

# *Spirulina Platensis* ve Phosmet, Ethion, Methyl Parathion pestisitlerinin Bioremediasyonu

## *Spirulina Platensis* and Bioremediation of Phosmet, Ethion, Methyl Parathion pesticides

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**Özetçe—** Bu çalışmada, CBRN (Kimyasal, Biyolojik, Radyoaktif, Nükleer) tehditlerinin Toksik Endüstriyel Kimyasal kategorisindeki bazı pestisitlerin mavi-yeşil mikroalglerle biyoremediasyon etkinliği araştırılmıştır. Söz konusu pestisitler insan, canlı ve çevre sağlığına oldukça zararlı olmakla beraber doğada kalıcılığı yüksektir. Son zamanlarda tarımsal alanda pestisitlere olan talebin ve kullanımının artması üretim, taşıma ve depolama alanlarında risk oluşturmaktadır. Kullanmış olduğumuz mikroalg hali hazırda başta tıp ve ilaç sanayisi olmak üzere önemli alanlarda kullanımının yanı sıra pestisitleri metabolize etme yeteneği de bilinmektedir. Pestisitlerin zararlı etkilerini yok etmek ya da en aza indirmek için çevre dostu olan biyoremediasyon tekniğinin kullanılması, doğaya ve geleceğe katkı sağlamaktadır. Seçilen mikroalg ve pestisitlerin daha önce araştırılmamış olması çalışmamızın özgünlüğünü kanıtlamaktadır. Pestisit remediasyon miktar ölçümleri elektrokimya yöntemi ile yapılmış ve *Spirulina platensis*'in 7. gün sonunda Phosmet, Ethion ve Metil Paration'u sırasıyla % 70.0, % 61.0, % 50.0 oranında ortamdaki uzaklaştırdığı tespit edilmiştir.

**Anahtar Kelimeler—**KBRN (Kimyasal, Biyolojik, Radyoaktif, Nükleer); Pestisit; Biyoremediasyon; Mikroalg.

**Abstract—** In this study, the bioremediation efficiency of some pesticides in Toxic Industrial Chemical category of CBRN (Chemical, Biological, Radioactive, Nuclear) threats with blue-green microalgae was investigated. Although these pesticides are highly harmful to human, living and environmental health, they are highly persistent in nature. Recently, the increasing demand and use of pesticides in the agricultural field poses risks in production, transportation and storage areas. The microalgae we have used are already known for its ability to metabolize pesticides, as well as its use

in important areas, especially in the medicine and pharmaceutical industry. Using environmentally friendly bioremediation technique to eliminate or minimize the harmful effects of pesticides contributes to nature and the future. The fact that the selected microalgae and pesticides have not been investigated before proves the originality of our study. Pesticide remediation amount measurements were made by electrochemistry method and it was determined that *Spirulina platensis* removed Phosmet, Ethion and Methyl Paration by 70.0%, 61.0%, 50.0%, respectively, at the end of the 7th day.

**Keywords—**CBRN (Chemical, Biological, Radioactive, Nuclear); Pesticide; Bioremediation; Microalgae.

### I. INTRODUCTION

Wastes which cannot be processed or consumed in time such as various metals, residual oils, slag and mine wastes, hospital wastes that are left over from materials used by processing or consumption in the industrial sector, are defined as industrial waste. These wastes cause irreversible long and short term damages to human health, nature, the environment and all living things. Considering the waste generation worldwide; storage and disposal of toxic industrial chemical wastes is very costly and difficult [1]. If toxic industrial chemicals are not disposed of properly, they can survive in nature for a long time. Among 87,000 commercial chemicals, 53 have been identified as persistent, bioaccumulative and toxic by the US Environmental Protection Agency (USEPA, 2000). Unless remediation of toxic industrial chemicals, whose harmful effects continue for many years, it puts living things and nature at risk. Marine pollutants can be carried

far and wide fields by air and water currents, ocean currents and migrating fishes. For this reason, pollution is not only limited to geographical boundaries, but also becomes a worldwide problem [2] .

Pesticides are known as a group of chemicals used to destroy insects, weeds, fungi, bacteria etc. Pesticides are generally categorized according to the target organism and the prefix of each category changes to describe the target. For example aviscides, rodenticides, insecticides, miticides, molluscicides, nematocides, herbicides, fungicides, algicides, bactericides, and viricides [3,4] .

Uncontrolled and unconscious use, accidental or deliberate release of these substances, accidents in the stages of production, destruction and storage threaten the health of all living things and also harm nature. It has been reported that pesticides can enter the food chain and accumulate gradually in higher-level organisms and even in the human body through biomagnification [5]. Toxic industrial chemicals can be cause of poisoning, respiratory diseases, cardio-toxic diseases, allergic reactions in the acute period, they can lead to irreversible health problems such as chronic diseases and even death. It is also known that they contribute to the formation of disorders such as neurodegenerative diseases, birth and developmental disorders, endocrinological and metabolic diseases, neurological disorders, hematological malignancies, reproductive system disorders and cancer as a result of prolonged exposure [5,6] .

