



Research Article.

Effects of crude extracts and the aqueous fraction of *Carica papaya* leaves on the zootechnical performance and survival of juvenile *Clarias gariepinus* raised in soilless tanks in Cameroon

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ABSTRACT

High mortality rates among juvenile *Clarias gariepinus* due to disease pose a major constraint to aquaculture development in sub-Saharan Africa. Excessive use of synthetic antibiotics leads to risks of bacterial resistance and environmental pollution. This study evaluates the effect of crude extracts and the aqueous fraction of *Carica papaya* leaves administered through immersion on the growth and survival of juvenile *C. gariepinus*. One hundred and twenty (120) juveniles with an initial mean weight of 19.95 ± 0.70 g were divided into three duplicate treatments: control (T0), crude extract (T1, $3 \text{ ml} \cdot \text{L}^{-1}$), and aqueous fraction (T2, $3 \text{ ml} \cdot \text{L}^{-1}$). The trial lasted 28 days. The parameters evaluated were survival rate, weight gain, specific growth rate, and feed conversion ratio. The highest survival rate was recorded with treatment T2 ($92 \pm 7.5\%$), while the maximum average weight gain was also observed in this treatment. Due to limited replication (duplicate tanks, $n=2$), these results should be considered preliminary and require validation with a higher number of replicates.

KEYWORDS

Clarias gariepinus;
Carica papaya;
Plant extracts;
Survival;
Growth;
Sustainable Aquaculture.

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1. INTRODUCTION

Aquaculture potentially contributes to food security and poverty reduction in developing countries (FAO, 2022). However, bacterial disease outbreaks constitute a major threat to its sustainability in tropical regions, leading to significant economic losses. African catfish (*Clarias gariepinus*) is the most farmed species in West and Central Africa, playing a vital economic role for rural communities (Kenfack *et al.*, 2019). Bacterial infections, particularly those caused by *Aeromonas hydrophila* and *Pseudomonas fluorescens*, can cause mortality rates of up to 50-80% in intensive farming systems, thereby compromising food security and farmers' livelihoods (Renault *et al.*, 2021). Intensive use of synthetic antibiotics remains the common response to these health problems, but this practice promotes antimicrobial resistance and contamination of aquatic ecosystems (FAO, 2020). *Carica papaya* leaves represent a potential sustainable alternative (Srivastava *et al.*, 2016; Ukwé *et al.*, 2021), as the latter contain bioactive compounds such as carpaine and flavonoids with documented antimicrobial and immunostimulant properties (Akah *et al.*, 1997; Yujun *et al.*, 2003; André Scalbert, 1991). Although the toxicity effects of *Carica papaya* seed extract on *Clarias gariepinus* has been investigated in several studies (Eyo, 2013; Ayotunde *et al.*, 2010; Oparaku *et al.*, 2024), the optimal dose-response effects of *C. papaya* leaf extracts

administered through immersion on *Clarias gariepinus* growth and disease resistance remain to be quantified, notably in the Cameroonian region. This study aims to evaluate the effects of crude and aqueous papaya leaf extracts administered through water bath immersion on growth performance, feed conversion ratio, and survival of juvenile *Clarias gariepinus*.

2. MATERIALS AND METHODS

2.1. STUDY LOCATION

The study was conducted in the Central Region, Mfoundi Department, Yaoundé IV District, and more specifically in the Mimboman neighborhood (Fig. 1). The latter is located between 3°31'0" - 3°62'06" North Latitude and 11°26'38" - 11°45'11" East Longitude and at an average altitude of 712m.

2.2. BIOLOGICAL MATERIAL

A total of one hundred and sixty (160) collected juveniles of *Clarias gariepinus* with an initial mean weight of 19.95 ± 0.70 g, were acclimated for two weeks in the laboratory. During the acclimation period, the fish were fed regularly (three times daily (8:00 AM, 1:00 PM, 4:00 PM) with the Imported Blue Crown brand, which was distributed manually, according to recommendations by Van (1995) and Uys (1999). The water was continuously oxygenated using aquarium aerators.

