

## Emerging Technologies in Screening for Colorectal Cancer: CT Colonography, Immunochemical Fecal Occult Blood Tests, and Stool Screening Using Molecular Markers

Bernard Levin, MD; Durado Brooks, MD, MPH; Robert A. Smith, PhD; Amy Stone, MCo

**Dr. Levin** is Vice President for Cancer Prevention, The University of Texas MD Anderson Cancer Center, Houston, TX.

**Dr. Brooks** is Director, Colorectal and Prostate Cancers, Cancer Control Sciences Department, American Cancer Society, Atlanta, GA.

**Dr. Smith** is Director, Cancer Screening, Cancer Control Sciences Department, American Cancer Society, Atlanta, GA.

**Ms. Stone** is President, Amy Stone Scientific and Medical Communications, Inc., Decatur, GA.

This article is available online at: <http://CAonline.AmCancerSoc.org>

**ABSTRACT** The American Cancer Society's (ACS) Colorectal Cancer Advisory Group held a workshop on new technologies for the early detection of colorectal cancer and adenomatous polyps as part of a regular review of ACS guidelines for colorectal cancer screening. The Advisory Group formally reviewed CT colonography, immunochemical fecal occult blood tests (FOBT), and stool screening using molecular markers, and also addressed other technologies including capsule video endoscopy. With the exception of immunochemical stool testing, the ACS has determined that at this time there is insufficient evidence to recommend these technologies for routine colorectal cancer screening. Based on recommendations of the Advisory Group, only a minor modification has been made to the ACS's Recommendations for Screening and Surveillance of the Early Detection of Adenomatous Polyps and Colorectal Cancer. (*CA Cancer J Clin* 2003;53:44-55). © American Cancer Society, 2003.

### INTRODUCTION

Colorectal cancer is the third most frequently diagnosed cancer among men and women in the United States and the second leading cause of cancer death. According to current estimates, colorectal cancer will develop in about 5.9 percent of the population over their lifetime.<sup>1</sup> Excluding deaths from lung cancer, colorectal cancer is the most common cause of cancer death for men and women combined.

Colorectal cancer is a type of cancer for which screening is particularly effective. Screening can detect adenomatous polyps, precursors to cancer that can be successfully removed thereby preventing the cancer from occurring.<sup>2</sup> Screening can also detect early stage colorectal cancer when it is very amenable to treatment, as evidenced by the fact that 90 percent of patients diagnosed with localized disease are alive five years after diagnosis.<sup>3</sup> Currently, however, only about 37 percent of colorectal cancers are diagnosed at this stage, reflecting low rates of awareness about the disease and a recent screening rate of less than 40 percent of all people who should be screened.<sup>4</sup> Although several effective screening methods are available, including FOBT, flexible sigmoidoscopy, double contrast barium enema, and colonoscopy, some are more acceptable to patients than others. For many individuals, one or more of these procedures

**Author disclosures:** Bernard Levin, MD, serves on the Scientific Advisory Board of Enterix, Inc. Theodore Levin, MD, has received research support from EXACT Sciences Corporation, and anticipates commencing a study with and receiving research support from Enterix, Inc. Richard Wender, MD, serves on the Scientific Advisory Board of Enterix, Inc.

is uncomfortable or unpleasant, and some of the tests require preparation that many consider to be worse than the procedure.<sup>5</sup> Additionally, for a variety of reasons, physicians do not routinely recommend all risk-eligible patients undergo colorectal cancer screening. These facts, coupled with a lack of awareness about the disease and screening as well as insurance issues (inconsistent coverage and/or lack of insurance), probably account for the low prevalence of screening among Americans aged 50 and older.

Several new technologies for detection of colorectal cancer and adenomatous polyps are described as less invasive, more accurate, and more palatable to the public than the conventional screening tests. Given the potential relevance these new technologies may have to the ACS colorectal cancer screening guidelines,<sup>6</sup> and the fact that their use is being promoted to the public in various clinical settings, the American Cancer Society convened a workshop to review the current status of emerging technologies.

The technologies reviewed at the April 2002 meeting of the American Cancer Society Colorectal Cancer Advisory Group were:

- CT colonography (virtual colonoscopy).
- Immunochemical Fecal Occult Blood Tests with a focus on the InSure™ immunochemical test.
- Detection of altered human DNA in stool samples.

For this periodic review of new advances in colorectal cancer screening, published articles were identified using MEDLINE and other sources (National Library of Medicine bibliographies of identified articles, personal files of panel members, and unpublished manuscripts), and leading experts in these new technologies were invited to a one-day workshop to present the latest data related to the new screening tests. Advisory group members reviewed literature in advance of the meeting, and also considered unpublished evidence presented at the workshop to inform

deliberations about the current state of evidence for these new screening technologies. When evidence was insufficient or lacking, the final recommendations incorporated the expert opinions of the panel members. During the workshop and subsequent conference calls, consensus was reached on the key issues within the guideline recommendations.

