57 range
Previous Deep-vein Thrombosis	19	19%
Family history of varicose veins	61	61%
History of Superficial thrombophlebitis	17	17%
History of Spontaneous varix rupture	4	4%
Session-level Summaries (n=126)		

Stocking use: knee high	62	49%
Stocking use: thigh high	29	23%
Stocking use: Over-the-counter support (<20 mmHg)	34	27%
History of Surgical vein stripping	18	14%
History of chemical sclerotherapy	10	8%
History of endovenous thermal ablation	5	4%

Table 1: Demographic characteristics.

Nearly all patients (92%) reported using compression hose prior to treatment with EFS. Patients most commonly sought treatment for the symptoms of pain and swelling. Nine patients had a venous ulcer and 2 patients had experience prior cutaneous bleeding due to their varicose veins.

Varicose vein characteristics are given in Table 2. The mean and median vein size treated was 5 cm (3 to 10 mm). Most patients were Clinical Etiologic Anatomic Pathophysiologic classification CEAP class 2 (48%), 20% CEAP class 3, 19% CEAP class 4 and 7% CEAP class 5-6. The median Villalta score at baseline (n=60 for baseline, 3 month and 6 month follow up complete) was 7.25 (interquartile, IQR, range: 3.75-9.00).

Characteristic	**N**	**%**
Indication		
Pain	103	82%
Swelling	94	75%
Thrombophlebitis	16	13%
Ulcer	9	7%
Hemorrhage	2	2%
Other indication: itching, burning, cramps, skin changes	26	21%
Saphenofemoral junction incompetent	54	43%
Deep vein obstruction present	1	1%
Great saphenous vein varicose	92	73%
GSV+tributary varicosities	105	83%
Small saphenous vein varicose	7	6%
SSV+tributary varicosities	9	7%
Above knee incompetent perforating veins	8	6%
Below knee incompetent perforating veins	93	74%
CEAP class 0	1	1%
CEAP class 1	4	3%
CEAP class 2	60	48%
CEAP class 3	25	20%
CEAP class 4a	19	15%
CEAP class 4b	5	4%
CEAP class 5	8	6%
CEAP class 6	1	1%
Vein diameter	5 mm median	3-10 range

Table 2: Vein treatment characteristics (n=126 sessions).

Table 3 summarizes the treatment session characteristics. Among the 114 limbs with a week 1 follow-up measure, 50 (44%) had a record of a reinjection at the 1-week follow-up visit. Among the 74 limbs with a 3-month follow-up record, 4 (5%) had a record of a reinjection after the 1-week follow-up visit and prior to or at the 3-month follow-up visit. Among the 81 limbs with a 6-month follow-up record, 2 (2%) had a record of a reinjection after the 3-month follow-up visit and prior to the 6-month follow-up visit.

Initial Treatment		
Sclerotherapy procedure	**N**	**%**
Tessari/STS initial treatment	126	100%
STS concentration 1%	2	2%
STS concentration 3%	123	98%
STS Sclerosant volume 1 ml	2	2%
STS Sclerosant volume 2 ml	124	98%
Total foam volume 4 ml	2	2%
Total foam volume 8 ml	123	98%
Number of injections per session: 1	120	95%
Number of injections per session: 2*	5	4%
Full intraluminal injection	120	95%
Failed initial attempt	4	3%
Re-injection		
Re-injection at 1 week follow up	50 (n=114)	44%
Re-injection within 3 month follow up	4 (n=74)	5%
Re-injection within 6 month follow up	2 (n=81)	2%
*one patient with 8 injections per session		

Table 3: Treatment characteristics.

Of the 126 initial procedures, 1-week follow-up obliteration data are available for 112 limbs (89%), 3-month follow-up obliteration data are available for 74 limbs (59%), and 6-month follow-up data are available for 81 limbs (64%). At the 1-week visit (following initial treatment, n=112 with obliteration data), complete obliteration of all injected varicosities was observed for 41 (37%) of the limbs. Table 4 summarizes the treatment obliteration results. At the 3-month visit (following initial treatment, n=74 with obliteration data), complete obliteration of all injected varicosities was observed for 58 (78%) of the limbs. At the 6-month visit (following initial treatment, n=81 with

obliteration data), complete obliteration of all injected varicosities was observed for 59 (73%) of the limbs.

	1 week	3 months	6 months
Obliteration: all varicosities	41/112 (37%)	58/74 (78%)	59/81 (73%)
Obliteration: 80-99% varicosities	50/112 (45%)	16/74 (22%)	11/81 (14%)
Obliteration: 50-79% varicosities	0/112	0/74	0/81
Obliteration: minimal with most varicosities patent	21/112 (19%)	0/74	1/81 (1%)
Venous stasis symptom improvement (VEINS)	99/108 (92%)	69/72 (96%)	71/76 (93%)

Table 4: Outcomes of obliteration of treated vein and improvement in venous stasis symptoms.

