The occurrence of heavy metals in Qarun Lake and their influence on microbial biodiversity

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ABSTRACT

Qarun Lake represents an important part of Egyptian history; it is surrounded by many wild and urban environments. In this study, water samples were collected from ten different regions, during different seasons in (2020, and 2021). The water samples were analyzed for their physical properties and the concentrations of the possible heavy metals. One Actinobacteria isolate was identified as *Streptomyces noursei* based on the partial sequencing of 16S rDNA and, six fungal isolates were fully identified as *Aspergillus niger, Aspergillus versicolor, Aspergillus terreus, Aspergillus flavus, Aspergillus fumigatus, and Fusarium oxysporum* based on the partial sequencing of 18S rDNA. Fortunately, *S. noursei* showed antifungal efficacy against three of the tested fungal isolates, with the inhibition zones (18, 16, and 19 mm) against *A. niger, A. Fumigatus,* and *F. oxysporum* respectively. Different heavy metals were detected in stations 7, 8, 9, and 10. The highest values of Pb, Cu, Zn, Ni, Cd, and Cr were 35.3, 12.5, 56.6, 12.5, 3.21, and 21.5 respectively. The total viable count of fungi (TVFC) varied between $0.7x10^2$ CFU/mL at site 7 and $5.5x10^2$ CFU/mL at site 1. We can conclude that Qarun Lake contains pathogenic fungi as well as actinomycetes with high antifungal activity, and that these microorganisms are closely related to the distribution of heavy metals in Qarun Lake water.

Keywords: Qarun Lake; microbial biodiversity; actinobacteria; heavy metals; antifungal activity.

INTRODUCTION

To comprehend the consequences of heavy metals on water and living things, it is crucial to investigate their presence in aquatic ecosystems. Heavy metals have received a lot of attention in recent years in the study of aquatic environments due to their possible harmful effects, persistence, and bioaccumulation issues (Censi et al., 2006; Carr and Neary, 2008). Lakes are perfect habitats for diverse microbial communities (Tahoun, 2023). Qarun Lake is one of the oldest lakes in the world. It began as a freshwater lake and nowadays has changed to become a salt lake owing to climate changes and anthropogenic activities. The environment of Qarun Lake has been subjected to many changes since the beginning of the19th century that have led to drastic variations in its elements. In addition, it was subjected to various exploitation regimes (Abdelmageed et al. 2022). Furthermore, the desert of El-Fayoum is abundant in a number of natural resources, including subsurface water, minerals, and stone deposits. This is in addition to the area's abundant animals and biodiversity. A number of native birds, ducks, and migrating birds like gulls are also thought to call the area home (Mohamed and El-Raey, 2019). The lake is crucial for the Egyptian fisheries because it produces overall catches and a variety of species that are economically beneficial (Gohar, 2022). Although being a protected area since 1989, Qarun Lake has not been shielded from contamination. Qarun Lake is a storehouse of many fungi that have the ability to resist salinity and pollution. Qarun Lake suffered drastic chemical, natural and microbial variations through the previous years. As a result, its salinity gradually rises, having a significant impact on Qarun Lake's biota. Also, the nitrogen load from the overabundance of water used for agriculture contributes to the buildup of organic compounds in the lake water. (Sabae and Ali 2004). The unique source of lake water inflow is water from drainage and the evaporation of freshwater is the unique source of outflow water from lake, this increases salinity, which affected the ecosystem and reduced fish production (Abdelbaki 2021). About 70% of the drainage water of Al-Fayoum city (i.e., the drainage of agriculture, sewage drainage, and industrial drainage) is discharged in Qarun Lake. Observing the lake may provide early warning indicators of ecosystem depletion due to, for instance, pollutant inputs, nutrient additions, or reductions in the pollutants coming from the drain of El-Wadi by implementing automated filters like in El-Batts drain, this filter will reduce salts in the lake and increase salt extraction factory numbers to reduce salt intensity As an EMISAL factory (Abdelbaki 2021). The main goals of this study are to evaluate fungal distribution in Qarun Lake water and follow up on its biodiversity

throughout the seasons, study physical properties and heavy metal contamination in Qarun Lake water and their effect on fungal dispersal through Qarun Lake, and, most importantly, biologically control the fungi distributed through Qarun Lake over the years because this strongly affects the fish stocks and the environmental balance.

MATERIALS AND METHODS

Study area

In the current study, water samples were collected from Qarun Lake during the winter of 2020 and the summer of 2021. Qarun Lake (Figure 1), placed in the north of Al-Fayoum City and distant 80 km southwest of Cairo. Al-Fayoum is one of Egypt's natural developments and a resource that has helped human culture for some 8,000 years. Water samples were collected in subsurface (25 cm) at ten stations to cover the largest lake area. A Ruttner water specimens bottle of 1 L volume was used to collect the samples that were kept in plastic bottles for chemical analysis, and microbial specimen were collected using sanitized glass bottles and then brought to the laboratory in an ice-filled insulated container during transport.

Sample collection

In this study; samples were collected at different seasons from December 2020 to Jon 2021, 10 samples in each season. Water samples were collected in sterilized screw-capped bottles by Austin (1988), and then transmitted to a sterile polythene bag. Sampling took place at 10 different stations from Qarun Lake to cover the largest lake area, as the following in table (1).

Isolation of actinobacteria

For actinomycetes isolation, serial dilutions were made up to 10^{-4} and spread on starch nitrate agar (SNA) plates. The media were inoculated with one ml of diluted samples. Plates were incubated at 28 ^{o}C for 5-10 days. After incubation, colonies with powdery texture and folded and branching filaments without or with aerial mycelia were subcultured on SNA slants that were preserved at 4 ^{o}C (Sharma and Jagtap 2022).

Isolation of fungi

Serial dilutions were made up to 10^{-4}. Fungal isolates were isolated from water samples by spreading method on surface plates of Potato dextrose agar (PDA) media, and then the plates were incubated aerobically at 28°C for 4 -7 days (Jäger et al., 2022).

Identification of actinobacteria and fungi

Phenotype characterization

After 3 days of incubation, isolates were examined under a light microscope to investigate the microscopic characters of isolates. Morphological characterization was based on classical macroscopic techniques of color, form, shape, margin, and elevation of the pure colonies. Cultural characteristics of actinomycetes isolates were examined by visible observation of a 7-day old culture that was grown on SNA medium. Morphology and sporulation of colonies were investigated under a light microscope using the cover slip method (Williams et al., 1989). Substrate and aerial mycelium, branching, and the nature of the colony were noted after 5–10-day intervals. After growth on medium and on cover slips, the cover slips were withdrawn and mounted on the glass slide with one drop of methylene blue (0.3 g in 10 ml distilled water). The cover slips were fixed with Canada balsam and observed under a light microscope.

