Histologic Predictors of Renal Cell Carcinoma Response to Interleukin-2-Based Therapy

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Summary: The authors examined pathology from patients with renal cancer (RCC) treated with IL-2 to determine response rates for clear cell and variant RCC and to identify histologic features that predict response. Pathology specimens were reviewed by a single pathologist who was blinded to both the prior pathology interpretation and the therapeutic response. Findings were correlated with response to IL-2 therapy. Evaluable pathology specimens were obtained from 231 patients. Of 163 primary RCCs, the response rate was 21% (30/146) for patients with clear cell versus 6% (1/17) for patients with variant or indeterminate type RCC ($P = 0.20$). For clear cell carcinomas, response to IL-2 was associated with the presence of alveolar features and the absence of papillary and granular features. Patients with more than 50% alveolar features and no granular or papillary features had a 39% response rate (14/36). Patients with alveolar and granular features representing less than 50% of the specimen and no papillary features had a 19% response rate (15/77). The response rate for the others was 3% (1/33). This model was then applied to an independent sample of 68 metastasis specimens. Response rates in the three prognostic groups and for patients with non-clear cell cancers were 25% (5/20), 9% (2/22), 0% (0/16), and 0% (0/10), respectively. Median survivals for all patients with clear cell tumors by risk group were 2.87, 1.36, and 0.87 years, respectively ($P < 0.001$). These data suggest that patients with non-clear cell RCC or with clear cell RCC with papillary, no alveolar, and/or more than 50% granular features respond poorly to IL-2 and should be considered for alternative treatments. Investigation of other tumor-related predictors of IL-2 responsiveness is warranted.

Key Words: renal cancer, IL-2, histologic features

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Metastatic renal cell carcinoma (RCC) remains a vexing therapeutic challenge. High-dose IL-2, the only FDA-approved therapy, produces tumor regression in 15% to 20% of patients,[1,2] with only 7% of patients showing durable long-term freedom from recurrence. High-dose IL-2 treatment is also associated with considerable toxicity and expense, making it an impractical standard. Improved treatment strategies and/or better methods of identifying patients likely to benefit from IL-2-based therapy are needed.

Kidney carcinomas have been shown to comprise several distinct subtypes that differ in genetics as well as in histologic appearance and clinical behavior.[3,4] The majority of conventional (clear cell) carcinomas possess mutations in the Von Hippel-Lindau gene located at chromosome sub-band 3p25, while papillary (chromophil) carcinomas have alterations in copy numbers, typically trisomies of chromosomes 7 and 17, and losses of chromosome Y.[3,5–7] In addition, type I and type II papillary carcinomas differ in their chromosomal abnormalities, the latter more commonly showing losses in chromosome Xp,[8] while many type I and familial papillary carcinomas have mutations in the c-met oncogene (locus 7q31), which encodes for hepatocyte growth factor receptor.[9] Chromophobe and collecting duct carcinomas presumably have other associated genetic abnormalities yet to be fully defined. The relative responsiveness of these various histologic subgroups to IL-2-based therapy has never been fully evaluated.

Studies of RCC populations have identified clinical and histologic factors that predict for poor survival with or without specific therapy.[10–24] For example, Motzer et al reported that in patients receiving IFN, poor survival is associated with low Karnofsky performance status, high serum lactate dehydrogenase, low hemoglobin, high "corrected" serum calcium, and time from initial RCC diagnosis to start of therapy of less than 1 year.[21] Similar clinical prognostic features have been identified in patients receiving various IL-2-based treatments.[22–24] A recent multivariate analysis confined to patients who received IL-2 after nephrectomy reported survival to be inversely associated with lymph node involvement, constitutional symptoms, metastases involving sites other than bone or lung or multiple sites, a TSH level more than 2.0 mIU/L, or sarcomatoid histology.[22] Others have reported high nuclear grade and rhabdoid and sarcomatoid phenotypes to be associated with more aggressive disease.[14,15,25–27] Prior to the distinction of papillary, chromophobe, and collecting duct tumor types from conventional carcinomas, morphologic features such as patterns of cell arrangement, (tubular, papillary, and alveolar), degree of granularity or eosinophilia of the cytoplasm, and extent

of inflammation and/or necrosis were determined to have possible prognostic significance.[11–17] Worse survival was associated with granular rather than clear cytoplasm and fusiform or spindle-shaped cells.

