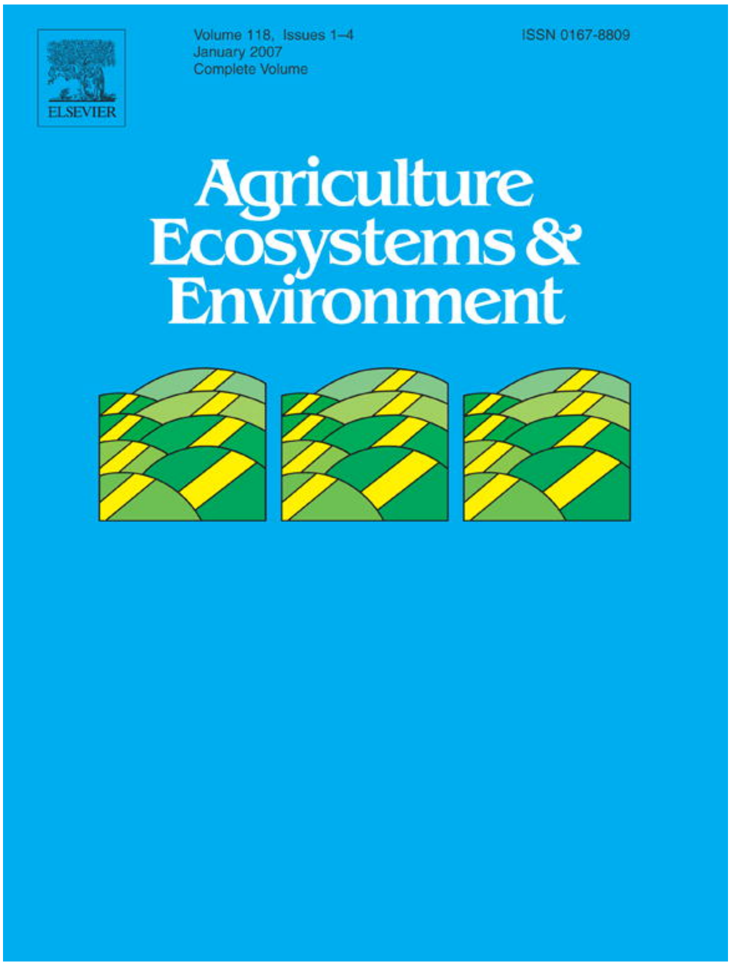

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Biogas production from maize and dairy cattle manure—Influence of biomass composition on the methane yield

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Abstract

There is an increasing world wide demand for energy crops and animal manures for biogas production. To meet these demands, this research project aimed at optimising anaerobic digestion of maize and dairy cattle manures. Methane production was measured for 60 days in 1 l eudiometer batch digesters at 38 °C. Manure received from dairy cows with medium milk yield that were fed a well balanced diet produced the highest specific methane yield of 166.3 Nl CH_4 kg VS^{-1}. Thirteen early to late ripening maize varieties were grown on several locations in Austria. Late ripening varieties produced more biomass than medium or early ripening varieties. On fertile locations in Austria more than 30 Mg VS ha^{-1} can be produced. The methane yield declined as the crop approaches full ripeness. With late ripening maize varieties, yields ranged between 312 and 365 Nl CH_4 kg VS^{-1} (milk ripeness) and 268–286 Nl CH_4 kg VS^{-1} (full ripeness). Silaging increased the methane yield by about 25% compared to green, non-conserved maize. Maize (*Zea mays* L.) is optimally harvested, when the product from specific methane yield and VS yield per hectare reaches a maximum. With early to medium ripening varieties (FAO 240–390), the optimum harvesting time is at the “end of wax ripeness”. Late ripening varieties (FAO ca. 600) may be harvested later, towards “full ripeness”. Maximum methane yield per hectare from late ripening maize varieties ranged between 7100 and 9000 Nm^3 CH_4 ha^{-1}. Early and medium ripening varieties yielded 5300–8500 Nm^3 CH_4 ha^{-1} when grown in favourable regions. The highest methane yield per hectare was achieved from digestion of whole maize crops. Digestion of corns only or of corn cob mix resulted in a reduction in methane yield per hectare of 70 and 43%, respectively. From the digestion experiments a multiple linear regression equation, the Methane Energy Value Model, was derived that estimates methane production from the composition of maize. It is a helpful tool to optimise biogas production from energy crops. The Methane Energy Value Model requires further validation and refinement.

Keywords: Anaerobic digestion; Maize varieties; Harvesting time; Harvesting technique; Methane Energy Value Model

1. Introduction

Biogas production from agricultural biomass is of growing importance as it offers considerable environmental benefits (Chynoweth, 2004) and is an additional source of income for farmers. Renewable energy is produced. The principle of a closed circuit is strengthened, because particularly the nitrogen is being hold stronger in the system (Möller,

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2003). Methane emissions during manure storage are reduced and the fertiliser quality of the digestate is high. Suitable substrates for the digestion in agricultural biogas plants are: energy crops, organic wastes, and animal manures. Maize (*Zea mays* L.), herbage (Poacae), clover grass (*Trifolium*), Sudan grass (*Sorghum sudanense*), fodder beet (*Beta vulgaris*) and others may serve as energy crops (Chynoweth et al., 1993; Gunaseelan, 1997; Weiland, 2003; Tong et al., 1990). Maize is the most dominating crop for biogas production. Maize is considered to have the highest yield potential of field crops grown in Central Europe.

Open questions are quality needs, the yield potential considering the given limits in water availability and thermal time and the integration of energy maize in sustainable cropping systems to minimize negative effects on the environment and to maximise net energy yield (Kauter and Claupein, 2004).

Economic efficiency of anaerobic digestion depends on the investment costs, on the costs for operating the biogas plant and on the optimum methane production (Chynoweth, 2004; Walla and Schneeberger, 2005a).

