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Impact of crop–manure ratios on energy production and fertilizing characteristics of liquid and solid digestate during codigestion

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The influence of maize silage–manure ratios on energy output and digestate characteristics was studied using batch experiments. The methane production, nutrients availability (N and P) and heavy metals' content were followed in multiflask experiments at digestion times 7, 14, 20, 30 and 60 days. In addition, the available nutrient content in the liquid and solid parts of the digestate was evaluated. Aanaerobic digestion favoured the availability of nutrients to plants, after 61 days 20–26% increase in NH_4^+ and 0–36% increase in PO_4^{3-} were found in relation to initial concentrations. Digestion time and maize addition increased the availability of PO_4^{3-}. Inorganic nutrients were found to be mainly available in the liquid part of the digestate, i.e. 80–92% NH_4^+ and 65–74% PO_4^{3-}. Manure had a positive effect on the methane production rate, whereas maize silage increased the total methane production per unit volatile solids in all treatments.

Keywords: anaerobic digestion; codigestion; digestate; energy crops; maize silage; manure

1. Introduction

Manure is a residue that significantly contributes to environmental disturbances including imbalances in nutrient cycles via eutrophication and acidification, and climate change due its related emissions of greenhouse gases and the fossil fuels consumed to handle it.[1]

Anaerobic digestion is one of the technological options available to handle this residue, interesting because it can stabilize it producing energy in the form of biogas while leaving nutrients which are better available for plants.

Biogas energy production from manure is limited due to its high fibre content and low solids content.[2] Therefore, in the last decade and given the supportive European and national legal frameworks for green energy production that guarantee tariffs and support initial investments, codigestion with energy crops has been increasing dramatically. In Germany, for example, the number of digesters using energy crops has increased from about 100 in 1990 to nearly 7800 in 2012.[3] The use of energy crops is an interesting complementary option given they can significantly increase the energy output per unit digester volume.

Maize is the most dominant energy crop used for methane production in Europe, largely attributable to its high-energy yield per hectare of land as compared with other energy crops.[4] Codigestion of maize with manure is also widely implemented because of their complementary positive effect: manure adds nutrients and alkalinity providing digestion stability, whereas maize contains more digestible carbohydrates.[5–8]

In addition to the biogas, the residual product, i.e. digestate, is an important outcome of the digestion process which needs to be properly managed. The digestate has attractive characteristics for reuse in agriculture as the proportion of N readily available for plant growth, as compared with N in undigested manure, is higher in the first year after application.[9,10] In comparison with artificial fertilizers and undigested manure, digestate has been shown to have a more positive impact on maize growth and total N in maize plants grown in acid soils during their early vegetative stages of growth.[11] In this way, digestate can be used as organic fertilizer in crop production, substituting mineral fertilizers and potentially reducing the overall energy demand in a cropping system.[1]

Despite the attractive characteristics of the digestate, attention has been raised regarding its reuse especially in locations where nitrates presence is already high.[12] There is also concern about the potential introduction of environmental risks to population and ecosystems through

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bioaccumulation of harmful heavy metals in the food chain via water or soil pollution.[13,14] Because of the former, the digestate is sometimes transported to alternative sites with concomitant energy and economic costs.[15] Digestate separation processes are already in place; however, a better understanding is needed regarding the properties of liquid and solid fractions with regard to nutrient and heavy metals' content.

In the past, research has been conducted on codigestion as described in [16], using different manure–crop mixtures containing less than 40% crop content in completely stirred tank reactors (CSTRs) at 20 days hydraulic retention time (HRT). It has been reported that at ratios exceeding 30–40% crop to manure and HRTs < 20 days, digestion becomes instable showing fluctuations in pH and volatile fatty acids (VFAs).[16,17] However, our previous research showed that digesting maize by itself is feasible even at HRTs as low as 20 days.[18]

Therefore, with this research work it is our objective to revise the effect on energy production of different crop/manure ratios increasing maize content up to 100% of the mixture and varying the applied digestion time. Digestate properties of liquid and solid fractions, including nutrients and heavy metals' content, are further examined in order to conclude about reusability of the effluent.

