NMR visible cholesterol content predicts ultimate treatment response in HCT-116 xenografts treated with Flavopiridol, CPT-11 or the combination

J-H. Chen[1], Y. V. Wu[1], R. O'Connor[1], B. U. Laxa[1], P. DeCarolis[1], E. R. Brill[1], and S. Singer[1]
[1]MSKCC, New York, NY, United States

Introduction: Current methods for assessing therapeutic response are largely based on changes in tumor size after multiple cycles of chemotherapy and are often inaccurate and are only detectable late in the course of treatment. The objective of this study was to use an *ex vivo* NMR biomolecular analysis of tumor tissue following a single cycle of chemotherapy to predict ultimate therapeutic effect after several cycles of chemotherapy in a human colon cancer (HCT-116) xenograft model. The hypothesis is that tumor tissue responding to effective treatment disrupts cellular integrity, results in an increase in NMR detectable cholesterol, and serves as an early biochemical marker of treatment response.

Methods: SCID mice were inoculated with 5 million HCT-116 cells on both flanks. Treatment started when the tumor size was approximately 150 mm^3. Mice were treated with either CPT-11 100 mg/kg IP once a week or Flavopiridol 11 mg/kg IP once a week or the combination (CPT-11 100 mg/kg IP followed by Flavopiridol 3 mg/kg IP seven h after) once a week. The tumors from the left flank were harvested at day 4 after the treatment started and were snap frozen at -80°C. Mice were allowed to complete the treatment course for a total of 23 days to determine the final treatment effect. Magic-angle-spinning (MAS) NMR spectra were acquired on approximately 17.5 mg tumor tissue.

Results: The time-dependent tumor size under treatment is shown in Fig. 1a and the NMR detected cholesterol content is shown in Fig. 1b. The HCT-116 tumor responds to the treatment of CPT-11 or the combination of CPT-11/Flavopiridol but only slightly to the Flavopiridol alone. At day 4 following treatment, when there is no statistical difference in tumor size, the NMR detected cholesterol is 3-fold and 2.34-fold higher in the CPT-11 and combination treated tumors than the control tumor (p=0.003, 0.027, n=9), respectively while there is no statistical difference between Flavopiridol treated tumor and control tumor (p=0.310, n=9).

Conclusions: The NMR detected cholesterol measured shortly after the initial drug dose correlates with response and predicts ultimate treatment outcome in this HCT-116 model xenograft prior to any discernable change in tumor size. These results suggest that NMR visible cholesterol serves as an early biomarker of therapeutic response and would provide a more efficient approach for selecting active drug regimes.

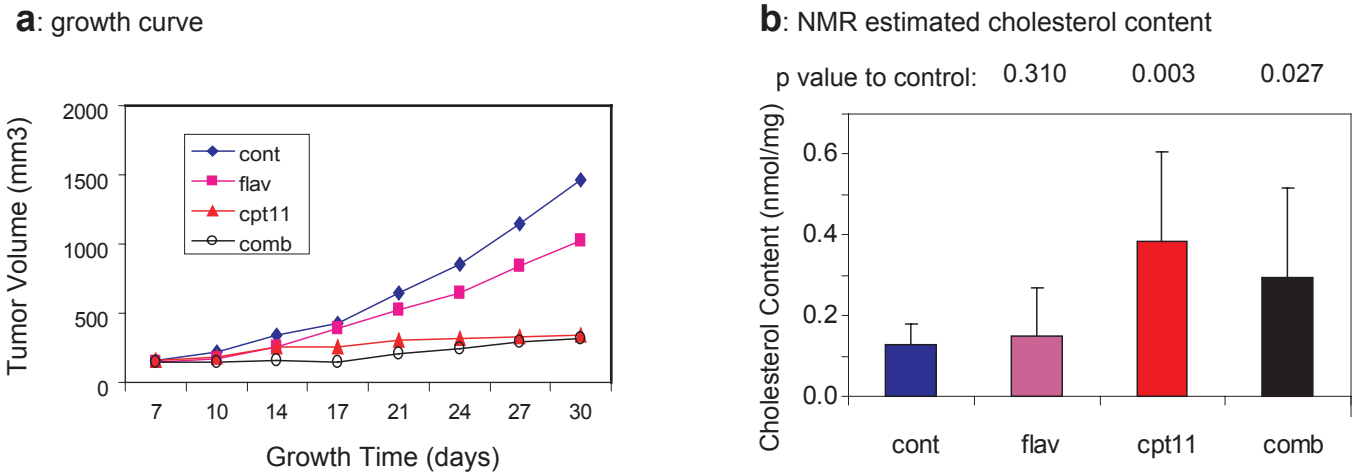

Fig. 1. a). the growth curve of tumor on the right flank under different treatments. b). NMR measured cholesterol content in the tumor resected at day 4 from mouse left flank.

Reference: 1. M. Motwani, C. Jung, F. M. Sirotnak, Y. She, M. A. Shah, M. Gonen and G. K. Schwartz. Augmentation of Apoptosis and Tumor Regression by Flavopiridol in the Presence of CPT-11 in Hct116 Colon Cancer Monolayers and Xenografts. Clin. Canc. Res. 2001, 7, 4209-4219. 2.

2. Chen JH, Singer S. High-Resolution Magic Angle Spinning NMR Spectroscopy. In: Lindon JC, Nicholson JK, Holmes E, editor. The Handbook of Metabonomics and Metabolomics. New York: Elsevier; 2007:p113-148