A maximum methane yield is especially important with the digestion of energy crops as these – in contrast to animal manures or organic wastes – have production costs that have to be covered by the methane production (Walla and Schneeberger, submitted for publication). When energy crops are digested, the methane yield per hectare must be maximised – always bearing in mind not only the single crops, but environmentally friendly crop rotations that deliver maximum methane yields.

The quality of energy crops, used for biogas production, is determined on the field. The content and availability of substances which are able to produce methane is influenced by variety, cultivation and stage of maturity at harvesting time (Amon et al., 2005). Chandler et al. (1980) found several relationships between substrate biodegradability and substrate composition. An estimation of the potential to produce methane of energy crops and animal manures is essential. Maximum methane yield requires adequate and efficient nutrient supply for micro-organisms in the digester.

Existing models concentrated on picturing the kinetics of anaerobic digestion and showing influences such as, e.g. pH value, NH_4–N content, or content of volatile fatty acids (Angelidaki et al., 1993; Batstone et al., 2000, 2001; Henze et al., 1986; McCarty and Mosey, 1991; Pavlostathis and Gossett, 1986). They are only valid for specific areas of digestion of organic wastes. These models were not developed to estimate methane yield from energy crops and to optimise nutrient supply for micro-organisms in the digester of agricultural biogas plants.

Buswell (1936) and Boyle (1977) developed a model that estimates biogas composition (CH_4, CO_2, H_2S und NH_3) from the chemical composition of organic substrates: C, H, N and S. This model does not estimate the methane yield that can be achieved from digestion of organic substrates. Structural substances, especially lignin, are key influences for the digestibility of organic substrates in biogas plants (Amon et al., 2002a; Scherer, 2002; Wellinger et al., 1984). They determine the degradability and thus the methane yield that can be produced through anaerobic digestion. The models of Buswell (1936) and Boyle (1977) do not integrate the influence of lignin. Another shortcoming for the introduction of this model on commercial farms is that it requires C, H, N and S content to be known, which is normally not the case. In the area of animal nutrition, extensive databases are available on the composition of crops that can be fed to animals (e.g. crude fibre, protein, fat content). If a model was developed that can use these databases as input factors, additional costly substrate analyses would not be necessary and commercial farms could easily apply such a model.

Methane production from organic substrates mainly depends on their content of substances that can be degraded to CH_4 and CO_2. Composition and biodegradability are key factors for the methane yield from energy crops and animal manures. Crude protein, crude fat, crude fibre, cellulose, hemi-cellulose, starch and sugar markedly influence methane formation (Amon et al., 2002b, 2003, 2004a; Balsari et al., 1983).

Fig. 1 illustrates influences on the biomass quality considering as example maize for all stages of biogas production. Key influences on the quality of maize for anaerobic digestion can already be found in phase I, when maize is grown on the field. Location, climate and maize variety are important. Plant management and the stage of vegetation when maize is harvested must be optimally chosen to maximise the methane yield. In phase II (harvest, conservation and supply) farmers can positively influence methane yield by choosing the optimum harvesting time and conservation technology and by possibly applying additives. In phase III, energy in the organic substrates is transformed to methane energy in the biogas. Environmental conditions in the digester such as pH, temperature or inhibitors and the nutrient composition of organic substrates determine the methane yield. Amount and quality of the biogas and of the digestate in phase IV result from the influences shown in phases I–III.

The research project aimed at optimising methane production from maize and dairy cattle manure. Influence of performance and feeding intensity on dairy cattle manure composition and on the methane yield from dairy cattle manure was investigated.

Experiments with maize aimed at finding options that achieve a maximum methane yield per hectare. A new model – the Methane Energy Value Model – was developed that estimates methane yield from the nutrient composition of maize via regression models. Factors investigated were: quality criteria for anaerobic digestion of maize, suitability of maize varieties and achievable methane yields per hectare, influence of silaging, optimum harvesting time and optimum harvesting technology.

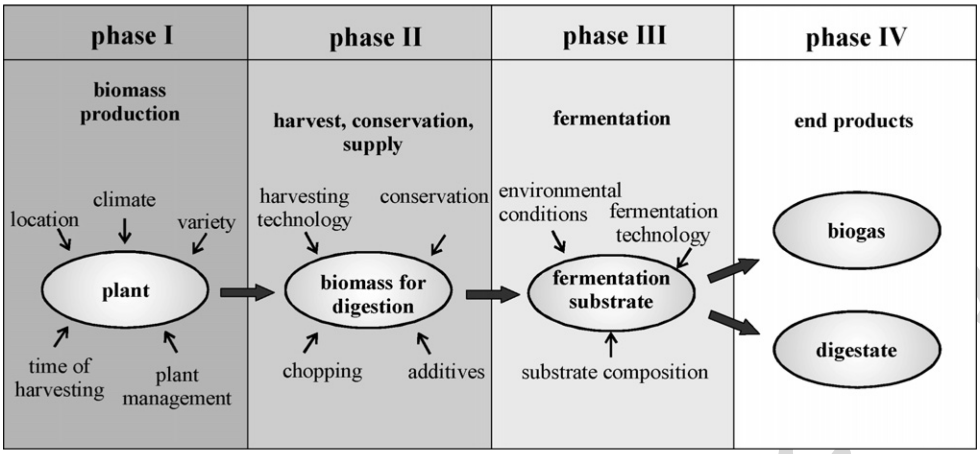

Fig. 1. Influences on biogas production from maize along the production process.

2. Materials and methods

2.1. Dairy cattle manure

The Federal Research Institute for Agriculture in Alpine Regions (HBLFA Raumberg-Gumpenstein) conducted feeding trials with dairy cattle at contrasting milk yields and feeding intensities. The animal diets are listed in Table 1. Milk yield ranged from 11.2 to 29.2 l milk per cow and day. Animal diets differed in their concentrate level and in forage composition (hay, grass silage, maize silage). Methane production from the contrasting dairy cattle manure was measured in eudiometer batch digesters (see Section 2.3).