Many of these chemicals are designed to disrupt the physiological activities of the target organism, leading to dysfunction and decreased vitality. Pesticide residues are an important source of contamination to environmental factors such as air, water and soil. Approximately two million tons of pesticides are used annually in the world. Increasing the amount of pesticide use for high agricultural production means much more environmental (soil, water and air) pollution. It has been determined that pesticides have high lipophilicity, bioaccumulation, long half-life and long-distance transport potential, causing air, water and soil contamination even years after their use [7]. According to a study by WHO, 80% of all pesticides are used by developing countries. Due to legal gaps, inappropriate market regulations, and people's ignorance and unconsciousness, agricultural workers in developing countries are exposed to high levels of agrochemicals, including pesticides [8].

Pesticide exposure has been recorded as the primary occupational hazard among people engaged in agriculture in developing countries [9]. Farmers are considered to be the main risk group, but formulators, loaders, mixers, production workers and agricultural workers are among the most vulnerable groups. Non-occupational hazards arise due to the pollution of the ecosystem or habitat as a whole. It should not be forgotten that pesticides can also be used for sabotage and terrorist purposes due to their high

toxicity. In addition, it is known that approximately two million hospitalizations have occurred due to complications of the organophosphate insecticide exposure and it is estimated to cause approximately 100000 deaths per year in the world. Due to their stability in the environment, the use of pesticides has been banned in many developed countries, but they are still used in developing countries [10].

Bioremediation is an environmentally friendly healing technique that uses the natural ability of microbes such as algae, fungi and bacteria to degrade / detoxify organic and inorganic contaminants from industrial wastewater. Bioremediation technique uses the metabolic properties of microorganisms to remove toxic contaminants. For this reason, it is an environmentally friendly and economical technique that stands out with its wide and operational application areas, less sludge production and simple structural installation. Among the qualities that make bioremediation superior to other methods are the low cost of the process and the harmlessness of by-products formed after bioremediation [11,12].

Micro-algae are microscopic photosynthetic organisms found in both marine and freshwater environments. They serve as the source of oxygen and the first link of food chains in water systems. Many microalgae species have a gelatinous extracellular polysaccharide layer surrounding their cells, called envelopes, sheaths, or capsules [13]. Microalgae contain lipids and fatty acids as membrane components, storage products, metabolites and energy sources. Most of the simple, single-celled microalgae are used in the food industry. The pigment substances they contain are used as additives in the pharmaceutical industry, medicine, pharmacy and cosmetic products. In addition to these, they are also used as human and animal food. Especially in developing countries, they are consumed as a food source due to their rich protein content [14]. There are two main reasons why microalgae are preferred in bioremediation technique. The first is that they help oxygenate wastewater pools poor in dissolved oxygen, thanks to their ability to produce oxygen through photosynthesis. The second is that they have the feature of an organism that can grow rapidly in sea and inland waters rich in nitrogen and phosphate and take an active role in excessive reproduction [15].

*Spirulina* is a photosynthetic, filamentous, spiral-shaped, multicellular blue-green microalgae. The most important species are *Spirulina maxima* and *Spirulina platensis*. *Spirulina*, which does not have any toxic effects, is considered to be the miracle food used effectively in viral attacks, tumor growth, anemia and malnutrition. Optimum growth temperature for *Spirulina platensis* is between 35.0 ° C and 37.0 ° C. *Spirulina* is a type of alga that can be monocultured because it prefers high pH levels. Growth temperature for *Spirulina platensis* is minimum 18.0 ° C and maximum 39.0 ° C [16].

Phosmet is a non-systemic organophosphate insecticide used in both plants and animals. Phosmet is used in various fruit and ornamental plants as well as vines for the control of aphids, absorbers, mites and fruit flies. Like other organophosphates, phosmet is an inhibitor of the cholinesterase enzyme. Acute phosmet poisoning leads to nausea, vomiting, abdominal cramps and diarrhea. Acute period effects in excessive exposure may cause muscle spasms, loss of muscle coordination, confusion, respiratory distress and increased body secretions. Phosmet's oral LD 50 value is 113 mg/kg in female rats and 369 mg/kg in male rats while 23.1 mg/kg in female mice and 50.1 mg/kg in male mice [17].

Methyl parathion is an organophosphate insecticide widely used for the control of various insects and pests. Although its use is increasing in developing countries it has been banned in many countries [12]. Toxic effects of methyl parathion include headache, nausea, diarrhea, difficulty breathing, excessive sweating, incoordination and unconsciousness. Methyl parathion toxicity causes serum cholinesterase inhibition, cardiovascular system disorders (cardiovascular lesions, abnormal heart rate and increased heart-body ratio), reproductive and nervous system diseases (headache, muscle weakness, insomnia, dizziness and loss of consciousness). Methyl parathion is acutely toxic at low doses when administered orally (LD50 = 4 mg / kg body weight) or by inhalation (LC50 = 0.13 mg / L). WHO classified Methyl parathion as "extremely dangerous chemical" [18].

