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## Recent advances in pulp vitality testing: A review

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### Abstract

The assessment of pulp vitality is a crucial diagnostic procedure in the practice of dentistry. Recent studies have shown that blood circulation and not innervations is the most accurate determinant in assessing pulp vitality as it provides an objective differentiation between necrotic and vital pulp tissue.

**Keywords:** Pulp vitality, blood flow, objective test

### Introduction

Diagnosis is often derived from personal and cognitive experiences. Good diagnosticians use past experience, based on knowledge and diagnostic tools. To become a successful diagnostician, one must develop a number of assets. The most important of these are knowledge, interest, intuition, curiosity, and patience<sup>[1]</sup>.

The purpose of diagnosis is to determine what problem the patient has and why does he have that problem. Ultimately, this will directly relate to what treatment, if any, will be necessary. Providing the wrong treatment for a patient could not only intensify patient's symptoms but make it even more difficult to arrive at a correct diagnosis. In simple words, diagnosis is the process whereby the data obtained from questioning, examining and testing are combined by the dentist to identify deviation from normal. The diagnostic aids can be wonderful allies to a judicious clinician in the process of decision making and can lead to both periapical and pulpal diagnosis<sup>[2]</sup>.

In 2008, the American Association of Endodontists held a consensus conference to standardize diagnostic terms used in endodontics. The goals were to propose universal recommendations regarding endodontic diagnoses; develop a standardized definition of key diagnostic terms that will be generally accepted by Endodontists, educators, test construction experts, third parties, generalists and other specialists, and students; resolve concerns about testing and interpretation of results; and determine the radiographic criteria, objective test results, and clinical criteria needed to validate the diagnostic terms<sup>[3]</sup>.

Recent studies have shown that blood circulation and not innervations is the most accurate determinant in assessing pulp vitality as it provides an objective differentiation between necrotic and vital pulp tissue. Therefore, the aim of this review is to discuss blood flow determination devices in pulp vitality testing.

### Laser Doppler flowmetry

- Pulp vitality implies that blood supply is present within the tissues. Hence, only a test that actually measures or assesses pulp blood flow can be called a vitality test. Laser Doppler flowmetry (LDF) is a noninvasive, painless, electro optical technique, which allows the semi-quantitative recording of pulpal blood flow. It measures blood flow even in the very small blood vessels of the microvasculature
- Laser Doppler method was used by Yeh and Cummins to estimate the velocity of red blood cells in capillaries. LDF was developed to assess blood flow in microvascular systems, e.g., in the retina, gut mesentery, renal cortex, and skin. It has since been widely adopted for the measurement of blood flow especially in soft tissues.
- This method was used in dentistry to study blood flow in oral tissues. Gazelius *et al.* used it first for measuring pulpal blood flow in 1986. The source of light used to measure the pulp blood flow was helium-neon (He-Ne) laser light.

