Prevalence and pathology of gastric tumours in Indian oil sardine (Sardinella longiceps) from Parangipettai coastal waters, southeast coast of India

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1. Introduction

Gastric tumour is a benign or malignant nature epithelial and non-epithelial cellular uncontrolled proliferation of the stomach cells or their progenitor[1]. The radiographical observations are generally used to assess skeleton deformities, bone demineralization and tumours[2-5]. Histopathological examination is usually used to assess the manifestation of diseases, health of organisms and evaluation of the organ structure, which reflect the morphological structure of the cells and tissues[6,7]. In human, the gastric tumour diagnosis is predominantly based on the histological pattern[1]. Similarly, neoplasms of lower animals are also classified based on higher animals[8]. They are classified histologically as epitheloid and non-epitheloid subtypes[9].

Tumours in fish have been reported in almost of all organs[10]. But gastric tumour related reports are meager, such as harbor porpoise, blue shark and sea bass[11-13]. The Indian oil sardine [Sardinella longiceps (S. longiceps)] is distributed abundantly along the east and west coasts of India[14]. They are highly suffered with skin neoplasm[2]. This study reports prevalence and histological observation of the stomach tumours in S. longiceps from Parangipettai coastal waters, south east coast of India.

2. Materials and methods

2.1. Study area and sample collection

The Indian oil sardine was directly collected from the fishing boats and auction yard of the Parangipettai fish landing centre since January 2014 to December 2014. The collected fishes were examined for the stomach tumour infection. The length and weight of normal and tumour infected fish was measured. Stomach tumour infected fish were dissected and the tumour...
diameter were measured, weight and sex of the fish were recorded. The prevalence of gastric tumours in collected *S. longiceps* was analyzed.

### 2.2. Radiography

The collected fish were thoroughly washed with tap water followed by distilled water and radiographed at 100 mA in 45 kV.

### 2.3. Histopathology

Tumour lesions were excised and preserved in 10% neutral phosphate buffered formalin fixative. The biopsies were washed with tap water and dehydrated with different grades of ethyl alcohol, cleaned with xylene and embedded into a paraffin wax. The thin section of 3 μm was incised with rotary microtome. The sections were stained with haematoxylin and eosin (H & E) [15]. The tissues were observed under phase contrast microscope (Nikon Eclipse Ts 100).

### 3. Results

#### 3.1. Gross pathology

The stomach tumour infected fishes showed distended abdomen. The swelling occupied the coelomic cavity and both lateral side and ventral sides were enlarged (Figure 1).

**Figure 1.** Gross observation of *S. longiceps*. A: Normal; B: Stomach tumour; Rulers show the length (cm).

#### 3.2. Radiography

The radiographic analysis of the gastric tumour infected fish clearly exhibited the enlarged tissue mass of stomach which merged with part of the alimentary canal. The normal structural arrangement of other visceral organs were disturbed (Figure 2).

**Figure 2.** Radiograph of *S. longiceps*. A: Normal stomach; B: Stomach tumour.

#### 3.3. Tumour prevalence

The seasonal prevalence of stomach tumour (Gastroma) in *S. longiceps* landed at Parangipettai landing center are given in Table 1. Of 16321 individuals, 31 were infected with stomach tumour, with an overall prevalence of 0.19%. The higher seasonal prevalence (0.34%) was observed during monsoon, followed by pre-monsoon (0.32%). Month wise higher prevalence (0.63%) was observed during October followed by September (0.54%), August (0.36%), November (0.20%) and July (0.10%). Sex wise higher prevalence was 58.06% (18) observed in female, when compared with males 41.94% (13).

**Table 1**

| Season     | Month | No. of fish examined | No. of tumour infected fish | Prevalence (%) |
|------------|-------|----------------------|------------------------------|----------------|
|            |       |                      |                              | Month          | Season |
| Post-monsoon| Jan   | 355                  | 0                            | 0.00           | 0.00   |
|            | Feb   | 486                  | 0                            | 0.00           |        |
|            | Mar   | 958                  | 0                            | 0.00           |        |
| Summer     | Apr   | 1425                 | 0                            | 0.00           | 0.00   |
|            | May   | 1932                 | 0                            | 0.00           |        |
|            | Jun   | 1813                 | 0                            | 0.00           |        |
| Pre-monsoon| Jul   | 1969                 | 2                            | 0.10           | 0.32   |
|            | Aug   | 1648                 | 6                            | 0.36           |        |
|            | Sep   | 1665                 | 9                            | 0.54           |        |
| Monsoon    | Oct   | 1754                 | 11                           | 0.63           | 0.34   |
|            | Nov   | 1492                 | 3                            | 0.20           |        |
|            | Dec   | 824                  | 0                            | 0.00           |        |

#### 3.4. Description of the tumour

Gastroma in sardines varied in their appearance and size. Most of the tumours were white to reddish in colour and few were in reddish brown. Multi-lobed tumours were found in most of the fishes, whereas few were single lobed. They were consistency hard muscle with smooth surface (Figure 3). The neoplasms ranged in size between 1.9–4.3 cm in diameter and 7.8–18.3 g in weight. The normal stomach weight varied between 0.8–1.2 g. The stomach weight of the normal fish represented between 1.23%–1.60% of their total body weight, but tumour infected stomach occupied 12.46%–
17.29% of their total body weight. The infected fish alimentary canal contains meager quantity of food matrix than normal fish. The infected fishes was found to have immature gonads.

