Ethanol production from maize silage as lignocellulosic biomass in anaerobically digested and wet-oxidized manure

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Abstract

In this communication, pretreatment of the anaerobically digested (AD) manure and the application of the pretreated AD manure as liquid medium for the simultaneous saccharification and fermentation (SSF) were described. Furthermore, fermentation of pretreated maize silage and wheat straw was investigated using 2 l bioreactors. Wet oxidation performed for 20 min at 121 °C was found as the most suitable pretreatment conditions for AD manure. High ammonia concentration and significant amount of macro- and micro-nutrients in the AD manure had a positive influence on the ethanol fermentation. No extra nitrogen source was needed in the fermentation broth. It was shown that the AD manure could successfully substitute process water in SSF of pretreated lignocellulosic fibres. Theoretical ethanol yields of 82% were achieved, giving 30.8 kg ethanol per 100 kg dry mass of maize silage.

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1. Introduction

Ethanol obtained from biomass has potential to become an important sustainable transportation fuel in the near future. Ethanol can be produced via fermentation from any simple sugar or starchy material, and an interesting feedstock is lignocellulosic biomass (e.g. from agricultural wastes and forest residues) which constitutes approximately 50% of all land produced biomass. Implementation of efficient ethanol production from lignocellulose can be a breakthrough in the fuel market (Lee, 1997; Chum and Overend, 2001; Wyman, 2003; Kim and Dale, 2004; Demirbas, 2005). Optimisation of the existing processes needed to convert lignocellulose into bioethanol (physical/chemical pre-treatment, enzymatic hydrolysis, and fermentation) is necessary and has been investigated by several researchers (Stenberg et al., 1998; Varga et al., 2003; Lloyd and Wyman, 2005). The global prospective and potential of bioethanol production from lignocellulosic biomass are discussed in several recent papers given, e.g. by Lee (1997), Zaldivar et al. (2001), Chum and Overend (2001), Wyman (2003), Kim and Dale (2004), Demirbas (2005) and Ballesteros et al. (2006).

1.1. Anaerobically digested (AD) manure as water and nutrient source

Water is a basis for all biotechnological processes and in bioethanol fermentation a great amount is required. Price of process water is estimated to be around 0.16 EUR/ton (Wingren et al., 2003). According to economical evaluations of ethanol production from lignocellulosic materials the second most costly operation in the production process
is the SSF (simultaneous saccharification and fermentation) step, accounting for approximately 28% of the total (Mielenz, 2001; Wingren et al., 2003). One way to minimize the cost of SSF is by minimizing the addition of the water, nutrients, and elements during fermentation. The AD manure is a great source of nitrogen which is essential in yeast fermentations and in addition it contains satisfactory amount of other nutrients which can improve yeast fermentation such as potassium, calcium, magnesium, iron, manganese, zinc, or copper (Thomsen et al., 2005a).

Usually, urea or yeast extract is added to the fermenters as a nitrogen source. Cost of urea is around 160 EUR/ton (Chemical Industry Intelligence – www.icispricing.com). On the other hand, digestate is considered a waste product from biogas production, and as a result it does not have an established market value. The only cost is the transport price, which could be reduced to minimum, if the biogas and bioethanol processes were integrated – or at least located in the same area.

During typical anaerobic digestion only 50% of the feedstock is converted into biogas. Lignocellulosic material pass through the biogas reactor almost unconverted. The remaining liquid after anaerobic digestion is used as an agricultural fertilizer. However, it may contain too many macro-nutrients, which – if not consumed by the plants – can pollute ground waters (Holm-Nielsen et al., 1997). Additionally, the anaerobic digestion process does not kill all of the pathogens. Pretreatment of AD manure, e.g., by wet-oxidation, would make the lignocellulosic fraction accessible to microbial conversion, and it would kill unwanted microorganisms in the manure. Results concerning an optimization of the pretreatment process of the AD manure were presented by Thomsen et al. (2005b).

The lignocellulosic sugars are utilized during fermentation whereas lignin and other non-fermentable compounds remain in the fermentation broth, which makes the ethanol stillage suitable as a carbon-rich fertiliser. Combining the biogas and bioethanol processes allows utilization of all biomass components. The nutrients not consumed during livestock or energy production will return to soil as a biofertilizer. The stillage will contain less nutrients and will, as a result, not be harmful for the environment, thus it would be much more suitable fertilizer for soils. The anaerobic fermentation (both biogas and bioethanol) causes destruction of solid matter and a reduction of viscosity of the liquid phase, which make easier to apply as a fertiliser. When it is used on the farmland it mixes and diffuses easier with the soil, compared to raw manure. Lower pH value after bioethanol fermentation compared to AD manure will prevent ammonia evaporation; consequently loss of the nitrogen will be declined.

