REVIEW ARTICLE

Edward W. Campion, M.D., *Editor*

Cancer of Unknown Primary Site

Gauri R. Varadhachary, M.D., and Martin N. Raber, M.D.

From the Department of Gastrointestinal Medical Oncology, University of Texas M.D. Anderson Cancer Center, Houston. Address reprint requests to Dr. Varadhachary at the Department of Gastrointestinal Medical Oncology, Unit 426, University of Texas M.D. Anderson Cancer Center, 1515 Holcombe Blvd., Houston, TX 77030-4009, or at gvaradha@mdanderson.org.

N Engl J Med 2014;371:757-65.
DOI: 10.1056/NEJMra1303917

Cancer of unknown primary site is a heterogeneous group of cancers for which the anatomical site of origin remains occult after detailed investigations.[1,2] The emergence of sophisticated imaging, immunohistochemical testing, and molecular-profiling tools has influenced our approach to unknown primary cancer, although it has also increased the ambiguity of designations for this disorder. In the era of tailored therapeutic strategies, this situation presents both an opportunity and a challenge.

The past four decades have seen a shift in our understanding of unknown primary cancer (Fig. 1). First, improved imaging techniques increased our confidence in the classification of some cancers as having an occult primary origin. Later, subsets of unknown primary cancers with an apparently favorable prognosis were identified, primarily on the basis of histopathological findings, the pattern of spread, and serum markers.[2] Subsequently, with the advent of new immunohistochemical markers and advances in diagnostic pathological tests, tissue-of-origin profiles were described that assigned additional putative primary sites to unknown primary cancer on the basis of immunohistochemical patterns.[3-6] Current research involves the application of proteomic and genomic tools to unknown primary cancer.

Cancer of unknown primary site was once viewed almost as a separate type of cancer, with the assumption that, regardless of the site of origin, the tumors in unknown primary cancers shared biologic properties, perhaps including rapid progression and dissemination, which contributed to their presentation. This view drove the conduct of phase 2 empirical trials over the past three decades, with the goal of developing standard chemotherapy regimens that would be effective in all patients with unknown primary cancer. The underlying assumption was that variations in presentation would not have a substantial effect on therapeutic approaches or survival.

Our view of unknown primary cancer has evolved as our understanding of cancer biology in general has matured to become much more personalized. Many people now believe that tumors in unknown primary cancer may retain the signature of the putative primary origin and that extending the management of known cancers to subtypes of unknown primary cancer can contribute to advancements in therapies for this disease. Cancer of unknown primary site could even be seen as the epitome of personalized medicine, with individualized treatment driven by the mutational status of each patient.

The biologic events that allow the primary site to remain obscure after the development of metastases have not yet been defined. Studies that have shown chromosomal abnormalities, microvessel density, aneuploidy, and overexpression of several genes suggest that these abnormalities are not unique to unknown primary cancer.[7-11] With the use of the Sequenom MassARRAY platform, a study involving consecutive patients with unknown primary cancer showed a low rate of mutations (in 18% of patients).[12] No new, low-frequency mutations were found with the use of a panel of mutations involving the phosphatidylinositol 3-kinase

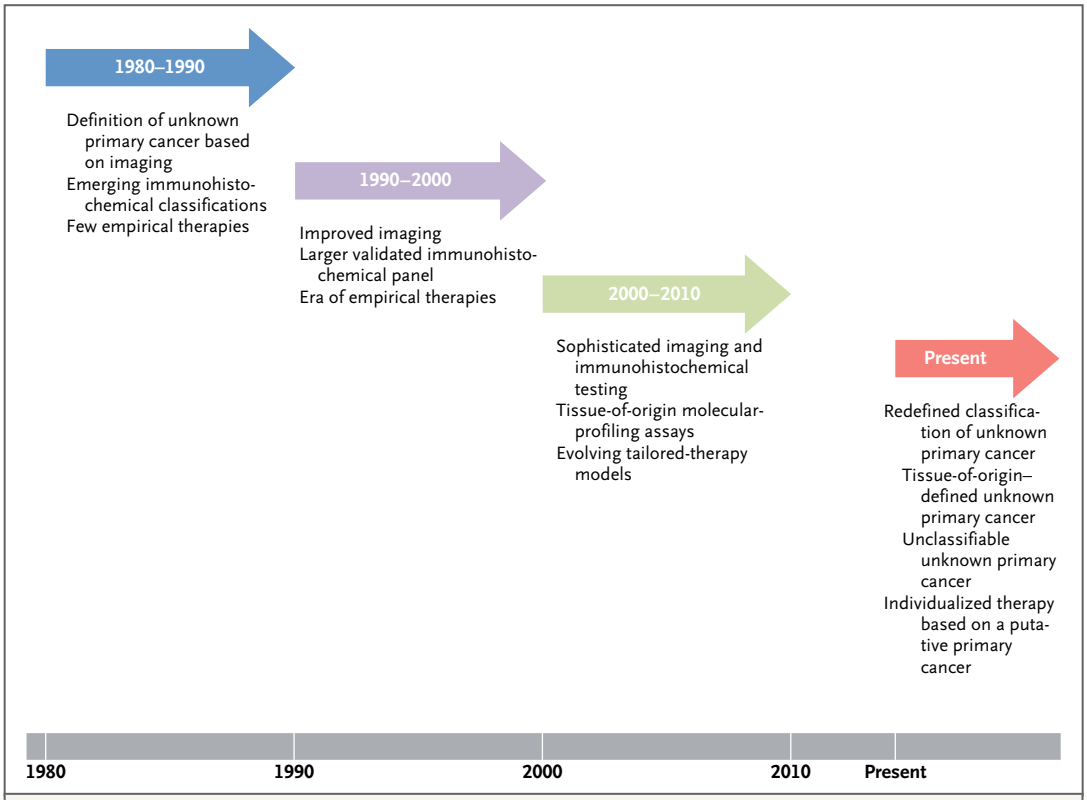

Figure 1. Classification of Unknown Primary Cancer through the Decades.

