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# Multi-Parametric Analysis Based on Physico-Chemical Characterization and Biochemical Methane Potential Estimation for the Selection of Industrial Wastes as Co-Substrates in Anaerobic Digestion

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**Abstract:** Anaerobic digestion is considered as one of the most feasible waste-to-energy technologies for the valorization of organic wastes. It can be applied to many different substrates but the mono-digestion of a single substrate usually has some important drawbacks due to the physico-chemical characteristics of the substrate. A feasible solution is the simultaneous co-digestion of several substrates with different composition and characteristics, so that synergetic effects may be generated and physico-chemical characteristics may be compensated, thus reaching higher process efficiencies and biogas production rates. In this work, a multi-parametric analysis for the objective comparison of industrial wastes was developed in order to help with decision making about their suitability as a co-substrate in anaerobic co-digestion. Criteria considered for this analysis included sample composition, C/N ratios, theoretical biochemical methane potential (BMP), and other important issues such as production rates, seasonality, and the distance to the WWTP or pre-treatment requirements. Results showed that, among the 13 evaluated wastes, 2 of them showed a higher potential for being used in anaerobic co-digestion: 1. Fried corn from the snack food industry and 2. Wet fatty pomace from the olive oil industry. Both wastes showed high estimated BMP values, high lipid and carbohydrate content, and C/N ratios in a proper range to improve the low C/N ratio of sewage sludge. Other wastes such as olive pomace (dry), skinless corn (not fried), and grape pomace from the winery industry may also be used as co-substrates. As a conclusion, this procedure based on a selection matrix can be considered as a useful tool to help both producers and WWTP operators to make decisions about the potential applicability of specific industrial wastes as co-substrates in anaerobic co-digestion.

**Keywords:** anaerobic co-digestion; biochemical methane potential; co-substrates; industrial wastes; valorization

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## 1. Introduction

In recent years, solid waste generation reached critical levels, especially in developed countries where huge amounts of wastes are generated at homes, industries, and other human activities [1]. Due to the environmental and human health consequences resulting from inappropriate waste management, the evaluation of new alternatives for waste treatment has become an essential issue for researchers, managers, and municipalities. At the same time, a more and more restrictive legal framework is continuously changing to improve waste management, not only in the European community but also in most countries around the world. In this context, one of the most important legal requirements is focused on the reduction in the amount of solid wastes that are sent to a landfill, thus promoting more efficient waste management, valorization, recycling, and reuse, thus prioritizing different ways of treatment rather than a landfill and trying to achieve the sustainable development goals (SDG) in the circular economy (CE) framework. In the European Union, the objective to be reached is to reduce the amount of solid wastes sent to landfills below 10% of the total amount of solid urban wastes before 2035 [2]. For organic wastes, the most common treatment is composting but, in recent decades, anaerobic digestion is gaining attention and it is being considered as one of the most feasible waste-to-energy technologies for the energetic valorization of organic wastes [3]. Regarding this fraction, the situation in Spain is promising, as the percentage of organic waste that undergo efficient valorization treatments such as composting or anaerobic digestion is increasing every year, thus reducing the disposal of organic wastes at landfills [4].

Besides the current waste management situation, the huge increase in energy demand must also be considered for justifying the development and use of waste-to-energy infrastructures. In this respect, the Spanish legal framework for energy and climate policy is governed by the objectives of the European Union, which are reflected in the Integrated National Energy and Climate Plan 2021–2030 (PNIEC) [5]. This plan proposes the use of renewable energies as the path to achieve energetic self-sufficiency and to reach the objective of decarbonization in the near future. New installations of renewable energies such as solar, wind, geothermal, or biomass need to be built but also the existing potential needs to be exploited for this purpose.

In this context, anaerobic digestion is a good alternative to provide a solution for both problems. On the one hand, it efficiently produces a low-cost renewable energy, which may be directly used in the installation itself or may be injected into the natural gas distribution network after purification [6] and, on the other hand, it is currently considered one of the most feasible options for organic waste valorization, because the high moisture content of these wastes makes it difficult to use other energetic valorization treatments such as pyrolysis or combustion [3]. Moreover, it brings great environmental benefits, such as the reduction in odor and greenhouse gas (GHG) emissions and the revalorization of a digestate as organic fertilizer [7]. Thus, an organic biomass becomes a resource with a great energy potential thanks to its high availability from both municipal and industrial sources [8].

Through anaerobic digestion, organic matter is biologically transformed into a biogas, which is mainly composed of methane ($CH_4$), carbon dioxide ($CO_2$), and hydrogen ($H_2$), whose concentrations vary depending on several aspects such as the resource used as a substrate or the operational conditions [9,10]. This process can be applied to many different organic wastes such as sewage sludge from wastewater treatment plants (WWTPs), manure, the organic fraction of municipal solid waste (OFMSW), wastes from the food industry, agricultural wastes, etc. [8,11]. Other organic wastes with a high heating value (HHV) such as those obtained from the olive oil industry [12], fruits and vegetables produced in markets and stores [13], or even forest and pruning residues [14] have also been investigated for this purpose. However, the mono-digestion of a single substrate usually has some important drawbacks, since the substrate may not have the appropriate physico-chemical characteristics for reaching high efficiencies in $CH_4$ production. A feasible solution for this problem is the simultaneous co-digestion of several substrates with different composition and characteristics, so that synergetic effects may be generated, thus compensating for physico-chemical characteristics and reaching higher process efficiencies and biogas production rates [12,15,16]. These synergies may be reflected in the C/N ratio, biochemical methane potential (BMP), buffering capacity, and dilution of inhibitory substances, as well as the seasonality damping [17,18]. Other advantages of co-digestion lie in the possibility of treating both liquid and solid substances, in such a way that the same facilities can be shared for the treatment of many different wastes with different properties and origins, and the possibility of using existing facilities such as WWTPs, thus reducing investment costs and improving the profitability of these plants or the better quality of the digestate in terms of nutrient content, thus increasing its value as a fertilizer compared to that obtained from mono-digestion [19].

The most widely used co-substrates in WWTPs are wastes produced in agri-food industries [20]. To determine the suitability of a waste to be used as a co-substrate in anaerobic co-digestion, lab-scale tests can be carried out to evaluate the experimental BMP [21,22]. However, these experimental methods are expensive, time-consuming, and very sensitive to operational conditions. Moreover, BMP values usually present significant differences when compared to real systems, as the operation mode and test conditions significantly differ from those selected in a full-scale anaerobic digester [23]. For these reasons, many researchers have turned to theoretical models for rapid BMP predictions, especially for comparative purposes and at pre-selection stages, in such a way that those wastes that reach higher predictions may be experimentally tested in a second research phase. In this way, experimental work is reduced and operational costs are saved, as only those wastes that were potentially viable are tested in a lab. In these situations, although the theoretical models are idealized and simplified representations of reality and they may overestimate BMP, their usefulness as a tool for the comparison and pre-selection of the most suitable co-substrates is remarkable, as they are fast, reliable, and easy to use [18].

Different theoretical methods can be found in the literature [24,25]. The first models were based on biological reactions and Monod-type equations describing biomass growth and they were able to distinguish between the substrate biodegradable and non-biodegradable fractions [26]. Recent models such as Anaerobic Digestion Model No. 1 (ADM1) [27] were able to simulate all the biochemical and physico-chemical reactions that take place during the anaerobic digestion process to predict biogas formation, but they significantly increase their complexity and require the calibration of a large number of kinetic and stoichiometric parameters to be used with guarantees [28]. For this reason, simple BMP estimation methods based on the chemical composition of the substrate are still considered as a useful tool for co-substrate selection. One of the most widely used theoretical models is based on the biomass elemental composition and it is known as Buswell's stoichiometric model [29,30], whose simplicity makes it an appropriate tool for comparing many different substrates and discarding those whose results did not reach a minimum value. Other models based on biodegradable components such as lipids, proteins, or carbohydrates may improve the accuracy of BMP predictions, although they usually overlook the influence of some components such as lignin [31]. Authors such as Xu et al. [32] stated that the highest accuracy in BMP predictions was achieved with multiple linear regression models that experimentally correlated the organic content of the waste with the $CH_4$ yield. Moreover, some studies were also published that applied infrared spectroscopy for direct BMP predictions [33]. All these methods had a common purpose, which is reducing the excessive time required by the experiments traditionally used to estimate such potential.

