Pancreatic neuroendocrine tumors (P-NETs) constitute a subset of pancreatic mass lesions characterized by diverse clinical presentations. Despite their inherent malignant potential, the timely identification and treatment of these tumors are critical for achieving favorable clinical outcomes. This study aims to shed light on the heterogeneous tumor biology of P-NETs and the management strategies employed at a tertiary care center in Pakistan.

Method: A retrospective study encompassing all patients with a biopsy-confirmed diagnosis of P-NETs at Shifa International Hospital between January 1st, 2016, and June 30th, 2021, was conducted. Meticulous data extraction from pathology records and thorough searches of medical records were performed to gather relevant demographic and clinical information.

Results: A total of 24 patients were retrieved from our database, with 13 (54%) female patients. The mean age was 49.5 ± 16.3 years. Eight out of the 24 patients presented with abdominal pain. Most patients (14 out of 24) had lesions in the pancreatic head region. In three cases, lesions exhibited multicentricity. The mean lesion size measured 4.4 ± 2.3 cm. Three of the 24 patients displayed distant liver metastasis at the presentation time. Nineteen out of the 24 patients underwent surgical resections, while endoscopic ultrasound (EUS)-guided biopsy was performed in 4 out of 24 cases. EUS-guided tissue biopsy yielded accurate diagnoses in all four cases.

Conclusion: Most P-NETs are non-functional, and there is an almost equal distribution between male and female patients. Solitary lesions predominate, and metastasis is uncommon at initial presentation. EUS-guided fine needle biopsy stands out as a dependable diagnostic modality for P-NETs.

Keywords: Neuroendocrine tumors, Pancreas, Clinical presentation, Management, Diagnosis

Introduction
Pancreatic neuroendocrine tumors (P-NETs) originating from the diffuse neuroendocrine cells are pretty uncommon and include a heterogeneous group of tumors of the pancreas. P-NETs comprise only 1 to 2% of all pancreatic malignancies, but their incidence has gradually increased over the
past few decades. Most P-NETs are indolent, but they do have malignant potential. The biological behavior of an individual P-NET is unpredictable. However, tumor grade, lymph node or liver metastasis, and tumor size help assess prognosis and clinical outcome. P-NETs can be functional (i.e., associated with clinical symptoms about hypersecretion of a specific hormone), nonfunctional, sporadic, or associated with familial syndromes; indolent in the long-term; or aggressive and life-threatening. Genetic disorders associated with P-NETs include multiple endocrine neoplasia type 1, von Hippel-Lindau syndrome, and neurofibromatosis type 1. P-NETs are typically hypervascular on imaging and undergo enhancement on arterial and venous phase images. Computed tomography (CT) scan has been reported to have significant prognostic value about pathological tumor grade as well as pancreatic duct involvement. Significant CT features that correlated with higher pathological grade include larger tumor size, non-hyperattenuation, presence of distant metastases, CT ratio, ill-defined tumor margins, lower sphericity, heterogeneous enhancing, lower attenuation values, vessel involvement, cystic degeneration, bile duct dilatation, and vascular invasion. Nuclear imaging with octreotide is helpful in the diagnosis of occult tumors not detected by anatomical imaging. Endoscopic ultrasound (EUS) is usually performed in conjunction with other imaging modalities, as it helps confirm the size and characteristics of these lesions and obtain tissue diagnosis simultaneously. EUS is the mainstay modality for diagnosing P-NETs with high diagnostic accuracy. According to the WHO classification scheme, the diagnosis of this group of tumors is based on the tumor's histopathology and the assessment of proliferation fraction. However, the former can be challenging due to the lack of well-defined histologic criteria and that the established criteria of >20 mitoses/10 high-power fields or Ki67>20% may not sufficiently distinguish well-differentiated P-NETs from poorly differentiated NETs. Over the last few decades, significant progress has been made in diagnosing, understanding the pathophysiology, and managing P-NETs. There is a lack of data on P-NETs from South Asia. Therefore, our experience with P-NETs is presented regarding the demographic features, characteristic findings on laboratory and imaging studies, and histopathology.

