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Distribution of Yeast Species and Risk Factors of Oral Colonization among the Residents at Nursing Homes in Taiwan

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Abstract

Introduction: *Candida* species are part of commensal microflora and etiological agents of different infections in humans. Fungal infections contribute to approximately one million deaths annually.

Objective: The present study is to determine the distribution of yeast species and the risk factors of yeast colonization in the oral cavities of residents at 10 nursing homes in Taiwan.

Methods: Oral rinses of residents were collected. All yeast isolates were subjected to the matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF MS) for species identification. The sequences of the internal transcribed spacer (ITS) region and/or the D1/D2 region of ribosomal DNA were used for assessing species identification.

Results: Among 158 of 204 residents colonized by yeasts, 52 were colonized by *Candida albicans* alone and 106 were colonized by at least one non-*C. albicans* yeast species. Interestingly, 88 of the 158 subjects were colonized by more than one species. Among the 320 isolates, comprising 12 fungal genera and 33 species, *C. albicans* (40.3%) was the dominant species, followed by *C. glabrata* (15.9%), *C. parapsilosis* (10.6%), *C. tropicalis* (6.9%), and *Trichosporon asahii* (5.6%). Age, chewing betel nuts, having dentures, periodontal disease, or other chronic diseases are expected risk factors for being colonized by yeast. Interestingly, we also identified two less expected risk factors: being female and brushing teeth more than once a day. Furthermore, residents colonized by > 10 colony-forming unit (CFU) of yeast (OR, 6.156; 95% CI 2.709–13.989; $p = 0.001$) or age > 70 years (OR, 2.435; 95% CI 1.057–5.607; $p = 0.037$) were independent factors associated with multi-species colonization.

Introduction

There are approximately one billion dermatological cases annually in the world. Fungal infections contribute to approximately one million deaths annually [1]. Due to the increased size of populations at risks — such as people receiving chemotherapy, those infected with human immunodeficiency virus (HIV), and the elderly — the prevalence of invasive fungal infections has increased significantly [2-4]. *Candida* species are the most frequently isolated fungal pathogens causing morbidity and mortality in seriously immunocompromised hosts. The observation that opportunistic *Candida* species exist as part of commensal microflora in humans and are usually the etiological agents causing infections [5,6] supports that colonization with *Candida* species is an independent risk factor for invasive candidiasis [7]. Hence, investigating yeast colonization among those high-risk populations may help us understand the basal line of species distribution and identify the risk factors associated with yeast colonization and multiple species colonization.

A previous study found that chronic diseases, medication, poor oral hygiene, reduced salivary flow, and/or the impairment of the immune system are risk factors for Candida infections among the elderly [8]. The Taiwan Surveillance of Antimicrobial Resistance of Yeasts (TSARY), a national survey in Taiwan, found that with increasing age there was a significant increase in the percentage of candiduria [9]. *Candida* species can be detected as commensal microorganisms in the oral cavity of 20–50% of the population, increasing with age [10]. Furthermore, another study in Finland reported that as many as 75% of elderly people were colonized by yeast in the oral cavity [11]. In addition, the frequency of 18.9%–29.2% of the colonization/infection cases with more than one species increasing with age [12].

It has been reported that residents in the nursing home have a high degree of dental/denture plaque, which is a reservoir of pathogenic microbes [13]. The rate of detection of *Candida albicans* (66.7%) in gargled samples from elderly patients was similar to that of *Streptococcus pneumoniae* (63%) but higher than those of methicillin-resistant *Staphylococcus aureus* (14.8%), and *Pseudomonas aeruginosa* (5.6%) [14]. Besides, they also found that the densities of *C. albicans* cells in the elderly were significantly higher than those in the healthy young group [14]. In Taiwan, 49 of 323 (15.2%) examined students and staff at the National Health Research Institutes were colonized by low-density yeasts [15]. In addition, approximately 50% (45–59%) of outpatients in Taiwan with human immunodeficiency virus (HIV) were colonized by yeasts [16-19]. However, oral yeast colonization among the elderly in Taiwan has not been previously investigated. Thus, in the present study, we determine the distribution of yeast species and the risk factors for yeast colonization among residents at nursing homes in Taiwan.

Methods

Study and data collection

The study titled "Elderly Care: Oral Yeast Colonization and Tinea Capitis in Non-hospitalized Elder Population" was approved by the Research Ethics Committee of the National Health Research Institutes (EC1040411-E). A total of 239 residents at 10 nursing homes in central Taiwan were enrolled after informed consents were obtained. We did not screen for the oral health of the residents before enrollment. Residents younger than 50 years or those who failed to provide a sample of oral rinse were excluded. A total of 204 residents were included in the present study. At the time of collection, an oral pathologist examined the oral health of all individuals. A standardized data-collection form was used to retrieve demographic characteristics (e.g., age, gender, height, weight, and education), underlying medical conditions (e.g., diabetes mellitus, heart disease, and hypertension), and information related to dental care.

