



Title	Impacts of PAFE on the relative CSH of Candida albicans
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57 Impact of Oral Disease among the Elderly in Hong Kong. A.S. McMILLAN*, E.C.M. LO, M.C.M. WONG, P.F. ALLEN. (Universities of Hong Kong and Newcastle upon Tyne, UK)

The institutionalized elderly often have limited access to dental care and may have untreated oral disorders that can seriously affect their quality of life. We aimed to describe the psychosocial and functional impact of oral conditions in the institutionalized elderly in Hong Kong using the 49 statement Oral Health Impact Profile (OHIP), previously translated and validated by us in Hong Kong Chinese. 285 residents (aged 60-80 years) in homes for the aged completed the Chinese OHIP in an interview with a Cantonese-speaking research assistant. A group of age-matched non-institutionalized elderly attending elderly daytime social centres completed the OHIP in the same way. OHIP sub-scales were computed and summary scores calculated by summing negative impacts across the 49 statements (OHIP-SC). Chi-square tests were used to compare the % of respondents reporting negative impacts. Two sample t-tests were used to compare OHIP sub-scales and summary scores. A general feature in both groups was the low percentage of subjects reporting negative impacts. The % of institutionalized reporting negative impacts for OHIP statements was lower than the non-institutionalized in 11 out of 49 and higher in one ($p < 0.05$). They also had significantly lower scores in three OHIP domains (physical disability, psychological disability & handicap) and the OHIP-SC ($p < 0.05$). The OHIP scores of the elderly in Hong Kong were low, especially in the institutionalized. This suggests that their quality of life is not significantly affected by their oral condition. Supported by CRGC-HKU & RGC, HKSAR)

58 Patterns of change in the oral health of older adults over seven years. A. JOKOVIC*, M. STEPHENS, D. LOCKER (Faculty of Dentistry, University of Toronto, Canada).

This study investigated patterns of change in the self-perceived oral health status of community-dwelling Canadians aged 50 years and over. Data were collected at baseline, 3 and 7 years from 422 participants in the Ontario Study of the Oral Health of Older Adults, of whom 60 were edentulous when recruited. Self-perceived oral health status was measured using four indicators: a 6-item chewing index, a 9-item oral pain inventory, an 11-item inventory of other oral symptoms and a 7-item psychosocial impact index. Each indicator gave rise to a numerical score that could be reduced to a dichotomy to give an estimate of the prevalence of oral health problems. Changes in proportions over time were evaluated with Cochran's Q test for k related samples and McNemar's test for paired samples. At baseline, the least prevalent oral health problem was psychosocial impact (dentate 8.8%, edentulous 18.3%; $p < 0.05$), and the most prevalent was oral symptoms other than pain (dentate 63.8%, edentulous 75.0%; ns). While the level of psychosocial impact fluctuated during the two observation periods, the proportions of those reporting chewing, pain and other oral symptoms increased steadily and significantly ($p < 0.001$, Cochran's Q). McNemar's tests indicated that the critical changes for chewing problems occurred between the baseline and 3-year follow-up and for oral symptoms between the 3- and 7-year follow-ups. The prevalence increase over seven years was more marked among the edentulous, ranging from 13.1% (oral symptoms other than pain) to 30.0% (oral pain). These results suggest that oral health status of older adults declines significantly over time, with the decrements being more pronounced among the edentulous. Supported by: Ontario Ministry of Health Grant #04170.

59 Measuring the outcomes of dental care for older adults. D. LOCKER (Faculty of Dentistry, University of Toronto, Toronto, Canada)

A number of methods have been used to measure change in self-perceived oral health. These include global transition judgements and change scores derived from repeat administrations of OHRQOL measures. This paper examines the use of these two approaches in terms of their sensitivity to the outcomes of dental care provision to older adults. Data were collected as part of the Ontario Study of the Oral Health of Older Adults, a survey of community-dwelling persons aged 50 years and over. 611 subjects were interviewed and clinically examined at baseline and three-year follow-up. The OHRQOL of these subjects was measured using a battery of subjective oral health status indicators (SOHSI). 495 subjects reported at least one dental visit during the three-year observation period. The dental records of 408 were obtained and provided data on the number of visits and the volume and value of dental services received. Using the global transition judgements, 10.4% of subjects reported improved oral health, 70.8% that their oral health had remained the same and 18.8% that it had deteriorated over three years. The mean number of dental services received by these three groups were: 15.7, 9.8, 10.7; $p < 0.0001$. The change scores derived from the SOHSI revealed a similar amount of change. However, correlations between these scores and the number of services received were below 0.10 and none were significant. This indicates that the SOHSI is not a good evaluative measure for assessing the outcomes of dental care for older adults. Supported by Ontario Ministry of Health Grant # 04170.

