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Organic waste as a sustainable feedstock for platform chemicals

M. Coma, E. Martínez-Hernández, F. Abeln, Sofia Raikova, Joseph Donnelly, T.C. Arnot, Michael J. Allen, D.D. Hong, C.J. Chuck

Biorefineries have been established since 1980s for biofuel production, switching lately from first to second generation feedstocks in order to avoid the food versus fuel dilemma. In a lesser extent, many opportunities have been investigated to produce chemicals from biomass using by-products of present biorefineries, simple waste streams. Current facilities apply intensive pre-treatments to deal with single substrate types such as carbohydrates. However, most organic streams such as municipal solid waste or algal blooms present a high complexity and variable mixture of molecules, which makes difficult specific compound production and separation. Here we focus on flexible anaerobic fermentation and hydrothermal processes that can treat complex biomass as a whole to obtain a range of products within an integrated biorefinery concept.

Waste generation is the second major challenge of our society. Nearly 50% of the average composition of global waste, 3M tons per day, is organic material. This accounts for the main source of greenhouse gases (GHG), and it includes household, food manufacturing and pre-factory resources, being the rest paper, plastic, glass, metal and others. These waste streams contain various compounds, most of which contain untapped energetic or economic value. Current waste management practices in decreasing order of added value to the organic waste include: animal feed, composting, incineration and landfill. Although a number of facilities direct their waste to land spread agents, these facilities only represent between 26% and 46% of organic waste and most of it is still disposed in landfills.

1. Introduction

Two main challenges of our society are the depletion of fossil resources and the increasing waste generation. In order to reduce the dependence on oil, but also mitigate climate change in transport and chemical sectors, alternative production chains are necessary. This involves a shift towards renewable resources which are not finite and can be easily regenerated.

While energy economy can be based on various alternative raw materials (win, sun, water, biomass, nuclear fission and fusion), the material economy of substances mainly depends on biomass, in particular plant biomass. Therefore, biorefineries as bioresource-converting systems, analogous to petroleum-based refineries, will be the key for the access to the bioeconomy: an integrated production of biobased products (food, feed, chemicals and materials) and bioenergy (fuels).

Waste generation is the second major challenge of our society. Nearly 50% of the average composition of global waste, 3M tons per day, is organic material. This accounts for the main source of greenhouse gases (GHG), and it includes household, food manufacturing and pre-factory resources, being the rest paper, plastic, glass, metal and others. These waste streams contain various compounds, most of which contain untapped energetic or economic value. Current waste management practices in decreasing order of added value to the organic waste include: animal feed, composting, incineration and landfill. Although a number of facilities direct their waste to land spread agents, these facilities only represent between 26% and 46% of organic waste and most of it is still disposed in landfills.

1.2. Towards a more sustainable biorefinery.

Biofuels such as bioethanol and biodiesel produced from seeds, grains and sugar, the so-called first generation (1G), exponentially increased since 1980s due to their easy...
applicability within existent engines without modifications, their renewability, their biodegradability, their lower emission generation and the ability to increase the security of supply and provide a steady income to farmers. However, energy carriers produced from crops have caused inflation in food prices and led to the food versus fuel crisis. Hence, the production of bioenergy from alternative sources such as agricultural and domestic organic wastes, the substrates for second generation (2G) biofuels which are mainly composed of lignocellulosic biomass, are now in positive shift towards green energy production. 2G biofuels generated from non-crop feedstocks release the pressure on food markets; however, there is a concern over competing land use or required land use changes. Therefore, the third generation (3G) biofuels are derived from past agricultural substrates, waste vegetable oils, microbes or microalgae as the viable alternative energy resource. The biorefinery concept embraces a wide range of technologies to separate biomass resources into their building blocks (carbohydrates, proteins, triglycerides and others) which can be converted not only to biofuels, but also to value added products and chemicals. Most of the existing biofuels and biochemicals are currently generated in single production chains and not within a biorefinery concept, and thus their exploitation is thereby limited. Modern biotechnology focusses on biofuels. Biogas production uses a resilient ecosystem of diverse microorganisms to co-convert multiple organics into methane. Bioethanol production is only possible with single bio-based substrates and single yeast strains, and its efficiency strongly depends on the bioavailability of carbohydrates. In general, these processes convert only a fraction of organics into biofuel and other outcomes as a low value co-product or a waste. To overcome this drawback, waste streams should be managed via a biorefinery system which integrates technologies flexibly and where all product outcomes are considered. The design and development of multi-purpose biorefineries that generate a variety of products as consequence of integrated, sequential, non-competitive processes is considered strategic to reach this goal.

