

Influence of Iron Availability on Nutrient Utilization and Phytoplankton Growth in the Syrian Coast

Hazem Krawi¹, Firuz Darwich², Hussam Eddin Laika³, Samar Joulak⁴

¹: Associate Professor - Marine Chemistry Department - High Institute of Marine Research – Tishreen University- Lattakia – Syria

²: Associate Professor - Marine Biology Department - High Institute of Marine Research – Tishreen University- Lattakia – Syria

³: Associate Professor - Marine Chemistry Department - High Institute of Marine Research – Tishreen University- Lattakia – Syria

⁴: Ph.D. student - Marine Chemistry Department - High Institute of Marine Research – Tishreen University- Lattakia- Syria

Abstract

This research aims to study the effect of iron enrichment on the growth of phytoplankton and nutrients available in some areas of the Syrian coast. The experiment was conducted during the period from 3 to 24 April 2018. Samples were collected from the estuary of the Big Northern River and the marine area opposite the sports city. Samples then were transferred to laboratories of the Higher Institute of Marine Research, where semi of the water that was sampled from each site was enriched by iron and the other semi was kept as control samples. Samples were kept in laboratory conditions, and periodic samples were taken within 2-3 days with the aim of conducting measurements of nutrient concentrations and chlorophyll a. The experiment lasted 21 days.

A small effect of iron enrichment was observed in both studied sites. Whereas the growth rates of phytoplankton biomass in control and enrichment flasks were close. The absence of response of the phytoplankton for the iron stress in the estuary of the Northern River can be explained by warmness this region in iron. But in the sports city area, this can be attributed to phosphate depletion. The application of this type of experiment to different areas of the Syrian coast considers an important point to determine the role of iron in influence on the primary productivity in our region.

Keywords - Iron; Primary productivity; phytoplankton growth; Bottle incubation; Nutrient utilization; Syrian coast.

I. INTRODUCTION

Eutrophic events generally respond to a range of conditions, such as temperature and light, as well as the availability of nutrient form [1]. When the concentrations of nitrate and phosphate are high, primary production is limited by other controlling factors. More specifically, productivity is thought to be controlled by the supply of iron [2]. The biological

availability of iron to microorganisms and their fundamental mechanisms has far-reaching implications for many natural systems and diverse research areas, such as ocean biochemistry, carbon and climate cycles, harmful algal blooms, soil and plant research, bioremediation, disease and medicine. Within the framework of ocean sciences the lack of supply and limited bioavailability of Fe to phytoplankton is believed to limit primary production and reduce carbon dioxide withdrawal in the atmosphere in vast ocean regions [3]. Of all the trace metals, iron is particularly prominent in biochemical catalysis ([4], [5]). It is a major factor controlling the growth of phytoplankton and inhibits the growth of phytoplankton in up to 40% of the ocean [6].

Iron is usually transported to the marine environment through three routes: river inputs, atmospheric deposition and seabed processes, such as interpretation, re-suspension of sediment and hydrothermal respiration. ([7], [8], [9]).

We must take into account the many cellular processes in which this micronutrient plays a central role when studying the effects of iron restriction on phytoplankton or adaptation to low iron environments. The iron element is indispensable for enzymatic stimulation and electron transfer reactions according to the flexible oxidation chemistry, and is therefore closely related to the acquisition, absorption and use of the underlying resources. Iron limitation will therefore influence a wide range of metabolic pathways within phytoplankton, most prominently photosynthesis [10].

Iron metabolism intersects with a range of macronutrients, including phosphorous [11], nitrogen and silica in diatoms [12]. Previous studies on iron reduction have shown that iron should be treated not only as a direct limiting factor for phytoplankton growth, but also as an indirect control of the biogeochemical cycle of nutrients [13].

It has been recognized that the growth of phytoplankton in the three major HNLC 'High Nutrient Low Chlorophyll' regions (i.e., subarctic

Pacific Ocean, equatorial Pacific upwelling and Antarctic Ocean), is restricted by the low availability of soluble iron in the photic zone ([14], [15], [16], [17], [18]). Although these regions have common features, such as abundant major nutrients, their temperature regimes are distinctively different [19].

Its significance within aquatic systems came to the fore with John Martin's pioneering work in High Nutrient Low Chlorophyll ocean regions ([20], [21]). The resulting Iron Hypothesis sparked great interest in iron as a key limiting nutrient for marine primary production and as an important factor in the global carbon cycle [22]. Subsequent studies have shown that phototrophic microorganisms are Fe limited in vast regions of the world's oceans and some fresh water environments ([23], [24], [25], [26], [27]).

This study is the first in the Syrian coast, which shows the effect of iron enrichment on the growth of phytoplankton and its relevance to the nutrients available in our marine waters, which has not been studied until now. Iron plays an important role in controlling the primary productivity because phytoplankton growth is determined by the

availability of iron in many areas of global waters. Iron lack hinders the biological use of available nutrients and also affects the quantitative and qualitative composition of phytoplankton in those areas. Therefore, there is an urgent need to conduct such the present study in the Syrian marine waters in order to evaluate the role of enrichment of marine water by iron and its impact on primary productivity in the coastal waters of Lattakia because it lacks such studies.