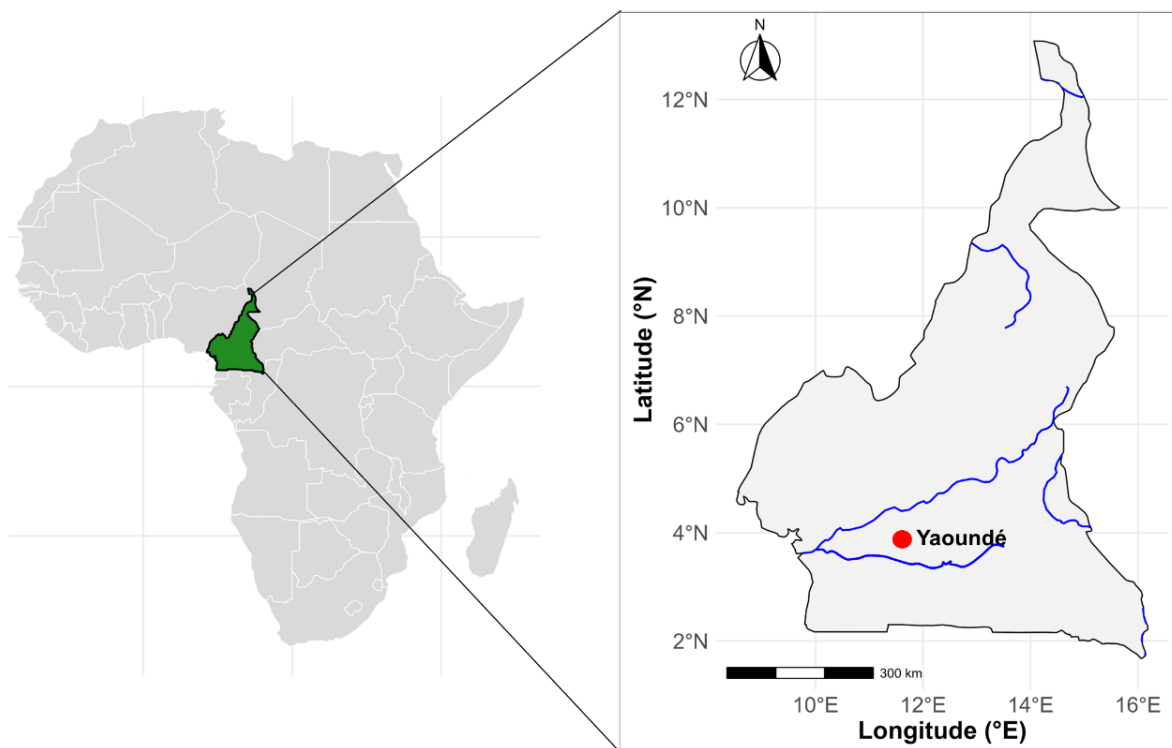


Figure 1. Location of the study area

2.3. PROCESS FOR OBTAINING CRUDE EXTRACTS AND THE AQUEOUS FRACTION OF (*CARICA PAPAYA*) LEAVES

To achieve the set objectives, papaya leaves were harvested, dried at ambient temperature (30°C) for two weeks, then round into powder and weighed (precision balance 0.1 g). Extraction was performed by maceration: 3 kg of powder were macerated in 10 L of 96% methanol in a hermetically sealed container for two days at room temperature. The mixture was filtered through cotton.

The solvent was subsequently removed using a rotary evaporator under reduced pressure, a technique commonly employed to condense plant extracts while minimising the thermal degradation of bioactive compounds (Azmir *et al.*, 2013; Dai & Mumper, 2010), yielding a concentrated crude

extract (Sulistianto, 2004). For the aqueous fraction, 50% of the crude extract was fractionated by adding distilled water (equal proportions), then steam-distilled to separate insoluble compounds. The solution was centrifuged at 3000-4000 rpm for 10-15 minutes, then decanted to recover the supernatant (aqueous fraction) stored under refrigeration.

