Biomarkers to Help Guide Management of Patients with Pulmonary Nodules

Edward F. Patz, Jr.[1,2], Michael J. Campa[1], Elizabeth B. Gottlin[1], Priscilla R. Trotter[1], James E. Herndon II[1], Don Kafader[3], Russell P. Grant[3], and Marcia Eisenberg[3]

[1]Department of Radiology and [2]Department of Pharmacology and Cancer Biology, Duke University Medical Center, Durham, North Carolina; and [3]Laboratory Corporation of America Holdings, Burlington, North Carolina

Rationale: **Indeterminate pulmonary nodules are a common radiographic finding and require further evaluation because of the concern for lung cancer.**

Objectives: **We developed an algorithm to assign patients to a low- or high-risk category for lung cancer, based on a combination of serum biomarker levels and nodule size.**

Methods: **For the serum biomarker assay, we determined levels of carcinoembryonic antigen, α_1-antitrypsin, and squamous cell carcinoma antigen. Serum data and nodule size from a training set of 509 patients with (n = 298) and without (n = 211) lung cancer were subjected to classification and regression tree and logistic regression analyses. Multiple models were developed and tested in an independent, masked validation set for their ability to categorize patients with (n = 203) or without (n = 196) lung cancer as being low- or high-risk for lung cancer.**

Measurements and Main Results: **In all models, a large percentage of individuals in the validation study with small nodules (<1 cm) were assigned to the low-risk group, and a large percentage of individuals with large nodules (≥3 cm) were assigned to the high-risk group. In the validation study, the classification and regression tree algorithm had overall sensitivity, specificity, and positive and negative predictive values for determining lung cancer of 88%, 82%, 84%, and 87%, respectively. The logistic regression model had overall sensitivity, specificity, and positive and negative predictive values of 80%, 89%, 89%, and 81%, respectively.**

Conclusion: **Integration of biomarkers with lung nodule size has the potential to help guide the management of patients with indeterminate pulmonary nodules.**

Keywords: lung cancer; diagnosis; biomarkers

Indeterminate pulmonary nodules are a common radiographic finding, and require further evaluation because of the concern for lung cancer. Patient work-up depends on the clinical scenario and the size and morphology of the abnormality. Most lesions are small (<1 cm) and most patients are followed with sequential computed tomography (CT) studies (1). Other individuals are subjected to additional imaging with positron emission tomography (PET) or undergo a biopsy to establish a diagnosis. A more efficient and cost-effective strategy that would avoid delays in diagnosis, decrease radiation, and reduce the need for invasive procedures would have a significant impact on patient management.

Biomarker analysis is potentially one element of such a diagnostic strategy, but the discovery and validation of tumor-specific markers has been difficult. We previously reported on a panel of serum markers to be used for the diagnosis of lung cancer (2). We now focus on using a combination of these markers plus nodule size (1, 3, 4) to stratify patients as to the risk of lung cancer (i.e., low or high risk). This approach seems to provide information about the risk of lung cancer in patients with indeterminate pulmonary lesions, and may be used in conjunction with clinical presentation to develop a more effective management strategy.

AT A GLANCE COMMENTARY

Scientific Knowledge on the Subject

Nodules are difficult to diagnosis as either benign or lung cancer from imaging results alone.

What This Study Adds to the Field

The addition of serum biomarker levels to nodule size data results in assignment of lung cancer risk to patients with indeterminate lung nodules.

METHODS

Overall Study Design

This study was designed to evaluate a combination of three serum proteins and nodule size to differentiate benign from malignant pulmonary nodules in patients at risk of having lung cancer. We first analyzed a training set of 509 patients with indeterminate nodules using classification and regression tree (CART) and logistic regression analyses that assigned patients to a low- or high-risk category. These models were then tested in an independent, masked validation set of 399 patients.

Serum Marker Assays

We established a lung cancer biomarker assay (LCBA) that measures the serum levels of carcinoembryonic antigen (CEA), α_1-antitrypsin (AAT), and squamous cell carcinoma antigen (SCC). The make-up of the LCBA is as follows: CEA (Roche Cobas Modular Analytics E170 Assay; Roche, Indianapolis, IN); AAT (Roche COBAS Integra Tina Quant Assay); and SCC (CanAg SCC EIA; Fujirebio, Göteborg, Sweden). All samples in the training set were run in duplicate. For the validation study, the SCC assay was run in duplicate, and CEA and AAT assays were run on automated analyzers in singlicate per the manufacturer's instructions.

