Available online at http://www.journalcra.com

International Journal of Current Research
Vol. 8, Issue, 09, pp.39390-39394, September, 2016

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

ISSN: 0975-833X

RESEARCH ARTICLE

NONINVASIVE PROCEDURES FOR DIAGNOSING ORAL SQUAMOUS CELL CARCINOMA - CURRENT CONCEPTS AND FUTURE

[1]Dr. Asha, M. L., [*,2]Dr. Aprajita Dua, [3]Dr. Chaitanya Reddy, [4]Dr. Mahesh Kumar H. M. and [5]Dr. Basetty Neelakantam Rajarathnam

[1]Department of Oral Medicine and Radiology, Dr. Syamala Reddy Dental College, Hospital and Research Centre, Bangalore, MDS
[2]Department of Oral Medicine and Radiology, Dr. Syamala Reddy Dental College, Hospital and Research Centre, #111/1, SGR College Main Road, Munnekolala, Marathahalli (Post), Bangalore- 560037, (MDS)
[3]Department of Public Health Dentistry, Dr. Syamala Reddy Dental College, Hospital and Research Centre, Bangalore, MDS
[4]Department of Oral Medicine and Radiology, Dr. Syamala Reddy Dental College, Hospital and Research Centre, Bangalore, MDS
[5]Department of Oral Medicine and Radiology, Dr. Syamala Reddy Dental College, Hospital and Research Centre, Bangalore, MDS

ARTICLE INFO

Article History:
Received 05th June, 2016
Received in revised form
23rd July, 2016
Accepted 18th August, 2016
Published online 30th September, 2016

Key words:

Oral Squamous Cell Carcinoma,
Non Invasive Diagnostic Aids.

ABSTRACT

Oral squamous cell carcinoma (OSCC) has a remarkably high incidence worldwide, and a fairly serious prognosis, encouraging further research into advanced technologies for noninvasive methods of making early diagnosis, ideally in primary care settings. Clinical trials and other information published till the recent years has been studied and a review of noninvasive methods of diagnosing OSCC, including oral brush biopsy, optical biopsy, saliva-based oral cancer diagnosis, and others were included. It is clear that screening for and early detection of cancer and pre-cancerous lesions have the potential to reduce the morbidity and mortality of this disease. Advances in technologies for saliva-based oral diagnosis and optical biopsy are promising pathways for the future development of more effective noninvasive methods for diagnosing OSCC that are easy to perform clinically in primary care settings. Minimally invasive interventions are critical to improving healthcare efficiency, enhancing the quality of care provided, and reducing cost. The trend is toward facilitating the making of early diagnoses of OSCC by General Practitioners or dentists possible in primary care settings. This review thus provide an insight into the current concepts and future of noninvasive procedures for diagnosing oral squamous cell carcinoma.

Citation: Dr. Asha, M. L., Dr. Aprajita Dua, Dr. Chaitanya Reddy, Dr. Mahesh Kumar H. M. and Dr. Basetty Neelakantam Rajarathnam, 2016. "Noninvasive procedures for diagnosing oral Squamous cell carcinoma - Current concepts and future", *International Journal of Current Research*, 8, (09), 39390-39394.

INTRODUCTION

Cancers of the oral cavity, oropharynx, hypopharynx, pharynx, and larynx, are included in head and neck squamous cell carcinoma (HNSCC), which being the sixth most common cancer worldwide. (Saman, 2012) constitutes about 5% of all cancers globally, with about 0.7 million new cases being diagnosed annually in India. (Reddy *et al*., 2014) The highest incidence rates occur in three developing countries (Pakistan, Brazil and India) and one developed country (France) (Ford and Farah, 2013). Accounting for 96% of all oral cancers, squamous cell carcinoma (SCC) is usually preceded by dysplasia presenting as white epithelial lesions on the oral mucosa (leukoplakia). Leukoplakias develop in 1–4% of the population (Regezi *et al*., 2007). Malignant transformation, which is quite unpredictable, develops in 1–40% of leukoplakias over 5 years (Regezi *et al*., 2007). Dysplastic lesions in the form of erythroplakia (red lesions) carry a 90% risk of malignant conversion (Regezi *et al*., 2007). Tumour detection is further complicated by a tendency towards field cancerisation, leading to multicentric lesions, all of which may not be clinically visible (Acha *et al*., 2005). These benign lesions are often biopsied surgically; in most cases multiple follow-up biopsies are indicated. The following disadvantages

