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Myrmecodia pendens Bulb Extract in the Lele Dumbo (*Clarias gariepinus*) Feed: Effects on the Growth Performance, Survival, and Blood Indices

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Abstract

This feeding experiment was performed to determine the effects of *Myrmecodia pendens* bulb extract (MBE) supplementation in fish feed on the growth, survival, and hemato-biochemical profile of *Clarias gariepinus*. A group of fish was fed with 0.25; 0.50; 1.0; 2.0% MBE and compared to the control group (without MBE) for 75 days of observation. At the end of the feeding trial, growth parameters, hematological profile such as red blood cells (RBC), white blood cells (WBC), Hemoglobin (Hb), Hematocrit (Htc), differential leukocyte, blood plasma biochemistry (glucose, total albumin, cholesterol, and triglyceride), the hepatosomatic (HSI) and intestinal somatic index (ISI) were measured. Survival of all fish was also counted every two weeks. Supplementation MBE above 0.25% resulted in significantly higher final biomass weight (FBW), body weight gain (BWG), daily weight gain (DWG), and average weekly gain (AWG). Meanwhile, the fish group fed dietary MBE above 1.0% had a significantly higher specific growth rate (SGR) (3.32±0.15) than other groups. Fish fed 1.0% of MBE also showed a better value of feed conversion ratio (FCR) (1.13±0.03), Hb, and HSI compared to other groups. Survival, neutrophil, monocyte, and ISI of all groups were not affected by any concentration of MBE supplementation. Dietary MBE above 0.5% enhanced RBC, WBC, Hematocrit, platelet (PLT), lymphocyte, blood plasma biochemistry such as glucose, total albumin, and triglyceride. The cholesterol of fish fed MBE in the diet showed incrementally enhanced. The present finding suggested that 1.0% MBE in the diet of *Clarias gariepinus* is recommended to enhance growth, survival, and blood profiles.

INTRODUCTION

Ant nest plant (*Myrmecodia pendens*) contains bioactive compounds such as glycoside, vitamin, mineral, flavonoid, tocopherol, polyphenol, and tannin (Engida *et al.*, 2013; Sanjaya *et al.*, 2014; Sudiono *et al.*, 2015) which are useful as antioxidant and anticancer. The ant-nest plant also has an abundance of high antioxidant properties and medical activities (Hanh *et al.*, 2016; Hertiani *et al.*, 2010; Soeksmanto *et al.*, 2010). Generally, the ant nest plant which can be found in several regions in Indonesia such as, Kalimantan and Papua uses as a traditional biomedicine such as supplement to recover after child birth in women and breastfeeding period (Firdausy and Nurlaila, 2016). The previous report stated that the ant-nest plant enhanced growth and blood profiles of *Pangasianodon hypophthalmus* (Nugroho *et al.*, 2019), boosted macrophage phagocytosis activity and lymphocytes proliferation (Sumardi *et al.*, 2013). Thus, the extract of this ant-nest plant might be the potential to be applied as a growth enhancer and immunomodulatory in fish such as *Clarias gariepinus*.

The *Clarias gariepinus* or known as catfish increasingly become an important commercial species in Europe, Africa, and part of Asia, including Indonesia. It is also one of the pivotal fish species cultured either indoor or outdoor in both tropical and subtropical regions (Sousa *et al.*, 2013; Yakubu *et al.*, 2014). The *C. gariepinus* has a high fecundity, resistances to diseases, and is easy to the captive, making its commercial importance species (Haylor and Mollah, 1995; Ljubobratovic *et al.*, 2015; Noor El-Deen *et al.*, 2014).

