# Prevalence of Pathogenic Microorganisms in Two Commercially Important Marine Fishes of Nizampatnam, East Coast of India

KoteswaraRao V<sup>1</sup>, YedukondalaRao P<sup>1</sup>, JJ Babu Geddam<sup>2</sup>, Naveen Kumar R<sup>3</sup>, Bala Naveen Kannegalla<sup>5</sup>, Premchand K<sup>1</sup>, Prasad Undrajavarapu<sup>6</sup>

<sup>1</sup>Department. of Marine Living Resource, Andhra University, Visakhapatnam, Andhra Pradesh, INDIA

<sup>2</sup>Scientist – E, Clinical Division, National Institute of Nutrition (ICMR), Hyderabad, Telangana, INDIA

<sup>3</sup>Food and Drug Toxicology Research Centre, National Institute of Nutrition (ICMR), Hyderabad, Telangana, INDIA

<sup>4</sup>Manager (R &D),Dept. of Statistics, Parishodhan, Hyderabad, Telangana, INDIA

<sup>5</sup>Department of Biotechnology, AcharyaNagarjuna University, Guntur, Andhra Pradesh, INDIA

#### Abstract

The present study was under taken on microbiological quality of two commercially important marine fishes Upeneusvittatus and Sphyraenaflavicauda collected from nizampatnam, east coast of India. The skin, meat, intestine and gills of two fishes were analyzed for total bacterial count (TBC), Escherichia coli, Total fecal coliform (TFC), Staphylococcus aureus, Salmonella spp., Shigella spp., yeast and moulds count and vibrio spp. Isolation and identification of pathogenic bacteria was done according to US-FDA bacteriological analytical manual.

The result of the study shows that pathogenic bacteria were found in both fishes. The load of Vibrio spp. (4.2-4.6 log) and E. coli (4.3 log) were high when compared to other pathogenic bacteria. The fish samples found to contain high pathogenic bacterial load and necessary action should be taken to control the pathogenic microorganisms in these fishes before marketed.

**Keywords:** Upeneusvittatus, Sphyraenaflavicauda, microbial quality, TBC, TFCC, US-FDA, nizampatnam

# I. INTRODUCTION

Fish and Fish products are the most important source of proteins in human diet [1], It contains all ten essential amino acids which are useful to the entire human kind<sup>[2]</sup>. Fish is also a Vitamin and Mineral-rich food <sup>[3]</sup>. The pathogenic microorganisms present in the marine water normally contaminate the fish and causes undesirable changes in the fish. It is additionally viewed as that the nature of fish relies upon the nature and quality of water where fishes are collected and also the status of the fish landing centre. Microbial pollution on fishes depends on the condition that can be exchanged for sustenance items straight forwardly through contact by individual developments during cleaning procedure [4]. The contamination of microorganism in the interior fish organs results in breakdown of the immunological guard component framework<sup>[5]</sup>.

The aim of the study is to determine the microorganisms from skin, meat, intestine and gills of two commercial important fishes i.e., *Upeneusvittatus, Sphyraenaflavicauda*, of Nizampatnam fish landing centre.

# II. STUDY LOCATION

The fish samples were collected from the fish collection center of Nizampatnam situated at east coast of India (Figure-1). The fishes *Upeneusvittatus, Sphyraenaflavicauda*are available throughout the year and used for human consumption. The fish landing centre of Nizampatnam has modern facilities like electrical balances, insulated packing systems, etc.

# III. MATERIALS AND METHODS:

#### A. Sample Collection:

Upeneusvittatus (goat fish) and Sphyraenaflavicauda (barracuda) were collected in a sterile polythene bag from Nizampatnam fish landing centre and brought to the lab for further analysis in aseptic condition.



Figure 1: Sampling Area -Nizampatnam Outline Map

#### B. Microbial Analysis

The microbiological parameters were done according to US-FDA bacteriological analytical manual.

Microbial analysis was done for four parts of each fish. These parts are skin, meat, intestine and gills of fish. These parts are analyzed for Total bacterial count (TBC), *E. coli*, Total fecal coliform count (TFCC), Yeast and moulds, *Salmonella spp.*, *Shigellaspp and Vibrio* species.

#### C. Sample Preparation:

25 gram of each part meat, intestine of fish was added in to 225 ml of buffered peptone water. For microbial analysis of skin and gills, 1 g of the sample was added to 9 ml of buffered peptone water. The sample was mixed thoroughly in the buffered peptone water and 0.1ml of this sample was inoculated on to different selective media by spread plate method.

# IV. ENUMERATION, ISOLATION AND IDENTIFICATION OF ISOLATES

Different media was used for the isolation of different pathogenic bacteria. Nutrient agar for total bacterial count, Mac -Conkey agar for E. coli, manital salt agar for S.aureus, sabouraud dextrose agar for yeast and moulds, Salmonella Shigella agar for Salmonella spp., Shigella spp., Thiosulfate citrate bile salt (TCBS) agar for Vibrio spp. After the inoculation the plates were incubated at  $37^{\circ}$ c for 24-48h. For yeast and moulds the plates were incubated at room temperature for 48 hours.

# V. MICROBIAL COUNT

The mean number of colonies counted was expressed as log colony forming units (log10cfu/gm)

# VI. RESULT AND DISCUSSION

Microbiological quality of different parts of fishes is shown in Chart 1 & 2.

The Total Bacterial count range was more in Upeneusvittatus Skin, gills (log 4.6 to log 4.9) and lowest in meat, Intestine and Gills of Sphyraenaflavicauda. The bacterial flora of the fish depends on the environmental conditions where it was caught.

