CHAPTER 137 CANCER OF UNKNOWN PRIMARY SITE

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Cancer of unknown primary (CUP) site is a clinical syndrome that represents many types of cancers. Patients are considered to have CUP if no primary site is identified after a standard clinical and pathological evaluation. As diagnostic techniques improve, the spectrum of patients with CUP continues to evolve.

Patients with CUP are common. The exact incidence is unknown because many of these patients are "assigned" other diagnoses and therefore are not accurately represented in tumor registries (see section "Carcinoma of Unknown Primary Site as a Distinct Clinical Syndrome"). Nonetheless, in the United States, CUP accounted for approximately 2% of all cancer diagnoses reported by Surveillance, Epidemiology, and End Results (SEER) registries.[1] International registries from seven other countries have reported incidences ranging from 2.3% to 7.8%.[2] The authors believe a more realistic estimate of the incidence of these patients is 5% of all invasive cancers, approximately 80,000 to 90,000 patients per year in the United States.

Within this heterogeneous patient group, there are a wide variety of clinical presentations and histologic tumor types. Most patients have metastatic carcinoma; however, some neoplasms are difficult to categorize using histologic features alone. Improvements in the evaluation of tumors using immunohistochemical (IHC) staining has aided in differential diagnosis, particularly in separating carcinomas from neoplasms of other lineages. The recent development of molecular gene profiling of tumors promises to aid in defining the tissue of origin of various metastatic adenocarcinomas.

Treatment for patients with CUP has improved slowly and has been the focus of only scattered clinical trials. Early autopsy studies, which showed a preponderance of primary sites considered at the time to be untreatable (lung, pancreas, stomach, colon, liver), have added to the negativity surrounding the diagnosis of CUP.[3] Nevertheless, several important patient subsets, identified either by clinical or pathologic features, are now known to benefit from specific first-line therapy. Also during the past 20 years, treatment has improved for many advanced solid tumors. Standard treatment now improves survival for patients with advanced cancers of the colon, lung, ovary, breast, stomach, kidney, gallbladder, and others. Novel agents for the treatment of cancer are being developed at an accelerated rate. Improved diagnosis of patients with CUP is therefore critical, so that site-specific treatments can be applied.

This chapter is divided into three major sections. The first section, greatly expanded in this edition, reviews the pathologic evaluation of patients with CUP. New information regarding the emerging role of molecular tumor profiling is included. In the second section, the clinical evaluation of CUP patients is summarized. Situations in which results from the pathologic evaluation direct the clinical evaluation are addressed. Finally, the treatment of patients with CUP is discussed, with special focus on specific treatable patient subsets.

PATHOLOGIC EVALUATION

Histologic examination of a biopsy tumor specimen remains the gold standard for initial evaluation and provides a practical classification system on which to base subsequent evaluation. In the broad category of CUP, there are five major light microscopic histologic diagnoses: (1) poorly differentiated neoplasm, (2) poorly differentiated carcinoma (with or without features of adenocarcinoma), (3) well-differentiated and moderately well-differentiated adenocarcinoma, (4) squamous cell carcinoma, and (5) neuroendocrine carcinoma. Sarcoma and melanoma are also occasionally diagnosed without an obvious primary tumor site, and management of these patients follows established guidelines.

These histologic diagnoses vary to some extent with respect to clinical characteristics, recommended diagnostic evaluation, treatment, and prognosis. The approximate size of the various groups and subsets of patients are illustrated in Figure 137.1.

Histologic Subtypes

Poorly Differentiated Neoplasms of Unknown Primary Site

If the pathologist cannot differentiate a general category of neoplasm (e.g., carcinoma, lymphoma, melanoma, sarcoma), the tumor is designated a poorly differentiated neoplasm. A more precise diagnosis is essential because many patients in this category have responsive tumors. Approximately 5% of all patients with CUP (4,000 patients annually in the United States) present with this diagnosis by routine hematoxylin and eosin (H&E) light microscopy, but few remain without a defined lineage after specialized pathologic study.[4–7] The most frequent tumor for which effective therapy is available is non-Hodgkin's lymphoma. In reported series, 35% to 65% of poorly differentiated neoplasms were found to be lymphomas after further pathologic study.[4,5] Most of the remaining tumors in this group are carcinomas, including poorly differentiated neuroendocrine tumors. Melanoma and sarcoma together account for less than 15% of all patients.

Immunohistochemical staining, electron microscopy, and genetic analysis are helpful in the differential diagnosis. The most common cause of a nonspecific light microscopic diagnosis is an inadequate or poorly handled biopsy specimen. If

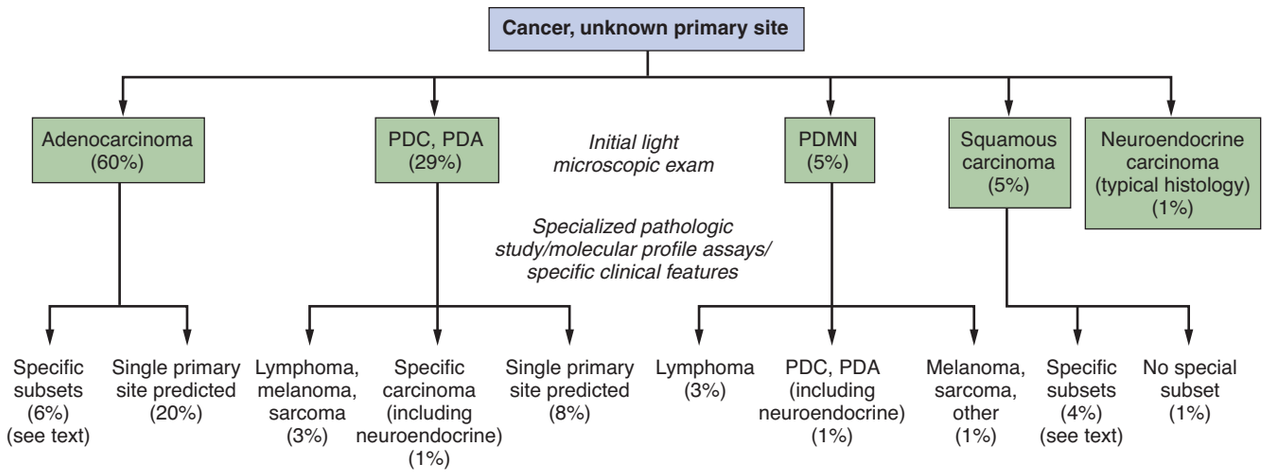

FIGURE 137.1 Relative size of various clinical and histologic subgroups of patients as determined by clinical and pathologic evaluations. PDC, poorly differentiated carcinoma; PDA, poorly differentiated adenocarcinoma; WD, well differentiated; PDMN, poorly differentiated malignant neoplasm.

possible, fine needle aspiration biopsy should not be performed as an *initial* diagnostic procedure because the histologic pattern is not preserved and the ability to perform special studies is limited.

Poorly Differentiated Carcinoma

Poorly differentiated carcinomas (PDC) account for approximately 30% of CUP (about 25,000 patients annually in the United States). In approximately one third of these patients, some features of adenocarcinomatous differentiation can be identified (poorly differentiated adenocarcinoma). Some patients have extremely responsive neoplasms, and therefore careful pathologic evaluation is crucial.

Histopathologic features that can differentiate chemotherapy-responsive tumors from nonresponsive tumors have not been identified.[8] Even with careful retrospective review of these tumors, responsive tumors of well-defined types (e.g., germ cell tumor, lymphoma) are only rarely identified.

All PDCs should undergo additional pathologic study with IHC staining (see section "Immunohistochemical Staining"). In selected tumors, electron microscopy, karyotypic/cytogenetic analysis, and gene expression profiling are also appropriate. Although site-directed or -tailored therapy based on these diagnoses seems reasonable now, there are still only preliminary data supporting an improved outcome with this approach for these patients.

Electron microscopy can be useful for a small minority of these carcinomas and should be reserved for tumors in which IHC is not contributory. Lymphoma can be diagnosed reliably in most instances in those tumors mistakenly believed to be carcinoma. In addition, sarcoma, melanoma, mesothelioma, and neuroendocrine tumors occasionally are defined by subcellular features.

Identification of cytogenetic abnormalities may be useful in patients with PDC. In reference to germ cell tumors, Motzer et al.[9] performed karyotypic analysis on tumors in 40 young men with extragonadal germ cell syndrome or midline carcinomas of uncertain histogenesis. In 12 of the 40 patients abnormalities of chromosome 12 (e.g., i[12p]; del [12p]; multiple copies of 12 p) were diagnostic of germ cell tumor. Other specific abnormalities were diagnostic of melanoma (two patients), lymphoma (one patient), peripheral neuroepithelioma (one patient), and desmoplastic small cell tumor (one patient). Of the germ cell tumors diagnosed on the basis of genetic analysis, five patients achieved a complete response to cisplatin-based chemotherapy. This confirms the authors' previously formulated hypothesis that some of these patients have histologically atypical germ cell tumors. These genetic findings can be diagnostic in these patients.

Autopsy data looking specifically at patients with PDC are limited. Based on the limited necropsy data the authors have accumulated, it appears that primary sites are found in only a minority of these patients (about 40%). These findings are contrary to those for well-differentiated or poorly differentiated adenocarcinoma of unknown primary site, in which the primary site is found in most patients (about 75%) at autopsy.[3,6]

Adenocarcinoma

Well-differentiated and moderately well-differentiated adenocarcinomas are the most common tumors identified by light microscopy and account for 60% of CUP diagnoses (about 50,000 patients annually in the United States). These are the patients that many physicians associate with the entity of CUP. Typically, patients with adenocarcinoma of unknown primary site are elderly and have metastatic tumors at multiple sites. The sites of tumor involvement frequently determine the clinical presentation; common metastatic sites include lymph nodes, liver, lung, and bone.

The diagnosis of well-differentiated or moderately well-differentiated adenocarcinoma is based on light microscopic features, particularly the formation of glandular structures by neoplastic cells. The authors have considered patients with well-differentiated or moderately well-differentiated adenocarcinoma as one group. All adenocarcinomas share histologic features, and the primary tumor site usually cannot be determined by histologic examination. Certain histologic features are typically associated with a particular tumor type (e.g., papillary features with ovarian cancer and signet ring cells with gastric cancer). However, these features are not specific enough to be used as definitive evidence of the primary site.

The identification of relatively cell-specific antigens by IHC staining has improved the ability to predict the site of origin in patients with adenocarcinoma of unknown primary site.[7,10] Panels of IHC stains are most useful and are often directed by clinical features (e.g., sites of metastases, gender). Molecular tumor profiling assays also appear relatively accurate and often provide additional diagnostic information. Both of these new diagnostic modalities should be considered in the pathologic

evaluation of adenocarcinoma of unknown primary site (see sections "Immunohistochemical Staining" and "Gene Expression Profiling and Cancer of Unknown Primary Classification").

Squamous Carcinoma

Squamous carcinoma of unknown primary site represents approximately 5% of patients with CUP (about 4,000 patients annually in the United States). Effective treatment is available for patients with certain clinical syndromes (approximately 90% of patients), and appropriate clinical evaluation is important.

The diagnosis of squamous carcinoma is usually definitively made by examination of histology. Additional pathologic evaluation is usually not necessary. However, IHC staining or molecular studies should be considered in patients with poorly differentiated squamous carcinoma, particularly if the clinical presentation is atypical.

Neuroendocrine Carcinoma

Neuroendocrine carcinomas with widely varying clinical and histologic features are represented in patients with CUP. Neuroendocrine tumors account for approximately 3% of all CUP (about 3,500 patients annually in the United States). Improved pathologic methods for diagnosing neuroendocrine tumors have resulted in the recognition of an increased incidence and wider spectrum of these neoplasms.

Two subgroups of neuroendocrine carcinoma can be routinely recognized by histologic features. Well-differentiated or low-grade neuroendocrine tumors share the same histologic features as carcinoids and islet cell tumors and frequently secrete bioactive substances. A second histologic group (variously described as small-cell carcinoma, atypical carcinoid, or poorly differentiated neuroendocrine carcinoma) has typical neuroendocrine features and an aggressive histology.

A third group of neuroendocrine carcinomas appears histologically as a poorly differentiated neoplasm or poorly differentiated carcinoma. Accurate identification of these tumors requires IHC staining and occasionally electron microscopy or molecular tumor profiling.

