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Research Article

Fatty Acids Composition and Sources of Organic Matter in Surface Sediments of Four River Nile Sub-Branches, Egypt

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Abstract

Sedimentary Fatty Acids (FAs) were investigated in surficial sediment of four sub-branches (canals) of River Nile, Egypt, to distinguish their origin of organic matter. Seventeen stations were investigated for four seasons from spring, 2014 till winter, 2015. Analysis of variance (ANOVA) with *post hoc* suggested significant seasonal differences in total fatty acids, fatty acid classes and main fatty acids with obvious increase in spring. The dominant Saturated Fatty Acids (SFA) in sediment samples were C16:0 and C18:0. Dominant monounsaturated fatty acids (MUFA) included C16:1 ω 7 and C17:1 ω 7 whereas, C18:1 ω 9 was less important. Fatty acids indicators including microalgal classes, bacteria, sewage and terrestrial plants were used to distinguish their contribution to surficial sediment organic matter. The results indicated that microalgae were major contributor to sediment organic matter, alternatively, land plant organic matter was nearly absent from the sedimentary organic matter in the four canals. Between microalgae, diatoms were prevalence over cyanobacteria and green algae. The seasonal changes in fatty acids composition were driven by the changes in species composition of microalgal communities rather than the direct effect of environmental characteristics.

Key words: Fatty acids, organic matter, fatty acids indicators, microalgal classes

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Streams and rivers supply important ecosystem services, such as drinking water supply, fish production, opportunity for recreational activities and the collection, transport and processing of pollutants and contaminants originated from the surrounding landscape (Millennium Ecosystem Assessment, 2005). The river Nile is the life artery of Egypt. Throughout the known Egyptian history; the Nile had dominating influence on the economy, culture, public health, social life and political aspects (Abdel-Hamid *et al.*, 1992). At Delta Barrage, the river Nile bifurcates into two main branches namely Damietta and Rosetta and four other subbranches (which called rayahs in native tongue). The four sub-branches are El Rayah El Tawfiky, El Rayah El Menoufy, El Rayah El Behery and El Rayah El Nasery. These sub branches or canals extend for hundreds of kilometers from the Delta Barrage to the most northern of Nile Delta. Alongside the two main Nile branches, the four rayahs are the main water source for irrigation, drinking, industries and navigation of all governorates of Nile Delta, specially the most western and eastern of Nile Delta, in addition to Alexandria governorate (NIOF., 2015).

Sedimentary Organic Matter (OM) contains a diverse range of lipid compounds derived from organisms live within aquatic ecosystems and their catchments, with differences in lipid composition directly reflect the different biota (Pearson *et al.*, 2007). The application of lipid biomarkers to provide more specific and sensitive information rather than bulk elemental and isotopic techniques has become widespread (Bianchi, 2007). Traditionally, fatty acids occur almost ubiquitously in sediments and their distributions have been widely used to identify OM sources. Fatty acids have been used extensively for assessing the sources and fates of organic matter in marine, coastal and estuarine ecosystems (Meziane and Tsuchiya, 2000; Countway *et al.*, 2007) and more recently freshwater rivers and lakes (Mortillaro *et al.*, 2011; Boechat *et al.*, 2014; Fang *et al.*, 2014; Honeyfield and Maloney, 2015).

Environmental studies involving FAs in aquatic ecosystems commonly focus on their use as markers for bacterial or algal presence and origin of organic matter (terrestrial, aquatic), as well as indicators of prey nutritional quality for consumers (Arts *et al.*, 2009). Fatty acids profiles provide a measure of the entire periphyton community, including heterotrophic bacteria, filamentous cyanobacteria, flagellates and nematodes (Da Silva *et al.*, 2008) and therefore, offer a more complete picture of fatty acids found in the basal resources in the system. The FAs have rarely been tested as indicators of human impacts on ecosystems, but could be

used as markers for urbanization impacts, such as sewage discharge (Jarde *et al.*, 2005) or as markers for changes in key ecosystem processes, such as the response of primary production to enhanced nutrient availability in agricultural streams (Boechat *et al.*, 2014). Analyses of individual fatty acids have been shown to be powerful for deciphering their origins in recent sediments. The difference in characteristic chain lengths and composition of aquatic and terrestrial plants have made the distribution of fatty acids an effective biomarker tool for assessing biogenic sources of organic matter in terrestrial and lacustrine ecosystems (Fang *et al.*, 2014). Relationships between community species composition and fatty acids have been documented (Sushchik *et al.*, 2010; Honeyfield and Maloney, 2015). Therefore, the efficacy of using fatty acids as a stream bioassessment tool and to assess stream water quality must be considered (Honeyfield and Maloney, 2015). This study planned to evaluate, with respect to the pool of sedimentary OM, the relative contribution of the direct import of anthropogenic, riverine and terrestrial derived OM to the sediment of the four canals.

