

# Final Progress Report for Research Projects Funded by Health Research Grants

Instructions: Please complete all of the items as instructed. Do not delete instructions. Do not leave any items blank; responses must be provided for all items. If your response to an item is “None”, please specify “None” as your response. “Not applicable” is not an acceptable response for any of the items. There is no limit to the length of your response to any question. Responses should be single-spaced, no smaller than 12-point type. The report **must be completed using MS Word**. Submitted reports must be Word documents; they should not be converted to pdf format. Questions? Contact Health Research Program staff at 717-783-2548.

1. **Grantee Institution:** Thomas Jefferson University
2. **Reporting Period (start and end date of grant award period):** 06/01/12-08/29/14
3. **Grant Contact Person (First Name, M.I., Last Name, Degrees):** Carol Prem
4. **Grant Contact Person’s Telephone Number:** 215-955-1407
5. **Grant SAP Number:** 4100059197
6. **Project Number and Title of Research Project:** 1 - Occult Tumor Burden as a Marker Stratifying Therapy to Eliminate Racial Disparities in Colon Cancer
7. **Start and End Date of Research Project:** 06/01/12-08/29/14
8. **Name of Principal Investigator for the Research Project:** Scott A. Waldman, MD, PhD, FCP, FAHA
9. **Research Project Expenses.**

9(A) Please provide the total amount of health research grant funds spent on this project for the entire duration of the grant, including indirect costs and any interest earned that was spent:

\$ 744,156

9(B) Provide the last names (include first initial if multiple individuals with the same last name are listed) of **all** persons who worked on this research project and were supported with health research funds. Include position titles (Principal Investigator, Graduate Assistant, Post-doctoral Fellow, etc.), percent of effort on project and total health research funds expended for the position. For multiple year projects, if percent of effort varied from year to year, report in the % of Effort column the effort by year 1, 2, 3, etc. of the project (x% Yr 1; z% Yr 2-3).

Last Name, First Name	Position Title	% of Effort on Project	Cost
Waldman	Principal Investigator	10%	\$ 90,006.20
Hyslop	Co-Investigator	10%	34,406.90
Myers	Co-Investigator	5%	23,732.30
Andrel	Statistical Analyst	20%	37,674.76
Bonaccorso	Clinical Nurse Coordinator/Data Manager	12% Yr 1	23,215.06
Cocroft	Data Manager	5%	9,412.92
DiCarlo	Clinical Research Assistant	40%	58,451.57
Haaf	Clinical Nurse Coordinator	15% Yr 1; 8% Yr 2	31,701.48
Leong	Research Assistant	80% Yr 1; 95% Yr2	105,338.92
Haslam	Clinical Research Assistant	2% Yr 2	826.87
Palotto	CRU Director	5% Yr 2	6,359.18
Puchalski	Clinical Research Nurse	<1% Yr2	126.78
Pullaro	Clinical Res Coordinator	2% Yr 2	1,246.91
Snook	Researcher	8% Yr 2	5,345.54
Vizza	RN	2% Yr 2	1,476.56

9(C) Provide the names of **all** persons who worked on this research project, but who *were not* supported with health research funds. Include position titles (Research Assistant, Administrative Assistant, etc.) and percent of effort on project. For multiple year projects, if percent of effort varied from year to year, report in the % of Effort column the effort by year 1, 2, 3, etc. of the project (x% Yr 1; z% Yr 2-3).

Last Name, First Name	Position Title	% of Effort on Project
None		

9(D) Provide a list of **all** scientific equipment purchased as part of this research grant, a short description of the value (benefit) derived by the institution from this equipment, and the cost of the equipment.

Type of Scientific Equipment	Value Derived	Cost
None		

**10. Co-funding of Research Project during Health Research Grant Award Period.** Did this research project receive funding from any other source during the project period when it was supported by the health research grant?

Yes \_\_\_\_\_ No  X

If yes, please indicate the source and amount of other funds:

## 11. Leveraging of Additional Funds

11(A) As a result of the health research funds provided for this research project, were you able to apply for and/or obtain funding from other sources to continue or expand the research?

Yes  X  No \_\_\_\_\_

If yes, please list the applications submitted (column A), the funding agency (National Institutes of Health—NIH, or other source in column B), the month and year when the application was submitted (column C), and the amount of funds requested (column D). If you have received a notice that the grant will be funded, please indicate the amount of funds to be awarded (column E). If the grant was not funded, insert “not funded” in column E.

Do not include funding from your own institution or from CURE (tobacco settlement funds). Do not include grants submitted prior to the start date of the grant as shown in Question 2. If you list grants submitted within 1-6 months of the start date of this grant, add a statement below the table indicating how the data/results from this project were used to secure that grant.

A. Title of research project on grant application	B. Funding agency (check those that apply)	C. Month and Year Submitted	D. Amount of funds requested:	E. Amount of funds to be awarded:
GUCY2C-targeted adoptive T cell therapy to treat metastatic colorectal cancer	<input type="checkbox"/> NIH <input type="checkbox"/> Other federal (specify: _____) <input checked="" type="checkbox"/> Nonfederal source (specify: <u>PhRMA</u> )	Sept 2013	\$100,000	\$
GUCY2C-specific tolerance in colon cancer patients	<input type="checkbox"/> NIH <input type="checkbox"/> Other federal (specify: _____) <input checked="" type="checkbox"/> Nonfederal source (specify: <u>PhRMA</u> )	Feb 2014	\$100,000	\$100,000
GUCY2C-targeted adoptive T cell therapy for metastatic colon cancer	<input type="checkbox"/> NIH <input type="checkbox"/> Other federal (specify: _____) <input checked="" type="checkbox"/> Nonfederal source (specify: <u>AACR</u> )	Feb 2014	\$67,500	\$
GUCY2C-specific CD4+ T cell tolerance mechanisms and outcomes in tumor immunity	<input type="checkbox"/> NIH <input type="checkbox"/> Other federal (specify: _____) <input checked="" type="checkbox"/> Nonfederal source (specify: _____)	April 2014	\$100,000 (annual)	\$100,000

	<u>Margaret Q. Landenberger Research Foundation</u> )			
Elimination of Adoptive Cell Therapy (ACT) Toxicity by Hypoxic Regulation of Antigen Receptors	<input type="checkbox"/> NIH <input type="checkbox"/> Other federal (specify:_____) <input checked="" type="checkbox"/> Nonfederal source (specify: <u>WW Smith</u> )	June 2014	\$125,000	\$
GUCY2C-specific CD4+ T cell tolerance mechanisms and outcomes	<input checked="" type="checkbox"/> NIH <input type="checkbox"/> Other federal (specify:_____) <input type="checkbox"/> Nonfederal source (specify:_)	June 2014	\$232,500	\$
GUCY2C endocrine axis at the nexus of diet and hyperphagia in obesity	<input checked="" type="checkbox"/> NIH <input type="checkbox"/> Other federal (specify:_____) <input type="checkbox"/> Nonfederal source (specify:_)	June 2014	\$250,000	
Hormone suppression silencing GUCY2C is required for colorectal cancer	<input checked="" type="checkbox"/> NIH <input type="checkbox"/> Other federal (specify:_____) <input type="checkbox"/> Nonfederal source (specify:_)	July 2014	\$250,000	
Silencing the GUCY2C-guanylin paracrine hormone axis is required for colorectal cancer	<input type="checkbox"/> NIH <input checked="" type="checkbox"/> Other federal (specify: <u>DOD</u> ) <input type="checkbox"/> Nonfederal source (specify:_)	September 2014	\$150,000	
Elimination of adoptive cell therapy (ACT) toxicity by hypoxic regulation of antigen receptors	<input checked="" type="checkbox"/> NIH <input type="checkbox"/> Other federal (specify:_____) <input type="checkbox"/> Nonfederal source (specify:_)	October 2014	\$125,000	
Hypoxic regulation of antigen receptors in adoptive cell therapy	<input type="checkbox"/> NIH <input type="checkbox"/> Other federal (specify:_) <input checked="" type="checkbox"/> Nonfederal source (specify: <u>ACS</u> )	October 2014	\$125,000	
APC-β-catenin regulates the GUCY2C tumor suppressor axis in colorectal cancer	<input checked="" type="checkbox"/> NIH <input type="checkbox"/> Other federal (specify:_____) <input type="checkbox"/> Nonfederal source (specify:_)	October 2014	\$250,000	

The GUCY2C paracrine hormone axis at the intersection of radiation exposure and the GI syndrome	<input type="checkbox"/> NIH <input checked="" type="checkbox"/> Other federal (specify: <u>DOD</u> ) <input type="checkbox"/> Nonfederal source (specify:_)	October 2014	\$387,959	
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11(B) Are you planning to apply for additional funding in the future to continue or expand the research?

