Research Article

Saccharification and fermentation of pretreated banana leaf waste for ethanol production

Meenakshi Suhag[1] · Anil Kumar[2] · Joginder Singh[3]

Abstract

The present work reveals the potential applicability of banana leaf waste as feedstock for bioethanol production as very less work has been reported on this feedstock. For achieving the same, the performance of crude cellulases of *Aspergillus niger* JD-11 is explored for saccharification of untreated and pretreated (alkaline, dilute acid and steam) banana leaf waste. Therefore, to determine the best saccharifying conditions, the effects of various parameters such as enzyme loading (5–15 FPU/g), temperature (40–50 °C), surfactant addition (0.05–0.15% Tween 80 and PEG 6000) and substrate concentrations (2–6%) were studied at 150 rpm and pH 5.0. The crude cellulases yielded maximum reducing sugars (524.83 mg/g) from the dilute acid pretreated banana leaf waste at 45 °C, 15 FPU/g enzyme loading and 2% substrate loading in the presence of 0.15% PEG 6000 (w/v) as surfactant. The fermentation of enzymatic hydrolysate was carried out using initial reducing sugars concentration of 40 g/L in the medium at pH 5.5, 150 rpm and 30 °C for 30 h with the help of *Saccharomyces cerevisiae*. Ethanol production was estimated at every 6 h and maximum ethanol yield of 0.38 g/g sugar and productivity of 1.28 g/L/h was obtained after 12 h of fermentation of banana leaf waste hydrolysate. Thus, the easily obtained banana leaf waste could be a promising feedstock for bioethanol production.

Abbreviations

LB	Lignocellulosic biomass
w/v	Weight/volume
SSF	Solid state fermentation
MS	Mandels and Sternberg's media
rpm	Rotations per minute
mL	Millilitre
FPU	Filter paper activity units
DNSA	Dinitrosalicylic acid
FPase	Filter paperase
RS	Reducing sugars
PEG	Polyethylene glycol 6000
YPD	Yeast peptone dextrose
v/v	Volume/volume

1 Introduction

To achieve sustainable development as well as to combat the challenges of first generation biofuels, there is huge concern concurrently about the utilization of lignocellulosic materials/wastes which are abundant, easily available and relatively cheaper. Moreover, the bioconversion of these second-generation wastes, generated from many agricultural and forestry activities, to energy can not only promote self-sufficiency but also breed a new industry of jobs and hence economic growth leading to the upliftment of entire society [5, 52, 59]. Worldwide bioethanol articulated itself as most demanding engine fuel because it acts as octane enhancer for cleaner combustion in

✉ Joginder Singh, jssdahiya@gmail.com; Meenakshi Suhag, meenakshisuhag@gmail.com; Anil Kumar, anildhaka2k9@gmail.com | [1]Institute of Environmental Studies, Kurukshetra University, Kurukshetra, Haryana 136119, India. [2]Department of Botany, Pt. NRS Government College, Rohtak, Haryana 124001, India. [3]Laboratory of Environmental Biotechnology, Department of Botany, AIJHM College, Rohtak, Haryana 124001, India.

unleaded gasoline thereby reducing air pollution [12]. The foremost raw materials utilized for the production of bioethanol are corn in the US and sugarcane in Brazil [6]. Also, there are several studies reported in the literature regarding exploitation of second-generation biofuels, *i.e.*, bioethanol production from different lignocellulosic feedstocks like sugarcane bagasse [36], wheat straw [13], sorghum straw [25], switchgrass [48] and grasses [29] etc. Likewise, banana also represents one of the important fruit crop of tropical and sub tropical regions [1] and generates huge amount (~ 220t/hectare) of waste residues (i.e., stem, leaves and rhizomes) which are primarily lignocellulosic in nature [37] and hence may act as good raw material for the production of bioethanol [53]. Furthermore, banana peels are generally disposed off in the open, as also observed over nearby water-bodies or on roads, which further become one of the greatest causes of environmental pollution [33]. In India, the waste residues of banana fruit constitute a significant factor of urban solid wastes which are difficult to manage under normal conditions and sometimes burnt unethically [7]. Oberoi et al. [31] estimated that more than 1.6 million tonnes of dry banana peels are being produced by India every year which may directly serve as the source of agricultural lignocellulosic waste as its leaf fibers are composed of β-(1-4)-D glucose units in their crystalline microfibrils of the cellulose chains [60]. Whereas, non-cellulosic matter has 20–25% hemicellulose, 25–30% protopectin, 10–12% lignin, 1–2% insoluble pectin and the remaining being cellulose [40]. Usually after harvesting the fruits, the leaves are left behind in the field which take many months for their natural degradation [4, 9]. Therefore, using banana waste as lignocellulosic biomass appeared suitable since they are relatively low in lignin [15] but rich in carbohydrates which are necessarily required for bioethanol production.

