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COMMUNICATION III

The Use of Live, Frozen and Pottasium Permanganate Treated *Moina micrura* for Catfish (*Clarias macrocephalus*) Larvae

ABSTRAK

Moina micrura *yang hidup, yang dibekukan dan yang diolahkan dengan kalium permanganat ($KMnO_4$) telah diberikan kepada lava ikan keli* (Clarias macrocephalus) *selama 20 hari. Pengolahan dengan $KMnO_4$ sebelum dimakan bertujuan untuk mengurangkan paras mikroorganisma yang terdapat di dalam kultur* M. micrura. *Kadar pertumbuhan dan nisbah penukaran makanan untuk* M. micrura *hidup dan yang diolah dengan $KMnO_4$ tidak mempunyai perbezaan bererti.* M.micrura *yang dibekukan mempunyai kadar pertumbuhan yang rendah dan nisbah penukaran pemakanan yang tinggi. Nilai-nilai ini mempunyai perbezaan yang bererti apabila dibandingkan dengan yang diperolehi untuk* M. micrura *yang tidak diolah. Keputusan ini mencadangkan bahawa* M. micrura *masih mempunyai kualiti pemakanan walaupun selepas olahan dengan 1 μM $KMnO_4$ dan ia sesuai sebagai makanan lava ikan.*

ABSTRACT

Live, frozen and pottasium permanganate ($KMnO_4$) treated Moina micrura *were fed to catfish* (Clarias macrocephalus) *larvae for 20 days. The aim of $KMnO_4$ treatment prior to feeding was to minimise the level of microorganisms present in the* M. micrura *culture. The growth rates and feed conversion ratios for the live untreated and $KMnO_4$ treated* M. micrura *were not significantly different. Frozen* M. micrura *had a significantly lower growth rate and significantly higher feed conversion compared with the live, untreated* M. micrura. *The results on relative growth and feed conversion of fry fed the treated and untreated suggest that* M. micrura *retained its nutritive qualities even when treated with 1 μM $KMnO_4$ and is suitable as a larval feed.*

INTRODUCTION

The recent intensification of catfish *Clarias macrocephalus* culture in Malaysia has resulted in increased demand for its fry. However, a major constraint faced by fish farmers presently is low rates of larval survival and growth, often due to fungal and bacterial infection present in the natural food provided (Lau & Plumb 1981, Viveen *et al.* 1986) or unsuitable artificial larval feeds.

Natural food organisms such as *Daphnia pulex* (Hecht 1981) and rotifers (Lubzens *et al.* 1987) have been widely used as first feeds for larvae. However, live feeds have several disadvantages such as seasonal availability and the introduction of pathogens and parasites into the hatchery since they cannot be sterilised (Uys and Hecht 1985). To overcome the availability problem, frozen or freeze-dried natural food can be used but these have been found to be unsuitable (Brett 1971; Fluchter 1980).

Locally, *Moina micrura* is used to feed newly hatched catfish larvae for the first four weeks of life. However, we have experienced inconsistent larval survival when *M. micrura* was used and low larval survival was often due to fungal and bacterial infection such as from *F. columnaris.* The infected fish were treated with pottassium permanganate ($KMnO_4$) because of its therapeutic properties against external bacterial and protozoan infections (Lau and Plumb 1981 and Duncan 1987). Since the fish are stress-sensitive at the larval stage, $KMnO_4$ treatment was often too late and resulted in high mortalities.

In this study, the effectiveness of freezing and $KMnO_4$ treatment on *M. micrura* to obtain optimal growth and survival of larvae as compared to untreated *M. micrura* was tested. *M. micrura* was treated with KMn_4O prior to feeding the larvae with the aim of reducing the level of microorganisms present in the *M. micrura* culture and consequently reducing the risks of larval

infection. Preliminary studies of $KMnO_4$ treated and untreated *M. micrura* cultures to determine the presence of microorganisms has shown the former to contain a lower colony count. Since $KMnO_4$ is a strong oxidant, toxicity problems which frequently arise during $KMnO_4$ treatment were avoided by using a concentration of oxidant that was high enough to eliminate the microorganisms but low enough in toxicity to ensure survival of the Moina and subsequently the larvae.

MATERIALS AND METHODS

Moina micrura was obtained from a nearby moina cultivation farm. Diet 1 was untreated *M. micrura*, Diet 2, *M. micrura* treated with 1uM $KMnO_4$ and Diet 3, *M. micrura* frozen at -400C. $KMnO_4$ treated *M. micrura* (Diet 2) was rinsed thoroughly with water and the frozen *M. micrura* (Diet 3) was thawed at room temperature prior to feeding. The nutrient content of all the three diets was found to be very similar (Table 1).

TABLE 1
Proximate analysis of *Moina micrura*
(% dry weight)

	Diet 1	Diet 2	Diet 3
Crude protein [1]	60.72	61.73	64.05
Crude lipid [2]	18.12	14.45	19.52
Carbohydrate	9.95	13.68	5.67
Ash	11.21	10.14	10.76
Dry matter	7.81	7.49	7.77
Moisture [3]	93.40	92.51	92.23

[1] Measured as Kjeldahl-N x 6.25
[2] Extracted in Chloroform-Methanol
[3] Untreated *Moina micrura*

Clarias macrocephalus larvae were obtained by induced spawning using human chorionic gonadotropin (Mollah & Tan 1983). The larvae were transferred from the breeding tanks to the experimental aquariums on the final day of yolk absorption and before exogeneous feeding started. The nine experimental aquariums were filled with 30 liters of fresh water, aerated and stocked with 100 larvae. Water exchange rate was 5 lhr^{-1} and the temperature was maintained at 29°C throughout the feeding trial. All experiments were carried out with three replicates per treatment.

