

1 **Environmental assessment of Biorefinery processes for the valorization of lignocellulosic**
2 **wastes into oligosaccharides**

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8 **Abstract**

9 The European Commission is adopting strategies in order to "closing the loop" of product life cycles in
10 industrial production systems through better recycling and re-use under the perspective of circular
11 economy. One of the most relevant goals in the application of this approach is to convert low-value
12 side streams into more valuable products. In this sense, the conversion of lignocellulosic biomass into
13 biofuels and chemicals is a major challenge not only from the technological but also from the
14 economic and environmental perspectives.

15 In this study, the production of pectin-derived oligosaccharides (POS) from sugar beet pulp (SBP) was
16 environmentally assessed by means of the Life Cycle Assessment methodology under a cradle-to-
17 gate approach. Two different scenarios at pilot scale were considered: Scenario 1 based on
18 conventional autohydrolysis at high temperature and Scenario 2 based on enzymatic hydrolysis.

19 The outcomes of this environmental study are highly dependent on the production yield of the target
20 compounds (POS) and the valorisation strategy considered. In fact, the POS yield of the
21 autohydrolysis approach is around 20% higher than in the enzymatic one. According to the results,
22 Scenario 1 reports the worst results when a functional unit based on the amount of valorised material
23 (100 kg of oven-dried SBP) is considered. However, the profile entirely changes when a unit based on
24 the economic revenue (1 €) is managed. Therefore, attention should be paid on the selection of the
25 functional unit since decision making strategies should highly depend on it.

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27 Without waiting for the opportunity to conduct LCA of already-developed processes of the biorefinery
28 system, the development of new alternatives must be carried out with sustainability in mind.
29 Accordingly, the proposal of valorisation strategies for secondary streams should include the analysis
30 of the environmental impacts associated to each alternative, even at the pilot plant stage.

31 **Keywords:** environmental profile; life cycle assessment; pectin-derived oligosaccharides; prebiotics;
32 sugar beet pulp

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1. Introduction

Sugar beet pulp is an abundant co-product from the sugar industry (Al-Tamimi et al., 2006; Zheng et al., 2013). Around 5 Mtons are produced in European countries (Ziemiński et al., 2014), of which nearly 20% is produced in Spain mainly for animal feed (Martínez et al., 2009a; Concha Olmos and Zúñiga Hansen, 2012).

The sustainable development of bio-processes in the short and medium-term is receiving special attention due to the fast depletion of fossil fuels, their fluctuating price as well as the increasing concerns on greenhouse gases (GHG) emission (Huisingsh et al., 2015; Montazeri and Eckelman, 2015; Philp, 2015). Therefore, the current industry must then face the challenge to transform industrial low-value side products into more value-added materials (Pin et al., 2014; González-García et al., 2016; Van Uytvanck et al., 2014). In this sense, the implementation of the biorefinery scheme in the framework of the sugar beet industry must promote the innovation technologies that increase not only the yield and profitability of the processes but also that improve the environmental profile (Huisingsh et al., 2015; González-García et al., 2016; Russell and Shiang, 2013).

Thus, sugar beet pulp (from now SBP) - a lignocellulosic source, could be considered a promising by-product with potential valorisation in a biorefinery scheme due to its composition and abundance. In the sugar beet processing system, the main product is sucrose, which accounts for 10-12% of the total biomass processed. However, the analysis of the SBP composition also comprises 20-25% cellulose, 25-36% hemicelluloses (mainly arabinans), 20-25% pectin, 10-15% protein and 1-2% lignin (Concha Olmos and Zúñiga Hansen, 2012; Ziemiński et al., 2014).

Although feed formulation is the main use of SBP (Concha Olmos and Zúñiga Hansen, 2012), other applications are also possible (Ziemiński et al., 2014), such as in the production of composites (Liu et al., 2005), galacturonic acid and arabinose (Leijdekkers et al., 2013) or biogas (Ziemiński et al., 2014) as well as in the papermaking industry due its low lignin content (Bellido et al., 2015). In recent studies, SBP has been evaluated as a potential source for the production of functional carbohydrates,

59 paying attention to neutral and acidic oligosaccharides by means of a direct enzymatic saccharification
60 treatment (Concha Olmos and Zúñiga Hansen, 2012; Martínez et al., 2009a).

61 Oligosaccharides are attracting increasing interest as prebiotic functional food ingredients (Rastall,
62 2010; Patel and Goyal, 2011) as they may confer health benefits on the host associated with the
63 modulation of microbiota (Martínez et al., 2009b), especially pectin-derived oligosaccharides (POS)
64 (Rastall, 2010; Patel and Goyal, 2011). POS can be produced from waste biomass or low-cost
65 byproducts by applying a selective fractionation based on physical, chemical and enzymatic methods
66 (Sun and Hughes, 1998; Patel and Goyal, 2011).

67 To the best of our knowledge, only one environmental study has been published with special focus on
68 the environmental footprints of different production schemes of soluble saccharides of polymeric and
69 oligomeric nature from woody residual streams (González-García et al., 2016). In this work, the
70 assessment of the environmental impacts associated to the valorisation of SBP from the sugar refining
71 industry for the production of POS was performed. To do this, a Life Cycle Assessment (LCA) was
72 undertaken to analyse two valorisation scenarios based on different extraction routes considering SBP
73 as raw material. On the basis of experimental results carried out at semi-pilot scale, the environmental
74 *hotspots* responsible of the largest environmental impacts were identified. This environmental
75 methodology considers all the resources (mass and energy balances) required to make a product, the
76 wastes generated as well as the environmental burdens associated with the product (Goedkoop et al.,
77 2008). In addition, this methodology can provide useful information to stakeholders and policy makers
78 on decision making of processes under development. The principles established by ISO standards
79 (ISO 14040, 2006) were followed in this research study.

