BULLETIN OF THE IRAQ NATURAL HISTORY MUSEUM
Iraq Natural History Research Center & Museum, University of Baghdad
https://jnhm.uobaghdad.edu.iq/index.php/BINHM/Home
Copyright © Bulletin of the Iraq Natural History Museum Online ISSN: 2311-9799-Print ISSN: 1017-8678

Bull. Iraq nat. Hist. Mus.
(2022) 17 (2): 291-301. https://doi.org/10.26842/binhm.7.2022.17.2.0291

ORIGINAL ARTICLE

DISTRIBUTION AND PHYLOGENETIC OF FRESHWATER MUSSEL *UNIO TIGRIDIS* BOURGUIGNAT, 1852 (BIVALVIA, UNIONIDAE) FROM GREATER ZAB RIVER, IRAQ

Shwan Khursheed Bashê
Biology Department, Education College, Salahaddin University-Erbil, Erbil, Iraq
E-mail: shwan.raman@su.edu.krd

Recived Date: 27 August 2022, Accepted Date 16 November 2022, Published Date:20 December 2022

This work is licensed under a Creative Commons Attribution 4.0 International License

ABSTRACT

Freshwater mussels are a guild of stationary, suspended-feeding species; they perform significant ecological functions like nitrogen cycling, bioturbation that gives oxygen and habitat that other creatures need to survive, and increasing water clearance by filtration. Knowledge of the freshwater mussel *Unio tigridis* Bourguignat, 1852, distribution, and molecular study in Iraq was inadequate. In the current study, this species of freshwater Mussels belonging to the family Unionidae was collected from different locations in the Greater Zab River, from April to August 2022. The average water temperature of the site was arranged between (17.8 to 36.1 C°). All previous studies in the Kurdistan Region and Iraq were based on morphological characters and the current study was the first report of *Unio tigridis* that was confirmed by molecular genetics and *COI* gene, analyzed phylogenetically using Maximum Likelihood and Maximum Parsimony Methods.

Keywords: *COI*, Distribution, Phylogenetic, *Unio tigridis*, Greater Zab, Iraq

INTRODUCTION

Freshwater mussel *Unio tigridis* Bourguignat, 1852 (Mollusca: Bivalvia: Unionidae) are a guild of stationary, suspended-feeding species that play vital roles in rivers and streams (Bolotov *et al*., 2019; Hamli and Al Asif, 2021; Hamli *et al*., 2021). Its essential aquatic ecosystem constituents, which perform significant ecological functions like nitrogen cycling, bioturbation that gives oxygen and habitat that other creatures need to survive, and increasing water clearance by filtration (Zieritz *et al*., 2016; Klishko *et al*., 2017; Lopes-Lima *et al*., 2021).

The Unionidae is the richest of the six extant freshwater mussel families, there are currently 674 species known globally, with (80%) of them being widely dispersed over temperate North America and Eurasia in addition to tropical Mesoamerica, Africa, and southeast Asia (Huber, 2010, 2015). According to the International Union for Conservation of Nature's (IUCN) assessment of the majority of freshwater mussel species (517), 6% extinct, 7% vulnerable, 9%

near threatened, 10% endangered, 13% critically endangered and 37% of least concern(IUCN, 2016). Freshwater mussel descriptions from the early 19th century up till the mid-20th century included hundreds of *Unio* species described from many regions of the world (Huber, 2015).

U. tigridis Bourguignat, 1852, the most prevalent species, found from Syria to Iraq, had been evaluated by the IUCN Red List and reported as Least Concern (LC), containing an elongate solid shell buried obliquely in the mud (Plaziat and Younis, 2005). In Iraq, the first recorded of this species in Habbaniya Lake by Najim (1959), in Euphrates by Al-Bassam (2005), and in the Greater Zab River by Ali (2007). Previously, Unionid mussels' species depended on shell traits for identification before the development of molecular technology (Lopes-Lima *et al*., 2017). However, it depending alone on shell morphology found difficult, due to the morphological convergences between species and the high level of phenotypic variability within species (Klishko *et al*., 2017). Several newly released studies that used region of cytochrome oxidase subunit I gene of mitochondrial DNA (mt DNA-COI) for the biogeography and relationships of aquatic mussels in various Asiatic regions fundamentally altered our knowledge of diversity patterns (Choi, 2016; Lopes-Lima *et al*., 2020). Unfortunately, no recent investigation had been carried out on molecular phylogenetic data for any endemic Iraqi invertebrates especially freshwater mussels.

