Journal of Clinical Periodontology
Official Journal of The European Federation of Periodontology
Founded by the British, Dutch, French, German, Scandinavian, and Swiss Societies of Periodontology

J Clin Periodontol 2009; 36: 913–914 doi: 10.1111/j.1600-051X.2009.01456.x

Journal of Clinical Periodontology

Guest Editorial

Of mice and men: animal models of human periodontal disease

Daniel H. Fine

Department of Oral Biology, University of Medicine and Dentistry of New Jersey, Newark, NJ, USA

Key words: bone loss; micro-computed tomography; periodontal disease; *P. gingivalis*

Accepted for publication 21 June 2009

Fine DH. Of mice and men: animal models of human periodontal disease. J Clin Periodontol 2009; 36: 913–914. doi: 10.1111/j.1600-051X.2009.01456.x.

Animal models can provide critically important information regarding periodontal disease pathogenesis (Graves et al. 2008). This benefit results from the fact that these models can assess disease in a longitudinal manner on a cellular and molecular level. As such, animal models can provide otherwise unattainable information at the host level in spite of shortcomings that are known to exist (Graves et al. 2008). To optimize their utility, animal models need to be approached with the same rigour that is applied to clinical models of human disease (Fine et al. 2001). Moreover, information obtained from animal models should be interpreted with the same caution used to analyse human models of disease. One critical issue related to the article by Wilensky and colleagues rests in the area of comparison of *Porphyromonas gingivalis*-induced host responsiveness or the lack thereof. All infections, without exception, are dependent on the dose and form of the infectious agent as well as the route of infection (Gibson et al. 2004). Further, the challenge with a foreign antigen should be put in the context of the time of exposure. While animal models of periodontal disease are still in their early stages of exploration these factors need to be considered.

With respect to *P. gingivalis*-induced periodontal disease in rodents, at first, it was difficult to understand how *P. gingivalis* could either colonize or survive in the oral cavity of rodents in spite of antibiotic suppression of the flora and the use of carboxymethyl cellulose to add to the ability of *P. gingivalis* to adhere (Evans et al. 1992). This was particularly true because *P. gingivalis* is difficult to grow in the laboratory due to its mandatory requirement for hemin and menadione-like substances and anaerobic growth conditions, which would not appear to be available in the mouths of healthy mice (Gibbons & Macdonald 1960). The question arose as to how this bacterium could survive in a supragingival environment in a disease-free mouse (Kesavalu et al. 1997). It was assumed that the use of lavage, gavage and the copraphagic nature of rodents led to re-innoculation and ultimate colonization (Chang et al. 1994). Genetic studies have shown that *P. gingivalis* possesses a variety of genes or segments of genes that allow it to attach to many surfaces such as teeth, soft tissue, etc. (Lamont & Jenkinson 1998). *P. gingivalis* also has a group of proteinases, known as gingipains, that allow it to degrade connective tissue and activate bone-resorbing cells (Lamont & Jenkinson 1998). Taken together, it would seem that this bacterium has the machinery required to colonize and cause disease in humans, and as more and more animal studies have accumulated over time, it appears that *P. gingivalis* can colonize and cause disease in mice (Baker et al. 2000, Baker & Roopenian 2002). We can conclude from this mass of data that the adaptability of this microbe has been severely underestimated (Hart et al. 2004).

