Original Research Article

Nutritional and anti-nutritional compositions of rice bran as a potential animal feed.

Abstract

Rice bran is a byproduct of rice milling and highly enriched in nutrients such as carbohydrates, protein, and minerals. It is a potential candidate for animal feed formulations, including poultry feed, due to its nutritional, economic benefits and, high energy content, which helps to improve overallperformance and productivity of birds. The presence of these amino acids in rice bran makes it a valuable ingredient in feed formulations, as it helps in meeting the protein requirements of the birds and supporting muscle development.

The proximate analysis of raw rice bran was conducted according to AOAC prescribed standards and the micronutrients/antinutritional components were evaluated using atomic absorption spectroscopy. In addition to high energy content and protein, rice bran was found also to be rich in minerals such as calcium, phosphorus, and potassium. These minerals are essential for bone development, egg production, and overall health and immunity of poultry. Incorporating rice bran in poultry feed formulation helps in ensuring adequate mineral intake, thereby improving the overall well-being of the birds. Rice bran also has a favorable fat profile, with higher content of unsaturated fatty acids compared to other feed ingredients. These fatty acids have been shown to have positive effects on poultry health, including improved egg quality, enhanced immune function, and reduced incidence of certain diseases.

The moisture content of rice bran was determined as 4.3338% and the crude fiber content was evaluated as 16.3675%. Carbohydrate, Protein, and lipids were determined as 26.3900%, 14.4469%, 19.6418% respectively. The total ash content was 18.8126%.The proximate compositions compared favorably well to commercial feed formulations for starter, grower, and finisher meals. The moisture content for the starter, grower and finisher meals were 5.6804%, 8.1933% and 8.4552% respectively, crude fiber content was 4.3100%, 3.6450% and 3.1425% respectively. The carbohydrate contents are 41.2677%, 41.8456% and 49.4032%. The crude fat contents are 12.0354%, 12.3676% and 12.6180% while crude protein contents are 26.2688%, 27.5813% and 21.8906%. The total ash content was measured at 10.4377%, 6.3672% and 4.4905%.

The trace mineral composition of the rice bran (mg/100g) were Calcium (63.9800), Magnesium (227.0620), Potassium (440.8960), Phosphorus (20.0658), Sodium (54.8480), Iron (88.5630),and Copper (1.9450). The heavy metal composition includes lead (0.8020) and cadmium was below the detection limit of the M5 Thermo Scientific atomic absorption Spectrometer used for the analysis.The anti-nutrient composition of the rice bran in mg/100g were Anthraquinones (0.0134), Phenolics (0.0296), Tannins (0.2655), Saponins (6.4470), and Phytic acid (62.0466).

In conclusion, inclusion of rice bran in poultry feeds formulations with added additives meets the nutritional requirement and compared favorably with commercially available poultry feeds. There is also need for the addition of phytase enzyme (anti-nutritional component) to degrade phytic acid in the rice bran in other to improve the nutrient availability.

INTRODUCTION

Intakeofwholegrainshasbeenshowntoreducetheriskofdiabetesandcardiovasculardiseaseinpopulationstudiesdueto protective factors including dietary fiber, vitamin E and othernutrients.[1,2]Mostofthedietaryfiberfromplantsources,includingcerealbrans,isclassifiedasinsolubledietaryfiber (IDF).TheIDFofcerealbranismainlycomposedofcellulose,hemicelluloseandligninthatcontainseveralfunctionalgroups such as alcohols, aldehydes, ketones, carboxylic acid,phenolicandetherlinkages.[3]Thesegroupshaveastrongaffinitytobindwater,oil,ortoxic metalions.However,to use these cereal brans they require some level of pre-treatmentthathelpstoexposethebindingsitesorincreasetheporosity[4,5].Therefore,severalcommonphysicalandchemicalpretreatments such as micronization,[6] enzymatic treatment[7] aswell as some inorganic and organic bases, acids, and salt solutions[3] treatments have been reported. The extent of physicalpretreatment strongly depends on particle size while enzymeprocesses usually require complex steps and high dosage ofcostlyenzymesaswellasprecisеregulationofreactiontemperature. In many instances, acidic pretreatment is foundmoresuccessful,primarilyduetoeasyremovalofimpurities andionsthatmightblockthefunctionalgroupsorporousstructures.[3,8,9]

Ricebran(RB)isabyproductofricemilling andisleft overinlargequantitybythericeindustryeveryyear.Inrecentyears,most of the rice bran produced is used as animal feed ingredient, fertilizer,and fuel[10]. But there is an underestimated potential for high-value rice bran production because of its high content of IDF.Ricebran(RB)iscomposedofabout27%DF[7]andalmost90%of the IDF accounts for rice bran DF as the main component[11].Though, a few studies for improving insoluble rice bran fiber(IRBF)functionalpropertiessuchasfatbindingandemulsifying capacity to stabilize emulsions of the food system[7] or theadsorptioncapacityoffiberfortheremovalofNi (II)fromaqueoussolution[3]havebeenreported,inmostofthesestudies,the use of rice bran was based on its granular structure, insolubilityinwater,chemicalstabilityandlocalavailability.

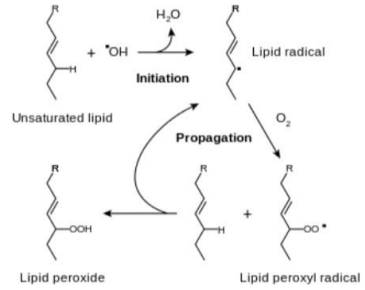
Sincethechemicalpre-treatmentscanpotentiallymodifythecellsurfaceeitherbyremovingormaskingthegroupsorexposing more porous structure, therefore, it was reported that theeffect of inorganic acid

concentration on the composition andmicrostructure might expose various functional propertiesof IRBF. The study aimed to simplify the extractionprocess,reducethecost,andimprovethephysicochemicalproperties of IRBF for food system enrichment, and to understand therelationship between microstructure andphysicochemicalpropertiesofthe fiber.Acid inducedmodificationofcerealfibersandfurthercharacterization of theirphysicochemicalattributesintermsofwaterholdingcapacity(WHC),oilbindingcapacity(OBC),swellingcapacity (SWC)andcation-exchangecapacity(CEC) were reported[12,13].Also, acid–baseregimes were applied onricebranandtheirstructuralandphysicochemicalcharacteristics were evaluated.

PhysicochemicalCharacteristicsofRiceBran

Rice bran, contributes about 10% of whole grain weight, is obtained duringmillingasaby-product.Everyyearproductionofricebranisincreasingastheproductionofpaddyincreases;theworldricebranproductionin2014–2015isabout23.80 million tonnes, and during 2015–2016, it is estimated to be 38.50 milliontonnes(SEA,India).Interestinthefieldofricebranisincreasingduetoitsamazingnutrientcontent and its health benefits.Raw rice bran has characteristics bland flavor, slightly bitter, and sweet intaste.Ricebranisnutritionallyrichinprotein,fiber,lipids,andantioxidants;italsocontainsanti-nutritionalfactors,suchastrypsininhibitorandlectins.Brancontainsless soluble fiber and cholesterol-lowering and other health-enhancing properties.These include rice bran oil, plant sterols, tocopherols, oryzanol, and β-sitosterol(Sairam et al. 2011). Although the nutritional and food potential of rice bran havebeen recognized, the consumption of rice bran in human foods has been limited toa very small quantity due to quick rancidity of the bran as shown on Fig 1. Stabilization process willinactivate the rancid-causing enzyme and anti-nutritional factors (Sharma et al.2004). As bran deteriorates due to the presence of lipase enzyme, the flavor of rice bran also changesto incipientrancid, musty,and sour.

