Nitrous oxide emissions in soils fertilized with pig manure: soil processes and

strategies of control and mitigation

Emissões de óxido nitroso em solos adubados com dejetos suínos: processos no solo e estratégias de controle e mitigação

Emisiones de óxido nitroso en suelos fertilizados con estiércol porcino: procesos de suelo y

estrategias de control y mitigación

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Vilmar Muller Júnior

ORCID: https://orcid.org/0000-0002-2908-9411 Universidade Federal de Santa Catarina, Brazil E-mail: vilmar.muller@ifpr.edu.br Jucinei José Comin ORCID: https://orcid.org/0000-0002-6176-6208 Universidade Federal de Santa Catarina, Brazil E-mail: j.comin@ufsc.br Guilherme Wilbert Ferreira

ORCID: https://orcid.org/0000-0002-1768-2058 Universidade Federal de Santa Catarina, Brazil E-mail: guilhermewferreira@hotmail.com

Jorge Manuel Rodrigues Tavares

ORCID: https://orcid.org/0000-0002-4222-4251 Instituto Politécnico da Beja, Portugal E-mail: jorge.tavares@ipbeja.ptr **Rafael da Rosa Couto** ORCID: https://orcid.org/0000-0003-1071-4165 Instituto Federal Catarinense, Brazil E-mail: rrccouto@hotmail.com **Paulo Belli Filho** ORCID: https://orcid.org/0000-0003-4338-2830 Universidade Federal de Santa Catarina, Brazil E-mail: paulo.belli@ufsc.br

Abstract

Nitrous oxide (N₂O) is one of the main gases that contributes to the greenhouse effect. With a Global Warming Potential (GWP) 265 times greater than that of carbon dioxide (CO₂), over a 100-year horizon, N₂O also has the potential for the depreciation of the ozone layer. The activities related to agriculture and livestock are responsible for approximately 60% of the global anthropogenic emissions of this gas to the atmosphere. In Brazil, the sector corresponds to 37% of total emissions. The objectives of this review article were: (i) To verify which are the main processes involved in N₂O emissions in soils fertilized with swine manure; (ii) What are the direct emissions on these soils under different management systems, and; (iii) What are the possible strategies for controlling and mitigating N₂O emissions. Therefore, an exploratory and qualitative research of articles was carried out using the following keywords: óxido nitroso', 'nitrous oxide', 'N₂O', 'nitrogênio', 'nitrogen', 'suínos, 'pig, 'swine', 'dejetos', 'manure' and 'slurry'. Effects of pig diet, manure treatment systems, presence of heavy metals in the soil and moisture content of manure on N₂O emissions were verified. Therefore, we recommend integrated studies of the quantitative and qualitative impacts of the levels and sources of nitrogen in the animals' diets on N₂O emissions after the application of these wastes to the soil. We also recommend studies related to the effects of copper and zinc contents added to the soil via swine manure on enzymes that catalyze the biotic denitrification process in the soil.

Keywords: Environmental management; N₂O; Greenhouse gas emissions; Organic fertilization; Microbial community.

Resumo

O óxido nitro (N₂O) é um dos principais gases que contribuem para o efeito estufa. Com um Potencial de Aquecimento Global 265 vezes maior que o dióxido de carbono (CO₂), em um horizonte de 100 anos, também apresenta potencial de depreciação da camada de ozônio. As atividades agropecuárias são responsáveis por aproximadamente 60% das emissões antropogênicas globais deste gás. No Brasil, este setor corresponde por 37% do

total das emissões. Os objetivos deste artigo de revisão foram: (i) Verificar quais são os principais processos envolvidos nas emissões de N_2O em solos adubados com dejetos suínos; (ii) Quais são as emissões diretas nesses solos sob diferentes sistemas de manejo, e; (iii) Quais são as possíveis estratégias de controle e mitigação das emissões de N_2O . Para tanto, foi realizada uma pesquisa de caráter exploratório e qualitativo de artigos utilizando as seguintes palavras-chave: 'óxido nitroso', 'nitrous oxide', ' N_2O ', 'nitrogênio', 'nitrogen', 'suínos, 'pig, 'swine', 'dejetos', 'manure' e 'slurry'. Foram verificados efeitos da dieta dos suínos, dos sistemas de tratamento dos dejetos, da presença de metais pesados no solo e dos teores de umidade dos dejetos nas emissões de N_2O . Sendo assim, recomendamos estudos integrados dos impactos quantitativos e qualitativos dos teores e fontes de nitrogênio nas dietas dos animais sobre as emissões de N_2O após a aplicação desses dejetos ao solo. Também recomendamos estudos relacionados aos efeitos dos teores de cobre e zinco adicionados ao solo via dejetos de suínos sobre as enzimas que catalisam o processo de desnitrificação biótica no solo.

Palavras-chave: Gestão ambiental; N₂O; Emissões de gases de efeito estufa; Adubação orgânica; Comunidade microbiana.

Resumen

El óxido nitroso (N₂O) es uno de los principales gases que contribuyen al efecto invernadero. Con un potencial de calentamiento global 265 veces mayor que el dióxido de carbono (CO₂), en un horizonte de 100 años, también tiene un potencial de depreciación de la capa de ozono. Las actividades agrícolas son responsables de aproximadamente el 60% de las emisiones antropogénicas globales de este gas. En Brasil, este sector representa el 37% de las emisiones totales. Los objetivos de este artículo de revisión fueron: (i) Verificar cuáles son los principales procesos involucrados en las emisiones de N₂O en suelos fertilizados con estiércol porcino; (ii) Cuáles son las emisiones directas en estos suelos bajo diferentes sistemas de manejo, y; (iii) Cuáles son las posibles estrategias para controlar y mitigar las emisiones de N₂O. Por lo tanto, se realizó una investigación exploratoria y cualitativa de artículos utilizando las siguientes palabras clave: óxido nitroso', 'nitrous oxide', 'N₂O', 'nitrogênio', 'nitrogen', 'suínos, 'pig, 'swine', 'dejetos', 'manure' y 'slurry'. Se verificaron los efectos de la dieta del cerdo, los sistemas de tratamiento de estiércol, la presencia de metales pesados en el suelo y el contenido de humedad del estiércol sobre las emisiones de N₂O. Por lo tanto, recomendamos estudios integrados de los impactos cuantitativos y cualitativos de los niveles y fuentes de nitrógeno en la dieta de los animales sobre las emisiones de N₂O luego de la aplicación de estos desechos al suelo. También recomendamos estudios relacionados con los efectos del contenido de cobre y zinc agregado al suelo a través del estiércol de cerdo sobre las enzimas que catalizan el proceso de desnitrificación biótica en el suelo.