80 **2. Materials and methods**

81 **2.1 Goal and scope definition**

82 This study aims to assess the environmental performance of different extraction routes of SBP from
83 the sugar industry to obtain a POS extract with high content in arabino-oligosaccharides and

84 oligogalacturonides. Side-streams are also derived from the system which could be used for further
1 85 applications such as a solid stream rich on cellulose for bioethanol production and a liquid stream rich
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3 on monosaccharides. The description of both valorisation scenarios is included below.
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8 87 ▪ Scenario 1 based on the autohydrolysis of SBP (hydrothermal processing under non-
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10 88 isothermal conditions) performed in a stirred and pressurised stainless steel reactor at 163°C
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12 89 (Martínez et al., 2009b).

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16 90 ▪ Scenario 2 based on the enzymatic hydrolysis of SBP performed by a combined enzyme
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18 91 concentrate of Cellulast and Viscozyme at 37°C (Martínez et al., 2009a).

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22 92 A cradle-to-gate approach was considered in the scenarios under assessment that is, considering the
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24 93 extraction of raw materials to produce the required inputs and the production of POS but not the
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26 94 phases of use and/or final disposal of POS. This perspective was assumed since the production
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28 95 systems are at semi-pilot scale and the products are not available in the markets yet. Among the
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30 96 processes considered throughout the production life cycle of both valorising schemes, centrifugation,
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32 97 membrane concentration and freeze-drying are performed after the extraction phase.
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36 98 Considering that SBP is a co-product of the sugar refining process, the background process of SBP
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38 99 production (activities directly linked to the sugar production) was excluded from the assessment. Thus,
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40 100 derived environmental impacts were entirely allocated to the sugar.
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45 101 The LCA functional unit must be selected carefully to allow comparisons between the valorising
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47 102 systems under study. Thus, the functional unit is defined as the valorisation of 100 kg of dry SBP at
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49 103 the factory gate, provided by a local pulp factory (Martínez et al., 2009b).
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53 104 **2.2 Description of the SBP valorisation scenarios**

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57 105 Each valorisation scenario destined to the production of POS was divided in five different subsystems
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59 106 which are depicted in **Figure 1**: hydrolysis (autohydrolysis - SS1.1 or enzymatic hydrolysis - SS1.2),
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107 centrifugation (SS2), concentration (SS3), diafiltration (SS4), freeze-drying (SS5) and wastewater
108 treatment (SS6). The subsystems were assessed from the production of raw materials (resources) up
109 to the final product at the pilot plant gate. The analysis of a pilot scale process located in the Faculty of
110 Sciences from the University of Vigo (Orense, NW Spain) was performed. As indicated above, further
111 processing and transport activities were excluded from the system boundaries as well as the activities
112 related with the production of the SBP used as raw material. In both scenarios, SBP (70-72%
113 moisture) was provided by a local pulp factory, homogenised in a single lot to avoid compositional
114 differences, and stored in polyethylene bags at -18°C until use.

115 <Figure 1 around here>

116 *Scenario 1 – Autohydrolysis treatment*

117 SBP is subjected an aqueous processing (autohydrolysis treatment – SS1.1) with tap water in a ratio
118 of 12 g water per g dry pulp. In this work, the reactor was heated up to achieve a maximum
119 temperature of 163°C, the optimum conditions reported for the production of pectic-oligomers from
120 SBP (Martínez et al. 2009a). The autohydrolysis treatment permits the selective breakdown of pectic
121 polymers to give valuable soluble hydrolysis products such as pectin-oligomers (POS) and a solid
122 fraction rich in cellulose and lignin. At the end of treatment, the reactor was rapidly cooled to 60°C for
123 12 min, and the liquors were separated from the spent solids by centrifugation (SS2). **Table 1** shows
124 the composition of the autohydrolysis liquors.

125 <Table 1 around here>

126 *Scenario 2 – Enzymatic Hydrolysis treatment*

127 Based on previous studies (Martínez, 2009b), POS mixtures were obtained by enzymatic processing
128 of SBP (SS1.2) using commercial enzymes (Celluclast 1.5L cellulases from *Trichoderma reesei* and
129 Viscozyme 1.5L endopolygalacturonases from *Aspergillus aculeatus*). The experimental conditions
130 considered were the following: water/solid ratio of 12 g/ g dry pulp, endopolygalacturonase/solid ratio

131 of 10 U¹/g, cellulase/endopolygalacturonase ratio of 0.725 FPU²/U and reaction time of 12.8 h. Sodium
132 acetate buffer was used to maintain the medium at pH~5. At the end of process, liquors were
133 separated by centrifugation (SS2). The experiments were carried out at 37°C in Erlenmeyer flasks with
134 orbital agitation (150 rpm). **Table 2** shows the composition of the hydrolysates obtained by enzymatic
135 treatment.

