



Evaluation of the Nutritional, Mineral, and Estimated Metabolizable Energy Values of Agricultural Biomass Raw Materials Used for the Development of Concentrated Fermented Feed for Beef Cattle in Mukdahan Province, Northeast Thailand

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Abstract: This study evaluated the nutritional, mineral, and estimated metabolizable energy (EME) profiles of eight locally available agricultural biomass raw materials to support the development of a cost-effective concentrated fermented feed (CFF) for beef cattle in Mukdahan Province, Northeast Thailand. Finely ground maize kernels, soybean meal, palm kernel meal, rice bran, dried cassava chips, cassava pulp, washed cassava peels, and fresh cassava starch residue were analyzed for proximate composition, EME, and essential minerals (Ca, Mg, P, Fe, Zn, Na, and K), with all values reported on an as-fed basis. A CFF formulation (1,000 kg batch) was developed from these raw materials and compared with five commercial beef cattle feeds. Soybean meal, palm kernel meal, and rice bran exhibited the highest protein contents, while rice bran showed the greatest calculated EME value (4097 kcal/kg of sample). Cassava-based by-products were rich in carbohydrates but relatively low in protein and fat. Mineral analysis revealed substantial variation among raw materials, with high concentrations of P, Mg, Ca, and K in several biomass sources. The formulated CFF contained $12.16 \pm 0.21\%$ crude protein, $3.29 \pm 0.05\%$ fat, and $47.56 \pm 0.90\%$ carbohydrates. The calculated EME content was 2685 kcal/kg of sample, which is highly comparable to the nutrient profiles of commercial feeds available in the market. Importantly, CFF production costs were markedly lower (4.50 THB/kg) than those of commercial products (10.67–15.33 THB/kg), demonstrating significant economic advantages. Overall, the results confirm that locally sourced biomass can be effectively used to formulate nutritionally adequate, mineral-rich, and low-cost fermented feed, thereby contributing to sustainable beef cattle production and reducing farmers' feed expenses.

Keywords: Agricultural biomass; fermented feed; beef cattle nutrition; nutritional composition; estimated metabolizable energy (EME) evaluation

1. Introduction

Thailand is an agriculture-driven nation with approximately 20 million people (28% of the population) engaged in the sector. Agriculture is vital to national economic development, with crop cultivation—including rice, rubber, cassava, oil palm, maize, and soybean—serving as both key export commodities and essential raw materials for livestock feed and various agro-industries [1–5]. According to the Land Development Department (2019), agricultural land covers approximately 153 million rai (47.77% of the total area) and supports 5.8 million farming households. This extensive base ensures national food security and provides a steady supply of raw materials for domestic and export markets, thereby strengthening economic stability and long-term competitiveness [5–7]. The livestock sector is also an important component of Thailand's agricultural economy. Data from the Department of Livestock Development (DLD, 2021) indicate that the country had 3,302,406 livestock farmers, accounting for approximately 16.5% of total agricultural households. Most farmers raise beef, dairy, and buffalo cattle, swine, and native chickens. Notably, the beef cattle population reached 7,364,967 head in 2021. In Livestock Zone 4 (Central Northeast and Upper Northeast regions), Mukdahan Province alone recorded about 17,477 beef cattle farmers managing 79,071 head, generating an estimated total income of 2.4 billion Thai Baht (based on an average sale price of 30,000 Baht per head). These statistics highlight beef cattle production as a viable pathway to enhance rural income and economic resilience [8–10].

Feed cost is recognized as the most significant expense in cattle production systems, accounting for more than 60% of the total production cost. The continuous rise in raw material prices—particularly for imported soybean meal—has intensified this financial burden. Consequently, developing cost-effective feed formulations using locally available agricultural by-products has become a practical strategy to reduce production costs while simultaneously creating market opportunities for local crop producers [11–13]. Several studies have addressed this issue. Sooksoi [14] reported that rice straw used as roughage by dairy farmers in Sa Kaeo Province had low nutritional quality, characterized by a low protein content (2.78%) and high levels of cell wall and lignocellulosic fiber (78.05% and 56.89%, respectively). Munier et al. [15] demonstrated that corn cob silage significantly improved average daily gain and body condition score in Donggala cattle, with inclusion levels of 30.5–31% yielding the best results ($p < 0.05$). Dagaew et al. [16] further showed that cassava solid yeast waste (CSYW) could completely replace soybean meal in concentrate diets for Thai native beef cattle without adverse effects on feed intake, digestibility, or rumen fermentation, while increasing rumen ammonia–nitrogen and total bacterial populations ($p < 0.05$).

Despite the potential of agricultural by-products, a research gap exists regarding the mineral and EME profiling of biomass unique to the Mukdahan border trade zone. While previous studies focused on single-ingredient replacements or silage quality, Mukdahan offers a unique industrial landscape with high volumes of residues from sugar, cassava, and oil palm plants. Lack of integrated data on these local materials hinders the development of a CFF tailored to regional production. Without precise EME and mineral evaluations on an as-fed basis, farmers rely on expensive commercial feeds or unbalanced residues, which limits efficiency. This study addresses this gap by mapping local biomass to develop a scientifically formulated, cost-effective CFF. Additionally, fermentation technology enhances feed quality through pre-digestion, where microbial enzymes degrade complex carbohydrates and anti-nutritional factors, improving bioavailability and palatability. Fermented feeds also provide beneficial microorganisms and organic acids that stabilize ruminal pH and improve fermentation efficiency. These biological improvements enhance feed intake and metabolic health, offering a superior alternative to non-fermented local diets.

In Nong Sung District, Mukdahan Province, the Nong Sung Agricultural Cooperative Limited has established an integrated beef cattle production model encompassing upstream (breeding management), midstream (feed development), and downstream (processing and marketing) operations. This holistic system strengthens the economic sustainability of both livestock and crop farmers. However, the recent escalation in feed prices, driven by the high cost of imported protein sources, has negatively affected production profitability and farmer income. Therefore, this study was designed with the primary objective to conduct a comprehensive laboratory-based evaluation of eight locally available agricultural biomass raw materials in Mukdahan Province. Specifically, the research aims to: (1) determine the proximate composition and essential

mineral profiles (Ca, Mg, P, Fe, Zn, Na, and K) of regional biomass; (2) EME values based on chemical analysis; and (3) develop a cost-effective CFF formulation by comparing its laboratory-derived nutritional profile and production cost with commercial alternatives. It should be noted that this study focuses exclusively on the nutritional and economic characterization of the feed, and does not include in vivo animal performance trials.

