

## Toxicity of Al-Dura Oil Refinery Wastes Towards Some Freshwater Phytoplanktons

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### ABSTRACT

Petroleum-derived hydrocarbon wastes are one of the most dangerous aquatic environmental pollutants, the production and export of oil are regarded as the main sources of these wastes. Discharging of the oil refinery wastes to the aquatic ecosystems can cause hazardous and harmful effects to its food chain levels especially algae, depending on the released concentrations. The present study experiments were conducted with axenic culture of the green algae *Chlorella vulgaris* and *Scenedesmus dimorphus*. Different concentrations of the oil wastes (25, 50, 75 and 100 %) from three selected locations (SO1, SO8 and SO12) at the refinery treatment unit of Al-Dura refinery were prepared.

Decreasing in the algal growth rates associated with increasing in the doubling time of the cells were detected for the both strains when treated with tested concentrations of the oil refinery during the exposure period that took 96 hr. The reduction was clear with *C. vulgaris*, but it was gradual in the case of *S. dimorphus*. An accelerating increasing in the algal growth inhibition averages accompanied with increasing in the wastes concentrations as well as time of exposure. The differences in the calculated EC50 values for both strains indicate differences in the toxic effects of the oil wastes in addition to their sensitivity towards such pollutants.

**Keywords:** Toxicity; Algae; Batch culture; Refinery waste; Growth rates; Biotest.

### سُمِّيَة مَخْلُفَات مَصْفَى الدَّوْرَة النِّفْطِي فِي بَعْض هَائِمَات المِيَاه العَذْبَة

#### الخلاصة :

تُعتبر المَخْلُفَات النِّفْطِيَة أخطر الملوّثات البيئيّة في البيئَة المائيّة، حيث تمثّل عمليات إنتاج النفط وتصديره أهم مصادر التلوّث بهذه المَخْلُفَات. بالإضافة إلى ذلك فإن لدَفَق مَخْلُفَات مَصْفَى إنتاج المشتقات النِّفْطِيَة المطروح في البيئات المائيّة قد يكون سبباً في إحداث تأثيرات حادّة وخطرة في السلاسل الغذائيّة المائيّة بشكل عام وفي مجاميع الهائمات النباتيّة كمستوى إغذائيّ بشكل خاص إعتدالاً على التراكمات المطروحة لهذه المَخْلُفَات. تضمنت الدراسة الحاليّة تقييم الأثار السُمِّيَة الحادّة لمَخْلُفَات مَصْفَى الدَّوْرَة النِّفْطِي فِي سلاّتين للهائمات النباتيّة *Chlorella vulgaris* و *Scenedesmus dimorphus* تابعين إلى قسم الطحالب الخضراء من خلال تعريضهما إلى تراكيز عديدة لمَخْلُفَات نِفْطِيَة (25، 50، 75 و 100%) تنتمي إلى مراحل معالجة مختلفة (SO1, SO8, SO12) ضمن وحدة معالجة المَخْلُفَات النِّفْطِيَة فِي المَصْفَى.

أظهرت نتائج البحث الراهن إنخفاضاً ملحوظاً في معدلات نمو الهائمات المدروسة والذي تزامن مع زيادة في زمن تضاعف هذه الكائنات عند معاملتها بتراكيز المَخْلُفَات المحضّرة خلال فترة تعريض

حاد إمتدت الى 96 ساعة. وتجدر الإشارة الى أن الإنخفاض في نمو الكائنات المجهرية المختبرة كان على أشده في الطحلب الأخضر الأحادي الخلية *Chlorella vulgaris*، بينما أظهر نظيره *Scenedesmusdimorphus* تناقصاً تدريجياً في النمو. وفي المقابل تم تشخيص زيادة مُضطردة في معدلات تثبيط نمو الطحالب والتي أقرنت بزيادة كل من التراكمات المختبرة فضلاً عن تقدم زمن التعريض. وفي نفس السياق، فإن نتائج حساب متوسطات التراكمات الفعالة لكلا السلالتين المختبرتين تؤكد وجود إختلافات في التأثيرات السُمية للمخلفات النفطية إعتياداً على مرحلة المعالجة بالإضافة الى حساسية الأحياء المختبرة في الدراسة تجاه المادة الملوثة.