Corresponding author: Aprajita Dua,
Department of Oral Medicine and Radiology, Dr. Syamala Reddy Dental College, Hospital and Research Centre, #111/1, SGR College Main Road, Munnekolala, Marathahalli (Post), Bangalore- 560037, (MDS)

of surgical biopsies can discourage patients from agreeing to further diagnostic biopsies: (i) fear and stress; (ii) pain and damage to healthy tissue; (iii) risk of infection; (iv) temporary disability and discomfort; and (v) cosmetic concerns. This warrants the need for noninvasive methods for diagnosing OSCC are summarised in List 1. (Omar, 2015) Despite significant advances in cancer treatment, early detection of cancer and its curable precursors remains the best way of optimising patient survival and quality of life. Advancements in diagnosis and detection for oral cancer have been listed in List 2. (Renuka Verma *et al*., 2015)

Light induced fluorescence spectroscopy

When cells interact with light they become excited and re-emit light of varying colours (fluorescence) and this can be detected by sensitive spectrometers. All tissues fluoresce due to the presence of fluorescent chromophores (fluorophores) within them. FS can detect these substances and provide characteristic spectra that reflect biochemical changes occurring within the tissue. The resultant spectra not only detect the fluorescence but also are sensitive to the cellular components that absorb light, eg. haemoglobin. (Sharwani *et al*., 2006) The commonly detected fluorophores include NADH, collagen, elastin and co-factors such as flavins (FAD, FMN) (Bigio and Bown, 2004). The fluorescence can either occur as autofluorescence (induced by UV light), or as a laser-induced phenomenon and may also be enhanced by either topical or systemic application of 5-aminolaevulinic acid (5-ALA) and can be used for single-point or imaging measurements. 5-ALA is a precursor of the fluorescent photosensitiser, protoporphyrin IX (PpIX), and can be administered systemically or applied topically to the oral mucosa and facial skin. (Sharwani *et al*., 2006) A schematic diagram representing the light induced fluorescent spectroscope is shown in Figure 1.

Clinical Methods Vital staining – Toluidine Blue/Lugol's Iodine Chemiluminescence: vizilite Oral CDx **Visualization Adjuncts Tissue Auto fluorescence** VELscope In Vivo Confocal Microscopy **Photo diagnosis** Auto fluorescence Spectroscopy Fluorescence Photography Narrow band imaging	**Saliva-based oral cancer diagnostics** Molecular Methods DNA Ploidy & Quantification of nuclear DNA content Tumor Markers & Bio Markers PCR Based diagnostic aids

List 1: Summary of the most common non-invasive methods for diagnosing oral squamous cell carcinoma

SPECTROSCOPY	IMAGING TECHNIQUES
▸ Light-induced fluorescence spectroscopy ▸ Elastic scattering spectroscopy ▸ Orthogonal polarization spectral (OPS) imaging	➢ Photoacoustic imaging ➢ 2nd harmonic generation ➢ Teraherlz imaging

List 2: Advancements in diagnosis and detection for oral cancer

- Dysplastic and malignant tissues, have a different spectral characteristics, tend to have increased red fluorescence and decreased green fluorescence.
- Significant increase in the red/green fluorescence ratio is an accurate predictor of dysplasia and malignancy. (Sharwani *et al*., 2006)

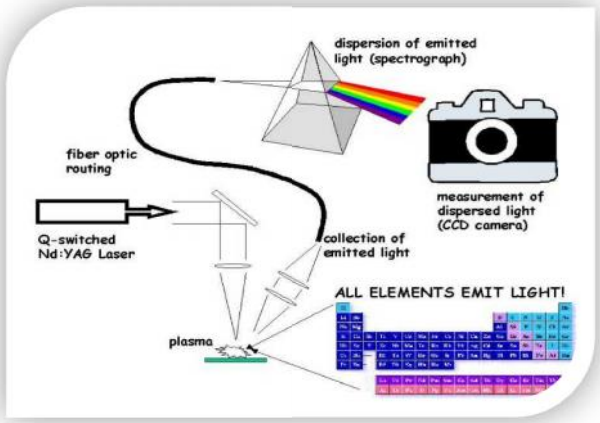

Figure 1. Schematic diagram showing the light induced spectroscopic technique

Advantages:

Noninvasive method with a high level of specificity

Disadvantage:

Low sensitivity has been reported for fluorescence spectroscopy (Sharwani *et al*., 2006)

Elastic Scattering Spectroscopy

Elastic Scattering Spectroscopy (ESS) is an emerging technique that generates a wavelength dependant spectrum that reflects structural and morphological change within tissues. Elastic scattering implies that the light returns with the same kinetic energy as the incident photons. The incident light can undergo single, or more commonly, multiple scattering events before being collected again at the same surface by an optical probe and the resulting data is analysed. The acquired data reflects both the scattering and absorptive properties of that tissue. (Bigio and Mourant, 1997) This scattering process has been shown to occur at gradients in the optical index of refraction resulting from differences in densities that occur at a cellular and sub-cellular level. The structures that induce the scattering (collectively known as scattering centres) are the nucleus, chromatin concentration, and sub-cellular organelles; other scattering centres include structural proteins, lipids and erythrocytes. ESS has been shown to be sensitive to nuclear size, chromatin content and nuclear/cytoplasmic ratio which are all criteria that the histopathologist looks for when establishing malignancy within a tissue (Mourant *et al*., 1998; Perelman *et al*., 1997; Wallace and Van Dam, 2000; Mourant *et al*., 2000; Drezek, 2003). It has an advantage of being simple non invasive method of tissue interrogation. ESS has been shown to be sensitive to nuclear size, chromatin content and nuclear/cytoplasmic ratio which are all criteria that the histopathologist looks for when establishing malignancy within

a tissue. (Sharwani *et al*., 2006) In one study, sensitivity of 72.7% and specificity of 75% were obtained after assessment of clinically suspicious oral lesions in 25 patients, uaing ESS and the spectra could be combined as a result of analysing different anatomical sites. (Sharwani *et al*., 2006) Assessment of nodal metastases using ESS had a sensitivity of 98% and specificity of 68%, but false positives were found in 40.5%. (Jerjes *et al*., 2004)

Orthogonal polarization spectral (OPS) imaging

Quantitative and qualitative analysis of tumor vasculature is of paramount importance for the assessment of microvascular function and for the development of novel anti-vascular strategies. (Pahernik *et al*., 2002) In vivo and in vitro studies have shown that tumor growth is dependent upon neovascularization. (Folkman, 1990; Folkman, 1995; Srivastava *et al*., 1986; Weidner, 1993) In order for this process to take place, tumors produce growth factors which stimulate the proliferation, migration and differentiation of endothelial cells. (Petruzzelli, 1996) In several tumor systems, the degree of angiogenesis can be correlated with tumor aggressiveness and clinical outcome. Moreover, targeting the angiogenic pathway of tumors could be an important development for more effective treatment strategies.(O'Reilly *et al*., 1996) Recently a new in vivo microscopic technique, called orthogonal polarization spectral (OPS) imaging, has been introduced to the clinical observation of the human microcirculation allowing first time observations of a large number of microcirculatory properties in human health and disease. (De Backer *et al*., 2002; Groner *et al*., 1999; Lindeboom *et al*., 2005; Mathura *et al*., 2001; Pennings *et al*., 2004; Spronk *et al*., 2002) Polarized light (550 ± 70 nm) traveling through a light guide and passing through a series of lenses is absorbed by hemoglobin (Hb) in erythrocytes and thus projecting an image of circulating dark bodies flowing through tissue vasculature. The light wavelength of 550 ± 70 nm is the isobestic point for Hb. Through the use of this spectroscopic method, OPS imaging technology produces high contrast images of the microcirculation.

Advantages

- This patented "virtual backlighting" technology makes it possible to visualize and measure real time images of the microcirculation, without the use of fluorescent dyes or transillumination.
- In vivo visualization of human microcirculation makes it possible to acquire high resolution images of the oral mucosa.

