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Fast and Economical Biosorption of Lead (II) Heavy Metal in Aqueous Solutions by *Bacillus licheniformis sp.*

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ABSTRACT
Lead is used in many areas of industry. Considering the prevalence of use, there is an intense lead and heavy metal content in the wastes resulting from these applications, and the toxic pollution caused by these metals affects the nearest water source directly or indirectly. Lead poses a serious threat to all life forms in the ecosystem, even at low concentrations in water. Lead recovery with environmentally friendly methods is both easier and more economical. *Bacillus licheniformis sp.* type was used as a biosorbent in the study. Nearly 98.4% of lead was removed by using the batch biosorption method, at 25 °C, pH 5.5, with an adsorption capacity of 42.92 in 120 minutes. from the water. The properties of the biosorbent, such as its morphological appearance, were characterized by scanning electron microscopy (SEM). Besides, the functional groups affecting biosorption in the surface structure were investigated by fourier transform Infrared spectroscopy (FT-IR)while its resistance to heat treatment was measured by thermal gravimetric analysis (TGA-DTA). The lead(II) element content in the aqueous solution was also determined by inductively coupled plasma optical emission spectroscopy (ICP-OES).

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ÖZET
Kurşun endüstrinin birçok alanınında kullanılmaktadır. Kullanım yaygınlığı düşünülürse buna bağlı olarak bu uygulamaların sonucunda ortaya çıkan atıklarda yoğun kurşun ağır metal içeriği bulunmakta ve büyük oranda en yakın su kaynağını doğrudan ya da dolaylı olarak etkilemektedir. Suda yüksek konsantrasyonlarda bulunması ile tüm yaşam formları için ciddi tehdit oluşturmaktadır. Bunların çevre dostu yöntemlerle iyileştirilmesi hem daha kolay hem de ekonomiktir. Çalışmamızda Dicle Nehri topraklarından izole edilen *Bacillus licheniformis sp.* kullanılarak sulardaki kurşun miktarı kesikli biyosorbsiyon yöntemi kullanılarak 25 °C de pH 5.5 da 120 dakikada 42.92 biyosorbsiyon kapasitesiyle % 98.4 oranında önemli ölçüde iyileştirildi. Biyosorbentin taramalı alan mikroskopisi (SEM) kullanılarak morfolojik görünümleri, fourier İnfrared spektroskopisi (FT-IR) dataları ile yüzey yapısında biyosorbsiyona etki eden fonksiyonel gruplar ve ısıl işlem karşısında gösterdiği direnç de termal gravimetrik analiz (TGA-DTA) verileri ile karakterize edildi. Sulu çözeltideki kurşun element içeriği İndüktif olarak eşleşmiş plazma optik emisyon spektroskopisi (ICP-OES) cihazı ile tespit edildi.

Çevre Bilimi

Araştırma Makalesi

Makale Tarihçesi
Geliş Tarihi :
Kabul Tarihi :

Anahtar Kelimeler
Atık su
Bacillus licheniformis sp.
Biyosorpsiyon
Biyosorbent
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INTRODUCTION

Wastes generated as a result of the operation of industrial facilities directly or indirectly affect water resources. The heavy metals in the content of these wastes cause serious consequences by threatening all life forms with their toxic effects and their capacity to result in environmental pollution (Chellaiah, 2018). Considering the necessity of water for all living organisms, increased toxicity due to heavy metal pollution in water resources is a crucial problem (Amirah et al., 2017). Lead (Pb) is the leading cause of heavy metal pollution. It is one of the toxic metals that cause environmental pollution and various health problems in many parts of the world (Hoyle-Gardner, 2021).

Many people, especially children, are exposed to heavy metals, which have toxic effects even at very low concentrations, in the environment. When these accumulate in the body, the immune system is suppressed. Their neurotoxicity and ability to inhibit the activity of critical enzymes related to the synthesis of vital biomolecules, as well as their carcinogenicity are some of the biomagnifications (Syed and Chinthala, 2015).

Lead is frequently used in the manufacture of car batteries, the printing industry, paint manufacturing, petrochemicals, fuel consumption, and photographic materials. There is a high concentration of lead content in the wastes resulting from these processes. Lead is one of the most dangerous heavy metals and is toxic even at low concentrations. It is highly soluble in water and is very harmful to all tissues in the human body, especially the kidneys and immune system (Morcali and Baysal, 2019; Zahra, 2012). Therefore, it is necessary to effectively reduce its concentration in aqueous environments and to choose the best method for improving the quality of the water (Zhang et al., 2013).

There are different methods and techniques for the removal of heavy metals from fluids. Chemical precipitation (Eltarahony et al., 2020), ion exchange (Dąbrowski et al., 2004), solvent extraction (Fillipi et al., 1998), reverse osmosis (Bakalár et al., 2009), evaporation (Mattenberger et al., 2008), adsorption (Baran and Duz, 2019) methods are some of them. Among these, heavy metal removal by the adsorption technique is widely used. The metal holders used for this technique are called adsorbents (Kouli et al., 2020).

