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Acute Hepatopancreatic Necrosis affecting penaeid shrimp culture in Mexico

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— Abstract—

Emerging diseases that affect crops on shrimp farms cause significant economic losses to aquaculturers. In most cases, good management practices are not implemented in production systems, because of the established biosecurity standards that are not adequate ones, favoring the introduction and dispersion of pathogenic organisms, such as the case of Acute Hepatopancreatic Necrosis (AHPND), an infection that appeared at the end of 2013 in shrimp crops in Mexico. It also analyzes and discusses the impacts and the control measures that this transboundary disease faces in the Mexican territory from the outbreak.

Keywords:

Shrimp farming; biological contingency; AHPND

For the world's aquaculturists, dedicated to shrimp fattening, diseases are a real threat in terms of economic and social impacts, their appearance is related to the recent increase in globalization and the commercial volume of the aquaculture sector that it has created new market opportunities for farmed aquatic animals, but, simultaneously, they have facilitated new mechanisms, by which these microorganisms can spread to new areas, thus, aquatic animal diseases are one of the most serious limitations for expansion and the development of sustainable aquaculture. These transboundary diseases are usually caused by viruses, but the pathogen can also be a bacteria or a parasite (FAO, 2020), leading to a considerable decrease in the records of farmed shrimp production.

Addressing this problem in a particular way, regionalized in those farms where its presence has been manifested and verified, it is urgent to start programs for the prevention of local diseases to eliminate the vertical transmission of these pathogens. Shrimp farming is facing a great challenge regarding the development of control and prevention strategies for this pathology, at present AHPND is caused by at least two different species of Vibrio, this opens the possibility of spreading to other species, therefore molecular diagnostic techniques must undergo constant sensitivity and specificity evaluations (Varela *et al.*, 2017).

This document presents the official aquaculture health mechanisms adopted to attend the biological contingency due to the appearance of Acute Hepatopancreatic Necrosis (AHPND) and the results obtained from its application.

RECENT ACUTE HEPATOPANCREATIC NECROSIS

AHPND represents a real threat, a team of researchers from the University of Arizona has managed to isolate the strain and use it to infect healthy shrimp with AHPND, and this is the method known as Koch's postulates (FAO, 2013). There are documented reports that AHPND initially appeared in 2009. It is a new disease that affected shrimp farms located in southern China and Hainan Island in 2010. The disease manifests itself within 20 or 30 days after being introduced to the fattening ponds with the postlarvae, the clinical signs include lethargy, reduced hepatopancreas, red and empty intestine, soft and dark exoskeletons, and spots on the shell with affectations to the species, both *Penaeus monodon* and *Litopenaeus vannamei* present the same pathology (Lightner, 2012).

During 2010 and 2011, similar mortalities were recorded in Vietnam and Malaysia; these new cases shared some characteristics with what happened in 2009. Later in 2012, Thailand was affected by AHPND with farmed shrimp mortalities of 20-30%. In Latin America and particularly in the shrimp farms of the Mexican Republic, until that date there were no official reports on the incidence of the disease, however, in August 2013, Dr. Donald Lightner confirmed the presence of this disease at the "Sixth Meeting of the Inter-American Committee on Aquatic Animal Health", carried out in Yucatán, Mexico (Sánchez *et al.*, 2014).

The impact by AHPND in Mexico has been critical with a decrease in the production rates of farmed shrimp in the order of 60%, from 100,321 t of live weight produced in 2012 to 60,292 t in 2013, a partial recovery for 2014 with 86,950 t recorded and an extraordinary rebound in 2015 with 130, 344 t reported by the production units (Figure 1).

