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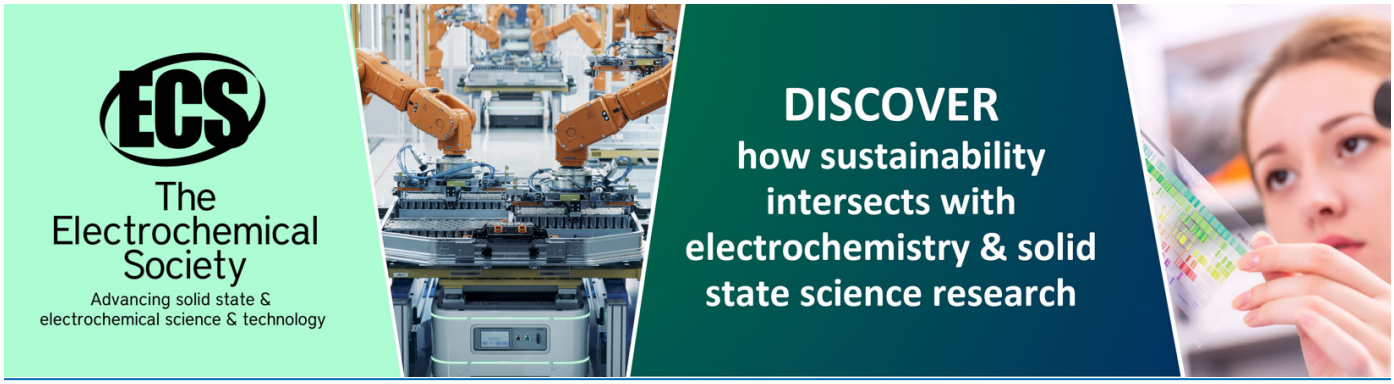

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Nutritional Value Content, Biomass Production and Growth Performance of *Daphnia magna* Cultured with Different Animal Wastes Resulted from Probiotic Bacteria Fermentation

Vivi Endar Herawati*, R A Nugroho, Pinandoyo and Johannes Hutabarat

Department of Aquaculture, Faculty of Fisheries and Marine Science, Diponegoro University., Semarang 50275, Indonesia

email : anshinvie@yahoo.com

Abstract. Media culture is an important factor for the growth and quality of *Daphnia magna* nutrient value. This study has purpose to find the increasing of nutritional content, biomass production and growth performance of *D. magna* using different animal wastes fermented by probiotic bacteria. This study conducted using completely randomized experimental design with 10 treatments and 3 replicates. Those media used different animal manures such as chicken manure, goat manure and quail manure mixed by rejected bread and tofu waste fermented by probiotic bacteria then cultured for 24 days. The results showed that the media, which used 50% chicken manure, 100% rejected bread and 50% tofu waste created the highest biomass production, population and nutrition content of *D.magna* about 2111788.9 ind/L for population; 342 grams biomass production and 68.85% protein content. The highest fatty acid profile is 6.37% of linoleic and the highest essential amino acid is 22.8% of lysine. Generally, the content of ammonia, DO, temperature, and pH during the study were in the good range of *D. magna's* life. This research has conclusion that media used 50% chicken manure, 100% rejected bread and 50% tofu waste created the highest biomass production, population and nutrition content of *D. magna.*

Keywords: Nutrient value, Fermentation, *D. magna*, Animal wastes, probiotic bacteria

1. Introduction

D. magna is one of the most potential natural feed for fish larvae due to its high nutritional content [1]. Nutritional content of *D. magna's* largely depends on its culture medium as the growth of phytoplankton, which is the feed for *D. magna*, depends on it [2]. The most commonly used culture medium nowadays is the used chicken manure [3]. Another less common one is a combination of chicken dung, bran, and copra waste [4]. The organic materials in bran are highly nutritious for the growth of *D. magna*.

The use of organic fertilizers in culture media including the wastes/feces of chicken, goat and quail mixed with the rejected bread and tofu waste fermented with the probiotic bacteria has not so far been conducted as the use of organic fertilizer could impact the growth performance and content of *D*. magna. The highest nutrients - particularly for the content of N, P and Ca in organic fertilizer are the food sources of *D. magna*. The chicken waste containing N (4.75%); P (3.57%) and Ca (4.80%); quail waste containing N (4.06%); P (2.96%) and Ca (2.57%); and goat waste/feces

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containing N (2.36%); P (2.96%) and Ca (3.41%) [1]. Furthermore, the analysis on the dried materials of tofu waste contained crude protein (27.09%), crude fibre (22.85%), fat (7.37%), ash (35.02%), and extract material without nitrogen/BETN (6.87%) [5], [6]. The rejected bread contained the crude protein (12.63%), crude fibre (0.13%); crude fat (4.63%); ash (4.19%) and the extract material without nitrogen (58.42%) [6], [7].