(*Received in original form October 1, 2012; accepted in final form December 18, 2012*)

Supported by Laboratory Corporation of America Holdings.

Author Contributions: E.F.P., concept, design, data analysis, literature search, data interpretation, and writing. M.J.C., concept, design, data analysis, literature search, data interpretation, and writing. E.B.G., design, data analysis, literature search, data interpretation, and writing. P.R.T., data collection. J.E.H., design, data analysis, data interpretation, and writing. D.K., design, data analysis, data interpretation, and writing. R.P.G., design, data analysis, data interpretation, and writing. M.E., design, data analysis, data interpretation, and writing

Correspondence and requests for reprints should be addressed to Edward F. Patz, Jr., M.D., Duke University Medical Center, Department of Radiology, Box 3808, Durham, NC 27710. E-mail: patz0002@mc.duke.edu

This article has an online supplement, which is accessible from this issue's table of contents at www.atsjournals.org

Am J Respir Crit Care Med Vol 188, Iss. 4, pp 461–465, Aug 15, 2013

Originally Published in Press as DOI: 10.1164/rccm.201210-1760OC on January 10, 2013
Internet address: www.atsjournals.org

Imaging Studies, Nodule Size, and Pathologic Classification

All patients enrolled in either the training or validation study had a CT study of the thorax, performed for a variety of indications including the following: possible respiratory neoplasm, pulmonary nodule, interstitial lung disease, pulmonary embolism, pneumonia, or chest pain. CT studies at our institution were performed using multiple contiguous 5-mm sequential axial images through the thorax.

The size of the nodule or focal abnormality was extracted by the study coordinator (P.R.T.) from the official clinical CT report. In the validation study, 18 patients (4%) had their CT study performed at another institution, and the tumor size was determined from the pathology report. The longest dimension on axial CT images or pathologic report was recorded.

All patients with lung cancer had histologic confirmation and pathologic staging. Patients without lung cancer had a histologic diagnosis, CT resolution or 2-year stability of the lesion, or clinical observation without evidence of lung cancer.

Patient Selection

All patients enrolled in this study signed an informed consent form approved by Duke University's Institutional Review Board in accordance with an assurance filed with and approved by the Department of Health and Human Services. We focused on patients who had lesions greater than 5 mm in whom there was a concern for lung cancer. The training set included a total of 509 archived serum samples, collected between January 2001 and June 2007. A total of 153 of these 509 training serum samples were provided by the Mayo Clinic. Included with the serum sample of each patient were the serum draw date, CT date, nodules size, and diagnosis including histology and stage (lung cancer = 77; no cancer = 76). There were 298 patients with a confirmed diagnosis of lung cancer and 211 individuals without cancer who met the following inclusion criteria: (*1*) no prior history of lung cancer, (*2*) no currently known extrathoracic malignancy, and (*3*) a CT of the thorax with an indeterminate pulmonary nodule. All patients had blood drawn and their serum was stored immediately at -80°C in the laboratory repository. Clinical data including age, sex, race, past medical history, pathologic diagnosis, and imaging findings were recorded in a confidential trial database.

The independent validation set included sequential patients between August 2007 and June 2011, with an indeterminate pulmonary nodule or focal opacity detected on thoracic CT. There were 203 patients with lung cancer and 196 patients without lung cancer. Serum was collected from each subject and stored at -80°C. After enrollment was complete, the samples were thawed for the first time, and 500-μl aliquots were placed in deidentified tubes with randomly assigned unique study identification numbers.

Statistical Methods

The association between various characteristics of a pulmonary nodule (serum levels of AAT, SCC, CEA, and nodule size) and the presence of lung cancer was assessed within the training set data using CART methodology (5, 6) and logistic regression. The CART model was generated using the Gini index that favors even splits with a minimum size of 5 for terminal nodes and 10 for parent nodes. The probability of cancer was assumed to be 50% and the optimal tree was the tree with minimum cost regardless of size. Ten-fold cross-validation was used in generating this model.

