Research Article

Nutritional Characterization of Edible Viscera of an Autochthonous Swine Breed of Assam, India

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Abstract | Scientific information about the nutritional quality on meat by-products is very scarce when compared with muscle meats (e.g., physicochemical composition, meat quality, sensory and their utilization etc.). The meat by-products are widely accepted as edible foods by the consumers. Therefore, the current study aims to examine the nutritional composition of edible viscera namely, liver, kidney, spleen, heart, large intestine and small intestine of Doom pig of Assam, India. The samples were collected from semi-extensively reared pig farms of Kokrajhar district, Assam. The approximate composition range of these edible viscera was found to be: moisture 50.67-68.47%, fat 0.47-9.85%, ash 0.16-0.92%, protein 6.85-22.36%, carbohydrate 10.18-24.65% and calorie 124.31-198.16 k/cal/100g. Among the samples the maximum protein, carbohydrate and calorie were found in liver. Most numbers of the essential amino acids was found in spleen with lysine contributing the largest amount. Fat content was maximum in large intestine, similarly the saturated fatty acid content too was found highest in large intestine. Furthermore, the saturated fatty acid, mono unsaturated fatty acid and poly unsaturated fatty acid content range from 35.97-51.42%, 2.03-3.44% and 2.05-18.79% respectively. The study reports that the edible viscera are excellent sources of nutrients. The data may be utilized by the nutritionist and dieticians which may help in value addition and the promotion of the consumption of edible viscera, as well as their utilization in meat processing industry.

Keywords | Edible viscera, Autochthonous Doom pigs, India, Proximate, Amino acids, Fatty acids.

Received | July 27, 2022; **Accepted** | August 30, 2022; **Published** | September 25, 2022
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Citation | Daimari R, Narzari S, Sarmah J (2022). Nutritional characterization of edible viscera of an autochthonous swine breed of assam, India. Adv. Anim. Vet. Sci. 10(10): 2222-2227.
DOI | http://dx.doi.org/10.17582/journal.aavs/2022/10.10.2222.2227
ISSN (Online) | 2307-8316

INTRODUCTION

The Doom, commonly known as 'Desi' by rearers, is an autochthonous pig breed found in western part of Assam. It has good meat texture, taste and palatability and its meat is sold at higher market prices than other crossbreed pigs. The pig has larger body size, high prolificacy and survival rate even in the low input system that is highly polymorphic in nature (Zaman et al., 2014). Doom is the first registered indigenous pig breed of Assam with accession no. INDIA_PIG_0200_DOOM_09006 (NBAGR, 2008). The Doom pig's viscera are extensively consumed by the local population as edible foods. Consumption of edible viscera is advocated by nutritionists because of its high content of essential amino acids (EAA), vitamins and minerals (Kicinska et al., 2019).

Till recently meat scientists has focused on muscle meats (Thomas et al., 2016a; Thomas et al., 2016b; Daimari et al., 2022) whereas now focus has been shifted to the edible viscera which is also consumed as low cost human food. Due to lack of scientific studies limited data has been generated on its nutritional content. Edible viscera are rich in nutrient content such as minerals (P, Fe, Cu, Mg, I, Ca, K, Na, Se, Zn, Mn) required by human body (Cordian et al., 2002). They are also known for high quantities of essential fatty acids such as arachidonic acid, the omega-3 fatty acids, EPA and DHA (Cordian et al., 2002; Park et al., 1993; Nicklas et al., 2014). Edible viscera also contain folate, choline and B12 which are difficult to obtain elsewhere (Oloruntuba et al., 2019).

Therefore, the present study determines the proximate, amino acid and fatty acid content in six edible organs namely; heart, kidney, liver, small intestine, large intestine and spleen of Doom pig of Assam, India.