- Pettersson and Oberg in 1991 used LDF instrument to assess the viability of pulp in intact and traumatized teeth.
  - They used an infrared laser diode with a longer wavelength that gave better penetration than the He-Ne wavelength. Sasano *et al.* designed, developed, and tested a transmitted laser- light flow meter that used high- powered laser light to monitor the pulpal blood flow of teeth rather than the conventional light flow- meter apparatus. Konno *et al.* in 2007, modified the apparatus and demonstrated that a high powered (5 MW vs 2 MW) transmitted light flow meter apparatus is a better tool than the conventional back scattered light flow meter apparatus in evaluating changes in pulpal blood flow in molar intrusion (animal model).
  - LDF has been found to be reliable and able to predict revascularization of the pulpal tissue. Adaption of LDF in everyday dentistry is still being actively pursued.
  - This technique involves directing a “Laser” probe held steady by a stabilizing splint made of polyvinyl siloxane (PVS) or acrylic. The PVS splinting also helps to hold the probe at an optimum angle (90°) and attenuate and reduce the ‘noise’ by providing a degree of isolation of a tooth from the surrounding tissue.
  - The laser beam is directed toward the tissue being tested and reading the reflected light that is cattered back by the moving blood cells. This reflected light is different from the incident light as it undergoes a Doppler frequency shift. This fraction of light that is scattered back from the illuminated area is detected and then processed to give a signal which is a measure of the blood flow in the dental pulp
  - The total backscattered light is processed to produce an output signal which is commonly recorded as the concentration and velocity (flux) of cells using an arbitrary term “perfusion units” (PU), (2.5 volts of blood flow = 250 PU). It is thought that the predictive modeling may provide clinicians with the opportunity to identify such teeth and initiate specific treatments.
  - Adverse outcomes are seen associated with a significant decrease in values on subsequent visits as compared to normal control teeth and favorable outcomes were seen associated with significant increase in the values on subsequent visits. It was shown that the laser can penetrate densely upto 4 mm depth and less densely for upto 13 mm length this would imply that even with good isolation, the signal contaminating nonpulpal artefactual signals aka ‘noise’ cannot be eliminated and therefore there is a likelihood of false results<sup>[4]</sup>.
- A. This original technique utilized a light beam from a helium–neon (He–Ne) laser emitting at 632.8 nm, which, when scattered by moving red cells underwent a frequency shift according to the Doppler principle
- B. A fraction of the light back-scattered from the illuminated area, shifted frequency in this way. This light was detected and processed to produce a signal that was a function of the red cell flux. This information was used as a measure of blood flow, the value being expressed as a percentage of full-scale deflection at a given gain. This method was adopted to monitor blood flow in intact teeth in animals and in man. Other wavelengths of semiconductor laser have also been used: 780 nm and 780–820 nm. Zang *et al* demonstrated greatly improved results using forward scattering detection as opposed to conventional backward scattering detection.
- C. These results were confirmed by Sasano. Odor *et al.* reported that the 810 nm wavelength showed good sensitivity but poor specificity and that the 633 nm wavelength showed good specificity but poor sensitivity. Nonlaser light (peak output at 576 nm) has also been used for the detection of pulpal perfusion. In general, infrared light (780–810 nm) has a greater ability to penetrate enamel and dentine than shorter wavelength red light (632.8 nm).
- D. LDF techniques are united in their validity for pulp vitality testing as they reflect vascular rather than nervous responsiveness. Due to some of the inherent problems associated with this technology, Sasano *et al.* considered it to be limited in its usefulness for human pulp vitality testing. The lasers used for LDF are usually at a low-power level of 1 or 2 mW and no reports on pulp injury by this method have been made. The other use of laser for diagnostics related to endodontics was the application of an excimer laser system emitting at 308 nm for residual tissue detection within the canals.

### Principle and working

- The technique depends on the Doppler principle whereby light from a laser diode incident on the tissue is scattered by moving RBCs and as a consequence, the frequency is broadened. The frequency broadened light, together with laser light scattered is photo detected and the resulting photocurrent processed to provide a blood flow measurement. LDF is an optical measuring method that enables the number and velocity of particles conveyed by a fluid flow to be measured.
- The particles (1–20  $\mu\text{m}$ ) must be big enough to scatter sufficient light for signal detection but small enough to follow the flow faithfully. The original technique used a light beam from a helium–neon (He–Ne) laser emitting at 632.8 nm. Other wavelengths of semi-conductor laser have also been used: 780 nm and 780–820 nm. Laser light is transmitted to the dental pulp by means of a fibre optic probe placed against the tooth surface. (FIG 54)
- Two equal-intensity beams (split from a single beam) intersect across the target area. The scattered light beams from moving red blood cells are frequency-shifted whilst those from the static tissue remain unshifted in frequency. The unshifted light is returned by an afferent fibre within the same probe to photodetectors in the flowmeter and the signal is produced.
- The LDF output signal or Flux can be simplified as a function of the product of red blood cells’ concentration as well as their mean velocity. It should be emphasized that the optical properties of a tooth change when the pulp becomes necrotic and this can produce changes in the LDF signal that are not due to differences in blood flow. In fact, as red blood cells represent the vast majority of moving objects within the tooth measurement of the Doppler-shifted backscattered light serves as an index of PBF. LDF evaluates dynamic changes in blood flow by detecting blood cell movement in a small volume of tissue (about 1 mm<sup>3</sup>). Most current laser Doppler devices give readout, in addition to the flux, in perfusion units (PUs) (.FIG 55,)
- If a wave with frequency  $\omega$  is scattered from a moving particle with velocity  $v$ ; the Doppler shift can be written as

