Antibiotic resistance associated with aquaculture in Vietnam



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The fishery sector is an important source of income, employment and food supply in Vietnam. In 2014, Vietnam was ranked the world's fourth largest exporter and the third largest producer of farmed food fish. Vietnam seafood export has attained the value of over US$6.0 billion since 2011 and reached a peak of US$7.9 billion in 2014. However, many problems and diseases confront sustainable development of the fishery sector and overuse of antibiotics is considered a major challenge. Antibiotics are used in aquaculture for both therapeutic and prophylactic reasons. Various antimicrobials used in human medicine are also used for food animals even for non-therapeutic use. The use of antibiotics in health management of aquaculture farming is of great concern due to possible residues in aquatic products and in the development of antibiotic resistance. In 2005 the Vietnamese government first promulgated a list of medicines, chemicals and antibiotics that are banned or limited for use in aquaculture and this is regularly updated and amended to tackle the growing problem of antibiotic resistance.

The fishery sector in Vietnam

Fisheries is a key national economic sector that produces 4–5% GDP and 5–6% of the total national turnover of Vietnam. Aquaculture in Vietnam has changed from a self-sufficient sector in the early 1960s into a concentrated commodity production and this model has become the most important contributor in the fishery sector since 2007. In Vietnam, the Mekong River Delta in the south and the Red River Delta in the north have been the major areas for wild caught fishing as well as intensive aquaculture farming for decades. Aquaculture production in the Mekong Delta has contributed more than 70% of the country's total production, in which catfish (fresh water) and black tiger and white leg shrimp (brackish water) are the three most important commercial species grown (Figures 1, 2). Since the 1990s, fishery products have been the third biggest export commodity of the country. Fishery exports earned over US$1 billion in 2000, US$3 billion in 2006, US$6 billion in 2011 and reached a peak of US$7.9 billion in 2014. Currently, Vietnamese fishery products are exported to more than 140 countries and territories in five continents. According to the Food and Agriculture Organisation of the United Nations (FAO), nearly 10% of the population in Vietnam obtains their main income from Fisheries. More than four million workers including a large proportion of women are employed in aquaculture and fishery activities (Figure 3).

Antibiotic resistance in aquaculture in Vietnam

In Vietnam, only limited studies on antibiotic uses and resistance in aquaculture were conducted and/or published before 2005. A study on antibiotic residues and resistance against Norfloxacin (NOR), Oxolinic acid (OXO), Trimethoprim (TRI) and Sulfamethoxazole

Figure 1. Seabass or Barramundi (*Lates calcarifer*) farming in Vietnam.

Figure 2. Tra catfish feeding in a farm in the Mekong Delta, Vietnam.

(SUL) has been conducted in four shrimp farming locations (Thai Binh, Nam Dinh, Can Gio-Ho Chi Minh City and Ca Mau provinces) in mangrove areas in Vietnam in 2003[1]. The results showed the presence of antibiotic residues and a high incidence of bacterial resistance to the four antibiotics observed in these shrimp ponds in different mangrove areas in Vietnam. Another study on antibiotic resistance of 123 bacterial isolates (from water, sediment and different fish farms (catfish, tilapia, common carp and gourami) in five provinces in the Mekong Delta) found that 90% of the isolates were resistant to Tetracycline (TET), 76% to Ampicillin (AMP), 100% to Chloramphenicol (CHL), 65% to Nitrofurantoin (NIT) and 89% to Trimethoprim-Sulphamethoxazole (SXT).

A number of extensive research projects on antibiotic resistance of commensal bacteria from several aquaculture species have been conducted by our groups. In 2003, a study on the resistance of 101 isolates (belonging to the family Enterobacteriaceae) from frozen catfish of the Mekong Delta showed high frequencies of resistance (80–100%) to Oxytetracycline (OTE), CHL, Streptomycin (STR), SXT and AMP (unpublished data). The levels of multiple resistance were also high, of which 100% of isolates were resistant to at least four antibiotics. A research study published in 2005 reported the resistance of 92 isolates (including Enterobacteriaceae (49.1%), Pseudomonas spp. (35.2%) and Vibrionaceae (15.7%) families) from three catfish farms in the Mekong Delta[2]. Rather high incidences

Figure 3. Tra catfish filleting in a seafood processing company in the Mekong Delta, Vietnam.

