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 717-231-2825.

1. **Grantee Institution:** Trustees of the University of Pennsylvania

2. **Reporting Period (start and end date of grant award period):** 1/1/2011 – 12/31/2014

3. **Grant Contact Person (First Name, M.I., Last Name, Degrees):** Gearline Robinson-Hall, BSF

4. **Grant Contact Person's Telephone Number:** 215-746-6821

5. **Grant SAP Number:** 4100054874

6. **Project Number and Title of Research Project:** 11- Customization of Advanced Non-small Cell Lung Cancer Treatment Based on Emerging Histologic and Molecular Markers

7. **Start and End Date of Research Project:** 7/1/2011 – 12/31/2014

8. **Name of Principal Investigator for the Research Project:** Corey J Langer, MD

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:

   $ 609,554.43

   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
Albelda, Steven	Professor	1% Yr1-2; 2% Yr3	10,000.02
Werner, Diana	Nurse	5% Yr1;13% Yr 2; 100% Yr 3-4	162,216.40
Mick, Rosemarie	Research Investigator	4% Yr 3; 7% Yr4	15,844.56
Judy, Brendan	Research Specialist	8% Yr1; 32% Yr2	10,097.37
Cheng, Guanjun	Temp. Extra Person	67%Yr1	30,056.19

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
Langer, Corey	Principal Investigator	5%
Bauml, Joshua	Fellow, now Asst Professor in Medical Oncology	5%
Vachani, Anil	Asst Professor, Pulmonary	5%
Ahmed, Saman	Project Coordinator, Thoracic TCE	5%

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_____ No__X_____

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 awarded:
AP26113-11-101 (PI) ARIAD: Phase I/II Study of the Safety Tolerability, Pharmacokinetics and Preliminary Anti-Tumor Activity of the Oral ALK/EGFR Inhibitor AP26113	☐NIH ☐ Other federal (specify:_____) ■ Nonfederal source (specify: _Ariad)	06/11/2012-	$	$37,502
RP2 Trial of AP26113 at Two Different Dose in ALK (+) NSCLC Previously Treated with Crizotinib	☐NIH ☐ Other federal (specify:_____) ■ Nonfederal source (specify: Ariad_)	12/2014	$142,184	$ 10/20/2014
CO-1686-008: Phase I/2 Open-label Safety, Pharmacokinetic and Preliminary Efficacy Study of Oral CO-1686 in Patients with Previously Treated Mutant EGFR Non-Small Cell Lung Cancer (NSCLC)	☐NIH ☐ Other federal (specify:_____) ■ Nonfederal source (specify: _Clovis_____)	11/20/2014	$	$83,000

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 have used the work generated by this grant to leverage internal grants within our institution, including the ongoing Translational Center of Excellence (TCE) award that will enable us to expand immunotherapy trials in thoracic malignancy and to compare molecular profiles with patient-specific immune profiles. As detailed in the project spearheaded by Drs. Singhal and Albelda, we will start by analyzing resected specimens where tissue availability is abundant. In addition, we have used this grant to leverage ongoing work with Takeda and Millenium analyzing circulating tumor free DNA in advanced small cell carcinoma of the lung and our Transgenomic work evaluating molecular profiles in advanced NSCLC patients about to receive systemic chemotherapy for the first time. This work is ongoing. Finally, as a result of our work with ALK (+) NSCLC, we will be a regional site for a major, randomized phase II trial of AP26113 in the management of patients whose disease has progressed on front-line crizotinib; and we are involved in an ongoing study assessing the activity of Rociletinib (CO-1686) in the management of acquired resistance due to T790 in patients with EGFR mutation. This preliminary project promises to branch out as this agent goes forward with further testing in the first-line and second-line setting.

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

In addition to finishing the papers generated by this grant, we plan to expand on the work done with EGFR and KRAS mutations to other emerging molecular markers that govern the behavior of NSCLC. This includes EML4/ALK, for which we have received industry funding and are seeking additional funding, as well as BRAF, HER2, RET, ROS1, etc. Each of these markers has the potential to alter the therapeutic landscape of advanced NSCLC. We are also collaborating with Carmen Guerra of Internal Medicine evaluating the testing of markers in underserved populations, determining if disparities exist. Emerging data suggest that those with greater mutation burden are more likely to respond to newer immunotherapies, including Nivolumab, recently approved in second line squamous cell carcinoma of the lung. We plan to correlate personalized diagnostic results, including EGFR and KRAS, with immune profiles and therapeutic outcomes on PD1 and PDL1 inhibitors. Finally, as outlined above, we will be exploring the role of new agents in the management of acquired therapeutic resistance in patients with ALK translocation and EGFR mutation.

