



# ARTIFICIAL NEURAL NETWORK (ANN) MODELING FOR HYDROGEN PRODUCTION IN A CONTINUOUS ANAEROBIC SLUDGE BLANKET FILTER (ASBF)

P. Mullai<sup>1</sup>, M. K. Yogeswari<sup>2</sup>, K. Sridevi<sup>3</sup>, P. Ronald Ross<sup>4</sup>

<sup>1,2,3</sup> Pollution Control Research Laboratory, Department of Chemical Engineering,

<sup>1,2,3</sup> Faculty of Engineering and Technology (FEAT), Annamalai University, Annamalai Nagar – 608 002, India.

<sup>4</sup> Zoology Wing, Directorate of Distance Education (DDE), Annamalai University, Annamalai Nagar, India.

[pmullai@yahoo.in](mailto:pmullai@yahoo.in)<sup>1</sup>; [indhukrishnan@gmail.com](mailto:indhukrishnan@gmail.com)<sup>2</sup>; [sridevi.kothandapani@gmail.com](mailto:sridevi.kothandapani@gmail.com)<sup>3</sup>

[ross\\_1971@yahoo.co.in](mailto:ross_1971@yahoo.co.in)<sup>4</sup>

## ABSTRACT

A laboratory scale anaerobic sludge blanket filter (ASBF) was operated for 217 days in two phases to investigate the effect of substrate concentration (2.53 – 13 g glucose/L) and hydraulic retention time (6 – 30 h HRT) at mesophilic temperature (32 ± 2°C) and constant pH 5.5 for hydrogen production. At the optimized condition of 11 g glucose/L at 24 h HRT, 99.9% glucose degradation with 2.25 mol H<sub>2</sub>/mol glucose was achieved. The obtained data were modeled by an artificial neural network (ANN). The performance function - determination coefficient (R<sup>2</sup> = 0.9981) value obtained between experimental and simulated hydrogen production rate revealed that ANN model could reliably be used as a simulation model in ASBF.

**Keywords:** Anaerobic sludge blanket filter; ANN model; Hydrogen production rate; Hydraulic retention time (HRT)

## 1. INTRODUCTION

Search for alternative renewable energy have gained utmost importance, in particular for upcoming world stability. Worldwide, 13 terrawatts (1 TW=10<sup>12</sup> W=3.2 EJ/year) of energy was utilized from burning fossil fuels (Goldemberg and Johansson, 2004). This in turn, adds about 6 gigatons per year of C (as CO<sub>2</sub>) to the atmosphere. The reserve of fossil fuel will be exhausted sooner or later. In the present situation, world's energy requirement will increase 6 folds by 2100 (Dunn, 2002). In India, the demand for energy is also increasing day by day and it ranks fifth in the world in terms of energy consumption. The consumption of various energy sources in India is oil (32.3%), natural gas (7.7%), coal (54.4%), nuclear (1.2%), and hydro (4.5%). Approximately 96 % of hydrogen produced from fossil fuels is highly energy-intensive and not environmental friendly. So, there comes a

circumstance to recognise a cost-effective, sustainable, environmental friendly and renewable resource. The rapid decline of energy reserves can be offset by making use of hydrogen, the sustainable, alternative source for fossil fuels (Kothari et al., 2010; Mullai et al., 2013).

Hydrogen is clean, non-polluting, carbon-free, inexhaustible, recyclable and environmental friendly fuel that produces water instead of green house gas emissions when combusted. It is light and generates 2.75 fold greater energy yield (122 KJ/g) than hydrocarbons (Ren et al., 2006). Hydrogen production by biological method gains more consideration than the conventional methods due to their low energy requirement and investment. Amongst, dark fermentation is more feasible method which has more commercial values and offers an excellent provision for practical application of

hydrogen in various fields (Levin et al., 2004; Kothari et al., 2010). Many studies on biohydrogen production using various bioreactors have been carried out including batch reactor (Logan et al., 2002; Mullai et al., 2013), fed-batch reactor (Chin et al., 2003), continuous stirred tank reactor (Lara et al., 2010), packed-bed reactor (Chang et al., 2002), fluidized-bed reactor (Wu et al., 2003), membrane bioreactor (Lee et al., 2009), and up-flow anaerobic sludge blanket reactor (Yu et al., 2002).

