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Spirulina (*Arthrospira fusiformis*) as a potential protein source in practical diets for fry mariculture of Rufiji tilapia (*Oreochromis urolepis urolepis*)

Deogratias P. Mulokozi^{1,2,*}, Matern S. Mtolera², Aviti J. Mmochi²

¹ Tanzania Fisheries Research Institute, Kigoma Centre, P.O. Box 90 Kigoma, Tanzania

² Institute of Marine Sciences, University of Dar es Salaam, P.O. Box 668, Zanzibar, Tanzania

Corresponding author: deomulokozi@gmail.com

Abstract

The effects on growth performance, feed utilization, survival and whole fish body proximate composition of replacing fishmeal (*Rastrineobola argentea*) with a locally available spirulina species (*Arthrospira fusiformis*) as a protein ingredient in the feed of Rufiji tilapia (*Oreochromis urolepis urolepis*) fry was examined. Fishmeal replacement with *A. fusiformis* was carried out at 5%, 15%, 25%, 35% and 100% (S5, S15, S25, S35 and S100), and the effect of the replacement was compared with the control diet (S0, 0% spirulina). Fish fry were stocked at an average initial weight of 0.57 g at 10 fish per 0.1 m³ and cultured for 60 days using full strength salt water (30-35 ppt). *A. fusiformis* was isolated from Momella Lake in Arusha National Park, Tanzania and propagated using selected culture media. It was observed that fish fry fed spirulina at a 5% fishmeal substitution level diet had better ($p < 0.05$) final weight (8.48), average daily weight gain (0.132), specific growth rate (4.47), feed conversion rate (2.08) and protein efficiency ratio (1.37) compared to the control fish group. Growth performance in fish fed diet S15 was comparable with the control group ($p > 0.05$). Spirulina supplementation had no effect on fish survival rates and meat quality. It appears that the Momella Lake spirulina may be an appropriate growth-stimulating plant protein when used as a feed additive in Rufiji tilapia mariculture.

Keywords: Rufiji tilapia; Spirulina; *Arthrospira fusiformis*; Growth performance; Meat quality; Mariculture

Introduction

Globally, aquaculture is currently the most rapidly growing food production sector, with an annual growth rate of more than 5.8% (FAO, 2018). In line with global aquaculture growth, there is an increased demand for nutritionally sufficient and economically affordable aqua feeds (Thilsted *et al.*, 2016). Protein remains the most expensive ingredient in these feeds and the most crucial factor affecting growth performance of fish (Jose *et al.*, 2007). Thus, one of the constraints to aquaculture sustainability and growth is the shortage of readily available cheap protein sources (Machena and Moehl, 2001). Fishmeal is the most preferred feed supplement in intensive and semi-intensive aquaculture systems (El-Saidy and Gaber, 2004). This is partly because it is considered most palatable and rich in essential amino acids, fatty acids, energy and minerals (Tacon, 1993; Hardy, 2010). However, because global fisheries have leveled off at a time

when there is increasing demand to feed the growing human population and to supply a fast-expanding fish meal industry, the availability of fish meal is also limited (FAO, 2016). Moreover, from an environmental perspective, overfishing the wild fishery for fishmeal production is unjustifiable (Wijkström, 2009).

In the search for new, readily available protein sources, algae are attracting the attention of nutritionists as one possibility to supplement the world protein shortage, particularly in developing countries (Becker, 2007; Roy and Pal, 2015). Cultivated microalgae and cyanobacteria have long been integrated with hatchery production of many farmed finfish, shellfish and other commercially important aquaculture species (Muller-Feuge, 2010). Alongside these well-established microalgae uses in aquaculture hatcheries, there is currently a drive to substitute fishmeal with algae or cyanobacteria in formulated animal feeds, both for aquaculture and

terrestrial animals (Nath *et al.*, 2012; Sheikhzaden *et al.*, 2012). One possibility is to utilize primary producers like spirulina (*Arthrospira spp.*), which supports huge populations of flamingos in Tanzania's soda lakes.

Spirulina are multicellular, filamentous, gram negative, non-toxic species of cyanobacteria belonging to two separate genera, namely *Spirulina* and *Arthrospira*, consisting of about 15 species (Habib *et al.*, 2008). Of these, *Arthrospira spp.* are the most common and widely available, possessing diverse biochemical compounds of biological and nutritional significance such as protein (55-70 %), minerals (calcium and iron) and vitamins (Habib *et al.*, 2008). It grows and thrives in warm alkaline lakes including the soda lakes of East Africa such as Elmenteita, Lishateni and Momela in Tanzania, and Nakuru in Kenya (Grant, 2006; Kaaya *et al.*, 2007). It can be harvested from such lakes and used directly to serve as a protein source in fish feeds, or to seed artificial mass production systems.