MATERIALS AND METHODS

Study area: At the north of Cairo, at Delta Barrage, River Nile bifurcates into two main branches namely Damietta and Rosetta and four other subbranches which are El rayah El Tawfiky, El rayah El Menoufy, El rayah El Behery and El rayah El Nasery. The four rayahs or canals have been used for irrigation, drinking and navigation in one of the most populated area in Egypt. Recently, many power plants were constructed, specifically at El Behery and El Nasery canals. When the water of the four canals run short, they replenish from Rosetta and Damietta branches, except El Nasery canal which replenishes from El Behery canal at Kom Hamada city. El Behery canal replenishes from Rosetta branch at El Mahmoudia city, whereas, El Tawfiky and El Menoufy canals replenish from Damietta branch at El Mansoura and Zifta cities, respectively. The four canals vary in length from 170 km (El Menoufy) to 230 km (El Behery). They are as deep as the Nile main branches but less in width. Four stations were selected at each canal, two before and two after the replenishing point, in addition to one station before the bifurcation of River Nile at the Delta Barrage (Fig. 1).

Sampling: Surface sediment was collected using Ekman grab, where sampling was repeated until undistributed samples were collected. Samples of the upper 0.5 cm layer were putted

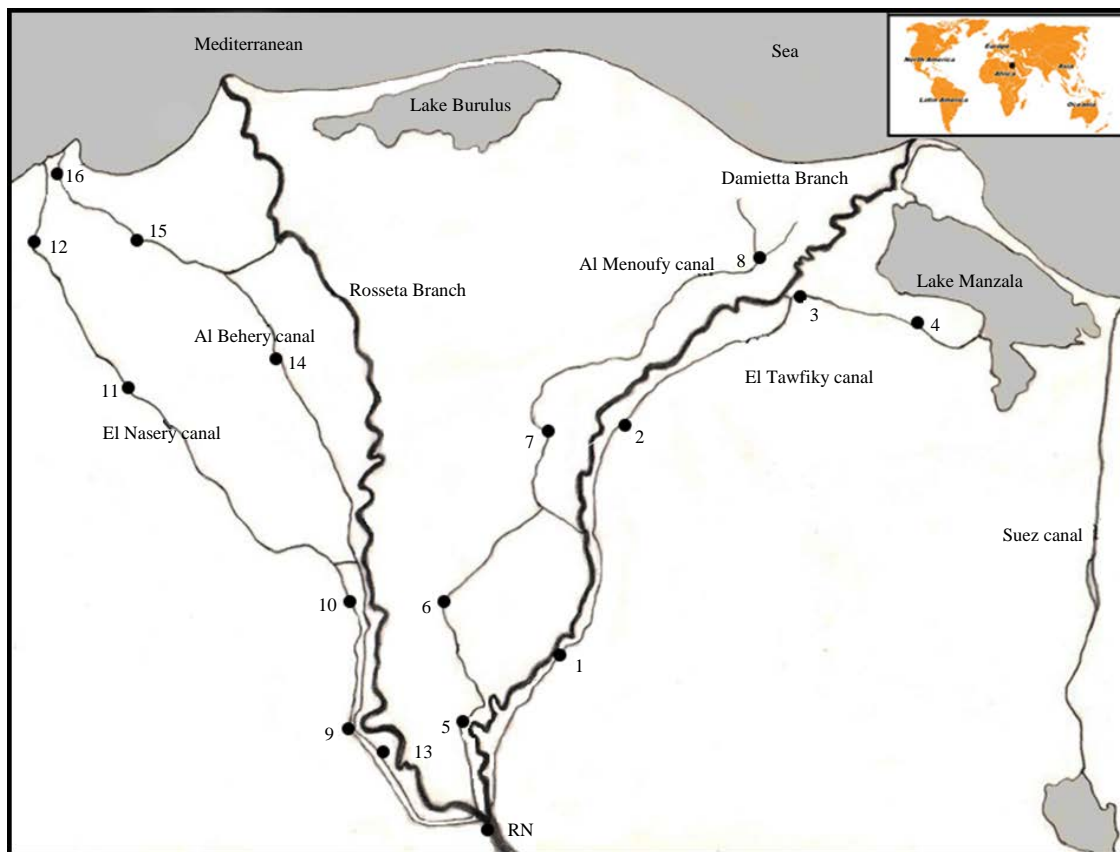


Fig. 1: Map showing the sampling stations in the four canals

in 100 mL glass bottle. All collected samples were stored on ice and kept in the dark until returned to the laboratory, where the samples were stored at -80°C till analysis.

