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First DNA Barcoding-based Inventory of Suez Gulf Fishes in Egypt and its Implication for Species Diversity

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Abstract—The Suez Gulf is one of the most important water bodies north of the Red Sea that contribute significantly in fish production in Egypt. The current study represents the first molecular identification of marine fish species from the Suez Gulf in Egypt based on DNA barcoding. A total of 96 DNA barcodes using a 662 bp-long fragment of the mitochondrial cytochrome oxidase subunit I (*COI*) gene were generated from 32 species that represent 26 genera and 19 families. The average genetic divergence based on Kimura two-parameter model between families and species were 0.274 and 0.256, respectively. The genetic distance among families varied greatly with the lowest genetic distance of 0.190 obtained between Carangidae and Atherindae, while Sphyraenidae and Moronidae had genetic distance of 0.472. The percent GC content was above average across species and ranged between 51.74% in *Acanthopagrus bifasciatus* and 66.37% in *Lagocephalus guentheri*. The Neighbor Joining tree showed clear clustering of the 19 fish families with species of the same family formed a single cluster. The findings of the current study support that *COI* barcode effectively identified Suez Gulf fish species. Moreover, the results of the current study will establish the first DNA barcode records for Suez Gulf fishes in Egypt which will contribute in future fish species identification in the Suez Gulf and the Red Sea.

Keywords: DNA barcoding, cytochrome oxidase subunit one gene (*COI*), Red Sea, Suez Gulf fish inventory, fish biodiversity

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INTRODUCTION

The Red Sea is one of the worldwide hotspots. It is known to harbor significant number of biodiverse fauna and biota, yet with high endemism rate as compared to surrounding marine water bodies (DiBattista et al., 2016). The significant endemism rate at the Red Sea is, in part, associated with its location at the northwest of Gulf of Aden strait of Bab al-Mandab. Many environmental conditions including temperature, salinity, and shallow depth can diminish the movement of species between the Red Sea and the Indian Ocean (Kemp, 2000). The Red Sea is linked to the Mediterranean via Suez Canal in the north, which causes a unidirectional spreading of species, named Lessepsian migration (Por, 1978). In addition, the Red Sea has a historical salty and warm depth with near isothermal and isohaline water (around 22°C and 40%, respectively) which is apparent in large parts of the shallow depth (Neumann and McGill, 1961), leading to a highly specialized and partially endemic fauna. The significant coral reefs coverage of over 16000 km^2 in the Red Sea is considered the main factor associated with elevated species diversity (Roberts et al., 2002). The Suez Gulf expands for 250 km between Suez in the north (Lat. 29°56′ N) to Shadwan Island in the south (Lat. 27°36′ N). Its width fluctuates between 20 and 40 km, with a constant depth of an average of 45 m. The Suez Gulf is considered as a part of the Red Sea due to barriers absence between the two water bodies, which suggest the establishment of most of Red Sea fish species in the Suez Gulf.

Since 1960s, species identification is considered a major focus for fisheries Food and Agriculture Organization (FAO). According to FAO reports, about 35% of the fisheries catch have not been identified into species (Lleonart et al., 2006). Nowadays, the influence of human on nature through environmental disturbances, and climate change has greatly altered many ecosystems (Triantafyllidis et al., 2011). However, the monitoring and conservation of fish biodiversity must rely on precise identification of species (Dayrat, 2005). DNA barcoding is an efficient way of species identification (Hebert et al., 2003; Hajibabaei et al., 2007; Ali et al., 2019). DNA barcoding secures quick and precise species identifications based on a short

consistent fragment of the genome. DNA barcoding has gained universal popularity as an efficient tool for species identification (Hebert et al., 2003). This technique is especially helpful when studies aimed to identify species translocated from different geographical locations (Bergsten et al., 2012).

Although, The Red Sea is a biodiversity rich water body with large varieties of fish and other marine organisms, few studies dealt with estimating the number of Red Sea fish species or making reliable inventories for the established species in Red Sea. Moreover, the reported checklist for Red Sea fishes relied only on the morphological characteristics of fish species. Golani and Fricke, (2018) provided a checklist for Red Sea fishes which counts 1207 species, representing 164 families, of these, 339 from the Suez Gulf. Whereas the total number of endemic species in the Red Sea is 174 species, of these, 8 are endemic to the Suez Gulf (Golani and Fricke, 2018). In Egypt, Akel and Karachle (2017) reported that the Egyptian marine waters encountered a total of 956 marine fish species (based on FishBase and the Global Biodiversity Information Facility). Of those, 592 species are present only in the Red Sea, 263 species are present only in the Mediterranean Sea, and the remaining 101 species are reported from both seas.