MATERIALS AND METHODS

Sample collection

The study was approved by the Institutional Animal Ethics Committee; Bodoland University vide letter no- IAEC/BIOTECH/2019/3. Samples of Doom pig's edible viscera were obtained from six semi-extensive indigenous pig farms in Kokrajhar district of Assam, India, with live weights of around 48-51 kg. These indigenous pigs were given locally available feed like local vegetation, remnants of crop and kitchen waste. Ten months old pigs (n=12) were slaughtered in a commercial abattoir, later six edible viscera samples were collected from each of the 12 pigs. The heart (53.46±1.35 g), liver (887.31±23.30 g), kidney (189±2.42 g), spleen (78.36±1.38 g), small intestine (432.26±21.52 g) and large intestine (1432±64.30 g) were conventionally chilled for 24 hours in a chiller at 4 ° C. 24-hours post-mortem, the viscera's were homogenized using a mechanical grinder. The experiments were performed at the Department of Biotechnology, Bodoland University.

Proximate Analysis

The proximate composition (moisture, ash, fat, protein, carbohydrate and calorie) of the edible viscera were determined according to the methods of analysis of the AOAC (2005). The carbohydrate content was calculated by the difference method, following conversion formula (FAO 2003):

Available Carbohydrate (%) = 100- [Moisture (%) + Ash (%) + Crude Protein (%) + Fat (%)]

The nutritive value or calorific value in kcal/100 g was calculated with the help of the following equation (James 1995):
Calorific value (kcal/100 g) = 4 × Protein (%) + 9 × Fat (%) + 4 × Carbohydrate (%)

Amino Acid Analysis

The amino acid content of the samples was determined on 0.001 g of oven dried and defatted samples. The samples were dissolved in 2mL of milliQ-water and incubated at 45° C in thermomixer for 30 minutes. 8mL of methanol was added to precipitate the proteins and incubated overnight at -20° C. The solutions were centrifuged at 4000 rpm for 30 minutes and the supernatant was transferred to another tube. The supernatant was evaporated under nitrogen gas at 60 ° C to complete dryness. Derivatization was done by adding 350 μL of Borate buffer, 20 μL of AccQ-Tag ulta reagent to the sample and incubated for 10 minutes at 55 ° C. After incubation 2 μL is loaded on the instrument, which is quantified using a Sigma standard. Buffer for Mobile Phase A: AccQ- Tag Ultra eluent A1 and for Mobile Phase B: AccQ-Tag Ultra eluent B. Amino acids were determined using a WATERS Acquity (make) UPLC system.

Fatty Acid Analysis

For fatty acid determination, the dried samples were subjected to lipid extraction with chloroform/methanol (Folch et al., 1957). The lipid extract was esterified with BF_3- methanol (Joseph et al., 1992) for preparation of FAME's (Fatty Acid Methyl Esters). The fatty acid composition of each aliquot was estimated by Gas Chromatography. GC-MS analysis of sample extracts was carried out with Perkin Elmer (USA), Model: Clarus 680 GC & amp; Clarus600C MS comprising a liquid auto-sampler. The Software used in the system was TurboMass Ver.6.1.2. The peaks were analyzed using data analysis software NIST-2014. The capillary column used is 'Elite- 5MS' having dimensions- length- 60 m, ID- 0.25 mm and film thickness- 0.25 μm and the stationary phase is 5% diphenyl 95% dimethylpolysiloxane. Helium gas (99.99%) was used as carrier gas (i.e. mobile phase) at flow rate of 1 ml/minute. An injection volume of 2 μl was employed in split less mode. Injector temperature was 280°C and ion-source temperature 180°C. The oven temperature was programmed at 60°C (for 1 minute), with an increase at the rate 7°C/minutes to 200°C (hold for 3 minutes) then again increased at rate of 10 °C/min to 300 °C (hold for 5 min).The total run time is ~ 39 minutes. Solvent delay was kept for 8 minutes. MS Protocol Mass Spectra was taken in Electron Impact positive (EI+) mode at 70 eV. For MS scan, a solvent delay of 8 minute was provided with m/z range 50-600 amu.