In this work, a methodology for the multi-parametric comparison and selection of the most feasible industrial organic wastes to be used as co-substrates in anaerobic digestion has been tested. Parameters such as the production rates, seasonality, distance to the WWTP, pre-treatment requirements, and physico-chemical parameters such as the C/N ratio, theoretical BMP estimation, and biochemical composition (carbohydrate content), were included in a multi-parametric selection matrix, weighted and punctuated. All these parameters allowed for carrying out a fast and low-cost comparative analysis of different industrial organic wastes and provided a useful tool for the decision making and pre-selection of the most promising co-substrates for this purpose.

## 2. Materials and Methods

### *2.1. Pre-Selection of Co-Substrates*

The first step carried out for this study was an extensive search and location of potential co-substrates to be used for anaerobic digestion. For this purpose, more than 50 agro-food industries were contacted in order to obtain information about the organic wastes generated in their manufacturing processes and about other issues such as the distance to the biofactory, generation potential (ton/year or $m^3$/year), and seasonality of these wastes.

Based on the information provided by all these agro-food companies, a shorter list including only the most suitable candidates was drawn up following the criteria of the proximity to the WWTP, and the production of organic wastes and suitability, according to the results found in the literature. In total, 13 potential co-substrates were selected to be later analyzed and evaluated as anaerobic digestion co-substrates, trying to obtain a realistic and homogeneous representation of the variety of industrial organic wastes available in this area. These companies provided samples of each substrate for physico-chemical analyses.

### *2.2. Physico-Chemical Analyses*

The composition and characteristics of each substrate were required for theoretical BMP estimation [30]. Moreover, physico-chemical parameters such as pH may highly influence the process and they can even cause process inhibition and they need to be checked [8,34]. For this purpose, several laboratory analyses for the physico-chemical characterization of organic wastes were carried out in three different facilities. On the one hand, elemental analyses were carried out at the Scientific Instrumentation Center (CIC), while lipid, protein, and carbohydrate analyses were carried out at the Biomedical Research Center (CIBM). The remaining analyses were performed at the laboratory of the Environmental Technologies Area. All these installations belong to the University of Granada. The methodology used for each analysis is briefly described as follows.

#### 2.2.1. pH and Conductivity Measurements

A Crison pH 25 pH-meter previously calibrated at pH values of 4.0, 7.0, and 9.0 was used to determine the pH of liquid samples. For this analysis, the sensor was submerged inside the sample while stirring until a stable value was reached.

In the same way, the electrical conductivity (EC) of liquid samples was measured using a Crison model CM 35 conductivity meter including automatic temperature compensation, which was calibrated at the conductivity values of 147 µS/cm, 1413 µS/cm, and 12.88 ms/cm. The analyses were also carried out by immersing the sensor inside the sample while stirring until a stable value was reached.

#### 2.2.2. Moisture and Ash Content

Analyses carried out to determine moisture and ash content were carried out according to the Standard Methods for the Examination of Water and Wastewater [35]. For moisture measurements, a known weighted amount of each sample was placed in a previously dried porcelain crucible, weighed, and introduced into a stove at a temperature of 105 °C for 24 h in order to reach the total loss of the water contained in the sample. After that, the crucible containing the dry sample was weighed and put back into the oven for 15 min more. It was weighed again and this operation was repeated three times to verify that the weight remained constant. Moisture content was determined according to Equation (1), where $W_{\text{wet}}$ is the weight of the wet sample and $W_{\text{dry}}$ is the weight of the dry sample:

$$\text{Moisture content } (\%) = \frac{W_{\text{wet}} - W_{\text{dry}}}{W_{\text{wet}}} \times 100 \quad (1)$$

The same crucibles containing dry samples were placed in a muffle furnace at 550 °C for 15 min to obtain the weight of ashes after combustion. After this time, combustible organic matter was fully volatilized and only the inert compounds converted into ashes remained in the crucibles. They were taken out of the furnace and weighed to calculate the percentage of ash content, TS and VS according to Equations (2)–(4), where $W_{\text{ashes}}$ is the weight of ashes. Moreover, organic matter was estimated as the percentage of volatile solids in the sample.

$$\text{Ash content } (\%) = \frac{W_{\text{ashes}}}{W_{\text{dry}}} \times 100 \quad (2)$$

$$TS\ (\%) = \frac{W_{dry}}{W_{wet}} \times 100 \quad (3)$$

$$VS\ (\%\ respect\ to\ TS) = \frac{(W_{dry} - W_{ashes})}{W_{dry}} \times 100 \quad (4)$$

#### 2.2.3. Elemental Analysis

Samples were prepared prior to the analysis. They were dried and milled until reaching a 1 mm average particle size. Later, an elemental analyzer model, THERMO SCIENTIFIC Flash 2000, was used for the estimation of carbon (C), hydrogen (H), nitrogen (N) and sulfur (S) content. Dried samples (1 mg) were taken into an aluminum boat and loaded into the analyzer. Peak values were used to quantify the presence of C, H, and N [36], sulfur was analyzed using a trace S detector, and once the CHNS and ash contents were known, oxygen (O) was calculated using Equation (5) [12]. The C/N ratio was also calculated based on the C and N content of each sample.

$$\%\ O = 100 - \%\ C - \%\ H - \%\ N - \%\ S - \%\ Ashes \quad (5)$$

#### 2.2.4. Biochemical Composition

Prior to the biochemical analyses, samples were milled and separated in two different aliquots containing 20 and 80 g of each sample. Both aliquots were frozen until the analysis. The 20 g aliquot was thawed, weighed, and placed in the oven at 105 °C to obtain a dry sample. Likewise, its ash content was determined and organic matter content was also estimated. Simultaneously, the 80 g frozen aliquot was weighed and lyophilized. Later, it was milled again to improve the extraction of fatty matter. Samples were digested with sulfuric acid for at least 60 min and the Kjeldahl method was used to determine the percentage of N, which was later converted into proteins. Finally, the hydrolysis of the samples with hydrochloric acid was carried out for lipid estimation using the Soxhlet extraction method with petroleum ether and once the lipid analysis was completed, carbohydrate content was calculated using Equation (6) [18]:

$$\%\ CH = 100 - (\%\ Moisture + \%\ Ashes + \%\ Proteins + \%\ Lipids) \quad (6)$$

#### 2.2.5. Estimation of BMP

Theoretical BMP was estimated using the stoichiometric Buswell equation based on the elemental composition of the substrate [29,30]. CHNSO data obtained from the elemental analysis were used to estimate the maximum BMP based on the stoichiometry of the redox reaction illustrated in Equation (7) and assuming the total conversion of organic matter into $CO_2$ and $CH_4$. Cellular synthesis was not taken into account, since it was assumed that all donated electrons were only used for metabolic energy. According to these assumptions, $CH_4$ potential might be estimated using Equation (8).

$$C_aH_bO_cN_d + \left(\frac{4a - b - 2c + 3d}{4}\right)H_2O \rightarrow \left(\frac{4a + b - 2c - 3d}{8}\right)CH_4 + \left(\frac{4a - b + 2c + 3d}{8}\right)CO_2 + dNH_3 \quad (7)$$

$$Y_{CH_4 Buswell} = \frac{(4a + b - 2c - 3d) * 22.4}{(12a + b + 16c + 14d) * 8} \quad (8)$$

#### 2.2.6. Selection Matrix

Finally, a selection matrix was generated in order to evaluate and prioritize the co-substrate potential. These matrixial analyses have been widely applied for the selection of alternatives in a wide variety of fields such as waste management [37,38] and they are usually employed when several options need to be narrowed down to one choice and a large number of criteria may affect the decision [39]. In these cases, the matrix format may help to simplify and better understand all the parameters influencing the final selection.