1.3. Organic waste as feedstock.

Organic waste streams are a sustainable alternative to fossil-based resources as they do not compete directly with food crops. ‘Waste’ covers any organic material apart from the primary material for which the plants were originally grown (e.g. corn stover from maize), but it also applies to any biomass-derived by-product for which supply greatly exceeds demand (e.g. glycerol from biodiesel). Nearly all wastes currently have some value, for instance agricultural waste is used as soil improver in the fields, but the future looks forward to obtain higher value from them. Most biorefineries utilise the available feedstocks without upstream concerns which are grouped in different categories such as lignin, carbohydrates, proteins and triglycerides (from fat and oils). Lignin, from woody biomass, can be used as fuel but is also the only large volume renewable that comprises aromatics. Bulk chemicals can be obtained after the hydrolysis of carbohydrate residues to their monomers to obtain, among others, bioethanol, butanol and lactic from fermentation, or furfural and 5-hydroxymethyl-furfural produced from acid-catalysed dehydration of pentoses. Proteins residues (e.g. distillers grains) are valorised as animal feed, but the ideal scenario would isolate the non-essential amino acids as chemical feedstocks.

Rapid industrialisation across the developing world has led to a number of adverse effects on the environment. The severe dumping of plastics into water courses has fouled our oceans, rivers, lakes and estuaries; while excessive nutrient run-off from intense agricultural activity has prompted unchecked and persistent micro- and macroalgal (seaweed) blooms, which are potential feedstocks for 3G biorefineries. Harmful algal blooms (HABs) can produce anoxic zones, kill wildlife and produce toxic compounds responsible for death, illness and/or a direct restriction of commercial activities such as fishing and tourism. Whilst relatively chemically inert, there is an increasing body of evidence of the detrimental impact of plastics on aquatic wildlife and trophic food webs, in addition to the obvious impact of detritus on the aesthetics of the environment. It is of particular relevance that these vastly different anthropogenic pollutants often become entwined as the complex organic matrix associated with HABs.

Sustainable feedstocks can include energy crops grown on marginal land, agricultural and forestry residues, municipal solid waste and other novel feedstocks such as algae and other aquatic plants and microbial biomass. However, variability of quantity and composition reduces the technological and economic feasibility of potential value conversion processes. The types of biomass sources not falling into the categories mentioned above, commonly used in biorefineries, are organic fraction of the Municipal Solid Waste (MSW), manure, sewage sludge, wild fruits and crops, proteins and residues from fresh fruit and vegetables or food waste (FW). The physical and chemical characteristics of this wide spectrum biomass resources vary largely and, therefore, there are more suited for systems that can recover the potential of the organics as a whole. Examples might be anaerobic digestion (AD) to produce biogas or hydrothermal systems to produce crude oil substitute.

1.4. Biologically based conversion for complex biomass.

AD is based on a mixed microbial biotechnology (MCB) originated from the waste treatment field. Compared with pure culture based industrial biotechnology, MCB does not require sterilisation, it has a high adaptive capacity and can use mixed substrates thanks to microbial diversity and it is possible to be operated as a continuous process. The biogas produced within AD is the final product of a long chain of reactions including hydrolysis of polymers to monomers and oligomers, oxidation of these products within primary fermentation generating volatile fatty acids (VFA), lactate and ethanol, as well as hydrogen and carbon dioxide, and a final secondary
fermentation. All these biochemical reactions are enclosed within the carboxylate platform in which carboxylates are the intermediate or final targets from the conversion of biomass to chemicals or biofuel.

Hydrogen generated during acidogenesis (primary fermentation) has highest energy content per unit weight of any known fuel (143 GJ tonne\(^{-1}\)) and is the only fuel that is not bound to any carbon. In comparison to combustion of methane which is generated by AD, hydrogen is considered a cleaner technology as it does not involve carbon dioxide. However, methane can be generated alongside hydrogen in low quantities either by typical acetoclastic methanogens (from acetate) or by hydrogenotrophic organisms (from H\(_2\) and CO\(_2\)). Mixture of hydrogen and methane is known as biohythane (46-57% H\(_2\), 43-54% CH\(_4\), 0.4% CO\(_2\)) and it is a perfect fuel owing to its clean nature than methane, high fuel efficiency, improved heat efficiency and making engines easy to ignite with less input energy. The value of these technologies does not rely only on the biogas obtained anaerobically, but also on the other biobased products present on the fermentation broth which may have high commercial value in the market.

Different types of products within the carboxylate platform will depend on the substrate composition and operational conditions which will rule the syntrophy between different organisms and determine the final microbial community. In all microbial fermentations where organic carbon is both electron donor and acceptor of redox reactions, methane presents the lowest Gibbs energy change and thus an homogeneous end-product will be generated irrespective of the substrate. Thus, to switch from biogas to biochemical production in a MCB methane production must be inhibited by working at AD suboptimal conditions. Preliminary studies are required from diverse organic streams as composition variability will modify the synergy between organisms and thus the biological reactions and final product.