Lignocellulosic biomass (LB) conversion into bioethanol mainly consists of four basic steps, viz., pretreatment, hydrolysis/saccharification, fermentation and product distillation. The recalcitrant nature of any LB is one of the physical barriers which can be overcome to some extent by using different pretreatment methods to augment the reachable surface area of cellulose which finally boost up the yield of saccharification [41, 56]. The most commonly used methods for the treatment of different lignocellulosic biomasses are steam, dilute acid and alkali pretreatments [44]. After pretreatment, hydrolysis process is required to hydrolyze the LB into monomeric sugar units which are ultimately fermented into bioethanol. Acid hydrolysis and enzymatic hydrolysis are the two basic methods for saccharification of LB. However, enzymatic hydrolysis of LB is a key approach [46] as it is performed at mild conditions of temperature and pH, having no corrosion problem and usually produces improved glucose yields as compared to acid catalyzed hydrolysis [6, 35, 47]. Enzymatic hydrolysis is commonly accomplished by a battery of hydrolytic and oxidative bacterial and fungal enzymes which are capable to synergistically break down the cellulose, hemicellulose and lignin [34]. The degraded cellulosic monomers are fermented to obtain bioethanol which can be utilized for different purposes after distillation.

In this study, enzymatic hydrolysis of pretreated banana waste with acetone fractionated partially purified cellulases of *Aspergillus niger* JD-11 was investigated keeping the practical fact of [47] in mind that the main challenges of low yield and high cost of hydrolysis could be overcome by optimizing various process parameters [18, 30]. Accordingly, the present work was envisaged to analyze the efficient production of fermentable sugars through enzymatic hydrolysis of pretreated banana waste in respect of enzyme loading, temperature, concentration of surfactant as well as substrate.

2 Materials and methods

2.1 Substrate

Musa paradisiaca L. leaves (= banana leaf waste) were collected from new fruit and vegetable market, Rohtak city (28.8955°N, 76.6066°E) in Haryana, India. The collected banana leaf waste was washed thoroughly with tap water and finally with distilled water. The banana leaf waste was then dried in the oven at 70 °C till constant weight, grounded (0.2 to 1.0 mm size) and stored in sealed plastic bag for further use.

2.2 Pretreatment

The alkali and acidic pretreatment was carried out by soaking powdered banana leaf waste in aqueous solutions of 0.1 N NaOH and 0.1 N H_2SO_4 respectively in the ratio of 1:10 (w/v). For steam pretreatment, powdered banana leaf waste was soaked in distilled water in the ratio of 1:10 (w/v). The mixture of each flask was autoclaved at 121 °C for one hour. The pretreated biomass was washed extensively with tap water and finally with distilled water until neutral pH was obtained. For further use, the pretreated biomass was oven-dried at 70 °C till constant weight and stored in air tight plastic bags at room temperature.

2.3 Cellulase production and its harvestation

Cellulase production by *Aspergillus niger* JD-11 was achieved under optimized solid state fermentation (SSF). The alkali pretreated dried banana leaf waste (5 g), with particle size between 0.2 to 1.0 mm, taken

in 500 mL Erlenmeyer flasks was moistened with 20 mL modified Mandels and Sternburg's (MS) basal medium [24] containing (g/l) proteose peptone, 2.5,yeast extract, 7.5; $(NH_4)_2SO_4$, 10; KH_2PO_4, 5.0; $MgSO_4.7H_2O$, 0.3; urea, 0.3; $CaCl_2$, 0.3; $MnSO_4.H_2O$, 0.0016; $ZnSO_4.7H_2O$, 0.0014; $FeSO_4.7H_2O$, 0.005; $CoCl_2$, 0.002 and Tween 80, 0.1% (v/v). The initial pH of moistening media was set to 6.0 and autoclaved at 121 °C for 20 min. Then, the cooled flasks were inoculated with 1.0 mL of spore suspension having 5×10^7 spores. Spore suspension was prepared from five days old culture slants of *Aspergillus niger* JD-11 by adding 10 mL sterilized distilled water containing 0.1% Tween 80 (v/v). All the contents of flasks were mixed well and incubated at 30 °C under static conditions.

