An-Najah National University

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ALGAE FOR NUTRIENTS REMOVAL FROM WASTEWATER: THE APPLICATION OF ARTIFICIAL LIGHT FOR ENHANCED ALGAL GROWTH

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Dedication

I dedicate this thesis to my support and the light of my eyes in this life, my dear father.

To the one who lights my path with her sincere prayers, my dear mother.

To my brothers and sisters, the source of inspiration and encouragement.

To my small family, my husband, my other half, my life partner and companion.

To my daughters, the secret of my happiness, the light of my eyes and the beat of my heart.

To the proud Gaza, to the proud Gaza that does not know defeat and surrender.

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My family, my friends, to every person who left a beautiful mark on my scientific journey and supported me to overcome difficulties and obstacles, and encouraged me, to everyone who helped me move forward and increased my determination to succeed in this scientific achievement, thank you from the heart.

Declaration

I, the undersigned, declare that I submitted the thesis entitled:

ALGAE FOR NUTRIENTS REMOVAL FROM WASTEWATER: THE APPLICATION OF ARTIFICIAL LIGHT FOR ENHANCED ALGAL GROWTH

I declare that the work provided in this thesis, unless otherwise referenced, is the researcher's own work, and has not been submitted elsewhere for any other degree or qualification.

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Date: **10/10/2024**

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Abstract

Treating municipal wastewater using algae to remove nutrients is considered one of the most successful, environmentally friendly, and inexpensive natural methods, as algae grow naturally in wastewater if the appropriate conditions are available for its growth, such as nutrients, temperature, duration, and intensity of exposure to light, as well as the presence of bacteria that form a symbiotic relationship with algae to help it grow. This research studied the effect of artificial light on the growth of algae and its activity in removing nutrients from municipal wastewater.

Three beakers of municipal wastewater in which the algae grew, with a volume of 5 liters as a pilot scale of patch reactors, were used and placed in the same conditions which were Temperatures and mixing for 15 days, with varying durations of exposure to artificial light. In the first sample, the algae was exposed to light 24 hours a day and was called the light sample. The second beaker was exposed to light for 19 hours, the fifth was exposed to natural daylight and was called half light sample, and the third sample remained in the day and night cycle, where it was called the natural sample.

After 15 days of comparing the three samples, although the results were close in the two light and half-light samples, the algae in the light sample were the most efficient in removing nutrients from wastewater, as their efficiency in removing COD was (95%) and phosphorus was (96%). (99.8%) ammonium, TSS concentration 737.4 mg/L, and chlorophyll A pigment concentration of 129.4 μg/L. In the half-light sample, the effectiveness of algae in removing COD was (93%), phosphorus was (94%), ammonium was (99.8%), TSS concentration was 237.4 mg/L, and chlorophyll A concentration was 57.2 μg/L. Finally, in the natural sample, the efficiency in removing COD was (93%), phosphorus was (6%), ammonium was (56%), TSS concentration was 174 mg/L, and chlorophyll A concentration was 8 μg/L. These results show the effectiveness of algae as a natural method for removing nutrients from wastewater and the positive effect of light on improving algae growth and its efficiency in the treatment process.

Keywords: Algae; Wastewater Treatment; Nutrients Removal; Artificial Light; Algae Growth.

Chapter One

Introduction and Theoretical Background

1.1 General Background

Water scarcity is a serious global issue that challenges humans. Despite extensive research on water scarcity from a global to a local scale, this problem is exacerbated in urban areas such as large cities, where population growth, industrial and urban development all hurt the quantity and quality of available water (Li et al., 2020; Srinivasan et al., 2013). This calls for the need to purify and treat polluted water resulting from various human activities, so the goal of wastewater treatment is the removal of biochemical oxygen demand (BOD), chemical oxygen demand (COD), suspended solids, nutrients (NO_3^--N, NO_2^--N, NH_4^+-N, and PO_4^{3-}-P), coliform bacteria, and toxicity (Abdel-Raouf et al., 2012; Gerba & Pepper, 2015). The main steps in the wastewater treatment process are preliminary treatment, primary treatment, and secondary treatment (Mohsenpour et al., 2021; Sonune & Ghate, 2004). The effluent from the primary and secondary treatment processes still contains considerable nitrogen, phosphorus, and pathogens. This method, which acts as a tertiary treatment step, will increase the dissolved oxygen content, which helps to reduce pathogens present in the treated wastewater, especially in anaerobic wastewater treatment, where nitrogen and phosphorus are absorbed by algae (Rathod, 2015).

Algae and bacteria coexist in wastewater. Microalgae are classified as phototrophs; they need light as an energy source and carbon dioxide to produce carbohydrates and ATP (Oregon State University, 2011). For that reason, the algae consume the carbon dioxide produced by the bacteria for the process of photosynthesis, and in a reciprocal relationship, the algae produce the oxygen that the bacteria consume in the process of decomposing organic matter in the wastewater (Tang et al., 2016).

The growth of algae in the wastewater treatment process depends on various parameters, such as physical, chemical, and biological. Other parameters can also affect algae growth, such as mixing, dilution rate, depth, addition of bicarbonate, and harvesting frequency (Schnurr & Allen, 2015).

A new technique for treating municipal wastewater through algal-bacterial symbiosis interaction to reduce nutrient content will be studied in this research by applying artificial light to the process in the absence of daylight, studying the effect of this artificial light on algae growth and nutrient removal efficiency, and comparing these results with daylight results.

1.2 Objectives

The main objectives of this research are:

1. To study the effect of the application of artificial light during the night on the removal of nutrients and COD from municipal wastewater.
2. To study the recommended period for the algae exposure to artificial light.
3. To determine the efficiency of nutrients and COD removal by using algal technology with different periods of exposure to artificial light.

1.3 Literature Review

1.3.1 Municipal Wastewater

Water shortages in Palestine and everywhere in the world urge the replacement of the demand for freshwater for agriculture with alternative sources and new, less expensive, and environmentally friendly technologies. The global solution to the problem of water shortage is the treatment of wastewater, which is classified into four categories: domestic wastewater resulting from residences and commercial institutions, industrial wastewater from industries (Kataki et al., 2021; Sonune & Ghate, 2004), infiltration/inflow, and stormwater resulting from flooding due to rainfall (Panasiuk et al., 2015). The definition of municipal wastewater or sewage is a mixture of household waste, which can be dissolved or suspended materials. Therefore, high-quality sewage service must be provided to urban areas with increased population density and increased production of household waste. This requires careful planning and a solid infrastructure. Wastewater treatment processes affect public health and the environment. They also produce problems that are difficult to solve, such as sludge, and require a lot of energy to operate (Abusoglu et al., 2012).

In Palestine, sewage networks exist and serve the main cities and urban areas partially or completely, and some West Bank camps are also connected to sewage collection networks. In rural areas, sewage networks serve less than 30 towns partially, while the majority dispose of their wastewater through unlined pits. In the Gaza Strip, the infrastructure situation is poor concerning sewage disposal, as more than a third of the generated wastewater is discharged into sewage collection tanks, open drains, and pits. There are always trends to improve the sewage system and drainage networks in Palestine, by expanding the spread and investment in new sewage networks, performing periodic maintenance on them, and connecting them to homes so that they include all residential areas in the country. These trends are carried out under the management of municipalities and large facilities in cooperation with the Palestinian Water Authority (PWA, 2013).

1.3.2 Municipal wastewater treatment

The main objective of municipal wastewater treatment has always been to reduce its content of suspended solids, oxygen-demanding materials, dissolved inorganic compounds, and harmful organisms (Sikosana et al., 2019), as well as remove the nutrients (Wang et al., 2017). With the development and increase in research and studies, additional treatment processes have emerged to improve the means of waste disposal in municipal wastewater. The basic methods of municipal wastewater treatment are divided into four stages: preliminary treatment, the coarse solids found in raw wastewater and large materials are removed in this process. These solids and large materials contain paper, plastics, cloth, with fecal matter (Sonune & Ghate, 2004); primary treatment, beginning with degranulation, screening, grinding, and sedimentation, tanks of sedimentation usually circular or rectangular with slopped bottoms, its function is to remove settleable solids and floating materials, also this process helps decrease the load on secondary treatment units, and it removes about 40 percent of biochemical oxygen demand (Guyer, 2018); secondary treatment, which involves the oxidation of dissolved organic matter by the use of biologically active sludge, which is then filtered, in this stage occur biodegradation process, which means the decomposing of suspended solids by microorganisms, and the reduction of pathogens. The wastewater resulting from primary treatment undergoes biological treatment by different methods like aeration tanks, sewage lagoons, or in a flow-through filter, which can remove approximately (85%) of BOD and approximately (90%) COD, the ratio between BOD/COD in wastewater is 0.5. At the end of secondary treatment typically a disinfection step is applied (Gerba & Pepper, 2015); and tertiary treatment, where advanced biological methods are used to remove nitrogen as well as chemical and physical methods such as granular filtration, activated carbon adsorption (Gerba & Pepper, 2015), UV disinfection, ultrafiltration systems and sedimentation (Illueca-Muñoz et al., 2008).

1.3.3 Growth of algae

Since the need for wastewater treatment and reuse began, studies and research have been concerned with finding low-cost, easy-to-implement, and environmentally friendly treatment methods. For that, wastewater treatment with algae has been studied as a method with a more environmentally friendly approach (Abdel-Raouf et al., 2012). Algae are classified as plants. They are aquatic plants that grow in water and wet places. Some are terrestrial, and all of them are autotrophs. They make their food because they contain chloroplasts, which can be unicellular or multicellular. Algae are a special group of plants because they do not grow true roots, stems, or leaves (Sahoo & Baweja, 2015). The growth of algae in the wastewater treatment process depends on various parameters, such as physical factors like light and temperature, chemical factors like the availability of nutrients and carbon dioxide, and biological factors like symbiosis with bacteria and virus infections. Also mixing rate, bicarbonate existing, depth, and harvesting rate of algae, all these factors can affect on growth of algae in wastewater (Gatamaneni et al., 2018; Rathod, 2015). Algae and bacteria are present together in wastewater, and the interactions between them are of great importance, as bacteria affect the growth of algae by both stimulation and inhibition. Also, bacteria affect the algal biomass, its formation, the removal of nutrients, and the determination of its presence. Some bacteria secrete enzymes such as glucosides, chitinases, and cellulases, which work to break down the algal cell wall, and thus the algae are decomposed, then bacteria absorb the compounds which are produced from algae cell decomposition, This is known as the "survival competition" between algae and bacteria. (Saravanan et al., 2021).

Algae are a member of the plant family. They are organisms that grow in water and depend on photosynthesis for their nutrition. Their cells do not contain stems, roots, or leaves (Bajpai, 2019). The symbiotic relationship between algae and bacteria is important in the wastewater treatment process and photosynthesis in algae, as it has a positive effect and is an important factor for completing this process, through gas exchange, as algae consume carbon dioxide resulting from the aerobic respiration of bacteria when decomposing organic matter, while algae produce the oxygen needed by bacteria, thus increasing the effectiveness of algae in removing nutrients from wastewater, including nitrogen and phosphorus. Studies have shown the possibility of using harvested algae biomass in the production of biogas (Wang et al., 2018). The gas exchange process between algae and bacteria and the symbiotic relationship between them falls under the photoactivated sludge (PAS) system, which is based on the symbiosis of algae and bacteria in wastewater, where algae provide oxygen gas that bacteria need to breathe without the need for external air, this system works to enhance the growth of algae in the wastewater (Mohamed et al., 2021).

Stirring is one of the factors that has an impact on the growth of algae and its efficiency in treating wastewater. It works to enhance the growth of biomass in the algae and ensure homogeneity in the aquatic environment and the distribution of nutrients in a way that facilitates the algae's access to them and their removal from the water. It is necessary to know the appropriate stirring rate to apply in the water treatment process because rapid stirring negatively affects the algae cells. The gentle stirring process is important in that it works to increase the distribution of gases so that they are easily exchanged in the water between microorganisms and prevents the sedimentation of algae, so they remain stuck in the tank, which ensures easy it provides access to nutrients and thus increases its efficiency in treating wastewater (Isiya & Sani, 2020).

Algae in wastewater go through different life phases depending on the surrounding conditions, such as the availability of nutrients, light, and carbon dioxide. In the beginning, the algae begin to grow through the process of photosynthesis and consuming organic materials and nutrients from nitrogen and phosphorus needed to build the cell. The growth period continues until the nutrients are removed from the aquatic environment, but after a period of time, the algae must be harvested to avoid overgrowth because it negatively affects the treatment system. Also, the presence of algae cells increases the concentration of suspended materials in the water which negatively affects the environment, especially when using water for irrigation of crops, especially drip irrigation, because algae cells can lead to blockage of the drippers. Algae are similar to other organisms, they have a life cycle in which they go through stages from the beginning of their formation to the period of decomposition, and their life cycle begins with the lag stage, which is the formation stage or the initial period where they grow slowly in this stage, the second stage is the stage of increasing biomass where the cells divide and grow rapidly and this is called the exponential period, after a period of time the nutrients begin to decrease due to their consumption by the algae, so the division and reproduction of the cells becomes limited and this is called the period of declining relative growth, then the stationary stage begins, where the nutrients are close to being finished and the biomass does not increase and the growth of the cells becomes slow down, in this stage algae reach to maximum growth, and when the nutrients are removed, especially nitrogen and phosphorus, the algae enter the death stage where the algae cells decompose and their activity ends, and when the algae cells decompose, it release nitrogen and phosphorus into the water, and when the appropriate conditions are available, the algae cells return to grow again (Farag, 2013).

1.3.4 Temperature

Algae are important organisms that treat wastewater from nutrients. It is an effective, environmentally friendly and economically inexpensive method. There are several factors that play an important role in the growth and activity of algae in the wastewater treatment process. These factors include the temperature of the surrounding environment and the photoperiod. Studies have proven the effect of temperature on the growth of algae. Very high temperatures and very low temperatures have a negative effect and lead to a reduction in the growth rate of algae. Short light periods also negatively affect the activity of algae cells. Temperatures and photoperiod vary according to the seasons. In the summer, temperatures are high and the day is long, which means a longer light period. Conversely, temperatures are low in the winter and the photoperiod is shorter. Algae grow better and their effectiveness increases in the summer. This depends on the type of algae, but in general, algae cells grow and increase in warm temperatures. Temperatures between 20 and 30 °C are considered suitable for providing a good environment for photosynthesis and the growth of algae cells, which increases the release of oxygen consumed by bacteria and thus increases their activity along with algae. This works to increase Its effectiveness in wastewater treatment and nutrient removal. Although algae need warm temperatures to be active, alternating temperature regimes are best for algae activity and increasing their efficiency in removing organic compounds, nitrogen, and phosphorus compounds from wastewater (Xu et al., 2021). But when using a constant temperature system in the wastewater in which algae grow, especially *Chlorella vulgaris*, the optimal temperature is approximately 30 °C, as this temperature plays an important role in increasing the production of algae biomass in addition to the presence of carbon dioxide and its effect on the production and increase of the content of the green pigment chlorophyll (Chinnasamy et al., 2009).

1.3.5 Carbon Dioxide

Since algae are considered plants and autotrophs, one of the main factors for their growth and survival is the availability of carbon dioxide in an appropriate concentration, as algae consume carbon dioxide as a source of carbon in the process of photosynthesis, which its mechanism is represented by algae absorbing carbon dioxide with water. In addition to light energy, glucose and oxygen are produced. The success of the photosynthesis process has an important impact on increasing the biomass of algae and thus increasing their efficiency in treating wastewater and removing nutrients. Therefore, maintaining an appropriate concentration of carbon dioxide ensures an increase in the ability of algae to grow and enhances its efficiency in the treatment process (Nguyen et al., 2022). The sources of carbon dioxide in wastewater are multiple, the easiest and most important of which is atmospheric air, where algae absorb carbon dioxide present in the atmospheric air, especially during the mixing process, as mixing allows the exchange of gases. It is also possible to obtain carbon dioxide through dissolved organic matter in wastewater when it decomposes. If carbonates and bicarbonates are present in wastewater, they contribute to increasing the concentration of carbon dioxide when they interact (Abraham et al., 2023). The process of aerobic respiration is carried out by bacteria through the decomposition of organic matter in wastewater, as illustrated by the following reaction:

$$Organic\ Compounds + O_2 \xrightarrow{Aerobic\ Bacteria} CO_2 + H_2O + Energy \qquad (1)$$

This process is considered a good source of carbon dioxide, as it is produced from the decomposition of these organic materials, and this explains the symbiotic relationship between bacteria and algae in the wastewater treatment process (Fallahi et al., 2021).