The health of fish can be evaluated by determining the immune status of fish using a blood profile (Chandel *et al.*, 2009). The blood profile such as red blood cells (RBC), white blood cells (WBC), hemoglobin level (Hb), and differential WBC (lymphocyte, monocyte, granular, and neutrophil) is a pivotal tool that can be performed to determine fish physiology (Inama *et al.*, 1993; Nugroho *et al.*, 2017; Nugroho *et al.*, 2016). Moreover, white blood cells have been generally used as an indicator to monitor the health indices of fish because white blood cell is an important part of the innate immune system, regulating fish immune defense (Ekman *et al.*, 2013; Zhou *et al.*, 2010). Besides blood profile, the intestine and hepar which are important digestive organs in the digestion system of nutrients from the feed are also pivotal organs to be used as a health indicator. Therefore, evaluating these organs are considered necessary as the digestive system is a good indicator for the nutritional status of fish and may relate to growth indices on feeding nutrition (Chowdhary *et al.*, 2013; Heikkinen *et al.*, 2006; Krogdahl *et al.*, 2003).

However, the information regarding the effects of *M. pendens* bulbs extract (MBE) on the growth and blood indices of *C. gariepinus* is limited. To evaluate the health and growth performance of fish, various physiological tools such as: the increase of either total leukocyte or differential leukocyte count (Adel *et al.*, 2015), phagocytosis activity (Bennani *et al.*, 1995; Chi *et al.*, 2016; Haugland *et al.*, 2012) and other blood parameters have been also successfully performed as indicators of the health and immune status of fish (Abidin *et al.*, 2016; Couto *et al.*, 2016; Jiang *et al.*, 2015). The survival rate has been also applied in a variety of fish as a pivotal physiological tool (Cai *et al.*, 2015). Thus, this research purpose was to determine the effects of different concentrations of dietary MBE addition on the growth and hemato-biochemistry of the blood profile of MBE-fed *C. gariepinus*. The fish survival rate was also recorded to evaluate the success of MBE supplementation in the diet.

METHODOLOGY

Place and Time

The present study had been conducted for 5 months (January-May 2020), starting from the preparation and trial of the study. All preparation, including extraction of Ant nest plant bulb, fish acclimatization, and experimental study had been performed in the Animal physiology, development and molecular laboratory, Department of Biology,

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Research Materials

The Ant nest plant bulb was provided from the local traditional market, Indonesia. The ant nest plant bulb was washed, cut, and ground, resulting in a powder of ant nest plant bulb. The bulb powder was then extracted by using ethanol 96% for 2 days, followed by filtration. The filtrate was evaporated (Rotary evaporator), resulting in a crude extract. The crude extract was stored at 4 °C until being used. Meanwhile, fish were obtained from Local fish farmer, Samarinda East Kalimantan and acclimated for one week at Laboratory of Animal Physiology, Department of Biology, Universitas Mulawarman, Kalimantan Timur.

Research Design

A completely randomized design (CRD) which is the simplest design has been used in this present study. The current study used independent variables in the form of ant nest plant bulb extract with 5 various concentrations, including control. All various concentration was in triplicates, containing 15 fish per replication.

Work Procedures

Basal and Test Diet Preparation

The basal diet was a commercial diet (Hi Pro Vite FF-888), containing 36-38% crude protein, 2% crude lipid, 10% ash, 12% moisture, and 2% crude fiber. Meanwhile, a test diet was obtained by adding basal diet at different concentrations of MBE (0.25; 0.50; 1.0; 2.0%) and repelletized (0.5 mm in diameter, 0.5 mm in length) using a mincer and then dried in the oven at 50 °C. The dried pellets were cooled, placed at room temperature, and packed with plastic bags. the pellet was stored in a dark room, until being used as a control-basal diet (Without MBE addition) and test diets.

Animals and Experimental Preparation

In total two hundred and twenty-five fish (27.48±0.16 g initial weight) and randomly grouped into five triplet groups of fifteen fish each group. Each group of fish was then placed in a plastic tank container (60 L sized, 40 L freshwater in each tank). For 75 days, fish in each group was fed with several concentrations of MBE. Temperature, pH, and Dissolve Oxygen (DO) were measured every two weeks using a routine thermometer, pH meter, and TOA-dkk pH HM-7, TOA instrument, Japan. The fish in each plastic container tank was fed with a basal or treatment diet (3% of the bodyweight of fish per day). The remaining uneaten feed and feces were taken out by siphoning before adding fresh water.

