What's new: Management of venous leg ulcers

Approach to venous leg ulcers

Afsaneh Alavi, MD,[a] R. Gary Sibbald, MD,[a,b] Tania J. Phillips, MD,[c] O. Fred Miller, MD,[d] David J. Margolis, MD, PhD,[e] William Marston, MD,[f] Kevin Woo, RN, PhD,[g] Marco Romanelli, MD, PhD,[h] and Robert S. Kirsner, MD, PhD[i]

Toronto and Kingston, Ontario, Canada; Boston, Massachusetts; Danville and Philadelphia, Pennsylvania; Chapel Hill, North Carolina; Pisa, Italy; and Miami, Florida

Learning objectives

After completing this learning activity, participants should be able to evaluate and treat the symptoms and signs of early venous disease, prevent or delay the occurrence of venous leg ulcers, assess the differential diagnosis of leg ulcers, and delineate an approach to the evaluation of leg ulcers.

Disclosures

Editors

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Leg ulcerations are a common problem, with an estimated prevalence of 1% to 2% in the adult population. Venous leg ulcers are primarily treated in outpatient settings and often are managed by dermatologists. Recent advances in the diagnosis and treatment of leg ulcers combined with available evidence-based data will provide an update on this topic. A systematized approach and the judicious use of expensive advanced therapeutics are critical. Specialized arterial and venous studies are most commonly noninvasive. The ankle brachial pressure index can be performed with a handheld Doppler unit at the bedside by most clinicians. The vascular laboratory results and duplex Doppler findings are used to identify segmental defects and potential operative candidates. Studies of the venous system can also predict a subset of patients who may benefit from surgery. Successful leg ulcer management requires an interdisciplinary team to make the correct diagnosis, assess the vascular supply, and identify other modifiable factors to optimize healing. The aim of this continuing medical education article is to provide an update on the management of venous leg ulcers. Part I is focused on the approach to venous ulcer diagnostic testing. (J Am Acad Dermatol 2016;74:627-40.)

Key words: leg ulcers; lipodermatosclerosis; venous disease; venous leg ulcers; wound healing.

From the Departments of Medicine (Dermatology)[a] and Public Health,[b] University of Toronto; Department of Dermatology,[c] Boston University School of Medicine; Department of Dermatology,[d] Geisinger Health System, Danville; Department of Dermatology,[e] University of Pennsylvania, Perelman School of Medicine, Philadelphia; Department of Surgery,[f] University of North Carolina, Chapel Hill; Faculty of Nursing,[g] Queen's University, Kingston; Department of Dermatology,[h] University of Pisa; and the Department of Dermatology and Cutaneous Surgery,[i] University of Miami.

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Correspondence to: Afsaneh Alavi, MD, Wound Care Centre, Women's College Hospital (Main Bldg), 76 Grenville St, 5th Fl, Toronto, ON M5S 1B2, Canada. E-mail: afsaneh.alavi@utoronto.ca.

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Abbreviations used:

ABPI:	ankle brachial pressure index
CEAP:	clinical, etiology, anatomy, and physiology
DVT:	deep venous thrombosis
LDS:	lipodermatosclerosis
$TcPCO_2$:	transcutaneous pressure of carbon dioxide
$TcPO_2$:	transcutaneous pressure of oxygen
VLU:	venous leg ulcer

INTRODUCTION

Leg ulcers are a common problem, with an estimated prevalence of 1% to 2% in the adult population, similar to the prevalence of psoriasis and alopecia areata.[1-3] With shifting demographics toward an aging population, sedentary lifestyles, an increased prevalence of obesity, and the emergence of various chronic diseases, leg ulcers will likely continue to be a significant burden on the health care system.[4,5] Despite the myriad potential causes of leg ulcers, a majority are caused by vascular abnormalities, with venous disease being the most common cause. Recent advances in the diagnosis and treatment of leg ulcers combined with the latest available evidence indicate a need for an update on this topic. A systematized approach and the judicious use of advanced expensive therapeutics are critical. The aim of this continuing medical education article is to provide an update on venous leg ulcers (VLUs). Part I is focused on the diagnostic approach to VLUs. In part II, the current medical and surgical management options will be reviewed.

EPIDEMIOLOGY

Key points

- **Approximately 1.5 to 3 per 1000 adults have active leg ulcers in North America**
- **Venous leg ulcers are more common in elderly patients, but 22% of individuals develop their first venous leg ulcers by 40 years of age, and 13% by 30 years of age**

The overall incidence of venous disease has been documented to be 76.1 per 100,000 person-years.[6] It is estimated that approximately 1.5 to 3 per 1000 North American adults have active leg ulcers.[7] Although chronic leg ulcers may be caused by many pathologies, upwards of 70% are related to venous disease, and approximately 20% are caused by arterial insufficiency or mixed arteriovenous disease.[8,9] The annual prevalence for individuals 65 to 95 years of age is reported as 1.69%; the overall male incidence is 0.76% and the female incidence is slightly higher (1.42%).[10] Previous epidemiologic studies identified a number of risk factors for venous disease, including the following: advanced age, female sex,[11,12] a family history of leg ulcers, non-Hispanic white race, obesity, a history of deep venous thrombosis (DVT) or phlebitis, previous serious traumatic leg injury, chronic lower extremity edema, a sedentary lifestyle, and any occupation requiring prolonged long periods of standing.[13-15]

Although VLUs are more common in elderly patients, 22% of individuals develop their first VLUs by 40 years of age and 13% before 30 years of age, affecting their ability to work and participate in social activities. As a result, many patients living with chronic leg ulcers experience a diminished quality of life, acute and chronic pain, and associated physical disabilities.[16] While upwards of three quarters of VLUs heal after 6 months, the annual reported recurrence rates range from 6% to 27%.[17,18] High recurrence rates may be attributable to persistence of underlying disease and a number of psychosocial and economic factors.[7,19] However, even when best practice pathways are implemented, only 50% to 75% of leg ulcers achieve complete healing after 6 months of treatment.[20]

