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INVESTIGATIVE REPORT

Bacteriological Study of Epidermal Cysts

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The aim of this study was to determine whether bacterial infection plays a significant role in the inflammatory process of epidermal cysts. Samples from 152 patients (115 cases of inflamed and 37 of uninflamed epidermal cysts) were subjected to aerobic and anaerobic bacterial culture and the isolates were investigated. The rate of bacterial growth and the recovered anaerobes were significantly greater in the inflamed than the uninflamed epidermal cysts. However, it is difficult to determine whether recovered isolates from epidermal cysts represent "infection" or "colonization". In conclusion, this study revealed the predominance of anaerobes in inflamed cysts, strongly suggesting that anaerobes play a role in the inflammatory process. ***Key words: epidermal cysts; bacterial culture.***

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Epidermal cysts are closed sacs with a definite wall that results from proliferation of surface epidermal cells. Production of keratin and lack of communication with the surface are responsible for cyst formation. Epidermal cysts can become infected, and when an abscess develops they may need to be surgically drained, followed by administration of systemic antibiotics. It is a matter of debate whether bacterial infection plays a role in the inflammatory process. Some authors reported that 2 groups of inflamed and uninflamed epidermal cysts did not differ significantly with respect to the number of bacterial isolates and aerobes, anaerobes or potential pathogens cultured (1, 2). In other reports, *Staphylococcus aureus* (3, 4) and indigenous anaerobes (4, 5) were predominantly isolated from inflamed epidermal cysts, suggesting that they may have played a role in the inflammatory process (3–5).

In this study, we obtained aerobic and anaerobic bacterial culture specimens from inflamed and uninflamed epidermal cysts. We then investigated the positive rate of culture growth and species of recovered isolates in order to determine whether bacterial infection plays a significant role in the inflammatory process of epidermal cysts.

SUBJECTS AND METHODS

Subjects

The subjects were 152 patients clinically diagnosed with epidermal cysts at Osaka City General Hospital and Osaka City Juso Hospital between January 1998 and December 2005. They were non-immunocompromised adult patients with no history of recent systemic antibiotics. There were 115 patients with inflamed epidermal cysts (72 men and 43 women) aged 18–79 years (mean age 43.3 years) and 37 patients with uninflamed epidermal cysts (20 men and 17 women) aged 24–83 years (mean age 51.1 years).

Methods

An inflamed epidermal cyst was defined as a known cyst that developed a fluctuant soft-tissue swelling surrounded by erythema and contained a localized collection of purulent material. An uninflamed cyst was defined as an intradermal nodule with no evidence of inflammation. Each cyst was incised with a sterile surgical blade and then a sterile swab was inserted into it. After written informed consent was obtained, samples were collected by the swab method, before treatment with antibiotics. In the aerobic culture, samples were cultured on sheep blood agar plates and chocolate agar plates in 5.5% CO_2 at 35ºC for 3 days. Colonies formed were identified based on colony morphology, Gram stain, and catalase and coagulase productivity. In the anaerobic culture, samples were immediately streaked on 7% sheep blood agar prepared with Brucella (Kyokuto Pharmaceutical Co., Tokyo, Japan) and incubated at 35ºC for 48 hours in an anaerobic jar with an atmosphere of 5% CO_2 + 95% N_2.The organisms grown anaerobically under such conditions were then identified using Rapid ANA II System (for biochemical identification of medically important anaerobic bacteria, RE-MEL Inc., Norcross, GA, USA) and Rapid ANA II Code Compendium (6). Institutional ethical review boards at the Osaka City General Hospital and Osaka City Juso Hospital approved the study protocol.

RESULTS

Of the 115 inflamed cysts, 45 were from the head and neck, 38 from the trunk and 27 from the buttocks and inguinal area, whereas of the 37 uninflamed cysts, 16 were from the head and neck, 15 from the trunk, 3 from the gluteal and inguinal regions and 3 from the extremities (Table I).

Of the 115 inflamed cysts, 91 (79.1 %) yielded bacterial growth (a total of 116 isolates). Of the 37 uninflamed cysts, 20 (54.1%) cysts yielded bacterial growth (a total of 21 isolates), showing that isolates were significantly more frequently recovered from inflamed cysts than uninflamed cysts (χ^2 test, p=0.003).