To create a selection matrix, first of all, criteria to be considered must be selected and a weight needs to be assigned to each one depending on their specific importance. In this case, the significance of each parameter was indicated as a percentage. The C/N ratio and BMP (20% each) were supposed to be the most significant parameters to decide if these wastes were suitable for anaerobic co-digestion or not. Meanwhile, criteria such as seasonality or production capacity (5% each) were considered less relevant. Once the criteria were selected and weighed, the scores for each alternative were defined in such a way that a high score represents a favorable situation and a low score represents an unfavorable situation. In this study, three scores were selected: 1, 3, and 5. A score of 1 indicated that this waste was not recommended for this purpose, 3 indicated that it could be recommended for this purpose, and 5 indicated that this substrate was highly recommended according to this specific criterion. Finally, each score was multiplied by the corresponding weight and the total score for each alternative was calculated as the sum of the weighted scores for all the criteria. Those options that reached the highest final scores were supposed to be the best candidates for being selected as co-substrates [40].

## 3. Results

### *3.1. Pre-Selection of Co-Substrates*

Taking into account the review carried out prior to the pre-selection of co-substrates as well as criteria such as proximity or seasonality, different industries were chosen to analyze their organic wastes as potential co-substrates. These wastes are summarized in Table 1.

**Table 1.** Industrial wastes evaluated as co-substrates.

| Waste | Estimated Production Rate | Seasonality | Distance to the WWTP |
|---|---|---|---|
| Fried corn | 1000 kg/d | All year | 12 Km |
| Skinless corn | 2000 kg/d | All year | 12 Km |
| Sulfuric black water | 120 $m^3$/month | All year | 12 Km |
| Milk serum (whey) | Variable | All year | 7.5 Km |
| Pig slurry | 5000 L/d | All year | 40 Km |
| Olive pomace 1 | Variable | November–March | 52 Km |
| Wet fatty pomace | Variable | November–March | 23 Km |
| Olive pomace 2 | Variable | November–March | 23 Km |
| Egg shells | Unknown | All year | 7.6 Km |
| Chicken manure | Variable | All year | 7.6 Km |
| Asparagus stalks | 1000 kg/d | February–May | 41 Km |
| Grape pomace | 10,000 kg/y | February–April | 28 Km |
| Pineapple peels | 21,000 kg/d | All year | 22 Km |

Fried and skinless corn were produced in a food company, which produces sunflower seeds, crispy corn, and other types of nuts and snacks. Moreover, as the corn needs to be introduced into a solution of water and sulfuric acid to remove the skin before entering the line of production, there is also another liquid waste called "sulfuric black water", which is composed of a mixture of corn, water, and sulfuric acid. From a cheese factory, the milk serum or whey was also evaluated. Pig slurry and chicken manure were included in the list as potential co-substrates and egg shells were also considered. Related to the olive oil manufacturing process, several wastes may be considered for this purpose. In this study, two different olive pomace samples (known as "orujillo") were analyzed in order to compare their differences when they come from different installations, as one of them (olive pomace 1) was a fresh sample directly taken at the exit of the process and the other one (olive pomace 2) was stored outside the plant for several months. Olive pomace is formed by dry pulp, stones, and olive skin; it is solid and its oil content is low because it has been previously degreased. On the other hand, wet fatty pomace (known as "alpeorujo") was also analyzed. In this case, it is a pasty liquid composed of stones, pulp, oil, and olive skin and with a high content in water. From the wine manufacturing industry, dried grape pomace was also included in this study and finally, some agro-food industry wastes were also considered. Due to the seasonality characteristics in the production of fruits and vegetables, the available agro-food organic wastes were asparagus stalks and pineapple peels. Moreover, for comparative purposes, sewage sludge entering the anaerobic digester at the "Biofactory Granada-Sur" was also analyzed.

### 3.2. Physico-Chemical Analyses

Prior to the elemental and biochemical analyses, liquid samples were measured for pH and EC. All the samples were also analyzed for moisture, ashes, TS, and vs. determination. Data obtained with all these analyses are presented in Table 2.

**Table 2.** Physico-chemical analyses of industrial wastes.

| Waste | Humidity (%) | Ashes (%) | Organic Matter (%) | TS (%) | VS regarding TS (%) | pH | EC (mS/cm) |
|---|---|---|---|---|---|---|---|
| Fried corn | 2.15 ± 0.24 | 0.84 ± 0.06 | 97.01 ± 0.22 | 97.85 ± 0.24 | 99.14 ± 0.06 | - | - |
| Skinless corn | 48.55 ± 2.62 | 0.45 ± 0.10 | 51.00 ± 2.57 | 51.45 ± 2.62 | 99.13 ± 0.16 | - | - |
| Sulfuric black water | 77.31 ± 4.48 | 6.94 ± 0.13 | 15.76 ± 4.38 | 22.69 ± 4.48 | 69.43 ± 5.79 | 1.30 ± 0.96 | - |
| Milk serum (whey) | 93.62 ± 3.46 | 0.67 ± 0.05 | 5.71 ± 3.47 | 6.38 ± 3.46 | 89.50 ± 7.28 | 6.82 ± 0.07 | 8.16 ± 1.32 |
| Pig slurry | 91.00 ± 1.22 | 0.42 ± 0.05 | 8.58 ± 1.21 | 9.00 ± 1.22 | 95.31 ± 0.70 | 7.68 ± 0.16 | 18.16 ± 4.86 |
| Olive pomace 1 | 29.38 ± 2.26 | 1.33 ± 0.08 | 69.29 ± 2.31 | 70.62 ± 2.26 | 98.12 ± 0.16 | - | - |
| Wet fatty pomace | 67.92 ± 2.44 | 2.75 ± 0.16 | 29.33 ± 2.42 | 32.08 ± 2.44 | 91.43 ± 0.73 | 5.95 ± 0.13 | 20.4 ± 3.14 |
| Olive pomace 2 | 8.33 ± 0.59 | 1.33 ± 0.09 | 90.34 ± 0.68 | 91.67 ± 0.59 | 98.55 ± 0.11 | - | - |
| Egg shells | 6.70 ± 0.63 | 92.27 ± 0.83 | 1.03 ± 0.72 | 93.30 ± 0.63 | 1.11 ± 0.77 | - | - |
| Chicken manure | 73.10 ± 3.94 | 10.05 ± 0.46 | 16.84 ± 4.35 | 26.90 ± 3.94 | 62.63 ± 7.74 | - | - |
| Asparagus stalks | 92.78 ± 2.75 | 0.56 ± 0.08 | 6.66 ± 2.70 | 7.22 ± 2.75 | 92.24 ± 3.53 | - | - |
| Grape pomace | 15.16 ± 0.42 | 14.54 ± 1.61 | 70.30 ± 1.41 | 84.84 ± 0.42 | 82.86 ± 1.86 | - | - |
| Pineapple peels | 87.33 ± 4.07 | 0.57 ± 0.08 | 12.10 ± 4.14 | 12.67 ± 4.07 | 95.50 ± 2.74 | - | - |

pH highly influences biological processes. In particular, low pH values may inhibit the microbial activity of methanogenic bacteria, which are very sensitive to operational parameters [8]. According to authors such as Marchetti et al. (2020) [34], the preferred pH value for the biological anaerobic process to produce $CH_4$ is between 6.5 and 8.5. In this study, measured pH values ranged from 5.95 ± 0.13 to 7.68 ± 0.16 for samples such as wet fatty pomace, whey, and pig slurry. The only one that obtained pH values below this range was the sulfuric black water, which is highly acidic and reached a pH value as low as 1.3 ± 0.96 due to its high sulfuric acid content (7.22 ± 3.14% of S, shown in Table 3). For this reason, the biological process could be inhibited due to the acidification of the medium in case this waste is used as a co-substrate and it has been discarded for being used in anaerobic digestion. On the other hand, EC measurements showed average values for pig slurry and wet fatty pomace around 18 and 20 mS/cm, respectively, which are similar to the range usually obtained for the sewage sludge that enters an anaerobic digester in conventional activated sludge WWTPs [41]. The whey sample showed a lower value (around 8 mS/cm), indicating a weak presence of salts in this waste.