Growth and Survival

On the initial and final day of the feeding trial, the initial (IW), final weight (FW), initial (IBW) and final biomass weight (FBW), body weight gain (BWG), daily weight gain (DWG), average weekly gain (AWG), specific growth rate (SGR), feed conversion ratio (FCR), feed efficiency (FE), and survival rate (SR), were calculated to measure the growth indices and feed utilization of fish fed with different concentrations of MBE. All growth indices were measured following previous research (Abdel-Tawwab *et al.*, 2015; Githukia *et al.*, 2015; Havas *et al.*, 2015; Omosowone *et al.*, 2015). Meanwhile, the survival rate of fish in each tank was noted every 2 weeks and calculated following the formula previously used by Okomoda *et al.* (2017).

Blood Profile

At the end of day 75, blood samples (n=6 fish per tank) were taken from the tail. Total leukocyte (10^3 per mm^3), the percentage of neutrophil, lymphocyte, monocyte, and Red blood cell (RBC), and

Hemoglobin (Hb) were evaluated by using Hematology Analyzer Mindray (BC2800, Mindray® Shenzhen, China). Meanwhile, plasma biochemistry glucose, total cholesterol, triglyceride was measured following the protocol of the assay kits (Sigma Aldrich, USA). Albumin was determined by using bromocresol green reagent and detected with a microplate reader (HBS-1101 Microplate Reader, China) at 630 nm.

Viscera Index

Hepar and intestines of the fish (n=6 per tank) were collected and weighed to measure the hepatosomatic (HSI) and intestinal somatic index (ISI) at the end of day 75. Both HIS and ISI were calculated using the equation described by Zhao *et al*., (2015).

Data Analysis

All data obtained are shown as means ± standard error (SE) and data were analyzed using SPSS version 24 (SPSS, Inc., USA). The data of the percentage of leukocyte, neutrophil, lymphocyte, monocyte, and survival were transformed to arcsine and subjected to one-way ANOVA, followed by Duncan Multiple Range Test to evaluate significant differences among the group of treatments. All significant tests were at $P<0.05$ levels.

RESULTS AND DISCUSSION

The average temperature, pH, DO, nitrate, ammonia during project research was 26.13 ± 0.12 °C, pH 7.45 ± 0.21, DO 6.01 ± 0.21 ppm, nitrite 0.10 ± 0.02 ppm, ammonia 0.10±0.01 ppm that classified in the range for *C. gariepinus* culture. The present finding showed that fish fed MBE with 0.5-1% concentration had significantly higher (P<0.05) final weight, final biomass weight, BWG, DWG, AWG, and SGR. Fish fed 1% of MBE in the diet had significantly better FCR (1.13±0.03) than control and 0.25-0.5%. The highest FCR (2.04±0.04) was found in the fish supplemented with 2% MBE in the diet. Survival of all fish groups was not affected by any concentration of MBE supplementation in the diet of *C. gariepinus* (Table 1).

Table 1. Mean ± SE of growth parameters and visceral somatic index of *Clarias gariepinus* fed *Myrmecodia pendens* bulb ethanolic extract in the diet for 75 days.

