3. MATERIALS AND METHODS

3.1 Study Area:

3.1.1. Nubaria canal:

Location

Nubaria Canal is the largest main canal in Western Nile Delta. It is mainly fed from the Nile River. Nubaria Basin is located in Behira Governorate. It is situated by Longitudes 30^{o} 05'and 30^{o} 45' East, and Latitudes 30^{o} 30'and 31^{o} 00' North. [72] The location is situated 418 kilometres north (7°) of the approximate center of Egypt and 101 kilometers North West (319°) of the capital Cairo. It is mainly fed by fresh water from El-Rayah El-Nassery and El-Rayah El-Beheiry. It serves a total area of 373,800 hectare (ha). [73]

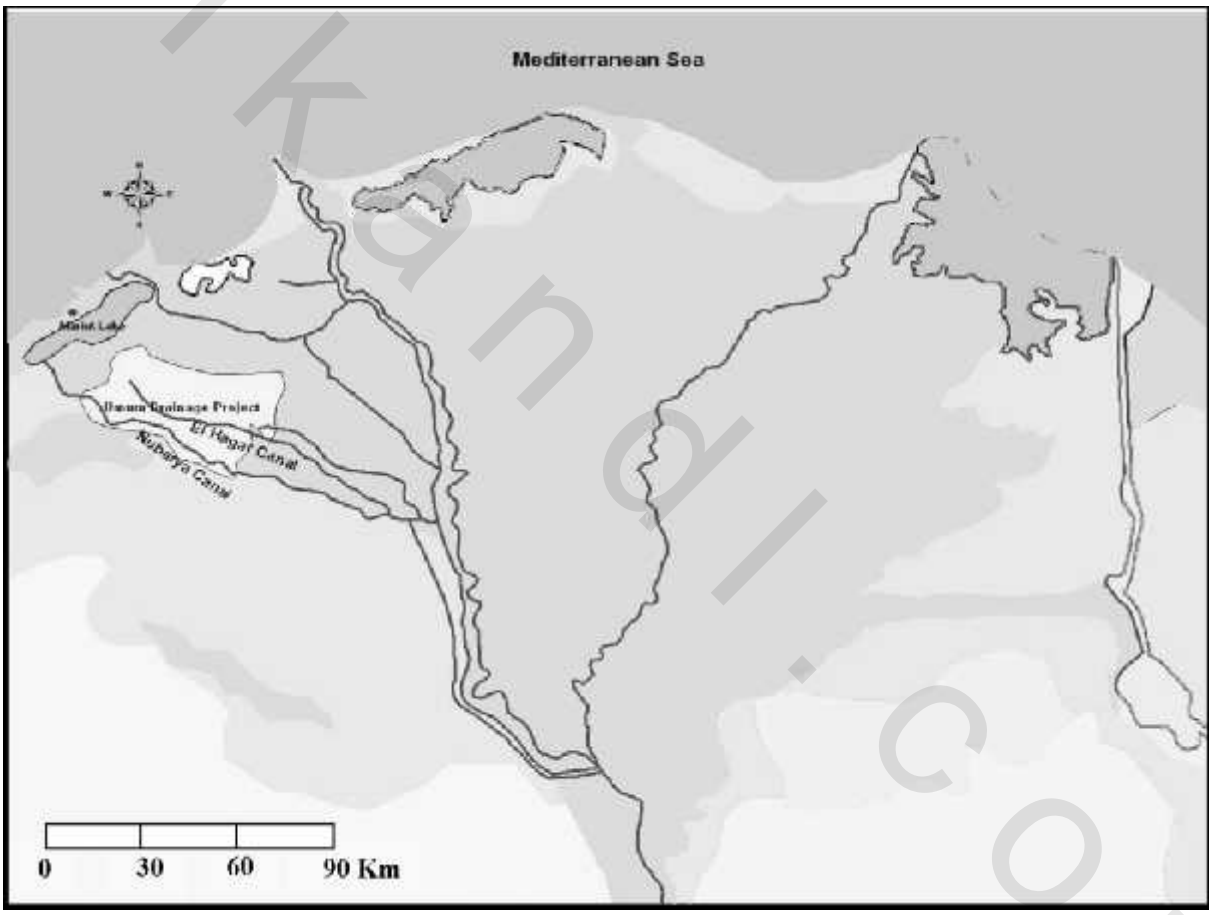

Figure (3.1): Geographic location of Nubaria Canal [73]

The canal has been developed several times to cope with the horizontal expansion in the newly reclaimed areas in western desert. Behind the intake at Km 46, there is an area of 280,000 hectare has to be irrigated with 5 BCM per year.