---

#### CT COLONOGRAPHY AND VIRTUAL COLONOSCOPY

CT colonography is an imaging procedure that uses computer programming to combine multiple, helical CT scans in order to create two- or three-dimensional images of the interior of a patient's colon. These images can be rotated for different views and even combined for a complete view of the colon that can be "flown through." The term "virtual colonoscopy" was coined in 1994 by researchers at Wake Forest University who described this procedure as simulating conventional colonoscopy.<sup>7</sup> Other terms, including CT pneumocolon and CT colography have been used, but "CT colonography" has become the accepted medical terminology with "virtual colonoscopy" often being used as the popular lay term.<sup>8</sup>

#### Procedure

Practitioners in the field have developed the following basic principles for performing CT colonography:<sup>9</sup>

1. Prior to the test, a thorough colonic cleansing is required. Most clinicians recommend a 48-hour low-residue diet and an over-the-counter preparation of phospho-soda and bisacodyl. This preparation leaves less residual fluid in the bowel compared with polyethylene glycol electrolyte solution. As with colonoscopy, the success of the procedure is influenced by the completeness of this

**Members of the American Cancer Society's Colorectal Cancer Advisory Group**

Chair: Bernard Levin, MD, is Vice President for Cancer Prevention, The University of Texas MD Anderson Cancer Center, Houston, TX.

Vice Chair: Tim Byers, MD, is Professor, Preventive Medicine and Biometrics, University of Colorado Health Sciences Center, Denver, CO.

Briggs W. Andrews, Esq., is Senior Vice President, General Counsel, Carilion Health System, Roanoke, VA.

Lovell Jones, PhD, is Professor of Gynecologic Oncology, Professor of Biochemistry and Molecular Biology, and Director, Center of Excellence for Research on Minority Health, and Director, Experimental Gynecology/Endocrinology, Head, Breast Cancer Nutrition Research Group, Department of Gynecologic Oncology, The University of Texas MD Anderson Cancer Center, Houston, TX.

Gordon Klatt, MD, is Medical Director, Cancer Programs, Mt. Ranier Surgical Association, Tacoma, WA.

Cynthia LeBlanc, EdD, is Deputy Superintendent, Hayward Unified School District, Richmond, CA.

Theodore Levin, MD, is Staff Physician, Gastroenterology Department, Kaiser Permanente Walnut Creek Medical Center, Walnut Creek, CA, and Physician/Scientist, Kaiser Permanente Division of Research, Oakland, CA.

Pam McAllister, PhD, is Chair, Board of Directors, Colorectal Cancer Network, Madison, WI.

Sigurd Normann, MD, PhD, is Professor, Department of Pathology, Immunology and Laboratory Medicine, University of Florida, Gainesville, FL.

cleansing. This means that CT colonography technology currently does not remove one of the obstacles to colonoscopy frequently cited by patients—the need for bowel preparation.

2. The colon is insufflated with room air via a rectal tube to the maximum level tolerated by the patient. Carbon dioxide may be substituted, which may reduce patient discomfort, but does add to the complexity and cost of the procedure.

3. Helical CT scanning is performed in a single breath-hold, generally using 5 mm collimation and reconstruction intervals of 2 to 3 mm with the patient in both supine and prone positions to redistribute the gas into segments of the colon that may have been collapsed. New multidetector CT scanners can scan faster (less than 15 seconds, hence a shorter breath-hold), and use thinner reconstruction intervals and collimations for finer anatomic detail.

4. The data from the CT scans are displayed as 2D and/or 3D images. Generally, but not universally, 2D images on a computer workstation are used for lesion identification, and when a suspicious area is encountered, a 3D view can be used for further examination. To obtain the 3D view, the CT data are processed on the same workstation equipped with specialized software. The resulting virtual environment and endoluminal “fly-through” allows the physician to view the entire colon relatively quickly. This is an advantage since the helical CT technology

produces so many slices that the reader needs the computer post-processing to efficiently analyze all the data. The 3D “virtual reality” environment also allows viewing of hidden surfaces of folds and flexures from different angles.

Using 2D and 3D images together allows differentiation between complex haustral folds and polyps, for example, or between retained stool and polyps.

**Current Evidence**

Results from major centers in the United States show accuracy of CT colonography to be comparable to conventional colonoscopy for the detection of polyps greater than 10 mm with few false positives.<sup>9</sup> In the hands of highly experienced radiologists, polyps greater than 10 mm are detected with sensitivity and specificity approaching 90 percent, with sensitivity falling to 50 percent for polyps 5 mm in size. Some published studies have also found CT colonography to be effective in detecting frank colon cancers with a sensitivity of 100 percent and no false positives.<sup>10-12</sup> Table 1,<sup>13-15</sup> highlights results from three recent clinical trials from academic centers studying the sensitivity of CT colonography.<sup>9</sup> The sensitivity of CT colonography for detecting individual polyps of various sizes (No. Per Polyp) is substantially greater for individual polyps > 10 mm compared with individual polyps less than 10 mm in size. For patients with at least one polyp larger than 6 mm (No. Per Patient), CT colonography had nearly equivalent sensitivity for polyps 6 to 9 mm compared with polyps > 10 mm. Insofar as these trials represent a small series of patients examined by experts investigating CT colonography, the generalizability of these findings to the screening setting requires further study.

It is important to note that most published studies of virtual colonoscopy involve individuals undergoing diagnostic studies or

high-risk patient groups, as is common in studies of new screening tests, in order to have a higher prevalence of occult disease. These studies also emanate from radiologists who are highly experienced in the relevant techniques. Large clinical trials are underway comparing colonoscopy with CT colonography in average-risk and high-risk screening populations.<sup>16</sup>

#### Advantages/Indications of CT Colonography

CT colonography offers the advantage of potentially identifying cancers in the colon that may not be adequately assessed or identified by conventional endoscopy, e.g., those located near complex haustral folds. Additionally, it has the ability to image the colon proximal to occlusions and redundant loops; and in many settings it is becoming the technique of choice for completing examination of the colon after failed or incomplete colonoscopy.

