



# Article Influence of Guar Meal from Pig Compound Feed on Productive Performance, Nitrogen Metabolism, and Greenhouse Gas Emissions

Gabriel Mihaila<sup>1</sup>, Mihaela Habeanu<sup>2,\*</sup>, Nicoleta Lefter<sup>3</sup>, Anca Gheorghe<sup>2</sup>, Mihaela Dumitru<sup>3</sup>, Iuliana Marin<sup>4</sup>, Livia Vidu<sup>1</sup>, Carmen Georgeta Nicolae<sup>1</sup>, Dana Popa<sup>1</sup> and Monica Marin<sup>1,\*</sup>

- <sup>1</sup> Faculty of Animal Productions Engineering and Management, University of Agronomic Sciences and Veterinary Medicine of Bucharest, 59 Marasti Blvd., District 1, 011464 Bucharest, Romania; gabriel.mihaila@madr.ro (G.M.); livia.vidu@usamv.ro (L.V.); carmen.nicolae@usamv.ro (C.G.N.); dana-catalina.popa@igpa.usamv.ro (D.P.)
- <sup>2</sup> Research Station for Sericulture Baneasa, 013685 Bucharest, Romania; anca.gheorghe@scsbaneasa.ro
- <sup>3</sup> Animal Nutrition and Biotechnology Department, National Research-Development Institute for Biology and Animal Nutrition, Calea Bucuresti No. 1, 077015 Balotesti, Romania; nicoleta.ciuca@ibna.ro (N.L.); mihaela.dumitru@ibna.ro (M.D.)
- <sup>4</sup> Faculty of Engineering in Foreign Languages, National University of Science and Technology POLITEHNICA, 060042 Bucharest, Romania; iuliana.marin@upb.ro
- \* Correspondence: mihaela.habeanu@scsbaneasa.ro (M.H.); monica.marin@usamv.ro (M.M.); Tel.: +40-728380172 (M.H.); +40-720657597 (M.M.)

Abstract: Guar (*Cyamopsis tetragonoloba*) is an annual legume tolerant to drought. Guar meal (GM) is a protein- and carbohydrate-rich co-product generated after the mechanical separation of the endosperm from the germ and hull of guar seed. GM has received considerable interest in animal feed as an alternative to soybean meal (SM). In this study, we aimed to assess the nitrogen (N) balance indicators, performance, carcass traits, and main greenhouse gas (GHG) emissions resulting from enteric fermentation (E-CH<sub>4</sub>) and manure (M-CH<sub>4</sub> and N<sub>2</sub>O). Two tests were performed: (i) a biological trial on 45 pigs (15 animals/group) and (ii) a digestibility test in metabolism cages (N = 15, 5 replicates/group). Three different diets were given to the pigs: one diet was based on 0% GM (SM diet); in the second, GM-50%, GM replaced 50% of the SM; and the third was GM-100%, in which GM fully replaced the SM. The GM and SM diets were analyzed for their proximate composition. A model based on prediction equations was used to estimate the GHGs. GM up to 10% in the diets of finishing pigs did not significantly impact growth performance or carcass traits, although a slight increase in neutral detergent fiber (NDF) was observed. GM up to 10% improved N digestibility (p < 0.0001), net protein utilization (p < 0.0001), the biological value of protein, coefficients of metabolizability, and the coefficient of the total tract's apparent digestibility. Irrespective of its dietary proportion, GM decreased total nitrogen output (TNO, p = 0.11). A highly significant impact was noted for N<sub>2</sub>O and E-CH<sub>4</sub> (for DM, p < 0.0001), as well as a significant impact for E-CH<sub>4</sub>, expressed as g CO<sub>2</sub> Eq (p = 0.007), and g CO<sub>2</sub> Eq. LU (livestock unit, p = 0.005), also reported as ADG (p = 0.024). Manure, M-CH<sub>4</sub>, was not significantly influenced. In conclusion, GM can replace up to 100% SM and is thus a valuable byproduct that does not alter animal performance and can positively impact  $N_2O$ and E-CH<sub>4</sub>.

Keywords: greenhouse gas emissions; guar meal; nitrogen; performance; pigs

# 1. Introduction

The livestock sector is a notable consumer of natural resources. Classical diets for pigs are based on a mix of maize and soybean meal (SM) as the primary energy and proteinrich feedstuff. However, there is a growing discrepancy between production, availability, and demand [1,2]. Only non-genetically modified (non-GM) soybean is permitted in the



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**Copyright:** © 2023 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). European Union. With this background, supply chain disturbances and reduced packing plant capacity have caused considerable difficulties [3]. Romania is a country that relies on the importation of SM at a fluctuating price. In addition, the frequency of drought might lead to an increased gap between the feed supply and the nutritional needs of animals.

The identification of well-known feedstuffs and the use of locally accessible vegetable resources are required to address feed deficits. High priority has been given to the search for solutions to improve existing feed resources efficiently. The availability of non-conventional forage resources is increasing, although most of these resources need to be more palatable [4]. Guar (*Cyamopsis tetragonoloba*) is an atmospheric nitrogen (N)-fixing annual legume that is drought-tolerant and environmentally friendly. Guar meal (GM) is a co-product of the guar gum industry that is not genetically altered and is characterized by an elevated protein level (around 50%), as well as carbohydrates. GM is composed of germs and hulls that remain after the mechanical separation of the endosperm from the germ and guar seed hulls [5,6]. The nutritional value of GM was described in detail by Biel and Jaroszewska [7]. The protein concentrations in GM range from 33 to 60% and have favorable amino acid profiles. On the other hand, anti-nutritive components such as guar gum (mannan), saponins, and trypsin inhibitors limit GM's usage in animal feed [8].