$$\Delta\omega = |v||kI - ks|\cos\beta,$$

where  $kI$  is the incident wave vector,  $ks$  is the wave vector of the scattered wave, and  $\beta$  is the angle between the velocity vector and the scattering vector, which is defined as ( $kI-ks$ )

### Indications

1. Estimation of the pulpal vitality: the diagnosis of a tooth with a necrotic pulp may be difficult particularly when referred pain is present. In these situations, a suitable test and its precise interpretation are of paramount importance.
2. Pulp-testing in children: sensibility tests are not reliable in children, because they are subjective and rely upon patient's response. LDF is a suitable method for the measurement of PBF in deciduous incisors.
3. Periapical radiolucencies may have nonendodontic origins, so application of vitality tests, such as LDF can help in differential diagnosis of these radiographic views.
4. It monitors age related changes in PBF. Using this system, it has been shown that the hemodynamics in the human pulp is reduced with age.
5. Monitoring the effect of exercise on PBF. It has been indicated that PBF varies during exercise, with a mean percentage change of 38% from the level at rest
6. Monitoring of reactions to local and systemic pharmacological agents (including local anesthetic solutions).
7. Monitoring of reactions to electrical or thermal pulp stimulation.
8. Monitoring reactions to orthodontic procedures.
9. Measuring PBF after orthognathic surgery. Among patients who undergo a segmental maxillary osteotomy or Le fort I osteotomy, significant reduction in pulpal sensibility has been noted in teeth in the osteotomized segment or maxilla.
10. Measuring of PBF after traumatic injuries: Traumatized teeth may have their innervations damaged and give a negative response to pulp tests although their blood circulation and thus their true vitality is functional. LDF is an accurate and objective technique for assessment of pulpal vitality in these teeth.
11. Monitoring of revascularization of replanted teeth: LDF readings correctly predict the pulp status in vital vs nonvital teeth [6].

### Advantages

- Accurate
- Reliable
- Reproducible
- Nonpainful
- Luxation injuries

Useful in young children whose responses are unreliable and its noninvasive nature helps to promote patient cooperation and acceptance.

### Limitations

- Too expensive for a device to use in a dental office.
- The sensor should be maintained motionless and in constant contact with the tooth for accurate readings.
- The laser beam must interact with the moving cells within the pulpal vasculature.
- It is generally agreed that LDF assessment for human teeth should be performed at 4 weeks following the initial

trauma and repeated at regular intervals until 3 months

- Blood pigments within a discolored tooth crown can also interfere with laser light transmission. Care must be taken to ensure that the false positive results are not obtained from the stimulation of supporting tissues [6].

