Research Article

Analysis of Proximate Body Composition under the Influence of Different Ammonia Concentration in Confined Condition of *Pangasius pangasius*

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Abstract | Proximate body composition was analyzed to determine the effect of ammonia exposure on various body constituents (water, ash, fat, protein, and organic content) of *P. pangasius*. Oven dried method was used for estimation of water content; muffle furnace for ashing, chloroform: methanol method for fat content and protein content was estimated by the difference in mass of other constituents. Variation in the percentage of water and protein content was observed, and minimum water content and maximum protein content were observed in control (T_0). Regression analysis was done by a relationship of various body constituents with body size (weight and length), all of the constituents showed non-significant except relation with protein content as the control group showed a highly significant correlation ($P<0.001$; $r=0.897$) with percent water indicating inverse relation. While, condition factor showed non-significant correlation ($P>0.05$) with other body constituents except % ash with condition factor in T_4. It is, therefore, concluded that body composition of *P. pangasius* can be estimated directly from water content, body weight or length of fish using predictive regression models developed in this work with a reasonable amount of accuracy.

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Introduction

Fish play an essential role in a country's income, nutrition, employment, and earnings. The composition of fish must be known to make the best use of it. When compared to chicken, buffalo, mutton, and goat meat, fish is safer and healthier to consume. Fish is the best source of high-value protein compared to other protein sources (Louka *et al.*, 2004). Because of current nutritional and medicinal attention on carp fish, it's vital to have a working knowledge of the species proximate analyses. Proximate body composition involves an examination of crude fat, moisture, crude protein, carbohydrate, and ash contents of fish. The moisture percentage in fish muscles is a reliable predictor of their crude protein, relative energy, and fat contents (Cui and Wootton, 1988). When compared to all other animals, fish is more efficient in converting food into body tissues. The reason for their greater food conversion efficiency is ascribed to their reduced dietary energy needs, which is a result of their assimilation of a protein-rich diet. Because of this, fish is an important part of a healthy, well-balanced diet. Fish contains almost all of the essential and required nutrients that humans need. To determine fish's body value, proximate analysis is used to investigate their body composition (Soltan and Tharwat, 2006; Kousar *et al*., 2021). Any edible animal's body composition, including fish, is a vital indication of its functional and biological status. Hence, measuring body composition has been the most important aspect in determining physiological status, it is however a time-taking procedure. Proximate analysis is used to assess a fish's body composition by measuring several constituents such as water content, protein, ash content, fat, organic contents, and fibers (Jakhar *et al*., 2012).

The purpose of the present study is to examine changes in the relative proportions of the body constituents in relation to body weight, length and condition factor with effect of different concentration of ammonia. Predictive equations are developed to describe these relationships in *P. pangasius*.

Materials and Methods

Sample collection and experimental site

50 samples of *Pangasius pangasius* (Length ranges from 24.4 to 34.4cm while weight 103.2 to 343.5g, respectively) were collected from Fish Farm, Al-Raheem fish hatchery, near Murad Abad (Latitude 30°20′0″N; Longitude 71°5′0″E) Muzaffargarh, Punjab, Pakistan. An experimental trial was conducted in Animal house Biopark, Bahauddin Zakariya University, Multan (Latitude 30°16'02.19"N; Longitude 71°30'05.76"E) from 04 September 2019 to 02 December 2019 for 90 days.

Experimental layout

Samples were acclimatized according to the condition in Biopark, Bahauddin Zakariya University, Multan, Pakistan. The experimental trial was divided into five groups (Four ammonia exposure and one control group). Ammonia exposure took place in 50-liter water haler/hapas having 10 fishes in each group. Solution of Ammonia chloride (NH_4Cl) (Sigma) was dissolved in each haler with T_0(Control without ammonia); T_1(0.25mg/L); T_2(0.5mg/L); T_3(0.75mg/L) and T_4(1mg/L). Fishes were fed 2% of their body weight twice a day (8 am and 8 pm). Water in haler was thoroughly exchanged once a week and the same concentration was made in haler. The dried specimen was examined for water, ash, fat, and protein content by covering each specimen in the pre-weight aluminum foil. After covering in the aluminum foil in a ship manner the specimen was kept in the oven at 70 °C to dry the specimen for approximately 15-20 days. The samples were then measured twice until samples became fully dry.