1.3.6 pH

Another factor that affects algae growth is pH, which is a measure of how acidic or basic the water is. The pH value is measured from 1.0 to 14.0, where a value from 1.0 to 7.0 indicates that the medium is acidic, and from 7.0 to 14.0 indicates that the medium is basic. The effect of algae on pH varies according to their type. Algae tend to be active in a neutral medium. The process of photosynthesis in algae is affected by the pH value, as the presence of the necessary nutrients and the appropriate temperature resulting from sunlight activates the process of photosynthesis and algae consume carbon dioxide, which works to raise the pH value. pH imbalance significantly affects ammonia balance and thus nitrogen, which is important for algae metabolism, is affected, as are carbon dioxide levels and uptake, which affects the vital functions of algae including enzyme and protein activity (Alkhamis et al., 2022). The pH value is related to the level of carbon dioxide, the higher the concentration of carbon dioxide in the water, the higher the pH value, which positively affects the activity of algae and the photosynthesis process necessary for their growth, and the lower the concentration of carbon dioxide causes a decrease in pH, which inhibits the activity of algae (Rodríguez-Torres et al., 2021). It is essential to maintain a suitable pH during the wastewater treatment process to ensure that algae grow properly in a suitable environment and increase their activity and efficiency in removing nutrients during the treatment process.

1.3.7 Salinity

Salinity is defined as the amount of salts contained in soil or water. Salts are able to dissolve in water medium and are transported with the movement of water. The amount of salinity of water is affected by temperature. The higher the temperature, the more soluble salts are in the water. The amount or degree of salinity of water can be measured as electrical conductivity (EC) or total dissolved solids (TDS) (Srivastava et al., 2021; Velmurugan et al., 2020). Inorganic dissolved salts such as sodium and magnesium salts are responsible for the salinity of wastewater, which has multiple sources, such as industrial sources and agricultural sources, some of these industries such as pharmaceuticals, textiles, petrochemicals and other various industries (Srivastava et al., 2021).

In the aquatic environment, salinity of water affects on algae growth and activity, although algae cells have the ability to adapt to different salinity levels by controlling the osmotic pressure inside the cell, algae are affected by the salinity of the medium, as salinity affects the metabolic processes of the algal cell. Studies have shown that the salinity concentration tolerated by algae varies depending on the type of algae present in the aquatic medium. In general, algae can tolerate a salinity level of up to approximately 50 mg/L, as salinity below this concentration stimulates the growth of algal cells and their components, while salinity levels of up to 300 mg/L are considered lethal to algal cells (Al-Enazi, 2020), so the imbalance in the salinity level from the appropriate value negatively affects the chemical composition of the cell and its components, such as fats, carotenoids, steroids, carbohydrates and sterols, and when salts in the water are ionized, the imbalance in ions affects the permeability of the algal cell membrane. At high or low salinity levels below the permissible level, algae are forced to adapt to changes in the surrounding environment in order to continue their growth and activity, so they work to regulate the photosynthesis process responsible for energy production and regulate its consumption, and also work to reduce the formation of cell protein, and control the fluidity of the membrane in a manner consistent with the environment to relieve pressure on the algal cell (Novosel et al., 2022).

1.3.8 Total Suspended Solids

Total suspended solids (TSS) are considered important indicators of water quality, as their increased concentration in water hinders the growth of microorganisms used in wastewater treatment processes, because they may contain substances that cause toxicity to microorganisms, and cause turbidity in the water, as they are known as non-settled solid particles that remain suspended in the aquatic medium (Verma et al., 2013; Zhuang et al., 2019).

Suspended solids affect the aquatic environment and microorganisms, including algae. The biomass of bacteria in wastewater is expressed as COD or volatile suspended solids (VSS), while algae are components of (TSS) in wastewater, and are affected by high concentrations of suspended solids. When the concentration of suspended materials in wastewater increases, the photosynthesis process of algae is negatively affected, because suspended solids block light from the aquatic environment, and they also hinder the gas exchange process in the aquatic medium, which prevents algae from consuming carbon dioxide necessary for the photosynthesis process so that their cells grow and reproduce. Suspended solids have the ability to absorb nutrients from the aquatic medium, which leads to a deficiency of these materials that algae feed on. These suspended materials can also cause toxicity to algae and microorganisms because they can transport pollutants with their particles (Bilotta & Brazier, 2008). Total suspended solids concentration is an indicator of water quality, and its concentration is measured by filtration, oven drying, and weighing before and after drying (Adjovu et al., 2023).

1.3.9 Nutrients

Nutrients are a very important parameter for algae growth. These nutrients contain especially the elements phosphorus (P), nitrogen (N), and carbon (C), along with micronutrients like iron (Fe) and silicon (Si), in addition to sulfur, potassium, and magnesium (Bajpai et al., 2014).

1.3.9.1 Phosphorus

Phosphorus is a non-metallic element that is chemically active in terms of reactions. It is located in the fifth group of the periodic table. Its symbol is P, and its atomic number is 15. It is found in nature as phosphate because it is very reactive and cannot be a pure, free element. It has two forms: white phosphorus and red phosphorus (*Phosphorus - Wikipedia*, n.d.). Phosphorus plays an important role in the human body and is essential in teeth, bones, and cell membranes. It also helps regulate the function of the heart, nerves, and muscles, maintains normal blood acidity, and activates various enzymes. It is obtained from many foods and is available as a nutritional supplement (Chan & Uribarri, 2017). Phosphorus (P), whether organic or inorganic, is important for the balance of aquatic ecosystems. It is considered a nutrient for photoautotrophic organisms. Some genes are classified and regulated in some way by knowing their phosphate content (Li et al., 2022). Algae need phosphorus to grow, which is considered an essential compound for algae metabolic processes, photosynthesis, energy transfer, and signaling. It also plays an important role in the production of fats and amino acids. Phosphorus is necessary to produce cellular components in algae, such as RNA, DNA, and phospholipids. Phosphorus must be available at a rate of (0.03–0.06%) to ensure proper algae growth, constituting approximately (1%) of the total mass of algae (Yaakob et al., 2021).

The concentration of phosphorus in domestic wastewater varies depending on the source of this element. Its main sources are human waste, household detergents, and liquid waste resulting from some industrial and commercial facilities. The nature of the agricultural area also affects the concentration of phosphorus. As is known, phosphorus is an essential element in agricultural fertilizers, and therefore the concentration of phosphorus increases in wastewater resulting from areas where there is a lot of agricultural activity (Kroiss et al., 2011).

1.3.9.2 Nitrogen

Nitrogen is a gas with no color or odor and exists as a molecule consisting of the bonding of two atoms of the element N_2. Nitrogen gas is one of the components of the air and constitutes approximately (78%) of the atmosphere. Nitrogen is a nonmetal located in group five of the periodic table. It has the symbol N and the atomic number 7 (*Nitrogen - Wikipedia*, n.d.). Nitrogen is important in our lives as it is involved in the formation of the nucleic acids DNA and RNA, which are the molecules that carry genetic information in the cells of all living organisms. Nitrogen is essential in the formation of amino acids, which seem to form the basic building blocks of proteins. In plants, when there is a lack of nitrogen, the plant becomes unable to make the proteins necessary for the growth of plant cells, and thus a problem occurs in the growth of the plant. When nitrogen is increased in the plant, it increases the biomass, so we notice an increase in the growth of leaves and stems. Therefore, it is important to know the concentration of nitrogen necessary for each type of plant, as this element is essential in the composition of agricultural fertilizers, and when its concentration increases, it can lead to the poisoning of animals that feed on fertilized plants (Aczel, 2019).

In the human body, nitrogen is important in the formation of tissue proteins and many compounds involved in hormones, neurotransmitters, the immune system, and antioxidant defenses. Therefore, the concentration of nitrogen in the body must be within the normal range in terms of quantity and quality to ensure that the body performs its normal functions (Tessari, 2007). Nitrogen is considered one of the essential nutrients for algae growth in wastewater treatment processes using algae, as it feeds on these elements and helps reduce their concentration and remove them from wastewater. Nitrogen greatly affects the biochemical composition of algae due to its important role in the synthesis of proteins, lipids, and carbohydrates. The higher the nitrogen concentration, the greater the algae growth, and the lower the concentration in the culture medium, the lower the growth and the higher the lipid production. Nitrogen can be present in wastewater in the form of nitrate, nitrite, urea, and ammonium, but in algae seeding operations, nitrate (NO_3^-) and ammonia salts (NH_4^+) are used, and it is common to use nitrate in culture media because it is more stable (Yaakob et al., 2021). Sources of nitrogen in domestic wastewater are human waste, chemical detergents, and agricultural fertilizers (Berger et al., 2022).

1.3.9.3 Heavy metals

1.3.9.3.1 Iron

Iron is a chemical element with the symbol Fe. It is found in nature in abundance, as it is the fourth largest in terms of quantity as an element present on the surface of the Earth. It is of great importance in most mineral industries and is also included in the pharmaceutical and agricultural industries (Silver, 1993). It is one of the heavy elements that play an important role in the production of energy and the transport of gases, including oxygen. It is involved in the formation of DNA in the cells of algae and microorganisms, which makes it important in the process of photosynthesis and the formation of the green pigment chlorophyll. It is considered one of the nutrients that algae need to grow and develop, but the presence of iron should not exceed a low concentration approximately 50 ppm (Zada et al., 2021) so as not to negatively affect the life of the algae. Hence the importance of iron in producing biomass for algae cells and improving the treatment process (Blanco-Vieites et al., 2022).

1.3.9.3.2 Silicon

Silicon (Si) is a chemical element located in the fourth group in the periodic table. This explains why silicon combines the properties of metals and non-metal materials. Silicon has many uses in various industries, such as the manufacture of solar cells, the manufacture of computers, and other electronic devices (Cheng et al., 2016). Silicon is considered a micronutrient that has an effect on microorganisms in the aquatic environment, especially algae. Its effect can be positive or negative. Algae can absorb silicon molecules and accumulate them inside the cells. Therefore, high concentrations of this element increase the pressure on algae cells, which leads to their stress, imbalance, and damage to their membrane. Consequently, their absorption of light necessary for photosynthesis decreases, and the level of chlorophyll A pigment decreases. This negatively affects the growth of algae and increases their biomass and can cause their poisoning and death. As for the positive effects of silicon on algae, it improves the photosynthesis process in the cell by increasing the content of the green chlorophyll pigment, providing structural support for the algal cell and improving its hardness, which increases algae growth and thus increases its efficiency in the wastewater treatment process. Therefore, it is necessary to maintain an appropriate concentration of silicon to which algae are exposed in wastewater during the treatment process, optimum concentration of silicon depends on type of algae species in wastewater, but in general the concentration of silicon should not exceed about 50-100 mg/L (Shariati & Shirazi, 2019).

1.3.9.3.3 Magnesium

Magnesium is a chemical element with a symbol (Mg), located in the second group in the periodic table and it's a metallic element. The energy fields are one of the important applications in which the element magnesium is included. It has a unique structural composition that makes it able to replace or complement other metals in the fields of energy conversion and storage (Shin et al., 2023). Magnesium is also widely used in the aerospace and aerospace fields due to its hardness, strength, and ease of manufacture, so it is used in rockets, aircraft engines, and helicopters. Also, because of the flexibility and low density of magnesium, it is used in the automobile industry, as it has good castability (Froes et al., 1998).

In the wastewater field, the presence of magnesium compounds in wastewater can negatively and positively affect algae growth and reproduction. On the positive side, magnesium ions improve algae growth and reproduction in wastewater because they are considered one of the nutrients that help algae increase biomass and increase their activity and vitality. On the other hand, during the process of sag and coagulation in the treatment process, when the pH rises, magnesium hydroxide precipitates form, and on their way to precipitate, they drag the algae cells with them. Thus, the algae precipitate and are removed from the water along with other sediments. This explains the effect of magnesium ions on the life cycle of algae in wastewater (Semerjian & Ayoub, 2003), algae need about (5-15 mg/L) of magnesium for biomass production and for protein and carbohydrates which is necessary for algae cells (Salman et al., 2023).

1.3.9.3.4 Sulfur

Sulfur is a chemical element located in group six of the periodic table. It has symbol S, its atomic number is 16. It is a non-metallic element. It has the ability to form many compounds in combination with other elements and has multiple uses in various aspects of life. One of its common compounds is carbon dioxide gas, which is released into the atmosphere and causes acid rain, which is known to have a negative impact on the environment, humans, and plant life. (Francioso et al., 2020). Sulfur has many uses and is important in the agricultural field, as it enters into the composition of vitamins that are important for plants and soil, and it also works to fix nitrogen in the soil to improve agricultural production. Sulfur is also used as a soil fertilizer because of its importance in forming amino acids necessary for plants and improving the process of photosynthesis (Lucheta & Lambais, 2012). Sulfur is widely distributed worldwide and is often used in the form of acids. It is used in many industrial fields, such as water treatment, fertilizer manufacturing, pesticides and fungicides, detergents, fireworks, metal manufacturing, and steel analysis, and also in the chemical and pharmaceutical industries such as cosmetics and personal care products. It is also used in the manufacture of high-quality, corrosion-resistant and frost-resistant concrete, and other various industrial and agricultural applications (Netherlands Organisation for Applied Scientific Research, 2017b). Sulfur improve the growth of algae in wastewater, where it is considered a micronutrient for them. It is an important nutrient for photosynthesis and the formation of essential amino acids such as cysteine and methionine, which are needed to make protein in the algae cell. Sulfur also helps increase the biomass of algae cells, which leads to an increase in the efficiency of algae in removing nitrogen and phosphorus from wastewater. It is possible that recycling of the sulfur element occurs during the treatment process, as hydrogen sulfide gas is released from anaerobic digestion processes, so the algae use this gas as a food source, and this works to provide the sulfur element permanently, although sulfur is considered an essential nutrient for the growth of algae cells. However, an imbalance in its concentration at high or low levels affects the efficiency of algae growth, in the study (Hughes et al., 2018), showed that the appropriate concentration of Na^2SO_4 for algae growth in wastewater is 304 μM.

1.3.9.3.5 Potassium

Potassium is a metallic element located in the first group of the periodic table. It has the symbol K and the atomic number 19. One of its characteristics is that its color is silvery white, and it oxidizes quickly, as it loses its luster immediately upon oxidation and exposure to air. The best way to preserve it is with oil or grease (Tiecke, 2019). Potassium is of great importance in many industrial fields and in aspects of plant and animal life. It is used in the form of potassium oxide in the respiratory equipment. Its use is also widespread as an essential element in plant fertilizers and fireworks. It is also a necessary element that humans and animals depend on because it is part of the various body fluids that help the body's systems perform their functions. Potassium is an important element in the human body, as it affects the functions and efficiency of cells, especially nerve cells. In industrial fields, its salts and compounds are widely used in bread making, medicines and medical preparations, glass making, detergents, photography, leather tanning, preservatives in the food industry, water treatment, and other various industries (Netherlands Organisation for Applied Scientific Research, 2017a). In the wastewater treatment process, it is essential to maintain the appropriate concentration level of potassium because of its great importance in this process, as it affects the growth of algae cells in wastewater, and is considered one of the essential macronutrients that work to grow algae. Potassium improves the photosynthesis process of algae by activating enzymes such as the rubisco enzyme responsible for this process, and maintaining the osmotic balance in the algae cell by regulating the process of water absorption and turgor pressure. Osmotic balance is of great importance in helping algae deal with the pressures and environmental changes surrounding them, in order to improve cell growth and increase their efficiency. Among the environmental pressures that algae may be exposed to, such as increased salinity and drought, potassium also helps increase biomass by regulating the process of cell division and increasing their growth, which leads to increasing the efficiency of algae in the wastewater treatment process (Xu & Pan, 2020), studies showed that optimum amount of potassium which needed for enhanced algae growth in wastewater depends on the type of algae species of algae, but in general, it is found that (0.3 wt%) of potassium is useful for growth of algae cell, and addition of 1.0 g/L of NPK fertilizer is considered the optimum for growth of some types of algae (Mtaki et al., 2021).