On the other hand, few studies were conducted for the identification of marine fish species in the Red Sea, based on molecular approaches. Trivedi et al. (2016), provided *COI* barcodes for only six fish species belonging to Red Sea in Saudi Arabia, these species included *Epinefhelus chlorostigma*, *Siganus rivulatus*, *Carangoides bajad*, *Carangoides bajad*, *Lutjanus ehrenbergii* and *Pristipomodies filamentosus*. Bariche et al. (2015) recorded the invasion of approximately 90 species of marine fishes into the Mediterranean Sea based on DNA barcoding using *COI* partial sequence. In Egypt, Ahmed et al. (2016) provided barcodes for four cryptic fish species of family Mullidae based on *COI* partial sequences. Abbas et al. (2016) used *COI* barcode partial sequen (Folmer et al., 1994) to characterize five different crabs' species that were collected from the northern part of the Egyptian Red Sea. Abbas et al. (2017) have studied the phylogenetic relationships among fish species of family Sparidae based on *COI*. The same family was further studied by Ibrahim et al. (2020) based on *Cyt b*, RAPD and ISSR markers. The western king prawn *Melicertus latisulcatus* from Suez Gulf, Red Sea was molecularly characterized by Abbas et al. (2018). Also, Galal-Khallaf et al. (2019) employed DNA barcoding to authenticate some fish species of family Serranidae inhabiting the Red Sea.

Due to the lack of biodiversity assessment focused on the Red Sea and the scarcity of studies that accurately identified the Red Sea fishes, there is an urgent need to explore the biodiversity of the Red Sea. Special attention needs to be directed to Suez Gulf, which is considered the most productive fishing ground alongside the Egyptian sector of the Red Sea where over 64% of Egyptian Red Sea fish production belong (Mehanna and El-Gammal, 2007). This makes the Suez Gulf an ideal fishery that can represent the biodiversity of the Red Sea.

In this study, a partial sequence of Mitochondrial Cytochrome c oxidase subunit 1 gene (*COI*) was employed to barcode and identify the commercial marine fish species collected from the Suez Gulf, in Egypt. Mitochondrial Cytochrome c oxidase subunit 1 (*COI*) is considered an efficient barcode region of different animal species (Hebert et al., 2003), particularly, the identification of fish species (Lakra et al., 2011; Deichmann et al., 2017). Here, we report the first inventory of representative marine fish species in Suez Gulf in Egypt based on DNA barcoding. This inventory will serve as a reference library to compare DNA sequences against in the future studies. Furthermore, the genetic diversity of different Red Sea fish species will be assessed. The information obtained in the current study will provide guidelines in the conservation efforts of the gulf fish species in Egypt.

MATERIALS AND METHODS

Fish Sampling and Morphological Identification

Approximately 150 fish samples were collected from the commercial catch of Suez Gulf, north of the Red Sea, Egypt (between 29°58′ N and 32°31′ E). The collected fish samples were transferred frozen to the genetics laboratory, National Institute of Oceanography and fisheries. The collected samples were sorted into 32 different species according to their external features (Froese and Pauly, 2017). Sample photos of the studied species are presented in Supplementary Table S1. For each species, three fish samples were selected for barcoding where small pieces of the dorsal muscle were sampled and preserved in absolute ethanol in –20°C for DNA isolation and PCR amplification.

DNA Extraction, Amplification, and Sequencing

DNA was isolated from the muscle tissues of the sampled fishes, using phenol chloroform method (Ibrahim et al., 2020). Briefly, about 50 mg muscle tissue were ground through a tissue lyser using 500 of Tris-EDTA buffer (100 mM Tris-HCl, 10 mM EDTA, pH 8). The ground tissues were lysed in water bath at 56°C with 0.5 mg mL^{-1} of proteinase K. DNA was precipitated using ice cold absolute ethanol and washed twice with 70% ethanol. DNA was dissolved in TE buffer and preserved at 4°C for PCR amplification. For species barcoding, primer pairs described by Kochzius et al., 2010 on the basis of the universal primers of Ward et al., 2005: *COI*-Fish-F (5'-TTCTCA ACTAACCAYAAAGAYATY GG-3') and *COI*-Fish-R (5'-TAGACT TCT GGG TGG CCR AAR AAY CA-3'), were used to amplify a partial *COI*

