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Symptomatic Diseases Surveillance of Climbing Perch (*Anabas testudineus)* under various Recirculatory aquaculture Systems (RAS) of Haryana

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ABSTRACT: Recirculatory aquaculture system (RAS) provides appropriate water quality parameters and a pathogen free environment during the culture period because productive growth of fish depends on various factors like: water quality, pathogen free environment, stocking density and seed quality etc. Sometimes, disease outbreaks in RAS reveal the favourable conditions of pathogens which lead into a higher rate of mortality in this super intensive system where water exchange is limited. Pathogenic infection in RAS causes mass mortality once it comes out. In this concern, our surveillance is aimed to identify the climbing perch common symptomatic disease outbreaks in RAS of Haryana state. During 2021-2022 survey we examined randomly total 14 RAS of distinct districts in Haryana and on each site we collected 20-40 symptomatic infected pieces of Climbing Perch (*Anabas testudineus*). As per farmer interrogation we observed that dispersion of fish infection varies with stocking densities. RAS farmers with high stocking densities (10000-25000/50000 litre tanks) in our surveillance got various pathogenic infections in their stock as compare to optimum stocking density farms (3000-8000/50000 litre tanks) respectively. Tail and fin rot, red spot and white cotton like growth on body were the most common symptomatic infections in climbing perch under RAS systems of Haryana. This survey on distinct RAS farms of Haryana also highlighted the most common treatment measures that a farmer adopted to get rid from these mortal diseases.

Keywords: Stocking density, Infection, RAS, Pathogen, Disease.

INTRODUCTION

Blue transformations of fisheries sector have a prominent role to fulfill the world's upcoming demand of protein and food security. Now a day's fish farmers and aqua entrepreneurs are adopting intensive scale of fish farming practices and technologies to double their income and production. Because of these approaches India become global leaders in fisheries sector after china with total share of 7.58% in world's production of fisheries (Anonymous, 2022). Advancement of fisheries industry will be very productive for its growth and expansion. Moreover, culture of valuable fish species under intensive systems like: Recirculatory aquaculture systems (RAS) and Biofloc technology will be productive in prospective of doubling fish farmer's income. Modernizations in fisheries sector comes up with the introduction of Pradhan mantra matsya sampada yojanan (PMMSY). Moreover, RAS has an enclosed super intensive system intended to manage the high value fish species culture on higher stocking densities and the development of these kind aquaculture system and expansion bring about some serious aspects of pathogens occurrence during the culture Sergaliyev *et al.* (2017). These systems are often intensive in operation with high stocking densities where pure oxygen supplementation is provided for biofiltration to remove ammonia and dissolved carbon dioxide Summerfelt *et al.* (2015). Water physicochemical properties can be more unstable in RAS systems than in large ponds or flow-through systems, where stocking densities is lesser than RAS. Fluctuations in water quality parameters result in sudden disease outbreak or significant losses during the production cycle (Banrie, 2013). There is no hesitation that in RAS more sophisticated conditions were provided to fish for their better survival and production but this does not always commit pathogen free environment under water Sergaliyev *et al.* (2017). Climbing perch (Anabas testudineus) locally famous name is kabai or kawai in India (Fish Base, 2022). Kabai is a freshwater fish species and they have an ability to live under low

