**Invited review**

# Antimicrobial function of human saliva—how important is it for oral health?

Jorma Tenovuo
Institute of Dentistry and Turku Immunology Centre, University of Turku, Turku, Finland

Tenovuo J. Antimicrobial function of human saliva—how important is it for oral health? Acta Odontol Scand 1998;56:250–256. Oslo. ISSN 0001-6357.

Human saliva contains a number of physical, physicochemical, and chemical agents that protect oral tissues against noxious compounds, in particular those produced by various microorganisms. Among such protective factors, the flushing effect of saliva flow is the most important one, not only because it so effectively removes exogenous and endogenous microorganisms and their products into the gut but also because a steady supply of saliva guarantees continuous presence of both non-immune and immune factors in the mouth. A great number of studies with controversial results have been published regarding various individual agents and their possible association to oral health, particularly to dental caries. It appears that no single chemical agent is far more important than the others. For example, patients with selective IgA deficiency have normal levels of non-immune defense factors and often display a compensatory increase in the other immunoglobulin isotypes. The concerted action of all agents in whole saliva, both saliva- and serum-derived, provides a multifunctional protective network that is collapsed only if salivary flow rate is substantially reduced. In this mixture of defense factors, many show additive or even synergistic interactions against oral pathogens. Increased knowledge of the molecular functions of various agents has made it possible to prepare oral hygiene products that include host-derived antimicrobial agents instead of synthetic agents. Although the clinical efficacy of such products is still unsatisfactory and poorly described, new technologies, for example in the production of specific antibodies against oral pathogens, may considerably improve the antimicrobial power of these products. □ *Antimicrobials; dental caries; host defense; immunoglobulins; peroxidases; saliva*

*Jorma Tenovuo, Institute of Dentistry and Turku Immunology Centre, University of Turku, Lemminkäisenkatu 2, FIN-20520 Turku, Finland*

Human saliva not only lubricates oral tissues, making oral functions such as swallowing and speaking possible, but also protects oral tissues, teeth, and mucosal surfaces in many ways. The main protective factor is the constant flow of saliva from the mouth into the gut, and this 'flushing effect' transfers, for example, food debris and exogenous and possibly noxious agents (food-borne mutagens, bacteria, viruses) into the gut. Some of the mutagens (1–3) or exogenous microorganisms (4, 5) are detoxified and killed by innate components of human saliva, but salivary components also display remarkable antiviral activity both in vitro and in vivo (6–10). Also, endogenous oral microflora and many of its harmful metabolic products are inhibited or neutralized by salivary components (Fig. 1). These host factors, apart from salivary flow rate and buffer effect, which are responsible for saliva's protective and antimicrobial functions, are listed in Table 1.

## Ontogeny of salivary antimicrobial agents

*Non-immune agents*

During early childhood the non-immune salivary factors work already at almost full capacity (11). In mixed saliva of newborn babies, lysozyme, salivary peroxidase, and the peroxidase-generated antimicrobial agent hypothiocyanite ($OSCN^-$/HOSCN) are already at levels similar to those of adults, whereas lactoferrin, myeloperoxidase, and total protein are still significantly less abundant (11, 12). The levels of all antimicrobial agents in infants' saliva depend very much on the sample collection technique, because these same agents exist also in nasal and lacrimal secretions, which easily contaminate saliva in restless children (11). Non-immune factors are not notably affected by the emergence of teeth (which makes serum contribution possible), length of breast-feeding, or history of antibiotic treatment (11, 13). All non-immune factors, including lactoferrin and myeloperoxidase, are at adult levels by the early teenage years (14). All these factors remain at high concentrations even among elderly people (up to the age of 90 years), unless a considerable number of teeth are extracted, which leads to a depleted influx of serum-derived agents (15). In cases where serum transudation is enhanced, such as after tonsillectomy (16, 17) or as a consequence of periodontal (18) or lingual (19) inflammations, serum-derived components (mainly myeloperoxidase, lysozyme, and lactoferrin) display increased concentrations in whole saliva. To conclude, these findings indicate that non-immune factors maintain their individual levels throughout life, but oral infections often increase the levels of those components that are found also in human serum.

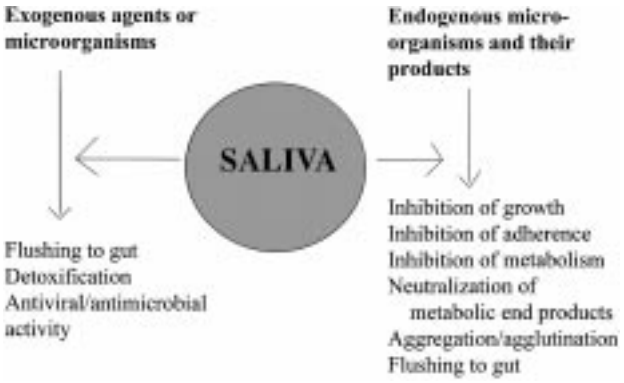


Fig. 1. Physical, physicochemical, and chemical components of whole saliva protect the human mouth against both exogenous and endogenous noxious agents by several mechanisms.

