Duplex mapping of 2036 primary varicose veins

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Objective: **To produce a comprehensive anatomical and hemodynamic ultrasound scan mapping of the whole limb in patients with primary varicose veins (VVs).**
Design: **An analysis of venous duplex scans performed on patients referred for treatment of primary VVs.**
Methods: **A total of 2036 limbs were evaluated, looking for the origin of VVs in the saphenous systems and in the perforating vein (PV) systems, as well as for the presence of non-saphenous reflux.**
Results: **The sapheno-femoral junction (SFJ) of the great saphenous vein (GSV) was involved in 1330 limbs (65.3%). We have noted that finding reflux in the groin does not imply that it originates at that point necessarily, as reflux from the pelvis or abdominal wall can also cause primary VVs (SFJ reflux, 41.9% and competent SFJ with reflux from proximal veins, 35.4%). We also noted that analyzing only the presence of reflux in the SFJ of the GSV would miss 10.9% of limbs of reflux in the SFJ of the anterior accessory GSV. In 237 limbs (11.6%), reflux was observed in the popliteal fossa. In the PV system, we distinguished those PVs with retrograde flow that acted as an origin of the VVs, and other PVs that acted as re-entry points. Based on this difference, the location identified as the most frequent origin of VVs in the PV system was the thigh, specifically in the group of PVs of the medial thigh of the femoral canal, with 85 PVs with a total of 238 incompetent PVs identified. Pure non-saphenous reflux was observed in 162 limbs (8%).**
Conclusion: **The assumption that the origin of VVs would be exclusively in the sapheno-femoral or sapheno-popliteal junction, is a mistaken attitude and a comprehensive duplex scan mapping is recommended. (J Vasc Surg 2009;49: 681-9.)**

Duplex ultrasonography (DU) scan has revolutionized the diagnostic and therapeutic management of chronic venous insufficiency (CVI). Nowadays, many authors recommend this imaging test for the assessment of varicose disease, especially when there is a chance to offer the patient surgical treatment.[1-4] Therefore, the current underuse of this test is only due to time, cost, and staff implications.[1] Performance of DU for varicose veins (VVs) assessment is useful to identify the origin of the venous reflux, the relationships of the different venous systems among themselves, and their possible anatomical variations. Recurrence rates of 52-65% at 5 years after VVs surgery have been published.[5,6] It is thought that inappropriate surgery in the first place, due to lack of an adequate anatomical and hemodynamic assessment, is the basis for these high recurrence rates.[7] The best method to obtain better surgical results is through the effective identification of the main cause of reflux in the original surgical intervention. Although there have been several studies on the use of DU in the assessment of VVs, most of them have focused on one particular aspect, such as the examination of the sapheno-femoral junction (SFJ) or sapheno-popliteal junction (SPJ) alone.[2,3,8] The aim of the present study was to produce a comprehensive anatomical and hemodynamic ultrasound scan mapping of the whole limb in patients with primary VVs to evidence all possible patterns of reflux of the limb, and significant anatomical variations of the superficial venous system that can influence the therapeutic strategy.

PATIENTS AND METHODS

We reviewed retrospectively, from an existing prospective database, the ultrasound scan mapping of the lower limbs venous systems for all patients referred to the Vascular Surgery Outpatients Clinic for assessment of their primary VVs, to evaluate for possible surgical treatment, between January 1998 and August 2004. Patients were unselected consecutive assessments. The rates of different reflux patterns detected by DU were investigated, and this information was plotted in paper format as a hemodynamic and anatomic ultrasound scan mapping. All duplex scan mappings were performed or supervised by a vascular surgeon with 10 years experience in the diagnostic management of CVI by DU, according to protocols and standard criteria recommended by the Spanish Society for Angiology and Vascular Surgery[9] and previous international publications.[10,11] The interpretation of all the duplex scan mappings was carried out by a single vascular surgeon. Patients were excluded if they had a confirmed history of deep vein thrombosis in the assessed lower limb, or venous surgery of any kind, or sclerotherapy of VVs in the affected limb (except for sclerosis of reticular veins or telangiectases).

