E-ISSN: 2347-5129
P-ISSN: 2394-0506
(ICV-Poland) Impact Value: 5.62
(GIF) Impact Factor: 0.549
IJFAS 2020; 8(1): 16-19

www.fisheriesjournal.com
Received: 10-11-2019
Accepted: 12-12-2019

Jazer Anne M Tattao
Department of Aquatic Resources, Ecology and Management, College of Fisheries, Central Luzon State University, Science City of Muñoz, Nueva Ecija, Philippines

Ravelina R Velasco
Department of Aquatic Resources, Ecology and Management, College of Fisheries, Central Luzon State University, Science City of Muñoz, Nueva Ecija, Philippines

Jemuel S Doctolero
Department of Aquaculture, College of Fisheries, Central Luzon State University, Science City of Muñoz, Nueva Ecija, Philippines

Corresponding Author:
Jazer Anne M Tattao
Department of Aquatic Resources, Ecology and Management, College of Fisheries, Central Luzon State University, Science City of Muñoz, Nueva Ecija, Philippines

Potential of ashitaba (*Angelica keiskei* Koidzumi) leaves as a feed additive on the diet of Nile tilapia fry (*Oreochromis niloticus* L.)

Jazer Anne M Tattao, Ravelina R Velasco and Jemuel S Doctolero

Abstract
The interest of using natural materials such as medicinal plants has increased its acceptability as feed additives to fish for the attainment of efficient feed utilization. Ashitaba (*Angelica keiskei)* is considered as medicinal plants that contains high nutritive value and diverse bioactive components. Due to the increasing costs and short supply of fish-based feed ingredients efforts have been directed to search for an alternative. The study was conducted to determine the effectiveness of Ashitaba leaves as feed additive on the diet of Nile tilapia. Four experimental diets were prepared, composed of different levels of Ashitaba such as 0% (commercial feeds), 5%, 10% and 15%. Results of the study showed that the control group attained comparable growth and survival with that of fish whose feeds were incorporated with Ashitaba leaves. The study concluded that addition of up to 10% Ashitaba leaves could be used for the culture of Nile tilapia without negative effect on its growth and survival. Further, utilization of Ashitaba leaves as feed additive can potentially improve the growth performance and survival rate of Nile tilapia fry.

Keywords: *Angelica keiskei* Koidzumi, Nile tilapia, *Oreochromis niloticus* L

Introduction
The rapid growth of tilapia culture has stimulated the expansion of tilapia feed production and a search for novel protein sources to replace fish meal and vegetable or plant sources are promising alternatives which are naturally abundant and high in protein content (Garduño-Lugo & Olvera-Novoa, 2008) [1]. World Health Organization encourages the use of medicinal herbs and plants to substitute or minimize the use of chemicals through the global trend to go back to the nature (Palhares *et al.,* 2015) [2]. Attempts to use natural materials such as medicinal plants could be widely accepted as feed additives to enhance feed utilization and aquaculture production (Dada, 2015) [3].

Ashitaba has long been considered a folk remedy, used as a diuretic, laxative, analeptic and galactagogue in the Izu islands for a long time (Ohnogi *et al.,* 2012) [4]. It is a popular botanical medicine containing diverse bioactive components including prenylated chalcones, linear and angular coumarins, and flavanones (Caesar & Cech, 2016) [5]. The plant is nutritious as it is high in vitamins and mineral content (Japan Bio Science Laboratories Co., Ltd., 2009) [6] and have unique and rare class of flavonoids called chalcones which an aromatic ketone that forms a central core for a variety of important biological compound that have different properties like antibacterial, antifungal, anti-inflammatory and anti-tumor (Chavan *et al.,* 2016) [7].

Attaining truly sustainable growth in the aquaculture industry will depend on a progressive decrease in the use of marine protein and lipids in feeds for farmed fish (Francis *et al.,* 2001 [8]; Francesco *et al.,* 2004 [9]. Fish feed has created the need to search for an alternative due to the increasing costs and short supply (Bimbo & Crowther, 1992) [10]. The fish feeds needs to be fortified with feed additives that are edible substances to enhance the feed quality which in turn enhances growth performance and reduces mortality in fish (Dada, 2015) [3]. Some researchers found that the use of plant based protein in fish feeds improved growth performance (Mzengereza & Kang'ombe, 2016) [11].

