Biorefining Process of Carbohydrate Feedstock (Agricultural Onion Waste) to Acetic Acid

Ho Myeong Kim, † In Seong Choi, † Seoyoun Lee, † Jung Eun Yang, † Seul-Gi Jeong, † Ji Hye Park, † Seung Hee Ko, † In Min Hwang, † Ho Hyun Chun, † Seung Gon Wi, † Jin-Cheol Kim, ‡ and Hae Woong Park*, †

†R&D Division, World Institute of Kimchi, Gwangju 61755, Republic of Korea
‡Asian Pear Research Institute and Division of Applied Bioscience & Biotechnology, Chonnam National University, Gwangju 61186, Republic of Korea

ABSTRACT: The biorefining of agricultural waste into green chemicals has clear potential for improving global environmental sustainability. In this study, we evaluated the potential of acetic acid production from carbohydrate feedstock (onion waste, OW) as a more environmentally friendly source than feedstock produced from natural gas. In particular, OW is an ideal feedstock for the biorefining process as it contains a sufficient amount of carbohydrates (69.7%). Five days of the simultaneous saccharification and two-step fermentation (SSTF) process produced acetic acid from OW more efficiently than the simultaneous saccharification and cofermentation (SSCF) process. SSTF produced 19.3 g/L acetic acid and recorded the highest conversion yield (90.5%) from OW (6% substrate loading, w/v). These results suggested that acetic acid can be efficiently and sustainably produced from OW by the SSTF process.

1. INTRODUCTION

Ongoing development and global population growth result in ever-increasing amounts of waste production. 1 Meanwhile, the biorefining of waste materials to produce green chemicals has become increasingly important worldwide; this can form the foundation of a future economy based on biological chemicals and bioactive compounds. Agricultural waste, which accounts for over 30% of worldwide agricultural productivity, is an ideal resource for producing fermentable sugar such as glucose, fructose, and galactose. 1, 2 For example, onions are a major agricultural product whose global production increased to 97.8 million tons in 2017 owing to their beneficial medicinal and nutritional effects (FAO). Although onions and their by-products are rich in fiber and bioactive compounds with high nutritional value, 500 000 tons of onion waste (OW) are discarded annually in Europe; this is becoming an environmental issue in Spain, the Netherlands, and the United Kingdom. 3−5 Therefore, researching upcycling processes for converting onion waste into green chemicals has become increasingly important, especially in Asia, which accounts for ~63.7% of global onion production.

Acetic acid is generally used as a food preservative or for the production of commercial chemicals, 6 especially vinyl acetate monomer, which polymerizes as a poly(vinyl acetate) for a variety of commercial uses. 7 The global market for acetic acid is expected to increase at a compound annual growth rate of more than 4.30% from 2019 to 2024. 8 The Asia-Pacific region is estimated to lead the market with ~34% of total global demand because of rapid growth in the global textile market. However, as acetic acid production is primarily based on natural gas, the ongoing depletion of global fossil fuel resources makes the diversification of production processes urgent. Diverting this toward agricultural wastes will have a significant positive environmental impact. While the introduction of oxygen into petroleum-based chemicals is chemically difficult, bio-based chemicals naturally contain oxygen. 9

In this study, we investigated one approach to utilizing OW for acetic acid production using Saccharomyces cerevisiae and Acetobacter aceti and compared the simultaneous saccharification and two-step fermentation (SSTF) and simultaneous saccharification and cofermentation (SSCF) methods.

2. RESULTS AND DISCUSSION

2.1. Chemical Composition of OW. The efficient and sufficient use of agricultural feedstock can be improved by the sustainable use of biorefinery processes. 10, 11 Sufficient
carbohydrate content and production yield are necessary to produce acetic acid from agricultural waste. OW can be used for the production of biochemical or bioactive compounds such as bioethanol, rare sugar, and quercetin because it has a sufficient carbohydrate content and efficient bioactive compound.\textsuperscript{1,5} The chemical composition of OW (Table 1) is affected by agronomic methods, harvest time, cultivar, maturity stages, storage time, and environmental conditions.\textsuperscript{12} The OW dry matter tested in this study had 69.7% carbohydrate content composed mainly of glucose (35.1%), fructose (21.5%), sucrose (7.6%), galactose (4.0%), mannose (0.6%), xylose (0.5%), and arabinose (0.4%) (Table 1). Carbohydrate content varies among different biomass types, including wheat straw (57.7%), rice straw (57.3%), rapeseed (68.6%), corn stover (67.7%), corn stalk (62.0%), banana waste (28.0%), and sugarcane bagasse (67.0%).\textsuperscript{13,14} OW clearly has a suitable carbohydrate content for acetic acid production using the fermentation process of \textit{S. cerevisiae} and \textit{A. aceti}.

