IJPRP

Indian Journal of Pathology: Research and Practice 8(5) September - October 2019
DOI: http://dx.doi.org/10.21088/ijprp.2278.148X.8519.16

Original Research Article

Study on Spectrum of Neuroendocrine Neoplasms of Digestive System

Nivedita SB[1], Gauri Kelkar[2]

[1]Consultant, Ashwini Sahakari Rugnalaya Ani Sanshodhan Kendra Nyt, Solapur, Maharashtra 413003, India. [2]Assistant Professor, Department of Pathology, Ashwini Rural Medical College, Hospital & Research Centre, Kumbhari, Solapur, Maharashtra 413006, India.

Corresponding Author:

Gauri Kelkar, Assistant Professor, Department of Pathology, Ashwini Rural Medical College, Hospital & Research Centre, Kumbhari, Solapur, Maharashtra 413006, India.

E-mail: drlalitagauri@gmail.com

Received on 29.06.2019,

Accepted on 24.07.2019

Abstract

Background: According to the survey data from the Surveillance, Epidemiology, and End Results (SEER) program, the incidence of malignant GI-NETs is increasing, probably due to increased physician awareness and improved diagnostic techniques. The present study was undertaken to study on occurrence and spectrum of neuroendocrine neoplasms in the digestive system. *Method:* This hospital-based study was conducted at the Kidwai Memorial Institute of Oncology, Bangalore. The present study consists of 118 cases of Digestive System NENs encountered during a 10 year study period. Retrospective cases were retrieved from the registers of Histopathology and Immuno histochemistry divisions and slides were analyzed. For the prospective study, complete clinical data was obtained from the patient and clinical records. Data regarding routine laboratory investigations, abdominal ultrasound, CT Scan, MRI, upper or lower GI endoscopy and peroperative findings, medical treatment and follow up data were analyzed. *Result:* Oesophagus was the most common site (35%), followed by the stomach (23%), small intestine (22.0%), large intestine (6.%), rectum (7%), pancreas (4.2%), one case each of appendix, gall bladder, common bile duct and metastasis to the liver. In the oesophagus, NEC was the most common diagnosis. G1 NETs were the most common NENs of the stomach, small intestine and pancreas. G2 NETs were most common in the large intestine, gall bladder, CBD, rectum & liver metastasis. *Conclusion:* The incidence of neoplasms is increasing because of early diagnosis by use of modern diagnostic methods and better physician awareness. Use of WHO 2010 classification helps in the classification of NENs and grading will impact the treatment and prognosis of individual patients.

Keywords: Neuroendocrine neoplasms; Digestive system; Immunohistochemistry; WHO classification.

How to cite this article:

Nivedita SB, Gauri Kelkar. Study on Spectrum of Neuroendocrine Neoplasms of Digestive System. Indian J Pathol Res Pract. 2019;8(5):620–631.

Introduction

Gastrointestinal neuroendocrine tumours (GI-NETs) are a genetically diverse spectrum of malignant solid tumours, a large number of which arise from the secretory cells of the diffuse neuroendocrine cell system which produce peptides causing characteristic hormonal syndromes. GI-NETs can be clinically symptomatic, i.e., 'functioning', or silent, i.e., 'non-functioning'. The estimated prevalence of neuroendocrine tumours (NET) in all parts of the body is 1 to 2 cases per 1,00,000 people, of which the gastrointestinal (GI) tract is the most common site.[1,2]

According to the survey data from the Surveillance, Epidemiology, and End Results (SEER) program, the incidence of malignant GI-NETs is increasing, probably due to increased physician awareness and improved diagnostic techniques. There are various modes of diagnosis of GI-NETs. These are based on clinical symptoms, hormone levels, radiological and nuclear imaging and histological confirmation. At the time of diagnosis, most patients with NETs present with metastatic disease, with regional or distant metastasis observed in 50% of patients. Initial metastases are usually noted in the regional lymph nodes, followed by the liver and finally in distant sites such as bone[1]

The majority of NETs are non-functioning and are diagnosed incidentally during an unrelated procedure. The clinical symptoms of functioning NETs generally arise after the tumour has metastasized to the liver [1]. NETs are classified based on the WHO 2010 classification of Tumours of the Digestive System as "Neuroendocrine tumour" which includes G1 and G2, and "Neuroendocrine carcinoma" which includes G3 and MANEC.NETs have a better prognosis than NECs and MANECs.[1,3]

Reporting of NENs[4]

The prerequisites of reporting NENs are knowledge of the exact site, size and the distance from the resection margins in excised specimen.

Microscopically-

- Mitoses counted per 10 HPF
- Ki67 Index

The assessment of endocrine function should be provided only on specific clinical request. The diagnosis of NENs should contain:

- Classification of the lesion (NET/NEC)
- Grade (G1/G2/G3)
- TNM stage
- Cell type and functional activity upon specific request

Objective:

To determine occurrence and spectrum of neuroendocrine neoplasms in the digestive system

Materials and Methods

This hospital-based study was conducted at the Kidwai Memorial Institute of Oncology, Bangalore. The present study consists of 118 cases of Digestive System NENs encountered during a 10 year study period (January 2005 – December 2014). The study was undertaken after the approval of the hospital ethics committee. Retrospective case numbers from January 2005–December 2013 were retrieved from the registers of Histopathology and Immuno histochemistry divisions and slides were analyzed. For the prospective study, complete clinical data was obtained from the patient and clinical records. Data regarding routine laboratory investigations, abdominal ultrasound, CT Scan, MRI, upper or lower GI endoscopy and peroperative findings, medical treatment and follow up data were analyzed.