Several studies have evaluated *Arthrospira spp.* as a potential fish meal substitution in feeds for tilapia and other fish species (Lu *et al.*, 2002; Takeuchi *et al.*, 2002; Abdel-Tawwab and Ahmad, 2009; Dernekbası *et al.*, 2010; Belal *et al.*, 2012; El-Sheikh *et al.*, 2014; Velasquez *et al.*, 2016). The results show considerable variation in the degree of success for partial and complete replacement depending on the spirulina strains, farmed fish species as well as the growth stage of fish. In addition to their potential use in aquaculture as protein sources, *Arthrospira* species are reported to be a good immunostimulant in fish, poultry and rats (Abdelkhalek *et al.*, 2017; Abdel-Daim *et al.*, 2018; Kata *et al.*, 2018).

Tanzania is endowed with a long coastline, and farming the sea with high value marine species could be more profitable than land-based farms. According to the World Bank (2016), it is estimated that humans will soon run out of areas that will be able to produce enough food to provide for the entire population of earth, due to freshwater scarcity. The same report points out that continuous human population growth will lead to increasing competition for limited water resources, which are likely to become more constrained due to adverse climate change-associated effects such as drought and flooding. It would therefore make sense to explore new ways in which the ocean can be used to cultivate food in the future.

Rufiji tilapia (*Oreochromis urolepis urolepis*) occurs in the southeastern rivers, reservoirs and satellite lakes

in Tanzania, where it is an economically important fish to the communities of these areas (Lamtane, 2008). This species has considerable potential for culture in saline waters in order to expand the farming area, and improve income and livelihoods of coastal communities.

Despite being found mainly in freshwater and brackish environments, *O. urolepis urolepis* can be cultured in salt water without adverse effects on growth and survival rates. For example, Nehemia *et al.* (2014a) reported 100 % survival and acceptable growth performance when *O. urolepis urolepis* was cultured using sea water at 35 ppt. Similar results were reported by Nehemia *et al.* (2014b) and John (2016). On the other hand, Ulotu *et al.* (2016) found best growth and survival rates at a salinity of 25 ppt when *O. urolepis urolepis* was cultured in water with salinity values of 5, 15, 25 and 35 ppt.

Rufiji tilapia, like other tilapiine species, is a typical omnivorous fish species. Therefore, it was hypothesized that spirulina may have potential as a protein source for *O. urolepis urolepis*. Spirulina (*A. fusiformis*) collected from Momela Lake was used in this study as a partial and complete fish meal substitute, and its effects on growth performance, survival rates and biochemical composition were tested on Rufiji tilapia fry cultured in full-strength salt water.

Material and methods

Collection and culture of spirulina

Water samples containing a mixture of microalgae was collected from the Big Momela Lake in Arusha National Park, Tanzania. After sampling, water samples were immediately transported in a cool box to the Department of Botany, University of Dar es Salaam for isolation. Isolation was carried out using serial dilution and streak plating microbiology techniques. Spirulina were identified according to Ciferri (1983) and Komarek and Lund (1990). Isolated spirulina was then cultured outdoors at the Institute of Marine Sciences in Zanzibar for three months.

Cultivation was carried out using ten 100 l half-metal drums lined with a plastic sheet to prevent rust (Fig. 1). Filtered tap water was left for 48 hours to allow for chlorine evaporation and used to make a culture medium composed of the following ingredients (Singh, 2006) per litre: sodium bicarbonate (8 g); potassium sulphate (0.5 g); sodium chloride (5g); magnesium sulphate (0.16 g); ferrous sulphate (0.05 g); Urea (0.2 g); and phosphoric acid (0.2 ml). Inoculation