1.5. Hydrothermal based conversions for complex biomass. The algal biomass and plastics each represent different, yet significant, opportunities and problems from a remediation perspective. Algal biomass requires extensive drying to give a suitable feedstock for processing, rendering direct combustion or pyrolysis routes, normally used for plastic waste, uneconomic. However, suitable conversion technologies for wet biomass, such as anaerobic digestion (AD) and fermentation, are not able to convert the energetically rich plastics. Hydrothermal liquefaction (HTL) offers an interesting opportunity for simultaneous processing of heterogeneous organic material. In HTL, biomass is processed wet, with solid loadings of 5-20%, at 280-350 °C under pressures of up to 180 bar. HTL results in four product phases, including a bio-crude oil, which can be processed into fuels and chemicals similarly to crude oil, an aqueous phase containing nitrate and phosphate based micronutrients, CO\(_2\) and solid residue containing the inorganic elements and further carbon.

1.6. A multi-product integrated biorefinery for organic waste. Biorefineries must be developed using process design, technology integration, and analysis of sustainability and economics. We work towards a biorefinery that will produce different products from various sustainable feedstocks by integrating upstream, processing, and downstream stages, taking into account both biological and thermo-chemical technologies and their integration for maximum recovery. Various wild yeast strains have been applied to either produce lipids from depolymerised lignocellulose or microbial palm oil substitutes. Bacterial communities have been investigated for the production of antimicrobials, by using intermediate products of anaerobic digestion and bioethanol obtained from side-streams. Fuel precursors as bio-oil have been obtained by physical treatment of algal biomass. Although recent advances in bio-based feedstock processing have been achieved, a holistic perspective is still required to allow efficient technology integration which is justified by sustainability and economic analysis. To achieve this, previous knowledge of the substrate behaviour in each technology is required. Evaluation of the processes and their integration can be achieved thanks to systems modelling. Process system analysis tools enable integration of multi-step processes for maximisation of energy and resource recovery efficiency, mitigation of emissions, waste and cost, achieving an interdisciplinary approach for sustainable feedstock valorisation. In this work we evaluated two separate biological and thermochemical systems for organic waste streams, the latter used with plastics as main impurities of the feedstock. The final aim was to unravel the energy and chemical potential of different substrate compositions to be applied on an integrated biorefinery concept.

2. Results and Discussion

2.1. Biological conversion of food and liquid waste to biogas and chemicals.

The anaerobic digestion process is applied for complex biomass conversion into methane. Its microbiome containing hydrolyser, fermenter and methanogenic organisms in syntrophy allow the oxidation of variable substrates to a single product. High concentrations of VFA or low pH cause inhibition of methanogenesis. Limiting methane production increases the possibility to recover chemicals and alternative fuels from these systems. Under conditions of overloading and in the presence of inhibitors (e.g. free ammonia, high salts), methanogenic activity cannot remove hydrogen and volatile organic acids as quickly as they are produced. The result is the accumulation of acids, the depletion of buffering capacity and the depression of pH to levels that, sometimes, also inhibit other fermentation processes. Several carboxylic acids fermentation has been pursued at low pH (i.e. below or close to the lowest pKa) although higher titer were achieved at neutral pH, which in carboxylic acid production is between pH 6-8. At a pH around 6, short carboxylates can be recovered as bulk chemicals, further conversion of them is possible to medium-chain VFA.
with higher value or hydrogen can be simultaneously generated as alternative fuel. Therefore, the evaluation of fermentation products and biomethane potential by adjusting loads and pH within MCB allows the determination of maximum energy and chemicals production and composition. Here we tried the influence of food to microorganisms ratio during anaerobic conversion of food waste (FW) and determined the acidification potential (AP) and biochemical methane potential (BMP) of various organic substrates.

In terms of soluble COD, substrate alone or F/M of 10 and 5 solubilised a fraction of the pCOD as can be observed by the increase in solubles at the end of the experiment. Sugars and lactic acid where primary converted the propionic acid (51-54%), acetic acid (24-36%) and ethanol (2-15%) with traces of butyric (2.5-5%) and valeric (3-11%) acids. Not surprisingly, the conditions which presented the lowest ethanol (F/M of 5), produced 3.5% of caproic acid. This is in line with possible chain elongation of VFA from common organisms present in AD (e.g. Clostridium) which can reverse the β-oxidation reaction under reductive conditions supplied by hydrogen and ethanol as electron donor. Therefore, a concentration of 1 g VS L−1 (F/M of 5) was chosen as the optimal to evaluate the acidification potential (AP) of substrates as minimal carbon losses were detected for the total COD while the inoculum concentration even allowed biological upgrade of short VFA into more valuable chemicals such as caproic acid. From a ratio of F/M of 1 main losses were detected by biogas production while optimal methane production was obtained at F/M of 0.5 in line with the values in the literature.