Sample collection and fungal cultures

The sample collection was according to the previous report [20]. Approximately 20 ml of oral rinse containing saline was obtained from each resident from September to October 2105. All rinses were kept in 50 ml centrifuge tubes at room temperature and transported to the central laboratory in a research institute within 24 h. The rinses were then centrifuged and re-suspended in 1 ml of 0.85% NaCl. A total of 0.05 ml of a suspension of each sample was streaked onto Chromagar Candida media (CHROMagar, Paris, France) [21]. All medium plates were incubated at 35°C for 2 days. If present, colonies from each medium plate were selected for further analyses. Additional colonies were selected from the medium plates when more than one morphotype was present. One isolate of each species from each resident was analyzed.

Identification

All isolates were subjected to MALDI-TOF MS for species identification using MALDI Biotyper software version 3.1. When isolate identifications were inconsistent with the color on CHROMagar Candida agar medium or when uncommon species were reported, the sequences of the internal transcribed spacer (ITS) region and/or the D1/D2 region of ribosomal DNA

were used for species identification. The ITS regions were amplified by the primers ITS1, 5'-TCCGTAGGTGAACCTGCGG-3, and ITS4 5'-TCCTCCGCTTATTGATATGC-3'; and the D1/D2 regions were amplified by the primers NL1 5'-GCATATCAATAAGCGGAGGAAAAG-3' and NL4 5'-GGTCCGTGTTTCAAGACGG-3' [22]. There were 6 isolates identified as *Trichosporon mucoides* by MALDI-TOF MS. However, we could not distinguish *T. mucoides* from *Trichosporon dermatis*, and the database of MALDI-TOF MS does not contain *T. dermatis*. Thus, we named them as *T. mucoides/dermatis* in the present study. In addition, we failed to identify two isolates; one was named as belonging to the *Trichosporon* species and the other as ND, standing for not determined.

Statistical methods

The results were analyzed with SPSS software for Windows, version 12.0. Items on the data-collection form were tested for association with frequency (incidence) yeast colonization and multiplicity of species. The factors including age, gender, body mass index (BMI), education, wheelchair use, smoking, betel nut chewing, number of missing teeth, number of denture, frequency of brushing teeth, mouthwash use, having dry mouth, having periodontal disease and/or tartar, having chronic disease (hypertension, diabetes mellitus, heart disease, osteoporosis, stroke, gout, arthritis, kidney disease), and taking medicine were assessed. The Chi-squared test was applied for categorical variables, and Student's t-test was used for continuous variables. Logistic regression was applied to assess the independent effects of factors with values less than 0.05 in univariate analysis. A *p*-value less than 0.05 was considered significant.

Results

Study population

During the study period, 204 residents were enrolled in the present study. The average number of participants per nursing home was 20, ranging from 9 to 48. Their demographic data are shown in (Table 1). The average age was 77.1 years, ranging from 50 to 95, with 153 (75%) older than 70 years. They were 106 females (52%). The sample population had an average of 10 dentures each. A total of 163 (79.9%) had chronic diseases, and 57 (27.9%) needed wheelchairs.

Species distribution

A total of 320 isolates comprising 12 fungal genera and 33 species were identified. There were 265 Candida; 27 Trichosporon; 7 Saccharomyces; 5 each of Exophiala and Magnusiomyces; 2 each of Fereydounia, Lachancea, and Pichia; and one each of Cryptococcus, Lodderomyces, Rhodosporidium, Rhodotorula, and undetermined one. The colonization species were *C. albicans* (129, 40.3%), *Candida glabrata* (51, 15.9%), *Candida parapsilosis* (34, 10.6%), *Candida tropicalis* (22, 6.9%), *Trichosporon asahii* (18, 5.6%), *Saccharomyces cerevisiae* (7, 2.2%), *Candida pararugosa* (6, 1.9%), *T. mucoides/dermatis* (6, 1.9%), *Exophiala dermatitidis* (5, 1.6%), *Magnusiomyces capitatus* (5, 1.6%), *Candida guilliermondii* (4, 1.3%), *Candida krusei* (4, 1.3%), *Candida orthopsilosis* (3, 0.9%), and others (26, 8.1%).

Of the 158 residents colonized by yeasts, 70, 41, 29, 11, 5, and 2 were colonized by 1, 2, 3, 4, 5, and 6 species, respectively (Figure 1, Table S1 and Table S2). Overall, *Candida* species accounted for 82.8% of these isolates. Furthermore, 106 were colonized by at least one non-*C. albicans* yeast species. Among the 70 residents colonized by a single species, 52 (74.3%) were colonized by *C. albicans*, followed by 5 *C. parapsilosis*, 4 *T. asahii*, 3 *C. glabrata*, and one each of *C. guilliermondii*, *C. pararugosa*, *C. tropicalis*, *Candida utilis*, *Trichosporon faecale*, and *Trichosporon* sp. Some species were more prevalent than others. Among the 5 residents colonized by 5 species, 3 were colonized by species composition C (*C. albicans*/*C. glabrata*/*C. parapsilosis*/2 others). Among the 29 residents colonized by 3 species, 11, 8, and 5 were colonized by species composition N (*C. albicans*/*C. glabrata*/*C. tropicalis*), P (*C. albicans*/*C. glabrata*/other), and Q (*C. albicans*/*C. parapsilosis*/other), respectively. Among the 41 residents colonized by 2 species, 17, 14, and 5 were colonized by species composition V (*C. albicans*/*C. glabrata*), W (*C. albicans*/other) and X (*C. albicans*/*C. parapsilosis*), respectively. All four residents colonized by *C. krusei* were also co-colonized by *C. albicans*, *C. albicans*/*C. glabrata*, *C. albicans*/*C. glabrata*/*C. tropicalis*, and *C. albicans*/*C. parapsilosis*/*Candida pararugosa*/*C. tropicalis*.

