

Comparative resistance of channel catfish (*Ictalurus punctatus*) and channel catfish female × blue catfish (*I. furcatus*) male hybrid catfish against *Aeromonas hydrophila*

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Abstract

Fish handling represents a significant stressor within aquaculture systems, frequently compromising the integrity of crucial mucosal barriers. This mechanical disturbance can lead to the development of wounds and skin injuries, thereby providing entry points for opportunistic bacterial pathogens and increasing the fish's susceptibility to disease. This study assessed the survival of channel catfish (*Ictalurus punctatus*) and channel catfish female × blue catfish (*I. furcatus*) male hybrids (CB hybrids) following *Aeromonas hydrophila* infection after passive integrated transponder (PIT) tag injection. The genotype of channel catfish and CB hybrids was identified using the PCR analysis of follistatin (*fst*) and hepcidin antimicrobial (*hamp*) gene polymorphisms. The findings demonstrated a noteworthy 15% increase in the overall survival rate in CB hybrid catfish compared with channel catfish. Furthermore, statistical analysis revealed a significantly prolonged ($P=0.005$) mean survival time in the CB hybrid group following *A. hydrophila* exposure. Beyond disease resistance, the study also assessed growth performance. Notably, CB hybrid catfish demonstrated significantly greater mean body weight compared to channel catfish in both dying ($P= 0.016$) and surviving individuals ($P<0.0001$). Collectively, these results provide evidence of faster growth of CB hybrid catfish in earthen ponds and higher survival rates following handling stress, skin injuries, and exposure to an infectious agent. These traits are of considerable economic importance for sustainable and efficient catfish farming and Aquaculture.

Introduction

Aquaculture growth remains the fastest among agricultural sectors (1). However, the U.S. catfish industry has contracted since it peaked in 2003, when 300,278 tons were processed, to 136,531 tons in 2014 (2). United States catfish sales reached USD 447 million in 2022, 5% higher than in 2021, while the total area of water surface allocated to catfish farming declined from 23,593 hectares in 2021 to 22,604 hectares in 2022 (3). The primary fish farmed in the US has been channel catfish (*Ictalurus punctatus*), with a more recent transition to CB hybrid catfish produced by hybridizing channel catfish females with blue catfish (*I. furcatus*) males. It was recently reported that 55-70% of the catfish industry has used CB hybrid catfish (4). This transition is the result of the outstanding performance of CB hybrid catfish when compared to both parental species (5,6), including growth rate, feed conversion, resistance to disease-causing agents, tolerance to environmental stressors, harvestability, and performance in intensive production systems (7-10). These performance enhancements have propelled CB hybrid catfish to broad adoption in commercialized catfish operations in the United States. In both research settings and commercial aquaculture operations, individual fish identification is common to track performance metrics and genetic histories. Identifying individual fish can be achieved using different methods. The most common methods are coded wire tags (11), hot branding (12), anchor tags (13), visible implant elastomer tags (VIE) (14), and passive integrated transponder (PIT) tags (15). The PIT tag technology was introduced 40 years ago for individual tagging of fish and has since become an effective method for tagging large numbers of individuals (16). PIT tags offer several benefits over other tagging methods, including higher long-term retention rates, greater accuracy, easier tag identification, and greater reuse. PIT tags can be recovered from the fish, cleaned and disinfected, and reused to tag other fish, reducing costs. The Gram-negative *Aeromonas hydrophila* is one of the primary causes of Motile Aeromonas Septicemia (MAS), with worldwide distribution in cultured and feral fish (17). MAS has frequently affected not only cold-water but also warm-water fishes (18), resulting in high mortality and economic losses (19,20). Several farmed freshwater fish species, including channel catfish, are susceptible to MAS (21). In most cases, *A. hydrophila* is regarded as a secondary opportunistic pathogen, widespread in aquatic environments, living in ponds or stream waters, feeding on dissolved organic compounds (22), and infecting fish when stressed or in a state of immune compromise (23). Several stressors can predispose catfish to *Aeromonas* infections, such as hypoxia, high ammonia levels (24), high nitrite levels (25), and handling stress (23). Systemic infections with *Aeromonas* developed readily in channel catfish with skin abrasions at high temperatures (24°C) and under overcrowding,

