



Minimally-invasive diagnostic approaches in periodontics: laser Doppler imaging and optical coherence tomography

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Abstract

Evaluating the health status of the gingival tissue represents an important objective in the dental daily practice. Inflammation changes the microcirculatory and micro-morphological dynamics of human gingiva which cannot always be determined only by current clinical examination methods and which may lead to unpleasant complications if not diagnosed and treated at the right moment. Laser Doppler Flowmetry and Laser Doppler Imaging could be a useful, noninvasive, sensitive, reproducible, and harmless methods for measuring gingival blood flow in humans. Optical Coherence Tomography is a non-invasive method of imaging the dental microstructure and can potentially be used to evaluate periodontal tissues. It offers an ‘optical biopsy’ of tissues at a 2–3 mm depth. These techniques provide a different evaluation of periodontal inflammation and can either replace traditional clinical examinations or serve as adjuncts for periodontal diagnosis.

Keywords Gingival blood flow · Non-invasive periodontal diagnosis · Optical biopsy · Biologic imaging

Quick reference/description

Owing to the limitations of the current clinical tools of periodontal diagnosis, adjunctive or alternative diagnostic techniques that utilize non- or minimally-invasive tools are essential for an accurate diagnosis. The benefits of these novel diagnostic tools are their non-invasive nature, peri-procedural patient comfort and no requirement of tissue extraction. A new paradigm of periodontal diagnosis would ultimately facilitate the effective management of patients with periodontal disease.

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Overview

Diagnostic approaches for periodontal diagnosis	Indications
Laser Doppler imaging	<ul style="list-style-type: none"> - Determination of tissue vitality - Evaluation of marginal gingival health status - Monitoring tissue response to periodontal therapy - Identification of patients at risk for adverse reactions like irreversible inflammation, avascular necrosis and tissue loss
Optical coherence tomography	<ul style="list-style-type: none"> - Evaluation of the health of periodontal tissues - Determination of probing depth and clinical attachment level - Qualitative and quantitative detection of morphological changes in dental hard and soft tissues - Detection of dental caries and oral cancer - Evaluation and prediction of tooth response under orthodontic forces - Prediction of gingival phenotype before surgical periodontal procedures

Materials/instruments

- Laser Doppler flowmetry probe
- Gingival dam
- High-speed handpiece
- Fissure bur
- Rigid opaque plastic tube
- Laser Doppler periodontal probe
- Optical coherence tomography probe

Procedure

Periodontal diseases are characterized by gingival or periodontal inflammation, or tissue destruction. Its diagnosis includes a thorough periodontal examination involving the patient's medical and dental histories, detection of clinical signs of

inflammation and evaluation of other signs or symptoms like pain, ulcers, presence of plaque and calculus, probing depths, pattern and extent of bone loss and clinical attachment loss.

Periodontal probing and conventional radiography are the traditional tools of routine periodontal examination. Patient discomfort and diagnostic inaccuracy are prevalent with periodontal probing. Effective monitoring of disease status, progression, and treatment response is also impaired by its low sensitivity and reliability. With conventional radiography, early detection of periodontal disease is not possible due to underestimation of bone loss. Detecting the exact location (buccal or lingual) of bony defects is difficult in two-dimensional radiography, despite the exposure to ionizing radiation. It also does not provide information about soft tissues. Routine utilization of CBCT for periodontal diagnosis is unjustified due to its relatively poor spatial resolution and exposure to high levels of ionizing radiation.

Owing to the pitfalls of the current clinical tools of periodontal diagnosis, adjunctive or alternative approaches using non- or minimally-invasive diagnostic tools are necessary for an accurate diagnosis. The key advantages of these diagnostic tools are their non-invasive nature, perioperative patient comfort and no requirement of tissue excision. However, these approaches need meticulous examination due to the multifactorial character of periodontal disease. A new paradigm for periodontal diagnosis would ultimately facilitate the management of periodontal disease (Fig. 1).

