

# Predictors of Long-Term Outcome in Patients with Well-Differentiated Gastroenteropancreatic Neuroendocrine Tumors After Peptide Receptor Radionuclide Therapy with $^{177}\text{Lu}$ -Octreotate

Samer Ezziddin<sup>1</sup>, Mared Attassi<sup>1</sup>, Charlotte J. Yong-Hing<sup>2</sup>, Hojjat Ahmadzadehfar<sup>1</sup>, Winfried Willinek<sup>3</sup>, Frank Grünwald<sup>4</sup>, Stefan Guhlke<sup>1</sup>, Hans-Jürgen Biersack<sup>1</sup>, and Amir Sabet<sup>1</sup>

<sup>1</sup>Department of Nuclear Medicine, University Hospital, Bonn, Germany; <sup>2</sup>Department of Radiology, University of British Columbia Hospital, Vancouver, British Columbia, Canada; <sup>3</sup>Department of Radiology, University Hospital, Bonn, Germany; and <sup>4</sup>Department of Nuclear Medicine, University Hospital, Frankfurt, Germany

Outcome analyses for patients with gastroenteropancreatic neuroendocrine tumors (GEP NET) after peptide receptor radionuclide therapy (PRRT) are still limited, especially with regard to the impact of the Ki-67 index. Using a single-center analysis, we aimed to establish predictors of survival. **Methods:** We retrospectively analyzed a consecutive cohort of 74 patients who had metastatic GEP NET and underwent PRRT with  $^{177}\text{Lu}$ -octreotate (mean activity of 7.9 GBq per cycle, aimed at 4 treatment cycles at standard intervals of 3 mo). Patients (33 with pancreatic NET and 41 with nonpancreatic GEP NET) had unresectable metastatic disease graded as G1 or G2 (G1/G2) and documented morphologic or clinical progression within less than 12 mo or uncontrolled disease under somatostatin analog treatment. Responses were evaluated according to modified Southwest Oncology Group criteria. Potential predictors of survival were analyzed with the Kaplan–Meier curve method (log-rank test) and multivariate analysis ( $P < 0.05$ ). **Results:** The response rates were 36.5% partial response, 17.6% minor response, 35.1% stable disease, and 10.8% progressive disease for the entire cohort; 54.5% partial response, 18.2% minor response, 18.2% stable disease, and 9.1% progressive disease for pancreatic NET; and 22.0% partial response, 17.1% minor response, 48.8% stable disease, and 12.2% progressive disease for nonpancreatic GEP NET. The median progression-free survival and overall survival were 26 mo (95% confidence interval, 18.3–33.7) and 55 mo (95% confidence interval, 48.8–61.2), respectively. Besides the Ki-67 index, a Karnofsky performance score of less than or equal to 70%, a hepatic tumor burden of greater than or equal to 25%, and a baseline plasma level of neuron-specific enolase of greater than 15 ng/mL independently predicted shorter overall survival (hazard ratio, 2.1–3.1). Patients with a Ki-67 index of greater than 10% still had median progression-free survival and overall survival of 19 and 34 mo, respectively. **Conclusion:** The results of this study demonstrated the favorable response and long-term outcome of patients with G1/G2 GEP NET after PRRT. Independent predictors of survival were the Ki-67 index, the patient's performance status (Karnofsky performance scale score), the tumor burden, and the baseline neuron-specific enolase level. Even patients with a Ki-67 index of greater than 10%

seemed to benefit from PRRT, with a good response and a notable long-term outcome. We present the first evidence, to our knowledge, that even in patients with metastatic disease the distinction between G1 and G2—in particular, between G1 (Ki-67 index of 1%–2%) and low-range G2 (Ki-67 index of 3%–10%)—provides prognostic stratification.

**Key Words:** gastroenteropancreatic tumors; neuroendocrine tumors; peptide receptor radionuclide therapy;  $^{177}\text{Lu}$ -octreotate; tumor grading

**J Nucl Med 2014; 55:183–190**

DOI: 10.2967/jnumed.113.125336

**P**eptide receptor radionuclide therapy (PRRT) is a highly efficient modality for the systemic treatment of gastroenteropancreatic neuroendocrine tumors (GEP NET) (1–4). The compound [ $^{177}\text{Lu}$ -DOTA<sup>0</sup>,Tyr<sup>3</sup>]octreotate ( $^{177}\text{Lu}$ -octreotate) is frequently used for this purpose. Outstanding response and survival data are available (1), and with the growing importance of this treatment modality, the relevance of outcome predictors (2) is becoming increasingly meaningful for multidisciplinary management of tumors.

The first analysis of the impact of the Ki-67 index on the efficacy of PRRT has been reported (5). Interestingly, it was demonstrated that indices of up to 20% had no discernible influence on tumor response. However, analyses of the impact of the proliferation parameter on survival in the context of PRRT were not yet available. We aimed to assess the impacts of various baseline variables, including the Ki-67 index, on outcome in the well-characterized population of patients in whom the previously reported factor analysis was performed.

## MATERIALS AND METHODS

We retrospectively analyzed a consecutive cohort of patients who had well-differentiated GEP NET and a known Ki-67 index ( $\leq 20\%$ ) and underwent PRRT at our institution. In addition to giving written informed consent for their treatment, patients also gave written informed consent for the scientific analysis of their data; the local ethics committee approved the study. All 74 patients were part of a previously published study (5), and 23 of the patients were included in a more recent analysis of bone metastatic GEP NET (4). None of the patients was included in a therapy trial.

Received Apr. 23, 2013; revision accepted Sep. 6, 2013.

For correspondence or reprints contact: Samer Ezziddin, Department of Nuclear Medicine, University Hospital, Sigmund-Freud-Strasse 25, D-53105 Bonn, Germany.

E-mail: [samer.ezziddin@ukb.uni-bonn.de](mailto:samer.ezziddin@ukb.uni-bonn.de)

Published online Jan. 16, 2014.

COPYRIGHT © 2014 by the Society of Nuclear Medicine and Molecular Imaging, Inc.