II. MATERIAL AND METHODS

A. Sampling Regions:

The samples were collected from two different sites: the marine area opposite the big northern river and the marine area opposite the sports city. The big northern river flows south of the Latakia city. Its waters are flooded with many industrial installations and communities, while the sea water opposite the sports city is relatively remote from outside sources.



Figure 1. the estuary of the big north river region (the right one), The sports city region (The left one).

B. Experiment design

The experiment began on April 3, 2018 and continued for 21 days. Samples of surface water were collected for the two studied sites on 3 April and were stored directly in 30 liter polyethylene containers and transferred to laboratories of the Higher Institute for Marine Research. Semi of the amount of water sampled from each site was enriched with iron (10 nM FeCl₃ iron dissolved in 0.05 N (HCl) was added per liter of the sample. The other semi is kept as control specimens. Water divided and placed in 5 liter bottles of polycarbonate. The samples were kept in laboratory conditions (temperature and lighting) and the water was moved daily in a circular and gentle manner. Samples were taken with a 2-3 day interval for measurements of nutrient and chlorophyll a concentrations.

C. The used analyses methods

The method of Kurhoff [28] was used to determine the concentration of ammonium carbonate

in seawater, which is based on the interaction of ammonia with hypochlorite in the alkaline medium to give chloride monomer, which in turn reacts with phenol with an excessive amount of hypochlorite forming blue endophenol which absorb the light at 630 nm wavelength.

The standard method for determining the dissolved nitrite electrolytes in sea water, according to Robensen and Benschneider, is based on the interaction of nitrite with the sulfonyl amide hydrochloride forming diazonium, which is associated with [n-(1-naphthalene)-ethylene dichloride dichloride]. This interaction leads to the formation of an azo pigment that absorbs light at 540 nm wavelength [28]. The concentration of nitrate was determined by following the method of determining the nitrite itself after returning the nitrate to the nitrite using a column of copper cadmium [28].

The Riley and Murphy method was used to determine the concentration of phosphorus inorganic

oxides based on the interaction of ammonium molybdates with phosphates in the presence of triple antimony as an intermediary to obtain phosphomolybdate acid. The resulting complex is due by ascorbic acid to give the blue phosphomolybdenum which absorbs light at length Wave 885 nm [28].

To determine the concentration of dissolved silicate electrolytes in the water, the Corulf method was used to form silicomolybdic acid when acidic samples are treated with molybdate solution, which is due to the blue silicolipid complex by ascorbic acid and oxalic acid. This complex absorbs light at 810 nm wavelength [28].

To determine the concentration of chlorophyll dyes in water, the Jeffrey and Humphrey method [29] was used. The samples were filtered on cellulose filters and then crushed by a vertical mixer. Chlorophyll a was extracted from samples of in the dark by acetone 90%. Then the absorption of samples was measured by a spectrophotometer and the chlorophyll a concentration was determined using arithmetic equations [29].

The dissolved iron concentrations were determined by the Varian 220 atomic absorption device, where the water samples (500 ml) were filtered using filter filters (0.45 μm), then liquid-liquid extraction was performed using APDC and chloroform [30]. Water salinity and temperature were determined using a WTW-Multi 340 i field meter. All required absorption measurements were performed using a ZUZI Spectrophotometer (Models 4211/20).

III. RESULTS AND DISCUSSION

The primary concentrations of nutrients in the estuary of the big Northern River (K) were 25.101,

3.063, 108.931 and 34.477 $\mu\text{mol} / \text{L}$ for nitrates, phosphates, silicates and ammonia, respectively (Table 1). While the primary concentration of nutrients in the sports city area (M) was 10.857, 0.330, 5.261 and 3.733 $\mu\text{mol} / \text{L}$ for nitrate, phosphate, silicate and ammonia, respectively (Table 1). These values converged with the values recorded in many previous local studies conducted in these two regions ([31], [32], [33]).

The station K was characterized by the primary nutrient concentrations were higher than the station M because estuaries were directly subject to the impact of agricultural and human activities and land-based sources of pollution, leading to higher concentrations of nitrates and phosphates.

While the station M is relatively far from pollution sources as many studies recorded ([34], [35], [33]).

The concentrations of silicates in station K were also higher due to the richness of the rivers with it [36] which resulting from the current erosion along the runway of the river and its subsequent transition to marine waters. The primary concentrations of nutrients were directly reflected on the primary concentrations of chlorophyll a in the two studied stations because the availability of nutrients in the medium contributes to controlling the growth of phytoplankton and the changes in their biomass and their specific composition ([37], [38]).

Station K, which was characterized by higher concentrations, recorded the highest value of chlorophyll a of (6.283 $\text{mg} \cdot \text{m}^{-3}$), while the M station recorded the value of (1.611 $\text{mg} \cdot \text{m}^{-3}$), consistent with many studies conducted at the Syrian coast which pointed to increasing a phytoplankton abundance in stations subject to the influence of human activities ([31], [32], [33]).