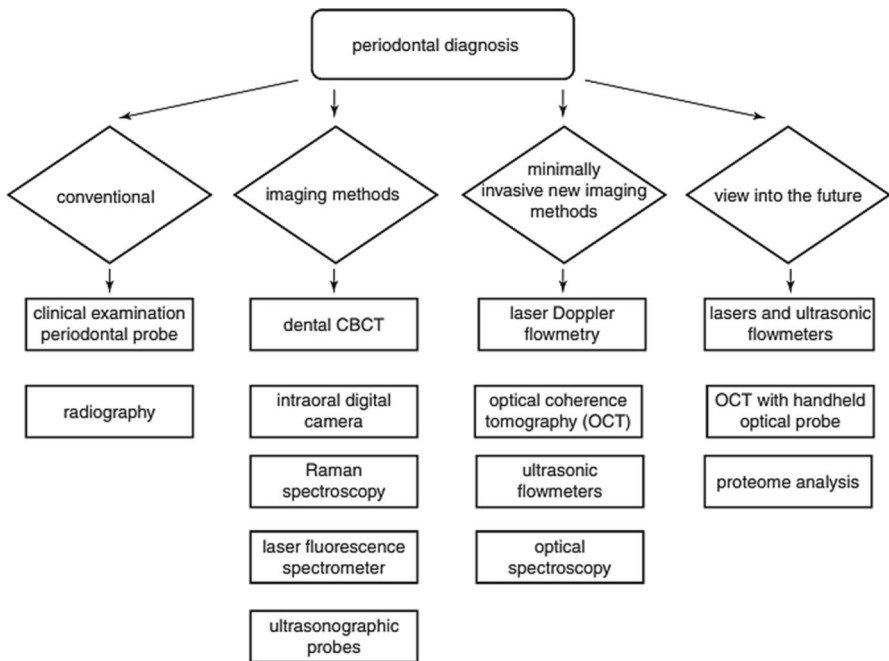


Fig. 1 Overview of existing techniques, currently available new minimally invasive imaging methods, futuristic views

Periodontitis causes destruction of the tooth supporting tissues and tooth loss with several implications on systemic health leading to a negative impact on the quality of life. It is regarded as a major factor in the global burden of oral diseases. There appears to be a strong association between gingival health and gingival blood flow (GBF). Vascular inflammatory changes are related to blood flow changes and can act as early signs of pathology in the gingival tissues.

Periodontal inflammation can cause ulcers in the gingival sulcus and expose the blood capillaries to microbial biofilms. Periodontal pathogens are translocated and released into the blood from the sulcus disrupting the microcirculation. Conversely, microcirculatory dysfunction can affect tissue perfusion and lead to organ dysfunction.

The novel diagnostic tools can be useful in the prediction, prevention and management of gingivitis and periodontitis. Each diagnostic approach gives distinct information about the vascular changes within a particular tissue volume. Hence, combined use of these tools is advisable for optimal diagnosis and treatment of periodontal disease. These techniques provide a different evaluation of periodontal inflammation and can either replace traditional clinical examinations or serve as adjuncts for periodontal diagnosis.

Periodontal diagnostic procedures aim to provide data about the type, severity and location of periodontal disease for effective treatment planning and disease monitoring. Traditional diagnostic tools are usually inadequate in detecting active disease sites, establishing accurate diagnosis and effectively planning treatment. They are also insufficient for quantitative monitoring of therapeutic response and for judging patient susceptibility to disease progression. Sensitive and specific diagnostic tests for periodontal diseases are being developed to improve the treatment.

Laser Doppler flowmetry

Laser Doppler flowmetry (LDF) was introduced more than 30 years ago as a non-invasive and real-time technique for measurement of perfusion. LD methods can exhibit changing blood flow wave patterns in different types of gingival tissue and maintain consistency of same-subject measurements over time. In gingivitis, altered vascular architecture and microvasculature are the earliest signs of inflammation. If the morphological vascular inflammatory changes are associated with blood flow changes, they can act as early predictors for the onset of gingival pathology. Certain steps were followed by the authors to obtain LDF measurements of GBF.