2. Materials and methods

2.1. Experimental design

Two types of test were performed, a multiflask batch experiment and a biological methane potential (BMP) test. The first was made to assess different codigestion ratios of maize and manure working under total solids (TS) conditions closer to a full-scale digestion, and the second was performed to calculate the ultimate biodegradability of the input materials and mixture ratios. In the first test, the nutrient and heavy metals' content in the liquid and solid phases also were analysed by using the full content of the bottles per measurement.

2.2. Multiflask experiment

The batch digestion experiments were performed in duplicate using 500 ml serum bottles applying four different input ratios of maize silage and manure, i.e. 100%, 70%, 50% and 30% maize, on volatile solids (VS) basis, and six different digestion periods, i.e. 0, 7, 14, 20, 30 and 61 days. The experiments were carried in two runs, C100 and C50M50, in the first, and C70M30, C30 and M70 in the second. A 5% TS content was applied in both experiments to minimize the diffusion limitation.

The bottles were incubated at 35°C providing continuous mixing at 120 rpm. Serum bottles were inoculated with inoculums adapted to codigesting maize and manure and buffered using sodium bicarbonate according to requirements for optimal pH conditions (0–8 g $NaHCO_3$ l^{-1}).[18] Gas was collected using gas bags attached to the batch bottles. Gas composition, intermediates and digestate characteristics, i.e. pH, nutrients and heavy metals, were followed in time. Distinction was made between the solid and liquid portions of the digestate by centrifuging for 3 min at 3000 rpm (IEC, International Equipment Company, Centra CL3), each portion was analysed separately for inorganic nutrient content. This centrifugation speed was chosen in order to compare the data with the relevant literature [19] and practical applications where centrifugation of manure takes place at speeds up to 4000 rpm.[20] In addition, distinction between inorganic and organic fractions of N and P was made in both fractions by analysing for the inorganic portion of the respective nutrient. Figure 1 shows the approach used for the nutrient analyses and calculations. The shaded blocks correspond to fractions analytically assessed whereas the white blocks were assessed by subtraction.

2.3. Biological methane potential

In addition to the multiflask batch experiments, the ultimate biodegradability of the input materials and mixture ratios was assessed in duplicate using an optimized BMP test protocol previously developed.[21] The remaining methane

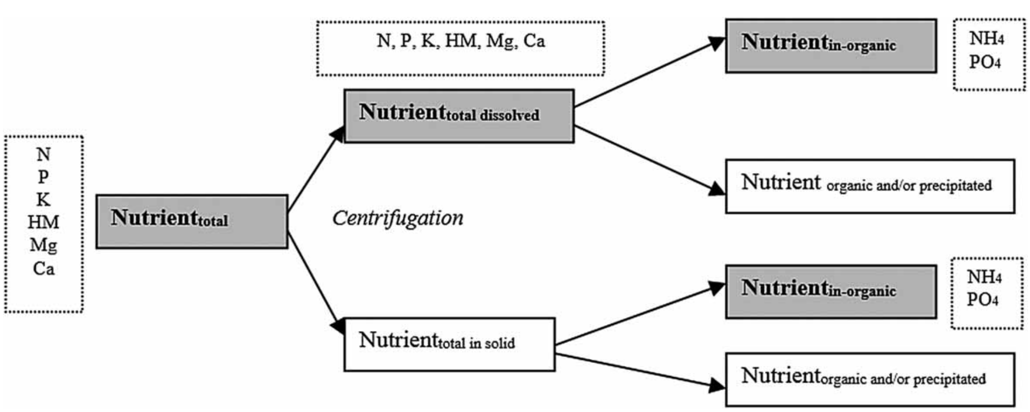

Figure 1. Scheme showing the approach used for nutrient analyses and calculations.

Table 1. Characteristics of input materials.