compared with fish with intact skin at cooler temperatures (18°C) and low density (26). Clipping of the adipose fins in channel catfish also promotes the development of MAS in disease challenges (27). *Aeromonas hydrophila*, particularly the virulent strain (vAh), causes significant pathology in channel catfish with a range of diverse and severe clinical signs, impacting both dermal and internal organ systems (28). Skin lesions frequently include severe ulceration, often presented with hemorrhagic dermatitis, and petechial hemorrhages (28). Internal organ involvement is characterized by splenomegaly with necrosis, gastric hemorrhage, lesions in the liver and kidneys, ascites, and brain hemorrhages (28). Recently, a virulent Asian-origin strain of *A. hydrophila* caused MAS outbreaks in West Alabama (29), and *A. hydrophila* was identified as the primary pathogen (30). The virulent *A. hydrophila* infection resulted in catastrophic economic losses of \$23.88 million from 2015 to 2021, due to fish mortality and costly veterinary treatments (31).

Currently, no studies have investigated whether fish handling and skin injuries from PIT tag injections facilitate *Aeromonas* infection in channel catfish and CB hybrid catfish, or whether these genetic types differ in resistance under these circumstances. Understanding the characteristics and patterns of *A. hydrophila* infection following PIT tag injection can enhance management practices, thereby reducing fish mortality associated with the PIT tag injection process. In this study, we compared the growth rates of channel catfish and CB hybrid catfish, identified their genotype phenotypically and molecularly using PCR, and examined their resistance to a natural *A. hydrophila* infection after the intramuscular injection of PIT tags.

Materials and methods

Ethical approval

This experiment was conducted at the Fish Genetics Research Unit, E. W. Shell Fisheries Research Center, Auburn University, AL, USA. The experimental procedures of the current study were approved by the Auburn University Institutional Animal Care and Use Committee (AU-IACUC). They followed the Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC) protocols and guidelines.

Experimental fish

CB hybrid catfish were half-sib to channel catfish because they resulted from crossing the same channel catfish parent with a blue mix strain (9). Both channel catfish and CB hybrid catfish fry were of the same age and size (approximately 80 mg). They were stocked communally in the same earthen pond (400 m²) with a 1-meter water depth at a density of nine fry per cubic meter. Fry feeding was conducted according to Qin *et al.* (32). Briefly, the fry was fed to satiation with a 50% protein commercial catfish feed

(Purina® AquaMax® Fry Powder (Purina, St. Louis, MO, USA)). As the fry grew, they were fed Purina® AquaMax® Fry Starter 100, then Purina® AquaMax® Fry Starter 200, and finally Purina® AquaMax® Fry Starter 300 (36-48% protein) twice a day. Finally, fingerlings were fed to satiation using a 32–36% protein floating fingerling feed. The fish were reared under the same conditions during the two-year rearing period. Before PIT tagging, the fish were seined and placed in holding tanks with continuous water flow and aeration. For the current study, we randomly selected 231 two-year-old fish (123 channel catfish and 108 CB hybrid catfish). All fish appeared healthy, with no visible signs of disease.

Injection of PIT tags

Before PIT tagging, fish anesthesia was achieved with 100 ppm tricaine methane sulfonate (MS-222) (Western Chemical Inc., Ferndale, WA, USA) buffered to a pH of 7.0 – 7.5 using sodium bicarbonate. PIT tags (10 mm) were purchased from Biomark (Boise, ID, USA). Then, PIT Tagging was performed by careful injection into the dorsal muscle near the dorsal fin origin, with the needle inserted at a 45° angle. PIT tags were injected by the same person in the same injection site using PIT tag injectors of the same gauge needles and injection technique for all the fish in the study. To minimize handling stress, individual fish weights were recorded concurrently with PIT tagging. After PIT tag injection, fish were distributed randomly and equally across three flow-through holding tanks (approximately 3.0 m × 0.47 m × 0.61 m, with a capacity of 837 liters) at a density of 77 fish (41 channel catfish and 36 hybrid catfish) per tank, with well-oxygenated running water for recovery. All fish in the study were reared under the same environmental conditions and feeding regimens.

