

Report of *Poecilia mexicana* in a fish assemblage from a Gulf of Mexico blind estuary

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Abstract. The fish assemblage composition at the end of estuarine inlet closed period (May 2013) from Laguna El Llano blind estuary located in Veracruz State, Mexico, is described here as under hypersalinity (73 PSU), low turbidity and well oxygenated waters. The fish assemblage was comprised of four fish species: freshwater opportunistic shortfin molly *Poecilia mexicana*, two marine opportunistic flathead gray mullet *Mugil cephalus* and gray snapper *Lutjanus griseus*, and diadromous catadromous fat snook *Centropomus parallelus*; but in this assemblage *P. mexicana* was the most abundant species, and were collected mature males and females in superfetation stage, while another three species were in standard lengths correlated to undifferentiated and juvenile gonadic stages. In trophic terms, *P. mexicana* and *M. cephalus* were first order detritivorous consumers, *L. griseus* and *C. parallelus* feed on fish and crustacean mainly. This is the first report of these species under hypersalinity water conditions in Mexico.
Key Words: *Poecilia mexicana*, blind estuary, hypersalinity, Gulf of Mexico, Poeciliidae.

Introduction In the coastal zone, the estuaries are the most productive ecosystems, and are also physiologically stressing to organisms due to the salinity variations, however they offer important natural services for fishes such as feeding, refuge and nursery areas (Brito 2012; Needles et al 2015).

Until now, explanations about abiotic and biotic processes have been proposed based on estuaries which have a permanent communication with the open sea; however, it has been recently acknowledged that these processes are different in temporarily sand-isolated estuaries, known as blind or obstructed inlet. Inlet opening and closing dynamics are regarded as a key factor to explain the ecological processes of the biota which they occupy (Roy et al 2001; Schallenberg et al 2010). As opposed to open inlet estuaries, in blind estuaries environmental processes are more variable and unpredictable, thus making it more difficult to recognize seasonally consistent ecological patterns (Taljaard et al 2009).

In general terms, fish assemblages from open inlet estuaries are formed by estuarine-dependant marine species, which arrive in early growth life stages, often this guild contributes the most abundance, biomass, and species richness; besides coexisting with less tolerant marine species, estuarine species are able to complete the whole life cycle in these ecosystems, and freshwater species guild are considered as transient or accidentally visitors, with low species richness and abundances (Potter et al 2015; Whitfield 2015).

From this guild, several *Poecilia* species are able to colonize occasionally or permanently fresh, brackish, and estuarine waters from the eastern United States to the Caribbean Sea and northeastern Argentina. Previously, some species have been reported as salinity tolerant in Gulf of Mexico such as the Mexican molly, *Poecilia sphenops*, and the sailfin molly, *Poecilia latipinna* (Vega-Cendejas et al 2013).

The shortfin molly *Poecilia mexicana* has a wide distribution and is common in freshwater systems along the Atlantic slope and several estuaries and coastal lagoons of the Gulf of Mexico (Rodríguez-Varela et al 2010; Mercado-Silva et al 2012). This paper describes the occurrence of shortfin molly, *P. mexicana* forming an estuarine fish

assemblage in a Mexican blind estuary with seasonally-consistent hypersalinity conditions.

Material and Method

Study area. Laguna El Llano Estuary (LELE) is located at 19°38'25.22" - 19°40'05.41" N, 96°23'54.44" - 96°24'39.44" W, near to Actopan city, Veracruz State, Mexico (Figure 1). It has 226 ha of water surface, the inner shoreline is < 1 m depth, but depths in central channel range from 2 to 4 m deep. The climate is sub-humid warm with a summer rainy season (Aw2); mean annual precipitation reaches 1,286 mm, whereas the annual mean water temperature is 24°C (Arriaga-Cabrera et al 1998). LELE is unique among other Mexican estuarine ecosystems because its inlet is closed to the sea by a sand berm annually, from December to May of each year, with null freshwater discharge and rising evaporation rate, this condition causes stagnancy and hypersalinity in water.