The logistic model provided an estimate of the probability that cancer was present as a function of nodule diameter, CEA, SCC, and AAT. Two threshold probabilities were considered for predicting a cancer diagnosis: a calculated probability of 0.5 and higher, and a threshold probability of 0.3. The 0.5 threshold is a standard probability and the 0.3 threshold is intended to reduce the number of false-negative diagnoses.

An independent validation study was designed to test the accuracy of the CART and logistic regression models developed from the training set. Each model was used to assign each subject within the validation dataset a "lung cancer" (high-risk group) or "no lung cancer" (low-risk group) designation. After classification of all patients, the diagnosis was revealed and the sensitivity, specificity, and positive and negative predictive values for each model were determined. This validation study was designed to specifically assess the sensitivity and specificity of the derived classification algorithm for the presence or absence of cancer within a patient group representative of all patients with a pulmonary nodule or focal pulmonary abnormality.

The study was designed to include at least 384 patients with an indeterminate pulmonary nodule (192 patients with cancer and 192 patients without cancer). In this study the probability that a cancerous nodule is diagnosed as cancerous without the benefit of a pathology assessment is 50%. If the classification algorithm increased that probability to 75%, then it would be considered potentially clinically useful in the diagnosis of cancer in patients with a pulmonary nodule. If the classification algorithm increased the probability of detecting a cancerous tumor to 65%, it would not be worthy of clinical use. With 192 patients with cancer, there was 90% power to differentiate between a sensitivity rate of 65% (null hypothesis) and 75% (alternative hypothesis) assuming $\alpha = 0.05$. A similar argument can be made for the determination of benign lesions when a nodule is truly benign. With 192 patients without cancer, there was 90% power to differentiate between a specificity rate of 65% and 75% assuming $\alpha = 0.05$.

RESULTS

Training Set Results

A total of 509 archived serum samples were included in the training dataset. Patient characteristics including age and sex are summarized for the 298 patients with a confirmed diagnosis of lung cancer and the 211 individuals without cancer in Table 1. The distribution of benign and malignant nodules according to lesion size (small, <1 cm; intermediate, 1 to <3 cm; and large, ⩾3 cm) approximately reflected the expected proportion of cases as reported in the literature (Figure 1A). The training dataset is provided in Table E1 in the online supplement.

The CART using nodule size, AAT, CEA, and SCC developed to predict diagnosis is provided in Figure E1. With 17 nodes, nine of which are terminal, this model is rather complex. The sensitivity, specificity, and positive and negative predictive values

TABLE 1. TRAINING SET: PATIENT DEMOGRAPHICS AND CLINICAL PROFILES

Demographic	Cancer (*n* = *298*)	Control (*n* = *211*)
Age, yr*	63.8 ± 10.6	62.5 ± 11.9
Range	28–84	23–84
Sex, number of patients*		
Male	108	66
Female	113	69
Stage, number of patients		
I	122	
II	40	
III	77	
IV	47	
Unknown	12	
Histology, number of patients		
Adenocarcinoma	145	
Adenosquamous cell carcinoma	3	
Bronchioloalveolar carcinoma	2	
Squamous cell carcinoma	74	
Large-cell carcinoma	13	
Small-cell carcinoma (limited)	2	
Non–small-cell carcinoma, unspecified	40	
Adeno/bronchioloalveolar carcinoma	11	
Large-cell/adenocarcinoma	1	
Large-cell neuroendocrine carcinoma	1	
Large-cell/small-cell carcinoma	1	
Small-cell/squamous cell carcinoma	1	
Not specified	4	

*These numbers were determined without 153 deidentified specimens provided by the Mayo Clinic (n = 153; lung cancer = 77, no cancer = 76).