136 <Table 2 around here>

137 The autohydrolysis and enzymatic liquors (from SS1.1 and SS1.2 respectively) were treated for
138 purification and concentration using a membrane processing unit (SS3 and SS4) in order to get a
139 stream rich on POS. The experiments were carried out using a polymeric spiral membrane with
140 molecular weight cut-off of 1 kDa and pressure of 8 bar. The processing was carried out at room
141 temperature.

142 The processing started with the concentration of both streams to achieve a volume/concentration
143 (VCR) ratio of 5 (SS3). For additional refining, water was added to the retentate to reach initial volume,
144 and the resulting solution was concentrated again (discontinuous diafiltration – SS4) to achieve the
145 same VCR. At the end of processing, two streams were obtained, a retentate rich on POS and a
146 permeate rich on monosaccharides and other non-saccharide compounds. Finally, the retentate
147 stream was freeze-dried (SS5) and permeates from SS4 and SS5 in both scenarios were sent to a
148 wastewater treatment plant (SS6) for further treatment.

149 **2.3 Inventory data collection and allocation approach**

150 A reliable environmental assessment requires the collection of Life Cycle Inventory (LCI) data. In this
151 study, inventory data for the foreground system (direct inputs and outputs for each stage) such as
152 electricity requirements (estimated with power and operational data from the different units: reactors,
153 centrifuges, membranes, orbital shakers and freeze-dryers) as well as the use of chemicals, enzymes

¹ U: unit of enzymatic activity. It is defined as the amount of enzyme catalyzing the formation of 1 µmol of D-galacturonic acid per minute at 37°C and pH 5.

² FPU: Filter Paper Activity

154 and water were average data of the pilot plant (primary data). A summary of the primary data is
155 displayed in **Table 3** for the different valorisation scenarios. According to this table, the POS
156 production yield (the main desired product) is around 20% higher in the autohydrolysis based scenario
157 than in the enzymatic one. This result could be expected due to the enzymatic treatment commonly
158 produces low weight POS which should derive on largest losses ratios in the purification units
159 (membranes).

160 <**Table 3** around here>

161 Concerning the background system, the inventory data corresponding to the production of the different
162 inputs to the systems (electricity, sodium acetate and tap water) and the wastewater treatment
163 process were taken from Ecoinvent database® v3.1. Regarding sodium acetate, the specific
164 production process is not included in the database and therefore, a representative process
165 corresponding to the production of inorganic chemicals was considered instead of excluding it from the
166 system boundaries. Regarding the production of enzymes (Viscozyme and Celluclast), information
167 concerning the environmental burdens has been directly supplied by Novozymes³ under confidential
168 issues. Detailed information with regard to the background processes is summarised in **Table 4**.

169 <**Table 4** around here>

170 All the scenarios under assessment are multi-outputs systems with more than one product. Allocation
171 of the environmental impacts is required for multi-functional processes and the selection of an
172 allocation approach can have a strong effect on the results. According to the functional unit chosen
173 based on the amount of waste valorized per batch (100 kg of dry SBP), no allocation procedure was
174 required. The economic value of all the bioproducts differs significantly, which should motivate the use
175 of an economic based approach. Therefore, an alternative functional unit in terms of economic value
176 was considered based on the expected economic revenues of the bioproducts obtained in each
177 scenario. **Table 5** reports the market prices considered for the different co-products and the expected
178 net revenue.

³ <http://www.novozymes.com> .

179 <Table 5 around here>

180 2.4 Impact assessment methodology

181 LCA evaluates the environmental burdens by identifying resource and energy consumption as well as
182 emissions to different environmental compartments associated to the life cycle of the process under
183 assessment, including the identification of priority areas to apply improvement actions (ISO 14040,
184 2006). The characterisation factors reported by the Centre of Environmental Science of Leiden
185 University - CML 2001 method (Guinée et al. 2001) v2.05 were considered in this study for the
186 analysis. The following impact categories were evaluated: abiotic depletion potential (ADP),
187 acidification potential (AP), eutrophication potential (EP), global warming potential (GWP),
188 photochemical oxidation potential (POFP), ozone layer depletion potential (ODP), human toxicity
189 potential (HTP), terrestrial ecotoxicity potential (TEP), freshwater aquatic ecotoxicity potential (FEP)
190 and marine aquatic ecotoxicity potential (MEP). Moreover, SimaPro 8.02 was the software used for
191 the computational implementation of the LCI (PRé Consultants, 2016).

192 3. Results and discussion

193 The environmental assessment in terms of characterisation results obtained for both schemes is
194 displayed in **Table 6**. As previously indicated, the production of SBP, the main raw material, was
195 excluded from the study because it was managed as a waste stream from sugar factories. Thus,
196 environmental burdens derived from sugar processing are totally allocated to the main product of the
197 production systems, i.e. sugar.

198 <Table 6 around here>

199 According to the environmental results presented in **Table 6**, remarkable differences can be identified
200 between both valorising schemes of SBP for the production of POS, which should be directly related
201 with differences on the production systems. Although they share a number of stages in common
202 (centrifugation – SS2, concentration – SS3, diafiltration – SS4, freeze-drying – SS5 and wastewater

203 treatment – SS6), the main difference between both routes is focused on the first stage or subsystem,
204 that is the hydrolysis of the SBP (SS1.1 and SS1.2 for scenarios 1 and 2, respectively). In Scenario 1,
205 it is a hydrothermal processing under non-isothermal conditions carried out at high temperature
206 (163°C). On the contrary, Scenario 2 considers an enzymatic step based on the use of a cocktail of
207 enzymes at moderate temperature (37°C). This fact entails differences on the electricity requirements,
208 being 565 times larger in Scenario 1 than Scenario 2. However, Scenario 2 requires enzymes whose
209 production also entails energy consumption. Accordingly, Scenario 1 reports the worst environmental
210 results in most categories except ODP, HTP, MEP and TEP. A detailed analysis of these results will
211 be discussed below.