*Immunoglobulins*

Salivary antibodies against oral pathogens, such as mutans streptococci, are induced by the common mucosal immune system, which produces protective antibodies on mucosal surfaces, including those in human mouth (20). The gut-associated lymphoid tissues (GALT), in particular Peyer's patches, are essential since they contain precursor IgA B cells with a potential to populate target tissues on mucosal surfaces. IgA-committed B cells from GALT migrate to glandular epithelia (e.g. in lacrimal, mammary, and salivary glands) and eventually synthesize secretory IgA (sIgA) antibodies in saliva (20). Also, local stimulation of minor salivary glands by oral antigens may lead to the development of sIgA antibodies (21). Thus, duct-associated lymphoid tissues (DALT) of minor salivary glands can also be regarded as an integral part of oral mucosal defense (Fig. 2).

Salivary sIgA antibodies to mucosal bacteria, such as *Escherichia coli*, *Streptococcus mitis*, and *Streptococcus salivarius*, begin to appear as early as the first weeks of life (22), and they approach adult levels by 1 to 2 years of age. In general, the appearance of specific antibodies in saliva correlates with the colonization of the mouth by the respective microorganisms, and most children over 3 years of age already have salivary sIgA antibodies reactive with whole cells or serotype antigens of mutans streptococci (23–30). The total concentration of sIgA in saliva tends to increase with age, but response to specific antigens, such as *Streptococcus mutans* glycosyltransferases, is highest between 30 and 65 years of age (29). Of the two isotypes of sIgA, $IgA_1$ is more prevalent among infants than adults and the elderly (29).

Serum-derived antibodies, mainly of IgG isotype, also exist in human whole saliva. Such IgG antibodies to oral streptococci have been detected in children's sera as early as the first year of life (31–33). Although the frequency of samples positive for IgG antibodies against mutans streptococci varies greatly in different populations, it seems that virtually all children develop *S. mutans*–specific serum antibodies during preschool age. The main route of entry for IgG antibodies into whole saliva is the gingival crevice (13, 34). Saliva, on the other hand, has been reported to increase serum IgG retention on *S. mutans* cells, in particular among caries-resistant subjects (35). Thus, it seems likely that serum-derived IgG antibodies to oral pathogens, such as mutans streptococci, could also have a significant role in host defense. In fact, results supporting this protective role have been obtained when the extent of serum IgG response, oral levels of mutans streptococci, and preschool caries increment have been studied (36, 37).

The mechanism of induction of serum IgG to mutans streptococci is not known in detail, but the following hypothesis has been postulated (28, 33, 36). In predentulous babies, maternally derived mutans streptococci pass through the oral cavity and are swallowed (Fig. 2). Serotype and bacteriocin analyses have indicated that fecal and oral isolates of mutans streptococci among individuals are usually the same, suggesting that ingested streptococci indeed enter the intestinal canal. However, in young children the epithelial structure of the intestine is still incomplete, thus allowing antigens to pass through the mucosa. Also, the mucosa-protecting sIgA system is immature in early childhood, and therefore it is likely

Table 1. Major antimicrobial agents in human whole saliva

| Agent | Major target/function |
|---|---|
| Immune factors | |
| Secretory IgA | Inhibition of adhesion |
| IgM | Enhancement of phagocytosis (?) |
| IgG | Enhancement of phagocytosis |
| Non-immune, innate factors | |
| Salivary and myeloperoxidase | Antimicrobial, decomposition of $H_2O_2$ |
| Lysozyme | Gram-positive bacteria, *Candida* |
| Lactoferrin | Gram-positive and -negative bacteria |
| Agglutinins | Agglutination/aggregation of a number of microorganisms |
| Histatins (histidine-rich peptides) | Antibacterial, antifungal |
| PRPs (proline-rich peptides) | Antibacterial, antiviral |
| Cystatins | Antiviral |
| Polymorphonuclear leukocytes | Phagocytosis |

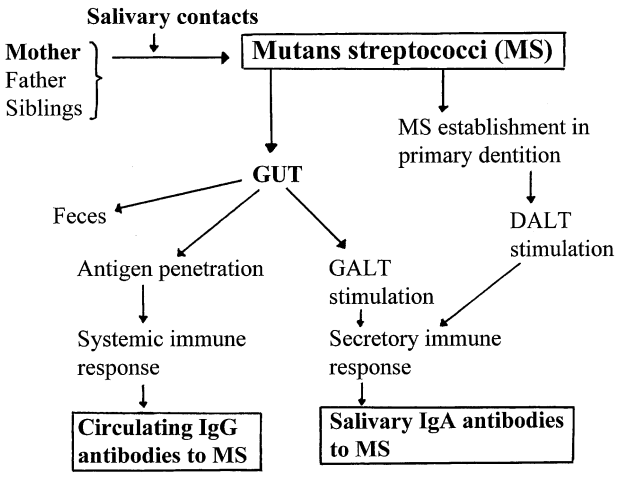


Fig. 2. Proposed scheme for the development of specific serum and salivary antibodies against mutans streptococci in children (details in the text; modified from Tenovuo (30)). GALT = gut-associated lymphoid tissues; DALT = duct-associated lymphoid tissues.