At the 1-week visit (following initial treatment), improvement in venous stasis symptoms by patient report (VEINS) was observed for 99 (92%) of the 108 limbs with venous stasis data. At the 3-month visit (following initial treatment), improvement in venous stasis symptoms by patient report was observed for 69 (96%) of the 72 limbs with venous stasis data. At the 6-month visit (following initial treatment), improvement in venous stasis symptoms by patient report was observed for 71 (93%) of the 76 limbs with venous stasis data.

Following initial treatment, varicosity recurrence was reported by 1 (1%) of the 114 limbs with data reported. A total of 1 (1%) of the 78 limbs at 3 months, and none of the 74 limbs at 6 months, demonstrated varicosity recurrence.

The change in Villalta score for severity of venous symptoms over time following endovenous foam sclerotherapy and change in quality of life measured by the SF-36 questionnaire is given in Table 5. Villalta score data are available for: 100 participants (125 legs) at the baseline visit, 72 participants (75 legs) at the 3-month visit, and 75 participants (76 legs) at the 6-month visit. There were 60 participants (60 legs) who had Villalta score data for all of the baseline, 3-month, and 6-month visits. In 60 participants (60 legs) who had data for all baseline, 3-month, and 6-month visits, there was a statistically significant improvement in Villalta scale over the 6-month follow-up period. In 72 participants (72 legs) who had data for both baseline and 3-month visits, there was also a statistically significant improvement in Villalta scale. This same finding was seen in 75 participants (75 legs) who had data for both baseline and 6-month visits.

	Baseline median	3-month median	6-month median	p
Villalta score (n=60)	7.25 (3.8-9.0)	2 (1-4.5)	1.5 (1.0-3.5)	<0.0001
PCST (n=58)	49.1 (35.2-54.9)	50.4 (40.5-56.3)	50.6 (41.2-56.2)	0.0668
MCST (n=58)	55.4 (52.1-58.7)	56.6 (52.1-58.8)	56.8 (51.4-59.1)	0.6688

Table 5: Change in quality of life measured by Villalta score and SF-36 (Mental and Physical components) for patients with baseline, 3- and 6-month follow-up data available. Data reported as median (25^{th} percentile-75^{th} percentile).

The SF-36 Quality of life questionnaire was scored according to the RAND method. Physical and mental component summary scores were calculated based on the standard methods. The physical component summary score (PCST) are available for 98 participants at the baseline visit, 74 participants at the 3-month visit, and 75 participants at the 6-month visit. There were 58 participants who had PCST data for all of the baseline, 3-month, and 6-month visits. In 58 participants who had data for all baseline, 3-month, and 6-month visits, and for 67 participants who had data for both baseline and 6-month visits, there was a trend to improvement in PCST (p=0.057).

The Mental component summary score (MCST) are available for 98 participants at the baseline visit, 74 participants at the 3-month visit, and 75 participants at the 6-month visit. There were 58 participants who had MCST data for all of the baseline, 3-month, and 6-month visits, 71 participants who had data for both baseline and 3-month visits, and 67 participants who had data for both baseline and 6-month visits. No statistically significant changes in MCST were seen in comparison in any groups versus baseline.

Thrombotic biomarker changes following EFS

Biomarker data are available for 100 participants (126 legs) at the baseline visit, 79 participants (81 legs) at the 3-month visit, and for 72 participants (74 legs) at the 6-month visit. There were 63 participants (64 legs) who had biomarker data for all of the baseline, 3-month, and 6-month visits. In these patients, there was a statistically significant decline in median PPL compared to baseline over the 6 month follow-up period. In the 79 participants (81 legs) who had data for both baseline and 3-month visits, there was a statistically significant increase in median FM, and decrease in median PPL. In the 72 participants (74 legs) who had data for both baseline and 6-month visits, there was a statistically significant decline in median PPL at 6 months compared to baseline.

At 3 months, there were 70 patients (72 legs) who had clinical outcome and biomarker data.

There were no significant changes in D-dimer, fibrin monomer (FM) or microparticles (PPL) at 3 months compared to baseline in

those with ≥ 80% obliteration of their varicose veins. There was a statistically significant lower median D-dimer, but not in fibrin monomer or PPL in veins with complete obliteration (n=54, median 0.30) as depicted in Table 6 compared to those with less than complete obliteration at 3 months (n=14, median 0.59); however this different was no longer statistically significant when the change from baseline was considered. There were no statistical associations between D-dimer, FM or PPL at 6 months compared to baseline in any patients stratified by obliteration status of their veins.