Less information has been reported on predictors of response to specific therapies, including IL-2. Clinical factors that have been variably associated with response to IL-2-based therapy include good performance status, one metastatic site, no bone metastases or prior IFN therapy, prior nephrectomy or erythropoietin production, treatment-related thrombocytopenia, no thyroid dysfunction, or rebound lymphocytosis, and post-treatment elevations of blood TNF-α and IL-1 levels.[28,29] Cangiona et al reported that despite a poor overall prognosis, patients with sarcomatoid RCC were still able to respond,[30] but little additional information is available regarding the relationship of other pathologic and morphologic features to tumor response in patients receiving IL-2-based therapy.

Between 1990 and 2001, the Cytokine Working Group (CWG) performed seven separate clinical trials involving 388 patients with metastatic RCC using a variety of IL-2-based regimens.[31] These trials have included patients with variant RCCs. Tumor responses were seen in 63 patients (19 complete and 44 partial responses) for a response rate of 16%. An additional 56 patients were treated with IL-2 at Beth Israel Deaconess Medical Center (BIDMC) outside of these CWG trials, with tumor responses observed in 12% of patients. We performed a detailed histologic review and classification of pathology slides obtained from these patients to determine the response to IL-2 for patients with clear cell or variant RCC and within in the clear cell subtype to identify morphologic features predictive of response to IL-2-based therapy.

METHODS

Pathology slides were requested from all 388 patients treated between 1990 and 2001 on CWG clinical trials. All patients had previously provided informed consent to participate in the CWG clinical trial, which included the possibility of central review of their pathology material. Pathology specimens were also obtained from patients treated on non-CWG clinical trials involving IL-2 at BIDMC between 1997 and 2001. The current investigation was approved by the Institutional Review Board of BIDMC.

The CWG trials for which pathology specimens were collected are described elsewhere.[31] Eligibility criteria for these trials included histologically confirmed stage IV RCC, measurable and clearly progressive disease, Eastern Cooperative Oncology Group performance status of 0 or 1, excellent organ function, and no prior IL-2-based therapy. Treatment included inpatient regimens involving high-dose IL-2 alone or in combination with IFNα or low-dose IL-2 alone or outpatient regimens involving IL-2 in combination with IFNα with or without 5-flurouracil.[31,32]

Two hundred sixty-five specimens were obtained from 239 patients. Specimens were obtained from 183 patients on CWG trials and all patients on BIDMC trials. Eleven specimens from eight patients were eliminated from the analysis either because of insufficient material (n = 4) or because they were fine-needle aspirates (n = 7). In addition, 21 patients had two specimens and 1 patient had three specimens. In these instances, kidney and then lung metastasis specimens were preferentially analyzed. Other metastatic sites were analyzed only when neither kidney nor lung specimens were available. Only a single specimen per patient was included in the analysis, so that any rule developed for the kidney specimens could be assessed in an independent group of patients with specimens obtained from the other sites. Thus, we report results based on 231 pathology specimens, from 231 distinct patients.

Cases were classified as IL-2 responders or non-responders for this analysis, based on the response reported in the initial clinical study. Responders included patients with either complete or partial responses (>50% regression of measurable tumor deposits). Patients were considered non-responders if there was only a minor response (25%–50% regression of measurable tumor deposits) or no response to IL-2 treatment. Survival was calculated from the date of initiation of IL-2 treatment until date of either death or last follow-up.

Histologic evaluation was performed using standard H&E-stained sections and specially stained slides available from the original pathologic examination. All cases were evaluated according to a protocol based on the consensus classification of carcinoma types[4] and the UICC staging and Fuhrman nuclear grading systems,[10] with additional data fields designed to capture morphologic features that might permit additional analyses. Semiquantitative assessments of necrosis and tumor-infiltrating lymphocytes (TILs) were performed using an Olympus BH-2 microscope with standard 10× oculars and objectives.

All pathology assessments were performed by the same pathologist (M.P.U.), blinded to the response to therapy and to the original reading of the slides. The type of carcinoma was recorded as clear cell, which may have variable amounts of clear or granular/eosinophilic components; papillary with subtyping of type I (basophilic) and type II (eosinophilic) as defined by Delahunt and Eble[33]; chromophobe; collecting duct; indeterminate; or other. The designation "indeterminate" was reserved for cases having insufficient well-preserved tissue to permit accurate classification, or for neoplasms so poorly differentiated that assignment by type was not possible.