that gut mucosa serves as a route of entry for pathogens to evoke a systemic IgG response to oral bacteria at an early age. This assumption is supported by our studies (36, 38) in which children who had had frequent salivary close contact with their mothers during the first year of life had significantly more serum IgG antibodies to mutans streptococci during childhood compared with children whose mothers had avoided such contact. However, the avidity of these antibodies in children with frequent close contact was significantly lower than those with less frequent contact.

## Immunodeficiency and dental caries

Immunodeficiency provides a unique model to evaluate the significance of salivary antimicrobial agents for oral health. The most common form of primary immunodeficiency is selective IgA deficiency (39, 40), with a frequency of 1:300 to 1:3000, depending on the population studied. Selective IgA deficiency is classically defined as a serum IgA level less than 0.05 g/L, but serum IgG and IgM levels are usually normal or even elevated (41). Patients with serum IgA deficiency normally lack also salivary sIgA (as well as sIgA on other mucosal surfaces), although rare exceptions may exist (42).

Subjects with IgA deficiency are often asymptomatic, but if symptoms exist they usually appear as increased frequency of respiratory infections, pneumonia, intestinal infections, and autoimmune diseases (43). Salivary IgA is in general significantly lower among infection-prone children than healthy controls (44). All these observations suggest that immunodeficient patients should also be susceptible to oral infections, such as periodontal diseases and dental caries. However, this is not supported by the available data. Among IgA-deficient subjects, both significantly higher (45, 46) and lower (47, 48) caries experience has been reported in comparisons with age- and plaque-matched controls. Also, more recent studies by Norhagen Engström et al. (49), Dahlén et al. (50), and Kirstilä et al. (51) failed to show any enhanced risk for dental caries or periodontal diseases among subjects with immunodeficiency. Interestingly, both Arnold et al. (52) and Kirstilä et al. (51) have found that immunocompromised subjects have normal (or even slightly elevated) levels of all non-immune antimicrobial agents in saliva, which in this way may form a 'backup system' in these relatively unusual cases. Furthermore, some of the IgA-deficient subjects compensate for the lack of IgA by producing secretory IgM (53), and these subjects do not differ from others with respect to caries susceptibility (52). It should also be kept in mind that more frequent antibiotic therapy of immunodeficient patients could resist the development of dental caries and periodontal diseases (48). Mutans streptococci have retained their susceptibility to antibiotics (54) in spite of continuous and increasing pressure by common antimicrobials.

To conclude, it seems that naturally evoked salivary antibodies do not have enough power to influence the development of dental caries or periodontal diseases, at least not among adolescents and adults. The non-immune factors seem to work effectively enough in all individuals, provided that proper salivary flow rate supplies these agents constantly to the oral cavity.

## Are all antimicrobial agents equally important?

Although a large number of studies have been published about individual antimicrobial agents in saliva, no clear evidence exists that any one of them is more important than the others. A good example is the previously described IgA deficiency, which does not seem to have any major impact on oral health. In studies where clinical parameters have been analyzed with respect to the concentrations or activities of oral defense factors, inconclusive results have been reported (13, 55, 56). Several reasons, such as differences in study design and populations, sample collections, assay methods, and statistical techniques, could have influenced the results considerably (56). Perhaps even more important is the fact that the number of salivary antimicrobial agents existing simultaneously in saliva and in plaque fluid is large, that their concentrations tend to be correlated (57), and that they depend on age (11) as well as salivary flow rate in an individual manner (58, 59).

Interestingly, many of the salivary antimicrobial agents interact with each other, in most cases in a synergistic or additive way. For example, such interactions have been reported between sIgA and peroxidase (60), lactoferrin and peroxidase (61, 62), lactoferrin and lysozyme (63), and lysozyme and histatins (64). Although these observations are from in vitro experiments, it is likely that such concerted effects exist also in vivo in mixed saliva, where

Table 2. Examples of methods to activate or enhance host defense in human saliva

| |
|---|
| 1. Increase of salivary flow rate and buffer effect: |
| – sugar-free chewing gums, sucking tablets, lozenges, xylitol, etc. |
| 2. Lactoperoxidase system-, lactoferrin-, and lysozyme-containing products: |
| – dentifrices, chewing gums, mouthrinses, moisturizing gels |
| 3. Immunization regimens: |
| – systemic (intramuscular) or oral immunization |
| – passive immunization (preformed IgG or IgY antibodies) |

all the components are simultaneously present. Thus, salivary antimicrobial factors form a network in which no single component is necessary for the overall antimicrobial capacity of the host's defense system.

