Infrared Microspectroscopy of Breast Tumor Tissue Sections and Tumor DNA

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The application of infrared (IR) spectroscopy to probe the chemical composition and structure of complex biological systems such as human cells, tissues and body fluids is a rapidly growing field. In order to evaluate the possibilities of IR spectroscopy as a new tool for the diagnosis of breast tumor tissue, we are currently applying two approaches: (i) the analysis of isolated DNA from normal and cancerous breast tissue and (ii) the mapping of thin sections of breast tissue through microscope optics.

IR spectra obtained from nanograms of DNA extracted from tissue that was histologically found to be normal and from tumor cells revealed minor differences in certain spectral regions, which could be of diagnostic value. For the practical use, however, this approach was found to suffer from problems associated with the microheterogeneity of tumor tissue samples.

IR microspectroscopy is known to enable to identify and to map functional groups in tissues by using vibrational modes as an intrinsic contrast mechanism. We have collected IR spectra of unstained human breast tumor tissue slices on an infrared transparent window under a conventional IR microscope equipped with a computer-controlled movable stage. This allowed the automated aquisition of spectra from microregions as small as 20 µm in diameter over a defined two-dimensional grid of the tissue section. Neighbouring slices were mounted on conventional glass slides for standard histological examination and/or immunohistochemical staining. The alternate use of thin sections for infrared analysis and for staining allowed the precise location of tissue structures, which is essential for the correlation of spectroscopic and histological data. For imaging major constituents within breast tissue sections, the intensities of characteristic IR absorption bands were plotted as a function of the x,y position, known as “chemical” imaging. For the identification of more subtle differences in tissue structure and biochemistry pattern recognition techniques, such as cluster analysis and Artifical Neural Network analysis, were applied. Our results demonstrate that the sensitivity of IR microspectroscopy towards changes in cellular biochemistry and variations in breast tissue architecture is high. At the same time, this work also demonstrates the need for collecting spectra with higher spatial resolution at the level of single tumor cells (5-10 µm). In order to achieve this, we have collected infrared spectra by using a microscope equipped with a focal plane array detector (Bruker FALCON^{TM}). In this case, the spatial resolution is determined by the size of each individual detector element, which was 4.1 µm. Moreover, multiple detector elements (4096) enable spectra at each pixel to be collected simultaneously, which drastically reduces the time required to mapp a certain tissue area. The potential of IR microspectroscopic imaging as a non-subjective complement to established histological examination of breast tumor tissue sections will be discussed.