2.1.2. Effect of organic loading. An F/M ratio of 5 was found to be optimal for AP when comparing the inoculum concentrations. However, microbes inherent in the feedstock when dealing with organic wastes also provide hydrolytic and acidogenic activity when incubated for fermentation purposes (F/M = ∞, Fig. 2a). Therefore, the quantity of organics, independent of the F/M ratio provided by the inoculum, may also increase the microbial community and modify the AP outcome when dealing with waste streams. Figure 3 presents various combinations of organic loading rates and inoculum, all with F/M ratios equal or over 1, with the exception of the control inoculum (this time with 1 g VS L−1).

As initial AP tests indicated, F/M ratios of 1 did not lead to fermentation products; at minimum loadings nearly no VFA, ethanol or biogas were produced while at higher loads of 5 g COD L−1 all organics were converted to biogases (Fig. a). This results were also confirmed by the minimal VFA production rate

Figure 2. a) Gas production during the first days of anaerobic fermentation; b) Total COD composition at start and end of each experiment; c) Soluble COD composition at start and end of each experiment. Tests were carried at various food to microorganism ratio (F/M, g COD g−1VS). F/M = ∞ refers to only substrate (5 g COD L−1) while F/M = 0 to only inocula (10 g VS L−1)
or even negative from day 3, indicating consumption of VFA to methane production (Fig 3b). From an F/M ratio of 2 and above, all tests produced similar or higher concentrations than the substrate alone (F/M = ∞, 5(0)). We observed that with similar F/M ratios, an increase of inocula boosted the total VFA production and reduced the presence of propionate by 10-30% in the final composition (e.g. 5(0.5) to 10(1) or 5(0.75) to 7.5(1)). Ethanol predominated after 7-day fermentation within the tests with maximum organic loading over inoculum; however, an ethanol peak of 0.7 g COD L⁻¹ was observed at day one and consumed afterwards for tests 5(0.75), 5(1) and 5(2). Lactic acid was only consumed from the substrate except for test 10(1) which peaked 0.5 g COD L⁻¹ after one day and depleted during consecutive days. Ethanol and lactate consumption, as electron donors for chain elongation reactions, should have increased longer chain VFA. This case was only correlated with 10(1) and 5(1) with the production of caproic acid. Finally, the tests with higher inoculum concentrations but F/M over 2 gave comparable or higher VFA concentrations than the rest and an improved VFA production rate for the first two days of fermentation as well as during the last day (Fig 3b). Although higher loading rates were providing an improved VFA concentration, the conversion yield (Y\textsubscript{VFA}), which stands for the conversion of total COD to VFA, was reduced below 50% in such cases. The maximum yield of 70% was obtained at 5(1), in line with conversions from the literature obtained for sugar based substrates to methane or to VFA\textsuperscript{31,17}. While similar values for VFA production rates and Y\textsubscript{VFA} were obtained for 7.5(2) and 5(2), ethanol production, and thus possible further valorisation to long chain fatty acids, were limited. Therefore, the optimal test conditions of F/M = 5 with 1 g VS L⁻¹ were adopted for further AP tests.

2.1.3. Effect of substrate composition on VFA and methane production. Optimised AP and BMP tests were carried out for six different liquid wastes (LW1-LW6) to determine the conversion potential to VFA or methane. Figure 4 presents the initial composition of the added substrate (5 g COD L⁻¹, except for LW2 which was a diluted substrate) and the end composition of both AP and BMP tests.

![Figure 4. VFA and methane production and composition during 7-day acidification potential (AP) tests and a 30 day biochemical methane potential (BMP) test for 6 different liquid wastes. Cont stands for control while ini stands for initial composition.](image)

Substrates with high proportion of ethanol or sugars in their initial composition (LW1, LW5 and LW6) presented the highest VFA production as well as Y\textsubscript{VFA}, being over 90% for the substrate mainly composed by ethanol. Ethanol oxidation to acetate did not only improve the increase in VFA, but provided the required energy to initiate chain elongation reactions and, thus, generate butyrate from acetate and further caproate from butyrate. Nearly no biogas, only 18 mL, was produced from LW2, being 4% CO\textsubscript{2} and the rest other than methane, supposedly hydrogen. This would have also enhanced the reductive conditions required for chain elongation. In the case where sugars were present within the initial substrate, the monomers had to undergo acidification to produce the required ethanol and acetate, accompanied with maximal biogas production other than methane (25% CO\textsubscript{2}, 75% H\textsubscript{2}), therefore chain elongation was only extended to butyrate during the 7-day fermentation.

