

Rapid Communication**First record of the red swamp crayfish *Procambarus clarkii* (Girard, 1852) (Decapoda: Astacidea: Cambaridae) in Tunisia**Mohamed Wajih Bouaoud^{1,*}, Faouzia Charfi-Cheikhrouha¹ and Mohamed El Gtari^{1,2}¹University of Tunis El Manar, Faculty of Sciences, Department of Biology, LR18ES 06 Diversity, Management and Conservation of Biological Systems, 2092 Manar II, Tunisia²University of Carthage, Higher Institute of Fisheries and Aquaculture of Bizerte, BP 15, 7080, Menzel Jemil, TunisiaAuthor e-mails: mohamedwajih@yahoo.fr (MWB), facharfi@yahoo.fr (FCC), elgtari.mohamed@gmail.com (MEG)

*Corresponding author

Citation: Bouaoud MW, Charfi-Cheikhrouha F, El Gtari M (2020) First record of the red swamp crayfish *Procambarus clarkii* (Girard, 1852) (Decapoda: Astacidea: Cambaridae) in Tunisia. *BioInvasions Records* 9(2): 349–356, <https://doi.org/10.3391/bir.2020.9.2.20>

Received: 24 November 2018**Accepted:** 28 June 2019**Published:** 20 April 2020**Handling editor:** Christoph Chucholl**Thematic editor:** David Wong**Copyright:** © Bouaoud et al.

This is an open access article distributed under terms of the Creative Commons Attribution License (Attribution 4.0 International - CC BY 4.0).

OPEN ACCESS**Abstract**

An investigation of artificial water ecosystems at the Cap Bon, in north-eastern Tunisia, led to the first record of the invasive alien freshwater species *Procambarus clarkii* in Tunisia. Its distribution in hill lakes, dams and streams in this area was investigated and some parameters of Gombare Lake population were studied from December 2017 to March 2018. During the four monthly samplings in Gombare Lake, 116 specimens were collected, sexed and measured. Preliminary results on the sampled population were reported. The introduction and potential impacts of *P. clarkii* in Tunisia are discussed and management recommendations are given.

Key words: invasive species, Cap Bon, Lebna dam, Gombare lake, Guitoune lake**Introduction**

Procambarus clarkii (Girard, 1852), known as Louisiana red crayfish or red swamp crayfish, is a freshwater decapod species native to northern Mexico (Hernandez et al. 2008), southern and southeastern United States (Hobbs H, Jass JP, huner JV 1989; Quan et al. 2014). This species is considered, according to the Convention on Biological Diversity (CBD2002) and the World Conservation Union, as an Invasive Alien Species.

P. clarkii has been assessed as Least Concern in the IUCN Red List of Threatened species. Being highly invasive, this species is not impacted upon by any threats (IUCN Red List 2010). Because of its commercial aquaculture success in its native area, *P. clarkii* has been introduced in many countries all over the world (Hobbs et al. 1989; Liu et al. 2011). It is also widely distributed in Asia, especially in China where it is now found in almost all types of freshwater bodies, including lakes, rivers and even paddy fields (Yanhe et al. 2015).

In Europe, the first introduction of *P. clarkii* was into Spain in 1973 for aquaculture purposes (Habsburgo Lorena and Laurent 1979). Then the species was introduced eastward across the continent, invading a wide range

