# Effects of Environmental Warming on Clarias gariepinus Growth and Physiology

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Library Investigation

#### <u>Abstract</u>

Fresh water makes up about 0.8% of the Earth's surface and is home to six percent of its species. Environmental warming is projected to have a significant effect on freshwater systems as the century progresses. They are susceptible to irreversible damage because fresh water availability and temperature is dependent of climate. The *Clarias gariepinus*, or the African Catfish, is popular among aquaculture in Africa and Asia because of its rapid growth, quick reproduction, and hardiness. The goal of my thesis was to examine the effect of environmental warming and elevated temperatures on the growth and physiology of *Clarias gariepinus* populations through a review of current literature. *Clarias gariepinus* that inhabited water temperatures of 40°C experienced lethality. Gastric Emptying Time (GET) decreased as water temperature rose. Food Conversion Rate (FCR) and Efficiency (FCE) both increased as water temperature rose. An increased GET with an increased FCE is consistent with similar research. Based on previous work this should have also increased growth rate, but it did not in these studies. No significant difference was observed between the growth of populations in different temperatures. Studies on other freshwater fish species have been done and some exhibit negative ecological results. Environmental warming effects on additional freshwater species have been variable. Only a small temperature range was observed in *C. gariepinus*, more values can depict if growth increases, decreases, or is sustained at higher temperatures. Comprehensive research needs to be done to examine trophic cascade effects.

## **Overview**

Environmental warming is currently occurring at an alarming rate in the world. A 41% increase in greenhouse gas emissions has increased the temperature of the atmosphere 1°C since 1750 and is expected to increase an additional 2°C by 2050 (Polley et al., 2013). This increase in atmospheric temperature will then affect freshwater ecosystems as well because freshwater availability and temperature are dependent on climate. Just about 0.8% of the Earth's surface is freshwater and it is home to 6% of its species. Freshwater is already exposed to multiple stressors like human disturbance and often is hurt by urbanization (Woodward, 2010). If we were to lose our freshwater ecosystems how could that affect human life?

The African catfish is native to Africa and can be found in parts of Asia today. It has become very useful in fish farming. Fish farming is very useful in these countries to help solve food shortages (Prokešová et al., 2015). The African catfish is so desired in fish farming because

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it tastes good when cooked, can grow quickly, reproduces often, can live in low oxygenated waters, and is resistant to many diseases. (Alexandrova et al., 2021). My thesis will examine the effect environmental warming may have on these fish that are so important to societies now. We examined two studies that placed the fish in differing environmental temperature ranges. Things like growth, metabolism, and blood were analyzed. Fish that were placed in water temperatures at 40°C died shortly after being introduced. This was the highest temperature fish were exposed to and the only one where every test subject died due to temperature (Ogunji and Awoke, 2017). It was discovered that as water temperature rose so did the African catfish's ability to digest food and convert it into energy. That energy could then be used to grow and reproduce. Although no major differences were seen with growth at different temperatures, the data did show increased digestion and conversion ability. They also exhibited a greater ability to empty their stomach after eating. This could mean a greater appetite and ability to grow over longer periods of time (Kashimuddin et al., 2021).

Similar research was done on other freshwater species like largemouth bass, rainbow trout, and an entire freshwater ecosystem food chain from the highest to lowest level. Largemouth bass showed a better ability to lower their energy burned when relaxed. This gave them the ability to dedicate more energy to converting food into mass (White and Wahl, 2019). The study on rainbow trout discovered that when the water temperatures were highest in the summer it stunted their appetite and growth (Morgan et al., 2001). The study on the entire freshwater ecosystem food chain found that different levels of the chain were affected in different ways. The highest and lowest levels exhibited greater growth but the two middle layers of the food chain each decreased in abundance as temperatures rose in the water (Mulhollem et al., 2016).

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More studies need to be done taking into account the specific effects of a raised water temperature on freshwater ecosystems at different ranges. This will paint a better picture on what we may expect to see in the future if we do not stop our current environmental impacts. More specifically, more research needs to be done on the African catfish and the effect this may have on them. The fact they died at 40°C is alarming and testing should be done at a wider temperature range. If African catfish were severely impacted in a negative way that could mean a major food crisis in many regions of Africa and Asia that rely on their farming. More research on these effects can help the government make more educated decisions regarding the status of many environmental laws or urbanization decisions. Reducing human impact is the only way we can combat this issue.