Kinetic parameters obtained from both AP and BMP corroborated that VFA production relies on the hydrolytic capacity of the substrate as well as the activity of the microbiome. Lower VFA rates were obtained for those substrates with lower hydrolysis constants (LW2-3-4). In such
cases, substrates should be pretreated to enhance the initial step of fermentation or only applied for biomethane production with larger retention times. For sugar based substrates, even relatively low hydrolysis constants would allow a high recovery of VFA together with a potential biohydrogen recovery. These systems also presented the highest pH decrease during fermentation, thus potential inhibitions should be taken into account. Finally, ethanol based substrates can be also considered for VFA recovery with the added value of production of longer chain VFA.

<table>
<thead>
<tr>
<th>Acclimation Potential (AP)</th>
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<td>LW 6</td>
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2.2. Hydrothermal co-liquefaction (HTL) of algal and plastic wastes.

While hydrothermal co-liquefaction of plastics in water has been examined with lignocellulose, no reports detail the effect of plastics on the HTL of micro- or macroalgae. To assess this application, we studied the microalgal species *Arthrospira platensis* (Spirulina) and the macro alga *Ulva* spp. (*Ulva*), both of which are common and problematic bloom formers in areas such as Viet Nam. All samples were liquefied at 310 °C over 60 minutes. Under these conditions an oil yield of 34% was obtained for Spirulina, while the liquefaction of Ulva produced only 7% biocrude (Fig. 5a). While this is substantially lower than the Spirulina, it is common for macroalgae to contain higher polysaccharide and ash contents, and correspondingly have lower oil yields. For example, other members of this genus have been reported to yield oils from 18-32 % under similar conditions.

Plastics such as polyethylene (PE) and polypropylene (PP) are more thermally stable than the algal biomass, and under the conditions tested both plastics failed to degrade when processed separately to the biomass. However, on co-liquefaction, both PE and PP demonstrated significant degradation. For example, on the addition of both PE and PP, the overall oil yield remains approximately stable for Spirulina, despite proportionally less biomass being present in the reaction mixture, where the addition of both plastics increased the bio-crude yield dramatically for Ulva. Presumably, as the biomass begins to degrade, these secondary products impact on the thermal stability of the polymers, which then react and decompose. The hydrocarbon polymer subsequently becomes a hydrogen donor, and this hydrogen can stabilise radicals formed during biomass decomposition and prevent recondensation to solid residues. This effect has been observed on co-liquefaction of lignocellulose with HOPE, for example.

The bio-crude fraction was analysed by elemental analysis (Fig. 6a). On addition of PE or PP to the Spirulina biomass, the C:H ratio decreased compared with the bio-crude from the pure Spirulina. The effect was even more pronounced at higher plastic loadings. A similar trend was also observed for the bio-crude produced from Ulva. The N content was also reduced in the bio-crudes with as little as 3.3% being observed. Nitrogen tends to be present in aromatic heterocycles and as such requires extensive hydrotreatment for removal from the resulting crude upon chemical upgrading. Bio-oils containing lower nitrogen are therefore significantly easier to process.

![Figure 5. HTL of Spirulina and Ulva spp. (310 °C, 60 mins) where a) product mass balance of the biomass b) Bio-crude yield with increasing PE content c) Bio-crude yield with increasing PP contents](image-url)
GC-MS analysis of the bio-crudes demonstrated that the majority of the lighter fraction comprised of aromatic compounds, fatty acids and nitrogen containing heterocycles. While this does not change substantially for the co-liquefaction products, an increasing hydrocarbon content (C_{10}-C_{22}) was observed on the addition of PP and PE. On the liquefaction of Ulva, a significant proportion of additional saturated hydrocarbons were observed around the C_{20} range for both PP and PE liquefaction (Fig. 6b). This strongly suggests that the plastic is partially fragmenting and partitioning into the bio-crude fraction. 1H NMR analysis of the biocrudes demonstrated that the crude is relatively similar when PE is introduced, with approximately the same CH_2:CH_3 ratios. However, on addition of PP, a larger proportion of CH_3 groups were present, suggesting either deposition of the PP polymer chain or the production of shorter chain moieties from the biomass. To estimate the total carbon content from the biomass, ^{13}C dating of the biocrudes was undertaken, and demonstrated that between approximately 21 - 61% of the carbon in the bio-crude came from the biomass source, for the 20% polymer loadings. This equates to approximately 10-15% of the available carbon in the Spirulina biomass depositing into bio-oil and between 45-55% of the carbon available in Ulva depositing into the bio-oils.