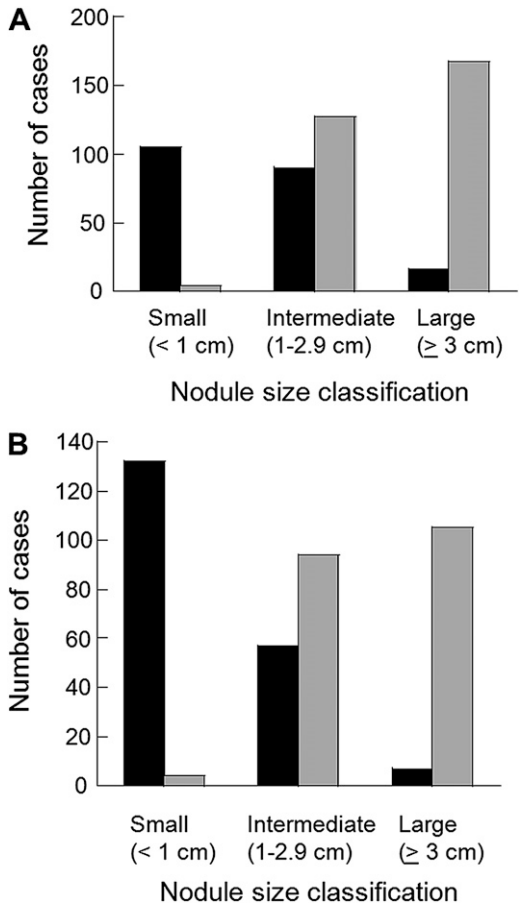

Figure 1. Distribution of benign nodules (*black bars*) and malignant nodules (*gray bars*) according to lesion size in the (*A*) training and (*B*) validation sets.

associated with this model within the training set are 90%, 82%, 88%, and 86%, respectively.

The logistic regression model used the same four features (nodule size, AAT, CEA, and SCC) to predict the diagnosis. The model, along with odds ratio estimates for 1 unit change in a predictor, is shown in Table E2. If a patient with a calculated cancer probability of 0.5 or higher is considered to have cancer and one with a probability less than 0.5 is considered not to have cancer, the overall sensitivity, specificity, and positive and negative predictive values of the model are 82%, 84%, 88%, and 76%, respectively. If the calculated cancer probability threshold is changed to 0.3 or higher, the overall sensitivity, specificity, and positive and negative predictive values are 94%, 63%, 78%, and 89%, respectively.

The overall logistic regression model was applied to samples within the three nodule size categories (small, <1 cm; intermediate, 1 to <3 cm; and large, ⩾3 cm). The sensitivity, specificity, and positive and negative predictive values within each of these size categories in the training set are shown in Table 2.

Validation Study Results

A total of 399 patients fulfilled the eligibility criteria and were included in the independent validation study. The diagnosis of primary lung cancer was established in 203 patients (50.8%) and a benign abnormality in 196 (49.2%). One patient with a slow-growing lesion designated as without lung cancer was later diagnosed with a mucosa-associated lymphoid tissue lymphoma. She has been followed for more than 2 years without additional therapy or evidence of disease. Patient demographics for the validation study are shown in Table 3.

The distribution of benign and malignant nodules according to lesion size is shown in Figure 1B. The validation study dataset is provided in Table E3.

LCBA data and nodule size were entered into the CART algorithm derived from the training set, and a "lung cancer" (high-risk) or "no lung cancer" (low-risk) diagnosis was assigned for each patient. The sensitivity, specificity, and positive and negative predictive values for determining which patient had lung cancer in the validation study were 88%, 82%, 84%, and 87%, respectively.

LCBA data and nodule size were entered into the logistic regression model derived from the training set, and a "lung cancer" (high-risk) or "no lung cancer" (low-risk) diagnosis was assigned for each patient. Assuming a probability threshold of 0.5 for assigning a patient to the high-risk group, the sensitivity, specificity, and positive and negative predictive values for all patients in the validation study were 80%, 89%, 89%, and 81%, respectively. Assuming a threshold of 0.3, these estimates are 92%, 74%, 79%, and 90%, respectively. The sensitivity, specificity, and positive and negative predictive values according to nodule size category in the validation study are shown in Table 4.

To investigate the added value of the serum markers as compared with nodule size alone, the sensitivity, specificity, and positive and negative predictive values according to nodule size category in the validation study with sequential addition of the serum markers is shown in Table E4. The likelihood ratio test comparing nested models is shown in Table E5; this analysis found that CEA added significant value to nodule size alone

TABLE 2. LOGISTIC REGRESSION MODEL PERFORMANCE WITHIN SIZE CATEGORIES OF THE TRAINING SET

Nodule Size Range	Cancer (*n*)	No Cancer (*n*)	Measure	LR Model with 0.5 Threshold	LR Model with 0.3 Threshold
Large	167	16	Sensitivity	100%	100%
			Specificity	0%	0%
			PPV	91%	91%
			NPV	NE	NE
Intermediate	127	90	Sensitivity	60%	87%
			Specificity	81%	47%
			PPV	82%	70%
			NPV	59%	72%
Small	4	105	Sensitivity	0%	75%
			Specificity	99%	87%
			PPV	0%	17%
			NPV	96%	99%

Definition of Abbreviations: LR = logistic regression; NE = not estimable; NPV = negative predictive value; PPV = positive predictive value.