CT colonography also offers the patient a choice regarding polypectomy. Since small and likely hyperplastic polyps are not likely to progress to cancer in the near term, there may be little therapeutic benefit in removing them. However, leaving small polyps behind means reassessment on a regular basis to monitor them, and CT colonography may be

determined to be an appropriate way to follow such patients. In the future, if CT colonography proves to be a useful screening test, it may offer a 'roadmap' for endoscopists performing therapeutic colonoscopies showing the number and location of polyps to be removed.

#### Limitations of CT Colonography

A number of limitations of CT colonography have been identified, including challenges that are common to other visual inspection tests of the colon as well as those unique to CT colonography:

- False-positive readings occur in about 15 percent of cases, and may result in unnecessary follow-up colonoscopy. The main causes of false-positive results have been:
  1. Retained stool.
  2. Diverticular disease, which results in areas of the colon that are poorly distensible.

Patricia T. Patterson, RN, MN, ET, was Nurse Navigator, York Cancer Center, York, PA, and is currently Regional Director of Cancer Control, Pennsylvania Division, American Cancer Society, Pittsburgh, PA.

David Rothenberger, MD, is Professor and Chief, Divisions of Colon and Rectal Surgery and Surgical Oncology, Department of Surgery, University of Minnesota, and Associate Director for Clinical Research and Programs, University of Minnesota Cancer Center, Minneapolis, MN.

Alan Thorson, MD, is Associate Professor of Surgery and Program Director, Section of Colon and Rectal Surgery, Creighton University School of Medicine, Omaha, NE, and Clinical Associate Professor of Surgery, University of Nebraska College of Medicine, Omaha, NE.

David Vining, MD, is Assistant Professor of Diagnostic Radiology, Virtual Endoscopy Center, Wake Forest University School of Medicine, Winston-Salem, NC.

Richard Wender, MD, is Chair, Department of Family Medicine, Thomas Jefferson University, Philadelphia, PA.

#### Presenters and Affiliations

CT Colonography: Joseph Ferrucci, MD, is Professor and Chair, Department of Radiology, Boston Medical Center, Boston University School of Medicine, Boston, MA.

InSure™ Fecal Occult Blood Testing: Graeme P. Young, MD, FRACP, is Professor of Gastroenterology and Academic Head of GI Services, Flinders University of South Australia, Adelaide, Australia.

DNA-based Stool Screening Methods: David A. Ahlquist, MD, is Professor, Gastroenterology and Internal Medicine, Mayo Clinic, Rochester, MN.

TABLE 1

Results of Virtual Colonoscopy: Sensitivity for Polyp Detection (Colonoscopic Correlations) <sup>9*</sup>							
Study	Date	Patients	Sensitivity				
			No. Per Polyp (%)			No. Per Patient (%)	
			Polyps > 5 mm	6-9 mm	>10 mm	6-9 mm	> 10 mm
Boston University, Fenlon, et al. <sup>13</sup>	11/99	100	58	36(82)	22(91)	(92)	(96)
Mayo Clinic, Fletcher, et al. <sup>14</sup>	9/00	180	263	142(47)	121(75)	(88)	(85)
University of California San Francisco, Yee, et al. <sup>15</sup>	6/01	300	223	141(80)	82(90)	(93)	(100)

\*Reprinted with permission from the *American Journal of Roentgenology*.

3. Thick or complex haustral folds misinterpreted as polyps or masses.

4. Metal and motion artifacts: metal artifacts (i.e., image streak artifact) result from x-rays passing through metallic hip prostheses, Herrington rod implants, etc. (motion artifacts are due to respiratory excursions or other body motion).

- An unknown ability to detect flat adenomas. While relatively rare, flat adenomas are thought to be more aggressive than typical adenomatous polyps; therefore, their detection is critical. No data are available on the ability of CT colonoscopy to detect these lesions. However, hyperplastic polyps, which are softer than adenomatous polyps and therefore flatten against the surface of the air-distended colon, are not as easily detected with CT colonoscopy. This has led to speculation that perhaps the technology would not be as sensitive for the detection of flat adenomas.

- Lack of standards for performance, training, and reading of scans.

- CT colonography is not therapeutic, i.e., polyps cannot be removed during the procedure. In a recent study of over 3,000 US Army veterans (with mean age of 64 years) undergoing colonoscopy, 38 percent had one or more neoplastic lesions, and 8 percent had advanced lesions (i.e., polyps  $\geq$  10 mm). If a polyp is found that needs to be removed, the patient will need to undergo conventional colonoscopic polypectomy. It is important to note, however, that there are differing opinions about whether or not this is a shortcoming, since most individuals undergoing screening do not require diagnostic evaluation or intervention.

- The cost of CT colonography may be higher than conventional colonoscopy. The cost analysis of this procedure should include the time required to perform the procedure and the time required for a radiologist to interpret the resulting images. Additionally, all positive tests must be followed up with conventional colonoscopy.

## Discussion

A growing body of research suggests that many small polyps, i.e., those less than 10 mm, may not progress to cancer in the patient's lifetime, and may be of little clinical significance to the average adult.<sup>17,9</sup> Therefore, the fact that CT colonography may not be as accurate in detecting these smaller polyps may ultimately prove to be of little significance. Indeed, CT colonography may be best at detecting the polyps that should be removed, and thus the real potential benefit of CT colonography may be to segregate the population into those who need colonoscopy from those who don't. However, more research is needed to answer these questions. Studies are also needed on the ability of CT colonography to detect flat adenomas.

