Statistical optimisation of saccharification process using Amorphophallus paeoniifolius tubers into fermentable sugars for bioethanol production in stirred tank batch reactor (STBR)

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Abstract
Current technologies to produce biofuels from various renewable feedstocks have considerably captured vast scientific attention since they can be used as an alternative fuel. Bioethanol being one of the most interesting biofuels and due to its positive impact on the environment has been categorised significantly in terms of scientific and technological investments. The aim of this study was to investigate tubers of Amorphophallus paeoniifolius biomass as a feedstock for bioethanol production. The composition analysis of A. paeoniifolius tubers revealed high carbohydrate content (78.30 ± 0.33%). The feedstock was subjected to physicochemical pre-treatment by treating with dilute acid followed by pressure cooking. The pre-treatment factors were optimised by CCD using RSM approach. The optimum condition was found to be 1.32% (v/v) of HCl, 5.83% (w/v) of elephant foot yam biomass and 66.84 min of pressure-cooking time yielding 45.87 g/L of total sugar. The second-order polynomial equation was generated for the saccharification of biomass and validated with R² 0.89. The fermentation of pre-treated biomass in the presence of Saccharomyces cerevisiae MTCC170 yielded 22.12 ± 0.62 g/L of bioethanol at 120 h utilising 92% of initial total sugar. The resultant ethanol yield and productivity was estimated to be 0.51 g/g and 0.30 g/(L/h), respectively. The Gompertz model equation was applied to experimental data using nonlinear regression with least square method and the kinetic fermentation parameters such as maximum ethanol concentration (Pₘ), production rate (rₚₘ), and lag phase (h) were estimated to be Pₘ = 21.90 g/L, rₚₘ = 0.57 g/(L/h) and tₐ = 8.22 h.

Keywords Amorphophallus paeoniifolius · Central composite design (CCD) · Stirred tank batch reactor (STBR) · Bioethanol

1 Introduction
Rise in global warming in recent years has promoted the research to focus on various aspects of alternative renewable fuel sources. This has captivated the production and utilisation of various bioresources for their progressive application thus making an attempt in reducing the fossil fuel consumption significantly [1]. Although substantial research has been collaborated in the resultant production of bioethanol, there is still a need for steady implementation of processes and optimisation techniques [2]. It is reviewed that the present biofuel demand is expected to rise between 250 and 500 billion litres per year till 2045 [3]. To meet this exorbitant demand, there is a necessity to quantify a number of unexplored feedstock in order to meet the concerned capacity production with its advancement. As per the availability of reports, some of the successfully explored potential starchy sources like wheat [4, 5], corn [6, 7], barley [8, 9], sweet potato [10], maize [11], rice [12], potato [13], cassava [14],and waste food grains [15] already has gained importance with defined technology and resources. Though these food crops have a huge demand in agriculture, there are all possibilities that they may pose threats of food vs. fuel conflicts [16]. But
surplus volume of selected starchy sources can always be grown in field to compensate this status. It is estimated that 2.7 billion tons of these crops are harvested annually and are available for use [17]. The total annual production of starchy feedstock depends on the fertile soil, available land and productivity. In the USA, 35% of the harvested maize grain is utilised for bioethanol production which yields 53 million litres of bioethanol annually [18]. Cassava starch grown in subtropical countries is another starchy residue used for bioethanol production and is compatible with current corn to ethanol technology [19]. Sorghum is grown in dry region and can become a good substitute for ethanol production [20].

Production of bioethanol is governed by various process parameters such as degree of hydrolysis, inoculum size, broth pH, and fermentation time [21]. Yeast plays a critical role in fermenting wide range of hexose and pentose sugars into bioethanol, thus enhancing the quantity and rate of bioethanol production [22]. Optimisation of pre-treatment process parameters is a critical step in the production of an effective and economic process. In the last decades, various research have been focusing on identification and demonstration of pre-treatment techniques for bioethanol production [23–25]. However, the nature of feedstock decides the type of pre-treatment and economy of process. To reduce the cost of overall process, the factors affecting the pre-treatment steps need to be identified at an optimum condition [26]. An optimisation technique adapts systematic statistical approach to standardise the maximum output based on the input given to the process. It is also carried out to achieve maximum or minimum response, depending on the process parameters [27].