2. Materials and Methods

2.1 Materials and sampling procedure

Agricultural biomass raw materials were collected from various locations in Mukdahan Province, Thailand, during the period from October to December, 2023. To ensure data representativeness, each biomass material was collected in three independent batches ($n = 3$) from different local sources, including sugar mills, cassava starch factories, palm oil extraction plants, and community rice mills. The collected materials included finely ground maize kernels (FMK), soybean meal (SBM), palm kernel meal (PKM), rice bran (RB), dried cassava chips (DCC), cassava pulp after starch extraction (CSP), washed cassava peels (WCP), and fresh cassava starch residue (CSR) as displayed in Figure 1. For the formulated feeds, the CFF was developed in collaboration between the Nong Sung Agricultural Cooperative Limited and the Appropriate Technology Center at Sakon Nakhon Rajabhat University. A total of 3 independent 1,000-kg CFF batches were produced and sampled for analysis. Furthermore, five commercial beef cattle feeds were procured from different local distributors: one pelleted feed (12% protein, C1), three pelleted feeds from different manufacturers (14% protein, C2, C3, and C4), and one powdered feed (14% protein, C5), as shown in Figure 2. From each commercial product, three separate bags were randomly sampled as replicates. All biomass, CFF, and commercial feed samples were oven-dried at 60°C until constant weight was reached, then finely ground to pass through a 1-mm screen to ensure uniformity for subsequent analytical procedures. To ensure consistent comparison across all analyses, including proximate composition, minerals, and EME values, all nutritional data in this study are reported on an as-fed basis. This approach was selected to provide practical information that aligns with local farmers' actual feeding practices.



Figure 1. Agricultural biomass raw materials collected from Mukdahan Province, Thailand, including finely ground maize kernels, cassava pulp (after starch extraction), dried cassava chips, rice bran, washed cassava peels, soybean meal, palm kernel meal, and fresh cassava starch residue used for CFF formulation.



Figure 2. Concentrated fermented feed (CFF) was developed in collaboration between the Nong Sung Agricultural Cooperative Limited and the Appropriate Technology Center, SNRU, along with commercial beef cattle feeds (C1–C5) collected from the Mukdahan market for comparative analysis.

The key reagents used in this study included analytical reagent (AR) grade chemicals as follows: sodium hydroxide (NaOH), boric acid (H_3BO_3), hydrochloric acid (HCl), sulfuric acid (H_2SO_4), copper (II) sulfate ($CuSO_4$), acetone (C_3H_6O), hydrogen peroxide (H_2O_2), nitric acid (HNO_3), strontium chloride hexahydrate ($SrCl_2 \cdot 6H_2O$), and perchloric acid ($HClO_4$), all supplied by Ajax Finechem (Australia). Hexane (C_6H_{14}) of AR grade was obtained from KEMAUS (Australia). Standard solutions of calcium (Ca), magnesium (Mg), zinc (Zn), iron (Fe), sodium (Na), potassium (K), and phosphorus (P) were also AR grade and supplied by Ajax Finechem. In addition, methylene blue and methyl red were used as analytical indicators.

2.2 Preparation of the biomass raw materials and beef cattle feed samples

The collected biomass was oven-dried and ground into a fine powder before being sealed in zip-lock bags for subsequent analysis. The moisture content of the samples was determined following the method described by Phewphong et al. [17], Suwannatrai et al. [18], and Preecharram et al. [19, 21]. Initially, empty beakers were dried in a hot-air oven at 105 °C until a constant weight was obtained and then cooled in a desiccator. The weight of the dried beaker (W_B) was recorded. Approximately 5 g of the sample was weighed into the beaker, and its total weight (W_S) was recorded. The beakers containing the samples were then dried in the oven at 105 °C for 3 h, cooled in a desiccator for 30 min at room temperature, and reweighed. The drying and weighing process was repeated at 30 min intervals until a constant weight (W_T) was achieved. Each determination was performed in triplicate. The percentage of moisture content was calculated using Eq. (1):

$$\text{Moisture content (\%)} = [W_S - (W_T - W_B) / W_S] \times 100 \quad (1)$$

where W_S is the weight of the sample before drying (g), W_T is the weight of the beaker with the dried sample (g), and W_B is the weight of the empty beaker (g).

2.3 The proximate analysis of the biomass raw materials and beef cattle feed samples

2.3.1 Determination of crude protein content

The total crude protein content of the biomass raw materials and beef cattle feed samples was determined following the AOAC method [20] and previous studies by Phewphong et al. [17] and Preecharram et al. [19, 21]. Briefly, 1.0 g of the finely ground sample was weighed into a digestion tube. Two catalyst tablets containing 10 g of potassium sulfate (K_2SO_4) and 1 g of copper (II) sulfate ($CuSO_4$) were added to accelerate digestion. Subsequently, 15 mL of concentrated sulfuric acid was carefully added, and the mixture was gently shaken to homogenize. The digestion tube was then placed in a digestion apparatus equipped with an acid fume trap and heated at 420 °C for approximately 1 h or until the solution became clear green, indicating complete digestion. The tube was removed and allowed to cool to room temperature while the fume trap remained active to absorb any residual acid vapors. For steam distillation, the distillation apparatus was first rinsed with distilled water and then prepared by adding 80 mL of distilled water, 50 mL of 32% (w/v) sodium hydroxide, and 30 mL of 4% boric acid containing mixed indicators (methylene blue and methyl red) into a 250 mL conical flask. The digestion tube was attached to the distillation unit, ensuring that the glass tip was

immersed in the solution. The system was closed and distillation was performed for approximately 5 min. After distillation, the contents of the conical flask were titrated with 0.1 N hydrochloric acid (HCl) until the color changed from green to purple. All analyses were performed in triplicate. The total nitrogen content was calculated and converted to crude protein using the following equations (2) and (3):

$$\text{Nitrogen (\%)} = [(0.014007) \times (V_1 - V_2) \times N_{\text{HCl}} / \text{Sample weight (g)}] \times 100 \quad (2)$$

$$\text{Protein (\%)} = \text{Nitrogen (\%)} \times 6.25 \quad (3)$$

where V_1 is the volume of 0.1 N HCl required to titrate the sample (mL), V_2 is the volume used for the blank titration (mL), N_{HCl} is the exact normality of the HCl used (0.1 N), and 0.014007 is the milliequivalent weight of nitrogen. The conversion factor of 6.25 was used to convert nitrogen content to protein content.

