

Colonization of Venous Leg Ulcers by Microorganisms

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SUMMARY The aim of the study was to identify the microorganisms that most commonly colonize leg ulcers. The study included 70 patients of both sexes, aged 31-85 years. There were 49 (70%) female and 21 (30%) male patients, mean age 69.16 and 61.62 years, respectively. Nineteen different types of microorganisms were isolated in study patients: one in 18, two in 38, and three pathogens in 13 patients. Only one of study patients had four microorganisms isolated at the time. The most common microorganisms were *Pseudomonas aeruginosa* found in 25 and *Streptococcus aureus* in 19 patients.

KEY WORDS: venous leg ulcers; colonization, microorganism; *Pseudomonas aeruginosa*; *Staphylococcus aureus*

INTRODUCTION

Venous hypostatic ulcer is an open wound, usually located on the lower leg skin damaged by chronic venous stasis. Chronic venous hypertension with consequential cutaneous capillary bed distention and increased capillary wall permeability plays a major role in the development of venous ulcer (1-3). The prevalence of venous hypostatic ulcer in the population has been estimated to 1% (4). Chronic wounds are not sterile as they are usually colonized but not necessarily infected by microorganisms, implying that antibacterial treatment may not always be required. In case of wound infection, pain and inflammation of the surrounding tissue occur, accompanied by pronounced exudation and suppuration, occasionally with fever and lymphadenitis. Although the process remains localized, healing of the wound is retarded. *Staphylococcus aureus*, alone or in combination with other gram-negative bacteria, is the bacterium most commonly isolated in venous leg ulcers (5).

PATIENTS AND METHODS

The study included 70 patients (49 female and 21 male) aged 31-85, with venous leg ulcers, hos-

pitalized at Department of Dermatology and Venereology, Dr. Josip Benčević General Hospital, Slavonski Brod. Wound swab was obtained after 24 hours of saline application. The specimen was obtained from the wound edge towards healthy skin or from the deepest part of the wound expected to contain the highest number of pathogens. The specimen was immediately transferred to Laboratory of Microbacteriology, Public Health Institute of the Brod-Posavina County for analysis. Specimens were put on a solid medium (blood agar and Salmonella Shigella Agar SS-agar), and on a liquid medium (thioglycolate medium, fluid). The specimen was incubated in aerobic conditions, thermo status, for 18-24 hours in an atmosphere with 5%-10% CO₂ at 35° C. History data showed the patients had not been taking any parenteral, *per os* or topical antimicrobial therapy for a week before the study.

RESULTS

The study included 70 patients, mean age 65.39 (age range 31-85) years. There were 49 (70%) female and 21 (30%) male patients, mean age 69.16 and 61.62 years, respectively (Figs. 1

Table 1. Number of pathogens isolated in study patients (N=155)

Pathogen	n
<i>Pseudomonas aeruginosa</i>	25
<i>Staphylococcus aureus</i>	19
<i>Staphylococcus epidermidis</i>	16
<i>Serratia marcescens</i>	16
<i>Pseudomonas</i> spp.	16
<i>Acinetobacter</i> spp.	12
Diphtheroids	12
<i>Streptococcus</i> spp.	6
<i>Streptococcus</i> β-hemolytic group G	5
<i>Proteus mirabilis</i>	5
<i>Morganella morganii</i>	4
<i>Escherichia coli</i>	3
<i>Enterobacter</i> spp.	3
<i>Klebsiella pneumoniae</i>	3
<i>Klebsiella</i> spp.	3
<i>Providencia</i> spp.	2
<i>Proteus vulgaris</i>	2
<i>Citrobacter</i> spp.	2
<i>Enterococcus</i> spp.	1

and 2). The largest number of patients were in the 7th and 8th decade of life. The duration of venous leg ulcers was found to considerably vary (Fig. 3). A total of 19 different pathogens were isolated in 155 isolates obtained from study patients (Table 1). *Pseudomonas aeruginosa* was the most common pathogen isolated in 25 patients, followed by *Staphylococcus aureus* found in 19, and *Staphylococcus epidermidis*, *Serratia marcescens* and *Pseudomonas* spp. in 16 patients each (Fig. 4). Less commonly isolated pathogens were *Enterococcus* spp., *Acinetobacter*, *Proteus vulgaris*, *Enterobacter* spp., *Klebsiella pneumoniae*, *Providencia* spp., *Klebsiella* spp. and *Citrobacter* spp.