Molecular identification

DNA samples were isolated from tested fungal strains through the Easy Pure Genomic DNA Extraction Kit, and DNA samples were measured for their purity and appropriate concentration through Nano-Drop. Four DNA samples of tested fungi were subjected to PCR reactions at Sigma Company.A PCR reaction was achieved for 18s rDNA gene amplification of the fungal isolates by using two universal fungal primers, FW 5-ATGGGCAAGGCACCAAATAA-3 and RW 5-TGGAAATGGATC CAAGAATG-3, and 16s rDNA gene amplification of the actinomycetes isolate using the universal primers. F 5′- AGA GTT TGA TCC TGG CTC AG-3′ and R 5′-GGT TAC CTT GTT ACG ACT T-3. PCR conditions were adjusted as follows : 6 µl of template DNA solution and 8.5 µl of master mix, which includes dNTPs mix, MgCl2, Taq polymerase, and PCR buffer. Primers were added separately after preparation from lyophilized stock (1 µmol/l of each primer). PCR conditions were adjusted at denaturation step at 92^{o}C, annealing step at 56^{o}C and extension one at 72^{o}C.The number of PCR cycles was 36 cycles using thermo cycler PCR. The amplified DNA product along with a DNA marker [gene ruler 100 bp DNA ladder (SM0241)] was separated by electrophoresis using 1% agarose gel. The gel was stained with ethidium bromide, and the banding profile

was recorded using the UV-gel documentation system. The amplified DNA product was purified and sequenced. Four sequences were analysed and tested against the most closed sequences on GenBank NCBI through BLST, and phylogenetic trees were designed through MEGA 7 software. (Hentschel et al. 2001).

Chemical, Physical and microbial characterization

Physical, chemical, and microbial studies of water samples were done according to the methods described by the American Public Health Association (APHA) (Beutler et al. 2014) in Fayoum Drinking Water and Sanitation Company. Water temperature (°C) measured by S/N:98608 instrument temperature and (pH) meter, pH measured by pH meter (Khalil et al. 2015). The total viable fungal count (TVFC) measured by the pour plate method was used for the enumeration of total fungal counts at 28°C using plate count PAD medium containing a pH of 6.8± 0.2.

Antifungal activity of actinomycetes extracts.

Actinomycete isolate was grown on starch nitrate medium and then incubated for ten days at 28°C for the production of secondary metabolites. Then the medium of fermentation was centrifuged at 8,000 rpm for 20 min. The supernatant was extracted by ethyl acetate (1:1), and then organic layers were evaporated to gain active substance. Theantifungal action of actinomycete extract was achieved by the standard agar-well diffusion method using PDA medium (Khattab et al., 2022).

RESULTS

The current review focuses on the microbial, chemical, and physical conditions of Qarun Lake. All of these initiatives are meant to persuade the government to put greater effort into halting or controlling the various lake pollution features. The current review was conducted through the examination of heavy metals in Qarun Lake water that influence the dispersal of various fungi.

Physical analysis of the water samples from Qarun Lake

In subsurface water, physical analyses (pH, temperature, and salinity) were determined. pH diverted from 7.0 at site 1 to 8.8 at site 5. A higher value of pH was seen in winter at site 5, while a lower value of pH was seen at site 1 through the summer. The temperature altered from 11.9 °C at site 1 to 31.6 °C at site 6. A higher value of temperature was noted through the summer at site 6, while a lower value was noted at location 5 through the winter. Salinity differed from 11.6% at site 1 to 37.4% at site 7. A higher value of salinity was noted through the summer at station 7, while a lower value of salinity was noted at station 1 through the winter. Data are noted in tables (2 and 3).

Chemical analysis of Qarun Lake water samples

Aluminum and iron concentrations were found in the ranges of 0.25–1.11 and 0.25–1.35 ppm, respectively. The lowest value of Al was 0.25 ppm at site 2 during the summer, while the highest value was 1.1 ppm at site 10 through winter. The lowest value of Fe was 0.25 ppm at site 2 through summer, while the highest value was 1.35 ppm at site 10 during the summer season. Manganese concentrations were found in the range of 19.2–139 µg/L. The lower value of Mn was 19.2 µg/L at site 7 through summer, while the higher value was 139 µg/L at site 8 through summer; copper and lead were detected only in stations 8, 9, and 10. The highest value of Cu was 12.5 µg/L at site 10 through the summer, while the highest value of Pb was 35.3 µg/L at site 10 through the summer. Cadmium and chromium were detected only in stations 9 and 10. The highest value of Cd was 3.21 µg/L at site 10 through the summer, while the highest value of Cr was 21.5 µg/L at site 10 during the summer. Zinc and nickel were detected only at stations 7, 8, 9, and 10. The highest value of Zn was 56.6 µg/L at site 10 during the summer, while the highest value of Ni was 12.5 µg/L at site 9 during the summer. The result recorded that beryllium (Be), cobalt (Co), molybdenum (Mo), selenium (Se), strontium (Sr), vanadium (V), titanium (Ti) and tellurium (Te) were not detected in all samples collected from Qarun Lake water. Data are noted in tables (2 and 3).

Microbial analysis and dispersal of fungi in Qarun Lake water

Throughout the study period (2020–2021), water samples were collected from 10 locations at Qarun Lake and tested for fungus on PAD media. No fungal count was detected in stations 8 and 9; TVFC in other locations of Qarun Lake varied between $0.7x10^2$ CFU/mL at site 7 in winter and $5.5x10^2$ CFU/mL at site 1 in summer. Data are noted in tables (2 and 3).

Isolation of fungi

Serial dilution was made up to 10^{-4}. Fungi were isolated from water samples by spreading method on surface plates of PDA media and afterwards, the plates were

incubated for 4–7 days at 28°C. In this study, 45 fungal isolates were isolated from samples collected from different locations in Qarun Lake through different seasons, after growth, fungal isolates were subjected to purification processes by inoculating each different isolate on different plate. After the incubation period, it was examined under a light microscope to study the microscopic characters of isolates.

Isolation of actinobacteria.

Dilution was made up to 10^{-4} and spread on SNA plates. After incubation, colonies with powdery texture, folded and branching threads without or with aerial mycelia were subcultured on SNA slants. Slants were preserved at 4°C until further use. *Streptomyces spp.* was isolated from Qarun Lake for selection of potent *Streptomyces spp*. One pure isolate of actinomycetes was tested for antifungal actions against several fungi in Qarun Lake.

Identification of fungi and actinomycetes isolates.

Morphological identification of fungi and actinomycetes isolates.

The results note that the SH2 isolate was in chains and filamentous in form, with gray aerial mycelium, white substrate mycelium, no pigments produced and with spherical spores (Figure 2).

