The 104th Annual Meeting of the American Academy of Periodontology
in collaboration with the Canadian Academy of Periodontology,
the Japanese Academy of Clinical Periodontology,
and the Japanese Society of Periodontology.

# Abstracts of JACP/JSP Poster Session

October 27-30, 2018
Vancouver Convention Centre, Vancouver, BC, Canada

The Japanese Academy of Clinical Periodontology
The Japanese Society of Periodontology

# 第104回アメリカ歯周病学会共催
# カナダ歯周病学会・日本臨床歯周病学会・
# 日本歯周病学会　2018年大会

**The 104th Annual Meeting of the American Academy of Periodontology**
**in collaboration with the Canadian Academy of Periodontology,**
**the Japanese Academy of Clinical Periodontology,**
**and the Japanese Society of Periodontology.**

## JACP/JSP ポスターセッション抄録集

一般演題（基礎研究，臨床研究），症例報告，歯科衛生士演題

## Abstracts of JACP/JSP Poster Session

**General (Basic Research, Clinical Research), Case Report, Dental Hygiene**

会期：2018年10月27日～10月30日
会場：バンクーバー・コンベンションセンター

October 27-30, 2018
Vancouver Convention Centre, Vancouver, BC, Canada

# 第104回アメリカ歯周病学会共催
# カナダ歯周病学会・日本臨床歯周病学会・
# 日本歯周病学会　2018年大会　JACP/JSP ポスター発表・
# 若手研究者支援協賛企業

---

アサヒプリテック株式会社

医歯薬出版株式会社

長田電機工業株式会社

株式会社ガイドデント

科研製薬株式会社

クインテッセンス出版株式会社

サンデンタル株式会社

ストローマン・ジャパン株式会社

タカラベルモント株式会社

デンツプライシロナ株式会社

株式会社ナルコーム

株式会社白鵬

ヒューフレディ・ジャパン合同会社

ライオン歯科材株式会社

（50音順）

# General
# (Basic Research)

GB-01

## Application of S-PRG nanofillers for novel periodontal therapeutic approach

Hirofumi Miyaji*[1,2], Kayoko Mayumi[2], Erika Nishida[1], Saori Miyata[2], Kanako Shitomi[2], Tomokazu Furihata[2], Yukimi Kanemoto[2], Tsukasa Akasaka[3], Tsutomu Sugaya[2]

[1]Clinic of Endodontics and Periodontics, Hokkaido University Hospital.
[2]Department of Periodontology and Endodontology,
[3]Department of Biomaterials and Bioengineering, Faculty of Dental Medicine, Hokkaido University

*Background and objective:* Surface pre-reacted glass-ionomer（S-PRG）fillers release six types of ions, such as F, Bo and Sr, to exert tooth remineralization and antibacterial effects. In present study, we designed the tooth surface coating technique with S-PRG nanofillers and conducted the antibacterial examinations.
*Materials and Methods:* S-PRG fillers（Shofu Inc., Kyoto, Japan）mixed with distilled water was fractionated by sedimentation, and the collected supernatant was used as dispersion of S-PRG nanofillers with an average particle size of 500 nm. Ion-releasing ability, cytocompatibility and antibacterial activity of S-PRG nanofillers were evaluated. The adhesion of the nanofillers on tooth surface was also examined using human tooth block（approval number: 17-222）after immersion into the nanofiller dispersion and subsequent ultrasonic cleaning（10 sec）. As a preclinical study, nanofiller dispersion was injected into periodontal pockets of 12 premolars with experimental periodontitis created in beagle dogs, and the percentage of sites with bleeding on probing（BOP%）was measured postoperatively.
*Results:* S-PRG nanofillers released 6 types of ions and possessed great antibacterial activity as well as low cytotoxicity. In adhesion test, nanofillers stably and uniformly adhered to dentin and cementum surface. Preclinical study revealed that the BOP% remarkably decreased after S-PRG nanofiller application. At 4 weeks postoperation, BOP% of S-PRG and control（no application）groups was 12.1% and 52.8%, respectively.
*Conclusion:* The coating technique with S-PRG nanofillers would acquire the antibacterial tooth surface. This study may provide a novel treatment strategy for periodontal disease.

GB-02

## Effects of parathyroid hormone and neutral self-assembling peptide on periodontal healing

Wataru Yoshida*[1], Takahiro Takeuchi[1], Takahiro Bizenjima[1], Daisuke Matsugami[1], Toshikatsu Nakazaki[2], Fumi Seshima[1], Atsushi Saito[1,3]

[1]Department of Periodontology, Tokyo Dental College, [2]Nakazaki Dental Clinic,
[3]Oral Health Science Center, Tokyo Dental College

*Background and objective:* Administration of intermittent parathyroid hormone（PTH）has been shown to enhance periodontal healing. Self-assembling peptides（SAP）hydrogels have similar biological properties to extracellular matrix（ECM）and can be used as a 3-dimentional scaffold. The purpose of this study is to investigate the effects of systemic intermittent administration of PTH and local application of a neutral SAP hydrogel（SPG-178）on the healing of surgically created periodontal defects in rats.
*Materials and Methods:* Micro-structure of SPG-178 was observed with a scanning electron microscope（SEM）. Ten weeks-old male Wistar rats were randomly assigned to two subgroups: PTH administration and saline group. In each animal, two standardized periodontal defects were surgically created mesially of the maxillary first molars. The defects were treated with SPG-178 or left unfilled. Micro-CT, histological and immunohistochemical analyses were performed.
*Results:* SEM observation showed nanofiber and network structures, similar to native ECM. In the unfilled group, the PTH administration yielded significantly greater bone volume fraction（BV/TV）than saline control at 4 weeks（$p<0.05$）. PTH + SPG-178 group showed significantly greater BV/TV than saline + unfilled group at 2 and 4 weeks（$p<0.05$）. At 4 weeks, histological assessment showed that newly formed bone in the defect area in the PTH + SPG-178 group appeared to be greater than other groups. Proportion of PCNA-positive cells above newly formed bone in the PTH + SPG-178 group was significantly greater than that in the saline + unfilled at 2 weeks（$p<0.01$）.
*Conclusion:* These findings suggest that the PTH administration and the SPG-178 application enhance periodontal healing.

GB-03

## Calcium metabolism in periodontium with experimental periodontitis of rats

Ryutaro Kuraji*[1,2], Ya-Hsin Wu[2], Shuichi Hashimoto[3], Saki Mishiro[2], Hiroshi Ito[2], and Yukihiro Numabe[2]

[1]The Department of Life Science Dentistry, The Nippon Dental University,
[2]The Department of Periodontology, School of Life Dentistry at Tokyo, The Nippon Dental University,
[3]The Nippon Dental University

*Background and objective:* A ligature placement for tooth is a method to produce the experimental periodontitis in rodents. However, previous studies mostly have not reported calcium metabolism in the periodontal tissue inflamed. In the present study, the $^{45}Ca$ dynamics in the tissue was analyzed after ligature placement.
*Materials and Methods:* The maxillary right first molar tooth（M1）in eight-week-old male Wistar rats was ligated with silk suture for 1, 3, 7, and 28 days; a ligation side. While the left M1 without ligature placement was utilized as a control side. $^{45}CaCl_2$ was applied intraperitoneally 24 hr before the terminal point. The rat left-and-right palatal mucosa, molar teeth, and maxillas were anatomically separated and collected. The $^{45}Ca$ concentration in each section was determined with a liquid scintillation counter. Moreover, amounts and bone mineral density（BMD）of the alveolar bone were analyzed by μCT.
*Results:* At 1 day after ligature placement, $^{45}Ca$-levels incorporated into each section in the ligation side significantly decreased by 10% as compared with that in the control side, but those at 3 and 7 day conversely increased. The amount and BMD of alveolar bone in the ligation side were lower than the control side from 3 to 7 days, but increased to near the control value after 28 days.
*Conclusion:* There was correlation between the $^{45}Ca$-levels incorporated and BMD in the alveolar bone. These results suggested that $^{45}Ca$ uptake into the periodontium per 24 hr was consistent with the bone remodeling in the experimental periodontitis.

GB-04

## Cyclic stretch inhibits IL-1β secretion by attenuating NLRP3 inflammasome activation

Eiji Nemoto*[1], Kentaro Maruyama[1], Yukihiko Sakisaka[1], Mizuki Suto[1], Hiroyuki Tada[2], Takashi Nakamura[3], Satoru Yamada[1]

[1]Department of Periodontology and Endodontology, [2]Department of Oral Immunology,
[3]Department of Dental Pharmacology, Tohoku University Graduate School of Dentistry

*Background and objective:* Macrophages are immune cells that play diverse roles in host defenses and tissue homeostasis. In mechanical microenvironments such as periodontal ligament, macrophages receive mechanical signals that regulate various cellular functions. However, the mechanisms by which mechanical signals influence the phenotype and function of macrophages in the process of inflammation have not yet been elucidated in detail. We examined the effects of cyclic stretch on NLR family, pyrin domain-containing 3（NLRP3）inflammasome activation in J774.1, a murine macrophage cell line, and mouse primary bone marrow-derived macrophages.
*Materials and Methods:* Gene and protein levels were assessed by real-time polymerase chain reaction analysis and enzyme-linked immunosorbent assay（ELISA）or Western blot analysis, respectively.
*Results:* Cyclic stretch inhibited adenosine monophosphate（ATP）-stimulated interleukin（IL）-1β secretion in lipopolysaccharide（LPS）-primed macrophages using ELISA and Western blot analyses. Cyclic stretch-mediated inhibition of IL-1β secretion was caused by the suppression of caspase-1 activity. The addition of compound C, a specific inhibitor of adenosine monophosphate-activated protein kinase（AMPK）, to LPS-primed macrophages inhibited IL-1β secretion as well as caspase-1 activation, suggesting that AMPK signaling is involved in ATP-triggered IL-1β secretion. Furthermore, the phosphorylation of AMPK induced by ATP in LPS-primed macrophages was significantly suppressed by cyclic stretch, indicating that cyclic stretch negatively regulates IL-1β secretion through the inhibition of caspase-1 activity by attenuating the AMPK pathway.
*Conclusion:* Our results suggest that mechanical stress functions to maintain homeostasis through the prevention of excessive inflammasome activation in macrophages in mechanical microenvironments such as periodontal ligament.

GB-05

## LIPUS inhibits IL-1β secretion through NF-κB signaling pathway in macrophages

Mizuki Suto*, Eiji Nemoto, Kentaro Maruyama, Yukihiko Sakisaka, Satoru Yamada

Department of Periodontology and Endodontology, Tohoku University Graduate School of Dentistry

*Background and objective:* Low-intensity pulsed ultrasound（LIPUS）, which acts on cells as a mechanical stress, has been reported that LIPUS treatment has a potential to activate osteogenesis. However, little is known regarding an effects of LIPUS treatment on inflammation. In this study, we examined the effect of LIPUS on J774.1, a murine macrophage cell line, stimulated with *E. coli* lipopolysaccharide（LPS）.
*Materials and Methods:* J774.1macrophages were maintained in RPMI-1640 containing 10% fetal bovine serum, 100 μg/ml penicillin-streptomycin. Cells were stimulated using LIPUS-generating device（ITO Co., Tokyo, Japan）. LIPUS signal consisted of a series of 1.5MHz and was delivered at an intensity of 30 or 60mW/cm$^2$. The cells were treated with LIPUS for 60 min and further cultured up to 6 h and then analyzed by real-time PCR, Western blot, and ELISA.
*Results:* Real time PCR revealed that treatment of cells for 6 h with LPS upregulated the gene expression levels of pro-IL-1β. This expression was inhibited by the first-60 min treatment of LIPUS under the condition of 60 mW/cm$^2$ and 1.5 MHz. LIPUS treatment significantly inhibited the expression of IL-1β triggered by LPS plus ATP at protein levels as assessed by ELISA and Western blot analysis. Furthermore, LIPUS treatment significantly inhibited the LPS-induced protein expression of phosphorylated-p65 NF-κB by Western blot.
*Conclusion:* Those finding suggest that LIPUS exerts anti-inflammatory effects on LPS-stimulated J774.1 by inhibiting NF-κB signaling. LIPUS may be powerful therapeutic tool for the NF-κB-associated diseases such as periodontal diseases.

GB-06

## Application of microperforated titanium mesh for tissue engineered regeneration

Yukihiko Sakisaka*[1], Kentaro Maruyama[1], Jingyu Zhang[1], Hiroshi Ishihata[1], Eiji Nemoto[1], Keiichi Sasaki[1], Takeshi Hatsuzawa[2], Satoru Yamada[1]

[1]Department of Periodontology and Endodontology, Tohoku University Graduate School of Dentistry,
[2]Laboratory for Future Interdisciplinary Research of Science and Technology, Institute of Innovative Research, Tokyo Institute of Technology

*Background and objective:* Currently, pore sizes of titanium mesh for the graft retainer applied to osteogenic regenerative surgery have a range from 50μm up to 5mm. Therefore, it cannot avoid that soft tissues intrude to the ready occupied space for the bone regeneration. We fabricated a new style retainer as the titanium membrane with piercing pores of 25×25 micron square. This study was aimed to apply the titanium membrane as a tissue barrier and cell-interactive 3D scaffold for periodontal regeneration therapy. We investigated the performance of migration of cells cultivated on the fabricated material.
*Materials and Methods:* Mouse pre-osterblastic MC3T3-E1 cells were cultured on the pure titanium membrane with 20μm thickness and 25μm ×25μm square through-holes at 75μm intervals. We evaluated the growth and characters of cells on the titanium membrane by immunocytochemistry and observation under SEM.
*Results:* Histological observations of the cells cultured for 96 hours detected orientation to produce pseudopodiums of the cell bodies to be attached to the edge around the square holes. Certain of cells migrated and gathered their bodies into the holes and retained in the through path. The tissue layer covered with the cells on the titanium surface in a sheet-like configuration was further organized and thickened for 1 month cultivation without any separation from the titanium mesh.
*Conclusion:* The morphological effect of micro array of square holes on the titanium membrane assisted growth and migration of cells. The membrane might be applicable to the periodontal surgery as using a barrier membrane for tissue regeneration.

GB-07

## Combination therapy using iPS cells and EMD for periodontal regeneration

Yukino Hisanaga*[1,3], Eiichi Suzuki[1], Hideto Aoki[1], Masahiro Sato[1], Akiko Saito[2,3], Toshifumi Azuma[2,3], Atushi Saito[1,3]

[1]Department of Periodontology, Tokyo Dental College, [2]Department of Biochemistry, Tokyo Dental College, [3]Oral Health Science Center, Tokyo Dental College

*Background and objective:* Induced pluripotent stem cells（iPSCs）are a candidate cell source in periodontal regenerative therapy. We previously reported the combined use of enamel matrix derivative（EMD）with atelocollagen sponge（ACS）scaffold enhances the early stage of osteoblastic differentiation of mouse induced pluripotent stem cells（miPSCs）in vitro. In the present study, we evaluated the effect of the combination healing of bone defects in vivo.
*Materials and Methods:* miPSCs were established from fibroblasts harvested from subcutaneous tissues of 4 weeks old male ICR mice. Following embryonic body formation, dissociated miPSCs were seeded onto ACS with or without EMD. There were cultured in osteoblast differentiation medium（OBM）for 14 days. Critical-sized（5 mm）calvarial defects were created in the parietal bone of 12 weeks old male mice. Mice were assigned to 4 different groups: unfilled, ACS, ACS + miPSCs and ACS + EMD + miPSCs. The healing was evaluated by micro computed tomography, histological analysis（HE and von Kossa staining）.
*Results:* At 7 days, compared to the unfilled group, healing of bone defect was observed in ACS, ACS + miPSCs and ACS + miPSCs + EMD groups. ACS + miPSCs and ACS + miPSCs + EMD groups showed a greater level of von Kossa staining at 14 days compared to the ACS group.
*Conclusion:* Combined use of miPSCs with ACS and EMD may promote healing of bone defects in vivo.

