EVALUATION OF VARIOUS FILTERS IN RECIRCULATION AQUACULTURE SYSTEMS ON SURVIVAL RATE, CORTISOL, GROWTH AND OSMOTIC PERFORMANCE IN *CHANNA MARULIOIDES* AS DOMESTICATION EFFORT

Rizal. A. Hutagalung[1] S. Anggoro[2] Suryanti Suryanti [3] M. R. Muskananfola[3]
Researcher Prof. Assist. Prof. Assist. Prof.
[1,] Dept. Aqua. Res., Man. Aqua. Res., Fac. Fis. and Mar. Sci., Diponegoro Univ., Indonesia;
[1,] Dept. Mar. Sci. and Fish., Aquaculture, Pontianak State Polytechnic, Indonesia
[2,3]Dept. Aqua. Res., Man. Aqua. Res., Fac. Fis. and Mar. Sci., Diponegoro Univ., Indonesia;
Email: rizalakbarhutagalung020503@gmail.com.

ABSTRACT

This study aims to determine the effectiveness of various filter materials in RAS such as synthetic materials and organic materials and there an impact on plasma cortisol, osmotic performance rate, growth rate and survival rate of *Channa marulioides*. The cultivation process in the RAS system is carried out for 100 days with 50 individuals in each different treatment, namely T1: Synthetic filter; T2: Organic filter; T3: Mixture of synthetic and organic filter; Control: No Filter and RAS. Measurements of plasma cortisol and osmotic performance levels were carried out at the beginning and end of cultivation which had an impact on growth and survival rates. The results of the study showed a decrease in cortisol levels (ng/ml), namely: T1: 9.24, T2: 9.66, T3: 10.37 which showed that there was no significant difference in each treatment (P> 0.05) but in T1, T2, and T3 were significantly different with control treatment: 1.32 (P<0.05). The same trend occurred when observing the level of osmotic performance with the highest decrease in T3 on 6 scales; T2 on a scale of 5.5; T1 has 5 scales, and Control has 3 scales. Reducing the level of cortisol and osmotic performance hormones with the RAS system can increase energy efficiency by reducing stress levels, and osmotic performance so that it can be optimized for growth and survival in adaptation.

Keywords: Aquaculture; Osmoregulation; RAS; Stress Hormone; Water Quality, life below water

تقييم المرشحات المختلفة في أنظمة الاستزراع المائي لإعادة التدوير على أداء الكورتيزول والتناضحي والنمو في جهد تدجين تشانا ماروليويدس

هوتاغالونغ .ريزال[1] أنغورو[2] سوريانتي سوريانتي[3] موسكانانفولا .ر .م[3]
باحث استاذ استاذ مساعد استاذ مساعد

قسم. أكوا. ريس ، رجل. أكوا. ريس ، القوات المسلحة الكونغولية. السمك. ومار. اصابات النخاع الشوكي., جامعة ديبونيغورو., إندونيسيا;[1]
[1] قسم. مارس. اصابات النخاع الشوكي. والأسماك., تربية الأحياء المائية, بوليتكنيك ولاية بونتياناك, إندونيسيا
قسم. أكوا. ريس ، رجل. أكوا. ريس ، القوات المسلحة الكونغولية. السمك. ومار. اصابات النخاع الشوكي., جامعة ديبونيغورو., إندونيسيا,[2,3]

المستخلص

تهدف هذه الدراسة إلى تحديد فعالية مواد الترشيح المختلفة في راس مثل المواد الاصطناعية والمواد العضوية وهناك تأثير على الكورتيزول البلازما ، ومعدل الأداء التناضحي ، ومعدل النمو ومعدل البقاء على قيد الحياة من شانا ماروليويدس. تتم عملية الزراعة في نظام راس لمدة 100 يوم مع 50 فردا في كل معاملة مختلفة ، وهي تي 1: مرشح الاصطناعية ؛ تي 2: مرشح العضوية؛ تي 3: خليط من مرشح الاصطناعية والعضوية ؛ التحكم: لا مرشح و راس. تم إجراء قياسات الكورتيزول في البلازما ومستويات الأداء التناضحي في بداية ونهاية الزراعة والتي كان لها تأثير على معدلات النمو والبقاء على قيد الحياة. أظهرت نتائج الدراسة انخفاضا في مستويات الكورتيزول (نانوغرام/مل) ، وهي: تي 1: 9.24 ، تي 2: 9.66 ، تي 3: 10.37 والتي أظهرت أنه لا يوجد فرق كبير في كل علاج (ع> 0.05) ولكن في تي 1 ، تي 2 ، و تي 3 كانت مختلفة بشكل كبير مع العلاج السيطرة: 1.32 (ع<0.05). حدث نفس الاتجاه عند ملاحظة مستوى الأداء التناضحي مع أعلى انخفاض في تي3 على 6 مقاييس ؛ تي2 على مقياس 5.5; تي1 لديه 5 موازين، والتحكم به 3 موازين. خفض مستوى الكورتيزول وهرمونات الأداء التناضحي مع نظام راس يمكن أن تزيد من كفاءة الطاقة عن طريق الحد من مستويات التوتر، والأداء التناضحي بحيث يمكن أن يكون الأمثل للنمو والبقاء على قيد الحياة في التكيف

الكلمات المفتاحية: تربية الأحياء المائية؛ تنظيم التناضح؛ رأس؛ هرمون التوتر؛ جودة المياه، الحياة تحت الماء