Ethion is a chemical used as a pesticide in agriculture. Pure Ethion is a clear and yellowish liquid with an unpleasant smell of sulfur. Besides being used in spray form for pest control, ethion is also used as a liquid adsorbed on powder or granules. Exposure to Ethion occurs through inhalation, absorption and retention in the skin. Ethion, which is widely used as an insecticide, causes toxic effects in humans. Ethion chemically reacts in the brain and nerves with an important enzyme called acetylcholinesterase, and this reaction can stop the enzyme from working properly. Consequently, the signals between the muscles and nerves are disrupted and involuntary spasms, nausea, sweating, urination, loss of vision, and confusion may occur. Ethion LD50 values via gavage have been reported as 65 mg/kg for male Sherman rats and 27 mg/kg for females [19].

In this study, we aimed to investigate the effectiveness of the bioremediation technique with *Spirulina platensis* in order to minimize the toxic effects of Phosmet, Ethion and Methyl Paration on the environment and living things.

## II. MATERIAL AND METHODS

### 2.1. Algal Strain (*Spirulina Platensis*)

*Spirulina platensis* culture was obtained from the Sum *Spirulina platensis* facility in liquid form as study material.

For the production of *Spirulina platensis*, Zarrouk medium culture medium was used.

### 2.2. Chemicals

Phosmet (Cas No 732-11-06), Methyl parathion (Cas No 298-00-0) and Ethion (Cas No 563-12-2) purchased from Sigma-Aldrich company.

### 2.3. Experimental Design

*Spirulina platensis* cultures used in the study are cultivated in Zarrouk culture medium and they were positioned in the laboratory environment to receive direct sunlight on the glass side. Temperature measurements were taken twice a day, in the morning and in the afternoon. The illumination of the culture bottles was set as a 12 hour photoperiod (12 hours dark, 12 hours light). Intensity and healthy reproductive levels of the cultures were monitored daily. Temperature and pH measurements of the culture bottles were made with ADVA brand Ad12 model device. Before the daily optical density measurement, the culture bottles were mixed and homogenized and samples were taken. The samples were placed in quartz tubes and read at 670 nm wavelength with UV-Vis spectrophotometer.

Pesticide stocks were prepared using 33.0% ethyl alcohol under the fume hood. For each pesticide used in the study, 100.0 mL of culture was taken and labeled individually in glass bottles. 25.0 mL each of 10.0 ppm stock Phosmet and Methyl parathion were added to the culture bottles. 20.0 mL of 12.0 ppm stock Ethion was added to the culture. The initial concentration for each pesticide culture mix was set at 2.0 ppm. Culture bottles with pesticide cultivation were observed for seven days, 5.0 mL samples were taken from each of the bottles on the 2nd, 4th, and 7th days. The samples taken were filtered using 30 µm filter paper.

### 2.4. Electrochemical cell assembly

Voltammetric studies were performed with BAS 100B / W Model device. Ag / AgCl electrode was used as reference electrode, glassy carbon electrode (GCE) was used as working electrode and platinum electrode was used as counter electrode. In voltammetric experiments the oxygen in the environment must be removed, as the dissolved oxygen in the cell will be reduced on the glassy carbon electrode surface. For this reason, nitrogen gas was passed through the electrochemical cell continuously for 10 minutes, preventing the reduction of oxygen in solution again.

### 2.5. Electrochemical analysis

2.0 mL of 0.1 M phosphate buffer selected as the support electrolyte was taken into the electrochemical cell and nitrogen gas was passed for 10 minutes. For the square wave voltammetry (SWV) method, a continuous potential was applied to the working electrode in the potential range of +0.0 V / + 1.5 V, and voltammograms were recorded for

each of the three pesticides in this potential range and calibration graphs were created. Electrochemical analyzes were performed at +0.75 V for Phosmet, +1.0 V for M. Parathion and +1.2 V for Ethion.

## 2.6. Preparation of calibration charts

The solutions were prepared by taking appropriate amounts of pesticide stock solutions (100.0 ppm) and diluting with methanol. In order to determine the linearity range, the standard solutions were added to the electrochemical cell at increasing concentrations and voltammograms were recorded. Calibration graphics were drawn by plotting the concentration versus peak current values. The smallest concentration that can be observed and the concentration ranges that can be quantified from the calibration graphs were determined and evaluated statistically.

## III. RESULTS

### 3.1. *Spirulina platensis* Culture Follow-up Results:

Laboratory-scale *Spirulina Platensis* culture was followed for 10 days in Hatay / Iskenderun in 2020 / September. Temperature, pH, and optical density values of *Spirulina Platensis* culture bottles are given in Table I.