2.2. Maize for anaerobic digestion

The following maize varieties and locations were included in the experiments:

Year 2001—location: Gross Enzersdorf, Lower Austria (dry region); varieties: PR39G12 (FAO 240), Sandrina (FAO 270), Clarica (FAO 310), Monalisa (FAO 360), Ribera (FAO 390); seeding: 2001-04-26; early harvest: 2001-08-21 (118 days after seeding); medium harvest: 2001-09-03 (131 days after seeding); late harvest: 2001-09-19 (147 days after seeding).

Year 2002—location Ludersdorf, Styria (favourable region for maize production); varieties: Benicia (FAO 300), Ribera (FAO 390), Phönix (FAO 290), Atalante (FAO 290), Saxxo (FAO 380); seeding: 2002-04-30; early harvest: 2002-08-08 (100 days after seeding); medium harvest: 2002-09-12 (143 days after seeding); late harvest: 2002-10-29 (190 days after seeding).

Year 2003—location Ludersdorf, Styria; varieties: Tonale, PR 34G13, Tixxus LZM 650, CSO 271 (FAO-600), Garbure, Ribera, Saxxo, Conca, DKS4626 (FAO 380-400); seeding: 2003-04-25; early harvest: 2003-07-31 (97 days after seeding); medium harvest: 2003-08-25 (122 days after seeding); late harvest: 2003-09-23 (151 days after seeding).

In course of the vegetation period, the following parameters were determined for all varieties: nutrient composition, gross energy, dry matter and organic dry matter content at milk ripeness, wax ripeness and full ripeness; specific methane yield and biogas quality during anaerobic digestion in eudiometer batch experiments; methane yield per hectare for each harvesting time.

In addition, the influence of harvesting technology on the methane yield was investigated. Whole maize crops, corns only, corn cob mix, and maize without corns and cob were anaerobically digested and methane yields were compared. Methane production from silaged maize compared to green,

Table 1
Diet and milk yield of dairy cattle that delivered the manure for the digestion experiments

Treatment	Concentrate [kg DM]	Hay	Grass silage	Maize silage	Milk yield [l day^{-1}]
Dairy-1	0	5.2	10.4	0	11.2
Dairy-2	0	5.4	6.4	5.8	11.2
Dairy-3	4.6	4.0	4.8	5.2	17.6
Dairy-4	5.8	5.0	10.0	0	16.0
Dairy-5	11.0	3.2	3.8	3.6	29.2
Dairy-6	10.0	3.0	6.2	0	29.2

DM = dry matter.

non-conserved maize was measured, as well. A detailed description of cultivation, plant management, and harvesting of maize can be found in Amon et al. (2002b, 2003).

2.3. Measuring methane production

Substance and energy turnover during anaerobic digestion of maize and dairy cattle manure were measured in 1 l eudiometer batch digesters at 38 °C. Methane yields from each treatment were measured in three replicates.

Measurements were conducted according to DIN 38 414 (1985). Each eudiometer consists of six digesters. A water bath tempers the digesters. A magnetic stirrer mixes the substrates for 10 s every 10 min. The biogas is collected in an equilibrium vessel and the biogas production is monitored every day. Biogas production is given in norm litre per kg of volatile solids (Nl (kg VS)$^{-1}$), i.e. the volume of biogas production is based on norm conditions: 273 K, and 1013 mbar (Beitz and Küttner, 1987). Biogas quality (CH_4, H_2S, NH_3) was analysed 10 times in course of the 6-week digestion. Each variant was replicated two to four times. Biogas production from inoculum alone was measured as well and subtracted from the biogas production that was measured in the digesters that contained inoculum and biomass.

Maize was chopped after harvest, prior to the ensiling process. Particle size was 0.5–3.0 mm. Inoculum was received from two biogas plants that digest energy crops (maize, sun flower, grass) at 38 °C. Hydraulic residence time was 70–80 days. 30–70 g maize silage were digested together with 350 g inoculum. Maize silage:inoculum ratio was 1:2 (basis: dry matter). With the digestion of dairy cattle manure, the manure:inoculum ratio was 7:1 (basis: dry matter). This resulted in a dry matter content of the sample of 9% which corresponds to the dry matter content that is commonly found on commercial biogas plants.

Methane concentrations in the biogas were analysed by a Gas Data LMS NDIR analyser (accuracy: ±1–3% of measurement reading). The analyser was calibrated every 10th sample with a 60% CH_4 calibration gas. NDIR readings were validated at regular intervals by gas chromatographic analysis of CH_4 concentration in the biogas. A Shimadzu 14B GC with HP-Plot molecular sieve 5A, and thermal conductivity detector (TCD) was used in isothermal mode. Oven, detector, and injector were operated at 40, 150, and 105 °C, respectively. H_2S concentration in the biogas was analysed two times per week with different Dräger tubes (1D, measurement range (m.r.): 1–200 ppm; 100 A, m.r.: 100–200 ppm, 0.2%/A, m.r.: 0.2–7 vol.%). H_2S concentration was analysed via a chemical reaction: $H_2S + Pb^{2+} = PbS$ (brown colour) + 2H. Accuracy was ±5–10% of measurement reading. NH_3 concentration was measured with Dräger tubes Type 5/b ammonia (measurement range: 5–100 ppm). A pH indicator gives a blue colour if it comes in contact with NH_3 (accuracy: ±10–15% of measurement reading).

Substrates were analysed prior to digestion for pH, dry matter (DM), crude protein (XP), crude fibre (XF), cellulose (Cel), hemi-cellulose (Hem), starch (XS), sugar (XZ), lignin (ADL), crude fat (XL) and ash (XA) with standard analysing procedures. Gross energy (GE) was measured with a calorimeter and is given as MJ per kg of dry matter. A detailed methodology description can be taken from Amon et al. (2003).