The fermentation of the fertilizer has been proven to be effective in the increase of the nutrient of culture media. The aims of the fermentation are to produce a product (food materials) that contains the nutrients, to have a longer storage time, and to have a better organoleptic characteristics and nutritional components [1]. The mass-cultured *D. magna* in this research used probiotic bacteria as it improves both biomass production and nutritional content of *D. magna*. Probiotic bacteria are supportive for the health of organisms [8]. It also serves to decompose and ferment organic materials [9]. Decomposition is a biological process that makes the most of bacteria's ability to produce growth substances, hormones, vitamins, and other enzymes [3], [10].

The purpose of this study is to find nutritional content, biomass production and growth performance of *D. magna* using different animal wastes fermented by probiotic bacteria to increase its nutrient content, biomass production and the growth. The fertilizer itself is fermented with probiotic bacteria (*Lactobacillus casei* and *Saccharomyces cerevisiae*)

2. Materials and Method

2.1 Fermentation Stage

The fermentation stage is the preparation of the ratio of molasses, water and pro-biotic. The ratio used was 1: 1, i.e. 1mL of molasses, 1 mL of probiotic bacteria and 100 mL of solvent. The organic materials used (chicken manure, goat manure, quail manure, rejected bread and tofu wastes) are dried. The treatments used in this research were: A. rejected bread 100 gr/L + tofu waste 100 gr/L; B. Chicken manure 50 gr/L + rejected bread 50 gr/L + tofu waste 100 gr/L; C. Chicken manure 50 gr/L + rejected bread 100 gr/L + tofu waste 50 gr/L; D. Chicken manure 100 gr/L + rejected bread 50 gr/L + tofu waste 50 gr/L; E. Goat manure 50 gr/L + rejected bread 50 gr/L + tofu waste 100 gr/L; F. Goat manure 50 gr/L + rejected bread 100 gr/L + tofu waste 50 gr/L; G. Goat manure 100 gr/L + rejected bread 50 gr/L + tofu waste 50 gr/L; H. Quail manure 50 gr/L + rejected bread 50 gr/L + tofu waste 100 gr/L; I. Quail manure 50 gr/L + rejected bread 100 gr/L + tofu waste 50 gr/L; I. Quail manure 100 gr/L + rejected bread 50 gr/L + tofu waste 50 gr/L; J. 50 gr/L rejected bread 100 gr/L + tofu waste 100 gr/L.

Fertilizers with a combination weight of 200g/L were given pro-biotic bacteria (*L. casei* and *S. cerevisiae*) that were already activated for 3 hours [9], [11]. They were left for fungi to grow and acidic smell to develop. Table 1 and 2 shows the result of nutrient analysis for organic fertilizer before fermented and after fermented for 14 days with pro-biotic bacteria for the mass-cultured *D. magna.*

2.2 Water Quality

The water quality during the research was maintained at 28-29^{0}C temperature, 0.3 ppm DO and 8,1-8,2 pH, which is ideal. This is in line with the statement of previous research [1], [12], [13] that the proper temperature for *D. magna* culture is 25-30^{o}C, DO at 0.3-0.6 ppm, and pH at 6.5-9. Excellent water quality helps grow phytoplankton and algae for *D. magna* to propel its growth.

2.3 D. magna Culture

The 100 ind/l *D. magna* was spread for each pool containing 200 g/L fermented organic fertilizer. Observation for the abundance of D. *magna* was conducted every two days to monitor the population of *D. magna*. The water (20-25%) of this culture was replaced, and its pH level was monitored every morning at around 7 to maintain the quality. The pH was maintained at its maximum with the addition of 1 L of dolomite /1.000 L of water.

2.4 Statistical Analysis

This research employed the complete random design with 10 treatments and 3 repetitions. The weight of biomass was analyzed using variant analysis to figure out differences among treatments. The parameters analyzed were growth, biomass production, and nutritional content of *D. magna*.

Proximate analysis. The proximate chemical composition of the samples was determined using a standard procedure [6], [14]. The crude protein content was calculated by multiplying the total nitrogen factor. The carbohydrate content was estimated by the difference.

Essential amino acid profile. The amino acid composition of the sample was determined using HPLC (Shimadzu LC-6A) [6], [14].

Fatty acid profile. The fatty acid composition of the sample was determined using a gas chromatograph (Shimadzu) [6], [14].

3. Results and Discussion

The results of the research found the culture media before and after being fermented experiencing an enhancement in which N before the fermentation was in the range of N: 1.19 - 2.75%; P: 0.19 - 0.54%; and K (Potassium): 0.15 – 0.59% and after the fermentation was in the range of 2.12-3.84%; P: 0.27-1.74%; and K (Potassium): 0.69-1.21%. Table 1 and Table 2 show the results of the analysis on N, P and K (Potassium) culture medium before and after fermentation.

Table 1. The results of the analysis on N, P and K (Potassium) culture medium before fermentation.

