DOI:10.15150/lt.2017.3153



Studies of the decomposition of fibre components in the biogas process by means of in-sacco-batch method

Claudia Demmig, Frank Höppner, Dietmar Ramhold, Michael Nelles

The substrates used in biogas plants are characterized on the basis of their biogas potential. The in-sacco-batch method is an approved method for animal nutrition which determined the digestibility of ingredients of different feedstuffs. The digestibility and the rate of degradation of plant components and fibre components in the biogas process are important parameters. The batch fermentation test for estimating the biogas potential of substrates has no significance in this regard. For the optimization of the biogas process the digestibility of fibre components plays an important role. In this paper the he in-sacco-batch method was adapted for the biogas potential tests. The method allows the characterization different fermentation additives in view of the digestibility of substrates.

Keywords

Biogas potential, lignocellulose, degree of utilization, in-sacco-batch method, process optimization

In biogas production from energy crops, provision of the necessary substrate represents the main cost factor. Major energy suppliers in agricultural biogas production include silages from maize and grass, the loss-free harvesting and conservation of which can lead to substantial cost savings. Timing of harvesting or cutting of these energy crops is dependent on vegetation development and weather conditions. The process of ensiling has been known for decades and can be supported by certain applications such as use of silage additives. In forage production, as well as in the biogas production, the aim must be conservation of the energy potential in the field with as little loss as possible for subsequent application in animal nutrition or in the biogas plant. Application of the in-sacco-batch testing method on a laboratory scale is aimed at, on the one hand, proving this method is suitable for characterisation of energy crop plants while also representing an informative way of investigating biogas production potential. On the other hand, the method should also demonstrate the relationship between anaerobic degradation and timing of the energy crop harvest.

During the vegetation period the relationship between cell contents and cell wall constituents alters, depending on fibre composition (JUNG und BUXTON 1993). Cell contents such as proteins, lipids and sugars from grass and maize silage reduce with increasing plant maturity. Meanwhile, cell wall constituents cellulose, hemicellulose and lignin, increase. In biogas production, cell contents are readily convertible substances. Depending on degree of lignification, and time spend in the biogas process, cell wall constituents are difficult to degrade, thus limiting extent of substrate degradability. Hydrolysis is the first step in the biochemical breakdown of crop biomass by bacteria (ZVERLOV et al. 2010). The hydrolytic breakdown of particularly resistant cellulose fibres in the cell walls takes place especially slowly (SCHWARZ 2004) Furthermore, only a few microorganisms are capable of com-

pletely degrading these structures (Köllmeier et al. 2012). The substrate degradation of a straw-hay mix (50:50) is investigated by Köllmeier et al. (2012). Before the substrate mix is fed into the biogas process, substrate fibre is milled to 5 mm length and dried. Degree of degradation of the straw-hay mix is observed in association with starting pH value. Results show that, with increasing pH, extent of degradability is greater. Additionally, the influence of fermentation residue on degradation was assessed. The higher the percentage of fermentation results, however, it is not clear whether during drying the fermentation acids and alcohols, according to WEISSBACH and STRUBELT (2008) were taken account of in calculation of the extent of degradation.