Palabras clave: Gestión ambiental; N₂O; Emisiones de gases de efecto invernadero; Fertilización orgânica; Comunidad microbiana.

1. Introduction

Nitrous oxide (N₂O) is one of the main gases that contribute to the greenhouse effect. With a global warming potential (GWP) 265 times that of carbon dioxide (CO₂), N₂O also has ozone depletion potential over a 100-year time horizon (IPCC, 2013). Human activities have significantly increased global N₂O emissions. The increase of N₂O concentration in atmosphere affect the environmental conditions, in addition to increasing the scale of phenomena associated to climate change. An increase of 20% in N₂O concentration in the atmosphere has been estimated since 1970, at a steady rate of 0.73 ± 0.03 ppmv/yr⁻¹ (IPCC, 2013). N₂O represents 16% (7.84 Gt CO₂-eq/yr⁻¹) of the total greenhouse gas (GHG) emissions from 2000 to 2010 (IPCC, 2013). On a global scale, uncultivated soils account for most N₂O emissions (6.6 Tg N/yr⁻¹), followed by agricultural soils (4.7 Tg N/yr⁻¹). Agricultural soils make up 27% of total N₂O emissions (Syakila and Kroeze, 2011) and 60% of human-caused emissions (Aguilera et al., 2013). Among the largest gas-emitting countries and blocks of countries are China (18.6%), the United States (9.1%), the European Union (8.4%; 28 countries), India (7.6%) and Brazil (6.8%), which together represent approximately 50.6% of global human emissions. In Brazil, agriculture is one of the main sectors responsible for emissions and accounts for approximately 37% of the national N₂O emissions (BRASIL, 2014).

Manure from animal production represents 30 to 50% of total emissions and is among the sources that contribute to N_2O emission in the agricultural sector. Table 1 shows the GHG emissions (Tg CO₂-eq/yr⁻¹) from the storage and treatment of animal manure, as well as its application to the soil.

Emission source	Tg CO ₂ -eq/yr ⁻¹			
Emission source	World	Brazil		
Manure storage and treatment				
Pig manure	91.55	3.54		
Cattle manure	161.49	4.42		
Poultry manure	20.99	0.92		
Others ¹	77.78	0.63		
Total	351.81	9.51		
Manure aplication in soil				
Pig manure	34.27	3.19		
Cattle manure	81.75	4.80		
Poultry manure	28.76	1.50		
Others ¹	47.04	1.45		
Total	191.82	10.94		

Table 1. Greenhouse gas emissions from animal manure.

¹ goat, sheep, buffalo, donkey, etc. $Tg = 10^{12}$ g. Fonte: FAO (2016).

Globally, the application of swine manure to the soil is responsible for approximately 18% of GHG emissions to the atmosphere from the use of animal manure as fertilizers. In Brazil, this practice is responsible for 29% of emissions. This high emission value can be explained by the large volume of pig manure produced daily in Brazil. In general, a pig in growing-finishing phase (25 to 120 kg body weight) produces approximately 4.5 L of manure per day (FATMA, 2014; Tavares et al., 2014). With an approximate herd of 35 million head of growing-finishing pigs, an estimated 158 million liters of manure are produced per day in Brazil. If the breeding sows are added to this herd, there is a total of 39.9 million head, reaching an approximate 270 million liters of manure per day.

Considering its fertilizer potential, the use of pig manure in agriculture is an alternative to the improper disposal of these materials in soil and water. It lessens environmental impacts, in addition to reducing the use of mineral fertilizers and the costs of purchase. The use of manure contributes to nutrient cycling of pig farming and increased crop yields. Pig manure is composed of nitrogen (N), predominantly as ammonium (N-NH₄⁺), in addition to phosphorus (P), potassium (K), copper (Cu⁺²), zinc (Zn⁺²) (Tiecher et al., 2013) and organic carbon in soluble and particulate forms. Table 2 shows the average physical and chemical characterization of the liquid pig manure in nursery (32 production cycles, n = 54,715 piglets) and grower-finisher (33 production cycles, n = 13,276 pigs) phases (Tavares, 2016).

Manure	Nursery			Grower-Finisher			
	Average	Max.	Min.	Average	Max.	Min.	
TS (g L ⁻¹)	40.9±20.2	74.5	14.9	58.2±14.9	91.8	33.7	
FS (g L ⁻¹)	9.8±3.9	16.9	5.5	14.6±3.3	22.1	9.5	
COD (g L ⁻¹)	-	-	-	74.8±14.9	111.3	47.7	
TOC (g L ⁻¹)	17.1±9.2	30.8	4.5	-	-	-	
TN (g L ⁻¹)	3.3±1.4	6.2	1.3	5.3±1.1	7.2	3.6	
$N-NH_4^+$ (g L ⁻¹)	1.6±0.5	2.8	1	3.1±0.6	4.5	2.5	
TP (g L ⁻¹)	0.7±0.3	1.3	0.3	1.2±0.3	1.8	0.7	
K (g L ⁻¹)	$1.9{\pm}0.8$	3.9	1.1	2.2±0.5	3.6	1.5	
Cu (mg L ⁻¹)	37±19	82	9	31±12	62	11	
Zn (mg L ⁻¹)	303±147	540	43	53±15	90	27	
рН	6.5±0.1	6.7	6.4	7.5±0.3	8.2	6.9	

Table 2. Average physical and chemical characterization of liquid pig manure (dry weight basis).

TS: total solids; FS: fixed solids; COD: chemical oxygen demand; TOC: total organic carbon; TN: total nitrogen; N-NH4⁺: ammonium nitrogen; TP: total phosphorus; K: potassium; Cu: copper; Zn: zinc; pH: hydrogen potential. Source: Adapted from Tavares (2016).

As shown in Table 2, pig manure has wide variations in the concentration of solids, carbon, pH and nutrients. This variability in manure makes it difficult to adopt a single treatment or use strategy as fertilizer, which increases the associated environmental risks. Due to its physical characteristics (predominantly in the liquid phase), manure is disposed in soil close to the pig production facilities. As it consists of large amounts of water, it has not been financially attractive to transport manure over long distances. Thus, large amounts of fertilizer are applied to soils of the same areas (Aita et al., 2015) and often above levels foreseen in current legislation. Continued manure application may have positive or negative effects in soil microbial communities. Data from the literature show that areas fertilized with liquid pig manure have higher N₂O emissions compared to areas using synthetic fertilizers (Decock, 2014). However, these effects are associated to several factors, such as the characteristics of pre-existing microbial communities in soil as well as the management and the physical, chemical and microbiological properties of manure.