Table 1. Primary concentrations of chlorophyll a and nutrients at the big North river station (K) and Sports city station (M).

Station	Chl a ($\text{mg} \cdot \text{m}^{-3}$)	NO_3^- ($\mu\text{mol/L}$)	PO_4^{3-} ($\mu\text{mol/L}$)	SiO_4^{4-} ($\mu\text{mol/L}$)	NH_4^+ ($\mu\text{mol/L}$)
K	6.283	25.101	3.063	108.931	34.477
M	1.611	10.857	0.330	5.261	3.733

Changes in Chl a and nutrients concentrations were followed over time in samples enriched with Fe and in controls.

The experiment showed low growth ability of phytoplankton in the first days of the experiment in both control and enrichment flasks and in both studied sites. Chlorophyll a concentrations in site K increased during the first nine days of the experiment by only 1% and 5% in both control and enrichment flasks, respectively. While the M samples showed no clear growth of phytoplankton during the first nine days. The value of chlorophyll throughout this period was close to the primitive value. Similar studies have

shown that the effect of iron enrichment begins in the early days of the experiment if phytoplankton in the studied area subject to the impact of iron stress ([39], [40], [41], [19], [13]).

Phytoplankton began to grow clearly in all circles starting on the 12th day of incubation, where phytoplankton showed clear growth ability in station K in the enrichment samples Fe (39.739 $\text{mg} = \Delta\text{Chl a}$) and control samples (37.946 $\text{mg} = \Delta\text{Chl a}$). The growth continued during the incubation period (21

days) recording the highest value of chlorophyll a concentrations at the end of the experiment which attained 46.021, 44.229 mg.m⁻³ = Max Chl a in both C and Fe respectively. While phytoplankton growth was lower in station M (2.411, 2.067 mg = ΔChl a in both C and Fe respectively), that due to the fact that primary concentrations of nutrients and chlorophyll a were low (Table 1). The concentrations of chlorophyll a at station M had the highest value on the 15th day of incubation, with 4.311, 4.115 mg.m⁻³ = Max Chl a in both C and Fe respectively. Then chlorophyll a concentrations began to decline after the fifteenth day of incubation. That due to the depletion of all phosphates in the medium recorded at the end of the experiment the value of 3.678 mg.m⁻³ in flask C and 4.022 mg.m⁻³ in the Fe flask. Where the study of re. [33] showed the persistence of phytoplankton to grow if nutrient availability and decline after nutrient depletion from the medium.

The increase in Chl a concentration was correlated with major nutrients consumption.

Nutrient consumption of phytoplankton at station K was significant during incubation period and in both sites (Table 2). The availability and consumption of available nutrients continued until the end of the experiment and was accompanied by a decrease in nutrient concentration with the increase in

phytoplankton biomass due to photosynthesis activity of phytoplankton [42], because nitrogen and phosphorus are essential elements for the growth of phytoplankton [43]. While the availability of silicates is necessary for the growth of the diatoms as it enters the building of its siliceous shield ([36], [44]).

The consumption of phytoplankton for nutrients at site M was evident in control and enrichment treatments to the fifteenth day of the experiment. Where phosphate was completely depleted from the two mediums. The percentage of the consumed nitrate during this period was 96.97% and the silicate consumption was 95.91%. The highest values of chlorophyll were 4.311, 4.115 mg.m⁻³ in both C and Fe respectively. It was noted that phytoplankton continued to consume the nitrates and silicates available in the medium even after the phosphates were depleted from the medium. This is explained by the fact that phytoplankton cells took their nitrate concentrations in the middle and deposited them in their bodies [45]. Especially in different types of central and rational diatoms [46]. Other studies have shown that diatoms have the ability to consume silicates from the medium to raise their cell contents from silicates and support their silica shield without any growth ([47], [48]).

Table 2. Total change in concentrations of chlorophyll a and nutrients in control vials (C) and enrichment (Fe) in the Northern River (K) and the Sports City (M)

The station	The treatment	ΔChl a mg	Δ NO ₃ ⁻ μmol	Δ PO ₄ ⁻³ μmol	Δ SiO ₄ ⁻⁴ μmol	Δ NH ₄ ⁺ μmol
K	C	37.947	9.890	1.319	37.289	21.814
	Fe	39.739	8.710	1.510	46.619	13.981
M	C	2.067	7.041	0.330	4.650	1.976
	Fe	2.411	7.155	0.330	4.374	1.391

A little effect of iron enrichment on growth of phytoplankton was observed in both stations, whereas the growth of phytoplankton in control flasks (C) and enrichment (Fe) was close (Table 2). This agrees with a number of studies in multiple regions showing a similar growth of phytoplankton in both mediums ([39], [40], [27]). On other hand, another laboratory and field studies in other areas where phytoplankton suffer from iron deficiency stress showed that biomass increased significantly when iron water was enriched ([39], [40], [41], [19], [13], [6]).