- Position the LDF probe 4 mm above the cervical line of the upper incisors and distance it with a gingival dam to avoid pressure on the gingiva (Fig. 2).

- Use a silicone rubber holder to secure the gingival LDF probe in position. A small hole for the laser probe is placed in the holder, 4 mm away from the gingival margin, using a 1.5 mm diameter fissure bur in a high-speed handpiece (Fig. 3).

- The probe is inserted into a rigid opaque plastic tube (diameter – 1.5 mm; length-0.1–0.2 mm longer than the fiber) after calibration and disinfection (Fig. 4). The tube decreases movement artifacts of the fiber inside the impression.

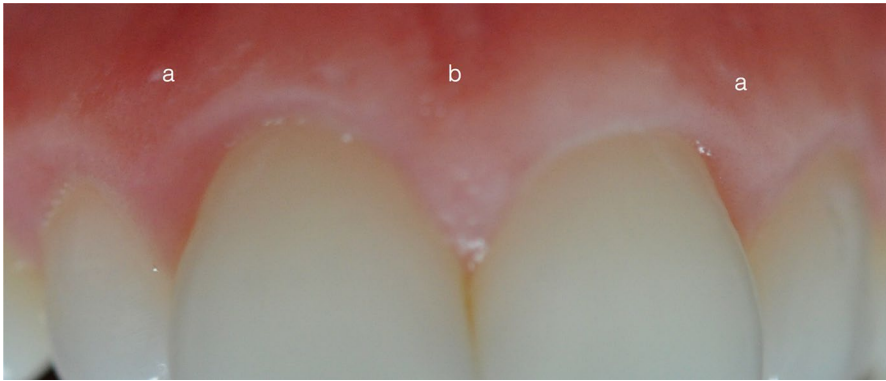


Fig. 2 Desired position of the LDF probe above the cervical line of the upper incisors. **a** Supposed gingivitis area **b** supposed healthy area



Fig. 3 Silicon rubber holder. **a** creating the holes for the laser probe with a fissure bur in a high-speed handpiece **b** the correct positioning of the holes for the laser probe

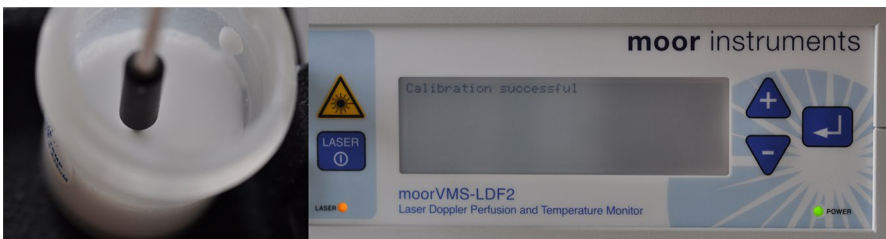


Fig. 4 Correct calibration and disinfection of the laser probe

- The plastic tube is inserted in a canal in the impression and positioned afterwards. A guiding mark on the fiber permits consistent placement of the probe to ensure reproducibility of the LD signal acquisition (Fig. 5).

Laser Doppler flowmetry provides data about blood flow in the marginal gingiva. The blood flow in different parts of the gingiva (marginal, interdental or attached gingiva or alveolar mucosa) as well as in different sites of the marginal gingiva is different. LDF variability can be affected by scattering from the surrounding tissues and morphological characteristics (gingival thickness, periodontal biotype, age and

