

Bacteria in Ulcera Crurum

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Bacterial cultures derived from 432 chronic leg ulcers were analysed retrospectively to determine which bacteria are most commonly found in these ulcers. The study covered a 2-year period. Two-thirds of the patients were over 70 years of age. *Staphylococcus aureus* was found in nearly half of the ulcers studied, *Pseudomonas* sp. in one-third, pyogenic streptococci and enterococci in every fifth and *Proteus* sp. in every tenth. The frequency by which pyogenic streptococci were isolated was about 10 to 20 times as high as previously reported. Obligate anaerobic bacteria were also frequently isolated. The sensitivity of the isolates from the second year to antimicrobial agents likely to be chosen if systemic therapy were required is also reported. The results are discussed in relation to previous findings. (Received October 26, 1987.)

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Ulcus cruris is a common, self-limiting but often chronic disease. The estimated incidence of ulcera crurum varies from 0.4 to 3% (1, 2). The bacterial flora recovered from the ulcers seems to vary (3-5) and infections such as cellulitis and erysipelas of the surrounding tissues and septicemia occasionally although rarely originate from the ulcers (6, 7). Bacterial cultures from the ulcers are frequently performed and may be useful in estimating the prevalence and sensitivity to antimicrobial agents of bacteria growing in ulcers treated in a given unit and in deciding which bacteria in the ulcers might be most likely to cause septic and other complications.

We therefore analysed in a retrospective study the distribution of bacteria growing from chronic leg ulcers and report here the findings of such an analysis and discuss them in relation to previous reports. The sensitivity of some of the isolated bacteria to certain antimicrobial agents is also reported.

MATERIALS AND METHODS

Bacterial cultures derived from chronic leg ulcers and sent for analysis by the City of Helsinki primary health care centres, hospitals and nursing homes during a period of 2 years were analysed. The clinical diagnosis was chronic leg ulcer in all the patients included in the study. The patients were mainly elderly women. Seventy-one percent were over 67 years of age, 94% over 50 years of age, while 2% were less than 50 years old. The age of 4% of the patients studied was not known. The total number of the samples analysed was 432. The samples were sent to the laboratory in Stuart transport tubes (Difco Laboratories, Detroit, USA, glass tubes). Aerobic and anaerobic cultures were performed and the bacteria identified as described by Lennette et al. (8).

Streptococcus pyogenes were grouped using Streptex reagents (Wellcome Diagnostics, UK). Alpha- and non-hemolytic streptococci were considered to be viridans group streptococci. Bile-resistant streptococci hydrolysing aesculin were considered to be enterococci and were not analysed further. Staphylococci possessing deoxyribonuclease were considered to be *S. aureus* and those lacking this enzyme, *S. epidermidis*. The differentiation was confirmed by using the slide agglutination test (Staphyslide, bioMérieux, France) when necessary. API 20E, APINE, API 20A and API20Strep (API System, France) were also used when necessary. All bacterial growth, including

members of the normal skin flora such as *Staphylococcus epidermidis*, diphtheroids, propionibacteria and *Bacillus* sp., was recorded. The disc diffusion method using discs from Rosco (Rosco Neosensitabs, Denmark), was used to test the antimicrobial sensitivity of some of the isolated bacteria.

RESULTS

Ninety-two percent of samples derived from chronic leg ulcers yielded microbial growth. The number of bacterial species isolated per ulcer varied from 1 to 7. Only one bacterial species was isolated from 28% of the ulcers, while 2 to 3 bacterial species per ulcer was the most frequent finding (Fig. 1). The average number of bacterial species per ulcer was 2.3.

The most frequently isolated bacterial species from the cultures of chronic leg ulcers was *Staphylococcus aureus*; it grew from 44% of the ulcers and comprised 20% of all the isolated bacteria (Table I). *S. aureus* was the only bacterial species isolated from 16% of the samples. The next most frequent isolate was *Pseudomonas* sp., which grew from 31% of the chronic leg ulcers and comprised 14% of the bacterial isolates. Staphylococci and *Pseudomonas* were followed by pyogenic streptococci and enterococci. *Streptococcus pyogenes* was isolated together with *S. aureus* in 11% of the samples. Pyogenic streptococci and enterococci were isolated from 20% of the ulcers and comprised 10% and 9% of the isolates, respectively. About 12% of the ulcers yielded obligate anaerobic rods, mainly of the family Bacteroides. About half of the Bacteroides belonged to the fragilis group. 8% of the ulcers yielded anaerobic cocci, peptostreptococci. *Candida* sp. was found in 3.5% of the samples.