Parameter	Unit	Maize	Manure	Maize inoculum	Codigestion inoculum
SMA[a]	$g COD gVS^{-1} day^{-1}$	–	–	0.36/0	0.19/0
TS	$gTSg^{-1}$	0.31	0.08	0.09	0.09
VS	$gVSg^{-1}$	0.30	0.07	0.07	0.08
ρ	$g l^{-1}$	395	1004	959	913
COD_{total}	$gCOD gVS^{-1}$	1.17	1.24	1.09	1.06
$COD_{soluble}$	$gCOD gVS^{-1}$	0.50	0.05	0.27	0.34
VFA_{total}	$gCOD gVS^{-1}$	0.06	0.05	0.01	0.05
N_{total}	$mg gTS^{-1}$	12.3	37.2	42.7	42.3
C_{total}	$mg gTS^{-1}$	444	408	401	398
C/N_{ratio}	–	34.60	19.98	16.55	15.79
P_{total}	$mg gTS^{-1}$	1.66	8.37	8.95	8.89
K_{total}	$mg gTS^{-1}$	8.79	17.46	44.26	44.50
Ca_{total}	$mg gTS^{-1}$	1.92	17.57	42.24	11.81
Mg_{total}	$mg gTS^{-1}$	1.70	9.07	4.78	6.72
Fe_{total}	$mg gTS^{-1}$	0.05	0.68	1.22	2.38
$PO_{4-} - P$	$mg gVS^{-1}$	–	6.08	1.41	1.06
$NH_4^+ - N$	$mg gVS^{-1}$	3.19	12.05	18.98	21.81
Alkalinity	$gCaCO_3 l^{-1}$	4.62	6.08	1.41	1.06
pH	–	3.72	7.03	7.92	7.91
Cd	$\mu g gTS^{-1}$	<DL	0.08	0.20	0.29
Cr	$\mu g gTS^{-1}$	<DL	1.03	0.39	6.28
Cu	$\mu g gTS^{-1}$	4.52	69.27	16.14	30.56
Ni	$\mu g gTS^{-1}$	<DL	4.43	4.45	3.05
Pb	$\mu g gTS^{-1}$	0.94	1.77	2.76	2.80
Zn	$\mu g gTS^{-1}$	31	170	104	158

[a]SMA, specific methanogenic activity of the inoculums was carried out following the procedure described in [18].
Note: <DL Below detection limit.

potential in the digestate was then calculated as the difference between the ultimate biodegradability and that obtained at the 61-day digestion time using the multiflask experiment.

2.4. *Origin and characteristics of substrates and inoculums*

Two different inoculums were used, an adapted suspended inoculum from a digester processing only maize and an inoculum adapted to codigesting maize and manure. The suspended inoculum adapted to maize silage digestion originated from the second reactor of a two-phase CSTR system operating on maize silage at 60 days HRT each at 40°C (Corntec, Germany). The codigestion inoculum originated from an anaerobic plug-flow digester codigesting manure, maize and grass silage at 14 days HRT at 41°C. The maize silage originated from a farm in Leer, Germany, it has ensiled for approximately a year. The manure had its origin in the northern part of the Netherlands where it was taken fresh from the manure pit constructed under the cow housing. The cows were fed with a ration of grass silage, maize silage, a waste product from grain fermentation and concentrates. All materials were stored for 1 week prior to be used in the experiments. Full characterization of the input materials is given in Table 1.

2.5. *Analytical methods*

In order to keep ammonium nitrogen in the digestate samples, the total nitrogen was analysed by adding a 0.44 M tartaric acid ($C_4H_6O_6$) solution to the samples in a ratio of 250 g of sample with 300 g of acid. The samples were left to acclimatize for 1 h and dried at 70°C during 1 night (Binder artikel nr: 9010–0080, Tuttlingen, Germany). Samples used for total carbon, calcium, potassium and heavy metals' content were also dried under the same conditions without acid. After drying, the samples were stored or directly grinded to decrease the particle size to ensure a representative sample. Samples for heavy metals, Ca, Mg and K total, were destructed in a microwave (Milestone high Performance Microwave Digestion Unit MLS 1200 mega, Milestone Microwave Laboratory Systems, Sorisole, Italy) by adding approximately 0.5 g of sample and 10 ml of aqua regia (7.5 ml HCl and 2.5 ml HNO_3). After destruction, the samples were quantitatively washed (with Millipore water) and filtrated with Schleicher & Schuell 589[1] ash-free filter paper circles (Schleicher & Schuell GmbH, Germany) into 50 ml flasks. The samples obtained were diluted four times before measuring.