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Table I. *Location and numbers of isolates from inflamed and uninflamed epidermal cysts*

	No of cysts (% of total number of cultures)	
	Inflamed	Uninflamed
Location		
Face, neck, or scalp	45 (39)	16 (43)
Trunk	38 (33)	15 (41)
Buttocks, inguinal area	27 (24)	3 (8)
Extremities	5 (4)	3 (8)
Numbers of isolates		
Positive cultured growth	91 (79)	20 (54)
Aerobes only	56 (49)	18 (48)
Anaerobes only	24 (21)	1 (3)
Aerobes and anaerobes	11 (9)	1 (3)
Negative cultured growth	24 (21)	17 (46)

Aerobic bacteria were recovered from 67 of the 115 inflamed epidermal cysts and 19 of the 37 uninflamed cysts, with no significant difference (χ^2 test, p=0.461). The predominant aerobes from the inflamed cysts, were coagulase-negative staphylococci (CNS) (43 isolates, including 18 *S. lugdunensis*, 6 *S. epidermidis* and 19 other CNS), followed by *Corynebacterium* spp. (11 isolates) and *S. aureus* (9 isolates). From the uninflamed cysts, 13 CNS isolates (2 *S. lugdunensis*, 5 *S. epidermidis* and 6 other CNS) and 4 *Corynebacterium* spp. were frequently isolated (Table II). *S. aureus* was more frequently isolated from the inflamed cysts than uninflamed cysts, but no significant difference was found (Fisher's test, p=0.114)

Anaerobes were isolated from 35 (30.4%) of the 115 inflamed epidermal cysts and 2 (5.4%) of the 37 uninflamed epidermal cysts, showing that anaerobes were significantly more frequently isolated from the inflamed cysts (χ^2 test, p=0.002). *Peptostreptococcus spp.* was the most frequently isolated from the inflamed cysts (26 isolates: 13 *P. magnus* and 9 *P. asaccharolyticus*), followed by *Propionibacterium* spp. (5 isolates), *Bacteroides* spp. (5 isolates) and *Prevotella* spp. (3 isolates). In contrast, only 2 isolates of *P. magnus* were isolated from the uninflamed cysts (Table II).

The distribution of the aerobic and anaerobic isolates, including CNS, *Corynebacterium* spp., *S. aureus*, *Peptostreptococcus* spp. and *Propionibacterium* spp. was equal in all the anatomic sites (Table III). However, 3 of 5 isolates of *Bacteroides* spp. were isolated from the gluteal region.

Table II. *Micro-organisms cultured from epidermal cysts*

	No. of epidermal cysts (% of total number of cultures)	
Micro-organism	Inflamed	Uninflamed
Aerobes		
CNS		
lugdunensis	18	2
epidermidis	6	5
Other	19	6
Staphylococcus aureus	9	0
Corynebacterum spp.	11	4
Streptococcus spp.	4	0
Proteus mirabilis	2	1
Escherichia coli	1	0
Klebsiella pneumoniae	1	0
Micrococcus spp.	1	1
Morganella morganii	1	0
Serratia proteamaculans	1	0
Enterococcus spp.	1	0
Anaerobes		
Peptostreptococcus		
magnus	13	2
asaccharolyticus	9	0
Other	4	0
Propionibacterium		
acnes	1	0
Other	4	0
Bacteroides		
fragilis	2	0
Other	3	0
Prevotella spp.	3	0
Porphyromonas asaccharolyticus	1	0
Peptococcus spp.	1	0

CNS: Coagulase-negative staphylococcus

DISCUSSION

Epidermal cysts occasionally become inflamed, with an erythematous soft tissue swelling containing a purulent material. In surgical drainage, a purulent cheesy discharge is excreted. This inflammatory process may not be a simple bacterial skin infection; however, micro-organisms may have some influence on the inflammation mechanism of epidermal cysts. We have studied the bacterial isolates from inflamed and uninflamed epidermal cysts using techniques to recover anaerobic and aerobic bacteria. The number of recovered isolates and positive rate of bacterial growth in the inflamed

Table III. *Characterization of inflamed and uninflamed epidermal cysts according to body site*