Thus, the objectives of this review were: (i) Verify which are the main processes involved in N_2O emissions in soils fertilized with pig manure; (ii) What are the direct emissions on these soils under different management systems, and; (iii) What are the possible N_2O emissions control and mitigation strategies.

2. Methodology

Regarding the research characteristic, the study has an exploratory and qualitative character (Pereira, 2018). Data recovery was performed in articles published in scientific journals without defining a specific period, using the search tools of the Portal de Periódicos Capes (CAPES / MEC, Brasília, DF, Brazil). Keywords used in the searches included 'óxido nitroso', 'nitrous oxide', 'N₂O', 'nitrogênio', 'nitrogen', 'suínos, 'pig, 'swine', 'dejetos', 'manure' and 'slurry'. The publications obtained were imported, and duplicate occurrences were checked, using an EXCEL spreadsheet. To minimize bias, some criteria were applied: (i) emissions measured during the development cycle of agricultural crops in the field, with the development of crops in the soil; (ii) emissions reported in experiments that include treatments with and without nitrogen

fertilizer application; (iii) N source of chemical or organic synthesis, and; (iv) areas with and without irrigation. In order to verify the effect of mitigation strategies on N_2O emissions, studies using slow-release fertilizers, enzyme inhibitors or coated with polymers were considered. The majority of articles published since the 2000s were recovered, in a total of 85 publications. They were pre-selected for reading and inclusion in this literature review was one that met the selection criteria used. To verify the direct emissions in soils fertilized with swine manure, studies were used in which corn (grains or silage), black oats or wheat were grown, resulting in a total of eight publications.

3. Main Soil Processes Involved in N₂O Emissions

Approximately 70% of the global soil N₂O emissions are a result of microbial nitrification and denitrification processes (Syakila and Kroeze, 2011). The metabolic pathways responsible for emissions are extensive, often producing simultaneous N₂O emission and consumption (Butterbach-Bahl et al., 2013). The addition of chemical or organic fertilizers induces changes in microbial communities (Suleiman et al., 2016), which consequently affect N₂O emissions. These microorganisms are associated with rhizosphere, soil particles and decomposing organic material, and respond quickly to changes in soil management and fertilization. Thus, they are good indicators of the changes in soil properties. Microbial communities responsible for N₂O emissions are vast and contain strictly nitrifying organisms and facultative anaerobes. Therefore, it is important to define fertilization strategies according to use and crop. It is important to emphasize that communities modulate their activity according to nutrient, carbon, and oxygen availability, in addition to soil moisture (Meng, Ding, and Cai, 2005).

When applied in both organic forms (e.g., hippuric acid - $C_9H_9NO_3$) and free forms (e.g., ammonia - NH_3), nitrogen compounds in pig manure are used as energy sources by soil organisms, especially by bacteria and archaea. Electron consumption for reducing one atom of O_2 to water occurs in the nitrification process (Heil, Vereecken, and Brüggemann, 2016). This produces some intermediates such as hydroxylamine (NH_2OH) and nitrite (NO_2^-), generating nitrate (NO_3^-) as the final product (Figure 1).



Figure 1. Schematic representation of the nitrification process.

Source: Authors.

In the first stage of the nitrification process the formation energy ($\Delta G^{0,2}$) is positive, that is, the reaction does not occur spontaneously, requiring external energy for it to occur. This energy can be provided by both biotic factors (e.g. microbial activity) and abiotics (e.g. the presence of electrons in the soil solution). Conversion of hydroxylamine (NH₂OH) to nitrite (NO₂⁻) occurs quickly, since the $\Delta G^{0,2}$ value of the reaction is negative (spontaneous reaction).

After the oxidation of NH_3 to NO_3^- , it can then be used in other pathways (e.g., uptake by plant and microorganisms). When nitrate is present in the soil solution, it can be used as an electron acceptor by the same autotrophic ammonia-oxidizing bacteria. It is then reduced to nitric oxide (NO) and N₂O by nitrification-denitrification processes (Figure 2).



Figure 2. Schematic representation of the denitrification process.

Source: Authors.

When nitrate is present in the soil solution, it can be used as an electron acceptor by the same autotrophic oxidizing bacteria as ammonia. It is then reduced to nitric oxide (NO) and N₂O by nitrification-denitrification processes (Figure 2). The reduction of nitrate to N₂O requires electrons and H⁺ ions present in the solution, or supplied by soil microorganisms. A description of the main pathways involved in soil N₂O emission and consumption is presented below.

3.1 Biotic denitrification

Biotic denitrification is a bacterial and fungal respiratory process that occurs under anaerobic conditions, and it is the main source of N_2O emission. Microorganisms use nitrate, nitrite and soluble nitrogen gases as electron acceptors instead of oxygen (Phillipot et al., 2007). The conversion of soluble nitrogen oxides to N_2O is catalyzed by enzymes, which are inhibited in the presence of oxygen. Because it is a heterotrophic process, denitrification requires labile sources of carbon as substrate (Butterbach-Bahl et al., 2013).

3.2 Abiotic denitrification

Abiotic denitrification occurs predominantly under conditions of high nitrate concentration and in soils with pH <5.0. The chemical decomposition of hydroxylamine and nitrite occurs during nitrification to form NO, N₂O and eventually N₂. The decomposition of ammonium nitrate may also occur in the presence of light, moisture and reactive surfaces. The importance of this pathway as a source of N₂O emission is significantly lower compared to the biotic process (Butterbach-Bahl et al., 2013).

In addition to these processes, nitrite may react with several metals in soil, producing gases such as N_2O through reactions with Fe⁺³ and Cu⁺² ions (Heil, Vereecken, and Brüggemann, 2016).

3.3 Dissimilatory nitrate reduction

Nitrate ammonification or dissimilatory reduction of nitrate to nitrite and ammonium (DNRA) is a process conducted by both facultative and obligate anaerobes under strictly anaerobic conditions. This process occurs predominantly in hydromorphic soils and its magnitude varies according to the availability and relationship between C and N (Butterbach-Bahl et al., 2013).

3.4 Ammonia oxidation

The oxidation of ammonia to hydroxylamine and then to nitrite is an autotrophic process, which occurs mainly by the action of bacteria of the genus *Nitrosomonas*, *Nitrosolobus* and *Nitrospira* (associated with acidic soils). In addition to the formation of nitrite (NO₂), other oxidation products of ammonia are N₂O and H⁺ ions, generating temporary acidification of the medium as the final product. Under aerobic conditions, N₂O amounts to less than 1% of oxidized ammonia and this ratio increases as oxygen availability is reduced (Heil, Vereecken, and Brüggemann, 2016).