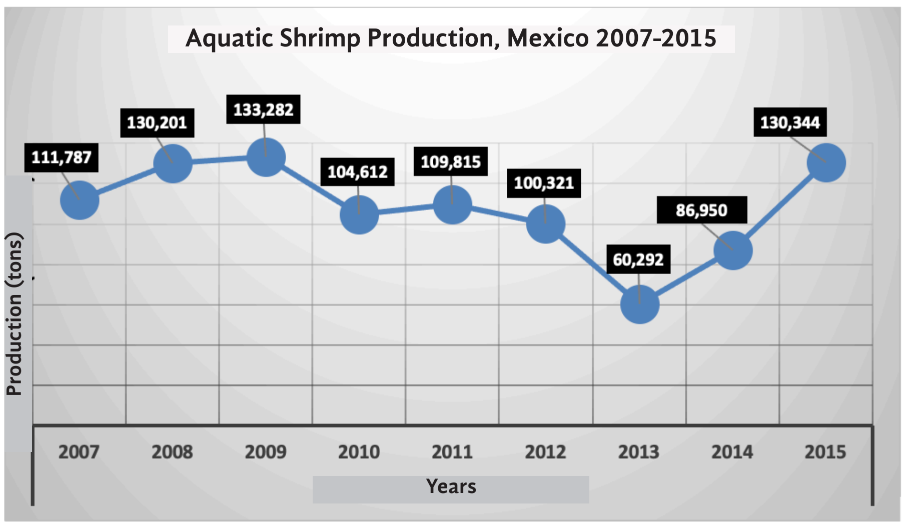

Figure 1. Shrimp aquaculture production in Mexico (Tons / Year live weight). Source: Yearbook: Fishery And Aquaculture Statistics. (CONAPESCA, 2007- 2015)

The causative agent is attributed to the pathogenic strain of Vibrio parahaemolyticus, which is introduced orally through the debris found in the water column and the bottom of the pond, colonizing the digestive tract, producing toxins and causing in the acute phase a hepatopancreas (HP) cell dysfunction where there is cell destruction, in addition, they produce the detachment of tubular epithelial cells, hemolytic inflammation and very marked HP necrosis. In the terminal phase, in addition to the shedding of epithelial cells, a massive bacterial secondary infection occurs (Navarro, *et al.*, 2013). It is an enteric bacterium whose natural habitat is marine waters since they require salt for their development (Rodríguez, *et al.*, 2014).

ACTIVATION OF THE NATIONAL AQUACULTURE HEALTH EMERGENCY DEVICE

The biological contingency was initially presented in three states in the northwest of the Mexican Republic, as part of the efforts to contain it on August 22, 2013, it was published in the Official Federal Gazette (DOF. 22.08.2013), the agreement that implements the national aquaculture health emergency device, in the terms of article 116 of the General Law on Sustainable Fisheries and Aquaculture, to control and eradicate the occurrence of atypical mortalities in shrimp production units in the states of Nayarit, Sinaloa, and Sonora, having validity of six months from its entry into force. The National Service of Agri-Food Health and Quality (SENASICA) coordinated these activities through the Directorate of Animal Health, establishing sanitary actions in the territory that includes the aforementioned states, including those as in the particular case of the state of Chiapas, where the AHPND disease spread.

In response to this emergency, in Mexico, a series of strategies and zoosanitary measures of urgent and coordinated application was established for the diagnosis, prevention, control, and eradication of the occurrence of atypical mortalities in the production units of shrimp farms (DOF 22.08.2013) that initially affected these three states. The lines of action were defined with the following biosafety measures:

- Control of the mobilization of products, by-products, and inputs for shrimp farming.
- Quarantine and elimination of organisms in which the presence of this etiological agent has been detected.
- Producer training.

In terms of economic impact, the most important diseases that affect the cultivation of shrimp belonging to the *peneidae* family in Asia, the Indo-Pacific region, and in America, are of infectious origin, among these, the most relevant are caused by viruses, protozoa, and bacteria (Table 1). The pandemics caused by the shrimp viruses of this family, such as the White Spot, Taura Syndrome, to a lesser degree the Infectious Hypodermal and Hematopoietic Necrosis Virus, and the Yellowhead disease have caused the shrimp industry to lose crops, jobs, and income from exports (Lightner & Pantoja, 2001) and more recently from the appearance of AHPND.