N_{total} and C_{total} were measured using a CE-instruments 1110 CHNS-O Elemental Analyzer with a CHNS column rapped in teflon, length 2 m, external diameter of 6 mm and internal diameter of 4 mm. P_{total}, $NH_4^+ - N$ and $PO_{4-} - P$

were measured using an Auto Analyzer Skalar type 1520 (SAN^{plus} System). P_{total} and $P_{totaldissolved}$ samples were prepared according to Dutch standard methods (NEN 7434, 1998). The inductively coupled plasma optical emission spectrometry (ICP-OES) system used to measure heavy metals content was a Varian Vista-MPX CCD Simultaneous ICP-OES. The total chemical oxygen demand (COD_{total}) analysis was performed by oxidizing the sample in potassium dichromate, the general procedure is described in [21]. To obtain the $COD_{soluble}$ and the total nitrogen dissolved the Dr. Lange tests LCK 514 and LCK 238 were used (Dr. Lange, Düsseldorf, Germany). After the procedure, the samples were measured in a Dr. Lange Xion 500 model LPG-385 photo-spectrometer (Hack Lange GMBH, Düsseldorf, Germany).

The gas composition was measured with a 'Hewlett Packard 5890A (Palo Alto, USA)' gas chromatograph, with the following characteristics: oven temperature: 45°C, injection port: 110°C, detector temperature: 99°C; column length measuring oxygen, nitrogen and methane: 30 m, model Molselve 0.53 mm × 15 µm and column to measure carbon dioxide: 25 m, model Paraplot 0.53 mm × 20 µm. To measure the VFA component of a sample, a Hewlett Packard 5890A gas chromatograph combined with a Hewlett Packard 6890 series injector (Palo Alto, USA) was used. The temperatures of the flame ionization detector, injection port and columns were 280°C, 200°C and 130°C, respectively. The column used was an Altech 14539 AT™-Aquawax-DA with a length of 30 m, internal diameter of 0.32 mm and a 0.25 µm thick coating.

3. Results

3.1. Ultimate biodegradability and multiflask biodegradation

The ultimate biodegradability assessed under optimal test conditions, i.e. with regard to nutrient addition and the use of inoculum mixtures including granular sludge, lasted about 38–40 days. Results showed a proportional increase in specific methane production or BMP values with the increase in the maize fraction of the supplied substrate. The multiflask test showed less clear results with regard to the relationship of the amount of methane produced and the amount of maize in the mixture due to the influence of methane production from the blank bottles (Figure 2). To show the effect of this factor, a correction value is shown in which the methane production from multiflask experiments is corrected with a fixed figure equivalent to the inoculum methane production under BMP test conditions.

Bottles with manure addition were faster in degrading the organic matter, showing around 85% of the methane production after 30 days in codigestion bottles as compared with only 66% in the bottles with solely the crop treatment (Figure 3).

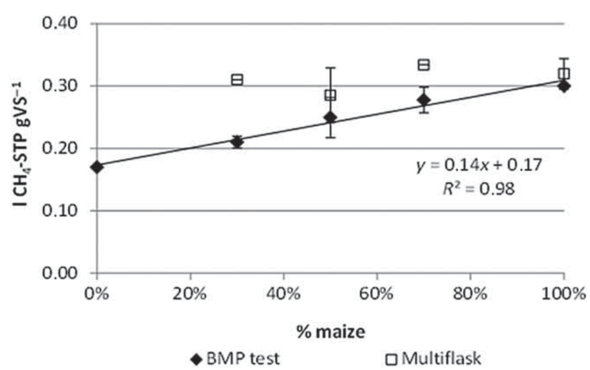

Figure 2. BMP and multiflask biodegradation after 61 days incubation of different mixtures of maize silage and manure (STP = standard temperature and pressure).