Enzyme harvesting was done on 6th day of incubation period by adding 100 mL citrate buffer (0.05 M, pH 4.8) in each culture flask and then keeping the flasks for one hour at 30 °C in incubator shaker at 200 rpm. The entire slurry was then squeezed through a muslin cloth. The culture extracts were centrifuged at 4 °C for 15 min at 10,000 rpm to remove spores and debris. The crude enzyme extract was concentrated by acetone fractionation with increasing volume of solvent. Chilled acetone was added to pre-chilled crude enzyme extract in the ratio of 1:0.5 and the mixture was saved at 20 °C for four hours, followed by centrifugation at 10,000 rpm for 15 min at 4 °C. For fractional precipitation, the supernatant was regained and acetone was added step by step to further increase the ratio to 1:4. From each step, the pellets were allowed to dry at room temperature to remove the residual acetone. Then, the pellets were resuspended in 2.0 mL of 0.05 M citrate buffer (pH 4.8) and dialyzed overnight.

2.4 Cellulase estimation

The cellulase activity was measured as filter paper activity units (FPU) according to IUPAC method of [17]. Whatman No. 1 filter paper strip of dimension 1×6 cm (~50 mg) was placed into each assay tube and were saturated with 1.0 mL of citrate buffer (0.05 M, pH 4.8). After adding 0.5 mL of appropriately diluted enzyme, the tube was incubated at 50 °C for one hour in water bath. At the end of incubation period, the enzyme reaction was terminated by the addition of 2.0 mL of DNSA (Dinitrosalicylic acid) reagent [26]. The tubes were incubated for five minutes in a boiling water bath and the color developed was read at 540 nm against a reagent blank. Filter paperase (FPase) activity was calculated by following the concept that 0.37 FPU (Filter Paper Unit) of enzyme will release 2.0 mg of glucose under the above assay conditions and was specified as FPUs.

2.5 Saccharification

Saccharification of unpretreated, alkaline, dilute acid and steam pretreated banana leaf waste was conducted in 250 mL screw-capped Erlenmeyer flasks having 50 mL of 0.05 M citrate buffer (pH 5.0) using acetone fractionated crude cellulases of *Aspergillus niger* JD-11 at 150 rpm for 70 h in an incubator shaker. One mL sample was periodically removed from each flask and centrifuged at 4 °C for 10 min at 10,000 rpm to remove unhydrolyzed residue and the clear supernatant was analysed for reducing sugars (RS). To find the best saccharification conditions of banana leaf waste, the effect of various factors such as enzyme loading (5–15 FPU/g), temperature (40, 45 and 50 °C), surfactant concentration [0.05–0.15% Tween 80 (v/v); 0.05–0.15% polyethylene glycol 6000 (PEG 6000) (w/v)] and substrate concentration [2 to 6% (w/v)] were studied selecting one variable at one time.

2.6 Analysis of reducing sugars

The estimation of reducing sugars liberated during enzymatic saccharification was done according to DNSA method [26]. The amount of total reducing sugars was calculated as follows:

$$Reducing\ sugars\ yield\ (mg/g)$$
$$= \frac{Amount\ of\ reducing\ sugars\ produced\ after\ enzymatic\ hydrolysis}{Amount\ of\ dry\ pretreated\ biomass}$$

2.7 Fermentation and ethanol production from enzymatic hydrolysate

Yeast strain, *Sacchromyces cerevisiae* (*S. cerevisiae*), obtained from the Department of Microbiology, Maharishi Dayanand University, Rohtak was maintained on yeast peptone dextrose (YPD) agar slants and was sub-cultured at regular intervals. The fermentation was done in 250 mL screw-capped Erlenmeyer flasks with a working volume of 100 mL. The fermentation of enzymatic hydrolysate obtained from enzymatic hydrolysis of dilute acid pretreated banana waste was carried out with the help of *S. cerevisiae*. The fermentation was done using initial reducing sugars concentration of 40 g/L in the medium at pH 5.5, 150 rpm and 30 °C for 30 h and ethanol production was estimated at every 6 h. The fermentation medium was sterilized by autoclaving at 121 °C for 15 min, cooled and inoculated with 24 h old seed culture (5% v/v) of *S. cerevisiae*. One mL of the samples was withdrawn at regular intervals and centrifuged at 10,000 rpm for 10 min at 4 °C. The supernatant was filtered using filters of 0.45 μm size

and the ethanol content was estimated by gas chromatography as outlined in NREL Laboratory Analytical protocol # 011 [50].

