



REVIEW

# Agave biomass: a potential resource for production of value-added products

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## Abstract

The recent research has been focussed on the alternative renewable energy sources. Lignocellulosic biomass has emerged as a potential alternate for biofuels, industrial enzymes, and organic acids production. There is great interest in finding new biomass sources (for biofuels) that can be grown in semi-arid and arid lands. The biomass generated by the plants related to the genus *Agave* is one of the promising sources for biotechnological conversion into various value-added products. This review describes the biomass generation by different species of *Agave* and the composition of *Agave* residues. The recent research and developments focused on the conversion of *Agave* biomass and residues into different useful products such as bioethanol, biobutanol, biogas and biomethane, biohydrogen, cellulases, xylanases, inulinases, polyhydroxybutyrate, and succinic acid have also been discussed.

**Keywords** Agave bagasse · Tequila industry · Bioethanol · Biomethane · Biobutanol · Lignocellulolytic enzymes

## Introduction

The environmental issues are associated with the utilization of fossil fuels that are depleting continuously. Therefore, renewable and environmentally sustainable sources of energy are required for the society. The use of lignocellulosic biomass (LCB) for biofuels and other value-added products has emerged as a sustainable alternative to fossil fuels. LCB is one of the most abundantly available and renewable biomass resource on earth (Budzianowski 2017; Cunha et al. 2019). Plants of the genus *Agave* have the potential for biofuels and high value-added products. *Agave* presents several advantages over other plants such as lower water requirement and high productivity in arid and semi-arid areas (Subedi et al. 2017). Furthermore, plants that belong to genus *Agave* are well adapted to high temperatures and drought conditions. *Agave* is a genus of 200–300 species

that are available around the world but the majority of them are native of Mexico (Sarah et al. 2011; Perez-Pimienta et al. 2017a; Silva-mendoza et al. 2020). Although, *Agave* is the native of Mexico and Central America but it propagated around the globe including East Africa, Philippines, Indonesia, and Australia. *Agave americana*, *Agave angustifolia*, *Agave fourcroydes*, and *Agave sisalana* are among the species that are commonly found in the warmer regions of the world. Historically, it was grown for food, animal feed, fiber, ornamental beauty, and alcoholic beverages (Cedeno-Cruz 2003; Mielenz et al. 2015). Different species of *Agave* are chiefly grown for the manufacturing of alcoholic beverages like tequila, mezcal, bacanora, and pulque from fructose. Stem and basal leaves combinedly known as core is used for the extraction of fructose (Caspeta et al. 2014; Láinez et al. 2019). The genus *Agave* belongs to the monocot family Agavaceae and plants of this genus are succulent. *Agave* plants have thick and fleshy leaves that generally end in to sharp points (Escamilla-treviño 2012). In Mexico, the tequila industry generates a huge quantity of agave bagasse and most of this is disposed off in the fields that generates leachate and odor polluting the environment. Moreover, it also provides the suitable conditions for pests and disease generation (Crespo et al. 2013; Valdez-guzmán et al. 2019). Therefore, proper management of this biomass waste is necessary. The valorization of agave biomass wastes into

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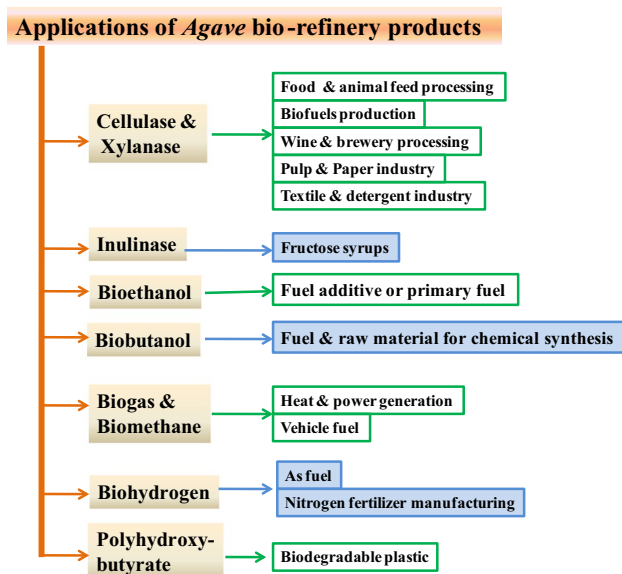
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value-added products is an environmentally viable option. *Agave* leaves and waste biomass have been employed for the production of cellulases and xylanases (Huitron et al. 2008; Contreras-hernández et al. 2018; Silva-mendoza et al. 2020). *Agave* biomass waste is a promising source for bioenergy due to abundant availability and renewability. Moreover, it does not compete with human food demand. The bioenergy products and enzymes find several important applications (Fig. 1) (Breton-Deval et al. 2018; Valdez-guzmán et al. 2019).

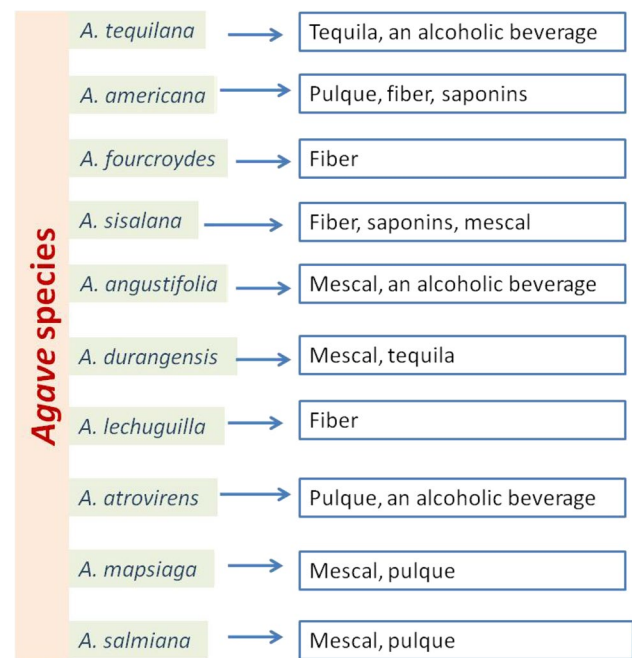
*Agave* biomass is a sustainable resource for the generation of biofuels and biochemicals. This review provides the very recent research and development in pretreatment technology, hydrolysis of *agave* biomass and fermentation of reducing sugars into valuable products.

## Agave biomass generation and chemical composition

Different species of *Agave* are cultivated for beverage and other purposes that generate a large amount of residues (Fig. 2). In this section of the review, the generation of different types of residues from various species of *Agave* has been discussed. *Agave tequilana* (blue agave) is dominantly cultivated for the manufacturing of tequila, a Mexican alcoholic beverage (Escamilla-treviño 2012). It is a perennial arid plant that needs 8–12 years for maturation and the fructans are accumulated in the pinecone of this plant. Pinecones are extracted for syrup that is used for the production of Tequila. The extraction process generates a fibrous



**Fig. 1** Value-added products produced from agave biomass and their applications



**Fig. 2** Different species of *Agave* and their traditional products

residue that is known as bagasse (Barrera et al. 2016; Palomo-Briones et al. 2018). This generated bagasse is approximately 40% of initial dry mass of pinecons. In 2018, 346,700 tons of tequila bagasse was generated in Mexico (Cedeno-Cruz 2003; Palomo-Briones et al. 2018; Gonz et al. 2019).