Artificial neural network (ANN) has been used to predict hydrogen production since it has many favorable features such as efficiency, generalization, and simplicity. ANN can be considered as non-linear statistical model and a very powerful tool to represent complex non-linear systems (Prakasham et al., 2011). Principally, ANN is a black box (hidden layers) consisting of a sequence of complicated equations for the computation of outputs based on a given series of input values (Jorjani et al., 2008). These layers include an input layer, one or more hidden layers, and an output layer (Figure 1). The use of ANN could be seen in number of modeling studies, such as, modeling of submerged bioreactor treating cheese whey wastewater (Cinar et al., 2006), prediction of chemical desulfurisation of tabas coal (Jorjani et al., 2008), immobilised cell biofilter treating hydrogen sulphide vapors (Rene et al., 2008), estimation of hydrogen production in genetically modified *E. coli* fermentation (Colunga et al., 2010), electrocoagulation of copper from simulated wastewater (Bhatti et al., 2011), membrane bioreactor treating hypersaline oily wastewater (Pendashteh et al., 2011), optimization of key fermentation parameters for hydrogen production (Prakasham et al., 2011), and determination of gold by atomic absorption spectroscopy in industrial wastewater samples (Ebrahimzadeh et al., 2012).

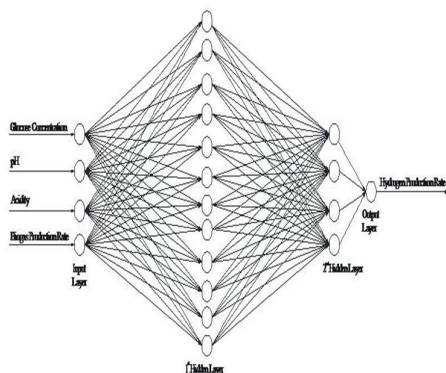


Figure 1 ANN model used in the study

This study proposed to use an anaerobic sludge blanket filter (ASBF), which is a conglomeration of the positive features of the most versatile anaerobic high-rate reactors, anaerobic filter (AF) and the

upflow anaerobic sludge blanket (UASB) reactor for biohydrogen production. The experiments, and the application of ANN modeling for hydrogen production using ASBF are investigated and discussed.

## 2. MATERIALS AND METHODS

### ANAEROBIC SLUDGE BLANKET FILTER (ASBF)

A laboratory-scale anaerobic sludge blanket filter (ASBF), made of perspex tube (Lark Innovative Fine Teknowledge, Chennai, India) was used.

It had a total volume of 5.097 L with an internal diameter of 104 mm and a height of 600 mm and the active liquid volume was 4.502 L. The reactor consisted of three sections: (i) upflow anaerobic sludge blanket (UASB) reactor at the bottom (350 mm in height), (ii) anaerobic filter (AF) at the middle (180 mm height) and (iii) a headspace of 70 mm at the top. At the bottom of the first section, the UASB, an inlet pipe of 8 mm (internal diameter) was attached. A peristaltic pump was used to pump the feed into the reactor through silicon tube which was connected with the inlet pipe. This pipe size was quite enough in avoiding choking by the biomass.

A distributor was attached to the inlet pipe for homogeneous distribution of the influent. A brass check valve of ¼ inch size was fixed at the bottom of the reactor to facilitate the sludge withdrawal. The second section, AF was packed with Fujino spirals (Fujino Spirals India Private Limited, Chennai, India). This packed section retains the suspended sludge within the reactor. At the upper end of the second section, an outlet was provided for the discharge. In the head-space, a provision was made at the topmost part of the reactor for flow of gas. This outlet was connected to a wet gas flow meter.

### Seed Sludge

The municipal sewage sludge collected from a pumping station at Chidambaram, Tamil Nadu, India and cow dung slurry were mixed and sieved with a mesh of 1mm was used as seed sludge.

