Incidence of human pathogenic bacteria in shrimp feeds - A study from India

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Abstract

The incidence of various human pathogenic bacteria in commercially available and home-made shrimp feeds used on some farms in India was analyzed. The Total Heterotrophic Bacteria in the commercial feed samples ranged between 10^3–10^5 cfu g^{-1} and those in the farm-made feeds between 10^6-10^7 cfu g^{-1}. No bacteria of significance to human health were found to be associated with any of the commercial feed samples analyzed, while farm-made feeds analyzed during the study showed a high incidence of various human pathogens such as *Vibrio parahaemolyticus*, *V. cholerae*, *Escherichia coli* and *Staphylococcus aureus*. Possible modes of contamination in feeds and ways to prevent them are discussed.

Introduction

Of the various hazards related to cultured aquatic products, the most important are the ones that are associated with human pathogenic bacteria. A number of pathogenic and potentially pathogenic bacteria belonging to the genus *Vibrio*, *Staphylococcus*, *Escherichia* and *Salmonella* have been isolated from cultured shrimps and their environments in India (Ahmed et al. 1995; Sugumar et al. 2001). In most instances, contamination has been attributed to the poor quality of source water used in the farms, pollution due to sewage and also the improper and unhygienic handling of shrimps by the farm workers. However, another possible reason for contamination could be due to the increased incidence of various human pathogenic bacteria in aquaculture feeds used in the culture systems. The use of wet or moist animal-based feeds has been identified as one of the factors that could be responsible for the increasing incidence of food safety hazards in aquaculture (Reilly et al. 1998). However, this factor has been overlooked by many researchers and hence, not much information is available with respect to the Indian scenario. The present study is an attempt to analyze the incidence of human pathogenic bacteria in commercial and farm-made feeds used in the various shrimp culture systems of south India to ascertain the role of feeds as possible vectors of these bacteria into the culture systems.

Bacteriological analysis

Samples of commercial and farm-made feeds used in the various shrimp culture systems of south India (Table 1) were obtained from the respective farm sites. Samples of commercial pelleted feed were taken from the feed bags, while farm-made feed samples were collected after they were prepared at the farm site. They were then transferred to sterile polythene bags and taken to the laboratory where the bacteriological analyses were carried out as per the methods outlined in the USFDA (1995). The Total Heterotrophic Bacteria (THB) in the feed samples was enumerated by the agar pour plate technique and results were obtained after 48 hours of incubation at 37° C. Specific agars were then used to isolate various pathogenic strains such as Thiosulphate Citrate Bile Sucrose (TCBS) agar for *Vibrio*, Tergitol-7 for *Escherichia*, Baird Parkers (BP) agar medium for *Staphylococcus*, Brilliant Green and/or Bismuth Sulphite agar medium for *Salmonella* and KF agar for *Streptococcus*. The plates were then inoculated and incubated at 37° C overnight and up to seven days. Specific colonies were then sub-cultured and the pure culture was used for further biochemical tests (Sneath and Holt 1986; Alsina and Blanch 1994).

Table 1. Details of different feed samples used for the study

Sample	Type /Nature of feed	Specification	Culture system/Location/Coast
A	Commercial	Grower	Semi-intensive-Andhra Pradesh -E. Coast
B	Commercial	Finisher	Semi-intensive- Andhra Pradesh - E. Coast
C	Commercial	Starter	Semi-intensive- Andhra Pradesh - E. Coast
D	Commercial	Grower	Semi-intensive- Tamil Nadu - E. Coast
E	Farm-made	-	Traditional – Kerala-W. Coast
F	Farm-made	-	Traditional – Kerala-W. Coast

Table 2. Total Heterotrophic Bacteria associated with the shrimp feed samples analyzed

Feed sample	THB cfu g^{-1}
A	4.18×10^3
B	4.01×10^5
C	5.22×10^3
D	4.68×10^3
E	3.78×10^6
F	5.01×10^7

Results

The THB in the commercial shrimp feeds ranged from 10^3 to 10^5 cfu g^{-1} (Table 2). No human bacterial pathogens were isolated from the commercial shrimp feeds. However, with regard to farm-made shrimp feeds, a higher bacterial load of 10^6 to 10^7 cfu g^{-1} (Table 2) was observed together with the incidence of various pathogens such as *Staphylococcus*, *Vibrio* and *Escherichia* (Table 3).