Parameter	Groups				
	Control	0.25%	0.5%	1%	2%
IW (g)	27.38±0.44[a]	27.24±0.47[a]	27.68±0.42[a]	27.53±0.61[a]	27.54±0.47[a]
FW (g)	85.09±1.23[a]	86.81±1.90[a]	142.26±1.62[b]	138.99±1.81[b]	137.38±1.10[b]
IBW (g)	412.46±0.27[a]	411.40±0.24[a]	394.10±1.67[a]	413.42±1.71[a]	413.16±1.14[a]
FBW (g)	1242.38±33.14[a]	1281.85±21.81[a]	2021.35±91.71[b]	2024.38±40.34[b]	1987.48±26.05[b]
BWG	57.59±1.23[a]	59.39±1.90[a]	114.75±1.53[b]	111.42±3.92[b]	109.84±1.11[b]
DWG	0.76±0.01[a]	0.79±0.02[a]	1.53±0.02[b]	1.48±0.05[b]	1.46±0.17[b]
AWG	5.75±0.12[a]	5.93±0.19[a]	11.47±0.15[b]	11.14±0.39[b]	10.98±1.31[b]
SGR	1.5±0.01[a]	1.53±0.02[a]	2.19±0.01[b]	2.15±0.04[b]	2.13±0.12[b]
FCR	1.68±0.01[a]	1.62±0.007[a]	1.51±0.07[a]	1.13±0.03[b]	2.04±0.04[c]
Survival (%)	91.11±8.89[a]	100.00±0.00[a]	100.00±0.00[a]	100.00±0.00[a]	100.00±0.00[a]

Note: IW = Initial weight, FW = Final weight, IBW = Initial biomass weight, FBW = Final biomass weight, BWG = Body weight gain, DWG = Daily weight gain, AWG = Average weekly gain, SGR = Specific growth rate, FCR = Feed conversion ratio, HSI = Hepatosomatic indices, ISI = Intestinal somatic indices. Different alphabets (a,b,c) indicate significantly different means for different group of diets at $p<0.05$. Control diet without MBE (*Myrmecodia pendens* bulbs ethanolic extract) supplementation.

Further, supplementation MBE higher than 0.5% in the diet of fish affected on RBC, WBC, hematocrit, PLT, and lymphocyte. Meanwhile, fish fed 0.25-2% MBE in the diet resulted in significantly increased hemoglobin.

However, neutrophils and monocytes of fish were not affected by any concentration of MBE supplementation in the diet (Table 2). Fish fed MBE in a diet higher than 0.5% resulted in significantly different glucose, total albumin, and triglyceride. The incremental addition of MBE in the diet showed significantly stepping up Cholesterol in the blood plasma of fish until 1% of MBE (Table 3).

Table 2. Mean ± SE of blood profiles of *Clarias gariepinus* fed *Myrmecodia pendens* bulb ethanolic extract in the diet for 75 days.

Parameters	Groups				
	Control	0.25%	0.5%	1%	2%
RBC ($10^6 \mu L^{-1}$)	0.98 ± 0.09^a	0.86 ± 0.14^a	1.16 ± 0.13^a	1.61 ± 0.01^b	$1.60 \pm 0.04a^b$
WBC ($10^3 \mu L^{-1}$)	13.34 ± 1.78^a	16.30 ± 0.77^{ab}	18.79 ± 2.66^{ab}	26.89 ± 2.05^c	20.94 ± 2.12^{ab}
Hemoglobin (g dL^{-1})	5.71 ± 0.12^a	7.04 ± 0.52^b	7.41 ± 0.41^b	8.82 ± 0.12^c	8.84 ± 0.13^c
Hematocrit (%)	14.31 ± 1.43^a	15.98 ± 1.15^{ab}	15.37 ± 0.86^a	18.54 ± 0.37^b	16.54 ± 0.63^{ab}
PLT ($10^3 \mu L^{-1}$)	16.14 ± 0.54^a	15.85 ± 0.20^a	27.71 ± 0.24^a	38.42 ± 0.63^b	25.28 ± 0.11^{ab}
Neutrophil ($10^3 \mu L^{-1}$)	0.35 ± 0.05^a	0.67 ± 0.04^a	0.65 ± 0.03^a	0.91 ± 0.03^a	1.38 ± 0.04^a
Lymphocyte ($10^3 \mu L^{-1}$)	12.83 ± 1.77^a	8.39 ± 1.16^b	16.57 ± 1.02^a	23.02 ± 0.29^b	19.37 ± 1.89^c
Monocyte ($10^3 \mu L^{-1}$)	0.14 ± 0.04^a	0.17 ± 0.07^a	0.24 ± 0.09^a	0.30 ± 0.01^a	0.32 ± 0.05^a

Note: RBC = Red blood cell, WBC = White blood cell, PLT = Platelet. Different alphabets (a,b,c) indicate significantly different means for different group of diets at $p<0.05$. Control diet without MBE (*Myrmecodia pendens* bulbs ethanolic extract) supplementation.