2.4. Statistical data analysis

Statistical data analysis was carried out with the software package SPSS, version 11.5 (SPSS Inc., 2005). Each treatment was measured in three replicates. In a first step, the data were summarised by descriptive statistics. Mean, standard deviation and frequency distributions of the data were determined. Differences between treatments were tested with comparative statistics. Variance analysis methods were applied to find significant differences in the means. The following tests and procedures were used: ANOVA and the one factorial post hoc tests “Student–Newman–Keuls” and “Scheffe”. Homogenity of Variances was analysed with the Levene test statistic. Normal distribution was checked by the rule $0.9 < \text{mean} < 1.1$ and $3\ s < \text{mean}$ (Sachs, 1992). The Methane Energy Value Model was developed by carrying out a multifunctional analysis of full regression models (Sachs, 1992).

3. Results and discussion

3.1. Biogas production from dairy cattle manure

Table 2 gives the nutrient composition of the contrasting dairy cow manures: pH, dry matter (DM), crude protein (XP), crude fibre (XF), cellulose (Cel), hemi-cellulose (Hem), lignin (ADL), crude fat (XL), ash (XA) and gross energy (GE). Biogas and methane yield per norm litre of volatile solids are listed as well.

Dairy cows of the treatments dairy-1 and dairy-2 had a low milk yield, dairy-3 and dairy-4 had a medium milk yield and dairy-5 and dairy-6 had a high milk yield. In each level of intensity, manures with contrasting crude protein levels were produced. The manures with the higher crude protein levels (dairy-1, 3, and 6) gave higher methane yields during anaerobic digestion. Lignin in the manure reduced the specific methane yield. The higher the feeding intensity and the milk yield, the greater was the reduction in methane yield through an increase in lignin content. Manure of the treatment dairy-3 was received from cows with medium milk yield that were fed a well balanced diet. Forage consisted of hay, grass silage and maize silage. Concentrate was supplemented according to the cows' requirements. Manure of the treatment dairy-3 produced the highest specific methane yield of 166.3 Nl CH_4 (kg VS)$^{-1}$.

Table 2
Composition of dairy cow manure and specific biogas and methane yield

Treatment	Composition of dairy cow manure [g (kg DM)$^{-1}$]									Gas yield[a] [Nl (kg VS^{-1})]		
	pH	DM[b]	XP	XF	Cel	Hem	ADL	XL	XA	GE [MJ]	Biogas	Methane
Dairy-1	6.95	143.7	162.6	265.9	194.7	144.0	162.1	46.4	157.1	15.8	208.2	136.5
Dairy-2	6.79	128.8	154.3	265.8	227.3	175.9	128.2	34.5	155.0	17.3	213.1	131.8
Dairy-3	6.60	135.0	156.6	310.1	250.8	190.3	124.7	23.8	131.7	14.6	245.8	166.3
Dairy-4	6.60	159.6	150.6	279.5	164.1	187.9	183.3	29.1	162.8	19.3	222.5	143.1
Dairy-5	6.70	148.5	180.2	273.3	161.8	208.7	190.4	28.5	148.4	15.6	238.9	125.5
Dairy-6	6.66	157.3	296.5	248.5	210.1	195.5	121.7	30.3	167.8	16.8	267.7	159.2

DM = dry matter; XP = crude protein; XF = crude fibre; Cel = cellulose; Hem = hemi-cellulose; ADL = lignin; XL = crude fat; XA = crude ash; GE = gross energy.

[a] Nl = norm litre (273 K, 1.013 bar).

[b] [g (kg FM)$^{-1}$].

Brachtl (2000) and Thomé-Kozmiensky (1995) digested cattle manure and found biogas yields between 200 and 300 l biogas (kg VS)$^{-1}$.

Braun (1982) conducted an intensive literature search on biogas production from cattle manure and found a range between 140 and 266 Nl biogas (kg VS)$^{-1}$. The range corresponds well with our experiments that gave biogas yields of 208–268 Nl (kg VS)$^{-1}$. Most of the biodegradable carbon in cattle feed is already digested in the rumen and in the gut. Thus, cattle manure has a lower potential to produce biogas than pig or poultry manure. CH_4 concentration in the biogas is lower (Weiland, 2001).

In agreement with our results, Balsari et al. (1983) found the lignin and cellulose content of cattle diets to influence biogas and methane production from dairy cattle manure. A model was developed that estimates biogas and methane yield from carbohydrate, fat und protein content of cattle manure. Lignin content in cattle manure, which is determined by lignin content in the animal diet, was a key influence on biogas production. Feed lignin content correlates with the vegetation period and a variation can be observed in course of the year. Amon et al. (2001) measured methane production at a commercial biogas plant for 1 year. The biogas plant digested dairy cattle and pig farmyard manure. Specific methane production was not constant throughout the year. When the dairy cattle diet changed from winter feed to summer feed, specific methane production increased. Winter feed consisted mainly of hay. In spring and summer fresh clover grass was fed.

3.2. Biogas production from maize

Maize was harvested at three different times in course of the vegetation period. Net total maize yield per hectare, and specific methane yield per kg VS were measured at each harvesting time. Methane yields per hectare was calculated. Correlations between harvesting technology and methane yield were investigated.

A regression equation was established that estimates methane production from anaerobic digestion of maize from its nutrient composition.