As the technology currently exists, CT colonography requires bowel preparation. Most investigators have found that patients find the preparation more unpleasant than the procedure, both for conventional and virtual colonoscopy. New protocols exploring stool tagging with pre-exam consumption of a radio-opaque material or through the use of "electronic cleansing" (computerized or digital elimination of stool based on density measurement) hold the potential to reduce or even eliminate the amount of preparation involved, creating an advantage of CT colonography over colonoscopy.<sup>18</sup>

Another practical issue is the fact that there is a learning curve for radiologists to master the technology. Even though the technology is becoming widespread, it lacks the oversight of national standards, which should include the training of radiologists, standardization of protocols, and certification of the technology. These issues are currently being addressed by leading practitioners in the field.

Another concern is the presence of radiologic detection of extracolonic abnormalities, many of which may require evalu-

ation.<sup>19</sup> A medical consensus needs to emerge on the management of these incidental findings.

Further computing advances, including computer-assisted diagnosis, will likely make CT colonography easier and more accurate to use. However, there is a concern that the newer generation of CT colonography technology uses higher doses of radiation. This concern is alleviated somewhat by the fact that if screening begins at age 50 and occurs every five to seven years, the cumulative radiation doses result in only small, theoretical increases in the risk of developing cancer, which are measurably offset by the benefit of early detection of colorectal cancer. Nevertheless, as with other screening procedures, potential harms must be recognized and obviated where possible.

#### Conclusion

The advisory group concluded that CT colonography is a compelling, emerging technology that shows considerable promise, but it has not yet been studied in a typical screening population; therefore, whether or not it has comparable or superior performance compared with conventional tests is unknown.

---

#### EMERGING TECHNOLOGIES RELATED TO STOOL SCREENING: IMMUNOCHEMICAL FECAL OCCULT BLOOD TESTS AND MOLECULAR MARKERS

Conventional stool testing for colorectal cancer has many positive aspects. It is noninvasive. Samples can be collected in the privacy of a person's home. It detects lesions throughout the length of the large bowel. It requires no bowel preparation, and it is inexpensive. Additionally, clinical trials have shown that mortality rates from colorectal cancer can be reduced by up to 33 percent with annual screening with FOBT.<sup>20-22</sup>

However, current recommendations for

FOBT using guaiac-based tests still require some dietary preparation, e.g., abstention from red meat and NSAIDs for three to five days prior to taking the test.<sup>23</sup> In addition, FOBT must be performed annually to increase the chance of detecting intermittent blood, and it carries high rates of false-negative and false-positive results. False-negative results represent missed advanced lesions, and false-positive readings cause great anxiety, discomfort, and potential harm from unnecessary tests as well as increased costs as further tests are performed.

#### Immunochemical Fecal Occult Blood Testing

Tests that examine the stool for the presence of blood currently rely on two main methods: guaiac and immunochemical.

The guaiac tests are the most commonly used and include brand names such as Hemoccult II® and Hemoccult II® SENSEA® (Beckman Coulter, Inc., Fullerton, CA). The test consists of three cards impregnated with guaiac that, when treated with a developer containing hydrogen peroxidase, give a color-coded result based on the presence or absence of peroxidase-like activity of the heme in the stool. In the United States, the majority of FOBTs performed use the guaiac-based method. These tests can yield false-positive results if certain foods, vitamins, or drugs (including meat, some raw fruits and vegetables, vitamin C tablets or high levels of foods containing vitamin C, or aspirin) are ingested in the days before taking the test.<sup>6,23,24</sup> Since cancers and precancerous polyps bleed intermittently, the tests need to be repeated, generally obtaining multiple stool samples from consecutive bowel movements in order to minimize the possibility of missing blood in the stool. Research is underway to develop tests that maintain the advantages of traditional FOBT (privacy, non-invasiveness, ability to collect samples at home), while increasing the sensitivity and specificity of the tests.

The immunochemical tests, such as HemeSelect (SmithKline Diagnostics, San Jose, CA), InSure™ (Enterix, Inc., Falmouth, ME), FlexsureOBT® (SmithKline Diagnostics, San Jose, CA), and others employ a more complex reaction that uses monoclonal and/or polyclonal antibodies that detect the intact globin protein portion of human hemoglobin. If hemoglobin is present in the stool, the labeled antibody will attach to its antigens, creating a positive test result. The problem of dietary restriction is solved by the use of immunochemical FOBTs since they do not react with non-human hemoglobin or with uncooked fruits and vegetables that may contain peroxidase activity that could cause a false-positive result with the guaiac-based tests. Immunochemical tests have, in general, performed well in clinical studies. However, for technical and commercial reasons, these tests have not found wide usage among US physicians.<sup>25,26</sup>

#### *InSure™ Fecal Occult Blood Test*

The Advisory Committee reviewed data on InSure™—a new immunochemical FOBT, which was approved for use by the Food and Drug Administration (FDA) in January 2001. Like other immunochemical stool tests, InSure™ detects the globin portion of the hemoglobin molecule. Since globin does not survive passage through the upper GI tract, the test is specific for occult bleeding in the colon and rectum.<sup>27</sup> The InSure™ immunochemical test differs from earlier immunochemical tests in that the sampling procedure is simpler and more user friendly, and the test result is quantifiable in terms of the amount of blood present in the sample. Current FDA approval of the test, however, is not based on quantification of blood volume, but rather a qualitative interpretation of the test conducted by a human reader based on color coding.