The present study of Drs. Wilensky and colleagues is commendable. Their use of micro-computed tomography has added to the accuracy and reproducibility of interpretation of bone loss in the mouse model of periodontal disease and thus moved the model to a new level of competence (Wilensky et al. 2005). Further, because of the sensitivity of the methodology, the authors have been able to reduce the number of mice in each of the study groups to assess differences in bone loss in infected mice as compared with their uninfected controls. This of course makes for a more economical and humane model because of the limited number of mice required. Further, this report provides support and extends observations indicating that different strains of *P. gingivalis* cause different levels of disease (Baker & Roopenian 2002). Similar data have also been shown recently for *Aggregatibacter actinomycetemcomitans*-infected rodents (Fine et al. 2009). Moreover, it has been shown that different strains of rodents have different susceptibilities to a challenge from the same strain of *P. gingivalis* (Baker et al. 2000, Baker & Roopenian 2002). This holds true for *A. actinomycetemcomitans* as well (Schreiner et al. 2008). These observations parallel what is seen in human disease, particularly in the case of *A. actinomycetemcomitans*-related disease where the JP2 strain of *A. actinomycetemcomitans* appears to be more virulent, and where individuals of African heritage appear to be more susceptible to *A. actinomycetemcomitans* infection (Haubek et al. 2008). Taken together, these similarities support the utility of an animal model of periodontal disease.

However, with these considerations taken into account, it is still a concern that most reports of *P. gingivalis*-induced periodontal disease in mice, including this one, fail to report the infecting dose and the direct recovery of the organism from experimentally infected animals (Gibson & Genco 2001). This is especially important in

studies where one strain of *P. gingivalis* is compared with another with the goal of assessing strain-dependent pathogenesis. Moreover, while it is well known that growing *P. gingivalis* in the laboratory can be fraught with difficulty, current DNA methodologies, such as real-time PCR, allow for recovery and estimation of the infecting dose in a quantitative manner.

In this study by Wilensky and colleagues antibody titres were used to indicate exposure to *P. gingivalis* as well as an indicator of the host response. Can we assume that a low IgG titre means a lower level of host responsiveness to *P. gingivalis* 53977, or, is it an indication of a lower infecting dose of *P. gingivalis* 53977? Either could be true; however, with respect to antibody titres, antibody levels represent a past history of exposure to the challenging antigen. Without demonstrating some equivalence of the infecting dose among the strains of *P. gingivalis* used, and the time-dependent changes in antibody titres resulting from that infecting dose, it is difficult to conclude that one strain is more pathogenic than the other. With this said, the periodontal literature has taught us that the assessment of the bacterial challenge and the host response to that challenge is best accomplished at the site of the infection (Assuma et al. 1998). There is no doubt that periodontal disease consists of a local infection that is directed at the surrounding tissues that support the tooth in its alveolus. The advantage of an animal model is that we can examine the local response in a time-dependent and site-specific and thus tissue-specific manner (Assuma et al. 1998). The models available for both *P. gingivalis* and *A. actinomycetemcomitans* have improved to the point where it is possible to advance to the next level, a level that permits us to examine the bacterial challenge at the local site as well as the host response to that challenge at the local level in a quantitative manner.

As we begin to gain a greater understanding of the periodontal disease process, it becomes more and more apparent that we need to consider both the bacterial initiator and the host response to these so-called ''pathogenic'' oral microbes if we hope to gain a better understanding of disease. The statements made herein are not intended to detract from the importance of the manuscript by Wilensky and colleagues but are aimed at pointing out that both the bacterial challenge as well as the host response to that challenge need to be considered in a quantitative, time-dependent, site- and tissue-specific manner so that the conclusions drawn from the data obtained can be extrapolated to the broader biological questions that we seek to answer.

References

Assuma, R., Oates, T., Cochran, D., Amar, S. & Graves, D. T. (1998) IL-1 and TNF antagonists inhibit the inflammatory response and bone loss in experimental periodontitis. *Journal of Immunology* **160**, 403–409.

Baker, P. J., Dixon, M., Evans, R. T. & Roopenian, D. C. (2000) Heterogeneity of *Porhpyromonas gingivalis* strains in the induction of bone loss in mice. *Oral Microbiology and Immunology* **15**, 27–32.

Baker, P. J. & Roopenian, D. C. (2002) Genetic susceptibility to chronic periodontal disease. *Microbes and Infection* **4**, 1157–1167.

Chang, K. M., Ramamurthy, N., McNamara, T., Evans, R., Klausen, B., Murray, P. & Golub, L. (1994) Tetracyclines inhibit *Porphyromonas gingivalis*-induced alveolar bone loss in rats by a non-anti-microbial mechanism. *Journal of Periodontal Research* **29**, 242–249.