The solid residue fraction was also increased on the addition of plastic with higher loadings of plastic producing higher yields for Spirulina and Ulva (Fig. 7a), though this was far more pronounced for the macroalgae. The additional solid residue showed an increasing carbon and decreasing nitrogen content (Fig. 7b, 7c) both factors suggesting that some of the plastic waste was depositing in this phase, with more plastic available to deposit from the Ulva sample due to lower deposition in the bio-oil.

Figure 6. Analysis of the bio-crude fractions where a) is the carbon:hydrogen ratio and nitrogen content of the bio-crude with plastic content b) GC –analysis of the bio-crude derived from Ulva c) the ratio of CH2 to CH3 moities estimated from the 1H NMR d) % fossil carbon in the bio-crude.

Figure 7. Analysis of the solid residue from the HTL of Spirulina and Ulva (310 °C, 60 mins) where a) solid residue yield with increasing plastic b) carbon % from the elemental analysis c) wt% nitrogen.
This exciting preliminary study demonstrates that the co-liquefaction of algae and plastics has significant potential for remediation of complex organic pollutants. Indeed, the additional plastics result in higher conversions to bio-crude which was generally of a higher quality than biocrude produced from algal biomass alone.

2.3 Organic waste biorefinery system integration.

A biorefinery is a facility for the sustainable conversion of biomass and waste feedstocks, through the integration of physical, chemical, biochemical and thermochemical processes, into multiple products. The analogy to today’s crude oil refineries suggests that adopting process systems engineering principles, such as feedstock fractionation, multiple product portfolio, process flexibility and process integration, should be applied in biorefinery concepts to achieve high efficiency levels. In this section an integrated biorefinery system is devised based on the process technologies investigated in this work for the co-processing of wastes. The integrated co-processing of different waste streams in a biorefinery fashion has shown potential for improved economics and also as a technological solution towards the circular economy.

Figure 8 shows systems integration of a waste biorefinery concept combining biochemical and thermochemical processes to produce platforms for biofuels or chemical production.

Figure 8 also shows the integration of alternative VFAs conversion into other products. The biochemical processing by yeast can produce palm oil substitute which can be sold for cosmetics or food industry or for biodiesel production. The chemical synthesis route involves esterification and hydrogenation into mixed alcohols including ethanol, propanol and butanol which can be sold as biofuels or as platform chemicals. The chemical synthesis route would require finding a way to supply the hydrogen required. Thus, another integration route would be needed, possibly via anaerobic fermentation which could be tuned to produce VFAs and hydrogen, or via steam reforming of biogas or gasification of solid residues. The selection of best integration alternatives for a sustainable waste biorefinery will require extensive analysis and optimisation at all levels. Figure 9 shows a systematic framework for biorefinery process design and integration. The figure also shows the goals at each level and the research areas and tools needed for a truly multidisciplinary approach to accelerate the development of sustainable biorefinery systems. Once the nature of the feedstock(s) is defined, the various steps in the framework involve the following:

Metabolic modelling. With the advancement of metabolic and genetic engineering as well as computational capabilities, starting with the microorganism cells as the core of a
biochemical process, as opposed to the reactor vessel, is now possible. To this end, metabolic engineering has largely contributed with mathematical modelling of biochemical reaction networks. Systems biology is advancing in getting knowledge about microbial communities’ behaviour and their structure and function. Therefore it is now possible to understand relation between microbial metabolisms, culture conditions and productivity to optimise microbial production in a biorefinery. This will allow to synergistically tune mixed cultures such as in the anaerobic fermentation process shown in this paper.

**Process system simulation.** VFAs are generally obtained in the reaction effluent in a diluted form together with other by-products, water and unreacted biomass. To obtain a marketable product, a proper combination of separation units (filtration, centrifugation, flash separators, distillation columns, liquid-liquid extraction columns, among others) are required. Process models are used in simulations to analyse the performance of the process integrated as a whole (reaction and separations) so that it can be optimised. The resulting mass and energy balances are the basis for the next levels of analysis and optimisation.

**Process integration.** It is important that energy and material inputs are used as efficiently as possible to offset fossil energy needs and greenhouse emissions. This involves a standard step in chemical process design and can employ pinch analysis to target energy recovery and make the biorefinery more energy efficient. Another important aspect is the integration of the various processes in the biorefinery through material streams exchange. For example, some processes can be sources of CO$_2$ (e.g. fermentation) and others can be sinks of CO$_2$ (e.g. algae cultivation) thus balancing sources and sinks can provide higher efficiency levels.