## INTRODUCTION

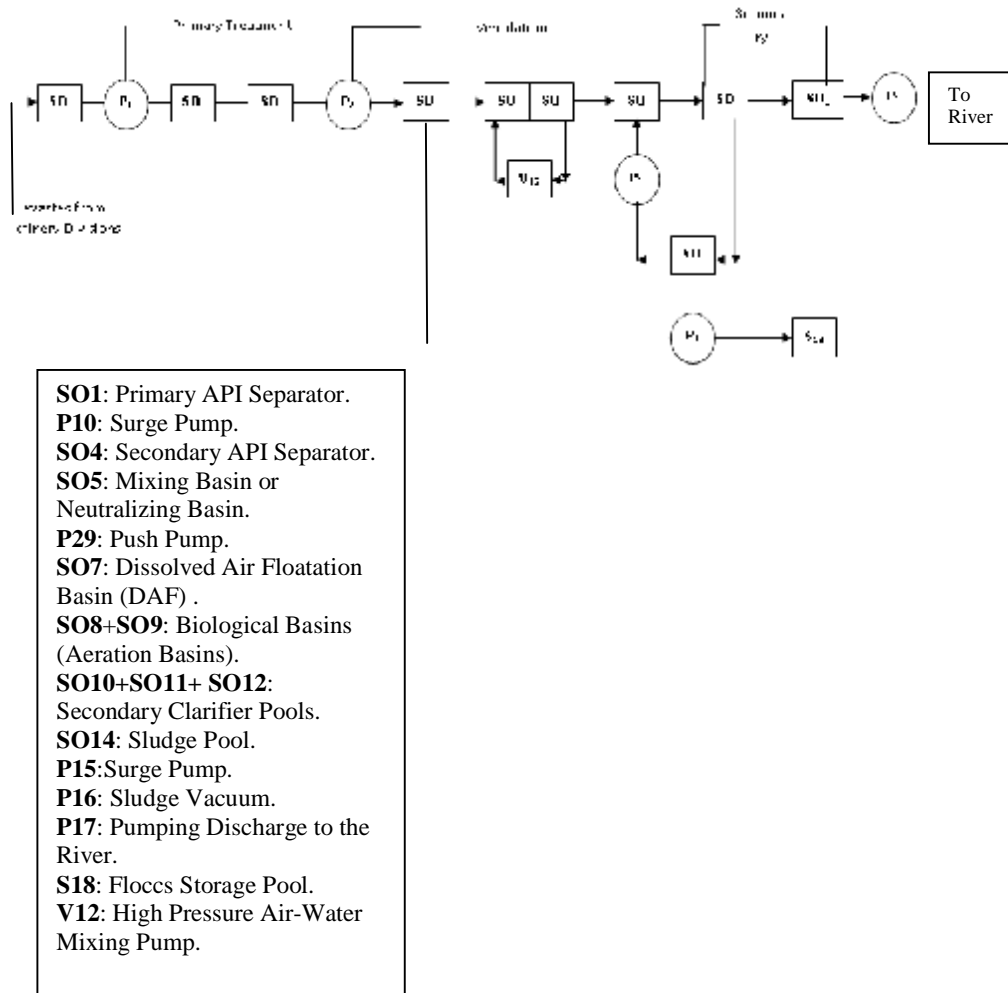
The fast expanding of petroleum and petroleum products industries has inevitably resulted in the discharge of oil wastes to the environment and became a source of pollutants entering the aquatic ecosystems throughout the world. The annual influx of petroleum into the marine environment is estimated to be between 1.1-7.2 million metric tons [1]. In addition, it was estimated [2] that 28%-30% of spilled oil enters freshwater environment. Oil refinery wastes release high levels of hydrocarbons to water; in addition to these, natural seepage from ground and human industrial activities other than petrochemistry are also considered sources of dangerous wastes [3]. Although, there is increasing interest in using algae as applicable tools for self-cleaning and bioremediation as well as bioindication of a polluted environment [4], Till now, a little data are available that related with such applications specially with complex wastes, in comparison with the role of bacteria in the biodegradation of the industrial effluents. Refined petroleum products, particularly fuel oils, has been reported to be more toxic to microalgae than crude oil [5, 6]. Likewise, the toxic effects of the oil refinery wastes was documented in *C. pyrenoidosa*, *Oocystis pusilla* and *Oscillatoria quadripunctulata* by using bioassay [7]. From other side, the increasing in the phytoplankton biomass influence the biogeochemical cycle of persistent organic pollutants such as polychlorinated biphenyls (PCBs) in aquatic environments [8]. A considerable studies on algal communities in respect to oil pollution has been studies [9, 10, 11, 12]. Recently, Algal bioassay consider an indispensable part of the test batteries in water pollution monitoring as a result to the ecological role for these microorganisms that playing in the aquatic ecosystems as a primary producers of the food chain, as well as their sensitivity towards water contamination rather than fish or invertebrates [13]. The purpose of this study is to detect the toxicity of treated and non-treated wastes of Al-Dura oil refinery (Iraq, Baghdad) in the tow isolated species *Chlorella vulgaris* and *Scenedesmus dimorphus* and to demonstrate the usefulness of algae for monitoring the effectiveness of the industrial effluent treatment as well as oil pollution in aquatic ecosystems.

## Material and Methods

### Sampling procedure

The wastewater treatment unit of Al-Dura oil refinery incorporates many treatment pools such as API separators for oil removal, mechanical, chemical and biological treatment pools (Fig.1). The effluent exposing to different treatment stages through passing in the parts of the system and finally discharge into a nearby stream that opens to the adjoining estuary.