60 *Candida albicans* Switch Phenotypes from HIV+ Patient Differ in Fluconazole Susceptibility. T. SIFRI* and K. VARGAS (Univ. of Iowa, Iowa City, IA).

Candida albicans and related species have been shown to switch at high frequency between a number of general phenotypes distinguishable by colony morphology. Studies have suggested that phenotypic switching is involved in pathogenesis and has been shown to affect characteristics such as: antigenicity, sensitivity to PMN and virulence in the mouse. Since antifungal drug resistance has been an increasing problem in immunocompromised individuals, it was the purpose of this study to evaluate whether different switch phenotypes of the same strain of *Candida albicans* vary in their antifungal susceptibility to fluconazole. A *C. albicans* strain isolated from an HIV+ individual was used for this study. After establishing the minimum inhibitory concentrations (MIC) to fluconazole for five switch phenotypes from this strain (smooth white, ring, star, wrinkled and heavily myceliated), growth kinetic assays were conducted. Switch phenotypes were adjusted to 1×10^7 cells in YPD broth and mixed with either no antifungal (control) or 2X, 4X or 6X the MIC and incubated aerobically at 35°C. At 0, 1, 2, 3, 4, 6, 8, 12 and 24 hours, aliquots were removed from each tube and plated onto modified Lee's agar plates, incubated at 25°C for 7 days. Colonies were then counted for phenotype stability and killing by the antifungal. Statistical analysis was done using one-way ANOVA and Tukey post-hoc tests. Our results showed that at even 6X the MIC, the star, ring and heavily myceliated phenotypes grew as well as the control. At all MIC concentrations, the smooth white phenotype was significantly ($p < 0.05$) inhibited by fluconazole and growth of the wrinkled phenotype was only affected at 4X and 6X the MIC ($p < 0.05$). Differences in growth rates among the phenotypes were also found to be statistically significant at 4X and 6X MIC ($p < 0.05$). These results suggest that phenotypic switching may enable *C. albicans* to escape threatening environments, and enable it to become resistant to antifungal treatment. This research project was supported by NIH grant DE00364 and a Dows Student Research Award, The University of Iowa College of Dentistry.

61 The Influence of Media Composition and Growth Phase on the Susceptibility of *Candida albicans* to Histatin 5. U. LENDEMANN*, S. KIM, R. F. TROXLER & F. G. OPPENHEIM (Boston University Goldman School of Dental Medicine, Boston, Massachusetts).

The effects of antimicrobial drugs on pathogenic organisms often depend upon the growth conditions employed. This is particularly true for agents which are lethal only against metabolically active target organisms. The anticandidal activity of histatin 5 requires cellular energy, and indeed, we previously found that the concentrations of histatin 5 lethal to 50% of cells (LC_{50}) was $12.7 \pm 2.2 \mu\text{M}$ for a culture exponentially growing in Sabouraud dextrose broth whereas an LC_{50} -value of $132 \pm 27 \mu\text{M}$ was observed for stationary cells. In this study, we tested how the chemical composition of the growth medium affects the susceptibility of *C. albicans* towards histatin 5. Cells were grown on minimal media containing mineral salts, vitamins and glucose, fructose, glycerol or succinate as source of carbon and energy. LC_{50} -values were 2.0, 1.9, 1.7 and 2.1 μM for cells grown on glucose, fructose, glycerol and succinate, respectively. These results show first, that cells grown on minimal media are more susceptible to histatin 5 than cells grown on Sabouraud dextrose broth and second, that the LC_{50} -values do not significantly depend on the carbon source. Similar LC_{50} -values were observed for both exponentially growing and stationary phase cells when the compositions of the minimal media were selected such that nitrogen rather than carbon was the growth-limiting nutrient. These results show that a better reproducibility of LC_{50} -values can be obtained when growing cells under nitrogen-limited conditions in chemically defined minimal media. Supported by NIH/NIDR grants DE05672 and DE07652.