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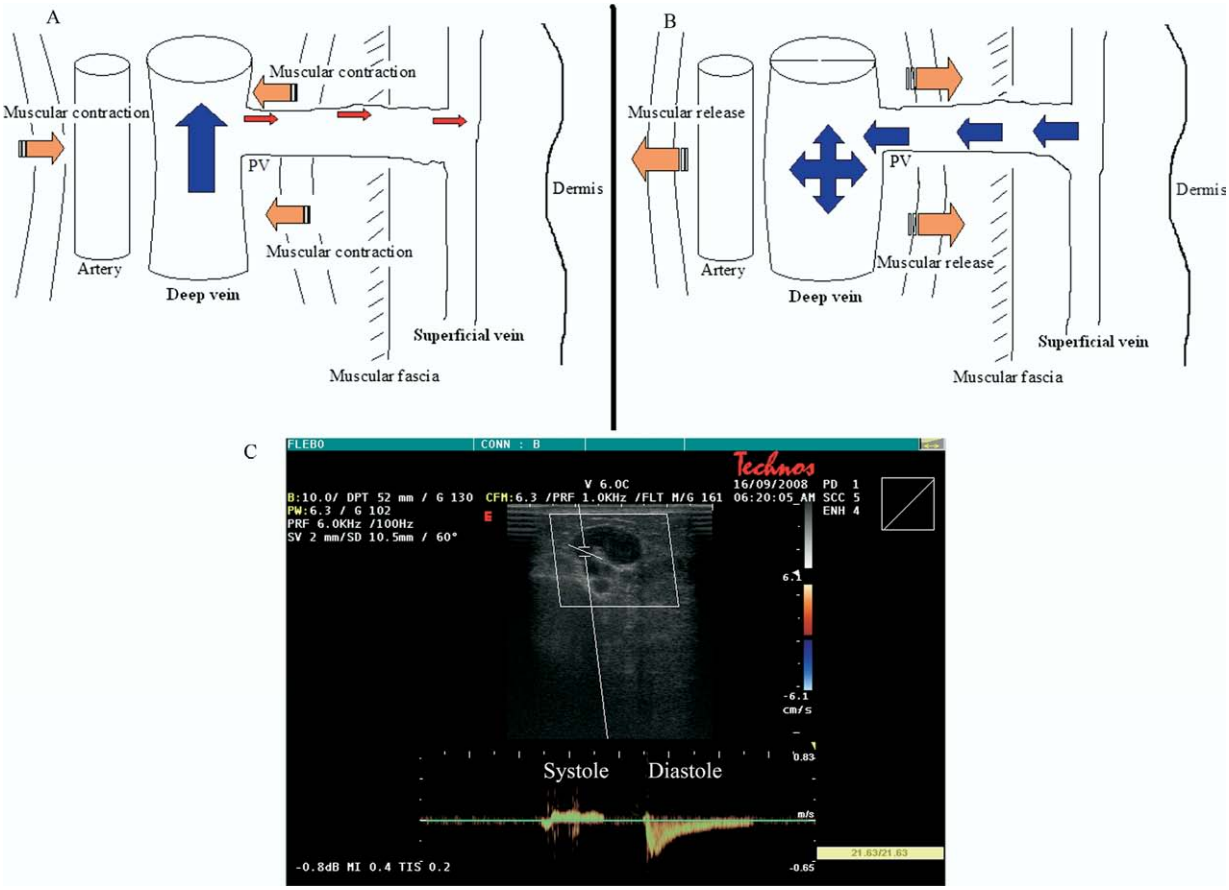


Fig 1. A, The re-entry perforating vein (PV) would have a retrograde flow (*red arrows*) in muscular contraction (systole; *orange arrows*) which would ascend through the superficial venous tract. **B,** But during the muscular release (diastole; *orange arrows*), the lower pressure generated in the deep venous system would add to the hydrostatic pressure of the blood column from the superficial venous tract, thereby generating an anterograde flow which would be greater than the initial retrograde flow (*blue arrows*). **C,** Duplex examination of the PV during Paranà maneuver.[13] ✣: retained flow.

Clinical status was classified according to the C part of the CEAP.[12]

The test was performed in the standing position by using an Esaote-Technos MP ultrasound scanner (Genoa, Italy) with a lineal multi-frequency probe (5-10 mHz). The assessment included compression-release tests, active foot dorsiflexion and relaxation, and Valsalva and Paranà maneuvers.[13] An analysis of the venous refluxes in the saphenous systems, its junctions, their varicose tracts, and in the perforating venous (PV) systems was also carried out, as well as an analysis of the presence of non-saphenous reflux.

From a hemodynamic point of view, we conceive venous pathology as a retrograde circuit (veno-venous shunt), determined by a venous reflux point, an incompetent venous tract, and finally a re-entry point to the deep venous system.

Reflux was defined as a flow in an inverse direction to the physiological flow with duration greater than 0.5 seconds after provocation maneuvers in all systems (superficial, deep, and PV). Venous reflux point was defined as the point through which there is a passage of blood from the deep to the superficial system, and, in general, as the point which will originate VVs.[9] On the contrary, the re-entry point was defined as the point through which the blood returns from the superficial to the deep system.[9] The incompetent venous tract connects both points, and due to the existence of gradient of pressure between both, the reflux occurs.

The PV system deserves special mention. We distinguished those PVs with retrograde flow that acted as a venous reflux point, and other PVs that acted as re-entry points (Fig 1-3).