Over the years, we have seen an improvement in diagnostic methods, including imaging, pathological testing, and molecular markers. These advances have helped define the current taxonomy of unknown primary cancer.

(PI3K)–AKT pathway, MEK pathway, receptors, and downstream effectors. Furthermore, there are major obstacles to conducting the trials that would be required to show definitively that unknown primary cancer with a putatively identified source behaves the same way as metastatic disease with a similar, known primary site.

CLINICAL EVALUATION

FOCUSED IMAGING

In the absence of contraindications, a baseline computed tomographic (CT) scan of the chest, abdomen, and pelvis with the use of intravenous contrast material is the standard of care, as supported by the National Comprehensive Cancer Network and National Institute for Health and Clinical Excellence radiology guidelines for unknown primary cancer.[13,14] Patients should then be approached in a directed fashion.[15] Currently, magnetic resonance imaging (MRI) of the breasts is indicated in women presenting with isolated axillary adenopathy and adenocarcinoma, if findings on mammography and ultrasonography are negative. The absence of a breast mass on MRI is associated with a low probability of finding a tumor at mastectomy.[16-18] Invasive testing (with bronchoscopy, upper endoscopy, colonoscopy, etc.) should be limited to symptomatic patients and to those with imaging or pathological abnormalities indicative of a primary cancer, since these patients may have a higher yield, as compared with asymptomatic patients without clinicopathological abnormalities, in efforts to detect a primary cancer.

CURRENT ROLE OF PET-CT IMAGING

In patients who have renal insufficiency or who cannot take iodine, positron-emission tomography (PET)–CT or MRI can be used. Elective use of PET-CT is currently limited to patients with squamous-cell lymphadenopathy of the neck (cer-

vical carcinoma of unknown primary site).[19-21] In these patients, PET-CT may help guide the biopsy, determine the extent of disease, facilitate the planning of radiation therapy, and help with surveillance. These patients are also candidates for pan-endoscopy (indirect and direct laryngoscopy, bronchoscopy, and upper endoscopy) and staging bilateral tonsillectomies.[22]

Apart from the above indication, the role of PET-CT is unclear. Several small studies have evaluated the usefulness of PET in patients with unknown primary cancer. Moller et al. reviewed ^{18}F-fluorodeoxyglucose (FDG) PET as a diagnostic test in patients with extracervical cancer of unknown primary site.[23] They identified four studies (involving a total of 152 patients), which were retrospective and heterogeneous with respect to inclusion criteria, study design, and diagnostic workup before the use of FDG-PET–CT. The primary tumor was detected by means of FDG-PET–CT in 39% of the patients with extracervical cancer of unknown primary site. The lung was the most commonly detected primary tumor site (in approximately 50% of the patients in whom the tumor was detected). The pooled estimates of the sensitivity, specificity, and accuracy of FDG-PET–CT in the detection of the primary tumor site were 87%, 88%, and 88%, respectively. The authors concluded that FDG-PET–CT may have a role in identification of the primary tumor in extracervical cancer of unknown primary site; however, prospective studies with more uniform inclusion criteria are warranted.

Although they have not been studied prospectively, PET-CT scans may be useful in selected patients with solitary metastases before definitive locoregional therapies and in follow-up of patients with disease predominantly involving bone. Figure 2 shows the challenges in evaluating small primary renal cancers that can be missed with a PET scan. Even with the sophisticated imaging available today, some very small primary sites remain occult; in the future, however, better technologies may reveal very small cancers and challenge unknown primary cancer as an entity.

SERUM TUMOR MARKERS

Tumor markers are generally not considered to be diagnostic, and among the adenocarcinoma markers, there is considerable variability. Elevated levels of carcinoembryonic antigen or cancer antigens 125, 19-9, and 27.29 are nonspecific and not helpful in identifying the primary tumor site. In men who present with adenocarcinoma and osteoblastic metastases, a prostate-specific antigen (PSA) test is recommended. Elevated levels of the beta subunit of human chorionic gonadotropin (hCG) and alpha-fetoprotein in men with undifferentiated or poorly differentiated carcinoma (especially those with a midline tumor) suggest the possibility of an extragonadal germ-cell (testicular) tumor.[24] Alpha-fetoprotein should also be considered in patients with a potential diagnosis of hepatoma. Although tumor markers are not particularly helpful in diagnosing a specific primary tumor, they may be helpful in monitoring the response to treatment.

PATHOLOGICAL FEATURES AND MOLECULAR PROFILING

General Considerations

In most patients with unknown primary cancer, pathological findings supersede the interpretations of radiologic testing. Adequate tissue sampling, ideally by means of a core biopsy, is essential, as is communication between the treating oncologist and the pathologist. Most therapeutic phase 2 trials have defined unknown primary cancer as limited to epithelial cancers. In these trials, patients with metastatic lymphomas, melanomas, and sarcomas who presented without a known primary tumor were excluded, since management of these cancers is based on the specific stage and histologic findings. In practice, however, one must consider the expanded differential diagnosis, including nonepithelial tumors, when dealing with an unclassified cancer.

