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The role of nasal cavity disinfection in the bacteriology of chronic sinusitis*

Ph. Rombaux[1], S. Collet[2], M. Hamoir[1], Ph. Eloy[2], B. Bertrand[2], F. Jamart[3], J. Gigi[4]

[1] Department of Otorhinolaryngology, Cliniques Universitaires Saint Luc, Brussels, Belgium
[2] Department of Otorhinolaryngology, Cliniques Universitaires Mont Godinne, Brussels, Belgium
[3] Department of Biostatistics, Cliniques Universitaires Mont Godinne, Brussels, Belgium
[4] Microbiology Department, Cliniques Universitaires Saint Luc, Brussels, Belgium

SUMMARY

Objectives: *Several factors may influence the results of bacteriological studies in chronic rhinosinusitis. We investigated the potential role of nasal cavity disinfection in the bacteriology of the bulla ethmoidalis in patients suffering from chronic sinusitis.*

Material and Methods: *Bacteriology of the bulla ethmoidalis was studied in 176 consecutive adult patients presenting a chronic sinusitis refractory to standard medical treatment who underwent functional endoscopic sinus surgery. Two different techniques were used: (A) a technique with nasal vestibule and facial disinfection with chlorhexedin (N = 89 patients and 165 samples) vs. (B) a technique with facial, nasal vestibule and nasal cavity disinfection with a povidone-iodine solution followed by a cleansing of the nasal cavity (N = 87 patients and 166 samples).*

Results: *Culture rate was 89.6% (183 bacterial isolates) for technique (A) vs 76.5% (164 bacterial isolates) for technique (B) ($p < 0.001$). Major bacteria encountered in the (A) group and in the (B) group were respectively: Coagulase Negative Staphylococcus: 77 vs 40 isolates ($p < 0.001$); Coagulase positive Staphylococcus: 44 vs 30 isolates ($p = 0.061$); Streptococcus pneumoniae: 4 vs 5 isolates; Others: Streptococcus sp.: 12 vs 16 isolates; Haemophilus influenzae: 8 vs 6 isolates; Enterobacteriacea: 33 vs 53 isolates ($p = 0.013$) and others Gram Negative Bacilli: 3 vs 7 isolates.*

Conclusions: *The standard (A) technique to study the bacteriology of the bulla ethmoidalis in patients with chronic sinusitis yielded a higher percentage of positive culture and of bacterial isolates than a more advanced (B) technique. This is mainly due to the higher percentage of contaminant bacteria such as Coagulase Negative Staphylococcus recovered with the standard technique. Enterobacteriacea and others Gram Negative Bacilli were more often encountered into the bulla ethmoidalis with the technique where disinfection of the nasal cavity was performed.*

Key words: bacteriology, bulla ethmoidalis, chronic rhinosinusitis, swab sampling.

INTRODUCTION

Chronic rhinosinusitis is usually caused by impaired paranasal sinus ventilation and drainage disorders located in the ostiomeatal complex. At least partially, chronic rhinosinusitis is a microbiological disease in which virus, bacteria and fungi play a distinct role [1]. Antibiotic therapy has been advocated for the treatment of acute bacterial rhinosinusitis (ABRS) but its role in chronic rhinosinusitis remains doubtful [2]. However, empiric antibiotic therapy covering bacteria frequently encountered in chronic infected sinuses remains an accepted method of treatment for chronic rhinosinusitis [3]. Bacteriological studies in chronic rhinosinusitis recover different results depending on many factors such as: way of sampling, site of sampling, culture medium and characteristics of the patient (previous surgery or not, previous antibiotherapy or not) (Table 1). For all these reasons, the study of the bacteriology of chronic rhinosinusitis did not yet achieve a general consensus. Maxillary sinus antral puncture seems to be the method of choice to study the bacteriology of maxillary acute sinusitis and proper techniques of sampling into the middle meatus seem to correlate with maxillary antral culture [4]. For chronic rhinosinusitis, endoscopically guided sinus cultures into the ethmoid sinus become the method of choice when considering the bacteriology of this disease [5]. However, sampling techniques may be quite different using the swab technique or the suction aspirate technique. Moreover, transporta-

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Table 1. Controversies surrounding studies performed to study the bacteriology of rhinosinusitis.