PATHOPHYSIOLOGY OF VENOUS DISEASE/VENOUS ULCERS

Pathogenesis of venous disease

Key point

- **Valve dysfunction, outflow obstruction, arteriovenous malformation, and calf muscle pump failure contribute to the pathogenesis of venous disease**

The venous system is constructed like a ladder, with deep and superficial veins forming the 2 sides connected by perforator veins as the rungs (Fig 1). The calf muscle pump acts as a "peripheral heart," propelling venous blood toward the heart during calf muscle pump contraction. Unidirectional valves in the vein allow blood flow in 1 direction toward the heart and prevent reverse flow or reflux. However, pooling of venous blood (venous disease) can occur if: (1) the valves are damaged from congenital conditions, trauma, recurrent infection, or inflammation caused by a DVT resulting in reverse flow or leakage around the closed valves; (2) there is obstruction associated with previous clotting with a DVT, or outflow obstruction caused by obesity, pregnancy, or a pelvic mass/growth; (3) arteriovenous malformations as a congenital disease composed of abnormal connections of arteries and veins; or (4) the calf muscle pump is ineffective because of muscle wasting, immobility, or limited

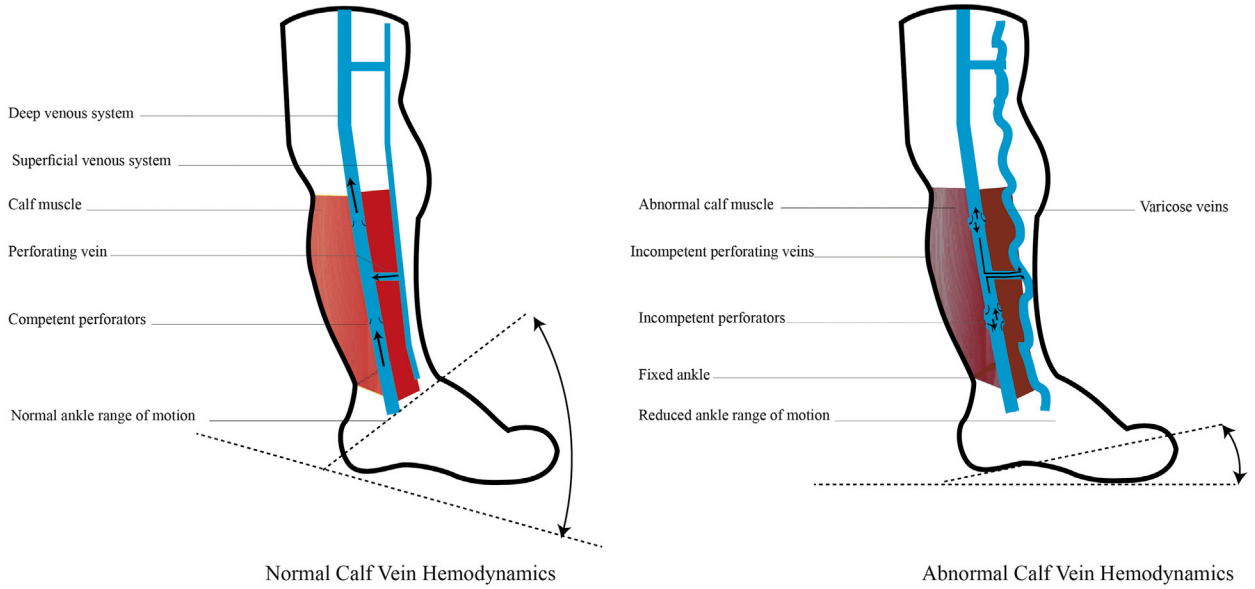


Fig 1. Venous disease. Calf vein hemodynamics.

ankle mobility (eg, neuromuscular disease, arthritis, or previous injury).

The calf muscle provides an important mechanism to propel venous blood flow toward the heart. During calf muscle pump contraction, the deep veins empty and blood flows from the superficial to the deep veins through the perforating veins. Venous pressure therefore drops during ambulation.[21-23] Venous reflux or obstruction—either superficial or deep—is linked to venous disease and the clinical manifestations of chronic insufficiency. An elevated venous pressure creates a retrograde buildup of pressure into the venules of the skin, leading to sustained increased ambulatory venous pressures (also called venous hypertension) that has been associated with development of leg ulceration.[23]

None of these theories fully explain the underlying pathophysiology that leads to venous leg ulceration, including fibrin cuff theory, inflammatory trapping, and altered cytokine, growth factor, and matrix metalloproteinase profiles.[24] None of these theories fully explain the observed pathophysiology.

In 1982, Burnand et al[25,26] postulated that an elevated intravascular pressure stretches the vascular wall and dilates the endothelial pores, allowing extravasation of red blood cells into the dermal tissue, promoting hemosiderin deposition and hyperpigmentation of the skin. Fibrinogen leaks into the interstitium and polymerizes to become fibrin that gathers around the capillary walls in bands called "fibrin cuffs."[25,26]

It has been speculated that these fibrin cuffs cause skin changes by compromising oxygen diffusion and entrapping various growth factors (ie, the growth factor trap hypothesis).[27] In addition, venous hypertension leads to margination and activation of various inflammatory cells (ie, the white cell trap hypothesis). Histologic studies indicate that venous disease is associated with the accumulation and adhesion of macrophages and T-lymphocytes in the perivascular and dermal matrix. A number of phospholipids, including phosphatidylcholine, a chemokine for macrophages responsible for a cascade of inflammatory responses, can be found attached to incompetent valves. High levels of interleukin (IL)-1 alfa, IL-1 beta, and tumor necrosis factor-alfa can be found in the wound fluid of difficult to heal VLUs. Other abnormalities associated with chronic recalcitrant VLUs include the upregulation of protease activity, especially matrix metalloproteinase, abnormalities of erythrocyte innate immunity, and factor XIII—mediated inhibition of fibrinolysis.[28,29]

Pathogenesis of nonhealing wounds

Key point

- **The relationship between inflammation and protein expression in venous leg ulcers is not clear**