The quantities of nitrates and phosphates consumed in our experiments were close (Table 2), in addition to the large similarity in the changes in the concentrations of these electrodes during the experiment in the control and enrichment treatment

and in both sites as shown in figures (1, 2). While the above studies showed significant consumption of nitrates and phosphates in the media to be enriched by iron compared with the control media. As for silicate, Table (3) shows that the two studied regions showed no significant differences in nutrient ratios between the control and enrichment wells, thus enhancing the belief that these areas are not subject to iron stress. Where previous studies have shown areas under the stress of iron deficiency that the consumption increases when enriched iron, but less than nitrates and phosphates.

A study of [40] and [41] showed that the consumption of (SiO₄⁻⁴; NO₃⁻) in the control vial was double or more than that in the vials of enrichment because the diatoms under stress of iron deficiency

also increased its consumption and use of silicates in enhancing its silicic shield, making silicate a specific catalyst for growth (colimiting) in areas under iron stress [49]. Where re. [6] mentioned that the addition of iron can increase the rate of consumption of salicylic acid and the production of bio silica for algae, and that increased iron stress can slow growth rate, and increases the silicification of certain types of algae.

The difference in the consumption of silicates between the control and enrichment treatment in the

Northern river region can be attributed to stimulating iron for the growth of silicated algae in the enrichment treatment (Table 2). Where the study of [50] showed that increased iron air supply in the central equatorial Pacific Ocean led to a change in the major primary producers of coccolithophores (alkenones) to diatoms (epibrassicasterol) and dinoflagellates (dinosterol).

Table (3): Nutrient consumption ratios ($\text{NO}_3^-:\text{SiO}_4^{4-}:\text{PO}_4^{3-}:\text{NO}_3^-:\text{SiO}_4^{4-}$) in control vials (C) and enrichment (K) in the two sites [north river (K) and sports city (M)]

Site	Treatment	$\text{SiO}_4^{4-}:\text{NO}_3^-$	$\text{SiO}_4^{4-}:\text{PO}_4^{3-}$	$\text{NO}_3^-:\text{PO}_4^{3-}$
K	C	3.770	28.271	7.498
	Fe	5.352	30.874	5.768
M	C	0.660	14.091	21.336
	Fe	0.611	13.255	21.682

Based on our findings, we note that the limited effect of iron supply, which may be due to the richness of the area of the estuary of the Big Northern River by the iron ($2.1 \mu\text{g} / \text{L}$) because it is directly subject to the impact of human activities and land-based sources of pollution which bring with it big amount of iron make the area not susceptible to iron stress and increased concentration of iron in the middle did not affect the growth of phytoplankton. A similar study in northern Monterey Bay on the Pacific ocean showed that chlorophyll production and nutrient consumption in this area were not affected by the addition of iron due to the abundance of iron

amounts coming with river water to this region [40]. In the samples of the sports city region, we also note that iron enrichment does not affect the growth of phytoplankton, although iron concentrations are lower than the first region ($0.0364 \mu\text{g} / \text{L}$). This can be attributed to the depletion of phosphates from the medium similar to re. [39] in the northwest Indian Ocean where the lack of nitrogen in this region has led to growth of phytoplankton in similar ways in the control and enrichment communities, While the growth rate was high in the enrichment treatment compared to the control one when accompanied iron enrichment with the enrichment of nitrogen.

