

Supplementary Information

1. Experimental

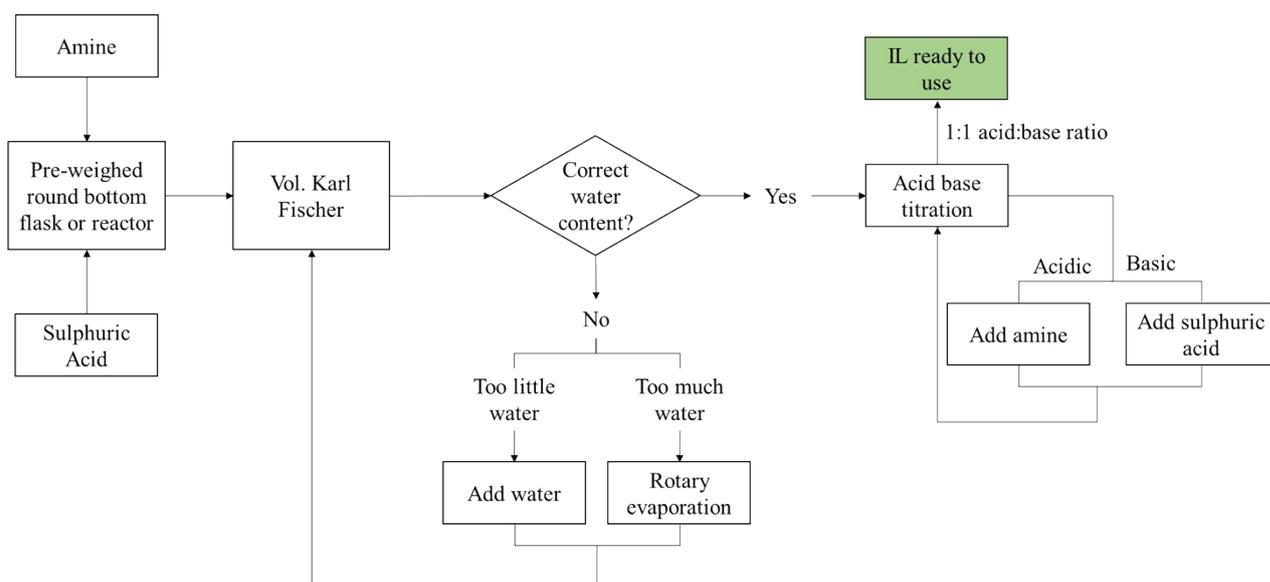


Figure S.1. General schematic for ionic liquid synthesis, in this case a protic HSO₄ ionic liquid such as [TEA][HSO₄] or [Ch][HSO₄]

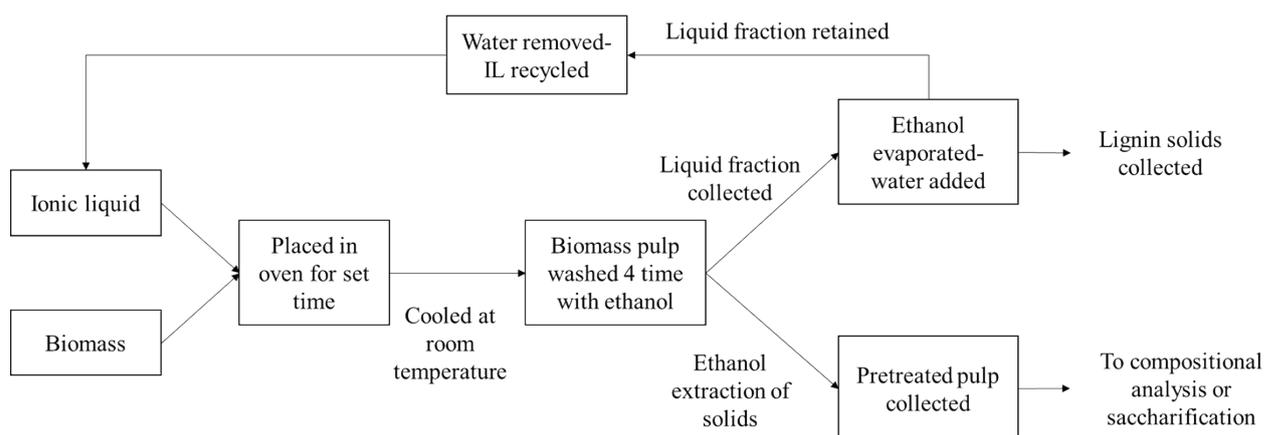


Figure S.2. Schematic for the IonoSolv pretreatment

1.1 Rice Straw Grinding and Moisture Content

Each of the rice straw varieties were milled using the Wiley Mill Retsch SM 2000 then sieved. Particles were between 180-850µm in size for pretreatment, except for Rc 25 which turned into a powder upon milling. Moisture content was determined following National Renewable Energy Laboratory (NREL) protocols (2). Specifically, approximately 1g of rice straw sample was placed onto pre-weighed aluminum foil and weighted to determine $m_{air\ dried}$. The folded foil containing the biomass was placed in an oven at 105 °C to dry overnight. Once removed, the sample was placed in a desiccator to cool for 15 minutes prior to weighing to measure $m_{oven\ dried}$. The moisture content is determined by:

$$moisture_{ovendried}(\%) = \frac{m_{air\ dried} - m_{oven\ dried}}{m_{oven\ dried}} \times 100 \quad (S1)$$

1.2 Ionic Liquid Synthesis

Food-grade ionic liquid [Ch][HSO₄] and non food-certified ionic liquid [TEA][HSO₄] was synthesized with a 20% water weight. The synthesis workflow is generalised in Figure S.1. A reactor was utilised for the synthesis of [TEA][HSO₄], which mechanically and simultaneously fed in the reagents (5M sulfuric acid with 99% triethylamine), stirred, and cooled the contents. The reactor was connected to a computer that monitored the progress of the reaction via conductivity of the output; however, water content was not controlled during this process and evaporation of the water in the synthesised ionic liquid was required. Once synthesis was completed, the ionic liquid was transferred from the reactor to a pre-weighed container and a rotary evaporator was utilised to remove the excess water.

Manual synthesis of [Ch][HSO₄] was conducted via the combination of choline hydroxide with 5M sulfuric acid. A calculated quantity (as well as known weight) of 5M sulfuric acid was placed into a separatory funnel. The choline hydroxide was placed into a 1000 mL round bottom flask (RBF) with a stir bar. The weight of the flask and magnetic stir bar was recorded prior to its placement in a bowl containing ice (which was used as a heat sink for the cooling of these exothermic reactions). The separatory funnel was hung over the opening of the RBF and allowed to drip slowly into the contents of the RBF. The entire process was conducted in a fume hood. Once the acid had finished dripping, the separatory funnel was rinsed with distilled water to ensure all contents were flushed into the RBF. The synthesised ionic liquid was left to stir for several hours in the ice bath. Whether synthesised manually or via the ionic liquid reactor, a syringe extracted 0.1 mL of the sample for Karl Fischer volumetric titration to determine water content of the sample. A rotary evaporator eliminated water from the ionic liquid and titration measurements were taken. The evaporation process was repeated until the [TEA][HSO₄] or [Ch][HSO₄] was at 20% water weight. A 0.1 mL sample was extracted and submitted for titration using the compact titrator to determine the acid-base ratio. If the sample was too acidic, the appropriate amount of amine was added to bring the ratio to balance and if the sample was too basic the appropriate amount of sulfuric acid was calculated and added. The sample was then titrated again, and adjustments made until the 1:1 ratio was achieved.

1.3 F. *Venenatum* Elemental Analysis

The fermentation broth samples were freeze-dried for 2 days in a vacuum at less than 0.01 bar at room temperature. The freeze-dried samples were sent for elemental analyses by OEA Laboratories Ltd. The elemental analyses for carbon (C), hydrogen (H), nitrogen (N) and Sulphur (S) were performed using an automatic combustion elemental analyser (Thermoquest EA1110 elemental analyser).

Milligram sub-samples are weighed on a microbalance into tared tin capsules with a small quantity of vanadium pentoxide which serves as a combustion aid for sulphur. The capsules are sealed and loaded into the elemental analyser autosampler carousel. The autosampler purges the sample chamber with oxygen to exclude air and provides a means of introducing the sample into the reaction furnace tube without pressure loss.

A controlled flow of helium as carrier gas is maintained through the elemental analyser analytical circuit. When the elemental analyser sequence is started, the helium carrier is replaced briefly by a measured dose of high purity oxygen. After a few seconds the oxygen dose arrives at the combustion zone in the upper half of the combustion tube operating at 1000°C. The autosampler drops the encapsulated sample into the combustion tube to meet the oxygen. The sample is burned instantaneously followed by the oxidation of the tin capsule giving a localised temperature in excess of 1800°C for a few seconds (flash combustion). The

resulting combustion gases are carried over a catalyst (tungstic oxide) and high purity reactive copper in the lower half of the reaction tube. These materials ensure complete oxidation of the gases, remove excess oxygen and other gaseous interferences and reduce any oxides of nitrogen that might have been formed.

A gas chromatography (GC) column separates and elutes the gaseous constituents into four peaks - elemental nitrogen (nitrogen), carbon dioxide (carbon), water vapour (hydrogen) and sulphur dioxide (sulphur) which are quantified by a thermal conductivity detector (TCD). The peak responses are identified and integrated by the elemental analyser software and compared to known reference compounds traceable to international standards.

Total oxygen (O) trace level in freeze dried samples is determined using an automatic elemental analyser. Milligram sub-samples are weighted and introduced into the pyrolysis furnace tube without pressure loss. A controlled flow of helium is maintained through the elemental analyser analytical circuit. When the elemental analyser sequence is started the autosampler drops the encapsulated sample into the pyrolysis tube operating at 1060°C. The sample is instantaneously pyrolysed. The resulting pyrolysed gases are carried over a catalyst (nickelised carbon granules) in the lower half of the combustion tube. The catalyst ensures complete conversion of any oxygen gases into carbon monoxide. A GC column separates and elutes the CO which is quantified by TCD. The peak response is identified and compared to known reference compounds traceable to international standards.

2. Economic Evaluation

There are multiple steps involved in the economic analysis of the process. These are described in Figure S.3. and a description of the corresponding sections is discussed below.

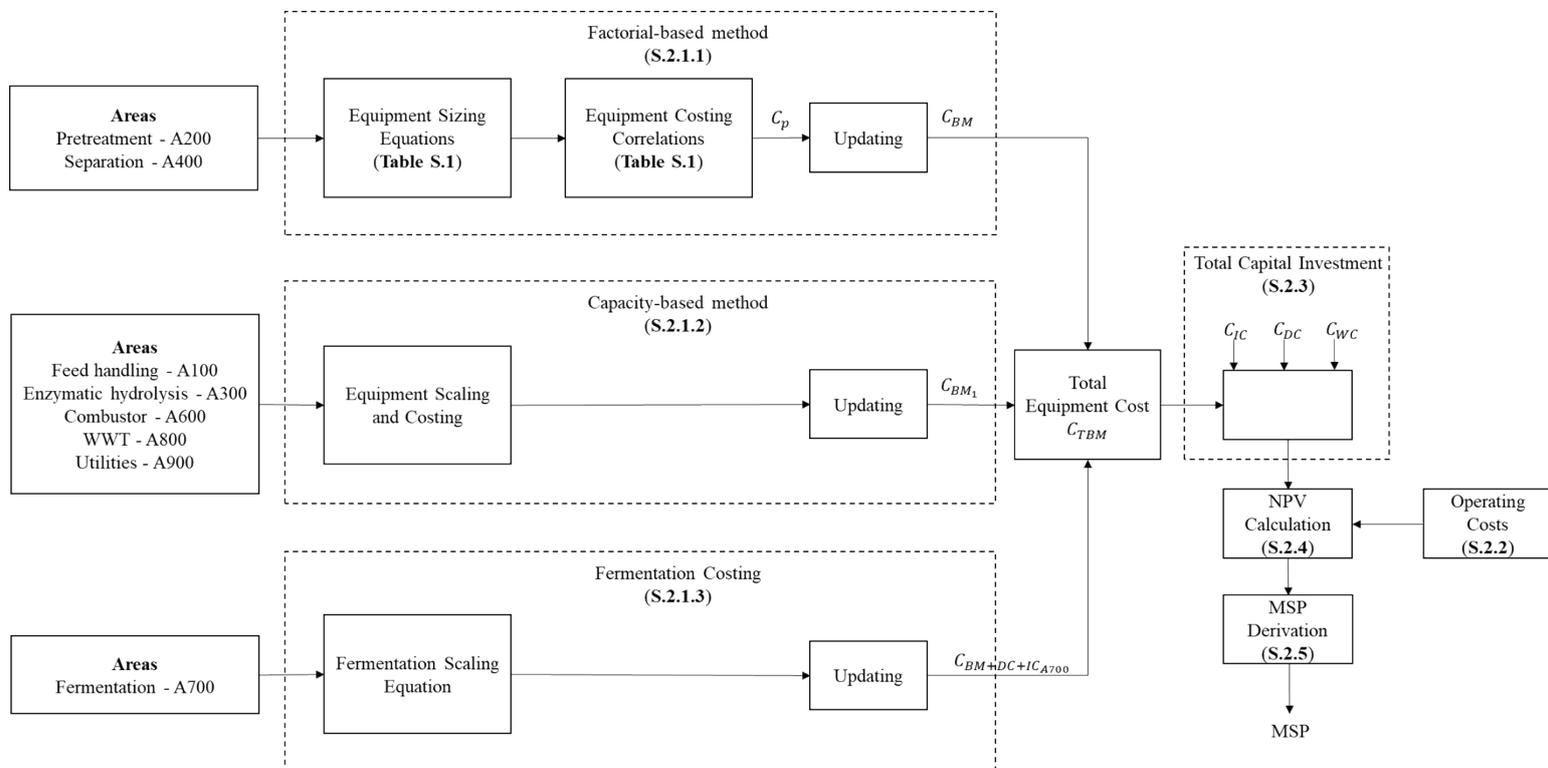


Figure S.3. Summary of economic evaluation methodology and their corresponding sections where the methodology is described in further detail. Updating in all cases refers to altering the cost of the equipment to reflect the general inflation of equipment prices as time passes. In each case, the cost correlations or equipment quotes are for a given year, therefore, their costs must be updated appropriately to the present time for fair comparison.

2.1. Equipment Sizing and Costing

To determine capital cost of the process, different methods were used. Factorial-based methods (S.2.1.1) were used for original design used in this work (Pretreatment (area A200) and separation (area A400)). Feed handling, enzymatic hydrolysis, the combustor, WWT, and some utility capital costs (areas A100, A300, A600, A800 and A900 respectively) were costed via capacity scaling (S.2.1.2) with known costs from Davis, Tao (4). Finally, the fermentation process (area A700) was costed in a similar capacity-based method (S.2.1.3), however, this was based on publicly available information regarding fermentation costs(1).