Estimation of water

To assess the percentage of water content in *Pangasius pangasius*, each sample was dried to almost equal weight in an electric oven at 70 °C. The dried fish samples were firmly grinded to make powder in a blender and powder was conserved into plastic boxes to refrain them from the direct air contact the plastic boxes were labeled with marker by assigning them the numerical numbers. The overall amount of water material in fish was measured via the given components:

Percentage of water = Moist mass of body — Dry mass of the body

The proportion of water within the entire fish was calculated via the following method.

% water = Overall amount of H_2O/ Moist body mass × 100

Estimation of ash

Analysis of the ash was achieved at the pattern of powder. The amount of one gram sample was taken from every plastic box and these samples were taken in warmth resilient crucibles. China Crucibles were then kept in an Electric deaden Oven for 24 hours at a temperature of 550 °C to isolate the sample from fuel and combustion including Ashes and other gasses if any. Samples placed in the china crucibles were assessed on an electric digital balance. Fat content percent in all specimens was measured using the given system:

Percentage of ash = Weight of sample (initial) — Loss of weight while heating

Total percentage of ash was calculated via formula.

Total % of wet ash = Overall sum of ash in sample/ Sample Wet Wt. × 100

Total % of dry ash = Overall sum of ash in sample/ Sample Dry Wt. × 100

Estimation of fat

The entire amount of fat contents from the dry tissues of the samples was measured along with the lipids from wet tissues had been mined in a 1:2 aggregate of chloroform and methanol respectively (Salam and Davies, 1994). In this method, 1 gm pattern powder for every fish was poured into the bottles, and mingling up of chloroform and methanol of 25 ml was poured into all flasks. The mixture was centrifuged after keeping in the bottle overnight. The clean solution was eliminated in bottles after taking its weight. Flasks were kept on a hot dish at 40-50 °C for a couple of days. The solvent was evaporated from the bottles leaving the lipid measurements proportionally. Fat content was measured on electric-powered numerical stability at the nearest 0.01g.

The % of lipid in moist and dry body mass of samples was calculated by given recipe.

Amount of lipid = Initial mass of sample – Final mass of sample

Percentage of lipid (moist) = Total lipid in sample/ Moist mass of sample × 100

Percentage of lipid (dry) = Total lipid in sample/ Dry mass of sample × 100

Estimation of protein

The value of protein substance in *Pangasius pangasius* fish was observed by weighing the relationship between body mass and other elements such as water, fat, ash, and others.

The overall quantity of protein in fish could be calculated as follow: Overall content of protein = Dry mass of sample – (Content of ash + Content of lipid)

Percentage of protein was calculated by the given equations.

Of wet protein =Content of protein in given sample/ Mass of sample (wet) × 100

Of dry protein =Content of protein in given sample/ Mass of sample (dry) × 100

Condition factor

To determine the condition factor elements discussed in Fulton's theory are extensively and broadly used in the fish ecology. It is as follows:

Condition factor = Weight/(length)^3 × 100

Statistical analysis

A statistical investigation was consisting of the calculation of correlation coefficients and regression evaluation to know the relation between the different variables. T-test was carried out the standard of assessment and scheming of information was performed with the assistance of the computer program MS-Excel data analysis tool. Correlation coefficients had been taken into consideration at $p<0.001$, $p<0.01$, and $p<0.05$.

Results and Discussion

10 samples from each treatment group and a total of 50 samples were studied to analyze the proximate body composition of *P. pangasius,* under exposure to ammonia in different concentrations. The wet weight of *P. pangasius* was ranged from 110.4 to 221.1 g with a mean value of 155.41g in To, 193.53 to 287.1g with a mean of 238.78 g in T_1, 133.7 to 297.4 with a mean 237.30g in T_2 and 181.2 to 271.4g with mean 224.31g in T_3 and 141.1 to 241.1g with mean 196.72 g in T_4, respectively. Total length was 28.1 to 33.2 cm, 28.4 to 30.2, 25.1 to 32.3, 29.3 to 31.3, and 27.7 to 29.8 cm with means of 31.3cm, 29.29cm, 30.44, 20.45 and 28.9 for T_o, T_1, T_2, T_3, and T_4, respectively. Values for condition factor was 0.80, 0.95, 0.84, 0.79 and 0.81, respectively, for *P. pangasius* under ammonia exposure (Table 1). Analysis of variance among various body constituents in wet and dry weight, where all constituents showed a significant difference (Table 2).