fragment. The target *COI* fragment were amplified in a 30 µl PCR mixture using the BIOLINE master mix (2X My-Taq Red Mix, Meridian bioscience, USA) according to the manufacturer's instructions. The PCR mixture contained 15 µL master mix, 0.7 µL of each primer (final concentration 0.25 µM) and DNA template with final concentration of 20 ng/µL. PCR amplification was conducted in a BIO-RAD PCR system (T100 96-well Thermal Cycler, BIO-RAD, USA) using the following thermal profile: initial denaturation at 95°C for 5 min, followed by 35 cycles of a denaturation step at 94°C for 30 s, annealing temperature at 52–55°C (varied among different species), for 30 s and 30 s extension at 72°C, with a final extension of 7 min at 72°C. Successful amplification were confirmed by loading 2.5 µL of the PCR products onto 2.5% agarose gel containing 2 µL of ethidium bromide (100 mg/mL), and electrophoresed. PCR products with acceptable bands size (approximately 700 bp) were purified using PCR/Agarose DNA purification kit (Intron Biotechnology, Korea) and sequenced using ABI's Big Dye Terminator kit on the Applied Biosystems 3500 Genetic Analyzer Sequencer (Hitachi, Japan) (Abbas et al., 2011). Sequences of all species were deposited in GenBank/EMBL/DDBJ genetic databases with the accession numbers LC543868 to LC543963.

COI Sequence-Based Identification

All species sequences were aligned and edited using BioEdit (Hall, 1999). Following sequence alignment and editing, an average of 662 bp *COI* partial sequence was obtained across all species. All sequences used in the current study were deposited in GenBank/EMBL/DDBJ genetic databases with the accession numbers LC543868 to LC543963. Species identification based on *COI* barcode sequences followed two main approaches. Firstly, BLAST searches was conducted using FASTA sequences for each sample to identify highly similar sequences on GenBank (https://blast.ncbi.nlm.nih.gov/Blast.cgi, accessed on May 1st, 2020). Also, a single *COI* sequence for each sepceis (accession numbers LC571884 to LC571904 and LC572148 to LC572158) were identified using the Barcode of Life Data System—BOLD (Ratnasingham and Hebert, 2013). BOLD assign *COI* sequence of the studied specimens to its respective Barcode Index Number (BIN) which cluster the speceismin sequence with closely congruent species. Secondly, we used barcode gap analyses as implemented in Automated Barcode Gap Discovery (ABGD) at the web interphase (https://bioinfo.mnhn.fr/abi/public/abgd/abgdweb.html, accessed on May 1st, 2020) which assign sequences into presumed species according to the barcode gap with no prior species information (Puillandre et al., 2011). ABGD was used with Kimura two-parameter distance model (K2P) with transition/transversion ratio (TS/TV) set to 1.5, 10 recursive steps, X (relative gap width) = 1.0 and the remaining parameters set to default values ($P_{min} = 0.001$, $P_{max} = 0.1$, Nb bins = 20).

Average genetic divergence within families and species were estimated based K2P (Kimura, 1980) as implemented in MEGA X (Kumar et al., 2018). Nucleotide diversity (π) for each species was estimated as the average number of nucleotide differences per site between two sequences according to (Nei and Miller, 1990) with Jukes and Cantor correction (Jukes and Cantor, 1969) using DnaSP (Rozas et al., 2017). The percentage of total GC content (% GC) was also calculated for all species using BioEdit. Genetic distance among studied families was estimated according to K2P as implemented in MEGA X. The phylogenetic tree was constructed using Neighbor–Joining tree method (Saitou and Nei, 1987) based on K2P distance. To better represent the clustering pattern of fish families, the NJ tree was generated based on a single *COI* sequence of 585 bp across all studied species. The NJ tree branches robustness was estimated by bootstrapping using 1000 pseudoreplicate datasets.

RESULTS

Sequence-Based Identification and Species Delimitation

A total of 96 partial sequences of *COI* gene were obtained from 32 species belong to 26 genera and 19 families. Sequencing of the partial *COI* gene produced sequences with lengths ranged between 585 to 688 bp with an average of 662 bp. No stop codons, insertions, or deletions were observed in any of the amplified sequences. BLAST search was used to compare the obtained *COI* sequences for the 32 collected species to the available data in the GenBank database yielded 11 species (34.5%) showed 100% similarity, 16 species (50%) showed ≤ 99% identity and 5 species (15.6%) showed ≤ 98% similarity (Table 1). In addition, the BOLD database assigned the studied species to an existing BINs that matched the morphological description (Table 1).

Barcode gap analysis using ABGD delimited the studied species into different 32 putative partitions (species) with prior maximal distance, of $P = 0.0359$ and $P = 0.001$, respectively with each species was assigned into a separate group (Fig. 1). The average genetic divergence between families ranged between 0.190 and 0.472 with an average of 0.274 (Table 2). Between species, genetic divergence ranged between 0.092 and 0.474 with an average of 0.256 (Table 2). Sequence analysis of the *COI* partial gene showed that percent GC content varied between species and ranged between 51.74% in *Acanthopagrus bifasciatus* and 66.37% in *Lagocephalus guentheri* (Fig. 2).