2.1.1. Factorial-based methods

For Areas 200 and A400 factorial-based methods were used to size and cost equipment. The costing equations are provided by Seider, Lewin (5) and are provided in Table S.1. First, the equipment is sized through appropriate design equations in order to determine the required size factor for the factorial-based costing equations. The costing equations relate this size factor, S , to the equipment base purchase cost C_B for the given year of publication of the correlations (Table S.1). Typically, this base cost represents the equipment cost using carbon-steel. Adjustments are then made to C_B to account for other design factors, such as a selected material that is different to the base material, in which case a material factor (F_m) is used to account for this (S2). The resultant cost is the f.o.b purchase cost, C_p which is the purchase cost of the selected piece of equipment to the required specifications in the year the correlations were produced, but does not account for other expenses such as instrumentation and piping, or transportation and installation costs. The f.o.b purchase cost is then updated to the current year through scaling of indices (I_{year} – CE index (Chemical Engineering Plant Cost Index)), and multiplied by the bare module factor (F_{BM}) to obtain the bare-module cost (C_{BM}) for the current year (S3). C_{CBM} is then the cost of the delivered and installed equipment on-site. The CE index for 2019 was used which was 603.

$$C_p = F_m C_B \quad (S2)$$

$$C_{BM} = C_p \left(\frac{I_{CE - Current\ year}}{I_{CE - Base\ year}} \right) F_{BM} \quad (S3)$$

Supplementary Information

Table S.1. Sizing and costing equations used as part of the factorial-based method of equipment evaluation found in (5). C_p and C_v refer to the f.o.b purchase cost. S in costing correlations represents a non-typical size factor. F^M is the material factor which adjusts the f.o.b cost to account for a change in construction material.

Equipment	Values from Aspen	Sizing Equations	Costing Correlations
Mixers	Volumetric Flowrate, Q	$V = Q * t_{hold\ up}$ $D = \frac{4V}{\pi AR^3}$ $H = D * AR$ $AR = 5$ $W = \pi(D_i + t_s)(L + 0.8D_i)t_s\rho$ $\rho = 490 \frac{lb}{ft^3}$ $t_s = \frac{P_d D_i}{2SE - 1.2P_d}$ $P_d = \exp[0.60608 + 0.91615[\ln(S)] + 0.0015655[\ln(S)]^2]$	<p>Horizontal (1,000 < W < 920,000 lb)</p> $C_v = \exp\{5.6336 + 0.4599[\ln(W)] + 0.00582[\ln(W)]^2\}$ <p>(3 < D_i < 12ft)</p> $C_{pL} = 2275(D_i)^{0.2094}$ <p>Vertical (4,200 < W < 1,000,000 lb)</p> $C_v = \exp\{7.1390 + 0.18255[\ln(W)] + 0.02297[\ln(W)]^2\}$ <p>(3 < D_i < 21ft) and 12 < L < 40ft</p> $C_{pL} = 410(D_i)^{0.73960}(L)^{0.70684}$ $C_p = F^M C_v + C_{pL}$
Pressure Vessels	Flowrate out of reactor	$V = Q \times \tau$ $W = \pi(D_i + t_s)(L + 0.8D_i)t_s\rho$ $\rho = 490 \frac{lb}{ft^3}$	<p>Horizontal (1,000 < W < 920,000 lb)</p> $C_v = \exp\{5.6336 + 0.4599[\ln(W)] + 0.00582[\ln(W)]^2\}$ <p>(3 < D_i < 12ft)</p> $C_{pL} = 2275(D_i)^{0.2094}$ <p>Vertical (4,200 < W < 1,000,000 lb)</p> $C_v = \exp\{7.1390 + 0.18255[\ln(W)] + 0.02297[\ln(W)]^2\}$ <p>(3 < D_i < 21ft) and 12 < L < 40ft</p> $C_{pL} = 410(D_i)^{0.73960}(L)^{0.70684}$ <p>Towers (9,000 < W < 2,500,000 lb)</p> $C_v = \exp\{10.5449 - 0.4672[\ln(W)] + 0.05482[\ln(W)]^2\}$ <p>(3 < D_i < 24ft) and 27 < L < 170ft</p>

Supplementary Information

			$C_{PL} = 341(D_i)^{0.63316}(L)^{0.80161}$ $C_p = F_M C_V + C_{PL}$
Distillation Columns	Column Diameter	$H = D * PS$ $PS = \text{Plate Spacing} = 0.6m$ $W = \pi(D_i + t_s)(L + 0.8D_i)t_s\rho$ $\rho = 490 \frac{lb}{ft^3}$ $t_s = \frac{P_d D_i}{2SE - 1.2P_d}$ $P_d = \exp[0.60608 + 0.91615[\ln(P_o)] + 0.0015655[\ln$	<p>Towers ($9,000 < W < 2,500,000 \text{ lb}$)</p> $C_v = \exp\{10.5449 - 0.4672[\ln(W)] + 0.05482[\ln(W)]^2\}$ <p>($3 < D_i < 24ft$) and $27 < L < 170ft$</p> $C_{PL} = 341(D_i)^{0.63316}(L)^{0.80161}$
Decanters	Density of Continous Phase, Density of Dispersed phase. Mass Flow, volumetric flowrates	$t_{separation} = \frac{100\mu}{\rho_{dispersed} - \rho_{continuous}}$ $V = \frac{t_{separation}Q}{0.95}$ $D = \frac{16V^{\frac{1}{3}}}{5\pi}$ $L = AR * D$ $t_s = \frac{P_d D_i}{2SE - 1.2P_d}$ $P_d = \exp[0.60608 + 0.91615[\ln(P_o)] + 0.0015655[\ln$	<p>Horizontal ($1,000 < W < 920,000 \text{ lb}$)</p> $C_v = \exp\{5.6336 + 0.4599[\ln(W)] + 0.00582[\ln(W)]^2\}$ <p>($3 < D_i < 12ft$)</p> $C_{PL} = 2275(D_i)^{0.2094}$

Supplementary Information

		$W = \pi(D_i + t_s)(L + 0.8D_i)t_s\rho$ $\rho = 490 \text{ lb/ft}^3$	
Flash Vessels	Vapour density, liquid (bottoms) density, Liquid volumetric flowrate, vapour volumetric flowrate	$u_{\text{settling}} = 0.07 \frac{(\rho_L - \rho_v)}{\sqrt{\rho_v}}$ $D_{\text{min}} = \sqrt{\left(\frac{4V_v}{\pi \times u_{\text{settling}}}\right)}$ $V_{\text{hold up}} = V_L * t_{\text{hold up}}$ $h_L = \frac{V_{\text{hold up}}}{\pi \frac{D^2}{4}}$ $H = 0.4 + \max(D_v, 1) + \max\left(\frac{D_v}{2}, 0.6\right) + h_L$ $W = \pi(D_i + t_s)(L + 0.8D_i)t_s\rho$ $\rho = 490 \text{ lb/ft}^3$ $t_s = \frac{P_d D_i}{2SE - 1.2P_d}$ $P_d = \exp[0.60608 + 0.91615[\ln(P_o)] + 0.0015655[\ln(P_o)]^2]$	<p>Vertical ($4,200 < W < 1,000,000 \text{ lb}$)</p> $C_v = \exp\{7.1390 + 0.18255[\ln(W)] + 0.02297[\ln(W)]^2\}$ <p>($3 < D_i < 21 \text{ ft}$) and $12 < L < 40 \text{ ft}$)</p> $C_{PL} = 410(D_i)^{0.73960}(L)^{0.70684}$
Heat Exchangers	Heat duty, Hot stream temperatures in and out, cold stream temperatures in and	$LMTD = \frac{(T_{h,in} - T_{c,out}) - (T_{h,out} - T_{c,in})}{\ln\left[\frac{T_{h,in} - T_{c,out}}{T_{h,out} - T_{c,in}}\right]}$	<p>Fixed head heat exchanger ($150 \leq A \leq 12,000 \text{ ft}^2$)</p> $C_B = \exp\{11.4185 - 0.9228[\ln(A)] + 0.0986[\ln(A)]^2\}$ $C_p = F_p F_M F_L C_B$

Supplementary Information

	out.	$A = Q \times U \times LMTD$	$F_M = a + \left(\frac{A}{100}\right)^b$ <p>For $100 \leq P \leq 2,000 \text{ psig}$</p> $F_p = 0.9803 + 0.018\left(\frac{P}{100}\right) + 0.0017\left(\frac{P}{100}\right)^2$ <p>Double-Pipe heat exchangers ($2 \leq A \leq 200 \text{ ft}^2$)</p> $C_B = \exp\{7.2718 + 0.16[\ln(A)]\}$ $C_p = F_p F_M C_B$ <p>For $600 \leq P \leq 3,000 \text{ psig}$</p> $F_p = 0.8510 + 0.1292\left(\frac{P}{600}\right) + 0.0198\left(\frac{P}{600}\right)^2$
Pumps	Volumetric Flowrate through pump, Work.		<p>Centrifugal pump ($10 \leq Q \leq 5,000 \text{ gpm}$)</p> $S = Q(H)^{0.5}$ <p>Q = flow rate in gpm, H = pump head in ft</p> $C_B = \exp\{12.1656 - 1.1448[\ln(S)] + 0.0862[\ln(S)]^2\}$ $C_p = F_T F_M C_B$ <p>Electric Motors ($1 \leq P_c \leq 700 \text{ Hp}$)</p> $C_B = \exp\{5.9332 + 0.16829[\ln(P_c)] - 0.110056[\ln(P_c)]^2 + 0.0000001[\ln(P_c)]^3\}$ $C_p = F_T C_B$
Washing	Solids mass flowrate into washer. Number of washing stages.		<p>Roll Press ($150 \leq \dot{m}_s \leq 12,000 \text{ lb/hr}$)</p> $C_p = \exp\{10.9807 - 0.4467[\ln(F)] + 0.06136[\ln(F)]^2\}$ <p>*Each C_p represents the cost of one washing stage. In the process, 4 washing stages are required.</p>
Filter	Filtrate flowrate	$A = \frac{\dot{m}_{\text{Filtrate}}}{FR (1500 \text{ lb/day.ft}^2)}$	<p>Rotary-drum vacuum filter ($100 \leq A \leq 800 \text{ ft}^2$)</p> $C_p = \exp\{11.796 - 0.1905[\ln(A)] + 0.0554[\ln(A)]^2\}$ <p>Pressure leaf filter ($30 \leq A \leq 100 \text{ ft}^2$)</p> $C_p = 1385A^{0.71}$

Supplementary Information

Thickener	Solids flowrate	$A = C_1 * \frac{\text{tons solids}}{\text{day}}$ $C_1 = 10$	<p>Thickener, concrete ($8000 \leq A \leq 125,000 \text{ ft}^2$) $C_p = 2720A^{0.58}$</p> <p>Thickener, steel ($80 \leq A \leq 8,000 \text{ ft}^2$) $C_p = 3810A^{0.58}$</p>
Vacuum System	Vapour Flowrate Pressure (torr), Vapour and liquid density (kg/m3), Vapour and liquid volume flowrate	$u_{\text{settling}} = 0.07 \frac{(\rho_L - \rho_v)}{\sqrt{\rho_v}}$ $D_{\text{min}} = \sqrt{\left(\frac{4V_v}{\pi \times u_{\text{settling}}} \right)}$ $V_{\text{hold up}} = V_L * t_{\text{hold up}}$ $\text{Air Leakage} \left(\frac{\text{ft}^3}{\text{min}} \right) = (5 + \{0.0298 + 0.03088[\ln(P)] -$	<p>Liquid-ring pumps ($50 \leq S \leq 350 \text{ ft}^3/\text{min}$) $C_p = 8,250S^{0.37}$</p>

2.1.2. Capacity-based method

For areas A100, A300, A600, A800 and A900, the equipment costs were scaled by capacity according to the costs in (6). Equation S4 is used to cost the combustor, WWT, enzymatic hydrolysis and some utility capital costs, based on existing estimates of these costs.

$$C_{BM1} = C_{BM2} \left(\frac{Capacity_2}{Capacity_1} \right)^n \left(\frac{I_1}{I_2} \right) IF \quad (S4)$$

Where C_{BM1} represents the cost of the equipment in this study and C_{BM2} represents the equipment cost of the base scenario, which in this case is found in (6). Capacity₁ and Capacity₂ are the process flows/streams that the equipment is scaled on for this work and the base-case respectively. I_1 and I_2 are the Chemical Engineering Plant Index (CE) for the present year and base year respectively. The CE index for 2019 was used which is 603. IF is the installation factor of a particular piece of equipment and varies between types of equipment. n is the scaling exponent, which takes a different value depending on the equipment being scaled.

2.1.3. Fermentation Costing

The fermentation hierarchy (Area 700) was costed through the scaling method similar to Equation S4. Data from Moore, Robson (1) was used to determine the cost for a certain capacity of mycoprotein production, in which, in 1997 a total investment of \$97.5m for two fermenters providing between 10,000-13,000 additional capacity. Therefore, the specific scaling equation for fermentation costing is:

$$C_{BM + DC + IDCA700} = 97.5 \left(\frac{P_{capacity}}{12500} \right)^{0.6} \left(\frac{I_1}{I_2(1997)} \right) \quad (S5)$$

Where $C_{BM + DC + IDCA700}$ is the bare-module cost of all the equipment in the fermentation area, $P_{capacity}$ is the production capacity of lignocellulosic-mycoprotein, I_1 and I_2 are the CE index for the present year (2019 = 603) and 1997 respectively, and 12500 refers to the capacity of the two fermenters built in 1997. $C_{BM + DC + IDCA700}$ includes already the direct and indirect costs associated with this equipment due to the cost of the current fermenters being the overall investment costs. Therefore, in Supplementary S.2.3, the fermentation area (A700) is excluded when determining the direct and indirect costs.

2.1.4. Utility Costing

Process water and electricity were estimated via the procedure defined by Ulrich and Vasudevan (7) in which a two-factor cost equation is used to account for both inflation and energy costs (S6).

$$C_{S,u} = a (CEPCI) + b (C_{S,f}) \quad (S6)$$

Where $C_{S,u}$ is the utility price, $C_{S,f}$ denotes the fuel price (\$/GJ), a and b are utility cost coefficients, and $CEPCI$ adjusts for inflation. a and b (Table S.2) are taken from Ulrich and Vasudevan (7). $CEPCI$ was defined as 603 for the year 2019. Natural gas was chosen for fuel with an associated $C_{S,f}$ of 8.11 \$/GJ (8).

Table S.2. Utility cost coefficients based on the costing method described in Ulrich and Vasudevan (7)

Component	Cost Coefficients	
	a	b
Electricity	1.3×10^{-4}	0.010
Process Water	$7.0 \times 10^{-5} + 2 \times 10^{-6} q^{-0.6}$	0.003
Refrigerant ($T = 263 K$)	$0.5Q_c^{-0.9}(T^{-3})$	$1.1 \times 10^6(T^{-5})$

Costs relating to disposal of flu ash were assumed to cost \$28.86/tonne(4).

2.2. Operating Costs

After the fixed costs were found, the variable operating costs were then estimated. A few different aspects contribute to the overall production cost of the final product. The main categories are feedstock costs, utility costs, labour-related operations, maintenance, operating overhead, property taxes and insurance, depreciation and general expenses. These categories are further detailed in Table S.3 and the assumed calculation of these is provided. Once the sub-components of these categories are calculated, the overall production costs can be found.

Table S.3. Costing sheet for annual production costs of SCP pastes. Operating overhead (OO), and general expenses (GE) are non-specific and taken from(5). Maintenance (M), depreciation (D) and property taxes and insurance (PTnI) are adjusted in line with the operating assumptions in (6). Operations (O) is determined via the methodology described in S.2.2.1.

