Prognostic factors influencing survival from metastatic (stage IV) gastroenteropancreatic well-differentiated endocrine carcinoma

Cosimo Durante¹, Houda Boukheris², Clarisse Dromain³, Pierre Duvillard⁴, Sophie Leboulleux¹, Dominique Elias⁵, Thierry de Baere², David Malka⁶, Jean Lumbroso¹, Joël Guigay⁶, Martin Schlumberger¹, Michel Ducreux⁶ and Eric Baudin¹

¹Service de Médecine Nucléaire et de Cancérologie Endocrinienne ²INSERM u605, ³Département de Radiologie,

⁴Département d'Anatomo-Pathologie, ⁵Département de Chirurgie and, ⁶Département de Médecine, Institut Gustave-Roussy, Université Paris XI, 39 rue Camille Desmoulins, 94805 Villejuif Cedex, France

(Correspondence should be addressed to E Baudin; Email: baudin@igr.fr)

Abstract

Survival of metastatic gastroenteropancreatic well-differentiated endocrine carcinoma (GEP WDEC) is not well characterized. We evaluated the long-term outcome and prognostic factors for survival in 118 patients with distant metastases from GEP WDEC. Inclusion criteria were 1) pathological review by a single pathologist according to the present WHO criteria, 2) absence of previous therapy apart from surgery, 3) complete morphological evaluation within 3 months including somatostatin receptor scintigraphy, and 4) follow-up at Gustave-Roussy Institute until death or study's end. Clinical, biological marker, and pathological parameters were analyzed in univariate and multivariate statistical models. Survival after the first complete imaging work-up of the metastatic disease was determined using Kaplan–Meier method. Overall, survival for 5 years after the diagnosis of metastatic disease was 54%. In multivariate analysis, age (hazard ratio (HR): 1.05, 95% confidence interval (CI): 1.01–1.08, $P=0.01$), the number of liver metastases (HR: 3.4, 95% CI: 1.4–8.3, $P=0.01$), tumor slope (HR: 1.1, 95% CI: 1.0–1.1, $P=0.001$), and initial surgery (HR: 0.3, 95% CI: 0.1–0.8, $P=0.01$) were predictive of survival. Five-year survival was 100%, 91% (95% CI, 51–98%), 62% (95% CI, 37–83%), and 9% (95% CI, 6–32%) when patients had 0, 1, 2, 3 or more poor prognostic features respectively. This study enables the stratification of metastatic GEP WDEC patients into distinct risk groups. These risk categories can be used to tailor therapeutic approaches and also to design and interpret clinical trials.

Endocrine-Related Cancer (2009) 16 585–597

Introduction

Gastroenteropancreatic neuroendocrine tumors (GEP NET), recently renamed endocrine tumors, are a group of tumors defined by the expression of a set of structural proteins and hormonal products common to neurons and endocrine cells (Plöckinger *et al.* 2004, Baudin 2007).

Defining prognostic factors for survival is a critical objective in oncology to evaluate therapeutic results, to tailor therapeutic trials and the aggressiveness of treatment. This is particularly true in GEP tumors that

can be associated with a diverse natural history. Epidemiological studies of GEP NET have reported increase age as a major prognostic feature (Quaadvlieg *et al.* 2001, Zar *et al.* 2004, Lepage *et al.* 2007) but conflicting results regarding the prognostic effect of gender (Beasley *et al.* 2000, Quaadvlieg *et al.* 2001, Modlin *et al.* 2003, Zar *et al.* 2004, Tomassetti *et al.* 2006, Lepage *et al.* 2007). Additionally, in previous multivariate analysis from tertiary referral centers, pathological differentiation and stage at diagnosis have

been identified as the strongest prognostic features of GEP NET. Indeed, differentiation of GEP NET classified as poorly compared with well-differentiated (WD) carcinoma (Johnson *et al.* 1986, Greenberg *et al.* 1987, Madeira *et al.* 1998, Travis *et al.* 1998, Rindi *et al.* 1999, Lim *et al.* 2005, Panzuto *et al.* 2005, Asamura *et al.* 2006, Faggiano *et al.* 2007, Lepage *et al.* 2007, Pape *et al.* 2008) and stage (Lo *et al.* 1996, Pelosi *et al.* 1996, Burke *et al.* 1997, Madeira *et al.* 1998, Kirshbom *et al.* 1999, Rindi *et al.* 1999, Shebani *et al.* 1999, Onaitis *et al.* 2000, Quaedvlieg *et al.* 2001, Rigaud *et al.* 2001, Solorzano *et al.* 2001, Hochwald *et al.* 2002, Plöckinger *et al.* 2004, Panzuto *et al.* 2005, Tomassetti *et al.* 2006, Baudin 2007, Pape *et al.* 2008) as long been recognized the strongest prognostic parameters before the primary location. More recently, lymph node metastases (Hellman *et al.* 2002, Cardillo *et al.* 2004, Lim *et al.* 2005, Tomassetti *et al.* 2005, Baudin 2007, García-Yuste *et al.* 2007, Rea *et al.* 2007, Pape *et al.* 2008) and proliferative index have emerged as major determinant of prognosis in GEP tumors of the lung (Travis *et al.* 1998, Beasley *et al.* 2000, Lim *et al.* 2005, Asamura *et al.* 2006, Baudin 2007) stomach and pancreas (Chaudhry *et al.* 1992, Pelosi *et al.* 1996, Rindi *et al.* 1999, Rigaud *et al.* 2001, Hochwald *et al.* 2002). At the end, a new classification emphasizing the role of TNM (Tumor, Nodes, Metastases) classification and proliferative index has emerged (Rindi *et al.* 2006, 2007, Bettini *et al.* 2008, Fischer *et al.* 2008).

The vast majority of previous studies but one has focused on GEP tumors of various stages. However, progress in subgroup of GEP NET patients characterized by a given differentiation and stage is expected since these two parameters are the strongest prognostic parameters of GEP NET and since each stage of GEP NET carries a specific set of clinical characteristics.

We therefore conducted a single-center, retrospective multivariate analysis of previously untreated, metastatic (stage IV), GEP WD endocrine carcinoma (WDEC), 1) to identify prognostic factors related to survival not affected by the therapy at the time of definition and 2) to evaluate the prognostic value of morphologically defined tumor burden slope.

Materials and methods

Patients

Two hundred and fifteen consecutive patients referred to Gustave-Roussy Institute between January 1994 and June 2001 for treatment of a metastatic GEP well-differentiated endocrine carcinoma (GEP WDEC) was

included in a retrospective analysis. Inclusion criteria were: 1) pathological diagnosis of metastatic GEP WDEC (stage IV) according to WHO and TNM classification diagnostic criteria and confirmed by a single pathologist (PD; *n*=11 patients excluded, because tumor samples were not available: Solcia *et al.* 2000, Heitz *et al.* 2004, Travis *et al.* 2004, Rindi *et al.* 2006, 2007); in the absence of a solid or diffuse histologic architectural pattern and/or presence of necrosis, a mitotic index >10 or a Ki-67 score >10% was not considered sufficient as a single criteria to classify the tumor as poorly differentiated; 2) initial visit including no prior treatment for GEP WDEC, except for surgery (*n*=53 patients excluded); 3) distant metastatic disease analyzed by complete morphological evaluation, including somatostatin receptor scintigraphy performed within 3 months of study entry (*n*=13 patients excluded); and 4) diagnostic work-up and follow-up in a single institution (Gustave-Roussy Institute; *n*=20 patients excluded). All of the patients who met the inclusion criteria for this study underwent a clinical examination and biological and morphological testing at the time of initial visit and every 3–6 months until the end of the study (October 31, 2004) or death.