Ulcers are usually contaminated by several pathogens. In our group, a single pathogen was isolated in 17 patients, whereas two pathogens were isolated in 38, three pathogens in 13, and four pathogens in only one patient (Fig. 5).

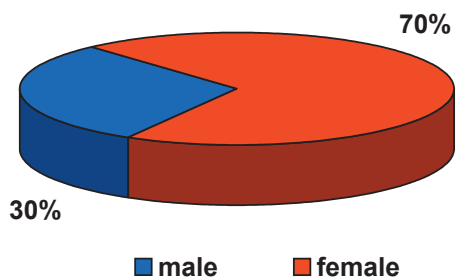


Figure 1. Sex distribution of pathogens isolated in study patients.

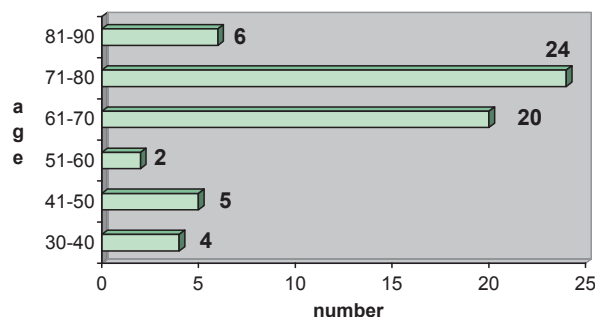


Figure 2. Age distribution of pathogens isolated in study patients.

DISCUSSION

Chronic venous ulcers are usually colonized by microorganisms, however, wound infection with the isolated pathogens is extremely rare and accompanied by a typical clinical picture. None of our patients had wound infection. *Pseudomonas aeruginosa* is the agent most commonly isolated from venous ulcers, followed by *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Serratia marcescens*, *Proteus* spp., etc. Eriksson *et al.* (5) report on *Staphylococcus aureus*, and Schraibmann (6) on gram-negative microorganisms as the most commonly isolated pathogens.

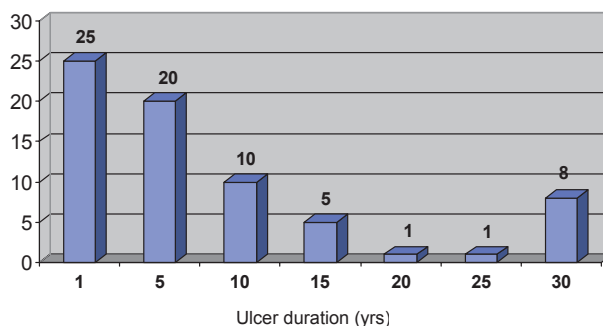


Figure 3. Distribution patients according to ulcer duration

Normal skin flora consists of a great variety of bacteria, fungi and protozoa. The composition of normal skin flora depends on the patient's general condition, age, pregnancy, nutritional status, personal hygiene, and living conditions. Normal skin flora usually contains the aerobes *Staphylococcus epidermidis*, *Staphylococcus saprophyticus*, *Streptococcaceae* and *Peptococcaceae*, and the anaerobes diphtheroids (7).

Staphylococcus aureus belongs to the family of gram-positive cocci, and is characterized by fast and easy development of resistance to antibiotics. *Staphylococcus aureus* produces many enzymes and exotoxins involved in the pathogenesis of in-

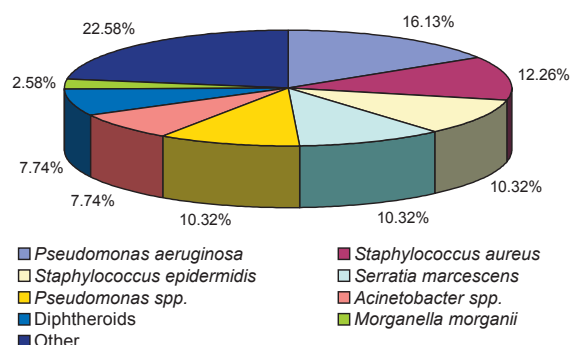


Figure 4. Percentage of pathogens isolated in study patients

fections. The characteristic staphylococcal infections are abscess, furuncle, carbuncle, wound infection, impetigo contagiosa and SSSS syndrome (8). *Staphylococcus aureus* is a common cause of nosocomial infections, especially in immunocompromised patients (8).