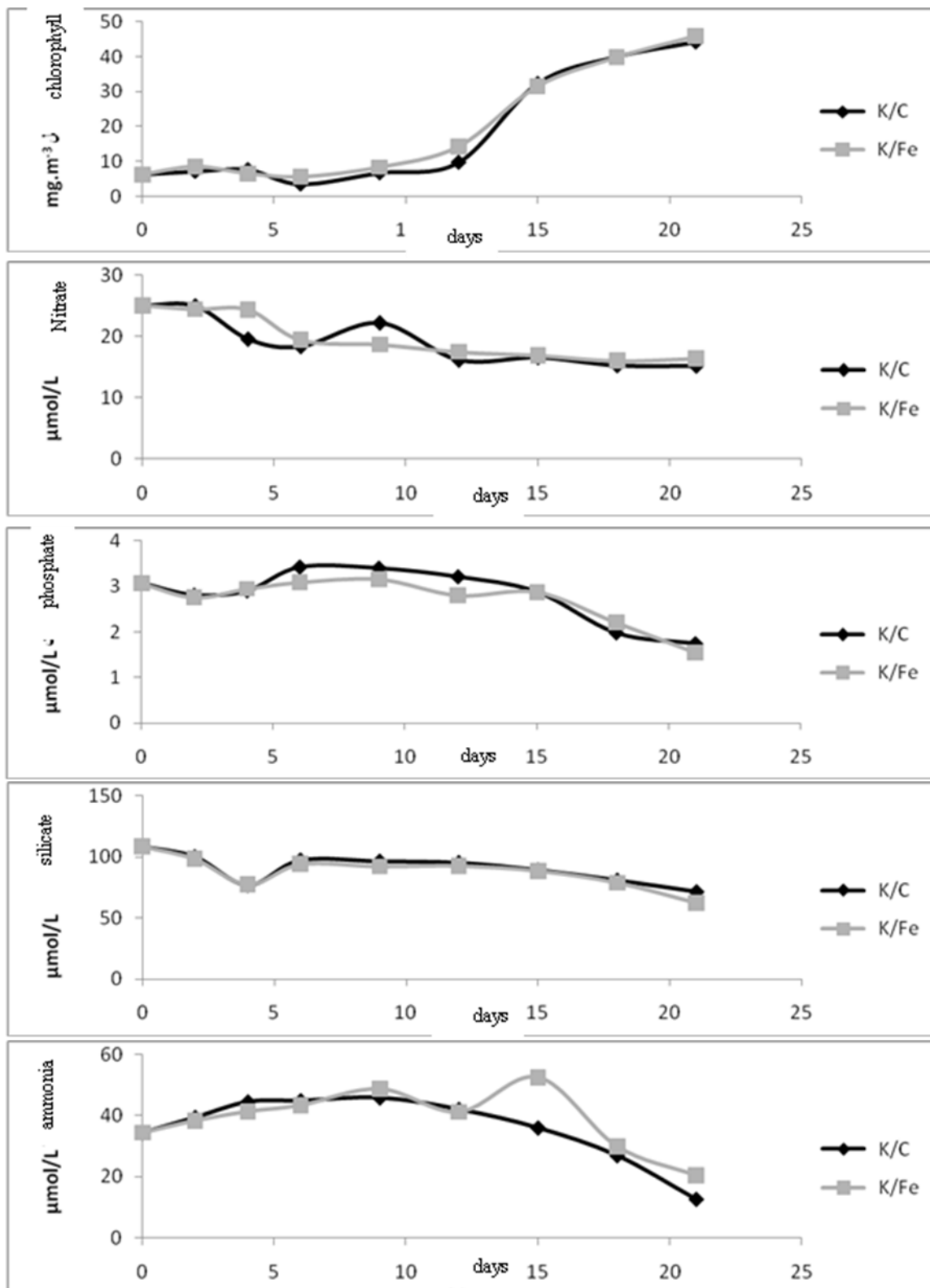


Figure (1): The chronological development of concentrations of chlorophyll a, nitrates, phosphates, silicic acid and ammonia in control vials (C) and enrichment (Fe) for the big northern river station (K)