Treatment given for GEP WDEC after enrolment in the study is presented in Table 1. Results of these therapies have been previously published (Mitry *et al.* 1999, Aparicio *et al.* 2001, Elias *et al.* 2003, Roche *et al.* 2003, 2004, Delaunoy *et al.* 2004).

Table 1 Therapeutic management of gastroenteropancreatic well-differentiated endocrine carcinoma (GEP WDEC)

Type of treatment ^a	Number	Percent
Treatments before study initiation	52	44
Primary tumor surgery	46	39
Liver metastases surgery	8	15
Others metastases surgery	12	23
Treatments after study initiation	116	82
Primary tumor surgery	30	26
Liver metastases surgery	24	21
Others metastases surgery	16	14
Systemic chemotherapy	70	60
Number of chemotherapy regimens		
1	27	39
2	23	33
≥3	20	28
Chemoembolization	44	38
Radiotherapy	24	21
Somatostatin analogs	77	66
Interferon	12	10
Peptide receptor radionuclide therapy	1	1

^aTreatments were not mutually exclusive.

Pathological, biological, and morphological work-up

Initial evaluation included a careful interview history and physical examination. Clinical, laboratory, imaging, and histologic data were collected prospectively and analyzed by skilled pathologist and radiologist specialists (PD, CD). During the course of study, chromogranin A (CGA) measurement and liver magnetic resonance imaging (MRI) became part of routine clinical practice in 1998 and September 2000 respectively, and incorporated into the study protocol.

Tumoral tissue was obtained from the primary tumor ($n=74$; 63%) or metastases ($n=44$; 37%), by either surgical resection ($n=75$; 64%) or biopsy ($n=43$; 36%) allowing proliferative index assessment. At least 10 high-power fields (HPFs) were examined on hematoxylin and eosin section to determine the mitotic index using a Zeiss Axioplan microscope ($40\times/0.7$ objective, $10\times$ ocular was used; each field corresponding to a surface of 0.33 mm^2). Ki-67 score was assessed using MIB1 (mindbomb homolog 1) stains (clone MIB1, Dako A/S, Glostrup, Denmark).

Laboratory investigations included serum gastrin, pancreatic peptides, urinary 5-hydroxyindoleacetic acid (5-HIAA) levels in patients with pancreas, ileum or lung endocrine tumors according to a standard clinical practice (Plöckinger *et al.* 2004, Baudin *et al.* 2007). Finally, serum glucose, serum calcium, neuron specific enolase or CGA were measured in all patients as previously described (Baudin *et al.* 1998, 1999, Baudin 2007).

To determine the extent of metastatic disease during the initial evaluation, patients underwent conventional imaging studies. Abdominal and chest computed tomography (CT) scan was performed with a conventional scanner and after 1998 with a spiral scanner; images were obtained before and after i.v. injection of iodinated contrast material. CT of the brain was performed, if clinical symptoms were suggestive of brain metastases. Liver MRIs were performed with a 1.5-T whole-body imager, and included both T_1 -weighted and fat-suppressed T_2 -weighted sequences but also dynamic contrast-enhanced MRIs as previously described (Dromain *et al.* 2005). Furthermore, all patients underwent somatostatin receptor scintigraphy after i.v. injection of 170–220 MBq of indium-111-DTPA-phe1-octreotide (Dromain *et al.* 2005). Only lesions larger than 1 cm in diameter were taken into account to determine the number of lung metastases on CT. Bone involvement was quantified by specifying the number of metastases detected by

somatostatin receptor scintigraphy. Bone-related clinical events (pain, spinal cord compression, and/or fractures) were also recorded. All liver CTs and MRIs were reviewed to determine the number of liver metastases and tumor slope. Indeed, in a subgroup of patients, in whom a second imaging work-up was carried out in our institution, spontaneous tumor slope was evaluated by performing a CT scan or MRI 3–6 months after initial assessment. Results of tumor slope were normalized per month to allow comparison between patients. Comparison of the sum of the longest diameters of target lesions at two imaging and/or, appearance of one or more new lesions were analyzed to classify the tumor slope as progressive (whatever the rate of progression) or stable (Therasse *et al.* 2000). Patients were either untreated or in the case of functioning GEP tumors, had received somatostatin analogs at the time of tumor slope assessment.

Statistical analysis

The following clinical information extracted from patients charts was recorded in all patients: gender, age (≤ 65 or > 65 years), embryologic origin of the primary tumor (foregut, midgut or hindgut), anatomical location of the primary tumor (lung, pancreas or ileum), presence or absence of hormone-related symptoms (≤ 3 episodes/day = no; > 3 episodes/day = yes), time elapsed between diagnosis of primary tumor and diagnosis of metastatic disease (synchronous; metachronous), liver involvement (yes/no), lung involvement (yes/no), bone involvement (yes/no), number of lung metastases (< 5 , 5–9, or ≥ 10), bone metastases (< 3 , 3–5 or > 5), number of organs involved with metastatic tumor (1, 2 or ≥ 3). Surgery before study entry and treatment given during the course of the study were also recorded.

Tumor mitotic count (< 2 , 2–10, or > 10 per 10 HPF) was available for 82 cases, Ki-67 index (< 2 , 2–10, or $> 10\%$) for 50 cases, and 86 patients had either tumor mitotic count or Ki-67 index. Very few tumors expressed mitotic counts above 20 or Ki-67 index above 20% explaining why these cut off values were not selected despite recent recommendations (34, 35). 5-HIAA or CGA (< 2 , 2–4 or ≥ 5) levels or at least one of these measurements were found in 60, 89, and 101 cases respectively. Eighty-five patients had information on the number of metastatic liver lesions (< 10 , ≥ 10). Tumor slope (0, if non-progressive or > 0 , if progressive) was determined over a 3–6 month period in 66 patients, including 19 patients receiving somatostatin analogs at the time of tumor slope assessment.

Overall, survival rates and 95% confidence intervals (CIs) were estimated using the Kaplan–Meier method (Kaplan & Meier 1958). Rates were computed with life-table analysis. The log-rank test (Lee 1986) was used to compare survival curves. Univariate and multivariate analysis of survival were performed with follow-up starting at the time of first complete imaging work-up, as described above, after diagnosis of distant metastasis. Univariate analysis included the above-mentioned variables. Multivariate analysis included only variables that were significant in the univariate analysis and was performed with a backward variable selection technique. The risk of death was computed as hazard ratio (HR). The effect of the variables that were significant in the multivariate analysis was evaluated using a stepwise Cox proportional-hazards regression model from the time of diagnosis of the first tumor metastasis to the end of the study or the patient's death (Cox 1972). For all analyses, two-sided tests were employed and the 0.05 level of significance was used. Ninety-five percent CIs were calculated for the HR estimated from the Cox proportional hazard regression analysis. All analyses were performed using SAS software (version 9.1, SAS Institute, Cary, NC, USA).