Recently, there has been an increasing interest in the biosorption method using biosorbent in adsorption applications in this field as it is environmentally friendly, easy, displays a high adsorption efficiency, and its application stages are economical (Shokoohi et al., 2020). Among the biosorbents used for these applications, there are certain studies where various biological sources such as bacteria (Abedinzadeh et al., 2020), yeasts (Lu and Wilkins, 1996), algae (Rangabhashiyam and Balasubramanian, 2019), fungi (Qin et al., 2020), shellfish (M. Keshvardoostchokami, L. Babaei, A.A. Zamani, A.H. Parizanganeh, 2017) and, plants (Turkyilmaz et al., 2018) were used.

Gram-positive bacteria are preferred in applications using bacteria as a biosorbent. Such bacteria have a thick peptidoglycan layer, a feature that increases metal removal by providing high metal adsorption (Reith and Mayer, 2011).

Bacillus licheniformis, one of the members of the Bacillus genus of the Bacillaceae family, is a gram-positive species commonly found in nature. These bacteria have enzymatic activities such as catalase and oxidase and also show spore-forming properties. *B. licheniformis* is a bacterium that offers positive advantages with its strong resistance to adverse environments, resistance to high temperatures, efficient production of various enzymes, and carrying no risk of pathogenicity. In addition to having a thick peptidoglycan layer on the cell wall, teichoic and teichouronic acids in their structure are responsible for 60% of metal binding. (Huang et al., 2020; Samarth et al., 2012). In recent years, different biomasses have been used to increase biosorbent efficiency.

This study aims to perform the rapid removal of lead Pb (II) heavy metal from the aqueous solution by the environmentally friendly and economical batch biosorption method using the gram-positive *Bacillus licheniformis sp.* (*B. licheniformis sp.)* strain isolated from the soils of the Tigris river. The maximum metal removal capacity was determined by determining the experimental conditions. TGA-DTA, SEM-EDX, FTIR, thermodynamic and kinetic parameters were used to evaluate biosorption changes. As a result, it was determined that the biosorbent used was effective in metal removal.

MATERIAL and METHOD

Isolation of Bacterial Strains

Soil samples were taken for bacterial isolation from the coastal soils of the Tigris river. 1 gram of the soil sample was weighed and mixed with 4.5 mL of sterile distilled water. Then it was incubated at 80 °C for 10 minutes (Lennete et al., 1985). Dilutions were made to obtain colonies from the prepared soil sample. The diluted sample was inoculated on a nutrient agar medium containing 10% NaCl and incubated at 37 °C. Colonies formed in the medium were identified according to their morphological structures. Colonies showing *Bacillus* morphology were inoculated on nutrient agar medium and were grown at 37 °C for 1 day (Sneath et al., 1986). Taxonomic identification of *Bacillus licheniformis* sp. was determined by Dr Hüsamettin Aygün from Dicle University.

Stock Solution

0.798 g of $Pb(NO_3)_2$ was taken and dissolved with a sufficient amount of deionized water and the total volume was completed to 500 ml concentrations (5.0, 10.0, 15.0, 20.0, 25.0, 30.0, 40.0 and 50.0 mg/l).

Instruments and Equipment

The characterization of the biosorbent was determined via Mattson 1000 model fourier ınfrared spectroscopy (FT-IR), Shimadzu TGA-50 model thermogravimetric analysis (TGA-DTA), and EVO 40 LEQ model SEM data. Perkin-Elmer OPTİMA 5300 inductively coupled plasma optical emission spectroscopy (ICP-OES) was used for determining the adsorbed Pb(II) amount.

Preparation of Biosorbent

Bacillus licheniformis sp. strain which was isolated from Tigris river soils was used as the bio sorbent for the application. This micro-organism, which is a member of the Gram-positive bacilli, has a thick peptidoglycan layer. The structure of the peptidoglycan layer is a large polymer formed by cross-linking N-acetylglucosamine (NAG) and N-acetylmuramic acid (NAM) pentapeptide chains. In addition, teichoic acid in the structure of such bacteria has a large metal binding capacity (Reith and Mayer, 2011; Samarth et al., 2012; Tocheva et al., 2013).

Appropriate amounts of media were prepared for the growth of biosorbent using nutrient agar, and nutrient broth solid forms. *B. licheniformis sp.* was inoculated in a nutrient broth agar medium. It was incubated for 24 hours at 37 °C. After the growth, the micro-organisms were taken from the solid medium and inoculated into the previously prepared 1.0 litter nutrient broth medium. It was allowed to grow in a shaking bath at 37 °C for 24 hours. The medium content was then centrifuged at 7.000 rpm for 15 minutes. The pellet at the bottom was subjected to a series of washings with sterile distilled water. The dried biomass was passed through a 180 µm sieve. It was then stored in a sterile container ready for use in the biosorption study.

Pb(II) Ion Biosorption by Batch Method

Physical biosorption with *B. licheniformis sp.* was performed using the batch method (Verma et al., 2013), (Nazarzadeh et al., 2018). The method is based on the principle of removing heavy metals from the aqueous solution by being held by the adsorbent via the agitation process (Mwandira et al., 2020; Nazmara et al., 2020).

