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Valorization of municipal organic waste into purified lactic acid

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HIGHLIGHTS

• Municipal organic waste conversion to lactic acid, challenges and opportunities.
• Enzymatic hydrolysis of lignocellulose and food waste into fermentable sugars.
• Fermentation into D and L lactic acid isomers dependent on bacterial strain.
• Continues fermentation resulted in highest productivity due to high cell density.
• Downstream purification of lactic acid using membrane and distillation approaches.

ABSTRACT

Municipal organic waste (biowaste) consists of food derived starch, protein and sugars, and lignocellulose derived cellulose, hemicellulose, lignin and pectin. Proper management enables nutrient recycling and sustainable production of platform chemicals such as lactic acid (LA). This review gathers the most important information regarding use of biowaste for LA fermentation covering pre-treatment, enzymatic hydrolysis, fermentation and downstream processing to achieve high purity LA. The optimal approach was found to treat the two biowaste fractions separately due to different pre-treatment and enzyme needs for achieving enzymatic hydrolysis and to do continues fermentation to achieve high cell density and high LA productivity up to 12 g/L/h for production of both L and D isomers. The specific productivity was 0.4 to 0.5 h⁻¹ but with recalcitrant biomass, the enzymatic hydrolysis was rate limiting. Novel purification approaches included reactive distillation and emulsion liquid membrane separation yielding purities sufficient for polylactic acid production.

1. Introduction

Huge amounts of municipal organic waste (biowaste) are generated in cities such as discarded food and fibrous lignocellulose such as vegetable leaves and wastepaper. It is estimated that over 2 billion tonnes of biowaste is generated per year and with a large part of it not managed in an environmentally safe manner thereby creating a huge environmental challenge (Kaza et al., 2018). Biowaste has either been disposed in untreated form or at best incinerated only utilizing the heating value. In this way, the energy content of biowaste is utilized, although the nutrients are lost. The European Union has banned that biowaste is disposed in landfills and is targeting that 65% of biowaste will be reused and recycled by the year 2023 (EC. Council, 2018; EC. Council, 1999). To recycle biowaste, separation and collection are implemented in EU and will be compulsory by the year 2023. Besides Europe, many countries worldwide, attempt to implement new technologies for the utilization of biowastes.

In recent years, awareness of the challenges our planet is facing has
changed the perception of how we treat waste globally. The awareness has led to new approaches for holistic production, consumption of products and has resulted in consideration of biowastes to be perceived as bioresources. This approach has also been connected to the economy, driven by the wish to combine sustainability with welfare and life quality offered by the high technological development achieved in the last 100 years. This has been formulated in the bioeconomy approach, which relies on renewable natural resources to produce food, bio-products, biochemicals, biofuels and energy. This approach will decouple our lifestyle from fossil fuel consumption, while at the same time prevent biodiversity loss and minimize the negative impacts on the environment. One element is to use waste and residual bioresources and thereby create a circularity or cascade-based system, where everything is reused with the least energy expenditure and waste production since outputs of one process are used in another process in the desired biorefinery concept.

Consideration of environmental sustainability and reduced utilization of raw materials are the key objectives of improved waste management. New biomass is also not an endless resource and cannot entirely substitute fossil fuel resources. With the current development, heat and electricity can efficiently be generated by other renewable sources such as solar and wind energy. Therefore, we need to focus on using biomass resources for production of valuable products. There are many suggestions of bioproducts, which can be based on biowaste as substrate. Thereby it has been proposed that biowaste can be used for the production of single-cell proteins (Khoshnevisan et al., 2019), succinic acid (Olajuyin et al., 2019), volatile fatty acids (Yin et al., 2016), polyhydroxyalkanoates (Nielsen et al., 2017) and many others. One very attractive use of biowaste is the production of lactic acid (LA), which is supported by the high content of LA in fermented biowaste (Ahmad et al., 2020; Zhang et al., 2021a). It has been reported that the natural flora in biowaste has a large content of lactic acid bacteria (LAB). Therefore, it can be assumed that unsterilized biowaste can naturally ferment the contained sugars into LA (Probst et al., 2013) as outlined in Fig. 1.

LA (2-hydroxypropionic acid) is the simplest hydroxycarboxylic acid which occurs in the optically active levoratory (L-LA) and dextrorotatory form (D-LA). Due to the reactive hydroxyl group (–OH) and carboxyl group (–COOH), LA can undergo several chemical conversion reactions, which makes it a versatile platform chemical. The most recent reviews about LA production and achievable products are outlined in Table 1. The topic of lactic acid research has been reviewed on substrate sources covering biowaste (Ahmad et al., 2020; Ajala et al., 2020; Alexandri et al., 2019; López-Gómez et al., 2020b; Nduko and Taguchi, 2021). Lactic acid production has been covered using catalysts (Razali and Abdullah, 2017) and fermentation with LAB (Martínez et al., 2013; Peng et al., 2020; Rawoof et al., 2021). Lactic acid purification by downstream processing have been studied (Ghaffar et al., 2014; Jantasee et al., 2017; Komesu et al., 2017a; Kumar et al., 2019; Li et al., 2021a) including prospects for polymer production (Hamad et al., 2018; Kowalewska and Nowacka, 2020; Michalski et al., 2019; Riaz et al., 2018; Yildirim et al., 2018). These reviews concentrate mainly on few steps in the value chain creating a need for a holistic review of these steps from carbon source until downstream processing. It is also important to consider the achieved purity and isomeric L and D forms for further utilization. Biowaste also consists of animal waste, which
includes residues from slaughterhouses, such as skin, meat waste, blood, hairs and bone residues rich in protein. Animal waste has been investigated for several purposes such as production of biogas (Angelidaki et al., 2006). However, animal waste has not been investigated for LA fermentation (Toldrà et al., 2012).