Parameters	Analysis Result										Method
	Poultry manure (%)	Chicken manure (%)			Goat manure (%)			Quail manure (%)			
	A	B	C	D	E	F	G	H	I	J	
Nitrate (N)	1,54± 0.06	2,24± 0.09	2,55± 0.07	1.23± 0.02	1,25± 0.08	2,23± 0.01	1,78± 0.08	1,19± 0.05	2,19± 0.03	2,26± 0.09	Kjeldahl
Phosphor (P)	0,19± 0.03	0,18± 0.07	0,25± 0.01	0,23± 0.03	0.17 ± 0.05	1.03± 0.09	0.45± 0.06	0,41± 0.02	0,23± 0.08	0.54± 0.07	AOAC 958.01. 2000
Potassium (K)	0,39± 0.02	0,27± 0.01	0,59± 0.05	0.26± 0.06	0.33± 0.09	0.45± 0.03	0,15± 0.03	0,21± 0.09	0,13± 0.03	0,54± 0.02	AOAC

Table 2. The results of the analysis on N, P and K (Potassium) culture medium after fermentation.

Parameters	Analysis Result										Method
	A	B	C	D	E	F	G	H	I	J	
Nitrate (N)	2,74± 0.05	3,40 ± 0.05	3,84 ± 0.03	2.43 ± 0.09	2,12 ± 0.08	3,29 ± 0.02	2,98 ± 0.06	2,89 ± 0.01	3,29 ± 0.03	2,37 ± 0.09	Kjeldahl
Phosphor (P)	0,27± 0.02	0,48 ± 0.04	1,43 ± 0.06	0,59 ± 0.03	1,14 ± 0.02	1,30 ± 0.01	1,16 ± 0.01	1,21 ± 0.07	1,33 ± 0.02	1,24± 0.09	AOAC 958.01.200 0
Potassium (K)	0,69± 0.09	1,21 ± 0.02	0,89 ± 0.08	0,86 ± 0.03	1,56 ± 0.03	1,11 ± 0.09	1,05 ± 0.05	1,41 ± 0.01	0,34 ± 0.03	0,96 ± 0.06	AOAC 958.01.200 0

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This research is the development of the research on the massive culture of *D. magna* using the chicken manure, rice bran and residue of copra fermented using the probiotic bacteria in 2015. The

research found that the organic material in the form of a variety of the fermented animal manures mixed with rejected bread and waste tofu experienced an increase for its nutrient quality. The increases of the content included N at 1.2%; P and K at 1% (Table 1 and Table 2), the improvement of the nutrient quality on the media fermented is the process of anaerobic dissimilation of organic compounds by the microorganisms activity or the extract of microorganisms cells. The factors affecting biomass and the nutrient content of *D. magna* included the nutrient quality of the media, availability of food in the form of phytoplankton, bacteria detritus and the environment [1], [2]. The organic materials in the fermented media can increase the number of bacteria and organic particles from decomposition result to increase the availability of nutrients in the media. This then influenced the population growth and biomass of *D. magna*. This is supported by the results of previous research [8] that fermentation is aimed to increase the number of microorganisms and activate the metabolism in food, resulting in new food products using microorganisms.

The increase of nutrient in the media, especially nitrate, functions to determine the level of phytoplankton present in the culture medium – in addition to bacteria and detritus - as the food source of *D. magna*. The results of this study stated that the promotion and the growth of phytoplankton population in waters are associated with the nutrient availability, especially nitrates and the sunlight confirmed by previous research [6], [15]. This argument has been proven through the results of this research that the high level of nutrient of media brought an influence on the growth and absolute biomass of *D. magna* the highest growth was found in media of the fermentation of 50 g/L of chicken manure + 100 g/L of rejected bread + 50gr/l tofu waste with a content of N and P at 3.84% ± 0.03 and 1.43% ± 12:06 respectively providing the growth of 211788.9 ind /L (Chart 1 and Table 3) and the highest biomass was at 345 g ± 2.64 (Graph 2) .The quality of the nutrient culture media is very influential for the supply of plankton and bacteria to boost the population growth and biomass of *D. magna*. The high level of organic materials can bring an effect on the density and biomass of *D. magna* [2]. Growth performance *D. magna* cultured using the different manures of chicken, goat, quail, rejected bread and tofu waste fermented using probiotic bacteria as illustrated in Figure 1:

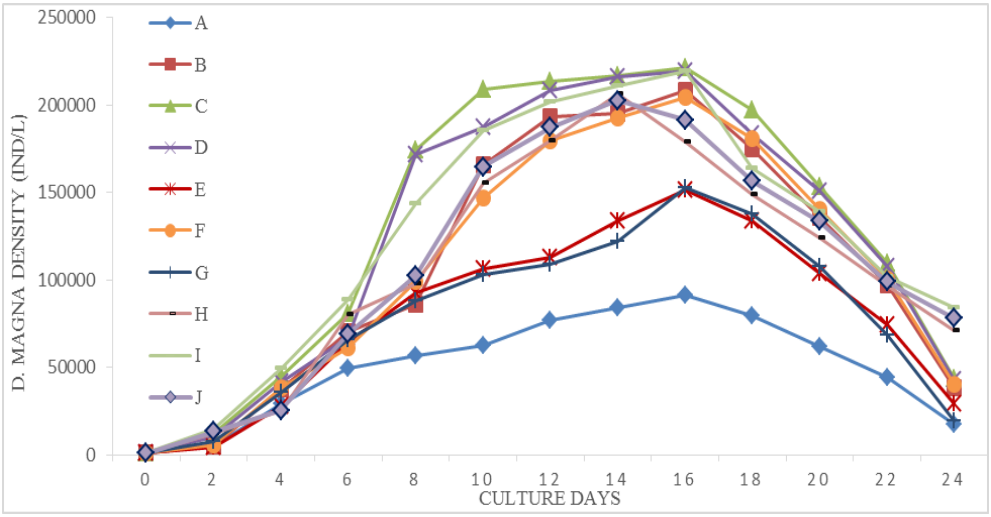

Figure 1. Growth performance of *D. magna* cultured using the different manures of chicken, goat, quail, rejected bread and tofu waste fermented using prebiotic bacteria.

The research found that the growth of *D. magna* massively cultured by means of a mix of animal different animal wastes with the rejected bread and tofu waste at the lag phase and death phase did not give any significant influence ($p<0.01$) among treatments in contrast to the exponential and stationary phases showing a significant influence ($p>0.01$) between treatments. The results of the highest growth

on *D. magna* mass massively cultured using the fermentation of 50 g/L of chicken manure + 100 g/L of rejected bread+ 50 g/L waste tofu (C) with a dense population of the lag phase at 46251.85 ind/L on day 4; exponential phase at 198955 ind/L on day 10; stationer phase at 211788.9 ind /L on day 16 and the death phase at 102177.8 ind/L on day 26. Table 3 presents the results of the growth of *D. magna* massively cultured using the fermentation of a mix of different animal wastes with rejected bread and tofu waste.

Table 3. Growth of *D. magna* massively cultured using the fermentation of a mix of different animal wastes with rejected bread and tofu waste.

Treatment	A (%)	B (%)	C (%)	D (%)	E (%)	F (%)	G (%)	H (%)	I (%)	J (%)
Lag	2809.63 ± 0.05^{a}	37622.22± 0.01 ^{a}	46251.85± 0.01 ^{a}	40048.15 ± 0.05 ^{a}	32437.04 ± 0.05^{a}	35085.19 ± 0.02^{a}	25646.3 ± 0.01^{a}	39578.9 ±0.04 ^{a}	42312.2 ±0.04^{a}	42256.7 ±0.02^{a}
Exponential	65418.52 ± 0.07^{bdc}	153233.3± 0.03 ^{a}	198955.6± 0.02 ^{a}	189140.7 ± 0.04 ^{a}	100863 ± 0.02^{a}	141863 ± 0.01^{dba}	90362.96 ± 0.06^{a}	167423.3 ±0.03 ^{a}	209656.7 ±0.02^{c}	174723.3 ±0.01^{a}
Stationer	85270.37 ± 0.03^{a}	208000 ± 0.02 ^{ac}	211788.9± 0.05 ^{ab}	209603.7 ± 0.02^{ba}	139548.1 ± 0.09^{da}	192714.8 ± 0.05^{bda}	90770.37 ± 0.05^{a}	178101.1 ± 0.06^{a}	195778.9 ± 0.07^{c}	189478.9 ±0,04^{a}
Death	41400 ± 0.02^{a}	90640.74± 0.09 ^{a}	102177.8± 0.04 ^{a}	100.418.5 ±0.08^{a}	69270.37 ±0.01^{a}	93788.89 ± 0.04^{a}	45323.93 ± 0.03^{a}	974233.3 ± 0.02^{a}	101356.7 ±0.02^{a}	100678.9 ±0.01^{a}

The growth of *D. magna* massively cultured using the fermentation of various manures, rejected bread and tofu waste did not show a significant effect on the lag phase ($P<0.01$) between the treatments. The lag phase occurred on the fourth day and the highest one occurred in the media of fermentation of 50g/L of chicken manures + 100 g/L of rejected bread + 50gr /L of tofu waste at 46251.85 ± 0.01 ind/L. This was because at the lag phase *D. magna* began to adapt to the new environment in which if the concentration of media was equal to the nature, it would provide the fast growth and if there would be any differences in the culture medium and in the growth at a concentration of media in nature, *D. magna* would require more time for growing. The difference between the concentration of culture media and liquid cell in plankton would bring an effect on the restitution of enzyme and substrate concentrations to the further level for growth and the presence of nutrients in the cells was through the diffusion process as a result of differences in the concentration between the culture medium and body fluid [1].

