

conditions, the highest attained lignin removal was larger than 80%. Even if the temperature positively influences lignin removal, it negatively affects the solid fraction yield, mainly containing the remaining cellulose and hemicellulose fractions, eventually leading to the fermentable sugars usable for subsequent bioprocesses. Decreasing the hydrolysis temperature to 60°C led to 71% delignification and 70% solid yield, which are not far from those predicted as optimal by the models considered in this work. Confocal laser scanning microscopy confirmed the delignification of the samples by a significant decrease in the characteristic autofluorescence of lignin. Scanning electron microscopy analysis of the samples indicated better exposition of the cellulose fiber structure. The polymer structure was mainly conserved, suggesting that the hydrolysis to develop reducing sugars may require tough conditions.

Data Availability Statement: All data have been exposed in the article or are available upon request.

Author Contributions: G.K. & M.P.B: Investigation, experimental data acquisition, data curation; GS: data curation, formal analysis, writing and visualization; M.C.: Conceptualization, experimental design, writing original draft, supervision, funding acquisition; C.D.B: Consultation, review, and editing; M.G.: Conceptualization, experimental design, consultation, funding acquisition.

Funding: The authors gratefully acknowledge the financial support from Universidad de Buenos Aires (UBACyT 20020170100604BA), UNSAM, CONICET (PIP 1122015-0100902CO, PIO), and Suomen Kulttuurirahasto (00210970).

Data Availability Statement: Not applicable.

Conflicts of Interest: The authors declare no conflict of interest.

References

- European Union, P.O. of the E. Masterplan for a Competitive Transformation of EU Energy-Intensive Industries Enabling a Climate-Neutral, Circular Economy by 2050. Available online: <http://op.europa.eu/en/publication-detail/-/publication/be308ba7-14da-11ea-8c1f-01aa75ed71a1> (accessed on 21 December 2021).
- International Energy Agency *Renewables 2018: Analysis and Forecasts to 2023.*; 2018; ISBN 978-92-64-30684-4.
- Vargas, F.; Domínguez, E.; Vila, C.; Rodríguez, A.; Garrote, G. Agricultural Residue Valorization Using a Hydrothermal Process for Second Generation Bioethanol and Oligosaccharides Production. *Bioresource Technology* **2015**, *191*, 263–270, doi:10.1016/j.biortech.2015.05.035.
- Toor, M.; Kumar, S.S.; Malyan, S.K.; Bishnoi, N.R.; Mathimani, T.; Rajendran, K.; Pugazhendhi, A. An Overview on Bioethanol Production from Lignocellulosic Feedstocks. *Chemosphere* **2020**, *242*, 125080, doi:10.1016/j.chemosphere.2019.125080.
- Gallone, B.; Mertens, S.; Gordon, J.L.; Maere, S.; Verstrepen, K.J.; Steensels, J. Origins, Evolution, Domestication and Diversity of *Saccharomyces* Beer Yeasts. *Current Opinion in Biotechnology* **2018**, *49*, 148–155, doi:10.1016/j.copbio.2017.08.005.
- Takada, M.; Chandra, R.; Wu, J.; Saddler, J.N. The Influence of Lignin on the Effectiveness of Using a Chemithermomechanical Pulping Based Process to Pretreat Softwood Chips and Pellets Prior to Enzymatic Hydrolysis. *Bioresource Technology* **2020**, *302*, 122895, doi:10.1016/j.biortech.2020.122895.
- Biofuels, Solar and Wind as Renewable Energy Systems: Benefits and Risks*; Pimentel, D., Ed.; Springer: Dordrecht, 2008; ISBN 978-1-4020-8653-3.
- McCaherty, J.; Wilson, C.; Cooper, G. 2019 ETHANOL INDUSTRY OUTLOOK - Renewable Fuels Association. **2019**.
- Liu, C.-G.; Xiao, Y.; Xia, X.-X.; Zhao, X.-Q.; Peng, L.; Srinophakun, P.; Bai, F.-W. Cellulosic Ethanol Production: Progress, Challenges and Strategies for Solutions. *Biotechnology Advances* **2019**, *37*, 491–504, doi:10.1016/j.biotechadv.2019.03.002.
- Vu, H.P.; Nguyen, L.N.; Vu, M.T.; Johir, M.A.H.; McLaughlan, R.; Nghiem, L.D. A Comprehensive Review on the Framework to Valorise Lignocellulosic Biomass as Biorefinery Feedstocks. *Science of The Total Environment* **2020**, *743*, 140630, doi:10.1016/j.scitotenv.2020.140630.