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Dentigenous infectious foci – a risk factor of infective endocarditis

Authors' Contribution:

- A** Study Design
- B** Data Collection
- C** Statistical Analysis
- D** Data Interpretation
- E** Manuscript Preparation
- F** Literature Search
- G** Funds Collection

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Summary

Background:

Dentigenous, infectious foci are frequently associated with the development of various diseases. The role of such foci in the evolution of endocarditis still remains unclear. This article presents the concluding results of an interdisciplinary study verifying the influence of dentigenous, infectious foci on the development of infective endocarditis.

Material/Methods:

The study subjects were 60 adult patients with history of infective endocarditis and coexistent acquired heart disease, along with the presence at least 2 odontogenic infectious foci (ie, 2 or more teeth with gangrenous pulp and periodontitis). The group had earlier been qualified for the procedure of heart valve replacement. Swabs of removed heart valve tissue with inflammatory lesions and blood were then examined microbiologically. Swabs of root canals and their periapical areas, of periodontal pockets, and of heart valves were also collected.

Results:

Microbial flora, cultured from intradental foci, blood and heart valves, fully corresponded in 14 patients. This was accompanied in almost all cases by more advanced periodontitis (2nd degree, Scandinavian classification), irrespective of the bacterial co-occurrence mentioned. In the remaining patients, such consistency was not found.

Conclusions:

Among various dentigenous, infectious foci, the intradental foci appear to constitute a risk factor for infective endocarditis.

key words:

dental infectious foci • endocarditis • periodontitis

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BACKGROUND

The existence of dentigenous infectious foci constitutes a potential danger to the human body, as they may become a source of severe systemic diseases involving the heart, joints, kidneys and other organs. This view is supported by a number of studies [1–11].

Infective endocarditis (IE) is an intravascular-type infection, affecting heart valves and other artificial devices such as pacemaker electrodes and prostheses. It is in most cases caused by bacteria, and is characterised by elevated morbidity and mortality rates, with the latter reaching 30% [12,13]. The incidence is 3 per 100,000 inhabitants annually [14].

Typical entry points are oral cavity and teeth 26%, gastrointestinal tract 12.5%, drug abuse 5%, skin 5%, urinary tract 4%, iatrogenic 9%, other 5.5%, and undetermined 33% [14]. The oral cavity is the most common point of entry, particularly in persons over 50 years old. The aortal valve is usually the location for the bacteria involved, again in patients over 50 years of age. The bacteria locate in 63.5% of cases on the valves, with concomitant defects. The remaining 36.5% locate on normal valves, in persons otherwise healthy. Methicillin-resistant *Staphylococcus aureus* species, diverse streptococcal strains (particularly *S. viridans*) and *Enterococcus faecalis* are the most frequent infective pathogens associated with native-valve endocarditis, and also in the late form of this disease following valve replacement [14,15]. In turn, the most frequent bacterial species present in gangrenous pulp is *Streptococcus viridans*. Among other species occurring there, *Staphylococcus aureus* has also been reported [16].

Therefore, the coincidence of seemingly the same (corresponding, consistent) bacterial flora typical for pulp gangrene and that for infected endocardium appears, in our opinion, not to be accidental. No clear evidence on this has, to date, been published.

This article thus attempts to fill this gap. Identification of infective foci, by types, which are potentially responsible for the development of infective endocarditis, was the study objective.

MATERIAL AND METHODS

The studied group consisted of 60 patients with infective endocarditis, concomitant acquired heart disease, and the occurrence of at least 2 dentigenous, infectious foci.

The group had earlier been qualified for heart valve replacement procedure, and all patients included in the study had the cardio-surgery.

The diagnoses of infective endocarditis, dental examination, and dental treatment were separated from cardiac surgery by up to 3 weeks. Full clinical examination was the basis for qualification in the study.

The study inclusion criteria were occurrence of active infective endocarditis, acquired heart disease, and the presence of 2 or more odontogenic, infectious foci. The existence of other infectious foci was excluded during the

examination by a clinical specialist. If other than dentigenous, infectious foci occurred, then, despite the presence of IE and an acquired heart disease, such patients were excluded from the study.

The study exclusion criteria were absence of any 1 of the 3 or more pathologies forming the required inclusion criteria, or, along with the presence of these inclusion criteria, a presence of 1 or more infectious foci at a location other than a denture. As mentioned above, the presence of any of such additional infectious foci had been excluded prior to forming the study group, in the course of medical examination by a specialist.

The following laboratory tests were performed in all patients, confirming the occurrence of endocarditis:

- blood cultures,
- cultures from heart valves, after their surgical removal,
- cultures from odontogenic, infectious foci,
- echocardiography, electrocardiography, and chest x-ray,
- electrophoresis of the serum proteins, the A.S.O. test, rheumatoid factor, seromuroid.

An infectious (also dentigenous) focus was defined as a lesion characterised by the presence of pathogens and/or capable of producing infection.

Dentigenous, infectious foci included:

- teeth with dead (devitalised) pulp or with its necrotic decay,
- resected teeth, with poorly filled root canals (due to anatomical or other difficulties),
- teeth without evident periapical lesions, and with seemingly well filled root canal (eg, endo-perio syndrome),
- residual roots,
- root cysts,
- periodontal pockets,
- inflammatory processes due to maleruption,
- periodontal changes leading to osteitis (eg, alveolar).