To use InSure™ a long-handled brush is used

to brush the surface of the stool while in the toilet bowl, thus trapping dislodged occult blood and water on the bristles. The brush is then dabbed onto the test card where the water dries into a special pad. Once dry, the hemoglobin is considered stable and the card is sent to a laboratory for testing.

Once the test card is received at the laboratory, an immunochromatographic test strip consisting of a test line and a control line is inserted into the test pad of the card. Liquid is added, causing any hemoglobin present to migrate up the test strip. If hemoglobin is present in the sample, it binds to a colloidal gold-conjugated polyclonal antibody against hemoglobin, and is labeled red. The control line has an antibody that reacts with the conjugate antibody, which also causes a red line to form indicating the test has functioned correctly. A positive result will produce two red lines on the test strip, whereas a negative result produces only one red line (i.e., the control line), provided that the test has functioned properly. The test is completed in approximately five minutes.

InSure™ was tested in a group of 240 people considered at high risk of developing colorectal cancer based on their personal or family history of colorectal cancer and adenomas, and also scheduled for colonoscopy. Two samples per patient were gathered. In this study, the test had a sensitivity for cancer of 87 percent (20/23) and a sensitivity for adenomas >10 mm of 47.4 percent (9/19). In a separate, normal population aged 40 and over that was evaluated for test specificity, specificity was 97.9 percent (88/90), and in a population under age 30, specificity was 97.8 percent (92/94).<sup>28</sup> Test results were verified by colonoscopy.

According to the manufacturer's FDA application, InSure™ has been tested in vitro against samples of myoglobin and hemoglobin from beef, chicken, fish, horses, pigs, rabbits, deer, sheep, and kangaroos with negative results. Similarly, when extracts of raw broccoli,

cantaloupe, cauliflower, horseradish, red radish, and turnip were added to the InSure™ test card, the results also were negative. These are the types of foods that give false positives with guaiac tests. Additionally, up to this point no evidence has been found that any of the toilet bowl deodorizers/fresheners or cleaners studied interfered with test function or accuracy.

To determine the effect of upper GI bleeding on InSure™—two healthy subjects ingested 20 ml of autologous blood immediately after the blood was obtained. Subjects collected samples with InSure™ one day prior to ingesting the blood and from each bowel movement after until six samples had been collected. None of the samples tested positive by InSure™ FOBT. Similar specificity for lower GI bleeding has been demonstrated for other immunochemical tests.<sup>24,27</sup>

#### ADVANTAGES AND LIMITATIONS OF !NSURE™ FOBT

Advantages of an immunochemical test compared with a guaiac test include:

- Improved specificity. Immunochemical tests will not react with non-human hemoglobin, vitamins, drugs, or peroxidase from food sources. InSure™ FOBT has also been shown to be non-reactive with blood from the upper gastrointestinal tract when bleeding is occult.

- Potential increase in patient compliance. Since no dietary restrictions are needed, and since InSure™ requires collection from only two stool specimens and is performed by swirling a brush in the toilet water with the stool, it may be more acceptable to the consumer than current FOBT tests with their higher testing and stool handling requirements.

Disadvantages of an immunochemical test compared with a guaiac test include:

- Limited clinical testing. InSure™ FOBT has not been tested in a large screening population of average-risk individuals, although trials are

underway in Queensland, Australia, and Chicago, Illinois, with additional studies being planned.

- Sensitivity limitations. While immunochemical tests have advantages over guaiac tests, they are still tests for occult blood, which may leak intermittently and may occur from sources in the colon or rectum other than cancers or large adenomas. Data indicate that the problem for detection created by intermittency is less marked with immunochemical than with guaiac tests because higher test sensitivity is not accompanied by significant degradation of specificity, as is the case with guaiac-based tests. In addition, because bleeding from adenomas occurs infrequently, the potential for CRC prevention through adenoma detection and removal is likely to be lower with this and all FOBT methods than with endoscopic and imaging screening modalities. However, when used annually, as recommended, the program sensitivity of FOBT is very high.<sup>20</sup>

#### Conclusion

Even though the Advisory Group noted that the background of studies for InSure™ was small, it believes immunochemical tests have advantages over guaiac tests. In the opinion of the Advisory Group, the recent studies of InSure™ combined with previously published reports on the performance of immunochemical tests for stool occult blood<sup>24,25,27,29</sup> provide a persuasive argument that these tests offer enhanced specificity in colorectal cancer screening compared with guaiac-based testing. Based on the advisory group's report, the ACS's Recommendations for Screening and Surveillance for the Early Detection of Adenomatous Polyps and Colorectal Cancer now include the statement: "in comparison with guaiac-based tests for the detection of occult blood, immunochemical tests are more patient-friendly, and are likely to be equal or better in sensitivity and specificity."<sup>4</sup>

### Screening Stool for DNA Mutations

Colorectal cancer is a disease in which many DNA mutations associated with the process of carcinogenesis have been characterized.<sup>30</sup> This characterization makes it potentially valuable to examine the stool for the presence of DNA with these different mutations as indicators of both pre-clinical and clinical disease. This is important since no single mutation has been found that is expressed in all colorectal cancers. Further, research has shown DNA to be a good marker. It is stable in the stool; it is shed continuously; and, through the use of amplification tests, such as polymerase chain reaction, it can be detected in minute amounts.<sup>31</sup> Early clinical studies in a small number of patients with a multi-target DNA-based stool assay approach suggest high sensitivity for both colorectal cancer and premalignant adenomatous polyps while maintaining high specificity.<sup>31</sup> Other investigators have published data on detection of APC gene mutations using a digital protein truncation assay with a similarly high specificity rate.<sup>32</sup>