Yes   X   No \_\_\_\_\_

If yes, please describe your plans:

We will apply for funding to continue to explore the relationship between GUCY2C mRNA in lymph nodes, and response to chemotherapy in patients with colorectal cancer. These applications will focus on large prospective multi-center clinical trials to define this utility. We will apply to NCI for funding. In addition, we will apply to the company who has licensed this technology and will commercialize it, Targeted Diagnostics & Therapeutics, Inc. for additional funding to support ongoing analyses.

**12. Future of Research Project.** What are the future plans for this research project?

We will expand these studies into a prospective multicenter blinded clinical trial to define the utility of GUCY2C to identify stage I-II colorectal cancer patients who are at risk for developing metastatic disease and who could benefit from adjuvant chemotherapy.

**13. New Investigator Training and Development.** Did students participate in project supported internships or graduate or post-graduate training for at least one semester or one summer?

Yes \_\_\_\_\_ No   X  

If yes, how many students? Please specify in the tables below:

	Undergraduate	Masters	Pre-doc	Post-doc
Male				
Female				
Unknown				
<b>Total</b>				

	Undergraduate	Masters	Pre-doc	Post-doc
Hispanic				
Non-Hispanic				
Unknown				
<b>Total</b>				

	Undergraduate	Masters	Pre-doc	Post-doc
White				
Black				
Asian				
Other				
Unknown				
<b>Total</b>				

**14. Recruitment of Out-of-State Researchers.** Did you bring researchers into Pennsylvania to carry out this research project?

Yes \_\_\_\_\_ No X \_\_\_\_\_

If yes, please list the name and degree of each researcher and his/her previous affiliation:

**15. Impact on Research Capacity and Quality.** Did the health research project enhance the quality and/or capacity of research at your institution?

Yes X \_\_\_\_\_ No \_\_\_\_\_

If yes, describe how improvements in infrastructure, the addition of new investigators, and other resources have led to more and better research.

These studies permitted us to recruit new faculty (Adam Snook) who is spearheading these analyses as well as developing related immunological approaches to prevent metastatic colorectal cancer in patients. Further, they permitted us to recruit an expert technologist in quantitative RT-PCR analysis that is serving as an institutional resource for biomarker development. Also, these studies permitted us to form relationships with regional healthcare systems to establish new collaborations that will be leveraged to enhance research capacity. Moreover, these studies have led to the building of relationships with regional biotechnology organizations which license technologies from the institution, support ongoing research with extramural funding, and grow the regional workforce and jobs in the biotechnology sector.

**16. Collaboration, business and community involvement.**

16(A) Did the health research funds lead to collaboration with research partners outside of your institution (e.g., entire university, entire hospital system)?

Yes  No

If yes, please describe the collaborations:

We developed productive collaborations with Fox Chase Cancer Center, University of Pittsburgh Medical Center, Christiana Health System in Delaware, Virtua Health System in New Jersey, and Allegheny Health System in Pennsylvania. Moreover, these studies solidified a working relationship with the regional biotechnology organization, Targeted Diagnostics & Therapeutics, Inc.

16(B) Did the research project result in commercial development of any research products?

Yes  No

If yes, please describe commercial development activities that resulted from the research project:

The technology that was the focus of this project was licensed by Targeted Diagnostics & Therapeutics, Inc. In turn, this technology was sublicensed from them by DiagnoCure in Quebec, Canada for commercialization to identify stage I and II colorectal cancer patients who could benefit from adjuvant chemotherapy.

16(C) Did the research lead to new involvement with the community?

Yes  No

If yes, please describe involvement with community groups that resulted from the research project:

One of the projects explored the barriers to using molecular diagnostics in the clinical practice setting. This required us to reach out to community physicians who manage patients with colorectal cancer to define those barriers, and approaches to overcome them to permit molecular diagnostics technology to benefit patients.

### **17. Progress in Achieving Research Goals, Objectives and Aims.**

List the project goals, objectives and specific aims (as contained in the grant agreement). Summarize the progress made in achieving these goals, objectives and aims for the period that the project was funded (i.e., from project start date through end date). Indicate whether or not each goal/objective/aim was achieved; if something was not achieved, note the reasons why. Describe the methods used. If changes were made to the research goals/objectives/aims, methods, design or timeline since the original grant application was submitted, please describe the changes. Provide detailed results of the project. Include evidence of the data that was generated and analyzed, and provide tables, graphs, and figures of the data. List published abstracts, poster presentations and scientific meeting presentations

at the end of the summary of progress; peer-reviewed publications should be listed under item 20.

This response should be a DETAILED report of the methods and findings. It is not sufficient to state that the work was completed. Insufficient information may result in an unfavorable performance review, which may jeopardize future funding. If research findings are pending publication you must still include enough detail for the expert peer reviewers to evaluate the progress during the course of the project.

Health research grants funded under the Tobacco Settlement Act will be evaluated via a performance review by an expert panel of researchers and clinicians who will assess project work using this Final Progress Report, all project Annual Reports and the project's strategic plan. After the final performance review of each project is complete, approximately 12-16 months after the end of the grant, this Final Progress Report, as well as the Final Performance Review Report containing the comments of the expert review panel, and the grantee's written response to the Final Performance Review Report, will be posted on the CURE Web site.

**There is no limit to the length of your response. Responses must be single-spaced below, no smaller than 12-point type. If you cut and paste text from a publication, be sure symbols print properly, e.g., the Greek symbol for alpha ( $\alpha$ ) and beta ( $\beta$ ) should not print as boxes ( $\square$ ) and include the appropriate citation(s). DO NOT DELETE THESE INSTRUCTIONS.**

#### **Project goals, objectives and specific aims**

The significance of the project is highlighted by the populations at risk, which include 1.5 million patients worldwide with colorectal cancer and the associated racial disparities in outcomes. The clinical impact can be appreciated by considering that one of the greatest gaps in management of these patients is identifying who will respond to chemotherapy. The commercial impact is underscored by the market size for diagnostics predicting therapeutic response in colorectal cancer, which exceeds \$1.2 billion annually worldwide. The feasibility of this project reflects the innovation of the team in creating this field, its history of >10 years of collaboration, and the advantage of an existing population of patients, specimens and analyses which this project will leverage. In that context, the broad research objectives include: (1) defining the utility of occult tumor burden analysis by GUCY2C RT-qPCR (quantitative reverse transcriptase-polymerase chain reaction) to identify African American (black) and Caucasian (white) patients with early stage (lymph node-negative; pN0) colon cancer at excess risk for developing recurrent disease who benefit from therapy; (2) defining barriers that restrict the adoption of molecular diagnostic tests by practicing physicians and limit commercialization success; and (3) advancing commercialization of this novel molecular paradigm. These objectives will be accomplished by executing specific aims, which include: (1) the Clinical Development Aim, in which a retrospective multicenter clinical trial central to the commercialization plan will be conducted that defines the utility of GUCY2C RT-qPCR as a predictive marker of therapeutic response in pN0 black and white patients; (2) the Health Services Research Aim, which will identify physician barriers to adopting molecular diagnostic tests that could restrict commercialization; and (3) the Commercialization Aim, which will

advance commercial deployment of occult tumor burden as a molecular diagnostic test to identify pN0 colon cancer patients who benefit from adjuvant chemotherapy. Commercialization will occur through a well-established and successful public-private partnership between Thomas Jefferson University and Targeted Diagnostics & Therapeutics, Inc., a Pennsylvania-based biotechnology company with a track record of success in commercializing GUCY2C-based diagnostics for managing patients with colorectal cancer.