Immunohistochemical Tumor Staining

Immunohistochemical staining is the most widely available specialized technique for the classification of neoplasms. Staining usually can be done on formalin-fixed, paraffin-embedded tissue, which broadens its applicability. Immunohistochemical antibodies are usually directed at normal cellular proteins. These proteins are commonly retained during neoplastic transformation. Many new antibodies are being developed against a variety of rather cell-specific proteins, making this area of diagnostic pathology a dynamic and evolving field.

Several important questions can usually be answered by IHC staining. The correct lineage of poorly differentiated neoplasms can be reliably identified in most instances[7,10–12] (Table 137.1). In particular, lymphomas (common leukocyte antigen

TABLE 137.1

IMMUNOHISTOCHEMICAL TUMOR STAINING PATTERNS IN THE DIFFERENTIAL DIAGNOSIS OF CANCER OF UNKNOWN PRIMARY

Tumor Type	Immunohistochemical Staining
Carcinomas	pan-cytokeratin AE1/3 (+), EMA (+), S100 (−), CLA (−), vimentin (−), CK7, 20 (variable)
Lymphomas	CLA (+), pan-cytokeratin AE1/3 (−), EMA (−), S100 (−)
Melanoma	S100 (+), HMB45 (+), melan-A (+), pan-cytokeratin (–), CLA (−)
Sarcoma	vimentin (+), desmin (+), CD117 (+), myogen (+), factor VIII antigen (+), pan-cytokeratin AE1/3 (usually–), S100 (usually –),CLA (−), HMB45 (−), melan-A (−)
Neuroendocrine	Epithelial stains (+), chromogranin (+), synaptophysin (+)
Specific Carcinomas	
Colorectal	CK20 (+), CK7 (−), CDX2 (+)
Lung: adenocarcinoma	CK7(+), CK20 (−), TTF1 (+)
Lung: squamous	CK7 (+), CK20 (−), P63 (+), CK5/6 (+)
Lung: neuroendocrine (small cell/large cell)	TTF1 (+), chromogranin (+), synaptophysin (+)
Breast	CK7 (+), ER (+), PR (+), GCDFP-15 (+), Her2/neu (+), mammogloblin (+)
Ovary	CK7 (+), ER(+), WT1 (+), mesothelin (+)
Bladder (transitional cell)	CK20(+), CK5/6(+), P63(+)
Prostate	PSA (+), CK7(−), CK20(−)
Pancreas	CK7(+), Ca19-9 (+), mesothelin (+)
Renal	RCC (+), CD10(+), pan-cytokeratin AE 1/3(+)
Liver	hepar1(+), CD10(+)
Adrenocortical	alpha-inhibin(+), melan-A(+), CK7(−), CK20(−)
Germ cell	PLAP(+), OCT4(+)
Thyroid/follicular/papillary	thyroglobulin(+), TTF1(+)

EMA, epithelial membrane antigen; S100, calcium binding protein expressed in melanocytes; CLA, common leukocyte antigen; CK, cytokeratin; HMB-45, anti-human melanosome antibody; melan-A, melanoma antigen; CD117, tyrosine kinase receptor (c-kit); CDX2, intestinal specific transcription factor; TTF-1, thyroid transcription factor-1; ER, estrogen receptor; PR, progesterone receptor; GCDFP-15, gross cystic fluid protein 15; WT1, Wilm's tumor transcription factor; p63, tumor suppression gene protein; PSA, prostate specific antigen; RCC, brush border of proximal kidney tubule antibody; CD10, common acute lymphocytic leukemia antigen; hepar1, hepatocyte paraffin 1 marker; PLAP, placental alkaline phosphatase; OCT4, octamen binding transcription factor-4.
(Derived from refs. 7, 10–16.)

staining) and poorly differentiated neuroendocrine carcinomas (chromogranin, synaptophysin staining) can be identified,[7,13] and staining for germ cell tumors (HCG, AFP, OCT4, PLAP) is suggestive in an appropriate clinical situation.

The ability of IHC staining to identify the origin of various adenocarcinomas has improved, but in most cases the staining results must be interpreted in the context of clinical and histologic features. An exception is the prostate-specific antigen (PSA) stain, which is very specific for prostate carcinoma.[7] Stains suggestive of other primary sites are summarized in Table 137.1; the use of panels improves specificity.[7,10–16]

Several problems are associated with the IHC stains. Technical expertise is required to perform these tests accurately and reproducibly, and proper interpretation requires an experienced pathologist. False-positive and false-negative results can occur with any of these stains. For example, some carcinomas stain with vimentin, some sarcomas stain with cytokeratins, and a wide variety of carcinomas do not always stain in the expected patterns.[7,17] The classic staining patterns as illustrated in Table 137.1 often overlap with staining patterns of other adenocarcinomas, forcing the pathologist to consider two or three possible primary sites. However, consideration of the clinical setting helps to direct the selection of the IHC stains and may narrow the spectrum of possibilities if staining patterns are not completely specific. For example, in a patient with mucin-positive adenocarcinoma and metastases limited to the liver, a CK20+/CK7− staining pattern provides strong evidence for the colon as a primary site. Conversely, IHC findings may lead to additional diagnostic procedures; in the above example, a colonoscopy should be performed and may result in the identification of a primary site.

In many cases, a single primary site cannot be identified with certainty even after histologic examination, IHC staining, and correlation with clinical features. Additional pathologic evaluation with either electron microscopy or a search for specific chromosomal abnormalities is useful in a few situations. In addition, molecular tumor profiling is a new technique that promises to be of broad importance in identifying the tissue of origin in patients with CUP.

Electron Microscopy

A diagnosis can be made by electron microscopy in some poorly differentiated neoplasms. Electron microscopy should be reserved for the study of neoplasms whose lineage is unclear after routine light microscopy and IHC staining. Electron microscopy is also reliable in undifferentiated sarcoma. Ultrastructural features such as neurosecretory granules (neuroendocrine tumors) or premelanosomes (melanoma) can suggest a particular tumor. Undifferentiated tumors can lose these specific ultrastructural features; therefore, the absence of a particular ultrastructural finding cannot be used to rule out a specific diagnosis. Electron microscopy is not able to distinguish among various adenocarcinomas and should not be used to identify a tissue of origin in patients with adenocarcinoma of unknown primary site.

Karyotypic or Cytogenetic Analysis

The existence of specific chromosomal abnormalities is well characterized in several hematopoietic neoplasms. Most B-cell non-Hodgkin's lymphomas are associated with tumor-specific immunoglobulin gene rearrangements, and typical chromosomal changes have been identified in some B-cell and T-cell lymphomas and in Hodgkin's lymphoma.[18,19] In the rare instance when the diagnosis of lymphoma cannot be definitively established by IHC staining, electron microscopy, molecular profiling, or chromosomal analysis [t(14:18); t(8:14); t(11:14) and others], the presence of an immunoglobulin gene rearrangement is diagnostic.

A few other nonrandom chromosomal rearrangements associated with nonlymphoid tumors have been identified and occasionally can be useful in the diagnosis of CUP. A chromosomal translocation, t(11:22), has been found in peripheral neuroepitheliomas, desmoplastic small round cell tumors, and frequently in Ewing's tumor.[20–22] A balanced translocation, t(15:19), resulting in the *BRDA-NUT* oncogene has been identified in children and young adults with carcinoma of midline structures of uncertain histogenesis.[23] An isochromosome of the short arm of chromosome 12 (i12p) and other chromosome 12 abnormalities are found in a large percentage of germ cell tumors.[24–26] A genomic hybridization technique has been developed that can detect extra 12p material in paraffin-embedded tissue specimens.[26]

Other nonrandom cytogenetic abnormalities include t(2:13) in alveolar rhabdomyosarcoma; 3p deletion in small-cell lung cancer; 1p deletion in neuroblastoma; t(X:18) in synovial sarcoma; and 11p deletion in Wilm's tumor. Epstein-Barr viral genomes have been identified in the tumor cells of patients with cervical lymph node metastases of unknown primary site, highly suggesting nasopharyngeal primaries.[27,28] The search for specific chromosomal abnormalities should be limited to patients with the histologic diagnoses of poorly differentiated neoplasm or poorly differentiated carcinoma, in whom IHC stains have failed to narrow the diagnostic spectrum. No specific chromosomal changes have been identified to aid in the evaluation of adenocarcinomas.

Gene Expression Profiling and Cancer of Unknown Primary Classification

Gene expression or molecular profiling of human neoplasms arose from DNA microarray analysis described about 15 years ago.[29,30] CUP patients represent a large group with a clinically undefined primary tumor site of origin and are ideal candidates for classification by molecular profiling.[31] Molecular profiling may identify the specific type of cancer present and, when used in concert with the clinical and pathological features, may be useful in predicting the primary tumor site of origin. Primary site identification in CUP will likely improve the therapeutic outcome by allowing site-specific therapy to be administered, rather than an empiric single regimen to all patients. In addition to defining the precise tumor type, molecular tumor profiling may aid in unraveling various gene-specific, cancer-activated, or overexpressed cellular pathways and in identifying new targets for therapy.[32,33]

A pivotal study in cancer classification and diagnosis was reported by Golub et al.[34] and demonstrated for the first time that patterns of gene expression alone could discriminate acute myeloid leukemia from acute lymphoblastic leukemia. Other investigators demonstrated that numerous cancer types could be classified accurately by measuring the differential expression of specific gene sets.[35–44] One basis of molecular profiling in recognizing specific cancer types is the identification of the genes responsible for the synthesis of proteins required for specific normal cellular functions (e.g., milk production in breast luminal duct cells, albumin production in hepatocytes, etc.) in the approximately 400 different normal cell types in humans. Cancer cells retain some normal cell-type specific functional characteristics in their gene expression profile, and usually their origin can be predicted, regardless of neoplastic differentiation.[42] Molecular profiling assays designed to determine the type of cancer are not measuring tumor-specific markers but, rather, gene expression dynamics in relation to cell lineage.

Retrospective Studies in Cancer of Unknown Primary Site

Molecular profiling assays have been validated in patients with metastatic tumors of known primary site. When applied to biopsy specimens from a metastatic site, various molecular assays have correctly predicted the primary site in 76% to 89 % of patients.[38–43] Correct identification of the primary tumor type in CUP is difficult to validate, since the primary tumor site is unknown and rarely becomes apparent during the subsequent clinical course of these patients. It would seem reasonable to assume a similar accuracy rate for these assays in predicting the primary tumor site by testing a metastasis in CUP, and this assumption is supported by the results of several retrospective studies in CUP patients (Table 137.2).[38,39,45–50] However, this validation has usually been indirect and is based on correlation with clinical features, pathology (including IHC stains), and response to treatment.

A complementary DNA microarray was utilized by Tothill et al.[38] on the biopsy specimens from 13 patients with CUP. The primary tumor site predictions were compared with clinicopathologic features. In 11 of 13 patients (85%) the molecular classification prediction was consistent with the most likely primary site as determined by the clinical and pathologic data.

Talantov et al.[39] included 33 patients with CUP in their tumor samples from 449 patients in a validation study of their reverse transcriptase polymerase chain reaction (RT-PCR) assay for known primary cancers. Twenty-two of 33 patients (77%) with CUP were assigned a primary tumor site, and 17 of those (85%) correlated with the prediction of the primary site made by IHC.

Varadhachary et al.[45] used a RT-PCR assay (same as Talantov et al.[39]) on biopsies of 120 patients with CUP. This assay was capable of recognizing only six primary cancer types. In 63 patients (61%), a primary site was predicted, and the clinicopathologic features and response to treatment were compatible with the predicted primary site in most patients. Twenty-three patients with colorectal profiles are of interest. Twelve patients who were retrospectively identified received empiric chemotherapy, usually with paclitaxel and carboplatin, and only two had an objective response. In contrast, 10 of the 11 prospectively identified patients received colorectal cancer regimens in either the first- or second-line setting, and 9 had objective responses to therapy.

A microarray assay was used by Bridgewater et al.[46] on biopsies from 21 CUP patients and results were correlated with clinical and pathologic features. The predicted primary site was "clinically feasible" in 18 (86%) based on clinicopathologic features. The authors felt the management of 12 patients would have been influenced had the assay results been available at the time of the initial diagnosis.