This review focuses on biowaste for LA fermentation and downstream processing required for the use as a platform chemical such as for production of PLA. The biowaste is divided into food wastes, lignocellulosic residues and animal wastes, which makes it possible to rethink the production strategy such as pretreatment and enzymatic hydrolysis of the waste fractions targeting at effective fermentation and downstream processing. Thereby it will be possible to stress out specific challenges of biowastes, which is due to the complex nature of organic matter and also the indigenous microbial conversions needed.

2. Biowaste characterization, pretreatment and hydrolysis

Biowaste consists of discarded food which is primarily starch and meat based and a lignocellulosic fraction containing wastepaper, vegetable residues, plant residues and cellulose pulp. The composition of these residues is shown in Table 2.

Table 2
Food composition and lignocellulose relating to biowaste composition. The edible food waste is modelled as consisting of protein, fat, carbohydrates and dietary fibers. The lignocellulosic part of food waste contains vegetable residues and paper etc.

| Food wastes | Fraction (%) | Protein (%) | Fat (%) | Carbohydrates (%) | Sugar (%) | Starch (%) | Plant fibers (%) |
|-------------|--------------|-------------|---------|-------------------|-----------|------------|-----------------|
| Bakery products | 13 | 8 | 17 | 42 | 27 | 14 | 1 |
| Bread | 34 | 8 | 12 | 49 | 15 | 30 | 4 |
| Fruit (dried) | 20 | 1 | 0 | 16 | 3 | 3 | 2 |
| Vegetables | 20 | 3 | 3 | 8 | 3 | 3 | 2 |
| Meat, fish | 13 | 16 | 36 | 0 | 0 | 0 | 0 |
| Edible waste | 100 | 6 | 12 | 27 | 11 | 14 | 2 |
| General waste | 18 | 19 | 56 | 30.2 | |
| Univ cafeteria | 18 | 26 | 50 | 12 | 29 | 8 |
| Fine restaurant | 15 | 33 | 46 | 18 | 16 | 12 |
| Italian restaurant | 16 | 26 | 44 | 12 | 23 | 9 |
| Grill restaurant | 13 | 27 | 51 | 10 | 28 | 13 |
| Lignocellulosic waste | | | | | | | |
| Lignin | | | | | | | |
| Carbohydrates | | | | | | | |
| Cellulose | | | | | | | |
| Mannan | | | | | | | |
| Xylan | | | | | | | |
| Hardwood | 28 | 65 | 46 | 3 | 16 |
| Softwood | 28 | 66 | 47 | 13 | 16 |
| Hardwood CTMP | 25 | 71 | 55 | 1.5 | 14 |
| Softwood CTMP | 27 | 64 | 52 | 6.9 | 5.1 |

This review
2.1. Bio waste amounts, value and composition

A study by the United Nations Food and Agriculture Organization found that one-third of the global food production is wasted corresponding to 1.3 billion tons with an estimated value of $750 billion (Pradhan et al., 2021). It has been shown that 3.8 tons of CO\textsubscript{2} are produced per ton if the waste is not properly treated. On the other hand, one-ton food waste could generate 847 kWh electricity or 89 GJ of heating potential when biologically treated for biogas production (Thi et al., 2016). In Europe, food waste at the consumer level accounts for 42% of the total food supply equivalent to 126 million tons by 2020.

A study has investigated food waste composition and separated it into the categories of bread, other bakery products, fruit/vegetables and meat (Hanssen et al., 2016). The waste composition is calculated in Table 2 and separated into 6% protein, 12% fat, 27% carbohydrates, 11% monomer sugar, and 2% dietary fibers. The data show that the protein content mainly originates from meat and carbohydrates from bakery products, vegetables and fruit. The food waste amounted to 58% of the biowaste while the rest 42% was lignocellulose and plant residues. The lignocellulosic part of the biowaste contains vegetable residues and pulp fibers from waste paper. These wastes are rich in cellulose, hemicellulose sugars and lignin (Wu et al., 2021).

Studies on food waste comparing different restaurant types have shown varied protein content of 13 to 18%, fat contents of 26 to 33% and total carbohydrate contents of 44 to 51%. The carbohydrate content of 10-18% sugar, 23-29% starch, 4% cellulose and 4-9% hemicellulose. These results agreed on carbohydrate content with an Asian study showing 54% carbohydrates, 2.1% organic nitrogen and 9% oil/grease (Thi et al., 2016). Variations are expected between countries, due to seasonal variations, fractions of lignocellulosic residues in the biowaste and applied sampling methods. The high content of protein and readily fermentable sugars makes especially food waste attractive for LA fermentation (Carmona-Cabello et al., 2020) (Table 2).

2.2. Pretreatment of lignocellulosic biowaste

The process overview for pretreatment and enzymatic hydrolysis aiming at fermentable sugars for LAB is shown in Fig. 1. Digestible food waste contains no lignin which makes it easy to hydrolyze enzymatically into fermentable sugars. Therefore lignin poor compounds such as seaweed is easier to hydrolyze (Thomsen et al., 2020) than lignin rich compounds such as wood and straw (Thomsen et al., 2008). In general, mechanical grinding of biowaste is needed to make it homogeneous. For lignocellulosic biowaste, pretreatment such as alkaline treatment, hydrothermal treatment (Thomsen et al., 2008), wet oxidation (Lissens et al., 2004), steam explosion and plasma treatment (Heiske et al., 2013) have been assessed to increase the enzymatic convertibility into fermentable sugars such as glucose and xylose. Pretreatment of lignocellulosic biomass has been reviewed recently by Sankaran et al. (2020). Pretreatment approaches aiming at LA fermentation includes acid hydrolysis (Hoeheneder et al., 2021; Ouyang et al., 2020) and ionic liquid treatment (Yadav et al., 2021) (Table 3). The acid hydrolysis has the advantage of making enzymatic hydrolysis avoidable due to hydrolysis of cellulose and hemicellulose into monomeric sugars. Hydrothermal treatment is used to open the lignocellulosic structure by relocation or oxidation of lignin into surface droplets, which enables the enzymatic saccharification with cellulose and hemicellulose hydroyzing enzymes (Rodrigues et al., 2015; Thomsen et al., 2008).