## Patients

The patient cohort consisted of 74 consecutive patients (mean age, 62.5 y; range, 34–83 y; 42 men and 32 women) who had well-differentiated GEP NET graded as G1 or G2 (G1/G2) according to the current World Health Organization classification; who were treated with PRRT at the University Hospital of Bonn; and who had complete restaging and follow-up (Table 1). Thirty-three patients had pancreatic NET, and 41 patients had nonpancreatic GEP NET; of the latter, 4 had foregut, 19 had midgut, and 2 had hindgut GEP NET, and 16 had GEP NET with an unknown primary tumor. Metastatic sites included the liver in 58 patients (78.4%), bone in 28 patients (37.8%), and other organs in 12 patients (16.2%). Previous treatments comprised surgery ( $n = 38$ ; 51.4%), biotherapy ( $n = 28$ ; 37.8%), chemotherapy ( $n = 18$ ; 24.3%), and locoregional treatment ( $n = 13$ ; 17.6%). PRRT was the first-line systemic therapy in 25 patients (33.8%). At baseline, before the initiation of PRRT, there was documented clinical progression ( $n = 16$ ; 21.6%) or morphologic progression ( $n = 56$ ; 75.7%) within less than 12 mo or uncontrolled disease under somatostatin analog treatment ( $n = 8$ ; 10.8%). For the purpose of the present study, hepatic tumor burden at baseline was retrospectively assessed according to pretreatment CT or MR imaging, and patients were categorized into 4 groups of liver involvement: none, less than 25%, 25%–50%, and greater than 50% of the liver volume. The cutoff value of 25% was then identified as the best predictive separator and consequently used for all survival analyses. Tumor uptake was classified according to the  $^{177}\text{Lu}$ -octreotate therapy scan of the first PRRT cycle (grade 3, greater than that of the kidney or spleen; grade 2, greater than that of the liver; and grade 1, approximately that of the liver). This was done to standardize the uptake score for the entire cohort because some patients had scintigraphic scans (e.g., OctreoScan; Covidien) and others had PET-based pretherapeutic somatostatin receptor imaging.

## Histopathology

Tumors were classified according to the current TNM staging and grading system for NET (6–8). All tumors were well-differentiated endocrine tumors, according to histopathology, with the presence of distant metastases (TNM stage IV). Histologic and immunohistochemical analyses, including determination of the Ki-67 proliferation index, were performed on resection specimens ( $n = 35$ ; 47.3%) or biopsy material ( $n = 39$ ; 52.7%). The Ki-67 index was expressed as the percentage of MIB1 antibody-stained tumor cells in areas in which the highest level of nuclear labeling was observed (6,7). The median time interval between the Ki-67 index assessment and the

initiation of PRRT (first treatment cycle) was 14.5 mo; in 38 patients (51.4%), the interval was greater than 12 mo. The time interval was insignificantly shorter for pancreatic tumors (median, 9 mo) than for nonpancreatic tumors (median, 16 mo) ( $P = 0.313$ ). Categorization of the proliferation of tumors as G1 (Ki-67 index of <3%), low-range G2 (Ki-67 index of 3%–10%), and high-range G2 (Ki-67 index of 15%–20%) was done retrospectively to analyze the potential impact on survival. There were no Ki-67 indices between 10% and 15%, in accordance with current standard practice for reporting the immunostaining of NET.

## PRRT

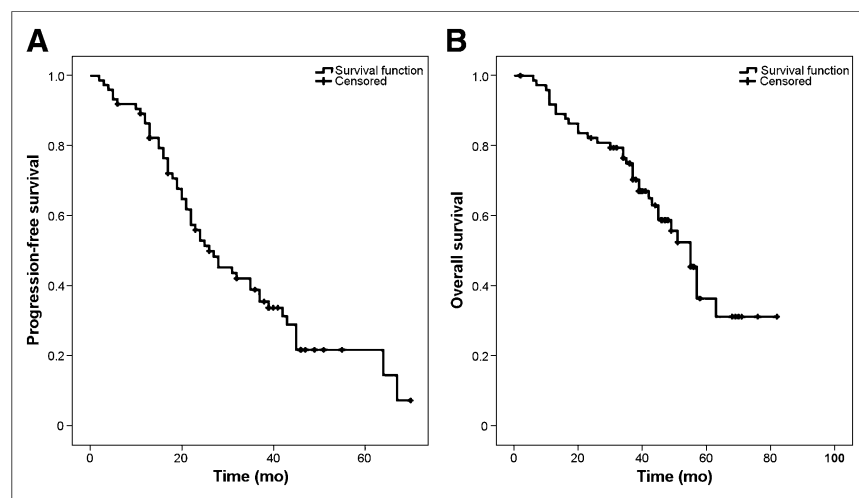
The inclusion criteria for treatment with PRRT were histologically confirmed, unresectable, metastatic GEP NET; sufficient tracer uptake (greater than or equal to that of normal liver) on baseline somatostatin receptor imaging; a glomerular filtration rate of greater than 30 mL/min/1.73 m<sup>2</sup>; a white blood cell count of greater than or equal to  $2 \times 10^9/\text{L}$ ; and a platelet count of greater than  $70 \times 10^9/\text{L}$ . PRRT was performed by the administration of a mean activity of 7.9 GBq of  $^{177}\text{Lu}$ -octreotate per treatment cycle, aimed at 4 courses at standard intervals of 3 mo (10–14 wk). At the time of administration,  $^{177}\text{Lu}$  (IDB Holland) had a specific activity of approximately 100–160 GBq/ $\mu\text{mol}$ . Peptide labeling (9,10) was performed to obtain an apparent specific activity of about 54 GBq/ $\mu\text{mol}$  (ratio of activity to the total amount of peptide). Nephroprotection was implemented with standard amino acid coinfusion according to the Rotterdam protocol (11,12) (2.5% lysine and 2.5% arginine in 1 L of 0.9% NaCl; infusion of 250 mL/h). Short-acting somatostatin analogs were required to be paused 1 d before the administration of  $^{177}\text{Lu}$ -octreotate, and long-acting analogs were required to be paused a minimum of 6 wk before PRRT. Informed consent was obtained from all patients before the initiation of therapy and before the administration of each treatment cycle.

## Response Assessment

Restaging was performed 3 mo after the termination of PRRT. Imaging consisted of CT or MR imaging according to the baseline imaging modality. Follow-up imaging was performed at 6-mo intervals after the first restaging. Responses were evaluated according to modified Southwest Oncology Group solid tumor response criteria as described previously (5,13) and were classified as partial response, minor response (25%–49% decrease in the sum of perpendicular diameters), stable disease, and progressive disease. Documented tumor progression at any time before the end of treatment led to the termination of PRRT and the classification of the patient as having progressive disease. Because complete remission was not observed in the present study, this term does not appear in the analyses. The results of standard functional imaging (pre- and posttreatment somatostatin receptor imaging) were not incorporated into the response characterization in the present study.

## Outcome and Statistical Analyses

The baseline characteristics of the study population were analyzed with regard to the tumor response. For this purpose, the Fisher exact test was applied after dichotomization for each factor and the resulting response: regression (partial response or minor response) versus nonregression (stable disease or progressive disease). Overall survival (OS)



**FIGURE 1.** Kaplan–Meier curves for PFS (A) and OS (B) of entire study cohort. Median PFS from start of treatment was 26 mo (95% CI, 18.3–33.7), and median OS was 55 mo (95% CI, 48.8–61.2).