Waste samples from three locations (SO1; SO8; SO12) at the treatment unit were collected in June 2013. The physico-chemical parameters for the (SO1 and SO12) were analyzed at the wastewater treatment unit laboratory according to standard methods [14], to detect the effective wastes concentrations at the wastewater treatment column and for toxicological impact assessment of the effluent on the tested algae (Table.1).



**Figure. (1) Sketch represents main components of wastes treatment unit for Al-Dura refinery (Iraqi ministry of oil/Midland refineries co. / AL-Dura refinery).**

**Table (1) Physico-chemical parameters for SO1 and SO12 pool wastes at Al-Dura oil refinery.**

Variables	Unit	SO1	SO12
Temperature	(°C)	32.00	29.00
pH	H	7.5	7.6
TDS	(mg/L)	1355	1392
TSS	(mg/L)	379	26
Sulphide	(mg/L)	0.34	0.017
Oil	(mg/L)	131	2
COD	(mg/L)	377	36
BOD	(mg/L)	34	8
Phenols	(mg/L)	2.2	0.022
PO4	(mg/L)	0.95	0.17
SO4	(mg/L)	254	307
N-NO2	(mg/L)	0.07	0.025
Turbidity	NTU	85	6.1
Fe	(mg/L)	1.5	0.285
Dissolved oxygen	(mg/L)	4.640	5.9

### Growth culture

All experiments were conducted with axenic culture of the green algae *Chlorella vulgaris* (Smith) beijerinck and *Scenedesmus dimorphus* (Turp.) kutzing (Chlorococcales, chlorophyta). The stock culture was obtained from Algal Unit of Water Research Center in Ministry of Science and Technology which were already isolated from Tigris-river (Baghdad- Iraq). The cultures were grown in Chu-no.10 medium according to [15] with modification made by [16, 17] at  $28 \pm 2^\circ\text{C}$  and light intensity ( $\sim 2500$  Lux) which was provided by white fluorescent lamps, under a light/dark regime of 16/8 hours for the duration of the experiments. After detecting the nutrients concentrations (N, P) in the wastes samples (Table.1), An optimal phytonutrient concentrations (10 mg/l nitrate; 5 mg/l phosphate and 1:10 mg/l N:P ratio), were calculated and added to both of control and treatments culture mediums as optimal chemical conditions in order to obtain higher growth rates and lower doubling time of cells. The buffered culture medium was finally adjusted to pH 7 with NaOH. The medium for preculture was autoclaved in 1000-ml polycarbonate flasks. Patterson's method was used to purify the culture to get an Axenic culture [18]. Growth of the microalgal cultures was measured daily along exposure period (96 hr) by counting culture aliquots in a Neubauer haemocytometer. For determination of chlorophyll-a, the procedure recommended by [19] was used, using 90% methanol as extraction solvent. Calculations were done using Lorenzen's equation (Eq. 1).

$$\mu\text{g Chl.-a/sample} = 11.9 [2.43 (\text{Db-Da}) \text{ V/L} \dots (1)$$

Where:

$\mu\text{g Chl.-a/Sample}$  = Chlorophyll-a concentration ( $\mu\text{g/ml}$ )

Db = Light density for Chl-a extraction before adding HCl at (665,750 nm)

Da = Light density for Chl-a extraction after adding HCl at (665,750 nm)

V = solvent volume

L = Light cell (Cuvette) length (cm)

### Algal bioassays

The collected waste samples from the three locations (SO1; SO8 and SO12) were filtered through Millipore filter paper (0.45  $\mu\text{m}$ ) and kept at 4°C to use in bioassay. A set of cultures was simultaneously raised in the maintenance medium. Different concentrations of the oil wastes (25, 50, 75 and 100%) were prepared by using 250 mL sterile conical flasks in triplicate and inoculated with  $1 \times 10^6$  cells/ml of the algal culture at the exponential phase of the growth for both of *C. vulgaris* and *S. dimorphus*. For control, algae were just incubated in culture medium. Determination of the algal biomass in presence or absence of the oil wastes, expressed as a specific growth rates which derived from both of the cells number counting ( $\mu$ ) and chlorophyll-a concentrations (K), in addition to the doubling time of cells (G) [20] (Eq. 2, 3). Also, growth Inhibition (GI %) as another indicator for the algal response towards oil wastes was calculated according to [21] (Eq. 4). The median effective concentrations (EC50) for the oil wastes were detected to identify the concentrations that causing the death for 50% of the tested algae after 96 hr. of exposure [22].

$$\text{Growth rate } (\mu \text{ or } K) = [\ln(X_2/X_1) / (t_2-t_1)] (\text{day})^{-1} \quad \dots(2)$$

Where:

$X_1$  = cell number per ml (cell $\times 10^6$  /ml) or Chl-a concentrations per ml ( $\mu\text{g/ml}$ ) at time T1

$X_2$  = cell number per ml (cell $\times 10^6$  /ml) or Chl-a concentrations per ml ( $\mu\text{g/ml}$ ) at time T2

$$\text{Doubling time (G)} = \ln 2/K \quad \dots(3)$$

$$\% \text{ GI} = \{(T-C)/C\} * 100 \quad \dots(4)$$

Where:

GI: Growth Inhibition (%).

