Chapter 5
Co-digestion of Animal Manure and Carcasses to Increase Biogas Generation

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Abstract Livestock productions are changing with scale production increasing and concentration in some geographical areas. As a consequence, the activity environmental sustainability is under concern especially for manure and carcass management, disposal, or treatment. The livestock production system has its own particularities for each rearing process, resulting in residues with different characteristics. News technologies for pre-treatment and treatment for these residues have been established. Anaerobic digestion is an alternative for treatment due to this process combines the waste stabilization producing renewable energy and biofertilizer. The different components of manure excreted by livestock could be influenced on the biodegradation and biogas production. Previous studies are corroborated in this chapter and highlighted the importance of process control and digestate application when the carcass and manure are digested. For the evaluation of the efficiency of treatment processes, reduce environmental risks, and sanitary aspects, the choice of biomarkers is imperative. This chapter presents an approach and review to legislation about the conditions and criteria for the use of manure and carcasses in biodigesters and subsequently biofertilizer.

Keywords Combined process · Process control · Environmental risks

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5.1 Livestock Production

The world food economy is increasing the demand by livestock products and consequently the global livestock production. Livestock is an important economic activity around the world, due to high-value products (Herrero et al. 2013). Livestock products (milk, beef, pork, and poultry meat) are supplied by four animal food systems (beef cattle, dairy cattle, pigs, and broilers) (Weindl et al. 2017). Swine, poultry, and cattle chains have representativeness importance in the global production with approximately 110, 71 and 61 million ton of meat in 2013, respectively, in addition, milk production is around 508 million ton (Gerber et al. 2013).

Livestock operations providing social benefits, mostly in the developing ones, however, are a major impact on the environmental quality through effluent production, large uses of water, and emission of greenhouse gases (GHGs) Sakadevan and Nguyen (2017). GHGs' emissions from cattle represent about 65% of total, while swine and poultry contribute with 9 and 8%, respectively. Table 5.1 is described the emission intensity of each chain.

Manure management practices that ensure the recovery and recycling of nutrients and energy contained in manure along chains can contribute to mitigation of GHG. In many parts of the world, where occurs the increasing of specialized livestock farms, without sufficient land for use these residues for crop production, increase the necessity of treatment alternatives (Petersen et al. 2007).

5.1.1 Cattle

USA is the major producer of bovine meat with 11.9 million ton in 2017, followed to Brazil (9.5), European Union (7.9), China (7.3), and India (4.3), and these countries represent 66% of the world production USDA (2018). For milk production, the leadership continues with USA (87 million ton), followed by India (50 million ton), China (36 million ton), Russia (31 million ton), Brazil (31 million ton), and Germany (29 million ton), and these countries represent approximately 50% of the world's total production (IFCN 2016).

Dairy systems (meat and milk production) are constantly changing due to the market demand and land occupation. The dairy systems are characterized by the following phases (Fig. 5.1) (FAO 2016a):

- Gestation: refers to the pregnancy period after mating, when the calf fetus develops prior to birth;

Table 5.1 Global production and emission intensity for livestock chains

Herd	Production (million tons in 2013)	Emission intensity (kgCO_2-eq kg^{-1} product)
Cattle	61.4	67.6
Chicken	71.6	5.4
Swine	110.2	6.1

Source Gerber et al. (2013)

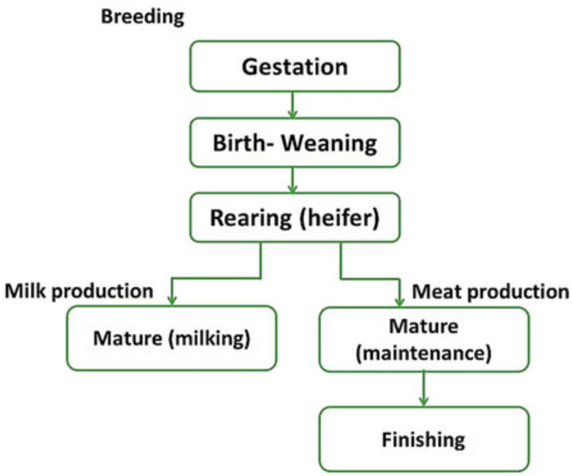

Fig. 5.1 Differences between systems of dairy milk and meat production. *Source* Adapted from FAO (2016a)

