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POSTHARVEST EFFECT ON *VIBRIO* SPP. IN SHRIMP (*PENAEUS* SPP.) SOLD BY VENDORS IN TRINIDAD, W. I.

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ABSTRACT: Postharvest processing (PPH) methods have been determined to reduce *Vibrio* spp. to non-detectable levels, and include cool pasteurization, cryogenic individual quick freezing (IQF) with extended storage, high hydrostatic pressure (HHP) processing, low-dose gamma irradiation and high-salinity treatment. In Trinidad and Tobago, glazing or direct contact with ice is another common process used by shrimp vendors that could be useful to minimise occurrence of *Vibrio* spp. in marine shrimp (*Penaeus* spp.). Sixty glazed shrimp composites were purchased from the five largest depots in Trinidad and were analysed for *Vibrio* spp. using a slightly modified methodology outlined in the US FDA Bacteriological Analytical Manual. The absence of *Vibrio* in the shrimp met international and local human consumption standards. In this study, *Vibrio* spp. was the only bacteria of natural inhabitants to seawater, especially in warm areas, that can contaminate live fish and shellfish. Direct contact with ice to the warm water shrimp by vendors avoided the survival and recovery of *Vibrio* spp. Rapid cooling of the shrimp by glazing can injure *Vibrio* and thus minimise public health concerns.

Keywords: high hydrostatic pressure, public health, marine, contamination.

Introduction

Seafoods, especially shellfish, could readily be contaminated with pathogenic microorganisms such as *Vibrio* because of the texture of their flesh and also their living habits in the microbe-laden habitats that they occupy (Colakoglu, Sarmasik, and Koseoglu 2006). Previous studies have reported *Vibrio* in a variety of shellfish that included shrimp, mussels and prawns from Turkey, Croatia, China and Taiwan (Fang, Huang, and Chen 1987; Strom and Paranjpye 2000; Jaksic et al. 2002; Yano et al. 2004; Colakoglu, Sarmasik, and Koseoglu 2006). Postharvest processing (PPH) methods have been determined to reduce *Vibrio* spp. to non-detectable levels, and include cool pasteurization, cryogenic individual quick freezing (IQF) with extended storage, high hydrostatic pressure (HHP) processing, low-dose gamma irradiation and high-salinity treatment (US FDA 2011). In Trinidad and Tobago, glazing or direct contact with ice is another common process used by shrimp vendors that could be useful to minimise occurrence of *Vibrio* spp. in marine shrimp (*Penaeus* spp.).

Materials and Methods

Sixty glazed shrimp composite samples were purchased from the five largest depots in Trinidad in 2009. Each shrimp composite of 908 g obtained from each vendor was placed into sterile bags and within two hours of purchase were transported to the Microbiology Laboratory in the Department of Food Production at The University of the West Indies (UWI) in an ice cooler to maintain a temperature of approximately

4°C. Samples were processed within 30 minutes of arrival at laboratory for *Vibrio* spp. using a slightly modified methodology outlined in the US FDA Bacteriological Analytical Manual.

A slightly modified version of the spread plate technique outlined by Kaysner and DePaola (2004) was used for this analysis. Twenty-five grams (25 g) of shrimp were homogenised in a Waring blender (Connecticut, U.S.A) with 225 mL alkaine peptone water (APW) that contained an additional 2% sodium chloride. Serial dilutions were made from 10^{-1} to 10^{-7} and then incubated for 6h at 37°C.

After incubation, a loopful (approximately 3-mm) of the APW culture was streaked from the surface pellicle to triplicate TCBS (Oxoid) plates. The plates were allowed to cool, inverted and incubated for 18-24 hours at 37°C. Plates containing between 30-300 colonies were counted. Presumptive yellow and green colonies of *Vibrio* were picked and streaked onto PCA plates with an additional 2% sodium chloride. Gram stain followed and biochemical tests for IMViC, oxidase, growth in (0,3, 6, 8 and 10) % sodium chloride, D-mannitol, sucrose, lactose and arabinose respectively. Water and agar controls were carried out in triplicate.

