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Slaughterhouse by-products treatment using anaerobic digestion

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ABSTRACT

The objective of the present study is to evaluate the use of animal by-products (ABP) as substrates for anaerobic digestion, aiming at methane production. Specifically, four ABP of Category 2 and 3, namely (i) stomach and rumen, (ii) stomach contents, (iii) breasts and reproductive organs and (iv) bladders and intestines with their contents, were selected. The methane potential of each ABP was initially determined, while the feasibility of anaerobic co-digestion of ABP with two agroindustrial waste, i.e. orange peels and olive leaves was also studied. To this purpose, Biochemical Methane Potential (BMP), as well as semi-continuous assays were respectively conducted. In the latter, the effect of the variation in the organic loading rate (OLR) on methane production was investigated. Results obtained from BMP assays showed that the samples containing breasts and reproductive organs, bladders and intestine, and stomach and rumen, had higher methane potentials of 815, 787 and 759 mLCH_{4,STP}/gVS, respectively. Moreover, according to the results of the semi-continuous assays, maximum methane yields between 253 and 727 mLCH₄/gVS_{fed} were obtained at an OLR of 0.8 gVS/L/d. The only case in which methanogenesis inhibition phenomena, due to increased ammonia concentrations, were observed, was the assay being fed with a mixture of breasts and reproductive organs and orange peels, at the highest OLR. This inhibition phenomenon was attributed to an inappropriate C/N ratio.

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1. Introduction

The high nutritional value of meat, i.e. high protein, bioavailable minerals and vitamins content, results in an increasing demand for livestock products and consequently, in increasing animal by-products generation originating in slaughterhouses. The characteristics and composition of slaughterhouse by-products vary considerably, depending on both the size of the slaughterhouse and the animal species that are slaughtered (Alvarez and Lidén, 2008). More specifically, the total amount of non-edible parts may vary from 20 to 45% of the live animal total mass (Reale et al., 2009). Most slaughterhouse by-products are contaminated with high numbers of microorganisms (Urlings et al., 1992), thus usually giving rise to significant environmental problems, due to both organic pollution and microbial load (Cuetos et al., 2008). The adoption of practices that will allow the sustainable management of such waste is a challenge for modern slaughterhouses (Franke-Whittle and Insam, 2013), with legislative restrictions and increased processing and disposal costs often being the cause of their inappropriate management (Arvanitoyannis and Ladas, 2008). The

European Union (EU) legal framework for slaughterhouse by-products management is determined by the European Regulation (EC) No. 1069/2009. According to this regulation, animal by-products (ABP) consist of full bodies or parts of animals, as well as products of animal origin that are not intended for human consumption, either because they are improper for consumption, as a result of their nature, or due to lack of commercial demand. More specifically, ABP are classified into three categories, namely Category 1, 2 and 3, which reflect the degree of risk, on the basis of risk assessments. The same Regulation signalizes that ABP disposal does not represent a sustainable choice, while it also recognizes that it would be in the interest of all citizens if a wide range of such by-products were used for various applications, provided that health risks are minimized. To this regard, the Regulation mentions the utilization of ABP of Category 2 and 3 for various applications, under certain conditions, which ensure public and animal health. Several different methods for ABP management have been proposed, including incineration, aerobic treatment, alkaline hydrolysis and anaerobic digestion (AD).

AD is a controlled biological degradation process, in which stabilization of organic substrates is achieved through the combined action of different microbial consortia, in the absence of oxygen. The main products of this process are biogas (a mixture of CH₄, CO₂ and other gases in traces) and digested sludge. The former

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Nomenclature

ABP	animal by-products	sCOD	soluble chemical oxygen demand [mg O ₂ /L]
ADF	acid detergent fiber [%]	SH	slaughterhouse by-products
ADL	acid detergent lignin [%]	STP	standard temperature and pressure
AW	agroindustrial waste	TA	total alkalinity [mg CaCO ₃ /L]
BMP	biochemical methane potential	TAN	total ammoniacal nitrogen [mg/L]
FAN	free ammonia nitrogen [mg/L]	TMP	theoretical methane potential [mL CH _{4,STP} /g VS]
HEM	hexane extractable matter	ThOD	theoretical oxygen demand [mg O ₂ /g VS]
HHV	higher heating value [kJ/kg]	TPH	total phenols [mg GAE/L]
HRT	hydraulic retention time [d]	TS	total solids [%]
LHV	lower heating value [kJ/kg]	VA	volatile acids [mg/L]
NDF	neutral detergent fiber [%]	VS	volatile solids [%]
OL	olive leaves		
OLR	organic loading rate [g VS/L/d]		
OW	orange waste		

