



FACULTY OF  
**DENTISTRY**  
Research Report  
**07/08**

## CONTENTS

- 01** / Foreword
- 02** / Faculty Research Strategy
- 03** / Strategic Research Initiatives
- 07** / Graduate Research Programmes
- 08** / Graduate Coursework Programmes
- 10** / Undergraduate Research Opportunities Programme (UROP)
- 12** / Research Highlights
- 19** / Completed Research Projects in FY 2007/08
- 25** / New Research Projects awarded for funding in FY 2007/08
- 31** / Research Collaborations
- 32** / Research Achievements
- 35** / Research Statistics

## Foreword by Vice Dean (Research)

As Faculty of Dentistry continues its relentless journey in the concerted pursuit of academic excellence, we now have the comrade in arms joining the allies. The inception of NUHS in January 2008 enables capitalization on this immense opportunity and presumably allows the synergy with our sibling medical clinical faculty and NUH, to better engage biomedical research, embracing the national thrust of engineering knowledge-based economic restructuring.

Nevertheless, Faculty's research, traditionally interdisciplinary in collaboration with institutional peers and/or industrial players, remains pretty much PI-lead and driven to improve the oral and craniofacial health with the intent to translate into enhanced quality delivery of holistic care and well being.

In alignment with this mission, the 3 research initiatives [CCRB, DBBP & CCRU] leveraging on the Faculty's existing research strengths have taken off in the past year serving as platforms to facilitate development of various exciting thematic research programs in the pipelines. The allocated additional research space of 92 square meters at level 3 of DSO building has made feasible the reorganization of the research infrastructure and the establishment of a new core research facility, the Oral Tissue Repository [OTR], to optimally support these budding strategic research activities.

Under the wings of these 3 provost-funded strategic research initiatives, Faculty has formulated plans to advance technologies in the set up of unique oral biofilm research program and state-of-the-art fluoride laboratory as well as imploring the capability of the ethically less controversial, readily accessible and operationally less invasively harvested oral tissue-derived adult stem cells that have been reported to be of potential for wide clinical exploitation.

As we take comfort in the staff contribution and steady progress to-date, our articulated research journey ahead is indeed promising, given as well the addition of new faculty stakeholders to our small faculty capacity. Yet, the quest of acquiring research triumphs would certainly be faced with a myriad of challenges including the intricate research ethical issue that need to be delicately balanced. Faculty would just have to keep persevering, stay focused and power up the aspiration to achieving the not impossible peaks of research excellence.

*Associate Professor Varawan Sae-Lim  
Vice Dean (Research)*



**Faculty Vision:** To be a Dental Institution of International Distinction

**Faculty Mission:** To improve Oral Health through Academic Excellence, High Impact Research and Quality Clinical Service



**Research Mission:** To improve Oral, Dental and Craniofacial Health and the Delivery of Clinical Care



**Strategic Research Initiatives**



**Centre for Craniofacial  
Regenerative Biology (CCRB)**



**Dental Biophotonics and  
Biomaterials Programme (DBBP)**



**Craniofacial Clinical  
Research Unit (CCRU)**



## Centre for Cranio-facial & Regenerative Biology (CCRB)

Director: Assoc Prof Varawan Sae-Lim

### Overview:

In addition to the established research programs it facilitates under its wing among which includes the embryonic stem cells and oro-facial pain research, CCRB's strategy was to focus in the past year, the set up of key infrastructure and capability to allow nurturing of the budding research that harness oral tissue-derived adult stem cells. The initiative planned to commence with the capability of dental pulp stem cell aims to advance cell-based research exploitable in cranio-facial regenerative biology and other translational research entities.

### Progress in AY 2007/08:

With the approval of the Institutional Review Board, CCRB has embarked on the set up of Oral Tissue Repository [OTR] as a core research facility following International Society for Biological and Environmental Repositories (ISBER)'s Best Practices for Repositories. CCRB-OTR hopes to expedite the collection, from routine surgical procedures, the 'to-be-discarded' human oral tissues [dental pulp, gingiva, periodontal tissue, oral mucosa, bone, etc], as well as banking and retrieval of these biological tissue materials for specific research utilization. CCRB-OTR has also conducted the risk assessment and obtained the approval by Institutional Biosafety Committee (IBC).

### CCRB Progress Overview

CCRB maintained an overall steady progress in the past review period with cumulative achievements generally exceeding the targeted deliverables. To highlight, these included 2 research papers which won the Senior Travel Award (First Runner Up) at the 21st ASM, International Association for Dental Research / South-East Asian Division judged by the 3 international renowned researchers as well as the Best Clinical Research Project at the 18th South East Asian Association for Dental Education. One CCRB research team is also in the process of filing a potential cell-based therapy Invention Disclosure.

## Dental Biophotonics and Biomaterial Programme (DBBP)

Director: Assoc Prof Stephen Hsu

### Overview:

In this fiscal year, the biophotonic group has advanced with laser technologies to address scientific questions related to caries and root canal infection. The biomaterial group has proceeded to characterize the implant stability, remineralizing effect of glass ionomer cement, and biocompatibility of novel dental implant. The bio-imaging group has continued the computational and statistical modeling of morphological characterization of human head/teeth.

### Progress in FY 2007/08:

The number of research grants and awards achieved under DBBP, as listed in other parts of this report, has well exceeded the targeted projection. The thematic progress is summarized below:

- **Tooth-biofilm Interaction:**
  - o Dr. Anil's group has successfully applied optical tweezers (1064 nm) and Equipartition theorem-based stiffness measurements to quantify the adhesion force between bacteria and collagen of the treatment group, suggesting that the presence of calcium hydroxide or EDTA treatment may increase the adhesion force of E. faecalis to type-I collagen. Coupled with Light Activated

Disinfection (LAD), the MB in emulsion (PF4) was found to be the most effective photosensitization formulation for photooxidation, generation of singlet-oxygen to destroy biofilm bacteria. Working along this line, the student team has won the NUS Outstanding Undergraduate Research (NUS-OUR) Award. The other NUS-OUR winning team, supervised by A/P Hsu, has elucidated the potential cariostatic effect of Yakult™, a popular soft drink with pH=3.5, sucrose, glucose, and lactobacillus. Against the common belief, the acid production of the biofilm/plaque was found to be significantly reduced among the young adults with moderate caries risk after the 2-week daily consumption of Yakult™.

- **Laser-Tissue Interaction:**

- o A/P Hsu's team has successfully quantified site-specific micro-diffusion in enamel, with fluorescent techniques coupled with laser scanning confocal microscopy. The results revealed significant reduction of enamel diffusion after laser treatment, substantiating the "Organic blocking theory" proposed by A/P Hsu's team as one of the major mechanisms involved in the laser-induced caries prevention.

- **Bio-Imaging Research:**

- o A/P Foong's team continues to build accurate patient-specific models of the human face and dentition using the imaging modalities of Magnetic Resonance Imaging, Computed Tomography and surface scanning that are used for training and clinical decision making. A current project, funded by the Singapore Bio-Imaging Consortium, seeks to create these models for orthodontic treatment and jaw surgery without the use of ionizing radiation.

- **Biomaterial Research:**

- o A/P Keson Tan's group has continued to assess critical factors influencing the dental implant stability and published one article using finite element analysis to quantify the potential effects. The team has secured one ARF grant this year. Besides a start-up grant, A/P Ngo's team has published their work investigating the effects of environmental degradation and remin/demineralization cycling on physicochemical properties of glass ionomers. A/P Cao Tong's team has successfully secured a grant and evaluated the biocompatibility of novel 3-dimensional printing titanium dental implant with hESCs.

## Craniofacial Clinical Research Unit (CCRU)

**Director: Assoc Prof Jennifer Neo**

### Activities for CCRU 2007/2008

Dental diseases are amongst the most common health problems in the community, both dental caries and periodontal diseases are highly prevalent and require on-going management. Even though both diseases are non life-threatening, their repetitive nature creates a large burden on the individual and community. This burden can manifest as financial problems and a poorer quality of life and is amplified in the older individuals and medically compromised. The range of research themes to be pursued is designed to fulfill the stated mission statement of the Faculty of Dentistry (FOD) "to improve oral and cranial health and the delivery of care". It will lead to both prevention of dental disease and maintenance of oral function contributing to quality of life for the older Singaporeans.

One of the CCRU's vision is to become the regional knowledge base and centre of excellence in the field of clinical cariology and the three major research platforms which will be used are: artificial mouth biofilm, fluoride laboratory and digital data mining. Once the clinical protocols are established, Minimal Intervention will be the platform to dissipate the information to the profession and population at large.



To lay the foundation, the FOD hosted the following world experts in the fields of cariology, fluoride research and gerodontology to Singapore:

- London Dental Institute, London, United Kingdom
- A/Prof. Chris Sissons, University of Otago, Wellington, New Zealand
- Prof. Ron Ettinger, University of Iowa, Iowa, USA
- A/Prof. Jane Chalmers, University of Iowa, Iowa, USA
- Prof. Conrad Naleway, Loyola University, Illinois, USA
- Dr. Jim Grisdale, University of British Columbia, Vancouver, Canada

The following researchers also visited the FOD to discuss collaboration:

- Dr. Duncan Yu, R&D, Johnson & Johnson
- Dr. Aaron Pfarrer, R&D, 3M Espe



Dr Aaron M Pfarrer with his 3M team with Prof Neo

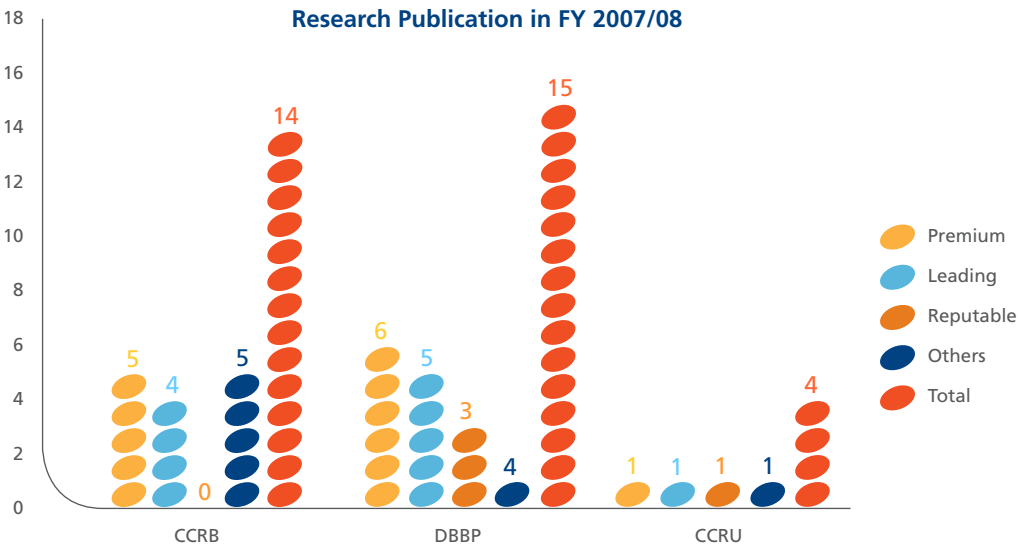
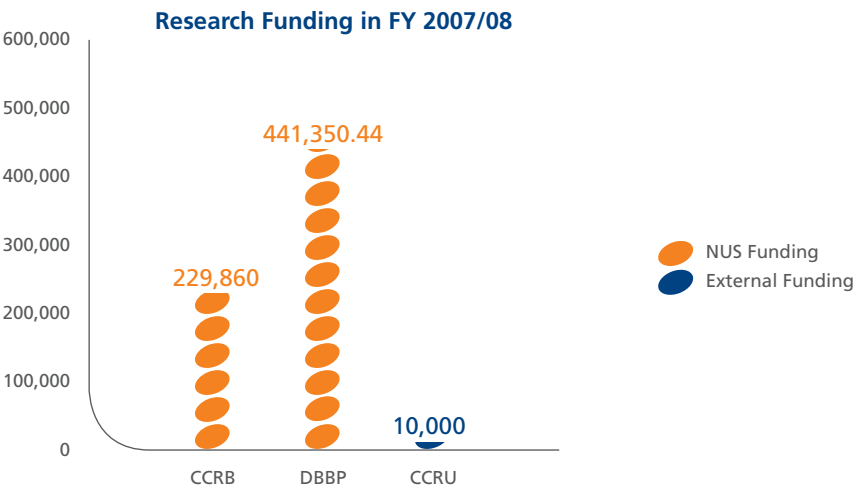
Special grants, from the RI scheme, allowed the set up of a state-of-the-art fluoride laboratory. The aim is to facilitate translational research and to position Singapore as a fluoride reference centre in Southeast Asian region. Currently, there are no reliable testing centres, in the region, the FOD facility will act as a 'gold-standard' against which other laboratories can standardize their methods and equipment to ensure that they are able to provide accurate results. With a fluoride laboratory, FOD can be a regional reference centre where fluoride in food (e.g. salt, milk), water, fingernails, saliva, blood, urine, toothpaste can be assayed from samples sent from neighboring countries.