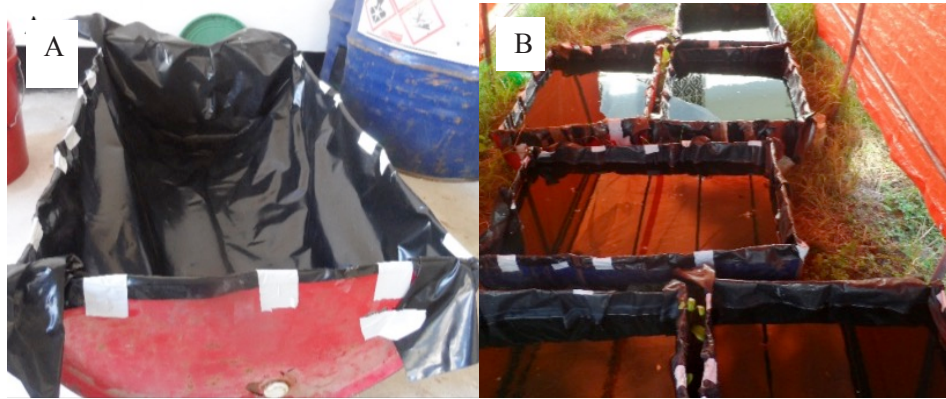


Figure 1. *A. fusiformis* outdoor culture; (A) culture basin preparation; (B) culture unit.

started with 20 l culture medium inoculated with 1 l spirulina inoculum as a starter culture, followed by scaling-up to 90 l within 7 to 10 days (Son *et al.*, 2019). Daily measurements of various pond parameters such as temperature, light intensity and salinity were undertaken to monitor the wellbeing of the culture.

Microscopic examination was done daily to detect any abnormal morphological changes and the presence of contaminating organisms such as other algae and protozoa. Large contaminants like insects, plant leaves and birds were excluded by placing a wire mesh on top of the culture basins. Harvesting was carried out in the morning hours to ensure cool temperatures with enough sunshine to dry the product. After sun-drying, the spirulina was stored in cool, dry environment to preserve its quality.

Fish diets and experimental running

The proximate chemical composition of the feed ingredients used is presented in Table 1. Six isonitrogenous test diets (Table 2) were formulated to contain 35% crude protein. Fish meal and maize bran (obtained from the local market) were prepared with a cereal grinding machine, while spirulina was ground to powder form using a mortar and pestle. In the control set (S0), experimental fish were fed with a 100% fishmeal protein diet. In the remaining treatments, prior-cultured dried spirulina meal protein replaced 5% (S5), 15% (S15), 25% (S25), and 35% (S35) and 100% (S100) of fishmeal protein. The experimental culture

system consisted of a series of eighteen 100 l plastic basins, divided into six triplicate treatments. The fish were cultured using seawater from the Pangani river estuary, pumped during the high tide to ensure high salinity of 33-35 ppt. Rufiji tilapia fry that had been raised in full strength sea water were obtained from the Institute of Marine Science Mariculture Centre (IMS-MC) hatchery at Pangani. Prior to the experiment, fish were acclimatized for two weeks while they were fed the control diet. Thereafter, the fish fry (0.57 ± 0.01 g) were randomly distributed into six experimental groups in triplicate at a density rate of 10 fry per 0.1 m^3 . Fish were fed at 5% of their body weight twice per day, at around 8:00 and 16:00. Complete water exchange was carried out every 10 days. Fish were group-weighted every 10 days and the amount of administered feed was adjusted accordingly. The experiment was conducted for 60 days.

Water quality monitoring

Physical parameters (temperature, pH, dissolved oxygen, and salinity) were checked daily using a hand-held thermo-pH meter (HANNA model no: HI 98128), oxygen meter (YIS Environmental model no: DO 200) and refractometer (EXTECH instruments model no: RF 20), respectively. Water samples for un-ionized ammonia analysis were collected twice a week in 500 ml plastic bottles and stored in a freezer at the IMS-MC before analysis in the laboratory. Concentration of ammonia in the water samples was determined according to the guidelines of UNESCO (1993).

Table 1. Chemical proximate composition (% Dry matter) of the ingredients used in experimental diets.

Ingredients	Dry matter	Crude protein	Crude lipid	Crude fiber	Ash
Fish meal	94.43	63.0	7.62	0	6.51
Spirulina meal	94.63	57.25	7.42	9.14	5.23
Maize bran	89.59	9.23	1.74	7.95	3.8

Table 2. Ingredient and proximate composition of the experimental diets.

