ARTIFICIAL NEURAL NETWORK MODEL FOR BIOSORPTION OF METHYLENE BLUE BY DEAD LEAVES OF *POSIDONIA OCEANICA* (L.) DELILE

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Abstract: In the present study, an alternative promising evaluation method was recommended for dead leaves of *Posidonia oceanica* (L.) Delile as an adsorbent for biosorption of Methylene Blue (MB). The data from batch experiments were modeled by using Artificial Neural Network (ANN). The optimal operation conditions for biosorption of MB by *P. oceanica* dead leaves were found for pH, adsorbent dosage, temperature and initial dye concentration as 6, 0.3 g, 303 K and 50 mg/L, respectively. The adsorption reached equilibrium after 30 minutes. According to the results of sensitivity analysis, relative importance of temperature, dye concentration, pH, adsorbent dosage and process time on the biosorption of MB were 33%, 27%, 21%, 10% and 8%, respectively. Minimum mean square error (MSE) was found as 0.0169 by ANN modeling. The present study reveals a novel strategy for adsorption studies to utilize the highly accumulated biomass of dead leaves of *P. oceanica* in Turkish coastlines instead of burning these dead leaves.

Key words: *Biosorption, modeling, artificial neural network, Posidonia oceanica*

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1. Introduction

In many third-world countries, industrial wastewaters including heavy metals, textile dyes etc. are directly discharged into the aquatic environments such as sea,

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lake and river. These pollutants are resistant against sun and temperature and also are harmful for human health through food chain [1]. Many wastewater treatment methods applied for removal of resistant pollutants are based on adsorption, filtration, oxidation, and sedimentation, etc. [2]. Among these treatment methods the adsorption of pollutants has advantages in view of their application easiness. Moreover, adsorption of various pollutants by using waste biomasses such as seaweeds [3], orange peel [4], rice husk [5] and sugar cane dust [6] provides an evaluation method for utilization of waste biomasses. In addition, it prevents environmental pollution by using these biomasses and decreases the cost of wastewater treatment [7]. Methylene blue (MB) is one of the most commonly used dyes in some important industries such as paper, dye, cotton, wool and microbiology [8]. Moreover, MB is accepted as a model dye for adsorption studies [7-9].

Turkey has long coastlines on the Mediterranean Sea, the Black Sea and the Aegean Sea. These coastlines provide various habitats for the proliferation of sea grasses and seaweeds. Leaves of sea plants such as *Posidonia oceanica, Cymodocea nodosa*, *Zostera noltii* and *Z. marina* are decomposed before winter season and dead biomasses of them are accumulated on the beaches (Photo 1). Unfortunately, waste biomasses of sea grasses and seaweeds have not yet been utilized economically in Turkey, some of which are burned on the beaches. The total primary production of only *P. oceanica* has been estimated in the range of $5x10^6$-$5x10^7$ ton per year in the Mediterranean region [10, 11]. The evaluation of *P. oceanica* as an adsorbent material was first proposed by Ncibi, Mahjoub, and Seffen [12]. According to Ncibi, Mahjoub, and Seffen's [12] study, leaf sheaths of *P. oceanica* were used for MB adsorption. However, in Turkish coastlines the accumulation of dead leaves on the beaches has been observed commonly. Therefore, in the present study, it was aimed to test the adsorption capacity of this part of *P. oceanica* to provide further information to the scientific literature. There are many adsorption studies in scientific literature where traditional methods such as kinetic, isotherm and thermodynamic models have been used. However, in the present work, we preferred to use Artificial Neural Network (ANN) to model the adsorption process. The reason is that ANN is a nonlinear, powerful computational tool which is highly capable of solving classification, modeling and association problems by simulating the basic functions of the mammalian brain [13, 14]. There are two phases in utilizing the neural network in a real-world problem: The first phase is called training/learning in which the connection strengths among the neurons, called weights, are tuned to model complex relationships between inputs and outputs. The second phase is testing phase in which the weights determined in the learning phase are used to produce approximately correct results for new input values that are not used in the training phase.

In recent years, use of modeling approaches in adsorption studies has become popular because of their robustness, reliability and speed. In addition, with modeling by ANN, it is possible to predict the behavior of the system and design the components for control and fault detection of adsorption process successfully [15-17]. On the other hand, traditional adsorption models (kinetics, isotherms, thermodynamics, etc.) give specific information about the system and it is impossible to explain the entire behavior of the system by using only one traditional model. Examples of the use of ANN to model batch adsorption processes can be

Photo 1 *Posidonia oceanica (L.) Delile meadows from Turkish coastline (Dikili).*

found in the literature such as individual and binary sorption of cadmium and zinc ions by *Sargassum filipendula* [18], removal of acid orange 7 by powdered activated carbon [19], adsorption of Pb(II) ions by Antep pistachio (*Pistacia vera* L.) shells [20], biosorption of a triphenylmethane dye solution (Basic Green 4) by *Chlamydomonas* sp. [21], removal of chlorophenol by coconut fiber carbon [22], biotreatment of a triphenylmethane dye solution (Malachite Green) by *Vaucheria* sp. [23], competitive adsorption of phenol and resorcinol by using carbonaceous adsorbents (activated carbon, wood charcoal and rice husk ash) [24], biotreatment of a triphenylmethane dye solution (Malachite Green) by *Cladophora* sp. [25], and removal of Lanaset Red G by *Chara contraria* [26].

