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Microbiological studies on denture-induced stomatitis in children

Keiko Shimizu, DDS Fumio Shimizu, DMD, PhD Kikuo Kamiyama, DDS, DDSc

Abstract

Denture-induced stomatitis in children was studied microbiologically. Quantitative cultural studies on Candida and bacteria were done in 17 denture wearers with or without palatal denture-induced stomatitis (erythema score 0-3) and in 16 healthy nondenture wearers. It was found that significantly higher numbers of Candida both in denture plaque and in saliva were cultured from the denture wearers as the clinical condition of the palatal mucosa deteriorated. Almost identical numbers of bacteria, however, were isolated from the denture wearers, irrespective of erythema score. It was also shown that the isolation rates of Candida both from denture plaque and from saliva was 100% in subjects with erythema scores of 1-3, but approached 50% in subjects with an erythema score of 0. Studies on the saliva from nondenture wearers revealed that both the isolated numbers of microbes and the isolation rate of Candida were almost identical with those of subjects with an erythema score of 0. The most frequently isolated species of Candida was Candida albicans A. These studies suggest that Candida might be an etiologic factor for denture-induced stomatitis in children.

Denture-induced stomatitis (DS) is a frequent complication in children and adults wearing removable dentures or space maintainers (Budtz-Jörgensen et al. 1975). DS is a term used to describe certain pathologic changes found in the oral mucosa of denture-bearing tissues (Budtz-Jörgensen 1974). These changes are characterized by a bright erythema and are found under dentures in both jaws, but most frequently in the maxilla (Bergendal and Isacsson 1983).

Several etiologic factors may be associated with DS. Microbes can be one of the candidates. Culture studies indicate that this condition is associated with a quantitative increase in yeasts on the mucosal surface of maxillary dentures and the lesions in the plate (Budtz-Jörgensen et al. 1975; Arendorf and Walker 1979). Other studies, however, suggest that bacteria may play a causative role in the disease (Van Reenen 1973; Holbrook 1979). Second, allergic response of the host to resins has been suggested as one causative agent (Giunta et al. 1979; Devlin and Watts 1984). Third, physical injury induced by a poorly fitting denture can be a significant etiologic factor (Stohler 1984). In addition, some of the factors described may work together. Many studies on DS have been done in adults, but there is no known report in the literature on DS in children.

In the present study, DS in children was investigated microbiologically. This report suggests that Candida might be one possible cause of the disease.

Materials and Methods

Normal Subjects

Sixteen healthy, nondenture wearers were recruited from dental outpatients who visitedthe pedodontic clinic of Tohoku University Dental Hospital (six males and 10 females, average age seven years, seven months). Individuals with a medical condition or those taking drugs were excluded from the study.

Subjects with Palatal Denture

Seventeen individuals who wore a maxillary denture as a space maintainer were divided into four groups, depending on their erythema score (Bergendal and Isacsson 1980, Table 1). The erythema score was determined by the same examiner, taking oral photographs as records (six males and 11 females, average age seven years, four months).

Collection of Denture Plaque

Plaque for the study was collected by washing the mucosa surface of the maxillary denture with a sterile toothbrush in 2 ml phosphate buffered saline (PBS). Plaque was then passed four to five times through a 25-gauge needle to dissolve the aggregates.

Table 1. Erythema Score

0: Normal pink
1: Slightly erythematous mucosa
2: Moderately erythematous mucosa
3: Pronouncedly erythematous mucosa

Salivary Samples

Each participant deposited approximately 1 ml of mixed, whole, unstimulated saliva into a sterile plastic tube. The saliva was passed four to five times through a 25-gauge needle to dissolve the aggregates.

Microbiological Methods

Several dilutions of samples up to 10^6 were prepared in PBS and 0.1 ml was inoculated onto agar containing 5% rabbit defribrinated blood. The agar plates were anaerobically incubated in jars containing sodium borohydride, sodium bicarbonate, and citric acid[a] for six days at 37°C and then the number of colony forming units (CFU)/ml was calculated. Counts of Candida were carried out by inoculating 0.1 ml of undiluted, 10^{1} and 10^{-2}-fold diluted suspensions onto Candida GE agar plates.[b] CFU/ml were determined after incubation in air for two days at 37°C. Candida species were identified using a Candida check kit.[c]

Results

CFU in Denture Plaque

Figure 1 shows the CFU of Candida and bacteria from denture plaque collected from individuals with an erythema score of 0-3. CFU of Candida increased with higher erythema scores. At erythema scores of 0, 1, 2, and 3, average CFU/ml was $10^{1.1\pm0.6 (SD)}$, $10^{3.2\pm0.7}$, $10^{4.4\pm0.8}$, and $10^{4.8}$, respectively. There was a high degree of correlation ($r = 0.73$, $P < 0.01$) between the Candida CFU and the erythema score. The CFU/ml of total bacteria, however, were almost identical in any score, that is, $10^{8.0\pm0.9}$ (Fig 1). These data clearly show that there is a close relationship between the erythema score and the quantitative increase in Candida.