Discussion

Studies have shown that commercial fish feeds contain a mixed microflora, of which many are pathogenic or potentially pathogenic to fish and human (Trust 1971; Trust and Money 1972). The major bacterial genera associated with commercially available shrimp feeds were *Flavobacterium, Bacillus, Micrococcus* and *Alcaligenes* (Rajeev et al. Unpubl). Commercial feeds are considered unsatisfactory if they contain large bacterial populations even if the bacteria are non-pathogenic in nature and have not caused any changes to the pellet (Thatcher and Clarke 1968). Reports also indicate that the bacterial composition becomes evident in feeds containing 10^6 to 10^8 microorganisms/g. In the present study, the THB in the commercial feed samples was in the range of 10^3 to 10^5 cfu g^{-1} indicating that the bacterial counts were well within permissible limits.

The farm-made feeds analyzed showed a higher bacterial load of 10^6 to 10^7 cfu g^{-1} indicating the advent of bacterial contamination. It is known that aquaculture feeds that contain higher levels of moisture are prone to faster microbial spoilage due to the action of various bacterial enzymes (Jones 1987). Most of the farm-made feeds analyzed were wet feeds prepared from animal-based raw materials such as prawn meal, shrimp-head meal, fishmeal and clam meat which usually contain very high levels of moisture. This would have contributed to the higher bacterial loads as observed in these feed samples.

Public health bacteria, including *Escherichia coli* (Niemi and Taipalinen 1982), *Salmonella* (Trust 1971) and *Clostridium boutulinum* (Lalitha 1998; Sugita et al. 1989) have been reported in fish feeds. However, none of the samples of commercial shrimp feeds analyzed showed the presence of any such human bacterial pathogens. Modern and hygienic methods of feed processing and improved feed management practices at the farm sites have greatly reduced the incidence of such bacteria in shrimp feeds.

Trust and Wood (1973) had suggested that the ingredients used in feed formulation partially determine the nature of its microflora. Farm-made feeds used in the traditional shrimp culture systems are generally made from marine animal ingredients such as prawn meal, fishmeal and clam meat. These feed ingredients were mainly harvested and/or collected from contaminated coastal and estuarine waters and from unhygienic and poorly maintained landing centers and pre-processing units. Contamination by bacteria such as *Vibrio cholerae*, *V. parahaemolyticus*, *E. coli* and *Staphylococcus aureus* in one or more of these marine animal raw materials used for feed preparation would have led to the contamination in the final products also. The presence of such pathogenic bacteria has been reported in the Cochin backwaters, which is the main water source for the culture farms, and also the principal area for harvesting the raw materials used in feed preparation (Pradeep and Lakshmanaperumalasamy 1986a, b and Gore et al. 1979). In addition, the rather unhygienic handling practices followed by the workers at the farm site could also be a possible explanation for the contamination by indicator bacteria such as *S. aureus* and *E. coli*.

There is little awareness of the importance of good feed management practices in shrimp culture among traditional farmers. This has resulted in the use of poor quality raw materials and unhygienic feed preparation, which ultimately have led to the increased incidence of many human bacterial pathogens in aquaculture feeds.

From the results of the present investigation, it may be inferred that commercially available shrimp feeds used in the grow-out farms of south India are free from public health bacteria. However, farm-made feeds prepared from marine animal-based raw materials, especially those which are commonly used in traditional shrimp farms, are in most cases contaminated with various species of human pathogenic bacteria. Poor quality raw materials used for feed preparation and low personal hygiene at the farm sites have been found to be the main causes associated with the bacterial contamination in feeds. The importance of using good quality raw materials,

Table 3. Presence of human pathogenic bacteria in shrimp feed samples

Sample	*Vibrio cholerae*	*Vibrio parahaemolyticus*	*Salmonella typhi*	*Staphylococcus aureus*	*Escherichia coli*	*Streptococcus faecalis*
A	-	-	-	-	-	-
B	-	-	-	-	-	-
C	-	-	-	-	-	-
D	-	-	-	-	-	-
E	-	+	-	+	+	-
F	+	+	-	+	+	+

(+ present, - absent)

ensuring their hygienic storage and adoption of hygienic on-farm feed preparation techniques should be extended to traditional shrimp farmers to decrease the incidence of various pathogenic bacteria in shrimp feeds. These factors will reduce environmental pollution, enhance shrimp health and eliminate potential food safety hazards to humans.

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