The re-entry PVs would have a retrograde flow in muscular contraction (systole), which would ascend through the superficial venous tract, but during the muscular release (diastole), the lower pressure generated in the deep venous system would add to the hydrostatic pressure of the blood column from the superficial venous tract, thereby generating an anterograde flow which would be greater than the

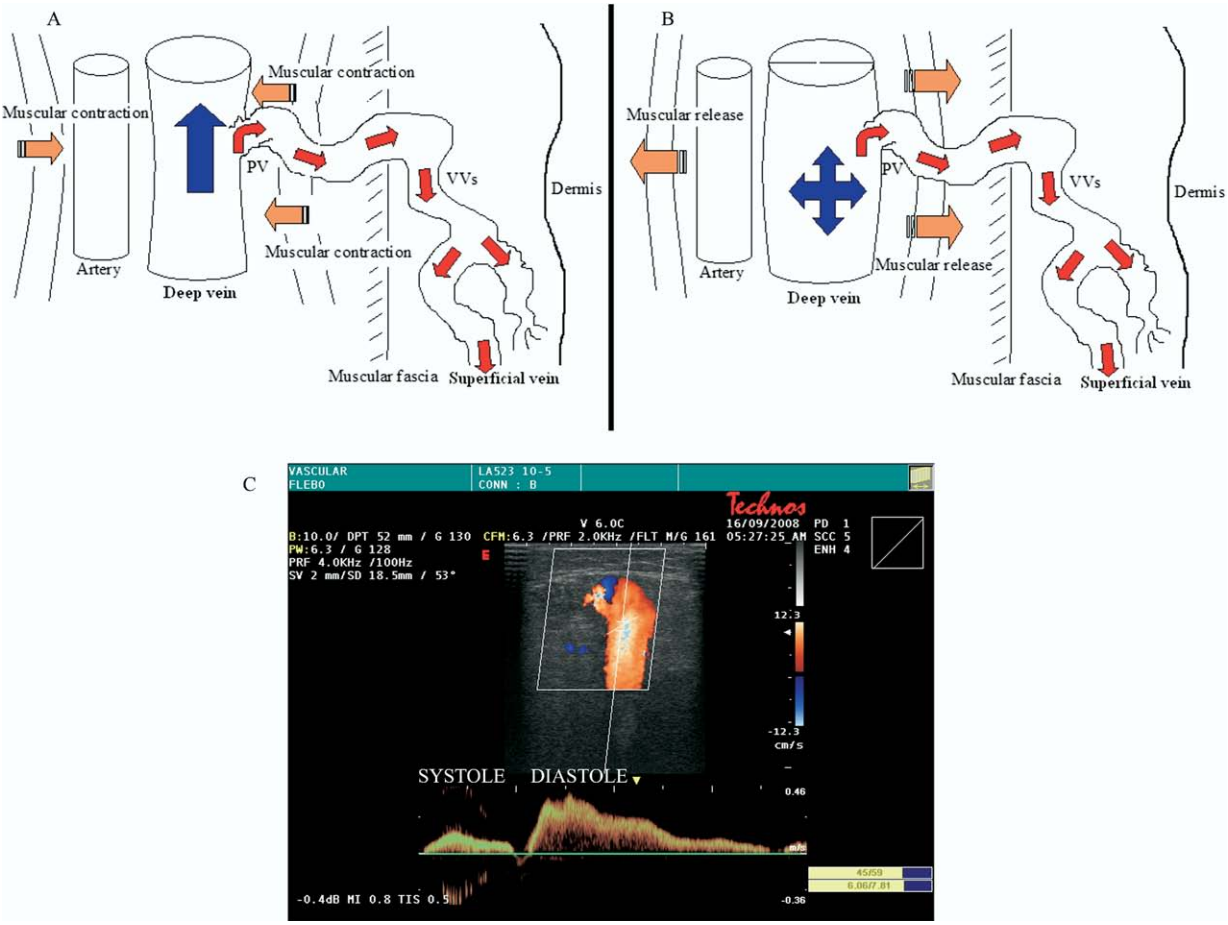


Fig 2. A, Perforating veins (PV) acting as venous reflux points are those showing retrograde flow (*red arrows*) during muscular contraction (systole; *orange arrows*) **(B)** and principally during muscular release (diastole; *orange arrows*). **C,** Duplex examination of the PV during Paranà maneuver.[13] *VVs,* Varicose veins; ✣: retained flow.

initial retrograde flow (Fig 1). PVs acting as a reflux venous point, on the other hand, would be those showing retrograde flow in muscular diastole (Fig 2), or retrograde flow in muscular systole which would be markedly greater than the anterograde flow in muscular diastole (Fig 3).[14]

Three types of reflux evidenced were based on their three sites: inguinal reflux, popliteal reflux, and non-saphenous reflux. For detailed drawings of normal hemodynamic ultrasound scan mapping and possible combinations of reflux see the Figs E1-E14 (online only).

Reflux in the groin. In the SFJ, the possibility of reflux both in the great saphenous vein (GSV) and anterior accessory great saphenous vein (AAGSV) was considered. For both, SFJ reflux was defined as reflux in the GSV or AAGSV accompanied by segmental incompetence in the common femoral vein, indicating the presence of terminal valve incompetence. Competent SFJ with reflux from proximal veins was defined as reflux in the SFJ of the GSV or AAGSV, but without incompetence at the level of the common femoral vein, which would demonstrate the competence of the terminal valve and would require searching for the origin of the reflux in some epigastric or pudendal collateral to characterize this reflux (competent SFJ with reflux from epigastric vein or with reflux from pelvis). In the case of AAGSV, segmental reflux in AAGSV, was also defined as the reflux in AAGSV that does not come from SFJ reflux or competent SFJ with reflux from proximal veins and that could be justified by a reflux within the valve of the AAGSV.