The overall mean percentage of water content in all groups ranged from 73.70 to 80.15% of the body weight of *P. pangasius*. Mean±S.E. values of water content were found 73.70±3.53, 75.62±1.66, 77.31±3.85, 78.69±2.51, and 80.15±2.44 in T_o, T_1, T_2, T_3, and T_4, respectively when exposed with different ammonia concentration. The highest percent value of water content was found in T_4, while the lowest was in T_o. Analysis of variance (ANOVA) study indicated a significant difference ($P<0.05$) among various treatments. A significant change in percent water, when exposed to ammonia concentration (Table 1). The least significant difference in percent water content of *P. pangasius* in different treatments indicated significant differences among all except in T_1 with T_2, and T_2 with T_1. The overall mean percentage of ash content in wet weight in all treatment groups was

Table 1: *Mean values and Ranges of various body constituents of Pangasius pangasius under ammonia exposure.*

Body constituents	Control		Treatment 1		Treatment 2		Treatment 3		Treatment 4	
	Mean ±S.D	**Range**	**Mean± S.D**	**Range**	**Mean ±S.D**	**Range**	**Mean ±S.D**	**Range**	**Mean± S.D**	**Range**
Content of water (%)	73.70± 3.53	68.61 to 77.71	75.62± 1.66	71.74 to 77.32	77.31± 3.85	72.33 to 82.78	78.69± 2.51	75.04 to 82.60	80.15± 2.44	76.02 to 81.0
Ash content (%Wet weight)	1.23± 0.66	0.82 to 2.96	1.21± 0.21	0.87 to 1.60	1.08± 0.26	0.84 to 1.67	1.11± 0.10	1.12 to 1.24	1.01± 0.28	0.68 to 1.59
Ash content (%dry weight)	4.57± 1.92	3.43 to 9.61	4.62± 0.96	3.85 to 7.03	4.85± 1.08	3.21 to 6.08	5.29± 0.94	4.20 to 7.07	5.07± 1.05	4.03 to 7.08
Fat content (%wet weight)	6.41± 1.54	5.22 to 10.34	6.17± 0.94	4.83 to 7.61	5.95± 2.04	4.64 to 10.84	5.46± 0.83	4.49 to 7.07	4.35± 1.46	4.20 to 6.59
Fat content (% dry weight)	24.32± 3.96	20.54 to 33.56	25.26± 3.10	20.03 to 31.05	26.13± 6.25	19.54 to 39.51	25.75± 3.49	20.91 to 32.42	22.14± 7.35	19.8 to 33.2
Protein contents (%wet weight)	18.66± 2.75	15.65 to 23.65	17.08± 1.31	15.46 to 19.44	15.65± 3.19	11.58 to 21.37	14.73± 2.30	11.66 to 18.39	14.48± 2.59	10.7 to 17.9
Protein contents (%dry weight)	71.10± 5.74	56.82 to 75.67	70.11± 3.19	64.09 to 75.30	69.02± 7.14	54.41 to 77.25	68.95± 3.84	62.13 to 74.20	72.78± 7.36	64.49 to 84.02
Organic content (% wet weight)	25.07± 3.18	21.23 to 30.20	23.25± 1.68	28.51to 30.16	21.60± 3.77	16.22 to 26.78	20.19± 2.55	16.16 to 24	18.83± 2.27	17 to 22.6
Organic content (% dry weight)	95.43± 1.92	90.39 to 96.57	95.38 ±0.96	92.97 to 96.15	95.15± 1.08	93.92 to 96.79	94.70± 0.94	92.85 to 96.20	94.92± 1.05	92.92 to 95.97

Table 2: *Analysis of variance among different body constituents in wet and dry weight of Pangasius pangasius.*

		Sum of squares	**Df**	**Mean square**	**F**	**Sig.**
% Water	Between groups	1316.968	4	329.242	226.970	0.00
	Within groups	58.024	40	1.451		
	Total	1374.992	44			
% Ash (WW)	Between groups	12.658	4	3.165	34.023	0.00
	Within groups	3.721	40	.093		
	Total	16.379	44			
% Ash (DW)	Between groups	387.731	4	96.933	65.060	0.00
	Within groups	59.596	40	1.490		
	Total	447.327	44			
% Fat (WW)	Between groups	28.938	4	7.234	20.171	0.00
	Within groups	14.346	40	.359		
	Total	43.284	44			
% Fat (DW)	Between groups	1654.977	4	413.744	68.907	0.00
	Within groups	240.177	40	6.004		
	Total	1895.154	44			
% Protein (WW)	Between groups	1997.210	4	499.303	333.979	0.00
	Within groups	59.800	40	1.495		
	Total	2057.011	44			
% Protein (DW)	Between groups	3625.994	4	906.499	77.293	0.00
	Within groups	469.122	40	11.728		
	Total	4095.116	44			
% Organic content (WW)	Between groups	1566.805	4	391.701	285.472	0.00
	Within groups	54.885	40	1.372		
	Total	1621.690	44			
% Organic content (DW)	Between groups	387.731	4	96.933	65.060	0.00
	Within groups	59.596	40	1.490		
	Total	447.327	44			