Horlings et al.[47] reported results of a microarray assay on biopsies from 38 patients. Sixteen of these patients had been given a diagnosis based on IHC results; molecular profiling results correlated with the IHC prediction in 15 of 16 patients. Twenty-two of the biopsies could not be classified by IHC staining results. However, molecular profiling predicted a primary site in 14 of these 22 patients (64%).

Monzon et al.[49] used a microarray assay on fresh-frozen biopsy specimens from 21 patients. The primary site of origin was predicted in 16 of 21 patients (76%) and was indeterminate in 5 (24%). In 10 of the 16 patients the assay predictions were consistent with the clinicopathologic suggestions of the primary site.

These small retrospective studies provide some indirect validation of the accuracy of the molecular assays. More direct evidence is now available from a study of CUP patients who had a primary site identified later during their clinical course (latent primary).[50] The authors identified 20 such patients who had primary sites identified 2 to 54 months (median 10 months) after the initial diagnosis of CUP. Four additional patients were later identified (unpublished data). The initial diagnostic biopsies were evaluated by an RT-PCR assay (Cancer Type ID, BioTheranostics, Inc.) capable of identifying

TABLE 137.2

MOLECULAR PROFILE ASSAY VALIDATION STUDIES IN CANCER OF UNKNOWN PRIMARY

Study (Ref.)	Assay (Ref.)	Assay Validated Indirectly by Clinicopathological Correlation[a]	Assay Validated by Latent Primary Site[b]
Tothill et al. (38)	Microarray (38)	13 patients 11 correlated (85%)	Not done
Talantov et al. (39)	RT-PCR (39)	22 patients 17 correlated (85%)	Not done
Bridgewater et al. (46)	Microarray(43)	21 patients 18 correlated (86%)	Not done
Monzon et al. (49)	Microarray (41)	21 patients 16 patients correlated (76%)	Not done
Varadhachary et al. (45)	RT-PCR (39)	120 patients 63 correlated (61%)	Not done
Horlings et al. (47)	Microarray (43)	38 patients 29 correlated (76%)	Not done
Greco et al. (50) and unpublished data	RT-PCR (42)	Not applicable latent primary site known	24 patients 18 predicted accurately (75%)
Greco et al. (unpublished data)	RT-PCR (42)	147 patients (127 evaluable) 83 correlated (66%) 59 patients with single site suspected (52 evaluable) 40 correlated (77%)	Not done

RT-PCR, reverse transcriptase polymerase chain reaction.
[a]Clinical and pathologic (immunohistochemistry) features only; no primary tumor site documented.
[b]Primary tumor site of origin later definitely identified.

32 tumor types. In 18 of 24 biopsies (75%), the primary tumor was accurately predicted (matched the latent primary site identified), providing direct validation of the accuracy and confirming the usefulness of a molecular profiling assay in classifying the primary tumor site in CUP patients.

Comparison of Immunohistochemical and Molecular Profiling Predictions

The small retrospective studies summarized in Table 137.2 suggest that molecular tumor profiling adds to the information obtainable by standard pathologic evaluation. To examine this question more closely, the authors began a prospective study in March 2008 in which all new CUP patients and selected CUP patients already being followed had a molecular profiling assay (Cancer Type ID). Results were correlated with clinical features, pathologic evaluation (including IHC staining), and response to treatment. Although the study is ongoing, the results in 171 patients provide useful information regarding the role of molecular profiling in diagnosis. Molecular tumor profiling provided putative diagnoses in 144 of 171 patients (84%); 22 patients had insufficient tumor in the biopsy specimen to allow successful assay, while 5 tumors had molecular profiles that were unclassifiable. A total of 21 different primary sites were identified; primary sites accounting for 5% or more of patients included intestine (16%), non–small-cell lung (11%), breast (9%), liver (6%), pancreas (5%), and ovary (5%) cancers.

The large majority of patients also had complete IHC profiling. A specific diagnosis based on IHC staining results was predicted in 59 patients (35%). In this group of patients, summarized in Table 137.3, the molecular profiling diagnosis was obtainable in 52 patients and was identical to the IHC prediction in 40 patients (77%). The high level of correlation in lung-adeno/large cell (74%), intestinal (predominantly colorectal; 93%), and breast cancer (100%) is notable; molecular profiling may be superfluous in these patients when the diagnosis is predicted by IHC.

TABLE 137.3

SINGLE PRIMARY SITE SUSPECTED IN CANCER OF UNKNOWN PRIMARY BASED ON IHC STAINING FEATURES: CORRELATION WITH MOLECULAR PROFILE DIAGNOSIS (N = 59)[a]

		Molecular Assay Diagnosis: Agreement with Suspected Primary Site	
Suspected Primary Site	Number	Number	%
Lung-adeno/large cell	19	14	74
Lung-neuroendocrine	3	2	66
Intestine	16	15	93
Breast	5	5	100
Melanoma	3	2	66
Germ cell	2	1	50
Liver	1	1	100
Ovary	1	0	0
Prostate	1	0	0
Sarcoma	1	0	0
Insufficient cells/RNA (Inevaluable)		7	
Total Evaluable	52	40	77

[a]Seven of 59 with insufficient cells/RNA to perform the molecular assay (excluded from analysis).

However, the correlation is lower in other tumor types and further decreases when IHC results are less specific. Ninety-seven of these patients had sufficient tissue for molecular profiling assays; results matched one of the diagnoses suggested by IHC in 43 (44%). In 47 patients with 2 or 3 suggested diagnoses by IHC, the molecular profiling prediction matched one of the two suspected diagnoses in only 20 (43%).

Summary

An increasing body of data now indicates that molecular tumor profiling can accurately predict the tissue of origin in a majority of patients with CUP. The correlation between IHC and molecular profiling is good when IHC predicts a specific primary site; in these patients, molecular profiling may not be necessary. However, in the majority of patients with adenocarcinoma, IHC is less specific and molecular profiling can provide valuable additional information. However, there are few published data regarding the impact of these diagnoses on patient treatment results. Until such data exists, these patients should still be considered to have CUP when planning management. In some cases, consideration of treatment based on the predicted primary site is now appropriate (see the "Treatment" section).

CLINICAL FEATURES AND EVALUATION

Most patients with CUP develop signs or symptoms at the site of a metastatic lesion and are diagnosed with advanced cancer. The subsequent clinical course is usually dominated by symptoms related to metastases; the primary site becomes obvious in only 5% to 10% of patients during their lifetime. At autopsy, a primary site is identified in about 75% of patients.[3,6] Primary sites in the pancreas, lung, colorectum, and liver account for approximately 60% of those identified. Primary sites in the breast, ovary, and prostate are uncommon in autopsy series, but preliminary data from molecular profiling series suggest that breast and ovarian primaries may be more common than previously recognized.

Although some clinical differences exist, there is substantial overlap between the clinical features of patients with adenocarcinoma, poorly differentiated adenocarcinoma, and poorly differentiated carcinoma. Patients with poorly differentiated carcinoma have been a somewhat younger median age and usually exhibit rapid tumor growth. These patients may also have more frequent location of dominant metastatic sites in the mediastinum, retroperitoneum, and peripheral lymph nodes. Because of the similarities, the clinical evaluation of patients with these histologies should follow the same guidelines. Patients with neuroendocrine carcinoma and squamous carcinoma of unknown primary site are discussed separately.

Clinical Evaluation

The recommended clinical evaluation for all patients is summarized in Table 137.4. In actuality, many of these procedures are usually done in the process of diagnosing CUP. Positron emission tomography (PET) scanning should be considered routine in the initial CUP evaluation, although definitive data in large numbers of patients have not been published.[51] Further evaluation for subsets of patients should be directed by results of the initial clinical and pathologic evaluations. Further focused evaluation may (1) identify a primary site, (2)

TABLE 137.4

INITIAL DIAGNOSTIC EVALUATION

- Complete history: including detailed review of systems
- Complete physical examination: including pelvic examination, stool for occult blood
- Complete blood cell count, comprehensive metabolic panel, lactate dehydrogenase, urinalysis
- Computed tomography scans of chest, abdomen, and pelvis
- Mammography in women
- Serum prostate-specific antigen in men
- Positron emission tomography scan in selected patients
- Pathology-including screening immunohistochemistry marker stains (CK7, CK20, TTF-1, CDX2)

narrow the spectrum of possible primary sites, or (3) identify specific treatable subsets of patients (see section "Treatable Subsets").

Table 137.5 summarizes the additional evaluation indicated for several common clinical presentations. Additional evaluation should be triggered by either clinical findings or IHC results during the initial evaluation. Although molecular tumor profiling is not yet considered a standard component of the diagnostic workup and is not included in Table 137.5, the authors believe this test will become standard in the future and can be considered in selected patients.

Neuroendocrine Carcinoma

Although the initial clinical evaluation is the same (Table 137.4), patients with neuroendocrine carcinoma require special consideration in determining appropriate treatment. Of major importance is the separation of this group into tumors with low-grade histology and indolent clinical course versus those likely to have an aggressive clinical course. This distinction can usually be made by the pathologist: patients with classical carcinoid tumors typically have indolent histology, while those with small-cell neuroendocrine carcinoma or poorly differentiated carcinoma with positive neuroendocrine IHC stains have aggressive cancers.

Low grade neuroendocrine carcinomas, when presenting with an unknown primary site, most frequently involve the liver. Other metastatic sites include lymph nodes (usually abdominal or mediastinal) and bone. Some are associated with various syndromes caused by secretion of bioactive peptides (carcinoid syndrome, glucagonoma syndrome, VIPomas, Zollinger-Ellison syndrome). Additional clinical evaluation in these patients should include serum or urine screening for these substances. In addition to the evaluation listed in Table 137.4, upper and lower gastrointestinal endoscopy should be performed, since some of these patients have detectable primary sites in the gastrointestinal tract.

Aggressive neuroendocrine carcinomas of unknown primary site are usually found in multiple metastatic sites and rarely secrete bioactive peptides. Patients with a history of cigarette smoking should be suspected of having a lung primary, particularly if the tumor has a small-cell histology, and a fiberoptic bronchoscopy should be performed. Patients with a positive tumor cell IHC stain for thyroid transcription factor-1 (TTF-1) should also have a bronchoscopy. Extra pulmonary small-cell carcinomas arising from a variety of other sites (salivary glands, paranasal sinuses, esophagus, pancreas, colorectum, bladder, prostate, uterus, cervix) have been described and are occasionally identified during clinical evaluation. Colonoscopy should be considered in patients with tumor IHC staining for CDX2.

The origin of these aggressive neuroendocrine carcinomas remains unclear. It is likely that some patients, with small-cell histology, have small-cell lung cancer with an occult primary tumor. However, more than half of these patients have no smoking history, and the absence of overt pulmonary involvement makes this diagnosis unlikely. It is probable that some of these tumors are undifferentiated variants of well-recognized neuroendocrine tumors (e.g., carcinoid tumor) without a recognizable primary site. In the undifferentiated form, the clinical and pathologic characteristics no longer resemble the characteristics of the more differentiated counterpart.

TABLE 137.5

FOCUSED DIAGNOSTIC EVALUATION OF PATIENT SUBSETS DEFINED BY INITIAL CLINICOPATHOLOGIC EVALUATION

Initial Evaluation	Additional Evaluation
Women with features of breast cancer (bone, lung, liver metastases, CK7+)	Breast magnetic resonance imaging ER, GCDFP-15, HER2 stains
Women with features of ovarian cancer (pelvic/peritoneal metastases; CK7+)	Pelvic/intravaginal ultrasound WT-1 stain
Mediastinal/retroperitoneal mass	Testicular ultrasound Serum HCG, AFP PLAP, OCT4 stains; FISH for i(12p)
Features of lung cancer (hilar/mediastinal adenopathy; TTF-1+)	Bronchoscopy
Features of colon cancer (liver/peritoneal metastases; CK20+/CK7−, CDX2+)	Colonoscopy
Poorly differentiated carcinoma, with or without clear cell features	Stains for chromogranin, synaptophysin, RCC, Hepar-1, HMB-45 (If Hepar-1+, obtain serum AFP; if neuroendocrine stains +, obtain octreotide scan)

HCG, human chorionic gonadotropin; AFP, α-fetoprotein; FISH, fluorescence *in situ* hybridization.