Figure 3. Gastric tumour colour and consistency of S. longiceps.
A: Normal stomach (arrow); B: Whitish hard muscle (arrow); C: Tumourous elongated fundus region exhibiting pink to red colour; D: Single lobular reddish smooth muscle; E: Multi-lobular reddish smooth muscle (arrow). F: Multi-lobular smooth muscle pink colour with red patches; A, B and C: Coiled alimentary canal; D, E and F: Uncoiled alimentary canal; Rulers show length (cm).

3.5. Histopathology

Histological examination of normal stomach tissue of S. longiceps showed the spindle shaped fibroblast cells intermingled with moderate collagen fibers and moderate basophil-like small nuclei cellular matrix (Figure 4). Tumour lesions showed highly differentiated gastric stratified squamous hyperplasia and also exhibited undifferentiated squamous dysplasia with abundant and prominent monocytoid morphology. Numerous undifferentiated glandular epithelial cells were observed. The glands were elliptical structure and widened the distance between the pits and glands. The glandular cells contain intracytoplasmic mucin and the cells were scattered in the lamina propria. Mitotic figures were observed in the glandular epithelial cells (Figure 5A). The tumour lesion composed hyalinised loose collagen fibres intermingled with hypocellular matrix (Figure 5B). Well differentiated matured hyperplastic adipose cells packed with large fat vacuoles, hence nuclei were found in the periphery (Figure 5C). An undifferentiated hyperplastic fibroblast cells with lower degree of collagen fibres were observed. Cellular anaplasia and high mitotic rate were observed (Figure 5D).

Figure 4. Cross section of normal stomach of S. Longiceps shows fibroblast cells intermingled with prominent collagen (arrow) and moderated small nuclei cells (arrow head). H & E.

Figure 5. Gastric tumours of S. longiceps shows 4 major patterns of cellularity.
A: Glandular epithelial cells containing intracytoplasmic mucin; B: High dense collagen fibre; C: Well differentiated hyperplastic mature adipose cells with large fat vacuoles; D: Undifferentiated hyperplastic fibroblast cell with lower degree of collagen fibrous. H & E.

4. Discussion

The present study is the first observation on stomach tumour in S. longiceps. The stomach tumour infected S. longiceps abdomen was found to be distended. However, viral infection and gonad neoplasm also leads to abdomen swelling[16,17]. The stomach tumours appear as a radiodense hard tissue mass, which may result in a broader differential diagnosis[18]. Tumour infected S. longiceps has dense tissues mass than the normal one, which clearly reflects in the radiograph, where the X-rays transmit was higher in normal than tumour infected fish.

Previously, stomach tumours have been reported in harbor porpoise (Phocoena phocoena), blue shark (Prionace glauca) and Asian sea bass (Lates calcarifer)[11-13,19]. Gopalakrishnan et al. has reported skin tumour (0.38%) in S. longiceps, Parangipettai coastal waters[2]. In fish, reports on stomach tumours are meager than skin tumour, because of their distinctive appearance and imperceptible pathological nature. The Sphyraena jello odontoma prevalence is higher in monsoon[5]. Similarly, this present study also exhibited
the higher seasonal prevalence during monsoon. Summer and post-monsoon season fish landing was examined, but tumour infection was absent. The seasonal variations may influence the fluctuation of tumour prevalence. In addition, the present study showed higher prevalence in female fishes, but sex do not contribute the higher prevalence of stomach tumour. However, fish feeding behavior may influence. Other wise, the higher female proportion than the male in sardine population may reflect the higher tumour prevalence in female fishes.

Gopalakrishnan et al. have reported the divergence of skin tumour location, appearance, consistency and size[2]. Similarly in this present study, the stomach tumour appearance and size varied. The stomach tumour location and consistency did not varied, because external tumours were originated from almost of the region so their location and consistency was varied. In addition, this present study observed the meager gut content, because the swollen stomach was having pain, indigestion and inappetence. Therefore, the stomach tumour infected malnutritioned fish contain immature gonads.

In this present study, histopathological observation of sardine tumour lesions consists of four different hyperplastic epitheloid and non-epitheloid cell types including glandular epitheloid, myofibirl, adipocytes and fibroblast. On the basis of the gross and histopathological findings the tumours were diagnosed as gastric adenoma, myofibroblastoma, lipoma and fibrosarcoma. The tumour infection was only observed in stomach rather than internal organs. No evidence of local invasion and distinct metastases were observed in these cases.

Conflict of interest statement

We declare that we have no conflict of interest.

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