The anti-nutritional properties of GM, as well as its low palatability, have led to doubts about the use of this byproduct. However, thermic treatment and limiting the levels of inclusion can eliminate these drawbacks. According to Abdel-Wahab et al. [9], a reduction in the levels of certain anti-nutritional factors (e.g.,  $\beta$ -mannan and saponin) can positively affect the health and performance of buffalo. In addition, this co-product is a natural feed ingredient as no chemicals or preservatives are used to obtain it. Previous studies highlighted changes in some growth, meat quality, and health parameters when using GM in poultry [10–12], cattle [13], sheep [14,15], goat [16], and buffalo [9]. Karpiesiuk et al. [2] and Hasan et al. [4] investigated the effects of using dietary GM supplementation as a cost-cutting technique on the performance and nutrient metabolism of pigs. Nonetheless, it remains critical to research the effects of using the GM co-products that remain after guar gum extraction on greenhouse gas (GHG) production.

Pig farming is growing steadily in terms of its complexity, industrialization, and intensification. The gases produced in the pig house impact not only the health and efficiency of the pig sector but also human health and quality of life. Relevant hazardous gases include gaseous N and its compounds such as nitrous oxide (N<sub>2</sub>O) and ammonia (NH<sub>3</sub>). Consequently, GHG emissions linked to global warming potential (GWP) have provoked public concern around the globe. Pork is one of the most commonly consumed meats [17]. According to Bälter et al. [18], food of animal origin emits more GHGs than feed of vegetable origin. Pig farming has a large impact on GHG production, especially enteric CH<sub>4</sub> (E-CH<sub>4</sub>) and manure-based CH<sub>4</sub> (M-CH<sub>4</sub>) and N<sub>2</sub>O in manure, while carbon dioxide (CO<sub>2</sub>) emissions are considered zero since plants reuse this gas through photosynthesis [19,20]. According to FAO [21], pigs produce lower E-CH<sub>4</sub> emissions (avg. 11%) than ruminants (more than 90%), but there are higher CH<sub>4</sub> emissions in their manure (more than 69% compared to ruminants, which produce less than 8%). The manure storage of N<sub>2</sub>O is more pronounced among chickens (avg. 66%) and pigs (avg. 20%) compared to that among ruminants (less than 7%) [21].

The reduction in GHG emissions via animal feed and manure management is a significant problem for sustainable pig farming. Biogenic CH<sub>4</sub> is a volatile organic compound produced by bacteria in pigs' digestive tracts (E-CH<sub>4</sub>) and feces through the anaerobic breakdown of organic matter [22,23]. The GWP of CH<sub>4</sub> is 25 times greater than that of CO<sub>2</sub> [24]. The microbial process in the N cycle and manure carbon content, along with the time required for storage and treatment, determines the amount of N<sub>2</sub>O emitted during storage and treatment [19,25]. The GWP of N<sub>2</sub>O is 298-fold greater than that of CO<sub>2</sub>. N<sub>2</sub>O originates only from manure [26] and accounts for about 26% of N<sub>2</sub>O production.

Taking into consideration the factors mentioned above, this study aimed to test two hypotheses: 1. The total substitution of SM with GM will affect the growth performance

and carcass characteristics of pigs; 2. GM has the potential to reduce N from manure and the main GHG emissions (CH<sub>4</sub> and  $N_2O$ ) in growing–finishing pigs.

#### 2. Materials and Methods

The trials took place in the IBNA Balotesti experimental Biobase located in Ilfov county in the southeastern part of Romania. This area is located in the central-eastern part of the Walachia Plain and is characterized by a temperate–continental climate, with dry and hot summers and cold winters.

Two experiments were carried out following protocol 7976/12/12, authorized by the Ethical Committee of the IBNA Balotesti and pursuant to Romanian Law No. 199/2018, which complies with the EU Directive 2010/63/EU on animal research.

GM is a protein- and carbohydrate-rich co-product generated after the mechanical separation of the endosperm from the germ and hull of natural guar seed that is broken and heat-treated (roasted for 3 min at 120–130 °C) to improve digestibility and palatability. GM has high nutritional value and a similar or reduced cost to SM. Section 3.1 provides a detailed comparison between GM's composition and that of soybean meal.

## 2.1. Animals and Housing

### Experiment 1. Experimental design.

This study used a total of forty-eight healthy, crossbred finishing Topigs pigs ((female Large White × Hybrid (Large White × Pietrain) × male Talent (mainly Duroc)), (2 replicates/group; 8 pigs/pen), 69.73  $\pm$  0.77 kg initial body weight (BW), 120  $\pm$  5 days old, with a similar sex ratio (mixed, with 4  $\Im$  and 4  $\sigma$  in each pen), and ear-tagged individually. The pigs were randomly assigned to three feeding groups for 35 days in a grow–finish shelter with strict environmental controls (21 °C; 60% relative humidity). The pigs received experimental diets for 35 days.

Experiment 2. N digestibility.

Following the procedure outlined in the law, using individual steel cages in an atmosphere-controlled room, a metabolic test was conducted over 21 days (7 days for accommodation) to assess N metabolism. A total of 15 barrows (Topigs hybrid pigs, BW avg. 89.6 kg  $\pm$  2 kg) were selected and split into three groups (5 replicates/group). Previous research has shown that three to six animals per group are statistically adequate for digestibility trials [27]. The pigs were individually housed and weighed. The digestibility trial weeks were divided into two balance periods (4 sampling days per period).