GB-08

## Effect of UV irradiation of titanium and zirconia on cultured cells

Takuya Hama*[1], Hirotsugu Morinaga[1], Takumi Sato[1], Yuta Shimizu[1], Tadashi Yasuda[1], Yasuaki Hotta[2], Yukimichi Tamaki[3], Toshiaki Shibutani[1]

[1]Asahi University School of Dentistry Department of Periodontology,
[2]Asahi University School of Dentistry Central Research Institute of Oral Science,
[3]Asahi University School of Dentistry Department of Dental Materials

*Background and objective:* Zirconia is garnering attention as an implant material that could replace titanium for implants. During implant procedures, it is important to protect the region where epithelial tissues come in contact with the implant material. The aim of this study was to investigate the efficacy of the use of excimer UV irradiated zirconia and titanium disks for early attachment to epithelial progeny cells.
*Materials and Methods:* Titanium and zirconia（ZPEX）, were used for this study. Each disk was polished before the experiment so that both disks had the same surface roughness. The disks were irradiated with excimer UV for 20 minutes and surface roughness and surface wettability were analyzed. Human gingival epithelial progenitor cells（CELLnTEC CO.）were seeded onto each disk. Each disk was then cultured in an incubator using media for 1, 3, 12, and 24 hours. Cells remaining attached to each disk were analyzed using a scanning electron microscope.
*Results:* Contact angle measurements indicated that the group in which disks were irradiated with excimer UV showed a significantly smaller contact angle and became hydrophilic. Cell counts for attached cells on excimer UV irradiated disks increased significantly compared to the number on disks that had undergone polishing alone between 1 to 24 hours after the initiation of incubation.
*Conclusion:* Results showed that excimer UV irradiation of the surface of titanium disks and zirconia disks promotes the early attachment of human gingival epithelial progenitor cells to the disks, thus suggesting that irradiation is effective for the early attachment of epithelial cells to the disks.

GB-09

## Effect of parathyroid hormone on osteoblastic differentiation of iPS cells

Masahiro Sato*[1], Eiichi Suzuki[1], Hideto Aoki[1], Yukino Hisanaga[1], Ayano Nakamura[1], Atsushi Saito[1,3], Toshifumi Azuma[2,3]

[1]Department of Periodontology, Tokyo Dental College, [2]Department of Biochemistry, Tokyo Dental College, [3]Oral Health Science Center, Tokyo Dental College

*Background and objective:* Parathyroid hormone（PTH）plays an important role in bone remodeling, and different actions have been reported depending on its administration method. Induced pluripotent stem cells（iPSCs）are promising as a cell source for regeneration of periodontal tissue due to their ability of proliferation and pluripotency. However the effects of PTH on iPSCs remain mostly unknown. The purpose of this study is to investigate the effects of PTH on osteoblastic differentiation of iPSCs in vitro.
*Materials and Methods:* Mouse iPSCs（miPSCs）were seeded on unadhesive plate to generate embryonic body（EB）. EBs were treated with 0.05% trypsin EDTA and seeded into atelocollagen sponge（ACS）in osteoblast differentiation medium（OBM）. Cells were divided into three groups: control, continuous PTH exposure（PTH-C）, and intermittent PTH exposure（PTH-I）groups. Alkaline phosphatase（ALP）staining and real-time PCR were performed on day 0, 7, 14 for evaluation of osteoblastic differentiation.
*Results:* Degree of ALP staining in PTH-I group appeared to be higher than the control group and PTH-C group. In real time PCR, expression of *Col1a1* in the PTH-I group on the day 14 was significantly higher（$p < 0.001$）than the other groups. There was a time-dependent increase in the expression of Osx in all groups. On day 14, the level was higher in the PTH-I group than other groups.
*Conclusion:* These results suggested that intermittent administration of PTH promotes early osteoblastic differentiation of miPSCs.

GB-10

## HLA genome editing in human dental pulp cells using ZFN

Shuhei Otari*[1], Hitomi Aoki[2], Yuta Shimizu[1], Tomoko Kawaguchi[3], Toshiyuki Shibata[3], Takahiro Kunisada[2],Toshiaki Shibutani[1], Ken-ichi Tezuka[2]

[1]Asahi University School of Dentistry Department of Periodontology, [2]Gifu University Graduate School of Medicine Department of Tissue and Organ Development Regeneration and Advanced Medical Science, [3]Gifu University Graduate School of Medicine Department of Oral and Maxillofacial Science

*Background and objective:* Human leukocyte antigen（HLA）plays a role in distinguishing between self and non-self. Currently, construction of a cell bank for transplants is underway by collecting dental pulp cells from individuals with HLA-A, B, and DRB1 which are haplotype homozygous and creating iPS cells from the cells. However, large costs and time are required to create the cell bank. Therefore, construction of cells carrying quasi-HLA haplotype homozygosity and induction of iPS cells were attempted by genome editing using zinc finger nucleases（ZFNs）.
*Materials and Methods:* A ZFN plasmid which specifically cleaves the HLA-A*02 locus was introduced into dental pulp cells（HLA-A*02, 33; B*44, -; DRB1*13,-）. The cells were fluorescent labelled with anti-HLA-A*02-FITC antibody and the HLA-A*02 minus fraction was isolated using FACS followed by expansion of culture containing the cell fraction. Genes were amplified by PCR and analyzed by sequencing. Next, Yamanaka 4 factors were introduced into the cells using the Sendai viral vector（Cytotune-iPS2.0）to induce iPS cells.
*Results:* Fraction with decreased expression of HLA-A*02 molecules was obtained by FACS in the ZFN-treated group. Results obtained by gene sequence analysis of cells in the fraction revealed presence of deletions and insertions specific to the HLA-A*02 allele, as well as presence of homologous recombination with the HLA-C and H regions. Currently, analysis of gene sequences of the HLA region in iPS cells induced using the SeV vector is underway.
*Conclusion:* ZFN specifically cleaved the HLA-A*02 region and induced deletions, insertions, and homology-directed repair mutations.

GB-11

## The effects of theaflavins on experimental periodontitis in rats

Ya-Hsin Wu*[1], Ryutaro Kuraji[1,2], Yuji Taya[3], Hiroshi Ito[1], Yukihiro Numabe[1]

[1]Department of Periodontology, The Nippon Dental University School of Life Dentistry at Tokyo
[2]Department of Life Science Dentisitry, The Nippon Dental University
[3]Department of Pathology, The Nippon Dental University School of Life Dentistry at Tokyo

*Background and objective:* Theaflavins (TF), the major polyphenol in black tea, has shown the ability to reduce inflammation and bone resorption. The aim of this study was to evaluate the effects of TF on experimental periodontitis in rats.
*Materials and Methods:* Thirty Wistar rats were divided into 5 groups: Control (glycerol; without ligation), Ligature (glycerol; with ligation), TF1 (1 mg/ml TF; with ligation), TF10 (10 mg/ml TF; with ligation), and TF100 (100 mg/ml TF; with ligation). To induce experimental periodontitis, ligatures were placed around maxillary first molars bilaterally. After that, rats were topically applied daily with 100 µl of glycerol or TF for 7 days. Bone resorption was assessed by micro-computed tomography (Micro-CT). Expression of mRNA for proinflammatory and osteoclastogenesis-related cytokines were evaluated using real-time RT-PCR in gingival tissue. H&E and TRAP staining were conducted for inflammation and osteoclast analysis.
*Results:* TF10 and TF100 groups but not TF1 caused significant inhibition of alveolar bone loss, reduction of inflammatory cells infiltration in the periodontium, as well as significantly reduced the numbers of TRAP-positive osteoclasts when compared to Ligature group. Furthermore, bone mineral density was increased in TF100 group than in Ligature group. Correspondingly, TF10 and TF100 groups significantly downregulated the gene expression of interleukin (*IL*)*-6*, *IL-8*, matrix metalloproteinase-9 (*Mmp-9*) and receptor activator of NF-κB ligand (*Rankl*), but not that of osteoprotegerin (*Opg*).
*Conclusion:* The present study suggests that topical application of TF reduce inflammation and bone resorption on experimental periodontitis. Therefore, TF may have therapeutic potential in periodontal disease.

GB-12

## Treatment of OPG-deficient mice with WP9QY recovers alveolar bone loss

Yuki Ozaki*[1], Masanori Koide[2], Nobuyuki Udagawa[2], Nobuo Yoshinari[1]

[1]Department of Periodontology, [2]Institute for Oral Science, Matsumoto Dental University

*Background and objective:* Osteoblasts express two key molecules for osteoclast differentiation, receptor activator of NF-κB ligand (RANKL) and osteoprotegerin (OPG). RANKL induces osteoclastogenesis, while OPG inhibits it by blocking the binding of RANKL to RANK. OPG-deficient ($OPG^{-/-}$) mice exhibit severe alveolar bone loss with enhanced bone resorption. WP9QY (W9) peptide binds to RANKL and blocks RANKL-induced osteoclastogenesis. Here, we show that treatment with W9 restores alveolar bone loss in $OPG^{-/-}$mice
*Materials and Methods:* W9 or risedronate (a bisphosphonate) was subcutaneously administered to 12-week-old $OPG^{-/-}$ mice. First, we analyzed of alveolar bone loss by *in vivo* µCT images. Secondary, we analyzed bone morphometry, histomorphometry, and immunocytochemistry of alveolar bone.
*Results:* Administration of W9 or risedronate to $OPG^{-/-}$mice significantly decreased the osteoclast number in the alveolar bone. Interestingly, treatment with W9, but not risedronate, enhanced Wnt/β-catenin signaling and induced alveolar bone formation in $OPG^{-/-}$mice. Expression of sclerostin, an inhibitor of Wnt/β-catenin signaling, was significantly lower in tibiae of $OPG^{-/-}$mice than in wild-type mice. Treatment with risedronate recovered sclerostin expression in $OPG^{-/-}$mice, while W9 treatment further suppressed sclerostin expression. Histomorphometric analysis confirmed that bone formation-related parameters in $OPG^{-/-}$mice, such as osteoblast number, osteoblast surface and osteoid surface, were increased by W9 administration but not by risedronate administration. These results suggest that treatment of $OPG^{-/-}$mice with W9 suppressed osteoclastogenesis by inhibiting RANKL signaling and enhanced osteoblastogenesis by attenuating sclerostin expression in the alveolar bone.
*Conclusion:* W9 may be a useful drug to prevent alveolar bone loss in periodontitis.

GB-13

## IL-1β enhances cell adhesion through laminin 5 and β4 integrin in gingival epithelial cells

Masaru Mezawa*, Yorimasa Ogata

Department of Periodontology, Nihon University School of Dentistry at Matsudo

*Background and objective:* Junctional epithelium（JE）attaches to the tooth enamel with hemidesmosomes. Laminin 5 and α6β4 integrin are important adhesion molecules of internal basal lamina of JE. IL-1β is a pro-inflammatory cytokine which mediates degradation of extracellular matrix through the functions of matrix metalloproteinases. The purpose of this study was to examine the effects of IL-1β on adhesion molecules in gingival epithelial cells.
*Materials and Methods:* Human epithelial cell line Ca9-22 cells were cultured in α-MEM containing 10% fetal calf serum until 70-80% confluent. Ca9-22 cells were stimulated by IL-1β（1 ng/ml）and examined mRNA levels of type Ⅳ collagen, α1 type Ⅰ collagen, cadherin 1, β4 integrin and laminin 5 subunits such as laminin-α3, -β3, -γ2 chain. Ca9-22 cells were co-stained by β4 integrin and laminin 5 antibodies, β4 integrin antibody and phalloidin（actin filaments), respectively. Immunostained co-localization images were analyzed using a confocal microscopy. The extent of co-localization was analyzed by Pearson correlation analysis.
*Results:* β4 integrin, laminin-α3, -β3, -γ2 chain, type Ⅳ collagen, α1 type Ⅰ collagen and cadherin 1 mRNA levels were increased by IL-1β（1 ng/ml）at 1 h. Laminin-γ2, type Ⅳ collagen and α1 type Ⅰ collagen mRNA levels were decreased by IL-1β at 3 h. IL-1β increased 1.4-fold co-localization of β4 integrin and laminin 5 after 1 h stimulation by IL-1β. There was no change of co-localization of actin filaments with β4 integrin after IL-1β stimulation.
*Conclusion:* These results suggest that co-localization of β4 integrin and laminin 5 was increased by IL-1 β.

GB-14

## Effect of implant placement on ectopically-induced trabecular bone

Shoji Miyazono*, Yoko Miyazaki, Junro Yamashita

Fukuoka Dental College, Graduate School

*Background and objective:* We were successful to induce massive cancellous bone formation in the tibial diaphysis by performing bone marrow penetration followed by daily parathyroid hormone（PTH）administration in rats. However, since the diaphysis is a hollow structure, it was thought that such an ectopically formed bone would be resorbed. Here, we report that implant placement in the ectopically formed bone prevents bone resorption.
*Materials and Methods:* Fourteen rats received bone marrow penetration using a round bur in the tibiae followed by daily PTH administration for 14 days. At day-14 half of rats received titanium screw implant placement. The remaining half received sham surgeries. Rats were euthanized at 21 days after the implant placement. Micro-computed tomographic and histological analyses were performed to assess the effect of implant placement in the ectopically formed bone.
*Results:* As anticipated, bone marrow penetration in the tibial diaphysis followed by PTH therapy induced tremendous cancellous bone formation in the bone marrow cavity of the diaphysis. No new bone was formed in the diaphysis without marrow penetration. While the ectopically formed bone was mostly resorbed after the cessation of PTH therapy, significant bone was observed when implants were placed. Such a bone was not newly formed due to implant placement since implant placement into the intact diaphysis caused minimum bone formation around the implants.
*Conclusion:* Implant placement slows bone resorption significantly in the ectopically induced bone. Findings suggest that augmented bone can be maintained by implant placement.

GB-15

## Acid-resistance and antibacterial properties of laser-processed and fluoride-incorporated apatite layer

Kanako Shitomi*[1], Hirofumi Miyaji[1], A. Joseph Nathanael[2], Maki Nakamura[2], Ayako Oyane[2], Tsutomu Sugaya[1]

[1]Department of Periodontology and Endodontology, Faculty of dental Medicine, Hokkaido University
[2]Nanomaterials Research Institute, National Institute of Advanced Industrial Science and Technology（AIST）

*Background and objective:* Laser-assisted biomimetic（LAB）process enables rapid and area-specific calcium phosphate（CaP）coating on the artificial substrate, by applying low energy pulsed laser irradiation to the substrate immersed in a supersaturated CaP solution. In the present study, sintered hydroxyapatite disc（sHA）was subjected to the LAB process in a supersaturated solution supplemented with different concentrations of fluoride ions. We assessed acid-resistance and antibacterial activity of the LAB-processed sHA.
*Materials and Methods:* sHA was immersed in a supersaturated CaP solution supplemented with NaF（0, 1 and 3 mM）and then irradiated with Nd: YAG laser light（355 nm, 6W/cm2）for 30 minutes. The irradiated surface was characterized by SEM, TEM, and EDX. To assess acid resistance, the LAB-processed sHA was immersed in a citric acid buffer for 3 days, and Ca and P in the buffer were quantified by ICP. To assess the antibacterial properties, *Streptococcus mutans*（*S. m*）was seeded on the LAB-processed sHA and anaerobically cultured for 12 hours. SEM observation of *S. m* and measurements of turbidity and colony forming unit were carried out.
*Results:* CaP layer was formed on the laser-irradiated region of sHA. By adding NaF to the supersaturated CaP solution, fluoride was incorporated in the CaP layer and its acid resistance was improved. Addition of NaF decreased the number of *S. m* on the LAB-processed sHA, turbidity of medium and colony number of bacteria.
*Conclusion:* Fluoride-incorporated CaP layer fabricated by the LAB process exhibited the acid resistance and antibacterial effects.

