Prediction of long term breast cancer survival using MR metabolomics

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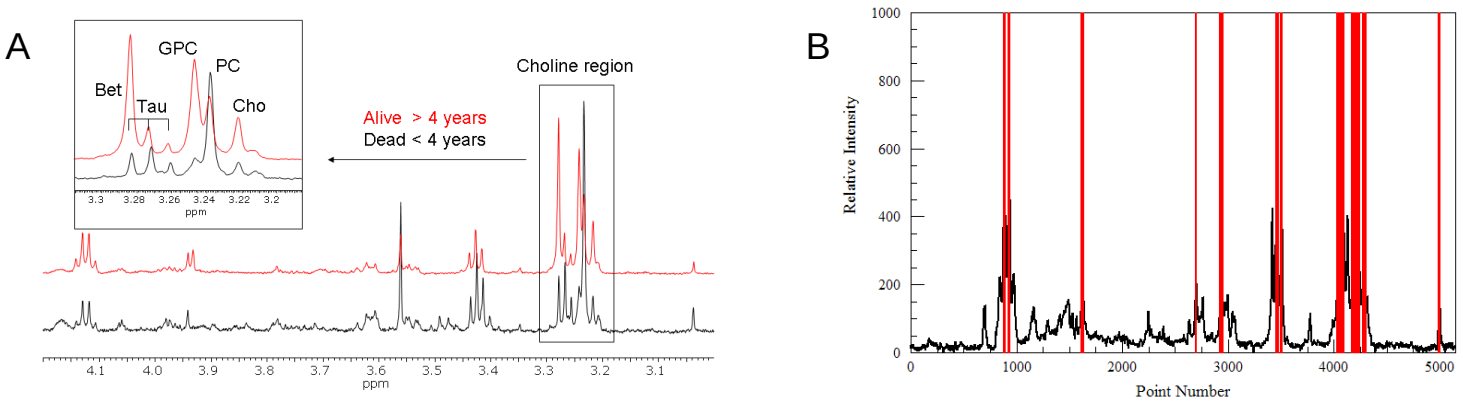
Introduction:

MR metabolomics can be defined as the systematic study of MR visible small-molecules. In tumour tissue, the metabolite profiles express the continuous endpoints of the gene and protein activity. It has been shown that biochemical properties described by magnetic resonance (MR) can characterise breast cancer (1, 2). The biochemical activity in cancer tissue is altered and high resolution magic angle spinning (HR MAS) metabolic profiles of breast cancer tissue correlate to clinical parameters such as lymphatic spread and hormone status (3). The purpose of the current study was to investigate the feasibility of MR metabolomics in prediction of long term breast cancer survival.

Experimental:

Breast cancer tissue was excised from patients with palpable breast cancer (diagnosed as invasive ductal carcinoma (IDC) grade I, II and III), receiving no neo-adjuvant treatment before the surgery. Tissue specimens were analysed in D2O-PBS, and HR MAS MR spectra were recorded on a Bruker Avance DRX600 spectrometer. Proton MR spin echo spectra were acquired with a total echo time of 285 ms and water presaturation (4 °C, spin rate 5 kHz). A pathologist scored the relative areas of normal and neoplastic epithelial elements visually, and spectra from biopsies with tumour-content less than 5% were excluded from further analysis. The survival status 4 years after surgery was known for all patients. This resulted in a dataset consisting of spectra from 40 patients, where 32 patients survived 4 years (represented by 38 spectra), and 8 patients (represented by 8 spectra) passed away before the 4 years limit. The spectral intensities in the 3.0-4.2 ppm region were scaled from 0-1000, and all points with an average value greater than 480 were selected for further analysis of class separation (survival status 4 years after surgery) by Support Vector Machine (SVM, Parameters: normalized polynomial kernel of degree =2; no data normalization; leave-one-out generalization error estimate; sequential minimal optimization algorithm; and C=1000.)



Results:

In Figure A, spectra of biopsies (tumour content 20-30%) from two patients with similar clinical status at the time of surgery but with different outcome regarding survival are compared. Both subjects were in the mid 70ties, diagnosed with IDC grade 3, hormone receptor positive (both ER and PgR) breast cancer with lymphatic spread. Distinct metabolite differences are visible in the spectra, especially in the expanded choline region. The 242 variables selected for SVM analysis, shown graphically in Figure B, include signals from the major metabolites such as cholines, taurine, lactate, glycine and myo-inositol. The actual versus the SVM predicted survival status is shown in Figure C. The black squares denote the actual status, and the red circles the predicted. Wrong predictions were only made for two subjects (number 4 and 43, indicated by arrows).

Figures: (A) Comparison of HR MAS MR spectra from two breast cancer patients with similar clinical status but different survival outcome. (Bet: betaine, Tau: Taurine, GPC: glycerophosphocholine, PC: phosphocholine, Cho: choline) (B) The chemical shifts (in total 242 points) chosen for SVM are indicated by the superimposed red vertical lines on a random spectrum. (C) Actual versus predicted class from SVM. 0: dead < 4 years, 1: alive > 4 years. The two erroneous predictions are indicated by arrows.

Discussion and conclusion:

This preliminary investigation suggests a role of MR metabolomics in prediction of long term breast cancer survival. Early identification of patients with poor prognosis may be important for the administration of more aggressive treatment and closer follow-up of this patient group. The approach is still limited by the sample number size, and further validation including more samples and blind testing are currently being designed in our lab.

References:

(1) Gribbestad et al. NMR Biomed 1994;7:181; (2) Sitter et al. NMR Biomed 2002;15:327; (3) Bathen et al. Breast Cancer Res Treat 2007;104:181