2.3. Enzymatic hydrolysis of lignocellulosic biowaste

2.3.1. Enzymatic hydrolysis mechanism

Production of fermentable sugars such as glucose, xylose, arabinose and mannose from cellulosic biowaste can be done by enzymatic hydrolysis with cellulases and hemicellulases (Kari et al., 2020). The cellulases includes endo-β-1,4-glucanases hydrolyzing amorphous cellulose so exo-β-1,4-glucanase can hydrolyze cellohiose from the cellulose chain ends. Finally, β-glucosidase hydrolyses the produced cellbiose into glucose. Hemicellulases such as endo-1,4-β-xylanase and β-xyllosidases hydrolyses the xylan chains and acetyl groups with xylan esterase. In cases with pectin rich biowaste polygalacturonase is needed for liberation of galacturonic acid (Liu et al., 2016). α-L-arabinofuranosidase, β-mannanase and α-mannosidase are needed to remove sidechains containing arabinose to gain access to the xylan chains and liberate all the fermentable sugars. Although xylans are non-crystalline, many hemicellulases are thus required to break their complex chemical structure.

2.3.2. Enzymatic hydrolysis strategies aiming at LA fermentation

Yields of LA achieved in enzymatic hydrolysis and fermentation with different LAB are shown with focus on waste composition (Table 3) and with focus on process mechanisms and L and D isomers of LA (Table 4). For instance, addition of exogenous glucoamylases can enhance yield and productivities utilizing the indigenous microbiome during continuous fermentation of biowaste (Peinemann et al., 2019). Similarly, supplementation with α-amylase stimulated hydrolysis and enhanced LA

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**Table 3**

Recent studies on lactic acid fermentation using biowaste streams and defined polysaccharides.

| Waste                  | Composition                  | Microorganism                  | Key topic              | Titer (g LA/L) | Productivity (g/L/h) | LA yield (g LA/g sugar) | Reference                  |
|------------------------|------------------------------|--------------------------------|------------------------|----------------|----------------------|------------------------|---------------------------|
| Lignocellulosic wastes |                              |                                |                        |                |                      |                        |                           |
| Spent sulfite liquor   | Hemicellulose                | Enterococcus munditii          | Simultaneous           | 56.3 ±         | 0.53 ± 0.05          | 0.88 ± 0.02            | (Hoheneder et al., 2021) |
| Alfalfa silage         |                              | Bacillus subtilis              | Inhibition             | 44.2           | 0.74                 | 0.709                  | (Ouyang et al., 2020)     |
| Wheat straw            | Glucose + Xylose             | Bacillus coagulans             | Inhibitor tolerance    | 26.3           | 0.8-104 h: 0.91     | 0.79                   | (Yadav et al., 2021)     |
| Hydrolysat             |                              | Lactobacillus plantarum        | Cellulose hydrolysis   | 37             | 0.77                 |                        |                           |
| Ironic liquid treated  |                              |                                |                        |                |                      |                        |                           |
| Rice straw             |                              |                                |                        |                |                      |                        |                           |
| Starch and food derived wastes |                  |                                |                        |                |                      |                        |                           |
| Apple waste            | Starch/pectin                |                                | Anaerobic co-digestion | 28             | 0.16                 | 0.65                   | (Lian et al., 2020)      |
| Potato waste           | Starch                       |                                | Anaerobic co-digestion | 8.9            | 0.31                 |                        | (Lian et al., 2020)      |
| Food waste             | Meat, rice, vegetables, tofu | Bacillus, Enterococcus,        | Salinity, Microbial    | 30             | 0.62                 | 0.75                   | (Li et al., 2021b)       |
| Bakery waste + Yeast extract | Lactobacillus               | Batch fermentation             |                        | 59             | 2.4                  | 0.84                   | (Alexandri et al., 2020) |
| Bakery waste + lucerne green juice |              |                                |                        | 62             | 2.6                  | 0.56                   |                           |
| Bakery waste + Yeast extract | Bacillus coagulans          | Continues (0.1 h\textsuperscript{-1}) | 46.9                | 4.7            | 0.69                 |                        |                           |
| Bakery waste + Yeast extract | Bacillus coagulans          | Continues (0.2 h\textsuperscript{-1}) | 57.9                | 11.6           | 0.87                 |                        |                           |
| Bakery waste + lucerne green juice | Bacillus coagulans          | Continues (0.1 h\textsuperscript{-1}) | 65.7                | 6.6            | 0.66                 |                        |                           |
| Bakery waste + lucerne green juice | Bacillus coagulans          | Continues (0.2 h\textsuperscript{-1}) | 55                 | 11.3           | 0.48                 |                        |                           |
productivity by 70% compared to the control tests using food waste (Wang et al., 2016).

Apart from amylases, cellulose-degrading enzymes are also exploited to improve the process metrics of LA production. The enzymatic hydrolysis and subsequently, the LA yield was increased due to the addition of cellulase (20 U/g total solids (TS)) and β-glucosidase (10 U/g TS) (Wang et al., 2016). These studies have shown positive results on LA production from lignocellulosic waste or from starch in food biowaste. In both cases LA is produced by fermentation of sugars achieved by enzymatic or acidic hydrolysis as shown in Table 3 and Fig. 1.