In our previous study, we have modeled the biosorption of MB by *P. oceanica* in a dynamic fixed-bed column system [27]. Therefore, in this study, we aimed to model the characteristic parameters of the biosorption of MB by using the same *P. oceanica* (L.) Delile material in batch system via ANN.

2. Methods

2.1 Sorbent preparation and characterization

Dead leaves of *P. oceanica* (L.) Delile were collected from coasts of Izmir in January 2009 (Photo 1). Dead biomasses were placed into plastic bags and transported to the laboratory. The dead leaves were washed with tap water to remove salt and then all impurities, and also epiphytic organisms were removed [11]. Finally the material was washed with distilled water. The dead leaves were dried at 70° C for 16 h. The dried materials were grounded by a crusher machine. Powdered *P. oceanica* dead leaves (PPO) with 500 μm particle diameter were used in the experiments. Functional groups on the dead leaves of *P. oceanica* were determined with Fourier Transform Infrared (FT-IR) spectrometer (Perkin–Elmer, Spectrum BX) [28].

2.2 Preparation of dye solution

MB was used as a model dye in the experiments. The stock solution with 1000 mg/L concentration of MB was prepared. Then dye solutions at several concentrations were prepared by diluting stock solution (20, 50, 75, 80 and 100 mg/L). All dye solutions were prepared with distilled water (pH=6.3).

2.3 Adsorption experiments

In order to determine the effects of different operation conditions, the experiments were performed by differing the initial pH (3.0 to 9.0), adsorbent dosage (0.05 to 1.00 g), temperature (293, 303, 313 and 323 K), contact time and initial dye concentration. In initial experiments, adsorbent dosage was selected as 0.3 g and polyethylene vessels were filled with 30 mL dye solution. In order to adjust the initial pH, 0.1 N NaOH and 0.1 N HCl solutions were used. Initial dye concentration and temperature were selected as 50 mg/L and 303 K, respectively, for determining the effect of initial pH. Initial dye concentration and volume of dye solution were chosen as 50 mg/L and 30 mL, respectively, for determination of the effect of adsorbent dosage. The mixtures were agitated at 100 rpm with a shaker (GFL 1092) for 3 h. In kinetic experiments, samples were taken out at preset time intervals (0-180 min) and centrifuged at 5000 rpm for 4 min. Dye concentrations in supernatants were determined at 665 nm by using Shimadzu UV-VIS 1601 spectrophotometer. The amount of dye adsorbed onto PPO at equilibrium was calculated by using the equation as shown below:

$$q_e = \frac{C_0 - C_e}{M}\, V, \tag{1}$$

where q_e is the adsorbed dye amount at equilibrium (mg/g), C_0 is initial dye concentration (mg/L), C_e is the dye concentration at equilibrium (mg/L), V is the volume of dye solution (L), and M is the amount of adsorbent (g).

2.4 Application of ANN model

In this work, ANN is used for modeling of adsorption process of MB onto PPO. ANN model is constructed on a multilayer feedforward architecture, and it is trained using the error backpropagation learning technique. Briefly, a feedforward architecture, in its basic form, has a layered structure: the input layer, one or more hidden layers and the output layer. Each neuron receives its input from the neurons in the previous layer, and each of those inputs is multiplied by its weight value. Weighted inputs are summed and passed through certain nonlinear/linear functions to produce output of each neuron, and output of first neuron is passed to the next layer as input of second neuron. Further details of dedicated neural networks of the type mentioned above are available in [13]. The schematic representation of the neural network in this study is shown in Fig. 1(a).

To reach the aimed goal, which is to learn the associations between a given set of input-output pairs, $\{(\underline{x}_1, \underline{t}_1), (\underline{x}_2, \underline{t}_2), \ldots, (\underline{x}_P, \underline{t}_P)\}$, we utilize an error backpropagation learning algorithm [14]. In this algorithm, firstly, a single input pattern $\underline{x}_p$ is presented to the network and the layers' responses $\underline{o}_p^k$, $k = 1, 2, \ldots, M-1$ are computed. Then, the error vector is determined in the output layer, and then it is propagated towards the network inputs. Finally, the weights are subsequently readjusted according to these error values. This procedure continues until certain prespecified termination criteria are satisfied.