Each case was also assessed for features such as sarcomatoid, rhabdoid, papillary, granular/eosinophilic, tubular, alveolar, and cystic morphologies, and it was noted whether or not these features represented greater than 50% of the carcinoma area. The architectural patterns of "tubular," "alveolar," and "cystic" growth arrangement were taken from the categorization outlined and illustrated by Bostwick and Eble.[34] The tubular pattern is also described as "acinar" by other authors.[35] Standard definitions were used for sarcomatoid,[27] rhabdoid,[25,26] and granular patterns. These patterns are illustrated in Figure 1.

The presence or absence of TILs and necrosis was noted for each neoplasm. TIL presence was graded in a semiquantitative fashion on a scale of 0 to 3, with 0 = no lymphocytes identified; 1 = lymphocytes apparent in only one high-power field (400×) or visible only at high power; 2 = lymphocytes present on scanning power in some but not all areas of the carcinoma and intermediate in concentration; 3 = abundant lymphocytes seen at 100× in many fields and in high concentration in at least two high-power fields. Necrosis was also

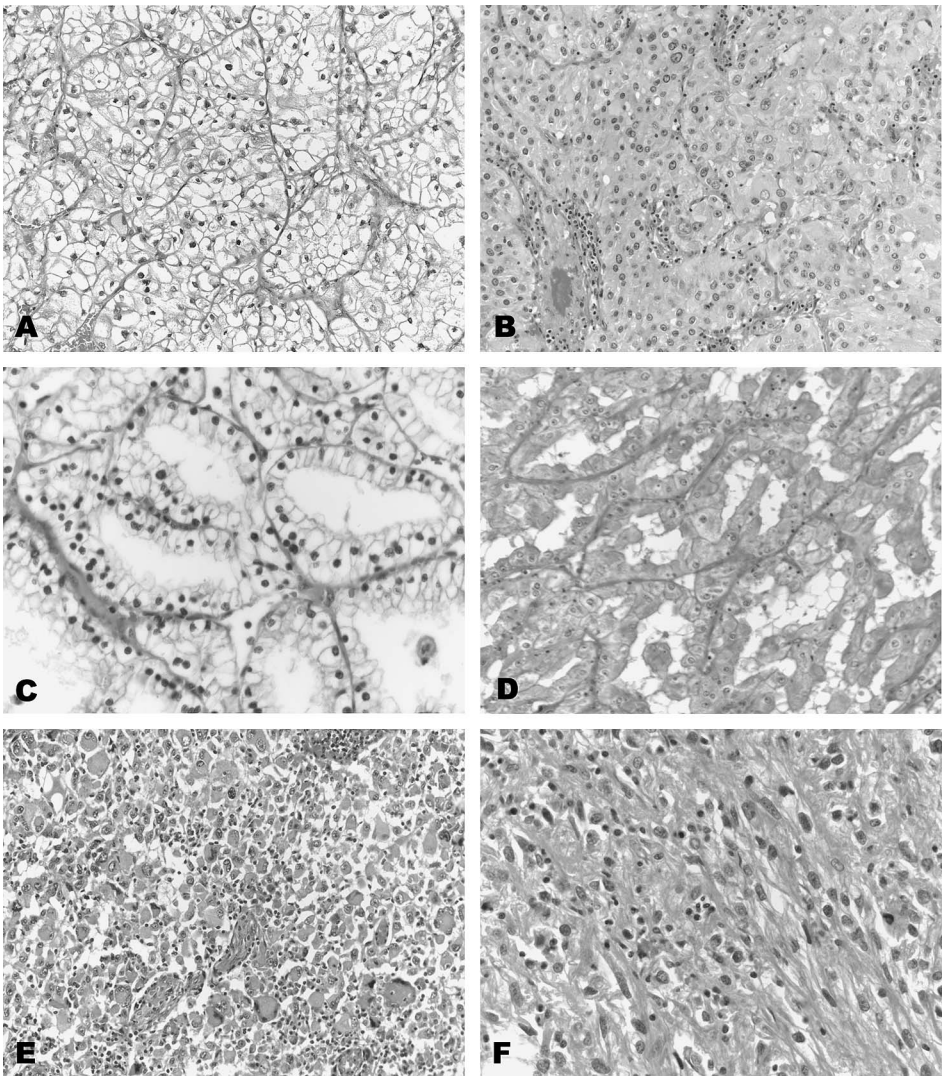

FIGURE 1. Examples of RCC conventional variant (clear cell type) exhibiting (A and B) alveolar morphology: the cells form a solid nested pattern bounded by a connective tissue septum showing prominent vascularity in an arborizing pattern that is delicate, branching, and reminiscent of the alveolar septae of the lung; C and D, tubular morphology: cells line the connective tissue septum, with a lumen-like space in the center; A and C, clear cell features: cells have a foamy cytoplasm; B and D, granular cell features: cell cytoplasm has an eosinophilic and finely granular appearance; E, rhabdoid features: cells are large and epithelioid cells with vesicular nuclei, prominent nucleoli, and prominent paranuclear hyaline or eosinophilic cytoplasmic globules, resembling the appearance of embryonic muscle cells; F, sarcomatoid features: cell possess high-grade spindled or polygonal morphology with occasional fascicular cell arrangement or storiform pattern, resembling a fibrosarcoma.

graded 0 to 3, with 0 = no necrosis identified, 1 = necrosis present in less than one high-power field (400×) and present in less than two areas; 2 = necrosis present at scanning power (100×) in less than two areas and occupying less than the area of one high-power field; 3 = necrosis present with confluence involving more than the area of one high-power field in at least two areas of the slide.