1.2. Maize silage as a carbon source

Thomsen (2005) describes the most common agricultural by-products, which can be used as a feedstock for ethanol production, and Weiland (2006) describes suitable crops for biogas production. Maize can be extensively used in biogas and bioethanol production (pre-stored as maize silage for all year round). Maize silage is the whole crop of maize plant harvested about 10–20 cm above the ground. It consists of the stem, leaves and cobs (spinals and grains). The whole harvested plants are cut into small pieces (about 1.5 cm) and ensiled anaerobically. In a correct ensilage lactic acid bacteria dominate the fermentation process. Best harvest time for proper ensiling is when the whole crop has a total solid content of approximately 30–33% dry matter, optimal for compaction. In Denmark nearly all grown maize is harvested for maize silage production.

The maize used for silage is mainly composed of hard degradable lignocellulosic material (stem, leaves) and in smaller amount of starchy material (grain). Big variations for different maize silages occur in starch content, cob development, dry matter content, dependent on weather and soil conditions, time of harvesting, climate, and ensiling period. At present maize silage is mainly used as animal fodder; however, it has a great potential to become a highly efficient material for bioenergy production.

The main goal of the research described in this paper is to investigate the features of pretreated maize silage as an efficient carbon source for microorganisms, as well as to examine whether a mixture of pretreated AD manure and solids from pretreated maize silage could be efficient as nutrient and carbon source for the ethanol fermentation process. The influence of wet-oxidation on maize silage is investigated. The hydrolysis yields for non-treated and pretreated maize silage, wheat straw, and AD manure, and as well as ethanol yields are determined. The comparison between the three different types of biomass (maize silage–energy crop, wheat straw–agricultural by-product, and AD manure–agricultural–animal by-product) is presented and described.

2. Methods

2.1. Raw materials

The anaerobically digested pig manure was obtained from the Snertinge Biogas Plant (Denmark). Before pretreatment the material was kept at −5 °C to avoid uncontrolled fermentation. The dry matter (DM) content of the manure was 51.1%.

The maize silage was delivered from Aalborg University Esbjerg. The maize was harvested a few months before experiments and collected from a diary farm. DM was approximately 25%.

The raw and pretreated wheat straw was delivered from a pilot scale pretreatment plant: IBUS, Integrated Biomass Utilization System located at Fynsværket in Odense (Thomsen et al., 2006). Table 1 shows the composition of sugars, proteins, and fatty acids of raw maize silage and straw.
Maize silage contains more valuable organic compounds per 1 kg of dry matter than wheat straw. Therefore, higher ethanol yield should be expected.

2.2. Pretreatment

Wet-oxidation pre-treatment was conducted in a loop reactor designed and constructed at Risø National Laboratory. The volume of the reactor is 2 l, with a working volume of 1 l. The metal autoclave was mounted on the rack make possible to control the temperature by immersing the reactor in a thermostatic bath. The detailed description of the autoclave is presented by Bjerre and Skammelsen Schmidt (1997).

The AD manure was wet-oxidized using 12 bar O₂ for 20 min at 121 °C and applied as a liquid nutrient rich medium for fermentation purpose. The ethanol yield for wheat straw in pretreated AD manure at different conditions is shown in Table 2.

The maize silage was pretreated for 15 min at optimal conditions for maize (corn) stover according to Varga et al. (2003): 195 °C, 2 g/l Na₂CO₃, 12 bar O₂. The wet material of non-milled maize silage (160 g wet material, 25% DM) was mixed with 11 of water and Na₂CO₃. After cooling down to about 35 °C, the pretreated material was separated by filtration into solid (filter cake) containing the cellulose sugars and a liquid fraction (hydrolysate) containing hemicellulose sugars and fermentation inhibitors. For further use in the fermentation process only the cellulose-rich solid fraction (98% DM) was used. It has been shown that after wet-oxidation process the cellulose is recovered in the solid phase (Thomsen et al., 2006). Within this study only cellulose was important because Saccharomyces cerevisiae is unable to ferment hemicellulose sugars.