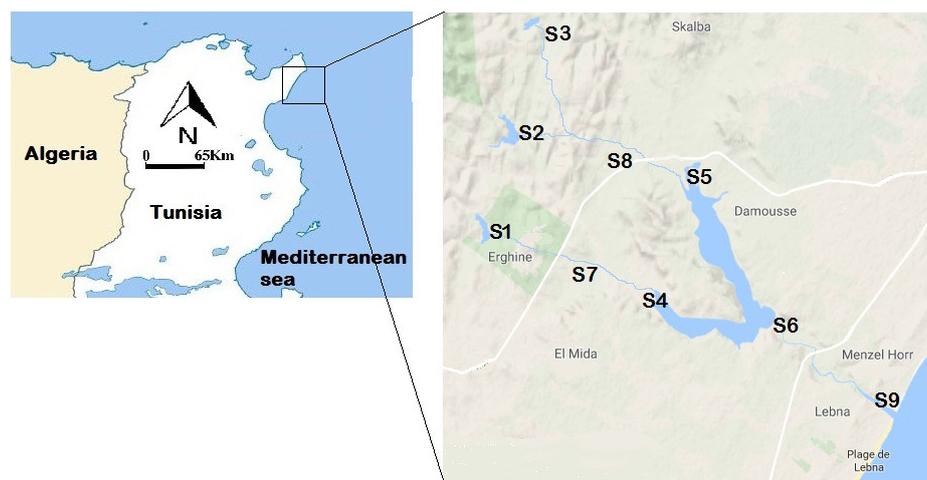


Figure 1. Study area: Location of Tunisia and sampling region (left); Sampling stations in Cap Bon locality (right); S1: Gombare hill Lake, S2: Soudane hill Lake, S3: Guitoune hill Lake; S4: Mouth of Wadi Boudokhane; S5: Mouth of Wadi El Wediane; S6: Southern east bank; S7: Wadi Boudokhane; S8: Wadi El Wediane; S9: Wadi Lebna.

of water courses and dams, lakes and marshy areas (Gherardi 2006; Kouba et al. 2014; Souty-Grosset et al. 2016). Since then, several records of *P. clarkii* have been reported in Central and Southern European countries (Souty-Grosset et al. 2016). According to Gherardi (2006), *P. clarkii* has become the dominant macroinvertebrate in several European countries.

In South Mediterranean countries, *P. clarkii* was recorded in Egypt (El Zein 2005), in Israel (Wizen et al. 2008), in Merja Zerga, Morocco, late 2008–early 2009 (El Qoraychy et al. 2015), and most recently in Malta (Vella et al. 2017).

In this paper, we report the first record of *P. clarkii* in fresh waters from north-eastern Tunisia where no native crayfish occur.

Materials and methods

Study site

The first specimen of the species was accidentally discovered at the Gombare Lake in 2014 by the first author, but no scientific work was performed until the end of 2017. A field survey was only conducted in 2017–2018, during one day per month from December 3rd to March 31st in the freshwater ecosystems including hill lakes, dams and streams. The study area (Figure 1) comprises three hill lakes: Gombare (22 ha; 36°47.433'N; 10°50.012'E), Soudane (6 ha; 36°49.352'N; 10°50.448'E) and Guitoune (20 ha; 36°50.187'N; 10°50.447'E), which are connected to the Lebna dam (750 ha; 36°44.694'N; 10°52.940'E) by two watercourses, the Boudokhane connecting Gombare Lake and Lebna dam spreading into the south bank of Lebna dam (at 36°44.694'N; 10°52.940'E), and El Wediane connecting Soudane and Guitoune Lakes to the north bank of the Lebna dam (at 36°46.505'N; 10°53.710'E). Lebna dam enters the sea through the Lebna wadi. The study site is an agricultural area belonging both to the sub-humid and the semi-arid bioclimatic zone (Smaoui 2015) with an average of 400 mm rainfall per year.



Figure 2. Japanese trap for decapods used in the present study.

Sampling

Sampling was carried out in each hill lake, i.e. Gombare (S1), Soudane (S2) and Guitoune (S3). In Lebna dam, three stations were selected: the mouth of the two wadis, Boudokhane (S4) and El Wediane (S5), and the south east bank (S6). Three stations, S7, S8 and S9 were selected along Boudokhane, El Wediane and Lebna wadi, respectively (Supplementary material Table S1).

