UTTAR PRADESH JOURNAL OF ZOOLOGY

42(6): 81-89, 2021
ISSN: 0256-971X (P)

NUTRITIONAL AND BIOLOGICAL ASSESSMENT OF NATIVE VERSUS BROILER CHICKENS AND ESTIMATING OF TRI IODO THYRONINE IN CHICKEN MUSCLE GC MS METHODS

J. MERLIN SELES[1*], R. KALIDOSS[1], K. ARUNPRASATH[1], M. MARIRAJ[1], M. SHENBAGAM[1], N. RAJAPRABHU[1] AND P. PONMURUGAN[1]

[1]Biomedical Research Laboratory, Department of Botany, Bharathiar University, Coimbatore – 641 046, Tamil Nadu, India.

AUTHORS' CONTRIBUTIONS

This work was carried out in collaboration among all authors. Author JMS designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors PP and RK managed the analyses of the study. Author RK managed the literature searches. All authors read and approved the final manuscript.

Article Information

Editor(s):
(1) Dr. Ana Cláudia Correia Coelho, University of Trás-os-Montes and Alto Douro, Portugal.
Reviewers:
(1) Raafat Khidr, Desert Research Center, Egypt.
(2) Oday Satar Abbas, Ibn Sina University of Medical and Pharmaceutical Sciences, Iraq.
(3) Karwan Yaseen Kareem, Salahaddin University, Iraq.

Received: 01 October 2020
Accepted: 03 December 2020
Published: 30 March 2021

Case Study

ABSTRACT

It is well known that the market price has been increased according to the increasing of population density which followed by the increasing in demand for chicken meat. However, researchers have shown that T3 growth hormone injection to broiler chicken to create a problem in human health hazard and reduction in nutritional value. The main objectives were to compare the nutritional and biochemical assessment in meat of native versus broiler chicken. To find out the nutritional value reading were carried out by spectrophotometry method.GCMS method is used to detect the T3 hormone (tri iodo thyronine) in chicken muscle.To find out the GCMS result prominent peaks were identified .Statistical analysis was carried out by using software IBM SPSS statistics 20 packages The results imply that broiler chicken extract was detected with T3 hormone needed to alert the society to avoid consume them.

Keywords: Triiodo thyronine; nutrition; biochemical minerals; native chicken; broiler chicken.

1. INTRODUCTION

Poultry is a domesticated bird to provide meat to human and plays an important role in the agricultural sector. Due to unemployment is national problem for educated youths and rural uneducated women, knowledge of poultry rearing to generate incomes is an important to an understanding of the role of poultry

**Corresponding author: Email: merlinseles@gmail.com;*

in rural areas. The importance of poultry export in 2000 is known with an estimate of 8.79 million tons, but double the tons of export in 2014 probably was much more significance than other products of livestock or meat products Davis, [1]. Earlier human being consumed native chicken raised from indigenous country breed and withstand extremes of climatic factors. Because of the comparatively introduction of growth hormone and antibiotic injection for early maturity in broiler chicken, girls are more susceptible to early maturity, human are less immune to drug resistant microbial pathogen. Probably, more faster growth in the broiler chicken, but not in the native chicken, more beneficial to growers. In particular, the consumer's price per Kg of broiler chicken is cheap and because of its disease tolerance, the consumers are highly dependent on broiler chicken marketing. The relative rates of growth and feed intake efficiency and increased its conversion into muscle tissue are dependent on growth hormone injection in broiler chicken. Consequentlly, many farmers concluded that broiler chicken and egg productions are advantageous due to its quick returns and higher weight returns [2,3]. In Namakkal, probably it is of more than 200 small growers and 150 entrepreneurs are indulging in broiler chicken production.

The efficacy of drugs to promote disease free marketed animals is well documented in the literature and demonstrated with proof in some in vivo experiments (Anderson et al., 1997) relating to health hazards to human. The remark of Hadded and Mashaly, (1991) suggested that all chicken meat are not completely "hormone free" and it is possible that manipulating growth hormones is of high risk of early induction of menstrual cycles in females.