3 Results and discussion

To evaluate the effect of pretreatment conditions on the digestibility in comparison with the control or unpretreated condition, enzymatic hydrolysis of alkaline, dilute acid and steam pretreated banana waste was carried out as shown in Fig. 1. The enzymatic hydrolysis was executed at 2% solid loading (w/v), 10 FPU/g of crude cellulase dosage, 50 °C and 150 rpm. The reducing sugars produced were estimated at every 10 h upto 70 h. The reducing sugars attained after enzymatic hydrolysis of pretreated banana waste were found to be ranging from 99.52 to 358.11 mg/g as compared to unpretreated biomass producing sugars yield of 33.83 to 55.03 mg/g. Maximum reducing sugars of 55.03, 157.91, 303.70 and 358.11 mg/g were obtained respectively from unpretreated, steam, alkali and dilute acid pretreated banana waste after enzymatic hydrolysis in 70 h. However, around 95% reducing sugars were obtained consistently in the first 40 h of the

enzymatic hydrolysis and therefore no significant increment in the release of reducing sugars could be achieved after 40 h. Corroborative results were also obtained by [10] as well as [19] who obtained maximum reducing sugars after 48 h of enzymatic hydrolysis. There were 2.86, 5.46, 6.51 times more reducing sugars production from steam, alkali and dilute acid pretreated banana waste respectively as compared to the unpretreated biomass. As the dilute acid pretreated biomass of banana waste yielded maximum reducing sugars during enzymatic hydrolysis, subsequent hydrolysis experiments were carried out using the same biomass only.

3.1 Effects of enzyme loading on enzymatic hydrolysis

To analyse the impact of cellulase loading on the yield of reducing sugars, the enzymatic hydrolysis of dilute acid pretreated banana waste was carried out at 2% solid loading (w/v), 50 °C and 150 rpm with cellulase dosage of 5 to 15 FPU/g and the reducing sugars produced were estimated at every 10 h upto 70 h. With increasing
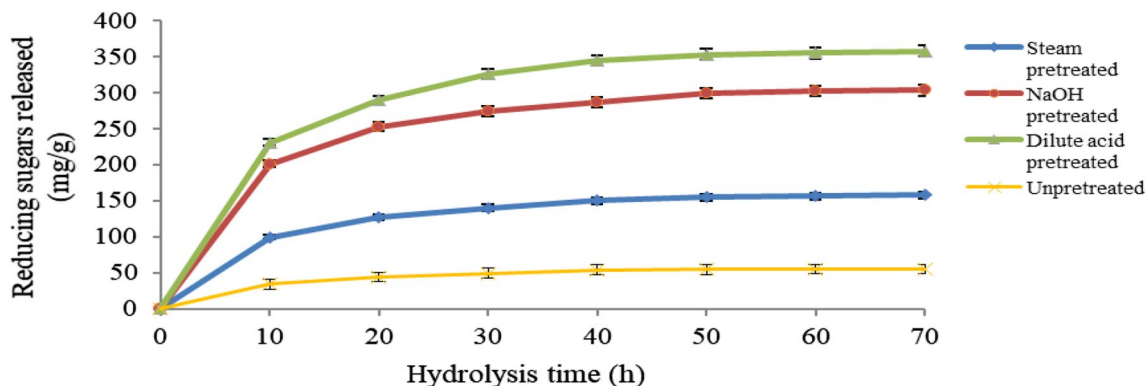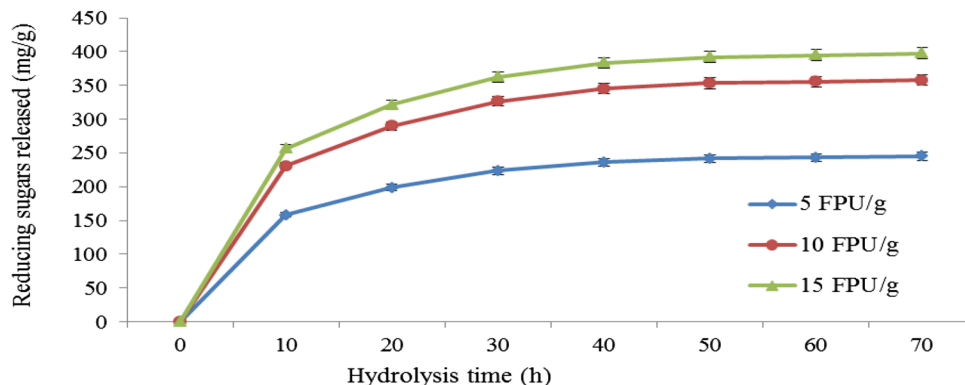

Fig. 1 Enzymatic hydrolysis of differentially pretreated banana waste biomass at enzyme dosage of 10 FPU/g, substrate loading of 2% (w/v), pH 5.0 and 50 °C

Fig. 2 Effect of enzyme dosage on enzymatic hydrolysis of dilute acid pretreated banana waste biomass at 2% substrate loading (w/v), pH 5.0 and 50 °C

cellulase loading of 5 to15 FPU/g Fig. 2, the reducing sugars enhanced from 245.31 to 397.57 mg/g.