The exponential phase occurred on the 10th day followed by a stationary phase on day 16. At the exponential phase and stationary phase, there was a significant influence ($P <0.01$) between the treatments. The length of the stationary phase was correlated with the duration of the *D. magna* to adapt to a new culture media. This was because the length of the stationary phase had an effect on the nutrient absorption into the culture medium by *D. magna*. The results of this research were consistent with previous research [1], [15], showing the discontinuation of the exponential phase due to the lack of nutrients in the increase of cell density. *D. magna* massively cultured using a mixture of rejected bread , tofu waste and various animal manures fermented using the probiotic bacteria provided a significant influence ($P >0.01$) at absolute biomass. *D. magna* massively cultured using the media of fermentation of 50 g/L of chicken manure + 100 g/L of rejected bread + 50 g/L of tofu waste (C) provided the highest absolute biomass at 345 g ± 2.64. Figure 2 presents the biomass of *D. magna* massively cultured using fermentation of a mixture of different manures, rejected bread and tofu waste.

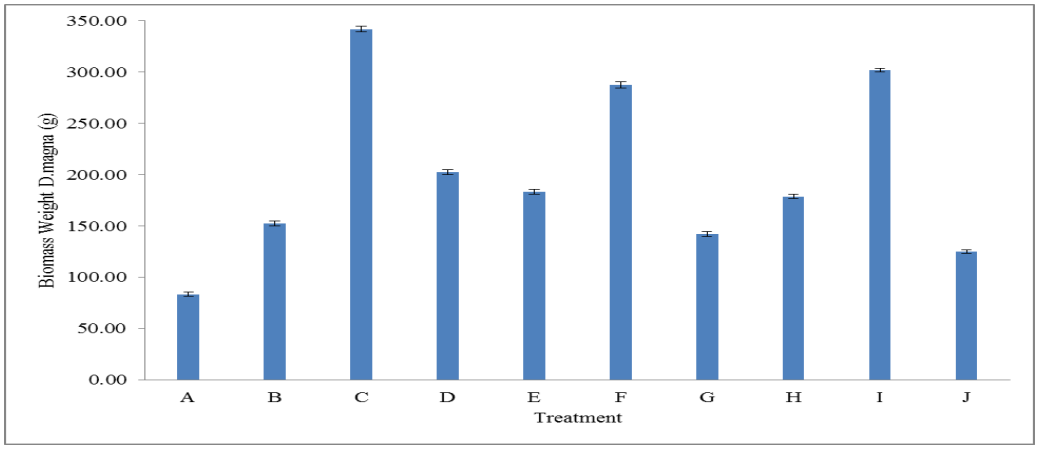

Figure 2. Biomass of *D. magna* massively cultured using fermentation of a mixture of different manures, rejected bread and tofu waste.

The media of fermentation of 50 g/L of chicken manure + 100 g/L of rejected bread + 50 g/L of tofu waste also resulted in a highest content of nutrients of *D. magna* that are 68.85 % ± 0.23 for protein and 8.84% ± 0.01 for fat. While, for the lowest level of nutrient content of *D. magna* massively cultured using the media of fermentation A (100 g/L of rejected bread + 100 g/L of tofu waste) with 60.32 % ± 0:04 and 7.16 %± 0:08 for protein and fat respectively. Table 4 presents the results of the analysis on the proximate of *D. magna* massively cultured using the fermentation of the mix of various animal manures, rejected bread and waste tofu.

Table 4. The result of the proximate composition of *D. magna* in mass culture using the different animal wastes resulted from probiotic bacteria fermentation.

Proximate	Poultry manure (%)	Chicken manure (%)			Goat manure (%)			Quail manure (%)		
	A	B	C	D	E	F	G	H	I	J
Crude ash	8.43 ± 0.07	8.67 ± 0.02	6.11 ± 0.08	6.52 ± 0.06	6.57 ± 0.09	6.60 ± 0.09	7.75 ± 0.23	7.75 ± 0.09	6.11 ± 0.09	7.99 ± 0.23
Crude lipid	7.16 ± 0.08	8.71 ± 0.03	8.84 ± 0.01	8.16 ± 0.09	8.34 ± 0.02	8.65 ± 0.03	6.95 ± 0.06	7.25 ± 0.02	8.14 ± 0.03	7.78 ± 0.06
Crude fiber	2.15 ± 0.03	3.05 ± 0.06	2.91 ± 0.02	2.59 ± 0.07	2.73 ± 0.01	2.81 ± 0.05	2.65 ± 0.19	3.37 ± 0.01	2.54 ± 0.05	2.89 ± 0.19
Crude protein	60.32 ± 0.04	65.18 ± 0.08	68.85 ± 0.23	65.55 ± 0.01	61.47 ± 0.03	63.56 ± 0.06	62.44 ± 0.03	62.75 ± 0.03	67.13 ± 0.06	63.04 ± 0.03
Carbohy Drate	21.94 ± 0.10	14.39 ± 0.01	13.29 ± 0.19	17.18 ± 0.07	20.89 ± 0.09	18.38 ± 0.10	20.21 ± 0.05	18.88 ± 0.09	16.08 ± 0.10	18.30 ± 0.05