Megacapillaries and bleeding can be visualized in oral squamous cell image in Figure 2. The dark background in an OPS image of oral squamous carcinoma. The dark background in an OPS image of oral squamous carcinoma in Figure 3. (Lindeboom *et al*., 2006)

Photo acoustic imaging

Biomedical photoacoustic (PA) imaging provides a means of visualizing the internal structure and function of soft tissues. The image contrast is provided by light-absorbing chromophores within the tissue. These may be naturally occuring, such as oxyhemoglobin and deoxyhemoglobin, or externally administered, e.g., dyes, nanoparticles, or other contrast agents. Figure 4 shows a schematic representation of its operation. (Cox *et al*., 2006) The combined ultrasound and photoacoustic imaging together with molecular specific gold nanoparticles can be used to detect cancer at an asymptomatic stage and can also be used to accurately plan and guide the photothermal therapeutic procedures and monitor the outcome. (Mallidi *et al*., 2009)

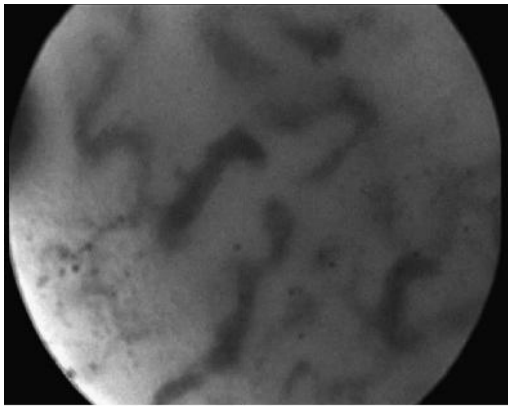

Figure 2. Megacapillaries and bleeding in oral squamous cell image

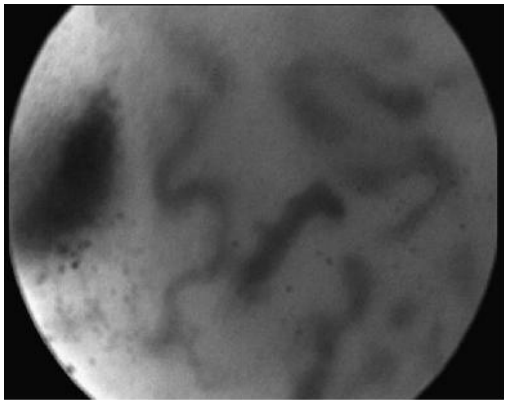

Figure 3. Notice the dark background in an OPS image of oral squamous carcinoma

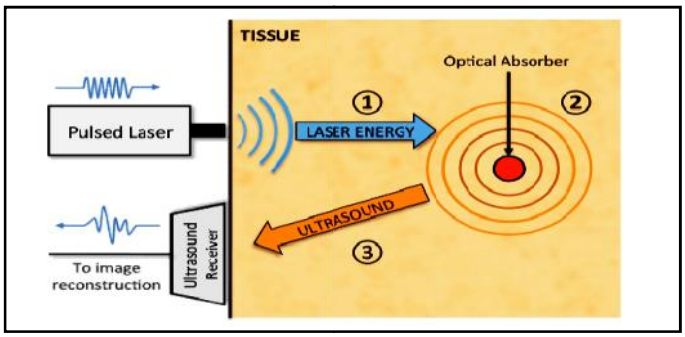

Figure 4. Operational function of photoacoustic imaging

Advantages

- Noninvasive diagnostic procedure
- Penetration depth achievable is more, in cms in contrast to other optical imaging modalities with a penetration of a few mms.

Disadvantage

- Because the absorbed energy density is the product of both the absorption coefficient and the fluence, this image has the disadvantage that it provides a somewhat indirect representation of the spatial variation of the optical coefficients and therefore the structure and physiology of the tissue. (Cox *et al*., 2006)

2ND harmonic generation

The content and structure of collagen is essential in governing the delivery of therapeutic molecules in tumors. Thus, simple histological staining of tumor tissue biopsies for collagen could be used to assess the accessibility of molecular therapeutics in tumors. it is possible to optically image fibrillar collagen in tumors growing using second-harmonic generation (SHG).

