# Choline profiles of breast cancer correlate to clinical tumor characteristics

**B. Sitter[1], T. F. Bathen[1], M. T. Grinde[1], H. Fjøsne[2], J. Halgunset[3], S. Lundgren[4], and I. S. Gribbestad[1]**

[1]Dept. of Circulation and Medical Imaging, NTNU, Trondheim, Norway, [2]Dept. of Surgery, St. Olav University Hospital, Trondheim, Norway, [3]Dept. of Laboratory Medicine, Children's and Women's Health, NTNU, Trondheim, Norway, [4]Dept. of Oncology, St. Olav University Hospital, Trondheim, Norway

## Background

Elevated levels of cholines have been described in brain, breast, prostate, colon, cervical cancers, and metastases using MRS (1). Also, the relative composition of the different choline compounds glycerophosphocholine (GPC), phosphocholine (PC) and choline have been found to change with treatment (2). Breast cancer is a heterogeneous disease, with highly varying tumor biology and treatment response, and the clinical parameters for decision of treatment regime and prediction of outcome should be extended. We wanted to investigate if the choline profiles in tissue from breast cancer patients correlate to clinical parameters related to patient prognosis.

## Experimental

Samples from tumor (n=286) and adjacent non-involved tissue (n=50) from breast cancer patients were analyzed by high resolution magic angle spinning (HR MAS) MRS according to previously described procedures (3). After HR MAS, tissue specimens were examined with histopathology, and only tumor samples with more than 5% tumor cells and confirmed non-involved samples were included. Spectra were analyzed by measuring GPC, PC and choline peak intensities and by partial least squares regression (PLS), and correlated to clinical characteristics of the breast cancer patients; histological grading, lymph node status, tumor size and estrogen receptor status. The spectral region between 3.2-3.3 ppm, including resonances from taurine, GPC, PC and choline was used as input for PLS performed with mean-centering and full cross-validation. The number of PCs to retain in the model was determined by the PC which minimizes residual variance and root mean square error of prediction.

## Results and Discussion

The calculated average spectra of non-involved tissues and invasive ductal carcinomas grade I, II III are shown in Figure 1. Adjacent non-involved tissues showed significantly different ratios PC/Cho and GPC/PC compared to tumor tissues. The different ratios are presented in Table 1. MR derived choline profiles of breast tumor tissue appear to be different with respect to tumor grade (Figure 1). However, there is a large variation within samples of the same grade, and no significant differences could be found. There was a trend of higher GPC/PC (0.83) in tumors from patients with negative lymph node status compared to patients with lymphatic spread (0.68, p=0.097). Choline profiles of estrogen receptor (ER) positive tumors were different from ER negative. PC/Cho were lower in ER negative tumor tissue compared to ER positive (Table 2), and PLS provided classification of ER status. The score plot of PC1 versus PC2 from PLS is shown in Figure 2. ER negative patients cluster in the lower left region of the plot. Six significant PCs were retained in the model, and the MR predicted versus the clinically measured ER status was significantly correlated both for calibration (r=0.67, p<0.01) and validation (r=0.48, p<0.01).

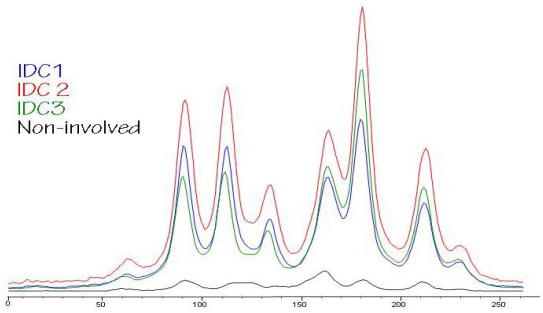


Figure 1. Calculated average spectra of non-involved (n=50), IDC I (n=28), IDC II (n=126) and IDC III (n=114).

Table 1
Mean choline ratios (±SD) for non-involved tissue and invasive ductal carcinomas grade I, II and III.

| | PC/Cho | GPC/Cho | GPC/PC |
|---|---|---|---|
| Non-involved | 0.90 (±0.60) | 1.64(±1.46) | 2.42(±2.98) |
| IDC I | 2.36(±0.95) | 1.38(±0.46) | 0.63(±0.23) |
| IDC II | 3.20(±2.47) | 1.53(±0.83) | 0.67(±0.49) |
| IDC III | 2.71(±1.56) | 1.62(±1.03) | 0.71(±0.41) |

Table 2
Mean choline ratios (±SD) for ER positive and negative tumors.

| | PC/Cho | GPC/Cho | GPC/PC |
|---|---|---|---|
| ER positive | 1.91(±1.47) | 1.46(±1.27) | 0.95(±0.91) |
| ER negative | 2.81(±1.87) | 1.54(±0.85) | 0.77(±0.85) |

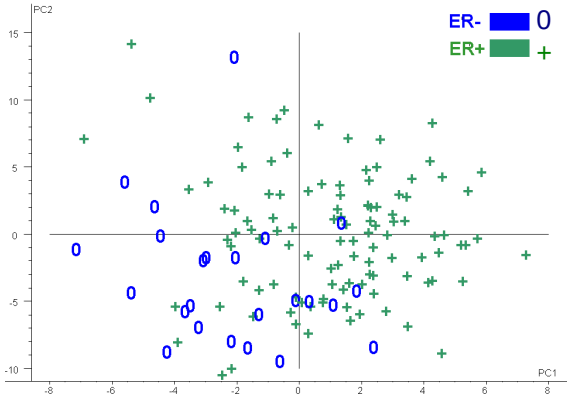


Figure 2. Score plot of the first two principal components from PLS with respect to estrogen components.

## Conclusion

MR spectroscopic profiles of the choline region in breast cancer tissue show a correlation to the hormone receptor status of the patient. The choline profiles display a high level of variance, enlightening the complexity of breast tumor biology and challenges in developing MR spectroscopy as a clinical tool. Further studies using multivariate data analysis are now being investigated.

## References

(1) Sorensen. J Clin Oncol 2006;24:3274; (2) Glunde, Serkova. Pharmacogenomics 2006;7:1109; (3) Sitter et al. NMR Biomed 2006; 19: 30