

Choline metabolism in basal-like and luminal-like breast cancer xenografts respond differently to doxorubicin and bevacizumab treatment

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Introduction

MRS examination of breast cancer patients before and after neoadjuvant therapy have suggested that changes in total choline concentration are associated with therapeutic outcome, and that MRS may be useful in therapy monitoring of breast cancer patients¹. However, the response of different subtypes of breast cancer to chemotherapy is not known in detail. MRS in breast cancer treatment monitoring would benefit from a better understanding of how individual choline metabolites respond to treatment in various subtypes of breast cancer. In this study, we have measured changes in the choline metabolic patterns of xenograft models representing basal-like and luminal-like breast cancer subtypes following treatment with bevacizumab and/or doxorubicin.

Methods

Basal-like and luminal-like orthotopic breast cancer xenograft model (MAS98.12 and MAS98.06, respectively) have previously been established and characterised^{2,3}. Animals were treated with bevacizumab (5 mg/kg ip injection at days 1, 4 and 7 after study start), doxorubicin (8 mg/kg single iv injection) or a combination of these treatments. Tumor tissue was harvested 3 days and 10 days after treatment. Untreated animals were used for controls at both sacrifice time points (n=3 in each group, totally 48 animals). The data from untreated controls at day 3 and 10 were pooled.

Tissue samples (14 ± 4 mg) were analysed using HR-MAS MRS performed on a Bruker AVANCE DRX 600 spectrometer (Bruker BioSpin, Karlsruhe, Germany). Single-pulse spectra were obtained according to previously described procedures⁴ including ERETIC for quantification. Peak areas were calculated using peak-fitting software (PeakFit v 4.12 by SeaSolve, USA) and selected metabolites were quantified. The metabolite concentrations were compared using a two-sided unpaired t-test with significance level $p < 0.05$.

Results

Significant reduction of tumor growth rate was seen in all treatment groups compared to untreated control animals ($p < 0.05$) in both xenograft models. The tumor growth rate was reduced more by doxorubicin and combination treatment groups than by the bevacizumab treatment. In the basal-like xenografts, all treatments gave increase in phosphocholine (PCho) and decrease in glycerophosphocholine (GPC) concentrations, with a statistically significant change in GPC/PCho ratio in all treatment groups both at day 3 and day 10. No significant change in total choline concentration (tCho) was observed in any treatment group. In the luminal-like xenografts, there was an initial decrease in PCho concentration in the combination treatment group at day 3 followed by increase in both PCho, GPC and tCho concentrations at day 10 ($p < 0.05$).

Discussion

In this study, tumor growth rates in two different subtypes of breast cancer were similarly affected by treatment with bevacizumab and/or doxorubicin. However, with respect to choline metabolite concentrations, the two subtypes showed differences in their response to therapy.

Previous studies have shown that chemotherapy may cause changes in choline metabolism in breast cancer, with reduction in PCho and/or tCho concentrations as the most prominent response to chemotherapy^{5,6}.

In basal-like xenografts, tCho remained unchanged in all treatment groups at both time points. However, the contribution of the individual choline metabolites was changed, as seen by the reduction in GPC/PCho ratio.

The response in basal-like xenografts differs from that reported in MCF-7 and MDA-mb-231 xenografts, where the GPC/PCho ratio was increased following docetaxel therapy⁵. In luminal-like xenografts, there was an increase in total choline 10 days, but not 3 days, post-treatment. No change in GPC/PCho ratio was observed.

Comparing the changes observed in these basal-like and luminal-like xenografts, it is seen that choline metabolite concentrations in different subtypes of breast cancer may respond differently to the same treatment. Although these data are limited with respect to the tumor models and treatments used, they demonstrate that changes in choline metabolism following chemotherapy may be more complex than previously acknowledged. As changes in choline metabolite concentrations after chemotherapy may depend both on disease model, treatment and time point of monitoring, further studies are required to investigate choline metabolite responses across animal models and treatments.

References

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Figure 1 Changes in tCho (top) and GPC/PCho ratio (bottom) in basal-like (filled labels) and luminal-like (open labels) xenografts following treatment with bevacizumab (▲), doxorubicin (▼) or combination (◆), and in untreated controls (●).

