Marine green algae *Codium iyengarii* as a good bio-sorbent for elimination of reactive black 5 from aqueous solution

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Abstract: The green seaweeds *Codium iyengarii* (*C. iyengarii*) was used to prepare as an adsorbent surface for the deletion of Reactive Black 5 (RB 5) from aqueous solution via adsorption. The batch technique was adopted under the optimal condition of amount of adsorbent, agitation time, concentration of dye, and at neutral and low pH. The depletion in concentration of the dye was monitored by Schimadzo 180 AUV/Visible spectrophotometer. It was initially monolayer adsorption, which showed multilayered formation later on with the passage of time at low and neutral pH. The Results displayed that adsorptive ability of *C. iyengarii* was 1.95-3.82mg/g with an elevation in primary application of dye contents (50ppm-70 ppm). The elimination data were well stable into the Langmuir and Freundlich adsorption isotherm equations. The Langmuir (R^2=0.9848) and Freundlich (R^2=0.9441) constants for biosorption of RB 5 on green algae were determined. The coefficient relation values suggested that the Langmuir isotherm was well fitted. It explained the interaction of surface molecules, which helps in well organization of dye molecules in a monolayer formation initially on algal biomass. The pseudo first and second order rate equations were applied to link the investigational statistics and found that the second order rate expression was found to be more suitable for both the models. The absorption spectrum of RB 5 before and after adsorption with respect to time was monitored which clearly indicate that *C. iyengarii* was much effective surface at very low quantity.

Keywords: Reactive Black b, green algae, biosorption**,** isotherm

INTRODUCTION

Biosorption practices have devoted a pronounced significance from ecological safety measurements because they are useful in elimination of harmful composites from manufacturing effluents. A number of industries commonly used dyes to color their finishing goods and discharge remaining colored water into natural running streams that causes severe damages to marine organism besides this it also destructed the symbolic fauna of ecosystem. Dye textile industry requires a massive quantity of water for the cleaning and washing purposes (Robinson *et al*., 2001) and discharge highly colored wastewater containing complex dyes effluent. Moreover, the dyes are deliberately design in such way to persist and showed resistance in degradation. There fore these waste waters are challenging to scientist who are engaged in removing these colorants via predictable physico-chemical as well as biological methods due to their composite and stable structure of the dyes (Azmat & Saleem, 2011; Azmat, *et al*., 2011, Uddin *et al*., 2012). Consequently, advancement in adsorption methods offers an attractive alternate for the management of colored waters (Lee *et al*., 1996). Several researches used numerous adsorbent like sewage sludge and peanut shell, activated carbon, silk cotton hull, naturally abundant biomass:, coconut tree saw dust and coir pith, moss, banana pith and water hyacinth roots, parthenium plant, bacteria and fungi (Graham, *et al*., 2001; Kadirvelu *et al* 2003,Low *et al*., 1995; Rajeshwari & Subburam, 2002; Fu & Viraraghavan,2001, McKay, 1996 ; Schiewer & Volesky (2000).Various studies were taken into consideration in search of low cost material which includes, saw dust, coir pith, pearl millet husk, date pits, buffing dust of leather industry, crude oil residue tropical grass, shells of olive and almond , coconut shell, wool waste, etc., as carbonaceous originators, intended for the deletion of colorants from receiving streams (Kadirvelu *et al*., 2003). Marine alga showed a great potential in green technologies, also very effective promising materials as a vital biosorbents in controlling the pollution of wastewater. They are freely accessible in bulk quantities, having a great metal linking capability, commonly due to carboxylic and sulfonate groups from the algal polysaccharides. In batch sorption studies *Sargassum muticum*, commonly used to remove dye colorant from wastewater and also describe the capacity of suitable adsorbent. (Critchley *et al* 1990, Jayaraj *et al*., 2011,. Al-Khafagy, 2012, Hasan & Ibrahim, 2011).