The Fuhrman nuclear grades I to IV were assessed for each carcinoma using standard definitions.[10] For each carcinoma the percentage area with each nuclear grade was estimated and recorded as less than 5%, 5% to 25%, 25% to 50%, or greater than 50%, with additional scoring of the predominant grade and the overall grade. The predominant grade was considered the grade occupying the greatest percentage area of the neoplasm. The overall grade was assigned as originally noted by Fuhrman et al[10] as "the most malignant or highest grade exhibited even if only focal."

Finally, all available staging data, including invasion of perinephric tissues, involvement of renal veins, specifying whether major, minor, or both types of veins were involved, and involvement of regional lymph nodes, were recorded.

Data Analysis

The primary analysis examined the association of pathologic features and response to IL-2. Given that the majority of cases had kidney specimens available, we developed predictive rules using data only from primary kidney carcinomas. Because over 90% of the kidney neoplasms were clear cell carcinomas, and because we observed only one response in a patient with non-clear cell carcinoma, we developed a predictive rule for clear cell carcinomas only. We screened each variable for an association with response, using either Fisher exact test (for a binary variable) or Mantel-Haenszel test for trend when there were ordered categories, with $P < 0.20$ used to identify potential associations between response and each variable in our database. Two-tailed P values, unadjusted for multiple comparisons, were used. Coding of ordered categorical variables was optimized to permit maximum separation of response rate. For example, we compared the degree of separation between

TABLE 1. Distribution of Specimens Requested vs. Received

	All Requested Specimens	All Requested BIDMC Specimens	All Requested CWG Specimens	Kidney Specimens Analyzed	Other Sites Analyzed
No. specimens	444	56	388	163	68
Mean age (yr)	55 ± 7	59 ± 10	54 ± 7	55 ± 9	55 ± 10
Gender (M/F) (% male)	314/130 (71%)	42/14 (75%)	272/116 (70%)	114/49 (70%)	49/19 (72%)
IL-2 treatment					
High-dose IL-2	206 (46%)	11 (20%)	195 (50%)	70 (43%)	32 (47%)
Low-dose IL-2 ± IFN	238 (54%)	45 (80%)	193 (50%)	93 (57%)	36 (53%)
Response rate (CR/PR)					
High-dose IL-2 treatment	40/206 (19%)	3/11 (27%)	37/195 (19%)	16/70 (23%)	7/32 (22%)
Low-dose IL-2 treatment	29/238 (12 %)	4/45 (8%)	25/193 (13%)	15/93 (16%)	0/36 (0%)
Total population	69/444 (16%)	7/56 (12%)	62/388 (16%)	31/163 (19%)	7/68 (10%)
Date of treatment					
1990–1996	196 (44%)	0 (0%)	196 (51%)	14 (9%)	5 (7%)
1997–2001	248 (56%)	56 (100%)	192 (49%)	149 (91%)	63 (93%)