#### 2.2. Treatments

To evaluate growth performance and carcass traits and estimate the main GHGs (CH<sub>4</sub> and  $N_2O$ ), the GM and SM were analyzed for ether extract (EE), crude protein (CP), crude fiber (CF), amino acids, and minerals before they were included in the diets.

The composition and nutritional characteristics of the diets are outlined in Table 1. Throughout both experiments, the diets remained the same. Three feeding treatments that met the nutritional requirements of the Topigs hybrid were formulated: (1) The control group (SM); (2) GM-50%, where 50% of the SM was replaced with GM; and (3) GM-100%, in which the SM was completely replaced with GM. The diets included crystalline amino acids, DL-methionine and L-Lysine-HCl, to meet the requirements for all three diets, as well as calcium carbonate and monocalcium phosphate to provide the Ca and P requirements. The fiber level was about 3.8% higher in the GM-100% diet vs. that in the SM and GM-50% diets; NDF was +8.8% higher in GM-50% vs. the SM diet, with an increase of 17.06% in the GM-100% vs. SM diet.

Items (g * kg feed <sup>-1</sup> )	SM	GM-50%	GM-100%					
Maize	499.1	520.3	517.3					
Wheat	150.0	150.0	150.0					
Rice bran	110.0	110.0	110.0					
Soybean meal (44%)	120.0	50.0	-					
Guar meal	-	50.0	100.0					
Sunflower meal	80.0	80.0	85.0					
DL-methionine	0.9	0.6	0.1					
L-Lysine-HCl	2.4	2.1	1.2					
Calcium carbonate	16.7	16.2	15.4					
Monocalcium phosphate	5.9	5.8	6.0					
Sodium chloride	4.0	4.0	4.0					
Choline premix	1.0	1.0	1.0					
Vitamin and trace mineral mixture <sup>1</sup>	10.0	10.0	10.0					
Analyzed composition (g * kg $^{-1}$ as feed bases)								
DM	881.0	882.0	883.0					
СР	155.2	151.6	155.9					
EE	38.8	39.9	40.5					
Crude fiber	47.6	47.7	49.5					
NDF	139.5	153.0	168.2					
ADF	57.5	63.0	70.5					
Ca	8.0	8.0	8.0					
Р	6.0	6.0	6.0					
Lys	8.8	8.8	8.8					
Met + Cys	6.7	6.7	6.7					
Calculated composition (g * kg $^{-1}$ as feed bases) $^{2}$								
ME, Mj as feed basis	12.6	12.8	12.9					
Ń	24.83	24.26	24.94					
Lys d	7.3	7.4	7.6					
Met + Cys d	5.6	5.6	5.7					

**Table 1.** Experimental diet composition based on two levels of GM that replaced 50% or 100% of the SM in the diets.

Abbreviation: dry matter (DM), crude protein (CP), metabolizable energy (ME), crude protein (CP), ether extract (EE), neutral detergent fiber (NDF), lysine (Lys), methionine + cysteine (Met + Cys), calcium (Ca), phosphorus (P), digestible (d). Diets 2 and 3 were formulated with the addition of GM to replace 50% (GM-50%) and 100% (GM-100%) of the SM in the control diet (SM) and to adjust the CP, respectively. <sup>1</sup> The vitamin–mineral premix administrated per kg of feed: (i) 6000 IU vitamin A; 800 IU vitamin D3; 20 IU vitamin E; 1 mg vitamin K3; 1 mg vitamin B1; 3.04 mg vitamin B2; 10 mg vitamin B3; 6.3 mg vitamin B5; 1.5 mg vitamin B6; 0.03 mg vitamin B7; 0.3 mg co; 60 mg antioxidant. <sup>2</sup> To calculate N content, we used a factor of 6.25; the ME was calculated using regression equations based on the chemical characteristics (ME = 5.01 DP + 8.93 EE + 3.44 GF + 4.08); for the calculation of digestible amino acid levels, the feed chemical composition and theoretical coefficients were determined by IBNA Balotesti. Feed was provided twice a day at 08:00 and 14:00 h. Water was available ad libitum.

## 2.3. Measurement and Sampling

The BW of each pig was recorded on an electronic scale at the start and end of the biological and digestibility trials. The pigs were fasted overnight before being weighed, and blood was collected to minimize postprandial nutrient contents. The average daily gain (ADG) and feed conversion ratio (FCR) were computed based on BW. We used the equations of Diaz et al. to calculate the Kleiber ratio (KR), relative growth rate (RGR, %) based on ADG, and metabolic BW (MBW<sup>0.75</sup>), along with BW and age [28].

The carcass traits (backfat thickness, *Longissimus dorsi* area, and lean meat percentage) were determined on the left side using a PIGLOG 105 ultrasonic apparatus (SFK-Technology, Denmark) fitted with a formula for assessing meat percentage:

Y = 64.39 - 0.28 Fat-1 + 0.14 LD muscle thickness - 0.55 Fat-2

where LD is the *Longissimus dorsi* muscle. Fat-1 was measured 7 cm sideways behind the last rib from the middle dorsal line, whereas fat-2 was measured 10 cm from the last rib to the cranial section and 7 cm sideways from the middle dorsal line.