1.3.10 Chemical Oxygen Demand (COD)

COD is chemical oxygen demand and BOD is biochemical oxygen demand, both are indicators of water quality, COD measures the amount of dissolved oxygen in water needed to oxidize total organic matter in water, while BOD measures the amount of oxygen consumed by bacteria in the process of aerobic respiration to decompose organic compounds in water, this is the difference between COD and BOD. The decomposition of organic matter produces carbon dioxide and water, it is a source of carbon dioxide and increases its concentration in water (Panawala, 2018). This leads to a negative impact on the nature and quality of water. The presence of nutrients and organic carbon upsets the ecological balance, reduces biodiversity, and causes an unpleasant odor. As the concentration of nutrients and organic carbon in wastewater increases, the water becomes more toxic (Cai et al., 2013). Wastewater treatment using algal-bacterial symbioses is an environmentally effective way to remove nutrients, especially nitrogen and COD. Oxygen, which results from the photosynthesis process carried out by algae, is consumed by bacteria to oxidize organic carbon present in the water. From oxidation, carbon dioxide gas is produced, which is consumed by algae again in the process of photosynthesis, through which nitrogen is absorbed and synthesized within the algae biomass. Through these periodically repeated processes, nitrogen and organic carbon are removed, and thus wastewater is treated with symbiosis between algae and bacteria (Lee et al., 2016).

1.3.11 Chlorophyll Pigment

Chlorophyll A is the green pigment responsible for the green color in plants and some microorganisms, such as algae and some bacteria. Chlorophyll A has the ability to essentially absorb light energy from the sun and convert it into chemical energy so that plants can carry out photosynthesis to make food. The light energy absorbed by this pigment falls within the wavelengths in the blue and red regions of the electromagnetic spectrum. The molecular structure of the pigment chlorophyll A allows it to interact with water and amino acids, and this helps it in the process of transmitting light energy and working efficiently during photosynthesis. The chlorophyll A molecule consists mainly of a porphyrin ring and a magnesium atom in the center, with a phytol tail attached to the thylakoid membrane. It also includes its composition of functional groups allows it to interact with various solvents (Björn et al., 2009).

The green chlorophyll pigment in plants and organisms that depend on photosynthesis to provide their food, especially algae, consists of another part in addition to chlorophyll A, which is chlorophyll B, whose function is to convert the photons of light energy that it absorbs into chemical energy that plants use in the process of photosynthesis to make food and grow. Chlorophyll B is characterized by its ability to absorb light photons with a wavelength longer than those absorbed by chlorophyll A, which leads to improving the process of photosynthesis and increasing its efficiency by absorbing a wider range of wavelengths of light through chlorophyll A and B. Also, one of the most important features of chlorophyll B is that it can adapt to solar radiation, so that its peak absorption of light corresponds to the spectral distribution of sunlight, which makes plants adapt well to different lighting conditions and increases the efficiency of the light absorption process (Kume et al., 2018).

Algae contain the chlorophyll pigment, which is responsible for the green colour of algae. It also has an essential role in the process of photosynthesis, and it consists of two types: chlorophyll A and chlorophyll B. Photons of light are projected onto the chlorophyll molecule in the algae to stimulate it, causing some of this light energy to be reflected and transmitted and some to be absorbed to be converted into chemical energy used in growth and biomass production. Chlorophyll B is the pigment responsible for absorbing light photons and transmitting them to chlorophyll a, the main participant in the process of photosynthesis. Chlorophyll B absorbs light with a wavelength between 410 - 480 nm and 600 - 685 nm, while chlorophyll a needs to absorb light with a wavelength between 380 - 450 nm and 600 - 670 nm (Kommareddy & Anderson, 2003).

1.3.12 Light

The type of algae used to remove nutrients from wastewater is green algae (Chlorophyceae). As mentioned earlier, algae are microorganisms and are also considered members of the plant family because they are autotrophs that make their food through photosynthesis, a physicochemical process in which autotrophs use light and carbon dioxide to convert them into energy for oxidation and reduction. Therefore, light is an important element in the work of algae, as it is a major source of energy in food production. Light propagates in the form of waves, or photons (Kommareddy & Anderson, 2003).

Sunlight is the natural source of light that algae use to grow. The amount of light intensity affects the growth process of algae and the way it coexists with bacteria. Also, the processes of photosynthesis and oxygen production in algae are affected by the intensity of light. Light intensity is one of the important factors that must be provided to algae at an appropriate value, because it is important in improving the efficiency of the wastewater treatment process, as it increases the growth of algae and improves the efficiency of the photosynthesis process by providing the necessary light, which leads to high efficiency in removing nutrients from wastewater and maintaining the ecological balance between algae and bacteria in the aquatic environment (Lin et al., 2019; Nzayisenga et al., 2020). Light intensity also plays a role in the formation of amino acids in the algal cell and affects their composition, as well as stimulates the content of fats and carotenoids (Maltsev et al., 2021). Another important factor that affects the growth and efficiency of algae is the duration of exposure to light. The longer the light period that algae are exposed to, the more efficient the photosynthesis process is, which depends primarily on light, which leads to improving the efficiency of removing nutrients from wastewater during the treatment process using algae (Lee et al., 2015). Using light with alternating cycles of light and dark has a good effect on stimulating the photosynthesis process, increasing the biomass of algae, and reducing energy consumption. Also, maintaining the provision of light under appropriate conditions ensures the healthy growth of algae cells and prevents their stress. In some cases, the high density of algae cells can obstruct the access of light to other cells and thus inhibit the growth process. Therefore, an appropriate density of cells must be maintained so that the environment is healthy for growth and reproduction (Maltsev et al., 2021).

1.3.13 Previous studies

Based on Abdel-Raouf et al. (2012), the use of algae in wastewater treatment processes was studied in this resaerch as a tertiary treatment process, as the primary and secondary treatment of wastewater resulting from various activities such as agriculture, domestic, and industrial uses occurs in which oxidation of organic materials and filtering of suspended materials occur. Some materials are not removed through primary and secondary treatment, such as nitrogen, phosphorus, and some heavy metals. These elements become harmful to the environment if their concentration exceeds the permissible limit. Thus, it was necessary to find a way to remove these elements from the treated water. This study concluded that the use of algae is of great importance in water treatment. Since increasing the production of biomass in algae and increasing its growth requires consuming nitrogen and phosphorus elements from the aquatic medium, the process of treating wastewater using algae is considered environmentally friendly and economically inexpensive compared to other treatment processes. (Abdel-Raouf et al., 2012).

Based on Zimmo et al. (2000), in this research, the efficiency of algae and duckweed containers and the difference between them in removing nutrients, especially nitrogen, from wastewater was studied. The time needed for this experiment was 15 days, where nitrogen was used at different concentrations. The nitrogen concentration in the first time was 50 mg nitrogen/L and 100 mg nitrogen/L in the second time. Applying nitrogen at different concentrations led to differences in environmental conditions between the containers in the experiments. The study showed that the dissolved oxygen (DO) concentration was higher in the algae-based container than in the duckweed container. Also, the pH was higher in the algae-based container than in the duckweed container, which means that the efficiency of algae is higher in removing nitrogen from wastewater. This is because the duckweed plants stay on the surface of water, in the container of duckweed, which leads to block sunlight from the algae and thus reduces the efficiency of the photosynthesis process. As for nitrogen removal, between (42%) and (62%) of the total nitrogen was removed from the duckweed based containers, and between (56%) and (95%) of the Kjeldahl nitrogen. The nitrogen removal rate was between (45%) in the algae-based containers as a treatment and (48%), and (48%) and (58%) for Kjeldahl nitrogen. This study proved that the difference in environmental conditions between containers based on duckweed and containers based on algae has an importatnt impact on the mechanism of nitrogen removal from wastewater (Zimmo et al., 2000).

According to Sayara et al. (2021), the effect of the symbiotic relationship between algae and bacteria on removing nutrients from wastewater was studied, as a 600 L sequensing batch photobioreactor (SBR) was used, distributed over three equal basins. After a 10 day incubation period, the effect of the symbiotic relationship between algae and bacteria was positive in improving the removal of nutrients from wastewater and in reducing greenhouse gas emissions, as the aerobic respiration of bacteria produces carbon dioxide, which the algae consume in the process of photosynthesis. Another result of this study was that (60%) of COD, (40%) of nitrogen (NH_4^+-N), and (90%) of phosphorus were removed, as the dissolved oxygen concentration during daylight periods was 6 mg/L (Sayara et al., 2021).

Based on Sayara et al. (2021), the effectiveness of retention time in the algae reaction was studied by feeding the reactors with the secondary flow of domestic wastewater and working under natural daylight/dark cycles. The results showed that the reaction time (RT) has a major role in the performance of the entire process, as the increase in algae biomass concentration was directly proportional to the increase in retention time. Thus, the COD removal rates increased significantly with increasing in (RT) value, also the phosphorus removal rates increased when RTs increased. Also, ammonium (NH_4^+-N) removal rates increased when RT increased (Sayara et al., 2021), that means, light is an important factor in algae growth and affects the efficancy of nutrients removal from wastewater.

According to Nzayisenga et al. (2020), this study investigated the effect of light intensity on the growth of microorganisms in aquatic media, especially algae. Different types of algae were used in wastewater, where four types were grown and exposed to three different values of light intensity (50, 150, 300 µmol/m².s). The study concluded that the fatty acid content in algae increased in the four types of algae, and the biomass of algae increased with increasing light intensity, thus improving algae growth and increasing their efficiency in removing nutrients from wastewater. The nitrogen and phosphorus content in wastewater decreased by more than (75%). It was concluded from this study that the higher the light intensity to which algae are exposed in wastewater, the better their growth and the more efficient they are in the treatment process. (Nzayisenga et al., 2020).

Based on Lee et al. (2015), this study investigated the effect of photoperiod on algae growth and efficiency in wastewater treatment. Three periods of light were studied: 12 hours: 12 hours; 36 hours: 12 hours; and 60 hours: 12 hours of dark/light cycles. This study concluded that, after 12 days, carbon concentration rates decreased in all photoperiods and were 72.3, 56.0, and 35.2 mg/L in the periods 12 hours: 12 hours, 36 hours: 12 hours, and 60 hours: 12 hours Respectively, the concentrations of nitrogen and phosphorus also decreased during the three periods: nitrogen concentration decreased to 4.8, 14.0, and 25.6 mg/L, and phosphorus concentration decreased to 0.6, 1.7, and 3.0 mg/L. The results of the study also showed that photoperiods affected the amount of biomass. After 6 days, the longest photoperiod to which the algae was exposed was the period in which biomass increased and the concentration of chlorophyll a increased (Lee et al., 2015).

According to Zimmo et al. (2002), this study was based on evaluating the differences in environmental conditions and their impact on the efficiency of wastewater treatment in two systems: ponds based on duckweed and ponds based on algae. This study lasted for 12 months, during which two systems were fed, each consisting of four equal basins, with a constant flow of partially treated wastewater. Among the results of this study, pH values were higher in ponds based on algae. Also, the concentration of dissolved oxygen (DO) was higher in ponds based on algae, so duckweed was more efficient than algae in removing organic matter (BOD) and TSS. As for fecal coliforms, their removal was higher in algae-based ponds. As for nitrogen removal, algae proved effective in removing (80%) of the nitrogen present in wastewater compared to duckweed, which removed

(55%) of the nitrogen. This study showed the effect of different environmental conditions in the two systems on the efficiency of wastewater treatment (Zimmo et al., 2002).

According to Nguyen et al. (2022), this study was based on a comparison between algae based wastewater treatment and bacteria based wastewater treatment. Among the results of this study is that the use of algae in wastewater treatment requires pretreatment for wastewater. Also, algae does not work at low nutrient concentrations because the efficiency of algae in their remedial work depends on the production of biomass, so they need nutrients for growth and cell reproduction. Algae also depend on carbon dioxide, which they obtain mainly from a symbiotic relationship with bacteria. It needs a retention time (HRT) of more than 10 days, compared to bacteria that need a few hours. Algae also depend on absorbing light, which increases their efficiency in the treatment process (Nguyen et al., 2022).

Based on Whitton et al. (2019), this research studied how the light types and its wavelengths affect the efficiency of algae in the form of beads in removing nutrients from wastewater during a reaction period of 3 hours. White light (400-700 nm) with a photon flux efficiency of 200 µmol/m^2.s, blue light with a wavelength of (465 nm), and red light (660 nm) were used. The study concluded that algae proved to be highly effective and efficient when white light was used in removing phosphate from wastewater, while the phosphate concentration decreased by 10.7 mg P/h per bead. When blue light was used, the phosphate concentration decreased by 10.2 mg P/h per bead, while when red light was used, the phosphate concentration decreased by 10.1 mg P/h per bead. The results concluded that white light is the best for improving algae growth and increasing their efficiency in the wastewater treatment process, because white light has the ability to cover a larger illumination area, as it has the ability to penetrate deeper (Whitton et al., 2019).

Based on Oruganti et al. (2022), this research investigated the effect of mixing rate on the efficiency of algae in the wastewater treatment process, as it is considered an important factor among the many factors that help improve the wastewater treatment process and remove nutrients from it through algae and bacteria. The presence of microorganisms in wastewater is affected by many factors, including pH, light intensity, temperature, availability of nutrients, and mixing rate. The study proved that the appropriate mixing rate ensures that microorganisms obtain the nutrients necessary for their growth in the aquatic medium. Thus, the importance of mixing lies in the fact that it is responsible for distributing nutrients in the aquatic environment, which facilitates their consumption to build algae and bacterial cells. Mixing also prevents the sedimentation of biomass by distributing it so that it remains suspended in the medium, to facilitate the interaction process between algae and bacteria and ensure the arrival of the light necessary for photosynthesis to the cells. Since bacteria and algae live in a symbiotic relationship based on the exchange of oxygen and carbon dioxide according to each other's needs, the mixing rate plays an important role in this relationship, as it allows gas exchange to ensure good ventilation and for the bacteria to obtain the oxygen needed for respiration, which in turn produces carbon dioxide that the algae consume for photosynthesis, which increases their efficiency in removing nutrients from wastewater (Oruganti et al., 2022).

According to Fan et al. (2021), in this research the nature of the symbiotic relationship between algae and bacteria and the effect of available light intensity on the domestic wastewater treatment process were studied. In light of this coexistence and aggregation of bacterial algae granular sludge (MBGS), the appropriate light intensity improves the growth of algae and increases its cells by stimulating the photosynthesis process which is based on algae consuming nutrients and removing them from wastewater. Light intensity also stimulates the vital activities of MBGS, which enhances cooperation between algae and bacteria in removing nutrients and treating water, as light intensity positively affects oxygen production in the MBGS system., which ensures balance and integrated work between algae and bacteria. According to the results of this research, when the light intensity was increased from 70 to 210 μmol/m^2.s, the efficiency of removing nutrients from wastewater increased, as (70.5%) of COD, (80.7%) Ammonia, and (73.9%) Phosphate were removed (Fan et al., 2021).

Based on Priyadarshi (2021), the effect of using different concentrations of algae on removing nutrients from wastewater was studied to find the best concentration that improved the efficiency of nutrient removal. The same conditions were provided in all stages, and six different *Chlorella vulgaris* algae types were used. In the first stage, (30%) and (40%) of algae were used. In the second stage, (20%) and (25%) of algae were used. In the third stage, (35%) and (45%) of the algae were used by fixing the temperature and neglecting the rate of evaporation. The experiment retention time is 18 days using a batch reactor. The results of this research showed that (30%) algae concentration was the optimal concentration for removing nutrients from wastewater, as (81%) of COD, (75%) nitrate, (99%) phosphorus, and (99%) ammonia were removed (Priyadarshi, 2021).