Consider a multi-layer feedforward neural network. The net input to neuron i in layer $k+1$ is calculated as below:

$$n^{k+1}(i) = \sum_{j=1}^{S_k} w^{k+1}(i,j)a^k(j) + b^{k+1}(i). \tag{2}$$

Here, $w^{k+1}(i,j)$ is the weight correlating neurons i and j, $a^k(j)$ is the output value of neuron j in layer k, and $b^{k+1}(i)$ is the bias term of neuron i in layer $k+1$. The output of neuron i is calculated as

$$o^{k+1}(i) = f^{k+1}(n^{k+1}(i)), \tag{3}$$

where f(.) is the activation function of the corresponding neuron. During the learning process the cost function to be optimized, Sum of Squares of Errors (SSE), is defined as

$$SSE = \frac{1}{2}\sum_{p=1}^{P}(\underline{t}_p - \underline{o}_p^M)^T(\underline{t}_p - \underline{o}_p^M) = \frac{1}{2}\sum_{p=1}^{P}\underline{e}_p^T\underline{e}_p, \tag{4}$$

where P is the number of input patterns, M is the number of layers in the network, $\underline{o}_p^M$ is the actual output value and $\underline{t}_p$ is the target (desired) value of the network when p-th input pattern, $\underline{x}_p$, is presented. Clearly, $\underline{e}_p = \underline{t}_p - \underline{o}_p^M$ is the output error for the p-th input under concern. In this study, the Levenberg-Marquardt (LM) algorithm, which combines the speed of the Newton algorithm with the stability of the steepest decent algorithm, is preferred to readjust the weights. The whole process presented in an algorithmic form is as follows [29]:

1. Initialize all weights to small random values.
2. Compute the SSE for all inputs.
3. Find the derivatives of the errors with respect to the weights and form the Jacobian matrix J.
4. Update weights by computing changes in weights: $\Delta\underline{w} = \left[J^T J + \mu I\right]^{-1} J^T\underline{e}$.
5. Recompute the SSE by using the updated weights. If this new SSE is smaller than the previous one, then decrease μ by β, accept updated weights and compute SSE. If not, just increase μ by β. Continue to process by going to step 3.

6. Terminate the algorithm when SSE is smaller than a predefined value, or maximum number of cycle is reached, or μ goes beyond its predefined range.

In this study, 102 experimental data sets were used to train and test the performance of ANN for the adsorption process. Each set contains 5 input variables; namely, adsorbent dosage, initial dye concentration, temperature, initial pH and contact time, and 1 output variable, namely, the adsorption efficiency. The range of variables and their means and standard deviations are presented in Tab. I. Care must be paid that no preprocessing or feature extraction methods are applied to experimental data. The only processing is the normalization of the input values. At this point, it is important to note that it was examined whether it was useful to apply Principal Component Analysis (PCA) to reduce the dimension and to uncorrelate the components of the input. Since any principal component that contributes less than 10% to the total variation was not found, it was not preferred to use PCA as a preprocessing procedure.

Input Name	**Range**	**Mean and Standard Deviation**
Dye concentration	10 – 100 mg.L^{-1}	59.6 $\pm$ 29.0 mg.L^{-1}
Temperature	293 – 323 K	306 $\pm$ 9 K
pH	3 – 9	6.0 $\pm$ 0.5
Adsorbent dosage	0.05 – 1.0 g	0.3 $\pm$ 0.07 g
Time	5 – 180 min	75.4 $\pm$ 58.2 min
Output Name		
Adsorption	0.99 – 9.96 mg/g	5.84 $\pm$ 2.83 mg/g

Tab. I *Ranges of inputs and output variables with their means and standard deviations.*

Since it is aimed to develop an ANN model whose unknown parameters are found by optimization and fitting processes of the training data, cross-validation is applied to determine the fitness of the model parameters to a hypothetical validation set. It was preferred to use leave-one-out cross validation because of limited number of experimental data sets. In this validation method, one uses one data set for validation and all the rest for training. This procedure is applied till each data set is used once for validation.

3. Results and Discussion

3.1 Sorbent characterization

Identified functional groups on the dead leaves of *P. oceanica* were hydroxyl; O-H (3200-3600 cm^{-1}), carboxyl; COOH (2800-3000 cm^{-1}), amine; NH_2(3200-3600 cm^{-1}), C-O (1000-1150 cm^{-1}), sulfonyl; S=O (1000-1150 cm^{-1}), carbonyl; C=O (1580-1700 cm^{-1}), and S-O (500-650 cm^{-1}). Intensities of peaks revealed that the most abundant functional groups on surface of the material were carboxyl and amine. Hence, dye molecules possibly might have been bound with the –OH, –C=O and –NH groups. In previous studies from our laboratory, SEM images of raw *P. oceanica* powder were obtained. The relevant information can be found in Aydin et al [30].