Coagulation biomarkers	Obliteration<100%			Obliteration=100%			P*
	N	Median (Q1#, Q3)	Mean (SD)	N	Median (Q1, Q3)	Mean (SD)	
D Dimer (µg/ml)	14	0.59 (0.41, 0.89)	0.75 (0.53)	54	0.3 (0.22, 0.56)	0.48 (0.53)	0.02
Liatest FM (µg/ml)	14	3.81 (2.87, 4.25)	3.75 (1.3)	54	4.68 (3.77, 5.16)	5.0 (2.93)	0.08
P PPL (seconds)	14	37.7 (33.2, 54.2)	41.7 (12.1)	54	40.7 (35.3, 47.2)	44.2 (19.0)	0.61

*Comparison between participants with obliteration <100% and those with obliteration=100%. Wilcoxon two-sample test was used to do the analyses because of the small sample size and skewed distribution of biomarkers. #Q1: the First Quartile; Q3: the Third Quartile; SD: Standard Deviation

Table 6: At the 3-month follow-up visit, the distribution of the biomarker values according to obliteration status (complete obliteration of the varicosities).

Complications following EFS are given in Table 7. At the 3-month follow-up visit, there were four (5.7%) of the 70 legs (69 patients) that developed DVT. Patients with DVT had a statistically lower change in D-dimer at 3 months (median change -0.31, IQR=-0.34 to -0.20) compared to baseline relative to patients who did not develop a DVT (median change 0, IQR=-0.09 to 0.12). At the 6-month follow-up visit, there were 3 (4.4%) of the 68 legs (67 patients) developed DVT. There was no statistically significant difference in the change in D-dimer, FM or PPL compared to baseline when comparing patients who did and did not develop DVT at the 6-month follow-up visit.

Adverse event	Count/patients	
	3 month	6 month
Hematoma	0/70 (0%)	0/67 (0%)
Thrombophlebitis	2/70 (2.9%)	0/67 (0%)
Trapped anticoagulum requiring drainage	0/70 (0%)	1/67 (1.5%)
Skin necrosis	1/67 (1.5%)	0/68 (0%)

Table 7: Adverse events following endovenous foam sclerotherapy.

Discussion

To our knowledge, our study represents evaluation of clinical and biomarker characteristics in the largest number of patients receiving EFS using sodium tetradecyl sulphate-room air foam with extended follow up to 6 months post treatment. Our study has several key findings. EFS was proven effective in vein obliteration. Our study showed in a consecutive series of patients treated with sodium tetradecyl sulfate (STS) EFS a vein obliteration rate of 73% in 81 patients followed up at 6 months. More importantly, 93% of patients reported improvement in their venous stasis symptoms assessed by improvement in the VEINS score and reduction in the Villalta score, confirming that vein obliteration does not entirely correlate with symptom improvement. It is important to note that 80% of our patients had documented above or below knee incompetent perforator veins indicating advanced deep venous pathology.

EFS is safe with a low rate of serious adverse effects. There were 4 patients with diagnosed DVT by the 3 month visit, and few with minor side effects of thrombophlebitis, trapped coagulum or skin necrosis. EFS improved perceived quality of life with improvement in the physical component summary score in treated patients at 3 months. The SF-36 QOL questionnaire and the VEINS score represent commonly used QOL assessment tools for venous disease and are therefore reflective of overall improvement in perceived health.

EFS caused an unpredictable change in markers of coagulation. D-dimer was relatively unaffected overall, but statistically increased in those with complete obliteration of their veins compared to those with less than complete obliteration. This may strengthen the hypothesis that thrombosis plays a role in the overall efficacy of EFS. Those with DVT, all of whom were treated, had lower levels of D-dimer compared to baseline, likely reflecting the effect of anticoagulation. While the percentage of patients with DVT post procedure seems high, it is unclear whether this complication represented true occlusive DVT or foam induced thrombosis with an unknown natural history. Conservatively, it was chosen to treat these patients with anticoagulation. Interestingly, those with more complete vein obliteration demonstrated an increased D-dimer at 3 month follow-up compared to those with less than complete obliteration, suggesting a role for coagulation activation in treatment effect. PPL decreased after vein treatment at 3 and 6 months compared to baseline. Since PPL reflects ongoing cell apoptosis or generalized cell death, treatment of underlying inflammation due to varicose veins and venous stasis may stabilize and/or reduce cell death, a finding that should be explored in further studies.