#### **Introduction**

Environmental warming continues to have a significant impact on precipitation patterns, increased wildfires, extreme droughts, glacial retreat, and atmospheric warming. Atmospheric warming leads to a modified amount and distribution of precipitation annually. It will increase the number of droughts and heat waves (Polley et al., 2013). Environmental warming directly affects the forage quantity, livestock production systems, soil C content, and livestock metabolism. Each effect has been a result of a 41% increase in greenhouse gases (GHGs) in the atmosphere since 1990 (Polley et al., 2013). GHGs block the emission of long wave infrared radiation into space, by doing this they alter the planet's ability to cool. Even if GHG usage was to decrease immediately, their presence would remain for hundreds of years (Polley et al., 2013). Increased GHG concentration has caused an increase of 1°C to the atmospheric temperature since 1750. It is projected to increase an additional 2°C by 2050 (Polley et al., 2013). Land

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surface temperatures in the northern hemisphere could see a 40% increase to the global average by the end of the 21<sup>st</sup> century (Shuhua, 2021). The rate of warming in the Amazon rainforest has been increasing 0.25°C every decade. Drying trends have been observed since the 1970s in the Amazon and it is projected to continue to increase throughout the 21<sup>st</sup> century. Rainforests themselves have a significant effect on global climates, in which deforestation itself is a driver of environmental warming (Malhi, 2008). A loss of 30-40% of the Amazon rainforest is expected to drive the region into a permanent drier state (Malhi, 2008). The Arctic is responsible for its effect on most of the world's oceans and global climate systems. Findings show that environmental warming's largest effect is the reduction of Arctic sea ice. The trend has lasted since the 1980s, and in the summer of 2018 the coverage of Arctic sea ice was recorded the lowest ever at 3 million square km (Shuhua, 2021). The melting of the Arctic sea ice has major ecological repercussions. It will alter the albedo of the Earth's surface because pure sea water only reflects 5% of solar rays. A mixture of sea water and sea ice is able to reflect 85% of solar rays. The Arctic sea ice is responsible for regulating many of the Earth's temperatures by reflecting that solar radiation into space. Sea water has absorbed an increased amount of heat. This led to a warming trend causing more sea ice to melt and the creation of a vicious cycle of environmental warming (Shuhua, 2021).

Environmental warming will immensely affect freshwater ecosystems. Despite already suffering from the exploitations of human usage, climate change adds to the risk (Woodward, 2010). The quantity of water is one of the initial effects first noticed due to increased run-off patterns from environmental warming (Dallas and Rivers-Moore, 2014). Water quantity can become limited by just a 10% change in precipitation that causes a 20-30% change to run-off (Dallas and Rivers-Moore, 2014). Prolonged precipitation overfills bodies of water, and they

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begin to exceed their water shed. Once the water shed is exceeded, run-off increases. Increased water temperatures lower the water's solubility to oxygen and other gases. Elevated water temperatures also contribute to a doubling of chemical reaction rates and a rise in microbial activity. Together, they can all be ruinous to freshwater ecosystems (Dallas and Rivers-Moore, 2014). Long periods of dry weather seen from environmental warming lowers the movement of water in streams and rivers. This is referred to as low flow and it directly results in decreased food production, water quality, and habitat availability (Sørensen et al., 2009). Regional climate change patterns like extreme droughts and precipitation contribute to low flow and increased surface run-off respectively (Dallas and Rivers-Moore, 2014). The reason that freshwater ecosystems are susceptible to irreversible damage is that many of the species are incapable of relocating habitat loss or destruction. Water sources are reduced by habitat fragmentation (Woodward, 2010). These ecosystems also suffer from exposure to multiple anthropogenic stressors like visitor disturbance and urbanization. Freshwater temperature and availability are also completely reliant on the climate (Woodward, 2010). Fresh water constitutes about 0.8% of the planet's surface and it houses 6% of the Earth's species. The biodiversity of all freshwater ecosystems is at risk from environmental warming regardless of the differing regional effects (Woodward, 2010).

The *Clarias gariepinus*, is referred to as the African sharp-tooth catfish or the African catfish (Figure 1). It is classified under *Clarias* spp and it is native to the subtropical and tropical fresh waters of Africa and Asia (Prokešová et al., 2015). Its potential as an aquaculture species spiked its cultivation in the 1990s. The species quickly spread throughout Europe, Asia, and Latin America. The fish was able to make its way into natural waters through flooding and escape from aquaculture ponds. Naturally, they are carnivorous, but they portray omnivorous

behavior. Their diet is composed of detritus, phytoplankton, fruits, seeds, birds, fish, and small mammals. They also can utilize vegetal protein from feeds (Prokešová et al., 2015). The muscle and caviar are composed of a full range of essential amino acids, high protein counts, and low lipid contents indicative of the fish having a high nutritional value (Alexandrova et al., 2021). The meat composition is on average: 15-20% crude protein, 4-7% crude fat, 3.5-4.5% ash, and 70% moisture. When consumed, studies show it can reduce cardiovascular disease (Alexandrova et al., 2021). They have very few intramuscular bones and are scaleless. They have a high fecundity and develop quickly, reaching commercial weight of 1kg in just one year and sexual maturity in less than one year (Păpuc et al., 2019). Females can spawn multiple times within one year (Prokešová et al., 2015). Although the mechanism is not understood the African catfish is an obligate with the capability to also be a facultative air breather. It possesses gills to breathe in water and other organs in order to breathe air. Dendritic organs, suprabranchial membranes, and gill fans allow the African catfish to breathe air (Prokešová et al., 2015).