Reflux in popliteal fossa. The possibility of reflux at the level of the SPJ through gastrocnemius veins or through terminal valve incompetence of the small saphenous vein (SSV) was investigated, as well as the possibility that the SSV would be incompetence through a refluxing Giacomini vein, or through refluxing VVs from another venous system.

Non-saphenous reflux. Under the term "non-saphenous" we included two types of VVs. First, those VVs whose reflux originated in the PV system. The PVs were classified as per recent recommendations by the Federative International Committee on Anatomical Terminology (FICAT).[15] Second, we have classified as "pure non-

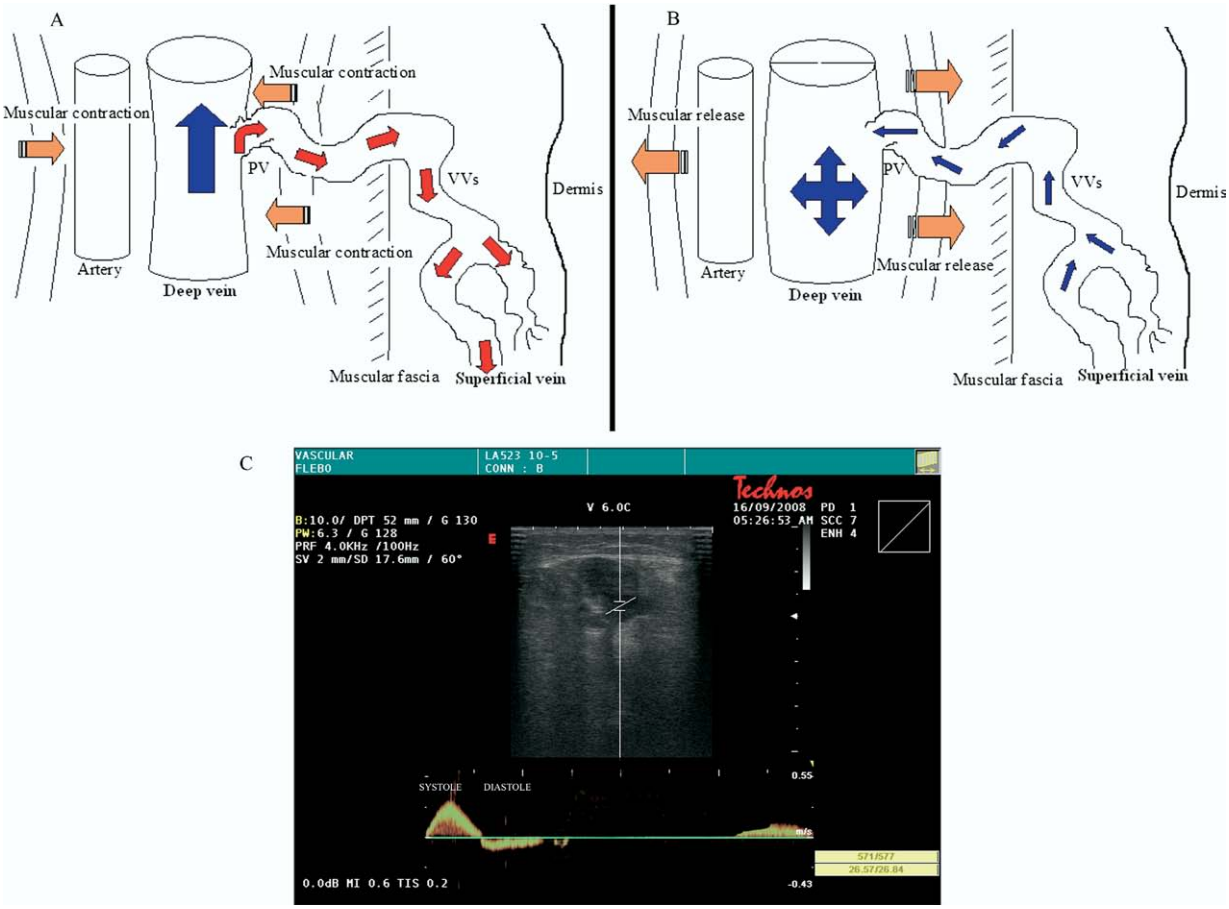


Fig 3. A, Perforating veins (PV) acting as venous reflux points are those showing retrograde flow (*red arrows*) during muscular contraction (systole; *orange arrows*), **(B)** which would be markedly greater than the anterograde flow (*blue arrows*) during muscular release (diastole; *orange arrows*). **C,** Duplex examination of the PV during Paranà maneuver.[13] *VVs,* Varicose veins; ✣: retained flow.

saphenous" reflux those VVs which reflux source was not related to saphenous junctions or the PV system. This type of reflux could include pelvic or epigastric reflux not afferent to saphenous junctions, with no reflux in trunk saphenous veins, and also could include reflux in tributaries from saphenous veins close to competent trunk saphenous veins.