Light Microscopy

On light microscopy, cancers of unknown primary site include well-differentiated and moderately differentiated adenocarcinoma (in 60% of patients), poorly differentiated carcinoma or adenocarcinoma (in 30%), poorly differentiated or undifferentiated malignant neoplasm (in 5%), and squamous-cell carcinoma (in 5%). In rare cases, patients with unknown primary cancer present with neuroendocrine cancer or mixed tumors, including sarcomatoid, basaloid, and adenosquamous carcinomas.[25]

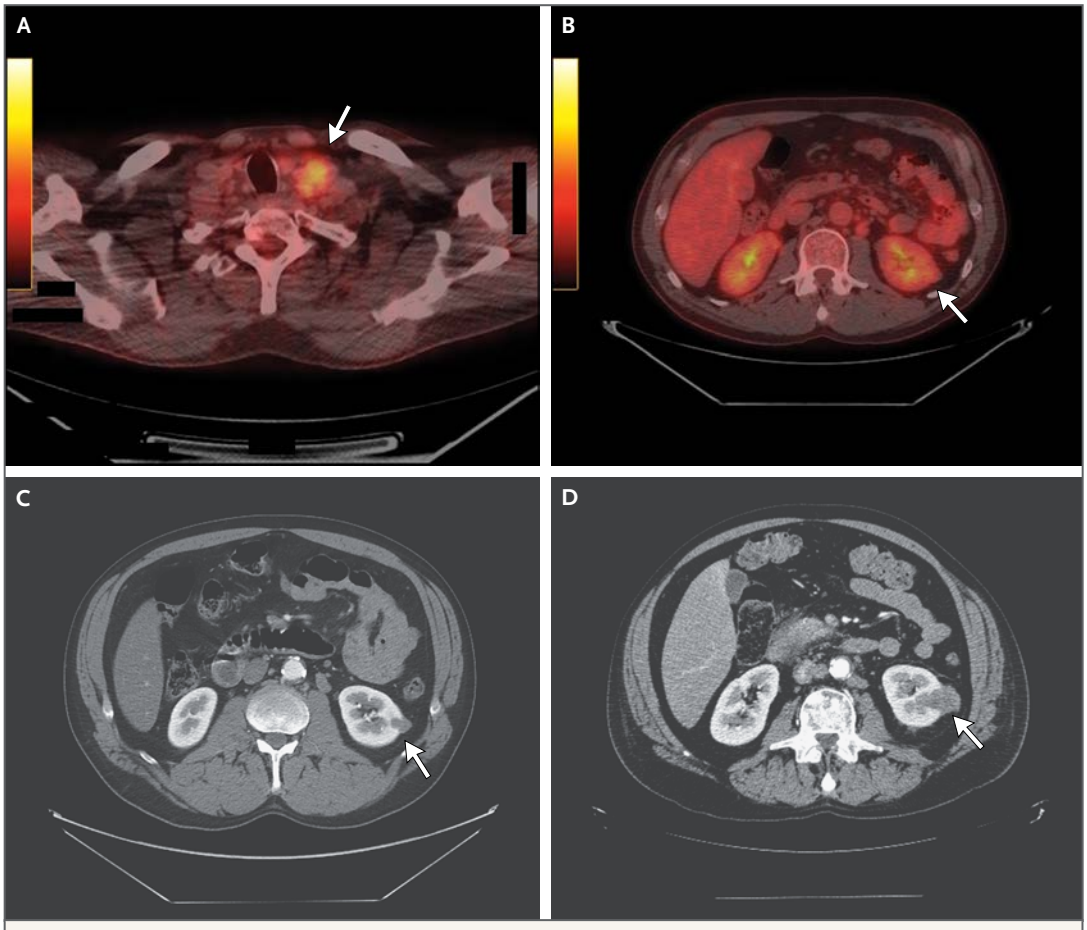

Figure 2. Drawback of Baseline ^{18}F-fluorodeoxyglucose Positron-Emission Tomography (PET)–CT as the Initial Imaging Method in Unknown Primary Cancer.

A 51-year-old man who was a smoker presented with neck adenopathy. Biopsy of the left supraclavicular lymph node revealed metastatic, poorly differentiated carcinoma. Immunostains were negative for cytokeratin (CK) 7, CK20, synaptophysin, chromogranin, S-100, melanoma antigen recognized by T cells (MART-1), prostate-specific antigen, thyroid transcription factor 1 (TTF1), inhibin, and thyroglobulin. The tumor was focally positive for Hep Par-1, CD10, and low-molecular-weight keratin, and final pathological results were reported as nonspecific. A PET-CT scan that was ordered as the baseline study in the head and neck oncology clinic showed multiple hypermetabolic nodes in the neck (Panel A, arrow). The PET-CT scan did not show a renal primary cancer, although in retrospect there was a hint of a small lesion (Panel B, arrow). The patient received chemotherapy with paclitaxel and carboplatin for unknown primary cancer favoring a lung-cancer profile. He had mild disease progression while receiving this regimen. In parallel, he underwent a tissue-of-origin molecular-profiling study that showed a kidney-cancer profile. Additional renal-specific immunohistochemical testing on the nodal tissue showed the tumor to be positive for PAX-8, renal-cell carcinoma, CD10, epithelial membrane antigen, and vimentin — findings that are consistent with conventional-type, metastatic renal-cell carcinoma. A CT scan obtained with the use of intravenous contrast material showed a mass (1.0 by 1.2 cm) in the lower pole of the left kidney (Panel C, arrow). The patient was treated with targeted therapies, including everolimus, axitinib, and pazopanib; he had a mixed response initially, followed by disease progression in lymph nodes, liver, bones, and the primary site (Panel D, arrow). This patient did not have unknown primary cancer on presentation; instead, he had metastatic renal-cell cancer that had been evaluated with a suboptimal workup. Unfortunately, even with accurate diagnosis, directed therapies do not have a clear therapeutic effect in most patients with advanced renal-cell cancer.