62 Histatin 5 Inhibits Respiration of Mitochondria Isolated from *Candida albicans*. E.J. HELMERHORST*, U. LENDEMANN, R.F. TROXLER, F.G. OPPENHEIM (Boston University Goldman School of Dental Medicine, Boston, MA).

Histatins are salivary proteins with anti-candidal activity *in vitro*. The susceptibility of *C. albicans* is strongly reduced by agents that interfere with mitochondrial activity such as sodium azide, an inhibitor of the cytochrome aa₃ complex of the respiratory chain, and by anoxic conditions. Previous studies have shown that histatin 5 has affinity for mitochondria, based on the *in situ* co-localization with a mitochondrion-specific marker. In the present study the interaction of histatin 5 with yeast mitochondria was further explored. For this purpose, mitochondria were isolated from *C. albicans* spheroplasts under hypo-osmotic conditions using a manual Potter-Elvehjem homogenizer. The mitochondria were well coupled based on a respiratory control ratio of 2.3. The antimicrobial magainin peptide PGL₂, which is known to possess uncoupling properties, increased State 2 respiration by a factor 3.7 at 6.7 $\mu\text{g/ml}$. At similar low concentrations, histatin 5 had no uncoupling effect, and stepwise increases of histatin levels led to complete inhibition of respiration. This inhibition could neither be reversed by addition of the uncoupler CCCP, nor circumvented by addition of sodium succinate. This suggests interference of histatin 5 with a respiratory chain component downstream of coenzyme Q, involving the cytochromes. The inhibition by histatin 5 was specific, since a control peptide of similar size at high concentration had no effect. The data show that histatin 5 inhibits respiration between coenzyme Q and molecular oxygen. Inhibition of respiration may well be the principle lethal effect of histatins against *C. albicans*. Supported by NIH/NIDCR Grants DE05672 and DE07652.

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WITHDRAWN

64 Impacts of PAFE on the relative CSH of *Candida albicans*. H.EGUSA^{1,2}, A.N.B. ELLEPOLA², H.NIKAWA¹, T.HAMADA¹, L.P.SAMARANAYAKE² (¹Hiroshima Univ. Hiroshima, Japan, ²Univ. of Hong Kong, Hong Kong).

Post antifungal effect (PAFE) is defined as the suppression of growth that persists following limited exposure of fungi to antimycotics and subsequent removal of the drug. The fungal pathogen *C. albicans* is the major aetiological agent of oral candidosis and the cell surface hydrophobicity (CSH) of this yeast is considered a critical factor contributing to its colonisation potential. As the concentration of topically prescribed antifungals reach sub-therapeutic levels at dosage intervals, the study of the polyene-induced PAFE and its impact on the CSH of oral *C. albicans* would be of clinical relevance. Hence the aims of this investigation were to measure the PAFE and CSH of 12 isolates of *C. albicans* following limited exposure (1 hour) to nystatin and amphotericin B and secondly to investigate the ultrastructural features of yeast cells following such antifungal exposure. The yeasts were exposed to sub-lethal concentrations of nystatin ($\times 2$ MIC) and amphotericin B ($\times 2$ MIC) for a period of 1 hour. Following subsequent removal of the drug, the PAFE and the CSH of the isolates were assessed by a turbidometric measurement of growth (Archs Oral Biol 1998; 43: 999-1007) and a biphasic aqueous-hydrocarbon assay (FEMS Microbiol Lett 1987; 48: 159-63), respectively. The mean duration of PAFE of nystatin and amphotericin B were $5.99 (\pm 0.49)$ hours and $8.73 (\pm 0.93)$ hours, respectively while the reduction in CSH following exposure to these drugs were 17.32% ($p < 0.05$ for 83% of the isolates) and 14.26% ($p < 0.05$ for 66% of the isolates), respectively. On scanning electron microscopy the exposed cells were seen to undergo collapse of the internal cell membrane leaving a "ghost-like" intact cell wall while a proportion of cells were deflated possibly due to efflux of cellular constituents. Some cells showed intense puckering of the cell wall resulting in a mulberry appearance. Taken together, these data elucidate additional mechanisms by which polyene antimycotics may operate *in vivo* to suppress candidal pathogenicity.