oxygen conditions because of extra respiratory organs (Hughes *et al.* 1986). Besides this, climbing perch has thick flesh and delicate tastes (Muchlisin, 2013). Kawai contains high values of iron and copper essentially needed for hemoglobin synthesis (Saha, 1971). Mostly found in small rivers, canals, and swamps Hossain *et al*. (2021). In India climbing perch has a high customer demand with high value price (Singh *et al*. 2018). Because of its high value fish farmer in Haryana state of India prefer to culture kawai under intensive culture systems to gain up the high profit and production under confined water systems (RAS). Moreover, Haryana is the only state in India where first RAS system installed in India during 2016 at Sultan fish farm (TAAS, 2020). Besides this, Haryana is on second position in India during 2021 according to annual per hectare fish production (7000 kg/hectare) followed by Punjab (Anonymous 2021a, 2021b). On the other side, Recirculating aquaculture systems (RAS) enjoy many advantages. Advanced recirculatory systems represent only 4.5% of the total aquaculture production as compare to fish farming in pond culture dominating means of production. However, by the end of 2030, RAS will produce the near about 40% of the total aquaculture output. Europe poised to be a leader in water reuse systems (Lux Researcher, 2015). On the other hand, Physical achievements of department of fisheries under Pradhan Mantri Matsya Sampada Yojana already approved 2870 RAS units in India to till date (Anonymous, 2022b, 2022). However, the prevention and treatment of diseases in RAS is little bit challenging, as the pathogens spread throughout the system, and the addition of chemicals and antibiotics disrupts the microbiome of the biofilters Almeida *et al.* (2019). Under different circumstances with this flow-through aquaculture like: high fish and pathogen densities, limited medication possibilities, in many cases newer or less studied cultured species make recirculation facilities prone to disease problems (Koski, 2013). There is very limited study available on most common climbing perch disease outbreaks in RAS systems of Haryana. Therefore, in our one year surveillance, we tried to investigate the symptomatic infectious diseases outbreak in climbing perch under water reuse system (RAS) with different stocking densities and their preventive or therapeutic treatment measures.

MATERIAL AND METHODS

A cross question interview based survey was conducted throughout the Haryana state on different RAS farms to understand the most common water reuse farming problems (Table 5). Our main aim was to identify the symptomatic infections of climbing perch under distinct stocking densities. During this study, we have collected 20-40 pieces of climbing perch on each RAS farm and on the basis of fish farmer's interrogation; we enlisted various symptomatic history and present situation of fish health. Moreover, we gathered this data from 14 distinct RAS farms of Haryana during 2021-2022 respectively. A survey questionnaire was used to extract the primary information regarding the stocking density of climbing perch under RAS tanks and the most common infectious symptoms, farmer's ability to diagnose fish disease with the help of infectious symptoms, fish health management practices, rate of mortality and treatments (Fig. 1).

Performa 7 – Fish health management in RAS

Fish sampling		
Fish daily monitoring		
Prophylactic measures		
First Disease outbreak		
Expert diagnosis or treatment		
Treatment details		
Total mortality with disease		
Aqua products for prevention		
Number of diseases diagnosed		
Total expenditure on treatment		

Performa 4 - Stocking management of RAS

Selection of fish species		
Source of seed		
Stocking Size of seed		
Price of seed		
Transportation		
Stocking time and frequency		
Stocking rate and ratio		
Survival of seed during stocking		
Diseases occurrence after stocking		
Feed size used during stocking		
Name of feed company		
Timing of feeding with frequency		

Fig. 1. Represents the RAS farmer's survey questionnaire proforma sample number 4 & 7.

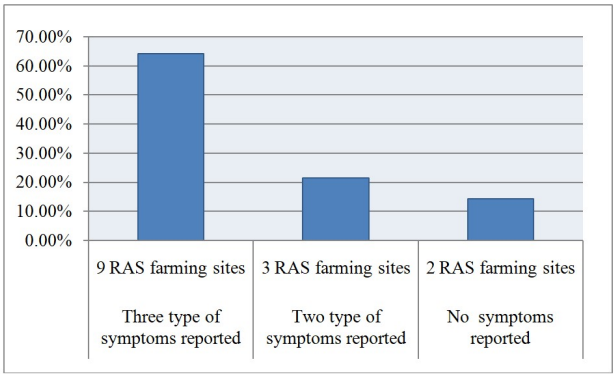
RESULTS AND DISCUSSION

All question queries were collected during survey time from different fish farmer's of Haryana distinct districts regarding stocking density, disease outbreak and mortalities during their culture practices. Besides this, fish farmers and aqua entrepreneur were able to express the symptomatic observation in his/her own words. This was easily understandable. We used simple statistical methods to calculate the surveillance primary data such as frequencies and percentages. Pie and bar charts were used to depict the analyzed variables. Moreover, we applied our clinical pathological sciences of fisheries to identify the exact causative agent of infectious disease with the help of symptomatic identification signs on fish body.