#### *EXACT Sciences Corporation's Test*

One stool test for DNA mutations has been studied in a number of small trials.<sup>33</sup> EXACT Sciences Corporation, in Maynard, MA, has created a prototype multicomponent test that targets point mutations at 15 mutational hot spots on *K-ras*, *APC*, and *p53* genes, mutations on *Bat-26* (a microsatellite instability marker), and long DNA (i.e., DNA not degraded by apoptosis). This multicomponent test was studied in a blinded clinical study in which 61 people each contributed a single stool sample. Of these 61 individuals, 22 were known to have colorectal cancer, 11 had large premalignant adenomatous polyps, and 28 had normal colons as shown on endoscopy. Analyzable DNA was recovered from all 61 stools evaluated. Each marker contributed to the findings. Results showed a sensitivity of 91 percent for cancer and 82 percent for large adenomas with an

initial specificity of 93 percent. Excluding *K-ras* from the analysis (since *K-ras* mutations can appear in histologically-normal colonic mucosa), sensitivities for cancer were unchanged but decreased slightly for large adenomas to 73 percent while specificity rose to 100 percent.

Cancers were detected with high sensitivity from sites throughout the colon. This was an important finding since it showed DNA is not degraded as it passes through the length of the colon.

#### *Advantages and Limitations of Stool DNA Mutation Testing*

During discussion of stool DNA mutation testing, the following points were raised as advantages:

- Neoplasm specific. DNA tests target specific mutations known to occur in colorectal cancers. Other conditions that may cause gastrointestinal bleeding will not cause a false-positive result.

- Consistent markers. DNA is shed continuously from colon cancer and its precursor polyps, thus only a single stool sample is needed for analysis. Bleeding from cancers or polyps usually occurs only intermittently, requiring the collection of multiple samples for occult blood testing.

- Highly accurate. The tests detect target mutations with a high degree of sensitivity and specificity.

- User-friendly. The test is non-invasive, allows for off-site collection, requires no preparation, and may allow for less frequent screening intervals as compared with annual FOBT.

- Reduced false-positive results. The test relies on functional, not spatial, detection of polyps and cancers. By using a molecular profile rather than a physical shape, location, or size, the test may reduce false-positive readings when compared with screening modalities utilizing imaging or direct visualization of lesions.

- Ability to detect cancers proximal to the

colon. Aerodigestive cancers (e.g., lung, esophageal, stomach, pancreas, colorectal) collectively account for more than half of all malignant deaths. Each of these cancers exfoliate cells (and hence DNA) into the GI tract or into other fluids (such as sputum or bile) that enter the GI tract, and preliminary studies show that altered DNA from these cancers can be detected through the stools.

- **Cost.** Since the lifetime risk of colorectal cancer is approximately six percent and only about half the population will develop adenomas, the test may ultimately improve cost effectiveness of screening for colorectal cancer by limiting the need for colonoscopy strictly to individuals with adenomatous polyps or cancer identified through this method. Further, testing for altered DNA in stool may also decrease the number of surveillance colonoscopies needed after therapy for colonic neoplasia.

The following limitations were also discussed with regard to DNA stool tests:

- **Lack of data from screening populations.** There is a need for clinical studies in the general population. With regard to this point, it was noted that EXACT Sciences Corporation has organized a trial comparing DNA stool testing to guaiac-based FOBT tests with 5,000 subjects that will be completed in 2003. Additionally, a study by the Mayo Clinic and the NIH with 4,000 subjects will be completed in 2004.

- **A need for test refinement.** New markers and testing methods are being defined continuously (APC protein truncation assay, hypermethylated DNA), and there is a lack of consensus on how many or which markers are necessary for the test to achieve an acceptable sensitivity level. More markers will increase sensitivity, while adding markers also adds cost to the procedure.

- **Automation.** Automation of the tests needs to be explored to reduce costs and improve efficiency and turn-around time.

- **Implementation.** The stool collection kits for this test are large, creating a potential space/storage problem in doctors' offices.

- **Expense.** Currently, these tests are expensive, costing over \$400 per test.

- **Patient acceptance/adherence.** This test requires the patient to send the entire bowel movement to the processing laboratory, something some patients may find unacceptable.

### *Conclusion*

The science of DNA stool testing is an emerging technology. While DNA stool testing holds the potential to be more specific than FOBT, the committee recognized that there are still unanswered questions regarding the test. Therefore, the committee concluded that remaining questions related to the most appropriate markers for DNA detection of cancer, on the best combination of markers, and on the results of studies in populations at average risk for colorectal cancer are required before DNA stool testing can be recommended as a test for average-risk individuals.

---

### CAPSULE VIDEO ENDOSCOPY

During this review, one additional new technology was considered—specifically, capsule video endoscopy (the camera in a capsule). Capsule video endoscopy was not included in the formal program, but was discussed in the context of providing guidance to physicians and their patients who may have questions due to recent media exposure.

Media outlets around the world have given coverage to a “camera in a capsule,” which, once swallowed, provides images of the digestive tract for up to eight hours.

