Optimization of pretreatment condition for ethanol production from *Cyperus difformis* by response surface methodology

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Abstract

This study investigated the potential of a new material, so-called small-flowered nutsedge, for bioethanol production. This plant causes the huge loss of rice yield as it competes nutrients, sunlight and other necessary elements with rice plant to grow. The project plans to transform its biomass into valuable product and bring profitable for farmers. The powered raw sample was treated with sodium hydroxide (NaOH) and hydrogen peroxide (H₂O₂) as followed the design experiment. The use of response surface method is helping researcher to save time and effort but still gain meaningful predicted value that closed to the actual value. The highest total sugar was given when the pretreatment condition is solid to liquid ratio of 0.05, 1% NaOH, 1% H₂O₂ for 72 h. The efficiency of hydrolysis can reach 47% after 24 h with cellulase enzyme at 50 °C, 150 rpm and highest ethanol concentration was obtained on the fifth day of fermentation.

Keywords Rice yield · Weed · *Cyperus difformis* · Alkaline pretreatment · Immobilized yeast · Bioethanol

Introduction

Bioethanol considered as fuel is mainly produced via biological processes which take advantages of yeast metabolism. With the current technologies, bioethanol can be biosynthesized from lignocellulosic biomass feedstock (herbaceous and woody plants, agricultural and forestry residues, and a large portion of municipal and industrial solid waste streams) without disturbing food-based source using biochemical and biological pathways (Kang et al. 2014). However, due to the strong recalcitrance of the cell wall structure, the conversion of lignocellulosic into bioethanol requires two more steps before biological fermentation, so-called pretreatment and saccharification (Kang et al. 2014). Generally, while pretreatment aims to disturb the biomass resistance, make cellulose chains more accessible to the enzymes, and transform the crystalline structure of cellulose, saccharification uses acid or enzyme to break down polysaccharide and cellulose into fermentable sugars. It is certain that effective

pretreatment is quite important to a successful enzymatic hydrolysis (Hendriks and Zeeman 2009). The simple sugar then is used as food for yeast and some type of bacteria that transform it into bioethanol and carbon dioxide (CO₂). *Saccharomyces cerevisiae* is the most common yeast strain adopted widely in both lab and industrial scale because of its tolerance to high ethanol concentration and globally distribution.

According to Küüt (2013), 1 kg of hay (grass biomass) containing 93.73% of total solid could be able to yield 72–104 mL of bioethanol. In another word, the grass biomass can be a promising feedstock for biofuel production due to its abundance, and low price. Small-flowered nutsedge (*Cyperus difformis*) is chosen to be a raw material in this study because this weed grows abundantly in rice fields, especially organic rice fields which causes a huge reduction of rice yield (Chauhan and Johnson 2009). There is the attempt of farmers to eliminate this harmful weed out of the field using manual as well as chemical methods which are time consuming and increase the production cost (Kuk et al. 2004; Saha and Cotta 2006). Being similar to other lignocellulosic biomass, the conversion of small-flowered nutsedge into bioethanol has to undergo two more additional steps: alkaline pretreatment and enzymatic hydrolysis. As stated by Saha and Cota (2006), alkaline-peroxide pretreatment was proved as an efficient method to cope with the

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recalcitrance of cell wall of non-woody materials, particularly wheat straw. According to Tarkow and Feist (1969), sodium hydroxide can result in delignification on grass biomass by breaking the ester bonds cross-linkage of lignin and hemicellulose, thus; the porosity on biomass surface increases. Furthermore, under alkaline conditions, H_2O_2 produces hydroxyl radical ($OH^\cdot$) which oxidizes lignin so that enhance the lignin removal process in pretreatment (Ahn et al. 2012). The main advantages of using alkaline pretreatment are high lignin degradation, lesser corrosion effect on equipment, and mild conditions required (Singh et al. 2016). Nevertheless, this method still possesses some drawback such as the production of unexpected salt, no separation of pentoses and hexoses (Singh et al. 2016).