- Birth—Weaning: is the period after birth up until the calf is weaned from either its mother's milk or a milk replacement substitute. This stage may have different durations depending on the production system;
- Rearing (heifer): refers to the stage where the female animal (heifer) gains weight postweaning, reaching approximately 65–80% of the adult weight;the heifer may be mated or may be transferred to the beef system for fattening or immediate slaughter. This stage defined that animal is used to milk or meat production;
- Mature (milking): refers to the stage where adult postpartum cows are milked;
- Mature (maintenance): the former refers to the stage where animals are at their minimum mature body weight or may be used for other purposes;
- Finishing: the stage when the body weight is deliberately increased for slaughter.

Mortality on production units can depend on health status and management level, being considered a routine mortality until 5% of herd annually (McConnel et al. 2015; FAO 2016a).

5.1.2 Poultry

Statistics from poultry industry demonstrated that USA is the major producer of meat with 18.7 million ton in 2017, followed to Brazil (13.1 million ton) European Union (11.8 million ton), and China (11.6 million ton), representing 60% the world production (Embrapa 2018; USDA 2018).

The poultry sector is structurally diverse; there are differences in the scale and types of housing, feeding systems, and animal genetics. In a modern system production, the broilers are raised in large, open, or fully enclosed houses. The floors of the houses are covered with litter consisting of wood chips, rice hulks, or peanut shells. Barns are frequently equipped with automatic systems to deliver feed and

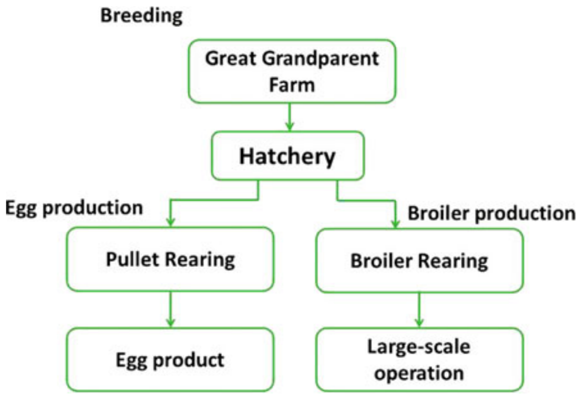

Fig. 5.2 Differences between systems of meat and egg in poultry chain. *Source* Adapted from FAO (2016b)

water (FAO 2016b). Figure 5.2 demonstrates the systems of meat and egg production. In this chain, the routine mortality was between 5 and 9% per year (CAST 2008).

5.1.3 Swine

China is the major producer of swine meat in the world with 53.4 million of ton in 2017, followed by European Union (23.6 million ton), USA (11.6 million ton), and Brazil (3.7 million ton), representing approximately 83% of total global production USDA (2018).

Swine production systems present a high variability ranging from very low (subsistence) to large-scale, in response to a factors socio-economic, markets and consumption. Globally, there is a wide variety of swine production systems, can be characterized by the following phases (FAO 2016c):

- Gestation: breeding females during gestation period;
- Breeding or farrowing: piglets until weighing 7–15 kg between 21 and 28 days of age;
- Nursery or Weaner: pigs, weighing 7–15 kg, reared to 25–35 kg at age 56–84 days;
- Growing to finishing: feeder pigs, weighing 25–35 kg, grown to market weight;

Swine production segregation is organized according to the countries characteristics (FAO 2016c). One example of segregation is demonstrated in Fig. 5.3, at where: farrow-to-feeder (gestation and breeding/farrowing), wean-to-finish (nursery/weaner and growing to finishing), Feeder-to-finishing (growing to finishing), farrow-to-wean (gestation, breeding/farrowing and nursery/weaner) fully integrated systems (gestation, breeding/farrowing, nursery/weaner, and growing to finishing).