Pseudomonas aeruginosa is a gram-negative anaerobic bacterium. Although a usual saprophyte in humans, it rarely causes infection in immunocompetent, normal individuals. The infection caused by this microorganism is mostly associated with a failure in the body's mechanisms of defense, which may imply disruption of a physical barrier such as skin. *Pseudomonas aeruginosa* shows high affinity for skin defects, including superinfection, especially in hospitalized patients. The infection caused by this pathogen is characterized by blue-green discoloration and typical malodor. This infection may remain localized, however, fulminant spread of the infection has also been described (9).

Besides these most common causative agents of chronic wound contamination, those less frequently observed include *Enterobacteriaceae*, *Escherichia coli*, *Klebsiella* and *Proteus*, which were also found in our study patients (10). The rate of isolation of these pathogens is by far lower. Apart from *Staphylococcus aureus* and *Pseudomonas aeruginosa*, *Staphylococcus epidermidis* was most commonly detected in our patients, followed by *Serratia marcescens*, diphtheroids and *Pseudomonas spp.* *Acinetobacter spp.*, group G β -hemolytic streptococci, *Proteus vulgaris*, *Enterobacter spp.*, *Streptococcus spp.*, *Proteus mirabilis*, etc. are extremely rarely observed, and in our study were isolated in one patient each (Table 1).

In physiologic conditions, different bacteria colonize body surface. A low count of pathogenic bacteria is not associated with an increase in the rate of inflammation, however, high colonization

may lead to inflammation. The presence of *Staphylococcus aureus* count of 10^5 - 10^6 /mm² has been described as sufficient to induce infection in standard conditions (10).

The initial stage of the infection proceeds free from any clinical signs of the disease, however, the bacteria keep penetrating the body where they multiply at a fast rate. Our patients showed no signs of wound infection or erysipelas, thus topical therapy was used in most patients. The subsequent course, i.e. development of infection, depends on the infectiosity, virulence and invasiveness of the agent, and on the general condition and immune status of the patient.

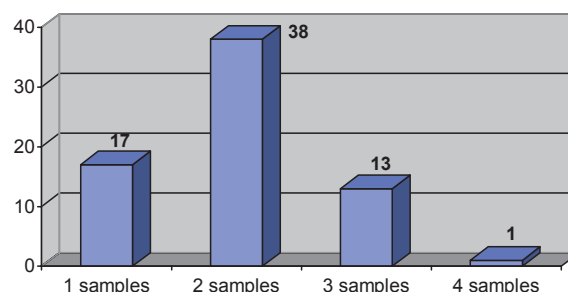


Figure 5. Pathogen distribution according to number of samples

Swab sampling for bacteriologic testing is not necessary in routine wound care. It is only recommended when one or more signs or symptoms indicative of wound infection are present, e.g., cellulitis, elevated temperature, pain, skin redness, lymphangitis, or ulcer enlargement. The diagnosis may be confounded by acute dermatitides such as contact dermatitis with erythema and wound enlargement.

The management of venous leg ulcers includes systemic antibiotic therapy according to antibiotic sensitivity report. Cellulitis, a common complication of venous leg ulcers, may be mistaken for noninfective erythema and edema that frequently surround the ulcer. Cellulitis is accompanied by the signs of tension or pain, lymphangitis, fever, nausea and leukocytosis (11,12).

CONCLUSION

Venous ulcers are chronic wounds that are obligatorily colonized by microorganisms. The presence of microorganisms need not imply an infection. In the absence of the clinical signs of infection, there is little evidence that wound healing is retarded by bacteria.

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