In this study, the pretreatment condition was optimal using response surface method (RSM). RSM is a combination of the mathematical and statistical techniques based on the fit of mathematical models to the experiment results produced from the designed experiment and the confirmation of the model (Antony 2014). RSM is usually applied for modeling and analyzing a process to study the relation among several independent factors and one or more response and the optimization of a process (Montgomery and Runger 2002; Ayeni et al. 2013). This combines experimental designs with a method of constructing new data points by first-order second-order polynomial equations in a sequential testing procedure. Experimental design has recently been used for studying the use of alternative fuels in internal combustion engines. It has been applied to predict and optimize many of biofuel process and its application such as biodiesel, biogas, biohydrogen, and bioethanol which were listed in the review of Mäkelä (2017).

Box-Behnken design (BBD) is the other useful tool of response surface methodology for optimizing model. The advantage of BBD is in pointing out the issue of where the experimental boundaries should be in general and in particular to avoid the unnecessary combination of treatment. However, BBD has only two significant limitations. The first is the number of experimental factors has to be equal or higher than three and the BBD should not be used for fitting other equations but second order polynomial as the below equation (Eq. 2) (Witek-Krowiak et al. 2014). In

general, the variable values are often coded to compare their effects within the design range. As following to the Eq. 1, the coded value is ranged from -1 to 1 and the center point is 0 Mäkelä (2017).

$$X_i = \frac{\theta_i - \theta_{\min}}{\Delta\theta/2} - 1, \quad (1)$$

where x_i denotes a coded value; θ_i , $\theta_{\min}$ and $\Delta\theta$ are the respective variable value, minimum variable value, and variable range (all in original units):

$$Y = \beta_0 + \sum_{i=1}^k \beta_i X_i + \sum_{i=1}^k \beta_{ii} X_i^2 + \sum_{i < j}^k \beta_{ij} X_i X_j + \epsilon, \quad (2)$$

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_4 X_4 + \beta_{11} X_1^2 + \beta_{22} X_2^2 + \beta_{33} X_3^2 + \beta_{44} X_4^2 + \beta_{12} X_1 X_2 + \beta_{13} X_1 X_3 + \beta_{14} X_1 X_4 + \beta_{23} X_2 X_3 + \beta_{34} X_3 X_4 + \epsilon,$$

where Y is response (sugar yield), β_0 is the intercept value, β_i ($i = 1, 2 \dots k$) is the first order model coefficient, β_{ii} represents the quadratic coefficients of X_i , and β_{ij} is the interaction effects. X_i and X_j are the input variables that influence the response variable and ϵ represents the random error. To evaluate the fitting as well as the statistical significance of obtained model, an analysis of variance (ANOVA) should be carried out. The general structure of an ANOVA analysis was given in Table 1. The main function and meaning of ANOVA analysis will be discussed in detail in “Results and discussion”.

Materials and methods

Collection and preparation of materials

Small-flowered nutsedge (*Cyperus difformis*) was collected in the rice fields located inside Maejo University campus ($18^\circ 53' 35.1''N$, $99^\circ 01' 10.8''E$). Dirt and visible contaminants were washed away using tap water. The samples were

Table 1 The general structure of an ANOVA table in an experimental design

Parameter	Degrees of freedom (df)	Sum of squares (SS)	Mean square (MS)	F value
Total corrected	$n - 1$	SS_{tot}		
Model	k	SS_{mod}	MS_{mod}	$MS_{\text{mod}}/MS_{\text{res}}$
Residual	$n - p$	SS_{res}	MS_{res}	
Lack of fit	$n - p - (n_r - m)$	SS_{lof}	MS_{lof}	$MS_{\text{lof}}/MS_{\text{pe}}$
Pure error	$n_r - m$	SS_{pe}	MS_{pe}	

Subscripts; *tot* total, *mod* model, *res* residual, *lof* lack of fit, *pe* pure error, MS SS/df. $p = k + 1$

then placed in a hot air oven at 60 °C and mashed into powder using high speed blender (Otto BE-127) that could pass through 1 mm mesh (Fig. 1). Finally, the powdered samples were kept in ziplock bags and put in a desiccator for further analyzing.