Anatomy of the CEAP classification. A retrograde flow segment does not necessarily entail to be the reflux origin. Many other possibilities exist, as has been previously described, and this was considered in our study. Nevertheless, the anatomical category ($A_{S,D,P}$) of the CEAP classification was defined, depending on the presence or absence of reflux in the different segments that constitute it, without any other hemodynamic consideration.[12] AAGSV was registered as segment in this classification.

We have considered that existed reflux in deep venous system only if such reflux was evident in a segment different to that it contains the venous reflux point (for example, the SFJ or the SPJ refluxes).

Interrelations between the saphenous system and the PV system. We recorded whether the segment of the GSV received a refluxing VV that originated from an incompetent SSV or AAGSV or from an incompetent PV, and made it to reflux. Similarly, we recorded this possibility for the segment of the SSV and the AAGSV.

RESULTS

A total of 2036 ultrasound scan mappings were carried out for 1595 patients. Bilateral disease was detected in 27.6%. A total of 431 patients were excluded because of having recurrent VVs. The male to female ratio was 1:3. The mean age of the patients examined was 48.2 years. We distinguished two categories according to clinical severity in CEAP classification: **mild to moderate CVI** (C1 to C3) and **severe CVI,** characterized by the presence of skin changes (C4 to C6). Mild to moderate CVI was present in 1768 limbs (86.8% of the series), whereas severe CVI was evident in 268 limbs (13.2%). The frequencies of all possi-

Table I. Frequencies of all possible venous reflux points of the limb in the superficial venous system of 2036 limbs

Reflux in the groin	1528 (75%)
Reflux in GSV	1330 (65.3%)
SFJ reflux of the GSV	694 (34.1%)
Competent SFJ of the GSV with reflux from proximal veins	660 (32.4%)
Competent SFJ of the GSV with reflux from pelvis	644 (31.6%)
Competent SFJ of the GSV with reflux from epigastric vein	27 (1.3%)
Reflux in AAGSV	221 (10.9%)
SFJ reflux of the AAGSV	87 (4.3%)
Segmental reflux in AAGSV	72 (3.5%)
Competent SFJ of the AAGSV with reflux from proximal veins	62 (3.0%)
Competent SFJ of the AAGSV with reflux from pelvis	59 (2.9%)
Competent SFJ of the AAGSV with reflux from epigastric vein	3 (0.1%)
Reflux in the popliteal fossa	237 (11.6%)
SPJ reflux	214 (10.5%)
SSV reflux though gastrocnemius veins	3 (0.1%)
SSV reflux though an incompetent VV	7 (0.3%)
SSV reflux though Giacomini vein	13 (0.6%)
Reflux in the PV system	238 (11.7%)
PV gluteal	3 (0.1%)
PV thigh	150 (7.4%)
PV knee	39 (1.9%)
PV calf	42 (2.1%)
PV ankle	4 (0.2%)
PV foot	0
Pure non-saphenous reflux	162 (8.0%)

Numbers of limbs (percentage). *GSV,* Great saphenous vein; *SFJ,* sapheno-femoral junction; *AAGSV,* anterior accessory great saphenous vein; *SPJ,* sapheno-popliteal junction; *SSV,* small saphenous vein; *VV,* varicose veins; *PV,* perforating venous.

ble venous reflux points of the limb of the superficial venous system were investigated (Table I).

Reflux in the groin. Topographically, the groin was the most frequent location where the venous reflux was identified (75%). The SFJ of the GSV was involved in 1330 limbs of the 2036 ultrasound scan mappings (65.3%). In 694 of these (34.1%) limbs, reflux was due to incompetence of the terminal valve, whereas in 660 limbs (32.4%) the SFJ was competent with reflux from proximal veins (competent SFJ with reflux from pelvis, 31.6% and from epigastric vein, 1.3%). Both kinds of reflux could be present in the same limb, which is why the sum of percentages of their frequencies does not represent the value 100. This situation also occurred at SFJ of the AAGSV, the popliteal fossa, and the PV system, because the same limb can have more than one venous reflux point.

In the groin, it was also considered that the AAGSV could be involved in the causation of reflux. In this study, in 221 limbs (10.9%), the venous reflux point was found in relation to the SFJ of the AAGSV, with the varieties of SFJ reflux (4.3%), competent SFJ with reflux from proximal veins (3%), and the reflux we term segmental reflux in AAGSV (3.5%).

Reflux in the popliteal fossa. In 237 (11.6%) of the 2036 ultrasound scan studies, reflux was observed in the popliteal fossa. SPJ reflux accounted for 10.5% of the total, whereas SSV reflux through the Giacomini vein was identified in 0.6%, through a refluxing VV in popliteal fossa in 0.3% and through gastrocnemius veins in 0.1% of the ultrasound scan mappings.