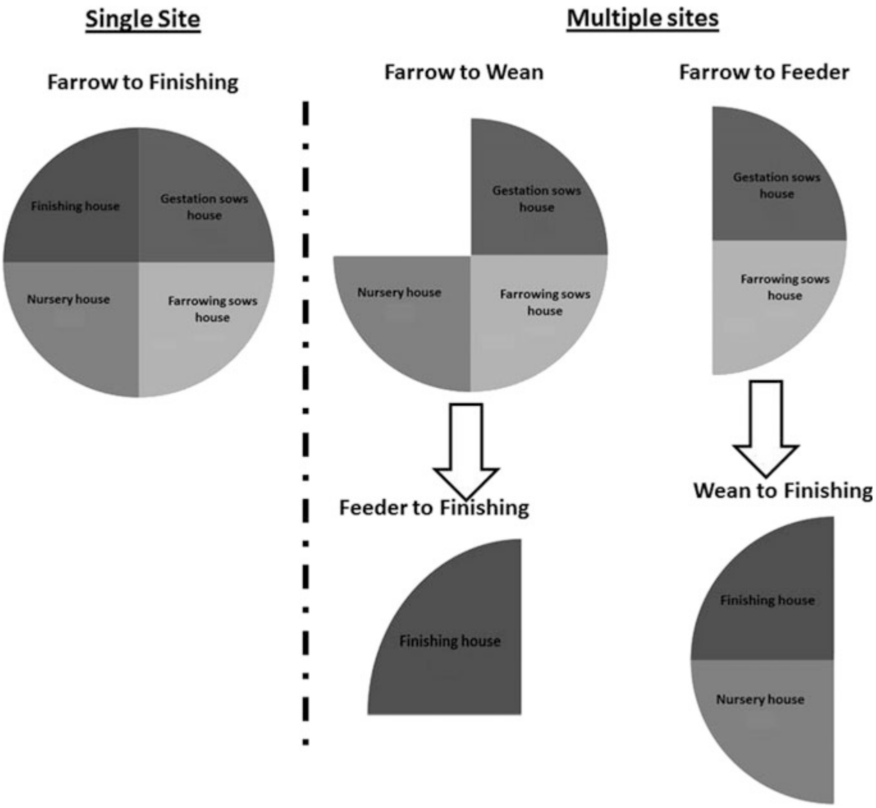

Fig. 5.3 Swine production systems and animal phases. *Source* Cestonaro do Amaral et al. (2016)

Mortality on production units can change depending on health status and management level, generally is between 3 and 9%. Likewise, the number of piglets stillbirths per litter can vary significantly, depending on litter size and sanitary status (FAO 2016c).

5.2 Management and Treatment of Animal Carcasses

There are different types of residues generated in livestock and poultry production that can be separated in: farm and industry levels. At farm level is generated mainly two residues, manure and dead animals, meanwhile, at industry level we have hatchery wastes, residues of meat, fat, feathers, blood, condemned carcasses, and others.

As the livestock industry grows, intensified for global food demands, the necessity of disposal alternatives that effectively manage carcasses and manure are increased. Simple and inexpensive methods such as burial are used for mortalities disposal on small farms, but they can lead to water and air pollution, and neither is practical for routine, large-scale use (Gooding and Meeker 2016). Responsible and safe animal carcass disposal is an important issue whole the world (Won et al. 2016), and need includes protection of environment, animal, and public health, due to animal carcass may contain pathogens, many of zoonotic importance (Berge et al. 2009; Zhong et al. 2017).

The methods used for carcass disposal include incineration, burial, rendering, composting, and anaerobic digestion.

- Incineration: is thermal-treatment method where animal carcasses or by-products are burnt at high temperatures (>850 °C), during this process is expected to destroy all infective pathogens (NABC 2004) (Fig. 5.4). The principal health concerns with the incineration of carcasses related to gaseous emissions and release of dioxins and furans from flue gas and fly ash, from incomplete combustion can settle in areas around carcass incinerators (Gwyther et al. 2011; Hseu and Chen 2017). Pollution control, it is necessary for the incineration installation, can reduce the risk of noxious emissions.
- From an environmental point, animal carcass incineration has a high energy demand that uses very high temperature (Gwyther et al. 2011). Furthermore, must be taken into consideration about biosecurity risks when transporting animal carcasses off-site (farms) in order to incineration facilities Stanford and Sexton (2006).
- Burial: To be applied this method should be considered, land topography, water table, and soil type of the available land will determine if burial is a valid, although has degradation need time and while production of noxious odors will continue during the degradation Stanford and Sexton (2006). In order to reduce the risk of transmission of bovine spongiform encephalopathy (BSE), the

Fig. 5.4 Animal carcass incinerator equipment. *Source* Lucas S. Cardoso

Commission Regulation (EC No. 1774/2009) prohibited in EU on-farm burning and burial for all fallen stock, irrespective of species susceptibility to prion diseases (except in specific situations, or in areas where access is practically impossible). In USA, burial/permitted landfilling is an accepted practice for animal carcass disposal in emergency management of animal mortalities (USDA 2012).

