SUNLIBB

Sustainable Liquid Biofuels from Biomass Biorefining

Grant Agreement no. 251132

Collaborative Project
EU 7th Framework Programme
ENERGY

Project duration: 1st October 2010 – 30th September 2014

Deliverable 1.3
"Insights into predictive value of fodder digestibility for saccharification potential"

Author: **Dr. Matthieu Reymond (INRA, Versailles, France)**

Workpackage: **1**
Workpackage Leader: **Dr. Matthieu Reymond (INRA)**

Due date: **September 2011 (Project Month 12)**
Actual submission date: **September 2011**
Revised version submitted: **December 2014**

Dissemination Level: **PU**

This project has received funding from the European Union's Seventh Programme for research, technological development and demonstration under grant agreement number 251132

SUNLIBB deliverables

Del No: 1.3	**Deliverable Name:** Insights into predictive value of stover digestibility for saccharification potential			
WP: 1	**Lead partner:** 6	**Dissemination level:** PU	**Delivery date (project month):** 12	**Actual delivery date:** 12

Objective:

Digestibility and saccharification are two traits that are related to degradability of biomass. The objective of this deliverable is to get insights into the correlation between these traits. Digestibilty is estimated *in vitro* by enzymatic solubity (using a mix of different enzymes, namely: amyloglucosydase from *Aspergillus niger*, cellulase Onozuka R-10) with a mild pre-treatment (pepsin at 40°C for 24h in acidic buffer HCl 0.1N). NIRS equations have been previously developed and transferred to Partner 6 (Deliverable 1.5), in order to predict this enzymatic solubilty of maize biomass. At the same time, saccharification has been performed at York University (Partner 1) using a water pre-treatment and another mix of enzymes (see WP5). Other differences can be noted between both quantifications such as the amount of material treated, time of hydrolysis and quantification methods for degradability. Table 1 summarizes the differences between both quantifications of biomass degradability.

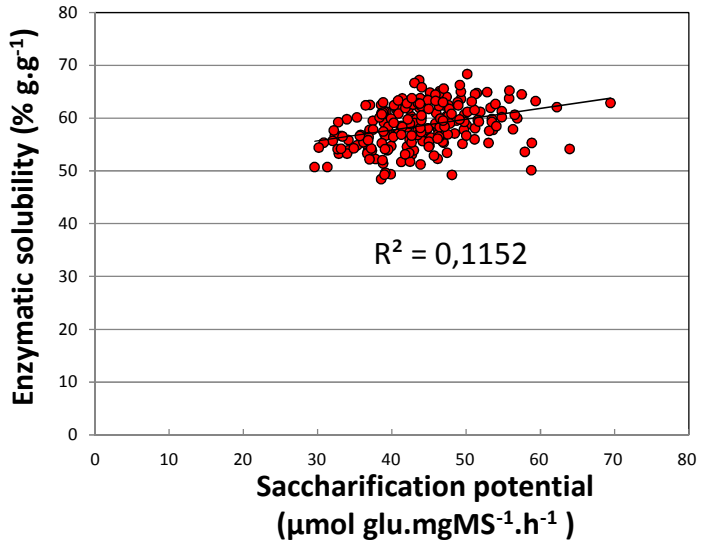
Table 1

Assays	**Digestibilty**	**Saccharification (Univ York)**
Amount/type of sample	DM (250mg)	DM (4mg)
Pretreatment	-Pepsidase -0,1N HCl	-water
Enzymatic Hydrolysis	Onozuka + amyloglucosidase NaAc Buff (24h)	Celluclast + Novozyme 188 NaAc Buff (8h)
Quantification	- Weight Loss (% $g.g^{-1}$)	- Released Sugars (µmol glu. $mgDM^{-1}.h^{-1}$)

In order to get insights into the correlation between these traits, both quantifications have been carried out on maize biomass using samples from a mapping population (D1.1).

Results:

The figure below represents a scatter plot for the relationship between enzymatic solubility versus saccharification potential.



Discussion /Conclusion:

Despite the fact that these two traits are both used to quantify degradability of biomass, their low correlation highlights the fact that they are significantly different. This low correlation is in part due to the protocols that are different (see table 1). The absence of correlation between these quantification methods can be due to:

- the amount of material. Only 4mg of dried mass is used in the High Through-put quantification of saccharification. This small quantity of material is perhaps not representative of the lignocellulosic biomass harvested for each line.

- a pre-treatment (mild and acid) is used for the quantification of digestibility, whereas no pre-treatment is applied in the case of saccharification.

- the duration of hydrolysis is also different between both quantification. Due to the amount of material in each protocol, the duration of hydrolysis is different, in order to fully release sugars from cell walls.

- finally, quantification of degradability is also different (% of weight loss for digestibility *versus* amount of equivalent glucose released per hour for saccharification) making difficult the comparison between both traits.

These results impact the way the selection should be driven. For digestibility, the protocol is being commonly used and adopted in the field of animal feeding. Selection of more digestible maize lines is driven using this protocol by private companies. However, the protocol for saccharification (nature of pre-treatment, choice of enzymes, duration of hydrolysis etc.) will vary between biorefinery scenarios proposed. This can impact the selection and the choice of biomass to be tailored for each scenario.