TABLE 3. VALIDATION SET: PATIENT DEMOGRAPHICS AND CLINICAL PROFILES

Demographic	Cancer ($n = 203$)	Control ($n = 196$)
Age, yrs	68.6 ± 10.1	58.9 ± 12.8
Range	39–100	26–88
Sex		
Male	121	83
Female	82	113
Stage		
I	94	
II	36	
III	42	
IV	27	
Unknown	4	
Histology		
Adenocarcinoma	94	
Adenosquamous cell carcinoma	1	
Bronchioloalveolar carcinoma	5	
Squamous cell carcinoma	70	
Large-cell carcinoma	6	
Small-cell carcinoma	1	
Non–small-cell carcinoma, unspecified	20	
Basaloid carcinoma	1	
Large-cell neuroendocrine carcinoma	3	
Pleomorphic carcinoma	1	
Squamous cell and adenocarcinoma	1	

($P < 0.001$). The addition of AAT and SCC to the model with nodule size and CEA was also statistically significant ($P = 0.04$).

DISCUSSION

A pulmonary nodule or focal pulmonary abnormality on imaging studies is a common radiographic finding (7, 8). With improvements in spatial resolution on CT, the number of patients with pulmonary nodules continues to rise. In the recent National Lung Screening Trial report, more than 24% of CT-screened participants had a pulmonary abnormality necessitating further evaluation because of concern for lung cancer (4). All of these indeterminate abnormalities create an undesirable burden on the healthcare system, because each lesion must be evaluated, with the understanding that most are benign and the prevalence of lung cancer is low. Current guidelines focus on small pulmonary nodules and recommend multiple sequential imaging studies to assess for growth, additional imaging with fluorodeoxyglucose (FDG) PET, or an invasive procedure, depending on risk for disease, clinical presentation, and size of the lesion. More recent data examining ground glass nodules suggest that these lesions typically have a more indolent behavior, and additional guidelines for these abnormalities are being developed by the Fleischner Society (9–12).

The use of tumor-specific, noninvasive biomarkers could modify this paradigm. From an imaging perspective, FDG-PET was introduced for the purpose of distinguishing a benign from malignant focal pulmonary abnormality (13, 14). However, because of the predominately small size of nodules, PET remains cost prohibitive and impractical. More recent advances in technology have prompted an interest in the discovery of biomarkers to complement anatomic imaging. However, translating technology into clinical practice is not an easy proposition, particularly given the heterogeneity of tumors.

We previously reported on a panel of serum markers discovered by a proteomic expression platform to be used for the diagnosis of lung cancer (2). The current study is an extension of that work, and focuses on using a combination of these markers with the most predictive imaging feature of lung cancer, nodule size, to stratify patients with indeterminate nodules into low- and high-risk groups. This risk designation is then intended to provide additional information about the likelihood of lung cancer and help clinicians as part of their decision process in guiding patient management.

This study developed several models using CART and logistic regression analysis. With the CART model we were able to assign 85% of all patients with lung cancer to the high-risk group, and 83% of all patients with benign nodules to the low-risk group. Those individuals assigned to a high-risk category could have more immediate workup with a biopsy, PET, or close surveillance. Patients in the low-risk group could be followed with less frequent sequential imaging to assess growth. It is interesting to note that in this algorithm three features (nodule size, CEA, and AAT) were needed to achieve a reasonable accuracy. This is consistent with studies that suggest only a few classifiers may be needed, and that additional features may not provide significant benefit (15, 16). The serum biomarker data need to be evaluated in the context of the clinical presentation.

Similarly, the logistic regression model assuming a threshold of 0.5 for risk assignment placed 80% of all patients with lung cancer in the high-risk category, and almost 90% of the patients with benign nodules in the low-risk category.

Ultimately these results need to be distilled and translated into clinical practice. We envision that for patients with a pulmonary nodule and a concern for lung cancer, the LCBA could be ordered and the results of this test, in conjunction with nodule size and clinical presentation, could lead to an efficient evaluation strategy. The laboratory will combine serum assay results with nodule size, and return to the clinician a high- or low-risk result.