Pretreated wheat straw was obtained from the pilot plant at Fynsværket in Odense (IBUS, Integrated Biomass Utilization System). The pretreatment conditions were as follows: temperature 195 °C, straw flow – 50 l/h, water flow – 250 l/h, with addition of 0.5% H₂O₂ (Thygesen et al., 2004; Thomsen et al., 2005a). The pretreated wheat straw was used as a carbon source in the optimisation of pretreatment of AD manure (Thomsen et al., 2005b).

2.3. Drying

The solids of wet-oxidized maize were dried and stored at 20 °C and 65% humidity. Drying was necessary in order to be able to store the biomass without biological or chemical decomposition, which could negatively influence on the laboratory experiments. In the full scale process, this step has to be avoided in order not to increase the process energy demand. According to Bjerre and Skammelsen Schmidt (1997), the chemical structure of the solid fractions stored at these conditions should be stable for up to 10 years. The total solid value of stored material is in the range of 95–98% DM (dry matter).

2.4. Strong acid hydrolysis

Strong acid hydrolysis was performed to characterise the composition of the dried raw materials and the solid fractions of wet-oxidized maize silage, wet-oxidized AD manure (121 °C, 12 bar O₂, 20 min), and pretreated wheat straw. The first hydrolysis step was performed at 30 °C for 60 min with 1.5 ml of H₂SO₄ (72 %) for 0.16 g DM. Then 42 ml water was added and the second step was performed at 121 °C for 60 min. The acid hydrolyzate was filtered and the glucose, xylose, and arabinose were quantified by HPLC (Aminex HPX-87H column (Biorad)). Klason lignin was calculated as the ash free residue after hydrolysis. The ash content was determined by heating for 3 h in oven at 550 °C.

2.5. Cellulose convertibility

The convertibility of cellulose to glucose with cellulase enzymes (Cellubrix, produced by Novozymes) was determined using enzymatic hydrolysis for 24 h, at 50 °C and pH 4.8. The enzyme loading was 30 FPU/g DM (Filter Paper Unit per gram dry matter). This analysis was performed on non-treated and pretreated maize silage, wet-oxidized AD manure (solid part), and pretreated wheat straw. The hydrolysis yield is given as the percentage of glucose produced based on the total amount of cellulose present in the substrate.

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Table 1

<table>
<thead>
<tr>
<th>Raw material</th>
<th>Cellulose (g/100 g DM)</th>
<th>Hemicellulose (g/100 g DM)</th>
<th>Lignin (g/100 g DM)</th>
<th>Proteins a (g/100 g DM)</th>
<th>Fatty acids a (g/100 g DM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maize silage</td>
<td>51.7</td>
<td>19.5</td>
<td>16.6</td>
<td>7.9</td>
<td>1.4</td>
</tr>
<tr>
<td>Wheat straw</td>
<td>33.9</td>
<td>23.0</td>
<td>19.1</td>
<td>3.3</td>
<td>0.5</td>
</tr>
</tbody>
</table>

a Møller (2005).

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Table 2

<table>
<thead>
<tr>
<th>Pretreatment conditions for AD manure</th>
<th>Ethanol yield (g/100 g TS_straw)</th>
</tr>
</thead>
<tbody>
<tr>
<td>121 °C, 20 min, no oxygen</td>
<td>20.7</td>
</tr>
<tr>
<td>121 °C, 20 min, 12 bar O₂</td>
<td>20.9</td>
</tr>
<tr>
<td>160 °C, 15 min, no oxygen</td>
<td>20.9</td>
</tr>
<tr>
<td>160 °C, 15 min, 12 bar O₂</td>
<td>20.6</td>
</tr>
<tr>
<td>195 °C, 10 min, no oxygen</td>
<td>21.2</td>
</tr>
<tr>
<td>195 °C, 10 min, 12 bar O₂</td>
<td>21.9</td>
</tr>
</tbody>
</table>

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2.6. Ammonia

The concentration of ammonia was measured in samples collected from the fermentation process performed in 21 fermenters to investigate the nitrogen consumption during SSF. The quantitative analysis was performed with the HPLC using the Hamilton PRPX211 column.

2.7. Fermentation

The wet-oxidised maize silage was milled in a universal grinder (type: MF 10 basic microfine having 1 mm sieve, produced by IKA) into powder before application in the SSF process. The microorganisms applied for the SSF was typical baker’s yeast (S. cerevisiae). As the first step, a prehydrolysis was carried out for approximately 24 h at 50 °C with addition of 15 FPU/g DM enzyme solution Cellubrix (Novozymes). After 24 h the temperature was lowered to 30 °C and new enzyme loading (20 FPU/g DM) and baker’s yeast were added, the simultaneous saccharification and fermentation was continued for approximately 6 days.

