Original Article

Performance of Multitarget Stool DNA Testing in African American Patients

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BACKGROUND: Multitarget stool DNA (mt-sDNA) is an approved method for colon cancer screening that is especially relevant for patients who cannot undergo colonoscopy. Although the test performance has been evaluated in a large clinical trial, it was limited to a predominantly white population. Given differences in the epidemiology and biology of colon cancer in African American individuals, the authors sought to compare the performance of mt-sDNA between racial groups. **METHODS**: The authors prospectively identified patients aged ≥40 years who were referred for colonoscopy at an academic medical center and 2 satellite facilities. Prior to the colonoscopy, the authors collected stool for mt-sDNA and fecal immunochemical testing (FIT). They compared the sensitivity, specificity, and receiver operating characteristic curve between African American and white patients for the detection of advanced lesions or any adenoma. **RESULTS**: A total of 760 patients were included, 34.9% of whom were African American. The prevalence of any adenoma (38.9% for African American patients and 33.9% for white patients) and that for advanced lesions (6.8% and 6.7%, respectively) were similar between groups. The overall sensitivities of mt-sDNA for the detection of advanced lesions and any adenoma were 43% and 19%, respectively, and the specificities were 91% and 93%, respectively. In general, mt-sDNA was more sensitive and less specific than FIT. When stratified by race, the sensitivity, specificity, and receiver operating characteristic curve area were similar between African American and white patients for both mt-sDNA and FIT. **CONCLUSIONS**: Test performance characteristics of mt-sDNA were comparable in African American and white patients. Given the lower uptake of colonoscopy in African American individuals, mt-sDNA may offer a promising screening alternative in this patient population. ***Cancer* 2018;124:3876-3880**. © *2018 American Cancer Society.*

KEYWORDS: African Americans, colon polyps/diagnosis, colonoscopy, colorectal neoplasms/diagnosis, DNA analysis, early detection of cancer.

INTRODUCTION

Routine screening for colorectal cancer and adenomatous polyps among adults aged ≥50 years is recommended, and approximately 60% of the target population currently is up to date with screening, primarily through the use of colonoscopy.[1] However, screening rates are consistently lower among individuals from lower socioeconomic strata, which is attributed in part to the lack of a regular health care provider and health insurance.[1] Studies also have reported lower colon cancer screening rates among African American individuals, which may be attributed in part to socioeconomic status[2,3] as well as barriers such as fear of the diagnosis, mistrust of the health care system, and a lack of provider recommendation. In addition, there are important differences in the biology and clinical behavior of colon cancer in African American individuals compared with white individuals, with an earlier age at onset, a greater percentage of right-sided cancers, and unique genetic mutations.[3,4]

Given the significant number of individuals who are not up to date with colonoscopy screening, alternative screening procedures have been developed. These include stool-based tests such as fecal immunochemical testing (FIT), radiographic procedures such as computed tomography colonography, and other endoscopic procedures such as flexible igmoidoscopy. Another noninvasive testing option is multitarget stool DNA testing (mt-sDNA), which includes a panel of methylation markers, oncogenes, and FIT, reduced to a single patient result via an algorithm. Although mt-sDNA primarily was designed as a cancer screening test, it also detects a greater percentage of advanced, precancerous

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lesions compared with FIT alone. To the best of our knowledge, the largest colorectal cancer screening trial of mt-sDNA to date enrolled nearly 10,000 participants, 10.7% of whom were African American, but did not stratify results by race or ethnicity.[5] Thus, we performed a prospective cohort study to evaluate and compare the test performance of the commercially available mt-sDNA (Cologuard; Exact Sciences Corporation, Madison, Wisconsin) in African American and white individuals. Our goal was to determine whether the sensitivity of mt-sDNA in African American patients was comparable to that in white patients and thus it is an appropriate screening test in this traditionally underserved patient population.