Results

Characteristics of patients at discovery of distant metastases

The characteristics of the 118 remaining patients enrolled into this retrospective study are listed in Table 2. Briefly, the study population included 60 men (51%) and 58 women (49%). The mean age was 57 years (s.d.: 13 years; range: 12–90 years) at the time of first complete imaging work-up of metastatic disease. The mean follow-up was 4.6 years (s.d.: 2.9 years; median: 4 years; range: 0.2–14 years) from the time of first complete imaging work-up of metastatic disease.

Concerning primary, 58 (57%) arose from the foregut, most from the pancreas (35 patients) and 39 (39%) arose from the midgut, most from the ileum (33 patients); four (4%) arose from the hindgut, all arising from the rectum (Table 2) and 17 (14%) remained unknown. Clinical symptoms related to hormonal overproduction included cutaneous flushing and or diarrhea in 24 (20%) patients. Multiple endocrine neoplasia type 1 (MEN1) were diagnosed in three patients.

Mitotic counts above 2 or 10 was observed in 35 and 10% of patients respectively, while Ki-67 index values above 2 or 10% was observed in 58 and 18% of patients respectively.

An increase (≥ 5 the high upper limit) in at least one hormonal marker was observed in 32 of 101 patients (32%), including CGA and or 5-HIAA in 20 (33%) and 17 (19%) patients respectively.

Synchronous or metachronous metastases were observed in 96 (81%) or 22 (19%) patients respectively. In the latter case, the first metastasis was diagnosed after a mean ($\pm$ s.d.) period of 6.1 ± 6 years (range 1–26 years) from the time of primary tumor diagnosis. At study entry, the majority of patients had liver metastases ($n=108$; 92%). Among the 85 patients for whom the number of liver metastases could be properly determined, 31 (37%) had 1–4 metastases, 19 (22%) had 5–9 metastases, and 35 (37%) had 10 or more. Extra-hepatic metastatic sites were seen in 49 of the study subjects (Table 3). Only three patients experienced bone pain; spinal cord compression or fractures were not observed. The spread of metastases involved one organ site in 73 (62%) patients, two in 41 (35%) and three in four (3%). Among the 29 (44%) patients who experienced a tumor progression, 11 (38%) and 18 (62%) patients experienced a tumor rate of progression below or above 20% respectively. Results were not significantly different when patients receiving therapy with somatostatin analogs were excluded from analyses. Patients in whom spontaneous tumor slope was analyzed differed for primary location (higher prevalence of non-pancreatic foregut primary; $P<0.01$) and frequency of surgery (higher prevalence of patients who underwent surgery before study entry; $P<0.01$) among parameters listed in Table 2.

Prognostic factors of survival

At the end of the study, 64 (54%) patients died. Deaths were related to tumor progression in 61 (95%) cases. Therefore, overall survival was studied. The 5- and 10-year overall survival rates from the time of detection of metastases were 54% (95% CI 45–63%) and 22% (95% CI 11–37%) respectively (Fig. 1A).

The following parameters were associated with a significant decrease in overall survival in the univariate analysis: age at the time of first complete imaging work-up of metastases discovery, foregut embryological origin, pancreatic primary site, more than 10 liver metastases, tumor-progression slope, increased CGA >5 levels, absence of surgery before study entry and absence of surgery for the primary tumor (Table 2). Owing to the small number of patients with information regarding CGA levels, this parameter was not included in the multivariate analysis.

Table 2 Characteristics of patients with metastatic well-differentiated endocrine carcinoma (WDEC). Univariate and multivariate analysis of overall survival

Variables	Number of patients	Number of deaths	Five-year survival (%)	Univariate analysis			Multivariate analysis		
				Unadjusted HR	95% CI	P ^a	Adjusted HR ^b	95% CI	P ^c
Total	118	66	54						
Gender									
Male	60	39	46						
Female	58	27	63	0.7	0.4–1.1	0.11	–	–	–
Age									
≤65	85	43	60						
>65	33	23	41	1.7	1.1–2.9	0.03	2.8	1.4–5.4	0.002
Foregut ^d									
No	43	18	65						
Yes	58	37	47	1.9	1.1–3.3	0.03	0.9	0.4–2.4	0.9
Midgut ^e									
No	62	38	49						
Yes	39	17	64	0.6	0.3–1.1	0.09	–	–	–
Hindgut ^f									
No	97	54	54						
Yes	4	1	75	0.3	0.1–2.6	0.3	–	–	–
Pulmonary primary									
No	84	46	52						
Yes	17	9	69	0.7	0.3–1.5	0.33	–	–	–
Pancreatic primary									
No	66	32	65						
Yes	35	23	36	2.1	1.2–3.6	0.01	1.6	0.8–3.3	0.18
Ileum primary									
No	68	39	52						
Yes	33	16	62	0.7	0.4–1.2	0.21	–	–	–
Flushing									
No	107	60	52						
Yes	11	6	53	0.8	0.5–1.2	0.22	–	–	–
Diarrhea									
No	100	52	58						
Yes	18	14	37	1.5	0.8–2.7	0.21	–	–	–
Mitotic count ^g									
<2	45	25	56						
2–10	29	12	71	0.7	0.4–1.5	0.41			
>10	8	5	37	1.8	0.7–4.6	0.2	–	–	–
Ki-67 index ^h									
<2	12	7	67						
2–10	29	11	60	0.7	0.6–1.5	0.3			
>10	9	5	44	1.9	0.7–5.4	0.2	–	–	–
CGA level ⁱ									
Normal	28	8	74						
2–4 USR	12	2	82	0.6	0.2–2.3	0.46			
≥5 USR	20	15	30	2.6	1.3–5.0	0.002	–	–	–
5-HIAA									
Normal	51	27	53						
2–4 USR	21	10	62	0.9	0.5–1.5	0.69			
≥5 USR	17	13	46	1.4	1.0–2.1	0.09	–	–	–
Metastasis discovery									
Synchr	96	55	53						
Metachr	22	11	61	0.9	0.8–1.0	0.09	–	–	–
Liver metastases									
No	10	4	67						
Yes	108	62	53	1.8	0.6–4.9	0.3	–	–	–

Table 2 continued

Variables	Number of patients	Number of deaths	Five-year survival (%)	Univariate analysis			Multivariate analysis		
				Unadjusted HR	95% CI	P ^a	Adjusted HR ^b	95% CI	P ^c
Total	118	66	54						
Number of liver metastases	50	20	75						
Lung metastases	35	24	36	2.1	1.2–3.9	0.01	2.4	1.3–4.6	0.005
Bone metastases	109	58	56						
	9	8	22	1.8	0.8–3.8	0.12	–	–	–
Number of organs with metastases	92	49	58						
	23	15	43	1.3	0.7–2.3	0.5	–	–	–
	73	38	57						
Tumoral progression ^l	41	26	51	1.2	0.9–1.7	0.25			
	4	2	50	1.0	0.3–2.6	0.94	–	–	–
Previous surgery ^k	37	14	77						
	29	21	39	2.8	1.4–5.6	0.003	2.9	1.3–6.6	0.01
	66	43	42						
Previous surgery for primary tumor	52	23	69	0.4	0.2–0.7	<0.001	0.4	0.2–0.7	0.002
	72	45	44						
	46	21	70	0.4	0.2–0.7	0.002	0.9	0.2–3.9	0.8

Abbreviations: CGA, Chromogranin A; 5HIAA, 5 hydroxyindolacetic acid; HR, Hazard risk; USR, Upper superior range; Synchr, Synchronous; Metachr, Metachronous.

