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Isolation of Candida Spp. in Dental Plaque of ECC Affected Children

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Introduction

The most important fungal pathogen in the oral cavity are yeasts of Candida genus. It is a common yeast living in the oral cavity of about one-half of population. Yeasts are noted for their ability to ferment monosaccharides and some disaccharides to ethanol and carbon dioxide. Yeasts were also found in the dental plaque.

Objectives

The study was carried out to identify yeasts isolated from the dental plaque samples obtained from children affected with ECC.

Material and Methods

Isolated yeast colonies obtained from the dental plaque smears cultivated on MRS agar at 37°C in 5% CO2 atmosphere (N = 58) were characterized. To quantify yeasts, further samples of dental plaque (N = 30) were smeared using sterile cotton swabs. The swabs were rinsed in 1ml of saline, 100µl of the solution were inoculated on Sabouraud dextrose agar and cultivated at 30°C. The number of colony-forming units was calculated and assessed as strong positive (+++), positive (++) and weakly positive (+). The species identification was established using morphological characteristics, characteristic growth on CHROMagar Candida, assimilation and fermentation tests (kits Auxalor 2 and ID 32C). Selected samples of cultivated colonies (N = 10) were investigated in a scanning (SEM) electron microscope. Extracted primary teeth covered by dental plaque were investigated under SEM (N = 20) and a transmission (TEM) electron microscope (N = 10).

Results

Among isolates cultivated originally on MRS agar 58 yeast strains were identified as Candida albicans (51 strains), Candida tropicalis (4 strains), Candida dubliniensis (2 strains) and Rhodotorula rubra (1 strain). In 30 samples evaluated quantitatively Caldida albicans was found to be strongly positive in 5 samples, positive in 7 samples and 2 samples were weakly positive as well as 1 sample of Candida sp. and 1 sample of Candida guilliermondii. 14 samples were found to be negative.
SEM and TEM investigations confirmed the presence of Candida albicans, both hyphal and budding forms were revealed.

Table 1: Isolates cultivated on MRS agar (N=58)

Type of the isolate	**Number of strains**
C. albicans	51
C. tropicalis	4
Rodotorula rubra	1
C. dubliniensis	2

Table 2: Quantitative detection and identification of Candida strains (N=30) from ECC affected children

Type of the isolate	**Quantity**	**Number of samples**
C. albicans	+++	5
	++	7
	+	2
Candida sp.	+	1
C. guilliermondii	+	1

Negative samples	0	14

+ = weakly positive ++ = positive +++ = strongly positive
Negative results were obtained in control samples from caries-free children (N=3)

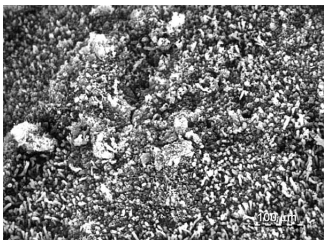

Fig. 1a. Large area of the tooth surface covered by dental plaque of different morphology.

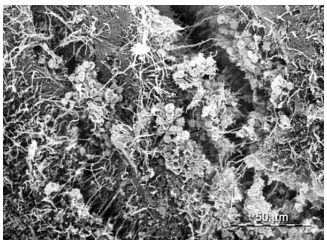

Fig. 1b. Higher magnification of the area revealed a yeast-like microflora

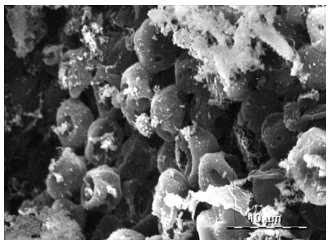

Fig. 1c Higher magnification demonstrated the dental plaque composed of Candida albicans

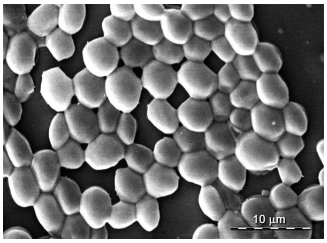

Fig. 2. Cultivated colony of Candida albicans from dental plaque smears

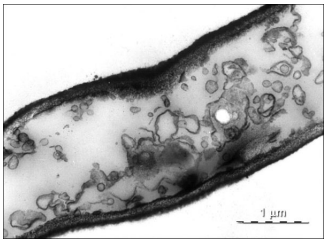

Fig. 3. Hyphal form of Candida albicans from dental plaque

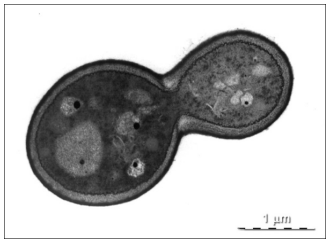

Fig. 4. Budding form of Candida albicans from dental plaque

Conclusions

Candida albicans and further yeast strains are constant components of dental plaque in ECC affected children and they can contribute by their carbohydrate fermenting ability to the destructive course of the disease.