Physicochemicalpropertiesareveryimportantparameters that determine the characteristics ofricebranduring food processing. Physicochemical properties include physical properties(bulkdensity,color,particlesize,wateractivity),functionalproperties(waterabsorption capacity, water solubility, and fat/oils absorption), and chemical properties (nutritionalcontent,antioxidantproperty).Thesepropertieswill guide maximum utilization ofricebraninfoodapplications.Inthisstudythenutritional and anti-nutritionalpropertiesofrice brananditsimportancewere investigated. The nutritional composition of the rice bran was compared with that of commercially available poultry feeds.Raw rice brans are often stabilized beforecommercial application.Theprocessofstabilizationalsoaffectsthepropertiesofbran.



The free radical pathway for the first phase of the oxidative rancidification of fats

Fig 1: Rancidity-CausingEnzymes in Rice Bran

Theprimaryreasonforthefailureofusingricebranasafoodsourceisthepresenceof rancid-causing enzymes, which are activated soon after the milling. If oil is notextractedsoonaftermilling,theoilishydrolyzedintofreefattyacids(FFA)andglycerol.Theresultin gfattyacidsincreasebranacidityandreducepH;produceoff-flavorandsoapytaste;andchangethefunctionalpropertiesofthebran.Ingrain,enzymes are present in the testa-cross layer of the grains, but oil vacuoles are present in aleurone and sub-aleurone layers and in germ. Upon milling, oil isexposed to lipases, causing rapid breakdown to free fatty acids at the rate of 5–7%oftheweightofoilperday.

Stabilization of Rice Bran

It is necessary to stabilize the rice bran by using suitable techniques for controllingundesirable reactions to avoid rancidity, off-flavor, refining loss, and deteriorationofnutrients.Stabilizationisaimedatdestructionorinhibitionoflipase,theenzyme that cause the development of FFA. This is done to reduce refining losses, whicharedirectlyproportionaltotheFFAcontent.Toprocessbranintoafoodgradeproductofgoodqualit yandhighindustrialvalue,allthecomponentswhich cause deterioration must be removed or their activity should be arrested. Importantinthis respect isinactivationofenzymesmustbecompleteandirreversible.Atthe same time, the valuable nutrients should be preserved. Bran, after proper stabilization, can serve as a good source of protein, essential unsaturated fatty acids,calories,andnutrients suchastocopherols andferulic acidderivatives.Two types of rancidity will occur in rice bran such as oxidative rancidity andhydrolytic rancidity. Oxidative rancidity is caused by the oxidation of the doublebondsofthefattyacids,whilehydrolyticrancidityiscausedbytheremovaloffattyacidsfrom the glycerol molecule

Several thermal methods are used for rice bran stabilization (to inhibit lipaseactivity). Heat treatment is the most common method to stabilize rice bran. Hightemperaturesabove120°Cdenaturetheenzymeresponsibleforlipiddegradationinricebranwitho utdestroyingthenutritionalvalueofthericebran(Thanonkaewet al. 2012). Most of the processes involve dry or moist heat treatment. It is suggested that moist heat treatment may be more effective than dry heat, but few processesthatusesteamhaveachievedsatisfactoryresults.Toachieveproperstabilization, every discrete bran particle must have a proper moisture content,depending upon the time and temperature of the treatment.

PhysicalProperties

Raw rice bran has light tan or yellowish-brown color, and it varies according to thevariety and the processing conditions. Most of the bran is stabilized beforecommercialapplications. In heat processing, the color of the bran changes to brown by losing its yellowish light color, and the bran stabilized by dry heat changes its color tobrown and extruded bran dark brown (Garcia et al. 2012). Upon heating, change inthe color may be attributed to the formation of Maillard reaction compounds andpartlyto the color of pigments of raw bran.Rice bran is composed of beta-carotene and lycopene, which is considered ascarotenoid, which gave its reddish-brown appearance. Lycopene and beta-carotene are precursorsofvitaminA,andbothofthemareantioxidantsinfoodandbiological system (Lambertsetal.2006).Carotenoids actasphoto-protectiveagentsbyabsorbing the potentially harmful light energy or by quenching singlet oxygen and trapping free peroxyl radicals.

WaterActivity (A_w)

Wateractivityofanymaterialisveryimportanttoavoidspoilageandtoextendshelflife. In the case of bran also, it is important to know the water activity to avoidhydrolysis. When A_w below 0.30 reaches primary adsorption zone, where watermolecules linked to carboxyl (COOH) group, which links to other molecules byhydrogen bonds, this water layer would not dissolve the food components, but itwould cover the food, which could lead to an acceleration of lipid oxidation. Theraw bran had A_w around 0.54 ± 0.03, and upon dry heat stabilization process, itreduces to 0.25 ± 0.0 (Garcia et al. 2012).

ParticleSize

Theparticlesizeofthebranvariesaccordingtothetypeofpolisherusedtoremovebranfromricek ernelandtheprocessingconditionswhichareusedforstabilization.The commercially available rice brans are sieved in different mesh sizes to get theproper particle size for specific application. For example, to get maximum oilyield after stabilization, the pellets will be formed so coarse bran is needed, and touseinfoodsystems, fineflouris required(Table1).

Bulk Density

Thebulkdensityofstabilizedbransignificantly improvewhencomparedtorawbranof0.500g/ml.Increaseinbulkdensityafterstabilizationisobservedt oincreasepercolation rate of the solvent during oil extraction and thus, better oil yield (Sharma et al. 2004). After extraction of oil, the bulk density oftendecrease as seen on the bulk density of different stabilized bran given inTable 2.

Table1 Ricebranparticlesizedistribution,%

Mesh	Particlesize(μm)	Rawbran	Moist heat stabilized bran
18	>1000	0	0
18–30	1000–595	2.4	18.6
30–50	595–297	30.0	32.7
50–80	297–177	12.2	18.5
80–100	177–149	8.5	10.8
<100	<149	46.7	19.4

Luh et al. (1991)

Table2 Functionalpropertiesofricebran

	Bulk density	Waterabsorption	Watersolubility	Fatabsorption

	(g/ml)	(g/100g)	(g/100g)	(g/100g)
Rawbran	0.500	143.68	7.57	96.32
Commercially defattedrice bran	0.340	240.00	–	210.00
Extruded stabilized bran	0.593	170.93	9.33	72.80
Dry heat stabilized bran	0.548	156.91	9.34	81.28

Sairam et al. (2011), Sharma et al. (2004)

FunctionalProperties

Toincorporatebraninanyfood application,thereisaneedtounderstanditsfunctionalcharacteristicssuch as water absorption capacity, water solubility, and fat absorption capacity. Thesefunctional properties will vary according to the bran source and processing conditions.Branisrichinproteinandfiber;withfiberhavinghighwater-holdingcapacity,hencebranwillabsorbthemaximumamountofwateraround143.68ml/100g,andasstabilizedricebran,themoisturecontentislessthanthatoftherawbran,andwaterabsorptioncapacityisincreased.Alsoindefattedbran,thewaterabsorptionismoreduetochangeintheporosityofthebran.During branheatstabilization,the starch molecules absorb water and get solubilized which cause water solubilityofthebran.Duringheattreatment,thereisachangeinthebedporosityandporesize of the bran due to the rupture of fatty vacuoles, and fat tends to leach out fromthetissue, which causes decrease in fat absorption.

Because of the high water-holding capacity of rice bran, it maintains moistureandfreshnessinbakedfoods.Rawricebrancanmaintainstabilityinemulsifiedlayeroffoodevenafter30minofheating.Itshowsthepossibilityofusingbranasfatemulsifiersinfoods(Tangetal.2003).Thefoamingcapacityaidsairincorporation, leavening and maintaining texture in baked foods and whipped toppings.Defatted rice bran absorbs more water and has good foaming capacity and stabilityin food. Raw rice bran has high foaming capacity as well as emulsification. Theextrudedricebranwith115.5%foamingvaluecouldbethebestbranforuseinfoodsystems(Table .3).

ChemicalProperties

Chemical properties include proximate and antioxidant properties of the bran. Foranyfoodmaterial,weneedtounderstandthecompositionand quantity per serving for improvement in human health. As indicatedearlier bran is a rich source of protein, fiber, and fat and other nutrients, havingunique antioxidants like oryzanol and tocopherol, which makes bran more interestingandpassionatetoincorporateintohuman food and animal feed compositions.