T: number of cells/ml in treatment culture.

C: number of cells/ml in control culture.

### Statistics

For assessment of the observed variance between control and treatments, a one-way statistical analysis of variance ( $P < 0.05$ ) in conjunction with Duncan's multiple range test was done also. Correlation factor was determined by Simple Linear Regression Equation [23].

### RESULTS

The bioassay results showed clear differences in the algal growth between treatments for both species *C. vulgaris* and *S. dimorphus*, when exposed to different concentrations of the refinery wastes from the studied locations during 96 hr.

The growth rates of the tested algae decreased in concentrations (25, 50, 75 and 100 %) respectively, during the exposure period. The reduction was clear in *C. vulgaris* with all concentrations and for all studied locations. Growth rates during 96 hr. at SO1, SO8 and SO12 (100%) reached to ( $\mu=0.224\pm0.0281$ ,  $0.231\pm0.0162$ ,  $0.301\pm0.0155$  respectively) (Fig.2). Similarly, was observed with respect to *S. dimorphus* at location (SO1), while gradually decreased with samples from SO8 and SO12 at all studied concentrations as long as exposure took place. The lowest growth values for the studied locations at 100% were ( $\mu=0.316\pm0.02$ ,  $0.391\pm0.027$ ,  $0.411\pm0.025$  respectively) (Fig.3).

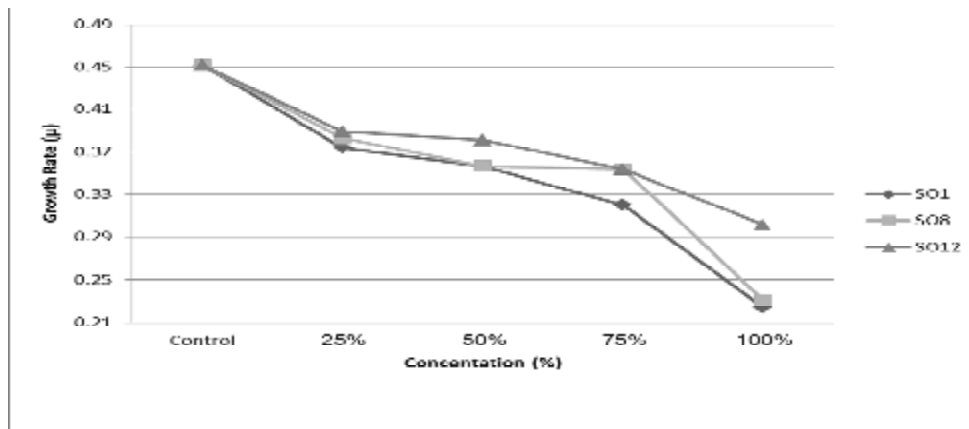
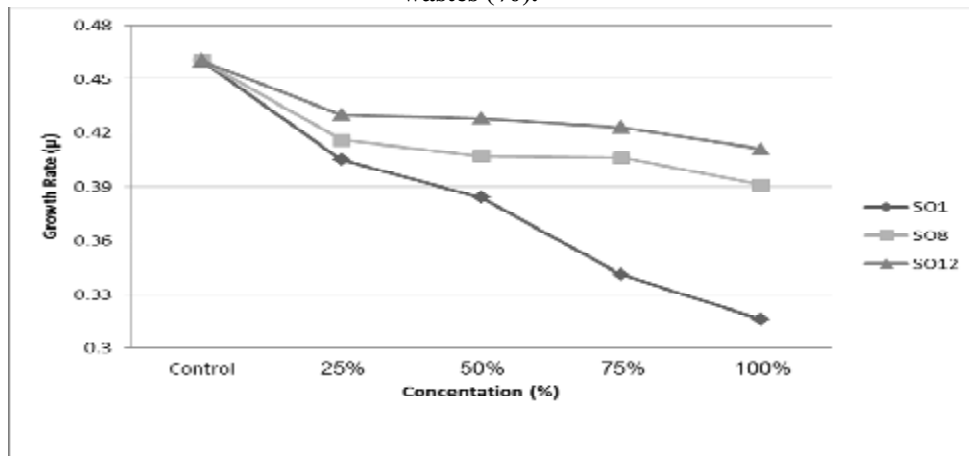


Figure (2) growth rate of Chlorella vulgaris based on the cells number counting when exposed to different concentrations of SO1, SO8 and SO12 wastes (%).



Figure(3) growth rate of Scenedesmus dimorphus based on the cells number counting when exposed to different concentrations of SO1, SO8 and SO12 wastes (%).