The traditional macroscopic methods of colour, form, shape, margin, and elevation of the pure colonies were used to characterize the morphology of fungi (Table 4). At 28°C, the majority of colonies can grow after 4 to 7 days of incubation. The colony traits noted for the different isolates are displayed in (Table 4). The isolated fungi displayed several spores, mycelia, and hyphae (Figures 3 and 4). The conidia were arranged in chains at the ends of the aerial hyphae or in a sac-like structure. The hyphae could be septate or aseptate.

Molecular identification of fungi and actinomycetes isolates.

All sequences underwent BLAST analysis and comparison to the GenBank NCBI's most closely related sequences, and MEGA 7 software was used to create phylogenetic trees. The isolates were identified as *Aspergillus niger*, *Aspergillus versicolor*, *Aspergillus terreus*, *Aspergillus flavus*, *Aspergillus fumigatus*, *and Fusarium oxysporum.* In addition to *Streptomyces noursei strain SH2*, with accession numbers ON969128, ON969129, ON969130, ON969131, ON969133 and ON969134 in addition to OP782087 respectively. The phylogenetic trees were built by using the MEGA-X program and the neighbor-joining method. The PCR product's sequencing was carried out at the GATC Corporation utilizing an ABI 3730xl DNA sequencer and forward- and reverse-specific primers. PCR products were analyzed and entered as query on BLASTn on the NCBI site, the seven fungal isolates were submitted and their nucleotide sequences, phylogenetic tree with strictly related sequences on GenBank (Figure 5).

Incidence of fungal isolates at different locations of Qarun Lake

Station 1 had a higher number of fungal isolates; fourteen fungal isolates were isolated from station 1 representing by 31%. Moreover, fungal isolates at stations 2, 3, 4, 5,6 and 7 ranged from three fungal isolates representing 7% at station 7 to seven fungal isolates representing 16% at stations 3, while only one fungal isolate representing (2%) was isolated at station 10 and no fungal isolates were isolated at stations 8 and 9 as shown in figure (6).

Screening the antifungal activity of Streptomyces extract

In this research, the results of actinomycetes extract testing as an antifungal agent by diffusion through agar wells against different fungal isolates including (*A. versicolor*, *A. flavus*, *A. niger*, *A. Fumigatus*, *A. terreus*, and *F. oxysporum*) to determine susceptibility patterns. The isolation of actinomycetes showed activity against most of the tested fungi. The strain SH2 exhibited strong antifungal activity (inhibition zones) against *A. niger* (18 mm), *A. Fumigatus* (16 mm) and *F. oxysporum* (19 mm), while *A. versicolor, A. flavus* and *A. terreus* were resistant as shown in figure (7).

DISCUSSION

Many research studies have developed that Qarun Lake has huge significance for Al-Fayoum city development. The development of the environmental and urban future of Qarun Lake needs to be a combined system of ecological organization and city development to solve the problems facing this area. Human-caused biodiversity shifts and biota deprivation in Qarun Lake are becoming increasingly dangerous (Wang et al. 2021). Furthermore, increasing the amount of organic substances has an impact on the eco-balance of aquatic systems as well as the formation of a sediment microbial society (Hornick and Buschmann 2018). Nutrients (as organic

matter) and microbial populations in lakes have been the subject of increased research in recent years. With its importance for both long-term economic growth and environmental protection, water is a crucial global environmental concern. The "water value" category is based on a number of physical and hydrochemical elements. (Poonam et al. 2013). Water samples underwent chemical and physical examinations using the techniques outlined in APHA (Beutler et al. 2014). pH values in this study ranged from 7.0 at site 1 to 8.8 at site 5, remaining on the alkaline side. In many aspects of life, the H^+ concentration is crucial. The pH has a significant impact on and is highly sensitive to living things. The summertime temperature at site 6 was 31.4 ^oC, while the wintertime temperature at site 1 was 11.9^oC. Since water temperature affects all aquatic microorganisms' life processes and promotes their active proliferations, Qarun Lake's greater water temperature during the summer months resulted in high TVFCs. A significant physical component regulating the growth of bacteria is temperature. Salinity varied between 11.6% at site 1 and 37.4% at site 7. Heavy metals dispersal in Qarun Lake were noted at the southern part of the lake more than other parts of Qarun Lake due to the drains in the south, like the El Mashrouh drain. This agreed with results obtained by (El-Sayed et al., 2015). The lowest value of Al was 0.25 ppm at site 2, while the highest value was 1.1 ppm at site 10, and the lowest value of Fe was 0.25 ppm at site 2, while the highest value was 1.35 ppm at site 10. The lower value of Mn was19.2 µg/L at site 7, while the higher value was 139 µg/L at site 8. Copper and lead were detected only in stations 8, 9 and 10. The highest value of Cu was 12.5 µg/L at site 10, while the highest value of Pb was 35.3 µg/L at site 10. Cadmium and chromium were detected only in stations 9 and 10. The highest value of Cd was 3.21 µg/L at site 10, while the highest value of Cr was 21.5 µg/L at site 10. Zinc and nickel were detected only at stations 7, 8, 9 and 10. The highest value of Zn was 56.6 µg/L at site 10, while the highest value of Ni was 12.5 µg/L at site 9. The result records that Be, Co, Mo, Se, Sr, V, Ti and Te were not detected in all samples collected from Qarun Lake water.

Optimal temperature has an important role in the diversity of microorganisms at all environments (Kumar et al. 2022). Several severe settings including saline liquids, hot springs, the surface of dried rocks, ocean pits, dry deserts, and very low pH, as well as polar environments have produced a variety of fungus groupings. (Hassan et al. 2016). In this work, 25 fungal isolates representing 56% were isolated from samples of summer season, while only 20 fungal isolates representing 44% were isolated from samples of winter season. Thus, this may be due to best condition of temperature for fungal growth at summer season comparing with winter season. Fungal growth and sporulation are directly affected by salinity for example; the increase of sporulation and fewer hyphae form salinities above 5%, (Mulder and El-Hendawy, 1999; Mandeel, 2006). High ambient salts and pH put a great deal of stress on living things, which could have an impact on their biodiversity as a whole (Grum-grzhimaylo et al. 2016). Extremozymes and extreme metabolites are produced by fungi in extreme habitats under osmotic stress. (Raddadi et al., 2018). By storing K^+ ions in their cells, fungi are able to minimize water loss at high osmolarities (Plemenitas et al., 2014), while others list metabolites as suitable organic solutes such as polyols, sugars, and amino acids (Roberts, 2005). In this work 14 Fungal isolates representing 31% were isolated at station 1.This may be due to the fact that station 1 recorded lowest level of salinity and few heavy metals detected (only Fe, Al and Mn) with comparing with other stations at Qarun Lake, number of fungal isolates isolated at station 2, 3, 4, 5, 6 and 7 ranging between 3 fungal isolates representing 7% at station 7 to 7 fungal isolates representing 16% at station 3. This may be due to the fact that stations 2, 3, 4, 5, 6 and 7 recorded a high level of salinity and few heavy metals detected when compared with other stations at Qarun Lake, while only 1 fungal isolate representing 2% was isolated at station 10 and no fungal isolates were isolated at stations 8 and 9. This may be due to the fact that stations 8, 9 and 10 recorded high level of salinity and a high level of heavy metals when compared with other stations at Qarun Lake. The total viable count of fungi (TFVC) in lake water varied between $0.7x10^2$ CFU/ml at sit 7 in winter (the relative low values of TFVC) and $5.5x10^2$ CFU/ml at sit 1 in summer (the relative high values of TFVC). Thismay be because site 7 recorded a high level of salinity and heavy metals, while site 1 recorded a low level of salinity and no heavy metals were detected there.