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				
Total				

	Undergraduate	Masters	Pre-doc	Post-doc
Hispanic				
Non-Hispanic				
Unknown				
Total				

	Undergraduate	Masters	Pre-doc	Post-doc
White				
Black				
Asian				
Other				
Unknown				
Total				

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.

    Briefly, our work here has helped enable us to implement the Center for Personalized Diagnostics (CPD), which in turn, has facilitated the treatment of patients with oncogene-driven NSCLC. In addition to the management of patients with EGFR mutation using commercially available products such as erlotinib and afatinib, we are actively evaluating investigational agents such as rociletinib (CO-1686) in the management of acquired resistance to EGFR mutation due to T790. Similar efforts are pending with Astellas Pharmaceutical. We have also leveraged this work into ongoing collaborations with Ariad

Pharmaceutical as we assess AP26113 in ALK (+) NSCLC patients who have developed resistance to crizotinib.

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__X_____ No_____

If yes, please describe the collaborations:

We have worked with industry, in the development of new agents targeting EGFR and ALK. See above (#15)

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

Yes_____ No___X_____

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

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

Yes_____ No____X_____

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

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 (α) and beta (ß) should not print as boxes (□) and include the appropriate citation(s). DO NOT DELETE THESE INSTRUCTIONS.

Expected Research Outcomes and Benefits

In a global sense, we expect this study to show that "customized" therapy based on molecular correlates, immunohistochemical correlates and clinical factors will lead to better care and improved outcomes compared to historic, empiric treatment in patients with advanced NSCLC. If we are able to do this, patients will directly benefit by receiving treatments that will enhance response rates to treatment, delay tumor progression, increase survival, and show less toxicity.

More specifically, we expect that

1) By improving tissue and serum collection approaches (i.e. by starting to do "reflex testing"), we will be able to apply current and developing predictive tests in a more useful fashion. We expect that advances in analysis of circulating tumor cells may be an important way in which we can obtain molecular correlates in patients without readily accessible tissue for biopsy, thus giving them access to these new tests and obviating the need for additional painful and expensive biopsy tests.
2) Thymidilate synthase staining, will be especially useful in helping us to decide which patients should receive pemetrexed therapy.
3) Patients with a heavier smoking history and those who continue to smoke will have worse outcomes. This could benefit patients by enabling us to pursue more intense anti-smoking strategies.

Summary of Research Completed

Objective #1 and #5: the collection of tumor tissue and serum to further elucidate the molecular typing of patients with advanced NSCLC;

Molecular testing project continues to proceed. Lynn Werner, the Research Coordinator hired for this project, under the aegis of GEI, has consented a total of 135 newly diagnosed, treatment-naïve, advanced NSCLC patients to date for biospecimen collection, including urine, blood, and tissue. 100 of these consents have occurred since the inception of this project. This material is banked under the auspices of Dr. Anil Vachani. Consent of new patients has ceased as of 2013, since these specimens are being pooled with specimens obtained prior to the inception of this project. In addition, as part of objective # 5, Lynn Werner has collected baseline smoking assessment surveys that will enable us to elucidate any linkage between current smoking status / total lifetime tobacco exposure and both therapeutic outcome and toxicity. As part of the GEI project, these surveys are paired to the biospecimen collection.

An early report from the GEI project centers on the relationship between mitochondrial (mt) DNA and both the incidence of and outcome in NSCLC. 465 incident cases were compared to 378 controls. Lower mtDNA content was associated with increased lung cancer risk and could potentially serve as a risk biomarker. However, mtDNA content was not associated with survival in NSCLC. The manuscript detailing this effort is in preparation and will likely be submitted to a peer-reviewed journal by the end of the summer, 2014. In addition, we have used material from the GEI study for a pemetrexed biomarker study (PMT), in conjunction with Steve Whitehead in Pharmacology; Recent evidence based on work by Dr. Whitehead suggests that differences in cellular folate metabolism may confer variation in tumor response to Pemetrexed. We investigated whether the pre-treatment intra-erythrocyte (RBC) level of 5-methyltetrahydrofolate (5-MTHF), a key component of folate metabolism is associated with response to Pemetrexed-based chemotherapy in newly diagnosed stage IV or recurrent lung adenocarcinoma. RBC 5-MTHF levels were quantified using a liquid chromatography/mass spectrometry-based method from blood drawn prior to initiation of chemotherapy. Clinical response was assessed by determining radiographic tumor response after 4-6 cycles of pemetrexed-based chemotherapy and categorized as partial response, stable disease, or progression. We explored the relationship between pre-treatment RBC 5-MTHF levels and response to pemetrexed-based chemotherapy. A log-binomial model was used to adjust for age and sex. 54 patients were enrolled and analyzed; median age was 61 (IQR 55-68) and 29 (54%) were female. RBC 5-MTHF ranged from 37 to 1,667 nM (mean 588, median 518). Following 4-6 cycles of chemotherapy, 23 patients (43%) had an objective radiographic response and 15 (28%) had stable disease. The remaining 16 patients (29.6%) had disease progression (7 during cycles 1-3, 9 during cycles 4-6). Patients with 5-MTHF levels in the lowest quartile were significantly less likely to achieve radiographic response compared to patients in the highest three quartiles (response rate 8% vs. 54%; risk difference 0.46, 95% CI 0.25-0.67). After controlling for age and sex, there remained a significantly higher likelihood of lack of response to pemetrexed for patients in the lowest 5-MTHF quartile (RR 6.86, 95% CI 1.01 – 46.92). We concluded that low pre-treatment RBC 5-MTHF levels were associated with an inferior response to Pemetrexed-based chemotherapy in stage IV lung adenocarcinoma. These findings were submitted as an abstract to the annual ASCO Meeting for 2015, but we recognize that much