*Agave durangensis* is another very important species that is utilized for the production of tequila and mezcal in Mexico. During the preparation of these beverages, the leaves are separated from the pine that is used for the production of alcoholic beverages. The by-product in the form of leaves has potential for their valorization into value-added products. Approximately, one million tons of *A. durangensis* are produced and processed that generates 1200 tons of leaves (Contreras-hernández et al. 2018). *Agave lechuguilla* is a non-timber forest plant that is a commonly found in arid and semiarid areas of northern Mexico. The heart or pulp stem along with attached leaf bases is known as cogollos that are harvested several times without harvesting complete plant. *A. lechuguilla* generates 4 tons of biomass per hectare of land annually with an average rainfall of 427 mm (Castillo et al. 2013; Morales-Martínez et al. 2017; Díaz-Blanco et al. 2018).

*Agave salmiana* is another important species that is available in the semiarid areas of north-central Mexico. It has massive leaves compared to other wild agaves and shows approximately 10-fold productivity compared to desert regions. This species along with *A. americana*, *Agave atrovirens*, and *Agave mapiaga* are placed in a group colloquially known as “magueys plqueros”. These three species generate

high economic revenue after *A. tequilana* in Mexico (Alfaro Rojas et al. 2007; Garcia-Moya et al. 2011; Láinez et al. 2018). Alcoholic beverages are produced from the stem of *A. salmiana* while the remaining residues including leaves are generally discarded. The leaves represent about 40–60% of the plant and constitute huge unused biomass that can be converted into several biotechnological products such as biofuels and biochemicals (Zamudio et al. 2009; Láinez et al. 2018).

LCB is composed of cellulose (30–60%), hemicelluloses (20–40%), and lignin (10–25%), extractives and minerals that are intermeshed with one another (Kumar et al. 2019a). The agave biomass and leaves are the potential sources of carbohydrates i.e. cellulose and hemicelluloses. Therefore, the agave residues represent a potential resource for generation of biofuels and other useful products. The chemical composition of various types of *Agave* residues is depicted in Table 1. The pretreatment and hydrolysis analysis of agave biomass confirmed that it contains higher content of amorphous cellulose and low content of lignin that makes agave biomass less recalcitrant for hydrolysis compared to other LCBs (Cushman et al. 2015).

## Pretreatment and hydrolysis of *Agave* residues

Cellulose and hemicelluloses are carbohydrates that make up to two-thirds of LCB that are recalcitrant to microbial attack and enzymatic hydrolysis due to availability of cementing layer of lignin (Gírio et al. 2010; Kumar et al. 2019a). Cellulose, hemicelluloses, and lignin together form a three-dimensional complex structure having covalent and non-covalent interactions (Sanchez 2009). The pretreatment process removes lignin and enhances the digestibility of cellulose to overcome its recalcitrance for enzymatic hydrolysis and fermentation stages. Pretreated lignocellulosic biomass acts as feedstock for the production of second-generation biofuels (Kumar et al. 2018). Various physical and chemical pretreatments such as steam explosion, ammonium fiber expansion, dilute acid, lime, and organic solvents have been proved to be effective for minimizing the recalcitrance of LCB for subsequent enzymatic hydrolysis (Mosier et al. 2005; Perez-Pimienta et al. 2015). The development of the pretreatment technology aims some general targets such as improved fractionation, minimum sugar degradation, and the minimum generation of inhibitors that directly affect the fermentation process (Sun et al. 2016; Aguirre-Fierro et al. 2020). Therefore, several kinds of pretreatments have been tested for the efficient utilization of agave biomass.

The pretreated lignocellulosic biomass is hydrolyzed either by acid or enzymatic hydrolysis. Acid hydrolysis

**Table 1** Chemical composition of *Agave* biomass residues

| Agave species  | Type of residue                        | Cellulose  | Hemi-celluloses | Lignin    | References                            |
|----------------|--|------------|-----------------|-----------|---------------------------------------|
| A. tequilana   | Bagasse                                | 45.6±1.6   | 22.8±0.7        | 19.3±1.5  | Perez-Pimienta et al. (2013)          |
|                |  | 42±2       | 20±1            | 15±1      | Saucedo-Luna et al. (2011)            |
|                |  | 42         | 22              | 18        | Corona-gonzález et al. (2016)         |
|                |  | 30.51±0.2  | 20.72±3.8       | 18.78±0.8 | Aguilar et al. (2018)                 |
|                | Bagasse (hydrothermal pretreatment)    | 65.78±1.6  | 30.20±0.77      | 7.87±0.08 | Aguilar et al. (2018)                 |
| A. tequilana   | Depithed leaves                        | 64.8       | 5.1             | 15.9      | Iñiguez-Covarrubias et al. (2001)     |
| A. lechuguilla | Biomass                                | 79.8       | 3–6             | 15.3      | Vieira et al. (2002)                  |
|                | Cogollos <sup>a</sup> (untreated)      | 20.18±1.7  | 11.08±0.8       | 21.79±0.5 | Díaz-Blanco et al. (2018)             |
|                | Cogollos (acid-pretreated)             | 40.81±1.8  | 1.61±0.46       | 39.92±3.1 | Díaz-Blanco et al. (2018)             |
|                | Cogollos (autohydrolysis pretreatment) | 40.98±1.2  | 1.52±0.47       | 44.25±0.7 | Morales-Martínez et al. (2017)        |
| A. fourcroydes | Biomass                                | 77.6       | 5–7             | 13.1      | Vieira et al. (2002)                  |
| A. sisalana    | Leaf fiber                             | 43         | 32              | 15        | McDougall et al. (1993)               |
| A. salmiana    | Bagasse                                | 47         | 13              | 10        | Garcia-Reyes and Rangel-Mendez (2009) |
| A. americana   | Leaf fiber                             | 68         | 16              | 4.9       | Mylsamy and Rajendran (2010)          |
|                | Stalk (untreated)                      | 37.5 (Glu) | 16 (Xyl+Gal)    | 18 (KL)   | Yang and Pan (2012)                   |
|                | Stalk (dilute acid pretreated)         | 51.2(Glu)  | 7.9 (Xyl+Gal)   | 31.8 (KL) | Yang and Pan (2012)                   |
|                | Stalk (NaOH pretreated)                | 58.8 (Glu) | 12.4(Xyl+Gal)   | 21.8 (KL) | Yang and Pan (2012)                   |

Gal galactose, Glu glucose, Xyl xylose, KL Klason lignin

<sup>a</sup>Cogollos: heart with attached leaf bases,

utilizes dilute or concentrated acids including  $\text{H}_2\text{SO}_4$ ,  $\text{H}_2\text{SO}_3$ ,  $\text{HCl}$ ,  $\text{HF}$ ,  $\text{H}_3\text{PO}_4$ ,  $\text{HNO}_3$ , and  $\text{HCOOH}$ . Acidic methods are simple but they have disadvantages such as strong equipment corrosion, inadequate acid recovery, and the requirement of a large amount of acids (Kumar et al. 2019a). Dilute acid hydrolysis methods need 2–5% of acid that is significantly lower than concentrated acid hydrolysis methods. Dilute acid hydrolysis pretreatment is performed at a higher temperature to achieve acceptable hydrolysis yield. At higher temperature fermentation inhibitors such as furfural and hydroxymethyl-furfural are generated (Verardi et al. 2012). Enzymatic hydrolysis is an environment-friendly alternative for acidic hydrolysis. It is performed at mild environmental conditions that need lower energy. LCB hydrolysis by enzymes is less corrosive process compared to acidic hydrolysis. Moreover, enzymatic hydrolysis does not release inhibitory compounds that have negative impact on fermentation process (Sarkar et al. 2012; Kumar et al. 2018; Alfaro and Arias 2020).

