Introductory of Excel Spreadsheet for comparative analysis of linearized expressions of Langmuir isotherm for methylene blue onto rice husk

Yu-Ting Huang[1] · Li-Chiun Lee[1] · Ming-Cheng Shih[2]* · Wei-Ting Huang[2]

[1.] Department of Nutrition, I-Shou University. No.8, Yida Rd., Jiaosu Village, Yanchao District, Kaohsiung City 82445, Taiwan, R.O.C.

[2.] Department of Biological Science and Technology, I-Shou University. No.8, Yida Rd., Jiaosu Village, Yanchao District, Kaohsiung City 82445, Taiwan, R.O.C.

*Corresponding author.

E-mail address: mchshih@isu.edu.tw

Tel: +886-7-6151100 ext.7321.

Fax: +886-7-6577056.

Present address: Department of Biological Science and Technology, I-Shou University. No.8, Yida Rd., Jiaosu Village, Yanchao District, Kaohsiung City 82445, Taiwan, R.O.C.

DOI: 10.29322/IJSRP.9.01.2019.p8587

http://dx.doi.org/10.29322/IJSRP.9.01.2019.p8587

Abstract: Isotherm models are generally implemented to define characterize an adsorption process. Throughout the earlier studies associated with adsorption process, less attention has been paid to choice of linearization methods through the analysis by using Excel spreadsheet. The present study was established to investigate mathematical and statistical properties of these linear types and compare different statistical criteria to specify the best linear curves, fitted to experimental data.Toward these goals, the data obtained from an empirical study on methylene adsorption onto rice husk were applied. The excel softeare is a perfect tool for researchers to solve these linear equations since the Excel is frequently bundled free with new computers. Also, The simple user interface allows researchers to easily use the software without special learning. Due to all these reasons, the *Data Analysis toolpak,* an analysis tool incorporated into Microsoft Excel for Windows, was applied to analyze and find the best linear type fitted to experimental data from five different types of linear expression of Langmuir isotherm equation.

Keywords: Langmuir; Adsorption; Isotherm; Excel

Introduction

Synthetic dyes are broadly applied in manufacturing such as pharmaceutical, textiles, printing, plastics and paper etc. These industries result in serious water pollution by disposing these effluents into streams, rivers ponds, lakes and other municipal sewer, since the factory discharge water are extremely coloration. In fact, the international environmental discharge standards of dyes are also becoming more severe, since the increasing concern for public health and water quality. Therefore, these industries are presently facing severe pressure to reduce the coloration from the factory discharge water. These industries have to remove these coloration wastes from these effluents before discharging them into water bodies to meet the international environmental standards (Hamdaoui and Naffrechoux 2007, Kaur and Kaur 2014, Sakr, Sennaoui et al. 2015, Talbi, Kabbaj et al. 2016). Some of them involve chemical oxidation, photodegradation, adsorption, electocoagulation and reverse osmosis (Basker, Shabudeen et al. 2014). Currently, the adsorption technique is regarded as an substitute technique in comparison with the other treatment techniques. This technique offers many benefits including its easy control and operation , lower environmental impacts, lower costs, high regeneration potential of the adsorbents, less energy and chemical requirements (Chowdhury and Das Saha 2011, Begum, Golam Hyder et al. 2016, Sharififard, Rezvanpanah et al. 2018). There are many parameters affecting the process efficiency , the adsorbent is the most important one. The frequently used adsorbent is activated carbon owing to their extraordinary mechanical and chemical resistance and porous structure (Sharififard, Rezvanpanah et al. 2018). However, they are not cheap and not appropriate for developing countries. Due to these disadvantages of activated carbons, researchers are searching for cheap, affordable and available materials as adsorbents. Some of inexpensive adsorbents have been used include coconut husk, red mud, fly ash and saw dust (Basker, Shabudeen et al. 2014).