Data collection and fish genotype identification

At the time the fish were anesthetized for PIT tag injection, we recorded each fish's body weight, sex, and genotype (channel or CB hybrid). The fish's genotype was identified phenotypically (5) and genetically (33,34). Anal fin tissue samples (10-20 mg) from each fish were placed in a sterile labeled 1.5-mL Eppendorf tube. DNA extraction was conducted according to Elaswad *et al.* (9) using the protocol of digestion with proteinase K, precipitating and washing DNA with isopropanol and 75% ethanol, and elution in TE buffer (10-mM Tris, 1-mM EDTA, pH 8.0). The genotype of individual fish was then confirmed by analyzing follistatin (*fst*) and hepcidin antimicrobial (*hamp*) gene polymorphisms as described by Waldbieser and Bosworth (34) and Perera *et al.* (33). The two bands represent one allele from the channel catfish parent and one allele from the blue catfish parent indicative of the individual being an F1 interspecific hybrid (34). The parent species are homozygous.

Aeromonas hydrophila infection

A natural *A. hydrophila* outbreak occurred in two holding tanks. Each tank contained fish of the two genotypes, channel catfish and hybrids, injected with PIT tags so that each fish could be considered a single experimental replicate of the genotype. Mortality began on the 4th day after PIT tag injection and continued until the 9th day, ceasing on the 10th day. Dead fish were collected, given a value corresponding to the time (day) of death, and necropsied to determine the cause of death. Fish were diagnosed at the Auburn University Fish Farming Center in Greensboro, Alabama, USA, which employs a full-time pathologist and handles more than a thousand cases per year. The causative agent was isolated on Tryptic Soy Agar from the kidneys and livers of five freshly dead fish. The bacterial colonies were then identified biochemically to be *A. hydrophila* according to Hossain *et al.* (35). The strain of bacteria was then identified as *A. hydrophila* (strain AL06-01) using specific PCR primers for *gyrB* and *C13R2* genes and sequencing of PCR products, following the protocol described by Hossain *et al.* (35). No other disease-causing agents were detected.

Statistical analysis

Body weights were examined for normality using the Shapiro-Wilk test. The normality assumption was not fulfilled ($P < 0.05$). The homogeneity of variance assumption was satisfied ($P > 0.05$) when tested using Levene's test of equality of variances. The Mann-Whitney U test was used to compare the body weights of channel catfish and CB hybrid catfish.

The day of PIT tag injection was considered day 0. Each dead fish was given a number corresponding to the day of death. Surviving fish were granted a value of 14 (after five successive days without mortality). Pearson's correlation coefficient between the fish's sex, body weight, and time to death was determined. The survival curves of channel catfish and CB hybrid catfish were evaluated using Cox's proportional hazards model (36). The body weight and sex of the fish were covariates in the test. The Wald test was used to explore the influence of body weight and sex on survival time.

To rule out the effects of body weight, where the CB hybrid catfish were larger, channel catfish and CB hybrid catfish with similar body weights were compared. The numbers of surviving and dead channel catfish and hybrid catfish in each category were compared using Fisher's Exact Test. All data analyses were performed with SPSS 23.0 software (IBM Corporation, Armonk, NY). Boxplots were prepared using the package "ggplot2" (Version 3.5.1) in R (Version 4.4.1). Survival curves were compared using the Kaplan-Meier test. Probability values < 0.05 were considered significant, and all data were provided as the mean \pm standard error (SEM).

Results

In addition to identifying channel and hybrid catfish based on morphological characteristics, we investigated polymorphisms in the *fst* and *hamp* genes. Figure 1 illustrates the polymorphism of the *fst* and *hamp* genes in channel catfish and CB hybrid catfish used in the present study. The results revealed one band for each gene in channel catfish (222 and 348 bp for *hamp* and *fst*, respectively). CB hybrid catfish exhibited two products for each gene (222 and 262 bp for *hamp*, and 348 and 399 bp for *fst*).