At this stage it is necessary to detect growth hormone triiodothryorine in chicken meat. Growth hormone triiodothryronine T3 regulates metabolism of early maturity. Because evaluation of the role of T3 hormone increase health hazards of human has been based largely on medical observations, the interpretations of our researchers have tended to be influenced by the comparative assessment of physical and chemical parameters of native Vs Broiler chicken meat and T3 hormone detection in such meat using GCMS analysis [4]. The main objective of present study was to analyse the differences of native versus broiler chicken meat muscles collected from local farms and shops on biometric parameters and T3 hormone residue analysis using GC - MS spectrum.

2. MATERIALS AND METHODS

Four chicken samples for analysis were collected for nutritional status, physical and chemical parameters characteristic as a comparative study of native versus broiler chicken. The first sample was reported as Valar fresh chicken. The second sample of chicken meat was Suguna and third was Shanthi and these all were chicken samples of a broiler type. Pure native breed obtained was subjected to remain stored at - 20 C along with other three broiler chicken samples in refrigeration.

2.1 Nutritional Measurements

The collected meat samples were subjected to complete analysis (moisture, crude protein, crude fibre, ether extract, total ash, calcium, phosphorus, Potassium, Magnesium, Manganese, Zinc, Iron, Ash, Fibrous proteins and Myosin proteins) as per A.O.A.C. (2003). Moisture and crude fat content were measured using the (AOAC, 1990); (AOAC Official Method PVM-1:2003 MEAT). Total fatty acid composition was determined through extraction using the procedure followed by Sukhija and Palmquist (1988) from the pulverized samples.

2.2 Determination of Moisture

The hot air oven drying method has been suggested as a method for examination of moisture content of broiler versus native chicken meat samples. Well dried crucible appeared clean greese free was weighed with sample weight of 50 g and marked W1. The crucible was left undisturbed in an oven at a temperature of 135+-2 degree Celsius for 2 hours. Then the crucible was placed in the desiccator for 30 minutes. The weight of crucible (W2) was weighed along with the oven dried sample.

The percent moisture was calculated by following formula

$$\text{Moisture (\%)} = \frac{\text{W1} - \text{W2}}{\text{Weight of sample}} \text{x } 100$$

Where,

W1-Initial weight of crucible+ sample,
W2-Final weight of crucible+ sample
Dry matter (%)=100-moisture(%).

2.3 Determination of Fat

The commonly used procedure is a Soxhlet extraction method. Approximately, five gram sample kept oven dried was used and kept them in fat free thimble. The meat fat content was extracted by siphoning petroleum ether. The ether was allowed to evaporate, leaving the extracted fat and transferred in a petri dish. The petri dish was removed and oven dried

extract at 103°C for 30 min was cooled in desiccators. The result of crude fat was determined using the following formula

$$\text{Crude fat}(\%) = \frac{\text{weight of ether extract}}{\text{Weight of sample}} \text{x } 100$$

The test for total lipid content was carried out with the procedure described by Spiricet et al., (2010).

2.4 Determining of Ash

The ash products obtained in this test were carried out by approximate amount (5 gm) of dried chicken meat sample which was left it in muffle furnace at 600°C for an hour. The empty crucible weight was denoted as W1. The ash obtained after the procedure was denoted as W3. The weight of sample was represented as W2. The following formula used to calculate % ash.

$$\text{Percent ash} = \frac{\text{Difference in weight of ash}}{\text{Weight of sample}} \text{x } 100$$

Difference in weight of Ash = W3-W1.