Advantages

- Using this noninvasive technique, one can estimate relative diffusive hindrance, quantify the dynamics of collagen modification after pharmacologic intervention and provide mechanistic insight into improved diffusive transport
- This technology could offer basic scientists and clinicians an enhanced ability to estimate the relative penetrabilities of molecular therapeutics. (Scully *et al*., 2008)

Terahertz imaging

Terahertz (THz) imaging, which utilizes the electromagnetic radiation spectrum between 0.1 and 10 THz, has been investigated to assess its potential to diagnose cancers by measuring the water content change and cell deformation of malignant tumors or by sensing the nanoparticle probes targeted at cancerous tumors. (Pickwell *et al*., 2005)

Conclusion

Minimally invasive interventions are critical to improving healthcare efficiency, enhancing the quality of care provided, and reducing cost. The trend is toward facilitating the making of early diagnosis of OSCC by General Practitioners or dentists possible in primary care settings. These noninvasive methods for diagnosing OSCC that are easy to perform clinically in primary care settings can prove as a boon to future oral cancer diagnosis. (Omar, 2015)

REFERENCES

Acha A, Ruesga MT, Rodriguez MJ, Pancorbo MA, Aguirre JM. 2005. Applications of the oral scraped (exfoliative) cytology in oral cancer and precancer. *Med Oral Patol Oral Cir Bucal*., 2:95–102.

Bigio I.J., S.G. Bown, 2004. Spectroscopic sensing of cancer and cancer therapy: current status of translational research, Cancer Biol. Ther. 3 (3) , 259–267, Epub March 16, 2004.

Bigio IJ, Mourant JR. 1997. Ultraviolet and visible spectroscopies for tissue diagnostics: fluorescence spectroscopy and elastic-scattering spectroscopy. *Phys Med Biol*., 42(5):803–14.

Cox BT, Arridge SR, Kostli KP, Beard PC. 2006. Two-dimensional quantitative photoacoustic image reconstruction of absorption distributions in scattering media by use of a simple iterative method. *ApplOpt*., 45:1866-1875.

De Backer D, Creteur J, Preiser JC, Dubois MJ, Vincent JL. 2002. Microvascular blood flow is altered in patients with sepsis. *Am J Respir Crit Care Med*., 166:98–104.

Drezek RA, Richards-Kortum R, Brewer MA, Feld MS, Pitris C, Ferenczy A, *et al*. 2003. Optical imaging of the cervix. *Cancer*, 98(Suppl. 9):2015–27.

Folkman J. 1995. Angiogenesis in cancer, vascular, rheumatoid and other disease. *Nat Med*., 1:27–31.

Folkman J. 1990. What is the evidence that tumors are angiogenesis dependent. *J Natl Cancer Inst*., 82:4–6.

Ford P.G, Farah CS. 2013. Early detection and diagnosis of oral cancer: strategies for improvement *Journal of Cancer Policy*, 1: 2– 7.

Groner W, Winkelman JW, Harris AG, *et al*. 1999. Orthogonal polarization spectral imaging: a new method for study of the microcirculation. *Nat Med*., 5:1209–12.

Jerjes W, Swinson B, Pickard D, *et al*. 2004. Detection of cervical intranodal metastasis in oral cancer using elastic scattering spectroscopy. *Oral Oncol*., 40:673–8

Lindeboom JA, Mathura KR, Ince C. Orthogonal polarization spectral (OPS) imaging and topographical characteristics of oral squamous cell carcinoma.

Lindeboom JA, Mathura KR, Ince C. 2005. The microcirculation during wound healing after oral and maxillofacial surgical procedures. In: Vincent JL, editor. Yearbook of intensive care and emergency medicine. Berlin Heidelberg: Springer-Verlag, p.519–27.

Mallidi S., T. Larson, J. Tam, P. P. Joshi, A. Karpiouk, K. Sokolov, and S. Emelianov. 2009. Multiwavelength photoacoustic imaging and plasmon resonance coupling of gold nanoparticles for selective detection of cancer. *Nano Lett*., 9:2825.

Mathura KR, Alic L, Ince C. 2001. Initial clinical experience with OPS imaging. In: Vincent JL, editor. Yearbook of intensive care and emergency medicine. Berlin Heidelberg: Springer-Verlag, p. 233–44.

Mathura KR, Bouma GJ, Ince C. 2001. Abnormal microcirculation in brain tumors during surgery. *Lancet*, 17:1698–9.

Mathura KR, Vollebregt KC, Boer K, de Graaf JC, Ubbink DT, Ince C. 2001. Comparison of OPS imaging and conventional capillaroscopy to study human microcirculation. *J Appl Physiol*., 91:74–8.