A reversed effect relationship was detected between growth rates and doubling time of cells, Growth rates decreased with increasing doubling time when algae exposed to increasing concentrations as well as time of exposure (Tables 2,3),

Whereas the waste toxicity increased with increasing of the concentrations in addition to exposure period in comparison with control for both species.

Generally, it could be noting that the samples from SO1 was the most toxic for both of species (*C. vulgaris*:  $K=0.234\pm 0.001$ ,  $G=30.22\pm 0.001$ ), (*S. dimorphus*:  $K=0.311\pm 0.001$ ,  $G=23.17\pm 0.009$ ), whereas SO12 was the least (*C. vulgaris*:  $K=0.308\pm 0.017$ ,  $G=23.89\pm 0.271$ ), (*S. dimorphus*:  $K=0.409\pm 0.005$ ,  $G=17.635\pm 0.025$ ).

**Table (2) Growth rates (K) and doubling times (G) of *Chlorella vulgaris* based on chlorophyll-a concentration with respect to the wastes concentrations (%).**

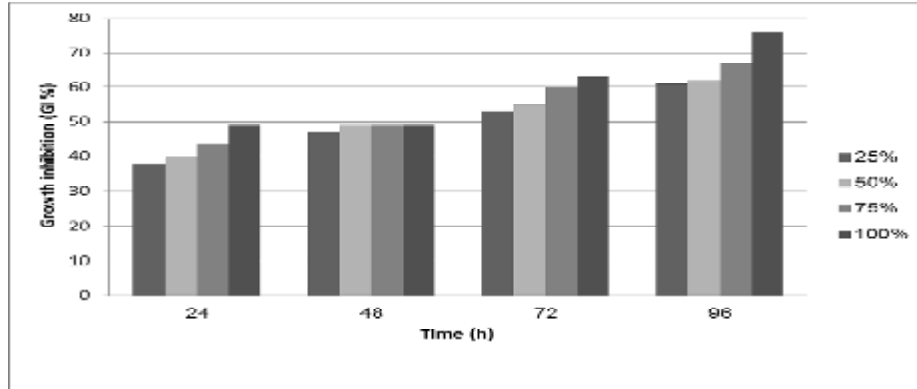
SO12		SO8		SO1		Concentrations (%)
Doubling time (G) hr	Growth rate (K)	Doubling time (G) hr	Growth rate (K)	Doubling time (G) hr	Growth rate (K)	
14.95±0.009	0.489±0.005	14.95±0.009	0.489±0.005	14.95±0.009	0.489±0.005	<b>Control</b>
18.88±0.001	0.393±0.001	19.79±0.015	0.365±0.002	18.56±0.077	0.389±0.001	<b>25</b>
21.10±0.013	0.341±0.001	20.48±0.056	0.3526±0.001	20.81±0.001	0.347±0.003	<b>50</b>
20.30±0.019	0.355±0.001	20.91±0.108	0.341±0.002	21.0±0.254	0.344±0.004	<b>75</b>
23.89±0.271	0.308±0.017	30.18±0.011	0.237±0.019	30.22±0.001	0.234±0.001	<b>100</b>

**Table (3) Growth rates (K) and doubling times (G) of *Scenedesmus dimorphus* based on chlorophyll-a concentration with respect to the wastes concentrations (%).**

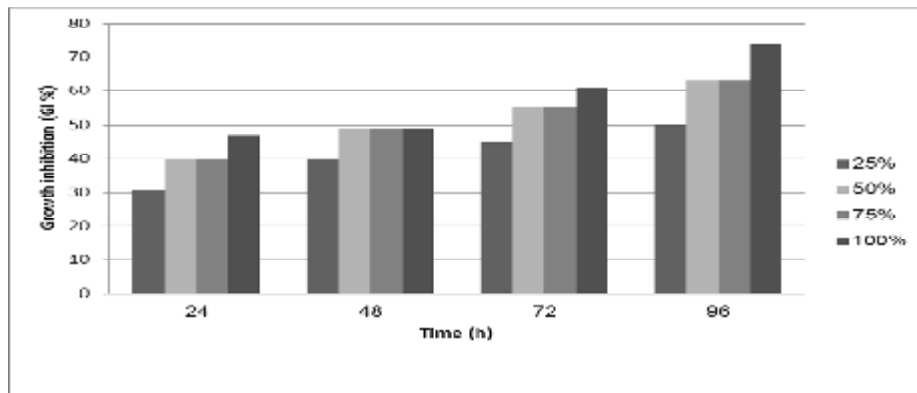
SO12		SO8		SO1		Concentrations (%)
Doubling time (G) hr	Growth rate (K)	Doubling time (G) hr	Growth rate (K)	Doubling time (G) hr	Growth rate (K)	
14.47±0.004	0.499±0.001	14.47±0.004	0.499±0.001	14.47±0.004	0.499±0.001	<b>Control</b>
16.80±0.020	0.431±0.030	17.53±0.040	0.412±0.001	17.88±0.040	0.404±0.002	<b>25</b>
16.72±0.050	0.432±0.080	17.74±0.009	0.407±0.001	19.83±0.050	0.362±0.090	<b>50</b>
17.756±0.023	0.406±0.009	17.29±0.090	0.403±0.002	21.13±0.020	0.337±0.001	<b>75</b>
17.635±0.025	0.409±0.005	17.76±0.042	0.406±0.009	23.17±0.009	0.311±0.001	<b>100</b>