GB-16

## Involvement of TRPV4 in bone remodeling by mechanical stress with periodontitis

Yoshimasa Taketani*[1], Takafumi Suzuki[1], Joichiro Hayashi[1], Kohei Hayashi[1], Makiko Ishii[1], Hideharu Otsuka[1], Junichi Tatsumi[1], Yoshiyuki Hakeda[2], Kitetsu Shin[1]

[1]Division of Periodontology, Department of Oral Biology and Tissue Engineering, Meikai University School of Dentistry,
[2]Division of Oral Anatomy, Department of Human Development and Fostering, Meikai University School of Dentistry

*Background and objective:* In clinical situations, excessive occlusal force with periodontitis can cause rapid alveolar bone resorption, but the molecular mechanism remains unclear. In this study, we focused on transient receptor potential vanilloid 4（TRPV4）, a mechanosensitive ion channel expressed on osteoblasts that is thought to contribute to bone mass maintenance. We investigated the role of TRPV4 under chronic inflammation to clarify the molecular mechanism of alveolar bone resorption with occlusal trauma.
*Materials and Methods:* Osteoblasts were isolated from the calvariae of 3- to 7-day-old C57BL/6J mice and cultured for 7 days in osteogenic medium（α-MEM containing 10% fetal bovine serum（FBS）, 0.1% ascorbic acid, and 0.2% β-glycerophosphate）with or without *Porphyromonas gingivalis* lipopolysaccharide（*P.g*-LPS）. The expression level of TRPV4 and the receptor activator of NF-κB ligand（RANKL）/osteoprotegerin（OPG）ratio at day 7 were determined by real-time PCR. Subsequently, cell growth was arrested at day 7 by incubation for 12 hours in α-MEM containing 0.5% FBS, then 4α-phorbol 12-13-dicaprinate（4α-PDD）, a selective agonist of TRPV4, was added to the medium under the assumption of mechanical stress. After 24 hours, the RANKL/OPG ratio was determined.
*Results:* At day 7, the expression level of TRPV4 and the RANKL/OPG ratio did not differ between the groups. However, after the addition of 4α-PDD, the RANKL/OPG ratio increased significantly in the cells cultured with *P.g*-LPS.
*Conclusion:* These results suggest that *P.g*-LPS enhances the response of TRPV4 and increases the RANKL/OPG ratio. TRPV4 may be involved in the bone remodeling induced by mechanical stress with periodontitis.

GB-17

## Influence of antimicrobial photodynamic therapy for LPS and bacteria

Meri Fukaya*, Yuji Matsushima, Ayaka Suzuki, Yoko Nakano, Satoshi Shirakawa, Takatoshi Nagano, Kazuhiro Gomi

Department of Periodontology Tsurumi University School of Dental Medicine

*Background and objective:* Active oxygen is generated by toluidine blue irradiated with the excitation light.These effects are applied to the dental field as the antimicrobial photodynamic therapy（a-PDT）. The purpose of this study is to evaluate the alteration of root surface and organic degradation and antibacterial action by a-PDT treatment.
*Materials and Methods:* In this study, FotoSan630（FS）and toluidine blue was used for a-PDT. The root surface, which performed SRP, was treated by a-PDT and observed by SEM. The organic resolution was evaluated by degradation of LPS and albumin. After a-PDT treatment（1min, 3min, 5min）, LPS was detected using LPS detection kit and albumin was evaluated by electrophoresis. Antibacterial action was evaluated by bacterial colony count after treatment by a-PDT.
*Results:* The deposition of root surface was reduced the test group compared with the control. Decomposition of LPS and albumin was found to be significantly reduced compared with control at 5 min of irradiation but did not completely decompose. The antimicrobial effect suppressed almost all the bacteria in 5 minutes of light irradiation.
*Conclusion:* The a-PDT can be applied to periodontal therapy because it is able to clean the root surface without damage, to degrade the LPS and organics and to have bactericidal potency.

GB-18

## Replacement of junctional epithelium by oral epithelium

Mayu Kato*[1,2], Ryo Aizawa[1], Junichi Tanaka[2], Sara Yajima-Himuro[1], Tatsuaki Seki[1], Keisuke Tanaka[1,2], Kenji Mishima[2], Matsuo Yamamoto[1]

[1]Department of Periodontology, School of Dentistry, Showa University
[2]Division of Pathology Department of Oral Diagnostic Sciences, School of Dentistry, Showa University

*Background and objective:* Junctional epithelium（JE）is originated from odontogenic epithelium. JE is believed to be gradually replaced by oral epithelium（OE）in a lifetime. However, the detailed process of replacement remains still unclear. The aim of this study was to clarify replacement of JE by OE using a GFP-positive tooth germ transplantation method.
*Materials and Methods:* GFP-positive tooth germs were transplanted into the hole of alveolar bone in C57BL/6-KI（$ROSA^{mT/mG}$）mice, which ubiquitously express tdTomato red fluorescent. On day 50, 140 and 200 after transplantation, JE around transplanted GFP-positive teeth was histologically analyzed. Next, the palatal region of JE around the transplanted GFP-positive teeth in C57BL/6 mice were excised. On day 30 after excision, the newly formed JE was histologically analyzed.
*Results:* In transplanted teeth, the dental pulp, the periodontal ligament and the JE expressed GFP on day 50. On day 140, GFP-positive JE was partly replaced by tdTomato-positive epithelium and completely replaced on day 200, while there was no difference in the expressions of integlin β4 and laminin5 between JEs before and after replacement by OE. In addition, there was no GFP-positive cell in the regenerated JE after excision and the regenerated JE also expressed both integlin β4 and laminin5.
*Conclusion:* These results confirm that JE derived from odontogenic epithelium are gradually replaced by OE. And the partly removed JE after gingivectomy is not cured by the surrounding JE, but OE.

GB-19

## Optical dissection of the mitophagy dynamics in HPDL cells

Motozo Yamashita*[1], Mio Suzuki[2], Kuniko Ikegami[1], Tomomi Nakamura[1], Arisa Nishikawa[2], Koji Miki[1], Manabu Yanagita[3], Masahiro Kitamura[2] and Shinya Murakami[2]

[1]Department of Periodontology, Osaka University Dental Hospital,
[2]Department of Periodontology, Osaka University Graduate School of Dentistry,
[3]Department of Oral Health, Kobe Tokiwa Junior College

*Background and objective:* Mitochondria is a representative organelle as a power generator for ATP supply and is critical for the proper cell metabolism. However, defective mitochondria generate excessive Reactive Oxygen Species（ROS）, which induce the damage of cellular proteins and DNA. Mitophagy, a part of autophagy is the cell clearance system for the damaged mitochondria with selective degradation. Previous study revealed that mitochondrial dysfunction correlated with senescent cells and chronic systemic diseases, involving metabolic syndrome that affect the progression of periodontitis. Thus, we aimed to dissect the mitophagy dynamics in senescent human periodontal ligament（HPDL）cells at single cell level, by using the newly developed pH responsive fluorescence probes.
*Materials and Methods:* Mitochondrial morphology was observed by transmission electron microscopy. Mitochondrial membrane potential was measured by a cationic dye, JC−1 labeling. Mitophagy dynamics in HPDL cells was evaluated by using mt−mkeima−Red which is a coral−derived acid−stable fluorescent protein tagged with mitochondria targeting signal peptide. Basal activity of autophagy in HPDL cells was quantified with a novel autophagic flux probe, GFP−LC3−RFP−LC3ΔG. Turnover of the GFP/RFP ratio indicates the autophagy dynamics at single cell scale.
*Results:* Senescent HPDL cells showed dysregulated mitochondrial morphology and membrane potential. Fluorescent autophagic probes dissected the defective mitophagy and ROS induction in senescent HPDL cells at single cell level.
*Conclusion:* Failure of mitophagy may induce the accumulation of damaged mitochondria thereby induce excessive ROS in senescent HPDL cells. Activation of impaired mitophagy may be a promising therapeutic target for the periodontitis in elderly populations.

GB-20

## Senescence−associated miR−34a accelerates periodontal stem−cell aging by targeting SATB2

Kuniko Ikegami*[1], Motozo Yamashita[1], Mio Suzuki[2], Arisa Nishikawa[2], Tomomi Nakamura[1], Koji Miki[1], Jirouta Kitagaki[2], Manabu Yanagita[3], Masahiro Kitamura[2] and Shinya Murakami[2]

[1]Department of Periodontology, Osaka University Dental Hospital,
[2]Department of Periodontology, Osaka University Graduate School of Dentistry,
[3]Department of Oral Health, Kobe Tokiwa Junior College

*Background and objective:* Periodontitis is characterized in part as an age−dependent chronic inflammatory disease with periodontal tissue destruction caused by dental plaque. Accumulation of environmental stress, such as bactericidal infection, Reactive Oxygen Species（ROS）and traumatic occlusal force is thought to induce senescence in periodontal tissue at cellular level. We previously reported that senescent human periodontal ligament cells（HPDL）secrete various inflammatory cytokines, referred to as the senescence−associated secretory phenotype（SASP）. A comprehensive analysis of microRNA identified miR−34a which is preferentially expressed in senescent HPDL as a regulator of inflammation. Noteworthy, bioinformatics analysis revealed that special AT−rich sequence−binding protein 2（SATB2）, a critical transcription factor for osteoblastogenesis and cleft palate deformity as a target of miR−34a. In this study, we aimed to explore the SATB2 regulation by miR−34a in senescent HPDL with focusing the periodontal stem−cell aging.
*Materials and Methods:* Replicative senescence was conducted to primary HPDL. Irreversible cell cycle arrest was judged with population doublings periods. SASP−related genes, IL−6, IL−8 and MMPs were examined by RT−qPCR and ELISA. A screening of miRNAs was performed using the human miRNAs array（Agilent）. Synthetic double strand mimics or competitive inhibitor oligos for hsa−miR−34a were induced in HPDL to clarify the miR−34a function.
*Results:* Senescent HPDL showed the decreased expression of stem−cell marker, SSEA−3 and hard tissue forming ability. While miR−34a was elevated, SATB2 expression was decreased in senescent HPDL. Exogenous miR−34a treatment suppressed the osteoblastic differentiation of HPDL by targeting SATB2.
*Conclusion:* miR−34a−SATB2 pathway may participate in stem−cell aging in senescent HPDL.

GB-21

## Concomitant suppression of osteoclasts and macrophages hinders osseous wound healing

Masahiro Hirano*, Hiroki Fujimoto, Akimichi Sugihara, Junro Yamashita

Fukuoka Dental College Graduate School

*Background and objective:* Concomitant use of antiresorptives with immunosuppressants increases a risk of osteonecrosts of the jaw（ONJ）after tooth extractions. However, precise mechanisms are unclear. The purpose of the study was to investigate the effect of macrophage depletion on oral osseous wound healing in animals on antiresorptive therapy.
*Materials and Methods:* Young adult mice received zoledronic acid（ZA）injections for 7 weeks. To suppress macrophages clodronate liposomes（CL-lipo）were given to mice. Tooth extractions were performed at 4 weeks after the intiation of the ZA and CL-lipo administration. Visual and micro-computed tomographic（mCT）evaluation was conducted every week to assess wound healing. Flow cytometric analysis of bone marrow and blood cells were performed to assess F4/80（+）Gr1（-）macrophages.
*Results:* It was confirmed that macrophages decreased in the bone marrow and blood. Bone fill in the extraction sockets was compromised in the CL-lipo/ZA group compared to control 3 weeks. However, at 2 and 4 weeks after tooth extractions bone fill was similar between two groups. Soft tissues healing delayed in the CL-lipo/ZA group compared to control. Monotherapy of either CL-lipo or ZA did not result in a delay in tooth extraction socket healing.
*Conclusion:* Since the combined administration of CL-lipo and ZA induced impaired osseous wound healing and monotherapy did not,it was considered that the macrophage depletion and antiresorptive therapy has a synergistic negative effect on osseous wound healing.

GB-22

## α-Cyano-4-hydroxycinnamic acid suppresses bone resorption

Hiroko Imai*[1,2], Kentaro Yoshimura[2], Yoichi Miyamoto[2], Masahiro Chatani[3], Masamichi Takami[3], Ryutaro Kamijo[2], Matsuo Yamamoto[1]

Departments of [1]Periodontology, [2]Biochemistry, and [3]Pharmacology, Showa University School of Dentistry

*Background and objective:* Monocarboxylate transporters（MCTs）are proton-linked membrane transporters. Among the 14 MCT subtypes, MCT-1, -2, -3 and -4 transport lactate and pyruvate across the plasma membrane. MCTs are involved in energy metabolism of muscle and nerve cells. However, their roles in osteoclasts are not reported. Here we analyzed the role of MCTs in osteoclast differentiation.
*Materials and Methods:* Bone marrow cells（BMC）isolated from 5- to 8-week-old male ddY mice were cultured for 3 days in the presence of M-CSF to obtain macrophages（BMM）. BMM were treated for 3 days with M-CSF and RANKL to induce osteoclastogenesis. We examined the effects of α-cyano-4-hydroxycinnamic acid（CHC）, an inhibitor of MCTs, on RANKL-induced osteoclastogenesis. Osteoclastogenesis was evaluated by activity staining of tartrate-resistant acid phosphatase（TRAP）and determination of TRAP activity. Osteoclasts differentiated in co-cultures of BMC and mouse osteoblastic UAMS32 cells were cultured on dentine disks in the presence or absence of CHC to examine the effect of CHC on bone resorbing activity of osteoclasts.
*Results:* BMM and osteoclasts expressed mRNAs for *Mct1*, *Mct2*, and *Mct4*. While the expression of *Mct1* was abundant in both BMM and osteoclasts, that of *Mct2* increased after differentiation into osteoclasts. CHC enhanced osteoclast differentiation from BMM. However, CHC suppressed bone resorbing activity of osteoclasts.
*Conclusion:* The results described above indicate that transport of monocarboxylates via MCTs plays important roles in differentiation and function of osteoclasts. Our findings also suggest a possibility that MCTs become a new therapeutic target for bone metabolic diseases.

GB-23

## Effect of Antiangiogenic agents in tooth extraction socket healing

Hiroki Fujimoto*, Masahiro Hirano, Akimichi Sugihara, Junro Yamashita

Fukuoka Dental College Graduate School

*Background and objective:* A combined administration of thalidomide（Thal）, bortezomib（Vel）, and zoledronic acid（ZA）is often used to treat multiple myeloma. In this particular population a risk of osteonecrosis of the jaw（ONJ）, which is impaired osseous wound healing in the oral cavity, is high. Thalidomide is known to have antiangiogenic effect. There are reports that the use of antiangiogenic agents increases a risk of ONJ. However, how antiangiogenesis plays a role in the development of ONJ is unclear.
*Materials and Methods:* Mice received a combination therapy of Thal/Vel/ZA for 7 weeks. In control, mice received a combination of Thal/Vel for 7 weeks. Tooth extractions were performed at 3 weeks after the initiation of the therapy. Visual and micro-computed tomographic（μCT）evaluation was conducted every week to assess wound healing. Flow cytometric analysis was performed to assess CD31（+）CD309（+）vascular endothelial cells in circulation. To further confirm antiangiogenic effects, tumor inoculation（B16F10 cells）were performed in another group of mice. Those mice received Thal/Vel therapy and tumor blood vessel density was assessed.
*Results:* Number of circulating CD31（+）CD309（+）cells, which are referred to as vascular endothelial cells, decreased in mice on Thal/Vel therapy regardless of ZA. Thus, Thal/Vel therapy suppressed angiogenesis. Oral wound closere was good in both groups. However, the μCT assessment revealed a significant difference in the bone fill at 7-day post-extraction.
*Conclusion:* Antiangiogenesis may not play a dominant role in the development of ONJ-like impaired wound healing in mice.