The adsorbate molecules attachment to the adsorbent surface by adsorption is a surface phenomenon, the mechanism of mass transfer dominate the rate of adsorption in adsorption equilibrium (Ullah, Nafees et al. 2017). To select appropriate adsorbents and optimize the adsorption efficiency for dye removal The investigation of the adsorbate molecules and adsorbents surfaces interaction at the interface of aqueous-solid can help to realize the adsorption mechanism, which is essential to select appropriate adsorbents and optimize the adsorption efficiency for dye removal (Lyubchik, Lyubchik et al. 2011). In other words, it is important to study the adsorption rate and adsorption equilibrium in optimization of the conditions of dye removal by the choosing materials (Khan, Riazi et al. 2000). The adsorption isotherm, which offers the idea relating to the amount of the adsorbate molecules adsorbed in the adsorbents surfaces as a function of the amount at equilibrium (Lyubchik, Lyubchik et al. 2011). The adsorption isotherm can be determined through the usage of the experimental data, the experimental data will be fitted with the suitable equations by numerical methods. Literature review shows that there are a number of predictive models which govern adsorption equilibrium and can be used to estimate the amount of adsorbate molecules adsorbed on the adsorbent surfaces, out of them the Langmuir isotherm model is the most frequently used (Ryu, Lee et al. 2001, Lyubchik, Lyubchik et al. 2011, Ebrahimian, Saberikhah et al. 2014, Ullah, Nafees et al. 2017).

In literatures, the linear regression analysis was commonly applied to determine the most fitted isotherm for dye removal(Kundu and Gupta 2006, Gimbert, Morin-Crini et al. 2008). This linear regression method is easily done by using Excel or similar spreadsheet software (Ncibi 2008, Hossain, Ngo et al. 2013, Huang and Shih 2016). In this study, statistical analysis with Excel was applied on 5 different types of linear expressions of the Langmuir isotherm equations to analyze the statistical results of these 5 different linear types on the basis of the experiment of methylene blue adsorption from its aqueous solution onto rice husk.

2. Materials and methods

The experimental data used in this research was got from the literature {Huang, 2016 #142}. Rice husk (RH) is used as adsorbent in the literature and the dye used in the literature was methylene blue (MB). The amount of dye adsorbed at equilibrium time (q_e) was calculated using the following equation (Djeribi and Hamdaoui 2008, Huang and Shih 2014):

Amount adsorbed at equilibrium time (q_e) = $\frac{(C_i - C_e)V}{M}$ (1)

where V(L) is the volume of the adsorption solution. M is the amount of adsorbent used (g). C_i and C_e (mg/L) are respectively the initial and equilibrium concentrations of MB in the liquid solution, respectively.

2.1. The Langmuir model

The Langmuir isotherm equation assumes that the adsorption takes place at specific homogeneous sites within the adsorbent and is expressed as (Ho 2006, Hamdaoui and Naffrechoux 2007, Karadag, Koc et al. 2007) :

$$q_e = \frac{q_{\max} K_L C_e}{1 + K_L C_e} \quad (2)$$

Where $q_{\max}$ (mg/g) is the maximum adsorption at the complete saturation of binding sites.; and K_L (L/mg) is the adsorption equilibrium constant of the Langmuir equation..

The linearized forms of Langmuir equation derived on the basis of Eq. [2] is expressed as :

$$\frac{C_e}{q_e} = \frac{1}{K_L q_{\max}} + \frac{C_e}{q_{\max}} \quad \text{type1} \quad (3)$$

Eq.[3] can be rearranged to obtain another 4 different linearized types of Langmuir expressions (Hamdaoui and Naffrechoux 2007):

$$\frac{1}{q_e} = \frac{1}{K_L q_{\max}} \frac{1}{C_e} + \frac{1}{q_{\max}} \quad \text{type2} \quad (4)$$

$$q_e = \frac{-q_e}{K_L C_e} + q_{\max} \quad \text{type3} \quad (5)$$

$$\frac{q_e}{C_e} = -K_L q_e + K_L q_{\max} \quad \text{type4} \quad (7)$$

$$\frac{1}{C_e} = K_L q_{\max} \frac{1}{q_e} - K_L \quad \text{type5} \quad (8)$$