Sampling procedures. The fish species were collected in LELE during May 2013 at end of closed inlet stage; this was the first sample in 12-month project between May 2013 and May 2014 about LELE fish community. Due to administrative authorizations, we could not begin to sample prior to May 2013.

The samplings were made in six sites near the red mangrove, *Rhizophora mangle* and the black mangrove, *Avicennia germinans* fringe zone (Figure 1). Sampling was restricted to depths of 0.8 to 1.2 m and could not be extended into the central channel (depth > 3 m), and the depth of mud soil profile. Each sampling site was located with a Garmin 10X GPS meter, dissolved oxygen (mg L^{-1}) and water temperature ($^{\circ}\text{C}$) was recorded with an Oakton DO 300 series meter, salinity (PSU, practical salinity units) with a Vista A366ATC refractometer, turbidity with a La Motte 2020 turbidimeter (NTU, nephelometric turbidity units), and pH with an Oakton pH 110 meter. The hypersalinity level (> 40 UPS) was defined according to Venice System of water salinity (Ito 1959).

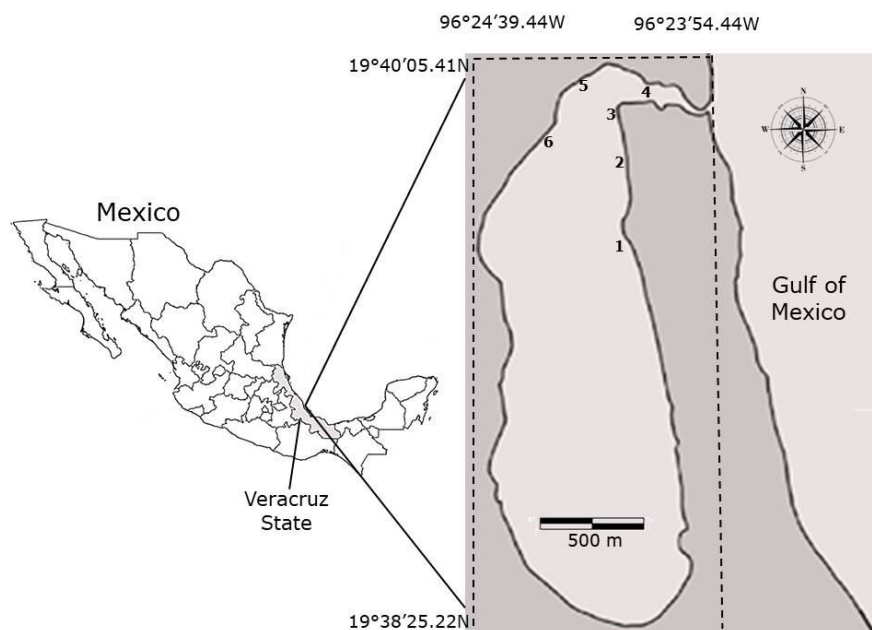


Figure 1. Laguna El Llano Estuary (LELE), Actopan municipality, Veracruz State, Mexico. The numbers are indicating the sampling stations (SS1 – SS6) in May 2013.

Fish species were captured at each sampling site using a single haul seine of 25 m long x 2 m high net constructed with 6.35 mm mesh. The total sampled area at each station was 300 m². All specimens were cooled in ice and then fixed in 70% ethanol in the field, in the laboratory specimens were washed in freshwater, and stored in 70% ethanol; each fish was measured in standard length (SL) rounded to the nearest 0.01 mm with a

caliper, and weighed individual total weight (TW), with a Cole-Parmer Simmetry EC400 electronic balance with 0.001 accuracy. The assemblage definition was used in accord to Fauth et al (1996); fish species common names were used in accord to Froese & Pauly (2017) and the ecological categories following Potter et al (2015) criteria. *P. mexicana* gonadic maturity stages and embryonic phases were defined and were identified with Riesch et al (2011), for the other species were applied Brown-Peterson et al (2011) criteria.