The effort in developing clinical cariology research by FOD was recognized by industry and one of the first companies to lend their support was 3M ESPE with the donation of research equipment. Thanks to this support, FOD is now equipped with the only Quantitative Light Fluorescent machine in the region.

The following research projects are ongoing:

- Effects of TCP and CPP-ACP on the remineralisation of artificial enamel initial lesions.
- Singapore oral microbiota: HNRCT patients, hyposalivation in vitro, impact and treatment
- The remineralisation of demineralised dentin by glass ionomer cement: the effects of apatite forming and non apatite-forming elements from glass ionomer

Research Statistics of the 3 Research Initiatives in FY 2007/08





## List of Conferred MSc and PhD Students in AY 07/08

### Tarun Kumar Maheshwari

<b>Main Supervisor</b>	<b>Degree</b>	<b>Thesis title</b>	<b>Awards won</b>
A/P Varawan Sae-Lim	MSc	Molecular Profile of Periodontal Tissues Following Tooth Replantation	Senior Travel Award (First Runner Up), 21st Annual Scientific Meeting IADR/SEA (Sept 2007)
<b>Department</b>	<b>Nationality</b>		
Restorative Dentistry	India		

### Lu Kai

<b>Main Supervisor</b>	<b>Degree</b>	<b>Thesis title</b>	
A/P Cao Tong	MSc	Assessment of Mitomycin C - Induced Cytotoxicity and Genotoxicity Utilizing Somatic Progenies of Human Embryonic Stem Cells	
<b>Department</b>	<b>Nationality</b>		
Oral and Maxillofacial Surgery	China		

### Pallavi Uppangala

<b>Main Supervisor</b>	<b>Degree</b>	<b>Thesis title</b>	
A/P Yeo Jin Fei	MSc	A Comparative Study of the Biological and Physical Properties of Viscosity Enhanced Root Repair Material (VERRM) and MTA	
<b>Department</b>	<b>Nationality</b>		
Oral and Maxillofacial Surgery	India		

### Sui Lin

<b>Main Supervisor</b>	<b>Degree</b>	<b>Thesis title</b>	
A/P Cao Tong	MSc	Comparison of Different Systems for Chondrogenic Differentiation of Human Embryonic Stem Cells: The Effect of Tgf-Beta 1	
<b>Department</b>	<b>Nationality</b>		
Oral and Maxillofacial Surgery	China		

### Saji George

<b>Main Supervisor</b>	<b>Degree</b>	<b>Thesis title</b>	<b>Awards won</b>
Dr Anil Kishen	Ph.D.	Investigation on the Bacterial Biofilm in Root-Canal Dentine and Elimination by Modified Light Activated Therapy	Lee Foundation Travel Award (2007)
<b>Department</b>	<b>Nationality</b>		
Restorative Dentistry	India		

List of MDS Students Conferred in AY 07/08

(Prosthodontics) Thesis Projects		
Student Name	Residency Intake	Thesis title
Tapan N Koticha	AY2005/06	Accuracy of Fit at Interface of Four Implant-Abutment Systems
Thesis Submission date	Supervisor(s)/ Dental co-supervisor	
2008	A/P Keson Tan	
(Oral and Maxillofacial Surgery) Thesis Projects		
Student Name	Residency Intake	Thesis title
Intekhab Islam	AY2005/06	A Retrospective Analysis of Dental Implant Failure Rates, Complications and Associated Risk Factors
Thesis Submission date	Supervisor(s)/ Dental co-supervisor	
2008	Dr Goh Bee Tin/ A/P Yeo Jin Fei	
Student Name	Residency Intake	Thesis title
Pujan Raj	AY2005/06	Determination of Incidence of Nerve Injury after Mandibular Third Molar Surgery
Thesis Submission date	Supervisor(s)/ Dental co-supervisor	
2008	A/P Yeo Jin Fei/ Dr Aidan Yeo	
(Orthodontics) Thesis Projects		
Student Name	Residency Intake	Thesis title
Boey Pui Yunn	AY2005/06	Soft Tissue Facial Profiling of Orthodontic Patients
Thesis Submission date	Supervisor(s)/ Dental co-supervisor	
2008	Dr Soh Jen Dr Chew Ming Tak	
Student Name	Residency Intake	Thesis title
Lee Ping Geraldine	AY2005/06	Oral Health Related Quality of Life in Orthodontic Patients: A Pilot Study
Thesis Submission date	Supervisor(s)/ Dental co-supervisor	
2008	Dr Soh Jen Dr Chew Ming Tak	
Student Name	Residency Intake	Thesis title
Lim Ming Yee	AY2005/06	Analysis of Palatal and Arch Form Changes Associated with Class i, Class ii Div 1 and Class iii Orthodontic Treatment
Thesis Submission date	Supervisor(s)/ Dental co-supervisor	
2008	Dr Soh Jen Dr Ong Sim Heng	

List of MDS Students Conferred in AY 07/08 *continued***(Periodontology) Thesis Projects**

<b>Student Name</b> Christina Cheok	<b>Residency Intake</b> AY2005/06	<b>Thesis title</b> The Biological Effects of Non-Surgical Periodontal Therapy on Diabetics and Healthy Individuals	<b>Awards(s) won</b> Third place in the International Dental Exhibition and Meeting (IDEM) 2008 Poster Competition, Singapore
<b>Thesis Submission date</b> 2008	<b>Supervisor(s)/ Dental co-supervisor</b> A/P Lim Lum Peng		
<b>Student Name</b> Chang Kok Meng	<b>Residency Intake</b> AY2005/06	<b>Thesis title</b> The Effects of Periodontal Intersection on The Halitosis Status of a Diabetic Population Compared with Health Controls	<b>Awards(s) won</b> Fourth place in the International Dental Exhibition and Meeting (IDEM) 2008 Poster Competition, Singapore
<b>Thesis Submission date</b> 2008	<b>Supervisor(s)/ Dental co-supervisor</b> A/P Lim Lum Peng		
<b>Student Name</b> Ang Chee Wan	<b>Residency Intake</b> AY2005/06	<b>Thesis title</b> Alveolar Ridge Preservation with Polycapactose Scaffold Compared to Extraction Alone for Implant Site Development: A Clinical, Radiographic and Histologic Study in Humans	
<b>Thesis Submission date</b> 2008	<b>Supervisor(s)/ Dental co-supervisor</b> A/P Lim Lum Peng/ Dr Chung Kong Mun		

**(Endodontics) Thesis Projects**

<b>Student Name</b> Tay Li Chye	<b>Residency Intake</b> AY2005/06	<b>Thesis title</b> Effects of The Endoactivator on Smear Layer and Debris Removal at The Apical 1/3 pf Curved Root Canals: A Sem Study	
<b>Thesis Submission date</b> 2008	<b>Supervisor(s)/ Dental co-supervisor</b> CI A/P Patrick Tseng/ Dr Anil Kishen		
<b>Student Name</b> Chung Tze Onn	<b>Residency Intake</b> AY2005/06	<b>Thesis title</b> Effects of Periodontal Cell Sheet on Delayed Replanted Teeth in Dogs - A Pilot Study	
<b>Thesis Submission date</b> 2008	<b>Supervisor(s)/ Dental co-supervisor</b> A/P Varawan Sae-Lim, A/P Phan Toan Thang		

## Listing of Students, Projects & Supervisors

### Academic Year 2007/2008

**Project Title : Pain Reduction Following Thrid Molar Surgery with 830nm GaA1As Low Level Laser Therapy of Energy Densities 50 & 100 J/cm<sup>2</sup>**

Zhou Shicai  
Chua Chew Kiat Simon, Jude  
Chan Pei Yuan  
Song Yi Lin  
Luo Wenyuan

**Supervised by**

Prof Loh Hong Sai  
A/P Yeo Jin Fei

**Project Title : A 3D Interactive and Visualisation Educational Platform in Oral Histopathology**

Zhuang Ronglin  
Lim Min Min  
Low Ming Hui  
Lu Zhiyin  
Yim Shao'en Joey Emilia  
Balasubramanian Suraj

**Supervised by**

A/P Yeo Jin Fei  
A/P Kelvin Foong  
A/P Keng Siong Beng

**Project Title : 3D Digitisation of Plaster models Using Computed Tomography- A Pilot Work on the Accuracy and Validation of the Approach**

Melvin Liew Kang Ming  
Ng Jing Hao  
Hoo Swee Tiang  
Tan Jiunn Ming Gary  
Ode Wataru  
See-Toh Kai Mun

**Supervised by**

A/P Kelvin Foong  
A/P Keng Siong Beng

**Project Title : Cytotoxicity Testing of Hemostatic Agents, Bonewax and Surgicel, on CRL-1486 Cells**

Leonardo Saigo  
Chew Qin'An Amelia  
Kwan Yi Alexia  
Chan Wai Seng  
Ong Guo An  
Sia Kia Suan Priscilla

**Supervised by**

A/P Cao Tong

**Project Title : Advanced Non-invasive Light Activated Disinfection (ANILAD): An Alternative to Caries Removal by Hand Instrumentation for Deep Occlusal Caries?**

Tan Hui Xian Melissa  
Cai Xingni  
Esther Ogawa  
Chong May May  
Gomes Mario Regino Wenliang

**Supervised by**

Dr Anil Kishen  
A/P Jennifer Neo

**Project Title : Fibre Post: Does Fit or Different Post Space Cleaning Methods Affect Retention?**

Tan Jun Da  
Joanne Ou Jiazhen  
Tang Hui Yun Marian Elizabeth  
Ho Swee Ferng Cindy  
Tan Wye Lynn  
Shella Tan Hui Lu

**Supervised by**

Dr Lim Kian Chong

## Faculty Research Day 2007

The Undergraduate Research Opportunities Programme (UROP) aims to cultivate research interest and provide every undergraduate in the Faculty of Dentistry, a chance to perform supervised research work. Students work in groups under the guidance of a research mentor to explore research ideas and test hypotheses. The programme commences in Year 2 Term 3 and lasts for about 2 years. Each group has to submit a final research report at the end of the programme. Selected groups will then present their findings during the annual Faculty Research Day.

Faculty Research Day 2007 was held on 9 November 2007 and saw seven groups presenting their findings to a panel of judges comprising Prof Murray Clyde Meikle, Prof Robert Gunnar Bergenholtz and A/P Hien Chi Ngo.

The results of the competition were as follows:

### Winning Group

**Group Members:** Lim Tian Wei, William, Teo Eu Gene, Cheng Jean Lynn (Miss), Wong Jian Zhang and Lim Zixiang

**Supervisor:** Dr. Anil Kishen

**Project Title:** Testing a Light Activated Therapy (ANILAD) to eliminate bacteria in tooth canal

The winning group represented the Faculty at the DENTSPLY Student Clinician Programme during the SEAIDE-IADR meeting.

### 1st Runner-up

**Group Members:** Foo Kee Yin, Jonathan, Tan Mei Na (Miss), Goh Siew Hor and Ivan Prabowo Budiharto

**Supervisor:** A/P Stephen Hsu

**Project Title:** Potential cariostatic effects of Yakult®

The 2<sup>nd</sup> place winners represented at SEAIDE.

### 2nd Runner-up

**Group Members:** Boey Qing Xia, Jasmine (Miss), Peh Yew Jia (Miss), Leo June Joo (Miss), Myat Mon Thinn (Miss), Chin Hui Fen, Sharon (Miss)

**Supervisor:** A/P Cao Tong

**Project Title:** Cytotoxicity of whitening gel with CRL-1486 human cell and MTT

### 3rd Runner-up

**Group Members:** Hu Shijia, Chia Ai Ping, Vanessa (Miss), Ngo Di Ying, Joanna (Miss), Teo Hee Hwa, Daniel

**Supervisor:** Dr. Anil Kishen

**Project Title:** Detection of horizontal and vertical cracks in extracted teeth using a microbend sensor

The 4<sup>th</sup> place winners represented at the 9<sup>th</sup> Dental Students' Scientific Conference, University of Malaya.