GB-24

## The effect of neutropenia and anti-resorptives in tooth extraction socket healing

Hikaru Takeyama*, Akimiti Sugihara, Hiroki Fujimoto, Masahiro Hirano, Shoji Miyazono, Junro Yamashita

Fukuoka Dental College Graduate School

*Background and objective:* Cyclophosphamide is used to suppress immume responses in patients receiving bone marrow transplant. Bone marrow suppression induces a decrease in neutrophil numbers. Bone trauma and infection cause necrotic bone formation. Neutrophil infiltration occurs in the connective tissue adjacent to the necrotic bone. Since the neutrophil infiltration is followed by the resorption of the necrotic bone, there would be interactions between neutrophils and osteoclasts. However, their synergistic roles during osseous healing are unclear. Here, neutropenia and osteoclast suppression were induced using Cyclophosphamide and bisphosphonate（ZA）and their effect on tooth extraction socket healing was investigated.
*Materials and Methods:* Mice were administered Cyclophosphamide for 7 weeks. Flow cytometric analysis of F4/80（-）Ly6c/6g（-）cells in the bone marrow, spleen, and blood was performed to confim neutropenia. Micro-computed tomographic（μCT）analysis of tibiae was performed to confim the antiresorptive effect of ZA. After the 4 weeks of treatment, tooth extraction of the maxillary first molars were conducted and bone fill in the sockets were assessed using in vivo μCT. Mice were euthanized at 4 weeks post-extractions.
*Results:* In vivo μCT assessment found no difference in bone fill in the extraction sockets between the experimental and control groups although neutropenia and the suppression of bone resorption were observed in the experimental groups. Epithelial wound closure occurred in 2 weeks post extractions in both groups with no differences.
*Conclusion:* These results suggest that the combination of neutropenia and the suppression of bone resorption does not have detrimental effect on tooth extraction socket healing in mice.

GB-25

## IL-1β induced by dental calculus promotes osteoclastogenesis in RAW-D cells

Atsutoshi Yoshimura*, Jorge Luis Montenegro Raudales, Ziauddin SM, Kanako Higuchi, Yukio Ozaki

Department of Periodontology and Endodontology, Nagasaki University Graduate School of Biomedical Sciences

*Background and objective:* We have shown that cellular uptake of dental calculus triggers nucleotide-binding oligomerization domain-like receptor family pyrin domain-containing 3（NLRP3）inflammasome activation, leading to the processing of the interleukin（IL）-1β precursor into its mature form in mouse macrophages. The purpose of the present study was to examine whether IL-1β induced by dental calculus in mouse macrophages can promote osteoclastogenesis in RAW-D cells.

*Materials and Methods:* Wild-type and NLRP3-deficient mouse macrophages were stimulated with dental calculus for 8 hours and the concentration of IL-1β in the culture supernatants was measured by ELISA. Then, these supernatants were added to the culture of RAW-D cells pretreated for 2 days with 10 ng/ml of receptor activator of nuclear factor kappa-B ligand（RANKL）. After 3 days, tartrate-resistant acid phosphatase（TRAP）staining was performed, and the TRAP-positive cells with 3 or more nuclei were counted as osteoclasts.

*Results:* Dental calculus induced IL-1β secretion in wild-type, but not NLRP3-deficient, mouse macrophages. RANKL-pretreated RAW-D cells incubated with the culture supernatants of wild-type mouse macrophages stimulated with dental calculus had significantly higher number of TRAP-positive cells compared to the cells incubated with the culture supernatants of NLRP3-deficient mouse macrophages stimulated with dental calculus.

*Conclusion:* Dental calculus induced the production of IL-1β via NLRP3 in mouse macrophages. IL-1β contained in the culture supernatants of wild-type mouse macrophages promoted osteoclastogenesis in RAW-D cells. These results suggest that dental calculus may contribute to periodontal tissue destruction by accelerating alveolar bone resorption.

# General
# (Clinical Research)

GC-01

## The Clinical Trial of Periodontitis Index Software - PerioPerio -An Interim Report-

Yoshio Motegi*

Motegi dental practice

*Background and objective:* We created periodontitis index software - PerioPerio - based on an algorithm of the following formula:

SL = - 203.9 F + 281.2（correlation coefficient: －0.8826）

SL: Lost periodontal ligament area

F: Anatomical coefficient

For this PerioPerio, we conducted clinical trials for 5 years to investigate the effectiveness in the diagnosis or screening of periodontitis.

*Materials and Methods:* We examined two values at 41,281 existing teeth of 916 patients 55 years old or older suffering from periodontal disease. A value at one point of the deepest point of the attachment level of target tooth was measured in mm unit. Then, the operator independently diagnosed the test target tooth to select a value of 0 to 4 from the five levels of guideline values: P0 to P4 of the periodontitis classification（Japanese Association for Dental Science, 2007）. 10 clinicians conducted it once a year for 5 years. Therefore, the target tooth was measured several times. At the time when two and a half year has elapsed, intermediate results are aggregated. For the analysis, the PerioPerio values vs. P0 to P4 were compared by F test and t-test. This clinical trial was conducted with an ethical review of The Japanese Academy of Clinical Periodontology.

*Results:* When comparing a PerioPerio value in each periodontitis classification of P0 to P4（unpaired t-test）, significance（$P < 0.01$）was observed in all groups.

*Conclusion:* As a result of comparing, clinical effectiveness of PerioPerio was suggested in diagnosis or screening of periodontitis.

GC-02

## The association of anti-phospholipid antibody with Behcet's disease

Mari Mori*[1], Toshiyuki Nagasawa[2], Osamu Uehara[3], Satsuki Kato[4], Izuru Sato[5], Kenichi Namba[6], Masayuki Tsutsumi[5,6], Nobuyoshi Kitaichi[5,6], Yasushi Furuichi[4].

[1]Division of General Dental Sciences, Department of Oral Rehabilitation, [2]Division of Advanced Clinical Education, Department of Integrated Dental Education, [3]Division of Disease Control and Molecular Epidemiology, Department of Oral Growth and Development, [4]Division of Periodontology and Endodontology, Department of Oral Rehabilitation, School of Dentistry, [5]Department of Ophthalmology, Health Sciences University of Hokkaido, [6]Department of Ophthalmology, Faculty of Medicine and Graduate School of Medicine, Hokkaido University

*Background and objective:* Anti-phospholipid antibody induces vascular thrombosis, resulting in myocardial infarction, stroke, and fetal loss. Behcet's disease is an inflammatory disorder characterized by recurrent oral aphthous ulcers, genital ulcerations, and ocular manifestations. Several investigators reported that anti-pospholipid antibody was increased in periodontitis patients, and periodontitis was severe in Behcet's disease patients. We have reported that periodontitis induced anti-pospholipid antibody, thorough the molecular mimicry between oral bacteria and beta-2 glycoprotein I. The purpose of this study was to evaluate the anti-pospholipid antibody in Behcet's disease patients.

*Materials and Methods:* Patients with Behcet's disease（n=15）and healthy subjects（n=20）were recruited in this study. Venous blood was collected from the subject, and antibody titer against beta-2 glycoprotein I dependent and/or independent anti-pospholipid antibody was measured. Whole saliva was collected from the patients with Behcet's disease. Meta-genome analysis was performed using DNA prepared from the patient's saliva.

*Results:* Anti-phospholipid antibody was elevated in Behcet's disease patients compared with the healthy subjects. Meta-genome analysis revealed that bacteria that bear molecular mimicry with human beta-2 glycoprotein I were detected in the saliva from the Behcet's disease patients.

*Conclusion:* Present study suggested that anti-pospholipid antibody was increased in the Behcet's disease patients, and several oral bacteria might induce the antibody through the molecular mimicry with beta-2 glycoprotein I.

GC-03

## Effectiveness of Improved Dental Model for Training of Pocket Probing

Yusuke Harada*[1], Masayo Sunaga[1], Yasuo Takeuchi[2], Akira Aoki[2], Shogo Maekawa[2], Yuichi Izumi[2], Atsuhiro Kinoshita[1]

[1]Department of Educational Media Development, Graduate School of Medical and Dental Sciences; Curricular Management Division, Institute of Education, Tokyo Medical and Dental University
[2]Department of Periodontology, Graduate School of Medical and Dental Sciences, Tokyo Medical and Dental University

*Background and Objective:* Since 2008, we have been developing a training model simulating the structure of the bottom of periodontal pockets and reported that it is effective for training of periodontal pocket probing. The aim of this study was to evaluate the effectiveness of our latest dental model, which has been improved in terms of manufacturing cost, versatility and durability.
*Materials and Methods:* Study participants included 43 fourth year dental students（students）, and 22 dental residents（residents）. We divided 24 artificial teeth into four groups of six teeth each; designated as groups A, B, C, and D. Participants probed these groups of teeth according to a randomly assigned order. For each group, participants self-verified their probing pocket depths against setup depths in order to improve their probing skill. Each Probing time was recorded. After completing the probing, participants answered a questionnaire to evaluate the improved model.
*Results*: Probing accuracy rates increased significantly from 79%（median）on the first probing to 85% on the last probing in students, and from 81% to 86% in residents（$p<0.01$）. Probing time reduced significantly from 423 s on the first probing to 295 s on the last probing in students, and from 309 s to 267 s in residents（$p<0.001$）. Questionnaire results showed that most participants, 98% of students and 100% of residents, considered the pocket probing training using this model effective.
*Conclusion:* Pocket probing training using this improved dental model was effective for improving probing accuracy and time.

GC-04

## Periodontal status in Japanese institutionalized elderly, covering 10 years

Satoshi Sekino*[1], Takeshi Kikutani[2], Fumiyo Tamura[2], Yukihiro Numabe[1]

[1]Department of Periodontology, School of Life Dentistry at Tokyo, The Nippon Dental University,
[2]Tama Oral Rehabilitation Clinic, The Nippon Dental University

*Background and objective:* The present study describes periodontal status in Japanese institutionalized elderly over the 10 years.
*Materials and Methods:* In 2006, a cross-sectional study was performed in 6 nursing home in Taito-ku,Tokyo in Japan. A total of 123 dentate residents aged 56-101 years were given a clinical examination comprising recording of plaque index（PII）, bleeding on probing（BoP）, clinical attachment level（CAL）and probing pocket depth（PPD）in 4 sites of all teeth present except third molars. Basic demographic information was also collected. In 2016, another cross-sectional study was performed in 180 dentate residents in 6 nursing homes in Taito-ku, Tokyo .
*Results:* The mean number of teeth increased from 12.2±8.2 to14.6±8.8 over 10 years. In 2006, proportion of surface which harboring visible plaque was 69.3% ±26.0. It was decreased to 53.6% ±31.7. The mean proportion of teeth with CAL ≥7mm in 2006 and in 2016 were 13.9% ±22.2 and 14.0% ±20.9, respectively. According to the CDC/AAP case definition, prevalence of severe periodontitis were 29.4% in 2006 and 29.1% in 2016.
*Conclusion:* Periodontal disease was still prevalent in current Japanese institutionalized elderly even though oral hygiene status were improved and the remained teeth were increased comparing with 2006. The new proper public health programs are necessary to establish.

GC-05

## Analysis of failed implants with peri-implantitis

Sho Sakamoto*[1], Takashi Kado[1,2], Yasushi Furuichi[1]

[1]Division of Periodontology and Endodontology, Department of Oral Rehabilitation,
[2]Division of Dental Education Development, Department of Integrated Dental Education, School of Dentistry,
Health Sciences University of Hokkaido

*Background and objective:* Prevalence of Peri-implantitis is reported as high rates. But we are less well understood the cause of peri-implantitis. In this study, prevalence of various microorganism around failed implants and physicochemical characteristics of such implant surfaces were investigated.
*Materials and Methods:* A total of 6 patients with 13 failed implants were recruited. Clinical parameters were obtained prior to the implant removal surgeries. At the time of implant removal, plaque samples were obtained by inserting sterile paper points into the sulcus or pockets. The paper points were placed in a sterile tube and the removed implants were putting into the bottle filled with formalin. Then, quantitative real-time polymerase chain reaction（PCR）was carried out to analyze the microbiologic composition. Additionally, X-ray fluorescence analysis（XRF）was performed to analyze the elements of removed implant surfaces.
*Results:* The prevalence rate of *P.gingivalis*（*P.g*）, *T.denticola*（*T.d*）, *T.forsythia*（*T.f*）, *F.nucleatum*（*F.n*）and *P.intermedia*（*P.i*）around the failed implant sites was significantly higher than that of the healthy tooth sites. The presence of zinc was showed from 10 failed implant surfaces, and only found in the subjects with multiple implant failures.
*Conclusion:* This study suggested that the amount of Red complex, *F. n*, and *P. i.* were associated with failed implants with peri-implantitis. Significance of zinc observed on the failed implant surfaces in the initiation and/or progression of peri-implantitis should further be investigated.