The diets of *P. mexicana* and *Mugil cephalus* were quantified using the grid method of Gido & Franssen (2007), the diatom species in the diet were identified with Prescott (1978) and Bellinger & Sigee (2010) keys; for another two species (*Lutjanus griseus* and *Centropomus parallelus*) were used the gravimetric diet analysis (Hyslop 1980), in all cases the specific diet composition was transformed to percentages.

Results and Discussion. May 2013 was the last month of close-inlet estuarine period, the hydrological records showed warm, lightly alkaline, low turbidity, well oxygenated, and hypersaline waters (Table 1), in June 2013 estuarine inlet was breached and the water exchange with the sea restarted, after that, *P. mexicana* was not collected in LELE again.

Table 1
May 2013 hydrological records in LELE during close estuarine inlet stage (see Figure 1 for sampling site locations (SS1 – SS6))

Parameter	SS 1	SS 2	SS 3	SS 4	SS 5	SS 6	Mean \pm SD
pH	7.59	8.49	7.6	7.61	7.6	7.62	7.75 \pm 0.36
O ₂ (mg L ⁻¹)	8.22	8.91	10.2	9.5	9.86	10.01	9.45 \pm 0.76
T (°C)	30.2	31.9	31.5	31.8	31.9	31.4	31.45 \pm 0.65
Turbidity (NTU)	23	19.1	13.6	10.37	10.45	29.1	17.60 \pm 7.52
Salinity (PSU)	71	74	72	70	76	76	73.17 \pm 2.56

NTU: nephelometric turbidity units; PSU: practical salinity units; SD: standard deviation.

Under these hydrological conditions the fish assemblage were formed with four species, two marine opportunistic species: flathead gray mullet *M. cephalus* and gray snapper *L. griseus*, one obligate diadromus catadromous species, the fat snook *C. parallelus*, and surprisingly the shortfin molly *P. mexicana* as freshwater facultative opportunistic species. The low species richness of this assemblage is typical of this environmentally restrictive estuarine stage (Whitfield et al 2008); moreover some reports pointed out the total fish disappearance when stagnation and hypersalinity conditions prevail during several years, but Vorwerk et al (2003) proposed this salinity level as a selective factor for true eurihalyne tolerant fish species, in this sense *P. mexicana* had showed a physiological aptitude never reported until now.

In open inlet estuaries the fish assemblages are dominated by euryhaline marine species, freshwater species guild contribution to species richness, abundance and biomass is low, for this reason Whitfield et al (2008) pointed out that in blind estuaries is possible to find freshwater endemic taxa. But *P. mexicana* had been reported from another Gulf of Mexico estuaries in low abundances but with frequent occurrence (Castillo-Rivera et al 2011), without a doubt *P. mexicana* is a freshwater species but showed physiological tolerance to salinity because it is a permanent inhabitant of oligohaline habitats in estuaries such as Alvarado Lagoonal System (Chávez-López et al 2015). The fish species records of abundance, weight, standard length (SL), and gonadic growth stages are showed in Table 2.

Table 2

Fish species abundance, biomass, standard length (SL) records, gonadic growth, and ecological guild from LELE in May 2013 under hyper-salinity conditions

<i>Species</i>	<i>Abundance</i>	<i>Biomass (g)</i>	<i>SL rank (mm)</i>	<i>Weight rank (g)</i>	<i>Gonadic growth</i>	<i>Ecological guild**</i>
<i>Poecilia mexicana</i>	170	69.5	4.3-59.2	0.04-5.48	*Oocytes, eggs, embryo in all growth stages	Freshwater opportunistic
<i>Mugil cephalus</i>	29	601.7	85-178	5.1-70.6	Immature, in development	Marine opportunistic
<i>Centropomus parallelus</i>	11	149.6	68 –130	5.1-25.2	Immature, in development	Diadromous catadromous obligate
<i>Lutjanus griseus</i>	7	99.4	53-122	4.1-38.6	Immature	Marine opportunistic

**P. mexicana* gonadic growth stages were defined with Riesch et al (2011) criteria; ** Specific ecological guild definition was made in accord to Potter et al (2015).