Amorphophallus paeoniifolius tuber (elephant foot yam) is a plant belonging to the family of aroids, Araceae. It is grown abundantly in the tropical areas of the world for its edible tubers. These tubers possess high starchy content amounting almost 85% of the total dry weight [28–30]. The present research aims to focus on unexplored source of elephant foot yam biomass (EFYB) and pre-treatment optimisation for bioethanol production. The optimisation of saccharification process was carried out using central composite design (CCD), and the experimental response was fitted to the second-order mathematical model. The hydrolysate obtained after the optimum pre-treatment process was subjected to fermentation in the presence of yeast Saccharomyces cerevisiae MTCC 170. Furthermore, fermentation studies were carried out in laboratory-stirred tank batch reactor (STBR), and the rate of ethanol formation and substrate utilisation was determined. The kinetic parameters of bioethanol fermentation were fit into the experimental data using nonlinear regression with least square method.

2 Materials and methods

2.1 Raw material

Elephant foot yam (EFY) was collected from the coastal district of Karnataka, India (12°52’01.5”N, 74°50’21.6”E). Full-grown 7–8-month-old yam was harvested for this study. The EFY was cleaned with fresh water to remove soil and other unwanted debris. The whole yam, except the dark brown outer skin was used as a substrate for pretreatment and bioethanol production. The EFY tubers were chopped into smaller pieces and sun dried for 3 days to remove excess moisture. The chopped tubers were further dried in hot air oven at 60 °C for 12 h. The bone-dry samples were milled to flour and stored in an airtight container for further use.

2.2 Compositional analysis of EFYB

The chemical compositions of EFYB such as carbohydrate, starch, protein and ash content were determined experimentally by adapting AOAC standard protocols [31]. The compositional analysis was carried out by taking 1 g of EFYB. The total sugar in the biomass was estimated by using spectrophotometric technique as per Dubois method [32].

2.3 Pre-treatment process

The powdered EFYB was subjected to treatment with dilute acid followed by pressure cooking to hydrolyse the carbohydrate to fermentable sugar. The process was carried out by taking 100 mL of acid in 250 mL screw-capped bottle to avoid any evaporation loss during pressure cooking. After the pre-treatment process, the hydrolysate was separated by filtering it through ceramic mesh filter and the amount of total sugar was quantified.

2.3.1 Acid pre-treatment

The pre-treatment of biomass with dilute mineral acid is proven to be one of the effective methods in industrial scale [33]. Dilute acid pre-treatment significantly reduces the reaction activation energy, thereby improving the cellulose hydrolysis rate [24]. The acid pre-treatment of EFYB was carried out by mixing the biomass with 100 mL of dilute hydrochloric acid (HCl). To study the correlation among the acid concentration and biomass handling capacity, the acid strength was varied between 1 and 9% (v/v) by
keeping EFYB load constant at 5% (w/v). Further, EFYB load was varied between 1 and 10% (w/v) by treating it constantly with HCl concentration at 3% (v/v).

### 2.3.2 Physicochemical pre-treatment

Physicochemical pre-treatment is a cost-effective and efficient technique with lower-energy consumption. In this process, steam penetrates into the biomass and expands cell wall which facilitates acid to hydrolyse the carbohydrates [34]. The acid-treated EFYB was pressure cooked in 12 L domestic cooker in 250 mL screw-capped bottle. The contents were pressure cooked at 121 °C at 15 psi by varying the residence time between 20 and 80 min. The volume of water inside the pressure cooker was maintained constant to avoid any variation in pre-heating time. Post-pressure cooking, the contents were cooled to ambient temperature and filtered. The total sugar released after the hydrolysis was determined.

### 2.4 Optimisation of pre-treatment parameter using CCD

The pre-treatment process parameters such as acid concentration, EFYB load and pressure-cooking time significantly affecting the degree of hydrolysis were optimised using response surface methodology (RSM). In RSM, the effect of pre-treatment factors on the release of total sugar was represented by second-order polynomial equation as shown in Eq. (1) [35–37].

$$\text{Total sugar} = a_0 + \sum_{i=1}^{3} a_i x_i + \sum_{i<j}^{k} a_{ij} x_i x_j + \sum_{i=1}^{3} a_{ii} x_i^2$$  \hspace{1cm} (1)

where $a$ is the regression coefficient and $x$ is the independent factor.

The experiments were designed and analysed using statistical software MINITAB 17 trial version. A central composite design with three pre-treatment factors taken in 5 coded levels (−α, −1, 0, +1 and +α) consisting of 20 experimental combinations were taken in this study. ± α corresponds to the star or axial point having a value of ± 1.68, and other coded values (−1, 0 and +1) represented the low, mid- and high-range value, respectively.