Materials and Methods

Study population
A retrospective descriptive study was conducted at Shifa Tameer-e-Millat University in Islamabad. Patients undergoing evaluation at Shifa International Hospital for the diagnosis of P-NET from January 1st, 2016, to June 30th, 2021, were included in the study. Patients were identified through pathology records retrieved from the institutional database. Medical records were scrutinized for demographic, clinical, and diagnostic variables. The clinical data under review encompassed age, gender, and symptoms presented at admission. Laboratory data included assessments of elevated hormone levels such as gastrin, insulin, vasoactive intestinal polypeptide (VIP), glucagon, chromogranin, and neuron-specific enolase. These parameters facilitated the distinction between functional and nonfunctional P-NETs. The diagnostic evaluation encompassed CT scan findings, with the pathological features of the tumor being also incorporated into the data collection process. Medical records were examined to determine if patients had undergone EUS-guided biopsy. The management plan and survival rate of patients were meticulously scrutinized and
evaluated throughout the study. All statistical analyses were executed using Statistical Package for the Social Sciences (SPSS) version 29. The Institutional Review Board (IRB) of the Human Research and Ethics Committee (EC) approved this study, with the ethical code number IRB #061-21. This study adheres to the principles of the Declaration of Helsinki.

**CT scan findings**
The meticulous assessment was conducted on the CT features, encompassing the following aspects: tumor site and size, peripancreatic vascular involvement, and upstream pancreatic duct dilatation.

**EUS- fine needle aspiration (EUS-FNA) examination**
Seasoned gastroenterologists conducted all procedures following the acquisition of informed consent from the patient. A 22-gauge fine needle biopsy was employed for the examination, with patients routinely administered a singular dose of intravenous antibiotics concurrent with the procedure.

**Results**
A total of 24 patients were retrieved from our database. 13 patients (54%) were females. The mean age of the patients was 49.5 ± 16.3 years. The most prevalent clinical presentation was abdominal pain, observed in 8 out of 24 patients (33.0%) (Table 1). Five (20.8%) patients presented with hypoglycemia, and three (12%) reported weight loss. The mean size of the lesion was 4.4 ± 2.3 cm (Figure 1). Fourteen out of the 24 (58.3%) patients had lesions involving the head region (Figure 2). In three cases, lesions were multicentric. 19 out of 24 (79.2%) patients underwent surgical resections. Three out of 24 patients had liver metastasis at the time of presentation. Two of these three patients had lesions in the tail of the pancreas. EUS was performed in 4 four cases, and a correct diagnosis of PNET was established in all four cases (Figure 3).

Histological diagnosis was based on morphology and immunohistochemistry (IHC). Synaptophysin staining was conducted in 23 out of 24 cases, with positive results in 22 out of 23 cases. The diagnosis relied on morphology in 2 cases. The Ki-67 index was reported in 23 cases, with ten histological specimens graded as G1, nine as G2, and five as G3 (Figures 4 and 5). The most frequently performed surgery was a Whipple's procedure, conducted in 10 out of 19 cases (52.6%). The final histological diagnosis indicated a well-differentiated tumor in 20 out of 24 (83.3%) patients. At the time of study analysis, 22 out of 24 (91.66%) patients were still alive.

**Discussion**
Our study reports 24 cases of P-NETs from a tertiary care center in Pakistan. An almost equal incidence of P-NETs was observed in both genders, with a slightly increased incidence in females. Our study reports a significantly higher proportion of non-functional tumors. A similar trend has been observed in recent studies. Patients in our report who were diagnosed with insulinoma had hypoglycemia on clinical presentation. On imaging, most patients presented with solitary lesions, and the head of the pancreas was the predominant location of the tumor. In 3 cases, patients had multicentric lesions, and one of these had distant metastasis to the liver. The mean size of the lesion was 4.4 +/- 2.3 cm, and it was noticed that lesions greater than 3 cm were associated with distant metastasis.