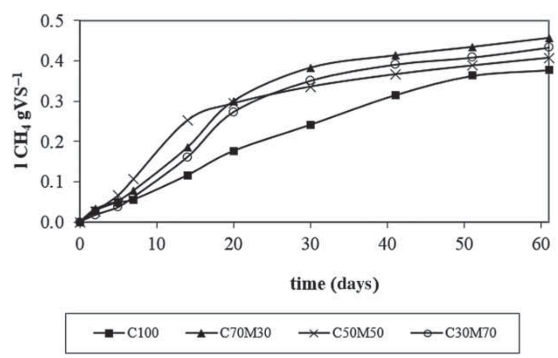

Figure 3. Methane production with time of the multiflask experiment without blank correction.

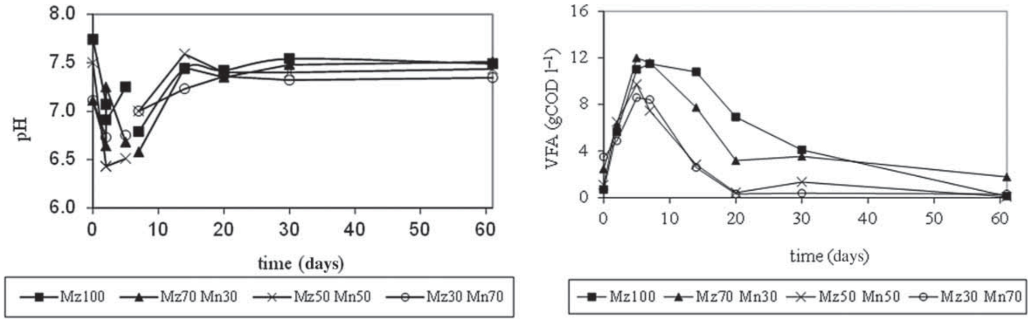

Figure 4. pH (left) and VFA evolution (right) during codigestion of maize silage and manure at different ratios in the multiflask batch experiment. Mz, maize and Mn, manure.

3.2. VFA and pH evolution

In the multiflask batch experiment, the bottles containing manure converted $COD_{soluble}$ and VFA more rapidly as compared with digesters fed with only maize silage. High VFA concentrations (8–12 g COD l^{-1}) were present during the first 15 days of the experiment; acetic and propionic acids accumulated in same proportions up to 5 g COD l^{-1} each. pH remained mostly in the neutral range during the