Specimens were captured at night when crayfish were more active using Japanese traps (Figure 2) for decapods baited by chicken meat. Traps were set overnight to avoid catching water turtles. In addition, all information pointing to the presence of crayfish at a sampling site, such as dead individuals and presence of burrows, was reported.

Laboratory work

Captured specimens were morphologically identified according to Morehouse and Tobler (2013). They were sexed according to the presence of modified pleopods in males as copulatory organs (Cilenti et al. 2017).

For all specimens the following morphological parameters were measured using a Vernier caliper (accuracy ± 1 mm) (Figure 3): total length (TL: the distance between the apex of the rostrum to the posterior margin of the pleon, excluding the telson), cephalothorax length (CL: distance from the apex of the rostrum to the mid-dorsal posterior margin of the carapace) and cheliped length of the fixed claw (ChL: distance between the tip of dactylus and the basis of propodus).

Average values with their standard deviation of the different measures, TL, CL and ChL, were calculated using Microsoft Excel 2010. Differences in



Figure 3. *Procambarus clarkii* (Dorsal view); TL: Total length, CL: Cephalothorax length.

Table 1. Specimens of *Procambarus clarkii* captured at Gombare Lake during four sampling surveys.

Date	03/12/2017	28/01/2018	08/02/2018	21/03/2018	Total
Specimens captured	22	34	30	30	116
Males	10	14	19	13	58
Females	12	20	11	17	58

these traits between males and females were assessed with t-tests using R software (v.3.5.3).

Ten specimens from Gombare Lake were preserved in 100% ethanol for further molecular studies and deposited at the Laboratory “Diversity, Management and Conservation of Biological Systems” Sciences Faculty of Tunis, University Tunis El Manar.

Results

All crayfish specimens captured in the sampling area were identified as *P. clarkii*. The species was not detected in Soudane Lake, Boudokhane wadi, El Wediane wadi and Lebna wadi but dead specimens occurred at Guitoune Lake (three specimens) and Lebna dam (two specimens) (Table S1). Burrows were detected at Gombare Lake, Guitoune Lake and Lebna dam. An abundant population of *P. clarkii* was sampled at Gombare Lake. At this station, a total of 116 specimens were captured through four sessions (22, 34, 30 and 30 specimens per sampling occasion, respectively) (Table 1). Table 2 summarizes the three morphological parameters measured (TL, CL and ChL) in total and for males and females, respectively, as well as standard deviation, t-test statistic (t) and p value. T-tests revealed no significant differences between TL and CL of males and females ($p > 0.07$), whereas males had significantly larger chelae than females ($p \leq 0.001$).

Table 2. TL, CL and ChL (in mm) of total specimens, males and females, and results of t-tests.

	Total Length TL (mm)			Cephalothorax Length CL (mm)			Cheliped Length ChL (mm)			
	Total	Males	Females	Total	Males	Females	Total	Males	Females	
Min	58	58	58	14	36	14	20	21	20	
Max	97	97	90	56	56	56	53	53	47	
Average	73	71.7	74.2	43.2	43.3	43.2	34.3	38.2	30.3	
Standard deviation	7.5	7	7.8	4.9	3.7	5.9	7.3	6.6	5.6	
t		-1.817			0.109			6.951		
Degree of freedom Df		114			114			114		
P value		0.072			0.913			≤ 0.001		


Figure 4. Burrows of *P. clarkii* at the Lebna dam at the mouth of the Boudokhane Wadi.

Burrows were found in clay soil at Lebna dam at the mouth of the two wadis, Boudokhane and El Wediane (Figure 4).

A survey conducted among three aquarium traders in Tunis revealed that *P. clarkii* is a popular sold species and imported as part of the aquarium industry.

Discussion

Our preliminary findings prove the presence of *Procambarus clarkii* in a northeastern region of Tunisia; this finding presents the first record of this invasive species in this country. Furthermore, genetic analyzes will be performed to determine the provenance of this species.