Starting from the left: Dr. Joanne Uy, A/P Varawan Sae-Lim, Prof Bergenholtz, Lim Zixiang, Lim Tian Wei, William, Prof Murray Meikle, Wong Jian Zhang, Teo Eu Gene, Cheng Jean Lynn (Miss), Dr. Anil Kishen and A/P Hien Chi Ngo

## MSc Project

**Thesis Title:** Molecular Profile of Periodontal Tissue Following Tooth Replantation

**Student :** Dr. Tarun Kumar Maheshwari

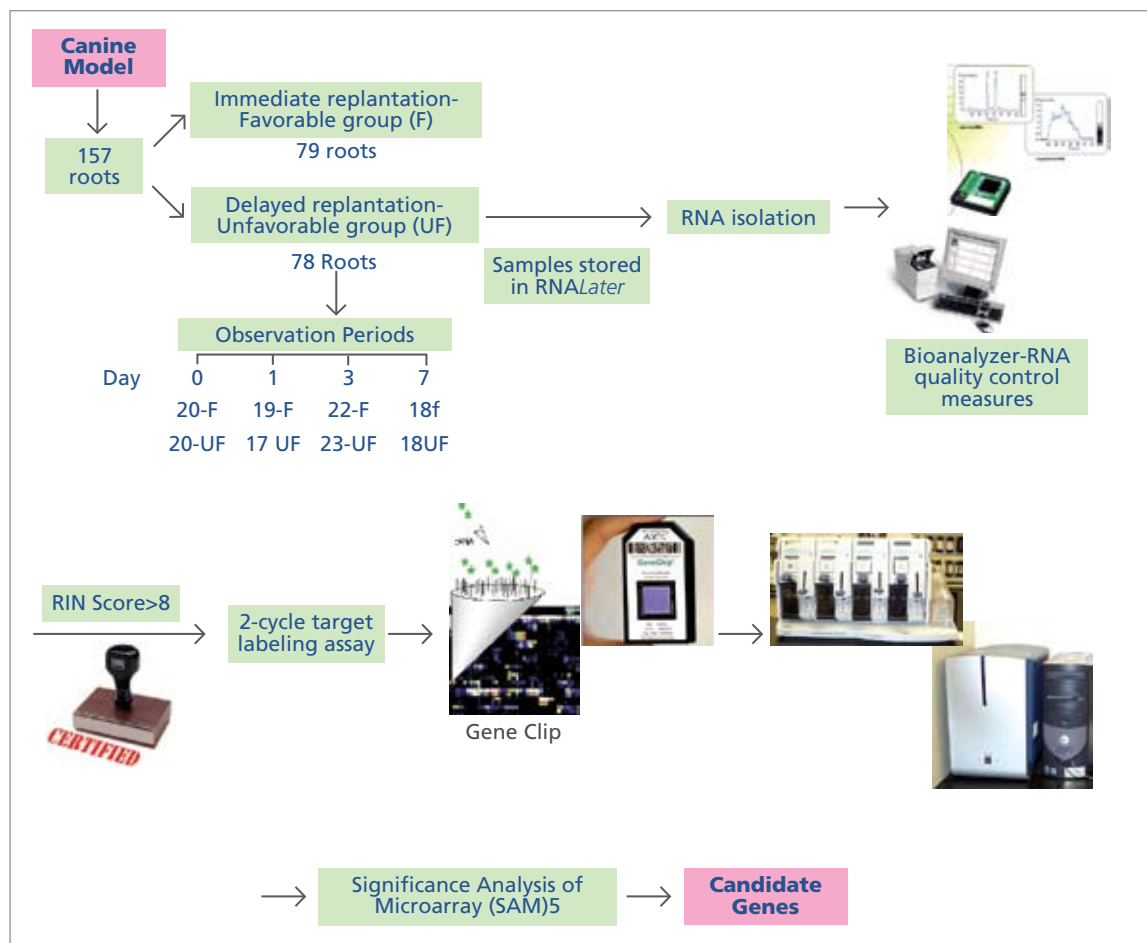
**Supervisor :** Assoc Prof Varawan Sae-Lim

*This project was awarded the Senior Travel Award (First Runner Up) at the 21st Annual Scientific Meeting IADR/SEA*

### Introduction and Objectives

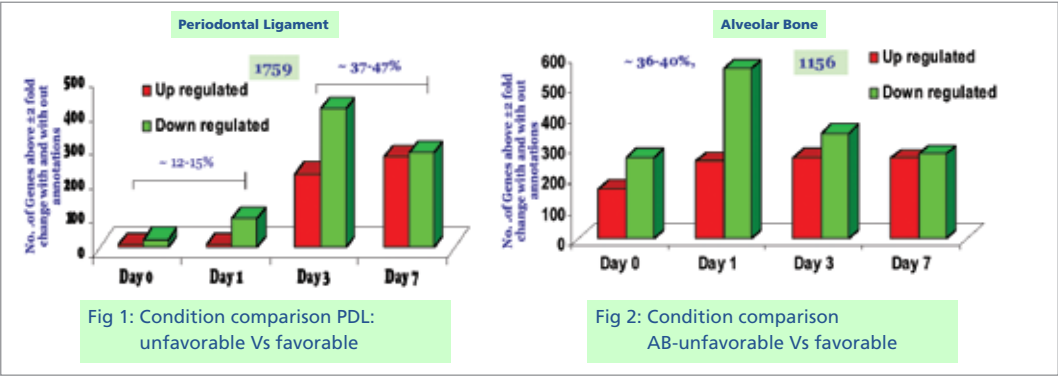
- Prolong delayed replantation of avulsed teeth unavoidably results in adverse consequences of ankylosis and replacement resorption that eventually lead to tooth loss<sup>1</sup>
- Periodontal ligament (PDL) and alveolar bone (AB) play a critical role in the biological response initiated towards healing process in replanted tooth<sup>2</sup>
- Using high-density oligonucleotide arrays, the molecular signatures of PDL and AB in delayed replanted teeth were determined in canine model

### Material and Methods





Results and Discussion



- Conditional Comparison [Unfavorable vs. Favorable]**
- AB & PDL-Difference in genetic expression pattern
  - AB-Highly vascularized, maintaining vitality during traumatic episodes2 with uniform expression
  - PDL-Paravascular progenitor cells differentiating at Day 3,7 coinciding with known wound healing process3,4

Table 1A: Temporal Expression in Immediate- Favorable Group							
Expression (9945)	All 3 Days		2 Days		1 Day		
Observation day	Day 1, 3&7	Day 1&2	Day 3&7	Day 1&7	Day 1	Day 3	Day 7
Up Regulated	0	0	99	0	0	28	24
Down Regulated	1	0	18	0	0	7	5

Table 2A: Temporal Expression in Immediate- Favorable Group							
Expression (3112)	All 3 Days		2 Days		1 Day		
Observation day	Day 1, 3&7	Day 1&2	Day 3&7	Day 1&7	Day 1	Day 3	Day 7
Up Regulated	4	4	11	10	17	12	13
Down Regulated	1	2	11	1	1	4	27

Table 1B: Temporal Expression in Delayed- Unfavorable Group							
Expression (9458)	All 3 Days		2 Days		1 Day		
Observation day	Day 1, 3&7	Day 1&2	Day 3&7	Day 1&7	Day 1	Day 3	Day 7
Up Regulated	2	0	152	2	0	0	24
Down Regulated	0	0	12	1	0	7	8

Table 2B: Temporal Expression in Delayed- Unfavorable Group							
Expression (3112)	All 3 Days		2 Days		1 Day		
Observation day	Day 1, 3&7	Day 1&2	Day 3&7	Day 1&7	Day 1	Day 3	Day 7
Up Regulated	9	2	20	4	6	10	11
Down Regulated	8	5	15	0	3	4	15

Temporal Comparison [Day 0 vs 1,3,7]

- AB**
- Maintains uniform genetic activity throughout early period of wound healing process
  - Highly vascularized, maintains potential to modulate gene expression in wound healing following the traumatic episode
- PDL**
- Lower genetic activity at days 0,1 with increased activity at day 3 onwards in both groups
  - Seems coinciding with perivascular progenitor cell differentiation and infiltration of inflammatory cells and angiogenesis

Fig 3: PDL- Biological functional categories

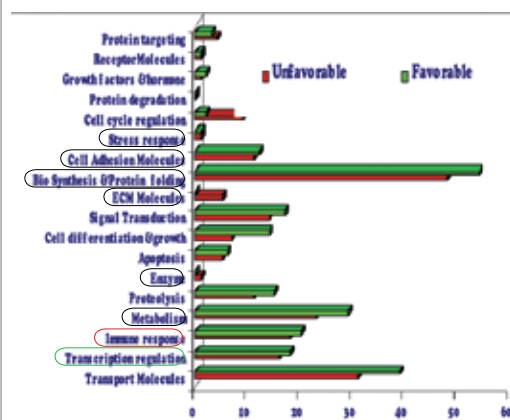
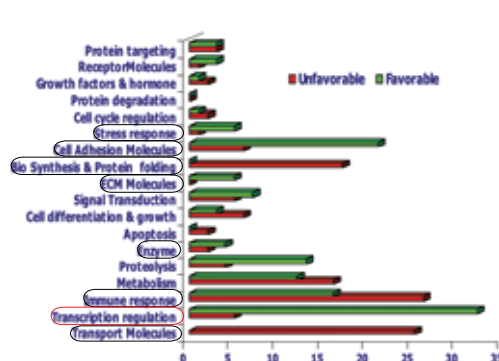


Fig 4: AB-Biological functional categories



### Biological functional Categories

- Similar and difference in functional categorical genetic expression between PDL and AB.
- Various functional genes played different roles in the regenerative process

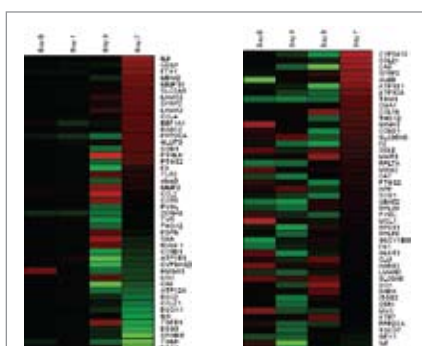


Fig 5: PDL- Genes

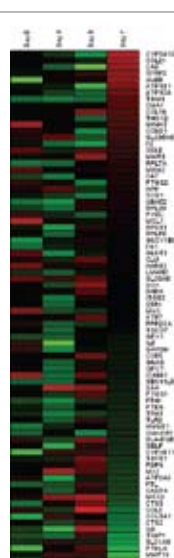


Fig 6: AB-Genes:

### Regeneration related functional genes:

- PDL- Genes known to suppress cell growth, cell adhesion, proliferation & enhance differentiation of osteogenic cell types. This genes can lead to the adverse consequences of replacement resorption and tooth loss.
- AB- genes that modulate regeneration belonging to functional categories like protein synthesis, immune response and angiogenesis.

### Conclusion

- Generally a lower gene expressions was seen in PDL compared to AB in both favorable and unfavorable groups.
- Onset of molecular events was almost immediate in the AB commencing from day 1. In PDL onset of events was delayed till day 3.
- This study suggests that PDL tissue in the unfavorable group had been severely stressed, undergo degradation with diminished potential to express genes which modulate regeneration and repairs.

## PhD Project

**Thesis Title:** Investigation on the Bacterial Biofilm in Root Canal Dentine and Elimination by Modified Light Activated Therapy

**Student :** Dr Saji George

**Supervisor :** Dr Anil Kishen

### Background

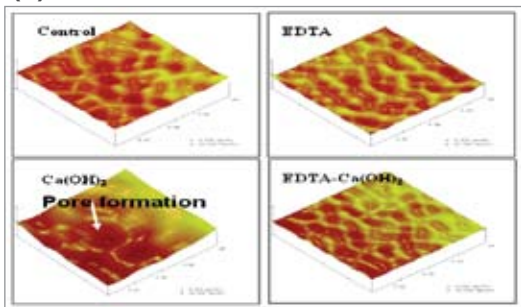
The possibility of using Light Activated Therapy (LAT) to disinfect the ‘root canals’ of infected teeth was examined in this project. This objective was achieved by designing a specific photosensitizer formulation’ and a treatment strategy, for infected one need to understand the physico-chemical nature and interaction of biofilm bacteria with root canal dentine.

### Factors that influence bacterial interaction with dentine

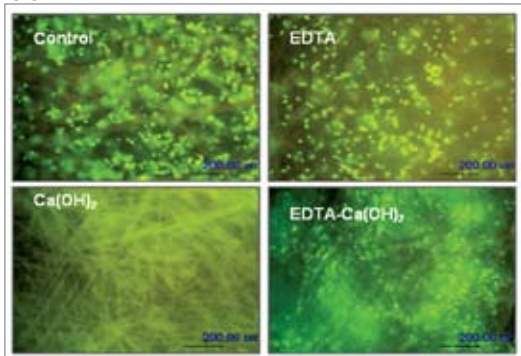
Main goal of this investigation was to establish the microbial and treatment related factors that determine bacterial adherence to dentine

tissues, a process that eventually leads to biofilm formation. Hydrophobicity of cells, one of the physical factor that help bacteria adhere to a biotic surface, was studied in a collection of oral bacteria that are often involved in biofilm mediated infections. Results showed that bacteria involved in the primary stage of oral biofilm development- *S. mutans*, *F. nucleatum*, and *P. gingivalis* are relatively more hydrophobic than *E. coli* and *E. faecalis*. However, hydrophobic nature of *E. faecalis* was significantly increased on subjecting them to starvation. Likewise, in a study using Atomic Force Microscopy and biochemical assays, we were able to show that, ‘pre-exposure’ to EDTA (chemical used to clean root canal) could make *E. faecalis* resist the high pH generated by Calcium hydroxide. These two studies highlighted the possible reasons of bacterial survival after root canal therapy and established that ‘survivors’ of root canal therapy could adhere to dentine which in later stages could develop into biofilm.