Received:22/11/2023, Accepted:28/2/2024

INTRODUCTION

Identification and utilization of a diversity of aquatic animal species is needed to increase product diversity, towards a sustainable aquaculture process which is currently a global issue (38). Currently the world of aquaculture is experiencing rapid development (50), because fish diversity contributes to socio-economic conditions so it needs to be maintained and developed. (5). Increasing market demand in the world fishing industry requires practitioners to be able to classify several species that have the potential to increase the diversity of aquaculture products, which has an impact on the stability of the aquatic environmental ecosystem as an economic source for locals (17, 26, 53). Freshwater fish represent half of all fish species and are the most threatened group of vertebrates (51). One of the causes of reduced availability of species in inland waters is the reduced environmental carrying capacity of natural habitats, and exploitation of fish along the river flow (8, 22). Southeast Asian waters are an aesthetic area for endemic fish species that have classification of potential species diversity, especially the Channidae family. One type of Channidae fish that has potential market demand is *Channa marulioides*, with its distribution in tropical fresh waters in Southeast Asia currently being supplied from natural catches, making the proliferation of this species increasingly worrying (24). .This condition makes this fish worthy of being classified as an endemic species that needs to be preserved through the design of containers and cultivation media following its natural, sustainable habitat. *C. marulioides* like channidae fish, they are generally fish that live in fresh water in areas with calm currents. (28). *C. marulioides* are often found in the waters of Malaysia and western Indonesia (21). The waters of rivers and lakes on the island of Kalimantan in these two countries have a high abundance of this species. The calm current water habitat type is a typical culture medium that is relevant in the scope of recirculation system fish cultivation. Manipulation of water flow in the form of a *Recirculation Aquaculture System* (RAS) system is an effort to maintain the condition of fish in their natural habitat. There is a strong relationship between water quality and fish production, so that RAS can provide better environmental conditions throughout the year (54). Recirculating system aquaculture is an effort to manipulate water flows by reusing water media repeatedly. RAS can process waste in cultivation containers (12); reviewing this, it can be recommended to implement a recirculation cultivation system (RAS) on snakehead fish species (25). The recirculating aquaculture system is a cultivation system that can be applied on land, indoors, and in a controlled manner to minimize interactions between environmental dynamics and production processes (2). Currently, information on the role of RAS in developing cultivation for certain species still needs to be improved, considering that the RAS system involves filter materials that can optimize cultivation media. Therefore, it is crucial to develop fish farming waste management through the identification of mechanical and biological filter materials to develop the aquaculture sector sustainably (35). The most crucial factor in the success of implementing RAS is the selection of the suitable filter media (29); information from research results about the effectiveness of different filter materials in *C. marulioides* cultivation schemes is very important in structured domestication planning. Domestication efforts require proper identification of the successful adaptation of *C. Macrolides* to the scope of cultivation. The hormone cortisol is a significant indicator of stress (43), fish health condition, and has been used to identify stressors, including organic pollutants (30). Apart from the dynamics of the cortisol hormone, the osmotic performance rate also has an essential role in physiological adaptation efforts. When the osmotic pressure of water approaches the osmotic pressure of fish body fluids, the energy consumed in regulating the osmotic pressure of the fish body to adapt to the external environment is the smallest, which is most conducive to fish survival and growth (19). Examining the importance of sustainable domestication efforts, it is necessary to provide information about identifying the dynamics of stress hormones and the osmotic performance rate in blood plasma, which impact the growth rate and survival of fish by implementing RAS filtering during the rearing process. This research aims to determine the effectiveness of different filter materials in RAS which have an impact on changes in the rate of cortisol performance, osmotic performance, as well as the growth rate and survival of *Channa marulioides* in the scope of the cultivation

MATERIALS AND METHODS

Time, place and research method

This research was conducted between May – June 2023; Blood plasma samples were collected at the Laboratory of the Faculty of Agriculture, Tidar University, Magelang, IndonesiaThe data obtained are observation data on the dynamics of cortisol levels in plasma, and the rate of osmotic performance in blood plasma and culture water media before and after cultivation. At the same time, information data was also collected regarding growth and survival rates during culture. The research was conducted using purposive random sampling technique. Sampling points are determined based on sample variations in each treatment, namely prioritizing observations of the distribution of different individual research objects with different parameters in the population distribution of each treatment. This is done with the aim of ensuring that there is no similarity in sampling the same individuals repeatedly.

C. marulioides cultivation

Observations were carried out by cultivating *C. marulioides* intensively for 100 days. The adaptation process of *C. marulioides* seeds in a closed cultivation container as an open concrete pond for 72 hours after the transportation process from the catch area. Preparation of a tank container measuring 100x50x50 cm with the water media used being fresh water without salinity originating from a groundwater spring treated with a 48-hour recirculation system. Water media samples before and after observation were tested for osmotic performance rate. Selection of observation objects in the form of *C. marulioides* fry originating from wild catches with a weight classification of <9.5 grams with a length of ±10 cm with a size uniformity of >95%. Fish seeds are stocked at a density of 50 fish/treatment/container. Blood was collected without killing the fish during the study. Technique for collecting blood in Puncturing the Caudal Vessel (Caudal Vessel); This is done by administering an anesthetic solution before injecting blood into the midline of the body behind the anal fin, then pulling the syringe through the needle into the muscle until it reaches the spine (spinal column). Take a 1 ml blood sample of *C. marulioides* fish fry before observation using a 2.5 ml syringe and store it in an EDTA tube to suppress the possibility of blood coagulation. Feed management, namely providing feed in the form of dried maggots at a dose of 3% of the biomass. Considerations for choosing to use dry maggot feed are; 1. The nature and eating habits of this fish are carnivorous in nature; 2. The availability of sustainable feed does not depend on natural catch; 3. Maggots contain animal protein which is needed for the metabolism of carnivorous fish. The frequency of feeding is three times a day, with feeding at night being the most dominant. Observations were carried out using an experimental method by comparing cortisol and osmotic performance rate of *C. marulioides* after being caught in nature and then reared in cultivation. As data to support the observation of growth, including length and weight growth rates. Fish are maintained in the RAS system with different filter materials for each treatment, namely: T1: Synthetic filter material: Bioball and Dacron foam; T2: Organic Filter: Activated Carbon and Coconut Fiber; T3: Filter Mix: Bioball, Activated Carbon, Dacron foam, and Fiber; Control: No filter and no recirculation.

Plasma cortisol sampling

Cortisol analysis is carried out as a serological examination to determine the levels in the blood plasma samples being observed. Blood samples were taken from fish that were raised without being killed and without anesthesia, then the fish were immediately returned to the relevant cultivation container for recovery without morphological defects according to their initial condition. 1 ml of blood sample is taken using a 2.5 ml volume syringe moistened with 3.8% sodium citrate as an anti-coagulant. Blood samples were placed in EDTA tubes and centrifuged at 3000 rpm for 10 minutes to separate the plasma. The examination uses an elisa reader with the *Labtron* brand type

LMPR-A12. Plasma cortisol hormone levels were measured using the RIA (radioimmunoassay) method using *Cortisol (^{125}I) Elissa Kit (Ref: RK-240CT) IZOTOP* material.