3. Results and discussion

For the current study, the experimental data were fitted to the 5 different linearized forms of Langmuir equations described above and analyzed by the Data Analysis Toolpak in Excel. Values of regression statistics were showed in table1. From the table 1, the multiple R of types 2 and 5 are closer to 1 than the other three linear types. This results indicated that the data clusters of types 2 and 5 are around the regression line. R² is the squared correlation coefficient, which is also called the coefficient of determination. As observed from table 1, the expression of type 2 and type 5 had the R^2 value over 0.999 for the MB adsorption on RH, the R^2 values of type 3 and type 4 expressions are between 0.95 and 0.96 , and the R^2 values of type 1 expression are between 0.98 and 0.99. The highest R^2 values of types 2 and 5 make them to be the best linear expressions of Langmuir equation among these five different types of linearized Langmuir isotherm equations. The values of *standard error* is used to evaluate the standard deviation about the regression. In fact, the standard error of the regression, also recognized as the standard error of the estimate. From table 1, it was observed that expression of type 2 and type 5 had the standard error values lower than 0.024 for the MB adsorption on RH, the standard error values of type 3 and type 4 expressions are higher than 0.02 and lower than 0.04, and the standard error values of type 1 expression are around 0.06. The low standard error values of type 2 and type 5 indicated that the distances between the experiment data points and the fitted values are smaller than the other three linear types. In fact, the standard error and R-squared in the goodness-of-fit section give you a numeric calculation of how well a model fits the experimental data. However, there are differences between the two statistics. Both of these measures, the R-squared offers the relative measure of the dependent variable variance that the model explains, and the values of R-squared are normally from 0 to 1. The standard error offers the absolute measure of the distance between the experimental data points and the regression line.

Table1: Regression Statistics for

	Line1	*Line2*	*Line3*	*Line4*	*Line5*
Multiple R	0.992345	0.999627	0.977407	0.977407	0.999627
R Square	0.984748	0.999254	0.955325	0.955325	0.999254
Adjusted R Square	0.979664	0.999005	0.940434	0.940434	0.999005
Standard Error	0.059962	0.024387	0.409062	0.046593	0.023986
Observations	5	5	5	5	5

The data analysis toolpak also offer the analysis of variance (ANOVA) table . The results were showed in table 2. In table 2, the output value of Significance F is the probability for retaining the null hypothesis , this values indicates the probability that the regression output could have been obtained by chance. A small Significance of F suggest that there is strong evidence for accepting the regression model. Normally, the validity of the Regression output will need the value less than 0.05. If Significance F is greater than 0.05, it's probably better to stop using this regression model. In table 2, the values of Significance F of these 5 different linear types are all less than 0.05, these low values of Significance F indicated that the regression output of these 5 linear types was not a chance occurrence. All these 5 linear types are acceptable to simulate the adsorption of MB by using RH.

Table 2: ANOVA

Line1					
	df	*SS*	*MS*	*F*	*Significance F*

Regression	1	0.696424	0.696424	193.6993	0.000803
Residual	3	0.010786	0.003595		
Total	4	0.70721			
Line2					
	df	*SS*	*MS*	*F*	*Significance F*
Regression	1	2.388348	2.388348	4015.961	8.66E-06
Residual	3	0.001784	0.000595		
Total	4	2.390132			
Line3					
	df	*SS*	*MS*	*F*	*Significance F*
Regression	1	10.73466	10.73466	64.15213	0.004063
Residual	3	0.501994	0.167331		
Total	4	11.23666			
Line4					
	df	*SS*	*MS*	*F*	*Significance F*
Regression	1	0.139267	0.139267	64.15213	0.004063
Residual	3	0.006513	0.002171		
Total	4	0.145779			
Line5					
	df	*SS*	*MS*	*F*	*Significance F*
Regression	1	2.310567	2.310567	4015.961	8.66E-06
Residual	3	0.001726	0.000575		

Total	4	2.312293			

The table 3 provides a summary of the model calculated from these 5 different linear types. The values for the X Variable 1 is the slope, and the values *of intercept* are identified as *y*-intercept. These values will bring into y = slope * x + intercept to calculate the predicated values. In table 3, the P-Values provide the possibility that they are real results and did not occur accidentally. In fact, the P-Values of X Variable 1 is same as *Significance F*. In other words, the p-value is less than 0.05 indicated that your experimental data offer enough evidence to conclude that the regression model will fit the experimental data better than the model with no independent variables. In table 3, the P-values of these 5 different linear types are all less than 0.05, these low values indicated that the regression output of these 5 linear types was not a chance occurrence. All these 5 linear types are acceptable to simulate the adsorption of MB by using RH.

