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Research Article

An overview of recent introductions of non-native crayfish (Crustacea, Decapoda) into inland water systems in Israel

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Abstract

Crayfish are known to effectively invade freshwater systems worldwide. Whilst no native crayfish species exist in Israel, three exotic species have been documented in recent years, and their introduction details are hereby discussed. Three of these introduction events, one of each species, were previously reported; one was established, one failed to establish, and the third was successfully eradicated. However, more recently, invasive crayfish have been independently detected multiple times, involving a variety of natural and artificial habitats. Altogether, three invasive crayfish species currently inhabit Israeli freshwaters: *Cherax quadricarinatus*, *Procambarus clarkii*, and *Procambarus virginalis*. They were found in fourteen independent cases, and at least five of them represent established populations. Similar to other crayfish invasions around the globe, we speculate that the invasive populations in Israel result from the intentional release of aquarium inhabitants, as well as from aquaculture escapees. The import and trade policy of aquatic organisms in Israel requires thorough revision to prevent future invasions. Thought should also be given to the prevention of the spread of the existing invasive populations as well as to their eradication, if possible.

Key words: *Cherax quadricarinatus*, *Procambarus clarkii*, *Procambarus virginalis*, aquarium release, invasive species, Levant

Introduction

Crayfish are considered amongst the most impactful invasive species worldwide (e.g. Savini et al. 2010). They prey on invertebrates and vertebrates, and graze on macrophytes, algae, and detritus. Their grazing, walking, and sometimes burrowing activity lead to turbid phytoplankton-dominated systems (Rodríguez et al. 2003, 2005; Haubrock et al. 2021). Crayfish outcompete local species, and their omnivorous diet endangers indigenous biota and simplifies food webs (Geiger et al. 2005; Kreps et al. 2016), resulting in the potential collapse of entire ecosystems. They alter habitat structure and water characteristics (Angeler et al. 2001). Economically, they may threaten fisheries in both commercial fishponds and hobbyist anglers, as well as other agri- and aquacultural effects (Lowery and Mendes 1977; Haubrock et al. 2021; Kouba et al. 2022; Madzivanzira et al. 2022).

In recent decades they rapidly spread in numerous countries, with a few species already being extremely common outside their native ranges of distribution (Souty-Grosset et al. 2016; Madzivanzira et al. 2020; Haubrock et al. 2021; Soto et al. 2023). Accumulating evidence suggests that their main mode of introduction is due to the pet trade: they are extremely popular amongst aquarium holders, and can often be found in shops and private aquaria, and later in natural habitats nearby (e.g. in Germany, Italy, and elsewhere; see Barbaresi and Gherardi 2000; Gherardi et al. 2011; Chucholl 2013; Weiperth et al. 2020; Scheers et al. 2021; Bláha et al. 2022; Soto et al. 2023). In addition, aquaculture was also identified as an important introduction pathway for some species (e.g. Savini et al. 2010; Haubrock et al. 2021).

No crayfish species naturally inhabit inland waters in Israel or adjacent Middle Eastern countries. However, introduced *Procambarus clarkii* (Girard, 1852) have been reported in Egypt and Cyprus (Ibrahim et al. 1995; Papatheodoulou et al. 2021), and *Cherax quadricarinatus* (von Martens, 1868) has been introduced but not established in Cyprus and Lebanon (Haubrock et al. 2021). In Israel, three cases of introduced crayfish species have been reported to date, all recently and probably due to intentional releases; they involve three different crayfish species: *P. clarkii* (Wizen et al. 2008), *C. quadricarinatus* (Snovsky and Galil 2011), and *Procambarus virginalis* Lyko, 2017 (Carneiro et al. 2023). The former was successfully eradicated, the second had never established a sustainable population, and the latter sustains a confined, yet stable, population. However, recent evidence—presented here for the first time—suggests that these were hardly the last cases of crayfish introductions in the country. To the best of our knowledge, we list here all the reported cases of crayfish invasions in Israel and add new recent invasion reports, which altogether paint a very disturbing picture.

Materials and methods

Data for this synthesis were collected from published literature (Wizen et al. 2008; Snovsky and Galil 2011; Carneiro et al. 2023), and additional documented cases based on either collected material that was deposited in natural history collections, or trusted non-scientific reports supported by photographs (personally shared with the authors or published on social media). The authors attempted to confirm all the reports and assess the status of the relevant populations. This aim was also achieved by extensive field surveys, where crayfish were collected using hand-nets (seldom assisted by traps), during the day- and nighttime, in introduction sites and nearby waterbodies. We found some additional leads on social media pointing to the potential presence of crayfish in other freshwater systems, but with no detailed information or photographs, these could not be confirmed and were omitted from the study.

All specimens are deposited at the Crustacea collections in the Steinhardt Museum of Natural History, Tel Aviv University (SMNH-TAU), and in the National Natural History Collections, Hebrew University of Jerusalem (NNHC, HUJ). Species were identified morphologically and molecularly. Morphological identification was based on Holdich (2009). To confirm morphological observations, the mitochondrial barcode gene Cytochrome Oxidase Subunit I (COI) was sequenced for representatives of all available populations. Specimens used for genetic analysis were fixed in 96% ethanol and stored at −20 °C pending analysis. Mitochondrial DNA was extracted from 5 mg of muscle tissue of specimens using DNA Blood and Tissue Kit (Qiagen, Germany), according to the manufacturer's specifications. After DNA extraction, the COI gene was amplified using PCR with the universal primers LCO1490 and HCO2198 (Folmer et al. 1994). Reaction conditions were as follows: 95 °C for 5 min, followed by 35 cycles of 95 °C for 30 s, 50 °C for 45 s, and 72°C for 1 min, and an additional elongation step of 72 °C for 5 min. The PCR products were purified and sequenced by Hylabs (Israel) and uploaded to the public NCBI GenBank database under the accession numbers detailed in Supplementary material Table S1.