MATERIALS AND METHODS

The study was conducted from 2012 to 2015 at 3 sites in metropolitan Cleveland, Ohio: 1) an urban, tertiary care academic medical center; 2) an affiliated suburban community hospital; and 3) an affiliated suburban ambulatory surgery center. The methods and results are reported in accordance with the STROBE (Strengthening the Reporting of Observational Studies in Epidemiology) guidelines.[6] Approval for the study was obtained from the University Hospitals institutional review board. Patients aged 40 to 80 years who were referred for colonoscopy by their health care providers were eligible for enrollment. Patients aged <50 years were largely referred because of a family history of colorectal cancer or nonspecific lower gastrointestinal symptoms such as constipation. Exclusion criteria were a known history of any malignancy, a prior history of adenomatous polyps or serrated neoplasia in the colon, previous colon resection, prior colonoscopy within 5 years, overt gastrointestinal bleeding, a diagnosis of ulcerative colitis or Crohn disease, or the inability to provide informed consent or understand English. In addition, for this analysis, we excluded patients who were of a racial group other than white or African American or were of unknown race.

Prior to undergoing colonoscopy and the bowel preparation, all patients collected stool for mt-sDNA that was processed according to a standard protocol as well as a commercially available FIT (OC-FIT CHEK; Polymedco CDP, LLC, Cortlandt Manor, New York). A questionnaire was completed prior to colonoscopy and included health risk factors including height and weight and whether the patient ever used tobacco products. Patients then underwent standard colonoscopy; the endoscopist was unaware of the mt-sDNA or FIT results but knew that the patient was enrolled in the study. All visible lesions were removed or, if not feasible, biopsied, and a positive test included findings of ≥1 adenomas, sessile serrated adenomas, or carcinomas. Advanced lesions were defined as an adenoma measuring ≥1 cm and/or containing high-grade dysplasia or adenocarcinoma. The location of the adenoma was divided into the rectum/rectosigmoid, left colon (sigmoid, descending, or splenic flexure), and right colon (transverse, hepatic flexure, ascending, or cecum) based on the colonoscopy report. All colonoscopies were performed by faculty endoscopists, all of whom met or exceeded established quality metrics.

Using the findings at colonoscopy as the reference standard, we determined the sensitivity, specificity, and area under the receiving operating characteristic (ROC) curve of both mt-sDNA and FIT for any adenoma and for advanced lesions. Test characteristics then were compared between white and African American patients.

RESULTS

A total of 844 patients agreed to participate in the current study. We excluded 57 patients for the following reasons: failure to collect a stool sample prior to colonoscopy (6 patients), patient did not keep appointment for colonoscopy (44 patients), and poor preparation at colonoscopy without rescheduling (7 patients). For this analysis, we further excluded 27 patients with a self-identified race other than African American or white (26 patients) or who were of unknown race (1 patient), leaving 760 subjects available for analysis.

The mean age of the cohort was 56.7 ± 8.0 years; 60.2% were female and 495 were white (65.1%) and 265 were African American (34.9%). Compared with white patients, African American individuals were more often female, but there was no difference noted with regard to the mean age (Table 1). The average body mass index was higher in African American patients and African Americans had a greater prevalence of smoking. One or more adenomas were found in 103 African American patients (38.9%) and 168 white patients (33.9%) (P = .36) and advanced lesions, including 2 cancers, were detected in 18 African American patients (6.8%) and 33 white patients (6.7%) (P = .12). Among patients with adenomas, there was a somewhat greater mean number of adenomas noted among African Americans. The distribution of adenomas in the colon also was similar between the groups and the prevalence of sessile serrated adenomas was comparable (Table 1).