^aP value univariate.

^bAdjusted HR: multivariate analysis was performed within three steps according to the variables categories which are respectively: clinical variables, the slope progression, and variables related to the treatment. We first computed adjusted risks in a multivariable model including clinical parameters that were significant in the univariate analysis, then in a second step, we added the slope variable to this first model. The third step consisted to generate a multivariate model with treatment variables that were significant in the univariate analysis.

^cP value for the multivariate analysis.

^dForegut included pancreas (n=35), thymus (n=3), larynx (n=1), stomach (n=1), duodenum (n=1) and lung (n=17).

^eMidgut included ileum (n=33), appendix (n=4), and right colon (n=2).

^fHindgut included rectum (n=4).

^gThe mitotic index was determined in 59 (72%) primary or 23 (28%) metastatic tumor specimens, and in 59 (72%) surgical or 23 (28%) biopsy samples.

^hThe Ki-67 score was determined in 33 (66%) primary or 17 (34%) metastatic specimens, and in 38 (76%) surgical or 12 (24%) biopsy samples.

ⁱBecause of the small number of patients with information regarding CGA levels, this parameter was not included in the multivariate analysis.

^jPercentage increase per month after a 3–6 month period of follow-up.

^kPrimary tumor and/or initial metastases surgery, before study entry.

Table 3 Site of metastatic disease according to site of primary tumor

Site of primary GEP WDEC	Location of metastases			
	Liver number (%)	Lung number (%)	Bone number (%)	Other ^a number (%)
Foregut				
Pancreas	35 (100)	2 (6)	8 (23)	4 (12)
Lung	16 (94)	2 (12)	8 (47)	4 (24)
Thymus–larynx	1 (33)	2 (50)	2 (50)	2 (50)
Stomach–duodenal	2 (100)	0	0	0
Midgut				
Ileum	31 (94)	0	2 (6)	9 (27)
Colon–appendix	5 (83)	0	0	3 (59)
Hindgut				
Colon–rectum	4 (100)	0	1 (25)	0
Unknown	14 (82)	3 (18)	2 (12)	4 (24)
Total	108 (65%)	9 (5%)	23 (14%)	26 (16%)

^aOther sites included: peritoneum, adrenal gland, ovary, skin, spleen and cerebellum.

In multivariate analysis (Table 4), an older age at diagnosis of metastases (HR:1.05; 95% CI 1.01–1.08) and the number of liver metastases over 10 (HR:3.4; 95% CI 1.4–8.3) were found to be predictors of decreased survival. When tumor slope and therapeutic modalities given during the course of the disease were introduced in the multivariate analysis, the significance of the two former criteria did not change and two new predictors were found: tumoral progression slope (HR:1.1; 95% CI 1.0–1.1) and initial surgery (HR:0.3; 95% CI 0.1–0.8). No other therapeutic options given during the course of the disease was found to alter the statistical analysis (details given in Table 5).

Using the four prognostic parameters identified in the multivariate analysis (age above 65 years, number of liver metastases above or equal to 10, a progressive tumor slope and absence of previous surgery), a subgroup of 56 patients with all these parameters available was identified. The survival significantly decreased with increasing numbers of risk factors: 100%, 91% (95% CI 51–98%), 65% (95% CI 37–83%), and 9% (95% CI 6–32%) 5-year survival was observed when patients had 0 (10 patients (18%)), 1 (12 patients (21%)), 2 (19 patients (34%)) or ≥3 (15 patients (27%)) poor prognostic features. The risk of death significantly rose moving from patients with one risk factor (HR:2.1; 95% CI:0.9–4.7; $P=0.15$) to patients with two or more risk factors (HR:4.8, 95% CI 1.5–15.3; $P=0.008$) and patients with three risk factors or more (HR:19.2, 95% CI 5.3–70.0; $P<0.0001$; Fig. 1B).

Finally, we analyzed tumor progression as a function of the number of liver metastases, Ki-67 and pancreatic location: tumor progression was observed in 10 out of

32 (18%) subjects with less than 10 liver metastases but in 14 out of 24 (58%) of those with 10 or more liver metastases ($P=0.04$); tumor progression was observed in 9 out of 23 (30%) subjects with Ki-67 less than 10% but in 7 out of 7 (100%) of those with Ki-67 equal or above 10% ($P=0.005$; Fig. 2). No relationship was found with the primary.

Discussion

Until now, very few studies have focused on metastatic WD GEP tumors, and to the best of our knowledge, this is only the second study that includes multivariate analysis in the study of these patients. Previously, Clancy *et al.* (2006) first found age and alkaline phosphatase level, probably as a surrogate marker of liver tumor burden, to be important prognostic parameters in patients with metastatic GEP tumors; gender, primary location, CGA level, and presence of liver metastases were not found to be predictive. Our study includes a comprehensive list of detailed prognostic parameters considered to be important in previous studies including pancreatic primary (Onaitis *et al.* 2000, Panzuto *et al.* 2005), extra-hepatic metastases (Yu *et al.* 1999, Panzuto *et al.* 2005), extent of liver metastases (Weber *et al.* 1995, Lo *et al.* 1996, Yu *et al.* 1999, Chamberlain *et al.* 2000, Solorzano *et al.* 2001), level of serum biological markers (Janson *et al.* 1997, Onaitis *et al.* 2000, Turner *et al.* 2006), presence of a functioning tumor (Pellikka *et al.* 1993), and also the spontaneous tumor burden slope (Madeira *et al.* 1998, Arnold *et al.* 2005).

The main issue regarding the prognostic stratification of GEP tumors relies in the heterogeneity of the study populations. Our aim was to focus the analysis on

Figure 1 (A) Overall, patient survival since metastases diagnosis. Number of patients: 118. Five-year overall survival rate: 54% (95% CI:45–63%). Ten-year overall survival rate: 22% (95% CI:11–37%). (B) Survival rate according to the number of poor prognostic factor.

a homogeneous subgroup of chemotherapy-naïve GEP WDEC patients regarding the stage and differentiation, subsequently treated with classical therapeutic options. All tumors samples and imaging were revised by single experienced specialists (PD, CD). Our analysis carries the limitation of retrospective studies but remains the

most detailed to date in this specific subgroup of patients.