larger, multicenter prospective studies are needed to validate RBC 5-MTHF as a predictive biomarker of pemetrexed in NSCLC.

As of January 2015, we intend to integrate these findings with our thoracic oncology database, now being developed by Dr. Langer and IT consultants, Oliver Mellet and Wallace Wormley, which, in turn, should enable us to garner enriched information regarding therapeutic outcomes and prognosis. This has become a particular focus over the past fiscal year (2013 – 2014), both under the aegis of this project and the newly funded Thoracic Center of Excellence, through which we have hired a project manager and database coordinator, Saman Ahmed as of 7/14. Her work on the Thoracic Oncology Database continues.

Objective #2: the development of a molecular pathology platform that incorporates simultaneous *reflex testing* of multiple candidate biomarkers;

We have also made considerable progress in initiating reflex testing of newly obtained NSCLC specimens, though as of 1/15, this has still not been formally enacted. This has required a complex collaboration with anatomic and molecular pathology [MP], as well as interventional radiology and pulmonology. Reflex testing was intended to "go live" during the summer of 2012, under the aegis of Dr. Christopher Watt, our collaborator in MP, and was to include all patients with non-squamous NSCLC as well as never smokers and remote, former smokers with squamous cell carcinoma of the lung. Unfortunately, this automatic approach has not yet occurred, though the newly constituted Center for Personalized Diagnostics (CPD), which went live in the 2nd Quarter of 2013, and partly grew out of this project, is highly responsive to our needs; the panel features a minimum of 40 genetic mutations and other molecular markers shared amongst solid tumors. Initial testing, by mutual agreement, includes EGFR mutation and ALK translocation, since these molecular abnormalities are "actionable" and we have had IRB-approved studies as well as FDA-approved agents that focus on these aberrant pathways. We have also included KRAS mutations, as well as panoply of additional, potentially "actionable" molecular aberrations, including BRAF, HER2, ROS1, RET etc, although not yet formally "actionable" with FDA-approved agents in advanced NSCLC. To date, as of the end of December, 2014, 2091 patients institution-wide have been tested, of which lung cancer constitutes the plurality with 695 or 33.2 % of the total; of 582 lung cancer patients on whom we have information, 107 or 18.3% have proven KRAS (+), and 81 or 13.9 % have proven (+) for EGFR mutations.. The data for ALK positivity are still pending. MEK inhibitors [MEKIs] appear to have activity in patients with this mutation, and we have also conducted formal trials at Penn employing MEKIs, as a partial outgrowth of this project.

In related projects stemming from this effort, Drs. Anil Vachani of Interventional Pulmonology, Charu Aggarwal [an attending in Medical Oncology hired since the inception of this project], former fellow, Anjana Ranganathan, former fellow and current colleague, Joshua Bauml MD, and medical resident Anna Brady (who has since left the institution), and I have assembled data on contemporaneous advanced NSCLC patients with and without KRAS mutation, showing that prognosis in the KRAS mutation (+) cohort treated with standard cytotoxic chemotherapy is not inferior to those who have KRAS wild type tumors. The results to date fly in the face of conventional wisdom. These observations were submitted as an abstract and presented as a poster at the 2012 ASCO Meetings; manuscript is being prepared. Prospective assessment of KRAS mutation status and its influence on therapeutic outcome continues under the aegis of Drs.

Aggarwal and Ahmed, with the CPD providing source data to our incipient database.