(A)



(B)



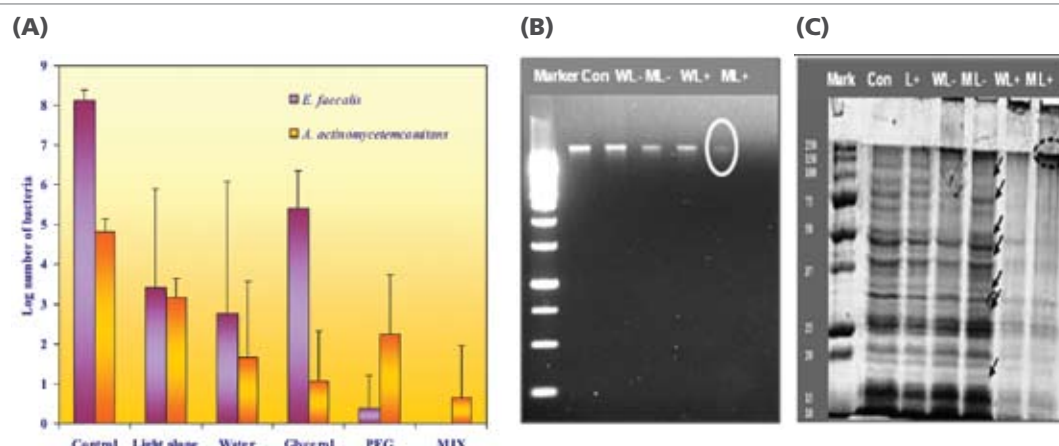
The Ca(OH)<sub>2</sub> induced damage to cell wall (A) and inhibition of bacterial adhesion to collagen sheet (B) were reversed by pretreatment with EDTA

### Antibiofilm effect of LAT

The antibacterial effectiveness of LAT was tested in bacterial biofilms developed in the root canals of extracted teeth under in vitro conditions. Results showed that LAT using the photosensitizer formulation ‘MIX’ could achieve 100% elimination of bacteria. However, bacteria became recalcitrant to LAT as well as to chemical disinfectant as the biofilm got matured. The lowered antibacterial effect was attributed to the pockets of viable bacteria probably protected by the mineralized biofilm matrix. However, it should be noted that the formation of thick mineralized biofilm under in vivo situation is highly unlikely. Nevertheless, a strategy that would allow the dissolution of biofilm matrix was suggested to enhance the effectiveness of LAT while treating matured biofilm. Studies were also conducted in

fibroblast cell lines using an experimental set up to model the clinical situation. The cytotoxicity of LAT was significantly less when compared to sodium hypochlorite, the chemical disinfectant

often employed during root canal therapy. These experiments showed that, LAT could kill bacteria at a faster rate compared to human cells.

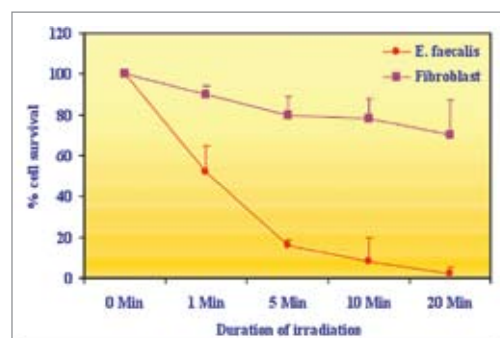


Among the different solvent systems tested the MIX showed maximum bacterial killing (A).

The mechanisms of bacterial killing mediated by LAT was found to be chromosomal DNA damage (B) and outer membrane protein degradation (C)

### Cytotoxicity of LAT

The next task in recommending a LAT based disinfection strategy for clinical treatment was to establish the cytotoxicity and compare with that of the currently applied disinfectants. Studies were conducted in fibroblast cell lines using an experimental set up to model the clinical situation. The cytotoxicity of LAT was significantly less when compared to sodium hypochlorite, the chemical disinfectant often employed during root canal therapy. Moreover, experiment was carried out to establish the specificity of LAT, where human cell lines and bacteria were simultaneously exposed to LAT. The result showed that, LAT could kill bacteria at a faster rate compared to human cells.



Bacteria (red line) were found to be killed at a faster rate than mammalian cells (purple line) when both the cells were simultaneously exposed to LAT.

### Conclusion

Our studies showed the potential of LAT as a root canal disinfecting agent. In addition the biofilm characterization studies gave new insight into the chemical and structural properties of matured bacterial biofilm and a potential reason for its recalcitrance towards antimicrobial agents.

## MDS Project

**Thesis Title:** The Effect of Periodontal Cell Sheet on Delayed Replanted Teeth in Canine Model (A proof of concept)

**Student :** Dr Chung Tze Onn [*Thesis in partial fulfillment for MDS (Endodontics)2008*]

**Supervisor :** Assoc Prof Varawan Sae-Lim

### Summary

Delayed replanted teeth present critical-sized periodontal defects resulting in adverse consequences of dentoalveolar ankylosis and replacement root resorption [Figure 1, arrow] that eventually leads to tooth loss. Previous physico-chemical therapeutic modalities have not shown predictable success in inhibiting such damaging outcome. In this study, we adopted cell-based therapy using autologous periodontal cell-sheet-wrapping technic in a canine model.

### Materials & Methods:

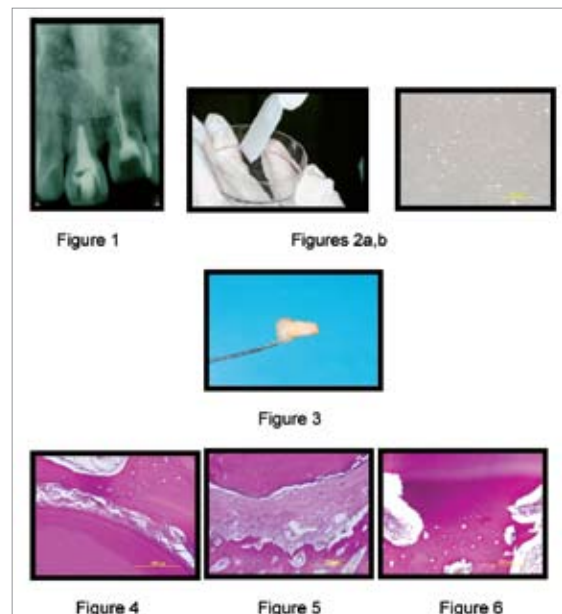
Canine roots were endodontically treated and extracted. Teeth in the negative and the positive control groups were replanted immediately and after one-hour bench-dry, respectively. Experimental roots were extracted for periodontal fibroblasts explant to form cell sheet [Figures 2a,b] while the roots were treated. The cell-coated roots [Figure 3] were subsequently replanted. After about 6 weeks, the roots cum jaw bone were harvested, histologically prepared and histomorphometrically evaluated. Statistical analyses were performed using Kruskal-Wallis and Mann-Whitney U tests.

### Results:

There was significantly less observation of replacement root resorption in the experimental group [Figure 4] employing periodontal cell-sheet-wrapping technic. The observation was not significantly different from that in the negative control group [Figure 5] where optimal healing was observed while the positive controls [Figure 6] showed significantly more replacement resorption as expected ( $p < 0.05$ ).

### Conclusions:

Periodontal cell sheet constructs were successful in preventing replacement resorption. Longer observation period is needed to validate the promising signs if the cell sheet integrates optimally with the alveolar bone.



## UROP-Faculty Research Day Winning Group

**Project Title** : Testing a Light Activated Therapy (ANILAD) to eliminate bacteria in tooth canal

**Group Members : Lim Tian Wei, William, Teo Eu Gene, Cheng Jean Lynn (Miss), Wong Jian Zhang and Lim Zixiang**

**Supervisor : Dr. Anil Kishen**

*The winning group represented the Faculty at the DENTSPLY Student Clinician Programme during the SEAADE-IADR meeting.*

Endodontic therapy presents a risk of failure because of the difficulty involved in controlling and eliminating infection. The major factors associated with root canal treatment failure are persistent microbial infection in the root canal system and/or periradicular tissue, as well as procedural errors, such as broken instruments and perforations. Light activated therapies (LAT) have recently become increasingly important adjuncts in root canal disinfection, where an exogenous photosensitizer absorbs the energy of a low-power laser and kills the target bacteria cell. The aim of this study was to investigate the efficacy of a modified two step LAT setup utilizing a tissue specific photo-sensitizer formulation (MIX) for sensitization followed by an liquid optical conduit for irradiation and to compare it with other anti bactericidal modalities in conventional root canal treatment.

A total of 55 prepared single rooted tooth radicular sections with similar root canal morphology were used for this study. An *Enterococcus faecalis* biofilm was developed for four days in the root

canal walls by incubation of the tooth specimens with the bacteria. *E. Faecalis* was chosen for this study because it was found to be extremely difficult to eradicate and form biofilms in established root canal infections. The comparative analysis of the microbial reduction by different antibacterial modalities was obtained in vitro in 55 prepared radicular sections through the following experimental groups, as shown in Figure 1.

The number bacteria surviving different treatment procedures are shown in Figure 2. The results showed that ANILAD is as effective, if not more so, in eliminating bacteria in the root canal system compared to sodium hypochlorite or other previous existing setups of LAT.

ANILAD has the potential to be used as an effective adjunct as an anti-bacteria treatment modality in treating root canal infections. Further in vivo studies are needed in order to ascertain the clinical value of the use of this modified setup of LAT in the disinfection of root canal in root canal therapy.

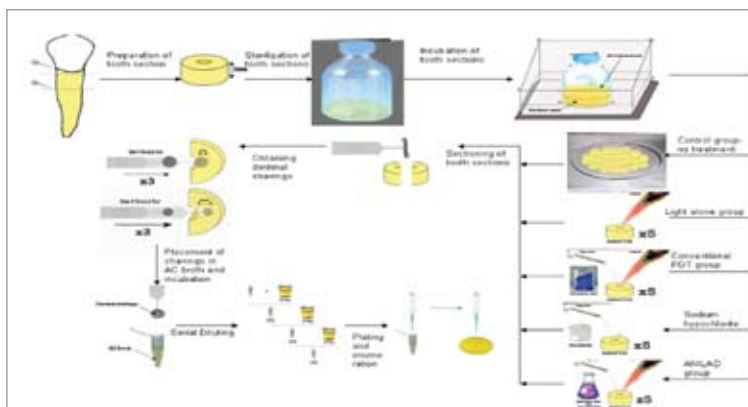


Figure 1: Schematic diagram showing the experimental procedure

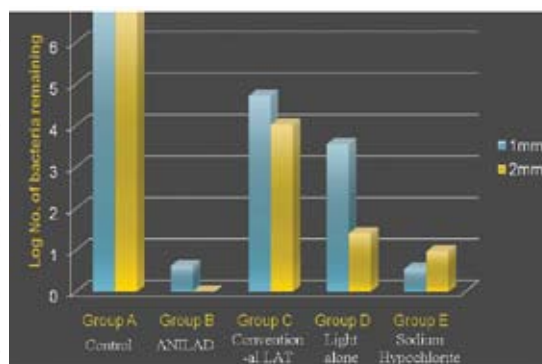


Figure 2: Graph showing the number of bacteria remaining after treatment in different experimental groups



## The Use of Dexmedetomidine for Conscious Sedation in Dental Surgery



**Principal Investigator:**  
**Dr Victor Fan**

**Co-Principal Investigator:**  
**Assoc Prof Ti Lian Kah, Department of Anaesthesia**

**Total Project Value:**  
**S\$49,020**

**Introduction:** The purpose of this study was to compare the efficacy and safety of Dexmedetomidine and Midazolam for conscious sedation in dental surgery and also compare the psychomotor recovery of patients receiving either dexmedetomidine or midazolam

**Abstract:** This study was a prospective, randomized, double-blinded study with 60 healthy ASA I or II patients who presented for outpatient dental surgery. A BIS monitor was also used in all patients. Patients were randomly assigned to receive either midazolam or dexmedetomidine and the degree of sedation was titrated. Patients in the midazolam group received an infusion of 0.005 mg/kg/min, while the patients in the dexmedetomidine group received an infusion of 0.1 mcg/kg/min, until the adequate sedation was achieved. Once the desired BIS was achieved, the patients were kept within 10% of that BIS level with intravenous infusions titrated to effect, starting at rates of 0.01 mg/kg/min midazolam and 0.2 mcg/kg/min dexmedetomidine respectively. Standard monitors were applied to the patients (NIBP, ECG

and pulse oximeter). Demographic and surgical data, including age, weight, height, gender, ASA status, medical history, type of surgery performed, length of surgery, and amount of local anaesthetic used was recorded. The amount of loading dose and maintenance dose required was recorded for both groups. Oxygen saturation (pulse oximeter), heart rate, continuous electrocardiograph (lead II), and non-invasive blood pressure was recorded on arrival, and continued till discharge.

**Results:** The mean HR, Systolic and diastolic blood pressure was significantly reduced in the Dexmedetomidine group. There was no significant difference in the respiratory rates, BIS and total volume (of the drugs used). However Dexmedetomidine group showed a greater reduction in anxiety ( $p < 0.032$ ) and better patient cooperation. There was no significant difference in the oxygen saturation or the duration of surgery in the two groups.