Evans, R. K., Sojar, B., Bedi, H. T., Sfinrwau, G. S., Ramamurthy, N. S., Golub, L. M. & Genco, R. J. (1992) Immunization with *Porphyromonas* (Bacteroides) *gingivalis* fimbriae protects against periodontal destruction. *Infection and Immunity* **60**, 2926–2935.

Fine, D. H., Goncharoff, P., Schreiner, H. C., Chang, K. M., Furgang, D. & Figurski, D. (2001) Colonization and persistence of rough and smooth colony variants of *Actinobacillus actinomycetemcomitans* in the mouths of rats. *Archives of Oral Biology* **46**, 1065–1078.

Fine, D. H., Schreiner, H., Nasri-Heir, C., Greenberg, B., Jiang, S., Markowitz, K. & Furgang, D. (2009) An improved cost-effective, reproducible method for evaluation of bone loss in a rodent model. *Journal of Clinical Periodontology* **36**, 106–113.

Gibbons, R. J. & Macdonald, J. B. (1960) Hemin and vitamin K compounds as required factors for the cultivation of certain strains of *Bacteriodes melaninogenicus*. *Journal of Bacteriology* **80**, 200–206.

Gibson, F. C. III & Genco, C. A. (2001) Prevention of *Porphyromonas gingivalis*-induced oral bone loss following immunization with gingipain R1. *Infection and Immunity* **69**, 7959–7963.

Gibson, F. C. III, Gonzalez, D. A., Wong, J. & Genco, C. A. (2004) *Porphyromonas gingivalis*-specific immunoglobulin G prevents *P. gingivalis*-elicited oral bone loss in a murine model. *Infection and Immunity* **72**, 2408–2411.

Graves, D. T., Fine, D., Tang, Y.-T., Van Dyke, T. E. & Hajishengalls, G. (2008) The use of rodent models to investigate host-bacteria interactions related to periodontal diseases. *Journal of Clinical Periodontolog* **35**, 89–105.

Hart, G., Shaffer, D. J., Akilesh, S., Brown, A. C., Moran, L., Roopenian, D. C. & Baker, P. J. (2004) Quantitative gene expression profiling implicates genes for susceptibility and resistance to alveolar bone loss. *Infection and Immunity* **72**, 4471–4479.

Haubek, D., Ennibi, O.-K., Poulsen, P., Vaeth, M. & Kilian, M. (2008) Risk of aggressive periodontitis in adolescent carriers of the JP2 clone of *Aggregatibacter* (Actinobacillus) *actinomycetemcomitans* in Morocco: a prospective longitudinal cohort study. *Lancet* **371**, 237–242.

Kesavalu, L., Holt, S. C. & Ebersole, J. L. (1997) *Porphyrmonas gingivalis* virulence in a murine lesion model: effects of immune alterations. *Microbial Pathogenesis* **23**, 317–326.

Lamont, R. J. & Jenkinson, H. F. (1998) Life below the gum line: pathogenic mechanisms of *Porphyromonas gingivalis*. *Microbiology and Molecular Biology Reviews* **62**, 1244–1263.

Schreiner, H. C., Markowitz, K., Moore, D., Miryalkar, . M., Diehl, S. R. & Fine, D. H. (2008) *A. actinomycetemcomitans* colonization and induced bone loss in three rat strains. *Journal of Dental Research* **87**, (Spec Iss B): #2170, www.dentalresearch.org.

Wilensky, A., Gabet, Y., Yumoto, H., Houri-Haddad, Y. & Shapira, L. (2005) Three-dimensional quantification of alveolar bone loss in *Porhpyromonas gingivalis*-infected mice using micro-computed tomography. *Journal of Periodontology* **76**, 1282–1286.

Address:
Daniel H. Fine
Department of Oral Biology
University of Medicine and Dentistry of New Jersey
185 South Orange Ave
Newark
NJ 07103
USA
E-mail: Finedh@umdnj.edu