In addition, Joshua Bauml MD, initially in collaboration with Chris Watt MD, the Assistant Director of Molecular Pathology, along with Corey Langer, MD, and Rosemarie Mick of the Biostatistics Core has reviewed the association of EGFR and KRAS status with race and outcome. Dr. Bauml finalized the results in the Spring of 2013 and subsequently published these findings in a paper entitled *Frequency of EGFR and KRAS mutations in patients with non small cell lung cancer by racial background: Do disparities exist?* in 2013 in Lung Cancer . This effort constituted a retrospective chart review of 513 NSCLC patients undergoing EGFR and KRAS mutational analysis at the Hospital of the University of Pennsylvania between May 2008 and November 2011, predating the development of our CPD platform. Clinical and pathologic data were abstracted from the patients' electronic medical record. Of 497 patients with informative EGFR mutation analyses, the frequency of EGFR mutation was 13.9%. The frequency of EGFR mutations was associated with race ($p<0.001$) and was lower in African American (AfA) patients compared to Caucasian (C) patients but did not reach statistical significance (4.8% vs 13.7%, $p=0.06$). Mean Charlson Comorbidity Index and number of cigarette pack years were significantly lower in patients with EGFR mutations ($p=0.01$ and $p<0.001$, respectively). Multivariable logistic regression analysis showed a significant association between race and EGFR mutation ($p=0.01$), even after adjusting for smoking status ($p<0.001$) and gender ($p=0.03$). KRAS mutation (study frequency 28.1%) was not associated with race ($p=0.08$; $p=0.51$ for AfA vs C patients), but was more common among smokers ($p<0.001$) and females ($p=0.01$). Based on multivariable analysis, even after adjusting for smoking status and gender, we found that race was statistically significantly associated with EGFR mutation, but not KRAS mutational status. To our knowledge, this is one of the largest series to date evaluating racial differences in EGFR and KRAS mutational status among patients with NSCLC.

Joshua Bauml has also looked at the influence of EGFR and KRAS mutations on survival, and based on his findings, he published a paper in *Clinical Lung Cancer*: *Bauml J, Mick R, Zhang Y, Watt CD, Vachani A, Aggarwal C, Evans T, Langer C. "Determinants of Survival in Advanced Non-Small Cell Lung Cancer (NSCLC) in the Era of Targeted Therapies, Clinical Lung Cancer, Volume 14, Issue 5, September 2013, Pages 581–591.* This survival analysis was based upon the 376 patients who received systemic treatment, and it was determined from date of initiation of systemic therapy. The median overall survival was 30.8 months (95%CI 24.7-36.9 months). Surprisingly, neither *EGFR* mutational status ($p=0.09$) nor *KRAS* mutational status (0.69) was associated with overall survival differences. Female sex ($p<0.001$), never smoker status ($p=0.01$), better PS ($p<0.001$), lower Charlson Comorbidity Index ($p<0.001$) and lower age-weighted Index ($p<0.001$) were associated with prolonged survival. The presence of bone metastases ($p=0.001$) and liver metastases ($p=0.004$) were also associated with shortened survival. Multivariable regression demonstrated that male gender ($p=0.002$), worse ECOG PS ($p=0.01$), metastases to bone ($p=0.03$) and higher age-weighted co-morbidity Index ($p=0.001$) were independent prognostic factors for shorter survival. In conclusion, in contrast to the observations of other researchers, *EGFR* and *KRAS*, in our series, have not functioned as prognostic determinants for NSCLC. Nevertheless, these patients appear to live twice as long as we had surmised in our original statistical prognostications, and certainly a lot longer than patients lived before the advent of routine molecular testing.

As a result of this and other efforts in other malignancies, the institution has developed the Center for Personalized Diagnostics (CPD), which includes tests for EGFR and KRAS mutation, as well as other actionable markers, including EML4/ALK, and other *potentially* actionable genetic abnormalities found in NSCLC. The CPD platform permits expedited, simultaneous testing of multiple markers, essentially maximizing the yield of limited tissue samples. These reports are now found in EPIC, our electronic medical record, separate from the original pathology reports; and while we have yet to develop true "reflex" testing, as we had originally intended, this current collaboration is iterative and highly sensitive to feedback, both from medical oncology and molecular pathology. It enables us to bypass the expense and inconvenience of commercial labs outside the institution. To date, since the inception of the CPD at the beginning of 2013, we have tested over 600 NSCLC specimens on campus.