GC-06

## The association between periodontitis and preterm low birth-weight in women with gestational diabetes

Yoko Koide*[1], Anna Osamura[2], Kyoko Kohashi[2], Tomohiro Oba[3], Yoshimasa Okamatsu[5], Katsufumi Otsuki[4], Yasubumi Maruoka[5], Akihiko Sekizawa[3], Tsutomu Hirano[2], Matsuo Yamamoto[1]

[1]Department of Periodontology, Showa University School of Dentistry, [2]Division of Diabetes, Metabolism, and Endocrinology, Department of Internal Medicine, Showa University School of Medicine, [3]Department of Obstetrics and Gynecology, Showa University School of Medicine, [4]Department of Obstetrics and Gynecology, Showa University Koto Toyosu Hospital, [5]Department of Special Needs Dentistry, Division of Community Based Comprehensive Dentistry, Showa University School of Dentistry

*Background and objective:* There have been little studies about the association between periodontitis and preterm low birth-weight infants in women with gestational diabetes（GDM）. The objective of this prospective cohort study was to investigate whether periodontal disease was risk factors of the preterm low birth-weight infants in women with GDM.
*Materials and Methods:* Fifteen women with GDM were enrolled into study. Before treatment of GDM, we performed oral examinations, and collected samples from blood and saliva. All subjects were received usual treatment of GDM. After delivery, medical records were consulted to determine each infant's birth weight and gestational age. Subjects were assigned to either good or poor periodontal condition.
*Results:* There was none preterm. 9 subjects were poor periodontal condition. 2 subjects of low birth-weight were all poor periodontal condition. The number of *T. denticola* was significantly higher in the poor periodontal condition. Triglyceride level was significantly higher in the good periodontal condition. On the other hand, all 5 subjects who had GDM diagnosis in 1st trimester were poor periodontal condition. Diabetes related markers were not significantly different between poor and good periodontal condition. Glycoalbumin level was significantly higher in subjects who had GDM diagnosis in 1st trimester, but total cholesterol, LDL cholesterol, and triglyceride level was significantly higher in subjects who had GDM diagnosis in 3rd trimester.
*Conclusion:* In this study, the effect of periodontal condition on gestational age was not investigated. It was suggested that should be considered time of GDM diagnosis and lipid metabolism in future study.【250 words】

GC-07

## A new screening system for periodontitis using Hepatocyte growth factor

Shogo Maekawa*[1], Shinta Suzuki[1], Akira Aoki[1], Sayaka Katagiri[1], Yuichi Ikeda[1], Kenichiro Ejiri[2], Sophannary Kong[3], Mizuki Nagata[4], Yoko Yamaguchi[5], Mitsuhiro Ohshima[6], Yuichi Izumi[1]

[1]Department of Periodontology, Graduate school of Medical and Dental Sciences, Tokyo Medical and Dental University, [2]Dental Clinic Eji, [3]Dentistry Department, Faculty of Health Sciences, University of Puthisastra, [4]Department of Orthodontics and Pediatric Dentistry, University of Michigan School of Dentistry, [5]Department of Biochemistry, Nihon University School of Dentistry, [6]Department of Biochemistry, Ohu University School of Pharmaceutical Sciences

*Background and objective:* Clinical diagnosis of periodontitis is performed by assessment of probing pocket depth, bleeding on probing, tooth mobility as well as radiographic examination. These conventional procedures are beneficial for precise diagnosis, however, they are impractical for medical mass screening. The aim of this study was to analyze the relationship between the level of hepatocyte growth factor（HGF）in oral rinse water samples and clinical parameters of periodontitis, and to evaluate the potential of a prototype HGF immunochromatographic paper test strip（p-HGF-TS）for diagnostic screening of periodontitis, in comparison with a commercially-available occult blood（hemoglobin）test strip（Hb-TS）.
*Materials and Methods:* One hundred twenty-five subjects were classified by Biofilm-Gingival Interface（BGI）classification（Offenbacher *et al*, 2007）. Rinse water samples were collected and clinical periodontal parameters were recorded. The presence of HGF and hemoglobin（Hb）in each sample was detected using a p-HGF-TS and an Hb-TS. And then, the concentrations of HGF and Hb was evaluated.
*Results:* The HGF concentrations in rinse water samples showed significant positive correlations with clinical parameters of periodontitis. The positive rate and read value on p-HGF-TS showed significantly high values especially in severe periodontitis compared to healthy subjects. Hb-TS showed generally higher positive rates than p-HGF-TS; however, it also showed false positive results in healthy subjects.
*Conclusion:* The concentration of HGF in oral rinse water showed close association with the progress of periodontitis and the p-HGF-TS has a potential for diagnosis of periodontitis although further refinement of the test strip is required.

GC-08

## Accelerated inflammation in peripheral artery disease patients with periodontitis

Keitetsu Kure*[1], Norio Aoyama[2], Hiroki Sato[1], Yuichi Izumi[1]

[1]Department of Periodontology, Tokyo Medical and Dental University
[2]Department of Oral Interdisciplinary Medicine, Kanagawa Dental University

*Background and objective:* Peripheral arterial disease（PAD）is arteriosclerosis in extremity that involves ischemia. Previous studies report that patients with periodontitis had a high risk of PAD, however evidence regarding the relationship between the two diseases has not yet been fully elucidated. In this cross-sectional study, we investigated the relationship by comparing the patients with PAD to those with arrhythmia（ARR）as a control.
*Materials and Methods:* Patients with PAD（n=25）or ARR（n=25）, who visited Tokyo Medical and Dental University Hospital, were examined. We recorded the dental clinical finding, which includes probing pocket depth（PPD）, bleeding on probing（BOP）, clinical attachment level（CAL）and the number of missing teeth, and collected periodontal bacteria and peripheral blood samples. We adjusted age, gender, prevalence of diabetes, hypertension, dyslipidemia, obesity and smoker rate between PAD and ARR groups. Real-time PCR was performed to detect bacterial counts, and ELISA method was used to measure anti-bacterial antibody titers and pro-inflammatory cytokine levels in serum. We used Student's t test, Chi-square test and Mann-Whitney's U test for the comparisons.
*Results:* PAD patients had more missing teeth（18.4 ± 2.0）and higher serum levels of C-reactive protein（1.57 ± 0.85 mg/dl）and tumor necrosis factor-alpha（70.3 ± 5.7 pg/ml）than ARR patients（12.0 ± 1.7, 0.38 ± 0.21 mg/dl, 39.3 ± 4.5 pg/ml, respectively）. Meanwhile, there was no statistical difference in other dental clinical measurements, bacterial antibody titers and bacterial counts between the groups.
*Conclusion:*
It was suggested that PAD patients had worsened oral and periodontal condition with enhanced systemic inflammation.

# Case Report

R-01

## Periodontal tissue regeneration around the tooth with orthodontic extrusion

Masahiko Kanenari*

Crystal Dental office

*Background and objective:* Prolonged retention of deciduous tooth may interrupt developing oral function including establishing lateral guidance. Attached gingiva of the extruded permanent tooth is reported to be lost after the forced extrusion, however we experienced the good treatment result with the impacted mandibular canine accompanied with the odontogenic cyst and supernumerary teeth.
*Case:* The patient was 24-year old female with the spaced maxillary anterior dentition. Impacted mandibular right canine with the odontogenic cyst and supernumerary teeth was extruded orthodontically. Bone defect and gingival recession with the loss of attached gingiva have been improved by periodontal regenerative therapy.
*Clinical procedure and outcome: The impacted canine was extruded by elastic with the lingual hook on the labial surface after extracting persistent* deciduous tooth and removing odontogenic cyst and supernumerary teeth. Malalignment was corrected by the full mouth orthodontic treatment. After elevated trapezoidal full- and partial-thickness flap to access bone defect, root plaining was performed. Enamel matrix derivative was applied and the bone graft material was placed on the labial side. Connective tissue harvested from palate was fixed more coronal to CEJ with sling suture after the graft site was covered with resorbable membrane fixed with periosteal suture. Then the flap was repositioned and sutured to cover connective tissue completely.
*Conclusion:* Esthetic problem by the lost attached gingiva and bone defect caused by the orthodontic extrusion and supernumerary teeth extraction was improved by root coverage procedure and periodontal regenerative therapy.

R-02

## A case report of comprehensive treatment including periodontal regenerative therapy, implant treatment, and prosthodontic treatment involving a patient with extensive severe chronic periodontitis

Hiroshi Funaki*

Hibiya Dental office

*Background:* Periodontal tissue regenerative therapy, implant treatment, and prosthetic treatment were performed, and favorable results were obtained.
*cases:* The patient was a 33-year-old male with chief complaints of repetitive full-mouth gum swelling and pain, and masticatory disturbance due to the mobility of teeth. Radiographic findings showed full-mouth horizontal bone resorption, and some vertical resorption. Intraoral findings included marked mobility of the overall teeth, a large amount of subgingival calculus, swollen gums, and deep periodontal pockets. The patient's general condition was favorable, and he was a non-smoker.
*Clinical Procedures and Outcomes:* Regenerative therapy using Enamel matrix derivative（EMDOGAIN） was performed for the lower preservable teeth, and tooth extraction and implant treatment were performed for the lower non-preservable teeth. All upper teeth were considered non-preservable, and, therefore, the maxilla was treated with a full-denture.
*Conclusion:* Gum swelling and pain, which were the patient's chief complaints, disappeared as a result of the treatment. Vertical bone resorption improved after periodontal regenerative therapy. Tooth mobility also improved. Masticatory disturbance in the edentulous area was improved by implant treatment.

R-03

## Treatment of Chronic Periodontitis with Apically-positioned Flap and Free gingival graft.: A Case Report

Takahiro Bizenjima*[1], Daisuke Irokawa[1], Sachiyo Tomita[1], Toshitake Takahashi[2], Atsushi Saito[1]

[1]Department of Periodontology, Tokyo Dental College, [2]Tokyo Dental Clinic

*Background:* Apically positioned flap（APF）can be used for pocket elimination and widening the zone of attached gingiva. When caries or fractures are extensive and subgingival, APF is used to expose intact tooth structure. Free gingival graft（FGG）case of generalized chronic periodontitis requiring surgical intervention including APF and FGG.
*Case:* The patient was a 53-year-old woman who presented with the chief complaint of loss of occlusion and bad breath. An initial examination revealed that 31.8% of sites with a probing depth（PD）of ≥ 4 mm. The level of plaque control as assessed by the O'Leary plaque control record（PCR）was 63.1%. Many subgingival caries were found in the cervical area of the teeth. Radiographic examination revealed horizontal bone resorption in almost region and deposition of subgingival calculus. A clinical diagnosis of generalized chronic periodontitis was made according to the classification of the American Academy of Periodontology（1999）.
*Clinical Procedures and Outcomes:* The patient underwent initial periodontal therapy. An improvement was observed in periodontal conditions at reevaluation. A series of APF were performed on #13, 14 and #21, 23, 24, 25, 27, and 43, 44, 45. FGG was performed on #34, 35. Following reevaluation, oral function was restored using a fixed bridge prosthesis and removable partial denture. Then the patient was placed on supportive periodontal therapy.
*Conclusion:* In the present case of chronic periodontitis, surgical interventions resulted in a decrease in probing depth and reestablishment of the biological width, and improved the architecture of soft tissues.

R-04

## Periodontal Plastic Surgery for Altered Passive Eruption: A Case Report

Tokihisa Mizokami*

Mizokami Dental Office

*Background:* Altered passive eruption causes not only poor esthetic but also difficulty in plaque control due to deep periodontal pocket. A feature almost always found is thick buccal bone, especially in interdental areas. The surgical technique for altered passive eruption is a periodontal plastic surgery including osteoplasty and, when necessary, osteotomy. Based on these facts, the treatment plan should include not only observing gingiva but the consideration on the positional relationship of the tooth to alveolar bone.
*Case:* Patient: 37-year old Female, First visit: 2015. 10. 21, Chief complaining: poor esthetics and caries
Diagnosis: 12, 21, 22; Altered passive eruption, 11, 21, 22; Caries
*Clinical Procedures and Outcomes:* 1.Initial preparation, 2. Re-assessment, CBCT examination, 3. Periodontal plastic surgery including osteoplasty and osteotomy, 4. Caries treatment, 5. Maintenance
Because of the usage of surgical guide with the incision line advanced to 2mm coronal to CEJ examined from CBCT data, the gingival morphology has almost not changed since immediately after the surgery. Excessive buccal marginal bone of #21 was removed and buccal interdental bone of #12, #11, #21,#22 was corrected its shape. The coronal part of the de-epithelialized interdental papillae heals by second intention.
*Conclusion:* Periodontal plastic surgery including osteoplasty and, when necessary, osteotomy for altered passive eruption contributed to enhancing the esthetic outcome and better plaque control. 2 years since the surgery, no gingival regression has observed. However, its prognosis may still need meticulous observation and maintenance care.

R-05

## A case report: Regenerative treatment for generalized severe chronic periodontitis

Takafumi Ueyama*

Ueyama Dental Clinic

*Background:* Regenerative therapy is known as an effective treatment for intrabony defect. This case report demonstrate that will enhance the success of periodontal treatment and improve the patient's periodontal problems.
*Case:* A 56 years-old female presented to our clinic with the chief complaint of tooth mobility. The patient has hypothyroidism and never smoked. Oral examination revealed that oral hygiene and bleeding score was poor（PCR 76%, BOP 88%）. Probing depth（PD）≧4mm was 64% and PD ≧7mm was 22%. Radiographic examination revealed moderate horizontal bone resorption and a few sites have severe intrabony defects.
*Clinical Procedures and outcome:* Initial periodontal treatment such as oral hygiene instructions, scaling and root plaining were performed. After two month, intrabony defect lesions were treated with regenerative therapy using Freeze-Dried Bone Allografts（FDBA）and Enamel Matrix Derivative（EMD）. Furthermore, resorbable membrane was applied to the one-wall defect lesion additionally. SPT was introduced when the recovery of the tissues was confirmed. PD and oral hygiene is stable for three years after surgery（PCR=16%, and BOP=17%, PD ≧4mm=10% ,PD ≧7mm=0%）.
*Conclusion:* The present results show the beneficial effects of regenerative therapy using FDBA, EMD, and resorbable membrane to intrabony defects. And SPT is required to maintain a good periodontal condition.

R-06

## Surgical treatment for teeth with furcation involvement: a case report

Daisuke Irokawa*[1], Takahiro Bizenjima[1], Sachiyo Tomita[1], Midori Oshima[2], Atsushi Saito[1]

[1]Department of Periodontology, Tokyo Dental College, [2]Oshima Dental Clinic

*Background:* The main purpose of treatment for furcation involvement is to facilitate maintenance and to prevent further loss of attachment. Here we report a chronic periodontitis case treated for furcation involvements.
*Case:* The patient was a 43-year-old man who requested treatment of periodontal disease. Baseline examination revealed 30.9% of sites with a probing depth of ＞4 mm and 29.2% of sites with bleeding on probing. The level of plaque control as assessed by the O'Leary plaque control record was 24.1%. Radiographic examination revealed vertical1 bone resorption in maxillary molars and lower anterior teeth. As for furcation involvements, buccal of #16 had grade 2, distal had grade 1 and mesial of #26 had grade 1. A clinical diagnosis of generalized chronic periodontitis was made.
*Clinical Procedures and Outcomes:* The patient underwent initial periodontal therapy. An improvement was observed in periodontal conditions at reevaluation. A series of flap operation were performed on #16, 17 and #25, 26 and #31, 32, 33and #41, 43. Root separation was performed on #16 of distal. Following reevaluation, oral function was restored using a fixed bridge prosthesis and removable partial denture. Then the patient was placed on supportive periodontal therapy.
*Conclusion:* In the present case of chronic periodontitis with furcation involvement, surgical interventions resulted in stable periodontal condition that facilitated favorable level of plaque control.

R-07

## Effect of orthodontic treatment for periodontitis with anterior crowding

Natsuki Okushi*

Heart Smile Dental Clinic

*Background:* Malocclusion is an exacerbating factor in periodontal disease. Toothbrushing on the lingual side tends to be ineffective, and the bone and gingiva on the labial side become thickened, without symptoms on the surface. According to Wennstrom, the gingival width and height maintain a ratio of 1: 1.5, so that a tooth with linguoversion has a high gingival line, giving rise to aesthetic dissatisfaction. I present a case of a patient with chronic periodontitis and anterior crowding in which the periodontal condition and aesthetics were improved by periodontal and orthodontic treatment.
*Case:* A 34-year-old female presented with aesthetic dissatisfaction with the maxillary anterior teeth. Periodontal examination revealed swelling and 4-mm pockets in the gingiva of the upper and lower anterior teeth and the upper right molar. Minor alveolar bone resorption and poor fit of a prosthetic appliance were observed on X-ray.
*Clinical Procedures and Outcomes:* After initial periodontal therapy and oral hygiene instruction, the anterior crowding was improved by orthodontic treatment. The occlusion was checked and a prosthetic appliance with well-fitting margins was inserted.
*Conclusion:* The orthodontic treatment made toothbrushing easier, and gingival swelling and bleeding on probing improved. Aesthetics improved as the gingiva of the lateral incisors moved labially and acquired normal thickness and height. Orthodontic treatment for chronic periodontitis with malocclusion is effective in improving periodontal disease, and can assist in ensuring prosthetic appliances are in harmony with the periodontal tissue, thus achieving a more stable prognosis.

