Purification of wastewater by Algae

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Abstract

The two micro algae chlorella & Scenedesmus were used in purification of wastewater with investment of sludge bacteria existing in the same water.

The two algae were isolated from Tigris river startly them grown in medium prepared in the lab independent on the water of the river as a basic medium in order to use these growths as algal inoculums in the later experiments.

These algae were tried to grow on the samples of wastewater in Baghdad to produce a biomass utilized as a source of food and feed. Lab experiments were achieved to study the effect of temperature, conc. of salt and shaking period on the extraction of protein taken from the mass of algal growth.

The conc. of NaCl = 0.6M gave a high protein percentage 52% among the other molar concentrations studied (0.2, 0.4, 0.6, 0.8, 1.0M). The Temperature 40C^o gave a higher protein percentage 51% among the other temperature degrees studied (20, 25,30,35,40,45 C^o), then the shaking period 12h gave a high percentage protein 55% among the periods studied (3,6,12,18,24,).

The 55% protein was the highest value in the all experiments applied in this study.

A field experiments were worked to clearance the wastewater and storage it in precipitation pool then in the primary settling basin, then to accelerating pool, finally to the secondary clarifier pool. Chemical

Oxygen Demand (COD) was determined in these pools to point the properties of wastewater , COD value decrease from 697 mg/L in the wastewater before enter first pool to 61 mg/L in the final pool treatment. The algae were concentrated in the secondary clarifier pool then were isolated and analyzed to determine the crude protein % content as 49.8% this protein percentage was qualified the protein in the eggs and beef, which to encourage to use these algae in food and feed. Also they contained 2.3% as phosphate and other important minerals.

The efficiency of the pool of treatment in purification of wastewater. BOD was determined which found decreased from 140 mg/L before the first pool to 9 mg/L after the treatment. In this way the efficiency was 93.6% as BOD removal. This efficiency was higher than three times of the BOD standard value worldly 30 mg/L. These treatments improved the advantage in the purification of wastewater and could reuse it in purposes of irrigation and drinking.

INTRODUCTION

Wastewater treatment is necessary before wastewater can be disposed of without producing significant undesirable or even harmful effects. However, some communities and municipalities still dispose of inadequately treated wastewater into natural bodies of water, either because they are indifferent to the consequences or because it is assumed that the body of water is sufficiently large and so located that dilution prevents hazards. Communities and municipalities can no longer rely on disposal of wastewater by dilution. There is an ever-increasing demand for domestic and industrial water, necessitating more reuse of waters that receive wastewater. [(5)]

Domestic wastewater or sewage consists of approximately 99.9 percent water, 0.02 to 0.03 percent suspended solids, and other soluble organic and inorganic substances. On a percentage basis, the amount of

solids appears small; however, the tremendous volume of material handled daily by a major municipal plant (e.g., several hundred millions of gallons) contains as much as 100 tons of solids. The chemical constituents, present in low concentration, nevertheless are extremely important and are subject to variations, between communities as well as within a community, even from hour to hour. Inorganic chemicals initially present in the water supply with likewise are present in the sewage; organic compounds are contributed through human excrement and other domestic wastes, and both organic and inorganic compounds are added by industrial wastes. For example, slaughterhouses, sugar factories, paper mills, and creameries add organic substances; mines and metal industries contribute acids and salts of metals and other inorganic wastes. The organic compounds in sewage are classified as nitrogenous or no nitrogenous. The principal nitrogenous compounds are urea, proteins, amines, and amino acids; the no nitrogenous substances include carbohydrates, fates, and soaps. **(4)**

Since the composition of wastewater varies, it is to be expected that the types and numbers of organisms will fluctuate. Fungi, protozoa, algae, bacteria, and viruses are present. Raw sewage may contain millions of bacteria per milliliter, including the coliforms, streptococci, anaerobic spore-forming bacilli, the proteus group, and other types originating in the intestinal tract of humans. Sewage is also a potential source of pathogenic protozoa, bacteria, and viruses. The causative agents of dysentery, cholera, and typhoid fever may occur in sewage. The poliomyelitis virus, the virus of infectious hepatitis, and the Coxsackie's viruses are excreted in the feces of infected hosts and thus may appear in sewage. Certain bacterial viruses are readily isolated from sewage. **(5)**

Predominant physiological types of bacteria may shift during the course of sewage digestion. In an anaerobic digester, facultative types

(*Enterobacter, Alcaigenes, Escherichia, Pseudomonas*, etc.) predominate during initial stages.