Anaplastic or atypical carcinoid tumors arising in the gastrointestinal tract are responsive to platinum-based chemotherapy, whereas carcinoid tumors with typical histology are usually resistant.[52] A few reports of patients with extrapulmonary small-cell carcinoma of unknown primary site have also documented chemotherapy responsiveness and occasional long-term survival after systemic therapy.[53,54] However, the term extrapulmonary small-cell carcinoma implies the existence of a known primary site; the tumors discussed here are more aptly described as neuroendocrine carcinoma of unknown primary site.

Squamous Carcinoma

As opposed to unknown primary cancers of other histologies, squamous carcinoma almost always presents with isolated metastases in the cervical or inguinal lymph nodes. The cervical lymph nodes are the most common metastatic site. Patients are usually middle aged or elderly, and frequently they have abused tobacco or alcohol, although recently these lesions have also been associated with human papilloma virus infection. When the upper or middle cervical lymph nodes are involved, a primary tumor in the head and neck region should be suspected. Clinical evaluation should include an examination of the oropharynx, hypopharynx, nasopharynx, larynx, and upper esophagus by direct endoscopy, with biopsy of any suspicious areas. Computed tomography (CT) of the neck better defines the disease in the neck and occasionally identifies a primary site. PET scanning is indicated, as it can identify primary tumor sites in a large number of these patients.[55] Detection of Epstein-Barr virus genome in the tumor tissue is highly suggestive of a nasopharyngeal primary site,[27,28] particularly in poorly differentiated carcinomas. Other genetic studies of squamous cell carcinoma of the head and neck region have shown genetic alterations in "normal tissue" as a precursor of invasive carcinoma.[56] Further study is indicated, as these findings do not yet have a practical application. When the lower cervical or supraclavicular lymph nodes are involved, a primary lung cancer should be suspected. Fiberoptic bronchoscopy should be performed if the chest radiograph and head and neck examinations are normal, as this has a high yield, frequently identifying a lung primary.[57]

Ipsilateral tonsillectomy has been advocated as a diagnostic modality in patients with a single node involving the subdigastric, midjugulocarotid, or submandibular areas, and bilateral tonsillectomy has been advocated in patients presenting with bilateral subdigastric adenopathy.[58] In one series of 87 patients who had tonsillectomy as part of their workup for cervical node presentations, 26% had a tonsillar primary identified.[59] The advantages of identifying the primary are worthwhile in this group of patients and include more a specific treatment plan, determination of prognosis, reduction of radiation therapy ports, and perhaps easier follow-up.

Most patients with squamous carcinoma involving inguinal lymph nodes have a detectable primary site in the genital or anorectal areas. Careful examination of the anal canal, vulva, vagina, uterine cervix, penis, and scrotum is important, with biopsy of any suspicious areas. Digital examination and anoscopy should be performed to exclude lesions in the anorectal area. Identification of a primary site in these patients is important because curative therapy is available for carcinomas of the vulva, vagina, cervix, and anus, even after spread to regional lymph nodes.

Metastatic squamous carcinoma in areas other than the cervical or inguinal lymph nodes usually represents metastasis from an occult primary lung cancer. Fiberoptic bronchoscopy should be considered.

TREATMENT

The heterogeneous group of patients with CUP contains some patients who experience long-term survival after appropriate treatment and others for whom treatment makes little or no impact. Patients who have a primary site defined clinically during their initial evaluation should no longer be considered to have CUP and should be treated appropriately for their defined tumor type. A second group of patients can be identified as having specific treatable clinical syndromes, even if the primary site is not identified. The management of these subsets is detailed in this section. Finally, a large group of patients retain the diagnosis of CUP and do not fit into any subset, even after appropriate clinical and pathologic evaluation. Empiric chemotherapy remains the standard treatment for these patients, and this is summarized separately. Site-specific therapy directed by the molecular profiling diagnosis in these patients is a developing area, and it is also briefly reviewed.

Favorable Subsets

Women with Peritoneal Carcinomatosis

Adenocarcinoma, particularly serous adenocarcinoma, causing diffuse peritoneal involvement is typical of ovarian carcinoma, although carcinomas from the gastrointestinal tract, lung, or breast can occasionally produce this clinical syndrome (Table 137.6). On occasion, women with diffuse peritoneal carcinomatosis have no primary site found in the ovaries or elsewhere in the abdomen at the time of laparotomy. These patients frequently have histologic features typical of ovarian carcinoma, such as papillary serous configuration or psammoma bodies, and also share clinical features, such as elevated serum cancer antigen 125 (CA 125) levels. It is now clear that many of these patients have a primary peritoneal carcinoma. These tumors are more common in women with a family history of ovarian cancer, and prophylactic oophorectomy does not always protect them from this tumor.[60] Like ovarian carcinoma, the incidence of primary peritoneal carcinoma is increased in women with *BRCA1* mutations.[61]

The site of origin of some of these carcinomas is from the peritoneal surface (primary peritoneal carcinoma) or from the fimbriated end of the fallopian tubes.[62,63] Because ovarian epithelium is in part an extension of the mesothelial surface, some carcinomas arising from the peritoneal (mesothelial) surface or the uterine tubes share a similar lineage (müllerian derivation) and biology with ovarian carcinoma. Support for this hypothesis has been strengthened by the demonstration of gene expression profiles nearly identical to ovarian carcinoma.[50] Treatment of these women using guidelines for advanced ovarian cancer (surgical cytoreduction followed by taxane or platinum chemotherapy) produces results similar to those seen in comparable stages of ovarian cancer[64,65] and should be the standard approach. Therefore, optimal management of these patients should follow guidelines for the management of advanced ovarian cancer.

Papillary serous peritoneal carcinomatosis has also been reported in men[66]; however, it is difficult to confirm the precise biology, and some of these tumors may be metastatic from an occult primary from elsewhere. The study of gene expression patterns in these patients may be very revealing, particularly if they match those seen in women. A trial of chemotherapy should be administered to good performance status patients.

TABLE 137.6

CARCINOMA OF UNKNOWN PRIMARY SITE: SUMMARY OF EVALUATION AND THERAPY OF RESPONSIVE SUBSETS

Carcinoma	Clinical Evaluation[a]	Special Studies	Subsets	Therapy	Prognosis
Adenocarcinoma (well-differentiated or moderately differentiated)[b]	Chest, abdominal CT scan; PET scan Men: Serum PSA Women: Mammogram Additional studies to evaluate symptoms, signs	Men: PSA stain Women: ER, PR, Other IHC (see text) Molecular Profiling assay (see text)	1. Women, axillary node involvement[b] 2. Women, peritoneal carcinomatosis[b] 3. Men, blastic bone metastases, high serum PSA, or PSA tumor staining 4. Single metastatic site[b] 5. Colon cancer profile	1. Treat as primary breast cancer 2. Surgical cytoreduction plus chemotherapy 3. Hormonal therapy for prostate cancer 4. Lymph node dissection, radiotherapy 5. Treat as metastatic colon cancer	Survival improved with specific therapy
Squamous carcinoma	Cervical node presentation[b] Panendoscopy PET scan Supraclavicular presentation[b] Bronchoscopy PET scan Inguinal presentation[b] Pelvic, rectal examinations, anoscopy PET scan	Genetic Analysis	1. Cervical adenopathy; nasopharyngeal cancer identified by PCR for Epstein-Barr viral genes 2. Supraclavicular 3. Inguinal adenopathy	1. Radiation therapy, neck dissection, chemotherapy 2. Radiation therapy, chemotherapy 3. Inguinal node dissection, radiation therapy, chemotherapy	Survival improved 1. 25%–50% 5-y survival 2. 5%–15% 5-y survival 3. 15%–20% 5-y survival
Poorly differentiated carcinoma, poorly differentiated adenocarcinoma	Chest, abdominal CT scans, serum HCG, AFP; PET scan; additional studies to evaluate symptoms, signs	IHC; electron microscopy; genetic analysis; molecular profiling assay (see text)	1. Atypical germ cell tumors (identified by chromosome 12 abnormalities) 2. Extragonadal germ cell syndrome (two features) 3. Lymph node-predominant tumors (mediastinum, retroperitoneum, peripheral nodes) 4. Gastrointestinal stromal tumors (identified by CD117 stain) 5. Other groups (see text)	1. Treatment for germ cell tumor 2. Cisplatin/etoposide 3. Newer chemotherapy 4. Imatinib 5. Newer empiric chemotherapy/or site-specific therapy	1. 40%–50% cure rate 2. Survival improved (10%–20% cured) 3. Survival improved 4. Survival improved 5. Survival improved
Neuroendocrine carcinoma	Chest, abdominal CT	IHC Electron microscopy Genetic analysis including molecular assay (see text)	1. Low-grade 2. Small-cell carcinoma (or Ewing's family of tumors) 3. Poorly differentiated	1. Treat as advanced carcinoid 2, 3. Carboplatin/etoposide or platinum/etoposide (or other)	1. Indolent biology/long survival 2, 3. High response rate survival improved; rarely cured

CT, computed tomography; PET, positron emission tomography; IHC, immunohistochemistry; PSA, prostate-specific antigen; ER, estrogen receptor; PR, progesterone receptor; HCG, human chorionic gonadotropin; AFP, α-fetoprotein.
[a]In addition to history, physical examination, routine laboratory tests, and chest x-ray films.
[b]May also present with poorly differentiated carcinoma, and management and outcome are similar.

Women with Axillary Lymph Node Metastases

Breast cancer should be suspected in women who have metastatic carcinoma in an axillary lymph node.[67] Men with occult breast cancer can present in this fashion, but these are rare. The initial lymph node biopsy should be stained for IHC breast markers including estrogen receptors, progesterone receptors, and *HER2*. Elevated levels provide strong evidence for the diagnosis of breast cancer.[68]

If no other metastases are identified, these patients may have stage II breast cancer with an occult primary, which is potentially curable with appropriate therapy. PET and magnetic resonance imaging have identified occult breast cancer even with normal mammography.[69–71] Modified radical mastectomy has been recommended in such patients, even when physical examination and mammography are normal. An invasive occult breast primary has been identified after mastectomy in 44% to 80% of patients. Primary tumors are usually less than 2 cm in diameter and may measure only a few millimeters; in occasional patients, only noninvasive tumor is identified in the breast. Prognosis after primary therapy is similar to that of other patients with stage II breast cancer.[67] Radiation therapy to the breast after axillary lymph node dissection represents a reasonable alternative primary therapy.[72] Either neoadjuvant or adjuvant systemic chemotherapy is indicated in this setting, following guidelines established for the treatment of stage II breast cancer.

Women with metastatic sites in addition to the axillary lymph nodes should be managed as if they have metastatic breast cancer. Hormone receptor and *HER2* status are of particular importance in these patients because they may derive major palliative benefit from hormonal therapy, chemotherapy, and trastuzumab. In the experience of the authors, a molecular profiling assay usually predicts breast carcinoma in these patients.

Men with Elevated Serum Prostate-Specific Antigen or Prostate-Specific Antigen Tumor Staining

Serum PSA concentrations should be measured in men with adenocarcinoma of unknown primary site. These tumors can also be stained for PSA. Even when clinical features (i.e., metastatic pattern) do not suggest prostate cancer, a positive PSA (serum or tumor stain) is reason for a trial of androgen deprivation.[73,74] In most of these patients, a needle biopsy of the prostate would confirm the primary site but may not be necessary for optimal clinical management. Osteoblastic bone metastases in the absence of other metastatic sites are also an indication for an empiric hormone trial, regardless of the PSA findings.

Extragonadal Germ Cell Cancer Syndrome

The extragonadal germ cell cancer syndrome was first described in 1979.[75–77] The full syndrome, which is seen in only a minority of patients, has the following features: (1) occurrence in men less than 50 years of age, (2) predominant tumor location in the midline (mediastinum, retroperitoneum) or multiple pulmonary nodules, (3) short duration of symptoms (less than 3 months) and a history of rapid tumor growth, (4) elevated serum levels of human chorionic gonadotropin (HCG), α-fetoprotein (AFP), or both, and (5) good response to previously administered radiation therapy or chemotherapy. If possible, cytogenetic evaluation for chromosome 12 abnormalities should be obtained, as previously discussed. Because these patients may have atypical germ cell tumors, treatment with cisplatin-based chemotherapy, as used in advanced poor-prognosis testicular cancer, is recommended.