2.5 Determining of Protein

Tests with sulphuric acid digestion of chicken meat sample to determine crude protein content was followed by Kjeldhal method. The tests depend on distillation of 5 mL of digested sample in Kjeltec system. The distillation tube received phenolphthalein indicator in drops and 40% sodium hydroxide of 10 mL. Characteristic ammonia was formed and collected into sulphuric acid containing conical flask. The ammonia was titrated against 0.1N sodium hydroxide. The amount of non reactive or non utilized acid or non neutralized sulphuric acid was calculated by the following formula

Per cent Crude Protein = 6.25* x % N

$$\% \text{ N } = \frac{(\text{S } - \text{ B}) \text{ x N x } 0.014 \text{ x D}}{\text{Weight of the sample x V}} \text{x } 100$$

Where S - Sample titration reading, B - Blank titration reading, N - Normality of Na OH D - Dilution of sample after digestion, V - Volume taken for distillation

2.6 Determination of Reducing Sugar Using DNS Method

A solution of 80% hot ethanol (50°C) was used as a solution for extraction of sugars in chicken meat samples. The supernatant was collected from centrifuge tube seperated by centrifugation of meat extract at 1130 x g for 10 minutes. After centrifucation of the extract, the impurities of certain pellets were removed by filtration using Whatman filter paper No. 1, leaving a crystal clear solution of filtrate in a test tube. The tube was left to air dry. The dried extract was redissolved indistilled water (2 mL) and centrifuged at 10,000 x g for 10 minutes. Finally, 1mL of this extract was subjected to react with 2 mL of DNS (0.5 g DNS, 8 gm sodium hydroxide, and 150 gm Rochelle salt in 500 mL distilled water) for 10 minutes. The absorbance was recorded in Spectrophotometer at 550 nm. The concentration of sugar in chicken extract was calculated from mg/mL of glucose solution prepared as standard. Korea Society of food Science and Nutrition (2000) procedure of reducing sugar test was preferred.

2.7 Determination of Non Reducing Sugar

2.7.1 Determination of pH

Test for pH was determined using an L1 120 meter (Elico, Geramany) fitted with a combined electrode, calibrated at pH 4.0 and 7.0 with standard buffer stored at room temperature (25°C). The procedure was carried out in three measurements and finally the average pH value was recorded.

2.7.2 Extraction and elution of triiodothyronine growth hormone residues

Triiodothyronine growth hormone residues was Soxhlet extracted from chicken muscles and eggs along with additive, anhydrous sodium sulphate (25.5 g) using hexane (250ml) followed by shaking for two hours on a mechanical shaker Haddad and Mashaly, [5]. The filtrate was concentrated to near dryness in a rotary vacuum evaporator. Co-extractives in the concentrated residues was removed using adsorption column chromatography, where five percent deactivated neutral alumina (25 g) served as adsorbent while hexane: diethyl ether (90:10 v/v) were used as eluting solvent Schenck and Donoghue, [6].

2.7.3 Analysis of triiodothyronine growth hormone residues

Concentrated triiodothyronine growth hormone pesticide residue was diluted with suitable ratio of hexane/acetone. Each sample was injected three times, besides the control and standards. Analysis was carried out using a Gas Chromatography equipped with Mass Spectroscopy in which nitrogen phosphorous detector (NPD and Ni-63 electron capture detector (ECD) are fitted. Residues identified by comparison of retention time with authentic standards and were quantified with point scan method

of known concentration of standard based on peak area. Samples containing pesticide residues were spiked with known amount of respective standards (Hadded and Mashaly, 1991, Shaikh et al. [7].

Observed values obtained from the result was compared with actual values be means of MRL values fixed by the World health Organization (WHO) and Food Adulteration Act, FAA as per the procedure prescribed by Lehotay et al. [8].

Statistical analyses were provided by using SPSS software packages.

3. RESULTS AND DISCUSSION

The native chicken meat biochemical parameters showed significant nutritive values compared to broiler chicken meat. The main reasons behind maximum nutritive value of native chicken could be due to differences in moisture content and other features. The color of Native was darker (Thick Red) and more yellow than Broiler chicken. The meat color was proportionate to the content of myoglobin that increases with age Jaturasitha, et al. [9] and breed Fletcher, [10] and hence it would make it darker.

The result findings of mean average major and minor biochemical parameters of native chicken showed higher nutritive value than broiler chicken.