3.2 ANN model and simulation data

In this section, a performance evaluation of the limitations and capabilities of multilayer feedforward networks for modeling of adsorption of MB onto PPO was provided. Network learning parameters and architectures were investigated for the determination of the best configuration.

Single and double hidden-layer neural network architectures with different hidden neuron sizes were tested to decide on the optimal configuration. Each one of architectures was tested 20 times with different initializations of the weights to take into account the probability of converging to local minima. Fig. 1(b) shows the Mean Square Error (MSE) of the validation set for different numbers of hidden neurons in a single layer ANN. According to the results, the lowest MSE is 0.0211 for 5 hidden neurons. MSE gets worse as the number of hidden neurons is increased beyond 5.

Two hidden-layer network architecture was also tested. In these numerical experiments, we kept the number of hidden neurons of the first hidden layer at 5 and varied the number of hidden neurons of the second hidden layer from 2 to 15. Results are presented in Fig. 1(c). It is clearly seen that the ANN with two hidden layers is superior to the one with single layer. The lowest MSE is obtained at 0.0169 when the number of hidden units is 8.

Experimentally determined adsorption results were well in line with predicted results estimated from neural network modeling. Data points were well distributed around y=x line and correlation coefficient was determined as 0.9982, which means ANN is good enough to predict experimental results obtained from adsorption of MB onto PPO in batch studies.

In all numerical experiments, hyperbolic tangent sigmoid (linear) functions were used for the activation functions of the neurons placed in hidden (output) layers. The parameters of the LM algorithm, μ and β, were set to 0.001 and 10, respectively.

In order to determine the relative importance or contribution of the input variables to the output variable, sensitivity analysis was applied. In the sensitivity analysis method, the partial derivatives of outputs of the network with respect to each input are calculated, and then a sensitivity matrix is formed for a set of training data [29, 31]. Results of sensitivity analysis can be seen in Fig. 1(d).

3.3 Effect of initial pH

The highest adsorption capacity (4.84 mg/g) was observed at pH 6.0 both for experimental and ANN modeling studies (Fig. 2(a)). According to Ncibi, Mahjoub, and Seffen [12], a sharp increase at pH 5.0 and no remarkable difference was observed in the MB adsorption. On the other hand, there was no strict difference in the adsorption values (4.6-4.9 mg/g) of Ncibi, Mahjoub, and Seffen's [12] study.