Single Site of Neoplasm

When only one site of neoplasm is identified (e.g., one node group, one mass), the possibility of an unusual primary tumor mimicking metastatic disease should be considered. Several unusual tumors could present in this fashion, including Merkel-cell neuroendocrine tumors; skin adnexal tumors (e.g., apocrine, eccrine, and sebaceous carcinomas); and even sarcomas, melanomas, or lymphomas that are mistakenly interpreted as metastatic carcinoma (pathologically and clinically). Patients with one site of involvement (brain, liver, adrenal, subcutaneous tissue, bone, intestine, lymph node, skin, or other sites) usually have metastatic carcinoma, and many other sites are present but are not detectable. Some of these patients may have a primary tumor at the single site that developed from embryonic rest cells or adult stem cells (see the section "Special Issues in Carcinoma of Unknown Primary Cancer-Biology of the Primary Tumor"). Before initiating local treatment, a PET scan is helpful to exclude other unsuspected metastatic sites.[78]

In the absence of any other documented metastatic disease, these patients should be treated with aggressive local therapy (i.e., resection, radiation therapy, or both) because a minority enjoy long-term, disease-free survival. In addition to definitive local therapy, the authors believe these patients should also receive either neoadjuvant or adjuvant chemotherapy with one of the newer regimens, but it is difficult to be certain if this treatment is superior to local therapy alone.

Patients with a single small site of metastasis frequently survive 1 year or longer and thus represent a favorable prognostic subgroup. In a reported group of patients presenting with single brain metastasis of unknown primary site, 15% remained progression free 5 years after definitive therapy.[79] The authors have treated and followed 36 patients with single site metastases (unpublished observations). All patients had local therapy (resection with or without radiotherapy) and most also received empiric chemotherapy regimens. The median survival in this group is 17 months; 1-, 2-, and 3-year survivals are 65%, 40%, and 28% respectively.

Squamous Carcinoma Involving Cervical or Supraclavicular Lymph Nodes

Squamous carcinoma of unknown primary site is unusual, but most frequently presents with unilateral involvement of the cervical lymph nodes. The clinical evaluation of these patients has been previously described. The recommended evaluation results in the identification of a head and neck primary site in almost 85% of patients.

When no primary site is identified, local treatment should be given to the involved neck. The reported results in more than 1,400 patients are derived primarily from retrospective single-institution experiences, often using a variety of local treatment modalities.[80] In many of these series, a large minority of patients had poorly differentiated carcinoma or adenocarcinoma. A substantial percentage, usually 30% to 40%, of patients achieved long-term, disease-free survival after local treatment modalities. The results obtained using radical neck dissection, high-dose radiation therapy, or a combination of these modalities have been similar. The volume of tumor in the involved neck influences outcome, with N1 or N2 disease having a significantly higher cure rate than N3 or massive neck involvement.[81] Poorly differentiated carcinoma also represents a poor prognostic factor in these patients. When resection alone is used as the primary treatment modality, a primary tumor in the head and neck subsequently becomes apparent in 20% to 40% of patients. Primary tumors surface less commonly when radiation therapy is used, presumably because of the eradication of occult head and neck primary sites within the radiation field. Radiation therapy dosages and techniques should be similar to those used in patients with primary head and neck cancer, and the nasopharynx, oropharynx, and hypopharynx may be included in the irradiated field.

The role of chemotherapy for metastatic squamous carcinoma in cervical lymph nodes is now generally accepted. A nonrandomized comparison of patients treated with local modalities alone or with local modalities combined with chemotherapy (cisplatin and 5-fluorouracil [5-FU]) showed a higher complete response rate (81% vs. 60%) and longer median survival time (more than 37 vs. 24 months) in patients receiving chemotherapy.[82] Combined modality treatment with concurrent chemotherapy and radiotherapy in locally advanced head and neck carcinoma is now standard and should be the treatment of choice for squamous cell carcinoma in cervical lymph nodes. In those who receive local therapy first, adjuvant platinum-based or taxane-based chemotherapy should be considered.

Patients with low cervical and supraclavicular nodes do not do as well because lung cancer is a frequent site of occult primary tumors, although skin, uterine, cervix, and anal canal are also possible primary sites. Molecular assays may be helpful in predicting the primary site. Patients with no detectable disease below the clavicle should be treated with aggressive local therapy because 10% to 15% of these patients have long-term, disease-free survival. Concurrent chemotherapy should also be considered for these patients.

Squamous Carcinoma Involving Inguinal Lymph Nodes

Most patients with squamous carcinoma involving inguinal lymph nodes have a detectable primary site in the genital or anorectal areas. For the unusual patient in whom no primary site is identified, inguinal lymph node dissection with or without radiation therapy to the inguinal area sometimes results in long-term survival.[83] These patients should also be considered for neoadjuvant or adjuvant chemotherapy.

Low-Grade Neuroendocrine Carcinoma

These tumors usually exhibit an indolent biology, and slow progression over years is likely. Management should follow guidelines established for metastatic carcinoid or islet cell tumors from known primary sites. Treatment with octreotide long-acting release (LAR) results in a marked increase in time to tumor progression and is a first-line treatment of low toxicity.[84] Depending on the clinical situation, appropriate management may also include local therapy (resection of isolated metastasis, hepatic artery ligation or embolization, cryotherapy, radiofrequency ablation). Several cytotoxic agents have some activity (streptozocin, doxorubicin, 5-fluorouracil, temozolomide), and preliminary results with targeted agents (sunitinib, everolimus) are promising. These neoplasms are usually refractory to intensive systemic chemotherapy, and cisplatin-based chemotherapy produces low response rates.[76]

Aggressive Neuroendocrine Carcinomas

Patients with aggressive neuroendocrine carcinoma of unknown primary site are those with either small-cell carcinoma or poorly differentiated carcinoma (often large cell) with neuroendocrine staining by IHC. Both of these histologies are initially responsive to combination chemotherapy, and all patients should be considered for a trial of treatment.

The authors initially reported a group of 29 patients with poorly differentiated neuroendocrine tumors[85] and have updated their experience to include 99 patients, 94 treated with combination chemotherapy. Most of these patients had clinical evidence of rapid tumor growth and metastases in multiple sites. Fifty-nine of 87 assessable patients (68%) responded to chemotherapy with a platinum-based combination regimen. Nineteen patients (22%) had complete responses, and 13 remained continuously disease free more than 2 years after completion of therapy.

The results of a prospective trial using the combination of paclitaxel, carboplatin, and oral etoposide in 48 patients (48 of the 99 previously listed) have been reported.[86] The majority of these patients were initially called poorly differentiated carcinoma (about 20% were small-cell carcinoma) but later defined as neuroendocrine tumors by IHC staining or electron microscopy. Most of these patients had several sites of metastasis, often with predominant tumor in the bones, liver, and nodes (particularly retroperitoneum and mediastinum). Patients received a maximum of four courses of chemotherapy with paclitaxel, carboplatin, and oral etoposide; stable or responding patients subsequently received weekly paclitaxel for 24 weeks. The overall response rate was 55% with six complete responses (13%). The median survival was 14 months and 12 patients remain alive from 15 to 45 months.

Data from clinical trials remain limited in this uncommon group of patients; however, current first-line chemotherapy should include the platinum-based regimens used for small-cell lung cancer. The addition of paclitaxel to a carboplatin and etoposide regimen increased toxicity, but did not appear to improve efficacy.[86] In the uncommon patient with a single site of involvement, radiation therapy with or without resection should be added to combination chemotherapy.

Poorly Differentiated Carcinoma

Although patients with poorly differentiated carcinoma form a relatively large and heterogeneous group, the inclusion of patients with highly treatable neoplasms within this group has been recognized since the late 1970s.[75–77] At that time, several young men with mediastinal tumors were reported who had complete response to combination chemotherapy. Elevated serum levels of HCG or AFP were common in these young men. Although the histology was not diagnostic, these patients were thought to have histologically atypical extragonadal germ cell tumors. Several other tumor lineages have subsequently been identified in some of these patients (i.e., thymic neoplasms, neuroendocrine tumors, midline carcinoma with t(15;19), sarcomas, melanomas, lymphomas), but others still defy precise classification.

Further evidence for the responsiveness of many other patients has accumulated since 1978. Based on the encouraging results in a few patients treated from 1976 to 1978, the authors prospectively studied the role of cisplatin-based therapy. In a series of reports, the authors documented a high overall response rate and long-term disease-free survival in a minority of these patients.[87–90] The 220 patients seen and treated, between 1978 and 1989, are of interest.[90] Most of the patients did not have clinical characteristics strongly suggestive of extragonadal germ cell tumor. However, involvement of the mediastinum, retroperitoneum, and peripheral lymph node groups was relatively common; these clinical features are now known to be associated with a more favorable prognosis. All patients who received initial treatment with two courses of cisplatin-based chemotherapy and responding patients received a total of four treatment courses. Major tumor responses were seen in 138 of 220 patients (62%), and 58 patients (26%) had complete response to treatment.

Of the 58 complete responders, 22 patients remained alive and relapse free (38%), representing 10% of the entire group of 220. These results in this large series of patients are historically important, since long-term survival in these patients had not been previously reported. At that time, the results also supported the notion that poorly differentiated histologic types represent more sensitive tumors than well-differentiated adenocarcinoma. Other investigators also demonstrated the responsiveness of selected poorly differentiated carcinomas.[91–96]

Complete responses were seen in a minority (10% to 20%) of these patients, and a small cohort (5% to 10%) comprised long-term, disease-free survivors. These results were usually seen with platinum-based chemotherapy.

The authors now are certain that their original prospective clinical trial of the 220 patients with PDC was heavily weighted with patients now known to represent favorable subsets, each with a relatively good prognosis. These subsets included (1) patients with two or more features of the extragonadal germ cell syndrome, (2) patients with poorly differentiated neoplasms otherwise not specified, (3) patients with anaplastic lymphoma diagnosed as carcinoma in years past but routinely diagnosed today by specialized pathology, (4) patients with primary peritoneal carcinoma, (5) patients with poorly differentiated neuroendocrine carcinoma, and (6) patients with predominant sites of tumor involving the retroperitoneum, mediastinum, and peripheral lymph nodes. The authors' more recent experience has excluded these now recognizable more favorable subsets of patients in their clinical trials. After these favorable subsets of patients are excluded, the remaining patients have a similar prognosis to the large majority of the well-differentiated adenocarcinoma group, and since 1996 the authors have included all these patients in new clinical trials.

Colorectal Cancer Profile

With the introduction of more effective cytotoxic agents and targeted therapies, the median survival of patients with metastatic colon cancer has increased from 9 to about 24 months.[97,98] It is therefore likely that the ability to identify the subset of CUP patients likely to have advanced colorectal cancer would lead to better treatment for these patients. The improved specificity of IHC staining for colon cancer, coupled with the recent availability of molecular tumor profiling assays, may now allow identification of this patient subset. Although results to date are derived from relatively few patients, the potential importance of this syndrome merits inclusion here.

Patients with typical clinical features (liver, peritoneal metastases), histology compatible with a lower gastrointestinal primary, and typical IHC staining (CK20+/CK7− and/or CDX2+) have been defined as having the "colon cancer profile." Several such patients described by Varadhachary et al.[99] had excellent responses to colorectal cancer regimens.

Preliminary data also indicate that a molecular profiling assay that confirms a colorectal origin may identify patients who respond to colon cancer therapy. The authors and colleagues performed a molecular assay (Veridex RT-PCR assay) on biopsies from 104 patients with CUP; in 23 patients, colon was predicted as the site of origin.[45] In 17 of 23 patients, colonscopy was negative (6 not done). Nine of 10 patients who received colorectal cancer regimens had objective responses. In contrast, 2 of 12 patients (retrospectively identified) had responded to empiric chemotherapy for CUP (usually with taxane- or carboplatin-based regimens).

The authors now have data on 21 additional CUP patients in whom molecular profiling assay (Cancer Type ID) predicted a colorectal origin. Table 137.7 shows the clinicopathologic features, treatment, and outcome of these 21 patients; similar data are also included regarding the 11 patients who received colorectal cancer treatment as reported in a previous publication.[45] Thirty of 32 patients had normal colonoscopy. Twenty-three of the 30 evaluable patients received standard first-line regimens for colorectal cancer, and 16 (69%) had objective responses. In addition, 7 of 13 patients (54%) responded to colon cancer regimens as second-line treatment. The median survival for the entire group was 20 months (range: 4–65+ months); the 2- and 4-year survivals were 59% and 30%, respectively.