Cost Factor	Assumed Factor
Raw Materials (RM)	
Biomass	\$0.049 /kg
[Ch][HSO ₄]	\$1 /kg
[TEA][HSO ₄]	\$1.24 /kg (9)
Ammonia	\$0.50 /kg
Cellulase	\$6.27 /kg
Lime	\$0.2 /kg (6)
Nutrient	\$0.747 /kg (Supplementary S.2.2.2)
Caustic Soda	\$0.15 /kg (6)
Utilities (U)	
Cooling water	Built into the model
Process water	See utility costing methodology (S.2.1.4)
Refrigerant	See utility costing methodology (S.2.1.4)
Electricity	See utility costing methodology (S.2.1.4)
Landfill	\$0.17 /drykg
Operations (O)	
Total labour	\$2,920,000 (Supplementary S.2.2.1)
Maintenance (M)	
Total maintenance	3% of ISBL
Operating overhead (OO)	
General plant overhead	7.1% of Maintenance (M) & Operations (O)
Mechanical department services	2.4% of M&O
Employee relations department	5.9% of M&O
Business services	7.4% of M&O
Property taxes and insurance (PTnI)	
	0.7% of C_{TDC}
Depreciation	
Direct plant	3% of C_{TDC}

General expenses (GE)

Selling expense	3% of sales
Direct research	4.8% of sales
Allocated research	0.5% of sales
Administrative expense	2.0% of sales
Management incentive compensation	1.25% of sales

^a See appendix S.8;

2.2.1. Labour Cost Assumptions

Labour costs were estimated from the current Quorn process and estimated personnel required in the NREL biochemical model. 600 employees were reported in 2010 (10) for 20,000 tonnes/year mycoprotein production, which would cover the employees required for the downstream process. In the NREL biochemical model, the bioethanol facility requires 50 employees (excluding on-site enzyme production employees) for 773,000 dry US. ton/year. The NREL model features similar infrastructure to the lignocellulosic-paste process, and therefore, the same type of labour was assumed to cover the upstream part of the process. The number of employees was scaled according to the capacity of feedstock of mycoprotein in the upstream or downstream areas respectively (Equations S7 and S8). UK average salary was £35,058 in February 2019, which is \$45,575 (conversion factor £1:\$1.30) (11).

$$Labour_{upstream} = 50 \times \left(\frac{Feedstock \left(\frac{\text{tonnes}}{\text{year}} \right)}{773,000 \frac{\text{tonnes}}{\text{year}}} \right)^{0.6} \quad (S7)$$

$$Labour_{downstream} = 600 \times \left(\frac{Mycoprotein Production \left(\frac{\text{tonnes}}{\text{year}} \right)}{20,000 \frac{\text{tonnes}}{\text{year}}} \right)^0 \quad (S8)$$

2.2.2. Nutrient Make-up and costing

Without detailed knowledge of nutrient medium composition for mycoprotein fermentation, available elemental analysis data taken from Moore, Robson (1) (Table S.4) combined with our own elemental analysis of the macro-elements of fermentation broth was used to

Table S.4. Approximate elemental composition of *F. Venenatum* showing the minor and trace elements that make up ~4% of the biomass as shown in Moore, Robson (1)

Element	Composition /%
Potassium	20.0
Sodium	N/A
Calcium	0.8
Magnesium	1.8
Iron	0.05
Copper	0.04
Manganese	0.12
Zinc	0.28

stoichiometrically define the minimum nutrient requirements.

The overall compounds considered to make-up part of the nutrient medium are shown in Table S.5. The compounds that were considered to supply the elements to the biomass were

Supplementary Information

primarily based from those given by Moore, Robson (1) (Table S.4), with any missing compounds taken from those used in Vogel's medium (3).

In specifying the phosphoric acid demand, the phosphorus content of the biomass was used to work out the stoichiometric molar demand of phosphorus, which was then back calculated to give a phosphoric acid mass requirement. A similar procedure was used for the minor and trace elements that account for the 3.3% of unaccounted mass in the elemental analysis that we conducted. However, they were calculated post-simulation. The mass of each compound in was then calculated by finding the mass of each chemical required to supply enough of each element for the formation of the biomass. The amount of each compound supplied per tonne of accounted (dry) biomass (from the elemental analysis) is shown in column 4 of Table S.5.

Although our elemental analysis included the sulfur content, the biomass was defined in Aspen plus without accounting for the sulfur. This decision was made to facilitate the calculation for the demand of the different nutrient compounds. Because a number of compounds in Table S.5 supply sulfates to the biomass, it was then assumed and verified that the sulfur demand in the biomass was met.

Table S.5. Assumed compounds used as part of the overall nutrient profile for *F. Venenatum* fermentation. Compounds were taken from the information available at Moore, Robson (1), for compounds that were still missing, Vogel's medium compounds were used to supply the required element (3).

Compound	Formula	Cost \$/kg	Demand (kg/t accounted dry biomass)
Phosphoric acid ^a	H ₃ PO ₄	0.7	^a
Potassium sulfate ^b	K ₂ SO ₄	1.23	42.5
Calcium chloride dihydrate ^b	CaCl ₂ ·2H ₂ O	0.15	2.80
Magnesium sulfate heptahydrate ^b	MgSO ₄ ·7H ₂ O	0.2	17.4
Ammonium iron(II) sulfate hexahydrate ^b	Fe(NH ₄) ₂ (SO ₄) ₂ ·6H ₂ O	1	0.335
Copper sulfate pentahydrate ^b	CuSO ₄ ·5H ₂ O	2	0.150
Manganese(II) sulfate ^b	MnSO ₄	0.4	0.352
Zinc sulfate heptahydrate ^b	ZnSO ₄ ·7H ₂ O	1	1.17

^a Phosphoric acid was defined separately in Aspen plus and featured in the stoichiometric equation that defined the fermentation reactor in Aspen Plus; Flow amount was calculated in Aspen plus directly.

^b All these compounds were group did not feature in the Aspen plus nutrient stream to the fermentation reactor, instead the amounts were calculated post simulation and a price of the nutrient stream as a whole was deduced.

Using this method, Total nutrient cost for the scenarios was \$0.747 /kg

2.3. Total Capital Investment

Once the cost of all pieces of equipment was found, the total capital investment C_{TCI} of the process could be determined. Firstly, the C_{BM} of all equipment was then summed to give the total bare module cost C_{TBM} (S9).

Supplementary Information

$$C_{TBM} = \sum_{i=1}^N C_{Bmi} \quad (S9)$$

After computing the equipment bare-module costs, the total direct costs, C_{DC} , were computed. These costs account for warehouse costs (4%), site development (9%), and additional piping (5%) (6). Typically, these costs would only be attributed to the main process sections (inside-battery-limits (ISBL)) which include pretreatment, enzymatic hydrolysis, separation and fermentation. However, because of the way that the fermentation area (A700) is costed, where total investment costs were scaled (S.2.1.3), it is assumed that additional direct costs relating to fermentation are already accounted for. Therefore, when determining the direct costs of the process, the ISBL is modified to not include the fermentation area. Therefore, C_{DC} is determined as per equation S10.

$$C_{DC} = 0.18C_{TBMISBL - A700} \quad (S10)$$

Where $C_{TBMISBL - A700}$ is the total bare module cost of all equipment considered in the ISBL not including fermentation (A700). The indirect costs, C_{IC} , were assumed to be 60% of C_{DC} which account for proratable expenses, field expenses, home office and construction fee, project contingency and other costs(6) (S11).

$$C_{IC} = 0.60C_{DC} \quad (S11)$$

From this, the Fixed Capital Investment (Total Depreciable Capital (C_{TDC})) can be found by summing the direct and indirect costs (S12)

$$C_{TDC} = C_{IC} + C_{DC} \quad (S12)$$

Assuming a working capital (C_{WC}) of 17.6% of C_{TDC} , The Total Capital Investment, C_{TCI} , is given by equation S13.

$$C_{TCI} = 1.176C_{TDC} \quad (S13)$$

2.4. Net Present Value (NPV) Calculation

Before calculating the MSP, the NPV of the ‘upstream’ process is required. For calculating the NPV, both fixed capital costs and annual operating costs are used. The NPV was calculated for a 30-year plant life. The net earnings (NE) and annual cash flow (ACF) for each year of plant operations were calculated from Equations S14 and S15 respectively.

$$NE = (S - C_{Ex. D} - D) \times (1 - ITR) \quad (S14)$$

$$ACF = (NE + D) - fC_{TDC} - C_{WC} \quad (S15)$$

$$PV = DF \times ACF \quad (S16)$$

$$NPV = \sum_i^{30} PV_i \quad (S17)$$

Where, S is the annual sales \$/yr, $C_{Ex. D}$ is the production cost less annual depreciation \$/yr, D is the annual depreciation \$/yr, ITR is the income tax rate, C_{TDC} is the total depreciable

capital, and C_{WC} is the working capital. The cash flow for each year is then discounted by the discount rate (0.1) and a cumulative Present Value (PV) is found (S16), from which the NPV, the sum of PV over the plant life, is calculated (S17).

2.5. Minimum Selling Price (MSP) Derivation

The MSP is defined as the selling price of the protein product for which the NPV is 0(4). Two MSPs were calculated. The first was the MSP of the protein paste (herein defined as MSP_{paste}), which is the product of the biorefinery process (upstream) modelled in this paper and was calculated via an iterative procedure to minimise the upstream process NPV. The second MSP ($MSP_{product}$) calculated was the final price of the product as sold at the company gate to external customers (i.e. the texturised product such as Quorn mince). This was calculated by considering approximate overall downstream processing costs and associating a price per kg to ‘upgrade’ the protein paste to the finished product, and then adding this to the aforementioned MSP_{paste} . Figure S.4 visualises these economic distinctions.

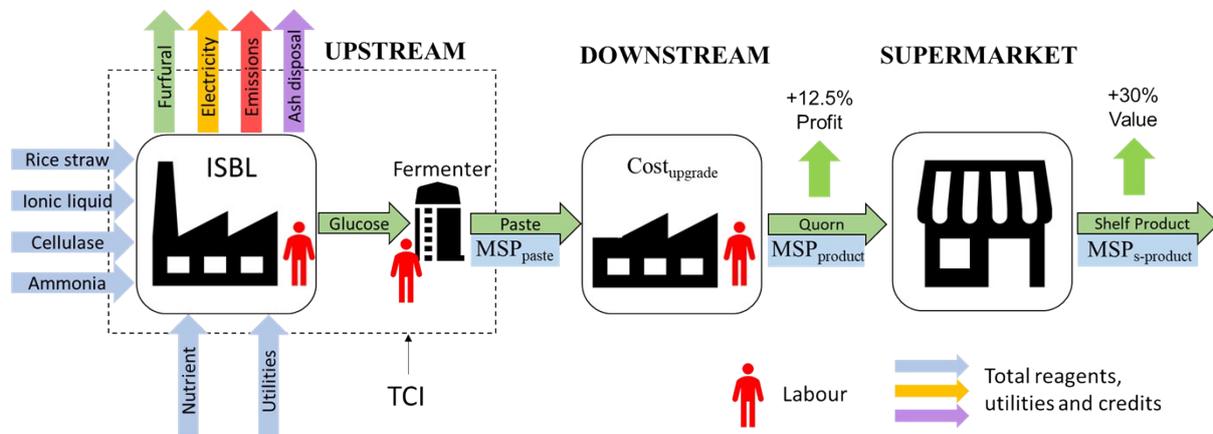


Figure S.4. Boundaries considered as part of economic comparison to power-to-protein process

The ‘upstream’ (accounted-for) part of the process converts initial feedstock (rice straw) to the paste, and the ‘downstream’ process, processes the paste to the texturised product. Labour cost is scaled and is split between upstream and downstream, with the upstream labour based on the estimates from NREL to cover a bioethanol facility which shares infrastructure with the upstream portion of this process. Downstream labour is based from reported quorn employee figures (10) (S.2.2.1).

The upstream MSP_{paste} is calculated through finding the price of paste at which the NPV is 0 for the upstream process (i.e. the units modelled in Aspen Plus). The downstream upgrading cost is calculated from dividing the proportion of the annual revenue estimated to cover downstream processing costs (as defined in the economic model of Molitor, Mishra (12)) and the downstream labour cost, by the annual production capacity. The resulting price is an approximate cost ($\$/kg$ mycoprotein paste) for processing the paste to the final product ($Cost_{upgrade}$). The processing cost was then scaled according to processing capacity by Equation S18.

$$Cost_{upgrade1} = \left(\frac{Capacity_1}{Capacity_2} \right)^{0.6} \left(\frac{I_{year_1}}{I_{year_2}} \right) (Cost_{labour} + Cost_{downstream}) \quad (S18)$$

Supplementary Information

Where $Cost_{upgrade1}$ is the upgrade price for the conversion of mycoprotein paste to final product. $Capacity_1$, and ($Capacity_2 = 20,000 \text{ tonnes/year}$) are the downstream processing capacity in the current scenario and base case scenario respectively, I_{year1} and I_{year2} are the CE index values for the 2019 and base case respectively (2014), and $Cost_{labour}$ and $Cost_{downstream}$ (\$127m) are the downstream labour costs and proportion of annual revenue to cover downstream processing costs respectively for the base case scenario.

The total capital investment (TCI) includes all costs of the equipment upstream including the fermenter.

Once both the MSP_{paste} , and $Cost_{upgrade}$ were found, the two were added together to obtain the $MSP_{product}$. 12.50% was added to this cost at the downstream processing exit gate to allow for profit. Further to this, 30% is assumed to be added throughout the supermarket portion of the value chain. This final calculated price is then the minimum selling price of the mycoprotein product at the supermarket ($MSP_{s-product}$), which achieves satisfactory returns for Quorn and the supermarket, which can therefore be compared to current production and other protein sources.

2.6. Current Predicted Minimum Selling Price

Table S.6. Inputs and assumptions for the prediction of the current minimum selling (production) price of mycoprotein paste.

Raw Materials and Utilities					
Inputs	kg/kg Paste	\$/kg	\$/kg paste	\$/year	Notes
Glucose	1.0999	0.6	0.6599	30,893,757	Glucose requirement from (10)
Ammonia	0.0449	0.2	0.0090	362,492	Stoichiometric NH_3 demand with 16% excess
Nutrient	0.0648	0.747	0.0484	1,954,608	Supplementary S.2.2.2
H2O	4.6324	0.0005	0.0023	92,608	Water required to dilute glucose demand to 10% glucose concentration with 50% recycle rate (12).
Electricity	3.4214	0.0995	0.3404	13,746,808	Assumed 2.75kWh/kg paste produced for aeration of fermenter, with this covered 50% of total plant-wide electricity. Total power

Supplementary Information

				125GWh/year divided by annual production capacity. (12)
Labour				
Labour				Notes
\$2,919,986.96				^a assumed same as upstream labour for lignocellulosic-mycoprotein process (S.2.2.1)
Capital Cost and Depreciation				
Fixed Capital Investment (FCI)	Annual Depreciation	Annual Depreciation charge	Notes	
\$176,233,870	3.3% (straight-line depreciation over 30 year lifespan)	\$5,874,462	Scaled according to Supplementary S.2.1.3, based on \$40,000 tonnes/year capacity with investment cost from (10). Direct and indirect costs were not added to the investment cost as it is assumed the reported investment cost includes these.	
Economic Parameters Used				
Parameter	Value			Notes
Tax rate (%)	35			
Plant salvage value (\$)	0			
Plant life (years)	30			
Annual depreciation (%)	3.3			
Discount rate (%)	2/10			Two rates are used. 2% for the MSP that gives no real returns but reaches a positive NPV only accounting for inflation. 10% allows for a return of investment that beats inflation.