Drs. Charu Aggarwal and Saman Ahmed, who took over as Project Manager for our Translational Center of Excellence, under the aegis of Dr. Corey Langer, have assessed outcomes in select individuals whose tumor was included in the CPD and correlated their mutation status with therapeutic outcome. Specifically, we conducted a retrospective analysis of Stage IV NSCLC patients treated at the Abramson Cancer Center with established EGFR and TP53 MT status using gene sequencing Overall Survival (OS) and Progression Free Survival (PFS) were determined for the p53 MT and wild type (WT) cohorts in TKI sensitive (exon 19, 21;SEN) and TKI non-sensitive (exon 18, 20, non- SEN) EGFR MT cohorts. Of 51 pts analyzed, there were 37 SEN and 14 non- SEN pts; 38 had TP53 MT and 13 were p53 WT pts. TP53 MT and WT groups had similar median age (67 vs. 63 yrs), PS (90% ECOG 0-1), smoking status (47% vs. 46% smokers) and T790M status (16% vs. 23%) and female pre dominance (55% and 69% respectively). Exon 19 deletions (69% vs. 29%), brain (69% vs 55%) and bone (69% vs. 39%) metastasis were more common in the TP53 WT group. At median follow-up of 20.7 mos (15.1 for 37 living pts), the overall median PFS was 15.5 mos (95% CI = 12.8 – 18.2 mos) and OS 46.3 mos (95% CI = 31.7 – 60.8 mos).

Cohort	No	Med PFS mos (95% CI)	Med OS mos (95% CI)
Non-SEN			
TP53 WT	2	Not estimable	Not estimable
TP53 MT	12	11.5 (10.8-12.2)	28.3 (4.3-52.4)
SEN			
TP53 WT	11	33.2 (3.3 – 63)	Not reached (no deaths)
TP53 MT	26	17.1 (13.8 -20.5)	46.3 (38.9-53.7)
Log rank test		p = 0.52	p = 0.12

Based on these results, we have concluded that the coexistence of TP53 MT is likely associated with worse clinical outcomes in NSCLC patients with either SEN or non- SEN EGFR MTs, but we have also concluded that larger sample sizes would be needed to firmly establish whether true associations exist.

Objective #3- evaluation of thymidylate synthetase (TS) status in tumor specimens as both a prognostic and predictive marker in patients receiving pemetrexed;

Lynn Werner, under the aegis of Drs. Vachani and Langer, has obtained consents for biospecimen collection from newly diagnosed patients with advanced NSCLC about to receive pemetrexed under the aegis of the TGM study. 39 patients through July 2014 have signed ICFs and undergone blood draws; since July 2013, 29 patients receiving Pemetrexed have consented and undergone biospecimen collection. We believe we will conclude accrual to this crucial project by the beginning of 2015 Despite the advent of "targeted therapy," chemotherapy remains the therapeutic backbone of most of our clinical efforts in the vast majority of patients; and Pemetrexed, over the past ten years, has emerged as a dominant agent in the non-sqaumous cell carcinoma population. It has been postulated that low levels of thymidylate synthetase (TS) in tumor specimens is both a prognostic and predictive marker in patients receiving pemetrexed, that low levels specifically correlate with better outcome. Recent data in malignant pleural mesothelioma from our institution, as delineated by Dr. Albelda, would call this assumption into question. Consequently, as part of this project, in addition to assays for TS, Dr. Vachani's lab will be looking at other putative markers (e.g. Folyl-polyglutamate synthase [FPGS]), and will correlate both TS and FPGS results with therapeutic outcome. We hope to have these data assembled by the early part of 2016 for either ASCO or AACR.

Objective #4- identification of circulating tumor cells (CTCs) in serum specimens collected at the start of treatment and at first radiographic evaluation, both to predict outcome and to determine or predict sensitivity to therapy based on molecular testing;
We have continued to work closely with the Veridex company to detect CTCs in small cell and non-small cell lung cancer. To date, over 60 patients have been consented for this project, with roughly 9 patients consented since July, 2014. In addition, in conjuction with Takeda/Millenia, we have embarked on a project analyzing CTCs as well as circulating free (cf) DNA in patients with relapsed or progressive SCLC, as a means of potentially identifying benefit to new agents in the management of this challenging disease. To date, since July, 2013, twelve patients have been accrued to this project. Preliminary data to date have shown a fairly good correlation between CTCs and cfDNA in relapsed patients.

Unlike small cell lung cancer, where CTC's are easy to detect, it has been difficult to detect CTC's in non-small cell lung cancer using the FDA approved, standardized antibody based system. This approach (Veridex, Cell Search System) uses antibodies to the epithelial molecule EPCAM coupled to magnetic ferrofluids to isolate CTCs. After collecting EPCAM+ cells, CTCs must also be cytokeratin positive and CD45 (a white blood cell marker) negative. We have collected serum from 20 advanced stage lung cancer patients. Only 4 of the 20 had detectable levels of CTC (>2 per 7.5 ml), and only one had more than 7 CTC. We have been trying to understand why. Our leading hypothesis is now that lung cancer cells undergo an epithelial to mesenchymal transition (EMT) when they leave the tumor and go into the blood stream. This means that these cells lose EPCAM expression and presumably gain new markers consistent with a mesenchymal phenotype.