**A. Specific Aims.** This program provides the clinical validation for commercializing a molecular diagnostic that identifies lymph node (node)-negative (pN0) colon cancer patients who benefit from adjuvant chemotherapy (therapy). There is an unmet need to improve therapeutic management of colorectal cancer, the 4th leading cause of cancer and 2nd leading cause of cancer mortality worldwide. In Pennsylvania (PA), the burden of colorectal cancer is higher than in the rest of the country, with ~15,000 cases annually, costing >\$200M. Importantly, there are stage-specific disparities in pN0 African Americans (blacks), who exhibit ~40% excess mortality compared to Caucasians (whites). One limitation to (1) managing pN0 patients and (2) reducing racial disparities is the absence of markers predicting therapeutic response.

This proposal advances an emerging paradigm employing GUCY2C as a predictive marker to direct therapy in pN0 colon cancer. GUCY2C is a protein expressed normally by intestinal cells, but universally over-expressed by colorectal tumors. There is a relationship between the quantity of occult tumor burden in nodes estimated by GUCY2C RT-qPCR (quantitative reverse transcriptase-polymerase chain reaction) and prognosis in pN0 colorectal cancer. Moreover, disproportionate occult tumor burden contributes to racial disparities in pN0 colon cancer. Here, we will validate the utility of GUCY2C RT-qPCR to optimize therapy and reduce racial disparities in pN0 colon cancer. Goals include: (1) establishing the utility of occult tumor burden to identify black and white pN0 colon cancer patients who benefit from therapy; (2) defining barriers to adoption of molecular tests by physicians that limit commercialization; and (3) advancing commercialization of occult tumor burden analysis as a predictive molecular test. Goals will be accomplished through: (1) the Clinical Development Aim, in which a retrospective trial will establish GUCY2C RT-qPCR as a predictive marker in pN0 black and white patients; (2) the Health Services Research Aim, which will identify physician barriers to adopting molecular tests; and (3) the Commercialization Aim which will leverage the established public-private partnership between Thomas Jefferson University (Jefferson) and Targeted Diagnostics & Therapeutics, Inc. (TDT) to advance this diagnostic test into commercialization. Specific Aims include:

**Aim 1. Establishing the utility of GUCY2C RT-qPCR to predict therapeutic benefit in pN0 colon cancer (Clinical Development Aim).** We will demonstrate that occult tumor burden predicts therapeutic benefit in black and white pN0 colon cancer patients using a retrospective case-control study design.

**Aim 2. Defining barriers to adoption of molecular tests by physicians (Health Services Research Aim).** We will identify barriers to adoption of GUCY2C RT-qPCR as a molecular diagnostic test by medical oncologists for managing pN0 colon cancer patients.

### **Aim 3. Commercializing occult tumor burden analysis by GUCY2C RT-qPCR for therapeutic decision-making in pN0 colon cancer (Commercialization Aim).**

#### **Research Outcomes and Benefits**

Outcomes and benefits of this research include:

(1) The *Clinical Development Aim* will define the utility of quantifying occult tumor burden, utilizing GUCY2C RT-qPCR, to identify pN0 black and white colon cancer patients at risk for recurrent disease who derive benefit from adjuvant chemotherapy. This study will be the first to employ a molecular diagnostic to effectively identify pN0 colon cancer patients who benefit from adjuvant chemotherapy, one major unmet clinical gap in colon cancer management. Also, this study will demonstrate that pN0 patients with excess risk related to race can be identified and treated, to reduce racial disparities in outcomes in colon cancer. Moreover, this study is absolutely required to advance the commercialization of this molecular diagnostic platform for the therapeutic management of patients with pN0 colon cancer.

(2) The *Health Services Research Aim* will identify factors that influence the adoption of molecular diagnostic tests by practicing physicians. There is a substantial time lag between development and adoption of new medical technologies, including molecular diagnostics, for use in routine clinical care. Delay in translating innovations into practice is one primary hurdle to commercial success for molecular diagnostic products. Although these issues have been explored for other diagnostic products, there are no data on the uptake of molecular diagnostics to stage and treat pN0 colon cancer patients. We will complete structured interviews with practicing physicians, using established decision analysis methods, to assess barriers to physician adoption of molecular diagnostics for staging pN0 colon cancer patients. Interview data will be used to identify specific issues underlying resistance to adoption, and to develop outreach strategies which inform the commercialization plan for GUCY2C RT-qPCR.

(3) The *Commercialization Aim* will leverage the studies described herein as an essential element in the strategy to commercialize occult tumor burden quantified by GUCY2C RT-qPCR to stratify therapeutic responsiveness in pN0 colon cancer. One of the greatest barriers to adoption of molecular tests by practicing physicians is the absence of clinical trials that are adequately powered and robust in statistical analysis that conclusively demonstrate clinical utility. The central component of the present project, the retrospective analysis of the predictive utility of GUCY2C RT-qPCR in black and white pN0 colon cancer patients (Aim 1), will provide the analytical and clinical validation of the utility of this diagnostic platform in patient management. Beyond the impact of this project on physician adoption, the present study is significant because it will demonstrate the utility of GUCY2C RT-qPCR in providing clinically actionable data defining management of pN0 colon cancer patients. The present project will provide the base of evidence for the use of GUCY2C RT-qPCR in clinical management decisions that are essential to promote physician adoption and reimbursement by third party payers, including Medicare, that are key to commercial success.

## **Summary of Research Completed**

### Aim 1. Establish The Utility Of GUCY2C RT-qPCR To Predict Therapeutic Benefit In Black And White Pn0 Colon Cancer Patients.

This study is a retrospective case-control analysis, of patients diagnosed  $\geq 5$  y previously, with stage I and II colon cancer in  $>12$  nodes. Cohorts consist of populations of both treated and untreated black and white patients, to evaluate the efficacy of quantitative GUCY2C real-time reverse-transcription PCR (RT-qPCR) as a predictive marker of the benefits of therapy. The study enables correlation of clinical outcomes with occult metastases, and reflects the variation in therapy in pN0 colon, excluding the rectum. Node availability reflects the current clinical standard and recent studies, demonstrating the importance of adequate nodal collections for molecular analysis. Patients were selected with  $\geq 5$  y of stable, disease-free survival, as the agreed upon clinical boundary. Cohorts were structured so that samples are available to assess the impact of GUCY2C according to race.

Requisite agreements were established with new collaborating clinical centers, including Methodist Hospital in Philadelphia, Virtua Health System in Southern New Jersey, and Christiana Health System which captures nearly the entire population of the State of Delaware, and we obtained the requisite Institutional Review Board approvals from these sites. This permits the accrual of a sufficient number of pN0 colon cancer patients who received therapy to support this analysis. The database has been revised to incorporate these centers and patients, and we continue to verify and validate the system for integrating clinical, demographic and molecular information. This study has identified  $\sim 200$  patients for analysis. We continue to obtain lymph nodes from these patients, and their relevant clinical and demographic data. Acquired tissue blocks from eligible patients are being assessed for their GUCY2C levels by RT-qPCR, to estimate occult tumor burden across the regional lymph node network. This study continues with the assessment of  $\sim 2,400$  lymph nodes for evidence of occult metastatic disease. These analyses are being supported by a grant from the licensee, Targeted Diagnostics & Therapeutics, Inc. This licensing agreement for commercialization, and the resources to continue these analyses are a key deliverable of this program.

### Aim 2. Defining Barriers To Adoption Of Molecular Tests By Physicians.

Initially, the research team planned to identify physician barriers to the adoption of genomic (GUCY2C) testing to identify early stage (pN0) colon cancer patients. This plan involved administering a survey questionnaire to 50 practicing medical oncologists who treat early stage colon cancer patients in order to identify motivational factors that were associated with physician intention to use GUCY2C testing. We discovered, however, that as medical oncologists and gastrointestinal (GI) surgeons were likely to encounter pN0 colon cancer patients, both types of clinicians would have the opportunity to recommend GUCY2C testing. Therefore, we decided to broaden the survey target population to include medical oncologists and GI surgeons. In addition, the emerging literature on genomic testing in clinical care suggests that physician receptivity to ordering such testing may be influenced not only by motivational factors intrinsic to clinicians, but also by the availability of and

access to such testing at their affiliated institutions. We planned to learn about factors that would influence genomic test availability and access by conducting semi-structured interviews with a sample of practicing pathologists who are responsible for analyzing patient biological specimens and for communicating test results to treating clinicians at Thomas Jefferson University. In order to remain within budgetary constraints of the project, we decided to modify the initial data collection plan, and administer at least 40 physician surveys and complete up to five pathologist interviews.