Inclusion criteria

1. All Digestive system NENs diagnosed during the defined period.
2. Availability of paraffin blocks and IHC slides for review.
3. Availability of clinical records for correlation.

Exclusion criteria

1. Cases which were diagnosed only on FNAC, with no cell block
2. Cases which were diagnosed only on morphology

Data collection procedure

This study included resection specimens, biopsies as well as cases referred from other centres which were proven to be neuroendocrine neoplasms by immunohistochemistry.

Variables including age, gender, tumour site, histologic cell type, mitotic activity and necrosis were evaluated, with the last three on routine H

& E stained sections. Similarly, the categorisation of small & large cell carcinoma was done on morphology. The tumours were assigned to categories according to the WHO 2010 classification.

In the prospective cases where primary tumour tissue or representative tissue blocks were available, three markers were performed i.e., Synaptophysin, Chromogranin and Ki-67. In the remaining cases where primary tumour tissue was not available, the slides of immunohistochemistry already done for Synaptophysin, Chromogranin and Ki 67 were retrieved.

In the cases which had been proved to be neuroendocrine on IHC with chromogranin and synaptophysin but which had not been assessed for the Ki 67 index, a tissue microarray (TMA) was constructed & Ki 67 immunostainingwas performed on the TMA (12 cores). Cases where there were no available slides for review but which had been proven to be neuroendocrine in the IHC register in the Department of Pathology were also included for the total count. In two of three cases where there was no block for Ki67 immnostaining, marked crush artefact precluded mitoticcounting.

Method for immunohistochemistry

Method: Two- step polymer- HRP detection system was used.

Principle: A detection system using a non-biotin polymer technology that makes use of only one major component, a polymer- HRP reagent. This reagent consists of both secondary immunoglobulin molecules and horse radish peroxidase molecules linked to a common dextran polymer backbone, thus eliminating the need for sequential application of link antibody and peroxidase conjugated antibody.

Primary antibodies (Table 1)

Table 1:

Antibody	Species	Clone	Dilution	Manufacturer
Anti-Chromogranin A (LK2H10)	Mouse	Monoclonal	1:300	BioGenex
Anti-Synaptophysin (Snp 88)	Mouse	Monoclonal	1:8	BioGenex
Anti-Ki-67 (EPR)	Rabbit	Monoclonal	1:700	BioGenex

Secondary antibody used

The secondary antibody used was the Biogenex Two- Step Polymer- HRP Reagent

Controls used

Positive and negative controls were included in each staining run. The positive controls were sections of known cases of chromogranin & synaptophysin positive pheochromocytoma. The positive controls for Ki 67 immunostaining were sections of reactive lymph node. Negative controls were tumour sections with omission of the primary antibody.

Steps of IHC:

Procedure (outline)

Tissue sections on triethoxysilane coated slides were dewaxed, treatedwith an antigen retrieval solution if required, blocked with hydrogen peroxide and 2% skimmed milk powder blocking solution and then incubated with a primary antibody. The primary antibody would bind to the antigen of interest. The bound primary antibody was detected by the addition of secondary antibody conjugated with horse radish peroxidase polymer and DAB substrate. When adequate colour development was seen, the slides were washed in water to stop the reaction, counterstained and covered with a mounting medium.

○ *Tissue section cutting*

Sections were cut at 3–4 micron thickness from tumour blocks and taken on totriethoxysilane (3-Amino propyl) coated slides and labelled with the patient's block number using a glass marker. These slides were incubated for 12–16 hours at 58° C prior to start of deparaffinization.

○ *Deparaffinization of tissue sections*

Slides were incubated in glass jars containing Xylene I (15 minutes), Xylene II (15 minutes),

absolute alcohol I (2 minutes) and absolute alcohol II (2 minutes). Following this, sections were transferred to running tap water for five minutes and then rinsed in distilled water for 5 minutes.

○ *Heat induced epitope retrieval*

Epitope retrieval was done by use of multiple epitope retrieval system (MERS) containing EDTA at pH 9 retrieval solution.

○ *Primary blocking*

Slides were incubated in 3% H_2O_2 in distilled water for 20 minutes.

○ *Secondary blocking*

The slide edges were wiped clean and placed on the IHC staining rack. The tissue sections were covered with 2% skimmed milk powder blocking solution and incubated at room temperature for 20 minutes.

○ *Addition of primary antibody*

Primary antibody solution in appropriate dilutions was added on tissue sections. Slides were incubated at room temperature for a specified time according to the protocol for each antibody. Slides were washed in TBS I and II staining jars for 2 minutes each.

○ *Addition of secondary antibody*

The tissue sections were covered with labelled polymer solution and incubated at room temperature as per kit instructions. Then the slides were washed in TBS I and II staining jars for 2 minutes each.