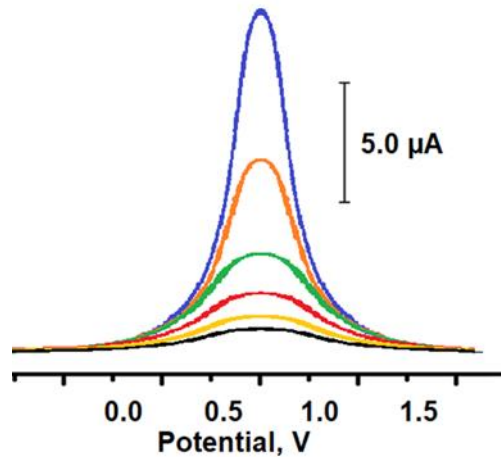
		Temperature (° C) Morning	Temperature (° C) Afternoon	pH	Absorbance (670 nm)
1.	Day	27.0	28.3	9.27	0.1876
2.	Day	28.7	28.3	9.40	0.5597
3.	Day	26.4	26.8	9.55	0.5950
4.	Day	25.2	26.4	9.60	0.5399
5.	Day	28.4	30.4	9.53	0.7660
6.	Day	25.7	27.8	9.65	0.7000
7.	Day	28.1	28.3	9.73	0.9463
8.	Day	26.8	28.7	9.76	0.9185
9.	Day	27.5	24.6	9.85	1.05226
10.	Day	26.3	28.7	9.87	1.1489

**Table I:** Temperature, pH, absorbance values of *Spirulina platensis* culture

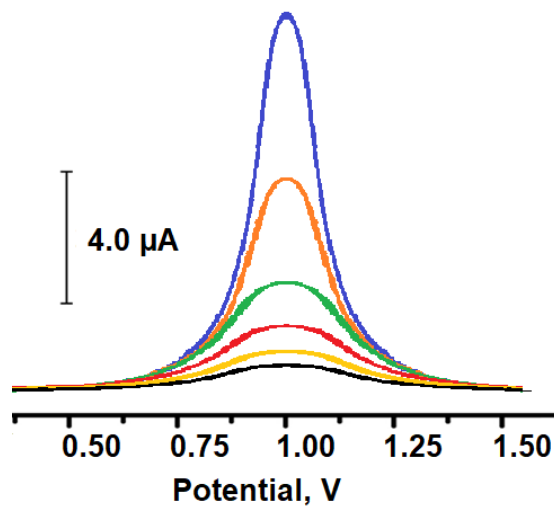
### 3.2. Voltammograms Recorded for All Three Pesticides for Electrochemical Determination:

The most suitable parameters were selected as 100 Hz for frequency, 5 mV for step height and 50 mV for pulse

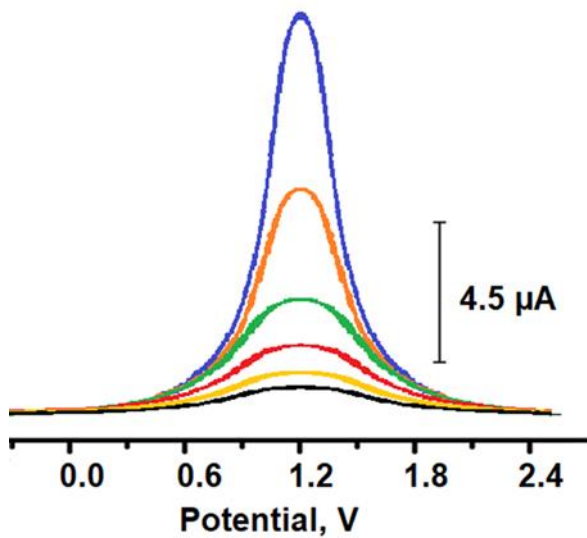
amplitude considering the peak shape and current values. Square wave voltammograms of all three pesticides at increasing concentrations in 0.1 M Phosphate buffer system are shown in Figures I, II and III.



**Figure I:** Square wave voltammograms of Phosmet (Smallest concentration: 0.5 ppm and Highest concentration: 10.0 ppm)

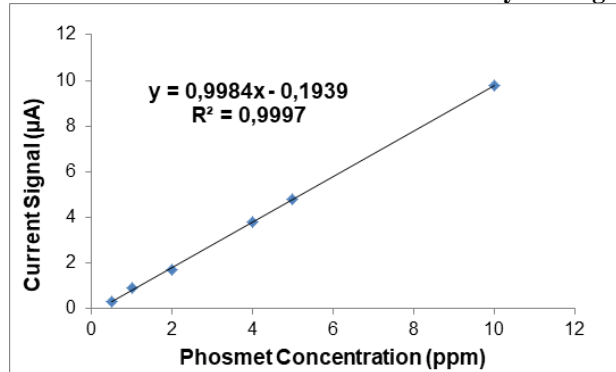


**Figure II:** Square wave voltammograms of Methyl Parathion (Smallest concentration: 0.5 ppm and Highest concentration: 10.0 ppm)