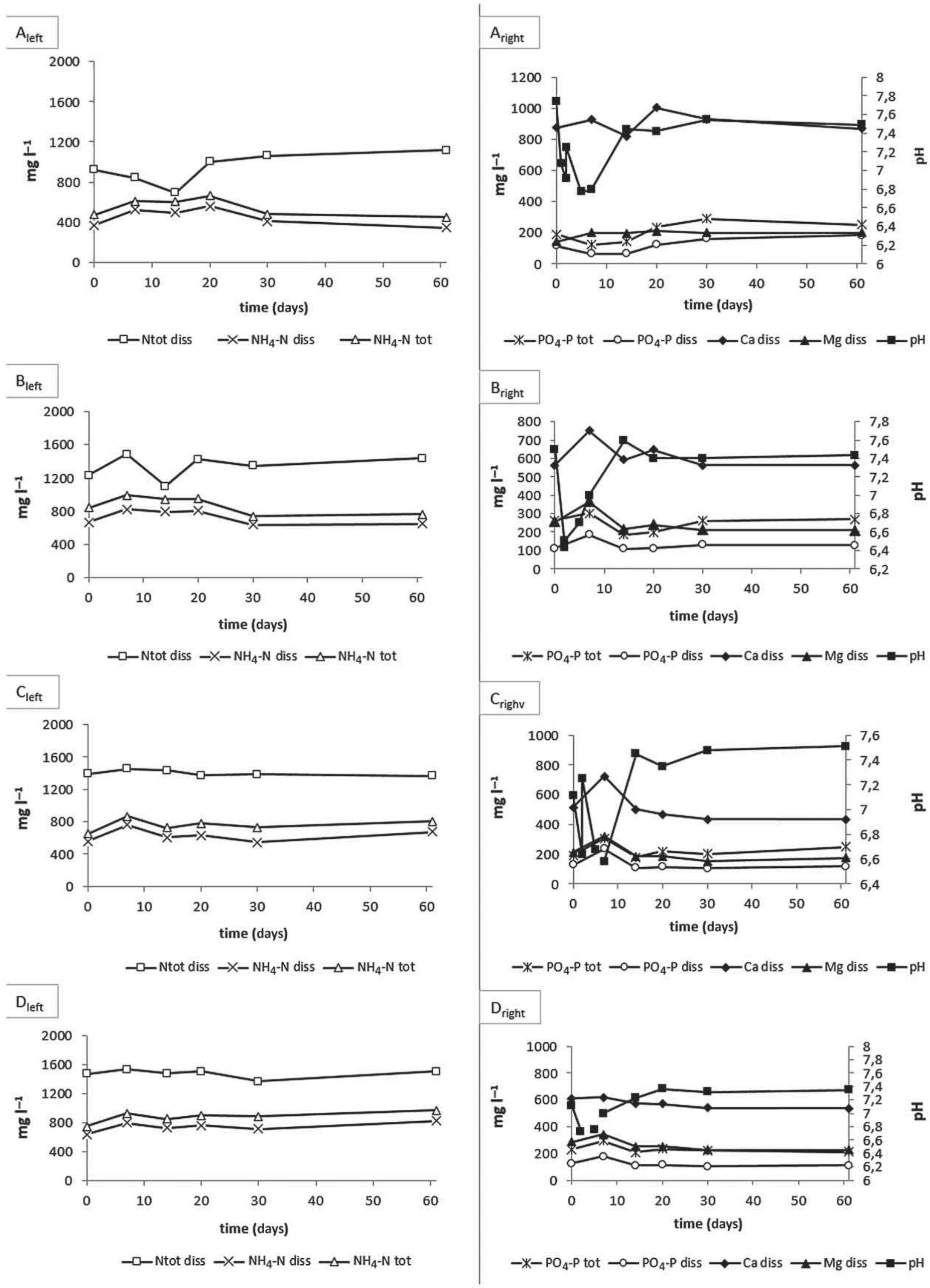

Figure 5. Nitrogen (left) and phosphate, calcium, magnesium and pH (right) evolution with time for different mixtures of maize (C) and manure (M). (a) C100; (b) C50M50; (c) C70M30 and (d), C30M70.

experiment, although fluctuations (6.5–7.8) occurred during the first 14 days of digestion (Figure 4).

3.3. Digestate properties

Total N and P content at the start of the experiment were higher in manure treatments. When incubation started, 38–49% of the total nitrogen was present in the liquid part of the mixture, and of this 40% was NH_4^+. Proportion of $NH_4^+ - N$ with regard to total N was comparable in all treatments, i.e. 19–23%.

Nitrogen evolution in time can be described as follows (Figure 5, left). Treatments showed an initial increase in the dissolved fraction during the first week of the experiment. NH_4^+ increased 25–35% in relation to initial amounts. The initial increase was followed by diminishment during the second week. Thereafter, concentrations returned to original values.

Regarding the presence of NH_4^+ in liquid and solid fractions; in general, 80–92% of was found in the liquid part of the digestate.