Figure 1. An adult Clarias gariepinus (Gao and Gurd, 2019)

African catfish rank as one of the most cultured freshwater finfish species in the world right now (Kashimuddin et al., 2021). They have been introduced into many countries' aquaculture for several reasons. The African catfish possesses one of the highest growth rates among fish species. Their breathing mechanisms allow them to live even in high stocking

densities of 300–400 kg m<sup>-3</sup>, low oxygen levels, high CO<sub>2</sub> levels, high ammonia concentrations, and organic pollution. (Prokešová et al., 2015). They have the ability to inhabit a wide range of habitats like traditional ponds, deep lakes, swamps, recirculating aquaculture system (RAS) tanks, or rivers. (Kashimuddin et al., 2021 and Radhakrishnan et al., 2011). The fish can adapt to new environments and are resistant to many diseases and parasites. Their fast development and diverse diet make them advantageous for cultivation. Several cost advantages come from their breathing mechanism. It allows them to live in hypoxic conditions saving oxygen reserves, electricity, and water volume. An oxygen level of 3-6mg L<sup>-1</sup> is still recommended to ensure metabolic processes can occur (Prokešová et al., 2015). This species is also one of the few fish that can withstand hydrogen sulfide as large adults (Prokešová et al., 2015).

Aquaculture is currently the quickest growing agricultural market. Scientific advances over the last 50 years have contributed to its rapid rise (Alexandrova et al., 2021). Almost 20% of Africa's protein intake comes from fish. Those fish populations are becoming very overstretched since most of those fish are wild caught. Fish farming alleviates the pressure on the ocean species and is working to solve Africa's food security issues. Population growth calls for the need to discover alternative protein sources like fish farming (Mulligan, 2015). Farmed fish are very efficient at converting feed into human edible food. About 10-15% of the calories fish consume is converted into food. That number is just 1% in beef production (Mulligan, 2015). The African catfish has become the preferred aquaculture species in African countries, surpassing Tilapia's popularity (Păpuc et al., 2019). Since 1980, the worldwide agriculture of African catfish has exponentially increased from 50 tonnes to 191,000 tonnes in 2010 (Prokešová et al., 2015). In 2016, 230,000 tonnes of *C. gariepinus* was produced worldwide. Nigeria and Kenya were two of the biggest producers reliant on the species (Păpuc et al., 2019).

Aquaculture continues to be a facet of our world that we may become even more reliant on as population numbers increase. *Clarias gariepinus* are a hardy and tolerant aquaculture species, but with the growing issues of environmental warming and climate change what can we expect to happen to this species when the impact to freshwater ecosystems is already evident? An adverse response to elevated temperatures from a tolerant species like the African catfish could be dangerous for other aquaculture or fish species. My thesis will examine the effect of environmental warming and elevated temperatures on the growth and physiology of *Clarias gariepinus* populations. It will look at two studies focusing on their growth, physiology, and other metabolic processes in different temperature ranges.

## **Current Investigations**

In 2017, a 56-day study was completed to examine the role temperature plays in the survival, growth performance, and hematology of *Clarias gariepinus* fingerlings. Each fingerling was randomly assigned a diet with a protein makeup of 41.97% dry matter, or a diet composed of 50% housefly maggot meal, 34% soya bean meal, 14.5% maize, 0.25% fish oil, 0.25% groundnut oil, and 1% vitamin premix. The mixture was pelleted, and sun dried. Ninety fingerlings made up the test group. They were acclimated for seven days and placed into nine different tanks equally. The initial average weight was  $4.33 \pm 0.03g$  among the fingerlings. The tanks were equally divided among three locations: the laboratory, the greenhouse, and the outdoors. Tanks were regulated with three water temperature measurements daily. The laboratory and greenhouse experimental tanks were monitored daily to ensure proper dissolved oxygen, pH, nitrate, nitrite, and ammonia levels. The experimental tanks were cleaned every three days and all fish were in static water. They were fed a ration of 5% body weight per day

twice a day. The percentage of food given daily was adjusted after the fingerlings were weighed in batches (Ogunji and Awoke, 2017).

Blood samples were taken from each fingerling at the conclusion of the experiment through the caudal vein. The blood was tested for the packed cell value (PCV). This was done with microhematocrit using a 25mm heparinized capillary tube. Red and white blood cell counts, and hemoglobin concentration was determined with methods by Blaxhal and Wedemeyer, respectively. Mean corpuscular hemoglobin (MCH) was calculated using: MCH (pg) = [Hb (g dl<sup>-1</sup>) x 10] / RBC (10<sup>6</sup> µl<sup>-1</sup>). Mean cell volume (MCV) was calculated using: MCV (fl) = Hct / RBC (10<sup>6</sup> µl<sup>-1</sup>). Mean corpuscular hemoglobin concentration (MCHC) was done using: MCHC (g l<sup>-1</sup>) = [Hb (g dl<sup>-1</sup>) x 10] / Hct x 100. Gross energy was calculated based on these factors: crude protein = 23.9kJ/g, crude lipids = 39.8kJ/g, and Nitrogen-free extract = 17.6kJ/g (Ogunji and Awoke, 2017).