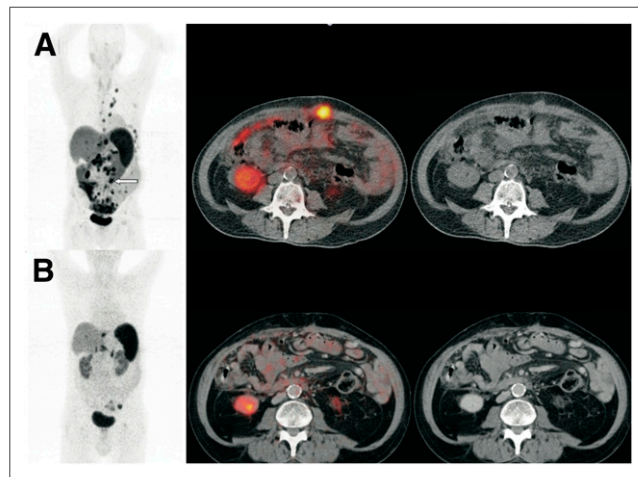
and progression-free survival (PFS) were analyzed with the Kaplan-Meier curve method (log-rank test) ( $P < 0.05$ ). Patients were censored at the start of another antiproliferative treatment, such as chemotherapy or salvage PRRT. Univariate analysis with the log-rank test was performed for each baseline factor. Multivariate analysis (Cox proportional hazards model) by use of the stepwise model with backward elimination was performed with significant variables from the log-rank test. The Fisher exact test was used for comparing proportions of regression in patient groups dichotomized for baseline characteristics. For all tests, a 2-sided  $P$  value of less than 0.05 was considered significant. The statistical software package SPSS (version 18.0; SPSS Inc.) was used to analyze the data.

## RESULTS

The median follow-up period was 47 mo (95% confidence interval [CI], 44.5–49.5), and the median OS of the entire cohort ( $n = 74$ ) (Fig. 1) was 55 mo (95% CI, 48.8–61.2). Thirty-four patients (45.9%) had died by the end of the study. No treatment-related deaths were observed. Relevant transient myelosuppression (grade 3 or 4; Common Terminology Criteria for Adverse Events, version 3.0) occurred in less than 10% of treatments. No irreversible toxicity, including renal toxicity (grade 3 or 4), was noted.

The observed response rates were 36.5% partial response, 17.6% minor response, 35.1% stable disease, and 10.8% progressive disease for the entire cohort; 54.5% partial response, 18.2% minor response, 18.2% stable disease, and 9.1% progressive disease for the pancreatic NET cohort; and 22.0% partial response, 17.1% minor response, 48.8% stable disease, and 12.2% progressive disease for the nonpancreatic GEP NET cohort. Figure 2 shows a partial response to PRRT in a patient with a pancreatic NET; the Ki-67 index was 20%. The analysis of various baseline factors for potential contributions to responses is shown in Table 2.

The median PFS was 26 mo (95% CI, 18.3–33.7) from the start of treatment (Fig. 1). The only factors contributing to time to progression in the univariate analysis were the Ki-67 index (cutoff, 10%; log-rank test,  $P = 0.02$ ) (Fig. 3A) and hepatic tumor burden



**FIGURE 2.** Regression of multiple metastases illustrated by  $^{68}\text{Ga}$ -DOTATOC PET/CT imaging before (A) and 3 mo after (B) PRRT in patient with metastatic NET of pancreas. Maximum-intensity-projection PET images (coronal views) are shown on left; fused and unfused CT images are shown on right. Selected lesion is indicated by arrow. This patient remained in partial remission for 20 mo; proliferation index (Ki-67 index) was 20%.

**TABLE 1**  
Patient Characteristics and Proportions Receiving PRRT as First-Line Treatment

Variable	Patients (n)	PRRT as first-line treatment*	$P$
Total	74	25 (33.8)	
Age			
$\leq 65$ y	35	13 (37.1)	0.563
$> 65$ y	39	12 (30.8)	
Tumor type			
P-NET	33	16 (48.5)	0.016
Other GEP NET	41	9 (22.0)	
Performance status			
KPS of $\leq 70$	27	5 (18.5)	0.035
KPS of $> 70$	47	20 (42.6)	
Functionality			
Nonfunctional	55	21 (38.2)	0.174
Functional	19	4 (21.1)	
CgA level			
$\leq 600$ ng/mL	45	13 (28.9)	0.267
$> 600$ ng/mL	29	12 (41.4)	
NSE level			
$\leq 15$ ng/mL	39	16 (41.0)	0.164
$> 15$ ng/mL	35	9 (25.7)	
Ki-67 index			
$\leq 10\%$	60	19 (31.7)	0.533
$> 10\%$	14	6 (42.9)	
$\leq 2\%$ (G1)	26	10 (38.5)	0.453
3%–10% (low-range G2)	34	9 (26.5)	
15%–20% (high-range G2)	14	6 (42.9)	
Hepatic tumor burden <sup>†</sup>			
$< 25\%$	37	17 (45.9)	0.027
$\geq 25\%$	37	8 (21.6)	
Tracer uptake			
$\leq$ Grade 2	9	2 (22.2)	0.709
$>$ Grade 2	65	23 (35.4)	

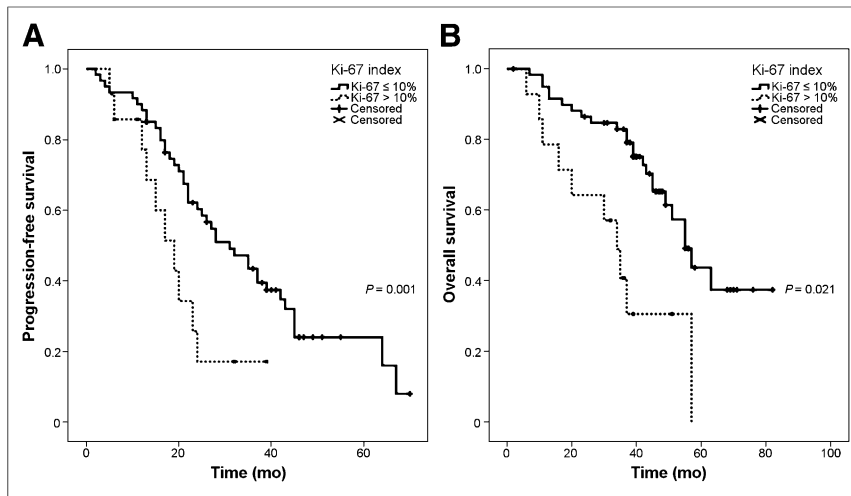
\*Reported as number of patients, with percentage in parentheses.

<sup>†</sup>Fraction of total liver volume.

P-NET = pancreatic NET; CgA = chromogranin A.

(cutoff, 25%; log-rank test,  $P = 0.006$ ). Patients with a Ki-67 index of greater than 10% had a median PFS of 19 mo (95% CI, 12.4–25.6), and patients with a Ki-67 index of less than or equal to 10% had a median PFS of 31 mo (95% CI, 22.1–39.9).