The studied species showed very low nucleotide diversity ($\pi = 0$) except *Rhabdosargus haffara* and *Siganus rivulatus* showed nucleotide diversity of 0.028 and 0.150, respectively. Genetic distance among families showed wide range, with the lowest genetic dis-

Table 1. List of Suez gulf spices barcoded using *COI* , their families, Barcode Index Number (BIN) and BLAST search results representing most similar sequences to Suez Gulf fish species

Family	Species	BINs ID	Most similar sequence	Accession no.	Percentage of identity, %
Sparidae	1. *Diplodus noct*	NA	*Diplodus noct*	MF123858.1	100.00
	2. *Acanthopagrus bifasciatus*	BOLD:ADF5006	*Acanthopagrus bifasciatus*	LC150892.1	100.00
	3. *Rhabdosargus haffara*	BOLD:ACG7708	*Rhabdosargus haffara*	KP308279.1	100.00
	4. *Argyrops spinifer*	BOLD:AAB3720	*Argyrops spinifer*	MT325508.1	100.00
	5. *Crenidens crenidens*	BOLD:ACU3055	*Crenidens crenidens*	LC155797.1	100.00
Mullidae	6. *Upeneus vittatus*	NA	*Upeneus vittatus*	KT823808.1	100.00
	7. *Parupeneus forsskali*	BOLD:ACU3747	*Parupeneus forsskali*	MH331820.1	100.00
	8. *U. pori*	BOLD:AAC1406	*U. pori*	KY176690.1	99.53
Carangidae	9. *Trachurus indicus*	BOLD:AAA8614	*Trachurus indicus*	KU179049.1	100.00
	10. *Decapterus maruadsi*	BOLD:AAB6796	*Decapterus maruadsi*	KY451627.1	98.74
Sillaginidae	11. *Sillago suezensis*	BOLD:AAA7605	*Sillago suezensis*	KY176638.1	99.37
Lethrinidae	12. *Lethrinus* sp.	BOLD:ADX4312	*Lethrinus* sp.	KY675565.1	99.80
	13. *L. borbonicus*	BOLD:AAB0511	*L. borbonicus*	KU499639.1	99.84
	14. *L. nebulosus*	BOLD:ABY6363	*L.nebulosus*	KU499797.1	99.55
Siganide	15. *Siganus luridus*	BOLD:AAL9467	*Siganus luridus*	MF409629.1	99.23
	16. *S. rivulatus*	BOLD:ABY0829	*S.rivulatus*	MF124055.1	99.84
Mugilidae	17. *Liza carinata*	BOLD:ABX8353	*Liza carinata*	KR861538.1	99.52
Platycaphalidae	18. *Platycephalus indicus*	BOLD:AAB2371	*Platycephalus indicus*	KR861547.1	100.00
Sphyraenidae	19. *Sphyraena chrysotaenia*	BOLD:AAD0400	*Sphyraena chrysotaenia*	KY176643.1	98.58
Gerreidae	20. *Gerres oyena*	BOLD:AAC1291	*Gerres oyena*	KU179069.1	99.25
Atherindae	21. *Atherinomorus forskalii*	BOLD:ACJ4684	*Atherinomorus forskalii*	KY176403.1	99.53
Synodontidae	22. *Trachinocephalus* sp.	BOLD:ADL2607	*Trachinocephalus* sp.	KX139521.1	99.69
	23. *Saurida lessepsianus*	BOLD:ACG7154	*Saurida lessepsianus*	KY176611.1	99.84
Monacanthiae	24. *Stephanolepis diaspros*	BOLD:ACF6911	*Stephanolepis diaspros*	KM538597.1	98.76
Nemipteridae	25. *Nemipterus randalli*	BOLD:AAE3907	*Nemipterus randalli*	KU236033.1	98.51
Clupeidae	26. *Sardinella aurita*	BOLD:AAB7268	*Sardinella aurita*	KM538516.1	99.39
Serranidae	27. *Serranus cabrilla*	BOLD:AAD1027	*Serranus cabrilla*	KY176628.1	98.12
Moronidae	28. *Pomadasys stridens*	BOLD:AAB8677	*Pomadasys stridens*	KM538505.1	99.04
	29. *Dicentrarchus punctatus*	BOLD:ADC6850	*Dicentrarchus punctatus*	LC317282.1	100.00
Tetraodontidae	30. *Lagocephalus suezensis*	BOLD:ACG7296	*Lagocephalus suezensis*	KU324616.1	100.00
	31. *L. guentheri*	BOLD:ADG5739	*L. guentheri*	KU324615.1	100.00
	32. *L. sceleratus*	BOLD:AAC5565	*L. sceleratus*	KU324609.1	99.83

NA—not available.

tance of 0.190 obtained between Carangidae and Atherindae, while Sphyraenidae and Moronidae showed genetic distance of 0.472 (Fig. 3). The Neighbor Joining tree showed clear division of the 19 fish families with all species of the same family formed a monophyletic group with no misplacement of species (Fig. 4). Also, the phylogenetic tree indicates the presence of five main clades that represent the 19 fish families, the first clade included species that belonged to four families: Sparidae, Platycaphalidae, Sphyraenidae, and Mullidae. The second clade harbored species from two families: Clupeidae and Synodontidae, while the third clade included species from families Carangidae, Atherindae, and Nemipteridae. The fourth clade included species from families Lethrinidae, Siganide, Mugilidae, and Tetraodontidae, while the fifth clade included species from families Serranidae, Sillaginidae, and Haemulidae. Moreover, species belonged to three families: Gerreidae, Monacanthiae, and Moronidae formed three non-clade groups.