The clinical characteristics of patients with metastatic WDEC warrant comment. The 5-year survival of 54% we report is within the range of that reported by others in referral center-based studies (Weber *et al.* 1995, Janson *et al.* 1997, Madeira *et al.* 1998, Solorzano *et al.* 2001, Panzuto *et al.* 2005, Pape *et al.* 2008), but higher than rates published in cancer registries (Quaadvlieg *et al.* 2001, Modlin *et al.* 2003). Death related to disease progression accounted for nearly all deaths in patients with metastatic GEP tumors in contrast to studies of GEP patients with non-metastatic disease (Greenberg *et al.* 1987, Yu *et al.* 1999, Søreide *et al.* 2000). We evaluated various primary tumor sites because primary location is a weaker prognostic parameter than differentiation and stage. Finally, mainly sporadic GEP WDEC analyzed before any systemic therapeutic intervention were enrolled making the role of mitotic index or tumor slope evaluable. In accordance with previous results, the majority of WDEC patients with metastatic disease had liver metastases of varying extent, followed by bone metastases (Gibril *et al.* 1998, Madeira *et al.* 1998, Hellman *et al.* 2002, Modlin *et al.* 2003, Plöckinger *et al.* 2004, Panzuto *et al.* 2005, Tomassetti *et al.* 2006, Baudin 2007, Pape *et al.* 2008).

Our study identifies age, the number of liver metastases, spontaneous tumor slope, and initial surgery as major predictors for overall survival in patients with metastatic GEP tumors. Advanced age has been found to be related to worse overall survival as well as disease-specific and relative survival in multiple studies (Greenberg *et al.* 1987, Janson *et al.* 1997, Gibril *et al.* 1998, Quaadvlieg *et al.* 2001, Solorzano *et al.* 2001, Zar *et al.* 2004, Asamura *et al.* 2006, Clancy *et al.* 2006, Lepage *et al.* 2007, Rea *et al.* 2007). Our study does not allow for discriminating the putative roles of co-morbid conditions, tolerance to therapy and/or tumor aggressiveness associated to increased age. Our study also suggests a prognostic

Table 4 Predictors for overall survival in patients with metastatic gastroenteropancreatic well-differentiated endocrine carcinoma (GEP WDEC; multivariate analysis)

Variables (characteristics)	Adjusted HR ^a	95% CI	P
Age at diagnosis of metastases ^b	1.05	1.01–1.08	0.01
Number of liver metastases ^c	3.4	1.4–8.3	0.01
Tumoral progression slope (%)	1.1	1.0–1.1	0.001
Previous surgery (no/yes) ^d	0.3	0.1–0.8	0.02

^a Each risk estimate is adjusted for all risk factors shown in the table.

^b Per additional year of age.

^c Less than 10 vs 10 or more liver nodules.

^d Primary tumor and/or initial metastases surgery, before study entry.

Table 5 Therapeutic management of gastroenteropancreatic well-differentiated endocrine carcinoma (GEP WDEC) after study initiation according to the number of poor prognostic factors

Treatments after study initiation ^a	Number of poor prognostic factors			
	0 (n=10)	1 (n=12)	2 (n=19)	≥ 3 (n=15)
Primary tumor surgery	1 (10%)	4 (33%)	6 (32%)	4 (27%)
Liver metastases surgery	3 (30%)	2 (17%)	3 (16%)	3 (20%)
Others metastases surgery	2 (20%)	1 (8%)	2 (11%)	0
Systemic chemotherapy	2 (20%)	5 (42%)	11 (58%)	13 (87%)
Number of chemotherapy regimens				
1	2 (20%)	1 (8%)	5 (26%)	6 (40%)
2	0	2 (17%)	4 (21%)	3 (20%)
≥ 3	0	2 (17%)	2 (11%)	4 (27%)
Chemoembolization	3 (30%)	2 (17%)	10 (53%)	5 (33%)
Radiotherapy	0	3 (25%)	4 (21%)	2 (13%)
Somatostatin analogs	6 (60%)	8 (67%)	15 (79%)	11 (73%)
Interferon	0	1 (8%)	2 (11%)	4 (27%)
Peptide receptor radionuclide therapy	0	0	0	0
None	0	0	1 (5%)	0
Number of treatments (%) ^b	17 (12%)	26 (19%)	53 (38%)	42 (31%)

^a The number of treatments has been evaluated in the subgroup of 56 patients in whom all the prognostic parameters identified in the multivariate analysis were available (age at diagnosis of metastases; number of liver metastases; tumoral progression slope; previous surgery).

^b Percentage over the total number of treatments performed after the study initiation.

role of the number of liver metastases at diagnosis in contrast to either metastatic disease at other sites or the number of organs with metastatic involvement. The less significant prognostic effect of bone metastases, also evidenced by the low number of bone-related morbid events, may be explained by increased bone mineral density associated with these lesions. Interestingly, CGA, a well-known surrogate marker of both tumor burden and/or functional activity of endocrine tumors (Baudin *et al.* 1998), was found to positively correlate with overall survival in univariate analysis and remains to be evaluated in multivariate analysis.

Figure 2 Ki-67 index as a function of tumor slope, defined by the percentage increase per month after a 3–6 month period of follow-up (correlation coefficient 43%, $P=0.016$).

We demonstrate an independent statistical correlation between tumor slope before and survival of metastatic GEP tumors. Indeed, in our study, this parameter was found to better reflect tumor aggressiveness than disease-free interval or proliferative index. Interestingly, stable disease without aggressive therapy was observed in 56% of GEP tumors over a 6 month time frame. Previous studies report the occurrence of spontaneous disease stabilization ranging from 13% of 31 and 26% of 65 evaluable patients (Arnold *et al.* 1996, Madeira *et al.* 1998, Clancy *et al.* 2006). Since tumor slope assessment is not yet standardized in the field of endocrine tumors, a comparison with previous studies is difficult. Our results suggest that tumor slope assessment improves prognostic classification of GEP tumors. However, it should be stressed that the period of time required for slope assessment delayed the prognostic classification and was frequently considered stressful by patients. Some other limitations of tumor slope assessment in our study should be mentioned. First, tumor slope was assessed in 56% of total patients, among which a higher frequency of midgut GEP tumors and previous surgery was noted. Second, somatostatin analog therapy was given to 28% of these patients who complained of symptoms attributed to functioning tumor. Finally, caution should be exercised when treating patients with high tumor burden, (e.g. more than 10 liver metastases), or high Ki-67 index (above or equal to 10%). Indeed, 58 or 100% of these

subgroups of patients were found to have progressive disease. In our series, previous surgery but not other therapies given during the course of the disease was associated with a favorable outcome as similarly reported by others (Lo *et al.* 1996, Madeira *et al.* 1998, Solorzano *et al.* 2001, Asamura *et al.* 2006). However, the design of our study preclude definite conclusion regarding the influence of therapies given during the course of our study on our results even if, major disequilibrium between each therapeutic option was not observed between prognostic subgroup (Table 5).