2.2. Immunogold Labeling Assay of Pectin in Onion. Figure 1 shows examples of overall pectin distribution in our onion samples. The monoclonal antibody LM20, which can recognize the homogalacturonan domain of pectin polysaccharides including the methylester groups, was selected to analyze the distribution of pectin within the onion’s structure.\textsuperscript{15} Pectin is a heteropolysaccharide mainly present in the primary cell wall and middle lamella of terrestrial plants;\textsuperscript{1} in onion, this mainly exists in the parenchyma cell walls.\textsuperscript{1}

We assessed the selectivity of pectin in onion because the structure of the primary cell walls plays a crucial role in their mechanical properties.\textsuperscript{17} Pectin affects the mechanical behavior of the multicellular epidermal surface.\textsuperscript{18} Our transmission electron microscope (TEM) results showed the following distribution of gold particles: parenchyma cell (29.8 particles/\textmu m\textsuperscript{2}), outer layer (13.9 particles/\textmu m\textsuperscript{2}), abaxial epidermis (9.3 particles/\textmu m\textsuperscript{2}), and vessel (3.0 particles/\textmu m\textsuperscript{2}) (Figure 1), suggesting that pectin is a major component of OW, allowing the efficient hydrolyzation of monosaccharides by pectinase treatment.

2.3. Optimization of Enzyme Loading Content. Enzymatic saccharification was conducted on 1% substrate (OW, w/v) with a variable loading of pectinase (0.0–19.2 mg/g OW) and cellulase (0.0–18.4 mg/g OW) at 45 °C. The production of fermentable sugar can be increased by increasing the enzyme loading contents. After a 24 h reaction, the fermentable sugar content was calculated by high-performance liquid chromatography (HPLC) analysis; the enzyme loading results were used to determine that the optimal loading with respect to economic feasibility was 4.8 mg/g OW of pectinase and 4.6 mg/g OW of cellulase (Table 2).

The enzyme demand for the production of fermentable sugar from lignocellulosic biomass is a major factor affecting production cost. Newman et al. (2013) reported that enzyme cost for the production of lignocellulosic sugar accounted for 54 and 25–38% of the total cost for steam-exploded pine wood and corn stover, respectively.\textsuperscript{19} To decrease the enzyme loading content, various pretreatments for lignocellulosic biomass have been used to improve the enzyme accessibility for cellulose and hemicellulose fiber. However, these processes increase the production cost of fermentable sugar. Unlike lignocellulosic biomass, agricultural waste (such as kimchi cabbage waste) can easily produce fermentable sugar using pectinase treatment without a pretreatment.\textsuperscript{20} Similarly, onion

### Table 1. Chemical Composition of Onion Waste

| dry matter (%) | Suc | Glu | Fru | total | Ara | Xyl | Man | Gal | Glu | total | Suc | Fru | total |
|---------------|-----|-----|-----|-------|-----|-----|-----|-----|-----|-------|-----|-----|-------|
| onion waste   | 7.6±0.2 | 25.4±0.4 | 21.5±0.4 | 54.5±0.4 | 0.4±0.0 | 0.5±0.0 | 0.4±0.0 | 0.5±0.0 | 0.4±0.0 | 0.5±0.0 | 0.4±0.0 | 0.5±0.0 | 0.4±0.0 |

\textsuperscript{a} Values represent the average over three replicates. Suc, sucrose; Glu, glucose; Fru, fructose; Ara, arabinose; Xyl, xylose; Man, mannose; and Gal, galactose.
juicing residue (4.2%), onion peel (4.2%), and brown skin (1.5%) are structurally soft with very low lignin content. Therefore, fermentable sugar from OW can be efficiently produced by enzymatic hydrolysis without pretreatment.