The present investigation was conducted to study the utility of a molecular technique as a DNA barcode approach for the identification, distribution, and estimate of a phylogeny of *U. tigridis*, the freshwater mussels from the Greater Zab River.

MATERIALS AND METHODS

Specimens' collection and identification

Freshwater mussels (No. 33) were collected and gathered by hand from various sites along the banks of canals, and irrigation ditches from Greater Zab River, West Erbil province, Iraq (Map 1), from April to August 2022. The water temperature was measured month wise using a temperature probe/ thermometer (HANNA instruments, HI 9811-5 for Temp., pH, EC and TDS-Romania). The collected mussel specimens brought alive and a dead shell to the laboratory. Selection has been made for the complete animals (shell not broken or eroded). The bivalves were boiled gently in 5% sodium hydroxide to be cleaned, then washed with water and dried for measurements (Abdul-Sahib and Abdul-Sahib, 2009). Morphologically, *U. tigridis* exhibit the traditional wedge-shaped shell used as diagnosed species (Lopes-Lima *et al*., 2021).

DNA extraction, COI amplification, and Sequencing

A biopsy was fixed at 99% ethanol and centrifuged 10,000 rpm for 3 minutes, then the ethanol was eliminated, the DNA was extracted using a commercial kit (Favorgen- Taiwan) in accordance with the manufacturer's instructions.

The DNA barcoding for mussel was achieved based on amplification and sequencing of the region of (mtDNA-COI) (~700 bp) (Folmer *et al.*, 1994), using forward primer LCO1490 and reverse HCO2198 primers. The PCR mixture (40 µl) contained: (a master mix, DNA template,

primers, and double-demonized water (ddH2O)). Under the following circumstances, PCR was carried out in a MJ Research, Applied Biosystem (AB) thermocycler: 94 C°/4 min; 35 cycles of 94 C°/30 sec; 48 C°/60 sec; 72 C°/90 sec, followed by one cycle of 72 C°/5 min. In order to see DNA under UV light, PCR products were separated on a 1.3% agarose gel and dyed with a safe dye (Bashê and Ali, 2019).

DNA sequencing was performed by using an ABI 3730XLs nucleotide sequence analyzer through Macrogen Inc. (Korea). The sequences were subjected to the Basic Local Alignment Search Tool for nucleotides (Blastn) implemented in the NCBI GenBank database. In addition, 11 sequences were obtained from GenBank that available on the website: http://www.ncbi.nlm.nih.gov/genbank/. All the DNA sequences were edited and aligned with (ClustalW algorithm), available in the MUSCLE program within EMBL-EBI (https://www.ebi.ac.uk/Tools/msa/muscle/).

Further estimates of *COI* variation and relationships among species by applying Maximum Likelihood (Kimura 2-parameter model) (Kimura, 1980), and using the method of Maximum Parsimony Branches associated with partitions that were replicated in fewer than 50% of bootstrap replicates are collapsed. In the bootstrap test (1000 replicates), the proportion of duplicate trees in which the linked taxa clustered together is displayed next to the branches (Felsenstein, 1985). The Tree-Bisection-Regrafting (TBR) technique was used to create the MP tree with search level 1 and the initial trees were created by randomly adding sequences (10 replicates) (Nei and Kumar, 2000). This analysis involved 12 nucleotide sequences; there were a total of 498 positions in the final dataset edited by Finch Tv program (version 1.4.0) and deposited in NCBI. Methods were conducted using MEGA X Kumar *et al.* (2018), including a sequence representative from many species currently present and available on GenBank.