**Economic analysis.** This is a well-established step towards selecting a process design by evaluating economic performance indicators such as payback, net present value, minimum selling price, economic margin potential. Traditional life cycle costing as well as the value analysis method for economic margin analysis of biorefinery processes could be used.

**Environmental impact analysis.** An environmental impact analysis is needed to select the process alternatives that have the lowest potential for causing damage to the environment, ecosystems and human health. Traditional tools include carbon footprint and water footprint analyses and the more holistic life cycle assessment (LCA). Trade-offs between environmental and economic objectives of a biorefinery system may arise during the analyses, thus to support decision making, a simultaneous evaluation of economic value and environmental impact (EVEI) analysis can be used.

The technologies studied in this paper have the potential of enabling high productivity and conversion of waste to valuable products in a biorefinery conceptualised through systems integration and employing a holistic multidisciplinary approach for systems optimisation. This approach could open new possibilities for biorefining waste for biofuels and chemicals in a sustainable manner.

3. Experimental

3.1. Biological anaerobic conversion

3.1.1. Substrates and inoculum. Inoculum from full scale anaerobic digesters (3.14 g TS L$^{-1}$; 2.19 g VS L$^{-1}$; 33 g COD L$^{-1}$) and food and liquid waste (Table 2) were supplied by GENeco (Wessex Water, UK).
Table 2. Physico-chemical characteristics of municipal organic waste.

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<th>Sugars g COD L^{-1}</th>
<th>Lactate g COD L^{-1}</th>
<th>Ethanol g COD L^{-1}</th>
<th>VFA g COD L^{-1}</th>
<th>TS %</th>
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<tbody>
<tr>
<td>FW</td>
<td>4.6</td>
<td>157 ± 11</td>
<td>7.80</td>
<td>11.14</td>
<td>7.65</td>
<td>8.78</td>
<td>10.48 ± 0.03</td>
<td>9.56 ± 0.06</td>
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<td>LW 1</td>
<td>4.2</td>
<td>45 ± 5</td>
<td>0.03</td>
<td>2.48</td>
<td>35.91</td>
<td>7.61</td>
<td>10.09 ± 0.01</td>
<td>0.86 ± 0.01</td>
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<tr>
<td>LW 2</td>
<td>7.5</td>
<td>7 ± 2</td>
<td>0.01</td>
<td>0.00</td>
<td>0.00</td>
<td>0.04</td>
<td>1.82 ± 0.01</td>
<td>0.69 ± 0.01</td>
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<td>LW 3</td>
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<td>0.63</td>
<td>0.44</td>
<td>0.00</td>
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<td>12.42 ± 0.24</td>
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<tr>
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<td>6.2</td>
<td>87 ± 14</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>2.82</td>
<td>3.58 ± 0.01</td>
<td>2.91 ± 0.06</td>
</tr>
<tr>
<td>LW 5</td>
<td>2.7</td>
<td>196 ± 24</td>
<td>78.66</td>
<td>1.26</td>
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<td>0.00</td>
<td>17.47 ± 2.39</td>
<td>15.56 ± 4.81</td>
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<tr>
<td>LW 6</td>
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<td>87.68</td>
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<td>0.00</td>
<td>0.00</td>
<td>8.32 ± 0.07</td>
<td>7.68 ± 0.26</td>
</tr>
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### 3.1.2. Acidification potential (AP) and biochemical methane potential (BMP) tests.
Batch experiments were carried out in 500 mL Schott® bottles immersed in a water bath at 35°C. Bottles were tapped with a rubber stopper containing three ports; one of them connected to an automatic biogas counter (Bioprocesso control AMPTS II), the second port was used for sampling and the third contained the vertical stirrer controlled by the system. The right amounts of substrate and inoculum were added according to the desired proportions (F/M) and COD and VS concentrations and topped up with tap water until a working volume of 400 mL. pH was initially adjusted to neutrality (7-7.5) with 2M HCl or NaOH. For AP, variable F/M ratios and controls without substrate were tested in triplicate for 7 days, sampling at days 1, 2, 4 and 7 of the experiment. Optimised AP tests consisted of an F/M = 5 with 5 g COD L^{-1} of substrate and 1 g VS L^{-1} of inoculum. Degree of acidification was calculated according Bengston et al. for BMP, 5 g COD L^{-1} of substrate and 10 g VS L^{-1} of inoculum (F/M = 0.5) were fixed for each substrate in triplicate, including controls for inoculum. Tests were carried for 30 days and sampling was performed at the end of the experiment. Specific Methane Production (SMP) and kinetic parameters were calculated according Angelidaki et al.