Table.3 Emulsificationandfoamingpercentageofbran

	Emulsification(%)	Foaming(%)
Rawricebran	46	9
Microwave-stabilizedbran	41	5
Extrusion stabilized bran	22	4

Tangetal.(2003)

The nutrients are distributed throughoutthealeuronelayerinvaryingquantity.Thenutrientcontentvariesaccordingtothesourceofbranandprocessingconditions;stabilizedbrancanretain maximum nutrients and the percentage of fat also increases. The bran is alsoa richsourceofsilicaduetothepresenceofricehullfragments.Commercialbranisafine,flourymaterialmadeupoftheouterlayerofbrownriceandpulverizedgermwith some hull fragments and some endosperm (Orthoefer,2005). The particle sizedistribution varies according to polishing percentage, i.e., the degree of milling,millingconditions,andtypeofmilling.Accordingtotheseconditions,thecompositionofbranalsovaries.Generally,alowdegreeofmillingispracticed.

The amounts of starch and other nutrients in the bran depend on the degree ofmilling and extent of kernel endosperm breakage during milling. Whitening is theprocess in which brown rice is subjected for removal of rice bran and germ to produce whiterice through application of pressure to grain which generates heat, causing cracking and broken rice. This results in less yield of rice paddy. Therefore, to avoid overheating of grains,three to four whitening machines are provided which are collectively known as amulti-breaksystem.Themulti-breaksystemwillprovidetherestingtimewhilemilling,tominimizeheatproductionduetofrictioninpolishingandalso serve as a port to the separation of bran from thegrain (Yilmaz 2016). The amount of bran removed from the whitening machine isknown as bran fraction of that particular multi-break pass, and mixture of outputsfromthesepasses isknown ascomposite ricebran (Table4).

Nutrients are not uniformly distributed throughout the bran layers, and theremoval of bran is due to friction and abrasion of the kernel and the surface ofpolisher. The removal of bran is not layer by layer; abrasion will occur in a non-uniformpattern which may remove endosperm layer along with the bran. In some portion, the bran will not be removed completely as it may have attached to the kernel, and insomeotherportion,therewillbecompleteremovalofbran,whichcausesvariations incompositions according to polisher and degree of milling.

The stabilization techniques change the properties of bran,and it retains the maximum amount of nutrients by inactivating the rancid-causingenzymes especially lipases. The proximate composition of different brans is giveninTable5.Oilqualityisimprovedbyinactivatingenzyme,increasingshelf-stability of the oil by reducing FFA of the bran. The FFA of raw bran is more ascompared to the stabilized one over the period of bran storage. The high amount ofFFAindicatesthe branrancidity,and itisno longerableto addvalue tohumandiet.

Rice bran contains 15–20% oil with three main fatty acids, i.e., palmitic acid(12–18%),oleicacid(40–50%),andlinoleicacid(30–35%).Thebranisarich

Table4 Compositionofricebranduringdegreeofmilling

	Degreemilling(%)	Protein	Fat	Fiber	Ash
1st cone	0–3	17.0	17.7	10.5	9.8
2nd cone	3–6	17.6	17.5	10.3	9.4
3rd cone	6–9	17.0	16.5	5.7	8.4
4th cone	9–10	16.7	14.2	5.7	7.5

Danforth and Orthoefer (1989)

Table.5 Theproximatecompositionofdifferentbrang/100g

	Rawbran	CDRB	Microwave
Moisture	11.23	11.1	8.4
Protein	14.63	17.2	17.5
Fat	16.4	0.66	17.5
Totaldietaryfiber	24.58	11.44	28.98
Ash	7.4	14.65	7.6
Carbohydrate	51.7	43.6	48.9

Sairam et al. (2011)

**CDRB*:commercially defatted ricebran

source of minerals also like iron, calcium, and phosphorus. As told earlier the branhasunique compounds like oryzanol and tocopherol (Orthoefer2005).

Thericebrancontainsauniquecomplexofnaturallyoccurringantioxidantcompoundslikeoryzanols,tocopherols,tocotrienols,andphytosterolsgroups.Ricebranoilcontainsahighnumberofantioxidantsliketocopherolapproximately1.0%(v/v)compared to other oil seeds and tocotrienol around 1.7% (v/v). Crude rice bran oilcontains ≤2% (v/v) of oryzanol that is a mixture of sterol esters of ferulic acid(Lloyd et al. 2000). The concentration of these antioxidants also depends on thedegreeofmillingasmillingprogressesthesecompoundsgotreducedandalsostabilizationtechnique which isused to inactivate lipolyticenzymes.

Application of Rice Bran in Food Industry

Rice bran is finding enormous applications in food industries for increasing thenutritional quality of processed foods. Rice bran being high in dietary fiber and inview of its therapeutic potential, its addition can contribute to the development ofvalue-addedfoodsorfunctionalfoodsthatcurrentlyareinhighdemand.Supplementation of rice bran has been successfully carried in various foods likebread,cakes,noodles,pasta,andicecreamswithoutsignificantlyaffectingthefunctionaland textural properties.

Theprimaryuseofricebranasanadditiveinfoodisduetoitshighfibercontent,whichmildlypromotesstoolregularity.Fromamarketingpoint,themostcommonlyavailable rice bran-derived product is the oil (Prasad et al. 2012). Rice bran oil hasanimpressivenutritionalqualitythatmakesitsuitablefornutraceuticalproducts.

When 5% rice bran was incorporated in pizza dough it was found to be stable for 60 days at -18^{o}C.Rice bran was incorporated in pasta in other to see its effect on the textural and antioxidant properties the pasta supplemented with rice bran was still acceptable for up to 4 months of storage. Similarly, Infrared stabilized rice bran was used in replacing wheat flour in bread in other to test the effect on B Vitamins and Minerals.It was observed to improve the amount of B Vitamins and minerals, especially niacin and phytic acid.Incorporating rice bran in pork meatballs at less than 10% was found to give a good textural property. Supplementing cookies with rice bran improved the dietary fiber content and mineral profile

Materials and Methods

Pre-treatment of rice bran

The fresh rice bran was dried at 60 ^{o}C in a forced-air oven and screened through a 40-mesh sieve. Defatting was conducted in duplicate by soaking the rice bran in n-hexane (1:5, w/v) at room temperature for 12 hours and then decanting the n-hexane. The defatted rice bran was first

air dried in a fume hood to remove residual hexane and then dried at 60 ^{o}C in a forced-air oven and kept in sealed bags for further use

Chemical composition analysis of rice bran
The proximate composition of the rice bran was determined using standard method of AOAC (2005)

Moisture content
The moisture content was determined by drying in an oven at 105 ^{o}C until constant weight was attained

Crude fat determination
The crude fat content of the rice bran will be determined using Soxhlet extraction method using petroleum ether as the extracting solvent. 5g of rice bran will be placed in the thimble and the thimble placed in the Soxhlet extractor. 90ml of the solvent (petroleum ether) will be placed in a round bottom flask and the whole setting will be placed on a heating mantle and the petroleum ether was allowed to boil this extraction will be allowed to continue for a period of 6 hours. All the solvent was collected after distillation. The sample was placed in an oven after which it was transferred to a desiccator. The weight of the sample is then taken representing the weight of the defatted sample.
Calculation
Weight of empty thimble = w1
Weight of thimble with sample =w2
Weight of sample = p
Percentage of crude fat = (w2 – w1)/p X 100

Protein determination
The protein content was determined using the Kjeldahl method.In this method 1g of the pretreated rice bran was added to kjeldahl salt and digested in concentrated sulphuric acid and the total organic nitrogen converted to ammonium sulphate. Ammonia is formed and distilled into boric acid solution under alkaline conditions. The borate anions formed was titrated with standardized hydrochloric acid.Thereafter, the nitrogen content calculated to represent the amount of crude protein in the sample. Since most proteins contain 16% of nitrogen thus a conversion factor of 6.25 was used
The pretreated rice bran (1g) was placed in a Kjeldahl flask. 10ml concentrated sulphuric acid and 5 g Kjeldahl salt as a catalyst, the destruction process carried out in a hood to clear green liquid.10ml of the cold solution with 10ml of 10% sodium hydroxide was distillated for 20 minutes and the distillate collected in Erlenmeyer containing 25ml 0.1N HCl. The excess HCl was titrated with 0.1N NaOH using indicator Tashiro.