TABLE 4. LOGISTIC REGRESSION MODEL PERFORMANCE WITHIN SIZE CATEGORIES OF THE VALIDATION SET

Nodule Size Range	Cancer (n)	No Cancer (n)	Measure	Logistic with 0.5 Threshold	Logistic with 0.3 Threshold
Large	105	7	Sensitivity	100%	100%
			Specificity	0%	0%
			PPV	94%	94%
			NPV	NE	NE
Intermediate	94	57	Sensitivity	60%	86%
			Specificity	81%	42%
			PPV	84%	71%
			NPV	55%	65%
Small	4	132	Sensitivity	25%	25%
			Specificity	98%	92%
			PPV	25%	8%
			NPV	98%	98%

Definition of Abbreviations: NE = not estimable; NPV = negative predictive value; PPV = positive predictive value.

With a clear understanding of risk of lung cancer, general guidelines for further management could be constructed.

Given the current data one could envision integrating the biomarkers with nodule size to guide management strategy, realizing that additional studies are needed to validate this type of approach. In the small nodule category, there were very few lung cancers diagnosed, and as expected by the low prevalence of malignancy in this size category, the standard logistic regression model threshold assigned most patients to the low-risk group. Only 2% of individuals in the low-risk group are expected to be diagnosed with lung cancer; these patients may not need as close surveillance. In those few individuals who eventually demonstrate nodule growth and are diagnosed with lung cancer, the risk of cancer progression to more advanced stage disease during the observation period seems low. Although there were only four small (<1 cm) lung cancers in the validation dataset, the three that were assigned to the low-risk group had their lesions followed before surgical resection (4 mo to 4 yr); all had early stage disease; none were treated with adjuvant therapy; and none have developed recurrence at least 2 years after the diagnosis. Approximately 3% (4 of 136) of individuals in the small-size category were assigned to the high-risk group, and approximately 25% of these patients will eventually be diagnosed with lung cancer; in these individuals a closer follow-up CT could be performed.

In the intermediate category, by using the logistic regression model with the 0.3 threshold, the numbers of patients with lung cancer assigned to the low-risk category are minimized. Eighty-six percent (86%) of all lung cancers in this size category were placed in the high-risk group, and 71% of patients in the high-risk category were diagnosed with lung cancer. Individuals assigned to this category could have an immediate FDG-PET or biopsy.

Individuals with lesions in the intermediate-size category assigned to the low-risk group could have an initial short-term follow-up CT to assess for growth, with the understanding that approximately one-third of individuals in this group will eventually be diagnosed with lung cancer. Alternatively, an immediate FDG-PET study could be performed.

In the large nodule size category, the prevalence of lung cancer is high. Some studies estimate that more than 90% of patients with larger than 3 cm lesions will have lung cancer (17). None of the algorithms were sufficient to exclude lung cancer in this group, and it does not seem that the biomarkers assayed in this study will add significantly to the decision process, although all patients in the validation study with a lesion greater than or equal to 3 cm and an elevated CEA (>3.22 ng/ml) had lung cancer (100% specificity, 68% sensitivity). Patients in the large-size category could have an FDG-PET scan, or an immediate biopsy (or resection), particularly if CEA is elevated. Alternatively, if there is an atypical clinical presentation and a biopsy does not show cancer, a low CEA may give more weight to a more conservative short-term follow-up approach, rather than taking the patient directly to surgery.

The current study suggests that integration of serum biomarker data with nodule size may complement current diagnostic strategies in the appropriate clinical context as long as the risks and benefits are understood. Because this study was conducted in the context of a tertiary care setting, additional studies are needed to determine the impact of integrating serum biomarker data with nodule size on a screening program. We believe a diagnostic strategy should minimize the number of patients with lung cancer placed in a low-risk group, but it does not seem that these patients, as categorized using our approach, will see a rapid progression of disease and no significant impact on long-term survival is expected. The proposed conservative strategy places some individuals without cancer in the high-risk category, but the number of additional diagnostic studies as compared with current standard of care will not change.