Genetic expression of beta catenin, c-myc, and properdin are elevated in keratinocytes of nonhealing VLUs. However, the relationship between inflammation and genetic expression in VLUs is not clear.[30,31] Transforming growth factor beta (TGF-β) receptors are downregulated in tissue samples from patients with chronic VLUs.[32] The attenuation of TGF-β signaling leads to activation of the small body

size against decapentaplegic signaling cascade and a subsequent loss of tissue hemostasis with associated hyperproliferation.[32] These finding express an abnormal gene signature often related to the refractory wounds along with the role of cytokines and inflammasomes or large proteins, which are potent inducers of ILs-1β and -18 during inflammation.[33]

Cost

Key points

- **Chronic wounds in general are responsible for $7 billion per year in annual health care costs worldwide, with venous leg ulcers being the most common type of leg ulcers**
- **The average cost for a venous leg ulcer is estimated as $16,000 per treatment episode in the North American population**
- **The prevalence of venous leg ulcers in the elderly is as high as 1% to 2% of the elderly population in North America**

As the population ages, the prevalence and economic burden of VLUs is increasing.[10,34,35] The annual health care system cost of VLU management in the United States has been estimated at $1.5 to $3.5 billion.[17,36]

Based on this analysis, the average cost for VLU therapy is estimated at $16,000 per treatment episode.[36] The total annual cost of the treatment of VLUs has been reported as $25 million in Scandinavia and $200 million in England.[1,37]

In a recent study, patients with VLUs consumed more medical resources compared to non-VLU patients, with more days missed from work and a 29% higher cost from lost work.[38] Chronic wounds in general are responsible for $7 billion per year in annual health care costs worldwide, with VLUs being the most common cause of leg ulcers.[10,39] The major costs have moved from inpatient costs to outpatient care and nondrug treatments; nursing visit times and bandaging systems are the most expensive components.

Spectrum of disease

Key point

- **Patients with venous disease have a spectrum of skin presentations from edema, hemosiderin staining, venous eczema, venous starburst of veins radiating distally from the medial malleolus (ie, corona phlebectatica paraplantaris), lipodermatosclerosis, and atrophie blanche along with ulcer formation**

Chronic venous disease refers to a spectrum of changes ranging from varicose veins and hyperpigmentation to stasis dermatitis to lipodermatosclerosis (LDS) and VLUs.[24] VLUs commonly present as shallow ulcers in the gaiter area—the area extending from midcalf to approximately 1 inch below the malleolus.[10,28] Patients with venous disease have a spectrum of skin presentations ranging from edema (pitting or nonpitting), hemosiderin staining, venous eczema, venous starburst of veins radiating distally from the medial malleolus (ie, corona phlebectatica paraplantaris [Fig 2]), LDS, and atrophie blanche along with ulcer formation.[40]

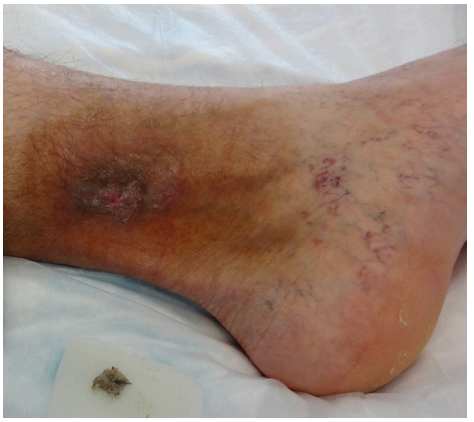


Fig 2. Corona phlebectatica paraplantaris or blood vessel burst extending distally from medial ankle.

In order to standardize the reporting and comparing of the diverse manifestations of chronic venous disorders, a comprehensive classification system (clinical, etiology, anatomy, and physiology [CEAP]) has been developed. An international committee of the 1994 American Venous Forum introduced a simplified classification for chronic venous disorders. CEAP is clinically used for those in vascular medicine but has not yet been validated or linked to clinical outcome.

The fundamentals of the CEAP classification include a description of the clinical class (C) based upon objective signs, the etiology (E), the anatomic (A) distribution of reflux and obstruction in the superficial, deep, and perforating veins, and the underlying pathophysiology (P), whether caused by reflux or obstruction.[41] Although CEAP classification has not been validated or been shown to correlate with outcome, it does provide a framework to classify disease (Table I).

LDS, or inflammation of the skin and fatty tissue causing woody changes in the dermis (C4), is part of this spectrum and presents as indurated plaques of the lower extremities (Fig 3). Acute phase LDS is exquisitely painful and is commonly misdiagnosed as cellulitis, phlebitis, inflammatory morphea, or other panniculitides. The chronic phase of LDS presents with induration and fibrosis that usually

Table I. Clinical, etiology, anatomy, and physiology classification of chronic venous disease

Clinical classification	Etiologic classification	Anatomic classification	Pathophysiology
C0: No visible or palpable signs of venous disease	Ec: Congenital	As: Superficial veins	Pr: Reflux
C1: Telangiectases or reticular veins	Ep: Primary	Ap: Perforating veins	Po: Obstruction
C2: Varicose veins	Es: Secondary	Ad: Deep veins	Pr,o: Reflux and obstruction
C3: Edema	En: No venous cause identified	An: No venous location identified	Pn: No venous pathophysiology identified
C4a: Pigmentation or eczema			
C4b: Lipodermatosclerosis or atrophie blanche			
C5: Healed venous ulcer			
C6: Active venous ulcer			
S: Symptomatic, including ache, pain, tightness, skin irritation, heaviness, and muscle cramps, and other complaints attributable to venous dysfunction.			
A: Asymptomatic.			