Detailed literature survey revealed that few reports were available in sorption studies of the dye using green seaweeds but no report was available on RB 5 sorption by green algae, *C. iyengarii*. It was abundantly available at Karachi Coast. The current research was undertaken to assess the effectiveness of locally collected biosorbent surface; prepared from marine green algae *C. iyengarii* for removal of Rb5 dye from aqueous medium. The article covered the biosorption phenomena in batch sorption processes at different operational parameters.

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Results were discussed in order to design adsorption treatment systems, knowledge of kinetic and plot of isotherm including efficiency of the surface for high mass of dye and low amount of surface.

MATERIAL AND METHOD

Collection of seaweeds for preparation of Biosorbent surface

Codium iyengarii, green seaweeds *was* selected and collected from Bullajii coastal area of Karachi Beach in the month of Feb. 2012 when it was abundantly available. The weeds were collected from free-floating sea tides and washed with sea water and transported in the test site. The seaweed samples were sucked and cleaned with tap water in order to remove any contamination and then further scrubbed with distilled water to eradicate the sand, epiphytes and saline spots. Washed algal biomass was then spread on a mat for evaporation of water molecule at room temperature for 3 d. After 3 d it was placed in an oven for complete dehydration at 50°C for 48 h. The parched green algae were then grinded in an electrical mortar and used without further treatment with any chemical for adsorption of reactive black 5.

Stock solution of RB 5 (Sigma Aldrich, CAS Number 17095-24-8, Empirical Formula (Hill Notation) $C_{26}H_{21}N_5Na_4O_{19}S_{62}$ Molecular Weight 991.82) was prepared in distilled water. Dilutions were made at the time of run. Structure formula of RB5 is showed in Fig. 1.

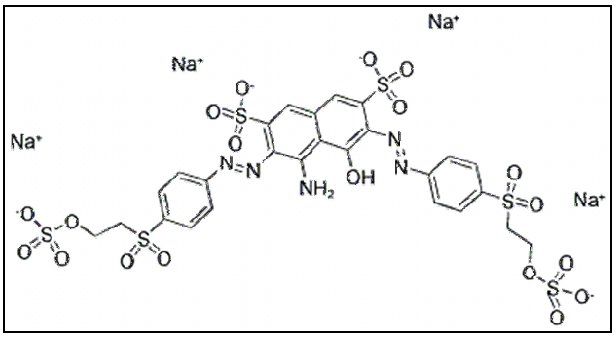


Fig. 1: structural formula of reactive black 5(RB 5)

Batch biosorption process

Adsorption of reactive black 5 was conceded in a set of experiments by stirring a static quantity of washed green algal powder with 200mL of dye solution at 120 rpm at room temperature for 3 h. At that time a minor volume of RB 5 solution was introverted at established time interval and optical density was recorded for determination of concentration of dye solution, using double beam UV/VIS 180 A spectrophotometer at λ_{max} =590. Adsorption capacity, *qe* (mg/g) and percentage (%) of dye uptake were calculated through equations, which are designate by Tan *et al* (2009)

Effect of agitation rate, pH and sorbent dose

The influence of several operational factors like quantity of sorbent was evaluated via contacting the coloring mixture through a sequences of algal biomass dose in the range of 0.1-2.0g at 120rpm, at room temperature for 2 h. Adsorption was monitored at low, neutral and high pH using acid, aqueous and alkaline medium to monitor the effects of pH on removal of dye on algae. For the estimation of the influence of agitation speed, algal biomass (0.25 g) was agitated at various agitation rates (50-250 rpm) at 30°C for 4 h.

Kinetic study and Isotherm Analysis

Langmuir (Eq. 1) and Freundlich (Eq. 2) models were tested for analysis of the dye -algal biomass bio-sorption process in a reaction mixture with variable biomass dose. Correlation coefficient values (R^2) derived for each isotherm were compared and applied to assess the association between surface and dye concentration at equilibrium.