Based on Nguyen et al. (2020), this study investigated the extent to which the different inoculation rates of algae and bacteria affect the efficiency of removing nutrients from wastewater and the nature and conditions of the symbiotic relationship between microalgae and activated sludge, as the inoculation rate plays an important role in the metabolism and growth processes of microorganisms and affects the interaction between these organisms and their adaptation in aqueous media. The ideal inoculation ratio ensures suitable environmental conditions for the growth of microorganisms in wastewater and facilitates their work in water treatment, removing nutrients and increasing biomass production, which ensures a highly efficient treatment process. One of the findings from this study was that a mixing ratio of 3:1 microalgae:bacteria in photobioreactor during 11 days was optimum, with (99%) of COD, (79%) TP, and (86%) TN removed (Nguyen et al., 2020)**.**

From the literature review, the benefit and effectiveness of algae are evident in using them as a method to treat wastewater and remove nutrients from it, especially nitrogen and phosphorus, and adopting them as a tertiary treatment step, as algae consume nitrogen and phosphorus to build cells, increase biomass, and produce the energy they need to be active in photosynthesis. Literature review and previous studies were used to identify the factors affecting algae growth and life cycle and the parameters necessary to design the experiment in this research, such as the algae life cycle, appropriate temperature, and the necessary mixing rate. However, the existing gap is that previous studies are few and insufficient regarding the effect of light, its intensity, and duration of exposure to it on algae and their effectiveness and activity in removing nutrients from wastewater during the treatment process.

Chapter Two

Materials and Methods

In this research, nutrients in municipal wastewater were removed by algae. Artificial light was used with different dark/light cycles, and a comparison was made to find out the best cycle in this process to increase the efficiency of the algae in removing nutrients.

2.1 Materials

Materials and conditions that were needed in the experiment of algae in wastewater treatment and nutrient removal by applying artificial light:

2.1.1 Wastewater samples

Municipal wastewater samples were taken in July from two sites; the first 15 Liters were taken from duckweed basins at BierZeit University in Ramallah, as shown in Figure (1). The second part of the wastewater was taken from primary sedimentation and secondary treatment at Wastewater Treatment Plant Nablus West.

Figure 1

Duckweed basin at BierZeit University in Ramallah

2.1.2 Light control cabinates

Two closed wooden cabinates were built with doors. The base is square, with a side length of 40 cm and a height of 50 cm. Two cabinates were used to control the lighting system used for algae growth in wastewater.

2.1.3 Reactors

Three beakers with a capacity of 5 L each were used as reactors. In this research, batch and semi-batch reactor systems were needed, whereas in the first period of the experiment, a semi-batch system was used for the reactor. At this stage, the algae seeding process was carried out, and later, this system was also used during the algae growth stage. Batch system was used later in the wastewater treatment stage for nutrient removal.

2.1.4 Light system

As mentioned previously, light is an important factor in algae growth, and in this research, it was used to study its effect on the effectiveness of algae in removing nutrients from wastewater. Double LED light strips of visible light with a wavelength of (380 nm–700 nm) with intensity (760 lux) (14.46 μmol/m².s) (Lighting, n.d.) were used, fixed inside the wooden boxes on both sides and on the top lid of the boxes.

2.1.5 Stirring system

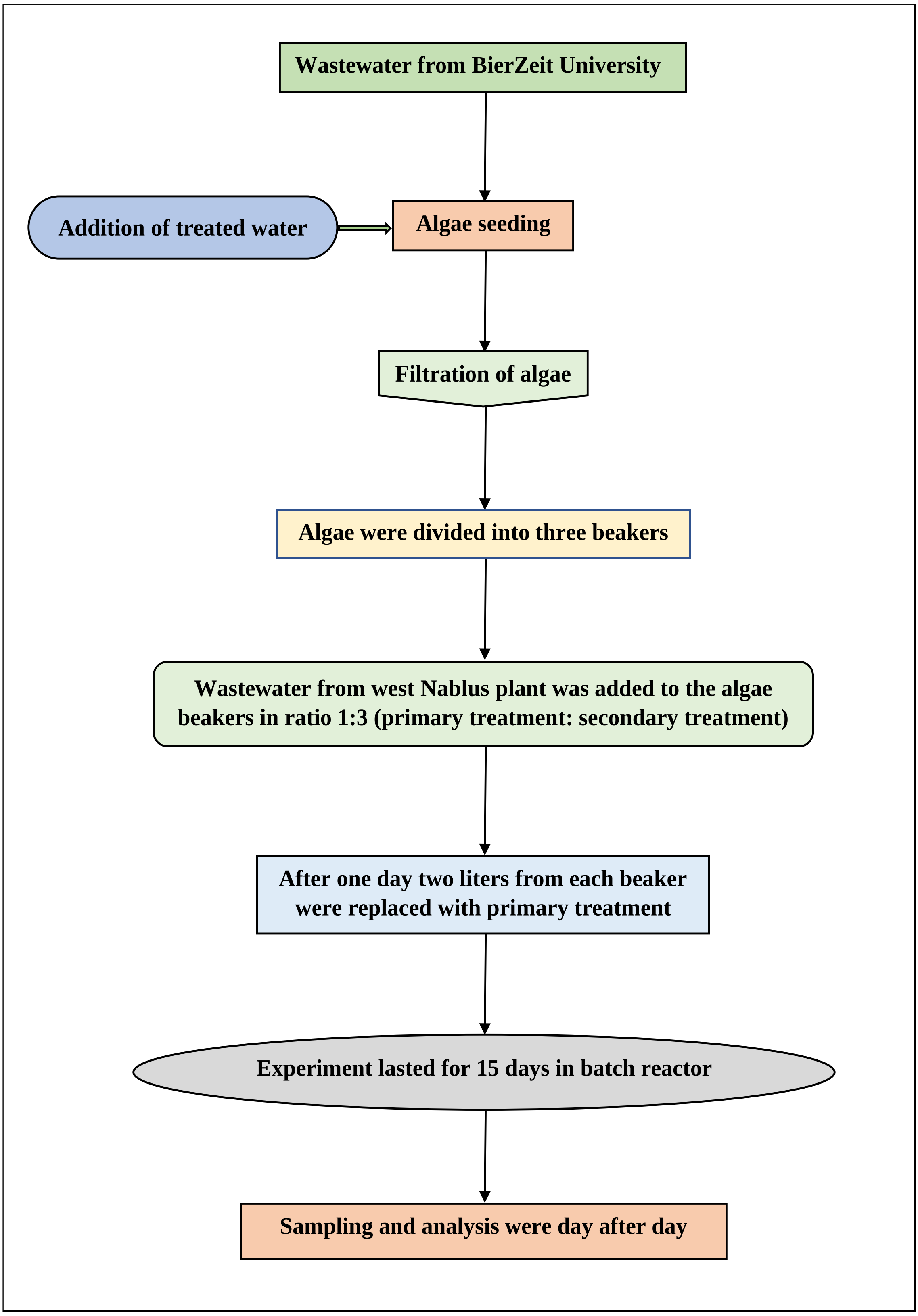
A magnetic stirrer apparatus was used for the stirring process with rate (100 rpm).

2.2 Methodology

The methodology in this study was divided into three steps, which are algae seeding, where the algae cells were grown and their biomass increased until they were ready for the treatment process, the second step was to determine the appropriate mixing ratio between primary treatment water and secondary treatment water, and the third was to use the algae in the process of treating wastewater and removing nutrients from it. The Figure (2) describes the methodology.

Figure 2

A Flow Chart describing general methodology



2.4.1 Algae seeding

As mentioned earlier, 15 Liters of wastewater were taken from duckweed basins at BierZeit University in Ramallah, and were filtered from duckweed, the wastewater was divided into three beakers with a capacity of 5 L for each. Duckweed is a type of algae that needs low concentrations of nutrients to grow in wastewater. It can remove heavy metals from wastewater through a process called phytoremediation. The treatment process using duckweed depends on several factors, such as the concentration of heavy metals in water and exposure time (Ali et al., 2015). At first, the samples of wastewater were filtered to remove duckweed species as shown in Figure (3.a).

After two days, 300 ml of treated water was added for each beaker, to encourage algae growth, and samples were put under artificial light with mixing in range 100 rpm, treated wastewater was added every two days for three times, the Table (1) shows the characteristics and nutrient content of treated water which was used to encourage algae growth in wastewater.

Table 1

Characteristics of treated wastewater

Parameter	Concentration (ppm)
Total Nitrogen (TN)	120
Ammonia (NH_3)	110
Phosphorus (PO_4^{-3})	25

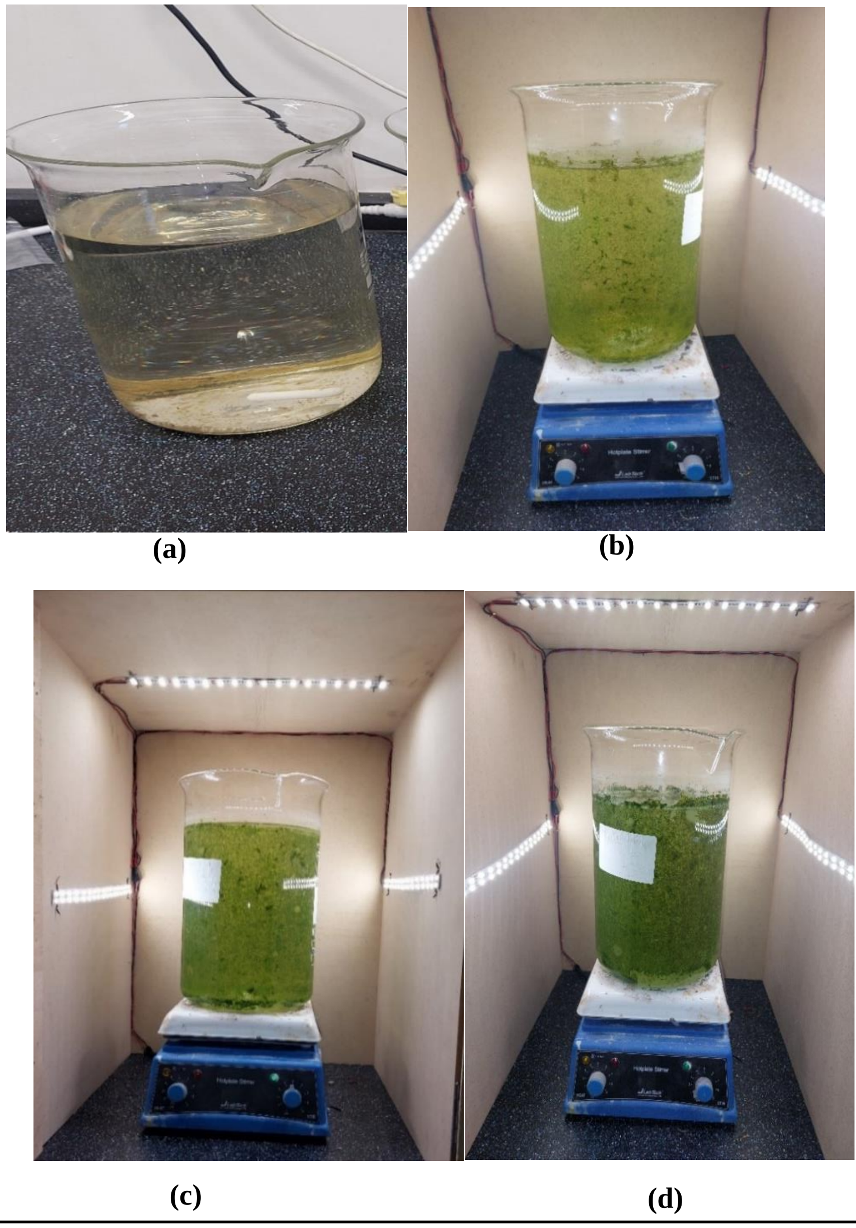
Two days after the first addition of treated water, the algae began to grow and this was evident by the appearance of its cells in the water and the color of the water in the beakers changed, as shown in Figure (3.b), at this time, the second addition of treated wastewater was added to the beakers.

After two days from second addition, the number of algae cells increased, and the color became darker due to the green chlorophyll pigment in the algae cells, as shown in Figure (3.c), and at this time, the third addition of treated wastewater was added to the beakers.

During the two days after the third addition, although the number of algae cells increased, they were in the process of decay as their color began to change to brown and the chlorophyll pigment in the cells decreased, which means that their activity decreased, they lost their vitality, they became unhealthy, and their life cycle began to end, this appears in Figure (3.d).

Figure 3

Algae seeding in wastewater sample



2.5 Effect of mixing ratio of primary and secondary treatment on algae growth

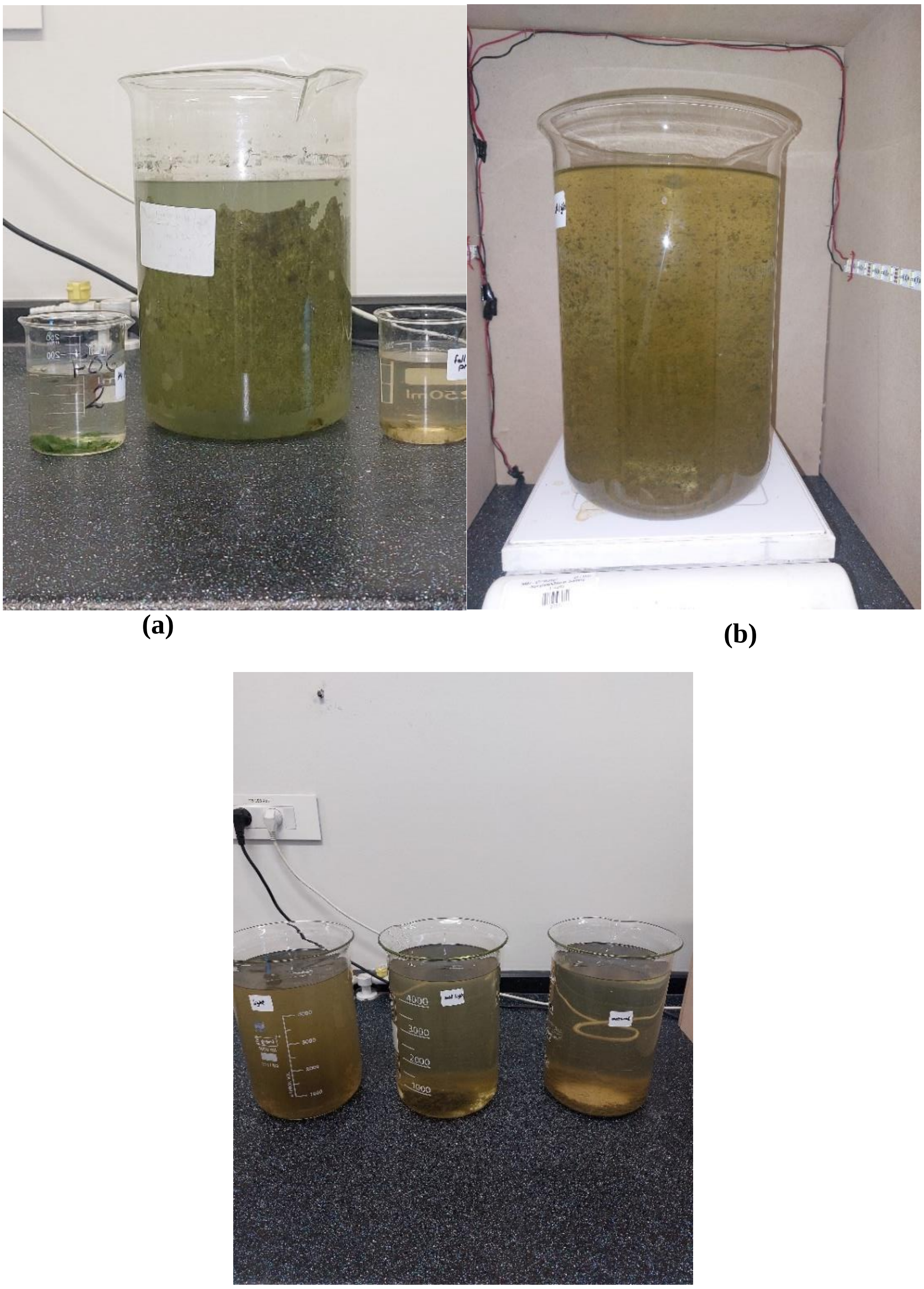
Algae seeding is the stage of stimulating algae to grow in wastewater and preparing it for use in experiments on the domestic wastewater treatment process. After the algae seeding stage was completed, a sample of algae cells was taken and distributed into two small beakers, each containing wastewater that would be used later in the experiment, which was taken from the Wastewater Treatment Plant Nablus West. Water samples were placed in different proportions of water from the primary and secondary treatments. The first beaker contained only water resulting from the primary treatment stage, and the second beaker contained a ratio of 1:3 (primary treatment: secondary treatment). This step was an initial experiment to study the behavior of algae in new water samples, to avoid shock to the algae when changing its environment, and to know the best mixing ratio between the primary treatment samples and the secondary treatment that should be started.

