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Growth and feed utilization in *Clarias gariepinus* fingerlings fed on *Acacia auriculiformis* leaf supplemented diets

AFE Oluwafunmilola Eunice and OMOSOWONE Olajumoke Olamiposi*

Department of Fisheries and Aquaculture Technology, School of Agriculture and Agricultural Technology, The Federal University of Technology, Akure, Ondo State, Nigeria.

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Effect of 'Earleaf' plant (*Acacia auriculiformis*) as feed additive was examined on *Clarias gariepinus* fingerlings (4.2±0.5 g) over a period 56 days against farm-made feed which served as the control. *A. auriculiformis* leaves were collected, air-dried (under shade), powdered and added at varying inclusion levels (0, 0.5, 1.0, 1.5 and 2.0%) to basal diets containing 40% crude protein. Fish were randomly distributed into tanks at 10 fish tank^{-1} with each treatment in triplicate. *C. gariepinus* were fed at 5% body weight between 8:00 – 9:00 and 16:00 – 17:00 h for 56 days. At the expiration of the feeding trials, results showed significant differences ($p<0.05$) in growth performance and nutrient utilization indices measured. The highest weight gain (WG), feed intake (FI), specific growth rate (SGR), protein efficiency ratio (PER) and best feed conversion ratio (FCR) were recorded in *C. gariepinus* fed *A. auriculiformis* at 1.5% supplementation level. Fish fed *A. aurculiformis* supplemented diets had higher WG (3.60 and 5.73 g) than those fed the control diet. Incorporating *A. auriculiformis* into fish diets did not have any adverse effect on the physiochemical water parameters measured as they were within the recommended ranges for raising warm water fish species. Results showed that diet supplementation with *A. auriculiformis* at 1.5 and 2.0% gave best growth performance and higher protein conversion, hence, the recommendation for diet supplementation at 1.5 or 2.0% inclusion levels.

Key words: Catfish, growth, *Acacia auriculiformis*, utilization.

INTRODUCTION

There is the rising need to improve food security, among economically challenged developing countries. Aquaculture has become an increasingly important option for improving animal protein intake from 40 to about 60% so as to fulfil a core deliverable sustainable development goal. This can only be achieved using high-quality feeds rich in protein and other essential nutrients to improve growth while still maintaining the animal's health (Soltan and El-Laithy, 2008). The use of natural products as cheaper sources of growth promoters is making waves in the aquaculture industry as opposed to the expensive synthetic antibiotics and other growth promoters.

*Corresponding author. E-mail: ooomosowone@futa.edu.ng. Tel: +2348034339189.

The use of medicinal plants (ginger cloves, funnel seeds, fenugreek seeds, garlic bulbs, black seeds, peppermint leaves, etc) as natural growth promoters have been proven to significantly improve weight gain, survival and feed conversion rates in fish by about 50% (El-Dakar, 2004; Shalaby, 2004).

The African catfish *Clarias gariepinus* is a major cultivated fish of high commercial value in Nigeria with a production of 257, 368 tons in 2014 using intensive culture systems (FAO, 2016). This is because of its ability to consume supplementary feeds (agricultural by-products), good conversion of feed to flesh (can utilize 2 g of feed to gain 1 g of flesh), resistance to disease, ability to reproduce in captivity, fast growth rate and tolerance to a wide range of environmental conditions such as temperature and dissolved oxygen. The economic importance of this species has increased tremendously as a result of its extensive use in aquaculture (De Graff and Janssen, 1996). In Nigeria, revenue generation from *C. gariepinus* was 766, 024 USD in 2014 compared to 519, 060 USD in 2010 (FAO, 2016).

Acacia auriculiformis, commonly called "Earleaf Acacia or Australian wattle" belongs to the family Fabaceae. The tree grows tall and thrives in poor soils and is well adapted to areas with extended dry seasons (Norman, 2000). Various extracts (leaf, stem, bark and root) of this plant have shown antioxidant benefit on living organisms generally (Singh et al., 2007). It is reported to be significantly effective against helminthic diseases, filariasis and microbial diseases (Mandal et al., 2005; Ghosh et al*.,* 1993). The root extract is known to be useful in the treatment of various aches, pains and sore eyes in man. The Aborigines of Australia were known to have used the bark extract to treat rheumatism effectively (Girijashankar, 2011).