Optimization of alkaline pretreatment using Box-Behnken design

In this study, the four factors of alkaline pretreatment were statistically optimized with RSM using Box-Behnken design (Box and Behnken 1960). The Design-Expert software (Stat-Ease, USA), version 11.0.3.0 was used to build and analyze the experimental design. The four independent variables chosen were ratio of solid/liquid, NaOH concentration (%), H₂O₂ concentration (%), and time (h). The low (− 1), middle (0), and high (1) levels for each factor were given in Table 2. The software displayed totally 27 base runs with 4 runs at the middle points. Repetition of experiments at the central values ensures the accuracy of the data long with reproducibility of the model.

The statistical significance of the model was estimated using analysis of variance (ANOVA) with *p* value less than 0.05 and insignificance of lack of fit tests. The variables that significantly affected the responses were determined using a confidence level above 95% which *p* value less than 0.05. Moreover, the goodness of fit of the model was evaluated by

Table 2 The low, middle, and high level of the factors by BBD for gooseweed and small-flowered nutsedge

Factor	Unit	Symbol	Coded level		
			− 1	0	1
Ratio of solid/liquid	–	A	0.05	0.175	0.3
NaOH	%	B	1	1.5	2
H ₂ O ₂	%	C	0.5	1	1.5
Time	Hour (h)	D	24	48	72

the determination of R^2 , predicted R^2 , and adjusted R^2 coefficients. Three-dimensional surface plots and contours were achieved to demonstrate the effects of independent factors on sugar concentration. A second order polynomial equation was used to test the effects of independent factors on the response to predict the optimal condition divided into linear, quadratic, and interactive components as below (Eq. 3).

$$Y = \beta_0 + \beta_1 A + \beta_2 B + \beta_3 C + \beta_4 D + \beta_{12} AB + \beta_{13} AC + \beta_{14} AD + \beta_{23} BC + \beta_{24} BD + \beta_{34} CD + \beta_{11} A^2 + \beta_{22} B^2 + \beta_{33} C^2 + \beta_{44} D^2, \quad (3)$$

where *Y* is the predicted response (sugar concentration g/g); β_0 is the intercept; $\beta_1, \beta_2, \beta_3$, linear coefficients; $\beta_{11}, \beta_{22}, \beta_{33}$, squared coefficients; $\beta_{12}, \beta_{13}, \beta_{23}$, interaction coefficients.

Fig. 1 Sampling location (a); small-flowered nutsedge in the rice field (b); powder making (c)

A, B, C are coded values for the ratio of solid/liquid, NaOH concentration, H_2O_2 concentration, and time.

Enzymatic hydrolysis

Hydrolysis process was carried out with commercial cellulase enzyme supplied by Union Science Company, Chiang Mai, Thailand. Enzyme assay described by supplier is cellulase (CMC) 2398 units/g, beta glucosidase 577 units/g, and pH 4. Conical flasks containing 200 mL pretreated sample were adjusted to pH 5 and added 2% (v/v) of cellulase enzyme. The mixture was kept at 50 °C and agitated at 150 rpm for 24, 48, and 72 h. The obtained hydrolysate is determined two parameters which are total sugar and reducing sugar. Total sugar and reducing sugar are estimated by following sulfuric-phenol and 3,5 dinitrosalicylic method, respectively (Dubois et al. 1956).

Fermentation

For fermentation, 100 ml of hydrolysate solution was fermented with 2% of yeast (*Saccharomyces cerevisiae* TISTR 5020) bead cells in 250 mL working volume fermenter. This yeast strain was collected at the Faculty of Science, Maejo University. The mixture was incubated at 35 °C for three to five days. The temperature fluctuated due to the high ambient temperature in April in Thailand. After three and 5 days, 50 ml aliquots of samples in the fermenter were collected to measure the percentage of ethanol using Ebulliometer (Dujardin-Salleron, Alcohol Burner).

Statistical analyses

The data obtained were expressed as mean $\pm$ standard deviation (SD). The statistical analyses were performed using one-way analysis of variance (ANOVA), IBM SPSS statistical package version 22.0 (IBM Corp., New York, USA). The statistical significances were reached when $p < 0.05$.