Immunohistochemical Testing

The use of immunohistochemical testing in unknown primary cancer is based on the premise that there is concordance in the expression profiles of primary and metastatic cancers. Immunohistochemical tests are typically tests of peroxidase-labeled antibodies against specific tumor antigens that help suggest the tumor lineage and can establish most lineages (carcinoma, lymphoma, sarcoma, melanoma, etc.). Most researchers believe that a search for the putative primary cancer, performed by means of immunohistochemical testing, is helpful in detecting tumors with a favorable prognosis and in planning a tailored therapy for the patient. Although individual immunohistochemical tests have modest specificity and sensitivity (with the possible exception of the PSA test), their predictive value may improve with grouping and recognition of patterns that are strongly indicative of specific tumors.[26] For example, the phenotype for positive thyroid transcription factor 1 (TTF1), with positive cytokeratin 7 (CK7), and the phenotype for positive cytokeratin 20, with positive homeobox protein CDX-2 and negative CK7, have been reported as very suggestive of lung and lower gastrointestinal cancer profiles retrospectively, although they have not been validated prospectively in the absence of a primary cancer. With the use of light microscopy and immunohistochemical testing, a single putative primary tumor may be assigned in up to 25% of cases of unknown primary cancer, and in the remaining cases, immunohistochemical testing is nonspecific.[27]

Currently, we lack a tiered and uniform approach to performing the stains. Additional limitations of immunohistochemical testing include factors affecting tissue antigenicity, interobserver and intraobserver variability in interpretation, and tissue heterogeneity and inadequacy. Most important, the clinical efficacy of immunohistochemical test–based management of unknown primary cancer has not been shown adequately. In one retrospective study, patients with CDX-2–positive cancers who were treated with regimens used for gastrointestinal cancers had a survival of more than 30 months,[28] but prospective validation of the therapeutic effect of immunohistochemical test–directed therapies for putative primary tumors is urgently needed. A differential diagnosis based on immunohistochemical testing can prompt more focused biomarker studies with potential therapeutic effect or actionable targets, which may allow patients with unknown primary cancer to enroll in biomarker-based early-phase studies.

Tissue-of-Origin Molecular Profiling

The premise for studying tissue-of-origin molecular-profiling assays in unknown primary cancers is that, when a large number of genes from known cancers are examined with the use of tools such as DNA microarray or quantitative real-time polymerase-chain-reaction (rt-PCR) assay, metastatic tumors have molecular signatures that match their primary origin. The performance of tissue-of-origin molecular-profiling assays in known cancers has been validated with the use of independent, blinded evaluation of sets of tumor samples, with an accuracy of approximately 90%.[29-31] The feasibility of using formalin-fixed samples obtained from small, core-needle biopsy or using samples obtained by means of fine-needle aspiration makes this method practical for use in the clinic setting.

Tissue-of-origin assays based on messenger RNA (mRNA) or microRNA have been studied in prospective and retrospective trials involving patients with unknown primary cancer.[32,33] Most of the studies have evaluated assay performance, although the challenge with validating the accuracy of an assay for unknown primary cancer is that, by definition, the primary cancer diagnosis cannot be verified. Thus, current estimates of the accuracy of tissue-of-origin testing have relied on indirect metrics, including comparison with immunohistochemical testing, clinical presentation, and the appearance of latent disease at the primary site. With the use of these measures, the assays suggest a plausible primary site in approximately 70% of the patients studied.[34-37] In the remaining patients, the results are clearly discordant with the working differential diagnosis, the sample is insufficient despite repeat biopsy (an issue that occurs with bone samples), or the assay is unable to designate a primary origin from its panel of cancers.[38]

At present, the only outcomes-based study has been a prospective, single-group study evaluating the role of the 92-gene assay to predict the tissue-of-origin and assay-directed, site-specific therapy in patients with unknown primary cancer.[39] The investigators found that the median overall survival of 12.5 months (95% confidence interval, 9.1 to 15.4) among patients who received assay-directed, site-specific therapy compared favorably with the results of previous studies that used empirical therapy. Biliary and urothelial cancer profiles accounted for 33% of the predictions. Unfortunately, firm conclusions regarding therapeutic effect cannot be drawn from this study, given the nonrandomized design, statistical biases, confounding variables, including use of subsequent lines of (empirical) therapy, and the heterogeneity of unknown primary cancers.

Without randomized, controlled trials it is difficult to gauge the therapeutic effect of tissue-of-origin molecular-profiling assays. Creative trial designs are urgently needed in order to study subsets of unknown primary cancers and the effect of these assays on survival and quality of life of patients.