212 *Contributions per subsystems involved*

213 **Figure 2a and 2b** displays the distribution of environmental burdens per subsystems involved in the
214 production schemes for Scenarios 1 and 2, respectively.

215 <**Figure 2** around here>

216 According to **Figure 2a**, the majority of the environmental burdens associated with Scenario 1 are
217 related to the freeze-drying step (SS5) and the autohydrolysis stage (SS1.1). Freeze-drying process
218 requires electricity consumption in order to obtain the final product (rich on POS and with low content
219 on monosaccharides) while autohydrolysis is also energy intensive since the reactor operates at
220 163°C. Both subsystems are responsible (on average) for 69% and 20% of the total contributions,
221 respectively (see **Figure 2a**) and the difference in energy requirement is very relevant: 3.5 times
222 higher in SS5 than in SS1.1 (**Table 3**). The equipment used in the subsequent steps (centrifuge and
223 membranes) requires the lowest amounts of electricity and therefore, their contributions to the
224 environmental profile are negligible regardless the impact category.

225 Concerning **Figure 2b**, modifications on the environmental profile can be identified. Once again SS1.2
226 and SS5 are the steps responsible for the majority of contributions to environmental impacts although
227 with differences on the relative values and categories. SS5 is the environmental *hotspot* in all the

228 categories with contributions ranging from 53% to 80% except in terms of MEP where SS1.2 is the
1
2 229 main contributing subsystem with a contributing ratio of 74%. Enzymes, acetate and electricity
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4 230 production are the processes behind this impact. Enzymes production is a high intensive process,
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6 231 which demands large amounts of energy. In addition, enzymes production requires the addition of
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8 232 chemicals for the formulation of the culture medium and, the background processes related to their
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10 233 production are the main responsible of contributions to MEP.

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14 234 If the source of the environmental burdens derived from SS1.2 is assessed in more detail, two inputs
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16 235 are identified as major responsible of impacts: the production of the enzyme cocktail for the enzymatic
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18 236 hydrolysis and the production of acetate for pH control. **Figure 3** displays the distribution of impacts
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20 237 derived from SS1.2 between the involved processes and, in line with the results, the production of
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22 238 acetate should be an environmental key factor, being responsible of the majority of the impacts in all
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24 239 the categories with ratios ranging from 54% to 88% except for MEP and TEP, where the production of
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26 240 the enzymes required for the hydrolysis contribute with 98% and 79% of the total impacts.

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30 241 <**Figure 3** around here>

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34 242 Total electricity requirements per batch in Scenario 2 are only 13% of the electricity consumed in
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36 243 Scenario 1 and enzymatic hydrolysis step reports the lowest energy requirements followed by SS2,
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38 244 SS3, SS4 and SS5, However, in Scenario 1 the highest electricity requirement is also observed in SS5
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40 245 followed by SS1.1, SS3, SS4 and SS2. **Table 3** displays the distribution of electricity requirements
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42 246 between the subsystems directly involved in the valorising systems, excluding SS6 since the
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44 247 wastewater treatment plant is beyond the premises of the manufacture plant.

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49 248 *Alternative functional unit and allocation strategies*

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53 249 According to the results reported so far, Scenario 1 presents much higher environmental burdens than
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55 250 Scenario 2 in six impact categories. These results are related to a functional unit based on the amount
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57 251 of SBP used: 100 kg of dry SBP. This functional unit can be considered useful when valorising
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59 252 systems are being analysed and where multiple by-products are obtained since it corresponds to the

253 amount of valorised material per batch (Pérez-López et al., 2014; González-García et al., 2016).
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2 254 Nevertheless, the amount of valuable by-products, which also present different market prices,
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4 255 depends on the scenario. Therefore, the potential revenue obtained per scenario should be different
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6 256 depending on the valorising strategy. The selection of the functional unit to report the results has just
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8 257 been previously discussed in numerous studies where biosystems have been environmentally
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10 258 analysed (Kim and Dale, 2006; Pérez-López et al., 2014; González-García et al., 2016). According to
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12 259 these studies, special attention must be paid on the selection of the functional unit to estimate the
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14 260 environmental profiles since strategic decisions highly depend on it.

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18 261 For that reason, an alternative unit based on the potential economic benefit expected from each
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20 262 valorisation scenario could be of interest. In addition, an allocation procedure was also proposed here
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22 263 considering an economic allocation approach. The alternative functional unit proposed is the
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24 264 generation of 1 € of economic revenue from the valorisation of SBP into high added value products.
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26 265 Market prices available in the literature (Mercopress, 2006; Marketing study, 2009; Sigma-Aldrich,
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28 266 2016; Megazyme, 2016) were managed and presented in **Table 5**.