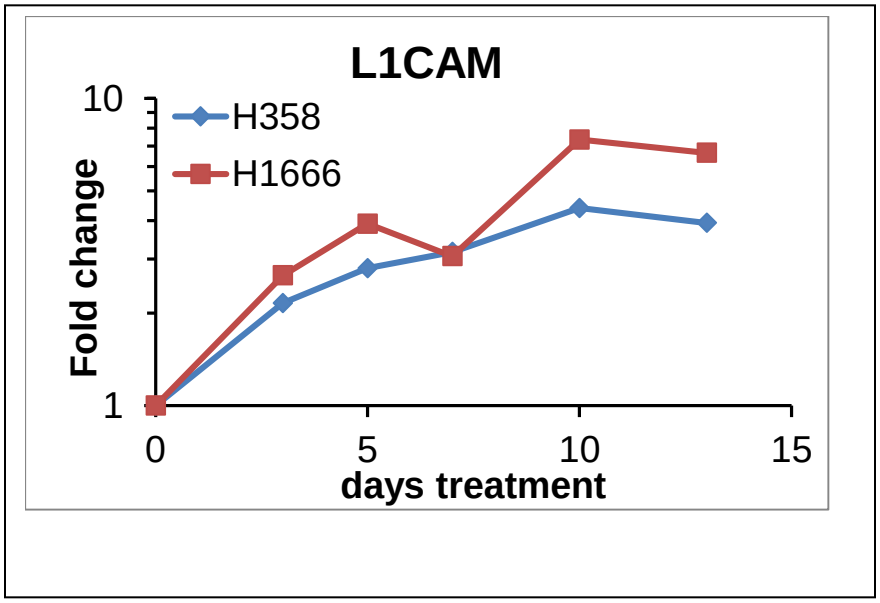
We are approaching this question by taking lung cancer cells in culture and exposing them to conditions that force them towards the mesenchymal phenotype (exposure to TGF-beta and

EGF). Under these conditions, they lose EPCAM and increase classic markers of EMT- i.e. an increase in vimentin. We are assembling these cells and evaluating them in two ways. First, we are testing new capture antibodies based on the literature. Our leading candidate is a surface protein called L1-CAM. As shown in **Figure 1** below, EMT induces strong surface expression of L1-CAM in our two lung cancer lines. We are now working with Veridenx to make a "ferrofluid" that will allow us to "pull" L1-CAM expressing cells from the blood.

In our second approach, we are taking baseline cells and cells that have undergone EMT and are extracting RNA. We will compare the two conditions and use bioinformatics to identify potentially new targets.

Once we find our optimal capture system using cell lines, we will collect serum from lung cancer patients and look for CTCs.

Figure 1. Two lung cancer cell lines were subjected to flow cytometry to detect surface expression of L1-CAM. The cells were studied at baseline (time 0) and then at varying times after exposure to TGF-beta/EGF to induce EMT. Both lines show strong upregulation of L1-CAM over time.



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

____X_No

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

_____Yes

___X__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? These data are pending

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.)

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
___X__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. Frequency of EGFR and KRAS mutations in patients with non small cell lung cancer by racial background: Do disparities exist?	Joshua Bauml, Rosemarie Mick, Yu Zhang, Christopher D. Watt, Anil Vachani, Charu Aggarwal, Tracey Evans, Corey Langer	Lung Cancer	2013	☐Submitted ☐Accepted ■Published
2. Determinants of Survival in Advanced Non-Small Cell Lung Cancer (NSCLC) in the Era of Targeted Therapies	Bauml J, Mick R, Zhang Y, Watt CD, Vachani A, Aggarwal C, Evans T, Langer C	Clinical Lung Cancer Vol 14, Issue 5, Sept 2013, Pgs 581–591.	2013	☐Submitted ☐Accepted ■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:

We plan to submit articles on the following topics to JTO, JCO and Lung Cancer:

1. The influence of KRAS mutations in outcome in advanced NSCLC patients receiving systemic chemotherapy
2. The interaction of p53 status with EGFR mutation in advanced NSCLC
3. The role of CTCs in influencing outcome in patients with SCLC
4. The relationship between pre-treatment RBC 5-MTHF levels and response to pemetrexed-based chemotherapy in advanced NSCLC

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.

None

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.

As a result of this project, we have leveraged our capacity, in collaboration with Pharma, to test new agents in the setting of therapeutic resistance to EGFR mutation and ALK translocation. These include Rociletinib (CO1686), which targets T790, which governs resistance in ~ 60% of patients with EGFR mutation and AP26113, which has demonstrated considerable therapeutic activity in ALK (+) patients who have developed resistance to crizotinib.