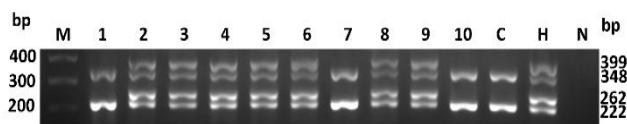


Figure 1: PCR results to identify the genotype of the fish used in the present study based on the differences in length of *hamp* (hepcidin antimicrobial) and *fst* (follistatin) genes. All the fish used in the current study were tested, but only 10 are presented here as examples. Channel catfish (*Ictalurus punctatus*) exhibited one product for each gene (222 bp for *hamp* and 348 bp for *fst*). Channel catfish female \times blue catfish (*Ictalurus furcatus*) male (CB) hybrid catfish showed two products for each gene (222 bp and 262 bp for *hamp*, and 348 bp and 399 bp for *fst*). The two bands represent one allele from the channel catfish parent and one from the blue catfish parent, indicating that the individual is an F1 interspecific hybrid (Waldbieser and Bosworth 2008). The parent species are homozygous. M: 1 Kb plus DNA ladder (Invitrogen). C: channel catfish control. H: CB hybrid catfish control. N: negative control. Samples 1, 7, and 10 are channel catfish, while the rest are hybrids.

At stocking, channel catfish and hybrid catfish that were half-sibs had the same mean body weight (approximately 80 mg). However, after two years of rearing in an earthen pond, the mean body weight (0.572 ± 0.017 kg) of hybrid catfish was significantly higher, with a 55% improvement in the average body weight, than that of channel catfish (0.370 ± 0.013 kg) (Figure 2 and Table 1). The mean body weight of females and males did not differ significantly within each genetic group ($P > 0.05$).

We observed significant pathology in diseased catfish, characterized by a variety of clinical signs affecting the skin. These dermal lesions included ulcerations with hemorrhagic dermatitis and petechial hemorrhages. Mortality began on the 4th day after PIT tag injection, continued till the 9th day, and stopped on the 10th day. Hybrid catfish survival 94.4% after *Aeromonas* infection was 15% higher than that of channel catfish 78.9%, as shown in Table 1 and Figure 2b. All dead fish from both genotypes weighed 640 g or less. Therefore, we repeated the analysis by comparing both genotypes with body weights ≤ 640 g, excluding fish > 640 g

because no mortality occurred in these fish. The cumulative rate of daily mortality was higher in channel catfish than in CB hybrid catfish from the first day of mortality until mortality stopped (Figure 3). No correlations were detected between death time and fish sex ($r = -0.059$, $n = 231$, $P = 0.370$). A weak positive correlation was found between the fish's body weight and death time ($r = 0.203$, $n = 231$, $P = 0.002$); therefore, body weight was added as a covariate in the Cox proportional hazards model.

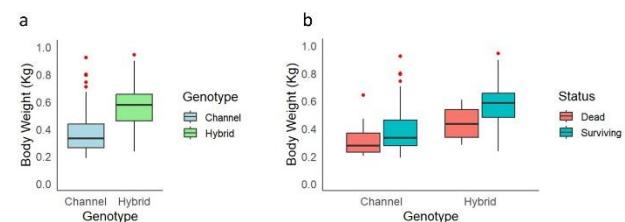


Figure 2: Body weight distribution of channel catfish (*Ictalurus punctatus*) and channel catfish female \times blue catfish (*I. furcatus*) male (CB) hybrid catfish after two years of rearing in an earthen pond (a) and following *Aeromonas hydrophila* infection (b), with "Surviving" or "Dead" representing the status of the fish after infection. Significant pathology was observed in diseased catfish, characterized by a variety of clinical signs affecting the skin. These dermal lesions included ulcerations with hemorrhagic dermatitis and petechial hemorrhages, and *Aeromonas hydrophila* was confirmed by the Auburn University Fish Farming Center, Greensboro, Alabama, USA.