The above-mentioned foci were divided into either intradental foci, periapical foci or periodontal foci.

Intradental foci (1st order) are defined by their localisation in a tooth's pulp chamber and pulp canal. Periapical foci (2nd order) are localised near the tooth's apex.

Periodontal foci are divided into 3 degrees: 1st degree: *levis* = mild, 2nd degree: *gravis* = severe, and 3rd degree: *complicata* = complicated (Table 1).

Periodontal pockets deeper than 3 mm were regarded as infectious foci. Such pockets are also referred to as pathological pockets.

Detailed clinical and laboratory examinations were performed in order to ascertain the existence of active (infectious) foci in the oral cavity:

- thorough history taking was conducted with a view to indicating possible association between an exacerbation in the alveolar process area and the symptoms of a focus-related disease,
- careful examination of the oral cavity was performed, with particular attention paid to:

- pulpless teeth (pulp vitality assessment, percussion, visual assessment of the periapical area: colour changes, undue eminence, fistula openings, palpation, Owinski symptom),
 - periodontal pockets: their depth and type of exudate,
 - clinical assessment of the periodontium; its inflammatory lesions were graded according to the Scandinavian classification,
- c. current condition and the hygiene of removable and fixed dentures,
- d. palpation of submental and submandibular lymph nodes,
- e. x-ray pantomographic examination in all subjects, and when necessary, additionally fluoro radiograms of the teeth that would constitute a possible disseminating focus.

The presence of other infectious foci, potentially located in skin, gastrointestinal tract, eyes, ears, nasal cavity with sinuses, urinary tract, uterine appendages, osteoarticular system, and others was excluded in the course of specialist clinical examination.

Medical histories of the patients concerned recent or recurrent infective endocarditis and coexisting acquired heart disease. In all patients the teeth diagnosed as constituting infectious foci were extracted. Since all patients included in the study had to have 2 or more dentigenous, infectious foci, the extraction of a corresponding number of foci was performed in all subjects.

The material for bacteriological studies included heart valves with inflammatory abnormalities, dentigenous foci, and blood. Conventional culture techniques were applied.

From 60 patients, 76 heart valves (29 mitral, 47 aortal) were taken during cardio-surgical valve replacement procedure. Cut fragments of valves were directly cultured on BHI (brain-heart infusion) broth (bioMerieux), Schaedler broth (bioMerieux) and on thioglycolic medium (bioMerieux), enriched with hemin (Sigma) and menadione (Merck).

The cultures were subsequently incubated up to 10 days at 37°C. Passages from the BHI broth and the Schaedler broth to blood discs and chocolate agar were performed daily, and in the case of observed growth on the thioglycolic medium a passage was conducted to 2 blood discs (Columbia agar, bioMerieux). One of the latter was incubated in anaerobic conditions, using Generbox anaer systems (bioMerieux). Representative bacterial colonies growing in aerobic conditions on solid medium were passed in order to obtain pure bacterial cultures, and were subsequently identified using biochemical tests, appropriately selected to match the species (Api 20E, Api Staph., Api Strepto, Api Coryne) or using the ATB computer system, employing corresponding diagnostic strips. In the case of observing growth exclusively in anaerobic conditions, pure bacterial cultures were diagnosed using Api 20A biochemical strips or the ATB system (Rapid ID 32A).

The ability to produce coagulase by *Staphylococci* was determined by test-tube technique, using rabbit plasma. The drug-resistance of the cultured microorganisms was assessed by diffusion-disc method according to the Polish National

Institute of Hygiene recommendations, using M-H medium (bioMerieux), or the latter medium with sheep blood.

The teeth recognized as potential infectious focus were extracted, after collection of all blood samples, within an 8-hour period, with prophylactic antibiotics administered intravenously (eg, clindamycin). The decision to apply antimicrobial protection was made despite the generally changing views concerning antibiotic prophylaxis accompanying dental procedures. Since tooth extraction is considered a highly invasive procedure, this decision was justified [17–20].

As the material for bacteriological examination of the odontogenic infectious foci served the roots of the extracted, gangrenous teeth, swabs from their periapical areas, from pathological periodontal pockets (>3 mm), and root canals were also taken. All dental, infectious foci encountered in a given subject, irrespective of their number, were cultured immediately after extraction, and the material was directly transferred to the laboratory.

The extracted teeth were cultured on the BHI broth (bioMerieux), while swabs from periapical areas were cultured on BHI broth, Schaedler broth, and on Columbia agar with sheep blood, kanamycin and vancomycin (bioMerieux). The BHI broth was incubated aerobically, whereas Schaedler broth and Columbia agar were incubated anaerobically with the use of gas-paks (BioMerieux).

Upon observing growth on the BHI medium, isolating passages were performed to blood discs, Sabouraud and Drygalski medium and mannitol salt agar (Chapman-agar), and after multiplication representative colonies were subsequently examined biochemically using appropriate biochemical strips (bioMerieux Api system or ATB computer system). If growth was observed only in anaerobic conditions, pure bacterial cultures were diagnosed using Api 20A or Rapid ID 32A biochemical strips (ATB computer system).