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32 267 **Figure 4** displays the comparative profile between both scenarios considering 1 € of economic
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34 268 revenue as functional unit and it considerably changes in comparison with the profile corresponding to
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36 269 a functional unit based on the amount of processed biomass per batch. According to the results
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38 270 reported, Scenario 1 should involve the lowest impacts in all the categories under study. The
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40 271 reductions on the impacts should be remarkable in terms of MEP and TEP, where the enzymatic
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42 272 treatment based scenario reports 3.8 and 1.6 times higher burdens than Scenario 1. Both impact
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44 273 categories are significantly affected by the contributions from the enzymatic hydrolysis subsystem
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46 274 (SS1.2) as depicted in **Figure 2b**, being the enzyme production the main responsible factor (see
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48 275 **Figure 2b**).

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53 276 <Figure 4 around here>

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278 If an economic allocation is considered on the basis of the production of POS and other by-products
279 with economic value, the share of the environmental burdens between the different products should be
280 carried out taking into account the allocation factors reported in **Table 4**. Thus, 96.9% and 95.6% of
281 the impacts reported in **Table 5** for Scenario 1 and 2 respectively should be allocated to POS. The
282 majority of burdens should correspond to the production of POS and negligible burdens should be
283 assigned to the remaining by-products (except the uronic acids), mostly due to the lower price of the
284 by-products (lignin, cellulose and hemicellulose). Around 3.1% and 4.4% of the impacts should be
285 attributed to uronic acids. Therefore, when referring to the amount of POS produced, the
286 environmental behaviour should be similar to the results reported in **Figure 4**, presenting Scenario 2
287 the worst profile.

288 According to González-García et al. (2016), purification activities (commonly concentration and freeze-
289 drying processes) as well as autohydrolysis can be identified as the major responsible of
290 environmental burdens when valorising routes into prebiotics are analysed. In our study, hydrolysis
291 and freeze-drying were also identified as environmental *hotspots* specifically due to the high electricity
292 requirements and enzymes (and chemicals) production. Thus, special attention and improvements
293 should be paid on both activities since, regardless the valorisation route and raw material processed,
294 are identified as critical steps.

295 *Limitations of study*

296 LCA methodology can be considered as a valuable and useful tool to support decision making
297 strategies, specifically for production schemes which are still under development. LCA helps to identify
298 the potential environmental risks associated with the production sequence in advance to their
299 complete implementation. Therefore, it can be considered a predictive support not only from
300 environmental but also from economic perspective. However, there are inherent difficulties in
301 performing a LCA of a new bioproduct or production system.

302 In this case study under development, the major obstacles include the uncertainty arising from the
303 immature nature of the technology and markets since the goal product (pectin-derived

304 oligosaccharides) is not still available at commercial scale. However, and according to the literature
1 305 (Gómez et al., 2016; Moniz et al., 2016) natural products and food ingredients with functional and
2 306 healthy properties are receiving growing interest by consumers. Therefore, research efforts are
3
4 307 required in order to enhance their efficiency and improve/optimize their production systems.
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6 308 Nowadays, the commercial oligosaccharides are inulin, lactose and sucrose (Moniz et al., 2016).
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8 309 Thus, the production of oligosaccharides by means of alternative sources such as lignocellulosic
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10 310 biomass is being boosted for multiple reasons not only economic but also environmental. In fact, xylo-
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12 311 oligosaccharides from lignocellulosic biomass are the only oligosaccharides available at present in the
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14 312 market although their market is still limited (Moniz et al., 2016).. The unwilling approach to provide
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16 313 extensive information on the processes under development could also be a limitation specifically in
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18 314 terms of the collection of good quality inventory data. This approach should be carefully managed
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20 315 specifically if the production systems are going to be analysed at pilot scale. Moreover, the lack of risk-
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22 316 associated information and the modelling assumptions (e.g. the use of proxy processes, system
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24 317 boundaries) are items that must be meticulously handled specially in decision making strategies..
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26 318 Moreover, it has been demonstrated that choice of the reference unit in terms of which the
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28 319 environmental results are reported and analysed can considerably change the valorisation strategy
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30 320 selected.
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40 322 **4. Conclusions**

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44 323 The valorisation of refinery side-stream products is receiving special attention and it is a challenge of
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46 324 today's industry. This study has analysed two different valorising routes of sugar beet pulp, by-product
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48 325 of the sugar industry, with the aim of obtaining POS, a product with prebiotic properties. According to
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50 326 the results, special attention must be paid on two specific stages: hydrolysis and freeze-drying of the
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52 327 final product. Moreover, the choice of the functional unit considered for analysis is really important
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54 328 since the results considerably change. The enzymatic based hydrolysis involved the requirement of
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56 329 enzymes whose production also requires energy. In contrast to the conventional hydrolysis carried out
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58 330 at high temperature, the consumption of electricity is 565 times lower. The valorising sequences
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331 analysed in this study appear to be attractive options to produce high-added value products with
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2 332 multiple applications. According to the outcomes, further research should be focused in order to
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4 333 improve the current valorising techniques at pilot scale. Inherent difficulties in performing the LCA of
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6 334 these new biorefinery systems related to the uncertainty arising from the immature nature of the
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8 335 technology are avoided in large scale systems. However, the results from this study help to identify the
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10 336 stages which require special effort to develop and optimize.

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Figure 1. System boundaries and process chain under study corresponding to the Scenario 1 - Autohydrolysis treatment and Scenario 2 – Enzymatic hydrolysis treatment. Processes marked in black were excluded from the system boundaries. Processes and lines marked in grey correspond exclusively to Scenario 2.