Given Imaging Ltd., an Israel-based company, developed the pill, formally called M2A™ Capsule. It was cleared by the FDA in August 2001 and is currently approved for sale in the United States, the European Union, Canada, Australia, New Zealand, and Israel. The M2A™ Capsule contains a tiny camera that snaps two pictures per second as it travels through the small intestine. It works in

## APPENDIX A

## American Cancer Society Guideline on Screening and Surveillance for the Early Detection of Colorectal Adenomas and Cancer—Average-Risk Women and Men aged 50 and Older

Test	Interval (beginning at age 50)	Comment
Fecal Occult Blood Test (FOBT)*	Annually	The recommended take-home multiple sample method should be used. All positive tests should be followed up with colonoscopy.*†
Flexible Sigmoidoscopy	Every 5 years	All positive tests should be followed up with colonoscopy.*
Fecal Occult Blood Test (FOBT)* and Flexible Sigmoidoscopy	FOBT annually and flexible sigmoidoscopy every 5 years	Flexible sigmoidoscopy together with FOBT is preferred compared with FOBT or flexible sigmoidoscopy alone. All positive tests should be followed up with colonoscopy.‡
Double Contrast Barium Enema	Every 5 years	All positive tests should be followed up with colonoscopy.
Colonoscopy	Every 10 years	Colonoscopy provides an opportunity to visualize, sample, and/or remove significant lesions.

\*FOBT as it is sometimes done in physicians' offices, with the single stool sample collected on the fingertip during a digital rectal examination, is not an adequate substitute for the recommended at-home procedure of collecting two samples from three consecutive specimens. Toilet bowl FOBT tests also are not recommended. In comparison with guaiac-based tests for the detection of occult blood, immunochemical tests are more patient-friendly, and are likely to be equal or better in sensitivity and specificity.

†There is no justification for repeating FOBT in response to an initial positive finding.

‡If colonoscopy is unavailable, not feasible, or not desired by the patient, double contrast barium enema (DCBE) alone or the combination of flexible sigmoidoscopy and DCBE are acceptable alternatives. Adding flexible sigmoidoscopy to DCBE may provide a more comprehensive diagnostic evaluation than DCBE alone in finding significant lesions. A supplementary DCBE may be needed if a colonoscopic exam fails to reach the cecum, and a supplementary colonoscopy may be needed if a DCBE identifies a possible lesion or does not adequately visualize the entire colorectum.

conjunction with the Given Diagnostic System, which includes a portable data recorder worn on a belt and a workstation that processes the data and produces a video of the images.

The FDA cleared the device based on animal and clinical studies of safety and effectiveness conducted by the manufacturer. Results show the camera pill was safe, without any side effects, and was able to detect abnormalities in the small intestine, including parts that cannot be reached by an endoscope.

Clinical studies of this technology to date have been limited to use of the device in the upper gastrointestinal tract (stomach and small intestine). Since the battery only lasts about eight hours, the camera begins to run down by the time it gets to the colon. The battery-life issue is also complicated by the slower transit time of material in the colon compared with the small intestine. The colon lumen is also

wider than that of the small intestine, making it possible for the camera to miss suspicious areas in the colon simply by being pointed in the wrong direction. In addition, fecal material in the colon will obscure the views of the bowel lumen wall, making interpretation of transmitted images much more difficult and less reliable. Although there have been inquiries about using the device for colorectal cancer screening and some sporadic entrepreneurial use, at the present time there is no evidence to support the use of capsule video endoscopy for the detection of colorectal polyps or cancers, and it is not being marketed for that purpose by the manufacturer. Refinement of equipment and technique as well as evaluation of the technology for detection of advanced colorectal neoplasia will be needed before this technology can be considered for diagnostic or screening use in the colon.

## CONCLUSION

After reviewing and discussing the technologies in this report, the Advisory Group concluded that while CT colonography and screening stool for DNA mutations are promising technologies, at this time they remain unproven as screening modalities for the general population. Therefore, at this time the Advisory Group recommends only a minor modification to the ACS's Recommendations for Screening

and Surveillance for the Early Detection of Adenomatous Polyps and Colorectal Cancer (Appendix A). The proposed modification is to append the guideline regarding fecal occult blood testing to include: "in comparison with guaiac-based tests for the detection of occult blood, immunochemical tests are more patient-friendly, and are likely to be equal or better in sensitivity and specificity." The Advisory Group will revisit these technologies in the future as additional data become available. CA