$1/q_e = 1/Q_o + 1/bQ_o\, C_e$ ---1

$\ln q_e = \ln k + [1/n] \ln Ce$ ---2

Linearized equation of pseudo-first-order kinetic model was presented in Eq. (3)

$\log (q_e - qt) = \log q_c - K_1\, t/2.0303$-----3

Pseudo-second-order kinetic analysis was expressed as in Eq. (4)

$1/q_t = 1/ K_2 q_e^2 + 1t/q_e$ ----4

UV/Visible spectral Analysis

Spectral change was recoded after different time interval and presented in neutral and acidic medium.

RESULTS

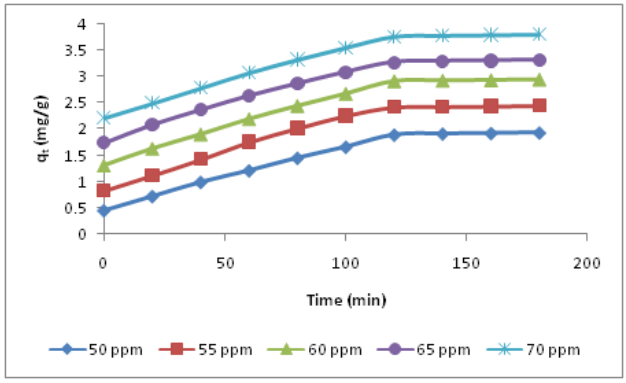
Preliminary experiments

The biosoption kinetics was monitored in batch system for controlling colorant toxicology to the safety of ecology of aquatic resources at Laboratory scale. For this purpose kinetics of the removal reactions between adsorbent and adsorbate were investigated at various operational parameters to test the suitability of low cost biosorbent, green algal biomass spectrophotometrically. Results showed the removal efficiency of the biomass with no oxidative products as in other advanced oxidation processes. The maximum removal of Rb 5 found to be 120-150 min for concentration range of (50-70ppm) at pH range of (4.5 & 7). The deletion kinetics with respect to various biomass concentration of green marine biomass (0.05-0.25g) showed that weight of biomass was effective in the range of 0.1 to0.2 gm keeping all other parameters as constant. The pH of reaction mixture was adjusted via acid and alkali in the range of 3 & 11 and found that most suitable pH was (4.5 & 7) for sorption kinetics. The obtained data was then used to analyze the best model of adsorption kinetics followed by order of reactions.

DISCUSSION

Influence of advocate time and dye concentration

The optimum time for maximum removal of RB 5 was investigated in a batch system at variable concentrations of dye dosages (50-70ppm) at neutral and low pH (4.5 & 7) keeping all other parameters as constant at room temperature. The contact time for removal of the RB 5 presented in the Fig: 2 & 3. It was clearly observed from the Fig. 2&3 that contact time necessary for adsorption of the dye to reach at equilibrium was about 150 min at pH 7 whereas at pH 4.5 it was 120 min for various concentrations of the dye (50-70ppm). It was also observed from the both the Fig. 2 & 3 that at low pH equilibrium reached more rapidly as compared to the pH 7. Moreover at this state the amount of dye adsorbed was in the state of dynamic equilibrium with the concentration of dye being biosorbed onto algal surface. It was clearly elucidated from the Fig. 2 & 3 that as the concentration of the dye increases the adsorption increases on the biosorbent surface from monolayer to multilayer to reached an equilibrium state.



Fig, 2: The variation of adsorption capacity with biosorption time at various concentration of RB 5 at 30°C (pH 4.5, W=0.2 g).

