

RAMAN SPECTROSCOPY FOR BREAST CANCER DETECTION: A SAMPLE PROCESSING STUDY

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Abstract: Raman spectra and images collected from human breast tissues were examined using 514-, 830- and 1064-nm excitations. Three tissue-processing techniques are commonly used in pathology and spectroscopy. This study investigates the effect of these processes.

Breast cancer affects one in eleven women over aged 50 in Australia [1]. Triple assessment, a combination of breast examination, mammogram and fine-needle biopsy, is the current standard for diagnosis [2]. Alternative diagnostic techniques, including magnetic resonance spectroscopy (MRS) *ex-vivo*, allow the distinction between pathologies with accuracies approaching 100% [3].

Ultimately, we wish to conduct parallel Raman studies with both MRS and pathology. Ideally, sample processing would be common to each of these diagnostic techniques in order to minimise variations. In this current study, we used Raman spectroscopy (RS) to analyse breast biopsy specimens. Samples were stored in phosphate buffer saline (PBS) solution at $-70\text{ }^{\circ}\text{C}$ prior to processing and were examined using three processing techniques: bulk tissue examination, cryosection and paraffin-embedded section. Bulk tissue samples required no chemical processing but were subject to dehydration during long collection times unless hydration is maintained. However, samples prepared by cryosectioning or by paraffin-embedding also present problems. Adjacent sections of the samples, each taken from cryosectioned and paraffin-embedded sections, were stained with hematoxylin and eosin for pathological examination, and for identification of suspect areas of interest for RS.

Fifty breast biopsies were studied using Raman spectroscopy prior to receipt of pathology reports. This was applied to at least two of the three tissue processing techniques using point spectroscopy, mapping and imaging. Raman maps and images, based on the $800\text{--}1800\text{ cm}^{-1}$ region, were constructed to visualise the distribution of DNA, protein and lipid within the tissue. Differences in the observed spectra can be related to the various sample processing. The results from different sample processing techniques showed differences. The future potential of RS for parallel studies with MRS and pathology will be discussed.

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References:

1. See <http://www.aihw.gov.au>, Australian Institute of Healthcare and Welfare, 2004
2. See <http://www.komen.org>, Susan G. Komen Breast Cancer Foundation, 2004
3. Mackinnon, W. B., P. A. Barry, et al. (1997). "Fine-needle biopsy specimens of benign breast lesions distinguished from invasive cancer ex vivo with proton MR spectroscopy." *Radiology* 204(3): 661-6.
4. J. Radford, B. Hernandez, D. Roman, *Histotechniques Manual 2002*, University of Sydney, Sydney, 2002