Supplementary Information

Loan equity (%)	40			
Loan interest (%)	8			
Loan term (years)	10			
Derived prices				
MSP (2% discount rate) (\$/kg)	1.37			
MSP (10% discount rate) (\$/kg)	1.65			

2.7. Crude Protein Comparison

Table S.7: Production costs of meats and a meat alternative.

Protein Source	Cost (\$/kg)	Standard Deviation (\$/kg)	Cost (\$/kg protein)	Standard Deviation (\$/kg protein)	Reference
Beef	4.29	0.0611	18.48	0.263	(13)
Lamb	5.80	0.689	26.47	3.14	(14)
Pork	2.03	0.065	9.75	0.313	(15)
Chicken	1.91	0.00805	9.02	0.0380	(16)

Table S.8. Details of average protein content derivation and price per product for Quorn and meat-based protein products.

Product	Protein Content (%)	Price (\$/kg)	Price (\$/kg protein)	Notes
Quorn	11.7	11.61	107.14	Values taken as an average across a range of Quorn products sourced from UK Supermarkets in May 2020
Beef	23.2	15.70	67.68	Protein content taken from (17). Beef price is an average of different products taken from (18).
Lamb	21.9	14.15	64.61	Protein content taken from (17). Lamb price is an average of different products taken from (19).
Pork	20.8	7.03	33.80	Protein content taken from (20) as an average of diced, stir-fry, steak and mince. Pork price is an average of different products taken from (21).
Chicken	21.2	4.13	19.52	Protein content taken from (22). Chicken price is taken from (23) for 2019 adjusted from lbs to kg.

3. Life Cycle Analysis

3.1. Methods

Land use methods were adjusted from the standard ReCiPe 2016 Midpoint method to account only for arable land use. These are provided in the supplementary file 'Land use methods.xlsx'.

3.2. Life Cycle Inventory

3.2.1. Key assumptions

Supplementary Information

Table S.9. Key assumptions used in the building of the Life Cycle Assessment in this work.

Factor	Assumption																													
Protein content	<p>When modelling the LCA for all protein sources considered in this study, the Functional Unit (FU) was a tonne of dry weight protein. The output of each model in the LCA is per tonne of total mass. Therefore, the quantity of product produced in the LCA model needs to be adjusted by the protein content to give the impact for the selected FU. Assumed protein contents are: Beef – 23.2%, Chicken – 21.2%, Pork – 20.8%, Lamb – 21.9%, and SCP (mycoprotein) – 12.6% (See Table S.8).</p> <p>For example: when evaluating the LCA model, the FU is 1 tonne of dry weight protein. Therefore, with a 23.2% protein content for beef,</p> $\frac{1 \text{ tonne dry weight protein (beef)}}{23.2\%} = 4.31 \text{ tonnes beef}$																													
Alternative protein sources	<p>For comparison of lignocellulosic-mycoprotein to alternative meat protein products, the LCA of lignocellulosic-mycoprotein was compared to that of beef, pork, chicken, and lamb. All meat products were taken at the slaughterhouse stage of meat production. Beef, chicken and pork were based on those in the agri-footprint database(24), and lamb based on that from Ecoinvent 3(25). Economic allocation was used.</p>																													
Alternative feedstocks	<p>For switchgrass and corn stover, the USLCI database was used(26). For miscanthus the database Ecoinvent 3 was used(25). For rice straw, an LCA was built based on in-field experimental data in Nguyen Van, Sander (27).</p>																													
Electricity grid mix	<p>The assumed mix used in this work was medium voltage electricity for the Rest of World (RoW) market from the Ecoinvent 3 database(25). The data is based on the year 2014. RoW is a designated geographical area which comprises the regions of the world not specifically defined within the Ecoinvent 3 database. Thus the datasets within the RoW geography are an estimated average across the RoW regions.</p> <p>The breakdown, based on share of production volume per 1 kWh of transported electricity of this mix is as follows (see ‘Electricity grid production mix’ sheet in ‘Inventory.xlsx’:</p> <table border="1"> <thead> <tr> <th>Source</th> <th>Share of production (%)</th> </tr> </thead> <tbody> <tr> <td>Peat</td> <td>0.03</td> </tr> <tr> <td>Oil</td> <td>3.95</td> </tr> <tr> <td>Coal</td> <td>41.55</td> </tr> <tr> <td>Natural gas</td> <td>20.47</td> </tr> <tr> <td>Biogas</td> <td>0.31</td> </tr> <tr> <td>Biomass</td> <td>0.42</td> </tr> <tr> <td>Nuclear</td> <td>11.27</td> </tr> <tr> <td>Wind</td> <td>3.22</td> </tr> <tr> <td>Hydroelectric</td> <td>18.46</td> </tr> </tbody> </table> <p>The resultant emission factors in the selected impact categories per kWh of transported electricity is as follows:</p> <table border="1"> <thead> <tr> <th>Impact Category</th> <th>Unit</th> <th>Value</th> </tr> </thead> <tbody> <tr> <td>Global warming</td> <td>kg CO₂ eq</td> <td>6.66 × 10⁻¹</td> </tr> <tr> <td>Terrestrial</td> <td>kg SO₂ eq</td> <td>3.64 × 10⁻³</td> </tr> </tbody> </table>	Source	Share of production (%)	Peat	0.03	Oil	3.95	Coal	41.55	Natural gas	20.47	Biogas	0.31	Biomass	0.42	Nuclear	11.27	Wind	3.22	Hydroelectric	18.46	Impact Category	Unit	Value	Global warming	kg CO ₂ eq	6.66 × 10 ⁻¹	Terrestrial	kg SO ₂ eq	3.64 × 10 ⁻³
Source	Share of production (%)																													
Peat	0.03																													
Oil	3.95																													
Coal	41.55																													
Natural gas	20.47																													
Biogas	0.31																													
Biomass	0.42																													
Nuclear	11.27																													
Wind	3.22																													
Hydroelectric	18.46																													
Impact Category	Unit	Value																												
Global warming	kg CO ₂ eq	6.66 × 10 ⁻¹																												
Terrestrial	kg SO ₂ eq	3.64 × 10 ⁻³																												

Supplementary Information

	acidification		
	Freshwater eutrophication	kg P eq	4.97×10^{-4}
	Land use	m ²	1.91×10^{-4}
	Water consumption	m ³	5.36×10^{-3}

3.2.2. [TEA][HSO₄]

Table S.10. Life Cycle Inventory for the production of [TEA][HSO₄] based on Chen, Sharifzadeh (9)

Chemical	Amount (kg/hr)	Normalised (kg/hr)
[TEA][HSO ₄]	22729.56	1
Sulfuric Acid	8949.75	0.39375
Triethylamine	9233.76	0.40624
Water	4546.04	0.20000
Utility		
Electricity	9.86361 (kWh)	0.000434 (kWh)
Cooling Water	4283.78	0.188468

3.2.3. [Ch][HSO₄]

Inventory for Choline Hydrogen Sulphate was based stoichiometric ratios of Sulphuric acid and choline hydroxide. Choline hydroxide synthesis was based on the reaction stoichiometry for a selected patented process(28), in which trimethylamine (TMA), ethylene oxide (EO), and water react in a 1:1:1 molar ratio to produce choline hydroxide. TMA and EO are fed at 1:1 molar ratio whilst water is provided in excess (2.2kg water/kg TMA) which produces a 48% choline hydroxide solution. The electricity and cooling water usage were assumed to be the same as for [TEA][HSO₄].

Table S.11. Life Cycle Inventory for the production of [Ch][HSO₄] based on experimental synthesis used in this paper based on synthesis of [Ch][OH] from Trimethylamine and ethylene oxide

Chemical	Amount (kg/hr)
[Ch][HSO ₄]	1
Sulfuric Acid	
Trimethylamine	0.4869
Water	0.1485
Ethylene Oxide	0.3631
Utility	
Electricity	0.000434 (kWh)
Cooling Water	0.188468

3.2.4. Mycoprotein

Supplementary Information

Table S.12. Life Cycle Inventory for the production of mycoprotein paste based on the input and output flows obtained from the process simulation flowsheet.

Chemical	Amount
<i>Inputs (kg/h)</i>	
Rice Straw (wet 20% moisture)	43598.62
[Ch][HSO ₄]	246.25
Cellulase	180.26
Ammonia	198.27
Lime	17.89
Caustic soda	94.58
Phosphoric Acid	230.37
Potassium sulphate	46.94
Magnesium sulphate	19.22
Copper sulphate	0.17
Manganese sulphate	0.39
Zinc monosulphate	1.29
Calcium chloride	3.09
Water (m ³ /h)	77.09
Natural gas (m ³ /h)	0.00
Electricity (kWh)	14486.46
Refrigerant	160.38
<i>Emissions (kg/h)</i>	
non-biogenic CO ₂	2114.81
Sequestered CO ₂ *	1882.21
CH ₄	2.07
N ₂	243617.20
O ₂	34243.07
Furfural	0.34
H ₂ O	53528.06
CO	42.77
NO ₂	42.77
NH ₃	4.55
H ₃ PO ₄	1.82
Acetic acid	<0.01
<i>Waste treatment (kg/h)</i>	
Fly ash	6222.04
WWT Brine	1043.04

* Sequestered into mycoprotein cell mass

3.2.5. Additional Information

The inventory data for all protein comparisons (Beef, lamb, chicken, and pork) and for lignocellulosic-mycoprotein from the selected feedstocks (Rice straw, miscanthus, strawgrass, and corn stover) is supplied in the additional file 'Inventory.xlsx'.

4. Experimental Results

4.1. Rice variety selection and compositional analysis

3 varieties were chosen for compositional analysis and pretreatment (Rc 25, Rc 442, Rc 400). The selection criteria include high-grain yields (Rc 442 and 400) and low-yield variety.

The compositional analysis (Table S.13) showed high ash contents present in the three varieties under investigation (27-33% of the dry biomass weight). This is much higher than the ash content found in our previous research (10-12%). In addition, lower glucan content was observed than their Indian counterparts (25-27% in these samples vs 44-45% using Indian rice straw/husks).

Table S.13. Compositional analysis of the three selected rice straw varieties Rc 25, Rc 400, and Rc 442 performed by Near Infrared Analysis (NIR) by Celignis. All values given in %.

Sample Name	Total Sugars	Glucose	Xylose	Mannose	Arabinose	Galactose	Rhamnose	Klason Lignin	AS ^a Lignin	Ash
Rc 25	40.32	25.25	10.11	0.39	2.56	1.48	0.26	14.01	2.54	32.80
Rc 400	44.85	27.55	12.52	0.47	2.78	1.32	0.21	13.75	2.32	27.21
Rc 442	41.75	26.34	10.61	0.32	2.66	1.48	0.19	13.82	2.21	31.01

^aAS= Acid Soluble

4.2. Pretreatment and Saccharification Experiments

Table S.14. Pulp yield of ionic liquid pretreatment of rice straw varieties (Rc 400, Rc 25, and Rc 442) with choline hydrogen sulphate ([Ch][HSO₄]) and triethylammonium hydrogensulphate ([TEA][HSO₄]) for 1 hour at 150°C.

Sample	Ionic Liquid	Pulp Yield (%)				Average	Standard Deviation
		1	2	3			
Rc 400	[Ch][HSO ₄]	66.5	74.9	71.5	70.9	4.2	
Rc 25	[Ch][HSO ₄]	74.1	72.2	69.1	71.8	2.6	
Rc 442	[Ch][HSO ₄]	72.8	74.4	74.9	74.0	1.1	
Rc 400	[TEA][HSO ₄]	49.2	49.9	51.8	50.3	1.3	

Table S.15. Composition (%) of the pretreated rice straw variety Rc 400 with [Ch][HSO₄] and [TEA][HSO₄]

Sample	Ionic Liquid	Glucan	Xylan	Mannan	Arabinan	Galactan	Lignin	Ash
Rc 400	[Ch][HSO ₄]	27.48	4.54	0.10	0.06	0.05	14.17	25.98
Rc 400	[TEA][HSO ₄]	48.02	3.90	0.13	0.04	0.03	10.10	35.52

Table S.16. Saccharification yield of pretreated pulp using [Ch][HSO₄] with Celluclast 1.5L and comparison to yield obtained from pretreatment with [TEA][HSO₄] and saccharification with Celluclast 1.5L and Cellic CTec 2. Pretreatment conditions: 1 hour at 150 C. Saccharification conditions: 7 days maintained at 50°C.

Sample	Ionic Liquid	Cellulase	Saccharification Yield (%) ^a				Standard Deviation
			1	2	3	Average	
Rc 400 - Untreated	-	Celluclast 1.5L	28.2	29.1	30.7	29.3	1.3
Rc 25 - Untreated	-	Celluclast 1.5L	30.6	31.4	30.6	30.9	0.5
Rc 442 - Untreated	-	Celluclast 1.5L	27.5	28.4	28.1	28.0	0.4
Rc 400 - Treated	[Ch][HSO ₄]	Celluclast 1.5L	43.7	40.4	43.0	42.4	1.8
Rc 25 - Treated	[Ch][HSO ₄]	Celluclast 1.5L	38.9	40.3	41.8	40.3	1.4
Rc 442 - Treated	[Ch][HSO ₄]	Celluclast 1.5L	40.5	36.9	38.0	38.4	1.9
Rc 400 - Treated	[TEA][HSO ₄]	Celluclast 1.5L	95.9	98.5	99.0	97.8	1.7

Supplementary Information

Rc 400 - Treated [TEA][HSO₄] Cellic CTec 2 91.6 92.4 94.1 92.7 1.2

^a Yield determined based on quantity of glucose obtained as a fraction of the total cellulose content in the untreated rice straw feedstock.

Table S.17. Time course experiments for pretreatment of rice straw variety Rc 400 with choline hydrogen sulphate ([Ch][HSO₄]) for times of 30, 45, 60 and 75 min at 150 C, and subsequent saccharification of pretreated pulp with Celluclast 1.5L or Cellic CTec 2. Saccharification conditions: 7 days maintained at 50°C.