Table 3: The values for the model's coefficients

Line1								
	Coefficients	*Standard Error*	*t Stat*	*P-value*	*Lower 95%*	*Upper 95%*	*Lower 95.0%*	*Upper 95.0%*
Intercept	0.944	0.042	22.55	0.000191	0.811	1.078	0.811	1.077
X Variable 1(Ce)	0.114	0.008	13.92	0.000803	0.088	0.141	0.088	0.141
Line2								
Intercept	0.086	0.015	5.61	0.011201	0.037	0.135	0.037	0.135
X Variable 1 (1/Ce)	1.016	0.016	63.37	8.66E-06	0.965	1.067	0.965	1.067
Line3								
Intercept	8.986	0.839	10.72	0.001737	6.318	11.65	6.318	11.65
X Variable 1 (qe/Ce)	-8.581	1.071	-8.01	0.004063	-12	-5.17	-12	-5.17

Line4								
Intercept	1.035	0.040	26.05	0.000124	0.908	1.161	0.908	1.161
X Variable 1 (qe)	-0.111	0.014	-8.01	0.004063	-0.16	-0.07	-0.16	-0.07
Line5								
Intercept	-0.084	0.016	-5.24	0.013536	-0.14	-0.03	-0.14	-0.03
X Variable 1 (1/qe)	0.983	0.016	63.3	8.66E-06	0.934	1.033	0.934	1.033

The Data Analysis Toolpak also offer the Residual Plot, Line Fit Plot, and Normal Probability Plot. The Residual Plots of these 5 linear types were presented in figure 1a-1e. The Line Fit Plots of these 5 linear types were presented in figure 2a-2e.

In the Residual scatter Plots, each point is one experimental data point where the accuracy of the prediction is on the y-axis. The values of y-axis indicate how bad the prediction was for that value. In fig 1a, the scatter plot displays a fairly random pattern. From the fig 1a, we can find the first residual is positive, the second is zero, the next two point are negative, and the last residual is positive. This random pattern shows that a linear type 1 offers a decent fit to the experimental data. In fig 1b, the first and second residuals are positive, the next two point are negative, and the last residual is positive. The scatter plot shows a fairly random pattern, it shows that a linear type 2 offers a decent fit to the experimental data. In fig 1c, the first residual is negative, the next three points are positive, and the final point is negative. The scatter plot shows a fairly random pattern, it shows that a linear type 3 offers a decent fit to the experimental data. In fig 1d, the first residual is negative, the next three points are positive, and the final residual is negative. The scatter plot shows a fairly random pattern, it shows that a linear type 4 offers a decent fit to the experimental data. In fig 1e, the first and second residuals are negative, the next two points are positive, and the final residual is negative. The scatter plot shows a fairly random pattern, it shows that a linear type 5 offers a decent fit to the experimental data. The fig 1a-1e, the scatter plots of residuals shows a fairly random pattern, suggesting that the relationship between the independent variables (x) and the transformed dependents variable (y) is linear. These figures suggest that the transformation of five linear types to achieve linearity were successful. Also, the residual distribution pattern of the linear type 2 and 5 were smaller than the other three linear types, it indicated that the transformed data of linear type 2 and 5 resulted in a better model.