Information about Ashitaba plant as added feed ingredient or feed additive for Nile tilapia is not yet previously studied and in order to determine its potential, the study aimed to assess the

Ashitaba leaves as feed additive on the diet of Nile tilapia by evaluating its effect on the growth and survival as well as by determining its nutritive value.

Materials and Methods

Experimental Fish and Set Up

Nile tilapia (*Oreochromis niloticus* L.) fry with a mean weight of 0.3 g were obtained from a private hatchery in Sapang Cauayan, Science City of Munoz, Nueva Ecija, Philippines. Twelve indoor aquaria 30x60x30 cm in size with 35 L capacity served as experimental unit and situated at the Wet Laboratory of the Freshwater Aquaculture Center - Central Luzon State University. Acclimation was done for seven days prior to stocking and each aquaria was stocked randomly with 30 Nile tilapia fry. Fish were fed three times a day at 9:00 a.m., 1:00 pm and 4:00 pm with a feeding rate of 10%. Maintenance of good water condition was done by supplying aeration and daily water exchange. Fish sampling was done every 15 days to monitor the growth and survival of the fish. The culture duration lasted for 45 days.

Experimental Treatments and Preparation of feed

The study had four treatments composed of different dietary levels of Ashitaba such as
0% (commercial feeds), 5%, 10% and 15% which were named as Treatment 1, Treatment 2,
Treatment 3 and Treatment 4, respectively. The powdered Ashitaba leaves were weighed using electronic weighing scale accordingly to its level of incorporation. The powdered leaves were mixed and homogenized thoroughly together with the commercial feeds using feed mixer

Proximate Analysis

Proximate analysis of the prepared feed was conducted at the Nutrition Laboratory of Freshwater Aquaculture Center, Central Luzon State University. Crude protein were determined following the Kjeldahl method (N x 6.25). Identification of Crude lipid content was done using Goldfisch method by extraction with petroleum ether. A drying oven was used to determine the moisture content set at 40 degree Celsius for six hours and while Ash content was measured by subjecting the sample in muffle furnace 600˚C for three hours.

Data Gathered and Statistical Analysis

The study determined the final weight, final length, specific growth rate, gain in weight and survival rate of the experimental fish. The data generated were analyzed using analysis of variance and comparison of means was done using Tukey's Range Test at significant level of (P<0.05). Statistical analysis was done using R-software.

Results and Discussion

Growth is measured in units of length and weight and feeding is one of the factors that promotes growth. Table 2 shows the growth performance of Nile tilapia fry in terms of final weight and length, specific growth rate, gain in weight and survival rate. Final weight revealed that all treatments were not significantly different with each other and thus attained comparable growth after 45 days of culture. Comparing the final weight of treatments with added Ashitaba leaves, the level of incorporation of 10% attained the highest result. In terms of final length, statistical analysis also revealed that all treatments were insignificantly different to each other however, diet with 10% added Ashitaba leaves also attained the highest result. Moreover, similar result was obtained in Specific Growth Rate. On the other hand, significance among treatments were obtained in terms of gain in weight where diet with 10% incorporated Ashitaba leaves was significantly higher compared to the control treatment.
Result on the survival rate further revealed that diet with 5% and 10% added Ashitaba leaves were significantly higher compared to the commercial feed (control treatment). However, result also revealed that as the level of Ashitaba leaves was increased up to 15%, reduction of survival was notable as diet with 15% Ashitaba leaves was significantly lower compared to the commercial feeds and the other level of incorporation (5% and 10%).

Table 1: Summary data on the growth of fish fed with and without powdered Ashitaba leaves.

