Markers of bone turnover in DENTISTRY

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Outline

- Remodeling of the bone
- Osteopontin
- RANK/RANKL/OPG/Osteocalcin
- Alkaline phosphatase
- Matrix metalloproteinases
- Sclerostin
- Osteogenesis imperfecta
- Periodontitis; point of care testing
- Peri-implantitis
- Conclusions
Biochemical markers of bone turnover

- include markers of bone formation, bone resorption and bone metabolism regulation
- determine abnormalities in bone remodeling
- can be used to monitor the rate of bone loss in various metabolic bone disorders
- enable monitoring the efficacy of drug therapy for osteoporosis and other bone disorders
- the use of clinically relevant biomarkers provides an important noninvasive, sensitive, rapid, and real-time tool to monitor bone activity at the whole skeleton level and to investigate potential mechanisms (Smith et al, 2017)
- enable development of point of care testing methods: rapid, user-friendly, low-cost
- early diagnosis
Remodeling of the bones

- Bone REMODELING: reshaping of the outline of the bone by selective resorption of bone in some areas and deposition in other areas
- DEPOSITION: addition of new bone by osteoblastic activity
- RESORPTION: removal of bone due to osteoclastic activity
- Bone enlargement
- Osteoblasts: synthesize the organic bone matrix, rich in collagen type I, osteocalcin, and alkaline phosphatase (Long 2012), and control the mineralization process (Schminke et al. 2015); some osteoblasts are predetermined to become osteocytes (Bonewald 2011)
- Ostocytes: are important regulators of bone remodeling in response to mechanical stress (example: occlusal load of the implant), and in response to hormonal signals (Long 2012)
- Control of cellular metabolism of the bone:
  - hormones—thyroid hormone (Long 2012)
  - transcription factors: SOX9 (Akiyama et al. 2005), RUNX2 (Kronenberg 2004), and OSX (Nakashima et al. 2002)
  - different signaling pathways via bone morphogenetic proteins (BMPs) (Rosen and Thies 1992), Hedgehog (St-Jacques et al. 1999), Notch (Hilton et al. 2008), Wnt, and fibroblast growth factor (FGF) (Long 2012).
ALVEOLAR BONE:
- Continually remodeled by osteoblasts, osteocytes, osteoclasts: to adapt to mechanical loading associated with tooth function.

MANDIBLE:
- Continuous
- Mechanobiological factors: chewing force, tooth loss

CEMENTUM:
- Less influenced by mineral metabolism
- Does not undergo physiological remodeling
- Forms by continual slow apposition, mediated by cementoblasts: deposition of ECM proteins (bone sialoprotein, osteopontin)
- Apposition and mineralization regulated by: phyrophosphates (PPi), progressive ankylosis protein, ectonucleotide phyrophosphatase phosphodiesterase 1 (Enpp1/ENPP1), alkaline phosphatase
Bone remodelling cycle
## Bone resorption stimulation and inhibition

<table>
<thead>
<tr>
<th>Protein</th>
<th>Stimulation</th>
<th>Inhibition</th>
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</table>
| **Systemic** | PTH  
PTHrP  
1,25(OH)2D3  
Thyroid hormone  
beta2adrenergic | Calcitonin  
Sex steroids (Estrogen, Testosterone) |
| **Local**   | RANKL  
Macrophage CSF  
IL 1,6, 7, 11, 15, 17  
TNF alpha  
FGF  
PGL | OPG  
TGF beta  
IL 4, 10, 13, 18 |
The most common causes: age and menopausal related osteoporosis and osteopaenia

Disease related causes of unbalanced bone turnover include hyperparathyroidism, hyperthyroidism, Paget’s disease, bone metastases, multiple myeloma and rickets
High rate of bone remodelling during childhood.

Peak bone mass is being developed during childhood to puberty.

More bone is lost than made after 25-30 years old.

