Peritoneal carcinomatosis from small intestinal neuroendocrine tumors: Clinical course and genetic profiling

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Background. *One-fifth of all patients with small-intestinal neuroendocrine tumors (SI-NETs) present with or develop peritoneal carcinomatosis (PC). Our aim was to determine the prognosis and genetic profiles of tumors in patients with PC compared with tumors in patients without PC.*
Methods. *We included SI-NET patients (cases with PC,* n = *73, and controls without PC,* n = *468) who underwent operation between 1985 and 2012. The Lyon prognostic index was used to correlate the amount of PC to survival. DNA samples from patients with (*n = *8) and without (*n = *7) PC were analyzed with a single-nucleotide polymorphism array (HumanOmni2.5 BeadChip, Illumina) to investigate genetic disparities between groups.*
Results. *Patients with PC had poorer survival (median 5.1 years) than controls (11.1 years). An advanced postoperative Lyon prognostic index was a negative prognostic marker for survival by multivariable analysis (*P = *.042). Patients with and without PC clustered differently based on loss of heterozygosity and copy number variation data from single-nucleotide polymorphism array of the primary tumors (*P = *.042).*
Conclusion. *SI-NET patients with PC have poor survival, which diminishes with increasing PC load after surgery. Clustering based on copy number variation and loss of heterozygosity data suggests different genotypes in primary tumors comparing patients with and without PC. (Surgery 2014;156:1512-22.)*

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The clinical incidence of small intestinal neuroendocrine tumors (SI-NETs) is increasing according to most cancer registries, the last decade ranging from 0.3 to 1.33/100,000.[1,2] The majority of patients with SI-NETs present with distant metastases, and the third most common path of dissemination after lymph node and liver metastases is peritoneal carcinomatosis (PC), which is reported in 19–33% of patients with SI-NET at specialized centers.[3,4]

In 2007, the European Neuroendocrine Tumor Society (ENETS) presented a consensus document about PC in gastroenteropancreatic neuroendocrine tumors (GEP-NETs).[5] The document concluded that data on PC in GEP-NETs are scarce and it also proposed that the Lyon prognostic index may be used to retrospectively classify PC. The Lyon prognostic index comprises 4 stages, ranging from small, localized nodules, to larger and diffusely spread nodules in the peritoneum (Table I). A supplemental system, global peritoneal score (GPS; Table II), which is used to assess the extent of liver and lymph node metastases, also was suggested by ENETS, because NET patients with PC often display synchronous spread to these organs.[5]

Cytoreductive surgery with peritonectomy (CRS) with the aim to remove all visible tumor, in combination with heated intraperitoneal chemotherapy (HIPEC), has been tried for SI-NET tumors in select centers.[6] One fear regarding the use of CRS and HIPEC in patients with NET has been the lack of knowledge concerning effective chemotherapy for NET with low proliferation.[6] Therefore, since 2008, Elias et al[6] ceased administering HIPEC and continued with CRS only; they reported promising 5-year overall survival of 69%. A majority of patients, however, experienced recurrence during follow-up, and no studies have yet thoroughly investigated the natural course of

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Table I. Lyon prognostic index to classify the extent of PC

Stage 0	No macroscopic disease
Stage I	Localized nodules in one part of the abdomen <5 mm in size
Stage II	Diffuse nodules spread to the whole abdomen <5 mm in size
Stage III	Localized or diffuse nodules 5–20 mm in size
Stage IV	Localized or diffuse nodules or masses >20 mm in size

PC, Peritoneal carcinomatosis.

SI-NET patients with PC exempted to CRS,[6] although a study from our own institution has showed that PC is a negative prognostic factor in these patients.[7]

Because PC is an indicator of a more advanced or more aggressive disease in patients with SI-NET, we hypothesized that the genetic profiles of tumors from patients with PC may be disparate from the tumors from patients without PC. Genome-wide loss of heterozygosity (LOH) screening with microsatellite markers the use of comparative genomic hybridization (CGH) arrays and single-nucleotide polymorphism (SNP) arrays have shown copy number (CN) loss and LOH of chromosome 18 in most primary tumors.[8-12] No studies have been performed with the primary aim to investigate genetic aberrations in tumor specimens from SI-NET patients with PC in contrast to those without PC.

Although the former study from our institution briefly mentions PC as a risk factor for death[7] this work focuses on the subset SI-NET patients that have PC to highlight specific risks and management concerns in these patients. Our first aim was to use the Lyon prognostic index and the GPS to demonstrate survival rates with these classifications. Our second aim, in light of the conceivably more aggressive disease in SI-NET patients with PC, to investigate plausible genetic differences in tumor specimens of patients with and without PC using a high-resolution SNP array for LOH and copy number variation (CNV) analysis.

PATIENTS AND METHODS

We included patients (cases with PC, $n = 73$, and controls without PC, $n = 468$) from a cohort of 672 consecutive patients with SI-NET who were diagnosed and admitted between 1985 and 2012 to the Departments of Surgery or Oncologic Endocrinology at Uppsala University Hospital (Fig 1). All patients had a histopathologic diagnosis of SI-NET based on microscopy and immunohistochemic staining of either liver biopsy material or operative specimens. In tumor specimens retrieved after 1997, the Ki-67 antigen was immunostained routinely at the pathology department, after which a number of cells in tumor hotspots were counted and the Ki-67 index given as a percentage of positive cells. Proliferation grade was classified with Ki-67 index; grade 1 = <3%, grade 2 = 3–20%, and grade 3 = >20%, as described by Klimstra et al.[13] Patients with a known proliferation rate exceeding a Ki-67 index of 20% at baseline (Grade 3 tumors; neuroendocrine carcinomas) and non-Swedish residents were not included (due to follow-up issues). Our rationale not to include grade 3 tumors is that these tumors generally have a different clinical course and also different treatment and they are thus commonly treated and studied as a separate entity to G1-2 tumors. Of the 574 patients who were subjected to laparotomy, 106 displayed signs of PC according to the operative reports from the first laparotomy. PC was defined as spread of separate tumor deposits within the peritoneal cavity away from the primary tumor (excluding mesenteric lymph node metastases). Biopsies or resections of PC were performed in 73 of these 106 patients, and PC was confirmed by histopathology in all 73 cases. The 468 patients who did not show PC at the first laparotomy were included as a control group, thus enabling us to compare tumor dissemination, survival, and reoperative procedure rates with SI-NET patients without PC (Fig 1). Of the 468 patients without PC at first laparotomy, 16 developed PC during follow-up.

Overall follow-up encompassed 7.3 ± 5.4 (mean ± SD) years, and these patients were followed until death or their last follow-up at the Departments of Surgery or Oncologic Endocrinology (until 1 January 2012). Causes of death were collected from the Swedish National Board of Health and Welfare (causes of death registry) and corroborated with available clinical data.

Assessment of tumor burden. We used the ENETS abdominal global PC score (GPS) to quantify the tumor burden for lymph node metastases, liver metastases, and PC (Table I).[5] The presence of mesenteric lymph node metastases and PC was assessed by retrospective examination of operative reports. Para-aortal lymph node metastases, extra-abdominal lymph node metastases, and liver metastases were assed with radiology. Lymph nodes were deemed pathologic if enlarged (largest diameter >1 cm), round-shaped, and accumulating intravenous contrast similar to the primary tumor