Isolation and identification of fungal isolates were done under aseptic conditions by cultivation on PDA and MEA media. Six fungal isolates were fully identified as *A. versicolor, A. flavus, A. niger, A. fumigatus, A.*

terreus and F. oxysporum depending on the partial sequencing of 18S rDNA. Moreover, one isolate of actinobacteria was identified as *S. noursei* depending on partial sequencing of 16S rDNA.

One of the major bacterial lineages that includes a diverse array of uncommon halophilic actinomycetes is the actinobacteria (Subramani and Sipkema 2019). Actinomycetes release a multitude of substances against pathogenic bacteria and fungi (Al-Ansari et al. 2019), particularly intriguing and promising possibilities for the release of chemicals with useful properties that appear to be actinomycetes (Benhadj et al. 2020). Actinomycetes are well known for producing a wide variety of bioactive substances, including various hydrolytic enzymes. Moreover, numerous studies have demonstrated that actinomycetes are capable of successfully metabolizing a variety of different substances, including carbohydrates, lipids, alcohols, proteins, and amino acids (Subramani and Aalbersberg 2012). In this work, the antifungal activity of isolated *S. noursei* was carried out by diffusion assay through agar wells against fungi isolated from Qarun Lake. Most potent isolation of actinomycetes strains exhibited activity against most fungi tested. The strain SH2 exhibited strong antifungal activity against *A. niger* (18 mm), *A. fumigatus* (16 mm) and *F. oxysporum* (19 mm), while *A. versicolor, A. flavus* and *A. terreus* were resistant. Actinomycetes from Qarun Lake showed the synthesis of various antimicrobial activities against various fungi.

CONCLUSION

In conclusion, we can say that the lake water is polluted with heavy metals because agricultural sewage wastes exploded into Qarun Lake, causing serious problems with the lake's water quality and affecting the distribution of fungi in the lake. *S. noursei* can contribute to controlling the distribution of fungi in the lake. *S. noursei* demonstrated antifungal efficacy against three of the fungal isolates tested, with the inhibition zones as follows: *A. niger, A. fumigatus* and *F. oxysporum* (18, 16, and 19 mm) respectively, while *A. versicolor, A. flavus* and *A. terreus* were resistant. The primary factors determining physical, chemical, and microbiological features are climate and the rate of evaporation of Qarun Lake water. We recommend that the pollution in Qarun Lake be reduced by using biological and chemical treatment for wastewater before discharging it into the lake and by increasing the number of salt extraction factories to reduce the salt concentration.

ABBREVIATIONS

Al: Alumnium; Fe: Iron; Co: Cobalt; Cu: Copper; Cr: Chromium; Zn: Zinc; Ni: nickel; Be: beryllium; Mo: molobednum; Se: selenium; Sr: stronatium; V: vanadium; Ti: titanium; Te tellurium; PCR: Polymerase Chain Reaction; *A. niger: Aspergillus niger; A. Fumigatus: Aspergillus Fumigatus; A.versicolor: Aspergillus versicolor; A. terreus: Aspergillus terreus; F. oxysporum: Fusarium oxysporum; A.flavus: Aspergillus flavus; S. noursei: Streptomyces noursei s*; APHA: American Public Health Association; TVFC: Total Viable Fungal Count; YEA: Yeast Extract Agar; CMA: Corn Meal Agar; SNA: Starch Nitrate Agar.

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Table 1: Water samples sites at Qarun Lake.

Stations	Features of stations	Longitude	Latitude
1	Opposite to El-Baron	30° 49′ 14.1″	29°29′ 7.854″
2	Opposite to El-Arab Resort	30° 48′ 26.94″	29° 28′ 45.882″
3	Opposite to El-Bats Drain	30° 47′ 42.03″	29° 28′ 31.212″
4	Opposite to Loaloah village	30° 46′ 53.898″	29° 28′ 6.708″
5	Opposite to El-Oprerg lesan	30° 45′ 54.306″	29° 28′ 6.048″
6	Far North-East of Lake	30° 45′ 4.542″	29° 28′ 10.152″
7	Opposite to Marina Resort	30° 44′ 29.694″	29° 28′ 22.74″
8	Opposite to Goharah Resort	30° 43′ 57.762″	29° 28′ 35.406″
9	Opposite to Abou Nema Resort	30° 43′ 38.442″	29° 28′ 38.544″
10	Malahet Mizar (Far Lake West)	30° 26′ 08.4″	29° 26′ 37.1″

Table 2: Water analysis of samples collected from different sites of Qarun Lake during the summer season.

No.	Factor	Unit	Station									
			1	2	3	4	5	6	7	8	9	10
1	Temperature	°C	30.9	29.4	29.8	31.1	30.8	31.6	31.0	30.6	31.2	29.9
2	Salinity	%	12.6	15.9	27.8	28.5	21.8	30.1	37.4	30.1	31.3	27.9
3	pH		7.0	7.9	8.0	7.5	8.2	7.8	7.4	7.7	8.1	7.8
4	Al	ppm	0.28	0.25	0.35	0.39	0.63	0.58	0.75	0.43	1.02	0.89
5	Be	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
6	Cd	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.21
7	Co	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
8	Cr	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	7.28	21.5
9	Cu	µg/L	ND	ND	ND	ND	ND	ND	ND	1.98	ND	12.5
10	Fe	ppm	0.28	0.25	0.33	0.39	0.63	0.68	0.75	0.83	1.12	1.35
11	Mn	µg/L	69.3	29.3	68.2	92.1	39.2	58.4	19.2	139	99.5	112
12	Mo	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
13	Ni	µg/L	ND	ND	ND	ND	ND	ND	6.25	11.7	12.5	6.98
14	pb	µg/L	ND	ND	ND	ND	ND	ND	ND	15.7	27.5	35.3
15	Sr	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
16	Ti	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
17	V	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
18	Zn	µg/L	ND	ND	ND	ND	ND	ND	19.2	17.8	28.1	56.6
19	Se	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
20	Te	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
21	TVFC	$\times 10^2$ CFU/mL	5.5	4.8	5.0	4.5	4.8	3.8	3.5	ND	ND	1.5

ND= not detected

Table 3: Water analysis of water samples collected from different locations of Qarun Lake during the winter season.