Analysis of the potential impact of various factors on OS is shown in Table 2. Of the contributing factors in the univariate analysis (log-rank test), only the Ki-67 index, tumor burden, Karnofsky performance score (KPS), and neuron-specific enolase (NSE) level at baseline remained significant in the multivariate analysis (Cox regression). Among the risk factors, a Ki-67 index of greater than 10%, a KPS of less than or equal to 70, and a plasma NSE concentration of greater than 15 ng/mL had hazard ratios of approximately 3; a hepatic tumor burden of greater than or equal to 25% was associated with a hazard ratio of 2.1 (Table 3). The Kaplan-Meier curves in Figures 3–6 illustrate the prognostic value of these factors. The use of PRRT as a first-line treatment for metastatic disease did not affect outcome in our cohort ( $P = 0.936$  for PFS;  $P = 0.364$  for OS).



**FIGURE 3.** PFS (A) and OS (B) stratified by tumor proliferation index (Ki-67 index of  $\leq 10\%$  vs.  $> 10\%$ ).

When the proliferation of tumors was stratified as G1 (Ki-67 index of  $< 3\%$ ), low-range G2 (Ki-67 index of  $3\%–10\%$ ), and high-range G2 (Ki-67 index of  $15\%–20\%$ ), the Kaplan–Meier

**TABLE 2**  
Morphologic Response to PRRT According to Various Baseline Factors

Variable	Patients (n)	Regression*	P
Total	74	40 (54.1)	
Age			
$\leq 65$ y	35	18 (51.4)	0.816
$> 65$ y	39	22 (56.4)	
Tumor type			
P-NET	33	24 (72.7)	0.005
Other GEP NET	41	16 (39.0)	
Performance status			
KPS $\leq 70$	27	13 (48.1)	0.476
KPS $> 70$	47	27 (57.4)	
Functionality			
Nonfunctional	55	32 (58.2)	0.289
Functional	19	8 (42.1)	
CgA			
$\leq 600$ ng/mL	45	23 (51.1)	0.634
$> 600$ ng/mL	29	17 (58.6)	
NSE			
$\leq 15$ ng/mL	39	23 (59.0)	0.484
$> 15$ ng/mL	35	17 (48.6)	
Ki-67 index			
$\leq 10\%$	60	32 (53.3)	1.0
$> 10\%$	14	8 (57.1)	
$\leq 2\%$ (G1)	26	13 (50.0)	0.873
$3\%–10\%$ (low-range G2)	34	19 (55.9)	
$15\%–20\%$ (high-range G2)	14	8 (57.1)	
Tracer uptake			
$\leq$ Grade 2	9	2 (22.2)	0.071
$>$ Grade 2	65	38 (58.5)	

\*Regression (partial response or minor response) is reported as number of patients, with percentage in parentheses.

P-NET = pancreatic NET; CgA = chromogranin A.

curve analysis revealed a significant impact on PFS and OS (Fig. 7). Even the distinction between G1 and low-range G2 tumors was significant for the outcome; the median OS for patients with low-range G2 tumors was 49.0 mo (95% CI, 37.3–60.7), whereas the median OS for patients with G1 tumors was not reached after 82 mo ( $P = 0.04$ ). The same was true for the distinction between low-range G2 and high-range G2 tumors (median OS of 34.0 mo; 95% CI, 25.8–42.2) ( $P = 0.038$ ). With regard to PFS, patients with G1 tumors had a significantly better outcome (median PFS of 43.0 mo; 95% CI, 37.4–48.6) than patients with low-range G2 tumors (median PFS of 24.0 mo; 95% CI, 16.6–31.4) ( $P = 0.008$ ), as shown in Figure 7.

For the small subgroup of patients with a Ki-67 index of greater than 10% ( $n = 14$ ), there was no significant impact of hepatic tumor burden ( $P = 0.628$ ) or tumor uptake ( $P = 0.586$ ). In patients with a Ki-67 index of less than or equal to 10% ( $n = 60$ ), a hepatic tumor burden of greater than or equal to 25% was associated with a shorter median OS (45 mo vs. not reached after 76 mo) ( $P = 0.016$ ), whereas tumor uptake again had no impact on survival ( $P = 0.832$ ).

Interestingly, patients in our cohort who had an unknown primary tumor had a significantly worse outcome than patients with nonpancreatic GEP NET for which the origin was determined (“carcinoid” of the foregut, midgut, or hindgut), that is, a shorter PFS ( $P = 0.001$ ) and a shorter OS ( $P = 0.003$ ).

## DISCUSSION

This retrospective study of 74 patients with well-characterized GEP NET showed the strength of PRRT with  $^{177}\text{Lu}$ -octreotate, even in patients with advanced metastatic disease. The overall response rates of 36.5% (partial response) and 54.1% (regression rate; partial response plus minor response) and the median PFS and OS (26 and 55 mo, respectively) are highly promising and compare favorably with those achieved with other treatment modalities (14–18). The data were analyzed in accordance with the new World Health Organization criteria (2010) and should help with comparisons of outcome data from different patient cohorts in various treatment studies. However, given the nature of this retrospective series, with multiple potential biases—including retrospectively selected cutoff points—care must be taken in applying the results to clinical practice. Rather, the results provide strong evidence and a rational starting point for a prospective evaluation of the prognostic impact of various factors.

Well-differentiated GEP NET, especially those of nonpancreatic origin, show only a minor response to systemic chemotherapy. Initial studies with various chemotherapeutic agents reported a limited clinical benefit and a high rate of overall toxicity (19–25). Subsequent studies reported similar response rates after treatment with a temozolomide-based regimen (26–29), with a median PFS of up to 18 mo. Recent trials evaluated a variety of novel targeted agents (14,17,18,30–33) with disappointing low response rates ( $< 20\%$ ) for gastrointestinal NET. The outcomes proved significantly better than those of placebo-treated control groups with pancreatic NET; the median PFS in patients treated with sunitinib

**TABLE 3**  
Univariate and Multivariate Analyses of Potential Factors Contributing to OS

Factor	OS (mo)		Univariate analysis* (P)	Multivariate analysis	
	Median	95% CI		Hazard ratio (95% CI)	P
All patients	55	49–61			
Age					
≤65 y	55	46–64			
>65 y	51	38–64	0.526		
Tumor type					
P-NET	57	48–66			
Other GEP NET	43	31–55	0.037		0.173
Performance status					
KPS ≤ 70	39	30–48			
KPS > 70	57	47–67	0.003	2.7 (1.3–5.5)	0.006
Functionality					
Nonfunctional	55	50–60			
Functional	43	NA	0.630		
CgA					
≤600 ng/mL	55	NA			
>600 ng/mL	43	32–54	0.052		
NSE					
≤15 ng/mL	57	45–69			
>15 ng/mL	45	40–53	0.032	2.8 (1.3–5.9)	0.006
Ki-67 index					
≤10%	55	47–62			
>10%	34	25–42	0.001	3.1 (1.4–6.9)	0.004
Hepatic tumor burden†					
<25%	NR	NA			
≥25%	43	37–50	0.007	2.1 (1.0–4.5)	0.044
Tracer uptake					
≤ Grade 2	55	NA			
> Grade 2	55	44–66	0.774		

\*Log-rank test.