- Rendering: in this process entails crushing animal carcasses and by-products into smaller particles, heating and separate fat and protein, transforming in meat and bone meal and tallow (Kalbasi-Ashtari et al. 2008). However, after problems with BSE, the feeding of meat and bone meal is currently prohibited in developed countries, owing to rendering plants do not play as significant a role in the disposal of animal wastes, to avoid the dispersion of pathogens (Franke-Whittle and Insam 2013). Tallow from rendering can be used in among other applications as soaps, washing powders, as lipids in the chemical industry and cosmetics (Kalbasi-Ashtari et al. 2008). Rendering, as for incineration, has a high energy demand but if tallow is recovered for subsequent energy production then the net GHG emissions are likely to be low. The main environmental concerns associated with rendering are related to gas and odor emissions (Gwyther et al. 2011). Figure 5.5 demonstrates a flour of animal carcass.
- Composting: is a simple technique that can be undertaken on-farm, typically the process involves the layering of carcasses between strata of carbon-rich substrate such as straw, sawdust, or rice hulks with a final covering of carbon-rich substrate over the entire pile (NABC 2004) (Fig. 5.6); it is a relatively inexpensive technology and the final product can be transformed in fertilizer (Wang et al. 2016). Composting of dead animals requires the addition of a carbon source to ensure proper *C*/*N* ratios, odor and leachate control and equipment requirements differ the composting process (Kalbasi et al. 2005). The time for composting is a concern due to characteristics of the organic material and pathogens reduction (Glanville et al. 2016), because the organic matter instability, recontamination by pathogenic organisms and ammonia emission (Lasekan et al. 2013).

Fig. 5.5 Flour of animal carcass. *Source* Monalisa Pereira

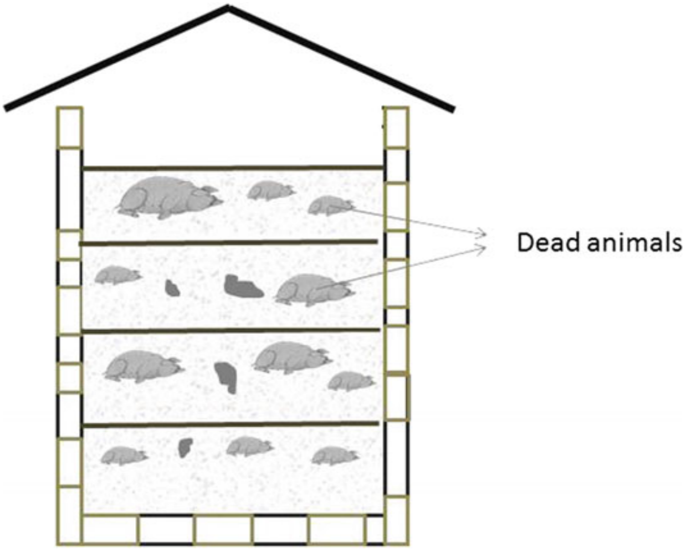

Fig. 5.6 Schematic of conventional composting system for dead animals

- Anaerobic digestion (AD): is a promising technology that combines a method for carcass disposal with renewable energy production, and other end products including liquid and solid fertilizers (digestate) (Zhang and Ji 2015). Anaerobic digestion of dead livestock is not permitted within current EU legislation without prior treatment of the carcass (sterilization) (EC No. 1069/2009). Figure 5.7 demonstrates an anaerobic co-digestion system that used animal carcass after pre-treatment.

Decision-makers should consider factors that compose each disposal technology (Table 5.2), including the principles of operation, costs, environmental considerations, advantages, and disadvantages of each technology (Baba et al. 2017).