ranged from 1.01 to 1.23, while 4.20 to 5.29 in percent ash dry weight respectively. The maximum value of ash content in wet weight was found in T_1 (1.23±0.66), while the minimum value in T_4 (1.01±0.28). As in percent ash dry weight, the mean value was found maximum in T_3 and minimum in the T_1 group. Analysis of variance (ANOVA) study indicated a significant difference (P<0.05) among various treatments. A significant change in percent Ash (WW and DW), when exposed to the ammonia concentration. The least significant difference for percent ash content (WW) of *P. pangasius* in different treatments indicated significant difference among all except in T_o with T_1 and T_2 with T_1. Percent fat content in wet weight in all treatment groups was ranged from 4.35 to 6.41, while in percent dry weight ranged from 22.14 to 26.13, respectively. The maximum value of percent fat content in wet weight was found in T_0, while the minimum value in T_4. As in percent fat dry weight, the mean value was found maximum in T_2 and minimum in T_4. Analysis of variance (ANOVA) study indicated a significant difference (P<0.05) among various treatments. A significant change in percent fat (WW and DW), when exposed to the ammonia concentration. The least significant difference for percent fat content (WW) of *P. pangasius* in different treatments indicated significant difference among all except in Control with T_1; T_2 with T_1 and T_3 with T_2. Percent protein content in wet and dry weight was found to be maximum in control group with a mean of 18.66±2.75 and 71.10±5.74, respectively. The mean value of percent protein in wet and dry weight was found minimum in the T4 group. Analysis of variance (ANOVA) study indicated a significant difference (P<0.05) among various treatments. A significant change in percent protein (WW and DW), when exposed to the ammonia concentration. The least significant difference for percent ash content (WW) of *P. pangasius* in different treatments indicated significant relationship.

Statistical analyses of percentage water and body constituents in wet and dry weight of *Pangasius pangasius* under different treatments and control condition, respectively. Percent water showed a non-significant correlation with percent ash wet and dry weight in all treatments. Percent fat (WW and DW) showed a non-significant correlation with percent water except percent water with fat wet weight in the control condition that showed a significant relation (r=0.711). Percent protein (WW) showed a non-significant correlation with percent water, while percent protein (DW) with percent water showed partial significant relation with T2 (r=0.686) and T3 (r=0.739); significant relation with T1 (r=0.832) and T4 (r=0.778); highly significant correlation in the control condition. Percent organic content (WW) showed a non-significant correlation with percent water, while percent organic content (DW) with percent water showed a highly significant correlation with all treatments except T4 which showed a non-significant correlation in *P. pangasius* (Table 3).

Regression analysis of body weight showed a non-significant (P>0.05) correlation with percent water in all treatments. Percent ash (WW and DW) showed a non-significant correlation with body weight in all treatments except T_2 which showed a partial significant correlation and highly significant in T_4. Percent fat (WW) showed a non-significant correlation in control and T_1, and a significant correlation with T_2, T_3, and T_4. In fat percent dry weight non-significant correlation with Control and T1; least significant with T2; significant with T_4 and highly significant in T_3. Percent protein (WW) showed a non-significant correlation in T_1, T_2, and T_4, and a significant correlation with To and T_3. In protein percent dry weight non-significant correlation with Control and T_1; partial significance with T_3; significant with T_2 and T_4. Percent organic content showed a non-significant correlation in all treatments, while percent organic content in dry weight showed the least signification with T_2 and a highly significant relation with T4 (Table 4). Total length showed a non-significant correlation with percent water. Ash wet weight showed non-significant relation with all treatment T_4, similarly in dry ash weight, all treatments showed non-signification except T_2 and T_4. Fat wet weight showed non-significant relation in all treatments except T_2 and T_4 that showed a significant correlation, as in dry weight highly significant correlation was observed in T_2 and T4. Percent protein showed a significant correlation with all except control. In dry condition significant with T_2 and highly significant with T_4. Organic content showed non-significant in all treatments except T_2 and T_4 (DW) which showed the least significance (Tables 4, 5). A study of regression analysis among various body constituents (Percent water, ash, fat, protein, and Organic content) with condition factor in wet and dry weight showed a non-significant correlation, except relation with percent ash that showed a significant relation in T4 (r= 0.865) as shown in Table 6.