Osmotic performace sampling

Osmotic performance rate analysis was conducted by calculating the difference in the osmotic performance of blood and water media at the beginning and end of the study. Blood from which body fluids (blood plasma) have been taken is added with 3.8% Vaculab K3 EDTA anticoagulant solution (3.5 grams of Nacitrate in 100 ml of Akadest). The *C. marulioides* rearing water medium was taken after measurements were carried out. The measurement results could be determined by the isoosmotic medium of the test animal, and these results were used as a basis during the cultivation phase. At the end of the study, 12 samples of 0.1 ml of fish blood were taken in each container to measure the blood osmolarity level for each treatment. Osmorality values of fish blood plasma samples were measured directly using an *Automatic Micro-osmometer Roebling Type 13/13 DR Autocal* by calculating the difference between the osmotic pressure of the blood and the osmotic pressure of the rearing medium.

Growth and survivle rate sampling

Data variables observed is the percentage of length gain, meen length and weight gain; these data parameters are to accommodate the growth performance of *C. marulioides* as an impact of treatment. Observations of the length of the fish were carried out at the intial and the final of the study by measuring the total length from the tip of the mouth to the direction of the tail of, in addition to measuring the body weight of the individual. In this research, growth and survival calculated using parameter formula as follows:

- Mean length gain (L) (cm) (40): Mean Final Length (cm) - Mean Initaial Length (cm)
- Length gain (LG) (%) (40): $\frac{\text{Final Lenght (cm)} - \text{Initial Length (cm))}}{\text{Initial Length (cm)}}$ x 100=
- Mean weight gain (W) (g) (1): Mean Final Body Weight (g) - Mean Initaial Body Weight (g)
- Survival rate (SR) (%) (40) : $\frac{\text{Final Number of Live Fish}}{\text{Initial Number of Live Fish Stocked}}$ x 100

Water quality

This research uses a water quality tool in the form of a dissolved oxygen meter with the *Lutron*® brand type *DO-5510* which can display data on oxygen solubility, temperature and pH. Using the ammonia checker tool with the *Hanna*® Brand type *HC HI700* with its reagent package can determine the real-time condition of ammonia levels in water media.

Data analysis

Statistical analysis data was processed using IBM SPSS Statistics 23 software to observe the effect of different filter material treatments in the RAS system on cortisol and osmotic performance rate with a confidence level of 95%. The correlation relationship between parameters is also calculated by calculating the logarithm of the R^2 relationship.

RESULTS AND DISCUSSION

Cortisol Plasma: Plasma cortisol levels were observed before and after the research, this was intended to determine changes in blood cortisol levels at the beginning and end of cultivation. Sampling was carried out considering the condition of fish adapted from nature. The observation results showed a decrease in plasma cortisol levels during the study in all treatments. This indicates that *C. marulioides* adapt to the cultivation environment by decreasing cortisol levels during cultivation. Observations were carried out in the laboratory, and the results were presented in Table 1.

Table 1. Plasma Cortisol levels (ng/ml)

Treatment	Pre Treatment	Post Treatment	Difference Cortisol (Pre Treatment–Post Treatment)
T1	27.8	18.14	9.24 ± 0.025[a]
T2	27.40	17.74	9.66 ± 0.013[a]
T3	28.37	18.00	10.37 ± 0.03[a]
Control	28.00	27.01	1.32 ± 4.09[b]

Note: The same superscript letter in the same row indicated no significant difference on significant level 5% (Tukey test)

The results of laboratory observations show that the difference in the use of filter material with a recirculation system in treatments T1 (9.24 ng/ml); T2 (9.66 ng/ml), dan T3 (10.37 ng/ml) is not significantly different (P>0.05), but is substantially different (P<0.05) from the control treatment without recirculation filtration (1.32 ng/ml). This research shows that the use of other filter materials has no impact on changes in cortisol levels in blood plasma. Even so, the application of the RAS system with filtration is considered efficient in reducing fish stress levels during cultivation. The decrease in plasma cortisol levels of *C. marulioides* during rearing indicates that the fish can adapt physiologically to the scope of cultivation. The recirculation system can reduce stress levels in the adaptation process to support fish growth and health and reduce factors that can reduce productivity (31, 42). RAS can also reduce stress levels contained in water due to the degradation processes of denitrification and nitrification and optimal water management (20, 34). The trends of decreasing cortisol in this study shows that fish caught in the wild can adapt to a controlled cultivation environment with the condition of relevant manipulation of the container and cultivation media. Reducing levels of the hormone cortisol plays an essential role in reducing stress levels in aquatic animals such as *C. marulioides*, followed by reducing the level of osmotic performance so that energy is optimized to increase growth rates Factors influencing fish-producing cortisol levels are initial handling, environmental conditions, temperature, and toxic levels in the water (46). The decrease in cortisol levels in this study was due to good handling and cultivation processes and a positive response to *C. marulioides* in the form of water flow and quality water management in the recirculation filtration system so that it could stabilize water temperatures and suppress dangerous toxic pollutants such as ammonia nitrate. Responses to stressful conditions in fish are divided into primary responses, including endocrine changes; secondary responses, metabolic changes; and tertiary responses, including animal performance (growth, immunity, and changes in behavioral patterns) (10). On the other hand, prolonged exposure to stress can lead to maladaptive responses that adversely impact physiological processes such as growth, behavior, reproduction, and immunity (44). Therefore, it is clear that cortisol concentrations in many matrices are highly correlated with circulating plasma concentrations and can be used as an alternative endocrinological tool to assess stress (30). This study shows that fish can optimally adapt to the cultivation media during cultivation. This research shows that differences in filter materials in recirculating aquaculture systems do not affect the level of osmotic performance of *C. marulioides* during cultivation, but this does not apply to control treatments without recirculating filtration. In cultivating fish species, it is essential to differentiate cortisol rate under homeostatic conditions and conditions outside the normal range. This is to consider variations in cortisol levels in one population (31). The cortisol hormone indicator can be used to evaluate the level of ecological adaptability in fish as captive animals from the wild (18). The difference in cortisol is significant in knowing the physiological response *of C. marulioides* fish caught in the wild as a form of adaptation to the culture media. Research results show that decreasing blood cortisol levels is a good sign for the physiology of fish domesticated from the wild. Controlled stress levels during rearing show positive things for sustainable fish rearing. Optimal metabolic processes and physiological organ adaptation during maintenance trigger a decrease in cortisol levels in the blood, resulting in more efficient energy mobilization. Cortisol levels are closely related to blood glucose levels, impacting energy efficiency (4) and fish health indicator (3). Cortisol levels are a crucial hormone in the physiology and biology of fish (47) in the process of transcriptional modulation of glucose, namely glucose levels in fish blood (6, 18) so that fish can meet their energy needs in stressful situations. The energy resulting from a decrease in cortisol needed during maintenance can be utilized by mobilizing growth reproduction processes and plays an essential role in the osmoregulation and acclimatization processes of aquatic animals (37). The effects of stress and cortisol on fish metabolism go beyond simply disrupting growth through energy redistribution, as the consequences of stress and increased cortisol levels in fish alter gut structure and further inhibit digestive function by reducing nutrient utilization (45). The hormone cortisol is a neuroendocrine component responsible for regulating animal stress responses (15, 32). Cortisol is the primary glucocorticoid hormone that regulates metabolic/physiological adaptation under stress conditions in teleost fish (16). In fish, the stress response includes activation of the hypothalamic-pituitary-interrenal (HPI) axis, culminating in the release of glucocorticoids from interrenal cells located in the head of the kidney (10). After that, the hypothalamus releases corticotropin-releasing factor (CRF) in the blood circulation (27). In some farmed fish, increased glycolysis activity in the blood is one of the early signs of releasing the hormone cortisol. The decrease in cortisol levels in this study shows that *C. marulioides* can adapt through stable physiological mechanisms under biomonitoring conditions. The osmotic metabolism of blood and water media is an effort to adapt fish to the environment, which is the primary stress factor (14) in individuals during cultivation.