This is followed by methane producers, which are strict anaerobes, e.g., *Methanobacterium, Methanosarcina,* and *Methanococcus* .Organic acids produced by the facultative bacteria are metabolized by the methane formers; the end products are methane and carbon dioxide. Large amounts of these gases are produced in anaerobic digesters. **(6)**

The biochemical oxygen demand (BOD) is a measure of the amount of oxygen used in the respiratory processes of microorganisms in oxidizing the organic matter in the sewage and for the further metabolism (oxidation) of cellular components synthesized from the wastes. One of the primary reasons for treating wastewater prior to its being returned to the water resource (e.g., stream or lake) is to reduce the drain on dissolved oxygen supply of the receiving body of water .The magnitude of the BOD is related to the amount of organic material in the wastewater-i.e., the more oxidizable organic material, the higher the BOD.The "strength" of wastewater is expressed in terms of BOD level.**(1)**

Biological treatment processes accomplish oxidation of the organic material in the liquid wastewater by microbial activity. The oxidation methods employed are: First, filtration by intermittent sand filters, contact filters, and trickling filters. Second, aeration by the activated-sludge process or by contact aerators. Third, oxidation ponds that are shallow ponds (20 to 60 cm in depth) designed to allow algal growth on the wastewater effluent. Use of oxidation ponds should be preceded by primary treatment. Oxygen for biochemical oxidation of nutrients is supplied from the air, but the release of O_2 during photosynthesis by the algae *Chlorella & Scenedesmus* provides an additional important source of oxygen **(1).** This procedure was applied in this study.

The aim of this study is to use the micro algae with sludge bacteria to purify treatment and reduce BOD of wastewater, that to make it as sufficient water to drinking or irrigation then aiming to re-extraction nutritional substances from the wastes by getting the algae that completed its role in the first stage.

The Methods:

The algae *Scenedusmus* spp. and *Chlorella* spp. were taken from laboratories of biochemical Technology branch/university of Technology.

These microalage were growing by using lighting incubator. The medium of growth was prepared as following: 20.0mM $NaHCO_3$, 5.0mM KNO_3, 5.0mM $MgSO_4$, 0.4mM KH_2PO_4, and 0.3mM CaCl2, 1.5mM FeCl3, 6.0mM EDTA, 0.0-1.0 M NaCl.

Those quantities were prepared in different concentrations of salt (NaCl). The development of growth was controlling by counting method using Haemocytometer. pH was supervised by pH-meter that was 7.0 – 6.8 at the starting of experiments .The growths were preserved as an inoculums for the next experiments.

Another experiments were achieved to grow these two algae by using water of Tigris river from middle of Baghdad, this water was analyzed to detect the nutritional minerals as reference[(2)]. The composition of this water was corrected by adding the deficient chemicals to be use as nutrient media for algal growth. The samples of Tigris river were taken from position of hanged bridge (Karada).

On the other hand the algae were tested to grow on the samples of domestic wastewater from Rustumia position, South of Baghdad, in where the wastewater of the city is treating. The genera *Chlorella* and *Scenedesmus* were dominant in this wastewater.

The dry weights of algal samples were determined according to [9], also the total nitrogen was determined by microkjeldahl method[2] whereas the BOD, COD, TSS and phosphate were determined according to [2]. The crude protein percentage of fungal growths was determined by kjeldahl method according to AOAC [2] as:

Crude protein % = Total Nitrogen % x 6.25

The protein was gained from the biomass of algal growths as mixture of the two algae *Scenedesmus* spp. & *Chlorella* spp. in ratio of 1:8. We studied the effect of temperature, salt concentration and time period on protein percentage which gained from the biomass of these algae. [4]

Laboratories Experiments:

In order to know the effect of different NaCl conc.(0.2,0.4, 0.6, 0.8, 1.0)M on extract the best protein percent from algae , we tried to extract algal protein in a medium in which parameters were constant at T= 30 C^0 and shaking period =3h .