### 2.4. Utilization of animal wastes as protein source

Animal production waste including meat, hair, nails, blood and trimming bones are rich in protein and produced at large quantities at slaughterhouses (Toldrà et al., 2012). Animal waste has not been studied specifically as protein source for LAB cell mass production aiming at LA fermentation. Protein sources have been achieved by protease treatment of meat byproducts such as blood and collagen (Arizhara et al., 2021). Addition of protein sources such as meat extract has been reported to increase the cell mass production and thus increase the LA productivity from 1.2 to 3.5 g/L/h (de la Torre et al., 2018). The implementation of animal waste is shown in Fig. 1.

### 3. Fermentation of biowaste to lactic acid focused on waste types

The recent studies on LA fermentation of biowaste have often been conducted using either fermentable sugars achieved from cellulose and hemicellulose in lignocellulosic waste or from starch in food biowaste. In both cases LA is produced by fermentation of sugars achieved by enzymatic or acidic hydrolysis as shown in Table 3 and Fig. 1.

#### 3.1. Lignocellulosic biomass and derived extractives

The cellulosic wastes tested for fermentation has been achieved from sources such as sulfite liquor derived from wood pulp production (Hoheneder et al., 2021), wheat straw acid hydrolysate (Ouyang et al., 2020) and rice straw treated with ionic liquids (Yadav et al., 2021). These biomasses all consist of lignocellulose which were pre-treated and hydrolyzed into the fermentable sugars glucose, xylose, arabinose and mannose. These pre-treatments also resulted in formation of fermentation inhibitors such as furfural and 5-hydroxymethylfurfural by degradation of sugar oligomers hydrolyzed from hemicellulose and cellulose at elevated temperature (150–200 °C) (Thomsen et al., 2009). The benefit of the ionic liquid [EMIM][OAc] tested on rice straw was thus to make the recalitrant cellulose part of the biomass enzymatic digestible at reduced temperature. The process mechanism is that the anions increased the solubility of lignin and hemicellulose without inhibitor formation resulting in increased enzymatic accessibility (Yadav et al., 2021).

The fermenting strain Enterococcus mundii assessed on the sulfite liquor could despite the fermentation inhibitors ferment 99% of the sugars including glucose, mannose, galactose, xylose and arabinose into LA. The fermentation duration was 50–120 h resulting in 99% LA yield. The average productivity was 0.73 g/L/h with 3.18 g/L as maximum. A titer of 56 g LA/L was achieved using 100 g/L of the hydrolysate sugars (Hoheneder et al., 2021). Bacillus coagulans assessed on the wheat straw hydrolysate was made tolerant to phenolic fermentation inhibitors by adaptive evolution proved by complete xylose conversion and an increase in productivity from 0.28 g/L/h in the first fermentation cycle to 0.97 g/L/h in the third adaptation cycle. The explanation for the improved tolerance was up-regulated oxidoreductases and phenolic acid decarboxylase. In addition, the study confirmed that the enzymatic hydrolysis was inhibited, which was avoided by water washing of the...
solid cellulosic fraction. Simultaneous saccharification and fermentation showed a rapid depletion of glucose and xylose after 8 h (productivity = 0.91 g/L/h) followed by a phase limited by enzymatic hydrolysis terminating after 104 h with a productivity of 0.25 g/L/h (Ouyang et al., 2020). The enzymatic hydrolysis using ionic liquid treated wheat straw also gave a high glucose yield of 92% of the theoretical during a similar fermentation time with Lactobacillus plantarum. It had a similar productivity of 0.77 g/L/h but a lower titer of 37 g LA/L (Yadav et al., 2021). All the assessed lactic acid fermenting strains E. mundtii, \textit{B. subtilis}, \textit{B. coagulans} and \textit{L. plantarum} could thereby conduct LA fermentation and the choice was thereby not critical for the LA productivity, which was 0.7 g/L/h on average while the titer increased versus the sugar concentration (Bai et al., 2020; Yadav et al., 2021) (Table 3).

3.2. Food waste containing starch and pectin

Food waste recently tested for LA fermentation are for a large extend starch related such as potato (Lian et al., 2020; Pradhan et al., 2021), wheat (Pradhan et al., 2021), apples (Lian et al., 2020) and bakery waste (Alexandri et al., 2020). The fermentation into LA requires enzymatic hydrolysis of the soluble polysaccharides into monosaccharides using α-glucosidase while the soluble proteins are hydrolyzed with protease. These enzymes were produced in a mixed culture approach studied by Li et al. (2021b). However, for bread derived starch, enzymes were added including Ban 2401 (endo-α-amylase) and Stargen™ 002 and Viscozyme for cellulose hydrolysis due to the lignocellulose content in the lucerne green juice (Alexandri et al., 2020).

The anaerobic co-digestion studied by Lian et al., (2020) using swine manure as inoculum could be done without enzyme addition for both apple and potato waste. The apple waste gave a higher LA concentration (28 g/L) than the potato waste (8.9 g/L) but the productivity was low (0.16 g/L/h). The use of a mixed community explained the low productivity with formation of side products such as butyric acid and acetic acid (Lian et al., 2020).

The co-fermentation of food waste at saline conditions resulted in a stable microbial community of Bacillus \textit{sp.}, Enterococcus \textit{sp.} and Lactobacillus \textit{sp}. The study showed a LA productivity of 0.61 g/L/h and an achieved LA concentration of 30 g/L (Li et al., 2021b), which are similar to the results achieved on the lignocellulosic residues (Table 3) as explained due to similar fermentable sugars such as glucose and xylose achieved during the hydrolysis step.

The bakery waste batch fermentation resulted in a higher LA productivity of 2.4 g/L/h with Bacillus coagulans as fermenting strain with a titer of 62 g/L (Alexandri et al., 2020) compared with potato/apple derived starch (Li et al., 2021b; Lian et al., 2020). When lucerne green juice hydrolysate was added replacing yeast extract a 6 h lag phase took place but no significant change occurred in the LA titer demonstrating that it was useful as nitrogen supplement. The glucose was fully fermented as it is a base carbon source for many LAB (Yadav et al., 2021).