Figure (4.a) shows that the algae in the first beaker (at the right), which contains a full sample of water from the primary treatment stage, was in shock and dead because this water contains high levels of nutrients and other substances. While the second beaker (at the left) contained a 1:3 ratio (primary treatment: secondary treatment), the algae was healthy and regained its activity. The algae were filtered and distributed into three beakers, each with a capacity of 5 liters. The wastewater used on the first day was a 1:3 ratio (primary treatment: secondary treatment), and this ratio was used as a first step as previously mentioned to avoid causing shock to the algae cells. Therefore, each beaker was filled with 1250 mL of wastewater from primary treatment and 3750 mL from secondary treatment, as shown in Figure (4.b).

On the second day, the beakers were placed until algae settled to the bottom, after which 2 liters of water in each beaker were replaced with primary treatment water, thus increasing the percentage of primary treatment water in each beaker to approximately more than half, as shown in Figure (4.c), at this time, preliminary analyzes were carried out to determine the initial concentrations of nutrients.

Figure 4

Effect of mixing ratio of primary and secondary treatment on algae growth



(a) **(b)**

(c)

2.6 Treatment by algae

In this project, the experiment was carried out in the laboratories of An-Najah National University by using beakers as pilot-scale batch and semi-batch reactors. The batch system was automated as follows: 3 hours mixing period in range 100 rpm, under constant laboratory temperature at 28 C, for 15 days, three samples with algae; the first was left until the algae grew naturally in daylight while monitoring its activity and the progress of the water treatment process in this case; the second was treated like the first sample, but with artificial light at night to encourage the algae to grow and continue its activity in the absence of daylight; and the third sample was treated with artificial light in the partial system (19 hours under light and 5 hours in natural daily light). Samples of wastewater were taken every day after day and analysis were done in laboratory, and results were recorded. A comparison of the results from the three samples was made, all tests were done in the laboratories of An-Najah National University in the Water and Environmental Studies Institute (WESI) and were been by the standards as per Standard Methods for Water and Wastewater Examination (Brandi & Wilson-Wilde, 2013).

2.7 Measurements of COD

The concentration of COD in wastewater was measured using the HANNA photometer apparatus in the laboratories of the Water and Environmental Studies Institute (WESI) at An-Najah National University. This analysis was made by adding 2 ml of wastewater sample to the COD vial with reagent and heating the sample to 150 °C for two hours by HANNA HI 839800. The vial was then left to cool, and the concentration was measured directly by HANNA HI 83214 photometer.

2.8 Measrenents of Nitrae

The concentration of Nitrate in wastewater was measured using the HANNA photometer apparatus. This analysis was made by adding 1 ml of wastewater sample to the NO_3^- vial, then the reagent was added to the vial. The vial was put in the apparatus and waited for 5 minutes before the concentration was measured directly by HANNA HI 83214 photometer.

2.9 Measurements of Ammonium

The concentration of Ammonium in wastewater was measured like the test of Nitrate, using the HANNA photometer apparatus. This analysis was made by adding 1 ml of wastewater sample to the NH_3 vial, then the reagent was added to the vial. The vial was put in the apparatus and waited for 5 minutes before the concentration was measured directly by HANNA HI 83214 photometer.

2.10 Measurements of Total Nitrogen

The concentration of TN in wastewater was measured using the HANNA photometer apparatus. This analysis was made by adding 0.5 ml of wastewater sample to the TN vial containing reagent and heating the sample to 105 °C for half an hour by HANNA HI 839800. The vial was then left to cool, then another reagent was added to the vial, and the concentration was measured by HANNA HI 83214 photometer.

2.11 Measurement of Phosphorus

The concentration of PO_4^{3-} in wastewater was measured using GENESYS 10S UV-VIS spectrophotometer apparatus in the laboratories of the Water and Environmental Studies Institute (WESI) at An-Najah National University. This analysis was made using the Ascorbic Acid method from Standard Methods for Water and Wastewater Examination (Brandi & Wilson-Wilde, 2013), four reagents were prepared for this test (Sulphuric Acid, Ascorbic Acid, Ammonium Molybdate solution, and Antimony Potassium Tartrate solution), these reagents were mixed and were added to the samples of wastewater sample, then this mixture was poured in cuvettes of a spectrophotometer to measuring phosphorus concentration.

The concentration of phosphate in wastewater sample was calculated according to the equation (2):

$$Conc.of\ Phosphate = apparatus\ reading \times 2.1 \tag{2}$$

Where:

Conc. of Phosphate in (mg/L).

2.1 is correction factor.

2.12 Measurement of Total Suspended Solids

The concentration of TSS was measured using a vacuum filter. The filter paper was weighed before starting, and then 100 ml of wastewater sample was taken and filtered with a vacuum to collect suspended solids in the sample on the filter paper. The filter paper was then taken to the oven, where it was heated to 105 °C, filter paper was left in the oven for at least two hours to completely dry. The filter paper was then reweighted after drying. The concentration of TSS was calculated according to the following equation (3):

$$TSS\ conc. = (\Delta weight \times 1000)/V_s \qquad (3)$$

Where:

TSS concentration in (mg/L).

Δweight: the difference between filter paper weight before filtering and after drying.

V_s: sample volume (L).

2.13 Measurement of Total Dissolved Solids

The concentration of TDS was measured like TSS using a vacuum filter. The beaker was weighed before starting, and then 100 ml of wastewater sample was taken and filtered with a vacuum. The filtered water was then taken in the beaker to the oven, where it was heated to 180 °C, the beaker was left in the oven for 24 hours to completely dry. The beaker was then reweighted after drying. The concentration of TDS was calculated according to the following equation (4):

$$TDS\ conc. = (\Delta weight \times 1000)/V_s \qquad (4)$$

Where:

TDS concentration in (mg/L).

Δweight: the difference between beaker weight before filtering and after drying.

V_s: sample volume (L).

2.14 Calculation of Chlorophyll A

Since algae are considered a family of plants and are autotrophs, the chlorophyll A pigment is important in the formation of algae because of its main role in photosynthesis. Chlorophyll a is an essential component in algae and plant cells. It is responsible for absorbing light energy from the light source and converting it into chemical energy that algae consume in the process of photosynthesis. Its importance also lies in its ability to produce carbohydrates in algae cells. Its distinctive green color indicates the health of the cells. The stronger the color, the more active and vital the algae cells are (Pareek et al., 2017).

The concentration of chlorophyll A was measured using GENESYS 10S UV-VIS spectrophotometer apparatus, the samples were prepared by filtering 100 ml of wastewater with algae using a vacuum filter and adding magnesium carbonate, then filter papers were placed in a desiccator and frozen to the after day. The reagents used in this test are (aqueous acetone solution, 0.1 N hydrochloric acid, and (1%) magnesium carbonate suspension) which were prepared in the laboratory, after day the samples were frozen, were placed in a centrifuge tube and 10 ml of aqueous acetone solution was added, then the samples were put in sonicator for 20 seconds at 5 settings. After that samples were placed in a dark box in the cold room to allow the extract to steep overnight. The extract was clarified using a centrifuging system, then absorbance was measured at wavelengths (750, 663, 645, 630, and 665 nm) using a spectrophotometer instrument (Sosa et al., 2019).

The concentration of chlorophyll A was calculated according to the following equations (5, 6, and 7):

$$Uncorr.Chlor.A = \frac{[11.64(Abs663) - 2.16(Abs645) + 0.1(Abs630)] \times E(F)}{V(L)} \quad (5)$$

Where:

Uncorrected chlorophyll A concentration in (μg/L).

F: dilution factor.

E: the volume of acetone used for the extraction (mL).

V: the volume of water filtered (L).

L: the cell path length (cm).

To calculate correct chlorophyll A, 0.1 ml of 0.1 N HCL was added to samples in spectrophotometer cells, and absorbance was remeasured for previous wavelengths. Corrected chlorophyll A was calculated according to the following equation (6):

$$Corrected\ Chlor.A = \frac{26.73(663_b - 665_a) \times E(F)}{V(L)} \quad (6)$$

And to calculate Pheophytin A, the following equation (7) was used:

$$Pheophytin\ A\ (\mu g/L) = \frac{26.73(1.7 \times [665_a] - 663_b) \times E(F)}{V(L)} \quad (7)$$

Where:

Corrected chlorophyll A concentration in (µg/L).

F: dilution factor.

E: the volume of acetone used for the extraction (mL).

V: the volume of water filtered (L).

L: the cell path length (cm).

665_a: the turbidity corrected Abs at 665 nm after acidification.

663b: the turbidity corrected Abs at 663 nm before acidification.

2.15 Calculations of Efficiency

Efficiency is an amount or percentage that generally expresses the extent of hitting the target and the quality of accomplishing a task by making optimal use of available resources and not wasting them to obtain the desired result, efficiency for nutrients removal can be calculated by:

$$Efficiency = \frac{C_i - C_a}{C_i} \times 100\% \quad (8)$$

Where:

C_i: initial concentration (mg/L).

C_a: final concentration (mg/L).

Chapter Three

Results

After the experiment was started, water samples were taken from almost every beaker, day after day. The first beaker was the one that was exposed to light throughout the day and was called (light sample). The second beaker was the one that was exposed to light for 19 hours at night and was left in the daylight for 5 hours and was called (half light sample), and the third beaker was the one that was left in natural light all day and was named (natural sample). The necessary nutrient analyses were performed, and the results were as follows:

3.1 COD removal

Samples were taken and analyses were performed every day after day during the 15 day experimental period. The results of COD removal from wastewater and the decrease in its concentration are shown in Table (2).

Table 2

COD concentration in three (light, half light, and natural) samples (mg/L)

days	Light	Half light	Natural
1 day	94	73	70
4 day	85	55	15
6 day	53	36	15
8 day	34	25	16
11 day	20	17	8
13 day	<5	<5	<5
15 day	<5	<5	<5

Table (2) shows the concentrations of COD in three samples during the 15 days of the experiment. The concentration of COD in the light sample was 94 mg/L on the first day until it reached less than 5 mg/L on the 13th and 15th day with efficiency (95%), while the concentration of COD in half light was 73 mg/L on the first day until it reached less than

5 mg/L on the 13th and 15th day with efficiency (93%), and the concentration of COD in natural was 70 mg/L on the first day until it reached less than 5 mg/L on the 13th and 15th day with efficiency (93%). COD removal from wastewater depends on the oxidation of organic materials that contain carbon bonds. Bacteria, whether aerobic or anaerobic, can oxidize organic materials, and aerobic bacteria depend on the concentration of oxygen in the surrounding environment so that they can decompose organic materials biologically. Here, the role of algae appears in the process of oxidation of organic materials, as they are autotrophic organisms that need carbon dioxide in order to carry out the process of photosynthesis and have the ability to decompose inorganic carbon. Thus, algae consume carbon dioxide for the process of photosynthesis, while they release the oxygen gas resulting from this process to return bacteria use it as an air source for the process of decomposing organic materials and converting them into carbon dioxide (Johnson et al., 2020). This explains the symbiotic relationship between algae and bacteria in removing and oxidizing organic materials present in wastewater. This means bacteria grow alongside the algae in the samples, and because of the symbiotic relationship between algae and bacteria, COD is removed from the wastewater effectively.

Figure (5.a) shows C/C_0 of COD for three samples, in the light and half light samples, the COD concentration decreases gradually until it reaches less than 5 mg/L, while in the natural sample, the concentration decreases dramatically and sharply until it reaches less than 5 mg/L. The COD removal process in the natural sample was more efficient than the light and half light samples, and this is due to the presence of bacteria that were highly effective in removing COD.

3.2 Nutrients removal

3.2.1 Phosphate removal

Samples were taken and analyses were performed every day after day during the 15 day experimental period. The results of Phosphate (PO_4^{3-}) removal from wastewater and the decrease in its concentration are shown in Table (3).

Table 3

Phosphate (PO_4^{3-}) concentration in three (light, half light, and natural) samples (mg/L)

days	Light	Half light	Natural
1 day	11.76	11.09	10.58
4 day	4.60	6.11	8.15
6 day	3.00	3.49	8.63
8 day	1.53	3.05	8.97
11 day	1.07	1.87	9.28
13 day	1.01	1.18	9.51
15 day	0.48	0.71	9.95

Table (3) shows the concentrations of PO_4^{3-} in three samples during the 15 days of the experiment. The concentration of PO_4^{3-} in the light sample was 11.76 mg/L on the first day until it reached 0.48 mg/L on the last day with efficiency (96%), while the concentration of PO_4^{3-} in half light was 11.09 mg/L on the first day until it reached 0.71 mg/L on the last day with efficiency (94%), and the concentration of PO_4^{3-} in natural was 10.58 mg/L on the first day until it reached 9.95 mg/L on the last day with efficiency (6%). Phosphate is one of the nutrients that must be removed from wastewater. It is an important element in algae growth, so algae use it for growth and metabolism and are thus removed from wastewater. Algae can effectively reduce phosphorus concentration in wastewater by up to (90%) if provided with appropriate environmental conditions such as temperature and light intensity. Algae use more than one step to remove phosphorus from wastewater, including absorption, where algae cells absorb phosphorus to use it for growth and increase the number of cells. The adsorption step that occurs on the surface of algae cells, then take up the portion of phosphate ions they need and precipitate the rest. This depends on the surrounding environmental conditions and the interactions between algae and other organisms in the wastewater (Delgadillo-Mirquez et al., 2016), Phosphate removal from the light was the most efficient of the three samples.

Figure (5.b) shows C/C_0 of PO_4^{3-} for three samples, in the light and half light samples, the PO_4^{3-} concentration decreased gradually until it reached a small quantity, while in the natural sample, in the first the concentration decreased slightly, then increased again after the fourth day. This means that after the fourth day, the algae began to decay and decompose, releasing phosphorus into the water again.

3.2.2 Ammonium and Nitrate removal

Samples were taken and analyses were performed every day after day during the 15 day experimental period. The results of Ammoium (NH_4^+) and Nitrate (NO_3^-) removal from wastewater and the decrease in its concentration are shown in Table (4).