The ability to produce coagulase by *Staphylococci* was determined by test-tube technique, using rabbit plasma. The drug-resistance of the cultured microorganisms was determined by the diffusion-disc method, according to the Polish National Institute of Hygiene recommendations, using the M-H medium (bioMerieux), or the same medium with sheep blood.

In all patients with acute endocarditis, samples (20ml) of venous blood were collected 6 times within 48 hours: in the first 24-hour period, 4 times every 6 hours; and during the second such period 2 times every 12 hours, in fully aseptic conditions. The blood samples were collected at the peak of the fever, or just before the predicted peak. For blood cultures, the HEMOLINE kits were used (bioMerieux), warmed to 37°C.

They consisted of 2 bottles: the aerobic one (green), and the anaerobic one (red).

They contained the appropriate liquid media, or there was only 1 bottle, containing biphasic medium, adjusted to accommodate both the aerobic and the anaerobic microorganisms. The biphasic medium consisted of solid phase, aside from the liquid one, of agar medium layer, along the bottle's wall. The material was inoculated on this medium by tilting the bottle

and rinsing the solid medium with the collected blood. After 16–24 hours of incubation at 37°C the potential growth of microorganisms on solid medium was assessed, from which isolation and passing were conducted. If no microorganism growth had been observed, the incubation was continued for 2 weeks, with continual monitoring of the media and daily culturing by turning the bottle. In order to obtain aerobic bacteria from agar medium, they were passed after 2 days to the chocolate (Levine) and mannitol salt (Chapman) media, as well as the Sabouraud medium enriched with blood. Cultures of anaerobic bacteria were obtained after 2 days from their inoculations on Schaedler blood medium. Preparations from blood and broth stained by Gram method were also performed. Representative colonies were subsequently biochemically tested after multiplication, with the use of appropriate biochemical Api strips. Drug-resistance of the cultured microorganisms was determined by the diffusion-disc method, using M-H medium (bioMerieux), or the same medium with sheep blood. Biphasic media were employed because they limit the contact with the examined material, and thereby the possibility of its accidental contamination.

The HEMOLINE media are rich, enabling the growth of a variety of microorganisms. They are distinguished by prompt growth, particularly of the following rods: *Enterobacteriaceae*, *Pseudomonas*, *Staphylococcus*, and *Streptococcus*. For even more precise bacterial diagnostics, the following media were additionally used:

- Hemomedican (bio Med) – transport-growth medium for aerobes,
- Signal (Oxoid) – for aerobic and anaerobic bacteria,
- Vacutainer (Becton, Dickinson) - for aerobic and anaerobic bacteria,
- Septi Chek (Roche),
- Isolator (Merck) – transport medium.

In all patients with subacute bacterial endocarditis, blood samples were collected only 2 or 3 times. The culture was observed and passed throughout 2 weeks. The diagnostic criteria for infective endocarditis as proposed by Durack et al from Duke University, recently modified, were adopted [21,22]. Overall, 2 major and 5 minor criteria, and the suggested modifications, were taken into consideration. Pathological criteria were histological or bacteriological demonstration.

Major criteria were positive blood cultures (microorganism specific for endocarditis or persistently positive blood cultures) and endocardial involvement (vegetations on valves, valve-rings, additional echoes in typical localisations, oscillation, intracardial abscess, new valvular incompetence or new perivalvular leak); modification: also positive Q fever serology, *Staphylococcus aureus* bacteriemia.

Minor criteria were history of a defect or other heart disease, predisposing to infective endocarditis, drug abuse), fever over 38°C, vascular symptoms (aortal embolism, septic pulmonary embolism, mycotic aneurysm), positive blood cultures (not meeting a major criterion), immunological reactions, echocardiography suggesting infective endocarditis but not meeting major criteria; modification: diagnosis based on echocardiography is no longer an additional criterion. For diagnosis of infective endocarditis, either 2 major criteria or 1 major and 3 minor or 5 minor criteria were

necessary. Irrespective of these criteria, of importance is also: a surgical procedure or infection within the last 2 months.

It has become possible, by applying these criteria, to increase the probability of formulating prompt and definitive diagnoses of endocarditis [21,23].

The European Society of Cardiology has recently proposed new guidelines concerning prevention, diagnosis, and treatment of this disease [24]. The periodontal diseases were diagnosed according to the Community Periodontal Index of Treatment Needs (CPITN). It is a modern tool that not only precisely assesses periodontal status but is also capable of indicating possible therapeutic approaches [25]. The method consists of the specially constructed periodontal probe, and the division of dentition into sextants, scoring index teeth in each sextant. The highest index found is recorded for each sextant, applying the following scores: 0 – healthy, 1 – bleeding on probing, 2 – calculus and/or overhanging restorations, 3 – pocket depth to 5.5 mm, 4 – deep pocketing of 5.5 mm or greater. If only 1 tooth existed in any one sextant, it was, by default, added to the nearest sextant. Periodontal tissues were assessed by the surface of the teeth. In addition, the number of existing teeth in each subject was recorded.