Two prospectively defined, blinded studies of difficult-to-diagnose primary cancers (several of them poorly differentiated cancers) have shown the cost-effectiveness of tissue-of-origin molecular profiling over immunohistochemical testing. Samples were evaluated by means of morphologic and immunohistochemical analysis or the tissue-of-origin molecular-profiling test. Accuracy was defined on the basis of comparison with the pathological features of a known primary cancer. In one study, the assay showed overall accuracy of 79% for tumor classification versus 69% for morphologic and immunohistochemical analysis ($P=0.02$).[40] The mean number of immunohistochemical stains used was 7.9 per case (range, 2 to 15). The other study had similar findings; the assay accurately identified 89% of the specimens, as compared with 83% accuracy with immunohistochemical testing ($P=0.01$).[41] In the subset of 33 patients with poorly differentiated or undifferentiated carcinoma, the accuracy was 91% with the assay versus 71% with immunohistochemical testing ($P=0.02$). These results have important implications for the management of unknown primary cancers and warrant a study of an integrated algorithm evaluating tissue-of-origin molecular profiling to complement the use of immunohistochemical testing in selected patients.

TREATMENT IN THE GENOMICS ERA

Despite its heterogeneity, unknown primary cancer traditionally has been treated largely as a single entity, primarily with platinum-based combination chemotherapies. Over the past two decades, several combination treatments have been evaluated, and these have led to a range of therapies available for patients with unknown primary cancer. Phase 2 studies of empirical regimens have shown response rates of 25 to 35% and survival ranging from 6 to 16 months.[42-44] Survival has been longer for patients with nodal, pleural, or serous peritoneal disease (14 to 16 months) than for patients with visceral metastatic disease (6 to 9 months). In most patients, the disease is disseminated and incurable. Additional prognostic factors guiding therapy decisions include lactate dehydrogenase and albumin levels, performance status, and number of sites of disease.[45]

Historically, the "favorable subset" designation was based on a presentation that overwhelmingly suggested a specific primary origin.[2] Patients who receive such a diagnosis often have a response to treatment that is based on the putative primary origin, and they may have prolonged survival and also a potential cure. These presentations (and their presumed primary origins) include adenocarcinoma in axillary lymph nodes in women (breast cancer), squamous-cell carcinoma in neck nodes (head and neck cancer), papillary or serous tumors in the peritoneal cavity in women (ovarian cancer), and poorly differentiated midline nodal disease in young men (germ-cell cancer), as well as metastatic neuroendocrine tumors and indolent, solitary metastases (the latter treated with definitive surgery, chemotherapy, radiation therapy, or a combination thereof). Some clinicians may view isolated or oligometastatic squamous-cell carcinoma in inguinal nodes as a favorable presentation, although the differential diagnosis is broader and includes anal, genitourinary, and gynecologic primary origins.

Case studies show the promise and challenge with determining the tissue of origin in

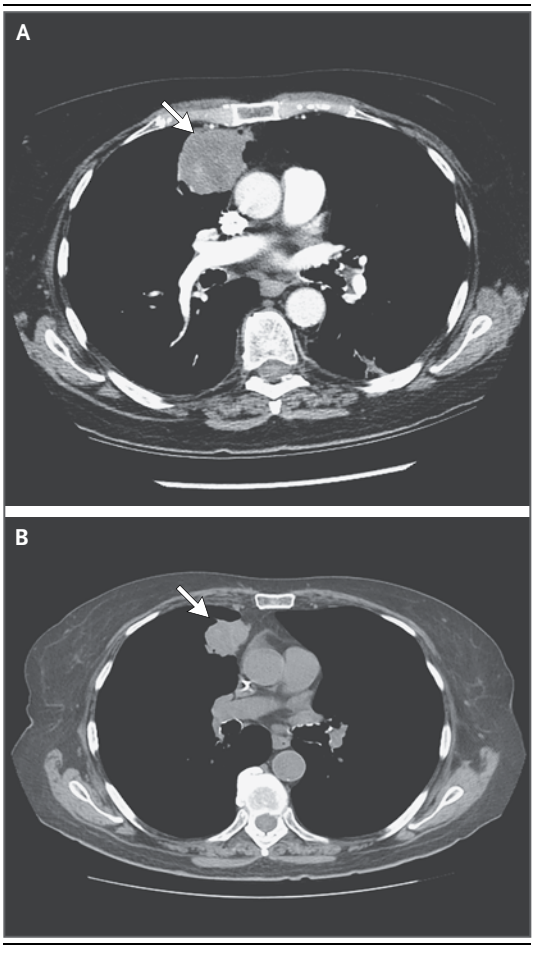

Figure 3. Discordant Immunohistochemical and Radiologic Findings in Assessing Unknown Primary Cancer.