Table 3 Experimental designed runs with actual and predicted values of alkaline pretreatment for small-flowered nutsedge

Run	Factor 1 A:ratio of S/L–	Factor 2 B:NaOH%	Factor 3 C: H_2O_2 %	Factor 4 D:time hours	Total sugar g/g		Residual
					Predicted value	Actual value	
1	0.05	1	1	48	0.1778	0.174	– 0.0038
2	0.175	1	1.5	48	0.0377	0.040	0.0023
3	0.175	1.5	0.5	24	0.0300	0.029	– 0.0010
4	0.3	1.5	0.5	48	0.0524	0.049	– 0.0034
5	0.175	1	0.5	48	0.0450	0.049	0.0040
6	0.175	1	1	72	0.0367	0.035	– 0.0017
7	0.3	1	1	48	0.0481	0.049	0.0009
8	0.05	1.5	0.5	48	0.1770	0.174	– 0.0030
9	0.05	1.5	1	24	0.1615	0.167	0.0055
10	0.175	1.5	1	48	0.0350	0.030	– 0.0050
11	0.05	2	1	48	0.1760	0.162	– 0.0140
12	0.175	2	0.5	48	0.0357	0.045	0.0093
13	0.3	1.5	1	24	0.0458	0.047	0.0012
14	0.175	2	1	24	0.0269	0.030	0.0031
15	0.175	1.5	1	48	0.0350	0.035	0.0000
16	0.175	1.5	1.5	72	0.0471	0.035	– 0.0121
17	0.175	1.5	1	48	0.0350	0.040	0.0050
18	0.3	1.5	1.5	48	0.0565	0.061	0.0045
19	0.175	1	1	24	0.0317	0.030	– 0.0017
20	0.175	2	1.5	48	0.0523	0.060	0.0077
21	0.3	1.5	1	72	0.0488	0.055	0.0062
22	0.05	1.5	1	72	0.1835	0.194	0.0105
23	0.3	2	1	48	0.0553	0.046	– 0.0093
24	0.05	1.5	1.5	48	0.1822	0.187	0.0048
25	0.175	1.5	1.5	24	0.0341	0.027	– 0.0071
26	0.175	2	1	72	0.0469	0.050	0.0031
27	0.175	1.5	0.5	72	0.0420	0.036	– 0.0060

Results and discussion

Optimization of alkaline pretreatment using Box-Behnken design

$$Y_2 = 0.0350 - 0.0626A + 0.0013B + 0.0023C + 0.0062D + 0.0023AB - 0.0002AC - 0.0047AD + 0.0060BC + 0.0038BD + 0.0003CD + 0.0768A + 0.0025B + 0.0052C - 0.0019D, \quad (4)$$

with regards to Eq 4 for small-flowered nutsedge, the coefficient of linear term (A: ratio of solid/liquid) has negative sign indicates that the increased value of the ratio of solid/liquid leads the decrease of sugar that can be seen clearly. With regards to the interaction between tested variables, the Eq. 4 shows that the interaction of ratio of solid/liquid with H_2O_2 concentration and time were found to be negative which implies that the more positive of ratio of solid/liquid is, the more negative the effect of H_2O_2 concentration (time) on sugar yield. This trend can be found on the interaction of the ratio of solid/liquid with NaOH concentration due to the very small value of coefficient. The coefficient of the squared term (time) is negative which means that the

Fig. 2 Experimental data plotted against RSM model predicted data of pretreatment for small-flowered nutsedge

maximum sugar yields at the central point and it decreases when there is an increasing or decreasing the time from central point. On the other hand, the positive values of the rest squared terms (ratio of solid/liquid, NaOH concentration, and H_2O_2 concentration) indicate the minimum value

Table 4 ANOVA analysis of model for optimization of pretreatment for small-flowered nutsedge

