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Evaluation of tropical feedstuffs used in ruminant animals for carbohydrate and protein fractions by CNCP system

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ABSTRACT

This research was carried out to assess the nutritional content of frequently used 8 cereal grains and 13 agro industrial by-products (AIBP) as per CNCPS along with their *in vitro* digestibility, gas and methane production. It was observed that lignin (% NDF) was lowest in white maize (5.28) and highest in gram khanda (30.93). NPN (%SP) content was highest in gram churi (73.32) and lowest in wheat grains (10.85). Among grains, values (%) for protein fraction P_A, $P_{B1,}$ $P_{B2,}$ P_{B3} and fraction P_C were found to be highest for oat (10.58), followed by wheat (26.85), pearl millet (72.08),barley (17.63) and pearl millet (7.09), respectively while for AIBP, maximum values were observed for gram churi (32.67), followed by beet pulp (56.98), gram khanda (82.61), paddy straw (43.39) and in gram husk (25.07). For AIBP, carbohydrate fractions, C_{B1} content was highest (42.71) in wheat bran and C_A was highest (61.74) in dried distillers grains with solubles (DDGS). It was observed that AIBP had more unavailable carbohydrate fraction (C_C) in comparison to grains. Average values of IVDMD and IVOMD for grains were observed to be 89.78% and 90.80%, and for AIBP 75.19% and 75.23%, respectively. Average gas production (ml/200 mg) was found to be 57.85 for grains and 33.60 for AIBP. It was found that average methane generation (g/kg IVDMD) was comparable for grains and AIBP. Therefore, the study's findings may help to clarify more about the digestibility and gas production of agro industrial by products and cereal grains for more precise and environment friendly feed formulation for dairy animals.

Keywords: Agro industrial by products, Cereal grains, CNCPS, *In vitro* digestibility, Methane production

Growing human population calls for use of agro-industrial by-products along with grains to completely utilise the capacity of livestock production. Currently, there is 47% shortage of protein-rich feedstuffs (NIANP, 2013; Kour et al, 2023), which reinforces case for utilisation of agro-industrial by products along with grains to maintain nation's feed balance. In India, milch animals are being typically fed grasses or crop leftovers, one or two concentrate feed items which are readily available locally ensuing deficiency of proteins, energy, minerals and vitamins (Garg *et al*, 2013, Datt *et al*, 2017). Moreover, anaerobic fermentation, leads to conversion of most of the protein and carbohydrate (CHO) components in feeds to final products like microbial protein, CH_4, CO_2 and volatile fatty acids.

A diet's energy content can be lost by up to 6-12% of gross energy value due to methane emissions from intestinal sources, and it depends upon both the quality and quantity of the diet fed to the animal as it influences how quickly the rumen content is digested and passed to lower tract (Van Soest, 1994). Takahashi (2001) reported that enteric CH_4 formation could be impacted by the type of fermented carbohydrates consumed, including cellulose, hemicelluloses and soluble dietary residues.

The feed's chemical composition, including the protein fractions and CHO, as well as its ingestion and digestibility determines the characterisation of the end products. Reduced generation of CH_4, an increase in propionate and decrease in acetate are the fermentation outcomes of plant low cell wall materials such as grains. (Widiawati and Thalib, 2007; Sharma *et al*, 2013, Sharma *et al*, 2016). The amount of fibre and the rate at which digesta passes are the two main variables which influence the breakdown of roughages. Degradation of DM and NDF varies depending on binding of cellulose and lignin content, intrinsic characteristics and components. Cornell Net Carbohydrate and Protein System

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(CNCPS) groups the feed carbohydrate and proteins into different fractions based ontheir degradability pattern in the rumen.

Further, the escalating environmental activism has strongly integrated life cycle assessment together with livestock operations to reduce enteric emission of greenhouse gases, including methane (CH4) and nutrient excretions from manure (Patra, 2017). Accordingly, methods to reduce these pollutants may help achieve cleaner, more environmentally friendly and sustainable animal production (Eisler *et al.* 2014). Despite this awareness, there aren't many published studies on this topic. To estimate the amount of CH_4 produced *in vivo* by ruminants' digestive systems, data on CH_4 generation from various forages could be obtained using *in vitro* ruminal fermentation. Given the aforementioned details, the primary objectives were established to evaluate the nutritive value of commonly used feedstuffs/agro industrial by products as per CNCPS.