The presence of *P. clarkii* burrows at the mouth of the two wadis, Boudokhane and El Wediane connecting Lebna dam, is probably due to its spread from Gombare Lake and Guitoune Lake. Maybe is it the beginning of the colonization of these areas? To confirm this hypothesis and to identify the source of this alien species in these areas, molecular genetic analyses are needed.

However, for Guitoune Lake and the Lebna dam, the very small number of *P. clarkii* captured can be explained by the presence of its two predators, *Micropterus salmoides* (Lacépède, 1802) and *Sander lucioperca* (Linnaeus, 1758), trapped in recreational fishing by the first author. The absence of these two predators at Gombare Lake may explain the abundance of *P. clarkii* in this lake.

According to Loureiro et al. (2015), the impressive ability of this species to successfully colonize a wide range of environments is a consequence of its behavioral and biological characteristics. Indeed, biological characteristics of *P. clarkii*, such as early maturity, rapid growth rates, high numbers of offspring (Paglianti and Gherardi 2004) and life cycle plasticity (Gutiérrez-Yurrita and Montes 1999; Gutiérrez-Yurrita et al. 1999; Gherardi et al. 2000) allow this species to invade different environments. In addition, this species is able to withstand extreme environmental conditions related to its burrowing activity (Huner and Barr 1984). The latter is the most negative impact caused by its presence in Egyptian Nile waters, leading to the weakening of small dikes, dikes and aquaculture ponds, erosion along streams and flooded fields (Khalil et al. 2015). Similar damage to agricultural fields as well as holes and collapse banks are also reported in Italy (Barbaresi et al. 2004). The burrowing activity of *P. clarkii*, once introduced, increases the turbidity of the water, thus reducing primary production (Rodríguez et al. 2003). Gherardi (2006) has demonstrated the negative impact of this species on macrophytes and invertebrates in Lago della Doccia (Italy) and adverse impacts on molluscs and vertebrates, including amphibian larvae, have been also reported (Gherardi et al. 2001; Renai and Gherardi 2004).

Thus, the introduction of *P. clarkii* into Tunisian fresh waters is a major concern. We suggest that prompt action should be taken to eradicate this species, as it is still confined in Gombare Lake, before being too late. It is also necessary to implement legislation prohibiting the transport of live crayfish and immediate destruction if caught, as happened in France (Vigneux et al. 2002).

Moreover, stringent controls should be conducted on aquarium traders to prevent the introduction of *P. clarkii* into other freshwater habitats.

Acknowledgements

The authors are grateful to Catherine Souty-Grosset and anonymous referees for their comments that improved the manuscript. The research was supported by the laboratory “Diversity, Management and Conservation of Biological Systems”, Faculty of Sciences of Tunis.

References

- Barbaresi S, Tricarico E, Gherardi F (2004) Factors inducing the intense burrowing activity of the red-swamp crayfish, *Procambarus clarkii*, an invasive species. *Naturwissenschaften* 91: 342–345, <https://doi.org/10.1007/s00114-004-0533-9>
- CBD (2002) Convention on Biological Diversity (CBD). <https://www.cbd.int> (accessed 8 November 2018)