No.	Factor	Unit	Station									
			1	2	3	4	5	6	7	8	9	10
1	Temprature	°C	12.0	12.5	12.9	12.3	11.9	12.2	13.5	13.2	12.9	12.8
2	Salinity	%	11.6	11.9	21.2	18.3	22.9	22.9	37.3	31.2	28.9	21.2
3	pH		7.5	8.1	8.0	7.4	8.8	8.1	7.8	7.6	7.9	8.5
4	Al	ppm	0.33	0.41	0.45	0.31	0.53	0.66	0.59	0.27	0.86	1.1
5	Be	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
6	Cd	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	1.53	1.85
7	Co	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
8	Cr	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	4.53	18.5
9	Cu	µg/L	ND	ND	ND	ND	ND	ND	ND	0.99	6.96	11.8
10	Fe	ppm	0.31	0.41	0.35	0.31	0.53	0.46	0.59	0.97	0.96	1.08
11	Mn	µg/L	26.5	29.2	88.2	57.3	66.3	98.2	26.6	99.2	66.6	118
12	Mo	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
13	Ni	µg/L	ND	ND	ND	ND	ND	ND	ND	6.27	12.4	8.47
14	pb	µg/L	ND	ND	ND	ND	ND	ND	ND	16.2	22.4	18.5
15	Sr	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
16	Ti	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
17	V	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
18	Zn	µg/L	ND	ND	ND	ND	ND	ND	15.9	ND	47.6	11.8
19	Se	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
20	Te	µg/L	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
21	TVFC	$\times 10^2$ CFU/mL	1.2	1.5	1.0	2.0	2.5	1.0	0.7	ND	ND	1.4

ND= not detected

Isolate	Morphological characters				
	Top color	Bottom color	Margin	Form	Elevation
Fsp1	Yellowish-green	Yellowish-gold	Entire	Irregular	Raised
Fsp2	Cream white	Brownish gold	Entire	Circular	Raised
Fsp3	Cream white	Brownish gold	Entire	Circular	Raised
Fsp7	Cream with purple rings	Brownish red	Entire	Circular	Raised
Fsp11	Cream black	sulfur-yellow	Entire	Circular	Raised

Table 4: Morphological characteristics of fungal isolates.

Fsp12	gray	Cream	Undulate	Circular	Flat

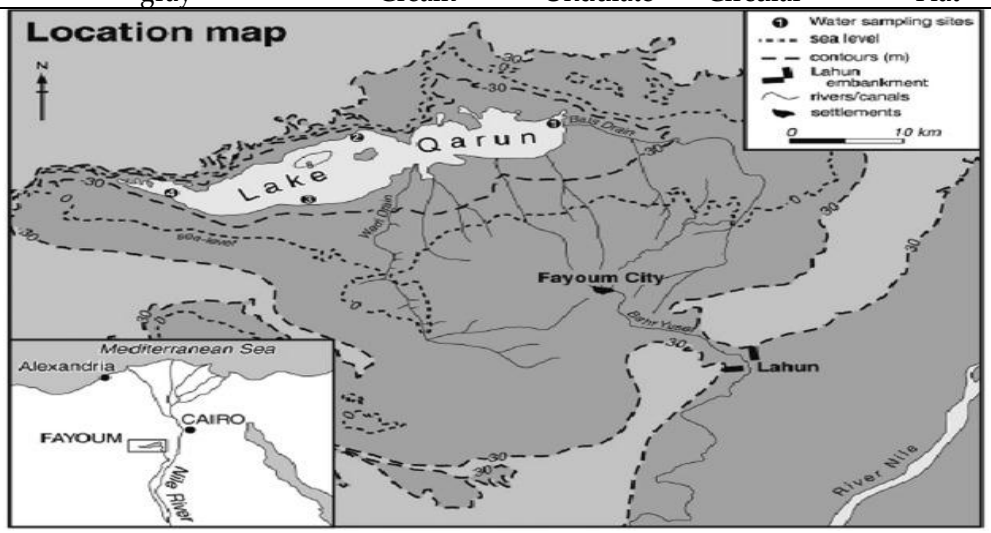
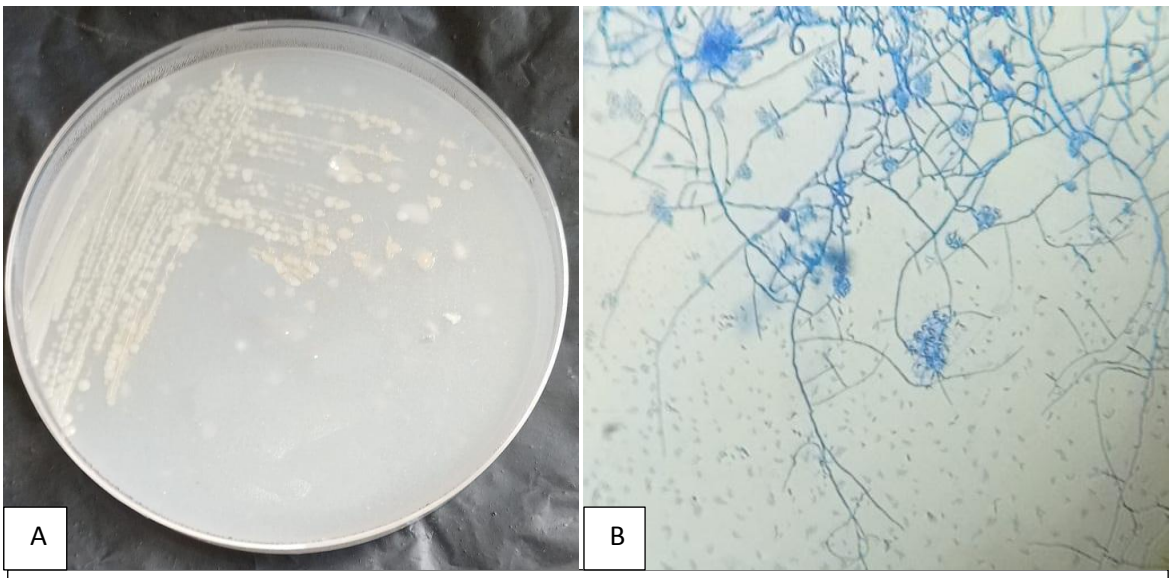
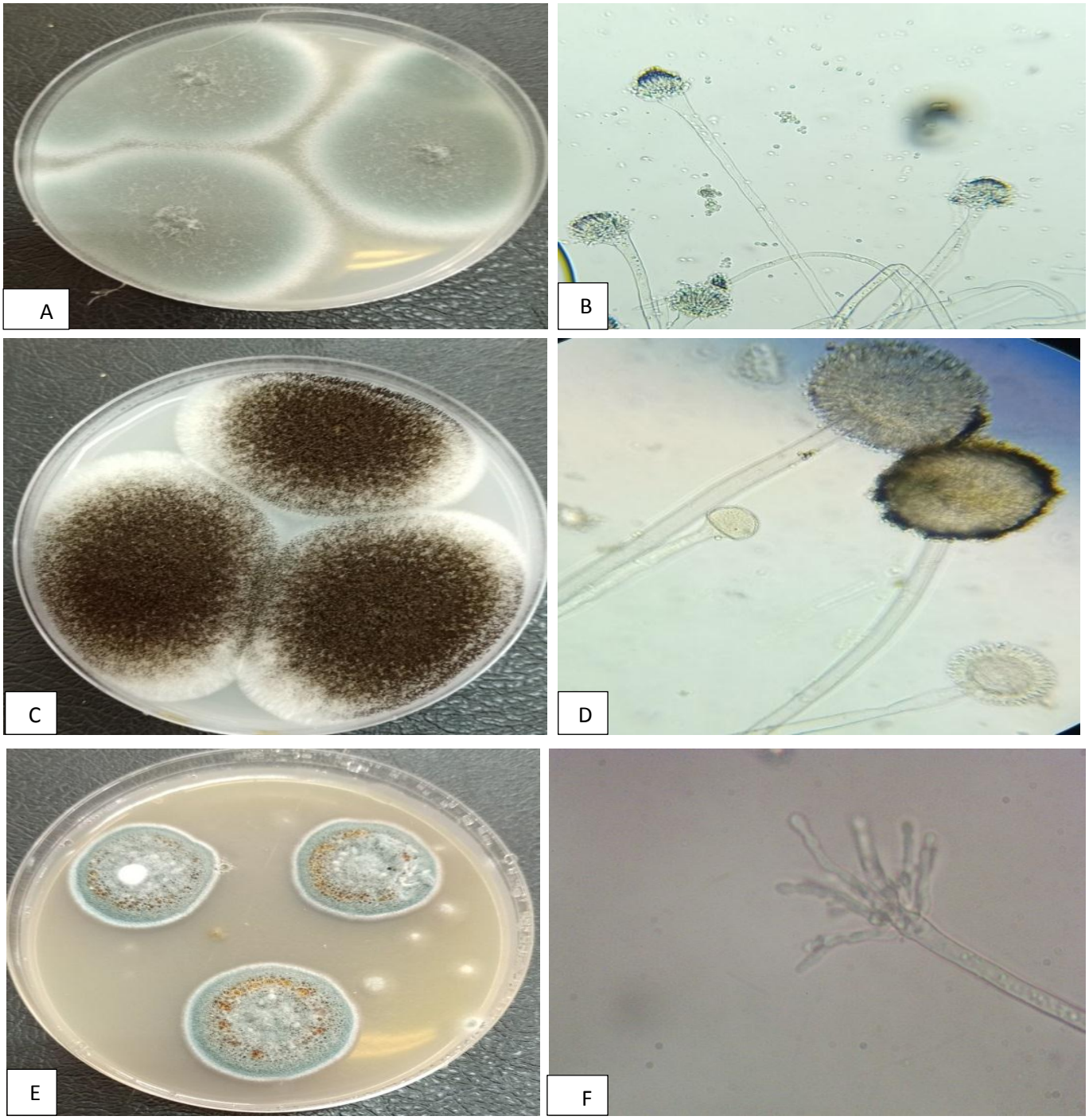