The in-sacco-batch method in the biogas field has been also described by MARÍN-PÉREZ and WEBER (2012) and MARÍN-PÉREZ et al. (2012), amongst others. MARÍN-PÉREZ und WEBER also presented hydrolysis as limiting step in lignocellulose degradation within the biogas process and investigated the degradability of lignocellulose at different pH levels and with addition of various microorganism cultures. In this case, a dried material was selected as substrate, a mix of straw and hay. The results of this work show that pH value and temperature both influence degradability. Higher degrees of degradability were found by temperatures between 45 and 50°C and at pH values between 6.2 and 6.3. Investigations by (Teller and VANBELLE 1990) into In-Sacco digestibility of grass silages fed to heifers confirm that the time of harvesting has an influence on degradability within the rumen. An influence of wilting on ruminal degradability could not be demonstrated, however. PERČULIJA et al. (2011) investigated degradability by sheep of dry matter and NDF of grass silages harvested at three different times, once again by the In-Sacco method. In these investigations, too, the silages were dried and milled before going into nylon bags and the rumens. The results show that the degree of dry matter degradation is dependent on the harvest time or maturity of grass. The extent of dry matter degradation decreases in-line with increasing crop plant maturity. In the investigations by WEISS et al. (2011) with batch fermentation of grass silage, a zeolite was given so that the colonisation of microorganisms and enzyme activity could be investigated. The zeolite was added in a polyamide sack to investigate the colonisation of microorganisms within a defined area in order to exclude inoculum influences. Degradability degree of the different constituents of grass silage was not observed in these investigations. WROBLEWITZ et al. (2014) investigated the influence of carbon dioxide enrichment of the atmosphere on growth of the maize plant as well as on maize grain nutrient digestibility by cattle. A significant effect of atmospheric CO₂ on degradation of crude protein and aNDFom (ash free Neutral-Detergent-Fibre) could be demonstrated. The degradation kinetic as well as degradability degree were negatively influenced.

Material and Methods

At the Julius Kühn Institute in Braunschweig, a trial plot with a population of Lolium multiflorum (Italian ryegrass) was harvested at four different cutting times, each representing a new growth after an earlier harvest. The first cut took place at beginning of ear emergence, with the three following cuts carried out after ear emergence. After each cut, the material was wilted, homogenised and then ensiled over 90 days in 1.5 litre preserving jars (Weck). The ensiling took place with threefold repetition for each opening day. Following conclusion of the ensiling process, the silage was comprehensively analysed (Table 1). The trial location Braunschweig Völkenrode lies 75 m above sea level with long-term average temperature of 8.7 °C and precipitation 619 mm. Soil type is 30-40 point silty sand (glevic cambisol). The in-sacco-batch trials took place according to BANEMANN et al. (2012) and SÜDEKUM (2005) being schematically presented in (Figure 1). Compared with a biogas potential determination, the fermentation took place in an in-saccobatch trial in a porous nylon bag (ANKOM Technology) with 50 µm mesh. The fermentation residue remaining within the nylon bag can be examined in detail after differing incubation periods. In these trials, incubation periods of 4, 7, 10, 15 and 42 days are selected. Every incubation day was subject to threefold repetition. The fermentation took place in 60 l clamp-ring barrels as batch fermenters, kept in constant-temperature heat cabinets at 40°C, mesophilic conditions. The ball valve on the lid of each container has a gas storage bag (Dr.-Ing. RITTER Apparatbau) attached by coupling connector (CPC Colder Products) for collecting the

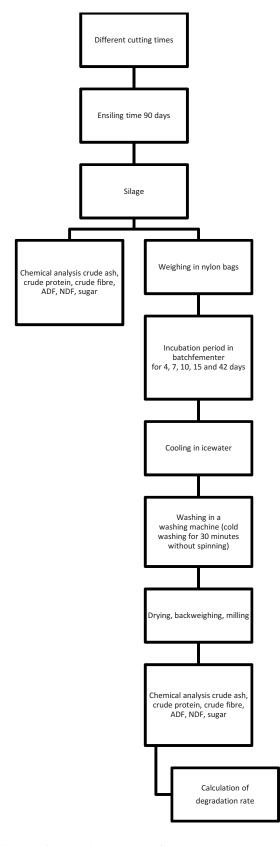


Figure 1: Schematic structure of in-sacco-batch trials

biogas produced. For replacing biogas collection bags at the filling point, the system allows loss-free interruption of flow.

