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An integrating geospatial technology with microbiology in isolating and characterizing selenite-reducing bacteria from two mangrove areas along the Red Sea, Egypt

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ABSTRACT

The wide anthropogenic use of selenium compounds represents the major source of selenium pollution worldwide causing environmental problems and health concerns. The current study aimed at isolating sixteen bacterial strains, capable of transforming toxic SeO32- to non-toxic elemental selenium, from sediment, water, and mangrove leaves in two mangrove study areas (17 km south of Safaga and 35 km north of Quseir). Geospatial technology was used to detect the location of the mangrove areas. Microbes were isolated using membrane filter technique and direct bacterial plating on salt peptone (SP) agar supplemented with 5mM sodium selenite. Morphological, biochemical, Scanning Electron Microscope (SEM), and Transmission Electron Microscope (TEM) analysis of two bacterial isolates were identified using 16S rRNA gene sequencing. Results showed that the red colonies of sixteen isolates signify the reduction of selenite to red elemental selenium. Phylogenetic analysis proved that the two tested isolates affiliated to (*Cobetia amphilecti* and *Vibrio alginolyticus*) with accession numbers of MN099349 and MN099350 respectively. SEM and TEM analysis confirmed the presence of globular particles of insoluble selenium outside and inside the cells of the two selected bacteria. The higher bacterial count in south Safaga compared with wadi Abu Hamrah ensured by drainage pattern which shows that there are main drains that pour directly in mangrove stand especially in south Safaga stand. In conclusion, bacterial isolates can transform toxic selenite to non-toxic red elemental selenium which could be further used for bioremediation of contaminated locations. To our knowledge, these selenite reducing bacteria were not detected and /or isolated from the Egyptian Red Sea coast before as well as both identified strains are novel and well-characterized bacterial aerobic selenite reductase.

INTRODUCTION

In 1817, Berzelius discovered Selenium (Se) (**Montes, 2012**) in several natural and anthropogenic sources (Savard et al., 2009). Selenium occurs in four states; [SeO_4^{-2}], selenite [SeO_3^{-2}], selenide (Se^{-2}), and elemental selenium [Se^0] (**Rehan *et al.*, 2018**). Though humans require trace quantities of selenium for cellular functions, elemental

selenium and selenite oxyanions are lethal in even small dosages (**Fordyce, 2005**). Due to the human activities, selenium contamination is considered to be the main environmental pollution found almost everywhere (soil, water) (**David *et al.*, 2015**) Selenium has widespread uses in industrial and agricultural processes, which are responsible for its high toxic levels in the environment (**Pierru *et al.*, 2006**).

Microorganisms have been shown to play major role in the reduction of different Se oxyanions **(Nancharaiah and Lens, 2015)**. Moreover, selenium bio-reduction mechanism has attracted attention to be used for remediation of selenium oxyanions-contaminated sites (**Gonzalez-Gil *et al.*, 2016**).

Both halophilic and halotolerant microorganisms are suitable for biomineralization in saline soils because they have several adaptation mechanisms **(Ventosa, 2004),** and can produce many functional compounds as extracellular, hydrolytic enzymes that are used in biomedical science and chemical industries **(Chen and Liu., 2013)**.They are distributed widely in hypersaline environments, all over the phyla and the orders of bacterial domain (**Hedi *et al.*, 2009**), and classified into three groups: slightly halophiles, moderately halophiles and extremely halophiles with optimum NaCl concentration of 2– 5, 5–20, and 20–30%, respectively according to their response to NaCl (**Kerkar, 2004**).

Mangrove ecosystems occur in the transitional area between marine and terrestrial environments **(Thi *et al.*, 2014)** that are rich in marine microorganisms and unique halophilic flora. The ecosystem importance originates from its protection of coastal zones from corrosion and its contribution of recycling nutrients in wetland environments **(Twilley *et al.*, 1992)**. In Egypt, *Avicennia marina* (black mangrove) and *Rhizophora mucronata* (red mangrove) (**Basheer *et al.*, 2019)** inhabit many sites along the Sinai Peninsula and the Red Sea shoreline.

Remote sensing used for mapping mangroves (including biomass and carbon stocks) saving time and money and at a wider scale than field measurements **(Pham *et al.*, 2019).**

The current study aimed at applying geospatial technology in detecting the mangrove ecosystem areas along the Red Sea from which halophilic microorganisms capable of transforming toxic SeO32- to non-toxic elemental selenium were isolated.

MATERIALS AND METHODS

Study area

Two mangrove areas along the Red Sea were selected for the current study (Figure 1). The first one is located 17 Km south of Safaga (26° 37' 01.04"N and 34° 00' 39.96" E) whereas the second one is located 35 km north of Quseir (Wadi Abu Hamrah) (26° 23' 58.04"N and 34° 07' 01.19" E).

Sampling

The study collected samples of water, sediment, and mangrove leaves from the two mangrove study areas during November 2018. From each area, two samples of water (500 ml; depth 20 cm) were collected and then mixed thoroughly to make one composite sample. Moreover, the study collected two groups of leaves for microbial variation detection. The first group was five leaves from different plants and the second from different heights of one plant. Finally, two sediment samples (depth 40 cm) were collected using a plastic coring tube of 25 cm in diameter and 10 cm in length. All

samples were immediately placed in ice sterilized bags and brought to the laboratory where they were kept in a refrigerator at 4°C until they were processed and analyzed in the laboratory within 24-48h.