Sampling methods (swab - aspirate)
Patient : age, acute vs chronic, immunocompetent, previous therapy, previous surgery,…
Site of culture
Transport method and media
Culture technique
Processing in the lab
Culture results expression
Susceptibility testing

tion, processing in the lab, culture results, expression and susceptibility testing may also lead to difficulties in interpreting the results [6]. We investigated the potential role of nasal cavity disinfection in the bacteriology of the bulla etmoidalis in patients suffering from chronic rhinosinusitis.

The aim of this work was to compare two different methods of nasal disinfection before sampling and to underline the differences between contaminant and pathogenic bacteria obtained by the two methods.

MATERIALS AND METHODS

A prospective study was conducted at the Saint Luc university clinic between January 2000 and December 2002. Consecutive adult patients with chronic sinusitis refractory to standard medical treatment underwent functional endoscopic sinus surgery under general anesthesia.

In this study, antibioprophylaxy was given at the end of the surgical procedure, therefore not influencing sampling.

The diagnosis of chronic rhinosinusitis was based on clinical, radiographic and endoscopic findings. Patients were excluded if they showed factors that may influenced the microbiology of the sinuses such as: atrophic rhinitis, immunodepression, history of previous sinus surgery, odontogenic disease, nasal topical treatment and/or antibiotherapy within 10 days before surgery, cystic fibrosis, patients in intensive care or with a nasogastric tube and extramucosal mycotic sinusitis. Two different methods of disinfection were performed in this study. The choice between the different methods was randomly made.

Technique of sampling

In the first (A) technique, the face and the nasal vestibule of the patients were disinfected with a chlorehexedin solution. Vasoconstrictive agents on cottonoid pledgets were left in place for 10 minutes and then removed before surgery. Then a vertical incision of the uncinate process in order to visualize the bulla ethmoidalis was performed. Bulla ethmoidalis was opened using sterile-cutting forceps. A sterile cotton-tipped swab was introduced into the nasal cavity, while retracting the nasal ala with the endoscope, and then placed into the bulla ethmoidalis.

In the second (B) technique, the face and the nasal vestibule of the patients were disinfected with a povidone-iodine solution. Cottonoid pledgets with vasoconstrictive agents were placed into the nasal cavity for 10 minutes and then removed. After vasoconstriction of the nasal mucosa, the nasal cavity was disinfected with 20 ml of povidone-iodine solution and then cleansed with 20 ml of physiologic serum. After the disinfection, the swab was placed into the bulla ethmoidalis using the same technique and the same precautions as in the (A) technique.

In the (A) and (B) techniques, the swab samples medium for transfer were respectively a Stuart's medium and a thioglycolate medium.

On arrival at the laboratory, the samples were incubated for 24 hours under aerobic conditions. If a growth developed after 24 hours, a subculture was made on: Columbia blood agar (Becton – Dickinson) in a capnophilic atmosphere at 35°C; Mueller Hinton blood with Bacitracine and V factors (Becton – Dickinson) in a capnophilic atmosphere at 35°C; - Wilkins amukin blood (Becton – Dickinson) in an anaerobic atmosphere at 35°C.

Identification of bacterial isolates was based on conventional biochemical, enzymatical and phenotypical biotyping.

The use of rabbit plasma revealed the presence of free and bound coagulase and served to distinguish *coagulase negative Staphylococcus* (CoNS) and *coagulase positive Staphylococcus* (CPS) or *Staphylococcus aureus.*

To investigate Methicillin-resistant Staphylococcus aureus (MRSA), an Oxascreen medium was incorporated into the study as well as an E-test System (AB Biodisk) on Mueller Hinton blood (Blood – Merieux) to specify the level of Streptococcus pneumoniae to Penicillin.

Total number of samples was reported for each group of patients. Culture rate was the number of positive cultures divided by the total number of samples. Bacterial isolates was the total number of bacteria found in the positive culture. Statistic analysis was performed by a Chi-Squared test.

RESULTS

During this period, a total of 176 patients underwent functional endoscopic sinus surgery for refractory chronic ethmoidal sinusitis. There were 89 patients (165 samples) who have been studied with the (A) technique and 87 patients (166 samples) with the (B) technique.

The patient population in each group was similar in terms of gender, age and comorbid conditions. The (A) and (B) methods of sampling give respectively as positive culture: 89.6 % (148/165 samples) vs. 76.5 % (127/166 samples) ($p < 0.001$). Total number of bacterial isolates was 183 microorganisms with the (A) method and 164 microorganisms with the (B) method ($p < 0.001$). When considering the total number of positive cultures, it was possible to specify the culture as mono- or polymicrobial: (A) method: 98 positive cultures with one microrganism, 32 with two microorganisms and 7 with three microorganisms;

(B) method: 91 positive cultures with one microorganism, 29

Table 2. (A) Method with facial and nasal vestibule disinfection with chlorhexedin. (B) Method with facial, nasal vestibule and nasal cavity disinfection with Povidone-iodine and nasal cavity cleansing before sampling.