The batch fermenters each contain 20 kg inoculation sludge (mesophilic, municipal waste water treatment plant). The dry matter content (DM) of the sewage sludge is 2.56% FM, the organic dry matter (oDM) being 1.29%. Within this is placed the grass silage substrate in nylon bags. The substrate amount of grass silage, weighed in the nylon bags, is calculated applying equation 1 according to VDI 4630 (VDI 2004):

$$\frac{\text{oDM}_{\text{substrate x weight of sample}_{\text{substrate}}}}{\text{oDB}_{\text{inoculation sludge}} \text{ weight of sample}_{\text{inoculation sludge}} < 0.5$$
(Eq. 1)

In order to reduce headspace volumes, 20 litres water was added. The pH rose slightly through this addition of water, but not significantly. Inert nitrogen was used to drive any air out of the headspace. In conclusion, the batch fermenter was closed airtight. For determining the blank value, only batch fermenters filled with inoculated sludge and water were used. Furthermore, to control inoculated sludge activity, three batch fermenters with inoculum and microcrystalline cellulose (Merck KGaA) were applied. The biogas produced was collected via drum-type gas meter (Dr.-Ing. RITTER Apparate-bau, TG 10, with water sealing liquid and calibration half-yearly) after 4, 7, 10, 15 and 42 days. With a Visit 03 gas detector (Messtechnik EHEIM, measuring precision $\pm 2\%$, calibration half-yearly with test gas), the volume percentage potential of methane, carbon dioxide and hydrogen sulphide could be measured. On the same day as the gas measurement, the collection of material from the nylon bags took place in threefold repetition. The volume of biogas is depending on atmospheric pressure, temperature and vapour pressure of the water. The biogas yield is calculated in accordance to VDI 4630 (VDI 2004) (standard pressure 1013,25 mbar, standard temperature 273,15 K).

After taking the material out of the bags, the nylon bags were placed in ice water to stop microbial metabolic activity. Then the nylon bags were placed in a washing machine (Bomann) and washed in a cold water program for 30 minutes, subsequently dried in a drying cabinet (UF 750, Memmert) at 65 °C through to weight constancy. After backweighing, the remaining fermented material was ground via an ultracentrifuge mill (ZM 200, Retsch, mesh size 1mm). Content constituents were analysed as sugar (VDLUFA B III 7.1.1), ADF (VDLUFA B III 6.1.1), NDF (VDLUFA B III 6.1.1), crude fibre (VDLUFA B III 6.1.1), hemicellulose and crude protein (VDLUFA B III 4.1.1) with each measurement duplicated. Calculation of biogas and methane yield was determined according to VDI 4630 (VDI 2004). With drying of silages, fermentation acids and alcohols are volatile. Because of this, dry matter contents have to be corrected. Hereto, the volatile organic acids were determined by High Pressure Liquid Chromatography (HPLC Smartline, Knauer Wissenschaftliche Geräte GmbH) and these applied in the corrected formula for grass silages according to WEISSBACH and STRUBELT (2008). As HPLC column, an Aminex HPX-87 column 300 x 7.8 mm (BIO-RAD) is used. Pack material in this column is a sulphonated divinyl-benzine-styrine-copolymer. The HPLC fluxing agent is 0.02n sulphuric acid. The flow rate, at pressures between 70 and 90 bar and temperature of 30°C, 0.6 ml/min. Applied as UV detector is a Smartline UV Detector 2500, as IRR detector a Smartline RI Detector 2300 and as liquid pump a Smartline Pump 1000 (all Knauer Wissenschaftliche Geräte GmbH). Fermentation acid content was calculated as mean value from UV and RI detectors. Only sugars and alcohols were detected by the IR detector.

Results

The chemical composition of the respective silages for the in-sacco-batch trial is presented in (table 1). The silage is sampled using the In-Batch method without milling or drying, so that the procedure is as comparable as possible to that carried out with substrates in practice.