Then to study the effect of different temperature (20, 25, 30, 35, 40, 45) C^0 on the best extract of algal protein we try to stabilize NaCl conc. = 0.6 M and shaking period = 3 h.

And to study the effect of different shaking periods of algal culture (3, 6, 12, 18, 24) hours, we stabilized the previous optimum parameters as (NaCl = 0.6 M and T = 40 C^o).

Field Experiments:

The domestic sewage was to flow through the sieves to remove the solid, coarse and fine materials then stored in the primary setting basin for 3 hours. After that, the water run to acceleration basin in which stay to 3-4 days to fall into a behind region in the basin in which the water rotates continuously with velocity 15 cm/ sec. to prevent sedimentation of algae in the bottom of the basin and to stay in the surface exposure to the

sunlight which needed by algae to produce an oxygen which demanded by bacteria to do the biological processes on the organic wastes and to purify the water. After that the water went to the secondary setting basin as in the figure 1.

The algae were then concentrated in the secondary setting basin and isolated to analyze a sample of it as in the following method: dry sample was solidified for 4 hours on 550-600C^{o} in quartz container. The ash was hydrolyzed in 5.5N HCl. The concentrations of metals was determined by using atomic absorption system, where as the sodium and potassium were extracted from dried algae for 30 minutes in solution of 1 N ammonium acetate, pH = 7.0. After filtration, the concentration of sodium and potassium were detected by system of flame photometer. The standard methods in the reference **(3)** were used to achieve other analyses.

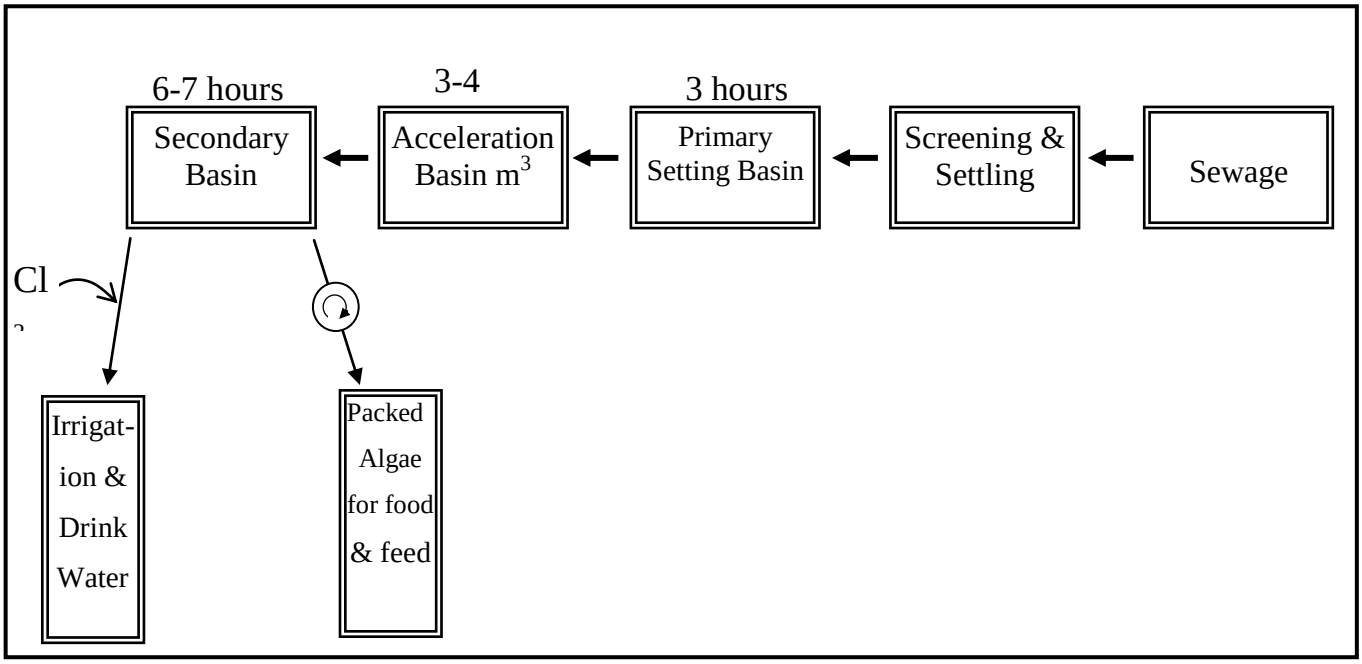

Fig.1:**Diagram of domestic wastewater treatment in which the field experiments were done**