Ingredients (g/100g)	Diets					
	Control	S5	S15	S25	S35	S100
Fish meal ^a	51.0	46.0	37.0	29.0	20.0	0.0
Spirulina meal	0.0	5.0	15.03	25.0	35.0	56.0
Maize bran	39.0	39.0	38.0	35.0	34.0	38.0
Sunflower oil	6.0	6.0	6.0	7.0	7.0	6.0
Binder (Cassava)	2.0	2.0	2.0	2.0	2.0	2.0
Premix ^b	2.0	2.0	2.0	2.0	2.0	2.0
Total	100	100	100	100	100	100
Proximate composition (% DM)						
Dry matter	92.22	92.23	91.19	91.87	92.1	92.45
Crude protein	35.19	35.29	35.0	35.19	35.39	35.2
Crude lipid	10.51	10.31	9.84	9.53	9.06	6.59
Crude fiber	3.18	3.14	3.14	3.06	2.98	2.7
Ash	4.78	4.74	4.67	4.6	4.53	4.09
NFE ^c	46.34	46.52	47.36	48.1	48.04	51.42
GE(kcal/kg) ^d	4937	4931.3	4906	4918.6	4884	4781.5

^a Locally occurring Lake Victoria sardine (*Rastrineobola argentea*).

^b Locally manufactured commercial premix (per kg mixture): vitamin: A, 500000 IU; D3, 1000000 IU; E,1500 IU; B1, 600 mg; B2, 2500 mg; B6, 125 mg; B12, 7.5 mg; K,1250 mg; C, 200 mg. Minerals: 1.5 mg, CuSO₄; 90 mg, MnSO₄; 300 mg, MnI₂; 70 mg, ZnO; 5500 mg, C₆H₅NO₃; 5000 mg, C₁₈H₃₂CaN₂O₁₀.

^c NFE (Nitrogen free extract) = 100 - (crude protein + crude lipid + crude fiber + ash).

^d GE (gross energy): calculated using conversion factors 5.65, 9.45 and 4.22 kcal/g for protein, lipids and carbohydrate respectively (NRC,1993).

Growth performance and feed utilization.

After the feeding trial, fish from each basin were collected, weighed, and counted. The parameters of fish growth and feed utilization were calculated according to the following equations:

Average daily weight gain (g/fish) (ADG)

$$ADG = (W2 - W1) / T \dots\dots\dots 1$$

Where: W1 = Mean initial weight (g)
 W2 = Mean final weight (g)
 T= experimental period

Specific growth rate (SGR)

$$SGR = \left(\frac{\ln W2 - \ln W1}{T} \right) \times 100 \dots\dots\dots 2$$

Where: Ln = Natural Logarithm
 W1 = Mean initial weight (g)
 W2 = Mean final weight (g)

Feed conversion ratio (FCR)

$$FCR = \frac{\text{Dry feed intake (g)}}{\text{Live weight gain (g)}} \dots\dots\dots 3$$

Where: Dry feed intake (FI) = Total feed consumed during the 60-day trial

Protein efficiency ratio (PER)

$$PER = \frac{\text{Live weight gain (g)}}{\text{Protein intake (g)}} \dots\dots\dots 4$$

Where: Protein intake = % Crude protein x FI

Proximate analysis

Ingredients, practical diets and whole-body chemical composition were analysed according to AOAC (1984). Dry matter was calculated from weight loss after oven drying of the fresh samples at 105°C for 48 hrs continuously. Ash content was determined by incinerating