Parameter	Unit	Cutting time 1 (CT 1)	Cutting time 2 (CT 2)	Cutting time3 (CT 3)	Cutting time 4 (CT 4) 28.09.2011	
	•	11.05.2011	15.06.2011	02.08.2011		
DMc content ¹⁾	%-FM	27.02 ± 0.32	35.61 ± 0.53	47.92 ± 0.62	54.30 ± 0.26	
Crude ash	%-DM _c	6.07 ± 0.02	7.66 ± 0.10	7.77 ± 0.09	8.57 ± 0.20	
Crude protein	%-DM _c	11.92 ± 0.29	10.64 ± 0.12	9.97 ± 0.13	9.40 ± 0.08	
Sugar	%-DM _c	10.23 ± 0.39	9.77 ± 0.58	9.61 ± 0.42	10.11 ± 0.94	
Crude lipid	%-DM _c	2.54 ± 0.82	3.06 ± 0.06	3.13 ± 0.05	3.60 ± 0.03	
Crude fibre	%-DM _c	20.46 ± 0.15	24.34 ± 0.42	24.51 ± 0.33	26.37 ± 0.31	
ADF	%-DM _c	25.70 ± 0.38	27.80 ± 0.68	28.41 ± 0.28	29.78 ± 0.28	
NDF	%-DM _c	50.38 ± 0.82	48.20 ± 0.38	50.75 ± 0.34	51.48 ± 0.49	
Hemicellulose ²⁾	%-DM _c	24.68	20.40	22.34	21.70	
Lactic acid	%-FM	1.71 ± 0.08	1.82 ± 0.12	1.86 ± 0.10	0.90 ± 0.17	
Acetic acid	%-FM	0.57 ± 0.02	0.65 ± 0.03	0.83 ± 0.28	0.50 ± 0.07	
Ethanol	%-FM	0.11 ± 0.02	0.08 ± 0.07	0.14 ± 0.01	0.10 ± 0.04	
pH-Wert		3.84 ± 0.01	4.04 ± 0.01	4.33 ± 0.10	4.81 ± 0.12	

Table 1: Content of raw nutrients and fibre substances of grass silage (mean ± standard deviation of three replicates

¹⁾ DM_c = corrected dry matter content

²⁾ Calculated as difference.

As with Table 1, the dry matter content increased through wilting from cutting date 1 through to cutting date 4 from 27.02 to 54.30 % FM. The wilting result reflects practical conditions and is aimed at minimising the production of fermentation liquid containing readily available carbohydrates. The sugar content decreased from 10.23 % DM_c at cutting date 1 to 10.11 % DM_c at cutting date 4. Cutting date 2, with a sugar content of 9.77 % DM_c, just as did cutting date 3 with sugar content 9.61 % DM_c, showed a lower sugar content compared with those of the silages from cutting dates 1 and 4. The structural materials, brought together analytically within the parameter NDF, rose from 50.38 to 51.48 % DM_c, whereby the silages from the second cut returned an NDF content of 48.20 % DM_c. The ADF content reflecting the lignocellulose fraction rose from 25.70 to 29.78 % DM_c. To be expected from such results is also a reduction in digestibility within the anaerobic process in sequence with cutting dates. Because cutting dates 2 to 4 represent regrowth after harvest, the contents show only limited differences in composition. The fermentation acid contents shown in table 1 represent those found in practice. The fermentation acids and alcohols are lost during drying for the subsequent analysis and have to be adjusted for in the dry matter corrections (WEISSBACH and STRUBELT 2008).

In Figure 2 the degree of dry matter degradation in the biogas process is presented in relationship to incubation time and cutting date. High degradation rates are already achieved in the first four days of incubation. These are explained through conversion of readily available carbohydrates, including

sugars. Further growth is reduced in extent and occurs more slowly. After 42 days, maximum degradation degree of 95.09% is achieved with cut 1 (see Table 2). With the subsequent cuts, degree of degradation reduces within the same period of incubation. After 42 days, degradation degree for cut 4 is only 85.46%, converting 9.63% less dry matter than cut 1. This trend is also expressed in biogas yield which reduces from 571 to $522 \, l \, kg^{-1}$ DM.