○ *Preparation and addition of 5% DAB chromogen solution*

The appropriate amount of buffer needed from the substrate bottle (DAKO Emission Kit) was transferred into a calibrated test tube. For each one mL buffer, one drop of solution from the chromogen bottle was added and mixed immediately as per the kit instructions. The tissue sections were covered with 100 mL of DAB chromogen solution and incubated at room temperature for 3 minutes and then washed with distilled water for 5 minutes.

○ *Counterstain*

Tissue sections were overlaid with Harris Hematoxylin for 20 to 30 seconds and then washed in running tap water for 2 minutes.

○ *Dehydration of tissue sections*

Dehydration was done by incubating in jars containing 96% ethanol, absolute alcohol, and xylene placed in sequential order for 3 minutes and 10 minutes respectively.

○ *Mounting*

Sections were mounted using DPX and an appropriate cover slip and incubated at 50°C for 15 minutes. After drying, the slides were labelled with patients block number.

Statistical analysis

Descriptive statistics such as mean, SD and percentage was used. Data was analysed by test for proportions with use of R software. Statistical values of $p < 0.05$ were considered as significant.

Results

In the present study, 65 (55.0%) were males and 53 (45%) were females. The mean age at presentation was 52.23 years. The mean age at presentation for

Table 2: Basic characteristics

Characteristics	No. of cases	Percentage
Gender		
Male	65	55
Female	53	45
Age group (years)		
< 20	1	0.8
21–30	9	7.6
31–40	15	12.7
41–50	28	23.7
51–60	28	23.7
61–70	27	22.9
71–80	8	6.8
> 80	2	1.7

males in the study was 53.1 years, for females it was 51.15 years. Most of the cases were in the age group of 40 to 70 years (70.3%). The youngest patient was 19 years old and the oldest patient was 84 years old (Table 2).

Of the NENs which were studied, oesophagus was the most common site (35%), followed by the stomach (23%), small intestine (22.0%), large intestine (6.%), rectum (7%), pancreas (4.2%), one case each of appendix, gall bladder, common bile duct and metastasis to the liver. In the present, majority of specimen belongs to biopsy (77.6%) followed by resected (22.4%). Majority of cases have clinical symptoms of dysphagia and dyspepsia (71.4%) followed by weight loss (17.9%), and vomiting (7.1%) (Table 3).

Table 3: Clinical parameters

Clinical parameters	No. of cases	Percentage
Location		
Oesophagus	41	34.7
Stomach	27	22.9
Small intestine	26	22.0
Large intestine	15	12.7
Rectum	8	6.8
Pancreas	5	4.2
Appendix	1	0.8
Gall bladder	1	0.8
Common bile duct	1	0.8
Metatasis	1	0.8
Specimen type		
Biopsy specimen	66	77.6
Resected specimen	19	22.4
Clinical symptoms		
Dysphagia and dyspepsia	40	71.4
Weight loss	10	17.9
Vomiting	4	7.1
Pain abdomen	2	3.6

Of the NENs which were studied, 33 (40%) cases were G1, 24 cases (29%) were G2, 21 (25.3%) cases were G3, MANEC accounted for three(4%) cases. Two NENs where no block was available were excluded from grading due to marked crush artefact of the biopsy due to of which mitotic count could not be done. Out of 118 cases, 83 cases were reviewed and reclassified according to the current WHO 2010 classification of neuroendocrine neoplasms of the digestive system. Two NENs where no block was available were excluded from the grading due to marked crush artefact of the biopsy due to which

Table 4: Grading of NENs

Grading Site	Grade 1	Grade 2	Grade 3	MANEC	Total
Oesophagus	6	8	18	–	32
Stomach	8	7	3	1	19
Small intestine	13	3	–	–	18
Appendix	–	–	–	1	1
Large intestine	2	1	–	–	3
Rectum	–	1	–	1	2
Pancreas	4	1	–	–	5
Metastasis liver	–	1	–	–	1
CBD	–	1	–	–	1
Gall bladder	–	1	–	–	1
Total	**33**	**24**	**21**	**3**	**83**

mitotic count could not be done. For the remaining 33 cases where there were no available slides for review, the diagnosis of NENs was taken from the IHC register in the Department of Pathology (Table 4).

Table 5: Most common grade of tumours in various sites

Site	Most common grade	Percentage(%)
Oesophagus	G 3	56.25%
Stomach	G 1	42.1%
Small intestine	G 1	72.0%
Large intestine	G 2	66.6%
Pancreas	G 1	80.0%

In the oesophagus, NEC was the most common diagnosis. G1 NETs were the most common NENs of the stomach, small intestine and pancreas. G2 NETs were most common in the large intestine, gall bladder, CBD, rectum & liver metastasis. In the stomach, appendix and rectum, there was one case each of MANEC (Table 5).

Table 6: Comparison of grades between sites

Site	Grade	*p*-value	Comment
Oesophagus Vs Stomach	G 1	p=0.0785	No significance
Oesophagus Vs SI	G 1	p=0.002	Significant
Stomach Vs SI	G 1	p=0.0698	No significance
Oesophagus Vs Stomach	G 2	p=0.4077	No significance
Oesophagus Vs SI	G 2	p=0.4636	No significance
Stomach Vs SI	G 2	p=0.1729	No significance
Oesophagus Vs Stomach	G 3	p=0.0054	Significant

The frequency of occurrence of grade 1 NET at small intestine is more compare to oesophagus and stomach (p = 0.002). The frequency of occurrence of grade 3 NEC at oesophagus is more compared to stomach and small intestine (p = 0.005) (Table 6).