3.1 Determination of Moisture

The availability of moisture content was found to be significant in chicken meat at any stage of nutritive analysis. The chicken moisture content was carried out in triplicates. It was found that the native chicken showed meats with lower moisture content as compared to other broiler chicken meats. The native chicken showed lowest meat 54.16% and maximum of 59.52% (Shanthi) and the moderate moisture content measured 58.10% (Valar); 58.64 (Suguna) (Fig. 1). The result showed also that broiler chicken showed maximum moisture content of 59.52 and minimum of 58.10% (Valar). This explains the reason for why the broiler chicken weighs heavier but poorer the nutritive value than lesser the weight and richer the nutritive value of native chicken. The results of the present study for this moisture content are not in agreement with the findings of Ezhil Vallavan et al. [11] who showed that the moisture content of native chicken's breast meat was 72.31% versus 73.12% in broiler chicken meat. They also found that the comparative assessment of moisture content of native chicken's thigh meat was 72.65% 70.94% in broiler chicken which showed that the native chicken moisture content of thigh meat was higher when compared with breast meat. Boni et al. [12] interpreted the differences in the moisture content of chicken meat according to its age. He pointed out that higher the age of bird lower the moisture content of chicken meat.

3.2 Determination of Protein Content

The maximum percentage of protein content of chicken meat differed significantly with the native versus broiler chicken meats. The result findings of protein content of native chicken meat showed differences to the greater extent of 15.5% versus 12.7%, 15.5, and 12.0% of valar, suguna and shanthi chicken meat respectively. The results of the present study represented that the native chicken protein content (15.5 mg/g. dry wt.) is comparable to Suguna chicken (145.31 mg/g) (Table 1); Similar study conducted by Ezhil Valavan et al. [11] who showed that the protein content of native chicken's breast meat was 23.38 ±0.07 and 23.69±0.54 of broiler chicken. But, the protein content of native chicken's thigh meat (mg/g) showed 19.41±0.70 and 21.93±0.35 of broiler chicken.

According to the present study, native chicken has protein content of 15.5% but is less nutritious than the findings of Ezhil vallavan et al. [11] who reported that the protein content of native chicken breast meat was 23.38±0.07.

The total nitrogen content obtained in the present study for broiler chicken such as Valar, Suguna and Shanthi was 14.5; 16.7 and 13.5 respectively. The total nitrogen content obtained for a native chicken was 18.5% and differences could be attributed to variation in the breed, feed, age at slaughter, the system of production, sex, processing, and the part of the cut as suggested by Haunshi et al. [13].

3.3 Determination of Carbohydrates

The mean average reducing sugar content varied significantly with the Native versus Broiler chicken (Valar, Suguna, and Shanthi). Native chicken are left to scavange any wider range of feeds. The present study reported that reducing sugar content of native chicken meat had higher the value of 15.5% versus 12.5%, 13.5, and 12.5 of Valar, Suguna and Shanthi broiler chicken meat respectively. Comparative assessment of native chicken reducing suger content (15.5 mg/g. dry wt.) was similar to Suguna chicken (13.5 mg/g) (Table 2). The study conducted by Chepkemoi et al. [14] showed that the carbohydrate content was highest in indigenous chicken (8.25%) but the commercial chicken meat showed the least carbohydrate content (2.57%). According to Haunshi

et al. [13] the nutritional content of poultry meat varies from one study to another and this can be attributed to variation in the breed, feed, age at slaughter, the system of production, sex, processing, and the part of the cut.

3.4 Determination of Ash

The results on comparative assessment of mean average ash content of Native versus Broiler chicken (Valar, Suguna, and Shanthi) meat showed that the concentration was 5.05mg/g, versus 2.30, 4.33, and 2.75 (mg/g. dry wt.) respectively (Table 3). Similar study conducted by Choe et al. [15] reported that crude ash content measured in the Korean native chicken was (0.96%) higher than broiler chicken meat (0.47%). The study conducted by Ezhil Valavan et al. [11] showed that the ash content of native chicken's breast meat was 2.09±0.35 and 1.41±0.04 of broiler chicken. But, the ash content of native chicken's thigh meat (mg/g) showed 1.65±0.17 and 1.17±0.05 of broiler chicken. The results are in agreement with the findings of Choe et al. [15] and Ezhil Valavan et al. [11].