From the nutrient content based upon the analysis on proximate as shown in Table 4 it was found that the highest level of the protein and fat on *D. magna* massively cultured using the media of fermentation of 50 g/L of chicken manure + 100 g/L of rejected bread + 50gr / L of tofu waste were at 68.85% ± 0:23. The result of this research was lower than previous research [1] in which on protein it was 73.90% ± 0.04 higher than the results obtained from the previous research reaching at 68.12% [16] and the other research reaching 4% [13] of wet weight. In this research, the highest level of fat was 8.84%± 0, 01 higher than previous research [1] at 7.89% ± 0.02. The high level of protein

content and the low level of fat from the study were due to the high level of nutrient in the culture media of *D. magna* where the nitrate and phosphate levels were getting higher. The higher the N and P content, the higher the protein in the cultivar [17]. The fat content was inversely related to the protein content. The results of this study were supported by the results of previous research that a higher protein content was always conversely comparable to fat because the fat work doubled in the body compared to the protein [18].

The profile of fatty acid of *D. magna* massively cultured using the mix of d rejected bread, tofu waste and different animal manures fermented using the highest probiotic bacteria on the media of 50 g/L of chicken manure + 100 g/L rejected bread + 50 g/L of tofu waste; those are linoleic fatty acid at 6.37% ± 0.02; while the lowest one was found in DHA at 0.06% ± 0.02 in treatment *D. magna* massively cultured using the media of fermentation of 100 g/L of rejected bread + 100 g/L of tofu waste. The profile of the fatty acid of *D. magna* massively cultured using the fermentation of a mixture of different animal manures, rejected bread and tofu waste is presented in Table 5.

Table 5. Profile of fatty acid of *D. magna* in mass culture using the different animal wastes resulted from probiotic bacteria fermentation.

Fatty acids profile (%)	A	B	C	D	E	F	G	H	I	J
Miristic	0,28±0.04	0,49±0,04	0,41±0,09	0,48±0,02	0,01±0,05	0,01±0,05	0,02±0,03	0.65±0.03	0.25±0.05	0.25±0.02
Pentadecanoic	0,10 ± 0,08	0,18±0,06	0,15±0,08	0,17±0,04	0,08±0,02	0,09±0,06	0,04±0,02	0.12±0.02	0.12±0.07	0.02±0.03
Palmitic	2,01 ± 0,06	2,29±0,08	3,59±0,04	1,97±0,08	0,12±0,01	1,14±0,09	0,14±0,06	1.52±0.01	3.52±0.03	2.23±0.01
Stearic	0,41 ± 0,07	1,65±0,02	2.91±0,09	0,52±0,03	0,08±0,05	1,11±0,07	0,10±0,08	0.50±0.037	2.50±0.05	1.75±0.05
Oleic/ω9	1,62 ± 0,05	0,95±0,03	2,61±0,01	0,89±0,08	0,05±0,03	1,07±0,02	0,06±0,07	1.04±0.08	2.04±0.06	0.23±0.08
Linoleic/ω6	0,54 ± 0,02	5,46±0,07	6,37±0,02	4,49±0,07	3,97±0,02	4,73±0,09	3,52±0,01	5.13±0.09	6.13±0.01	4.48±0.04
Linolenic/ω3	0,19 ± 0,06	2,38±0,09	4,32±0,01	3,39±0,03	156±0,07	2,54±0,05	1,90±0,04	2.38±0.02	3.38±0.08	3.45±0.03
Arachidic	0,02 ± 0,07	2,83±0,02	3,05±0,03	1,02±0,04	1.25±0,05	2,30±0,08	1,27±0,05	2.78±0.01	3.26±0.04	2.26±0.05
Arachidonic	0,07 ± 0,01	0,15±0,05	0,13±0,08	0,15±0,02	0,06±0,08	0,07±0,02	0,03±0,09	2.19±0.03	0.17±0.08	0.23±0.01
Eiksapentaenoic	0,27 ± 0,05	2,53±0,09	3,52±0,06	0,50±0,04	0,05±0,02	0,06±0,07	0,01±0,02	1.19±0.05	3.19±0.05	1.19±0.07
Omega 3	0,03 ± 0,02	4,01±0,04	5,91±0,04	3,99±0,08	2,02±0,04	4,06±0,08	3,04±0,08	4.25±0.05	5.25±0.08	3.17±0.02
Omega 6	0,63 ± 0,08	5.64±0,07	7,53±0,08	5,68±0,02	5,03±0,05	6,05±0,02	5,03±0,05	4.08±0.05	6.26±0.01	5.26±0.08
Omega 9	1,62 ± 0,05	0,95±0,01	0,61±0,01	0,89±0,08	0,60±0,03	0,48±0,02	0,63±0,03	0.59±0.06	0.89±0.04	0.29±0.09
Unsattu rated fatty Acid	0,88 ± 0,09	4,46±0,06	3,56±0,07	4,24±0,02	2,68±0,04	3,65±0,08	1,05±0,04	0.68±0.08	2.18±0.09	1.18±0.03
Satturated fatty acid	0,97 ± 0,01	1.09±0,09	2,80±0,08	1,49±0,07	0,97±0,02	1,24±0,02	1,52±0,05	0.14±0.02	1.17±0.01	0.17±0.09
Mono unsaturated fatty acid (MUFA)	1,63 ± 0,052	642±0,02	4,97±0,06	2,40±0,04	3,57±0,05	3,93±0,03	4,67±0,07	2.74±0.08	3.05±0.03	2.07±0.01
Polyunsaturated fatty acid (PUFA)	1,24 ± 0,08	3,82±0,08	4,58±0,04	2,83±0,02	3,17±0,01	4,31±0,09	4,50±0,03	3.65±0.03	4.25±0.05	3.25±0.02
AA	0,08 ± 0,09	0,15±0,04	0,13±0,07	0,15±0,09	2,18±0,03	2,71±0,03	2,89±0,01	2.12±0.02	6.12±0.07	2.02±0.03
DHA	0,06 ± 0,02	0,08±0,04	1,07±0,03	0,07±0,01	0,39±0,08	0,83±0,05	0,77±0,04	0.52±0.01	0.52±0.03	0.23±0.01
EPA	0,27 ± 0,03	1.53±0,02	2,52±0,06	1,50±0,07	0,09±0,02	0,08±0,08	0,11±0,06	0.50±0.037	0.50±0.05	0.75±0.05