2.3.2 Determination of fat content

The fat content of the samples was determined using the Soxhlet extraction technique, following the procedures described by Phewphong et al. [17], Preecharram et al. [19, 21], and Suwannatrai et al. [18, 22]. First, an aluminum dish was dried in a hot air oven at 105 °C for 1 h, cooled in a desiccator for 30 min, and reheated at 30 min intervals until a constant weight was obtained (M_1). Approximately 1 g of the dried sample was accurately weighed using a four-decimal analytical balance and transferred into a thimble (M_0). The thimble containing the sample was placed in the Soxhlet extractor, and 120 mL of analytical-grade hexane was added as the solvent. The extraction was carried out under reflux for 3 h. After extraction, the solvent was removed using a rotary evaporator, and the remaining oil residue was oven-dried at 105 °C for 30 min, cooled in a desiccator, and reweighed. The drying process was repeated every 30 min until a constant weight was achieved (M_2). All experiments were conducted in triplicate.

The percentage yield of the extracted oil was calculated based on the dry weight of the sample using Eq. (4):

$$\% \text{Yield of oil} = [(M_2 - M_1) / M_0] \times 100 \quad (4)$$

where M_0 is the initial sample weight (g), M_1 is the weight of the empty aluminum dish (g), and M_2 is the weight of the aluminum dish containing the extracted oil (g).

2.3.3 Determination of crude fiber content

The crude fiber content of the samples was determined from the defatted residues obtained after Soxhlet extraction, following the procedures described by Phewphong et al. [17], Preecharram et al. [19,21], Suwannatrai et al. [18, 22], and Salami and Afolayan [23]. Approximately 1 g of the defatted sample was placed in a sintered glass crucible. A total of 200 mL of 0.128 M sulfuric acid solution was added, and the mixture was heated to boiling for 30 min. During boiling, the volume was maintained constant by adding distilled water as needed. The mixture was then filtered through the sintered glass crucible under suction and washed with hot distilled water three times (50 mL each). Subsequently, 200 mL of 0.313 M sodium hydroxide solution was added, and the mixture was again boiled for 30 min while maintaining a constant volume by adding distilled water. The residue was filtered under suction and washed with hot distilled water three times (50 mL each).

The remaining residue was further washed with 25 mL of 0.128 M sulfuric acid, then with 25 mL of hot distilled water until the acid was completely removed, and finally washed three times with 50 mL of acetone. The crucible containing the residue was dried in a hot air oven at 105 °C for 1 h, then ignited in a muffle furnace at 550 °C until a white ash was obtained. The crucible was cooled in a desiccator and weighed. All analyses were performed in triplicate. The crude fiber content (%) was calculated using Eq. (5):

$$\text{Crude fiber content (\%)} = [(M_1 - M_2) / M_0] \times 100 \quad (5)$$

where M_1 is the weight of the crucible with the dried residue (g), M_2 is the weight of the crucible with ash after ignition (g), and M_0 is the sample weight (g).

2.3.4 Determination of ash content

The ash content of the samples was determined according to the standard procedures reported by Phewphong et al. [17], Preecharram et al. [19,21], and Suwannatrai et al. [18, 22]. Briefly, a clean crucible was dried in an oven at 105 °C until a constant weight was achieved, cooled in a desiccator, and weighed to obtain the empty crucible weight (W_B). Approximately 1 g of the sample (W_0) was placed into the crucible and incinerated in a muffle furnace at 550 °C until white ash was obtained. After cooling in a desiccator, the crucible containing the ash was weighed (W_T). The ash content was calculated using the following Eq. (6):

$$\text{Ash content (\%)} = [(W_T - W_B) / W_0] \times 100 \quad (6)$$

where W_T is the weight of the crucible with the ash (g), W_B is the weight of the empty crucible (g), and W_0 is the weight of the sample (g). All measurements were performed in triplicate to ensure accuracy.

2.3.5 Determination of carbohydrate content

The carbohydrate content of the samples was estimated by difference, based on the proximate composition including moisture, crude protein, crude fat, ash, and crude fiber, following the method described by Phewphong et al. [17], Preecharram et al. [19, 21], and Suwannatrai et al. [18, 22]. The carbohydrate content was calculated using the following Eq. (7):

$$\text{Carbohydrate (\%)} = 100 - (\% \text{Moisture} + \% \text{Protein} + \% \text{Fat} + \% \text{Ash} + \% \text{Crude Fiber}) \quad (7)$$

For samples with high moisture content, carbohydrate content was calculated on a dry-weight basis to ensure accuracy.

2.3.6 Determination of EME content

The EME content of the samples was estimated based on their proximate composition, following the approach described by Phewphong et al. [17], Preecharram et al. [19, 21], Suwannatrai et al. [18, 22], and Salami and Afolayan [23]. The energy values reported in this study are referred to as EME and are calculated from modified Atwater factors derived from proximate analysis data. It is important to acknowledge that these values represent theoretical energy yields rather than direct physiological measurements from in vivo ruminant digestion trials (in vivo ME). This estimation approach is commonly used in laboratory-scale nutritional screenings to provide a rapid and cost-effective energy profile of various feed ingredients. The calculation accounted for each macronutrient using the following energy values: 4 kcal/g for total carbohydrates, 4 kcal/g for crude protein, and 9 kcal/g for crude fat. The total energy content was then calculated as the sum of these contributions using Eq. (8):

$$\text{EME (kcal/100 g of sample)} = (4 \times \text{Carbohydrates}) + (4 \times \text{Protein}) + (9 \times \text{Fat}) \quad (8)$$

This approach provides estimates of the caloric value of biomass raw materials and beef cattle feed samples, allowing comparison of energy density across different formulations. Furthermore, the experimental results were processed and reported as kcal/kg of sample, as this unit is widely recognized, easy to understand, and commonly used in practical feed formulation.

2.4 Determination of mineral content in biomass raw materials and beef cattle feed samples

The mineral composition of the biomass raw materials and beef cattle feed samples was determined following standard procedures, based on the AOAC method [24] and the approaches described by Phewphong et al. [17], Preecharram et al. [19], and Bvenura and Afolayan [25]. Approximately 1 g of each sample was accurately weighed into a 100 mL beaker, after which 20 mL each of concentrated nitric acid (HNO_3) and hydrogen peroxide (H_2O_2) was added. The mixture was digested on a hot plate until a clear solution was obtained, ensuring that the solution did not completely dry. The digested solution was allowed to cool to room temperature, filtered, and the filtrate was collected in a 100 mL volumetric flask, adjusted to

volume with distilled water, and thoroughly mixed prior to analysis using an Atomic Absorption Spectrometer (AAS, iCE3000 series, Thermo, USA).