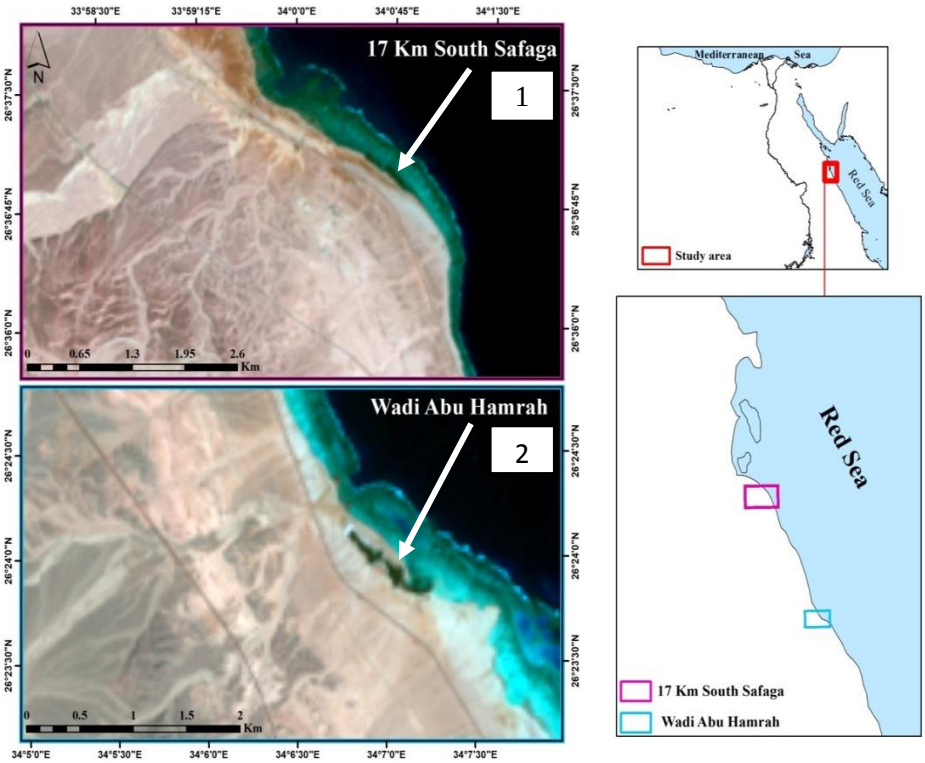

Fig. 1: The two study areas along the Red Sea, location 1 (17Km Sough Safaga) and location 2 (Wadi Abou Hamrah)

Isolation and purification of selenium reducing bacteria:

The bacteria were isolated using direct plating technique method in salt peptone agar medium (SP; g/l), that contained KCl 0.5; $MgSO_4.7H_2O$, 1; $CaCl_2.3H_2O$, 0.7 ; $MnCl.4H_2O$, 0.05; Peptone, 10; Yeast extract, 10; Agar, 15 **(Williams *et al*., 1989)** and nutrient agar medium (NA; g/l) that contained Peptone, 5.0; NaCl, 3.0; beef extract, 3.0; agar , 18 (**Mishra *et al* ., 2011**) with 1-5% NaCl (w/v) at pH 7.0±2. Both media were supplemented with 5mM selenite then incubated at 30 ^{0}C under aerobic conditions for 72 h.

Orange, pink and red colonies, indicating that the isolates could reduce selenite to elemental selenium; they were re-streaked on nutrient agar and salt peptone media without selenite to confirm that the color was not due to pigmentation. The pure cultures were isolated and maintained on selenite supplemented plates until use (**Mishra *et al*., 2011**).

Enumeration of metalloid-resistant bacteria

For enumeration of the isolated selenite reducing bacteria, one gram of the collected leaves of *Avicennia marina* and / or sediment samples were transferred to 100 ml sterile saline solution in 250 ml Erlenmeyer's flask. The flask, contexts were mixed thoroughly in the shaker for 5 minutes then filtered using Millipore filtration technique (**APHA, 1995**). Sea water samples were filtered using the same filtration technique. The Millipore filter paper was cultured on the surface of SP and/ or NA agar plates. The plates were incubated at 30°C for 72 hr. for counting the colony forming unit (CFU) and used to calculate counts / g. of the leaves and sediment and per ml of water samples. Two media used for the enumeration; salt peptone agar medium (SP), **(Williams *et al.*, 1989)**. and nutrient agar medium (NA) (**Mishra *et al.*, 2011**) supplemented with 1 and/ or 5% NaCl and with and without 5 mM sodium selenite.

Characterization of the bacterial isolates:

Identification of Na_2SeO_3 tolerant bacteria was done based on morphological characterization and biochemical test following **Williams *et al.* (1989)**.