The total profile of fatty acid (Table 5) found the most acidic saturated fats (SAFA) on *D. magna* massively cultured using the fermentation media of 50 g/L of chicken manure + 100 g/L of rejected bread+ 50 gr/L of tofu waste included the palmitic fatty acid and unsaturated fatty acids (PUFA) at the linoleic fatty acids. The palmitic fatty acid content was found at highest at 3.59% ± 0.04. The palmitic fatty acids serve as the energy storage for the phytoplankton and zooplankton. This statement is supported by the results of previous research that the saturated palmitic fatty acid has the function for energy storage later used for the biosynthesis process of saturated fatty acids (SAFA) [1], [18]. The palmitic fatty acid is the substrate of biosynthesis process of SAFA fatty acid [1], [19]. The highest linoleic fatty acid was at 6.37% ±0.02 and the results of this study were higher than that of previous research [1] at 0.2%. The linoleic fatty acid serves as a substrate in the formation of long chain of PUFA. The results of this study were confirmed through a statement of previous research that the linoleic fatty acid acts as the base substrate to form a long chain of Omega 6 and Omega 3 [1], [19], [20].

The results of research on the profile of amino acid showed the highest level of D. *magna* massively cultured using the fermentation of 50 g/L of chicken manure + 100 g/L of rejected bread + 50 g/L of tofu waste at the amino acid of non-essential glutamic acid of 31.2% ± 0.02 and the essential lysine amino acids at 22.8% ± 0.03. While, the lowest levels of *D. magna* massively cultured with the media of the fermentation A (100 g/L of rejected bread + 100 g/L of tofu waste) was the non-essential amino acid 2.7% ± 0.06 for arginine and essential amino acids of cysteine at 0.3% ± 0.01. The profile of amino acid of *D. magna* massively cultured using the fermentation of a mixture of different animal manures, rejected bread and tofu waste is presented in Table 6.

Table 6. Profile of amino acid of *D. magna* in mass culture using the different animal wastes resulted from probiotic bacteria fermentation.