### Start-up Process

Initially, at 24 h hydraulic retention time (HRT), the reactor was fed with the sewage and cow-dung slurry in the ratio of 1:9 (by volume) for one week. This retention time favored the growth of microbes on the packing section and at the bottom. The sewage content was then increased in steps of 10% by volume until it attained 100% by volume over 10-week duration with one week for each step rise. Thereafter, the fermentation solution as per recommendations of Venkatamohan et al. (2008) was fed into the reactor. One litre of the fermentation solution contained the following nutrients:  $\text{NH}_4\text{Cl}$  -

0.5 g,  $K_2HPO_4$  - 0.25 g,  $MgCl_2 \cdot 6H_2O$  - 0.3 g,  $NiSO_4$  - 0.016 g,  $CoCl_2$  - 0.025 g,  $ZnCl_2$  - 0.0115 g,  $CuCl_2$  - 0.0105 g,  $CaCl_2$  - 0.005 g,  $MnCl_2$  - 0.015 g, and  $FeCl_3$  - 0.025.

#### Substrate

Glucose was used as substrate. The substrate along with fermentation solution was maintained at the pH of 5.5. The feed was adjusted to  $5.5 \pm 0.05$  with 1 N NaOH or 1 N HCl as methanogenic bacteria were either killed or suppressed at that pH (Yu et al., 2002).

#### Reactor Operation

The reactor was operated continuously for 217 days to study the effect of substrate concentration at eight initial substrate concentrations (2.53 – 13 g glucose/L) in phase I and to study the effect of hydraulic retention time at six different HRTs of 30, 24, 18, 12, 9, and 6 h in phase II. When the substrate degradation efficiency was found to remain constant for three consecutive days, the steady-state conditions were assumed to have set-in. Changes in the loading were made only after “stable state” conditions of effluent glucose concentration persisted. The mesophilic temperature ( $32 \pm 2^\circ C$ ) was maintained throughout the study.

#### Monitoring and Analysis

Volatile fatty acid (VFA), pH, and biomass concentration in terms of volatile suspended solids (VSS) were measured according to the standard methods of APHA (1995). Glucose concentration was measured by DNS method using spectrophotometer (Elico, India) at a  $\lambda_{max}$  of 550 nm. Biogas released from the headspace of the reactor was measured by a wet gas flow meter (Toshniwal, India). Hydrogen in the biogas was determined by a gas chromatograph (Shimadzu, 221-70026-34, Japan) equipped with a thermal conductivity detector (TCD) and was packed with porapak Q column. Nitrogen was used as the carrier gas. The operating

The neural network toolbox of MATLAB was used to develop various artificial neural networks and allowed the user to quantitatively and graphically monitor the network training and prediction processes (Mullai and Rene, 2008). The model was trained using different combinations of parameters like influent glucose concentration, pH, acidity, and biogas production rate so as to achieve maximum determination coefficient values. This was achieved by vigorous trial and error method by keeping some training parameters constantly and slowly moving the other parameters over a wide range of values.

#### Determination Coefficient

The closeness of prediction between the experimental and simulated outputs was evaluated by

temperature of the column and detector were  $100^\circ C$  and  $120^\circ C$ , respectively.

### 3. ARTIFICIAL NEURAL NETWORK MODELING APPROACH

An artificial neural network using the feed forward algorithm is the most widely used neural network for prediction purposes. Neural networks acquire their name from the simple processing units in the brain called neurons which are interconnected by a network that transmits signals between them. These can be thought of as a black box device that accepts inputs and produces a desired output. Figure 1 illustrates a feed-forward ANN with four input layers, two hidden layers where 1<sup>st</sup> hidden layer has 12 neurons and 2<sup>nd</sup> hidden layer has 4 neurons with one output.