Table 1
Pathogens that affect shrimp cultures, according to their danger

Pathogen		Pathogen type	Category (Dangerousness)
Virus	TSV	Taura virus	C-1
	WSSV	White Spot Syndrome Virus	C-1
	YHV	Yellowhead virus	C-1
	IHHNV	Infectious haematopoietic and hypodermal necrosis virus	C-2
	BP	Bacauolovirus penaei	C-2
	MBV	Bacuolovirus of the P. *monodon* type	C-2
	BMN	Baculoviral intestinal necrosis	C-2
	HPV	Enteric parvovirus	C-2
Protozoa		Microspora	C-2
		Gregarious	C-3
Bacterias	NHP	Necrotizing hepatopancreatitis	C-2
		Vibrio sp.	C-3
	AHPND	Acute Hepatopancreatic Necrosis (*Vibrio parahaemolyticus*)	C-1*

* Of recent appearance in Mexico (2013)

Source: Modified table obtained from Lightner (2001) and Leyva (2010)

The pathogens considered within category C-1 are those that due to their effects on organisms and their high infectivity have the potential to produce catastrophic losses within cultivation systems (80% to 100%). While the pathogens in category C-2 show the ability to produce high mortalities in low shrimp cultures. In category C-3, pathogens that have minimal effects on crops are listed and in some cases, there are measures to eliminate them. The dangerousness of these pathogens on the list is determined by their virulence and potential to produce mortalities in aquaculture systems, however, many of the less dangerous pathogens can also cause economic losses due to the difficulty in marketing the product, since in some cases it can cause deformity, erosion of the shrimp cuticle or low growth of the organisms (Leyva, *et al*., 2010).

The shrimp farms established in the state of Chiapas, in the southeast of Mexico, have not been the exception to this problem, during the March-July 2014 production cycle, particularly in the El Fortín shrimp farm, with an area of 76 ha. of ponds for culture, there were accumulated mortality rates of 95%, according to fresh shrimp samples sent to diagnostic laboratories by the Comité Estatal de Sanidad Acuícola de Chiapas (CESACH), the results indicated that the cause of the infectious outbreak it would have been caused by the bacterium *Vibrio parahaemolyticus* (Figure 2) where clinical signs of the disease were observed (pale hepatopancreas, empty intestines, tubular deformation in hepatopancreas, expanded and previously red chromatophores), as a consequence of the sowing of contaminated shrimp postlarvae with this bacterium from a laboratory established in the state of Sinaloa, located in northwestern Mexico, the region where the infectious outbreak originated. Given the degree of damage that occurred in the El Fortín farm, the application of the national aquaculture emergency device was immediately implemented, establishing technical restrictions for its control and eradication.