Fermentation in shake flasks was carried out with 6 g DM of pretreated wheat straw added pretreated AD manure to obtain total mass of 100 g. The process was conducted in 250 ml flasks. Mixing was assured by magnetic stirrer. The pH value (4.76–4.84) was adjusted by addition of 0.01 M H2SO4 (5× or 10× dilution). After centrifugation for 10 min at 4000 rpm, the samples were filtered into HPLC vials and analysed by HPLC (column BioRad Aminex HPX87H).

The fermentation of pretreated maize silage in pretreated AD manure was carried out in 2-l fermenters (MINIFORS, Infors AG, Switzerland). The working volume was equal to 1.5 l. The mass of pretreated maize silage supplied to the reactor was equal to 68 g DM (which results from: mass of glucan in feedstock, g/100 g DM; TH is the theoretical ethanol yield, g/l. The relative ethanol yield is expressed as percentage of theoretical ethanol yield

\[
THP = \frac{\text{EtOH}_{\text{HPLC}}}{\text{TH}} \times 100\% \quad (1)
\]

where THP is the percentage of theoretical ethanol yield, %; EtOH_{HPLC} is the ethanol concentration determined by HPLC, g/l; TH is the theoretical ethanol yield, g/l.

Based on HPLC-analysis of the samples the practical ethanol yield (kg/100 kgDM) was calculated from obtained percentage of the theoretical one.

2.10. Hydrolysis yield

The relative hydrolysis yield of glucose was calculated from:

\[
Y_{\text{glucose}} = \frac{m_c}{m_g} \times 100\% \quad (2)
\]

where \(m_c\) is the mass of cellulose converted by cellulase in enzymatic convertibility test, g/100 g DM; \(m_g\) is the mass of glucan in feedstock, g/100 g DM.

3. Results and discussion

3.1. SSF of pretreated wheat straw in pretreated AD manure

Optimum pretreatment conditions for AD manure to be used in bioethanol production were chosen as 20 min at 121 °C with addition of oxygen (12 bar) according to Thomsen et al. (2005b). Due to the fact that the different pre-treatment conditions resulted in very similar ethanol yields (Table 2), it was decided to select the pretreatment conditions with the lowest energy input. Temperatures below 121 °C were not tested, because of the sterilization requirement.
The only distinction between AD manure pretreated with and without oxygen was a much stronger and unpleasant odour in samples where no oxygen was added. During wet-oxidation, a reduction of aromatic compounds occurred and lighter colour was observed in these samples compared to samples pretreated without oxygen, indicating degradation of aromatic compounds.

Influence of the AD manure on the fermentation process was investigated. SSF of pretreated wheat straw fibres in AD manure was compared to SSF carried out in water (with addition of urea as a nitrogen source) (results not shown). No difference in obtained ethanol yield was observed. This indicates that AD manure can be used as a water and nutrient source in ethanol fermentation from lignocellulosic materials.

Liao et al. (2004) and Wen et al. (2004) investigated dilute acid hydrolysis of manure lignocellulosics into fermentable sugars. Achieving glucose yield of 11.32 g/100 g manure (corresponding to about 40% cellulose conversion) they concluded that the animal manure can be converted into a valuable product such as ethanol and other chemicals.

In this project the anaerobically digested animal manure was used, therefore the readily available carbon had already been consumed by the methanogenic microorganisms, and no significant increase in ethanol yield was expected. Application of the AD manure as a water and mineral source in the ethanol-fermentation can reduce cost in the SSF step as well as diminish the input of fossil fuels (urea) in the process.

3.2. Cellulose concentration

Glucan in the form of glucose is the most important polymer for yeast fermentation. Comparison of glucan content in different raw and pretreated materials is shown in Table 3. Maize silage has significantly higher glucan content than wheat straw, even the non-treated materials contain more cellulose than pretreated straw. Maize silage having higher glucan content could as a result be a promising feedstock for bioethanol production compared to the traditional raw materials. The theoretical yield was calculated based on the total amount of glucan in the biomass. The solids in pretreated AD manure did not contain large amount of glucan, and as a result the application of wet-oxidized AD manure would not directly influence the ethanol yield. AD manure should be considered as a nutrient source.