Morphology

Gross

The gross appearance of resected specimens varied from small polypoidal lesions to large well circumscribed masses.

Of 19 resected specimens, 12 (63%) cases of NET were small, polypoidal, grey white to grey yellow in colour, with size varying from 1.5 to 4 cms. Most of them appeared confined to the mucosa and submucosa. None of the NETs showed surface ulceration.

Four NEC cases (21%) & three cases of MANEC (16%) were large globular masses, grey white to grey yellow in colour, varying in size from 4 to 6.5cms. Most of them showed surface ulceration and involvement of the entire thickness of the wall (Figs. 1-2).

Fig. 1: The gross appearance of a resected specimen of small intestine showing a well circumscribed polypoidmass with a yellowish surface, with unremarkable adjacent duodenalmucosa.

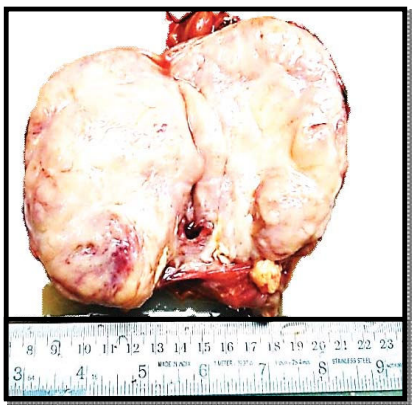


Fig. 2: The cut section varied from grey white to pinkish in appearance.

Microscopic features

On microscopic examination, NENs were graded and histologic typing was done. For resected specimens, in addition to the microscopic diagnosis, the status of resected margins (proximal, distal and circumferential) was evaluated. Dissected lymph nodes were evaluated for metastasis & when present, extracapsular spread.

Tumour blocks were evaluated for adequacy of representative tumour for immunostaining.

Of the 83 cases, 57 cases (69%) were G1 & G2. The tumour cells were arranged in insular and sheet like patterns. The nuclei were round to oval, with salt and pepper chromatin and abundant eosinophilic granular cytoplasm. They showed mitotic rates of <2–9 per 10 HPF and only one case (G2) showed areas of necrosis.

23 cases (28%) were G3. The tumour cells were classified into large cell and small cell types. Of 23 cases, 19 cases (83%) were small cell carcinoma with cells arranged predominantly in a sheet-like pattern. The tumour cells were small, round and ovoid with a high N:C ratio, scant granular cytoplasm, fine chromatin and inconspicuous nucleoli. Many of the biopsies showed marked crush artefact.

4 cases (17%) were large cell carcinomas, with tumour cells arranged in ill-defined nests and sheet like patterns. The cells were medium to large in size, with fairly abundant eosinophilic granular cytoplasm and large atypical nuclei with prominent nucleoli. They showed mitotic rates of >20 per 10 HPF & multifocal areas of necrosis in resected specimens. In addition, two oesophageal NECs showed full thickness dysplasia of the overlying squamous epithelium.

Three cases (4%) were MANEC in resected specimens, with features of both adenocarcinoma and neuroendocrine carcinoma. Each component comprised more than 30% of the tumour with relatively sharp demarcation between the two components. The adenocarcinoma component showed cells arranged in glandular patterns (2) or signet ring cells (1). The neuroendocrine component in 2 cases showed one case each of small and large cell NEC, with cells arranged in sheets, mitotic rates of >20 per 10 HPF and areas of necrosis. In one case, the neuroendocrine component was G1.

Of the 83 cases, 57 cases were G1 & G2, all of which (100%) showed diffuse and intense staining for synaptophysin and chromogranin. All 26 cases of G3 & MANECs combined showed diffuse & intense staining for synaptophysin (100%). However, staining for chromogranin was absentin 6 cases (23%) and patchy in 20 cases (77%).

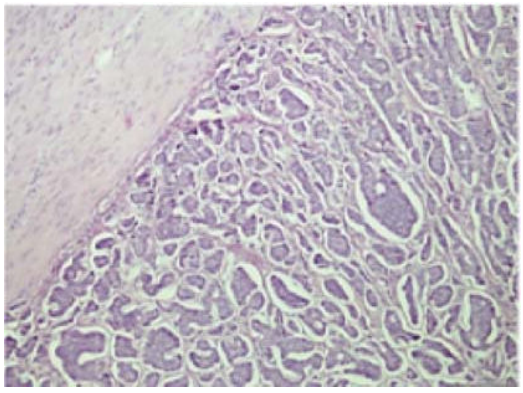
Ki 67 index in G1&G2 varied from <2 to 18% in 57 cases. It varied from >20 to 90% in G3 and MANECs in 26 cases.

Photographs of cases *(Fig. 3-8)*

Out of 56 patients who underwent biochemical investigation for serum chromogranin A, a raised value was noted in eight cases.