Although these data are derived from a relatively small group of patients from two institutions with some of the patients identified retrospectively, the treatment results are similar to those achieved in patients with known metastatic colon cancer. Further prospective studies are essential to confirm these results. In the meantime, the authors feel these results are sufficient to recommend treatment with colorectal cancer regimens for CUP patients with a colorectal cancer profile defined by either IHC staining or molecular profiling assay.

Empiric Therapy for Metastatic Carcinoma of Unknown Primary Site

Chemotherapy

Approximately 80% of patients with carcinoma of unknown primary site are not represented in any of the favorable prognostic clinical subsets (Table 137.6). In the past, empiric chemotherapy of various types has produced low response rates, very few complete responses, and even fewer long-term survivals.[2,80] The results of chemotherapy in several reported series of 10 or more patients from 1964 to 2002 are briefly summarized as follows. A total of 1,515 patients were reported in 45 trials.[2,80] The only single agent studied adequately in previously untreated patients was 5-FU, with response rates ranging from 0% to 16%. Cisplatin was evaluated as a single drug in only one series, with a response rate of 19%. Methotrexate, doxorubicin, mitomycin C, vincristine, and semustine had single agent response rates ranging from 6% to 16%. The FAM regimen (5-FU, doxorubicin, mitomycin C) and various modifications were used often, based on the demonstrated activity of these combination regimens in some gastrointestinal cancers. The combination of 5-FU and leucovorin has not been evaluated adequately but does not appear active in CUP patients with liver metastasis, a group most likely to have gastrointestinal primaries.[100] The overall response rates from all these prospective clinical trials varied from 8% to 39% (mean: 20%); the complete response rate was less than 1%. The median survival ranged from 4 to 15 months (mean: 6 months), and survival beyond 2 years was rare (although rarely reported).

Cisplatin-based combination chemotherapy regimens were also evaluated several years ago. In two small, randomized comparisons (subject to many confounding factors) of doxorubicin with or without cisplatin, no difference in median survival was observed, but there was more toxicity in the cisplatin-containing arms.[101,102] A third small, randomized trial did show the superiority of cisplatin, epirubicin, and mitomycin C compared with mitomycin C alone (median survival: 9.4 vs. 5.4 months).[91]

The authors have reviewed several reports of survival for large groups of patients with CUP[103–110] in an attempt to have some historical control data and better define the natural history of this syndrome. These reports were retrospective; therefore, treatments were not uniform, and some patients received no systemic therapy. In addition, these series usually contained patients now known to fit into specific treatable or favorable subsets. These historical series represent 31,419 reported patients. The median survival was 5 months, with a 1-year survival of 22% and 5-year survival of 5%. It is very likely that survival at 1 year and beyond is largely represented by subsets of patients with a more favorable prognosis who received local therapy (surgery or radiotherapy) or those with indolent tumors (e.g., carcinoids). Squamous cell carcinoma (usually in neck nodes) and well-differentiated neuroendocrine carcinoma (carcinoid, islet cell–type histology) reported from some of these series (N = 2,971 patients) had median, 1-year, and 5-year survivals of 20 months, 66%, and 30%, respectively. All

TABLE 137.7

CANCER OF UNKNOWN PRIMARY WITH INTESTINAL/ MOLECULAR PROFILE DIAGNOSES: CLINICOPATHOLOGIC CHARACTERISTICS AND RESULTS/SURVIVAL OF SITE-SPECIFIC THERAPY IN FIRST- AND SECOND-LINE SETTING

Age	Sex	Histology	Sites of Metastases	IHC + Marker	First-line Treatment	Response	Second-Line Treatment	Survival Response	(Months)
75	M	PDA	Liver, lung, bone	CK7	Folfox	PR	Folfiri/Ce	PR	19
56	F	Adeno	Peritoneum, ovary	CK20, CDX2	Folfox/B	PR			9+
46	F	PDA	Ovary	CK20, CDX2	Folfox/B	CR			19+
61	F	Adeno	Lung, nodes	CK20, villin	PC	PD	Cape/RT	PR	8
42	M	PDA	Peritoneum, testes	CK20, CDX2	Folfox	PR	Iri/Ce	PR	31+
54	F	PDA	Peritoneum, ovary, bone	CK20, CDX2, CK7	FU	NA	D/B	SD	30
53	M	Adeno	Liver, peritoneum	CK7, CK20, CEA	PC	PR			7+
52	F	PDA	Peritoneum, abdominal nodes	CK20, chromogranin, CK7	NA	NA			6
70	F	Adeno	Liver	CK7, CK20, CEA	Gem/Cis	SD	Folfox	PR	10+
55	M	PDA	Liver, bone	CK20, CDX2, CEA	Folfox/B	PR	Iri/Ce	NA	12+
58	F	Adeno	Liver, peritoneum, ovary	CDX2	PC	PD	Folfox	PR	38+
61	M	Adeno	Mesentery, omentum	CK7, CK20	Folfox	PR			9
47	F	Adeno	Neck node, lung, retroperitoneum	CK 20	Folfox/B	PR			6
53	F	Adeno	Retroperitoneum, liver	CK7, CK20, CDX2	Capox/B	PR			4
78	M	PDC	Pelvic mass	CK20, CDX2	Folfox/B	PR			16
53	M	PDA	Liver, lung, brain	CK7, villin	Capox	PR			5
48	F	PDA	Liver, retroperitoneum, mediastinum	CK7, Her2-neu	PC/T	PR	Capox	PD	6
63	F	Adeno	Pelvic mass	CK20, CDX2	Folfox/B	PR	Cape/DC/C/RT		65+
63	M	Adeno	Peritoneal, ascites	CK7, CK20, CDX2	Capox	PR			10+
54	F	Adeno	CA Omentum, peritoneal	CK20, CDX2	CE	PR	Folfox/B	PR	60+
68	F	PDC	Peritoneum, retroperitoneum	CK20	PC	PR			4+
46	M	PDC	Mediastinum	CK20, CDX2	Folfox/Iri/B	PR			32+
81	F	Adeno	Liver, lung	CK20, CDX2	Folfox/B	PR			5+
49	F	Adeno	Liver, peritoneum	CDX2	PCE	PR	Gem/Iri	PD	10
49	M	Adeno	Peritoneal mass	CK7, CK20, CDX2	Folfox	PD	Folfiri/B	PD	6+
68	F	Adeno	Liver, peritoneum, mediastinum	CDX2	Cape/B	PR			7
69	M	Adeno	Pelvic mass, bone	CK20, CDX2	Cape/RT	CR			29+
64	F	Adeno	Ovary, omentum	CK20, CDX2	Folfox/B	CR	Folfiri	PD	13
51	M	Adeno	Retroperitoneum	CK20	Capox	NA			13
49	M	PDA	Liver	CK20, CDX2	Gem/Iri	SD			10
47	F	PDA	Liver, lung, mesentery	CK20, CDX2	Cape/Iri/B	PR			22
62	M	Adeno	Retroperitoneal mass	CK7, CK20	FU/P/C	PD			4

IHC, immunohistochemistry; PDA, poorly differentiated adenocarcinoma; PDC poorly differentiated carcinoma; Adeno, adenocarcinoma; Gem, gemcitabine; Folfox, fluorouracil/leucovorin/oxaliplatin; Folfiri, fluorouracil/leucovorin/irinotecan; Iri, irinotecan; FU, fluorouracil; D, docetaxel; B, bevacizumab; Cis, cisplatin; Cape, capecitabine; Capox, capecitabine/oxaliplatin; P, paclitaxel; C, carboplatin, E, etoposide; T, trastuzumab; RT, radiotherapy; Ce, cetuximab; M. male, F, female; PR, partial response; CR, complete response; SD, stable disease; NA, not accessible; PD, progressive disease.

the remaining patients in these series had median, 1-year, and 5-year survivals of 6 months, 20%, and 5%, respectively.

In the past decade, empiric chemotherapy has improved for patients with adenocarcinoma and poorly differentiated carcinoma who do not fit into any of the treatable subsets.[111,112] The introduction of several new drugs with rather broad-spectrum antineoplastic activity from 1990 to 2000 and later with targeted mechanism-based therapies changed the approach to treatment and prognosis for patients with several common epithelial cancers. These drugs include the taxanes, gemcitabine, vinorelbine, irinotecan, topotecan, oxaliplatin, and several targeted agents (e.g., bevacizumab and erlotinib).

Since 1996, the Minnie Pearl Cancer Research Network/Sarah Cannon Oncology Research Consortium(MPCRN/SCORC) has completed nine sequential prospective phase 2 trials incorporating paclitaxel,[113,114] docetaxel,[114,115] gemcitabine,[116,117] gemcitabine/irinotecan,[118,119] bevacizumab/erlotinib,[120] and oxaliplatin[121] into the first-line or second-line

therapy for 692 patients. One additional phase 3 randomized prospective trial has been reported with 198 patients.[122] Only patients with CUP who were not included in a favorable subset were eligible for these trials. The first five phase 2 studies (396 previously untreated patients) have a minimum follow-up of 4 years. The total objective response rate for all patients treated in the first five clinical trials was 30% (107 of 353 evaluable patients), with 85 (24%) partial responders and 22 (6%) complete responders. With a minimum follow-up of 4 years and maximum follow-up of 11 years, the median survival was 9.1 months, and the 1-, 2-, 3-, 5-, 8-, and 10-year survivals were 38%, 19%, 12%, 10%, 8%, and 8%, respectively. The median progression-free survival was 5 months, and the 1-, 2-, 3-, 5-, 8-, and 10-year progression-free survivals were 17%, 7%, 5%, 4%, 3%, and 3%, respectively. The toxicity of these regimens was generally moderate, primarily myelosuppression, with a total of eight (2%) treatment-related deaths.

Long-term follow-up on the 264 patients in the first four trials is of interest since follow-up of this duration has not been previously reported from prospective trials. After a minimum follow-up of 6.5 years (range: 6.5 to 11 years); the median survival was 10.2 months; and the 1-, 2-, 3-, 5-, 8-, and 10-year survivals were 41%, 24%, 15%, 11%, 8%, and 8%, respectively. The actuarial survival curves for the 428 other patients treated in four additional phase 2 trials and the single phase 3 trial[122] look similar. There have been no significant survival differences when results from the phase 2 studies were compared. The phase 3 trial compared paclitaxel with carboplatin and oral etoposide to gemcitabine and irinotecan; stable and responding patients received follow-up treatment with gefitinib. The survival at 2 years was similar (15% vs. 18%), and gemcitabine and irinotecan was significantly less toxic.[122] Although both empiric regimens appear to improve the 2-year survival compared to historical controls, the modest efficacy underscores the need for newer therapeutic approaches for these patients.

Three second-line regimens have been recently evaluated in phase 2 trials, including gemcitabine[117] (39 patients), gemcitabine with irinotecan[118] (40 patients), and oxaliplatin with capecitabine[121] (48 patients). Modest activity was documented in this difficult patient group with clinical benefit seen in about 35%; median survivals were 4.0, 4.2, and 9.7 months, and 2-year survival rates were 20%, 12%, and 25%, respectively.

Analysis of all the previously untreated patients in the MPCRN/SCORC trials shows no difference in survival for adenocarcinoma versus PDC. Women survived significantly longer than men, and those with performance status 0 or 1 (Eastern Cooperative Oncology Group Scale) lived longer than those with performance status 2.

Several trials reported by others in the past decade[92,123–133] have substantiated the activity of the newer combination regimens (Table 137.8). These phase 2 trials usually contained combinations of newer broad spectrum cytotoxic drugs (paclitaxel, docetaxel, gemcitabine, irinotecan, vinorelbine, oxaliplatin). The primary end points of these trials were response rate or median survival. The 1-year survival was reported in 12 studies (532 patients), and survival at both 1 and 2 years was reported by eight of the studies (363 patients). The 1-year survival ranged from 25% to 52% (mean: 34.4%) and at 2 years from 5% to 18% (mean: 12.3%). Only one study reported a 3-year survival rate (11%). These survival results are very similar to the 396 patients reported by the previously detailed six MPCRN/SCORC studies. The survival of all 988 patients (532 from 12 studies plus 456 from 6 MPCRN/SCORC studies) are shown in Table 137.8. These survival data are unique, as survival at 2 years and beyond had not been previously appreciated.