3.5 Determination of Fat

The fatty acid composition of meat is influenced by factors other than diet including genotype, gender and age of the animal. According to De Smet et al. [6] genetic factors could affect the meat fatty acid composition, but to a lower extent than dietary factors. Genetic variability relates to differences between species, between breeds or lines, variation due to the crossing of breeds and variation between animals within breeds. Atteh [16] observed that fact content of muscles was dependent on the feeds of bird that should contain sufficient energy value.

The chicken feed plays a vital role in determining the fatty acid profile pictures of broiler tissues. The chicken feed rich in lipid could modify the fatty acid content of the chicken meat. Higher the fatty acid content of chicken meat in turn is beneficial to human health. For instance, the beneficial fatty acids are such as Oleic acids, Oleic acid plus Omega 3 fatty acids. Therefore, modification of chicken fatty acid profile may fulfill the demands of health cautious beef eating competitors. Eventually, the producers may also be benefited.

The results of present study found out that Valar chicken had a fat value of 9.3%, 5.5% fatty acid content and 3.7% lipid content. The Suguna chicken had a value of 10.3%, 6.0% fatty acid and 4.11% of lipid content. The Shanthi chicken meat had a value of 9.7% fat content, 3.5% fatty acid and 2.0% lipid. In comparison to native and broiler chicken, the Shanthi chicken showed poor fat, fatty acid and lipid contents. The Native chicken had the highest fat content of 11.64% while the fatty acid content was 8.5% and its total lipid content was 5.5%. The Suguna broiler chicken meat in this study had higher the fat, fatty acid and lipid profile than other broiler chicken samples such as Valar and Shanthi chicken (Table 4).

Choe et al. [15] observed that crude fat contents of the thighs of Korean native chicken was lesser (2.98%) than commercial broilers (4.74%). Jsturasitha et al. [9] investigated revealed Thai native chicken had lower fat content than broiler chicken such as (0.12 vs 0.34%, respectively).

3.6 Determination of Fiber Content

The comparative assessment of crude fiber content of Native versus Broiler chicken showed that fiber content of native chicken (13.5%) was higher than broiler chicken(range 11 -12.45.mg/g dry wt.). The results are presented in the (Table 5). Similar study conducted by Ezhil Valavan et al. [11] observed that native chicken breast meat and thigh muscle meat's fibre content were 0.18±0.01 and 0.22±0.03($P < 0.01$) respectively. The value was higher compared to breast and thigh muscles of broiler chicken.

3.7 Determination of pH

The pH value was higher in valar chicken (6.16). Jaturasitha et al. [9] reported that the pH level of Native chicken had to be lesser than that of Broiler chicken. They observed that previously the more aggressiveness of Native chicken lead to higher the stress which drew more glycogen into use. Eventually, this affected the process of post mortem glycolysis that had lead to more lactic acid accumulation. The low pH value in native chicken was evidently due to high accumulation of lactic acid at this stressful stage. The pH of the native chicken was 5.61. But the broiler chicken showed the values of 6.04 (Suguna) and 6.0 (Shanthi) samples (Table 6).

3.8 Determination of Minor Biochemical Parameters in Chicken Samples

The deficiency diseases of calcium for bone and tooth development in child (Rickets), iron for hemoglobin of RBC in pregnant women (anemia); are obviously dependent on mineral component of food stuffs as found in chicken meat. Native chicken had the highest mineral content. They were Calcium, Phosphorous, Potassium, Magnesium, Manganese, Zinc, Iron, Ash, Native chicken showed highest calcium content (6

mg/g dry wt). The results reported by Chepkemoi et al. [14] on zinc and iron content of indigenous native chicken were higher. The minor biochemical elements of Fibrous proteins, and Myosin proteins were significantly higher in native chicken samples (Table 7).