Regarding the TS and VS values, these results showed that fried and skinless corn reached the highest VS values (99.14 ± 0.06 and 99.13 ± 0.16% of TS, respectively), thus indicating that most of the solids were volatile and the ash content was very low. Other samples such as pig slurry or olive pomace also showed high VS percentages. On the contrary, egg shells showed the lowest one (1.11 ± 0.77%), indicating that almost 99% of the TS in egg shells were inert matter (Figure 1) that would not be consumed by bacteria in the anaerobic digesters [42]. As shown in Table 2, estimated organic matter contents were significantly higher for fried corn (97.01 ± 0.22%) and olive pomace 2 (90.34 ± 0.68%). Both olive pomace samples showed differences due to the water content of each sample, as olive pomace 1 was taken directly from the process and olive pomace 2 was stored and sun-dried for several months, so water was lost and higher solid contents were reached. On the other hand, egg shells (1.03 ± 0.72%), whey (5.71 ± 3.47%), and asparagus (6.66 ± 2.70%) showed the lowest values for organic matter. In the first case, this is due to the fact that this waste was composed mainly of inert mineral compounds (92.27 ± 0.83% ashes). On the contrary, whey and asparagus were mainly composed of water. According to these physico-chemical results, "sulfuric black water" was discarded due to the high sulfuric acid content and low pH showed by this sample [34]. Similarly, it can be assumed that egg shells should not be considered as a feasible co-substrate, as they were mainly composed of inert matter and they had a very low content in organic matter that could be used as a substrate for anaerobic bacteria [42].

**Table 3.** Elemental analyses of industrial wastes.

| Waste | N (%) | C (%) | H (%) | S (%) | O (%) | C/N Ratio |
|---|---|---|---|---|---|---|
| Fried corn | 1.38 ± 0.05 | 53.81 ± 4.37 | 11.89 ± 0.45 | 0.00 ± 0.00 | 32.08 ± 4.07 | 45.49 ± 4.51 |
| Skinless corn | 1.48 ± 0.13 | 45.04 ± 3.45 | 11.32 ± 0.22 | 0.00 ± 0.00 | 41.71 ± 3.48 | 35.50 ± 3.57 |
| Sulfuric black water | 1.95 ± 0.12 | 13.99 ± 0.47 | 6.79 ± 0.22 | 7.22 ± 3.14 | 63.11 ± 2.31 | 8.37 ± 0.72 |
| Milk serum (whey) | 1.64 ± 0.25 | 38.56 ± 3.56 | 9.33 ± 0.43 | 0.00 ± 0.00 | 49.80 ± 4.18 | 27.43 ± 2.86 |
| Pig slurry | 2.83 ± 0.37 | 42.38 ± 3.05 | 9.36 ± 0.38 | 0.24 ± 0.03 | 44.77 ± 3.28 | 17.47 ± 1.78 |
| Olive pomace 1 | 0.44 ± 0.03 | 49.57 ± 2.95 | 10.09 ± 1.95 | 0.00 ± 0.00 | 38.57 ± 1.14 | 131.44 ± 12.12 |
| Wet fatty pomace | 1.45 ± 0.07 | 54.04 ± 3.54 | 11.82 ± 0.28 | 0.00 ± 0.00 | 29.94 ± 3.83 | 43.48 ± 1.02 |
| Olive pomace 2 | 1.88 ± 0.21 | 48.18 ± 2.79 | 9.75 ± 1.05 | 0.00 ± 0.00 | 38.86 ± 2.54 | 29.90 ± 5.15 |
| Egg shells | 0.68 ± 0.02 | 1.36 ± 0.05 | 0.76 ± 0.03 | 0.00 ± 0.00 | 4.93 ± 0.74 | 2.34 ± 0.09 |
| Chicken manure | 6.91 ± 0.27 | 36.46 ± 2.94 | 6.99 ± 0.37 | 0.10 ± 0.03 | 39.49 ± 2.91 | 6.16 ± 0.70 |
| Asparagus stalks | 3.69 ± 0.27 | 44.68 ± 2.97 | 8.69 ± 0.27 | 0.15 ± 0.04 | 42.23 ± 3.19 | 14.13 ± 1.43 |
| Grape pomace | 4.18 ± 0.31 | 47.42 ± 3.68 | 7.50 ± 0.36 | 0.00 ± 0.00 | 26.36 ± 4.12 | 13.24 ± 1.21 |
| Pineapple peels | 0.90 ± 0.08 | 45.34 ± 3.74 | 9.18 ± 0.29 | 0.00 ± 0.00 | 44.01 ± 3.74 | 58.77 ± 7.27 |
| Sludge | 6.18 ± 0.33 | 42.64 ± 3.11 | 7.61 ± 0.34 | 0.29 ± 0.08 | 42.30 ± 3.07 | 8.05 ± 0.87 |

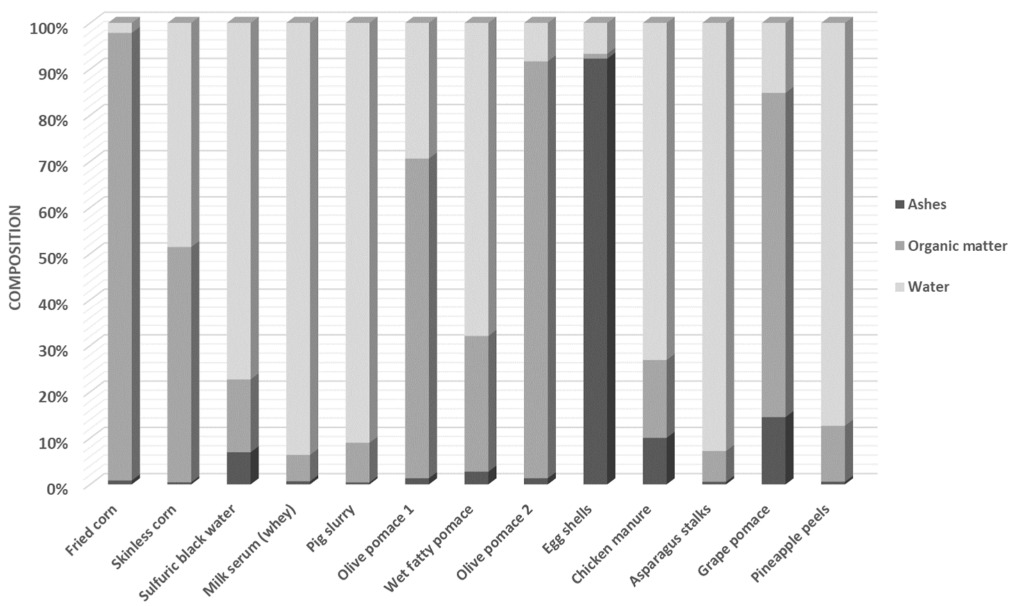


**Figure 1.** Water, ash, and volatile organic matter content for different organic wastes tested.

### 3.3. Elemental Analysis

Values obtained in this study to describe the CHNOS elemental composition of wastes are shown in Table 3. These results indicated that most of the evaluated substrates, including sewage sludge, had a high carbon content in a range from 36.46 ± 2.94 to 53.81 ± 4.37%. Only two of the wastes showed a carbon content below this range, which are egg shells and "sulfuric black water", already discarded. Similarly, it is worth mentioning that most of the wastes showed nitrogen contents lower than that of the sewage sludge and the only waste with a high nitrogen content similar to that obtained for the sewage sludge was chicken manure, with values of 6.91 ± 0.27% for chicken manure and 6.18 ± 0.33% for sewage sludge. Other wastes that showed high nitrogen contents were grape pomace (4.18 ± 0.31%), asparagus (3.69 ± 0.27%), and pig slurry (2.83 ± 0.37%). In relation to the hydrogen content, fairly homogeneous values were observed, while for sulfur content, only "black water", which contained sulfuric acid, had a significant sulfur percentage. In the literature, a great variability for the elemental composition of sewage sludge and many other anaerobic digestion feedstocks can be found as it highly depends on the specific characteristics and conditions of the samples [12,36,43].