Occurrence of infectious symptoms and rate of mortality on RAS farms. During our survey program mostly RAS farmers of Haryana region reported that they have observed three to two different kinds of infectious symptoms in climbing perch or kawai within 6-8 months of culture period respectively. Significantly, 64.28% RAS farmers reported three types of infectious symptoms on fish body and 21.42% RAS farmers observed only two types of infectious symptoms in climbing perch (Table 1). On the other hand, 14.28% RAS farmers had never noticed any kind of infectious symptoms in climbing perch or kawai during 6-8 months growth cycle respectively (Table 1). The most common reported symptoms were like: red spots and ulcers on fish body like fluid in body respectively, brownish or white cotton like growth on the skin, gills and caudal region of body and tail and fin region start breaking with infections on bas of caudal region respectively. Moreover, highest average rate of mortality also observed in three types of symptomatic RAS farms 34.44% as compare to lowest average rate of mortality 20% find out in two symptomatic RAS farms respectively (Table 3).

Table 1: Represents the different kind of symptoms percentage in various RAS farms of Haryana.

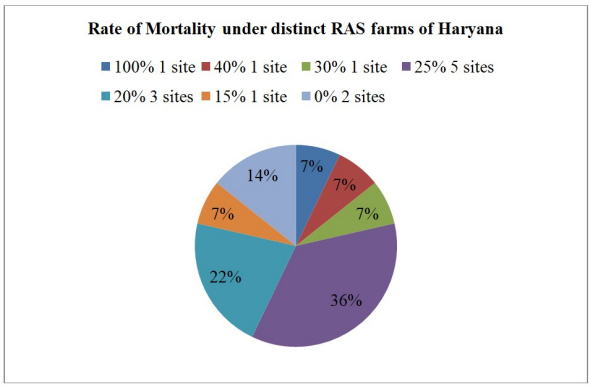
Number symptoms observed	Three type of symptoms reported	Two type of symptoms reported	No symptoms reported
Number of RAS Sites or farms	9 RAS farming sites	3 RAS farming sites	2 RAS farming sites
Percentage share	64.28%	21.42%	14.28%



Graph 1: Represents the distinct kind of reported symptoms percentage on various RAS farms of Haryana.

Table 2: Represents the fish (kawai) rate of mortality on different RAS farms in Haryana.

Rate of mortality (%)	Number of RAS sites or farms	Percentage
100%	1 site	7.14%
40%	1 site	7.14%
30%	1 site	7.14%
25%	5 sites	35.71%
20%	3 sites	21.42%
15%	1 site	7.14%
0%	2 sites	14.28%

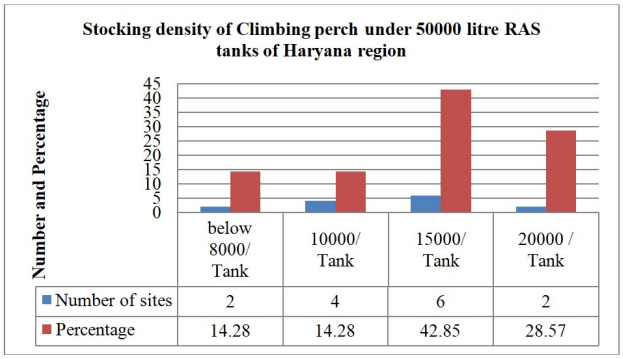


Graph 2: Represents the climbing perch (Kawai) rate of mortality on various RAS farms of Haryana.

Frequency of stocking densities in RAS farms. Stocking density rate in RAS has a significant role in prospective of their growth and survival rate respectively. Moreover, during our one year surveillance, we observed that mostly RAS farmers prefer to stock Anabas test udineus on higher rate in their RAS 50000 litre tanks as compare to lower stocking density rate (Graph 3). On the other side, we observed that different RAS farmers has lower survival rate on higher stocking density and higher rate of infectious symptoms as compare to lower stocking density farms respectively.

Table 3: Represents the fish symptomatic avg. rate of mortality on distinct RAS farms.