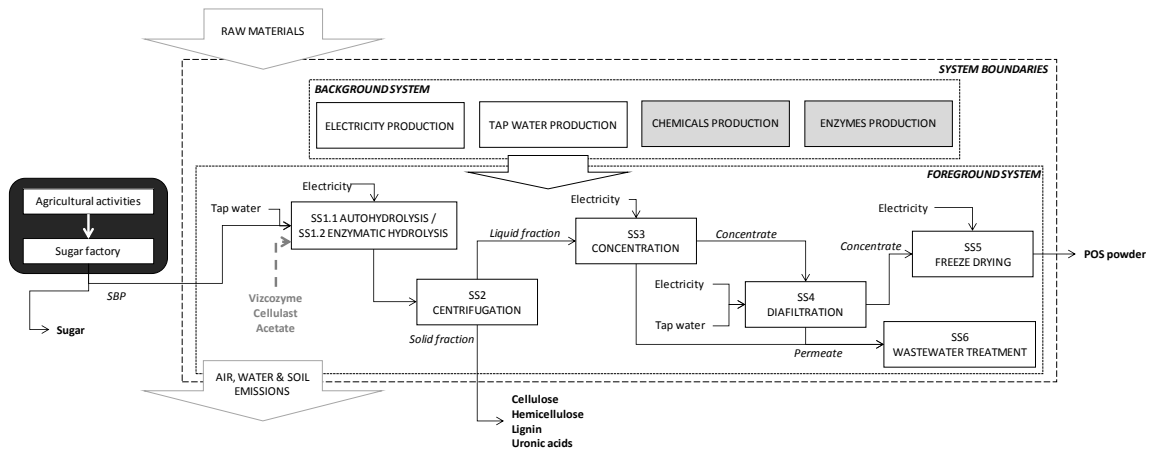
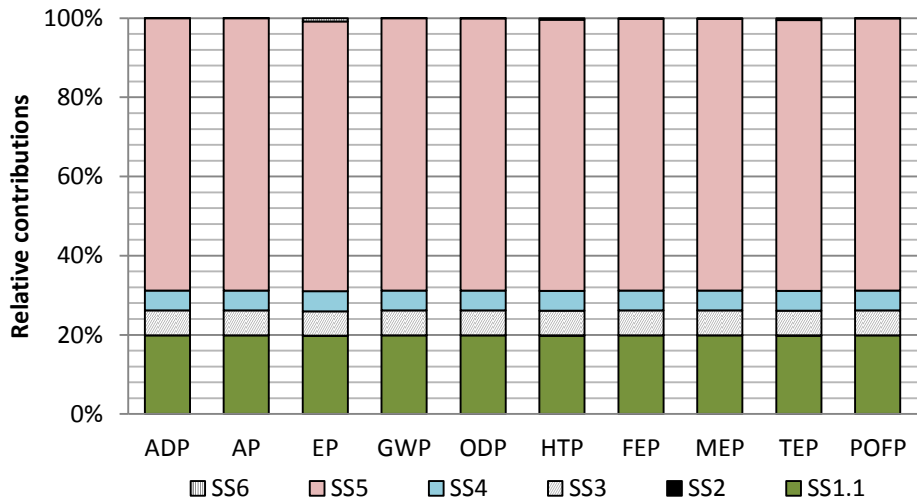
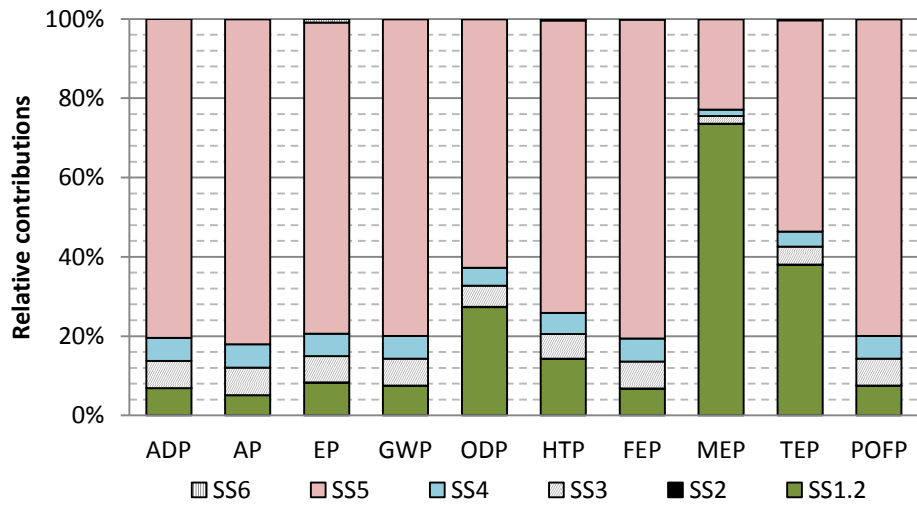


Figure 2. a) Distribution of environmental impacts per subsystems involved in Scenario 1; b) Distribution of environmental impacts per subsystems involved in Scenario 2. Acronyms: abiotic depletion potential (ADP), acidification potential (AP), eutrophication potential (EP), global warming potential (GWP), ozone layer depletion potential (ODP), human toxicity potential (HTP), freshwater aquatic ecotoxicity potential (FEP), marine aquatic ecotoxicity potential (MEP), terrestrial ecotoxicity potential (TEP) and photochemical oxidation potential (POFP).



a



b

Figure 3. Distribution of impact burdens derived from SS1.2 between the different processes involved. Acronyms: abiotic depletion potential (ADP), acidification potential (AP), eutrophication potential (EP), global warming potential (GWP), ozone layer depletion potential (ODP), human toxicity potential (HTP), freshwater aquatic ecotoxicity potential (FEP), marine aquatic ecotoxicity potential (MEP), terrestrial ecotoxicity potential (TEP) and photochemical oxidation potential (POFP).