the three prognostic groups when the best prognostic groups were required to have "no granular features" versus "no or <50% granular features." The final model was selected subjectively based on both heuristic considerations (eg, the frequency of the potential predictor; the association of one predictor with another; the optimal coding for the variable) and statistical significance. Once this prognostic model was developed, we applied it without modification to an independent sample of patients whose tumor specimens were collected from metastatic sites as an independent assessment of its validity. In addition, as a separate test of its validity, the model was applied to survival data for all 231 patients. Median survivals for the predefined predictive groups were calculated using a log-rank test applied to the Kaplan-Meier estimated survival distribution. $P < 0.05$ defined statistical significance.

RESULTS

Demographic and Specimen Features

Of the 231 specimens included in this analysis, 163 were from the kidney and 68 were from other sites. The patient, treatment, and response characteristics for specimens requested compared with those received and evaluable for analysis are displayed in Table 1. Most of specimens were collected from patients treated after 1996, and a disproportionate number of patients treated at BIDMC received low-dose IL-2 regimens; otherwise there was no discernible collection bias. Thirty-eight patients (16.5%) with specimens had responded to IL-2-based therapy, similar to the 15.5% response rate for the whole IL-2-treated population for which specimens were sought. Response rates were higher for patients with specimens who received high-dose IL-2 (23% vs. 12%) and for patients treated who received high-dose IL-2 with unresected primary tumors (ie, those with metastatic pathology specimens [22% vs. 0%]), as previously reported.[36]

Table 2 describes the specimens analyzed, while the sources of metastatic specimens are listed in Table 3. Twenty-seven specimens (17 renal and 10 from metastatic sites) were determined to be non-clear cell or indeterminate in type. Specimens received from patients treated on BIDMC trials had similar histologic features to those received from CWG, with 89% and 86% being clear cell, respectively.

TABLE 2. Description of Specimens Analyzed

	Kidney Specimens	Other Sites
n	163	68
Tumor type		
Clear cell	146 (90%)	58 (85%)
Chromophobe	2 (1%)	0
Papillary type I	2 (1%)	0
Papillary type II	10 (6%)	4 (6%)
Indeterminate	3 (2%)	6 (9%)
Tissue of specimen included in analysis		
Kidney	163 (100%)	
Lung		16 (24%)
Other sites		52 (76%)
Type of specimen included in analysis		
Resection	152 (93%)	3 (4%)
Biopsy	11 (7%)	55 (81%)
Fine-needle aspirate	0	10 (15%)

Predicting Response from Kidney Specimens

Tumor response by various pathologic features is presented in Table 4. Tumor responses were seen in 30 of

TABLE 3. Sites of Metastatic Carcinoma Specimens

Site	No.
Bone	21 (31%)
Lung	16 (24%)
Lymph node	11 (16%)
Liver	6 (9%)
Mucosal sites (vagina, GI tract)	5 (7%)
Soft tissue, NOS, including "chest wall"	4 (6%)
Brain	2 (3%)
Adrenal	1 (1%)
Pleura	1 (1%)
Skin	1 (1%)
Total	68 (100%)

146 patients with conventional renal carcinoma (21%) compared with only 1 of 17 patients with non-clear cell histology (6%) ($P = 0.20$). The one tumor response in a patient with non-clear cell carcinoma occurred in a patient originally classified as having a papillary carcinoma (chromophil carcinoma), type II. Subsequent analysis determined that this patient had a collecting duct carcinoma.

The arrangement of cells in clear cell carcinomas is most commonly seen in three patterns: alveolar, tubular, and cystic. We observed a statistically significant difference ($P = 0.02$) in response rate depending on the amount of alveolar component present in the tumors. Tumors with greater than 50% alveolar pattern had a 28% response rate (23 of 83 cases) compared with a 13% response rate (7 of 55 cases) in tumors with less than 50% alveolar pattern and with no responses in five patients with no alveolar pattern. Neither tubular morphology nor cystic morphology was significantly associated with response to IL-2-based therapy (both $P > 0.25$).