2.4.1 Determination of Magnesium (Mg) and Calcium (Ca)

The concentrations of Mg and Ca in the biomass raw materials and beef cattle feed samples were determined using an AAS. The digested sample solutions prepared as described in Section 2.4 were used for this analysis. For the preparation of reagents, a mixed acid solution of nitric acid (HNO₃, 69–70%) and perchloric acid (HClO₄, 69–72%) in a 1:1 v/v ratio was prepared and thoroughly mixed. Additionally, a strontium chloride solution was prepared by dissolving 61 g of SrCl₂·6H₂O in 420 mL of 36–38% HCl, then diluting to 1000 mL with distilled water. Working standard solutions of Mg and Ca at concentrations of 0, 2, 4, 6, 8, and 10 ppm were prepared by adding 5 mL of the strontium chloride solution to each flask, followed by pipetting appropriate volumes (0, 1, 2, 3, 4, and 5 mL) of 10 ppm stock solutions into 50 mL volumetric flasks, and adjusting the volume to the mark with distilled water. The digested sample solutions were then analyzed using AAS, and the absorbance values were compared with those of the working standards to determine the concentrations of Mg and Ca in the samples. The mineral content in the samples was calculated using the following eq. (9):

$$\text{Mg or Ca (\%)} = [(\text{ppm} \times \text{dilution factor}) / (\text{Wt. of sample (g)} \times 10^6)] \times 100 \quad (9)$$

where ppm is the concentration obtained from the AAS measurement, the dilution factor accounts for any dilution of the digested sample solution, and Wt. of sample is the weight of the original sample used for digestion. This approach ensures that the concentrations of Mg and Ca are accurately determined from the solutions prepared in Section 2.4, providing a direct link between sample digestion and mineral analysis.

2.4.2 Determination of Na, K, Zn, and Fe

The concentrations of Na, K, Zn, and Fe in the biomass raw materials and beef cattle feed samples were determined using the digested sample solutions prepared in Section 2.4. For reagent preparation, a mixed acid solution of nitric acid (HNO₃, 69–70%) and perchloric acid (HClO₄, 69–72%) in a 1:1 v/v ratio was used. Working standard solutions for each mineral were prepared at appropriate concentrations: 0, 2, 3, 4, and 5 ppm for Na, K, and Zn, and 0, 1, 2, 3, 4, and 5 ppm for Fe. The standards were prepared by pipetting suitable volumes of 100 ppm stock solutions into 50 mL volumetric flasks and adjusting the volume with distilled water. The digested sample solutions were analyzed using an AAS. The absorbance values of the samples were compared to those of the working standards, and the concentrations of each mineral were calculated using the following eq. (10):

$$\text{Mineral (\%)} = [(\text{ppm} \times \text{dilution factor}) / (\text{Wt. of sample (g)} \times 10^6)] \times 100 \quad (10)$$

where ppm is the concentration obtained from the AAS measurement, the dilution factor accounts for any dilution of the digested sample solution, and Wt. of sample is the weight of the original sample used for digestion. This procedure ensures accurate determination of Na, K, Zn, and Fe from the digested solutions prepared as described in Section 2.4.

2.4.3 Determination of P

The P content in the biomass raw materials and beef cattle feed samples was determined using the digested sample solutions from Section 2.4. A molybdovanadate reagent was prepared by dissolving 40 g of ammonium molybdate in 400 mL of hot distilled water and 2 g of ammonium metavanadate in 300 mL of hot distilled water, followed by the addition of 450 mL of HClO₄ (69–72%) and thorough mixing. The final solution was diluted to 2000 mL with distilled water. Working standard solutions of P at concentrations of 1, 2, 4, 6, 8, 10, and 12 ppm were prepared by pipetting suitable volumes of a 100 ppm stock solution into 50 mL volumetric flasks and adjusting the volume to the mark with distilled water. The digested sample solutions were analyzed using a UV-Vis spectrophotometer. The absorbance of each sample was compared to the standard calibration curve, and the phosphorus content was calculated using the eq. (11):

$$P (\%) = \frac{[(\text{ppm} \times \text{dilution factor}) / (\text{Wt. of sample (g)} \times 10^6)] \times 100}{1} \quad (11)$$

Where ppm is the phosphorus concentration obtained from the spectrophotometer, the dilution factor accounts for any sample dilution, and Wt. of sample is the weight of the original sample. This method ensures that phosphorus content is accurately determined from the digested solutions prepared in Section 2.4.

2.5 Analysis of Feed Cost for Beef Cattle

The feed cost analysis in this study was conducted by collecting and comparing data on commercially available cattle feed and feed formulated by Nong Sung Agricultural Cooperative Limited. Detailed information was gathered for each feed type, including the feed name, composition of raw materials, and the cost of each ingredient and the final feed product. The data collection process involved direct surveys and interviews with local farmers to ensure accuracy and representativeness of the feed types commonly used in the region. The analysis aimed to evaluate the economic efficiency of different feed formulations by calculating the total feed costs required to raise beef cattle under local farming practices in Nong Sung District, Mukdahan Province. This comparative approach provides insights into potential cost savings and the financial feasibility of using cooperative-formulated feed versus commercially available feed. The resulting data serve as a practical database that can support farmers and cooperative members in strategic planning, budgeting, and optimization of feeding strategies for sustainable beef cattle production.

3. Results and Discussion

3.1 Analysis of the nutritional composition of biomass raw materials for CFF

This study investigated the nutritional composition of various biomass raw materials intended for the development of CFF formulations. The objective was to establish a scientific basis for selecting suitable and locally available ingredients for cost-effective feed production. Eight biomass feed ingredients—FMK, SBM, PKM, RB, DCC, CSP, WCP, and CSR—were analyzed for their proximate composition. The proximate composition of these materials—focusing on protein, carbohydrate, and fat—was determined, as these nutrients are essential for muscle development, energy supply, and growth performance in beef cattle. Among them, protein content plays a particularly critical role in formulating feeds tailored to different growth stages, including the starter (under 1 year), grower (around 2 years), and fattening (2–3 years) phases [26, 27].

As shown in Table 1 and Figure 3, SBM, PKM, and RB contained the highest protein levels (43.81%, 14.21%, and 11.75%, respectively), while fat was highest in RB (12.69%), PKM (9.70%), and FMK (2.22%), significantly exceeding cassava-based by-products (0.31–1.03%). These variations result from the biochemical characteristics of plant tissues, genetics, and processing methods. Cassava by-products are carbohydrate-rich, whereas soybeans and palm kernels accumulate higher levels of lipids and proteins, consistent with prior studies on starch and oilseed processing [28]. Industrial operations—such as starch extraction and oil pressing—substantially alter nutrient profiles by concentrating or removing macronutrients, such as residual oil retention in PKM [29]. Furthermore, environmental factors such as soil fertility and harvest maturity, along with the physiological roles of different plant parts (e.g., bran vs. root), contribute to the observed nutrient variability [30]. Consequently, rice bran and oilseed meals naturally exhibit higher protein and lipid contents than root-based biomass.