The factors such as acid concentration (A), EFYB load (B) and pressure-cooking time (C) were taken as independent variable, and total sugar released during the hydrolysis was taken as a response or dependent variable. The CCD levels were taken by conducting pre-treatment experiment in which one factor was varied by retaining other factors at fixed value. In design, the mid-value (0) was selected by increasing the range of factors until a maximum response (total sugar) was obtained. The experimental combination for CCD and response is tabulated in Table 1. To minimise any error in measurement, the total sugar was taken in triplicate.

### 2.5 Culture and media preparation

Saccharomyces cerevisiae MTCC 170 was procured from Microbial Type culture collection and GenBank (MTCC), Pune, India. The lyophilised culture was revived in 100 mL media composition of 2 g glucose, 2 g peptone and 1 g yeast extract. After the reactivation, culture was subjected to gram staining to check for any possible contamination.

### 2.6 Bioethanol fermentation in STBR

Bioethanol fermentation was carried out using laboratory-stirred tank batch reactor (STBR) (Fig. 1) with 1 L capacity, manufactured by Borg Scientific, Tamil Nadu, India. The glass reactor was fed with 700 mL of production media, consisting of EFYB hydrolysate pre-treated under optimal pre-treatment condition. The media was autoclaved at 121 °C for 20 min, and pH was adjusted to 6.9–7 using a pH controller. The reactor and content were cooled to room temperature by circulating chilled water through cooling coil. Sterile silicon tubes were connected to inlet port, sampling port and inoculation port. The aerobic condition was maintained by...
sparging 2 mL/min sterile air through a nozzle sparger. The vent was connected with sterile 0.2 m air filter.

*S. cerevisiae* cultured in activation media, and 6 h log phase culture was taken for inoculation; 5% (v/v) of log phase culture was transferred to the reactor aseptically through the inoculation port using peristaltic pump. Contents inside were kept under suspension using turbine impeller of variable speed, during the operation of the reactor. Cell maintenance under suspension was set at 50 rpm to prevent any cell death due to nutrient depletion and also to avoid cell disruption due to rotation of agitator blade.

Sampling was carried out at 24 h interval and sampling port was regularly sterilised using IPA to avoid any contamination. These samples were analysed using gas chromatography (GC) for ethanol estimation, subsequently measuring bioethanol concentration. The reduction in total sugar and ethanol yield was also accounted.

### 2.7 Analysis

The analysis of ethanol samples were done in GC (Shimadzu Corporation, Tokyo, Japan) equipped with a flame ionisation detector (FID) and ZB-Wax capillary column (30 m × 0.25 mm × 0.25 μm) using high-purity N₂ (99.99%) as the carrier gas. The temperature of the injector and detector was maintained at 180 °C and 200 °C, respectively. The column temperature was increased at controlled rate by holding initial temperature at 40 °C for 1 min. Further column temperature was increased at a ramp rate of 10 °C/min to 80 °C and hold for 1 min. The column temperature was raised at 25 °C/min to 150 °C and hold for 1 min. The analysis was carried out by taking 0.2 μL of fermentation sample.

### 2.8 Prediction of kinetic model

Kinetic model of any fermentation process describes the formation of fermentation product with respect to time. Thus, kinetic model generated for a process modulates the design for process control, reducing the operational cost, enhancing the product quality and yield, respectively [38]. In the present investigation, bioethanol production was modelled by modified Gompertz model (Eq. 2). This model predicts the kinetic terms of bioethanol fermentation such as maximum rate of productivity, maximum bioethanol concentration and production lag time. These kinetic values can be used to scale up the bioethanol fermentation process [39, 40].

where \( P \) is the ethanol concentration (g/L), \( P_m \) is the maximum ethanol concentration (g/L), \( r_{pm} \) is the maximum productivity of bioethanol, \( t_L \) and \( t \) are the time of lag phase and fermentation time, respectively. Bioethanol yield over the total sugar consumption and % sugar utilisation was calculated as per Eq. 3 and Eq. 4 shown below [40].

\[
Y_{P/S} = \frac{P_f - P_0}{S_0 - S_f} \tag{3}
\]

\[
\% \text{sugar consumption} = \left( 1 - \frac{S_f}{S_0} \right) \times 100 \tag{4}
\]

where \( Y_{P/S} \) is the bioethanol yield, \( P_f \) and \( P_0 \) are the final and initial bioethanol concentration (g/L), \( S_f \) and \( S_0 \) are the final and initial sugar concentration (g/L).