- Cilenti L, Alfonso G, Gargiulo M, Chetta FS, Liparoto A, D'Adamo R, Mancinelli G (2017) First records of the crayfish *Procambarus clarkii* (Girard, 1852) (Decapoda, Cambaridae) in Lake Varano and in the Salento Peninsula (Puglia region, SE Italy), with review of the current status in southern Italy. *BioInvasions Records* 6: 153–158, <https://doi.org/10.3391/bir.2017.6.2.11>
- El Qoraychy I, Fekhaoui M, El Abidi A, Yahyaoui A (2015) Biometry and demography of *Procambarus clarkii* in Rharb Region, Morocco. *AACL Bioflux* 8(5): 751–760
- El Zein G (2005) Introduction and impact of the crayfish *Procambarus clarkii* in the Egyptian Nile. *L'Astaciculteur de France* 84: 1–12
- Gherardi F (2006) Crayfish invading Europe: the case study of *Procambarus clarkii*. *Marine and Freshwater Behaviour and Physiology* 39: 175–191, <https://doi.org/10.1080/10236240600869702>
- Gherardi F, Barbaresi S, Salvi G (2000) Spatial and temporal patterns in the movement of the red swamp crayfish, *Procambarus clarkii*, an invasive crayfish. *Aquatic Sciences* 62: 179–193, <https://doi.org/10.1007/PL00001330>
- Gherardi F, Renai B, Corti C (2001) Crayfish predation on tadpoles: A comparison between a native (*Austropotamobius pallipes*) and an alien species (*Procambarus clarkii*). *Bulletin Français de la Pêche et de la Pisciculture* 361: 659–668, <https://doi.org/10.1051/kmae:2001011>
- Gutiérrez-Yurrita PJ, Montes C (1999) Bioenergetics and phenology of the reproduction of the redswamp crayfish *Procambarus clarkii* in Doñana National Park, SW-Spain. *Freshwater Biology* 42: 561–574, <https://doi.org/10.1046/j.1365-2427.1999.00484.x>
- Gutiérrez-Yurrita PJ, Martínez JM, Bravo-Utrera MÁ, Montes C, Ilhéu M, Bernardo JM (1999) The status of crayfish populations in Spain and Portugal. In: Gherardi F, Holdich DM (eds), Crayfish in Europe as alien species. How to make the best of a bad situation? Rotterdam: A.A. Balkema, pp 161–192, <https://doi.org/10.1201/9781315140469-12>
- Habsburgo Lorena AS, Laurent PJ (1979) Present situation of exotic species of crayfish introduced into Spanish continental waters. *Freshwater Crayfish* 4: 175–184
- Hernandez L, Maeda-Martinez A, Ruiz-Campos G, Rodriguez-Almaraz G, Alonzo-Rojo F, Sainz JC (2008) Geographic expansion of the invasive red crayfish *Procambarus clarkii* (Girard 1852) (Crustacea: Decapoda) in Mexico. *Biological Invasions* 10: 977–984, <https://doi.org/10.1007/s10530-007-9175-0>
- Hobbs H, Jass JP, Huner JV (1989) A review of global crayfish introductions with particular emphasis on two North American species (Decapoda, Cambaridae). *Crustaceana* 56: 299–316, <https://doi.org/10.1163/156854089X00275>
- Huner JV, Barr JE (1984) Red swamp crawfish: biology and exploitation. Baton Rouge, Louisiana, LA: Louisiana Sea Grant College Program, Center for Wetland Resources, Louisiana State University, 184 pp
- IUCN Red List (2009) IUCN Red List of Threatened Species. <https://www.iucnredlist.org/> (accessed 8 November 2018)
- Khalil MT, Fishar MR, Khadra AM (2015) Assessment of burrowing behavior of *Procambarus clarkii*, in Fatmia Drain, Sharqya Governorate, Egypt; Case Study Overview. *Aquatic Biology and Fisheries* 19: 117–124, <https://doi.org/10.21608/ejabf.2015.2262>
- Kouba A, Petrusek A, Kozák P (2014) Continental-wide distribution of crayfish species in Europe: update and maps. *Knowledge and Management of Aquatic Ecosystems* 05: 1–31, <https://doi.org/10.1051/kmae/2014007>
- Liu X, Guo Z, Ke Z, Wang S, Li Y (2011) Increasing potential risk of a global aquatic invader in Europe in contrast to other continents under future climate change. *PLoS ONE* 6: e18429, <https://doi.org/10.1371/journal.pone.0018429>
- Morehouse R, Tobler M (2013) Crayfishes (Decapoda: Cambaridae) of Oklahoma: Identification, distributions, and natural history. *Zootaxa* 3717: 101–157, <https://doi.org/10.11646/zootaxa.3717.2.1>
- Loureiro TG, Anastácio PM, Araujo PB, Souty-Grosset C, Almerão MP (2015) Red swamp crayfish: biology, ecology and invasion - an overview. *Nauplius* 23: 1–19, <https://doi.org/10.1590/S0104-64972014002214>
- Paglianti A, Gherardi F (2004) Combined effects of temperature and diet on growth and survival of young-of-year crayfish: a comparison between indigenous and invasive species. *Journal of Crustacean Biology* 24: 140–148, <https://doi.org/10.1651/C-2374>
- Quan AS, Pease KM, Bereinholt JW, Wayne RK (2014) Origins of the invasive red swamp crayfish (*Procambarus clarkii*) in the Santa Monica Mountains. *Aquatic Invasions* 9: 211–219, <https://doi.org/10.3391/ai.2014.9.2.10>
- Renai B, Gherardi F (2004) Predatory efficiency of crayfish: Comparison between indigenous and non-indigenous species. *Biological Invasions* 6: 89–99, <https://doi.org/10.1023/B:BINV.0000010126.94675.50>
- Rodríguez FC, Becares E, Fernández-Aláez (2003) Shift from Clear to Turbid Phase in Lake Chozas (NW Spain) Due to the Introduction of American Red Swamp Crayfish (*Procambarus clarkii*). *Hydrobiologia* 506: 421–426, <https://doi.org/10.1023/B:HYDR.0000008626.07042.87>