2.4. Fermentable Sugar Production by OW Concentration. After enzymatic hydrolysis, the OW samples recorded fermentable sugar contents of 20.1, 38.7, 56.1, 72.3, and 86.3 g/L and conversion yields of 98.2, 94.5, 91.3, 88.3, and 84.4% for the 3, 6, 9, 12, and 15% substrates, respectively (Figure 2). As the substrate concentration increased, the sugar conversion yield decreased and the sugar production content increased. Previous studies documented conversion yields for coffee waste and jack beans of 71.9 and 87.8% for substrate concentrations of 10 and 8%, respectively. These results suggested that OW can be used as a major feedstock for biofining.

2.5. Acetic Acid Production. Hydrothermal reaction and bioconversion processes have been widely used for the production of acetic acid from biomass. In the former, lactic acid is produced from biomass and then converted to acetic acid via an oxidant, obtaining acetic acid yields of 26, 22, and 23% from glucose, cellulose, and starch, respectively. In the latter, two-step fermentation efficiently produces acetic acid by first producing ethanol by yeast such as A. pasteurianus and S. cerevisiae through the fermentation of sugar, then producing acetic acid from ethanol by acetic acid bacteria such as Acetobacter spp. and Gluconacetobacter spp. For example, Wang et al. (2013) reported the production of acetic acid from glucose using a mixed fermentation of S. cerevisiae and A. pasteurianus in a batch and fed-batch culture process.

In this study, we used the SSTF and SSCT processes to determine the best approach for the production of acetic acid from OW. There was a significant difference in acetic acid concentration between fermentation methods (Table 3). Acetic acid concentration increased with the increase of the OW concentration from 2 to 10% (F = 388.1, df = 4,120, P < 0.001) and with the increase of the fermentation period from 1 to 5 days, except at the two least concentrations (2−4%) of OW (F = 721.0, df = 5,120, P < 0.001) (Figures 4 and 5). At 6−10% OW concentrations, with both fermentation methods, the greatest acetic acid concentration was already achieved after 4 days of fermentation. No further increase occurred thereafter.

2.5.1. SSTF. After initial simultaneous saccharification and fermentation (SSF), a significant difference in ethanol production was observed with OW concentrations (F = 676.8, df = 4,10, P < 0.001). The OW samples recorded ethanol concentrations of 6.8 g/L with 2% OW and 31.2 g/L with 10% OW and conversion yields of 95.7 and 87.9%, respectively. Ethanol production increased and ethanol conversion yield decreased slightly when OW concentration increased from 2 to 10% (Figure 3). Subsequently, acetic acid,

---

**Figure 1.** Immunogold labeling analysis of OW sample using LM20. (a) Tunic and (b) abaxial epidermis, (c) parenchyma cells, and (d) vascular bundle of 1st bulb scale. CU: cuticle layer, AE: abaxial epidermis, ML: middle lamella, P: parenchyma, PCW: primary cell wall, VP: vascular parenchyma, SCW: secondary cell wall, and V: vessel. Pectin content in different areas is given below the imagery.
production increased with the increase of OW concentration ($F = 518.9, df = 4, 60, P < 0.001$) and the fermentation period from 0 to 5 days ($F = 468.5, df = 5, 60, P < 0.001$) (Figure 4).

In a similar study, the cofermentation of yeast and acetic acid bacteria strains for 9 days resulted in efficient acetic acid production of 25.88 g/L from food waste.29 In particular, the conversion yield of acetic acid was 73.7% after 5 days, despite