Status and risk factors for yeast colonization

Risk factors for yeast colonization were determined (Table 2). Having more dentures ($p < 0.001$) or being older ($p < 0.001$) were risks for being colonized by yeast. Approximately 80% of residents colonized by yeast were older than 70 years ($p = 0.033$). Those who brushed their teeth more than once a day ($p = 0.015$), had a chronic disease ($p = 0.027$), or were female ($p = 0.048$) were more likely to be colonized by yeasts. According to multivariate analysis, having more dentures (OR, 1.052; 95% CI 1.008–1.099; p=0.021) is a risk for being colonized by yeast. Furthermore, residents having yeast colony-forming unit (CFU) >10 (OR, 6.156; 95% CI 2.709–13.989; $p = 0.001$) or age > 70 years (OR, 2.435; 95% CI 1.057–5.607; $p = 0.037$) were more likely to be colonized by multiple species.

Table 1: Risk factors of residents colonized by yeasts

Characteristics	Total (N = 204)		Colonized by Yeast (N = 158)		Not-colonized by Yeast (N = 46)		Univariate	Multivariate
	mean	SD	mean	SD	mean	SD	*p* value	*p* value, OR (95% CI)
Age, years (mean ± SD)	77.1	9.73	78.4	9.17	72.7	10.37	**<0.001**	
BMI	22.7	3.58	22.6	3.63	23.0	3.43	0.451	
Number of missing teeth	15.8	10.13	16.5	10.30	13.3	9.21	0.06	
Number of dentures	10.8	10.97	12.1	11.28	6.2	8.42	**<0.001**	**0.008, 1.06 (1.015-1.107)**
	No.	%	No.	%	No.	%	p value	
Female	106	52	88	55.7	18	39.1	**0.048**	0.974, 0.987 (0.447-2.178)
Age > 70 years	153	75	124	78.5	29	63	**0.033**	0.949, 1.027 (0.454-2.324)
Education							0.815	
None	27	13.2	22	13.9	5	10.9		
Elementary School	99	48.5	74	46.8	25	54.3		
Junior High School	32	15.7	26	16.5	6	13		
High School	28	13.7	23	14.6	5	10.9		
College or above	18	8.8	13	8.2	5	10.9		
Wheelchair use	57	27.9	42	26.6	15	32.6	0.423	
Smoking	18	8.8	15	9.5	3	6.5	0.905	
Betel nut chewing	6	2.9	2	1.3	4	8.7	**0.001**	0.081, 0.183 (0.027-1.236)
Frequency of brushing teeth > once a day	112	54.9	94	59.5	18	39.1	**0.015**	0.257, 1.55 (0.726-3.309)
Mouthwash	51	25	39	24.7	12	26.1	0.847	
Dry mouth	75	36.8	61	38.6	14	30.4	0.312	
Periodontal disease and/or Tartar	127	62.3	92	58.2	35	76.1	**0.028**	0.238, 0.598 (0.254-1.406)
Chronic disease	163	79.9	132	83.5	31	67.4	**0.027**	0.078, 2.155 (0.916-5.065)
Hypertension	91	44.6	72	45.6	19	41.3	0.609	
Diabetes mellitus	46	22.5	40	25.3	6	13	0.08	
Heart disease	35	17.2	28	17.7	7	15.2	0.692	
Osteoporosis	14	6.9	11	7	3	6.5	1	
Stroke	12	5.9	9	5.7	3	6.5	0.735	
Gout	10	4.9	8	5.1	2	4.3	1	
Arthritis	10	4.9	10	6.3	0	0	0.121	
Kidney disease	4	2	3	1.9	1	2.2	1	
Taking medicine	143	70.1	115	72.8	28	60.9	0.245	
No, number.								

Figure 1: Distribution of species composition of residents colonized by multiple species.