The mentioned data were statistically analyzed using SPSS, Version 26.0 and demonstrated as mean ± SEM (standard error of mean).

RESULTS AND DISCUSSION

Proximate Composition

The mean values of proximate composition of Doom pig are tabulated in Table 1. Spleen and small intestine had the highest moisture content (68.47% and 64.72%, respectively), while kidney, heart and liver had significantly lower

Table 1: Proximate composition of edible viscera Doom pig.

	Kidney	Small Intestine	Spleen	Liver	Heart	Large Intestine
Moisture (%)	61.93±1.43	64.72±2.18	68.47±0.99	50.67±1.22	55.25±1.57	64.11±1.26
Fat (%)	2.28±0.28	0.58±0.03	0.47±0.02	1.12±0.17	1.16±0.56	9.85±0.51
Ash (%)	0.78±0.18	0.51±0.02	0.92±1.04	1.19±0.09	0.75±0.13	0.16±0.00
Protein (%)	20.93±0.18	10.12±0.40	19.83±0.11	22.36±0.52	18.23±0.48	6.85±0.48
Carbohydrate (%)	14.15±1.016	24.05±1.83	10.18±1.05	24.65±1.03	24.59±1.98	19.02±1.13
Calorie (kcal/100g)	160.94±7.26	141.97±8.63	124.31±3.57	198.16±5.40	181.80±3.58	192.17±6.85

moisture contents (61.93%, 55.25% and 50.67% respectively). Also the crossbred pigs of Thailand had high moisture content (79.96%) than the present study (Chanted et al., 2021). Another study by Seong et al., (2014), determined the proximate content in pork-by products of crossbred pigs (Landrace×Yorkshire×Duroc) of South Korea, when they found high content of moisture with an overall mean value of 76.38%.

Among the edible viscera determined, large intestine had the highest fat content (9.85%), followed by kidney (2.28%), heart (1.16%), liver (1.12%), small intestine (0.58%) and spleen (0.47%). The present study had similar data with that of fat content of crossbred pigs of Thailand (Chanted et al., 2021). Another study reported high amount of fat content in large intestine (19.54%), followed by heart (4.55%) and lowest in spleen (0.97%) (Seong et al., 2014), which was similar with our study, even though contents were high. High content of fat (5.86-25.97%) was also found in the muscles of different beef breeds (Ba et al., 2013; Moon et al., 2006). Fat not only helps in building up energy but also helps in vitamin absorbance.

Liver had the highest content of ash (1.19%) followed by spleen (0.92%) and large intestine had the least ash content with a mean of 0.16%. Similar data were also found in the pork by-products of crossbred Korean pigs (Seong et al., 2014). The ash content determines one of the most important properties of food i.e. the nutritional value, quality and physiochemical factors (Keran et al., 2009).

Liver and kidney had the highest protein content with a mean of 22.36% and 20.93% respectively, followed by spleen (19.83%) and heart (18.23%) while lowest was found in small intestine (10.12%) and large intestine (6.85%). Previous studies by Seong et al. (2014), too reported high protein content in liver (22.05%) and lowest in large intestine (8.45%). Similar values of protein content were also reported in the liver of similar species (Kim et al., 2008).

The carbohydrate content determined by difference was found to be highest in liver (24.65%), heart (24.59%) and small intestine (24.05%) respectively, while lowest was found in spleen with a mean of 10.18%.

The nutritional calorie/energy value of edible viscera showed that liver had the highest energy content (198.16 kcal/100g), followed by large intestine (192.17 kcal/100g). While heart (181.80 kcal/100g) and kidney (160.94 kcal/100g) had significantly lower content whereas small intestine (141.97 kcal/100g) and spleen (124.31 kcal/100g) had the lowest energy content. On the other hand, studies by Seong et al. (2014), reported highest calorie content in large intestine, followed by pancreas and liver respectively.