Figure 5, right, shows P evolution with time. At the start of the incubation, 32–38% of total P was available as PO_4^{3-} in all the treatments. This fraction was mostly present in the liquid part of the digestate (65–74%). Evolution of PO_4^{3-} in time showed no variation in treatments containing manure, except for day 7 when an increase in PO_4^{3-} was evident. The treatment containing only maize showed a different pattern as the proportion of PO_4^{3-} increased with time. The maximum amount of PO_4^{3-} oscillated between 28% and 63%, whereas the proportion of PO_4^{3-} to total P by the end of the experiment was higher as compared with the start of incubation (20–36% increase) in all treatments except in the treatment with a higher manure content. A similar behaviour was observed in Ca and Mg evolution (Figure 5, right).

The evolution of PO_4^{3-} in the liquid and solid parts of the digestate showed no variation in time with respect to the start of incubation.

Nutrients other than nitrogen and phosphorus and heavy metals were also followed during the experiment.

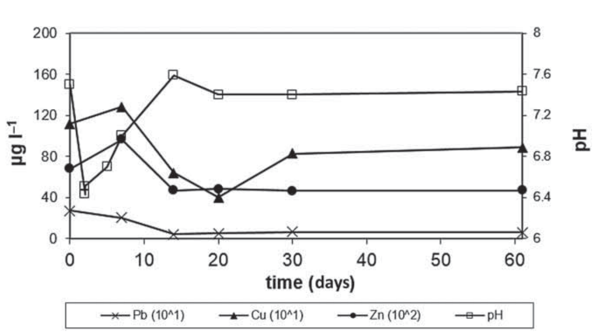

Figure 6. Evolution of heavy metal concentration in liquid portion of the digestate in treatments containing equal proportions of maize and manure.

The concentrations of heavy metals in the course of the batch experiments fluctuated in different ranges as follows: Cd 0.3–0.7 $\mu g\, g TS^{-1}$; 2.6–5.1 $\mu g\, g TS^{-1}$; Ni 6.2–8.0 $\mu g\, g TS^{-1}$; Zn 164–425 $\mu g\, g TS^{-1}$; Cu 39–113 $\mu g\, g TS^{-1}$ and Cr 6.0–11.4 $\mu g\, g TS^{-1}$. Potassium was mostly present in the liquid part of the digestate. Dissolved heavy metal concentrations were higher during the first 14 to 20 days of digestion, thereafter concentrations remain unchanged (Figure 6). Sulphate concentrations, on the other hand, mainly show an increase from day 30 to day 61.

4. Discussion

4.1. Energy production from codigestion of maize and manure

Our results clearly showed a linear increase in the BMP of crop manure mixtures, with an increasing proportion of maize in the mixture. Similar findings were reported in [17] during the digestion of straw–manure mixtures in the fed-batch reactors suggesting that this linear relationship is maintained as long as optimal conditions are ensured.

The obtained net amounts of methane in the multiflask batch experiments exceeded the BMP values. Since the main difference among the two tests was the nutrient content, it is hypothesized that the lower nutrient availability in multiflask test bottles containing maize and manure as compared with the multiflask blank bottles, i.e. those containing only inocula, lower endogenous methane production from inoculum material was present in the test bottles leading to an overestimation of the methane production in the multiflask batch experiment when blank results were subtracted. Therefore, when the value is corrected using the maximum inoculums' biodegradability value, results show better resemblance with those produced by the BMP test.

4.2. Availability of nutrients during codigestion of maize and manure

In this study up to 20–26% increase in N availability was found during the multiflask batch test. Previously Lehtomaki et al. [16] reported an increase in amounts of NH_4^+ in relation to N_{total} of 26%, 25% and 14% during the continuous codigestion at 20 days HRT of manure with sugar beet tops, grass and straw, respectively. Similarly, also other authors have reported the increase in ammonium availability in the digestate as compared with initial conditions in pilot- and full-scale digestion.[22,23] Apparently, the phenomenon is common for both continuous flow and batch digesters operating under stabilized conditions.