Fig. 5.7 Anaerobic co-digestion of swine carcass and manure. *Source* Monalisa Pereira

Table 5.2 Advantages and disadvantages of methods for livestock carcasses disposal

Disposal methods	Advantages	Disadvantages
Incineration	Superior disease control high volume of waste reduction	Expensive, equipment, and fuel required; ash requires disposal; gas emissions;
Burial	Easy and inexpensive	Possible groundwater contamination tracking of sites required low degradation
Rendering	No generation of residues. Hide and tallow recycled	Logistic limitations; odor and gas emissions
Composting	Organic fertilizer production, easy technology, pathogens inactivation	Need control the time of composting due to odor emission and regrowth of pathogens
Anaerobic digestion	Renewable energy and organic fertilizer production	Necessity of pre-treatment of carcasses

5.3 Anaerobic Digestion Process Using Animal by-Products

5.3.1 Biochemical Methane Potential (BMP)

Due to a large production of livestock and poultry products, thousands of tons of organic by-products in the form of carcass, viscera, feet, head, bones, blood, and feathers are generated. Studies have suggested that residues that contain high concentrations of proteins and lipids (such as carcasses and animal products) are attractive substrates for biogas production (Rajagopal et al. 2014; Zhang and Ji 2015). The BMP test can be very helpful to estimate the biogas generation capacities of different substrates (Table 5.3).

The residues have a high methane potential, on the other hand, mono-digestion methods are susceptible to inhibition due to the accumulation of volatile fatty acids and/or unionized ammonia, resulting in toxicity for methanogenic archaea (Béline et al. 2017), reducing the methane production. One alternative to reduce this effect is simultaneous anaerobic co-digestion with others residues (e.g., manure), which may contribute to the dilution of inhibitory compounds originated during decomposition (Rajagopal et al. 2014). Using livestock manure with the substrate for co-digestion has shown to be an alternative treatment option.

Anaerobic co-digestion (AcoD) between manures and C-rich residues overcome these problems by maintaining a stable pH, within the methanogenic range, and reducing the ammonia concentration by dilution while enhancing methane production (Mata-Alvarez et al. 2011, 2014; Zhang et al. 2016). Most part of studies were conducted using livestock manure to establish different residues, with different types of reactors submitted at different operating parameters as temperature, organic loading rate (OLR), and hydraulic retention time (HRT) (Nasir et al. 2012).

Table 5.3 Biochemical methane potential of different residues of animal by-products

Animal	Material	BMP ($L_{N\ CH4}\ kg^{-1}_{VSadd}$)	Refs.
Bovine and swine	Digestive tract content	400	Luste et al. (2009)
	Meat and bone meal	390	Pitk et al. (2012)
	Fat	978	Pitk et al. (2012)
Swine	Meat tissue	976	Borowski and Kubacki (2015)
	Intestinal waste	826	Borowski and Kubacki (2015)
	Meat	575	Hejnfelt and Angelidaki (2009)
	Carcass	600	Tápparo et al. (2018)
	Solid slaughterhouse	580	Rodríguez-Abalde et al. (2011)
	Manure	406–1157 (biogas)	Cestonaro do Amaral et al. (2016)
Bovine	Soft offal	650	Ware and Power (2016)
	Paunch	228	Ware and Power (2016)
	Manure	204	Kafle and Chen (2016)
Poultry	Intestine residues	512	Yoon et al. (2014)
	Blood	250	Yoon et al. (2014)
	Solid slaughterhouse	460	Rodríguez-Abalde et al. (2011)
	Manure and feather	342	Yoon et al. (2014)
	Feather	210	Salminen and Rintala (2002)
	Meat	500	Salminen and Rintala (2002)
	Litter	259	Kafle and Chen (2016)

5.3.2 *Co-digestion of Animal Carcass*

As discussed above, co-digestion is an interesting alternative to reduce inhibitory effects of carcasses degradation under anaerobic conditions. Tápparo et al. (2018) described biochemical methane potential of swine carcass is around 1076 ± 48 $L_{Nbiogas}\ kg^{-1}_{VSadd}$ until five times more than swine manure. During co-digestion, the potential of methane yield is incremented until 6% per each $Kg_{carcass}$ added at m^3 of manure.