	Face, neck, scalp	Trunk	Buttocks, inguinal area	Extremities
Inflamed (n = 115)				
Aerobes				
CNS	12	15	14	2
Staphylococcus aureus	5	2	2	0
Corynebacterium spp.	2	5	4	0
Others	6	3	1	2
Anaerobes				
Peptostreptococcus spp.	9	12	5	0
Propionibacterium spp.	2	3	0	0
Bacteroides spp.	1	1	3	0
Others	4	0	1	0
Uninflamed (n = 37)				
Aerobes				
CNS	6	5	1	1
Corynebacterium spp.	0	4	0	0
Others	2	0	0	0
Anaerobes				
Peptostreptococcus spp.	2	0	0	0

CNS: Coagulase-negative staphylococcus

group were significantly more frequent than in the uninflamed group, and the anaerobes were significantly more frequently recovered from inflamed cysts.

Diven et al. (1) investigated 25 inflamed and 25 uninflamed epidermal cysts, and Ohata et al. (2) investigated 40 inflamed and 32 uninflamed epidermal cysts, and they both concluded similarly that the dominant recovered isolates from both groups were CNS, *Peptostreptococcus* spp. and *Propionibacterium* spp., while the 2 groups did not differ significantly with respect to number of isolates, "no growth" cultures, and aerobic or anaerobic organisms cultured. They were unable to discern any pattern to the types of organisms or the quantities cultured from inflamed and uninflamed cysts (1, 2). In contrast, others reported that *S. aureus* (3, 4) and some anaerobes, such as *Prevotella*, *Peptostreptococcus* spp. and *Bacteroides* spp. (3–5), were more frequently isolated from inflamed cysts. Brook (3) investigated 231 infectious epidermal cysts, and isolated 153 isolates of aerobes (81 isolates of *S. aureus* and 7 isolates of CNS) and 162 isolates of anaerobes (55 isolates of *Bacteroides* spp., 85 isolates of *Peptostreptococcus* spp., and 7 isolates of *Propionibacterium* spp.), suggesting the involvement of *S. aureus* in inflammation (3, 4). Nishijima et al. (5) investigated 53 and 32 inflamed and uninflamed epidermal cysts, respectively, and found significant isolation of *Propionibacterium*, *Prevotella*, *Veillonella* and *Bacteroides* from the inflamed epidermal cysts (5).

Anaerobes were isolated significantly frequent from the inflamed, rather than from the uninflamed, epidermal cysts. *Peptostreptococcus* spp. accounted for 64% of anaerobic isolates, and was similarly predominant in other reports (57% (5), 56% (2) and 52% (3)). In our study, *P. magnus* and *P. asaccharolyticus* accounted for 33% and 21% of anaerobic isolates, respectively. Brook & Frazier (7) reported that 133 *P. magnus* and 84 *P. asaccharolyticus* isolates were found among 1260 isolates recovered from bacterial skin infections, suggesting that these Gram-positive cocci are important in the induction of skin soft tissue infection and inflammation of bones, joints and soft tissues (3, 8, 9). Similar to CNS, a cell wall component of gram-positive cocci, peptidoglycan, may act as a superantigen and induce the production of inflamed cytokines, including interleukin (IL)-1β and tumour necrosis factor (TNF)-α, leading to inflammation.

Other than *Peptostreptococcus* spp., a significant isolation of *Bacteroides* spp. (*B. melaninogenicus* and *B. fragilis*) from inflamed epidermal cysts was reported by Brook (3), and *Propionibacterium*, *Prevotella*, *Veillonella* and *Bacteroides* by Nishijima et al. (5). Higaki et al. (10) reported the predominance of *Propionibacterium* and *Prevottella* in an investigation of numerous bacterial skin infections (10). Lipopolysaccharides contained in the outer cell membrane of gram-negative bacilli, such as *Bacteroides*, *Fusobacterium* and *Veillonella*, have endotoxin-like properties, and exhibit pathogenic features (11). *P. acnes* releases lipase, which may produce free fatty acids, and protease, which may destroy hair follicle walls, thus contributing to the process of inflammation (12).

This study highlights the predominance of anaerobes in inflamed epidermal cysts, strongly suggesting that anaerobic bacterial infection may play a significant role in the inflammatory process (5). Questions regarding pathogenesis and appropriate treatment, including the proper selection of antimicrobial therapy, should be addressed in additional studies.

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