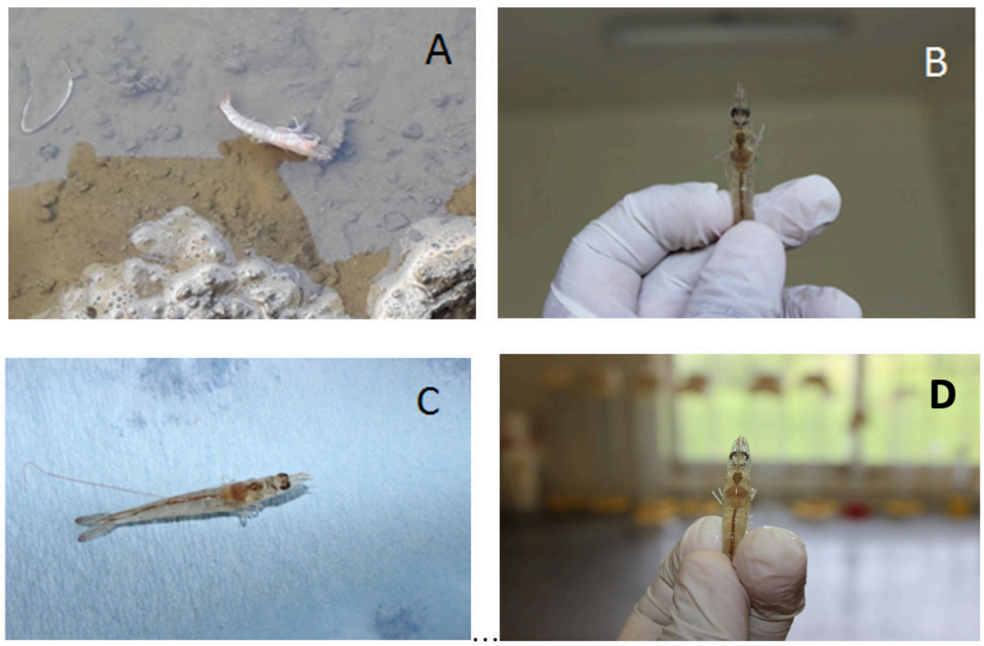

Figure 2. Massive shrimp mortality (A). Hepatopancreas affected in juvenile shrimp collected (B). Laboratory analysis of living organisms (C and D)

In this sense, a study carried out by Akazawa and Eguchi (2013), on AHPND in a comprehensive shrimp farm in the Malaysian peninsula, the results indicated that the disease originated with infected postlarvae and spread rapidly to all fish ponds of the farm.

For its part, the World Organization for Animal Health (OIE, 2016), to establish standards and codes to improve sanitary safety and international trade in aquatic animals, among which are penaeid shrimp, in 2016, published an updated list of pathogenic viral and bacterial diseases of crustaceans to be considered as a mandatory declaration by the affiliated member countries of this organization, the OIE list includes Acute Hepatopancreatic Necrosis Disease, among which are also:

1) Taura Syndrome Virus (TSV)
2) White Spot Syndrome Virus (WSSW)
3) Yellow Head Virus (YHV)
4) Infectious haematopoietic and hypodermal necrosis virus (IHHNV)
5) Infectious myonecrosis virus (IMN)
6) Whitetail disease (ECB)
7) Acute hepatopancreatic necrosis (AHPND)
8) Necrotizing hepatopancreatitis (NHP)
9) Acute hepatopancreatic necrosis disease (AHPND)

These diseases transcend due to the negative impact they have on shrimp farming production systems, with the probability of affecting the natural populations of this species. The competent authorities of the importing and exporting countries must be obliged to comply with the sanitary regulations of the Aquatic Code, during the activities of early detection, notification, and control of pathogens in aquatic animals, avoiding their spread, through the international trade of aquatic animals and derived products, as well as the establishment of unjustified trade barriers (OIE, 2015).

Given these examples of health emergencies and to face contingencies due to new and rapidly spreading diseases, in the 10th Meeting of FAO's Subcommittee on Aquaculture held in 2019, the causes, factors, and pathways of appearance of aquatic animal diseases were analyzed and four indicators were established for their study: i) trade and movement of products and live aquatic animals; ii) knowledge of pathogens and their hosts; iii) aquatic animal health management, and iv) ecosystem changes.

In the shrimp sector, one of the most complex paradigms is biosecurity, to strengthen it requires raising the level of action, health mapping, the formulation of contingency plans and drills, the implementation of compensation programs, and an increase in the level of knowledge about biosafety, the review of fresh food, greater control of the environment, the appropriate disposal of organic waste, the manipulation of microbial communities and the promotion of applied research (Figueredo, *et al.*, 2020).

Six years after the biological contingency arose, the country's health authorities have reacted favorably to stop the advance of the disease, the control measures implemented by farmers from Nayarit, Sinaloa, and Sonora, for the eradication of the bacteria, have been seen reflected with the increase in shrimp production two years after the outbreak; from 48,022 t of shrimp produced in 2013, it went to 112,426 t for 2015. In absolute terms, this annual increase corresponds to 64,716 t of live shrimp produced by aquaculture practices, equivalent to 134.1% in the recovery rates, concerning what was produced in the year in which the biological contingency occurred (Figure 3).