Several parameters analyzed in this study deserve further comment. First, the primary location of the tumor was not found to be a major prognostic factor in metastatic WD GEP tumors even if pancreatic location was associated with a poorer outcome at univariate analysis. Second, our study does not confirm a main role of mitotic index to lead the therapeutic strategy in metastatic GEP tumors as already found by others (Burke *et al.* 1997, Van Eeden *et al.* 2002, Panzuto *et al.* 2005), but not all (Pape *et al.* 2008). Remarkably, our results suggest a good correlation between positive slope and Ki-67 equal or above 10% but heterogeneous results in case of Ki-67 below 10%. Both tissue heterogeneity, cut-off levels of mitotic index used in this study but also various impact of mitotic index as a function of primary location may explain this result in stage IV GEP WDEC. Third, presence of a functioning WDEC should no more be considered as a major prognostic indicator thanks to progress in control of functioning symptoms including somatostatin analog therapy. Association of a GEP tumor as part of a MEN1 could not be evaluated in our study.

By combining the prognostic indicators described in our study, different outcome can be predicted. For patients with no risk factor, a 'wait-and-see' policy or low-toxicity profile therapy should be scheduled. On the other hand, among the 25% of our study population expressed at least three poor prognostic factors, 5-year disease-specific survival was only 9% (95% CI 0.6–33%), which suggests the use of early therapeutic approaches. Finally, in case of one negative prognostic feature (which account for 39% of our study population), or two negative prognostic features (which account for 36% of our study population), the 5-year disease-specific survival was 95% (95% CI 71–99%) or 62% 5-year survival (95% CI 35–80%) respectively additional studies are required. However, we suggest close monitoring of patients with more than 10 liver metastases or Ki-67 above 10%.

In conclusion, our study highlights prognostic indicators associated with survival in patients with

metastatic WDEC. Number of liver metastases together with age and tumor progression slope may serve to divide what is a heterogeneous population of patients into more homogeneous subgroups to whom therapeutic interventions, including therapeutic trials, might be tailored.

Declaration of interest

The authors have no conflict of interest to disclose.

Funding

This research was supported (in part) by the Intramural Research Program of the NIH and the National Cancer Institute.

Acknowledgements

The authors thank Diane Prestone for reviewing and constructive remarks. Houda Boukheris had a fellowship from the International Agency for Research on Cancer (IARC). Present address: National Cancer Institute, Division of Cancer Epidemiology and Genetics, Radiation Epidemiology Branch, Bethesda, MD, USA.

References

- Aparicio T, Ducreux M, Baudin E, Sabourin JC, De Baere T, Mitry E, Schlumberger M & Rougier P 2001 Antitumor activity of somatostatin analogues in progressive metastatic neuroendocrine tumours. *European Journal of Cancer* **37** 1014–1019.
- Arnold R, Trautmann ME, Creutzfeldt W, Benning R, Benning M, Neuhaus C, Jürgensen R, Stein K, Schäfer H, Bruns C *et al.* 1996 Somatostatin analogue octreotide and inhibition of tumour growth in metastatic endocrine gastroenteropancreatic tumours. *Gut* **38** 430–438.
- Arnold R, Rinke A, Klose KJ, Müller HH, Wied M, Zamzow K, Schmidt C, Schade-Brittinger C, Barth P, Moll R *et al.* 2005 Octreotide versus octreotide plus interferon-alpha in endocrine gastroenteropancreatic tumors: a randomized trial. *Clinical Gastroenterology and Hepatology* **3** 761–771.
- Asamura H, Kameya T, Matsuno Y, Noguchi M, Tada H, Ishikawa Y, Yokose T, Jiang SX, Inoue T, Nakagawa K *et al.* 2006 Neuroendocrine neoplasms of the lung: a prognostic spectrum. *Journal of Clinical Oncology* **24** 70–76.
- Baudin E 2007 Gastroenteropancreatic endocrine tumors: clinical characterization before therapy. *Nature Clinical Practice. Endocrinology and Metabolism* **3** 228–239.
- Baudin E, Gigliotti A, Ducreux M, Ropers J, Comoy E, Sabourin JC, Bidart JM, Cailleux AF, Bonacci R, Ruffié P *et al.* 1998 Neuron-specific enolase and chromogranin A as markers of neuroendocrine tumours. *British Journal of Cancer* **78** 1102–1107.