Amino Acid Content

The amino acid content in edible viscera of Doom pig is depicted in Table 2. Our data show a large variation among the determined edible viscera of both essential and non-essential amino acids. Among the edible viscera, spleen had the highest levels of most Essential Amino Acids (EAA) such as lysine, phenylalanine, isoleucine and leucine, whereas heart and large intestine had the lowest levels of most EAA. Spleen had the highest content of lysine (10.33 %), followed by kidney (5.29%), liver (5.02%) and heart (5.02%). Methionine (0.94%) too was found highest in spleen. Studies by Seong et al. (2014), too showed similar results where lysine was found highest in liver, while that of methionine was found highest in pancreas and liver, followed by spleen. Small intestine and heart had the highest content of valine. On the other hand isoleucine was found highest in kidney (8.57%), followed by large intestine (8.54%) and lowest was found in heart (0.03%). Leucine (2.34%) and phenylalanine (8.61%) were both found highest in spleen and that of tryptophan (0.85%) was found highest in liver. Valine was found highest in pancreas and liver as reported by Seong et al. (2014). It is found that most of the EAA are found highest in pancreas followed by liver and heart, which was quite similar with our findings (Seong et al., 2014). While that of amino acid content in muscle *longissimus thoracis et lumborum* of Ghungroo pigs of Assam reported quite similar findings with the edible viscera of present study (Thomas et el., 2016b).

Human body cannot produce EAAs and must be provided from outside. Human body cannot function normally without these amino acids. Earlier studies have reported

Table 2: Amino acid composition in edible viscera of Doom pig.

Moles %	Kidney	Small Intestine	Spleen	Liver	Heart	Large Intestine
Essential Amino acids						
Lysine	5.29±0.04	1.24±0.01	10.33±0.01	5.02±0.01	5.02±0.01	1.45±0.01
Methionine	0.15±0.02	0.03±0.01	0.94±0.01	0.23±0.01	0.01±0.00	0.02±0.01
Valine	0.14±0.00	5.34±0.02	0.04±0.01	0.15±0.02	4.11±0.00	0.87±0.00
Isoleucine	8.57±0.33	5.89±0.33	5.11±0.01	0.25±0.01	0.03±0.01	8.54±0.30
Leucine	2.15±0.02	0.15±0.02	2.34±0.01	1.34±0.01	0.15±0.02	0.51±0.01
Phenylalanine	1.14±0.02	3.92±0.01	8.61±0.14	5.84±0.01	0.03±0.01	0.44±0.02
Tryptophan	0.07±0.01	0.12±0.03	0.46±0.02	0.85±0.02	0.04±0.01	0.26±0.02
Non-Essential Amino Acids						
Serine	0.11±0.012	5.22±0.02	2.02±0.02	8.87±0.01	3.68±0.21	1.87±0.04
Glutamine	1.22±0.01	7.61±0.31	0.83±0.04	7.23±0.28	1.75±0.00	0.05±0.02
Arginine	1.60±0.02	1.76±0.31	4.20±0.05	12.67±0.05	2.26±0.02	3.32±0.02
Glycine	4.92±0.02	5.34±0.32	2.98±0.31	1.79±0.12	9.32±0.02	9.30±0.01
Aspartic acid	1.46±0.01	5.22±0.02	0.82±0.03	13.05±0.02	2.15±0.02	1.77±0.00
Glutamic acid	8.92±0.02	10.82±0.09	2.61±0.04	17.12±0.32	0.16±0.00	1.77±0.04
Alanine	0.03±0.00	0.02±0.00	4.46±0.01	4.54±0.07	0.02±0.01	0.01±0.00
Proline	4.12±0.01	0.05±0.01	3.33±0.02	13.81±0.01	0.03±0.00	0.36±0.12
Cysteine	1.34±0.02	1.79±0.05	1.45±0.01	2.13±0.02	1.34±0.00	0.01±0.00
Tyrosine	0.23±0.01	0.04±0.01	1.25±0.01	1.57±0.01	0.04±0.01	0.02±0.01

Table 3: Fatty acid composition (expressed in FAME %) in edible viscera of Doom pig.