3.2.1. Influence of silaging on the specific methane yield

Investigations on the influence of silaging on the specific methane yield were carried out with the maize variety Ribera (FAO 380). Three replicates of ensiled and green maize were anaerobically digested. Ensiling conditions were optimal for the production of lactic acid: maize was chopped, compacted and stored under anoxic conditions. Degradation of sugars to lactic acid goes along with a very small energy loss of about 3% (Buchgraber et al., 1994). Maize silage yielded 289 Nl CH_4 VS^{-1} (standard deviation of three replicates $\pm$10.8 Nl CH_4 VS^{-1}). Green, non-conserved maize only produced 225 Nl CH_4 per kg VS (standard deviation of three replicates $\pm$7.1 Nl CH_4 VS^{-1}) which is ca. 25% less than silaged maize. During the silaging process lactic acid, acetic acid, methanol, alcohols, formic acid, H^+ and CO_2 are formed. These products are important precursors for methane formation (Madigan et al., 2000). Another reason for the increase in specific methane yield could be a pre-decomposition of crude fibre in course of the silaging process, which improves the availability of nutrients for the methanogenic metabolism.

3.2.2. Influence of harvesting time on the biomass yield, on the specific methane yield and on the methane yield per hectare

The influence of harvesting time on the biomass yield, on the specific methane yield and on the methane yield per hectare is illustrated with late ripening maize varieties (FAO ca. 600). Results from investigations from early and medium ripening varieties (FAO 240–390) can be found in Amon et al. (2004a,b,c).

Fig. 2 gives the biomass yield per hectare of late ripening maize varieties in course of the vegetation period. Dates were gained from the following maize varieties: Tonale, PR34G13, Tixxus, LZM 600, CSO271 (FAO 600), Garbure, Ribera, Saxxo, Conca, DKC4626 (FAO 380–400). Biomass yield of late ripening maize varieties (FAO 600) increased until full ripeness of the maize plants. Earlier experiments with early and medium ripening varieties (FAO 300–400) only showed an increase in biomass yield until wax ripeness. The latest harvest at full ripeness resulted in a loss in net total

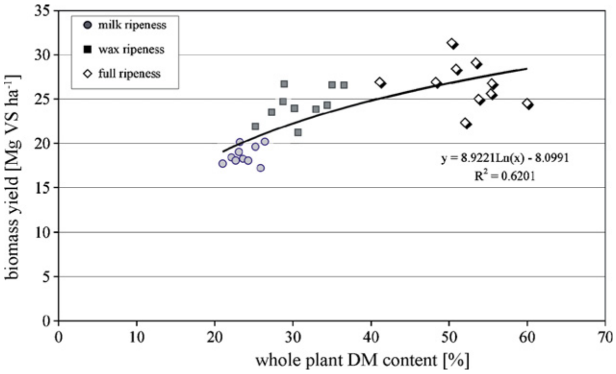

Fig. 2. Biomass yield of late ripening maize varieties at different stages of vegetation.

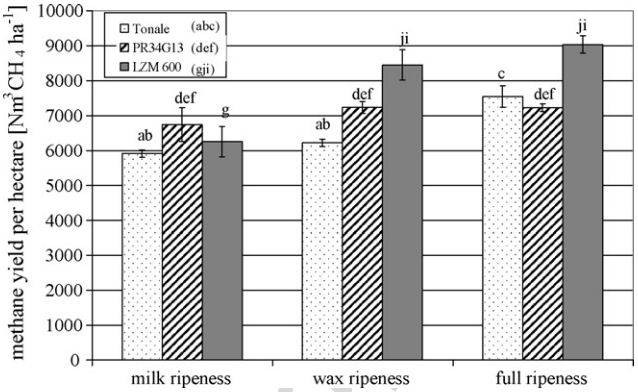

Fig. 3. Methane yield per hectare of late ripening maize varieties at different stages of vegetation with standard deviation from three replicates per variety and vegetation stage. Different letters indicate significant differences at $p < 0.05$.

biomass yield (Amon et al., 2002b, 2003). The reduction in biomass yield from late harvesting of early ripening maize varieties may be due to respiration and/or breakage losses (Zscheischler et al., 1984). According to Zscheischler et al. (1984) the optimum harvesting time for maize is reached at a dry matter content of 30–35%. Maize can then easily be silaged and gives maximum biomass yields.

At milk ripeness, the VS yield varied between 17.2 Mg VS ha^{-1} (Garbure) and 20.2 Mg VS ha^{-1} (Conca, LZM 600). At wax ripeness, the VS yield increased to 21.9–26.7 Mg VS ha^{-1}. At full ripeness, 22.3–31.4 Mg VS ha^{-1} had been produced. In Austria, the mean maize yield in the years 2000–2003 was ca. 43 Mg fresh matter ha^{-1}. Assuming a dry matter content of 30%, this corresponds to a medium yield of 12.9 Mg VS ha^{-1}. The medium maize yield in the EU (EU15) is about 42.1 Mg ha^{-1}, which corresponds to ca. 12.6 Mg VS ha^{-1} (Eurostat, 2003).

The methane yield per hectare is the product of biomass yield and specific methane yield per kg VS. Fig. 3 gives the methane yield per hectare in course of the vegetation period for three late ripening maize varieties. Schumacher et al. (2006) found similar methane yields per hectare from maize grown in Germany. The specific methane yield is shown in Table 3. It ranged from 312 to 365 Nl CH_4 kg VS^{-1} (milk ripeness) to 268–286 Nl CH_4 kg VS^{-1} (full ripeness). The specific methane yield declined towards full ripeness. Oechsner et al. (2003) carried out digestion experiments in discontinuous digesters according to the "Hohenheim biogas yield test". Substrates were digested for 36 days at 37 °C. When maize was harvested at or near full ripeness at a dry matter content of 30–42%, medium biogas yield was 375 Nl CH_4 kg VS^{-1}. Harvesting before wax ripeness at a dry matter content of 22.2% resulted in methane yields between 310 and 350 Nl CH_4 kg VS^{-1}.