Moreover, it is important to recognize that the digestion time exerted an effect in N availability in our experiments. The increase in NH_4^+ concentrations during the first week of the experiments can be attributed to the hydrolysis of proteins present in the individual components. The subsequent decrease during the second week could be related to possible

Table 2. Heavy metal assessed in the multiflask experiment as compared with EU legislation.

Metal country/unit	Cd ($\mu g\,gTS^{-1}$)	Pb ($\mu g\,gTS^{-1}$)	Hg ($\mu g\,gTS^{-1}$)	Ni ($\mu g\,gTS^{-1}$)	Zn ($\mu g\,gTS^{-1}$)	Cu ($\mu g\,gTS^{-1}$)	Cr ($\mu g\,gTS^{-1}$)
EU recommendation	20	750	16	300	2500	1000	1000
EU maximum	40	1200	25	400	4000	1750	1500
This study	0.3–0.7	2.6–5.1	–	6.2–8.0	164–425	39–113	6.0–11.4

Source: [25] and data from this research.

binding and precipitation of the soluble nitrogen cation with the phosphate anion, i.e. via struvite for example, given the pattern observed related to PO_4^{3-} and pH behaviour, as is further elaborated as follows.

In the case of phosphorus an increase in availability was also found. Observed PO_4^{3-} concentrations coincided with pH changes in the medium, especially evident in all treatments having manure addition. Phosphate is known to form many precipitates, such as Fe3(PO4)2, Ca3(PO4)2, MgNH4PO4·6H2O (struvite) and AlPO4.[24] These precipitates dissolve when the digestate becomes more acidic, which is the case during the first period of digestion in this study. Further evidence of the occurrence of this phenomenon is magnesium and calcium which were found to follow a similar pattern as phosphorus concentrations in the digestate. Furthermore, the absence of such a behaviour on the maize treatment can be attributed to the much lower content of Ca, Mg and Fe in maize silage as compared with manure. Despite the possible precipitation observed for magnesium and calcium, the main part of these nutrients was found to be dissolved. After 20 days of digestion, magnesium, calcium and heavy metal concentrations become more or less stable corresponding with a stable pH. A clear positive effect of anaerobic digestion in the availability of phosphorus is seen by the end of the experiments, when the pH is neutral.

4.3. Heavy metal content and availability

Total amounts of heavy metals assessed in the treatments did not exceed the European regulations (Table 2). These results are in accordance with reported results from the assessment of different types of digestates.[9] The fluctuations and changes in the dissolved metal concentrations showed a pattern related to pH fluctuations at the start of the experiment, thereafter remaining constant. Such a pattern implies that a low pH present in the digestate poses higher risk of mobility of heavy metals in the environment and should be taken into consideration when deciding upon the digestate reuse.

5. Conclusions

Codigestion of maize and manure showed different effects from each component in the mixture. Manure had a positive impact on the methane production rate, and in the conversion of intermediates during the experiment and on the total nutrient content of the digestates. Maize silage favoured the ultimate amount of methane produced as well as the mineralized phosphorus.

Anaerobic digestion increased the availability of nutrients. During the experiments NH_4^+ increased up to 25–35% depending on the treatment, whereas maximum increase in PO_4^{3-} oscillated between 28% and 63%.

Digestion time was related to the behaviour of inorganic nutrients. After 7 days maximum amounts of NH_4^+ and PO_4^{3-} were reached thereafter diminishing and further stabilizing.

Fluctuations in dissolved inorganic nutrients coincide with pH changes in the medium. Calcium, magnesium and heavy metals presence in the liquid fraction of the digestate also showed a pH-related pattern. This finding provides an indication that the digestion time, particularly in relation to pH, can be of relevance when deciding about the digestate field applications given its relation to nutrient mineralization.

Furthermore, inorganic nutrients were found to be mainly available in the liquid part of the digestate, 80–92% NH_4^+ and 65–74% PO_4^{3-}, the proportion of total mineralized nutrients in the liquid and solid parts of the digestates remaining mostly constant. The previous implying that separation and diversion of the liquid fraction of the digestate can have a significant impact on nutrient balances in the field, regardless of time.

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