Fatty acids extraction, methylation and quantification:

Freeze-dried sediment was extracted using modified Folch method (Taipale *et al.*, 2011; Galloway *et al.*, 2013). After addition of chloroform, methanol and water to 1 g sediment in a culture tube, the mixture was sonicated for 10 min, vortexed for 30 sec and finally centrifuged for 3 min at 3000 rpm to separate the phases. The lower (organic) phase was removed to a separate culture tube. Additional chloroform was added to the original sample to replenish the volume removed and the process of sonication, vortexing, centrifugation and removal was repeated two more times. The extracted organic layers were pooled and evaporated under nitrogen. Once the layers were dry, 1 mL of toluene and 2 mL of a 1% solution of sulfuric acid in methanol were added to re-suspend the lipids. This mixture was vortexed to ensure uniformity and then placed into a 50°C water bath for 16 h to allow for methylation of the FAs. Once the mixture was cooled,

2 mL of 2% KHCO_3 solution and 5 mL of 1:1, hexane : diethyl ether, were added, followed by vortexing and centrifugation for 2 min at 1500 rpm to separate the phases. The organic layer was again removed (this time the upper phase). To ensure total removal of FAs methyl esters (FAME), an additional 5 mL of hexane: diethyl ether, were added. The previous step was repeated and pooled the organic layers from the extractions. These organics were evaporated under nitrogen and the FAME subsequently dissolved into 1.5 mL hexane for Gas Chromatography (GC) analysis.

Fatty acids were analyzed by Gas Chromatography-Mass Spectrometry (GC-MS) using Agilent 7890 Series GC system interfaced to 5975 inert MSD with triple-axis detector MS. The total GC run time was 50 min and the carrier gas was helium at flow rate of 1.22 mL min^{-1} . The initial oven temperature was held at 90°C for 1 min and then reached 290°C in 6 min, after which it was held at this temperature for 30 min. Compound identification was performed by comparison with the chromatographic retention characteristics, mass spectral library of the GC-MS data system and external standards (Sigma-Aldrich) and quantified using total ion peak area and calibration curves of the external standards.

Table 1: Sources, variables and references of different fatty acids indicators

Source	Variable	References
Σ C16:0, C16:1 ω 7 and C20:5 ω 3	Diatoms (DFA)	Hill <i>et al.</i> (2011)
C16:1 ω 7/C16:0	Diatoms (DFA2)	Hill <i>et al.</i> (2011)
Σ C18:2 ω 6c, C18:3 ω 6 and C18:3 ω 3	Green algae (GFA)	Arts <i>et al.</i> (2009)
Σ C14:0, C16:0, C18:0, C18:1 ω 9t and C18:1 ω 9c	Cyanobacteria (CFA)	Arts <i>et al.</i> (2009)
Σ C16:0, C16:1 and C18:0	Domestic sewage (DSFA)	Reveille <i>et al.</i> (2003)
Σ C13:0+C15:0+C17:0	Bacteria (BFA)	Volkman <i>et al.</i> (1980) and Harvey (1994)
Σ C ₁₅₋₂₁ / Σ C ₂₀₋₂₄	Terrestrial plants (TARFA)	Volkman <i>et al.</i> (1980)
Σ C18:2 ω 6+C18:3 ω 3	Terrestrial plants (TPLFA)	Parrish <i>et al.</i> (1995) and Napolitano <i>et al.</i> (1997)

Statistical analysis: Differences in total abundance among canals and seasons were tested separately using one-way ANOVA followed by Tukey *post hoc* test. Table 1 shows the sources and variables of different fatty acids indicators.

RESULTS

A total of 34 fatty acids were identified from the surficial sediment samples including saturated (SFA), polyunsaturated (PUFA) and monounsaturated (MUFA) fatty acids. The dominant SFA found in sediment samples were C16:0 (palmitic acid) and C18:0 (stearic acid) which were found in all 60 samples. Dominant MUFA included C16:1 ω 7 (palmitoleic acid) and C17:1 ω 7 (cis-10-heptadecenoic acid, HPD) whereas, C18:1 ω 9 (oleic acid) was less important. The FAs composition in the four canals was marginally dominated by Saturated Fatty Acids (SFAs) over unsaturated fatty acids, with mean $\Sigma_{SFA}:\Sigma_{USFA}$ ratio of 1.16 ± 0.03 . Between the USFA family, ω 3 group had lower importance compared with ω 7 family which had highest importance during the four seasons with highest abundance in spring and least in winter (Tukey HSD < 0.05). The PUFA were present at all sediment samples but with low importance compared with MUFA, C22:6 ω 3 (cis-4, 7, 10, 13, 16, 19-docosahexaenoic acid) was the most important. The essential fatty acid (EFA) 22:6 ω 3 occurred in all canals with a range between 1.64 and 7.81 $\mu\text{g g}^{-1}$. The ANOVA analysis revealed significant seasonal differences in total fatty acid contents of sediment organic matter, different indicators and dominant fatty acids but no significant differences between canals were found. C17:1 ω 7 had high significant differences ($p < 0.0007$) between canals but showed non-significant temporal variations ($p > 0.2$). Total FAs decreased from spring, 2014 to winter, 2015 (Table 2) within a wide range of 408.82-86.28 $\mu\text{g g}^{-1}$ dry sediment. The *post hoc* test revealed that sediments in spring (64.01 ± 2.2 , $n = 15$) were enriched in FA relative to sediments collected in other seasons. Fatty acid contents before the bifurcation of the Nile non-significantly differed from the stations at the four canals but it was considerably higher than the annual mean.

