

Determination of cut-off value of total choline concentration (tCho) for the differentiation of malignant, benign and normal breast tissues by in-vivo proton MR spectroscopy at 1.5 T in a large cohort of women.

R. G. Sah¹, U. Sharma¹, R. Parshad², and N. R. Jagannathan¹

¹Department of NMR & MRI Facility, All India Institute of Medical Sciences, New Delhi, Delhi, India, ²Department of Surgery, All India Institute of Medical Sciences, New Delhi, Delhi, India

Introduction: The differentiation of malignant and benign lesion is vital in the diagnosis of breast cancer for appropriate treatment. Recent in-vivo MRS experiments demonstrated that alterations in the levels of choline-containing metabolites (tCho) are associated with malignant transformation in breast cancer. However, several studies also reported the observation of tCho in benign, normal breast tissues and in lactating breast (1-3). Therefore, a quantitative estimation of the absolute concentration of tCho in normal, malignant and benign breast tissues is necessary for differentiation of various tissues. Earlier we standardized and reported tCho concentration using water as an internal reference (4). The aim of the present study was to determine a cut-off value of tCho concentration for the differentiation of malignant, benign and normal breast tissues using ROC analysis in a large cohort of women.

Material and Methods: 155 women including 101 cytologically proven infiltrating ductal carcinoma (IDC) patients (44.3 ± 12.4 yrs); 25 benign (32.07 ± 8.01 yrs); and 29 normal volunteers (34.35 ± 10.31 yrs) were recruited for this study. Written informed consent was obtained and Institutional ethical committee approved the study. Patients were evaluated clinically and tumor size measured using Vernier calipers. MR investigations were performed using a phased array breast matrix coil at 1.5 T (Siemens Avanto). Following the scout image, T2-weighted coronal images were obtained using standard SE sequence and fat suppressed MR images in the transverse, sagittal and coronal planes. Contrast-enhanced MRI was carried out using a fat-saturated 3D FLASH where-ever indicated for appropriate localization of the lesion. The in-vivo proton MRS using a single voxel PRESS sequence was carried out. Typical water peak line width ranged from 8 to 22 Hz and water+lipid suppression was achieved using MEGA pulse. The experimental parameters used were: TR=1500 ms, TE=100 ms, averages=128, total acquisition time was 3.18 minutes. An additional spectrum of the same voxel without water and lipid suppression was obtained for the concentration calculation using water signal as an internal reference. The tCho concentration was calculated using the modified equation reported by Baik et al for 1.5 T (4-6). Experiments were carried at two time points in 10 patients showed that concentration values determined were within 5%. All statistical analyses were carried out using statistical software SPSS 11.5 and STATA.

Results: Typical MR spectra obtained from a malignant breast tissue of patient with IDC (A); benign breast tissue (B) and from normal breast tissue of a volunteer (C) are shown in Figure-1. tCho was observed in 96/101 malignant, 22/25 benign lesions and 16/29 normal volunteers. The tCho concentration for malignant tumor was 4.0 ± 2.9 mmol/kg (range 0.2-19.8 mmol/kg) was significantly higher compared to benign (1.45 ± 0.92 mmol/kg, range 0.04-2.7) and normal breast tissues (0.57 ± 0.37 mmol/kg, range 0.1-1.3) (Fig.2). The concentration of tCho between volunteers and benign lesions was not significantly different. A cut-off value to discriminate normal, benign and malignant breast tissues was worked out using ROC analysis (Fig-3). Accordingly, a cut-off value of 2.24 mmol/kg for tCho [sensitivity 73.9%, specificity 72.7% with area under curve (AUC) 0.85] was obtained to differentiate malignant from benign diseases. Similarly, a cut-off value of 1.02 mmol/kg for tCho (sensitivity 93.75%; specificity 93.33%; AUC 0.96) was obtained to for differentiation of malignant from normal breast tissues.