3.9 Screening of Triiodothyroxine Growth Hormone using GC-MS Method

The results on estimation of triiodithyroxine growth hormone residues in various native and broiler chicken muscles using GC – MS method showed that there was a significant variation in the results of native versus broiler chicken sample analysis. The results indicated that there was a prominent peaks obtained in the GC-MS results in broiler chicken samples. This was compared with standard marker of triiodothyroxine (sigma). The hormone was not detected in Native chicken meat samples. The result may be considered with the use of this hormone in poultry industries to get more weight of muscles towards environmental exploitations (Fig. 2).

3.10 Statistical Applications

One way ANOVA was carried out for comparison of chicken meat sample results using IBM SPSS statistics 20 packages.

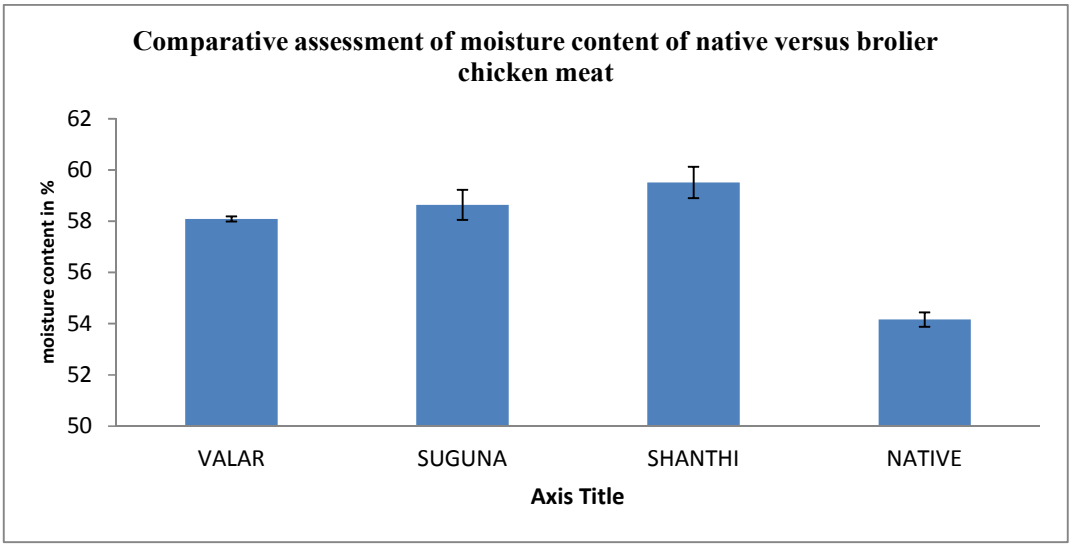

Fig. 1. Determination of moisture content of chicken meat samples

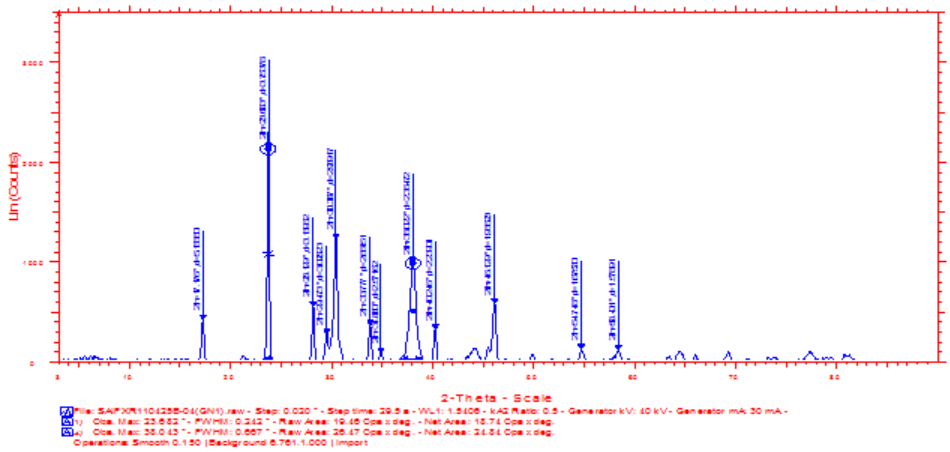

Fig. 2. Estimation of triiodothyronine growth hormone residues in various native and broiler chicken muscles using GC-MS method