Results and Discussion

The findings revealed that no *Vibrio* was present in the shrimp that met the International Commission for the Microbiological Specification of Food (ICMSF 1986) and the Trinidad and Tobago Food and Drugs (2007) standard for human consumption. In this study, the *Vibrio* spp was the only bacteria of natural inhabitants to seawater especially in warm areas that can contaminate live fish and shellfish. Direct contact with ice to the warm water shrimp by vendors avoided the survival and recovery of *Vibrio* spp. Rapid cooling of the shrimp by direct contact with ice can injure *Vibrio* and thus minimise public health concerns.

Vibrio spp. include gram-negative, facultative anaerobic, non-spore-forming bacilli which are oxidase positive and halophilic (Jaksic et al. 2002). *Vibrio,* such as *V. damsel*, *V. alginolyticus*, *V. fluvialis, V. vulnificus* and *V. parahaemolyticus,* are indigenous to the marine environment and shrimp (Murdoch 1993; Hosseini et al. 2004; Su and Liu 2007) but the most commonly encountered organisms of this group were *V. cholerae* and *V. parahaemolyticus*, usually associated with enteric disease (Warnock III and MacMath 1993). The routes of transmission from the environment to man include consumption of undercooked or raw seafood or shellfish (Jaksic et al. 2002).

Vibrio, a Concern in Seafood Outbreaks and Illnesses

Vibrio parahaemolyticus is a common cause of foodborne illnesses in many Asian countries, including China, Japan and Taiwan, and is recognized as the leading cause of human gastroenteritis associated with seafood consumption in the United States (Su and Liu 2007). In Taiwan, during the period 1986 to 1995, the three most common bacteria involved in food-borne disease outbreaks were *Vibrio parahaemolyticus* (35% of 555 outbreaks), *Staphylococcus aureus* (30% of 555 outbreaks), and *Bacillus cereus* (18% of 555 outbreaks) (Pan et al. 1997). Individuals

who are at the greatest risk of serious illness and mortality from water and foodborne enteric microorganisms include the very young, the elderly, pregnant women, and the immunocompromised (Gerba, Rose, and Hass 1996).

Outbreaks of *Vibrio parahaemolyticus* gastrointestinal illness occurred on two Caribbean cruise ships in the late 1974 and early 1975, affecting 697 passengers and 27 crew members (Lawrence et al. 1979). Epidemiologic evidence incriminated seafoods served on the ships as the vehicles of transmission. Similarly, in Washington, during the 3-month period from late summer to fall of 1981, investigations revealed that six sporadic gastrointestinal cases reported to the public health agencies were due to *Vibrio parahaemolyticus* and was associated with eating raw oysters that were harvested at Willapa Bay (Nolan et al. 1984).

Shellfish and shrimp have also been incriminated in cholera outbreaks in many countries including USA, Philippines, Malaysia, Gilbert Island and Sardinia (Dutt, Alwi, and Velauthan 1971; McIntyre et al. 1979; Kuberski, Flood and Tera 1979; Salmaso et al. 1980; Pavia et al. 1987; Rabbani and Greenough III 1999). The largest outbreak was a pandemic in South America in the early 1990s when *V. cholerae* O1 caused more than 400,000 illnesses and 4,000 deaths, in Peru; lightly fermented fish, ceviche, was the cause of the outbreak (Wolfe 1992; Suárez and Bradford 1993). The cholera disease is caused by infection of the small intestine by *Vibrio cholerae* O1 and O139 and is characterized by massive acute diarrhoea, vomiting, and dehydration; death occurs in severe, untreated cases (Rabbani and Greenough III 1999).

Vibrio vulnificus which occurs naturally in warm ocean waters (Murdoch 1993) has been identified as a cause of severe seafood-related illnesses among individuals with liver dysfunction, with a mortality rate of >40% (Doyle 1994). This pathogen can also cause sickness and death in persons with HIV-AIDS, chronic renal insufficiency, cancer, diabetes, steroid dependent asthma, and chronic intestinal disease, according to the U.S. Public Health Service (Murdoch 1993). *V. vulnificus* is an opportunistic human pathogen representing the leading cause of seafood fatality in the United States (Wong et al. 2005).

Conclusion

It is well-established worldwide that *Vibrio* have been suspected in seafood outbreaks and caused health concerns to consumers. Hence, postharvest processing (PPH) methods such as glazing should be established for *Vibrio* to prevent potentially hazardous food-borne risk to consumers.

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