4. Factors Influencing N₂O Emissions

The processes that govern nitrogen loss include a complexity of biotic and abiotic factors, as well as management practices, climate and soil properties, which condition the dynamics of nitrogen and carbon in soil. Some of the factors that contribute to the increase of N_2O emissions are the availability of readily assimilable carbon sources from animal manure and agricultural and cover crop residues, as well as soil carbon.

We will now present how pig farming can influence the processes responsible for soil N_2O emission. The influencing factors are divided into: (i) pig nutrition, (ii) manure storage and treatment, (iii) manure composition, (iv) heavy metal presence, and (v) oxygen diffusion.

4.1 Pig nutrition

Reducing labile nitrogen and carbon contents in excreta by managing animal diet may potentially reduce GHG emissions, especially N_2O . In animal excreta, volatile forms of nitrogen are mainly present in urine (Montes et al., 2013). These nutrients are provided especially (or largely) in protein compounds made available in the diet. Thus, the management of the components and contents of the ingredients in the feed may be an important tool to reduce N_2O emissions.

Cu and Zn are used in pig diet as growth promoters and for the prevention/treatment of diarrhea. The effect of the presence of these metals will be presented and discussed in section 4.4.

4.2 Manure storage and treatment

The composting of pig manure is classified as an aerobic treatment. It can be done with passive aeration (natural) or active aeration (oxygen injection). It is an alternative to reduce the volume of manure, facilitating its treatment, transport and disposal to the soil. The addition of carbon-rich material (typically sawdust, shavings or rice husk) reduces the labile fractions of carbon and nitrogen and consequently N_2O emissions. In evaluating CH_4 and N_2O emissions during the composting process in passive aeration, active aeration and liquid manure storage systems, Thompson, Wagner-Riddle and Fleming (2003) found a reduction of 30% in the emissions of these gases in active aeration compared to liquid manure storage. On the other hand, the authors found an increase in emissions of approximately 300% in passive aeration compared to liquid manure storage. Active aeration systems are the most efficient in stabilizing the compost, reducing the anoxic zones and N_2O emissions (Osada, Kuroda, and Yonaga, 2000).

Lagoons are the most commonly used liquid manure system in Brazil. In legislation of the state of Santa Catarina, hydraulic retention time of manure in lagoons should be the equal to the number of days in which the application of manure to the soil was licensed, and should be no less than 40 days (FATMA, 2014). This period is intended to stabilize and reduce organic matter, reduce pathogens and adsorb phosphorus. The discharge rate of the lagoon is defined based on the availability

of farm areas suitable for the application of manure and the critical limit of phosphorus. An alternative to lagoons is the use of anaerobic biodigesters. However, the reduction of the organic loading rate through anaerobic treatment in biodigesters has contrasting effects. On the one hand, methane (CH₄) production occurs through the degradation of part of the organic matter in manure. On the other hand, the degradation of the carbon compounds generates a digest with lower organic loading rate, which may reduce emissions after application in soil. The agronomic value of manure from anaerobic treatment systems reduces emissions at the time of application and throughout the biological transformations occurring in soil (nitrification and denitrification), compared to the raw manure (Dennehy et al., 2017). Additionally, the use of anaerobic treatments is a possible alternative to increase the energy value, reducing the use of fossil fuels and the environmental impacts associated with this energy source, including the emission of greenhouse gases.

In evaluating N_2O emissions in soil fertilized with pig manure from different treatment systems (separation of solid and liquid phase, untreated and anaerobically treated), Bertora et al., (2008) found a significant decrease in the loss of N applied to soil as N_2O , in the following order: untreated liquid manure> liquid fraction> anaerobically treated liquid manure> solid fraction. The authors associate such effects to carbon, fiber and N-NH₄⁺ contents in manure.

4.3 Manure composition: carbon and other nutrients

Carbon and nutrient contents (mainly nitrogen) in pig manure are one of the main factors that impact the emissions of greenhouse gases after application in soil. Pigs are monogastric animals and compared to polygastric animals produce manure with high proportions of biodegradable carbon (Amon et al., 2007), which is available for soil microbial processes.

On average, 80% of N, 78% of P and 95% of K present in animal diet are found in raw manure, which depends on the animal species and the diet. In Brazil, manure is applied predominantly in liquid form in which nitrogen is mainly present in ammonium form. Once in the soil, this form of nitrogen is used as substrate for nitrification and denitrification.

4.4 Heavy metal presence

The presence of heavy metals promotes a decrease in the genetic diversity of microbial communities. On the other hand, some studies show that the presence of these metals may induce the tolerance of microbial communities over time (Philippot, Hallin, and Schloter, 2007). Nitrification and denitrification are sensitive to environmental changes and are influenced by the presence of soil contaminants. The enzymes that catalyze the reduction reactions are activated by the expression of *narG* and *napA* (nitrate reductase), *nirK* and *nirS* (nitrite reductase), *norB* (nitric oxide reductase) and *nosZ* (nitrous reductase) (Philippot, Hallin, and Schloter, 2007).

Studies in literature show the presence of heavy metals such as zinc (Zn^{+2}) , nickel (Ni^{+2}) and cadmium (Cd^{+2}) inhibit the expression of *nosZ*, especially copper (Cu^{+2}) (at concentrations $\geq 0.5 \text{ mg L}^{-1}$) (Gui et al., 2017). This gene is responsible for the activation of N₂O reductase, an enzyme that catalyzes the reduction of N₂O to N₂ and is activated by *nosZ* (Figure 3).



Figure 3. Schematic representation of the enzymes responsible for the denitrification process.

Source: Authors.

The process of reducing NO_3^- to N_2 (dinitrogen) is mediated by enzymes, as described in figure 3. The inhibition of the *nosZ* enzyme by the presence of metals such as Cu^{+2} , in the last stage of the biotic denitrification process, resulting in increases in N₂O emissions to the atmosphere. This phenomenon is likely to occur in areas with a history of surface applications of liquid pig manure. After 32 applications over 10 years, Tiecher et al. (2013) found Cu^{+2} contents available in the topsoil (0.00-0.05 m) varied from 11 to 111 mg kg⁻¹. It is important to note that approximately 90% of the copper content found in soil is in soluble form (in soil solution), wherein the surface layer approximately 30% is in the form of Cu^{+2} and 60% are bound to dissolved organic compounds (De Conti et al., 2016).

4.5 Oxygen diffusion

Oxygen diffusion is associated to soil physical properties, such as structure, texture, and organic matter content, in addition to management practices. Agricultural practices promote soil densification by reducing partial oxygen pressure. Also, with the addition of organic fertilizers in soil, there is increased availability of labile carbon, in addition to nitrogen used as substrate by heterotrophic microorganisms. Thus, the formation of anaerobic microsites occurs by oxygen consumption, generating the necessary conditions for denitrification (Meng, Ding, and Cai, 2005). Denitrification occurs preferentially under conditions water-filled pore space (WFPS, %) above 80%.