In regard to growth inhibition (GI %), an inhibition effects associated with the same concentrations was observed, also a linear effect relationship was detected among growth inhibition from hand and concentrations as well as exposure period from another hand. There were increasing in the inhibition effects on the algal growth with increasing in the concentrations of the wastes as well as time of exposure. The inhibitory effects of SO1, SO8 and SO12 wastes proceeded with a much higher rate compared with the control resulting in death of the treated algae and decline of the growth rates as long as exposure period. Results demonstrated that the wastes from SO1 caused the higher inhibitory effects on the algal growth than SO8 and SO12 in both species after 96 hr. of exposure (76, 74 and 65 % respectively for *C. vulgaris*), (60, 53 and 41 % respectively for *S. dimorphus*). Moreover, *C. vulgaris* appeared more sensitive by showing the largest growth

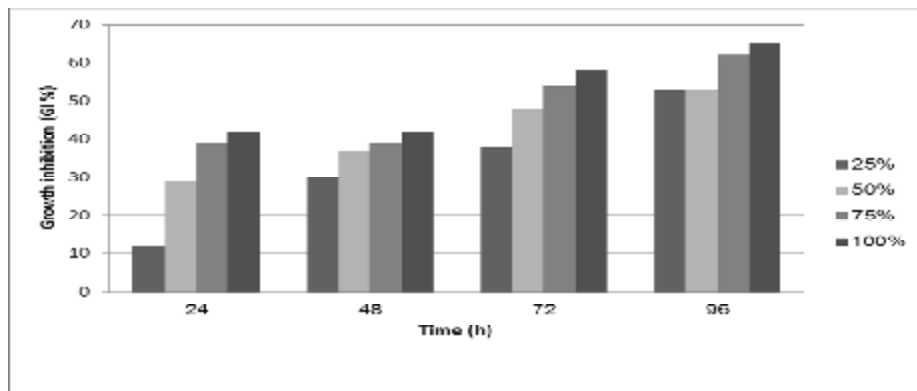
inhibition values than *S. dimorphus* for all concentrations and studied locations during time of exposure (figs. 4-9).



Figure(4) Growth inhibition (GI %) of *Chlorella vulgaris* when exposed to different concentrations of SO1 wastes during 96 hr.

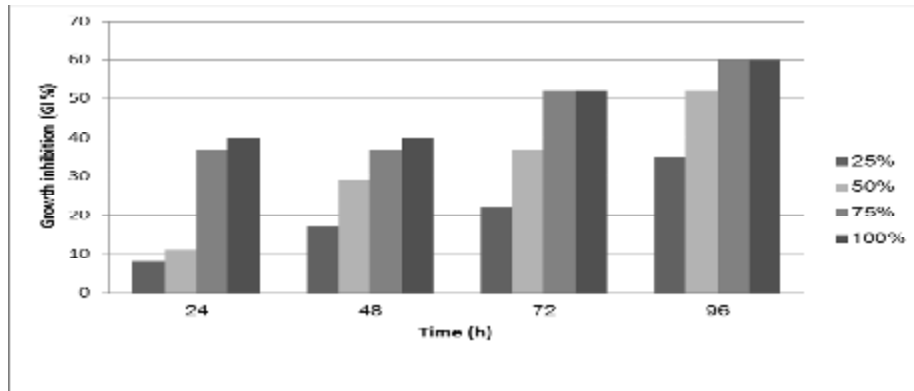


Figure(5) Growth inhibition (GI %) of *Chlorella vulgaris* when exposed to different concentrations of SO8 wastes during 96 hr.

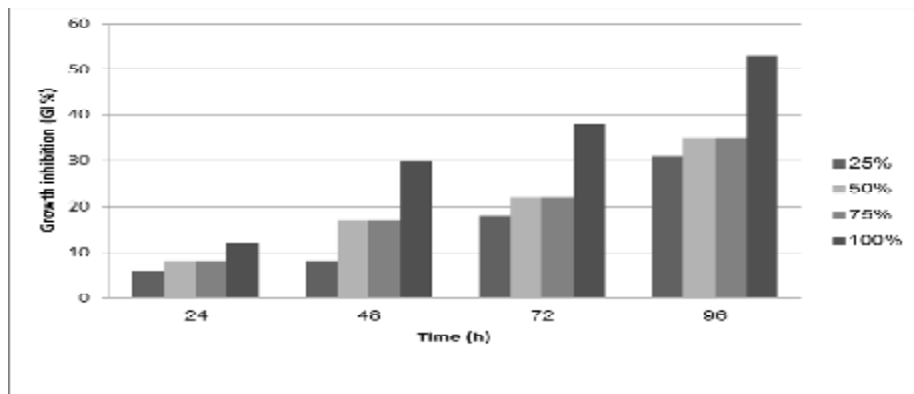


Figure(6) Growth inhibition (GI %) of *Chlorella vulgaris* when exposed to different concentrations of SO12 wastes during 96 hr.