Table II. Frequencies of all possible venous reflux points of the limb in the PV system of 2036 limbs

Groups of PVs	*n (%)*
Gluteal PVs	
Superior gluteal PV	0
Midgluteal PV	0
Lower gluteal PV	3 (0.15%)
Thigh PVs	
PV of the femoral canal	85 (4.17%)
Inguinal PV	1 (0.05%)
Anterior thigh PV	22 (1.08%)
Lateral thigh PV	16 (0.79%)
Posteromedial thigh PV	13 (0.64%)
Sciatic PV	7 (0.34%)
Posterolateral thigh PV	5 (0.25%)
Pudendal PV	1 (0.05%)
Knee PVs	
Medial knee PV	3 (0.15%)
Suprapatellar PV	6 (0.29%)
Lateral knee PV	8 (0.39%)
Infrapatellar PV	1 (0.05%)
Popliteal fossa PV	21 (1.03%)
Leg PVs	
Paratibial PV	3 (0.15%)
Posterior tibial PV	
- upper	10 (0.49%)
- middle	11 (0.54%)
- lower	9 (0.44%)
Anterior leg PV	2 (0.1%)
Lateral leg PV	0
Medial gastrocnemius PV	1 (0.05%)
Lateral gastrocnemius PV	3 (0.15%)
Intergemellar PV	3 (0.15%)
Para-Achillean PV	0
Ankle PVs	
Medial ankle PV	3 (0.15%)
Anterior ankle PV	1 (0.05%)
Lateral ankle PV	0
Foot PVs	
Dorsal foot PV	0
Medial foot PV	0
Lateral foot PV	0

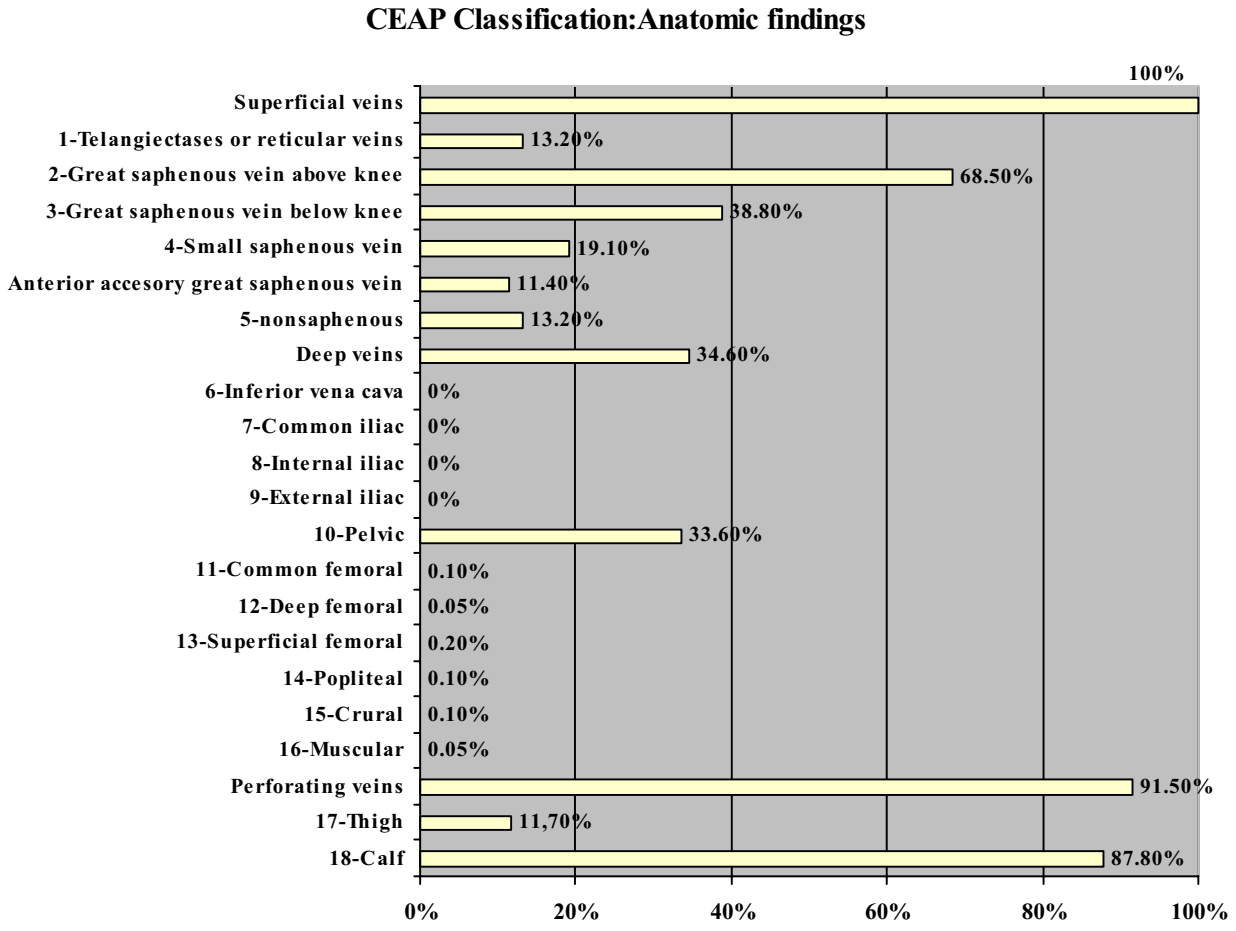
Numbers of limbs (percentage). *PV,* Perforating venous.