The proximate composition aligns with previous reports, with minor variations due to sources and processing. Suksroi [14] and Promthep and Siriamornpun [31] reported protein levels in cassava residues and maize consistent with our findings. Notably, the fat content observed in rice bran (12.69%) and palm kernel meal (9.70%) in this study is higher than some reported averages. This elevation is likely due to mechanical extraction methods, such as expeller pressing, which typically yield higher residual oil content than solvent extraction. Furthermore, the high carbohydrate content in cassava-based materials (55.54%–83.49%) reflects the efficiency of the local processing stage in preserving starch fractions. Variation within species was also noted by Kraiprom et al. [32] regarding oil-palm fractions. Recent studies by Budianto [33] and Bamikole [34] further indicate that such nutrient fluctuations in maize and cassava-based diets directly affect ruminal fermentation, digestibility, and growth performance. These findings emphasize that plant genotype and processing methods determine nutrient profiles, underscoring the need for precise synchronization in CFF

formulation to enhance productivity. Generally, Proximate composition is driven by genotype, soil fertility, and processing. Feed selection should align with the specific nutritional needs of beef cattle growth stages. High-protein SBM, PKM, and RB support muscle development in pre-weaning calves, while the high fat content in PKM and RB provides energy for the growing stage. For fattening, SBM, FMK, and WCP offer an ideal protein-fat balance for weight gain. Utilizing local biomass reduces costs while ensuring nutrient adequacy. However, raw material selection must also account for regional availability, affordability, and storage stability within farming communities.

Table 1. Proximate composition (%) of biomass raw materials used for CFF formulation for beef cattle.

Biomass raw material	Moisture (%)	Ash (%)	Fat (%)	Protein (%)	Fiber (%)	Carbohydrate (%)
Finely ground maize kernels (FMK)	8.00±0.25	1.84±0.08	2.22±0.12	6.83±0.15	3.55±0.10	77.56±0.28
Soybean meal (SBM)	8.40±0.20	6.86±0.04	1.86±0.09	43.81±0.42	6.24±0.25	32.83±0.54
Palm kernel meal (PKM)	3.29±0.18	3.86±0.07	9.70±0.21	14.21±0.30	18.52±0.31	50.42±0.52
Rice bran (RB)	4.53±0.15	5.82±0.06	12.69±0.26	11.75±0.22	3.08±0.08	62.12±0.35
Dried cassava chips (DCC)	6.38±0.18	4.41±0.05	0.49±0.02	2.22±0.10	3.01±0.10	83.49±0.27
Cassava pulp–after starch extraction (CSP)	9.79±0.22	3.99±0.08	0.46±0.03	2.52±0.09	13.62±0.20	69.62±0.32
Washed cassava peels (WCP)	10.42±0.24	9.13±0.07	1.03±0.04	6.62±0.14	17.26±0.22	55.54±0.30
Fresh cassava starch residue (CSR)	16.09±0.30	5.80±0.09	0.31±0.03	1.99±0.08	16.64±0.20	59.17±0.25

Note: Values are expressed as mean ± standard deviation (SD, n = 3), and all nutritional, mineral, and EME values are expressed on an as-fed basis.

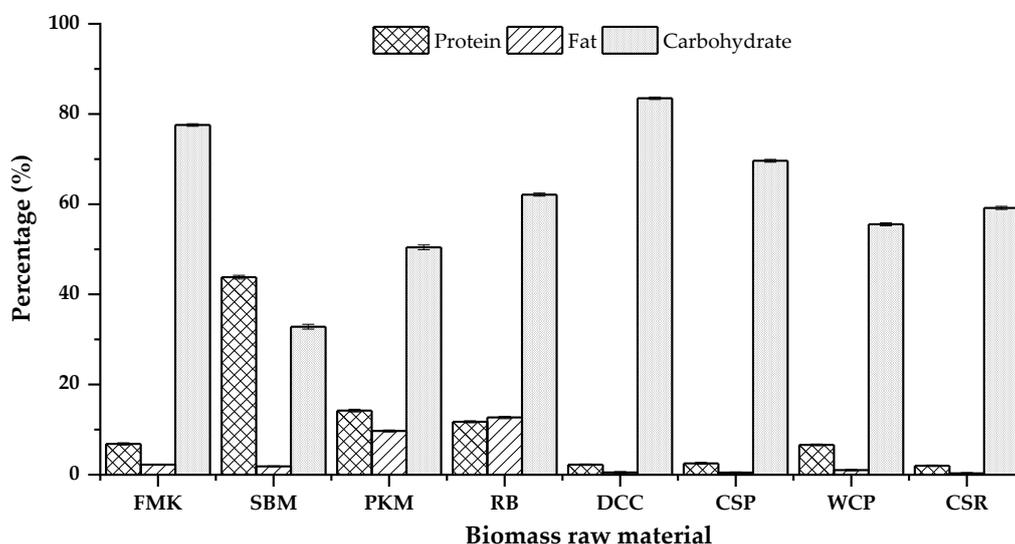


Figure 3. Proximate composition (carbohydrate, protein, and fat contents) of biomass raw materials used for CFF formulation.

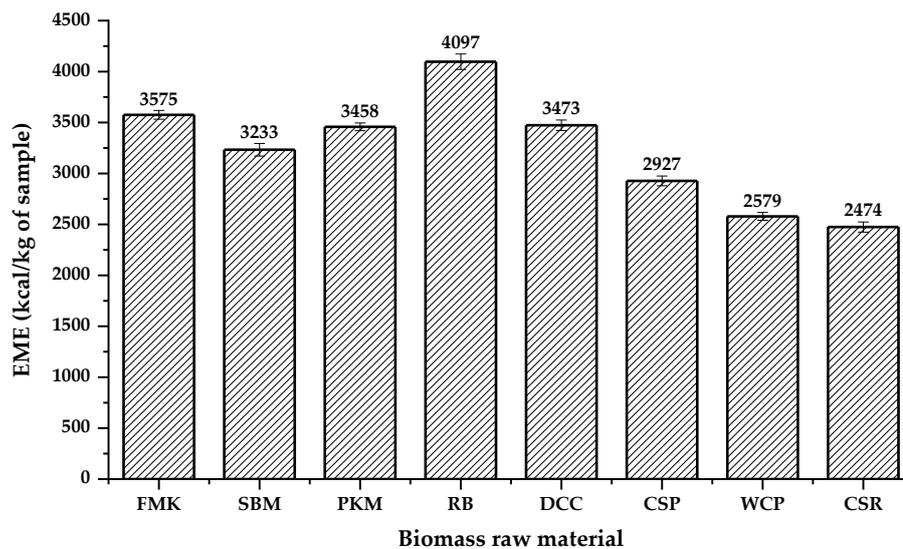


Figure 4. EME values of biomass raw materials used for CFF formulation.