## Can antimicrobial capacity of saliva be enhanced or restored?

Activation of the natural antimicrobial capacity of human whole saliva has been attempted in a number of ways (Table 2). Of course, increasing the flow rate with a concomitant increase in the output of saliva-borne factors has a high priority: it is very important in particular for patients with hyposalivation and/or xerostomia (65, 66). Lozenges that release buffering agents have also been developed to improve the quality of residual saliva (67, 68). For subjects with low salivary flow rate and symptoms of dry mouth, commercial products containing natural innate defense factors have been designed to restore or replace the host's own lacking defense systems. The first such commercial products were those designed to activate oral peroxidase systems (66, 69).

The actual antimicrobial agent produced by human oral peroxidases in vivo is hypothiocyanite ($OSCN^-$/HOSCN), which has a wide inhibitory spectrum against pathogenic bacteria, viruses, and yeasts (70–73). $OSCN^-$/HOSCN also shows an inverse relation with glucose-induced dental plaque acid production (74), and studies suggest that it is likely to be protective against initial phases of dental caries (14, 59). The limiting component for the production of $OSCN^-$/HOSCN is hydrogen peroxide (75), and therefore the endogenous peroxidase system in saliva can be activated or restored by elevating the amount of $H_2O_2$ in saliva by introducing either $H_2O_2$-generating enzymes or by introducing all the peroxidase system components (lactoperoxidase, $SCN^-$, $H_2O_2$-generating enzymes) into the mouth in the form of oral hygiene products (69). Examples of such products are some dentifrices (Zendium®, Biotene®), moisturizing gels (Oralbalance®), and mouthrinses (Biotene®, Zendium®). There is no evidence that the use of these products could promote oral health among persons with normal salivary flow rate, but in xerostomic patients they have been shown to heal irritated oral mucosa (76), reduce gingival inflammation (77), decrease oral anaerobic flora (78), and relieve the subjective symptoms of oral dryness (76, 79). Thus, these products seem to be beneficial for subjects with symptoms of dry mouth. It may be mentioned that also lysozyme and lactoferrin have been incorporated in some of these products to further mimic the in vivo situation, and that fluoride ions in these products have an additive effect with $OSCN^-$/HOSCN against mutans streptococci (80).

Several attempts to enhance the immune protection in the oral cavity have also been made. So far most of these studies have focused on developing immunization regimens against *S. mutans*. Because of the potential side effects when using whole cells of mutans streptococci for systemic immunization, recently more emphasis has been paid to oral immunization by purified antigens (81, 82), and to passive immunization in which preformed antibodies to *S. mutans* have been applied directly to the dentition (83–85).

Interesting approaches, currently under extensive research, to enhance the immune response against mutans streptococci are production of specific antibodies in bovine milk (83, 86), hen egg yolk (85), or transgenic plants, such as tobacco (87). Although clinical data regarding the efficacy of these antibodies to inhibit mutans streptococci or prevent dental caries are still very limited, they are already known to be potent anti-streptococcal agents. For example, specific antibodies in bovine immune whey inhibit extracellular polysaccharide formation by *S. mutans* (86), prevent the adherence of *S. mutans* to saliva-coated hydroxyapatite (88), aggregate both *S. mutans* and *Streptococcus sobrinus* cells (88), and support the phagocytosis and killing of mutans streptococci by human leukocytes (Loimaranta et al., unpublished). Furthermore, immune whey and $OSCN^-$/HOSCN act in an additive way against *S. mutans* (89), thus supporting the previously described additive or synergistic interactions between various host defense factors. All the above findings indicate that further development of products with non-immune or immune agents (or both) may in future be an important way to prevent oral infectious diseases without antibiotics or other synthetic antimicrobials.

*Acknowledgements.*—Studies of salivary defense factors in the author's laboratory have been supported by the Academy of Finland, the Sigrid Juselius Foundation, Patentsmedelsfondet, the Finnish Dental Society, and the Finnish Technology Development Centre.

