ABSTRACT

Objectives: The Rapid Automated Bacterial Impedance Technique (RABIT) monitors real time bacterial activity by measuring the carbon dioxide released during bacterial metabolism. It is routinely used by industries as well as health agencies to screen for microbial contamination and / or to estimate product shelf life. This research validates the use of this technique to measure the changes in bacterial activity in morning saliva, post use of the Triclosan containing Blend-a-Med Medic Nacht toothpaste.

Methods: A randomized, double blind, crossover study was conducted with 20 volunteers. After a "washout period" of one week using a standard fluoride toothpaste and manual toothbrush, volunteers were divided into two groups and were asked to use the test products: Blend-a-Med Medic Nacht (containing Triclosan) and Blend-a-Med Medic Weiss (control without an antimicrobial) for evening brushing. Saliva samples were collected the following morning. Then after a second "washout period" of 2 days on standard dentifrice, the same process was repeated with a treatment crossover. The samples were analyzed by the Zone of diffusion assay to detect the presence of biologically active Triclosan. Also changes in bacterial metabolism were quantified using RABIT.

Results: The morning saliva samples collected after Triclosan treatment showed a significantly higher antimicrobial activity as measured by the Zone of diffusion assay compared to the control leg (treatment 72.2% ± 10.56%, control leg 35.3% ± 11.59%, p=0.0033). Also a significant reduction in bacterial metabolism was observed using the Impedance technique (Average AUC: control 309.2, treatment 273.5, p=0.0153).

Conclusion: Due to the reduction in salivary flow rate, the mouth is more susceptible to bacterial activity at night. Use of the toothpaste containing antimicrobial such as Blend-a-Med Medic Nacht can help control bacterial activity until morning brushing as demonstrated by Impedance technique and zone of diffusion assay.

INTRODUCTION AND PURPOSE

This study was designed to validate the application RABIT methodology for monitoring real time bacterial metabolism in saliva samples. Here we assess the potential of the technique to monitor the overnight efficacy of a Triclosan containing formulation in parallel with traditionally used zone of diffusion technique. RABIT technology is versatile in that it allows the monitoring of: the performance of actives which may have a diffusion limitation (e.g. Zinc, CPC) traditionally agar techniques. Blend-a-Med Medic Nacht (MN) dentifrice with overnight protection (collected ~10 hours post brushing with Medic Nacht) – showing the antibacterial diffusion inhibiting growth of the S. aureus pre-seeded on the plate.

The samples were all divided and randomized so that Right (R) or Left (L) cheek, Lower (L) or Upper (U) gum and saliv (2ml split into 4x 5ml) were analysed by both RABIT and Zone of diffusion testing. Scientists were 'blinded' to the treatment randomisation schedule until post statistical analysis.

Zone of Diffusion Assay: For testing presence of residual Triclosan, samples were placed onto agar pre-seeded with diluted (1:10) S.aureus (239.40*). The samples were then incubated for 18 hours and the following day the plates were checked for growth of S. aureus. The distance between the edge of the colony and the edge of the sample was measured and a percentage of growth inhibition was calculated.

RESULTS

A clear ‘zone’ can be seen around the saliva sample collected ~10 hours post brushing with Medic Nacht (Average AUC: control 309.2, treatment 273.5, p=0.0153). This result suggests a significantly higher antimicrobial activity as measured by the Zone of diffusion assay compared to the control leg (treatment 72.2% ± 10.56%, control leg 35.3% ± 11.59%, p=0.0033). Also a significant reduction in bacterial metabolism was observed using the Impedance technique (Average AUC: control 309.2, treatment 273.5, p=0.0153).

CONCLUSION

To our knowledge this is the first time the overnight effect of Triclosan on bacterial metabolism has been profiled using this sensitive technology. Rapid Automated Bacterial Impedance Technique (RABIT) showed that the Triclosan formulation (MN) actively inhibited activity of oral bacteria for up to 14 hours after brushing with the product.

Toothbrushing testing demonstrated that there is biologically active Triclosan present in the morning mouth after brushing the night before.