The measured fatty acids ranged from C12-C24, with a typical strong even/odd predominance (annual average ratio of 3.68). Samples were characterized by a trimodal distribution with a prominent C_{max} at C16, C17 and C18. A prevalence of Lower Molecular Weight (LMW) fatty acids (<C20), was detected specifically in spring, compared with the High Molecular Weight (HMW). The $\Sigma_{LMW}:\Sigma_{HMW}$ (TARFA) varied significantly between seasons ($p < 0.005$), ranged from 3.56 in summer to 17.61 in spring. The mean LMW contributed to 85.7% of the total fatty acids. The presence of higher plant fatty acids (TARFA) in sediments from all the four canals was very low. The levels of 24:0 decreased from spring (annual mean of 5.95 $\mu\text{g g}^{-1}$) toward winter (4.86 $\mu\text{g g}^{-1}$ dry sediment). The TPLFA (summation of 18:2 ω 6 and 18:3 ω 3, indicator of terrestrial plant) was very low, its values never exceed 0.05 $\mu\text{g g}^{-1}$. The values of TPLFA were highest in winter (0.022) and least in spring (0.016 $\mu\text{g g}^{-1}$).

Fatty acids common in domestic sewage (Σ_{SEW}) were detected in high concentrations at all sampling stations, resulted in $\Sigma_{SEW}:\Sigma_{TFA}$ ratios between 0.29 and 0.67 (Table 1). In spite of the iso- and ante-isoforms fatty acids characterized bacterial contribution to sedimentary organic matter was not analyzed during this study, other bacterial FAs indicator (summation of C13:0, C15:0, C17:0, C16:1 ω 7 and C18:1) were detected at all sampling stations. The $\Sigma_{BACT}:\Sigma_{TFA}$ ratio varied significantly between seasons ($p < 0.00001$) but non-significant between regions. The ratio of $\Sigma_{BACT}:\Sigma_{TFA}$ ranged between 0.04 and 0.73 in all samplings. The calculated indicators of diatom dominance based on fatty acids showed seasonal differences (Table 2). The ratio of 16:1 ω 7/16:0 was lower in summer (mean; 0.18) and highest in winter (mean; 0.33) and the sum of 16:0, 16:1 ω 7 and 20:5 ω 3, was higher in spring (96.6 $\mu\text{g g}^{-1}$ dry sediment) than other seasons. Green algal indicators showed clear higher values in spring and summer (6.4 and 6.23 $\mu\text{g g}^{-1}$ dry sediment, respectively) and lower in winter and autumn (5.7 and 5.9 $\mu\text{g g}^{-1}$ dry sediment, respectively). Cyanobacteria indicator, $\Sigma_{CFA}:\Sigma_{TFA}$, was highest in spring (0.51) and least in summer (0.42). The calculated indicators showed significant temporal variations ($p < 0.05$).

Table 2: Composition and mean absolute concentrations in $\mu\text{g g}^{-1}$ dry sediment (± 0.1 SD) of single fatty acids, FAs classes as well as FAs class ratios along the four canals in four successive seasonal sampling campaigns