Table 4

Ammoium (NH_4^+) and Nitrate (NO_3^-) concentration in three samples (mg/L)

days	Light		Half light		Natural	
	NH_4^+	NO_3^-	NH_4^+	NO_3^-	NH_4^+	NO_3^-
1 day	58	0	61	0	54	0
4 day	23	2.7	39	2.5	41	0.1
6 day	9	4.6	21	3.1	34	0.2
8 day	<0.1	2.1	15	1.8	29	0.5
11 day	<0.1	0.8	6	0.5	26	0.3
13 day	<0.1	0.2	<0.1	0.1	22	0.1
15 day	<0.1	0	<0.1	0	20	0

Table (4) shows the concentrations of NH_4^+ and NO_3^- in three samples during the 15 days of the experiment. The concentration of NH_4^+ in the light sample was 58 mg/L on the first day until it reached less than 0.1 mg/L on the 8th day with efficiency (99.8%), and the concentration of NO_3^- in this sample started at zero and reached maximum on the 6th day which 4.6 mg/L then decreased again to reach zero on the last day, while the concentration of NH_4^+ in the half light sample was 61 mg/L on the first day until it reached less than 0.1 mg/L on the 13th day with efficiency (99.8%), and the concentration of NO_3^- in this sample started at zero and reached maximum on the 6th day which was 3.1 mg/L then decreased again to reach zero on the last day, and the concentration of NH_4^+ in natural was 45 mg/L on the first day and it reached 20 mg/L on the last day with efficiency (56%), while the concentration of NO_3^- in natural sample started at zero and reached maximum on 8th day which was 0.5 mg/L then decreased again to reach zero on the last day. Algae cells can absorb ammonium and nitrates from wastewater, as they are compounds that contain nitrogen, which is an essential element in building cells for growth and in the photosynthesis process carried out by algae. Thus, the concentration of nitrogen compounds in wastewater decreases (Delgadillo-Mirquez et al., 2016). The concentration of nitrates during the wastewater treatment process depends on the concentration of ammonium, as an oxidation process of ammonium occurs, and this process is called nitrification. The nitrification process is carried out by two types of bacteria, which produce nitrite, and then, with the continuation of the oxidation process, nitrate is produced (Holmes et al., 2019). Since nitrate is produced from the oxidation of ammonium, this explains the increase in the concentration of nitrate whenever the concentration of ammonium decreases, as the concentration of nitrate begins at zero, then the ammonium concentration begins to decrease when it is oxidized, and the nitrate concentration begins to increase, and with the presence of algae that can absorb the nitrogen element from its compounds, the concentration returns to decrease. Nitrates are returned to zero, so ammonium and nitrates are eliminated by algae.

Figure (5.c) shows C/C_0 of NH_4^+ for three samples, in the light and half light samples, the NH_4^+ concentration decreased gradually until it reached small quantity, while in the natural sample, the concentration decreased slightly, from the figure it appears that in the light sample, the ammonium removal process was faster and with high efficiency.

3.2.3 Total Nitrogen removal

Samples were taken and analyses were performed every day after day during the 15 day experimental period. The results of Total Nitrogen (TN) removal from wastewater and the decrease in its concentration are shown in Table (5).

Table 5

Total Nitrogen (TN) concentration in three (light, half light, and natural) samples (mg/L)

days	Light	Half light	Natural
1 day	69	75	60
4 day	38	49	54
6 day	22	36	47
8 day	14	21	44
11 day	9	12	41
13 day	5	9	35
15 day	4	7	28

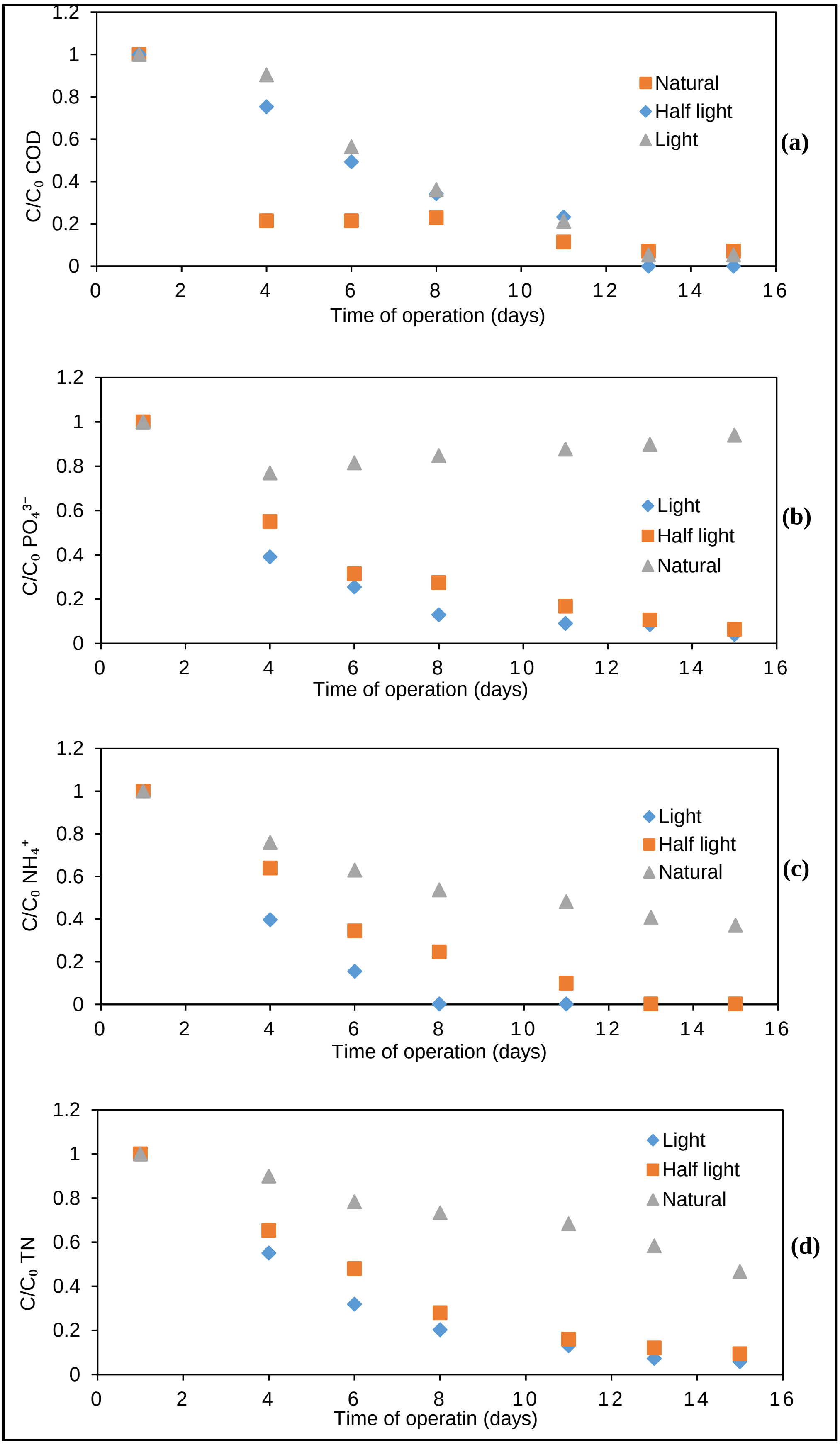
Table (5) shows the concentrations of TN in three samples during the 15 days of the experiment. The concentration of TN in the light sample was 69 mg/L on the first day until it reached 4 mg/L on the last day with an efficiency (94%), while the concentration of TN in half light was 75 mg/L on the first day until it reached 7 mg/L on the last day with efficiency (91%), and the concentration of TN in natural was 60 mg/L on the first day until it reached 28 mg/L on the last day with efficiency (53%). Total nitrogen is the sum of nitrogen compounds present in wastewater, and measuring total nitrogen concentration is important in evaluating the removal of nitrogen from wastewater during the treatment process. As mentioned previously, algae have proven effective in absorbing nutrients and using them in the processes of growth and metabolism. The most important of these nutrients is nitrogen, as algae combine with bacteria and work effectively to remove a large percentage of total nitrogen, which works to improve water quality and get rid of nutrients. Nitrogen element undergoes transformation processes during the process of removing it from wastewater, the nitrification process is carried out by specialized aerobic bacteria called *Nitrosomonas*, which begin by converting ammonia into nitrite, followed by *Nitrobactor* bacteria that convert nitrite into nitrate. The denitrification process is carried out by anoxic bacteria and is based on reduce nitrate to nitrogen gas and carbon dioxide. (Jia & Yuan, 2016). During 15 days of the experiment, the half light sample had the highest total nitrogen removal efficiency.

Figure (5.d) shows C/C_0 of TN for three samples, in the light and half light samples, the TN concentration decreased gradually until it reached a small quantity, while in the natural sample, the concentration decreased slightly.

C/C_0 represents the percentage decrease in concentration and the nature of this reduction. These results can be further analyzed through kinetic analysis, where design parameters can be concluded from the kinetic analysis.

Figure 5

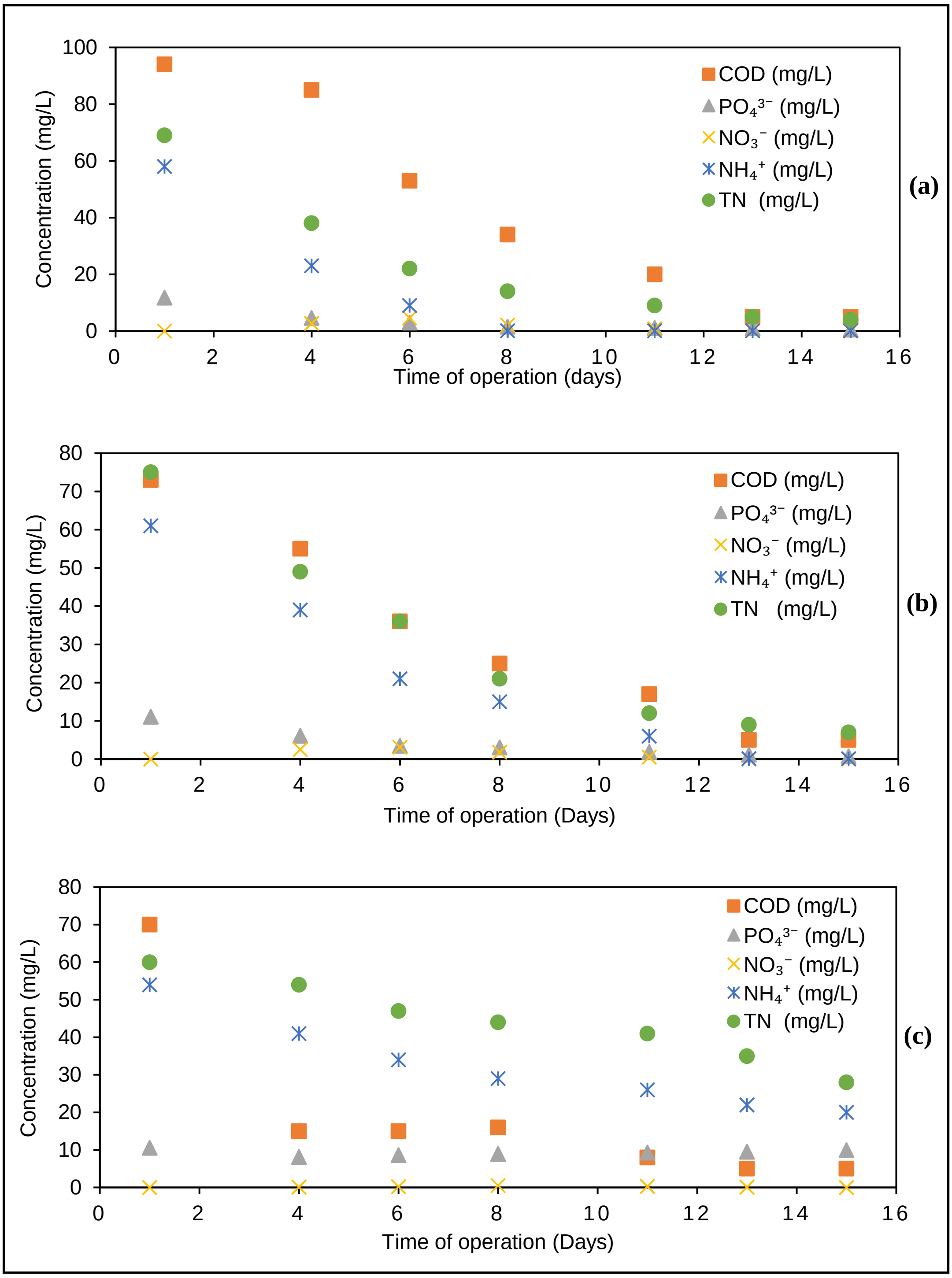
C/C₀ of (COD, PO_4^{3-}, NH_4^+, and TN) for three samples



Following Figures (6(a-c)), shows concentrations of COD and nutrients in three (light, half light, and natural) samples.

Figure 6

Concentrations of (COD, PO_4^{3-}, NO_3^-, NH_4^+, and TN) in three samples



3.3 Total Suspended Solids

The total suspended solids were measured as containing different particles, the most important of which are algae cells and bacteria in wastewater (Azarian et al., 2007). Therefore, the concentration of algae cells was considered the total suspended solids. While the total dissolved solids in wastewater are defined as a mixture of organic substances resulting from the decomposition of algae and various mineral salts such as calcium, potassium, magnesium, etc. (Pushpalatha et al., 2022), the following are the results of measuring TSS and TDS.

Table 6

TSS and TDS concentration in three samples (mg/L)

parameter	Light	Half light	Natural
TSS	737.4	237.4	174
TDS	743	782	951

Table (6) shows the concentrations of TSS and TDS in three samples, TSS concentration in the light sample was 737.4 mg/L, and TDS concentration in this sample was 743 mg/L, while TSS in the half light sample was 237.4 mg/L and TDS in this sample was 782 mg/L, and in natural sample TSS was 174 mg/L, and TDS in this sample was 951 mg/L. These results show that the concentration of TSS in the light sample was the highest. Since the value of TSS indicates the concentration of algae cells and their number, this means that in the light sample, the growth of algae and the number of their cells were higher and better than in the other samples. Also, the concentration of TDS in the light sample was lower, which means dissolved solids, including decomposing algae cells, were less in this sample. Based on these results, the efficiency of algae in the treatment process was better in the light sample.

3.4 Chlorophyll A concentration

Chlorophyll A is the green pigment produced by algae based on several factors, including the availability of light and the availability of nutrients, the most important of which are nitrogen and phosphorus. This pigment absorbs light to help algae in the process of photosynthesis (Adams et al., 2021). Therefore, the concentration of chlorophyll A has an important effect on the growth and effectiveness of algae and is an indicator of the efficiency of algae and their vitality in water. the following are the results of measuring Chlorophyll A concentration.

Table 7

Concentration of Chlorophyll A in three samples (µg/L)

parameter	Light	Half light	Natural
Corrected Chlorophyll A	129.4	57.2	8.0
Pheophytin A	229.9	302.0	-23.0
Uncorrected Chlorophyll A	268.8	237.4	-2.5

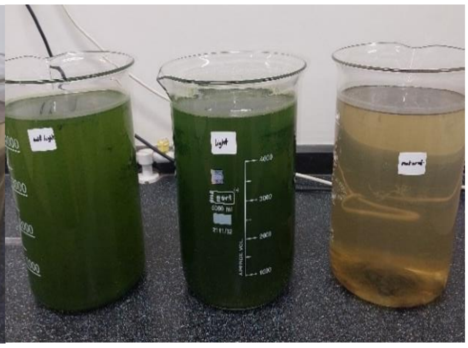
Table (7) shows the concentration of chlorophyll A in three samples, the concentration of corrected chlorophyll A in the light sample was 129.4 µg/L, and uncorrected chlorophyll A in this sample was 268.8 µg/L, while the concentration of corrected chlorophyll A in the half light sample was 57.2 µg/L, and uncorrected chlorophyll A in this sample was 237.4 µg/L, in the natural sample, the concentration of corrected chlorophyll A was 8 µg/L, and uncorrected chlorophyll A in this sample was -2.5 µg/L. These results show that the concentration of chlorophyll A pigment in the light sample was the highest. Since the chlorophyll A value indicates the concentration and activity of algae cells, this means that in the light sample, the algae growth and its activity in absorbing nutrients from wastewater were higher and better than in the other samples. It also appears from the results that the concentration in the natural sample was negative values, meaning that it did not contain chlorophyll pigment, This indicates that the algae cells in it were decomposed and inactive, as was shown in previous analyses of this sample. Based on these results, the efficiency of algae in the treatment process was better in the optical sample.

Algae activity was monitored during the experiment through visual observation and through nitrogen and phosphorus uptake, and this was shown by the results of sample analysis during the experiment period. Following figures (7(a-f)) show samples during the 15 days of the experiment, It is noted in the figures the stages of algae growth during the days of the experiment, where the algae cells were clear in the water and their density was increasing over the days. Also, the green color was getting darker, which meant an increase in the concentration of chlorophyll pigment and an increase in algae growth and activity, and this was clear in the two light and half light samples. The natural sample was brown, and it was clear that the algae cells were decayed and inactive. This was evident in the results of the sample analyses.