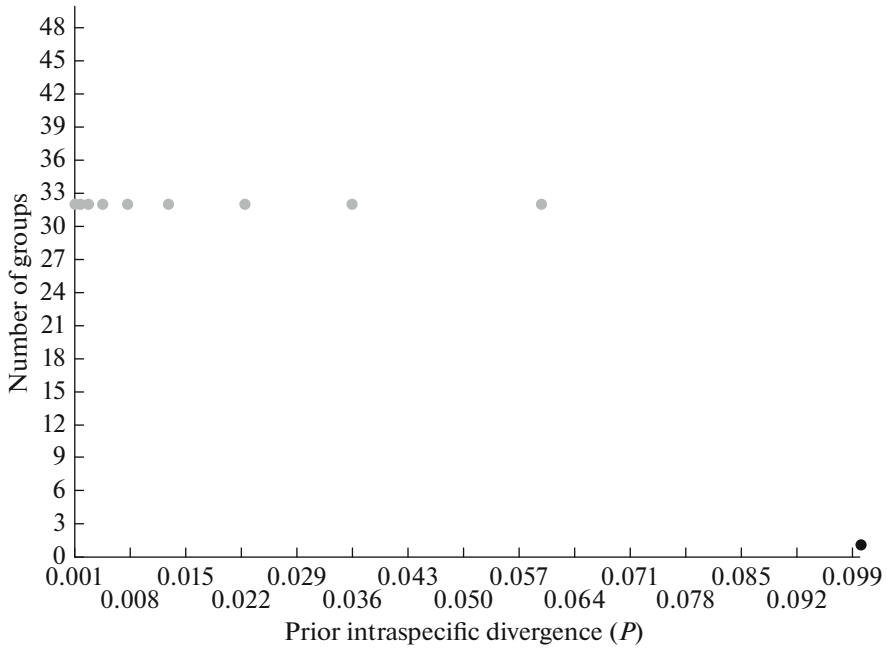

Fig. 1. Number of groups inferred from Automated Barcode Gap Discovery analysis based on prior intraspecific divergence; recrussive (●), and inetial (⚬) partition.

DISCUSSION

The herein study introduces the first inventory of the Suez Gulf marine fishes in Egypt based on DNA barcoding. DNA barcoding is proved to be a compelling tool to identify different genetic groups inhabiting similar or different habitat based on the differences between a short region of the genomic sequences (Rasmussen et al., 2009; Ward et al., 2009). The high level of congruency between morphology and DNA-based fish identifications using *COI* was documented in different fish species (Hebert et al., 2003; Kochzius et al., 2010; Landi et al., 2014; Bariche et al., 2015). In this study, 32 fish species belonging to 26 genera and 19 families of Suez Gulf, Red Sea were characterized. The universal primer amplified the target region of the species, generating 96 *COI* barcodes of 662 bp. No insertion, deletion or stop codons were found in all the sequences, supporting the hypothesis that all the amplified sequences derive from a functional mitochondrial *COI* sequences (Ward et al., 2005). The barcode sequences differentiated among the taxonomic levels of the 32 species examined. The effectiveness of species characteristics using molecular methods is assessed by its ability in quantifying the intraspecific homogeneity and interspecific heterogeneity levels (Halldén et al., 1994). The Mitochondrial *COI* gene has been reported, as an appealing and efficient barcode for species identification in differing categories of fishes, in Australian marine fishes (Ward et al., 2005), Canadian freshwater fishes (Hubert et al., 2008), ornamental fishes of North America (Steinke et al., 2009), marine fishes of India (Lakra et al., 2011); marine fishes from Japan (Zhang and Hanner, 2011); Caribbean and western vital Atlantic fishes (Weigt et al., 2012); Mediterranean Sea and Cantabric Sea fishes (Ardura et al., 2013); Red Sea fishes (Trivedi et al., 2014); alien fish species of Mediterranean Sea that was probable Red Sea origin (Bariche et al., 2015) and Egyptian freshwater fishes (Ali et al., 2020). By comparing the obtained *COI* sequences for the 32 collected species to the available data in the GenBank database via BLAST searches, the majority of the collected species (94%) showed high similarity (≥98%) when compared to their references on the GenBank database. Nevertheless, *Lethrinus* sp. and *Trachinocephalus* sp. showed lower sequence similarity (<97%) with the reference sequences on GenBank database. Both species were possibly identifiable only at genus level which is common practice in a number of previous studies (Deichmann et al., 2017; Nneji et al., 2019). The incongruous identity of the sequence at the species level not essentially be considered as a failure of the DNA barcodes to differentiate among species (Bariche et al., 2015). However, it can be regarded to