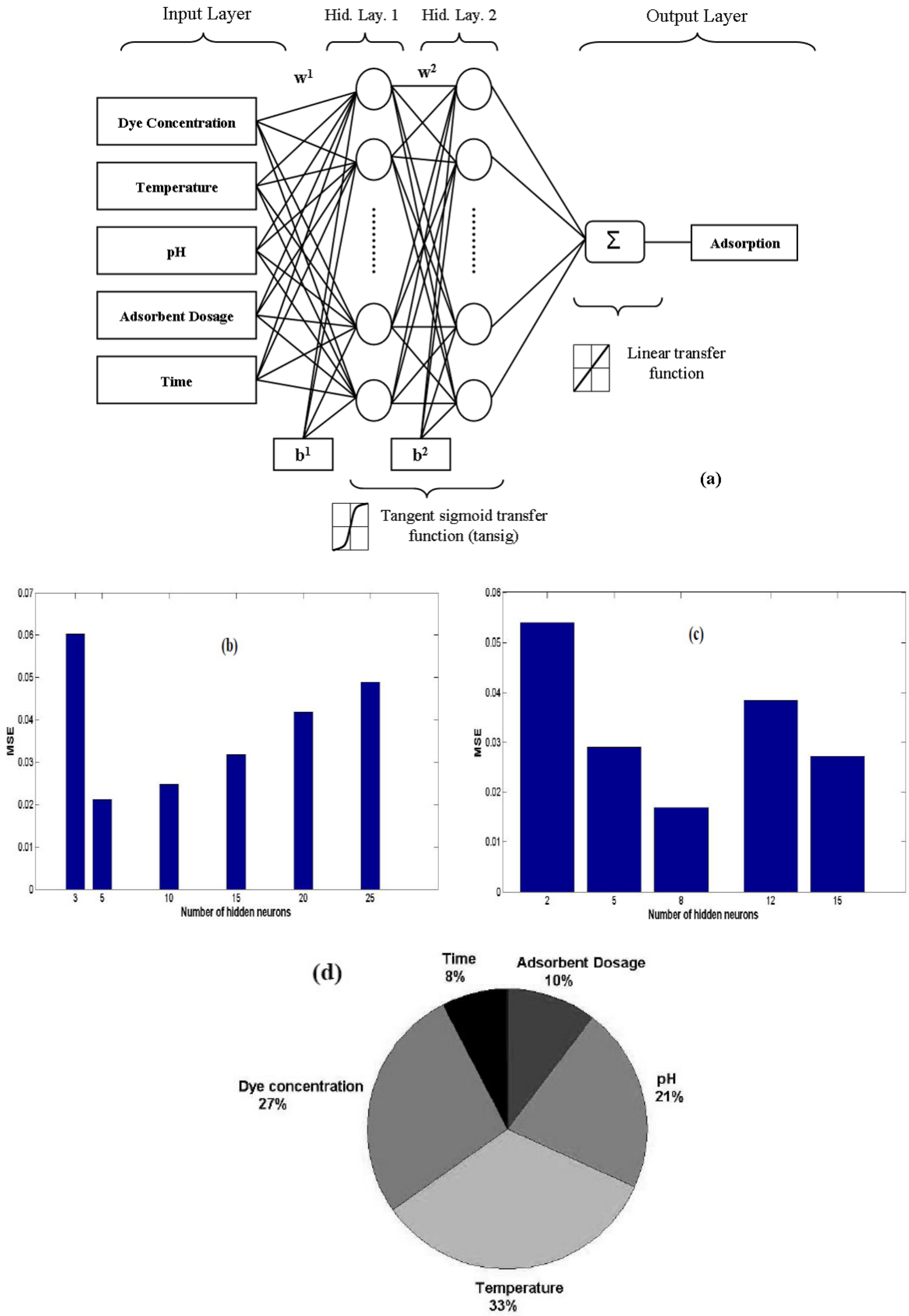


Fig. 1 *(a) Schematic representation of ANN architecture. (b) MSE for different number of hidden neurons in the single hidden layer architecture. (c) MSE for different number of hidden neurons in the second hidden layer. (d) Relative importance of different inputs.*

Statistical test was applied to the data since experiments were performed as three replicates and no statistical difference (p>0.05) between the adsorption value at pH 3.0 and pH 9.0 was found. Results of ANN modeling for effect of initial pH on adsorption of MB onto PPO were well in agreement with experimental results. Therefore, it can be said that PPO from Turkish coastlines can be used in this pH range (3.0 to 9.0) with an adsorption value as 4.8 mg/g.

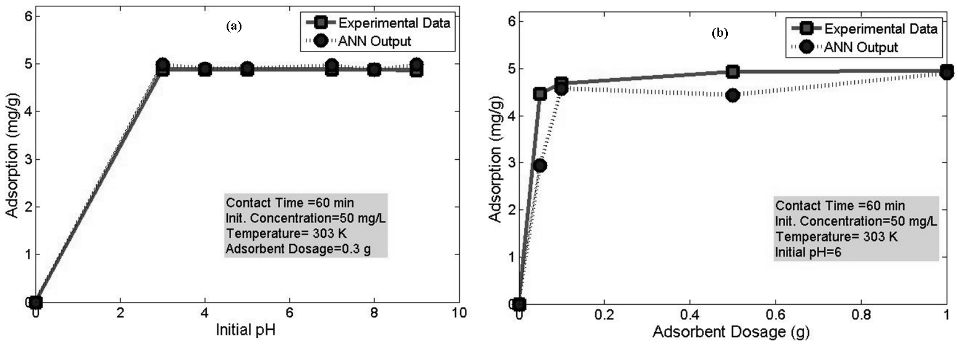


Fig. 2 *Effect of (a) initial pH, and (b) adsorbent dosage on the biosorption of MB by PPO.*

3.4 Effect of the adsorbent dosage

The effect of adsorbent dosage for dye adsorption from aqueous solution determined from batch and ANN modeling studies was shown in Fig. 2(b). The results showed that, when the adsorbent dosage increased, the removal of the MB was also increased. Similar result, increased adsorption with increasing adsorbent dosage, was reported by Ncibi, Mahjoub, and Seffen [12].

3.5 Effect of the contact time and temperature

The effect of contact time to the adsorbed dye onto PPO at various temperatures was studied (293, 303, 313 and 323 K). There were no remarkable differences among q values at all studied temperatures. Effect of temperature on adsorption efficiency was shown in Fig. 3. The results showed that after 30 minutes the adsorption between dye and PPO was reached to equilibrium in all dye concentrations. On the other hand, an agreement between ANN modeling and experimental results for the effects of temperature on adsorption of MB onto PPO can be seen in Fig. 3.

3.6 Effect of initial dye concentration

Effect of initial MB concentration on adsorption efficiency was shown in Fig. 4. According to Fig. 4, adsorption efficiency was increased with increasing initial MB concentration. Good agreement between experimental and ANN modeling results can also be seen in Fig. 4.