MATERIALS AND METHODS

Sampling sites: The site of experiment was at ICAR-National Dairy Research Institute in Karnal, Haryana, India. Samples for the experiment were gathered from the Institute and neighbouring villages in the same region (29.68°N, 76.98°E).

Collection and processing of samples: Samples of grains and AIBP comprised of maize - *Zea mays* (yellow and white varieties), wheat – *Triticum aestivum*, pearl millet –*Pennisetum typhoides*, sorghum (*Sorghum bicolor*) (White and red varieties), barley-*Hordeum vulgare*, oat – *Avena sativa*, wheat bran –*Triticum aestivum*, rice bran – *Oryza sativa*, de-oiled rice bran (DORB) – *Oryza sativa*, rice polish- *Oryza sativa*, maize gluten meal (MGM)- *Zea mays*, gram churi-*Cicer arietinum*, gram husk-*Cicer arietinum*, gram khanda- *Cicer arietinum*, distillers dried grain solubles (DDGS), wheat straw- *Triticum aestivum*, paddy straw-*Oryza sativa*, sugarbeet or beet pulp-*Beta vulgaris*, and distillery/maize bran *Zea mays* were gathered from neighbouring villages or the National Dairy Research Institute's experimental farm. Samples were oven dried for 48 h at 60°C and ground up in a Wiley mill to pass through a 1 mm screen.

Chemical evaluations: Samples of grains and agro-industrial by products were examined using the AOAC (1995) standard procedures for ash, nitrogen (N), ether extract (EE), and dry matter (DM). Fibre Tech (FibraPlus FES 6, Pelican, Chennai, India) was utilised to calculate the neutral detergent fibre (NDF) in accordance with Van Soest *et al*. (1991) approach. Sodium sulfite and heat-stable alpha amylase were not utilised in NDF calculation. In accordance with AOAC (1997; method 973.18), acid detergent fibre (ADF) was analysed. Both NDF and ADF were stated with residual ash included. Sulfuric acid-mediated cellulose solubilization in the ADF residue allowed for the determination of lignin (sa) (Van Soest *et al*. 1991).

Carbohydrate fractionation: The CNCPS system, which categorises portions of carbohydrates into four categories based on the rates of their degradation, was used to estimate the carbohydrate fractions. Different fractions are C_A (sugars and immediately degradable CHO); C_{B1} (starch and pectins that degrade in an intermediate manner); C_{B2} (plant cell wall that degrades slowly); and C_C (unavailable/ lignin-bound plant cell wall are the others). The calculation of total CHO (grams/kg dry matter) involved deducting the quantities of ash, CP, and EE from 1000. The difference between total CHO and SC was used to estimate non-fiber carbohydrates, while the difference between NDF and NDIP was used to compute structural carbohydrates (SC). To solubilize pigments, lipids, free sugars and waxes, samples were extracted using ethyl alcohol in order to estimate the amount of starch present. In order to measure glucose colorimetrically using standard glucose, the starch-rich residue was treated with anthrone-sulphuric acid after being solubilized with perchloric acid (Sastry *et al*. 1991).

Crude protein fractionation: As per CNCPS sample crude protein was divided into five segments (Sniffen *et al.* 1992). Different segments or fractions are fraction P_A, as (non-protein crude protein, NPCP), which is calculated as the difference between total crude protein and true crude protein precipitated using 0.5 M sulfuric acid and 0.30 M sodium tungstate. The difference between true crude protein and buffer-insoluble crude protein, which is determined using borate phosphate buffer (pH 6.7-8) and newly made sodium azide solution (1 g/10 ml), is known as fraction PB1 (buffer soluble CP, BSP). Acid detergent soluble crude protein (ADSP), or fraction PB3, was established as the difference between neutral detergent insoluble crude protein and acid detergent insoluble crude protein. Neutral detergent soluble crude protein (NDSP), or fraction PB2, was calculated as buffer insoluble crude protein minus neutral detergent insoluble crude protein. It is believed that percentage P_C which is ADICP (acid detergent insoluble crude protein) is indigestible. For N, all the fractions-including crude protein-were examined in triplicate.