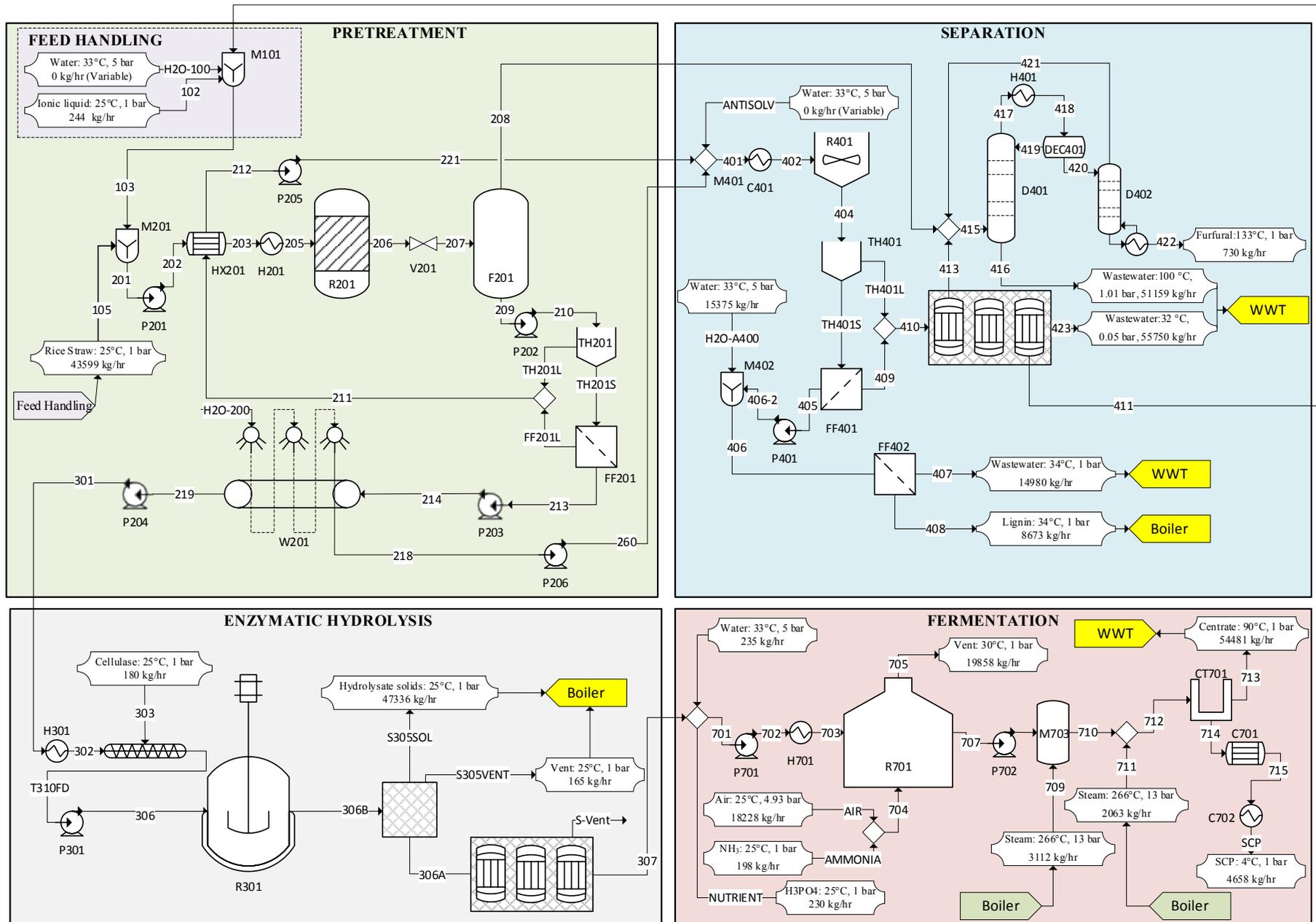
Time (min)	Pulp Yield (%)					Standard Deviation	Cellulase	Saccharification Yield (%) ^a					Standard Deviation
	1	2	3	Average	1			2	3	Average			
30	66.0	67.6	69.6	67.7	1.8	Celluclast 1.5L	15.2	38.6	41.6	31.8	14.5		
						Cellic CTec 2	76.8	48.7	51.8	59.1	15.4		
45	71.3	75.2	74.0	73.5	2.0	Celluclast 1.5L	39.7	39.2	39.7	39.5	0.3		
						Cellic CTec 2	45.9	45.1	43.6	44.9	1.1		
60	66.5	74.9	71.5	70.9	4.2	Celluclast 1.5L	43.7	40.4	43.0	42.4	1.8		
						-	-	-	-	-	-		
75	72.8	67.1	70.5	70.1	2.9	Celluclast 1.5L	31.3	31.1	32.1	31.5	0.6		
						Cellic CTec 2	38.0	34.6	35.2	36.0	1.8		

^a Yield determined based on quantity of glucose obtained as a fraction of the total cellulose content in the untreated rice straw feedstock.

5. Process

5.1. Process Diagram

Supplementary Information



Supplementary Information

	H20-200	218	219	306	306B	S305SOL	306A	S305VENT	307	NUTRIENT	703
Temperature (°C)	33.0	35.3	33.1	48.1	48.0	24.6	30.5	24.6	32.0	25.0	30.0
Pressure (bar)	5.0	1.0	1.0	5.1	1.0	1.0	1.0	1.0	3.0	1.0	3.0
Mass Liquid Fraction	1.0	1.0	0.8	0.8	0.8	0.6	1.0	0.0	1.0	1.0	1.0
Mass Solid Fraction	0.0	0.0	0.2	0.2	0.2	0.4	0.0	0.0	0.0	0.0	0.0
Mass Flows (kg/hr) - TOTAL	232382.3	146862.9	109640.3	109821.5	109821.5	47335.8	232974.0	165.5	54937.7	230.4	55403.1
H ₂ O	232382.3	145136.8	87677.0	87678.0	87129.1	29967.7	227649.0	3.2	49613.3	0.0	49848.3
Glucose	0.0	27.8	0.6	0.6	5490.0	274.5	5215.5	0.0	5215.5	0.0	5215.5
Galactose	0.0	2.9	0.1	0.1	0.1	0.0	0.1	0.0	0.1	0.0	0.1
Mannose	0.0	1.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Xylose	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Arabinose	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Cellobiose	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Soluble lignin	0.0	20.9	0.4	0.4	0.4	0.0	0.4	0.0	0.4	0.0	0.4
HMF	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Furfural	0.0	29.6	0.6	0.6	0.6	0.0	0.6	0.0	0.0	0.0	0.0
Acetic acid	0.0	6.5	0.1	0.1	0.1	0.0	0.1	0.0	0.1	0.0	0.1
NH ₃	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
O ₂	0.0	0.0	0.0	0.0	0.0	0.2	0.0	37.7	0.0	0.0	0.0
N ₂	0.0	0.0	0.0	0.0	0.0	0.4	0.0	124.6	0.0	0.0	0.0
CO ₂	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Cellulose	0.0	0.0	9013.2	9013.2	4072.6	4052.3	20.4	0.0	20.4	0.0	20.4
Galactan	0.0	0.0	223.0	223.0	223.0	221.9	1.1	0.0	1.1	0.0	1.1
Mannan	0.0	0.0	42.6	42.6	42.6	42.4	0.2	0.0	0.2	0.0	0.2
Xylan	0.0	0.0	1514.1	1514.1	1514.1	1506.5	7.6	0.0	7.6	0.0	7.6
Arabinan	0.0	0.0	711.7	711.7	711.7	708.2	3.6	0.0	3.6	0.0	3.6
Lignin	0.0	0.0	4319.6	4319.6	4319.6	4298.0	21.6	0.0	21.6	0.0	21.6
Actate	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Ash	0.0	0.0	6103.8	6103.8	6103.8	6073.3	30.5	0.0	30.5	0.0	30.5
Cellulase	0.0	0.0	0.0	180.3	180.3	179.4	0.9	0.0	0.9	0.0	0.9
[Ch][HSO ₄]	0.0	1637.2	33.4	33.4	33.4	10.9	22.5	0.0	22.5	0.0	22.5
SCP	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
H ₃ PO ₄	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	230.4	230.4

Supplementary Information

	704	705	707	710	712	713	714	402	404	TH401L	TH401S
Temperature (°C)	14.3	30.0	30.0	68.0	90.0	90.0	90.0	37.0	37.0	37.0	37.0
Pressure (bar)	1.0	1.0	1.0	5.0	5.0	1.0	1.0	2.0	1.0	1.0	1.0
Mass Liquid Fraction	0.0	0.0	1.0	1.0	1.0	1.0	0.7	1.0	1.0	1.0	0.7
Mass Solid Fraction	0.0	0.0	0.0	0.0	0.0	0.0	0.3	0.0	0.0	0.0	0.3
Mass Flows (kg/hr) - TOTAL	18426.1	19858.1	53963.4	57075.5	59138.8	54480.8	4658.0	465199.4	465199.4	441546.0	23653.3
H ₂ O	357.0	497.2	52096.7	55208.8	57272.1	53790.9	3481.1	207764.9	207764.9	200600.9	7164.0
Glucose	0.0	0.0	0.0	0.0	0.0	0.0	0.0	4145.8	0.0	0.0	0.0
Galactose	0.0	0.0	0.1	0.1	0.1	0.1	0.0	430.6	0.0	0.0	0.0
Mannose	0.0	0.0	0.0	0.0	0.0	0.0	0.0	194.2	0.0	0.0	0.0
Xylose	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Arabinose	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Cellobiose	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Soluble lignin	0.0	0.0	0.4	0.4	0.4	0.4	0.0	3113.9	0.0	0.0	0.0
HMF	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Furfural	0.0	0.0	0.0	0.0	0.0	0.0	0.0	4414.4	4414.4	4262.2	152.2
Acetic acid	0.0	0.0	0.1	0.1	0.1	0.1	0.0	967.5	967.5	934.1	33.4
NH ₃	198.3	4.1	23.0	23.0	23.0	21.6	1.4	0.0	0.0	0.0	0.0
O ₂	4162.4	577.3	0.1	0.1	0.1	0.1	0.0	0.0	0.0	0.0	0.0
N ₂	13708.4	13707.7	0.7	0.7	0.7	0.6	0.0	0.0	0.0	0.0	0.0
CO ₂	0.0	5071.6	11.9	11.9	11.9	11.2	0.7	0.0	0.0	0.0	0.0
Cellulose	0.0	0.0	20.4	20.4	20.4	6.1	14.3	0.0	0.0	0.0	0.0
Galactan	0.0	0.0	1.1	1.1	1.1	0.3	0.8	0.0	0.0	0.0	0.0
Mannan	0.0	0.0	0.2	0.2	0.2	0.1	0.1	0.0	0.0	0.0	0.0
Xylan	0.0	0.0	7.6	7.6	7.6	2.3	5.3	0.0	0.0	0.0	0.0
Arabinan	0.0	0.0	3.6	3.6	3.6	1.1	2.5	0.0	0.0	0.0	0.0
Lignin	0.0	0.0	21.6	21.6	21.6	6.5	15.1	0.0	7884.4	0.0	7884.4
Actate	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Ash	0.0	0.0	30.5	30.5	30.5	9.2	21.4	0.0	0.0	0.0	0.0
Cellulase	0.0	0.0	0.9	0.9	0.9	0.3	0.6	0.0	0.0	0.0	0.0
[Ch][HSO ₄]	0.0	0.0	22.5	22.5	22.5	21.1	1.4	244168.1	244168.1	235748.9	8419.3
SCP	0.0	0.0	1577.7	1577.7	1577.7	473.3	1104.4	0.0	0.0	0.0	0.0
H ₃ PO ₄	0.0	0.0	144.4	144.4	144.4	135.6	8.8	0.0	0.0	0.0	0.0

Supplementary Information

	423	411	406	407	408	416
Temperature (°C)	32.5	38.5	33.6	33.6	33.6	99.7
Pressure (bar)	0.0	1.0	1.0	1.0	1.0	1.0
Mass Liquid Fraction	1.0	1.0	0.7	1.0	0.1	1.0
Mass Solid Fraction	0.0	0.0	0.3	0.0	0.9	0.0
Mass Flows (kg/hr) - TOTAL	55749.5	306583.9	23653.2	14980.3	8672.9	51158.9
H ₂ O	54826.4	60987.4	15553.6	14776.0	777.7	49870.4
Glucose	0.0	0.0	0.0	0.0	0.0	0.0
Galactose	0.0	0.0	0.0	0.0	0.0	0.0
Mannose	0.0	0.0	0.0	0.0	0.0	0.0
Xylose	0.0	0.0	0.0	0.0	0.0	0.0
Arabinose	0.0	0.0	0.0	0.0	0.0	0.0
Cellobiose	0.0	0.0	0.0	0.0	0.0	0.0
Soluble lignin	0.0	0.0	0.0	0.0	0.0	0.2
HMF	0.0	0.0	0.0	0.0	0.0	0.0
Furfural	679.3	1203.3	3.8	3.6	0.2	1107.7
Acetic acid	243.8	435.6	0.8	0.8	0.0	180.5
NH ₃	0.0	0.0	0.0	0.0	0.0	0.0
O ₂	0.0	0.0	0.0	0.0	0.0	0.0
N ₂	0.0	0.0	0.0	0.0	0.0	0.0
CO ₂	0.0	0.0	0.0	0.0	0.0	0.0
Cellulose	0.0	0.0	0.0	0.0	0.0	0.0
Galactan	0.0	0.0	0.0	0.0	0.0	0.0
Mannan	0.0	0.0	0.0	0.0	0.0	0.0
Xylan	0.0	0.0	0.0	0.0	0.0	0.0
Arabinan	0.0	0.0	0.0	0.0	0.0	0.0
Lignin	0.0	0.0	7884.4	0.0	7884.4	0.0
Actate	0.0	0.0	0.0	0.0	0.0	0.0
Ash	0.0	0.0	0.0	0.0	0.0	0.0
Cellulase	0.0	0.0	0.0	0.0	0.0	0.0
[Ch][HSO ₄]	0.0	243957.7	210.5	200.0	10.5	0.0
SCP	0.0	0.0	0.0	0.0	0.0	0.0
H ₃ PO ₄	0.0	0.0	0.0	0.0	0.0	0.0

Supplementary Information

Table S.19. Summary of equipment type, size and cost for base case scenario ([Ch][HSO₄], Celluclast 1.5L). Equipment is sized and costed through the methodology defined in (5).

Tag Number	Equipment	Units	Size	Quantity	Bare Module Cost (CBM) (\$)
Feed Handling (A100)*					
M_101	Mixing tank	cum	16.16	1	\$400,553
Pretreatment (A200)					
P201_M	Motor	kW	40.8	1	\$11,761
P202_M	Motor	kW	17.2	1	\$5,182
P203_M	Motor	kW	0.3	1	Negligible
P204_M	Motor	kW	12.0	1	\$3,975
P205_M	Motor	kW	7.5	1	\$3,031
P206_M	Motor	kW	5.7	1	\$2,689
P201_P	Pump	gal/min	966.0	1	\$92,013
P202_P	Pump	gal/min	981.1	1	\$92,730
P203_P	Pump	gal/min	47.7	1	Negligible
P204_P	Pump	gal/min	431.0	1	\$65,176
P205_P	Pump	gal/min	888.3	1	\$88,314
P206_P	Pump	gal/min	654.0	1	\$76,803
HX_201	Heat Exchanger	sqm	716.5	1	\$1,760,507
R_201	Reactor System (Scaled from NREL)			1	\$77,859,405
FF_201	Filter	sqm	34.1	1	\$2,019,353
FF_202	Filter	sqm	0.0	1	Negligible
F_201	Flash Vessel	cum	11.1	1	\$338,835
M_201	Feed Mixing Tank	cum	1537.0	1	\$675,615
H_201	Heat Exchanger	sqm	29.4	1	\$208,139
TH_201	Thickener	sqm	538.1	1	\$1,228,463
Separation (A400)					
C_401	Heat Exchanger	sqm	59.1	1	\$276,602
P401_M	Motor	kW	<0.1	1	Negligible
P401_P	Pump	gal/min	34.2	1	Negligible
R_401	Precipitation Tank	cum	59.9	1	\$827,174
FF_401	Filter	sqm	12.6	1	\$1,251,403
M_402	Mixing Tank	cum	1.9	1	\$160,854
FF_402	Filter	sqm	7.1	1	\$397,600
MEE	Triple-Effect Evaporator				\$33,378,442
D_401	Distillation Column	cum	10.1	1	\$1,720,585
H_401	Condensor	sqm	80.2	1	\$324,081
DE_401	Decanter	sqm	5.1	1	\$75,483
D_402	Distillation Column	cum	0.2	1	\$459,289
TH_401	Thickener	cum	193.5	1	\$678,749

*Feed handling is a combination of NREL feed handling design and an extra mixing tank for Ionic Liquid recycle (M101)

5.2. Physical Properties

The properties of components defined in the simulation are a combination of those native to the Aspen's own chemical databanks and custom property databanks

produced my National Renewable Energy Laboratory (NREL)(29) and employed in their bioethanol model(6).

