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Research Aids in Conservative Dentistry and Endodontics- Instrumental Methods

Dr. Teena Sheethal Dsouza
Senior Lecturer, Department of Conservative Dentistry & Endodontics,
A.B.Shetty Memorial Institute of Dental Sciences,
Nitte- Deemed to be University

Dr. Raghu Srinivasan
Professor,
Department of Conservative Dentistry & Endodontics,
AECS Maaruti College of Dental Sciences

Dr. Kelvin Peter Pais
Senior lecturer,
Father Mullers Medical College and Hospital

***Abstract:*- Man from time immemorial has been on a quest to better this life and living; From starting fire to keep warm and cook, through the invention of the wheel, to the concepts of vaccination and the off late quest for better health using physical, chemical and biological technology. As Albert Einstein said,**

"If we knew what it was we were doing, it would not be called research, would it?"

Hence, research forms an integral part of survival and advancement of life for man. Yesterday's technology has been replaced either by modifications of existing ones or by all new concept driven technologies. Like other fields, the medical/dental field has seen a drastic change in life savings research.

Keywords: Research, dental materials, conservative dentistry and endodontics

INTRODUCTION

It is very well said that research begins from home. With research, there is control and autonomy over what is learnt. The new horizons of a subject can be confirmed, clarified, pursued or even discovered. It is the systematic investigation into the study of materials and sources in order to establish facts and reach new conclusions[1].

With the introduction of several materials and equipments, the field of conservative dentistry and endodontics has been on a swift move. However, its translation into clinical practice is not being substantiated enough with clinical research and appropriate aids.

The subject has seen advances, but extrapolation of results and techniques for use has been of question. Hence research aids help in bringing knowledge and newer designs and techniques to better the profession and the professional. With this, research with quality is accessible to all through publications and thus bring about change in concept and ideologies. Research aids form the basis of the subject and thus will influence the future role of any material, technique or treatment[2]. This review article summarizes the various instrumental research aids for chemical analysis used in the field of conservative dentistry and endodontics.

PART 1: INSTRUMENTAL METHODS OF CHEMCAL ANALYSIS

X-RAY DIFFRACTION ANALYSIS:

Max Von Laue in 1912 observed the diffraction of X-rays, which marked an significant event in the realm of science. Since then, X-ray diffraction (XRD) studies have unveiled a great deal of valuable information about many ordered atomic structures seen in a plethora of day to day materials[3].

About 95% of solid objects encountered in daily life are crystalline in nature. When x-rays interact with a crystalline substance, one observes a particular pattern that can be described as a diffraction pattern. The spacing of planes of chemical substances to form crystals is not similar or identical in all analogous directions; hence a unique result is obtained for each substance[4].

Diffraction apparatus:

The X-ray diffraction apparatus is somewhat similar to an optical grating spectrometer. When collimation is expected in one plane only, the X-ray tube emits a beam that passes through a bundle of metal tubes or through a series of narrow slits.

The photographic and electric detection of the diffracted X-ray beam is by means of the ionization produced in a gas, by scintillation counting, or by the photoelectric effect produced in the semiconductor elements germanium or silicon[4].

Applications in dentistry:

- Lehman and Isard, 1969: X ray diffraction method was used for the detection of crystalline and other constituents of dental porcelain[5]
- Cai and Watanabe, 2001: X-ray diffractometer was employed to study dental alloy-ceramic interfaces[6].
- Amal and Carlos, 2006: X-ray diffractometer (Rigaku) was used to identify the structural and physical characterization of the unknown composition of resilon composite[7].
- Islam and Chng, 2006: powder X-ray diffractometry was used to compare the primary

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ingredients which were present in ProRoot MTA, ordinary Portland cement and white Portland cement[8].

MAGNETIC RESONANCE SPECTROSCOPY

Nuclear magnetic resonance (NMR) is a noninvasive method that yields a subatomic picture of molecular chemical, physical and transport processes[9]. It measures the absorption of electromagnetic radiation in the radiofrequency region of roughly 4MHz to 750MHz, which corresponds to a wavelength of about 75m to 0.4m.

Its applications include polymer analysis, pharmaceutical determinations, invivo studies of phosphorous metabolism and magnetic resonance imaging[10].

Principle:

Here, a sample or material is subjected to 2 magnetic fields simultaneously, one that is stationary H, and the other varying at some radiofrequency f=4MHz or higher. The interaction between matter and electromagnetic forces that is detected and absorbed, can be observed as a change on the signal developed by a radiofrequency detector and amplifier[4].

Dental applications of NMR:

Magnetic resonance spectroscopy has proved its reliability in the study of dental materials.

i. Pires and Nunes, 2001: The authors proved its reliability in the study of the photopolymerization of resin-based dental materials[11].
ii. Pires et al, 2004: The authors subjected a commercial glass ionomer (Fuji II) to different NMR techniques. The curing kinetics of the cement were observed that revealed two distinct phases: the gelation and maturation phases[12].
iii. Grande and Plotino (2006) using NMR, demonstrated the chelating effects of EDTA and flushing action of sodium hypochlorite. They concluded that a final flush with sodium hypochlorite cannot curb the chelating effects of EDTA in a clinical scenario[13].
iv. Nowicki and Sem (2011) used NMR to determine the chemical constitution of the reaction precipitate between sodium hypochlorite (NaOCl) and chlorhexidine (CHX) and also the relative molecular weight of the components[14].