Osmotic Performance

In this study, crucial to observe physiological activity in the form of osmotic performance levels in measuring the ability of fish to adapt to water. The osmotic performance rate was observed in each treatment at the beginning and end of cultivation. The decrease in osmotic performance rate drastically in blood osmotic pressure (D), which ranged between 23-28 mOsm/l H_20, while the osmotic medium (M) ranged from 19-21 mOsm/l H_20. The results of observing the level of osmotic performance are presented in Table 2.

Table 2. Osmotic Performance Rate

Osmorality	T1 (Baseline)	T1 (Final)	T2 (Baseline)	T2 (Final)	T3 (Baseline)	T3 (Final)	Control (Baseline)	Control (Final)
Blood Osmostic (mOsm/l H_20) = D	30	25	29	25,3	30	23	30	28
Water Osmotic Media (mOsm/l H_20) = M	20	20	19	20	20	19	20	21
Omsotic Performance Level (M-D)	10	5	10	4,5	10	4	10	7
∑ Cultivation Osomotic Level	5 ± 0.079^a		$5,5 \pm 0.075^a$		6 ± 0.080^a		3 ± 5.63^b	

Note: The same superscript letter in the same row indicated no significant difference on significant level 5% (Tukey test).

The research results show that there is a relatively similar trend in results when observing cortisol levels. There were no significant differences in each treatment ($P>0.05$), except for the control treatment. There were no significant differences in treatments T1 (5 mOsm/l H_20) and T2 (5.5 mOsm/l H_20), as did treatment T3. It can be seen that the treatment T3 (6 mOsm/l H_20) showed the highest decrease in osmotic performance rate, then in the T2, T1, and control treatments, the decrease in the osmotic performance rate was seen to be the smallest with a value of 3 mOsm/l H_20. The decreasing trend in cortisol levels and osmotic performance rate during maintenance, indicates optimal physiological metabolism in increasing ion recirculation in the body so that energy utilization is more efficient. The dynamics of cortisol lperformance rate are essential in ion secretion mechanisms and controlling osmoregulation in fish (36). Differences in osmotic performance rate in the rearing process are related to the stress level of fish during cultivation. The decrease in osmotic performance also has the same pattern as the decrease in cortisol levels as an indicator of stress in fish. Examining these results requires an in-depth analysis of the relationship between plasma cortisol levels and the osmotic performance rate in this study related to stress dynamics in domestication efforts.

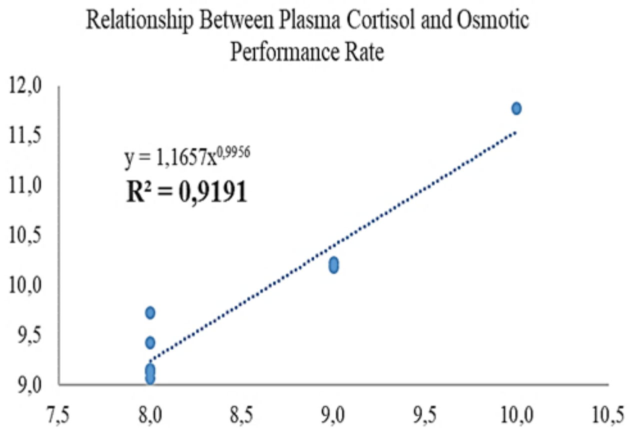

Figure 1. Relationship Between Decreasing Cortisol Levels and Osmotic Performance Rate.

Based on the Figure.1, the relationship between cortisol levels and osmotic performance rate in this study shows a close relationship (R^2: 91), or a relationship of 91%. In fish, the hormone cortisol plays an essential role in responding to stress, namely in osmoregulation metabolism (15). Cortisol synergizes with *Growth Hormone* (GH) on increasing gill Na+/K+-ATPase activity (37), the initial mechanism for fish to adapt through the osmoregulatory system. This study shows a decrease in cortisol as a stress hormone and in the osmotic performance rate. A decrease in the osmotic performance rate followed by a significant decrease in stress hormones has a positive impact on the effectiveness of energy use, thereby impacting the optimization of growth metabolism (9,41), reproduction through gonad development (33) during cultivation.

Growth and Survivle Rate

This research process also shows that domesticated caught *C. marulioides* experienced positive growth both in terms of length and weight gain. The absolute increase in length ranges from 6-9 cm, with an increase in weight ranging from 9-13 grams. Meanwhile, *C. marulioides*, in the scope of cultivation, can maintain life in a population ranging from 69% - to 98%.. The results of growth performance and survival during cultivation are presented in Table 3.

Table 3. Growth Rate and Survival Rate

Performance	Treatment			
	Control	T1	T2	T3
Mean length gain (L) (cm)	6,3 ± 0,39[b]	9,1 ± 0,014[a]	8,8 ± 0,023[a]	9,3 ± 0,014[a]
Length gain (LG) (%)	67,4 ± 9,38[b]	83,8 ± 7,82[a]	82,28 ± 7,79[a]	85,76 ± 7,89[a]
Mean weight gain (W) (g)	11,17 ± 1,97[b]	17,82 ± 0,19[a]	17,41 ± 0,24[a]	18,55 ± 0,22[a]
Survival rate (SR) (%)	69,3 ± 13,02[b]	98 ± 3,72[a]	96 ± 4,18[a]	92 ± 4,28[a]

Note: The same superscript letter in the same row indicated no significant difference on significant level 5% (Tukey test).

