



Introduction

Although many study the human oral cavity for bacteria, different species of yeast that are also present are often overlooked. Therefore, the different strains of yeasts found in the mouth have not yet been identified. This study focuses on developing a PCR-based technique to identify species of yeast in the mouth. The relationship between the species of yeast found in patients with periodontal disease versus healthy patients was also analyzed.

Objectives

To test the method of isolating and identifying yeast species. To determine the pattern of yeast species in healthy versus periodontal patients.

Methods

Samples of plaque from four different sites in the mouth were collected from ten patients with periodontal disease and ten patients with healthy periodontium. Plaques samples were resuspended in water and boiled to release genomic DNA. A two-step PCR method was used to amplify the internal transcribed spacer

Primer name	Forward	Reverse	Amplicon size	PCR
<u>NESTED PCR</u>				
NSA3f/NLC2r	AAACTCTGTCGTGCTGGGGATA	GAGCT GCATT CCCAAACAACT C	~1100 bp	1 st
NSI1f/NLB4r	GATT GAATG GCTTAGT GAGG	GGATTCT CACCCTCTATGAC	~ 910 bp	2 ND

The PCR product was then separated via agarose gel electrophoresis. Amplicons with correct base pair lengths were gelpurified and sequenced. Specific species of yeasts were identified using the Basic Local Alignment Search Tool from The National Center for Biotechnology Information by comparing the sequenced DNA with a national database. The relationships between the yeast species found in healthy patients and periodontal t'ante una data materia a

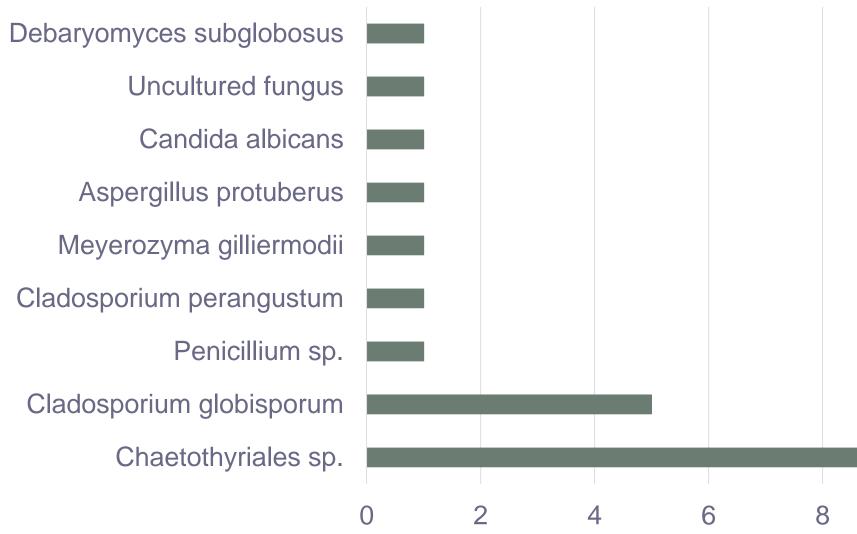
Species of Yeast Identified in the Plaque of **Healthy Patients versus Periodontal Patients** Michelle Fong¹, Der Thor², Tamer Alpagot³, Nan Xiao²

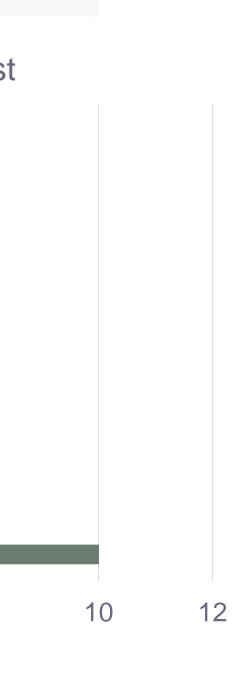
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Results: Healthy Patients

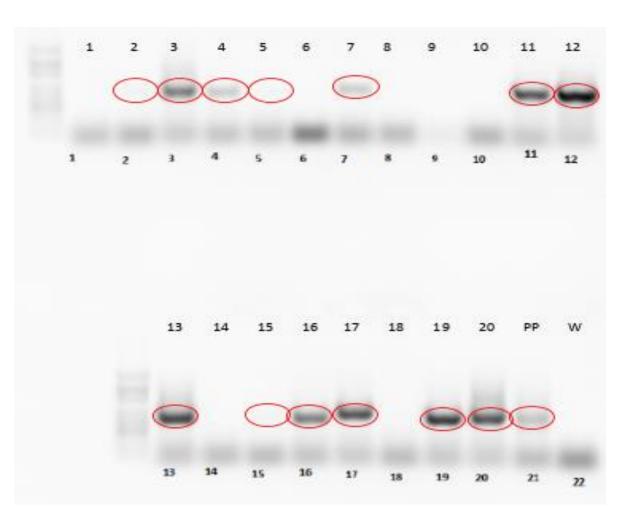
8 9 10 11 12 13 0 14 15 16 17 18 19 20 PP W \circ