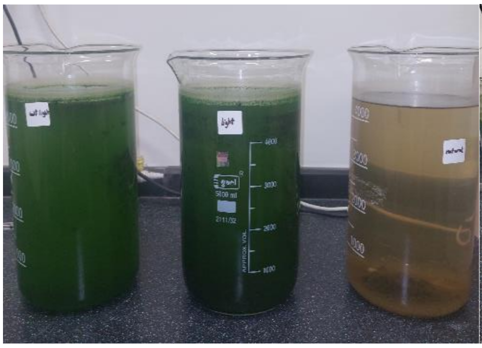
Figure 7

Samples during the 15 days of the experiment

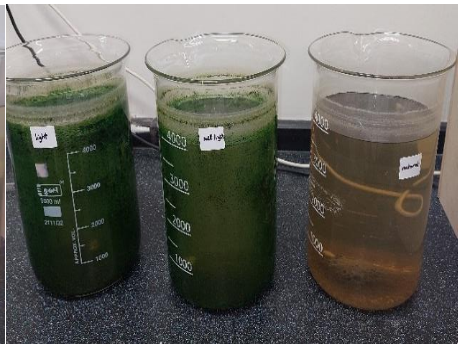
(a) *Samples on the fourth day of the experiment*



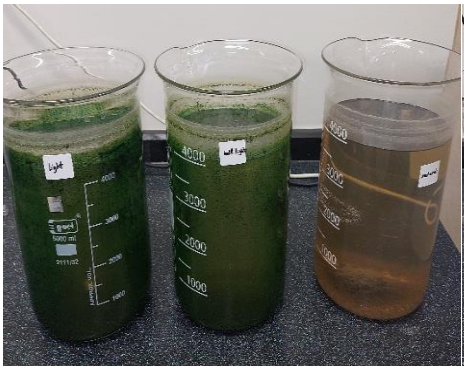
(b) *Samples on the sixth day of the experiment*



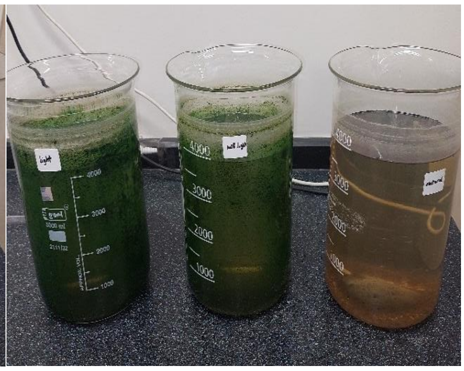
(c) *Samples on the eighth day of the experiment*



(d) *Samples on the eleventh day of the experiment*



(e) *Samples on the thirteenth day of the experiment*



(f) *Samples on the fifteenth day of the experiment*

3.5 Discussion

Some of the different parameters used in this study were based on the results of previous studies in the field of wastewater treatment and nutrient removal using algae as a natural and environmentally friendly method. The Table (A.7) in Appendix (A) shows some of the previous studies with their results, parameters, and different methods used, discusses the differences and similarities, and compares them with the results of this research.

According to Sayara et al. (2021), the effect of the symbiotic relationship between algae and bacteria on removing nutrients from wastewater was studied, as a 600 L sequencing batch photobioreactor (SBR) was used, distributed over three equal basins. After 10 days incubation period, the effect of the symbiotic relationship between algae and bacteria was positive in improving the removal of nutrients from wastewater and in reducing greenhouse gas emissions, as the aerobic respiration of bacteria produces carbon dioxide, which the algae consume in the process of photosynthesis. Another result of this study was that (60%) of COD, (40%) of nitrogen (NH_4^+-N), and (90%) of phosphorus were removed, as the dissolved oxygen concentration during daylight periods was 6 mg/L (Sayara et al., 2021). In comparison between results from this study, after 15 days of incubation in a batch reactor, when the light was used for 24 hours per day, nutrients were removed with high efficiencies, COD was removed with an efficiency of (95%), ammonium (99.8%), and phosphorus removed with efficiency (96%), this means that the treatment process with algae by applying artificial light achieved high efficiency and quality.

Based on Zimmo et al. (2000), The efficiency of algae and duckweed containers and the difference between them in removing nutrients, especially nitrogen, from wastewater was studied. The time needed for this experiment was 15 days, where nitrogen was used at different concentrations. The nitrogen concentration in the first time was 50 mg nitrogen/L and 100 mg nitrogen/L in the second time. Applying nitrogen at different concentrations led to differences in environmental conditions between the containers in the experiments. The study showed that the dissolved oxygen (DO) concentration was higher in the algae-based container than in the duckweed container. Also, the pH was higher in the algae-based container than in the duckweed container, which means that the efficiency of algae is higher in removing nitrogen from wastewater. This is because the duckweed plants stay on the surface of water, in the container of duckweed, which leads to block sunlight from the algae and thus reduces the efficiency of the photosynthesis process. As for nitrogen removal, between (42%) and (62%) of the total nitrogen was removed from the duckweed based containers, and between (56%) and (95%) of the Kjeldahl nitrogen. The nitrogen removal rate was between (45%) in the algae-based containers as a treatment and (48%), and (48%) and (58%) for Kjeldahl nitrogen. This study proved that the difference in environmental conditions between containers based on duckweed and containers based on algae has an important impact on the mechanism of nitrogen removal from wastewater (Zimmo et al., 2000). In comparison between this study and our study, the results for algae based in this study the efficiency of algae in removing total nitrogen was (56%) and (95%) for two initial concentrations of nitrogen, in our study during 15 days, initial total nitrogen was 69 mg/L for light sample, and the final concentration was 4 mg/L with efficiency (94%).

According to Zimmo et al. (2002), this study was based on evaluating the differences in environmental conditions and their impact on the efficiency of wastewater treatment in two systems: ponds based on duckweed and ponds based on algae. This study lasted for 12 months, during which two systems were fed, each consisting of four equal basins, with a constant flow of partially treated wastewater. Among the results of this study, pH values were higher in ponds based on algae. Also, the concentration of dissolved oxygen (DO) was higher in ponds based on algae, so duckweed was more efficient than algae in removing organic matter (BOD) and TSS. As for fecal coliforms, their removal was higher in algae-based ponds. As for nitrogen removal, algae proved effective in removing (80%) of the nitrogen present in wastewater compared to duckweed, which removed (55%) of the nitrogen. This study showed the effect of different environmental conditions in the two systems on the efficiency of wastewater treatment (Zimmo et al., 2002). In comparison with our study, the algae proved its high efficiency in removing nitrogen which was (94%) during 15 days of the experiment in light sample.

According to Nguyen et al. (2022), this study was based on a comparison between algae based wastewater treatment and bacteria based wastewater treatment. Among the results of this study is that the use of algae in wastewater treatment requires pretreatment for wastewater. Also, algae does not work at low nutrient concentrations because the efficiency of algae in their remedial work depends on the production of biomass, so they need nutrients for growth and cell reproduction. Algae also depend on carbon dioxide, which they obtain mainly from a symbiotic relationship with bacteria. It needs a retention time (HRT) of more than 10 days, compared to bacteria that need a few hours. Algae also depend on absorbing light, which increases their efficiency in the treatment process (Nguyen et al., 2022). In comparison between results from this study, The water that was used was a mixture of primary treatment water and secondary treatment water. The use of light in this study had a positive effect on increasing the growth and reproduction of algae and improving the photosynthesis process, as algae grow with bacteria in a symbiotic relationship where bacteria consume oxygen and release carbon dioxide, which algae absorb and depend on in the photosynthesis process. The experiment lasted for 15 days, where algae proved effective in removing nutrients, especially nitrogen and phosphorus, from wastewater with high efficiency. The use of algae in wastewater treatment is a natural, effective, environmentally friendly, and economically inexpensive treatment method.

According to Whitton et al. (2019), this research studied how the light types and its wavelengths affect the efficiency of algae in the form of beads in removing nutrients from wastewater during a reaction period of 3 hours. White light (400-700 nm) with a photon flux efficiency of 200 μmol/m^2.s, blue light with a wavelength of (465 nm), and red light (660 nm) were used. The study concluded that algae proved to be highly effective and efficient when white light was used in removing phosphate from wastewater, while the phosphate concentration decreased by 10.7 mg P/h per bead. When blue light was used, the phosphate concentration decreased by 10.2 mg P/h per bead, while when red light was used, the phosphate concentration decreased by 10.1 mg P/h per bead. The results concluded that white light is the best for improving algae growth and increasing their efficiency in the wastewater treatment process, because white light has the ability to cover a larger lightining area, as it has the ability to penetrate deeper (Whitton et al., 2019). In comparison with the results of this study, visible light (380 nm - 700 nm) with an intensity of 760 lux, equivalent to (14.46 μmol/m^2.s) (Lighting, n.d.) was used to enhance algal growth during a retention period of 15 days. Light was used in two systems, the first for 24 hr, and the second for 19 hr with artificial light and 5 hr of natural daylight. In both systems, the efficiency of phosphate removal by algae was high, with (96%) of phosphate removed in the first system and (94%) of phosphate removed in the second system. High phosphate removal efficiency was achieved using visible light with different systems and low light intensity.

According to Oruganti et al. (2022), This study investigates the effect of mixing rate on the efficiency of algae in the wastewater treatment process, as it is considered an important factor among the many factors that help improve the wastewater treatment process and remove nutrients from it through algae and bacteria. The presence of microorganisms in wastewater is affected by many factors, including pH, light intensity, temperature, availability of nutrients, and mixing rate. The study proved that the appropriate mixing rate ensures that microorganisms obtain the nutrients necessary for their growth in the aquatic medium. Thus, the importance of mixing lies in the fact that it is responsible for distributing nutrients in the aquatic environment, which facilitates their consumption to build algae and bacterial cells. Mixing also prevents the sedimentation of biomass by distributing it so that it remains suspended in the medium, to facilitate the interaction process between algae and bacteria and ensure the arrival of the light necessary for photosynthesis to the cells. Since bacteria and algae live in a symbiotic relationship based on the exchange of oxygen and carbon dioxide according to each other's needs, the mixing rate plays an important role in this relationship, as it allows gas exchange to ensure good ventilation and for the bacteria to obtain the oxygen needed for respiration, which in turn produces carbon dioxide that the algae consume for photosynthesis, which increases their efficiency in removing nutrients from wastewater (Oruganti et al., 2022). These results are consistent with the results of this study, as providing appropriate conditions for the experiment had a positive impact on achieving high efficiency in removing nutrients from wastewater. Artificial light with an intensity of 760 lux was used for full time and half time to improve the photosynthesis process of algae. Also mixing rate of 100 rpm for 5 hours a day was used and a daily temperature of 28 °C. All of these factors resulted in high effectiveness in improving the interaction between algae and bacteria and, thus, high efficiency in removing nutrients from wastewater.

Based on Fan et al. (2021), in this research, the nature of the symbiotic relationship between algae and bacteria and the effect of available light intensity on the domestic wastewater treatment process were studied. In light of this coexistence and aggregation of bacterial algae granular sludge (MBGS), the appropriate light intensity improves the growth of algae and increases its cells by stimulating the photosynthesis process which is based on algae consuming nutrients and removing them from wastewater. Light intensity also stimulates the vital activities of MBGS, which enhances cooperation between algae and bacteria in removing nutrients and treating water, as light intensity positively affects oxygen production in the MBGS system., which ensures balance and integrated work between algae and bacteria. According to the results of this research, when the light intensity was increased from 70 to 210 μmol/m².s, the efficiency of removing nutrients from wastewater increased, as (70.5%) of COD, (80.7%) Ammonia, and (73.9%) Phosphate was removed (Fan et al., 2021). Compared to the conditions and results of this research, the effect of using light was positive on algae and on the nutrients removal from wastewater during the 15 day retention time of the experiment. The use of a light intensity of 760 Lux was highly efficient in enhancing the algae growth and its symbiotic relationship with bacteria, which appeared through its high efficiency in wastewater treatment from nutrients.

According to Priyadarshi, (2021), in this research the effect of using different concentrations of algae on removing nutrients from wastewater was studied to find the best concentration which improved the efficiency of nutrient removal. The same conditions were provided in all stages, and six different *Chlorella vulgaris* algae type were used. In the first stage, (30%) and (40%) of algae were used. In the second stage, (20%) and (25%) of algae were used. In the third stage, (35%) and (45%) of the algae were used by fixing the temperature and neglecting the rate of evaporation. The experiment retention time is 18 days using a batch reactor. The results of this research showed that (30%) algae concentration was the optimal concentration for removing nutrients from wastewater, as (81%) of COD, (75%) nitrate, (99%) phosphorus, and (99%) ammonia were removed (Priyadarshi, 2021). The results showed that the concentration of algae is an important factor in the process of treating wastewater from nutrients, adequate concentration of algae must be maintained, as increasing the concentration to a high percentage increases the turbidity of the water, which leads to blocking light from some internal cells and reducing the efficiency of the photosynthesis process, thus causes decreasing the efficiency of the treatment process.

According to Nguyen et al, (2020), this study investigated the extent to which the different inoculation rates of algae and bacteria affect the efficiency of removing nutrients from wastewater and the nature and conditions of the symbiotic relationship between microalgae and activated sludge, as the inoculation rate plays an important role in the metabolism and growth processes of microorganisms and affects the interaction between these organisms and their adaptation in aqueous media. The ideal inoculation ratio ensures suitable environmental conditions for the growth of microorganisms in wastewater and facilitates their work in water treatment, removing nutrients and increasing biomass production, which ensures a highly efficient treatment process. One of the findings from this study was that a mixing ratio of 3:1 microalgae:bacteria in photobioreactor during 11 days was optimum, with (99%) of COD, (79%) TP, and (86%) TN removed (Nguyen et al., 2020). In comparison with our study, Algae has proven its effectiveness in removing nutrients from wastewater using a batch reactor with a light intensity of 14.46 $\mu mol/m^2.s$ and a mixing rate of 100 rpm during an incubation period of 15 days. Because the symbiotic relationship, algae and bacteria grew naturally in wastewater, this relationship is stimulated by mixing and improving oxygen and carbon dioxide gases exchange between these organisims, to ensuring healthy growth of algae and bacteria and facilitating thier work in removing nutrients. The partnership between algae and bacteria proved highly efficient in removing nutrients, with (95%) of COD, (96%) TP, and (94%) TN removed.

Chapter Four

Conclusions and Recommendations

4.1 Conclusions

In this research, the effect of applying artificial light on the growth of algae in wastewater and its effectiveness in removing nutrients was studied. Three samples of wastewater were used and placed in the same environmental conditions with different durations of application of artificial light on each of them. The efficiency of algae was high in the light sample and the half-light sample, and the relationship was direct between algae growth and the duration of exposure to light, as the longer the period of light exposure, increased algae growth and increased its activity in removing nutrients. In the sample that was exposed to light for 24 hours (the light sample), the algae was more effective and efficient than the other samples. At the same time, the results were close between the light sample and the sample that was exposed to light for 19 hours (the half light sample) in terms of removing nutrients from wastewater. Still, there is a difference between the two samples in the concentration of TSS and the concentration of chlorophyll A pigment, which indicates the activity and vitality of the algae cells in the medium. The algae in the light sample were the most efficient in removing nutrients from wastewater, as their efficiency in removing COD was (95%) and phosphorus was (96%). (99.8%) ammonium, TSS concentration 737.4 mg/L, and chlorophyll A pigment concentration of 129.4 μg/L. In the half-light sample, the effectiveness of algae in removing COD was (93%), phosphorus was (94%), ammonium was (99.8%), TSS concentration was 237.4 mg/L, and chlorophyll A concentration was 57.2 μg/L. Finally, in the natural sample, the efficiency in removing COD was (93%), phosphorus was (6%), ammonium was (56%), TSS concentration was 174 mg/L, and chlorophyll A concentration was 8 μg/L. It was concluded from this study, the important effect of light duration on the growth of algae and its activity in removing nutrients from wastewater. From an economic standpoint, the light period of 19 hours can be considered the suitable duration of the applied artificial light. Exposing algae to continuous light for a long period of time leads to exhaustion, so they must be given a break from light to ensure healthy and natural growth and also to avoid overgrowth of algae, which reduces the efficiency of water treatment and nutrient removal because it works to block light and prevent it from reaching all cells appropriately. It causes a clogging system and increases the turbidity of the water. It was also concluded from this study that it is necessary to purify the water from algae after completing the nutrient removal through a suitable filtration process, so that the algae do not enter the death phase and release nitrogen and phosphorus back into the water, also, after proper treatment for the filtered algae, they can be collected, dried and used as animal feed. The results of this study can be relied upon in developing the tertiary treatment stage in wastewater treatment plants to produce water suitable for agriculture by applying artificial light during the night to enhance algae growth and remove nitrogen and phosphorus from wastewater. Referring to the Palestinian specifications and standards regarding treated water used in agriculture, the concentration of total phosphorus and total nitrogen at 30 mg/L is considered high quality water and classified as A, which means that the tertiary treatment process by applying artificial light requires 4-5 days as retention time to partially remove nitrogen and phosphorus to comply with Palestinian specifications, because agricultural water requires the presence of nitrogen and phosphorus at a certain concentration. Economically, the application of the tertiary treatment method using artificial light is inexpensive, as 1 meter of artificial light used costs approximately 1.5 $, and the price of 1 kWh is 0.16 $. Suppose this system is applied as a small operating unit in rural areas with an operating capacity of approximately 300 m^3 for the algae treatment reactor with the application of artificial light to increase efficiency for 4 days at a rate of 19 hours/day. In that case, the cost of purchasing artificial light is 59 $, and the operating cost of electricity is 17 $ for 4 days.