The cultivation results with different filter materials in the recirculation aquaculture system did not affect the growth rate in terms of mean length gain (L), mean weight gain (W), or length gain (L). The observations showed that the T3 treatment had the best values for length, namely 9,3 cm, absolute weight (18. 55 grams), and relative growth (85,76%), followed by the T1 and T2 treatments. The results of the data analysis showed that the different filter material treatments were not significantly different ($P>0.05$). However, each different filtration material treatment showed significant differences ($P<0.05$) in the control with no recirculation filtration. The survival rate of fish during rearing had the same trend as the growth rate. Namely, the difference was not significant for each filtration material treatment. Manipulating the container in the form of recirculation and adding filters has been proven to maintain fish survival, ranging between 90-95%. The different filtration treatments showed a significant difference from the control ($P<0.05$), namely without recirculating filtration, which showed that the success of *C. marulioides* in cultivation life in one population was 60-70%. The body will optimally absorb nutrients from feed by increasing body metabolism through energy efficiency to increase growth so that domestication efforts are getting closer to perfection in this context. However, there needs to be further research related to the effectiveness of the growth rate and survival rate of *C. marulioides* during cultivation as an indicator of success in fish cultivation. This study shows a similar trend in decreasing cortisol levels with increasing weight and length and fish survival. This is presented in the relationship graph in Figure 2.

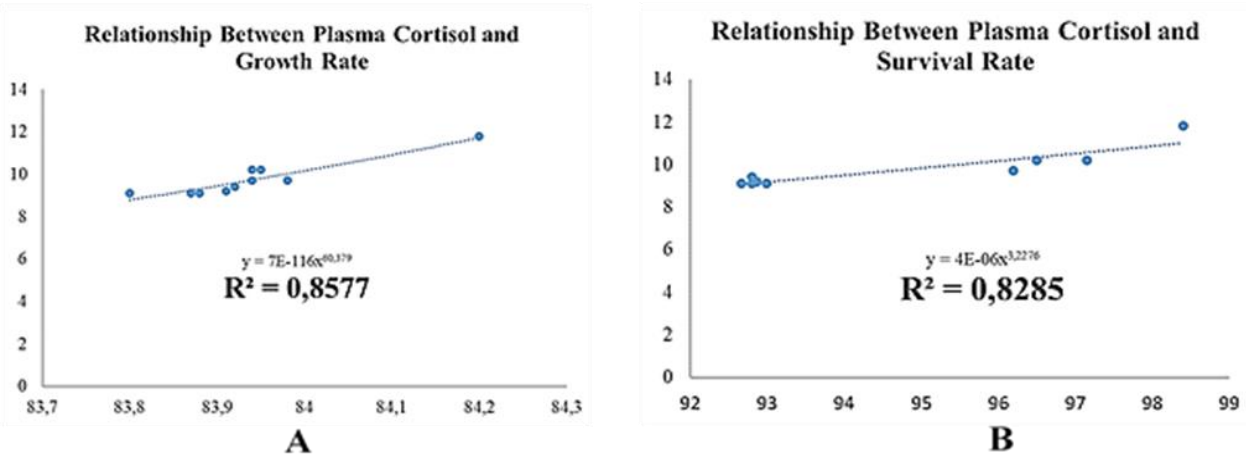

Figure 2. Relationship Between Plasma Cortisol with Growth (A) and Survival (B).

Based on Figure 2, the research results show that the decrease in cortisol levels and growth rate have a close relationship, namely reaching 85.77%. Reducing stress levels also has an impact on the fish's ability to maintain life, with an R^2 relationship value of 82.85%. Reducing cortisol levels has an important role in increasing growth rate, where in this condition the fish will be more optimal in utilizing nutrients in energy metabolism (13) to increase length and weight. Stress in fish affects the ability to maintain life in the scope of cultivation. Fish in stressful conditions release the hormone cortisol through physiological metabolism; this will affect the somatic growth process in aquatic animals (13). In the case of cultivation, serum cortisol levels increase, indicating stress and increased mortality (1, 49). This research shows that fish that experience decreased cortisol levels experience favorable growth. In addition to the energy efficiency obtained from adapting the osmotic performance rate, the fish can grow by increasing length and weight during cultivation. Optimal osmotic performance rate also causes optimal growth due to implementing a recirculation aquaculture system with filtration. *C. marulioides* adapt well during cultivation, and it can be seen that decreasing cortisol levels, which is an indicator of stress, has an impact on growth and survival.

Water Quality

The vital water quality parameters to observe are temperature, pH, oxygen, and ammonia levels. Impacts the recirculation aquaculture system applied in the *C. marulioides* cultivation process is good water quality. Research shows that the recirculation system in cultivating *C. marulioides* can maintain temperatures ranging between 25^0C – 27^0C, pH 5-6, oxygen solubility 4-5 mg/L, and ammonia 0.001 mg/L – 0.002 mg/L. This is significantly different from the results of measuring water media without filtration recirculation in the control treatment. The results of water quality measurements are presented in Table 4.

Table 4. Water Quality

Treatment	Temp. (^{0}C)	pH	Disolved Oxygen (mg/L)	Amonia (mg/L)
T1	25,5	5,8	4,9	0,02
T2	25,8	5,9	5,1	0,02
T3	25	5,6	5	0,01
Control	27,5	6	1,8	0,1

This research also looked at the effects of recirculating filtration on water quality during cultivation. Water quality factors simultaneously have a fairly strong correlation with cortisol (48), in this study showing that fish can adapt to the aquatic environment created in domestication efforts. The measurement results in this study are classified as ideal water quality for living media for *C. marulioides*. It can be seen that there is a decrease in cortisol levels as an indicator of stress and a decrease in the osmotic performance rate as an indicator of optimal adaptation to the water media. The application of aquaponics with a recirculation aquaculture system with a temperature range of 28^0 C – 31^0 C, pH 6 -8, and oxygen solubility of 4 – 6 mg/l has a positive impact on the channa fish, namely *C. Striata* (11). In aquaculture, stress plays a vital role in fish survival due to water quality and dissolved oxygen levels, ultimately reducing fish growth and quality, thereby affecting efforts to domesticate endangered fish species (39). Good water quality management is the manipulation of cultivation water media appropriate to its natural habitat and impacts the level of adaptation through growth and sirvival (7) and reduces stress experienced by fish and prevents the proliferation of disease (23). The recirculating aquaculture system is a water quality index control that is important for the survival and growth of aquaculture objects because RAS is suitable for the concept of sustainable and healthy modern fisheries development (52). The application of recirculation with filtration in *C. marulioides* cultivation media can increase the growth and survival of fish so that it can optimize cultivation results in the future.