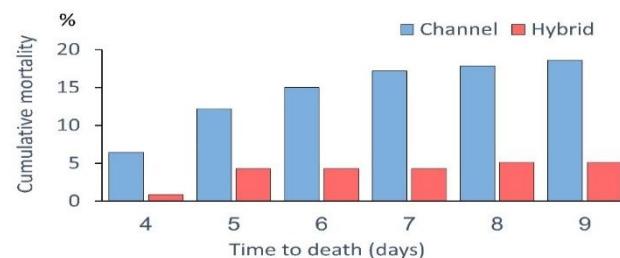


Figure 3: Daily cumulative % mortality in channel catfish (*Ictalurus punctatus*) and channel catfish female \times blue catfish (*Ictalurus furcatus*) male (CB) hybrid catfish following *Aeromonas hydrophila* infections. CB hybrid catfish had significantly lower daily cumulative mortality than channel catfish ($P = 0.097$). Fish were individually pit-tagged at the same site in the dorsal musculature and communally stocked in three flow-through holding tanks (837 liters each) at a density of 77 fish per tank. Significant pathology was observed in diseased catfish, characterized by a variety of clinical signs affecting the skin. These dermal lesions included ulcerations with hemorrhagic dermatitis and petechial hemorrhages, and *Aeromonas hydrophila* was confirmed by the Auburn University Fish Farming Center, Greensboro, Alabama, USA.

Table 1: The number, average body weight (kg), and mean survival time (days) of surviving channel catfish (*Ictalurus punctatus*) and channel catfish female \times blue catfish (*I. furcatus*) male (CB) hybrid catfish studied in the current experiment. Fish were individually pit-tagged at the same site in the dorsal musculature and communally stocked in three flow-through holding tanks (837 liters each) at a density of 77 fish per tank

Genotype	Total no.	Mean body weight (kg) \pm SEM	Mean survival time (days) \pm SEM	Dead fish			Surviving fish		
				No.	Percent	Mean body weight (kg) \pm SEM	No.	Percent	Mean body weight (kg) \pm SEM
Analysis of all fish									
Channel catfish	123	0.370 ^b \pm 0.013	12.19 ^b \pm 0.32	26	21.1	0.312 ^b \pm 0.021	97	78.9	0.385 ^b \pm 0.016
CB hybrid catfish	108	0.572 ^a \pm 0.017	13.51 ^a \pm 0.20	6	5.6	0.537 ^a \pm 0.109	10 ₂	94.4	0.574 ^a \pm 0.017
Overall	231	0.463 \pm 0.124	12.80 \pm 0.20	32	13.9	0.354 \pm 0.169	19 ₉	86.1	0.492 \pm 0.244
Analysis of fish in the body weight range (0.2-0.64 kg)									
Channel catfish	116	0.342 ^b \pm 0.010	12.05 ^b \pm 0.34	26	22.4	0.312 ^b \pm 0.021	90	77.6	0.350 ^b \pm 0.012
CB hybrid catfish	75	0.494 ^a \pm 0.015	13.31 ^a \pm 0.28	6	8.0	0.537 ^a \pm 0.109	69	92.0	0.490 ^a \pm 0.112
Overall	191	0.401 \pm 0.010	12.55 \pm 0.24	32	16.8	0.354 \pm 0.023	15 ₉	83.2	0.410 \pm 0.010

All data are presented as the mean \pm standard error of the mean (SEM). Different superscript letters in a column indicate a significant difference ($P < 0.05$). Significant pathology was observed in diseased catfish, characterized by a variety of clinical signs affecting the skin. These dermal lesions included ulcerations with hemorrhagic dermatitis and petechial hemorrhages, and *Aeromonas hydrophila* was confirmed by the Auburn University Fish Farming Center, Greensboro, Alabama, USA.