Table 3: *Analysis of variance among different body constituents in wet and dry weight of Pangasius pangasius.*

		Sum of squares	Df	Mean square	F	Sig.
% water	Between groups	1316.968	4	329.242	226.970	0.00
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	Total	1374.992	44			
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	Total	1621.690	44			
% Organic content (DW)	Between groups	387.731	4	96.933	65.060	0.00
	Within groups	59.596	40	1.490		
	Total	447.327	44			

Body composition is predicted using % water and its correlations with other body constituents (protein, ash, and fat). It can be assessed even without the time, cost, or expertise required in most lab and biological fields (Hartman and Margraf, 2008). Microwave techniques to assess water content in fish were used for analysis by Crossin and Hinch (2005), by measuring water content, the non-significant relation between fat and water and fat content for a rapid and cost-effective index to the content of relative energy (Hislop *et al.*, 1991). As in our study % water showed a non-significant correlation with fat except for control in wet weight.

Various researchers have reported an inverse relation between % fat and protein and percent water in whole fish (Shearer, 1994; Osibona *et al.*, 2009). Because many studies have established prediction equations, the current study indicated that the body composition of fish may be determined using regression models.

Ash is only a minor part of the fish body constitution. In *Pangasius pangasius*, the ash value in wet weight ranged from 1.01 to 1.23, while 4.20 to 5.29 in dry weight. The percentage of ash is in agreement with the study percentage of ash 1.22% in *C. idella* (Scherer *et al.*, 2006), 1.21% in *P. fluviatilis* (Orban *et al.*, 2007), 1.2% in *C. gariepinus* and and *T. zillii* (Osibona *et al.*, 2009). Far from this finding Ali *et al.* (2001) found 4.3% ash in *C. punctata*, Kamal *et al.* (2007) found 3.7% in *C. batrachus*, 3.1% in *H. fossilis*, and 3.1% in *A. testudineus* and 3.3% in *C. punctata* show deviation in ash wet weight. This data is consistent with O'Connor *et al.* (1981), who showed both increasing and decreasing trend in ash content with increase in body weight of fish (Gunther *et al.*, 2005).

Table 4: *Statistical analysis of body weight with various body constituents of P. pangasius.*