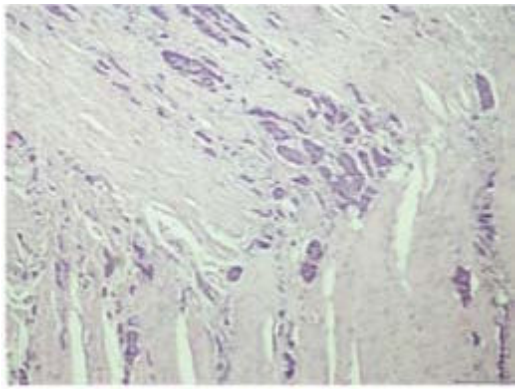
Treatment & follow up:

Of 56 cases, 30 patients had taken treatment. Surgery was the modality of treatment for 22 cases. Sixteen



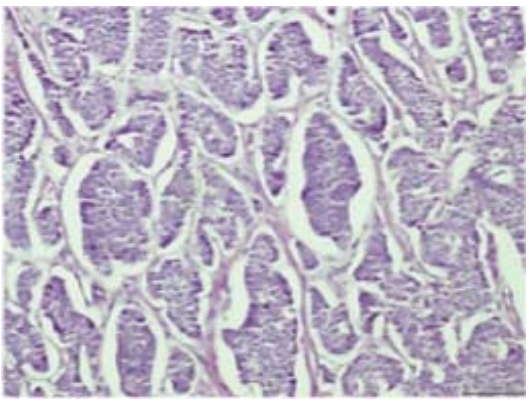
a

Fig. 3a: Resected specimen of Grade 1 NET, H&E stain (x40)



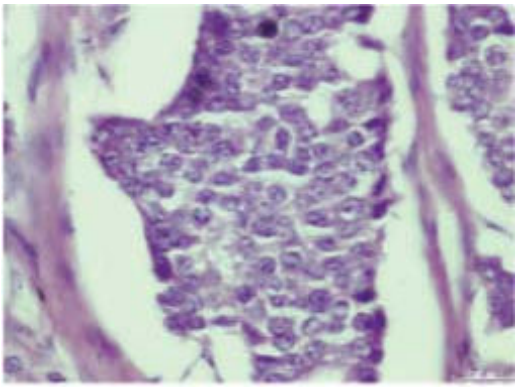
b

Fig. 3b: Tumour cells infiltrating the muscularispropria, H&E stain (x40)



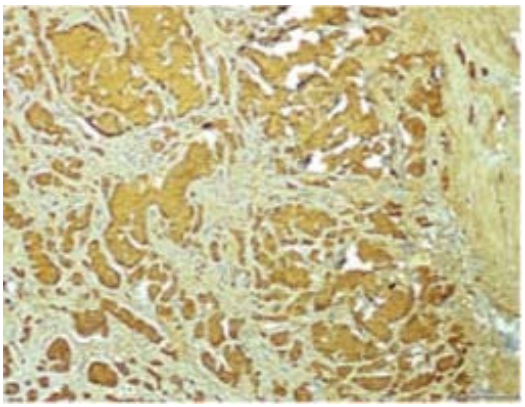
a

Fig. 4a: Tumour cells in anorganoid pattern, H&E stain(x100)



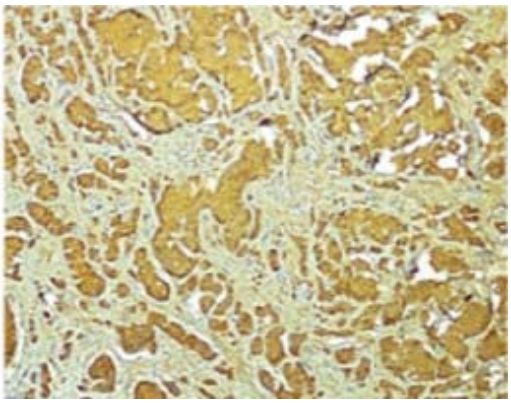
b

Fig. 4b: Small round tumour cells with scant eosinophilic cytoplasm and round nucleiwith salt and pepper chromatin, H&E stain (x400)



a

Fig. 5a: Tumour cells showing diffuse & intense cytoplasmic positivity for Chromogranin Aimmunostain (x40)



b

Fig. 5b: Tumour cells showing diffuse & intense cytoplasmic positivity for Synaptophysinimmunostain (x40)

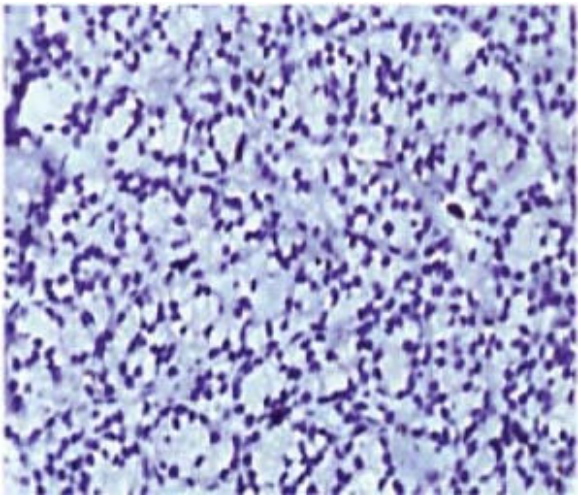


Fig. 6: Tumour cells show <2% of proliferative index in G1 NET (Ki 67immunostain x100)