In vitro dry matter digestibility (IVDMD) and in vitro organic matter digestibility (IVOMD): Truly degradable feed was estimated using Goering and Van Soest (1970) approach. Approximately, 500 mg of substrate was taken in 100 ml bottles as triplicate. Warm (about 39°C) 1095 millilitres of distilled water, 730 ml of rumen buffer solution (35.0 g NaHCO3 and 4 g NH4HCO3), 365 ml of macro mineral solution (6.2 g KH2PO4, 5.7 g Na2HPO4, 2.22 g NaCl, and 0.6 g MgSO4.7H2O made up to 1 litre with distilled water), and 0.23 ml of micro mineral solution (10.0 g MnCl2.4H2O, 13.2 g CaCl2. 2H2O, 1 g CoCl2.6H2O, 8.0 g FeCl2.6H_2O made up to 100 ml volume with distilled water) and rumen fluid (660 ml) was added to it and comprised of reduced medium, 60 ml of freshly manufactured reduction solution with 580 mg Na_2S. $9H_2O$ and 3.7 ml of 1 M-NaOH, together with 1 ml of resazurine (0.1 g made up to 100 ml with distilled water) (Makkar *et al*. 1995). Using a magnetic stirrer equipped with a heated plate, the mixture was continuously swirled at 39°C under CO_2. Each bottle was filled with 40 ml of the rumen-fluid medium, which was then incubated for 24 h (24 h for all feeds except dry roughages, which were incubated for 48 h) at 39°C in a water bath. The entire bottle's contents were transferred into a 1000 ml beaker (spout less) during incubation. About 40 cc of double strength NDS was used to wash the bottles, and they were refluxed for one hour at 100°C. The contents were dried in a hot air oven set at 90°C after being filtered through a sintered glass crucible. To find the OM content, the residue in each crucible was ashed for 2 h at 550°C in a muffle furnace.

In vitro gas production: Pressure transducer approach was utilized to calculate *in vitro* gas production (Theodorou *et al.* 1994). Two adult male Murrah buffaloes (fistulated) maintained on wheat straw and concentrate diet (65:35 dry matter basis) were used to collect rumen fluid prior to their feeding. Before the incubation period started, the rumen fluid was bubbled with CO_2 and filtered through two layers of cheese cloth. The incubation medium was prepared by sequentially mixing the buffer solution, macro-mineral solution, micro-mineral solution, and resazurin solution (Menke and Steingass, 1988). Three 150 ml serum vials were filled with 1 g of air-dried sample material. As blanks, three serum bottles devoid of substrate were utilised. After briefly gassing the sample and control serum vials with CO_2, 65 ml of medium was added. After continuously fluxing each bottle with CO_2, 3 ml of reducing solution were added. Bottles were gassed with CO_2 until the pink hue vanished. Each bottle's head space gas pressure was adjusted using the gas pressure transducer before to inoculation in order to modify the LED display's zero reading. About 8 ml of ruminal fluid inoculum were injected into serum bottles using a 10 ml syringe. The bottles were sealed and then incubated at 39°C. The amount of gas produced (measured in millilitres) was determined after a 24 h period.

Methane measurements: Gas chromatography (Nucon 5700 Microprocessor controlled gas chromatograph, Okhla, New Delhi, India) fitted with a stainless steel column packed with Porapak-Q and a Flame Ionisation detector was used to measure the amount of methane in the total gas from three bottles that were incubated for each of the samples for 24 h. One µl of gas was extracted using a Hamilton syringe and manually injected into the gas analyzer (GC) using the push-and-pull method of sample injection. The GC was calibrated using standard amounts of CO_2 and CH_4. Methane was also measured from blanks at the chosen fermentation times, and the results were used to adjust for CH_4 generated by the inoculum.