Anaerobic digestion needs equilibrated concentrations of carbon (organic matter) and nitrogen (nutrients) to be carried out in an efficient way. For this reason, one of the most influencing parameters in the anaerobic digestion process is the C/N ratio [8,44]. Usually, sewage sludge from WWTPs is characterized by a low C/N ratio and high nitrogen content [45], so if co-substrates with low C/N ratios (high N contents) are used for co-digestion, the accumulation of compounds such as free ammonia may occur. These compounds are toxic for methanogenic bacteria and inhibit the process [46]. In such cases, to ensure the stability of the biological process, co-substrates may be used for increasing the C/N values of the mixture entering the anaerobic digester and reaching high carbon and low nitrogen concentrations inside the reactor [16,43,45]. However, co-substrates must be carefully selected because high C/N ratios may also be harmful for the process as it is possible to cause a decrease in pH and the interruption of the biological activity due to nutrient scarcity [47]. In general, the literature agrees that for a proper activity inside a digester, a C/N ratio in a range from 20 to 30 should be ensured [7].

In this study, according to Figure 2, samples with the highest values of the C/N ratio were olive pomace 1 (131.44 ± 12.12), fried corn (45.49 ± 4.51), wet fatty pomace (43.48 ± 1.02), and pineapple (58.77 ± 7.27), so it could be assumed that they could be good candidates as co-substrates. However, the first one could probably lead to unstable conditions due to a lack of nutrients [47]. On the other hand, the mixture of sewage sludge and co-substrates such as chicken manure (6.16 ± 0.70), grape pomace (13.24 ± 1.21), and asparagus (14.13 ± 1.43) could not reach optimum C/N values as all of them have low C/N ratios [7].

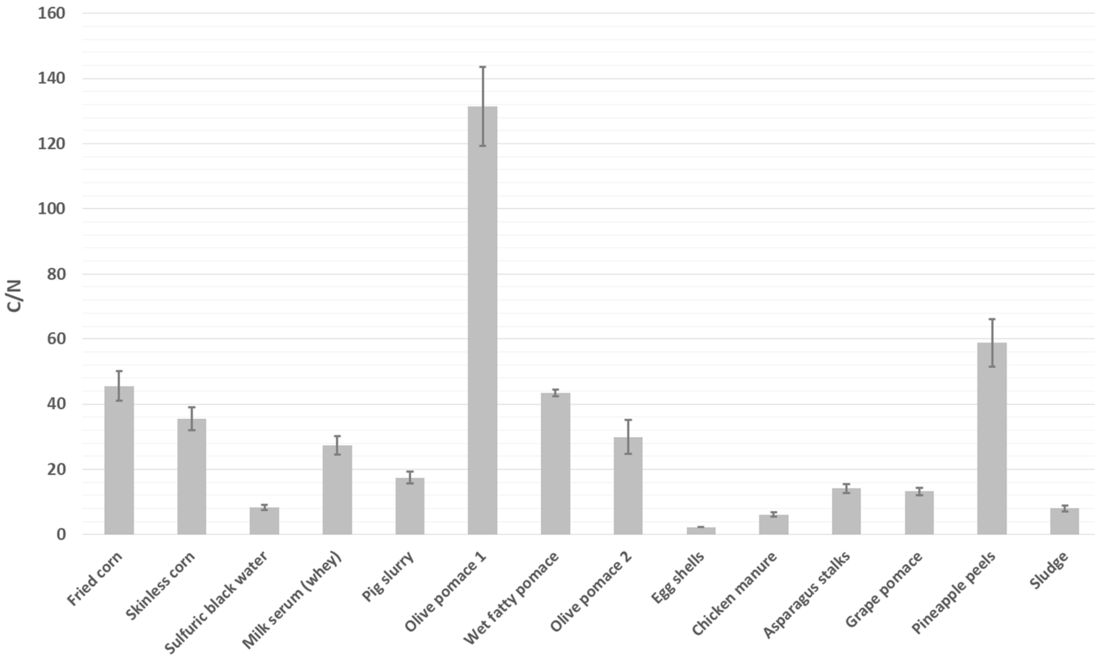


**Figure 2.** C/N ratio for different organic wastes tested.

### *3.4. Biochemical Analysis*

Table 4 shows the biochemical composition of co-substrates. "Black water" was not analyzed since its high sulfuric acid content and low pH ruled it out as a candidate to be used as a co-substrate. Moreover, samples of pig slurry, chicken manure, and sewage sludge could not be analyzed since these analyses were carried out in a laboratory for food characterization and, for sanitary reasons, it must maintain sanitized conditions that are incompatible with the introduction of this type of samples. The rest of the samples were biochemically analyzed.

**Table 4.** Biochemical analyses of industrial wastes.

| Waste | Lipids (%) | Proteins (%) | Carbohydrates (%) | Energetic Value, kcal/100 g |
|---|---|---|---|---|
| Fried corn | 32.45 ± 6.09 | 5.94 ± 0.39 | 58.62 ± 5.79 | 550 ± 41.2 |
| Skinless corn | 1.42 ± 0.14 | 4.16 ± 0.24 | 45.42 ± 4.48 | 211 ± 38.3 |
| Sulfuric black water | -- | -- | -- | -- |
| Milk serum (whey) | 0.01 ± 0.00 | 0.56 ± 0.09 | 5.14 ± 0.30 | 23 ± 4.7 |
| Pig slurry | -- | -- | -- | -- |
| Olive pomace 1 | 2.45 ± 0.21 | 1.93 ± 0.16 | 64.91 ± 7.06 | 289 ± 12.3 |
| Wet fatty pomace | 1.13 ± 0.20 | 2.27 ± 0.34 | 25.92 ± 0.77 | 123 ± 8.8 |
| Olive pomace 2 | -- | -- | -- | -- |
| Egg shells | 2.10 ± 0.16 | 5.84 ± 0.48 | 0.69 ± 0.06 | 45 ± 7.7 |
| Chicken manure | -- | -- | -- | -- |
| Asparagus stalks | 0.09 ± 0.02 | 1.32 ± 0.10 | 5.25 ± 0.20 | 27 ± 5.4 |
| Grape pomace | 0.72 ± 0.05 | 16.14 ± 0.20 | 53.43 ± 1.81 | 285 ± 10.2 |
| Pineapple peels | 0.12 ± 0.04 | 0.59 ± 0.04 | 11.38 ± 0.30 | 49 ± 3.2 |
| Sludge | -- | -- | -- | -- |

According to the literature [12,48], energy-rich co-substrates with high fats and easily degradable carbohydrate contents resulted in high biogas yields. In this case, the co-substrate that showed a higher lipid content was fried corn as it was fried using vegetable oil. The carbohydrate content of this sample was also high (58.62 ± 5.79%), although other food wastes, specifically pomace derived from the olive oil industry (64.91 ± 7.06%), grape pomace (53.43 ± 1.81%), and skinless corn (not fried in oil), also showed high carbohydrate contents. The average energetic values of the above-mentioned wastes ranged from 211 kcal/100 g for skinless corn to 550 kcal/100 g for fried corn. On the contrary, wastes such as whey (23 kcal/100 g), asparagus (27 kcal/100 g), egg shells (45 kcal/100 g), and pineapple peels (49 kcal/100 g) showed low energetic values, which predict low biogas production during the anaerobic co-digestion process.

Some disadvantages must also be considered, as the anaerobic digestion of feedstocks with an excessively high content of lipids and fats may be highly complex due to a pH decrease and accumulation of long chain fatty acids (LCFA) or volatile fatty acids (VFA) inside a digester. In these conditions, anaerobic microorganisms, mainly methanogenic bacteria, are very sensitive to these substances and biological activity may be inhibited by the presence of high amounts of fatty acids [34,47]. In addition, fats may cause operational problems due to characteristics such as the viscosity of these co-substrates. These operational problems must be taken into account when experimental installations at the lab, pilot, or full scale are designed for co-substrates such as fried corn and may be solved if these fatty co-substrates are mixed with easily degradable compounds such as carbohydrates [49,50].