The study was supported by Project 1M0528 from the Czech Ministry of Education.

Literature

1. Marsh P., Martin M.V.: Oral Microbiology, Wright, 2001, p. 153-162
2. Samaranayake L.P.: Essential Microbiology for Dentistry, Churchill Livingstone, 2002, p. 142-147, 239-250

Abbreviations

ECC = early childhood caries
TEM = transmission electron microscope
SEM = scanning electron microscope

This Poster was submitted by Prof. MUDr. Martina Kukletová.

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Poster Faksimile:

Isolation of Candida Spp. in Dental Plaque of ECC Affected Children

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Introduction

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Aim

The study was carried out to identify yeasts isolated from the dental plaque samples obtained from children affected with ECC.

Material and Methods

Isolated yeast colonies obtained from the dental plaque smears cultivated on MRS agar at 37 °C in 5% CO_2 atmosphere (N=58) were characterized. To quantify yeasts, further samples of dental plaque (N=36) were smeared using sterile cotton swabs. The swabs were rinsed in 1 ml of saline, 100 µl of the solution were inoculated on Sabouraud dextrose agar and cultivated at 30 °C. The number of colony-forming units was calculated and assessed as strongly positive (+++), positive (++), and weakly positive (+). The species identification was established using morphological characteristics, characteristic growth on CHROMagar Candida, assimilation and fermentation tests (kits Auxalor 2 and ID 32C). Selected samples of cultivated colonies (N=10) were investigated in a scanning (SEM) electron microscope. Extracted primary teeth covered by dental plaque were investigated under SEM (N=20) and a transmission (TEM) electron microscope (N=10).

Results

Among isolates cultivated originally on MRS agar 58 yeast strains were identified (Table 1) as *C. albicans* (51 strains), *C. tropicalis* (4 strains) *Rhodotorula rubra* (1 strain) and *C. dubliniensis* (2 strains). In 36 samples evaluated quantitatively (Table 2) *C. albicans* was found to be strongly positive in 5 samples, positive in 7 samples, and 2 samples were weakly positive as well as 1 sample of *Candida* sp. and 1 sample of *C. guilliermondii*.

SEM and TEM investigations confirmed the presence of *C. albicans*, both hyphal and budding forms were revealed.

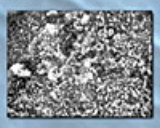
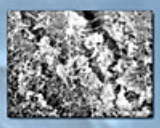


Fig. 1. SEM

a. Large area of the tooth surface covered by dental plaque of different morphology



b. Higher magnification of the area revealed a yeast like microflora

c. Higher magnification demonstrated the dental plaque composed of *Candida albicans*

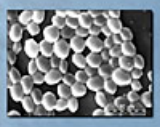


Fig. 2. SEM

Cultivated colony of *Candida albicans* from dental plaque smears

Fig. 3. TEM

Hyphal form of *Candida albicans* from dental plaque

Fig. 4. TEM

Budding form of *Candida albicans* from dental plaque

Table 1. Isolates cultivated on MRS agar (N=58)

Type of the isolate	Number of strains
C.albicans	51
C.tropicalis	4
Rodotorula rubra	1
C.dubliniensis	2

Table 2. Quantitative detection and identification of *Candida* strains (N=36) from ECC affected children

Type of the isolate	Quantity	Numer of samples
C. albicans	+++	5
	++	7
	+	2
Candida sp.	+	1
C. guilliermondii	+	1
Negative samples	0	14

\+ = weakly positive
++ = positive
+++ = strongly positive

Negative results were obtained in control samples from caries-free children (N=3)

Conclusion

Candida albicans and further yeast strains are constant components of dental plaque in ECC affected children, and they can contribute by their carbohydrate fermenting ability to the destructive course of the disease.

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