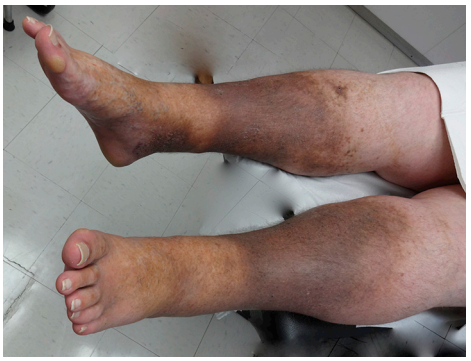


Fig 3. Lipodermatosclerosis. Note the inflammation of the skin presenting as indurated fibrosing plaques of the lower extremities.

precedes any associated ulceration.[28] If LDS is left untreated, complications may arise and the woody fibrosis may extend distally to the feet and toes, eventually resulting in venolymphedema (and association of venous disease and lymphedema).

VLUs commonly present as shallow ulcers in the area extending from the midcalf to approximately 1 inch below the malleolus (ie, the gaiter area; Fig 4).[10] The border is often serpiginous, with an irregular shape, and the wound base generally has a preponderance of pink granulation and yellow fibrinous tissue. Most patients have edema and discomfort or aching that is worse at the end of the day and may be exacerbated by dependency.

Differential diagnoses of venous disease/ venous leg ulcers

Key points

- **Approximately 10% of lower extremity wounds are atypical with less common etiologies**

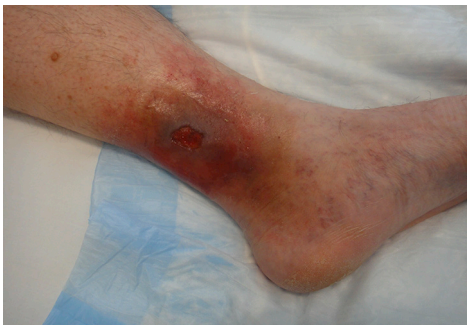


Fig 4. Venous leg ulcers. Note the shallow ulcer with a yellow fibrinous base that is commonly found on the medial aspect of the ankle.

- **Histology is often essential for the diagnosis of an atypical wound**

Although the majority of chronic wounds are caused by vascular, neuropathic, and pressure etiologies, the early diagnosis of atypical wounds is critical. An estimated 10% of lower extremity wounds are caused by less common etiologies, including infections, skin cancers, metabolic disorders, inflammatory processes, and other diagnoses. Fig 5 lists the differential diagnoses for painful ulcers.

Histology is often essential for the diagnosis of an atypical wound. In a retrospective study of 350 biopsy specimens obtained from chronic wounds, 29.7% were identified as atypical, with malignancy detected in 24 patients (17%).[9] In a study on 144 patients with VLUs, Senet et al[42] obtained at least two 6-mm punch biopsy specimens, 1 at the wound edge and 1 in the wound bed, in the most clinically suspicious areas. In this study, the overall frequency of skin cancer in patients with chronic leg ulcers was reported to be as high as 10.4%.[42]

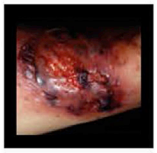
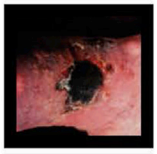
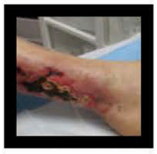
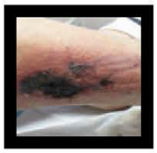
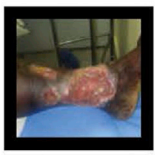
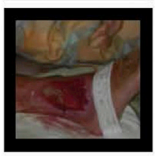
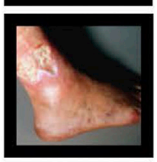
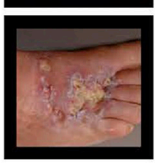
Picture	Painful ulcers	Location	Characteristics	Common associations	Diagnostics	Management
	**Pyoderma gangrenosum**	any	inflammed, undermined elevated edges, sterile pustules	inflammatory bowel disease,hematoproliferative disorders, rheumatoid arthritis, antineutrophilic cytoplasmic antibody positive vasculitis	history and clinical characteristics	topical steroid [39,21] topical tacrolimus [39,21] oral corticosteroids [39, 40] cyclosporine [39,40,21] anti TNF alpha [21] mycophenolate Mofetil [39,40]
	**Vasculitis**	dependent areas	palpable purpura, atrophie blanche, livedo reticularis, pustules	autoimmune disorders, cryoglobulinemia, infections	biopsy, urinalysis, medical work-up	oral corticosteroids [41, 43] cyclosporine [41] dapsone [41] colchicine [41-43] methotrexate [42] azathioprine [42]
	**Martorell ulcer**	posterior lateral overlying achilles tendon	livedo reticularis necrosis	hypertension, diabetes, vitamin K antagonists	biopsy	surgery [8,11] skin graft [8,11] control of blood pressure [11] prostaglandin E [135] platelet-derived growth factor [12] hyperbaric oxygen [44]
	**Calciphylaxis**	any	livedo reticularis necrosis	chronic renal failure, hyperparathyroidism, kidney transplant recipients, warfarin	biopsy, history	control of hyperparathyroidism or parathyroidectomy [45] Sodium thiosulfate [45-47] warfarin/enoxaparin [45-47] hyperbaric oxygen [45-47] tissue plasminogen activator [48]
	**Sickle cell ulcer**	medial ankle	very painful polycyclic wound, absence of vital granulation tissue	sickle cell anemia	history, sickle cell prep	blood transfusion [49,50] hydroxyurea [49-51,52] arginine butyrate [53] nitric oxide [53] Hypomethylating agents [53] compression therapy local wound care
	**Arterial leg ulcer**	around lateral malleolus, pretibial area, dorsum of foot and toes	punched out skin defect, often with eschar and/or necrotic border	coronary arterial disease, intermittent claudication, rest pain	ankle-brachial-index(ABI), duplex, angiography	Revascularization [28] Medical therapy [28] local wound care pain management
	**Hydroxyurea ulcer**	medial and lateral ankle	painful progressive skin defect with white ulcer base and absence of vital granulation tissue	history of essential thrombocytosis, polycythemia, vera, myeloproliferative disorder	history of medication use	cessation of hydroxyurea [54,4] skin substitutes [54] compression therapy local wound care pain management
	**Anti-phospholipid syndrome**	any	necrotic ulcer livedoreticularis	livedo racemosa, purpura, ecchymosis, acrocyanosis,raynaud's, venous thromboembolism, arterial thrombosis, fetal loss	IgG/IgM anticardiolipin antibody, lupus anticoagulant, VDRL, beta2-glycoprotein 1	Anticoagulants [55] systemic corticosteroids [56] intravenous Immunoglobulins [57]

Fig 5. Differential diagnosis of painful leg ulcers. (The authors acknowledge Professor Jurge Hafner's help in making the table.)