Table 3. Mean±SE of blood plasma biochemistry of *Clarias gariepinus* fed *Myrmecodia pendens* bulb ethanolic extract in the diet for 75 days.

Parameters (mg dL^{-1})	Groups				
	Control	0.25%	0.5%	1%	2%
Glucose	14.30 ± 0.06^a	14.45 ± 0.08^a	$14.50.33 \pm 0.08^a$	15.71 ± 0.05^b	16.24 ± 0.03^c
Total albumin	4.35 ± 0.07^a	4.45 ± 0.06^a	4.46 ± 0.04^a	5.16 ± 0.06^b	5.13 ± 0.07^b
Cholesterol	141.90 ± 1.75^a	113.28 ± 0.76^b	101.51 ± 0.51^c	93.78 ± 0.52^d	93.16 ± 0.53^d
Triglyceride	78.78 ± 0.63^a	79.15 ± 0.31^a	78.46 ± 0.69^a	71.28 ± 0.61^b	64.30 ± 1.55^c

Note: Different alphabets (a,b,c,d) indicate significantly different means for different group of diets at $p<0.05$. Control diet without MBE (*Myrmecodia pendens* bulbs ethanolic extract) supplementation.

Furthermore, in visceral value, the highest hepatosomatic index (HSI) was found on fish fed 1-2% MBE in the diet, while the intestinal somatic index (ISI) was not affected by the addition of any concentration of MBE in the diet of fish (Table 4).

Table 4. Hepatosomatic (HSI) and intestinal somatic index (ISI) of *Clarias gariepinus* fed dietary *Myrmecodia pendens* bulb extract (MBE) in the diet for 75 days.

Parameters	Groups				
	Control	0.25%	0.5%	1%	2%
HSI	1.11 ± 0.41^a	1.23 ± 0.31^a	1.54 ± 0.25^a	2.74 ± 0.42^b	2.94 ± 0.32^b
ISI	1.62 ± 0.32^a	3.33 ± 0.66^a	1.66 ± 0.26^a	1.80 ± 0.20^a	2.41 ± 0.40^a

Note: Different alphabets (a,b) superscripts on the same row indicate significantly different means for a different group of diets at $p<0.05$. Control diet without MBE (*Myrmecodia pendens* bulbs ethanolic extract) supplementation.

Recently, the use of the plant as a feed additive to replace antibiotics for enhancing the growth parameters, health indices and meat quality of fish due to the phytochemicals such as flavonoids, phenolics, and pigments is gaining in popularity. Dietary inclusion of some plant-derived substances has been also considered and proved to have a great economic value in the aquaculture field. The application of plant extracts in aquaculture fields to boost growth factors and immunity has attracted researchers due to their active ingredient (Abdel-Tawwab *et al.*, 2018a; Adeshina *et al.*, 2018; Farsani *et al.*, 2019; Rahman *et al.*, 2018; Tan *et al.*, 2018).

The active ingredient derived from ethanolic plant extracts has been confirmed, containing active compounds namely saponin, triterpenoid, flavonoid, alkaloid, phenolic, and tannin (Barrett *et al.*, 2018; Ogunleye *et al.*, 2019). Some important secondary metabolite phytochemical compounds which abundant with antioxidant properties has been also found in ant nest plant (Sari *et al.*, 2017) that might be useful for the animal, such as fish. The present finding revealed that the MBE addition in the diet of *C. gariepinus* improved growth indices such as final weight, final biomass weight, BWG, DWG, AWG, and SGR. This improvement might be due to the occurrence of phytochemical compounds which can act as primary antioxidants that related to fish physiology (Rattanachaikunsopon and Phumkhachorn, 2007) and as a growth stimulant on juvenile *Pargus major* (JI *et al.*, 2007), *Carassius auratus* (Ahilan *et al.*, 2010), *Catla catla* (Kaleeswaran *et al.*, 2011).