The Pb (II) ion solutions were prepared at varying concentrations from 5.0 mg/L to 50.0 mg/L. As a result of the experiments, the optimum pH for biosorption was determined as 5.5. Pb ion solutions at varying concentrations of 5.0-50.0 mg/L in a 100 mL glass flask were mixed with 25 mg of biosorbent. The biosorption was carried out at 150 rpm at different temperatures (25, 35 and 45 °C) for 60 minutes in the shaken environment. The biosorption capacity was calculated using the following equation (Masoumi and Ghaemy, 2014).

$$qe=((Co-Ce).V)/m \quad (1)$$

In the equation qe = adsorption capacity of the adsorbent (mg/g), C_0 = initial concentration of adsorbed substance (mg/L), Ce = concentration of adsorbed substance at equilibrium (mg/L), V= volume of solution (L), m = weight of biosorbent (stands for mg).

Desorption of Adsorbed Metal Ions in Biosorbent

B. licheniformis sp. were weighed 25 mg and placed in 100 ml flasks. The optimum pH was adjusted at a concentration of 5 ppm by dilution from the previously prepared Pb(II) stock solution. The metal solution was placed in flasks and shaken at 150 rpm for 30 minutes. The sample was taken, centrifuged, and the amount of lead was determined in the ICP-OES device. Then, HCl (hydrochloride) and HNO_3 (nitric acid) solutions prepared at 0.01, 0.05 and 0.10 M (molar) concentrations were added to the remaining biosorbent in the tubes and centrifuging was performed. The Pb ion analysis was performed on the ICP-OES device. The metal concentration (Cd) recovered at the end of desorption was subtracted from the bio-sorbed metal concentration (Ca). The obtained difference was divided by the metal concentration (Ca) and multiplied by 100 to calculate the % recovery.

$$\%A=((Ca-Cd))/Ca \times 100 \quad (2)$$

RESULTS and DISCUSSION

Characterization of Biosorbent

Functional Groups of surface structure with FT-IR spectroscopy data

In the FT-IR spectroscopy data performed in the range of 450.0 and 40,000 cm^{-1} in figure 1 and table 1, the strong asymmetric stretch band at 3267.24 cm-1 belonging to the –OH and –NH functional groups on the surface of the biosorbent shifted as a result of the interaction between the adsorbed metal and the adsorbent. Frequency shifts in the 1727.06 and 1633.16 cm-1 bands of the Amide I and Amid II groups also occurred due to C–O- stretching. In addition, 1230.78 and 1230.78 due to P=O and C–O- stretching. The shifts in the 1052.42 bands show that Pb(II) metal is bound through these groups (Kariuki et al., 2017; Sabri et al., 2018; Zhang et al., 2013).

Table 1. Functional group frequencies of the surface of Pb (II) loaded/unloaded by *B. licheniformis sp.* (v, cm^{-1})
Çizelge 1. Pb (II) yüklenmiş/yüklenmemiş B. licheniformis sp.'in yüzey yapılarına ait fonksiyonel grupların frekansları (v, cm^{-1})

	–OH, –NH	–CH2-	Amide I	Amide II	–OH bending	C-H, COO–	P=O (phosphodiester)	C–O-Stress
Pb Unloaded	3277.67	2929.61	1727.06	1633.16	1440.35	1385.23	1230.78	1052.42
Pb loaded	3267.24	2928.77	1725.66	1631.01	1440.35	1390.32	1228.11	1050.26

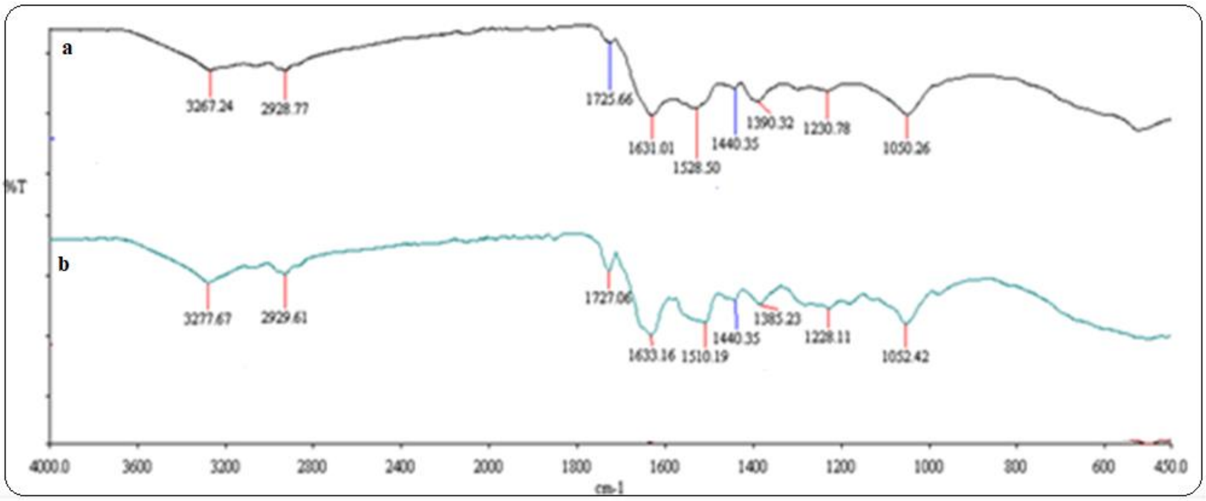

Figure 1. FTIR spectrum data of *B. licheniformis sp.* biosorbent a. After Pb(II) metal ion biosorption, b. Before biosorption
Şekil 1. B. licheniformis sp. biyosorbentinin FTIR spektrum verileri; a). Pb(II) metal iyonu biyosorpsiyonu sonrası, b). Biyosorbentinin uygulama öncesindeki.