**Table 2. Enzyme Optimization under Different Enzyme Loading Contents**

| onion waste | pectinase (mg/g OW) | cellulase (mg/g OW) | glucose (mg/mL) | fructose (mg/mL) | total (mg/mL) |
|-------------|---------------------|---------------------|-----------------|-----------------|--------------|
| 1           | 0                   | 0                   | 0.253           | 0.216           | 0.469        |
| 2           | 0                   | 2.3                 | 0.264           | 0.217           | 0.481        |
| 3           | 0                   | 4.6                 | 0.273           | 0.222           | 0.494        |
| 4           | 0                   | 9.2                 | 0.295           | 0.228           | 0.523        |
| 5           | 0                   | 18.4                | 0.318           | 0.232           | 0.549        |
| 6           | 2.4                 | 0.0                 | 0.308           | 0.234           | 0.542        |
| 7           | 2.4                 | 2.3                 | 0.312           | 0.234           | 0.546        |
| 8           | 2.4                 | 4.6                 | 0.338           | 0.238           | 0.576        |
| 9           | 2.4                 | 9.2                 | 0.349           | 0.241           | 0.590        |
| 10          | 2.4                 | 18.4                | 0.355           | 0.245           | 0.600        |
| 11          | 4.8                 | 0.0                 | 0.319           | 0.235           | 0.554        |
| 12          | 4.8                 | 2.3                 | 0.334           | 0.241           | 0.575        |
| 13          | 4.8                 | 4.6                 | 0.340           | 0.247           | 0.587        |
| 14          | 4.8                 | 9.2                 | 0.35            | 0.249           | 0.599        |
| 15          | 4.8                 | 18.4                | 0.362           | 0.249           | 0.612        |
| 16          | 9.6                 | 0.0                 | 0.326           | 0.241           | 0.567        |
| 17          | 9.6                 | 2.3                 | 0.335           | 0.242           | 0.577        |
| 18          | 9.6                 | 4.6                 | 0.347           | 0.247           | 0.595        |
| 19          | 9.6                 | 9.2                 | 0.354           | 0.251           | 0.605        |
| 20          | 9.6                 | 18.4                | 0.370           | 0.251           | 0.622        |
| 21          | 19.2                | 0.0                 | 0.332           | 0.241           | 0.573        |
| 22          | 19.2                | 2.3                 | 0.336           | 0.242           | 0.578        |
| 23          | 19.2                | 4.6                 | 0.347           | 0.251           | 0.598        |
| 24          | 19.2                | 9.2                 | 0.380           | 0.252           | 0.631        |
| 25          | 19.2                | 18.4                | 0.386           | 0.253           | 0.639        |

*Values represent the average over three replicates.

**Table 3. Analysis of Variance Results for the Effect of Fermentation Method, OW Concentration, and Time on Acetic Acid Production**

| df | SS  | MS  | $F$ value | $P$ value |
|----|-----|-----|-----------|-----------|
| method | 1   | 952.7 | 952.7 | 918.1 | <0.001 |
| OW conc. | 4   | 1611.0 | 402.7 | 388.1 | <0.001 |
| time | 5   | 3740.1 | 748.2 | 721.0 | <0.001 |
| method × OW conc. | 4   | 788.1 | 197.0 | 189.9 | <0.001 |
| method × time | 5   | 273.4 | 54.7 | 52.7 | <0.001 |
| OW conc. × time | 20  | 1119.4 | 56.0 | 53.9 | <0.001 |
| method × OW conc. × time | 20  | 529.9 | 26.5 | 25.5 | <0.001 |
| residuals | 120 | 124.5 | 1.0  | 1.0  | 1.0  |

*SS: sample size; MS: mean square.*
the high production of acetic acid (23.0 g/L) from ethanol (31.2 g/L) achieved at 10% (w/v) onion substrate (Figure 3). It is thought that A. aceti can tolerate acetic acid concentrations of up to 2.3%. These results indicated that OW with a 6% (w/v) substrate concentration was suitable for acetic acid production due to its high conversion yield (90.5%). At lower ethanol concentrations, the acetic acid production tended to decrease over time as it oxidized to CO₂ and H₂O.³⁰ Thus, we achieved good acetic acid productivity using the rapid processing method.

2.5.2. SSCF. SSCF has been widely used for the production of bioethanol from xylose-rich lignocellulosic biomass.³¹ In this study, sugar and ethanol were co-fermented at 32 °C using SSCF for the efficient production of ethanol because the optimum temperature for S. cerevisiae is 32 °C ± 2, while that for acetic acid bacteria is 30 °C.