Number of RAS sites or farms (Surveyed)	Total farms rate of mortality (%)	Number of symptoms observed	Average rate of mortality (%)
9	310%	3	34.44 %
3	60%	2	20 %
2	0%	0	0 %

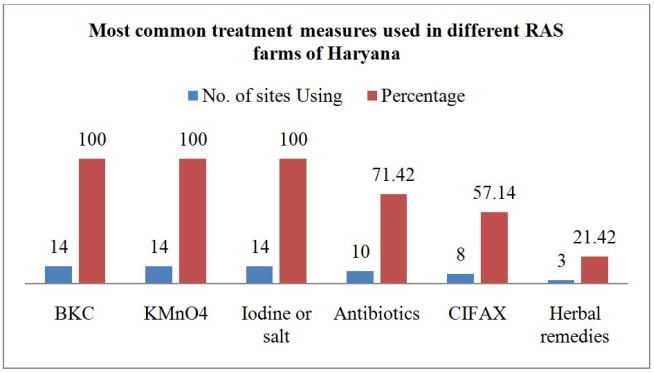


Graph 3: Represents the different stocking densities of climbing perch (Kawai) on various RAS farms in Haryana.

Most common treatment measures on RAS farms. In our survey we noticed mostly fish farmers prefer to use few sanitizers and disinfectants like: Benzalkonium chloride (BKC), $KMnO_4$ and Iodine liquid or salt as a primary most common treatment measure on their RAS farms. Besides this, 72.42% RAS fish farmers prefer to treat their infected fish stock with antibiotics as a secondary most common feed based treatment in their fish diet respectively (Table 4). Moreover, use of herbal treatment was not so popular among the RAS farmers only 21.42% RAS farms prefer to use it as a treatment measure in case of infection. We observed that, use of CIFAX was only 57.14% on different RAS farms during infectious conditions as compare to BKC and $KMnO_4$ respectively (Graph 4).

Table 4: Depicts the different kind of most common treatment measures on various RAS farms of Haryana.

Most common treatment measures	No. of RAS sites Using	Percentage
BKC (Benzalkonium chloride)	14	100 %
$KMnO_4$	14	100 %
Iodine or salt	14	100 %
Antibiotics	10	71.42 %
CIFAX	8	57.14 %
Herbal remedies	3	21.42 %



Graph 4: Represents the different kind of most common treatment measures on various RAS farms of Haryana.

Possible diseases diagnosis on the basis of clinical symptoms. As per clinical signs and farmer's interrogations, we observed mainly possible chances of Epizootic ulcerative syndrome (EUS), Dropsy, Tail and fin rot disease, Saprolegniasis and Cotton wool diseases in climbing perch (Kawai) on mostly RAS farms except on 2 RAS sites where no infectious symptoms reported due to optimum stocking density practices as per our analysis (Table 5) (Kumar *et al.,* 2022; Sergaliyev *et al*., 2017; John & George 2012; Robert *et al*., 2003).