As the population ages, the rate of peripheral arterial disease is increasing. Based on several studies, a venous etiology was identified in >50% to 75% of leg ulcers, with mixed arteriovenous ulcers accounting for ≤15% of the remaining patients.[23,43]

APPROACH TO VENOUS LEG ULCERS

Key points

- **A comprehensive history and physical examination are essential in the evaluation of chronic venous insufficiency and ulceration**
- **Varicose veins in the medial thigh and calf may implicate involvement of the great saphenous vein; dilated veins in the anterolateral thigh and knee are related to anterior saphenous insufficiency**

A comprehensive history and physical examination are required for an accurate diagnosis (Fig 6). Information should include symptoms, exacerbating and alleviating factors, medical history (particularly of previous DVTs), and other clotting disorders. Coagulation disorders are a significant but often

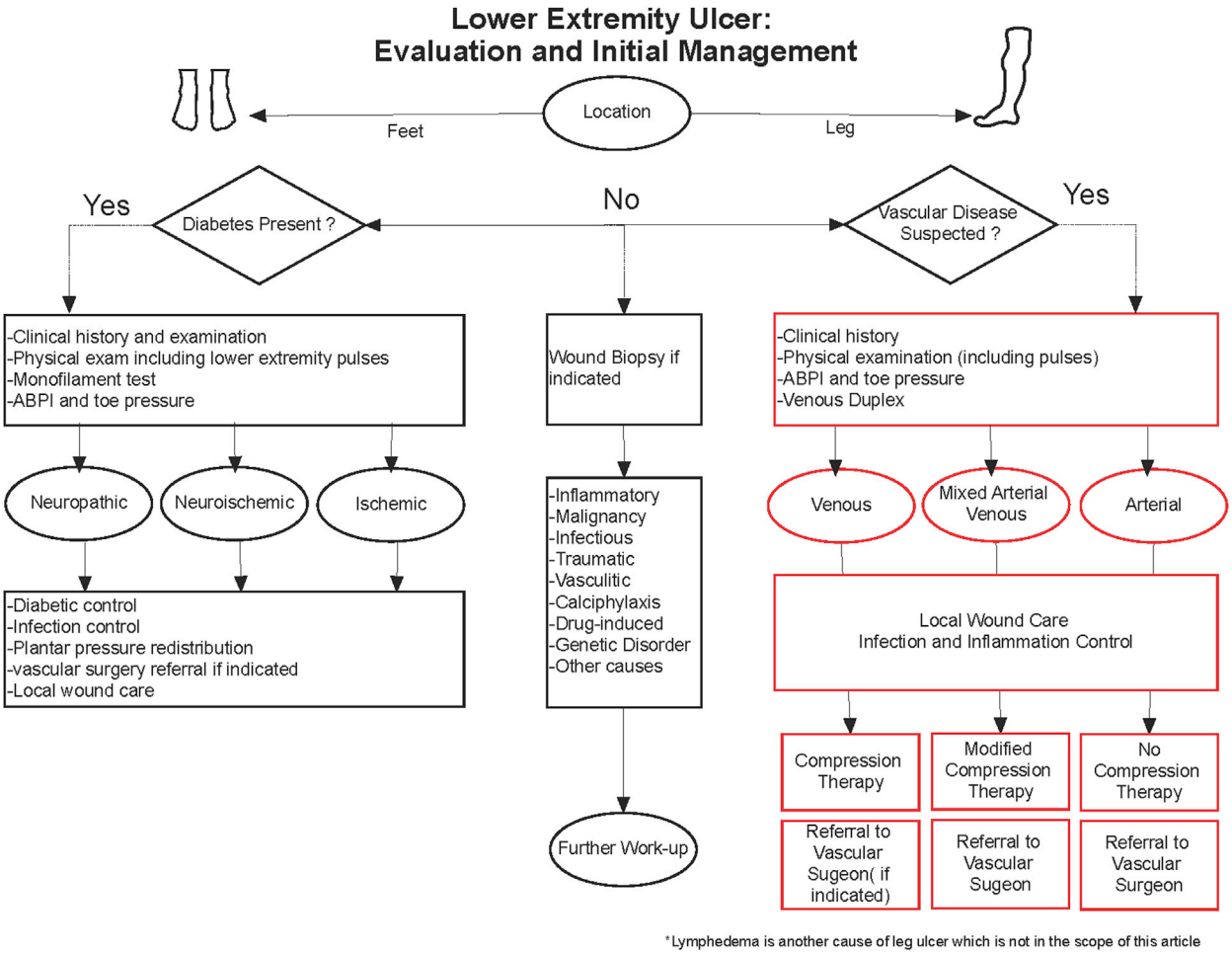


Fig 6. Evaluation and initial management of a lower extremity ulcer.

unrecognized risk factor for venous disease, especially in younger patients.[44,45] Factors that affect the calf muscle pump, such as arthritis, should be documented. Typical complaints of VLUs include: aching and heaviness, often at the end of the day, fatigue, restless legs, night cramping, and itching often associated with venous eczema and swelling. Symptoms can be exacerbated by prolonged standing or sitting, and difficult to control edema may be affected by the menstrual cycle. It is important to carefully assess patients with VLUs to be sure that the symptoms are in fact originating from chronic venous disease and not coexisting conditions, such as cardiac failure. A physical examination focusing on skin changes, limb size and shape, and vascular assessment is critical. The pattern of varicosities provides a clue to the possible location of venous disease. Varicose veins in the medial thigh and calf may implicate involvement of the great saphenous vein; dilated veins in the anterolateral thigh and knee are related to anterior saphenous insufficiency.[46] Other clinical findings may include edema, hemosiderin pigmentation, LDS, and venous eczema.