During the digestibility trial, samples were collected 4 days/week (two balance periods) from 08.00 h to 09.00 h to determine the N contents of the feces and urine. After weighing the total urine collected, 10% aliquots of urine were subsampled and stored at 4 °C for analysis. H<sub>2</sub>SO<sub>4</sub> at a concentration of 25% was added to each urine container to decrease the pH and diminish nitrogenous component volatilization. In the same way, after weighing the total feces samples, a quantity of about  $10\% \pm 0.4$  g was subsampled and stored at 4 °C prior to the N analyses. The samples were processed according to the procedure outlined by Hăbeanu et al. [29]. In the same manner, a blank digest was used. The samples were digested in the presence of catalyzers with H<sub>2</sub>SO<sub>4</sub> and then distilled and titrated. Class A glassware was employed for transvasation, dilution, and storage. The total N production was estimated after measuring N intake and excretion.

Blood samples (~6 mL) were taken via jugular venipuncture in heparin tubes (2 replicates per animal). Then, the samples were centrifuged for 15 min at 3000 rpm (iFuge D06, Neuation Technologies Pvt. Ltd., Gujarat, India), and the plasma obtained was placed in Eppendorf safe-lock tubes and stored in a freezer at -20 °C until urea nitrogen (PUN) analysis (Spotchem EZ SP-4430 chemistry analyzer).

#### 2.4. Analytical Laboratory Procedure

The proximate composition of the GM, SM, and diets was assessed at IBNA Balotesti in duplicate for dry matter (DM), CP, EE, CF, and their fractions compared to neutral detergent fiber (NDF) and acid detergent fiber (ADF), using the procedures outlined in Commission Regulation (EC) no. 152 (OJEU, 2009). Briefly, the semiautomatic traditional Kjeldahl technique, using a Kjeltek auto 1030 Tecator (SR EN ISO 5983-2:2009 [30] AOAC 2001.11 [31]), was employed to determine the CP. CF extraction was performed using an intermediate filtration method (standard SR EN ISO 6865:2002 [32]). Van Soest extractions were used to assess NDF and ADF according to SR EN ISO 16472:2006 [33] and SR EN ISO 13906:2008 [34]. A Raw Fiber Extractor FIWE 6 (Velp Scientifica, Usmate Velate, MB, Italy) was used for the analysis.

The amino acid composition was determined in duplicate via high-performance liquid chromatography, using HPLC Surveyor Plus Thermo Electron equipment (Waltham, MA, USA) fitted with a HyperSil BDS C18 column (Thermo Electron, Waltham, MA, USA) using silica sized at 250 mm  $\times$  4.6 mm  $\times$  5  $\mu$ m, as previously described by Varzaru et al. [35] and Gheorghe et al. [36].

In the digestibility trial, from the total quantity of collected feces,  $10\% \pm 0.4$  g subsamples were taken for analysis.

The samples were digested with  $H_2SO_4$  in the presence of catalyzers to assess the N concentration, followed by distillation and titration. The semiautomatic Kjeldahl method was carried out on a Kjeltec Auto 1030 Analyzer (Hillerod, Denmark). The reagents for mineral concentration were supplied by Merck (Darmstadt, Germany). Class A glassware was used for transvasation, dilution, and storage. Stock solutions traceable to standard reference material (NIST) were used for calibration. N retained (NR), total N output (TNO), and N digestibility were determined by measuring N intake (on a DM basis) and N excretions according to the methods described by Hăbeanu et al. [29] and using equations described by Hlatini et al. [27]. The coefficient of the total tract apparent digestibility (CTTAD), the coefficient of metabolizability (CAM), the biological value of protein (BVP), and the net protein utilization were calculated using the following equations:

$$CTTAD = (N intake - fecal N output)/N intake$$
 (1)

CAM = N intake - N fecal output - N urinary output/N intake (2)

$$BVP = N retained / N digested$$
 (3)

$$NPU = N retained / N intake.$$
 (4)

Blood samples were taken from the jugular vein, placed in heparin tubes (in duplicate), and then centrifuged for 15 min at 3000 rpm to separate the plasma. We used a chemical analyzer (Spotchem EZ SP-4430) to test the N and urea N in the plasma (PUN).

## 2.5. CH<sub>4</sub> (E-CH<sub>4</sub> and M-CH<sub>4</sub>) and N<sub>2</sub>O Emissions

In this study, to obtain models for estimating GHGs, the input and experimental output data were incorporated into prediction equations established by the IPCC [19,37] and prior works. The TNO assessed in our feeding trial was integrated into the equation for N<sub>2</sub>O prediction proposed by Philippe and Nick [26]:

$$N_2O = TNO * 0.2/100 * 44/28$$
 (5)

where 0.2% is the conversion factor of the N excreted into  $N_2O$  (IPCC, 2006) [19] in manure storage pits under animal housing, and 44/28 is the ratio of the molar mass of  $N_2O$  compared to that of N.

EvaPig<sup>®</sup> software, version 2.0.3.2, created by the French National Institute for Agricultural Research, Metex Nvistago, and the French Association of Zootechnie, was used for compound feed verification.

In this work, to determine the E-CH<sub>4</sub>, expressed as the CO<sub>2</sub> equivalent (g Eq.CO<sub>2</sub> \* day<sup>-1</sup>), we applied Philippe and Nick's equation [26]:

$$E-CH_4 = 0.012 * dRes * DM intake$$
(6)

where dRes are digestible residues.