TGA-DTA Analysis Data

The thermal gravimetric analysis was used to determine the biosorbent mass loss. (Zendehdel et al., 2019). The temperature range was determined as 25-1000 °C (Amirah et al., 2017). Figure 1, in the TGA and DTA data of the biosorbent mass losses, it is seen that the 4.70 % mass loss in the 25-107 °C range is due to the physically adsorbed water from the biosorbent. It is seen that the mass loss, which is 5.51% at temperatures of 107-198 °C, is related to water, which occurs by the combination of cellulosic -OH groups (Maranescu et al., 2019). The 37.51% mass loss that occurred in the range where the temperature was 198-337 °C occurred with carbonization. At the intermediate value where the temperature is 337-804 °C, a mass loss of 45.55% occurred, which shows that carbon is converted into carbon dioxide (Baran and Duz, 2019) (Figure 2 and Table 2).

Table 2. TGA-DTA data of biosorbent mass loss rates
Çizelge 2. Biyosorbentin TGA-DTA ile kütle kayıp oranları

Temperature range	% Mass loss
25-107 °C	4.70
107-198 °C	5.51
198-337 °C	37.51
337-804 °C	45.55

SEM Micrograph Images

The appearance of the morphological structures of the biosorbent before and after biosorption was evaluated via SEM images. When the SEM micrograph is examined, it is seen that there is a metallic glow with the effect of the metal adsorbed on the bacteria in their morphological appearance after the interaction with the Pb (II) metal ion (Ayucitra et al., 2017; Dahaghin et al., 2017). In addition, significant morphological shrinkage and irregular shape appearance (Su et al., 2020) occurred in the biosorbent (bacterial cells) after metal biosorption against the control biosorbent without metal interaction (Figure 3).

Biosorption Isotherm

The biosorption 5.0, 10.0, 15.0, 20.0, 25.0, 30.0, 40.0, and 50.0 mg/L concentrations at 298, 308, and 318 kelvin (K) temperatures versus metal ion equilibrium concentration versus metal ion amount adsorbed per milligram of biosorbent via the Langmuir (Taleb et al., 2020) and Freundlich (Khameneh and Moharreri, 2020) equations the compatibility with the adsorption isotherm was tested.

The Langmuir equation

Molecules adsorbed on the surface are in the form of a monolayer. The adsorption does not occur on the entire surface, but in the form of intermittent coverings. The energy of adsorption is constant on the whole surface and there is no interaction between the molecules attached to the surface (Nasab et al., 2020).

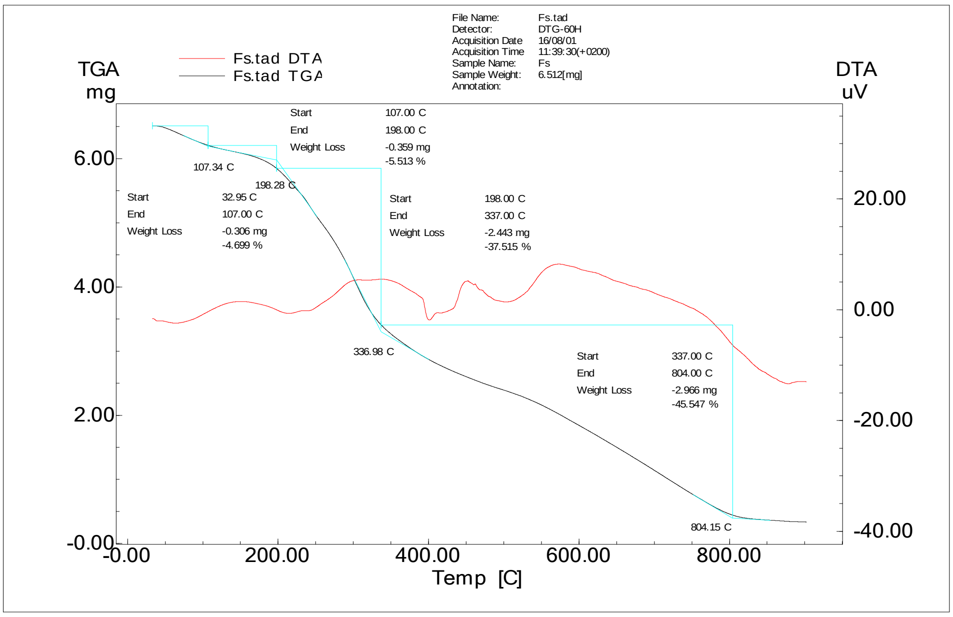

Figure 2. Mass losses of biosorbent against heat treatment with TGA-DTA data
Şekil 2. Biyosorbentin TGA-DTA verileri ile ısıl işlem karşısında kütle kayıpları