Aguirre-Fierro et al. (2020) used a high-pressure  $\text{CO}_2$ – $\text{H}_2\text{O}$  pretreatment method for *A. tequilana* bagasse conversion into ethanol. The solid phase obtained from high-pressure  $\text{CO}_2$ – $\text{H}_2\text{O}$  mixture pretreatment at temperature 150–190 °C, was subjected to enzymatic hydrolysis. The enzymatic saccharification resulted in a yield of 75 mol% of polysaccharides available in *A. tequilana* bagasse that were fermented into ethanol.

The response surface methodology (RSM) based on factorial central composite design was applied for optimization of dilute acid and alkaline pretreatment of agave bagasse. The solid loadings from 3 to 30% were tested to obtain the maximum release of sugar during enzymatic saccharification by Novozyme Cellic CTec2 and HTec2. The alkaline pretreatment by 1.87% of NaOH for 50.3 min with 13.1% solid loading was found most suitable for hydrolysis. The acid concentration of 2.1%, solid loading of 8.5%, and the hydrolysis time of 33.8 min were the optimal conditions for dilute acid pretreatment (Ávila-lara et al. 2015). The acid hydrolysis of *A. tequilana* Weber bagasse was optimized. The hydrolysis was performed in two sequential batches; the first stage resulted in 236 g/kg of dry matter and 26.9 g/L of fermentable sugars at optimized conditions i.e. 151 °C, 2%  $\text{H}_2\text{SO}_4$ , and 10 min of reaction time. In the second stage, biomass produced in the first stage was hydrolyzed for 30 min at 175 °C, 2%  $\text{H}_2\text{SO}_4$  and the hydrolysis released 15 g/L of fermentable sugars along with 90 g/kg of dry matter. The sequential batch stages produced 326 g fermentable sugars from 1 kg of dry matter that corresponds to a theoretical value of 48.50% (Saucedo-Luna et al. 2010). Delfín-Ruiz et al. (2019) conducted acidic pretreatment for xylose production from the bagasse of four different Agave species including *A. tequilana* Weber var. *azul* (AT), *A. americana* var. *oaxacensis* (AA), *A. karwinskii* (AK), and

*A. potatorum* (AP). Box–Behnken design was used for the optimum release of xylose and minimum generation of acetic acid. The xylose concentrations of 23.18, 27.63, 31.8, and 24.42 g/L were obtained from AT, AA, AK, and AP bagasse respectively. The higher concentration of  $\text{H}_2\text{SO}_4$  resulted in improved xylose concentration, but also a higher concentration of acetic acid (1.02 g/L in AT, 2.3 g/L in AA, 3.46 g/L in AK, and 2.01 g/L in AP). This study indicated that the bagasse from these species of *Agave* is a promising feedstock for bioethanol and xylitol production using dilute acid pretreatment (Delfín-Ruiz et al. 2019).

Recently, ionic liquids as pretreatment agents have gained attention and several studies have demonstrated that imidazolium-based ionic liquids have potential for biomass pretreatment. Ionic liquids are relatively environmentally beneficial compared to acidic, alkaline and organosolv pretreatment methods. Ionic-liquids show several environmental advantages such as low vapor pressure, non-flammability, and phase behavior. Ionic-liquids also have higher chemical and thermal stability. Moreover, these pretreatments make easy recovery of cellulose with the addition of antisolvents such as water or ethanol (Singh et al. 2009; Perez-Pimienta et al. 2013, 2017b; Trinh et al. 2015). Furthermore, ionic liquids pretreatment also results in improved enzymatic hydrolysis as well as effective recovery of reducing sugars without their degradation as compared to traditional pretreatment methods (Li et al. 2010; Perez-Pimienta et al. 2013). Although, the major challenge for commercial utilization of ionic liquids pretreatment is the relatively high cost (of ionic liquids) that ranges from 1 USD up to 800 USD per kg. The cost of ionic liquids may vary due to purity grade and the source (Konda et al. 2014; Perez-Pimienta et al. 2017b). In recent years, several researchers have reported the pretreatment of agave bagasse using ionic liquids (Perez-Pimienta et al. 2013, 2015, 2016a, b). Perez-Pimienta et al. (2013) performed ionic pretreatment of agave bagasse and switchgrass using 1-ethyl-3-methylimidazolium acetate ([C2mim][OAc]). The pretreatment was carried out at 15% biomass solid loading and reaction time of 3h. The temperature was maintained in a range of 120–160 °C. The pretreatment improved the digestibility of both substrates effectively. A delignification of 45.5% and 38.4% was observed for agave bagasse and switchgrass respectively at a temperature of 160 °C (Perez-Pimienta et al. 2013). Three pretreatments namely ammonia fiber expansion (AFEX), autohydrolysis, and ionic liquid were performed for agave bagasse and their efficiency was compared. Results demonstrated that all the carbohydrates available in biomass were preserved by AFEX pretreatment while 62.4% of xylan was degraded by autohydrolysis. The ionic liquid pretreatment caused 25% of lignin in wash streams (Perez-Pimienta et al. 2016a). Ternary ionic liquid-water system that had [C2mim][OAc], 1-butyl-3-methylimidazolium acetate ([C4C1Im][OAc]) and



water at different ratios was used for the pretreatment of agave bagasse and municipal solid waste (MSW) blend (1:1). Ionic pretreatment efficiently disrupted the agave bagasse and MSW and the efficiency was decreased when water was above 40%. A glucan conversion of 94.1 and 83.0% were obtained using an ionic liquid pretreatment with approximately 10 and 40% of water respectively. Results showed that the recycled ionic liquid-water system gave comparable results to that of the fresh ionic liquid-water system (Perez-Pimienta et al. 2017b). Different types of pretreatments that have been tested for agave biomass have been reviewed in Table 2.

## Biofuel production using *Agave* residues

The hydrolysis or saccharification process releases the reducing sugars that are metabolized by different microorganisms and converted into various valuable products such as biofuels, organic acids, xylitol, and microbial polysaccharides (Kumar et al. 2019a). The residues such as bagasse, leaves, stems, and cogollos from *Agave* plants have also been evaluated for different bioenergy and value-added products (Table 3).

### Bioethanol

Bioethanol production involves biomass collection, deconstruction of LCB into simple sugars, and conversion of sugars to biofuels and biochemicals (Li et al. 2019). The depolymerization of this complex polymer is required for its efficient utilization. The hydrolysis of celluloses is performed by cellulase enzyme complex having endoglucanase, exoglucanase, and  $\beta$ -glucosidase. The glucose generated by the cellulase hydrolysis is fermented into ethanol (Kumar et al. 2019a). Endoxylanases and  $\beta$ -xylosidases are involved in the depolymerization of xylan into monosaccharides. The hydrolysis of hemicelluloses mainly releases xylose, a pentose monomer sugar (Bhardwaj et al. 2019). *A. lechuguilla* cogollos were pretreated by auto-hydrolysis prior to enzymatic saccharification and the reducing sugars were fermented in to bioethanol. Auto-hydrolysis was performed in a Parr reactor at different severity factor with a solid/liquid ratio of 1/6 (w/v) at 200 rpm. Enzymatic hydrolysis of the solid fraction was performed to release the reducing sugars and it resulted in a maximum yield of 60.85%. A glucose concentration of 59 g/L was observed in the hydrolysate. The hydrolysate was fermented with *Saccharomyces cerevisiae* ATCC 4126 that produced 25.4 g/L (91% of the theoretical value) of ethanol (Ortiz-Mendez et al. 2017). Biomass residues from the *A. tequilana* and *A. salmiana* were pretreated with ammonia fiber expansion (AFEX) process and enzymatic hydrolysis was carried out at 17–20% of