The IPCC's [19,37] equations were used to estimate manure CH<sub>4</sub> emissions:

$$CH_4$$
, manure = VS \* B0 \* MCF \* fSMD (7)

where VS means solid volatile excretions per day resulting from organic solid manure substances, calculated using the IPCC's [37] Tier 1 method with the following equation:

$$VS = VSt * BW * 1000^{-1} (kg * day^{-1}).$$
(8)

Here, VSt = 4.9, where B0 denotes the maximum  $CH_4$  production in the pigs' manure (0.45 m<sup>3</sup>  $CH_4 * kg^{-1} VS * 0.67$ , which is the factor used for converting m<sup>3</sup> into kg; IPCC [37]); MCF is the  $CH_4$  conversion factor for the waste management system used, depending on the climate region, expressed in %; MCF (4% for solid storage in a dry, temperate region; IPCC [37]); and fSMD is the fraction of manure used by each management system. In our study, the solid storage system applied to the entire amount of manure (fSMD = 1).

## 2.6. Statistical Analyses

correlation was defined as a coefficient value > 0.7, and a moderate correlation was defined as a value below 0.69 and up to 0.5 (Akoglu cited by [38]); higher values were defined as significantly affected.

The group size was established based on Charan and Kantharia [39], where E = Total number of animals – Total number of groups, and E = the degree of freedom of the analysis of variance. In our study, the E value was 45, which is higher than 20. Therefore, including more animals would not increase the probability of obtaining significant findings.

#### 3. Results

## 3.1. Feedstuff Chemical Composition

Data on the chemical composition of protein-rich sources showed that GM has higher levels of analyzed CP (+15.4%), EE (+43%), and limitative amino acids (AA) (+34% lysine and +42% Met + Cyst) (Table 2). The lysine level was found to be higher in both feedstuffs. The diets were formulated to include 50 g or 100 g GM/kg feed. The addition of GM into the diet increased the NDF and ADF levels. These fiber fractions potentially impacted the responses of the animals.

Nutrients, %	SM <sup>1</sup>	GM <sup>1</sup>		
Dry matter	87.74	90.73		
CP	44.0	52.02		
EE	1.69	2.98 7.73 39.92		
Fiber	6.29			
NDF	12.44			
ADF	7.45	20.49		
Main AA				
Lysine	2.75	4.21		
Met.	0.64	1.11		
Cyst.	0.67	1.28		
Met. + Cyst.	1.31	2.38		
Minerals				
Ca	0.20	0.70		
Р	0.60	0.60		

Table 2. Comparison of the compositions of SM and GM.

<sup>1</sup> Salmonella, aflatoxins, and PCBs like dioxin below the detection limit.

Throughout the experiments, we did not observe any health problems or the refusal of feed among the animals.

#### 3.2. Growth Performance in the Biological Trial

Irrespective of the GM inclusion level, the carcass traits and growth performance were not significantly affected (Figure 1).

#### 3.3. N Digestibility

Table 3 presents the intake data, mean values, and SEMs for the balance indicators. While N, fiber, and ADF decreased (p > 0.05), the NDF fraction increased in the group fed GM, regardless of the percentage of inclusion. This result was reflected in the fecal N composition, with a significant decrease observed in the experimental groups compared to the SM group. However, no influence on urinary N was observed (p > 0.05). Therefore, although a decrease in TNO was determined, this decrease was not significant. A significantly higher impact was obtained for N excretion in the % intake, N digestibility, NPU, CAM, and CTTAD indicators.



**Figure 1.** Intake and descriptive statistics of the growth parameters and carcass traits of the fattening pigs fed two levels of a GM diet that replaced 50% or 100% SM (SM diet). p > 0.05, with no significant difference between the mean. The measurements were performed with PIGLOG 105 on live animals to determine their carcass traits. The number of observations was 48. Abbreviations: average daily feed intake (ADFI, Kg); dry matter intake (DMI, Kg); body weight (BW, Kg); metabolic BW (MBW<sup>0.75</sup>); average daily gain (ADG, Kg); relative growth rate (RGR, %); Kleiber ratio (KR, Kg); LD, *Longissimus dorsi*; fat thickness (mm); LD area (mm); lean meat (%); standard error of the mean (SEM).

**Table 3.** N metabolism of the fattening pigs fed two levels of a GM diet that replaced 50% or 100% of the SM diet.

Items <sup>1</sup>	SM	GM-50%	GM-100%	SEM	<i>p</i> -Value <sup>2</sup>			
Intake, g * day <sup>-1</sup>								
Feed	2550	2360	2168	86.92	0.18			
Ν	76.96	74.84	72.21	2.44	0.12			
Fiber	147.7	146.9	143.6	4.75	0.14			
NDF	413.9	399.2	287.8	16.02	0.29			
ADF	148.0	141.3	132.8	4.66	0.07			
N balance, g * day $^{-1}$								
Fecal N	8.73 <sup>a</sup>	7.08 <sup>b</sup>	6.86 <sup>b</sup>	0.25	0.01			
Urinary N	33.34	33.27	29.86	1.18	0.13			
TNÓ	42.07	40.35	36.72	1.41	0.11			
NR	34.89	34.49	35.48	1.06	0.12			
N excretion of % intake	54.65 <sup>a</sup>	53.80 <sup>b</sup>	50.67 <sup>c</sup>	0.34	0.007			
N digestibility, %	88.63 <sup>a</sup>	90.49 <sup>b</sup>	90.46 <sup>b</sup>	0.16	< 0.001			
NPU	45.34 <sup>a</sup>	46.19 <sup>b</sup>	49.33 <sup>c</sup>	0.34	< 0.001			
BVP	51.0 <sup>a</sup>	51.2 <sup>a</sup>	54.5 <sup>b</sup>	0.34	0.05			
CTTAD	0.89 <sup>a</sup>	0.90 <sup>b</sup>	0.90 <sup>b</sup>	0.001	0.01			
CAM	0.45 <sup>a</sup>	0.46 <sup>a</sup>	0.49 <sup>b</sup>	0.003	< 0.001			
PUN, mg/dL	26.61	26.84	27.27	0.62	0.19			