can be used for energy applications and the latter to potentially improve soil fertility (Singh et al., 2010). AD constitutes one of the possible methods for treating ABP and at the same time, for producing energy in the form of methane (Hejnfelt and Angelidaki, 2009). ABP are characterized by high organic carbon contents and are rich in lipids and proteins, thus making them interesting feedstock for anaerobic digestion, due to the potential for elevated biogas production. On the other hand, the composition of ABP may sometimes delay the hydrolysis step and consequently, the rate of the overall process. In fact, fat hydrolysis results in the production of glycerol and long chain fatty acids, with the accumulation of the latter, having been associated with possible inhibition on the activity of methanogens (Angelidaki and Ahring, 1992). In addition, high fat contents may cause flotation and biomass wash-out phenomena. Furthermore, inhibitory effects due to high ammonia concentration levels, as a result of protein degradation may also be manifested. These inhibitory effects, are mainly related to the fact that free ammonia can passively diffuse into the cell, leading to phenomena such as proton imbalance and/or potassium deficiency (Pitk et al., 2013). Moreover, the formation of inhibitory substances and/or the lack of nutrients in the medium, often results in a drop in methane production. The aforementioned problems are often attributed to the non-appropriate C/N ratio. In fact, high C/N values could lead to lack of a nitrogen source for the methanogenic microorganisms, whereas low C/N values could lead to nitrogen accumulation as free ammonia, resulting in toxic effects on the growth of methanogens (Khalid et al., 2011). Apart from these issues, another important parameter for the anaerobic process is the amount of substrate being fed to the digester on a daily basis, in terms of volatile solids, i.e. the organic loading rate (OLR). This parameter represents the biological conversion capacity of an anaerobic digestion system. With a low OLR, the anaerobic digester is running inefficiently, whereas a high OLR, poses the risk of system failure due to overloading (Chen et al., 2014). In order to avoid the above mentioned problems, as well as the manifestation of nutrient imbalance phenomena, co-digestion is often applied. In such systems different biodegradable waste are mixed and treated together, in order to improve methane production (Serrano et al., 2013). In fact, this strategy encompasses multiple benefits, such as an improved nutrients balance, dilution of inhibitory substances, processing of different substrates in the same reactor, a better alkalinity balance, increased load of biodegradable organic matter, etc. (Chen et al., 2008; Khalid et al., 2011; Pagés-Díaz et al., 2015).

In Greece, a considerable portion of meat production results from sheep and goats slaughtering. In fact, in 2014, these products accounted for 20% of the total produced quantity (EUROSTAT,

2016). A significant number of such slaughtering activities takes place in small-scale facilities, in which incineration is the only available safe management method for ABP. However, at the same time, the high energy demand of this method results in high operational costs for these facilities. The objective of this study is to assess the viability of slaughterhouse by-products as a renewable energy source, through their use as substrates in anaerobic digestion systems. To this purpose, four different ABP from sheep and goats slaughtering, of Category 2 and 3, namely (i) stomach and rumen, (ii) stomach contents, (iii) breasts and reproductive organs and (iv) bladders and intestines with their contents, were investigated. These ABP were initially used to carry out biochemical methane potential (BMP) assays under mesophilic conditions, in order to determine their methane potential. Subsequently, the ABP samples with the highest methane potential values were selected for conducting mesophilic anaerobic digestion assays in semi-continuous operation mode, in which ABP were co-digested with two typical Mediterranean agroindustrial waste, namely orange peels and olive leaves. During the latter assays, the effect of the variation in OLR on methane production was also investigated. The specific types of sheep and goats slaughtering by-products used in the present study, have not been investigated before as substrates for anaerobic digestion under similar conditions, neither in mono-digestion, nor in co-digestion assays. In addition, to the authors' knowledge, combining these types of by-products with agroindustrial waste, such as orange peels and olive leaves, had not been previously attempted. Therefore, the present study, apart from presenting the above mentioned specific novelties, also contributes to the general topic of sustainable slaughterhouse by-products management. The contribution of this study is especially emphasized when considering the importance of researching the feasibility of management and treatment methods, which valorize regional materials in their place of production.

2. Materials and methods

2.1. Substrates and inoculum

The ABP were collected from the municipal slaughterhouse situated in Chania, Crete (Greece), in which mainly sheep and goats and in lesser amounts pigs and cattle are slaughtered. Four different ABP from sheep and goat slaughtering were investigated in this study, namely stomach and rumen (SH1), stomach contents (SH2), breasts and reproductive organs (SH3) and bladders and intestines with their contents (SH4).

A pretreatment procedure was followed for all ABP, during which the materials were left in an oven at 80 °C for five days, in

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