Sample Sites:

Table 3.1: Nubaria sample site

SAMPLE SITE	LOCATION
N[(1)]C[(2)] 0.5	Boline Bridge
ND[(3)] 5.0	Delingate Drain
NC 6.0	Kheneza Bridge
NC 7.0	Rayyah El Nassery
NC 28.7	Boustan Bridge and Regulator
ND 29.0	Boustan Drain
NC 45.2	Al-Abd Bridge
ND 46.5	El-Umum Drain
NC 50.3	El Modier Bridge (Boustan Canal)
ND 51.0	Drain #3
ND 55.05	Drain #1
NC 60.6	Janaclis Regulator
NC 81.0	Noubaria WTP[(4)]
NC 96.0	Borg El Arab (KM 40) WTP

(1)N: Nubaria (2) C: Canal (3) D: Drain (4) WTP: Water Treatment Plant

Nubaria canal is a main irrigation canal at the north west of Egypt (Fig. 1) [(73)]. It was designed to irrigate eight hundred thousand acres of agricultural lands, along its course which extends 100 km. Several hydraulic structures control the flow and water levels through the canal. [(72)]

3.1.2 Mahmodia Canal:

Al-Mahmoudia canal in northern edge of Beheira Governorate, west part of Nile Delta, has important role in the economic development and prosperity of the people in Beheira and Alexandria Governorates. It has been exploited to support agriculture, fisheries, public water supply, industry, hydroelectric power and recreation. [(3)]

Al-Mahmoudia canal is located at the northern edge of Beheira Governorate. The canal off-takes from Rosetta branch at km 194.200. The actual served area for the canal is 130,200 hectares. The total length of the canal is 77.170 km and there are seventy canals off-take from this canal. Al-Mahmoudia canal has three sources of water; two fresh water sources which are from Rosetta branch via El-Atf pump stations at the head of the canal, and Al-Khandaq Eastern canal at km 13.200 on Al-Mahmoudia canal, the third is drainage water from Zarkon drain at km 8.500 on Al-Mahmoudia canal via Edko irrigation pump station which lifting part of Zarkon drain water into Al-Mahmoudia canal. The canal receives pollutants from point and non-point sources. [(74, 75)]

