## **OFDFA Final Report**

Type of Award: Orthodontic Faculty Development Fellowship Award

Name of Principal Investigator: Dr. Karthikeyan Subramani

**Title of Project:** *In vitro* Evaluation of Antimicrobial Activity and Force Decay of Chlorhexidine Hexametaphosphate Nanoparticle Coatings on Orthodontic Elastomeric Chains

Period of AAOF Support: 07-01-16 to 06-30-18

Amount of Funding: \$ 20,000

#### Summary/Abstract:

Objective: The purpose of this *in vitro* study is to evaluate antimicrobial activity of eluate from orthodontic elastomeric chains (OEC) coated with chlorhexidine (CHX) hexametaphosphate (HMP) nanoparticles (NP).

Methods: OECs were coated with CHX-HMP-5 (5mM CHX and HMP), CHX-HMP-1 (1mM CHX and HMP), CHX-5 (5mM CHX) and CHX-1 (1mM CHX) solutions. Uncoated OECs were used as control group. The release of CHX from coated OECs was evaluated for a period of 28 days at time points of day 1, 2, 3, 5, 7, 14, 21 and 28 with a UV spectrophotometer (n=5/group). Antimicrobial property of the eluate collected at day 1 was tested with *Streptococcus mutans (S. mutans)* and *Lactobacillus rhamnosus (L. rhamnosus)* microbial cultures by measuring the zone of inhibition after 4 days (n=3/group). OECs were observed by scanning electron microscope (SEM) to observe surface characteristics between: (1) uncoated and coated OECs and (2) before and after release of CHX. Force decay measurement of all groups (n=20/group) were measured at time points of day 1, 2, 3, 5, 7, 14, 21 and 28 with a Lutron force gauge.

Results: OECs coated with CHX-HMP-5 and CHX-HMP-1 released CHX over 28 days. SEM analysis showed that OECs were coated with NPs. CHX-HMP-5, CHX-HMP, CHX-5 and CHX-1 groups exhibited zones of inhibition of *S. mutans* (14 cm<sup>2</sup>, 8 cm<sup>2</sup>, 10 cm<sup>2</sup>, 7.5 cm<sup>2</sup>) and L. rhamnosus (6.1 cm<sup>2</sup>, 5 cm<sup>2</sup>, 6.1 cm<sup>2</sup>, 5.9 cm<sup>2</sup>. The largest loss of force occurred over the first 24 hours followed by a relative plateau over the remaining experimental protocol. There was no significant differences between group means at the following time points: 7 days, 14 days and 28 days as determined by a one-way ANOVA (p>0.05).

Conclusion: CHX-HMP NP coated OEC releases CHX in a sustained manner over a period of 28 days capable of inhibiting the growth of *S. mutans* and *L. rhamnosus*. The coating did not affect the force decay of the OECs. Use of such coating on OECs can exhibit antibacterial effect and reduction of biofilm buildup and prevent white spot lesions and offers promising clinical applications which needs to be explored further.

## **Response to the following questions:**

## 1. Were the original, specific aims of the proposal realized?

Yes.

## 2. Were the results published?

The study has resulted in the following 2 manuscripts which are soon to be submitted to orthodontic/biomaterials journals after final review by co-authors. The AAOF support has been acknowledged in both manuscripts.

(i) **Karthikeyan Subramani**, Han Na Seo, Prashanti Bollu, Kishore Chaudhry, Jian-Feng Zhang, Kenneth Rosenthal. *In vitro* Evaluation of Antimicrobial Activity of Chlorhexidine Hexametaphosphate Nanoparticle Coatings on Orthodontic Elastomeric Chains.

(ii) **Karthikeyan Subramani**, Joseph Dougherty, Prashanti Bollu, Kishore Chaudhry, Jian-Feng Zhang, Kenneth Rosenthal. *In vitro* Evaluation of Force Decay of Chlorhexidine Hexametaphosphate Nanoparticle Coated Orthodontic Elastomeric Chains.

#### 3. Have the results of this proposal been presented?

Yes. I received the prestigious **"Milo Hellman Award"** from AAO on 5<sup>th</sup> May 2019 at the AAO Annual Meeting, Los Angeles CA for this research project. The findings have been presented as posters at the local and national research conferences. The AAOF support has been acknowledged in all oral and poster presentations.

(i) Poster presented at Roseman University 2018 Research Symposium, Henderson, NV on March 14, 2018. **Karthikeyan Subramani**, Han Na Seo, Prashanti Bollu, Kishore Chaudhry, Jian-Feng Zhang, Kenneth Rosenthal. *In vitro* Evaluation of Antimicrobial Activity of Chlorhexidine Hexametaphosphate Nanoparticle Coatings on Orthodontic Elastomeric Chains.

(ii) Poster presented at Roseman University 2018 Research Symposium, Henderson, NV on March 14, 2018. **Karthikeyan Subramani**, Joseph Dougherty, Prashanti Bollu, Kishore Chaudhry, Jian-Feng Zhang, Kenneth Rosenthal. *In Vitro* Evaluation of Force Decay of Chlorhexidine Hexametaphosphate Nanoparticle Coated Orthodontic Elastomeric Chains.

(iii) Poster (#: 1083) presented at 2018 AADR Annual Meeting Fort Lauderdale, Fl on March 23, 2018. **Karthikeyan Subramani**, Han Na Seo, Joseph Dougherty, Sarah Rafo, Prashanti Bollu, Kishore Chaudhry, Kenneth Rosenthal. *In vitro* Evaluation of Antimicrobial Activity and Force Decay of Chlorhexidine Hexametaphosphate Nanoparticle Coatings on Orthodontic Elastomeric Chains.

(iv) Poster presented at 2018 PCSO Annual Meeting, Monterey, CA on October 12, 2018. **Karthikeyan Subramani**, Joseph Dougherty, Prashanti Bollu, Kishore Chaudhry, Jian-Feng Zhang,

Kenneth Rosenthal. *In Vitro* Evaluation of Force Decay of Chlorhexidine Hexametaphosphate Nanoparticle Coated Orthodontic Elastomeric Chains.

(v) **"Milo Hellman Award"** lecture on 5<sup>th</sup> May 2019 at the AAO Annual Meeting, Los Angeles CA. **Karthikeyan Subramani**. *In Vitro* Evaluation of Force Decay of Chlorhexidine Hexametaphosphate Nanoparticle Coated Orthodontic Elastomeric Chains.

# 4. To what extent have you used, or how do you intend to use, AAOF funding to further your career?

I am humbled to be the recipient of the prestigious 2019 **"Milo Hellman Award"** from the AAO for this research project. The funding has allowed me make significant progress in my area of research interest (nanomaterials), mentor two orthodontic residents on this project. It has also helped me to attend continuing education courses, which has helped me to apply the knowledge gained from these CE courses in my clinical and didactic teaching to the residents. Part of the OFDFA has been utilized as a salary supplement and I thank the AAOF for their generous support. I look forward to apply for the Biomedical Research award during the next awards cycle. I certainly believe that the continued support from AAOF will be immensely helpful to achieve my academic career goals.