Determination of the ash content of rice bran
Ash is a measure of total amount of inorganic substances, including trace and major minerals (Calcium, Potassium, Manganese etc.) as well as toxic substances (Lead, Mercury, Cadmium etc.)
After all the fat and lipid had been removed by the soxhlet extraction, 100g of the residue was washed with boiled distilled water until the filtrate is acid-free. Washed with 100ml 0.313 N boiled NaOH and reflux for 30 minutes. The residue was filtered with Buchner funnel and

washed with K_2SO_4 10%, distilled water and ethanol 95%, respectively. The residue was transferred to weight porcelain container and dried at 105 ^oC and cooled in desiccator and weighed.

Determination of the trace element contents using atomic absorption spectroscopy

When a solution is aspirated into a flame the heat of the flame will cause the solution to evaporate. The microcrystals remaining are partially or wholly decomposed into elements in the gaseous form (atomization). Some of these atoms of metal are of a particular energy of a characteristic wavelength and becomes excited to a higher electronic level or, they may absorb energy from the flame and become thermally excited. The atom lose their excitation energy either as heat by collision with other atoms, or as radiation of a characteristic wavelength as the electron returns to a lower excited state or to the ground state

The term atomic absorption refers to the absorption of energy from a light source with a consequent decrease in the radiant power transmitted through the flame. measurement of this absorption corresponds to atomic absorption spectroscopy.

Atomic absorption spectroscopy was used for the determination of the trace element content of the rice bran. AAS will be used for the determination of the amount of Calcium, Sodium, Iron, Magnesium, Iron, Potassium, Copper, Cadmium and Lead that are present in the rice bran.

RESULTS AND DISCUSSION

DETERMINATION OF MOISTURE CONTENT OF RICE BRAN

The moisture content is a measure of the percentage moisture lost due to drying at a temperature of 105 C according to Udo and Oguwede (1986) method, specified weight of the pretreated rice bran was weighed (w1) into pre weighed crucibles (w0) and placed into a hot drying oven at 105 C for three hours , the crucibles were removed , cooled in desiccators and weighed. The process of drying, cooling and weighing were repeated until a constant weight (w2) was obtained.

The weight loss due to moisture was obtained by the equation

$$\text{Moisture (\%)} = \frac{w1 - w2}{w1 - w0} \times 100$$

Where : w0 is weight of the empty crucible in grams

W1 is weight of the empty crucible + sample in grams

W2 is weight of the dried sample + empty crucible in grams

The moisture content was carried out using duplicate samples and the average of both was taken

s/n	W0	W1	W2	% moisture	Average % moisture
Rice bran Sample A	0.42817	3.4637	3.3280	4.4712	
Rice bran Sample B	0.4513	3.4754	3.3485	4.1963	4.3338

Sample A

Moisture Content = $\frac{3.4637 - 3.3280}{3.4637 - 0.4287} \times 100$

$= \frac{0.1357}{3.0350} \times 100$

$= \frac{13.5700}{3.0350}$

$= 4.4712$

Sample B

Moisture Content = $\frac{3.4754 - 3.3485}{3.4754 - 0.4513} \times 100$

$= \frac{0.1269}{3.0241} \times 100$

$= \frac{12.6900}{3.0241}$

$= 4.1963$

Average Moisture Content = $\frac{4.4712 + 4.1963}{2}$
Of pretreated Rice Bran

$= \frac{8.6675}{2}$

$= 4.3338$

DETERMINATION OF ASH CONTENT OF RICE BRAN

This is a measure of the residue remaining after combustion of the dried sample in a furnace at a temperature of 550 C for 5 hours according to James (1995). 2g of the pretreated rice bran was weighed (w1) into pre weighed empty crucible (w0) and placed into a Lento Muffle furnace at 550 C for 5 hours. The ash was cooled in a desiccator and weighed (w2). The weight of the ash was determined by the difference between the pretreated rice bran, pre weighed and the ash in the crucible

Percentage Ash was obtained by the equation

Ash (%) = $\frac{w2 - w0}{w1 - w0} \times 100$

Where : w0 is weight of the empty crucible in grams
W1 is weight of the dried sample + empty crucible in grams
W2 is weight of the ashed sample + empty crucible in grams

The ash content was carried out using duplicate samples and the average of both was taken

s/n	W0	W1	W2	% moisture	Average % moisture
Rice bran Sample A	22.4101	23.5631	22.6362	19.6097	
Rice bran	25.4367	26.6884	25.6622	18.0155	18.8126

Sample B

$$\text{Ash Content of Sample A} = \frac{22.6362 - 22.4101}{23.5631 - 22.4101} \times 100$$

$$= \frac{0.2261}{1.1530} \times 100$$

$$= \frac{22.6100}{1.1530}$$

$$= 19.6097\%$$

$$\text{Ash Content of Sample B} = \frac{25.6622 - 25.4367}{26.6884 - 25.4367} \times 100$$

$$= \frac{0.2255}{1.2517} \times 100$$

$$= \frac{22.5500}{1.1530}$$

$$= 18.0155\%$$

$$\text{Average Ash Content of Rice Bran} = \frac{19.6097 + 18.0155}{2}$$

$$= 18.8126\%$$

DETERMINATION OF CRUDE FIBRE CONTENT OF RICE BRAN

The percentage of crude fibre was determined by the method of Ude and Oguwele (1986). 2g of the pretreated rice bran was weighed (w1) into 1 dm3 conical flask water (100ml) and (20ml) of 20% sulphuric acid were added and boiled gently for 30 minutes . the content was filtered through whatman filter paper. The residue was scrapped back into the flask with a spatula water (100ml) and 20ml of 10% sodium hydroxide were added and allowed to boil gently for 30 minutes. The content was filtered, and the residue was washed thoroughly with hot distilled water then rinsed once with 10% hydrochloric acid and twice with ethanol and finally three times with petroleum ether. It was allowed to dry and scrapped into crucible and allowed to dry overnight at 105 C in an air oven, it was then removed and cooled in a desiccator the sample was weighed (w1) and dried at 550 C for 90 minutes in a Lento Muffle furnace, it was finally cooled in a dessicator and weighed again (w2) . The percentage crude fibre was calculated using the equation.

$$\text{Crude fibre (\%)} = \frac{w1 - w2}{w} \times 100$$

Where, w is weight of the sample in grams
w1 is weight of the dried sample = wa – w0
w2 is weight of the ashed sample = wb – w0
w0 is weight of the empty crucible
The crude fiber content was carried out using duplicate samples and the average of both was taken

s/n	W0	Wa	Wb	% moisture	Average % moisture
Rice bran Sample A	16.2308	16.5855	16,2545	16.5500	
Rice bran Sample B	17.8418	18.2077	17.8840	16.1850	16.3675

Crude Fibre Content of Sample A
Wo = 2g
W1 = 16.5855 – 16.2308
= 0.3547
W2 = 16.2545 – 16.2308
= 0.0237

$$\text{Crude Fibre \% Sample A} = \frac{0.3547 - 0.0237}{2} \times 100$$
$$= \frac{0.3310}{2} \times 100$$
$$= \frac{33.1000}{2}$$
$$= 16.5500$$

Crude Fibre Content of Sample B
Wo = 2g
W1 = 18.2077 – 17.8418
= 0.3659
W2 = 17.8840 – 17.8418
= 0.0422

$$\text{Crude Fibre \% Sample A} = \frac{0.3659 - 0.0422}{2} \times 100$$
$$= \frac{0.3237}{2} \times 100$$
$$= \frac{32.3700}{2}$$
$$= 16.1850$$

$$\text{Average Crude Fibre \% of Rice Bran} = \frac{16.5500 + 16.1850}{2}$$

= 16.3675%

DETERMINATION OF CRUDE PROTEIN CONTENT OF RICE BRAN

The crude protein content of the rice bran was determined using the micro Kjeldahlmetod described by AOC (1990). The principle of the method is based on the transformation of protein and that of the other nitrogen containing organic compounds, other than nitrite and nitrates into ammonium sulphate by acid digestion.