After the completion of the 56 day study, growth was analyzed. Weight gain was determined by subtracting the initial weight from the final weight. Specific Growth Rate (SGR) was calculated based on this formula:  $SGR = (lnW_2 - lnW_1) / [(T_2 - T_1) \times 100]$  where  $W_1$  is the initial weight,  $W_2$  is the final weight, and  $T_1$  and  $T_2$  are the time in days. Food conversion ratio (FCR) was done by dividing the feed fed by the live weight gain. Protein efficiency ratio (PER) is equivalent to the live weight gain divided by the protein fed. All data was compiled into a one-way analysis of variance (ANOVA; Ogunji and Awoke, 2017).

Fingerlings emplaced into the greenhouse experimental tanks experienced mortality once the water temperature reached 40° C. All fingerlings in the 40°C water were deceased on the 8<sup>th</sup> day of habitat exposure. No significant mortality was observed in the other experimental areas. Temperature values differed significantly (p < 0.05) between the three locations, with the

greenhouse recording the highest temperature. The water levels were similar among the three locations. Dissolved oxygen, nitrate, nitrite, pH, and ammonia were not significantly different in each location (p < 0.05; Fig. 1). MML201 represents the laboratory tank 20L treatment, MMO202 represents the outdoor tank 20L treatment, and MMG203 represents the greenhouse tank 20L treatment (Fig.1, 4, 5, and 6).

Parameter	MML20 <sup>1</sup>	MMO20 <sup>2</sup>	MMG20 <sup>3</sup>
Temp (°C)	26.53±0.01b	$26.06{\pm}0.01^{a}$	$31.52{\pm}0.00^{\circ}$
DO (mg/l)	5.17±0.14 <sup>a</sup>	5.33±0.17 <sup>a</sup>	4.65±0.57 <sup>a</sup>
pH	6.71±0.12 <sup>a</sup>	6.72±0.12 <sup>a</sup>	7.16±0.17 <sup>a</sup>
Nitrate (mg/l	) 1.96±0.05 <sup>a</sup>	1.96±0.05 <sup>a</sup>	1.62±1.11ª
Nitrite (mg/l)	0.02±0.01 <sup>a</sup>	0.02±0.01ª	$0.00{\pm}0.00^{a}$
Ammonia	2.56±0.72 <sup>a</sup>	2.37±0.14 <sup>a</sup>	3.00±0.00 <sup>a</sup>
(mg/l)			

Figure 2. Figure reproduced from Ogunji and Awoke, 2017. Table depicts the parameters for water quality in each of the experimental tanks for each treatment. Units for temperature were  $^{\circ}C$  and mg/l for every other parameter. The only exception being pH. Values in the same row with different superscripts are significantly different (p<0.05).

The nutrient composition of the maggot meal and the soybean meal used in the

experiment is presented in Fig. 2. The experimental diet used was composed of 41.97% crude

protein with only 3.56% of the diet being crude fat (Fig. 3).

Proximate components (%)	Maggot meal (MGM)	Soybean meal (SBM)
Crude protein	44.87	43.78
Crude fat	7.38	3.67
Crude fibre	6.88	6.48
Crude Ash	7.95	5.96
Moisture content	7.35	5.63
NFE	25.57	34.48

Figure 3. Figure reproduced from Ogunji and Awoke, 2017. Table depicts the composition of both the maggot meal (MGM) and the soybean meal (SBM) used for the fingerling diet. Table

separates the components by protein, fat, fiber, ash, moisture content, and nitrogen-free extract.

Parameter	Diet
Dry Matter	91.95
Crude protein	41.97
Crude fat	3.56
Crude fibre	2.54
Crude Ash	7.69
Moisture content	8.05
NFE	36.19

*Figure 4. Figure reproduced from Ogunji and Awoke, 2017. Table breaks down the components of the experimental diet used in the study. Like other figures, NFE is the nitrogen-free extract.* 

Fingerlings that were fed the experimental diet during the 56 day period experienced an increase in their initial mean weight from 4.33g to 13.10g. Individual measurements from MML20 fish displayed the highest MWG and SGR, followed by fish from the MMO20 group. MWG was the highest in MML20 (Fig. 4). MMG20 had no data for all parameters because all the fish died before measurements were taken. No significant difference was found in PER between either group (Fig. 4).