Factors influencing selenite reducing bacterial growth

To determine the optimum temperature and pH for the growth of the strain, the cultures were incubated at a temperature range of 15-50 °C with intervals of 5 °C and pH values of 5-10.5. Also, growth of the strains were evaluated at different percentage of NaCl values (0-30 % NaCl) by using Luria Bertani broth medium (LB; g/l), that contained bacteriological peptone, 10; yeast extract, 5.0; NaCl, 5.0 supplemented with 5 mM selenite under aerobic conditions **(Khalilian *et al.*, 2015).** pH values were adjusted by 0.1and 1N HCl, and 0.1 and 1 N NaOH**.**

Phylogenetic analysis

The two selected isolates (one moderate halophile and one halotolerant) were identified using 16S rRNA sequencing. The obtained gene sequence was searched in Basic Local Alignment Search Tool (BLAST) and online interactive Tree Of Life (iTOL) tool was used for phylogenetic tree construction (**Letunic and Bork, 2006**). The sequence was submitted to National Center for Biotechnology Information (NCBI) and the accession number was obtained.

Transmission electron microscopy (TEM) and scanning electron microscopy (SEM) analysis to determine selenium particle within isolate cell

TEM and SEM observations were carried out according to **Zhang *et al.* (2018)** to study the location of Se reduced by the bacterial strains (*Cobetia amphilecti* and *Vibrio alginolyticus*).

Analytical analysis

Determination Total Dissolved Selenium (TDSe)

Total dissolved selenium (TDSe) was determined in the three samples collected (sediment, water and leaves) from mangrove ecosystem, Red Sea, using spectroscopic method according to the method of **APHA (2017**).

Statistical analysis

The study computed correlation analysis with p-value < 0.05 using R programming language. The multivariate clustering analysis was done using ClustVis online tool (**Metsalu and Vilo, 2015**).

Digital elevation model

According to **Sarika (2005)**, rainfall is the major source of trace metals into the mangrove. Hence, it is important to extract the drainage pattern of the two study areas to detect the area that may receive the higher content of selenium. Consequently, the study used Digital Elevation Model (DEM) for deriving the drainage network along South Safaga and Wadi Abu Hamrah. DEM, the digital representation of the earth surface terrain **(Balasubramanian, 2017)**, was obtained from Shuttle Radar Topography Mission (SRTM) data available at the United States Geological Survey website (USGS), with 30 m spatial resolution. Different processes were carried out for the extraction of drains including clipping the boundary of the study area, creating a flow direction raster by using flow direction and flow accumulation, deriving of stream, and stream order using ArcGIS 10.1.

RESULTS AND DISCUSSION

The current study isolated two novel gram-negative, selenite-reducing bacteria, *Cobetia amphilect*i and *Vibrio alginolyticus*. These bacteria converted selenite to elemental selenium (Se^0; red colored colonies).

Isolation and purification of selenite reducing bacteria

Isolation of the moderately halophiles resistant to selenite was performed using a culture medium maintaining rapid bacterial growth. Altogether 16 bacterial isolates were isolated from sediment, water and leaves samples collected from the mangrove forest, Red Sea , Egypt, using SPA and NA media supplemented with and/ or 5mM sodium. Appearance of reddish color after 24-48 h indicated that selenite reducing bacteria has been enriched which converted soluble selenite (Na_2SeO_3) into red colored elemental selenium (Se^0) (A) when compared to control plate which failed to show a change in color when incubated under same conditions (B) (Fig. 2).

Reduction of Selenite:

Microorganisms can carry out the conversion of SeO_3^{2-} to Se^0 via several mechanisms **(Kessi, 2006)**. SeO_3^{2-} reduction can be catalyzed by reductases, including the periplasmic nitrite reductase, sulfite reductase, and dimethyl sulfoxide (DMSO) reductase **(Afkar *et al.,* 2003).** Several thiol mediated reactions have also been observed to reduce selenite to elemental selenium **(Nancharaiah and Lens, 2015)**.

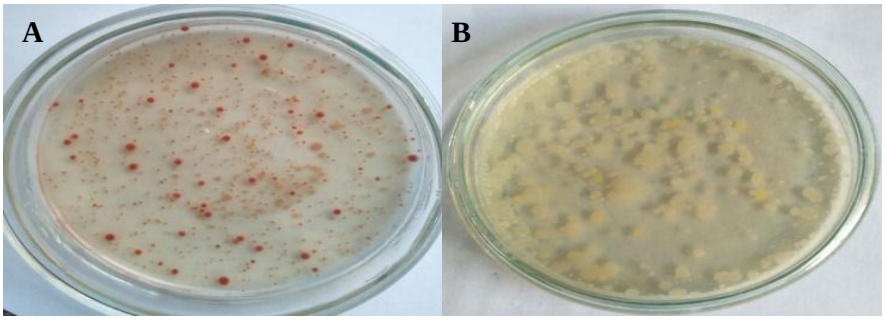

Fig.(2): (A) The formation of red colonies in presence of sodium selenite, (B) colorless colonies in absence of sodium selenite

Enumeration of bacteria

Culturable bacterial populations of leaves, water, and sediment samples of Avicennia marina ecosystem were enumerated. The results in table (1) indicated generally high counts for halophilic bacteria in location 1 comparing with those from location 2 of the mangrove area. This high count might be attributed to the high nitrogen and carbohydrate contents of the leaves of *Avicennia marina* plant from the same site (**Khalaf, 2002)**.

