





Iraq, there is no studies were done to re-classify the snails at molecular level. *Limax flavus* was founded in different parts in the world, but it's the first time was recorded in Iraq. This work was described *Limax flavus* genus as new recorded in Iraq. Morphological and genetic studies were done to confirm the classification of this genus. Descriptions of species in the majority of slug studies are depended on one individual or a small series of specimens. This fact hinders the estimation of inter- and intraspecific variations presented in these characters (Nitz *et al.* 2009). The apparent lack of diagnostic characters of external morphology, such as a well-developed shell is one of the major problems in slug research. The shape, body size, vestigial shell and coloration are all very variable and potentially misleading (Klee *et al.* 2007). Nearly all species of *Limax* genus are poorly known but there are some poorly studies on molluscans in Iraq (Shehab & et al 2015, Ameen 2018), and much historical identification is doubtful. Variable coloration was found in *Limax* species, ranging from creamy white through brownish to black. The long of living animal was up to 19.6 cm; sole length up to 19 cm (16.7 cm in ethanol), mantle length up to 5.8 cm (5.4 cm in ethanol); width up to 2.2 cm (up to 1.7 cm in ethanol), keel length in ethanol up to 4.6 cm. Discrimination of *Limax* species cannot be based on one or two morphological character sets alone, therefore the various characters value for discrimination and identification must be considered (Nitz *et al.* 2009). New possibilities of dealing with taxonomic problems were offered by using the application of molecular biology methods (Davis 1994). Using of molecular techniques to analyze relationships between populations and species has become widespread, and nucleotide sequence variations in the mtDNA offer a powerful tool in molecular phylogenetic of marine organisms (Carvalho & Pitcher 1995). MtDNA markers have been widely used for population genetic structure analyses (Kocher *et al.* 1989). The use of molecular biology methods in the studies of population genetics and evolution has increased dramatically during the last decade (Tripath *et al.* 2013). There is little information available on the amplification success of the *COI* gene fragment in gastropod tissues (Skujiene & Soroka 2003). The molecular tree study based on *COI* gene sequence data strongly supports the results based on morphology and behavior (Nitz *et al.*, 2009). Currently, one of the most widely used genes for phylogeny, systematics and the identification of species is *COI* (Huelsken *et al.* 2011). Nucleotide sequences of nuclear and MtDNA analyses were provides us a basis for taxonomic and phylogenetic considerations (Skujiene & Soroka 2003). The DNA sequences of cytochrome c oxidase subunit I (*COI*) and barcode gene, may serve as additional valuable character set for phylogenetic analyses and subsequent identification (Nitz *et al.* 2009). This study showed some differences in nucleotide sequences in comparison with the same nucleotide sequences for *Limax flavus* which published in NCBI data. Two primary evolutionary mechanisms that cause population differentiation are genetic draft and natural selection (Hufford & Mazer 2003). Development of ecological adaptation or ecotype will resulted by natural selection (Awodiran & Ogunjobi 2016). Also, these two forces may interact with other factors, such as breeding system, life-history traits, dispersal and other evolutionary and ecological processes to determine the genetic structuring patterns that are observed in the field (Gow *et al.* 2004). This

study revealed a good similarity with *Limax flavus* specimen published in NCBI data and this help us for confirming that the collected genus is *Limax flavus*.

## V. CONCLUSIONS

In conclusion, the collected specimen species was the first record in Iraqi environment and the sequences results confirms that this specimen is *Limax flavus*. *COI* gene sequence in this work was deposited in the NCBI gene bank repository ([www.ncbi.nlm.nih.gov/nucleotide/MF034732](http://www.ncbi.nlm.nih.gov/nucleotide/MF034732)).

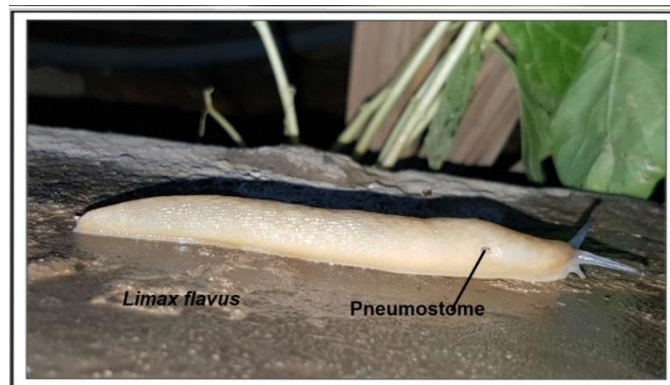
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TABLE I: PRIMERS FOR COI GENE AMPLIFICATION

| Primer      | Sequence (5' → 3')               |
|-------------|----------------------------------|
| LCO1490 (F) | 5'-GGTCAACAAATCATAAAGATATTGG-3'  |
| HCO2198 (R) | 5'-TAAACTTCAGGGTGACCAAAAAATCA-3' |

Fig. 1: External feature of the collected *Limax flavus* (L., 1758)