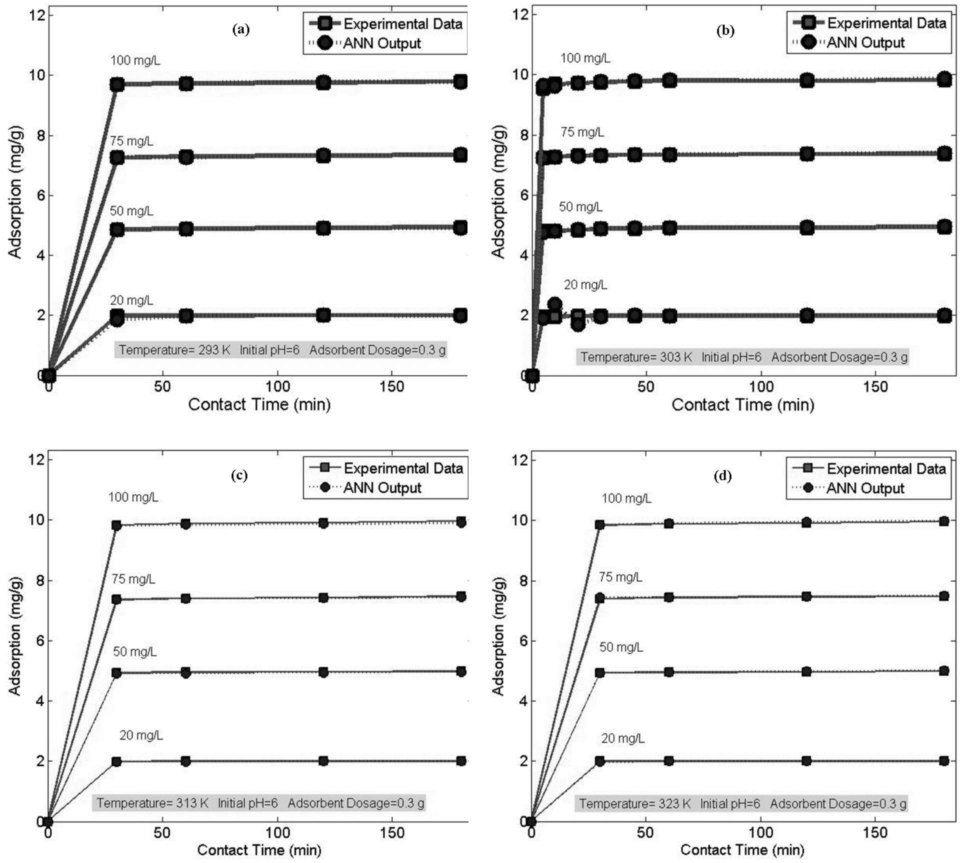


Fig. 3 *Effect of temperature on the biosorption of MB by PPO: (a) 293 K, (b) 303 K, (c) 313 K, (d) 323 K.*

3.7 Traditional evaluations

As it has been reported in vast amount of scientific publications on adsorption in the scientific literature, biosorption data generally have been evaluated by using traditional methods such as kinetics, isotherm and thermodynamics approaches. In this study, traditional approaches were also applied to the experimental data. According to the results, the adsorption capacities of *P. oceanica* dead leaves were found as 1.99, 4.94, 7.38, 7.90 and 9.83 mg/g in different initial dye concentrations of 20, 50, 75, 80 and 100 mg/L, respectively, at 303 K. The adsorption data were fitted well with the pseudo-second order model [32, 33] compared to pseudo-first order kinetics [34]. Also, the adsorption data in the present study supported the chemisorption. The rate constant at 303 K for 100 mg/L initial dye concentration was observed as 0.628 g.mg^{-1}.min^{-1}(R^2=1.00). Well known isotherm models such as Langmuir, Freundlich and Dubinin-Radusckevich were also used to model the experimental data [35-37]. According to isotherm results, the maximum biosorption