Table (1): The total bacterial counts in water (ml), sediment (gm) and leaf of dry weight of Avicennia marina at two sites. Both count in SP containing 5Mm sodium selenite and 5% NaCl

Samples	location 1	location 2
Sediment	**117**	**6**
Water	**100**	**68**
Leaves	**70**	**9**

The correlation analysis between count of water, sediment, and leaves revealed a high positive correlation (direct proportion, dark blue) between leaves and sediment count and weak positive (faint blue) correlation between both leaves and sediment with water at p-value of 0.05 (Figure 3a). The correlation network revealed that leaves stand as a key link between water and sediment (Figure 3b).

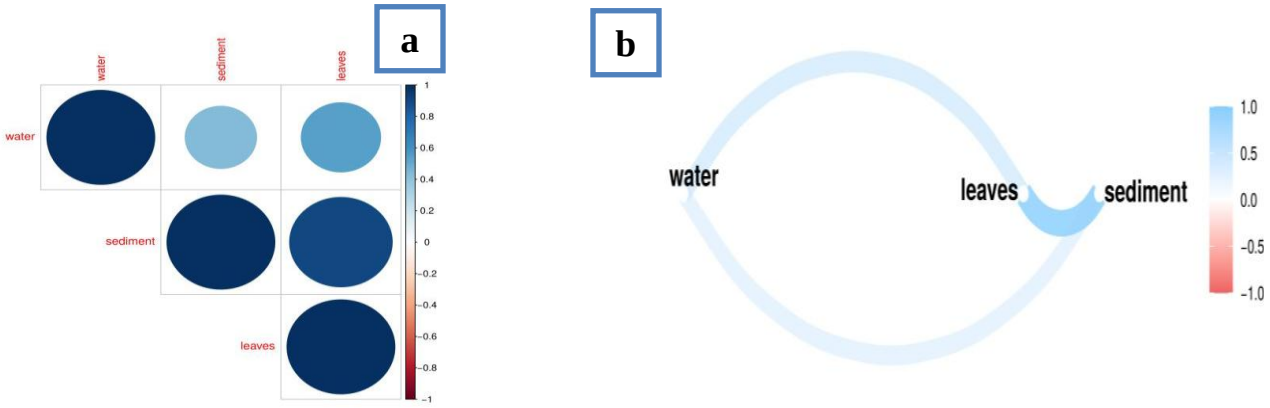

Figure 3: (a) The correlation analysis between water, sediment and leaves count at p-value of 0.05. (b) Correlation network between water, leaves and sediment at p-value of 0.05.

The correlation between bacterial count (Figures 4c and 4d) showed that high positive correlation (direct proportion) between Wadi Abu Hamrah- salt peptone agar medium with Se (WH-SPA-with-Se) and 17Km south Safaga- Nutrient Agar medium-with Se (k17SS-NA-with-Se), 17Km south Safaga-salt peptone agar medium with Se (k17SS-SPA-with-Se) and Wadi Abu Hamrah- salt peptone agar medium with Se (WH-SPA-with-Se), and 17Km south Safaga-Nutrient Agar medium-without Se (k17SS-NA-without-Se) and Wadi Abu Hamrah-Nutrient Agar medium-without Se (WH-NA-without-Se). While negative correlation (reverse fit, red color) can be seen between k17SS-SPA-without-Se and k17SS-SPA-with-Se. In this regard, the correlation network showed that, WH-SPA-without-Se, k17SS-SPA-without-Se and WH-SPA-without-Se formed the core of statistical correlation network among other counts. Where any change in these counts could influence changes in the other count.

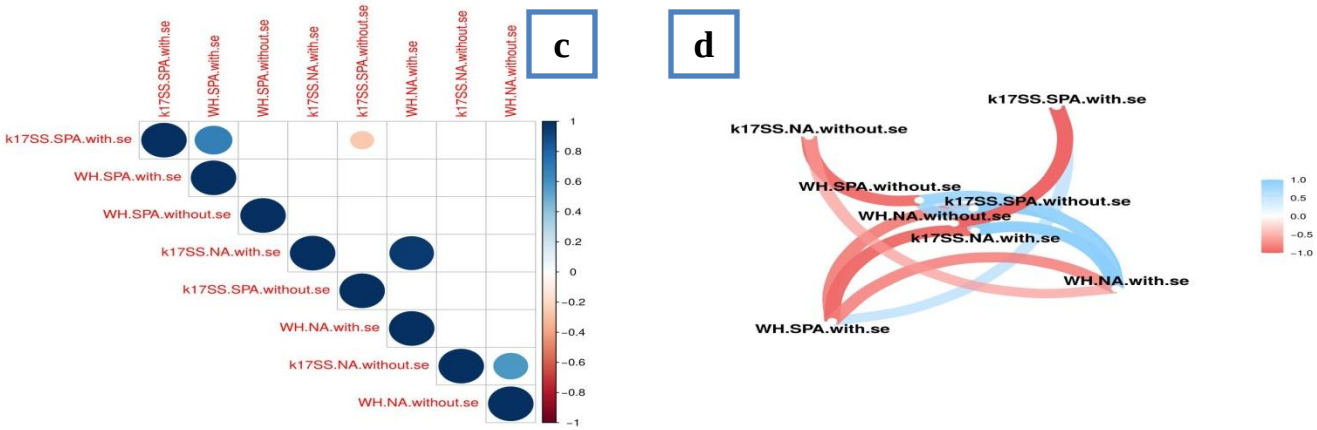

Figure (4): (c) The correlation analysis between bacterial count at p-value of 0.05 and (d) count correlation network at p-value of 0.05. (WH: Wadi Abou Hamrah; k17SS: 17Km Sough Safaga)