The arterial system should be assessed in all patients, including palpation of pulses, assessment of foot temperature, and measurement of the ankle brachial pressure index. Patients should also undergo testing for peripheral neuropathy and ankle range of motion.

Margolis et al[47] found an association of VLUs with other comorbidities, and it is therefore important for physicians to consider a proper assessment for associated diseases, including anemia, diabetes mellitus, and depression.[47]

Assessment of factors that delay healing

Key points

- **Failure of venous leg ulcer healing has been correlated with a larger initial area of the wound, longer duration of the wound, history of venous ligation or vein stripping, history of hip or knee replacement surgery, an ankle brachial pressure index <0.8, and >50% of the wound being covered by fibrin**
- **Large wound size, nonadherence to compression therapy, the involvement of**

all 3 venous valvular systems (ie, superficial, perforating, and deep venous), older age, being overweight (a body mass index ≥25 kg/m^2), and history of deep venous thrombosis are associated with nonhealing and venous leg ulcer recurrence

It is important to assess risk factors for the development of leg ulcers and factors that delay the healing of VLUs. VLUs at the highest risk for nonhealing could potentially benefit from the advanced therapies to treat stalled but healable venous ulcers. Failure of VLU healing has been correlated with a larger initial area of the wound, longer duration of the wound, a history of venous ligation or vein stripping, a history of hip or knee replacement surgery, an ankle brachial pressure index <0.8, and >50% of the wound covered by fibrin (firm yellow base).[1,34] Franks et al[48] studied 411 patients with nonhealing VLUs and determined associations with larger wound size, longer wound duration, poor lower limb joint mobility, and general immobility.[48] The rate of recurrence for VLUs has been reported to be as high as 37% at 3 years and 48% at 5 years.[49]

In an unselected group of 157 VLUs studied by Labropoulos et al,[49] 80% of the ulcers healed (131/157) with a proper diagnosis, appropriate surgical intervention, adequate compression therapy, and optimal local wound care. Triple venous system disease or involvement of superficial, perforating, and deep veins was the greatest risk factor associated with nonhealing. These findings suggest a potential role of venous valvular surgery to correct incompetence.[18,49]

In summary, a number of factors have been identified to be associated with nonhealing and recurrent VLUs: larger wound size, nonadherence to compression therapy, triple venous valvular system disease, older age, overweight patients with a high body mass index (>25 kg/m^2), and a history of DVT.[49] A history of DVT was detected in 60% of patients with VLUs; however, more patients may have an undiagnosed DVT. Patients with nonhealing ulcers (refractory to 6 months of treatment) had a 5-fold greater chance of having a history of DVT.[49] Patients with VLUs are commonly overweight and also have a relative nutritional deficiency that needs to be addressed.[3] Environmental factors, especially cold temperatures, may also play an important role in the onset of chronic leg ulcers.[23] VLU prevalence shows a reduction in VLUs in summer and a rise in VLUs in winter. In addition, there was a statistically significant negative correlation between higher temperatures and new ulcer onset.[23]

Wound assessment

Key points

- **Accurate and consistent wound measurement is important to monitor the healing rate**
- **If a wound is not 30% smaller by week 4 it is unlikely to heal by week 12, and the patient should be reassessed for the appropriate diagnosis and management**

The documentation of the wound location, wound area, and characteristics is important for the monitoring of healing and treatment effectiveness. Wound assessment requires an accurate measurement that is precise, user-friendly, and reproducible.[50]

Traditional wound area measurements include measuring length and width in perpendicular distances of wound borders (ie, the longest length with the greatest width at right angles), manual tracing, and digital photography. These methods are inconsistent and sometimes inaccurate. Wound tracings that calculate the area via digital software are slightly better than linear measurement.[51,52] The wound surface is measured by a tracing of the wound surface on a sterile disposable contact layer and then calculating the wound area to provide more accurate and reproducible wound measurement. Software programs can also calculate wound dimensions from a photograph of the lesion.[53] This method avoids any contact with the surface of the wound, reducing potential pain and bacterial contamination of the wound surface.

Many observational studies support the correlation between improvements of the geometric parameters of the wound margin, a healthy wound bed, and progression to wound healing.[54] Cardinal et al[54] reported that wounds with symmetrical convex geometries (ie, oval or circular wounds) at baseline heal better than wounds with large concavities, multiple segments, and skin islands at the margins. In summary, wound documentation is important for documenting healing rates. If a wound is not 30% smaller by week 4 it is unlikely to heal by week 12, and the patient should be reassessed for the appropriate diagnosis and management.[55] Stalled but healable chronic wounds are ideal candidates for advanced therapies.

Wound biopsy

Key points

- **The proper site from which to obtain the biopsy specimen depends on the etiology of the wound and appropriate selection of the biopsy site**

Table II. Common stains and markers used in wound pathology*

Stains and markers	Function
Hematoxylin–eosin	Regular
Periodic acid–Schiff	Vessel walls and basement membrane, fungi
Phosphotungistic acid-hematoxylin	Stains muscle fibers and fibrin, especially fibrin thrombi
Verhoff–van Gieson	Stains elastin to differentiate a venule from an arteriole
von Kossa stain	Identify the calcium deposits.
Fite	Leprosy/acid-fast bacilli
Zeihl–Neelsen	Tuberculosis/acid-fast bacilli
Giemsa	Leishmaniasis
Perl potassium frocyanate	Hemosiderin
CD31	Endothelial marker for vascular lesions
D2-40	Detects podoplanin in lymphatic endothelial cell
Factor VIII	a marker for mast cells and platelet thrombi

*Data from Labropoulos et al[49] and Cardinal et al.[54]

- **Multiple biopsy specimens are occasionally required for difficult diagnostic situations, and especially for the detection of localized malignancy**