Gradual loss of bone and strength as we age.
Bone remodeling
The contrasting roles of Wnt and Lrp5 signaling in bone remodeling

Patricia Ducy, and Gerard Karsenty J Cell Biol 2010;191:7-13
Markers of bone turnover

**Bone formation:**
- Matrix PROTEINS:
  - osteocalcin
  - PINP (procollagen type I N-terminal propeptide)
- ENZYME:
  - bone-specific alkaline phosphatase (bone isoform)

**Bone resorption:**
- beta crosslaps (Beta-CTx)
- ICTP (carboxyterminal cross-linked telopeptide)
- sclerostin
- NTX (amino-terminal cross-linked telopeptide of Type I collagen) (urine)
- pyridinium cross-links: pyridinoline (urine) and deoxypyridinoline
- tartrate-resistant acid phosphatase isoenzyme 5b (TRAP 5b)
- cathepsin K
TRAP = tartrate-resistant acid phosphatase
Wnt signaling pathway

'Wingless/Integrated'

*Ther Adv Musculoskel Dis* (2013) 5(1) 13-31
**Osteoprotegerin (OPG):** prevents RANK-RANKL contact, thereby reducing osteoclastogenesis.

**RANKL** (receptor activator of nuclear factor kB ligand): binds to RANK receptor of the osteoclasts, activating osteoclastogenesis and induces bone resorption.
OSTEOPOROSIS

• Definition:
  • reduced bone density
  • impaired bone integrity, increased fragility

• Chronic multifactorial disease

• Classification:
  • Type 1 (primary): postmenopausal, senile
  • Type 2 secondary: drugs: heparin, GCC, Li, anticonvulsivants, immunosuppresants; several diseases, immobilisation
### Bone density (WHO)

<table>
<thead>
<tr>
<th>Bone Density</th>
<th>Diagnosis</th>
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<tbody>
<tr>
<td>&gt;833 mg/cm²</td>
<td>Normal</td>
</tr>
<tr>
<td>833-648 mg/cm²</td>
<td>Osteopenia</td>
</tr>
<tr>
<td>&lt;648 mg/cm²</td>
<td>Osteoporosis</td>
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</table>
Pathophysiology of osteoporosis

Cytokines, hormones, drugs, mechanical stress

RANKL: receptor activator of nuclear factor k beta ligand; OPG: osteoprotegrin;
Roles of pro-inflammatory cytokines in osteoclastogenesis
Bone metabolism enhanced by growth factors
Osteopontin (OPN)

- multifunctional, noncollagenous sialoprotein, expressed in osteoblasts, odontoblasts and osteocytes, component of the mineralized, extracellular matrix of bone and teeth (Sodek et al, 2000)
- essential role in bone and periodontal remodeling and orthodontic tooth movement, biomineralization, wound healing, apoptosis, bone metastasis (Sodek et al, 2000)
- bone remodeling: osteoclastogenesis and osteoclast activity through CD44- and αvβ3-mediated cell signalling, formation of podosomes, osteoclast survival and motility, regulatory effect on hydroxyapatite crystal growth, inhibits the mineralization of osteoblast cultures (Singh et al, 2018)
- SNPs in OPN coding gene Spp 1 associated with orthodontically induced root resorption (Singh et al, 2018)
- its expression is regulated by cytokines, hormones, growth factors and mechanical stress (Singh et al, 2018)
<table>
<thead>
<tr>
<th>Expression and upregulation of OPN</th>
<th>Downregulation of OPN</th>
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<tbody>
<tr>
<td>Transcription factors: Runx2 and Osterix</td>
<td>cGMP-dependent protein kinase</td>
</tr>
<tr>
<td>Inorganic phosphate</td>
<td>Biphosphonates</td>
</tr>
<tr>
<td>Hypophosphatemia, hypocalcemia</td>
<td>ERK inhibitor</td>
</tr>
<tr>
<td>Hormones: Glucocorticoids, 1,25(OH)2D3, PTH</td>
<td></td>
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<tr>
<td>Vitamins: retinoic acid</td>
<td></td>
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<tr>
<td>Inflammatory mediators: TNF alpha, IL-1 beta, TGF beta</td>
<td></td>
</tr>
<tr>
<td>Mechanical stress</td>
<td></td>
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</table>

Osteopontin and podosome formation

Singh et al. Progress in Orthodontics (2018) 19:18
Osteopontin regulates dentin and alveolar bone development and mineralization

B.L. Foster a,*, M. Ao b, C.R. Salmon c, M.B. Chavez a, T.N. Kolli a, A.B. Tran b, E.Y. Chu b, K.R. Kantovitz d, M. Yadav e, S. Narisawa e, J.L. Millán e, F.H. Nociti Jr c, M.J. Somerman b