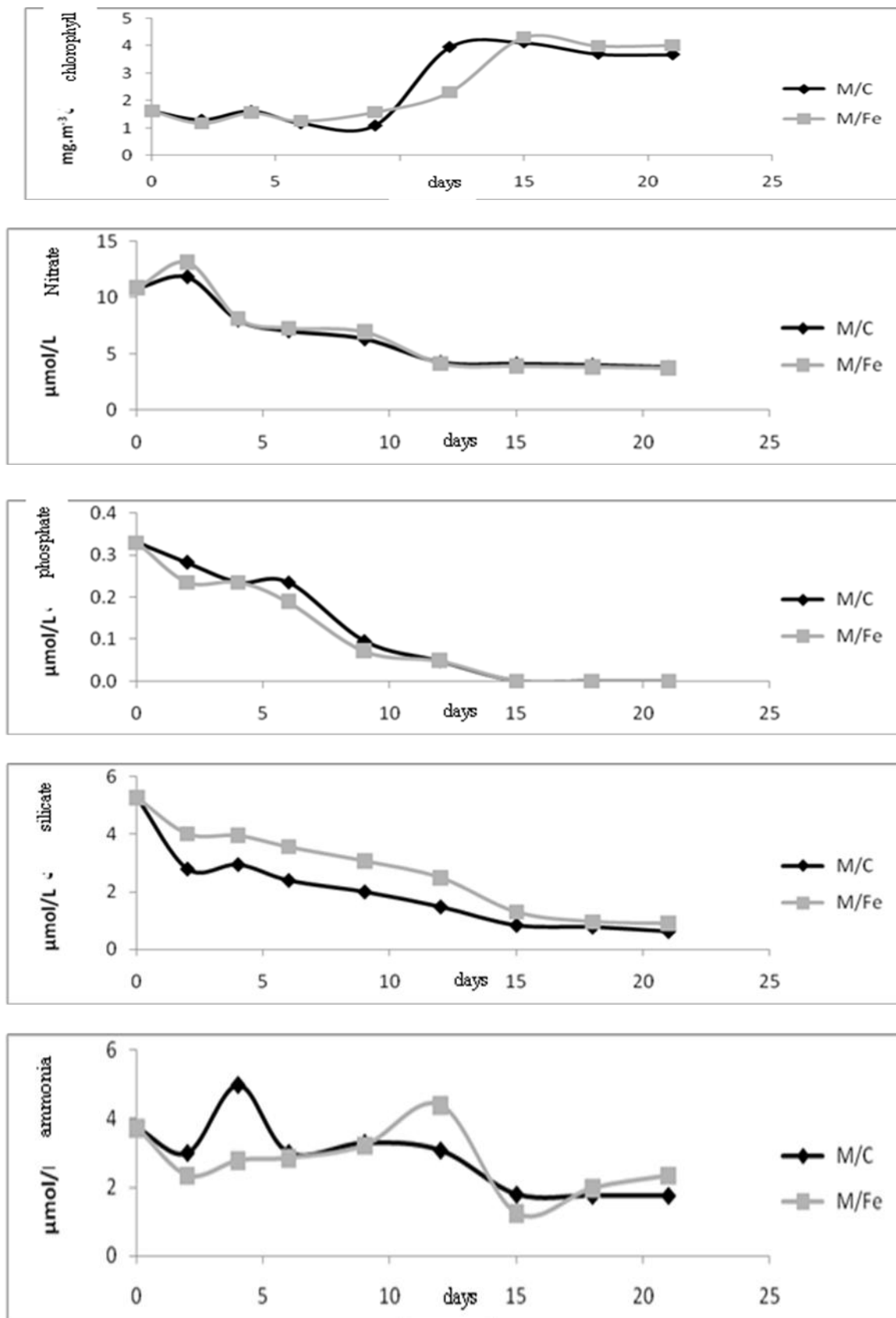


Figure (2): The chronological development of concentrations of chlorophyll a, nitrates, phosphates, silicans and ammonia in control vials (C) and enrichment (Fe) for the sport city (M)

VI. CONCLUSIONS

1. The primary concentrations of chlorophyll a and nutrient in the Big Northern River estuary (K) recorded values higher than those recorded in the sports city area (M).
2. Phytoplankton showed clear growth ability in station K in both the control and enrichment media. Growth continued throughout the incubation period and nutrients remained in the medium until the end of the experiment. Growth of phytoplankton was lower in station M and stopped on the 15th day of incubation in both of which are due to phosphate depletion.
3. A small effect of iron enrichment was observed in both studied sites, where the net growth rates of phytoplankton biomass in control and enrichment flasks were close.
4. Phytoplankton cannot be affected by iron stress in the area of the estuary of the Big Northern River due to the richness of this region with iron, but in the sports city area this can be attributed to the depletion of phosphates from the medium.
5. The application of this type of experiment to different areas of the Syrian coast is important to find out the role of iron in influencing the primary productivity in our region.
6. It is useful to apply these experiments under controlled laboratory conditions (lighting, heat, nutrients) where international studies have shown the effect of these factors on the role of iron in the growth of phytoplankton.