When body weight was not included in the analysis, the comparison of survival curves of the two genotypes indicated a significantly higher mean survival time in hybrid catfish (13.51 ± 0.2 days) than in channel catfish (12.19 ± 0.32) ($P=0.002$). Including body weight as a covariate in the analysis revealed its effect on the survival of the two genotypes. Body weight (Table 1) affected survival time, as indicated by higher survival in fish with larger body weight ($P=0.033$). When body weight was included, the p -value of comparing survival curves between the two genotypes (Figure 4) was changed from $P=0.002$ to $P=0.093$.

In general, for both channel catfish and hybrid catfish, dead fish had a smaller mean body weight when compared to the surviving fish (Table 1 and Figure 2). Still, the means were significantly different in channel catfish only ($P=0.019$). The mean body weights of surviving and dead hybrid catfish were not considerably different ($P=0.375$). The mean body weight of dead hybrid catfish (0.537 ± 0.109 kg) was 72% larger than that of dead channel catfish (0.312 ± 0.021 kg) (Table 1, $P=0.016$), which is biologically and economically significant. The mean body weight of dead hybrid catfish (0.537 ± 0.109 kg) was not significantly different from that of surviving channel catfish (0.385 ± 0.016 kg) ($P=0.102$). However, dead hybrid catfish were 49% larger than surviving channel catfish. Surviving hybrid catfish had a significantly larger mean body weight (0.574 ± 0.017 kg) than dead (0.312 ± 0.021 kg) and surviving (0.385 ± 0.016 kg) channel catfish (Table 1, $P<0.0001$). When the analysis was repeated by comparing both genotypes with

body weights ≤ 640 g, the effect of body weight difference on the survival of channel catfish (77.6%) and hybrid catfish (92.0%) was insignificant ($P=0.302$) (Table 1). Then, we used the Kaplan-Meier test to compare the survival curves of channel and CB hybrid catfish, which revealed a remarkably longer mean survival time for CB hybrid catfish ($P=0.005$).

Discussion

In the present study, we assessed the survival of channel catfish and hybrid catfish following a natural *A. hydrophila* infection associated with PIT tag injection. We identified the genotype of channel catfish and hybrid catfish using PCR of two polymorphic genes (*fst* and *hamp*), which is more accurate than phenotypic identification. The PCR product lengths were as expected for each genotype and agreed with previous studies (33,34). The CB hybrid catfish had a 15% higher survival rate and 11% longer mean survival time than channel catfish. Also, after two years of rearing in an earthen pond, body weight was significantly larger in CB hybrid catfish than in channel catfish. The overall comparison of survival revealed an effect of body weight: larger fish had higher survival rates. However, comparing survival among fish with similar body weights showed no significant effect of body weight on survival time. Together, these findings of faster growth rates and resistance to key bacterial pathogens, such as *A. hydrophila*, support the transition to CB hybrids as the primary catfish for culture. In this case, there is no trade-off as CB hybrid catfish has outstanding performance

compared to both parental species (5,6), including growth rate, feed conversion, resistance to several disease-causing agents, tolerance to environmental stressors, harvestability, and performance in intensive production systems (7-10). Thus, the current results demonstrate even more added genetic value for the hybrid.