The major objective of this study was to evaluate the effect of the natural herb; *A. auriculiformis* as a growth promoter in *C. gariepinus* fingerlings, thus, deviating from the conventional use of synthetic additives. The conventional additives commonly used in this part of the globe are the antibiotic growth promoters which are not readily available to the local fish farmers.

MATERIALS AND METHODS

Preparation of test ingredient and experimental diets

Fresh leaves of *A. auriculiformis* were collected from the matured tree within the main campus and authenticated by the Department of Crop Soil and Pest Technology, The Federal University of Technology, Akure, Ondo State, South-Western Nigeria. Leaves (fresh) of about 1.5 g were air dried at room temperature or under shade (25 – 27°C) and milled to fine powder using Maulinex electric blender. All dietary ingredients; fish meal, soybean meal, groundnut cake, yellow maize, rice bran, vegetable oil, bone meal, etc were purchased in a local market (Oja-Oba). Ingredients were then weighed on a top load balance (METLER TOLEDO, PB 8001 LONDON) and milled to powder (less than 20 µm). *A. auriculiformis* leaf powder (AALP) was carefully added at varying inclusion (0.5, 1.0, 1.5 or 2.0) % levels to five iso-nitrogenous (40% crude protein) basal diets (Table 1). All ingredients were thoroughly mixed using a kitchen mixer to obtain a homogenous meal mixture. The mixture was then steam pelleted through a 0.6 mm diameter die opening using Hobart pelletizer (A-2007 MODEL, UK).

Pellets were oven-dried at 50°C for 48 h, cooled to room temperature before been bagged in airtight containers and refrigerated prior to use. Experimental diets were designated as AALP1 (control), AALP2 (0.5% *A. auriculiformis*), AALP3 (1.0% *A. auriculiformis)*, AALP4 (1.5% *A. auriculiformis)* and AALP5 (2.0% *A. auriculiformis)* based on the test ingredient inclusion levels. Proximate compositions of experimental diets were assayed according to Association of Official Analytical Chemists, AOAC (2010) methods.

Experimental design

The experimental site was Teaching and Research Farm of the Department of Fisheries and Aquaculture Technology, The Federal University of Technology, Akure, Ondo State, Nigeria. All analyses took place at the Central Research Laboratory of the University. The experimental design was completely randomized with a single source of variation; the quantity of test ingredient (*A. auriculiformis* leaf powder) in each diet.

C. gariepinus fingerlings (4.20 ± 0.15) g were procured from KEAFI Farms in Akure, Ondo State, Nigeria and transported in oxygenated bags to experimental site. Fish were acclimated to laboratory conditions for 14 days, during which they were fed with a commercial diet-Durante Feeds. *C. gariepinus* were randomly distributed at a density of 10 fish tank^{-1} into fifteen aerators-fitted rectangular glass tanks with each treatment in triplicate. Each experimental unit is a 60 L water capacity plastic tank filled to 40-L level.

Feeding trial

Experimental diets were fed to fish twice daily at 5% body weight in two equal portions between 8:00 - 9:00 h and 17:00 - 18:00 h for 56 days. Experimental tanks were cleaned weekly and culture water replaced with fresh water on alternate days.

Experimental analyses

Chemical compositions of dry *A. auriculiformis* leaf and pooled fish samples were analyzed using methods described by Association of Official Analytical Chemists, AOAC (2010). Some water quality parameters such as dissolved oxygen, ammonia, nitrate, pH and temperature were monitored daily using a multi parameter kit (digital YSI Meter; Model 57 and Knick Portamess; Model 912). Fish in each experimental unit were batch-weighed (10 fish/tank or batch) fortnightly using Citizen's sensitive weighing balance with maximum capacity of 5 kg. The following growth and feed utilization parameters were evaluated following appropriate procedure:

Weight Gain (MWG) (g) = $WG = \frac{W_f - W_i}{n}$

Specific Growth Rate (SGR %day^{-1}) = $SGR = \frac{ln\frac{W_f}{n} - \frac{W_i}{n}}{t} \times 100$

Where: W1 and W2 are natural log of initial and final weight and T is number of experimental days.