Simple Nitrogen + H_2SO_4 (aq) --------- $(NH4)_2SO_4$ (aq)

$(NH_4)_2SO_4$ (aq) + 2NaOH (aq) ……. $2NH_3$ (aq) + $2H_2O$ + Na_2SO_4 (aq)

NH_3 (aq) + H_3BO_3 (aq) …… NH_4 (aq) + H_2BO_3 (aq)

H (aq) + H_2BO_3 (aq) ……. H_3BO_3

The Kjeldahl salt was prepared by dissolving 23g of sodium sulphate with 4g of copper II sulphate

To 1g of the pre-treated rice bran was added Kjeldahl catalyst and 10ml of concentrated sulphuric acid in a flask this was heated for 4 hours until it was completely digested and the colour turned clear green to the digested product was added distilled water to the 100ml marks.

DISTILLATION

To 20ml of the digested product was added 20ml of 40% NaOH and was distilled over 5ml of 4% boric acid with three drops of Tachero indicator (methylene blue, methyl red in ethanol). The product of the distillation with boric acid was titrated with 0.1M HCl.

$$N\% = \frac{(Vx - Vb) \times Nacid \times 0.01401}{W \times \text{volume used}} \times 100$$

Where Vx – Vb = titre value = 3.30ml

Nacid = 0.1M

W = 1g

$$N\% = \frac{3.30 \times 0.1 \times 0.01401}{1 \times 0.2} \times 100$$

$$= 0.0046233 \times 100$$

$$N\% = \frac{0.4623}{0.2}$$

$$= 2.3115$$

% Protein = N% X 6.25

= 2.3115 X 6.25

= 14.4469%

DETERMINATION OF CRUDE FAT OT THE RICE BRAN

The crude lipid content of the rice bran was determined using Soxhlet extraction procedure described by Udo and Oguwele(1986). Specified weight of the fresh rice bran (w0) was placed into a porous thimble and covered with a clean white cotton wool, 200ml of n – hexane was poured into a 250ml extraction flask which was previously dried in the oven at 105 C and weighed (w1) . The porous thimble was placed into the soxhlet and the rest of the apparatus was assembled. Extraction was done for 6 hours. the thimble was removed carefully, and the extraction flask placed in a water bath so as to evaporate the n- hexane and then dried in the oven at a temperature of 105 C to completely free the solvent and the moisture. It was cooled in a dessicator and reweighed (w2).
The percentage crude lipid was calculated using the equation below.

$$\text{Crude lipid (\%)} = \frac{w1 - w2}{W0} \times 100$$

Where : w0 is weight of the sample in grams
w1 is weight of the sample + thimble before extraction
w2 is weight of the dried sample + thimble after extraction

The crude lipid content was carried out using duplicate samples and the average of both was taken

s/n	W0	W1	W2	% lipid	Average % lipid
Rice bran Sample A	1.5619	2.3852	2.0755	19.8284	
Rice bran Sample B	1.5456	2.3542	2.0535	19.4552	19.6418

Crude lipid % Content of Sample A
Wo = 1.5619
W1 = 2.3852
W2 = 2.0755

$$\text{Crude lipid \% Sample A} = \frac{2.3852 - 2.0755}{1.5619} \times 100$$
$$= \frac{0.3077}{1.5619} \times 100$$
$$= \frac{30.9700}{1.5619}$$
$$= 19.8284\%$$

Crude lipid % Content of Sample A
Wo = 1.5456
W1 = 2.3542
W2 = 2.0535

Crude Fibre % Sample B = $\frac{2.3542 - 2.0535}{1.5456} \times 100$

$= \frac{0.3007}{1.5456} \times 100$

$= \frac{30.0700}{1.5456}$

=19.4552

Average Crude lipid % of Rice Bran = $\frac{19.8284 + 19.4552}{2}$

$= \frac{39.2836}{2}$

=19.6418%

DETERMINATION OF CARBOHYDRATE CONTENT OF RICE BRAN

The percentage is obtained by subtracting the other nutrients, ash and moisture from 100

% Carbohydrates = 100% - (% Crude fat + % protein + % crude fibre + % Ash + % Moisture)

= 100% - (19.6418 + 14.4469 + 16.3675 + 18 .8126 + 4.3338)

= 100% - 73.6026

% Carbohydrates = 26.3974

SUMMARY OF PROXIMATE ANALYSIS OF RICE BRAN

Crude Fat	=	19.6418
Crude Protein	=	14.4469
Crude Fibre	=	16.3675
Ash	=	18.8126
Moisture	=	4.3338
Carbohydrate	=	26.3974

ATOMIC ABSORPTION SPECTROSCOPY FOR THE DETERMINATION OF THE TRACE ELEMENT CONTENT OF RICE BRAN

1g of the rice bran was weighed into a conical flask 20ml of nitric acid was added in a fume cupboard . the flask was placed in a hot plate set at 70 C and digestion was carried out until the fume clears after 4 hours. The content was filtered into a 100ml standard volumetric flask. This was then transferred into sample bottles for AAS analysis.

Calculation

Concentration of metal ion in mg/kg = $\frac{\text{Conc} - \text{Blanc}}{\text{Wt. of Sample}} \times 100$

Calcium Content = $\frac{6.6419 - 0.2439}{1} \times 100$

= 6.398 x 100
= 639.8mg/kg
= 0.6398mg/g

Magnesium Content = $\frac{22.7700 - 0.0638}{1} \times 100$
= 22.7062 x 100
= 2270.6200mg/kg
= 2.2706mg/g

Potassium Content = $\frac{44.1303 - 0.0407}{1} \times 100$
= 44.0896 x 100
= 4408.9600mg/kg

= 44.0896mg/g

Sodium Content = $\frac{2.2177 - - 3.2671}{1} \times 100 = \frac{2.2177 + 3.2671}{1} \times 100$
= 5.4848 x 100
= 548.4800mg/kg
= 0.5485mg/g

Iron Content = $\frac{9.1112 - 0.2549}{1} \times 100$
= 8.8563 x 100
= 885.63mg/kg
= 0.8856mg/g

Copper Content = $\frac{0.2282 - 0.0337}{1} \times 100$
= 0.1945 x 100
= 19.4500mg/kg
= 0.0195mg/g

Cadmium Content = Below detection limit

Lead Content = $\frac{0.0919 - 0.0117}{1} \times 100$
= 0.0802 x 100
= 8.0200mg/kg
= 0.8020mg/100g

PROXIMATE ANALYSIS OF COMMERCIALLY AVAILABLE POULTRY FEEDS (ULTIMA FEED) MANUFACTURED BY CROWN FEEDS LIMITED

DETERMINATION OF MOISTURE CONTENT OF POULTRY FEED

The moisture content is a measure of the percentage moisture lost due to drying at a temperature of 105 C according to Udo and Oguwede(1986) method, specified weight of the poultry feed (