Constituents	Treatments	r	a	b	SE(b)	t-value (b=0)
Body weight vs Percent water	Control	0.598 ns	58.88462	0.01603	0.008118	1.974624
	Treatment 1	0.044 ns	69.72597	0.000961	0.008152	0.117885
	Treatment 2	0.318 ns	69.72681	0.004444	0.005007	0.887557
	Treatment 3	0.502 ns	76.11983	-0.0101	0.006561	-1.5394
	Treatment 4	0.061 ns	78.62263	0.004132	0.025178	0.164112
Body weight vs Percent ash wet weight	Control	0.413 ns	0.852119	-0.00114	0.000946	-1.20507
	Treatment 1	0.065 ns	0.772946	-0.00047	0.002701	-0.17401
	Treatment 2	0.707*	2.002407	-0.00452	0.00171	-2.64327
	Treatment 3	0.0005 ns	1.528735	3.89E-06	0.002804	0.001387
	Treatment 4	0.982***	5.169009	-0.01635	0.001176	-13.9031
Body weight vs Percent ash dry weight	Control	0.353 ns	2.138301	-0.00244	0.002447	-0.99714
	Treatment 1	0.062 ns	2.533191	-0.00143	0.008567	-0.16692
	Treatment 2	0.704*	6.663301	-0.01472	0.005604	-2.6267
	Treatment 3	0.078 ns	6.331534	-0.00215	0.010275	-0.20925
	Treatment 4	0.936***	23.46999	-0.07082	0.010027	-7.06293
Body weight vs Percent fat wet weight	Control	0.101 ns	2.64656	0.0012	0.004446	0.269906
	Treatment 1	0.091 ns	2.998499	0.000742	0.003056	0.242801
	Treatment 2	0.772**	4.481535	-0.00406	0.00126	-3.22222
	Treatment 3	0.868**	8.34975	-0.01919	0.004137	-4.63863
	Treatment 4	0.851**	10.39952	-0.02645	0.006145	-4.30431
Body weight vs Percent fat dry weight	Control	0.255 ns	6.115711	0.007186	0.010272	0.699572
	Treatment 1	0.119 ns	9.885084	0.002855	0.008999	0.317257
	Treatment 2	0.725*	14.84239	-0.01181	0.004227	-2.79394
	Treatment 3	0.882***	33.0666	-0.07831	0.015779	-4.96293
	Treatment 4	0.788**	48.59651	-0.11812	0.034873	-3.38715
Body Weight vs Percent protein wet weight	Control	0.836**	37.6167	-0.01609	0.00398	-4.04271
	Treatment 1	0.074 ns	26.50258	-0.00123	0.006216	-0.19788
	Treatment 2	0.323 ns	23.78925	0.004137	0.004579	0.903472
	Treatment 3	0.778**	14.00169	0.029284	0.00892	3.28296
	Treatment 4	0.539 ns	5.808844	0.038674	0.022789	1.697047
Body weight vs Percent protein dry weight	Control	0.160 ns	91.7459	-0.00474	0.011021	-0.43009
	Treatment 1	0.041 ns	87.58172	-0.00143	0.013046	-0.10961
	Treatment 2	0.807**	78.49431	0.026527	0.007329	3.619457
	Treatment 3	0.763*	60.60186	0.080462	0.025694	3.131548
	Treatment 4	0.859**	27.93351	0.188943	0.042381	4.458201
Body weight vs Percent organic contents wet weight	Control	0.577 ns	40.26326	-0.01489	0.007959	-1.87084
	Treatment 1	0.025 ns	29.50108	-0.00049	0.007124	-0.06878
	Treatment 2	0.006 ns	28.27078	7.87E-05	0.004605	0.01709
	Treatment 3	0.505 ns	22.35144	0.010096	0.006519	1.548704
	Treatment 4	0.183 ns	16.20836	0.012222	0.024712	0.494578
Body weight vs Percent organic contents dry weight	Control	0.353 ns	97.8617	0.002444	0.002447	0.998774
	Treatment 1	0.062 ns	97.46681	0.001429	0.008567	0.166803
	Treatment 2	0.704*	93.3367	0.014722	0.005604	2.627052
	Treatment 3	0.078 ns	93.66847	0.002148	0.010275	0.209051
	Treatment 4	0.936***	76.53001	0.070819	0.010027	7.06283

Table 5: *Statistical analysis of total length with various body constituents of P. pangasius.*