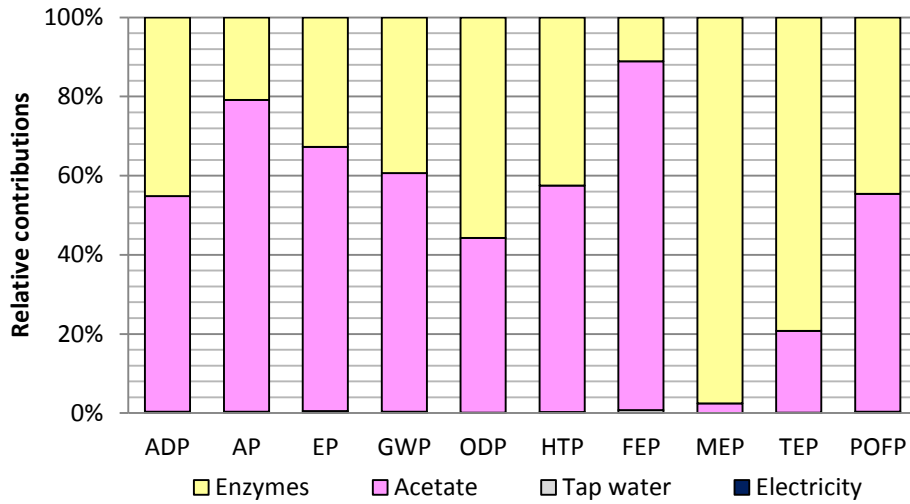
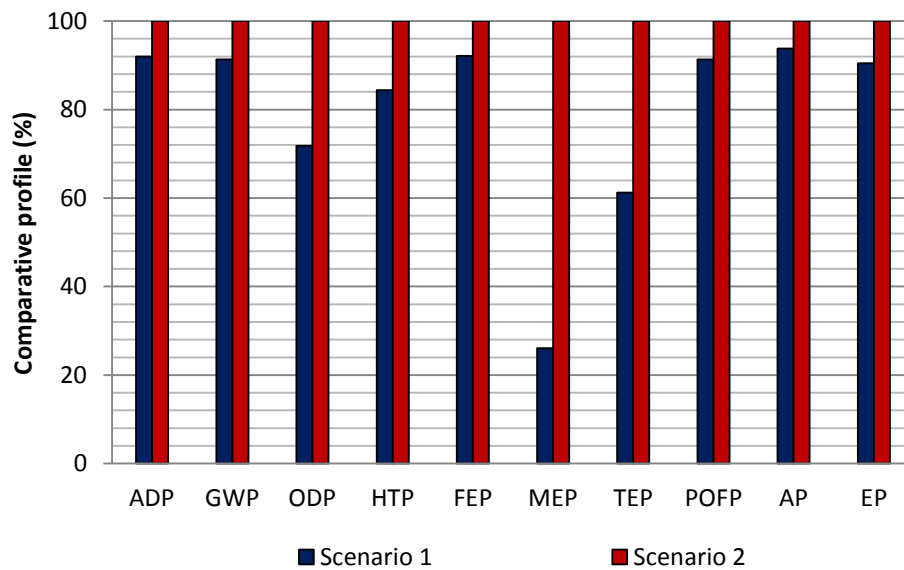


Figure 4. Comparative environmental profile considering 1 € of potential revenue as functional unit. Scenario 1 – Autohydrolysis treatment; Scenario 2 – Enzymatic hydrolysis treatment. Acronyms: abiotic depletion potential (ADP), global warming potential (GWP), ozone layer depletion potential (ODP), human toxicity potential (HTP), freshwater aquatic ecotoxicity potential (FEP), marine aquatic ecotoxicity potential (MEP), terrestrial ecotoxicity potential (TEP), photochemical oxidation potential (POFP), acidification potential (AP) and eutrophication potential (EP).



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4**Table 1.** Composition of liquid fraction (rich on pectic oligosaccharides) obtained by autohydrolysis treatment.

Component	Composition
<i>Monosaccharides</i>	
Glucose	2.36 g L ⁻¹
Galactose	2.43 g L ⁻¹
Arabinose	0.39 g L ⁻¹
<i>Pectin-derived oligosaccharides (POS)</i>	
GlcOS	1.61 g L ⁻¹
GalOS	2.96 g L ⁻¹
AraOS	14.44 g L ⁻¹
AcO	2.06 g L ⁻¹
OGalA	11.86 g L ⁻¹
<i>Volatile compounds</i>	
Acetic acid	0.38 g L ⁻¹
<i>Other non-volatile compounds</i>	
	8.21 g L ⁻¹

GlcOS = glucooligosaccharides; GalOS = galactooligosaccharides; AraOS = arabinooligosaccharides; AcO = acetyl substituents in oligomers; OGalA = oligogalacturonides

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9 **Table 2.** Composition of hydrolysates (rich on pectic oligosaccharides) obtained by enzymatic processing.