Map (1): Sampling sites on the Greater Zab River, north of Iraq.

RESULTS

Mitochondrial DNA *COI* from freshwater mussels was sequenced, the results (100%) confirmed the present *U. tigridis* (Pl. 1), the shell solid and elongate lying obliquely in the mud, with its rear end extending from the bottom in pond, marshes channel, and rivers. The average temperatures from April-October 2022 were arranged (17.8-36.1 C°). The obtained sequencing result was deposited in GenBank database under the following accession numbers (OP087400) for *U. tigridis*.

Plate (1): *Unio tigridis*, collected from Greater Zab River; (A) External shell, (B) Bivalve shell, (C) Visceral parts (Scale bar=2 cm).

In Phylogeny, Kimura 2 parameter (K2P) was used for intraspecific and interspecific variations. The pairwise distances between the *Unio* are shown in Table (1). Intraspecific genetic diversity within *U. tigridis* was from zero to (0.005), within *U. Delphinus* (Reis *et al*., 2013), from zero to (0.042) and *C. elongatulus* (Araujo *et al*., 2018), was zero to (0.040). The maximum genetic distance observed *Margaritifera homsensis* was (0.228).

Table (1): Estimates of evolutionary divergence sequences between and within *U. tigridis* species (Overall standard deviation is 0.14 with standard error 0.01).

No.	Species	1	2	3	4	5	6	7	8	9	10	11
1	*U. tigridis* (current study)	0.000										
2	*U. tigridis MZ511105.1*	0.001										
3	*U. tigridis MZ511087.1*	0.005	0.001									
4	*U. delphinus* EF571442.1	0.040	0.036	0.038								
5	*U. delphinus* EF571415.1	0.042	0.038	0.040	0.001							
6	*U. elongatulus* KX399981.1	0.040	0.036	0.038	0.044	0.046						

7	*U. pictorum* EU548056.1	0.044	0.040	0.042	0.040	0.042	0.034					
8	*U. pictorum* KY930353.1	0.774	0.779	0.785	0.762	0.768	0.839	0.812				
9	*U. foucauldianus* KX399998.1	0.042	0.038	0.040	0.023	0.025	0.038	0.038	0.778			
10	*U. foucauldianus* KX400006.1	0.042	0.038	0.040	0.023	0.025	0.038	0.038	0.778	0.00		
11	*Margaritifera_homsensis* KX550090.1	0.228	0.222	0.219	0.211	0.214	0.228	0.222	0.014	0.212	0.212	

DISCUSSION

The current work was the first to analyze the biogeographic and phylogenetic of *U. tigridis* freshwater mussels from the Greater Zab River, and it might be served as a baseline study for the investigated species and for the other available mussel species as well. Our result showed that, in the both tree maximum likelihood and parsimony, that *U. tigridis* was monophyletic in the same clade with (MZ511087.1) which was reported from the far East Asia by Lopes-Lima *et al*. (2021) (Diag. 1); but this topology might be changed with the addition of more loci, also had strongly related to (MZ511105.1); Which were also related and matched to the other deposited sequences, and the outgroup *Margaritifera homsensis (*KX550090.1*)* recorded by Vikhrev *et al*. (2018) that was found more divergence species with *U. tigridis*.