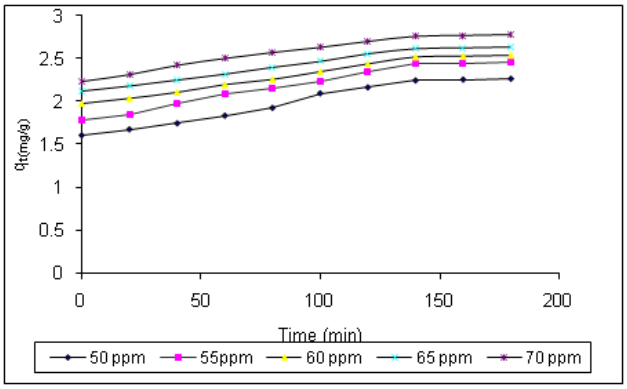


Fig. 3: The variation of adsorption capacity with biosorption time at various concentration of RB 5 at 30°C (pH 7, W = 0.2g)

The time compulsory to reached equilibrium was the equilibrium time and the amount of dye adsorbed reveals the maximum removal of the dye from the solution. This was the maximum adsorption capacity of the adsorbed dye under optimal condition. The maximum capacity of the surface (table 1) directly related with dye concentration (1.95-3.82 mg/g). These results proposed that biosorbent prepared from the green marine alga proved to be an efficient dye eliminator without producing sludge. Spectral analysis of the reaction mixture at various dye concentration before and after agitation time with fixed algal mass was shown in Fig. 4 and reflects the successful adsorption of the dye onto surface.

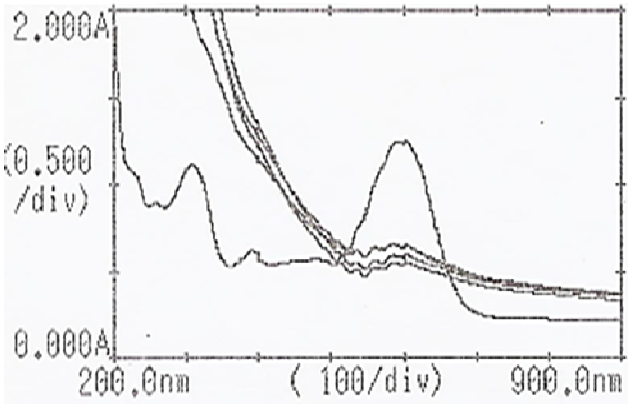


Fig. 4: Effect of dosage of sorbent on spectral change of RB 5

Effect of sorbent dosage

Effect of sorbent dosage on removal of RB 5 by *C. iyengarii* was observed through varying the quantity of seaweeds (0.05-0.25g) while keeping other variable constant. It was observed that the amount of sorbent used played an important role in removal of dye color. The sorbent dosage was effective in the range of 0.1 to 0.2g where maximum decoloration of the dye was observed. This may be due to more available surface, which contributed to an increase in the dye sorption. Consequently more dye was adsorbed. However at this point at all operational parameter *C. iyengarii* had reached the saturation point of 0.2g which considerably satisfactory for biosorption of RB5 with almost 99% of removal of the dye (Fig.5).

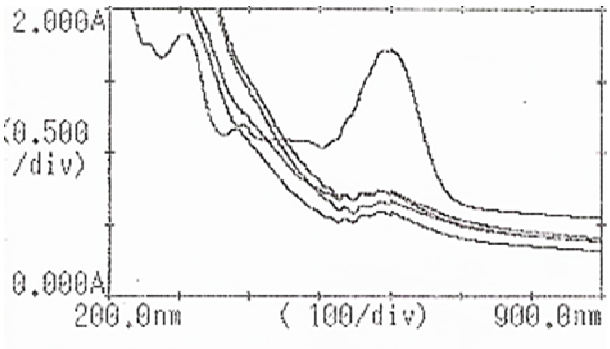


Fig. 5: Spectral change of RB 5 at various concentrations and fixed weight of sorbent

The results of current investigation were similar to those of earlier ‘s outcome who also reported that the adsorption capability of the surface was related with the available vacant surfaces on adsorbent. (Jayaraj *et al* 2011 a &b). The effect of change of sorbent dosage was presented in the Fig. 5 where it was clearly observed that sorbent dosage was effective in color removal of RB 5 (Azmat *et al*., 2013). This experiment propose that the *C. iyengarii* demonstrated to be very good effective surface at very low weight (0.2g) as compared to previous work reported in the literature (Tan *et al*., 2012). The change in initial amount of sorbent with the fixed concentration of dye on surface was presented in the Fig. 5. It is evidently pragmatic that the dye adsorbed on the surface of biosorbent and surface was effective in dye removal from the solution. The effective removal of RB 5 on algal surface may be related with the surface molecules (S, OH, NO) present, which can attract the dye molecule through interaction due to which dye molecule fixed on the surface of biosorbent.