The antimicrobial sensitivity of the *Staphylococcus aureus*, *Pseudomonas*, enterococcal and *Proteus* isolates from the second year of the study is given in Table II. All the isolated

Table I. Prevalence of certain bacteria in chronic leg ulcers

Isolate	n (%) ^a	% of ulcers ^b
<i>Staphylococcus aureus</i>	182 (20.2)	44
<i>Staphylococcus epidermidis</i>	94 (10.4)	23
<i>Streptococcus pyogenes</i> ^c	86 (9.5)	21
Enterococcus	80 (8.9)	20
<i>Pseudomonas</i> spp. ^d	127 (14.1)	31
<i>Proteus</i> spp. ^e	65 (7.2)	16
Other enterobacteriaceae ^f	110 (12.2)	24
Obligate anaerobic rods ^g	58 (6.4)	12
Obligate anaerobic cocci	36 (4.0)	8
Others ^h	65 (7.2)	14

^a n (%), number of isolates (% of all isolates).

^b % of ulcers, % of ulcers yielding the bacterium.

^c 32 (37%) of the pyogenic streptococci belonged to group A, 37 (43%) to group G, 13 (15%) to group B, 3 (3%) to group C and 1 to group F.

^d 93 (76%) of the pseudomonas isolates were *Ps. aeruginosa*, 10 (8%) *Ps. maltophilia*, 2 (2%) *Ps. fluorescens*, 17 (14%) were not identified to species level. Four *Flavobacterium* spp. and one *Alcaligenes faecalis* are included in this group.

^e 44 (69%) of the proteus isolates were *Pr. mirabilis*, 13 (20%) *Pr. vulgaris*, 6 (9%) were *Morganella morganii* and 1 (2%) *Providencia* sp.

^f Other enterobacteria included *E. coli*, *Enterobacter* sp., *Klebsiella* sp., *Citrobacter* sp., *Serratia* sp.

^g 32 (55%) of the anaerobic rods were of *Bacteroides fragilis* group, 24 (41%) of *Bacteroides nonfragilis* group and 2 (3%) were *Clostridium perfringens*.

^h Difteroids, viridans streptococci, propionibacteria, *Acinetobacter* sp., *Micrococcus* sp., *Moraxella* sp., *Bacillus* sp.

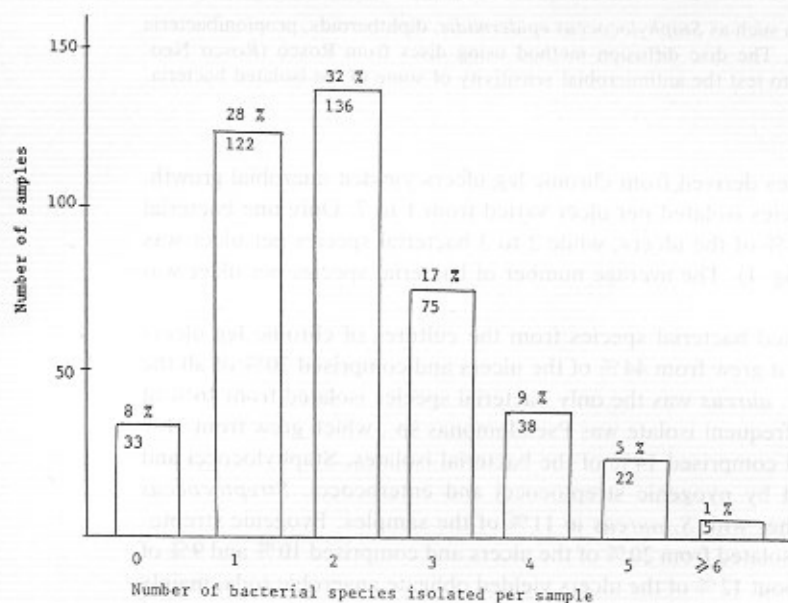


Fig. 1. Numbers and percentages of samples from venous leg ulcers yielding one to six or more bacterial species, or showing no growth.

Table II. Percentages of isolated *Staphylococcus aureus*, *Pseudomonas* sp., *Proteus* sp. and enterococci fully sensitive to commonly used antimicrobial agents

Bacteria	Antimicrobial agent	% sensitive (n/N) ^a	
<i>Staphylococcus aureus</i> ^b	Cloxacillin	100	(84/84)
	Cephalotin	100	(83/83)
	Tetracyclines	94	(79/84)
	Erythromycin	89	(75/84)
	Clindamycin	100	(84/84)
	Sulphatrimethoprim	96	(81/84)
<i>Pseudomonas</i> spp. ^c	Ceftazidime	89	(51/57)
	Gentamycin	91	(52/57)
	Tobramycin	93	(41/44)
	Amikacin	93	(37/40)
	Piperacillin	91	(40/44)
	Ciprofloxacin	84	(36/43)
	Aztrenonam	73	(27/37)
<i>Proteus</i> sp. ^c	Ampicillin	60	(18/30)
	Cefuroxime	67	(20/30)
	Tetracyclines	13	(4/30)
	Gentamycin	87	(26/30)
	Tobramycin	87	(26/30)
	Piperacillin	90	(27/30)
	Sulphatrimethoprim	73	(22/30)
Enterococcus	Ampicillin	100	(35/35)
	Tetracyclines	11	(4/35)
	Erythromycin	14	(5/35)
	Sulphatrimethoprim	89	(31/35)

^a n=number sensitive; N=number tested.