Table 2. Summary of genetic divergence within families and species based on the on Kimura two-parameter distance model

	Minimum	Maximum	Mean	SE
Within families	0.190	0.472	0.274	0.004
Within species	0.092	0.474	0.256	0.002

SE—standard error.

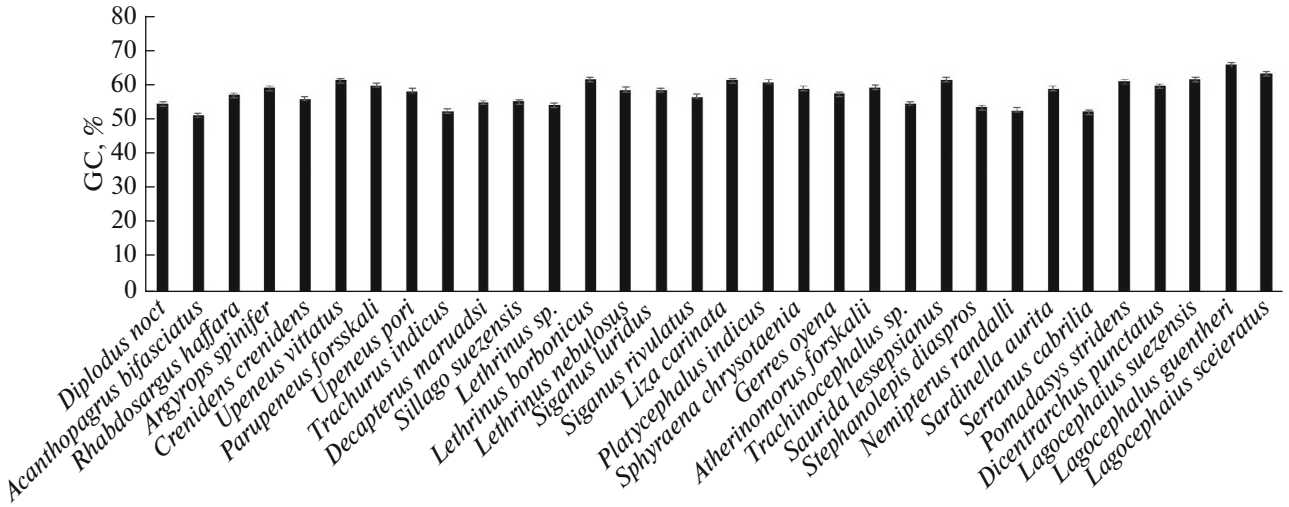

Fig. 2. Percent of total GC content of Suez Gulf fish species.