**OPN**
- expressed during tooth development by alveolar bone osteoblasts and cementoblasts
- regulates formation and mineralization of dentin and bone
- influences tissue properties of periodontal ligament (PDL) and pulp
- does not control cementum apposition
OSTEOMALACIA

**Definition:** inadequate mineralization of osteoid

**Causes:** vitamin D deficiency in ADULTS

**Risk factors:**
- age
- rigid vegetarian diet
- reduced endogenous production, vitamin D malabsorption, renal tubular disorders, RF anticonvulsivants
Pathophysiology of osteomalacia

Excessive subperiostal accumulation of OSTEOID → Bone DEFORMITIES

**Bone**

- **Osteoid**
- **ABNORMAL Mineral crystallization**
  - $[Ca]$
  - [phosphates]

↓vit. D

↓[Ca]

↑PTH

↑[Ca], ↑renal excretion of phosphates

inadequate MINERALIZATION
Alkaline phosphatase (ALP)

- circulating alkaline phosphatase is derived from bone, liver and placenta (3 isoenzymes)
- normal value: 30-120 UI/L (Bone isoenzyme 15-40 UI/L)
- membrane bound glycoprotein derived from leukocytes, macrophages, osteoblasts and fibroblasts, resident bacteria of the sulcus or pockets (Gupat et al, 2018)
- the bone fraction may be:
  - increased in: hyperparathyroidism, Paget’s disease, tumors or bone metastases (osteolysis), osteomalacia, rickets; bone growth, fracture repair
  - decreased in malnutrition.
- in gingival crevicular fluid: increased due to abundance of PMN at the site of periodontal inflammation; decreases after non-surgical periodontal therapy (Kanjappu et al, 2012; Gupta et al, 2018)
Osteocalcin

- an important marker of bone formation and osteoblastic activity with tissue specific expression
- non collagenous, vitamin K dependent, calcium binding protein
- synthesized by osteoblasts under the same conditions as alkaline phosphatase, being a marker of bone remodeling
- increases in disorders characterized by increased bone remodeling: Paget’s disease, bone metastases, primary hyperparathyroidism;
- physiologically increased in children, particularly during the first year of life and during puberty, period characterized by rapidity of physical growth (Cioffi et al, 1997; Marginean et al, 2017)
- decreases in diseases characterized by low bone remodeling (hypoparathyroidism) and chronic periodontitis (Gupta et al, 2018)
- in gingival crevicular fluid
- significant correlations among BMD, BMI, and Osteocalcin results in postmenopausal women
- each examination of BMD, BMI, and Osteocalcin can be used to identify the risk of osteoporosis in postmenopausal women.
- simple examination of osteocalcin can be used to predict mandibular bone loss.
Beta-crosslaps

• secreted and released into the bloodstream by osteoclasts during bone resorption together with a mixture of protease which degrade the collagen fibrils into molecular fragments.

• an important marker for the degradation of mature type I collagen and stable, specific marker of bone resorption:
  • type I collagen → C-terminal telopeptides (CTx) → beta-CTx

• highest values in neonates, reaches a nadir between 1 and 9 years of age and increases again during early adolescence in both sexes, attaining its peak earlier in girls than in boys (Crofton et al, 2002)

• pathological elevated levels are associated with osteoporosis, osteopenia, hyperparathyroidism, hyperthyroidism, Paget’s disease

• detect response to bone formation and anti-resorptive therapies for bone disease

• monitoring the therapeutic effects of estrogen replacement therapy on bone mineral density and bone turnover in postmenopausal women treated for osteoporosis and individuals diagnosed with osteopenia

• in children: detecting and managing metabolic bone disorders (osteogenesis imperfect, Paget disease, rickets, and secondary osteoporosis)
Normal values for Beta-crosslaps

- It is recommended to draw blood as fasting morning samples due to a circadian rhythm of Beta-CTx release. All follow up samples should also be taken in the fasted state.