### 3.2. Hydrothermal liquefaction (HTL).

#### 3.2.1. Substrate.
*Ulva lactuca* Linnaeus (Ulva) was collected from Xom Con, Nha Trang, Khanh Hoa province, Viet Nam on June 10, 2016. Prior to analysis and conversion, the macroalga was freeze-dried and milled to <1400 μm diameter. *Spirulina platensis* strain, classified as *Arthospira (Spirulina)platensis* was obtained from Hidumi Pharma Green Science Joint – Stock company, Viet Nam, and used without subsequent purification.

#### 3.2.2. Reactors.
Batch bomb-type reactors were fabricated according to literature precedent using stainless steel Swagelok® tube fittings. The reactor body consisted of a length of ½” tubing capped at one end, and connected at the other to a pressure gauge, thermocouple, and needle valve. The total internal volume of the reactors was ca. 9 mL.

#### 3.2.3. HTL procedure.
An adapted reaction procedure, based on previous studies, was followed. In a typical reaction, the reactor was loaded with 0.5 g total solids (made up of biomass and 0–20% plastics), and 5 mL freshly deionized water. The reactor was pressurised to 30 bar with compressed air, and heated within a vertical tubular furnace set to 400 °C until the specified reaction temperature was reached (310 °C+/- 10 °C, 60 min), then removed from the furnace and allowed to cool to room temperature.

After cooling, the pressure was released via the needle valve. Following this, the aqueous phase was decanted from the reactor contents and filtered through a filter paper pre-dried overnight at 60 °C. The product yield in the water phase was determined by leaving a 2.5 mL aliquot to dry in a 60 °C oven overnight, and scaling the residue yield to the total aqueous phase mass.

To separate the remaining bio-crude oil and char phase, the reactor was washed repeatedly using chloroform until the solvent ran clear, and filtered through the same filter paper used to separate the aqueous phase (after drying for a minimum of 1 h). The filter paper and collected char were washed thoroughly with chloroform to remove all remaining bio-crude. The filtrate was collected, and solvent removed in vacuo. The char yield was calculated from the mass of the retentate collected on the filter paper after drying overnight in an oven at 60 °C.

Three repeat HTL runs of *Spirulina* with no additional plastic were carried out to determine the standard deviation in mass balances under the conditions examined.

### 3.3. Analysis.
Total and volatile solids were determined by gravimetric standard 2540G. Total and soluble COD were analysed with colorimetric kits (LC014, Hach Lange®). HPLC (1260 Infinity, Agilent) was used to determine sugars, VFA and ethanol with an Aminex® HPX-87H column (Bio-Rad) and a refractive index detector using 5mM H2SO4 as mobile phase at 35 °C and 0.6 mL min. GC (7890A, Agilent) was used to with an HP-PLOT/Q column at 35 °C for 5 min using He as carrier gas and FID for CH4 and TCD for CO2.

Elemental analysis was carried out externally at London Metropolitan University on a Carlo Erba Flash 2000 Elemental Analyser to determine CHN content. (Elemental analyses were...
carried out at least in duplicate for each sample, and average values are reported).

Analysis of the biocrude was carried out by $^1$H NMR spectroscopy and GC-MS. $^1$H NMR spectroscopic measurements were carried out at 298 K using a Bruker AV400 spectrometer, operating at 400 MHz for $^1$H. Typically samples were analyzed in CDCl$_3$, and spectra were referenced to the residual CHCl$_3$ peak from the solvent ($δ$ 7.26 ppm). GC-MS analysis was carried out using the Agilent 7890A Gas Chromatograph equipped with a CP-Sil capillary column (25 m x 0.250 mm internal diameter) and a He mobile phase (flow rate: 1.2 ml min$^{-1}$), coupled with an Agilent 5975C MSD. Approximately 50 mg of each sample was dissolved in 100 ml hexane and 1 μl of each solution was loaded onto the column, pre-heated to 40 °C. This temperature was held for 1 minute and then heated to 250 °C at a rate of 10 °C min$^{-1}$ and then held for 10 minutes.

$^{13}$C analysis was undertaken by Beta Analytic Inc. (Florida, USA) according to ISO/IEC 17025:2005

Conclusions

Organic waste valorisation with complex and variable composition is possible with anaerobic fermentation processes and/or HTL processes. To unravel the chemical and energy potential during biological conversions, assessment of the substrate with BMP or AP tests is required, the latter optimised at F/M of 5 to allow all chemical conversions to occur. Feedstocks with high hydrolytic kinetics are recommended to firstly undergo fermentation instead of AD to recover chemicals from the plastic itself could improve the economic viability of the process. Systems integration and a holistic multidisciplinary approach could open new possibilities for biorefining waste for biofuels and chemicals in a sustainable manner.

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References