Protein fed = % protein × Total diet consumed

$$\text{Feed conversion ratio (FCR)} = \frac{\text{Total feed consumed}}{\text{Weight gained}}$$

$$\text{Feed intake (FI) (g)} = \frac{5\%\ \text{Body weight} \times \text{Experimental period}}{\text{Number of fish stocked}}$$

$$\text{Protein efficiency ratio (PER)} = \frac{\text{Weight gain}}{\text{Protein intake}}$$

Statistical analysis

The effects of *A. auriculiformis* leaf powder as an additive on growth, nutrient utilization and body composition of *C. gariepinus* were statistically analyzed using one-way analysis (ANOVA). Data was first tested for homogeneity of variance and suitably transformed where significant differences were observed. Values generated were presented as mean (± Standard error). Significant differences ($p<0.05$) among means were compared and separated using Turkey's multiple range test (Zar, 1996) where applicable.

RESULTS

Chemical compositions of experimental diets

The result of proximate analysis of experimental diets is presented in Table 1. Table 2 shows chemical composition of *A. auriculiformis* leaf meal having a high concentration of nitrogen free extract. Crude protein, NFE and gross energy contents of experimental diets were not significantly different ($p>0.05$). Crude protein content in experimental diets were iso-nitrogenous and not significantly different ($p>0.05$) indicating that no prejudice was introduced in the process of compounding experimental diets (Table 1). Results showed that there was a linear increase of crude lipid in feed (from 5.45% in AALP1 - control to 7.55% in treatment AALP5) as the quantity of test ingredient (*A. auriculiformis*) increased. Also, a similar but opposite trend was observed in ash and crude fibre contents as the quantity of *A. auriculiformis* increased from 4.69 to 3.39%.

Water quality parameters

The values of water quality parameters measured are shown in Table 5. There were no significant differences ($p>0.05$) in all the parameters measured among treatments. Dissolved oxygen ranged from 5.76 to 6.35 mg l^{-l}, temperature from 26.40 to 27.80°C, while pH ranged from 8.20 to 8.34. Values of nitrate concentration were between 0.02 and 0.09 mgL^{-l} while ammonia concentration was between 0.001 and 0.002 mg L^{-l}.

Growth performance and nutrient utilization

At the expiration of the experiment, growth evaluation indices were used to assess response of *C. gariepinus* to *A. auriculiformis* leaf powder supplemented diets and the result is presented in Table 3. There were no significant differences ($p>0.05$) in the initial stocking weight of *C. gariepinus* in all treatments. Also, no mortality was recorded during the experimental period and fishes accepted diets administered as they were observed to be actively fed. Results showed that *C. gariepinus* fed diet AALP4 (1.5% *A. auriculiformis*) had the highest mean weight gain (13.63 g) while the least was recorded fish fed the control diet. The increase in weight did not follow a linear trend, that is, it was not dependent on the quantity of the test ingredient. Feed intake increased with inclusion levels of test ingredient with the highest value (50.32 g) recorded in fish fed diet AALP4 (1.5 g *A. auriculiformis*). Similarly, there were significant differences ($p<0.05$) and improvement in feed conversion ratio (FC), specific growth rate (SGR) and protein efficiency ratio (PER) of *C. gariepinus* fed *A. auriculiformis* supplemented diets in comparison with the control. The best FCR (3.70), SGR (1.09) and PER (0.34) were recorded in fish fed diet AALP4.

Chemical body composition of *C. gariepinus*

Chemical composition (dry matter) of *C. gariepinus* fed different inclusion levels of *A. auriculiformis* supplemented diets is presented in Table 4. Moisture and crude lipid contents in fish were not significantly different ($p>0.05$) in fish in all the treatments. Crude protein contents were significantly different ($p<0.05$) in *C. gariepinus* fed test diets when compared with the control, but not statistically different within the test diets treatment groups, although fish fed diet AALP5 had the highest body protein retention. A linear increase in body ash content of *C. gariepinus* was observed with the highest recorded in fish fed diet AALP5 while a downward (decrease) trend was observed in NFE content with the highest recorded in fish fed diet AALP1 (control).