- The patient needs to fast prior to testing: 12 hours before the test no dietary or multivitamins supplements which contain Vitamin B7 and biotin

<table>
<thead>
<tr>
<th>Type</th>
<th>Gender</th>
<th>Age-Group</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Beta Cross Laps</td>
<td>UNISEX</td>
<td>All age groups</td>
<td>35-950 pg/mL</td>
</tr>
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</table>
Matrix metalloproteinases are a family of zinc and calcium dependent endopeptidases. They play a key role in periodontal extracellular matrix degradation and remodeling (Gupat et al, 2018) in gingival crevicular fluid: MMP-3 (Kanjappu et al, 2012), MMP-8, MMP-9 (Kinney et al, 2014; Skurska et al, 2015), MMP-13 (Pawar et al, 2015). They are increased in periodontitis; and decrease after non-surgical periodontal therapy.
Tissue inhibitors of matrix metalloproteinases restrict extracellular breakdown in gingival crevicular fluid:

- Increases with progression of periodontal disease
- Decreases after non-surgical periodontal therapy (Kumar et al, 2013)
Sclerostin

- a protein product of the SOST gene
- inhibits osteoblast activity via antagonism of the Wnt signaling pathway
- elevated in osteoporosis, immobilization-induced bone loss, rheumatoid arthritis, multiple myeloma and bone metastases (Rey et al, 2010; Gaudio et al, 2010; Colucci et al, 2011)
- therapeutic target of great interest for the fields of bone and cancer research
Calprotectin

- Involved in the inflammatory reaction: coordination of adhesion and migration of neutrophils or monocytes
- Important contribution to host defense against infection (Becerek et al, 2011)
- Levels in gingival crevicular fluid higher in patients with periodontitis
Structure of collagen with its terminal telopeptides.

Puri, Shweta, "Are Bone Turnover Markers and Vitamin D levels associated with Frequency of Complete Denture Relines?" (2015)
Significant positive correlations between C-terminal telopeptide and frequency of reline

Puri, Shweta, "Are Bone Turnover Markers and Vitamin D levels associated with Frequency of Complete Denture Relines?" (2015)
Puri, Shweta, "Are Bone Turnover Markers and Vitamin D levels associated with Frequency of Complete Denture Relines?" (2015)
Regulators of calcium and phosphate metabolism

- **PTH in plasma** *(NV=10-65 ng/L)*. Dosage is performed by radioimmunoassay (RIA) and is a valuable tool for assessing parathyroid gland function.

- The dosage of vitamin D metabolites: 25 hydroxycholecalciferol level *(NV=30-80g/L)* and 1,25 dihydroxycholecalciferol *(NV=20-75 g/L)*, are useful for diagnosis of disorders characterized by insufficient exposure to ultraviolet light and vitamin D metabolism disorders.

- Serum calcitonin *(NV<10 ng/L)* - useful in the diagnosis of bone metastasis.
Therapy with pamidronate in children with osteogenesis imperfecta

Abstract: Osteogenesis imperfecta (OI) is a genetic disease characterized by excessive bone fragility with fractures consecutive to minor trauma. Considering lack of standardization of therapy with pamidronate in children, it was our aim to present our experience over a period of 10 years regarding evolution and treatment in patients diagnosed with osteoporosis and OI. Nine patients diagnosed with OI were admitted to the First Pediatric Clinic, Timisoara. They were investigated (clinical, biomarkers of bone metabolism and imaging studies), and a quality-of-life questionnaire was used to evaluate the impact of OI. Treatment was performed with pamidronate 1 mg/kg/cycle, every 3 months. The patients were evaluated every 3 months. The most frequent was type III (three patients), and two patients were diagnosed with type II, while the other patients were diagnosed with other forms such as types IV, V, VI and VIII. The clinical expression was polymorphic, and the number of fractures was variable. Bone pain ameliorated just after the first cycle of pamidronate, while the activity and mobility increased quickly. Osteodensitometry in children over 12 years showed a decreased bone mineral density (BMD) with a significant improvement after treatment. The values of the bone alkaline phosphatase and osteocalcin changed after the antiresorptive treatment, and the quality of life of the children and their family improved. Treatment with pamidronate is beneficial for the patient, family and society, increases mobility and bone density, improves quality of life and reduces family dependence in children with OI.