CONCLUSIONS

Efforts to domesticate *C. marulioides* by rearing fish in a new, controlled environment through various filter materials in a recirculating aquaculture system did not have a significant effect on reducing cortisol levels, osmotic performance levels, growth rates, and fish survival during cultivation. However, it is recommended to apply a recirculation aquaculture filtration system in *C. marulioides* cultivation efforts using synthetic filter materials and natural materials such as bioball, dacron foam, activated charcoal, and coconut fiber. Reducing cortisol levels during maintenance impacts optimal *C. marulioides* adapting to cultivation media with good water quality by reducing osmotic performance rate during cultivation. Reducing the stress level of fish during rearing and the ability to adapt to the media impacts energy efficiency for growth and survival in controlled cultivation environments.

ACKNOWLEDGMENTS

We would like thank's to BPPT (Higher Education Financing Center) Ministry of Education, Culture, Research and Technology of the Republic of Indonesia; and LPDP (Education Fund Management Institute) Ministry of Finance of the Republic of Indonesia, as funders for this research activity. Thanks are also expressed to all academics at the Faculty of Fisheries and Marine Sciences, Diponegoro University – Semarang – Indonesia, and the Aquaculture Study Program, Department of Marine and Fisheries Sciences, Pontianak State Polytechnic for their academic support.

REFERENCES

1. Al Alalaq, M.A., L. T. Al–Hadedee, and A. M. S. Al-Rubeii. 2023. Effect of Iron Oxide Nanoparticles Prepared by Chemical Method on the Kidneys, Liver and Brain of Male Mice. IOP Conference Series: Earth and Environmental Science, 1252(1), 012132. DOI 10.1088/1755-1315/1252/1/012132

2. Ahmed N. and G.M. Turchini. 2021. Recirculating aquaculture systems (RAS): Environmental solution and climate change adaptation. Journal of Cleaner Production. (297) 126604. DOI: 10.1016/j.jclepro.2021.126604.

3. Afiyah, M.D. Fira, M.B. Santanumurti, M.T. Jamal, A. Muttaqin, S. Subekti, and P.D.W. Sari. 2023. Blood glucose profile as a rapid method for observing Koi carp (*Cyprinus carpio*) health status - case study of ectoparasites in Blitar, Indonesia. Revista Brasileira de Parasitologia Veterinaria 32(2): e014622. DOI: 10.1590/S1984-29612023019.

4. Al-Taee S.K., M.T. Annaz, M.S. Al-Badrany, and A.H. Al-Hamdani. 2021. Biochemical and behavioral responses in carp fish exposed to tricaine methane sulfonate (MS-222) as anesthetic drug under transport conditions. Iraqi Journal of Veterinary Sciences. 35 (4) : 719-723.

5. Alobaidy E.A. and O.K. Jbara. 2021. Production risk analysis of fish farming projects in fishponds and floating cages a case study in diyala governorate. Iraqi Journal of Agricultural Sciences. 52 (2): 403 – 410. https://doi.org/10.36103/ijas.v52i2.1301

6. Aluru N. and M.M. Vijayan. 2009. Stress transcriptomics in fish: a role for genomic cortisol signaling. General and Comparative Endocrinology. (164) : 142–150. DOI: 10.1016/j.ygcen.2009.03.020.

7. Arianto S.R., F.A. Syah, L.A. Sari, A.L. Nafisyah, S. Arsad, and N. Musa. 2023. Analyze the toxicities of benzalkonium chloride as a COVID-19 disinfectant in physiological goldfish (Carassius auratus). Veterinary World. 16(7): 1400–1407. doi: 10.14202/vetworld.2023.1400-1407

8. Aziz M.S.B., A.H. Neaz, R.M. Md. Mostafizur, A. Md. Mehedi, and M.H. Mohammad. 2021. Decline in fish species diversity due to climatic and anthropogenic factors in Hakaluki Haor, an ecologically