Number of Sites Found with Specific Yeast





Results: Periodontal Patients



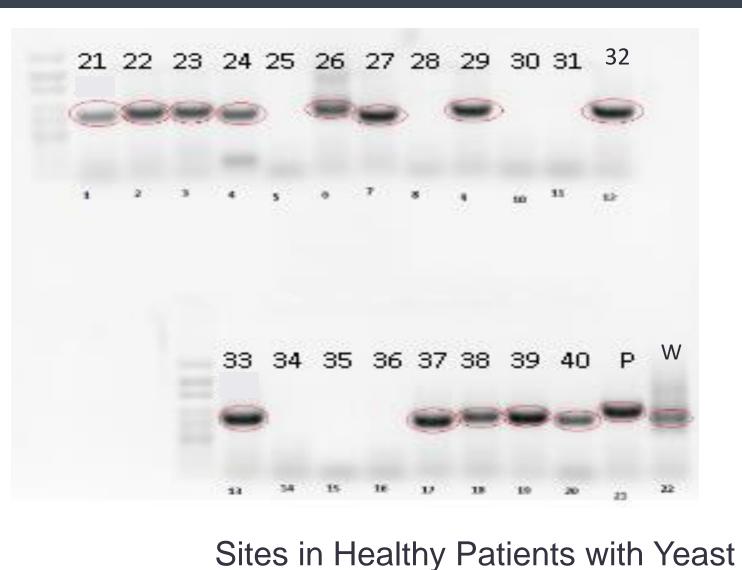
Species of Yeasts Found at Healthy and Perio Sites

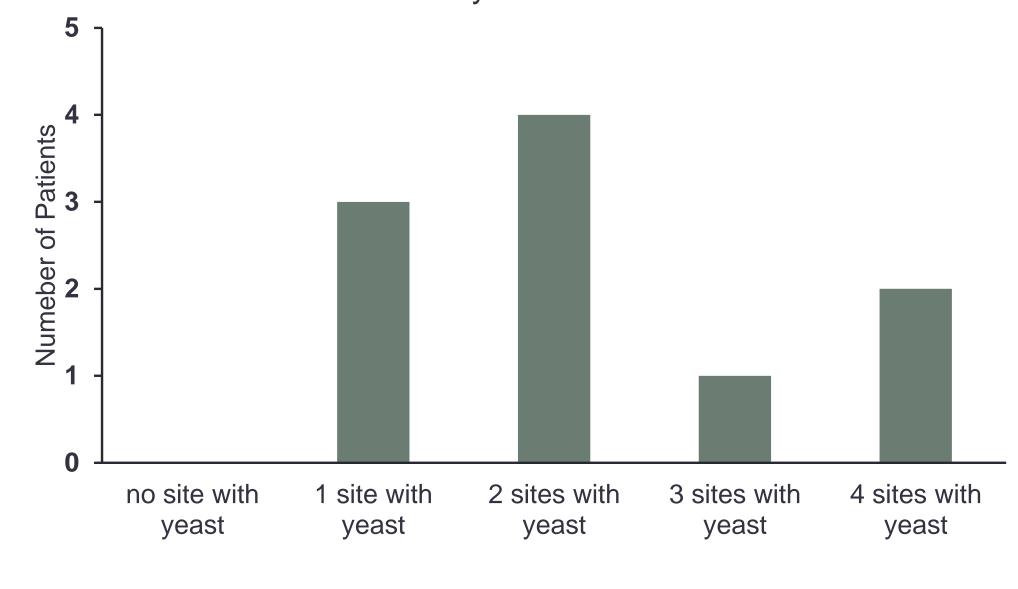


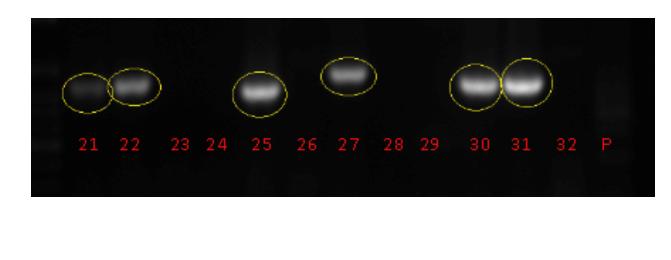
Cladosporium globisporum Penicillium chloroleucon Cladosporium cladosporioides Kabatiella microsticta Meyerozyma guilliermodii Candida albicans Exophiala jeanselmii Aureobasidium pullulans Filobasidium chernovii Uncultured fungus clone J228 Uncultured fungus clone CMH166

