

PRETREATMENT OF VARIOUS FEEDSTOCKS FOR LACTIC ACID PRODUCTION; DETECTION OF SUGARS, ORGANIC ACIDS AND FURANICS IN LIQUID FRACTIONS

Paulien Harmsen*, Steef Lips, Robert Bakker

Wageningen UR Food and Biobased Research, Bornse Weiland 9, 6708 WG

Wageningen, The Netherlands

**Paulien.Harmsen@wur.nl*

ABSTRACT

Barley straw, sugarcane bagasse and empty fruit bunches were pretreated under acid- and alkaline conditions. Solid phase was separated from the liquid phase and the concentration of dissolved monomeric sugars, organic acids and furanics was determined. Acid hydrolysis yielded monomeric xylose concentrations (max 20 g/l) whereas for alkaline hydrolysis less than 1 g/l was found. Organic acids and furanics were detected with high concentrations when dried empty fruit bunches were used. Acetic acid was found in all samples (max 3 g/l), concentrations were higher under alkaline conditions.

I. INTRODUCTION

The use of lignocellulosic biomass for biofuels has gained much interest the last few decades, but also chemical building blocks from renewable resources form a huge potential and in particular lactic acid for the production of the biodegradable plastic polylactic acid (PLA). To date, lactic acid is mainly produced from starch originating from corn, but a sustainable production process at a scale meeting future demands for PLA requires the use of second generation biomass such as lignocellulosics.

Given the large amount of pretreatment methods and lignocellulosic biomass available, a fast screening method is necessary to set boundaries and to determine suitable conditions for the preparation of enzymatic hydrolysates for fermentation studies at larger scale. In this study three model feedstocks were selected, *i.e.* barley straw, sugarcane bagasse and empty fruit bunch, and pretreated by acid- and alkaline hydrolysis. After pretreatment the solid phase was separated from the liquid phase, and the composition of the fractions was analyzed. The concentration of dissolved monomeric sugars, organic acids and furanics (major inhibitors to enzymatic hydrolysis and fermentation to lactic acid) in the liquid phase is described here.

II. EXPERIMENTAL

Biomass: barley straw was obtained from the Netherlands, sugarcane bagasse from Thailand and empty fruit bunches from Malaysia. The empty fruit bunches were dried at 60 °C before shipment to the Netherlands to prevent microbial degradation. Dry matter of all feedstocks was >90%.

Pretreatment: 0.1L-reactors (not stirred) were filled with biomass and water with a fixed L/S-ratio of 10. Acid hydrolysis was performed at 140° C for 30 min with H₂SO₄-concentrations ranging from 0-8 wt% on dry biomass. Alkaline hydrolysis was performed at 120 °C for 60 min with NaOH-concentrations ranging from 0-12 wt% on dry biomass.

Analysis: monomeric sugars were detected on a HPAEC-PAD (Dionex Corporation, USA), organic acids and furanics on a HPLC (Waters Pharmaceutical Division, Milford, MA, USA).

III. RESULTS AND DISCUSSION

Acid hydrolysis

Results of the acid hydrolysis of barley straw are presented in figure 1. Xylose concentration was maximal 19 g/l while the concentration of glucose and arabinose remained below 3 g/l. Based on the amount of xylose dissolved, hydrolysis of hemicellulose has taken place and expected degradation products include acetic acid and furfural. These components were indeed detected in the hydrolysis liquid as shown in figure 1, but also formic acid and succinic acid were found. HMF, a degradation product of glucose, may further degrade to formic acid and levulinic acid, and levulinic acid can further oxidize to succinic acid. Given the hydrolysis conditions it is possible that glucose degradation (from hemicellulose and/or cellulose) has taken place, but more research is necessary to validate the concentration of formic acid and succinic acid in the sample.