The pretreated maize silage gives almost 40% higher theoretical ethanol yield than pretreated wheat straw, and based on this, the maize silage from maize crop would be the best feedstock for ethanol production. However, maize silage is considered as an energy rich animal feed, whereas the wheat straw is regarded as an agriculture by-product and consequently has a lower price, which means that bioethanol can be produced at lower costs. On the other hand, the higher price of maize silage could be compensated by the higher bioethanol yield.

Arabian and xylan are the main composites of hemicellulose, and a major part of these sugars are solubilized and extracted into the filtrate during wet-oxidation. Fig. 1 presents the concentration of hemicellulose sugars in raw and pretreated materials (fibre fractions). The amount of hemicellulose sugars in the fibres of straw and maize silage decrease during pre-treatment: 30% of the xylan was extracted from wheat straw fibres and 70% from maize silage fibres. The extraction of hemicellulose from the fibers is considered as an important factor in the improvement of the enzymatic convertibility of the cellulose fibres. The high hemicellulose extraction implies that maize silage can easily be fractionated and converted to bioethanol. In both cases all the arabinan was transferred to the liquid. The pretreated AD manure contains some amount of xylan and arabinan, which confirms that the degradation of lignocellulosic solids in the AD manure during wet-oxidation is not complete. Strong buffer capacity of digestate keeps constant pH during the hydrolytic reaction. It can slow down the degradation, which is in favour in acidic environment.

The ash content in the manure is significantly higher than in other materials (Fig. 1). This is evidence of high nutrient content (e.g. phosphorus, potassium, nitrogen) in the manure (Thomsen et al., 2005a). The lignin content is similar for all presented biomass materials. Only in the pretreated maize silage it is lower (below 10 g/100 g TS), which indicates that part of the lignin is solubilised during the pretreatment process. Fig. 2 shows changes in sugars, lignin, and ash content after application of wet-oxidation.

In maize silage the concentration of glucan is increased by about 30% compared to the raw material. In ethanol fermentation glucan is the most important component, as it consists of C6 sugars, which can be fermented directly

<table>
<thead>
<tr>
<th>Lignocellulosic biomass</th>
<th>( Y_{\text{ETOH}}^T ) (g/100 g TS)</th>
<th>( Y_{\text{FUMH}}^T ) (g/100 g TS)</th>
<th>( m_c ) (g/100 g TS)</th>
<th>( Y_{\text{glucose}} ) (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Solids from pretreated AD manure</td>
<td>6.3</td>
<td>3.6</td>
<td>2.7</td>
<td>42.9</td>
</tr>
<tr>
<td>Non-treated wheat straw</td>
<td>33.9</td>
<td>19.2</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Pretreated wheat straw</td>
<td>47.5</td>
<td>26.9</td>
<td>28.9</td>
<td>60.8</td>
</tr>
<tr>
<td>Non-treated maize silage</td>
<td>51.7</td>
<td>29.3</td>
<td>32.2</td>
<td>62.3</td>
</tr>
<tr>
<td>Pretreated maize silage (solids)</td>
<td>66.4</td>
<td>37.6</td>
<td>59.8</td>
<td>90.1</td>
</tr>
</tbody>
</table>

Where \( m_c \) is the mass of cellulose converted by cellulase in enzymatic convertibility test.
by commercial yeast strains (S. cerevisiae). More than half of the lignin in maize silage was solubilised during the pretreatment process, which is considered an advantage in the hydrolysis step, where lignin can bind the cellulolytic enzymes, causing a decrease in activity and as a result a decrease in ethanol yield (Martin et al., 2007).

As expected from the efficient extraction of hemicellulose and lignin from the cellulose fibres during pretreatment, the enzymatic conversion of maize silage was very efficient. It should be noted, that the process parameters for pretreatment of the maize silage were not optimized in this study (the process conditions were adopted from experience on pretreatment of corn stover) but still a glucose yield of about 90% was achieved (Table 3). It might be concluded that the maize silage is an excellent material for a bioethanol production. Even without pretreatment a yield of 62% was achieved. However, application of wet-oxidation (at 195°C, 12 bar O₂, 2 g/l Na₂CO₃) significantly increases accessibility of cellulose for enzymatic hydrolysis. The pretreated maize silage has the highest convertibility among the presented biomasses.