- Smaoui A (2015) Bioclimat et Végétation de la Tunisie et des régions prospectées pendant le 12ème ITER Mediterranem de OPTIMA. *Boccone* 27: 13–20, <https://doi.org/10.7320/Bocce27.1.013>
- Souty-Grosset C, Anastácio PM, Aquiloni L, Banha F, Choquer J, Chucholl C, Tricarico E (2016) The red swamp crayfish *Procambarus clarkii* in Europe: impacts on aquatic ecosystems and human well-being. *Limnologica - Ecology and Management of Inland Waters* 58: 78–93, <https://doi.org/10.1016/j.limno.2016.03.003>
- Vella N, Vella A, Mifsud CM (2017) First Scientific Records of the Invasive Red Swamp Crayfish, *Procambarus clarkii* (Girard, 1852) (Crustacea: Cambaridae) in Malta, a Threat to Fragile Freshwater Habitats. *Natural and Engineering Sciences* 2: 58–66, <https://doi.org/10.28978/nesciences.328931>
- Vigneux E, Thibault E, Marnell F, Southy-Grosset C (2002) National legislation, EU directives and conservation. *Bulletin français de la pêche et de la pisciculture* 367: 887–898, <https://doi.org/10.1051/kmae:2002092>
- Wizen G, Galil BS, Shlagman A, Gasith A (2008) First record of red swamp crayfish, *Procambarus clarkii* (Girard, 1852) (Crustacea: Decapoda: Cambaridae) in Israel - too late to eradicate? *Aquatic Invasions* 3: 181–185, <https://doi.org/10.3391/ai.2008.3.2.8>
- Yanhe L, Xianwu G, Liping C, Xiaohui B, Xinlan W, Xiaoyun Z, Songqian H, Weimin W (2015) Inferring Invasion History of Red Swamp Crayfish (*Procambarus clarkii*) in China from Mitochondrial Control Region and Nuclear Intron Sequences. *International Journal of Molecular Sciences* 16: 14623–14639, <https://doi.org/10.3390/ijms160714623>

Supplementary material

The following supplementary material is available for this article:

Table S1. Sampling locations and records of *Procambarus clarkii*.

This material is available as part of online article from:

http://www.reabic.net/journals/bir/2020/Supplements/BIR_2020_Bouaoud_etal_Table_S1.xlsx