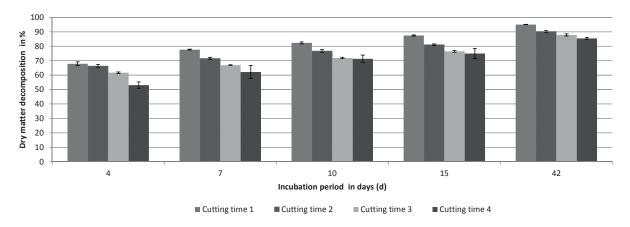
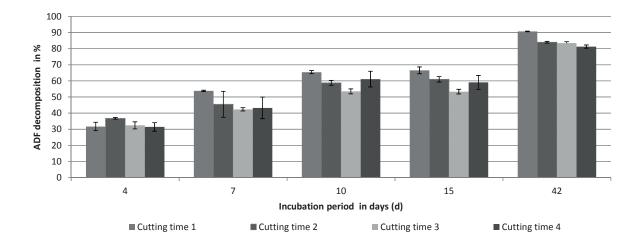


Figure 2: Content of raw nutrients and fibre substances of grass silage (mean ± standard deviation of three replicates

In Figure 3 the degree of lignocellulose (as ADF) degradation is shown. With increasing incubation time, the degree of degradation increases after 42 days to a maximum 90.67% with cut 1 material. In comparison, subsequent cuts achieve lesser degrees of ADF degradation between 81.27 and 83.96%, thus differing only slightly. The greater difference in degradation rate between cut 1 and subsequent cuts can be attributed to the reduced ADF content of these silages compared with the material from cut 1 (Table 1).



Figures 3: ADF decomposition in relation to incubation period and cutting time (mean ± standard deviation of three replicates)

In Figure 4 is presented the degree of dry matter degradation and methane yield of the silages from the different cuts with an incubation time of 42 days. The methane yield from the first cut is 291 l kg⁻¹DM. The methane yields from cuts 2 to 4 are shown as 19, 3 and 25 l kg⁻¹ DM less than from cut 1 (Figure 4). Despite the reduced dry matter degradation of 7.21 and 2.38% by cut 3 compared to cuts 1 and 2, the methane yield is, with 288 l kg⁻¹ DM, higher than that from cut 2 with a methane yield of 272 l kg⁻¹ DM and a dry matter degradation degree of 90.26%. The cut 4 substrate, at 85.46%, shows the least degree of dry matter degradation. This is also reflected in the reduced methane yield of 266 l kg⁻¹DM. The content of structural substances is highest with this cut and influences the dry matter degradation and, with that, the methane yield of this silage.

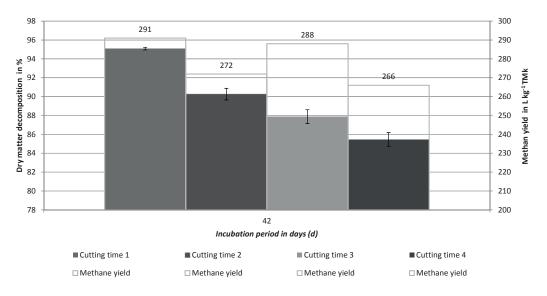


Figure 4: Dry matter decomposition and methan yield after 42 days of incubation in relation to cutting time

Table 2 presents the degree of degradation of the structural substances and lignocellulose after a period of 42 days in the biogas process in relation to the cut the material comes from. It shows clearly that the digestibility of all fractions reduces with the subsequent regrowths. The biggest differences in the degrees of degradation are to be seen in cut 1 compared with subsequent cuts. Cut 1, with a dry matter degree of degradation of 95.09%, achieves a biogas yield of 571 l kg⁻¹ DM. The dry matter degree of degradation of cut 2 is less than cut 1 by an absolute 4.83%. The biogas yield is 8.24% less than that from cut 1. Cut 3 has, in comparison to cut 2, a 2.38% absolute lesser degree of dry matter degradation. The biogas yield is 553 l kg⁻¹ DM and is 29 l kg⁻¹ DM higher than that from cut 2. The reason for this can be the differing chemical compositions of the silages (Table 1).