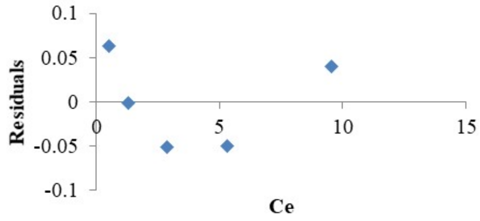


Figure 1a. Ce Residual Plot

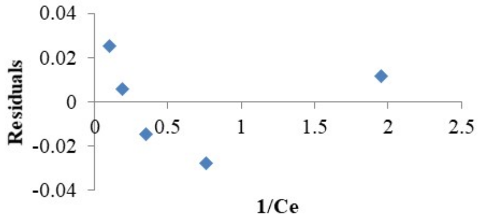


Figure 1b. 1/Ce Residual Plot

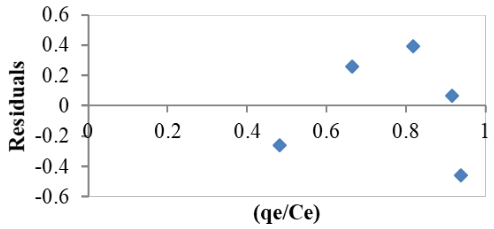


Figure 1c. (qe/Ce) Residual Plot

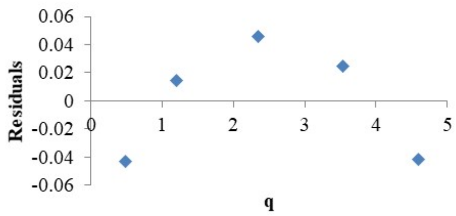


Figure 1d. q Residual Plot

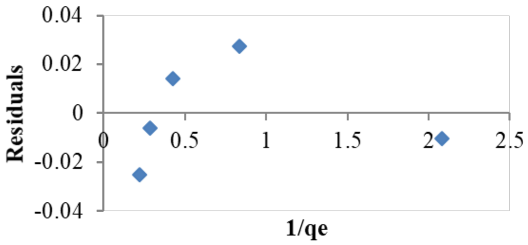


Figure 1e. 1/qe Residual Plot

The fig 2a-e were the graphs of the predicted values superimposed on the experimental data points, which will display how well the calculated values of these five different linear regression equation fits the experimental data. The predicted values and experimental data points of overlap were very high in fig 2a-e, suggesting that the transformation of five linear types to achieve linearity were successful.

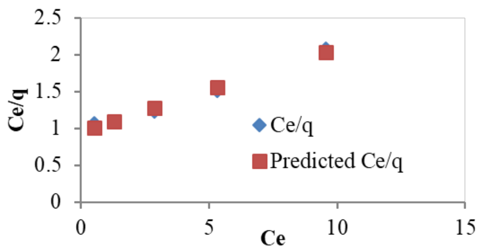


Figure 2a. Ce Line Fit Plot

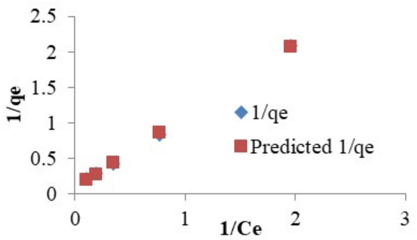
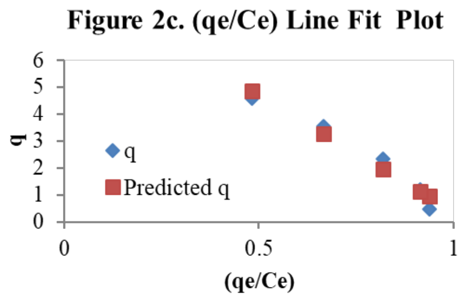
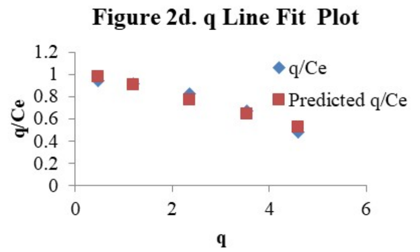
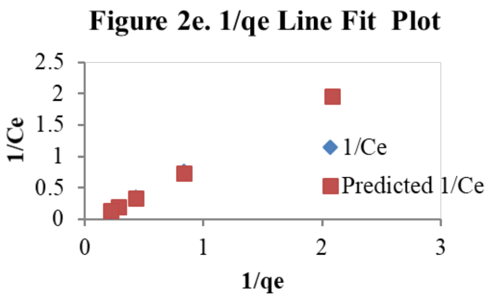


Figure 2b. 1/Ce Line Fit Plot







From the previous research, the comparison between the experimental points and predicted values for the adsorption of MB onto RH by these 5 linear type of Langmuir isotherm equations indicated that the type 1 linear expressions of Langmuir isotherm equation can predict the experimental data very well {Huang, 2016 #142}. Although the type 2 and type5 linear expressions of Langmuir isotherm equation failed to predict the q_e values and C_e values for final point of the adsorption of MB onto RH. All these results indicated that the good predictive values are still available for most of the experimental data of these 5 linear types.