An experiment was done to know characteristics of wastewater in different stages of purification. The BOD values were selected to do this comparation between the crude sewage before entered the basin, and the water in the primary setting basin, acceleration basin and secondary

basin. Whereas a final experiment was done to know the efficiency of treatment basin in the purification and in the removing of BOD, this was made by determination of BOD conc. in the water enter the basin or the water exit it. Also measurement of total suspended solids (TSS), total N-N and to phosphorus as (PO_4^{-3}).

Results and Discussion:

Table 1 shows the chemical constituents of minerals of Tigris river. River water was utilized as a nutrient medium for algal growth after correct the constituents and used to finish the laboratory experiments. It appeared clearly from the table 1 that the water of the river had important minerals such as phosphorus and nitrogen more than demanded, this was good economically.

Table 1: Mineral constituents of Tigris river

Material	Concentration
Ca	350.0 mg/L
Mg	422.0 mg/L
Fe	0.06 mg/L
HCO_3^-	141.0 mg/L
$NO_3 - N$	10.0 mg/L
$PO_4 - P$	1.0 mg/L
SO_4^{-2}	3240.0 mg/L

Fig 2 explained the effect of NaCl concentration, temperature and shaking period on the ratio of crude protein in the algae which was growing in the media of wastewater which prepared in the lab. The figure appeared that the concentration of NaCl = 0.6 M gave a higher concentration of protein 52% comparably with other concentrations.