<sup>1</sup> The number of observations N = 15 (5 replicates per group). Abbreviations: total N output (TNO); N retained (NR); net protein utilization (NPU); biological value of protein (BVP); coefficient of total tract apparent digestibility (CTTAD); coefficient of metabolizability (CAM), plasma urea nitrogen (PUN). <sup>2 a-c</sup> Means values with different superscripts in the same row differ, ignificantly (p < 0.05), distinctly significant ( $p \le 0.01$ ), highly significant ( $p \le 0.001$ ). Values without letters—insignificant effect ( $p \ge 0.05$ ). SEM—standard error of means.

## 3.4. CH<sub>4</sub> (E-CH<sub>4</sub> and M-CH<sub>4</sub>) and N<sub>2</sub>O Emissions Estimated

Table 4 outlines the level of the main GHGs ge nerated via the enteric fermentative process and in the manure. When expressed as DMI bases, N<sub>2</sub>O decreased significantly in the GM-50% and GM-100% groups vs. the SM-fed group. A significant decrease was observed for E-CH<sub>4</sub>, expressed as the g of CO<sub>2</sub> Eq \* day<sup>-1</sup> (9% lower in the GM-50%-fed group and 21% lower in the GM-100%-fed group compared with the SM diet) and reported as the livestock unit (LU), considered to be 500 kg LW (live weight) based on Philippe and Nick [26] (10% and 22% lower in the GM-50% and GM-100% groups compared to the SM group). The highest influence (*p* < 0.0001) was obtained when evaluated using the DMI bases (8% and 15% lower in the experimental groups vs. that in the SM group). Conversely, the dietary addition of GM did not significantly influence M-CH<sub>4</sub>.

**Table 4.** Mean GHG level (g CO<sub>2</sub> Eq. N<sub>2</sub>O, E-CH<sub>4</sub>, and M-CH<sub>4</sub>)  $\pm$  SEM for the fattening pigs fed two levels of GM that replaced 50% or 100% of the SM.

Items <sup>1</sup>	SM	GM-50%	GM-100%	SEM	<i>p</i> -Value <sup>2</sup>			
Intake, g * day $^{-1}$								
DMI	2216.0	2078.0	1929.0	7.62	0.19			
TNO	42.07	40.35	36.72	1.41	0.11			
N ex.	61.80	58.20	51.10	2.14	0.12			
$N_2O$ (g $CO_2$ Eq * day <sup>-1</sup> )	39.41	37.80	34.40	1.32	0.13			
$N_2O$ (g $CO_2$ Eq. $LU^{-1*}$ day <sup>-1</sup> )	193.3	182.75	167.35	9.1	0.09			
$N_2O$ (g $CO_2$ Eq. ADG, kg <sup>-1</sup> * day <sup>-1</sup> )	42.78	39.44	36.45	1.7	0.19			
$N_2O$ (g CO <sub>2</sub> Eq. DMI, kg <sup>-1</sup> * day <sup>-1</sup> )	14.41 <sup>a</sup>	13.88 <sup>b</sup>	13.36 <sup>c</sup>	0.08	< 0.001			
E-CH <sub>4</sub> (g CO <sub>2</sub> Eq * day <sup>-1</sup> )	41.87 <sup>a</sup>	38.09 <sup>ab</sup>	33.08 <sup>b</sup>	1.24	0.007			
E-CH <sub>4</sub> (g CO <sub>2</sub> Eq. $LU^{-1} * day^{-1}$ )	205.39 <sup>a</sup>	184.15 <sup>ab</sup>	160.96 <sup>b</sup>	6.91	0.005			
E-CH <sub>4</sub> (g $O_2$ Eq. ADG, kg <sup>-1</sup> *day <sup>-1</sup> )	45.46 <sup>a</sup>	39.76 <sup>ab</sup>	35.04 <sup>b</sup>	1.77	0.024			
E-CH <sub>4</sub> (g CO <sub>2</sub> Eq. DMI, kg <sup>-1</sup> * day <sup>-1</sup> )	15.31 <sup>a</sup>	14.02 <sup>b</sup>	12.90 <sup>c</sup>	0.18	< 0.001			
dRes (g, as DM bases)	51.05 <sup>a</sup>	46.73 <sup>b</sup>	43.01 <sup>b</sup>	0.61	0.007			
$VS(g^*day^{-1})$	501	507	504	5.7	0.71			
M-CH <sub>4</sub> (g $CO_2$ Eq * day <sup>-1</sup> )	151.13	152.91	152.16	1.74	0.92			
M-CH <sub>4</sub> (g $O_2$ Eq. $LU^{-1} * day^{-1}$ )	738.69	738.73	738.66	0.01	0.25			
M-CH <sub>4</sub> (g $\overrightarrow{CO}_2$ Eq. ADG, kg <sup>-1</sup> * day <sup>-1</sup> )	162.64	160.43	159.88	3.17	0.93			
M-CH <sub>4</sub> (g CO <sub>2</sub> Eq. DMI, $kg^{-1} * day^{-1}$ )	55.72	58.51	61.65	1.91	0.39			

<sup>1</sup> The number of observations N = 15 (5 replicates per group). Abbreviations: daily N excretion rate (N ex.); volatile solids (VS); dRes: digestible residue; <sup>1</sup> considering the global warming potential of 25 for CH4. LU means livestock unit = 500 kg LW (used in [14]). <sup>2 a-c</sup> Means values with different superscripts in the same row differ, ignificantly (p < 0.05), distinctly significant ( $p \le 0.01$ ), highly significant ( $p \le 0.0001$ ). Values without letters—insignificant effect ( $p \ge 0.05$ ). SEM–standard error of means.