3.2 Nutritional EME analysis of biomass raw materials for CFF formulation

In this study, the nutritional EME content of eight biomass raw materials intended for beef cattle feed was analyzed as illustrated in Figure 4. The experimental results were processed and reported on a kcal/kg sample (as-fed basis) to facilitate practical application and standardized comparison, as this unit is widely recognized in livestock feed formulation. The calculated EME content of the samples ranged from 2474 to 4,097 kcal/kg. Among the materials analyzed, RB exhibited the highest EME density at 4097 kcal/kg, primarily due to its superior fat (12.69%) and protein (11.75%) levels, which aligns with recent studies highlighting RB as a nutrient-rich feedstuff. In contrast, CSR provided the lowest EME value (2474 kcal/kg), while other materials such as FMK, PKM, and SBM showed intermediate to high values of 3575, 3458, and 3233 kcal/kg, respectively. These findings are consistent with prior research indicating that cassava by-products are effective EME sources for ruminants; for instance, cassava chips have been estimated via indirect calorimetry to provide an EME of approximately 10.63 MJ/kg DM (equivalent to 2327 kcal/kg as-fed) for lactating cattle. The observed variations in EME content are attributed to differences in chemical composition, which are further influenced by plant variety, cultivation conditions, and harvest stages. Therefore, selecting biomass for beef cattle should involve a balanced consideration of EME density, protein-mineral profiles, and the economic efficiency of replacing conventional feed sources with agro-industrial by-products to enhance overall returns.

3.3 Analysis of essential minerals (Ca, Mg, P, Fe, Na, Zn, and K) of biomass raw materials for CFF formulation

The analysis of essential minerals—Ca, Mg, P, Fe, Na, Zn, and K—in the biomass raw materials used for developing the CFF formulation showed that these minerals were present in all eight feed ingredients, including FMK, SBM, PKM, RB, DCC, CSP, WCP, and CSR, as presented in Table 2. In this study, the minerals were classified into three groups based on their physiological functions and importance: bone-supporting minerals (Ca, Mg, and P), blood- and nervous system-supporting minerals (Fe and Zn), and minerals that support the nervous and muscular systems (Na and K) [39]. Among the bone-supporting minerals, P was the most abundant, with RB containing the highest concentration and DCC the lowest. Mg was also highest in RB, whereas the other samples contained Mg at relatively similar levels (0.0732–0.2961%). Ca was found at the highest level in CSR and at the lowest level in FMK. Regarding blood and nervous system-supporting minerals, CSR had the highest Zn content, while DCC exhibited the lowest. For Fe, WCP contained the highest concentration, whereas RB had the lowest among the biomass samples. The relatively higher Fe content observed in WCP compared to other ingredients may be attributed to residual soil particles adhering to the outer peel surface, a common occurrence in agro-industrial root processing. These results indicate substantial variation in mineral composition across biomass types, highlighting the importance of selecting appropriate

raw materials to ensure that cattle diets meet both energy and essential mineral requirements for growth, skeletal development, and physiological functions.

Table 2. Mineral composition of biomass raw materials used for CFF formulation.

Biomass raw material	Na (%)	Zn (%)	Fe (%)	K (%)	Mg (%)	Ca (%)	P (%)
Finely ground maize kernels (FMK)	0.0122	0.0034	0.0138	0.4051	0.1044	0.0475	0.1813
Soybean meal (SBM)	0.0222	0.0057	0.0164	0.4099	0.2961	0.2997	0.4969
Palm kernel meal (PKM)	0.0126	0.0037	0.0178	0.4679	0.1942	0.2826	0.3105
Rice bran (RB)	0.0163	0.0076	0.0109	1.6250	0.7665	0.0801	1.3203
Dried cassava chips (DCC)	0.0147	0.0013	0.1714	0.0529	0.0732	0.1358	0.0036
Cassava pulp after starch extraction (CSP)	0.0471	0.0020	0.0335	0.2701	0.1398	0.5576	0.0195
Washed cassava peels (WCP)	0.3019	0.0036	0.4635	0.4628	0.1198	0.4877	0.0278
Fresh cassava starch residue (CSR)	0.0268	0.0901	0.3185	0.6378	0.1993	0.5584	0.2957

Note: All experimental values presented in Table 2 are the mean of three independent replicates with relative standard deviations (%RSD) less than 5%, and all nutritional, mineral, and EME values are expressed on an as-fed basis.

Regarding nervous and muscular system minerals, K concentrations exceeded Na in all samples, with RB, CSR, and PKM containing the highest K levels. Na was generally low, with WCP showing the highest and FMK the lowest concentrations (Table 2). These findings align with those of Burhan et al. [40] and Bayero et al. [41], who reported similar mineral hierarchies across various biomass sources, confirming that concentrations vary widely by species and source. The variation in essential minerals across the eight biomass types is likely driven by regional soil characteristics, fertilization regimes, and harvesting methods [17, 42, 43]. In Mukdahan's Nong Sung District, samples exhibited high levels of major minerals (Ca, Mg, P, and K) derived from soil fertilization, while minor minerals (Na and Zn) remained low, consistent with previous reports. Notably, specific trace elements, such as Fe in WCP, were exceptionally high (0.4635%). While these materials provide a foundation for bone, nervous, and muscular health, farmers should follow Department of Livestock Development recommendations for mineral supplementation to ensure a precise nutritional balance for optimal growth and physiological function.

3.4 Nutritional analysis of the CFF formulated from biomass raw materials compared with commercial beef cattle feed

This study evaluated a CFF formulated from eight local biomass materials, produced at a 1,000 kg scale. The mixture comprised CSR (600 kg), a DCC/CSP/WCP blend (200 kg), FMK (60 kg), PKM (20 kg), SBM (54 kg), and RB (54 kg). Supplements including urea (4 kg), dicalcium phosphate (3 kg), mineral premix (3 kg), and molasses (2 kg) were added to optimize nitrogen and phosphorus levels and fermentation efficiency. Following 10 days of anaerobic fermentation (28–30 °C), the CFF contained 12.16% crude protein, 3.29% crude fat, and 47.56% carbohydrates (Table 3, Figure 5), meeting the requirements for growth and EME supply in beef cattle. These results align with Anorach [44], who reported ~12% protein in fermented pumpkin-cassava substrates. Similarly, Puttha and Tangtaweewipat [45] observed protein levels of 8.40% in fermented maize and 10.6% in Napier grass silage. These findings confirm that local biomass is a viable substrate for nutritionally balanced, cost-effective fermented feed.

Table 3. Proximate composition of the CFF formulated from biomass raw materials compared with commercial beef cattle feeds.