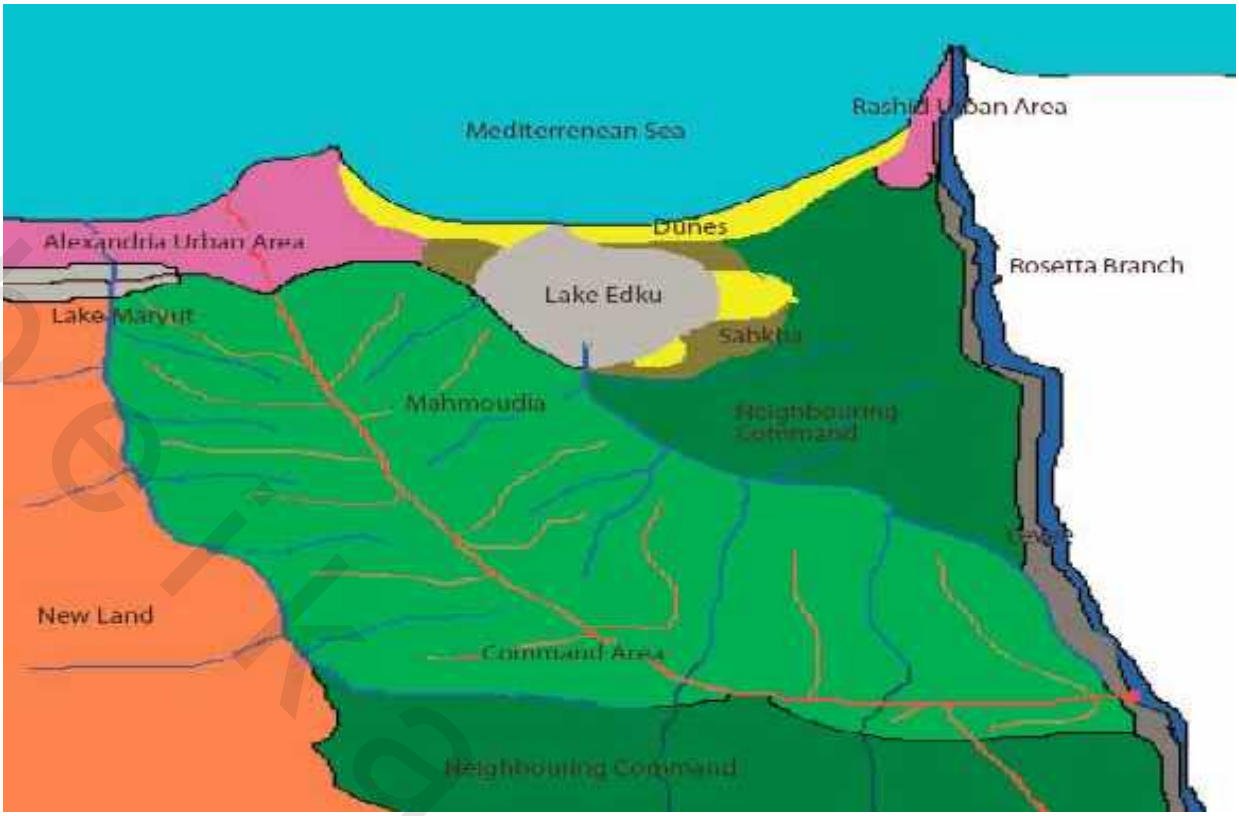

Figure (3.2): Geographic location of Mahmoudia Canal [76]

Sample Sites:

Table 3.2: Mahmoudia sample sites

SAMPLE SITE	LOCATION
M[1]C[2]-0	Rosetta Branch of the Nile where it enters Mahmoudia
MC-1	Mahmoudia City in front of Atef Pump Station
MD[3]-8.9	Zarkon Drain
MC-9	Zarkon
KC-15	Khandak before Mixing
MC-15.8	Khandak Canal after Mixing in Zawyat Ghazal Village
MC-26	Abou Homos
MC-42	Kafr El Dawar
MC-55	Korshid
MC-61.3	Siouf WTP[4] Intake
MC-66	Nozha WTP Intake
MC-73	Manshia WTP Intake

(1)M:Mahmoudia (2)C: Canal (3)D: Drain (4)WTP:Water Treatment Plant

3.1.3. Selection of Sampling Sites:

The sampling sites chosen for this study were the Nubaria and Mahmoudia canals. Nubaria canal was chosen because it is the largest main canal in western Nile Delta, and Mahmoudia canal serves a total command area of about 305000 feddans through 70 branch canals.

The major sources that affected the water quality and the trophic status of the two canals were agricultural runoff (through non-point source), animal husbandry effluents, untreated and semi-treated domestic effluents and industrial waste water and textile (through point sources).

This study was investigated physical, chemical and bacteriological water quality parameters in Nubaria and Mahmoudia canals through one year.

The selection of the sampling sites were choose to cover canals and show changes that happened in water quality through two canals. The minimum distance between sampling points is 1000 m.

The sampling sites were selected by GPS (Global Positioning System).

Table 3.3: Nubaria and Mahmoudia sample site codes

Nubaria Canal

Sample Point	Code
Boline Bridge	N01
Delingate Drain	N02
Kheneza Bridge	N03
Rayyah El Nassery	N04
Boustan Bridge	N05
Boustan Drain	N06
Al-Abd Bridge	N07
El-Umum Drain	N08
El Modier Bridge	N09
Drain #3	N10
Drain #1	N11
Giancalis Regulator	N12
Noubaria WTP	N13
(KM 40) WTP	N14

Mahmoudia Canal

Sample Point	Code
Rosseta branch	H01
Zarkon Drain	H11
Zarkon	H02
End of Khandak	H03
Zawyat Ghazal	H04
Abou Homos	H05
Kafr El Dawar	H06
Korshid	H07
Siouf WTPIntake	H08
Nozha WTP Intake	H09
Manshia WTP Intake	H10