2.4. PHYTOCHEMICAL ANALYSIS

The phytochemical compounds were analyzed by cold water extraction according to standard procedures (Nwamarah *et al.*, 2019). The total alkaloid content was determined by spectrophotometry. The total alkaloid, flavonoid, saponin, and tannin contents are expressed in Table 1.

Table 1. Phytochemical composition of papaya leaf extracts

Phytochemical composition	Concentration (mg/100g)
Flavonoids	866.53 ± 27.24
Alkaloids	1569.13 ± 92.58
Saponins	898.07 ± 20.67
Tannin	310.50 ± 11.51

2.5. EXPERIMENTAL SETUP

The experimental setup consisted of six 80-liter tanks connected in parallel, each containing 30 liters of groundwater and 20 juvenile fish. Due to facility constraints, replication was limited to duplicate tanks per treatment (n=2). Results should therefore be

considered preliminary and validated in future trials with at least three replicates. The three treatments (Table 2) were completely randomized.

Juvenile *C. gariepinus* were divided into three groups, each in duplicate. Water was changed every two days. Control fishing was conducted weekly in the morning after fasting to avoid the effect of undigested feed on weight. Mean weight was determined by individual weighing of specimens (scale precision ±1 g). The tanks were cleaned by scrubbing the sides with a clean sponge to remove feed residue and ensure proper water aeration. The water temperature and pH were measured using a multi-parameter monitor before and after the administration of the products.

Table 2. Characteristics of the different treatments

Treatments	Method of administration	Dose (ml/l)	Duration (h)
T0 (control lot)	None	0	0
T1 (crude extract of papaya leaves)	Bath	3	48
T2 (aqueous fraction of papaya leaves)	Bath	3	48

2.6. DATA PROCESSING AND STATISTICAL ANALYSIS

2.6.1. Growth and Survival Characteristics

The growth performance and survival of the experimental organisms were evaluated based on standard quantitative indicators. The parameters provided an assessment of the effects of the experimental conditions on the organisms' performance over the study period, using the

following formulas:

Survival rate (SR in %):

$$SR = (\text{Final number} / \text{Initial number}) \times 100$$

Weight gain (WG in g):

$$WG = \text{Mean final weight} - \text{Mean initial weight}$$

Specific growth rate (SGR in %/d):

$$SGR = [(\ln \text{Final weight} - \ln \text{Initial weight}) / \text{Time}] \times 100$$

Feed conversion ratio (FCR):

$$FCR = \text{Feed distributed} / \text{Weight gain}$$

2.7. STATISTICAL ANALYSIS

Data were analyzed by one-way ANOVA at the 5% significance level. Due to limited replication ($n=2$), statistical results should be interpreted with caution and considered as descriptive trends.

3. RESULTS

3.1. EFFECTS OF CRUDE EXTRACTS AND AQUEOUS FRACTION OF *CARICA PAPAYA* LEAVES ON THE SURVIVAL RATE OF JUVENILE *CLARIAS GARIEPINUS*

The survival rate during the experiment is shown in Figure 2. It reveals that the lowest survival rate was recorded in the group treated with the crude extract (T1), with a rate of $77.50 \pm 12.50\%$, compared to those treated with the aqueous fraction (T2), with a rate of $92 \pm 7.50\%$, and the subjects in the control group (T0), with a rate of $90.00 \pm 6.66\%$. The aqueous fraction treatment resulted in the highest survival rate of 92.5%.

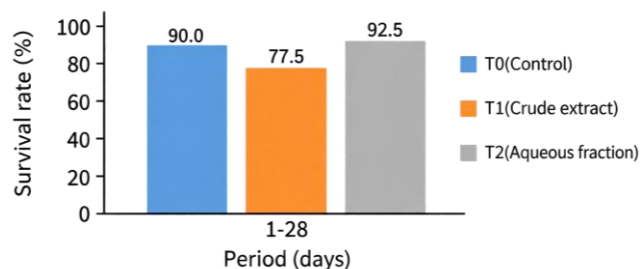


Figure 2. Survival rates as a function of time period

3.2. EFFECTS OF CRUDE EXTRACTS AND AQUEOUS FRACTION OF *CARICA PAPAYA* LEAVES ON THE AVERAGE WEIGHT GAIN OF JUVENILE *CLARIAS GARIEPINUS*