REFERENCES

- [1] VILMIN, L., MOGOLLÓN, J. M., BEUSEN, A.H.W., BOUWMAN, A. F. (2018). Forms and subannual variability of nitrogen and phosphorus loading to global river networks over the 20th century. *Global and Planetary Change*.
- [2] HOPPE, C.J.M.A., KLAAS, C., OSSEBAAR, S., days M.A., CHEAH, W., LAGLERA, L.M., SANTOS-ECHEANDIA, J., ROST, B., WOLF-GLADROWA, D.A., BRACHER, A., HOPPEMA, M., STRASS, V., TRIMBORN, S. (2017). Controls of primary production in two phytoplankton blooms in the Antarctic Circumpolar Current. *Deep-Sea Research II* 138 63–73.
- [3] SHAKED, Y., LIS, H. (2012). Disassembling iron availability to phytoplankton. *frontiers in microbiology*. Vol. 3.
- [4] MOREL, F.M.M., PRICE, N. M. (2003). The biogeochemical cycles of trace metals in the oceans. *Science* 300, 944–947
- [5] SHCOLNICK, S.; KEREN, N. (2006). Metal homeostasis in cyanobacteria and chloroplasts. Balancing benefits and risks to the photosynthetic apparatus. *Plant Physiol.* 141, 805–810.
- [6] ZHAO, P.; GU, W.; HUANG, A.; WU, S.; LIU, C.; HUAN, L.; GAO, S.; XIE, X.; WANG, G. (2018). Effect of iron on the growth of *Phaeodactylum tricornutum* via photosynthesis. *J. Phycol.* 54, 34–43.
- [7] USSHER, S.J., E.P. ACHTERBERG and P.J. WORSFOLD. (2004). Marine biogeochemistry of iron. *Environ. Chem.* 1: 67–80.
- [8] SEVERMANN, S., MCMANUS, J., BERELSON, W.M., HAMMOND, D.E., (2010). The continental shelf benthic iron flux and its isotope composition. *Geochimica et Cosmochimica Acta* 74, 3984–4004.
- [9] TAGLIABUE, A., BOPP, L., DUTAY, J.-C., BOWIE, A.R., CHEVER, F., JEAN-BAPTISTE, P., BUCCIARELLI, E., LANNUZEL, D., REMENYI, T., SARTHOU, G., AUMONT, O., GEHLEN, M., JEANDEL, C., (2010). Hydrothermal contribution to the oceanic dissolved iron inventory. *Nature Geoscience* 3, 252–256. Takeda, S., 1998. Influence of iron availability.
- [10] SCHOFFMAN H, LIS H, SHAKED Y and KEREN N (2016) Iron–Nutrient Interactions within Phytoplankton. *Front. Plant Sci.* 7:1223.
- [11] MILLS, M.M., RIDAME, C., DAVEY, M., LAROCHE, J., AND GEIDER, R.J. (2004). Iron and phosphorus co-limit nitrogen fixation in the eastern tropical North Atlantic. *Nature* 429, 232–292. doi:10.1038/nature02550
- [12] ARMBRUST, E. V. (2009). The life of diatoms in the world's oceans. *Nature* 459, 185–192.
- [13] SONG, S.; XINMING, P.; YONGSHAN, Z. (2009). In vitro iron enrichment experiments in the Prydz Bay, the Southern Ocean: A test of the iron hypothesis. *Sun S et al. Sci China Ser D-Earth Sci.* vol. 52, no. 9, 1426–1435.
- [14] COALE, K. H., JOHNSON, K. S., FITZWATER, S. E., GORDON, R. M., TANNER, S., CHAVEZ, F. P., et al. (1996). A massive phytoplankton bloom induced by an ecosystem-scale iron fertilization experiment in the equatorial Pacific Ocean. *Nature*, 383, 495–501.
- [15] MARTIN, J. H., COALE, K. H., JOHNSON, K. S., FITZWATER, S. E., GORDON, R. M., TANNER, S. J., et al. (1994). Testing the iron hypothesis in ecosystems of the equatorial Pacific Ocean. *Nature*, 371, 123–129.
- [16] TSUDA, A., TAKEDA, S., SAITO, H., NISHIOKA, J., NOJIRI, Y., KUDO, I., et al. (2003). A mesoscale iron enrichment in the western subarctic Pacific induces a large centric diatom bloom. *Science*, 300, 958–961.
- [17] SUGIE, K., NISHIOKA, J., KUMA, K. et al. (2013) Availability of particulate Fe to phytoplankton in the Sea of Okhotsk. *Mar. Chem.*, 152, 20–31.
- [18] JOHNSON, Z. I., SHYAM, R., RITCHIE, A. E. et al. (2010) The effect of iron-and light-limitation on phytoplankton communities of deep chlorophyll maxima of the western Pacific Ocean. *J. Mar. Res.*, 68, 283–308.
- [19] NOIRI, Y.; KUDO, I.; KIYOSAWA, H.; NISHIOKA, J.; TSUDA, A. (2005). Influence of iron and temperature on growth, nutrient utilization ratios and phytoplankton species composition in the western subarctic Pacific Ocean during the SEEDS experiment. *Progress in Oceanography* 64 149–166.
- [20] MARTIN JH, FITZWATER S. (1988). Iron deficiency limits phytoplankton growth in the north-east Pacific subarctic. *Nature* 331: 947–975.
- [21] MARTIN JH. (1990). Glacial-interglacial CO₂ change: the iron hypothesis. *Paleoceanography* 5: 1–13.
- [22] MARTIN JH, GORDON RM, FITZWATER SE. (1991). The case for iron. *Limnol Oceanogr* 36: 1793–1802.
- [23] MOORE JK, DONEY SC, GLOVER DM, FUNG IY. (2001). Iron cycling and nutrient-limitation patterns in surface waters of the World Ocean. *Deep Sea Res Pt II* 49: 463–507.
- [24] MCKAY RML, BULLERJAHN GS, PORTA D, BROWN ET, SHERRELL RM, SMUTKA TM et al. (2004). Consideration of the bioavailability of iron in the North American Great Lakes: development of novel approaches toward understanding iron biogeochemistry. *Aquat Ecosyst Health* 7: 475–490.
- [25] BOYD PW, JICKELLS T, LAW CS, BLAIN S, BOYLE EA, BUESSELER KO et al. (2007). Mesoscale iron enrichment experiments 1993–2005: synthesis and future directions. *Science* 315: 612–617.
- [26] NORTH RL, GUILDFORD SJ, SMITH REH, HAVENS SM, TWISS MR. (2007). Evidence for phosphorus, nitrogen, and iron colimitation of phytoplankton communities in Lake Erie. *Limnol Oceanogr* 52: 315–328.
- [27] LIS, H.; SHAKED, Y.; KRANZLER, C.; KEREN, N.; MOREL, F. M. (2015). Iron bioavailability to phytoplankton: an empirical approach. *The ISME Journal* 9, 1003–1013.