solid loadings with a commercial enzyme loading of 20 mg protein/g glucan. The enzymatic hydrolysis was continued for 72 h at 50 °C and pH 4.8. The enzymatic hydrolysis resulted in approximately 85% of recovery of sugars. A glucose and xylose fermenting yeast, *S. cerevisiae* 242A (LNH-ST) was used for fermentation that resulted in 40 g/L of bioethanol (90% of metabolic yields) without any washing step or nutrients supplementation. The results proved that AFEX pretreatment can be a potential technology for the conversion of agave residues to biofuels and biochemicals (Gómez et al. 2018). Mielenz et al. (2015) studied biofuel production from the whole plant of twelve different species of *Agave*. Whole *Agave* plants were hydrolyzed with proper enzyme blend of biomass-degrading enzymes including inulinase and ethanol was produced from ten species of *Agave* using *S. cerevisiae*. *A. neomexicana* produced ethanol by  $119 \pm 11$  mg/g biomass. Saucedo-Luna et al. (2011) carried out the pretreatment of *A. tequilana* bagasse at 147 °C with 2%  $H_2SO_4$  for 15 min which resulted in the release of reducing sugars by 25.8 g/L. The pretreated residue was hydrolyzed by a commercial enzyme that gave the reducing sugars yield of 73.6% (41 g/L). The obtained reducing sugars were fermented with native yeast *Pichia caribbica* UM-5 that resulted in 8.99 g (56.57% of theoretical value) of ethanol from 50 g of bagasse (Saucedo-Luna et al. 2011). Table 4 shows fermentation conditions and bioethanol yield obtained from different agave related biomass.

### Biobutanol

Biobutanol is the biofuel of future and it is potential alternate of gasoline. 1-butanol has several advantages over ethanol such as energy density, engine compatibility, and safety (Kumar et al. 2016a). 2,3-butanediol is a chemical compound that has various important applications like biofuels and chemical precursors. The production cost of 2,3-butanediol is a critical issue and its biological production using low-cost renewable lignocellulosic materials is mainly focussed by the researchers. 2,3-butanediol production by *Klebsiella oxytoca* UM2-17 was carried out using acid pretreated *A. cupreata* bagasse. Enzymatic hydrolysis released 25.8 g/L of fermentable sugars in hydrolysate that was fermented by *K. oxytoca* UM2-17 to produce 2,3-butanediol (10.3 g/L). Besides that 0.28 g/L of ethanol was also produced as co-product (Pasaye-anaya et al. 2019). *Clostridium beijerinckii* BA101 was able to produce acetone and n-butanol and it was used for fermentation of reducing sugars generated by the hydrolysis of selected species of *Agave*. Biomass residues from *A. americana*, *A. salmiana*, and *A. tequilana* were fermented by *C. beijerinckii* BA101 that produced approximately similar yields of n-butanol and acetone per gram of biomass utilized (Mielenz et al. 2015).

**Table 2** Pretreatments and hydrolysis of agave biomass

| Agave biomass                  | Pretreatment                              | Conditions |   | Enzymatic hydrolysis conditions   | Yield of fermentable sugars                             | References                    |
|--------------------------------|---|------------|---|---|---|-------------------------------|
|                                |   | Type       |   |   |   |                               |
| <i>A. tequilana</i> bagasse    | Ionic liquid pretreatment                 |            | Milled biomass treated with 1-ethyl-3-methylimidazolium, temperature: 120–160 °C, treatment time: 3 h                                 | Cellulase, CTec2 (20 g protein/kg glucan), xylanase, Htec2 (2 g protein/kg xylan), temperature: 55 °C, pH: 4.8                        | Sugars yield: 6.66 kg m <sup>-3</sup>                   | Perez-Pimienta et al. (2015)  |
| <i>A. tequilana</i> leaves     | Dilute acid pretreatment                  |            | H <sub>2</sub> SO <sub>4</sub> : 1–4% (v/v) temperature: 115, 120, 130 °C, treatment time: 30, 60, and 90 min                         | Cellic <sup>®</sup> CTec2 (Novozymes): 1.4–6.9 FPU/g cellulose, temperature: 50 °C, pH: 5.0, agitation: 50 rpm, hydrolysis time: 96 h | 267.9 mg/g that corresponds to 69% of theoretical yield | Rijal et al. (2016)           |
| <i>A. lechuguilla</i> cogollos | Dilute acid pretreatment                  |            | H <sub>2</sub> SO <sub>4</sub> : 0.5–1.5 %, Solid loading: 10%, temperature: 160–200 °C,  | Cellic CTec3 (Novozymes): 15 FPU/g pretreated solid, temperature: 50 °C, pH: 4.8, agitation: 150 rpm, hydrolysis time: 72 h           | Hemicellulosic sugars: 87%, glucose: 68%                | Díaz-Blanco et al. (2018)     |
| <i>A. salmiana</i> leaves      | Acid and alkaline sequential pretreatment |            | (i) H <sub>2</sub> SO <sub>4</sub> : 1 % (v/v) for 90 min at 121 °C<br>(ii) NaOH 3.4% for 70 min at 121 °C                            | Cellclast 1.5L (Novozyme): 2.93–17.07 FPU/g cellulose temperature: 50 °C, pH: 4.8, agitation: 150 rpm, hydrolysis time: 72 h          | Sugars: 50 g/L or 94.49%                                | Láinez et al. (2019)          |
| <i>A. tequilana</i> bagasse    | Hydrothermal high-pressure pretreatment   |            | CO <sub>2</sub> -H <sub>2</sub> O mixture at high pressure-temperature: 150–190 °C, treatment time: 10–50 min                         | Cellulase: 15 FPU/g glucan temperature: 50 °C, pH: 4.8, agitation: 150 rpm, hydrolysis time: 96 h                                     | 75.8 % of carbohydrates                                 | Aguirre-Fierro et al. (2020)  |
| <i>A. tequilana</i> bagasse    | Hydrothermal pretreatment                 |            | Solid:liquid ratio (1:10) temperature: 160–180 °C, treatment time: 30 min   | Cellic <sup>®</sup> CTec2 (Novozymes): 20 FPU/g glucan, temperature: 50 °C, pH: 4.8, agitation: 150 rpm, hydrolysis time: 96 h        | 102.62 g/L or 99.53%                                    | Aguilar et al. (2018)         |
| <i>A. tequilana</i> bagasse    | Thermo-mechano-chemical process           |            | Twin-screw extruder was used 5% NaOH (w/v) supplied A liquid to solid ratio of 7.5 maintained   | Different commercial enzymes: 19 FPU/g dry matter, temperature: 50 °C, pH: 5.5, hydrolysis time: 72 h                                 | 69.5 g/L or 73%   | Montiel et al. (2016)         |
| <i>A. tequilana</i> bagasse    | Hydrothermal pretreatment                 |            | Milled bagasse combined with solid/liquid ratio of 1:10 (w/v), temperature: 160–220 °C, treatment time: 10–50 min, agitation: 200 rpm | Cellic Ctec2 (Novozyme): 15 FPU/g glucan, temperature: 50 °C, pH: 4.8, agitation: 150 rpm, hydrolysis time: 72 h                      | Glucose: 195.6 g/L or 98%                               | (Pino et al. 2019)            |
| <i>A. tequilana</i> bagasse    | Ionic liquid and alkaline pretreatment    |            | 1-butyl-3-methylimidazolium chloride, temperature: 120–160 °C, treatment time: 3 h  | Cellulclast 1.5 L and Novozyme 188: 30 FPU/g of glucan and 60 CBU/g glucan, temperature: 55 °C, pH: 4.8, hydrolysis time: 72 h        | 7.8 g dm <sup>-3</sup>                                  | Perez-Pimienta et al. (2016b) |