	Treatments			
	1	**2**	**3**	**4**
Final Weight (g)	0.94±0.66^{a}	0.70±0.09^{a}	0.82±0.07^{a}	0.69±0.05^{a}
Final Length (cm)	2.94±2.54^{a}	3.48±0.96^{a}	3.51±4.37^{a}	2.85±5.92^{a}
Specific Growth Rate	1.41±1.46^{a}	0.90±0.19^{a}	1.15±0.15^{a}	0.86±0.11^{a}
Gain inweight (g)	0.25±0.01^{a}	0.40±0.09^{ab}	0.52±0.07^{bc}	0.39±0.05^{ac}
Survival %	83.33±0.02^{b}	96.67±0.01^{a}	93.33±0.02^{a}	73.33±0.03^{c}

Note: Means with different letters are significantly different at 5% level of significance

The growth curves of the experimental fish in various treatments are presented in Figure 1. Increasing growth of fish in various treatments as the culture period progresses was observed. In Figure 1, the difference in the increase of weight begins at day 15 and progressively increase up to 45th day. This is a clear indication of allometric growth pattern whereby the size of the fish is related to the total mass, as the length increases the weight also increases.

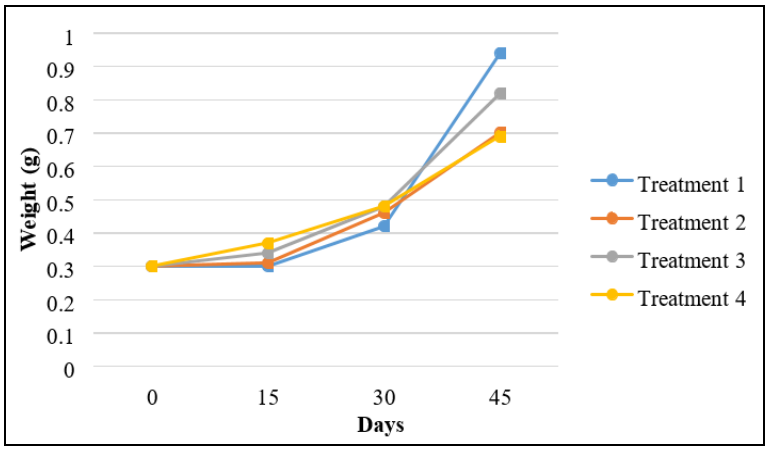

Fig 1: Growth pattern of Nile tilapia

The result on water quality parameters such as dissolved oxygen, temperature and pH were presented in Table 2. Result revealed that there were no significant differences (P□0.05) attained among all of the treatments. According to Boyd and Pillai (1985) [12], warm-water fish grow best at temperatures between 25°C and 32°C for dissolved oxygen (5.2-7.5mg/l) and waters with pH values of above 6.5 to 9 at daybreak are considered best for fish production. The level of the different water quality parameters evaluated were within the range of recommended level for Nile tilapia culture. Thus, it was notable in this study that the water quality parameters of each treatment were not affected by the addition of powdered Ashitaba leaves.

Table 2: Summary of water quality parameters.

Treatments	Dissolved Oxygen (ppm)	Temperature (°C)	pH
1	6.19 ± 0.05[a]	24.7 ± 0.01[a]	8.11 ± 0.01[a]
2	6.18 ± 0.07[a]	24.5 ± 0.32[a]	8.12 ± 0.01[a]
3	6.67 ± 0.9[a]	24.6 ± 0.06[a]	8.10 ± 0.02[a]
4	6.21 ± 0.11[a]	24.7 ± 0.04[a]	8.11 ± 0.01[a]

Note: Means with different letters are significantly different at 5% level of significance.

Proximate analysis (Table 3) revealed that crude protein level from all the experimental diets were within the range of 28.83% to 30.75%. The experimental diets incorporated with Ashitaba leaves had slightly higher analyzed crude protein level than the control treatment. On the other hand, crude fat was higher on the control than those of the feed with Ashitaba leaves. Similar result was also obtained in moisture content and ash content.