### **Medical Oncologist and GI Surgeon Surveys.**

To address the goal of administering a survey questionnaire to clinicians, we initially obtained a mailing list of 211 medical oncologists and surgeons practicing in the Greater Philadelphia area that included clinician name, mailing address, and telephone number. Following established methods, we sent individuals on the list a mailing that included an introductory letter that described the purpose of the study and invited response via provision of written consent, and completion and return of an enclosed survey questionnaire or completion of an online version of the survey. The mailing also advised the recipient that s/her would be compensated (\$125) for completion of the survey, and included a federal W-9 form for completion and return in a postage-paid return envelope. We also included a postcard that allowed the recipient to opt-out of the survey that could be sent back in the return envelope. About a month after this initial survey mailing, the research team sent non-respondents a reminder that included a cover letter, another copy of the survey questionnaire, an opt-out card, and a return envelope. About 60 days later, the research team attempted to contact non-respondents by telephone to encourage response. **Figure 1** shows that after exclusion of individuals with inaccurate mailing address and/or telephone number (n=60), who did not currently see pN0 colon cancer patients (n=45), and who were deceased (n=2), this effort resulted in the identification of 104 physicians who were eligible and available to complete the survey. Of this number, 43 (41%) physicians completed the survey, 18 (18%) declined to participate, and 43 (41%) were lost to follow-up.

In accordance with the Complete Diagnostic Evaluation (CDE) Model, which we have used in prior research, the physician survey included items on clinician practice environment (e.g., hospital-versus community-based practice), physician background (e.g., sociodemographic characteristics), physician experience (exposure to pN0 colon cancer patients), and current practice used in staging (e.g., use of histopathology versus genomic testing in disease staging for pN0 colon cancer patients). We assessed physician perceptions about staging and treatment by asking the extent to which they thought histopathology and genomic testing were in staging pN0 colon cancer patients (very accurate, somewhat accurate, not accurate, don't know). We also asked physicians to respond to statements about stress related to making treatment recommendations for pN0 colon cancer patients, and their confidence in recommendation of treatment to those patients. Items using a Likert-type response set (Strongly Disagree – 1 to Strongly Agree -5) were used to elicit responses. Stress and confidence index scores were computed, along with the scores for constituent items. Physician approach to staging and treatment planning was measured in terms of the types of testing currently being used to stage pN0 colon cancer patients they see. Using the same type of response set, we also asked physicians to indicate their readiness to incorporate GUCY2C

testing into treatment planning, that is, “I would treat patients with pN0 colon cancer who have abnormal GUCY2C test results much more aggressively than patients with a normal test result.”

To assess physician receptivity to genomic testing (GUCY2C) for pN0 colon cancer patients – the primary dependent variable, we asked physicians if they agreed or disagreed with the following statements: “I think that all patients with pN0 colon cancer should have a GUCY2C test.” and “I think that GUCY2C test results should be considered when treatment is recommended for pN0 colon cancer patients.” Respondents were provided with Likert-type responses for these items that ranged from “strongly disagree” (1) to “strongly agree” (5). Responses to these items were summed and averaged to arrive at an overall score. Scores were then categorized into “Agree” (>3) and “Not Agree” (<= 3) categories.

We also asked physicians to respond to two open-ended questions by writing in those factors that would encourage them to or discourage them from ordering GUCY2C testing. These responses were categorized using standard content analysis methods, and category frequencies were computed. Respondents reported that environmental (e.g., documented predictive value, acceptance by medical community), patient-related (e.g., younger age, early stage disease, patient fitness), or test-related (e.g., low cost, high accuracy, ready availability, ease of interpretation) factors would encourage them to order the test. Physicians also reported that environmental (e.g., lack of efficacy data, limited peer adoption), patient-related (e.g., older age, late stage disease, patient frailty), or test-related (e.g., high cost, poor accuracy, low availability, difficulty of use) factors would discourage their use of the test.

Finally, summary statistics were calculated. Specifically, frequencies and percentages for categorical variables and means and standard deviation for continuous variables were determined. Fisher’s Exact testing was used to assess statistically significant associations between categorical variables and the dependent variable; and the Wilcoxon test was used to identify continuous variable differences relative to the outcome. Covariates associated with the outcome variable at the  $p \leq 0.2$  level were included in a multivariable logistic regression model. Backwards selection was used to determine the model, with retention of those independent variables that were associated at p-value of 0.05. Because of the small sample size, exact p-values are reported.

**Table 1** shows that in terms of practice environment, most survey respondents (77%) said that their practice was hospital based. Physician background and experience measures were distributed as follows: male (84%), white (69%), and  $\geq 50$  years of age (49%). In terms of experience, respondents reported: < 20 years in practice (37%);  $\geq 5$  pN0 patients seen in past year (86%), and  $\geq 1$  pN0 patients seen in past year with recurrence (53%). Regarding current practice in staging, 56% of respondents reported that they relied on histopathology, while 44% used histopathology combined with genomic testing. Data on physician perceptions about staging showed that only 14% of physicians thought that histopathology staging was a “very accurate” method for colon cancer staging, and the same proportion thought that genomic testing was a very accurate method of staging; while a surprising 64% reported, that combined histopathology and genomic testing was very accurate. Interestingly, 67%, 61%, and 91% of respondents, respectively, agreed that these approaches to staging provided them

with sufficient information to recommend a treatment plan for their pN0 colon cancer patients. However, 49% of respondents reported that deciding on the type of treatment to recommend for these patients was stressful. When asked about receptivity to GUCY2C testing, 45% of respondents reported that they thought all pN0 colon cancer patients should be tested in this manner, and 51% indicated that GUCY2C results should be considered when treatment is recommended for these patients. Finally, 40% of physicians said that they would treat patients with an abnormal GUC2YC result more aggressively than those with a normal GUCY2C result.

In univariable analyses (**Table 1**), we found several variables to be positively associated ( $p \leq 0.20$ ) with physician receptivity to ordering GUCY2C testing. These variables include practice environment ( $p=0.002$ ); physician perceptions about GUCY2C testing (i.e., belief that GUCY2C testing provides information needed to develop a treatment plan) ( $p=0.031$ ); anxiety about recommending treatment for pN0 colon cancer patients ( $p=0.098$ ); ease of making treatment decisions for pN0 colon cancer patients ( $p=0.009$ ); belief in GUCY2C test accuracy and combined histopathology and GUCY2C test accuracy ( $p=0.124$  and  $p=0.039$ , respectively); and readiness to treat patients with abnormal GUCY2C results aggressively ( $p=0.029$ ).

Multivariable analysis results displayed in **Table 2** show that physicians who considered making treatment decisions for pN0 colon cancer patients to be easy were significantly less receptive to GUCY2C testing than those who did not (OR=0.04, CI: 0.004, 0.30). Physicians who said they that would treat patients with abnormal GUCY2C test aggressively were significantly more receptive to GUCY2C testing (OR=16.91, CI: 2.62, 109.11).

**Table 3** shows that that the most commonly reported factor that would encourage physicians to order GUCY2C testing is the belief that such testing was more accurate than histopathology (51%). The most frequently mentioned factor that would discourage physicians from ordering GUCY2C testing is their concern about the limited evidence currently available on test accuracy (52%).

### **Pathologist Interviews.**

Ronald E. Myers, PhD initially contacted a senior pathologist in the Department of Pathology, Thomas Jefferson University Hospital to initiate the process of identifying and interviewing pathologists who have had experience working with medical oncologists and GI surgeons on diagnostic testing and staging of colon cancer patients. Prior to this initial encounter, the research team had developed a semi-structured interview guide for use in the planned interviews. The interview guide included the following questions: What Type of Test is GUCY2C? What Factors are Likely to influence Use of GUCY2C? How Would Results of GUCY2C Affect Treatment Planning? Participating pathologists were compensated with \$125 for completing the interview.

The senior pathologist volunteered that he had had such experience and identified four other pathologists with similar experience. Dr. Myers was able to arrange and conduct an in-person interview with each consenting pathologist. During the interviews, Dr. Myers posed

each question and kept written notes of responses. Following each interview, Dr. Myers reviewed the interview notes, produced a written summary of respondent comments, and categorized emergent themes reflected in those comments. Findings from this analysis are summarized below.