Effect of pH

Results showed that the pH of reaction mixture played an important role for continuation of the biosorption. It was important because algae and dye contained many functional groups having ionization sites for interaction to each other. The sorption of RB 5 onto green biomass was investigated in a reaction mixture having pH range between 3 and 11.0 by addition of diluted NaOH and HCl solution ($0.1mol^{-}dm^{3}$). The initial dye concentration and the weight of algal mass were maintained at 50ppm and 0.2g respectively at $25^{o}C$ The sorption ability of biosorbent surface increased with increasing initial solution pH from 3 to 7 and then it remains constant It was maximum for 4.5 and 7 indicating that the optimum pH 4.5 and 7. The algal biomass contain significant compounds constitutes of hydroxylic and carboxylic groups. It was observed that the sorption process was highest at low pH. This may be related to that; at pH 4.5 these functional groups become neutral due to the presence of H^{+} while strong attraction with RB 5 due the amino groups of the algal compounds were established with surface. However at high pH, OH ions were in excess due which the amino groups of surface become neutral and the hydroxylic and carboxylic groups are in anionic forms provided the weak electrostatic attraction with dye due to which adsorption become constant. There were several reasons provided for justifying the biosorption of dyes by the marine algae biomass relative to pH. The superficial of the marine algae comprises of vast integral of responsive axes. At low pH range, the biosorbent surface of the algae acquires positively charged hence creating the H^{+} ions compete effectively with color molecule cations producing reduction in the quantity of dye adsorbed (mg/g). At alkaline pH, the surface of the biomass becomes negatively charged, which increases the interaction of positively charged dye cations with the surface of algae via the electrostatic forces of attraction. All experiments were carried out at neutral pH 7, for the percent uptake of dye (Fig. 2) because adsorption was become constant at this pH and equilibrium has been reached subsequently (Critchley *et al.*, 1990, Jayaraj *et al*., Al-Khafagy 2012, Hasan & Ibrahim, 2011, Qamer *et al*., 2013).

Biosorption Isotherm Analysis

The data obtained from above investigation was applied by fitted them to two well-known adsorption isotherms (Eqn. 1) model, Langumir and Freindlich isotherm (Eqn.2).

This was an essential phase to find appropriate model that can be used to explore the overall biosorption efficiency and to characterize the RB 5 sorption process in the coordination using various amount of *C. iyengarii* biomass. Langumir adsorption isotherm undertakes that adsorption occur homogenously with monolayer formation onto a surface comprising finite number of available uniform line of attack for adsorption of dye with no movement of adsorbate in the plane surface. While Frendlich isotherms an empirical model employed heterogeneous surface in which the energy term in Langumir equation varies as a function of the surface coverage (Kannan *et al.*, 2010). The applicability of both isotherm equations was compared by linear equation correction coefficient R^2 (figs. 6 & 7).

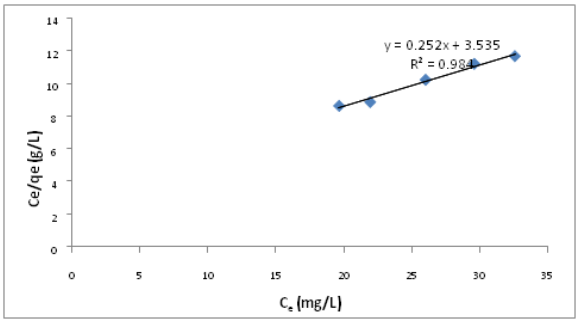


Fig. 6: Langmuir adsorption isotherm for the biosorption of RB5 on *C. iyengarii* (30°C sorbent dosage effect).