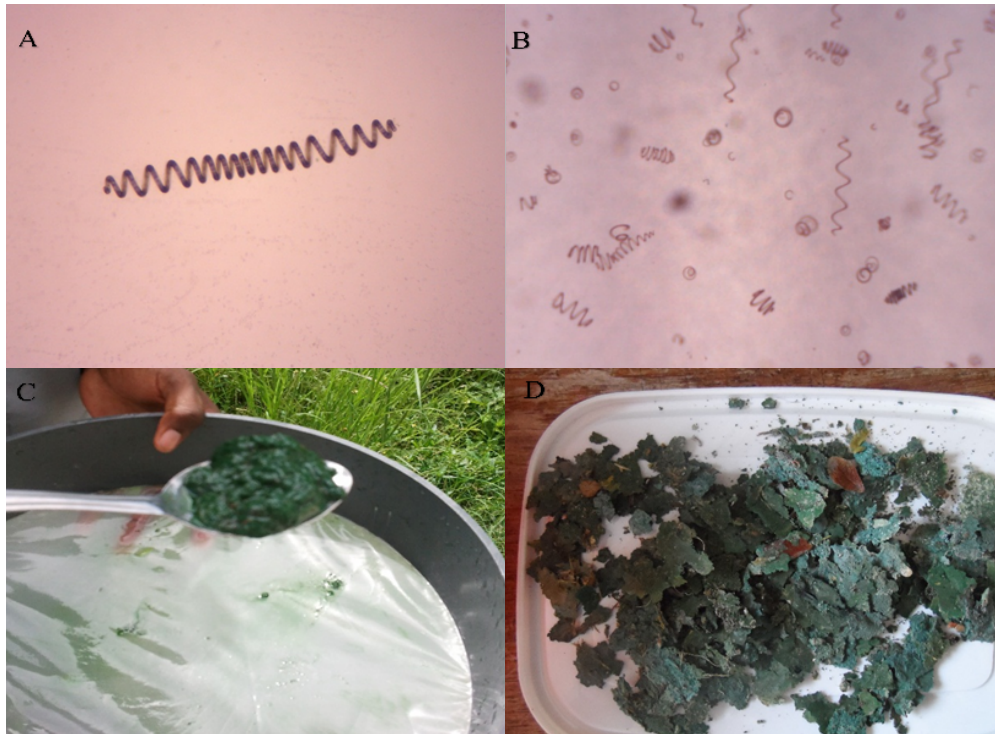


Figure 2. Morphology of spirulina, *A. fusiformis*. (A) x 10 magnification. (B) x 4 magnification. (C) fresh. (D) dried mass.

the fresh samples in a muffle furnace at 550 °C for 3 hrs. Crude protein (N X625) and crude fat was determined using micro kjeldahl and soxhlet extraction methods, respectively. Crude fiber was determined using an ANKOM fiber analyzer.

Data analysis

Results were recorded as means with standard error (\pm SE) calculated from the residual mean square. One-way analysis of variance (ANOVA) was used to determine differences between treatment means at $p < 0.05$. Post-hoc analysis was carried out where significant differences were detected between treatments by using Tukey's Honest Significant Difference (THSD) Test. Analysis was performed using Statistica software (Stat soft.) version 7 (Nunes *et al.*, 2015). Homogeneity of variance was checked using Levene's test.

Results

Spirulina identification

Cultured spirulina was identified based on microscopic characteristics and guidelines after Ciferri (1983), and Komarek and Lund (1990). The isolated spirulina were solitary with multicellular cylindrical trichomes. Most filaments were helical consisting of shorter than broader cells with clear and visible transverse cross walls (Fig. 2A and B).

General observations from the fish experiment

Fish fry in all treatments consumed their assigned experimental diets with full acceptance. Cultured spirulina had a proximate composition of 57.25% CP, 94.63% DM, 7.42% EE, 9.14% fiber and 5.35 % ash content (Table 1). Ingredients and proximate composition of the experimental diets are shown in Table 2. Water

Table 3. Physical-chemical parameters of water during experimental period.

Parameter	Minimum	Maximum	Mean \pm SE
Temperature (°C)	24.5	29.3	27.1 \pm 0.13
pH	7.5	8.2	7.9 \pm 0.02
Unionized ammonia (mg/l)	0.007	0.13	0.02 \pm 0.01
Dissolved oxygen (mg/l)	5.61	8.71	6.79 \pm 0.21
Salinity (ppt)	33	35	33.4 \pm 0.1

Table 4. Growth performance, feed utilization and survival rate of Rufiji Tilapia, *O. urolepis urolepis* fry, fed on experimental diets (mean \pm SE, n=3).

Parameter	Experimental diets					
	S0	S5	S15	S25	S35	S100
IW (g)	0.57 \pm 0.0 ^a	0.58 \pm 0.01 ^a	0.58 \pm 0.02 ^a	0.58 \pm 0.01 ^a	0.56 \pm 0.01 ^a	0.57 \pm 0.01 ^a
FW (g)	7.25 \pm 0.01 ^a	8.48 \pm 0.01 ^b	6.65 \pm 0.0 ^{ac}	6.31 \pm 0.01 ^c	5.98 \pm 0.02 ^c	3.78 \pm 0.02 ^d
WG (g)	6.68 \pm 0.01 ^a	7.9 \pm 0.01 ^b	6.07 \pm 0.02 ^{ac}	5.73 \pm 0.09 ^c	5.42 \pm 0.02 ^c	3.203 \pm 0.02 ^d
ADG (g fish ⁻¹ day ⁻¹)	0.111 \pm 0.0 ^a	0.132 \pm 0.01 ^b	0.101 \pm 0.02 ^{ac}	0.1 \pm 0.02 ^c	0.09 \pm 0.03 ^c	0.053 \pm 0.02 ^d
SGR (%g day ⁻¹)	4.23 \pm 0.01 ^a	4.47 \pm 0.03 ^b	4.07 \pm 0.02 ^{ac}	3.99 \pm 0.02 ^c	3.94 \pm 0.03 ^c	3.14 \pm 0.03 ^d
FI (g feed fish ⁻¹)	14.95 \pm 0.01 ^a	16.47 \pm 0.02 ^b	14.04 \pm 0.02 ^{ac}	13.51 \pm 0.01 ^{cd}	12.84 \pm 0.01 ^d	9.1 \pm 0.003 ^e
FCR	2.24 \pm 0.0 ^a	2.08 \pm 0.0 ^b	2.31 \pm 0.0 ^{ac}	2.35 \pm 0.0 ^c	2.39 \pm 0.01 ^c	2.58 \pm 0.01 ^d
PER	1.28 \pm 0.0 ^a	1.37 \pm 0.0 ^b	1.24 \pm 0.0 ^{ac}	1.21 \pm 0.0 ^c	1.19 \pm 0.03 ^c	1.0 \pm 0.02 ^d
Survival rate (%)	100 \pm 0.0 ^a	100 \pm 0.0 ^a	100 \pm 0.0 ^a	100 \pm 0.0 ^a	100 \pm 0.0 ^a	100 \pm 0.00 ^a