Starter, Grower and Finisher) was weighed (w1) into pre weighed crucibles (w0) and placed into a hot drying oven at 105 C for three hours , the crucibles were removed , cooled in desiccators and weighed. The process of drying, cooling and weighing were repeated until a constant weight
(w2) was obtained.
The weight loss due to moisture was obtained by the equation

$$\text{Moisture (\%)} = \frac{w1 - w2}{w1 - w0} \times 100$$

Where, w0 is weight of the empty crucible in grams
W1 is weight of the empty crucible + sample in grams
W2 is weight of the dried sample + empty crucible in grams
The moisture content was carried out using duplicate samples and the average of both was taken

s/n	W0	W1	W2	% moisture	Average % moisture
Starter Sample A	0.3455	2.4310	2.3397	4.3778	
Starter Sample B	0.2758	2.5286	2.3671	6.9829	5.6804
Grower Sample A	0.1782	2.3335	2.1558	8.2448	
Grower Sample B	0.1920	2.5723	2.3785	8.1418	8.1933
Finisher Sample A	0.2407	2.5014	2.2827	9.6740	
Finisher Sample B	0.1657	2.3035	2.1488	7.2364	8.4552

Starter Sample A

$$\text{Moisture Content} = \frac{2.4310 - 2.3397}{2.4310 - 0.3455} \times 100$$

$$= \frac{0.0913}{2.0855} \times 100$$

$$= \frac{9.1300}{2.0855}$$

$$= 4.3778\ \%$$

Starter Sample B

$$\text{Moisture Content} = \frac{2.5286 - 2.3671}{2.5286 - 0.2158} \times 100$$

$$= \frac{0.1615}{2.3128} \times 100$$

$$= \frac{16.1500}{2.3128}$$

= 6.9828 %

Average Moisture Content = $\frac{4.3778 + 6.9828}{2}$
Starter Meal

= $\frac{11.3607}{2}$

= 5.6804 %

Grower Sample A
Moisture Content = $\frac{2.3335 - 2.1558}{2.3335 - 0.1782} \times 100$

= $\frac{0.1777}{2.1553} \times 100$
= $\frac{17.7700}{2.1553}$

= 8.2448 %
Grower Sample B

Moisture Content = $\frac{2.5723 - 2.3785}{2.5723 - 0.1920} \times 100$
= $\frac{0.1938}{2.3803} \times 100$
= $\frac{19.3800}{2.3803}$

= 8.1418 %

Average Moisture Content = $\frac{8.2448 + 8.1418}{2}$
Grower Meal

= $\frac{16.3866}{2}$

= 8.1933 %

Finisher Sample A
Moisture Content = $\frac{2.5014 - 2.2827}{2.5014 - 0.2407} \times 100$

$= \frac{0.2187}{2.2607} \times 100$

$= \frac{21.8700}{2.1553}$

$= 9.6740\ \%$

Finisher Sample B

$\text{Moisture Content} = \frac{2.3035 - 2.1488}{2.3035 - 0.1657} \times 100$

$= \frac{0.1547}{2.1378} \times 100$

$= \frac{15.4700}{2.1378}$

$= 7.2364\ \%$

$\text{Average Moisture Content Finisher Meal} = \frac{9.6740 + 7.2364}{2}$

$= \frac{16.9104}{2}$

$= 8.4552\ \%$

DETERMINATION OF ASH CONTENT OF POULTRY FEED

This is a measure of the residue remaining after combustion of the dried sample in a furnace at a temperature of 550 C for 5 hours according to James (1995). 2g of the poultry feed was weighed (w1) into pre weighed empty crucible (w0) and placed into a Lento Muffle furnace at 550 C for 5 hours. The ash was cooled in a desiccator and weighed (w2). The weight of the ash was determined by the difference between the poultry feeds, pre weighed and the ash in the crucible Percentage Ash was obtained by the equation

$\text{Ash}\ (\%) = \frac{w2 - w0}{w1 - w0} \times 100$

Where : w0 is weight of the empty crucible in grams

W1 is weight of the dried sample + empty crucible in grams

W2 is weight of the ashed sample + empty crucible in grams

The ash content was carried out using duplicate samples and the average of both was taken

s/n	W0	W1	W2	% moisture	Average % moisture
Starter Sample A	20.0547	21.1321	20.1668	10.4047	
Starter Sample B	18.6838	19.8270	18.8035	10.4706	10.4377
Grower Sample A	15.5247	16.7682	15.6013	6.1600	

Grower Sample B	28.8740	29.9996	28.9480	6.5743	6.3672
Finisher Sample A	19.7184	21.0083	19.7777	4.5937	
Finisher Sample B	12.5734	13.7619	12.6255	4.3837	4.4905

Starter Sample A

Ash Content = $\frac{20.1668 - 20.0547}{21.1321 - 20.0547} \times 100$

= $\frac{0.1121}{1.0774} \times 100$

= $\frac{11.2100}{1.0774}$

= 10.4047 %

Starter Sample B

Ash Content = $\frac{18.8035 - 18.6838}{19.8270 - 18.6838} \times 100$

= $\frac{0.1197}{1.1432} \times 100$

= $\frac{11.9700}{1.1432}$

= 10.4706 %

Average % Ash Content = $\frac{10.4047 + 10.4706}{2}$
Starter Meal

= $\frac{20.8753}{2}$

= 10.4377 %

Grower Sample A

Ash Content = $\frac{15.6013 - 15.5247}{16.7682 - 15.5247} \times 100$

= $\frac{0.0766}{1.2435} \times 100$

= $\frac{7.6600}{1.2435}$

= 6.1600 %

Grower Sample B

Ash Content = $\frac{28.9480 - 28.8740}{29.9996 - 28.8740} \times 100$

= $\frac{0.0740}{1.1256} \times 100$

= $\frac{7.4000}{1.1256}$

= 6.5743 %

Average % Ash Content Grower Meal = $\frac{6.1600 + 6.5743}{2}$

= $\frac{12.7343}{2}$

= 6.3672 %

Finisher Sample A

Ash Content = $\frac{19.7777 - 19.7184}{21.0083 - 19.7184}$

= $\frac{0.0593}{1.2899} \times 100$

= $\frac{5.9300}{1.2899}$

= 4.5973 %

Finisher Sample B

Ash Content = $\frac{12.6255 - 12.5734}{13.7619 - 12.5734} \times 100$

= $\frac{0.0521}{1.1885} \times 100$

= $\frac{5.2100}{1.1885}$

= 4.3837 %

Average % Ash Content = $\frac{4.5973 + 4.3837}{}$

Finisher Meal 2

= 8.9810
= 4.4905 %

DETERMINATION OF CRUDE FIBRE CONTENT OF POULTRY FEED

The percentage of crude fibre was determined by the method of Ude and Oguwele (1986). 2g of the poultry feed was weighed (w1) into 1 dm3 conical flask water (100ml) and (20ml) of 20% sulphuric acid were added and boiled gently for 30 minutes . the content was filtered through whatman filter paper. The residue was scrapped back into the flask with a spatula water (100ml) and 20ml of 10% sodium hydroxide were added and allowed to boil gently for 30 minutes. The content was filtered, and the residue was washed thoroughly with hot distilled water then rinsed once with 10% hydrochloric acid and twice with ethanol and finally three times with petroleum ether. It was allowed to dry and scrapped into crucible and allowed to dry overnight at 105 C in an air oven , it was then removed and cooled in a desiccator the sample was weighed (w1) and dried at 550 C for 90 minutes in a Lento Muffle furnace, it was finally cooled in a dessicator and weighed again (w2) . The percentage crude fibre was calculated using the equation.

$$\text{Crude fibre (\%)} = \frac{w1 - w2}{w} \times 100$$

Where : w is weight of the sample in grams
w1 is weight of the dried sample = wa – w0
w2 is weight of the ashed sample = wb – w0
w0 is weight of the empty crucible

The crude fibre content was carried out using duplicate samples and the average of both was taken

	W0	W1	W2	% Crude fibre	Average % moisture
Starter Sample A	13.6495	13.7565	13.6530	5.1750	
Starter Sample B		10.1043	10.0354	3.4450	4.3100
Grower Sample A	24.9978	25.0338	25.0017	1.6050	
Grower Sample B	18.1067	18.1533	18.1130	2.0400	1.8225
Finisher Sample A	13.3981	13.4720	13.4154	2.8300	
Finisher Sample B	14.0067	14.0844	14.0153	3.4550	3.1425