The methane content in the biogas ranged from 55 to 62% (mean: 58.5%, $n = 100$). H_2S (mean: 140.6 ppm; $n = 60$) and

Table 3
Composition and specific methane yield from late ripening maize varieties

Treatment		Composition of maize varieties														CH_4-yield Nl CH_4 (kg VS)$^{-1}$	
Maize variety	Harvest no.	[% DM]												[% FM]		Specific CH_4-yield	S.D.
		XP	XL	XF	XA	XX	ADL	Cel	Hem	C	XS	Sugar	C/N	DM	VS		
Tonale	**1**	10.1	1.4	34.5	5.3	48.8	6.4	36.2	25.3	49.6	1.20	0.3	24.2	19.4	18.4	334	5.7
Tonale	**2**	7.9	2.1	26.2	4.8	59.0	5.3	28.6	38.0	49.9	20.2	1.0	39.6	29.8	28.3	283	4.9
Tonale	**3**	6.9	1.5	20.3	2.9	68.3	4.8	22.2	30.4	50.1	32.1	2.9	45.1	43.1	41.8	280	11.4
PR34G13	**1**	9.2	1.2	30.8	4.1	54.7	8.6	33.8	25.4	50.6	4.1	1.5	24.9	18.0	17.2	366	26.2
PR34G13	**2**	7.8	2.5	23.8	4.5	61.4	5.5	26.1	32.7	50.5	27.4	0.8	33.5	28.2	26.9	302	7.0
PR34G13	**3**	7.2	2.2	26.3	3.5	60.7	6.7	28.9	35.9	50.9	25.5	2.4	46.2	43.0	41.4	268	4.2
Tixxus	**1**	7.9	1.2	34.9	4.9	51.1	5.3	37.1	26.4	50.3	2.9	0.3	37.0	19.4	18.4	n.m.	n.m.
Tixxus	**2**	6.9	2.3	24.7	5.2	61.0	4.5	25.0	35.5	50.3	25.5	1.1	44.1	30.2	28.6	322	11.7
Tixxus	**3**	5.9	2.6	23.4	4.2	63.9	4.6	23.8	36.2	51.0	30.9	4.8	52.1	52.9	50.7	n.m.	n.m.
LZM 600	**1**	7.8	1.3	35.6	4.1	51.2	7.5	37.3	26.1	50.4	1.2	0.5	43.5	18.1	17.4	313	21.4
LZM 600	**2**	6.7	2.4	27.2	5.3	58.4	6.1	27.5	33.7	49.6	22.6	0.4	42.1	29.0	27.5	326	16.1
LZM 600	**3**	6.7	2.4	18.7	2.8	69.4	4.3	19.3	34.2	49.3	44.6	0.3	42.2	48.0	46.7	287	7.8

n.m. = Not measured; harvest no. 1 = harvest after 97 days of vegetation at milk ripeness; harvest no. 2 = harvest after 122 days of vegetation at wax ripeness; harvest no. 3 = harvest after 151 days of vegetation at full ripeness; FM = fresh matter; XP = crude protein; XL = crude fat; XF = crude fibre; XA = crude ash ; XX = nitrogen free extracts; ADL = lignin; Cel = cellulose; Hem = hemi-cellulose; XS = starch; C/N = C:N ratio; DM = dry matter; VS = volatile solids; Nl = norm litre (273 K, 1.013 bar).

NH_3 (mean: 20.7 ppm, $n = 27$) content in the biogas were low. Methane yield per hectare was highest at full ripeness. It ranged from 7226 $Nm^3\ CH_4\ ha^{-1}$ (PR 34G13) to 9039 $Nm^3\ CH_4\ ha^{-1}$ (LZM 600). With PR 34G13 and LZM 600, the biggest increase in the methane yield per hectare was observed from milk ripeness to wax ripeness. At full ripeness, only a small additional increase was observed.

It was shown, that biomass yield and specific methane production develop in opposite directions in course of the vegetation period. The methane yield per hectare is predominantly influenced by the maize variety and by the time of harvesting. Gunaseelan (1997) measured the biogas yield from sweet sorghum (Rio cultivar) and found that different plant parts, harvesting frequency, plant age, clonal variations, nutrient addition, and particle size reduction have a substantial effect on the CH_4 yield.

Maize is optimally harvested, when the product from specific methane yield and VS yield per hectare reaches a maximum. With early to medium ripening varieties (FAO 240–390), the optimum harvesting time is at the ‘‘end of wax ripeness’’. Maize has then a dry matter content of 35–39% (Amon et al., 2004c). Late ripening varieties (FAO ca. 600) may be harvested later, towards ‘‘full ripeness’’ at a dry matter content of ca. 44%. On fertile locations, late ripening varieties should be grown as these make better use of their potential of biomass production.

3.2.3. Influence of harvesting technology on the methane yield per hectare

Maize can be harvested as whole maize crops, maize corns or corn cob mix.

When maize is used for energy production in biogas plants, the harvesting technology must be chosen that delivers the highest methane yield per hectare. The harvesting technology determines the biomass yield per hectare and the specific methane yield from the digested substrate. Fig. 4 shows the biomass yield of whole maize crops, maize corns, corn cob mix and maize without corns and cob.

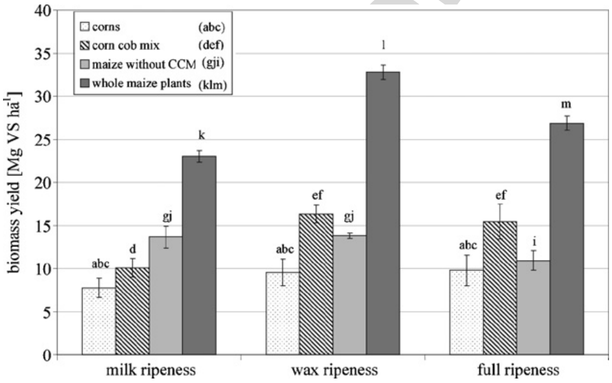

Fig. 4. Biomass yield from whole maize crops, maize without corns and cob, corn cob mix and corns only at different stages of vegetation (varieties: Benicia, Ribera, Saxxo) with standard deviation from three replicates per treatment and vegetation stage. Different letters indicate significant differences at $p < 0.05$.