**Clinical Significance:** Dexmedetomidine can work as well as midazolam in outpatient dental procedures and can be considered as an alternative to midazolam. It has been shown to be reliable, safe with well documented pharmacological and hemodynamic properties. It can be safely used to provide desired sedation levels to patient during outpatient dental procedures. An oral paper was presented at the 8th Asian Congress of Oral and Maxillofacial Surgeons on 4th November 2008 and a written paper is now being prepared for submission.

## Innovative Non-Invasive Laser Treatment for Preventive of Enamel Demineralization (Tooth Decays)



**Principal Investigator:**  
**Assoc Prof Stephen Hsu**

**Total Project Value:**  
**S\$799,450**

### Summary/Achievements

Four articles have been published in the same premier journal ("Journal of Dentistry"), and another was published in a major conference (SPIE proceedings), in addition to two postgraduate thesis, eighteen conference abstracts and 2 local & 5 international research awards won by the team members.

The inhibitory effect of laser to tooth decay has been further confirmed by using two Er:YAG lasers of different brands and by in vitro and in situ models. Different demineralization solutions were used in the acidic challenging process, such as standard demineralization solution, coke, and other acidic drinks, to assess the variability of laser-induced inhibition on enamel demineralization. The optimal range of laser parameter has been identified. The laser-enamel interaction has been preliminarily modeled to determine the optimal range of photothermal impact that may maximize the beneficial effect and minimize the detrimental effect. A non-invasive method using laser scanning confocal light microscopy (LSCM)

for quantification of enamel micro-diffusion has been developed to verify the "Organic blocking theory". Furthermore, a non-invasive mineral profiling technique using micro CT has been applied to evaluate various laser therapies. In summary, the characterization of lased/heated enamel has been performed using polarized light microscope (PLM), laser scanning confocal light microscopes (LSCM) with Fluorescence Recovery After Photobleaching (FRAP) and Fluorophore Transport Study (FTS) technique, Fourier Transform Raman spectroscopy (FT-Raman), micro-Raman spectroscopy (micro-Raman), micro-FTIR spectroscopy, micro-XRD, Scanning electron microscope (SEM) and Microcomputerised tomography (micro-CT), Microradiography (MRG), Harmonic Generation (HG), Atomic Force Microscopy (AFM), Contact Angle, Zeta Potential, and Fluorescence Microscopy etc.

To further elucidate the mechanism of laser-induced caries prevention for clinical application, preliminary results on the physiochemical surface properties (contact angles, surface free energy, zeta potential) of three bacterial strains and heated enamel/saliva coated enamel have been obtained. These results are critical in determining the alteration of the adhesion force between the oral bacteria and enamel surfaces after heat/laser treatment. Two new grant proposals, based on these new findings and preliminary data of this project, has been approved.

## Tissue Reaction to Implanted Viscosity Enhanced Root Repair Material (VERRM)



**Principal Investigator:**  
**Assoc Prof Yeo Jin Fei**

**Total Project Value:**  
**S\$45,382**

**Introduction:** Mineral Trioxide Aggregate (MTA) as a root-end filling material has generated a lot of interest due to its superior sealing ability and biocompatibility. While MTA possesses superior sealing ability and is less cytotoxic compared to traditional root-end filling materials such as Super-Ethoxy Benzoic Acid (super-EBA) and Intermediate Restorative Material (IRM), it has poor handling characteristics. A novel root-end filling material i.e. Viscosity Enhanced Root Repair Material (VERRM), with similar chemical composition but improved handling characteristics was recently developed. This material has been tested and was found to fulfill the physical properties requirements for use as a root-end filling material.

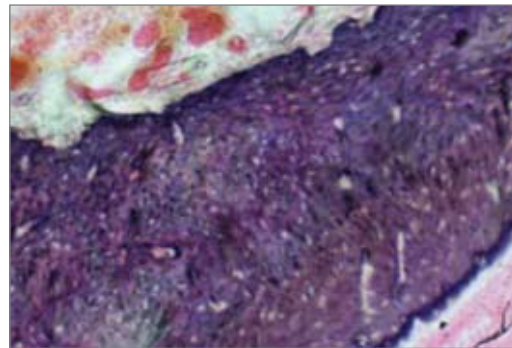
**Objective:** The aim of this study is to examine the tissue reactions to Viscosity Enhanced Root Repair Material (VERRM) implanted in the mandible of guinea pigs and to compare the reactions to those induced by MTA.

**Materials & Methods:** Under aseptic conditions, fifteen adult male guinea pigs were anesthetized. Tissue flaps were raised and bony cavities were created in the mandibles of the animals with burs. The materials MTA and VERRM were then implanted in these bony cavities. MTA and VERRM were implanted using Teflon cups as the carrier for the materials. The animals were randomly divided into 3 groups of 5 animals each. Each animal received one implant in the mandible. The animals were euthanized after a period of 80 days and the tissues were processed for histological examination using the Exakt cutting and grinding system.

**Results:** Both the materials showed similar tissue reactions. The absence of inflammatory reactions suggested that both the materials are biocompatible. (Please see photomicrograph showing the histologic evidence confirming the lack of reactions in the connective tissue adjacent to the VERRM).

**Conclusion:** The results of this study indicated that VERRM is a biocompatible material when implanted into the mandible of guinea pigs. It was also observed that this material behaves in a similar manner to MTA. VERRM would therefore have the potential to be used as a root-end filling material. However, further investigations are needed in order to determine its suitability for clinical use as a root-end filling material.

[This report is compiled from the MSc Thesis entitled: "A comparative study of the biological and physical properties of Viscosity Enhanced Root Repair Material (VERRM) and MTA" submitted by Dr. Pallavi Uppangala.]



**Photomicrograph showing the histologic evidence confirming the lack of reactions in the connective tissue adjacent to the VERRM**

## Role of Brain Phospholipases A<sub>2</sub> in Orofacial Pain



**Principal Investigator:**  
**Assoc Prof Yeo Jin Fei**  
**Total Project Value:**  
**S\$178,000**

### Summary/Achievements

This study was carried out, using inhibitors to secretory phospholipase A<sub>2</sub> (sPLA<sub>2</sub>, 12-epi-sclalaradial), cytosolic phospholipase A<sub>2</sub> (cPLA<sub>2</sub>, AACOCF<sub>3</sub>), or calcium-independent phospholipase A<sub>2</sub> (iPLA<sub>2</sub>, bromoenol lactone), to compare possible contributions of central nervous PLA<sub>2</sub> isoforms to the development of allodynia after facial carrageenan injection in mice. C57BL/6J (B6) mice showed increased responses to facial stimulation using a von Frey hair (1 g force), at 8 h, 1 day, and 3 days after facial carrageenan injection. On the other hand, BALB/c mice did not show increased responses at any of the time points. In

both B6 and BALB/c mice, intracerebroventricular injection of inhibitors to each of the three PLA<sub>2</sub> isoforms significantly reduced responses to von Frey hair stimulation at 8 h and 1 day after facial carrageenan injection, but at 3 days after injection, only the sPLA<sub>2</sub> inhibitor had an effect (Fig. 1 and Fig. 2). Since BALB/c mice did not show increased responses after facial carrageenan injection, the reduction in responses actually indicates that there is loss of normal sensitivity to von Frey hair stimulation after intracerebroventricular injection of each of these inhibitors, in this strain of mice. The effects of PLA<sub>2</sub> inhibitors are unlikely to be due simply to inhibition of arachidonic acid generation, since intracerebroventricular injection of arachidonic acid also had an anti-nociceptive effect. These results support an important role of central nervous PLA<sub>2</sub> in neurotransmission and pain transmission. The findings of this project indicate that PLA<sub>2</sub> isoforms could be a novel and important therapeutic target for orofacial pain.

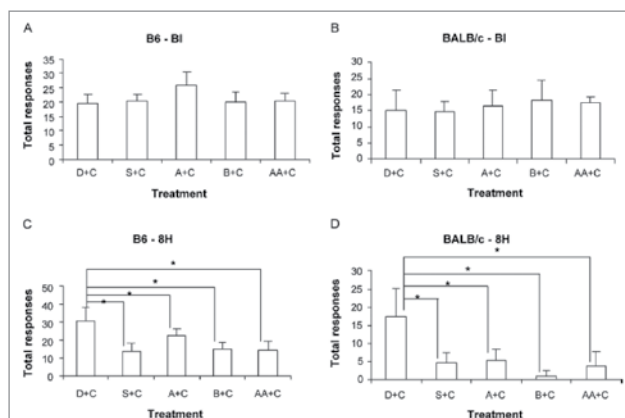


Fig. 1. Total responses in mice before treatment, and at 8 h after treatment with DMSO, PLA<sub>2</sub> inhibitors, or arachidonic acid, and carrageenan. (A and C): B6 mice. (B and D): BALB/c mice. (A and B): Total responses in mice before treatment. Abbreviations indicate treatments that the mice received the following day. There were no statistically significant differences between the mean total responses of any of the groups, in either the B6 or the BALB/c mice, i.e. the starting values were matched. (C and D): Total responses in mice that received intracerebroventricular injection of DMSO (5 ml, vehicle control), PLA<sub>2</sub> inhibitors (0.01 mmol in DMSO), or arachidonic acid (0.01 mmol in DMSO), and facial injection of carrageenan, 8 h after injections. Both strains of mice showed significantly fewer total responses to von Frey hair stimulation of the face, after injection with inhibitors to each of the three isoforms of PLA<sub>2</sub>, or arachidonic acid, compared to DMSO. Abbreviations indicate treatments received: D+C, S+C, A+C, B+C, AA+C indicate DMSO, 12-epi-sclalaradial, AACOCF<sub>3</sub>, BEL, or arachidonic acid, plus carrageenan, respectively. BI and 8H: before injections and 8 h after injections. Analyzed by 1-way ANOVA with Bonferroni's multiple comparison post hoc test. Asterisks indicate statistically significant differences (P<0.05).

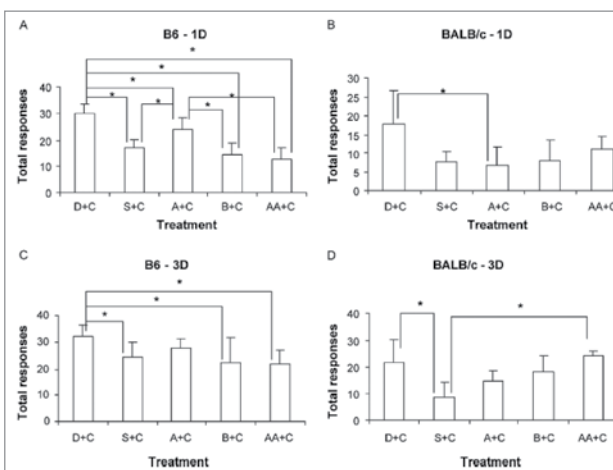


Fig. 2. Total responses in mice treated with DMSO, PLA<sub>2</sub> inhibitors, or arachidonic acid, and carrageenan, at 1 and 3 days after injections. (A and C): B6 mice. (B and D): BALB/c mice. (A and B): Total responses in mice, 1 day after injections. B6 mice (A) showed significantly fewer total responses to von Frey hair stimulation after injection with inhibitors to each of the three isoforms of PLA<sub>2</sub>, or arachidonic acid, compared to DMSO. BALB/c mice (B) showed significantly fewer total responses after injection with AACOCF<sub>3</sub>, compared to DMSO. (C and D): Total responses in mice, 3 days after injections. Both strains of mice showed significantly fewer total responses after injection with 12-epi-sclalaradial, compared to DMSO. Abbreviations indicate treatments received: D+C, S+C, A+C, B+C, AA+C indicate DMSO, 12-epi-sclalaradial, AACOCF<sub>3</sub>, BEL, or arachidonic acid, plus carrageenan, respectively. 1D, 3D: 1 and 3 days after injections. Analyzed by 1-way ANOVA with Bonferroni's multiple comparison post hoc test. Asterisks indicate statistically significant differences (P<0.05).

## Rapid Optical Method to Monitor Caries Activity in Children



**Principal Investigator:**  
**Dr Anil Kishen**

**Total Project Value:**  
**\$537,965**

### Summary/Achievements

Despite many advances in dentistry today, a reliable, cost effective and easy-to-use chairside method to monitor caries activity in children is still lacking. The multifactorial etiology of dental caries is considered to be the main obstacle in this development. The cardinal step in caries process is the formation of acid following carbohydrate intake, which lowered the pH below the critical level, resulting in the dissolution and disintegration of dental hard tissues. In addition, certain salivary proteins are known to facilitate bacterial adherence and colonization to the tooth surface. The levels of salivary total-protein have been associated with both caries activity and periodontal disease conditions.