Each layer consists of neurons which are connected to the neurons in the previous and following layers by connection weights ( $W_{ij}$ ). These weights are adjusted according to the mapping capability of the trained network. An additional bias term ( $\theta_j$ ) is provided to introduce a threshold for the activation of neurons. The input data ( $X_i$ ) is presented to the network through the input layer, which is then passed to the hidden layer along with the weights. The weighted output ( $X_i W_{ij}$ ) is then summed and added to a threshold to produce the neuron input ( $I_j$ ) in the output layer. This is given by Equation (1).

$$I_j = \sum W_{ij} X_i + \theta_j \quad (1)$$

This neuron input through an activation function  $f(I_j)$  to produce the desired output  $Y_j$ . The most commonly used activation function is the logistic sigmoid function which takes the form of Equation (2).

$$f(I_j) = \frac{1}{1 + e^{-I_j}} \quad (2)$$

computing the determination coefficient values as shown in Equation (3) (Rene et al., 2008).

$$R^2 = \left[ \frac{\sum_{i=1}^N (Y_{simulated_i} - \bar{Y}_{simulated})(Y_{experimental_i} - \bar{Y}_{experimental})}{(N-1)S_{Y_{simulated}} S_{Y_{experimental}}} \right]^2 \quad (3)$$

where,  $Y_{simulated_i}$  = Simulated values made by the model

$Y_{experimental_i}$  = Experimental values

$N$  = Number of cases analyzed

$\bar{Y}$  = Average value

$S_Y$  = Standard deviation

#### Average Percentage Error

The average percentage error (APE) used to make quantitative analysis on the simulated results were defined in Equation (4).

$$APE = \frac{1}{n} \sum_{i=1}^n \frac{|X_{1(i)} - X_{2(i)}|}{X_{1(i)}} * 100\% \quad (4)$$

where  $X_1$  and  $X_2$  = Experimental and simulated result sets

$n$  = Number of observations

## 4. RESULTS AND DISCUSSION

### Phase I – Effect of Substrate Concentration

During the initial period, the reactor was fed with 2.53 g glucose/L at 24 h HRT with consistent initial pH of 5.5. After 32 days, the steady state glucose degradation of 53.54 % and hydrogen production rate of 110 mL/d were attained. Minimal glucose consumption and hydrogen yield of 0.65 mol of hydrogen/mol of glucose might be due to inadequate time available for the biomass for acclimatization. On 32<sup>nd</sup> day, the effluent pH and volatile fatty acid (VFA) were 4.95, and 750 mg/L, respectively. For the various glucose concentrations of 3.60, 4.56, 4.89, 6.85, 9.00, 11.00, and 13.00 g/L, the steady state glucose degradation efficiency were 61.06, 73.24, 91.76, 97.18, 99, 99.9, and 96.14% respectively on days, 55, 98, 139, 148, 158, 165, 176 (Figure 2). The corresponding hydrogen production rates were 225, 366, 642, 906, 2184, 3080, and 2976 mL/d (Figure 3) and hydrogen yields were 0.82, 0.88, 1.09, 1.15, 1.97, 2.25, and 1.91 mol of hydrogen/mol of glucose.

It was observed that biohydrogen production rate increased with an increase in glucose concentration from 3.60 to 11.00 g/L. Similar trend was reported by Amorim et al. (2012) in their work on hydrogen production using glucose as substrate. A maximum of 99.9 % glucose degradation efficiency was attained using 11 g/L with hydrogen production rate of 3080 mL/d. Further increase of glucose concentration to 13 g/L, the glucose degradation efficiency and hydrogen production rate decreased to 96.14 %, and 2976 mL/d, respectively (Table I). A substantial decrease in the hydrogen production might be due to higher substrate concentration. This could be attributed to deficient fermentation with substrate or product inhibition during the hydrogen production process (Kyazze et al., 2006; Zhang et al., 2007; Mullai et al., 2011a). Final pH was decreased from 4.48 to 3.3 (Figure 4) but the VFA was increased from 865 to 1980 mg/L on 176<sup>th</sup> day. The observed pH drop was considered as favorable for the effective functioning of the anaerobic bacteria with the inhibition of methanogenic bacteria (Yu et al., 2002). The higher VFA values indicated the arresting of organic acid accumulation within the reactor to enable hydrogen production coupled with

substrate degradation (Lo et al., 2008; Venkatamohan et al., 2008).