Indicator Organisms like *E. coli* and Total Fecal Coliform Count (TFCC), were found in both the fishes. The range of E.coliwas from log 3.3 to log 4.3 and it is higher in Sphyraena, upeneusskin. The population of E. coli was less in Sphyraena intestine. Staphylococcus aureuspopulation was ranged from log 0.00 tolog 3.7, and it is higher in Sphyranea skin when compared toUpeneus meat.

Almost, all the values of *E. coli* are exceeded the FSSAI limits, which is not more than  $20/\text{gm}^{[6]}$ , like that TFCC values are also exceeded the IAMS limits, which should not be more than  $100/\text{gm}^{[7]}$ . The

Presence of *E. coli* and fecal coliform interfere the quality of fish. The presence of *E. coli* in higher range indicates the contamination of the samples during handling and processing <sup>[8]</sup>.Fish harvested around fecal contaminated water can carry Salmonella[9]

The Present study shows that *yeast* and *moulds* contamination was more in *Upeneus*Skin than *Sphyranea*meat. The log value was ranged betweenlog 3.0 to log 3.9.

Salmonella spp., and Shigellaspp., were found in all samples which are highly pathogenic, this may be because of improper handling and processing at Nizampatnam fish landing centre. Salmonella population was ranged from log 3.5 to log 4.31 its contamination was more in Sphyraenaskin when compared to its meat. Shigella spp., ranged from log 3.0to log 4.4, where its population was more in Sphyraena intestine, gills and less in Upeneus skin.

*Vibrio* species was also found in all parts of the fish samples, According to the International Association of Microbiology Societies Fresh and Frozen fishes should be free from *Vibrio*<sup>[10]</sup>. The *Vibrio* population was more in *Sphyranea* skin and less in *Sphyranea* intestine and gills

# VII. CONCLUSION

Microbial Flora on Skin and Gills will transient the condition of the water and handling<sup>[11]</sup>. Organism in the intestine shows the contamination in digestive track. The good quality fish should have Total Bacterial count not more than 5, 00,000/gm as per the FAO. E. coli should not be exceeded 20cfu/gm. Total Fecal Coliform should not be exceeded more than 100cfu/gm as per FAO<sup>[12]</sup>. Plate count and coliform count have been used regulating microbial quality of the food. In the present study, Total Bacterial count readings are normal, but other pathogenic organisms are above the ICMSF guidelines<sup>[13]</sup>. To modify the situation, it is necessary to follow the code of practice while handling of the fish post harvesting procedures and maintain the cleanness in the fish landing centre.

The present study, provide the information about the pathogenic load of the fish parts which results in causing foodborne diseases.

#### ACKNOWLEDGEMENT

We would like to express our sincere thanks to the Department of Marine Living Resources, Andhra University, Visakhapatnam and Microbiology and Immunology Division, National Institute of Nutrition (ICMR), Tarnaka, Hyderabad for providing necessary facilities to carry out the research.

Chart 1: Microbial Population in Varies Parts of Upeneusvittatus Fish Sample



Chart 2: Microbial Population In Varies Parts of Sphyraneaflavicauda Fish Sample



# REFERENCE

- Varadharajan D., Pushparajan N., and Soundarapandin P., Fish Resources in Machilipatnam Coast. South East Coast of India, Inter. J. Pharma Biol. Archives. 3(4), 871-876 (2012).
- [2] Moghaddam, H.N., M.D. Mesgaran, H.J. Najafabadi and R.J. Najafabadi., Determination of Chemical Composition, Mineral Content and Protein quality of Iranian Kilka fish meal, Inter. J. Poult. Sci., 6: 354-361 (2007)
- [3] Asuquo, F. E., I. Ewa-oboho, E.F. Asuquo and P.J. Udo, Fish species used as biomarkers for heavy metal and Hydrocarbon contamination for cross river, Nigeria, The Environ, 24: 29-26, (2004)
- [4] Boyd R.F., General Microbiology, Published by times Mirror/Mosby College, 459-461. (1984)
- [5] Gillespie L.A., Adak G.K., O Brien S.J., Brett M.M. and Bolton F.J., General Outbreaks of infectious intestinal disease associated with fish and shellfish, England and Wales, 1992-1999, Common Dis. Public Health, 4, 117-123 (2001)
- [6] http://www.fssai.gov.in/home/food-standards/fssregulations.html. Accessed on 15th March 2017.
- [7] International association of microbiological societies (IAMS), (www.microbialstandard.com), 1962
- [8] PrithwirajJha, Rudra Prasad Roy and Bharat S. Application of sensory and microbial analysis of assess quality of fish in Siliguri city, West Bengal, India. J. Env. Biol. 31(5) 587-594 (2010)
- ShashiKiran, MunishBatra, Umapati V Jitentdra Kumar and M.K. SaxenaDetection of Virulence in Salmonella Typhimurium Isolated from Ganga Water, pp. 363-366, (2009)
- [10] Hadin E.L., Radu S., Chen C.H. and Nishibuchi M, Prevalence of potentially pathogenic Vibrio species in the seafood marketed in Malaysia, J. Food Prot, 67(7) 1469-1475 (2004)
- [11] Marin M.cahill,Bacterial flora of fishes A review, microbial ecology, volume 19 issue 1, pp 21-41(1990)
- [12] Food and Agricultural Organization (FAO) Food Safety Regulations Applied to Fish by the Major Importing Countries, FAO Fisheries Circular No. 825 FAO, Rome, (1989)
- [13] International Commission on Microbiological specification for food (ICMSF), Sampling Plan for fish and shellfish, in: Microorganism in foods. Sampling for Microbiological Analysis: Principles and Scientific Applications, 2(2) University of Toronto Press, Toronto, Canada, 181-1996 (1986)