TABLE 1. Clinical Characteristics of Study Participants

Characteristic	White n = 495	African American n = 265	P
Age (mean ± SD), y	56.6 ± 8.1	57.2 ± 8.0	.57
Female, no. (%)	282 (57.0)	176 (66.4)	.03
BMI (mean ± SD)	28.2 ± 6.0	32.2 ± 7.9	<.0001
Current or previous smoker, no. (%)	214 (43.2)	158 (59.6)	<.001
Adenomas, no. (%)	168 (33.9)	103 (38.9)	.36
No. of adenomas per patient (mean ± SD)[a]	1.7 ± 1.3	2.1 ± 1.6	.07
Advanced adenomas, no. (%)	33 (6.7)	18 (6.8)	.12
Distribution of adenomas[b]			
Rectum/rectosigmoid	18 (6.4)	26 (12.2)	.49
Left colon	81 (28.7)	72 (33.8)	.28
Right colon	183 (64.8)	115 (53.9)	.20
Sessile serrated adenomas, no. (%)	30 (6.1)	10 (3.8)	.14

Abbreviations: BMI, body mass index; SD, standard deviation.

[a]Mean number of adenomas among patients with at least 1 adenoma.

[b]Distribution among all adenomas found.

TABLE 2. Performance Characteristics of mt-sDNA and FIT According to Race

		Sensitivity		Specificity		ROC Curve Area		P	
		Advanced Lesions	Any Adenoma	Advanced Lesions	Any Adenoma	Advanced Lesions	Any Adenoma	Advanced Lesions	Any Adenoma
Overall	mt-sDNA	43%	19%	91%	93%	0.67	0.56		
Overall	FIT	32%	11%	97%	98%	0.64	0.54		
African American	mt-sDNA	50%	20%	92%	95%	0.71	0.57	.42	.48
White	mt-sDNA	39%	17%	91%	93%	0.65	0.55		
African American	FIT	35%	11%	97%	97%	0.66	0.54	.74	.98
White	FIT	33%	11%	97%	98%	0.64	0.54		

Abbreviations: FIT, fecal immunochemical test; mt-sDNA, multitarget stool DNA testing; ROC, receiver operating characteristic.

Test characteristics of mt-sDNA and FIT for the overall cohort, African American patients, and white patients are shown in Table 2. In general, mt-sDNA was more sensitive and less specific than FIT for all adenomas as well as advanced lesions. When stratified by race, the sensitivity, specificity, and ROC curve area for mt-sDNA were similar between African American and white patients (ROC curve area: $P = .42$ and $P = .48$, respectively, for advanced lesions and any adenoma). Similarly, the test characteristics for FIT did not differ by racial group (ROC curve area for advanced lesions and any adenoma: $P = .74$ and $P = .98$, respectively).

There were a total of 40 patients with ≥1 sessile serrated adenomas in the current study (Table 1). The sensitivity of mt-sDNA and FIT were 28% and 2%, respectively, and the corresponding specificities were 94% and 98%, respectively. For those sessile serrated adenomas that measured ≥1 cm, the sensitivities were 42% and 13%, respectively, and the specificities were 92% and 98%, respectively.

DISCUSSION

Although colorectal cancer is one of the most common causes of cancer mortality in the United States, both the incidence and mortality can be reduced through the use of screening with the detection of early-stage cancer and the removal of precursor adenomas. African American individuals are more likely to be diagnosed with and to die of colorectal cancer than any other racial group.[3,4,7,8] Moreover, since 1960, although the mortality rate for white individuals has declined by 39%, it has increased by 28% in African American individuals.[8] The incidence of colorectal cancer among African Americans also remains 15% to 23% higher than in white individuals and other racial groups.[7] Given the higher colorectal cancer

incidence and mortality in African American individuals as well as a somewhat lower population-based uptake of colonoscopy, the feasibility of alternative screening methods is worthy of investigation. Because there may be differences in tumor-related and genetic factors in colorectal cancer affecting African American individuals, it is important to identify any differences in the performance characteristics of screening tests across racial groups. In the current prospective study, which was comprised of one-third African Americans, we found that mt-sDNA testing performed equally as well as in other ethnic groups for detecting advanced adenomas or any adenomas. These findings, coupled with a previous study that demonstrated comparable test acceptability in African American patients and others,[9] suggest that mt-sDNA could be an effective approach to increasing colorectal cancer screening in African American individuals. In the previous study, the most commonly cited reasons for preferring mt-sDNA were the absence of bowel preparation, no loss of work, and the ease of the test.