After a 5 day processing, acetic acid yields of 1.0–14.9 g/L were obtained from OW substrate concentrations (v/w) of 2–10% (F = 39.5, df = 4,60, P < 0.001) (Figure 5). Acetic acid concentration increased with the fermentation period (F = 289.2, df = 4,60, P < 0.001). The initial acetic acid productivity was higher with a lower substrate concentration, but the highest acetic acid output was recorded at 4–10% (w/v) OW substrate concentration. Overall, SSTF was more efficient than SSCF for acetic acid production because it optimized ethanol production (F = 918.1, df = 1,120, P < 0.001) (Table 3). Also, acetic acid strongly inhibits the SSF process for enzymatic hydrolysis and ethanol fermentation.³⁴ These results suggest that considering economic feasibility and productivity, SSTF is more suitable than SSCF for the production of acetic acid from OW. However, SSCF offers several advantages despite its lower productivity, including reduced capital cost and continuous removal of end-production inhibition on enzymatic hydrolysis.³⁵ Some studies have reported that SSF is more productive than separate hydrolysis and fermentation (SHF) in ethanol production.³⁶,³⁷ For example, ethanol production by SSF from corn stover recorded a 13% higher ethanol conversion yield than SHF.

2.6. Overall Mass Balance for the Production of Acetic Acid. After the comparison of the SSTF and SSCF processes, we designed an overall mass balance for the ideal production of acetic acid from OW, in which 100 kg of OW contained 7.6 kg sucrose, 35.1 kg glucose, and 21.5 kg fructose (Figure 6). For SSTF, ethanol production (6% substrate loading, v/w) was conducted with cellulase, pectinase, and S. cerevisiae at 32 °C for 48 h; then, acetic acid production was conducted with A. aceti at 30 °C for 72 h, resulting in acetic acid production of 32.2 kg. For SSCF, acetic acid production (6% substrate loading, v/w) was conducted with cellulase, pectinase, S. cerevisiae, and A. aceti at 32 °C for 120 h, resulting in an acetic acid production of 24.9 kg.

3. CONCLUSIONS

This study evaluated the possibility of producing acetic acid from OW. OW contains a sufficient amount of monosaccharides (69.7%). The SSTF and SSCF processes for 60 g of OW (6% substrate loading, v/w) can produce 19.3 and 14.9 g of acetic acid, respectively. Thus, we conclude that OW can be a major natural feedstock, and SSTF is an ideal process for the production of acetic acid.

4. EXPERIMENTAL SECTION

4.1. Raw Materials and Chemical Composition Analysis. We obtained OW from an onion field in Muan, South Korea. The OW was dried by a lyophilizer at −80 °C for 7 days, ground to particles using an electric grinder, and stored at −20 °C until further use. The soluble sugar content (sucrose, glucose, and fructose) of the OW was analyzed by high-performance liquid chromatography (HPLC) with a refractive index detector (2414; Waters, Milford, MA); a REZEX RPM (Phenomenex, Torrance, CA) column (300 × 7.8 mm) was used at 85 °C by adding deionized water at a flow rate of 0.6 mL/min. The insoluble sugar content (arabinose, xylose, mannose, galactose, and glucose) of the OW was analyzed by gas chromatography (GC). The sample pretreatment and GC analysis method followed that given by Choi et al.³⁵

4.2. Immunogold Labeling Assay of Pectin in Onion. Onion pieces were fixed in 4 °C for 6 h using 0.1% glutaraldehyde (v/v) and 4% paraformaldehyde (v/v) in a 50 mM sodium cacodylate buffer (pH 7.2). After washing with a
sodium cacodylate buffer, the samples were dehydrated in a graded ethanol series and embedded in LR white resin (London Resin Co., London, U.K.). Ultrathin sections (80 nm) were mounted on nickel grids of 300 mesh. After washing, the grids were treated with LM20 primary antibodies (PlantProbes, Leeds, U.K.) at 4 °C for 2 days, then labeled with an anti-Rat IgG antibody (Sigma, St. Louis, MO) conjugated to 10 nm gold particles at 25 °C for 3 h. After staining with 4% uranyl acetate, the sections of pectin distribution were studied using a transmission electron microscope (TEM, JEM-1400; JEOL, Tokyo, Japan).