Source	Sum of squares	df	Mean square	F value	p value	Conclusion
Model	0.0863	14	0.0062	72.76	< 0.0001	Significant
A-ratio of S/L	0.0470	1	0.0470	554.80	< 0.0001	
B-NaOH	0.0000	1	0.0000	0.2518	0.6249	
C- H_2O_2	0.0001	1	0.0001	0.7712	0.3971	
D-time	0.0005	1	0.0005	5.53	0.0366	
AB	0.0000	1	0.0000	0.2390	0.6337	
AC	2.500E-07	1	2.500E-07	0.0030	0.9576	
AD	0.0001	1	0.0001	1.07	0.3224	
BC	0.0001	1	0.0001	1.70	0.2168	
BD	0.0001	1	0.0001	0.6640	0.4310	
CD	2.500E-07	1	2.500E-07	0.0030	0.9576	
A ²	0.0315	1	0.0315	371.65	< 0.0001	
B ²	0.0000	1	0.0000	0.3805	0.5489	
C ²	0.0001	1	0.0001	1.71	0.2158	
D ²	0.0000	1	0.0000	0.2313	0.6392	
Residual	0.0010	12	0.0001			
Lack of fit	0.0010	10	0.0001	3.87	0.2229	Not significant
Pure error	0.0001	2	0.0000			
Cor total	0.0873	26				
Std. dev.	0.0092		R ²		0.9884	
Mean	0.0717		Adj R ² *		0.9748	
C.V. %	12.84		Pred R ² *		0.9349	
			Adeq Precision*		22.8306	

Adj R²* adjusted R², Pred R²* predicted R², Adeq precision* adequate precision

Fig. 3 **a** 3D plot showing the effect of NaOH concentration and ratio of S/L on sugar yield, and **b** 3D plot showing the effect of H₂O₂ concentration and ratio of S/L on sugar yield

of sugar observed at the central points of these parameters. Table 3 performs the number of experimental runs along with the predicted and observed responses.

ANOVA analysis for the chemical pretreatment model has a F value of 72.76 with low probability value ($P < 0.05$) (Table 4). Furthermore, the lack of fit p value of 0.2229 implies the lack of fit is not significant relative to the pure error. The significant value for model and non-significant value of lack of fit proved the validity of the obtained quadratic model (Rawat et al. 2013; Das et al. 2015). The factors that significantly affected the responses have a confidence level above 95% which p value less than 0.05 as show in

Fig. 4 **a** 3D plot showing the effect of time and ratio of S/L on sugar yield, and **b** 3D plot showing the effect of time and NaOH concentration on sugar yield

Table 4. While the p value for each model term that A , D , and A^2 have significant effect on the total sugar, the other terms that B , C , AB , AC , AD , B^2 , C^2 , D^2 were insignificant. However, these factors could not be reduced to support hierarchy of the model because of high adjusted determination coefficient (adjusted $R^2 = 0.9748$). The goodness of fit of the model was evaluated by the determination of R^2 , predicted R -squared, and adjusted R^2 coefficients. The predicted R^2 of 0.9349 is in reasonable agreement with the adjusted R^2 of 0.9748; i.e. the difference is less than 0.2. Adequate Precision measures the signal to noise ratio and a ratio greater than 4 is desirable (Cai et al. 2012). The ratio of 22.831 indicates an adequate signal. This model can be used to navigate the design space. Guan and Yao (2008) suggested that R^2

Fig. 5 a 3D plot showing the effect of H₂O₂ and NaOH concentration on sugar yield, and b 3D plot showing the effect of Time and H₂O₂ concentration on sugar yield

Fig. 6 Total sugar (TS) and reducing sugar (RS) during enzymatic hydrolysis

Table 5 Total sugar, reducing sugar and ethanol concentration

	Total sugar (g/l)	Reducing sugar (g/l)	Ethanol (g/l)
Start	30.92 ± 0.78	29.51 ± 1.01	0
After 3 days	9.69 ± 0.85	5.24 ± 1.21	7.89 ± 1.58
After 5 days	10.88 ± 0.93	6.87 ± 0.47	12.36 ± 3.73

should be at least 0.80 for the good fit of model. In this case, the value of correlation co-efficient (R^2) and adjusted co-efficient (adj. R^2) are 0.9884 and 0.9748, respectively, which illustrate that the fit of RSM model is significant and can be used to predict the optimal setting up (Fig. 2). 3D-response surface and contour figures (Figs. 3, 4 and 5) obtained by the analysis of the experimental data of BBD, demonstrate a correlation between the two variables in same time while managing the third variable at fixed level.