However, the disadvantage content was not fully fermented confirming the need for enzymatic hydrolysis similar to findings by Pradhan et al., (2021). Shifting to continues fermentation increased the productivity to 11.6 g/L/h at a dilution rate of 0.2 h⁻¹. The increase was explained by an increase in cell density from 4200 to 9600 cells/μL (Table 3). Shifting to lucerne as nutrient source did not change the titer and productivity significantly. However, free glucose was observed when the dilution rate was increased to 0.2 h⁻¹. Despite this problem the results show the benefit in adding nutrient rich lucerne green juice to starch waste such as bread with a low protein content (Table 2) increasing the prospect of fermenting biowaste into LA.

4. Fermentation of biowaste to lactic acid with bacterial focus

LA fermentation is a relatively quick microbially mediated process for the production of one of the two stereoisomers (L- and D-LA) or their racemic mixture. LAB have some specific phenotypic characteristics as their DNA have fewer GC base pairs compared to AT, are Gram-positive, are facultative anaerobes, do not form spores, tolerate acidic conditions (pH < 5), are immobile, and can ferment vast carbon sources having LA as end-product (Martínez et al., 2013). Despite the majority of LAB can optimally grow between 30 to 37 °C, some species are tolerant to temperatures up to 50 °C (Bosma et al., 2017).

Lactic acid bacteria are categorized into homo- and heterofermentative species. The principle of the homofermentative process is shown in Fig. 2a with lactic acid as the principal metabolite producing two mole LA and two mole ATP per mole glucose. The intermediate pyruvate is produced by glycolysis in the Embden–Meyerhof–Parnas pathway (Romano and Conway, 1996). In the heterofermentative process mentioned as the phosphoketolase pathway for C6 sugar, a mixture of 1 mol LA, 1 mol CO₂ and the by-product 1 mol ethanol are produced per mole glucose. For C5 sugar, 1 mol LA and 1 mol acetate are produced per mole xylose (Endo and Dicks, 2014) as shown in Fig. 2b. The heterofermentative species are divided into facultative and obligate types. The facultative type including \textit{L. plantarum} and \textit{L. pentosus} applies the homofermentative process when glucose is available and the phosphoketolase pathway when only C5 sugar is available. Finally, the obligate hetero-fermenters such as \textit{L. sanfranciscensis} and \textit{L. brevis} (Prücker et al., 2015) always uses the phosphoketolase pathway to dissipilate both C5 and C6 sugars.

4.2. Fermentation of biowaste to lactate

Considering that biowaste can contain a mixture of organic household wastes, restaurant wastes, garden wastes and industrial food wastes; understandably, the content of degradable organics including hexoses and pentoses such as hemicellulose, cellulose, and starch can vary greatly in the waste stream. Despite the high sugar content of biowaste (Table 2), the availability of free monomeric sugars is not high. In contrast, starch, cellulose, and hemicellulose represent a big share in the waste stream. Hence, the addition of exogenous enzymes or inoculation with hydrolytic microbes would be needed to hydrolyze the complex polysaccharides into oligomers and monosaccharides. Table 2 shows a rough estimate of the biowaste composition and justifies the need for hydrolysis of cellulose, hemicellulose, and pectin to fermentable sugars. Overall fermentation results on biowaste are outlined in Table 4 assessing a range of LAB bacteria.

4.2.1. Challenges for increased process metrics

Biowaste has a high content of food residues and contains a surplus of nutrients (e.g., nitrogen, phosphorus, magnesium, potassium, calcium) improving microbial growth and subsequently the LA production (Kwan et al., 2017; López-Gómez et al., 2020b). However, biowaste can be quite heterogeneous as mentioned, and has also different characteristics in terms of pH, salinity, and inhibitors (e.g., ethanol) creating stress conditions under which the homofermentative strains can shift the metabolism towards formic acid by the action of pyruvate-formate lyase (Martínez et al., 2013; Mayo et al., 2010). While LAB species could markedly proliferate in the biowaste under non-controlled conditions (Probst et al., 2013), antagonism between different bacteria for sugars utilization creates a non-ideal environment for LA optimization.

4.2.2. Exploitation of native microbiome

Natural inhabitants in the fresh biowaste provide the initial seed and within the native microbiome, lactobacilli can thus proliferate and dominate the community. In this frame, Probst et al., (2013) found that...
the heterolactic Lactobacillus brevis, homolactic Lactobacillus plantarum, and their closest genera accounted for more than 70% of the community. L. brevis are heterofermentative LAB with the ability to ferment xylose (Fig. 2), low ethanol tolerance, and low performance at acidic conditions (Bosma et al., 2017; Cui et al., 2011). On the other hand, the homofermentative L. plantarum spp. are found in numerous fermented food products (Behera et al., 2018) and their dominance in biowaste native flora has a great impact on LA production. Despite the generally low potential of LAB to hydrolyze complex sugars, L. plantarum has high amylolytic capacity favoring their presence in starchy food waste streams (i.e., potato, corn, wheat, rice) to perform simultaneous hydrolysis and fermentation of LA (John et al., 2007). For example, LAB with high amylolytic capacity can adapt their metabolism to starch degradation during continuous conversion and depletion of C5 and C6 sugars (Dreschke et al., 2015). Moreover, L. plantarum species can grow well over a wide pH range favoring their establishment in biowaste (Sakai et al., 2000).