Values in the same row having different superscripts are significantly different ($p < 0.05$). Where: IW stands for initial weight, FW for final weight, ADG for average daily gain, SGR for specific growth rate, FCR for feed conversion ratio and PER for protein efficiency ratio.

quality parameters ranged from 24.5-29.3 °C, 7.5 - 8.2, 0.007-0.013 mg/l, 5.61-8.71 mg/l and 33-35 ppt for temperature, pH, unionized ammonia, dissolved oxygen (DO) and salinity, respectively (Table 3).

Growth performance

Growth performance of Rufiji tilapia fed with different experimental diets are presented in Table 4 and Fig. 3. Replacement of fishmeal by spirulina at different levels had a significant effect ($p < 0.05$) on the fish growth (FW, AWG, ADG and SGR). Fish fed with the S5 diet showed significantly ($p < 0.05$) higher growth

than those fed the control diet, S0. The lowest fish growth was observed in fish fed with the S100 diet. There was an increasing trend in FW, AWG, ADG and SGR in the S5, S0, S15, S25, S35 and S100 treatments respectively. Fish fed with the S15 diet had comparable ($p > 0.05$) growth to those fed with the control diet, S0. Similarly, there was no statistical difference ($p > 0.05$) in growth performance between fish fed with S15, S25 and S35 diets, the three diets however performed significantly higher ($p < 0.05$) compared to S100. No fish mortality was observed during the whole experimental period.

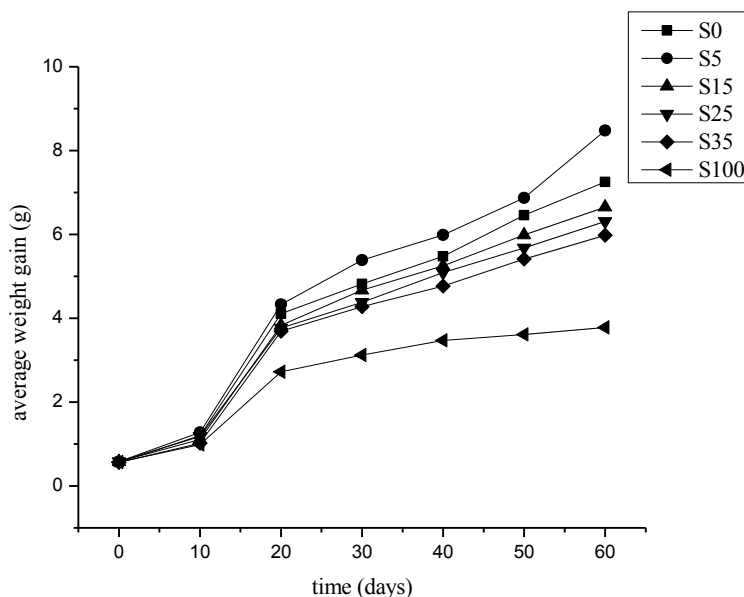


Figure 3. Changes in weight gain (g) of Rufiji tilapia, *O. urolepis urolepis* fry fed different levels of spirulina for 60 days.

Table 5. Chemical proximate composition (% on DM basis) of whole body of Rufiji tilapia, *O. urolepis urolepis*, before and after the experiment (mean \pm SE, n=2).