Fatty acids	Spring, 2014				Summer, 2014				Autumn, 2014				Winter, 2015								
	R	T	M	B	N	R	T	M	B	N	R	T	M	B	N	R	T	M	B	N	
C6:0	0.50	0.10	0.00	1.10	0.40	0.20	3.10	2.40	0.10	0.00	0.10	1.20	1.10	0.40	0.20	0.40	0.20	0.30			
C8:0	2.30	2.30	2.60	2.30	2.30	2.30	2.30	2.30	2.30	2.30	2.30	2.40	2.30	2.30	2.30	2.30	2.30	2.30	2.30		
C10:0	3.40	1.10	0.60	1.10	0.90	1.40	3.30	2.10	0.40	1.90	2.00	2.70	2.00	0.50	2.40	2.40	2.30	2.10			
C11:0	0.60	0.60	0.60	0.80	0.30	0.30	0.50	0.60	0.60	0.50	0.70	0.30	0.60	0.30	0.50	0.30	0.40	0.40			
C12:0	1.00	1.30	2.10	1.50	3.20	1.00	0.90	1.00	1.10	1.30	1.40	0.50	1.10	1.30	0.70	0.80	0.80	0.80			
C13:0	0.40	0.40	0.80	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40			
C14:1 ω 5	18.30	0.80	1.00	0.80	0.90	0.90	0.80	0.80	0.90	0.90	1.00	0.80	0.80	0.80	0.80	0.80	0.80	0.90			
C14:0	3.20	5.30	10.70	7.90	8.90	3.90	2.50	3.60	5.30	5.70	4.90	2.30	4.60	11.60	4.50	2.80	3.30	5.10			
C16:1 ω 5	1.30	1.40	1.50	1.40	1.60	1.20	1.10	1.20	1.30	1.20	1.30	1.10	1.10	1.00	1.20	1.20	1.20	1.10			
C15:0	1.40	2.00	3.60	3.10	3.30	1.70	1.00	1.40	2.00	2.00	1.40	0.90	1.90	3.80	1.3	1.10	1.60	2.00			
C16:1 ω 7	7.50	17.10	31.50	28.80	32.40	4.30	4.60	7.40	13.60	7.90	11.70	5.30	14.10	29.90	12.30	8.70	9.30	15.70			
C16:0	51.40	60.20	124.20	68.10	66.80	42.00	34.90	40.70	64.60	39.90	56.10	38.80	44.20	54.00	34.70	34.80	36.60	38.90			
C17:1 ω 7	79.70	72.00	75.40	78.40	77.40	45.80	73.40	56.90	18.00	52.00	74.40	92.00	50.90	3.40	58.80	28.80	59.70	41.30			
C17:0	1.30	1.60	2.60	2.00	2.40	1.60	1.20	1.20	1.40	1.60	1.70	1.00	1.40	1.80	1.30	1.10	1.40	1.30			
C18:2 ω 6	1.70	1.70	1.70	1.70	1.70	1.70	1.70	1.70	1.70	1.30	1.70	1.70	1.70	1.70	1.70	1.70	1.70	1.70			
C18:3 ω 3	1.50	3.10	1.60	1.60	1.80	1.70	1.60	1.80	1.50	1.60	1.70	1.50	1.60	1.60	1.90	1.70	1.50	1.60			
C18:1 ω 9c	4.40	5.30	6.00	5.60	6.80	5.30	4.90	4.50	4.30	5.10	5.20	3.60	5.10	4.80	4.70	4.60	4.80	4.60			
C18:1 ω 9t	1.00	1.10	1.10	1.10	1.10	1.10	1.10	1.10	1.10	1.10	1.10	1.10	1.10	1.00	1.10	1.10	1.10	1.10			
C18:0	20.80	21.50	37.10	19.90	18.80	17.90	15.80	15.50	27.00	13.80	21.60	16.60	17.00	20.00	14.70	15.80	16.40	16.00			
C20:4 ω 6	2.10	1.90	1.40	2.00	0.00	1.70	1.30	1.80	1.90	1.40	1.60	1.80	1.80	1.70	1.70	1.30	1.30	1.70			
C20:5 ω 3	2.20	3.20	3.80	4.10	3.10	2.20	2.20	2.80	2.70	2.30	2.40	2.00	2.50	2.00	2.10	2.00	2.10	2.20			
C20:3 ω 6	1.70	1.70	1.70	1.80	1.80	1.70	1.70	1.70	1.70	1.70	1.70	1.70	1.70	1.70	1.70	1.70	1.70	1.70			
C20:2 ω 6	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.10	1.10	1.50			
C20:1 ω 9	1.30	1.40	1.70	1.60	1.20	1.50	1.20	1.30	1.30	1.30	1.60	1.40	1.30	1.50	1.40	1.30	1.40	1.40			
C20:3 ω 3	1.50	1.60	1.60	1.60	1.60	1.50	1.50	1.60	1.50	1.50	1.10	1.50	1.50	1.50	1.50	1.50	1.60	1.50			
C20:0	1.30	1.30	1.80	1.40	1.50	1.30	1.10	1.10	1.20	1.40	1.40	1.00	1.40	1.20	1.20	1.10	1.20	1.20			
C21:0	1.70	1.90	1.60	1.60	1.60	1.40	1.70	1.60	2.00	1.50	1.60	1.80	1.50	1.10	1.50	1.30	1.50	1.40			
