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(54) **METHODS FOR DETERMINING
SUSCEPTIBILITY TO DENTAL CARIES**

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See application file for complete search history.

(56) **References Cited**

U.S. PATENT DOCUMENTS

7,309,759 B2 * 12/2007 Krieger et al. 530/300

OTHER PUBLICATIONS

Goebel et al. "Peptides, vol. 21, pp. 757-765, 2000".*

Sequence alignment for Seq ID No. 1.*

Burgess et al. (J of Cell Biology, 1990 vol. 111, pp. 2129-2138).*

Lazar et al (Molecular and Cellular Biology, 1988, vol. 8, pp. 1247-1252).*

Mizukawa et al. (Oral Surgery Oral Medicine Oral Pathology vol. 87, No. 5, May 1999).*

Dale et al. (J ournal of Oral Pathol. Med. vol. 30, pp. 321-327, Feb. 2001).*

Bals, R., and J.M. Wilson, "Cathelicidins—A Family of Multifunctional Antimicrobial Peptides," *Cell Mol. Life Sci.* 60:711-720, 2003.

Bonass, W.A., et al., "Expression of β -Defensin Genes by Human Salivary Glands," *Oral Microbiol. Immunol.* 14:371-374, 1999.

Dale, B.A., and L.P. Fredericks, *Antimicrobial Peptides in Human Health and Disease*, Horizon Scientific Press, Hethersett, United Kingdom, 2001, Chap. 8, "Antimicrobial Peptides in the Oral Environment: Expression and Function in Health and Disease," pp. 223-252.

Dale, B.A., and S. Krisanaprakornkit, "Defensin Antimicrobial Peptides in the Oral Cavity," *J. Oral Pathol. Med.* 30:321-327, 2001.

Dale, B.A., et al., "Localized Antimicrobial Peptide Expression in Human Gingiva," *J. Periodont. Res.* 36:285-294, 2001.

Dunsche, A., et al., "The Novel Human Beta-Defensin-3 Is Widely Expressed in Oral Tissues," *Eur. J. Oral Sci.* 109:121-124, 2002.

Ganz, T., "Defensins: Antimicrobial Peptides of Innate Immunity," *Nat. Rev. Immunol.* 3:710-720, 2003.

Ganz, T., et al., "Defensins: Natural Peptide Antibiotics of Human Neutrophils," *J. Clin. Invest.* 76:1427-1435, 1985.

Hicks, J., et al., "Biological Factors in Dental Caries: Role of Saliva and Dental Plaque in the Dynamic Process of Demineralization and Remineralization (Part 1)," *J. Clin. Pediatr. Dent.* 28(1):47-52, 2003.

Hollox, E.J., et al., "Extensive Normal Copy Number Variation of a β -Defensin Antimicrobial-Gene Cluster," *Am. J. Hum. Genet.* 73:591-600, 2003.

Joly, S., et al., "Human β -Defensins 2 and 3 Demonstrate Strain-Selective Activity Against Oral Microorganisms," *J. Clin. Microbiol.* 42(3):1024-1029, 2004.

Linzmeier, R.M., and T. Ganz, "Human Defensin Gene Copy Polymorphisms: Comprehensive Analysis of Independent Variation in α - and β -Defensin Regions at 8p. 22-p. 23," *Genomics*, 2005 <http://www.sciencedirect.com>.

Loesche, W.J., "Role of *Streptococcus mutans* in Human Dental Decay," *Microbiol. Rev.* 50(4):353-380, 1986.

Maisetta, G., et al., "Activity of Human β -Defensin 3 Alone or Combined With Other Antimicrobial Agents Against Oral Bacteria," *Antimicrob. Agents Chemother.* 47(10):3349-3351, 2003.

Mars, W.M., et al., "Inheritance of Unequal Numbers of the Genes Encoding the Human Neutrophil Defensins HP-1 and HP-3," *J. Biol. Chem.* 270(51):30371-30376, 1995.

McKay, M.S., et al., "Immunomagnetic Recovery of Human Neutrophil Defensins From the Human Gingival Crevice," *Oral Microbiol. Immunol.* 14:190-193, 1999.

Murakami, M., et al., "Cathelicidin Antimicrobial Peptides Are Expressed in Salivary Glands and Saliva," *J. Dental Res.* 81(12):845-850, 2002.

Nagaoka, I., et al., "Synergistic Actions of Antibacterial Neutrophil Defensins and Cathelicidins," *Inflamm. Res.* 49:73-79, 2000.

Nishimura, E., et al., "Oral *Streptococci* Exhibit Diverse Susceptibility to Human β -Defensin-2: Antimicrobial Effects of hBD-2 on Oral *Streptococci*," *Curr. Microbiol.* 48:85-87, 2004.

Sahasrabudhe, K.S., et al., "Expression of the Antimicrobial Peptide, Human β -Defensin 1, in Duct Cells of Minor Salivary Glands and Detection in Saliva," *J. Dental. Res.* 79(9):1669-1674, 2000.

Selsted, M.E., and A.J. Ouellette, "Mammalian Defensins in the Antimicrobial Immune Response," *Nat. Immunol.* 6(6):551-557, 2005.

Tanaka, D., et al., "Sensitivity of *Actinobacillus actinomycetemcomitans* and *Capnocytophaga* spp. to the Bactericidal Action of LL-37: A Cathelicidin Found in Human Leukocytes and Epithelium," *Oral Microbiol. Immunol.* 15:226-231, 2000.

An Nieuw Amerongen, A., et al., "Salivary Proteins: Protective and Diagnostic Value in Cariology?" *Caries Res.* 38:247-253, 2004.

(Continued)

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(57) **ABSTRACT**

The present invention provides methods for determining whether a human being is susceptible to dental caries. The methods each include the steps of measuring the amount of α -defensins HNP 1, HNP 2 and HNP 3 in saliva obtained from a human being, and determining whether a reduced amount of the α -defensins HNP 1, HNP 2 and HNP 3 is present in the saliva, thereby determining whether the human being is susceptible to dental caries.

13 Claims, No Drawings