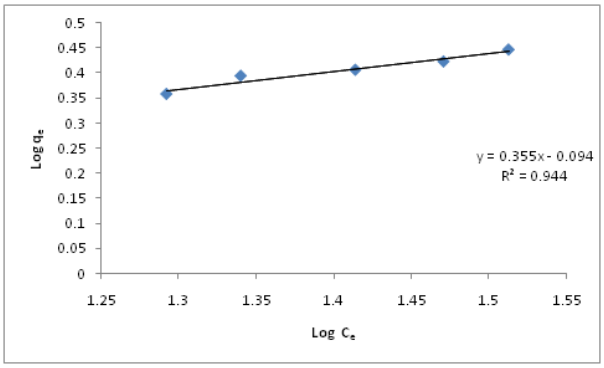


Fig. 7: Freundlich adsorption isotherm for the biosorption of reactive black b on *Codiumiyengarii* (at 30°C sorbent dosage effect).

Table 1: Comparison of the pseudo first and second order adsorption rate constants and calculated experimental q_e values for different initial concentration of reactive black 5 at pH 4.5

RB 5 (mg/L)	Q_{eexp} (mg/g)	Second order kinetic model							
		k (min)	Q_{ecal} (mg/g)	R^2	SSE (%)	k_2 (g/mg/min)	Q_{ecal} (mg/g)	R^2	SSE (%)
50	1.95	2.48	-0.026	0.943	0.16	0.436	15.39	0.936	4.2
55	2.46	2.55	-0.027	0.955	0.028	0.363	8.47	0.970	1.9
60	2.96	3.09	-0.031	0.936	0.041	0.310	5.51	0.981	0.81
65	3.35	2.31	-0.025	0.956	0.33	0.282	3.64	0.990	0.09
70	3.82	2.64	-0.027	0.949	0.37	0.248	2.91	0.991	0.28

Table 2: Comparison of the pseudo first and second order adsorption rate constants and calculated experimental q_e values for different initial concentration of reactive black 5 at pH 7

RBb (mg/L)	Q_{eexp} (mg/g)	Second order kinetic model							
		k_1 (min^{-1})	Q_{ecal} (mg/g)	R^2	SSE (%)	k_2 (g/mg/min)	Q_{ecal} (mg/g)	R^2	SSE (%)
50	2.28	0.0213	1.159	0.927	0.354	0.425	4.3433	0.9921	0.58
55	2.48	0.0211	1.155	0.916	0.419	0.394	3.4513	0.9942	0.66
60	2.55	0.0231	1.065	0.901	0.469	0.384	2.7075	0.996	0.68
65	2.65	0.0196	1.176	0.933	0.466	0.372	2.24	0.9972	0.72
70	2.8	0.0196	1.224	0.953	0.498	0.353	1.811	0.9981	0.77

Table 3: Langumir and Freundlich adsorption isotherm constant for biosorption of Reactive Black 5 (sorbent dosage effect)

Langumir Adsorption constant	Present	Literature Reported
Q_o	0.0.2526	43.29 Tan *et al* (2010)
B	3.5353	1.974
R^2	0.9841	0.777
Frendlich Isotherm constant		
n	0.3557	1.9868
K (mg/g) $(1/mg)^{1/n}$	0.0942	32.46
R^2	0.9441	0.996

By comparing the constant of both isotherm models, it was proved that both isotherm were well fitted and explained that initially monolayer formation takes place that later on at high dosage of the dye converted into the multilayer formation. It was suggested that *C. iyengarii* involves random distribution of vacant or reaction sites which occupy by more than one molecule. The effect of increase in sorbent dosage, the adsorption mechanism complied well with the Freundlich model (R^2=0.9441). It was established that the biosorption of RB 5 may be attributed to the functional groups of the algal surface which made up of cellulose via the multiple layer process. Value of an important Freundlich constant (n) is 0.3557, signifying a promising adsorption process. The determined Langmuir (R^2=0.9848) and Freundlich (R^2=0.9441) constants for biosorption of RB 5 on green algae suggested that the Langmuir isotherm was well fitted to explain the interaction of surface molecules which helps in well organization of dye molecules in a monolayer formation on biosorbent surface. The results of current results emphasized the viability and credibility of marine biomass as a low cost biosorbent in handling colorant sewage.