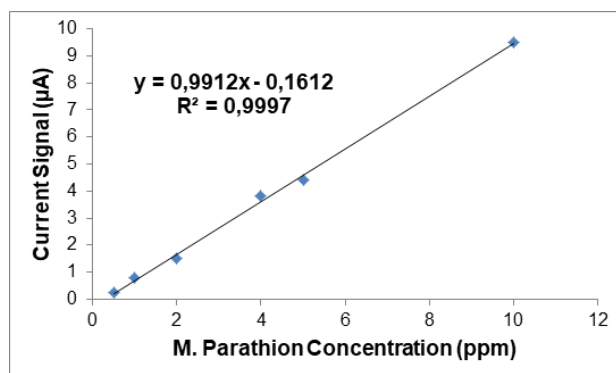


**Figure III:** Ethion's square wave voltammograms (Smallest concentration: 0.5 ppm and Highest concentration: 10.0 ppm)

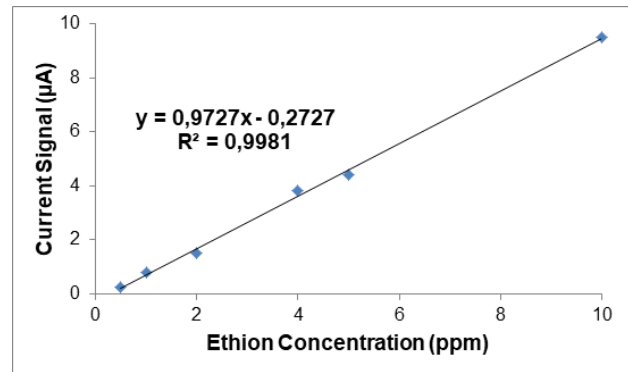
### 3.3 Calibration Curves and Linearity Ranges:



**Figure IV:** Calibration curve obtained by Phosmet's SWV method (n = 6)



**Figure V:** Calibration curve obtained by the SWV method of Methyl Parathion (n = 6)



**Figure 6:** Calibration curve obtained by Ethion's SWV method (n = 6)

### 3.4. Checking the significance of the calibration graphics for the pesticides used for nonlinearity:

The observed peak currents for all three pesticides were plotted against their concentration in order to create calibration curves using the SWV method and to determine the linearity intervals. Linear calibration curves were obtained for all three pesticides in the range of 0.50 - 10.00 ppm (Figure IV, Figure V, Figure VI). The significance of the separation from linearity of the calibration graphs obtained for all three pesticides was checked (Table II, Table III and Table IV).

For Phosmet  $F_H = 11659.05 > F_T = 5.59$ ; M. Parathion  $F_H = 12175.27 > F_T = 5.59$  and Ethion, values of  $F_H = 10972.77 > F_T = 5.59$  ( $p < 0.05$ ) were obtained and it was found that the departure from linearity was insignificant. In addition, by checking the significance of the correlation coefficients, these coefficients were found to be statistically significant values. (For Phosmet  $t_H = 98.12 > t_T = 2.48$ , for M. Parathion  $t_H = 93.42 > t_T = 2.48$ , for Ethion  $t_H = 88.23 > t_T = 2.48$ ,  $p < 0.05$ ). The pesticide results calculated by the SWV method in *Spirulina Platensis* culture (n = 6) are given in Table V.

Checking the Significance of the Correlation Coefficient						
n	r	Sr	$t_H$	$t_T$		
6	0.9997	$7.479 \times 10^{-3}$	98.12	2.48		
Nonlinearity Significance Control						
RKT	YOAKT	RAK T	RAK O	RKO	$F_H$	$F_T$
961178.13	955178.18	498.1	81.6	951378.18	11659.05	5.59

**Table II:** Statistical calculations for the correlation coefficient of the calibration curve obtained from Phosmet's analysis with SWV method and the significance control of the departure from linearity. (RKT Regression Sum of Squares; YOAKT Y Total of Squares Departed from the Mean;

RAKT Departure from Regression Sum of Squares; RAKO Regression Mean Square; FH Calculate F value found; FT Table F value)

Checking the Significance of the Correlation Coefficient						
n	r	Sr	$t_H$	$t_T$		
6	0.9997	$7.317 \times 10^{-3}$	93.42	2.48		
Nonlinearity Significance Control						
RKT	YOAKT	RAKT	RAKO	RKO	F <sub>H</sub>	F <sub>T</sub>
958364.18	952169.21	437.2	79.50	967934.22	12175.27	5.59

**Table III:** Statistical calculations for the significance control of separation from linearity with the correlation coefficient of the calibration curve obtained from the analysis of M. Parathion with the SWV method.

Checking the Significance of the Correlation Coefficient						
n	r	Sr	$t_H$	$t_T$		
6	0.9981	$7.217 \times 10^{-3}$	88.23	2.48		
Nonlinearity Significance Control						
RKT	YOAKT	RAKT	RAKO	RKO	F <sub>H</sub>	F <sub>T</sub>
954798.36	94397.21	387.6	83.1	911837.21	10972.77	5.59

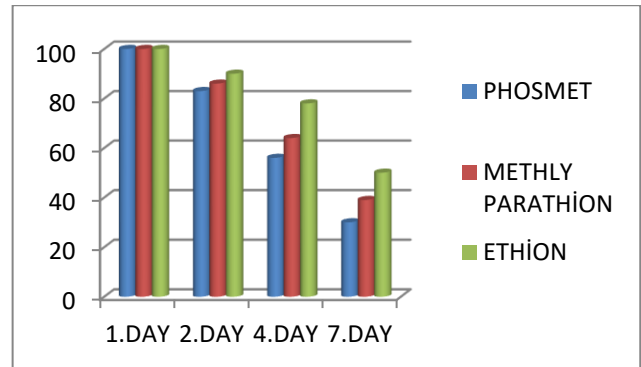
**Table IV:** Statistical calculations for the correlation coefficient of the calibration curve obtained from Ethion's analysis with the SWV method and the significance check of the departure from linearity.