Parameters	MML20	MMO20	
Initial Weight (g)	4.34±0.26 <sup>a</sup>	4.33±0.03ª	
Final Weight (g)	13.10±1.47°	11.17±1.93°	
Weight Gain (g)	8.76±1.21°	6.83±1.94°	
FCR <sup>1</sup>	$1.61\pm0.10^{b}$	1.67±0.73 <sup>b</sup>	
SGR <sup>2</sup>	1.96±0.10 <sup>c</sup>	1.64±0.30°	
PER <sup>3</sup>	0.21±0.03°	0.16±0.05°	

Figure 5. Figure reproduced from Ogunji and Awoke, 2017. Table details the growth performance for the fingerlings fed the experimental diet. The values are means based on the feeding groups and locations. Values in the same row with different superscripts are significantly different (p < 0.05).

Dietary protein was higher in all the treatments in relation to its initial value. Fish from MMO20 garnered more body protein. This was significantly different compared to the other treatments (p < 0.05; Fig. 5). Moreover, crude ash, crude fiber, crude fat, and moisture were significantly lower from the beginning measurements (p < 0.05; Fig. 5).

Compo- nents (%)	Initial status	MML20	MMO20
Crude	$58.97 \pm 0.00^{a}$	63.97±0.06 <sup>b</sup>	71.28±0.00 <sup>e</sup>
protein			
Crude fat	$8.25 \pm 0.01^{d}$	$5.87 \pm 0.06^{a}$	6.89±0.00°
Crude ash	$12.49 \pm 0.01^{d}$	$7.58 \pm 0.06^{a}$	$9.07 \pm 0.00^{b}$
Moisture	8.4±0.01 <sup>e</sup>	6.68±0.06°	5.25±0.00 <sup>a</sup>
NFE <sup>1</sup>	11.71±0.01°	$15.90 \pm 0.14^{d}$	7.51±0.00 <sup>a</sup>

Figure 6. Figure reproduced from Ogunji and Awoke, 2017. Table is used to determine the difference in composition of the carcass from the initial and final measurements for the fingerlings. All values are means for each group. Values with different superscripts in the same row are significantly different with p < 0.05. NFE means were determined using this formula: NFE = 100 - (% protein + % fat + % ash).

The PCV significantly decreased for all treatments (p < 0.05; Fig. 6). Hb measurements were also significantly different for all locations (p < 0.05; Fig. 6). RBC measurements

significantly decreased from their initial status for each experimental group, however the

treatment groups did not differ significantly in WBC values (Ogunji and Awoke, 2017).

Parameters	Initial	MML20	MMO20
PCV (%)	26.00	20.00	20.00
	$\pm 0.58^{b}$	$\pm 1.54^{a}$	$\pm 0.58$ <sup>a</sup>
Hb (g/100 ml)	8.70	6.60	6.60
	$\pm 0.58^{a}$	±0.23ª	$\pm 0.17^{a}$
WBC (10 <sup>3</sup> ) mm <sup>-3</sup>	78.00	57.00	58.00
	$\pm 57.74^{ab}$	$\pm 230.94^{a}$	$\pm 173.20^{a}$
RBC (10 <sup>3</sup> ) mm <sup>-3</sup>	6.40	3.80	6.90
	$\pm 0.12^{a}$	$\pm 0.17^{a}$	$\pm 0.14^{a}$
MCHC (%)	30.56	33.09	33.01
	±3.13 <sup>a</sup>	$\pm 0.76^{a}$	$\pm 0.09^{a}$
MCH (pg)	7.36	17.93	9.58
	$\pm 0.18^{a}$	±0.71°	±0.41 <sup>b</sup>
MCV (fl)	40.62	52.57	29.00
	$\pm 0.17^{b}$	±0.65°	$\pm 1.34^{a}$

Figure 7. Figure reproduced from Ogunji and Awoke, 2017. Table depicts the hematological data from the subjects. Values presented are means of the feeding groups and different superscripts within the same row represent significant difference (p < 0.05).

In relation to the previous study, more recent research was conducted in 2021. This was done by Kasihmuddin, Ghaffar, and Kumar Das in Malaysia. Two hundred *Clarias gariepinus* fingerlings were gathered from a local fish supplier and transported to the laboratory of University Kebangsaan Malaysia. Each fingerling had a mean weight of  $15.0g \pm 2.0g$ . The fish were acclimated in 26°C temperature water for one week. 180 of the fish were randomly divided into 12 recirculating 94L aquaria, each tank holding 15 fish. The fingerlings were divided into four groups, and each was exposed to a different experimental temperature of 26, 28, 30, or  $32^{\circ}$ C. The tanks were adjusted 1°C until they reached the experimental temperature. Once the respective tank reached its desired temperature the fingerlings were given an additional two days to acclimate. After that, initial body weight measurements (W<sub>i</sub>) were taken. The fish were

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anesthetized using  $\alpha$  – methyl quinoline prior to being weighed. The experiment began immediately after W<sub>i</sub> was taken. The study was carried out for 70 days from February until April. They were fed 2% of their mean body wet weight twice a day. The amount was adjusted every week to sustain continued growth. Pellets that were uneaten were collected daily for calculations. Water level, pH, salinity, and photo-regimen were monitored and sustained throughout to prevent inaccurate results. Fish were weighed weekly for dietary adjustments and calculation purposes (Kasihmuddin et al., 2021).