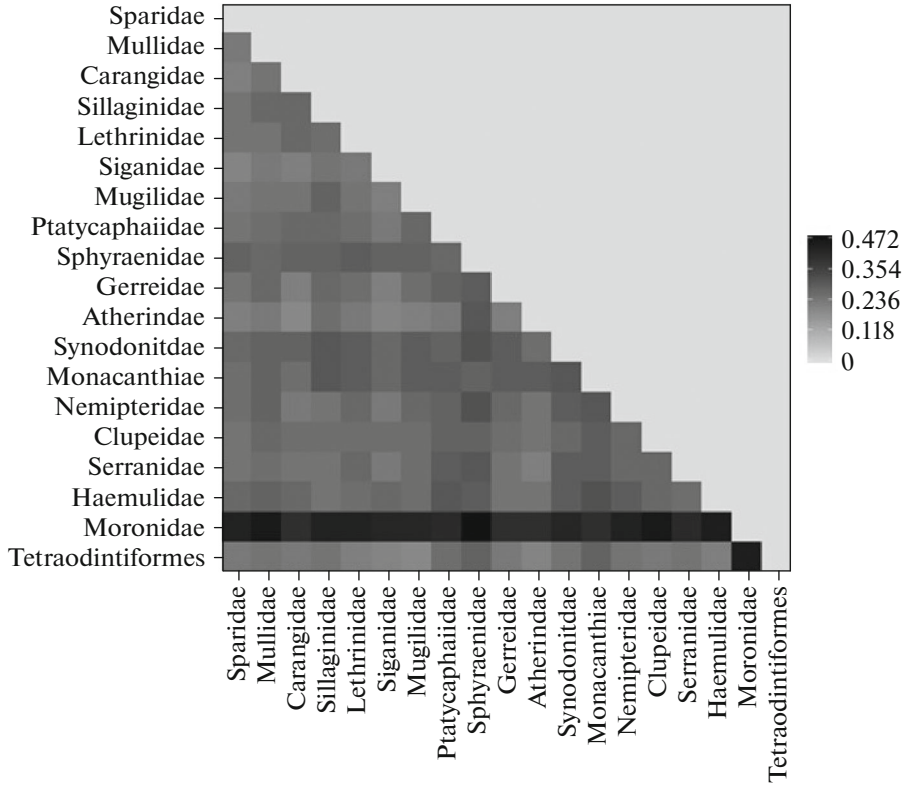

Fig. 3. Heatmap showing the distribution of pairwise genetic distances (K2P) among 19 fish families from Suez Gulf.

the lack of completely matching reference sequences on the GenBank database so they can be referred as "sp" (Hinchliff and Smith, 2014). This recommends the urgent need to an inclusive barcoding effort for providing more reference sequences to the GenBank database. The average K2P distance was higher within families (0.274) compared to (0.256) within species. The observed increase of genetic variation alongside the increase in taxonomic level, supporting a noticeable variation of genetic divergence on the species boundaries that was supported with the observation by Hubert et al., (2008). For Indian marine fishes, Larka et al. (2011) estimated the genetic divergence as 0.30% between species and 9.91% between families. Keshin

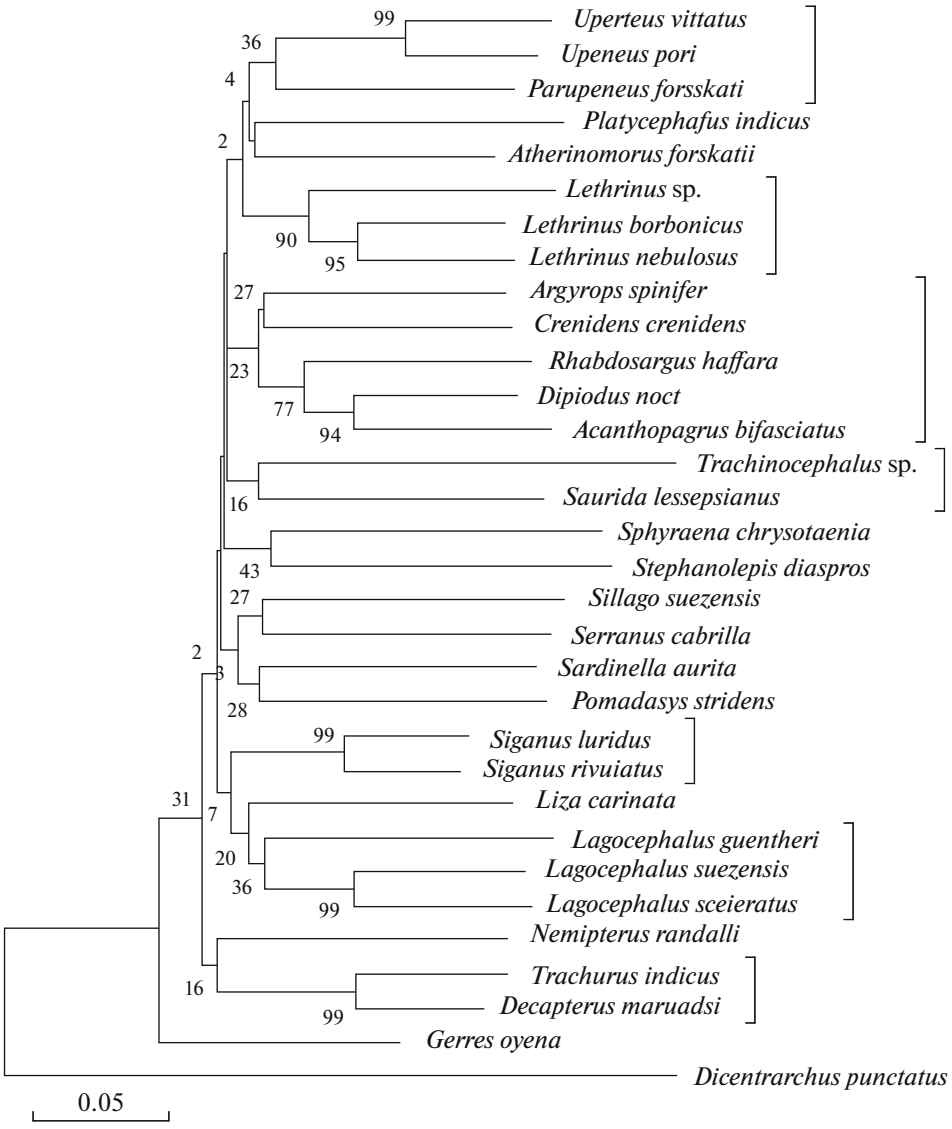

Fig. 4. Neighbor Joining tree of Suez Gulf fish species based on *COI* partial gene sequence using K2P distance, brackets represent families with more than one species. Numbers above branches are bootstrap values.