Conclusion

Isotherm is applying to predict and explicit the inert characters of biosorption of MB by RH. The experimental data have been modelled using Langmuir equations, which is a non-linear equation. The complexity of nonlinear equations increases the difficulty of modeling. In this context, linearization of non-linear modelling is a powerful technique used to fit the experimental data to the non-linear model. The invention of computers has made the modelling of data quick, reliable and easier for the users. This paper try to analyze the results obtained from 5 different linearization types of Langimur to find the suitable linear type for users with basic knowledge of excel and do not need a massive understanding of the mathematics behind the processes. The purpose of this research is to use Statistical Analysis with Excel to analyze the effect of linear transformation on the Langmuir isotherm equations. The function of data analysis toolpak of excel were applied to analyze the different linear types. From the *Regression Statistics* tables and ANOVA table, the output obtained from the data analysis toolpak by using these five linear forms of Langmuir equations indicate that the best-fit linear types of the same Langmuir isotherm equation were linear type 2 and type 5. The predicted q_e values and C_e values for the adsorption of MB onto RH by these 5 different linear expressions of Langmuir isotherm equations indicated that type 1 expressions provide reliable evaluates {Huang, 2016 #142}. However, the last point of predicted q_e values and C_e values of type 2 and type 5 expression of Langmuir isotherm equations were found to be the worse fit data of Langmuir isotherm equations. In fact, the good predictive values are still available for most of the experimental data of these 5 linear types. Compared to the computational complexity of the nonlinear form, linearization of nonlinear equations simplifies computational complexity by using the spreadsheet software. From the results, the data analysis toolpak can help researcher to judge goodness of fit from calculated estimates.

References:

Basker, A., P. S. Shabudeen, P. V. Kumar and A. Shekhar (2014). "SEQUESTRATION OF BASIC DYE FROM TEXTILE INDUSTRY WASTE WATER USING AGRO-WASTES AND MODELING WITH ANOVA."

Begum, S. A., A. Golam Hyder and N. Vahdat (2016). "Adsorption isotherm and kinetic studies of As (V) removal from aqueous solution using cattle bone char." Journal of Water Supply: Research and Technology-Aqua 65(3): 244-252.

Chowdhury, S. and P. Das Saha (2011). "Comparative analysis of linear and nonlinear methods of estimating the pseudo-second-order kinetic parameters for sorption of Malachite green onto pretreated rice husk." Bioremediation Journal 15(4): 181-188.

Djeribi, R. and O. Hamdaoui (2008). "Sorption of copper (II) from aqueous solutions by cedar sawdust and crushed brick." Desalination 225(1-3): 95-112.

Ebrahimian, A., E. Saberikhah, M. Emami and M. Sotudeh (2014). "Study of Biosorption Parameters: Isotherm, Kinetics and Thermodynamics of Basic Blue 9 Biosorption onto Foumanat Tea Waste." Cellulose Chemistry Technology 48: 735-743.

Gimbert, F., N. Morin-Crini, F. Renault, P.-M. Badot and G. Crini (2008). "Adsorption isotherm models for dye removal by cationized starch-based material in a single component system: error analysis." Journal of Hazardous Materials 157(1): 34-46.

Hamdaoui, O. and E. Naffrechoux (2007). "Modeling of adsorption isotherms of phenol and chlorophenols onto granular activated carbon Part II. Models with more than two parameters." Journal of Hazardous Materials 147(1-2): 401-411.

Hamdaoui, O. and E. Naffrechoux (2007). "Modeling of adsorption isotherms of phenol and chlorophenols onto granular activated carbon: Part I. Two-parameter models and equations allowing determination of thermodynamic parameters." Journal of Hazardous materials 147(1): 381-394.

Ho, Y.-S. (2006). "Isotherms for the sorption of lead onto peat: comparison of linear and non-linear methods." Polish Journal of Environmental Studies 15(1): 81-86.

Hossain, M., H. Ngo and W. Guo (2013). "Introductory of Microsoft Excel SOLVER function-spreadsheet method for isotherm and kinetics modelling of metals biosorption in water and wastewater." Journal of Water Sustainability.

Huang, Y.-T. and M.-C. Shih (2014). "Linear regression analysis for the adsorption kinetics of methylene blue onto acid treated rice husk."

Huang, Y.-T. and M.-C. Shih (2016). "Effect of linearized expressions of Langmuir equations on the prediction of the adsorption of methylene blue on rice husk." source: International Journal of Scientific and Research Publications 6(4): 549-554.

Karadag, D., Y. Koc, M. Turan and M. Ozturk (2007). "A comparative study of linear and non-linear regression analysis for ammonium exchange by clinoptilolite zeolite." Journal of Hazardous Materials 144(1-2): 432-437.