Using 39.54 kJ/L CH_4 and 0.716 mg/ml CH_4 factors, the observed methane was converted to energy and mass values, respectively and its concentration was estimated in relation to the total gas (Santoso *et al*. 2007).

Statistical analysis: The GLM technique of SAS (2002)

Table 1. Chemical composition of the common feed ingredients and fodder

Feed ingredients/ fodder	Class	CP (%DM)	NDF (%DM)	EE (%DM)	Starch (% DM)	SP (%DM)	NPN (%SP)	Lignin(sa) (% NDF)
Maize (yellow)	Grains	9.91	13.68	3.38	74.00	2.11	27.84	6.36
Maize (white)	Grains	8.03	14.6	2.40	73.19	1.57	47.20	5.28
Wheat	Grains	15.83	14.64	1.59	72.92	4.74	10.85	8.13
Pearl millet	Grains	10.63	12.08	2.70	63.63	1.33	16.60	12.23
Sorghum (white)	Grains	9.30	11.26	2.06	66.77	2.39	22.03	14.38
Sorghum(red)	Grains	9.10	13.50	2.35	69.32	1.78	30.70	10.69
Barley	Grains	9.10	25.89	2.21	46.72	1.68	45.31	15.14
Oat	Grains	11.68	22.10	3.66	70.14	1.79	69.00	17.94
Wheat bran	AIBP	16.03	33.26	1.62	69.64	2.56	43.52	19.09
Rice bran	AIBP	9.28	15.14	6.80	43.63	2.99	66.89	29.72
DORB	AIBP	11.46	35.41	1.47	47.28	2.17	29.02	22.99
Rice polish	AIBP	13.47	31.53	19.31	44.75	4.54	21.60	22.46
MGM	AIBP	64.26	16.43	5.27	17.44	19.40	53.49	20.63
Gram churi	AIBP	16.66	30.36	3.55	54.41	7.42	73.32	13.04
Gram husk	AIBP	3.51	75.65	0.27	92.59	1.45	23.45	30.85
Gram khanda	AIBP	9.96	48.69	1.41	62.80	0.69	60.65	30.93
DDGS	AIBP	44.05	20.44	3.46	20.29	7.55	36.67	6.07
Wheat straw	AIBP	4.63	74.40	1.73	46.42	0.38	50.53	12.41
Paddy straw	AIBP	4.39	72.36	2.72	43.36	0.67	67.22	6.04
Beet pulp	AIBP	10.96	43.77	1.45	22.60	7.25	13.87	9.06
Distillery bran	AIBP	18.17	44.59	2.57	31.30	5.65	44.34	6.38

Table 2. Carbohydrate fractions of the feed ingredients and fodder as per CNCPS (% of total CHO)

Feed ingredients/ fodder	Class	C_A	C_{B1}	C_{B2}	C_C
Maize (yellow)	Grains	21.49	61.17	14.85	2.49
Maize (white)	Grains	22.11	60.37	15.37	2.15
Wheat	Grains	21.60	58.15	16.65	3.61
Pearl millet	Grains	31.77	55.57	8.37	4.30
Sorghum (white)	Grains	29.30	58.88	7.27	4.56
Sorghum(red)	Grains	26.32	59.43	10.16	4.09
Barley	Grains	38.47	33.74	16.82	10.98
Oat	Grains	22.43	52.68	13.26	11.64
Wheat bran	AIBP	18.62	42.71	19.17	19.50
Rice bran	AIBP	45.51	35.25	3.73	15.51
DORB	AIBP	32.04	29.27	13.80	26.89
Rice polish	AIBP	25.15	21.56	24.18	29.11
MGM	AIBP	52.31	11.05	4.16	32.49
Gram churi	AIBP	28.09	33.52	25.74	12.65
Gram husk	AIBP	1.49	18.66	19.25	60.60
Gram khanda	AIBP	16.20	27.35	13.63	42.83
DDGS	AIBP	61.74	15.72	16.30	6.25
Wheat straw	AIBP	8.37	7.25	58.69	25.70
Paddy straw	AIBP	4.50	3.45	78.21	13.84
Beet pulp	AIBP	39.02	11.31	37.58	12.09
Distillery bran	AIBP	36.07	16.14	38.83	8.96