Table 2 (continued)

| Agave biomass  | Pretreatment                            |   | Enzymatic hydrolysis conditions   | Yield of fermentable sugars   | References                      |
|--|---|---|---|---|---------------------------------|
|  | Type                                    | Conditions  |   |   |                                 |
| <i>A. atrovirens</i> biomass                           | Enzymatic pretreatment                  | <i>Trametes versicolor</i> laccase: Agave biomass of 1 mm particle size treated with 3 U/g, temperature: 45 °C, treatment time: 12 h, pH: 4., agitation: 160 rpm    | Cellulclast 1.5L: 15 FPU/g substrate, temperature: 50 °C, pH: 4.8, agitation: 150 rpm, hydrolysis time: 72 h                            | 2.81 ± 0.13 g/L or 63.26 ± 2.98   | Lo (2016)                       |
| <i>A. americana</i> stalk                              | Acidic, sulphite, alkaline pretreatment | H <sub>2</sub> SO <sub>4</sub> (2%), Na <sub>2</sub> SO <sub>3</sub> (4%), NaOH (8%), all pretreatments conducted at temperature: 180 °C and treatment time: 30 min | Cellulase: 15 FPU/g cellulose, β-glucosidase: 30 CBU/g cellulose, temperature: 50 °C, pH: 4.8, agitation: 160 rpm, hydrolysis time: 72h | Cellulose conversion: Acidic (43.5%), sulphite (53.8%), and alkaline (68.9)                           | Yang and Pan (2012)             |
| <i>A. atrovirens</i> leaves, <i>A. salmiana</i> leaves | Acidic and alkaline pretreatment        | (i) HCl: 1% at 121 °C for 15, 30, and 45 min<br>(ii) NaOH: 2% at 121 °C for 15, 30, and 45 min  | Enzyme: 6000 U/L, temperature: 46–50 °C, pH: 4.6, agitation: 50–150 rpm, hydrolysis time: 96 h  | Cellulose conversion by 61.81 and 42.39% for <i>A. atrovirens</i> and <i>A. salmiana</i> respectively | Medina-Morales et al. (2017)    |
| <i>A. tequilana</i> bagasse                            | Alkaline-oxidative                      | (i) NaOH: 6% at 120 °C for 1 h<br>(ii) H <sub>2</sub> O <sub>2</sub> : 6% at 30 °C for 24 h without agitation   | Different enzymes with variable dosages, temperature: 50 °C, agitation: 150 rpm, hydrolysis time: 72 h                                  | 352.18 g/kg of dry biomass  | Velázquez-Valadez et al. (2016) |

**Table 3** Value-added product from different types of agave biomass

| Agave species            | Type of residue used | Value-added product                 | References   |
|--------------------------|----------------------|-------------------------------------|--|
| <i>A. tequilana</i>      | Bagasse              | Ethanol                             | Barrera et al. (2016), Gómez et al. (2018), Hernández et al. (2019)  |
|                          | Leaves and stems     | Xylanase, inulinase, and pectinase  | Huitron et al. (2008)  |
|                          | Leaves               | Ethanol                             | Gómez et al. (2018)  |
|                          | Bagasse              | Hydrogen                            | Arreola-vargas et al. (2015a), Contreras-dávila et al. (2017), Valdez-guzmán et al. (2019), Mu et al. (2020) |
|                          | Bagasse              | Methane                             | Arreola-vargas et al. (2015b), Arreola-vargas et al. (2015a), Corona and Razo-flores (2018)                  |
|                          | Bagasse              | Succinic acid                       | Corona-gonzález et al. (2016)  |
|                          | Bagasse              | Polyhydroxybutyrate                 | Gonz et al. (2019)   |
| <i>A. durangensis</i>    | Leaves               | Cellulase, inulinase                | Contreras-hernández et al. (2018)  |
| <i>A. lechuguilla</i>    | Cogollos             | Ethanol                             | Morales-Martínez et al. (2017)   |
|                          | Cogollos             | Hydrogen                            | Garza and Moreno-dávila (2018)   |
| <i>A. salmiana</i>       | Leaves               | Endoglucanase, $\beta$ -glucosidase | Silva-mendoza et al. (2020)  |
|                          | Leaves               | Ethanol                             | Gómez et al. (2018)  |
|                          | Bagasse              | Ethanol                             | Gómez et al. (2018)  |
| <i>Agave neomexicana</i> | Bagasse              | Ethanol                             | Mielenz et al. (2015)  |
| <i>Agave cupreata</i>    | Bagasse              | 2,3-butanediol                      | Pasaye-anaya et al. (2019)   |

## Biogas and biomethane

Biogas is one of the most promising renewable biofuel that chiefly consists of methane and carbon dioxide. It is produced by anaerobic digestion that generates two usable products namely biogas and digestate. It is either used to generate heat and power or upgraded to biomethane. Upgraded biomethane is injected into the gas grid or utilized as a fuel for vehicles. The digestate can be used as a biofertilizer for soil (Seppala et al. 2013). Tequila industry in Mexico generates agave bagasse, a byproduct that has not been handled properly up to now, even though it could be a low-cost resource for energy generation (Weber et al. 2020). Weber et al. (2020) performed steam explosion pretreatment of agave bagasse before anaerobic digestion. The methane yield of  $235 \text{ mL}_N \text{ g}_{VS}^{-1}$  was achieved with pretreated bagasse that was 11% higher compared to anaerobic digestion (AD) of untreated bagasse. Corona and Razo-flores (2018) tested the sequential process to maximize the recovery efficiency from *A. tequilana* bagasse. During the sequential process, in the first stage of process a continuous stirred tank reactor (CSTR) was employed for hydrogen production. The effluent generated from CSTR was used as substrate for methane manufacturing in the second stage. The energy conversion efficiency (56%) was improved significantly during the two stages of the continuous process compared to one-stage hydrogen production (8.2%) (Corona and Razo-flores 2018). Arreola-vargas et al. (2015b) utilized acid hydrolysate of cooked and uncooked *A. tequilana* for methane production. Results indicated that cooked bagasse was hydrolyzed more effectively for sugar release. The total reducing sugars of