Constituents	Treatments	R	a	b	SE(b)	t-value (b=0)
Total body length vs Water percent	Control	0.419 ns	54.07285	0.280064	0.228872	1.223671
	Treatment 1	0.321 ns	78.26305	-0.28364	0.31626	-0.89686
	Treatment 2	0.041 ns	71.1482	-0.01205	0.108934	-0.11062
	Treatment 3	0.615 ns	88.95485	-0.496	0.240037	-2.06635
	Treatment 4	0.199 ns	95.18183	-0.54486	1.012311	-0.53823
Total body length vs Percent ash wet weight	Control	0.126 ns	0.843026	-0.00866	0.02565	-0.33762
	Treatment 1	0.053 ns	1.121295	-0.01574	0.110614	-0.1423
	Treatment 2	0.627 ns	3.45062	-0.08282	0.038882	-2.13003
	Treatment 3	0.017 ns	1.375489	0.005062	0.11248	0.045004
	Treatment 4	0.641*	14.59836	-0.43759	0.197573	-2.21483
Total body length vs Percent ash dry weight	Control	0.071 ns	1.922149	-0.01236	0.06495	-0.1903
	Treatment 1	0.072 ns	4.153692	-0.06697	0.35035	-0.19115
	Treatment 2	0.644*	11.63791	-0.27815	0.124622	-2.23195
	Treatment 3	0.075 ns	8.37017	-0.08279	0.41235	-0.20078
	Treatment 4	0.758*	77.41285	-2.3486	0.763017	-3.07804
Total body length vs Percent fat wet weight	Control	0.131 ns	1.728322	0.038793	0.110271	0.351797
	Treatment 1	0.451 ns	-1.2196	0.150069	0.112062	1.33916
	Treatment 2	0.792 **	6.134871	-0.08594	0.024988	-3.43925
	Treatment 3	0.541 ns	18.64684	-0.4796	0.281703	-1.7025
	Treatment 4	0.801**	34.63321	-1.0186	0.287658	-3.54101
Total body length vs Percent fat dry weight	Control	0.251 ns	2.370323	0.176292	0.255954	0.688764
	Treatment 1	0.410 ns	-1.24588	0.403313	0.338135	1.192757
	Treatment 2	0.873***	20.96837	-0.29323	0.061792	-4.74544
	Treatment 3	0.595 ns	80.08622	-2.12144	1.080858	-1.96274
	Treatment 4	0.932***	190.8121	-5.72502	0.836742	-6.84204
Total body length vs Percent protein wet weight	Control	0.647*	43.3558	-0.3102	0.137828	-2.25063
	Treatment 1	0.221 ns	21.83526	0.14931	0.248765	0.600205
	Treatment 2	0.684*	19.26631	0.180806	0.072855	2.481724
	Treatment 3	0.643*	-8.97718	0.970544	0.436725	2.222323
	Treatment 4	0.682*	-44.4134	2.00105	0.810698	2.468305
Total body length vs Percent protein dry weight	Control	0.222 ns	95.70753	-0.16394	0.270967	-0.60502
	Treatment 1	0.237 ns	97.09219	-0.33634	0.519007	-0.64805
	Treatment 2	0.842**	67.39372	0.571384	0.138229	4.133604
	Treatment 3	0.521 ns	11.54361	2.204234	1.362882	1.617333
	Treatment 4	0.897***	-168.225	8.073625	1.501055	5.378634
Total body length vs Percent Organic contents wet weight	Control	0.422 ns	45.08413	-0.2714	0.21992	-1.23409
	Treatment 1	0.388 ns	20.61566	0.299379	0.268766	1.113902
	Treatment 2	0.377 ns	25.40118	0.09487	0.08805	1.077456
	Treatment 3	0.612 ns	9.669658	0.490942	0.239607	2.048947
	Treatment 4	0.360 ns	-9.78019	0.982451	0.960225	1.023147
Total body length vs Percent Organic contents dry weight	Control	0.071 ns	98.07785	0.012356	0.06495	0.190239
	Treatment 1	0.072 ns	95.84631	0.066974	0.35035	0.191163
	Treatment 2	0.644 *	88.36209	0.27815	0.124622	2.231949
	Treatment 3	0.075 ns	91.62983	0.082791	0.41235	0.200778
	Treatment 4	0.758*	22.58715	2.348601	0.763017	3.078045

Table 6: *Statistical analysis of condition factor with various body constituents of P. pangasius.*

Constituents	Treatments	r	a	b	SE(b)	t-value (b=0)
Condition factor (x) vs Percent water (y)	Control	0.256 ns	60.04589	3.492148	4.965992	0.703213
	Treatment 1	0.378 ns	67.2085	2.898014	2.681217	1.080858
	Treatment 2	0.532 ns	68.59712	2.600074	1.563676	1.662796
	Treatment 3	0.185 ns	74.87367	-1.28419	2.57618	-0.49849
	Treatment 4	0.230 ns	75.48219	4.866932	7.756997	0.627425
Condition factor (x) vs Percent ash (y)	Control	0.448 ns	1.072105	-0.62542	0.470923	-1.32807
	Treatment 1	0.110 ns	0.927754	-0.28203	0.954842	-0.29537
	Treatment 2	0.147 ns	1.206332	-0.32992	0.836294	-0.3945
	Treatment 3	0.011 ns	1.506069	0.029654	0.968214	0.030628
	Treatment 4	0.865**	5.650687	-4.55364	0.995777	-4.57295
Condition factor (x) vs Percent fat (y)	Control	0.028 ns	3.078625	-0.17013	2.265879	-0.07508
	Treatment 1	0.275 ns	3.92917	-0.79484	1.047116	-0.75908
	Treatment 2	0.004 ns	3.511133	0.008897	0.69416	0.012817
	Treatment 3	0.664 ns	8.069622	-5.0693	2.154605	-2.35277
	Treatment 4	0.596 ns	9.946104	-5.8483	2.978074	-1.96379
Condition factor (x) vs Percent Proteins (y)	Control	0.276 ns	35.80338	-2.69659	3.54355	-0.76099
	Treatment 1	0.311 ns	27.93458	-1.82114	2.10263	-0.86612
	Treatment 2	0.509 ns	26.68541	-2.27905	1.456353	-1.5649
	Treatment 3	0.486 ns	15.55064	6.323832	4.287895	1.47481
	Treatment 4	0.244 ns	8.921021	5.535	8.296413	0.667156
Condition factor (x) vs Percent organic content (y)	Control	0.219 ns	38.882	-2.86672	4.824689	-0.59418
	Treatment 1	0.390 ns	31.86375	-2.61599	2.32799	-1.12371
	Treatment 2	0.532 ns	30.19654	-2.27016	1.362785	-1.66582
	Treatment 3	0.181 ns	23.62026	1.254536	2.565016	0.489095
	Treatment 4	0.014 ns	18.86713	-0.3133	7.944067	-0.03944