(20–60%) of resistance to OTE, CHL, SXT, NIT, Nalidixic acid (NAL) and AMP were observed. The multiple antibiotic resistance (MAR) index values indicated that the three farms corresponded to high-risk exposed-antibiotic sources. In a recent publication, a larger collection of samples (catfish, water and sediment) was collected from 50 catfish culture ponds (belonging to 15 catfish intensive farms) in the Mekong Delta, Vietnam[3]. All 116 *Pseudomonas* spp. and 92 *Aeromonas* spp. isolates recovered from these samples were tested for their resistance to 13 antimicrobial agents belonging to eight families. High levels of resistance to AMP, SXT, and NAL were observed. Lower frequencies of resistance occurred against Ciprofloxacin (CIP), NOR, TET, Doxycycline (DOX), Gentamycin (GEN), Neomycin (NEO), STR, and Kanamycin (KAN). The percentages of multiple resistance (resistance to three or more antimicrobial classes) was 96.6% for *Pseudomonas* and 61.9% for *Aeromonas* spp. The MAR indices showed that *Pseudomonas* and *Aeromonas* isolates were high-risk sources of contamination where antibiotics were commonly used. The level of resistance to CHL and NIT was still high despite these antibiotics being banned for use in aquaculture in Vietnam since 2005, and this resistance is possibly due to the co-transfer of resistance genes. Our recent study on the resistance of 167 *Vibrio* spp. isolated from blood cockles (16 farms of the six provinces in the Mekong Delta) showed a low incidence of resistance to nine tested antibiotics (TET, NAL, NOR, CIP, SXT, NIT, CHL, STR, KAN), with a high level of resistance to AMP only. Unfortunately, very high levels of resistance of bacteria in aquaculture environments or products have also been reported in many countries which have intensive aquaculture activities such as China[4], USA[5], Thailand[6], India[7], Indonesia[8], Australia[9], Italy[10] and Chile[11] implying the heavy use of antibiotics in aquaculture worldwide and this provides a warning on the effects of antibiotic usage in aquatic ecosystems, and the world wide emergence of antibiotic resistance in aquatic bacteria.

Integrons and the transferability of antibiotic resistant genes

In bacteria, horizontal gene transfer is the principle mechanism responsible for the spread of antibiotic resistance genes[12]. Horizontal gene transfer is facilitated by mobile genetic elements such as conjugative plasmids, transposons, and phages[13]. Integrons are not themselves mobile elements but are associated with mobile genetic elements (transposons or conjugative plasmids) enabling efficient intra- or interspecies transmission[14]. In Vietnam, studies on integrons and resistance gene transfer mechanisms have been conducted in clinical isolates[1,15–19]. However, very limited studies have been published on non-human isolates such as food-borne pathogens[20], animal pathogens[19] and catfish pathogens[21]. Recently we have done an extensive study on integrons and the transferability of antibiotic resistant genes of commensal bacteria in catfish aquaculture in Vietnam[3]. We found that the commensal isolates of catfish harbour a pool of mobile genetic elements such as plasmids and integrons, which contain various antibiotic resistance gene cassettes. Conjugation and transformation experiments demonstrated the successful transfer of all or part of the resistance phenotypes of isolates to the recipient strains of different genera and sources. Class 1 integrons associated with plasmids have facilitated the emergence and dissemination of antibiotic resistance in aquaculture environments. The transformation and conjugation experiments also indicated that CHL resistance phenotype was co-transferred in association with SXT, AMP, TET resistance phenotypes. In addition, the CHL resistance genes (*catB*) have often been associated with class 1 integrons, therefore, the co-selection is important for CHL resistance dissemination. This suggests that, in the absence of chloramphenicol selection pressure, CHL resistance is co-transferred and maintained due to gene linkage to genes encoding resistance to antimicrobials that are widely used in food animals or aquaculture[22]. The other studies worldwide also indicate that aquatic environments or products are reservoirs of antibiotic resistance and class 1 integrons have facilitated the dissemination of antibiotic resistant genes such as in the USA[23], China[24], Thailand[25], Northern Europe and North America[26], Europe, Japan and the USA[27], and Japan[28].

Conclusion

The high frequencies of resistance observed in aquaculture bacterial isolates in Vietnam, and in worldwide aquatic environmental and product isolates, reflect the global spread of resistance due to extensive use of antibiotics in aquaculture. Aquaculture resident bacteria could serve as a reservoir of resistant genes if they harbour a pool of mobile genetic elements that can readily be transferred intra- and interspecies.

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