The presence of lymph node metastases or extrahepatic metastases; the depth of invasion; the rapid rate of growth; various histologic features (differentiation, mitotic rates, growth indices, vessel density, vascular endothelial growth factor [VEGF], and CD10 metalloproteinase expression); necrosis; presence of cytokeratin; elevated serum alkaline phosphatase levels; older age; presence of circulating tumor cells; and flow cytometric results, such as the presence of aneuploidy, are all important prognostic factors for the development of metastatic disease (Table 113-5). For patients with GI-NETs (carcinoids), additional associations with a worse prognosis include the development of the carcinoid syndrome (especially the development of carcinoid heart disease), male sex, the presence of a symptomatic tumor or greater increases in a number of tumor markers (5-hydroxyindolacetic acid [5-HIAA], neuropeptide K, chromogranin A), and the presence of various molecular features. With pNETs or gastrinomas, a worse prognosis is associated with female sex, overexpression of the Ha-*ras* oncogene or p53, the absence of multiple endocrine neoplasia type 1 (MEN 1), higher levels of various tumor markers (i.e., chromogranin A, gastrin), and presence of various histologic features (immunohistochemistry for c-KIT, low cyclin B1, loss of PTEN/TSC-2, expression of fibroblast growth factor-13) and various molecular features (Table 113-5). The TNM classification systems and the grading systems (G1–G3) have important prognostic value.

A number of diseases due to various genetic disorders are associated with an increased incidence of NETs **(Table 113-6)**. Each one is caused by a loss of a possible tumor-suppressor gene. The most important is MEN 1, which is an autosomal dominant disorder due to a defect in a 10-exon gene on 11q13, which encodes for a 610-amino-acid nuclear protein, menin **(Chap. 408)**. Patients with MEN 1 develop hyperparathyroidism due to parathyroid hyperplasia in 95–100% of cases, pNETs in 80–100%, pituitary adenomas in 54–80%, adrenal adenomas in 27–36%, bronchial carcinoids in 8%, thymic carcinoids in 8%, gastric carcinoids in 13–30% of patients with Zollinger-Ellison syndrome, skin tumors (angiofibromas [88%], collagenomas [72%]), central nervous system (CNS) tumors (meningiomas [<8%]), and smooth-muscle tumors (leiomyomas, leiomyosarcomas [1–7%]). Among patients with MEN 1, 80–100% develop nonfunctional pNETs (most are microscopic with 0–13% large/symptomatic), and functional pNETs occur in 20–80% in different series, with a mean of 54% developing Zollinger-Ellison syndrome, 18% insulinomas, 3% glucagonomas, 3% VIPomas, and <1% GRFomas or somatostatinomas. MEN 1 is present in 20–25% of all patients with Zollinger-Ellison syndrome, 4% of patients with insulinomas, and a low percentage (<5%) of patients with other pNETs.

Three phacomatoses associated with NETs are von Hippel–Lindau disease (VHL), von Recklinghausen's disease (neurofibromatosis type 1 [NF-1]), and tuberous sclerosis (Bourneville's disease) (Table 113-6). VHL is an autosomal dominant disorder due to defects on chromosome 3p25, which encodes for a 213-amino-acid protein that interacts with the elongin family of proteins as a transcriptional regulator **(Chaps. 118, 339, 407, and 408)**. In addition to cerebellar hemangioblastomas, renal cancer, and pheochromocytomas, 10–17% develop a pNET. Most are nonfunctional, although insulinomas and VIPomas have been reported. Patients with NF-1 (von Recklinghausen's disease) have defects in a gene on chromosome 17q11.2 that encodes for a 2845-amino-acid protein, neurofibromin, which functions in normal cells as a suppressor of the *ras* signaling cascade **(Chap. 118)**. Up to 10% of these patients develop an upper GI-NET (carcinoid), characteristically in the periampullary region (54%). Many are classified as somatostatinomas because they contain somatostatin immunocytochemically; however, they uncommonly secrete somatostatin and rarely produce a clinical somatostatinoma syndrome. NF-1 has rarely been associated with insulinomas and Zollinger-Ellison syndrome. NF-1 accounts for 48% of all duodenal somatostastinomas and 23% of all ampullary GI-NETs (carcinoids). Tuberous sclerosis is caused by mutations that alter either the 1164-amino-acid protein hamartin (TSC1) or the 1807-amino-acid protein tuberin (TSC2) **(Chap. 118)**. Both hamartin and tuberin interact in a pathway related to phosphatidylinositol 3-kinases and mammalian target of rapamycin (mTOR) signaling cascades. A few cases including nonfunctional and functional pNETs (insulinomas and gastrinomas) have been reported in these patients (Table 113-6). Mahvash disease is associated with the development of α-cell hyperplasia, hyperglucagonemia, and the development of NF pNETs and is due to a homozygous P86S mutation of the human glucagon receptor.