Analytical analysis

Table (2) displayed different values for Se concentration in the two study areas. Se concentration in the first location (17 Km Sough Safaga) was greater than in the second one (Wadi Abou Hamarah) which explains the difference in counts in both locations as the first location contained higher count than the second. The abundance and activity of bacteria in mangrove ecosystems was attributed to leaf exudates from leaf litter on water (**Ashour *et al.*, 2011**).

Table (2): values in both sites (location 1; 17 Km south of Safaga and location 2; 35 km north of Quseir at Wadi Abou Hamarah)

Location	Se	Location	Se
17 Km South Safaga	**mg/ Kg**	**Wadi Abu Hamrah**	**mg/ Kg**
Sediment	656.61	**Sediment**	538.73
Water	17.90	**Water**	11.97
Plant	419.78	**Plant**	386.54

Characterization of the bacterial isolates:

In the present study, sixteen bacterial isolates were selected from twenty-six isolates from mangrove water, sediment, and mangrove leaves ecosystem. According to phenotypic characterizations of the bacteria isolated, the entire sixteen isolates were shown to be Gram-negative, spore former, motile, and aerobic rod (Table 3). The growth characterization was performed to evaluate the variation of salt concentration in the media that greatly affects the growth of bacterial strains. It was essential to see the effect of salt concentration on selenite (Se (IV)) reduction **(Mishra *et al.*, 2011)** using SP medium containing 5mM sodium selenite.

The bacterial response to different NaCl concentration showed that the selenite reduction of the strains was marginally affected up to 15% NaCl and thereafter sharply declined on further increase of salt concentration. For all 16 isolates, NaCl optimum was ranged from 1-13%, most isolates were classified as moderate halophiles and as extreme halotolerant while two of the isolates as haloversatile. **Borg *et al.* (2009)** reported that the selenite reduction virtually stopped at 20% of salt in the media. The reduced activity is primarily due to detrimental effect of the ionic stress on the bacteria at higher salt concentrations which inhibits bacterial growth (Table 4)**.**

pH is an important parameter that influences the microbial reduction of Se(IV). It was observed that the reduction of Se (IV) (5 mM) increased with an increase of pH and attained a maxima in between pH 7.0 and 9.0. Further increase of pH showed a marginal effect on Se (IV) reduction as shown in (Table 4). The bacterial response to different pH

values were ranged from 5 to 10. It may be noted that the reduction of Se (IV) to Se^0 at neutral to alkaline pH is similar to the observation of **Lortie *et al.*, (1992)**.

Temperature is another factor that affects the microbial reduction of Se (IV). The bacterial response to different temperature values found that most strains are mesophilic bacteria that preferred temperature between 30 and 37 ^{0}C as shown in (table 4).

As a result of microbial reduction of selenium oxyanions the red elemental selenium particles with either crystalline or amorphous structure were produced (**Losi and Frankenberger, 1997 and 1998).** Most previous studies on microbial Se (IV) reduction to elemental selenium reported that the product of Se (IV) reduction was spherical selenium nanoparticles (**Xu *et al.*, 2018**).

Table (3): Morphological and main biochemical characteristics of the bacterial isolates

Characteristics	**Isolated strains**															
	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**
Cell shape	**Rod bacilli**															
Gram reaction	-															
Colony Color with selenite	Red	Red	Red-Orange	Red	Dark red	Red				Faint red	Red	Faint red	red	dark red	Red	Faint red
Endospore-forming	+															
Motility	+											-	+			
Catalase	+															
Blood Haemolysis	-				+	-	+	-					+	-		
Gelatin	-	-	+	-	+	-	+	-								
Starch	-	-	+	-	+	-	+	-					+	-		

Table (4): Salt, temperature and pH tolerance values for the sixteen pure bacterial isolates grown in salt peptone broth medium and incubated for 72 hr. at 35°C in case of salt and pH (**Min:** Minimum **O:** Optimum **Max:** Maximum)