Advantages:

- NMR is non-destructive, non-sample consuming and is insensitive to sample opacity.
- NMR spectroscopy techniques provide detailed metabolic information by analysis of cell extracts, supernatants, biological fluids and live biological samples.
- NMR provide direct, time-resolved monitoring of metabolite concentrations.

Disadvantages:

- Its inherent low sensitivity due to the low energies involved
- lower concentration detection thresholds
- increased cost.
- technique sensitivity[9].

GAS CHROMATOGRAPHY

The pioneers of the development of gas chromatography were Martin and Synge, who in 1941 demonstrated the gas-liquid partition chromatograms for analytical uses. It works on the principle that by which a moving gas phase passing over a stationary sorbent separates a mixture into its constituents[10].

Advantages:

- Fast analysis
- High efficiency – leading high resolution
- Non destructive- enabling coupling to mass spectrometres- an instrument that measures the masses of individual molecules that have been converted into ions i.e, molecules that have been electrically charged
- High quantitative accuracy[15].

Disadvantages:

- Limited to volatile samples
- Not suitable for samples that degrade at elevated temperatures (thermally labile)
- Requires MS detector for analyte structural elucidation[15].

Applications in dentistry

i. Manabe et al, 2000: recommended GC-MS as a reliable methodology for detecting the trace amounts of xenoestrogenic substance bisphenol A in dental materials[16].
ii. Ortengren et al, 2004: used GC-MS to assess the influence of pH and time on the degradation and elution of organic substances from the composite resin material, Z-100[17].
iii. Basrani et al, 2010: used gas chromatography to determine 4-chloroaniline and its derivatives formed in the interaction of sodium hypochlorite and chlorhexidine[18].

ATOMIC ABSORPTION SPECTROSCOPY:

Atomic absorption spectroscopy is a sensitive technique for determining the elemental composition of an analyte by its electromagnetic or mass spectrum. The technique makes use of the wavelengths of light specifically absorbed by an element. They correspond to the energies needed to promote electrons from one energy level to another higher energy level.

Principle:

The principle is that free atoms (gas) generated in an atomizer can absorb radiation at a specific frequency. Atomic absorption spectroscopy quanifies the absorption of ground state atoms in the gaseous state. The atoms absorb ultraviolet or visible light and make transitions to higher electronic energy levels.

The analyte concentration is determined from the amount of absorption. Concentration measurements are usually determined from a working curve after calibrating the instrument with standards of known concentration[19] .

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Selected studies using atomic absorption spectroscopy:

i. Wataha and Malcolm, 1995: used atomic absorption spectroscopy to determine the release of cytotoxic elements of high noble, noble and silver based dental casting alloys[20].
ii. Spano et al, 2009: evaluated the concentration of calcium ions and smear layer removal by using root canal chelators using atomic absorption spectrometry[21].
iii. Thangaraj and Ballal: used atomic absorption spectroscopy to determine the Calcium loss and its effect on microhardness of the root canal dentin following treatment with aqueous solution of 17% ethylenediaminetetraacetic acid[22].
iv. Taneja and Kumari, 2014: to assess the effect of different chelating agents on the calcium loss and its subsequent effect on the microhardness of the root dentin[23].

INFRARED AND RAMAN SPECTROSCOPY:

Since 1905, when William W. Coblentz obtained the first infrared spectrum, vibrational spectroscopy has become an important analytical tool in research and in technical fields[19].

Raman spectral analysis is often compared with the well-known infrared absorption (IR) spectroscopy. While the IR technique measures the light absorption by specific molecules using a broadband light source, the Raman technique measures the characteristic Raman emission induced from molecules under monochromatic laser irradiation. Frequency differences between laser light and Raman emission signals correspond to vibrational frequencies of the molecules. Some vibrational bands observable by the IR absorption technique also appear in the Raman spectrum. Other bands, however, are unique to either one of the two techniques because of the difference in the selection rules governing the two different processes (light absorption and Raman emission). When both techniques are combined, more bands can be identified; therefore, the IR and Raman techniques are often regarded to be complementary[24].

Advantages of Raman spectroscopy over IR spectroscopy:

i. Raman signals are emitted in the form of light scattering and can be observed from all directions, unlike the co-linear optical arrangement found in IR.
ii. The axes of excitation light and detection can be chosen independently, resulting in a considerable instrumental flexibility in the Raman technique.
iii. An optical microscope can be incorporated easily into a Raman spectroscopic system: Specimens can be scanned with a lateral resolution (~ 1 mm), or very small volumes of specimens can be examined.
iv. Polarization characteristics of the Raman signals from crystalline minerals can provide additional spectral information.
v. Simple non-destructive sample preparation,
vi. Linear response to chemical concentrations, and
vii. Easier spectral/band analysis compared with the IR absorption technique.

Disadvantages:

i. A problem of fluorescence exhibited by most biological materials when irradiated by laser light
ii. Fluorescence spectra due to organic materials often dominate the much weaker Raman signals. Raman spectroscopy based on near infrared Fourier transform (NIR-FTR) spectrometers has become popular in the medical and dental fields, because it offers solutions to the fluorescence problems and operational complexities.
iii. Increased cost[25]

Dental applications:

- Van Meerbeek et al, 1993: used the Raman technique to analyze dental adhesive resins[26]
- Alex et al, 2005: presented a method that combined optical coherence tomography (OCT) and Raman spectroscopy to provide morphological information and biochemical specificity for detecting and characterizing incipient carious lesions found in extracted human teeth[27]

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