Obtaining a biopsy specimen of a wound is an easy and helpful diagnostic procedure to identify less common etiologies if a wound has unusual presentation, is in an unusual location, if malignancy is suspected (such as nonhealing wounds in burn scars), and in wounds that fail to heal after standard care.[9,56]

Wound biopsy specimens provide valuable diagnostic histologic findings, including the diagnosis of malignancy, infection, and other causes.[56] In some cases, evaluation may include tissue culture or obtaining an additional biopsy specimen for immunofluorescence to detect immune complexes (ie, vasculitis, connective tissue disease, or inflammatory skin disorders) using specialized transport media.[56] The proper site from which of obtain the biopsy specimen depends on the etiology of the wound and appropriate selection of the biopsy site. Multiple biopsy specimens are occasionally required for difficult diagnostic situations, especially for the detection of localized malignancy. The preferred techniques for obtaining wound biopsy specimens are punch or elliptical biopsy specimens taken from the edge of the ulcer to compare the ulcerated area and surrounding skin.[50] Hematoxylin–eosin is the most widely used stain in wound pathology, but the selection of special stains depends on the differential diagnosis (Table II).[50,57]

The biopsy specimen—usually taken from the center of the wound—should be sent for culture to rule out viral, bacterial, fungal, and atypical infections. Atypical mycobacterial and deep fungal infections characteristically occur in immunosuppressed individuals or because of direct inoculation. Special staining can be used to identify the organism and direct appropriate and selective antimicrobial therapy.[9]

There is a reluctance to obtain a biopsy specimen from a patient with LDS because of the risk of nonhealing.[40,58] The biopsy specimen is commonly consistent with sclerosing panniculitis, and the most dramatic changes occur in the subcutaneous fat. However, the clinical picture of LDS is protean, and the limited biopsy specimens obtained show a variety of changes based on the stage of the disease. The presence of other pathologies, such as pyoderma gangrenosum or vasculitis, as the basis of venous disease is not unlikely.

VASCULAR ASSESSMENT: ARTERIAL AND VENOUS

After a comprehensive clinical assessment, subsequent noninvasive and sometimes invasive investigations may be indicated to confirm the diagnosis and plan treatment options. Assessment of the arterial system to rule out mixed arteriovenous disease is important. Up to 25% of patients with a VLU have concomitant peripheral arterial disease.[59,60]

Investigation of the arterial system

Investigation of the arterial system includes a review of both micro- and macrocirculation. Microcirculation assessment includes transcutaneous oxygen saturation ($TcPO_2$), laser Doppler flowmetry, and transcutaneous carbon dioxide saturation ($TcPCO_2$) measurements and capillaroscopy. Macrocirculation assessment includes the ankle brachial pressure index (ABPI) and toe pressure, Doppler arterial waveforms, duplex ultrasonography, angiography, and magnetic resonance imaging.

Table III. Arterial measurements related to vascular supply of the leg*

ABPI	Toe pressure	Toe brachial index	Ankle Doppler wave form	Diagnosis
>0.8	>80 mm Hg	>0.6	Normal/triphasic	No relevant arterial disease
>0.5	>50 mm Hg	>0.4	Biphasic	Some arterial disease: modify compression
>0.4	>30 mm Hg	>0.2	Biphasic/monophasic	Arterial disease predominates
<0.4	<30 mm Hg	<0.2	Monophasic	High risk for limb ischemia

ABPI, Ankle brachial pressure index.
*Data from Sibbald et al.[68]

Microcirculation/transcutaneous oxygen

Key points

- **Transcutaneous oxygen saturation, laser Doppler flowmetry, and transcutaneous carbon dioxide saturation are sensitive indicators of microcirculation**
- **The transcutaneous oxygen saturation measurement reflects oxygen supply to the end organ (the skin) by the combination of the macro- and microcirculation**

Different techniques can evaluate microcirculation in patients with wounds. $TcPO_2$, laser Doppler flowmetry, and $TcPCO_2$ measurements and capillaroscopy have been used clinically.[61] Transcutaneous oximetry measures tissue oxygenation in superficial skin layers by placing an electrode on the skin surface. $TcPO_2$ is correlated with arterial oxygen pressure in neonatal skin, but the variation in adult skin thickness interferes with this linear relationship.[61,62] However, this method should not be used in isolation. The values are influenced by edema, skin temperature, and infection.[62] $TcPO_2$ is an important value, especially because it reflects the net oxygen supply to the end organ (the skin) by macro- and microcirculation.

Macrocirculation

ABPI and toe brachial pressure index. ABPI is a noninvasive screening tool that offers 85% sensitivity and 97% specificity to detect arterial occlusive disease.[63] The ankle systolic blood pressure alone reflects the amount of blood flow to the ankle that is influenced by central blood pressure. The ankle pressure is divided by the best estimate of the central pressure (the higher of the 2 brachial blood pressures), which results in the ABPI (ie, ABPI is equal to the ankle Doppler pressure [for each leg] divided by the highest brachial Doppler pressure).

Normal ABPI measurements range from 0.9 to 1.3.[64,65] In general, indices <0.8 signify some arterial disease and may be associated with intermittent claudication; indices <0.5 indicate severe ischemia. The upper cutoff point of 1.3 has been generally accepted. Allison et al[66] identified a strong association of cardiovascular disease with ABPI >1.4, which is most commonly seen in those with noncompressible calcified vessels.

The ABPI Doppler measures macrovascular arterial disease and may overestimate the true pressure reading in patients with arterial calcification and advanced atherosclerosis caused by diabetes mellitus.[64,67,68] About 80% of patients with diabetes and 20% of nondiabetic patients have calcified, noncompressible arterial vessels.[67] In these cases, ankle vessels are not compressible with a cuff—such that any ABPI >1.3 may be related to spuriously elevated ankle pressures. These individuals have unreliable ABPI measurements, and a direct toe systolic pressure (or toe brachial pressure index) is more reliable because the digital arteries are rarely heavily calcified (Table III). The current standard for detection of significant peripheral arterial disease is an ABPI <0.8 or a toe pressure <55 mm Hg.[68]

Doppler arterial waveforms. Doppler arterial waveforms can be attained at the same time as assessment of the ankle systolic pressure with the Doppler probe. A normal waveform is triphasic and becomes biphasic if mild disease exists proximal to the probe insonation placement site and monophasic in the presence of more severe disease.