Kaur, H. and R. Kaur (2014). "Removal of Rhodamine-B dye from aqueous solution onto Pigeon Dropping: Adsorption, Kinetic, Equilibrium and Thermodynamic studies." J Mater Environ Sci 5(6): 1830-1838.

Khan, A. R., M. R. Riazi and Y. A. Al-Roomi (2000). "A thermodynamic model for liquid adsorption isotherms." Separation and purification technology 18(3): 237-250.

Kundu, S. and A. Gupta (2006). "Arsenic adsorption onto iron oxide-coated cement (IOCC): regression analysis of equilibrium data with several isotherm models and their optimization." Chemical Engineering Journal 122(1): 93-106.

Lyubchik, S., A. Lyubchik, O. Lygina, S. Lyubchik and I. Fonseca (2011). Comparison of the thermodynamic parameters estimation for the adsorption process of the metals from liquid phase on activated carbons. Thermodynamics-Interaction Studies-Solids, Liquids and Gases, InTech.

Ncibi, M. C. (2008). "Applicability of some statistical tools to predict optimum adsorption isotherm after linear and non-linear regression analysis." Journal of Hazardous Materials 153(1-2): 207-212.

Ryu, Y. K., S. J. Lee, J. W. Kim and C.-H. Leef (2001). "Adsorption equilibrium and kinetics of H 2 O on zeolite 13X." Korean Journal of Chemical Engineering 18(4): 525-530.

Sakr, F., A. Sennaoui, M. Elouardi, M. Tamimi and A. Assabbane (2015). "Étude de l'adsorption du Bleu de Méthylène sur un biomatériau à base de Cactus (Adsorption study of Methylene Blue on biomaterial using cactus)." Journal Materials Environment Sciences 6: 397-406.

Sharififard, H., E. Rezvanpanah and S. H. Rad (2018). "A novel natural chitosan/activated carbon/iron bio-nanocomposite: Sonochemical synthesis, characterization, and application for cadmium removal in batch and continuous adsorption process." Bioresource technology 270: 562-569.

Talbi, M., M. Kabbaj, O. Belghazi and T. Ainane (2016). "A statistical approach based on the full factorial experiment for optimization of dyes adsorption on biomaterials prepared from mint and tea." J. Mater. Environ. Sci. 7(4): 1379-1385.

Ullah, H., M. Nafees, F. Iqbal, S. Awan, A. Shah and A. Waseem (2017). "Adsorption kinetics of Malachite green and Methylene blue from aqueous solutions using surfactant-modified Organoclays." Acta Chimica Slovenica 64(2): 449-460.

ACKNOWLEDGEMENT

The authors acknowledge the effort of all students and graduates of Bioprocess Engineering and Fermentation Lab in Department of Biological Science and Technology, I-Shou University for their experiment operation.

AUTHORS

First Author –Yu-Ting Huang, assistant professor, Department of Nutrition, I-Shou University. No.8, Yida Rd., Jiaosu Village, Yanchao District, Kaohsiung City 82445, Taiwan, R.O.C., email address: ythuang@isu.edu.tw.

Second Author–Li-Chiun Lee, assistant professor, Department of Nutrition, I-Shou University. No.8, Yida Rd., Jiaosu Village, Yanchao District, Kaohsiung City 82445, Taiwan, R.O.C..

Third Author–Ming-Cheng Shih, assistant professor, Department of Biological Science & Technology, I-Shou University. No.8, Yida Rd., Jiaosu Village, Yanchao District, Kaohsiung City 82445, Taiwan, R.O.C., email address: mchshih@isu.edu.tw.

Fourth Author–Wei-Ting Huang, Department of Biological Science & Technology, I-Shou University. No.8, Yida Rd., Jiaosu Village, Yanchao District, Kaohsiung City 82445, Taiwan, R.O.C., email address: c960099@gmail.com.

Correspondence Author –Ming-Cheng Shih, email address: mchshih@isu.edu.tw, alternate email address: mchshih@gmail.com, Tel: +886-7-6151100 ext.7321, Fax: +886-7-6577056, Present address: Department of Biological Science & Technology, I-Shou University. No.8, Yida Rd., Jiaosu Village, Yanchao District, Kaohsiung City 82445, Taiwan, R.O.C.