and Atar, (2013) reported that pairwise genetic distance K2P increased with increasing taxonomic level (i.e., 0.32, 9.62, 17.90, and 22.40% for conspecific, congeneric, confamilial, and between order, respectively). The barcoding gap analysis in our study indicated that there is a significant maximal distance among species indicating presence of a "barcode gap" as a delaminating principle and an indicator of species differentiation (Pappalardo et al., 2015). This result confirmed the validity and effectiveness of the applied barcode (*COI*) for species differentiation. The average percent of GC content in our sequence analysis of the 662 bp *COI* gene was estimated in *Acanthopagrus bifasciatus* as the lowest (51.74%) while the highest percentage (66.37%) obtained for *Lagocephalus guentheri*. These results corresponded reasonably well to the results found in Australian (Ward et al., 2005), Candian (Hubert et al., 2008) and Taiwanese fish species (Bingpeng et al., 2018), especially with respect to the higher GC content of the teleosts. The presence of higher GC content plays an important role in the stability of the mtDNA (Johnston and Williams, 2016), where the nucleic acids with stronger GC bonds may be less affected by spontaneous mutations and thus more protected from environmental mutagens (Samuels, 2005). Also, the observed higher level of GC content in some species (≥60%) would provide more protection of the genome where the GC-rich mtDNA are advantageous when exposed to oxidative conditions (Johnston and Williams, 2016). The phylogenetic relationship between species was revealed, where related species were grouped together in sub-clusters while

unrelated species were spread apart. Although one of the barcoding goals is to search for delineate species boundaries, however, there is truly a phylogenetic sign in *COI* gene sequence data. Congeneric species continually clustered together and, in most cases, accomplished the confamilial species (Bariche et al., 2015). When the NJ tree was examined, there was a clear clustering pattern that might be informative concerning phylogenetic relationships between conspecific and confamilial levels. Ward et al. (2005) proposed that the information collected from 655-bp sequence of a single mitochondrial gene probably used for phylogenetic study, nevertheless, is not suitable for deep phylogenetic resolution. The level of genetic divergence declined with the lower taxonomic ranks which is in line with previous reports in marine fish species (Wang et al., 2020), which probably associated with substantial genome saturation (Bingpeng et al., 2018). In the current study, most of the barcoded species are of commercial interest, whose capture is effectively influencing the marine fish production in Egypt (GAFRD, 2019). On the other hand, approximately, 31.25% (10 out of 32 species) of the collected fish species represent Lessepsian migrants. The majority of Lessepsian migrant fishes are possibly derived from the population existing in northern Red Sea (Tikochinski et al., 2013; Bariche et al., 2015). In the present study, the fishes were collected from Suez Gulf which located at the northern part of the Red Sea which can normally follow the path of the Lessepsian migration from the Red Sea to the Mediterranean Sea. These species included *Parupeneus forsskali*, *Upeneus pori, Sillago suezensis*, *Platycephalus indicus, Sphyraena chrysotaenia*, *Atherinomorus forskalii*, *Stephanolepis diaspros*, *Pomadasys stridens*, *Lagocephalus suezensis* and *Lagocephalus sceleratus* (www.fishbase.org). Endemic species in the northern Red Sea, specifically in the Suez Gulf, was conferred by Golani and Ritte (1999); Por (1978) and others. According to those authors, the Suez Gulf had many species that isolated as a result of enormous variations of temperature and sea levels throughout the glacial periods, recurrently becoming hypersaline, and as a consequently resulting in excessive endemism (Tikochinski et al., 2013). However, the record of both *Sardinella aurita* and *Serranus cabrilla* in the Suez Gulf was astonishing where, they were only distributed in Mediterranean Sea (Froese and Pauly, 2017; Golani and Fricke, 2018) which implies a pattern of anti-lessepsian migration.

CONCLUSION

The current study was the first to deal with an essential component of biodiversity in Egypt, marine fishes, which represent the broadest sector of fishes globally. The current study also focused on the molecular identification of 32 commercially important Red Sea species which included either endemic or Lessepsian migrants. The study has strongly authenticated the efficiency of *COI* gene for fish species barcoding and provides DNA barcodes for the 32 studied species to be used as references for future marine fish identification.

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COMPLIANCE WITH ETHICAL STANDARDS

Conflict of interest. The authors declare no conflict of interest.

Statement on the welfare of animals. Ethical approval did not require as the samples were collected from the commercial fisheries then transferred frozen to the laboratory.

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