Figure 1: Location of Qarun Lake at Al-Fayoum governorate (Abdelbaki, 2021).



A. Colonies of isolates SH2 on starch nitrate agar medium.
B. Light microscope showing aerial mycecillium and spores of SH2 isolate (mag. Power 20×40).

Figure 2: Morphological and microscopic characters of isolate SH2.



A. Colonies surface of *Aspergillus Fumigatus* on PDA medium**.**

B. Light microscope showing conidiophores and spores of *Aspergillus Fumigatus* (mag. Power 20×40).
C. Colonies surface of *Aspergillus Niger* PDA medium**.**
D. Light microscope showing conidiophores of *Aspergillus Niger* (mag. Power 20×40).
E. Colonies surface of Aspergillus versicolor PDA medium.
F. Light microscope showing conidiophores of Aspergillus versicolor (mag. Power 20×40).

Figure 3: Morphological Characters of different fungal isolates.

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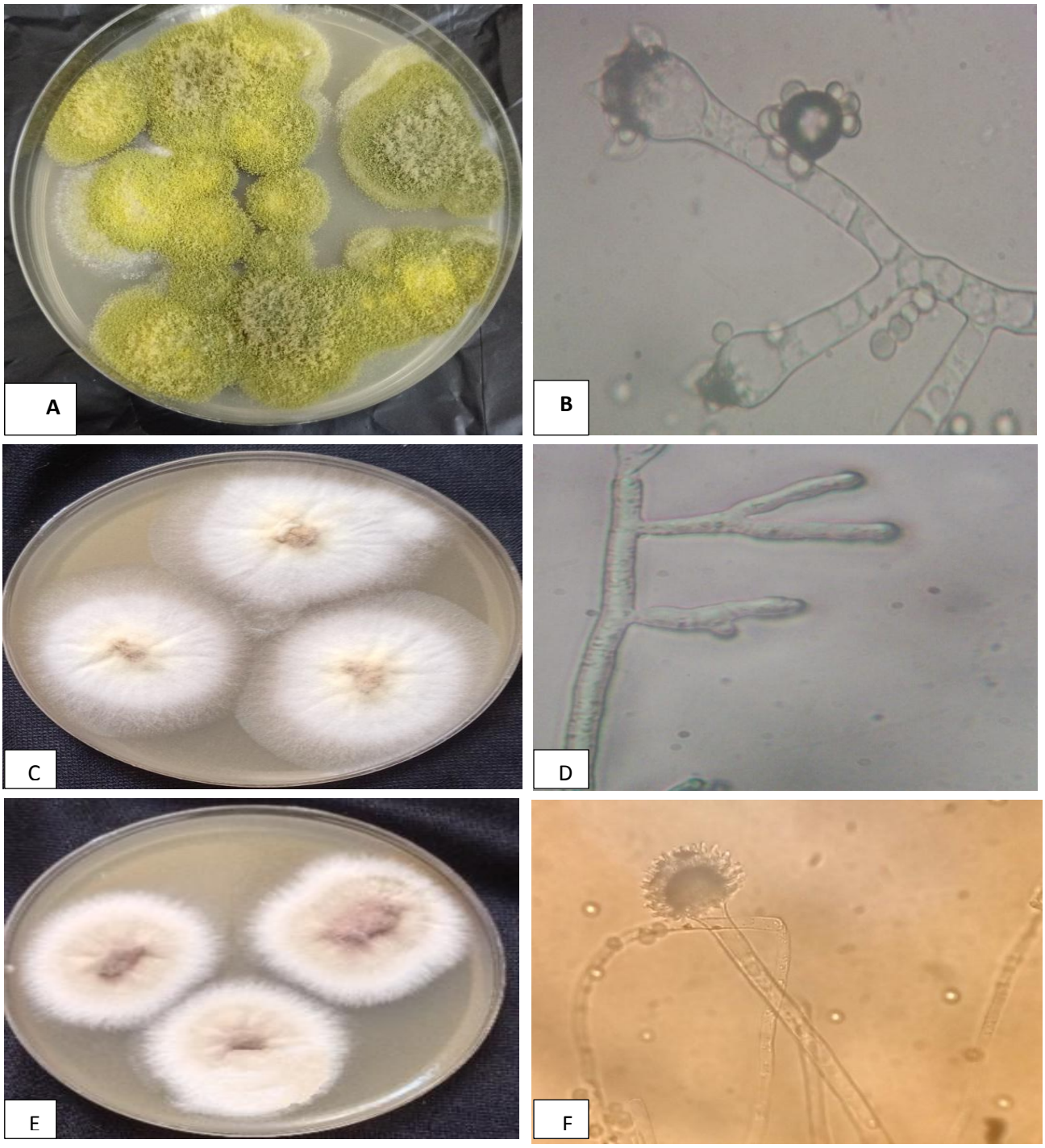