Reflux in the PV system. The location identified as the most frequent venous reflux point in this system was the thigh, more specifically, in the group of PVs of the medial thigh of the femoral canal, with 85 of a total of 238 incompetent PVs identified (35.7%) (Table II).

In ten limbs there were two incompetent PVs identified as venous reflux points in the same extremity and in one limb three incompetent PVs were identified.

Pure non-saphenous reflux. In 162 limbs (8%) VVs refluxes were reflected in the ultrasound scan mapping.

Reflux pattern according to the anatomy of the CEAP classification. Graph 1 shows the frequency of the different venous reflux segments involved according to the CEAP classification.



Graph 1. The frequencies of the different incompetent venous segments involved according to the CEAP classification.

Intersaphenous relationships and relationships with the PV system. The SSV, without any venous reflux points in its system, was incompetent when receiving a refluxing VV from an incompetent GSV in 126 limbs (6.2%), from an incompetent AAGSV in 17 limbs (0.8%), and when receiving a refluxing VV originating from a venous reflux point in the PV system in 21 limbs (1%).

The GSV, without any venous reflux points in its system, was incompetent when receiving a refluxing VV from an incompetent SSV in 43 limbs (2.1%), from an incompetent AAGSV in 22 limbs (1.1%), and when receiving a refluxing VV originating from a venous reflux point in the PV system in 65 limbs (3.2%).

The AAGSV without any venous reflux points in its system was incompetent when receiving a refluxing VV from an incompetent GSV in only one limb (0.05%) and when receiving a refluxing VV originating from a venous reflux point in the PV system in three cases (0.1%). There were no limbs in which the AAGSV was incompetent when receiving a refluxing VV from an incompetent SSV.

DISCUSSION

Using Duplex ultrasonography scan, we searched and identified the venous reflux points as the locations where the refluxes originated. We then plotted them on an anatomical and hemodynamic ultrasound scan mapping. Although there are several studies assessing patients with VVs by using DU, most have focused on the SFJ or SPJ only.[3,8,16,17] Certainly, reflux in the SFJ is the most common one. In our study, reflux in the groin was found in 75% of the cases. The frequency of reflux in the SFJ, according to the studies reviewed, is between 53% and 89.8%.[1-3,8,16-20] We have noted that finding reflux in the groin does not imply that it originates at that point, as reflux from the pelvis or abdominal wall can also cause primary VVs (SFJ reflux, 41.9% and competent SFJ with reflux from proximal veins, 35.4%). We also noted that analyzing only the presence of reflux in the SFJ of the GSV would miss 10.9% of limbs of reflux in the SFJ of the AAGSV. Seidel et al,[21] found that the AAGSV was incompetent in 8.09% of cases (86/1063), Ricchi et al,[22] in 14%

and Labropoulos et al,[23] in 8.8%. Juan-Samsó reported that the technical fault of not identifying the existence of AAGSV reflux was responsible for 12% of recurrences.[24] The SFJ is a complex anatomical and functional unit, and few studies on prevalence of reflux at this level take this peculiarity into account. Thus, although it is common to read the distinction between SFJ reflux and competent SFJ with reflux from proximal veins in the studies that make reference to the strategy known as Cure Conservatrice et Hemodynamique de l'Insuffisance Veineuse en Ambulatoire (CHIVA),[20] very few studies specify its differentiation and frequency,[25,26] and only an occasional study would exceptionally mention the possibility of the AAGSV being the only origin of the VVs.[22] Jiang et al[27] found an incidence of non-saphenofemoral venous reflux in 6.1% of cases of primary VVs. Esteban et al,[20] in a study assessing the CHIVA procedure in a series of 225 patients treated surgically for primary VVs, found competent SFJ with reflux from proximal veins of the GSV in 26.2% of the cases.

In our study, within the group of inguinal refluxes, SFJ refluxes (SFJ reflux of GSV, AAGSV, and segmental reflux in AAGSV) represented 55.8% (853/1528) and the limbs with competent SFJ with reflux from proximal veins (competent SFJ with reflux from proximal veins of GSV and AAGSV) represented 47.2% (722/1528). Recently, and consistent with our results, Capelli et al[17] reported that approximately half the refluxes in the SFJ presented with a competent terminal valve. The Phlebology International Union Consensus, published in 2006, already highlights the clinical importance of differentiating SFJ reflux from competent SFJ with reflux from proximal veins.[28]

In 11.6% of limbs, we found reflux in the popliteal fossa. The frequency of reflux in SPJ, according to the studies reviewed, should be somewhere between 9.4% and 28%.[1-3,8,16,19,20,29,30] In our study, SPJ reflux accounted for 10.5% of the limbs. In the popliteal fossa, the majority of limbs were SPJ reflux (90.3%), but in the remaining 9.7% we noted that in the majority (86.6%), the origin of the VV would be far from the popliteal fossa, through an incompetent Giacomini vein or through a refluxing VV. This indicates that surgery on the popliteal fossa in these limbs would be inappropriate. The presence of an incompetent VV in the popliteal fossa as cause of reflux has been previously described[1,18,30] and in our study it accounted for 2.9% of all refluxes in popliteal fossa.