Parameters	Kidney	Small Intestine	Spleen	Liver	Heart	Large Intestine
SFA	39.03±0.01	46.88±3.20	43.55±0.02	35.97±0.31	39.98±0.34	51.42±0.29
MUFA	2.69±0.15	2.72±0.19	3.44±0.11	2.03±0.02	2.55±0.27	2.06±0.64
PUFA	2.91±0.48	6.26±0.28	18.79±0.15	12.72±0.89	5.28±0.43	2.05±0.35
Palmitic acid (C16:0)	18.68±0.59	24±23±0.24	20.58±0.33	12.26±0.58	19.20±0.29	26.40±0.92
Stearic acid (C18:0)	12.38±0.13	18.74±0.11	16.00±0.05	18.62±0.01	14.81±0.14	22.57±0.18
Lauric Acid (C12:0)	2.61±0.39	3.29±0.56	3.00±0.05	1.90±0.13	1.60±0.34	2.08±0.49
Myristic acid (C14:0)	2.60±0.30	3.57±0.58	1.86±0.49	1.53±0.34	2.43±0.56	2.75±0.56
Behenic acid (C22:0)	2.11±0.24	2.65±0.30	2.74±0.56	1.70±0.26	1.65±0.31	2.37±0.30
Arachidic acid (C20:0)	0.77±0.33	0.75±0.02	0.34±0.01	0.74±0.02	1.30±0.01	0.74±0.02
Alpha Linolenic acid-ALA (C18:3-cis, n-3)	0.25±0.07	0.23±0.09	0.36±0.06	0.42±0.15	0.38±0.18	0.14±0.05
Linolenic acid-LA (C18:2-cis)	0.14±0.02	0.03±0.01	0.16±0.02	0.31±0.02	0.04±0.01	0.05±0.00
Arachidonic acid-ARA (C20:4 C, n-6)	2.26±0.02	5.76±0.02	18.24±0.02	13.44±0.06	4.82±0.02	1.65±0.02
Oleic acid (C18:1n-9)	0.65±0.02	1.08±0.01	1.24±0.01	0.86±0.02	0.26±0.02	0.74±0.01
Palmitoleic acid (C16:1n-7)	1.76±0.32	1.58±0.32	0.96±0.07	1.06±0.07	1.56±0.34	1.07±0.09
Myristoleic acid (C14:1n-7)	1.07±0.34	0.92±0.68	1.76±0.32	0.49±0.31	1.07±0.31	0.64±0.30

that the levels of EAA in edible viscera are not reduced after cooking or due heating treatment because of the low reducing sugar content of edible viscera do not cause secondary degradation reactions (Aristoy and Toldra, 2011). From our present study, it is observed that the edible viscera especially spleen, liver and heart are good sources of EAA's.

The non-essential amino acids detected in edible viscera of Doom pig are serine, glutamine, arginine, glycine, aspartic acid, glutamic acid, alanine, proline, cystine and tyrosine. Among the non-essential amino acids (NEAA), glutamic acid (17.12%), proline (13.81%), aspartic acid (13.05%) and arginine (12.67%) represented the highest amount of non-essential amino acids.

Fatty Acid Content

The fatty acid composition (expressed in FAME %) are depicted in Table 3. The highest percentage of saturated fatty acid (SFA) was in the small intestine (52.54%) and large intestine (51.42%). Lower and similar percentages of SFA's were found in spleen (43.55%), heart (39.98%), kidney (39.03%) and lowest value was obtained in liver (35.97%).

Spleen, small intestine and kidney showed significantly higher mono-unsaturated fatty acid (MUFA) percentages (3.44%, 2.72% and 2.69%, respectively), whereas lower and significantly similar percentages were found in large intestine and liver (2.06% and 2.03%, respectively). Higher percentages of poly-unsaturated fatty acid (PUFA) was obtained in spleen and liver (18.79% and 12.72%, respectively), whereas lower and similar percentages are obtained in small intestine (6.26%) and heart (5.28%). Lowest was reported in kidney and large intestine (2.91% and 2.05%).