The cell mass of *Fusarium Venenatum* was determined to be $CH_{1.98}N_{0.173}O_{0.647}P_{0.0151}$ (S.5.6). The enthalpy of formation for the *Fusarium Venenatum* was determined by determining the enthalpy of combustion h_c via the Patel-Erickson equation (S19)(30).

$$h_c = -111.14 \frac{kJ}{mol} E \quad (S19)$$

Where E is the number of electrons transferred to oxygen during combustion to the combustion products (S20).

$$E = 4n_C + n_H - 2n_O - 0n_N + 5n_P + 6n_S \quad (S20)$$

Where n_C , n_H , n_O , n_N , n_P , and n_S represent the number of C, H, O, N, P and S atoms in the *F. Venenatum* composition respectively.

After determining h_c , the enthalpy of formation, $(h_f)_{bio}$ is determined by S21(31).

$$\begin{aligned} (h_f)_{bio} &= n_C(h_f)_{CO_2} + \frac{1}{2}n_H(h_f)_{H_2O} + \frac{1}{4}n_P(h_f)_{P_4O_{10}} + n_S(h_f)_{SO_3} + \frac{1}{2}n_K(h_f)_{K_2CO_3} \\ &+ n_{Ca}(h_f)_{CaO} + \frac{1}{2}n_{Fe}(h_f)_{Fe_2CO_3} - h_c \end{aligned} \quad (S21)$$

Where $(h_f)_X$ represents the enthalpy of formation of combustion product X. Where no combustion products are produced (because of element X assumed/is 0 in *F. venenatum*), then the term is omitted. An overview and example of the whole calculation is given in(32). The heat of formation was determined to be $-158.56 \text{ kJ.mol}^{-1}$.

[TEA][HSO₄] is defined through the NIST-TRC database, however, no such definition for [Ch][HSO₄] exists. Therefore, the properties of [TEA][HSO₄] were also used for [Ch][HSO₄] but the enthalpy of formation and molecular weight were changed.

In order to determine the enthalpy of formation of the ionic liquids, the molecular structure of the anion and cations were first drawn in Chem Draw 3D, and the geometry of the structure was optimised using tools inbuilt into the software. Subsequently, Molecular Orbital PACKage (MOPAC) was used to obtain the enthalpy of formation of the cation and anion. From this, the enthalpy of formation of the ionic liquid is determined via equations S22(33), S23 and S24(34).

$$\Delta H_f^o(\text{ionic salt}, 298 \text{ K}) = \quad (\text{S22})$$

$$\Delta H_f^o(\text{cation}, 298 \text{ K}) + \Delta H_f^o(\text{anion}, 298 \text{ K}) - \Delta H_L$$

$$\Delta H_L = U_{POT} + \left[p \left(\frac{n_M}{2} - 2 \right) + q \left(\frac{n_x}{2} - 2 \right) \right] RT \quad (\text{S23})$$

$$U_{POT} = \gamma \left(\frac{\rho_m}{M_m} \right)^{\frac{1}{3}} + \delta \quad (\text{S24})$$

Where the value of n_M and n_x is dependent on the nature of the ions. For monoatomic ions, they are equal to 3, 5 for linear polyatomic ions and 6 for nonlinear polyatomic ions. p is the oxidation state of the cation and q is the oxidation state of the anion. U_{POT} is the lattice potential energy, ρ_m is the density, M_m is the molecular mass, and γ and δ depend on the charge ratio between cation and anion of the ionic liquid. For [Ch][HSO₄], the density was assumed the same as [TEA][HSO₄] due to limited literature available. The property parameters used to define the two ionic liquids are given in Table S.20.

Table S.20. Defined property parameters for ionic liquids [TEA][HSO₄] and [Ch][HSO₄] in the Aspen Plus flowsheet model.

Property	[TEA][HSO ₄] ^a	[Ch][HSO ₄] ^b	Units
MW	199.3	201.235	g.mol ⁻¹
T _b	337.1	337.1	°C
T _c	644.3	644.3	°C
ΔH_f^o	-909,529	-884100	kJ.kmol ⁻¹
V _c	0.62	0.62	m ³ .kmol ⁻¹
Acentricity	0.74	0.74	
P _c	4732	4732	kPa
Density	1143	1143	kg.m ⁻³

^a based on parameters provided in the NIST-TRC database

^b based on same parameters as [TEA][HSO₄] with changes to MW and ΔH_f^o

5.3. Feed Handling

The ‘uniform-format feedstock supply system’ envisages a biomass delivery system where feedstock (rice straw feedstock composition described in Table S.21) is pre-processed in specialised depots for different categories of biomass before being transported to a central shipping terminal from which it can be blended to desired specifications.

Supplementary Information

The milled feedstock is received on site in trucks. Hoppers mediate the passage of the biomass from the dumpers to a number of conveyor belts towards short-term storage. More conveyors take the biomass from storage towards receiving bins at the pretreatment reactor. In tandem, recycled ionic liquid is mixed with fresh water in a mixing tank and fresh ionic liquid to achieve the specified water loading for pretreatment (20%) and sufficient quantity of solvent mixture for ionic liquid pretreatment (solvent/biomass = 5 wt/wt). The recycled IL enters from the separation area in which the degree of evaporation is set to reach an approximate 20% water content of the recycled IL stream, therefore, only a relatively small amount of water is required.

Table S.21. Compositional analysis and adjusted composition of rice straw feedstock (sample Rc 442) based on dry mass

	Compositional Analysis	Normalised to 100%	Adjusted for 10% ash and 1.7% Acetate	Normalised to 20% moisture
Cellulose	27.55	31.26	36.54	29.23
Xylan	12.52	14.44	16.88	13.51
Lignin	16.07	18.23	21.31	17.05
Galactan	1.32	3.15	3.69	2.95
Mannan	0.47	0.53	0.62	0.50
Arabinan	2.78	1.50	1.75	1.40
Acetate	-	0	1.70	1.36
Ash + others	27.21	30.87	17.50	14.00

5.4. Pretreatment

5.4.1. Pulp Yield and Delignification

Table S.22. Reactions and conversions for pretreatment reactor for base case scenario.

Reaction	Conversion basis	Conversion (%)
Cellulose \rightarrow 2H ₂ O + HMF	Cellulose	0
Xylan + H ₂ O \rightarrow Xylose	Xylan	0
Xylan \rightarrow Furfural + 2H ₂ O	Xylan	0.74290
Lignin \rightarrow Soluble Lignin	Lignin	0.41895
Galactan \rightarrow Galactose Oligomer	Galactan	0
Mannan \rightarrow Mannose Oligomer	Mannan	0
Arabinan \rightarrow Arabinose Oligomer	Arabinan	0
Acetate \rightarrow Acetic Acid	Acetate	1
Xylan \rightarrow Xylan Oligomer	Xylan	0
Mannan \rightarrow HMF + 2H ₂ O	Mannan	0
Mannan + H ₂ O \rightarrow Mannose	Mannan	0.80389
Galactan + H ₂ O \rightarrow Galactose	Galactan	0.63476
Galactan \rightarrow HMF + 2H ₂ O	Galactan	0
Arabinan + H ₂ O \rightarrow Arabinose	Arabinan	0
Arabinan \rightarrow Furfural + 2H ₂ O	Arabinan	0.44657
Cellulose + H ₂ O \rightarrow Glucose	Cellulose	0.29280

For the washing of the pulp to remove ionic liquid and other solubles, a belt washer was chosen due to the high number of stages that one piece of equipment achieves, therefore reducing overall equipment cost, as well as for the attainable outlet solids consistency (~17.5%) which is in-line with the feed specification of the washed pulp to enzymatic hydrolysis of less than 20% solids due to the difficulty in pumping at a solids content higher than 20% (4).

5.5. Enzymatic Hydrolysis

Supplementary Information

Table S.23. Reactions and conversions for enzymatic hydrolysis at 48C, 1 atm

Reaction	Conversion basis	Conversion (%)
Cellulose + H ₂ O → Glucose	Cellulose	0.54814 ^a
Xylan + H ₂ O → Xylose	Xylan	0*

^a Conversion determined by determining the conversion required to meet the experimentally determined conversion of cellulose in rice straw feedstock to glucose sugars (Table S.15)

* In the case of xylose utilisation of *F. Venenatum*, with conversion of xylan to xylose equals that of cellulose to glucose.

5.6. Fermentation Definition

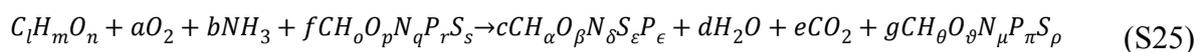
Table S.24. Elemental analysis of the live fermentation broth, results are % /dry weight

Element	%/dry weight	%/dry weight, normalised for S content of 0	Molar amount	Molar amounts relative to Carbon
Carbon	42.05	44.06	3.67	1
Hydrogen	6.95	7.28	7.28	1.98
Nitrogen	8.48	8.89	0.63	0.173
Oxygen	36.32	38.06	2.38	0.647
Sulphur	1.26	0	0	0
Phosphorus	1.64	1.72	0.055	0.0151

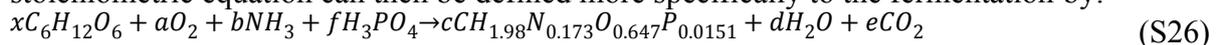
Fusarium Venenatum defined chemical formula: $CH_{1.98}N_{0.173}O_{0.647}P_{0.0151}$

To define the fermenter in Aspen, an RStoic model was used which requires stoichiometric reactions to determine the output stream composition. In order to define the stoichiometric equation with limited data (such as the respiratory quotient) usually a degree of reduction balance is required. However, the assumption of no significant extracellular products is made, therefore, only the knowledge of the yield of biomass per unit of substrate (glucose) is required to solve the series of equations which define the stoichiometric relations. Approximately 136 g protein kg⁻¹ glucose is obtained through fermentation(1). With a protein content of 45%, this would represent a yield of 302.2 g cell biomass kg⁻¹ glucose.

A general stoichiometric relation can be defined by the equation:



Where $CH_oO_pN_qP_rS_s$ is a general term to represent additional substrates and $CH_\theta O_\vartheta N_\mu P_\pi S_\rho$ is a general term to represent additional products. $CH_oO_pN_qP_rS_s$ is the fermentation microorganism, in this case defined by $CH_{1.98}N_{0.173}O_{0.648}P_{0.0151}$. Sulphur was not considered in the molecular formula for reasons discussed in (S.2.2.2). In this process, Glucose (C₆H₁₂O₆) is the primary substrate. The additional substrate considered in this process was H₃PO₄. The stoichiometric equation can then be defined more specifically to the fermentation by:



Where 7 stoichiometric coefficients need to be determined. It is convention to fix the coefficient of the substrate, therefore, $x = 1$, leaving 6 coefficients to be determined.

Based on the number of elements, a system of equations of five elemental relations can be defined:

Carbon Balance

$$l = c + e = 6 \quad (S27)$$

Supplementary Information

Hydrogen Balance

$$m + 3b = \alpha c + 2d = 12 + 3b = 1.98c + 2d \quad (\text{S28})$$

Oxygen Balance

$$n + 2a = \beta c + 2e = 6 + 2a = 0.647c + 2e \quad (\text{S29})$$

Nitrogen Balance

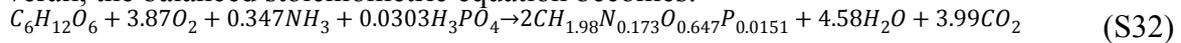
$$b = \delta c = 0.173c \quad (\text{S30})$$

Phosphorus Balance

$$f = \delta \epsilon = 0.173\epsilon \quad (\text{S31})$$

Therefore, with 5 independent relations, and 6 coefficients to be determined, a degree of freedom remains to fully specify the system. In this case, the yield of biomass to glucose substrate allows the solving of the system.

Overall, the balanced stoichiometric equation becomes:



A similar procedure was applied for xylose as substrate.

Table S.25. Reactions and conversions for fermentation

Reaction	Conversion basis	Conversion (%)
Main Reaction		
Glucose + 0.347NH ₃ + 3.87O ₂ + 0.0303H ₃ PO ₄ → 2 <i>F. Venenatum</i> + 3.99 CO ₂ + 4.58H ₂ O (for Xylose utilisation)	Glucose	1
Xylose + 0.2859NH ₃ + 3.24668O ₂ + 0.02496H ₃ PO ₄ → 1.653 <i>F. Venenatum</i> + 3.347 CO ₂ + 3.8297H ₂ O	Xylose	0*

* In the case of xylose utilisation of *F. Venenatum*, conversion equal to that of glucose

5. Scenario Evaluations

5.1. Total Capital Investment

Table S.26. Capital cost components and their values (in \$m) of the process for all four experimental scenarios at 40,000 tonnes/year production capacity.

Cost Component	Cost (\$m)			
	Scenario 1	Scenario 2	Scenario 3	Scenario 4
Area 100	16.84	16.22	10.54	10.54
Area 200	84.53	81.42	52.77	52.77
Area 300	49.94	49.19	38.42	38.42
Area 400	38.87	37.29	18.89	18.89
Area 600	34.82	33.76	24.26	24.26
Area 700*	160.36	159.83	164.11	164.11
Area 800	49.78	47.80	26.79	26.79
Area 900	5.81	5.72	4.96	4.96
Total	440.97	431.23	340.75	340.75
Warehouse 4%	6.93	6.72	4.40	4.40
site development 9%	15.60	15.11	9.91	9.91
Additional piping 5%	7.80	7.56	4.95	4.95
Total Direct Costs	471.30	460.62	360.01	360.01
Indirect Costs 60%	282.78	276.37	216.01	216.01
FCI (TDC)	754.08	736.99	576.02	576.02
Working Capital 5% of FCI	14.14	13.82	10.80	10.80

TCI **768.22** **750.80** **586.82** **586.82**

* Area 700 (fermentation) already includes its share of direct and indirect costs (See Supplementary S.2.5)

5.2. Production Costs

Table S.27. Breakdown of operating costs (\$M/year) for each scenario. The fraction of each wider operating cost category as a proportion of total operating costs (TOC) is provided at the end of each section.