Amino Acid Profile	A (%)	B (%)	C (%)	D (%)	E (%)	F (%)	G (%)	H (%)	I (%)	J (%)
Histidine	1.9±0.09	4.6± 0.08	8.0± 0.05	4.6± 0.06	3.8 ± 0,07	6.6± 0.06	2.2 ±0,05	1.18±0.07	1.38±0.08	4.31±0.06
Serine	3.8±0.04	7.9± 0.09	12.1 ± 0.08	6.9± 0.01	4.9 ± 0,06	5.9± 0.01	4.6± 0,09	0.93±0.08	2.97±0.01	2.71±0.09
Arginine	2.7±0.06	7.3± 0.06	10.6 ± 0.06	8.4 ±0.02	3.1 ± 0,05	4.4± 0.03	3.4 ±0,07	6.96±0.03	8.33±0.02	7.99±0.07
Glycine	7.6±0.03	12.2± 0.04	17.60± 0.09	12.0±0.08	11.3 ± 0,04	13.0± 0.08	8.5 ± 0,04	1.66±0.04	2.09±0.23	1.66±0.01
Aspartic Acid	6.1±0.08	14.5± 0.09	16.5± 0.04	16.9±0.04	9.5 ± 0,03	15.9± 0,04	7.8 ± 0,07	1.44±0.10	1.41±0.19	1.08±0.07
Glutamic Acid	15.4±0.09	24.4 ± 0.05	31.2± 0.02	27.9±0.01	19.5 ± 0,01	20.9± 0.01	17.5± 0,01	21.15±0.23	23.44±0.02	21.29±0.10
Threonine	5.4±0.03	8.9± 0.07	15.2± 0.08	8.7± 0.02	6.9 ± 0.04	9.7± 0.02	6.8 ± 0,05	0.16±0.26	1.16±0.03	0.11±0.13
Alanine	14.6±0.06	19.4± 0.06	23.1± 0.03	17.1±0.06	15.5 ± 0.01	18.1± 0.06	17.2±0,08	1.53±0.19	2.62±0.17	1.42±0.09
Proline	5.7±0.04	9.3 ± 0.09	6.8±0.09	8.7 ±0.04	7.1 ± 0.09	9.7± 0.04	6.8 ± 0,09	0.64±0.10	3.63±0.10	0.48±0.08
Cystine	0.3±0.01	0.4 ± 0.05	0.8±0.07	0.3 ±0.05	0.3 ± 0.07	0.4± 0.05	0.4 ± 0,07	0.36±0.03	0.45±0.26	0.29±0.03
Lysine	10.1± 0.09	16.8 ± 0.07	22.8± 0.03	15.± 0.03	14.5± 0.04	17.2± 0.07	12.3±0,09	19.43±0.45	21.46±0.09	17.94±0.4
Tyrosine	3.5±0.08	7.1 ± 0.04	10.9± 0.05	7.0 ±0.08	6.7 ± 0.06	5.7± 0.07	3.5 ± 0,07	6.88±0.19	8.83±0.15	7.48±0.01
Metheonine	1.8±0.05	5.0± 0.08	7.4 ±0.06	5.6 ±0.01	4.2± 0.08	4.7± 0.06	2.5 ± 0,06	6.06±0.03	8.08±0.02	7.79±0.05
Valine	7.7±0.08	13.1 ± 0.05	16.5± 0.04	11.5±0.05	11.5±0.01	13.1± 0.03	9.7 ± 0,03	8.05±0.12	12.53±0.11	11.69±0.0
Isoleucine	6.1±0.05	9.9 ± 0.02	13.33± 0.02	8.9 ±0.03	8.8± 0.02	10.3± 0.08	6.9 ± 0,01	10.62±0.09	12.64±0.17	11.53±0.1
Leucine	6.9±0.09	15.8 ± 0.08	19.2± 0.08	13.8 ±0.06	8.2 ± 0.04	10.8± 0.02	7.9 ± 0,04	8.94±0.01	9.37±0.45	7.88±0.09
Phenylalanine	6.6±0.05	12.8 ± 0.09	17.2± 0.09	13.6 ±0.06	12.1 ± 0.06	11.3 ± 0.03	7.1 ± 0,09	10.49±0.12	13.72±0.07	11.53±0.3
Tryptophan	2.17±0.09	2.5 ± 0.08	2.9±0.05	1.5± 0.01	2.2± 0.02	2.9± 0.02	2.2 ± 0,07	1.23±0.23	2.23±0.15	1.19±0.12

D. magna massively cultured using the media of fermentation of 50 g /L of chicken manure + 100 g/L of rejected bread + 50 g/L of tofu waste also provided the highest profile of amino acids in which for the non-essential amino acids of glutamic amino acid reached 31.2% ± 0:05 which was higher than previous research [1] at 0.76%. Glutamic amino acid acts as the natural component in the mixing in the metabolism and as the building block for proteins. This statement is in line with previous research stating that the glutamic amino acid is a natural component present in every living thing and plays an important role in metabolic processes [1], [21]. *D. magna* massively cultured using the fermentation medium of 50 g/L of chicken manure + 100 g/L of rejected bread + 50gr / L tofu waste provided the highest lysine essential amino acids at 22.8% ± 0.03. This result was higher than that of the previous research [1], reaching at 0:44%. The function of the lysine amino acid was as a building framework of vitamin B1 and anti-virus, helped in calcium absorption, stimulated the appetite, and helped in the production of carnitine to convert fatty acids into energy [1], [21], [22].

D. magna massively cultured using the media fermentation of 50 g/L of chicken manures + 100 g/L of rejected bread + 50gr / L of tofu waste based upon the research results provided an increase to the growth, and the highest biomass production. The nutritional quality was based on the analysis on proximate, profile of amino acid and amino fatty acid on *D. magna* massively cultured using the same media as well.

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