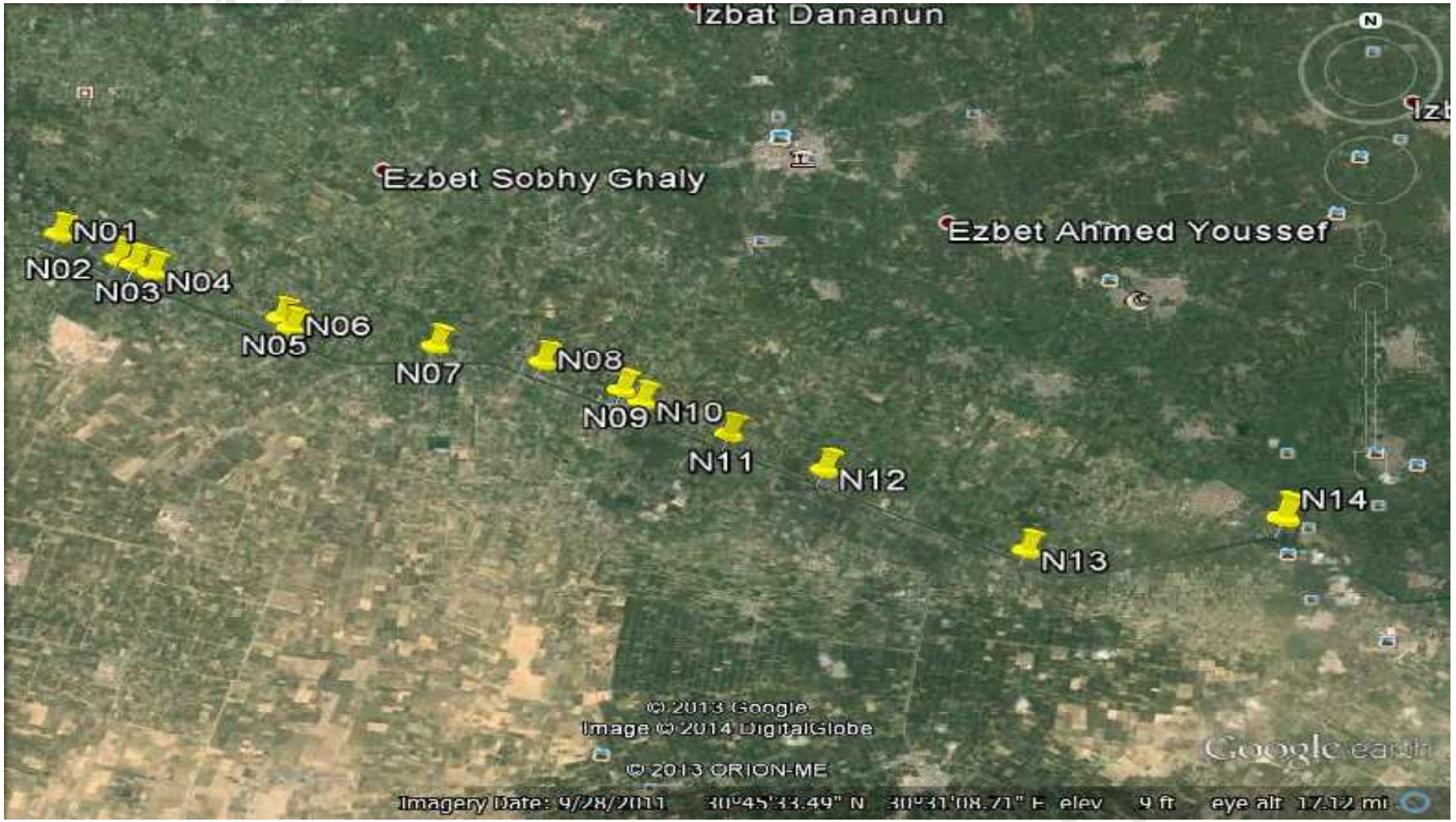

Figure (3.3): Satellite image of Nubaria canal showing location of sampling sites

Figure (3.4): Satellite image of Mahmoudia canal showing location of sampling sites

3.2. Field trips

24 field trips were performed during October 2012 to September 2013 in Nubaria and Mahmoudia canals. The selection of these periods to cover the seasonal variations throughout the year. This period covers all operational changes and weather conditions. The area of the study area is 96 Km and 73 Km in Nubaria and Mahmoudia canals.