Table 3: Proximate Analysis of the feed added with Ashitaba

Treatments	Moisture Content	Dry Matter	Ash Content	Crude Protein	Crude Fat
1	13.00	87.00	12.00	30.00	5.00
2	6.89	93.10	11.90	30.26	1.78
3	6.82	93.18	11.75	30.75	4.80
4	6.60	93.39	11.80	28.83	4.66

The positive effects in fish growth performance can be attributed to the bioactive constituents of Ashitaba leaves such as flavonoids, coumarins, and chalcones (Kim, 2013) [13]. In the study of Panche (2016) [14] flavonoids is a group of natural substances with variable phenolic structures, are found in fruits, vegetables, grains, bark, roots, stems, flowers, tea and wine which is attributed to their anti-oxidative, anti-inflammatory, anti-mutagenic and anticarcinogenic properties coupled with their capacity to modulate key cellular enzyme function. Coumarins (1-benzopyran-2-one) are chemical compounds in the benzopyrone class of organic compounds found in many plants which possess a variety of biological properties, including antimicrobial, antiviral, anti-inflammatory, antidiabetic, antioxidant, and enzyme inhibitory activity as stated in the study of Poumale *et al.* (2013) [15]. On the study of Chavan *et al.* (2016) [7], they mentioned that Chalcone is an aromatic ketone that forms a central core for a variety of important biological compounds, which are collectively known as chalcones. This bioactive compounds possess different activities like antibacterial, antifungal, anti-inflammatory and anti-tumor depending on the substitution made on them. The study of Nagata *et al.* (2007) [16] showed that intake of Ashitaba leaves promotes excretion of feces and bile acids to rats which has significant effect on keeping the gastrointestinal system healthy. Moreover, the study of Olafisoye *et al.* (2017) [17] mentioned that Ashitaba is considered a functional medicinal plant that contains various invaluable vitamins and minerals. The following mentioned benefits of Ashitaba leaves contributed for the positive result obtained on the growth performance and survival of Nile tilapia fed with added levels of Ashitaba leaves.

Nutrient sources of plant origin are found to contain a wide variety of anti-nutritional substances that proved to be toxic to animals, affecting growth and health and exhibiting deleterious effects. Most of the anti-nutrients incorporated at levels as protein sources in fish feed do not lead to mortality but produce adverse physiological effects, decreased growth and health conditions (Hajra *et al.*, 2013) [18]. There are some processing methods to inactivate or reduce the presence of anti-nutrients in plant-based feed ingredient and one of the studied effective method is through exposure to higher temperature on the preparation of the diet (Penaflorida, 1995 [19]; Siddhuraju and Becker, 2005) [20]. Relating to the current study, preparation of the diets with incorporated Ashitaba leaves were subjected to high air temperature exposure (100 ^{0}C for 4 hr). This method addressed the concern about the presence of anti-nutritional factors in Ashitaba leaves as comparable growth performance was attained by Nile tilapia fry fed with Ashitaba leaves compared to commercial diet. It can be concluded that utilization of plant-based feed additive may limit the presence of anti-nutritional factors through exposing the plant materials during the preparation of the diet in an elevated air temperature.

Conclusion

The study concluded that utilization of Ashitaba leaves as feed additive can potentially improve the growth performance and survival rate of Nile tilapia fry. Further, incorporation of Ashitaba leaves with a level of up to 10% could be utilized for the culture of Nile tilapia without negative effect on its growth and survival. No previous studies were conducted on the utilization of Ashitaba leaves as feed additive to aquaculture fish thus, this effort gives new viewpoint on the usage of medicinal and functional plants like Ashitaba as an additive to fish feed to promote good growth and survival.