Tumor histology categorizes the cancer into categories that include: adenocarcinoma (most colon cancers), epidermoid carcinomas, or other rarer types of cancer. Histological examination of the tumor also determines the grade level of the cancer. Well-differentiated tumors are graded higher than moderately and low-differentiated tumors. Tumor grade is an important factor that will determine the prognosis of the cancer. In clinical medicine, “histopathology” refers to the examination of a biopsy or surgical specimen by a pathologist, after the specimen has been processed and sections of the specimen have been placed onto slides. Genomic tests can detect changes in chromosomes, genes, or proteins that may not be observed on standard histologic analysis, and may also be used to determine disease stage. GUCY2C is a gene on chromosome 12p12 that encodes guanylate cyclase 2C, a receptor for E coli heat-stable enterotoxin. GUCY2C testing may be used in staging colon cancer patients.

Standard histopathologic analysis of patient biological material is done as “reflex” testing. Such testing is triggered automatically, because it is included on the list of tests considered to be standard care and medically actionable. Medically actionable tests provide information that not only help to determine a patient’s prognosis, but also can be useful in directing treatment that is in accordance with accepted guidelines. Genomic testing is an example of “discretionary” testing that may or may not be incorporated into treatment guidelines, and may or may not be medically actionable.

GUCY2C testing is currently a discretionary “prognostic” genomic test for pN0 colon cancer patients. That is, GUCY2C testing is intended to provide information about patient risk for recurrence that clinicians can use to decide on treatment. It is not a “predictive” genomic test (e.g., KRAS and BRAF) that is used to determine whether a patient is or is not likely to respond to a given therapeutic intervention.

While both medical oncologists and GI surgeons may order genomic testing, medical oncologists are more likely than GI surgeons to do so in order to obtain information that can inform the decision as to whether to recommend chemotherapy. One pathologist commented that for most GI surgeons, “After surgery, they are done.” Thus, to some degree the likelihood that GUCY2C testing is considered for use among pN0 patients is influenced by whether these patients are referred to a medical oncologist following surgery.

Physician ordering of the GUCY2C is likely to be influenced a number of factors. If GUCY2C is incorporated into clinical guidelines, the test would be considered to be a reflex test, and would be ordered routinely for all pN0 colon cancer patients. GUCY2C is currently a discretionary test. Therefore, individual physicians would have to make the decision about whether to order the test. That decision will be influenced by the strength of data on test efficacy reported in the literature, clinical experience with patients who have had a

recurrence, determination that test results provide information that is medically actionable, use of the test by professional colleagues, and demand by patients. Other factors that are likely to influence physician use of the test are cost (especially cost to the patient), timeliness of results (which is influenced by whether the test can be done in-house or must be sent out for analysis), and the availability of bio-specimen material for conducting the test (whether testing is done on fresh tissue or tissue blocks).

It should be noted that pathologists are willing and able to perform both reflex and discretionary tests that are ordered. However, the performance of discretionary tests is viewed as somewhat more problematic than reflex tests. That is, discretionary tests may require the assignment of technical staff to collect and process biological material for analysis. Discretionary tests done in house may require complex analyses that take more time and effort than reflex testing. Discretionary tests that are sent outside the institution involve the assignment of staff to manage the logistics of material processing for delivery to a third party for analysis. Furthermore, pathologists and technical staff are not compensated directly for the additional services that are associated with discretionary test use in house or outside the institution.

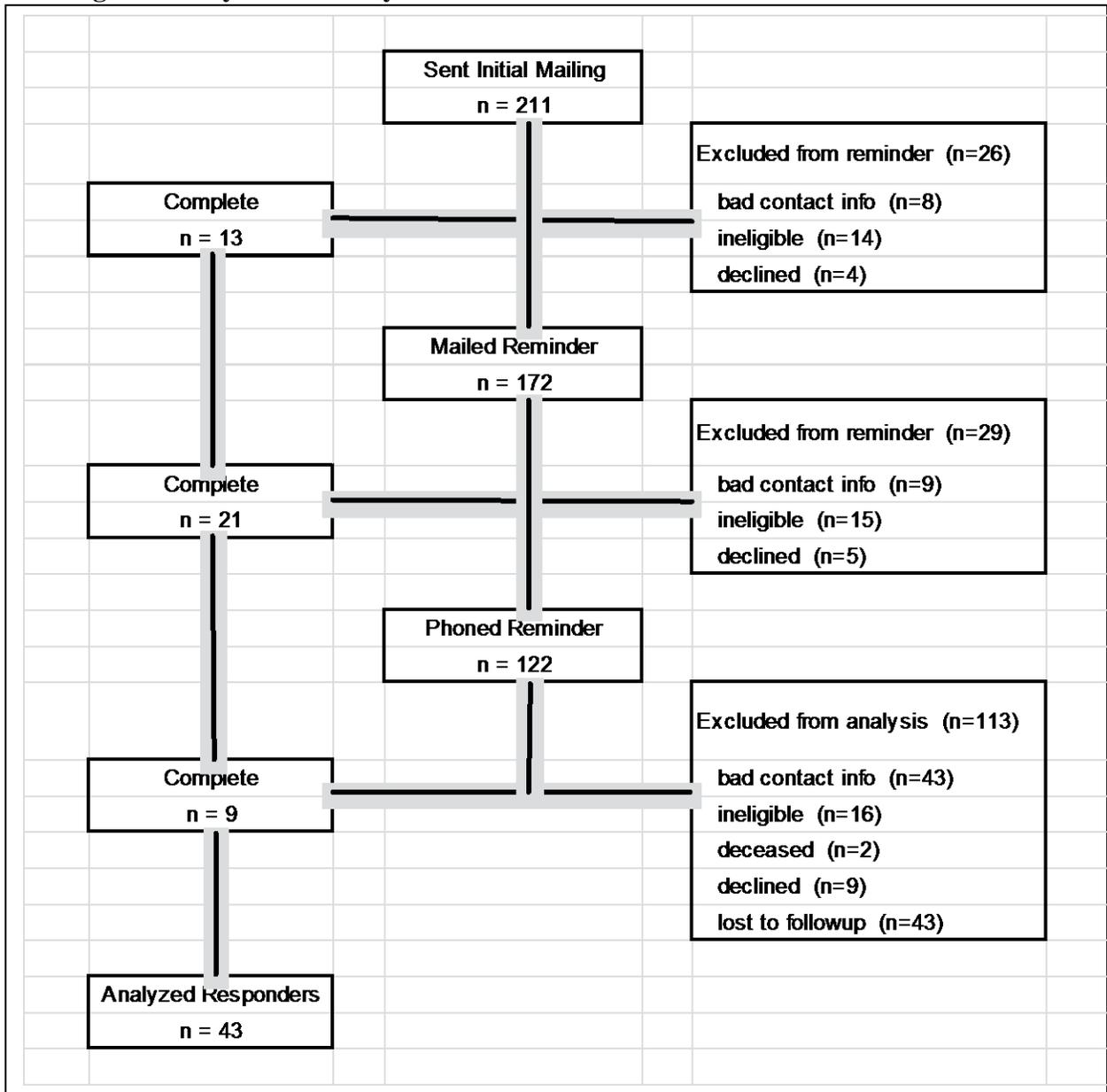
It was noted that medically actionable tests are more likely to be viewed favorably by pathologists than tests that are not. For the latter type of test, clinicians may find themselves in an ambiguous legal situation if test results are abnormal and no clinical action is taken. Furthermore, pathologists are not compensated for processing biological materials and sending those materials outside the institution for analysis. It was also noted that testing that is done on an in-patient basis is compensated at a set rate in accordance with standard institutional policy. However, pathologists can bill for test services when testing is done on an out-patient basis

Finally, respondents suggested that the adoption of GUCY2C testing would be increased if (1) there were strong scientific evidence supporting test efficacy, (2) clinicians and institution tumor board members were educated about the test, (3) testing was inexpensive, (4) testing could be done in-house, (5) testing and test interpretation were simple, and (6) test results were available quickly. Increased public awareness of the test may also promote GUCY2C adoption.

**Aim 3. Commercializing occult tumor burden analysis by GUCY2C RT-qPCR for therapeutic decision-making in pN0 colon cancer.**

The technology to quantify occult tumor burden across the regional lymph node network as a predictive marker of response to chemotherapy in Stage I-II colorectal cancer patients was licensed by Targeted Diagnostics & Therapeutics, Inc. They are providing grant funding to complete the analyses initiated under this proposal. Moreover, they are supporting the costs of patent prosecution related to these technologies. In turn, Targeted Diagnostics & Therapeutics sublicensed the technology to DiagnoCure in Quebec, Canada for commercialization.