- Baudin E, Bidart JM, Rougier P, Lazar V, Ruffié P, Ropers J, Ducreux M, Troalen F, Sabourin JC, Comoy E *et al.* 1999 Screening for multiple endocrine neoplasia type 1 and hormonal production in apparently sporadic neuroendocrine tumors. *The Journal of Clinical Endocrinology and Metabolism* **84** 69–75.
- Beasley MB, Thunnissen FB, Brambilla E, Hasleton P, Steele R, Hammar SP, Colby TV, Sheppard M, Shimosato Y, Koss MN *et al.* 2000 Pulmonary atypical carcinoid: predictors of survival in 106 cases. *Human Pathology* **31** 1255–1265.
- Bettini R, Boninsegna L, Mantovani W, Capelli P, Bassi C, Pederzoli P, Delle Fave GF, Panzuto F, Scarpa A & Falconi M 2008 Prognostic factors at diagnosis and value of WHO classification in a mono-institutional series of 180 non-functioning pancreatic endocrine tumours. *Annals of Oncology* **19** 903–908.
- Burke AP, Thomas RM, Elsayed AM & Sobin LH 1997 Carcinoids of the jejunum and ileum: an immunohistochemical and clinicopathologic study of 167 cases. *Cancer* **79** 1086–1093.
- Cardillo G, Sera F, Di Martino M, Graziano P, Giunti R, Carbone L, Facciolo F & Martelli M 2004 Bronchial carcinoid tumors: nodal status and long-term survival after resection. *Annals of Thoracic Surgery* **77** 1781–1785.
- Chamberlain RS, Canes D, Brown KT, Saltz L, Jarnagin W, Fong Y & Blumgart LH 2000 Hepatic neuroendocrine metastases: does intervention alter outcomes? *Journal of the American College of Surgeons* **190** 432–445.
- Chaudhry A, Oberg K & Wilander E 1992 A study of biological behavior based on the expression of a proliferating antigen in neuroendocrine tumors of the digestive system. *Tumour Biology* **13** 27–35.
- Clancy TE, Sengupta TP, Paulus J, Ahmed F, Duh MS & Kulke MH 2006 Alkaline phosphatase predicts survival in patients with metastatic neuroendocrine tumors. *Digestive Diseases and Sciences* **51** 877–884.
- Cox DR 1972 Regression models and life-tables. *Journal of the Royal Statistical Society, Series B* **34** 187–220.
- Delaunoy T, Ducreux M, Boige V, Dromain C, Sabourin JC, Duvillard P, Schlumberger M, de Baere T, Rougier P, Ruffie P *et al.* 2004 The doxorubicin–streptozotocin combination for the treatment of advanced well-differentiated pancreatic endocrine carcinoma: a judicious option? *European Journal of Cancer* **40** 515–520.
- Dromain C, de Baere T, Lumbroso J, Caillet H, Laplanche A, Boige V, Ducreux M, Duvillard P, Elias D, Schlumberger M *et al.* 2005 Detection of liver metastases from endocrine tumors: a prospective comparison of somatostatin receptor scintigraphy, computed tomography, and magnetic resonance imaging. *Journal of Clinical Oncology* **23** 70–78.
- Van Eeden S, Quaedyvlieg PF, Taal BG, Offerhaus GJ, Lamers CB & Van Velthuysen ML 2002 Classification of low-grade neuroendocrine tumors of midgut and unknown origin. *Human Pathology* **33** 1126–1132.
- Elias D, Lasser P, Ducreux M, Duvillard P, Ouellet JF, Dromain C, Schlumberger M, Pocard M, Boige V, Miquel C *et al.* 2003 Liver resection (and associated extrahepatic resections) for metastatic well-differentiated endocrine tumors: a 15-year single center prospective study. *Surgery* **133** 375–382.
- Faggiano A, Sabourin JC, Ducreux M, Lumbroso J, Duvillard P, Leboulleux S, Dromain C, Colao A, Schlumberger M & Baudin E 2007 Pulmonary and extrapulmonary poorly differentiated large cell neuroendocrine carcinomas: diagnostic and prognostic features. *Cancer* **110** 265–274.
- Fischer L, Kleeff J, Esposito I, Hinz U, Zimmermann A, Friess H & Büchler MW 2008 Clinical outcome and long-term survival in 118 consecutive patients with neuroendocrine tumours of the pancreas. *British Journal of Surgery* **95** 627–635.
- García-Yuste M, Matilla JM, Cueto A, Paniagua JM, Ramos G, Cañizares MA & Muguruza I 2007 Typical and atypical carcinoid tumours: analysis of the experience of the Spanish multi-centric study of neuroendocrine tumours of the lung. *European Journal of Cardio-Thoracic Surgery* **31** 192–197.
- Gibril F, Doppman JL, Reynolds JC, Chen CC, Sutliff VE, Yu F, Serrano J, Venzon DJ & Jensen RT 1998 Bone metastases in patients with gastrinomas: a prospective study of bone scanning, somatostatin receptor scanning, and magnetic resonance image in their detection, frequency, location, and effect of their detection on management. *Journal of Clinical Oncology* **16** 1040–1053.
- Greenberg RS, Baumgarten DA, Clark WS, Isacson P & McKeen K 1987 Prognostic factors for gastrointestinal and bronchopulmonary carcinoid tumors. *Cancer* **60** 2476–2483.
- Heitz PhU, Komminoth P, Perren A, Klimstra DS, Dayal Y, Bordi C, Lechagon J, Centeno BA & Klöppel G 2004 *Tumours of Endocrine Organs. World Health Organisation Classification of Tumors*. pp 175–208. Lyon: IARC Press.
- Hellman P, Lundström T, Ohrvall U, Eriksson B, Skogseid B, Oberg K, Tiensuu Janson E & Akerström G 2002 Effect of surgery on the outcome of midgut carcinoid disease with lymph node and liver metastases. *World Journal of Surgery* **26** 991–997.
- Hochwald SN, Zee S, Conlon KC, Colleoni R, Louie O, Brennan MF & Klimstra DS 2002 Prognostic factors in pancreatic endocrine neoplasms: an analysis of 136 cases with a proposal for low-grade and intermediate-grade groups. *Journal of Clinical Oncology* **20** 2633–2642.
- Janson ET, Holmberg L, Stridsberg M, Eriksson B, Theodorsson E, Wilander E & Oberg K 1997 Carcinoid tumors: analysis of prognostic factors and survival in 301 patients from a referral center. *Annals of Oncology* **8** 685–690.