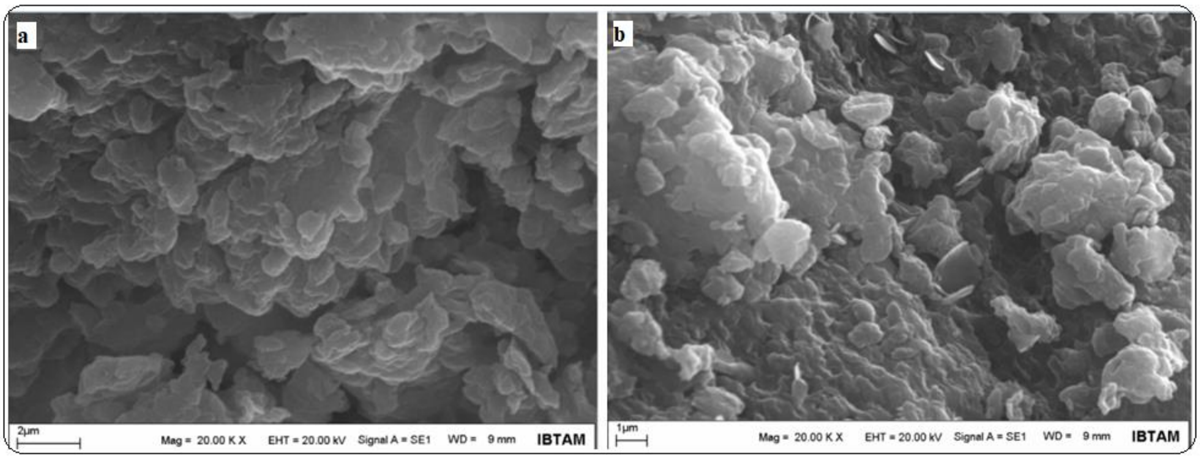

Figure 3. SEM micrograph images of the biosorbent, (a) before metal interaction and (b) after metal interaction
Şekil 3. Biyosorbentin SEM görüntüleri, (a) metal etkileşiminden önceki ve (b) metal ile ekileşiminden sonra ki mikrografisi

Qe =Qmax bCe/(1+bCe) (3)

When equality is established;

(Ce)/Qe=1/Qmaxb+1/Qmax Ce (4)

Qe (mg); the number of metal ions adsorbed on unit amount of adsorber, Qmax(mg); monolayer capacity, Ce (mg); concentration of the metal in the equilibrium solution, b (L); It represents the Langmuir adsorption equilibrium constant. In this equation, when Ce versus Ce/Qe is plotted, Qmax is calculated from the slope of the line and b constant is calculated from the shift value.

The Freundlich equation:

The Freundlich equation shows that there is a direct proportionality between the metal concentration in the solution and the adsorbent surface. As the metal concentration increases, the adsorbed concentration also increases. This experimental equation is based on multilayer adsorption on a heterogeneous surface. Although the Freundlich isotherm is widely used, it cannot provide sufficient information for monolayer adsorption capacity, unlike the Langmuir model. The Freundlich equation (Wu et al., 2020) is expressed as:

Logqe=(logk)+1/n logCe (5)

qe: the number of metal ions adsorbed on the unit amount of adsorbent, Ce: the concentration of the metal in the equilibrium solution, k: the adsorption capacity of the biomass, n: the effect of the concentration on the adsorption capacity.

Adsorption isotherm data with Langmuir and Freundlich equations are given in table 3 and figure 4.

Table 3. Freundlich and Langmuir constants for Pb(II) biosorption on biosorbent
Çizelge 3. Biyosorbent üzerinde Pb(II) biyosorpsiyonuna ilişkin Freundlich ve Langmuir sabitleri

	Freundlich constant			Langmuir constant		
T (K)	**K_F (1/dak)**	**n**	**R^2**	**Q_m (mg/g)**	**B (L/mg)**	**R^2**
298	14.25	3.36	0.778	33.67	1.151	0.9952
308	17.58	3.51	0.7878	38.46	1.699	0.9931
318	21.04	3.65	0.8082	42.92	2.354	0.9920

(**K_F**: Freundlich Constant (adsorption capacity L/g), **Q_m** : Langmuir constant maximum metal sorption (mmol/g); **n:** Freundlich Constant, **b** : Langmuir adsorption equilibrium constant (L/mmol) ,**R^2:** Correlation coefficient.)

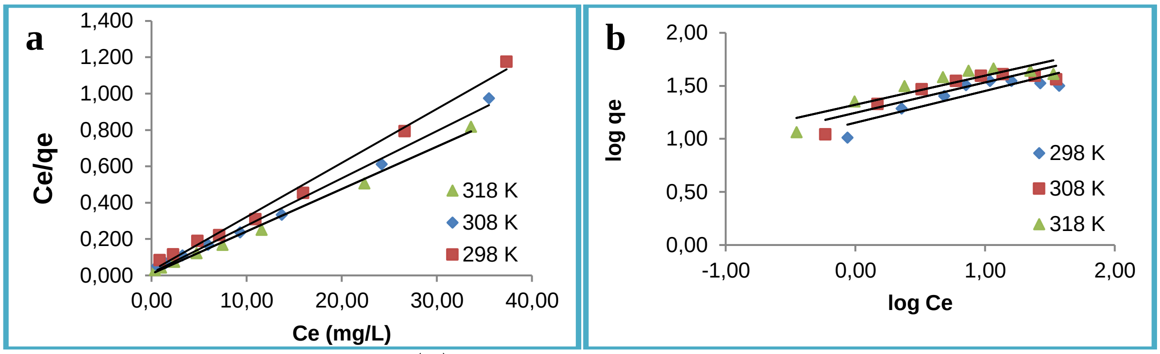

Figure 4. The biosorption isotherms of Pb(II) of the biosorbent at different temperatures **a.** Langmuir type, **b.** Freundlich type radiographs
Şekil 4. Biyosorbentin farklı sıcaklıklarda Pb(II)'nin biyosorpsiyon izotermlerinin a. Langmuir tipi, b. Freundlich tipi grafileri