**Figure 1. Physician Survey Schema**



**Table 1. Univariable Analyses of Physician Receptivity to Ordering Genomic Risk Assessment (GUCY2C) for pN0 Colon Cancer Patients**

	<b>Total (N=43) n (%)</b>	<b>Receptive (n=24) n (%)</b>	<b>Not Receptive (n=19) n (%)</b>	<b>p-value</b>
<b>Practice Environment:</b>				
<b>How would you describe your practice setting?</b>				<b>0.002</b>
Community-based or Other	25 (58.14)	9 (36.00)	16 (64.00)	
Hospital-based	18 (41.86)	15 (83.33)	3 (16.67)	
<b>Physician Background:</b>				
<b>Age</b>				1.000
<50	22 (51.16)	12 (54.55)	10 (45.45)	
≥50	21 (48.84)	12 (57.14)	9 (42.86)	
<b>Gender</b>				0.211
Male	36 (83.72)	22 (61.11)	14 (38.89)	
Female	7 (16.28)	2 (28.57)	5 (71.43)	
<b>Race</b>				1.000
Asian	13 (30.95)	7 (53.85)	6 (46.15)	
White	29 (69.05)	17 (58.62)	12 (41.38)	
<b>Hispanic/Latino</b>				0.442
No	42 (97.67)	24 (57.14)	18 (42.86)	
Yes	1 (2.33)	0 (0.00)	1 (100.00)	
<b>Years in Practice</b>				0.817
<10	5 (11.63)	3 (60.00)	2 (40.00)	
11-20	11 (25.58)	5 (45.45)	6 (54.55)	
20+	27 (62.79)	16 (59.26)	11 (40.74)	
<b>Board Certified</b>				--
No	0 (0.00)	0 (0.00)	0 (0.00)	
Yes	43 (100.00)	24 (55.81)	19 (44.19)	
<b>Physician Experience:</b>				
<b>During the past 12 months, how many newly diagnosed pN0 colon cancer patients have you personally seen in your practice?</b>				0.678
<5	6 (13.95)	4 (66.67)	2 (33.33)	
≥5	37 (86.05)	20 (54.05)	17 (45.95)	
<b>During the past 12 months, approximately what percentage of your pN0 colon cancer patients have had a recurrence?</b>				0.240
None	20 (46.51)	11 (55.00)	9 (45.00)	
>1	23 (53.49)	13 (56.52)	10 (43.48)	
<b>On average how many pN0 colon cancer patients do you see each month?</b>				0.509
<5	30 (69.77)	18 (60.00)	12 (40.00)	
≥5	13 (30.23)	6 (46.15)	7 (53.85)	

**Physician Perceptions About Staging and Treatment:**

<b>I believe that staging by histopathology can provide the information I need to recommend a treatment plan.</b>				1.000
Not Agree	14 (32.56)	8 (57.14)	6 (42.86)	
Agree	29 (67.44)	16 (55.17)	13 (44.83)	
<b>I think that molecular diagnostic testing can provide the information I need to recommend a treatment plan</b>				0.209
Not Agree	17 (39.53)	7 (41.18)	10 (58.82)	
Agree	26 (60.47)	17 (65.38)	9 (34.62)	
<b>I believe that combined staging by histopathology and molecular testing can provide the information I need to recommend a treatment plan</b>				<b>0.031</b>
Not Agree	4 (9.30)	0 (0.00)	4 (100.00)	
Agree	39 (90.70)	24 (61.54)	15 (38.46)	
<b>Not being sure of appropriate treatment for patients is stressful</b>				0.745
Not Agree	14 (32.56)	7 (50.00)	7 (50.00)	
Agree	29 (67.44)	17 (58.62)	12 (41.38)	
<b>I am concerned that I might be held accountable for the consequences of treatment</b>				0.543
Not Agree	22 (51.16)	11 (50.00)	11 (50.00)	
Agree	21 (48.84)	13 (61.90)	8 (38.10)	
<b>I feel anxious, because I am not sure that treatment to recommend</b>				0.098
Not Agree	30 (69.77)	14 (46.67)	16 (53.33)	
Agree	13 (30.23)	10 (76.92)	3 (23.08)	
<b>I feel uncomfortable about making a strong recommendation for treatment</b>				0.500
Not Agree	31 (72.09)	16 (51.61)	15 (48.39)	
Agree	12 (27.91)	8 (66.67)	4 (33.33)	
<b>I worry about malpractice in treatment</b>				0.523
Not Agree	29 (67.44)	15 (51.72)	14 (48.28)	
Agree	14 (32.56)	9 (64.29)	5 (35.71)	
<b>Deciding what to recommend when I talk about treatment to my patients is easy for me</b>				<b>0.009</b>
Not Agree	14 (32.56)	12 (85.71)	2 (14.29)	
Agree	29 (67.44)	12 (41.38)	17 (58.62)	
<b>I am sure about what to recommend when I talk to my patients about treatment</b>				0.500
Not Agree	12 (27.91)	8 (66.67)	4 (33.33)	
Agree	31 (72.09)	16 (51.61)	15 (48.39)	
<b>It is clear what treatment choice is right for my patients</b>				0.760
Not Agree	20 (46.51)	12 (60.00)	8 (40.00)	
Agree	23 (53.49)	12 (52.17)	11 (47.83)	
<b>I would like to have a more accurate way of determining risk for recurrence for my patients</b>				1.000
Not Agree	2 (4.65)	1 (50.00)	1 (50.00)	
Agree	41 (95.35)	23 (56.10)	18 (43.90)	
<b>I am not satisfied with the accuracy of current approaches for staging my patients</b>				0.230
Not Agree	18 (41.86)	8 (44.44)	10 (55.56)	

Agree	25 (58.14)	16 (64.00)	9 (36.00)	
<b>Stress (combined scale), mean(sd)</b>	3.09 (0.70)	3.19 (0.06)	2.97 (0.80)	0.362
<b>Stress (combined scale)</b>				0.223
Not Agree (<=3)	22 (51.16)	10 (45.45)	12 (54.55)	
Agree (>3)	21 (48.84)	14 (66.67)	7 (33.33)	
<b>Confidence (combined scale), mean(sd)</b>	3.80 (0.51)	3.74 (0.49)	3.87 (0.54)	0.275
<b>Confidence (combined scale)</b>				1.000
Not Agree (<=3)	3 (6.98)	2 (66.67)	1 (33.33)	
Agree (>3)	40 (93.02)	22 (55.00)	18 (45.00)	
<b>Perceived accuracy of tests used in staging</b>				
<b>Histopathology alone</b>				0.395
Very Accurate	6 (14.29)	3 (50.00)	3 (50.00)	
Somewhat Accurate	33 (78.57)	18 (54.55)	15 (45.45)	
Not Accurate	3 (7.14)	3 (100.00)	0 (0.00)	
Don't Know	0 (0.00)	0 (0.00)	0 (0.00)	
<b>GUCY2C testing alone</b>				0.124
Very Accurate	6 (14.63)	4 (66.67)	2 (33.33)	
Somewhat Accurate	24 (58.54)	15 (62.50)	9 (37.50)	
Not Accurate	3 (7.32)	3 (100.00)	0 (0.00)	
Don't Know	8 (19.51)	2 (25.00)	6 (75.00)	
<b>Combined Histopathology and GUCY2C testing</b>				<b>0.039</b>
Very Accurate	27 (64.29)	18 (66.67)	9 (33.33)	
Somewhat Accurate	11 (26.19)	6 (54.55)	5 (45.45)	
Not Accurate	0 (0.00)	0 (0.00)	0 (0.00)	
Don't Know	4 (9.52)	0 (0.00)	4 (100.00)	
<b>Physician Approach to Staging and Treatment Planning:</b>				
<b>Which do you most often rely on to plan pN0 treatment?</b>				0.217
Histopathology alone	24 (55.81)	11 (45.83)	13 (54.17)	
Histopathology and GUCY2C	19 (44.19)	13 (68.42)	6 (31.58)	
<b>Physician Readiness to Incorporate GUCY2C Testing into Treatment Planning:</b>				
<b>I would treat patients with pN0 colon cancer who have abnormal GUCY2C test results much more aggressively than patients with a normal molecular diagnostic test result</b>				<b>0.029</b>
Not Agree	25 (59.52)	10 (40.00)	15 (60.00)	
Agree	17 (40.48)	13 (76.47)	4 (23.53)	
<b>Physician Receptivity to Genomic (GUCY2C) Testing:</b>				
<b>I think GUCY2C test results should be considered when treatment is recommended</b>				--*
Not Agree	21 (48.84)	3 (14.29)	18 (85.71)	
Agree	22 (51.16)	21 (95.45)	1 (4.55)	
<b>I think that all patients with pN0 colon cancer should have a GUCY2C test</b>				--*
Not Agree	23 (54.76)	4 (17.39)	19 (82.61)	
Agree	19 (45.24)	19 (100.00)	0 (0.00)	

\*These variables were averaged to create the receptivity outcome variable; as such only descriptive statistics are provided.