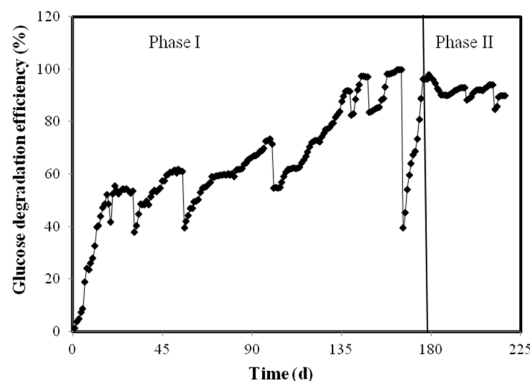


Figure 2 Glucose degradation efficiency for 217 days

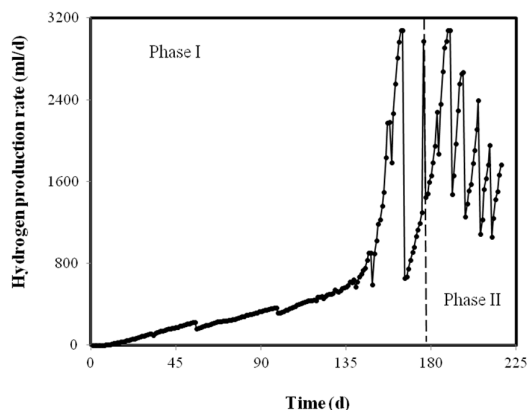


Figure 3 Hydrogen production rate for 217 days

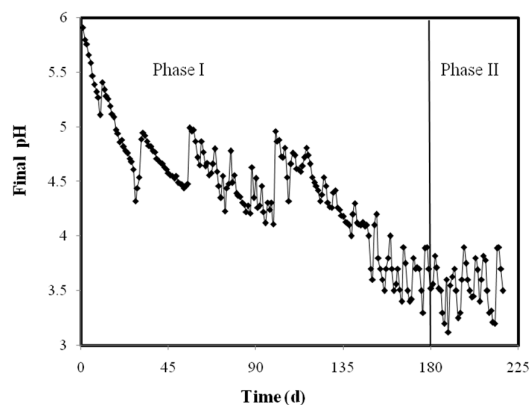


Figure 4 Profile of final pH for 217 days

### Phase II – Effect of HRT

In this phase (177- 217 days), the HRT was gradually varied from 30 to 6 h at initial glucose concentration of around 11g/L and constant pH of 5.5. For the six different HRTs of 30, 24, 18, 12, 9, and 6 h, the steady state glucose degradation efficiency were 92.7, 99.9, 93, 92, 94, and 89.9 %, respectively.

respectively on 183, 190, 197, 205, 211, and 217 days. The corresponding hydrogen production rates were 2280, 3083, 2666, 2395, 1956, and 1769 mL/d. Hydrogen yield of 1.80 mol of hydrogen/mol of glucose was obtained at 30 h HRT. Longer HRT of 30 h favored the growth of biomass (4.05 g VSS/L) rather than the hydrogen yield (Reyes et al., 2012). It might be also due to deficient fermentation processes or accumulation of fermentation products. When the HRT was shortened to 24 h, hydrogen yield increased to 2.25 mol of hydrogen/mol of glucose. When the HRT was further brought down to 18, 12, 9, and 6 h, on 197, 205, 211, and 217 days, obviously a clear fall in the hydrogen yield of 2.17, 1.91, 1.53, and 1.46 mol of hydrogen/mol of glucose

was noticed. This decrease of the hydrogen yield and glucose degradation efficiency (Table I) might be due to washout of greater amount of biomass from the reactor (Mullai, 2002; Yang et al., 2006). As the HRT was decreased, the final pH also decreased and found to be 3.71, 3.55, 3.6, 3.69, 3.4, and 3.5 at steady state conditions. This might be attributed to acid production leading to product inhibition. Generation of acid products caused changes in the metabolic pathways of the biomass might be also reason for drop in pH (Feng et al., 2008; Amorim et al., 2012). This phase conclusively demonstrated that the HRT of 24 h is the best suited for achieving a greater reactor performance.