An increase of 38% in the yield of reducing sugars was gained from banana waste at cellulase loading of 15 FPU/g as compared to cellulase loading of 5 FPU/g. Owing to this reason, the cellulase loading of 15 FPU/g pretreated substrate was used in rest of the saccharification experiments as the same yielded highest reducing sugars. Corroborative results were also observed by [54] when they found 45% enhancement in the yield of reducing sugars with the increase in cellulase loading from 3 to 15 FPU/g pretreated newspapers. However, Rodhe et al. [38] found the optimum enzyme loading of 25 FPU/g for the maximum hydrolysis of sorghum straw and achieved 70% (546 mg/g) hydrolysis yield but [3] reported maximum reducing sugars yield of 731 mg/g from alkali treated water-hyacinth biomass at an enzyme loading of 8 FPUs and 2400 U of β-glucosidase per gram of the biomass. Collaterally, Kim et al. [22] reported 83% glucan hydrolysis of ammonia pretreated barley husk at the enzyme loading of 15 FPU/g glucan while [42] accomplished maximum 59.8% hydrolysis of NaOH pretreated sunflower hulls with crude cellulases of *Trichoderma reesei* at dosage of 25 FPU/g substrate.

## 3.2 Effects of temperature on enzymatic hydrolysis

To determine the optimum temperature required for saccharification of dilute acid pretreated banana waste, the enzymatic hydrolysis was performed at 40–50 °C at substrate loading of 2% (w/v) in 0.05 M citrate buffer (pH 5.0) and cellulase loading of 15 FPU/g for 70 h and reducing sugars released were estimated at every 10 h. Maximum reducing sugars yield of 346.63, 455.91 and 397.57 mg/g were achieved respectively at 40, 45 and 50 °C Fig. 3. As the maximum reducing sugars yield was obtained at 45 °C, all the succeeding hydrolysis experiments were performed at 45 °C. Aswathy et al. [3] also accounted maximum reducing sugars yield of 731 mg/g obtained from alkali treated water-hyacinth biomass at 45 °C. Although Saha and Cotta [39] reported maximum yield of 428 mg/g of total sugars from pretreated rice hulls at 45 °C but [55] and [38] found

maximum rate of hydrolysis at 50 °C. Thus, the reduced enzyme activity at higher temperature of 50 °C could have occurred owing to the thermal inactivation of enzymes as also reported by [20].

3.3 Effects of surfactant addition on enzymatic hydrolysis

Several studies have reported the enhancement of enzymatic saccharification yields by using various surfactants [38, 57, 58]. In the present study also, the effect of surfactants on enzymatic hydrolysis was investigated by the addition of Tween 80 (v/v) and PEG 6000 (w/v) in the range of 0.05 to 0.15%. The experiments of enzymatic hydrolysis were conducted at 2% substrate loading, enzyme dosage of 15 FPU/g in 0.05 M citrate buffer (pH 5.0) and 45 °C and the reducing sugars were estimated at every 10 h up to 70 h. As presented in Fig. 4, with 0.05, 0.1 and 0.15% Tween 80 added, hydrolysis of dilute acid pretreated banana waste resulted in maximum reducing sugars yield of 470.39, 480.58 and 501.94 mg/g respectively, whereas with the addition of 0.05, 0.1 and 0.15% PEG 6000, maximum reducing sugars yield of 488.49, 510.91 and 524.83 mg/g was obtained respectively. Thus, the hydrolysis yield was increased by 3.2, 5.4 and 10.1% with the addition of 0.05, 0.1 and 0.15% Tween 80 and by 7.1, 12.1 and 15.2% with the addition of 0.05, 0.1 and 0.15% PEG 6000 respectively as compared to the control. Therefore, the results clearly indicated towards the maximum hydrolysis of the substrate with the addition of 0.15% PEG 6000 (524.83 mg/g) as compared to 0.15% Tween 80 (501.94 mg/g). An identical observation was also reported by [23] when they reported that the addition of PEG 6000 at 50 mg/g biomass of hydrothermally pretreated wheat straw caused an increase of glucose yield by 23% at 5% (w/w) loading. However, the positive effect on enzymatic hydrolysis by addition of PEG on various steam pretreated lignocellulosic substrates was also reported by [45] but [32] claimed that addition of PEG not only enhanced enzymatic hydrolysis yield but also improved enzyme recycling by minimizing the enzyme activity loss owing to adsorption during the hydrolysis which might have occurred because the