The effectiveness and safety of EFS has been previously evaluated by our group. In a comprehensive meta-analysis of 104 articles including more than 11,000 patients, we found an anatomic closure rate of 85%, with 91% of patients reporting reduction in vein-related symptoms [4]. There are few randomized comparative trials to thermal-based ablation, however, EFS in recent studies shows equal efficacy with few adverse effects [11]. Recently, there has been emphasis on assessment of improvement in venous symptoms and quality of life after EFS rather than vein closure. Darvall et al. showed statistically improved QOL measured by the SF-12 and vein specific Aberdeen Varicose Vein Symptom Score in 296 patients who underwent EFS sustained at 12

months follow up [12]. More recently, Biemans and colleagues showed similar improvement in the Chronic venous insufficiency QOL questionnaire and EuroQol 5 scores in 240 patients randomized to EVLT, EFS or conventional surgery at 3 months and not associated with anatomic vein closure [13]. Our findings of improvement in vein specific symptoms, and overall quality of life following EFS support its efficacy.

To date, we have found no comprehensive analysis of the coagulation effects of EFS. The effect of sclerotherapy on hemostasis activation and thrombosis has been evaluated in small studies. Ikeda et al. studied 41 consecutive patients who were randomized to surgical therapy of their varicose veins versus injection sclerotherapy with hypertonic saline had blood sampling at baseline and on the 7th a d 28th day post procedure [14]. In both groups, the thrombin antithrombin complex (TAT), D-dimer and fibrinogen concentrations were significantly elevated at day 7 compared to baseline, however the C-reactive protein (CRP) showed no change. Superficial thrombosis occurred more frequently in the sclerotherapy group and was associated with elevated TAT.

In contrast, Hamel-Desnos studied 40 patients who underwent foam sclerotherapy for treatment of the GSV or SSV with 1%polidocanol/room air and measured coagulation markers at baseline, and to 28 days post procedure [15]. Of the markers measured, D-dimer increased at day 1 and 7 post procedure in those prescribed compression hose or not. In addition, TAT increased significantly at day 1, and PF4 levels reduced by day 7. However, few levels were seen above the normal ranges. The authors concluded that foam sclerotherapy had minimal effect on markers of coagulation. Similarly, Fabi et al. studied coagulation factors after up to 30 ml of 0.25% STS foam was injected in 20 patients' reticular veins with follow up blood sampling at 15 minutes post injection [16]. There was no significant pre- to post-sclerotherapy change in any fibrinolysis, coagulation times, or thrombin activation. Most recently, Shadid and colleagues [6] studied 8 patients treated with foam sclerotherapy (polidocanol 3%, 1 to 4 dilution with room air) and measured thrombin generation paramaters (CAT), TAT, D-dimer, fibrinogen, vWf Ag, and PPL before and after treatment at 30 minutes, one and four hours and at one week. They found significant changes in coagulation parameters including reduction of endogenous thrombin potential, initial decrease than increase in fibrinogen, initial increase then decrease in D-dimer and TAT and decrease then increase in PPLs. The authors concluded that this pilot study showed that EFS caused a marked and acute activation of coagulation with a compensatory inhibitor effect on thrombin generation. They hypothesized that despite an acute procoagulant effect of EFS, the tendency to hypercoagulability was dampened. The diversity of findings regarding activation of coagulation in these studies highlights our limited understanding of the *in-vivo* effects of EFS, especially specific to sclerosant and gas diluent. Large cohort studies using modern techniques of EFS are required to further elucidate these mechanisms.

There are several limitations to our study. First, the sample size is modest limiting the strength of the findings and the number lost to follow up at 6 months was more than anticipated. However, previous studies evaluated *in-vivo* coagulation effects in much smaller numbers of patients. Also, our patients often underwent repeated injections to ensure treatment of all truncal and accessory varicose veins, that made analysis of effects of single injection EFS more difficult. The number of injections per patient and the concentration of sclerosant also varied, although most patients received 3% STS. However, repeated injection at varied intervals represents current sclerotherapy practice. At the time of design of this study, the revised Venous Clinical Severity Score (VCSS), a common varicose vein QOL measure, had not been fully validated and incorporated into our practice, and therefore was not prospectively collected. This score would have been useful in measurement of treatment effect. Instead, the Villalta score used for patient with post-thrombotic syndrome was used to evaluate severity of venous disease. The number of patients with complete follow up to 6 months was incomplete; two-thirds of patients were available for evaluation at the end of the study.

In conclusion, we have found that EFS using STS-room air is effective in treatment of varicose veins and related symptoms with overall improved quality of life and low rate of adverse effects. EFS produced variable effects on coagulation including elevated in D-dimer that was associated with extent of vein obliteration, and measured microparticles that decreased over time suggesting a decrease in cell apoptosis after EFS or perhaps a hypothesis of decreased vein inflammation. As foam based treatment of varicose veins becomes more common with the recent FDA approval of commercial microfoam, *in-vivo* biological effects should be further explored.

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