### *3.5. BMP Estimation*

According to the theoretical BMP results (Figure 3), those wastes that presented the greatest BMP were 1. Wet fatty pomace (0.74 ± 0.05 $Nm^3CH_4/kg$); 2. Fried corn (0.72 ± 0.04 $Nm^3CH_4/kg$); and 3. Grape pomace (0.63 ± 0.06 $Nm^3CH_4/kg$). According to these data, binary mixtures of sewage sludge–wet fatty pomace or sewage sludge–fried corn, as well as ternary mixtures of sewage sludge–wet fatty pomace–fried corn or sewage sludge–wet fatty pomace–grape pomace, could be the more efficient mixtures of co-substrates to be experimentally tested. Moreover, samples of skinless corn, olive pomace (both fresh or stored), and pineapple peels may also be considered as proper candidates for this purpose. In the Discussion section, these results are compared with theoretical and experimental BMP values found in the literature for these substrates.

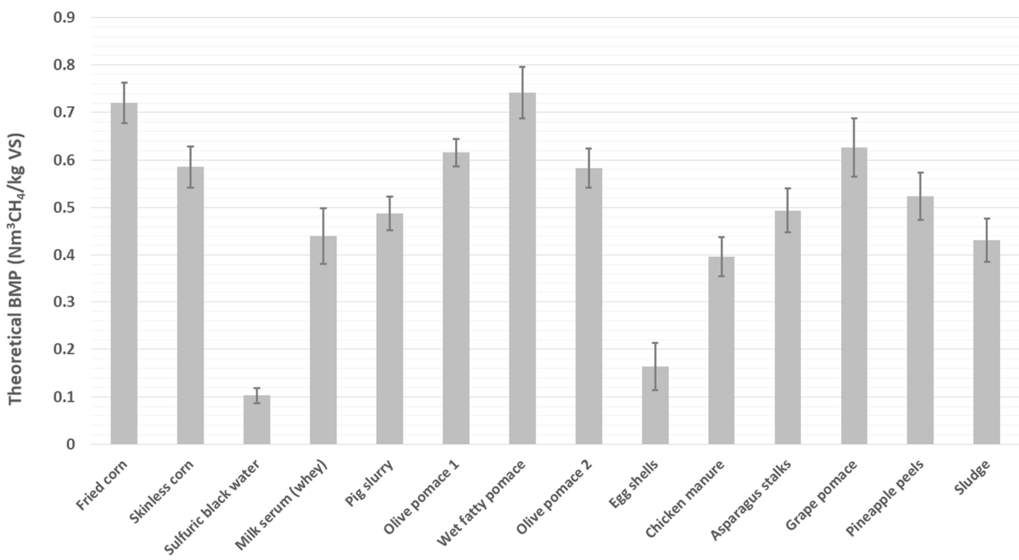


**Figure 3.** Theoretical BMP for different organic wastes tested.

### *3.6. Multi-Parametric Selection Matrix*

In order to facilitate the final selection of the most suitable organic wastes to be used as co-substrates in the anaerobic digesters of WWTPs, a comparative multi-parametric analysis was carried out on a matrix basis. This type of analyses may include different criteria considered as influential for anaerobic process performance and viability [40]. In this case, the most significant physico-chemical and biochemical characteristics of organic wastes were considered, as well as the amount of waste produced or the seasonal dependence of each one. In addition, the distance from the origin to the WWTP was included as it is an important parameter to evaluate transport costs. In the same way, if the waste requires any type of pre-treatment, this needs to be included as pre-treatments lead to an increase in energetic costs. Different scores (1, 3, or 5) were given to each co-substrate for every criterion, based on the ranges shown in Table 5. This table also shows the weight selected for each criterion.

**Table 5.** Set of parameters and ranges considered for the selection matrix.

| **Criterion** | **Score** | | | **Weight** |
|---|---|---|---|---|
| | **1** | **3** | **5** | |
| Production | Low (<1000 kg/d) | Medium (1000–10,000 kg/d) | High (>10,000 kg/d) | 5% |
| Seasonality | Seasonal generation of waste (<3 months per year) | Seasonal generation of waste (3–9 months per year) | Continuous annual generation (>9 months/per year) | 5% |
| Storage | Not possible | Short periods of time | Long periods of time | 10% |
| Distance to WWTP | >30 km | 15–30 km | <15 km | 10% |
| Pre-treatment | Strong milling and/or chopping | Soft milling and/or chopping | Not required | 10% |
| C/N ratio | <20 or >100 | 20–40 | 40–100 | 20% |
| BMP | <0.2 | 0.2–0.5 | >0.5 | 20% |
| Organic matter | <20% | 20–60% | >60% | 10% |
| Carbohydrates | <20% | 20–50% | >50% | 10% |

Figure 4 shows the scores applied to each co-substrate. Sulfuric black water was not included as it was previously discarded. Carbohydrate content for pig slurry and chicken manure could not be analyzed in the lab of the CIBM due to biological risks that could not be assumed in a food lab. For this reason, values for these two wastes were taken from the literature [18,24]. The code of colors is green: 5-point score, highly recommended; orange: 3-point score, slightly recommended; and red: 1-point score, not recommended.

| | Production | Seasonality | Storage | Distance | Pre-treatment | C/N ratio | BMP | Organic matter | Carbohydrates |
|---|---|---|---|---|---|---|---|---|---|
| Fried corn | 3 | 5 | 5 | 5 | 3 | 5 | 5 | 5 | 5 |
| Skinless corn | 3 | 5 | 5 | 5 | 3 | 3 | 5 | 3 | 3 |
| Milk serum (whey) | 3 | 5 | 1 | 3 | 5 | 3 | 3 | 1 | 1 |
| Pig slurry | 3 | 5 | 3 | 3 | 5 | 1 | 3 | 1 | 1 |
| Olive pomace 1 | 5 | 3 | 5 | 1 | 5 | 5 | 5 | 5 | 3 |
| Wet fatty pomace | 5 | 3 | 5 | 5 | 5 | 5 | 5 | 3 | 5 |
| Olive pomace 2 | 5 | 3 | 5 | 5 | 5 | 3 | 5 | 5 | 3 |
| Egg shells | 1 | 5 | 5 | 5 | 3 | 1 | 1 | 1 | 1 |
| Chicken manure | 1 | 5 | 3 | 5 | 5 | 1 | 3 | 1 | 1 |
| Asparagus stalks | 3 | 1 | 1 | 3 | 1 | 1 | 3 | 1 | 1 |
| Grape pomace | 1 | 3 | 5 | 3 | 5 | 1 | 5 | 5 | 5 |
| Pineapple peels | 5 | 5 | 1 | 3 | 1 | 5 | 5 | 1 | 1 |

**Figure 4.** Multi-parametric selection matrix scores for different organic wastes.

Following this, every score was multiplied by the weight given to this criterion. The sum of values corresponding to each co-substrate gives a total weighted score in a range from 0 to 5, which is indicative of the suitability of this waste as a co-substrate. These data are shown in Figure 5. The wastes that reached the highest scores were wet fatty pomace and fried corn (4.7), followed by olive pomace (4.3) and skinless corn (3.9).