A. Colonies surface of *Aspergillus flavus* PDA medium**.**
B. Light microscope showing conidiophores and spores of *Aspergillus flavus* (mag. Power 20×40).
C. Colonies surface of *Fusarium oxysporum* on MEA medium**.**
D. Light microscope showing conidiophores of *Fusarium oxysporum* (mag. Power 20×40).
E.Colonies surface of *Aspergillus terreus* on PDA medium**.**
F. Light microscope showing conidiophores of *Aspergillus terreus* (mag. Power 20×40).

Figure 4: Morphological Characters of different fungal isolates.

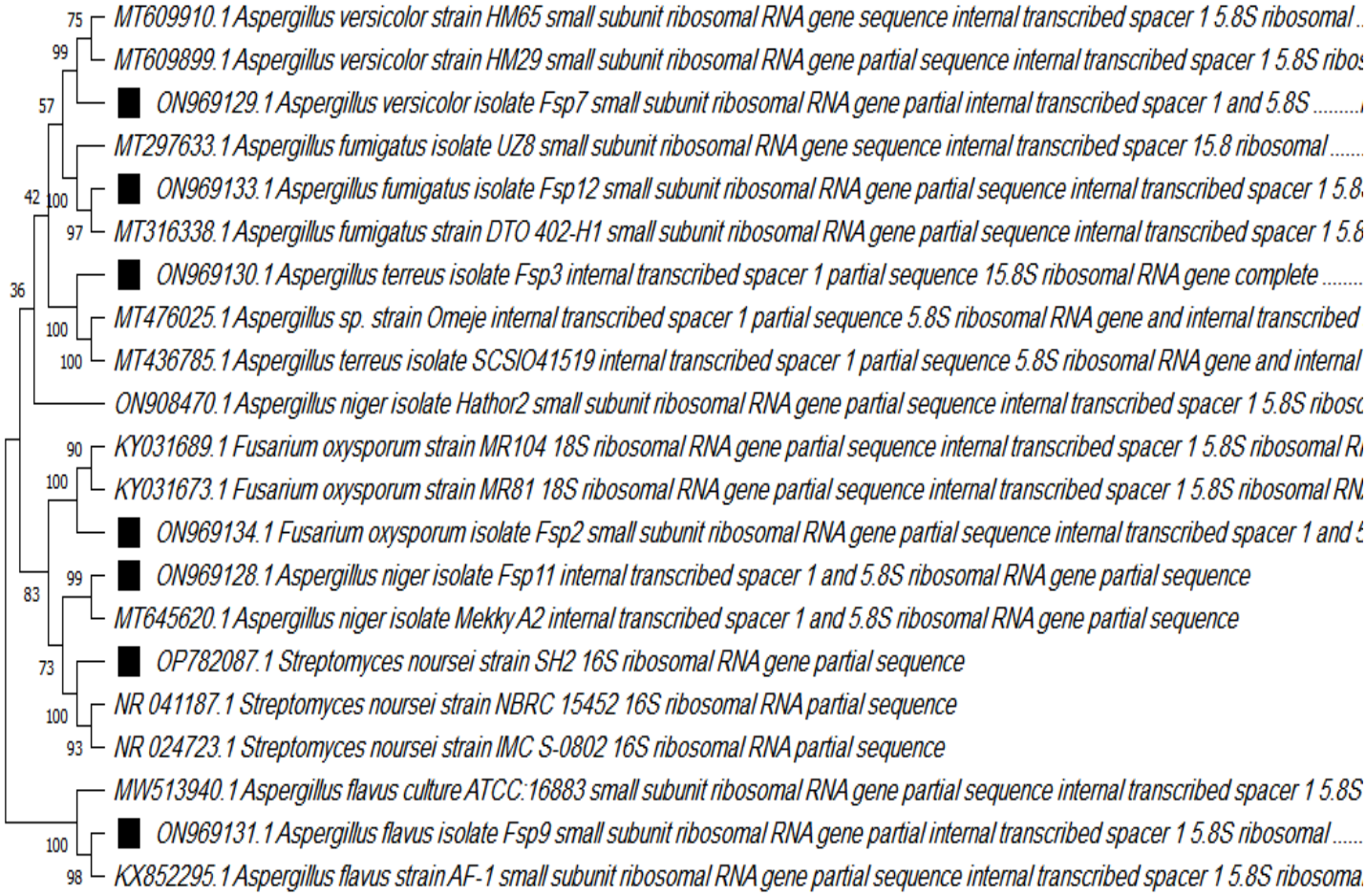


Figure 5: Phylogenetic analysis of different fungal and actinomycetes isolates, Neighbor-joining tree displaying the phylogenetic position of these isolates and phylogenetically related members of thire genus.