**Table I. Performance of ASBF under steady state conditions for 217 days**

Time (d)	Substrate concentration (g glucose /l)	HRT (h)	Glucose degradation efficiency (%)	Final pH	VFA (mg/l)	Hydrogen production rate (ml/d)	Hydrogen yield (mol H <sub>2</sub> / mol glucose)
0-32	2.53	24	53.54	4.95	750	110	0.65
33-55	3.60	24	61.06	4.48	865	225	0.82
56-98	4.56	24	73.24	4.31	915	366	0.88
99-139	4.89	24	91.76	4.0	1057	642	1.09
140-148	6.85	24	97.18	3.7	1250	906	1.15
149-158	9.00	24	98.2	3.6	1345	2184	1.97
159-165	11.00	24	99.9	3.4	1500	3080	2.25
166-176	13.00	24	96.14	3.3	1980	2976	1.91
177-183	10.97	30	92.7	3.71	2400	2280	1.80
184-190	11.00	24	99.9	3.55	1500	3083	2.25
191-197	10.80	18	93	3.6	1270	2666	2.17
198-205	10.97	12	92	3.69	1200	2395	1.91
206-211	10.95	9	94	3.4	1185	1956	1.53
212-217	10.90	6	89.9	3.5	1130	1769	1.46

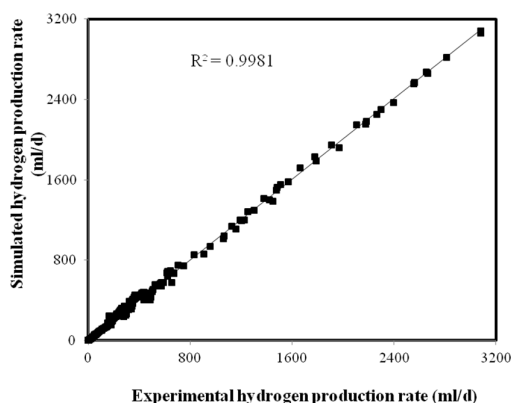
## 5.

### IMPLEMENTATION OF ANN MODEL

A total of 217 days raw data were used for the ANN model. From 217 data, alternate data of about 109 data were used for training, and the rest for testing the data on hydrogen production rate. The network understands the trend contained in the data and correlates the inputs and outputs by finding the optimum set of weight that minimizes the differences between the simulated value and the actual output value (Statsoft, 1998). Hydrogen production rate was

predicted using the influent glucose concentration, pH, acidity, and biogas production rate, as the input values. It was observed that all the data were simulated accurately using the ANN model (Figure 5) with R<sup>2</sup> value of 0.9981. The model was trained using the different combinations of input values to attain maximum determination R<sup>2</sup> values. The result of the test data for hydrogen production rate was very successful. Similarly, Cinar et al. (2006) and Pendashteh et al. (2011) had got satisfactory results

on training and testing data and had a good correlation between the measured and predicted values when treating the cheese whey wastewater and hyper saline oily wastewater respectively. The calculated value of average percentage error is 4.005. The closer the average value of APE is to zero, the model fits well (Mullai et al., 2011b). As the APE values were high, more experimental data are required to increase the validity of the proposed model.



**Figure 5 Comparison between experimental and simulated hydrogen production rate for ANN modeling**

## 6. CONCLUSIONS

An initial glucose concentration of 11g/L and HRT of 24 h were found to be the best combination of conditions, within the ranges studied, for maximum hydrogen yield and glucose degradation efficiency. The present investigation revealed that the presence of twelve neurons in the first hidden layer was effective for ANN model for biohydrogen production rate with four input variables. The simulated hydrogen production rate was in good agreement with the experimental values. This study also indicated the robustness of the ANN model for predicting performance of ASBF.