^b 76% of the isolated strains produced β -lactamase.

^c 77% of the *Pseudomonas* spp. and 90% of *Proteus* spp. were sensitive to neomycin.

Staphylococcus aureus strains were sensitive to cloxacillin, cephalothin and clindamycin and 90% or more to tetracyclines, erythromycin and sulphatrimethoprim. About 90% of the *Pseudomonas* species isolated were sensitive to piperacillin, ceftazidime and the aminoglycosides tested, gentamycin, tobramycin and amikacin. 84% were sensitive to ciprofloxacin and 73% to aztreonam. Piperacillin and aminoglycosides were effective against approx. 90% of the *Proteus* strains isolated, while 73% of the *Proteus* isolates were sensitive to sulphatrimethoprim and 60 and 67%, respectively, to ampicillin and cefuroxime. All enterococci isolated were sensitive to ampicillin and approx. 90% to sulphatrimethoprim.

DISCUSSION

Few of the chronic leg ulcers carry organisms which could be considered members of the normal skin flora. The pathogenic bacteria growing in the ulcers may, however, bear no correlation to the clinical status or healing of the wounds and may be largely unaffected by the local treatment(s) given (2). Although routine bacterial cultures of chronic leg ulcers are rarely indicated and should not be encouraged, occasional surveys can be useful. Thus knowledge of the bacteria growing in the ulcers treated in a given unit and their antimicrobial sensitivity is useful for epidemiological reasons and in predicting the most likely bacteria causing systemic or other infections derived from the ulcers and hence the treatment most likely to cover the infective agents.

The frequency by which certain bacteria are isolated from chronic leg ulcers may be influenced by e.g. the local conditions in the ulcer, the treatments given and the environmental microbial flora available at treatment units. Therefore the bacterial findings in different reports can vary (2-5). In patients studied in this report, *Staphylococcus aureus* was the bacterial species most frequently isolated from chronic leg ulcers. Nearly half of the ulcera crurum yielded *S. aureus*. In previous reports, *S. aureus* has been reported in 20-60% of the ulcers (2, 3). Every third chronic leg ulcer studied here yielded *Pseudomonas* sp., every fifth pyogenic streptococci and/or enterococci, every tenth *Proteus* sp. and/or obligate anaerobic rods. Pyogenic streptococci, mainly of groups G and A, were isolated from these patients about 10 to 20 times more often than described in previous reports (2, 3). Enterococci, found in 20% of the ulcers studied here, have previously been reported in 9-12% of ulcers (2, 3). The frequency at which *Pseudomonas* spp. were isolated was equal to or higher than in previous reports, 31% as opposed to 3-32% (2-5), while *Proteus* sp. was found in 16% of the ulcers vis-à-vis reported 1-6% (2-5). The numbers of anaerobes isolated from chronic leg ulcers has been reported to vary from less than 1% to 40% (2-5). A high frequency of anaerobes has been associated with diabetic foot ulcers (4, 5). Although the isolated bacteria generally were sensitive to the antimicrobial agents commonly used in systemic therapy, it is noteworthy that about 15% of the isolated *Pseudomonas* spp. were resistant to ciprofloxacin and 25% to aztreonam, although these two drugs were not in use (not registered in Finland) at the time of the study. The reasons for this are not known.

This retrospective study of chronic leg ulcers in elderly patients of whom two-thirds were more than 70 years of age revealed that in this group of patients, pyogenic streptococci were found in 20% of the ulcers, i.e., up to 20 times more frequently than previously reported. These ulcers could thus be an important source of septic and other complications such as erysipelas and sepsis caused by pyogenic streptococci. Antimicrobial therapy of e.g. sepsis should, if chronic leg ulcer is suspected as a primary focus, also cover pyogenic streptococci in addition to staphylococci and Gram-negative rods. Obligate anaerobes may also be involved in infectious complications of these ulcers, and should, at least in certain

patient groups, be taken into account in the therapy. A clinical follow-up of the patients was, however, not available and thus the frequency of streptococcal and other complications remains unknown.

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