Optical spectroscopy is a highly sensitive technique that can determine biochemical parameters from saliva both quantitatively and qualitatively. This study aimed to apply the principles of optical spectroscopy to monitor bacterial-mediated acidogenic-profile and total-protein concentration of saliva. Experiments were conducted using optical spectroscopic sensor (OSS) to monitor the bacterial-mediated acidogenic-profile of saliva mixed with sucrose from 70 children of age group 6-12 years. Bromophenol blue, a pH sensitive photoindicator, was used to monitor the rate of acid production by salivary bacteria following the addition of sucrose. The findings from the OSS were compared with the past caries experience (caries

scores: dmfs index), caries activity determined using mutans streptococci counts (Dentocult SM) and Lactobacilli counts (Dentocult LB) and pH shifts (pH-meter). The OSS showed a decrease in the absorption intensity at 595nm wavelength of saliva-sucrose sample with decrease in the pH. The half-life decay value determined from the OSS differed significantly between the caries-active (CA), caries-inactive (CI) and caries-free (CF) groups ( $P<0.05$ ). Children in CF group (90%) showed low MS and LB counts. However, this correlation was weak in cases of the CA and CI groups. The OS was able to distinguish children showing different caries activity. Experiments were also conducted to quantify the total-protein concentration of saliva using OSS. Absorption intensities for various protein concentrations were characterized using a protein sensitive indicator coomassie brilliant blue (CBB). Finally, considering the ability of the OSS to detect multiple parameters, a fiber optic based sensor incorporating two sensing regions for pH (methyl red as the photosensitizer) and protein (CBB as the photosensitizer) was fabricated using the principles of fiber optic evanescent wave spectroscopy (FOEWS) and sol-gel technology. This experiment demonstrated the ability of FOEWS to quantify total-protein concentrations and monitor the acidogenic-profile of saliva using a single sensor.

This study highlighted the ability of OSS to determine bacterial-mediated acidogenic-profile of saliva, which can be used as a caries activity indicator. The FOEWS developed was able to monitor the acidogenic-profile as well as quantify the total-protein content of saliva. The OSS can find significant application for the chairside measurement of clinically important parameters from saliva.

## User-Centric Intelligent Clinical Support System for Temporomandibular Disorders and Other Chronic Pain Conditions



**Principal Investigator:**  
**Assoc Prof Keson Tan**

**Total Project Value:**  
**S\$70,000**

### Summary/Achievements

Chronic facial pain is a multi-disciplinary problem. Amongst the four recognizable orofacial chronic pain complexes (Temporomandibular disorders [TMD], Atypical Facial Pain, Atypical Odontalgia and Oral Dysaesthesia), TMD is the most prevalent. TMD refer to a collection of medical and dental conditions affecting the temporomandibular joints and/or the muscles of mastication, as well as contiguous tissue components. The total number of people experiencing TMD-related pain is estimated at more than 10 million in America alone. Concerns regarding the welfare of TMD sufferers as well as safety and efficacy of TMD care have prompted NIDCR (National Institutes of Health, USA) to target TMD as one of its high priority research areas.

A critical obstacle to the further understanding of TMD is the lack of standardized diagnostic criteria for defining subtypes of TMD. The Research Diagnostic Criteria for TMD (RDC/TMD) was established by Dworkin & co-workers (1992) to address this lack. RDC/TMD allows for the standardization and replication of research into the most common forms of muscle and joint-related TMD. Its dual axis approach also allows physical diagnosis to be coordinated with operationalized assessment of pain-related behaviors, psychological distress and psychosocial function associated with TMD and other chronic pain conditions.

RDC/TMD is usually administered by pen and paper. The data collected are then entered manually and batch processed by a mainframe computer.

Therefore, there is a time lag between patient history taking/examination and the generation of the diagnosis. A project to create an on-line diagnostic tool based on the RDC/TMD was earlier undertaken by NUS. This computerized diagnostic system (NUS TMD v1.1) allowed for direct data input by patients/clinicians, chairside generation of Axis I and II findings, and automatic archiving of data in SPSS or other tab-delineated formats for data mining and global exchange. A trial program for this original tool involving 17 international centers in American, Europe, Asia and Australia was conducted. Preliminary findings of this program and the future evolution of RDC/TMD, warrants the development of the next generation of multi-lingual, clinician centered support system with intelligent multimedia interfaces and patient prognosis/treatment tracking engines.

The current project focused on developing a multi-lingual and multi-platform enabling electronic RDC/TMD system that can be used across the world. User-centered analysis on NUS TMD v1.1 was conducted. Data collected were used to enhance NUS TMD v1.1 resulting in the development of NUS TMD v2.0. TMD v2.0 incorporates several new functionalities including the diagnoses listing of the American Academy of Orofacial Pain guidelines (AAOFP). It has been employed clinically at the National University Hospital (NUH) as well as the National Dental Centre (NDC) and used as a platform to develop the new rules-based (Java xml) system. The latter offered significant advantages over a hard-coded system engine. The Java xml system and system's patient diagnostic classification validation were checked using a large patient base database. Several rounds of UAT were conducted and the system is currently awaiting pilot clinical trials. When fully implemented and deployed, the system will enable the establishment of a global network for TMD research and data pooling to further the scientific knowledge base on TMD.



## Utilizing Differentiated Progenies of Human Embryonic Stem Cells for Cytotoxicity and Genotoxicity Screening of Dental and Biomedical Products



**Principal Investigator:**  
**Assoc Prof Cao Tong**

**Total Project Value:**  
**S\$179,850**

### Summary/Achievements

Live animal models, primary cultures or established cell lines of vertebrates are commonly used to analyze the cytotoxic and genotoxic potential of newly developed drugs, xenobiotics, and various dental and biomedical products. However, these assays poorly reflect human physiology. An ideal alternative is to utilize differentiated progenies of human embryonic stem cells (hESC) for in vitro toxicology screening, because these can better mimic the physiological cell types within the human body. Additionally, the unrestricted proliferative capacity of hESC can provide a limitless source of differentiated somatic cells for this purpose. Hence, we endeavour to develop a technically-simple, cost-effective and replicable system of generating differentiated hESC progenies for toxicology screening. This is

been achieved through differentiation of hESC within the standardized culture. The data shows that differentiated fibroblastic hESC progenies can be reproducibly and efficiently generated through such a culture system. These are then applied to cytotoxicity and genotoxicity screening tests specified by the International Organization for Standardization (ISO) and the Organization for Economic Co-operation and Development (OECD) guidelines. Cytotoxicity is evaluated by the MTT/MTS, crystal violet and neutral red assay. Genotoxicity is assayed using single Cell Gel Electrophoresis (Comet Assay) for assessing DNA damage. Fluorescence In-Situ Hybridization (FISH) is also undertaken to evaluate specific chromosomal aberrations. Both PNA-FISH and multiplex FISH (mFISH) are employed. The hESC-derived fibroblastic progenies are also compared with established cell lines and primary cultures, to determine whether these provide a better cellular model for safety testing. This study thus allows for evaluation of the utility of hESC-derived progenies as in vitro models for toxicology screening, at the cellular, molecular and genomic level.

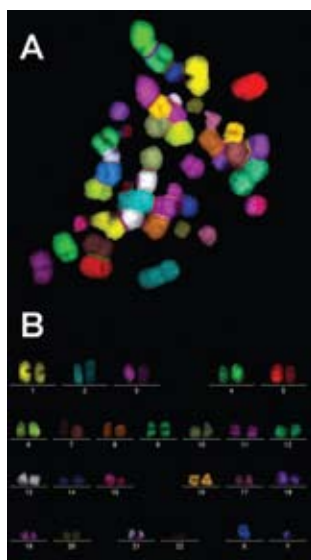


Figure 1. Untreated hESC control metaphase spread (A) and karyotype (B), as observed under mFISH

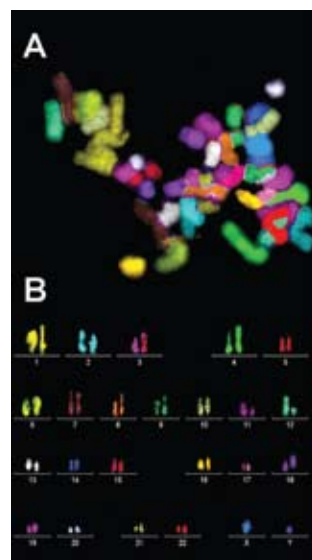


Figure 2. 0.06µg/ml Mitomycin-C treated hESC metaphase spread (A) and karyotype (B), as observed under mFISH

## Radiographic Assessment of Bone Regeneration in Alveolar Sockets with Scaffold after Teeth Extraction



**Principal Investigator:**  
**Dr Fan Tai Weng Victor**

**Total Project Value:**  
**S\$10,000**

**Aims:** To assess the effectiveness of a PLGA bio-scaffold in keeping the extracted tooth socket integrity, allow new bone formation and prevent the loss of alveolar bony height due to bone resorption.

**Abstract:** After dental extraction, there is socket contraction and loss in alveolar bone height due to resorption. The introduction of a bioresorbable scaffold should help to maintain socket integrity and should gradually resorb away to prevent risks of infection and foreign body reaction. The clinical trial was aimed to show the effect of placement of a bioscaffold in the alveolar socket in maintaining alveolar bone height with no adverse reactions and complete resorption with natural bone healing.

40 extraction cases were randomly divided into 2 groups: with and without insertion of the scaffold and radiographic assessments were done with the conventional OPG with a customized splint for each patient. The alveolar bone height were measured with 3 points taken on the x rays, before and after the extractions at 2 and 3 months intervals and the data collected by a computerized software scanner and analysed. The results had shown a significant reduction in ridge resorption after an alveolar scaffold was placed in those extractions sockets.

The bioscaffold was made of a resorbable material called PLGA which is poly-lactate-glycolic acid and produced by Bio-Scaffold International. This scaffold was jointly developed by faculty team, Dr Victor Fan and A/P Cao Tong as a TEC project with grants of \$750 million dollars from the Prime Minister's Office and the clinical trial was the final phase towards commercialization of the scaffold.

**Clinical relevance:** This is a novel concept in alveolar ridge preservation as prior to this there has been no rigid scaffolds used in such procedures. The scaffold has major advantages in that it is easy to place with no surgery involved. PLGA bio-scaffolds can be used in the extracted teeth socket to maintain space and yet allow bony healing, preserve the alveolar ridge and minimize ridge resorption after teeth extraction. With a better alveolar ridge, it will minimize the need for surgery for bone grafting prior to dental implantation and provide better support for denture prosthesis. Potentially all patients after extraction can have a bioscaffold inserted. Further development of this bioscaffold can lead to applications in reconstructive surgery such as mandibular reconstructions, cranio-maxillofacial applications (eg for skull defects, orbital floor reconstruction etc) and orthopaedic reconstructions. As a spin off from this project, a pilot animal study for jaw reconstruction was done.



## Accuracy of Fit of Implant-Abutment Systems Interfaces



**Principal Investigator:**  
**Assoc Prof Keson Tan**

**Total Project Value:**  
**S\$ 47,520**

**Aim:** To measure the fit tolerance of the listed component pair combinations new implant systems with different external and internal connection interfaces using the CMM.

- (a) implant to impression copings
- (b) impression coping to implant replica
- (c) implant replica to ceramic (Zirconia) abutments
- (d) abutment to implant

**Abstract:** Improperly fitting implant prostheses can lead to excessive localised stress levels in implants and prosthetic components in clinical service. Misfit or distortion in implant prostheses arise from errors from 2 sources: 1) distortion introduced during the clinical and laboratory stages of prosthesis fabrication; and 2) the machining tolerance / inaccuracy of the manufactured components themselves. The high resolution and measurement accuracy of Coordinate Measuring

Machines (CMM) has not yet been applied to define the tolerance of the interfaces of the newer implant-abutment systems that have been introduced in the past decade. These newer interfaces include internal connections like the 3i Certain and the NB Replace Select tri-cam, and newly introduced CAD-CAM ceramic (Zirconia) abutments which have also incorporated a modified metallic "inter-component" that makes the fit characteristics of these ceramic abutments likely different from their respective machined metallic predecessors. In addition, the tolerance of implant to impression coping to implant analog to abutment fitting component combinations of these newer systems interfaces is unknown. These machining tolerances contribute to the final level of inaccuracy or misfit and can contribute to the cumulative distortion in laboratory and clinical procedures.

**Clinical Relevance:** Identification of problematic tolerance levels and definition of these transfer inaccuracies in terms of the x, y and z rotational displacements as well as x, y and z linear displacements (6 degrees of freedom) will provide the clinicians with relevant information in assessing the clinical suitability of newer systems and the clinical service performance of implant prosthetic components.



Figure 1: Implant component feature measurement by ruby probe tip of CMM

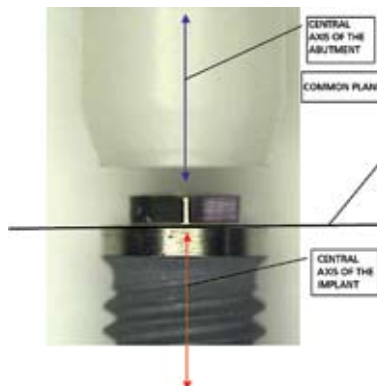


Figure 2: Common plane and the central axes of the superior and inferior implant components.