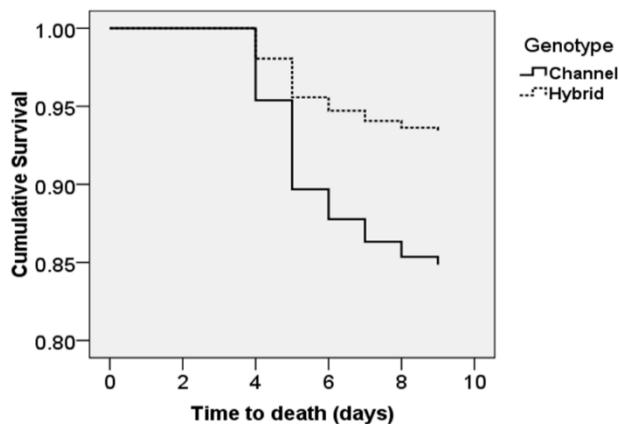


Figure 4: Plot of survival curves of channel catfish (*Ictalurus punctatus*) and channel catfish female \times blue catfish (*Ictalurus furcatus*) male (CB) hybrid catfish following *Aeromonas hydrophila* infections. CB hybrid catfish had significantly higher cumulative survival than channel catfish ($P=0.005$). Fish were individually pit-tagged at the same site in the dorsal musculature and communally stocked in three flow-through holding tanks (837 liters each) at a density of 77 fish per tank. Significant pathology was observed in diseased catfish, characterized by a variety of clinical signs affecting the skin. These dermal lesions included ulcerations with hemorrhagic dermatitis and petechial hemorrhages, and *Aeromonas hydrophila* was confirmed by the Auburn University Fish Farming Center, Greensboro, Alabama, USA.

In aquatic environments, fish are constantly exposed to stressors and microorganisms, including bacteria, viruses, fungi, and parasites, with mucosal barriers being the first line of defense against these pathogens. Fish skin is a physical barrier that serves several functions, including protection against microbes and stressors, osmoregulation, and sensory reception (37). The fish's skin is covered with a mucus layer rich in several immune factors, including mucins, lysozymes, immunoglobulins, proteases, and antimicrobial peptides (38). Also, the skin was reported to be colonized by a rich diversity of bacteria that constitute its microbiome, which plays a vital role in regulating host physiology and immunity (39). Any disruption of the mucosal barrier is expected to increase the fish's susceptibility to opportunistic bacteria. *A. hydrophila* occurs naturally in most freshwaters and is considered an opportunistic pathogen in fish culture, leading to substantial losses when predisposing stressors

exist (40). Fish handling is known to disturb the mucus layer on the skin. Also, PIT tag injection is a stressor. It involves penetrating the skin barrier, which can predispose fish to secondary bacterial infections. In the present study, channel catfish and CB hybrid catfish experienced the same handling stress, were anesthetized with the exact MS-222 dosage, and were PIT-tagged at the same location by the same person. Therefore, differences in survival after PIT tag injection could be attributed to fish-related factors. Moreover, hybrid fish benefit from enhanced immune responses due to the combination of genetic traits from both parental species. For instance, CB hybrid catfish exhibit superior disease resistance compared to their parental species. This is partly due to transgressive gene expression, as CB hybrid catfish showed significant upregulation of immune-related genes and pathways compared to both parental species (8). Similarly, in hybrid yellow catfish, Toll-like receptor (TLR) expression was upregulated after exposure to *A. hydrophila*, thereby mediating the innate immune response (41).

Previous studies have revealed that CB hybrid catfish offer production advantages over channel catfish, including greater resistance to disease-causing agents and better tolerance to environmental stressors (7-10). Our findings show that CB hybrid catfish exhibited higher resistance against *Aeromonas* infection than channel catfish. In addition, fish handling and high stocking density can be considered environmental stressors that negatively affect disease resistance. A recent study on silver catfish (*Rhamdia quelen*) reported that higher stocking densities could impair the number of cutaneous secretory cells, subcutaneous dermal thickness, and lysozyme activity in the epidermal mucus (42). CB hybrid catfish outperformed channel catfish at higher stocking densities, indicating greater stress resistance (43,44). These findings may explain the higher survival rates of CB hybrid catfish following stress from handling and PIT tag injection.

The survival rate of CB hybrid catfish was 15% higher, and the average survival time was notably more extended than that of channel catfish. This is important from an economic perspective. The mortality rate in the current study was 21.1% for channel catfish and 5.6% for CB hybrid catfish, reflecting background mortality that may go unnoticed in fish farms. This is something farmers may experience during a regular grow-out period, so, in the end, this could enhance profitability in routine culture conditions where higher survival would translate into profit. Also, the longer survival time after infection provides a longer opportunity for fish treatment than in those that die sooner, especially under current US law, which delays treatment of sick fish because a veterinarian must prescribe antibiotics. We conducted the survival analysis twice, once with and once without including body weight measurements as a covariate. We detected a body-weight effect on survival when all fish were included, indicating that larger fish have a higher chance of survival. However, when fish of both

genotypes with body weights ≤ 640 g were compared, body weight was not significant. Regardless of whether body weight affects survival, rearing CB hybrid catfish would be an added benefit because they grow faster and are more resistant to disease, leading to higher harvest weights and survival, ultimately increasing profits.