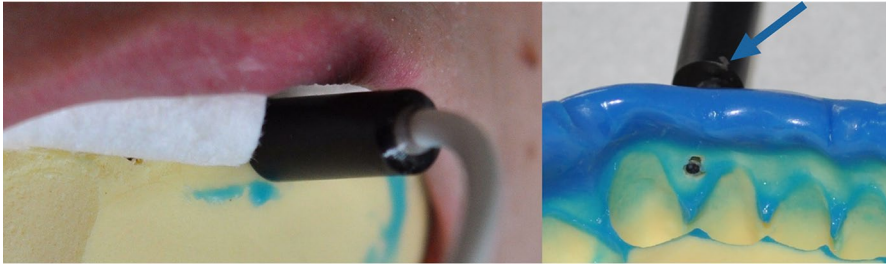


Fig. 5 Correct positioning of the laser probe in the silicon rubber holder and in the mouth

epithelial thickness). Mechanical stimulation of gingiva increases GBF in the papillary region. Marginal blood flow can also be affected by restorations or plaque accumulation. Sub-gingival restorative margins result in higher blood flow values.

Gingival microcirculation responds dynamically to the development and progression of gingivitis. LDF is an objective non-invasive method of monitoring tissue response to periodontal therapy. It can be used to quantify GBF after periodontal surgery and map the changing patterns of microvascular blood flow during wound healing (Fig. 6). GBF decreases immediately following anesthesia and also immediately following surgery. LDF techniques can exhibit the effects of smoking on GBF.

LDF can detect changes in GBF after orthodontic force application. These changes are correlated to the degree of force applied for teeth displacement with variation in individual responses to the same degree of force depending on the extent of tooth displacement and available interdental space. Accurate repositioning of the LDF probe is required for reliable blood flow measurements. LDF can be useful as a non-invasive, sensitive, reproducible and safe method of measuring gingival microcirculation. It can also aid in clarifying the role of GBF dynamics in

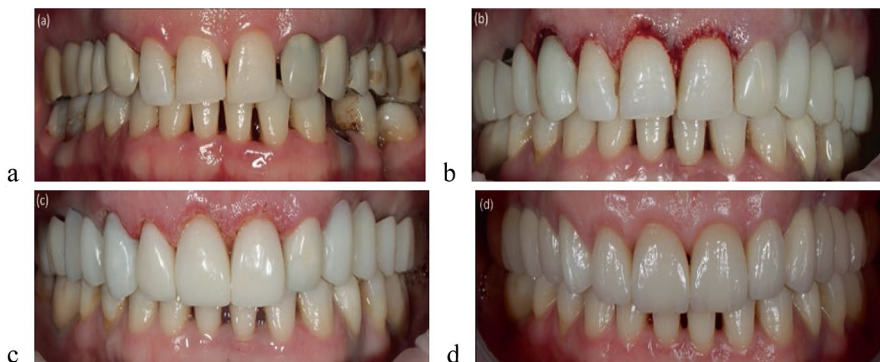


Fig. 6 Example of clinical situation, where LDF can be used to evaluate the healing process after periodontal surgery **a** initial intra-oral status **b** immediately after laser surgery **c** 24 h after laser surgery with indirect provisional restorations **d** clinical intra-oral view 2 months after final ceramic restorations

clinical gingivitis. Consequently, clinical signs of inflammation can be correlated with changes in GBF.

A laser Doppler periodontal probe has been developed for intrasulcular measurement of GBF. It is considered as an unbiased non-invasive technique of monitoring tissue response to periodontal therapy. LDF techniques can monitor post-surgical gingival recovery and provide information about the microvascular changes during healing.

Laser Doppler imaging

Laser Doppler imaging (LDI) works by scanning a monochromatic laser across a tissue surface. According to the Doppler principle, light that is backscattered from moving erythrocytes, undergoes a frequency shift proportional to its velocity. Most