Starter Sample A

W1 = wa – w0 = 13.7565 – 13,6495

= 0.1070

W2 = wa – = 13.6530 – 13.6495

= 0.0035

Crude fibre Content = $\frac{0.1070 - 0.0035}{2} \times 100$

= $\frac{0.1035}{2} \times 100$

= $\frac{10.3500}{2}$

= 5.1750 %

Starter Sample B

Crude fibre Content = $\frac{10.1043 - 10.0354}{2} \times 100$

= $\frac{0.0689}{2} \times 100$

= $\frac{6.8900}{2}$

= 3.4450 %

Average Crude fibre Content = $\frac{5.1750 + 3.4450}{2}$

Starter Meal

= $\frac{8.6200}{2}$

= 4.3100

Grower Sample A

Crude Fibre Content = $\frac{25.0338 - 25.0017}{1} \times 100$

= $\frac{0.0321}{1} \times 100$

= $\frac{3.2100}{1}$

= 3.2100 %

Grower Sample B

Crude Fibre Content $= \frac{18.1538 - 18.1130}{1} \times 100$

$= \frac{0.0408}{1} \times 100$

$= \frac{4.0800}{1}$

$= 4.0800\ \%$

Average Crude Fibre Content Grower Meal $= \frac{3.2100 + 4.0800}{1}$

$= \frac{7.2900}{2}$

$= 3.6450\ \%$

Finisher Sample A

Crude Fibre Content $= \frac{13.4720 - 13.4154}{2} \times 100$

$= \frac{0.0566}{2} \times 100$

$= \frac{5.6600}{2}$

$= 2.8300\ \%$

Finisher Sample B

Crude Fibre Content $= \frac{14.0844 - 14.0153}{2} \times 100$

$= \frac{0.0691}{2} \times 100$

$= \frac{6.9100}{2}$

$= 3.4550\ \%$

Average Crude Fibre Content Finisher Meal $= \frac{2.8300 + 3.4550}{2}$

$$= \frac{6.2850}{2}$$

$$= 3.1425\ \%$$

DETERMINATION OF CRUDE PROTEIN CONTENT OF RICE BRAN

The crude protein content of the poultry feed was determined using the micro Kjeldahl method described by AOC (1990). The principle of the method is based on the transformation of protein and that of the other nitrogen containing organic compounds , other than nitrite and nitrates into ammonium sulphate by acid digestion.

Simple Nitrogen + H_2SO_4 (aq) --------- $(NH_4)_2SO_4$ (aq)

$(NH_4)_2SO_4$ (aq) + 2NaOH (aq) ……. $2NH_3$ (aq) + $2H_2O$ + Na_2SO_4 (aq)

NH_3 (aq) + H_3BO_3 (aq) …… NH_4 (aq) + H_2BO_3 (aq)

H (aq) + H_2BO_3 (aq) ……. H_3BO_3

The Kjeldahl salt was prepared by dissolving 23g of sodium sulphate with 4g of copper II sulphate

To 1g of the poultry feed was added Kjeldahl catalyst and 10ml of concentrated sulphuric acid in a flask this was heated for 4 hours until it was completely digested and the colour turned clear green. to the digested product was added distilled water to the 100ml marks.

DISTILLATION

To 20ml of the digested product was added 20ml of 40% NaOH and was distilled over 5ml of 4% boric acid with three drops of Tachero indicator (methylene blue, methyl red in ethanol). The product of the distillation with boric acid was titrated with 0.1M HCl.

$$N\% = \frac{(Vx - Vb) \times N_{acid} \times 0.01401}{W} \times 100$$

Where Vx – Vb = titre value

N_{acid} = 0.1M

FOR STARTER MEAL

W = 1g

N acid = 0.1M

Vx – Vb = 6.0 ml

$$N\% = \frac{6.0 \times 0.1 \times 0.01401}{0.2} \times 100$$

$$= \frac{0.008406 \times 100}{0.2}$$

$$N\% = \frac{0.8406}{0.2} = \frac{8.4060}{2}$$

= 4.2030

% Protein = N% X 6.25

= 4.2030 X 6.25

= 26.2688 %

FOR GROWER MEAL

W = 1g

N acid = 0.1M

Vx – Vb = 6.3 ml

$$N\% = \frac{6.3 \times 0.1 \times 0.01401}{0.2} \times 100$$

$$N\% = \frac{0.008826 \times 100}{0.2}$$

$$N\% = \frac{0.8826}{0.2} = \frac{8.8260}{2} = 4.4130$$

% Protein = N% X 6.25

= 4.4130 X 6.25

= 27.5813 %

FOR FINISHER MEAL

W = 1g

N acid = 0.1M

Vx – Vb = 5.0 ml

$$N\% = \frac{5.0 \times 0.1 \times 0.01401}{0.2} \times 100$$

$$N\% = \frac{0.007005 \times 100}{0.2}$$

$$N\% = \frac{0.7005}{0.2} = \frac{7.0050}{2} = 3.5025$$

% Protein = N% X 6.25

= 3.5025 X 6.25

DETERMINATION OF CRUDE FAT OF POULTRY FEED

The crude lipid content of the poultry feed was determined using Soxhlet extraction procedure described by Udo and Oguwele(1986). Specified weight of the poultry field (w0) was placed into a porous thimble and covered with a clean white cotton wool, 200ml of n – hexane was poured into a 250ml extraction flask which was previously dried in the oven at 105 C and weighed (w1) . The porous thimble was placed into the soxhlet and the rest of the apparatus was assembled. Extraction was done for 6 hours .the thimble was removed carefully and the extraction flask placed in a water bath so as to evaporate the n- hexane and then dried in the oven at a temperature of 105 C to completely free the solvent and the moisture. It was cooled in a dessicator and reweighed (w2).

The percentage crude lipid was calculated using the equation below.

$$\text{Crude lipid (\%)} = \frac{w1 - w2}{W0} \times 100$$

Where : w0 is weight of the sample in grams
w1 is weight of the sample + thimble before extraction
w2 is weight of the dried sample + thimble after extraction

The crude lipid content was carried out using duplicate samples and the average of both was taken

s/n	W0	W1	W2	% Crude Lipid	Average % Crude lipid
Starter Sample A	0.6471	1.6816	1.6030	12.1465	
Starter Sample B	0.5166	1.6508	1.5892	11.9242	12.0354
Grower Sample A	0.5201	1.5651	1.5015	12.2284	
Grower Sample B	0.5525	1.5696	1.5005	12.5068	12.3676
Finisher Sample A	0.5924	1.6213	1.5462	12.6604	
Finisher Sample B	0.5956	1.5971	1.5222	12.5756	12.6180

Crude lipid % Content of Starter A
Wo = 0.6471
W1 = 1.6816
W2 = 1.6030

$$\text{Crude lipid \% Starter A} = \frac{1.6816 - 1.6030}{0.6471} \times 100$$
$$= \frac{0.0786}{0.6471} \times 100$$
$$= \frac{7.8600}{0.6471}$$
$$= 12.1465\%$$

Crude lipid % Content of Starter B
Wo = 0.5166
W1 = 1.6508
W2 = 1.5892

$$\text{Crude lipid \% Starter B} = \frac{1.6508 - 1.5892}{0.5166} \times 100$$
$$= \frac{0.0616}{} \times 100$$

$$= \frac{0.5166}{6.1600}$$
$$= \frac{0.5166}{}$$
$$=11.9242$$

Average Crude lipid % of Starter meal = $\frac{12.1465 + 11.9242}{2}$

$$= \frac{24.0707}{2}$$
$$=12.0354\ \%$$

Crude lipid % Content of Grower A

Wo = 0.5201
W1 = 1.5651
W2 = 1.5015

Crude lipid % Grower A = $\frac{1.5651 - 1.5015}{0.5201} \times 100$

$$= \frac{0.0636}{0.5201} \times 100$$
$$= \frac{6.3600}{0.5201}$$
$$=12.2284\ \%$$