The magnitude of the R^2 values of the isotherms plotted using the experimental data of Pb(II) metal on the biosorbent in Table 1 shows that the isotherms are compatible with the Langmuir type (Wen et al., 2018). Monolayer biosorption capacities (Qm) ranged from 32 to 44 mg/g depending on temperature. There is no significant difference between the monolayer biosorption capacity of the biosorbent. With the increase in temperature, the biosorption capacity also increased steadily (Ajmal et al., 2003).

Biosorption Kinetics

The biosorption kinetics of Pb(II) was evaluated using two kinetic models. These are Pseudo-First Order and Pseudo-Second Order kinetic models. The kinetic models determine how the constant uptake rate of the adsorbent affects the residence time at the solid and solution interface (Zu et al., 2020).

Pseudo-First Order kinetic model equation:

$$\log(qe-qt)=\log qe-k\_1/2,303\ t \qquad (6)$$

qe and qt; the number of metal ions adsorbed at equilibrium and time t (mg/g), k1; stands for pseudo-first-order adsorption rate constant (min-1) (Yang et al., 2014).

Pseudo-second degree kinetic model equation:

$$dqt/dt= k2(qe-qt)^2 \qquad (7)$$

qe and qt; the amount of metal ions adsorbed at equilibrium and time t (mg/g), k2; is expressed as the pseudo-second degree adsorption rate constant (g/mg min) (Zhang et al., 2013).

To explain the biosorption mechanism, the kinetic data of Pb(II) metal ion removal was modelled. From the experimental data fixed by linear regression analysis in equations 6 and 7, pseudo-first-order coefficients of determination and pseudo-second-order kinetic models were evaluated. All parameters related to both kinetic models are indicated in Table 4 and shown in Figures 5 (a, b). When the correlation coefficients (R^2) from each kinetic model are compared, it is seen that they fit the pseudo-second kinetic model. (Zendehdel et al., 2019).

Activation Energy and Thermodynamic Parameters in Biosorption

Since the best kinetic model for the biosorption of Pb(II) on Fs was determined as the pseudo-second-order kinetic model, the k2 rate constants in the Arrhenius equation were applied to calculate the activation energy of the adsorption process. Arrhenius equation is given below: (Baran and Duz, 2019).

$$\ln k=\ln Ao - Ea/RT \qquad (8)$$

Table 4. Biosorption kinetics of Pb(II) at different temperatures Pseudo-First and Pseudo-Second order rate constants

Çizelge 4. Farklı sıcaklıklarda Pb(II)'nin biyosorpsiyon kinetiği Pseudo-First ve Pseudo -Second mertebe hız sabitleri

	Pseudo-First order constant			Pseudo-Second order constant		
Temperature (°C)	**Kpf** (1/dak)	q_e (mg/g)	R^2	**Kps** (g/mgdak)	q_e (mg/g)	R^2
25	0.0309	5.6247	0.9526	0.0221	22.1239	0.9948
35	0.0474	3.6966	0.9825	0.0362	23.6407	0.9960
45	0.0859	3.2107	0.9476	0.0520	24.5098	0.9977

(Kpf: Pseudo-first order constant, Kps: Pseudo-second order constant, qe: The number of metal ions adsorbed on 1 g of adsorbent at equilibrium, R^2: Correlation coefficient)

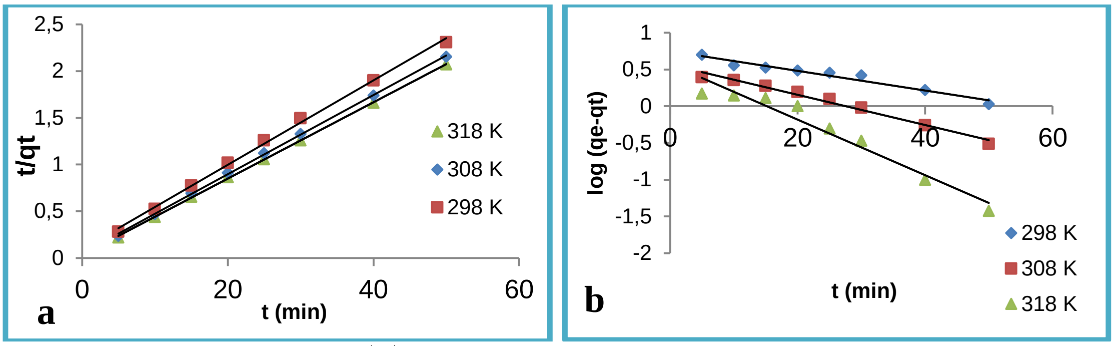

Figure 5. Biosorption kinetics of Pb(II) of the biosorbent at different temperatures a. Pseudo-second order b. Pseudo-first order graphs

Şekil 5. Biyosorbentin farklı sıcaklıklarda Pb(II)'nin biyosorpsiyon kinetiği a. Yalacı-ikinci mertebe b. Yalacı-birinci mertebe mertebe grafikleri

In the equation, k; the value of the rate constant calculated from the kinetic model with which the biosorption is compatible, Ea; Biosorption activation energy (J/mol), Ao; Arrhenius constant, R; Ideal gas constant (8.314 J/mol.K), T; It represents temperature (Kelvin).