The patient was a 77-year-old nonsmoking woman who was seen initially in the thoracic oncology department. CT of the chest that was performed as follow-up for pneumonia revealed a 5.7-cm "lung cancer" in the right upper lobe, with minor fissure and right-middle-lobe involvement (Panel A, arrow). Biopsy of the mass revealed a moderately differentiated adenocarcinoma, strongly and diffusely positive for homeobox protein CDX-2, CK20, villin, and carcinoembryonic antigen and negative for CK7 and TTF1. The pathology report concluded that the findings favored cancer of the lower gastrointestinal tract, including the appendix and colorectum. Upper endoscopy and colonoscopy showed no abnormalities. There was no clear evidence of a primary cancer in the small bowel or appendix. After a short course of preoperative fluorouracil and oxaliplatin, the patient underwent resection of the mass (Panel B, arrow). The final pathology report was unchanged (presumed metastatic colorectal cancer). The patient received the same chemotherapy after surgery. Subsequent colonoscopies were negative. The patient was enrolled in a microRNA tissue-of-origin clinical trial, and molecular assay confirmed a colon-cancer profile. Although the benefit of chemotherapy for this presentation is unknown, clinicians who provide care for patients with unknown primary cancer have to integrate the pathological information in therapy decisions. Common presentations of unknown primary cancer with a colon-cancer profile are isolated carcinomatosis and ovarian metastases.[46]

unknown primary cancer (Fig. 3 and 4). Patients with immunohistochemical test results suggesting a single diagnosis make up approximately 25% of patients with unknown primary cancer.[26,27,47] Examples include the TTF1-positive and CK7-positive lung-cancer profile; the CDX-2–positive, CK20-positive, and CK7-negative gastrointestinal-cancer profile; and the GCDFP (gross cystic disease fluid protein) 15–positive or mammaglobin-positive, CK7-positive breast-cancer profile. Frequently, the pattern of disease spread supports the immunohistochemical test results, and treatment algorithms are based on the putative primary origin. On occasion, the immunohistochemical and radiologic findings are discordant (Fig. 3A and 3B).

For the remaining 75% of patients, the differential diagnosis based on immunohistochemical testing is broad. Figure 4 shows a mass predominantly involving the liver that is suggestive of cholangiocarcinoma on the basis of radiologic findings. Pathological findings are typically nonspecific in such cases, and it is not unusual for cholangiocarcinoma to be called an unknown primary cancer. In patients without diagnostic (specific) results on immunohistochemical testing, a platinum-based regimen that is based on the clinicopathological presentation is often chosen, and protocol-based genomics studies, including tissue-of-origin molecular profiling and next-generation sequencing, may be useful. We lack specific and effective drugs for several cancer profiles, and treatments overlap for many cancers. However, as new therapies are developed for common known cancers, molecular tools for unknown primary cancers may be the cornerstone of decision making.

More broadly, there is an extensive push toward personalizing cancer care with the use of next-generation sequencing to identify driver mutations in individual tumors. Currently, we do not have a detailed understanding of the complex cross talk and signaling pathways involved in individual cancers. Vemurafenib, which targets the oncogenic *BRAF* V600E mutation, has

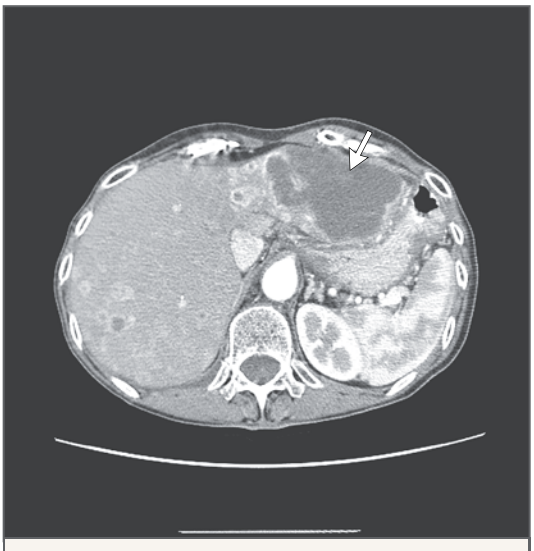

Figure 4. Distinguishing between Unknown Primary Cancer and Cholangiocarcinoma.

Given that hepatic presentation is common in unknown primary cancer, it is important to consider biliary cancers in the differential diagnosis of unknown primary cancers, especially in patients who have large, solitary hepatic masses with or without satellite nodules. Predominant liver lesions can be accompanied by bone, lung, or omental metastases. Multiphase scanning showing enhancement during the delayed phase is suggestive of a cholangiocarcinoma (arrow). Immunohistochemical testing is nondiagnostic, and a large battery of stains should be avoided in the evaluation of an unknown primary cancer.

single-agent activity in *BRAF*-mutated melanoma but not *BRAF*-mutated colorectal cancer.[48,49] This finding shows the important role of cellular context in therapeutics, and its role in unknown primary cancers requires further evaluation. Although the management of unknown primary cancer is changing at a rapid pace, until new technologies have been validated and are widely available, we should not lose sight of the fundamental principle that the use of focused clinicopathological testing and expert clinical judgment is critical in choosing the best therapies for patients.

FUTURE DIRECTIONS

Unfortunately, efforts to study unknown primary cancer with the use of collaborative research and new approaches have lagged behind efforts to study other solid-tumor types. Because of the heterogeneous presentations of unknown primary cancer, it is a challenge to adequately answer important questions involving new therapies, immunohistochemical testing, biologic features, and tissue-of-origin molecular profiling with the use of the traditional, prospective, phase 3 randomized designs. Innovative trial designs, the establishment of international consortia, and the application of genomic and proteomic techniques to subsets of unknown primary cancer will help us in our research efforts as we continue to expand therapeutic options to patients. The success of the next-generation sequencing approach will require both additional molecular insights and new drugs that are effective against specific mutations. Should this approach prove effective, the treatment of unknown primary cancers may merge with that of known primary cancers.

Disclosure forms provided by the authors are available with the full text of this article at NEJM.org.