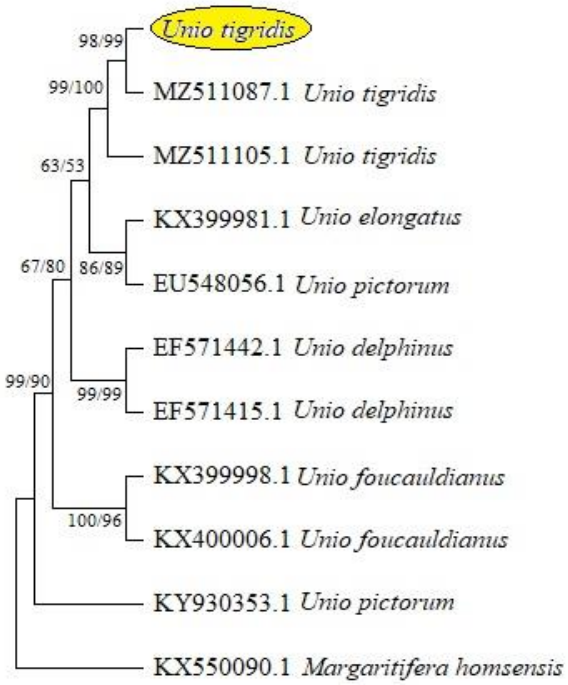

Diagram (1): Both the Maximum Likelihood (Kimura 2-parameter model) and Maximum Parsimony methods were applied to infer the evolutionary history, all nodes supported by high bootstrap (ML/MP).

According to species divergence, the *COI* gene was frequently employed as a barcode since it has been subject of the most research and is considered as a standard for analyzing divergence among Unionidae species (Araujo *et al*., 2018) (Tab. 1). Notably, in the phylogenetic tree of *U. tigridis*, the rebuilt species tree coincides with the general topology of the concatenated matrix studies.

DNA barcoding provided a remarkably valuable approach for confirming the identity of a species (Davison *et al*., 2009). The sequenced data presented in the current study were derived from the *COI* gene; it was currently one of the most widely used for phylogeny, systematics, and species identification. At present, there were many reference DNA barcode sequences belonging to Gastropod species available in GenBank which could be used for identification purposes. To date, there has been no study in Iraq regarding the molecular and phylogenetic of this investigated species. Nevertheless, the conclusions of this single-locus or multilocus DNA species delimitation algorithms could not be directly converted into new taxonomies. The Unionidae have frequently been recovered as paraphyletic by phylogenetic analysis throughout the last decade (Graf and Cummings, 2006). However, their findings were unsubstantiated and based on insufficient taxon and character sampling (Whelan *et al*., 2011); only two molecular character sets had been used to assess Unionoida family-group level associations through COI gene (Whelan *et al*., 2011), Other Unionidae phylogenetic analysis had relied on specific gene region. While, Lopes-Lima *et al*. (2017) hypothesized the subfamily's evolutionary connection based on COI.

The worldwide distribution and diversity of mussels species is to several reasons considering; habitat destruction, degradation, and alteration caused by increasing populations of human, industrialization, and changes in land use in addition to spread of non-native exotic species (Zieritz et al., 2016 - Hamli et al., 2021), enrichment water with nutrients resulting in phytoplankton and macrophyte blooms (Sharip and Zakaria, 2007), and heavy metal concentrations often reach deadly thresholds for freshwater mussels (Nobles and Zhang, 2015). According to the present findings, water temperatures fluctuated ranges between (17.8 to 36.1 C°), this result was similar to Payton *et al*. (2016) in USA, which recorded highest temperatures in summer average (35 to 40 C°) during particularly hot weather, which was near or exceeds the high temperature tolerant ranges of several aquatic organisms, and substantial changes in temperature might had an impact on the balance of ecosystem. That results in species-specific variations in sensitivity to factors including physiological tolerance, limited adaptability to abiotic stresses, and life history aspects. In another way, observed an increase of only ~3°C above normal water temperatures was enough to cause 33% mortality.

CONCLUSIONS

In the current study, the first report based on molecular technique *COI* for confirmation diagnosis of *Unio tigridis*, distribution along Greater Zab River, and phylogenetic methods (Maximum Likelihood and Maximum Parsimony) with other species.

AKNOWLEDGMENTS

I wish to thanks Dr. Rebwar Hamasalih, Ph. D. in Molecular Biology, for his valuable help and to Biology department for facilitating way for conducting this research.

CONFLICT OF INTEREST STATMENT

"The author has no conflict of interest to declare."