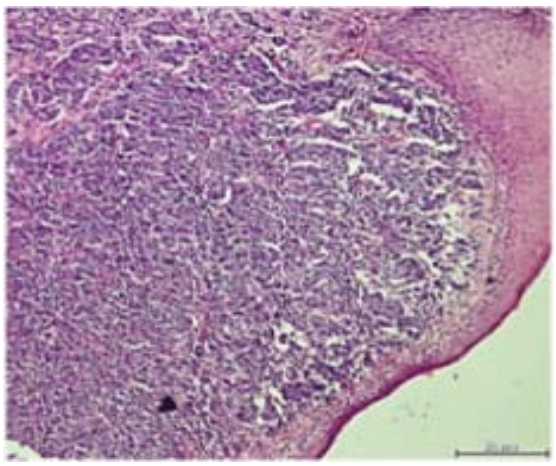


Fig. 7a: Grade 3 NEC oesophagus, resected specimen, H&E stain (x40)

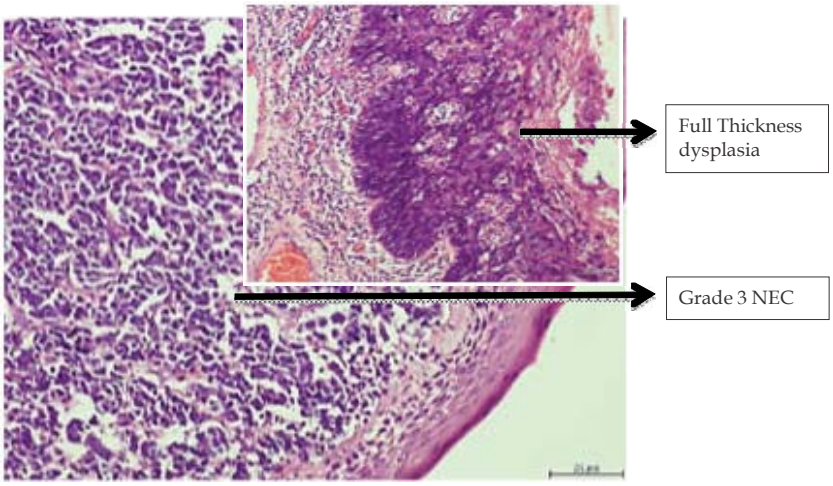


Fig. 7b: Tumourcells arranged in diffuse sheets with the adjoiningsquamous epithelium exhibiting high-grade dysplasia, H&E stain (x100)

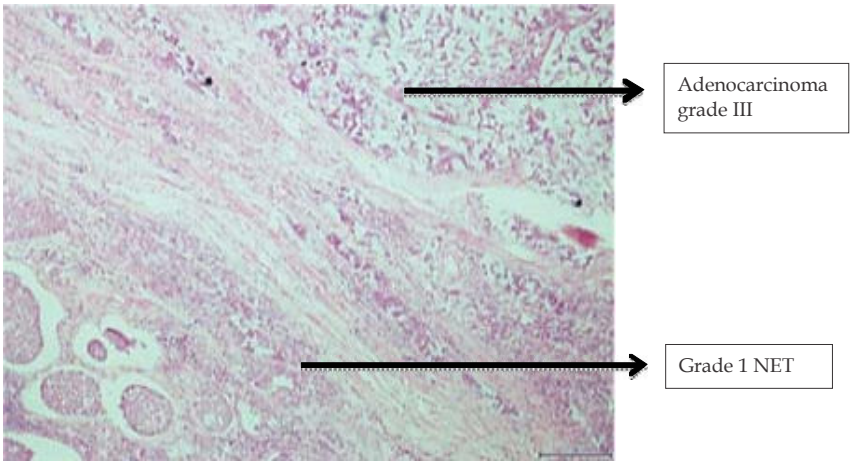


Fig. 8a: Grade 1 NET of duodenum with adjoining area showing adenocarcinoma Grade III, resected specimen, H&E stain (x40)

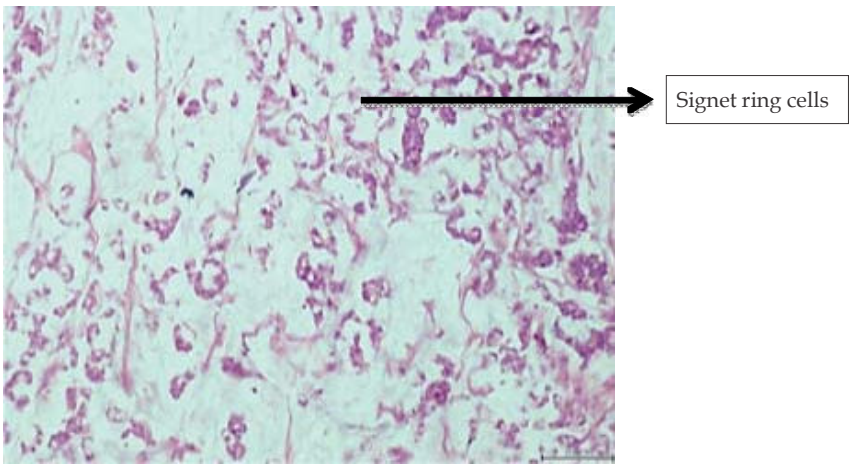


Fig. 8b: Shows adenocarcinoma cells with signet ring morphology in a mucinous background, H&E stain(x100)

cases with localised disease which included NETs (G1 & G2) were operated upfront. In 6 borderline resectable cases which included G2 & G3 tumours, neoadjuvant chemotherapy was given. In 8 cases of NECs with liver metastases, palliative treatment was administered.