Massé et al. (2008) and Rajagopal et al. (2014) investigated psychrophilic AcoD of swine carcasses and swine manure in a sequence batch reactor (SBR) operated at 25 °C. Their results showed an increase in biogas production and no inhibition at rates of 20 and 40 $kg_{carcass}\ m^{-3}_{manure}$ (that represents up to eight times commercial swine farm mortality rates) (Massé et al. 2008). However, at carcass loading

Table 5.4 Operational conditions of animal carcasses anaerobic co-digestion

Material	Organic loading rate	Reactor type	Temperature (°C)	Refs.
Swine carcass and manure	3.2 g COD L^{-1} d^{-1}	SBR	20–25	Massé et al. (2008)
Swine carcass and manure	3.2 g COD L^{-1} d^{-1}	SBR	25	Rajagopal et al. (2014)
Swine carcass and sugar beet pulp	–	Batch scale	35	Kirby et al. (2018)
Beef carcass, algae, and manure	–	Batch scale	40	Pratt et al. (2013)
Swine carcass and vinasse	6.8 ± 0.4 $kg_{vs\ add}$· m^{-3} d^{-1}	Batch scale	35	Dai et al. (2015)

rates > 230 $kg_{carcass}$ m^{-3}_{manure} simulating emergency disease outbreak, the system was resulting in accumulation of volatile fatty acids and biogas inhibition (Rajagopal et al. 2014).

Several studies have tested different operational conditions for livestock and poultry carcass co-digestion with manure and others residues and are summarized in Table 5.4.

5.3.3 *Sanitary Aspects of Animal Carcass Anaerobic Digestion*

The AD process may be a sustainable method for on-farm carcasses management converting into biogas and organic fertilizers, with environmental and socioeconomic benefits (Hidalgo et al. 2018); however, when the reactors are operated in psycrophilic and mesophilic temperatures, the AD process itself is not sufficient to guarantee sanitary safety aspects (Viancelli et al. 2013; Fongaro et al. 2014; Tápparo et al. 2018).

Temperature is considered the main factor that influences the pathogens inactivation during anaerobic digestion (Franke-Whittle and Insam 2013) due to temperature increase can cause denaturation of proteins in the cell membrane, because it is more permeable and allowing diffusion of compounds into the cytoplasm Ziemba and Peccia (2011). Considering sanitary aspects, for animal by-products use in biogas plants, a pre-treatment is necessary to avoid pathogens dissemination in environment.

Regulation (EU) No 142/2011 (2011) determined that the process must be monitored and *E. coli* and Enterococcus counts must not exceed 1000 (3.0 log10) CFU/g, absence of *Salmonella* and *Clostridium perfringens*, reduction of infectivity of thermoresistant viruses and products must be subjected to a reduction in spore-forming bacteria, where they are identified as a relevant hazard (Commission Regulation (EU) No 142/2011 2011).

5.3.4 Compounds that May Cause Inhibition During AcoD Using Animal by-Products

The AcoD can be inhibited by some parameters that can compromise seriously the biogas-generating process. Some of these parameters that need attention during animal by-products anaerobic digestion are described below. Sometimes, these parameters present synergic effect, making difficult to determine the exact cause of decline in process performance (Moestedt et al. 2016).

5.3.4.1 Free Ammonia (FA)

The anaerobic digestion of livestock wastes and materials rich in proteins can increase total ammoniacal nitrogen (TAN) in digestate, that can cause inhibition of methanogenic microorganisms due to shifting of chemical equilibrium to FA resulting in low methane production (Yenigün and Demirel 2013; Kunz and Mukhtar 2016). The mechanism that explains FA inhibition says that it can freely permeate cell membranes resulting in the change in intracellular pH, increasing the cell maintenance energy requirement, and inhibition of specific enzyme reactions (Tao et al. 2017). Bayr et al. (2012) reported that one FA concentration of 635 mg L^{-1} promotion an inhibition of 50% on methane producing during the digestion of slaughterhouse by-products. High levels of FA also lead to an increase on volatile fatty acids concentration (VFA) during AD process, and this situation indicates an imbalance on microbiological community and facilitates foam generation (Kirchmayr et al. 2011; Resch et al. 2011). Previous studies about swine carcass and manure co-digestion in laboratory scale demonstrated an increase around 10 mg L^{-1} of NH_3–N for each $kg_{carcass}$ added per m^{-3}_{manure}, (Table 5.5).