R-08

## Periodontal regeneration therapy for moderate chronic periodontitis with horizontal bony defect: a case report

Takao Kannari*

Aozora Dental Office

*Background:* This case report describes improvement of the horizontal bony defect by periodontal regeneration therapy for a patient with moderate chronic periodontitis. Implants and removable partial denture were used for partially edentulous to achieve occlusal support.
*Case:* This 57-year-female with moderate chronic periodontitis underwent periodontal regeneration therapy after initial preparation for periodontitis. Enamel matrix derivative（EMD）, bone graft material（Bio-oss®）, and a resorbable collagen membrane（Bio-gide®）were used to the mesial of #34 with horizontal bony defect. The guided bone regeneration（GBR）was performed simultaneously for partially edentulous from #35 to #37. Implants were placed after 6 months, and free gingival graft was performed with the secondary surgery.
*Clinical Procedures and Outcomes:* After 7 years the surgery, clinical evaluation and intraoral radiograph revealed improvement of horizontal bony defect at the #34, which had undergone periodontal regeneration therapy. And free gingival graft were performed due to insufficient keratinized mucosa to ensure adequate oral hygiene, no peri-implant inflammation was observed and plaque control remains favorable.
*Conclusion:* Combination therapy with EMD and bone graft material and membrane would seem to be effective of intrabony defect with unfavorable architecture. Considering the anatomical differences between the implant and natural teeth, the keratinized mucosa around implants is necessary. Proper initial treatment and regular maintenance for the patients with periodontitis are essential prior to implantation.

R-09

## A case of multiple anterior tooth defects treated using a computer simulation-based guidance system

Chujo Kitamura*[1], Mitsuhiro Iwata[2], Koichiro Fujibayashi[3], Hiroyuki Takino[4]

[1]Kitamura Dental Clinic, [2]Sakura Dental Clinic, [3]Fujibayashi Dental Clinic, [4]Takino Dental Clinic

*Background:* With the advent of cone beam computed tomography（CBCT）and other diagnostic software, highly accurate dental implantation and surgical treatment plans are now possible in the field of prosthetic dentistry. We report our experience related to the use of a computerized guidance system.
*Case presentation:* A 56-year-old woman presented with a chief complaint of maxillary anterior fixed partial denture loss. Positions #11, #12, #23 and #37 could not be preserved due to dental root fracture. The maxillary anterior alveolar ridge exhibited deficient hard and soft tissues.
*Treatment protocol:* 1）Initial periodontal therapy, 2）Secondary assessment, 3）Implantation plan development using software, 4）Guided bone regeneration（GBR）, 5）Implantation using guidance system, 6）Secondary surgery, 7）Occlusion by temporary prosthetic device/modification of crown shape, 8）Delivery of final prosthetic device, 9）Tertiary reassessment, and 10）Maintenance.
*Treatment course:* After the initial therapy, GBR was performed using a titanium frame, a bone filling agent, and an absorbable membrane. Implantation/GBR was then performed using the computerized guidance system. Following secondary surgery by punch-out, the occlusion/crown verification was assessed using temporary prostheses. Subsequently, the final prostheses were delivered.
*Conclusion:* In this case, precise implantation of multiple anterior tooth defects was possible with the help of a treatment plan to predict final prosthetic device configuration, using a computerized guidance system. GBR using a titanium frame prior to implantation enabled three-dimensional hard tissue reconstruction, and satisfactory results were obtained in terms of both functionality and aesthetics.

R-10

## Treatment of gingival fenestration with connective tissue graft

Daichi Kita*[1], Takashi Kinumatsu[2], Yoshihito Ishii[3], Shigeko Yamamoto[4], Atsushi Saito[1, 5]

[1]Department of Periodontology, Tokyo Dental College, [2]Kinumatsu Dental Clinic, [3]Mizuno Dental Clinic, [4]Yamamoto Dental Clinic, [5]Oral Health Science Center, Tokyo Dental College

*Background:* Gingival fenestration is defined as the exposure of tooth surface due to loss of the overlying bone and gingiva. Although information on gingival fenestration is limited, techniques for its treatment such as a connective tissue graft（CTG）, various flap designs, guided tissue regeneration, and orthodontic treatment have been reported. We report localized chronic periodontitis with gingival fenestration requiring surgical treatment with a CTG.
*Case:* The patient was a 32-year-old man presenting with the chief complaint of esthetic impairment and gingival twitching due to gingival fenestration. At baseline, periodontal examination revealed 3% of sites with a probing depth of ≥ 4 mm and 8.9% with bleeding on probing. Radiographic examination revealed vertical bone loss in #15 and 36, together with buccal fenestration in the lower right central incisor（#41）. Baseline examination revealed localized periodontal breakdown, including gingival fenestration in #41.
*Clinical Procedures and Outcomes:* Initial periodontal therapy comprised plaque control and scaling and root planing. Following suppression of inflammation, occlusal adjustment was performed in the anterior teeth. As periodontal plastic surgery, combined use of an elevated flap and a CTG was applied at #41. Following reevaluation, the patient was placed on maintenance care.
*Conclusion:* In the present case of localized chronic periodontitis with gingival fenestration, initial periodontal therapy and surgical treatment with a CTG resulted in the improvement of the patient's periodontal condition. It has remained stable over a 1.5-year period.

R-11

## A case of regenerative therapy for a true combined endodontic-periodontal lesion

Kazuaki Shinoda*

Ohana Dental Clinic

*Background and objective:* Dental pulp and periodontal tissue often determine the fate of teeth, and a disease involving both can pose a significant challenge. Such combined endodontic-periodontal lesions involve difficult decision-making regarding treatment and operative methods.
*Materials and Methods:* A 51-year-old male presented with a chief complaint of lower-left swelling and pain. There was a periodontal pocket（11 mm）reaching the distal root apex of the lower-left first molar with bleeding and pus discharge. Regenerative therapy was conducted using enamel matrix derivative, xenograft（Bio-oss®）, and cytoplast membrane after the endodontic treatment.
*Results:* Two months after the endodontic treatment, regenerative periodontal therapy was using a microscope. Follow-up examinations continued for ten months postoperatively. The final prosthesis was inserted after healing periodontal tissue
*Conclusion:* With appropriate materials, diagnostic techniques, and operative methods, regenerative therapy may improve outcomes for endodontic-periodontial lesions that were previously difficult to treat.

R-12

## Regenerative Surgery of severe periodontal defects Using fibroblast growth factor（FGF-2）. case report

Taishi Hirose*, Makiko Hirose, Takahiro Shiomi, Tetsushi Hirose

Yokohama Kannai perio-Implant Institute（Yuraku Dental Office）

*Background and objective:* We report regenerative procedure for severe periodontal defects with fibroblast growth factor（FGF-2）
*Materials and Methods:* A 50-years old female complained of severe pain of gum on the posterior tooth. The patient is in excellent health and reports no history of any systemic disease or any problem, which would preclude dental treatment. Tooth #1, #16, #17, #32 are missing due to the treatment from in the past.
Upon perio-examination of the patient, we formulated a diagnosis of generalized severe periodontitis. Pocket depth ranging from 3 to 12 mm with some gingival recession. Radiographically, we observed severe horizontal and vertical bone loss around posterior tooth. After obtaining informed consent and phase 1 therapy, we began a regimen of regenerative treatment with fibroblast growth factor. Prior to the regenerative treatment, subtraction X-ray were taken and mobile tooth were splinted.
*Results:* Pocket depth ranging from 2 to 4 mm with a few gingival recession. Some clinical attachment gain found most of defective sites.
*Conclusion:* Regenerative therapy with FGF-2 might be an effective treatment for severe periodontal defects.

R-13

## A case of regenerative therapy supported by occlusal stabilization

Takeshi Ajioka*

Ajioka Dental Clinic

*Background:* It is essential to establish a vertical stop at the molars for occlusal stabilization of the whole jaw.
Here we report a case of attempted occlusal stabilization in a patient with molar furcation involvement. The periodontal tissue of the molars was stabilized through regenerative therapy using Emdogain.
*Case:* The patient was a 66-year-old non-smoking female. She was first examined in July 2015, presenting with pain in the front teeth. Class 2 furcation involvement was present in 26 and 46.
The patient was diagnosed with chronic periodontitis.
*Treatment and result:* Initially, non-surgical therapy was conducted. At the same time, a poorly fitting prosthesis was removed, and a provisional restoration was placed. Regenerative therapy was conducted using Emdogain on 26 and 46 with residual furcation involvement. Follow-up examination was conducted after placement of the provisional restoration. We proceeded to place the final restoration as no issues were observed during follow-up. Approximately one year postoperatively, the patient's progress was favorable.
*Conclusion:* For patients with chronic periodontal disease, occlusal stabilization in the jaw with regenerative therapy is effective for long-term intraoral stabilization.

R-14

## Periodontal regeneration using Fibroblast Growth Factor-2 with Minimally Invasive Surgical Technique

Kotaro Tsuchida*

Tsuchida Dental Office

*Background and objective:* The procedures using Enamel matrix derivative with Minimally Invasive Surgical Technique（MIST）for deep intrabony defect have shown good results. These case series were evaluated using Fibroblast Growth Factor-2（FGF-2）with MIST.
*Materials and Methods:* 10 intrabony defects in 5 periodontal patients were treated. The sites had a probing depth（PD）≧ 8mm and intrabony component depth ≧5mm. MIST approach was utilized for surgical access. Following surgical debridement using microscope, FGF-2（Regroth®）was placed into the intrabony defect. After 6 months, the surgical sites were reevaluated. The effect of therapy was evaluated by assessing Probing Depths（PD）, Clinical Attachment Level（CAL）, and radiographic bone fill.
*Results:* The mean PD reduction amounted to 6.3mm. Mean gain CAL was 5.3mm. Radiographic bone fill was observed after 6 months, suggesting possible periodontal regeneration.
*Conclusion:* Short-term results from these case series indicate the use of FGF-2 with MIST resulted in a favorable outcome in the treatment of deep intrabony defect.

R-15

## A case of severe periodontitis utilizing interdisciplinary approach

Keiichiro Takahashi*

Takahashi Dental Clinic

*Objective:* In modern periodontal therapy, a variety of therapeutic options must be skillfully combined to achieve a stable long-term prognosis. We report a case in which we achieved an excellent long-term effect through a strategic combination of implant, orthodontic, and periodontal regenerative therapies.
*Case report:* The patient was a 45-year-old woman who required full-mouth periodontal and occlusal therapies. Occlusal reconstruction was achieved with orthodontic therapy in conjunction with implant therapy. Improvement in the anterior dental region was achieved through whole-mouth periodontal-prosthetic therapy, using periodontal regenerative techniques. We also report her condition 5 years after treatment.
*Therapy and results:* After the initial whole-mouth periodontal therapy was performed, the patient underwent orthodontic therapy using a temporary anchorage device. In the molar region, an implant was inserted after performing a sinus lift with extensive guided bone regeneration. Provisional restoration in the anterior region was achieved with periodontal regenerative therapy using Emdogain, which stabilized the periodontal tissue and bite. Eventually, the patient transitioned to a periodontal prosthesis.
*Conclusion:* We used a strategic combination of orthodontic, implant, periodontal regenerative, and prosthetic therapies in a case of periodontitis and occlusal collapse, accompanied by severe bone defects. This technique achieved improvement and a positive prognosis. The effectiveness of a interdisciplinary approach is evidenced by the stability of the patient's condition post-treatment even after 5 years.

R-16

## Periodontal regenerative therapy and orthodontic treatment for generalized aggressive periodontitis: a case report

Takayuki Kurihara*

Kinen Dental Clinic

*Background:* Aggressive periodontitis occurs in people from their teens to their 30s. In Japan, one out of every 1,000 to 2,000 people suffers from this disease. Although generally healthy except for their periodontitis, rapid periodontal tissue destruction can be observed. In most cases, once the patient perceives the symptoms, the periodontal disease has already significantly progressed.
*Case:* Initial visit: May, 2013 Age: 32 Sex: Male Chief complaint: Right lower jaw hurts due to swelling.
Past medical history: N/A Family medical history: Aggressive periodontitis was suspected in his grandmother
Smoking history: 20 cigarettes per day for 14 years
*Clinical Procedures and Outcomes: Initial periodontal treatment from June, 2013 to December, 2014*
*The patient suffered from low motivation, and his oral condition necessitated surgical periodontal therapy. However, as a smoker, the prognosis was not likely to be positive, even with surgery. While conducting the initial periodontal therapy, efforts were made to motivate the patient. After 18 months, he reduced his cigarette consumption to 5 cigarettes per day. After the patient's oral condition and gum inflammation improved, and after he requested, we transitioned to surgical periodontal therapy.*
*Conclusion:* Currently, orthodontic treatment and prosthodontic treatment have finished, but some periodontal pockets still remain. Because smoking is related to the impaired tissue healing, it is suggested that we should continue supportive periodontal therapy with caution.

R-17

## A case of autotransplantation 30 years after transplantation

Hiroyuki Kimura*[1], Taro Eida[1], Tsuyoshi Kumano[1], Makoto Yokota[2]

[1]Kimura Dental Clinic, [2]Yokota Jyuku

*Background:* Recently, autogenous tooth transplantation has received renewed recognition as a predictable procedure for restoring tooth defects. Here, we report a case of autogenous tooth transplantation that was successful for 30 years.
*Case:* The patient was a 24-year-old female. On March 29, 1989, she presented for treatment of decay in 37. She had no remarkable medical history and was a non-smoker. Intraoral findings revealed that 37 showed residual root conditions and 38 was semi-impacted. X-ray examination revealed horizontal contact between 38 and 37.
*Clinical Procedures and Outcomes:* Tooth 37 was diagnosed as periodontally hopeless. We obtained informed consent to perform transplantation. After extracting 37, we successfully transplanted 38 into the 37 site. We then performed root canal treatment on July 25. After confirming stability of the periodontal tissue, we placed a prosthesis on October 16. Post-transplantation went well and the patient had no complaints. After transplantation, bone condition was good; no abnormalities were observed in X-ray findings. The patient was shifted to maintenance. Even after treatment for 30 years, probing pocket depth on transplanted teeth was 3 mm, with no discomfort. Periotest values were 0-5. 3DCT indicated no root resorption or other abnormal findings.
*Conclusion:* In this case, autologous tooth transplantation was performed after tooth extraction; it has been clinically successful for 30 years. This case showed high predictability, even in molar teeth that experienced strong biting force. As saving the periodontium helps to maintain natural tooth function, this case suggested that we should consider transplantation before extraction.

R-18

## Use of orthodontic and periodontal therapy to a moderate periodontitis

Shinichi Saika*

Saika Dental Clinic

*Background and objective:* Patients suffered by moderate and/or more progressed periodontitis often have pathological tooth migration. For those cases, Orthodontic therapy can contribute to allocate occlusal forces uniformly by recovering the dental arch continuity and correcting teeth axes so that it enables to recover the masticatory function, stabilize the dentition and to improve the oral cleansability.
*Case:* Patient: 61-year old female First visit: June 20th. 2015
Chief complaint: anxious for anterior tooth to fall out
Medical history: Nothing Particular
Diagnosis: Generalized moderate chronic periodontitis
*Clinical Procedures and Outcomes:* 1）periodontal initial therapy 2）re-evaluation 3）periodontal surgery（tissue attachment therapy） 4）re-evaluation 5）orthodontic therapy 6）re-evaluation 7）maintenance. Periodontal surgery of tissue attachment therapy was performed for the site with deeper periodontal pocket than 4mm after confirming BOP positive and improvement of PCR following periodontal initial therapy. Orthodontic therapy improved malalignment and corrected teeth axes. Re-evaluation revealed the satisfactory result of improvement in periodontal tissue and esthetic recovery.
*Conclusion:* Tooth malalignment causes poor plaque control and leads to the inflammation. In addition, inclined teeth axes, mal-positioned teeth with abnormal lateral forces and traumatic occlusion are harmful to periodontal tissue.
That may increase the risk of periodontal disease and difficulty in treatment.
It suggests that including Orthodontic therapy into periodontal treatment contributes to 1）eliminate the damage to periodontal tissue, 2）accelerate remodeling of periodontal tissue, and the prognosis with long-term stability by decreasing the difficulty and increasing the efficacy of the treatment.