The survival data reported with newer empiric regimens during the past decade appear superior to historical retrospective control survival data and to the combined data from multiple prospective clinical trials reported from 1964 to 2002.[2,80] Although the median survival of this group has not changed dramatically, a larger number of patients derive major benefit from treatment, as indicated by the number of 1- and 2-year survivors. The survival curve has been shifted to the right and the survival at 2 years is comparable to the 1-year survival of historical control patients. Comparison of the existing phase 2 trials does not allow definition of an optimum regimen; several

TABLE 137.8

SURVIVAL IN PATIENTS WITH CANCER OF UNKNOWN PRIMARY AND UNFAVORABLE PROGNOSTIC FACTORS: SELECTED PHASE 2 TRIALS IN THE PAST DECADE

Year of Publication (Ref.)	No. of Patients	Regimen	Median Survival (months)	1-Year Survival (%)	2-Year Survival (%)	3-Year Survival (%)
2000 (92)	33	PC	10	25	5	NR
2001 (123)	34	PFUL	8.3	26	NR	NR
	17	CE	6.4			
2003 (125)	30	GemCisE	7.2	36	14	NR
2004 (127)	37	PCis	11	38	11	NR
2004 (128)	35	GemD	10	43	7	NR
2004 (126)	102	CDoxE	9	35.3	18	11
2005 (130)	22	PC	6.5	27	NR	NR
2006 (131)	66	GemCis	13.6	52	NR	NR
	33	GemVCis	9.6	30	NR	NR
2006 (124)	51	GemC	7.8	26	12	NR
2007 (133)	42	PC	8.5	33	17	NR
2007 (129)	47	OxIri	9.5	40	NR	NR
2007 (132)	33	GemCapeC	7.6	35.6	14.2	NR
1997–2009 (113–119, 134)	456	Multiple regimens	9.1	38	19	12
Total	988		9.0	35[a]	14[a]	12[a]

Gem, gemcitabine; Cape, capecitabine; Ox, oxaliplatin; Iri, irinotecan; V, vinorelbine; Cis, cisplatin; B, bevacizumab; FU, fluorouracil; C, carboplatin; P, paclitaxel; L, leucovorin; Dox, doxorubicin; E, etoposide; Er, erlotinib; D. docetaxel; NR, not reported
[a]Mean survivals of all studies

two-drug combinations appear similar and are currently acceptable. Many patients with adenocarcinoma or poorly differentiated carcinoma who do not fit or conform to any recognized favorable subset can now attain substantial clinical benefit from the new drug combinations, and a trial of treatment should be considered in all patients with acceptable performance status.

However, we believe that the era of empiric chemotherapy for patients with CUP is nearing its end. Improved diagnosis using IHC stains and molecular tumor profiling are likely to provide a more rational framework for decision making regarding therapy. Increasing numbers of targeted agents are now available or in clinical development, and their utility will most likely be defined by the identification of critical molecular abnormalities.

Targeted Therapy

A number of agents targeting pathways critical to cancer cells have been incorporated into the standard therapy of various solid tumors. It is likely that some patients in the heterogeneous group of CUP patients would also benefit from these targeted agents. Although there has been limited clinical experience with targeted agents, definite activity has been documented.

The combination of bevacizumab and erlotinib was evaluated in a group of 51 patients.[120] Thirty-seven patients had received previous chemotherapy (24 patients, 1 regimen; 13 patients, 2 regimens), and 14 patients were previously untreated but with poor prognostic features (advanced liver metastasis, bone metastasis or three or more visceral sites of metastases). All patients received bevacizumab 10 mg/kg intravenously every 2 weeks and erlotinib 150 mg orally daily. Forty-seven of 51 patients received at least 8 weeks of therapy; 5 patients (10%) had a partial response, and 29 patients (61%) had stable disease (many with tumor shrinkage). The median survival was 7.4 months with 33% of patients alive at 1 year and 18% at 2 years. Patients tolerated this therapy well (grade 3 or 4 toxicity of any type less than 10%, except fatigue at 16%). Survival seemed superior to second-line chemotherapy previously reported and was similar to results of many first-line chemotherapy trials.

This trial was followed by a first-line phase 2 study evaluating standard chemotherapy (paclitaxel and carboplatin) plus targeted therapy (bevacizumab and erlotinib).[134] Sixty patients received four cycles of these four agents repeated at 21-day intervals, followed by bevacizumab and erlotinib continued until tumor progression. Forty-nine of 60 patients completed the induction therapy, and 44 (73%) received the maintenance targeted drugs. Thirty-two patients (53%) had objective responses to treatment, and 18 others were stable. At a median follow-up of 19 months, the median progression-free survival was 8 months; 38% of the patients were progression free at 1 year. The median survival was 12.6 months, and the 2-year survival was 27%. There was no unexpected severe toxicity. This empiric regimen was relatively effective, and further empiric approaches using targeted therapy with chemotherapy are reasonable to investigate.

Prognostic Factors

The identification of prognostic factors in the population of patients with CUP continues to evolve as the group is divided into an increasing number of subsets. By definition, patients who fit into the favorable treatment subsets (see section "Treatable Subsets") have favorable prognosis compared to the remaining patients. As new treatable subsets are identified, the clinical features of the remaining patients can be expected to change. Therefore, results of previous analyses of prognostic factors, conducted primarily in patients receiving empiric chemotherapy, may no longer apply to the current population.

At M. D. Anderson Cancer Center, a large heterogeneous group with various histologic subtypes was analyzed retrospectively.[135,136] Patients with clinical features of extragonadal germ cell tumors were excluded, and only a minority of patients with PDC received cisplatin-based treatment. Clinical and pathologic features identified as favorable prognostic features included limited number of metastatic sites, tumor location in lymph nodes (including mediastinum and retroperitoneum) other than the supraclavicular lymph nodes, and female sex. Adverse prognostic factors included adenocarcinoma histology (as compared to other histologies) and liver metastasis.

Van der Gaast et al.[137] evaluated 79 patients with PDC and found three groups with median survivals of 4 years, 10 months, and 4 months based on performance status and serum alkaline phosphatase levels. A minority of their patients were long-term survivors following chemotherapy.

Culine et al.[138] have also defined a prognostic model based on retrospective analysis of 150 patients with various histologies. Patients with several known favorable prognostic subsets were excluded. Patients with good performance status and normal serum lactate dehydrogenase (LDH) levels had significantly better median survival (11.7 vs. 3.9 months) and 1-year survival (45% vs. 11%) after cisplatin-based chemotherapy. The LDH level was more predictive of prognosis than was the presence of liver metastasis.

Seve et al.[139] investigated a population of 317 patients in a Canadian center seen from 1998 to 2004 and found low serum albumin and lymphopenia to be important prognostic factors. A group of good-risk patients (normal serum albumin and no liver metastasis) had a median survival of about 1 year compared to 3.5 months ($P < .0001$) for poor-risk patients (low serum albumin with or without liver metastasis). These findings were validated in a group of 81 patients seen at two French centers from 2000 to 2004. Only 116 of the 317 patients in the initial test series were treated with chemotherapy, raising the question of the usefulness in patients in a setting appropriate for chemotherapy. Nonetheless, a number of easily obtainable clinical parameters appear to offer important prognostic information.

The authors examined prognostic factors in a large group of patients with PDC who were treated with cisplatin-based chemotherapy.[90] In this group, favorable features included mediastinal or retroperitoneal tumor location, metastases at less than three sites, age less than 35 years, female gender, negative smoking history, and normal LDH and carcinoembryonic antigen (CEA) levels.

In summary, prognostic factors that have been repeatedly identified are related to tumor location, extent of tumor, performance status, and measures of general health status (serum albumin, lymphocyte count). None of these features is surprising, since most have been repeatedly identified as prognostic factors in patients with various solid tumors. These factors should be considered when designing, interpreting, or comparing results of clinical trials in patients with CUP.

Site-Specific Treatment Directed by Results of Molecular Tumor Profiling

The emergence of molecular tumor profiling, as well as IHC staining of improved specificity, raises the question as to whether treatment guided by these results is superior to empiric therapy for CUP. At present, there are insufficient clinical data available to answer this question. However, the authors believe that the fragmentary information now available suggests a future change in the paradigm of treatment.

Since the biology of CUP is different from that of other cancers (as evidenced by the fact that the primary site does not become apparent), there has been speculation that these cancers will also respond differently to treatment. If so, the ability to identify the tissue of origin may not lead to improved therapy. However, most clinical data suggest that CUP represents a collection of cancer types, which, if identified, will respond to site-specific therapy in a predictable way. The successful treatment

of patients in several of the treatable subsets supports this argument. For example, women with adenocarcinoma that involves the peritoneum respond to ovarian cancer treatment, patients with squamous carcinoma that present in neck nodes have successful outcomes following treatment for head and neck cancer, and so forth. Furthermore, preliminary data now suggest that patients identified as having a colorectal primary site by molecular tumor profiling have good responses to site-specific therapy for colon cancer (see section "Colorectal Cancer Profile").

Prospective evaluation of site-specific therapy selected on the basis of molecular profiling or IHC results is urgently needed. Demonstration of the superiority of site-specific versus empiric therapy will be most likely in tumor types where treatment efficacy has improved and differs from empiric CUP therapy (e.g., colorectal, renal, hepatic, biliary tract). In other tumor types where site-specific and empiric CUP therapy are similar (e.g., ovary, non–small-cell lung) or in situations where all therapy is relatively ineffective (e.g., pancreas), such differences remain currently difficult to demonstrate. However, an acceptance of the site-specific treatment approach, based on prospective validation in selected tumor types, would allow generalization to additional tumor types as improved tumor-specific treatments are developed.

SPECIAL ISSUES IN CARCINOMA OF UNKNOWN PRIMARY SITE

Biology of the Primary Tumor

The biology of the primary tumor in CUP remains an enigma. The majority of patients harbor a clinically occult primary tumor site, as demonstrated by autopsy series.[3,6] It is remarkable that many of these invasive primary tumors measure less than 1 cm and some only a few millimeters. Rarely a latent primary tumor site is found many weeks or months after the initial diagnosis of CUP. The mechanism explaining very small clinically occult invasive primary tumor sites remains unknown but almost certainly will be clarified by a better understanding of the molecular mechanisms controlling primary tumor growth and metastasis. There are several other potential explanations for the apparent absence of a primary cancer in some of these patients. First, some of these primary cancers may inexplicably regress or involute entirely, despite the fact that metastasis already occurred. This theory is supported by the scarring seen occasionally in the testicle of male patients with metastatic germ cell neoplasms (i.e., "burned-out primary"). Second, some of these tumors may have arisen from embryonic epithelial "rest cells" that are fully differentiated but did not complete their appropriate migration *in utero* to their designated tissue or organ. Extragonadal germ cell tumors with primaries in the mediastinum, retroperitoneum, or undescended testicular cancer are known examples of this phenomenon. Third, some of these patients have unrecognized primary neoplasms such as an extragonadal germ cell tumors, thymic neoplasms, lymphomas, melanomas, or sarcomas, which arise from these lineages virtually anywhere in the body. Fourth, the pathogenesis of some of these carcinomas may result from a specific genetic lesion present in all cells, and these tumors might be expected to have a similar gene expression distinct from specific carcinomas of recognized primary sites, as is suggested by the unusual occurrence of metastatic adenocarcinoma of unknown primary site in monozygotic twin brothers with primary immunodeficiency disorder (X-linked hyperimmunoglobulin M syndrome).[140]

Finally, some of these neoplasms may arise from adult undifferentiated pluripotent stem cells with an ability to differentiate to multiple lineages.[141–145] Hematopoietic stem cells appear to be able to give rise to or transform into liver cells as well as muscle, gastrointestinal, skin, and brain cells.[141] Reserve precursor stem cells exist within the connective tissue compartments throughout postnatal life[144] and can form any lineage in any tissue if they undergo neoplastic transformation. Therefore, some tumors might continue to reflect the differentiation or transformation of adult stem cells and may be "tumors of adult stem cells." For example, seemingly metastatic adenocarcinoma in bone, liver, lymph node, or elsewhere may, in fact, arise in these sites from an adult stem cell with the capacity to become any type of cell and to develop as a "primary" neoplasm in any of these tissues.[142]

Although carcinomas of unknown primary share a metastatic phenotype, it is currently unknown whether these tumors share specific molecular abnormalities. Karyotypic analysis of unknown primary carcinomas demonstrates multiple chromosomal abnormalities, but these are not unique and are shared with advanced solid tumors of known primary sites (e.g., various chromosomal 1p abnormalities).[146] Similarly, overexpression of *p53*, *bcl-2*, *c-myc*, *ras*, and *HER2* has been observed in some CUP, but are not specific.[147–151] Although the search for a CUP-specific molecular profile continues, none has yet been identified. At present, most evidence suggests that CUP retains typical site-specific molecular abnormalities and can be identified by molecular tumor profiling; however, this does not preclude the coexistence of CUP-specific molecular abnormalities.