Table 1. Comparative assessment of total protein content of native versus broiler chicken meat sample

S. No.	Test Sample	Average Protein content mg/g	Average Nitrogen content mg/g
1.	Native	15.5±0.87[a]	18.50±0.5[a]
2.	Valar	12.74±0.12[c]	14.50±0.43[c]
3.	Suguna	14.31±0.45[b]	16.68±0.17[b]
4.	Shanthi	12.07±0.75[c]	13.50±0.26[d]

Table 2. Comparative assessment of reducing and non reducing sugar content in native vs. broiler chicken samples

S. No.	Test Sample	Average reducing sugar content mg/g	Average total sugar content mg/g
1.	Native	15.5±0.3[a]	17.31±0.96[a]
2.	Valar	12.5±0.46[c]	12.5±0.86[b]
3.	Suguna	13.5±0.56[b]	14.33±1.15
4.	Shanthi	12.5±0.46[c]	13.73±0.87[b]

Table 3. Comparative assessment of ash content of native vs. broiler chicken meat

S. No.	Test sample	Ash content mg/g
1.	Native	5.07±0.51[a]
2.	Valar	2.33±0.49[c]
3.	Suguna	4.33±0.15[b]
4.	Shanthi	2.75±0.22[c]

Table 4. Comparative analysis of total fat, fatty acid and lipid content of native vs. Broiler chicken meat samples

S. No.	Test Sample	Average fat content mg/g	Average fatty acid content mg/g	Average lipid content mg/g
1.	Native	11.64±0.16[a]	8.5±0.17[a]	5.5±0.10[a]
2.	Valar	9.33±0.15[d]	5.5±0.17[c]	3.74±0.05[c]
3.	Suguna	10.3±0.26[b]	6.07±0.06[b]	4.11±0.12[b]
4.	Shanthi	9.7±0.1[c]	3.53±0.06[d]	2.08±0.16[d]

Table 5. Determination of Fiber content of Native Vs broiler Chicken samples

S. No	Test sample	Average fiber content
1.	Native	13.5±0.50[a]
2.	valar	12.45±0.10[b]
3.	Suguna	12.2±0.23[b]
4.	Shanthi	11.53±0.27[c]

Table 6. Comparative analysis of pH of Native Vs Broiler chicken

S. No.	Test Sample	pH
1.	Native	5.6±0.10[b]
2.	valar	6.17±0.15[a]
3.	Suguna	6.04±0.05[a]
4.	Shanthi	6.03±0.06[a]

Table 7. Comparison of minor biochemical parameters of native versus broiler chicken

Biochemical parameters*	Native chicken	Valar	Suguna	Shanthi
Calcium	6.1±0.27	2.53±0.06	4.34±0.141	3.7±0.05
Phosphorus	4.74±0.076	2.56±0.16	3.51±0.09	2.71±0.08
Potassium	3.6±0.14	2.52±0.10	2.30±0.03	2.02±0.12
Magnesium	2.51±0.11	1.49±0.09	2.33±0.05	1.30±0.09
Manganese	5.52±0.058	1.50±0.05	5.50±0.10	2.50±0.08
zinc	3.08±0.171	2.53±0.06	2.70±0.030	1.71±0.06
Iron	3.5±0.07	1.49±0.09	2.5±0.04	2.0±0.07
Ash	5.05±0.090	2.30±0.10	4.31±0.02	2.7±0.04
Fibrous proteins	8.49±0.100	5.50±0.12	6.00±0.22	3.52±0.02
Myosin proteins	2.53±0.10	1.50±0.02	2.04±0.07	1.51±0.04