27.9 g/L and 18.7 g/L were obtained from the hydrolysate of cooked and uncooked bagasse respectively. The cooked bagasse hydrolysate contained 5-hydroxymethylfurfural, therefore methane production was carried using uncooked bagasse hydrolysate. Constant methane production of  $0.26 \text{ L CH}_4/\text{g COD}$  was obtained by anaerobic sequencing batch reactors without nutrient addition (Arreola-vargas et al. 2015b). Another study reported acidic and enzymatic hydrolysis of *A. tequilana* bagasse and the hydrolysate was fermented to produce methane and hydrogen through single and two-stage AD. Results showed that two-stage anaerobic digestion resulted in higher energy recovery. Hydrogen production was  $3.4 \text{ mol H}_2/\text{mol hexose}$  during the acidogenesis phase of the two-stage process using enzymatic hydrolysate at a concentration of 40%. While the highest methane yield ( $0.24 \text{ L CH}_4/\text{g COD}$ ) was achieved during the methanogenic phase of two-stage anaerobic digestion using 20% of both hydrolysate. The total recovery of energy in two-stage process was observed 3.3 times compared to single-stage process (Arreola-vargas et al. 2015a). Breton-Deval et al. (2018) studied biomethane production from *A. tequilana* bagasse using batch and sequencing batch reactors (AnSBR). *A. tequilana* bagasse was pretreated with HCl and  $\text{H}_2\text{SO}_4$  and the optimization of pretreatment parameters including temperature, acid concentration, and reaction time was performed through central composite design. The findings demonstrated that maximum sugar recovery was obtained with HCl ( $0.39 \text{ g total sugars/g bagasse}$ ) compared to  $\text{H}_2\text{SO}_4$  ( $0.26 \text{ g total sugars/g bagasse}$ ) under optimal conditions. Moreover,  $\text{H}_2\text{SO}_4$  generated a higher concentrations of inhibitors such as furans and  $\text{CH}_3\text{COOH}$ . AD showed that



**Table 4** Agave biomass for bioethanol production

| Agave biomass                  | Ethanogenic microorganism               | Fermentation conditions   | Ethanol yield   | References                    |
|--------------------------------|---|---|---|-------------------------------|
| <i>A. tequilana</i> bagasse    | <i>S. cerevisiae</i>                    | Temperature: 32 °C, fermentation time: 12 h, pH: 5.5, agitation: 100 rpm  | 65.2 g/L with conversion efficiency of 98.4% of theoretical value | Rios-González et al. (2017)   |
| <i>A. tequilana</i> bagasse    | <i>E. coli</i> strain MS04              | Temperature: 37 °C, fermentation time: 48 h, pH: 7.0, agitation: 60 rpm   | 12.1–12.7 kg of ethanol per kg of bagasse                         | Pérez-Pimienta et al. (2017c) |
| <i>A. tequilana</i> biomass    | <i>S. cerevisiae</i> NCIM 3622          | Fermentation time: 5 days, pH: 4.8  | 45.55%  | Shashikala and Kumar (2019)   |
| <i>A. tequilana</i> bagasse    | <i>S. cerevisiae</i>                    | Temperature: 35 °C, fermentation time: 48 h, agitation: 150 rpm           | 36.4 ± 0.95 g/L or 72% of maximal theoretical yield               | Aguirre-Fierro et al. (2020)  |
| <i>A. tequilana</i> bagasse    | <i>S. cerevisiae</i>                    | Temperature: 28 °C, fermentation time: 72 h, agitation: 150 rpm           | 107.9 gallons per metric ton                                      | Barrera et al. (2016)         |
| Agave bagasse                  | <i>S. cerevisiae</i> strain Super-Start | Temperature: 37 °C, fermentation time: 72 h                               | 0.25 g/g or 86% of the maximum theoretical yield                  | Caspeta et al. (2014)         |
| <i>A. sisilana</i> biomass     | <i>S. cerevisiae</i>                    | Temperature: 40 °C, fermentation time: 6 days, agitation: 120 rpm         | 0.30 g/g dry biomass  | Punnapayak et al. (1999)      |
| <i>A. tequilana</i> bagasse    | <i>S. cerevisiae</i>                    | Temperature: 35 °C, fermentation time: 72 h agitation: 200 rpm            | 32 g/L that corresponds to 85% of glucose available               | Montiel et al. (2016)         |
| <i>A. tequilana</i> bagasse    | <i>S. cerevisiae</i> PE-2               | Temperature: 30 °C, fermentation time: 12 h agitation: 150 rpm            | 55.02 g/L that corresponds to 87.56% of ethanol yield             | Aguilar et al. (2018)         |
| <i>A. tequilana</i> bagasse    | <i>S. cerevisiae</i>                    | Temperature: 35 °C, fermentation time: 72 h agitation: 150 rpm            | 36.4 ± 0.95 g/L or 72% of maximum theoretical yield               | Aguirre-Fierro et al. (2020)  |
| <i>A. salmiana</i> leaves      | <i>K. marxianus</i>                     | Temperature: 30 °C, fermentation time: 24 h agitation: 150 rpm            | 92. 88% of maximum theoretical yield                              | Láinez et al. (2019)          |
| <i>A. lechuguilla</i> cogollos | <i>E. coli</i> strain MM160             | Temperature: 37 °C, fermentation time: 288 h, pH: 7.0, agitation: 400 rpm | 73.30% of maximum theoretical yield                               | Díaz-Blanco et al. (2018)     |
| <i>A. tequilana</i> leaves     | <i>S. cerevisiae</i>                    | Temperature: 30 °C, fermentation time: 24 h, agitation: 50 rpm            | 38.6 g/L or 68% of maximum theoretical yield                      | Rijal et al. (2016)           |

the hydrolysate obtained from HCl pretreatment was a better substrate for methanogenesis. The results indicated that mild hydrolysis conditions were suitable for methanogenesis as compared to sugars recovery. The hydrolysate obtained from mild conditions was utilized to feed the AnSBR for 110 days. The maximum CH<sub>4</sub> yield and production of 0.28 NL CH<sub>4</sub>/g-COD and 1.04 NL CH<sub>4</sub>/day were obtained at cycle time of 3 days (Breton-Deval et al. 2018).

*A. tequilana* bagasse was tested for biomethane production using different pretreatment methods. The pretreatment methods including biological, enzymatic, chemical, and hydrothermal were evaluated based on carbohydrates solubilization (CHO), chemical oxygen demand (COD), and biological methane potential (BMP) from hydrolysates. All of

these pretreatments except chemical treatment act similarly to give an average yield of 0.16 ± 0.02 g<sub>COD</sub>/g. The chemical pretreatment resulted in a 2.6 times higher methane yield with a 0.95 of CHO/COD ratio. Chemical pretreatment generated a higher concentration of inhibitors such as furfural hydroxymethyl-furfural, and phenols compared to biological and enzymatic pretreatments. Most hydrolysates resulted in approximately the same BMP, on average 219 ± 15 mL/g<sub>CODin</sub>. The hydrolysates from different pretreatments were differentiated based on lag phase and methane production rates (Valdez-Vazquez et al. 2020). Recently, ionic acids derived from biological materials have been paid attention as a potential approach for the pretreatment of LCB. Pérez-Pimienta et al. (2020) studied methane production

from bioderived ionic acid pretreated *A. tequilana* bagasse. The pretreatment conditions were optimized using ionic liquid cholinium lysinate ([Ch][Lys]) as pretreatment agent. The highest total sugars yield was obtained at pretreatment temperature of 124 °C, reaction time of 205 min, and solid loading of 20%. The ionic pretreatment attained a higher delignification (45.4%) and changes the chemical linkages in biomass. Although, the cellulose crystallinity was found to increase up to 0.62 (pretreated) compared to untreated (0.51). The mass balance analysis showed that 38.2 kg glucose and 13.1 kg xylose were converted into 12.5 kg of methane per 100 kg of *A. tequilana* bagasse that corresponds to 86% of the theoretical yield of methane.