Component	Composition
<i>Monosaccharides</i>	
Glucose	4.89 g L ⁻¹
Galactose	2.90 g L ⁻¹
Galacturonic acid	3.50 g L ⁻¹
Arabinose	0.31 g L ⁻¹
<i>Pectin-derived oligosaccharides (POS)</i>	
GlcOS	6.59 g L ⁻¹
GalOS	3.02 g L ⁻¹
AraOS	11.21 g L ⁻¹
AcO	1.62 g L ⁻¹
OGalA	10.29 g L ⁻¹
<i>Volatile compounds</i>	
Acetic acid	0.55 g L ⁻¹
<i>Other non-volatile compounds</i>	
	13.87 g L ⁻¹

GlcOS = glucooligosaccharides; GalOS = galactooligosaccharides;
AraOS = arabinooligosaccharides; AcO = acetyl substituents in
oligomers; OGalA = oligogalacturonides

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12 **Table 3.** Summary of main relevant inventory data per functional unit for POS powder production from SBP
 13 under two valorising scenarios: Scenario 1 - . Autohydrolysis treatment and Scenario 2 - Enzymatic hydrolysis
 14 treatment.

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	Scenario 1	Scenario 2
Inputs		
<i>Autohydrolysis (SS1.1)</i>		
SBP (oven-dried)	100 kg	--
Tap water	945 kg	--
Electricity	1802.3 MJ	--
<i>Enzymatic hydrolysis (SS1.2)</i>		
SBP (oven-dried)	--	100 kg
Tap water	--	912.5 kg
Electricity	--	3.19 MJ
Viscozyme	--	0.18 kg
Celluclast	--	3.30 kg
Sodium acetate	--	30.0 kg
<i>Centrifugation (SS2)</i>		
Electricity	3.51 MJ	3.51 MJ
<i>Concentration (SS3)</i>		
Electricity	567 MJ	577.8 MJ
<i>Diafiltration (SS4)</i>		
Electricity	456.3 MJ	487.8 MJ
Tap water	726.2 kg	768.4 kg
<i>Freeze-drying (SS5)</i>		
Electricity	6234 MJ	6808 MJ
Outputs		
POS powder from SS5	25.75 kg	21.80 kg
Purified POS	25.40 kg	20.60 kg
Monosaccharides	0.35 kg	1.20 kg
Wastewater to treatment (from SS3+SS4)	1452.4 kg	1536.8 kg
Solid fraction from SS2	390.6 kg	338.9 kg
Cellulose (dry basis)	14.60 kg	6.70 kg
Hemicellulose (dry basis)	7.71 kg	10.33 kg
Lignin (dry basis)	7.50 kg	7.70 kg
Uronic acids (dry basis)	3.95 kg	4.90 kg

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21 **Table 4.** List of the main ecoinvent database ® processes considered in this study

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Input	Ecoinvent database ® process
Electricity	Electricity, medium voltage {ES} market for Alloc Rec, U
Tap water	Tap water {Europe without Switzerland} market for Alloc Rec, U
Sodium acetate	Chemical, inorganic {GLO} market for chemicals, inorganic Alloc Rec, U
Wastewater	Wastewater, average {CH} treatment of, capacity 1E9l/year Alloc Rec, U

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24 **Table 5.** Market prices and net economic yield expected for each valorising scenario as well as allocation
25 factors based on economic allocation approach.

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Co-products	Market price	Allocation factors	
		Scenario 1	Scenario 2
POS powder	16.5 €·g ⁻¹⁽¹⁾	96.9%	95.6%
Lignin	302 €·t ⁻¹⁽²⁾	0.0%	0.0%
Cellulose	686 €·t ⁻¹⁽²⁾	0.0%	0.0%
Hemicellulose	102 €·t ⁻¹⁽³⁾	0.0%	0.0%
Uronic acids	3.39 €·g ⁻¹⁽⁴⁾	3.1%	4.4%
Economic yield			
Scenario 1	438,279 €·batch ⁻¹		
Scenario 2	376,319 €·batch ⁻¹		

¹Megazyme (prices of arabinan); ²Mercopress, 2006; ³Marketing study, 2009; ⁴Sigma-Aldrich, 2016

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30 **Table 6.** Impact assessment characterisation values corresponding to the production of POS powder under two
31 valorising schemes, per functional unit (100 kg of oven-dried SBP at the factory gate) and relative environmental
32 difference between both scenario.

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Impact categories	Scenario 1	Scenario 2	Scenario 1 /Scenario 2
ADP (kg Sb eq)	9.45	8.82	+7%
GWP (kg CO ₂ eq)	1299	1221	+6%
AP (kg SO ₂ eq)	12.37	11.33	+9%
EP (kg PO ₄ ⁻³ eq)	2.35	2.23	+5%
ODP (kg CFC-11 eq)	8.23·10 ⁻⁵	9.84·10 ⁻⁵	-16%
POFP (kg C ₂ H ₄ eq)	4.51·10 ⁻¹	4.24·10 ⁻¹	+6%
HTP (kg 1,4-DB eq)	235	239	-2%
TEP (kg 1,4-DB eq)	2.74·10 ⁻¹	3.84·10 ⁻¹	-29%
FEP (kg 1,4-DB eq)	365	340	+7%
MEP (kg 1,4-DB eq)	1385	4458	-69%

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