The larvae were fed to satiation twice daily. Before feeding, fecal matter and excess feeds were siphoned out and half the volume of water drained. The feeding trial was terminated on the 20th day and all the fish in the tanks were counted and weighed. The relative growth rate and the feed conversion ratio (FCR), defined as the total feed taken per unit of fish weight gain, was calculated. The results obtained were analysed using ANOVA at 5% level of significance followed by Duncan's new multiple-range test for comparison of the means obtained (Steel & Torrie 1960).

RESULTS AND DISCUSSION

Survival time of *M. micrura* treated with 1µM $KMnO_4$ was not significantly different from untreated ones. The results at the end of the 20-day feeding trial are summarised in Table 2.

Table 2
Average weight gain, feed conversion and survival rate for *Clarias macrocephalus* larvae fed diets containing different types of *M. micrura*.

	Diet 1	Diet 2	Diet 3
Initial Weight (g)	0.0112	0.0131	0.0092
Final Weight (g)	0.0512	0.0461	0.0362
Weight gain	0.040^{a}	$0.033^{a,b}$	0.027^{b}
Feed Conversion	1.34^{a}	$1.66^{a,b}$	1.87b
Survival	97.0^{a}	95.0^{a}	98.6^{a}

Means in the same row with the same superscript are not significantly different ($P<0.05$).

Both Diets 1 and 2 which consisted of live *M. micrura* were preferred by the larvae compared with a tentative and delayed acceptance of the frozen *M. micrura*. Dabrowski and Bargeda (1984) observed that coregonid larvae accepted live food more readily than artifical diets. The food intake of larvae is dependent on such factors as mobility (Braum 1978), chemical attractants (Applebaum 1979) and physical characteristics such as particle size and texture (Dabrowski & Bargeda 1984) of the feeds. Thus it is possible that the mobile live and $KMnO_4$ treated *M. micrura* (Diets 1 and 2) were chemically and physically more attractive.

The relative growth and survival rates of larvae and the feed conversion ratios for the experimental diets are presented in Table 2. Larvae fed on live untreated (Diet 1) and $KMnO_4$ treated (Diet 2) *M. micrura* had similar growth rates whereas larvae fed on frozen *M. micrura* had the lowest growth rate. Fish fed Diet 1 showed a significantly higher growth rate than that fed Diet 3. However, no significant difference between Diets 2 and 3 was observed. Several observations have been reported regarding the use of frozen zooplankton as an experimental food for fish larvae. Flüchter (1980) reported that *Artemia* remained completely effective as larval food after being slowly frozen, although a gradual loss in an essential growth substance was observed. They found that whitefish larvae completed metamorphosis equally well when fed with *Artemia* that had been shock frozen in liquid nitrogen (-196° C) up to at least 2 hours after thawing or with live *Artemia.* Biochemical analysis of frozen and freeze-dried zooplankton showed that proteases and enzymes of intermediary metabolism were not diminished by freezing, freeze-drying or by storage at -18°C (Grabner *et al.* 1981). However, upon thawing, these freeze damaged cell immediately release as much as 70-75% of proteases and much more of amino acids into the water within 10 minutes. Thus frozen zooplankton is not suitable as a larval diet due to leaching of nutrients once they are thawed.

The feed conversion ratio was lowest for Diet 1 but this was not significantly lower than that of Diet 2. Diet 3 had a significantly higher conversion ratio compared with Diet 1. This is consistent with the fact that the nutritive value of frozen *M. micrura* is lower than that of live of $KMnO_4$ treated samples.

The survival rates of the larvae for all the treatments were high and not significantly different. This suggests that $KMnO_4$ treatment and slow freezing can eliminate or lower the level of pathogens present in the *M. micrura* culture. However, the high survival of larvae fed on untreated *M. micrura* during the experiment suggest that the batch of *M. micrura* used as control was either not highly contaminated or that the fish larvae were physiologically healthy enough to withstand infection. Previous experience using untreated *M. micrura* had often resulted in inconsistencies in larval survival; low larval survival was found to be due to fungal and bacterial infections.

The lack of significant difference in the relative growth rates and feed conversion ratios of larvae fed with untreated and $KMnO_4$ treated *M. micrura* indicated that the nutritive quality of the treated *M. micrura* is comparable to the untreated samples. Hence, instead of treating the larvae with $KMnO_4$ only when they are infected, which is often too late, *M. micrura* itself should also be treated with $KMnO_4$ prior to feeding to avoid microbial infections. Presently, studies are being carried out to determine the lowest level of $KMnO_4$ required to effectively eliminate these microorganisms with minimal toxicity to the *M. micrura* and larvae.

CONCLUSION

The use of $KMnO_4$ to treat *M. micrura* prior to feeding ensures a high survival rate of *Clarias macrocephalus* larvae. It is therefore recommended that *Moina micrura* obtained from farms be treated routinely with $KMnO_4$ to minimise larval infection caused by microorganisms present in this natural food.

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