4.3. Optimization of Enzyme Loading Content. We purchased cellulase (Celluclast 1.5 L) and pectinase (Pectinex SP-L) from Novozyme A/S (Bagsvaerd, Denmark) for enzymatic saccharification of OW. The cellulase and pectinase activities were 0.356 filter paper unit/mg protein and 240 international unit (IU)/mg protein, respectively. To optimize the enzyme loading content, enzymatic saccharification was performed on 1% substrate (OW, w/v) with variable loading of pectinase (0.0–19.2 mg/g of OW) and cellulase (0.0–18.4 mg/g of OW) for 24 h at 45 °C. After the enzymatic hydrolysis, the soluble sugar contents were calculated by HPLC with a glucose and fructose standard curve.

4.4. Enzymatic Saccharification According to OW Concentration. The enzymatic saccharification of OW was conducted in 100 mL of citrate buffer (0.05 M, pH 4.8) containing 3–15% (w/v) dry matter, pectinase (4.8 mg/g OW), and cellulase (4.6 mg/g OW) at 45 °C in a 500 mL Erlenmeyer flask. After 48 h incubation, soluble sugar content and sugar conversion yield were calculated using the standard curve and initial sugar content (69.7%) of the OW, respectively.

4.5. Acetic Acid Production. We tested both the SSTF and SSCP processes to determine the best method for acetic acid production from OW.

4.5.1. SSTF. We first produced ethanol from 100 mL of OW containing 2.0–10.0% (w/v) dry matter, pectinase (4.8 mg/g OW), and cellulase (4.6 mg/g OW), 1 mL of S. cerevisiae (KCCM 11293), 0.3% yeast extract, 0.3% malt extract, and 0.5% peptone at 32 °C for 48 h. After the reaction, acetic acid fermentation for 100 mL hydrolysates (2.0–10.0%) with 1 mL of A. aceti (KCCM 40229) was conducted at 30 °C for 5 days. The ethanol and acetic acid content were analyzed by HPLC using a refractive index detector (2489; Waters) at 210 nm and Aminex HPX-87H (Bio-Rad, Hercules, CA) column (300 × 7.8 mm) and with a UV-vis detector (2489; Waters) at 210 nm and Aminex HPX-87H (Bio-Rad, Hercules, CA) column (300 × 7.8 mm), respectively.

4.5.2. SSCP. We produced acetic acid using 100 mL of OW containing 2.0–10.0% (w/v) dry matter, pectinase (4.8 mg/g OW), and cellulase (4.6 mg/g OW), 1 mL of S. cerevisiae, 1 mL of A. aceti, 0.3% yeast extract, 0.3% malt extract, and 0.5% peptone at 32 °C for 5 days. After the reaction, the acetic acid content was analyzed by HPLC.

4.5.3. Statistical Analysis. All experiments were triplicated in a completely randomized block design. Data were analyzed using PASW software (Version 17, SPSS Inc., CA). Analysis of variance tests was used to determine the significant differences between treatments at P < 0.05 using Tukey’s honestly significant difference test. Data are expressed as means with standard deviations.

## AUTHOR INFORMATION

Corresponding Author

*E-mail: haewoong@wikim.re.kr. Tel: +82-62-610-1728. Fax: +82-62-610-1850.

ORCID

Hae Woong Park: 0000-0002-5181-1255

Notes

The authors declare no competing financial interest.

## ACKNOWLEDGMENTS

This research was supported by the World Institute of Kimchi (KE1901-1), funded by the Ministry of Science and ICT, Republic of Korea.