Bacteriology	(B) n = 166	(A) n = 165	Statistical significance
Aerobes	162	183	
Gram +	96	139	$p < 0.001$
Staphylococcus Coag. Negative	40	77	$p < 0.001$
Staphylococcus aureus	30	44	$p = 0.061$
Streptococcus Group C,G	0	4	
Streptococcus pneumoniae	5	4	
Streptococcus milleri	1	3	
Streptococcus viridans	15	5	
Enterobacter faecalis	1	0	
Corynebacterium pseudodiphteria	2	2	
Bacillus cereus	2	0	
Gram -	66	44	$P = 0.011$
Haemophilus influenzae	6	8	
Stenotromonas maltophilia	4	0	
Acinetobacter baumanii	2	0	
Acinetobacter junii	1	0	
Pseudomonas aeroginusa	0	3	
Enterobacteriacea ;	53	33	$P = 0.013$
E . Coli	14	7	
Enterobacter aerogenes	4	0	
Enterobacter cloacae	4	4	
Klebsellia pneumoniae	5	2	
Klebsellia oxytoca	6	4	
Serratia marcescens	1	5	
Citrobacter diversus	3	0	
Citrobacter freundii	3	4	
Proteus vulgaris	2	1	
Proteus mirabilis	11	6	
Anaerobes	2	0	

with two microorganisms and 5 with three microorganisms. Gram positive bacteria recovered 139 strains with the (A) method vs. 96 strains with the (B) method ($p < 0.001$), *Staphylococcus aureus*: 44 vs 30 ($p = 0.061$); *Coagulase negative Staphylococcus*: 77 vs. 40 ($p < 0.001$). Gram negative bacteria recovered 44 strains with the (A) method vs 66 strains with the (B) method ($p = 0.011$) and Enterobacteriacea: 33 vs 53 ($p = 0.013$).

Anaerobes could only be demonstrated in two samples.

Table 2 summarizes the differing results obtained using the two different methods of sampling with the statistical significance for the mostly encountered bacteria.

Table 3 gives the percentage of recovery expressed with different denominators such as the total number of positive cultures, the total number of bacterial isolates and the total number of cultures performed for the *Enterobacteriacea* group, the *Staphylococcus aureus* and the *Coagulase negative Staphylococcus.*

None of the *Staphylococcus* species have shown a decreased susceptibility to Methicillin, neither *Streptococcus pneumoniae* to Penicillin.

DISCUSSION

Although the standard method of sampling and disinfection yielded a higher percentage of positive cultures than a more advanced one, it seems that this fact is related to a higher percentage of bacteria usually seen as contaminant. Pathogenic bacteria such as *Enterobacteriacea* were mostly recovered with a proper method of sampling and with proper disinfection of the nasal cavity.

Many factors may influence the different results in the recovery rates between our two methods: nasal fossa disinfection, nasal vestibule disinfection, or the medium of culture. Simultaneously modifying the disinfection technique and the culture medium does not allow for adequate analysis of any individuals variable. However, this current study helps to underline the importance of nasal vestibule and of nasal fossa disinfection.

In order to avoid contamination of the swab when performing studies on bacteriology in the bulla ethmoidalis, it has been advocated to respect some rules: proper disinfection of the nasal vestibule and of the nasal fossa, endoscopic endonasal control and retracting the nasal ala with the endoscope when entering the swab into the nasal fossa. A protected device for the swab is an interesting tool which needs further investigation. A recent study with this device from Tantilipikorn et

Table 3. (A) Method with facial and nasal vestibule disinfection with chlorhexedin. (B) Method with facial, nasal vestibule and nasal cavity disinfection with Povidone-iodine and nasal cavity cleansing before sampling

Bacterial isolates (%)	Total number of positive culture n = 127 (B)	Total number of positive culture n = 148 (A)	Total number of bacterial isolates n = 164 (B)	Total number of bacterial isolates n = 183 (A)	Total number of culture n = 166 (B)	Total number of culture n = 165 (A)
Enterobacteriacea	41.7%	22.3%	32.3%	18.0%	31.9%	20.0%
Staphylococcus Coagulase negative	31.5%	52.0%	24.4%	42.0%	24.1%	46.7%
Staphylococcus aureus	23.6%	29.7%	18.3%	24%	18.1%	26.7%.

al.[7] has suggested that endoscopically-guided aspiration of pathological secretions is not better than properly-obtained swabs in directing antimicrobial therapy for chronic rhinosinusitis (swab at the sinus ostium or in the sinus cavity of patients who have had previous sinus surgery).