CFU in Saliva

The authors were interested in whether similar findings could be observed in studies on saliva. As shown in Figure 2(A), the CFU of Candida in the saliva increased with a higher erythema score, showing results similar to those in the denture plaque. There was a high degree of correlation ($r = 0.69$, $P < 0.01$) between the quantitative increase in Candida and the erythema score. The CFU/ml of total bacteria, however, were approximately $10^{7.8\pm0.5}$ at any score [Fig 2(A)]. The saliva from nondenture wearers was then examined. It was observed that the CFU/ml of Candida and total bacteria were $10^{0.8\pm1.2}$ and $10^{7.6\pm0.4}$, respectively [Fig 2(B)]. These values were similar to those in denture wearers with erythema score of 0.

[a] Gas pak - Becton Dickinson and Co; Cockeysville, MD.

[b] Candida GE agar - Nissui Pharmaceutical Co; Tokyo, Japan.

[c] Candida check - Iatron Lab, Inc; Tokyo, Japan.

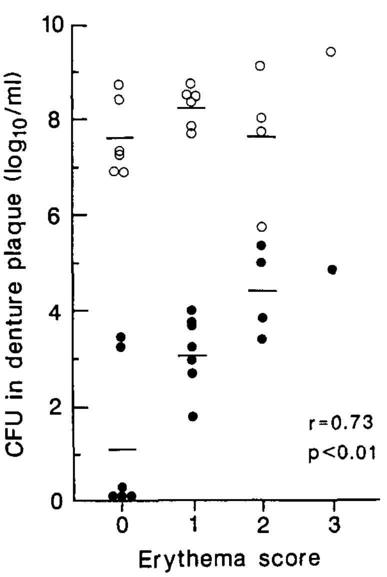

FIG 1. The CFU of Candida (•) and bacteria (○) in denture plaque obtained from denture wearers. "___" indicates the mean of values. There was a high degree of correlation ($r = 0.73$, $P < 0.01$) between the number of Candida and the erythema score.

Isolation Rate of Candida

Table 2 shows the isolation rate of Candida from denture plaque and saliva, calculated from Figures 1 and 2. The isolation rate of Candida from denture plaque was 33.3% for erythema score of 0, and 100% for erythema scores of 1-3. Similar findings were observed in the saliva study. In addition, the isolation rate of Candida from saliva obtained from the nondenture wearers was 43.8% and similar to that of denture wearers with erythema score of 0 (Table 2).

TABLE 2. Isolation Rate of Candida

Samples	Denture Wearer (Score) 0	Denture Wearer (Score) 1-3	Non-Denture Wearer
	(%)	(%)	(%)
Denture plaque	2/6 (33.3)	12/12 (100)	
Saliva	3/6 (50)	12/12 (100)	7/16 (43.8)

Identification of Candida Species

In the next experiment, the authors identified the

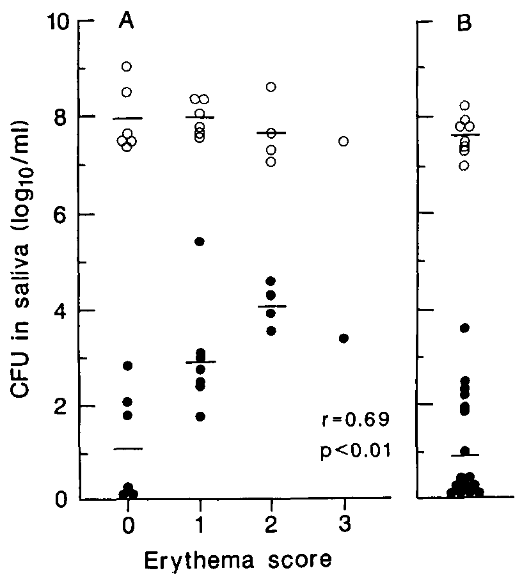

FIG 2. The CFU of Candida (•) and bacteria (○) in saliva obtained from denture wearers with various erythema scores (A) and nondenture wearers (B). "___" indicates the mean of values. There was a high degree of correlation ($r = 0.69$, $P < 0.01$) between the quantitative increase in Candida and the erythema score.

Candida species as described in the Materials and Methods section. *Candida albicans A* was shown to be the most frequently isolated species from denture plaque and saliva (Table 3).