The activation energy of Pb(II) biosorption was found to be 33.70 (table 5). Results with activation energy (Ea) of between 5 and 50 kJ/mol are considered physical adsorption (Deniz and Saygideger, 2010).

Table 5. Activation energy (Ea) values in biosorption of Pb(II) at different temperatures

***Çizelge 5.** Farklı sıcaklıklarda Pb(II)'nin biyosorpsiyonunda aktivasyon enerjisi (Ea) değerleri.*

T (K)	1/T	kps	Inkps	E_a (kj/mol)
298	3.36×10^{-3}	0.0221	-3.8106	
308	3.25×10^{-3}	0.0362	-3.3202	33.70
318	3.14×10^{-3}	0.0520	-2.9561	

The thermodynamic parameters, which are functions of temperature under constant pressure (1atm) and differ with increasing temperature, are calculated from the following equations:

$$\Delta G^o = -RTlnKd \quad (9)$$

In the equality; Kd, equilibrium constant, Ca; the amount of metal ion adsorption at equilibrium (mol/L), Cs; expressed as the amount remaining in solution at equilibrium (mol/L) (Deniz and Saygideger, 2010).

$$\Delta G^o = \Delta H^o - T\Delta S^o \quad (10)$$

$$lnKd=(\Delta S^\circ)/R-(\Delta H^\circ)/RT \quad (11)$$

In equations; ΔH°, ΔS°, and ΔG° represent standard enthalpy, entropy, and free energy changes, respectively (Biswas et al., 2020). Thermodynamic parameters obtained for the removal of Pb(II) with Fs biomass are given in Table 6. In addition, the negative value of ΔG indicates that the biosorption process

It is an indication that it happens spontaneously. The positive value of ΔS° is a value indicating an increase in disorder at the solid-liquid interface during biosorption. As a result, the positive ΔH° indicated that the biosorption process was endothermic (Ajmal et al., 2003; Subramani et al., 2019).

Effect of Biosorbent Dose and Time on Biosorption

The effect of increasing the amount of biosorbent in Pb(II) solution with an initial concentration of 10 mg/L on metal biosorption was investigated. It was observed that increasing the amount of biosorbent caused a decrease in the equilibrium concentration. The use of biosorbent in high concentrations causes a decrease in the surface area effective for biosorption due to agglomeration in parts (Mousavi et al., 2019) (Figure 6(a)). The interaction time in biosorption is an important parameter in adsorbing Pb (II) ions by biosorbent. As seen in Figure 6(b), Pb(II) ions were adsorbed by the biosorbent very quickly in a short time. Prolongation of time slowed down the biosorption reaction (Heraldy et al., 2018).

Table 6. Thermodynamic parameters of biosorption of Pb(II).
Çizelge 6. Pb(II)'nin biyosorpsiyonunun termodinamik parametreleri.

T(K)	1/T	Inb	ΔG° (kj/mol)	ΔH° (kj/mol)	ΔS° (j/molK)
298	3.36×10^{-3}	0.141	-0.568		
308	3.25×10^{-3}	0.530	-1.527	28.18	95.82
318	3.14×10^{-3}	0.856	-2.485		

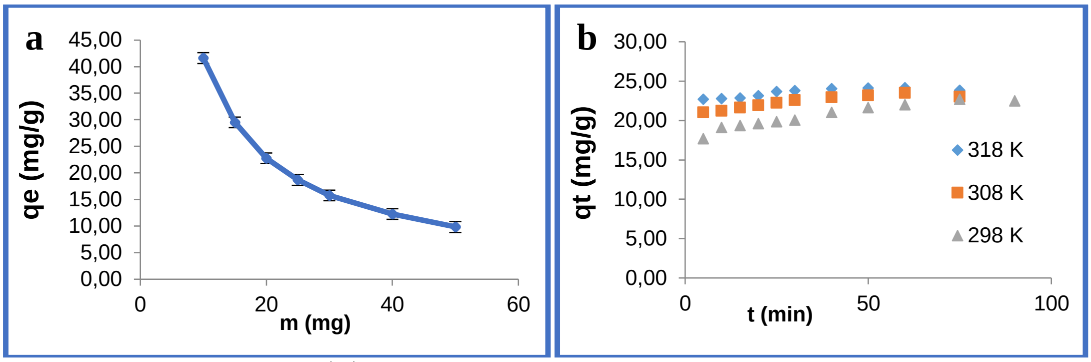

Figure 6. a. Effect of adsorbent dose on Pb(II) biosorption at 25 °C b. Effect of time on the biosorption of Pb(II) by Fs biosorbent at different temperatures
Şekil 6. a. 25 °C'de Pb(II) biyosorpsiyonuna adsorbent dozu etkisi b. biyosorbentin farklı sıcaklıklarda Pb(II)'nin biyosorpsiyona zamanın etkisi.