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_____ No___X_____

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

a. Title of Invention:

b. Name of Inventor(s):

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

d. 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___

If yes, indicate date patent was filed:

e. 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____
If yes, indicate number of patent, title and date issued:
Patent number:
Title of patent:
Date issued:

f. Were any licenses granted for the patent obtained as a result of work performed under this health research grant? Yes_____ No____

If yes, how many licenses were granted?_____

g. Were any commercial development activities taken to develop the invention into a commercial product or service for manufacture or sale? Yes___ No____

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_____ No____X_____

If yes, please describe your plans:

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.

BIOGRAPHICAL SKETCH

NAME	POSITION TITLE
Corey J. Langer, M.D.	Member of the Abramson Cancer Center, University of Pennsylvania
eRA COMMONS USER NAME: CLANGER	

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, and include postdoctoral training.)*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	YEAR(s)	FIELD OF STUDY
Boston University	B.A.	1981	Med. Science, Minor in English
Boston University	M.D.	1981	Combined 6 year program in Medicine

PERSONAL STATEMENT:

Corey Langer, M.D., Professor of Medicine at the University of Pennslvania, is the Clinical Director of Thoracic Oncology at the Abramson Cancer Center and Chair of the Medical Oncology Committee for the Radiation Therapy Oncology Group (RTOG), now NRG. He brings extensive clinical trial experience to research efforts both at Penn in the Cooperative Group system (RTOG and ECOG). For the 28 years, since completing fellowship, he has led or co-led over 120 clinical trials in both small cell (SCLC) and non-small cell lung cancer (NSCLC), in the adjuvant, locally advanced (LA), and advanced disease setting, and at least 20 trials in Head and Neck Cancer (HNC). He is the author or co-author of over 200 peer-reviewed papers, primarily focusing on the treatment of smoking-related malignancies. He is a core member of both the RTOG/NRG and ECOG HNC and Thoracic Committees. His clinical research efforts have extended from phase I to phase III studies, with an emphasis on new drug combinations and special populations, including the elderly and those with compromised performance status. His work in both advanced and LA-NSCLC helped show that increasing age, by itself, was not a negative prognostic factor, that PS trumped age in this regard. He has a particular interest in combined modality therapy, which dates back to fellowship when he first investigated the role of concurrent platinum-based chemotherapy and radiation (XRT) in unresectable LA-NSCLC. His research efforts culminated in a phase III trial (RTOG 9410), which he helped spearhead and which demonstrated a survival advantage for concurrent chemo-radiation over sequential therapy, the erstwhile standard of care. Since that time, he has investigated the role of new(er) agents in combination with local XRT as well as the still "radical" notion of radiosensitizing chemotherapy (cisplatin and paclitaxel) and **re**-irradiation in patients whose lung and HNCs have recurred after definitive XRT and who are no longer resectable, first in phase I at Fox Chase Cancer Center, and then in phase II through the RTOG (9911). He is currently collaborating in investigating proton beam re-irradiation with his colleagues at Penn. The latter study has produced longterm survivors, who likely would have succumbed with systemic therapy alone. He also led an ECOG trial evaluating the role of concurrent C225 and cisplatin/XRT in locally advanced, treatment-naïve, unresectable HNC (ECOG 3303). He helped usher in the use of carboplatin/paclitaxel as as a treatment standard in advanced NSCLC, and was amongst the first medical oncologists in the US to investigate the role of concurrent irinotecan and definitive cisplatin with standard XRT in limited stage SCLC,; more recently, he was one of the leaders of a randomized phase II trial of carboplatin/etoposide +/- obatoclax, a BCL2 inhibitor, in extensive stage SCLC, which he presented at both ASCO [6/11] And IASLC [7/11] in Amsterdam, Holland, and which showed a borderline significant survival advantage. He currently leads clinical research efforts at Penn in thoracic malignancy as part of the Interdisciplinary Thoracic Oncology Program (I-TOP); and, in the current era of molecularly driven cancer therapy, he has created a working coalition with Anatomic and Molecular Pathology delineating and documenting patient-specific "actionable" mutations and translocations, which are now linked to studies investigating novel therapies in adenocarcinoma of the lung. In coordination with Steve Albelda of the Pulmonary Division at Penn, he is currently leading research studies focused on the identification and implications of circulating tumor cells (CTCs) in SCLC and the immunotherapy of NSCLC. Finally, he spends considerable time mentoring junior investigators as they launch their own clinical trials.