Isolate No.	Salt tolerance				Heat toleransce				pH tolerance			
	Range (%)			Description	Range (^{o}c)			Description	Range			Description
	Min	O	Max		Min	O	Max		Min	O	Max	
1	1	5	25	Extreme Halotolerant	25	30	45	Mesophile	5	6.5	10	Neutrophilic
2	1	7	25	Moderate Halophile	25	30	45	Mesophile	5	8	10	Alkalophilic
3	1	1	25	Halotolerant	25	37	45	Mesophile	5	10	10	Alkalophilic
4	1	5	25	Extreme Halotolerant	25	37	45	Mesophile	5	9.5	10	Alkalophilic
5	1	5	25	Extreme Halotolerant	25	37	45	Mesophile	5	7.5	10	Alkalophilic
6	1	7	25	Moderate Halophile	25	37	45	Mesophile	5	8	10	Alkalophilic
7	1	3	25	Haloverstile	25	30	45	Mesophile	5	7.5	10	Alkalophilic
8	1	5	25	Extreme Halotolerant	25	30	45	Mesophile	5	9.5	10	Alkalophilic
9	1	5	25	Extreme Halotolerant	25	37	45	Mesophile	5	7	10	Neutrophilic
10	1	13	25	Moderate Halophile	25	30	45	Mesophile	5	8.5	10	Alkalophilic
11	1	7	25	Moderate Halophile	25	37	45	Mesophile	5	7	10	Neutrophilic
12	1	7	25	Moderate Halophile	25	30	45	Mesophile	5	7.5	10	Neutrophilic
13	1	3	25	Haloverstile	25	30	45	Mesophile	5	8	10	Alkalophilic
14	1	5	25	Extreme Halotolerant	25	37	45	Mesophile	5	7.5	10	Alkalophilic
15	1	5	25	Extreme Halotolerant	25	30	45	Mesophile	5	7.5	10	Alkalophilic
16	1	10	25	Moderate Halophile	25	30	45	Mesophile	5	8.5	10	Alkalophilic

Transmission electron microscopy (TEM) and scanning electron microscopy (SEM) analysis to determine selenium particle within isolate cell

Figure (5) shows the characterized reduced product (elemental selenium Se^0) associated with the cell, by SEM and TEM showed in (Fig.5). SEM and TEM microphotographs of the elemental selenium revealed globular both outside and inside the bacterial cells in the media. SEM analysis revealed the formation of Se^0 nanoparticles around the elongated bacterial cell. Further analysis by TEM revealed the formation of nanospheres through the reduction of Se (IV) by different bacterial strains. Also, **Yee *et al.* (2007)** reported similar observation indicating the ability of the bacteria to accumulate Se around and inside cells.

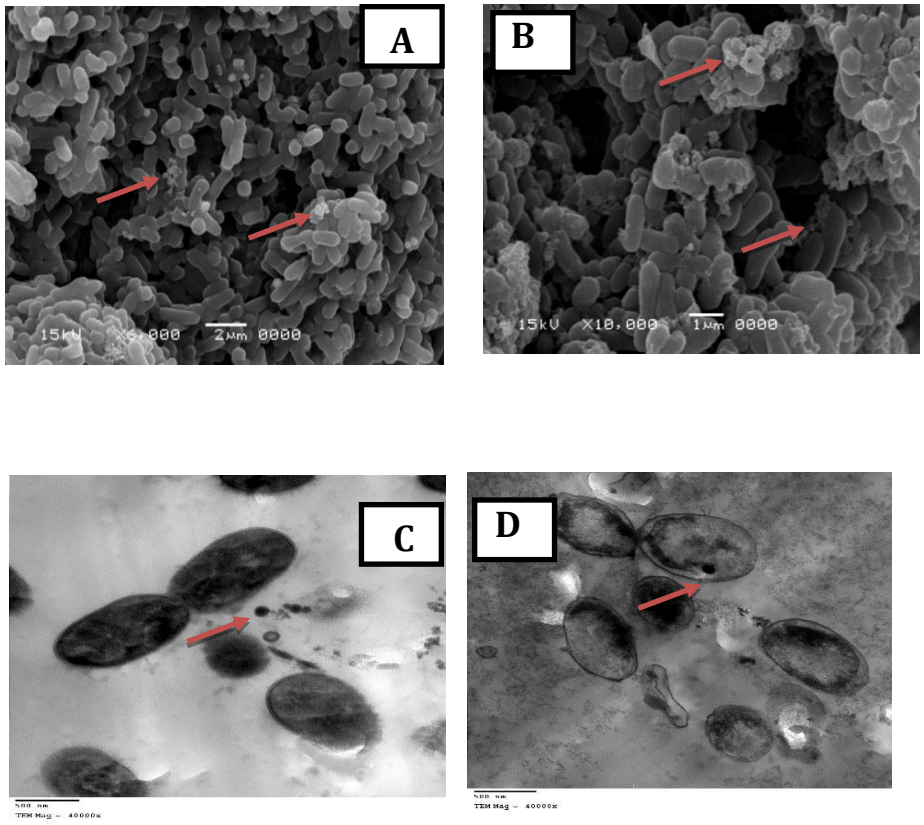

Figure (5): TEM and SEM micrographs of *Cobetia amphilecti* (A&C) and *Vibrio alginolyticus* (B &D) exposed to5 mM selenite obtained after 72h of growth. The arrows indicate the presence of spherical selenium particales outside and inside the cells.

For 16srRNA sequences of the two selected isolates

The NCBI-Blast analysis was successfully used to identify strains 2 and 3.16S DNA sequences annotation and their corresponding species. The NCBI-Blast tool assigned strain 2 to *Cobetia amphilecti* MSN1517 with similarity score 97.5 % and sequence coverage of 96% and strain 3 as Vibrio alginolyticus MSN1517 with similarity score 95 % and sequence coverage of 95% (Fig. 6) with accession numbers of MN099349 and MN099350 for strains 2 and 3, respectively.