Parameter	Unit	Cutting time 1 (CT 1)	Cutting time 2 (CT 2)	Cutting time 3 (CT 3)	Cutting time 4 (CT 4)	F-Value
DM degradation	%	95.09 ^a ± 0.14	90.26 ± 0.62	87.88 ± 0.73	85.46 ± 0.73	138.92**
ADF degradation	%	$90.67^{a} \pm 0.21$	83.96 ^b ± 0.55	$83.38^{b} \pm 0.89$	81.27 ^b ± 1.00	92.55**
NDF degradation	%	91.72 ± 0.28	85.06 ± 0.85	84.01 ± 0.86	82.69 ± 0.87	84.58**
Hemicellulose degradation ¹⁾	%	93.23 ± 0.51	86.77 ± 1.75	84.72 ± 0.83	84.44 ± 0.76	44.10**
Biogaspotential	∣kg ⁻¹ DM _c	571 ± 2.15	524 ± 5.23	553 ± 3.47	522 ± 4.98	5.82*
Methanpotential	l kg⁻¹ DM _c	291 ± 1.27	272 ± 3.98	288 ± 2.52	266 ± 2.68	5.54*

Table 2: Anaerobic degradation of lignocellulose und fibre components as well as biogas yield and methan yield of grass silage in relation to cutting time (mean \pm standard deviation of three replicates)

¹⁾ Calculated as difference, *p<0,05. **p<0,01: Comment: Values with different suspended letters (a,b) are significantly different at the 5%-level.

The results of the single factor variance analysis (Anova, Microsoft Excel) with a significance level of 0.05 and 0.01 are also presented in table 2. The cut number has a significant effect (F (3.8) = 92.55, p < 0.01) on the degree of degradation of ADF on incubation day 42, as well as a significant influence on the degree of degradation of the dry matter (F (3.8) = 138.92, p < 0.01) and of NDF (F (3.8) = 84.58, p > 0.01) on incubation day 42. With the later cuts, there is a significant reduction in the degree of degradation of DM, ADF and NDF. The time of cut has also a significant influence on biogas yield (F (2.9) = 0.63, p < 0.05) and methane yield (F (2.9) = 0.66, p < 0.05) on incubation day 42. The ADF degree of degradation from cuts 2 to 4 is significantly higher than the ADF degree of degradation of cut 1, although ADF content in the cut 1 silage is less than that from cuts 2 to 4. Although NDF content in the starting material does not differ greatly, the NDF degree of degradation after 42 days of incubation in cut 4 silage is 9.03% less than that of cut 1 silage. A reason for this could be the crude fibre content, including lignin, which differs by 5.91%.

The anaerobic degradation of grass silage in the biogas process is influenced by degree of maturity and vegetative phase of the biomass at harvest. The further the degree of maturity has progressed, the slower the substrate is able to be utilised in the biogas process. The degradation degree of DM correlates very negatively (correlation number K = 0.98) with the DM content of the silage. The NDF content of the original material correlates slightly negatively (correlation number K = 0.14) with the degree of NDF degradation. Whereas the ADF content of the original material correlates very negatively (correlation number K = 0.97) with the ADF degree of degradation.

KöLLMEIER et al. (2012) found a relationship between pH value and temperature. The higher the pH and the lower the temperature, the higher the resultant degree of degradation. In contrast to the trials reported here, the original material (a mix of straw and hay) was milled to 4 mm length. The degree of degradation hereby related only to the hydrolysis phase and therefore lay within a range of < 34%. The trial reported here observes an incubation period of 42 days and is therefore able to show a substantially higher degree of degradation. Furthermore, KöLLMEIER et al. (2012) demonstrated a higher cellulose degradation degree with increasing pH values. The batch trials evaluated in this work were carried out within a pH range from 7.3 to 7.6. Conclusions regarding influence of pH on degradation degree could not be demonstrated. Because in practice there are few renewable energy biogas production plants where separation of hydrolysis from methane production is implemented structurally and technologically, these trials concentrated on a single-stage process. The results from PERČULIJA et al. (2012) show that, with increasing maturity, the degree of dry matter degradation falls.