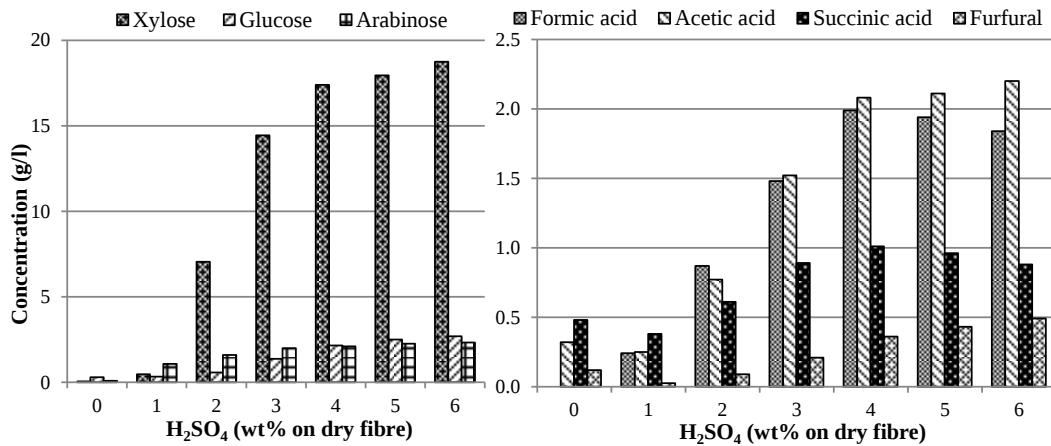


Figure 1. Acid hydrolysis of barley straw. Concentration (g/l) of monomeric sugars (left) and organic acids/furans (right) in the liquid phase. Rhamnose, galactose or mannose was < 1 g/l; lactic acid and levulinic acid were not detected, 5-HMF was < 0.1 g/l.

For sugarcane bagasse the results are given in figure 2. The amount of dissolved sugars is comparable to barley straw with a slightly lower xylose concentration. The amount of acetic acid formed is lower (approximately 1 g/l) and the amount of furfural is comparable; no succinic acid was detected.

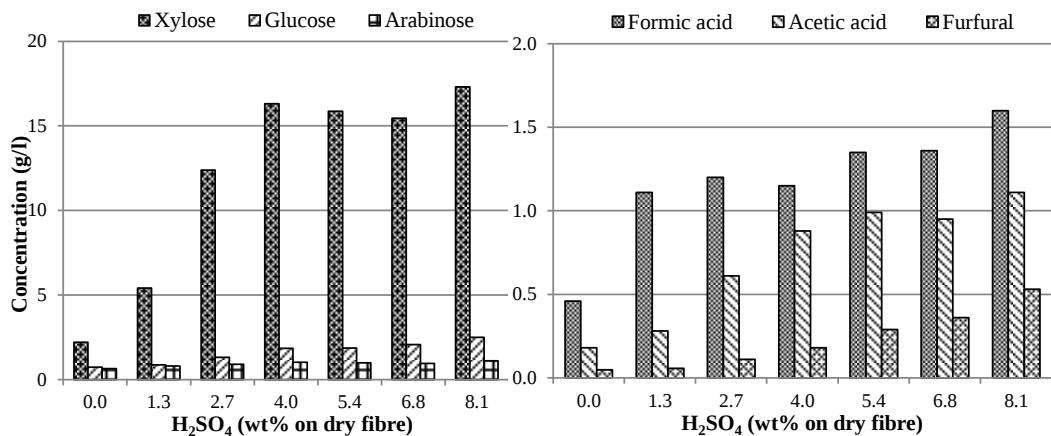


Figure 2. Acid hydrolysis of sugarcane bagasse. Concentration (g/l) of monomeric sugars (left) and organic acids/furans (right) in the liquid phase. Rhamnose, galactose or mannose was < 0.5 g/l; lactic acid, levulinic acid and succinic acid were not detected, HMF was < 0.1 g/l.

Acid hydrolysis of the dried empty fruit bunches resulted in a liquid with a very low xylose content (figure 3); only above 6 wt% H₂SO₄ the xylose concentration was more than 5 g/l. Probably sugar degradation had taken place and this was also confirmed by the amount of organic acids detected. High concentrations of formic acid, acetic acid and succinic acid were found; levulinic acid was also present in the liquid but the concentration decreased with increasing H₂SO₄ content or increasing severity of the pretreatment. It is possible that levulinic acid is further oxidized to succinic acid. There is reason to believe that the drying process of the empty fruit bunches prior to the pretreatment was too severe and caused (partial) burning or degradation. Given the high dry matter content of >90% and the dark brown, black color of the bunches this is most likely; fresh bunches normally contain a lot of water (>70%).

Of the acid pretreatments the combined severity factor R'_0 (Chum, Johnson *et al.* 1990) was calculated. This factor involves the reaction temperature T (°C), reaction time t (min) and pH (to include catalytic effects of applied or released acids) and is described by the following formula:

$$R'_0 = [H^+]R_0 = (10^{-pH}) \left(t * e^{\frac{T-100}{14.75}} \right)$$

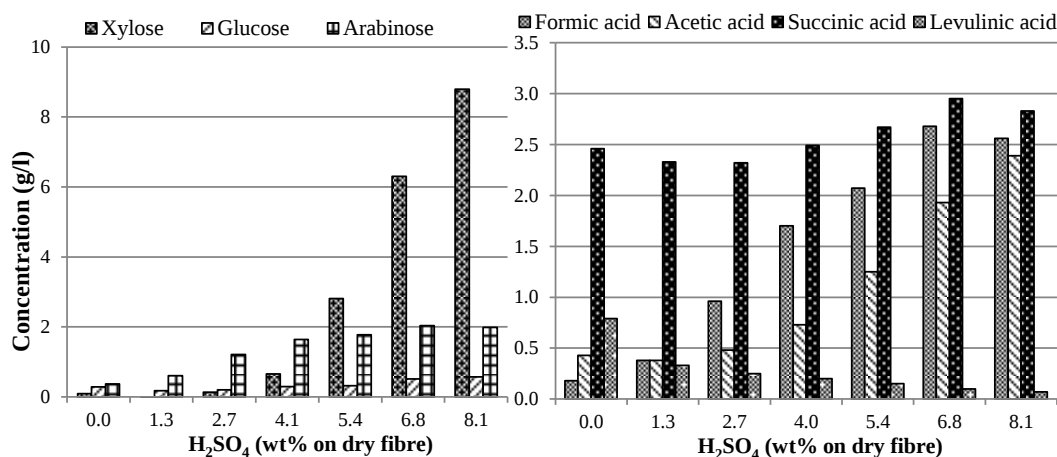


Figure 3. Acid hydrolysis of empty fruit bunches. Concentration (g/l) of monomeric sugars (left) and organic acids/furancs (right) in the liquid phase. Rhamnose, galactose or mannose was < 0.5 g/l; lactic acid, furfural or HMF was < 0.2 g/l.

Figure 4 illustrates the relation of the total sugar content in the liquid phase with the severity of the acid pretreatment. From this figure can be seen that the amount of sugars from the dried empty fruit bunches is lower at the same severity factor compared to barley straw or sugarcane bagasse. Furthermore it is clear that the severity factor should be above 0 to for the pretreatment to have some impact on the feedstock.

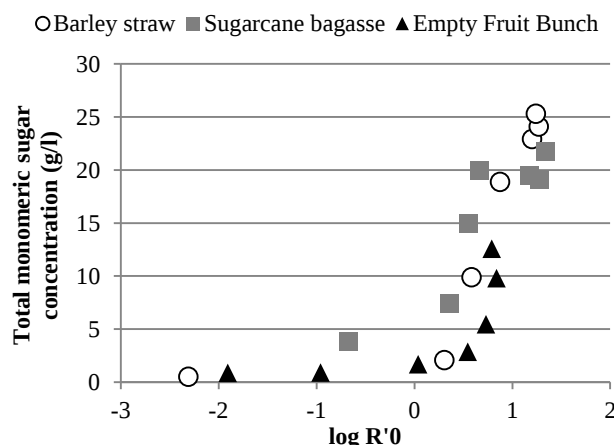


Figure 4. Total sugar concentration (g/l) as function of the combined severity factor $R'0$ for acid-hydrolyzed barley straw, sugarcane bagasse and empty fruit bunch.

Alkaline hydrolysis

For alkaline hydrolysis of barley straw the concentration of dissolved monomeric sugars is almost zero (<0.2 g/l), for sugarcane bagasse it is slightly higher (<2 g/l). It is known that for alkaline hydrolysis the main mode of action is dissolution of lignin, and to a lesser extent hydrolysis of hemicellulose to monomeric sugars. However, it is worthwhile investigating the content of oligomeric sugars as previous work showed that released sugars are mainly present as oligomers. The concentration of organic acids in alkaline pretreatment liquids for barley straw and sugarcane bagasse are presented in figure 5. Typical acid-catalyzed sugar degradation products like 5-HMF and furfural were not detected, and also levulinic acid was not found. The formic acid concentration was lower compared to acid hydrolysis but the acetic acid concentration was comparable or even higher. This illustrates that deacetylation of hemicellulose yielding acetic acid is best catalyzed under alkaline conditions.

Alkaline hydrolysis of dried empty fruit bunches (figure 6) showed different results regarding the formation of organic acids, whereas the concentrations of monomeric sugars was comparable to barley straw and sugarcane bagasse (<0.1 g/l). For the organic acids high concentrations of levulinic acid (maximal 9 g/l) and succinic acid (maximal 3 g/l) were detected, indicating that the drying step prior to pretreatment caused degradation of the bunches.