## REFERENCES

(1) Kim, H. M.; Song, Y.; Wi, S. G.; Bae, H. J. Production of D-tagatose and bioethanol from onion waste by an integrating bioprocess. *J. Biotechnol.* 2017, 260, 84–90.
(2) Ashworth, G.; Azevedo, P. Agricultural Wastes: Agriculture Issues and Policies; Nova Science Pub Inc: U.K., 2009. ISBN: 978-1-60741-305-9.
(3) Arshad, M. S.; Sohail, M.; Nadeem, M.; Saeed, F.; Imran, A.; Javed, A.; Amjad, Z.; Batool, S. M. Status and trends of nutraceuticals from onion and onion by-products: a critical review. *Cognet Food Agric.* 2017, 3, No. 1280254.
(4) Benitez, V.; Molli, E.; Martin-Cabrejas, M. A.; Aguiler, Y.; Lopez-Andreu, F. J.; Cools, K.; Terry, L. A.; Esteban, R. M. Characterization of industrial onion wastes (*Allium cepa L.*): Dietary fibre and bioactive compounds. *Plant Foods Hum. Nutr.* 2011, 66, 48–57.
(5) Choi, I. S.; Cho, E. J.; Moon, J. H.; Bae, H. J. Onion skin waste as a valorization resource for the by-products quercerin and biosugar. *Food Chem.* 2015, 188, 537–542.
(6) Awad, H. M.; Diz, R.; Malek, R. A.; Othman, N. Z.; Aziz, R. A.; Enshasy, H. A. Efficient production process for good grade acetic acid by *Acetobacter aceti* in shake flask and in bioreactor cultures. *E-J. Chem.* 2012, 9, 2275–2286.
(7) Ilkgur, F. H.; Becer, C. R. Lignocellulosic biomass: a sustainable platform for the production of bio-based chemicals and polymers. *Polymer.* 2015, 6, 4497.
(8) Mordor Intelligence. Acetic Acid Market — Segmented by Application, End-user Industry, and Geography - Growth, Trends, and Forecast (2019–2024). 2018, https://www.mordorintelligence.com/industry-reports/acetic-acid-market.
(9) Dale, B. E. ‘Greening’ the chemical industry: research and development priorities for biobased industrial products. *J. Chem. Technol. Biotechnol.* 2003, 78, 1093–1103.
(10) Alexandri, M.; Schneider, R.; Papapostolou, H.; Ladakis, D.; Koutinas, A.; Venus, J. Restructuring the conventional sugar beet industry into a novel biorefinery: fractionation and biocconversion of sugar beet pulp into succinic acid and value-added coproducts. *ACS Sustainable Chem. Eng.* 2019, 7, 6569–6579.
(11) de Jong, E.; Higson, A.; Walsh, P.; Wellics, M. Bio-based chemicals, value added products from biorefineries IEA Bioenergy, Task42 Biorefinery, 2012.
(12) Pokluda, R. Nutritional quality of Chinese cabbage from integrated culture. *Hort. Sci.* 2008, 35, 145–150.
(13) Rezania, S.; Din, M. F. M.; Mohamad, S. E.; Sohali, J.; Taib, S. M.; Yusof, M. B. M.; Kamyab, H.; Darajeh, N.; Ahsan, A. Review on pretreatment methods and <tep-common:author-query>AQ1: Please provide the DOI number for ref 13, or indicate if the DOI number does not exist.</tep-common:author-query>ethanol production from cellulosic water hyacinth. *BioResources* 2017, 12, 2108–2124.
(14) Shahzadi, T.; Mehmoond, S.; Irshad, M.; Anwar, Z.; Afroz, A.; Zeeshan, N.; Rashid, U.; Sughrata, K. Advances in lignocellulosic
biotechnology: a brief review on lignocellulosic biomass and cellulases. Adv. Biosci. Biotechnol. 2014, 5, 246–251.
(15) Verberghbrugen, Y.; Marcus, S. E.; Haeger, A.; Ordaz-Ortiz, J. J.; Knox, J. P. An extended set of monoclonal antibodies to pectic homogalacturonan. Carbohydr. Res. 2009, 344, 1858–1862.
(16) Cañal, K. H.; Mohnen, D. The structure, function, and biosynthesis of plant cell wall pectic polysaccharides. Carbohydr. Res. 2009, 344, 1879–1900.
(17) Cosgrove, D. J. Plant cell wall extensibility: connecting plant cell growth with cell wall structure, mechanics, and the action of wall-modifying enzymes. J. Exp. Bot. 2016, 67, 463–476.