For clear cell carcinomas, granular features appeared to predict a poor response to IL-2-based therapy. For tumors with more than 50% granular morphology, the therapeutic response rate was only 5% (1 of 19 cases) compared with 19% (13/67) for tumors with less than 50% granular morphology and 29% (16/56) for tumors without any granular morphology ($P = 0.03$). When we controlled for the presence of rhabdoid features in our analysis, there remained an independent association between granular features and a poor response to IL-2-based treatment. When papillary morphology was assessed as a morphologic component rather than as a tumor type, there was some evidence that it was associated with a lack of response ($P = 0.20$), so it was considered further when developing an overall predictive rule. Neither the presence nor the proportion of rhabdoid or sarcomatoid phenotypes showed any association with response. Similarly, we did not find any association between tumor necrosis, TILs, or nuclear grade and response to IL-2-based therapy.

For renal specimens, we were also able to assess for renal vein involvement. There was a high response rate among patients with major vein only involvement, which led us to compare the overall response rates of those with major vein involvement (30%, 18/60) to those without major vein involvement (14%, 12/86 patients, including 3 responses in 14 patients without adequate material to assess this feature) ($P = 0.02$). As this feature could not be applied to metastatic sites, it was not included in subsequent models.

Application of statistical modeling techniques to these data yielded several predictive models. The model in Table 5 was selected as it involves only four variables, provides reasonably sized groups for each prognostic category, and has a highly significant trend ($P < 0.0001$). This model classified patients with clear cell tumors into three groups. Patients with tumors containing papillary features or more than 50% granular features or without alveolar features had a poor chance of response to IL-2 (3% response rate, 1 response in 33 patients). In contrast, patients with significant (>50%) alveolar and no granular or papillary features had a 39% response rate (14/36 patients). Patients with either less alveolar features or some granular features (<50%) had an intermediate chance of responding to IL-2 (19%, 15/77 patients). The trend across these response groups remained highly significant ($P < 0.001$) even after adjustment for type of IL-2 treatment (high vs. low dose).

TABLE 4. Response Rate by Specific Factors in Kidney Specimens

Tumor Type	
Non-clear cell	1/17 (6%)
Clear cell	30/146 (21%)

Individual Factors in Clear Cell Tumors	**Coding**		
	None	**<50%**	**>50%**
Sarcomatoid	22/104 (21%)	7/23 (30%)	1/15 (7%)
Rhabdoid	19/86 (22%)	8/43 (19%)	2/14 (14%)
Papillary	30/132 (23%)	0/9 (0%)	0/5 (0%)
Granular/eosinophilic	16/56 (29%)	13/67 (19%)	1/19 (5%)
Tubular	11/61 (18%)	17/61 (28%)	2/20 (10%)
Alveolar	0/5 (0%)	7/55 (13%)	23/83 (28%)
Cystic	26/130 (20%)	4/9 (44%)	0/7 (0%)
Solid	18/79 (23%)	3/9 (33%)	0/2 (0%)

	Coding			
TILS and Necrosis	**None**	**Grade 1**	**Grade 2**	**Grade 3**
TILs	5/16 (31%)	6/27 (22%)	9/32 (28%)	2/20 (10%)
Necrosis	2/7 (29%)	4/15 (27%)	2/19 (11%)	13/56 (23%)

Nuclear Fuhrman Grade	**Coding**			
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
Predominant grade*	2/6 (33%)	10/33 (30%)	14/87 (16%)	4/20 (20%)
Overall grade*	0/0 (0%)	2/10 (20%)	11/55 (20%)	17/81 (21%)

Metastatic Invasion	**Coding**		
	No	**Missing/Unknown**	**Yes**
Invasion of perinephric tissue	7/43 (16%)	2/17 (12%)	21/86 (24%)
Regional lymph nodes	5/25 (20%)	16/95 (17%)	9/26 (35%)
Renal vein invasion	8/48 (17%)	3/14 (21%)	19/84 (23%)
Renal vein invasion: by known site			
No known site†	11/62 (18%)		
Major vein only	12/25 (48%)		
Minor vein only	1/24 (4%)		
Both major and minor veins	6/35 (17%)		

*"Predominant grade" is the nuclear Fuhrman grade occupying the greatest percentage area of the carcinoma. "Overall grade" is the highest nuclear Fuhrman grade observed.

†Includes missing information.