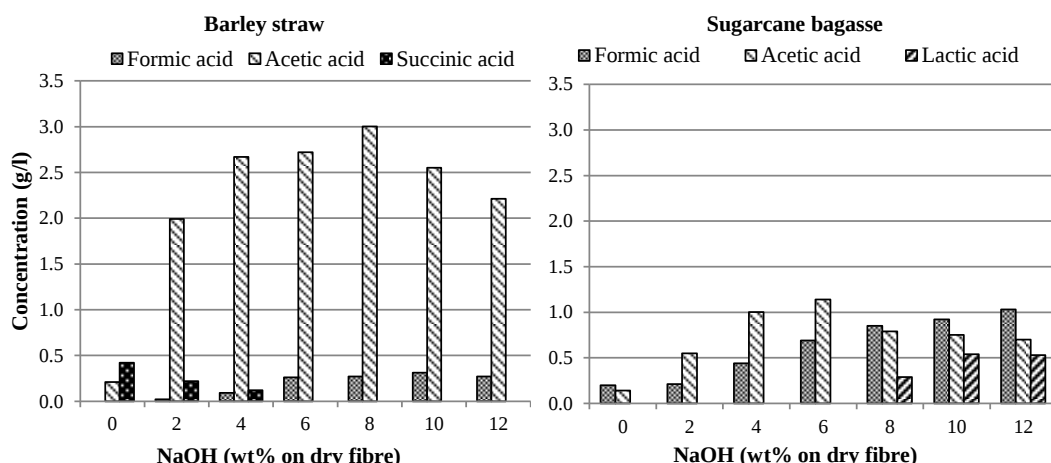


Figure 5. Alkaline hydrolysis of barley straw (left) and sugarcane bagasse (right). Concentration of organic acids (g/l). Levulinic acid, 5-HMF and furfural were not detected.

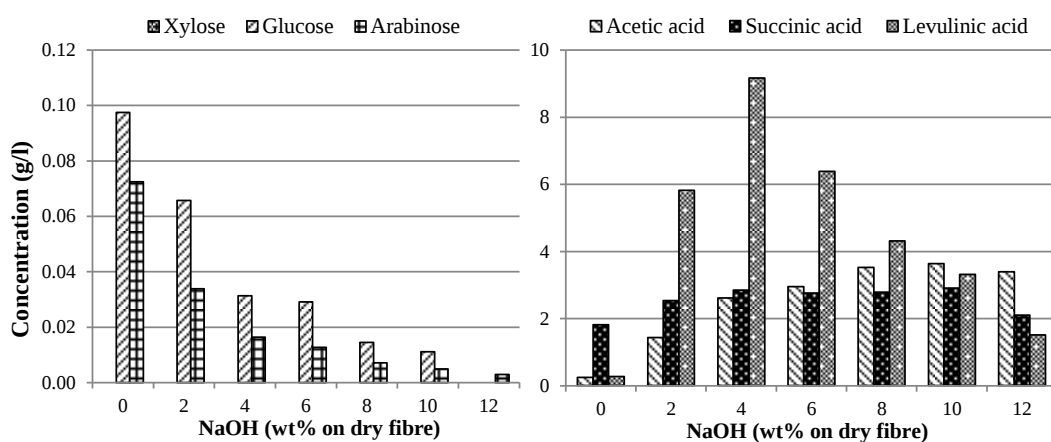


Figure 6. Alkaline hydrolysis of empty fruit bunch. Concentration of monomeric sugars (left) and organic acids (right) in g/l. 5-HMF and furfural were not detected; formic acid was <0.1 g/l.

IV. CONCLUSIONS

Results of biomass pretreatment is dependent on a number of parameters, one of them is the type of biomass. Differences in amounts of cellulose, hemicellulose, lignin and structural differences of those like amount of acetyl groups will influence the results of the pretreatment. Further work will focus on combining the results described here with the composition of the raw biomass and composition of the solid phases obtained after pretreatment. Furthermore, drying of the biomass prior to pretreatment can have a major impact as shown here for the empty fruit bunches. Experiments will be repeated with fresh material as empty fruit bunches remain an interesting source of fermentable sugars for lactic acid fermentation.

Type of pretreatment (acid or alkaline hydrolysis) and the applied conditions (time, temperature, catalyst concentration) have a major effect on the concentration of dissolved components in the liquid phase. Concentrations of xylose, furfural, HMF and hydrolysis yields can also be related to the combined severity factor in case of acid pretreatment. This severity factor can be used to predict the results of experiments done under various conditions.

V. ACKNOWLEDGEMENT

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VI. REFERENCES

Chum, H., D. Johnson, *et al.* (1990). "Pretreatment-Catalyst effects and the combined severity parameter." *Applied Biochemistry and Biotechnology* 1990, 24-25(1), 1-14 (research article)