REFERENCES

1. Briasoulis E, Tolis C, Bergh J, Pavlidis N. ESMO Minimum Clinical Recommendations for diagnosis, treatment and follow-up of cancers of unknown primary site (CUP). Ann Oncol 2005;16:Suppl 1:i75-i76.
2. Greco FA, Hainsworth JD. Cancer of unknown primary site. In: DeVita VT Jr, Hellman S, Rosenberg SA, eds. Cancer: principles and practice of oncology. 8th ed. Philadelphia: Lippincott Williams & Wilkins, 2008:2363-87.
3. Rubin BP, Skarin AT, Pisick E, Rizk M, Salgia R. Use of cytokeratins 7 and 20 in determining the origin of metastatic carcinoma of unknown primary, with special emphasis on lung cancer. Eur J Cancer Prev 2001;10:77-82.
4. Jagirdar J. Application of immunohistochemistry to the diagnosis of primary and metastatic carcinoma to the lung. Arch Pathol Lab Med 2008;132:384-96.
5. DeYoung BR, Wick MR. Immunohistologic evaluation of metastatic carcinomas of unknown origin: an algorithmic approach. Semin Diagn Pathol 2000;17:184-93.
6. Dennis JL, Hvidsten TR, Wit EC, et al. Markers of adenocarcinoma characteristic of the site of origin: development of a diagnostic algorithm. Clin Cancer Res 2005;11:3766-72.
7. Pavlidis N, Briassoulis E, Bai M, Fountzilas G, Agnantis N. Overexpression of C-myc, Ras and C-erbB-2 oncoproteins in carcinoma of unknown primary origin. Anticancer Res 1995;15:2563-7.
8. Briasoulis E, Tsokos M, Fountzilas G, et al. Bcl2 and p53 protein expression in metastatic carcinoma of unknown primary origin: biological and clinical implications: a Hellenic Co-operative Oncology Group study. Anticancer Res 1998;18:1907-14.
9. Hainsworth JD, Lennington WJ, Greco FA. Overexpression of Her-2 in patients with poorly differentiated carcinoma or poorly differentiated adenocarcinoma of unknown primary site. J Clin Oncol 2000;18:632-5.
10. Hillen HF, Hak LE, Joosten-Achjanie SR, Arends JW. Microvessel density in unknown primary tumors. Int J Cancer 1997;74:81-5.
11. Karavasilis V, Malamou-Mitsi V, Briasoulis E, et al. Angiogenesis in cancer of