The Doppler pulse wave is influenced by arterial distensibility and arterial stiffness. The pulse waves can be either detected by audible Doppler or documented by pulse wave velocity. A higher pulse wave velocity correlates with the stiffer arterial wall. The relationship between ABPI and wave velocity has been studied.[69]

Duplex ultrasonography. Duplex ultrasonography is a noninvasive, accurate test for the evaluation of the flow in the arteries and veins. It provides accurate noninvasive information relating to cross-sectional areas and provides the ability to view vessel walls in a longitudinal plane from several angles.

Duplex ultrasonography detects changes in the velocity of red cells. Duplex ultrasonography imagers are now available as relatively

inexpensive lightweight, portable machines. This high level of sensitivity may allow the noninvasive duplex technique to replace angiography as the criterion standard in these patients.[34] Experienced vascular technologists working in an accredited vascular laboratory provide expert vascular images for meaningful interpretation. Partnership with a high-quality vascular laboratory is important to obtain useful arterial and venous measurements.

Angiography. Angiography remains the criterion standard for arterial assessment because of its ability to successfully outline the entire arterial system. It has an advantage over various noninvasive technologies for patients who are obese, individuals with extensive vessel calcification, and in the presence of bilateral diffuse atherosclerotic disease or arteriovenous malformations. The assessment of pressures across a stenotic vascular lesion provides accurate hemodynamic information of the pressure gradient created by the lesion and, therefore, the severity of the arterial stenosis. One of the primary criticisms of angiography is the production of anatomic images to represent a functional deficit. Intra- and interoperator interpretation are also variable. Direct mortality rates caused by cardiac arrest or stroke are low; however, significant morbidity can arise from bleeding from the arterial puncture site, cholesterol and thrombus embolization to the legs, arterial wall dissection, contrast dye allergy, renal failure from the dye related hemoconcentration, and arteriovenous fistula formation. These complications prohibit the frequent use of angiography either for diagnostic or follow-up purposes. Therefore, angiography is now primarily reserved for preoperative evaluation, interventional procedures, thrombolysis, and emergency situations where other modalities are not available.

Magnetic resonance imaging. A magnetic resonance imaging scan using gadolinium to enhance the contrast is a diagnostic tool that can detect small and large vessels and the ability of the perfused tissue to extract oxygen. Increasingly detailed anatomic information may be obtained using computed tomography of magnetic resonance angiography. Between duplex ultrasonography and computed tomography or magnetic resonance angiography, the diagnosis of the severity of arterial disease and its location can be determined. Percutaneous angiography is only performed to define a target vessel for surgical bypass.

Investigation of the venous system

Investigation of the venous system can be conducted using venous Doppler ultrasonography, color flow duplex ultrasonography, air plethysmography, or venography.

Venous Doppler ultrasonography. Venous Doppler ultrasonography with the use of a handheld Doppler unit for assessment of the venous system is not recommended because of the higher sensitivity and specificity of duplex ultrasonography.

Color flow duplex ultrasonography. This highly operator-dependent, noninvasive test provides both anatomic and flow data, allowing for detailed and accurate assessments of reflux and patency within individual veins. The accuracy of duplex ultrasonography for defining venous reflux in the deep, superficial, and perforator veins is important. With duplex ultrasonography, the operator can first identify the vein in question with the ultrasound function, and then investigate the specific vein for reflux or obstruction—this is not possible with handheld duplex ultrasonography.

This test has the capacity to distinguish thrombus age, clot mobility at the tip of the clot, and the length of the free-floating tail of the clot. Venous thrombus can be detected in the large vessels down to the level of the popliteal vein, and good visualization of the deep and superficial calf veins is possible. Recent studies reported that duplex scanning is the best test to monitor thrombi and to check reflux after DVT. The exact localization of reflux within the superficial and deep systems can also be determined using this technique. This procedure is different than the procedure used to evaluate DVT. However, it is important to remember that an anatomic diagnosis of venous disease by duplex ultrasonography does not always correspond to a clinical diagnosis.[13]

Air plethysmography. This simple, noninvasive test quantitatively assesses venous reflux, obstruction, and poor calf muscle pump function. A polyurethane tubular air chamber surrounds the entire leg and is connected to a computer. Readings are taken throughout the procedure, and small changes in limb volume reflecting an increased venous blood volume are detected electronically through the use of an air-filled chamber or sleeve surrounding the limb. Plethysmography is able to identify abnormal venous function by rapid refilling of the venous tree, resulting in leg swelling with limb dependence that can be caused by either abnormal reflux or obstruction. This test is almost always abnormal in cases of venous ulceration.[70]

Venography. Venography is an invasive test that has been replaced by color flow duplex ultrasonography for most clinical indications. This technique provides additional information on thrombus age, valve damage, and a much wider view of the venous system for reconstructive surgery. In cases of venous

obstruction, it provides a road map for stenting procedures to reopen obstructed venous segments. The risks include severe contrast media allergy, associated cardiac arrest, or acute renal failure.

In conclusion, venous ulcers are the most common leg ulcers and must be distinguished from other vascular ulcers, including mixed arteriovenous and arterial ulcers. The differential diagnosis of leg ulcers includes a wide range of entities that must be identified. When wounds are not healing at the expected rate in spite of treatment, skin biopsy specimens should be obtained and bacterial cultures might be helpful diagnostic tools. Specialized arteriovenous studies are most commonly noninvasive. The ABPI can be performed with a handheld Doppler unit at bedside, and duplex Doppler ultrasonography performed by the vascular laboratory can identify segmental defects and potential surgical candidates. Vascular laboratory studies of the venous system can also predict which patients will benefit from surgery. Successful leg ulcer management requires an interprofessional team to make a specific diagnosis and to assess the vascular supply and other modifiable factors for optimal healing.

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