### 3 Results and discussion

#### 3.1 Physicochemical pre-treatment of EFYB

The compositional analysis of EFYB accommodated for 11.56 ± 0.30% protein, 6.7 ± 0.12% moisture, 3.44 ± 0.027% ash content and 78.30 ± 0.33% carbohydrate. This rich carbohydrate content in EFYB proves to be a potential source for bioethanol production. The rich concentration of carbohydrates in biomass eases the hydrolysis process and thus increases the sugar yield per weight of biomass, thus increasing the overall bioethanol yield and productivity. Parameter values obtained in this study were in good agreement to the reports by Behera et al. [30] and Suriya et al. [29].
marginal change in the composition could be attributed to geographical location, analysis method and accuracy.

Pre-treatment of EFYB was processed by treating with dilute HCl followed by pressure cooking. The effect of acid concentration, EFYB load and pressure cooking time on total sugar is presented in Fig. 2. From the figure, it is evident that the change in pre-treatment parameter greatly affected the hydrolysis rate. Increase in EFYB load indicated a positive effect on total sugar release till 6% (w/v) thereby releasing 43.38 ± 0.29 g/L of sugar (Fig. 2a), respectively. Further increase in biomass load beyond 6% (w/v) stabilises the hydrolysis process. This could be attributed due to an increase in biomass load, thus increasing the surface area further leading to poor wettability of biomass surface.

The hydrolysis process increases at a steeper rate by increasing the time of pressure cooking of acid treated biomass. The pressure point inside the pressure cooker was attained at 15 min of heating. The maximum sugar release (41.38 ± 0.45 g/L) was observed at 40 min of pressure cooking (Fig. 2b). However, extending the pressure cooking time resulted in a significant loss of total sugar (32.95 ± 0.29 g/L) which could be due to evaporation loss and degradation of released sugar to form inhibitory compounds [41–43]. It was also observed that higher residence time of pressure cooking yielded reduced volume of hydrolysate making the process unfavourable for fermentation. Thus, the present study indicates that high temperature (121 °C) and lower residence time had an impact to the method and was more favourable for hydrolysis.

The effect of dilute HCl on total sugar release was studied by varying the acid concentration from 1% (v/v) to 9% (v/v) (Fig. 2c). Increase in acid concentration to 3% (v/v) effectively released the total sugars (46.11 ± 1.8 g/L). Studies have been reported regarding utilisation of mineral and organic acids in the pre-treatment process to obtain C6 and C5 sugars [34]. However, sugar loss steadily increased by further increasing the acid concentration above 5% (v/v), which further resulted in a decrease in total sugar release. This conversion of carbohydrate to sugar was effective in dilute acid concentration as well utilising lesser time of hydrolysis.

### 3.2 Optimisation of physicochemical pre-treatment condition

The experiments were performed as per the factor combination shown in Table 1, and response was measured. The statistical significance of experimental data was performed by analysis of variable (ANOVA) and regression analysis presented in Table 2. Using response surface method, the release of total sugar was analysed with second-order polynomial equation. ANOVA results describe that pressure-cooking time and quadratic terms of EFYB load and pressure-cooking time had a $p$ value of less than 0.05, indicating the appropriate significant effect on total sugar release. The interaction among the pre-treatment factors having $p > 0.05$ did not have any significant effect on response. From the Fisher’s $F$-test, the model $F$ values reported in this study were observed to be 9.53. This higher $F$ value and the lack-of-fit $F$ value (3.90) indicate that the model was best fit. The insignificant $p$ value ($p = 0.081$) also supports the fitness of the derived model. The high coefficient of determination value for total sugar ($R^2 = 0.896$) also support the significance of the model generated.

The regression equation in coded form was deduced to represent the release of total sugar as given in Eq. 1.

\[
\text{Total sugar (g/L)} = 33.9 + 5.41A + 11.01B + 1.081C + 0.41A^2 - 0.588B^2 - 0.00804C^2 - 1.418AB - 0.0067BC + 0.0033BC
\]  

3-D interaction plot for total sugar release is presented in Fig. 3. The interaction plot for the physicochemical pre-treatment was studied at constant hold values of 3% (v/v) HCl, 6% (w/v) EFYB and 40 min of pressure-cooking time. From Fig. 3a, it can be described that lower and higher values of HCl and EFYB load had a negative effect on rate of hydrolysis at lower pressure cooking time and sugar released increased as time of pressure cooking was raised to a higher level (Fig. 3b). These observations depict that at lower temperature and pressure acid cannot penetrate deep into the cell wall and thus hydrolysis of carbohydrate.
seems to be ineffective. Similar trend was observed for increase in EFYB load and time. An increase in residence time of biomass inside the cooker enhances the sugar release but at higher residence time, sugar gets degraded into phenolic compounds. This can be observed in Fig. 3c, wherein total sugar concentration was found to be reduced at higher residence time.