Fig. 3 Effect of temperature on enzymatic hydrolysis of dilute acid pretreated banana waste biomass at enzyme dosage of 15 FPU/g, 2% substrate loading (w/v) and pH 5.0
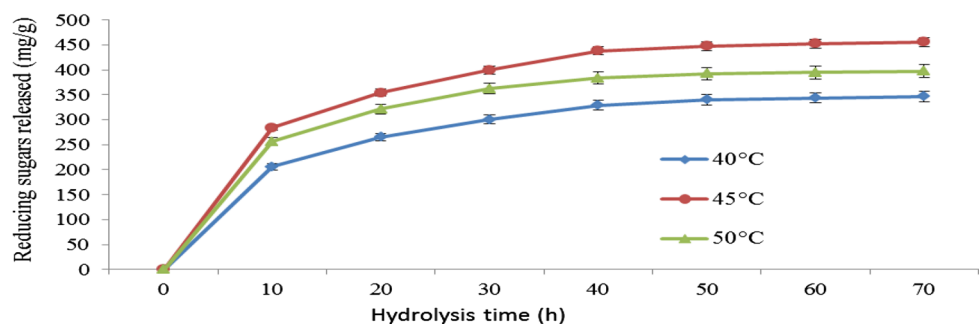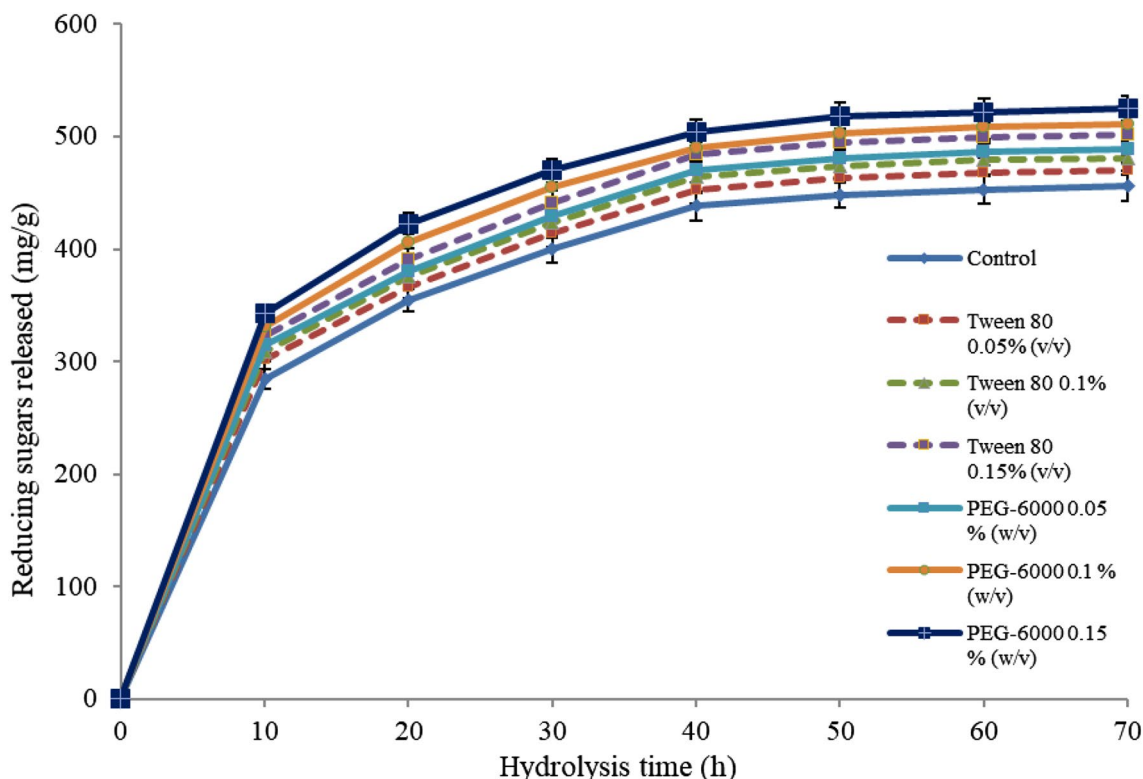
Fig. 4 Effect of different concentrations of surfactants on enzymatic hydrolysis of dilute acid pretreated banana waste at 45 °C, enzyme dosage of 15 FPU/g, 2% substrate loading (w/v) and pH 5.0