†Fraction of total liver volume.

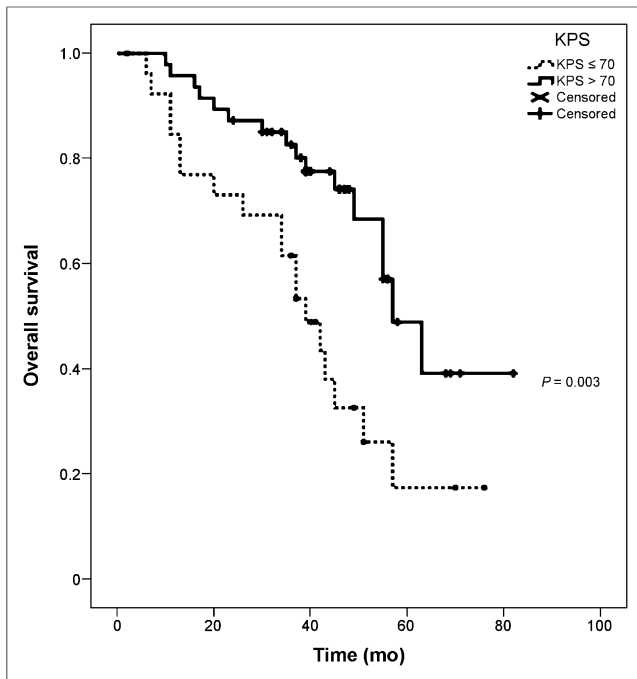
P-NET = pancreatic NET; CgA = chromogranin A; NR = not reached; NA = not available because of censored cases.

or everolimus was up to 12 mo (30,34). Our finding of a median PFS of 27 mo and a median PFS of 25 mo for nonpancreatic GEP NET and pancreatic NET, respectively, compare favorably with the findings for systemic treatment in these historic control groups. However, these comparisons should be considered with caution and lack substantial validity, although they may provide some indication of efficacy in the absence of comparative trials in the field of PRRT.

The strongest predictor of outcome in our patient cohort was the proliferation marker Ki-67 index. It contributed to PFS and was 1 of 4 independent variables affecting survival after PRRT. Although the Ki-67 index is being increasingly recognized as a powerful determinant of survival in patients with GEP NET (35–38), its relevance in unresectable metastatic disease and potential cutoff values in the intermediate proliferative range for any treatment modality are still undefined because of a lack of data. In addition, the only published evidence for PRRT, to our knowledge, is an analysis of the impact of the Ki-67 index on responses (5), with no survival data available at that time. That study proved that indices within the entire G1/G2 range (i.e., Ki-67 of ≤20%) had no discernible proliferation–response relationship. Because the documented response was PFS for at least 12 mo (restaging performed >12 mo after the start of treatment), the reported data were clinically meaningful. With the data from the present study,

it becomes clear that even though G2 tumors with a Ki-67 index of greater than 10% respond in a manner similar to that of tumors with a Ki-67 index of less than or equal to 10%, they show earlier progression after PRRT (median PFS of 19 mo vs. 31 mo) and produce shorter survival times (median OS of 34 mo vs. 55 mo). However, even this “impaired” survival of patients with NET in the “upper” G2 range is encouraging (median PFS and OS of 19 and 34 mo, respectively). It appears to be at least equal to that achieved with other treatment modalities (14), with reported OS of 11–24 mo (well-differentiated metastatic GEP NET) and 7–27 mo (well-differentiated metastatic pancreatic NET). We are aware that we should be cautious when making comparisons with historic control groups; nevertheless, it is fair to state that even the presented outcome for the subgroup with an unfavorable prognosis (Ki-67 of >10%) still indicates effective treatment and clearly does not provide an argument against performing PRRT in this subgroup.

Interestingly, stratification into G1 (Ki-67 index of <3%), low-range G2 (Ki-67 index of 3%–10%), and high-range G2 (Ki-67 index of 15%–20%) also had a prognostic impact on PFS and OS (Fig. 7). In particular, the distinction between G1 and low-range G2 tumors was significant with regard to PFS and OS ( $P = 0.008$  and  $P = 0.04$ , respectively). Although it is well known that G1 and G2 tumors have different prognoses because of a divergence



**FIGURE 4.** OS stratified by pretreatment KPS ( $\leq 70$  vs.  $> 70$ ).

in the tendency to metastasize, we present the first evidence, to our knowledge, that even in patients with metastatic disease the G1/G2 distinction—in particular, between G1 (Ki-67 index of 1%–2%) and low-range G2 (Ki-67 index of 3%–10%)—provides prognostic stratification in a uniformly treated cohort. However, it remains unclear whether Ki-67 acts simply as a broad prognostic marker (i.e., patients with tumors with higher Ki-67 indices would naturally be expected to have shorter times to progression and shorter survival times, perhaps independent of treatment) or as a more specific predictor of the efficacy of PRRT.

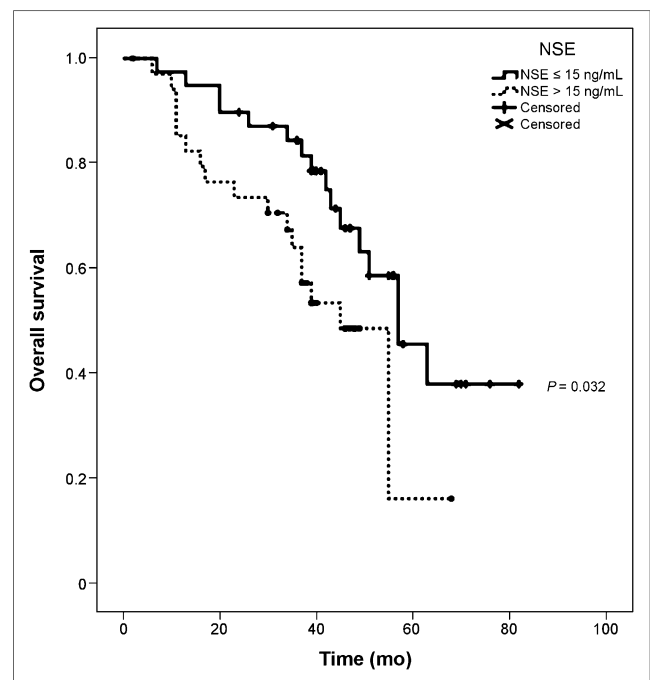
Other factors contributing to OS were the patient's performance status (Karnofsky performance score) (Fig. 4) and the plasma NSE level (Fig. 5). It is not surprising that the Karnofsky parameter predicted survival (2), although it is noteworthy that patients with a KPS of less than or equal to 70% still had a median OS of 39 mo (95% CI, 30–48) after PRRT. Also, the baseline NSE level was recently reported to be negatively associated with survival in patients with pancreatic NET in an interventional study (39), suggesting a prognostic impact of this secretory tumor marker. Although this marker was confirmed to be a general predictor for OS in our cohort, it lacked a respective significant impact in the subgroup analyses of pancreatic tumors ( $P = 0.06$ ) and nonpancreatic tumors ( $P = 0.12$ ). Larger investigations may eventually identify tumor subcategories in which an elevated baseline NSE level is of major prognostic relevance.