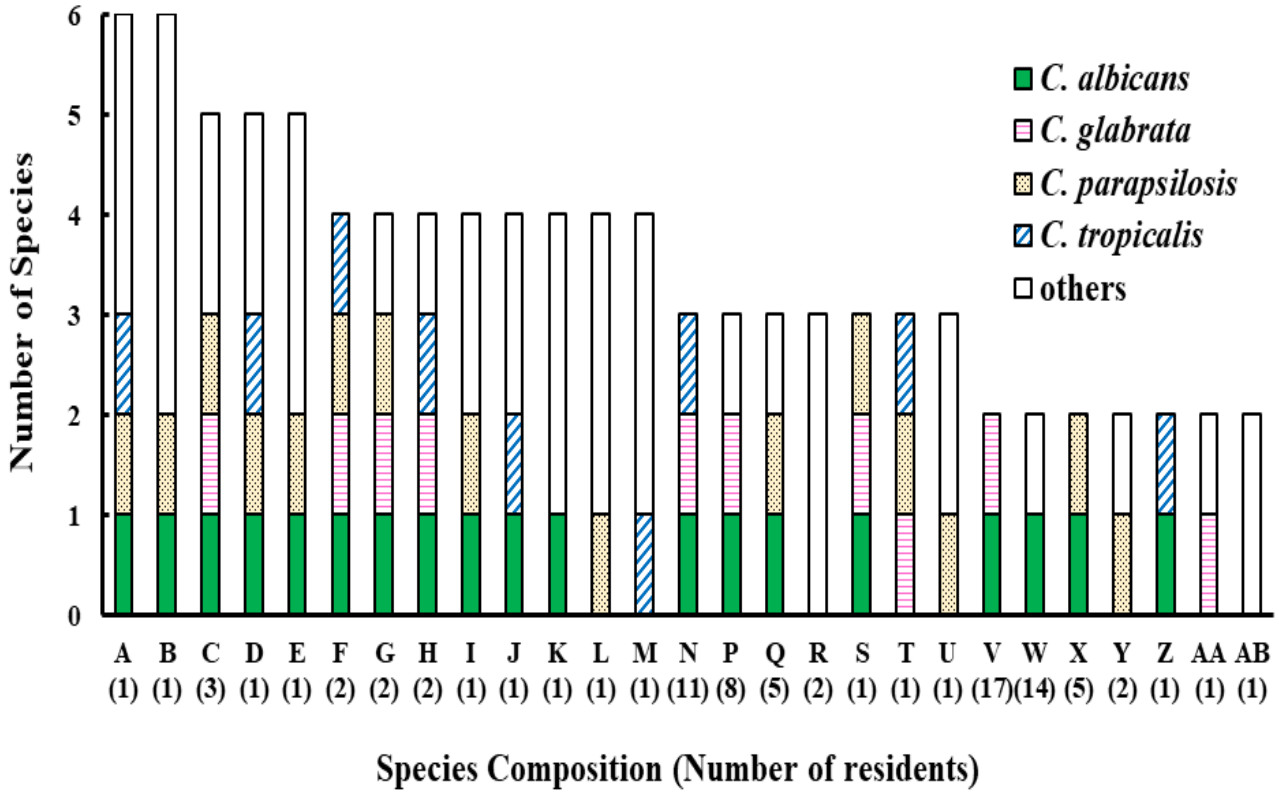

Table 2: Risk factors of residents colonized by multiple species

Characteristics	Total (N=158)		More than one species (N=88)		One species (N=70)		Uni-variate	Multivariate
	mean	SD	mean	SD	mean	SD	*p* value	*p* value, OR (95% CI)
Age	78.4	9.17	79.8	8.75	76.7	9.44	**0.032**	
BMI	22.6	3.63	22.5	3.76	22.7	3.48	0.793	
Number of missing teeth	16.5	10.30	16.4	10.35	16.7	10.31	0.847	
Number of dentures	12.1	11.28	12.7	11.31	11.5	11.29	0.504	
	No.	%	No.	%	No.	%	p value	
Yeast CFU > 10	117	74.1	78	88.6	39	55.7	**<0.001**	**0.001, 6.156 (2.709–13.989)**
Female	88	55.7	52	59.1	36	51.4	0.335	
Age > 70	124	78.5	75	85.2	49	70	**0.021**	**0.037, 2,435 (1.057–5.607)**
Education							0.675	
None	22	13.9	14	15.9	8	11.4		
Elementary school	74	46.8	41	46.6	33	47.1		
Junior high school	26	16.5	16	18.2	10	14.3		
High school	23	14.6	10	11.4	13	18.6		
College or above	13	8.2	7	8	6	8.6		
Wheelchair use	42	26.6	25	28.4	17	24.3	0.56	
Smoking	15	9.5	8	9.1	7	10	1	
Betel nut chewing	2	1.3	1	1.1	1	1.4	1	

Frequency of brushing teeth > once a day	94	59.5	48	54.5	46	65.7	0.155	
Mouthwash	39	24.7	21	23.9	18	25.7	0.789	
Dry mouth	61	38.6	35	39.8	26	37.1	0.736	
Periodontal Disease and/or tartar	92	58.2	51	58	41	58.6	0.938	
Chronic disease	132	83.5	76	86.4	56	80	0.273	
Hypertension	72	45.6	41	46.6	31	44.3	0.773	
Diabetes mellitus	40	25.3	22	25	18	25.7	0.918	
Heart disease	28	17.7	17	19.3	11	15.7	0.556	
Osteoporosis	11	7	8	9.1	3	4.3	0.348	
Arthritis	10	6.3	4	4.5	6	8.6	0.34	
Stroke	9	5.7	5	5.7	4	5.7	1	
Gout	8	5.1	4	4.5	4	5.7	0.733	
Kidney disease	3	1.9	1	1.1	2	2.9	0.585	
Taking medicine	115	72.8	68	77.3	47	67.1	0.25	
No, number.								