The optimal values of selected factors for the pretreatment condition of small-flowered nutsedge is solid to liquid ratio of 0.05, NaOH concentration of 1%, H₂O₂ concentration of 1% for 72 h. The total sugar concentration was achieved 0.183 g/g from the regression equation which is near to the experimental value (0.194 g/g ± 0.003).

Hydrolysis/saccharification

Figure 6 shows the changes in reducing sugar after 72 h of enzymatic hydrolysis. After 24 h of the reaction, the amount of reducing sugar increased significantly which makes up 47% of total sugar. It is stated that hemicellulose fraction in small-flowered nutsedge is higher than cellulose (Vu et al. 2017). Besides, hemicellulose is formed of many different monosaccharide units belonging to hexoses and pentose groups so that the hydrolysis of this component has to be carried with the presence of various specific enzymes that can be able to degrade both pentose and hexoses polymers (Vu et al. 2018). In this case, the presence of only cellulase enzyme is not enough to break down all the polysaccharide into simple sugars. This leads to the uncompleted process and causes low sugar yield.

It can be seen from Fig. 6. that the value of total sugar has a slight fluctuation during the hydrolysis time. Total sugar was determined using sulfuric-phenol methods in which all the oligosaccharide in the sample is hydrolyzed completely into single sugar units under the concentrated acid condition (Aswathy et al. 2010). As mentioned above, the limitation of cellulase enzyme to hydrolyze hemicellulose fraction can be a reason for the unchanged amount of sugar. In conclusion, with cellulase enzyme, the hydrolysis process can be done at 50 °C, 150 rpm for 24 h.

Table 6 The highest bioethanol concentration from different feedstocks

Material	Pretreatment and saccharification	Ethanol concentration (g/L)	References
Small-flowered nutsedge	Alkaline-peroxide pretreatment; cellulase enzyme	12.36	Present study
Gooseweed	Alkaline pretreatment; cellulase enzyme	11.84	Vu et al. (2018)
Water hyacinth	Enzymatic hydrolysis	4.4	Aswathy et al. (2010)

Ethanol production

The hydrolysate was collected and fermented with 2% (w/v) *S. cerevisiae* at 35 °C for 5 days. The parameters such as total sugar, reducing sugar, and ethanol were recording at each period of time and performed as below table (Table 5). Ethanol was produced after three days and reached the highest 12.36 ± 3.73 (g/l) after 5 days. In the meantime, sugar contents decreased dramatically after 3 days (Table 5). Even though the obtained ethanol in this research is lower than the one obtained from other lignocellulosic, the ethanol yield can be improved by upgrading pretreatment and hydrolysis method. Table 6 shows the comparison of the highest ethanol concentration after fermentation from different lignocellulosic feedstock.

Conclusion

Bioethanol production from lignocellulosic biomass is a promising route due to the abundance of feedstock. A harmful weed, namely small-flowered nutsedge was studied for bioethanol fermentation. The study conducted alkaline-peroxide pretreatment with the help of response surface methodology to optimize the pretreatment condition. A followed step is enzymatic hydrolysis with cellulase enzyme and achieved 47% of hydrolysis efficiency. The gained bioethanol in this study indicates that small-flowered could be stood as a potential feedstock for bioethanol production. However, a further research is needed to improve the hydrolysis efficiency using different types of enzymes.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest in the publication of this paper.