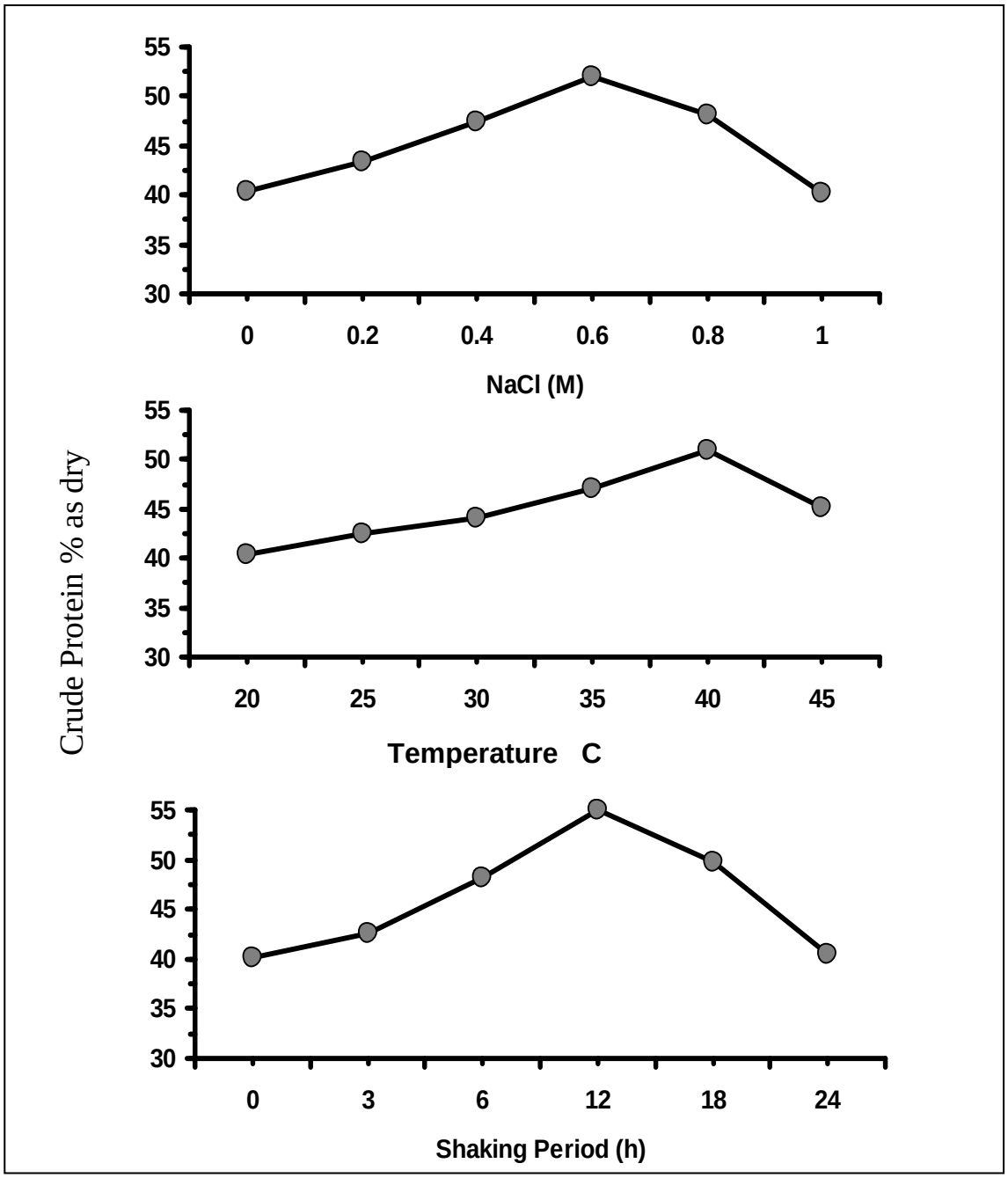

Fig. 2: Effect of NaCl concentration, temperature, and shacking period on getting of crude protein of algae growing on wastewater

The temperature = 40C^{o} gave a higher concentration of protein 51%. The experiments of effect of shaking period showed that the period = 12 hours gave a higher concentration of protein 55% which considered as a highest concentration of protein gained in these experiments. Theses

ecological parameters affected the results of protein concentrations because it's physiological effects on the growth of algae and on biosynthesis of protein inside the alive cells[8] .

It showed in table-2 the chemical composition of algal biomass growing in sewage, it appeared a ratio of crude protein 49.8%, this high

Table 2: Composition of algae growing on wastewater

Concentration g/100g dry weight	Constituents
49.8	Crude Protein
6.0	Ash
1.8	Ca^{++}
2.3	PO_4^{-3}
1.0	K^{+}
0.2	Fe^{++}
0.2	Na^{+}

eggs 48.8%, and to be like protein ratio in muscular meat 57.1%[7]. In addition to content phosphorus and other mineral salts which considered as important in nutrition of man and domestic animals, these algae can be fructified in feeding of it. The contents of algal biomass are considered as a high level nutrition compared with commercial sing-cell protein of yeast, fungi and bacteria that used in global zone.

In table-3 could be seen decrease COD from 697 mg/L in the original wastewater to 61 mg/L in the setting basin , and to 70 mg/L in the treatment basin , this ratio was good to reclaim the polluted water and make it to have good properties and unharmful.

Table 3: BOD values of purification stages of wastewater in basin of experiment

Conc. mg/L	Purification stages
697	Raw Sewage
388	Primary Settling Basin
70	Treatment Basin
61	Secondary Clarifier

Table-4 explained the properties of water coming out the basin after treated by algae .The results appeared to be agreed with conditions which detected by Environmental Protection Agency (EPA) , in the world law at 1972 in Clean Water Act [(3)] .

These conditions are (30, 30, 30) – i.e.:

30 mg/L maximum value of BOD (Biological oxygen demand).

30 mg/L maximum value of VSS (Volatile suspended substances).

30 cell/mL maximum value of MPN (Most probable number of bacteria in water).

Table 4: Efficiency of treatment basin in purification of wastewater

Analysations	Inlet water mg/L	Outlet treated water mg/L	Displacement %
BOD consumption	140.0	9.0	93.6
TSS	120.0	12.0	90.0
Total N – N	23.0	0.8	96.5
Total P- PO_4^{-3}	36.0	2.8	92.2

The BOD value in the table 4 was less than third part of the maximum demanded value (30 mg/L) that reached after treatment to 9 mg/L only; this meant the efficiency of treatment basin was 93.6% in

remove of BOD from the treated water that the BOD decreased before basin from 140 mg/L to 9 mg/L.