Discussion

We used a traditional epidemiological approach to establish a basal line of yeast colonization among residents at nursing homes in Taiwan. In the present study, we found that up to 77.5% of residents at nursing homes in central Taiwan were colonized by yeast, a rate similar to the 75% reported in Italy [23] and higher than the 67% reported in France [24]. Consistent with previously reports [8, 25-27], age, chewing betel nuts, having dentures, periodontal disease, other chronic diseases are expected risk factors for being colonized by yeast. Interestingly, we also identified two less expected risk factors: being female and brushing teeth more than once a day. A nationwide study of 36,026 Korean adults found that women brushed their teeth more often than men did, but men retained more natural teeth than women [28]. There is no doubt that brushing teeth is important for maintaining oral health. However, improper oral care can also introduce harmful effects [29]. In addition, aggressive and/or incorrect tooth brushing may also lead to dental damage. A long-term oral healthcare program increased the proportion of elderly residents in a nursing home having an acceptable score from 36% to 70% [30]. Hence, whether being female and brushing teeth more than once a day are still risks for being colonized by yeast after proper oral care needs further investigation.

Of the 33 species recovered in the present study, only 4 species (*Candida ethanolica*, *Pichia manshurica*, *Rhodosporidium toruloides*, and *Trichosporon jirovecii*) have not been reported to cause diseases in humans. There were six residents colonized by *C. pararugosa*, which was first reported in 2017 to cause bloodstream infections [31]. Hence, it is worth noting that in addition to those common species that cause diseases in humans, the remaining rare species recovered from the present study may also be potential pathogens causing diseases.

Among 126 oral Candida isolates from patients suffering from caries and chronic periodontitis, 75 were *C. albicans*, 18 were *C. parapsilosis*, 11 were *C. dubliniensis*, 6 were *C. guilliermondii*, 5 were *C. lipolytica*, 5 were *C. glabrata*, 4 were *C.tropicalis* and 2 were *C.krusei* [32]. In Japan, *Candida* species in tongue dorsa were detected in 162 of 266 participants older than 60 years old. *Candida albicans, C. glabrata*, *C. tropicalis*, and *C. parapsilosis* were detected in 142, 60, 5, and 2 participants, respectively. No *C. krusei* was detected in that survey [26]. Interestingly, the species distribution in this study is different from those in Taiwan. Oral yeast colonization of 323 healthy individuals with a mean age of 33.9 years (range 18–60) was studied previously. Among the 52 isolates, comprising 13 species, 57.7% were *C. albicans*, 15.4% were *C. parapsilosis*, and 3.9% were *C. glabrata*. No *C. tropicalis* was detected in that study.[15]. In the TSARY in 2014, 34.9%, 27.5%, 25.8%, 5.1%, and 1% of 1139 isolates from patients were *C. albicans*, *C. glabrata*, *C. tropicalis*, *C. parapsilosis*, and *C. krusei*, respectively. The observation that species distribution among the residents at nursing homes in the present study is more similar to that of patients than that of the healthy young population may be due to the high proportion (79.9%) of residents at nursing homes with chronic diseases.

In the present study, we found that among the 158 residents colonized by yeasts, 55.7% were colonized by more than one species. It is worth noting that 24 residents (Figure 1, species combination A, C, D, F, G, H, N, S, and T) were co-colonized by at least three of the four most common *Candida* species causing diseases in humans (*C. albicans*, *C. glabrata*, *C. parapsilosis*, and *C. tropicalis*). Previously, we detected HIV-infected patients were also colonized by multiple yeast species [16-19]. Of the 49 health population colonized by yeasts, 3 (6.1%) were co-colonized by more than one species [15]. Furthermore, more than one species were detected from specimens from 36 of 1116 (3.4%) patients in TSARY in 2010 and 44 of 1092 (4.0%) patients in TSARY in 2014 [33,34]. Consistent with our findings, of the 162 elderly colonized by *Candida* species in Japan, 27.2% were colonized by more than one species. A combination of *C. albicans* and *C. glabrata* was the most prevalent one [26]. In Spain, there were also 9 of 132 (7%) patients colonized by multiple species [35]. In addition, in a large survey, 498 of 5671 (8.8%) culture-positive samples contained more than one species. There were 476 colonized by two species, 21 by three species, and one by four species. The most common combination was *C. albicans*/*C. glabrata* and *C. albicans*/*C. krusei*, representing approximately 70% of all cases [36]. Approximately 11% of patients in long-term geriatric care were colonized/infected by a combination of *C. albicans*/*C. glabrata* [24].

The fact that different *Candida* species have different susceptibilities to antifungal drugs illustrates the potential for personalizing candidemia treatment [37]. For example, fluconazole, one of the most commonly prescribed antifungal drugs, is not suggested to treat infections caused by *C. krusei* or *C. glabrata* [38]. Furthermore, the increasing number of reports of multiple species colonization points to the importance that health care personals be aware that fungal infections, especially invasive infections, may be caused by multiple species with different susceptibilities to antifungal drugs.