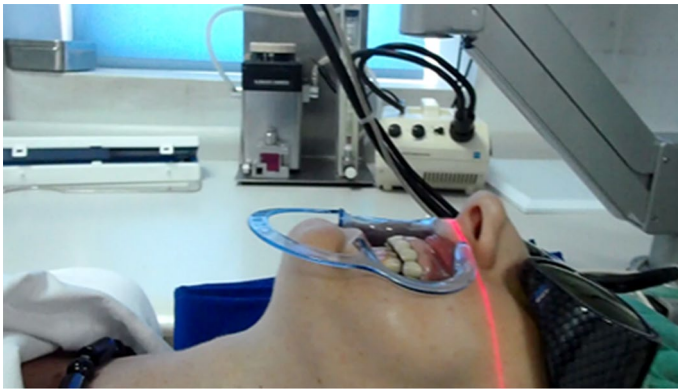


Fig. 7 Laser Doppler Imaging setup with a helium–neon laser

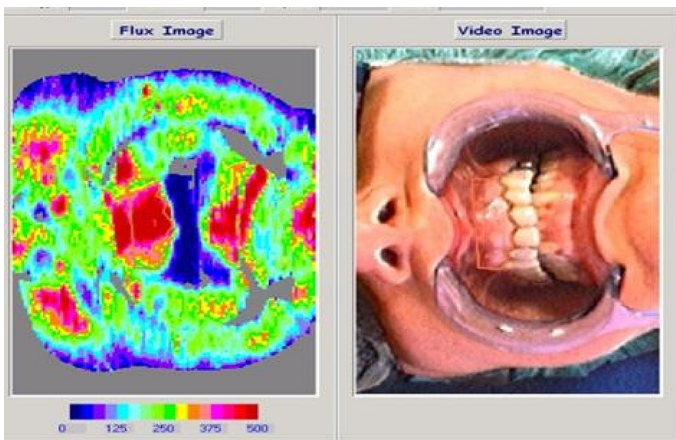


Fig. 8 Example of laser doppler image of gingival blood flow

laser Doppler setups use a helium–neon laser (RED, 632.8 nm) to provide an estimate of perfusion to a depth of 1–1.5 mm into the dermis (Fig. 7). It mainly measures the perfusion in arterioles, venules and capillaries. LDI provides a ‘snapshot’ of perfusion at any given point (Fig. 8).

The advantages of LDI over LDF are:

- No need of direct tissue contact (maximum distance: 19 cm)
- Measurement repeatability
- Capability for global analysis of blood flow in the area of interest
- Easy learning curve for surgeons

Regular postoperative assessment of flap perfusion by LD line scanning can be a practical alternative to complex and invasive monitoring techniques. An advantage of LDF over LDI is that it provides a constant measure of blood flow at any specified point.

The major advantages of LD techniques are their non-invasiveness, and their ability to measure tissue microcirculation flux and fast stimulus-induced changes in perfusion. LDF is an important tool to evaluate gingival and pulpal microcirculation, and mapping tooth vitality. It can assess the degree and duration of inflammation or ischemic episodes, which can detect patients at risk for adverse reactions like irreversible inflammation, avascular necrosis and tissue loss. LDF is suitable for determining tissue vitality in most clinical situations and can be used with other indices to assess marginal gingival health status.

Doppler flowmetry measures blood flow within a small tissue volume ($\sim 1\text{mm}^3$) that causes poor reproducibility due to the spatial heterogeneity of tissue blood flow and movement artifacts. Recent use of ‘integrated probes’ has improved its reproducibility. These probes use multiple collecting fibers positioned in a ring around a central light delivery fiber resulting in increased spatial resolution. However, LDI offers a larger surface area measurement and should be preferred in tissue areas with high spatial variability, despite the marked difference in cost.

Optical coherence tomography

Optical coherence tomography (OCT) is a non-invasive method of imaging the dental microstructure and can potentially be used to evaluate periodontal tissues. It offers an ‘optical biopsy’ of tissues at a 2–3 mm depth. OCT was proposed as a biologic imaging system by Huang et al. in 1991. In vivo dental OCT images show important anatomic structures for diagnostic evaluation of both hard and soft oral tissues. Periodontal tissue contour, sulcular depth and connective tissue attachment are seen at high resolution with OCT. Hence, it can identify active periodontal disease before substantial bone loss occurs.