R-19

## Regenerative therapy and dental implants for severe periodontitis: a case report

Hiroshi Okuda*

Okuda Dental Clinic perio-implant center

*Background:* In order to maintain long-term stability following treatment for a vertical bone defect or furcation involvement, it is important to minimize the depth of periodontal pockets and to regulate the bone and gingival levels of natural and restored teeth. This will create an environment that is easy to maintain.
*Case:* Patient: 58-year-old woman
Initial examination: April 2009
Chief complaint: Discomfort in the lower left tooth region
Systemic disease: Nothing of note
*Treatment:* Regenerative periodontal therapy was conducted to improve residual periodontal pockets following initial periodontal treatment. Regarding Class II furcation involvement on #17 and #27, although postoperative assessment confirmed bone regeneration, definitive surgery was performed to improve the residual periodontal pockets. The mandibular molars were extracted and guided bone regeneration was performed.
*Results:* For the maxillary molars, sufficient keratinized gingiva and a shallow gingival sulcus were obtained. As for the mandibular molar region, the bone level was successfully matched to the height of the adjacent teeth using guided bone regeneration.
The treatment was successful in matching the bone and gingival levels between natural teeth and implants as well as in creating an environment that is easy to maintain.
*Conclusion:* The patient has good plaque control and has been coming in every 3 months for maintenance. 5 years post-operation, favorable results have been maintained. Maintenance will be continued in the future.

R-20

## Five-year follow up of periodontal-guided tissue regeneration for vertical bone loss

Seiji Ozaki*

Ozaki Dental Clinic

*Background and objective:* Initial examination: 53-years old male（Februarry 2013）
Chief complaints: swelling, pain, and mobility（#41）
Symptom: Overall redness and swelling of gums
Findings: Bop（+）,P.D: 3mm or less54.8％,4~5mm22.1％,6mm or more23.1％ , vertical bone loss（#36 and 45）and premature contact（#27 and36）
Diagnosis: Generalized moderate chronic periodontitis. Occlusal trauma
Therapeutic policy: 1）initial preparation, 2）re-evaluation, 3）periodontal surgery, 4）re-evaluation, 5）oral function recovery, and 6）SPT
Therapeutic goal: 1）periodontal tissue stabilization, 2）establishment of bilateral molar occlusal support, and 3）anterior guidance
*Clinical Procedure:* 1）initial preparation, 2）re-evaluation, 3）periodontal-guided tissue regeneration（#36 and 45）, 4）implant treatment（#46）, 5）oral function recovery, 6）re-evaluation, and 7）SPT
［Results］Periodontal tissues became stable（#41）and the alveolar bone condition at the sites where periodontal-guided tissue regeneration was performed for vertical bone loss（#36 and 45）was stable.
*Conclusion:* Based on the initial preparation, we improved the periodontal tissues, eliminated premature contact, and established bilateral molar occlusal support. This resulted in anterior guidance and allowed for the stabilization of all periodontal tissues. Cleaning, occlusal examination/adjustment, and maintenance of the patient's motivation toward homecare during SPT every four months were the most important factors in this case.

R-21

## Ridge augmentation using modified onlay-interpositional graft: a case report

Shu Hoshi*[1, 2], Tatsuya Akizuki[2], Takanori Matsuura[2], Yuichi Izumi[2]

[1]Hoshi Dental Office
[2]Department of Periodontology, Graduate School of Medical and Dental Science, Tokyo Medical and Dental University

*Background:* Severe periodontitis leads to an extreme reduction of alveolar ridge volume, causing an esthetic and functional disorders. To resolve these problems, ridge augmentation using modified onlay-interpositional graft was performed and good clinical results are achieved.
*Case:* The patient was a 62-year-old man who suffered from severe chronic periodontitis. Alveolar bone resorption reached the apical part of the right upper central incisor and lateral incisor. The patient had no systematic disease and no history of smoking.
*Treatment and Results:* Soft tissue ridge augmentation was planned to reconstruct the reduced alveolar ridge because the patient did not want implant treatment. Three months after tooth extraction of the right upper central incisor and lateral incisor, soft tissue ridge augmentation using modified onlay-interpositional graft was performed. According to the method of the original onlay-interpositional graft procedure reported by Seibert and Louis, the recipient site epithelium was removed on the labial side of the ridge to prepare the recipient bed for the graft segment. The method was modified to preserve the recipient site epithelium to keep the keratinized gingiva width and soft tissue volume. To increase the volume of soft tissue, additional connective tissue was harvested from the distal side of the right upper first molar mucosa and grafted into the recipient site. The anterior area was treated with a tooth supported fixed prosthesis. The patient was followed up for 4 years, and the volume of reconstructed ridge was maintained.
*Conclusion:* The modified onlay-interpositional graft is an effective procedure to reconstruct the reduced alveolar ridge. Moreover, the volume of the reconstructed soft tissue could be maintained for a long period of time.

R-22

## Gingival Cul-de-sac: Complications Consequential to Root Coverage Procedures using a Subepithelial Connective Tissue Graft

Makoto Ono*

Shijo-Karasuma Perio Implant Center

*Background:* There have been reports of post-operative complications following root coverage treatment for gingival recession（GR）using a subepithelial connective tissue graft（SCTG）. This case report chronicles the formation of a gingival cul-de-sac caused by the creation of a pocket between the flap and graft, a complication which followed the SCTG-based procedure.
*CASE:* The patient was male, a smoker, forty-eight years old, and complained of poor esthetics. There were multiple teeth with GR in the upper anterior area.
*Clinical Procedures and Outcomes:* The GR was treated with a coronally advanced flap using a SCTG and Enamel Matrix Derivative. The graft was fully covered by the flap after surgery. However, 2 weeks after the operation, a white, creamy discharge was observed, indicating that a gingival cul-de-sac had formed. Twenty months after the operation, the separation of the flap and the graft had become obvious, and the discharge was still observed. To excise the cul-de-sac, the outer layer of gingiva was carefully removed. The grafted tissue remained on the denudated root surface and it matured to a conspicuous, unaesthetic appearance.
*Conclusion:* In cases where SCTG is used in root coverage, a gingival cul-de-sac may occur, even if the graft is fully covered by the flap. In this case, the cul-de-sac failed to heal naturally over the 20-month period of observation. By removing the outer layer of the cul-de-sac, the tissue of the graft remained, covering the root surface.

R-23

## Root coverage for multiple recession of natural teeth

Ryuichi Shirane*

Shirane Dental Clinic

*Background and objective:* Gingival recession is a common periodontal problem which clinicians are confronted with. Although root coverage is one of the most valid procedures for gingival recession, the clinical outcome is crucially influenced by technical and site-specific factors. On this poster, I would like to present you my clinical case of root coverage for deep and multiple gingival recessions.
*Materials and Methods:* The patient was 68 years old male. He noticed the change of his gingival margin with age.
Gingival recessions on maxilla and mandibular canine area became advanced near mucogingival junction. There were slight malalignment on maxilla left anterior teeth, but no attachment loss around gingival recession. Gingival bio-type of this patient was thick-flat.
After initial treatment, root coverage procedures using coronally advanced flap（CAF）with subepithelium connective tissue graft（SCTG）were applied three times to #33,34 in August 2016, #23,24 in January 2017 and #12,13 in September 2017. Tunnel procedure was applied under a surgical microscope.
*Results:* After three surgeries, the patient did not have any complaint, and SCTG did not undergo necrosis.
90％ of root coverage was obtained using Langer& Langer technique for #33,34.
Tunnel technique for #23, 13 achieved complete coverage, while the same procedure for #23,24 leaded to approximately 60 ％ root coverage because of teeth malposition.
*Conclusion:* Root coverage for gingival recession is quite beneficial method from aesthetic and functional point of view. Meanwhile, the selection of the procedure requires rigorous examination of each case. Therefore, it is essential to consider accumulated previous evidence and apply careful procedure in order to achieve successful result.

R-24

## A case report of class II furcation involvement in the mandibular molar with generalized advanced chronic periodontitis utilized periodontal regenerative therapy

Misa Satomi*[1], Masahiko Nikaido[2]

[1]Pacific Century Place Marunouchi Dental Clinic, Satomi Dental Clinic, [2]Nikaido Dental Office

*Background:* We report a case of a patient with class II furcation involvement of #36 utilized periodontal regenerative therapy; growth factor（platelet-derived growth factor: rh-PDGF-BB; GEM21s®）, mineralized freeze-dried bone allografts（FDBA）, and absorbable collagen membrane（Bio-Gide®）. Before the treatment, informed consent was obtained from the patient after explaining the use of unapproved drugs and materials.
*Case:* A 57-year-old man had the chief complaints of mobility and pain on mastication with #45. His medical history revealed unremarkable, but he had smoked 20 cigarettes a day for 35 years. Poor plaque control（PC）, gingival enlargement and redness, supra and subgingival calculus were observed throughout the whole mouth with plaque control record（PCR）of 100％, bleeding on probing（BOP）of 100％, and average probing pocket depth（PPD）of 4.2 mm. Radiographical finding revealed that vertical bone resorption and radiolucency in the furcation of the molars were observed for many teeth. The mesial PPD and lingual midsection PPD of #36 were 6 and 5 mm, respectively, #36 had no mobility and class II furcation involvement on the lingual side.
*Treatment*: 1. Initial preparation（oral hygiene instruction（OHI）, scaling/root planing（Sc/Rp）, smoke cessation training, extraction）
2. Re-evaluation
3. Implant placement（#45-46）
4. Surgical periodontal treatment（#15, #24: rh-PDGF-BB+FDBA, #35-36: rh-PDGF-BB+FDBA+Bio-Gide®）
5. Re-evaluation
6. Prosthodontics
7. SPT.
*Conclusions:* #36 with class Ⅱ furcation involvement was closed by rh-PDGF-BB, FDBA and Bio-Gide®. #36 was stable at the post-SPT 6-month follow-up, we will continue to provide SPT to maintain his dentition and implants.

R-25

## Dispersion of occlusal support area in periodontal therapy: A Case Report

Nobuhiko Inagaki*

Midorigaoka Dental Clinic

*Background and objective:* Excessive occlusal force has been known as the factor enhancing the degree of periodontal destruction of periodontally compromised tooth. Occlusal adjustment as a part of periodontal therapy has been controversial for years. The purpose of this case report is to demonstrate the effects of occlusal adjustment, associated with periodontal therapy, on periodontal parameters.
*Materials and Methods:* A 60−year−old female patient presented with discomfort of #24 and 46. #17 has been extracted 7−8 years ago due to poor prognosis. Class III furcation involvement, mobility class II as well as vertical bone loss were observed on multiple teeth. Patient was diagnosed as localized chronic moderate periodontitis.
*Results:* Following initial therapy, #26 was removed due to poor prognosis, but #27 was preserved and well functioned. The periodontal regeneration, using Enamel Matrix Derivative（EMD）has been applied to vertical bone loss on the mesial of #36 as well as the root separation was performed for the furcation involvement of #46. #48 was transplanted to maxillary right partial edentulous area and #35 and 45 were moved orthodontically for occlusal force distribution. A significant improvement of periodontal condition was achieved after therapy.
*Conclusion:* This case demonstrated the significance of periodontal treatment in conjunction with occlusal adjustment to reduce the progression of periodontal attachment loss. A success to key to treat chronic periodontitis involved occlusal trauma, especially for patients with multiple missing teeth was to adjust occlusion to increase the number of tooth supporting occlusion following inflammation control.

R-26

## Periodontal treatment in localized severe chronic periodontitis: A Case Report

Dai Kawanabe*

Kawanabe Dental Clinic

*Background and objective:* The reaction of periodontal tissue to treatment is strongly impacted by the age of the patient and other modifying factors. In the present case, we actively treated a patient who presented with conditions that appeared advantageous for the performance of periodontal treatment. A significant improvement in the patient's condition was achieved.
*Materials and Methods:* The patient was a 41−year old woman who complained of discomfort in #13 and #32. Class II furcation involvement in #17 and #27, and vertical bone loss in #12, #13, #24, and #32 were confirmed through radiographic examination. There was +2 mobility in #32, and +1 mobility in #13, #14, #17, #23, and #26. Based on the examination, the patient was diagnosed with localized severe chronic periodontitis.
*Results:* Reevaluation following the initial treatment led to the extraction of #48.
We performed periodontal tissue regenerative therapy using enamel matrix derivative（EMD）on #12, #13, and #32, which presented with vertical bone loss. We performed trisection of the furcation involvement in #17 and #27. After treatment, a significant improvement in the periodontal condition was observed.
*Conclusion:* This case verifies the importance of periodontal treatment. Factors that indicate a good response to periodontal treatment and surgery include young age of the patient, a high level of plaque control, and absence of risk factors such as smoking.
It is important to carefully examine treatment selection, such as periodontal surgery, considering the patient's background in addition to reevaluation.

R-27

## Effective Management of Periodontal and Peri-implant problems With Er: YAG Laser

Taichen Lin*[1,2,3], Cheng-Chia Yu[2,4], Yoichi Taniguchi[1,5], Paul Pao-Ying Lin[6], Chen-Ying Wang[7], Akira Aoki[1], Chun-Cheng Chen[2,3]

[1]Department of Periodontology, Graduate School of Medical and Dental Sciences, Tokyo Medical and Dental University（TMDU）, Tokyo, Japan, [2]School of Dentistry, Chung Shan Medical University, Taichung, Taiwan, [3]Department of Dentistry, Chung Shan Medical University Hospital, Taichung, Taiwan, [4]Institute of Oral Science, Chung Shan Medical University, Taichung, Taiwan, [5]Taniguchi Dental Clinic, Sapporo, Japan, [6]Professors Dental Clinic, Taipei, Taiwan, [7]Department of Periodontology, National Taiwan University Hospital, Taipei, Taiwan

*Background and Objective:* The Er: YAG laser（ErL: 2,940 nm）is very useful for dental treatment such as soft and hard tissues management as well as dental implant surface debridement. Three patients with different clinical problems were included in this report, and ErL was applied under various conditions, including flapless esthetic crown lengthening, implant placement with simultaneous guided bone regeneration, and socket preservation accompanied with implant surface debridement.
*Materials and Methods:* The first patient had an unhormonious gingival contour in the anterior maxillary zone, and ErL was used to perform gingivectomy as well as flapless bone contouring. The second patient had missing teeth at #36 and #37 sites with severe bony defects. After bone grafting around the embedded two implants, non-contact ErL irradiation was performed to induce blood coagulation on the surface and maintain the bone graft. The third patient had a severe bony defect at a missing #26 tooth site and peri-implantitis on the mesial side of #27 implant, and the bony defect and implant surface were carefully debrided by ErL. Then, the defect was filled with bone graft and covered with a resorbable membrane for socket preservation.
*Results:* Following ErL therapy, the first patient obtained a harmonious and esthetic anterior cervical line of teeth, and the other two patients exhibited favorable bone regeneration in the treated bone defects.
*Conclusion:* Results of the current report suggested that ErL therapy is beneficial and effective in periodontal and peri-implantal managemant, leading to a favorable wound healing as well as successful outcomes.