4. CONCLUSION

In India the study analyzed the nutritional composition of broiler chicken gained a permanent position.Last year we have studied and the result supported that the nutritional parameters of native chicken was higher than broiler chicken. The GCMS test reported that the tri iodo thyronine (T3) hormone was not found in native chicken. The value of statistical test results showed and proved that the native chicken is better than broiler chicken.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Davis CG. Factors influencing global poultry trade. International Food and Agribusiness Management Review. 2015;18(A).
2. De Smet S, Raes K, Demeyer D. Meat fatty acid composition as affected by fatness and genetic factors: A review. Anim Res. 2004;53: 81-89.
3. AOAC. Official Methods of Analysis (12th ed.). Association of Official Analytical Chemists, Washington D.C., USA; 1995.
4. Andersson N. Skakkebaek. Exposure to exogenous estrogens in food: Possible impacton human development and health. Eur. J. Endocrinol. 1999;140(6):477-485.
5. Haddad EE, Mashaly MM. Effect of thyrotropin-releasing hormone, triiodothyronine, and chicken growth hormone on plasma concentrations of thyroxine, triiodothyronine, growth hormone, and growth of lymphoid organs and leukocyte populations in immature male chickens. Poult. Sci. 1990;69(7):1094-1098.
6. Schenck FJ, Donoghue DJ. Determination of organochlorine and organophosphorus pesticide residues in eggs using a solid phase extraction cleanup. J. Agric. Food Chem. 2000;48:6412-6415.
7. Shaikh B, Rummel N, Donoghue DJ. Determination of sulfamethazine and its major metabolites in egg albumen and egg yolk by high performance liquid chromatography.J. Liq. Chromatogr. Rel. Technol. 1999;22:2651–2662.
8. Lehotay SJ, Lightfield LR, Harman-Fetcho JA, Dan J. Donoghue. Analysis of pesticide residues in eggs by direct sample introduction/gas chromatography/tandem mass spectrometry. J. Agric. Food Chem. 2008;49: 4589–4596.
9. Jaturasitha1 S, Leangwunta1 A, Leotaragul A, Phongphaew1 T, Apichartsrungkoon N, Simasathitkul T, Vearasilp L, Worachai U, Meulen R. A comparative study of thai native chicken and broiler on productive performance, carcass and meat quality, Conference on International Agricultural Research for Development, Deutscher Tropentag. Witzenhausen. 2002;9-11.
10. Fletcher D. Broiler breast meat color variation, pH, and texture. J. Poult. Sci. 1999;78:1323-1327.

11. Ezhil Valavan S, Omprakash AV, Bharathidhasan A, Ramesh Saravanakumar V. Comparison of nutrient composition of native chicken and commercial broiler under Indian condition. IJAPSA. 2016;02(12):7–11.
12. Boni I, Nuru H, Noryati I. Comparison of meat quality characteristics between young and spent quils. International Food Research Journal. 2010;17:661–666.
13. Haunshi S, Sunil D, Kadirvel G. Comparative studies on egg, meat, and semen qualities of native and improved chicken Varieties Developed for Backyard Poultry Production. Tropical Animal Health and Production. 2010;42(5):1013–19.
14. Chepkemoi M, Macharia JW, Sila D, Oyier P, Malaki P, Ndiema E, Agwanda B, Obanda V, Ngeiywa KJ, Lichoti J, Ommeh SC. Physical characteristics and nutritional composition of meat and eggs of five poultry species in Kenya. 2017;29(8).
15. Choe JH, Kichang N, Samooel J, Binna K, Hyejeong Y, Cheorun J. Differences in the quality characteristics between commercial korean native chickens and broilers. Korean J. Food Sci. Ani. Resour. 2010;30:13-19.
16. Atteh JO. Principles and practice of Livestock feed manufacturing. Aldek Printers, Ilorin, Nigeria. 2002;10–30.