Carcinoma of Unknown Primary Site as a Distinct Clinical Syndrome

The authors have found it amazing over the past three decades how often patients and their referring physicians (often oncologists) are frustrated by CUP. Physicians are often somewhat obsessed with finding the primary site or at least with giving the patient a more specific diagnosis. There are many reasons underlying these feelings. Some patients think their oncologist may not be a very good diagnostician and seek the advice of others. Some oncologists feel relatively inadequate and wonder what other test(s) they might order; some have been relatively tentative, not feeling confident in recommending any therapy. Certainly a reasonable evaluation of these patients and their tumors is indicated, being aware of possible primary sites and the relevance in particular patients. However, once these considerations and evaluations are complete, the physician should stop, discuss the issue with the patient and family, and accept the clinical syndrome as CUP. Patients are better served, and physicians eventually feel more comfortable and therefore manage these patients more effectively once their patients accept and understand this diagnosis. The authors now believe improved diagnostic techniques, including more specific IHC marker stains and molecular profile assays, will change the nature of these conversations in the future. Nonetheless, these patients will still lack anatomically defined primary sites and will therefore remain a distinct population.

A second practical issue in the United States is the determination of reimbursement for chemotherapy by Medicare for cancer diagnoses. Other than U.S. Food and Drug Administration approval for a specific tumor type, reimbursement for chemotherapy is most typically determined by Medicare (and some other third-party insurers) by consulting compendia—Medicare Drug Policies or the National Comprehensive Cancer Network Compendium. The list of "approved" drugs is based on published literature showing "effectiveness" or clinical benefit in a specific tumor type. This is an arbitrary system. For many years CUP was not included in any of the listings. Four drugs are currently listed as indicated for these patients (paclitaxel, carboplatin, cisplatin, and etoposide).

Medicare usually does not pay for any drug not listed as being indicated. For this reason, many patients with CUP are coded by oncologists as having other diagnoses. These diagnoses usually represent a "good guess" or statistical probability, based on clinicopathologic features. For example, patients with lung lesions or mediastinal node involvement are often coded as having non–small-cell lung cancer; patients with liver metastases are coded as colon or pancreatic cancer. Furthermore, patients are at times assigned a diagnosis based on the pathology report alone (e.g., adenocarcinoma consistent with pancreatic or colon primary) or by cytokeratin-staining results. This activity causes the true incidence of unknown primary cancer to be underestimated but also allows for reimbursement for some drug costs by a system that otherwise has not "approved" therapies for these patients.

There are now enough clinical and pathologic data to classify patients confidently and global acceptance of this syndrome will help these patients establish an identity, stimulate more interest by physician investigators, and eventually improve the general understanding of these patients and their tumors.

Isolated Pleural Effusion

An isolated malignant pleural effusion is most frequently a manifestation of a peripheral lung carcinoma (usually adenocarcinoma). The diagnosis of mesothelioma, or, rarely, a metastatic tumor from other sites, should also be considered. In a series of 42 patients, a primary lung cancer was eventually found in 15 patients (36%).[152] The primary may not be apparent even after chest tube drainage. Cytology usually shows adenocarcinoma; positive TTF-1 and CK7 stains support a diagnosis of lung carcinoma. Other IHC stains (i.e., calretinin in mesothelioma) or a molecular profiling assay may also assist in defining a primary site. In one small series of patients,[152] chemotherapy produced symptomatic improvement in 29 of 37 patients, and 30 of 37 patients had their pleural effusion reduced by chemotherapy; median survival was 12 months (range: 3–60 months).

In evaluating a female patient with an isolated pleural effusion, the possibility of an occult ovarian carcinoma or primary peritoneal carcinoma should be considered. Although this presentation is rare, these tumors are often relatively sensitive to treatment. This diagnosis is possible even when CT and PET scans of the abdomen and pelvis are normal. In such cases, an elevated CA 125 level suggests the diagnosis.

Germ Cell Tumors with Metastases of Other Histologies

On occasion, patients with germ cell tumors, particularly extragonadal primaries, may have a metastatic lesion that consists of only somatic tumor cells. This is particularly true for neuroendocrine or sarcomatous differentiation but can include any histology. Patients therefore may be diagnosed as having a neuroendocrine tumor or sarcoma. In these rare instances, a primary germ cell tumor (usually extragonadal) is present elsewhere and subsequently is clinically apparent. It is difficult to make the diagnosis initially. An elevated plasma AFP or HCG level is suggestive. The presence of a mediastinal, retroperitoneal, or testicular mass supports this possibility. Chromosomal analysis, IHC staining, or a molecular assay may confirm the diagnosis of germ cell tumor. The treatment of choice is cisplatin-based chemotherapy. Surgical resection should be pursued if feasible. These patients have a worse prognosis than those with typical germ cell tumors, probably because the somatic cell tumors are less sensitive to chemotherapy.

Melanoma and Amelanotic Melanoma

Approximately 10% to 15% of all melanomas that present with an unknown primary site are believed to be amelanotic. The authors have viewed this diagnosis with considerable skepticism. At times, the only reason for the pathologic diagnosis is the similarity of the histologic pattern to melanoma, even though no pigment is demonstrated. In the authors' experience, detailed pathologic and molecular study has occasionally revealed a group of other specific diagnoses, including lymphomas, neuroendocrine tumors, germ cell tumors, sarcomas, and poorly differentiated carcinoma (not otherwise specified).

Melanosomes or premelanosomes seen on electron micrographs have been considered diagnostic of melanoma, but on rare occasion these structures are seen in other tumors. Some believe amelanotic melanomas do not always form premelanosomes, raising the question as to whether they are really melanomas. Immunohistochemical panels and a molecular profiling assay are also useful in supporting the diagnosis of melanoma. It is of interest that in the authors' original report of 220 patients with poorly differentiated carcinoma, 9 were later believed to be amelanotic melanoma on the basis of IHC stains or electron microscopy.[90] These particular patients generally responded well to cisplatin-based chemotherapy, and several had long-term survival, an unexpected result for melanoma.

The history of a resected, abraded, or frozen pigmented skin lesion would certainly favor a metastatic melanoma in an individual. In addition, the rare primary visceral melanoma should be considered (e.g., eye, adrenal, bowel, others) as the source of the disease in questionable cases. For patients with the diagnosis of amelanotic melanoma, particularly without diagnostic IHC stains and no history or clinical features to support this diagnosis, empiric treatment based on guidelines for CUP should be considered. Recently *BRAF* mutations have been found in approximately 50% of melanomas, and if present would also support a presumptive diagnosis of melanoma and consideration of a clinical trial with a *BRAF* inhibitor.

UNKNOWN PRIMARY CANCER IN CHILDREN

There are limited data in children, and, as expected, many of these neoplasms represent embryonal malignancies.[153] They are exceedingly rare. In those rare patients with carcinoma, not otherwise specified, the authors favor following the same management plan as for adults.

Midline Carcinoma in Young Adults and Children with t(15;19) and *BRD4-NUT* Oncogene

A few young patients have been recently described with carcinomas arising from midline locations and an associated chromosomal translocation t(15;19) (q13,p13.1).[23] Patients with this syndrome ranged in age from 3 to 35 years, most had PDC, and all had widespread metastasis. The primary tumor site was difficult to identify in many of these patients. The *NUT* (nuclear protein in testes) oncogene is common to all these tumors and supports their possible origin from a specific cell type, perhaps an early epithelial progenitor cell that is more common in the first two or three decades of life. Perhaps these tumors are an example of "stem cell tumors" (see section "Biology of the Primary Tumor in Special Issues in Carcinoma of Unknown Primary Site").

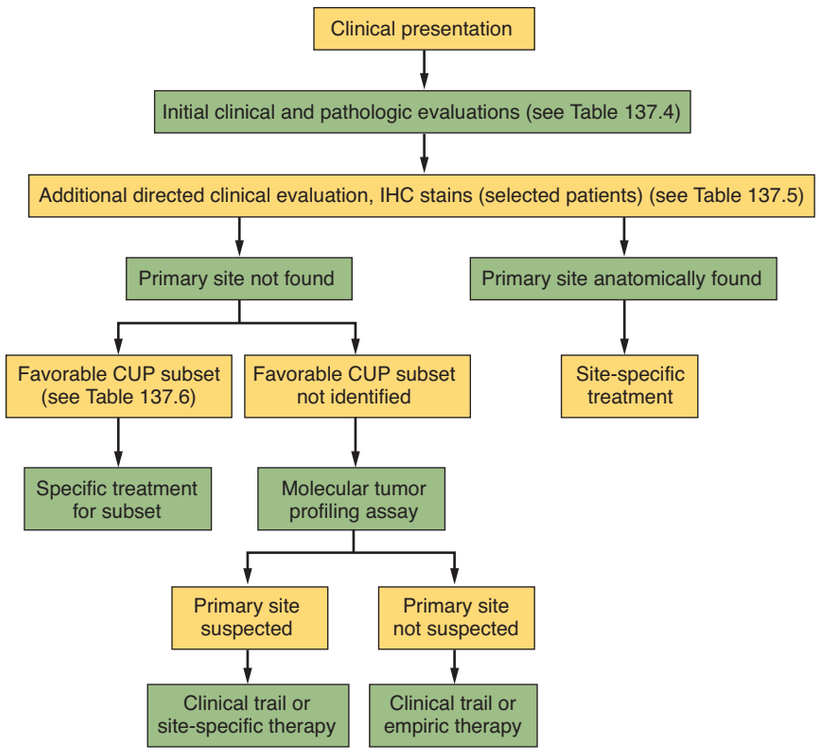

FIGURE 137.2 Suggested new management paradigm for the cancer of unknown primary site patient.

Despite intensive chemotherapies and radiation therapy, which produced initial good responses, all but one of these patients died from disease within 16 months (median: 7 months). Two additional patients were more recently reported: one patient with a tumor arising from the iliac bone (unknown primary site)[154] has been in complete remission for 13 years after combined modality therapy, and a second patient with mediastinal involvement (unknown primary site)[155] had a good response to secondary therapy with docetaxel and radiotherapy. They are clinically similar to the extragonadal germ cell cancer syndrome, and without a positive t(15;19), some of these patients could be included in that clinical syndrome and vice versa. Further knowledge of these *NUT*-rearranged carcinomas and improved treatment for these patients are likely to follow their more broad recognition.

FUTURE DIRECTIONS AND CHANGING TREATMENT PARADIGM

As described in this chapter, improving diagnostic methods are likely to change the diagnostic and therapeutic approach to patients with CUP in the near future. Although clinical data are currently incomplete, a change from empiric chemotherapy to site-specific therapy based on predictions from new diagnostic methods is predicted by both authors.

In Figure 137.2, we summarize what we believe to be the future management approach to patients with CUP. After standard initial clinical and pathologic evaluations, selected patients will have additional directed clinical evaluation or IHC staining of the tumor specimen. Patients with an identified primary site will be treated accordingly, and patients who fit into an identified favorable CUP subset will have appropriate subset-specific therapy (see section "Treatable Subset"). Patients in neither of these categories (i.e., patients who traditionally were candidates for empiric chemotherapy) will have molecular tumor profiling performed and will then be considered for site-specific therapy based on molecular profiling results interpreted in concert with clinical features and pathologic results.

The authors emphasize that the integration of molecular diagnostics into standard patient management is not yet supported unequivocally by clinical data. Continued clinical trials in this area are vital. Even with the ability to identify the tissue of origin, further improvements in the treatment of CUP are dependent on the development of improved treatments for advanced solid tumors.

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