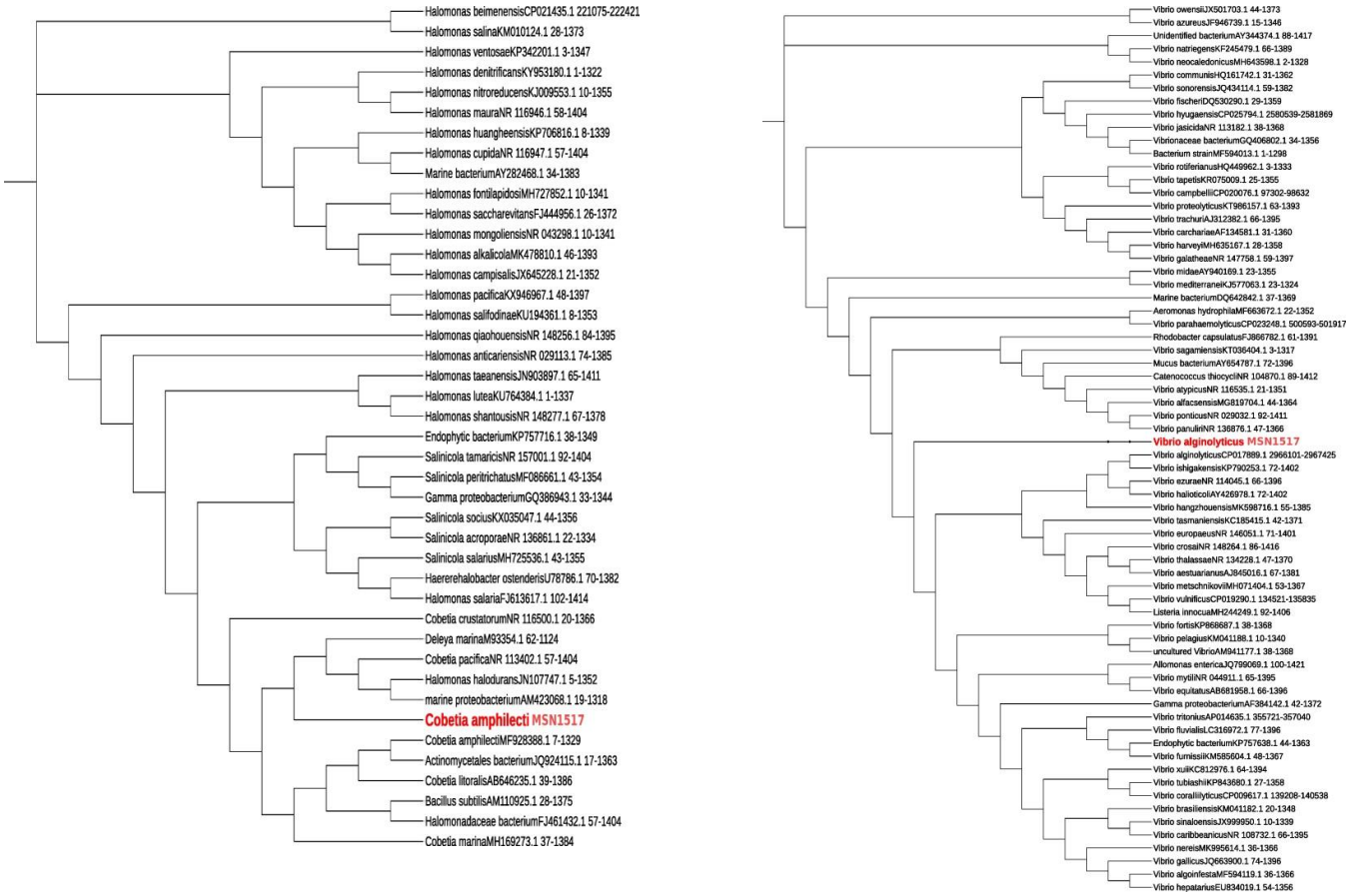

Figure (6): the phylogenetic tree constructed using 16S DNA sequences, where the isolates MSN1517 obtained form was *Cobetia amphilecti* and *Vibrio alginolyticus* highlighted.

Digital elevation model

DEM help in the extraction of the drainage network at the two study areas. Drainage pattern showing that there are different drains that pour directly in mangrove stands in both study areas especially in South Safaga stand which connected with main drain that might be a source of many elements and metals including selenium in mangrove ecosystem (Figs. 7a and b).