Application to Nonrenal Specimens

We then applied this model using alveolar, granular, and papillary features to the 58 clear cell pathology specimens obtained from metastatic sites (Table 6). Five tumor responses were found in the 20 patients in the best prognosis group (25%), with the other two responses occurring in the 22 patients in the intermediate prognosis group (9%). None of the 16 patients in the poor prognosis group responded ($P = 0.02$). As responses occurred only among those treated with high-dose IL-2, the analysis was repeated limited to this group, and the trend in response rate (5/14, 36%; 2/9, 22%; and 0/9, 0% in

TABLE 5. Response Rate by Prognostic Category: Kidney Tumors

Classification	
Best prognosis:	14/36 (39%)
Clear cell tumor	
No papillary features	
Alveolar features >50%	
No granular features	
Intermediate prognosis:	15/77 (19%)
Clear cell tumor	
No papillary features	
Alveolar features	
Granular features <50%	
Poor prognosis:	
Other clear cell patients	1/33 (3%)
Non-clear cell patients*	1/17 (6%)*
Total*	2/50 (4%)*

*Includes 3 patients without sufficient data to classify (none responded).

the best, intermediate, and poor prognosis groups, respectively) was again observed ($P = 0.05$). No responses were seen in the 10 patients with non-clear cell or indeterminate pathology in their metastatic lesions.

Survival Analysis

Survival curves for the three clear cell predictive categories (using all patients with clear cell histology) are shown in Figure 2. Median survivals were 2.87, 1.36, and 0.87 years for the good, intermediate, and poor prognosis groups, respectively ($P < 0.001$). Median survival was 1.41 years in the 27 patients with non-clear cell histology.

DISCUSSION

IL-2 leads to clinical response in 15% to 20% of patients with RCC, but it is associated with considerable toxicity. To reduce risk and to maximize potential benefits of IL-2 therapy, it is desirable to identify the patients who are most likely to respond to treatment. Previous clinical trials have identified clinical and laboratory features predicting poor survival after IL-2 therapy.[22–24] Others have identified clinical and laboratory factors that might be associated with response to IL-2-based therapy.[28,29] Several of these factors, including thyroid dysfunction, thrombocytopenia, or IL-1 production, are treatment-related and thus not useful in selection of patients for IL-2-based therapies. Other factors, such as absence of nephrectomy, presence of bone or liver metastases, or poor performance status, appear to be inadequate predictors of response in patients receiving high-dose IL-2 therapy.[36] Although these clinical and laboratory factors may help to identify patients with RCC whose life expectancy is short even with systemic immunotherapy, they do not help to predict which patients are likely to respond to treatment. Given that response to treatment remains a strong surrogate marker for clinical outcome, identification of factors that can predict for response remains a useful goal.

Little focus to date has been placed on histologic features of RCCs that might be predictive of response. With the identification of multiple distinct patterns of RCC and a new classification system, such an analysis seemed timely and, since pathologic information is available before treatment, of potential clinical value. In addition, with the established clinical utility of debulking nephrectomy before immunotherapy,[37] there is currently greater access to such tissue for histologic review.

Our analysis looked at a variety of pathologic features, first in primary carcinomas of the kidney and then in metastatic lesions, and then associated results with response to IL-2-based therapy. Looking at histologic type of carcinoma, we found a strong trend favoring response in patients with clear cell carcinomas compared with those with variant tumors, with the lone response in the latter subset occurring in a patient with a collecting duct carcinoma. The observation of poor response to immunotherapy in variant-type RCC is consistent with findings by Motzer et al,[38] who observed only one partial response

TABLE 6. Response Rate by Prognostic Category: Other Sites

Classification	
Best prognosis:	5/20 (25%)
Clear cell tumor	
No papillary features	
Alveolar features >50%	
No granular features	
Intermediate prognosis:	2/22 (9%)
Clear cell tumor	
No papillary features	
Alveolar features	
Granular features <50%	
Poor prognosis:	
Other clear cell patients	0/16*
Non-clear cell patients*	0/10*
Total*	0/26 (0%)

*Includes 6 patients without sufficient data to classify (none responded).