### Pulse Oximetry

- Pulse Oximetry has recently been adapted for use in dentistry. This technique has been the most commonly used technique for the measurement of oxygen saturation in medicine because of its ease and affordability.
- In 1940, Squire recognized that changes of red and infrared light transmission caused by pneumatic tissue compression permitted saturation to be computed. In 1950, Wood used this idea to compute absolute saturation continuously from the ratios of optical density changes with pressure in an ear oximeter. Takuo Aoyagi, an electrical engineer at Nihon Kohden company in Tokyo, realized that the pulsatile changes of oxygen saturation could be used to compute saturation from the ratio of ratios of pulse changes in the red and infrared. His ideas, equations, and instrument were adapted, improved, and successfully marketed by Minolta about 1978, stimulating other firms to further improve and market pulse oximeters worldwide in the mid- 1980s [4].
- This is an oxygen saturation monitoring device widely used in medical practice for recording blood oxygen saturation levels during the administration of intravenous anaesthesia. It was invented by Aoyagi in the early 1970s [21].
- Pulse oximetry is an entirely objective test, requiring no subjective response from the patient. The pulse oximeter sensor consists of two light-emitting diodes, one to transmit red light (640 nm) and the other to transmit infrared light (940 nm), and a photodetector on the opposite side of the vascular bed. The lightemitting diode transmits light through a vascular bed such as the finger or ear. Oxygenated haemoglobin and deoxygenated haemoglobin absorb different amounts of red/infrared light.
- The pulsatile change in the blood volume causes periodic changes in the amount of red/ infrared light absorbed by the vascular bed before reaching the photodetector. The relationship between the pulsatile change in the absorption of red light and the pulsatile change in the absorption of infrared light is analysed by the pulse oximeter to determine the saturation of arterial blood [21]. Earlier studies by Schnettler and Wallace reported a correlation between pulpal and systemic oxygen saturation readings using a modified ear pulse oximeter probe on a tooth [22]. They recommended its use as a definitive pulp vitality tester. Kahan and co-investigators subsequently developed a customized probe, in conjunction with a commercial pulse oximeter, for pulp vitality testing [23] [7].
- Unfortunately, the accuracy of the commercial instrument was disappointing, and was not considered to have predictable diagnostic value. The critical requirement of using pulse oximeter in dentistry is that the sensor should conform to the size, shape, and anatomical contours of teeth. Secondly, the sensor holder should also keep the light-emitting diode sensor and the photoreceptor as parallel as possible to each other so that the photoreceptor sensor receives the light transmitted through the tooth. Moreover, the sensor holder should allow firm placement

of the sensor onto the tooth to obtain accurate measurements.

- Pulse Oximetry uses red and infrared wavelengths in order to transilluminate a tissue and detects absorbance peaks due to pulsatile circulation and uses this information to calculate the pulse rate and oxygen saturation. The technology is based on a modification of Beer-Lambert's law: namely, the absorption of light by a solute is related to its concentration at a given wavelength. Pulse Oximetry also uses the characteristics of hemoglobin in the red and infrared range 'oxy' hemoglobin absorbs more light in the red range than 'deoxy' hemoglobin and vice versa in the infrared range. The tooth being tested is sandwiched between a photoelectric detector and an light emitting diode of red (640- 660 nm) and infrared (940 nm) lights held in a sensor holder. The devices may further be 'reflectance' type or 'transmission' type. The difference is in the type of light incident on the detector. This sensitivity test can be an ideal chair-side screening test [8, 9].

### Dual wavelength spectrometry

- Dual wavelength spectrophotometry (DWLS) is a method independent of a pulsatile circulation. It is a class of studies in the field of dynamic light scattering related to the investigation of the dynamics of particles within very short time intervals.
- Diffusion wave spectroscopy was introduced by W.L. Butler in 1962 for measuring minute absorption changes of highly turbid biological materials *in vivo*. It is a method independent of a pulsatile circulation. The presence of arterioles rather than arteries in the pulp and its rigid encapsulation by surrounding dentine and enamel make it difficult to detect a pulse in the pulp space. This method measures oxygenation changes in the capillary bed rather than in the supply vessels and hence does not depend on a pulsatile blood flow.
- Nissan *et al.* did an *in vitro* study to determine the feasibility of using DWS to identify teeth with pulp chambers that are either empty, filled with fixed pulp tissue or filled with oxygenated blood. Their findings indicated that continuous-wave spectrophotometry may be a useful method for testing pulp vitality.
- Oximetry by spectrophotometer determines the level of oxygen saturation in the pulpal blood supply with a dual-wavelength light source (760 and 850 nm). This approach is applicable in the case of dense media with multiple scattering, which is very important for tissues. DWS uniquely suited for the measurements of the average size of particles and their motion within the turbid macroscopically homogeneous highly scattering media [9].
- The presence of arterioles rather than arteries in the pulp and its rigid encapsulation by surrounding dentine and enamel make it difficult to detect a pulse in the pulp space. This method measures oxygenation changes in the capillary bed rather than in the supply vessels and hence does not depend on a pulsatile blood flow. Pulse oximetry is a method based on DWLS.
- DWLS detects the presence or absence of oxygenated blood at 760 nm and 850nm. The blood volume or concentration channel (760 nm plus 850 nm) is arranged to respond linearly to the increase in light absorption. The oxygenation channel (760 nm minus 850 nm) senses the