The biomass yield of whole plants was significantly different in the three harvests. Different letters indicate significant differences at $p < 0.05$. The highest biomass yield of whole plants was achieved in the vegetation stage wax ripeness. The biomass yield of maize without corn and cobs in the vegetation stages milk and wax ripeness was not significantly different, and declined to the vegetation stage full ripeness.

The biomass yield of corn cob mix was lowest at milk ripeness. The vegetation stage had no significant influence on the biomass yield of maize corns.

The specific methane yield was measured from the maize variety Benicia (FAO 300). Benicia was harvested at milk ripeness (22.3% DM), at wax ripeness (ca. 36.5% DM) and at full ripeness (48.4% DM). After 60 days of anaerobic digestion, whole maize crops (gross energy content 19.2 MJ kg VS^{-1}) had produced 326 Nl CH_4 kg VS^{-1} (±6.6 Nl CH_4 kg VS^{-1}, $n = 3$). Corn cob mix (GE = 17.3 MJ kg VS^{-1}) yielded 316 Nl CH_4 kg VS^{-1} (±7.5 Nl CH_4 kg VS^{-1}, $n = 3$). From corns only (GE = 16.7 MJ kg VS^{-1}) a specific methane yield of 309 Nl CH_4 kg VS^{-1} (±7.1 Nl CH_4 kg VS^{-1}, $n = 3$) was measured. Maize without corns and cob (GE = 18.2 MJ kg VS^{-1}) produced 274 Nl CH_4 kg VS^{-1} (±7.1 Nl CH_4 kg VS^{-1}, $n = 3$). Whole maize crops contained more nutrients that are suitable for methane production than corn cob mix or corns alone. Specific methane yield of all silages declined in course of the vegetation period. Biomass yield was measured at each harvesting time and the methane yield per hectare was calculated.

From the biomass yield of three maize varieties (Benicia, Ribera, Saxxo) and from the specific methane yield of the maize variety Benicia, the methane yield per hectare was calculated. The highest methane yield per hectare was achieved from digestion of whole maize crops. Digestion of maize without corn and cob, corn cob mix and corns only resulted in a reduction in the methane yield per hectare (Fig. 5). Harvesting at wax ripeness gave the highest methane yields per hectare. Methane yield at wax ripeness was 8778 (±231, $n = 3$) $Nm^3\ ha^{-1}$ for whole maize crops, 4961 (±311, $n = 3$) $Nm^3\ ha^{-1}$ for corn cob mix, 3744 (±341, $n = 3$) $Nm^3\ ha^{-1}$ for maize without corn and cob, and 2403 (±758, $n = 3$) $Nm^3\ ha^{-1}$ for corns only. Digestion of corns only gave only 30% of energy compared to digestion of whole maize crops. This means, that when maize is used for energy production, the whole maize crop should be harvested. Area requirement for a given energy production is then much smaller.

3.2.4. Methane Energy Value Model for maize

Amon et al. (2003) started to develop the Methane Energy Value Model (MEVM) that estimates methane production during anaerobic digestion from the composition of maize. With the results of the experiments presented above, the MEVM was further developed and its accuracy was further

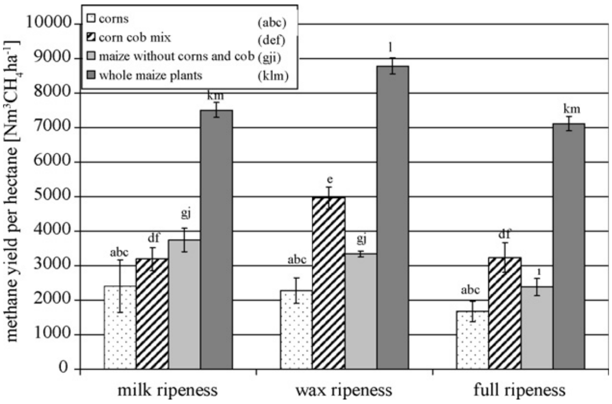

Fig. 5. Methane yield per hectare from whole maize crops, maize without corns and cob, corn cob mix and corns only at different stages of vegetation (varieties: Benicia, Ribera, Saxxo) with standard deviation from three replicates per treatment and vegetation stage. Different letters indicate significant differences at $p < 0.05$.

improved. More experiments and results were available on which the model could be based on. The Methane Energy Value gives the potential of maize silage to produce methane when anaerobically digested in a biogas plant.

Table 3 shows the nutrients that were analysed and the specific methane yield that was measured from experiments with late ripening maize varieties grown at Ludersdorf/Styria in 2003 and calculates the carbon:nitrogen ratio.

The maize varieties showed a characteristic methane production potential that was strongly dependent on their composition. The composition was mainly determined by the stage of vegetation. Crude protein (XP), crude fibre (XF) and cellulose (Cel) content declined in course of the vegetation period. Hemi-cellulose (Hem), N-free-extracts (XX) and starch (XS) content increased. The C:N ratio rose from ca. 24 on the first, early harvest (after ca. 97 days of vegetation) to >42 at the last, late harvest (after ca. 151 days of vegetation). Anaerobic digestion requires a C:N ratio between 10 and 30 (Schattauer and Weiland, 2004).

Table 4
Coefficients of regression, standard error and level of significance for the estimation of methane yield from maize silage from its composition

Nutrient [% DM]	Coefficient of regression	Standard error	Level of significance (p)
Crude protein	19.05	2.95	0.000
Crude fat	27.73	7.09	0.000
Cellulose	1.80	0.40	0.000
Hemi-cellulose	1.70	0.40	0.000

The regression equation is derived from 34 batches with maize, each batch was replicated three times.