One hundred and seventy (170) *P. mexicana* individuals were collected, 128 females in SL interval of 4.28 to 59.21 mm and 0.04 to 5.48 g in weight; 42 males were at 15.73 to 51.41 mm SL and 0.091 to 2.97 g in weight. Fish size classes 10-19 and 20-29 mm were best represented in sampling; both count to 90.5% for male's abundance, and 79.7% for female's abundance; the size class structure is showed in Table 2. Ten females among 36.3 to 50.7 mm SL showed some reproductive products as ova and embryos, in eight of these the embryos were in different development phases, particularly another four of them had developed oocytes and embryos at same time as a superfetation feature, males were found with developed gonopodium at 13 mm SL.

P. mexicana's diet in LELE was composed of detritus mainly (97.7%), other alimentary types were diatoms (1.5%), unidentified algae, foraminifera and crustacean remains (0.26% each one); in *M. cephalus* diet, detritus was the main food item (99%) and algae; in both food contents the observed diatoms belonged to the families Naviculaceae (*Navicula radiosa*, *N. chryptocephala*), Fragilariaceae (*Diatoma hemiale*), Surirellaceae (*Surirella robusta* var. *splendida*), Cymbellaceae (*Cymbella* sp.), and Bacillariaceae (*Nitzschia* sp.); both species formed a detritivorous trofic guild. *L. griseus* diet consisted of un-identified fish debris (47%), polichaeta (27%, Nereidae, possibly) and crustacean debris (26%), finally in *C. parallelus* diet were recorded fish debris only.

In this salinity condition a blind estuary can be viewed as an extreme habitat, less productive and with a lesser species diversity compared to adjacent oceanic shelf and freshwater rivers (Potter et al 2010; Laverty & Skadhauge 2015), moreover, in LELE, the mangrove vegetation is the sole autotrophic source that produces significant amounts of organic matter not grazed as living material, but instead is passed to the detritus food chain where this decaying material then is consumed by detritivore species; this evidence showed there was no feeding restriction to *P. mexicana*, because it is recognized as detritivore consumer in estuaries (Miller et al 2005; Martin & Saiki 2009), this being a great advantage to survive in hypersaline waters, because food was not an environmental limiting factor due to the detritus is an unlimited energy source; in fact, in this estuarine stage, *P. mexicana* must thrive against salinity changes more than food availability.

P. mexicana is a good exception to Whitfield's (2015) six reasons why freshwater fish species appear to be under-represented in estuaries, because in LELE it was able to cope physiologically with the large salinity ranges that occur in most estuaries, it adapted to the altered food resources in estuaries when compared with freshwater systems, competed with estuarine and marine fish species in estuaries, and overcame with connectivity issues between estuaries and upstream river systems. In general terms, Whitfield's (2015) arguments are well applied in permanent open inlet estuaries, but in blind estuaries the whole biota does suffer sudden environmental changes in weekly or

monthly spans, demanding quick physiological responses to organisms; of course, only those with better survivorship faculties will be able to colonize under these extreme conditions.

Conclusions. In this contribution was described *P. mexicana*'s participation in a fish assemblage collected at the end of the closed estuarine inlet stage in hypersalinity waters, were collected another three species: diadromous *C. parallelus* and two marine species, *M. cephalus* and *L. griseus*. For all species this is their first occurrence report in this salinity level; *P. mexicana* was the most abundant species and were identified several mature gonadic stages and gravid females in superfetation process, another three species were in young stages only.

The occupation of hypersaline aquatic habitats by *P. mexicana* is another example of the ability of cyprinodontid species to colonize adverse environmental conditions; the challenge for these organisms is to maintain osmotic homeostasis when facing extreme saline water levels. But this situation seemed mitigated for *P. mexicana* because it was able to grow, since food availability was not a limiting factor because their detritivore diet; *P. mexicana* in LELE was capable of using energy for reproduction; the embryo presence in females is a demonstration of a successful reproductive process with respect to the other euryhaline marine fish species, common estuarine inhabitants.

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