Records were kept using several different calculations.  $W_f$  represented final body weight, n was the total number of fish that survived at the end of the experiment, and t was the time in days. Body weight gain (BWG) was measured in grams using:  $BWG = (W_f - W_i) x n$ . Specific growth rate (SGR) was calculated using SGR =  $(\ln W_f - \ln W_i) / t x 100$ . Relative growth rate (RGR) was calculated with RGR =  $(W_f - W_i) / W_i x 100$ . Daily Growth Rate (DGR) was done using DGR =  $(W_f - W_i) / t x 100$ . Survival percentage was done by dividing n by the initial number of fish stocked multiplied by 100. Food consumption (FC) in grams per day was done using FC = food consumed (g) x t<sup>-1</sup>. Food conversion ratio (FCR) was a percentage calculation to measure how well the fingerlings can convert the feed into body mass, FCR = total feed fed / total weight gained. Lastly, food conversion efficiency (FCE), different from FCR, is the ability of the fish to convert the food into body mass, FCE = (Weight gain / Feed intake) x 100 (Kasihmuddin et al., 2021).

After final weight measurements were taken, fish were returned to their respective tanks and starved for 72 hours. Fish from each experimental group were chosen for gastric emptying time (GET) analysis. After feeding, five fish from each group were analyzed at every predetermined time. Fish guts were analyzed every two hours starting right after feeding up until

18 hours after feeding. The fish guts were photographed to track the movement of the food, once photographed the fish were fin clipped and returned to their tank to recover. A one-way analysis of variance (ANOVA) was completed once all data had been collected (Kasihmuddin et al., 2021).

A rise in temperature from 26 °C to 32 °C showed no significant difference (p > 0.05) when it came to the following parameters: W<sub>i</sub>, W<sub>f</sub>, BWG, SGR, RGR, DGR, survival percentage, and FC (Fig. 7). The initial weight showed no significant difference (p > 0.05) supporting the validity and accuracy of the experiment. Both FCR and FCE were affected by a rise in temperature and significantly different (p < 0.05; Fig. 7). FCR and FCE registered the highest at temperatures of 30°C and 32°C (Fig. 7).

Parameters	26 °C	28 °C	30 °C	32 °C	p-Value
W <sub>i</sub> (g)	$18.14 \pm 1.87$	$19.00 \pm 1.22$	$20.33 \pm 1.26$	$19.78\pm0.79$	0.26
$W_{f}(g)$	$50.57 \pm 4.12$	$50.31 \pm 5.79$	$53.09 \pm 5.21$	$49.18 \pm 4.75$	0.81
BWG (g)	$486.43 \pm 36.28$	$438.43 \pm 64.51$	$458.61 \pm 57.19$	$416.41 \pm 56.00$	0.82
SGR (% $day^{-1}$ )	$1.50\pm0.05$	$1.32\pm0.07$	$1.30\pm0.09$	$1.26\pm0.08$	0.10
RGR (%)	$187.59 \pm 10.50$	$155.86 \pm 12.19$	$154.28 \pm 14.50$	$146.16 \pm 13.84$	0.11
DGR (% $day^{-1}$ )	$46.33 \pm 3.46$	$44.74 \pm 6.58$	$46.80 \pm 5.83$	$42.49 \pm 5.71$	0.94
Survival (%)	$100 \pm 0.00$	$95.56 \pm 2.22$	$95.56 \pm 4.44$	$93.33 \pm 3.85$	0.72
FC (g day $^{-1}$ )	$0.76\pm0.06$	$0.77\pm0.11$	$0.84 \pm 0.10$	$0.85\pm0.11$	0.87
FCR	$2.01 \pm 0.03$ <sup>a</sup>	$1.79 \pm 0.03$ <sup>ab</sup>	$1.72 \pm 0.04$ bc	$1.64 \pm 0.02$ c	< 0.05
FCE (%)	$49.85 \pm 0.68$ <sup>a</sup>	$55.93 \pm 0.80$ <sup>b</sup>	58.36 ± 1.39 bc	$61.10 \pm 0.66$ c	< 0.05

Figure 8. Figure reproduced from Kasihmuddin et al., 2021. Table includes the measurements and calculations regarding the growth performance and feed utilization for the fingerling test subjects for each experimental group. Values are all means  $\pm$  standard error of mean values per treatment. Rows that have different superscripts indicate those that are significantly different (p < 0.05).