PIT tag injections are a commonly used identification method in fish and wildlife research. Fish can be PIT-tagged via different routes, including injection (45), surgery (46), and external attachment (47). Compared with other fish tagging methods, PIT tags demonstrated relatively higher tag retention rates (45). However, stress from handling and injections may disrupt the fish's mucosal defenses. Injuries similar to those induced by PIT tag injection needles can occur in catfish during transport or at higher stocking densities. The channel catfish and CB hybrid catfish possess sharp pectoral spines capable of causing injury, especially during seining or transport when many fish are crowded in a small water volume. In addition, injuries from fighting over feed in heavily stocked ponds cannot be ruled out. In the present study, the CB hybrid catfish exhibited higher survival after skin injury by PIT tag injectors. However, survival after the abovementioned injuries should be investigated, considering fish are not anesthetized during seining or transport.

Conclusion

The current study demonstrated the superior performance of CB hybrid catfish compared with channel catfish. CB hybrid catfish exhibited a 55% improvement in body weight, a 15% higher survival rate, and an 11% longer mean survival time following *Aeromonas hydrophila* infections after skin injury. These economically essential traits can translate into profit for catfish farmers. This study provides supporting evidence for switching from channel catfish to CB hybrid catfish aquaculture, given faster growth and greater disease resistance following skin injuries.

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Conflict of interest

The authors declare that they have no conflict of interest.

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الخلاصة

المتسلسل لجينات الفولبيستاتين (*fst*) والهبيدين المضاد للميكروبات (*hamp*). أظهرت النتائج أن معدل البقاء على قيد الحياة كان أعلى بنسبة ١٥٪، كما كان متوسط مدة البقاء على قيد الحياة أطول بشكل ملحوظ ($P=0.005$) في هجين أسماك السلور CB مقارنة بأسماك السلور القتيبة. علاوة على ذلك، أظهرت الأسماك الهجينية CB وزنًا متوسطًا للجسم أقل بشكل ملحوظ من سمك السلور القتيبة، سواء في الأسماك النافقة ($P=0.016$) أو الأسماك الباقية على قيد الحياة ($P<0.0001$). وتُقدم هذه النتائج مجتمعةً دليلاً على النمو المتتسارع لأسماك السلور الهجينية CB في البرك التربانية، بالإضافة إلى ارتفاع معدلات البقاء على قيد الحياة بعد إجهاد التعامل، وإصابات الجلد، والتعرض للعوامل المعدية. وتُعد هذه الصفات ذات أهمية اقتصادية كبيرة لتربيبة أسماك السلور وتربية الأحياء المائية بشكل مستدام وفعال.

يُمثل التعامل مع الأسماك عامل ضغط كبير في أنظمة تربية الأحياء المائية، مما يُضعف في كثير من الأحيان سلامه الحواجز المخاطية الأساسية. يمكن أن يؤدي هذا الإضطراب الميكانيكي إلى ظهور جروح وإصابات جلدية، مما يوفر نقاط دخول لمسببات الأمراض الجرثومية الانتهازية ويزيد من قابلية الأسماك للإصابة بالأمراض. هدفت هذه الدراسة إلى تقييم معدل البقاء على قيد الحياة لأسماك السلور القتيبة (*CB hybrids*) وأسماك السلور الهجينية (*Ictalurus punctatus*) الناتجة من تهجين إناث سلور قتيبة \times ذكور السلور الزرقاء (*I. furcatus*) بعد الإصابة بجرثومة الإيروموناس *Aeromonas hydrophila* عقب حلقها بعلامات التعريف الراديوبوئية السلبية. تم تحديد النمط الجيني لأسماك السلور القتيبة والهجينية باستخدام تفاعل البلمرة