Pig manure of growing-finishing phase presents solids contents of approximately 6% (Tavares, 2016). Its application on the soil surface may contribute to reduce the diffusivity of oxygen in soil. In evaluating the effect of liquid pig manure application on oxygen diffusion and N₂O production, Zhu et al. (2015) found a stimulus to the development of anoxic zones and consequently the production of N₂O. This behavior may be associated to the inhibition of the enzymes responsible for the reduction of N₂O to N₂. In addition to the effect of liquid pig manure application, Meijide et al. (2007) found found higher N₂O emissions after the application of liquid pig manure, coinciding with irrigations in which WFPS was above 70%. Soil moisture affect the frequency of *nirK*, *norB* and *nosZ* in soil, increasing in the number of copies of *nosZ*.

5. Direct Emissions of Soils Fertilized With Pig Manure

Studies carried out in several regions of the world (n = 8) show significant variability in N₂O emissions, with cumulative emissions varying from 0.4 to 6.4 kg N-N₂O ha⁻¹ in subtropical regions, 4.6 to 7.1 kg N-N₂O ha⁻¹ in temperate regions and 0.8 to 19.8 kg N-N₂O ha⁻¹ in Mediterranean regions (Table 3).

Reference	Country Climate		Crop	Source	Duration	Soil	Cumulative N ₂ O emissions	Emission factor ^b
					(day)	-	kg N-N ₂ O ha ⁻¹	(%)
Giacomini et al., 2006	Brazil	Subtropical	Black oat	APM	28	Sandy clay	0.4	0.2
Gonzatto et al., 2013	Brazil	Subtropical	Corn	APM	90	Sandy clay	1.2	nd
Gonzatto et al., 2013	Brazil	Subtropical	Corn	APM+OS	90	Sandy clay	3.2	nd
		Subtropical	Corn-					0.77
Aita et al., 2015	Brazil		wheat	APM+DCD	357	Loam	5.0	
		Subtropical	Corn-					1.36
Aita et al., 2015	Brazil		wheat	APM	357	Loam	6.4	
Chantigny et al., 2010	Canada	Temperate	Corn	APM	1095 ^a	Loam	4.6	2.4
Chantigny et al., 2010	Canada	Temperate	Corn	APM	1095 ^a	Clayey	7.1	3.1
Dambreville et al., 2008	France	Mediterranean	Corn	APM	325	Silty loam	0.8	0.38
Dambreville et al., 2008	France	Mediterranean	Corn	LPM	348	Silty loam	1.0	1.07
López-Fernández et al., 2007	Spain	Mediterranean	Corn	LPM	200	Sandy loam	4.6	1.02
López-Fernández et al., 2007	Spain	Mediterranean	Corn	LMPinc	200	Sandy loam	5.1	1.27
Meijide et al., 2007	Spain	Mediterranean	Corn	LPM	142	Sandy loam	8.3	1.3
Meijide et al., 2007	Spain	Mediterranean	Corn	CPM+U	142	Sandy loam	9.3	1.88
		Mediterranean	Corn					1.82
Louro et al., 2015	Spain		silage	LPMinj	126	Silty loam	19.8	

Table 3. Nitrous oxide emissions in areas fertilized with pig manure.

APM = pig manure after anaerobic treatment; APM+OS = pig manure after anaerobic treatment + oat straw; LPM = liquid pig manure; LPMinc = incorporated liquid pig manure; LPMinj = injected liquid pig manure; <math>DCD = dicyandiamide; CPM+U = pig manure compost + urea; nd = not available.

^aAproximately.

^bCalculated from nitrogen loss of the control treatment (Factor used by the IPCC: 1.25)

Source: Authors.

The highest emissions of N_2O seem to be related to soil type (influence of texture), application method (incorporated or injected), manure type (raw or treated), the joint use of manure and chemical fertilization, and the use of nitrification inhibitors. Studies show that emissions are also associated to the effects of soil moisture, carbon and nitrogen in manure, and manure storage time (this reduces contents of easily degradable carbon). Climate and local topographic effects, such as the presence of soils with hydromorphic characteristics also influence emissions. The applicability of global emission factors, such as those proposed by the IPCC (1.25%), is difficult at regional or local scales.

Internationally, another factor associated with the large variability of N_2O emission values may be linked to deficient and limited techniques used for measuring emissions. Static chamber is the most currently used method (Butterbach-Bahl et al., 2013).

6. Control and Mitigation Strategies

6.1 Managing pig nutrition

One of the strategies for reducing greenhouse gas emissions from animal manure is the management of crude protein levels in feed (Philippe and Nicks, 2015). Sanchez-Martín et al. (2017) evaluated the effect of the addition of fiber sources (consisting of N) on the diet of animals to reduce the concentrations of benzoic acid and hippuric acid in manure (feces and urine). The authors found that the addition of fiber sources caused reductions of 47 to 65% in N₂O emissions after manure application in soil.

In evaluating the emissions of N_2O in soil treated with manure from groups of piglets fed with 10 different diets, Velthof et al. (2005) found that regardless of soil texture (clayey or sandy), the lowest emissions were found in soils fertilized with manure from diets with lower protein contents. However, the variation of carbohydrate levels in the diet increased emissions in sandy soil.

6.2 Manure treatment

The use of technologies for pig manure treatment is an important strategy for reducing greenhouse gas emissions. Among the strategies that enable the reduction of N_2O emissions after application to the soil is decreasing the water content of manure, composting and improving anaerobic digestion.

Improving water supply efficiency for housed animals also contributes to reducing emissions after manure application in soil. The reduction of water content in manure through more efficient use of drinkers results in a smaller volume of manure. The use of nipple drinkers can reduce water losses by approximately 19% and 16% in comparison to bite-ball nipple and bowl. This was verified by Tavares et al. (2014) in a study carried out in 15 commercial farms in the growing-finishing phase. Thus, manure has higher solids contents (6% on average) (Tavares, 2016), contributing to the reduction of anaerobic sites in soil. Another alternative for reducing the moisture content in manure is the separation of the solid and liquid phases (Bertora et al., 2008), with subsequent composting of the liquid phase.

Composting of slurry is another alternative for reducing emissions. In evaluating the effect of the addition of liquid pig manure and pig manure compost on the soil, Kariyapperuma, Furon and Wagner-Riddle (2012) found reductions of up to 57% in soil N₂O emissions after the addition of treatments.