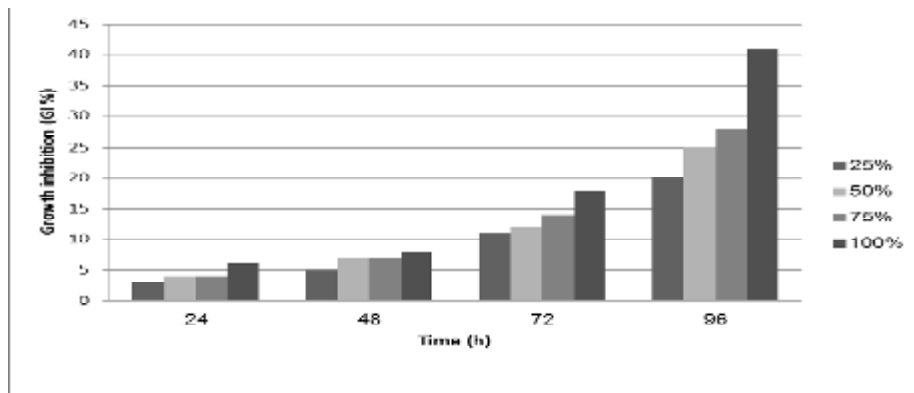




Figure(7) Growth inhibition (GI %) of *Scenedesmus dimorphus* when exposed to different concentrations of SO1 wastes during 96 hr.



Figure(8) Growth inhibition (GI %) of *Scenedesmus dimorphus* when exposed to different concentrations of SO8 wastes during 96 hr.



Figure(9) Growth inhibition (GI %) of *Scenedesmus dimorphus* when exposed to different concentrations of SO12 wastes during 96 hr.

The calculated EC<sub>50</sub> (%) of the oil wastes for both species showed gradual differences with different sampling locations after 96 hours of exposure. According to the samples taken from SO1, EC<sub>50</sub> values were less than with those calculated from either SO8 or SO12 locations (22.64, 25 and 27.03 % respectively for *C. vulgaris*), (61.65, 86.09 and 134.89 % respectively for *S. dimorphus*) (tab. 4). A relationship was defined between EC<sub>50</sub> values and growth inhibition (GI), decreasing of calculated EC<sub>50</sub> values accompanies with increasing in GI. Averages and wastes toxicity, which indicate that the wastes from SO1 was more toxic due to its ability to inhibit (more than 50 %) of algal growth at low concentration in case of *C. vulgaris*, whereas SO12 appeared less toxic to cause such inhibition specially with *S. dimorphus*. On the other hand, results showed that EC<sub>50</sub> values for *S. dimorphus* was higher than *C. vulgaris* which explain the sensitivity of the latter towards the oil wastes.

**Table (4) Median effective concentrations (EC<sub>50</sub>) of oil wastes for *Chlorella vulgaris* and *Scenedesmus dimorphus* after 96 hr.**

Species	Location	EC <sub>50</sub> (%)
<i>Chlorella vulgaris</i>	SO1	22.64 <sup>1</sup> ±0.01
	SO8	25±0.09
	SO12	27.03±0.07
<i>Scenedesmus dimorphus</i>	SO1	61.65±0.01
	SO8	86.09±0.10
	SO12	134.89 <sup>2</sup> ±0.02

<sup>1</sup> =Calculated EC<sub>50</sub> was less than tested concentrations.

<sup>2</sup> =Calculated EC<sub>50</sub> was more than tested concentrations.

Eventually, Statistical analysis showed significant variations between treatments and control. Growth rates of both species correlated negatively with the wastes concentrations ( $P < 0.05$ ) and positively between doubling time and concentrations, also positively was observed between growth inhibition and concentrations.

### Discussion

Overall the refinery wastes appeared to be toxic for both tested species during exposure period which continued to 96 hour. A reduction of the growth rates was observe when algal exposed to different concentrations of the oil wastes. The distinctive decreasing of algal biomass in the present study consisted with other several findings that attributed the effect of the oil wastes to its toxic components like oil hydrocarbons, phenols and other materials [24, 25, 26]. The results clearly demonstrate an accelerated decline in the growth of *Chlorella vulgaris* which might be due to the presence in high concentrations of a complex mixture of pollutants, as high concentrations of oils are expected to disrupt the structure and function of the plasma membrane and thus affect cell membrane permeability [27].