OCT is potentially a better diagnostic tool for determining attachment level in terms of reproducibility and reliability than traditional probing methods. The target tissue is imaged using a non-contact probe having a focal plane at a distance from the probe tip. The tissues lying within the depth of field of the probe optics are

imaged. The non-contact probe prevents soft tissue compression and enables direct geometric measurement of tissue dimensions in its natural state. OCT is a powerful tool for producing high-resolution, cross-sectional images of oral structures.

Detection and removal of sub-gingival calculus is essential for effective treatment of periodontitis. Sub-gingival calculus is difficult to detect as it is firmly attached to root surfaces within periodontal pockets. A fiber-probe swept-source optical coherence tomography (SSO-CT) technique combined with quantitative measurement of optical parameters can be used to distinguish sub-gingival calculus from sound enamel. The real-time imaging capability and high resolution of OCT can aid in monitoring periodontal ligament changes during orthodontic treatment.

OCT is a diagnostic tool that develops near-histologic tomographic images without a biohazard. Due to its high resolution and safety, it shows greater potential for use in periodontal diagnosis. Real-time OCT can also help to evaluate and predict precise tooth responses under orthodontic forces. It detects mechanical interfaces based on differences in light reflection. It also enables subsurface cross-sectional imaging with a 10-times higher resolution than typical ultrasound imaging systems. The advantages of OCT are:

- Use of non-ionizing near-infrared light
- Depth resolved imaging
- Rapid acquisition of data
- Three-dimensional imaging ability
- Capability to observe both hard and soft tissues
- Quantitative and qualitative assessments of oral tissues
- Requirement of minimal operator training to obtain reliable and reproducible data

OCT can non-invasively image the tooth microstructure. Several current OCT prototype systems operate in the Fourier domain (FD). FD-OCT provides simultaneous high-speed and wide field imaging. Depending on the illumination method, it can be classified as spectral domain OCT and SS-OCT. The required imaging depth to observe the gingival attachment is greater inside the furcation than in other areas due to the horizontal component of the furcal concavity.

The imaging depth of OCT is determined by two major factors: wavelength of the light source and numerical aperture of the light collecting optics. Longer central wavelength allows deeper tissue penetration. In highly scattering media like biologic tissue, the intensity of coherent backscattered light decays exponentially with depth. Increasing the wavelength markedly reduces the scattering coefficient of many biologic tissues, including enamel and gingiva but potentially decreases the spatial resolution of OCT images. A larger numerical aperture also improves imaging depth resolution as reflected light gets collected over a larger range of scattering angles.

Additional developments in dental OCT are required to improve imaging depth and quality before periodontal applications. Meticulous visualization of biologic tissues can be useful for early detection of active periodontal disease or its etiology (sub-gingival calculus). Captured images can be stored as permanent records for future comparison to gain valuable information during the diagnostic and