The population of the elderly is increasing significantly worldwide. We found that having more dentures or dry mouth are two major risk factors associated with multiple species of yeast colonization. Approximately 60% of residents had periodontal disease and/or tartar at the time of sampling. A study in Japan found that the incidence of fever of the elderly at the nursing home was associated with the oral Candida levels. The authors proposed that oral levels of Candida can be an indicator of oral hygiene concerns [39]. Thus, we encouraged those participants to seek help from dentists. Hence, it would be interesting and important to repeat sampling among the same population to assess the effect of providing information on proper oral care.

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Declaration of interest

Dr. Wen-Chi Cheng works in the R&D department of General Biologicals Corp.

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Table S1. Distribution of yeast species

Participant	No. of species	*C. albicans*	*C. glabrata*	*C. parapsilosis*	*C. tropicalis*	*T. asahii*	*S. cerevisiae*	*C. pararugosa*	Others
TCH-029	6	1	0	1	1	0	0	1	*C. krusei*, *Lodderomyces elongisporus*
GLC-019	6	1	0	1	0	1	0	1	*C. orthopsilosis*, *Trichosporon mucoides/dermatis*
GLC-011	5	1	1	1	0	0	0	0	*C. intermedia*, *C. lusitaniae*
GLC-005	5	1	1	1	0	0	0	0	*Pichia manshurica*, *Trichosporon mucoides/dermatis*
TCH-009	5	1	1	1	0	0	0	1	*Trichosporon sp.*
TCH-001	5	1	0	1	1	1	0	0	*C. guilliermondii*
FSC-042	5	1	0	1	0	0	1	0	*C. guilliermondii*, *Rhodotorula mucilaginosa*
GLS-007	4	1	1	1	1	0	0	0	
FSC-001	4	1	1	1	1	0	0	0	
TCH-014	4	1	1	1	0	0	0	0	*Lachancea fermentati*
HOP-006	4	1	1	1	0	1	0	0	
YEO-006	4	1	1	0	1	0	0	0	*C. krusei*
FSC-049	4	1	1	0	1	0	1	0	
HWA-012	4	1	0	1	0	0	0	1	*C. guilliermondii*
TCH-011	4	1	0	0	1	0	1	0	*Pichia manshurica*
HWA-008	4	1	0	0	0	1	0	0	*Exophiala dermatitidis*, *Trichosporon mucoides/dermatis*
FSC-038	4	0	0	1	0	0	0	1	*C. guilliermondii*, *C. kefyr*
HOPI-009	4	0	0	0	1	1	0	0	*Fereydounia khargensis*, *Trichosporon mucoides/dermatis*
WES-019	3	1	1	1	0	0	0	0	
WES-018	3	1	1	0	1	0	0	0	
WES-020	3	1	1	0	1	0	0	0	
GLS-008	3	1	1	0	1	0	0	0	
WES-013	3	1	1	0	1	0	0	0	
GLC-013	3	1	1	0	1	0	0	0	
GLC-015	3	1	1	0	1	0	0	0	
FAM-011	3	1	1	0	1	0	0	0	
HWA-002	3	1	1	0	1	0	0	0	
FSC-005	3	1	1	0	1	0	0	0	
FSC-016	3	1	1	0	1	0	0	0	
FSC-025	3	1	1	0	1	0	0	0	
GLC-010	3	1	1	0	0	0	0	0	*C. ethanolica*
FAM-005	3	1	1	0	0	0	0	0	*C. krusei*
GFC-014	3	1	1	0	0	0	0	0	*C. orthopsilosis*
FSC-029	3	1	1	0	0	0	0	0	*C. orthopsilosis*
TCH-037	3	1	1	0	0	0	0	0	*Exophiala dermatitidis*

TCH-010	3	1	1	0	0	0	0	0	*Magnusiomyces capitatus*
HWA-013	3	1	1	0	0	0	0	0	*Magnusiomyces capitatus*
HOP-008	3	1	1	0	0	1	0	0	
FSC-021	3	1	0	1	0	0	0	0	*Cryptococcus neoformans var grubii*
FSC-008	3	1	0	1	0	0	1	0	
GLC-008	3	1	0	1	0	1	0	0	
GFC-017	3	1	0	1	0	1	0	0	
TCH-015	3	1	0	1	0	1	0	0	
YEO-005	3	0	1	1	1	0	0	0	
GLS-002	3	0	0	1	0	1	0	0	*C. guilliermondii*
FSC-040	3	0	0	0	0	0	0	0	*C. metapsilosis*, *Exophiala dermatitidis*, *Rhodosporidium toruloides*
GFC-007	3	0	0	0	0	1	0	0	*Trichosporon jirovecii*, *Trichosporon mucoides/dermatis*
HOP-003	2	1	1	0	0	0	0	0	
HOP-005	2	1	1	0	0	0	0	0	
GLC-003	2	1	1	0	0	0	0	0	
HOP-010	2	1	1	0	0	0	0	0	
TCH-021	2	1	1	0	0	0	0	0	
TCH-024	2	1	1	0	0	0	0	0	
GLC-018	2	1	1	0	0	0	0	0	
FAM-004	2	1	1	0	0	0	0	0	
FAM-009	2	1	1	0	0	0	0	0	
HWA-007	2	1	1	0	0	0	0	0	
HWA-020	2	1	1	0	0	0	0	0	
HWA-035	2	1	1	0	0	0	0	0	
FSC-018	2	1	1	0	0	0	0	0	
FSC-023	2	1	1	0	0	0	0	0	
FSC-026	2	1	1	0	0	0	0	0	
FSC-033	2	1	1	0	0	0	0	0	
FSC-039	2	1	1	0	0	0	0	0	
WES-012	2	1	0	1	0	0	0	0	
GLC-012	2	1	0	1	0	0	0	0	
FAM-010	2	1	0	1	0	0	0	0	
FSC-024	2	1	0	1	0	0	0	0	
FSC-028	2	1	0	1	0	0	0	0	
WES-001	2	1	0	0	1	0	0	0	
FSC-048	2	1	0	0	0	0	0	0	*C. dubliniensis*
FSC-014	2	1	0	0	0	0	0	0	*C. krusei*
YEO-009	2	1	0	0	0	0	0	0	*C. norvegensis*
FSC-019	2	1	0	0	0	0	0	0	*C. norvegensis*
TCH-043	2	1	0	0	0	0	0	0	*Exophiala dermatitidis*
FSC-045	2	1	0	0	0	0	0	0	*Fereydounia khargensis*
YEO-002	2	1	0	0	0	0	0	0	*Magnusiomyces capitatus*