When the C:N ratio is too wide, carbon can not optimally be converted to CH_4 and the CH_4 production potential is not fully achieved. When maize was harvested at full ripeness, the C:N ratio was outside the optimum range with regard to producing a maximum specific methane yield. Co-digestion of substrates with a narrower C:N ratio could help to overcome this disadvantage. Location of maize cultivation and variety also influenced the nutrient composition of maize silage. Identical maize varieties grown at different locations differed in their composition (Amon et al., 2004a).

From the digestion experiments, a multiple linear regression equation was derived that estimates methane production from the nutrient composition of maize (Table 4):

$$\begin{aligned}
&\text{Methane Energy Value } [\text{Nl CH}_4\,(\text{kg VS})^{-1}]\\
&\quad = 19.05^{*}\,(\text{crude protein } [\%\text{ in DM}])\\
&\qquad + 27.73^{*}\,(\text{crude fat } [\%\text{ in DM}])\\
&\qquad + 1.80^{*}\,(\text{cellulose } [\%\text{ in DM}])\\
&\qquad + 1.70^{*}\,(\text{hemi-cellulose } [\%\text{ in DM}])
\end{aligned}$$

Table 5
Specific methane yield from anaerobic digestion of maize: measured values and values estimated with the Methane Energy Value Model

Treatment		Specific CH_4 yield measured	S.D.	Specific CH_4 yield estimated (MEWM)	Difference	
Maize variety	Harvest no.	[Nl CH_4 (kg VS)$^{-1}$]		[Nl CH_4 (kg VS)$^{-1}$]	[Nl CH_4 (kg VS)$^{-1}$]	[%]
Tonale	**1**	333.7	5.7	339.4	−5.7	−1.7
Tonale	**2**	283.2	4.9	324.8	−41.6	−14.7
Tonale	**3**	280.4	11.4	266.0	−14.4	5.1
PR34G13	**1**	365.9	26.2	313.6	52.3	14.3
PR34G13	**2**	302.1	7.0	320.7	−18.6	−6.2
PR34G13	**3**	268.2	4.2	311.4	−43.2	−16.1
Tixxus	**2.h**[a]	321.7	6.9	295.1	26.6	8.3
Tixxus	**2.h**[b]	312.8	11.7	299.7	13.1	4.2
Tixxus	**2.h**[c]	326.4	8.5	288.8	37.6	11.5
LZM 600	**1**	312.6	21.4	296.4	16.2	5.2
LZM 600	**2**	325.6	16.1	300.6	25.0	7.7
LZM 600h	**3**	286.8	7.8	286.9	−0.1	−0.0

Harvest no. 1 = harvest after 97 days of vegetation at milk ripeness; harvest no. 2 = harvest after 122 days of vegetation at wax ripeness; harvest no. 3 = harvest after 151 days of vegetation at full ripeness.

[a] Tixxus, 2nd harvest, digested with a mix of the inocula from biogas plants 1 and 2.

[b] Tixxus, 2nd harvest, digested with inoculum from biogas plant 1.

[c] Tixxus, 2nd harvest, digested with inoculum from biogas plant 2.

The nutrients crude protein (XP), crude fat (XL), cellulose (Cel) and hemi-cellulose (Hem) proved to have a significant influence on methane production. From their content – expressed as % in maize silage dry matter – the specific potential of maize to produce methane – its methane energy value – is estimated. The regression equation is based on the experiments shown in this paper and on experiments from earlier results (Amon et al., 2002b, 2003, 2004c). All trials are included that gave a specific methane yield between 250 and 375 Nl CH_4 (kg VS)$^{-1}$.

Table 4 shows coefficients of regression, standard error and level of significance of the regression model for the estimation of methane yield from anaerobic digestion of maize silage. The coefficients of regression are highly significant. They show the contribution of each nutrient to the net total methane yield. Crude fat (27.73) and crude protein (19.05) contribute most to the net total methane energy value of maize silage (Amon et al., 2004a).

Specific methane yields, measured in the eudiometer batch digesters, were compared to the values estimated with the Methane Energy Value Model (Table 5). Estimated values differed between 0.17 and 52 Nl CH_4 (kg VS)$^{-1}$ from the measured values. This corresponds to a difference of 0.1–14.3%. Mean difference was 1.5%. Additional experiments are necessary to further improve the accuracy of the Methane Energy Value Model. In particular, the role of starch for the methane yield has to be investigated in more detail.

4. Conclusions

Anaerobic digestibility of animal manures is markedly influenced by the animal diet and performance. The highest methane yield was achieved from manure that was received from cows with medium milk yield that were fed a well balanced diet.

Maize should be conserved as silage prior to anaerobic digestion as this increases the methane yield. Late ripening varieties (FAO ca. 600) make better use of their potential to produce biomass than medium or early ripening varieties. On fertile locations in Austria they can produce more than 30 Mg VS ha^{-1}. Maize is optimally harvested, when the product from specific methane yield and VS yield per hectare reaches a maximum. With early to medium ripening varieties, the optimum harvesting time is at the "end of wax ripeness". Late ripening varieties may be harvested later, towards "full ripeness". Farmers are advised to harvest maize when the dry matter yield per hectare reaches its maximum and maize can still be silaged.

Maximum methane yield is achieved from digestion of whole maize crops. Digesting corn cob mix, corns only or maize without corn and cob gives 43–70% less methane yield per hectare.

From the digestion experiments, the Methane Energy Value Model was developed. It estimates the methane yield from crude protein (XP), crude fat (XL), cellulose (Cel) and hemi-cellulose (Hem) of maize silage. The Methane Energy Value Model helps to optimise biogas production by the following capabilities: estimation of the methane production of organic substrates from their composition, estimation of the power of agricultural biogas plants in dependency of amount and composition of organic substrates that are digested, recommendations on varieties and optimum harvesting time of energy crops, and estimation of the methane yield per hectare of energy crops.

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