DAY	Phosmet / ppm	M. Parathion / ppm	Ethion / ppm
1.	2.000	2.000	2.000
2.	1.665 ± 0.003	1.734 ± 0.004	1.816 ± 0.003
4.	1.137 ± 0.002	1.289 ± 0.002	1.579 ± 0.004
7.	0.615 ± 0.001	0.797 ± 0.001	1.017 ± 0.002

**Table V:** Calculated pesticide results in *Spirulina platensis* culture (n = 6)

### 3.5. Bioremediation rates of Phosmet, Ethion and Methly parathion of *Spirulina platensis*:

As a result of the 7-day bioremediation experiment performed with *Spirulina platensis* culture; The rates of *Spirulina platensis* to bioremediate Phosmet, Ethion and Methly parathion are given in Figure 6 as percentages. The starting dose of 2.0 ppm was calculated as 100%.



**Figure VI:** Percentage results of the bioremediation effect of the *Spirulina platensis* culture on the pesticides used in the experiment by days.

During the *spirulina platensis* culture stage, daily temperature and absorbance measurements show that the culture is reproduced in a laboratory scale by adapting to natural conditions. The 7-day pesticide results calculated by SWV method in *Spirulina Platensis* culture for 2 ppm Phosmet, Ethion and Methyl Parathion determined as the initial concentration (Table V) show that *Spirulina Platensis* is effective in pesticide removal. In the study, it was determined that *Spirulina platensis* caused Phosmet to bioremediate by 17.0 %, 44.0 % and 70.0 % on day 2, day 4 and day 7, respectively (Figure VI). According to the results, it can be said that the bioremediation effect for Phosmet is approximately 20.0% in both days compared to the days. It was concluded that Methyl parathion bioremediated with *Spirulina platensis* at a rate of 14.0% on the 2nd day, 36.0% on the 4th day and 61.0% on the 7th day. Ethion's bioremediation rates by *Spirulina platensis* were determined as 10.0 % on day 2, 22.0 % on day 4 and 50.0 % on day 7.

## IV. DISCUSSION

Pesticides occupy an indispensable place in modern agriculture. It is widely used in other industries such as agriculture and animal husbandry to improve crop production and boost economic growth. In addition, the indiscriminate use of pesticides can result in pesticide contamination by causing them to accumulate in the environment.

Pesticides can be removed by microorganisms in two ways. The first is by an active process that requires energy from the outside, and the second by the physicochemical

interaction (passive way) owing to the chemical structures in the cell wall. It is also known that different mechanisms play a role in pesticide removal by microalgae, including bioadsorption, bioaccumulation and biodegradation. Microalgae have biochemical properties that undertake oxygen-mediated photosynthesis. They can survive in less suitable conditions such as high salinity, heat, cold, various light sources, osmotic pressure, and anaerobiosis. During photosynthesis, algae cells convert solar energy into chemical energy, which leads to the production of bioactive compounds. The production of these bioactive compounds can be associated with microbial and algal growth.

Kurade et al. reported that Diazinon, a widely used organophosphate group pesticide, was bioremediated by green microalga *Chlorella vulgaris* at a rate of 98.0% [20]. Similarly, it has been reported that *Microcystis novaceki* can bioremediate Methyl parathion more than 90.0% [21]. Abdel-Razek et al found that *Chlorella vulgaris* and *Spirulina platensis* removed 99.0% of Malathion from wastewater [22]. It has been determined that *N. Muscorum*, *A. oryzae*, and *S. platensis* have the ability to use Malathion as a source of biodegradation and phosphorus at a rate of 91%, 65% and 54% respectively [23].

In a study in which five green algae species (*Scenedesmus sp. MM1*, *Scenedesmus sp. MM2*, *Chlamydomonas sp. Stichococcus sp.* and *Chlorella sp.*) were used to biodegrade Fenamiphos, it was found that all algae species have a high level of detoxification ability, and especially *Chlorella sp.* showed removal efficiency as 99% [24]. In a study in which the adaptation of freshwater cyanobacteria and microalgae developed using an experimental model to lindane resistance was investigated, it was determined that bioremediation based on lindane-resistant cells (eliminated up to 99%) is effective in cleaning lindane and other chlorinated organic contaminated habitats [25].

In a long-term study investigating the bioremediation potential of *Chlorella vulgaris* on Atrazine, Molinate, Simazine, Isoproturon, Propanil, Carbofuran, Dimethoate, Pendimethalin, Metoalcholar, Pyriproxin pesticide mixture, it has been reported that the presence of growing algae causes biological removal of pesticides. In the same study, it was observed that the rate of pesticide elimination varied between 87% and 96.5% [26].