Keywords: osteoporosis, child, osteogenesis imperfecta, pamidronate
25(OH)D
Serum alkaline phosphatase
Osteocalcin
Beta-crosslaps
Z-score

Assessed:
1. hospital admission
3, 6 months after therapy start
annually

Figure 3 Dentinogenesis imperfecta with brown-yellow spots, malposition and friability.
Therapy with pamidronate in children with osteogenesis imperfecta

- Pamidronate in glucose 5% solution intravenous 0.5 mg/kg (1st dose), 1 mg/kg/cycle (next doses) over 3-4 hours; cycles of 1-3 days
- Follow up: 3-6 years
- Clinical and biological changes evaluated every 3 months during therapy
- Radiological and quality of life parameters: yearly
Therapy with pamidronate in children with osteogenesis imperfecta
Periodontitis: a chronic inflammatory disease of the gums with irreversible changes in the supporting structures of the teeth (gums, periodontal ligament, alveolar bone)

The gum and bone retreat away from the teeth, forming pockets between the bone and the tooth, enabling accumulation of bacteria, tartar, necrotic bone fragments.

Periodontitis: Imbalance between increased oral bacterial mass and host defense mechanisms

- Inflammation, immune mechanisms
- Virulence factors
  - Oral bacteria
  - Oral bacteria
- Host defense mechanisms

Mozos, 2017
Bacterial lipopolysaccharides (BLPZ)

- Activation of lymphocytes, monocytes, fibroblasts
- Inflammatory mediators: cytokines, metabolites of arachidonic acid
- MATRIX metalloproteinases
- Destruction of the extracellular matrix and bone
Specific pathogenic bacteria

Immune and inflammatory response of the host

Affecting connective tissue and bone

Clinical expression of periodontitis

Environmental and genetic factors

Atb, PMN

Ag, virulence factors

Cytokines, Pgl

MMP

Mozos, 2017
Biomarkers in periodontitis

Bacteria derived biomarkers:
Porphyromonas gingivalis, Treponema denticola, Tannerella forsythia, Actinobacillus actinomycetemcomitans, Prevotella intermedia

Host-derived biomarkers related to **soft tissue destruction**: interleukin 1 beta (IL1beta), IL8, TNF alpha, elastase, aspartate aminotransferase (AST), MMP-8, prostaglandins, C reactive protein (CRP), calprotectin, cathepsin G

Host-derived biomarkers related to **bone tissue destruction**: IL1beta, IL6, TNF alpha, pyridinoline crosslinked C-terminal telopeptide of type I collagen (PCCTC), RANKL, OPG, PGE2

Kido et al, 2017; Mozos 2017; He et al, 2018
Pathogenesis and associated biomarkers of human periodontitis

Physiological structure and response of the activated immune cells (PMNs, monocytes, fibroblasts) to periodontal pathogens and released proinflammatory cytokines and proteases

He et al, 2018. Trends in Biotechnology
Inflammatory mediators in periodontitis as markers of bone destruction

**IL1**: proinflammatory effect, bone resorption, stimulates release of MMP, PGE2

**IL6**: production of Atb, osteoclast formation, bone resorption

**IL8**: chemotactic effect for N, stimulates activity of MMP, collagen destruction

**TNF alfa**: proinflammatory effect, bone resorption, release of MMP, degrade connective tissue

**PGE2**: bone resorption, soft tissue destruction, MMP secretion

Mozos, 2017; He et al, 2018
Markers in periodontitis

**AST: aspartate amino transferase**, an enzyme enabling the transfer of an amino group from glutamic acid to oxalacetic acid; found in fibroblasts, epithelial cells, PMNs; indicates tissue destruction

**Elastase**: a serine endopeptidase stored in the granulocytes; released from dead neutrophils of the periodontium, can react with several proteins: fibrinogen, proteoglycans, elastin, collagen; participates in tissue breakdown and combating infection

**MMP-8**: degrades almost all basement membrane components and the extracellular matrix; it is produced and activated by host inflammatory mediators (IL-1beta, TNF alpha); more abundant in the active than the resting state

**Cathepsin G**: serine protease of the chymotrypsin family; antibacterial action; extracellular breakdown of ECM components at inflammatory sites

**Calprotectin**: found in the cytosol of PMNs, monocytes, gingival keratinocytes, oral epithelial cells

Zhou et al, 2012; He et al, 2018
Point of care testing platforms in periodontitis

**Lab on a chip (LOC) platforms**: small volumes of fluids and reagents, fast reaction; by miniaturizing and integrating complex laboratory procedures into a small chip; immunoassay: MMP-8, IL-1β, C-reactive protein in saliva (Christodoulides et al., 2007), calprotectin in GCF (Kido et al., 2012), bacterial cultures (Gaertig et al., 2015)