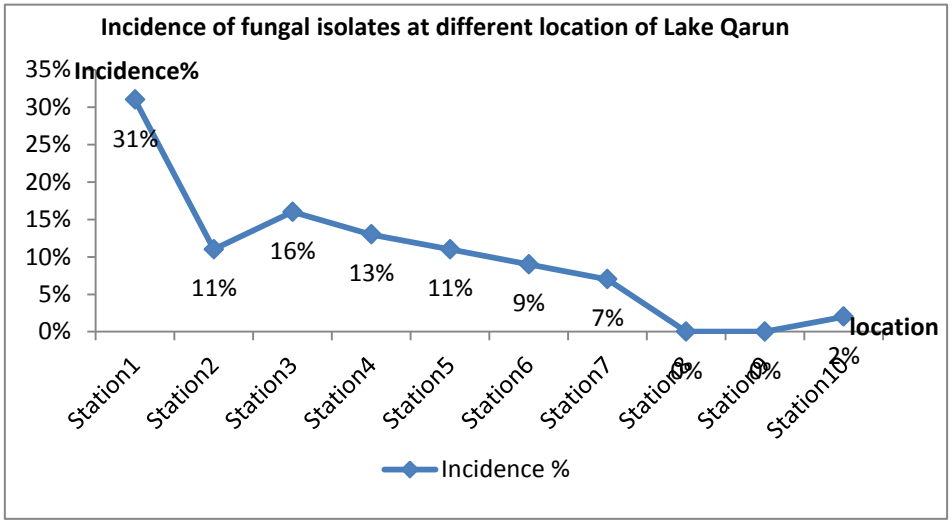
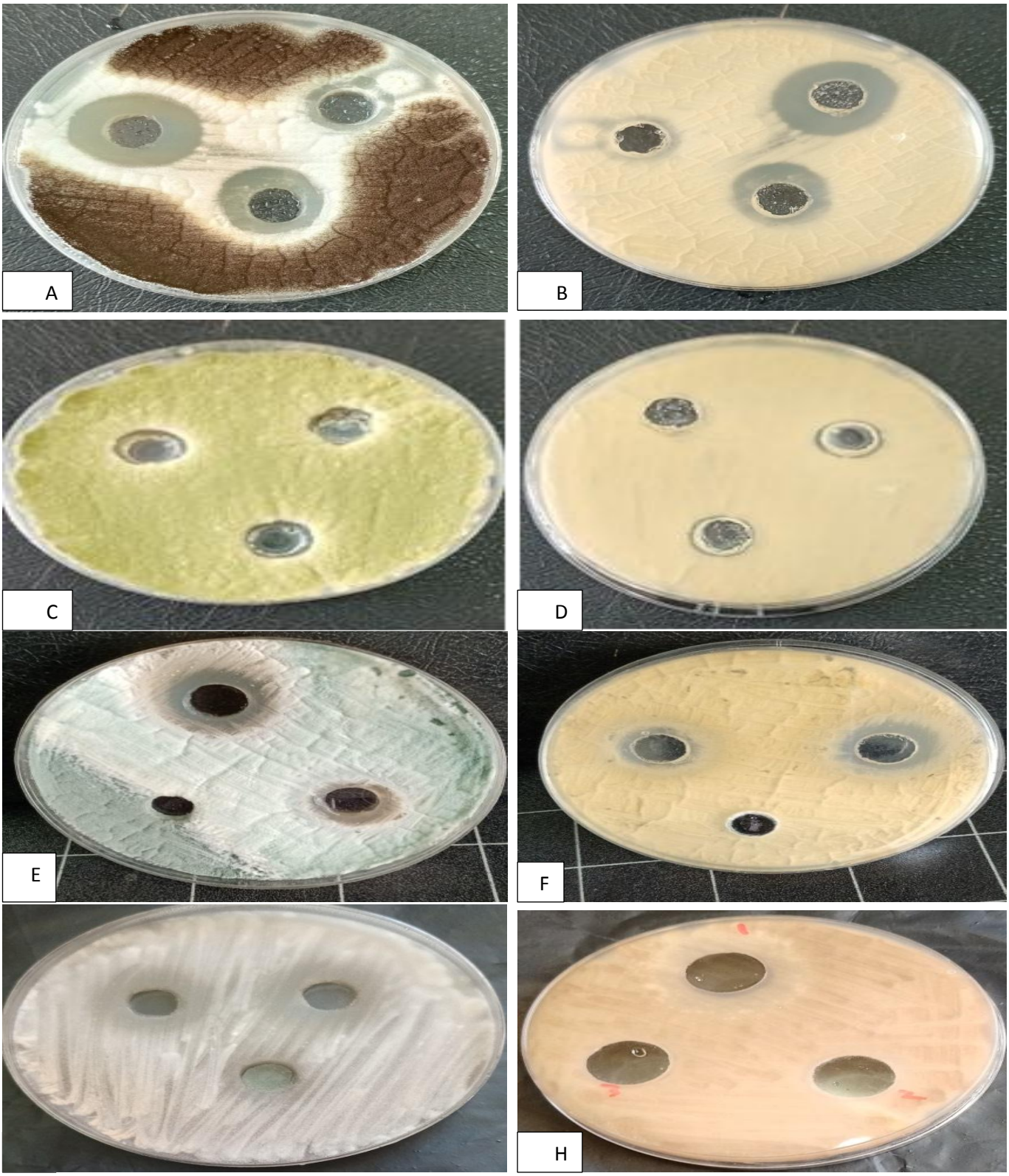


Figure 6: Incidence of fungal isolates at different location of Qarun Lake



A. and **B**. Antimicrobial activity of SH2 isolate on *A. niger*.
C. and **D.** Antimicrobial activity of SH2 isolate on *A.flavus.*
E. and **F**. Antimicrobial activity of SH2 isolate on *A. Fumigatus.*
G. and **H**. Antimicrobial activity of SH2 isolate on *F. oxysporum*.

Figure 7: Antifungal activity of SH2 isolate on fungal isolates.

انتشار المعادن الثقيلة وتأثيرها على التنوع الميكروبي في بحيرة قارون

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الملخص العربي

تعد بحيرة قارون من البحيرات التي لها طابع خاص حيث إن لها أهمية تاريخية في مصر وكذلك لأنها تحاط بالمجتمع الريفي. جمعت عينات المياه من عشرة أماكن مختلفة في بحيرة قارون وكذلك في مواسم مختلفة لكي نلاحظ تنوع الفطريات على مدار فصول السنة. كذلك أجريت تحاليل فيزيائية وكيميائية لقياس نسبة العناصر الثقيلة في عينات المياه لكى نلاحظ تأثيرها على تنوع الفطريات في مياه البحيرة حيث لاحظنا انتشار كل من النيكل والكروم والرصاص والنحاس والكادميوم والزنك في مناطق مختلفة من البحيرة وبنسب متفاوتة وكان لها دور في انتشار الفطريات في هذه المناطق. في هذه الدراسة عُزلت ستة أنواع مختلفة من الفطريات وتم تعريفهم على مستوى الشكل الظاهري والفحص الميكروسكوبي وكذلك على المستوى الجيني بالإضافة الى عزل نوع واحد من الاكتينوميستات وتم تعريفها أيضا مورفولوجيا وكذلك جينيا مع إجراء اختبار تأثير نشاطها ضد الفطري على الفطريات التي عُزلت من البحيرة حيث كان لها نشاط على بعض الفطريات التي عزلت من البحيرة والبعض الآخر كان مقاوماً لتأثيرها.

الكلمات الاسترشادية: بحيرة قارون، التنوع الميكروبي، اكتينوبكتريا، المعادن الثقيلة، النشاط المضاد للفطريات.