3.2.1. Collection of samples

900 water samples were collected during this study (25 sample for 2 canals during 1 month * 3 replicates *12 month).Samples were collected in 1 liter plastic bottle and 1 liter glass bottle refrigerated and brought to the laboratory for detailed physico-chemical and bacteriological analysis. Temperature and Dissolved Oxygen were determined in the field.

3.3 Water analysis

Analytical reagent grade chemicals purchased from (HACH, Merck, Panreac, Chemlab, Ultra Scientific, Scharlau, and Sigma-Aldrish).They were used without further purification ultra pure water was used for blank and standards preparation.

The physico-chemical and bacteriological analysis was performed following The Standard Method for the Examination of Water and waste Water. [77]

3.4. Physical and Chemical parameters

3.4.1Color

Visual Comparison method 2120B

Color is determined by visual comparison of the sample with known concentrations of colored solutions. Comparison also may be made with special, properly calibrated glass color disks. The platinum-cobalt method of measuring colour is the standard method, the unit of color being that produced by 1 mg platinum/L in the form of the chloroplatinate ion. The ratio of cobalt to platinum may be varied to match the hue in special cases; the proportion given below is usually satisfactory to match the color of natural waters.

3.4.2. Turbidity

Nephelometric Method 2130B

Turbidity was measured for samples by using turbidity meter HACH 2100AN.

This method is based on a comparison of the intensity of light scattered by the sample under defined conditions with the intensity of light scattered by a standard reference suspension under the same conditions. The higher the intensity of scattered light, the higher the turbidity. Formazin polymer is used as the primary standard reference suspension.

Turbidity can be determined for any water sample that is free of debris and rapidly settling coarse sediment. Dirty glassware and the presence of air bubbles give false results.

‘‘True color,’’ i.e., water color due to dissolved substances that absorb light, causes measured turbidities to be low. This effect usually is not significant in treated water.

3.4.3 Hydrogen ion concentration (pH)

Laboratory method 4500B

Hydrogen ion concentration was measured for samples by using pH meter HACH HQ 11d.

pH was measured using a pH meter. The pH meter was calibrated before making pH measurements. For calibration standard buffers of pH 4.00, 7.00 and 10.00 were used. pH of water indicates the hydrogen ion concentration in water. The concept of pH was put forward by Sorenson in 1909. It is expressed as the logarithm of the reciprocal of the hydrogen ion concentration in moles/liter at a given temperature. While the alkalinity or acidity measures the total resistance to the pH change or buffering capacity, the pH gives the hydrogen ion activity. The pH scale extends from 0 (very acidic) to 14 (very alkaline) with 7 corresponding to exact neutrality at 25ºC. pH was used in the calculation of carbonate, bicarbonate and CO_2, corrosion, stability index etc.

$pH = - \log ([H^+])$

3.4.4. Conductivity

Electrometric Method 4500B

Conductivity was measured for samples by using conductivity HACH HQ 14d.

Electrical conductivity (EC) is a measure of how conductive the water is to electrical current. Greater the ion concentration, greater is the EC. Generally higher the EC, higher is the total dissolved solids. Electrical Conductivity is an indirect measure for finding the total dissolved solids in a water body. To convert the electrical conductivity of a water sample (micro Siemens per cm, µS/cm) to the concentration of total dissolved solids (ppm), the conductivity must be multiplied by a factor between 0.46 and 0.9 (depending on the unique mixture of the dissolved materials). A widely accepted conversion factor is 0.67 TDS (ppm) = Conductivity {(µS/cm) x 0.67}.The instrument used for measuring conductivity is conductivity meter.

3.4.5 Chloride

Argentometric Method 4500- Cl^- B

Chloride was measured for samples by using Solarus Digital Burette 50 mL (9392050).

Chloride is a chemical the human body needs for metabolism (the process of turning food into energy). It also helps keep the body's acid-base balance. The amount of chloride in the blood is carefully controlled by the kidneys.

3.4.6 Alkalinity

Titration Method 2320B

Alkalinity was measured for samples by using Solarus Digital Burette 20 ml (9392020).

$$\text{Alkaliniy, mg CaCO3/L} = \frac{\text{A} * \text{N} * 50000}{\text{ml sample}}$$

Where:
A= ml standard acid used
N= Normality of Standard acid

3.4.7 Total Dissolved Solids

Total Solids at 103-105°C- 2540 B

Total dissolves solids (TDS) are naturally present in water or are the result of mining or some industrial treatment of water. TDS contain minerals and organic molecules that provide benefits such as nutrients or contaminants such as toxic metals and organic pollutants [(78)].