As for anaerobic digestion, improving the process reduces degradable carbon, energy availability for nitrifying microorganisms, and N₂O production in soil (Montes et al., 2013).

6.3 Fertilization strategies

The highest N₂O emissions occur mostly after the addition of manure to the soil, which are simultaneous with the peaks of nitrogen availability (Meng, Ding, and Cai 2005; Dambreville, Morvan, and Germon 2008; Gonzatto et al., 2013). Therefore, the addition of slow-release nutrient sources (e.g., pig manure compost or pelleted manure) may be an alternative to delay the processes of nutrient release, especially nitrogen. This promotes improved synchronization between release and the phase of greater crop demand. In addition, split fertilization may be another efficient alternative for reducing N₂O emissions. Shcherbak, Millar, and Robertson (2014) have shown through meta-analysis that nitrogen fertilization in doses \leq 50 kg N ha⁻¹ reduce N₂O emission factor more than the value recommended by the IPCC.

Another efficient alternative is the increased uptake of soil nutrients by crops. Genetic improvement programs for agricultural crops could select cultivars in locations with high nutrient content, thus allowing the development of varieties that will adapt to these conditions. Fertilizers are typically applied with high amounts of nutrients per hectare. This results in

readily available nutrients, especially nitrogen for the microbial processes responsible for N_2O emissions, which increase exponentially with increasing nitrogen levels per hectare (Shcherbak, Millar and Robertson, 2014).

The rate at which nitrogen preferentially present as ammonium in liquid manure is converted to nitrate may result in increased N₂O emissions. When converted to nitrate (NO_3^-) , ammonium nitrogen (NH_4^+) is used as substrate by the denitrifying bacteria.

The mechanism of nitrification inhibitors is to inhibit the bacteria responsible for the oxidation of NH_{4^+} to nitrite (NO_2^-). This will increase the residence time of NH_{4^+} in soil and consequently reduce N_2O and the risks of nitrogen leaching in the soil profile.

One of the most used nitrification inhibitors is dicyandiamide (DCD), which acts on the oxidation of hydroxylamine to NO_2^- , with a pronounced effect on *Nitrosomonas europaea* (Zacherl and Amberger, 1990). In evaluating the effect of liquid pig manure application combined with the use of DCD on N₂O emissions in corn and wheat cultivation in southern Brazil, Aita et al. (2015) found decreases of 60% when treatments were applied in single dose combined with DCD. Therefore, the use of nitrification inhibitors proves to be an efficient strategy, because it delays the nitrification process and reduces N₂O emissions (Meijide et al., 2007).

6.4 Soil and crop residue management

Soil management associated with crop and cover crop residues is a viable strategy from an agricultural standpoint. In the literature, inconsistent emission values are presented in areas managed under no-tillage and conventional tillage. In a twoyear study, Baggs et al. (2003) found higher N₂O emissions in soils managed under no-tillage with cereal residues in comparison to conventional tillage. Soils managed under no-tillage showed higher levels of moisture and organic carbon, forming anaerobic microsites that may increase emissions. On the other hand, Giacomini et al. (2006) did not find increased emissions with liquid pig manure applications in soil managed under no-tillage, compared to soil managed under minimum tillage. However, these authors reported that because of surface application in no-tillage, the formation of surface crusts may reduce oxygen diffusion, forming anoxic zones that contribute to emission peaks after rainfall events. Areas managed under no-tillage with less than 10 years of establishment tend to emit more N₂O than more consolidated systems (Six et al., 2004).

No-tillage combined with the use of cover crops contributes to increased carbon contents in the upper layers of the soil (Ghimire et al., 2017). The contribution of crop residues and their rotation promotes the increase of carbon stocks over the years, resulting in reduced N_2O and CO_2 emissions. The characteristics of crop residues influence the availability of carbon and nitrogen to the soil, and selecting cover crops is a way to reduce N_2O emissions. Several authors have found lower emissions in soils with addition of grass residues, which present higher C/N ratios, and higher lignin and cellulose contents. This increases the half-life ($t^{1/2}$) of the residues in the soil and reduces the release of carbon and nitrogen into the soil (Doneda et al., 2012).

The presence of plants during fallow periods can also be an efficient strategy to reduce N_2O emissions. López-Fernández et al. (2007) evaluated the effect of the presence and absence of corn plants on the magnitude of N_2O emissions and found reductions of approximately 34% in treatments fertilized with pig manure on soil with plants in comparison to those without plants.

6.5 Managing soil biodiversity

The use of soil management and fertilization techniques that promote unfavorable environmental conditions for denitrifying bacteria is a possible strategy to reduce N_2O emissions. Management strategies that increase stable carbon forms in the soil, which contribute to increased diameter of soil aggregates, promote the increase of total soil macroporosity (Loss et

al., 2015). As a result, there are fewer environments with low oxygen diffusivity, which contributes to reducing denitrification zones.

Another strategy is to enrich soil with bacteria capable of mitigating emissions. In analyzing N₂O production of indigenous microbial communities of incubated soils from Ireland, Sweden and England after soil enrichment with the addition of a strain of *Dyadobacter fermentans*, Domeignoz-Horta et al. (2016) reported the influence of soil pH values (r = 10.8; P < 0.01) and C/N ratio (r = 19.4; P < 0.01) on the ability of the bacterial strain to mitigate N₂O emissions.

7. Final Considerations

Microbiological processes are mainly responsible for N_2O emissions from the soil into the atmosphere. N_2O emissions are intensified when soils under natural conditions are converted to agricultural areas. This behavior is directly related to the effect of agricultural practices on the biological processes involved in N_2O emissions. Management and fertilization practices (e.g., pig manure) modify the biogeochemical nutrient cycles in agroecosystems, changing contents and availability of nutrient forms in soil, thus increasing N_2O emissions.

Efficient management of animal manure is one alternative to reduce N_2O emissions. The efficient use of water in animal production facilities and nutritional strategies that reduce nutrient contents in manure should be developed to minimize its pollution potential. The use of conservative soil management systems and the development of organic fertilization strategies that improve nutrient uptake efficiency by crops and increase carbon and nitrogen stocks in soil should be encouraged to reduce emissions.

We find few studies in the literature that address the effect of managing pig nutrition, their reflexes on the characteristics of manure and, consequently, on N_2O emissions. Therefore, as a suggestion for future research, we recommend integrated studies of the quantitative and qualitative impacts of the levels and sources of nitrogen in the animals' diets on N_2O emissions after the manure application to the soil. We also recommend studies related to the effects of copper and zinc contents added to the soil via pig manure on enzymes that catalyze the biotic denitrification process in the soil.