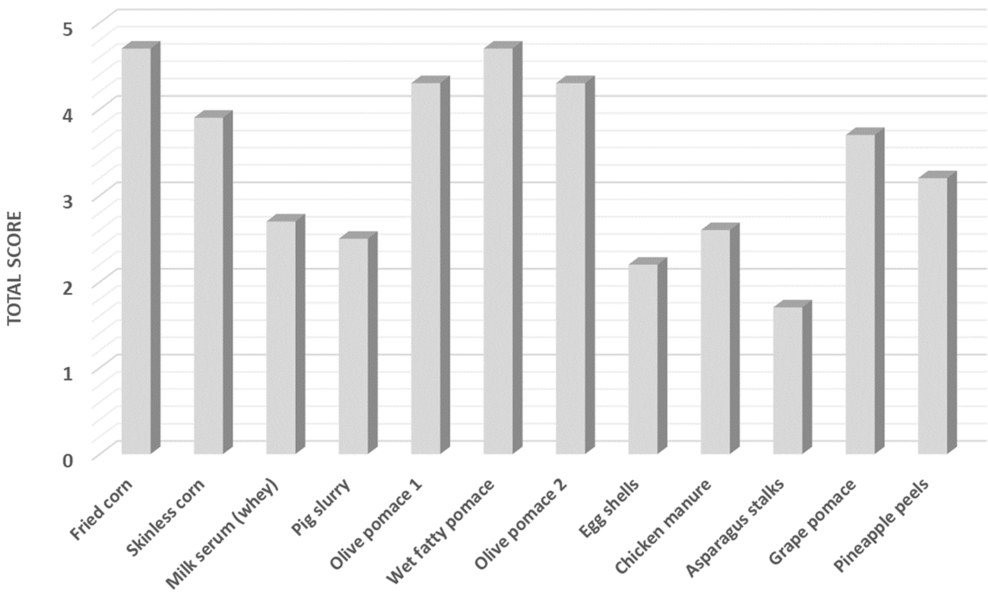


**Figure 5.** Total score for the multi-parametric comparison of different organic wastes.

A priority pyramid based on these data is shown in Figure 6. According to these results, two agri-food industries should be strongly considered as co-substrate suppliers: they are fried corn and olive oil industries. Not only wet fatty pomace and fried corn may be strongly recommended as potential co-substrates for anaerobic digestion but also other wastes from these industries may be considered for this purpose. On the other hand, co-substrates with high water contents such as whey or pig slurry resulted in lower scores in the selection matrix. Moreover, they might not be economically viable due to high transport costs and a low process efficiency improvement. In the same way, vegetables such as asparagus did not show optimal properties as a co-substrate.

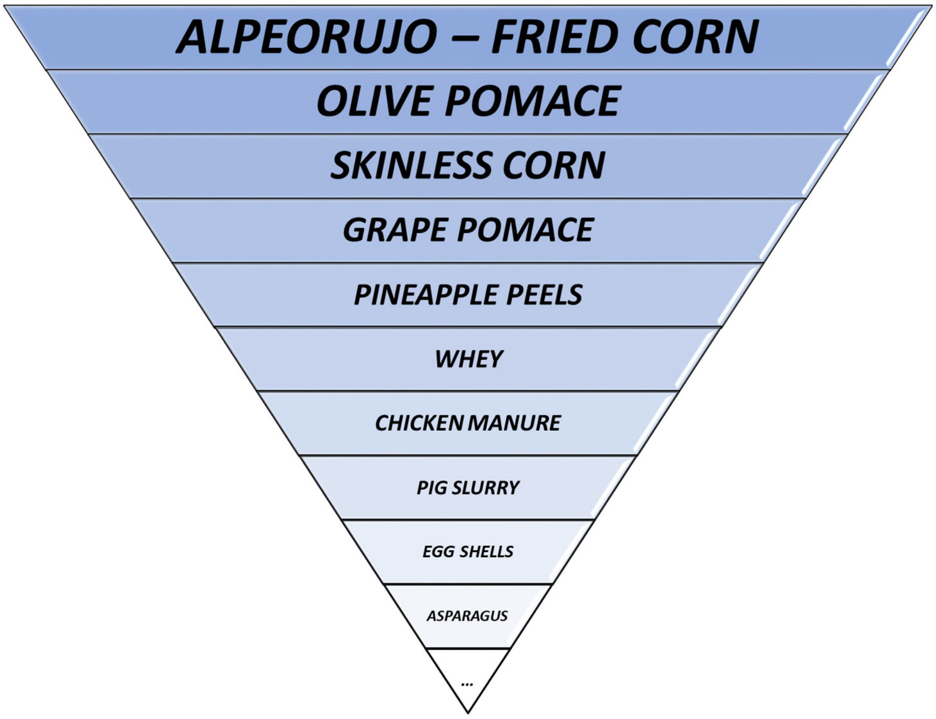


**Figure 6.** Priority pyramid for the pre-selection of different organic wastes tested.

## 4. Discussion

The sewage sludge entering anaerobic digesters in WWTPs is characterized by low C/N ratios and high nutrient contents, thus reaching low biogas production yields [36,43]. Wastes from agri-food industries are able to improve the nutrient balance, increase the C/N ratio, and optimize biogas production as they are usually easily biodegradable compounds with high carbon contents [7,8]. The same applies for other organic wastes such as herbaceous crops and pruning residues, which have a C/N ratio above the optimum range for anaerobic digestion and low quantities of nutrients [14,16]. The main drawback of these organic wastes is that they need high energy-demand pre-treatments to be used as co-substrates, which makes the valorization process more expensive. On the other hand, animal wastes such as slurry or manure provide a high buffer capacity but they usually have a high moisture content and a low C/N ratio, so their potential for biogas production is low and usually they may be used combined with sewage sludge and other co-substrates in ternary mixtures but it is not recommended to use them as sole substrates [8,24]. The variability of potential co-substrates and their changing characteristics leads operators to use many different binary or ternary mixtures of co-substrates, trying to compensate for the most important physico-chemical properties and composition of sewage sludge [44,50]. In order to decide which co-substrates may be used, both experimental and theoretical methods for BMP estimation may be used. One of these theoretical methods is the Buswell stoichiometric model, which has been widely used for different substrates [29,30].

Results described in the previous sections showed some of the most relevant characterization parameters of different organic industrial wastes. All of them provide useful information for selecting or discarding each organic waste as a co-substrate. According to these results, wastes with an optimum C/N ratio for anaerobic digestion were whey (27.43 ± 2.86) and olive pomace 2 (29.90 ± 5.15). These samples obtained estimated BMP values of 0.44 ± 0.06 and 0.58 ± 0.04 Nm$^3$ $CH_4$/kg, respectively. Whey was mainly composed of water (93.62 ± 3.46%) and it showed low values for carbohydrates (5.14 ± 0.30%) and organic matter (5.71 ± 3.47%). Moreover, one of the most important drawbacks of this co-substrate is that microbial activity may occur during storage and it may undergo fermentation or other biological processes that produce lactic acid and reduce pH, thus inhibiting the activity of methanogenic bacteria in an anaerobic digester, so it cannot be stored for long periods of time prior to entering the anaerobic digestion process [51].

On the other hand, olive pomace results are contradictory as two different samples from two olive oil industries were analyzed (olive pomace 1 and olive pomace 2), with C/N ratios of 131.44 ± 12.12 and 29.90 ± 5.15, respectively. C/N values found in the literature for this waste also showed significant differences [12,47]. These differences were probably due to the time that the waste was stored after being generated in the olive oil industry, as it was stored outside and biological composting processes may occur under sunny, humid, and aerated conditions, thus altering sample composition and reducing the C/N ratio [52]. In this case, olive pomace 1 was taken just after leaving the process and olive pomace 2 was taken after being stored for several months, showing humidity values of 29.38 ± 2.26 and 8.33 ± 0.59%, respectively. Carbohydrate content for the fresh sample of olive pomace was the highest one obtained in this study and reached 64.91 ± 7.06%. This value, together with the high volatile organic matter content (69.29 ± 2.31%), suggested a high amount of easily degradable organic matter [50]. The energetic value was also high compared to other evaluated wastes (289 ± 12.3 kcal/100 g) and these samples reached high scores in the selection matrix (4.3), so they are good candidates to be used as co-substrates.

Grape pomace is the sample that gave the highest value in proteins (16.14 ± 0.20%) and also showed a high carbohydrate content (53.43 ± 1.81%), energetic value (285 ± 10.2 kcal), and theoretical BMP (0.626 ± 0.06 Nm$^3$ $CH_4$/kg). On the contrary, its low C/N ratio (13.24 ± 1.21) and seasonality are important drawbacks. Nevertheless, it reached a total score in the selection matrix of 3.7 and it could also be used as a co-substrate. Authors such as El Achkar et al. (2016) [53] or Perra et al. (2022) [54] also evaluated anaerobic digestion as one of the most promising alternatives for this winery industry waste but they suggested that polyphenols and tannins could negatively affect the biological process.

Regarding the results obtained for fruits and vegetables such as asparagus or pineapple peels, it is remarkable that the asparagus waste is located at the bottom of the priority pyramid, even though it showed a high theoretical BMP (0.493 ± 0.05 Nm$^3$ $CH_4$/kg). Low lipid and carbohydrate contents (0.09 ± 0.02 and 5.25 ± 0.20%, respectively), as well as an excessively high water content (92.78 ± 2.75%) and a low C/N ratio (14.13 ± 1.43), strongly contributed to this position in the ranking of potential co-substrates. Authors such as Gunaseelan (1997) [55] also found similar results and significant differences among asparagus and other types of vegetables and they stated that its low biodegradability could be due to its woody structure. Moreover, this waste requires milling or chopping prior to entering a digester, making it difficult to use this waste as a co-substrate.