In the study investigating the bioaccumulation and degradation of Prometryne by *Chlamydomonas reinhardtii*, it was determined that prometryne was degraded by *Chlamydomonas reinhardtii* and removed from the environment rapidly [27]. Similarly, it has been reported that *Chlamydomonas reinhardtii* has high ability to degrade fluroxypyr in aquatic ecosystems [28].

As a result of the study, we observed that the Microalgae bioremediation technology in Hatay / Iskenderun can be implemented under natural conditions without temperature limitation at laboratory scale. In our study, the percentage of biodegradation of *Spirulina platensis* on Phosmet,

Methyl Malathion and Ethion pesticides was calculated by electrochemical measurements using Square wave voltammetry (SWV) method.

*Spirulina platensis* culture bioremediated Phosmet pesticide by 17.0% on the 2nd day, 44.0% on the 4th day and 70.0% on the 7th day; Methyl Parathion pesticide was found to be 14.0% on the 2nd day, 36.0% on the 4th day and 61.0% on the 7th day, whereas Ethion bioremediated on the 2nd day by 10.0%, on the 4th day with 22.0% and on the 7th day with 50.0%.

## V.CONCLUSION

From these data, we can conclude that the degradation of Phosmet, Methyl Parathion and Ethion by *Spirulina platensis* is closely related to an efficient cleansing / detoxification system. The difference in bioremediation rates of three different pesticides applied in equal doses under the same experimental conditions indicates that *Spirulina platensis* has different tolerance values among pesticides. However, the exact mechanism remains to be investigated.

Green algae are known to have a great ability to accumulate various organic substances, including pesticides, in aquatic environments. With this study, we have demonstrated that *Spirulina Platensis* can be useful as a reclamation method designed to improve environmental stress tolerance against Phosmet, Methyl Parathion and Ethion or in the improvement of wastewater ecosystems.

In order to eliminate or minimize the harmful effects of Phosmet, Ethion and Methyl Paration, it is very valuable to use the environmentally friendly bioremediation technique in our study in terms of contributing to the nature and the future.

In order to evaluate the reason for the variability of the bioremediation percentage, we think that extending the implementation period in new studies to be planned and taking culture samples in different periods will shed light on further studies. The difference in bioremediation results for three different pesticides applied in equal doses under the same experimental conditions indicates that the bioremediation effect of *Spirulina platensis* has a different tolerance effect among pesticides.

## AUTHOR CONTRIBUTIONS

Özlem GÜL executed experimental studies and literature research within the scope of her thesis.

Bahar BANKOĞLU YOLA contributed to the establishment of the experimental setup and experimental studies.

Prof. Dr. Mehmet Lütfi YOLA and Assoc. Prof. Dr. Tuğba Raika KIRAN is the responsible authors of the study.