Aside from the above-mentioned index, another well-established diagnostic method, the Scandinavian Classification, was also employed. In contrast with the CPITN, it involves radiology, and thus, the possibility of assessing lesions not visible to the naked eye (eg, vertical-horizontal alveolar resorption). Moreover, it includes the degree of tooth mobility, signs and symptoms of gingivitis, the presence of cervical line, and clinically visible loss of epithelial attachment of the gingiva (Table 1).

For statistical analysis the Fisher's and the Pearson's chi-square test were applied.

The subjects' rights have been protected by the University Ethics Committee, and informed consent was obtained. The project was financially supported by the university's own research resources.

RESULTS

The studied group comprised 60 patients: 32 males (53.30%), and 28 females (46.70%), aged 19–69 (average 49). Age and sex of particular subjects, with corresponding 3-fold culture results are presented in Table 10. The most patients had 2 major criteria for the diagnosis of endocarditis (31 subjects, 51.67%). In 27 subjects (45.00%) 1 major (13 patients had positive cultures, and 14 subjects had echocardiographic evidence) and 3 minor criteria occurred. Five minor criteria were present in 2 patients (3.33%).

Gram-positive bacteria (cocci and anaerobic rods) were cultured from dentigenous, infectious foci in 60 patients with acute or subacute infective endocarditis. Altogether, 328 teeth constituting a dental, infectious focus were extracted (average 5.47 per patient). The remaining, healthy teeth were not extracted.

Table 2 presents the types of dentigenous foci in the 8 patients with acute infective endocarditis¹. The foci in this group were

Table 1. Periodontitis criteria, Scandinavian classification.

	Feature	Periodontitis degrees			
		Gingivitis	Mild – 1 st	Severe – 2 nd	Complicated – 3 rd
1.	Bleeding during probing of gingival pockets or with pressing the gingiva	+	+	+	+
2.	Depth of gingival pockets	Up to 3.5 mm	4–5.5 mm	6 mm and more	Over 6 mm
3.	Concerning tissues	Gingiva	Uniform atrophy of tissues supporting teeth in sockets, slow bone atrophy to within 1/3 tooth's length	Uniform atrophy of tissues supporting teeth in sockets, slow bone atrophy to within 1/3 tooth's length	Irregular, bone loss of alveolar process' margin (crater). Tissue loss 2 nd and 3 rd -order in bifurcation area. Bone atrophy to within 1/3 tooth's length
4.	Cervical line visible	-	+	+	+
5.	Epithelial attachment's loss clinically visible	-	+	+	+
6.	Gingivitis symptoms	+	+	+	+
7.	Loose teeth	-	+	++ Supra- and subgingival calculus	+++ (2 nd -order) Supra- and subgingival calculus

Table 2. Types of dentigenous foci: intradental and periodontal in acute endocarditis.

Acute endocarditis			
Type of focus	Periodontitis degree	Number of subjects	%
Intradental (1 st order)	1 st	1	12.5
	2 nd	5	62.5
	3 rd	2	25.0
Total		8	100.0

$\chi^2=0.001$; $p=0.974$.

all intradental, along with any of the 3 degrees of periodontitis. The most frequently encountered was 2nd degree (62.50% of the examined group). Periapical foci were not found.

In spite of the relatively large percentage difference between periodontic degrees (62.50% 2nd degree vs. 37.50% 1st and 3rd degree) the calculation of statistical significance was not possible, due to the small number of patients with acute endocarditis (n=8).

Tables 3 and 4 present types of dentigenous foci in 52 patients with subacute infective endocarditis, with degrees of periodontitis. Intradental foci were analogically the most frequently encountered, along with 2nd degree periodontitis, in 22 patients (42.30%), whereas the least frequent were the periapical foci, with 1st degree periodontitis – only in 1 patient (1.90%). There is a moderate and significant association between the category of infective focus and periodontic degree. The occurrence of 2nd degree periodontitis was found to be significantly more frequent in the

intradental foci (42.30%), $p=0.024$. However, significantly less frequent was the occurrence of 3rd degree periodontitis in intradental foci (11.50% of the examined group), as compared with the periapical type (11.50% vs. 23.10%), $p=0.008$. No significant difference in frequency was established between intradental and periapical foci, 5.80% and 1.90%, respectively.

No statistical association was found between the periodontic degree and the course of endocarditis (acute or subacute) in patients with intradental odontogenic foci.

Table 5 presents the correlation between bacterial species cultured from heart valves and from dentigenous infective foci. A consistency was established between culture results from the 2 distinct tissue types and it was significantly greater in acute endocarditis.

Corresponding bacterial culture results from heart valves and from dentigenous infectious foci (irrespective of endocarditis form, whether acute or subacute) were obtained in 30 patients (50.00%). In 24 patients (40%) the cultures did

¹ $\chi^2=0.001$, $p=0.974$.

Table 3. Types of dentigenous foci: intradental, periapical and periodontal in subacute endocarditis.

Subacute endocarditis			
Type of focus	Periodontitis degree	Number of subjects	%
Intradental (1 st order)	1 st	3	5.8
	2 nd	22	42.3
	3 rd	6	11.5
Subtotal		31	59.6
Periapical (2 nd order)	1 st	1	1.9
	2 nd	8	15.4
	3 rd	12	23.1
Subtotal		21	40.4
Total		52	100.0

$\chi^2=7.903$; $p=0.019$; $C=0.363$.