Formulated and commercial feeds	Moisture (%)	Ash (%)	Fat (%)	Protein (%)	Fiber (%)	Carbohydrate (%)
Concentrated fermented feed (CFF)	11.74±0.20	4.60±0.08	3.29±0.05	12.16±0.21	32.39±0.80	47.56±0.90
The commercial beef cattle feeds C1	8.88±0.15	8.92±0.14	2.71±0.05	12.13±0.20	19.50±0.40	47.86±0.85
The commercial beef cattle feeds C2	7.76±0.14	8.22±0.12	1.60±0.03	10.90±0.18	13.22±0.30	58.30±1.00
The commercial beef cattle feeds C3	7.71±0.15	13.45±0.25	3.62±0.06	12.85±0.22	35.91±0.85	26.46±0.60
The commercial beef cattle feeds C4	5.93±0.12	16.87±0.30	0.48±0.01	17.34±0.30	20.85±0.50	38.53±0.75
The commercial beef cattle feeds C5	9.99±0.18	14.10±0.25	2.33±0.04	16.74±0.28	23.29±0.60	33.55± 0.70

Note: Values are expressed as mean ± standard deviation (SD, n = 3), and all nutritional, mineral, and EME values are expressed on an as-fed basis.

Comparison with Thaowarn [46] showed that TMR formulations (13.18–13.47% CP) were slightly lower than the theoretical 14% value. Such deviations often stem from natural variations in raw material quality, leading to discrepancies between calculated and analytical values. A similar trend was observed in commercial samples (C1–C5), where analyzed protein levels did not precisely match label declarations. These inconsistencies are supported by Kilama et al. [47] and Chen et al. [48], who identified significant batch-to-batch variability in commercial feeds and TMRs due to fluctuations in byproduct quality, mixing accuracy, and changes in dry matter content during storage. Consequently, these factors collectively influence the final nutrient profile delivered to livestock.

The higher fiber content in CFF compared to commercial pellets has key practical implications. While reducing energy density (EME), high fiber levels support rumen health by stimulating chewing and stabilizing ruminal pH via acetate production, preventing metabolic disorders such as acidosis. This makes CFF ideal for maintenance or grower rations, particularly for local or crossbred cattle. For finishing stages, CFF can be supplemented with high-energy ingredients or serve as a fermented roughage replacer. In contrast, commercial feeds are formulated for specific energy and protein targets. However, when CFF is used within a TMR strategy—blending roughage, concentrates, and minerals—it enhances ration consistency, reduces labor, and ensures a steady nutrient supply.

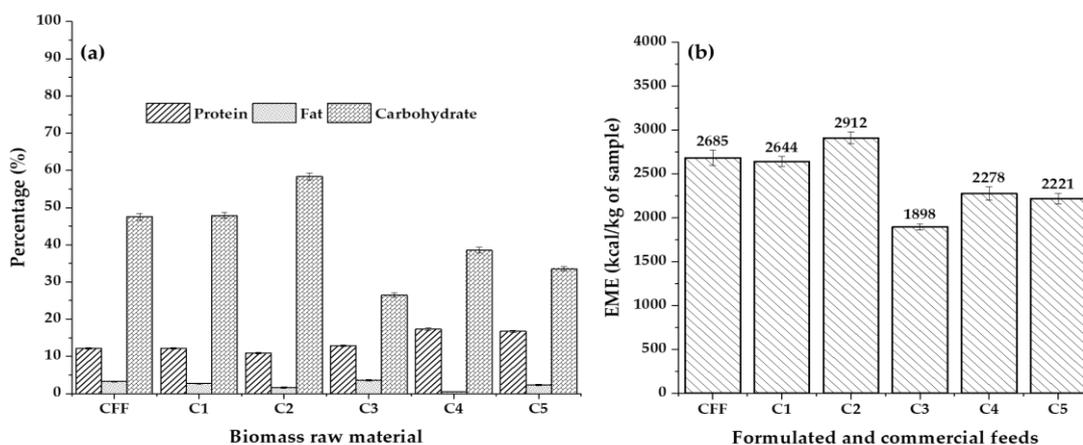


Figure 5. (a) proximate composition (carbohydrate, protein, and fat contents) and (b) EME values of the CFF formulated from biomass raw materials compared with commercial beef cattle feeds.

3.5 Nutritional EME analysis of the CFF formulated from biomass raw materials compared with commercial beef cattle feed

The EME evaluation revealed that the CFF, formulated from locally available biomass, possessed an EME value of 2685 kcal/kg of sample. This EME density places the CFF within the competitive mid-range compared to the examined commercial beef cattle feeds (Figure 5b). Among the commercial formulations, C2 exhibited the highest EME content (2912 kcal/kg), while the CFF outperformed several other commercial options, including C1 (2644 kcal/kg), C4 (2278 kcal/kg), and C5 (2221 kcal/kg). These results demonstrate that the CFF formulation provides a robust EME profile, making it a viable and high-quality alternative to standard commercial feeds. EME differences reflect variations in crude fat, non-structural carbohydrates, and fermentable fiber—key determinants in ruminant diets [17, 19]. The moderate EME in CFF aligns with the biochemical traits of cassava residues, RB, and PKM. While fermentation enhances digestibility and nutrient accessibility, it does not substantially increase caloric density [28, 36]. Despite lower EME, CFF is nutritionally adequate for smallholder or semi-intensive systems where cost-effective resources are vital. Improved ruminal degradability in fermented biomass can enhance microbial protein synthesis, partially compensating for lower gross EME [31, 38]. Furthermore, relying on local biomass offers economic and sustainability benefits, reducing dependence on high-cost commercial feeds [1, 5]. These findings corroborate evidence that fermented agro-industrial residues are viable, nutritionally acceptable, and environmentally aligned alternatives for ruminant production [35, 47, 48].

3.6 Analysis of essential minerals (Ca, Mg, P, Fe, Na, Zn, and K) of the CFF formulated from biomass raw materials compared with commercial beef cattle feed

The mineral composition of the CFF formulated from biomass raw materials demonstrated distinct variations when compared with commercial beef cattle feeds (C1–C5) (Table 4). Among the macrominerals, Ca was the predominant element in both CFF and commercial feeds, with Ca concentrations in CFF (0.4825%) comparable to those reported for commercial feeds (0.5323–0.5843% in most samples). Mg and P levels in CFF (0.1750% and 0.160%, respectively) were within the range typically observed in fermented feed ingredients and commercial formulations, reflecting their roles as essential structural and metabolic minerals [39–41]. For the trace minerals, Zn and Fe exhibited greater variability between feed types. CFF contained a higher Zn concentration (0.0308%) than all commercial feeds (0.0034–0.0064%), indicating that the biomass ingredients contributed a relatively higher Zn supply. Conversely, Fe content in commercial feeds varied widely (0.0546–0.1324%), whereas CFF showed moderate Fe levels (0.0878%), suggesting that mineral supplementation practices among manufacturers influence Fe concentration more strongly than fermentation-derived feed production. Regarding sodium and potassium, K was consistently more abundant than Na across all feed samples. CFF exhibited a K concentration of 0.1927%, similar to certain commercial formulations, while Na content was considerably lower (0.0268%), which is characteristic of biomass-based fermented feeds. The relatively low Na levels across feeds indicate the need for external salt supplementation in practical feeding systems, as recommended by livestock authorities.