POSITIONS AND HONORS *(Selected)*

Research and Professional Experience:

1990-2008	Chair of the FCCC Pharmacy and Therapeutics and Practice Monitoring Committees
1994-2008	Medical Director, Thoracic Oncology, FCCC
2008-	Professor of Medicine, Clinical Director, Thoracic Oncology, University of Pennsylvania
2009-2014	Co-Chair of the Cancer Therapeutics Core of the Abramson Cancer Center
2011-	Co-Chair of the Interdisciplinary Thoracic Oncology Program (I-TOP) at Penn
2013-	Co-Chair of the Penn Translational Center of Excellence in Thoracic Oncology

HONORS AND OTHER PROFESSIONAL ACTIVITIES (Selected)

1981	Phi Beta Kappa: Boston University
June 1984	Guth Award for Excellence in Patient Care, Graduate Hospital Residency Program
1986-87	American Cancer Society, Clinical Fellow, -
1987-	Member, ASCO
1993-	Journal of Clinical Oncology: Reviewer (1993-)
	Editorial Board Member (1996 – 1999; 2000 – 2003; 2005 – 2008; 2009 - 2012)
1996-	American College of Physicians, Fellow
1998-2008	Research Director of the Oncology Physician's Network (OPN) in the Delaware Valley (under aegis of FCCC)
2002-2008	Chair, American College of Surgeons, HNC National Database Committee
2002-	SWOG, Member, Data and Safety Monitoring Committee (DSMC)
2005-2009	Geriatric Oncology Consortium: Member Scientific Advisory Board
2002-	Chair of the Medical Oncology Subcommittee, RTOG, now NRG
2002-	Vice Chair of the RTOG, now NRG
2002- 2013	Member of the RTOG Publications and Translational Research Committee
2009- 2013	Co-Chair of the RTOG Internal Data Safety Monitoring Committee Board

PUBLICATIONS Relevant to Thoracic and Head and Neck Oncology (Selected since 2013)

30. Bauml, J., Mick, R., Zhang, Y., Watt, C. D., Vachani, A., Aggarwal, C., **Langer, C**. (2013). Frequency of *EGFR* and *KRAS* Mutations in Patients with Non Small Cell Lung Cancer by Racial Background: Do Disparities exist? *Lung Cancer 81*(3), 347–353. doi:10.1016/j.lungcan.2013.05.011

31, **Langer, C. J**., Novello, S., Park, K., Krzakowski, M., Karp, D. D., Mok, T.& Jassem, J. (2014). Randomized, Phase III Trial of First-Line Figitumumab in Combination With Paclitaxel and Carboplatin Versus Paclitaxel and Carboplatin Alone in Patients With Advanced Non–Small-Cell Lung Cancer. *Journal of Clinical Oncology*, JCO-2013.

32. Hirsh, V., A. Berger, G. Binder, **C. Langer**, T. J. Ong, M. Renschler, R. Bornheimer, S. Whiting, and G. Oster. "Cost Effectiveness of Nab-Paclitaxel Plus Carboplatin (nab-PC) Relative to Bevacizumab Plus Solvent-Based Paclitaxel and Carboplatin (B+ sb-PC) in Elderly Patients With Advanced Non-Small Cell Lung Cancer (NSCLC)." *International Journal of Radiation Oncology Biology Physics* 90, no. 5 (2014): S61-S62.

33. **Langer, Corey J**., Vera Hirsh, Amy Ko, Markus F. Renschler, and Mark A. Socinski. Weekly nab-Paclitaxel in Combination With Carboplatin as First-Line Therapy in Patients With Advanced Non–Small-Cell Lung Cancer: Analysis of Safety and Efficacy in Patients With Renal Impairment. *Clinical lung cancer* (2014).

34. Boggs, D. H., Robins, H. I., **Langer, C. J**., Traynor, A. M., Berkowitz, M. J., & Mehta, M. P. Strategies to Prevent Brain Metastasis in High-Risk Non–Small-Cell Lung Cancer: Lessons Learned From a Randomized Study of Maintenance Temozolomide Versus Observation. *Clinical lung cancer*, *15*(6), 433-440. (2014).

35. **Langer, C. J.,** Albert, I., Ross, H. J., Kovacs, P., Blakely, L. J., Pajkos, G. & GEM017 Investigators. (2014). Randomized phase II study of carboplatin and etoposide with or without obatoclax mesylate in extensive-stage small cell lung cancer. *Lung Cancer*, *85*(3), 420-428.

36. **Langer, C. J.,** Socinski, M. A., Patel, J. D., Sandler, A. B., Schiller, J. H., & Leon, L. (2015). Isolating the Role of Bevacizumab in Elderly Patients With Previously Untreated Nonsquamous Non–Small Cell Lung Cancer (in press)