LITERATURE CITED

Abdul-Sahib, I. and Abdul-Sahib, E. 2009. A new record of the freshwater clam, Anodonta vescoiana Bourguignat, 1857 (Mollusca: Bivalvia) from Al-Ezz River, Iraqi marshes. *Mesopotamian Journal of Marine Science*, 24 (1): 7-12. [Click here]

Al-Bassam, K. S. 2005. Strontium content and Na/Ca ratio in recent mollusk shells as salinity indicators in fluvial water systems. *Iraqi Bulletin of Geology and Mining*, 1 (1): 21-33. [Click here]

Ali, L. 2007. A study of macroinvertebrates community in the middle sector of Greater Zab River, Iraq. Ph. D. Dissertation, Department of Biology, College of Science for Women, University of Baghdad, Iraq, 123 pp. [Click here]

Araujo, R., Buckley, D., Nagel, K.-O., García-Jiménez, R. and Machordom, A. 2018. Species boundaries, geographic distribution and evolutionary history of the Western Palaearctic freshwater mussels *Unio* (Bivalvia: Unionidae). *Zoological Journal of the Linnean Society*, 182 (2): 275-299. [CrossRef]

Bashê, S. and Ali, L. 2019. First molecular identification and phylogenetic tree of *Petasiger exaeretus* Dietz, 1909 (Digenea: Echinostomatidae) from an intermediate host *Radix auricularia* (L., 1758) in Greater Zab River, Iraq. *Annals of Agricultural and Environmental Medicine*, 26 (3): 504-507. [CrossRef]

Bolotov, I. N., Konopleva, E. S., Vikhrev, I. V., Lopes-Lima, M., Bogan, A. E., Lunn, Z., Chan, N., Win, T., Aksenova, O. V. and Gofarov, M. Y. 2019. Eight new freshwater mussels (Unionidae) from tropical Asia. *Scientific Reports*, 9 (1): 1-15. [CrossRef]

Choi, S. C. 2016. Methods for delimiting species via population genetics and phylogenetics using genotype data. *Genes and Genomics*, 38 (10): 905-915. [CrossRef]

Davison, A., Blackie, R. L. and Scothern, G. P. 2009. DNA barcoding of Stylommatophoran land snails: a test of existing sequences. *Molecular Ecology Resources*, 9 (4): 1092-1101.[CrossRef]

Felsenstein, J. 1985. Confidence limits on phylogenies: An approach using the bootstrap. *Evolution*, 39: 783-791. [CrossRef]

Folmer, O., Black, M., Hoeh, W., Lutz, R. and Vrijenhoek, R. 1994. DNA primers for amplification of mitochondrial Cytochrome C Oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology*, 3 (5): 294–299. [Click here]

Graf, D. L. and Cummings, K. S. 2006. Palaeoheterodont diversity (Mollusca: Trigonioida+ Unionoida): what we know and what we wish we knew about freshwater mussel evolution. *Zoological Journal of the Linnean Society*, 148 (3): 343-394. [CrossRef]

Hamli, H. and Al Asif, A. 2021. Trends of bivalve (Bivalvia) research in East Malaysia: a systematic review. *Asian-Australasian Journal of Bioscience and Biotechnology*, 6 (2): 50-59.[CrossRef]

Hamli, H., Kamal, A. H. M., Idris, M. H., Gerusu, G. J., Ismail, J. and Abualreesh, M. H. 2021. Bivalves (Mollusca: Bivalvia) in Malaysian Borneo: status and threats. *Journal of Threatened Taxa*, 13 (11): 19553-19565. [CrossRef]

Huber, M. 2010. Compendium of bivalves. A full-color guide to 3,300 of the world's marine bivalves. A status on Bivalvia after 250 years of research. Hackenheim D: ConchBooks, 901 pp.[Click here]

Huber, M. 2015. Compendium of bivalves 2. A full-color guide to the remaining seven families. A systematic listing of 8'500 bivalve species and 10'500 synonyms. ConchBooks, 907 pp. [Click Here]

IUCN 2016. IUCN red list of threatened species. secondary IUCN Red List of Threatened Species. [Click here]