5.3.4.2 Volatile Fatty Acids (VFA)

Residues that contain high lipids concentration are difficult to degrade, such as animal by-products, hydrolysis must be coupled with the growth of hydrolytic bacteria (Vavilin et al. 2008). Lipids can cause flotation and during hydrolysis, by extracellular lipases, VFA are accumulated (Palatsi et al. 2011). Anaerobic

Table 5.5 Ammonia and free ammonia during swine carcass and manure co-digestion

Swine manure and carcass ratio (kg m^{-3})	Digestate	
	NH_3–N (mg L^{-1})	Free ammonia (mg L^{-1})
0	2180	208
35	2220	269
68	2850	320
100	3000	345

Source Authors

digestion process is stable at a VFA-to-alkalinity ratio below 0.4. However, a severe instability can occur when the volatile fat acids/alkalinity (VFA/AL) ratio exceeds 0.6 (Mézes et al. 2011). Due to the possible accumulation of VFA, the co-digestion with substrate with higher alkalinity has a good option for animal by-products as described by (Rajagopal et al. 2014) and (Tápparo et al. 2018).

5.3.4.3 Foaming Generation

Substrate composition (i.e., lipids and proteins higher) has effects on the AD process viscosity, which may contribute to the increase of foaming (Kougias et al. 2014). Lipids have a tendency to form aggregates and foam causing problems (Cuetos et al. 2008). The presence of foaming in a biodigester can represent operational problems with as reactor overflow and fouling of mixing system (Kougias et al. 2015).

Several studies demonstrated a decrease of methane production because foaming problems and accumulation of fats occurred in the reactor during digestion or co-digestion of animal by-products (Cuetos et al. 2008; Pitk et al. 2013; Borowski and Kubacki 2015; Pagés-Díaz et al. 2015).

An ideal ratio between animal by-products and others residues are necessary for the process occurred without declining in biogas production. If one substrate was identified to cause foam, it was kept generally out of the process if possible or at least reduced in the substrate mix until foaming stopped (Lindorfer and Demmig 2016).

5.4 Legislation Applied for Animal by-Products Treatment and Disposal

European Union follows a regulation about the treatment and disposal of animal by-products (ABP). The European regulation (EC No. 1069/2009) defines different residues into categories based on the risk and material origin:

- Category 1: is a high-risk material, includes animals suspected of being infected by a transmissible spongiform encephalopathy (TSE), wild, pet, and zoo animals;
- Category 2: includes manure and digestive tract content, killed or fallen animals, including animals killed to disease control purposes, fetuses and oocytes, embryos, semen which are not destined for breeding purposes;
- Category 3: is low-risk ABP and comprises the following: carcasses and parts of animals slaughtered, blood, placenta, wool, feathers, hair, horns, and hoof that did not show infected disease communicable.

EU regulation describes the anaerobic digestion as an alternative treatment for Category 2 material (after pre-treatment, pressure sterilization), and Category 3 (some materials need used pasteurization like pre-treatment (EC No. 1069/2009).

In Brazil, national legislation describes that animals that died due to mandatory notification diseases, according to IN 50 (MAPA 2013), is a high-risk material (similar to material of Category 1 described in EU regulation), and have a specific treatment according to, respectively, state legislation. However, for routine mortalities (that could be classified as Category 2), alternative treatments could be applied. With the purpose of to evaluate and develop technological solutions of correct disposal of dead animals along poultry, swine, and bovine chains Embrapa (Brazilian Agricultural Research Corporation) and Ministry of Agriculture, Livestock and Supply (MAPA) developed the project "TEC-DAM, Technologies for disposal of dead animal". One of the objectives of this project is to evaluate the conditions for the use of dead animals in the biogas production chain (Nicoloso et al. 2017).

Due to the less development of anaerobic digesters in USA, no specific regulations about utilized animal by-products are found. However, Wang et al. (2018) suggested that USA could follow the European Union regulations for pathogens control during anaerobic digestion.

5.5 Final Remarks

Residues with high lipids and protein content like animal by-products, especially carcass, have an excellent potential of biogas. However, it is necessary a good process control due to a possibility of free ammonia and volatile fat acids accumulation and consequently inhibitions on methane production and foam generation. Besides that health aspects should be considered for digestion, as like European recommendation, the pre-treatment is imperative to ensure the pathogens inactivation.

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