critical wetland in northeast Bangladesh. Heliyon. 7 (1) : e05861. DOI: 10.1016/j.heliyon.2020.e05861.
9. Barany A., N. Gilannejad, L.M. Alameda, V.L. Rodríguez, A. Astola, R.G. Martínez, J. Roo, J.L. Muñoz, and J.M. Mancera. 2021. Osmoregulatory Plasticity of Juvenile Greater Amberjack (Seriola dumerili) to Environmental Salinity. Animals. (11) : 2607. DOI: 10.3390/ani11092607.
10. Barton B.A. 2002. Stress in Fishes: A Diversity of Responses with Particular Reference to Changes in Circulating Corticosteroids. Integrative and Comparative Biology. (42): 517–525.
DOI: 10.1093/icb/42.3.517
11. Bich T.T.N., D.Q. Tri, C. Yi-Ching, and H.D. Khoa. 2020. Productivity and economic viability of snakehead *Channa Striata* culture using an aquaponics approach. Aquacultural Engineering (89) : 102057.
DOI: 10.1016/j.aquaeng.2020.102057.
12. Calderini M.L., S. Čedomir, T. Sami, and P. Katja. 2021. Filtration of nordic recirculating aquaculture system wastewater: Effects on microalgal growth, nutrient removal, and nutritional value. Algal Research. (60) : 102486.
DOI: 10.1016/j.algal.2021.102486.
13. Canosa L.F. and J.I. Bertucci. 2023. The effect of environmental stressors on growth in fish and its endocrine control. Frontiers in Endocrinology. (14) : 1109461.
DOI: 10.3389/fendo.2023.1109461.
14. Chen Z., M. Snow, C.S. Lawrence, A.R. Church, S.R. Narum, R.H. Devlin, and A.P. Farrell. 2015. Selection for upper thermal tolerance in rainbow trout (*Oncorhynchus mykiss Walbaum.* Journal of Experimental Biology. 218 (5) : 803–812.
DOI: 10.1242/jeb.113993.
15. Ellis T., H.Y. Yildiz, O.J. López, M.T. Spedicato, L. Tort, O. Øverli, and C.I. Martins. 2012. Cortisol and finfish welfare. Fish Physiology and Biochemistry (38) : 163–88. DOI: 10.1007/s10695-011-9568-y.
16. Fabbri E., A. Capuzzo, and T.W. Moon. 1998. The role of circulating catecholamines in the regulation of fish metabolism: An overview. Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology. 120 (2) : 177-192.
DOI: 10.1016/S0742-8413(98)10017-8.
17. Faisal, S.A., and M. S. Al-Khshali. 2023. Study of the chemical properties of the meat of some fish imported to Iraq. IOP Conf. Ser.: Earth Environ. Sci. 1262 072089.
DOI 10.1088/1755-1315/1262/7/072089
18. Geffory B. 2019. Measuring cortisol, the major stress hormone in fishes. Journal of Fish Biology. 94 (4) : 540-555.
DOI: 10.1111/jfb.13904.
19. Giffard M.I., M.A.H. Hernandez, R.J. Perez, and T.C. David. 2020. Effects of salinity on survival and plasma osmolarity of Totoaba macdonaldi eggs, larvae, and juveniles. Journal of Experimental Marine Biology and Ecology. (526) : 151339.
DOI: 10.1016/j.jembe.2020.151339.
20. Good C., J. Davidson, R.L Earley, J. Styga, and S. Summerfelt. 2017. The effects of ozonation on select waterborne steroid hormones in recirculation aquaculture systems containing sexually mature Atlantic salmon Salmo salar. Aquacultural Engineering. (79) : 9–16. DOI: 10.1016/j.aquaeng.2017.08.004.
21. Gustiano R., K. Kurniawan, and I. Kusmini. 2021. Bioresources and diversity of snakehead, *Channa Striata* (Bloch 1793): a proposed model for optimal and sustainable utilization of freshwater fish. IOP Conference Series: Earth and Environmental Science. (762) : 012012.
DOI: 10.1088/1755-1315/762/1/012012.
22. Hasan V., N.B. Mamat, J. South, F.P. Ottoni, M.S. Widodo, P. Arisandi, W. Isroni, R. Jerikho, D. Samitra, A.R. Faqih, C.P.H. Simanjuntak, and A.T. Mukti. 2022. A checklist of native freshwater fish from Brantas River, East Java, Indonesia. Biodiversitas. 23 (11): 6031-6939.
DOI: 10.13057/biodiv/d231158.
23. Heltonika B., A.O. Sudrajat, M.Z. Junior, Widanarni, M.A. Suprayudi, W. Manalu, and Y. Hadiroseyani. 2023. Immersion Effect of Estradiol-17β on Cannibalism of Asian Redtail Catfish (Hemibagrus nemurus) Post Larvae. Jurnal Ilmiah Perikanan dan Kelautan (JIPK). 15 (1): 95-105.
DOI: 10.20473/jipk.v15i1.38173.
24. Hutagalung R.A., S. Anggoro., Suryanti, and M.R. Muskananfola. 2023. Vitellogenesis activity of Channa marulioides, Bleeker, 1851, impact of folicle stimulating hormone as

reproductive domestication. Iraqi Journal of Agricultural Sciences. 54 (2): 589 – 597. https://doi.org/10.36103/ijas.v54i2.1735

25. Ibrahim L.A., M. Abu-Hashim, H. Shaghaleh, E. Elsadek, A.A.A. Hamad, and H.Y. Alhaj. 2023. A Comprehensive review of the multiple uses of water in aquaculture-integrated agriculture based on international and national experiences. Water. 15 (2) : 367. DOI: 10.3390/w15020367.

26. Isroni W., P.D.W. Sari, L.A. Sari, K. Daniel, J. South, R.A. Islamy, P.Y.A.P. Wirabuana, and V. Hasan. 2023. Checklist of mangrove snails (Gastropoda: Mollusca) on the coast of Lamongan District, East Java, Indonesia. Biodiversitas. 24(3): 1676-1685. DOI: 10.13057/biodiv/d240341

27. Kageyama K., Y. Iwasaki, and M. Daimon. 2021. Hypothalamic Regulation of Corticotropin-Releasing Factor under Stress and Stress Resilience. International Journal of Molecular Sciences. 22 (22) : 12242. DOI: 10.3390/ijms222212242.

28. Kpogue D.N.S., G.A. Mensah, and E.D. Fiogbe. 2013. A Review of biology, ecology and prospect for aquaculture of *Parachanna obscura*. Reviews in Fish Biology and Fisheries (23) : 41–50. DOI: 10.1007/s11160-012-9281-7

29. Lekang O.I. and H. Kleppe. 2000. Efficiency of nitrification in trickling filters using different filter media. Aquacultural Engineering. 21(3): 181-199. DOI:10.1016/S0144-8609(99)00032-1.

30. Lemos L.S., L.M. Angarica, R.A. Hauser-Davis, and N. Quinete. 2023. Cortisol as a stress indicator in fish: sampling methods, analytical techniques, and organic pollutant exposure assessments. International Journal of Environmental Research and Public Health. 20 (13): 6237. DOI: 10.3390/ijerph20136237.

31. Liu D., D.L. Straus, F.P. Lars, and T. Meinelt. 2018. Periodic bacterial control with peracetic acid in a recirculating aquaculture system and its long-term beneficial effect on fish health. Aquaculture. (485): 154-159. DOI: 10.1016/j.aquaculture.2017.11.050.

32. Lohr H. and M. Hammerschmidt. 2011. Zebrafish in endocrine systems: recent advances and implications for human disease. Annual Review of Physiology. (73): 183–211. DOI:10.1146/annurev-physiol-012110-142320.

33. Long X., X. Wu, L. Zhao, H. Ye, Y. Cheng, and C. Zeng. 2017. Effects of salinity on gonadal development, osmoregulation and metabolism of adult male Chinese Mitten Crab, Eriocheir sinensis. PLoS ONE. 12 (6): e0179036. DOI: 10.1371/journal.pone.0179036.