Table 4. Summary data concerning acute and subacute endocarditis v. periodontitic degrees for intradental foci.

Periodontitis degree	Acute endocarditis (N=8)		Subacute endocarditis (N=52)		Total (N=60)	
	N	%	N	%	N	%
1 st	1	12.5	3	9.7	4	10.3
2 nd	5	62.5	22	70.9	27	69.2
3 rd	2	25.0	6	19.4	8	20.5
Total	8	100.0	31	100.0	39	100.0

N=number of subjects; $\chi^2=0.214$; $p=0.898$.

not correspond, while in 6 patients (10.00%) they were negative (absent) (Table 6).

Table 7 presents bacterial species cultured from heart valves and from blood in both forms of endocarditis. From among 8 acute endocarditis patients, in 6 cases the cultures were positive and consistent (occurring both in heart valves and blood). The bacteria here belonged to the genus *Staphylococcus*.

In subacute endocarditis, corresponding (consistent) results were obtained in 15 patients (28.80%). *Staphylococcus*, *Streptococcus* and *Gemella* were the most frequently cultured bacteria here.

Table 8 presents both the regular, and percentage, corresponding results of cultures from dentigenous infectious foci and blood. This Table also illustrates the number of affected heart valves. There was no association between corresponding results of cultures from dentigenous infectious foci and blood, and the number of affected heart valves. Corresponding results of cultures from odontogenic infectious foci and blood from the whole studied group were obtained in 25 patients (41.70%). *Staphylococcus* occurred in 52.00%, and the remainder were *Streptococcus* (36.00%) and *Gemella* in 12.00%.

Table 9 presents results of corresponding bacterial flora cultures from dentigenous infectious foci, blood and heart valves (3-fold consistency) in patients with both acute and subacute

forms of endocarditis. Such results were obtained from the above-mentioned sources in 14 patients (23.3%). From among 8 patients infected by genus *Staphylococcus*, 6 had the acute form of endocarditis and the remaining 2 had the subacute form. However, in a total of 8 patients with subacute form (3-fold conformity of the culture results), the bacterial flora was more differentiated, and consisted of, in the 2 cases mentioned, genus *Staphylococcus*, in 4 cases genus *Streptococcus*, and in 2 patients genus *Gemella* was cultured.

Association between infection of aortal valve by genus *Staphylococcus* and infection of the mitral (and/or both mitral and aortal) valve was on the verge of statistical significance.

Association between the 3-fold consistency of the culture results, and the number of affected valves was also on the verge of significance.

Table 10 presents culture results from dentigenous, infectious foci, blood and heart valves with patient sex and age, as well as the type of valve defect and infectious focus.

No significant difference was found with respect to sex of the subjects here. Similarly, no such statistical difference occurred with regard to age.

There was also no correlation between age and the type of heart valve defect.

Table 5. Corresponding bacterial species cultured from heart valves and from dentigenous foci in acute and subacute endocarditis.

	Microorganism	Infective endocarditis		Total
		Acute	Subacute	
1	<i>Staphylococcus aureus</i>	3	1	4
2	<i>Staphylococcus epidermidis</i>	3	–	3
3	<i>Staphylococcus haemolyticus</i>	–	1	1
4	<i>Staphylococcus hominis</i>	1	2	3
5	<i>Streptococcus milleri</i>	–	2	2
6	<i>Streptococcus salivarius</i>	–	1	1
7	<i>Streptococcus sanguis</i>	–	7	7
8	<i>Gemella morbillorum</i>	1	2	3
9	<i>Aerococcus viridans</i>	–	1	1
10	<i>Proteus vulgaris</i>	–	1	1
11	<i>Corynebacterium pseudodiphtheriticum</i>	–	1	1
12	<i>Micrococcus luteus</i>	–	2	2
13	<i>Prevotella oris</i>	–	1	1
Total corresponding		8	22	30
Non-corresponding cultures		–	24	24
Negative cultures		–	6	6
Total		8	52	60

Table 6. Consistency of culture results from heart valves and from dentigenous infectious foci in acute and subacute infective endocarditis.

Endocarditis	Acute		Subacute		Total	
	N	%	N	%	N	%
Corresponding	8	100.0	22	42.3	30	50.5
Non-corresponding	0	–	24	46.2	24	40.0
Negative	0	–	6	11.5	6	10.0
Total	8	100.0	52	100.0	60	100.0

The association between 1st order focus and 3-fold consistency of culture results is significant and non-incident (p<0.017).

Moreover, regarding age of the subjects and type of infectious focus in acute endocarditis, it was established that *Staphylococcus aureus* species occurred in relatively younger subjects. Their average age was 42 years, the type of focus was 1st order (intradental), with 2nd and 3rd degree periodontitis, and aortal valve defect was the only type encountered.

However, in subacute endocarditis (in consistent culture results from dentigenous foci, blood and heart valves) the genus *Staphylococcus* occurred in 2 subjects, whose average age was 50 years; they had mitro-aortal valvular disease, 1st order foci, and 2nd and 3rd degree periodontitis. In 2 subjects

the species *Gemella morbillorum* was cultured from the 1st order foci, with 2nd degree periodontitis, and the valvular defect was mitro-aortal.