Expending the thickness models to examine porosity

Langumir thickness equation was used to explain seaweeds bio- sorbent as non-porous solids. It was observed that a direct relation exists between thickness and specific concentration of dye adsorbed (C_e/m) (Tan *et al* 2012). It was concluded that thickness equations predict C_e at various concentration for seaweeds materials. Plotting experimental data against thickness equation data yields a straight line for nonporous solids which reflect that biomaterial of seaweeds act as a good porous surface and also showed the capability of seaweeds to remove dye contaminate up to 70ppm (fig. 2 & 3). It was also observed that at pH 4.5 monolayer adsorption reached maximum value and poly layer

adsorption started and a deviated line for porous solids. The thickness of the adsorbed dye (fig. 3 pH 7) microspores volume of the surface is dominated whereas the size of microspores gets reduced 4.5. It was suggested that the thickness of the adsorbed dye at pH 7 was large as compared to at pH 4.5. It was clearly indicated from the figs. 2 & 3 that at pH 7 thickness of the layer was large as compared to low one that may be attributed to that biosorption was good enough at neutral pH (tables 1-3).

Biosorption kinetics Analysis

The kinetics of biosrption was studied using eqn.3 & 4 and tabulated into tables 1-3. The values for k_1 were calculated and reported in the table 3. Results tabulated in the Table 3showed that if value of R^2 were higher than 0.90 q_e agrees with the calculated one (table 3) obtained from three linear plots (figs. 6 & 7). This showed first order kinetics for biosorption of RB5 while from Fig.8, the plot of t/q vs t showed a linear relationship and settlement among experimental and calculated q_e values and the value of R^2 for the second order is about 0.99 indicating that biosorption of RB 5 onto green seaweeds followed second order kinetics (fig. 8).

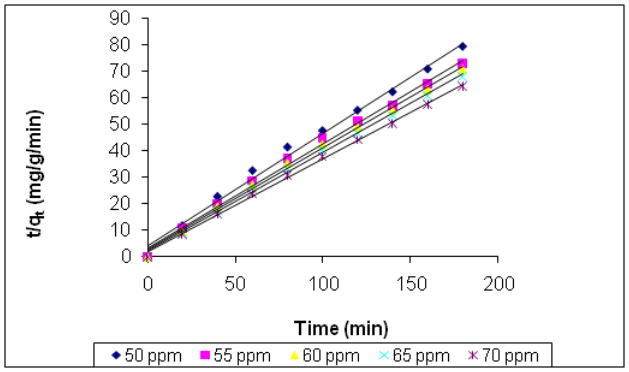


Fig. 8: Pseudo-second order kinetics for the biosorption of RB 5 on *C. iyengarii* at 30°C

This was established that pseudo second order assumed that biosorption of RB 5 was organized by chemical adsorption or chemisorption which comprises valiancy forces over interchange or reaction among the adsorbent surface OH group which help in adsorption of dye on the surface for de-coloration of solution.

CONCLUSION

The investigation about low cost surfaces for deletion of colorant from running streams suggested that the adsorption equilibrium achieved within less than 2h with constant stirring. Biosorption was directly related with amount of surface, agitation time and dye concentration. Very cost effective surface (0.2g) when compared with the reports available on marine algae for adsorption. It was proposed that it is promoting adsorbent for wastewater treatment

The results of isothermal study using *C. iyengarii* also showed that the experimental adsorption data is well fitted with Langmuir model.

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