R-28

## Treatment in collaboration with otolaryngologist to elevate sinus floor and implant installation

Akihiro Ishikawa*

Den-en-chofu Perio-Implant Center, Ishikawa Dental Clinic

*Background:* A collaboration treatment with an otolaryngologist may be necessary when installing maxillary molar implants for a patient who has acute or chronic inflammation in her maxillary sinus. In this case, the patient who had developed dental maxillary sinusitis was treated by an otolaryngologist who performed Endoscopic Sinus Surgery（ESS）on the natural ostium（antrostomy）.
*Case:* The patient was a 56-year-old female. She presented with # 26 pain and left cheek swelling. Opacity was found in her maxillary sinus and ethmoid sinus in a CT scan, and the natural ostium was closed. In addition, a paradoxically bent middle turbinate was recognized. Considering the need for an otolaryngologist, the patient was sent to a specialist. It was determined that the patient needed an antrostomy with ESS.
*Clinical Procedures and Outcomes:* Septoplasty was performed by ESS under general anesthesia. The natural ostium on the left side was enlarged. Subsequently, # 26 tooth was pulled out. The maxillary sinus became a normal mucosal. Since # 26 had an insufficient vertical bone, a sinus floor elevation was performed. The implant was installed after confirming that there were no problems in the patient's CT scan 6 months later.
*Conclusion:* When attempting to install implants in the maxillary posterior molar region, maxillary sinusitis may be present. With the closure of the natural ostium, dental treatment alone will not result in a cure. Therefore, it is possible to carry out implant treatment safely after restoring the maxillary sinus to a normal state through collaboration with an otolaryngologist.

R-29

## Effect of non-surgical periodontal therapy assisted by periodontal endoscope. Report of cases

Masahiko Nikaido*

Nikaido Dental Office

*Background and objective:* Although there have been numerous articles have been published regarding periodontal non-surgical therapy, how quality of the therapy impacts outcome of treatment has not been well documented and discussed. Recently, periodontal endoscope has been introduced and it allows us to meticulously debride a root surface with direct visualization. We report of cases utilized this new device.
*Case:* Case 1: A 30-year-old female patient who were diagnosed generalized aggressive periodontitis. Case 2: A 48-year-old female patient, who were diagnosed generalized advanced chronic periodontitis.
*Clinical Procedures and Outcomes:* After conventional scaling/ root planing, residual periodontal pockets in anterior region were non-surgically re-treated assisted by periodontal endoscope（Perioscolpy™）. We usually spent 20 to 30 minutes for each tooth. Re-evaluations were conducted 9 months after initial scaling root planning and 3 months after re-scaling and root planning assisted by periodontal endoscope. Periodontal probing depths were significantly reduced from 10mm to 4mm in case 1 and 12mm to 5mm in case 2. Both periodontal pocket did not show BOP（bleeding on probing）. In radiographic examination, bone regeneration was observed.
*Conclusion:* We have confirmed significant improvement of patients' periodontal conditions after meticulous re-scaling and root planing assisted by periodontal endoscope In the coming super aged society, efficacy of non-surgical periodontal therapy has to be re-examined and re-evaluated.

R-30

## Teeth auto-transplantation utilizing enamel matrix derivative and bone graft. A case report

Mitsuharu Inoko*

Inoko Dental Clinic

*Background and objective:* Teeth auto-transplantation is one of the treatment options to replace missing teeth. However, it has been considered that enough amount of bone at the recipient site is prerequisite and it limits the application of this therapy. In this case report, by utilized enamel matrix derivative（EMD）and bone graft, we were able to achieve teeth auto-transplantation for a case which did not have sufficient bone at the recipient site and maintain for a long term.
*Materials and Methods:* #11, 21 were extracted due to root fractures and labial plate was already missing. #12 and #22 were auto-transplanted into the extracted site and reconstruction with EMD, bone graft and collagen membrane were performed simultaneously.
*Results:* We have been observed this case for 14 years and auto-transplanted teeth are still functioning well without major complications such as ankylosis. In a recent radiographic examination, periodontal ligament and alveolar bone around two teeth were observed, suggested long term stable condition of this case.
*Conclusion:*
Generally, sufficient bone in the recipient site is prerequisite for a teeth auto-transplantation. However, by utilizing biologics and bone grafts, this therapy could be applied for a case with insufficient bone at the recipient site.

R-31

## Occlusal reconstruction for a patient with advanced periodontal disease. A case report

Takeshi Kamiyama*

Kamiyama Dental Clinic

*Background and objective:* In a treatment of advanced periodontal disease, occlusal discrepancies are common problem, which is associated with the progression of this disease and oral reconstruction is often required. We report a case which we accomplished comprehensive periodontal therapy and occlusal reconstruction for a patient with chronic advanced periodontitis.
*Materials and Methods:* A 64-year-old female patient presented with chief complaints of pain on mastication and esthetic problem of upper anteriors. Periodontal examination revealed deep intrabony pockets in molars and multiple hyper teeth mobility. Pathologic teeth migration was noted for upper anteriors. The patient also showed loss of vertical dimension due to severe teeth wear.
*Results:* After initial preparation, metal reinforced provisional crowns were utilized to increase vertical dimension and to achieve occlusal stability. Then periodontal regenerative therapies with enamel matrix derivative and bone graft were applied to eliminate deep intrabony defects. Upper anteriors were corrected with localized orthodontic therapy（LOT）and final protheses were placed. After active therapy, the patient has been placed in periodontal supportive therapy for 5 years without major complications.
*Conclusion:* In the treatment of this patient, metal reinforced provisional crowns contributed not only to maintain her occlusal dimension, but also to achieve optimal periodontal regeneration during an active therapy. We also succeeded to give the patient anterior guidance and canine disclusion by LOT, followed by full mouth reconstruction. Together with esthetic rehabilitation, whole treatment contributes to improve her quality of life. We continue to pay careful attention to obtain long term stability.

# Dental Hygiene

H-01

## A Case Report: Importance of Initial Therapy for The Chronic Periodontitis

Masami Sato*, Kazuyo Ikeda, Masahiko Ikeda

Ikeda Dental Clinic

*Background:* Normally, surgical treatment would be chosen as an option for the treatment of progressive chronic periodontitis. However, we experienced an accepted outcome from the treatment, which only chosen initial therapy expecting the individual's healing power on progressive periodontitis, had good time course of 15 years from the first visit, and, thus, report here.
*Case:* The patient was 40−years female with chronic periodontitis especially in the region of #45. Swelling, 5−9 mm probing depth, 1.5 of tooth mobility, vertical bone absorption from dental x−ray, were recognized at the first examination.
*Clinical Procedures and Outcome:* We hired the counseling technique tried gaining the motivation for the treatment from the patient. The physical treatment was done with instruction of teeth brush and occlusal adjustment clarifying the concept of healthiness oral hygiene. Only toothbrush was used for the brushing avoiding hurting the gingiva. We carefully performed scaling and root planing with the use of hand scaler. At the timing of 8 months later from the initial therapy, 2−4 mm probing depth and a reduction of vertical bone absorption from dental x−ray, were recognized. The tooth was subjected the orthodontic and prosthodontic treatments after the root canal treatment. The patient moved into the recall course and kept the periodontal condition good for 12 years.
*Conclusion:* In this case, we observed a significant healing with the treatment, which contains non−invasive initial therapy to the gingiva and dental root. It is suggestive that this method of initial therapy would be an effective option to chronic periodontitis.

H-02

## Microbiological testing for patients in supportive periodontal therapy

Michiko Ogata*, Shimizu Hiroyasu

Shimizu Dental Clinic

*Background and objective:* The primary etiology of the periodontal disease is bacterial plaque. The periodontal pathogenic bacteria greatly contribute to the progression of periodontal disease. This study reports the progression of periodontal disease and the change of periodontal pathogenic bacteria of patients in supportive periodontal therapy utilizing the microbiological testing.
*Materials and Methods:* Case#1 is a 50 years old, Asian male. The patient was diagnosed as localized severe chronic periodontitis. Non−surgical therapy was performed, followed by orthodontic treatment for lower anterior teeth. Finally, every 3−month supportive periodontal therapy was regularly done. Case#2 is a 53 years old, Asian female. The patient was diagnosed as localized severe chronic periodontitis. Non−surgical therapy was performed. After that, regenerative therapies were applied to three sites where deep periodontal pockets remained. Finally, every 3−month supportive periodontal therapy was regularly done. In these two cases, the microbiological testing was performed at the first examination and at the supportive periodontal therapy.
*Results:* In these two cases, as a result of OHI, definitive treatment with the regenerative therapy and orthodontic treatment, the periodontal condition was significantly improved. The red complex（*Pg, Tf, Td*）of periodontal pathogenic bacteria were observed at the first examination, but it decreased to the tolerance level at the supportive periodontal therapy. *P. intermedia* of the orange complex was however increased.
*Conclusion:* As a result of definitive treatment with the regenerative therapy and orthodontic treatment, the red complex decreased to the tolerance level because the periodontal attachment was gained and periodontal pocket became shallower. But *P. intermedia* was increased in shallower periodontal pocket. The microbiological testing may help to make proper supportive periodontal therapy planning and motivate a patient by comparing at the first examination and at the supportive periodontal therapy, because it can exhibit activity of periodontal disease.

H-03

## Orthodontic treatment of periodontal patients

Yuka Ogawa*, Kotaro Tsuchida

Tsuchida dental office

*Background and objective:* The pathologic tooth migration from periodontal disease may become the chief complaint of the patients, from aesthetic point of view. The purpose of this study was to investigate the changes in periodontal health after orthodontic treatment, especially Bleeding On Probing（BOP）, Probing Pocket Depth（PPD）, and the radiographic analysis.
*Materials and Methods:* This study was conducted with 15 patients（43 to 75 years old）diagnosed with pathologic tooth migration. After the periodontal treatment people received orthodontic treatment. All patients were investigated the changes of BOP, PPD from initial visit to supportive periodontal therapy.
*Results:* Compared with the stage of basic periodontal treatment, BOP and PPD got better on the Supportive Periodontal Therapy（SPT）following orthodontics.
*Conclusion:* For the patient who is affected by the pathologic tooth migration, the basic periodontal treatment is indispensable to orthodontic treatment. In addition to the careful removal of infection, we dental hygienist should have the patients recognize the importance of self-care during the orthodontic treatment and Supportive Periodontal Therapy（SPT）.

H-04

## 10-year favorable non-surgical prognosis of Generalized Severe chronic periodontitis

Yuko Shimoda*, Ryousuke Kondo, Rumi Fukamizu, Tetsuya Mizukami

Mizukami Dental Clinic

*Background and objective:* Short-term, efficient, less-invasive and non-surgical approach for the patients with progressed periodontal disease is one of the challenges in periodontal therapy. We reported the case study that experienced a good prognosis in the maintenance phase more than ten years with progressed periodontal disease patients that was treated with medication of azithromycin before SRP.
*Case*: Patient: 52-year old female, First visit: July 20th. 2007 Chief complaint: Restoration dislodgement Medical history: Asthma, non-smoker Diagnosis: Generalized moderate chronic periodontitis Examination and findings: #37 has chronic suppurative apical periodontitis. Inflammation was observed in gingiva around teeth. Peri-apical radiographs showed bone absorption. Initial PCR was 68%, PPD＞4mm was 59%, BOP was 100%. Bacteria test by PCR-Invader detected P. gingivalis.
*Clinical procedure and outcome:* 1. Initial therapy: Oral hygiene instruction, Scaling, full mouth SRP under medication of azithromycin and extraction of #37.
2. Re-evaluation
3. Oral function recovering therapy
4. Re-evaluation
5. Maintenance P.gingivalis was not detected after SRP. The value of PCR, PPD and BOP was dramatically improved at the end of treatment phase（PCR; 14%, PPD＞4mm; 0%, BOP; 0%）. Maintenance is performed every two months. Even after 10 years, the patient keeps good prognosis（PCR; 11.5%, PPD＞4mm; 0% BOP; 0%）and periapical radiograph shows regeneration of intrabony defect.
*Conclusion:* SRP with oral administration of azithromycin at the periodontal initial therapy improved periodontal parameters remarkably in the short period. One-time full mouth disinfection with medication of azithromycin is effective and has favorable prognosis.

H-05

## A case report of severe chronic periodontitis with an adult bronchial asthma

Eri Otsuka*

Nadyapark dental clinic

*Background and objective:* This presentation is to report a positive effect brought about by a comprehensive treatment to a generalized severe periodontitis with bronchial asthma.
*Case:* The patient was a 42-year-old male.The first visit was in January 2015. The patient had an adult bronchial asthma and an acute hepatitis for medical history.The chief complaint was bleeding of the gum.Clinical examination at the first visit revealed 77% of sites with a probing pocket depth（PPD）over 4mm and 64% of sites with bleeding on probing（BOP）. Plaque control record（PCR）was 68.3%.There were class Ⅱ mobility in #37/#18 and furcation involvement in #16/#2,#17/#3,#26/#14,#27/#15,#37/#18,#46/#30,#47/#31.On the X-ray, holizontal bone defect in the molar teeth and vertical born defect in #12/#7 were observed.P.g., T.d., P.i. were detected in real-time polymerase chain reaction method.The patient was diagnosed with severe chronic periodontitis.
*Clinical Procedures and Outcomes:* Azithromycin and metronidazole were prescribed during the scaling and root planning for 2weeks.The PPD, BOP and PCR were improved by initial preparation.After the second examination, the number of periodontal pathogenic bacteria had desreased and supportive periodontal therapy was started.Although the patient had an adult bronchial asthma from 2009, it was cured after the periodontal initial preparation.
*Conclusion:* Periodontal disease will relate to respiratory diseases such as pneumonia and asthma. So that, this case indicates adult bronchial asthma could be relieved by periodontal treatment with antibiotics.

H-06

## A Case Report: Effects of Applying Numerical Rating Scale in Treating Halitosis Associated with Advanced Periodontal Disease

Sayuri Kida*[1], Isao Ohashi[1], Masahiko Nikaido[2]

[1]Ohashi Dental Clinic, [2]Nikaido Dental Office

*Background:* Halitosis is one of the major symptoms associated with advanced periodontal disease. We applied Numerical Rating Scale（NRS）in the treatment of halitosis and could successfully improve it.
*Case:* 55 year -old female patient visited in our office with a chief complaint of halitosis. At the initial examination, she had 82% of sites with ≧4mm probing pocket depth（PPD）, 91% of sites with bleeding on probing（BOP）and a plaque control record（PCR）was 89%, was diagnosed as advanced periodontal disease. Her halitosis score with NRS was 10.
*Clinical Procedures and Outcomes:* Initial preparation was performed for the patient and NRS was markedly improved（NRS 1）. After completion of her dental treatment, she has been placed in a monthly maintenance program for 8 years. In a recent examination, all PPD are＜4 mm, no BOP and PCR was 6%.
Conclusion: NRS is a unidimensional measure of intensity. A respondent selects whole number（0-10）that best reflects the intensity of their feeling. In this case, she was extremely concerned about halitosis at her initial visit, since it impaired the interpersonal relationship, but as a result of initial preparation, halitosis became almost unnoticeable（NRS 1）. With that, she gained confidence in herself. We could successfully improve her halitosis by utilized NRS together with her periodontal condition and she is very satisfied with this treatment.

The 104th Annual Meeting of the American Academy of Periodontology in collaboration
with the Canadian Academy of Periodontology,
the Japanese Academy of Clinical Periodontology,
and the Japanese Society of Periodontology.
**Abstracts of JACP/JSP Poster Session**



Secretariat : c/o Atalacia.Ltd.
1-775, Funahashi-honmachi, Hirakara, Osaka
573-1116, Japan
TEL +81- 72-808-8125


The Japanese Academy of Clinical Periodontology
SINCE 1983
JACP
日本臨床歯周病学会


THE JAPANESE SOCIETY OF PERIODONTOLOGY
JSP
1958