Furthermore, the homo-fermentative genera of Pediococcus and Streptococcus are also well-known LA cell factories (Carr et al., 2002). At first, Pediococcus are proliferated during the food waste decomposition (Jiang et al., 2020; Lim et al., 2020). In the literature, Pediococcus acidilactici was successfully utilized since it can quickly grow on food waste producing high titer of LA and inhibiting the formation of acetic acid by other strains (Tran et al., 2019). Next, Streptococcus can naturally dominate biowaste related food waste as it can secrete extracellular amylases to hydrolyze starch and convert it directly to LA (Demichelis et al., 2017). This ability makes it a good candidate for simultaneous saccharification and fermentation with high yields and productivities (Pleissner et al., 2017).

4.2.3. Pure or open culture operation

Open culture fermentation can be used as approach, which has the characteristics of not being pre-sterilized and use a naturally evolved mixed culture in the fermentation. Although open culture fermentations have some unique characteristics such as no need for sterilization and exploitation of indigenous microbiomes, bi-products such as ethanol, acetic, propionic, and butyric acids are formed reducing the potential for achieving high LA yield and titer. At non-sterile conditions, other acidogenic native strains can proliferate and either compete for sugars utilization (Zhang et al., 2021b) or use LA as a carbon source (Wang et al., 2016) for by-product formation.

On the contrary, sterilization of biowaste could theoretically be applied to deactivate the competing microbes. On this topic, autoclavation was examined as a pretreatment technique to release monosaccharides and simultaneously, eliminate the activity of indigenous microbes before the inoculation with L. delbrueckii (Tsapekos et al., 2020). Nevertheless, sterilization led to a markedly lower yield (0.22 g/g total sugars) compared to the non-autoclaved treatment (0.66 g/g total sugars) revealing the robustness of mixed culture fermentation compared to pure culture fermentation. The robustness of the LA fermentation process makes logistics (collection, storage, transportation etc.) of the biowaste less critical. Despite the potential for achieving high efficiency during mixed culture fermentation, the risk of producing a racemic mixture of lactate is increased. The L- and D- enantiomers are produced based on LAB’s ability to encode L- and D-lactate dehydrogenase, respectively (Bosma et al., 2017). L-lactic acid fermentation is currently the dominant approach with only a few D-LA studies (Table 4).

4.2.4. D-Lactic acid fermentation

D-LA has been produced by fermentation of sugars from enzymatic hydrolyzed pulp mill residue and orange peel waste using L. coryniformis (de Oliveira Moraes et al., 2016) and L. delbrueckii (Bustamante et al., 2020; de la Torre et al., 2018), respectively. Fermentable sugars were produced by enzymatic hydrolysis using cellulase enzyme cocktails (Celluclast/Cellic CTeC2). The orange peels hydrolysis resulted in equal concentrations of glucose and the sum of fructose + galactose as fermentable sugars. The effect on nitrogen source was studied by de la Torre et al. (2018) including meat extract (ME), yeast extract (YE) and corn stover liquor (CSE). Effective sugar conversion was achieved with at least 1.2 g/L nitrogen supplement. The fraction of converted fructose + galactose was nearly 95%, for YE 87% and for CSL 67% resulting in cell mass concentrations of 7.48, 7.35 and 4.78 g/L, respectively. The achieved LA productivities were 3.4, 3.3 and 2.4 g/L h corresponding to similar biomass based LA productivities in the range of 0.46 – 0.49 h\(^{-1}\). This indicates that the fermentation rate was limited by the cell mass content similar to results on L-LA fermentation and thus increased proportional to the cell mass (Alexandri et al., 2020). The achieved LA concentrations were in the range of 39 – 57 g/L with yields of 0.84 – 0.97 g LA/g sugar. These results are similar to what is achieved for the production of L-LA (Bustamante et al., 2020; de Oliveira Moraes et al., 2016).

4.2.5. Opportunities to overcome the limitations

To round up, microbial diversity (i.e., indigenous microbes) and dynamicity (i.e., inoculation with pure strains) highly affect optical purity, yield, titer, and productivity. As alternative approaches to

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**Fig. 2.** Lactic acid production for homofermentative LAB fermenting glucose (a) and heterofermentative LAB fermenting C\(_6\) sugars and C\(_5\) sugars with ethanol and acetate as byproducts. The C\(_6\) and the C\(_5\) metabolism are in brown and green, respectively.
manipulate the microbiome and optimize process feasibility could be the controlled bio-waste storage time and logistics (Zhang et al., 2021a), pretreatment techniques (López-Gómez et al., 2020a; Yousuf et al., 2018), and inoculation with LAB after evolutionary or stepwise adaptation (Dreschke et al., 2015; Yang et al., 2015). The reported studies indicate that the LAB cell mass was limiting for both L-LA fermentation (Alexandri et al., 2020) and D-LA fermentation (de la Torre et al., 2018). Prospects for effective LA fermentation are improved considering implementation of sustainable N-sources such as corn stover liquor (de la Torre et al., 2018), green seaweed (Bentil et al., 2019) and slaughterhouse waste (Arhara et al., 2021).

5. Downstream processing

Downstream processing used to purify fermentation broth filtration approaches, reactive distillation, and molecular distillation. Results on the purification are shown in Table 5 and indicated in Fig. 1.

5.1. Downstream separation process for lactic acid recovery

Downstream separation is a major factor to be considered for a cost-effective LA production which usually represents 20–50% of the operation costs of the process (Alves de Oliveira et al., 2018). Several studies have been dedicated to improving the LA downstream separation from biowaste based fermentation aiming to reduce production cost and improve sustainability metrics.

Noteworthy, due to the physicochemical characteristics of the biowaste, the synthesis and design of the downstream process for the recovery and purification of LA can be a quite challenging separation task. LA has to be recovered from a highly diluted fermentation broth composed of suspended solids, cell biomass, residual sugars (e.g., glucose, xylose, fructose), bio-macromolecules (e.g., proteins, lipids, polysaccharides, nucleic acids), other carboxylic acids (e.g., formic, acetic, succinic, propionic, butyric acids), alcohols (ethanol, 1-propanol) and ions (e.g., K⁺, Na⁺, Ca²⁺, Mg²⁺).