4.2 Recommendations

According to the results of this study, the following points are some recommendations which useful for future studies in the field of municipal wastewater treatment by algae:

1. Apply different period intervals of light on algae in the wastewater, to find the optimum period of light that is effective in nutrient removal from wastewater which ensures healthy growth of algae without exhausting it.
2. Apply different light intensities to find the optimum light intensity that achieves the highest efficiency in algae growth and nutrient removal from wastewater.
3. It is recommended to search for new methods to utilize algae when harvested from wastewater after the nutrient removal process is complete, such as, using the produced algae as animal feed, after dried and proper treatment.
4. It is recommended to use this system in rural countries for agricultural purposes, and develop this system to apply in large wastewater treatment plants.
5. Lipid measurements for harvested biomass of algae, and study of the potential use for biofuel production.
6. Measurement and control of CO_2 concentration in the system to find the optimum concentration that achieves the highest efficiency in algae growth.
7. Perform mass balance for elements N, P, K, and C to determine important parameters of design criteria for this system.
8. It is recommended to study more about the photoactivated sludge (PAS) system and the effect of this system on algae growth and nutrient removal from wastewater.

List of Abbreviations

Abbreviation	Meaning
PAS	Photoactivated Sludge system
COD	Chemical Oxygen Demand
BOD	Biochemical Oxygen Demand
DO	Dissolved Oxygen
ATP	Adenosine Triphosphate
P	Phosphorus
N	Nitrogen
C	Carbon
Fe	Iron
Si	Silicon
RNA	Ribonucleic Acid
DNA	Deoxyribonucleic Acid
NO_3^-	Nitrate
NH_4^+	Ammonium
EC	Electrical Conductivity
SBR	Sequencing Batch Reactor
RT	Reaction Time
LED	Light Emitting Diode
WESI	Water and Environmental Studies Institute
TN	Total Nitrogen
PO_4^{3-}	Phosphate
TSS	Total Suspended Solids
VSS	Volatile Suspended Solids
TDS	Total Dissolved Solids
HCl	Hydrochloric Acid
C/C_0	Concentration/Initial Concentration
Ppm	Part per million
pH	Power of Hydrogen
K	Potassium
Mg	Magnesium
S	Sulfur

HRT	Hydraulic Retention Time
MBGS	Microalgal Bacterial Granular Sludge
PBR	Photobioreactor
TP	Total Phosphorous

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Appendices

Appendix A

Tables

Table A.1

Nutrient concentrations in light sample with efficiency

days	COD (mg/L)	PO_4^{3-} (mg/L)	NO_3^- (mg/L)	NH_4^+ (mg/L)	TN (mg/L)
1 day	94	11.76	0	58	69
4 day	85	4.6	2.7	23	38
6 day	53	3	4.6	9	22
8 day	34	1.53	2.1	<0.1	14
11 day	20	1.07	0.8	<0.1	9
13 day	<5	1.01	0.2	<0.1	5
15 day	<5	0.48	0	<0.1	4
Efficiency	95%	96%	-	99.8%	94%

- Light sample: is sample that was treated with exposure the sample for artificial light for 24 hours.

Table A.2

Nutrient concentrations in half light sample with efficiency

days	COD (mg/L)	PO_4^{3-} (mg/L)	NO_3^- (mg/L)	NH_4^+ (mg/L)	TN (mg/L)
1 day	73	11.09	0	61	75
4 day	55	6.11	2.5	39	49
6 day	36	3.49	3.1	21	36
8 day	25	3.05	1.8	15	21
11 day	17	1.87	0.5	6	12
13 day	<5	1.18	0.1	<0.1	9
15 day	<5	0.71	0	<0.1	7
Efficiency	93%	94%	-	99.8%	91%

- Half light sample: is sample that was treated with exposure the sample for artificial light for 19 hours, and 5 hours in natural daily light.

Table A.3

Nutrient concentrations in natural sample with efficiency

days	COD (mg/L)	PO_4^{3-} (mg/L)	NO_3^- (mg/L)	NH_4^+ (mg/L)	TN (mg/L)
1 day	70	10.58	0	54	60
4 day	15	8.15	0.1	41	54
6 day	15	8.63	0.2	34	47
8 day	16	8.97	0.5	29	44
11 day	8	9.28	0.3	26	41
13 day	<5	9.51	0.1	22	35
15 day	<5	9.95	0	20	28
Efficiency	93%	6%	-	63%	53%

- Natural sample: is sample that was left in natural daily light for 24 hours per day.

Table A.4

C/C_0 for nutrient concentrations in light sample

days	C/C_0 COD	C/C_0 PO_4^{3-}	C/C_0 NH_4^+	C/C_0 TN
1 day	1	1	1	1
4 day	0.9042	0.3911	0.3966	0.5507
6 day	0.5638	0.2551	0.1552	0.3188
8 day	0.3617	0.1301	0.0017	0.2029
11 day	0.2128	0.0910	0.0017	0.1304
13 day	0.0532	0.0859	0.0017	0.0725
15 day	0.0532	0.0408	0.0017	0.0580

Table A.5

C/C_0 for nutrient concentrations in half light sample

days	C/C_0 COD	C/C_0 PO_4^{3-}	C/C_0 NH_4^+	C/C_0 TN
1 day	1	1	1	1
4 day	0.7534	0.5509	0.6393	0.6533
6 day	0.4932	0.3147	0.3443	0.4800
8 day	0.3425	0.2750	0.2459	0.2800
11 day	0.2329	0.1686	0.0984	0.1600
13 day	0.0685	0.1064	0.0016	0.1200
15 day	0.0685	0.0640	0.0016	0.0933

Table A.6

C/C_0 for nutrient concentration in natural sample

days	C/C_0 COD	C/C_0 PO_4^{3-}	C/C_0 NH_4^+	C/C_0 TN
1 day	1	1	1	1
4 day	0.2143	0.7703	0.7593	0.9000
6 day	0.2143	0.8157	0.6296	0.7833
8 day	0.2286	0.8478	0.5370	0.7333
11 day	0.1143	0.8771	0.4815	0.6833
13 day	0.0714	0.8989	0.4074	0.5833
15 day	0.0714	0.9405	0.3704	0.4667

Table A.7

Discussion, methods of previous studies with similarities and differences with this research.

Method	Summary	Results	Reference
Photobioreactor operating under sequencing batch mode (SBR) fed with pre-treated domestic wastewater	The use of algae in wastewater treatment has an efficient and positive impact on greenhouse gases. Natural light for 10 days and a temperature of 14 to 23 °C was used to operate the photobioreactor in sequencing batch mode containing algae and bacteria.	• 60% COD removed • 40% ammonium removed • 90% phosphourus removed	(Sayara et al., 2021)
Nitrogen was applied at different initial concentrations in two experiments, in each experiment nitrogen was applied to duckweed based containers, and algae based containers.	The difference between the efficiency of duckweed and algae containers in removing nitrogen from wastewater has been studied on a laboratory scale. The experiment was conducted over a period of 15 days, and nitrogen was applied at different initial concentrations in two experiments.	• 42% and 62% TN was removed from the duckweed containers. • 56% and 95% of the Kjeldahl nitrogen was removed from duckweed containers. • 45% and 48% TN was removed by algae. • 48% and 58% for Kjeldahl nitrogen was removed by algae.	(Zimmo et al., 2000)
Two systems was used: ponds based on duckweed and ponds based on algae for 12 months, each system consisting of four equal basins, with a constant flow of partially treated wastewater.	This study was based on evaluating the differences in environmental conditions and their impact on the efficiency of wastewater treatment in two systems: ponds based on duckweed and ponds based on algae.	• 80% of nitrogen was removed by algae. • 55% of nitrogen was removed by duckweed. • Duckweed was more efficient than algae in removing organic matter (bod) and tss.	(Zimmo et al., 2002)
Comparison between algae and bacteria in wastewater treatment	Algae in wastewater treatment depend on the concentration of nutrients, which affects the growth of algae cells. Thus, algae can remove nutrients from wastewater with the help of bacteria that reproduce with it in a symbiotic relationship.	• Algae needs more than 10 days retention time for treatment. • Bacteria needs hours for treatment.	(Nguyen et al., 2022)

Comparison between white light with (400 – 700 nm) and blue light (465 nm) and red light (660 nm), with intensity (200 µmol/m².s), and retention time 3 hours, in phosphate removal from wastewater.	The effect of exposure of algae to light in wastewater on their efficiency in removing phosphate. The best lighting was to use white light with an intensity of 200 µmol/m².s.	• For white light phosphate decreased in rate 10.7 mg P/h per bead. • Blue light phosphate decreased in rate 10.2 mg P/h per bead. • Red light phosphate decreased in rate 10.1 mg P/h per bead.	(Whitton et al., 2019)
Wastewater treatment using symbiotic relationship between algae and bacteria and system modeling, and utilization of algae for biofuel production using microfluidic system.	The most important factors that affect the life cycle of algae with bacteria in wastewater are temperature, pH, and light intensity, which play an essential role in increasing algae biomass and removing nutrients.	• The symbiotic relationship in the algal-bacterial system has proven effective in improving the removal of nutrients from wastewater, by providing factors and conditions that help the development of the system, such as pH, light intensity, reaction time and temperature.	(Oruganti et al., 2022)
Study of light intensity on the algal-bacterial complex in domestic wastewater and its effect on the treatment process by increasing the light intensity from 70 to 210 µmol/m².s.	Light has an important role in the growth of microorganisms in wastewater, improving the treatment process, and removing nutrients. Also, the amount of light intensity has an essential role in stimulating the symbiotic relationship between algae and bacteria.	• 70.5% COD was removed. • 80.7% Ammonia removed. • 73.9% Phosphate removed. • Increased algal bacterial biomass.	(Fan et al., 2021)
Six concentrations of microalgae were used to treat wastewater, and their effect on nutrient removal was studied. In the first stage, the algae concentration was 30% and 40% for 18 days with a 24-hour retention period in batch reactor. The second stage, the algae concentration was 20% and 25%, and the third stage the algae concentration was 35% and 45%.	Study the importance of using algae in wastewater treatment and the effect of using different concentrations of microalgae on removing nutrients from wastewater, as the rate of algae growth and reproduction has a direct impact on nutrient removal.	• 81% COD removed. • 75% Nitrate removed. • 99% Phosphoruos removed. • 99% Ammonia removed. • 30% microalgae concentration achieved highest efficiency in nutrient removal.	(Priyadarshi, 2021)

Different ratios from microalgae:activated sludge were used. 1:0, 1:1, 3:1, 9:1, 0:1 (microalgae:activated sludge). With hydraulic retention time 11 days in PBR, 100 rpm mixing, 100 μmol/m².s.	Study the affect of using different ratios from microalgae:activated sludge on efficiency of nutrients removal from wastewater.	• The ratio of 3:1 microalgae:activated sludge was the optimum ratio for nutrient removal. • 99% COD removed. • 79% TP removed. • 86% TN removed.	(Nguyen et al., 2020)

جـامعـــة النجاح الوطنية
كليـــة الدراســـات العليـــا

استخدام الطحالب لإزالة المغذيات من المياه العادمة: تطبيق ضوء اصطناعي لتعزيز نمو الطحالب

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قدمت هذه الرسالة استكمالا لمتطلبات الحصول على درجة الماجستير في هندسة المياه والبيئة، من كلية الدراسات العليا، في جامعة النجاح الوطنية، نابلس– فلسطين.

2024

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الملخص

تعتبر معالجة مياه الصرف الصحي المنزلية باستخدام الطحالب لإزالة العناصر الغذائية من أنجح الطرق الطبيعية الصديقة للبيئة وغير المكلفة، حيث تنمو الطحالب بشكل طبيعي في مياه الصرف الصحي إذا توفرت الظروف المناسبة لنموها، مثل العناصر الغذائية ودرجة الحرارة ومدة وشدة التعرض للضوء، وكذلك وجود البكتيريا التي تشكل علاقة تكافلية مع الطحالب لمساعدتها على النمو. وفي هذا البحث تم دراسة تأثير الضوء الصناعي على نمو الطحالب ونشاطها في إزالة العناصر الغذائية من مياه الصرف الصحي المنزلية. وقد تم استخدام ثلاثة أكواب من مياه الصرف الصحي المنزلية التي نمت فيها الطحالب، بحجم 5 لترات كمقياس تجريبي لمفاعلات الدفعة، ووضعت في نفس الظروف ودرجات الحرارة والخلط لمدة 15 يومًا، مع فترات متفاوتة من التعرض للضوء الصناعي. في العينة الأولى، تعرضت الطحالب للضوء 24 ساعة يوميًا وسميت بالعينة الضوئية. تعرض الكأس الثاني للضوء لمدة 19 ساعة بما فيها فترة الليل، و 5 ساعات تعرض للضوء النهاري الطبيعي وسميت العينة نصف الضوئية، والعينة الثالثة بقيت في دورة الليل والنهار حيث سميت العينة الطبيعية. وبعد 15 يوما من مقارنة العينات الثلاث، ورغم أن النتائج كانت متقاربة في العينتين الضوئية ونصف الضوئية، إلا أن الطحالب في العينة الضوئية كانت الأكثر كفاءة في إزالة العناصر الغذائية من مياه الصرف الصحي، حيث بلغت كفاءتها في إزالة COD (95%) والفوسفور (96%). كما بلغت نسبة الأمونيوم (99.8%) وتركيز المواد الصلبة الذائبة الكلية 737.4 ملجم/لتر، وتركيز صبغة الكلوروفيل أ 129.4 ميكروجرام/لتر. وفي العينة نصف الضوئية كانت فعالية الطحالب في إزالة

COD(93%) والفوسفور (94%) والأمونيوم (99.8%) وتركيز المواد الصلبة العالقة 237.4 ملجم/لتر وتركيز الكلوروفيل أ 57.2 ميكروجرام/لتر. وأخيراً في العينة الطبيعية كانت فعالية إزالة COD (93%) والفوسفور (6%) والأمونيوم (56%) وتركيز المواد الصلبة العالقة 174 ملجم/لتر وتركيز الكلوروفيل أ 8 ميكروجرام/لتر. وتوضح هذه النتائج فعالية الطحالب كطريقة طبيعية لإزالة العناصر الغذائية من مياه الصرف الصحي والتأثير الإيجابي للضوء في تحسين نمو الطحالب وكفاءته في عملية المعالجة.

الكلمات المفتاحية: الطحالب، معالجة مياه الصرف الصحي، إزالة العناصر الغذائية، الضوء الاصطناعي، نمو الطحالب.