## Biohydrogen

Biohydrogen production through microbial fermentation is considered as an environmentally sustainable approach. It is an ideal biofuel that has the highest energy content. However, more research is required for its application at industrial scale (Contreras-dávila et al. 2017). Enzymatic hydrolysate of *A. tequilana* bagasse was used as substrate for continuous fermentation to produce hydrogen. The substrate was fed with organic loading rates (OLRs) in continuous tank reactor (CSTR) and trickling bed reactor (TBR) up to 87 days. In CSTR, 2.53 L H<sub>2</sub>/L-day and 1.35 mol H<sub>2</sub>/mol of volumetric hydrogen production rate (VHPR) and hydrogen molar yield (HMY) were obtained with OLR of 52.2 and 40.2 g COD/L-day respectively, that showed an inverse correlation. In TBR, both VHPR and HMY were improved up to 3.45 L H<sub>2</sub>/L-day and 1.53 H<sub>2</sub>/mol respectively with increasing OLR (52.9 g COD/L-day) (Contreras-dávila et al. 2017). The bagasse of *A. tequilana* Weber (var. azul) was evaluated for biohydrogen production by suspended and granular cell anaerobic bioreactors. The hydrolysate from acidic pretreatment was used for hydrogen production in batch mode and the carbohydrate concentrations higher than 6.3 g<sub>CHO</sub>/L was not found feasible. To attain maximum hydrogen productivity, granular biomass was observed to be more suitable for higher OLR compared to suspended biomass (Mu et al. 2020). Valdez-guzmán et al. (2019) used detoxified and undetoxified hydrolysate from *A. tequilana* bagasse for hydrogen production. Activated carbon was employed for the detoxification process at different concentrations and pH values. The results demonstrated maximum removal of acetic acid (89 %) using lower concentration of activated carbon of 1% (p/v). Moreover, the losses of fermentable sugars were found to be minimum. The effect of pH variation during detoxification process was not significant. The detoxified hydrolysate resulted in improved biohydrogen production (by 33%) compared to the undetoxified one and the hydrogen molar yields were 1.71 and 1.23 mol H<sub>2</sub>/mol<sub>sugar</sub> respectively (Valdez-guzmán et al. 2019).

*A. lechuguilla* cogollos were also utilized for hydrogen production under anaerobic digestion. Cogollos were pretreated by autohydrolysis in a high-pressure reactor using a solid/liquid ratio of 1:6 (w/v) at 190 °C for 30 min. Enzymatic hydrolysis of cogollos biomass was carried out to produce hydrolysate (55 g/L) that was fermented with a mixed microbial consortium to produce hydrogen. After optimization, a maximum yield of hydrogen was 3.48 mol H<sub>2</sub>/mol glucose consumed at 120 h (Garza and Moreno-dávila 2018). Agave bagasse from tequila distillery was hydrolyzed using two enzymatic preparations and individual enzyme preparation that resulted in binary agave bagasse hydrolysate (BH) and individual agave bagasse hydrolysate (IH). Both hydrolysates were employed for continuous hydrogen production using CSTR and TBR. The hydrolysates BH and IH were fed in CSTR that resulted in the VHPR of 13 and 2.25 L H<sub>2</sub>/L-day respectively. In TBR, BH and IH resulted in VHPR of 5.76 and 2.0 L H<sub>2</sub>/L-day respectively. This difference between reactors could be due to substrate availability that is intrinsic factor for microbial growth while the variation in VHPR between hydrolysates might be due the composition of enzymatic hydrolysates (de Jesús Montoya-Rosales et al. 2019). Different commercial enzyme preparations (Celluclast 1.5 L, Stonezyme, Zymapect, and Cellulase 50XL) were evaluated for the selection of suitable enzyme preparation for hydrogen and methane production from *A. tequilana* bagasse. Batch fermentation showed the highest hydrogen productivity (1.88 H<sub>2</sub>/L) using Celluclast 1.5 L that was 1.6–2% higher than other enzymes. Whereas, Zymapect demonstrated maximum methane productivity (1.32 L CH<sub>4</sub>/L) with a specific yield of 163 L CH<sub>4</sub>/kg bagasse that was 1.7–2.0-fold higher than other alternatives (Tapia-Rodríguez et al. 2019). Galindo-Hernández et al. (2018) performed oxidative delignification of *A. tequilana* bagasse for its improved saccharification and the effect of enzyme synergism was also tested for hydrogen and methane production. *A. tequilana* bagasse was pretreated with alkaline H<sub>2</sub>O<sub>2</sub> for lignin removal and the results demonstrated 97% delignification and 88% recovery of carbohydrates after 1.5 h of treatment. The enzymatic saccharification was carried out with cellulases and hemicellulases for extraction of fermentable sugars. The mixture of cellulases and hemicellulases resulted in 2-fold sugars yield and productivity as compared to single enzyme hydrolysis (cellulase) that indicated the synergism of enzymes during hydrolysis. The obtained hydrolysate was used as a feedstock for fermentation to produce hydrogen and methane. The results showed 1.5 and 3.6 times (215.14 ± 13 L H<sub>2</sub> and 393 ± 13 L CH<sub>4</sub> per kg bagasse respectively) of hydrogen and methane production compared to those obtained with hydrolysates from non-pretreated bagasse and hydrolysed with single enzyme (Galindo-Hernández et al. 2018).

Hydrogen production is a renewable and eco-friendly energy alternative. However, LCB conversion into hydrogen requires expensive enzymatic hydrolysis that can be bypassed through Consolidated Bioprocessing (CBP). This strategy was performed through the integration of cellulose utilizing microorganisms from the bovine ruminal fluid (BRF) and hydrogen-producing *Clostridium acetobutylicum* during fermentation. *A. lechuguilla* cogollos were acid pretreated and used for hydrogen production through CBP. CBP resulted in hydrogen production of 150 L H<sub>2</sub> per kg of biomass at 10% solid loadings, pH 5.5 and temperature of 35 °C at 264 h (Morales-Martínez et al. 2020).

## Use of Agave biomass for enzyme production

Enzymatic processing is a highly growing field of biotechnology. The production cost of enzymes is the major issue for their industrial utilization. The utilization of lignocellulosic-based waste materials as substrate can induce the production of highly active multifunctional enzymes cost-effectively. Agave biomass has been employed as a cost-effective substrate for cellulase, xylanase, and inulinase production (Ibrahim 2008; Kumar et al. 2019a). Cellulases and xylanases have a wide range of applications in pulp and paper industry, textile industry, food processing, brewery and wine making, animal feed processing, detergent preparation, and bioenergy industry (Kumar et al. 2016b; Gautam et al. 2018). Silva-mendoza et al. (2020) tested *A. salmiana* leaves for cellulase production using *Penicillium* sp. under solid-state fermentation (SSF). *A. salmiana* leaves were found to be a suitable substrate for cellulase production and induced endoglucanase (14.4 U/g substrate), exoglucanase (3.5 U/g substrate), and  $\beta$ -glucosidase (4.14 U/g). The leaves of *A. durangensis* were also used for the production of cellulase, inulinase, and  $\beta$ -fructofuranosidase by the strains of *Aspergillus niger*, *Aspergillus flavus*, *Neurospora crassa*, and *Alternaria* sp. under submerged fermentation. *A. durangensis* leaves were pretreated with ultrasound and high temperature and utilized for enzyme production. The ultrasound pretreatment resulted in higher cellulase and inulinase production while the thermal pretreatment improved the production of  $\beta$ -fructofuranosidase (Contreras-hernández et al. 2018). Huitron et al. (2008) employed *A. tequilana* waste as a substrate for cellulase, xylanase, pectinase, and inulinase production. The highest inulinase (1.48 U/mL) was produced by *Aspergillus* sp. CH-A-2010 while maximum xylanase (1.52 U/mL) and pectinase (2.7 U/mL) was produced by *A. niger* CH-A-2016. The enzyme activities were significantly higher with *A. tequilana* waste compared to lemon peel and polymeric carbohydrates (Huitron et al. 2008). *A. atrovirens* fiber was partially characterized, thermal