When compared with the SFA's, MUFA's and PUFA's content in muscle tissues of Ghungroo pig breed, they reported higher content (44.55%, 32.38% and 15.81% respectively) (Daimari et al., 2022). Earlier studies reported high SFA content in large intestine and spleen of crossbred pigs of Korea which was similar with our findings, similarly that of MUFA was found highest in large intestine and that of PUFA was found highest in liver and spleen (Seong et al,. 2014). Another study determined the fatty acid content in liver, kidney, heart and tongue of veal calves and suckler beef, where SFA's was reported highest in liver and that of MUFA was obtained highest in tongues and heart of beef and calves, while that of PUFA was reported highest in heart and kidney of beef (Florek et al., 2012).

SFA's are regarded as bad fatty acid as they are known to trigger various diseases mainly related to coronary heart disease. In regard to this, the Food and Agriculture Organization (FAO) and World Health Organization (WHO) has recommended dietary intakes of total fat and fatty acids for adult humans i.e. SFA content should be less than 10%, 15-20% MUFA and 6-11% PUFA (Burlingame et al., 2009). Therefore, nutritionist recommends reducing the intake of SFAs and thereby increasing the intake of PUFA's is encouraged.

The SFA's detected in our study are palmitic acid (C16:0), stearic acid (C18:0), lauric acid (C12:0), myristic acid (C14:0) and behenic acid (C22:0). Palmitic acid was detected the highest among the SFA's followed by stearic acid. The MUFA's detected in our study are oleic acid (C18:1n-9), palmitoleic acid (C16:1n-7) and myristoleic acid (C14:1n-7). And the omega-3 PUFA detected are alpha Linolenic acid-ALA (C18:3-cis, n-3) and omega-6 PUFA detected are linolenic acid-LA (C18:2-cis) and arachidonic acid-ARA (C20:4 C, n-6).

Limitations Of The Study

The data obtained in the study could not be compared with earlier studies on the nutritional content of edible offal of the same pig breed or any other autochthonous pig breeds of India as only limited data has been generated from similar works.

CONCLUSION

The present study determined the nutritional characteristics of pork by-products i.e. edible viscera on the basis of their proximate composition, amino acid and fatty acid content. The data obtained from our study defines that edible viscera especially spleen, liver and heart are good sources of nutrients such as proteins, EAA and PUFA (omega-3 and omega-6) as an essential fatty acid. Furthermore, when compared with the fatty acid content of the muscles, the edible viscera had lower fatty acid content. Thus, it may be concluded that the pork by-products i.e. edible viscera are suitable for human consumption. Also these by-products may be suitable for processing them into other final products in order to increase economic benefits. This is the first study to characterize the nutritional compositions of edible viscera which would provide not only the useful information for consumers but also the important databases for further investigations. Further; the data shall be of great importance in promotion of consumption of edible pork by-products as well as their utilization in meat processing. Attempts may be made to increase the commercial values of edible offal by using them in various meat products.

ACKNOWLEDGEMENTS

The authors are thankful to the Guwahati Biotech Park (GBP), Guwahati, India and the Sandor Speciality Diagnostics Private Limited, Hyderabad, India.

CONFLICT OF INTEREST

The authors report there are no competing interests to declare.

NOVELTY STATEMENT

The study highlights that the edible viscera of Doom swine breed are excellent sources of nutrients. The viscera by-products may be suitable for processing into other final products in order to increase economic benefits to the rearers.2

AUTHOR'S CONTRIBUTION

Rijumoni Daimari: Conceptualization of the study, manuscript writing, analysis and interpretation of data, formulation of proposed strategies. Silistina Narzari: Assisted in laboratory work and editing of the manuscript. Jatin Sarmah: Designed the study, manuscript finalization and revisions necessary.

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