Cost Factor	Cost (\$m/year)			
	Scenario 1	Scenario 2	Scenario 3	Scenario 4
Raw Materials (RM)				
Biomass	18.46	17.33	8.40	8.40
[Ch][HSO ₄]	2.09	1.94		
[TEA][HSO ₄]			0.93	0.93
Ammonia	0.86	0.85	0.89	0.89
Cellulase	9.77	9.17	5.56	5.56
Lime	0.03	0.03	0.02	0.02
Nutrient	1.95	1.94	2.02	2.02
Caustic Soda	0.06	0.06	0.06	0.06
Natural gas	-	-	-	-
RM Fraction of TOC (%)	26.41	25.79	19.50	19.50
Utilities (U)				
Process water	0.33	0.31	0.17	0.17
Refrigerant	0.01	0.01	0.01	0.01
Electricity	12.58	12.52	15.99	15.99
Landfill	9.14	8.58	4.14	4.14
U Fraction of TOC (%)	17.53	17.63	22.12	22.12
Operations (O)				
Total labour	2.92	2.81	1.82	1.82
O Fraction of TOC (%)	2.32	2.32	1.98	1.98
Maintenance (M)				
Total maintenance	10.01	9.83	8.23	8.23
M Fraction of TOC (%)	7.96	8.10	8.97	8.97
Operating overhead (OO)				
General plant overhead	0.92	0.90	0.71	0.71
Mechanical department services	0.31	0.30	0.24	0.24
Employee relations department	0.76	0.75	0.59	0.59
Business services	0.96	0.94	0.74	0.74
OO Fraction of TOC (%)	2.35	2.37	2.29	2.29
Property taxes and insurance (PTnI)				
	5.28	5.16	4.03	4.03
PTnI Fraction of TOC (%)	4.20	4.25	4.40	4.40
Depreciation				

Supplementary Information

Direct plant	25.14	24.57	19.20	19.20
D Fraction of TOC (%)	19.99	20.23	20.93	20.93
General expenses (GE)				
Selling expense	6.28	6.09	4.67	4.67
Direct research	10.05	9.75	7.47	7.47
Allocated research	1.05	1.02	0.78	0.78
Administrative expense	4.19	4.06	3.11	3.11
Management incentive compensation	2.62	2.54	1.94	1.94
GE Fraction of TOC (%)	19.23	19.32	19.59	19.59
Total Annual Operating Cost (\$M/year)	125.73	121.45	91.72	91.72

6. Economic Sensitivity Analysis

Table S.28. Assumptions varied in the economic sensitivity analysis and the corresponding change in the Minimum Selling Price (MSP \$/kg).

Assumptions Varied (min:baseline:max)	Change in MSP (\$/kg)		
	Min	Baseline	Max
IL Water Loading (30% : 20% : 10%)	0.09	0	-0.05
Biomass Loading (20% : 10% : 5%)	0.03	0	-0.02
Cellulase Price (3.14 : 6.67 : 9.41 \$/kg)	-0.14	0	0.14
Cellulose dosage (10 : 20 : 30 mg/g)	-0.14	0	0.14
Rice Straw Price (24.5 : 49 : 73.5 \$/tonne)	-0.26	0	0.26
Ionic Liquid Price (0.5 : 1 : 25 \$/kg)	-0.03	0	1.40
Xylose Utilisation (Yes : No)	-0.49	0	
Tax Rate (20% : 35% : 45%)	-0.35	0	0.34
Saccharification Yield (50.9% : 42.4% : 33.9%)	-0.60	0	0.85
Discount Rate (6% : 10% : 12%)	-0.85	0	0.43
FCI (-25% : 0% : +25%)	-0.50	0	0.50

7. Retro-techno-economic analysis (RTEA) supplementary

The following tables provide details of the trained kriging surrogate models for the RTEA. The tables list the training points (sample points of the input variables in the sample space), the observed output (MSP) of the simulation model, and the final kriging function parameters of the surrogate model as well as the

Table S.29. Sampling data for construction of kriging surrogate model in the retro-techno-economic analysis of the process model (Figure 5A). Kriging parameters (process mean μ and variance σ^2) are provided at the end.

Input Variables		Output Variables
Saccharification Yield (Fraction)	Dosage (g cellulase/g cellulose)	MSP (\$/kg)
0.4947	0.4947	5.1971
0.6526	0.6526	4.3243

Supplementary Information

0.8105	0.8105	3.7699
0.9368	0.9368	3.4507
0.5263	0.5263	5.0841
0.6842	0.6842	4.2724
0.8420	0.8420	3.7439
0.9683	0.9683	3.4371
0.4000	0.4000	6.2765
0.5579	0.5579	5.0118
0.7157	0.7157	4.2436
0.8736	0.8736	3.7376
0.9999	0.9999	3.4318
0.4316	0.4316	6.1053
0.5894	0.5894	4.9428
0.7473	0.7473	4.2161
0.9052	0.9052	3.7294
0.4631	0.4631	5.9285
0.6210	0.6210	4.8463
0.7789	0.7789	4.1726
0.4000	0.4000	6.0144
0.4000	0.4000	6.6436
0.9999	0.9999	3.2625
0.9999	0.9999	3.5336
0.4131	0.4131	6.0216
0.8903	0.8903	3.5227
0.8294	0.8294	3.9087
0.4759	0.4759	5.6317
0.7605	0.7605	4.0260
0.6028	0.6028	4.6799
0.6713	0.6713	4.4842
0.4806	0.4806	5.4823
0.7663	0.7663	3.9521
0.8870	0.8870	3.8183
0.9910	0.9910	3.4981
0.9506	0.9506	3.5174
0.7069	0.7069	4.4230
0.6118	0.6118	4.5671
0.5044	0.5044	5.5178
0.8203	0.8203	3.9860
0.9211	0.9211	3.6443
0.5481	0.5481	5.2611
0.6623	0.6623	4.5810
0.5695	0.5695	4.7538
0.7371	0.7371	3.9842
0.7898	0.7898	3.9958
0.6418	0.6418	4.5362
0.8662	0.8662	3.7164

Supplementary Information

0.4554	0.4554	5.5631
0.5801	0.5801	4.9357
0.9142	0.9142	3.5445
0.9840	0.9840	3.3633
0.5124	0.5124	5.3974
0.6952	0.6952	4.1841
μ	4.4791	
σ^2	0.4328	

Table S.30. Sampling data for construction of kriging surrogate model in the retro-techno-economic analysis of the process model (Figure 5B). Kriging parameters (process mean μ and variance σ^2) are provided at the end.

Input Variables		Output Variables
Cellulose Composition (Fraction)	Xylan Composition (Fraction)	MSP (\$/kg)
0.3737	0.4000	6.1934
0.4132	0.4211	5.6200
0.4526	0.4421	5.1411
0.4842	0.4632	4.7838
0.3816	0.4842	5.3604
0.4211	0.5053	4.9024
0.4605	0.5263	4.5067
0.4921	0.5474	4.2251
0.3500	0.5684	5.0531
0.3895	0.5895	4.6147
0.4289	0.6105	4.2541
0.4684	0.6316	3.9512
0.5000	0.6526	3.7301
0.3579	0.6737	4.4335
0.3974	0.6947	4.0788
0.4368	0.7158	3.7881
0.4763	0.7368	3.5397
0.3658	0.7579	4.0428
0.4053	0.7789	3.7412
0.4447	0.8000	3.4905
0.3500	0.4000	6.4584
0.3500	0.8000	4.0073
0.5000	0.4000	5.1992
0.5000	0.8000	3.2633
0.3532	0.4757	5.6865
0.4725	0.4088	5.2913
0.4479	0.6612	3.9304

Supplementary Information

0.3784	0.6442	4.4156
0.4085	0.6404	4.2415
0.4098	0.5594	4.6368
0.4401	0.5563	4.4615
0.3703	0.5378	5.0730
0.3768	0.7069	4.1640
0.4322	0.4546	5.1874
μ	4.5726	
σ^2	1.3520	

Table S.31. Sampling data for construction of kriging surrogate model in the retro-techno-economic analysis of the process model (Figure 5C). In the first scenario, xylose is assumed to not be utilised by *F.venenatum*. In the second scenario, xylose is assumed to be utilised by *F.venenatum*. Kriging parameters (process mean μ and variance σ^2) are provided at the end.

No xylose utilisation scenario			Xylose utilisation scenario		
Input Variables		Output Variables	Input Variables		Output Variables
Cellulose Composition (Fraction)	Xylan Composition (Fraction)	MSP (\$/kg)	Cellulose Composition (Fraction)	Xylan Composition (Fraction)	MSP (\$/kg)
0.3316	0.1300	5.3780	0.3316	0.1300	3.3404
0.3842	0.1368	4.9130	0.3842	0.1368	3.0942
0.4368	0.1437	4.5427	0.4368	0.1437	2.9023
0.4789	0.1505	4.2929	0.4789	0.1505	2.7768
0.3421	0.1574	5.2645	0.3421	0.1574	3.2245
0.3947	0.1642	4.8213	0.3947	0.1642	3.0084
0.4474	0.1711	4.4575	0.4474	0.1711	2.8348
0.4895	0.1779	4.2328	0.4895	0.1779	2.7170
0.3000	0.1847	5.7037	0.3000	0.1847	3.3544
0.3526	0.1916	5.1522	0.3526	0.1916	3.1077
0.4053	0.1984	4.7162	0.4053	0.1984	2.9170
0.4579	0.2053	4.3865	0.4579	0.2053	2.7581
0.5000	0.2121	4.1681	0.5000	0.2121	2.6517
0.3105	0.2189	5.5664	0.3105	0.2189	3.2191
0.3632	0.2258	5.0313	0.3632	0.2258	3.0025
0.4158	0.2326	4.6332	0.4158	0.2326	2.8294
0.4684	0.2395	4.3115	0.4684	0.2395	2.6826
0.3211	0.2463	5.4127	0.3211	0.2463	3.1194
0.3737	0.2532	4.9300	0.3737	0.2532	2.9252
0.4263	0.2600	4.5539	0.4263	0.2600	2.7623
0.3000	0.1300	5.7270	0.3000	0.1300	3.5049
0.3000	0.2600	5.6495	0.3000	0.2600	3.1836
0.5000	0.1300	4.1992	0.5000	0.1300	2.7555
0.5000	0.2600	4.1502	0.5000	0.2600	2.5926
0.3043	0.1603	5.6534	0.3042	0.1602	3.4108

Supplementary Information

0.4430	0.2230	4.4711	0.4636	0.1328	2.8453
0.4325	0.1888	4.5434	0.3674	0.1738	3.0830
0.3903	0.2162	4.8191	0.3378	0.2092	3.1238
0.3678	0.1740	5.0306	0.4429	0.2228	2.7751
0.3269	0.1749	5.4013	0.3904	0.2160	2.9361
0.4628	0.1328	4.3901	0.3267	0.1747	3.2467
0.3256	0.2015	5.4043	0.4321	0.1886	2.8570
0.4939	0.2299	4.1895	0.4094	0.1477	2.9824
0.3884	0.2367	4.8198	0.4640	0.1875	2.7704
0.3451	0.2578	5.1706	0.4970	0.2347	2.6263
0.4550	0.2567	4.3857	0.3452	0.2581	3.0050
0.4083	0.1474	4.7275	0.3496	0.2425	3.0228
0.3495	0.2425	5.1418	0.4549	0.2566	2.6979
0.3556	0.1406	5.1446	0.3557	0.1405	3.2058
0.4839	0.1964	4.2474	0.3874	0.2363	2.9065
0.4400	0.2420	4.4769	0.4018	0.2508	2.8394
0.3351	0.2277	5.2963	0.3704	0.1552	3.1144
0.4224	0.1622	4.6277	0.3348	0.2278	3.1040
0.4190	0.2143	4.6219	0.3136	0.2004	3.2518
μ	4.8354		μ	2.9788	
σ^2	0.1221		σ^2	0.1081	

Table S.32. Sampling data for construction of kriging surrogate model in the retro-techno-economic analysis of the process model (Figure 5D). Kriging parameters (process mean μ and variance σ^2) are provided at the end.

Input Variables			Output Variables
Cellulose Composition (Fraction)	Xylan Composition (Fraction)	Saccharification Yield (Fraction)	MSP (\$/kg)
0.3000	0.1711	0.4000	7.0991
0.3737	0.1779	0.4315	5.8746
0.4474	0.1847	0.4631	5.0122
0.3105	0.2258	0.4946	5.9670
0.3842	0.2326	0.5261	4.9984
0.4579	0.2395	0.5576	4.3201
0.3211	0.1300	0.5892	5.2175
0.3947	0.1368	0.6207	4.4272
0.4684	0.1437	0.6522	3.8804
0.3316	0.1916	0.6837	4.5876
0.4053	0.1984	0.7153	3.9484
0.4789	0.2053	0.7468	3.4874
0.3421	0.2463	0.7783	4.1046
0.4158	0.2532	0.8098	3.5605
0.4895	0.2600	0.8414	3.1719

Supplementary Information

0.3526	0.1505	0.8729	3.7717
0.4263	0.1574	0.9044	3.3027
0.5000	0.1642	0.9359	2.9629
0.3632	0.2121	0.9675	3.4473
0.4368	0.2189	0.9990	3.0423
0.3000	0.1300	0.4000	7.1293
0.3000	0.1300	0.9990	3.8094
0.3000	0.2600	0.4000	7.0493
0.3000	0.2600	0.9990	3.7595
0.5000	0.1300	0.4000	5.2069
0.5000	0.1300	0.9990	2.8520
0.5000	0.2600	0.4000	5.1550
0.5000	0.2600	0.9990	2.8056
0.4205	0.1336	0.4111	5.6724
0.3049	0.1884	0.8866	4.0598
0.3690	0.2570	0.9808	3.3736
0.3795	0.1337	0.4150	5.9979
0.4949	0.2145	0.4163	5.0637
0.3044	0.1803	0.9783	3.8095
0.4117	0.1329	0.4137	5.7193
0.3777	0.2563	0.9864	3.3152
0.4955	0.2157	0.4114	5.0925
0.3052	0.2082	0.8549	4.1437
0.4860	0.2022	0.9280	3.0136
0.3157	0.1541	0.7292	4.5401
0.4718	0.1393	0.8305	3.2975
0.4236	0.1614	0.7293	3.8087
0.3374	0.2503	0.6050	4.9048
0.4922	0.1964	0.5753	4.0743
0.3381	0.2514	0.5998	4.9279
0.3155	0.1543	0.7354	4.5159
0.4831	0.2013	0.9233	3.0313
0.4748	0.1389	0.8260	3.3004
0.4320	0.2208	0.4235	5.4258
0.4079	0.1937	0.8934	3.3987
0.4301	0.2546	0.4233	5.4196
0.3276	0.1674	0.5497	5.3910
0.4979	0.1679	0.4449	4.8389
0.4308	0.2213	0.4232	5.4347
0.4180	0.2585	0.4414	5.3537
0.3337	0.1671	0.5507	5.3236
0.4023	0.2586	0.6402	4.2453
0.4306	0.1943	0.8658	3.3627
μ	4.4449		
σ^2	1.4253		

8. LCA Supplementary

Table S.33. Life cycle impact assessment (LCIA) comparative results between beef, mycoprotein paste, chicken and tofu. Function unit = 1 tonne protein.

Impact category	Unit	Beef meat, fresh, from beef cattle, at slaughterhouse/IE Economic	Mycoprotein, economic alloc	Chicken meat, fresh, at slaughterhouse/NL Economic	Pig meat, fresh, at slaughterhouse/NL Economic	Sheep for slaughtering, live weight {GLO} market for Cut-off, S
Global warming	kg CO2 eq	172924	23660	30445	34469	46412
Terrestrial acidification	kg SO2 eq	2496	165	283	405	666
Freshwater eutrophication	kg P eq	55	13	3	3	11
Land use	m2a crop eq	229698	4390	21646	29043	83691
Water consumption	m3	1237	2232	218	193	889

Table S.34. Process contribution for lignocellulosic mycoprotein

	Land	Water	GWP	Terrestrial Acidification	Freshwater Eutrophication
Unit	m2a crop eq	m3	kg CO2 eq	kg SO2 eq	kg P eq
Mycoprotein Production	0.00	131.44	516.43	43.27	0.00
Straw Production	4180.88	1689.33	1925.49	5.21	0.45
Ionic Liquid Synthesis	0.07	11.91	582.43	2.92	0.16
Cellulase Production	199.81	284.00	1404.30	6.83	0.01
Nutrient Production	4.58	48.72	581.16	6.24	0.71
Ammonia Production	0.05	19.40	729.29	2.08	0.06
Natural Gas Production	0.00	0.00	0.00	0.00	0.00
Electricity	4.70	33.50	17289.39	96.35	11.38
Remaining Processes	0.13	13.37	631.27	1.99	0.26
Total	4390.2	2231.7	23659.8	164.9	3.9

Table S.35. Process assumptions varied in sensitivity analysis of LCA for lignocellulosic-mycoprotein production.