No follow-up was available for cases in the period 2005–2013, despite vigourous attempts to obtain this information. In the prospective arm, follow up of one year for 8 cases diagnosed in the year 2014 was available, which included one oesophageal NET (G1), two oesophageal NECs, one small intestinal NEC, two SI NET (G2) and one rectal and one stomach MANEC.

One of two oesophageal NECs showed metastasis to the lungs. One small intestinal NECs showed recurrence and one stomach MANEC case showed metastasis to the liver, pancreas and lungs. The oesophageal NET (G1) and SI NET (G2) did not show any recurrence or metastasis. It is not possible to correlate tumour grade with survival in view of the small number of cases with follow-up data.

Discussion

Gastrointestinal and pancreatic NENs are rare malignant tumours. Due to improved diagnostic and therapeutic modalities, they have gained attention over last few years. From all over the world, there is limited epidemiological data available for GI and pancreatic NENs. From India there are very few reports available on epidemiology of these tumours.[5,6]

Of the 118 cases, the majority of the patients were in the age group of 41 to 70 years with a mean age group of 52 years, which was similar to that of the study conducted by Amarapurkar DN *et al*.[2] The youngest patient was 19 years and oldest was 84 years.

In our study, we found 65 (55.08%) cases were male and 53(44.9%) cases were female. NENs were more common in males than females (M:F=1.2:1), similar to the studies of Amarapurkar DN *et al*.[2] and Tessa *et al*.[7]

The clinical data was available in 56 of 118 cases. Patients presented with dysphagia, dyspepsia, weight loss, vomiting and pain abdomen as the common symptoms. This is comparable to the studies of Amarapurkar DN *et al*.[2] & Tessa *et al*.[7]

The oesophagus was the most common site of NENs in our study, whereas stomach, pancreas, appendix/colon were the most common anatomical sites in the studies by Amarapurkar DN *et al*.,[2] and Tessa *et al* respectively.[7]

In the large series of Modlin and Sandor consisting of 8305 carcinoid tumours from the SEER database and 2 NCI archives, only 3 arose in the oesophagus, accounting for 0.02% of all carcinomas in the oesophagus and 0.05% of all GI NET cases.[8] In our study we found 41 (35%) cases of oesophageal NENs from total 118 cases. The reason for the high percentage of oesophageal NECs in the present study is probably a reflection of the large number of cases presenting with dysphagia who are referred to our government–run tertiary care cancer centre for palliative radiotherapy.

Histomorphological features were observed in the oesophageal NECs and cell typing was done. There were 29 cases of small cell neuroendocrine carcinoma and one case of large cell neuroendocrine

carcinoma. In two cases of oesophageal NECs, we observed full thickness dysplasia of the squamous epithelium. In a study of Indian patients by Bhavana *et al.* over a period of eight years, 11 cases of small cell carcinoma of the oesophagus were analysed.[9]

Primary oesophageal small cell carcinoma (SCC) is extremely rare, accounting for 0.4 to 2.8% of all oesophageal malignancies.[10,11] It is a highly aggressive malignancy and often presents with widespread metastasis at diagnosis, resulting in poor clinical outcome.[12] In the present study, we analysed the clinicopathologic features of all cases diagnosed as primary esophageal SCC over a period of ten years.

The oesophagus is reported as the most frequent digestive tract site of extra pulmonary SCC with an incidence of less than 1.5%.[12,13] Incidence of oesophageal SCC in our series was 25.4%. Oesophageal SCC usually affects individuals from the sixth to eighth decades with male predominance.[14,15] In our study, we found the majority of cases occurring in the age group of 40-80 years with male predominance, which was comparable to the study done by Bhavana *et al*.[9] On review of clinical data of these patients, 10 cases had history of smoking, which could be the one of the risk factors for NEC of oesophagus, comparable to the study done by Bhavana *et al*. [9]

In this study, inpatients who underwent endoscopy for dyspepsia, apolypoidal lesion in the oesophagus was an incidental finding. Most of these patients presented with loss of weight and loss of appetite. Four patients presented with liver metastasis and two patients with lung metastasis at the time of diagnosis, which is comparable to the study of Tessa *et al*[7].

Generally, the clinical manifestations of carcinoid tumours are vague or absent unless the tumour is large enough to create a mass effect or secretes bioactive mediators that create specific syndromes.[13] Depending on whether the secreted hormone is detectable and associated symptoms are present, GEP- NETs can be divided into functioning and non-functioning tumours.[15] From the clinical data which was available for 55 cases, none of them presented with carcinoid syndrome in our study. This was comparable to the study of Tessa *et al*.[7] In the study conducted by Amarapurkar *et al*. only 4.1% patients presented with the carcinoid syndrome.[2]

Out of 56 patients who underwent biochemical investigation for serum chromogranin A, a raised value was noted in eight cases. We can correlate this to secretory NENs. There is a direct correlation between the tumour burden and serum CgA levels with sensitivity and specificity of 92% and 96% in disease free patients and those with recurrent metastatic tumour.