As observed from the surface plot presented in Fig. 3, it being symmetric and optimum at the centre and within the range of level explored, curved lines in the plot (Fig. 3) prove the interactions among the factors with $p < 0.05$.

The reduced regression model was generated to get optimum conditions for pre-treatment from response optimiser tool. The optimised condition for total sugar release was 1.32% (v/v) of HCl, 5.83% (w/v) of EFYB and 66.84 min of pressure-cooking time. The generated model was validated by conducting experiment at optimal condition and comparing the experimental value with model predicted

### Table 2 ANOVA results for the optimisation of total sugar (g/L)

| Source                     | df | Adj SS  | Adj MS  | $F$-Value | $P$-Value* |
|----------------------------|----|---------|---------|-----------|------------|
| Model                      | 9  | 1343.13 | 149.24  | 9.53      | 0.001      |
| Linear                     | 3  | 1057.15 | 352.38  | 22.50     | 0.000      |
| A                          | 1  | 11.27   | 11.27   | 0.72      | 0.416      |
| B                          | 1  | 1.54    | 1.54    | 0.10      | 0.761      |
| C                          | 1  | 1044.35 | 1044.35 | 66.69     | 0.000      |
| Square                     | 3  | 221.34  | 73.78   | 4.71      | 0.027      |
| $A^2$                      | 1  | 2.41    | 2.41    | 0.15      | 0.703      |
| $B^2$                      | 1  | 79.61   | 79.61   | 5.08      | 0.048      |
| $C^2$                      | 1  | 149.10  | 149.10  | 9.52      | 0.012      |
| 2-Way interaction          | 3  | 64.64   | 21.55   | 1.38      | 0.306      |
| $A \times B$               | 1  | 64.35   | 64.35   | 4.11      | 0.070      |
| $A \times C$               | 1  | 0.14    | 0.14    | 0.01      | 0.926      |
| $B \times C$               | 1  | 0.09    | 0.09    | 0.01      | 0.912      |
| Error                      | 10 | 156.59  | 15.66   |           |            |
| Lack-of-fit                | 5  | 124.61  | 24.92   | 3.90      | 0.081      |
| Pure error                 | 5  | 31.98   | 6.40    |           |            |
| Total                      | 19 | 1499.73 |         |           |            |

*P ≤ 0.05 is significant.

![Fig. 3 a–c 3-D surface plot for the pre-treatment of EFYB for bioethanol production](image-url)
value. At optimum pre-treatment condition, the total sugar at 45.87 g/L was well correlated with the predicted value of total sugar at 43.48 g/L being a good fit model.

### 3.3 Ethanol fermentation in lab scale-stirred tank batch reactor

The direct fermentation of EFYB hydrolysate in STBR was carried out for different fermentation time intervals. The concentration of bioethanol and substrate utilisation with respect to fermentation time is shown in Fig. 4. The EFYB hydrolysate with initial sugar concentration of 46.87 g/L was taken as substrate for bioethanol fermentation in the presence of *S. cerevisiae*. The media was fed with 5% (v/v) of *S. cerevisiae*, and fermentation was carried out at 37 °C. The sugar concentration reduced to 3.92 g/L at the end of fermentation, which amounts to 92% total sugar utilisation in 120 h. The bioethanol concentration at 120 h of fermentation was found to be 22.12 ± 0.62 g/L. The yield and productivity of ethanol calculated at the end of 120 h fermentation was observed to be 0.51 g of ethanol/g of substrate and 0.30 g/(L/h), respectively.

High bioethanol yield and productivity shows that the EFYB hydrolysate was effectively converted into bioethanol and was free from any fermentation inhibitor. These fermentation parameter obtained in the present study is comparable with the values reported in the literature [44–49].

### 3.4 Estimation of kinetic parameters

The kinetic parameters of bioethanol production from EFYB by *S. cerevisiae* were estimated by fitting the experimental data to modified Gompertz model. This model represents the relationship between ethanol produced and fermentation time between experimental finding and model-calculated value (Fig. 5).

The experimental data of bioethanol fermentation was fitted to the proposed model equation using Microsoft Excel 2010 solver by nonlinear regression using least square technique. The calculated correlation coefficient was above 0.99 suggesting the accuracy of the model. The fitness of the generated kinetic model was predicted by calculating the mean square error (MSE) and variance. The lesser value (9.85 × 10⁻⁵) of MSE and variance (δ = 1.19 × 10⁻⁸) for bioethanol production thus supports the efficiency of the kinetic model generated.