## References

1. Nishioka H, Nishi K, Kyokane K. Human saliva inactivates mutagenicity of carcinogens. Mutat Res 1981;85:323–33.
2. Stich HF, Rosin MP, Bryson L. The inhibitory effect of whole and deproteinized saliva on mutagenicity and clastogenicity resulting from a model nitrosation reaction. Mutat Res 1982; 97:283–92.
3. Tenovuo J. The biochemistry of nitrates, nitrites, nitrosamines and other potential carcinogens in human saliva. J Oral Pathol 1986;15:303–7.
4. Purdy MA, Tenovuo J, Pruitt KM, White WE Jr. Effect of growth phase and cell envelope structure on susceptibility of

*Salmonella typhimurium* to the lactoperoxidase-thiocyanate-hydrogen peroxide system. Infect Immun 1983;39:1187–95.
5. Kamau DN, Doores S, Pruitt KM. Antibacterial activity of the lactoperoxidase system against *Listeria monocytogens* and *Staphylococcus aureus* in milk. J Food Protein 1990;53:1010–4.
6. Archibald DW, Cole GA. In vitro inhibition of HIV-1 infectivity by human salivas. AIDS Res Hum Retroviruses 1990;6:1425–32.
7. Bergey EJ, Gu M, Collins AR, Bradway SD, Levine MJ. Modulation of herpes simplex virus type 1 replication by human salivary secretions. Oral Microbiol Immunol 1993;8:89–93.
8. Gu M, Haraszthy GG, Collins AR, Bergey EJ. Identification of salivary proteins inhibiting herpes simplex virus 1 replication. Oral Microbiol Immunol 1995;10:54–9.
9. Courtois P, Van Beers D, De Foor M, Mandelbaum IM, Pourtois M. Abolition of herpes simplex cytophatic effect after treatment with peroxidase generated hypothiocyanite. J Biol Buccale 1990;18:71–4.
10. Mikola H, Waris M, Tenovuo J. Inhibition of herpes simplex virus type 1, respiratory syncytial virus and echovirus type 11 by peroxidase-generated hypothiocyanite. Antiviral Res 1995;26: 161–71.
11. Tenovuo J, Lehtonen O-P, Aaltonen AS, Vilja P, Tuohimaa P. Antimicrobial factors in whole saliva of human infants. Infect Immun 1986;51:49–53.
12. Mandel ID, Turett H, Alvarez J. Quantitation of salivary defense factors in human infants [abstract]. J Dent Res 1983;62:217.
13. Tenovuo J, Gråhn E, Lehtonen O-P, Hyyppä T, Karhuvaara L, Vilja P. Antimicrobial factors in saliva: ontogeny and relation to oral health. J Dent Res 1987;66:475–9.
14. Kirstilä V, Häkkinen P, Jentsch H, Vilja P, Tenovuo J. Longitudinal analysis of the association of human salivary antimicrobial agents to caries increment and cariogenic microorganisms: a 2-year cohort study. J Dent Res 1998;77:73–80.
15. Närhi TO, Tenovuo J, Ainamo A, Vilja P. Antimicrobial factors, sialic acid and protein concentration in whole saliva of the elderly. Scand J Dent Res 1994;102:120–5.
16. Lenander-Lumikari M, Tenovuo J, Puhakka HJ, Malvaranta T, Ruuskanen O, Meurman O, et al. Salivary antimicrobial proteins and mutans streptococci in tonsillectomized children. Pediatr Dent 1992;14:86–91.
17. Kirstilä V, Tenovuo J, Ruuskanen O, Suonpää J, Meurman O, Vilja P. Longitudinal analysis of human salivary immunoglobulins, nonimmune antimicrobial agents, and microflora after tonsillectomy. Clin Immunol Immunopathol 1996;80:110–5.
18. Saxén L, Tenovuo J, Vilja P. Salivary defence mechanisms in juvenile periodontitis. Acta Odontol Scand 1990;48:399–407.
19. Kullaa-Mikkonen A, Tenovuo J, Sorvari T. Changes in composition of whole saliva in patients with fissured tonque. Scand J Dent Res 1985;93:522–8.
20. McGhee JR, Michalek SM. Immunobiology of dental caries: microbial aspects and local immunity. Ann Rev Microbiol 1981;35:595–638.
21. Nair PNR, Schroeder HE. Duct-associated lymphoid tissue (DALT) of minor salivary glands and mucosal immunity. Immunology 1986;57:171–80.
22. Smith DJ, King WF, Taubman MA. Salivary IgA antibody to oral streptococcal antigens in predentate infants. Oral Microbiol Immunol 1990;5:57–62.
23. Lehner T, Murray JJ, Winter GB, Caldwell J. Antibodies to *Streptococcus mutans* and immunoglobulin levels in children with dental caries. Arch Oral Biol 1978;23:1061–7.