Kimura, M. 1980. A simple method for estimating evolutionary rates of base substitutions through comparative studies of nucleotide sequences. *Journal of Molecular Evolution*, 16 (2): 111-120. [CrossRef]

Klishko, O., Lopes-Lima, M., Froufe, E., Bogan, A., Vasiliev, L. and Yanovich, L. 2017. Taxonomic reassessment of the freshwater mussel genus *Unio* (Bivalvia: Unionidae) in Russia and Ukraine based on morphological and molecular data. *Zootaxa*, 4286 (1): 93-112. [CrossRef]

Kumar, S., Stecher, G., Li, M., Knyaz, C. and Tamura, K. 2018. MEGA X: Molecular evolutionary genetics analysis across computing platforms. *Molecular Biology and Evolution*, 35 (6): 1547.[CrossRef]

Lopes-Lima, M., Froufe, E., Ghamizi, M., Mock, K. E., Kebapçı, Ü., Klishko, O., Kovitvadhi, S., Kovitvadhi, U., Paulo, O. S., Pfeiffer III, J. M., Raley, M., Riccardi, N., Şereflişan, H., Sousa, R., Teixeira, A., Varand, S., Zanatta, D. T., Zieritz, A. and Bogan, A. E.

2017. Phylogeny of the most species-rich freshwater bivalve family (Bivalvia: Unionida: Unionidae): Defining modern subfamilies and tribes. *Molecular Phylogenetics and Evolution*, 106: 174-191. [CrossRef]

Lopes-Lima, M., Gürlek, M. E., Kebapçı, Ü., Şereflişan, H., Yanık, T., Mirzajani, A., Neubert, E., Prié, V., Teixeira, A., Gomes-dos-Santos, A., Barros-García, D., Bolotov, I. N., Kondakov, A. V., Vikhrev, I. V., Tomilova, A. A., Özcan, T., Altun, A., Gonçalves, D. V., Bogan, A. E. and Froufe, E. 2021. Diversity, biogeography, evolutionary relationships, and conservation of Eastern Mediterranean freshwater mussels (Bivalvia: Unionidae). *Molecular Phylogenetics and Evolution*, 163: 107261.[CrossRef]

Lopes-Lima, M., Hattori, A., Kondo, T., Lee, J. H., Kim, S. K., Shirai, A., Hayashi, H., Usui, T., Sakuma, K., Toriya, T., Sunamura, Y., Ishikawa, H., Hoshino, N., Kusano, Y., Kumaki, H., Utsugi, Y., Yabe, S., Yoshinari, Y., Hiruma, H., Tanaka, A., Sao, K., Ueda, T., Sano, I., Miyazaki, J.-I., Gonçalves, D. V., Klishko, O. K., Konopleva, E. S., Vikhrev, I. V., Kondakov, A. V., Gofarov, M. Y., Bolotov, I. N., Sayenko, E. M., Soroka, M., Zieritz, A., Bogan, A. E. and Froufe, E. 2020. Freshwater mussels (Bivalvia: Unionidae) from the rising sun (Far East Asia): phylogeny, systematics, and distribution. *Molecular Phylogenetics and Evolution*, 146: 106755. [CrossRef]

Najim, A. T. 1959. Notes on the distribution of some molluscs in Iraq. *Journal of Molluscan Studies*, 33 (4): 159-163. [CrossRef]

Nei, M. and Kumar, S. 2000. Molecular evolution and phylogenetics, Oxford University Press, 352 pp. [Click here]

Nobles, T. and Zhang, Y. 2015. Survival, growth and condition of freshwater mussels: Effects of municipal wastewater effluent. *PloS one*, 10 (6): e0128488. [CrossRef]

Payton, S. L., Johnson, P. D. and Jenny, M. J. 2016. Comparative physiological, biochemical and molecular thermal stress response profiles for two unionid freshwater mussel species. *Journal of Experimental Biology*, 219 (22): 3562-3574. [CrossRef]