The concept of integrating serum biomarkers with imaging findings has enormous potential (18–20). With the addition of biomarkers, a more efficient approach may be possible, although future studies need to address the frequency and use of sequential follow-up imaging or repeat serum marker evaluation. As we move forward with this type of combined diagnostic approach, additional markers will emerge and more efficient strategies can be developed to improve clinical management of these patients.

Author disclosures are available with the text of this article at www.atsjournals.org.

References

1. MacMahon H, Austin JH, Gamsu G, Herold CJ, Jett JR, Naidich DP, Patz EF Jr, Swensen SJ. Guidelines for management of small pulmonary nodules detected on CT scans: a statement from the Fleischner Society. *Radiology* 2005;237:395–400.
2. Patz EF Jr, Campa MJ, Gottlin EB, Kusmartseva I, Guan XR, Herndon JE II. Panel of serum biomarkers for the diagnosis of lung cancer. *J Clin Oncol* 2007;25:5578–5583.
3. Midthun DE, Swensen SJ, Jett JR, Hartman TE. Evaluation of nodules detected by screening for lung cancer with low dose spiral computed tomography. *Lung Cancer* 2003;41:S40.
4. Aberle DR, Adams AM, Berg CD, Black WC, Clapp JD, Fagerstrom RM, Gareen IF, Gatsonis C, Marcus PM, Sicks JD. Reduced lung-cancer mortality with low-dose computed tomographic screening. *N Engl J Med* 2011;365:395–409.
5. Breiman L. Classification and regression trees. Belmont, CA: Wadsworth International Group; 1984.
6. Steinberg D, Golovnya M, Tolliver D. Cart for windows user guide. San Diego, CA: Salford Systems; 2002.
7. Brandman S, Ko JP. Pulmonary nodule detection, characterization, and management with multidetector computed tomography. *J Thorac Imaging* 2011;26:90–105.
8. Weir G, Kos S, Burrill J, Salat P, Ho S, Liu D. Diagnosis and management of the solitary pulmonary nodule. *BMJ* 2011;343:d4866.
9. Goo JM, Park CM, Lee HJ. Ground-glass nodules on chest CT as imaging biomarkers in the management of lung adenocarcinoma. *AJR Am J Roentgenol* 2011;196:533–543.
10. Kim HK, Choi YS, Kim J, Shim YM, Lee KS, Kim K. Management of multiple pure ground-glass opacity lesions in patients with bronchioloalveolar carcinoma. *J Thorac Oncol* 2010;5:206–210.
11. Kim HY, Shim YM, Lee KS, Han J, Yi CA, Kim YK. Persistent pulmonary nodular ground-glass opacity at thin-section CT: histopathologic comparisons. *Radiology* 2007;245:267–275.
12. Lee HY, Lee KS. Ground-glass opacity nodules: histopathology, imaging evaluation, and clinical implications. *J Thorac Imaging* 2011;26:106–118.
13. Erasmus JJ, Patz EF Jr. Positron emission tomography imaging in the thorax. *Clin Chest Med* 1999;20:715–724.
14. Patz EF, Lowe VJ, Hoffman JM, Paine SS, Burrowes P, Coleman RE, Goodman PC. Focal pulmonary abnormalities: evaluation with F-18 fluorodeoxyflucose pet scanning. *Radiology* 1993;188:487–490.
15. Baker SG. Gene signatures revisited. *J Natl Cancer Inst* 2012;104:262–263.
16. Hand D. Classifier technology and the illusion of progress. *Stat Sci* 2006; 21:1–14.
17. Midthun DE, Swensen SJ, Jett JR. Clinical strategies for solitary pulmonary nodule. *Annu Rev Med* 1992;43:195–208.
18. Aloia T, Bepler G, Harpole D, Goodman PC, McAdams HP, Erasmus JJ, Herndon JE, Patz EF Jr. Integration of peripheral blood biomarkers with computed tomography to differentiate benign from malignant pulmonary opacities. *Cancer Detect Prev* 2001;25:336–343.
19. Patz EF Jr. Integration of biomarkers and imaging. *J Thorac Oncol* 2006; 1:78–80.
20. Bigbee WL, Gopalakrishnan V, Weissfeld JL, Wilson DO, Dacic S, Lokshin AE, Siegfried JM. A multiplexed serum biomarker immunoassay panel discriminates clinical lung cancer patients from high-risk individuals found to be cancer-free by CT screening. *J Thorac Oncol* 2012;7:698–708.