We have found 238 venous reflux points that have corresponded to incompetent PVs of different locations. When examining a patient with venous insufficiency, we may find PVs that have a retrograde flow, acting as venous reflux points, and other PVs acting as re-entry points.[14,31,32] This hemodynamic concept of the differentiation between PVs is not reflected in the majority of the studies done, which simply regard a PV as incompetent when there is a retrograde flow greater than 0.5 seconds or according to the size of the PV.[33] Based on this definition, the prevalence of incompetent PVs in primary VVs ranges from 2% to 61%,[2,3,34,35] depending on clinical severity. This implies that most of the re-entry PVs should be eliminated. However, it has been reported that a great number of PVs, previously described as incompetent, usually return to normality after a short stripping of the GSV.[33,36] Other authors simply do not regard them due to the lack of consensus about their hemodynamic role in varicose pathology.[37] We have differentiated those incompetent PVs that act as venous reflux points from those that act as re-entry points, so that in our study we have found a prevalence of incompetent PVs in primary VVs identified as the origin of the reflux in 11.7%. The most frequent location was the group of PVs of the femoral canal of the medial thigh. In our series, the frequency of PVs of the posterolateral thigh was 0.25%. Labropolous et al[38] found a prevalence of 0.92% in this location and warned that failure to identify them could be the cause of recurrent VVs and unnecessary surgery. Popliteal fossa PVs deserve special mention, because of their close relationship to the SPJ and possible cause of error by assuming that this is where the venous reflux point is located. In our series, we have recorded 21 incompetent PVs in popliteal fossa that represent 1.0% of the limbs studied. Other authors report prevalences of 0.8% and 2.2%.[30,39]

"Pure" non saphenous reflux presented in 8.0% of limbs. Labropoulos et at[40] reported a prevalence of reflux in tributary veins of GSV of 9.7% and, based on this fact, stated that the finding of an isolated reflux of a tributary vein without evidence of reflux in the trunk of GSV implies that the reflux may occur in an isolated segment or be multifocal without communication between them (theory of local changes in the venous wall).

We recorded the venous reflux segments according to the CEAP classification. A segmental venous reflux does not imply that such a segment is the origin of the reflux. Thus, we see that the segment of the SSV was incompetent in 19.6% of limbs, according to the CEAP classification, whilst we actually have only evidenced on SPJ reflux of the SSV in 10.5% of limbs, for example. The relationships between the various venous systems explain these differences. We have observed, for instance, that in 8.9% of limbs, there could be an incompetent SSV only due to these relationships. Another notable difference is found in the PV system. In our view, the most common location of the origin of the VVs in this system would be in the thigh and gluteal areas (64.3%), whilst according to the CEAP register, the commonest incompetent segment would be in the segment of the PVs of the calf (87.8%). For us, this is where most of the PVs that act as re-entry points would be located.

CONCLUSION

In our opinion, the assessment of primary VVs in lower limbs, especially if they are being evaluated for a surgical procedure, requires the performance of a diagnostic test to determine its hemodynamic behavior and allow a comprehensive morphological identification. Duplex ultrasonography scan is currently the most appropriate tool to meet these objectives. The assumption that the origin of the VVs would be exclusively in the SFJ or SPJ, is a mistaken

attitude resulting in the high rates of recurrences published. The implications from differentiating between SFJ reflux from competent SFJ with reflux from proximal veins in the groin are of great importance for a better surgical approach and a better understanding of the symptoms reported by the patient. Now, with the advent of new therapeutic techniques applied to varicose pathology (radio frequency, endolaser, and endovascular surgery), it is even more important to increase our knowledge of CVI from the morphologic and hemodynamic point of view. Duplex ultrasonography scan is the recommended tool for the strategic approach to CHIVA and, thanks to the development of DU, interest in and development of hemodynamic and morphologic ultrasound scan mapping has grown. Nevertheless, our work does not analyze therapeutic considerations, but rather emphasizes DU's potential for identifying the different and diverse origins of the venous reflux and their frequency. As vascular surgeons, we know the importance of the GSV as potential vascular graft for a possible future vascular reconstruction in patients with peripheral arterial disease. To detect and differentiate an incompetent AAGSV with competent GSV, or vice versa, is of great importance, as failing to do so prior to VV surgery could result in the unnecessary extraction of part of the competent venous network. Therefore, if DU assessments were done routinely, we would expect to learn more about the natural history of the disease and see lower recurrent rates after varicose surgery. Further studies would be required, however, to confirm these predictions.

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AUTHOR CONTRIBUTIONS

Conception and design: MG-G, SR, CV
Analysis and interpretation: MG-G
Data collection: MG-G, ST, EG, MG-A, DL
Writing the article: MG-G, ER
Critical revision of the article: MG-G, ER
Final approval of the article: MG-G, ER, SR, CV, ST, EG, MG-A, DL
Statistical analysis: ER
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Overall responsibility: MG-G

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