The kinetic parameters of the modified Gompertz model for bioethanol production was estimated and found to be $P_m = 21.90$ g/L, $r_{pm} = 0.57$ g/(L/h) and $t_L = 8.22$ h. The predicted value of productivity in the present research output was observed to be in good agreement with experimental value.

### 4 Conclusion

The process developed in this study was more economic due to the involvement of minimal unit operation and rapid saccharification process to release fermentable sugar to produce bioethanol. The present study investigated the potentiality of elephant foot yam (*A. paeoniifolius*) biomass as a successful feedstock for bioethanol production. The feedstock under investigation was hydrolysed prior to fermentation by the action of HCl acid followed with pressure cooking. The process parameter affecting the hydrolysate concentration was studied by varying one factor at a time and statistically

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**Fig. 4** Effect of fermentation time on bioethanol yield and substrate consumption for EFYB hydrolysate

**Fig. 5** Fitting the modified Gompertz model to experimental data
parameters were determined by using nonlinear regression and 0.30 g/(L/h), respectively. Gompertz model was accurately fit into bioethanol fermentation process, and kinetic parameters were determined by using nonlinear regression with the least square method.

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Declarations

Conflict of interest The authors declare no conflict of interest.

References

1. Lin Y, Tanaka S (2006) Ethanol fermentation from biomass resources: current state and prospects. Appl Microbiol Biotechnol 69:627–642
2. Khan I, Akhtar MW (2011) Bioenergy production from plant biomass: bioethanol from concept to reality. Nat Preced. https://doi.org/10.1038/npre.2011.6286.1
3. Zaky AS, Greetham D, Tucker GA, Du C (2018) The establishment of a marine focused biorefinery for bioethanol production using seawater and a novel marine yeast strain. Sci Rep 8:1–14. https://doi.org/10.1038/s41598-018-30660-x
4. Mikulski D, Klosowski G (2020) Microwave-assisted dilute acid pretreatment in bioethanol production from wheat and rye stilages. Biomass Bioenerg 136:105528. https://doi.org/10.1016/j.biombioe.2020.105528
5. Sayaslan A, Koyuncu M, Türker S et al (2018) Use of durum wheat clear flour in vital gluten and bioethanol production. J Cereal Sci 80:50–56. https://doi.org/10.1016/j.jcres.2018.01.014
6. LuizaAstolfi A, Rempel A, Cavanhi VAF et al (2020) Simultaneous saccharification and fermentation of Stevulina sp. and corn starch for the production of bioethanol and obtaining biopeptides with high antioxidant activity. Bioresearch Technol 301:122698. https://doi.org/10.1016/j.biortech.2019.122698
7. Szambelan K, Nowak J, Szwengiel A et al (2018) Separate hydrolysis and fermentation and simultaneous saccharification and fermentation methods in bioethanol production and formation of volatile by-products from selected corn cultivars. Ind Crops Prod 118:355–361. https://doi.org/10.1016/j.indcrop.2018.03.059
8. Liao B, Hill GA, Roesler WJ (2012) Stable expression of barley α-amylase in S. cerevisiae for conversion of starch into bioethanol. Biochem Eng J 64:8–16. https://doi.org/10.1016/j.bej.2012.02.004
9. Wang X, Tian S, Lou H, Zhao R (2020) A reliable method for predicting bioethanol yield of different varieties of sweet potato by dry matter content. Grain Oil Sci Technol. https://doi.org/10.1016/j.gaost.2020.06.002
10. Weber CT, Trierweiler LF, Trierweiler JO (2020) Food waste bioenergy advocating circular economy: bioethanol and distilled beverage from sweet potato. J Clean Prod 268:121788. https://doi.org/10.1016/j.jclepro.2020.121788
11. Chuck-Hernandez C, Perez-Carrillo E, Serna-Saldivar SO (2009) Production of bioethanol from steam-flaked sorghum and maize. J Cereal Sci 50:131–137. https://doi.org/10.1016/j.jcres.2009.04.004
12. Prasad S, Kumar S, Yadav KK et al (2020) Screening and evaluation of cellulolytic fungal strains for saccharification and bioethanol production from rice residue. Energy 190:116422. https://doi.org/10.1016/j.energy.2019.116422
13. Juodeikiene G, Cernauskas D, Vidmantiene D, et al (2014) Combined fermentation for increasing efficiency of bioethanol production from Fusarium sp. contaminated barley biomass. In: Catalysis Today 108–114. https://doi.org/10.1016/j.cattod.2013.09.028
14. Lyu H, Zhang J, Zhai Z et al (2020) Life cycle assessment for bioethanol production from whole plant cassava by integrated process. J Clean Prod 269:121902. https://doi.org/10.1016/j.jclepro.2020.121902
15. Melikoglou M, Turkmen B (2019) Food waste to energy: forecasting Turkey’s bioethanol generation potential from wasted crops and cereals till 2030. Sustain Energy Technol Assessments 36:100553. https://doi.org/10.1016/j.seta.2019.100553
16. Mohr A, Raman S (2015) Lessons from first generation biofuels and implications for the sustainability appraisal of second generation biofuels. Effic Sustain Biofuel Prod Environ Land-Use Res 63:281–310. https://doi.org/10.1016/j.enpol.2013.08.033
17. Vendruscolo F (2015) Starch: a potential substrate for biohydrogen production. Int J Energy Res 39:293–302. https://doi.org/10.1002/er.3224
18. Michael Sauer MS, Mattanovich D, Marx H (2014) Enabling processing technologies. In: Bisaria VS, Kondo A (eds) Renewable resources to commodity bioprocessing of renewable resources to commodity bioproducts, 1st ed. Wiley, p 13
19. Lu C, Zhao J, Yang ST, Wei D (2012) Fed-batch fermentation for n-butanol production from cassava bagasse hydrolysate in a fibrous bed bioreactor with continuous gas stripping. Bioresearch Technol 104:380–387. https://doi.org/10.1016/j.biortech.2011.10.089
20. Phukoeth Ph, Salakka M, Laopaiboon P, Laopaiboon L (2017) Kinetic models for batch ethanol production from sweet sorghum juice under normal and high gravity fermentations: Logistic and modified Gompertz models. J Biotechnol 69:627–642. https://doi.org/10.1016/j.jbiotec.2016.12.012
21. Nikolić S, Mojić L, Rakin M et al (2009) Effect of different fermentation parameters on bioethanol production from corn meal hydrolysates by free and immobilized cells of Saccharomyces cerevisiae var. ellipsoideus. J Chem Technol Biotechnol 84:497–503. https://doi.org/10.1002/jctb.2068
22. Mohd Azhar SH, Abdulla R, Jambo SA et al (2017) Yeasts in sustainable bioethanol production: a review. Biochem Biophys Reports 10:52–61
23. Zhang J, Shao S, Bao J (2016) Long term storage of dilute acid pretreated corn stover feedstock and ethanol fermentability evaluation. Bioresour Technol 201:355–359. https://doi.org/10.1016/j.biortech.2015.11.024
24. Rabemanolontsoa H, Saka S (2016) Various pretreatments of lignocellulosics. Bioresearch Technol 199:83–91. https://doi.org/10.1016/j.biortech.2015.08.029
25. Neves PV, Pitarelo AP, Ramos LP (2016) Production of cellulosic ethanol from sugarcane bagasse by steam explosion: effect of extraction technologies, content, acid catalysis and different fermentation technologies. Bioresour Technol 208:184–194. https://doi.org/10.1016/j.biortech.2016.02.085