Although no general consensus has yet been proposed, it seems that endoscopic endonasal guided culture of the bulla ethmoidalis is an appropriate method when considering the bacteriology of chronic ethmoidal sinusitis.

Paranasal sinuses are dogmatically considered as sterile though endoscopic guided cultures in healthy subjects into the middle meatus or in the spheno-ethmoidal recess have revealed the presence of some microorganisms: *Coagulase negative Staphylococcus, Corynebacterium species* and *Propionibacterium species* [8]. On the other hand, the nasal fossa, nasal vestibule and nasopharynx are usually colonized by a variety of microorganisms, which may become pathogenic when an upper airway viral infection is present for instance.

All these facts underline the importance of comparing the microbiologic results obtained in pathological conditions to those obtained in healthy subjects. Unfortunately, there is no study on the bacteriology found in the bulla ethmoidalis from healthy subjects for ethical reasons.

Bacteriology of chronic rhinosinusitis is rather understood as a polymicrobial disease where aerobic and anaerobic bacteria may be encountered. Besides microorganisms commonly found in the acute phase such as *Streptococcus pneumoniae, Haemophilus influenzae* and *Moraxella catharralis*, the bacteriology of chronic rhinosinusitis is more complicated, with most authors reporting *Coagulase negative Staphylococcus, Staphylococcus aureus, Enterobacteriacea* and anaerobic species. There is also a tremendous debate surrounding the role of fungi in the pathophysiology of the disease. Our present study focuses on the high percentage rate of *Enterobacteriacea* recovered with the proper (B) technique of sampling. This was also demonstrated by some studies performed in children and/or adults [5,9]. The role of *Enterobacteriacea* in chronic rhinosinusitis needs to be further investigated as its role in lower respiratory disease has already been highlighted [10].

The low percentages of anaerobes found in this study may be explained by our exclusion criteria (no odontogenic sinusitis) as well, at least for the thyoglycolate medium, by the swab samples medium. Thyoglycolate medium allows both aerobic and anaerobic growths and competitive growth may be present explaining the fact that aerobic bacteria are growing more rapidly than anaerobic bacteria and thus influence the percentage of anaerobic bacteria encountered. It should also be pointed out that the percentages of anaerobes found in chronic sinusitis may extremely differ from one study to an other [1].

One of the major goals of bacteriological studies is to give some support to empiric antibiotic therapy. Ideally, this antimicrobial agent selection should be based on endoscopic guided culture and a sensitivity study of the microorganisms. Nevertheless, empiric antibiotic therapy remains an accepted method. Gram negative bacteria, especially *Enterobacteriacea*, need to be targeted when considering the antibiotic therapy even though the disease is not viewed as a primarily bacteriologic disease [11].

One could argue that this study did not take into account the bacteriologic criteria consistent with sinus infection: purulent fluid on macroscopic examination, presence of neutrophils and bacteria identifiable by Gram staining or positive culture showing one or more predominant organism at the usual cut-off choice of 104 CFU/ml [2]. Moreover, chronic rhinosinusitis is currently viewed as a disease with bacterial colonisation followed by a host reaction to these colonizing bacteria [1]. Therefore, bacterial colonisation or bacterial infection must be distinguished. The bacterial colonisation of chronic ethmoidal sinusitis yielded different results depending on different disinfection techniques. The method with chlorehexedin disinfection on the face and in the nasal vestibule produced a higher percentage of positive cultures. This fact is related to a higher rate of contaminant bacterial isolates. The method where disinfection was performed using a povidone-iodine solution on the face, in the nasal vestibule and in the nasal cavity followed by a cleansing of the nasal cavity underlines the importance of colonisation with Enterobacteriacea in the bulla ethmoidalis of patients suffering from chronic rhinosinusitis.

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Ph. Rombaux, MD
Department of Otorhinolaryngology
Université Catholique de LOUVAIN
Hippocrate Avenue, 10
1200 Brussels
Belgium

Tel : 0032-2-764-1930
Fax : 0032-2-764-8935
E-mail : philippe.rombaux@orlo.ucl.ac.be