#### 3.5. Relationship between Input and Output Parameters

The Pearson coefficients from Table 5 show a strong correlation between specific input and output parameters. For example, the level of dietary GM is strongly positively correlated with the % N digestibility and NPU and negatively correlated with the N excretion of the % intake (p < 0.0001) and has a high-to-moderate correlation with E-CH<sub>4</sub> (r = -0.48; p < 0.01). However, for the other input parameters, such as the ADFI, DMI, fiber, and the fractions of ADF and NDF, the N intake presents a strong positive relationship with the TNO, NR, N<sub>2</sub>O, and E-CH<sub>4</sub> (p < 0.0001) and a negative moderate correlation with BVFP (with r ranging between 0.47 and 0.6 except for NDF).

Pearson Correlation (r)	TNO, g/day	NR, g/day	% N Dig	N Excretion of % Intake	NPU	BVFP	N <sub>2</sub> O, Eq CO <sub>2</sub>	E-CH <sub>4</sub> , Eq CO <sub>2</sub>	M-CH <sub>4</sub> , Eq CO <sub>2</sub>
Level of GM	-0.287	0.043	0.832 ***	-0.886 ***	0.886 ***	0.758 ***	-0.286	-0.487 **	0.045
ADFI	0.991 ***	0.972 ***	0.124	0.473 **	-0.473 **	-0.565 **	0.991 ***	0.937 ***	0.163
DMI g/zi	0.990 ***	0.974 ***	0.130	0.467 **	-0.467 **	-0.560 ***	0.990 ***	0.934 ***	0.164
Fiber intake, as DM bases	0.972 ***	0.992 ***	0.183	0.381 *	-0.381 *	-0.479 **	0.972 ***	0.898 ***	0.166
ADF intake, as feed bases	0.998 ***	0.955 ***	0.019	0.533 **	-0.533 **	-0.603 ***	0.998 ***	0.965 ***	0.154
NDF intake, as feed bases	0.849 ***	0.968 ***	0.456 *	0.105	-0.105	-0.246	0.850 ***	0.711 ***	0.173
N intake, as feed bases	0.989 ***	0.980 ***	0.101	0.452 *	-0.452 *	-0.535 **	0.989 ***	0.933 ***	0.160
N intake, as DM bases DM intake	0.988 ***	0.980 ***	0.098	0.449 *	-0.449 *	-0.530 **	0.988 ***	0.933 ***	0.160

**Table 5.** Pearson correlations between input and output parameters.

\*  $p \le 0.005$ —significant difference; \*\*  $p \le 0.01$ —distinct significant difference; \*\*\*  $p \le 0.0001$ —highly significant difference.

## 4. Discussion

GM is a concentrated protein source derived as a co-product of galactomannan extraction from guar seed. GM could be considered an appropriate feedstuff for animal feeding since the basic components of animal diets are frequently used in human nutrition. Furthermore, GM is nutritionally comparable to SM and is a reasonably inexpensive and readily accessible feed material that is also processed in large quantities for gum extraction.

This study showed the potential of GM to replace classical SM feedstuff, with a focus on nitrogen metabolism due to the relationship between TNO and  $N_2O$ . For the first time, we report data predicting GHG emissions ( $N_2O$  and  $CH_4$ ) resulting from the use of GM.

In previous research, dietary guar meal was not sufficiently studied as an alternative to traditional soybean meal [4]. Previous studies attempted to identify the ideal levels of dietary GM that could be used without adversely impacting performance [40], and few studies have investigated the impact of guar inclusion on carcass characteristics [41]. Antinutritional factors are widely recognized to restrict the amount of GM that can be included in the diet. For example, feeding animals can produce certain issues due to anti-trypsin and very viscous non-starch polysaccharides such as galactomannan polysaccharides, which raise the viscosity of the digesta and limit digestive enzyme activity as well as decrease nutritional digestibility [42]. These problems can be reduced by processing the meal; the anti-trypsin factor can be inhibited by toasting and by reducing the level of dietary inclusion. Pigs, chickens, and laboratory animals (rats and mice) have all been proven to suffer adverse effects from galactomannan, which is present in residual guar gum from meal [43]. Guar gum has been shown to inhibit rat and pig glucose absorption, which negatively impacts growth performance. However, research indicates that eating the gum's galactomannans, as found in guar meal, may enhance gut health by reducing the colonization of pathogenic bacteria in the gut. Furthermore, GM contains trypsin inhibitors involved in protein digestion.