**Table 2. Multivariable Analyses of Physician Receptivity to Ordering Genomic Risk Assessment (GUCY2C) for pN0 Colon Cancer Patients**

	<b>Odds Ratio</b>	<b>95% Confidence Interval</b>	<b>exact p-value</b>
<b>Physician Perceptions About Staging and Treatment:</b>			
<b>Deciding what to recommend when I talk about treatment to my patients is easy for me:</b>			
Agree vs. Not Agree	0.04	(0.004, 0.30)	0.001
<b>Physician Readiness to Incorporate GUCY2C Testing into Treatment Planning:</b>			
<b>I would treat patients with pN0 colon cancer who have abnormal GUCY2C test results much more aggressively than patients with a normal molecular diagnostic test result:</b>			
Agree vs. Not Agree	16.91	(2.62, 109.11)	0.002

**Table 3. Descriptive Statistics for Reported Physician Factors Influencing GUCY2C Testing**

	<b>Total (N=43) n (%)</b>	<b>Receptive (n=24) n (%)</b>	<b>Not Receptive (n=19) n (%)</b>
<b>Physician Decision Factors:</b>			
<b>Factors that would encourage physicians to order GUCY2C testing:</b>			
Environment	6 (13.95)	3 (50.00)	3 (50.00)
Patient	11 (25.58)	8 (72.73)	3 (27.27)
Physician	5 (11.63)	5 (100.00)	0 (0.00)
Practice	10 (23.26)	7 (70.00)	3 (30.00)
Test	33 (76.74)	16 (48.48)	17 (51.52)
<b>Factors that would discourage physicians from ordering GUCY2C testing:</b>			
Environment	3 (6.98)	1 (33.33)	2 (66.67)
Patient	14 (32.56)	10 (71.43)	4 (28.57)
Physician	7 (16.28)	6 (85.71)	1 (14.29)
Practice	6 (13.95)	4 (66.67)	2 (33.33)
Test	33 (76.74)	16 (48.48)	17 (51.52)

**18. Extent of Clinical Activities Initiated and Completed.** Items 18(A) and 18(B) should be completed for all research projects. If the project was restricted to secondary analysis of clinical data or data analysis of clinical research, then responses to 18(A) and 18(B) should be “No.”

18(A) Did you initiate a study that involved the testing of treatment, prevention or diagnostic procedures on human subjects?

Yes  
 No

18(B) Did you complete a study that involved the testing of treatment, prevention or diagnostic procedures on human subjects?

Yes  
 No

**If “Yes” to either 18(A) or 18(B), items 18(C) – (F) must also be completed.** (Do NOT complete 18(C-F) if 18(A) and 18(B) are both “No.”)

18(C) How many hospital and health care professionals were involved in the research project?

\_\_\_\_\_ Number of hospital and health care professionals involved in the research project

18(D) How many subjects were included in the study compared to targeted goals?

\_\_\_\_\_ Number of subjects originally targeted to be included in the study  
\_\_\_\_\_ Number of subjects enrolled in the study

**Note:** Studies that fall dramatically short on recruitment are encouraged to provide the details of their recruitment efforts in Item 17, Progress in Achieving Research Goals, Objectives and Aims. For example, the number of eligible subjects approached, the number that refused to participate and the reasons for refusal. Without this information it is difficult to discern whether eligibility criteria were too restrictive or the study simply did not appeal to subjects.

18(E) How many subjects were enrolled in the study by gender, ethnicity and race?

Gender:

Males  
 Females  
 Unknown

Ethnicity:

Latinos or Hispanics  
 Not Latinos or Hispanics  
 Unknown

Race:

- American Indian or Alaska Native  
 Asian  
 Blacks or African American  
 Native Hawaiian or Other Pacific Islander  
 White  
 Other, specify: \_\_\_\_\_  
 Unknown

18(F) Where was the research study conducted? (List the county where the research study was conducted. If the treatment, prevention and diagnostic tests were offered in more than one county, list all of the counties where the research study was conducted.)

Philadelphia, PA

**19. Human Embryonic Stem Cell Research.** Item 19(A) should be completed for all research projects. If the research project involved human embryonic stem cells, items 19(B) and 19(C) must also be completed.

19(A) Did this project involve, in any capacity, human embryonic stem cells?

- Yes  
 No

19(B) Were these stem cell lines NIH-approved lines that were derived outside of Pennsylvania?

- Yes  
 No

19(C) Please describe how this project involved human embryonic stem cells:

**20. Articles Submitted to Peer-Reviewed Publications.**

20(A) Identify all publications that resulted from the research performed during the funding period and that have been submitted to peer-reviewed publications. Do not list journal abstracts or presentations at professional meetings; abstract and meeting presentations should be listed at the end of item 17. **Include only those publications that acknowledge the Pennsylvania Department of Health as a funding source** (as required in the grant agreement). List the title of the journal article, the authors, the name of the peer-reviewed publication, the month and year when it was submitted, and the status of publication (submitted for publication, accepted for publication or published.). Submit an electronic copy of each publication or paper submitted for publication, listed in the table, in a PDF version 5.0.5 (or greater) format, 1,200 dpi. Filenames for each publication should include the number of the research project, the last name of the PI, and an abbreviated title of the

publication. For example, if you submit two publications for Smith (PI for Project 01), one publication for Zhang (PI for Project 03), and one publication for Bates (PI for Project 04), the filenames would be:

- Project 01 – Smith – Three cases of isolated
- Project 01 – Smith – Investigation of NEB1 deletions
- Project 03 – Zhang – Molecular profiling of aromatase
- Project 04 – Bates – Neonatal intensive care

If the publication is not available electronically, provide 5 paper copies of the publication.

**Note:** The grant agreement requires that recipients acknowledge the Pennsylvania Department of Health funding in all publications. Please ensure that all publications listed acknowledge the Department of Health funding. If a publication does not acknowledge the funding from the Commonwealth, do not list the publication.

Title of Journal Article:	Authors:	Name of Peer-reviewed Publication:	Month and Year Submitted:	Publication Status (check appropriate box below):
1. Translational medicine individualizes healthcare discovery, development and delivery.	Terzic, A.T. and Waldman, S.A.	Biomarkers in Medicine	2013	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
2. Information hierarchies optimize patient-centered solutions.	Terzic, A.T. and Waldman, S.A.	Clin Pharm Ther	2012	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
3. Obesity pharmacotherapy: What is next?	Colon-Gonzalez, F., Kim, G., Lin, J.E., Valentino, M.A., and Waldman, S.A.	Mol Aspects Med	2012	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
4. Molecular staging of node negative patients with colorectal cancer.	Hyslop, T. and Waldman, S.A.	J Cancer	January 2013	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
5. Guanylyl cyclase C as a biomarker in colorectal cancer.	Hyslop, T. and Waldman, S.A.	Biomarkers in Medicine	2013	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
6. Immunotherapeutic strategies to target prognostic and predictive markers of cancer.	Magee, M.S., Snook, A.E., Marszalowicz, G.P., and Waldman, S.A.	Biomarkers Medicine	2013	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
7. Advances in cancer immunotherapy.	Snook, A.E. and Waldman, S.A.	Discovery Med	2012	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
8. Mechanisms of weight regain following weight	Blomain, E., Dirhan, D.,	ISRN Obesity	March 2013	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted

loss.	Valentino, M., Kim, G., and Waldman, S.A.			<input checked="" type="checkbox"/> Published
9. GUCY2C at the intersection of obesity and colorectal cancer.	Kim, G., Lin, J.E., and Waldman, S.A.	Trends Endocrin Metab	2012	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
10. New advances in models and strategies for developing anti-obesity drugs.	Kim, G., Lin, J.E., and Waldman, S.A.	Exp Opinion Drug Discovery	2013	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
11. Colorectal cancer immunotherapy.	Xiang, B., Snook, A.E., Magee, M.S., and Waldman, S.A.	Discovery Med	2013	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
12. Translating colorectal cancer prevention through the guanylyl cyclase C signaling axis.	Blomain, E.S., Lin, J.E., Kraft, C.L., Trela, U.T., Rock, J.M., Aing, A.S., Snook, A.E., and Waldman, S.A.	Expert Rev Clin Pharmacol	2013	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
13. Intestinal GUCY2C prevents TGF-B secretion coordinating desmoplasia and hyperproliferation in colorectal cancer.	Gibbons, A.V., Lin, J.E., Kim, G.W., Marszalowicz, G.P., Li, P., Stoecker, B.A., Blomain, E.S., Rattan, S., Snook, A.E, Schulz, S. and Waldman, S.A.	Can Res	March 2013	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
14. Anti-obesity pharmacotherapy: New drugs and emerging targets.	Kim, G., Lin, J.E., Blomain, E.B., and Waldman, S.A.	Clin Pharm Ther	September 2013	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
15. Molecular insights provide the critical path to disease mitigation.	Waldman, S.A., and Terzic, A.	Clin Pharm Ther	2013	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
16. Managing the innovation supply chain to maximize personalized medicine.	Waldman, S.A., and Terzic, A.	Clin Pharm Ther	2014	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
17. Tumor radiotherapy creates therapeutic vaccine responses to the colorectal cancer antigen GUCY2C.	Witek, M., Magee, M.S., Xiang, B., and Waldman, S.A., Snook, A.E.	Internationa l Journal of Radiation Oncology, Biology, Physics	October 2014	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
18. Selective antigen-	Snook, A.E.,	Eur J	February	<input type="checkbox"/> Submitted

specific CD4+ T cell, but not CD8+ T- or B-cell, tolerance corrupts cancer immunotherapy.	Magee, M.S., Schulz, S., and Waldman, S.A.	Immun	2014	<input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
19. Gut-brain endocrine axes in weight regulation and obesity pharmacotherapy.	Merlino, D., Blomain, E.S., Aing, A.S., and Waldman, S.A.	J Clin Med	April 2014	<input type="checkbox"/> Submitted <input checked="" type="checkbox"/> Accepted <input type="checkbox"/> Published
20. The paracrine hormone for the GUCY2C tumor suppressor, guanylin, is universally lost in colorectal cancer.	Wilson, C., Lin, J. E., Li, P., Snook, A.E., Gong, J., Sato, T., Chengbao, L., Gironde, M.A., Rui, H., Hyslop, and Waldman, S.A.	Cancer Epidemiol Biomarkers Prev	April 2014	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
21. A Novel CDX2 isoform regulates alternative splicing.	Witek, M.E., Snook, A.E., Lin, J.E., Blomain, E. S., Xiang, B., Magee, M., and Waldman, S.A.	PLOS One	February 2014	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published
22. GUCY2C lysosomotropic endocytosis delivers immunotoxin therapy to metastatic colorectal cancer.	Marszalowicz, G.P., Snook, A.E., Magee, M.S., Merlino, D., Berman-Booty, L.D., Waldman, S.A.	Oncotarget	August 2014	<input type="checkbox"/> Submitted <input type="checkbox"/> Accepted <input checked="" type="checkbox"/> Published

**20(B) Based on this project, are you planning to submit articles to peer-reviewed publications in the future?**

Yes  X  No \_\_\_\_\_

If yes, please describe your plans:

A number of articles will be submitted focused on RT-PCR analysis of GUCY2C to detect metastatic cells in normal tissues; techniques to maximize the estimation of GUCY2C mRNA by RT-PCR, and the utility of GUCY2C qRT-PCR for identifying stage I-II colorectal cancer patients who could benefit from adjuvant chemotherapy. Also, articles will be submitted concerning barriers to physician utilization of molecular technologies to manage patients.

**21. Changes in Outcome, Impact and Effectiveness Attributable to the Research Project.**

Describe the outcome, impact, and effectiveness of the research project by summarizing its

impact on the incidence of disease, death from disease, stage of disease at time of diagnosis, or other relevant measures of outcome, impact or effectiveness of the research project. If there were no changes, insert “None”; do not use “Not applicable.” Responses must be single-spaced below, and no smaller than 12-point type. **DO NOT DELETE THESE INSTRUCTIONS.** There is no limit to the length of your response.

These studies will demonstrate the utility of molecular diagnostics to detect occult tumor metastasis that will change the way stage I-II colorectal cancer patients are managed, and their survival.

These studies identify barriers to integrating molecular diagnostics into clinical practice that will serve as a template for developing strategies to overcome those barriers.

These studies form the basis for licensing and commercializing the platform technology to use GUCY2C to detect occult tumor burden to define chemotherapeutic responsiveness in stage I-II colorectal cancer patients.

**22. Major Discoveries, New Drugs, and New Approaches for Prevention Diagnosis and Treatment.** Describe major discoveries, new drugs, and new approaches for prevention, diagnosis and treatment that are attributable to the completed research project. If there were no major discoveries, drugs or approaches, insert “None”; do not use “Not applicable.” Responses must be single-spaced below, and no smaller than 12-point type. **DO NOT DELETE THESE INSTRUCTIONS.** There is no limit to the length of your response.

Completion of these analyses will form the basis for applying GUCY2C quantification of occult tumor burden to identify stage I-II colorectal cancer patients who could benefit from adjuvant chemotherapy.

These analyses identified significant barriers to utilizing molecular diagnostics in clinical practice that will form the basis for developing strategies to overcome those barriers.

### **23. Inventions, Patents and Commercial Development Opportunities.**

23(A) Were any inventions, which may be patentable or otherwise protectable under Title 35 of the United States Code, conceived or first actually reduced to practice in the performance of work under this health research grant? Yes   X   No                     

If “Yes” to 23(A), complete items a – g below for each invention. (Do NOT complete items a - g if 23(A) is “No.”)

Title of Invention:

- a. Name of Inventor(s): Scott A. Waldman

- b. Technical Description of Invention (describe nature, purpose, operation and physical, chemical, biological or electrical characteristics of the invention):

Quantifying GUCY2C to estimate occult tumor burden in lymph nodes to identify stage I-II colorectal cancer patients who could benefit from adjuvant chemotherapy.

- c. Was a patent filed for the invention conceived or first actually reduced to practice in the performance of work under this health research grant?

Yes \_\_\_\_\_ No X

If yes, indicate date patent was filed:

- d. Was a patent issued for the invention conceived or first actually reduced to practice in the performance of work under this health research grant?

Yes \_\_\_\_\_ No X

If yes, indicate number of patent, title and date issued:

Patent number:

Title of patent:

Date issued:

- e. Were any licenses granted for the patent obtained as a result of work performed under this health research grant? Yes X No \_\_\_\_\_

If yes, how many licenses were granted? One

*Statement of Clarification: This license was applied for prior to the start of this project, and the license was awarded during this project for a process/technology used during the project.*

- f. Were any commercial development activities taken to develop the invention into a commercial product or service for manufacture or sale? Yes \_\_\_\_\_ No X

If yes, describe the commercial development activities:

23(B) Based on the results of this project, are you planning to file for any licenses or patents, or undertake any commercial development opportunities in the future?

Yes X No \_\_\_\_\_

If yes, please describe your plans:

At the completion of the analyses, a patent will be filed that describes quantifying GUCY2C to estimate occult tumor burden in lymph nodes to identify stage I-II colorectal cancer patients who could benefit from adjuvant chemotherapy.

**24. Key Investigator Qualifications.** Briefly describe the education, research interests and experience and professional commitments of the Principal Investigator and all other key investigators. In place of narrative you may insert the NIH biosketch form here. However, please limit each biosketch to 1-2 pages. . *For Nonformula grants only – include information for only those key investigators whose biosketches were not included in the original grant application.*

This is a nonformula grant; investigator biosketches are provided in the grant application .