This finding is similar to previous results performed by Izzreen and Fadzelly (2013) who used flavonoid-containing Green Tea, *Camellia sinensis* L that enhanced the growth of Nile Tilapia, *Oreochromis niloticus* (Abdel-Tawwab *et al.*, 2010). Moreover, phytochemical compounds such as triterpenoid, flavonoid, alkaloid, quinone, and phenolic have also been confirmed to increase many physiological indicators such as appetite, tonic, and immunity (Awad *et al.*, 2019; Chakraborty *et al.*, 2012; Sinha and Jindal, 2019).

Phytochemical content may be beneficial to enhance the innate immune system of fish to support their survival (Chakraborty *et al.*, 2012). The previous finding confirmed that phytochemical extract has successfully promoted the survival rate of *Cyprinus carpio* (Mohamad and Abasali, 2010); (*Oreochromis niloticus*) (Akinwande *et al.*, 2011), and marine ornamental fish (Dhayanithi *et al.*, 2013). Dhanalaxmi and Vastrad (2014) also stated that active phytochemicals from *Cinnamomum verum* increased the survival rate of *Oreochromis niloticus* post-*Aeromonas hydrophila* challenge (Abdel-Tawwab *et al.*, 2018b). The present study, however, revealed that *C. gariepinus* fed MBE any concentration did not affect the survival rate. In contrast, another study revealed that the presence of tannin in the plant might be harmful to fish at high doses and has negative effects on fish such as *Cyprinus carpio* and *Channa striatus* (Viswaranjan *et al.*, 1988).

Blood indices are a pivotal tool to determine fish health (He *et al.*, 2015; Mallik *et al.*, 2019; Suely *et al.*, 2016; Wang *et al.*, 2014). Blood indices such as red blood cells, white blood cells, hemoglobin, hematocrit value, and platelet are pivotal parameters to determine the physiological status of fish. Current research revealed that groups of fish fed with diet mixed with MBE showed significantly higher RBC, WBC, and Hb than the control group, confirming that MBE had beneficial to improved blood function properties.

A previous study found that the ethanolic extract of *M. tuberosa* that also contains phytochemical active such as phenolic improves the blood profile and immune system (Firdausy *et al*., 2016). The immunity of fish correlates with blood profile which is a pivotal indicator in the monitoring of fish health (Moazenzadeh *et al.*, 2017; Simide *et al.*, 2016; Soberon *et al.*, 2014). Blood indices such as RBC, WBC, Hb, Hct, and PLT can be used to evaluate fish physiological conditions (Bilen *et al.*, 2019; Velichkova *et al.*, 2019). The WBC is generally used to monitor fish's health status because it is an important parameter to their innate immune defense and functioning (Franz *et* *al.*, 2016; Kumar *et al.*, 2019). The present research found that WBC, Hb, and PLT of fish fed MBE above 0.25% in the diet showed significantly higher improvement than a control group. This finding is in line with previous studies, stating that plant extracts which contain active phytochemicals may enhance the value of WBC, Hb, PLT, neutrophil, monocyte, and lymphocyte in fish (Gavriil *et al.*, 2019; Babahydari *et al.*, 2014; Yuniar *et al.*, 2017).

The mechanism of MBE in increasing blood parameters in the fish is not clearly defined and needs further research. Nevertheless, Nair *et al.* (2002); Lyu and Park (2005) stated that flavonoids from plant extract may boost IL-2 (Interleukin 2) and INFγ (Interferon) as bio catalysator in WBC metabolism which is important in nonspecific cellular immunity. Further, this mechanism also helps in decreasing RBC hemolysis and protecting the bio membrane of RBC from oxidative damage that destructs by free radicals (Asgary *et al.*, 2005; Kitagawa *et al.*, 1992). Thus, the current finding is similar to past research, confirming that MBE is capable of an antioxidant that can be used to protect the heme iron of RBC and increase erythropoiesis (Hamed and El-Sayed, 2019; Shatoor, 2011; Uboh *et al.*, 2010).