3.4.8 Total Suspended Solids

Total Suspended Solids Dried at 103–105°C- 2540 D

$$\text{mg Total Suspended Solids/L} = \frac{(\text{A} - \text{B}) * 1000}{\text{Sample Volume, ml}}$$

Where:
A = weight of filter + dried residue, mg, and
B = weight of filter, mg.

3.4.9 Temperature

Temperature of water samples measured in field by the HACH HQ40d portable meter.

3.4.10 Hardness

EDTA Titrimetric Method 2340C

Total hardness was measured for samples by using Solarus Digital Burette 20 ml (9392020).

The determination of the total hardness of water is based on a complexometric titration of calcium and magnesium with an aqueous solution of the disodium salt of EDTA at pH value of 10. [(79)]

3.4.11 Dissolved Oxygen

Membrane Electrode Method 4500-O G

Dissolved Oxygen was measured for samples in field because this is a field test should be performed on site.

It measured by using HACH HQ40d portable DO.

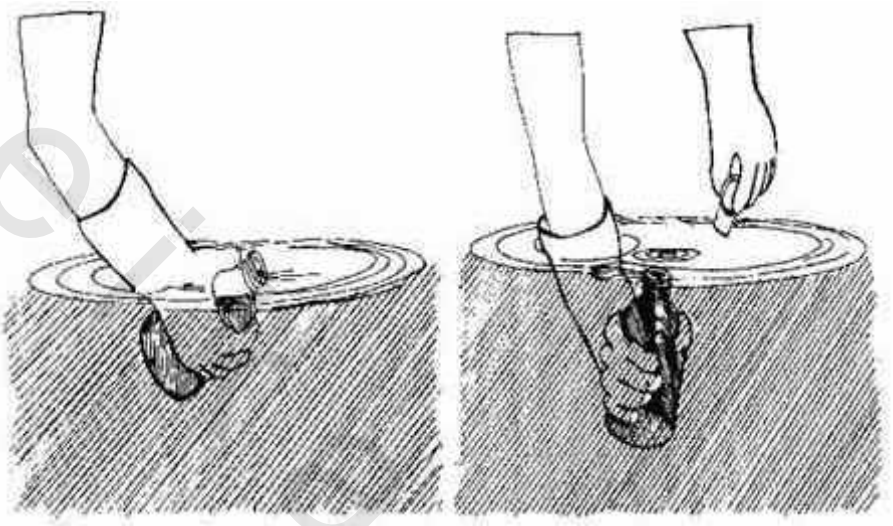

Figure (3.5): Taking a water sample for DO analysis [80]

Point the bottle downstream and fill gradually. Cap underwater when full.

3.4.12 Biochemical Oxygen Demand (BOD)

5210 B. 5-Day BOD Test

BOD was measured for samples by using HACH BOD Track.

A measured sample of sewage or wastewater is placed in one of the amber bottles on the apparatus and the bottle is connected the instrument. Above the sewage or water sample is a quantity of air, which contains 21 percent oxygen. Over a period of time, bacteria in the sewage consume dissolved oxygen to oxidize organic matter present in the sample. The air in the closed sample bottle replenishes the used oxygen, resulting in a drop in air pressure in the sample bottle. The BODTrak Apparatus measures the drop in pressure and displays results directly as mg/L BOD. During the test period (usually five days) the sample is continually agitated by a magnetic stirring bar. Carbon dioxide is produced by the oxidation of organic matter and must be removed from the system so that the pressure difference observed is proportional only to the amount of oxygen used. This is accomplished by the addition of a few crystals of lithium hydroxide in the seal cup of each sample bottle. The electromagnetic stirring mechanism provides adequate agitation to effectively maintain rapid transfer of oxygen from the liquid sample to the air above. The BODTrak Apparatus is free of leaks and has an effective carbon dioxide absorption system. The instrument also has accurate pressure sensors for reading pressure changes.