YEO-004	2	1	0	0	0	0	0	0	*Magnusiomyces capitatus*
HWA-017	2	1	0	0	0	0	0	0	*Magnusiomyces capitatus*
FSC-032	2	1	0	0	0	0	0	0	*Trichosporon mucoides/dermatis*
TCH-046	2	1	0	0	0	0	1	0	
FSC-041	2	1	0	0	0	0	1	0	
TCH-007	2	1	0	0	0	1	0	0	
WES-009	2	0	1	0	0	0	1	0	
TCH-031	2	0	0	1	0	0	0	0	*C. fermentati*
WES-025	2	0	0	1	0	1	0	0	
GFC-006	2	0	0	1	0	1	0	0	
TCH-013	2	0	0	0	0	0	0	0	*Lachancea fermentati*, *Exophiala dermatitidis*
WES-017	1	1	0	0	0	0	0	0	
WES-021	1	1	0	0	0	0	0	0	
WES-023	1	1	0	0	0	0	0	0	
WES-038	1	1	0	0	0	0	0	0	
GLC-002	1	1	0	0	0	0	0	0	
GLC-006	1	1	0	0	0	0	0	0	
GLC-007	1	1	0	0	0	0	0	0	
HOP-007	1	1	0	0	0	0	0	0	
GFC-002	1	1	0	0	0	0	0	0	
GFC-012	1	1	0	0	0	0	0	0	
GFC-015	1	1	0	0	0	0	0	0	
GFC-020	1	1	0	0	0	0	0	0	
TCH-019	1	1	0	0	0	0	0	0	
TCH-025	1	1	0	0	0	0	0	0	
TCH-027	1	1	0	0	0	0	0	0	
TCH-033	1	1	0	0	0	0	0	0	
TCH-041	1	1	0	0	0	0	0	0	
TCH-049	1	1	0	0	0	0	0	0	
GLS-003	1	1	0	0	0	0	0	0	
GLS-004	1	1	0	0	0	0	0	0	
GLS-005	1	1	0	0	0	0	0	0	
GLS-009	1	1	0	0	0	0	0	0	
WES-002	1	1	0	0	0	0	0	0	
WES-004	1	1	0	0	0	0	0	0	
WES-005	1	1	0	0	0	0	0	0	
WES-007	1	1	0	0	0	0	0	0	
WES-011	1	1	0	0	0	0	0	0	
GLC-014	1	1	0	0	0	0	0	0	
FAM-001	1	1	0	0	0	0	0	0	
FAM-002	1	1	0	0	0	0	0	0	
FAM-008	1	1	0	0	0	0	0	0	
HWA-001	1	1	0	0	0	0	0	0	