The blood biochemical properties are useful to reflect fish health conditions and nutritional metabolism that determine the health performance of the fish in response to dietary supplementation (Hassaan *et al.*, 2019; Turan and Gezer, 2018). The levels of triglycerides and cholesterol as energy metabolites are pivotal parameters in fish health (Eckel *et al.*, 2005). Either triglyceride or cholesterol has some important biological functions such as storing energy, signaling, and acting as pivotal structural components of cell membranes. The change value of both energy metabolites may lead to health disturbance in most vertebrate species. Moreover, triglycerides level is a general indicator of the health of liver function, while cholesterol level is a nutritional status (Brum *et al.*, 2018).

The current study revealed that Fish fed MBE in the diet above 0.5% resulted in significantly decreased cholesterol and triglyceride, while glucose and total albumin of fish were significantly increased. This finding is similar to the previous study, performed by Brum *et al.* (2018) who stated that cholesterol and triglycerides of Nile tilapia fed 0.5-1.5% clove basil in their diet showed a significant reduction. Other reports revealed that the albumin, cholesterol, glucose, and triglyceride levels in the blood plasma of *Oreochromis mossambicus* fed dietary medicinal plant extracts groups showed significantly higher than fish in control groups (Immanuel *et al.*, 2009).

In contrast, hybrid grouper (*Epinephelus lanceolatus* ♂ × *Epinephelus fuscoguttatus* ♀) fed dietary *Panax notoginseng* extract in the diet found significantly reduce glucose level in the plasma (Sun *et al.*, 2018). According to Ribeiro *et al.* (2016) feeding a fish for example Tambaqui *Colossoma macropomum* with diets containing plant extract improved plasma glucose and showed no stress in fish treatment groups, similar to those of the control group. Moreover, the increase in total albumin may also relate to sufficient feed and supplementation intake.

The value of hepatosomatic (HSI) and intestinal somatic index (ISI) are parameters that can be used as a liver and intestinal health indicator for fish (Chakraborty *et al.*, 2015; Elabd *et al.*, 2019). The present study found that supplementation of MBE between 1-2% resulted in significantly higher HSI than other groups, while dietary any concentration of MBE in the diet did not change ISI of *C. gariepinus*. This finding is supported by a past report that revealed that there were no significant differences in viscerosomatic index among all groups of hybrid grouper (*Epinephelus lanceolatus* ♂ × *Epinephelus fuscoguttatus* ♀) fed dietary ginkgo biloba leaf extract (Tan *et al.*, 2018).

Another report also stated that Nile tilapia (GIFT strain) fed an *Aloe vera* addition in the diet resulted in a significant increase in HSI, but viscerosomatic indices shown not significantly different (Panase *et al.*, 2018). The increasing HSI may be due to the phytochemicals in the MBE extract that can trigger a fish's hepatic cells to boost their ability to store biochemical nutrients in the body of fish such as glucose, amino acids, and lipid. Further, the biochemical nutrients can be released into the bloodstream, transferred to target cells, and converted into energy (Lucas and Watson, 2002). In addition, the high HSI also reflects the increment of liver cell size which can enhance growth, store more lipid in the fish body to maintain energy level, and combat some environmental stressors (Klaunig *et al.*, 1979; Panase *et al*., 2018).

CONCLUSION

The supplementation of *Myrmecodia pendens* bulb extract (MBE) supports growth, increases blood indices, and plasma biochemistry of *Clarias gariepinus.* Dietary 1% MBE in the feed of *C. gariepinus* is beneficial and recommended to increase the growth and blood parameters function of the fish. Nevertheless, further research needs to be done to evaluate the phytochemical active ingredient of those plants on fish physiology (including antioxidant activity and responses molecular). In addition, a challenge test using fish pathogenic bacteria concerning the effects of MBE supplementation need to be done to evaluate the effects of MBE on the immune system and other physiology parameters.

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