

Effect of acid and alkali formation on pH in the dental biofilm with reference to caries

Akademisk avhandling

Som för avläggande av odontologie doktorsexamen vid Sahlgrenska akademien,
Göteborgs universitet kommer att offentligen försvaras i Föreläsningssal F3,
Medicinaregatan 12E, Göteborg, fredagen den 15 juni 2018, kl. 13.00

av

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Leg tandläkare

Fakultetsopponent:

Professor Julia Davies

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Avhandlingen är av sammanläggningstyp och baseras på följande fem delarbeten:

I. Carlén A, Hassan H, Lingström P. The 'strip method': a simple method for plaque pH assessment. *Caries Res* 2010;44:341-344.

II. Hassan H, Lingström P, Carlén A. Plaque pH in caries-free and caries-active young individuals before and after frequent rinses with sucrose and urea solution. *Caries Res* 2015;4:18-25.

III. Dahlén G, Hassan H, Blomqvist S, Carlén A. Rapid urease test (RUT) for evaluation of urease activity in oral bacteria *in vitro* and in supragingival dental plaque *ex vivo*. *BMC Oral Health*. 2018;18:89-95.

IV. Hassan H, Ghali L, Wildeboer D, Sarwar S, Lingström P, Carlén A. Interproximal *in situ* plaque pH in relation to caries before and after short-term use of 1.5% arginine toothpaste. In manuscript.

V. Hassan H, Bjondahl F, Olofsson R, Dahlén G, Carlén A. Acid formation of supragingival dental biofilm bacteria isolated from caries-free and caries-active individuals - an *in vitro* study. In manuscript.

**SAHLGRENSKA AKADEMIN
INSTITUTIONEN FÖR ODONTOLOGI**
Göteborg 2018



UNIVERSITY OF
GOTHENBURG

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Abstract

Dental caries is a common multifactorial disease where a frequently low pH in the dental biofilm (dental plaque) plays an important role for caries occurrence and progress. The plaque-pH is lowered by acid formation from sugars by biofilm bacteria that also help to restore the pH by alkali formation from urea and the amino acid arginine. Despite the importance of pH for caries to occur, *in vivo* studies comparing the dental plaque-pH after acid and alkali formation are scarce. This may be due to the lack of methods that easily can be applied in the clinic. In this thesis, the dental plaque-pH after acid and alkali formation *in vivo* in relation to the individual caries status were examined. Two chair-side methods were also developed; the 'strip method' to measure *in situ* the interproximal, supragingival plaque-pH (Study I) and a microtiter plate format of RUT (Rapid Urease Test) to grade bacterial urease activity *in vitro* and *ex vivo* (Study III). Hopefully studies on pH and alkali formation in the dental plaque can contribute to finding measures for assessment of the individual caries risk. Studies on plaque-pH after sugar and urea challenges, pre- and post-adaptation periods to respectively acid and alkali formation from 1-week daily rinses with sucrose and urea, were examined in caries-free and caries-active individuals (Study II). Furthermore, the number of acid tolerant bacteria was examined as well as the ability of isolated acid tolerant bacteria to form acid from sugars and sugar alcohols *in vitro* (Study II, V). The pH response to a sugar challenge after 6-week usage of fluoride toothpaste with arginine was also examined in relation to caries (Study IV). Similar plaque-pH values and Stephan curves were obtained using the 'strip method' and the well-known 'microtouch method' before and up to 60 min following a sugar challenge (Study I). RUT showed a strong *in vitro* urease activity for the well-known urease active *Helicobacter pylori* and for strains of *Haemophilus parainfluenzae* but not for the more common plaque bacteria *Actinomyces spp.* and *Streptococcus mitis* (Study III). A higher urease activity in plaque at sites in the lower front compared to plaque at other sites was found as well (Study III, IV). Adaptation to acid formation resulted in lower plaque-pH after a sugar challenge and an increased number of acid tolerant bacteria in caries-free (CF) individuals (Study II). Adaptation to alkali formation resulted in somewhat higher pH values after a urea challenge in caries-active (CA) individuals (Study II). In concordance, acid formation was numerically increased in bacteria isolated from the CF group after acid adaptation and decreased in isolates from the CA group after alkali adaptation (Study V). In CA but not CF individuals, the usage of fluoride toothpaste with arginine resulted in increased plaque pH-values as well as increased saliva buffer capacity and pH (Study IV). It can be concluded that the 'strip method' and 'RUT' are applicable as chair-side methods, for the assessment of plaque acidogenicity and urease activity, respectively. Adaptation to sugar increased the acid formation and decreased the pH in the dental plaque in caries-free individuals. Adaptation to urea and arginine decreased the acid formation and increased pH in the dental biofilm in caries-active individuals

Keywords: acid, alkali, biofilm, caries, pH, supragingival plaque

ISBN 978-91-629-0512-5 (PRINT)

ISBN 978-91-629-0511-8 (PDF)

<http://hdl.handle.net/2077/5597>