To accomplish this separation task, many downstream process design alternatives have been described in the literature (Komesu et al., 2017b). In general, the conventional process for LA recovery and purification consists of filtration and centrifugation for biomass removal followed by neutralization and precipitation of the LA with calcium hydroxide yielding calcium lactate. Following this, a filtration process is used to recover the calcium lactate and sulfuric acid is used to dissolve the calcium lactate formed and recover the LA. This step generates large amounts of calcium sulfate (CaSO₄) that need to be regenerated into Ca(OH)₂ and H₂SO₄ or used industrially for production of gypsum boards. Alternatively, LA can be recovered by liquid-liquid extraction or ion exchange from the inorganic salt. Either solvent extraction, separation with membranes, evaporation, crystallization, or distillation processes are applied to concentrate the LA recovered from the previous step. At last, chromatography and ion exchange, are required for the removal of impurities.

For instance, Alvarado-Morales et al. (2021) investigated two downstream processes to recover LA from municipal biopulp based fermentation. A pre-purification step such as centrifugation, ultrafiltration, and activated carbon was in common to the two methods investigated. After the pre-purification step, ion exchange and vacuum distillation were applied in the first method resulting in LA recovery of 75.7 ± 1.5 % and purity of 72.5 ± 2.0 %, respectively. In the second method, a nanofiltration unit was included after the pre-purification step, which resulted in a higher LA purity of 82.0 ± 1.5 % but reducing the recovery to 65.0 ± 1.5 %.

On the other hand, Pleissner et al. (2017) recovered LA from the fermentation of food waste using micro- and nano-filtration units followed by mono- and bi-polar electrodialysis to concentrate the LA and to separate it from the salts. However, the concentration of ions was still high, and then an anion- and cation-exchange step was necessary which resulted in a decrease of Na⁺, K⁺, and Cl⁻ ions to less than 0.01 g/L but also LA concentration was decreased by 70% (54.1 g/L) with respect to its concentration after the electrodialysis step (171 g/L). Finally, to increase LA concentration, evaporation was applied resulting in a final LA concentration of 702 g/L with a recovery of 38% with respect to the LA present in the fermentation broth; clearly, a limitation of this method as 62% of the product was lost. On the other hand, the optical purity of the final product was 99.7% fulfilling the quality requirements for PLA synthesis. The advantages and disadvantages of these separation techniques and the combination of them are very well documented and reviewed in the literature (Komesu et al., 2017b).

Separation technologies for purification such as reactive distillation and molecular distillation have been implemented using emulsion liquid membranes (ELM), liquid membranes in Taylor flow and green ionic liquid ELM (Garcia-Aguirre et al., 2020; Komesu et al., 2017c; Li et al., 2021a; Mai et al., 2018; Murali et al., 2017). The combination of these with well-established separation techniques have opened a new window for design and synthesis of more sustainable and energetically efficient downstream separation processes for LA recovery.

| Table 5 |
|---|
| LA ferment. substrate | Purification strategy | LA recovery | Purity | Reference |
| Municipal biopulp | Centrifugation + ultrafiltration + distillation → ion exchange + distillation | 75.7% | 72.5% | (Alvarado-Morales et al., 2021) |
| Municipal biopulp | Centrifugation + ultrafiltration + distillation → Nanofiltration | 82.0% | 65.0% | (Alvarado-Morales et al., 2021) |
| Food waste | Nanofiltration → Mono + bipolar electrodialysis | 38% | 99.7% | (Pleissner et al., 2017) |
| Reactive distillation | Esterification between ethanol and lactic acid. Catalytical approach | 99.94% | Conc. 34 g/L | (Komesu et al., 2015) |
| Sugarcane | Esterification between ethanol and lactic acid → Ester hydrolysis | 99.95% | | (Mandegari et al., 2017) |
| Molecular distillation + Novel approaches | Extraction efficiency | | | |
| Molecular distillation and adiabatic crystallisation | 95% | | (Van Breugel et al., 2000) |
| ELM extraction | 99% | | (Garavand et al., 2018) |

5.2. Reactive distillation

Reactive distillation is applied specifically to reversible chemical reactions in the liquid phase, in which reaction equilibrium limits the conversion of reactants. Reactive distillation has been proposed as a promising reactive-separation process for the recovery of lactic acid with high recovery and productivity. The LA recovery involves a reversible reaction presented in Eq. (1). The forward reaction represents esterification of LA into ethyl acetate while the reverse reaction represents hydrolysis to LA in the presence of an acidic catalyst.

Lactic acid + ethanol $\rightleftharpoons$ ethyl acetate + H₂O

(1)

Homogeneous catalysts often employed are sulfuric acid and anhydrous hydrogen chloride. However, ion-exchange resins can also be used offering advantages such as low corrosion, ease of separation from the reactive mixture, no side reactions and can be re-used over the homogeneous catalysts. Alcohols such as methanol, ethanol, 2-propanol and butanol can be used in the esterification step. The advantage of using
ethanol is that it can be produced from renewable resources, although butanol and methanol are more attractive options from an economical point of view. The esterification with ethanol or 2-propanol is more expensive since a mass separation agent is needed to break the alcohol/water azeotrope.

Komesu et al. (2015) investigated the esterification reaction between ethanol and lactic acid in a reactive distillation column. The ethyl lactate yield attained was 99.94% under the following conditions: ethanol/LA molar ratio = 18.4, reboiler temperature = 125 °C and catalyst loading = 6 % (w/w). The lactic acid concentration obtained in the hydrolysis step was 34 g/L being 3 times higher than the initial concentration. However, in recent literature the studies for LA recovery are scarce from first and second-generation substrates-based-esterification using reactive distillation. However, simulation approaches using commercial simulators to model/simulate the recovery and purification of lactic acid from second-generation feedstocks-based fermentation have been investigated.