Treatment	Dry matter	Crude protein	Crude lipid	Ash
Initial	92.64 \pm 0.33 ^a	54.84 \pm 0.22 ^a	15.61 \pm 0.11 ^a	38.55 \pm 0.24 ^a
S0	92.72 \pm 0.16 ^a	50.71 \pm 0.44 ^a	14.65 \pm 0.18 ^a	20.96 \pm 0.62 ^b
S5	93.05 \pm 0.33 ^a	52.56 \pm 1.44 ^a	15.07 \pm 0.41 ^a	20.17 \pm 0.15 ^b
S15	95.18 \pm 0.43 ^a	51.34 \pm 1.25 ^a	14.72 \pm 0.00 ^a	21.14 \pm 1.38 ^b
S25	96.73 \pm 0.21 ^a	53.48 \pm 0.03 ^a	14.57 \pm 0.51 ^a	20.29 \pm 0.73 ^b
S35	95.70 \pm 0.3 ^a	45.50 \pm 4.4 ^a	12.8 \pm 1.23 ^b	19.7 \pm 0.290 ^b
S100	94.85 \pm 0.26 ^a	51.18 \pm 0.01 ^a	14.70 \pm 0.00 ^a	21.35 \pm 0.00 ^b

Values in the same column having different superscripts are significantly different ($p < 0.05$)

Feed utilization

A similar trend to growth performance was observed in the feed utilization parameters FCR and PER (Table 5). The lowest and highest FCR was exhibited in fish fed on S5 and S100 diets respectively, with the opposite trend observed in terms of PER. Both FCR and PER differed significantly ($p < 0.05$) among experimental diets. There was no significant difference ($p > 0.05$) in feed utilization between fish fed on S0 and S15, and also between S15, S25, S35 treatments. The lowest ($p < 0.05$) feed utilization values were observed in fish fed on the FM free diet (S100).

Fish biochemical composition

Results of carcass composition (DM, CP, CL and ash) of the whole-fish body at the start and the end of the experiment on fish fed fishmeal-spirulina substituted diet at different levels are presented in Table 5. There were no significant differences ($p > 0.05$) in terms of CP, DM and total ash content among experimental diets, except for CL which was significantly higher in the carcass of fish fed with the S35 diet.

Discussion

The potential of using single cell protein ingredients such as spirulina in fish feeds can be assessed on the basis of its protein content. Results from studies examining the nutritive value of spirulina vary greatly (Shah *et al.*, 2017). Madkour *et al.* (2012) reported protein content values (dry matter basis) of 37.79 to 47.1 %, and 52% when spirulina was cultured in reduced cost and synthetic media, respectively. Evaluating biochemical composition of spirulina using different culture media, Marrez *et al.* (2014) reported protein content ranging from 49.5 to 59 % dry matter. A 57% crude protein content of the spirulina used in this study was

within the ranges reported by Alvarenga (2011) and Belal *et al.* (2012), but differed from Yilmaz (2012), who reported crude protein ranges of up to 65%. The reasons for this variation could be attributed to the fact that spirulina in the present study was cultured under ambient conditions compared to the more controlled laboratory condition in the other studies.

Growth performance of Rufiji tilapia fed with spirulina supplementation at 5% inclusion were significantly higher than those fed with the control diet. Moreover, fish fed with 15% spirulina inclusion diet had comparable growth to those fed with the control diet. This indicates that spirulina inclusion of up to 15 % improved digestibility and feed intake, considering that spirulina is a single cell protein with no cell wall, and also contains high content of various nutrients such as vitamins and minerals (Habib *et al.*, 2008; Abdel-Tawwab and Ahmad, 2009). Similar findings were reported by Lu *et al.* (2002) when feeding raw spirulina to *Oreochromis niloticus* larvae at the onset of exogenous feeding. Additionally, *O. niloticus* was reported to display better weight gain and specific growth rates when fed with a 5 g spirulina kg⁻¹ diet (Abdel-Tawwab and Ahmad, 2009).

Apart from tilapia, spirulina has also been reported to improve growth in other fish species. Dernekbasi *et al.* (2010) for example, reported superior growth when spirulina was supplemented at 40% in practical diets for guppy, *Poecilia reticulata*. Rainbow trout (*Oncorhynchus mykiss*) fed with 7.5% spirulina in formulated feed showed significantly higher weight gain than with the fishmeal control diet (Teimouri *et al.*, 2013). Similarly, 5% fishmeal replacement with spirulina in diets of Parrotfish (*Oplegnathus fasciatus*) resulted in the highest

weight gain compared to other treatments (Kim *et al.*, 2014). On other hand, Ungsethaphand *et al.* (2010) did not find any significant differences in the growth performance of fish fed diets supplemented with spirulina at different levels to those fed the fishmeal control diet.