Assumptions Varied	Min (-%)	Baseline	Max (+%)
Water loading of ionic liquid (%)	10 (-50)	20	30 (50)

Supplementary Information

Biomass loading of ionic liquid (%)	5 (-50)	10	20 (100)
Cellulase dosage (mg/g)	10 (-50)	20	30 (50)
Saccharification yield (%)	36 (-20)	45.2	54.2 (20)
Xylose Utilisation	YES	NO	

Table S.36. Sensitivity LCIA results for 5 impact categories; Global warming, terrestrial acidification, freshwater eutrophication, land use (arable), and water consumption. Low values represent the lowest input value for sensitivity analysis (i.e. Water loading = 10%) and high value represents the highest input value (i.e. Water loading = 30%). Base value is the LCIA result of scenario 1.

	Low	Base	High
Global Warming			
Water Loading (10% : 20% : 30%)	23069.2	23659.7	24117.8
Biomass Loading (5% : 10% : 20%)	26362.9	23659.7	21749.6
Cellulase Dosage (10 : 20 : 30 mg/g)	22706.6	23659.7	24486.2
Saccharification Yield (33.9% : 42.4% : 50.9%)	23703.0	23659.7	23149.2
Xylose utilisation (Yes : No)	23088.5	23659.7	
Terrestrial Acidification			
Water Loading (10% : 20% : 30%)	161.6	164.9	167.4
Biomass Loading (5% : 10% : 20%)	180.0	164.9	154.2
Cellulase Dosage (10 : 20 : 30 mg/g)	161.2	164.9	167.7
Saccharification Yield (33.9% : 42.4% : 50.9%)	169.6	164.9	158.3
Xylose utilisation (Yes : No)	160.7	164.9	
Freshwater Eutrophication			
Water Loading (10% : 20% : 30%)	12.5	13.0	13.4
Biomass Loading (5% : 10% : 20%)	14.8	13.0	11.8
Cellulase Dosage (10 : 20 : 30 mg/g)	13.0	13.0	12.9
Saccharification Yield (33.9% : 42.4% : 50.9%)	12.5	13.0	13.1
Xylose utilisation (Yes : No)	12.8	13.0	
Arable land use			
Water Loading (10% : 20% : 30%)	4390.0	4390.2	4390.4
Biomass Loading (5% : 10% : 20%)	4391.2	4390.2	4389.2
Cellulase Dosage (10 : 20 : 30 mg/g)	4291.5	4390.2	4488.9
Saccharification Yield (33.9% : 42.4% : 50.9%)	5401.3	4390.2	3700.0
Xylose utilisation (Yes : No)	3817.5	4390.2	
Water consumption			
Water Loading (10% : 20% : 30%)	2235.0	2231.7	2229.3
Biomass Loading (5% : 10% : 20%)	2233.7	2231.7	2229.1
Cellulase Dosage (10 : 20 : 30 mg/g)	2089.7	2231.7	2373.7
Saccharification Yield (33.9% : 42.4% : 50.9%)	2718.4	2231.7	1898.4
Xylose utilisation (Yes : No)	1984.3	2231.7	

Supplementary Information

Table S.37. Summary of account of carbon sequestration of the process. Sources of negative CO₂ emissions came from biogenic CO₂ emissions and the carbon sequestered into *F. venenatum*.

	Flow (kg/hr)
Total CO₂ emissions	52500
Total biogenic CO ₂ produced in boiler	43401
Total CO ₂ from respiration of biogenic carbon sources	6983
Total biogenic CO₂	50384
Equivalent CO₂ sequestered in <i>F. Venenatum</i>	1882
Total sequestered CO₂	52267
<i>Net CO₂ produced</i>	233

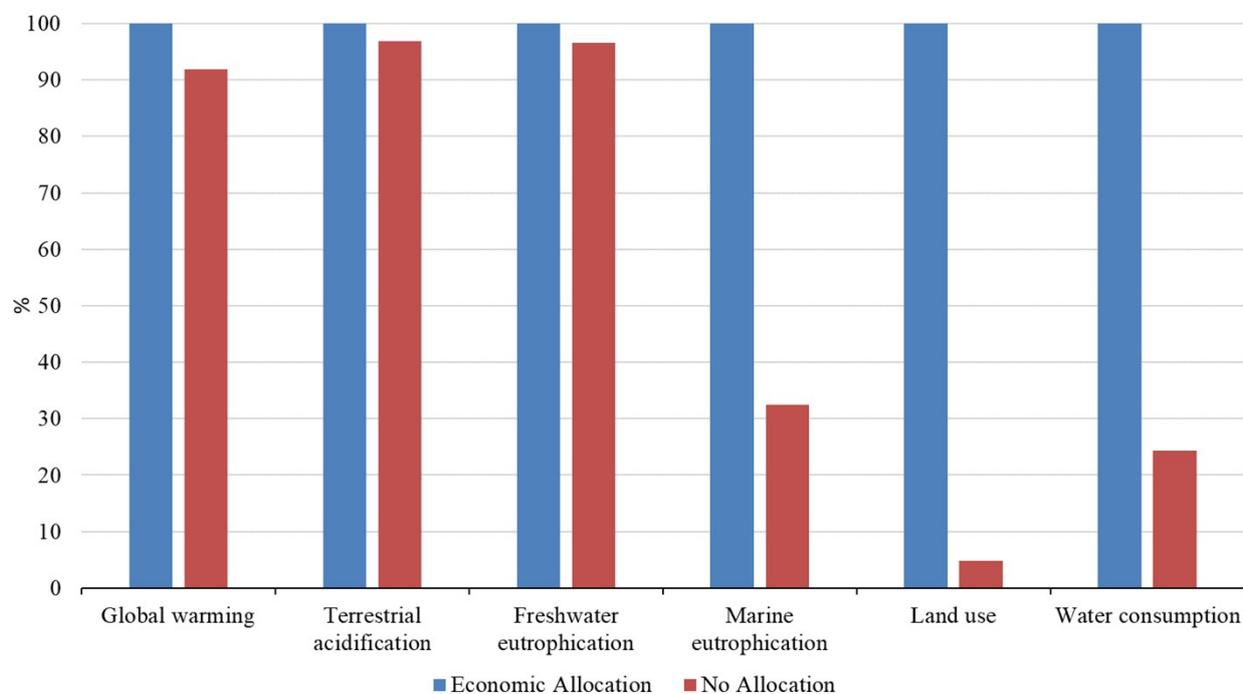


Figure S.5. Comparison of LCA results for lignocellulosic-mycoprotein when considering allocation vs no allocation of burden to rice straw

Table S.38. Reported literature rice straw compositions of three other works and composition of miscanthus, corn stover, and switchgrass

	Rice Straw				Miscanthus	Corn Stover	Switchgrass
	This work	(35)	(36)	(37)	(38)	(4) ^a	(39)
Cellulose	27.55(40.84)	28.10(37.69)	41.00(50.97)	32.15(36.26)	50.1(47.93)	35.05(43.00)	37.1(45.45)
Hemicellulose	17.09(25.34)	26.50(35.54)	21.50(26.72)	28.00(31.58)	22.4(21.43)	23.94(29.37)	32.1(39.33)
Lignin	16.07(23.82)	12.50(16.77)	9.90(12.31)	19.64(22.15)	26.8(25.64)	15.76(19.33)	6.3 ^c (7.72)
Ash	27.21(10)	18.1(10.00)	12.40(10.00)	11.33(10.00)	0.7(3.3 ^b)	4.93(6.6 ^b)	6.2(5.8 ^b)

^a Cellulose content is glucan, hemicellulose is the sum of xylan, arabinan, galactan and mannan; ^b Mean value given in parenthesis based on Kenney, Smith (40), cellulose, hemicellulose and lignin were normalised for the mean ash value of Miscanthus, Corn Stover and Switchgrass; ^c Value is of Acid Detergent Lignin (39)

References

1. Moore D, Robson GD, Trinci APJ. 21st Century Guidebook to Fungi. Cambridge, UK: Cambridge University Press; 2011.
2. Sluiter A, Hames B, Ruiz R, Scarlata C, Sluiter J, Templeton D, et al. Determination of structural carbohydrates and lignin in biomass, in: Laboratory Analytical Procedure (LAP). National Renewable Energy Laboratory. 2012.
3. Vogel HJ. A Convenient Growth Medium for *Neurospora crassa*. Microbial Genetics Bulletin 1956;13:42-7.
4. Davis R, Tao L, Scarlata C, Tan ECD, Ross J, Lukas J, et al. Process Design and Economics for the Conversion of Lignocellulosic Biomass to Hydrocarbons: Dilute-Acid and Enzymatic Deconstruction of Biomass to Sugars and Catalytic Conversion of Sugars to Hydrocarbons. National Renewable Energy Laboratory; 2015.
5. Seider WD, Lewin DR, Seader JD, Widagdo S, Gani R, Ng KM. Product and process design principles : synthesis, analysis, and evaluation 2017.
6. Humbird D, Davis R, Tao L, Kinchin C, Hsu D, Aden A, et al. Process Design and Economics for Biochemical Conversion of Lignocellulosic Biomass to Ethanol: Dilute-Acid Pretreatment and Enzymatic Hydrolysis of Corn Stover. National Renewable Energy Laboratory 2011.
7. Ulrich G, Vasudevan P. How to estimate utility costs. Chem Eng. 2006;113:66-9.
8. Eurostat. Gas prices by type of user 2018 [Available from: <https://ec.europa.eu/eurostat/tgm/refreshTableAction.do?tab=table&plugin=1&pcode=ten00118&language=en>].
9. Chen L, Sharifzadeh M, Mac Dowell N, Welton T, Shah N, Hallett JP. Inexpensive ionic liquids: [HSO₄]-based solvent production at bulk scale. Green Chem. 2014;16(6):3098-106.
10. Moore D, Robson GD, Trinci APJ. 21st Century Guidebook to Fungi. 2 ed. Cambridge: Cambridge University Press; 2020.
11. Scott K. Average advertised UK salary rises to £35,058 per year employee benefits 2019 [Available from: <https://employeebenefits.co.uk/advertised-salaries-uk-35058/>].
12. Molitor B, Mishra A, Angenent LT. Power-to-protein: converting renewable electric power and carbon dioxide into single cell protein with a two-stage bioprocess. Energy & Environmental Science. 2019.
13. AHDB. Monthly Deadweight Cattle Prices for Great Britain. 2020.

Supplementary Information

14. AHDB. Monthly Deadweight Sheep Prices for Great Britain. 2020.
15. AHDB. Monthly Deadweight Pig Prices for Great Britain. 2020.
16. Monthly Poultry Prices for the United Kingdom. In: Commission E, editor. 2020.
17. Williams P. Nutritional composition of red meat. *Nutrition & Dietetics*. 2007;64(s4 The Role of):S113-S9.
18. AHDB. Weekly Supermarket Red Meat Prices for Beef. In: Board AaHD, editor. 2000.
19. AHDB. Weekly Supermarket Red Meat Prices for Lamb. In: Board AaHD, editor. 2000.
20. Murphy KJ, Thomson RL, Coates AM, Buckley JD, Howe PR. Effects of eating fresh lean pork on cardiometabolic health parameters. *Nutrients*. 2012;4(7):711-23.
21. AHDB. Weekly Supermarket Red Meat Prices for Pork. In: Board AaHD, editor. 2000.
22. Marangoni F, Corsello G, Cricelli C, Ferrara N, Ghiselli A, Lucchin L, et al. Role of poultry meat in a balanced diet aimed at maintaining health and wellbeing: an Italian consensus document. *Food Nutr Res*. 2015;59:27606.
23. Council NC. Wholesale and Retail Prices for Chicken, Beef, and Pork. In: USDA, editor. 2020.
24. Durlinger B, Tyszler M, Scholten J, Broekema R, Blonk H. Agri-footprint; A life cycle inventory database covering food and feed production and processing. Vashon: American Center for Life Cycle Assessment; 2014. p. 310-7.
25. Wernet G, Bauer C, Steubing B, Reinhard J, Moreno-Ruiz E, Weidema B. The ecoinvent database version 3 (part I): overview and methodology. *The International Journal of Life Cycle Assessment*. 2016;21(9):1218-30.
26. NREL. U.S. Life Cycle Inventory Database. 2012.
27. Nguyen Van H, Sander BO, Quilty J, Balingbing C, Castalone AG, Romasanta R, et al. An assessment of irrigated rice production energy efficiency and environmental footprint with in-field and off-field rice straw management practices. *Sci Rep*. 2019;9(1):16887.
28. Process For Preparing Choline Hydroxide From Trimethylamine and Ethylene Oxide 2013.
29. Wooley RJ, Putsche V. Development of an ASPEN PLUS Physical Property Database for Biofuels Components. National Renewable Energy Laboratory; 1996 April 1996. Report No.: NREL/MP-425-20685.
30. Patel SA, Erickson LE. Estimation of heats of combustion of biomass from elemental analysis using available electron concepts. *Biotechnology and Bioengineering*. 1981;23(9):2051-67.
31. Battley EH. An empirical method for estimating the entropy of formation and the absolute entropy of dried microbial biomass for use in studies on the thermodynamics of microbial growth. *Thermochimica Acta*. 1999;326(1-2):7-15.
32. Popovic M. Thermodynamic properties of microorganisms: determination and analysis of enthalpy, entropy, and Gibbs free energy of biomass, cells and colonies of 32 microorganism species. *Heliyon*. 2019;5(6):e01950.
33. Gao H, Ye C, Piekarski CM, Shreeve JnM. Computational Characterization of Energetic Salts. *The Journal of Physical Chemistry C*. 2007;111(28):10718-31.
34. Jenkins HD, Tudela D, Glasser L. Lattice potential energy estimation for complex ionic salts from density measurements. *Inorg Chem*. 2002;41(9):2364-7.
35. Nassar MM. Thermal Analysis Kinetics of Bagasse and Rice Straw. *Energy Sources*. 1998;20(9):831-7.
36. Lee J. Biological conversion of lignocellulosic biomass to ethanol. *Journal of Biotechnology*. 1997;56(1):1-24.
37. Shawky BT, Mahmoud MG, Ghazy EA, Asker MMS, Ibrahim GS. Enzymatic hydrolysis of rice straw and corn stalks for monosugars production. *Journal of Genetic Engineering and Biotechnology*. 2011;9(1):59-63.
38. Gschwend FJV, Malaret F, Shinde S, Brandt-Talbot A, Hallett JP. Rapid pretreatment of *Miscanthus* using the low-cost ionic liquid triethylammonium hydrogen sulfate at elevated temperatures. *Green Chemistry*. 2018;20(15):3486-98.

Supplementary Information

39. Lemus R, Brummer EC, Moore KJ, Molstad NE, Burras CL, Barker MF. Biomass yield and quality of 20 switchgrass populations in southern Iowa, USA. *Biomass and Bioenergy*. 2002;23(6):433-42.
40. Kenney KL, Smith WA, Gresham GL, Westover TL. Understanding biomass feedstock variability. *Biofuels*. 2014;4(1):111-27.