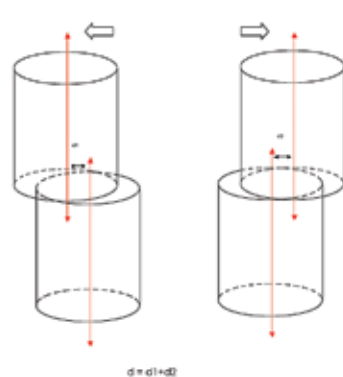


Figure 3: Movement limits defining tolerance (After Ma et al; 1997) in terms of relative displacements of central axes of components.

$d$  = total tolerance in XY plane  
 $d_1$  = tolerance in one direction  
 $d_2$  = tolerance in opposite direction

## Photothermal Modulation of Adherence of Bacteria on Human Enamel



**Principal Investigator:**  
**Assoc Prof Stephen Hsu**

**Total Project Value:**  
**S\$178,330**

### Aims:

- To evaluate the physiochemical properties of enamel modulated by heat and laser irradiation
- To characterize the physiochemical properties of three oral pioneer strains
- To quantify the nanoscale interactive forces between oral bacteria and enamel surfaces after laser/heat treatment and/or saliva coating

**Abstract:** Laser-induced caries prevention has been validated in the past decades. However, the laser effect on enamel surface related to bacterial colonization has not been characterized. An essential prerequisite for bacterial colonization is their ability to adhere to

a host surface, normally governed by long and medium range forces (primarily van der Waals and electrostatic forces). These forces depend on the physiochemical properties of substratum and the bacterial surface, such as hydrophobicity, free energy, and surface charges. In the oral cavity, streptococci (predominantly *Streptococcus sanguis*, *Streptococcus mitis*, and *Streptococcus oralis*) attach to the enamel pellicle within 2h after cleaning. The objectives of this study are 1) to characterize the physiochemical properties of three oral pioneer strains and lased/heated enamel surfaces, and 2) to quantify, using atomic force microscopy (AFM), the nanoscale interactive force between oral bacteria and enamel surface coated with saliva/calcium after heat and laser treatments. This study may elucidate the potential clinical effects of laser treatment on biofilm formation, in addition to the proven chemical/physical changes leading to inhibition of enamel demineralization.

**Clinical Relevance:** This study plays a critical role in translating the laboratory findings into the clinical setting and would facilitate the clinical application of laser therapies for caries prevention.

## Removal of Bacterial Biofilms Using Focused Ultrasound for Endodontic Disinfection



**Principal Investigator:**  
**Dr Anil Kishen**

**Total Project Value:**  
**S\$ 50,000**

**Aims:** This study aims to investigate the efficacy of collapsing-cavitation-bubbles produced by High-Intensity-Focused-Ultrasound (HIFU) to eliminate intracanal biofilm during root canal disinfection.

**Abstract:** High-Intensity-Focused-Ultrasound (HIFU) produces collapsing-cavitation-bubbles. This study aims to investigate the efficacy of collapsing-cavitation-bubbles to deliver antibacterial nanoparticles and disrupt biofilm to improve root canal disinfection. In stage-1, experiments were carried out to characterize the efficacy of collapsing-cavitation-bubbles to deliver the miniature plaster beads into a tubular channel model. In stage-2, experiments were conducted on root-dentin blocks to test the efficacy of HIFU applied at 27 kHz for 2 min to deliver antibacterial-nanoparticles into dentinal tubules. After stage-2 experiment the

samples were sectioned and analyzed using field emission scanning electron microscopy and Energy Dispersive X-ray analysis. Results- Stage-1 experiment showed that collapsing-cavitation-bubbles using HIFU delivered plaster beads along the entire length of the tubular channel. It was observed from stage-2 experiments that the diffusion of fluids alone was not able to deliver antibacterial-nanoparticles into dentinal tubules. The collapsing-cavitation-bubbles treatment using HIFU resulted in significant penetration upto 1000  $\mu\text{m}$  of antibacterial-nanoparticles into the dentinal tubules. The statistical analysis showed highly significant difference in the depth of penetration of antibacterial-nanoparticles between the two groups ( $<0.005$ ).

**Clinical relevance:** The cavitation bubbles produced using HIFU can be used as a potential method to deliver antibacterial-nanoparticles and disrupt biofilm to enhance root canal disinfection.

(A Shrestha, SW Fong, BC Khoo, A Kishen. Journal of the Acoustical Society of America 2008, Journal of Endodontics (in press))

## The Remineralization of Demineralized Dentin by Glass Ionomer Cement: The Effect of Apatite Forming and Non Apatite Forming Elements that can be Derived from Glass Ionomer



**Principal Investigator:**  
**Assoc Prof Hien Chi Ngo**

**Total Project Value:**  
**S\$169,000.44**

**Aims:** To study the mechanisms involved in the remineralization of partially demineralized dentin in the presence of glass-ionomer.

**Abstract:** As glass-ionomer contains both apatite forming and non-apatite forming elements; it is important to study how these are leached out of the material then infused into its immediate environments, be it saliva, biofilm or tooth tissues. A more complex situation, of particular interest, is how the apatite forming ions penetrate into demineralized dentin then precipitate to remineralize it. This study first aim is to determine those apatite and non-apatite forming elements which are leached and the likely quantity. A range of glass-ionomers will be tested against two variables, the level of pH (3.5, 4.5, 5.5 and 6.5) and maturation of the material. The analysis will be done using a range of analysis techniques: Inductively Coupled Plasma (ICP)

chromatography, ion selective electrode (ISE) and ion chromatography (IC). All data will be assembled to provide graphs of the concentration of all ions tested collated at progressive incremental levels, against time periods of collection. In the second phase, it is necessary to study the effects that a bacterial biofilm has in on ion release. In the last phase, the distribution of apatite forming elements (Sr, F, Ca and P) and non-apatite forming elements (Al and Na) in demineralized dentine will be studied using an in vitro model. The new data will be combined with the existing set of data in an attempt to determine the mechanism governing remineralization. The objective of this project is to elucidate the factors which control the ion release from glass ionomers and any associated remineralisation of dentine. The results from this study will add to the body of knowledge in a research area which is new to Singapore. It will also enhance the knowledge and understanding of the ion exchange which occurs with glass-ionomers when used in operative dentistry.

**Clinical significance:** These experiments will assist in the development of new materials for the treatment of deep caries lesion. The ultimate benefit would be the maintenance of pulp vitality in some of these teeth.



Research Collaborations in Financial Year 2007

Collaborating Organizations	Year	Department	Region
Universities			
University of Washington	2006 - 2007	Restorative Dentistry	International
Massachusetts General Hospital, Harvard University, USA	2007 - 2009	Restorative Dentistry	International
University of Texas Health Science Center at San Antonio, USA	2007 - 2009	Restorative Dentistry	International
Nanyang Technological University, Singapore	2006 - 2007	Restorative Dentistry	Local
University of Wisconsin Madison	2003 - 2009	Oral and Maxillofacial Surgery	International
Harvard University	2004 - 2009	Oral and Maxillofacial Surgery	International
Peking University	2007 - 2009	Oral and Maxillofacial Surgery	International
Tsinghua University	2006 - 2009	Oral and Maxillofacial Surgery	International
Zhejiang University	2005 - 2009	Oral and Maxillofacial Surgery	International
Saratov State University, Russia	2004 - 2007	Restorative Dentistry	International
Industries			
Bio-Scaffold	2004 - 2008	Oral and Maxillofacial Surgery	Local
Research Institutes			
SIMTech Pte Ltd, A*STAR	2007 - 2009	Oral and Maxillofacial Surgery	Local
Tissue Modulation Laboratory	2007 - 2009	Preventive Dentistry	Local

## Research Awards & Prizes Awarded in FY 2007/08

Contributor (s) (Staff / Student)	Name of Award	Awarding Agency	International or Local/Regional Award	Title of Article
A/P Lim Lum Peng, Dr. Christina Cheok	Third Place in the International Dental Exhibition and Meeting (IDEM) 2008 Poster Competition	IDEM 2008, Singapore	Regional	Tumor necrosis factor- in diabetics and healthy individuals with untreated periodontal disease
A/P Lim Lum Peng, Dr. Chang Kok Meng	Fourth Place in the International Dental Exhibition and Meeting (IDEM) 2008 Poster Competition	IDEM 2008, Singapore	Regional	The relationship between periodontal parameters and halitosis in patients with untreated periodontitis
A/P Stephen Hsu, Liu Yuanyuan, Teo Choon Meng, Jeremy, Teoh Swee Hin	Best Research Award (Cariology & Mineralized Tissue)	The 21st Annual Scientific Meeting IADR/SEA	Regional	Micro-CT Evaluation of laser-fluoride effect on enamel demineralization
A/P Varawan Sae-Lim, A/P Phan Toan Thang, Henry Yang, Dr. George Yip, Dr Tarun Kumar Maheshwari, Dr. Joseph Antoniraj Jude Aarthi	Senior Travel award (First Runner Up)	The 21st Annual Scientific Meeting IADR/SEA	Regional	Profiling Molecular Signals Elicited by Periodontal Tissue in Avulsion Injury
Dr. Anil Kishen, Dr. Ngo Di Ying, Joanna, Dr. Hu Shijia, Dr. Chia Ai Ping, Vanessa, Dr. Teo Hee Hwa, Daniel	Third Place in the International Category of Poster Presentation	9th Dental Students' Scientific Conference, University of Malaya	Regional	Detection of horizontal and vertical cracks in extracted teeth using a Microbend sensor
A/P Cao Tong, Dr. Boey Mao Jie, Dr. Eng Zhen Feng, Clement, Dr. Ee Zhongren, Jonathan and Dr. Mohamed Shafiq Bin Abdul Aziz	Best Clinical Research Project	South East Asia Association for Dental Education (SEAADE) GC Asia Student Prevention Table Clinician Competition 2007, Bali, Indonesia	Regional	Cytotoxicity Testing of Toothpastes on L929 and CRL-1486 Cells
Dr. Anil Kishen, Dr. Cheng Jean-Lynne, Dr. Lim Tian Wei, William, Dr. Teo Eu Gene, Dr. Wong Jian Zhang, Dr. Lim Zixiang	NUS Outstanding Undergraduate Researcher (OUR) Prize (GROUP category)	NUS	Local	Testing a Light Activated Therapy (ANILAD) to Eliminate Bacteria in Root Canal
A/P Stephen Hsu, Dr. Goh Siew Hor, Dr. Jonathan Foo Kee Yin, Dr. Ivan Prabowo Budiharto, Dr. Tan Mei Na	NUS Outstanding Undergraduate Researcher (OUR) Prize (GROUP category)	NUS	Local	Potential Cariostatic Prevention Effects of Yakult®

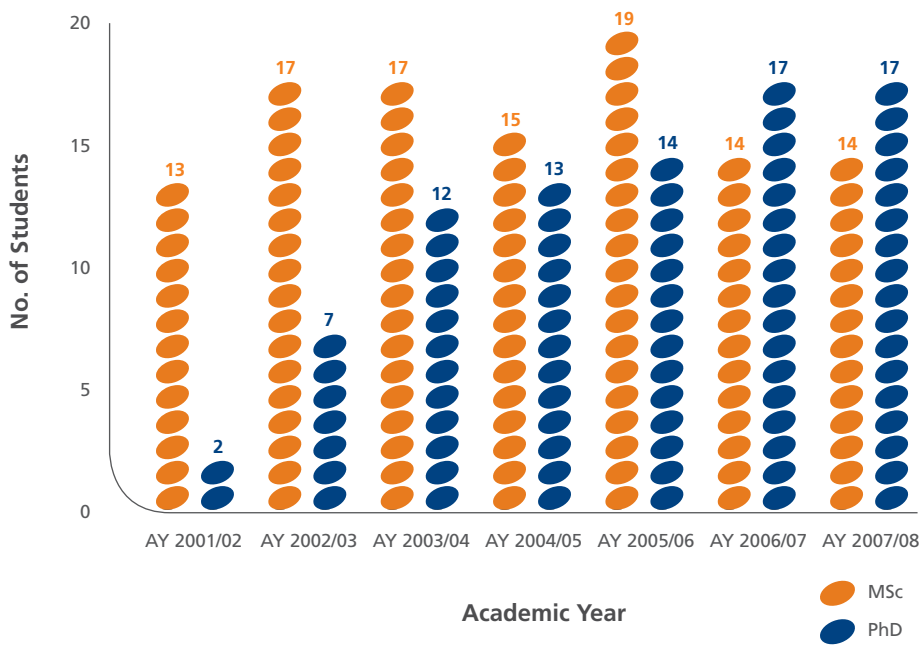
## Patents Awarded in AY 2007/08

Serial Number	Author Names	Department	Title of Patent	Status
112765	1) Renuga Gopal 2) Fujihara Kazutoshi 3) Vijay Kumar Ganesh 4) Prof Seeram Ramakrishna 5) Prof Chew Chong Lin 6) A/P Kelvin Foong 7) A/P Loh Poey Ling	Restorative Dentistry	Fiber reinforced Composite and Methods of Forming the Same	Granted by the Intellectual Property Office of Singapore (IPOS)
122125	1) Dr. Chng Hui Kheng 2) Dr. Tong Yen Wah 3) Adj A/P Adrian Yap	Restorative Dentistry	Viscosity Enhanced Root Repair Material	Granted by the Intellectual Property Office of Singapore (IPOS)