was utilised to conduct an analysis of variance on the data in a completely randomised design consisting of three field replicates. The model was:

$$Yij = \mu + Fi + Eij$$

where Fi is the fixed effect of the ith sample (i = 1–21) and Yij is the variable's individual observation. The total mean is represented as μ, and the random error linked to Yij that is not taken into account by the fixed effect is called Eij. Means were divided with Fisher's LSD and all statistical tests were at α= 0.05 level of significance. $P<0.05$ was used to classify mean differences as statistically significant.

RESULTS AND DISCUSSION

Chemical composition: The information regarding carbohydrate fractions, ether extract, crude protein, starch, NPN, Lignin and ash contents in grains and AIBP have been presented in the Table 1. The NDF (% of dry mater) was analyzed to be least in sorghum grains (11.26) and it was found to be highest in gram husk (75.65). Similarly, lignin (% NDF) was analyzed to be least in maize (white) (5.28) and maximum in gram khanda (30.93). The SP (%DM) varied from 0.38 in wheat straw to 19.40 in MGM. The gram churi contained the highest (73.32) NPN (%SP), while the wheat grains (10.85) had the lowest. The EE (%DM) varied between 0.27 (gram husk) and 19.31 (rice polish). Some workers (Bose *et al*. 2014, Das *et al*. 2014, Prusty *et al.* (2013) reported higher values for NDF (%DM) fraction, whereas Kamble *et al*. (2010) reported lower values. Lower values of lignin (%NDF) were reported for the grains. Further, Das *et al*. (2014) documented similar values for EE (%DM). Das *et al*. (2014) and Prusty *et al*. (2013) reported similar values of NDF for wheat straw. Accumulation of NDF, SP and lignin was higher ($P<0.001$) in AIBP, however, with grains the opposite happened. AIBP had a higher ($P<0.001$) SP concentration than grains.

Carbohydrate fractions: The data regarding the carbohydrate fractions of grains and AIBP (% CHO) have been presented in the Table 2. Among AIBP, C_{B1} content was highest (42.71) in wheat bran and C_A was highest (61.74) in DDGS followed by MGM (52.31). The higher C_A content of DDGS and MGM was possibly as a result of the presence of highly fermentable sugars, owing to the method involved in their processing. AIBP had higher unavailable carbohydrate fraction (C_C) compared to grains. Of the cell wall content (C_{B2}+C_C), a large part is available for digestion (C_{B2}) in paddy straw (84.96%) and wheat straw (69.54%), though the total cell wall content is very high. Grains have generally low indigestible cell wall content (C_C) (Avg. 5.48) compared to digestible cell wall content (C_{B2}) (Avg. 12.84), whereas, AIBP had generally equal fractions of both (27.17) for C_{B1} and 23.57 for C_A. The C_{B2} fraction consists of β-glucans and pectic materials that aren't digested by mammalian enzymes, thus considered among the fibre component (Lanzas *et al.* 2007). Among grains, C_{B1} fraction was highest in yellow maize (61.17%) whereas barley grain has lowest C_{B1} (33.74%) and rich in soluble sugars. But Prusty *et al*. (2013) reported better starch content in barley compared to soluble sugars. The amylase enzyme present in the endosperm might have caused the hydrolysis of starch granules to improve the sugar content of the grain in the present study. The similar carbohydrate fractions of maize and wheat grain indicated that maize could be effectively replaced by wheat grain without altering the content of sugar, soluble fiber, NDF and starch content. In corroboration to the present findings Gupta et al. (2011) found higher CB1 in grains and grain by-products, except for deoiled rice bran in which highest CC fraction was reported. CB2 fraction of wheat and paddy straw was highest among all carbohydrate fractions and the values corroborated with the reports of Kaithwas et al. (2020). Who reported negligible CA fraction in the cereal straws compared to present values.