26. El-Mekkawi SA, Abdo SM, Samhan FA, Ali GH (2019) Optimization of some fermentation conditions for bioethanol production from microalgae using response surface method. Bull Natl Res Cent 43:1–8. https://doi.org/10.1186/s42269-019-0205-8

27. Raja S, Murty VR (2013) Optimization of aqueous two-phase systems for the recovery of soluble proteins from tannery waste water using response surface methodology. J Eng (United States) 2013:1–10. https://doi.org/10.1155/2013/217483

28. Chattopadhyay A, Saha B, Pal S et al (2010) Quantitative and qualitative aspects of elephant foot yam. Int J Veg Sci 16:73–84. https://doi.org/10.1080/19315260903211852

29. Suriya M, Baranwal G, Bashir M et al (2016) Influence of blanching and drying methods on molecular structure and functional properties of elephant foot yam (Amorphophallus paeoniifolius) flour. LWT - Food Sci Technol 68:235–243. https://doi.org/10.1016/j.lwt.2015.11.060

30. Behera SS, Panda SH, Panda SK, Kumar A (2019) Biochemical analysis of elephant foot yam (Amorphophallus paeoniifolius) lacto-pickle with probiotic Lactobacillus plantarum. Ann Microbiol 69:577–590. https://doi.org/10.1007/s13213-019-01449-8

31. Becker J (2012) Plasmons as sensors. Assoc. Anal. Communities 1:141–144

32. Dubois G (1956) Smith (1956) M. Dubois, KA Gilles, JK Hamilton, PA Rebers and F. Smith, Colorimetric method for determination of sugars and related substances. Anal Chem 28:350–356