In the remaining patients the genus *Streptococcus* was cultured in all cases from 1st order focus, and with 2nd degree periodontitis.

DISCUSSION

In the 8 patients with acute endocarditis, only the intradental foci occurred. They were accompanied by 2nd degree periodontitis, 62.50% of the studied group, due probably to their oral hygiene, general dental condition, deep dental caries, occurrence of endo-perio syndrome (discussed below), with gingival pockets 5.5–6.0mm deep, and non-endodontic approach (x-ray confirmed).

Table 7. Bacterial species cultured from heart valves and blood in acute and subacute endocarditis.

Cultures from heart valves and blood			
Microorganism	Infective endocarditis		Total
	Acute	Subacute	
1 <i>Staphylococcus aureus</i>	3	1	4
2 <i>Staphylococcus epidermidis</i>	3	5	8
3 <i>Staphylococcus hominis</i>		2	2
4 <i>Streptococcus milleri</i>	–	1	1
5 <i>Streptococcus salivarius</i>	–	1	1
6 <i>Streptococcus sanguis</i>	–	2	2
7 <i>Gemella morbillorum</i>	–	3	3
Total corresponding	6	15	21
Non-corresponding cultures	–	21	21
Negative cultures	2	16	18
Total	8	52	60

$\chi^2=5.549$; $p=0.018$; $C=0.305$ it was significantly greater in acute endocarditis.

Table 8. Consistency of culture results from dentigenous, infectious foci and blood v. number of affected heart valves in acute and subacute endocarditis.

Microorganism	Endocarditis n=25		Subtotal	Per cent (%)	Valves		
	Acute	Subacute			Mitral	Aortal	Mitro-aortal
1 <i>Staphylococcus aureus</i>	3	3	6	24.0	1	3	2
2 <i>Staphylococcus epidermidis</i>	3	2	5	20.0	2	2	1
3 <i>Staphylococcus hominis</i>	–	2	2	8.0	–	1	1
4 <i>Streptococcus milleri</i>	–	3	3	12.0	1	1	1
5 <i>Streptococcus pneumoniae</i>	–	1	1	4.0	–	–	1
6 <i>Streptococcus salivarius</i>	–	1	1	4.0	1	–	–
7 <i>Streptococcus sanguis</i>	–	4	4	16.0	1	3	–
8 <i>Gemella morbillorum</i>	–	3	3	12.0	–	1	2
Consistent	6	19	25	100.0	6	11	8
Inconsistent	–	18	18		2	11	5
Negative	2	15	17		4	10	3
Total	8	52	60		12	32	16

$\chi^2=2.545$; $p=0.637$; n – number of bacterial cultures.

In those with subacute form of the disease, both intra-dental and periapical foci were established (40.0% and 60.0%, respectively). Second-degree periodontitis occurred in 42.3%, while 1st and 3rd degree occurred in 17.3%. The domination of 2nd degree periodontitis here was due, despite generally different bacterial flora, to the same causes, as in the case of patients with acute endocarditis, with the exception of endodontic treatment having

been applied in approximately half of the subacute endocarditis group.

Where the results of the cultures from dental infectious foci, blood and heart valves corresponded (ie, the same bacterial flora were cultured from these foci), in patients with acute as well as subacute endocarditis only the intradental foci showed up, along with 2nd degree periodontitis (83.30% and



Table 9. Total correspondence of culture results from dentigenous, infectious foci, blood, and heart valves in patients with acute and subacute endocarditis.

	Microorganism	Infective endocarditis		Total	Heart valves		
		Acute	Subacute		Mitral	Aortal	Mitral-aortal
1	<i>Staphylococcus aureus</i>	3	1	4	–	3	1
2	<i>Staphylococcus epidermidis</i>	3	–	3	1	2	–
3	<i>Staphylococcus hominis</i>	–	1	1	–	–	1
4	<i>Streptococcus milleri</i>	–	1	1	–	1	–
5	<i>Streptococcus salivarius</i>	–	1	1	1	–	–
6	<i>Streptococcus sanguis</i>	–	2	2	1	1	–
7	<i>Gernella morbillorum</i>	–	2	2	–	–	2
	Total	6	8	14	3	7	4

Table 10. Corresponding culture results from dentigenous foci, blood and heart valves, with regard to age, periodontitis degree and focal order (intradental and periapical). p<0.017.

Microorganism	Patients' age						Focus	Periodontitis degree				
	Females			Males				Intradental	Periapical	1 st	2 nd	3 rd
	M	A	M-A	M	A	M-A						
Acute endocarditis												
1	<i>Staphylococcus aureus</i>	–	42	–	–	–	1	–	–	1	–	
2	<i>Staphylococcus aureus</i>	–	–	–	–	–	1	–	–	1	–	
3	<i>Staphylococcus aureus</i>	–	53	–	–	–	1	–	–	–	1	
4	<i>Staphylococcus epidermidis</i>	–	–	–	42	–	1	–	–	1	–	
5	<i>Staphylococcus epidermidis</i>	–	–	–	–	46	1	–	–	1	–	
6	<i>Staphylococcus epidermidis</i>	–	–	–	–	56	1	–	–	1	–	
Subacute endocarditis												
1	<i>Staphylococcus aureus</i>	–	–	–	–	49	1	–	–	–	1	
2	<i>Staphylococcus hominis</i>	–	–	51	–	–	1	–	–	1	–	
3	<i>Streptococcus milleri</i>	–	28	–	–	–	1	–	–	1	–	
4	<i>Streptococcus salivarius</i>	–	–	–	53	–	1	–	–	1	–	
5	<i>Streptococcus sanguis</i>	–	61	–	–	–	1	–	–	1	–	
6	<i>Streptococcus sanguis</i>	–	–	–	49	–	1	–	–	1	–	
7	<i>Gemella morbillorum</i>	–	–	58	–	–	1	–	–	1	–	
8	<i>Gemella morbillorum</i>	–	–	51	–	–	1	–	–	1	–	