Our study reported positivity for synaptophysin in most patients on immunohistochemical analysis (figure 4B). This is consistent with literature reporting positivity for synaptophysin and other immunohistochemical markers, such as chromogranin A, which greatly aid the diagnosis.8-10 Chromogranin A provides
Insight into tumor burden, assists with treatment, and predicts prognosis with sensitivity, specificity, and accuracy of 66%, 95%, and 71%, respectively.\(^8\)\(^-\)\(^10\) P-NET diagnosis is based on a combination of pathology, imaging, and serum tumor markers. Other serum markers, such as neuron-specific enolase (NSE), pancreatic polypeptide (PP), pancreastatin, and subunits of human chorionic gonadotropin, can also aid in diagnosis. For functional P-NETs, patients often present with hormone-specific symptoms. Five patients (20.8%) in our study were diagnosed with insulinoma and presented with the clinical presentation of hypoglycemia. Our findings are consistent with the previous studies.\(^1\) Our study utilized CT as the primary imaging modality for tumor localization and vascular involvement, following established imaging guidelines for P-NETs.\(^11\)\(^-\)\(^12\) EUS-FNAC has a high sensitivity for grading malignancy in patients with small P-NETs, was performed in four cases, and correctly diagnosed P-NETs, demonstrating strong concordance between cytology and histopathological analysis.\(^4\) This is evident in the study by Eusebi et al., which included 91 patients undergoing 102 EUS procedures.\(^10\) The diagnostic sensitivity for EUS-FNA, EUS-FNB, and the combination of both methods was 88.4%, 94.3%, and 100%, respectively.\(^10\) Recent advances in P-NET imaging have significantly impacted management strategies. The use of Somatostatin receptor imaging (SRI) with radiolabeled SS analogs (SSA), such as 111 In-pentetreotide (Octreoscan) or 68Ga-DOTA-SSA PET/CT, has become popular in detecting P-NETs and monitoring recurrence, as these tumors have a high expression of somatostatin receptors (SSTRs).\(^13\) 68Ga positron emission tomography (PET) tracers have been reported to have higher diagnostic accuracy than traditional imaging modalities.\(^13\) Our study primarily relied on CT and EUS as the primary diagnostic modalities; PET scans were not performed on any of the patients. In our study, most patients had G1/G2 tumors, and the Ki-67 index was available for almost all cases. The histopathological analysis revealed well-differentiated P-NET in 83.3% of the patients, with an overall favorable prognosis. During the follow-up period, 91.6% of the patients remained alive and healthy. Our findings are consistent with another study done in Pakistan by Ali et al., which showed 5-year overall survival (OS) rates of 88%, 57%, and 0% for low, intermediate, and high-grade P-NETs and 94%, 79%, and 43% for complete, incomplete, and unresectable disease, respectively.\(^14\) Although P-NETs have a generally good prognosis after resection, about 21%-42% of patients have recurrence.\(^15\) Post-surgical prognostic factors for recurrence and mortality include tumor grade, stage, invasion, extent, and molecular markers.\(^16\) Furthermore, it has been reported that larger tumor size, high Ki-67 index, grade, and stage predict shorter OS, progression-free survival (PFS), and recurrence-free survival.\(^17\) In our study, 22 out of the 24 patients were alive and free from recurrence during the follow-up period. Although P-NETs are said to have low-grade malignant potential, about 40–80% of patients are metastatic at presentation, usually involving the liver (40–93%).\(^18\) This was also true for our study, as three patients had liver metastasis on presentation, and 2 out of these 3 patients had lesions involving the tail of the pancreas. Most of the patients in our study underwent curative surgical resection (79.2%). Pancreaticoduodenectomy (Whipple procedure) was the most common surgery (52.6%). This aligns with the existing literature, where surgical resection is the definitive treatment for achieving a cure.\(^19\) There is an ongoing debate regarding managing nonfunctional lesions <2 cm in
size, with no clear consensus between resection and surveillance. However, for patients with more significant than 2 cm P-NETs, invasive tumors, or radiographically positive lymph nodes indicating locally advanced disease, surgical resection is recommended as the primary treatment modality. After surgery, imaging is recommended once every 3 to 12 months for the first year and then every 6 to 12 months after that, according to guidelines from the National Comprehensive Cancer Network (NCCN) and the North American Neuroendocrine Cancer Tumor Society (NANETS). In our cohort, the average tumor size was 4.4 +/- 2.3, leading to most patients undergoing resection. As a result, the overall prognosis was favorable, with 91.7% of patients remaining alive and in good health during the follow-up period.