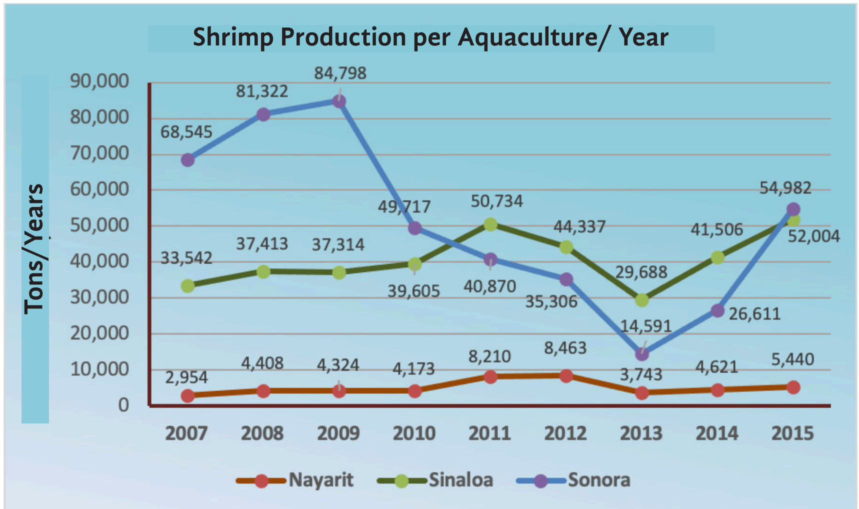

Figure 3. Shrimp Aquaculture Production in Three States Affected by EMS / AHPND (Tons / Year Live Weight). Source: Yearbook: Fishery And Aquaculture Statistics (CONAPESCA, 2007-2015).

AHPND resistance challenge investigations conducted from 2014 to 2016 in L. vannamei show that there is additive genetic variation in the Resistance Line and in the Growth Line, which can be exploited in breeding programs to increase AHPND resistance. (Castillo, *et al*., 2018).

CONCLUSION

Shrimp aquaculture is considered an expanding activity, the impetus for its growth and development lays its foundations from the use of intensive management techniques, which has caused the appearance and spread of infectious diseases to have begun to severely impact the shrimp crops, affecting in the short and medium-term the sustainability of this resource if biosafety protocols are not established to contain them, treat them in a timely, adequate and environmentally responsible manner. Given the threat of these transnational diseases in the case of AHPND, it is important to immediately activate a national emergency plan to respond in time with coordinated actions aimed at making efficient the care of the cases presented, executing the measures and protocols for the control of maximum security established to prevent its spread. Currently SENASICA, through the Mexico-United States Commission, carries out active surveillance for the detection of AHPND with monitoring and sampling in shrimp farms, to know the degree of affectation by this pathogen.

To respond quickly and effectively to the containment and eradication of transboundary aquatic animal disease outbreaks, it is important that countries strategically develop national contingency plans; achieving a rapid response to disease outbreaks and the detection of a new pathogen requires an effective level of surveillance, diagnosis, and dissemination, in addition to the participation of academia and the establishment of diagnostic laboratories strategically distributed throughout the country with the infrastructure and the technical capacity of specialized personnel for the timely identification of diseases of aquatic organisms of commercial interest.

Another aspect to consider is the restrictions imposed for the movement of shrimp larvae throughout the national territory, which must be produced in laboratories certified in the field of aquaculture health, which will guarantee that they are free from high-impact diseases such as AHPND.

In years after the outbreak of Vibrio parahaemolyticus, the Mexican aquaculture sector has faced this contingency from the implementation of good management practices of the production units and through the obtaining of shrimp postlarvae produced in laboratories from Central and mainly South American countries, with genetic lines resistant to diseases, which has made it possible to improve production volumes in recent years.

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