- Johnson LA, Lavin PT, Moertel CG, Weiland LH, Dayal YY, Doos WG, Geller SA, Cooper HS, Masse SR & Engstrom PF 1986 Carcinoids: the prognostic effect of primary site histologic type variations. *Journal of Surgical Oncology* **33** 81–83.
- Kaplan EL & Meier P 1958 Non parametric estimation from incomplete observations. *Journal of the American Statistical Association* **53** 457–481.
- Kirshbom PM, Kherani AR, Onaitis MW, Hata A, Kehoe TE, Feldman C, Feldman JM & Tyler DS 1999 Foregut carcinoids: a clinical and biochemical analysis. *Surgery* **126** 1105–1110.
- Lee ET 1986 *Statistical Methods for Survival Data Analysis*. Belmont, CA: Wadsworth.
- Lepage C, Rachet B & Coleman MP 2007 Survival from malignant digestive endocrine tumors in England and Wales: a population-based study. *Gastroenterology* **132** 899–904.
- Lim E, Yap YK, De Stavola BL, Nicholson AG & Goldstraw P 2005 The impact of stage and cell type on the prognosis of pulmonary neuroendocrine tumors. *Journal of Thoracic and Cardiovascular Surgery* **130** 969–972.
- Lo CY, van Heerden JA, Thompson GB, Grant CS, Söreide JA & Harmsen WS 1996 Islet cell carcinoma of the pancreas. *World Journal of Surgery* **20** 878–883.
- Madeira I, Terris B, Voss M, Denys A, Sauvanet A, Flejou JF, Vilgrain V, Belghiti J, Bernades P & Ruszniewski P 1998 Prognostic factors in patients with endocrine tumours of the duodenopancreatic area. *Gut* **43** 422–427.
- Mitry E, Baudin E, Ducreux M, Sabourin JC, Rufé P, Aparicio T, Aparicio T, Lasser P, Elias D, Duvallard P et al. 1999 Treatment of poorly differentiated neuroendocrine tumours with etoposide and cisplatin. *British Journal of Cancer* **81** 1351–1355.
- Modlin IM, Lye KD & Kidd M 2003 A 5-decade analysis of 13 715 carcinoid tumors. *Cancer* **97** 934–959.
- Onaitis MW, Kirshbom PM, Hayward TZ, Quayle FJ, Feldman JM, Seigler HF & Tyler DS 2000 Gastrointestinal carcinoids: characterization by site of origin and hormone production. *Annals of Surgery* **232** 549–556.
- Panzuto F, Nasoni S, Falconi M, Corleto VD, Capurso G, Cassetta S, Di Fonzo M, Tornatore V, Milione M, Angeletti S et al. 2005 Prognostic factors and survival in endocrine tumor patients: comparison between gastrointestinal and pancreatic localization. *Endocrine-Related Cancer* **12** 1083–1092.
- Pape UF, Jann H, Müller-Nordhorn J, Bockelbrink A, Berndt U, Willich SN, Koch M, Röcken C, Rindi G & Wiedenmann B 2008 Prognostic relevance of a novel TNM classification system for upper gastroenteropancreatic neuroendocrine tumors. *Cancer* **113** 256–265.
- Pellikka PA, Tajik AJ, Khandheria BK, Seward JB, Callahan JA, Pitot HC & Kvols LK 1993 Carcinoid heart disease. Clinical and echocardiographic spectrum in 74 patients. *Circulation* **87** 1188–1196.
- Pelosi G, Bresaola E, Bogina G, Pasini F, Rodella S, Castelli P, Iacono C, Serio G & Zamboni G 1996 Endocrine tumors of the pancreas: Ki-67 immunoreactivity on paraffin sections is an independent predictor for malignancy: a comparative study with proliferating-cell nuclear antigen and progesterone receptor protein immunostaining, mitotic index, and other clinicopathologic variables. *Human Pathology* **27** 1124–1134.
- Plöckinger U, Rindi G, Arnold R, Eriksson B, Krenning EP, de Herder WW, Goede A, Caplin M, Oberg K, Reubi JC et al. 2004 Guidelines for the diagnosis and treatment of neuroendocrine gastrointestinal tumours. *Neuroendocrinology* **80** 394–424.
- Quaedyvlieg PF, Visser O, Lamers CB, Janssen-Heijnen ML & Taal BG 2001 Epidemiology and survival in patients with carcinoid disease in the Netherlands: an epidemiological study with 2391 patients. *Annals of Oncology* **12** 1295–1300.
- Rea F, Rizzardi G, Zuin A, Marulli G, Nicotra S, Bulf R, Schiavon M & Sartori F 2007 Outcome and surgical strategy in bronchial carcinoid tumors: single institution experience with 252 patients. *European Journal of Cardio-Thoracic Surgery* **31** 186–191.
- Rigaud G, Missiaglia E, Moore PS, Zamboni G, Falconi M, Talamini G, Pesci A, Baron A, Lissandrini D, Rindi G et al. 2001 High resolution allelotyping of nonfunctional pancreatic endocrine tumors: identification of two molecular subgroups with clinical implications. *Cancer* **61** 285–292.
- Rindi G, Azzoni C, La Rosa S, Klersy C, Paolotti D, Rappel S, Stolte M, Capella C, Bordi C & Solcia E 1999 ECL cell tumor and poorly differentiated endocrine carcinoma of the stomach: prognostic evaluation by pathological analysis. *Gastroenterology* **116** 532–542.
- Rindi G, Klöppel G, Alhman H, Caplin M, Couvelard A, de Herder WW, Eriksson B, Falchetti A, Falconi M, Komminoth P et al. 2006 TNM staging of foregut (neuro)endocrine tumors: a consensus proposal including a grading system. *Virchows Archiv* **449** 395–401.
- Rindi G, Klöppel G, Couvelard A, Komminoth P, Körner M, Lopes JM, McNicol AM, Nilsson O, Perren A, Scarpa A et al. 2007 TNM staging of midgut and hindgut (neuro) endocrine tumors: a consensus proposal including a grading system. *Virchows Archiv* **451** 757–762.
- Roche A, Girish BV, de Baère T, Baudin E, Boige V, Elias D, Lasser P, Schlumberger M, Ducreux M et al. 2003 Transcatheter arterial chemoembolization as first-line treatment for hepatic metastases from endocrine tumors. *European Radiology* **13** 136–140.
- Roche A, Girish BV, de Baere T, Ducreux M, Elias D, Laplanche A, Boige V, Schlumberger M, Ruffe P & Baudin E 2004 Prognostic factors for chemoembolization in liver metastasis from endocrine tumors. *Hepato-gastroenterology* **51** 1751–1756.

- Shebani KO, Souba WW, Finkelstein DM, Stark PC, Elgadi KM, Tanabe KK & Ott MJ 1999 Prognosis and survival in patients with gastrointestinal tract carcinoid tumors. *Annals of Surgery* **229** 815–821.
- Solcia E, Klöppel G & Sobin LH 2000 Histological typing of endocrine tumors. In *World Health Organisation International Histological Classification of Tumors*, edn 2, pp 1–149. New York: Springer.
- Solorzano CC, Lee JE, Pisters PW, Vauthey JN, Ayers GD, Jean ME, Gagel RF, Ajani JA, Wolff RA & Evans DB 2001 Nonfunctioning islet cell carcinoma of the pancreas: survival results in a contemporary series of 163 patients. *Surgery* **130** 1078–1085.
- Söreide JA, van Heerden JA, Thompson GB, Schleck C, Ilstrup DM & Churchward M 2000 Gastrointestinal carcinoid tumors: long-term prognosis for surgically treated patients. *World Journal of Surgery* **24** 1431–1436.
- Therasse P, Arbuck SG, Eisenhauer EA, Wanders J, Kaplan RS, Rubinstein L, Verweij J, Van Glabbeke M, van Oosterom AT, Christian MC *et al.* 2000 New guidelines to evaluate the response to treatment in solid tumors. European Organization for Research and Treatment of Cancer, National Cancer Institute of the United States, National Cancer Institute of Canada. *Journal of the National Cancer Institute* **92** 205–216.
- Tomassetti P, Campana D, Piscitelli L, Casadei R, Santini D, Nori F, Morselli-Labate AM, Pezzilli R & Corinaldesi R 2005 Endocrine pancreatic tumors: factors correlated with survival. *Annals of Oncology* **16** 1806–1810.
- Tomassetti P, Campana D & Piscitelli L 2006 Endocrine tumors of the ileum: factors correlated with survival. *Neuroendocrinology* **83** 380–386.
- Travis WD, Rush W, Flieder DB, Falk R, Fleming MV, Gal AA & Koss MN 1998 Survival analysis of 200 pulmonary neuroendocrine tumors with clarification of criteria for atypical carcinoid and its separation from typical carcinoid. *American Journal of Surgical Pathology* **22** 934–944.
- Travis WD, Brambilla E, Müller-Hermelink CC & Harris CC 2004 *Pathology of the Lung, Pleura, Thymus and Heart*. pp 97–98. Lyon: IARC Press.
- Turner GB, Johnston BT, McCance DR, McGinty A, Watson RG, Patterson CC & Ardill JE 2006 Circulating markers of prognosis and response to treatment in patients with midgut carcinoid tumours. *Gut* **55** 1586–1591.
- Weber HC, Venzon DJ, Lin JT, Fishbein VA, Orbuch M, Strader DB, Gibril F, Metz DC, Fraker DL & Norton JA 1995 Determinants of metastatic rate and survival in patients with Zollinger–Ellison syndrome: a prospective long-term study. *Gastroenterology* **108** 1637–1649.
- Yu F, Venzon DJ, Serrano J, Goebel SU, Doppman JL, Gibril F & Jensen RT 1999 Prospective study of the clinical course, prognostic factors, cause of death, and survival in patients with long-standing Zollinger–Ellison syndrome. *Journal of Clinical Oncology* **17** 615–630.
- Zar N, Garmo H, Holmberg L, Rastad J & Hellman P 2004 Long-term survival of patients with small intestinal carcinoid tumors. *World Journal of Surgery* **28** 1163–1168.