Previous work established the unfavorable prognostic character of functioning pancreatic tumors (2). In the present study, the responses of pancreatic NET consisted of a partial response in 54.5% of patients, a minor response in 18.2% of patients, stable disease in 18.2% of patients, and progressive disease in 9.1% of patients. Therefore, regression (partial response plus minor response) was observed in 72.7% patients with pancreatic NET. The median PFS was 25 mo (95% CI, 17–33), and the median OS was 57 mo (95% CI, 48–66). These outcomes were notable and compared well with those

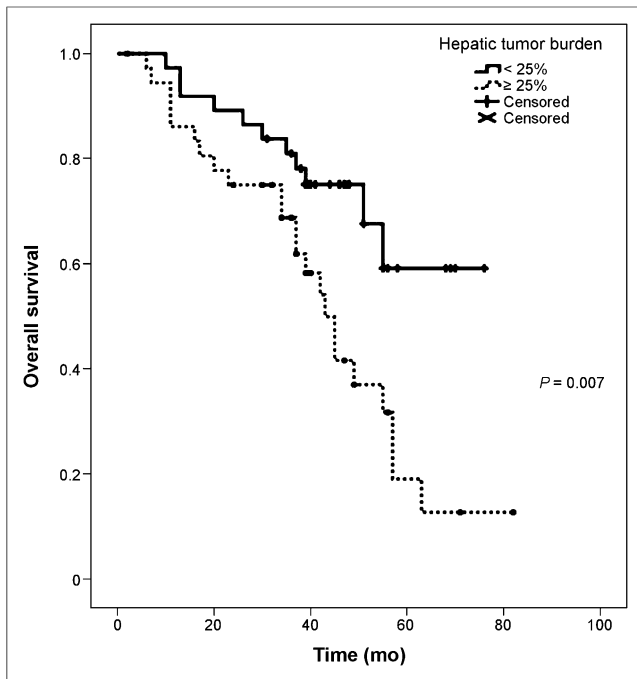
achieved with other current treatment modalities (14,18,27,30,40). For nonpancreatic GEP NET, the regression rate (partial response plus minor response) was 39.1%. The median PFS was 27 mo (95% CI, 16–38), and the median OS was 43 mo (95% CI, 31–55).

The known predictive factor tumor burden (Fig. 6) was confirmed by our analysis, in that patients with hepatic tumor involvement of greater than or equal to 25% of liver volume at baseline CT or MR imaging had a significantly shorter median OS (43 mo) than the remaining patients (median OS not reached after 76 mo). The difference remained significant in the subgroup with a Ki-67 index of less than 10% and proved to be an independent factor in the multivariate analysis (hazard ratio, 2.1) ( $P = 0.044$ ). Another factor of known prognostic relevance, tumor uptake (2,3), did not have a significant predictive impact on either PFS or OS. Because the first  $^{177}\text{Lu}$ -octreotate therapy scan was uniformly used to classify tumor uptake, this parameter may perform in a manner different from that of others. On the basis of the  $^{177}\text{Lu}$ -octreotate scan, most patients fell into the high-uptake group (grade 3; in our cohort, 65/74 patients), probably providing less optimal stratification than a  $^{111}\text{In}$ -DTPA-octreotide scan (2) or a  $^{90}\text{Y}$ -DOTATOC scan (3). Perhaps a quantitative measurement method or a qualitative assessment at some earlier point after treatment would allow better stratification on the basis of the degree of tumor uptake. It would make biologic sense to expect some correlation between radiation dose and response or outcome.

The main limitation of the present study is its retrospective nature. However, because of the general lack of available prospective studies, potential predictors of outcome for patients with NET have been predominantly analyzed by use of retrospective data (16,35,36,38,41–44). The advantage of the present study is that our cohort of patients underwent the same treatment modality. The parameter Ki-67 index is associated with inherent inaccuracy regarding the time and localization of assessments, but this fact reflects clinicians' reality and the use of the marker



**FIGURE 5.** OS stratified by pretreatment plasma NSE level ( $\leq 15$  ng/mL vs.  $> 15$  ng/mL).



**FIGURE 6.** OS stratified by pretreatment hepatic tumor burden (<25% vs.  $\geq 25\%$  of liver volume).

in routine settings as well as in clinical studies investigating and demonstrating the prognostic value of this parameter. It is not current practice to routinely assess the Ki-67 index at different sites during the course of disease, certainly because of the invasive nature of the assessment. The time lag–related inaccuracy of the proliferation index in the present study must be emphasized; however, despite the considerable interval between assessment and the start of PRRT (median, 14.5 mo; >12 mo in 38 patients), the immunohistochemical parameter proved to have high predictive power. Interestingly, even in the subgroup of

patients with an interval of more than 12 mo between assessment and the start of PRRT ( $n = 38$ ), the Ki-67 index was highly predictive of shorter survival times; the median OS in patients with a Ki-67 index of less than or equal to 10% was 55 mo, and that in patients with a Ki-67 index of greater than 10% was 34 mo ( $P = 0.003$ ). To overcome the obvious problem of temporal and spatial inaccuracy resulting from the invasive nature of the proliferation assessment, supplemental alternative methods for tumor grading, such as noninvasive, whole-body molecular imaging for metabolic grading with  $^{18}\text{F}$ -FDG (45,46) or proliferation markers might be helpful in the future.

## CONCLUSION

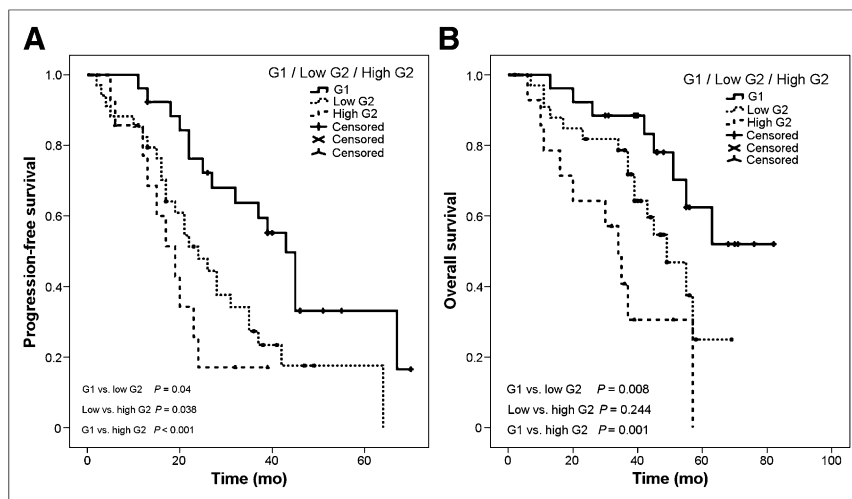
In the present study, we demonstrated a favorable outcome for patients with well-differentiated GEP NET graded as G1-2 after PRRT. Independent predictors of survival were the Ki-67 index, the patient's performance status, the hepatic tumor burden, and the baseline NSE level. Even patients with a Ki-67 index of greater than 10% seemed to benefit from PRRT in terms of response and long-term outcome. Although it is well known that G1 and G2 tumors have different prognoses because of a divergence in the tendency to metastasize, we present the first evidence, to our knowledge, that even in patients with metastatic disease the G1/G2 distinction—specifically, between G1 (Ki-67 index of 1%–2%) and low-range G2 (Ki-67 index of 3%–10%)—provides prognostic stratification in a uniformly treated cohort. However, given the nature of this retrospective series, with multiple potential biases—including retrospectively selected cutoff points—care must be taken in applying the results to clinical practice. Rather, the results provide preliminary evidence and a rational starting point for prospective studies to establish the impact of the proposed predictors.