24. Everhart DL, Rothenberg K, Carter WH, Klapper B. The determination of antibody to *Streptococcus mutans* serotypes in saliva for children ages 3 to 7 years. J Dent Res 1978;57:631–5.
25. Bamman LL, Gibbons RJ. Immunoglobulin A antibodies reactive with *Streptococcus mutans* in saliva of adults, children, and predentate infants. J Clin Microbiol 1979;10:538–43.
26. Gahnberg L, Smith DJ, Taubman MA, Ebersole JL. Salivary IgA antibody to glucosyltransferase of oral microbial origin in children. Arch Oral Biol 1985;30:551–6.
27. Camling E, Köhler B. Infection with the bacterium *Streptococcus mutans* and salivary IgA antibodies in mothers and their children. Arch Oral Biol 1987;32:817–23.
28. Tenovuo J, Lehtonen O-P, Aaltonen AS. Serum and salivary antibodies against *Streptococcus mutans* in young children with and without detectable oral *S. mutans*. Caries Res 1987;21:289–96.
29. Smith DJ, Taubman MA, Ebersole JL. Ontogeny and senescence of salivary immunity. J Dent Res 1987;66:451–6.
30. Tenovuo J. The microbiology and immunology of dental caries in children. Rev Med Microbiol 1991;2:76–82.
31. Berkenbilt DA, Bahn AN. Development of antibodies to cariogenic streptococci in children. J Am Dent Assoc 1971;83: 332–7.
32. Luo Z, Smith DJ, Taubman MA, King WF. Cross-sectional analysis of serum antibody to oral streptococcal antigens in children. J Dent Res 1988;67:554–60.
33. Tenovuo J, Lehtonen O-P, Aaltonen AS. Caries development in children in relation to the presence of mutans streptococci in dental plaque and of serum antibodies against whole cells and protein antigen I/II of *S.mutans*. Caries Res 1990;24:59–64.
34. Challacombe SJ, Russell MW, Hawkes JE, Bergmeier A, Lehner T. Passage of immunoglobulins from plasma to the oral cavity of rhesus monkeys. Immunology 1978;35:923–30.
35. Riviere GR, Bresnick SD, Miller JN. Saliva increases serum IgG retention on *Streptococcus mutans*. Oral Microbiol Immunol 1988;3: 129–33.
36. Aaltonen AS, Tenovuo J, Lehtonen O-P, Saksala R, Meurman O. Serum antibodies against oral *Streptococcus mutans* in young children in relation to dental caries and maternal close-contacts. Arch Oral Biol 1985;30:331–5.
37. Aaltonen AS, Tenovuo J, Lehtonen O-P. Increased dental caries activity in preschool children with low baseline levels of serum IgG antibodies against the bacterium *Streptococcus mutans*. Arch Oral Biol 1987;32:55–60.
38. Aaltonen AS, Tenovuo J, Lehtonen O-P. Maternal caries incidence and salivary close-contacts with children affect antibody levels to *Streptococcus mutans* in children. Oral Microbiol Immunol 1990;5:12–8.
39. Mestecky J, Russell MW, Jackson S, Brown TA. The human IgA system: a reassessment. Clin Immunol Immunopathol 1986;40: 105–14.
40. Marcotte H, Lavoie MC. Oral microbial ecology and the role of salivary immunoglobulin A. Microbiol Mol Biol Rev 1998; 62: 71–109.
41. Hanson LÅ, Björkander J, Oxelius V-A. Selective IgA deficiency. In: Chandra RK, editor. Primary and secondary immunodeficiency disorders. New York: Churchill Livingstone; 1983. p. 62–84.
42. Adner PL, Ekdahl H, Sandberg LO. Studies on salivary IgA. II. Salivary IgA in cases deficient of serum IgA. Swed Dent J 1974;67:349–52.
43. Koskinen S. Long-term follow-up of health in blood donors with primary selective IgA deficiency. J Clin Immunol 1996;16:165–70.
44. Lehtonen O-P, Tenovuo J, Aaltonen AS, Vilja P. Immunoglobulins and innate factors of immunity in saliva of children prone to respiratory infections. Acta Pathol Microbiol Immunol Scand [C] 1987;95:35–40.
45. Cole MF, Arnold RR, Rhodes MJ, McGhee JR. Immune dysfunction and dental caries. A preliminary report. J Dent Res 1977;56:198–204.
46. Legler DW, McGhee JR, Lynch DP, Mestecky JF, Schaeffer ME, Carson J, et al. Immunodeficiency disease and dental caries in man. Arch Oral Biol 1981;26:905–10.
47. Brown LR, Mackler BF, Levy BM, Wright TE, Handler SF, Moylan JS, et al. Comparison of the plaque microflora in immunodeficient and immunocompetent dental patients. J Dent Res 1979;58:2344–52.
48. Robertson PB, Mackler BF, Wright TE, Levy BM. Periodontal