34. Martins C., E.H. Eding, M.C. Verdegem, L.T. Heinsbroek, O. Schneider, J.P. Blancheton, E.R. d'Orbcastel, and J. Verreth. 2010. New developments in recirculating aquaculture systems in Europe: A perspective on environmental sustainability. Aquacultural Engineering. 43(3): 83–93. DOI: 10.1016/j.aquaeng.2010.09.002

35. Martins C.I., E.H. Eding, and J.A.J. Verreth. 2011. Stressing fish in Recirculating Aquaculture Systems (RAS): Does stress induced in one group of fish affect the feeding motivation of other fish sharing the same RAS. Aquaculture Research. 42 (9) : 1378-1384. DOI: 10.1111/j.1365-2109.2010.02728.x

36. McCormick S.D., M.L. Taylor, and A.M. Regish. 2020. Cortisol is an osmoregulatory and glucose-regulating hormone in Atlantic sturgeon, a basal ray-finned fish. J. Exp. Biol. (223) 18 : jeb220251. DOI: 10.1242/jeb.220251

37. Mommsen T.P., M.M. Vijayan, and T.W. Moon. 1999. Cortisol in teleosts: dynamics, mechanisms of action and metabolic regulation. Reviews in Fish Biology and Fisheries. (9) : 211–268. DOI: 10.1023/A:1008924418720.

38. Mohammed A. M., and M.S.Al-Khshali 2023. Effect of fertilization on growth characteristics of *cyprinus carpio* cultured in rice fields in iraq. Iraqi Journal of Agricultural Sciences. 54 (2): 447-454. https://doi.org/10.36103/ijas.v54i2.1719

39. Murugananthkumar M. and C. C. Sudhakumari. 2022. Understanding the impact of stress on teleostean reproduction. Aquaculture and Fisheries. 7 (5) : 553-561. DOI: 10.1016/j.aaf.2022.05.001.

40. Pechsiri J., and A. Yakupitiyage. 2005. A comparative study of growth and feed utilization efficiency of sex-reversed diploid and triploid Nile tilapia, Oreochromis niloticus L. Aquaculture Research. 36:45–51.

DOI: 10.1111/j.1365-2109.2004.01182.x.
41. Peña V.I., C. Narváez, and P. Sabat. 2016. Metabolic cost of osmoregulation in a hypertonic environment in the invasive African clawed frog Xenopus laevis. Biology Open. 5 (7) : 955-61.
DOI: 10.1242/bio.016543
42. Ramírez J.J.D.L., T.J.F. Garcíar, C.L. Felix, T.S. López, F.C.F. Sosa, and O.A.I. González. 2022. Effect of the water exchange rate in a recirculation aquaculture system on growth, glucose and cortisol levels in *Oreochromis niloticus*. Latin American Journal of Aquatic Research. 50 (2) : 267-275. DOI: 10.3856/vol50-issue2-fulltext-2790.
43. Ramsay J.M., G.W. Feist, Z.M. Varga, M. Westerfield, M.L. Kent, and C.B. Schreck. 2006. Whole-body cortisol is an indicator of crowding stress in adult zebrafish, Danio rerio. Aquaculture. (258) : 565–574.
DOI: 10.1016/j.aquaculture.2006.04.020
44. Rousseau K., P. Prunet, and S. Dufour. 2021. Special features of neuroendocrine interactions between stress and reproduction in teleosts. General and Comparative Endocrinology. (300) : 113634.
DOI: 10.1016/j.ygcen.2020.113634.
45. Sadoul B. and M.M. Vijayan. 2016. Stress and Growth. Fish Physiology. (35) : 167-205. DOI: 10.1016/B978-0-12-802728-8.00005-9.
46. Sadoul B. and B. Geffory. 2019. Measuring cortisol, the major stress hormone in fishes. Journal of Fish Biology. 94 (4) : 540–555. DOI: 10.1111/jfb.13904.
47. Samaras A. 2023. A Systematic review and meta-analysis of basal and post-stress circulating cortisol concentration in an important marine aquaculture fish species, European Sea Bass, *Dicentrarchus labrax*. Animals. 13 (8) : 1340.
DOI: 10.3390/ani13081340.
48. Sulmartiwi L., B.E.N.S. Aditama, I.U. Shohifah, and A.R.S.H. Saputra. 2022. Correlation of Density and Water Quality to Cortisol of Cantang Grouper Fish (Epinephelus fuscoguttatus X Epinephelus lanceolatus) in Floating Net Cages. Journal of Aquaculture and Fish Health. 11 (3): 402-411. DOI: 10.20473/jafh.v11i3.36661
49. Tahir D., M. Shariff, F. Syukri, and F.M. Yusoff. 2018. Serum cortisol level and survival rate of juvenile *Epinephelus fuscoguttatus* following exposure to different salinities. Veterinary World. 11 (3): 327-331. DOI: 10.14202/vetworld.2018.327-331
50. Taragusti A.S., P. Prayogo, and B.S. Rahardja. 2021. The effect of stocking density and the application of nitrobacter as ammonia decomposer in aquaponics system of *Clarias gariepinus* with Water Spinach (*Ipomoea aquatica*). Iraqi Journal of Veterinary Sciences. 35 (2): 217-222.
DOI: 10.33899/IJVS.2019.126116.1243.
51. Valdez J.W. and M. Kapil. 2019. Assessing the species in the CARES preservation program and the role of aquarium hobbyists in freshwater fish conservation. Fishes. 4 (4): 49.
DOI: 10.3390/fishes4040049.
52. Yang J., L. Jia, Z. Guo, Y. Shen, X. Li, Z. Mou, K. Yu, and J.C.W. Lin. 2023. Prediction and control of water quality in recirculating aquaculture system based on hybrid neural network. Engineering Applications of Artificial Intelligence. (121) 106002.
DOI: 10.1016/j.engappai.2023.106002.
53. Yuanawati D., H.S. Farizky, M.B. Santanurmurti, M.T. Jamal, L.M.I. Sani, H. Madduppa, and P.D.W. Sari. 2022. The newest COI molecular detection of Asian redtail catfish Hemibagrus nemurus (Valenciennes, 1840) in Progo River, Magelang, Central Java, Indonesia. Journal Of Advanced Veterinary And Animal Research. 9 (4): 591–600.
DOI: 10.5455/javar.2022.i628
54. Zhang S.Y., G. Li, H.B Wu, X.G. Liu, Y.H. Yao, L. Tao, and H. Liu. 2011. An Integrated Recirculating Aquaculture System (RAS) for land-based fish farming: The effects on water quality and fish production. Aquacultural Engineering. 45 (3): 93–102. DOI: 10.1016/j.aquaeng.2011.08.001.