(18) Kim, K.; Yi, H.; Zamil, M. S.; Haque, M. A.; Puri, V. M. Multiscale stress-strain characterization of onion outer epidermal tissue in wet and dry states. Am. J. Bot. 2015, 102, 12–20.
(19) Newman, R. H.; Vaidya, A. A.; Sohel, M. I.; Jack, M. W. Optimizing the enzyme loading and incubation time in enzymatic hydrolysis of lignocellulosic substrates. Bioresour. Technol. 2013, 129, 33–38.
(20) Kim, H. M.; Park, J. H.; Choi, I. S.; Wi, S. G.; Ha, S.; Chun, H. H.; Hwanh, I. M.; Chang, J. Y.; Choi, H. J.; Kim, J. C.; Park, H. W. Effective approach to organic acid production from agricultural kimchi cabbage waste and its potential application. PLoS One 2018, 13, No. e0207801.
(21) Jaime, L.; Molla, E.; Fernandez, A.; Martin-Cabrejas, M. A.; Lopez-Andreu, F. J.; Esteban, R. M. Structural carbohydrate differences and potential source of dietary fiber of onion (Allium cepa L.) tissues. J. Agric. Food Chem. 2002, 50, 122–128.
(22) Suutarinen, M.; Mustranta, A.; Autio, K.; Salmenkallo-Marttila, M.; Alvenainen, R.; Bucht, J. The potential of enzymatic peeling of vegetable. J. Sci. Food Agric. 2003, 83, 1556–1564.
(23) Kim, H. M.; Cho, E. J.; Bae, H. J. Single step purification of concanavalin A (ConA) and bio-sugar production from jack bean using glucosylated magnetic nano matrix. Bioresour. Technol. 2016, 213, 257–261.
(24) Kim, H. M.; Choi, Y. S.; Lee, D. S.; Kim, Y. H.; Bae, H. J. Production of bio-sugar and bioethanol from coffee residue (CR) by acid-chlorite pretreatment. Bioresour. Technol. 2017, 236, 194–201.
(25) Jin, F.; Zhou, Z.; Kishita, A.; Enomoto, H.; Kishida, H.; Moriya, T. A new hydrothermal process for producing acetic acid from biomass waste. Chem. Eng. Res. Des. 2007, 85, 201–206.
(26) Parmar, I.; Rupasinghe, H. P. V. Bio-conversion of apple pomace into ethanol and acetic acid: Enzymatic hydrolysis and fermentation. Bioresour. Technol. 2013, 130, 613–620.
(27) Huo, Z.; Fang, Y.; Yao, G.; Zeng, X.; Ren, D.; Jin, F. Improved two-step hydrothermal process for acetic acid production from carbohydrate biomass. J. Energy Chem. 2015, 24, 207–212.
(28) Wang, Z.; Yan, M.; Chen, X.; Li, D.; Qin, L.; Li, Z.; Yao, J.; Liang, X. Mixed culture of Saccharomyces cerevisiae and Acetobacter pasteurianus for acetic acid production. Biochem. Eng. J. 2013, 79, 41–45.
(29) Li, Y.; He, D.; Niu, D.; Zhao, Y. Acetic acid production from food wastes using yeast and acetic acid bacteria micro-aerobic fermentation. BioProcess Biosyd. Eng. 2015, 38, 863–869.
(30) Gomes, R. J.; Borges, M. F.; Rosa, M. F.; Castro-Gómez, R. J. H.; Spinosa, W. A. Acetic acid bacteria in the food industry:系统atics, characteristics and applications. Food Technol. Biotechnol. 2018, 56, 139–151.
(31) Olofsson, K.; Palmqvist, B.; Liden, G. Improving simultaneous saccharification and co-fermentation of pretreated wheat straw using both enzyme and substrate feeding. Biotechnol. Biofuels 2010, 3, 17.
(32) Mukhtar, K.; Asgher, M.; Afghan, S.; Hussain, K.; Zia-Ul-Hussain, S. Comparative study on two commercial strains of Saccharomyces cerevisiae for optimum ethanol production on industrial scale. J. Biomed. Biotechnol. 2010, No. 419586.
(33) Saichana, N.; Matsushita, K.; Adachi, O.; Frébort, I.; Frébortová, J. Acetic acid bacteria: A group of bacteria with versatile biotechnological applications. Biotechnol. Adv. 2015, 33, 1260–1271.
(34) Feng, Y.; Qj, X.; Jian, H.; Sun, R.; Jiang, J. Effect of inhibitors on enzymatic hydrolysis and simultaneous saccharification fermenta-