status of patients with abnormalities of the immune system. II. Observations over a 2-year period. J Periodontol 1980;51:70–3.
49. Norhagen Engström G, Engström P-E, Hammarström L, Smith CIE. Oral conditions in individuals with selective immunoglobulin A deficiency and common variable immunodeficiency. J Periodontol 1992;63:984–9.
50. Dahlén G, Björkander J, Gahnberg L, Slots J, Hanson L-Å. Periodontal disease and dental caries in relation to primary IgG subclass and other humoral immunodeficiencies. J Clin Periodontol 1993;20:7–13.
51. Kirstilä V, Tenovuo J, Ruuskanen O, Nikoskelainen J, Irjala K, Vilja P. Salivary defense factors and oral health in patients with common variable immunodeficiency. J Clin Immunol 1994; 14:229–36.
52. Arnold RR, Pruitt KM, Cole MF, Adamson JM, McGhee JR. Salivary antibacterial mechanisms in immunodeficiency. In: Kleinberg I, Ellison SA, Mandel ID, editors. Saliva and dental caries (a special supplement to Microbiology Abstracts): proceedings of a workshop on saliva and dental caries, 5–7 June 1978, School of Dental Medicine, Health Sciences Center, State University of New York at Stony Brook. New York: IRL Press; 1979. p. 449–62.
53. Norhagen Engström C, Engström P-E, Hammarström L, Söder P-Ö, Smith CIE. Immunoglobulin levels in saliva in individuals with selective IgA deficiency: compensatory IgM secretion and its correlation with HLA and susceptibility to infections. J Clin Immunol 1989;9:279–86.
54. Järvinen H, Tenovuo J, Huovinen P. In vitro susceptibility of *Streptococcus mutans* to chlorhexidine and six other antimicrobial agents. Antimicrob Agents Chemother 1993;37:1158–9.
55. Hocini H, Iscaki S, Bouvet J-P, Pillot J. Unexpectedly high levels of some presumably protective secretory immunoglobulin A antibodies to dental plaque bacteria in salivas of both caries-resistant and caries-susceptible subjects. Infect Immun 1993; 61:3597–604.
56. Rudney JD. Does variability in salivary protein concentrations influence oral microbial ecology and oral health. Crit Rev Oral Biol Med 1995;6:343–67.
57. Rudney JD, Smith QT. Relationships between levels of lysozyme, lactoferrin, salivary peroxidase and secretory immunoglobulin A in stimulated parotid saliva. Infect Immun 1985; 49:469–75.
58. Rudney JD. Relationships between human parotid saliva lysozyme, lactoferrin, salivary peroxidase and secretory immunoglobulin A in a large sample population. Arch Oral Biol 1989;34:499–506.
59. Tenovuo J, Jentsch H, Soukka T, Karhuvaara L. Antimicrobial factors of saliva in relation to dental caries and salivary levels of mutans streptococci. J Biol Buccale 1992;20:85–90.
60. Tenovuo J, Moldoveanu Z, Mestecky J, Pruitt KM, Månsson-Rahemtulla B. Interaction of specific and innate factors of immunity: IgA enhances the antimicrobial effect of the lactoperoxidase system against *Streptococcus mutans*. J Immunol 1982;128:726–31.
61. Lassiter MO, Newsome AL, Sams LD, Arnold RR. Characterization of lactoferrin interaction with *Streptococcus mutans*. J Dent Res 1987;66:480–5.
62. Soukka T, Lumikari M, Tenovuo J. Combined inhibitory effect of lactoferrin and lactoperoxidase system on the viability of *Streptococcus mutans*, serotype c. Scand J Dent Res 1991;99:390–6.
63. Soukka T, Lumikari M, Tenovuo J. Combined bactericidal effect of human lactoferrin and lysozyme against *Streptococcus mutans* serotype c. Microb Ecol Health Dis 1991;4:259–64.
64. Murakami Y, Nagata H, Amano A, Takagaki M, Shizukuishi S, Tsunemitsu A, et al. Inhibitory effects of human salivary histatins and lysozyme on coaggregation between *Porphyromonas gingivalis* and *Streptococcus mitis*. Infect Immun 1991;59:3284–6.
65. Tenovuo J. Oral defense factors in the elderly. Endodontol Dent Traumatol 1992;8:93–8.
66. Tenovuo J, Söderling E. Chemical aids in the prevention of dental diseases in the elderly. Int Dent J 1992;42:355–64.
67. Nilner K, Vassilakos N, Birkhed D. Effect of a buffering sugar-free lozenge on intraoral pH and electrochemical action. Acta Odontol Scand 1991;49:267–72.
68. Tenovuo J, Hurme T, Ahola A, Svedberg C, Ostela I, Lenander-Lumikari M, et al. Release of cariostatic agents from a new buffering fluoride- and xylitol-containing lozenge to human whole saliva *in vivo*. J Oral Rehabil 1997;24:325–31.
69. Tenovuo J, Lumikari M, Soukka T. Salivary lysozyme, lactoferrin and peroxidases: antibacterial effects on cariogenic bacteria and clinical applications in preventive dentistry. Proc Finn Dent Soc 1991;87:197–208.
70. Pruitt KM, Reiter B. Biochemistry of peroxidase system: antimicrobial effects. In: Pruitt KM, Tenovuo J, editors. The lactoperoxidase system, chemistry and biological significance. New York: Marcel Dekker; 1985. p. 143–78.
71. Tenovuo J, Mäkinen KK, Sievers G. Antibacterial effect of lactoperoxidase and myeloperoxidase against *Bacillus cereus*. Antimicrob Agents Chemother 1985;27:96–101.
72. Tenovuo J, Anttila O, Lumikari M, Sievers G. Antibacterial effect of myeloperoxidase against *Streptococcus mutans*. Oral Microbiol Immunol 1988;3:68–71.
73. Tenovuo J. Salivary peroxidase. In: Everse J, Everse KE, Grisham M, editors. Peroxidases: chemistry and biology. Vol I. Boca Raton (FL): CRC Press; 1991. p. 181–97.
74. Tenovuo J, Månsson-Rahemtulla B, Pruitt KM, Arnold R. Inhibition of dental plaque acid production by the salivary lactoperoxidase antimicrobial system. Infect Immun 1981;34: 208–14.
75. Pruitt KM, Tenovuo J, Fleming W, Adamson M. Limiting factors for the generation of hypothiocyanite ion, an antimicrobial agent in human saliva. Caries Res 1982;16:315–23.
76. Bánóczy JB, Dombi C, Czegledly A, Sari K. A clinical study with lactoperoxidase- containing gel and toothpaste in patients with dry mouth syndrome. J Clin Dent 1994;5:65–9.
77. van Steenberghe D, Van den Eynde E, Jacobs R, Quirynen M. Effect of a lactoperoxidase containing toothpaste in radiation-induced xerostomia. Int Dent J 1994;44:133–8.
78. Mulligan R, Navazesh M, Slots J. Antibacterial activity of an alcohol-free mouthwash with enzymes [abstract]. J Dent Res 1992;71:156.
79. Kirstilä V, Lenander-Lumikari M, Söderling E, Tenovuo J. Effects of oral hygiene products containing lactoperoxidase, lysozyme and lactoferrin on the composition of whole saliva and on subjective oral symptoms in patients with xerostomia. Acta Odontol Scand 1996;54:391–7.
80. Lenander-Lumikari M, Loimaranta V, Hannuksela S, Tenovuo J, Ekstrand J. Combined inhibitory effect of fluoride and hypothiocyanite on the viability and glucose metabolism of *Streptococcus mutans*, serotype c. Oral Microbiol Immunol 1997; 12:231–5.
81. Czerkinsky C, Russell MW, Lycke N, Lindblad M, Holmgren J. Oral administration of a streptococcal antigen coupled to cholera toxin B subunit evokes strong antibody responses in salivary glands and extramucosal tissues. Infect Immun 1989;57:1072–7.
82. Redman TK, Harmon CC, Lallone RL, Michalek SM. Oral immunization with recombinant *Salmonella typhimurium* expressing surface protein antigen A of *Streptococcus sobrinus:* dose response and induction of protective humoral responses in rats. Infect Immun 1995;63:2004–11.
83. Lehner T, Caldwell J, Smith R. Local passive immunization by monoclonal antibodies against streptococcal antigen I/II in the prevention of dental caries. Infect Immun 1985;50:796–9.
84. Filler SJ, Gregory RL, Michalek SM, Katz J, McGhee JR. Effect of immune bovine milk on *Streptococcus mutans* in human dental plaque. Arch Oral Biol 1991;36:41–7.
85. Hatta H, Tsuda K, Ozeki M, Kim M, Yamamoto T, Otake S, et al. Passive immunization against dental plaque formation in