Mutations in common oncogenes (*ras, myc, fos, src, jun*) or common tumor-suppressor genes (*p53*, retinoblastoma susceptibility gene) are not commonly found in either pNETs or GI-NETs (carcinoids) (Table 113-1). However, frequent (70%) gene amplifications in *MDM2, MDM4,* and *WIPI* inactivating the p53 pathway are noted in well-differentiated pNETs, and the retinoblastoma pathway is altered in the majority of pNETs. In addition to these genes, additional alterations that may be important in their pathogenesis include changes in the *MEN1* gene, *p16/MTS1* tumor-suppressor gene, and *DPC4/Smad4* gene; amplification of the HER-2/*neu* protooncogene; alterations in transcription factors (Hoxc6 [GI carcinoids]), growth factors, and their receptors; methylation of a number of genes that probably results in their inactivation; and deletions of unknown tumor-suppressor genes as well as gains in other unknown genes (Table 113-1). The clinical antitumor activity of everolimus, an mTOR inhibitor, and sunitinib, a tyrosine kinase inhibitor (PDGFR, VEGFR1, VEGFR2, c-KIT, FLT-3), support the importance of the mTOR-AKT pathway and tyrosine kinase receptors in mediating growth of malignant NETs (especially pNETs). The importance of the mTOR pathway in pNET growth is further supported by the finding that a single-nucleotide polymorphism (FGFR4-G388R, in fibroblast growth factor receptor 4) affects selectivity to the mTOR inhibitor and can result in significantly higher risk of advanced pNET stage and liver metastases (Table 113-5). Comparative genomic hybridization, genome-wide allelotyping studies, and genome-wide single-nucleotide polymorphism analyses have shown that chromosomal losses and gains are common in pNETs and GI-NETs (carcinoids), but they differ between these two NETs, and some have prognostic significance (Table 113-5). Mutations in the *MEN1* gene are probably particularly important. Loss of heterozygosity at the MEN 1 locus on chromosome 11q13 is noted in 93% of sporadic pNETs (i.e., in patients without MEN 1) and in 26–75% of sporadic GI-NETs (carcinoids). Mutations in the *MEN1* gene are reported in 31–34% of sporadic gastrinomas. Exomic sequencing of sporadic pNETs found that the most frequently altered gene was

TABLE 113-6 GENETIC SYNDROMES ASSOCIATED WITH AN INCREASED INCIDENCE OF NEUROENDOCRINE TUMORS (NETS) (GI-NETS [CARCINOIDS] OR PNETS)

Syndrome	Location of Gene Mutation and Gene Product	NETs Seen/Frequency
Multiple endocrine neoplasia type 1 (MEN 1)	11q13 (encodes 610-amino-acid protein, menin)	80–100% develop pNETS (microscopic), 20–80% (clinical): (nonfunctional > gastrinoma > insulinoma) GI-NETs (Carcinoids): gastric (13–30%), bronchial/thymic (8%)
von Hippel–Lindau disease	3q25 (encodes 213-amino-acid protein)	12–17% develop pNETS (almost always nonfunctional)
von Recklinghausen's disease (neurofibromatosis 1 [NF-1])	17q11.2 (encodes 2485-amino-acid protein, neurofibromin)	0–10% develop pNETs, primarily duodenal somatostatinomas (usually nonfunctional) Rarely insulinoma, gastrinoma
Tuberous sclerosis	9q34 (TSCI) (encodes 1164-amino-acid protein, hamartin), 16p13 (TSC2) (encodes 1807-amino-acid protein, tuberin)	Uncommonly develop pNETS (nonfunctional and functional [insulinoma, gastrinoma])

Abbreviations: GI, gastrointestinal; PNETs, pancreatic neuroendocrine tumors.