Similarly, it was reported that photosynthesis and cellular components consider the main target for the toxicity of the crude oil extracts in some freshwater phytoplankton's [28, 29, 30, 31]. Therefore, Crude oil up to 39  $\mu\text{l}/10\text{ ml}$  has been shown to inhibit growth of *C. vulgaris*, *Oocystis Sp.* and *Selenastrum capricornutum* by induced changes in the morphology of algae suggesting that the cell division or cell permeability is affected by the toxicants [32]. Further, [33] also documented that coccoid green algae increased at low oil concentration (10% v/v), but completely disappeared at 100% (v/v) concentration. Likewise, [34] observed decreasing in the cells number of *Scenedesmus* when exposed to water-soluble fractions of fuel oil, which supports our findings with oil wastes on growth of *S. dimorphus* in the present study. Furthermore, heavy-duty marine diesel oil (10 % concentration) has been shown to prevent the growth of a marine microalga *Isochrysis sp.*, whereas crude oil at a similar concentration caused little effect on the growth of this alga [35]. The gradual reduction in the growth in the case of *S. dimorphus* in comparison with *C. vulgaris* might be result to its ability to detoxify or metabolize some of the dissolved organic compounds. Also, it was documented that microalgae can assimilate petroleum hydrocarbons, for example, chlorococcales such as *Scenedesmus* are capable of assimilating organic solutes and may be facultative heterotrophs [36]. Moreover, [37] supported the above data, who stated the role of green alga *Scenedesmus* in the bioremediation of the crude oil, n-alkanes, poly aromatic hydrocarbons and the removal of nitrogen from wastewater. Thus, petroleum compounds in general have shown to either inhibit or stimulate algal growth, depending on the type and level of petroleum product and the algal species concerned [38, 39]. From other side, present data showed that decreasing in the algal growth rates associated with increasing in the doubling time of cell as another indicator for oil toxicity. [40] supported the above result who stated that the treatment of algal cultures of both species *S.obliquus* and *Nitzschia linearis* with crude oil led to prolongation the lag phase of the growth to 7<sup>th</sup> day with biomass less than control by 66% as well as increasing in doubling time of the cells.

With respect to the growth inhibition (GI %), the higher inhibitory effects caused by oil wastes in this study might be due to the toxic effects of the wastes fractions. Present observations indicated that the growth inhibition of *S. dimorphus* by the waste concentrations was less than *C. vulgaris*. This results are in good agreement with the findings obtained by [41] who reported that 0.1 mg/l of crude oil was responsible to inhibit the originally dominant blue-green algae which replaced then by the more resistant green alga (*S. quadricauda*). It is believed that some groups of algae can at most initiate the biodegradation of the hydrocarbons by oxidizing them to components of lower molecular weight, or by the transformation of petroleum hydrocarbons to more polar compounds of a carbon number equal to the parent compound [42]. Although we did not attempt to measure wastes concentrations in the culture medium during exposure period, algal growth inhibition tests can be very useful to detect the bioavailability fractions of the test compounds since the bioavailable fractions are expected to be responsible for toxicity.

According to the calculated EC<sub>50</sub> after 96 hr. of exposure, we can realize the gradually changing in the effective toxic concentrations of the wastes at the studied locations, which causing mortality for about 50% of the algal biomass. The results

showed that wastes samples taken from location SO1 were more toxic than from SO8 and the later were more toxic than SO12 in both species. Also, *C. vulgaris* appeared to be more sensitive than *S. dimorphus* for all studied locations at the oil refinery. In respect to present EC50 values, a similar results obtained by [43, 44] who detected the EC50, NOEC (No Observed Effect Concentration) and LOEC (Lowest Observed Effect Concentration) for *S. quadricauda* when exposed to two insecticides (Glyphosate and Paraquat) with different concentrations after 96 hours. The same was detect for green alga *C. saccharophila* after 96 hours of exposure to lead concentrations [45].

### CONCLUSIONS

The results of our study indicates many important points as follow:

1. There were differences in the tested algal sensitivities towards the toxic effects of Al-Dura refinery wastes depending on their concentrations and exposure period.
2. With regard to water pollution and thus tasks of biomonitoring, the situation in the capital of Iraq is typical for many developing countries: an oversized capital city which is hardly capable of coping with supply of basic goods and controlled removal of wastes including wastewater due to the overall economic situation in general and developing petroleum and petrochemical industries specially.
3. The method used here to estimate oil wastes burdens is simple and affordable and can also be applied elsewhere. It could thus become an integral part of biomonitoring in developing countries which now is restricted to few countries and to atmospheric inputs mainly. This is more important as a rapidly increasing population is going to enlarge burdens on natural water supply-including the necessity to tap possibly hazardous sources such as river water-continuously.
4. The present investigation established that the algae can be used effectively in assessing of the industrial effluent treatment efficiency and to identify potential environmental hazards at polluted sites and may be useful to establish guidelines for water quality.

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