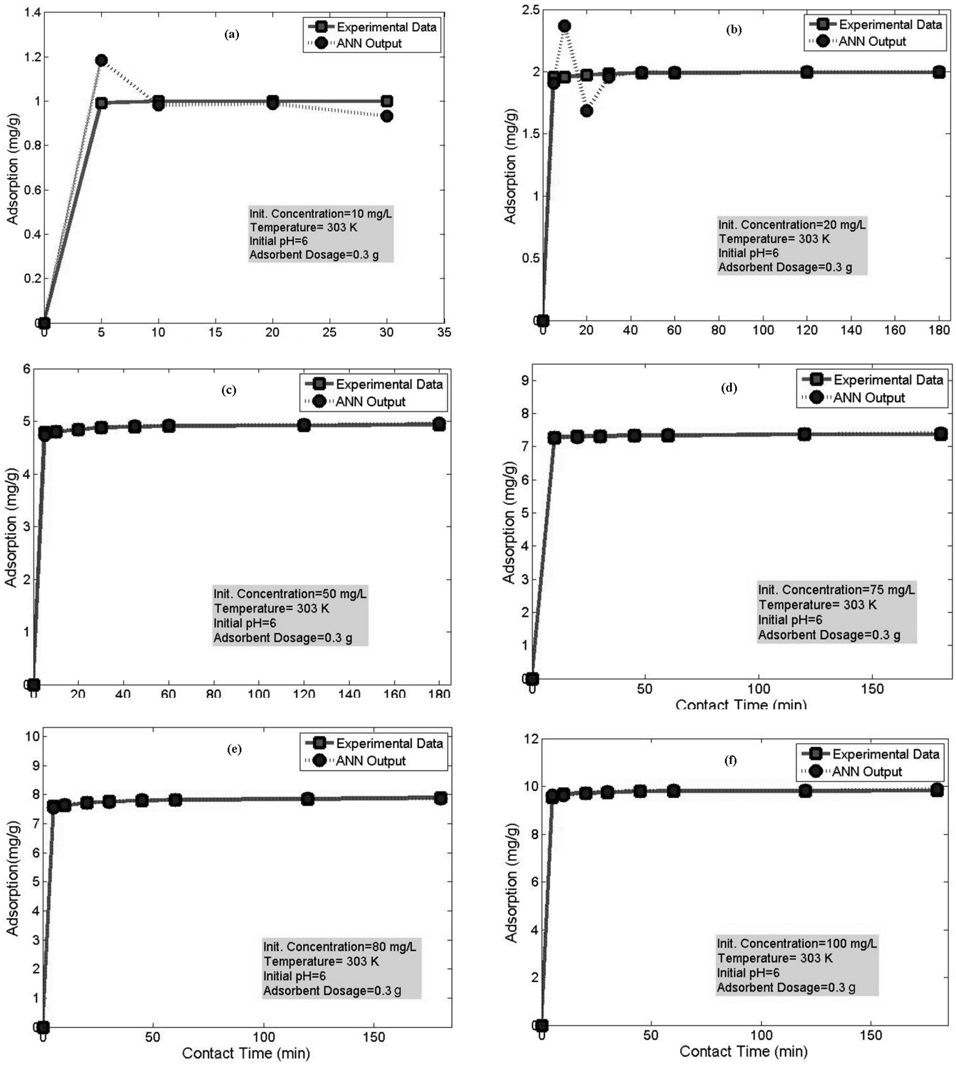


Fig. 4 *Effect of initial MB concentration on the biosorption of MB by PPO: (a) 10 mg/L, (b) 20 mg/L, (c) 50 mg/L, (d) 75 mg/L, (e) 80 mg/L, (f) 100 mg/L.*

capacities for Langmuir, Freundlich and Dubinin-Radusckevich were determined as 10.06 mg/g (313 K, R^2=0.978), 7.28 mg/g (323 K, R^2=0.978), 8.18 mg/g (313 K, R^2=0.897), respectively. According to results of thermodynamic parameters, ΔH was positive at all dye concentrations, and it indicates endothermic nature of interactions between dye and PPO. The negative values of Gibbs free energy showed that the adsorption was spontaneous at all temperatures. The results obtained with ANN method were compared with non-linear forms of traditional kinetics (Pseudo-first order kinetics and pseudo-second order kinetics) and isotherm models

(Langmuir and Freundlich isotherms) at selected conditions: 303 K and 50 mg/L initial dye concentration (Fig. 5). The results of comparison showed that ANN modeled the experimental results better than traditional models. The goodness of fit data for traditional models was given in Tab. II. ANN method models all the experimental data rather than a specific condition as applied for traditional models.

Model	**Equation**	**Parameters**	**Fitting Algorithm**	**Goodness-of-fit**
Pseudo First Order Kinetics	$q = q_e\left(1 - e^{-k_1/t}\right)$	$q_e = 4.884$, $k_1 = 0.7884$	Levenberg-Marquardt	R^2=0.9992
Pseudo Second Order Kinetics	$q = \frac{k_2\, q_e^2\, t}{1 + k_2\, q_e\, t}$	$q_e = 4.929$, $k_2 = 0.7041$	Levenberg-Marquardt	R^2=0.9999
Langmuir Isotherm	$q = \frac{q_m\, b\, C_e}{1 + b\, C_e}$	$q_m = 36.59$, $b = 0.1379$	Trust-Region	R^2=0.9507
Freundlich Isotherm	$q = K_f\, C_e^{1/n}$	$K_f = 4.762$, $n = 1.381$	Levenberg-Marquardt	R^2=0.9749

Tab. II *The goodness of fit data for non-linear traditional models used for modeling of experimental data.*