surfactants have been found to increase the enzyme stability by protecting them either from thermal denaturation by shear forces [21] or by adsorption onto lignin preventing the non-productive binding of enzymes to the surface of lignin [14]. Hence, the use of cost-effective surfactants must be investigated further as they could be able to reduce the enzyme loading, enhance enzyme activity and facilitate enzyme recovery and hence diminish the operational cost of the process [2].

3.4 Effects of substrate concentration on enzymatic hydrolysis

For analysing the effect of substrate concentration on the enzymatic hydrolysis, the experiments were conducted at different substrate loadings from 2 to 6% (w/v) in 0.05 M citrate buffer (pH 5.0) at cellulase dosage of 15 FPU/g and 45 °C and the reducing sugars were estimated at every 10 h upto 70 h. PEG 6000 (0.15% w/v) was added in the medium. It was done so as [30] reported that the cellulose conversion was influenced more by the substrate concentration as compared to agitation speed. As depicted in Fig. 5, no significant difference in the yields of reducing sugars could be observed within the substrate loading range of 2 to 6% during the enzymatic hydrolysis of

dilute acid pretreated banana waste. The maximum reducing sugars yields of 524.83, 519.98 and 500.46 mg/g were obtained at substrate loading of 2, 4, and 6% respectively. Therefore, the results clearly indicated that 2% substrate loading yielded maximum reducing sugars from the dilute acid pretreated banana waste. Similar findings were also observed by [8, 11, 16, 38]. However, with the increase in substrate concentration, the decrease in hydrolysis yield per gram of substrate might be because of the reduction in aqueous movable phase, inefficient solid mixing and end product inhibition [28, 49].
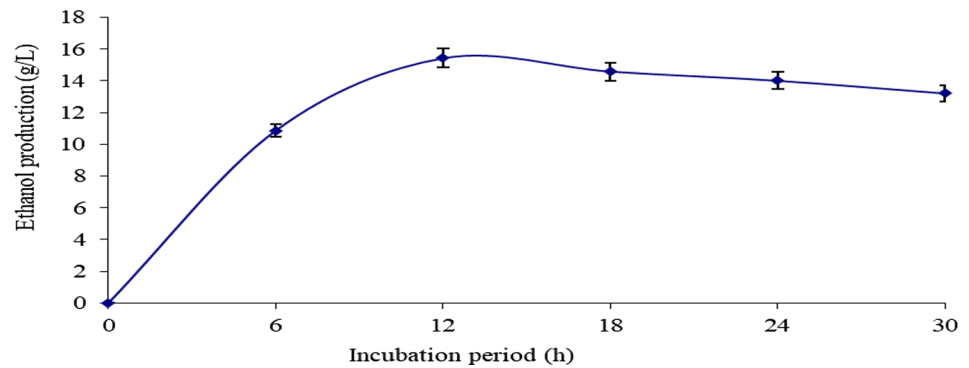
3.5 Ethanol production from enzymatic hydrolysate

Figure 6 depicts the time-course of fermentation profile from enzymatic hydrolysate of banana waste and the fermentation parameters are listed in Table 1.

In the present study, highest value of 15.43 g/L of ethanol was obtained with ethanol yield of 0.38 g/g sugar and productivity of 1.28 g/L/h after 12 h of fermentation of banana waste hydrolysate, containing initial reducing sugars concentration of 40 g/L, by *Saccharomyces cerevisiae*. Corroboratively, maximum ethanol level (21 g/L) was reached after 24 h of fermentation with 20% (w/w) banana peels [33] using *Kluyveromyces marxianus*. Although Singh