DISCUSSION

In recent years, focus has shifted from the use of synthetic substances as additives in fish diets to use of plants and its by-products for health reasons. Several medicinal plants have been used in nutritional studies in the field of aquaculture such as marjoram (*Majorana syriaca*), licorice (*Glycyrrhiza glabra*) roots, black (*Nigella sativa*) seeds, peppermint (*Mentha piperita*), caraway (*Carum carvi*) seed, fennel (*Foeniculum vulgare*) seed, fenugreek (*Trigonella fornum-graceum*) seeds and ginger (*Zingiber officinale*) cloves (El-Dakar et al., 2008; Khalil et al., 2009; Al-Absawy, 2010; Abdelhamid, 2010). There

Table 1. Gross and proximate compositions (% DM basis) of experimental diets.

Ingredients	AALP1 (Control)	AALP2	AALP3	AALP4	AALP5
Fish meal	23.00	23.00	23.00	23.00	23.00
Soybean Meal	26.00	26.00	26.00	26.00	26.00
Groundnut Cake	28.00	28.00	28.00	28.00	28.00
Yellow Maize	11.00	11.00	11.00	11.00	11.00
Vegetable Oil	4.00	4.00	4.00	4.00	4.00
Rice Bran	2.00	2.00	2.00	2.00	2.00
Bone meal	2.00	2.00	2.00	2.00	2.00
Vit. / Min. Premix*	2.00	2.00	2.00	2.00	2.00
Binder	2.00	1.50	1.00	0.50	0.00
Acacia auriculiformis leaf Powder (AALP)	0.00	0.50	1.00	1.50	2.00
Proximate composition (dry matter) of experimental diets (%)					
Crude protein	40.01±0.07^{a}	40.03±0.06^{a}	40.06±0.09^{a}	40.08±0.03^{a}	40.09±0.05^{a}
Ether extract	5.45±0.42^{a}	6.23±0.39^{b}	6.42±0.45^{b}	6.95±0.58^{b}	7.55±0.67^{c}
Ash	16.96±0.42^{c}	15.94±0.26^{b}	15.48±0.12^{b}	14.56±0.45^{a}	14.79±0.30^{a}
Crude fibre	4.61±0.77^{b}	4.23±0.04^{b}	3.91±0.29^{ab}	3.51±0.77^{a}	3.39±0.04^{a}
**NFE	26.46±0.02^{a}	26.11±0.41^{a}	25.33±0.30^{a}	25.39±0.51^{a}	25.43±0.89^{a}
***Gross energy (kcal 100 g $^{-1}$ DM)	400.55±5.04^{a}	406.17±6.24^{a}	403.69±6.12^{a}	408.30±6.31^{a}	413.93±6.75^{a}

Vitamin premix*: An Animal Care® Optimix Aqua product for catfish, containing the following per 5 kg of premix: A = 20 000 000 IU, D3 =2 000 000 IU, E = 200 000 mg, K3 = 10 000 mg, B2 =12 000 mg, B12 = 9 mg, B1 = 6 000 mg, B6 = 11 000 mg, C = 50 000 mg, Folic acid = 2 000mg, Niacin =80 000 mg, Calpan = 25 000 mg, Biotin = 100 mg, x Zinc =30 000 mg, Copper = 5 000 mg, Iron = 30 000 mg, Manganese = 50 000 mg,Iodine = 1 000 mg, Selenium = 100 mg, Antioxidant = 125 000 mg, **NFE: Nitrogen free extract, ***Gross energy: 5.65, 9.45, 4.0 and 4.0 kcal/g of protein, ether extract, crude fibre and NFE (Jobling, 1983). DM = dry matter.

Table 2. Proximate composition (dry matter) of *Acacia auriculiformis*.

Parameter	Composition (%)
Crude protein	14.69±0.77
Ether extract	2.08±0.17
Crude Fibre	33.42±0.54
Ash	7.01±0.14
Nitrogen free extract	35.48±0.23

has not been any report on *A. auriculiformis* as a growth enhancing agent in *C. gariepinus* fingerlings, hence, its use in this study. *A. auriculiformis* is a leguminous plant in nature and previous work on the seeds had been reported to contain 31.8% crude protein, 7.5% crude fibre and 49.2% NFE (Sathya and Siddhuraju, 2013). Its high NFE content was further confirmed in this study when the leaf was examined. Crude protein of 40% in experimental diets was within recommended range used by other researchers in compounding diets for *C. gariepinus*. Similar studies using medicinal plants in fish diets have crude protein ranging from 35-45% (Harikrishnan et al., 2011).