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References

Aguilera, E., Luis Lassaletta, A-C., Josette, G., & Antonio, V. (2013). The potential of organic fertilizers and water management to reduce N₂O emissions in Mediterranean Climate cropping systems. A Review. Agriculture, Ecosystems and Environment 164: 32–52. 10.1016/j.agee.2012.09.006.

Aita, C. J., Schirmann, S. B., Pujol, S. J., Giacomini, P. Rochette, D. A., Angers, M. H., Chantigny, R., Gonzatto, D. A., & Giacomini, A. D. (2015). Reducing nitrous oxide emissions from a maize-wheat sequence by decreasing soil nitrate concentration: effects of split application of pig slurry and dicyandiamide. *European Journal of Soil Science* 66 (2): 359–68. 10.1111/ejss.12181.

Amon, T., Barbara, A., Vitaliy, K, Werner, Z., Karl, M., & Leonhard, G (2007). Biogas production from maize and dairy cattle manure-influence of biomass composition on the methane yield. *Agriculture, Ecosystems and Environment* 118 (1–4): 173–82. 10.1016/j.agee.2006.05.007.

Baggs, E. M., M. Stevenson, M., Pihlatie, A. Regar, H., & Cook, G. C. (2003). Nitrous oxide emissions following application of residues and fertiliser under zero and conventional tillage. *Plant and Soil* 254 (2): 361–70. 10.1023/A:1025593121839.

Bertora, C., Francesco, A., Laura, Z., Jan, W. G., Gerard, V., & Carlo, G. (2008). Pig slurry treatment modifies slurry composition, N₂O, and CO₂ emissions after soil incorporation. *Soil Biology and Biochemistry* 40 (8): 1999–2006. 10.1016/j.soilbio.2008.03.021.

Brasil (2014). Estimativas anuais de emissões de gases de efeito estufa. (http://sirene.mcti.gov.br/documents/1686653/1706227/Estimativasd.pdf/0abe2683-e0a8-4563-b2cb-4c5cc536c336)

Butterbach-Bahl, K., E. M. Baggs, M. Dannenmann, R. Kiese, and S. Zechmeister-Boltenstern (2013). Nitrous oxide emissions from soils: how well do we understand the processes and their controls? *Philosophical Transactions of the Royal Society B: Biological Sciences* 368 (1621): 20130122–20130122. 10.1098/rstb.2013.0122.

Chantigny, M. H., Philippe Rochette, D. A., Angers, Shabtai Bittman, K. B., Daniel Massé, G. B., Nikita, E-H., & Marc-Olivier, G. (2010). Soil nitrous oxide emissions following band-incorporation of fertilizer nitrogen and swine manure. *Journal of Environment Quality* 39 (5): 1545. 10.2134/jeq2009.0482.

Dambreville, C., Thierry, M., & Jean, C. G. (2008). N₂O emission in maize-crops fertilized with pig slurry, matured pig manure or ammonium nitrate in Brittany. *Agriculture, Ecosystems and Environment* 123 (1–3): 201–10. 10.1016/j.agee.2007.06.001.

De Conti, L., Carlos, A., Ceretta, P. A. A., Ferreira, C. R., Lourenzi, E. G., Felipe Lorensini, T. L., Tiecher, C. M., Mylena, G. A., & Gustavo, B. (2016). Soil solution concentrations and chemical species of copper and zinc in a soil with a history of pig slurry application and plant cultivation. *Agriculture, Ecosystems and Environment* 216. Elsevier B.V.: 374–86. 10.1016/j.agee.2015.09.040.

Decock, C. (2014). Mitigating nitrous oxide emissions from corn cropping systems in the Midwestern U.S.: potential and data gaps. *Environmental Science & Technology* 48 (8): 4247–56. 10.1021/es4055324.

Dennehy, C., Peadar, G., Lawlor, Y. J., Gillian, E., Gardiner, S., Xie, L. D., & Nghiem, X. Z. (2017). Greenhouse gas emissions from different pig manure management techniques: A Critical Analysis. *Frontiers of Environmental Science & Engineering* 11 (3), 11. 10.1007/s11783-017-0942-6.

Domeignoz-Horta, L. A., M. Putz, A. Spor, D. Bru, M. C. Breuil, S., & Hallin, L. P. (2016). Non-denitrifying nitrous oxide-reducing bacteria - An effective N₂O sink in soil. *Soil Biology and Biochemistry* 103 (December): 376–79. 10.1016/j.soilbio.2016.09.010.

Doneda, A., Celso, A., Sandro, J. G., Ezequiel, C. C. M., Diego, A. G., Janquieli, S., & Rogério, G. (2012). Fitomassa e decomposição de resíduos de plantas de cobertura puras e consorciadas. *Revista Brasileira de Ciencia do Solo* 36 (6): 1714–23. 10.1590/S0100-06832012000600005.

Dunmola, A. S., M. Tenuta, A. P. Moulin, P., & Yapa, D. A. L. (2010). Pattern of greenhouse gas emission from a prairie pothole agricultural landscape in Manitoba, Canadian Journal of Soil Science 90: 243–56. 10.4141/CJSS08053.

FAO (2016). Nitrous oxide emissions in manure management and Manure applied to soils. (http://www.fao.org/faostat/en/#data)

FATMA (2014). Instrução Normativa Nº11 - suinocultura. (http://www.fatma.sc.gov.br/ckfinder/userfiles/arquivos/ins/11/IN%2011%20Suinocultura.pdf} (accessed Jun 2017)

Ghimire, R., Urszula, N., Prakriti, B., Augustine, K. O., & Jay, B. N. (2017). Soil organic matter, greenhouse gases and net global warming potential of irrigated conventional, reduced-tillage and organic cropping systems. *Nutrient Cycling in Agroecosystems* 107 (1): 49–62. 10.1007/s10705-016-9811-0.

Giacomini, S. J., Claúdia, P. J., Celso, A., Segundo Sacramento Urquiaga, and Bruno José Rodrigues Alves (2006). Emissão de óxido nitroso com a aplicação de dejetos líquidos de suínos em solo sob plantio direto. *Pesquisa Agropecuaria Brasileira* 41 (11): 1653–61. 10.1590/S0100-204X2006001100012.

Girotto, E., et al (2010). Acúmulo e formas de cobre e zinco no solo após aplicações sucessivas de dejeto líquido de suínos. *Revista Brasileira de Ciência do Solo* 34 (3): 955–65. 10.1590/S0100-06832010000300037.

Gonzatto, R., et al (2013). Volatilização de amônia e emissão de óxido nitroso após aplicação de dejetos líquidos de suínos em solo cultivado com milho. *Ciência Rural* 43 (9): 1590–96. 10.1590/S0103-84782013000900009.