The situation of pineapple peels was different. Although they also had a high water content (87.33 ± 4.07%) and low organic matter estimation (12.10 ± 4.14%), they showed a C/N ratio of 58.77 ± 7.27 due to the C of sugars. Lipid and carbohydrate contents were low (0.12 ± 0.04% and 11.38 ± 0.30%, respectively) and the theoretical BMP reached a value as high as 0.523 ± 0.05 Nm$^3$ $CH_4$/kg. Seasonality was not a negative issue as this waste is produced during the whole year. All these values helped it to climb up the priority pyramid, although its main drawbacks are, on the one hand, that it requires pre-treatment prior to entering anaerobic digesters and, on the other hand, that it is not able to be stored for long periods of time, as fermentation processes may take place and high concentrations of acids such as acetic acid are generated. In fact, some authors such as Chalchisa and Dereje (2021) [56] analyzed this acetic acid production to evaluate the possibilities for the revalorization of pineapple peels to produce vinegar. These authors showed that after 72 h of storage, the pH of pineapple peels decreased to values as low as 3.5, thus producing acidification inside anaerobic digesters in case they are used as a co-substrate after storage.

Finally, some remarks must be discussed related to those wastes that are at the top of the priority pyramid: fried corn and wet fatty pomace, which reached the same total score of 4.7 in the selection matrix analysis. Regarding the results for theoretical BMP, the highest value was obtained for wet fatty pomace ($0.742 \pm 0.05$ $Nm^3$ $CH_4$/kg) and fried corn ($0.72 \pm 0.04$ $Nm^3$ $CH_4$/kg), suggesting that besides the differences between theoretical assumptions and real conditions, these wastes will be those that reach the highest $CH_4$ production during anaerobic co-digestion. These results are in agreement with authors such as Petrovic et al. (2022) [12], who stated that substrates rich in lipids and carbohydrates gave the best results for BMP in lab-scale experimental tests. The waste that showed the highest lipid content during this study was fried corn, which also presented the highest content in carbohydrates ($58.62 \pm 5.79$%) and the highest energetic value ($550 \pm 41.2$ kcal/100 g). In addition, it had a high content in organic matter, with an estimated percentage of $97.01 \pm 0.22$%. The ratio of C/N was subsequently high ($45.50 \pm 4.51$), so it could significantly improve the C/N ratio of sewage sludge and, therefore, ensure the stability of the process. Fried and skinless corn samples, despite being similar substrates, revealed large differences in lipid content ($1.42 \pm 0.14$% for the skinless corn) and the energetic value ($211 \pm 28.4$ kcal/100 g), as fried corn had a high content in fats and oils from the frying process. The theoretical BMP obtained for skinless corn was lower than that obtained for the fried corn ($0.58 \pm 0.04$ and $0.72 \pm 0.04$ $Nm^3$ $CH_4$/kg, respectively) and the C/N ratio was $35.5 \pm 3.57$. In this case, fried corn would be a better choice for anaerobic digestion, but it may also be valorized for other purposes such as animal feed while skinless corn is not actually reused for any other purpose so it could be revalorized as a co-substrate.

The other top waste was wet fatty pomace, which not only reached the highest BMP value but also a high C/N ratio ($43.48 \pm 1.02$), with enough carbon content ($54.04 \pm 3.54$%) and a low but sufficient content of nitrogen ($1.45 \pm 0.07$%) [7]. Therefore, the mixture of sewage sludge with wet fatty pomace as a co-substrate would also improve the C/N ratio of a mixture inside a digester in comparison with the mono-digestion of sewage sludge. It had a carbohydrate content of $25.92 \pm 0.77$% and an organic matter percentage of $29.33 \pm 2.42$%. In addition, its energetic value was $123 \pm 8.8$ kcal/100 g. These results are in agreement with authors such as Pellera and Gidarakos (2016) [57], who also obtained the highest theoretical BMP for this waste when comparing with other agro-industrial wastes. Other authors such as Petrovic et al. (2022) [18] and Awe et al. (2017) [49] evaluated the co-digestion performance at the lab scale of this type of lipid-rich substrates with other organic substrates. The first group of authors obtained the highest biogas yields for a mixture of sewage sludge from a WWTP with the lipid-richest substrate and the second group also demonstrated that the mono-digestion of these oily substrates showed promising results, in spite of some operational problems that these substrates may cause such as biomass flotation, physical fouling, or process instability due to LCFA production and an overload of VFA. For this reason, they suggested that co-digestion was the best alternative for olive oil industrial wastes. Besides this, it is generated in high quantities in the Mediterranean area and, according to the International Olive Council (IOC) [58], Spain is the largest producer of olive oil in the world—the provinces of Jaén, Córdoba, and Granada being the ones with the highest production [59]. In the province of Granada, huge amounts of wastes from the olive oil industry such as wet fatty pomace or olive pomace are produced every year, with an average estimated production of wet fatty pomace of $441 \pm 84$ kton/year in the last 5 years [60]. If not treated, this waste may cause environmental problems such as leaching and underground water pollution, odors, insects, GHG emissions, and some other problems [47] and for this reason, finding a solution for the reuse and valorization of this waste is an important issue for local producers. On the other hand, its main disadvantage is the seasonality of this industry, which is usually active from October to March. During storage, the C/N ratio may vary due to biological composting processes [52], and authors such as Hernández et al. (2018) [61] tested the evolution of physico-chemical parameters of wet fatty pomace during 6 months and, besides it suffering from changes in moisture and composition during this period, they did not find significant variations in pH; thus, it is still a proper waste for anaerobic co-digestion.

Once the most promising wastes to be used as co-substrates are selected, the next step should be to evaluate different binary or ternary mixtures in experimental tests using lab-scale systems prior to determining the most suitable mixture to be used in the full-scale anaerobic digester of a WWTP.

## 5. Conclusions

The procedure previously described allows both producers and WWTP operators to make decisions about the potential applicability of specific industrial wastes as co-substrates in anaerobic co-digestion processes. Parameters such as pH, composition, the C/N ratio, and BMP are indicative of the waste suitability for this purpose and they can be easily determined, thus reducing the time and cost demanded by the experimental lab-scale tests of all these samples. Moreover, using a selection matrix, it is possible to objectively compare different potential co-substrates, considering not only physico-chemical properties but also other important issues such as production, the distance, pre-treatment requirements, transport costs, and the seasonality, which are all relevant for the pre-selection of substrates. Both parameters and scores may be modified by the user to adapt the selection matrix to each specific situation. In this way, those wastes that obtain the highest scores may be pre-selected as the most optimal candidates to be used as co-substrates in order to improve the anaerobic digestion of sewage sludge. On the other hand, those wastes that obtain low scores may be discarded, as their use for this purpose is not technically or economically feasible.

According to the results obtained in this study, those wastes that showed a higher potential for being used in anaerobic co-digestion were 1. Fried corn from the snack food industry and 2. Wet fatty pomace from the olive oil industry. Both wastes showed high BMP values, high lipid and carbohydrate content, and high C/N ratios in a proper range to improve the low C/N ratio of sewage sludge in anaerobic co-digestion. Moreover, in the case of wet fatty pomace, its production rate in Granada is high; it is able to be stored and it does not require any pre-treatment prior to entering a digester. For these reasons, it is the best candidate for this purpose. Other wastes such as olive pomace (dry), skinless corn (not fried), and grape pomace from the winery industry may also be used as co-substrates.

On the contrary, pineapple peels, although showing high BMP and C/N values, were penalized with the pre-treatment requirements and the difficulties of being stored for long periods of time, as they generate acetic acid, which may affect the anaerobic digestion process. Other wastes such as pig slurry or chicken manure provided low C/N values and carbohydrate content, which make them suitable for co-digestion with any other co-substrate but not as sole co-substrates with sewage sludge.

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