Plaziat, J.-C. and Younis, W. R. 2005. The modern environments of Molluscs in southern Mesopotamia, Iraq: A guide to paleogeographical reconstructions of Quaternary fluvial, palustrine and marine deposits. *Carnets de géologie*, (A01): 1-18. [CrossRef]

Reis, J., Machordom, A. and Araujo, R. 2013. Morphological and molecular diversity of Unionidae (Mollusca, Bivalvia) from Portugal. *Graellsia*, 69 (1): 17-36. [CrossRef]

Sharip, Z. and Zakaria, S. 2007. Lakes and reservoir in Malaysia: management and research challenges. Proceedings of Taal 2007: The 12th world lake conference, p. 1349-1355. [Researchgate]

Vikhrev, I. V., Bolotov, I. N., Altun, A., Gofarov, M. Y., Dvoryankin, G. A., Kondakov, A. V., Ozcan, T. and Ozcan, G. 2018. The revenant: rediscovery of Margaritifera homsensis from Orontes drainage with remarks on its taxonomic status and conservation (Bivalvia: Margaritiferidae). *Systematics and Biodiversity*, 16 (1): 69-80. [CrossRef]

Whelan, N. V., Geneva, A. J. and Graf, D. L. 2011. Molecular phylogenetic analysis of tropical freshwater mussels (Mollusca: Bivalvia: Unionoida) resolves the position of Coelatura and supports a monophyletic Unionidae. *Molecular Phylogenetics and Evolution*, 61 (2): 504-514.[CrossRef]

Zieritz, A., Lopes-Lima, M., Bogan, A. E., Sousa, R., Walton, S., Rahim, K. A. A., Wilson, J.-J., Ng, P.-Y., Froufe, E. and McGowan, S. 2016. Factors driving changes in freshwater mussel (Bivalvia, Unionida) diversity and distribution in Peninsular Malaysia. *Science of the Total Environment*, 571: 1069-1078.[CrossRef]

BULLETIN OF THE IRAQ NATURAL HISTORY MUSEUM

Bashê, S. K.

Bull. Iraq nat. Hist. Mus.
(2022) 17 (2): 291-301.

التوزيع و العلاقة النشوئية لبلح المياه العذبة *UNIO TIGRIDIS* BOURGUIGNAT, 1852 (BIVALVIA, UNIONIDAE) من نهر الزاب الكبير، العراق

شوان خورشيد باشىَ

قسم علوم الحياة، كلية التربية، جامعة صلاح الدين-أربيل، أربيل، العراق.

تأريخ الاستلام: 2022/8/27، تأريخ القبول: 2022/11/16، تأريخ النشر: 2022/12/20

الخلاصة

بلح البحر عبارة عن طائفة من الأنواع الثابتة والمعلقة التي تتغذى في المياه العذبة، التي تؤدي وظائف بيئية مهمة مثل دورة النيتروجين ، والاضطراب الحيوي الذي يمنح الأكسجين والموائل التي تحتاجها الكائنات الأخرى للبقاء على قيد الحياة ، وزيادة تصفية المياه عن طريق الترشيح. كانت المعرفة لنوع بلح المياه العذبة *Unio tigridis* Bourguignat, 1852، التوزيع، والدراسة الجزيئية في العراق غير كافية لتحديد النوع.

جمعت عينات هذا النوع من بلح البحر في الدراسة الحالية، والذي ينتمي إلى عائلة Unionidae في المياه العذبة لعدد من المواقع المختلفة في نهر الزاب الأكبر، من شهر نيسان 2022 إلى آب 2022. كانت متوسط درجة حرارة الماء في الموقع مابين 17.8 إلى 36.1م°.

استندت جميع الدراسات السابقة في إقليم كوردستان والعراق إلى الصفات المظهرية، في حين ان الدراسة الحالية كانت أول تقرير عن تشخيص النوع *U. tigridis* استنادا الى الوراثة الجزيئية و جين COI ، حيث تم تحليل النشوء والتطور باستخدام طرق الحد الأقصى من الاحتمالية والحد الأقصى من Parsimony.