C22:6 ω 3	2.20	1.80	3.60	3.10	2.60	4.00	3.10	3.40	2.30	3.00	3.50	1.80	2.50	2.50	2.10	2.60	2.70	3.20			
C22:2 ω 6	1.80	0.40	0.60	0.00	0.00	1.20	0.40	0.40	1.80	0.40	0.50	0.00	0.40	1.80	1.80	0.00	1.30	0.90			
C22:1 ω 9	1.90	0.90	1.90	1.40	1.10	2.00	1.10	1.30	1.30	1.90	0.70	1.10	1.80	1.30	0.30	0.50	2.10	0.60			
C22:0	3.50	3.50	4.60	3.90	5.00	3.70	3.10	3.00	3.00	3.80	3.90	2.50	3.40	3.20	2.90	3.10	3.50	3.20			
C23:0	1.70	1.40	2.00	3.50	2.30	2.00	1.60	1.60	1.60	1.90	1.80	1.40	1.70	1.60	1.50	1.50	1.80	1.60			
C24:1 ω 9	1.80	2.10	2.40	2.50	2.70	2.00	1.80	1.80	2.20	2.50	2.20	1.80	2.10	3.00	1.90	2.00	2.30	2.40			
C24:0	5.50	5.20	7.10	6.60	10.10	6.10	4.70	4.70	4.50	6.10	6.10	3.50	5.60	4.60	4.30	4.50	5.30	4.60			
TFA	233.10	228.70	342.10	264.20	267.70	168.50	183.20	176.00	177.70	172.70	222.20	198.70	183.50	170.60	172.80	136.40	176.50	166.60			
GFA ^b	5.50	6.40	7.10	6.70	7.90	6.50	5.90	5.60	5.30	6.10	6.20	4.60	6.20	5.90	5.80	5.60	5.80	5.70			
BFA ^d	15.60	27.10	44.90	40.60	46.10	14.00	12.70	15.70	22.40	17.60	21.10	11.80	23.50	41.30	20.70	16.60	18.10	24.70			
SFA	98.10	108.20	200.10	121.80	126.00	85.00	76.50	81.50	115.70	82.10	105.70	75.80	88.50	106.30	72.80	72.30	77.20	80.00			
USFA	135.00	120.50	142.00	142.40	141.60	83.50	106.70	94.60	62.00	90.50	116.50	122.90	95.00	64.30	99.90	64.10	99.30	86.60			
LMWFAs	205.00	205.70	311.60	234.30	234.80	140.50	155.10	149.60	154.40	145.00	195.00	177.40	157.00	146.60	150.00	114.30	149.60	142.50			
HMWFAs	20.00	17.30	23.80	22.70	25.40	22.50	17.60	17.70	18.50	21.30	20.30	14.00	18.90	18.90	16.30	15.50	20.50	17.80			
PUFA	17.90	18.40	19.40	20.90	16.30	19.20	16.70	18.20	18.20	16.70	17.40	14.80	16.90	17.50	17.50	15.20	16.80	17.60			
MUFA	117.10	102.10	122.60	121.50	125.30	64.20	90.00	76.40	43.80	73.80	99.10	108.10	78.20	46.80	82.50	48.90	82.60	69.10			
DSFA ^c	86.50	107.30	203.60	126.60	129.30	72.30	62.20	70.70	112.60	69.60	97.00	66.20	83.30	113.60	68.70	66.00	69.70	78.30			
DFA ^a	61.10	80.40	159.60	101.00	102.30	48.60	41.60	50.90	80.90	50.00	70.10	46.00	60.70	85.90	49.10	45.50	48.00	56.80			
DFA2 ^a	0.19	0.27	0.32	0.41	0.48	0.20	0.15	0.18	0.21	0.22	0.20	0.15	0.26	0.55	0.41	0.29	0.31	0.37			
CFA ^b	89.70	112.60	214.30	134.50	138.20	76.30	64.70	74.20	117.90	75.30	102.00	68.50	87.90	125.20	73.20	68.90	73.10	83.40			
TPLFA ^f	10.30	110.90	13.10	10.30	9.20	6.30	80.80	80.50	8.30	6.80	9.60	12.70	8.30	7.70	90.20	7.40	7.30	80.00			
TARFA ^e	0.014	0.023	0.011	0.015	0.013	0.021	0.020	0.021	0.018	0.017	0.016	0.016	0.018	0.019	0.022	0.026	0.018	0.022			
$\Sigma_{\text{SFA}}:\Sigma_{\text{USFA}}$	0.73	0.90	1.41	0.86	0.89	1.02	0.72	0.86	1.87	0.91	0.91	0.62	0.93	1.65	0.73	1.13	0.78	0.92			
$\Sigma_{\text{SEW}}:\Sigma_{\text{FAs}}$	0.37	0.47	0.60	0.48	0.48	0.43	0.34	0.40	0.63	0.40	0.44	0.33	0.45	0.67	0.40	0.48	0.40	0.47			
$\Sigma_{\text{BACT}}:\Sigma_{\text{FA}}$	0.07	0.12	0.13	0.15	0.17	0.08	0.07	0.09	0.13	0.10	0.09	0.06	0.13	0.24	0.12	0.12	0.10	0.15			