M – mitral valve; A – aortic valve; M-A – mitro-aortic valve.

87.50%, respectively). It is supposed that these latter foci are characterised by particular, 'toxic', affinity to valvular endocardium, especially in patients with acute, infective endocarditis, caused by genus *Staphylococcus*. Probably the bacteria inhabiting gingival pockets of depth up to 5 mm (2nd degree periodontitis) constitute the flora that are also most

virulent for endocardium. However, from the purely clinical point of view, the 2nd degree periodontitis is regarded as less of an issue in comparison with the 3rd degree (Table 1).

With reference to otherwise healthy patients, even the most recent reviews comparing endodontic and surgical

approaches and the one (resection, extraction, hemi-section, implant provision) do not clearly support either method of treatment over the other as regards long-term success [26–32].

This study's results, however, appear to indicate that tooth extractions are recommended during treatment of acute, infective endocarditis, where the cause is evident and clearly related to the presence of odontogenic infectious foci in general, and intradental foci in particular, accompanied by 2nd degree periodontitis. Endodontic treatment, typically administered in generally healthy (eg, non-endocarditic) patients with intradental, inflammatory processes, is insufficient and even hazardous in this high-risk group, with gangrenous pulp present in main and lateral root canals (specific to intradental foci). The gangrenous pulp in lateral root canals cannot be removed, and may be subject to mummification during endodontic treatment, constituting a constant, infective focus. Hence, the logical approach here is extraction of teeth with intradental foci and 2nd degree periodontitis.

In the case of subacute endocarditis, however, due to its long-lasting course and the diversified bacterial spectrum, the outcome of this procedure may prove to be less beneficial, though still viable. This specific coincidence of intradental foci with 2nd degree periodontitis may be more difficult to explain. It is possible that the endo-perio syndrome plays an important role here. Even a small amount of microorganisms may leave the root canal of a gangrenous tooth through lateral canals (root-periodontal canals) or through apical foramen to periodontium, gingivae and alveodental process [33]. Second-degree periodontitis causes hyperemia in the tissues surrounding teeth, which intensifies transport and penetration of the bacteria further out to the periphery, while gingival pockets exceeding 5 mm in depth may constitute a proper biotope only for the group of microorganisms with apparent affinity to the affected valvular endocardium. It has been suggested that elastase released by bacteria typically found in oral cavities may damage elastine, an element of the alveolo-dental ligament, which leads to its destruction and eventually to the deterioration of the adjoining periodontium. A purified elastase, in the form of cysteine protease (1 of the 3 protease classes typical for the species *Staphylococcus aureus*) has, through research, been obtained [34]. Can the gangrenous, dental pulp in the anaerobic domain of the root canal be a suitable biotope for the microorganisms able to spread on to periodontium and the adjacent tissues, and thus become virulent for the host? In all patients in this study an acquired heart disease had previously been diagnosed. Elastin is also a proteinic component of the cicatrised endocardium connective tissue. Would the affinity of *Staphylococci* to valvular endocardium be related to enzymes, active only in the conditions earlier profiled by plentiful bacteria of the infected periodontium, and hardly ever cultured from the infected heart valves? It is not known what enables the formation and stabilization of the pathogenic dental plaque, itself of altered ecology. The important differences, both in the host response to plaque bacteria, and the ecological interactions between the bacteria themselves, are the most likely explanation. Temporary occurrence of the potentially pathogenic microorganisms among bacterial flora of the plaque (itself of morbid ecology), by enhancing its formation may also result in the destructive disease of the periodontium. This initiating bacterial group may include

the genera *Staphylococcus* and *Streptococcus*, released from root canals. This premise is something intermediate between 2 other hypotheses, concerning the specific and non-specific plaque, neither of them are apparently directly related to the destructive disease of the periodontium. The relationship between 2nd degree periodontitis and intradental foci is complex. Even if the pathogenic factor is explicitly linked with the progress of periodontitis, it is difficult to establish whether it is the cause or the effect of the above-mentioned change in the bacterial flora of the plaque. The medical history of the patient alone may not suffice to determine whether the dental caries had originally led to the bionecrosis of the pulp, or that periodontal changes influenced the formation of the intradental, infectious foci in the studied group of 60 patients. It is difficult to say why the acute, infective endocarditis was accompanied by only intradental foci, whereas its subacute form developed in the presence of both types of foci. This study's results indicate that intradental and 2nd degree periodontitis may constitute a dentigenous, tooth-related risk factor of acute, infective endocarditis.