Our study addresses the scarcity of data on P-NET incidence in Pakistan and contributes as one of the few reports from this region. It offers valuable insights into the complex tumor biology and management strategies for P-NETs. However, several limitations deserve attention. First, the retrospective nature of the study should be noted. Secondly, patients from external facilities lacked sufficient follow-up information. Lastly, the study's small sample size should be considered when interpreting the results.

**Conclusion**

Most P-NETs are nonfunctional and solitary, exhibiting a similar distribution among both sexes. Targeted biopsies and immunohistochemistry (IHC) techniques can be employed to facilitate diagnosis. Surgical resections are a viable option for the majority of patients.

**References**


Table 1. Clinical presentation of patients with P-NETs

<table>
<thead>
<tr>
<th>Clinical Presentation</th>
<th>No.</th>
<th>% (age)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abdominal pain</td>
<td>8</td>
<td>33.3</td>
</tr>
<tr>
<td>Hypoglycemia</td>
<td>5</td>
<td>20.8</td>
</tr>
<tr>
<td>Weight loss</td>
<td>3</td>
<td>12.5</td>
</tr>
<tr>
<td>Jaundice</td>
<td>2</td>
<td>8.3</td>
</tr>
<tr>
<td>Incidental</td>
<td>1</td>
<td>4.2</td>
</tr>
<tr>
<td>Missing*</td>
<td>5</td>
<td>20.8</td>
</tr>
</tbody>
</table>

* Includes cases referred from outside facility; No.: Number

Figure 1. CT: This image illustrates a large, lobulated, well-circumscribed mass originating from the body of the pancreas, measuring $8.4 \times 9 \times 5.6$ cm.

CT: Computed tomography
Figure 2. This figure shows the distribution of P-NETs with regards to location.
P-NET: Pancreatic neuroendocrine tumors

Figure 3. This figure shows the endoscopic ultrasound: fine needle biopsy of a hypoechoic mass involving the body of pancreas.
Figure 4. Histological features of P-NETs - grade 1: This figure delineates the histological characteristics of Grade 1 well-differentiated neuroendocrine tumors (WD-NETs) of the pancreas. Panel A displays a photomicrograph revealing small, round, monotonous cells with coarse, salt-and-pepper nuclear chromatin and minimal atypia, organized in nests and trabeculae (H&E, ×200). Panel B exhibits synaptophysin immunohistochemical staining, demonstrating cytoplasmic positivity within the tumor cells (IHC, ×200). Panel C portrays Ki67 immunohistochemistry, showcasing rare positive cells (<1%) (IHC, ×200).

WD-NET: Well-differentiated neuroendocrine tumor; IHC: Immunohistochemistry; P-NET: Pancreatic neuroendocrine tumor
Figure 5. Histological features of P-NETs - grade 3: This figure illustrates the histological attributes of grade 3 well-differentiated neuroendocrine tumors (WD-NETs) of the pancreas. Panel A presents a photomicrograph displaying the trabecular arrangement of tumor cells (H&E, ×200). Panel B reveals Ki67 immunohistochemistry, indicating an elevated proliferative index (>20%) (IHC, ×200).

WD-NET: Well-differentiated neuroendocrine tumor; IHC: Immunohistochemistry