References

1. Garduño-Lugo M, AŁnge-Olvera M. Potential of the use of peanut (*Arachis hypogaea*) leaf meal as a partial replacement for fish meal in diets for Nile tilapia (*Oreochromis niloticus* L.). Aquaculture Research. 2008; 39:1299-1306.
2. Palhares RM, Gonçalves DM, dos Santos A, Figueiredo BB, Pereira CG, das Graças LBM *et al*. Medicinal plants recommended by the World Health Organization: DNA Barcode Identification Associated with Chemical Analyses Guarantees Their Quality. PLoS ONE. 2015; 10(5):e0127866.
3. Dada AA. Improvement of tilapia (*Oreochromis niloticus* Linnaeus, 1758) growth performance fed three commercial feed additives in diets. Journal Aquaculture Research Development. 2015; 6:4.
4. Ohnogi H, Hayami S, Kudo Y, Enoki T. Efficacy and safety of ashitaba (*Angelica keiskei*) on the patients and candidates with metabolic syndrome: a pilot study. JJCAM. 2012; 9:49-55.
5. Caesar LK, Cech NB. A review of the medicinal uses and pharmacology of Ashitaba. © Georg Thieme Verlag KG Stuttgart New York ISSN 00320943. Planta Medicine. 2016; 82:1236-1245.
6. Ashitaba Chalcone Powder. A Comprehensive Scientific Review. Japan Bio Science Laboratories Co., Ltd (JBSL) Internal report. https://www.dyna-nutrition.com/wpcontent/uploads/2016/04/Chalcones-WhitePaper-Apr172009.pdf. 17 April, 2009.
7. Chavan BB, Gadekar AS, Mehta PP, Vawhall PK, Kolsure AK, Chabukswar AR. Synthesis and medicinal significance of chalcones- A review. Asian Journal of Biomedical and Pharmaceutical Sciences. 2016; 6(56):01-07.
8. Francis G, Makkar PSH, Becker K. Antinutritional factors present in plant-derived alternate ¢sh feed ingredients and their ejects in ¢sh. Aquaculture. 2001; 199:197-227.
9. Francesco M, Parisi G, Medale F, Lupi P, Kaushik SJ, Poli BM. Effect of long-term feeding with a plant protein mixture based diet on growth and body/¢llet quality traits of large rainbow trout (*Oncorhynchus mykiss*). Aquaculture. 2004; 236:413-429.
10. Bimbo AP, Crowther JB. Fish meal and oil: Current uses. Journal of America Chemistry Society. 1992; 69:221-227.
11. Mzengereza K, Kang'ombe J. Effect of dietary salt (Sodium Chloride) supplementation on growth, survival and feed utilization of *Oreochromis shiranus* (Trewavas, 1941). Journal Aquaculture Research Development. 2016; 6:388.
12. Boyd C, Pillai V. Water Quality Management in Aquaculture, 1985.
13. Kim JH, Son YK, Kim GH, Hwang KH. Xanthoangelol and 4-Hydroxyderricin are the major active principles of the inhibitory activities against monoamine oxidases on *Angelica keiskei* K. Biomolecules & Therapeutics. 2013; 21(3):234-240.
14. Panche AN, Diwan AD, Chandra SR. Flavonoids: an overview. Journal of nutritional science. 2016; 5:e47.
15. Poumale HMP, Hamm R, Zang Y, Shiono Y, Kuete V. Medicinal Plant Research in Africa. First edition, Elsevier, London, 2013, 261-300.
16. Nagata J, Morino T, Saito M. Effects of dietary *Angelica keiskei* on serum and liver lipid profiles, and body fat accumulations in rats. Journal Nutrition Science Vitaminology. 2007; 53:133-137.
17. Olafisoye OB, Ojelade OD, Osibote OA. Trace elements and antioxidants in some medicinal plants. Research and Reviews in Bio Sciences. 2017; 11(3):111.
18. Hajra A, Mazumder A, Ganguly D, Mohanty B, Sharma A. Antinutritional factors in plant origin fish feed ingredients: the problems and probable remedies. In book: Advances in Fish Research. 2013; 11:193-202.
19. Penaflorida VD. Growth and survival of juvenile tiger shrimp fed diets where fish meal is partially replaced with papaya (*Carica papaya* L.) or camote (*Ipomea batatas* Lam.) leaf meal. Israel Journal Aquaculture Bamidgeh. 1995; 47:25-33.
20. Siddhuraju P, Becker P. Nutritional and antinutritional composition, *in vitro* amino acid availability, starch digestibility and predicted glycemic index of differentially processed mucuna beans (*Mucuna pruriens* var. utilis): An under-utilised legume. Food Chemistry. 2005; 91:275-286.