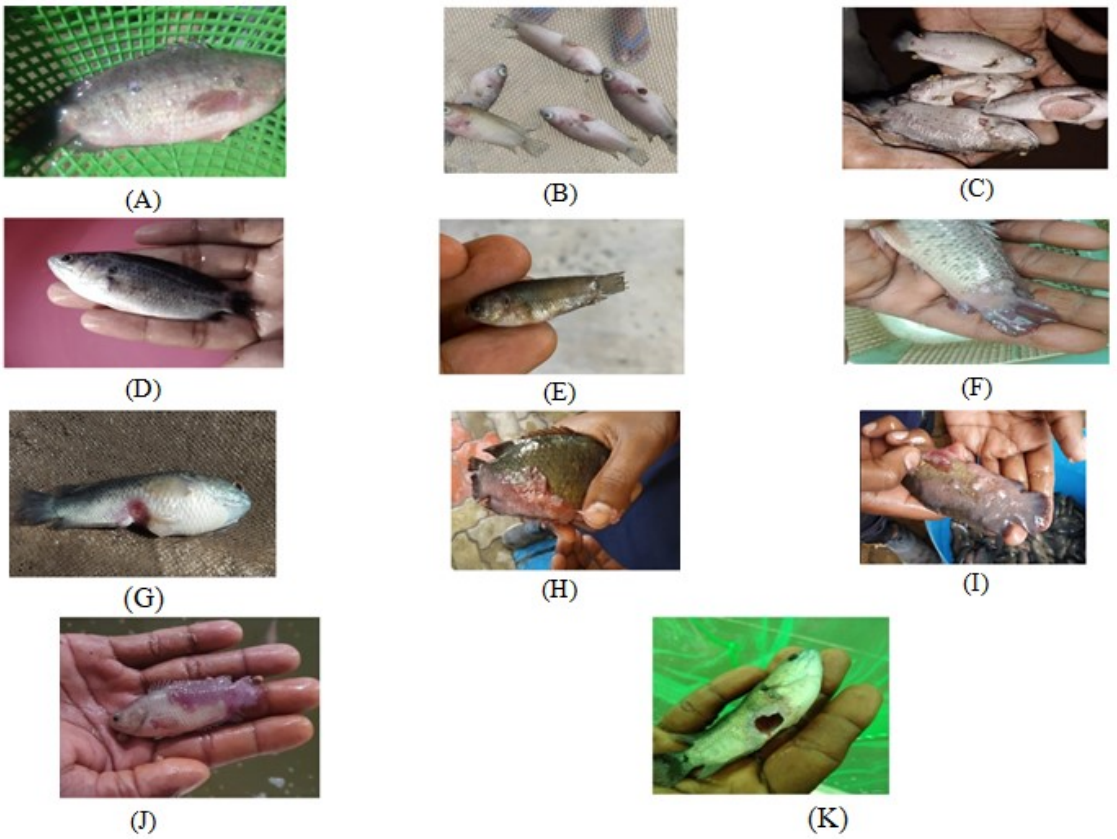
Table 5.

Location of RAS Farm	Fish stocking density (50000 = 50 K)	Fish symptomatic disease out break	Total mortality	Treatment details	Possible symptomatic diseases diagnosis
Site 1 RAS District Hisar, Haryana	10 k fishes/ 50 k litre	Tail and fin infection breakage, Red spot on ulcer with Fluid in body and white cotton like growth on body	30%	$KMnO_4$, BKC, CIFAX, Enrofloxacin 10%, tetracycline and Kmno4	Epizootic ulcerative syndrome (EUS), Dropsy, Tail and fin rot disease, Saprolegniasis and Cotton wool disease
Site 2 RAS District Fatehabad, Haryana	20 k fishes/ 50 k litre	Tail and fin infection breakage , Red spot ulcers with Fluid in body and white cotton like growth on body	100%	BKC, $KMnO_4$, CIFAX and tetracycline	Epizootic ulcerative syndrome (EUS), Dropsy, Tail and fin rot disease, Saprolegniasis and Cotton wool disease
Site 3 RAS District Sonipat, Haryana	15k fishes/ 50 k litre	Red spot, ulcers with fluid in body and white cotton like growth on body	20%	BKC, CIFAX, $KMnO_4$ and Iodine	Epizootic ulcerative syndrome (EUS), Dropsy, Saprolegniasis And Cotton wool disease
Site 4 RAS District Gurgaon, Haryana	12 k fishes/ 50 k litre	Tail and fin breakage, Red spot, ulcers, fluid in body and white cotton like growth on body	25%	BKC, $KMnO_4$, malachite green, iodine, tetracycline and Azithromycin	Epizootic ulcerative syndrome (EUS), Dropsy, Tail and fin rot disease, Saprolegniasis and Cotton wool disease
Site 5 RAS District Fatehabad, Haryana	15000 fishes / 50 k litre	Tail and fin base infection, Red spot and ulcers on body and white cotton like growth on body	20%	Tetracycline, BKC, Iodine, $KMnO_4$ and CIFAX	Epizootic ulcerative syndrome (EUS), Dropsy, Tail and fin rot disease, Saprolegniasis and Cotton wool disease
Site 6 RAS District Bhiwani, Haryana	15000 fishes / 50 k litre	Tail and fin breakage, Red spot, ulcers and with fluid in body and white cotton like growth on body	40%	Tetracycline, BKC, Iodine, and $KMnO_4$	Epizootic ulcerative syndrome (EUS), Dropsy, Tail and fin rot disease, Saprolegniasis and Cotton wool disease
Site 7 RAS District Sonipat, Haryana	8000 fishes / 50 k litre	No disease out break	0%	BKC, $KMnO_4$, spirulina, CIFAX, malachite green and rock salt	No diseases
Site 8 RAS District Rewari, Haryana	15000 fishes / 50 k litre	Tail and fin rot, red spot ulcers and white cotton like growth	20%	BKC, $KMnO_4$, CIFAX, turmeric and tetracycline	Tail and fin rot disease, Saprolegniasis and Cotton wool disease
Site 9 RAS District Rohtak, Haryana	15000 fishes / 50 k litre	Tail and fin rot, red spot ulcers and white cotton like growth	25%	BKC, Iodine, $KMnO_4$, spirulina, CIFAX, malachite green and rock salt	Epizootic ulcerative syndrome (EUS), Tail and fin rot disease, Saprolegniasis and Cotton wool disease
Site 10 RAS District Kaithal, Haryana	25 k fishes / 50 k litre	Red spot ulcers and white cotton like growth on body	25%	BKC, $KMnO_4$, spirulina, Iodine, malachite green and rock salt	Epizootic ulcerative syndrome (EUS), Saprolegniasis and Cotton wool disease
Site 11 RAS District	3000 fishes / 50 k	No	0%	BKC, Iodine, and	No diseases