3.3. Fermentation of pretreated maize silage

Ethanol fermentation was carried out using the pretreated maize silage (15 min, 195°C, 12 bar O₂, 2 g/l Na₂CO₃), as a carbon source, and pretreated/sterilised AD manure as a liquid medium (20 min at 121°C with 12 bar of oxygen). The experiment was carried out in duplicate. In order to achieve high glucose concentration at the
beginning of the process, a pre-hydrolysis step was applied before the fermentation. After addition of baker’s yeast the glucose content decreased immediately and reached the zero at the end of the process.

Approximately 50% of the theoretical ethanol yield was achieved already after about 4 h of the fermentation. The ethanol yield continued to increase until 40 h of fermentation, at which point the yield had reached about 75%. The average ethanol yield obtained was 82% of the theoretical.

The theoretical ethanol yield was calculated based on the amount of glucose in the fermentation broth. The ethanol yield obtained in SSF was slightly lower, than hydrolysis yield, shown in Table 3 (for pretreated maize silage: 90.1%), this might be due to inhibitory compounds in the fermentation broth formed during the wet-oxidation of the maize silage or AD manure.

Similar fermentation experiments were performed with pretreated wheat straw, and 70% of the theoretical ethanol yield was obtained (Thomsen et al., 2005b). In comparison, this shows that very efficient pretreatment of the maize silage was achieved.

Recently, research groups have presented results with xylose-fermenting *S. cerevisiae* (Sonderegger et al., 2004). Application of such a microorganism would definitely improve ethanol yield from lignocellulosic biomass. In these experiments only the fibre fraction (C-6 sugars) was used, and the calculated yields was based only on glucose sugars, hence the high theoretical yields.

AD manure is not rich in proteins and lipids, as these compounds are converted into biogas during the AD process. However, it contains several valuable elements (Thomsen et al., 2005a) and nitrogen in form of ammonia. These components are important for the fermentation process. It rich in phosphorous, potassium, calcium, and magnesium, it is relatively high in magnesium and iron, but it contains several different microelements such as Mn, Zn, or Cu at the concentration 0.01–0.1%. Compare to maize silage or wheat straw (Møller, 2005) it has much better potential to be valuable nutrients and minerals source for fermentation. Investigation of ammonia changes during fermentations in 2 l fermenters with pretreated maize silage was performed. During SSF the consumption of nitrogen was about 5% of the initial value (Fig. 3). During fermentation with straw, the ammonia usage was slightly higher – up to 10%. Fig. 3 indicates that the yeast utilised part of the nitrogen from the WO-AD manure. The content of ammonia would be sufficient for several recirculation of the WO-AD manure as process water.

To summarize, the bioethanol production from pretreated maize silage, can run efficiently without addition of yeast extract or similar nutrient solutions, when using the pretreated AD manure as process water. The remaining solids and liquid still rich in nutrients can serve as valuable bio-fertilizer and soil conditioner, respectively.

3.4. Estimation of ethanol yield from pretreated maize silage

The theoretical ethanol yield (based on the glucose content in the medium) was calculated for different biomasses and presented in Table 3. For practical applications it is good to know, how much ethanol can be obtained from 100 kg dry pretreated feedstock. During the fermentation in lab-scale 82% of the theoretical yield was obtained. The practical ethanol yield of wet-oxidized maize silage is equal to 30.8 kg of ethanol per 100 kg DM of maize silage. This result shows the significant potential of the maize silage as a substrate for ethanol production. Moreover, it is expected that addition of amylases in the SSF of maize silage can further increase, the ethanol yield due to the starch content in this biomass. However, within this study maize silage was considered only as a lignocellulosic feedstock and only cellulase enzymes were applied. The main

Fig. 3. Ammonia consumption during ethanol-fermentation.
purpose was to investigate, if manure could substitute process water in lignocellulose based ethanol fermentation. For fully estimation of the ethanol potential from maize silage, the influence of amylase-enzymes on ethanol yields should be examined.

4. Conclusions

This study showed that the wet-oxidized anaerobically digested (WO-AD) manure can efficiently serve as nutrient, nitrogen, and water supply in the SSF of lignocellulosic substrates. Moreover, the pretreatment process highly improves the enzymatic accessibility of cellulose in maize silage (from 62% to 90%), and efficient extraction of hemicellulose sugars from the maize silage fibres was achieved during pretreatment. The high content of available glucan resulted in high ethanol yield (82% of theoretical based on glucose content), which corresponds to 30.8 kg of ethanol per 100 kg dry wet-oxidized maize silage.

References