Fig. 5 Enzymatic hydrolysis of dilute acid pretreated banana waste at different substrate concentrations added with 0.15% PEG 6000 (w/v) at enzyme dosage of 15 FPU/g, pH 5.0 and 45 °C
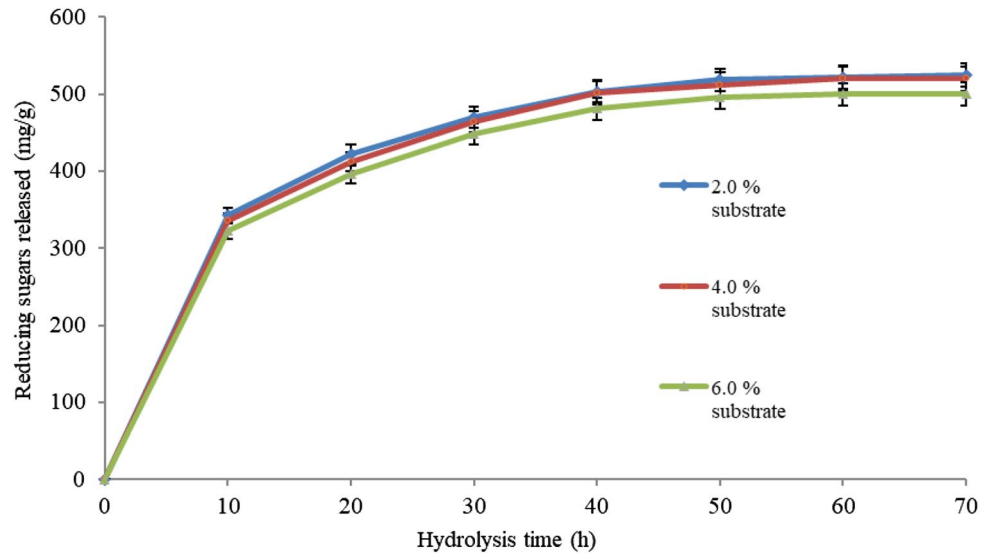
Fig. 6 Time-course of ethanol production by *S. cerevisiae* at 30 °C from enzymatic hydrolysate of banana waste

Table 1 Ethanol production from enzymatic hydrolysate using *S. cerevisiae*

Reducing sugar concentration in enzymatic hydrolysate (g/L)	Incubation time (h)	Ethanol concentration (g/L)	Ethanol yield (g/g sugar)	Ethanol productivity (g/L/h)
40	12	15.43 ± 0.52	0.38 ± 0.01	1.28 ± 0.03

and Bishnoi [43] obtained ethanol production of 4.3 g/L with ethanol volumetric productivity of 0.12 g/L/h and ethanol yield of 0.23 g/g by fermentation of water-hyacinth enzymatic hydrolysate with initial reducing sugars concentration of 35 g/L using *S. cerevisiae* but [55] found the maximum ethanol concentration of 12.39 g/L after 8 h of fermentation in the hydrolysate of horticultural waste with ethanol yield and productivity of 0.49 g/g of sugar and 1.55 g/L/h respectively. Collateral results were also

obtained by [51] who reported the ethanol production of 3.38 and 2.0 g/L with yield of 0.54 and 0.40 g/g from hydrolysates of fungal pretreated wheat straw and banana stem respectively by using *S. cerevisiae* for fermentation. Likewise, Geng et al. [16] found the ethanol production of 11.69 g/L with ethanol yield 0.43 g/g and ethanol productivity 1.46 g/L/h from hydrolysate of horticultural waste at eight hours using *S. cerevisiae*. Moreover, respective ethanol production of 14.4 and 14.9 g/L was obtained from enzymatic hydrolysate of water-hyacinth and water lettuce using *S. cerevisiae* in 9 h [27].

4 Conclusion

In the present study, the banana leaf waste is explored as a potential feedstock for bioethanol production. The feedstock was saccharified by making use of crude cellulases of *Aspergillus niger* JD-11 under varying conditions. The crude cellulases yielded maximum reducing sugars (524.83 mg/g) from the dilute acid pretreated banana leaf

waste at 45 °C, 15 FPU/g enzyme loading and 2% substrate loading in the presence of 0.15% PEG 6000 (w/v) as surfactant. Then, the fermentation of enzymatic hydrolysate was carried out using initial reducing sugars concentration of 40 g/L in the medium at pH 5.5, 150 rpm and 30 °C for 30 h with the help of *Saccharomyces cerevisiae*. Highest value of 15.43 g/L of ethanol with yield of 0.38 g/g sugar and productivity of 1.28 g/L/h was obtained after 12 h of fermentation of the enzymatic hydrolysate. Overall the results and findings provide significant insights that encourage further research in this area in such a way that the abundant banana waste could be envisaged as a budding source of bioethanol production. The ethanol yield may further be enhanced not only by using best recombinant yeast strains which can simultaneously ferment both hexoses and pentoses present in the enzymatic hydrolysate but also by optimization of fermentation parameters.

Compliance with ethical standards

Conflict of interest All contributing authors declare no conflicts of interest.

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