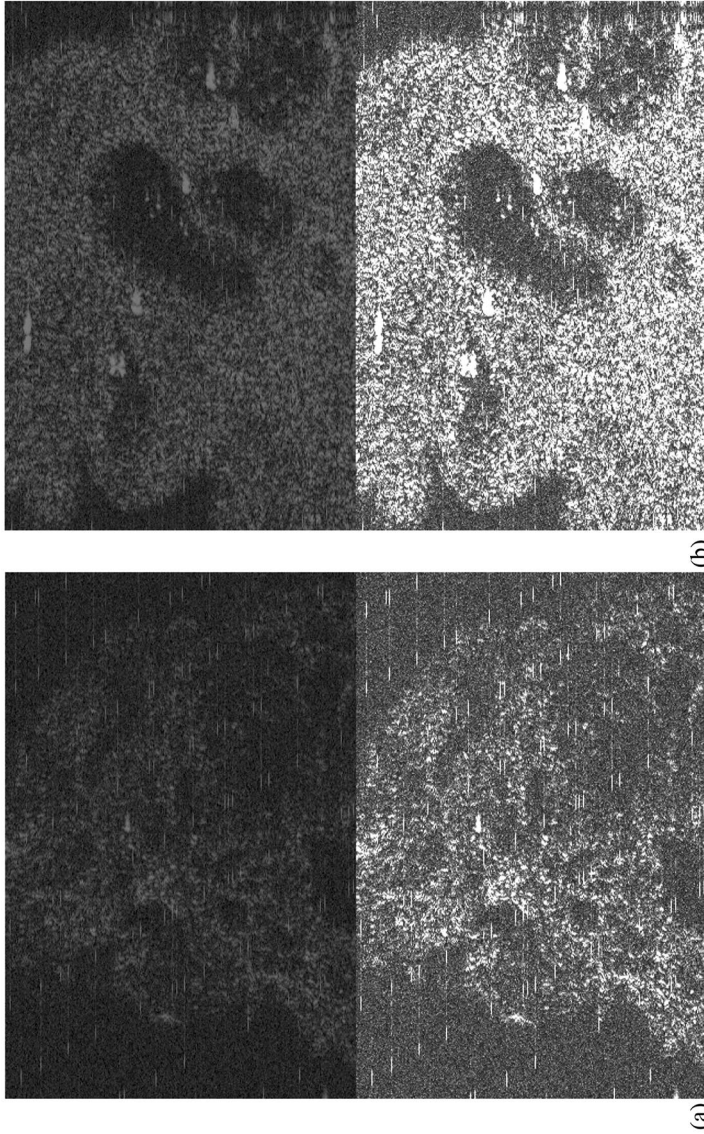


Fig. 9 En-face OCT: 2-D image of the buccal mucosa: **a** healthy tissue; **b** oral squamous cell carcinoma

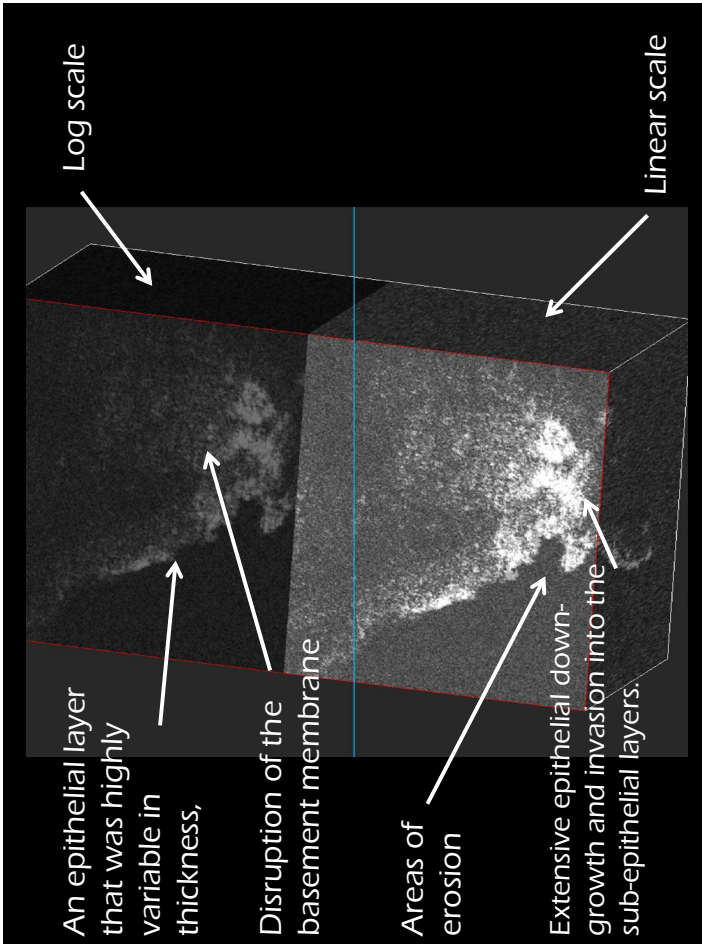


Fig. 10 OCT investigation: 3-D reconstruction of a pathologic oral mucosa

maintenance phases of periodontal therapy for detecting sites of disease recurrence or ineffective periodontal therapy.