In the trials reported here, this is also demonstrated for different incubation periods. Thereby, the anaerobic degradation of crop biomass is dependent on the degree of maturity of the plants and can be influenced by a targeted time of harvest. MARÍN-PÉREZ et al. (2011) could determine in batch trials that higher degradation rates are achieved, the higher the pH value and the lower the temperature in the trials. Investigated here was the hydrolysis phase. A result giving degradation rates in the total process was not achieved. Because a pH value dependency and a temperature influence were not observed in the trial reported here, although degradation rates on incubation day are available, a direct comparison is possible only to a limited extent.

Conclusions

With the methods described for investigating degree of degradation of structural substances and the lignocellulose fraction of grass silages in the biogas process, it is possible to examine silages that come from commercial farms without beforehand having to subject the material to preparation such as drying or milling. The wilting of harvested substrate prevents production of fermentation effluent which, because of its high content of readily available carbohydrates, represents a loss source, reflecting the situation in practice. In order to be able to better compare the results of the degrees of degradation of the different parameters, starting material should be wilted to a uniform dry matter content in subsequent investigations. Especially in the summer months, rising temperatures in the fermenters are repeatedly observed in agricultural biogas plants. Should this have an influence on degree of degradability, this could be demonstrated by the in-sacco-batch method.

The study reported here shows that the in-sacco-batch method is suitable for characterisation of energy crop plants with regard to degradation of the lignocellulose fraction and structural substances. It also demonstrates that estimates of biogas potential, whilst delivering a statement on expected yield, leave unconsidered the degree of material exploitation involved. It may be assumed that use of ensiling additives to optimise the biogas production process can influence the degree of silage exploitation. Additionally, the In- Sacco-Batch method is an instrument for demonstrating the efficiency of additives as already demonstrated by DEMMIG et al. (2011) in earlier studies. Moreover, the trials show that the yield of biogas, as well as the amount of structural substance and lignocellulose degradation, is dependent on the cutting/harvesting time. The in-sacco-batch method is, in comparison to a biogas potential investigation, substantially more complex. As basic research into the biogas process, such investigation methods should not be disregarded on cost grounds. The efficiency of a process can only be qualified and quantified through precise investigations.

References

- Banemann, D.; Nelles, M.; Mathies, E.; Ramhold, D. (2007): Adaptierung der In-Sacco-Methode für die Untersuchungen des Abbauverhaltens von Energiepflanzen in Biogas-Batch-Versuchen. In: Vorträge zum Generalthema: "Futtermitteluntersuchung und -bewertung – Grundlage für die Lebensmittelqualität", VDLUFA-Schriftenreihe Bd. 63, S. 277–284
- Demmig, C.; Höppner, F.; Banemann, D.; Nelles, M. (2011): Untersuchungen zur Abbaukinetik von Grassilagen in In-Sacco-Batch-Versuchen. In: Tagungsband zum 5. Rostocker Bioenergieforum, Universität Rostock, 02./03.11.2011, Rostock, S. 295–301
- Jung, H.G.; Deetz, D.A. (1993): Cell Wall Lignification and Degradability. In: Forage Cell Wall Structure and Digestibility, H.G. Jung, D.R. Buxton, R.D. Hatfield, J. Ralph (Ed.), ASA, CSSA, SSSA, Madison, WI., p. 315-346, doi:10.2134/1993.foragecellwall.c13