Rohtak, Haryana	litre			Kmno4	
Site 12 RAS District Ambala, Haryana	15000 fishes / 50 k litre	Fin and tail infection, Red spot on body and white cotton like growth on body	25%	BKC, Iodine, tetracycline and $KMnO_4$	Epizootic ulcerative syndrome (EUS), Tail and fin rot disease, Saprolegniasis and Cotton wool disease
Site 13 RAS District Karnal, Haryana	10000 fishes / 50 k litre	Red spot ulcers on body and white cotton like growth on body	15%	BKC, Iodine, tetracycline, CIFAX and $KMnO_4$	Epizootic ulcerative syndrome (EUS), Saprolegniasis and Cotton wool disease
Site 14 RAS District Rewari, Haryana	10000 fishes / 50 k litre	Fin and tail infection, Red spot ulcers on body and white cotton like growth on body	25%	BKC, Iodine, and $KMnO_4$	Epizootic ulcerative syndrome (EUS), Dropsy, Tail and fin rot disease, Saprolegniasis and Cotton wool disease

Table 5 depicts the RAS farmer's site location, symptoms, treatment details and possible diagnosis diseases.

In present surveillance, we analyzed that most of the RAS farmers of Haryana region doing their fish farming under this advance technology on very high stocking densities (10000-20000 fishes per 50000 litre tanks) with this impact they are facing infectious problems in their kawai (*Anabas testudineus*) stock respectively. Besides this, few RAS farmers have 0% rate of mortality due to (below 8000 fishes per 50000 litre tanks) optimum stocking densities (Table 5). Moreover, during the time of production cycle mostly farmers observing 3 types of symptomatic infections in their fish stock. On the other hand, BKC, $KMnO_4$ and iodine are the most common treatment measures that RAS farmer had taken as primary measure to deal with pathogenic infections. Whereas, antibiotics and CIFAX has also comes on secondary priority of RAS farmers against these infectious diseases in Haryana.



Images A to K: Represents the clinical symptoms of Anabas testudineus collected samples from different RAS farms of Haryana. (A), (B), (C), (H) and (I) Red spots and open Ulcers on body of fish. (F) and (J) Ulcers along with cotton like hyphae growth on body and fin splitting (arrow head), (D) and (E) Fins appear ragged and split, (K) Abdomen burst and with ulcers on body.

CONCLUSION

High value fish farming trend under RAS systems of Haryana on higher stocking densities becoming a main reason for outbreak of pathogenic infectious diseases on their farms and because of this farmer's facing huge economic lose due to high mortality rate. There are various issues in RAS farming of Haryana. However, our main aim of this surveillance was just to express the valuable information regarding appropriate level of stocking densities, possible pathogenic infections and common treatment measures of climbing perch under RAS systems of Haryana.

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Conflicts of Interest. None.

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