^{1}H MRS of Human Stool: A Simple, Non-invasive Approach for Diagnosing Colorectal Cancer?

T. Bezabeh[1], B. Levin[2], C. Johnson[2], and I. C. P. Smith[1]

[1]Institute for Biodiagnostics, National Research Council, Winnipeg, Canada; [2] Univ. of Texas M.D. Anderson Cancer Center, Houston, Texas, U.S.A.

Introduction

Colorectal cancer is one of the most common cancers in the western world. The lifetime risk that an individual in North America will develop colorectal cancer is about 5-6%. The mortality of colorectal cancer is relatively high. The clinical features associated with colorectal cancer include blood in the stool, anemia, abdominal pain and alteration of bowel habits. However, such symptoms become apparent only when the disease has advanced significantly. Prognosis for the patient depends largely on the stage of the disease and thus, a simple, non-invasive way of detecting the cancer at an early stage would have a significant impact on the success of therapeutic interventions. Magnetic resonance spectroscopy (MRS) is a technique that has the potential to detect small and early biochemical changes, and has been proven to be useful in the study of tissue biopsies. However, obtaining tissue biopsies usually involves an invasive procedure. Human stool, on the other hand, is available non-invasively and the collection does not present any risks to the subject. Moreover, the sample requires no special processing.

Methods

Human subjects who were scheduled for colonoscopy or surgery were recruited to donate a single sample of stool. Subjects were instructed to collect the first bowel movement after the start of their colonic preparation for colonoscopy or surgery the following day. Stool samples from 8 subjects (4 normal and 4 colon cancer patients) were shipped from U. T. M.D. Anderson Cancer Center in dry ice and kept frozen at -70 °C until the time of the experiment. Samples were thawed and an aliquot portion taken and suspended in PBS/D_2O buffer. The suspension was then put inside a capillary tube (filled to approximately one third of its volume) with one end closed with a teflon plug (1). This was then put in a standard 5 mm MR tube containing p-amino benzoic acid (PABA) that served as a chemical shift reference. All experiments were performed at 360 MHz (Bruker Instruments) at 25 °C with presaturation of the water signal. Both single pulse 1D (*90^opulse, NS = 256, RD = 2.41 s, TD = 4K, SW = 5000 Hz*), and 2D COSY (*NS = 128, NE = 180, TD = 2K, SW2 = 2906 Hz*) MRS experiments were performed on each stool sample.

Results and Discussion

There were easily noticeable differences in the 1D MR spectra of stool from normal subjects and those from cancer patients. As can be seen in Figure 1, the most striking results came from the 2D COSY MR spectra, where the appearance of a crosspeak at 1.3-4.3 (attributed to the methyl-methine couplings of bound fucose (2)) may serve as a marker for the presence of colorectal cancer. Whereas all the stool samples that came from cancer patients (n = 4) had this crosspeak in their COSY spectra, none of the normal ones (n = 4) showed the presence of this crosspeak.

This finding is consistent with earlier findings on colorectal cell lines and tissue biopsies where the presence of elevated levels of fucose on the cell surface was suggested to be a tumor marker (2,3). Besides indicating the presence and absence of cancer, the levels of fucose and the increase in complexity of the fucosylation patterns may correlate with the grade and/or the stage of the cancer. Although not as dramatic, there were also other notable differences in the COSY spectra. A multivariate analysis will be performed on the 1D MR spectra when an adequate patient cohort is reached. Moreover, MRS experiments will also be performed on stool samples from subjects with adenomas. By examining the 2D COSY spectral profile, one may be able to assess the malignant potential of adenomas, and thus, develop a more selective and efficient surveillance program for people with adenomas.

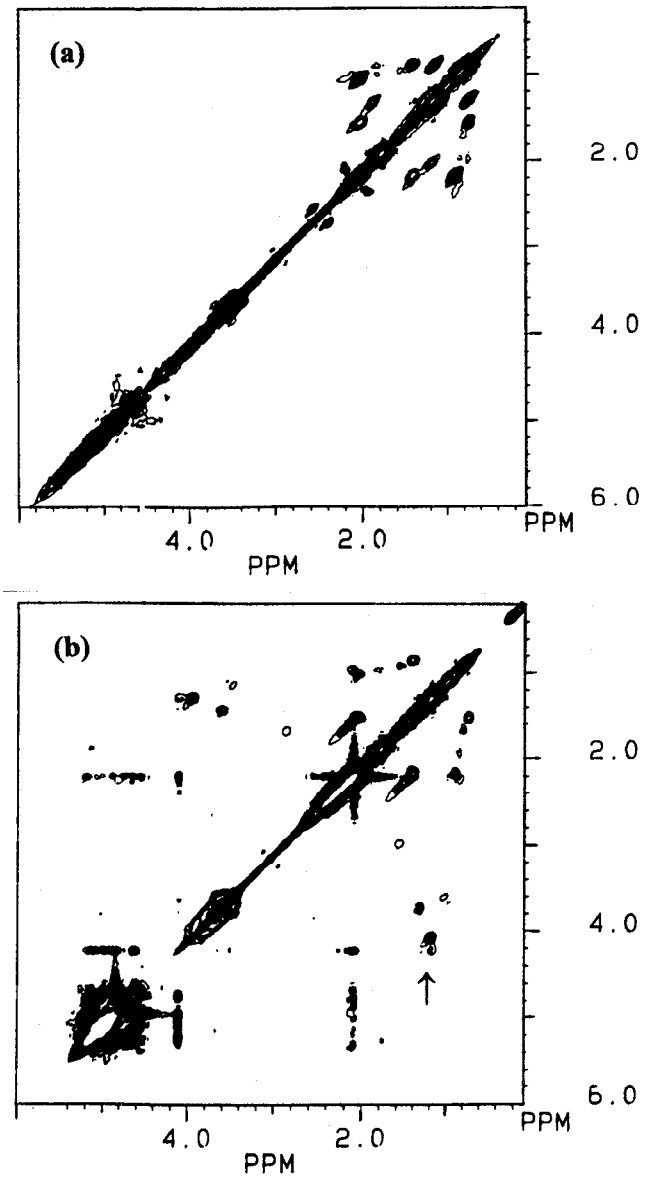

Figure 1. 360 MHz symmetrised COSY spectra of human stool from (a) normal subject, and (b) colon cancer patient.

Conclusion

This preliminary report shows convincing differences (in the fucose crosspeak) between the COSY spectra of human stool from healthy subjects and subjects with colon cancer. The potential role of MRS of stool as a simple, non-invasive clinical tool in the detection and surveillance of colorectal cancer is worthy of detailed examination.

Reference

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