The results also determined the GET of C. gariepinus is very dependent on the

temperature (Fig. 8). As temperature increased from 26°C to 32°C the GET shortened by an

interval of two hours (Fig. 8). The 32°C experimental group had the fastest digestion, in which

food was completely digested by the 10<sup>th</sup> hour. The 30°C experimental group completely digested its food after 12 hours, the 28°C experimental group took 14 hours and the 26°C experimental group took the longest at 16 hours (Fig. 8).

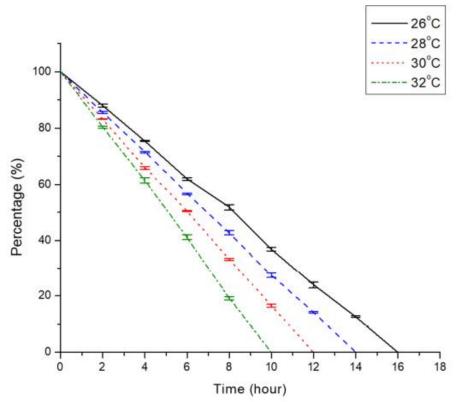


Figure 9. Figure reproduced from Kasihmuddin et al., 2021. Figure graphically illustrates the percentage of food remaining in the fish's digestive system over the time in hours. It is divided into two-hour intervals and each experimental group is represented by a different textured line. The black solid line is 26°C, the blue dashed line is 28°C, the red dotted line is 30°C, and the green dashed and dotted line represents 32°C. Error bars are present to depict the SE for the mean percentage values at each time point.

#### **Discussion**

Results suggest inconclusive and differing responses for *Clarias gariepinus* when present in warmer water conditions. In the first experiment conducted by Ogunji and Awoke, all the experimental fish died just eight days after exposure. The temperature had reached 40°C at this time and temperature that surpasses the optimum limits is detrimental to the wellbeing of the fish. This results from metabolic strain and negatively affects immunity levels, reproductive capacity, and growth (Cnaani, 2006). Other research done on the *Clarias gariepinus* determined that in extreme temperatures the species is unable to adapt and respond quick enough physiologically to survive. This leads to the mortality observed from changes in osmoregulatory functions and metabolic pathways (Adeyamo et al., 2003).

The most successful growth performance in Ogunji and Awoke's study was seen in fish from the experimental laboratory treatment tanks. However, FCR was not statistically significant between the treatment groups. Fish from the outdoor experimental group, that registered the lowest water temperature, were able to accumulate more body protein than the laboratory treatment group. In the laboratory experimental group, an overall decrease in the number of erythrocytes was observed. A reduction in Hb and PCV was detected from the initial status. However, none of these values fell outside the typical hematocrit range established by Dienye and Olumuji in 2014. Decreased PCV is indicative of a shortfall in the expected levels of RBC. A rise in temperature hinders RBC ability to carry oxygen and results in an overall increase in RBC count to compensate. A decrease in erythrocyte count not only causes a low oxygen availability but also has a cascade of metabolic consequences (Gross et al., 1996). Overall, fish that inhabited the ambient laboratory conditions performed better than the other treatment groups (Ogunji and Awoke, 2017).

In the second study completed by Kashimuddin et al., no significant difference was seen in the growth parameters at the various temperatures observed. There was no significant difference in the initial weight recorded to ensure experiment accuracy. Based on this it supports similar research that the optimum water temperature for the growth of the African catfish is between 26°C and 32°C. FCR and FCE increased as temperature rose. Appetite increases as temperature rises; this causes FCE to rise. A decreased FCR meant that the fish were able to utilize feed more efficiently, increase protease activity, and increase protein digestibility (Kashimuddin et al., 2021). Although the demonstrated increase in FCR and FCE was consistent with other studies the concern is the lack of significant difference in growth parameters. No significant difference was seen in growth parameters despite the feed utilization increase. Most likely, these values could have been skewed due to the mortality from cannibalism observed in every treatment group except those at 26°C. Genetic background could also have played a role in the FCE and FCR calculations (Kashimuddin et al., 2021).

GET of the African catfish was determined to be very reliant on temperature range in the experimental group (Kashimuddin et al., 2021). As water temperature rose, the gastric emptying time decreased 2 hours for every 2°C the water temperature increased. The experimental group inhabiting 32°C conditions took 10 hours to evacuate feed while the group inhabiting 26°C took 16 hours. GET was quicker as temperature rose because it was accompanied by an increase in digestive enzyme activity (Kashimuddin et al., 2021). This trend directly correlates with a study done on channel catfish that as FCE increased so did GET (Suja et al., 2009).

In addition to both studies, more detailed research needs to be completed that examines larger physiological parameters. The effect on overall growth for both studies was overall

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statistically insignificant. The first study could have benefitted from a fourth group with a median water temperature to provide more experimental data. The second study could introduce wider parameters of temperature to examine the exact optimum temperature for *C. gariepinus* development. Hematological decreases were seen in one study, but data was limited because there was a lack of temperature differentiation. A wider range of temperature in the tanks may have seen the hemoglobin, PCV, and RBC counts continue to lower outside the normal range. The experimental model of Kashimuddin et al. could be implemented with the tests from Ogunji and Awoke's study to paint a better picture of the hematological trend.