Protein fractions: The data regarding the protein fractions of grains and AIBP (% CP) have been given in the Table 3. The values (%) of the fraction P_A were highest in gram churi (32.67) and lowest in pearl millet grains (2.07). Among AIBP, the P_{B1} varied from (56.98) in beet pulp to (2.74) in gram khanda, but was highest for wheat (26.85) and lowest for oat (4.75) in grains. The P_{B2} values were found to be highest (82.61) in gram khanda and lowest

Table 3. Protein fractions of the feed ingredients and fodder as per CNCPS (% of crude protein)

Feed ingredients/ fodder	Class	P_A	P_{B1}	P_{B2}	P_{B3}	P_C
Maize (yellow)	Grains	5.90	15.34	66.44	8.61	3.71
Maize (white)	Grains	9.23	10.32	63.75	12.16	4.55
Wheat	Grains	3.28	26.85	60.86	4.68	4.34
Pearl millet	Grains	2.07	10.40	72.08	8.36	7.09
Sorghum (white)	Grains	5.45	19.28	62.26	7.00	6.01
Sorghum(red)	Grains	5.52	12.47	69.94	5.12	6.94
Barley	Grains	8.23	9.93	59.38	17.63	4.83
Oat	Grains	10.58	4.75	69.75	11.59	3.33
Wheat bran	AIBP	6.96	9.04	65.10	14.13	4.77
Rice bran	AIBP	21.56	10.67	52.46	8.37	6.63
DORB	AIBP	5.50	13.45	30.08	34.87	16.10
Rice polish	AIBP	7.27	26.39	56.87	3.74	5.72
MGM	AIBP	12.71	17.24	59.64	8.19	2.20
Gram churi	AIBP	32.67	11.89	46.30	3.44	5.70
Gram husk	AIBP	9.68	31.62	6.29	27.34	25.07
Gram khanda	AIBP	4.22	2.74	82.61	1.73	8.71
DDGS	AIBP	6.29	10.86	60.83	2.27	19.75
Wheat straw	AIBP	4.11	4.02	56.22	13.72	21.93
Paddy straw	AIBP	10.25	5.01	25.74	43.39	15.60
Beet pulp	AIBP	9.18	56.98	1.17	29.85	3.28
Distillery bran	AIBP	13.79	17.31	23.53	41.43	3.94

(1.17) in beet pulp. Values of this fraction among grains were highest for pearl millet (72.08) and lowest for barley (59.38). Similarly, the P_{B3} fraction was highest (43.39) in paddy straw and lowest (1.73) in gram khanda ,while the wheat and barley had lowest (4.68) and highest (17.63) values among grains, respectively. P_C fraction ranged from 25.07 in gram husk to 2.20 in maize gluten meal. Among grains, highest (7.09) and lowest (3.33) values were for pearl millet and oat, respectively. Comparable results were noted for maize grains by Sharma *et al.* (2018). Dixit and Kundu (2022) found fraction P_A for paddy straw 9.79%, which was similar to present study (10.25%). For fraction P_{B3} in paddy straw, they observed 36.7%, which is marginally lower than those noted in present study (43.39%). The results of this research were in accordance to the findings of other workers (Kamble *et al.* 2010, Prusty *et al.* 2013, Das *et al.* 2014).

IVDMD and gas production: Percentage *in vitro* dry matter digestibility (IVDMD) and *in vitro* organic matter digestibility (IVOMD) after 24 h of incubation were higher for grains as compared to AIBP (Table 4 and 5). Average values of IVDMD and IVOMD for grains being 89.78% and 90.80%, respectively and for AIBP 75.19% and 75.23%, respectively. Among grains, both highest IVDMD and IVOMD were observed for red sorghum (95.5 8%) and (95.98%), respectively while among AIBPs these figures

Table 4. *In vitro* digestibility, net gas and methane production in grains incubated for 24 hrs