- Köllmeier, T.; Zverlov, V.V.; Schwarz, W.H, D. (2012): Mikrobiologie der Hydrolyse von Pflanzenfasern in Biogasanlagen.
 In: BiogasPOTENZIALE erkennen, erforschen, erwirtschaften, 2. Öffentliches Symposium des BCN, 29.10.2012, IHK Potsdam, Bornimer Agratechnische Berichte Heft 79, S. 99-113
- Marín-Pérez, C.; Dandikas, V; Koch, K.; Lebuhn, M.; Gronauer, A. (2011): Einflussfaktoren auf die Hydrolyse eines Stroh- und Heumixes. In: Tagungsband zum 5. Rostocker Bioenergieforum, Universität Rostock, 02./03.11.2011, Rostock, S. 303–311
- Marín-Pérez, C.; Vasilis, D.; Koch, K.; Lebuhn, M.; Gronauer, A. (2012): The effect of cellulolytic microorganisms on the degradation of the solid residual fraction of straw and hay. International Conference of Agricultural Engineering, Proceedings, CIGR Ageng, 08.–12.07.2012, Valencia, Spain
- Marín-Pérez, C.; Weber, D. (2012): Möglichkeiten und Grenzen zweiphasiger Systeme zum Aufschluss lignocellulosereicher Substrate durch biologische Behandlung. Bornimer Agratechnische Berichte 79, S. 9–21
- Perčulija, G.; Vranić, M.; Kutnjak, H.; Leto, J. (2011): In sacco Dry Matter and NDF Degradability Grass Silage Harvested at Three Stages of Maturity. Bulletin UASVM Animal Science and Biotechnologies 68(1–2), pp. 58–62
- Schwarz, W.H. (2004): Cellulose Struktur ohne Ende. Naturwissenschaftliche Rundschau 8, S. 443-445
- Südekum, K. H. (2005): Möglichkeiten und Grenzen einer Standardisierung der In Situ-Methodik zur Schätzung des ruminalen Nährstoffabbaus. Übersichten zur Tierernährung (33)2, S. 71–86
- Teller, E.; Vanbelle, M. (1990): Dégradabilité in sacco d'ensilages d'herbe et contamination bactérienne des résidus. (In sacco degradability of grass silage and bacterial contamination of residues), Reproduction, Nutrition, Development 30, pp. 155–156
- Verein Deutscher Ingenieure VDI e.V. (2004): VDI 4630 Vergärung organischer Stoffe. Berlin, Beuth Verlag
- Weiß, S.; Zankel, A.; Lebuhn, M.; Petrak, S.; Smitsch, W.; Guebitz, G.M. (2011): Investigation of mircroorganisms colonising activated zeolites during anaerobic. Bioresource Technology 102, pp. 4353–4359
- Weißbach, F.; Strubelt, C. (2008): Die Korrektur des Trockensubstanzgehaltes von Grassilagen als Substrat für Biogasanlagen. Landtechnik 63(4), S. 210–2011, http://dx.doi.org/10.15150/lt.2008.818
- Wroblewitz, S.; Hüther, L.; Berk, A.; Lebzien, P.; Kluth, H.; Manderscheid, R.; Erbs, M.; Weigel, H-J.; Wätzig, H.; Dänicke, S. (2014): The impact of free air carbon dioxide enrichment (FACE) on nutrient digestibility of maize grains in pigs and broilerchickens and on ruminal in sacco degradability. Animal Feed Science and Technology 196, pp. 128–138
- Zverlov, V.V.; Hiegl, W.; Köck, D.E.; Kellermann, J.; Köllmeier, T.; Schwarz, W.H. (2010): Hydrolytic bacteria in mesophilic and thermophilic degradation of plant biomass. Engineering Life Science 10, pp. 528–536

Authors

Dipl.-Ing. Claudia Demmig is Ph.D. student at the University of Rostock on the Faculty of Agricultural and Environmental Sciences, Department Waste Management and Material Flow and works for the waste management company Rendsburg-Eckernförde as plant manager, Borgstedtfelde 15, 24794 Borgstedt, E-Mail: c.demmig@awr.de.

Dr. Frank Höppner is a scientific assistant at the Julius-Kühn-Institut, Federal Research Centre for Cultivated Plants, Institute for Crop and Soil Science, Bundesalle 50, 38116 Braunschweig.

Dipl.-Ing. Dietmar Ramhold is managing director of the ISF Schaumann Forschung and provides research in the fields of animal nutrition and process optimization and control of agricultural biogas plants.

Prof. Dr. mont. Michael Nelles is head of the Department Waste Management and Material Flow at the Faculty of Agricultural an Environmental Science, University of Rostock and managing director of the DBFZ Deutsches Biomasseforschungszentrum, Justus-von-Liebig-Weg 6, 18059 Rostock.