Mourant JR, Canpolat M, Brocker C, Esponda-Ramos O, Johnson TM, Matanock A, *et al*. 2000. Light scattering from cells: the contribution of the nucleus and the effects of proliferative status. *J Biomed Opt*., 5(2):131–7.

Mourant JR, Hielscher AH, Eick AA, Johnson TM, Freyer JP. 1998. Evidence of intrinsic differences in the light scattering properties of tumorigenic and nontumorigenic cells. *Cancer*, 84(6):366–74.

O'Reilly MS, Holmgren L, Chen C, Folkman J. 1996. Angiostatin induces and sustains dormancy of human primary tumors in mice. *Nat Med*., 2:689–92.

Omar E 2015. Current concepts and future of noninvasive procedures for diagnosing oral squamous cell carcinoma-a systematic review. *Head & Face Medicine,* 11: 6.

Pahernik S, Harris AG, Schmitt-Sody M, Krasnici S, Goetz AE, Dellian M, *et al*. 2002. Orthogonal polarization spectral imaging as a new tool for the assessment of antivascular tumour treatment in vivo: A validation study. *Br J Cancer*, 86:1622–7.

Pennings FA, Bouma GJ, Ince C. 2004. Direct observation of the human cerebral microcirculation during aneurysm surgery reveals increased arteriolar contractility. *Stroke*, 35:1284–8.

Perelman LT, Winn J, Wu J, Dasari RR, Feld MS. Photon migration of near-diffusive photons in turbid media: a Lagrangian-based approach. *J Opt Soc Am A Opt Image Sci.,* Vis 1997;14(1):224–9.

Petruzzelli GJ. 1996. Tumor angiogenesis. Basic science reviews. Head and Neck, 18: 283-91.

Pickwell E, Fitzgerald AJ, Cole BE, Taday PF, Pye RJ, Ha T, Pepper M, Wallace VP. Simulating the response of terahertz radiation to basal cell carcinoma using ex vivo spectroscopy measurements. *Journal of Biomedical Optics*., 2005 Nov 1;10(6):064021.

Reddy BK, Lokesh V, Vidyasagar MS, Shenoy K, Babu KG, Shenoy A, *et al.* 2014. Nimotuzumab provides survival benefit to patients with inoperable advanced squamous cell carcinoma of the head and neck: A randomized, open-label, phase IIb, 5-year study in Indian patients. *Oral Oncol*., 50:498-505.

Regezi JA, Sciubba JJ, Jordan RCK. 2007. Oral pathology: clinical pathologic correlations. 5th ed. St. Louis: Saunders.

Verma R, Chaturvedi M, Riaz A, Verma D, Mittal L, Mittal S. 2015. Diagnostic Adjuncts in Oral Cancer: a Review. *International Journal of Contemporary Pathology*, 1(1):55-60.

Saman DM. 2012. A review of the epidemiology of oral and pharyngeal carcinoma: Update. *Head Neck Oncol*., 4:1.

Scully C, Bagan JV, Hopper C, Epstein JB. 2008. Oral cancer: current and future diagnostic techniques. *Am J Dent*., 21:199–209

Sharwani A, Jerjes W, Salih V, *et al*. 2006. Assessment of oral premalignancy using elastic scattering spectroscopy. *Oral Oncol*., 42:343–9

Sharwani A, Jerjes W, Salih V, MacRobert AJ, El-Maaytah M, Khalil HS, Hopper C. 2006. Fluorescence spectroscopy combined with 5-aminolevulinic acid-induced protoporphyrin IX fluorescence in detecting oral premalignancy. *J Photochem Photobiol B*., 83:27-33.

Spronk PE, Ince C, Gardien MJ, *et al*. 2002. Nitroglycerin in septic shock after intravascular volume resuscitation. *Lancet*, 360:1395–6.

Srivastava A, Laidler P, Davies R, Horgan K, Hughes LE. 1986. The prognostic significance of tumor vascularity in intermediatethickness skin melanoma. *Am J Pathol*., 133:419–23.

Wallace M, Van Dam J. 2000. Enhanced gastrointestinal diagnosis: light-scattering spectroscopy and optical coherence tomography. *Gastrointest Endosc Clin N Am*., 10(1):71–80, VI.

Weidner N. 1993. Tumor angiogenesis: a review of current application in tumor prognostication. *Semin Diagn Pathol*., 10:302–13.

****