Gui, M., et al (2017). Effects of heavy metals on aerobic denitrification by strain Pseudomonas Stutzeri PCN-1. *Applied Microbiology and Biotechnology* 101 (4). Applied Microbiology and Biotechnology: 1717–27. 10.1007/s00253-016-7984-8.

Heil, J., H., & Vereecken, N. B. (2016). A review of chemical reactions of nitrification intermediates and their role in nitrogen cycling and nitrogen trace gas formation in soil. *European Journal of Soil Science* 67 (1): 23–39. 10.1111/ejss.12306.

IPCC (2013). Climate Change 2013: The Physical Science Basis. Climate Change 2013: The Physical Science Basis, no. January 2014: 1–169. 10.1017/CBO9781107415324.

Jongbloed, Age W. 2008. Environmental pollution control in pigs by using nutrition tools age. Revista Brasileira de Zootecnia 37: 215-29.

Kariyapperuma, K. A., et al (2012). Non-growing season nitrous oxide fluxes from an agricultural soil as affected by application of liquid and composted swine manure. *Canadian Journal of Soil Science* 92 (2): 315–27. 10.4141/cjss2011-059.

López-Fernández, S., J. A. Díez, P. Hernáiz, A. Arce, L. & García-Torres, A. V. (2007). Effects of fertiliser type and the presence or absence of plants on nitrous oxide emissions from irrigated soils. *Nutrient Cycling in Agroecosystems* 78 (3): 279–89. 10.1007/s10705-007-9091-9.

Loss, A., et al (2015). Carbono orgânico total e agregação do solo em sistema de plantio direto agroecológico e convencional de cebola. *Revista Brasileira de Ciencia do Solo* 39 (4): 1212–24. 10.1590/01000683rbcs20140718.

Louro, A., et al (2015). Nitrous oxide emissions from forage maize production on a humic cambisol fertilized with mineral fertilizer or slurries in Galicia, Spain. *Geoderma Regional* 5: 54–63. 10.1016/j.geodrs.2015.03.004.

Meijide, A., et al (2007). Nitrogen oxide emissions from an irrigated maize crop amended with treated pig slurries and composts in a Mediterranean Climate. *Agriculture, Ecosystems and Environment* 121 (4): 383–94. 10.1016/j.agee.2006.11.020.

Meng, L., Weixin, D., & Zucong, C. (2005). Long-term application of organic manure and nitrogen fertilizer on N_2O emissions, soil quality and crop production in a sandy loam soil. *Soil Biology and Biochemistry* 37 (11): 2037–45. 10.1016/j.soilbio.2005.03.007.

Montes, F, R., Meinen, C., Dell, A., Rotz, N., Hristov, J., Oh, G., Waghorn, et al., (2013). SPECIAL TOPICS -- Mitigation of methane and nitrous oxide emissions from animal operations: III. A review of animal management mitigation options. *Journal of Animal Science* 91: 5070–94. 10.2527/jas2013-6584.

Osada, T, K., & Kuroda, M. Y. (2000). Determination of nitrous oxide, methane, and ammonia emissions from a swine waste composting process. *Journal of Material Cycles and Waste Management*, 51–56. 10.1007/s10163-999-0018-1.

Pereira A. S., et al. (2018). *Metodologia da pesquisa científica*. UFSM. https://repositorio.ufsm.br/bitstream/handle/1/15824/Lic_Computacao_Metodologia-Pesquisa-Cientifica.pdf?sequence=1

Philippe, F., & Nicks, B (2015). Review on greenhouse gas emissions from pig houses : production of carbon dioxide , methane and nitrous oxide by animals and manure. *Agriculture*, *Ecosystems and Environment* 199: 10–25.

Philippot, L., et al (2007). Ecology of denitrifying prokaryotes in agricultural soil. Advances in Agronomy 96 (January): 249-305. 10.1016/S0065-2113(07)96003-4.

Sanchez-Martín, L. A., et al (2017). Diet management to effectively abate N_2O emissions from surface applied pig slurry. Agriculture, Ecosystems and Environment 239 (3): 1–11. 10.1016/j.agee.2016.12.007.

Shcherbak, I., N., & Millar, G. P. R. (2014). Global metaanalysis of the nonlinear response of soil nitrous oxide (N₂O) emissions to fertilizer nitrogen. *Proceedings of the National Academy of Sciences* 111 (25): 9199–9204. 10.1073/pnas.1322434111.

Six, J., et al (2004). The potential to mitigate global warming with no-tillage management is only realized when practised in the long term. *Global Change Biology* 10 (2): 155–60. 10.1111/j.1529-8817.2003.00730.x.

Suleiman, A. K.A., et al (2016). Temporal variability of soil microbial communities after application of dicyandiamide-treated swine slurry and mineral fertilizers. *Soil Biology and Biochemistry* 97: 71–82. 10.1016/j.soilbio.2016.03.002.

Syakila, A., & Kroeze, C. (2011). The global nitrous oxide budget revisited. *Greenhouse Gas Measurement and Management* 1 (1): 17-26. 10.3763/ghgmm.2010.0007.

Tavares, J. M.R., et al (2014). The water disappearance and manure production at commercial growing-finishing pig farms. *Livestock Science* 169 (C). Elsevier: 146–54. 10.1016/j.livsci.2014.09.006.

Tavares, J. M. R. (2016). Modelagem do consumo de água, produção de dejetos e emissão de gases de efeito estufa e amônia na suinocultura. Tese Doutorado, UFSC, Florianópolis, Brasil.

Thompson, A. G, & Fleming, R. (2003). Emissions of N₂O and CH₄ during the composting of liquid swine manure. *Environmental Monitoring and* Assessment, 87–104.

Tiecher, T. L., et al. (2013). Forms and accumulation of copper and zinc in a sandy typic hapludalf soil after long-term application of pig slurry and deep litter. *Revista Brasileira de Ciência do Solo* 37 (3): 812–24. 10.1590/S0100-06832013000300028.

Velthof, G. L., et al (2005). Gaseous nitrogen and carbon losses from pig manure derived from different diets. *Journal of Environmental Quality* 34: 698–706. 10.2134/jeq2005.0698.

Zacherl, B., & Amberger, A. (1990). Effect of the nitrification inhibitors Dicyandiamide, Nitrapyrin and Thiourea on Nitrosomonas europaea. *Fertilizer Research* 22 (1): 37–44. 10.1007/BF01054805.

Zhu, K., et al (2015). Heterogeneity of O2 dynamics in soil amended with animal manure and implications for greenhouse gas emissions. *Soil Biology and Biochemistry* 84: 96–106. 10.1016/j.soilbio.2015.02.012.