In 2019, a similar study was completed on largemouth bass populations to examine the effect environmental warming had on their growth and metabolism. Conversion efficiency was significantly higher and metabolic resting rate was significantly lower in populations inhabiting 30°C than those in 24°C water (White and Wahl, 2019). They found the reason is that a lower metabolic resting rate provides a greater supply of energy available for other processes. Their research determined that metabolic resting rate and growth are linked and can attribute to a higher FCE (White and Wahl, 2019).

Another study with freshwater trophic cascades in 2016 was done over 12 years and examined phytoplankton, zooplankton, planktivorous and piscivorous groups. They studied lakes that were naturally heated from their use in power plant cooling. The warmer waters demonstrated no significant differences in the phytoplankton population (Mulhollem et al., 2016). Warmer temperatures provide earlier algal growth but greater performance for herbivores may negate this. However, a significant decrease was seen in zooplankton populations present in waters over 30°C. This can be due to several factors. Hatchling abundance has been seen to decrease when spring temperatures occur earlier. Elevated temperatures can reduce the number

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of habitable locations for temperature-sensitive zooplankton. Lastly, the decrease could have been due to an increased predation from planktivorous fish. Since most fish are ectothermic it is expected that ontogeny will be affected by elevated temperatures (Mulhollem et al., 2016). Fish in the warmer lakes exhibited earlier onset of spawning. The peak spawning period occurred one month sooner. A lower density of planktivorous fish was observed in the warmer temperatures. The piscivorous species experienced greater growth most likely from less drastic winter temperatures and a longer growing season. Largemouth bass populations from the heated lakes were on average 20mm longer than their ambient counterparts (Mulhollem et al., 2016).

A simulated global warming environment was studied using rainbow trout in 2001 and made several conclusions. It determined that fish living in the upper threshold of their thermal tolerance could be severely at risk to global warming. They discovered that in the late summers, when water temperatures were highest, an increase of 2°C inhibited growth and appetite despite an excess of food available (Morgan et al., 2001). However, in the winter these populations benefitted from a 2°C increase in temperature seen with stimulated metabolism, appetite, and growth up to 60%. This study also discovered that fish with prolonged exposure to a 2°C increase in temperature exhibited a higher lethal temperature than fish from ambient populations. This means a gradual environmental warming may allow for fish to adapt. They concluded that although there may be metabolic benefits to these populations in the winter, fish living in the upper threshold of their thermal tolerance could see devastating effects in the summer (Morgan et al., 2001).

The effect environmental warming has on freshwater life is varying. Some species like largemouth bass benefit from increased temperatures while zooplankton populations suffer. African catfish in these studies did not have any adverse effects when within temperatures below

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40°C. This could make them even more popular in aquaculture for the future. Overall, this can be worrisome for freshwater biodiversity. It can make the importance of farmed fisheries even more prevalent in society with a growing world population. More research needs to be conducted on the thermal threshold for C. gariepinus to ensure their success. Without C. gariepinus, it could create a hunger issue in several regions that rely on their production (Mulligan, 2015). An isolated study just on the growth variation at different temperatures needs to be done to establish a optimum thermal temperature range. An examination on the resting metabolic rate like White and Wahl's experiment could be beneficial. If temperatures continue to rise it could reach a level that inhibits the growth of the African catfish to a point that another species may be more advantageous to farm. A more recent study should also be conducted on the effect environmental warming has on the entire aquatic ecosystem. More temperature values should be added to provide accurate trend depictions. Additional research can also be done to evaluate the effect warmer temperatures have on population density. Warm water is unable to withhold the oxygen levels that cooler waters do. Could this influence the survivability of populations and create a population cap? Spawning rate was examined by Mullhollem et al. but not in African catfish populations. They are also ectotherms so their ontogeny and physiology can be altered by temperature changes. If changes were seen in planktivorous populations spawning, then changes may be seen in C. gariepinus populations.

Lastly, the only way to prevent all these potential issues yet to be studied is to implement environmental warming mitigation immediately. Temperatures are rising globally at an accelerated rate (Shuhua, 2021). The research on the freshwater ecological effects is inconclusive. Lethality was demonstrated in elevated temperatures for the African catfish, so other fish may have a lower lethal temperature (Ogunji and Awoke, 2017). An increase in

research in this field can only benefit the issue. More awareness means greater support. The work being done in the Amazon to reduce deforestation and biofuels was influenced by the public support pushing government officials. Extensive research can allow governments to make urbanization decisions while considering the ecological ramifications. Mitigation of environmental warming is the only way to protect aquatic ecosystems (Fearnside, 2009).

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