OCT images permit measurement of gingival thickness, which is a strong predictor of gingival phenotype. Image sharpness is usually affected by axial resolution and signal-to-noise ratio but is still useful to discriminate between the epithelium and sub-epithelial connective tissue. This valuable information can affect the outcomes of periodontal therapy, gingival augmentation, root coverage and implants in esthetic areas. Estimation of epithelial thickness can be particularly useful during planning and execution of connective tissue grafting procedures as it can confirm the adequacy of undermining the epithelium at graft sites. Additionally, if palatal vessels are visualized, donor site bleeding can also be reduced.

Quantitative high-resolution OCT imaging can be used in real time during clinical procedures. The contrast between different adjacent tissues is higher when there is a greater difference in their water content. Hence, sub-gingival calculus has a higher signal intensity and contrast than the adjacent tooth surface.

OCT is an optical diagnostic tool based on interferometers. It uses a low-coherence broadband near-infrared light source to obtain excellent spatial resolution ($\sim 20 \mu\text{m}$) and real time images. It enables in situ cross-sectional imaging of tissue microstructures and can overcome the pitfalls of conventional methods of periodontal examination. Dental OCT detects qualitative and quantitative morphological changes in dental hard and soft tissues. It can also be used for early diagnosis of dental caries, periodontal disease and oral cancer (Figs. 9 and 10). Improvements in imaging depth and development of intra-oral sensors can make OCT a useful tool for periodontal applications.

Many factors affecting the intra-oral performance of OCT are improving. The wavelength choice requires optimization. Within the near-infrared window, the center wavelength determines the maximum depth of tissue penetration. When the wavelength is under 1000 nm, scattering is the main determinant as the sizes of light and tissue particles are similar. The Mie scattering theory is used to analyze this phenomenon. The absorption effect increases after 1000 nm and is maximum around 1400 nm. Water in tissues decays the input of light energy. Therefore, different wavelengths are used based on the nature of the target tissues. An OCT system with 1550 nm center wavelength is good for hard tissue measurements like enamel, dentin and alveolar bone but not ideal for soft tissues.

Target tissue composition and uniformity also affect imaging performance. Samples with rough surfaces or heterogenous composition show lower penetration depth and image contrast. The index difference between a sample and its background is also an important factor. Materials with similar refractive indices appear similar in OCT images.**

Future developments in OCT

Functional OCT (Doppler OCT and polarization-sensitive OCT) collects more information in biological tissues. DOCT gathers data on blood flow velocity and inflamed tissue volume, and PS-OCT collects data on structural orientation. For

dental in vivo imaging, improved optical probe design like probes for 3-D imaging, faster data acquisition and larger area scans are required. OCT can obtain images in seconds. However, equipment makers should optimize the balance between image quality and acquisition time for improved resolution. Future OCT systems should be telemedicine compatible with a picture archiving and communication system (PACS). This can be helpful in nursing care and in communities with limited access to dental care.

Pitfalls and complications

Drawbacks of LDF

- LDF can detect movement of red blood cells only in a small volume of tissue (1mm^3). Hence, variables like number of vessels with active flow, vessel diameter changes and flow in individual micro-vessels cannot be assessed. The small area of measurement can also affect the reproducibility of results as a minimal displacement of the optical probe can change the target site.
- Motion artifacts affect LDF measurements, and oral LDFRs show intra- and inter-individual variability.
- Uncalibrated blood flow readings are seen in each patient as LDF measurements are largely influenced by connective tissue thickness, local distribution of the vessels and the recording site (free, interdental or attached gingiva, or alveolar mucosa).
- The scattering properties of surrounding tissues affect flow readings in LDF.

Drawbacks of OCT

- OCT can image only a small sized target area (few mm^2) at any given time; thus, requiring hundreds or thousands of small images to cover an entire lesion.
- It has limited penetration depth that restricts clinical utility. This drawback can be overcome by selecting a high-quality light source, which increases the cost of the OCT system.
- Faster imaging speeds in OCT due to inadequate processing time can produce low quality images.

Further Reading

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