humans: effect of a mouth rinse containing egg yolk antibodies (IgY) specific to *Streptococcus mutans*. Caries Res 1997;31:268–74.
86. Loimaranta V, Tenovuo J, Virtanen S, Marnila P, Syväoja E-L, Tupasela T, et al. Generation of bovine immune colostrum against *Streptococcus mutans* and *Streptococcus sobrinus* and its effect on glucose uptake and extracellular polysaccharide formation by mutans streptococci. Vaccine 1997;15:1261–8.
87. Ma JK-C, Lehner T, Stabila P, Fux CI, Hiatt A. Assembly of monoclonal antibodies with IgG1 and IgA heavy chain domains in transgenic tobacco plants. Eur J Immunol 1994;24:131–8.
88. Loimaranta V, Carlén A, Olsson J, Tenovuo J, Syväoja E-L, Korhonen H. Concentrated bovine colostral whey proteins from *Streptococcus mutans/Strep. sobrinus* immunized cows inhibit the adherence of *Strep. mutans* and promote aggregation of mutans streptococci. J Dairy Res. In press 1998.
89. Loimaranta V, Tenovuo J, Korhonen H. Combined inhibitory effect of bovine immune whey and peroxidase-generated hypothiocyanite against glucose uptake by *Streptococcus mutans*. Oral Microbiol Immunol. In press 1998.