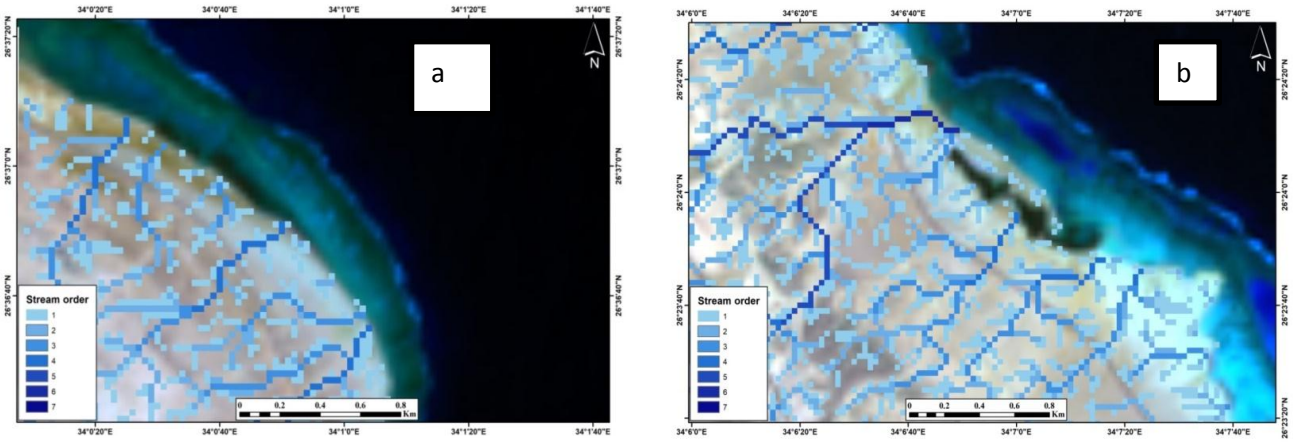

Figure (7): (a) Drainage network at South Safaga stand, (b): at Wadi Abu Hamrah stand

CONCLUSION

Selenium exists in the environment due to natural and anthropogenic activities that increased the contamination risk to the ecosystem. Conventional chemical methods for removing toxic oxyanions are expensive and require high energy or large quantities of chemical reagents, while microbial reduction of these toxic oxyanions is cost effective and supports green technology. In the current study, sixteen of selenite reducing microbial isolates showed potential to bioremediate the toxic form of selenium into less toxic form in a cost effective way. Two of these isolates well characterized halotolerant (*Cobetia amphilecti* and *Vibrio alginolyticus*) isolated from saline mangrove habitat, were found capable of reducing Selenite to elemental selenium even in the presence of high salt concentrations. Under optimized set of conditions (at 30-37^{0}C, initial pH 9.5-8) almost complete reduction of Se (IV) 5 mM was achieved within 48-72 h incubation. This selenite reducing bacteria were not detected and /or isolated from Red Sea Egyptian coast before, and the both identified strains are novel and well-characterized bacterial aerobic selenite reductase.

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الملخص العربى

دمج التكنولوجيا الجغرافية المكانية مع علم الأحياء الدقيقة في عزل ووصف البكتريا المختزلة للسيلينيت من منطقتين من المانجروف على طول البحر الأحمر، مصر.

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1- قسم النبات- كليه البنات للاداب والعلوم والتربية- جامعة عين شمس

2- قسم علوم البحار- الهيئة القومية للإستشعار من بعد وعلوم الفضاء

يمثل الاستخدام الواسع لمركبات السيلينيوم المصدر الرئيسي للتلوث بالسيلينيوم في جميع أنحاء العالم مسببا مشاكل بيئية و صحية. والهدف من هذه الدراسة هو عزل الكائنات الحية القادرة علي تحويل السيلينيوم من الصورة السامة للصورة غير السامة من منطقة المانجروف بالبحر الأحمر (17 كم جنوب سفاجا و وادي أبو حمرة). تم عزل ستة عشر سلالات بكتيرية من الرواسب والمياه وأوراق المانجروف. وقد تم اكتشاف مناطق المواقع بواسطة تقنيات الاستشعار عن بعد ونظم المعلومات الجغرافية. تم عزل الميكروبات باستخدام تقنية تصفية الغشاء والطلاء البكتيري المباشر على بيئة ببتون ملح الأجار مضاف اليها الصوديوم سيلينيت 5mM.تم عمل تحاليل مورفولوجية ، والكيميائية الحيوية وكذلك المسح المجهر الإلكتروني (SEM) والمجهر الإلكتروني للإرسال (TEM) و تسلسل الجينات 16S rRNA لاثنين من هذة العزلات.أوضحت النتائج أن البكتريا المعزولة قادرة علي اختزال الصوديوم سيلينيت الي عنصر السيلينيوم غير السام ودلالة ذلك هو تكوين المستعمرات البكتيرية حمراء اللون.وتم تعريف العزلات المختارة كـ *Cobetia amphilecti* و*Vibrio alginolyticu* مع عدد من الانضمام MN099349و MN099350.

وأكدت تحاليل SEM وTEM علي تكوين أجسام كروية من عنصر السيلينيوم غير قابل للذوبان داخل وخارج الخلايا. ارتفاع عدد البكتريا في منطقة الكيلو 17 مقارنة بوادي أبو حمرة الذي يضمنه نمط الصرف الذي يظهر أن هناك مصارف رئيسية تصب مباشرة في منطقة المانجروف خاصة في الكيلو 17 جنوب سفاجا. في النهاية تم عزل بكتريا قادرة علي تحويل السيلينيت السام الي عنصر السيلينيوم الأحمر غير السام. والتي يمكن استخدامها كذلك من أجل الإصلاح البيولوجي للمواقع الملوثة.علي حد علمنا انه لم يتم عزل هذة البكتريا من ساحل البحر الاحمر بمصر من قبل و تعتبر البكتريا المعرفة جينيا جديدة.