Crude lipid % Grower B

Wo = 0.5525
W1 = 1.5696
W2 = 1.5005

Crude lipid % Grower B = $\frac{1.5696 - 1.5005}{0.5525} \times 100$

$$= \frac{0.0691}{0.5525} \times 100$$
$$= \frac{6.9100}{0.5525}$$
$$=12.5068$$

Average Crude lipid % of Grower = $\frac{12.2284 + 12.5068}{2}$

$$= \frac{24.7352}{2}$$
$$=12.3676\ \%$$

Crude lipid % Content of Finisher A
Wo = 0.5924
W1 = 1.6213
W2 = 1.5462

$$\text{Crude lipid \% Finisher A} = \frac{1.6213 - 1.5462}{0.5924} \times 100$$
$$= \frac{0.0750}{0.5924} \times 100$$
$$= \frac{7.5000}{0.5924}$$
$$= 12.6604\ \%$$

Crude lipid % Content of Finisher B
Wo = 0.5956
W1 = 1.5971
W2 = 1.5222

$$\text{Crude Lipid \% Finisher B} = \frac{1.5971 - 1.5222}{0.5956} \times 100$$
$$= \frac{0.0749}{0.5956} \times 100$$
$$= \frac{7.4900}{0.5956}$$
$$= 12.5756$$

$$\text{Average Crude lipid \% of Finisher Meal} = \frac{12.6604 + 12.5756}{2}$$
$$= \frac{24.2360}{2}$$
$$= 12.6180\ \%$$

DETERMINATION OF CARBOHYDRATE CONTENT OF POULTRY FEED
The percentage is obtained by subtracting the other nutrients, ash and moisture from 100
STARTER MEAL
% Carbohydrates = 100% - (% Crude fat + % protein + % crude fibre + % Ash + % Moisture)

= 100% - (12.0354+ 26.2688 + 4.3100 + 10.4377+ 5.6804)
= 100% - 58.7323
% Carbohydrates = 41.2677
GROWER MEAL
% Carbohydrates = 100% - (% Crude fat + % protein + % crude fibre + % Ash + % Moisture)

= 100% - (12.3676 + 27.5813 + 3.6450 + 6.3672 + 8.1933)
= 100% - 58.1544
% Carbohydrates = 41.8456

FINISHER MEAL

% Carbohydrates = 100% - (% Crude fat + % protein + % crude fibre + % Ash + % Moisture)

= 100% - (12.6180 + 21.8906 + 3.1425 + 4.4905 + 8.4552)

= 100% - 50.5968

% Carbohydrates = 49.4032

SUMMARY OF PROXIMATE ANALYSIS OF STARTER MEAL

Crude Fat = 12.0354
Crude Protein = 26.2688
Crude Fibre = 4.3100
Ash = 10.4377
Moisture = 5.6804
Carbohydrate = 41.2677

SUMMARY OF PROXIMATE ANALYSIS OF GROWER MEAL

Crude Fat = 12.3676
Crude Protein = 27.5813
Crude Fibre = 3.6450
Ash = 6.3672
Moisture = 8.1933
Carbohydrate = 41.8456

SUMMARY OF PROXIMATE ANALYSIS OF FINISHER MEAL

Crude Fat = 12.6180
Crude Protein = 21.8906
Crude Fibre = 3.1425
Ash = 4.4905
Moisture = 8.4552
Carbohydrate = 49.4032

COMPARISON OF THE PROXIMATE ANALYSIS OF RICE BRAN AND ULTIMA POULTRY FEED MEALS

	RICE BRAN	STARTER MEAL	GROWER MEAL	FINISHER MEAL
CRUDE FAT	19.6418	12.0354	12.3676	12.6180
CRUDE PROTEIN	14.4469	26.2688	27.5813	21.8906
CRUDE FIBRE	16.3675	4.3100	3.6450	3.1425
ASH	18.8126	10.4377	6.3672	4.4905
MOISTURE %	4.3338	5.6804	8.1933	8.4552
CARBOHYDRATE	26.3900	41.2677	41.8456	49.4032

TEST FOR THE PRESENCE OF ANTI NUTRIENTS

TEST FOR ANTHRAQUINONES

Bomtragers reaction was used detect the presence of anthraquinones Aglycone in the extract. The pretreated rice bran was extracted with methanol and allowed to stand for 1 hour. To 8ml of the extract was added 2ml of 2M hydrochloric acid which was further heated in a hot water bath for 15 minutes.This was then cooled and filtered while the filtrate was then extracted with chloroform. The chloroform layer was separated and shaken with 10% potassium hydroxide solution. The total anthraquinone content was analysed by UV Spectrophotometer at wavelenght of 515nm (Harbone, 1998)
Absorbance = 1.1450
Blanc = 0.0100
From Beers law A = Elc
Where A is the absorbance
E is the molar absorptivity
l is the path length

A = 1.1450 – 0.010
= 0.134 IF E = 1 and l =1
Then A = c = 0.134 mg/kg
=0.0134mg/100g
% anthraquinines = 0.0134mg/100g sample

DETERMINATION OF TOTAL PHENOLIC COMPOUNDS OF RICE BRAN
1g of the sample was weighed and dissolved in 10ml of distilled water, 1ml of this solution was transferred to a test tube, then 0.5ml of 2N FolinCiocalten reagent and 1.5ml of 20% Na2CO3 solution was added and ultimately the volume was made up to 8ml with distilled water followed by vigorous shaking and finally allowed to stand for 2 hours after which the absorbance was taken at 765nm . these data was used to estimate the total phenolic content in the rice bran sample.
Absorbance of sample = 0.3330
Blanc = 0.0370
A = 0.3330 – 0.0370
= 0.296 Since E = 1 and l =1
Then A = c = 0.296 mg/kg
=0.0296mg/100g
% Phenolic compounds = 0.0134mg/100g sample

DETERMINATION OF TOTAL TANNINS
500mg of the sample was weighed into a 50ml plastic bottle, 50ml of distilled water was added and shaken for 1 hour in a mechanical shaker this was then filtered into a 50ml volumetric flask and made up to mark. Then 5ml of the filtrate was pipetted out into a test tube and mixed with 2ml of 0.1m Fecl3 in 0.1M HCl and 0.008M potassium ferrocyanide. The absorbance was measured at 385nm within 10 minutes.
Solution preparation
0.1M ferric chloride in 0.1M HCl was prepared by dissolving 16.221g FeCl3 in 1000ml of 0.1M HCl.
these data was used to estimate the total tannin content in the rice bran sample.
Absorbance of sample = 2.6800

Blanc = 0.0250

A = 2.6800 – 0.0250

= 2.6550 Since E = 1 and l =1

Then A = c = 2.6550 mg/kg

=0.2655mg/100g

Total Tannin compounds = 0.2655mg/100g sample

DETERMINATION OF TOTAL SAPONINS

20g of the pretreated rice was put into a conical flask and 100ml of 20% aqueous ethanol were added , the sample was heated over a hot water bath for 4 hours with continuous stirring at about 55 C the mixture was filtered and the residue re-extracted with another 200ml of 20% ethanol , the combined extracts were reduced to 40ml over a water bath at about 90 C . The concentrated extract was then transferred into a 250ml separating funnel and 2oml of diethyl ether was added to the extract and vigorously shaken. The aqueous layer was discarded and the purification process was repeated. 60ml of n – butyl alcohol was added and the combined n – butyl alcohol extract was washed twice with 100ml of 5% sodium chloride. The remaining solution was then heated in a water bath and after evaporation the sample were dried in the oven to a constant weight. (Harbone 1998).

Weight of empty beaker w0 = 103.2485

Weight of beaker + residue w1 = 104.5379

Weight of residue w1 – w0 = 1.2894

Weight of Saponins =1.2894g in 20g sample

Total Saponin content $= \frac{1.2894 \times 100}{20}$

= 6.4470g/100g sample

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