## DISCLOSURE

The costs of publication of this article were defrayed in part by the payment of page charges. Therefore, and solely to indicate this fact, this article is hereby marked “advertisement” in accordance with 18 USC section 1734. No potential conflict of interest relevant to this article was reported.

## ACKNOWLEDGMENTS

We are grateful to Professor Eric Krenning, Professor Dik Kwekkeboom, and Professor Wouter A.P. Breeman (Erasmus Medical Center, Rotterdam, The Netherlands) for sharing their invaluable experience in the receptor-targeting field and making somatostatin receptor-mediated treatment at all possible at our institution. Also, we thank Professor Richard P. Baum (Department of Nuclear Medicine and PET Center, Zentralklinik, Bad Berka, Germany) for his continuous critical and constructive input in this field. We also are thankful to the personnel of the Department of Nuclear Medicine and especially the nursing staff of the therapy ward.



**FIGURE 7.** PFS (A) and OS (B) stratified by tumor proliferation index into G1 (Ki-67 index of <3%;  $n = 26$ ), low-range G2 (Ki-67 index of 3%–10%;  $n = 34$ ), and high-range G2 (Ki-67 index of 15%–20%;  $n = 14$ ). Median PFS times were 43.0 mo (95% CI, 37.4–48.6), 24.0 mo (95% CI, 16.6–31.4), and 19.0 mo (95% CI, 12.4–25.6), respectively. Median OS was not reached after 82 mo, 49.0 mo (95% CI, 37.3–60.7), and 34.0 mo (95% CI, 25.8–42.2), respectively.