- [28] GRASSHOFF, K.; KREMLING, K., EHRHARDT, M. (1999), *Methods of Seawater Analysis*. 3rd. ed., Wiley-VCH, New York, 634.
- [29] JEFFREY, S. W.; HUMPHREY, G. F. (1975), New spectrophotometric equations for determining chlorophylls a, b, c1, and c2 in higher plants, algae and natural phytoplankton. *Biochem. Physiol. Pflanz*, Vol. 167, 191-194.
- [30] U.S. Environmental Protection Agency. (1983). General procedure for analysis by atomic absorption. P: 58-70.
- [31] OMRAN, M. Y. (1995). Investigation of nitrogen inorganic ions in Syrian coastal water. Master thesis, Tishreen University, 92pp. In Arabic.
- [32] JOLAK, S. K. (2013). Nutrient distribution in variable coastal waters and the effect of hydrochemical properties. Master thesis. Tishreen University, 103pp. In Arabic.
- [33] SULAIMAN, N. G. (2013). The role of nutrients (NO₃, PO₄, SiO₄) on phytoplankton growth in Lattakia coastal water. Master thesis, Tishreen University, 87pp. In Arabic.
- [34] SALMAN, M. DARWICH, F. KRAWI, H. (2017). Investigation of spatial and temporal variations of nutrient in the coastal water of Banias city. *Tishreen University Journal for Research and Scientific Studies Biological Sciences Series*, 39 (5). In Arabic.
- [35] JOULAK, S. LAIKA, H. E. KRAWI, H. (2003). Influence of iron availability on nutrient utilization and phytoplankton growth in the Syrian coast. *Tishreen University Journal for Research and Scientific Studies Biological Sciences Series*, 35 (2). 171- 189. In Arabic.
- [36] LEWIN, J. C. (1961). The dissolution of silica from diatom walls. *Geochimica et Cosmochimica Acta.*, Vol. 21, 1961, 182 – 198.
- [37] LAGUS, A.; SUOMELA, J.; WEITHOFF, G.; HEIKKILA, K.; HELMINEN, H.; SIPURA, J. (2004) Species-specific differences in phytoplankton responses to N and P enrichment and N:P ratio in the Archipelago Sea, northern Baltic Sea. *J. Plankton Res.*, Vol. 26 779 – 798.
- [38] SULLIVAN, C. W. (1976). Diatom mineralization of silicic acid I. Si(OH)₄ transport characteristics in *Navicula pelliculosa*. *Journal of Phycology*, Vol. 12, 390 – 396.
- [39] TAKEDA, S.; KAMATANI, A.; KAWANOBE, K. (1995). Effects of nitrogen and iron enrichments on phytoplankton communities in the northwestern Indian Ocean. *Marine Chemistry* 50 229-241.
- [40] HUTCHINS, D.; BRULAND, K. (1998). Iron-limited diatom growth and Si:N uptake ratios in a coastal upwelling regime. *NATURE*, VOL 393.
- [41] TAKEDA, S. (1998). Influence of iron availability on nutrient consumption ratio of diatoms in oceanic waters. *NATURE*. VOL 393.
- [42] RAGUENEAU, O.; LANCELOT, C.; EGOROV, V.; VERVLIMMEREN, J.; COCIASU, A.; DELIAT, G.; KRASTEVA, A.; DAOUD, N.; ROUSSEAU, V.; APOPOVITCHEV, V.; BRION, N.; POPA, L.; CAUWET, G. (2002). Biogeochemical Transformations of Inorganic Nutrients in the Mixing Zone between the Danube River and the Northwestern Black Sea. *Estuarine, Coastal and Shelf Science*, Vol. 54, 321- 336.
- [43] GOLTERMAN, H. L.; De OUDE, N. T. (1991). Eutrophication of lakes, rivers and coastal seas. The handbook of environmental chemistry, Vol. 5, (Part A), 79 – 124.
- [44] TRÉGUER, P. J., AND DE LA ROCHA, C. L. (2013). The world ocean silica cycle. *Ann. Rev. Mar. Sci.* 5, 477–501.
- [45] SOMMER, U. (1994). Are marine diatoms favoured by high Si:N ratios?. *Mar. Ecol. Prog. Ser.*, Vol. 115, 309 – 315.
- [46] DARWICH, F. (2006). Die untersuchung des wachstums der kieselalgen in abhängigkeit von verschiedenen nährstoffkonzentrationen und verhältnissen. Dr. rer. nat., Rostock, Germany, 101p.
- [47] BRODHERR, B. H. (2006). Nutrient dependent growth dynamics of diatom spring populations in the southern Baltic Sea. Rostock, Germany, 132p.
- [48] PAN'CI' C, M.; KIØRBOE, T. (2018). Phytoplankton defence mechanisms: traits and trade-offs. *Biol. Rev.*
- [49] Brzezinski, M. A., Baines, S. B., Balch, W. M. et al. (2011) Co-limitation of diatoms by iron and silicic acid in the equatorial Pacific. *Deep-Sea Res. Pt II*, 58, 493–511.
- [50] Badejo, A.; Seo, I.; Kim, W.; Hyeong, K.; Ju, S. Effect of eolian Fe-supply change on the phytoplankton productivity and community in central equatorial Pacific Ocean during the Pleistocene: A lipid biomarker approach. *Organic Geochemistry* 112 (2017) 170–17