Daful et al. (2016) investigated the environmental performance of lactic acid produced from lignocellulosic biomass and petrochemical sources using a life cycle approach. Lactic acid from the fermentation broth was purified via reactive distillation columns. Likewise, Gezae Daful and Gorgens (2017) performed a techno-economic analysis and environmental impact assessment of a process to produce LA from lignocellulosic feedstocks. The purification and recovery of LA from the fermentation broth consisted of a train of reactive distillation columns including esterification with ethanol and hydrolysis, respectively. Manegar et al. (2017) investigated the co-production of LA and ethanol from sugarcane via a multicriteria analysis based on economic evaluation, energy assessment, and environmental life cycle assessment. Reactive distillation was applied for LA recovery. LA produced from the esterification column was converted to lactic acid ethyl ester by the addition of ethanol, which was further separated by distillation. Pure LA (>99.5 % (w/w)) was subsequently recovered by hydrolysis of the ester in a second reactive distillation column.

5.3 Molecular distillation

Molecular distillation or short path distillation is a nonconventional unit operation of diffusional mass transfer employed for separation of homogeneous liquid mixtures with low volatility, high molecular mass, and high thermosensitivity. It is considered as a special case of evaporation where steam is generated on the liquid surface with the difference that there is practically no return of gaseous molecules to the liquid phase (no vapor-liquid equilibrium). This is achieved by setting the hot evaporation surface and the cold condensation surface closer to each other than the mean free path of the evaporated molecules. Therefore, the evaporated molecules easily reach the condenser, since the route is unobstructed. The distance between the evaporating and condensing surfaces is typically between 1 and 5 cm. In addition, as the process does not involve the use of a solvent as in extractive distillation, the product material is not polluted and no further purification is needed (Komesu et al., 2017c).

The industrial interest in the purification of LA by molecular distillation has been demonstrated by many published patents. Purac Biochem published a method for the industrial-scale purification of LA using molecular distillation and adiabatic crystallization to obtain a 95% pure lactic acid (Van Breugel et al., 2000). Brussels Biotech published a process comprising the following steps: pretreating a diluted solution of lactic acid, concentrating and re-concentrating the diluted solution, and distilling the lactic acid using molecular distillation to obtain the purified lactic acid (Van Gansbegeh et al., 2002). Archer Daniels Midland Company developed a method comprising two-step distillation processes (reactive and molecular distillation) for recovering lactic acid and ethyl lactate (Leboreiro, 2016).

5.4 Emerging extraction technologies

Currently, some new extraction techniques, such as ELM and liquid membrane in Taylor flow, have been successfully used to recover LA from fermentation broth. During the ELM based separation processes, ELM is first obtained upon emulsification of two immiscible phases (organic phase and internal phase) and then dispersing ELM into a third phase i.e. continuous feed phase by stirring for the extraction of low concentrated solute molecules (Li et al., 2021a).

The ELM method has been successfully applied to recover organic acids such as lactic acid from fermentation broth with an extraction efficiency of up to 99% under suboptimal conditions (Garavand et al., 2018). Innovative development of ELM technology is the use of green solvents (or vegetable oils) and ionic liquids to formulate the organic phase. Kumar et al., (2018) investigated the recovery of LA through green emulsion liquid membrane using rice bran oil as a green solvent. Under the optimal process parameters, the LA extraction efficiency was about 90%. Advantages of the ELM method such as large mass transfer area, ease of operation, high extraction efficiency for low solute concentration, and low energy requirements make this technique a promising option to develop more cost-effective downstream processes with less environmental impact. Nevertheless, the poor stability of the ELM technique is the major drawback for large-scale industrial implementation. In this case, liquid membrane in Taylor flow is a novel technology that aims to overcome the stability problems of emulsion systems while keeping the advantages of ELM (Pérez and Fontalvo, 2019).

Pérez et al. (2019) developed a fermentation system for lactic acid production based on a model for a hybrid liquid membrane in Taylor flow. Compared with the traditional batch fermentation, the fermentation time of the hybrid system was decreased by 7 h which is a significant improvement and the productivity and biomass concentration were increased by 2.58 g/L/h and 2.70 g/L, respectively. Unfortunately, this model does not take into account the molecular toxicity of the extrants, and thus the liquid membrane in Taylor flow technology is still in the experimental stage.

Because second generation-lactic acid fermentation is a mixture that is more complex than commercial lactic acid feedstock, due to the presence of residual sugars and other organic acids, the performance of these emerging separation process technologies may be influenced as reactive distillation is affected by the feed composition. In addition, the presence of lipids may adversely affect the reactive distillation process because they can compete with lactic acid in the esterification and hydrolysis reactions, or have a negative effect on the emulsification of two immiscible phases in the ELM process. Therefore, new experimental studies on lactic acid recovery from bio pulp based-fermentation broth with these novel technologies are required to develop more efficient and economically attractive downstream separation processes for industrial applications.

6. Conclusion

Municipal organic waste consists of food waste containing starch and protein and a lignocellulosic fraction of mainly cellulose and hemicellulose. Without proper treatment it is difficult to ferment to lactic acid due to lack of enzymatic hydrolysis of the lignocellulose. The food waste is easier to ferment as it is less recalcitrant. Several lactic acid bacteria can produce the hydrolytic enzymes needed for fermentation process. In cases of starch waste, nitrogen-rich additives such as lucerne green juice could thus replace expensive nutrients such as yeast extract. For downstream processing, reactive distillation gave high purity needed in production of polylactic acid.
Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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