The BODTrak Apparatus is a practical, convenient and economical answer to BOD testing.(81)

3.4.13 Chemical Oxygen Demand (COD)

5221 B Closed Reflux Method

COD was measured by using COD Reactor HACH

3.4.14 Ammonia

4500-NH_3 D Ammonia Selective Electrode Method

The ammonia-selective electrode method is applicable over the range from 0.03 to 1400 mg NH3-N/L.

Inorganic parameters

Inorganic parameters were measured by Spectrophotometer lambda 25 (Double beam UV / Visible) PerkinElmer.

3.4.15 Sulphate

Turbidimetric Method 4500-SO_4^{2-} E

A simple and precise turbidimetric method of determining sulfate S in water samples is described. It involves measurement of the turbidity formed when an aliquot of a barium chloride-gelatin reagent is added to an acidified sample. The method is sensitive and accurate and permits determination of microgram amounts of sulfate S present in water samples. (82)

3.4.16 Fluoride

SPADNS method 4500 D

This method relies on the fact that when fluoride reacts with certain zirconium dyes, a colorless complex anion and a dye are formed. The complex, which is proportional to the fluoride concentration, tends to bleach the dye which therefore becomes progressively lighter as the fluoride concentration increases. In the case of the fluoride ion reaction with Zr-SPADNS (sodium 2-(parasulphophenylazo-) 1,8-dihydroxy-3,6-naphthalene disulphonate), the resulting colored complex was measured in a spectrophotometer at 570 nm. Distillation is necessary for samples containing high concentration of dissolved solids. The dissolved solids interfere with the fluoride analysis. Alkalinity, aluminum, iron and sulphates have negative effect on the results while chloride and phosphate has positive effect. (83)

3.4.17 Nitrate

Ultraviolet Spectrophotometric Screening Method 4500-NO^{-3} B

3.4.18 Nitrite

4500-NO^{-2} B Colorimetric Method

Nitrite (NO^{-2}) is determined through formation of a reddish purple azo dye produced at pH 2.0 to 2.5 by coupling diazotized sulfanilamide with N-(1-naphthyl)-ethylenediamine dihydrochloride (NED dihydrochloride). The applicable range of the method for spectrophotometric measurements is 10 to 1000 μ g NO^{-2}-N/L.

3.4.19 Silica

4500-SiO_2 C- Molybdosilicate Method

3.4.20 Phosphate

4500-D Stannous Chloride Method

3.5 Heavy Metals

Metals such as lead (Pb), Manganese (Mn), Cupper (Cu), Iron (Fe), Molybdenum (Mo) and Cadmium (Cd) in water samples were determined by Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES) PerkinElmer 5300 DV using the EPA method 200.7. [84]

3.5.1. Principle of Method

The instrument measure characteristic atomic emission spectra by optical spectrometry. Samples were nebulized and the resulting aerosols were transported to plasma torch. Each elements has a specific emission spectra produced by radio frequency inductively coupled plasma, the spectra dispersed by a grating spectrometer, and the intensities of spectra monitored at specific wavelengths by a photosensitive device. Photocurrent from the photosensitive device processed and controlled by computer system. Background correction technique was required to compensate for variable background contribution to the determination of the analytes.

3.5.2. Collection of samples

Collect samples for Heavy metals analyses in glass bottle. Samples were collected filtrated, then add nitric acid (1+1) to filtrated samples to acidify that to pH < 2 and preserved at 4 °C in the refrigerator for metal analysis.

3.5.3 Procedure

Transfer 100 ml from preserved sample to a 250 ml beaker. Add 2 ml of (1+1) nitric acid and 1 ml of (1+1) hydrochloric acid to the beaker containing the measured volume of sample. Place the beaker on the hot plate for solution evaporation. The beaker should be covered with an elevated watch glass to prevent sample contamination from the fame hood environment.

Reduce the volume of the sample aliquot to about 20 ml by gentle heating at 85 °C. Do not boil to prevent loss of the HCl-H_2O azeotrope.

Allow the beaker to cool, then transfer the sample solution to a 50 ml volumetric flask, make to volume with reagent water, stopper and mix. The sample is now ready for analysis.

Quantification of metals was based upon calibration curves of standard solutions of respective limits.