TABLE 3. Identification of Species of Candida

Species	*Denture wearer* Denture plaque	*Denture wearer* Saliva	*Nondenture wearer* Saliva
	(%)	(%)	(%)
C. alb. A	66.7	81.8	66.7
C. alb. B	11.1	9.1	16.7
C. guillermondii	0.0	9.1	0.0
C. krusei	11.1	0.0	0.0
C. tropicalis	0.0	0.0	16.7
Unknown	11.1	0.0	0.0
Numbers tested	9	11	6

Discussion

In this experiment, denture-induced stomatitis in children was studied microbiologically. The authors demonstrated that significantly higher numbers of Candida, both in denture plaque and in saliva, were cultured as the severity of the palatal mucosa condition increased. Isolation rates of Candida, both from the denture plaque and from the saliva, were 100% in patients affected with DS (erythema score 1-3), while nearly 50% in denture wearers with erythema score of 0.

It is known that there may be several etiologic factors causing DS. Microbes can be one of the candidates, allergic response to resins has been suggested as another causative agent, and physical injury induced by a poorly fitting denture also can be a significant etiologic factor. In addition, some of these agents may work together.

Culture studies in humans indicate that DS is associated with a quantitative increase in yeast in the denture plaque and palatal lesions (Budtz-Jörgensen et al. 1975). An experimental model of yeast-induced DS also has been reported (Olsen and Bondevik 1978). Other studies, however, suggest that bacteria such as Klebsiella may play a role in the disease (Van Reenen 1973; Holbrook and Russell 1979). In the current experiments, a quantitative increase in Candida was shown in the patients, compared with the denture wearer with an erythema score of 0 and the nondenture wearers. The isolation frequency of Candida from patients with DS was 100% (twice as high as that in subjects without DS). Although no other bacteria were identified, these results suggest a significant correlation between DS in children and the quantitative increase in Candida.

Allergic response of the host against resins, particularly self-polymerizing resins, can be one etiologic agent.[1] Allergic contact stomatitis caused by resin, however, is rarely reported. In these clinical studies, there was a significant correlation between initially high Candida counts and improvement of the clinical condition of the palatal mucosa following antimycotic denture treatment, although in one patient antimycotic treatment had no effect (data not shown). These studies also support the possibility that Candida, rather than allergic response and physical injury, plays a major etiologic role in DS in patients.

It was shown that the studies on Candida in saliva seemed to reflect the state of Candida in denture plaque. This observation suggests that saliva can be used instead of denture plaque to study Candida in DS in children.

It would be interesting to study whether or not Candida isolated from patients with severe DS is more virulent than that from subjects with healthy palatal mucosa.

Also, what mechanisms are involved in the occurrence of Candida-induced stomatitis in children? How does the host immunologically respond to DS? To investigate such sequences, immulogical and histopathological studies on DS in children are under investigation.

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Drs. Keiko and Fumio Shimizu are assistant professors and Dr. Kamiyama is a professor, pedodontics, Tohoku University. Reprint requests should be sent to: Dr. Fumio Shimizu, Dept. of Pedodontics, Tohoku University School of Dentistry, Sendai, 980 Japan.

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Infection control

One of the complaints from the gay community has been that AIDS patients have had difficulty in obtaining dental care. Recently Heraeus Dental Products of New York did a three-state survey in Wisconsin, Indiana, and Illinois. While dentists overwhelmingly said they would use special precautions in treating high-risk patients, such as homosexual males and intravenous drug users, more than half the respondents said they would refuse treatment to AIDS sufferers. Why? The most reasonable answers that can be given are fear, society's prejudices, and ignorance.

Small wonder that there is fear. The number of cases in the United States is more than 28,000 (as of December, 1986) and doubles every 10 months. More than 150,000 have ARC (AIDS-related complex). There is neither a vaccine, nor a cure.

Most AIDS patients are either male homosexuals or bisexuals, intravenous drug users, or promiscuous heterosexuals. They do not fulfill the standards set by our religious forefathers, and are considered by most as social outcasts outside the mainstream of American life. As health care providers, we are not allowed to moralize or pass judgment. We have a duty to treat all patients who need our care, provided we have the ability.

It is unreasonable to assume that we are going to conquer AIDS soon, and it, along with hepatitis and herpes, has brought barrier techniques to dentistry. Rubber gloves, eye protection, and masks are the armamentarium that all dentists must adopt. This calls for behavioral changes on the part of dentists, because very few learned these techniques in school and must now learn their proper use. All dental health providers have this professional responsibility.

It is also necessary to use the autoclave or hot air oven to sterilize instruments and to use glutaraldehyde for those things that cannot withstand the heat. Sodium hypochlorite (bleach) or iodophors should be used for surface disinfection. We should not resheath or break needles, but should place them in a disposal system container for discard.

If dentists refuse to treat AIDS patients, then callers will not identify themselves as AIDS patients. If the dentist does not guess correctly (i.e., the situation doesn't warrant use of gloves and mask), then the dentist will have been exposed, as well as the staff, immediate family and other patients. It is very important that we treat all patients with appropriate aseptic procedures.

Today's dentists feel very strongly that there must be one standard of care. They know that they must accept infection control procedures. They know that they cannot have it both ways.