unknown primary: clinicopathological study of CD34, VEGF and TSP-1. BMC Cancer 2005;5:25.
12. Hale KS, Wang H, Karanth S, et al. Mutation profiling in patients with carcinoma of unknown primary using the Sequenom MassARRAY system. J Clin Oncol 2012;30:Suppl. abstract.
13. National Comprehensive Cancer Network. Occult primary (cancer of unknown primary), version 1. 2013 (http://www.nccn.org/professionals/physician_gls/f_guidelines.asp#site).
14. Taylor MB, Bromham NR, Arnold SE. Carcinoma of unknown primary: key radiological issues from the recent National Institute for Health and Clinical Excellence guidelines. Br J Radiol 2012;85:661-71.
15. Varadhachary GR. Carcinoma of unknown primary: focused evaluation. J Natl Compr Canc Netw 2011;9:1406-12.
16. Schelfout K, Kersschot E, Van Goethem M, et al. Breast MR imaging in a patient with unilateral axillary lymphadenopathy and unknown primary malignancy. Eur Radiol 2003;13:2128-32.
17. Olson JA Jr, Morris EA, Van Zee KJ, Linehan DC, Borgen PI. Magnetic resonance imaging facilitates breast conservation for occult breast cancer. Ann Surg Oncol 2000;7:411-5.
18. Lu H, Xu YL, Zhang SP, et al. Breast magnetic resonance imaging in patients with occult breast carcinoma: evaluation on feasibility and correlation with histopathological findings. Chin Med J (Engl) 2011;124:1790-5.
19. Joshi U, van der Hoeven JJ, Comans EF, Herder GJ, Teule GJ, Hoekstra OS. In search of an unknown primary tumour presenting with extracervical metastases: the diagnostic performance of FDG-PET. Br J Radiol 2004;77:1000-6.
20. Rusthoven KE, Koshy M, Paulino AC. The role of fluorodeoxyglucose positron emission tomography in cervical lymph node metastases from an unknown primary tumor. Cancer 2004;101:2641-9.
21. Rudmik L, Lau HY, Matthews TW, et al. Clinical utility of PET/CT in the evaluation of head and neck squamous cell carcinoma with an unknown primary: a prospective clinical trial. Head Neck 2011;33:935-40.
22. Randall DA, Johnstone PA, Foss RD, Martin PJ. Tonsillectomy in diagnosis of the unknown primary tumor of the head and neck. Otolaryngol Head Neck Surg 2000;122:52-5.
23. Moller AK, Loft A, Berthelsen AK, et al. 18F-FDG PET/CT as a diagnostic tool in patients with extracervical carcinoma of unknown primary site: a literature review. Oncologist 2011;16:445-51.
24. Greco FA, Vaughn WK, Hainsworth JD. Advanced poorly differentiated carcinoma of unknown primary site: recognition of a treatable syndrome. Ann Intern Med 1986;104:547-53.
25. Hainsworth JD, Greco FA. Treatment of patients with cancer of an unknown primary site. N Engl J Med 1993;329:257-63.
26. Oien KA, Dennis JL. Diagnostic work-up of carcinoma of unknown primary: from immunohistochemistry to molecular profiling. Ann Oncol 2012;23:Suppl 10:x271-x277.
27. Varadhachary GR, Spector Y, Abbruzzese JL, et al. Prospective gene signature study using microRNA to identify the tissue of origin in patients with carcinoma of unknown primary. Clin Cancer Res 2011;17:4063-70.
28. Varadhachary GR, Karanth S, Qiao W, et al. Carcinoma of unknown primary with gastrointestinal profile: immunohistochemistry and survival data for this favorable subset. Int J Clin Oncol 2013 June 28 (Epub ahead of print).
29. Pentheroudakis G, Golfinopoulos V, Pavlidis N. Switching benchmarks in cancer of unknown primary: from autopsy to microarray. Eur J Cancer 2007;43:2026-36.
30. Bloom G, Yang IV, Boulware D, et al. Multi-platform, multi-site, microarray-based human tumor classification. Am J Pathol 2004;164:9-16.
31. Tothill RW, Kowalczyk A, Rischin D, et al. An expression-based site of origin diagnostic method designed for clinical application to cancer of unknown origin. Cancer Res 2005;65:4031-40. [Erratum, Cancer Res 2005;65:8057.]
32. Kerr SE, Schnabel CA, Sullivan PS, et al. Multisite validation study to determine performance characteristics of a 92-gene molecular cancer classifier. Clin Cancer Res 2012;18:3952-60.
33. Pillai R, Deeter R, Rigl CT, et al. Validation and reproducibility of a microarray-based gene expression test for tumor identification in formalin-fixed, paraffin-embedded specimens. J Mol Diagn 2011;13:48-56.
34. Meiri E, Mueller WC, Rosenwald S, et al. A second-generation microRNA-based assay for diagnosing tumor tissue origin. Oncologist 2012;17:801-12.
35. Ferracin M, Pedriali M, Veronese A, et al. MicroRNA profiling for the identification of cancers with unknown primary tissue-of-origin. J Pathol 2011;225:43-53.
36. Horlings HM, van Laar RK, Kerst JM, et al. Gene expression profiling to identify the histogenetic origin of metastatic adenocarcinomas of unknown primary. J Clin Oncol 2008;26:4435-41.
37. Greco FA, Spigel DR, Yardley DA, Erlander MG, Ma XJ, Hainsworth JD. Molecular profiling in unknown primary cancer: accuracy of tissue of origin prediction. Oncologist 2010;15:500-6.
38. Varadhachary G. New strategies for carcinoma of unknown primary: the role of tissue-of-origin molecular profiling. Clin Cancer Res 2013;19:4027-33.
39. Hainsworth JD, Rubin MS, Spigel DR, et al. Molecular gene expression profiling to predict the tissue of origin and direct site-specific therapy in patients with carcinoma of unknown primary site: a prospective trial of the Sarah Cannon Research Institute. J Clin Oncol 2013;31:217-23.
40. Handorf CR, Kulkarni A, Grenert JP, et al. A multicenter study directly comparing the diagnostic accuracy of gene expression profiling and immunohistochemistry for primary site identification in metastatic tumors. Am J Surg Pathol 2013;37:1067-75.
41. Weiss L, Chu P, Schroeder BE, et al. Blinded comparator study of immunohistochemical analysis versus a 92-gene cancer classifier in the diagnosis of the primary site in metastatic tumors. J Mol Diagn 2013;15:263-9.
42. Culine S, Lortholary A, Voigt JJ, et al. Cisplatin in combination with either gemcitabine or irinotecan in carcinomas of unknown primary site: results of a randomized phase II study — trial for the French Study Group on Carcinomas of Unknown Primary (GEFCAPI 01). J Clin Oncol 2003;21:3479-82.
43. Briasoulis E, Kalofonos H, Bafaloukos D, et al. Carboplatin plus paclitaxel in unknown primary carcinoma: a phase II Hellenic Cooperative Oncology Group Study. J Clin Oncol 2000;18:3101-7.
44. Greco FA, Erland JB, Morrissey LH, et al. Carcinoma of unknown primary site: phase II trials with docetaxel plus cisplatin or carboplatin. Ann Oncol 2000;11:211-5.
45. Culine S, Kramar A, Saghatchian M, et al. Development and validation of a prognostic model to predict the length of survival in patients with carcinomas of an unknown primary site. J Clin Oncol 2002;20:4679-83.
46. Varadhachary GR, Raber MN, Matamoros A, Abbruzzese JL. Carcinoma of unknown primary with a colon-cancer profile-changing paradigm and emerging definitions. Lancet Oncol 2008;9:596-9.
47. Greco FA, Lennington WJ, Spigel DR, Hainsworth JD. Molecular profiling diagnosis in unknown primary cancer: accuracy and ability to complement standard pathology. J Natl Cancer Inst 2013;105:782-90.
48. Chapman PB, Hauschild A, Robert C, et al. Improved survival with vemurafenib in melanoma with BRAF V600E mutation. N Engl J Med 2011;364:2507-16.
49. Mao M, Tian F, Mariadason JM, et al. Resistance to BRAF inhibition in BRAF-mutant colon cancer can be overcome with PI3K inhibition or demethylating agents. Clin Cancer Res 2013;19:657-67.