3.6. Microbiological Parameters

All measurements were carried out according to The Standard Method for the Examination of Water and waste Water. [77]

Collect samples for microbiological examination in nonreactive borosilicate glass or plastic bottles that have been cleansed and rinsed carefully. When the sample is collected, leave ample air space in the bottle (at least 2.5 cm) to facilitate mixing by shaking, before examination.

3.6.1 Total Coliform

9222 B. Standard Total Coliform Membrane Filter Procedure

3.6.1.1 Principle

Red colonies with a metallic sheen produced within 24 h incubation at 35 °C on Endo type medium are considered members of the Coliform Group. This is based on the production of aldehydes from fermentation of lactose.

3.6.2.1 Procedure

An appropriate volume of water sample is filtered through Cellulose ester membrane filter that retains the bacteria present in the sample.

The filter is placed on Endo type selective medium, and the plate is incubated at 35 ± 0.5 °C. Typical Coliform colony has pink to dark- red color with a metallic surface sheen. Use a suitable optical device with 10-15 magnification and cool white fluorescent light source to determine colony counts on membrane filter.

3.6.2 Fecal Coliform

9222 D. Fecal Coliform Membrane Filter Procedure

Fecal coliform method same as total coliform method except (water bath 44.5 °C instead of incubator 35°C).

All apparatuses were obtained from the center laboratory and research of Alexandria Water Company.

3.7 Calibration and quality control

Blank Sample was analyzed at the beginning of each batch to ensure that all reagent interferences and glassware were under control and the method was suitable.

After processing calibration standards a verification standard was measured to determine recovery percentage. Calibration curves were checked by measuring different concentrations of standers in the beginning of the sample analyzed and at its end.

All results were expressed as mean ± SD. The Microsoft Excel spread sheet was used for the graphical presentation and correlation.

3.8 Method Validation and Uncertainty

To value a method, limit of quantification (LOQ), accuracy and precision should be calculated that were performed according to EURACHEM Guide for method validation (1998). [85]

To estimate the accuracy, the recovery percentage % R should be calculated by taking the average of the lowest, the midpoint and the highest standard level.

$$\% \, R = \frac{A}{B} * 100$$

Where:
A= Measured Concentration
B= Prepared Concentration

For example: If the lowest point is 40, the midpoint is 39.9 and the highest standard level is 39.99 and the prepared concentration is, then the average point is:

$$\frac{40 + 39.9 + 39.99}{3} = 39.96$$

Then the recovery percentage can be calculated by substituting the calculated value in the previous equation as:

$$\% \, R = 100 * \frac{39.96}{40} = 99.9\%$$

The LOQ defined as:

The concentration of the analyte which give 10 or more signal to noise ratio.

Relative Standard deviation (% RSD) is calculated as:

$$\% \, RSD = \frac{S}{\bar{A}} * 100$$

Where:
S = Standard deviation of check standard analyses.
$\bar{A}$= Mean of standard analyses.

From these accuracy and precision results, method uncertainty estimated based on EURACHEM Guide (2000). [86]

Table 3.4: LOQ, %RSD and %Recovery for studied parameters

Parameter	unit	LOQ	% RSD	% Recovery
Turbidity	NTU	0.15	0.5	90-110
pH		1	0.16	90-110
EC	µS / cm	2.04	0.045	90-110
Total hardness	mg/l as $CaCO_3$	1	0.48	90-110
Alkalinity, total	mg/l as $CaCO_3$	3.5	0.29	90-110
Chloride	mg/l as Cl^-	1.75	1.43	90-110
sulphate	mg/l asSO_4^{2-}	5	5.9	90-110
phosphate	mg/l asPO_4^{3-}	0.1	6.2	90-110
nitrate	mg/l asNO_3-	0.2	3	90-110
nitrite	mg/l asNO_2-	0.1	7.5	90-110
Silica	mg/l as SiO_2	2	3.4	90-110
Fluoride	mg/l as F^-	0.23	5.9	90-110
Aluminum	mg/L	0.009	6	90-110
Cadmium	mg/L	0.01	9	90-110
Cobalt	mg/L	0.007	6	90-110
Copper	mg/L	0.009	6	90-110
Iron	mg/L	0.006	9	90-110
Manganese	mg/L	0.004	7	90-110
Molybdenum	mg/L	0.005	7	90-110
Nickle	mg/L	0.009	5	90-110
Lead	µg/L	0.01	6	90-110
Zinc	mg/L	0.009	4	90-110