The mean gain values obtained with the different treatments are illustrated in Figure 3. Subjects in the control group (T0) recorded a lower mean value (15.13 ± 0.51 g), while those treated with the crude extract (T1) obtained a mean value (15.25 ± 0.077 g) close to that obtained in the group treated with the aqueous fraction (15.76 ± 0.60 g). However, the group treated with the aqueous fraction (T2) recorded the highest mean value.

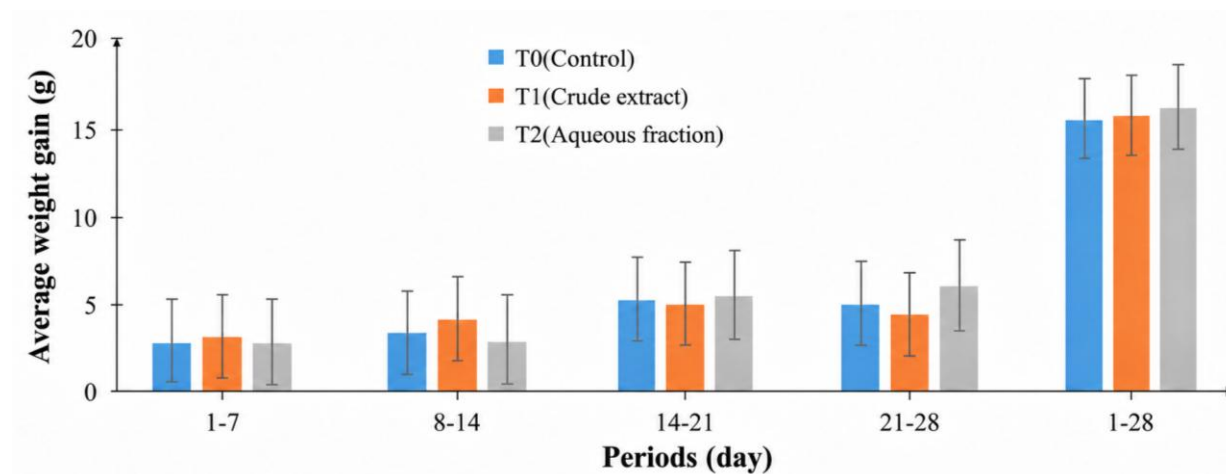


Figure 3. Evolution of weight gain over time

3.3. EFFECTS OF CRUDE EXTRACTS AND AQUEOUS FRACTION OF *CARICA PAPAYA* LEAVES ON THE SPECIFIC GROWTH RATE OF JUVENILE *CLARIAS GARIEPINUS*

Figure 4 shows the evolution of the specific growth rate as a function of the control fishing periods. Throughout the trial period, the specific growth rate increased during the first three weeks in each of the groups; the group treated with the crude extract (T1)

had the highest rate during the first two weeks, slowed down in the third week, and decreased at the end of the period with an average rate of $1.60 \pm 0.09\%$ g/day. The group treated with the aqueous fraction (T2) had a value of $1.45 \pm 0.08\%$ g/day, and the control group (T0) had a value of $1.51 \pm 0.03\%$ g/day. However, the group treated with the aqueous fraction (T2) exhibited the best performance.

