

## Deliverable 3.3

# Report on preliminary pretreatment results of combinations of feedstocks

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### PUBLISHABLE SUMMARY

The BIOrescue project aims to develop and demonstrate a new innovative biorefinery concept based on the cascading use of spent mushroom substrate (SMS) supplemented by wheat straw (and other seasonal underutilised lignocellulosic feedstocks). Within this biorefinery concept, the first step is a two-stage process (Separation and fractionation) which consists of a solid-liquid extraction and it is followed by a thermochemical pretreatment (second step) that aims to fractionate the SMS into different components that can be subsequently transformed and/or upgraded into valuable bio-based products.

This report includes the information about the thermochemical pretreatment assays carried out using SMS in combination with other substrates such as wheat straw (WS) and other underutilised lignocellulosic feedstocks (barley and oat straw). The optimum binary mixtures of SMS and WS and ternary using SMS, Barley and Oat Straw as underutilised feedstocks selected from the Western European Region have been presented in the previous deliverable (D2.4).

More in detail, this report contains the information regarding the composition of the pretreated samples obtained (soluble and insoluble fractions). Regarding the insoluble fraction, its digestibility after enzymatic hydrolysis using enzymes supplied by MetGen is reported. On the other hand, and the amount of lignin solubilised is also reported as it is being used for nanocapsules production within WP5. The conditions tested for thermochemical fractionation were selected based on best preliminary results obtained in previous assays carried out with the mixture of SMS and WS (see section 4.1).

The results obtained are very interesting when considering the provision of alternative feedstocks for securing the supply of a biorefinery plant. In conclusion, to optimise the pretreatment process to produce soluble sugars, the following issues have to be taken into account:

- Complexity and composition of the feedstock mixture:

- It seems that oat and barley are more recalcitrant to enzymatic hydrolysis than wheat straw. Thus, longer residence time is recommended for ternary mixture generating a solid more prone to enzymatic hydrolysis whereas for binary mixture the shorter residence time seems to perform better.
- Ternary mixtures generated slightly higher soluble glucose and xylose concentrations obtained after enzymatic hydrolysis and greater yields either compared to binary ones. Indeed, the amount of glucose solubilized after enzymatic hydrolysis for the combinations with barley and oat straw are similar or even slightly higher compared to the binary mixture used as control.

- Enzymatic hydrolysis yields and enzyme performance:

- There is still some room for improvement for glucose production for the mixtures containing oat and barley and also in case of the mixture just containing SMS and WS where there is still non-hydrolysed glucose remaining. In any case, the yields obtained for all these assays exceeded the threshold of 85%.
- Regarding xylose production, it is clearly shown that the shorter residence time the higher xylose values obtained.

In conclusion, it has been shown along the report that little differences have been found between using ternary or binary mixtures. Thus, both barley and oat could be perfectly used as alternative feedstocks in binary samples with SMS instead of wheat straw, reinforcing biorefinery's feedstock supply. Regarding the soluble compounds found in the liquid fraction and devoted for nanocapsules production, carbohydrate derived compounds and little amounts of lignin derived compounds have been found. The complete information about this specific topic will be included with more detail in D5.2. report.