Cereal grain	IVDMD%	IVOMD%	Gas production (ml/200mg)	Gas production (ml/200mgDM)	Methane (g/kgDM incubated)	Methane (g/kg IVDMD)
Maize (yellow)	90.79	90.74	66.49	2.66	36.49	40.19
Maize (white)	91.6	94.44	64.27	2.58	33.20	36.22
Wheat	92.27	93.47	54.55	2.18	27.89	30.23
Pearl millet	92.92	93.51	62.65	2.51	25.67	27.63
Sorghum (white)	94.08	95.41	52.40	2.10	23.26	24.73
Sorghum(red)	95.58	95.98	61.38	2.46	34.10	36.13
Barley	79.44	79.82	56.52	2.26	31.05	39.27
Oat	81.52	83.05	44.50	1.78	22.06	14.15

Table 5. *In vitro* digestibility, net gas and methane production in agro industrial by products incubated for 24 hrs

AIBP	IVDMD%	IVOMD%	Gas production (ml/200mg)	Gas production (mM/200mgDM)	Methane (g/kgDM incubated)	Methane (g/kg IVDMD)
Wheat bran	78.84	79.32	51.75	2.07	57.58	73.03
Rice bran	84.83	85.80	40.61	1.63	32.76	38.62
DORB	81.31	82.30	45.81	1.83	50.37	61.95
Rice polish	80.98	76.32	17.87	0.72	8.96	11.06
MGM	96.73	97.73	45.65	1.83	21.35	22.07
Gram churi	86.94	87.31	53.99	2.16	55.62	63.98
Gram husk	52.27	52.19	22.44	0.90	9.77	18.69
Gram khanda	79.81	80.38	39.08	1.56	21.70	27.19
DDGS	83.23	77.77	20.45	0.82	12.42	16.59
Wheat straw	51.09	51.99	23.41	0.94	12.85	25.15
Paddy straw	56.27	58.29	18.63	0.75	9.11	16.20
Beet pulp	88.084	90.27	45.62	1.83	21.49	24.71
Distillery bran	57.10	58.33	11.46	0.12	7.13	12.48

were noted for MGM (96.73%) and (97.73%), respectively. Reddy *et al.* (2021) obtained similar results for maize, sorghum, wheat and pearl millet except barley where IVDMD reported was little higher (91.55%). Average gas production (ml/200 mg) was found to be 57.85 for grains and 33.60 for AIBP. Among grains, highest values were noted for yellow maize (66.49) and lowest for oat (44.50), while among AIBP, the highest and lowest values were found to be for gram churi (53.99) and distillery bran (11.46), respectively. Similar results for maize, wheat, sorghum grains, paddy straw and wheat straw were observed (Kannan *et al.* 2017). Average methane production (g/kg IVDMD) for grains and agro industrial by-products was found to be similar, but the range was more in case of AIBP. Minimum values for grains were observed for oat (14.15) and maximum value was noted for yellow maize (40.19), while among AIBP, minimum values were found for rice polish (11.06) and maximum values were observed for wheat bran (73.03). Similarly, methane production (g/kg DM incubated) was highest for yellow maize (40.19) and minimum for (14.15) among grains. Among AIPB minimum values were observed for distillery bran (7.13) and maximum for wheat bran (57.58). These results were in line to those obtained by Aboagye *et al*. (2021), except for oat grains where methane production was reported to be 31 mg/kg DM against 22.06 mg/kg DM in present study.

The test feed samples were analysed and it was found that DDGS had the greatest C_A fraction (% CHO) (61.74%), followed by MGM (52.31%) among the AIBP, while among grains, barley (38.47%) had the highest C_A fraction. The most instantly degradable protein fraction (32.67% of CP) existed in gram churi, whilst the highest indigestible protein fraction (%CP) was assessed to be 21.18% in wheat straw (AIBP) and 7.09% in pearl millet (grains). Highest *in vitro* methane production (g/kg DM incubated) among AIBP and grains were noted for wheat bran (57.58) and yellow maize (36.39), respectively. In addition to chemical composition, in-depth understanding of CNCPS protein fractions of different feed ingredients has become more significant because of its pivotal role in integrating intestinal digestibility and ruminal degradability. This knowledge can thereafter be utilized to ascertain the RDP, UDP, and MP values for more precise feed formulations in ruminant nutrition and improved productivity.

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