Effect of pH on Biosorption

The effect of pH on the adsorption of Pb(II) metal ion on biomass was investigated in the range of 1.0-10.0 and adsorption capacities were calculated accordingly. As can be seen in Figure 7, the pH factor is an important parameter that affects the adsorption behaviour. It was observed that the highest capacity occurred at pH 5.5. In addition, pH affects the solubility of the metal and the functionality of the microbial sorbent surface (Pugazhendhi et al., 2018; Sabri et al., 2018).

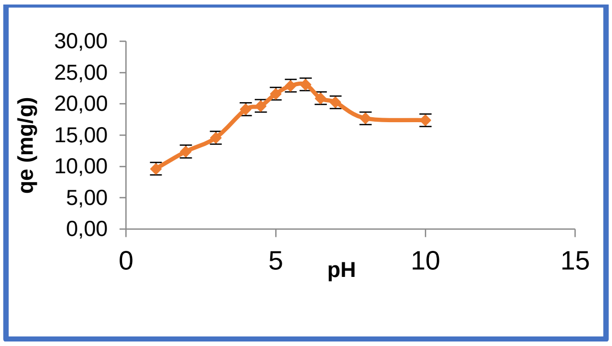

Figure 7. pH effect on biosorption
Şekil 7. Biyosorbsiyona pH'nın etkisi.

Desorption of Biosorbent

Metal recovery after biosorption is important in evaluating the efficacy and reusability of the biosorbent. In the recovery of Pb(II) metal ion adsorbed on the biosorbent as a result of biosorption, 0.01, 0.05 and 0.10 concentrations of HCl and HNO_3 solutions were applied and the most effective result was 0.10 NHO_3 solution with 98.6% lead recovery. (Table 7 and Figure 8).

Table 7. Desorption of Pb (II) metal ion adsorbed in biosorbent with HCl and HNO_3 (T=25 °C, Co=5 ppm, rpm=150 rpm, t=60 min and m=25 mg)
Çizelge7. Biyosorbentin HCI ve HNO_3 ile desorpsiyonu (T=25 °C, Co=5 ppm, hız=150 rpm, t=60 dak ve m=25 mg)

	Solution Concentration (mol/L)	% Desorption Efficiency
HCl	0.01	64.8±0.14
	0.05	75.9±0.19
	0.10	91.3±0.12
HNO_3	0.01	68.7±0.09
	0.05	79.4±0.28
	0.10	98.6±0.15

N=3, 95% confidence interval

In a study by Bangaraiah et al., they reported 75% heavy metal removal in a study using biomass. In another similar study, Hoyle-Gardner et al. reported that it has a metal recovery capacity of about 89% in lead biosorption. In conclusion, *Bacillus licheniformis* sp. The bacteria used in this study proved to be good adsorbents in metal removal in aqueous solutions. Biosorption conditions; Metal removal can be improved with parameters such as temperature, time, pH, amount of biosorbent and time. It shows that bacteria can be used effectively for Pb(II) removal from aqueous solutions and wastewater when appropriate experimental conditions are provided.

The studies carried out to remove Pb(II) metal ions from water by biosorption are given in Table 8.

Table 8. Biosorption studies for the removal of Pb(II) metal ions in water.
Çizelge 8. Pb (II) metal iyonunun sularda giderimi için yapılan biyosorbsiyon çalışmaları.

Biosorbent	Biosorbent Amount (g)	pH	T (°C)	Time (Hour)	% Removal	References
Talaromyces islandicus	4.56	5	30	2	90	(Sharma et al., 2020)
Eucalyptus camaldulensis	0.02	7	25	0.66	85	(Sabri et al., 2018)
Algae (Mixed culture)	5.52	6	30	1.1	95.43	(Mousavi et al., 2019)
Ralstonia solanacearum	0.1	6	35	12	90	(Pugazhendhi et al., 2018)
Sargassum muticum	0.3	5	20	0.5	96	(Hannachi and Hafidh, 2020)
B. licheniformis	0.7	6	30	12	98	(Wen et al., 2018)
B. licheniformis sp. This study	0.25	5.5	25	1	98.4	-

CONCLUSIONS

Since heavy metals are toxic, they are undesirable for ecological balance and living things. The most important of these is water pollution. Many methods are applied for metal removal in water. One of these methods is the biosorption method. In the study, bacterial biomass and lead biosorption in aqueous solutions was studied. In the study, Pb(II) metal ion, which has a toxic effect even at low concentrations, was detected in B. licheniformis sp. and was effectively removed with its biomass. The FT-IR, TGA-DTA, and SEM data were used to determine the characterization of the biosorbent. At the same time, factors affecting biosorption such as biosorbent dose, biosorption kinetics and isotherm, temperature, pH, and time were investigated. It was determined that *B. licheniformis sp.* biosorbent removed Pb(II) ions significantly rapidly with an adsorption capacity of 42.92. The biosorption method applied in the study has the advantage of being an environmentally friendly, economical and easy to apply process. In addition, the fact that the bacterial biomass (Fs) used is resistant to extreme conditions and there is no risk of pathogenicity provides a great advantage. Adsorption capacity can be increased by improving the factors affecting biosorption and changing the optimization conditions. In addition, the reproducibility of the used biomass was proven by desorption.

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Contribution of the Authors as Summary

The contribution of the authors is equal

Statement of Conflict of Interest

Authors have declared no conflict of interest

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