In the present study, performance and carcass traits did not yield a significant decrease when including GM in the diet. Even though GM increased the NDF fraction in the diet, we predicted that the pancreas' considerable increase in total enzymatic activity with age would yield an improvement in feed intake and nutrient digestibility. In the literature, most studies focused on broiler chickens. Lee et al. [44] observed a decrease in the BW and feed efficiency of chickens fed with high concentrations of GM, likely due to the existence of residual guar gum in the GM. According to Owusu-Asiedu et al. [45], the ADG and ADFI in pigs decreased with cellulose and guar gum during the first 7 days of being fed a high-NSP diet. Increased non-starch polysaccharide (NSP) content in the pigs' diets directly affected growth rate and voluntary feed intake. Pigs and their microbiota may

adjust to high-fiber diets over a longer period of time. A higher percentage of energy was also digested in the large intestine because of increased dietary NSP, which similarly decreased the total-tract energy digestibility and voluntary feed intake. In 2018, Karpiesiuk et al. [2] partially replaced SM with GM (25, 50, and 75%) in a pig-fattening diet. Pigs in the group fed diets containing 25% GM protein obtained the best performance, as evidenced by the lowest feed conversion ratio and the highest growth rate. On the other hand, Hasan et al. [4] observed a negative impact on ADFI and ADG when using GuarPro F-71 in young pigs' diets as a 75% replacement for SM, which showed a linear decrease as guar inclusion increased but not a decrease in feed efficiency, in conflict with our results and those of Heo et al. [46]. GM diets had no noticeable impact on ADFI in growing–finishing pigs [47]. The ADFI of Yorkshire Landrace pigs decreased when GM was added to their diets. There is a lack of information in the literature about how GM affects the quality of swine meat. Karpiesiuk et al. [41] added GM with 50% and 75% protein to pig diets and reported that the addition of GM may have negatively impacted performance; however, meat quality was not affected (unpublished data).

Milczarek et al. [40] proposed using a dietary GM level of 4% to obtain good performance and improve the carcass composition of broiler chickens, as well as the physicochemical qualities of their muscles. Conversely, in terms of carcass composition, dressing percentage, and carcass muscularity, chickens fed diets with a proportion of GM higher than 12% performed noticeably worse.

Nitrogen (N or its gaseous form, N<sub>2</sub>) and carbon (C) are components vital to life. Firstly, N<sub>2</sub> must be transformed by nitrifying bacteria to enter the feed in food chains as part of the N cycle and be used by plants and animals as a nutrient. Some of the consumed N is lost through organic or inorganic excretion. In anaerobic environments, nitrification and denitrification processes produce manure-based nitrous oxide (N<sub>2</sub>O). Oxygen encourages the formation of N<sub>2</sub>O [20,23,26].

The farm's manure storage releases both  $N_2O$  and  $CH_4$ . C is the fourth most frequent element in the Earth's crust. Global energy and the C cycle are driven by methanogenesis. The second-most common GHG after  $CO_2$ ,  $CH_4$  is produced by animals through enteric fermentation and their manure.

This increase is responsible for around 20% of the current trend in global warming. Presently, between 500 and 600 GT of the world's yearly  $CH_4$  emissions come from various habitats [47].  $CH_4$  remains in the atmosphere for nine to fifteen years and is over 25 times more effective than  $CO_2$  at retaining atmospheric heat [48]. The raising of livestock plays a substantial role in the build-up of  $CH_4$  in the atmosphere. As our understanding of GHGs continues to evolve, feeding factors remain incompletely explored.

To decrease the main greenhouse gases, various dietary approaches were investigated. These approaches included high fiber contents, weight increases, feed efficiency, rates of protein and fat deposition, slaughter weight, and carcass lean yield [49–51]. Kpogo et al. [49], for example, previously investigated the impact of multicarbohydrase enzymes on GHG in the diet using wheat millrun as a co-product. Even though adding 30% wheat millrun to pig diets increases their fiber content, the authors did not observe a significant effect on GHGs. Furthermore, adding multienzymes to wheat millrun diets did not significantly affect emissions. In 2022, Hăbeanu et al. [38] highlighted that the intestinal microbial community influences pigs' growth and intestinal health. Feeding pigs a higher level of fiber led to higher levels of the total bacteria identified, which influenced the total volatile fatty acids (VFAs). The authors noted an important decrease in E-CH<sub>4</sub> in pigs fed high-fiber diets featuring the addition of mustard and grapeseed cake.

The findings of the present study did not agree with those obtained by Kpogo et al. [49], which showed that N digestibility was not enhanced. Conversely, Chen et al. [52] found that utilizing a cocktail of non-starch polysaccharide enzymes in a corn–miscellaneous meal diet improved nutrient digestibility; nonetheless, the authors did not observe a significant influence on  $N_2O$  and  $CH_4$  emissions throughout the experiment. Our data regarding E-CH<sub>4</sub> are less comprehensive than those obtained by Hăbeanu et al. [38]. However, if we

take into consideration the data reported to LU, our data are similar to those obtained by Philippe et al. [17,26].

There is no evidence in the literature to suggest a low correlation between DMI, feed intake, N intake, and other parameters on  $M-CH_4$  emissions.

#### 5. Conclusions

The results of this study did not support our first hypothesis, which predicted that the performance of the pigs would decrease if GM completely replaced SM. Replacing 100% of the SM in the diet with GM can positively alter certain indicators of N metabolism. For N<sub>2</sub>O (based DM) and E-CH<sub>4</sub>, the positive effects of dietary GM were greater when utilizing a GM-100% diet. This result supports our second hypothesis, although the impact of the anaerobic process in the manure on M-CH<sub>4</sub> was less pronounced. This particular type of GHG decreases if organic matter is more digestible.

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