Discussion: Many recent in-vivo MRS studies have reported the observation of tCho in some benign lesions, normal tissues of healthy volunteers (1-3) and in lactating breast (1-3). Therefore, it is necessary to determine a cut-off value of tCho concentration for differentiation of various breast

tissues breast tissues. In the present study we calculated the absolute concentration of tCho in a large cohort of women using water as an internal reference and determined cut-off values using ROC analysis. The concentration of tCho in malignant tissues observed in the present study is in agreement with our earlier report (4) and with other studies reported in the literature (5-9). tCho observed in malignant tissues has contributions from free Cho, phosphocholine (PC) and glycerophosphocholine (GPC). However, studies reported high probability of increase in PC content in malignant transformation (10). In our study tCho was observed in 95% of malignant, 88% of benign and 55% of normal volunteers. Further, the wide range of the Cho concentration observed in malignant cases may be due to the heterogeneous nature of breast lesions (11). The non-observation of tCho in five malignant tumors may be attributed to the intrinsic tumor heterogeneity. It was reported that Cho detection may be difficult in diffusive enhancement type cancers because of the intermingling of tumor cells with adipose tissue (8). The observation of tCho in benign as well as in healthy volunteers may be due to biochemical changes like increase in the levels of phosphomonoesters especially during menstrual cycle (1-3). Thus, a cut-off value determined and reported here would help in providing the unambiguous diagnosis of breast lesions which can be used in a clinical setting along with contrast enhanced MRI characteristics.

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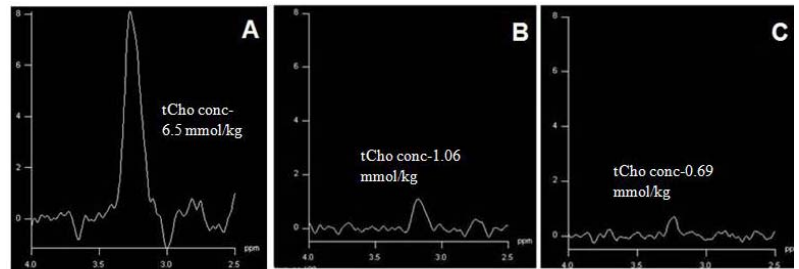


Fig-1-Proton MR spectra from (A) malignant tumor (B) benign and (C) normal breast tissue

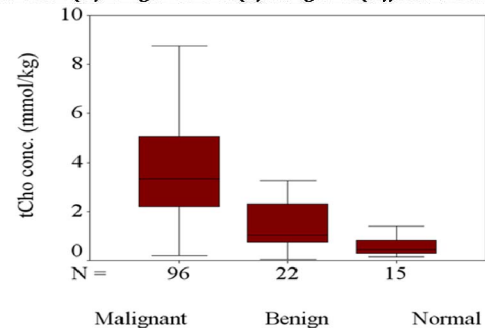


Fig-2: Box plot depicting mean tCho conc (mmol/kg) among malignant, benign and normal breast tissue

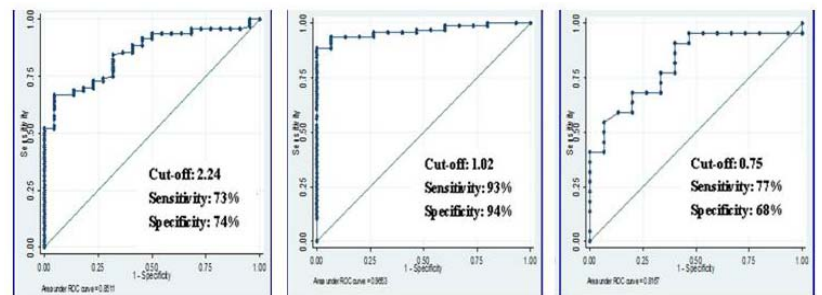


Fig-3: ROC showing the cut-off of tCho conc (mmol/kg) a) between malignant and benign b) between malignant and normal, c) between benign and normal breast tissue