Some medicinal plants have been reported to have growth stimulating effect when consumed by animals and this was also observed in this study as *C. gariepinus* fed *A. auriculiformis* supplemented diets had better weight gain than the control. Improved growth performance and better nutrient utilization was recorded in Nile tilapia fed fenugreek sprouts meal (Zaki et al., 2012). Goodarzi and Nanekarani (2014) reported an improvement in growth (50%) performance of broilers fed *Mentha pelegium* supplemented over the control. Similarly, Talpur (2014) who incorporated peppermint (*M. piperita*) as a feed additive in the diet of Asian seabass (*Lates calcarifer*) recorded improved growth. Furthermore, this result also agrees with that of Turan and Akyurt (2005) used red clover (*Trifolium pretense)* extract as a growth-enhancing agent in catfish, *C. gariepinus.* Mahdavi et al. (2013) also reported better growth indices when *Aloe vera* was

Table 3. Growth and nutrient utilization of *C. gariepinus* fed various levels of *Acacia auriculiformis* supplemented diets.

Parameter	AALP1 (Control)	AALP2	AALP3	AALP4	AALP5
Initial weight (g)	4.28±0.05^{a}	4.65±0.07^{a}	4.30±0.03^{a}	4.34±0.09^{a}	4.40±0.09^{a}
Final weight (g)	12.18±1.43^{b}	17.64±1.9^{a}	15.80±1.33^{ab}	17.97±1.82^{a}	16.50±1.27^{ab}
Weight gain (g)	7.90±1.50^{c}	12.99±1.92^{ab}	11.50±1.3^{b}	13.63±1.96^{a}	12.10±1.33^{b}
Feed intake (g)	34.10±4.72^{c}	49.40±7.22^{a}	44.24±4.74^{b}	50.32±5.73^{a}	46.20±6.33^{b}
FCR	4.31±0.35^{a}	3.80±0.70^{ab}	3.85±0.20^{ab}	3.70±0.56^{b}	3.82±0.45^{ab}
SGR (% day $^{-1}$)	0.82±0.04^{b}	1.04±0.01^{a}	1.02±0.06^{a}	1.09±0.04^{a}	1.04±0.06^{a}
PER	0.20±0.00^{b}	0.32±0.03^{ab}	0.29±0.01^{ab}	0.34±0.05^{a}	0.30±0.03^{ab}

Means in a given column with the same superscript letter were not significantly different at p<0.05. SGR=specific growth rate, PER = protein efficiency ratio, FCR = feed conversion ratio.

Table 4. Chemical composition of whole body of *C. gariepinus* fingerlings fed experimental diets (dry weight basis).

Parameter (%)	AALP1 (Control)	AALP2	AALP3	AALP4	AALP5
Moisture	6.48±0.13^{a}	6.24±0.35^{a}	6.41±0.015^{a}	6.49±0.00^{a}	6.27±0.03^{a}
Ash	16.90±0.3^{a}	16.93±0.17^{a}	16.94±0.415^{a}	17.82±0.10^{ab}	18.32±0.04^{b}
Lipid	9.18±0.53^{a}	9.22±0.57^{a}	9.47±0.40^{a}	9.63±0.450^{a}	9.86±0.28^{a}
Protein	62.94±0.02^{a}	63.40±0.90^{b}	63.46±1.21^{b}	63.65±0.48^{b}	63.74±0.15^{b}
NFE	4.50±0.04^{b}	4.21±0.09^{b}	3.72±0.10^{ab}	2.41±0.08^{a}	1.81±0.09^{a}

Means in a given column with the same superscript letter were not significantly different at p<0.05.

Table 5. Physio-chemical parameters of water in experimental units.