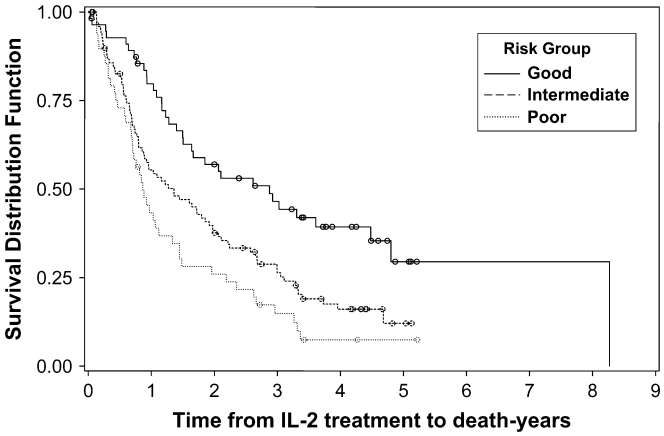

FIGURE 2. Kaplan-Meier survival curves from time of IL-2 therapy for all 204 patients with clear cell morphology separated into the three groups predictive of response to IL-2-based therapy.

among 31 patients with variant RCC treated with IFN alfa-2a, IL-2, or the combination. The single response in their analysis occurred in a patient with chromophobe histology. None of the 18 patients with papillary carcinoma responded to systemic IFN-based therapy. The difference in response rate observed between clear cell and variant RCC supports the exclusion of patients with non-clear cell carcinomas from IL-2 and/or IFN-based regimens.

Within the clear cell subtype, response to IL-2 was associated with the presence of alveolar features and the absence of granular or papillary features. An independent analysis of metastatic lesions supported the significance of the features identified in the initial analysis of primary kidney specimens. Combining the groups of patients with clear cell histology in kidney or metastatic specimens, it is evident that patients with any papillary, more than 50% granular, or no alveolar features are very unlikely to respond to IL-2-based therapy (1 response in 45 patients, 2%). In addition, the survival curves for the three prognostic groups in the model correspond closely with the responsiveness to IL-2, providing corroboration for the model. While it is impossible to distinguish the extent to which these survival differences reflect either distinct natural histories of these different histotypes or merely the predictable effect of tumor response on survival, these survival data suggest that the model may have prognostic as well as predictive capabilities.

The alveolar phenotype is the most classic of the described clear cell patterns. The lack of responsiveness of patients with tumors exhibiting papillary or granular features may result from additional mutations that either reduce immune responsiveness or induce tumor-associated immune suppression. As advanced RCC has been associated with immune suppression manifest by downmodulation of T-cell receptor ζ chain expression and diminished dendritic cell maturation,[39–41] further investigation of the relationship between the various histologic patterns and evidence of immune suppression is warranted.

Our experience suggests that patients whose RCC possesses poor predictive features including variant type, papillary or more than 50% granular features or tumors without any alveolar pattern should be considered for non-IL-2 based therapy. While this model requires validation from an independent data set, its use would enable the exclusion of approximately one third of patients from consideration for IL-2-based therapy. Unfortunately, sufficient clinical and laboratory data were not available in this retrospective, multi-institutional investigation to incorporate this information into the predictive model. However, in that most clinical features were established as prognostic variables for survival rather than as predictive features of response to IL-2, they may not have influenced the current model. As the survival data presented are based on a model developed from response rather than an independent analysis, it must be viewed cautiously. Furthermore, any true survival model involving histologic features should by necessity incorporate the well-established clinical prognostic features as described by Motzer and others.[19–24] Such an analysis was beyond the scope of this report. Nonetheless, it is conceivable that histologic features of clear cell tumors might contribute significant independent prognostic information to the existing clinical variables and might further identify patients who are poor candidates for IL-2-based therapy.

Our analysis combines patients who received multiple different IL-2 treatment regimens. Recent data from two large-scale randomized studies suggest that the response rate to high-dose IL-2 is significantly better than for low-dose IL-2 either alone[32] or in combination with IFNα.[36] Therefore, it is conceivable that patients with the favorable pathologic features might have a higher response rate if they were treated only with high-dose IL-2. Nonetheless, the fact that the majority of patients in the intermediate and even the good predictive categories did not respond to IL-2-based therapy indicates that more selective criteria are still needed. Given that tumor features are shown to be associated with response, this raises the hope that more sophisticated molecular analyses may identify specific gene expression products or surface proteins that might be predictive of response. Such tests could then be applied to pathology specimens to help identify the most appropriate therapy for a particular patient, be it IL-2 or a more recently developed therapy.

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