Improved fish growth has also been reported from studies that used other algal species to substitute fishmeal protein (Sarker, 2016). Tartiel *et al.* (2008), replacing fish meal with a combination of *Chlorella spp.* and *Scenedesmus spp.* in diets for *O. niloticus* at 10, 25, 50 and 70 % replacement levels, found that growth performance, FCR and protein productive were significantly higher in fish feed diets containing 50% algae. Similarly, Walker and Berlinsky (2011) reported improved feed utilization and growth in juvenile Atlantic cod (*Gadus morhua*) fed with 15% fishmeal substitution with a combination of dried *Nannochloropsis sp.* and *Isochrysis sp.* Vizcaíno *et al.* (2016) also concluded that *Tetraselmis suecica* and *Tisochrysis lutea* could replace up to 15% fishmeal in diets of gilthead sea bream fry.

In the present study, both FCR and PER increased with spirulina supplementation up to 15% inclusion. This is in agreement with Watanabe *et al.* (1990) and Takeuchi *et al.* (2002) who also found that feed supplemented with spirulina powder improved the feed conversion ratio in striped jack, *Pseudocaranx dentex*. Also, Belal *et al.* (2002) reported better FCR and PER when a 5 g spirulina kg⁻¹ diet was fed to *O. niloticus*. However, higher spirulina inclusion may result in poor feed utilization. This was found to be the case in the present study where both FCR and PER were negatively affected with higher and complete fishmeal replacement with spirulina. Similar results were reported by El-Sayed (1994) and Sharma and Panta (2012) who found that substitution with spirulina beyond 30% negatively affected fish growth. Additionally, Takeuchi *et al.* (2002) found that juvenile tilapia fed solely on the alga showed lower PER than commercial diets. On the other hand, Ungsethaphand *et al.* (2010) noted that feed utilization of hybrid red tilapia was not affected by spirulina supplementation. These variations might be attributed to differences in the spirulina concentration, the form of spirulina (raw or dried), fish species and size, as well as rearing condition.

Proximate biochemical composition of any edible organism forms an important aspect in food nutrition. The nutritive composition of fish can be greatly influenced by the type of feed they consume (Edea *et al.*, 2018). In the present study, there were no significant

differences in body crude protein and dry matter between fish groups fed spirulina and control diets. This indicates that spirulina supplementation did not compromise protein synthesis in fish. Similarly, Olvera *et al.* (1998) found that crude body protein and dry matter in *O. mosambicus* were not clearly affected by spirulina inclusion. Also, red tilapia fed with spirulina diets at 0, 5, and 10% levels did not show significant differences in carcass proximate composition compared to those fed on the control diet (Ungsethaphand *et al.*, 2010). On the other hand, lower lipid content in fish body fed 35% spirulina in the present study could be linked to a decrease in fish appetite, resulting in lower feed intake and nutrient utilization, which could lead to decreased lipid synthesis and deposition.

Tilapia is able to exist in a wide range of water temperature (from 12 to 42 °C), but the temperature to which they will adapt on sudden transfer depends on the temperature to which they were acclimated (Avella *et al.*, 1992). According to Popma and Lovshin (1995), the lethal lower temperature for most tilapia species is 10 - 11°C, while stress and diseases outbreaks occur at 37 - 38°C. In the present study, water temperature, dissolved oxygen and ammonia levels were within acceptable ranges reported by Makori *et al.* (2017), while pH (7.5 - 8.2) was within ranges reported by Ross (2000) who found pH tolerance in tilapia to be as low as 3 and as high as 11, and with the optimum range being from 7 - 9. On culturing Rufiji tilapia in salt water, Nehemia *et al.* (2014a) concluded that up to 35 ppt salinity was acceptable, which is within the range (33 - 35 ppt) in which fish were cultured in the present study.

Conclusions

The present study evaluated the potential use of a locally sourced spirulina, *A. fusiformis*, as a protein source in practical diets for Rufiji tilapia mariculture. The choice of this microalga was based on its relatively high protein content, local availability in soda lakes, and favorable climatic condition for its culture in Tanzania. The conclusion drawn from the present study is that *A. fusiformis* from Momela Lake can replace up to 15 % of the fishmeal protein in feeds for *O. urolepis urolepis* without adverse effects, and the best fish growth was observed at the 5% level.

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