Overall, differences in mineral profiles between CFF and commercial feeds can be attributed to variations in raw material composition, soil mineral status in cultivation areas, fertilization practices, and the specific formulation ratios used during feed production. These findings are consistent with previous reports indicating that the mineral content of forage and feed ingredients varies widely depending on agricultural conditions and processing methods. The results confirm that CFF provides essential macro- and micro-minerals beneficial for bone development, blood and nerve function, and muscular activity, although certain minerals may still require targeted supplementation to meet the full nutritional requirements of beef cattle [38–42].

3.7 Comparative analysis of feed cost for beef cattle

Feed ingredient cost is a major determinant in beef cattle diet formulation, particularly for smallholder and commercial operations. High-protein ingredients such as SBM and energy-rich corn kernels are relatively expensive due to high market demand and their nutritional value. RB, while locally abundant, commands moderate prices because of its potential for value-added products. In contrast, PKM and cassava by-products, including DCC, CSP, and WCP, provide substantial protein and energy at lower costs, making them attractive

alternatives for cost-sensitive feed formulations. The nutrient–cost analysis indicates that low-cost biomass materials, such as PKM and RB, can effectively substitute expensive ingredients without compromising protein and energy content. For example, PKM offers protein levels comparable to SBM at approximately one-third the cost. By optimizing the proportion of low-cost, nutrient-dense ingredients and reducing the use of high-cost feedstuffs, farmers can formulate economically efficient fermented concentrate feeds that meet the nutritional requirements of beef cattle.

Commercial feed prices (C1–C5) ranged from 10.67 to 15.33 THB/kg, whereas a 1,000 kg batch of fermented feed formulated with local biomass cost 4,500 THB, or 4.50 THB/kg (Table 5). This demonstrates that integrating locally available biomass by-products into fermented feed significantly reduces production costs while maintaining adequate nutritional quality. Ingredients such as DCC, WCP, and PKM contributed most to cost savings without compromising protein and energy levels. Furthermore, the economic advantage of low-cost biomass is amplified in large-scale production, where ingredient costs account for a major portion of total feed costs. Strategic blending of high- and low-cost ingredients allows flexibility to meet target protein and energy levels, supporting growth across different production stages. This approach also promotes sustainable livestock practices by utilizing agro-industrial residues, reducing waste, and improving feed cost-efficiency, particularly benefiting smallholder farmers with limited resources. Future studies should evaluate nutrient digestibility and growth performance of cattle fed these cost-effective fermented feeds under field conditions.

Table 4. Mineral composition of the CFF formulated from biomass raw materials compared with commercial beef cattle feed.

Formulated and commercial feeds	Na (%)	Zn (%)	Fe (%)	K (%)	Mg (%)	Ca (%)	P (%)
Concentrated fermented feed (CFF)	0.0268	0.0308	0.0878	0.1927	0.1750	0.4825	0.160
The commercial beef cattle feeds C1	0.4056	0.0064	0.0690	0.0751	0.1988	0.5843	0.2391
The commercial beef cattle feeds C2	0.3661	0.0062	0.0546	0.7129	0.1783	0.5323	0.1911
The commercial beef cattle feeds C3	0.5310	0.0058	0.1284	0.6247	0.1953	0.1185	0.1734
The commercial beef cattle feeds C4	0.3490	0.0034	0.1228	0.4667	0.1310	0.6500	0.1232
The commercial beef cattle feeds C5	0.2776	0.0057	0.1324	0.7956	0.2132	0.5774	0.2179

Note: All experimental values presented in Table 4 are the mean of three independent replicates, with relative standard deviations (%RSD) less than 5%.

Table 5. Example of cost calculation for the fresh fermented feed formulation.

Raw material	Fresh weight (kg)	Price per kg (THB)	Total cost (THB)
Cassava starch residue (CSR)	660	1.20	792
Dried cassava chips (DCC) + Cassava pulp after starch extraction (CSP) + Washed cassava peels (WCP)	200	7.00	1,400
Palm kernel meal (PKM)	20	7.30	146
Soybean meal (SBM)	54	24.00	1,296
Rice bran (RB)	54	10.00	540
Urea	4	25.00	100
Dicalcium phosphate	3	35.00	105
Mineral premix	3	35.00	105
Molasses	2	8.00	16
Total formulation weight	1,000 kg	-	4,500 THB
Cost per kilogram		4.50 THB/kg	

Note: Production costs were calculated based on local market prices in Mukdahan Province at the time of the study. In the original dataset, dried cassava chips (DCC) were priced at 7 THB/kg; since the combined 200 kg mixture includes DCC, CSP, and WCP without separate pricing, 7 THB/kg was applied as the representative value for this portion. It should be noted that ingredient prices may fluctuate depending on seasonal availability and regional market conditions.

4. Conclusions

This study demonstrated that agricultural biomass raw materials available in Mukdahan Province possess diverse nutritional and mineral compositions suitable for the development of CFF for beef cattle. Key protein-rich ingredients such as soybean meal, palm kernel meal, and rice bran, combined with carbohydrate-rich cassava by-products, provided a balanced nutrient foundation for feed formulation. Mineral analyses confirmed that the biomass sources supplied essential macro- and micro-elements required for skeletal development, metabolism, and physiological function. The formulated CFF exhibited nutritional values comparable to commercial beef cattle feeds, with adequate crude protein, fermentable carbohydrates, and essential minerals. Although its EME was moderately lower, the fermentation process is expected to potentially improve nutrient availability and palatability, though this requires further validation through digestibility trials. A major contribution of this work is the demonstration of substantial cost reduction—CFF production cost (4.50 THB/kg) was less than half of commercial feeds—highlighting its economic feasibility for smallholder farmers. The findings advance current knowledge on feed development using local biomass and reinforce the potential of agro-industrial residues for sustainable livestock production. Limitations include the lack of in vivo feeding trials and digestibility assessments. Future research should evaluate growth performance, actual nutrient digestibility, rumen fermentation characteristics, and long-term economic returns from incorporating CFF into actual farm management systems.

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