R: RN station, T: El Twafiky, M: El Menoufy, B: El Behery and N: El Narsery, ^a: Hill *et al.* (2011), ^b: Arts *et al.* (2009), ^c: Reveille *et al.* (2003), ^d: Volkman *et al.* (1980) and Harvey (1994), ^e: Volkman *et al.* (1980), ^f: Parrish *et al.* (1995) and Napolitano *et al.* (1997)

DISCUSSION

Surface sediment fatty acid profiles showed seasonal variability likely due to their strong relationships with seasonally variant environmental variables (Stewart *et al.*, 2005). This study supports this general trend, finding discernible seasonal patterns based on surface sediment fatty acid profiles. However, no significantly related environmental covariates were reported as environmental drivers such as light penetration and temperature. This may be supported the suggestion that the observed seasonal trends in fatty acid profiles most likely were an indirect results of environmental covariates but a direct result of algal community composition shifts. Each species likely had a unique fatty acid profile, thus seasonal compositional shifts resulted in an overall shift on the assemblage fatty acid profile (Sushchik *et al.*, 2010). The sampling locations of this study are acting as a sinking pool of highly diverse microorganisms originated from water column, macrophytes and surficial sediment fauna and flora. Phytoplankton structure in the four canals composed of 7 groups with diatoms, green algae and cyanobacteria, as main groups, with more than 150 species. The contribution of the three groups to the total density and community structure showed temporal variability with highly diverse species composition (NIOF., 2015). The littoral banks of the canals are occupied by more than 11 species of both submerged and emerged macrophytes (NIOF., 2015), especially those of submerged macrophytes can provide a hosting substrates for different organisms specifically epiphytic microalgae (Honeyfield and Maloney, 2015). With the high velocity of the water current and wind actions, epiphytic microalgae de-attach and sink to the sediment. The settlement of phytoplankton and de-attached epiphytic microorganisms in addition to the fauna and microflora of the surface sediment itself community can support the temporal shifts in the fatty acids profile in the four canals which agree with the findings of (Honeyfield and Maloney, 2015).

Two fatty acids indicators were used to identify the contribution of terrestrial plant input to the sedimentary organic matter in the four canals. Generally, fatty acids from higher plant waxes show carbon numbers >C20 with a maximum at C24 or C26, whereas, those from algae and bacteria range between C12 and C20 with a maximum at C16 (Simoneit, 1978; Gao *et al.*, 2008). The results showed a clear prevalence of Lower Molecular Weight (LMW) fatty acids (<C20) all over the year with very low presence of higher plant fatty acids (HMW) from all sites in the four canals, the annual

mean of the $\Sigma_{LMW} : \Sigma_{HMW}$ was 9.14. Hu *et al.* (2006) suggested that the high values of the $\Sigma_{LMW} : \Sigma_{HMW}$ ratios suggest that algae-derived fatty acids are the major source of total fatty acids in the surface sediments. Jaffe *et al.* (2006) conducted that the $\Sigma_{LMW} : \Sigma_{HMW}$ ratios of 1.8-23.2 mirrored that the contribution of fatty acids from algae is highly greater than that from higher plant waxes which had minor contribution. Terrestrial plants generally contain large amounts of 18:2 ω 6 and 18:3 ω 3 (Ackman, 1986). Parrish *et al.* (1995) examined the fatty acids in particulate matter in a shallow cove where terrestrial inputs were large and found amounts of 18:2 ω 6 and 18:3 ω 3 exceeding 4.7% of total fatty acids. Budge and Parrish (1998) suggested that the sum of 18:2 ω 6 and 18:3 ω 3 could be used as an indicator for the relative importance of terrestrial material. By examining marine sediment which was assumed to contain little or no terrestrial material, a threshold of 2.5 was assigned to this indicator. In this way, samples with values above this may be considered to have terrestrial material as a significant source of organic matter (Countway *et al.*, 2007). Preliminary studies with compound specific isotope analysis over support for this marker as the analyses showed that 18:2 ω 6 and 18:3 ω 3 in the riverine samples were depleted in ^{13}C compared to the other acids and had their origins in terrestrial plants, rather than in freshwater plankton. The riverine collected samples during this study contained much less of the summation of these two acids that still lower than 0.05, which mirrored the underestimation of terrestrial plants (allochthonous) as a contributor to sedimentary organic matter of the four canals.