Fat wet value in *P. pangasius* ranged from 4.35 to 6.41, while in percent dry weight ranged from 22.14 to 26.13. According to Hossain *et al.* (1999), the fat content of various species ranges from 1.87-9.55%. Although Gunther *et al.* (2005) found % fat in *S. namaycush*, the current values were within the range. Salam (2002) reported the high content of fat in *H. fossilis* (3.2%), while Mazumder *et al.* (2008) assessed *P. chola* (3% percent), *A. coila* (3.5%), *P. atherinoides* (2.2%), and *C. nama* (2.2%). Fish having a lipid content of less than 5% are termed lean (Ackman, 1989). Thus, fish such as *Pangasius pangasius* is good sources of high protein and low-fat content.

Fish are categorized into different groups, the high fat group (> 8%), medium fat group (4% to 8%), low-fat group (2% to 4%), and lean group (<2%) in wet body weight. The % fat content of catfish demonstrated a positive relationship with the total length and wet weight of fish. So, as a general rule, the content of fat is increased with size, as a result decreases in content of water. Each fish have a specific fat range, maximum in adult and minimum in the smaller size of fish (Peter, 1979). Fat content increases with fish development rate, according to Kalay *et al.* (2008). The proportion of fat tissue varies with age, with older animals having a higher proportion than younger animals (Degani, 1988).

Protein content in *P. pangasius* ranged from 18.66±2.75 and 71.10±5.74 respectively in the wet weight of fish. Contrary to the previous result different researchers reported differences in values of protein content. As Kamal *et al.* (2007) reported 15.6% in *M. vittatus* and 19% in *C. catla* by Ali *et al.* (2005). It indicates that tested fish are rich in protein since Stansby (1976) defined protein content as higher than 15%. So more than 15% value of protein content is reported in the present study. Exposure to ammonia affects the protein content, as ammonia cause decreases in the protein content. Weatherley and Gill (1987), protein concentrations had consistently strong correlations with fish body weight, hence it was logical to believe that estimations of body components versus body weight would be reliable.

The nutritional content of fish varies with species, body size (weight and length), and condition factor, according to studies on *A. nobilis* (Naeem and Salam, 2010) and *M. bleekeri* (Naeem and Ishtiaq, 2011) and other specimens (Yousaf *et al*., 2011; Ashraf *et al*., 2011; Naeem *et al*., 2016). The current research reveals a strong inverse relationship between water and fat content in *C. catla*, revealing that fat content in the fish body reduces as water level increases. Other research (Naeem and Salam, 2010; Naeem and Ishtiaq, 2011) shows comparable results, which support these results. Fish body composition is related to changes in growth (Shearer *et al*., 1994), length and weight (Naeem *et al*., 2010a, 2011a; Pervaiz *et al*., 2012), nutrition (Jobling and Miglavs, 1993; Raza *et al*., 2013; Ismat *et al*., 2013), reproduction in fish (Thorpe *et al*., 1998; Naeem *et al*., 2005, 2011b), and survival rate (Sogard and Olla, 2000) and quantity of elements (Naeem *et al*., 2010b). So, in the present study variation in results is mainly due to many factors that affect its body composition.

Conclusions and Recommendations

Body composition consists of water protein, ash, fat, and organic content. Water content increases with an increased concentration of ammonia, while reduction in protein content, while ash and fat contents showed a similar trend in the analysis. Deposition of water content results in the weight loss as observed in the growth pattern (weight and length gain).

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Novelty Statement

This study provides valuable insights into the nutritional responses of this species to ammonia-induced stress, aiding in better management practices.

Author's Contribution

Shoaib Hassan: Performed experiment, did statistical analysis and wrote manuscript.
Muhammad Naeem: Supervised the research work and reviewed the manuscript.

Conflict of interest
The authors have declared no conflict of interest.

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