33. Taherzadeh MJ, Karimi K (2008) Pretreatment of lignocellulosic biomass using a potential hybrid yeast strain. Appl Biochem Biotechnol 171:771–785. https://doi.org/10.1007/s13205-013-0167-8

34. Kumar P, Barrett DM, Delwiche MJ, Stroeve P (2009) Methods for pretreatment of lignocellulosic biomass for efficient hydrolysis and biofuel production. Ind Eng Chem Res 48:3713–3729. https://doi.org/10.1021/ie801542g

35. Sandesh K, Amin S, Kiran HT, Vaman Rao C (2017) Optimization of pretreatment of Saccharum spontaneum (Kans Grass) biomass for production of alcoholic biofuels. Res J Pharm Biol Chem Sci Optim 8:117–120

36. Sandesh K, Shishir RK, Vaman Rao C (2020) Optimization and comparison of induction heating and LPG assisted acid pretreatment of cocoa pod for ABE fermentation. Fuel 262:116499. https://doi.org/10.1016/j.fuel.2019.116499

37. Tripathi M, Bhatnagar A, Mubarak NM et al (2020) RSM optimization of microwave pyrolysis parameters to produce OPS char with high yield and large BET surface area. Fuel 277:118184. https://doi.org/10.1016/j.fuel.2020.118184

38. Dodić JM, Vučurović DG, Dodić SN et al (2012) Kinetic modeling of batch ethanol production from sugar beet raw juice. Appl Energy 99:192–197. https://doi.org/10.1016/j.apenergy.2012.05.016

39. Fan S, Chen S, Tang X et al (2015) Kinetic model of continuous ethanol fermentation in closed-circulating process with pervaporation membrane bioreactor by Saccharomyces cerevisiae. Bioresour Technol 177:169–175. https://doi.org/10.1016/j.biortech.2014.11.076

40. Srimachai T, Nuithitikul K, O-Thong S, et al (2015) Optimization and kinetic modeling of ethanol production from oil palm frond juice in batch fermentation. Energy Procedia 79:111–118. https://doi.org/10.1016/j.egypro.2015.11.490

41. Singh S, Khanna S, Moholkar VS, Goyal A (2014) Screening and optimization of pretreatments for Parthenium hysterophorus as feedstock for alcoholic biofuels. Appl Energy 129:195–206. https://doi.org/10.1016/j.apenergy.2014.05.008

42. Chaturvedi V, Verma P (2013) An overview of key pretreatment processes employed for bioconversion of lignocellulosic biomass into biofuels and value added products. 3 Biotech 3:415–431. https://doi.org/10.1007/s13205-013-0167-8

43. Shet VB, Palan AM, Rao SU et al (2018) Comparison of response surface methodology and artificial neural network to enhance the release of reducing sugars from non-edible seed cake by autoclave assisted HCl hydrolysis. 3 Biotech 8:1–8. https://doi.org/10.1007/s13205-018-1163-9

44. Yeh RH, Lin YS, Wang TH et al (2016) Bioethanol production from pretreated Miscanthus floridulus biomass by simultaneous saccharification and fermentation. Biomass Bioenerg 94:110–116. https://doi.org/10.1016/j.biombioe.2016.08.009

45. Zhao J, Xia L (2010) Bioconversion of corn stover hydrolysate to ethanol by a recombinant yeast strain. Fuel Process Technol 91:1807–1811. https://doi.org/10.1016/j.fuproc.2010.08.002

46. Kumari R, Pramanik K (2013) Bioethanol production from Ipomoea carnea biomass using a potential hybrid yeast strain. Appl Biochem Biotechnol 171:771–785. https://doi.org/10.1007/s13205-013-0398-5

47. Sathesh-Prabu C, Murugesan AG (2011) Potential utilization of sorghum field waste for fuel ethanol production employing Pachyseren tannophilus and Saccharomyces cerevisiae. Bioresour Technol 102:2788–2792. https://doi.org/10.1016/j.biortech.2010.11.097

48. Shet VB, Sanil N, Bhat M et al (2018) Acid hydrolysis optimization of cocoa pod shell using response surface methodology approach toward ethanol production. Agric Nat Resour 52:581–587. https://doi.org/10.1016/j.anres.2018.11.022

49. Chattopadhyay A, Tiwari AK, Singh D et al (2015) A systematic analytical study on lignocelluloses originated inhibitors in hydrolyzed biomass. Cellul Chem Technol 49:81–85

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