## REFERENCES

- [1] Maczulak AE. Pollution: Treating Environmental Toxins. New York: Infobase Publishing. ISBN: 9781438126333. 120, 2010.
- [2] Ward O, Singh A. Applied Bioremediation and Phytoremediation. Editors: Singh A, Ward OP. "Soil Bioremediation and Phytoremediation" Springer-Verlag Berlin Heidelberg, 1st edition, ISBN 978-3-662-05794-0.1,11,2004.
- [3] Uqab B, Mudasar S, Qayoom A, Nazir R. Bioremediation: a management tool. *Journal of Bioremediation & Biodegradation*, 7(2), 331,2016.
- [4] Gul UD, Yavuz SA. Pestisitler Kirlenmiş Ortamların Biyoremediasyonu. *Turkish Journal of Scientific Reviews*. 11 (1), 07-17, 2018.
- [5] Boudh S, Singh JS. Pesticide Contamination: Environmental Problems and Remediation Strategies. In: Bharagava R, Chowdhary P. (eds) Emerging and Eco-Friendly Approaches for Waste Management. Springer, Singapore. ISBN 978-981-10-8669-4, 245-269, 2019.
- [6] Varjani SJ, Agarwal AK, Gnansounou E, Gurnathan B. Introduction to Environmental Protection and Management. Editors: Varjani SJ, Agarwal AK, Gnansounou E, Gurnathan B. "Bioremediation: Applications for Environmental Protection and Management" Springer Nature Singapore Pte Ltd. Gateway East, Singapore, ISBN 978-981-10-7485-1, 1-7, 2018.
- [7] Jayaraj R, Megha P, Sreedev P. Organochlorine pesticides, their toxic effects on living organisms and their fate in the environment. *Interdiscip Toxicol*, 9(3-4), 90-100, 2016.
- [8] Smith A, Jong HM. Distribution of organochlorine pesticides in soils from South Korea. *Chemosphere* 43(2), 137-140, 2001.
- [9] Wasseling C, Aragón A, Castillo L, Corriols M, Chaverri F, De la Cruz E, Keifer M, Monge P, Partanen TJ, Ruepert C, Van Wendel de Joode B. Hazardous pesticides in Central America. *International Journal of Occupational and Environmental Health*, 7(4), 287-94, 2001.
- [10] McLellan J, Gupta SK, Kumar M. Feasibility of Using Bacterial-Microalgal Consortium for the Bioremediation of Organic Pesticides: Application Constraints and Future Prospects. In: Gupta SK, Bux F. (eds) Application of Microalgae in Wastewater Treatment. Springer, Cham. 341-349, 2019.
- [11] Bharagava RN, Saxena G, Mulla SI. Bioremediation of Industrial Waste for Environmental Safety Volume I: Industrial Waste and Its Management. Editors: Saxena G, Bharagava RN. "Introduction to Industrial Wastes Containing Organic and Inorganic Pollutants and Bioremediation Approaches for Environmental Management" Springer Nature Singapore Pte Ltd. ISBN 978-981-13-1891-7, 1-19, 2020.
- [12] Senthil Kumar P, Femina Carolin C, Varjani SJ. Pesticides bioremediation. In: Varjani SJ, Agarwal AK, Gnansounou E, Gurnathan B (eds) Bioremediation: applications for environmental protection and management. Springer Singapore, 197-222, 2018.
- [13] Ramanan R, Kim B-H, Cho D-H, Oh H-M, Kim H-S. Algae-bacteria interactions: evolution, ecology and emerging applications. *Biotechnol Adv*, 34(1), 14-29, 2016.
- [14] Singh S, Kate BN, Banerjee UC. Bioactive Compounds from Cyanobacteria and Microalgae: An Overview. *Critical Reviews in Biotechnology*, 25(3), 73-95, 2005.
- [15] Sisman AG, Oral R. Investigation of The Hormesis/Toxicity Potential of Manisa (Turkey) Urban Wastewater Treatment Plant by using *Selenastrum Capricornutum* Printz. *Fresenius Environmental Bulletin*, 23(5), 1183-1189, 2014.
- [16] Zhou T, Wang J, Zheng H, Wu X, Wang Y, Liu M, Liu Y. Characterization of additional zinc ions on the growth, biochemical composition and photosynthetic performance from *Spirulina platensis*. *Bioresource Technology*, 269, 285-291, 2018.
- [17] FAO Specifications and Evaluations for Agricultural Pesticide Phosmet. 2-20, 2019. Available from: <http://www.fao.org/3/CA2760EN/ca2760en.pdf>
- [18] Edwards F, Tchounwou P. Environmental Toxicology and Health Effects Associated with Methyl Parathion Exposure – A Scientific Review. *International Journal of Environmental Research and Public Health*, 2(3), 430-441, 2005.
- [19] Foster RLJ, Kwan B H, Vancov T. Microbial degradation of the organophosphate pesticide, Ethion. *FEMS Microbiology Letters*, 240(1), 49-53, 2004.
- [20] Kurade KB, Kim JR, Govindwar SP, Jeon BH. Insights into microalgae mediated biodegradation of diazinon by *Chlorella vulgaris*: Microalgal tolerance to xenobiotic pollutants and metabolism. *Algal Research*, 20, 126-134, 2016.
- [21] Fioravante IA, Barbosa FAR, Augusti R, Magalhães SMS. Removal of methyl parathion by cyanobacteria *Microcystis novacekii* under culture conditions. *Journal of Environmental Monitoring*, 12(6), 1302, 2010.
- [22] Abdel-Razek MA, Abozeid AM, Eltholth MM, Abouelenien FA, El-Midany SA, Moustafa NY, Mohamed RA. Bioremediation of a pesticide and selected heavy metals in wastewater from various sources using a consortium of microalgae and cyanobacteria. *Slov Vet Res*, 56 (22), 61-73, 2019.
- [23] Ibrahim WM, Karam MA, El-Shahat RM, Adwa AA. Biodegradation and Utilization of Organophosphorus Pesticide Malathion by Cyanobacteria. *Hindawi BioMed Research International*. 392682. 2014.
- [24] Cáceres T, Megharaj M, Naidu R. Toxicity and transformation of fenamiphos and its metabolites by two micro algae *Pseudokirchneriella subcapitata* and *Chlorococcum* sp. *Science of the Total Environment*, 398 (1), 53-59, 2008.
- [25] González R, García-Balboa C, Rouco M, Lopez-Rodas V, Costas E. Adaptation of microalgae to lindane: a new approach for bioremediation. *Aquat. Toxicol*. 109 (109), 25-32, 2012.
- [26] Mishra ESI. Biosorption potential of the microchlorophyte *Chlorella vulgaris* for some pesticides. *Journal of Fertilizers & Pesticides*, 8 (1), 5, 2017.
- [27] Jin ZP, Luo K, Zhang S, Zheng Q, Yang H. Bioaccumulation and catabolism of prometryne in green algae. *Chemosphere*, 87 (3), 278-284, 2012.
- [28] Zhang S, Qiu CB, Zhou Y, Jin ZP, Yang H. Bioaccumulation and degradation of pesticide fluroxypyr are associated with toxic tolerance in green alga *Chlamydomonas reinhardtii*. *Ecotoxicology*, 20 (2), 337-347, 2011.