The efficiency in remove TSS was 99.0% to decrease it from TSS = 120.0 mg/L to 12.0 mg/L after treatment and to be not cause hard water. Also the high removing of total nitrogen and total phosphorus were 96.5% and 92.2% respectively .The nitrogen and phosphorus would encourage the bacteria polluting water, if existed in high ratio, but the treatment decrease it into low values 0.8 mg/L and 2.8 mg/L respectively.

These results encouraged to reuse the water resulting from this treatments in order to refructify in the irrigation or in the drinking (after sterilization) in the regions that had little water.

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تنقية المياه الثقيلة باستخدام الطحالب

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الخلاصة

تم استخدام الطحالب الدقيقة *Chlorella & Scenedesmus* في تنقية المياه الثقيلة مع استغلال بكتريا الوحل *Sludge* الموجودة في المياه نفسها لأتمام المهمة .

وقد تم في البداية عزل هذين الطحلبين من مياه دجلة ونميا في وسط محضر في المختبر بأعتماد مياه النهر كوسط أساس لغرض استعمال هذه النموات كلقاح طحلبي في التجارب اللاحقة.

ثم جربت تنمية هذه الطحالب على عينات من مياه ثقيلة في بغداد للأستفادة من الكتلة الحيوية النامية كمصدر غذائي بروتيني يفيد التغذية والأعلاف . وقد أجريت تجارب مختبرية لدراسة تأثير تغير تراكيز الملح ودرجة الحرارة وفترة الرج في النسبة المئوية للبروتين المستحصل من كتلة الطحالب النامية.

فوجد ان تركيز NaCl = ٠,٦ M أعلى نسبة بروتين ٥٢,٠ % من بين التراكيز المولارية المجربة (٠,٢ ، ٠,٤ ، ٠,٦ ، ٠,٨ ، ١,٠) مولاري . وأعطت درجة الحرارة ٤٠ مْ أعلى نسبة بروتين ٥١,٠ % من بين درجات الحرارة المدروسة (٢٠ ، ٢٥ ، ٣٠ ، ٣٥ ، ٤٠ ، ٤٥) مْ . أما فترة الرج لمزارع الطحالب فقد أعطت الفترة ١٢ ساعة أعلى نسبة بروتين ٥٥

% من بين الفترات المعتمدة في الدراسة وهي (٣ ، ٦ ، ١٢ ، ١٨ ، ٢٤) ساعة وكانت هذه النسبة ٥٥ % بروتين هي الأعلى من بين كل التجارب المنجزة لدراسة المتغيرات المؤثرة في انتاج البروتين من الطحالب في هذه الدراسة .

كما أجريت تجارب حقلية تم فيها تصفية المياه الثقيلة ثم حجزها في حوض للترسيب ثم في حوض الترقيد الأولي ثم الى حوض التسريع وأخيرا الى حوض الترقيد الثانوي ، حيث تم تقدير قيم COD في هذه الأحواض لمعرفة مواصفات المياه الثقيلة فهبطت قيمة COD من ٦٩٧ ملغم/لتر في المياه الثقيلة قبل دخول الحوض الاول الى ٦١ ملغم/لتر في حوض المعالجة الاخير. جرى تركيز الطحالب في حوض الترقيد الثاني وتم فصلها وتحليل عينة منها فكان محتواها من البروتين الخام ٤٩,٨ % وهذا يضاهي ما موجود في البيض واللحم من بروتين مما يشجع في استغلال هذه الطحالب في التغذية والاعلاف . كما احتوت على ٢,٣ % فوسفات وغيرها من المعادن الاخرى المهمة في التغذية .

ثم درست كفاءة حوض المعالجة في تنقية المياه ، فقدرت قيمة BOD فوجد انها هبطت من ١٤٠ ملغم/لتر قبل الحوض الاول الى ٩ ماغم/لتر بعد المعالجة. وبهذا تكون نسبة ازالة BOD ما يعادل كفاءة ٩٣,٦ % . وقد كانت هذه الكفاءة أعلى ثلاث مرات تقريباً من القيمة المطلوبة عالميا للـ BOD والبالغة ٣٠ ملغم/لتر . فاثبتت هذه المعاملات جدواها في تنقية المياه الثقيلة وامكانية اعادة استعمالها لأغراض الري والشرب.