## REFERENCES

1. DEVCAN: Probability of developing or dying of cancer. Software, Version 4.0. Feur EJ, Wun LM. Bethesda, MD: National Cancer Institute; 1999.
2. Winawer SJ, Zauber AG, Ho MN, et al. Prevention of colorectal cancer by colonoscopic polypectomy: The National Polyp Study Workgroup. *N Engl J Med* 1993;329:1977-1981.
3. Ries LAG, Eisner MP, Kosary CL, et al. (eds). SEER Cancer Statistics Review, 1973-1999. National Cancer Institute. Bethesda, MD, 2002. Available at: [http://seer.cancer.gov/csr/1973\\_1999/](http://seer.cancer.gov/csr/1973_1999/). Accessed November, 2002.
4. Smith R, Cokkinides V, Eyre H. American Cancer Society guidelines for the early detection of cancer. *CA Cancer J Clin* 2003;53:27-43.
5. Vernon SW. Participation in colorectal cancer screening: A review [see comments]. *J Natl Cancer Inst* 1997;89:1406-1422.
6. Smith RA, von Eschenbach AC, Wender R, et al. American Cancer Society guidelines for the early detection of cancer: Update of early detection guidelines for prostate, colorectal, and endometrial cancers. Also: Update 2001—testing for early lung cancer detection. *CA Cancer J Clin* 2001;51:38-75.
7. Vining DJ, Gelfand DW. Non-invasive colonoscopy using helical CT scanning, 3D reconstruction, and virtual reality. Presented at: 23rd Annual Meeting, Society of Gastrointestinal Radiologists, Maui, HI, February, 1994.
8. Johnson CD, Hara AK, Reed JE. Virtual endoscopy: What's in a name? *AJR Am J Roentgenol* 1998;171:1201-1202.
9. Ferrucci JT. Colon cancer screening with virtual colonoscopy: Promise, polyps, politics. *AJR Am J Roentgenol* 2001;177:975-988.
10. Hara AK, Johnson CD, Reed JE, et al. Colorectal polyp detection with CT colonography: Two- versus three- dimensional techniques. *Work in progress*. [see comments]. *Radiology* 1996;200:49-54.
11. Hara AK, Johnson CD, Reed JE, et al. Detection of colorectal polyps with CT colonography: Initial assessment of sensitivity and specificity. *Radiology* 1997;205:59-65.
12. Royster AP, Fenlon HM, Clarke PD, et al. CT colonoscopy of colorectal neoplasms: Two-dimensional and three-dimensional virtual-reality techniques with colonoscopic correlation. *AJR Am J Roentgenol* 1997;169:1237-1242.
13. Fenlon HM, Clarke PD, Ferrucci JT. Virtual colonoscopy: Imaging features with colonoscopic correlation. *AJR Am J Roentgenol* 1998;170:1303-1309.
14. Fletcher JG, Johnson CD, Welch TJ, et al. Optimization of CT colonography technique: Prospective trial in 180 patients. *Radiology* 2000;216:704-711.
15. Yee J, Akerkar GA, Hung RK, et al. Colorectal neoplasia: Performance characteristics of CT colonography for detection in 300 patients. *Radiology* 2001;219:685-692.
16. American College of Radiology Imaging Network. Evaluating the accuracy of computerized tomographic colonography compared with pathology and colonoscopy for detecting clinically important colorectal neoplasia in a screening population (ACRIN Protocol A6664 - In Development.). Vol. 2002: American College of Radiology, 2002.
17. Eide TJ. Natural history of adenomas. *World J Surg* 1991;15:3-6.
18. Vining DJ, Pineau BC. Improved bowel preparation for virtual colonoscopy examinations [abstract]. *Gastroenterology* 1999;116:524A.
19. Hara AK, Johnson CD, MacCarty RL, et al. Incidental extracolonic findings at CT colonography. *Radiology* 2000;215:353-357.
20. Mandel JS, Bond JH, Church TR, et al. Reducing mortality from colorectal cancer by screening for fecal occult blood. Minnesota Colon Cancer Control Study. [published erratum appears in *N Engl J Med* 1993;329:672 (see comments)]. *N Engl J Med* 1993;328:1365-1371.
21. Hardcastle JD, Chamberlain JO, Robinson MH, et al. Randomised controlled trial of faecal-occult-blood screening for colorectal cancer [see comments]. *Lancet* 1996;348:1472-1477.
22. Kronborg O, Fenger C, Olsen J, et al. Randomised study of screening for colorectal cancer with faecal-occult-blood test [see comments]. *Lancet* 1996;348:1467-1471.
23. Ransohoff DF, Lang CA. Screening for colorectal cancer with the fecal occult blood test: A background paper. American College of Physicians [see comments]. *Ann Intern Med* 1997;126:811-822.
24. American College of Physicians. Suggested technique for fecal occult blood testing and interpretation in colorectal cancer screening. *Ann Intern Med* 1997;126:808-810.
25. Young GP, St John DJ, Winawer SJ, et al. Choice of fecal occult blood tests for colorectal cancer screening: Recommendations based on performance characteristics in population studies: A WHO (World Health Organization) and OMED (World Organization for Digestive Endoscopy) report. *Am J Gastroenterol* 2002;97:2499-2507.
26. Allison J, Tekawa I, Ransom L. A comparison of fecal occult-blood tests for colorectal-cancer screening. *N Engl J Med* 1996;334:155-159.
27. Rockey DC, Auslander A, Greenberg PD. Detection of upper gastrointestinal blood with fecal occult blood tests. *Am J Gastroenterol* 1999;94:344-350.
28. InSure, Summary of Safety and Effectiveness. Available at: <http://www.insurefobt.com/scientific03.html>. Accessed November, 2002.
29. Greenberg PD, Bertario L, Gnauck R, et al. A prospective multicenter evaluation of new fecal occult blood tests in patients undergoing colonoscopy. *Am J Gastroenterol* 2000;95:1331-1338.
30. Fearon ER, Vogelstein B. A genetic model for colorectal tumorigenesis. *Cell* 1990;61:759-767.
31. Ahlquist DA, Shuber AP. Stool screening for colorectal cancer: Evolution from occult blood to molecular markers. *Clin Chim Acta* 2002;315:157-168.
32. Traverso G, Shuber A, Levin B, et al. Detection of APC mutations in fecal DNA from patients with colorectal tumors. *N Engl J Med* 2002;346:311-320.
33. Ahlquist DA, Skoletsky JE, Boynton KA, et al. Colorectal cancer screening by detection of altered human DNA in stool: Feasibility of a multitarget assay panel. *Gastroenterology* 2000;119:1219-1227.