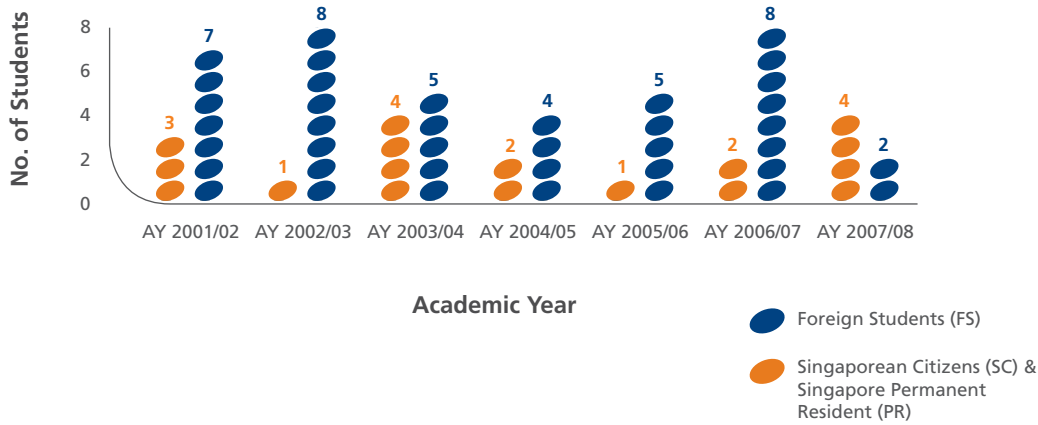
List of Editorial Board Memberships in AY 2007/08

Name of Staff	Name of Journal	Position Held	Journal Tier
A/P Foong Weng Chiong, Kelvin	Singapore Dental Journal	Section Editor	Reputable
A/P Hien Chi Ngo	Journal of Dentistry	Editorial Board Member	Premium
	Oral Health & Preventive Dentistry	Editorial Board Member	Others
A/P Lim Lum Peng	Hong Kong Dental Journal	Editorial Adviser	Others
	Oral Health & Preventive Dentistry	Editorial Board Member	Others
A/P Neo Chiew Lian, Jennifer	Journal of Dentistry	Editorial Board Member	Premium
	Journal of Operative Dentistry	Editorial Board Member	Premium
	Quintessence International	Editorial Board Member	Leading
A/P Ong Hui Lian, Grace	European Journal of Dental Education	Editorial Board Member	Leading
A/P Varawan Sae-Lim	Journal of Endodontics	Editorial Board - Scientific Advisory Panel	Premium
	Journal of Dental Traumatology	Editorial Board Member	Premium
A/P Tan Beng Choon, Keson	Journal of Oral Rehabilitation	Editorial Board Member	Others
Dr Anil Kishen	Singapore Dental Journal	Section Editor	Reputable
	Journal of Applied Oral Science	Editorial Board Member	Others
	Optics and Lasers In Engineering	Editorial Board Member	Leading
	Open Dentistry Journal	Editorial Board Member	Others
	Journal of Endodontics	Editorial Board - Scientific Advisory Panel	Premium
	Craniomandibular Functions	Editorial Board Member	Others
	Journal of Conservative Dentistry	Editorial Board Member	Others
Prof Loh Hong Sai	Singapore Dental Journal	Section Editor	Reputable
Prof Toshio Deguchi	American Journal of Orthodontics & Dentofacial Orthopedics	Editorial Board Member	Premium
	The Angle Orthodontist	Editorial Board Member	Leading

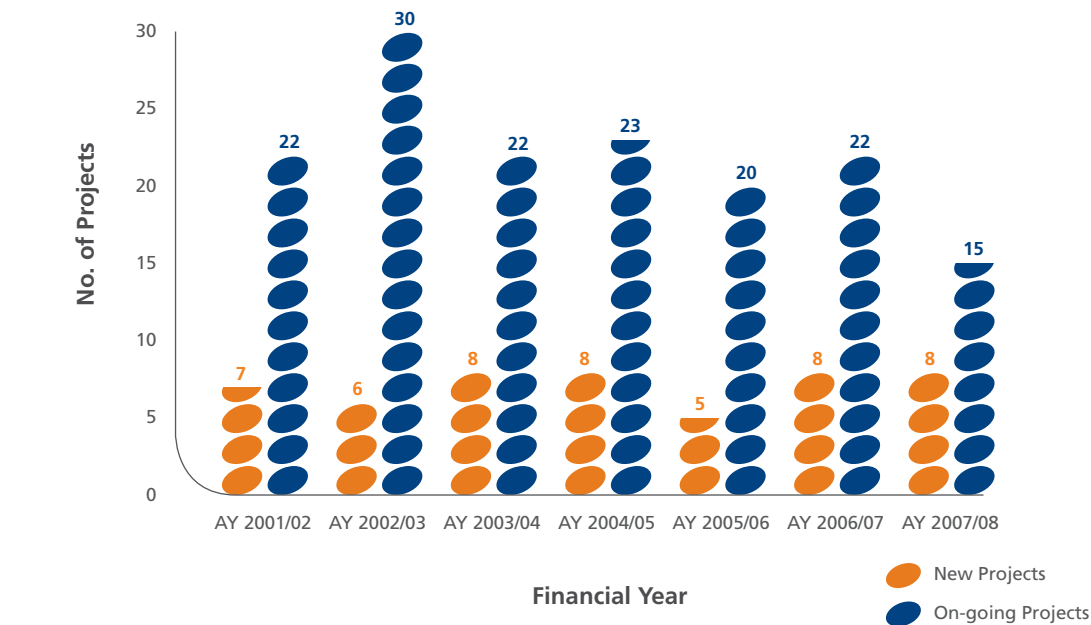
Enrolment of Research Students



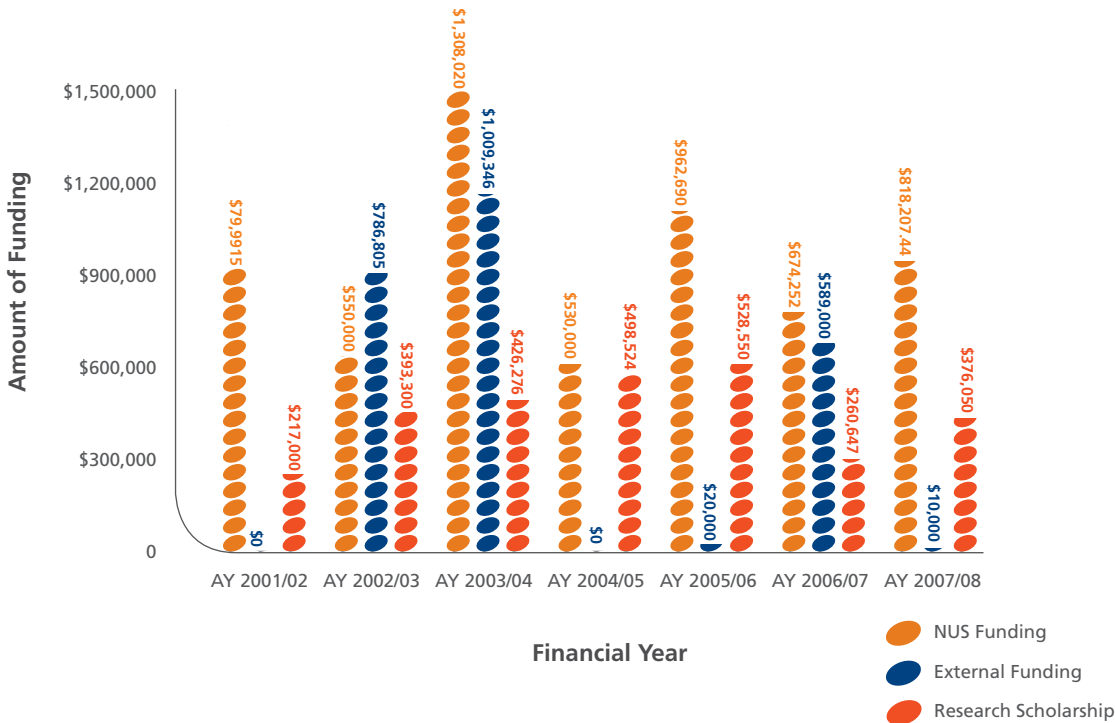
Research Students by Nationality

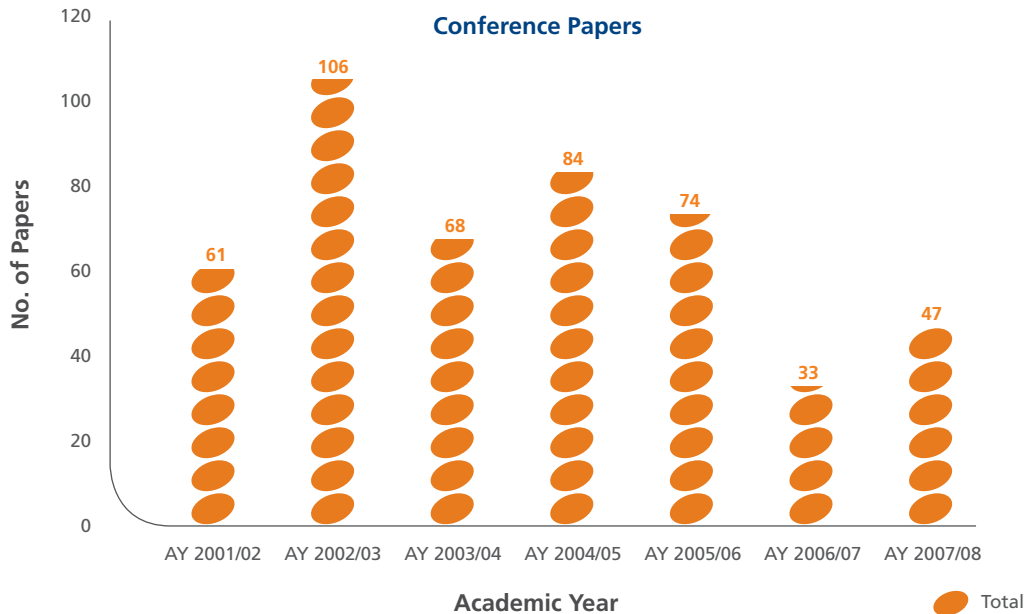
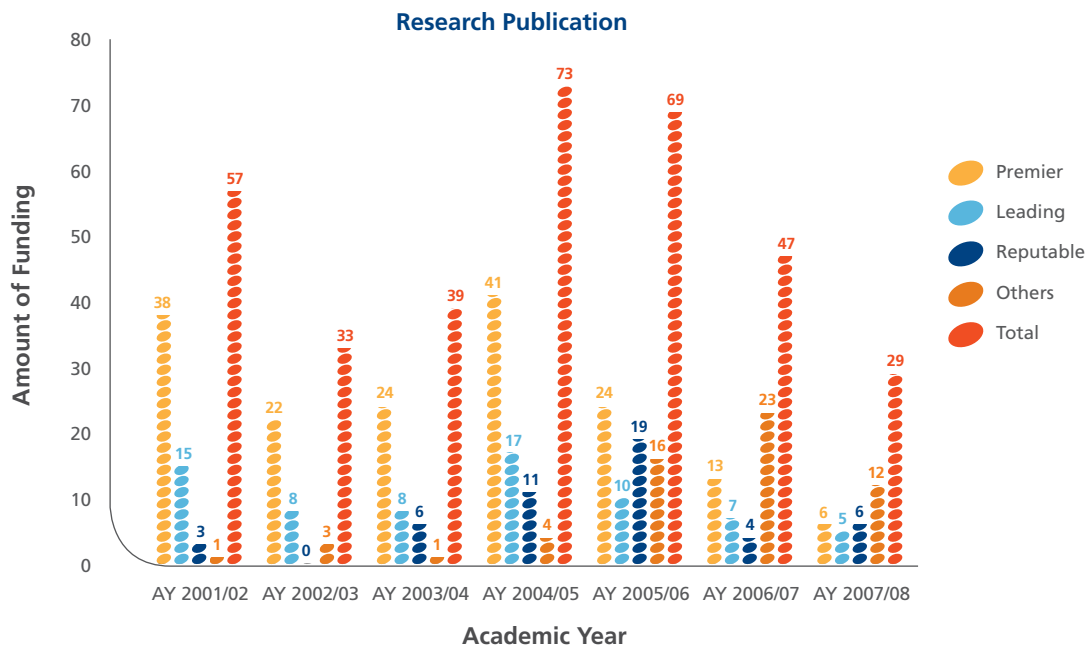


New and On-going Research Projects



Research Fundings





## Research Internships

The Faculty of Dentistry has received numerous students from various schools, both locally and internationally as research interns. These schools include Junior Colleges, Polytechnics, and overseas Universities. The duration of the internship varies from a few months to about a year. The internship can either be full-time or part-time.

These students receive guidance from the academic staff members from the Faculty, by learning and working under the specific research projects of the staff members.



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