Although to the best of our knowledge several studies to date have evaluated the ability of mt-sDNA to detect adenomas and advanced lesions, few have specifically examined its performance among specific racial or ethnic groups. In a prospective study of Alaska natives,[10] a population with lower access to colonoscopy, the sensitivity and specificity of mt-sDNA for detecting advanced lesions were 49% and 91%, respectively, which both were similar to those of a large, multicenter study.[5] Another recent study examined the uptake of colorectal cancer screening in Olmsted County, Minnesota,[11] where African American individuals comprise only 3.5% of the population. During a 12-month period, the use of both mt-sDNA and colonoscopy were lower in African Americans (6.3% and 1.5%, respectively) compared with the general population (8.1% and 3.6%, respectively). However, test characteristics were not compared between racial groups.

Although the current study included a modestly large sample of patients with a substantial percentage of African American individuals undergoing age-appropriate screening, we acknowledge some limitations. First, this was a single-center study in which patients were specifically recruited from a screening population. However, we have no a priori reason to question the generalizability of the findings to other centers and regions. Second, the mt-sDNA findings were reported as a single, qualitative positive or negative result, which is the reporting method in clinical practice. Thus, we were unable to ascertain whether the distribution of positive results for specific markers differed between racial groups. Third, both the American College of Gastroenterology[4] and US Multisociety Task Force on Colorectal Cancer[12] guidelines recommend the initiation of screening in African American individuals at age 45 years. However, due to sample size, we did not explicitly evaluate the test performance of mt-sDNA in African American patients aged 45 to 50 years. Finally, although colonoscopy was used as the reference standard for the calculation of the sensitivity and specificity of mt-sDNA, we recognize that it is imperfect and associated with a miss rate for both small and advanced lesions.[13] However, unless the adenoma detection rate at colonoscopy differed systematically between African American patients and others, the overall findings should not be biased. The absence of differences in lesion detection between white and African American patients also is less likely given the similar prevalence of serrated lesions and right-sided adenomas, both of which are potentially more difficult to visualize.

The results of the current study demonstrate similar test performance of mt-sDNA in African American individuals compared with members of other racial groups, suggesting its feasibility as a screening test in this traditionally underserved population.

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CONFLICT OF INTEREST DISCLOSURES

Sanford D. Markowitz has received royalties from Exact Sciences Corporation and has a licensed patent to Exact Sciences for Yes on Methylated Vimentin DNA, with royalties paid to Exact Sciences. Joseph E. Willis is associated with Lucid Diagnostics, a newly formed Barrett esophagus diagnostics company. In addition, Dr. Willis has a patent for Methylated markers for esophageal neoplasia licensed to Lucid Diagnostics with royalties paid to Case Western Reserve University. Barry M. Berger is a full-time employee of Exact Sciences Corporation, the manufacturer of the stool DNA technology used in the current study, and holds equity in the company.

AUTHOR CONTRIBUTIONS

Conceptualization: **Gregory S. Cooper**, **Sanford D. Markowitz**, **Dean E. Brenner**, and **Li Li**. Formal analysis: **Gregory S. Cooper**, **Zhengyi Chen**, and **Li Li**. Funding acquisition: **Gregory S. Cooper**, **Sanford D. Markowitz**, and **Li Li**. Investigation: **Gregory S. Cooper**, **Sanford D. Markowitz**, **Zhengyi Chen**, **Missy Tuck**, **Joseph E. Willis**, **Barry M. Berger**, **Dean E. Brenner**, and **Li Li**. Methodology: **Gregory S. Cooper**, **Sanford D. Markowitz**, and **Li Li**. Writing–original draft: **Gregory S. Cooper** and **Li Li**. Writing–review and editing: **Gregory S. Cooper**, **Sanford D. Markowitz**, **Joseph E. Willis**, **Barry M. Berger**, **Dean E. Brenner**, and **Li Li**.

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