### Acknowledgments

The authors are thankful to the authorities of the Annamalai University, India for facilities offered and University Grants Commission, India for funding the research project (F. No. 36-23/2008 (SR)) and Prof. Huu-Hao Ngo, University of Technology, Sydney, Australia for his valuable suggestions.

## REFERENCES

[1]. Amorim ELCD, Sader LT, Silva EL. 2012. Effect of substrate concentration on dark fermentation hydrogen production using an anaerobic fluidized bed reactor. *Appl Biochem Biotechnol* 166:1248-1263.

[2]. APHA. 1995. Standard methods for the examination of water and wastewater, 16<sup>th</sup> Edition, American Public Health Association, New York. 5.4 p.

[3]. Bhatti MS, Kapoor D, Kalia RK, Reddy AS, Thukral AK. 2011. RSM and ANN modeling of electrocoagulation of copper from simulated wastewater: Multi objective optimization using genetic algorithm approach. *Desalination* 274:74-80.

[4]. Chang JS, Lee KC, Lin PJ. 2002. Biohydrogen production with fixed-bed bioreactors. *Int J Hydrogen Energy* 27:1167-1174.

[5]. Chin HL, Chen ZS, Chou CP. 2003. Fed-batch operation using *Clostridium butylicum* suspension culture as biocatalyst for enhancing H<sub>2</sub> production. *Biotechnol Prog* 19:383-388.

[6]. Cinar O, Hasar H, Kinaci C. 2006. Modeling of submerged membrane bioreactor treating cheese whey wastewater by artificial neural network. *J Biotechnol* 123:204-209.

[7]. Colunga LMR, Garcia RG, Rodriguez ADL. 2010. Estimation of hydrogen production in genetically modified E. coli fermentations using an artificial neural network. *Int J of Hydrogen Energy* 35:13186-13192.

[8]. Dunn S. 2002. Perspectives towards a hydrogen future. *Cogen. On Site Power Product* 3:55-60.

[9]. Ebrahimzadeh H, Tavassoli N, Sadeghi O, Amini MM. 2012. Optimization of solid-phase extraction using artificial neural networks and response surface methodology in combination with experimental design for determination of gold by atomic absorption spectrometry in industrial wastewater samples. *Talanta* 97:211-217.

[10]. Feng C, Shimada S, Zhang Z, Maekawa T. 2008. A pilot plant two-phase anaerobic digestion system for bioenergy recovery from swine wastes and garbage. *Waste Manage* 28:1827-1834.

[11]. Goldemberg J, Johansson TB. 2004. World Energy Assessment; Overview 2004 update. United Nations Development Programme, USA. 88 p.

[12]. Jorjani E, Chelgani SC, Mesroghli S. 2008. Application of artificial neural networks to predict chemical desulfurization of tabas coal. *Fuel* 87:2727-2734.

[13]. Kothari R, Tyagi VV, Pathak A. 2010. Waste – to – energy: A way from renewable energy sources to sustainable development. *Renew Sust Energ Rev* 14:3154-3170.