**Paper-based platforms**: dipstick assays, lateral flow assays, microfluidic paper-based analytical devices; PGt in saliva/subgingival plaque, MMP-8 in saliva and GCF, nitrite in saliva, cathepsin-G and human neutrophil elastase in saliva

**Chairside tests**: disease activity, future progression, treatment efficacy; time consuming; PGt and bacterial cultures from subgingival samples/bacterial toxins and proteins, neutral proteases in GCF

**Wearable devices**: long term monitoring of physiological and biochemical factors, diagnosis and surveillance of oral diseases; requires external power and reader; salivary metabolites: lactate, uric acid, glucose

He et al., 2018. Trends in Biotechnology
LOC POCT platforms for periodontitis diagnosis

A. LOC (fluorescent sensors and optical system) to detect MMPs, IL-1beta, CRP in saliva (Christodoulides et al, 2007)
B. IMPOD (integrated microfluidic platform for oral diagnosis): microchip based on an ELFO immunoassay to detect salivary proteins in a small sample volume (Herr et al, 2007)
C. Microchip (ELISA): calprotectin in GCF (Kido et al, 2012)
D. PCR chip system (Gaertig et al, 2015)
Paper-based platforms for periodontitis diagnosis

Lateral flow assays with 4 layers to detect:

B. **PGI in subgingival plaque** (Imamura et al, 2015): initiation stage of periodontitis
C. **MMP-8 in GCF** (Mantyla et al, 2003)
D. **Nitrite detection in saliva** (Bhakta et al, 2014): potential biomarker of periodontitis
E. **MMP-8 and MMP-9 in saliva** (Yee et al, 2017)
F. **Cathepsin-G and human neutrophil elastase in saliva** (gold nanoparticles and magnetic beads) (Wignarajah et al, 2015)

Microfluidic paper-based analytical devices: hydrophilic main channel, branched channels, uptake zones, testing zones, hydrophobic wax barrier
Periodontitis diagnostics and future perspectives for POCT strategies

He et al., 2018. Trends in Biotechnology
Future development of POC periodontitis testing

- **Standard collection and treatment protocol for samples**
- **Prevention:** simple, qualitative or semi-quantitative methods
- **Prognosis:** quantitative methods (more accurate results)
- **Long distance medicine:** mobile app, website
- **Personal file record of every user:** (history, results)
- **Cloud database:** diagnostic results, therapy, prognosis
Peri-implantitis

- oral inflammatory disease affecting the surrounding tissue of the implant, predominately the bone; 30% of patients with dental implants (Schminke et al, 2015)
- long-term stability of dental implants: osseointegration into the alveolar bone (Brånemark et al, 1969)
- the clinical long-term survival of an osseointegrated implant depends on:
  - implant materials, two-piece implant systems (Tallarico et al, 2017), surface structure, cement excess (Pèsce et al, 2015)
  - bone quality (aggressive periodontitis), mechanical loading, genetic markers
  - food impaction into the periimplant sulcus (Bidra, 20114)
  - systemic diseases
  - bacterial biofilms, which leads to peri-implant infections: peri-implant mucositis and peri-implantitis (Esposito et al, 1998); results in bone loss and destruction of soft tissues (Scarano et al, 2004); loss of alveolar bone leads to instability and, finally, to loss of the implant (Lang et al, 2011); Porphyromonas gingivalis, Prevotella intermedia, Aggregatibacter actinomycetemcomitans, Treponema denticola, Tannerella forsythia, Fusobacterium nucleatum are associated with biofilm formation on dental implants (Leonhardt et al, 1999; Tallarico et al, 2017).
Gene expression of peri-implantitis:
A: microarray: upregulated genes (yellow), downregulated (blue)
B: osteogenic genes of bone matrix molecules are downregulated, IL-8 is upregulated
C: reverse transcription PCR: downregulation of osteogenic genes in inflamed periimplantitis tissue: just IL-8 and BMP-2 are upregulated

Reduced RUNX2 expression: fibro-osteoblastic cells generate a more fibrous tissue, with less osteogenic markers
Conclusions

• Bone remodelling is important for the proper metabolic functions and structure of the bone

• Osteopontin, osteocalcin, C-terminal telopeptide and vitamin D provide value information related to bone-turnover in dentistry

• **Combining multiple** biomarkers related to disease development provides more comprehensive information than single biomarkers, enabling a personalized treatment

• Point of care testing deserves special attention in assessing markers of bone-turnover
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