The ratio of C16:1/C16:0 can be used to distinguish between diatom- and other algal-derived organic matter and fatty acids. Values >1.6 are thought to be evidence of fatty acids originating from diatoms in different aquatic systems (Budge and Parrish, 1998). In this study, this ratio varied from 0.05-0.7 with mean of 0.33. This fatty acid-based index of diatom abundance did not correspond with the dominance of diatom reported in the water column, phytobenthos and epiphytic communities of the four canals (NIOF., 2015) and recently published data concerning dominance of diatoms in lotic systems in Egypt (epiphytic communities and epipelagic communities), (Abd El-Karim, 2014). The USFA compounds are hard to preserve in their original amounts in sediments owing to their labile characteristics of rapid loss through bacterial degradation and/or zooplankton grazing (Carrie *et al.*, 1998). However, due to the higher susceptibility of unsaturated fatty acids to the biological and chemical degradation of living microalgae during the sedimentation, the ratio of 16:1/16:0 in

surface sediments is usually well below 1 (Birgel *et al.*, 2004). However, during the process of sedimentation, unsaturated fatty acids are more vulnerable to biological and chemical degradation than saturated ones and the results in the ratio of C16:1/C16:0 in surface sediments being well below 1 (Birgel *et al.*, 2004). In this study, no value was found for the C16:1/C16:0 ratios that was >0.7, suggesting a deep degradation of this fatty acid.

The saturated short chain fatty acids dominated the total fatty acids in the four canals. The 16:0 (palmitic) and 18:0 (stearic) acids were the most important fatty acids along the four canals year round. Palmitic and stearic acids were detected as most abundant fatty acids in rivers (Boechat *et al.*, 2014; Berg *et al.*, 2002), freshwater lakes (Fang *et al.*, 2014), transitional (Budge and Parrish, 1998; Bergamino *et al.*, 2014) and marine (Volkman *et al.*, 2008) sediments. However, some fatty acids such as palmitic and stearic acids are ubiquitous. These fatty acids have highest concentrations in raw sewage (Quemeneur and Marty, 1994; Reveille *et al.*, 2003; Boechat *et al.*, 2014), periphyton assemblages (Honeyfield and Maloney, 2015), cyanobacterial mats in hyper saline environment and algal cultures (Cranwell *et al.*, 1987; Hempel *et al.*, 2012). Moreover, kitchen wastes derived from vegetable oils and animal fats show typical distributions of fatty acids dominated by palmitic and stearic acids (Boechat *et al.*, 2014). Pearson correlation (r) matrix showed no significant correlation between palmitic and stearic acids and water environmental parameters; NH₄-N, NO₃-N, NO₂-N, BOD, COD, DO, EC, pH, PO₄ and OM in sediment and mud sediment. On the other side, 16:0, 18:0 and most classes showed high significant correlation (0.56-0.74) with chlorophyll a concentrations (Chl-a is close to 200 µg L⁻¹) in both El Behery and El Towfiky canals (NIOF., 2015) which suggested that the main contributor to palmitic and stearic acids is autochthonous primary producers. Qin *et al.* (2010) and Xu *et al.* (2014) conducted that phytoplankton was expected to have a high contribution with total fatty acids and fatty acids classes that exceed allochthonous inputs to be the dominant source of OM in sediment based on the field observations of a high concentration of chlorophyll a (Chl-a is close to 1 mg L⁻¹) and its biphasic life cycle including both pelagic and benthic stages. Contradict the low concentrations of FAs in river sediment organic matter either indicated a small contribution of algae and other autochthonous autotrophic organisms, a fact also suggested by the low chlorophyll-a concentrations (2 µg L⁻¹) in Rio das Mortes, Brazil, river (Boechat *et al.*, 2014). The fact that

supports the idea of the autochthonous origin of palmitic and stearic acids is the decrease, non-significant, of the concentrations of these acids downstream the big cities located on different canals compared with upstream stations.

CONCLUSION

The fatty acids composition of the surface sediments shows that saturated fatty acids are dominated by 16:0 and 18:0, which are ambiguous biological markers of microalgae. However, the dominant MUFA included C16:1ω7 and C17:1ω7, whereas, C18:1ω9 was less important. Between the USFA families, ω3 group had lower importance compared with ω7 group. As confirmed by fatty acids investigation, microalgae are the major contributor to both organic matter and carbon cycling in surface sediment of the four canals the year round whereas, OM drive from terrestrial plant were overcome. Between microalgae, diatoms had relatively higher contribution than both green algae and cyanobacteria in the sedimentary organic matter. Still, bacteria and sewage were lower predictors of organic matter, whereas, the contribution of land-plant materials still underestimation. Both land-plant and bacterial fatty acids would need to have been heavily investigated to avoid uncertainties of the contribution of both to the organic matter and carbon cycle in the four canals.

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