oxygenated blood because of the greater absorption at 850 nm as compared to 760 nm. *In vivo* and *in vitro* studies were conducted to differentiate between pulp chambers that were empty, filled with oxygenated blood or fixed pulp tissue. DWLS was able to differentiate with reproducible readings between a pulp chamber of a vital and non-vital tooth *in vivo*. In young children, in cases of avulsed and replanted teeth with open apices, the blood supply is regained within the first 20 days after replantation but nerve supply lags behind [15].

- Repeated spectrophotometric readings taken at the start of the replantation and continuing upto 40 days later revealed an increase in blood oxygenation levels indicating a healing process and that the pulp of the avulsed tooth was recovering. Hence endodontic treatment need not be undertaken. Even though the instrument was not specifically designed for dental use, it was easy to use and can be developed as a pulp tester. A major advantage is that it uses visible light that is filtered and guided to the tooth by fibreoptics. Thus unlike Laser light, added eye protection is unnecessary for the patient and the operator. Still *in vivo* tests of this hypothesis are in progress. Influence of the gingival circulation cannot be ruled out and data on how large a mass of pulp tissue is needed for accurate readings must be determined. The test is noninvasive and yields objective results. The instrument is small, portable, relatively inexpensive and should be suitable for use in a private dental office [9].

### Transmitted Light Photoplethsmography

- This is an optical measurement technique that can be used to detect blood volume changes in the microvascular bed of tissue. The basic form of PPG technology requires only a few opto-electronic components: a light source to illuminate the tissue (e.g., skin or tooth) and a photodetector to measure the small variations in light intensity associated with changes in perfusion in the catchment (study) volume.
- The PPG sensing technology has been substantially improved since its origins in 1937. PPG has been compared with LDF in experiments on skin and was found to be of similar value. PPG has been applied in many different clinical settings, including clinical physiological monitoring, vascular assessment and autonomic function.
- TLP is a non-invasive technique used to monitor pulpal blood flow, and has been successfully applied in animal and human studies [115, 116]. It has been suggested that TLP incurs less signal contamination from the periodontal blood flow than is the case for LDF
- It is proposed that circulatory changes in human dental pulp can also be investigated with the PPG technique. Hemoglobin absorbs certain wavelengths of light, while the remaining light passes through the tooth and is detected by a receptor.
- The heart rate variability is composed of low- and high-frequency fluctuations, which are mediated by the sympathetic and the parasympathetic nervous systems. The baseline and the amplitude of the PPG signal also show fluctuations in the same frequencies. PPG assessments of dental pulp tissue viability have demonstrated pulsatile waveforms synchronous with a finger PPG reference in healthy subjects and the loss of pulsatility in patients with nonvital dental pulp. There was a significant negative correlation between the tooth

PPG signal and subject age in those with healthy teeth <sup>[10]</sup>.

### 133 Xenon Isotope

Radioactive materials for measurement of pulpal blood circulation were previously used in the radio-labelled microsphere injection method. A method utilizing a radiation probe with 133 xenon radioisotope to differentiate between vital and pulpless teeth on the basis of blood supply has been found effective. However, the use of radioactive materials is expensive, restricted on humans, and requires special licencing requirements. To this point the most promising of these experimental methods are those using the measurement of light passing through or deflected from the blood in the pulp <sup>[11]</sup>

### Conclusion

Diagnosis forms the basis of treatment. The vistas of endodontic diagnosis are ever evolving. Equipping one's natural diagnostic instinct with knowledge of contemporary advances would ensure that the clinician chooses the best possible diagnostic tools for his toolkit to help him and his patient along a safer and surer path of endodontic treatment

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