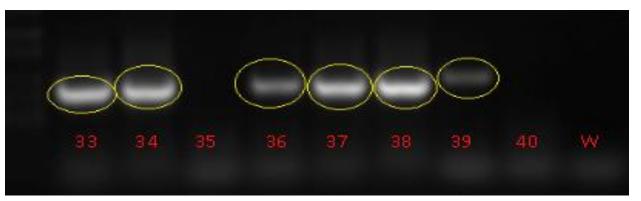
Acknowledgements:

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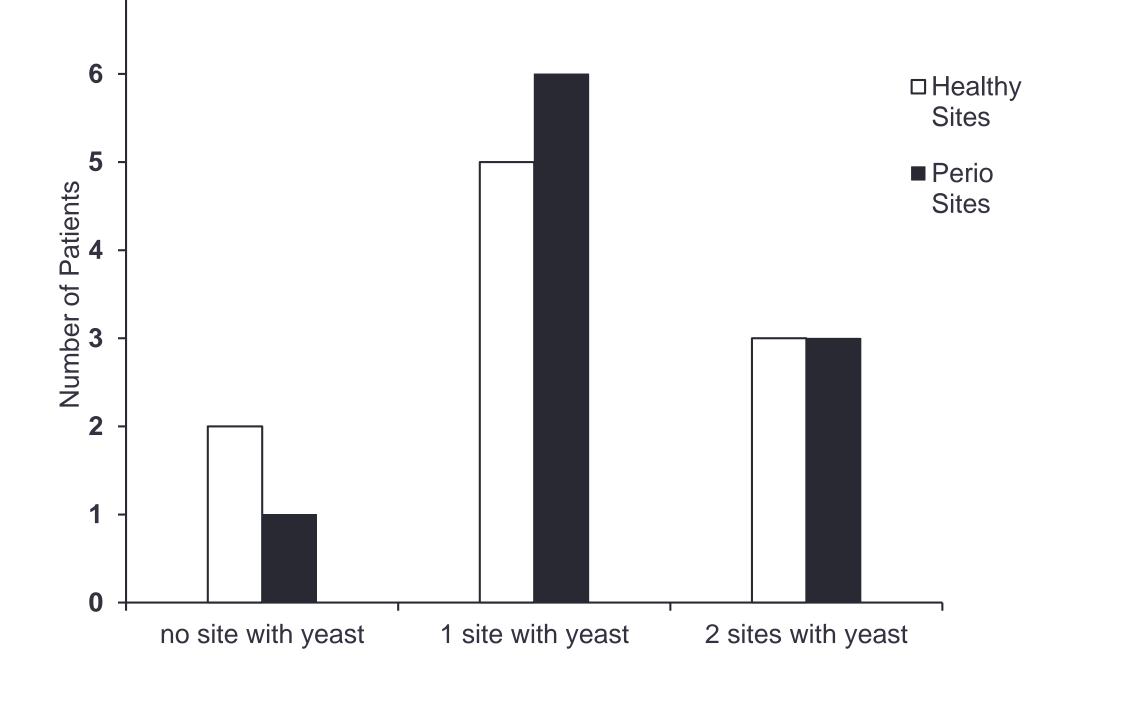
















Results

Many species of yeast were identified in the plaque of healthy patients and patients with periodontal disease. The major yeast species found in the plaque of individuals with a healthy periodontium was Chaetothyriales sp. and Cladosporium globisporum. The major yeast found in the plaque from healthy sites of individuals with periodontal disease was Candida albicans, while Penicillium chloroleucon and Cladosporium globisporum were identified in plaque from diseased sites. In perio sites, Chaetothyriales sp. and Cladosporium globisporum do not occur together, unlike in healthy patients.

Conclusions

The developed method was able to identify yeast species found in the plaque. From the results, there appears to be a difference in the species of yeasts found in healthy patients versus periodontal patients. Further studies with a larger patient pool will give more insight on the relationship between different yeast species in healthy and periodontal patients and any possible association with oral health.

References

1. Schoch CL, Seifert KA, Huhndorf S, Robert V, Spouge JL, Levesque CA, Chen W. 2012. Nuclear ribosomal internal transcribed spacer (ITS) region as a universal DNA barcode marker for fungi. Proc Natl Acad Sci USA. 109:6241-6246.

2. Lauer A, Baal JD, Baal JC, Verma M, Chen JM. Detection of *Coccidioides immitis* in Kern County, California, by multiplex. PCR. Mycologia. 2012;104:62-69. 10.3852/11-127.