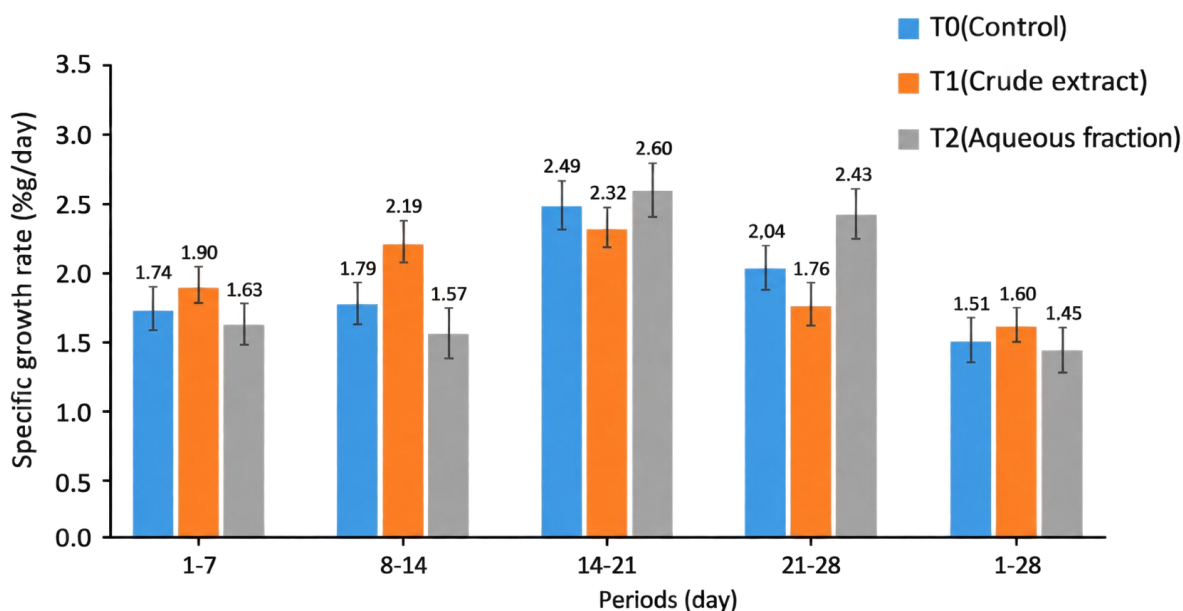


Figure 4. Specific growth rate as a function of time

4. DISCUSSION

The results obtained showed that the average survival rates recorded during the experiment were $77.5 \pm 12.5\%$ for treatment T1 (crude extract) and $92 \pm 7.5\%$ for treatment T2 (aqueous fraction). These values differ from those obtained by Ayotunde *et al.* (2011), who reported 85–90% and 70–88% for the two treatments, respectively. These results are relatively similar to those obtained by Afolabi *et al.*

(2014), who reported rates of 90–95% and 75–80% for the two treatments, respectively. This difference could be due to the rearing environment and conditions. The weight gain recorded during the experiment was 15.25 ± 0.07 g for treatment T1 (crude extract) and 15.75 ± 0.6 g for treatment T2 (aqueous fraction). The values obtained differ from those obtained by Ayotunde *et al.* (2011) and Ezekiel *et al.* (2011), who reported results of 230g and 120g, respectively for the two treatments after 8 weeks. On

the other hand, these results contradict those obtained by Ayotunde *et al.* (2011) and Ezekiel *et al.* (2011), which obtained a specific growth rate of 3–3.5% g/d for crude extracts and 2.5–3% g/d for the aqueous fraction. This could be due to the nutrient content of the different foods and the ingredients used. According to the results obtained from our research, the aqueous fraction appears to be more effective than crude extracts, which could be due to a lower concentration of bioactive compounds and the experimental conditions.

5. CONCLUSION

This preliminary study, which focused on the effect of using crude and aqueous extracts of papaya leaves (*Carica papaya*) as a preventative disease treatment on some zootechnical characteristics of juvenile *Clarias gariepinus* fry, aimed to test the hypothesis that preventative disease treatment with papaya leaf extracts could positively impact growth performance and survival rates in *Clarias gariepinus*. The results showed that *Clarias gariepinus* fry treated with the aqueous fraction of papaya leaves exhibited superior growth performance compared to those treated with the crude extract and those in the control group. Considering the limited replication (duplicate tanks, n=2), these results should be considered exploratory and validated by trials with higher replication. Observed growth differences (<1 g) fall within instrumental measurement error, while survival differences (>15%) warrant further investigation on the mechanisms of action of bioactive compounds from papaya leaves in aquaculture.

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