In patients with its subacute form, it takes a relatively long time for the intradental infective focus to develop (ie, from the necrotic dental pulp phase to the formation of a periapical focus). This was confirmed by medical histories taken from the studied group and by the results of x-ray examinations. Root canals that were not fully filled, with partially remaining necrotic pulp, were the possible cause. In other group of patients the cause would be deep caries, treated conservatively, resulting in (unbeknownst to the patient) necrotic pulp.

Furthermore, the most 'virulent' genus, *Staphylococcus*, is often absent in the bacterial flora of dead pulp in patients with the subacute form of endocarditis. In consistent culture results (odontogenic foci, blood, and heart valves) *Staphylococcus* occurred only in 2 such patients, and only when intradental foci were involved (Table 10). In the bacterial flora of devitalised pulp of patients with acute endocarditis, *Staphylococcus* was also, at all times, accompanied by *Streptococcus* and *Gemella*. It is equally conceivable that the *Streptococcus* bacteria influence the growth and virulence of the genus *Staphylococcus* [35]. The meaning of the coexistence of the 3 bacterial genera in the dead pulp environment, and their effect on initiation of acute, infective endocarditis, are unclear. The number of bacterial strains of the genus *Staphylococcus* occurring in root canals may be of consequence here. It has been demonstrated that inflammatory reactions within periapical tissue correlated with the amount of bacteria introduced and not with their genus [36, 37].

The possibility that the presence of either *Staphylococcus* or other bacterial strains could result from contamination from other sites must be regarded as highly unlikely. The specimens were collected in aseptic operating room conditions. The bacteriological lab environment, systematically monitored, practically excluded the chance of accidental contamination.

The Rosenow theory on the variability ("transmutation") of microorganisms may prove to be helpful in verification of these conjectures [38].

Evidence on the prevalence of both types of infective foci in a healthy population indicates that periapical foci occur

more frequently (73%), while the intradental foci occur in 27% of teeth with gangrenous pulp, either subject to root canal therapy or not treated [39]. The results of a series of studies conducted by ophthalmologists on their patients (with a total of 1393 teeth) show that only chronic, periapical foci occurred in that group [40]. It is remarkable that in patients with both acute and subacute endocarditis, the bacterial culture results pointed only to the intradental foci. Could they alone initiate the development of infective endocarditis? Why was the genus *Staphylococcus* at all times accompanied only by intradental foci?

The results presented here correspond with those of Kronfeld, who maintained that granuloma is not the site where the bacteria thrive but rather where they are eliminated [41]. It is known, however, that the connective tissue capsule of periapical granulomas constitutes a barrier, effectively preventing the bacteria from migrating outward. This shell, while limiting the microorganisms' ability to spread, also probably contributes to the changes in the environment of the root canal's gangrenous pulp, and thus its bacterial flora. It is also difficult to establish whether microorganisms can penetrate the inside of a granuloma, then thrive and multiply there. Precise studies, backed by electron microscopy, demonstrated that the bacterial presence is, in most cases, limited to the interior of the root canal, where they are separated from granulomas or capsules by a cast of polynuclear white blood cells [42].

However, in clinical practice there are cases of chronic inflammation of periapical tissue becoming exacerbated, in the form of endodontal abscess (ie, originating from gangrenous pulp), as well as cases of periapical lesions resistant to therapy. A study of 31 teeth with periapical changes, assisted by both light and electron microscopy, revealed bacteria in the canals of all 31 teeth examined [42]. On the other hand, in bacteriological examination of granulomas and root cysts, the same bacteria were found in only 5 cases. According to Nair, morphological assessment, aided by electron microscopy, clearly suggests that the bacteria did not penetrate into the granulomas in the course of a medical manipulation (tooth extraction), but having filtered through to the periapical lesions from root canal, they began to stimulate the host tissues to the immunological response [43]. Others, examining periapical granulomas in animal models, arrived at similar conclusions [44].

Some authors have maintained that the bacteria external to the root canal might exist in infected periapical changes and in the infected cyst [45–47]. Apart from these exceptions, the old theory of Harndt and Kronfeld that granulomas and cysts are free from bacteria is apparently still valid [41]. At least 1 author arrived at a similar conclusion [39]. The foregoing brief review of pertinent literature suggests that no single bacterial species or genus may be defined as etiologically specific in the domain of periapical tissue diseases. All bacteria residing in the root canal should be regarded as potential pathogens, and therapy should be directed at their eradication.

CONCLUSIONS

The concerted occurrence of the same bacterial flora on both the dental foci as well as on heart valves and in blood

in 23.30% of the studied group (14/60) appears to indicate the role of the dentigenous infective foci in the pathogenesis of infective endocarditis, especially its acute form.

In acute endocarditis, intradental foci were established in all cases (100%), and 2nd degree periodontitis was established in 62.50% of cases. In its subacute form, the intradental foci appeared in 59.60% of the group, whereas 2nd degree periodontitis appeared in 42.30%. Most of the bacterial strains cultured from intradental foci and 2nd degree periodontal lesions reveal affinity to valvular endocardium, suggesting that these infective foci, among all other dental foci, may be regarded as a dentigenous risk-factor of infective endocarditis.

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