References

- Ahn DJ, Kim SK, Yun HS (2012) Optimization of pretreatment and saccharification for the production of bioethanol from water hyacinth by *Saccharomyces cerevisiae*. *Bioprocess Biosyst Eng* 35(1–2):35–41
- Antony J (2014) Design of experiments for engineers and scientists, 2nd edn. Elsevier, London
- Aswathy US, Sukumaran RK, Devi GL, Rajasree KP, Singhania RR, Pandey A (2010) Bio-ethanol from water hyacinth biomass: an evaluation of enzymatic saccharification strategy. *Bioresour Technol* 101(3):925–930
- Ayeni AO, Hymore FK, Mudliar SN, Deshmukh SC, Satputea DB, Omoleye JA, Pandey RA (2013) Hydrogen peroxide and lime based oxidative pretreatment of wood waste to enhance enzymatic hydrolysis for a biorefinery: process parameters optimization using response surface methodology. *Fuel* 106:187–194
- Box GEP, Behnken DW (1960) Some new three level designs for the study of quantitative variables. *Technometrics* 2(4):455–475
- Cai BY, Ge JP, Ling HZ, Cheng KK, Ping WX (2012) Statistical optimization of dilute sulfuric acid pretreatment of corn cob for xylose recovery and ethanol production. *Biomass Bioenergy* 36:250–257
- Chauhan B, Johnson D (2009) Ecological studies on *Cyperus difformis*, *Cyperus iria* and *Fimbristylis miliacea*: three troublesome annual sedge weeds of rice. *Ann Appl Biol* 155(1):103–112
- Das S, Bhattacharya A, Haldar S, Ganguly A, Gu S, Ting YP, Chatterjee PK (2015) Optimization of enzymatic saccharification of water hyacinth biomass for bio-ethanol: comparison between artificial neural network and response surface methodology. *Sustain Mater Technol* 3:17–28
- Dubois M, Gilles KA, Hamilton JK, Rebers PA, Smith F (1956) Colorimetric method for determination of sugars and related substances. *Anal Chem* 28(3):350–356
- Guan X, Yao H (2008) Optimization of Viscozyme L-assisted extraction of oat bran protein using response surface methodology. *Food Chem* 106(1):345–351
- Hendriks ATWM, Zeeman G (2009) Pretreatments to enhance the digestibility of lignocellulosic biomass. *Bioresour Technol* 100(1):10–18
- Kang Q, Appels L, Tan T, Dewil R (2014) Bioethanol from lignocellulosic biomass: current findings determine research priorities. *Sci World J* 2014:298153
- Kuk YI, Kim KH, Kwon OD, Lee DJ, Burgos NR, Jung S, Guh JO (2004) Cross-resistance pattern and alternative herbicides for *Cyperus difformis* resistant to sulfonylurea herbicides in Korea. *Pest Manag Sci* 60(1):85–94
- Küüt A (2013) Characteristics of bioethanol fuel obtained from lignocellulose biomass in internal combustion reciprocating engines with spark- and compression-ignition. *Estonian University of Life Sciences, Tartu*, pp 25–28
- Mäkelä M (2017) Experimental design and response surface methodology in energy applications: a tutorial review. *Energy Convers Manag* 151:630–640

- Montgomery D, Runger G (2002) Applied Probability and Statistics to Engineering. McGraw Hill, Mexico
- Rawat R, Kumbhar BK, Tewari L (2013) Optimization of alkali pretreatment for bioconversion of poplar (*Populus deltoides*) biomass into fermentable sugars using response surface methodology. Ind Crops Prod 44:220–226
- Saha BC, Cotta MA (2006) Ethanol production from alkaline peroxide pretreated enzymatically saccharified wheat straw. Biotechnol Prog 22(2):449–453
- Singh R, Krishna BB, Kumar J, Bhaskar T (2016) Opportunities for utilization of non-conventional energy sources for biomass pretreatment. Bioresour Technol 199:398–407
- Tarkow H, Feist WC (1969) A mechanism for improving the digestibility of lignocellulosic materials with dilute alkali and liquid ammonia. ACS Publications, Washington D. C.
- Vu PT, Unpaprom Y, Ramaraj R (2017) Evaluation of bioethanol production from rice field weed biomass. ELSR 3(2):42–49
- Vu PT, Unpaprom Y, Ramaraj R (2018) Impact and significance of alkaline-oxidant pretreatment on the enzymatic digestibility of *Sphenoclea zeylanica* for bioethanol production. Bioresour Technol 247:125–130
- Witek-Krowiak A, Chojnacka K, Podstawczyk D, Dawiec A, Pokomeda K (2014) Application of response surface methodology and artificial neural network methods in modelling and optimization of biosorption process. Bioresour Technol 160:150–160