Grade 1 NETs were 34, Grade 2 NETs were 24 and Grade 3 NECs were 22, MANEC were 3 according to the current WHO 2010 classification of digestive system neuroendocrine tumours, which is comparable to the grading done by Zeng Y J *et al*.[16] When histomorphological pattern was studied, most cases showed organoid pattern in G1 and G2 NETs, whereas in G3 (NECs) diffuse sheets and necrosis was noticed in most cases. Many cases showed myoinvasion in G1 & G2 NETs, which was comparable to the other studies in the literature.

In follow up of 8 cases, patients with G1 & G2 had better prognosis and survival compared to G3 & MANEC, which was comparable to the study done by Pepe *et al.* and La rosa *et al*.[17,18]

Of 56 cases where we had clinical data, 30 patients had taken treatment. Surgery was the main modality of treatment for cases with local disease in 16 cases. In borderline resectable cases, neoadjuvant chemotherapy was given. Six cycles of chemotherapy were given in Grade 1 and G2 NETs with Octreotidelar & Everolimus or only Octreotide in four cases. For Grade 3 NECs, cisplatin & etoposide were administered in 10 cases.

Conclusion

Oesophagus emerged as the most common site of NENs in our study. The incidence of these neoplasms is increasing because of early diagnosis by use of modern diagnostic methods and better physician awareness of these neoplasms. Use of WHO 2010 classification helps in the classification of NENs and grading will impact the treatment and prognosis of individual patients.

Prior publication: Nil

Support: Nil

Conflicts of interest: Nil

Permissions: Nil

References

1. Oberg KE. Gastrointestinal neuroendocrine tumours. *Annals of Oncology*. 2010;(7)21:72–80
2. Amarapurkar DN, Juneja MP, Patel ND, *et al.* A retrospective clinicopathological analysis of

neuroendocrine tumours of the gastrointestinal tract. *Trop*ical Gastroenterology. 2010;31(2):101–04

3. Ehehalt F, Saeger H D, Schmidt C M, *et al.* Neuroendocrine tumours of the digestive system. Oncologist. 2009;14:456–67
4. Bosman FT, Carneiro F, Hruban RH. Theise N.D.(Eds):WHO classification of Tumours of Digestive System. IARC; Lyon 2010.
5. Hegde V, Mohandas KM, Ramadwar M, *et al.* Carcinoids-a changing trend. Indian Journal of gastroenterology. 2003;22:209–211
6. Radhakrishanan S, Subramoniam S. Colorectal carcinoids in south India. Tropical geographical Medicine. 1979;31:63–67
7. Joseph T and Shanthala PR. Gastroenteropancreatic Neuroendocrine Tumours – An Institutional Experience. International Journal of Biomedical Research. 2015;6(02):71–76.
8. Modlin IM and Sandor. An analysis of 8305 cases of carcinoid tumours. Cancer. 1997;79(4):813–829.
9. Bhavana *et al.* Primary Small Cell Carcinoma of TheEsophagus – An Eight Year Retrospective Study Journal of Clinical and Diagnostic Research. 2015 May;9(5):4–6.
10. Chen WW, Wang F, Zhang DS, *et al.* Primary small cell carcinoma of the esophagus: Clinicopathological study of 44 cases. BMC Cancer. 2014;14:222.
11. Lv J, Liang J, Wang J, *et al.* Primary small cell carcinoma of the esophagus. J Thorac Oncol. 2008;3(12):1460–1465.
12. Pantvaidya GH, Pramesh CS, Deshpande MS, *et al.* Small cell carcinoma of the esophagus: The Tata Memorial Hospital experience. Ann Thorac Surg. 2002;74(6):1924–27.
13. Law SY, Fok M, Lam KY, *et al.* Small cell carcinoma of the esophagus. Cancer. 1994;73:2894–99.
14. Nevárez A, Saftoiu A, Bhutani MS. Primary Small Cell Carcinoma of the Esophagus: Clinico - pathological Features and Therapeutic Options. Curr Health Sci J. 2011;37(1):31–34. E pub 2011 Mar 21.
15. Casas F, Ferrer F, Farrús B, *et al.* Primary small cell carcinoma of the esophagus: a review of the literature with emphasis on therapy and prognosis. Cancer. 1997;80(8):1366–72.
16. Zeng YJ, Liu L, Wu H, *et al*. Clinicopathological features and prognosis of gastroenteropancreatic neuroendocrine tumours: Analysis from a single-institution. Asian Pac J Cancer Prev 2013;14:5775–5781.
17. Pepe UF, Jann H, Muller–Nordan J *et al*. Prognostic relevance of novel TNM classification system for upper gastroenteropancreatic neuroendocrine tumours. Cancer. 2008;113:256–265
18. La Rosa S, Inzani F, Vanoli A *et al*. Histologic characterization and improved prognostic evaluation of 209 gastric neuroendocrine neoplasms. Hum. Pathol. 2011;42:1373–1384.