4. Conclusions

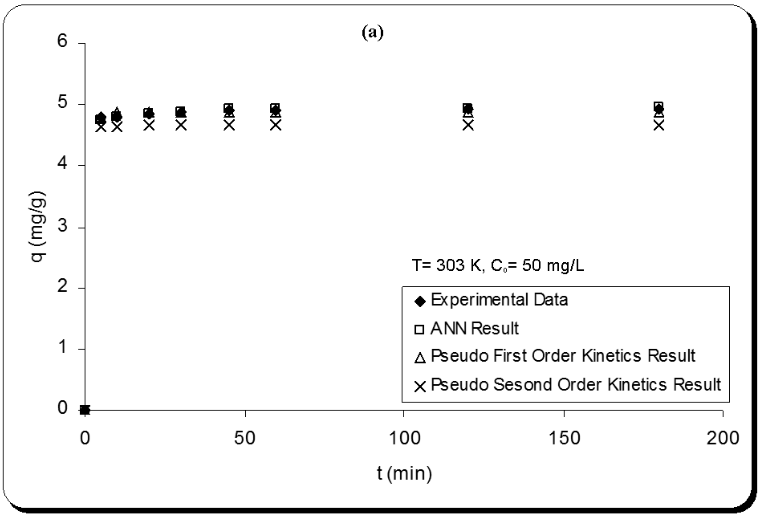
P. oceanica is one of the important bioindicator species for the ecosystem health of the Mediterranean Sea [37]. The accumulation of the dead leaves of *P. oceanica* on the beaches is a natural process. There are many advantages of this natural process:

1. This dead leaves protect beach sands against strong winter storms and prevent the coastal sand erosion [39].

2. These dead biomasses form a coastal ecosystem on the beaches where many animals live. Some birds use *P. oceanica* dead leaves as a nesting place [40].

These advantages are important for areas with no human activities. However, the accumulated dead leaves in populated areas are collected inasmuch as they give off bad odor and also are an eyesore especially in tourist places [11, 27, 28].

In France, there is an informative project on such dead leaves under way. The local people are informed about the advantages of these dead leaves. In some beaches, dead leaves are collected in the summer season from the beaches and then they are re-distributed to the beaches in winter to protect the beaches (Verlaque, M.; pers. commun.). Although this application is feasible for the hotels which are active only during the summer season. However, this application is unfortunately not applicable for the beaches that are active all-year-round such as some Aegean



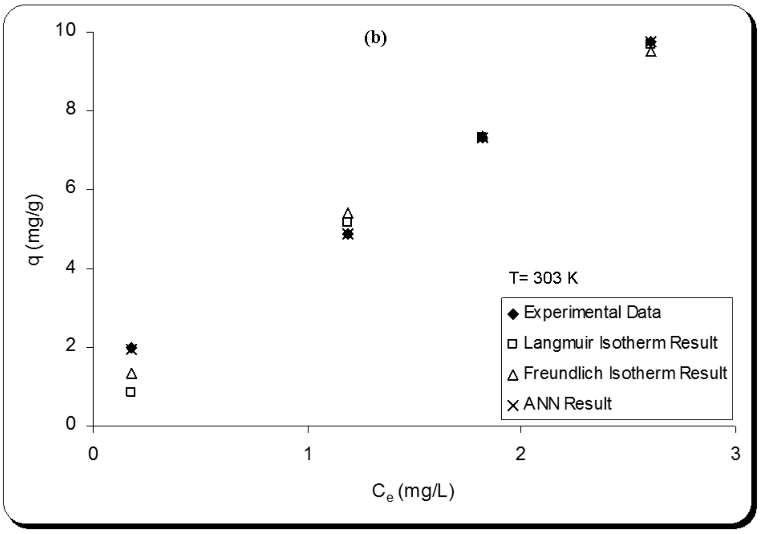


Fig. 5 *Comparison of artificial neural network results with traditional models: a) ANN vs. non-linear kinetics equations results and b) ANN vs. non-linear isotherm equations results.*

hotels. The present study proposes collection of dead leaves from the beaches where the tourist places are active throughout the year in view of tourist income, and also from some rocky places. It is known that dried biomasses of these species are burned during clean-up of some beaches. We showed in this study that this biomass can be used as an alternative low-cost adsorbent. Also, following the adsorption process, MB loaded *P. oceanica* dead leaves could be regenerated with different desorption techniques such as increasing ionic strength or temperature of washing solution, as we mentioned in our previous study [28]. In addition, modeling of methylene blue removal from aqueous solution by the dead leaves of *P. oceanica* that accumulated on Turkish coastlines was performed with ANN in the present study. According to the results of the present study, it can be observed from the presented figures that experimental and ANN modeling results were well in agreement. In conclusion, *P. oceanica* dead leaves, which are highly accumulated in the Mediterranean coastlines, can be used as an alternative and cheap adsorbent for adsorption of methylene blue from aqueous solutions. It is very important to note that the alive form of *P. oceanica* is an endangered species, therefore, this paper never proposes collection of alive form of this species under the water. The more researches are strongly warranted on the adsorption of other dyes by using the biomass mentioned in the present study.

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