Parameter	AALP1 (Control)	AALP2	AALP3	AALP4	AALP5
Dissolved oxygen (mg L $^{-1}$)	6.35±0.21^{b}	6.18±0.17^{b}	6.02±0.38^{a}	5.83±0.09^{a}	5.76±0.11^{a}
Temperature (°C)	26.40±0.43^{a}	27.60±0.32^{a}	27.20±0.15^{a}	27.70±0.23^{a}	27.80±0.15^{a}
pH	8.34±0.23^{a}	8.31±0.00^{a}	8.27±0.01^{a}	8.24±0.08^{a}	8.20±0.02^{a}
Nitrate (mg L $^{-1}$)	0.02±0.01^{a}	0.07±0.01^{a}	0.09±0.01^{a}	0.03±0.01^{a}	0.05±0.01^{a}
Ammonia (mg L $^{-1}$)	0.001±0.00^{a}	0.002±0.00^{a}	0.001±0.00^{a}	0.001±0.00^{a}	0.002±0.00^{a}

Means with the same superscript in the same row are not significantly different (p>0.05).

included in the diets of common Carp (*Cyprinus carpio*). Soosean et al. (2010) also corroborated the fact that *C. gariepinus* fed on medicinal plant extract supplemented diets exhibited faster growth than those fed with the control diet.

In this study, dietary supplementation of *C. gariepinus* diets with *A. auriciliformis* improved SGR in all treatments when compared with the control. The highest SGR was observed in fish fed diet AALP4 (1.09% day^{-1}). Previous study using fresh or dried garlic (*Allium sativum*) as a natural feed supplement on growth performance and nutrients utilization of the Nile Tilapia (*Oreochromis niloticus*) showed that fish fed with garlic supplemented diets showed high SGR over the control (Omosowone, 2011; Kumar et al., 2013). Similarly, Dada and Sonibare (2015) recorded improved weight gain (40%) using Siam weed (*Chromolaena odorata*) in the diet of *C. gariepinus*. In this experiment, *C. gariepinus* fed diet AALP4 had better FCR and PER compared with the other treatments and control. This finding is contrary to that of Pakravan et al. (2012) who reported no significant difference in FCR of common carp, *C. carpio* fed dietary willow herb, *Epilobium hirsutum*. However, other researchers (Yu et al., 2008; Dada and Ikuerowo, 2009) have reported improved FCR in *C. gariepinus* using medicinal plants.

Moisture and lipid contents of whole body composition of *C. gariepinus* fed all diets were not significantly different (p>0.05) showing that inclusion of *A. auriculiformis* leaf powder had no negative effect. This result is in agreement with those reported by Talpur (2014) and Pakravan et al. (2012) that inclusion of medicinal plant; fenugreek seed did not affect lipid and

moisture contents in Nile tilapia. Body protein composition of *C. gariepinus* fed *A. auriculiformis* supplemented was significantly different from control. It was observed that although protein retention in fish body was not different statistically, yet, *C. gariepinus* fed diet AALP5 had higher body protein content. Higher ash content was observed in the body of *C. gariepinus* fed diet AALP5 substantiating the assertion that *A. auriculiformis* is high in minerals (Girijashankar 2011). Generally, the result of proximate composition of *C. gariepinus* in this study agrees with reports of other researchers such as Pakravan et al. (2012), and Dada and Abiodun (2014) that used other medicinal plants.

The physiochemical parameters of water quality in experimental units measured during the experiment were not significantly different among all treatments except dissolved oxygen. There was a decrease in dissolved oxygen as the quantity of *A. auriculiformis* leaf powder increased in fish diets. Results of the dissolved oxygen though statistically different, were still the range values for culture of tropical fishes such as *C. gariepinus*. Similar results were reported by Tolan and Sherif (2007), Musa et al. (2013) and Samkelisiwe and Ngonidzashe (2014) using different medicinal plants and additives in the laboratory culture of tropical fishes. Water temperature is an important parameter in fish culture as it could affect protein intake either positively or negatively. Water temperature and other parameters; pH, nitrate and ammonia were within tolerance levels for aquaculture species. Furthermore, Lamai and Kolo (2013), Osuigwe et al. (2005), and Hussain (2004) also gave varying levels similar to those obtained in this project in which farm raised aquaculture species could thrive well.

Conclusion

This study has shown that incorporating *A. auriculiformis* leaf powder meal in the diet of *C. gariepinus* at varying inclusion levels led to higher growth and better nutrient utilization than the control. It can be concluded that under the experimental conditions used, inclusion of *A. auriculiformis* leaf powder in diet of African catfish fingerlings at 1.5 – 2.0% is suitable as a feed additive to produce visible changes in its growth and body composition.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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