[14]. Kyazze G, Perez NM, Dinsdale R, Premier GC, Hawkes FR, Guwy AJ,

- Hawkes AL. 2006. Influence of substrate concentration on the stability and yield of continuous hydrogen production. *Biotechnol Bioeng* 93:971-979.
- [15]. Lara CAAC, Latrille E, Steyer JP. 2010. Optimal control of hydrogen production in a continuous anaerobic fermentation bioreactor. *Int J of Hydrogen Energy* 35:10710 – 10718.
- [16]. Lee DY, Li YY, Noike T. 2009. Continuous H<sub>2</sub> production by anaerobic mixed microflora in membrane bioreactor. *Bioresource Technol* 100:690-695.
- [17]. Levin DB, Pitt L, Love M. 2004. Biohydrogen production: Prospects and limitations to practical applications. *Int J of Hydrogen Energy* 29:173-185.
- [18]. Lo YC, Chen WM, Hung CH, Chen SD, Chang JS. 2008. Dark H<sub>2</sub> fermentation from sucrose and xylose using H<sub>2</sub>-producing indigenous bacteria: Feasibility and kinetic studies. *Water Res* 42:827–842.
- [19]. Logan BE, Oh SE, Kim S, Van Ginkel SW. 2002. Biological hydrogen production measured in batch respirometers. *Environ Sci Technol* 36:2530-2535.
- [20]. Mullai P. 2002. Industrial wastewater treatment using a HUASB reactor, Ph.D thesis, Annamalai University, Annamalai Nagar. 207 p.
- [21]. Mullai P, Rene ER. 2008. A simple multi layered neural network model for predicting bacterial xylanase production by *Bacillus* sp. using *Avena sativa* as substrate. *Biotechnol* 7: 499-504.
- [22]. Mullai P, Ngo HH, Sabarathinam PL. 2011a. Substrate removal kinetics of an anaerobic hybrid reactor treating pharmaceutical wastewater. *JWS* 3:43-54.
- [23]. Mullai P, Arulselvi S, Ngo HH, Sabarathinam PL. 2011b. Experiments and ANFIS modelling for the biodegradation of penicillin - G wastewater using anaerobic hybrid reactor. *Bioresource Technol* 102:5492–5497.
- [24]. Mullai P, Yogeswari MK, Sridevi K. 2013. Optimisation and enhancement of biohydrogen production using nickel nanoparticles – A novel approach. *Bioresource Technol* 141: 212–219.
- [25]. Pendashteh AR, Razi AZ, Chaibakhsh N, Abdullah LC, Madaeni SS, Abidin ZZ. 2011. Modelling of membrane bioreactor treating hypersaline oily wastewater by artificial neural network. *J Hazard Mater* 192:568-575.
- [26]. Prakasham RS, Satish T, Brahmaiah P. 2011. Imperative role of neural networks coupled genetic algorithm on optimization of biohydrogen yield. *Int J of Hydrogen Energy* 36:4332-4339.
- [27]. Ren N, Li J, Li B, Wang Y, Li S. 2006. Biohydrogen production from molasses by anaerobic fermentation with a pilot-scale bioreactor system. *Int J of Hydrogen Energy* 31:2147–2157.
- [28]. Rene ER, Kim JH, Park HS. 2008. An intelligent neural network model for evaluating performance of immobilized cell biofilter treating hydrogen sulphide vapors. *Int J Environ Sci Tech* 5:287-296.
- [29]. Reyes JC, Celis LB, Mondragon FA, Flores ER. 2012. Different start-up strategies to enhance biohydrogen production from cheese whey in UASB reactors. *Int J of Hydrogen Energy* 37:5591-5601.
- [30]. Statsoft. 1998. *Statistica neural network*, Statsoft, Tulsa, OK.
- [31]. Venkatamohan S, Lalit Babu V, Sarma PN. 2008. Effect of various pretreatment methods on anaerobic mixed microflora to enhance biohydrogen production utilizing dairy wastewater as substrate. *Bioresource Technol* 99:59–67.
- [32]. Wu SY, Lin CN, Chang JS. 2003. Hydrogen production with immobilized sewage sludge in three-phase fluidized-bed bioreactors. *Biotechnol Prog* 19:828-832.
- [33]. Yang H, Shao P, Lu T, Shen J, Wang D, Xu Z, Yuan X. 2006. Continuous bio-hydrogen production from citric acid wastewater via facultative anaerobic bacteria. *Int J of Hydrogen Energy* 31:1306-1313.
- [34]. Yu H, Zhu Z, Hu W, Zhang H. 2002. Hydrogen production from rice winery wastewaters in an upflow anaerobic reactor by using mixed anaerobic cultures. *Int J Hydrogen Energy* 27:1359-1365.
- [35]. Zhang ZP, Tay JH, Show KY, Yan R, Liang DT, Lee DJ, Jiang WJ. 2007. Biohydrogen production in a granular activated carbon anaerobic fluidized bed reactor. *Int J of Hydrogen Energy* 32:185–191.