pretreated, and employed as substrate for cellulase production by *Trichoderma asperellum* under SSF. *T. asperellum* produced 12,860.8, 3144.4 and 384.4 U/g of endoglucanase, exoglucanase, and  $\beta$ -glucosidase respectively. *A. atrovirens* fiber was found to be a suitable carbon source for cellulase production (Nava-Cruz et al. 2016). *A. sisalana* biomass was used as raw material for simultaneous production of edible mushroom and extracellular enzymes by *Pleurotus* HK 37 under SSF. *Pleurotus* HK 37 resulted in maximum laccase production (27.3 U/mL) when the substrate was fully colonized. Maximum manganese peroxidase, lignin peroxidase, and xylanase activities were 8.9, 0.34 and 0.28 U/mL respectively on complete colonization (Muthangya et al. 2013).

## Conversion of Agave biomass in to other valuable products

Polyhydroxybutyrate (PHB) is a biodegradable and biocompatible polymer produced by microorganisms. It has the potential to replace petroleum-based plastics as packing material, disposable materials, and biomedical devices (Gonz et al. 2019; Tsang et al. 2019). Gonz et al. (2019) utilized tequila agave bagasse hydrolysate for the production of PHB by xylose-assimilating bacteria, *Burkholderia sacchari*. agave bagasse was acid hydrolysed that yielded 20.6 g/L of reducing sugars with xylose and glucose at 7:3 ratio. When *B. sacchari* was grown on undetoxified agave bagasse hydrolysate, phenolic compounds resulted in more than 60% inhibition of growth. Therefore, detoxification of hydrolysate was performed with resins, activated charcoal, and laccases. The activated charcoal (50 g/L) achieved maximum removal of growth inhibitory compounds (92%) from hydrolysate at pH 2 for 4 h. Finally, the detoxified agave bagasse hydrolysate was used as a substrate for PHB production in a two-step cultivation process, reaching a biomass production of 11.3 g/L. The PHB production was 2.67 g/L after 120 h of the incubation period (Gonz et al. 2019).

Succinic acid is an important specialty chemical that has several industrial applications. Succinic acid production by microbial fermentation is an environmentally friendly approach. Succinic acid is also included in the list of twelve platform chemicals from biomass by the US department of energy (Kumar et al. 2019b). It is a C4 dicarboxylic acid that finds several applications in the food, agricultural, and pharmaceutical industries (Kumar et al. 2016a). Bagasse hydrolysate from *A. tequilana* was used as a substrate for the production of succinic acid by *Actinobacillus succinogenes* in batch and repeated batch fermentation. Acidic and enzymatic hydrolysis of agave bagasse was optimized by response surface methodology (Corona-gonzález et al. 2016). H<sub>2</sub>SO<sub>4</sub>, HCl, and enzymatic hydrolysis resulted in 21.7, 22.4, and 19.8 g/L of sugars respectively, under

optimal conditions. Maximum production of succinic acid (0.446 g/g) was resulted by enzymatic hydrolysate with the productivity of 0.57 g/L h in a batch reactor. *A. succinogenes* was immobilized with agar in repeated batch fermentation and it produced a 33.6 g/L of succinic acid using 87.2 g/L monosaccharides after 5 cycles in 40 h, giving the productivity of 1.32 g/L h (Corona-gonzález et al. 2016).

## Future prospects

Rapidly growing population and industrialization have increased energy demand of the world that is expected to approach up to 739 quadrillions BTU in 2040. This high energy demand compels to find the renewable alternative sources. LCB is abundantly available and relatively inexpensive source for biofuel production (Sharma et al. 2020). Different types of residues such as bagasse, leaves, and cogollos are generated from different species of *Agave* that are found in different countries. The plants of genus *Agave* are capable to grow in semiarid regions with low water availability and high temperature. They are resistant to drought conditions and show higher sugar/products conversion yield. Therefore, *Agave* plants are potential feedstock for a modern bio-refinery concept (Perez-Pimienta et al. 2017c). The review of recent researches shows that agave biomass has been tested for the production of bioethanol (Aguirre-Fierro et al. 2020), biomethane (Valdez-Vazquez et al. 2020), biobutanol (Mielenz et al. 2015), biohydrogen (Morales-Martínez et al. 2020), succinic acid (Corona-gonzález et al. 2016), and polyhydroxybutyrate (Gonz et al. 2019). Several researchers studied the pretreatment and hydrolysis of agave biomass to obtain maximum yield of reducing sugars (Rios-Gonzalez et al. 2021). These reducing sugars can be the substrate for the microbial production of organic acids such as lactic acid, citric acid, fumaric acid, malic acid, and itaconic acid. The reducing sugars released from agave biomass can also be fermented into valuable microbial polysaccharides and xylitol. A very few studies reported the production of cellulases, xylanases, lignin degrading enzymes, and inulinase production using agave biomass as substrate. Agave biomass might be a potential low-cost substrate for the production of these industrial enzymes in future.

## Conclusion

The agave biomass is an ideal substrate for the second generation biofuels due to its ability to grow on marginal lands with minimum fertilizer and irrigation. It does not compete with human food supply. Different species of *Agave* including *A. tequilana*, *A. durangensis*, *A. lechuguilla*, *A. salmiana*, *A. Americana*, *A. angustifolia*, *A. fourcroydes*, and *A.*

*sisalana* are grown for different products such as alcoholic beverages (tequila, mescal, and pulque), fiber, ornamental beauty, animal feed, and chemicals such as saponins. These plants generate a substantial amount of biomass residues in the form of leaves, bagasse, stems, or cogollos. The disposal of these residues creates serious environmental problems. These residues can be a low-cost substrate for the production of valuable products such as biofuels, industrial enzymes, and organic acids. The conversion of agave biomass and residues into value-added products will minimize their disposal into environment. Moreover, the generation of biofuels, and organic acids from LCB will decrease dependency on fossil fuels, and conventional chemical methods of organic acids production. Effective pretreatment is essential for enzymatic hydrolysis and bioconversion of LCB into biofuels and biochemicals. Recently, new technologies of pretreatment such as high-pressure CO<sub>2</sub>-H<sub>2</sub>O and ionic liquids have been successfully utilized for agave residues. The utilization of agave biomass and residues for the production of value-added products will assist to achieve the goal of environmentally sustainable development.

## Declarations

**Conflict of interest** Authors declare that there are no conflicts of interest regarding the publication of this paper.

**Ethical statement and informed consent** Hereby, I, Dr. Amit Kumar consciously assures that for the manuscript “Agave biomass: A potential resource for production of value-added products” the following is fulfilled: (1) This work is the authors’ own original work, which has not been previously published elsewhere. (2) The paper is not currently being considered for publication elsewhere. (3) All sources used are properly disclosed (correct citation). (4) All authors have been personally and actively involved in substantial work leading to the paper, and will take public responsibility for its content. (5) All the authors mentioned in the manuscript have agreed for authorship, read and approved the manuscript, and given consent for submission and subsequent publication of the manuscript.

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