## REFERENCES

- Kwekkeboom DJ, de Herder WW, van Eijck CH, et al. Peptide receptor radionuclide therapy in patients with gastroenteropancreatic neuroendocrine tumors. *Semin Nucl Med*. 2010;40:78–88.
- Kwekkeboom DJ, de Herder WW, Kam BL, et al. Treatment with the radiolabeled somatostatin analog [<sup>177</sup>Lu-DOTA<sup>0</sup>,Tyr<sup>3</sup>]octreotate: toxicity, efficacy, and survival. *J Clin Oncol*. 2008;26:2124–2130.
- Imhof A, Brunner P, Marincek N, et al. Response, survival, and long-term toxicity after therapy with the radiolabeled somatostatin analogue [<sup>90</sup>Y-DOTA]-TOC in metastasized neuroendocrine cancers. *J Clin Oncol*. 2011;29:2416–2423.
- Ezziddin S, Sabet A, Heinemann F, et al. Response and long-term control of bone metastases after peptide receptor radionuclide therapy with <sup>177</sup>Lu-octreotate. *J Nucl Med*. 2011;52:1197–1203.
- Ezziddin S, Opitz M, Attassi M, et al. Impact of the Ki-67 proliferation index on response to peptide receptor radionuclide therapy. *Eur J Nucl Med Mol Imaging*. 2011;38:459–466.
- Rindi G, Kloppel G, Couvelard A, et al. TNM staging of midgut and hindgut (neuro)endocrine tumors: a consensus proposal including a grading system. *Virchows Arch*. 2007;451:757–762.
- Rindi G, Kloppel G, Alhman H, et al. TNM staging of foregut (neuro)endocrine tumors: a consensus proposal including a grading system. *Virchows Arch*. 2006;449:395–401.
- Rindi G. The ENETS guidelines: the new TNM classification system. *Tumori*. 2010;96:806–809.
- Breeman WA, De Jong M, Visser TJ, Erion JL, Krenning EP. Optimising conditions for radiolabelling of DOTA-peptides with <sup>90</sup>Y, <sup>111</sup>In and <sup>177</sup>Lu at high specific activities. *Eur J Nucl Med Mol Imaging*. 2003;30:917–920.
- Breeman WA, van der Wansem K, Bernard BF, et al. The addition of DTPA to [<sup>177</sup>Lu-DOTA<sup>0</sup>,Tyr<sup>3</sup>]octreotate prior to administration reduces rat skeleton uptake of radioactivity. *Eur J Nucl Med Mol Imaging*. 2003;30:312–315.
- Kwekkeboom DJ, Teunissen JJ, Bakker WH, et al. Radiolabeled somatostatin analog [<sup>177</sup>Lu-DOTA<sup>0</sup>,Tyr<sup>3</sup>]octreotate in patients with endocrine gastroenteropancreatic tumors. *J Clin Oncol*. 2005;23:2754–2762.
- Kwekkeboom DJ, Bakker WH, Kam BL, et al. Treatment of patients with gastroenteropancreatic (GEP) tumours with the novel radiolabelled somatostatin analogue [<sup>177</sup>Lu-DOTA<sup>0</sup>,Tyr<sup>3</sup>]octreotate. *Eur J Nucl Med Mol Imaging*. 2003;30:417–422.
- Green S, Weiss GR. Southwest Oncology Group standard response criteria, endpoint definitions and toxicity criteria. *Invest New Drugs*. 1992;10:239–253.
- Basu B, Sirohi B, Corrie P. Systemic therapy for neuroendocrine tumours of gastroenteropancreatic origin. *Endocr Relat Cancer*. 2010;17:R75–R90.
- Kwekkeboom DJ, Kam BL, van Essen M, et al. Somatostatin-receptor-based imaging and therapy of gastroenteropancreatic neuroendocrine tumors. *Endocr Relat Cancer*. 2010;17:R53–R73.
- Gupta S, Johnson MM, Murthy R, et al. Hepatic arterial embolization and chemoembolization for the treatment of patients with metastatic neuroendocrine tumors: variables affecting response rates and survival. *Cancer*. 2005;104:1590–1602.
- Yao JC, Phan AT, Chang DZ, et al. Efficacy of RAD001 (everolimus) and octreotide LAR in advanced low- to intermediate-grade neuroendocrine tumors: results of a phase II study. *J Clin Oncol*. 2008;26:4311–4318.
- Raymond E, Dahan L, Raoul JL, et al. Sunitinib malate for the treatment of pancreatic neuroendocrine tumors. *N Engl J Med*. 2011;364:501–513.
- Moertel CG, Lefkopoulo M, Lipsitz S, Hahn RG, Klaassen D. Streptozocin, streptozocin-fluorouracil or chlorozotocin in the treatment of advanced islet-cell carcinoma. *N Engl J Med*. 1992;326:519–523.
- Moertel CG, Hanley JA, Johnson LA. Streptozocin alone compared with streptozocin plus fluorouracil in the treatment of advanced islet-cell carcinoma. *N Engl J Med*. 1980;303:1189–1194.
- Engstrom PF, Lavin PT, Moertel CG, Folsch E, Douglass HO Jr. Streptozocin plus fluorouracil versus doxorubicin therapy for metastatic carcinoid tumor. *J Clin Oncol*. 1984;2:1255–1259.
- Bukowski RM, Tangen CM, Peterson RF, et al. Phase II trial of dimethyltriazenoimidazole carboxamide in patients with metastatic carcinoid: a Southwest Oncology Group study. *Cancer*. 1994;73:1505–1508.
- Rougier P, Oliveira J, Ducreux M, Theodore C, Kac J, Droz JP. Metastatic carcinoid and islet cell tumours of the pancreas: a phase II trial of the efficacy of combination chemotherapy with 5-fluorouracil, doxorubicin and cisplatin. *Eur J Cancer*. 1991;27:1380–1382.
- Bajetta E, Rimassa L, Carnaghi C, et al. 5-Fluorouracil, dacarbazine, and epirubicin in the treatment of patients with neuroendocrine tumors. *Cancer*. 1998;83:372–378.
- Cheng PN, Saltz LB. Failure to confirm major objective antitumor activity for streptozocin and doxorubicin in the treatment of patients with advanced islet cell carcinoma. *Cancer*. 1999;86:944–948.
- Kulke MH, Stuart K, Enzinger PC, et al. Phase II study of temozolomide and thalidomide in patients with metastatic neuroendocrine tumors. *J Clin Oncol*. 2006;24:401–406.
- Strosberg JR, Fine RL, Choi J, et al. First-line chemotherapy with capecitabine and temozolomide in patients with metastatic pancreatic endocrine carcinomas. *Cancer*. 2011;117:268–275.
- Fine RL, Gulati AP, Krantz BA, et al. Capecitabine and temozolomide (CAPTEM) for metastatic, well-differentiated neuroendocrine cancers: the Pancreas Center at Columbia University experience. *Cancer Chemother Pharmacol*. 2013;71:663–670.
- Ekeblad S, Sundin A, Janson ET, et al. Temozolomide as monotherapy is effective in treatment of advanced malignant neuroendocrine tumors. *Clin Cancer Res*. 2007;13:2986–2991.
- Yao JC, Shah MH, Ito T, et al. Everolimus for advanced pancreatic neuroendocrine tumors. *N Engl J Med*. 2011;364:514–523.
- Pavel ME, Hainsworth JD, Baudin E, et al. Everolimus plus octreotide long-acting repeatable for the treatment of advanced neuroendocrine tumours associated with carcinoid syndrome (RADIANT-2): a randomised, placebo-controlled, phase 3 study. *Lancet*. 2011;378:2005–2012.
- Shah MH, Young D, Kindler HL, et al. Phase II study of the proteasome inhibitor bortezomib (PS-341) in patients with metastatic neuroendocrine tumors. *Clin Cancer Res*. 2004;10:6111–6118.
- Duran I, Kortmansky J, Singh D, et al. A phase II clinical and pharmacodynamic study of temsirolimus in advanced neuroendocrine carcinomas. *Br J Cancer*. 2006;95:1148–1154.
- Kulke MH, Lenz HJ, Meropol NJ, et al. Activity of sunitinib in patients with advanced neuroendocrine tumors. *J Clin Oncol*. 2008;26:3403–3410.
- Jann H, Roll S, Couvelard A, et al. Neuroendocrine tumors of midgut and hindgut origin: tumor-node-metastasis classification determines clinical outcome. *Cancer*. 2011;117:3332–3341.
- Scarpa A, Mantovani W, Capelli P, et al. Pancreatic endocrine tumors: improved TNM staging and histopathological grading permit a clinically efficient prognostic stratification of patients. *Mod Pathol*. 2010;23:824–833.
- Pape UF, Jann H, Muller-Nordhorn J, et al. Prognostic relevance of a novel TNM classification system for upper gastroenteropancreatic neuroendocrine tumors. *Cancer*. 2008;113:256–265.
- Pape UF, Berndt U, Muller-Nordhorn J, et al. Prognostic factors of long-term outcome in gastroenteropancreatic neuroendocrine tumours. *Endocr Relat Cancer*. 2008;15:1083–1097.
- Yao JC, Pavel M, Phan AT, et al. Chromogranin A and neuron-specific enolase as prognostic markers in patients with advanced pNET treated with everolimus. *J Clin Endocrinol Metab*. 2011;96:3741–3749.
- Yao JC, Lombard-Bohas C, Baudin E, et al. Daily oral everolimus activity in patients with metastatic pancreatic neuroendocrine tumors after failure of cytotoxic chemotherapy: a phase II trial. *J Clin Oncol*. 2010;28:69–76.
- Vilar E, Salazar R, Perez-Garcia J, Cortes J, Oberg K, Tabernero J. Chemotherapy and role of the proliferation marker Ki-67 in digestive neuroendocrine tumors. *Endocr Relat Cancer*. 2007;14:221–232.
- Durante C, Boukheris H, Dromain C, et al. Prognostic factors influencing survival from metastatic (stage IV) gastroenteropancreatic well-differentiated endocrine carcinoma. *Endocr Relat Cancer*. 2009;16:585–597.
- Panzuto F, Nasoni S, Falconi M, et al. Prognostic factors and survival in endocrine tumor patients: comparison between gastrointestinal and pancreatic localization. *Endocr Relat Cancer*. 2005;12:1083–1092.
- Clancy TE, Sengupta TP, Paulus J, Ahmed F, Duh MS, Kulke MH. Alkaline phosphatase predicts survival in patients with metastatic neuroendocrine tumors. *Dig Dis Sci*. 2006;51:877–884.
- Binderup T, Knigge U, Loft A, Federspiel B, Kjaer A. <sup>18</sup>F-fluorodeoxyglucose positron emission tomography predicts survival of patients with neuroendocrine tumors. *Clin Cancer Res*. 2010;16:978–985.
- Garin E, Le Jeune F, Devillers A, et al. Predictive value of <sup>18</sup>F-FDG PET and somatostatin receptor scintigraphy in patients with metastatic endocrine tumors. *J Nucl Med*. 2009;50:858–864.