HWA-006	1	1	0	0	0	0	0	0	
HWA-010	1	1	0	0	0	0	0	0	
HWA-021	1	1	0	0	0	0	0	0	
HWA-046	1	1	0	0	0	0	0	0	
FSC-002	1	1	0	0	0	0	0	0	
FSC-003	1	1	0	0	0	0	0	0	
FSC-004	1	1	0	0	0	0	0	0	
FSC-006	1	1	0	0	0	0	0	0	
FSC-007	1	1	0	0	0	0	0	0	
FSC-012	1	1	0	0	0	0	0	0	
FSC-013	1	1	0	0	0	0	0	0	
FSC-015	1	1	0	0	0	0	0	0	
FSC-020	1	1	0	0	0	0	0	0	
FSC-027	1	1	0	0	0	0	0	0	
FSC-030	1	1	0	0	0	0	0	0	
FSC-031	1	1	0	0	0	0	0	0	
FSC-043	1	1	0	0	0	0	0	0	
FSC-044	1	1	0	0	0	0	0	0	
FSC-047	1	1	0	0	0	0	0	0	
HOP-001	1	1	0	0	0	0	0	0	
WES-016	1	0	1	0	0	0	0	0	
GFC-011	1	0	1	0	0	0	0	0	
TCH-016	1	0	1	0	0	0	0	0	
GFC-013	1	0	0	1	0	0	0	0	
TCH-006	1	0	0	1	0	0	0	0	
TCH-018	1	0	0	1	0	0	0	0	
HWA-014	1	0	0	1	0	0	0	0	
FSC-022	1	0	0	1	0	0	0	0	
TCH-002	1	0	0	0	1	0	0	0	
HWA-009	1	0	0	0	0	0	0	0	*C. guilliermondii*
TCH-012	1	0	0	0	0	0	0	0	*C. utilis*
YEO-003	1	0	0	0	0	0	0	0	ND
GLC-017	1	0	0	0	0	0	0	0	*Trichosporon faecale*
HWA-037	1	0	0	0	0	0	0	1	
YEO-007	1	0	0	0	0	1	0	0	
HWA-015	1	0	0	0	0	1	0	0	
FSC-035	1	0	0	0	0	1	0	0	
FSC-046	1	0	0	0	0	1	0	0	

Table S2. Distribution of species causing multiple species colonization

No. species	No. residents	*C. albicans*	*C. glabrata*	*C. parapsilosis*	*C. tropicalis*	*T. asahii*	*S. cerevisiae*	*C. pararugosa*	others
6	1	1	0	1	1	0	0	1	*C. krusei*, *Lodderomyces elongisporus*
6	1	1	0	1	0	1	0	1	*C. orthopsilosis*, *Trichosporon mucoides/dermatis*
5	1	1	1	1	0	0	0	0	*C. intermedia*, *C. lusitaniae*
5	1	1	1	1	0	0	0	0	*Pichia manshurica*, *Trichosporon mucoides/dermatis*
5	1	1	1	1	0	0	0	1	*Trichosporon sp.*
5	1	1	0	1	1	1	0	0	*C. guilliermondii*
5	1	1	0	1	0	0	1	0	*C. guilliermondii*, *Rhodotorula mucilaginosa*
4	2	1	1	1	1	0	0	0	
4	1	1	1	1	0	0	0	0	*Lachancea fermentati*
4	1	1	1	1	0	1	0	0	
4	1	1	1	0	1	0	0	0	*C. krusei*
4	1	1	1	0	1	0	1	0	
4	1	1	0	1	0	0	0	1	*C. guilliermondii*
4	1	1	0	0	1	0	1	0	*Pichia manshurica*
4	1	1	0	0	0	1	0	0	*Exophiala dermatitidis*, *Trichosporon mucoides/dermatis*
4	1	0	0	1	0	0	0	1	*C. guilliermondii*, *C. kefyr*
4	1	0	0	0	1	1	0	0	*Fereydounia khargensis*, *Trichosporon mucoides/dermatis*
3	11	1	1	0	1	0	0	0	
3	3	1	0	1	0	1	0	0	
3	2	1	1	0	0	0	0	0	*C. orthopsilosis*
3	2	1	1	0	0	0	0	0	*Magnusiomyces capitatus*
3	1	1	1	1	0	0	0	0	
3	1	1	1	0	0	0	0	0	*C. ethanolica*
3	1	1	1	0	0	0	0	0	*C. krusei*
3	1	1	1	0	0	0	0	0	*Exophiala dermatitidis*
3	1	1	1	0	0	1	0	0	
3	1	1	0	1	0	0	0	0	*Cryptococcus neoformans var grubii*
3	1	1	0	1	0	0	1	0	
3	1	0	1	1	1	0	0	0	
3	1	0	0	1	0	1	0	0	*C. guilliermondii*
3	1	0	0	0	0	0	0	0	*C. metapsilosis*, *Exophiala dermatitidis*, *Rhodosporidium toruloides*
3	1	0	0	0	0	1	0	0	*Trichosporon jirovecii*, *Trichosporon mucoides/dermatis*

2	17	1	1	0	0	0	0	0	
2	5	1	0	1	0	0	0	0	
2	3	1	0	0	0	0	0	0	*Magnusiomyces capitatus*
2	2	1	0	0	0	0	0	0	*C. norvegensis*
2	2	1	0	0	0	0	1	0	
2	1	1	0	0	1	0	0	0	
2	1	1	0	0	0	0	0	0	*C. dubliniensis*
2	1	1	0	0	0	0	0	0	*C. krusei*
2	1	1	0	0	0	0	0	0	*C. norvegensis*
2	1	1	0	0	0	0	0	0	*Exophiala dermatitidis*
2	1	1	0	0	0	0	0	0	*Fereydounia khargensis*
2	1	1	0	0	0	0	0	0	*Trichosporon mucoides/dermatis*
2	1	1	0	0	0	1	0	0	
2	1	0	1	0	0	0	1	0	
2	1	0	0	1	0	0	0	0	*C. fermentati*
2	1	0	0	1	0	1	0	0	
2	1	0	0	0	0	0	0	0	*Lachancea fermentati, Exophiala dermatitidis*
Total	88								
No, number.									