We compared the obtained sequences to (1) NCBI database by performing nucleotide blast search (blastn, https://blast.ncbi.nlm.nih.gov/), and (2) BOLD (Barcode of Life Data Systems) identification system for COI (http://www.boldsystems.org/). We further added the sequences to the constructed COI-based crayfish phylogenies. The closely related taxa (*C. destructor*, *C. bicarinatus*, *C. holthuisi*, *C. peknyi*, *C. mosessalossa*; *P. fallax*, *P. alleni*, *P. zonangulus*, *P. acutus*) were chosen as outgroups. The additional sequences were downloaded from NCBI GenBank (Supplementary material Tables S2–S4), added to the sequences of the collected specimens, and aligned using ClustalW in MEGA11 software (Tamura et al. 2021). Evolutionary models and parameter estimates were selected using the lowest AICc score obtained with ModelTest in MEGA11. Maximum likelihood (ML) trees were constructed in MEGA11 with 1000 bootstrapping replicates each.

Results

Fourteen independent cases (i.e., waterbodies with no hydrological connection between them) of crayfish introductions are currently known in Israel. First published only 15 years ago (Wizen et al. 2008), crayfish records have kept accumulating rapidly, with 2022 and 2023 (until August) being the most contributing years so far (Figure 1).

Overall, seven specimens from different populations in Israel were barcoded, including one individual of *Cherax quadricarinatus*, three individuals of *Procambarus clarkii*, and three individuals of *P. virginalis*. The introduced

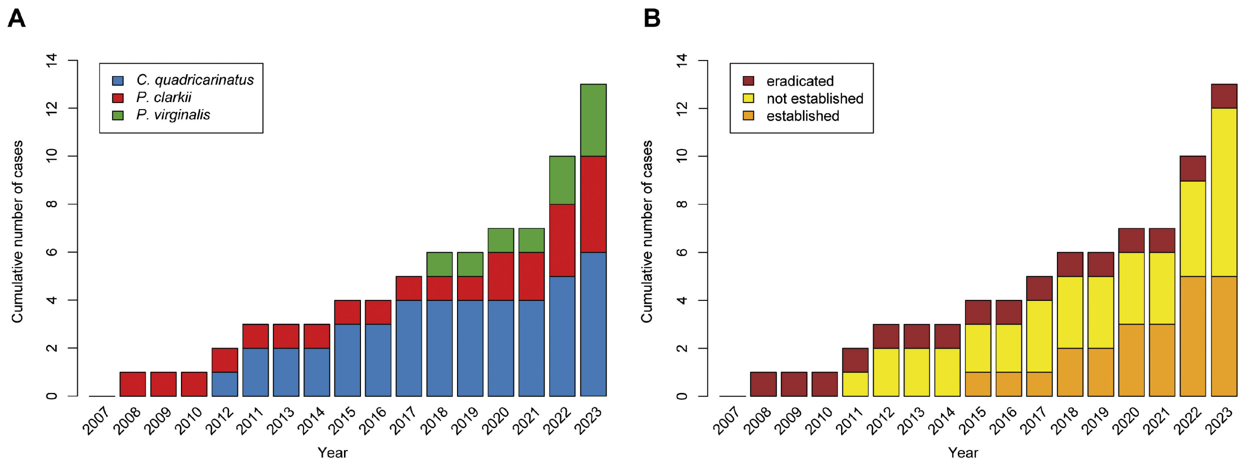

Figure 1. Cumulative number of crayfish introduction reports in Israel (identified species only), starting with the first documentation in 2008, indicating A – the crayfish species and B – the invasion status.

crayfish were found in fourteen sites (Table 1; Figure 2) representing a diversity of habitat types and different physical, hydrological, and chemical conditions (Figure 3). Morphological and molecular identification (% identity to GenBank COI sequences is presented in brackets below) confirmed the occurrence of three non-native crayfish species in Israel to date: *C. quadricarinatus* (100%), *P. clarkii* (99.84–100%), and *P. virginalis* (100%). The COI sequences were deposited in GenBank (http://www.ncbi.nlm.nih.gov/genbank/) and their accession numbers are presented in Table S1. The COI-based ML trees (Figures S1–S3) include both new and published sequences and confirm the morphology-based identification.

Following is a brief account of the known crayfish populations in Israel, most of them presented for the first time. Table 1 summarises these introduction cases.

Cherax quadricarinatus

Figure 4A

Originally from northern Australia and southern Papua New Guinea, this species is currently known from a wide range of tropical and subtropical areas, including in Africa, Asia, North and South America, and Europe (Haubrock et al. 2021). The species is popular worldwide for ornamental and aquacultural uses, *Cherax quadricarinatus* were introduced to Israel several times for aquaculture purposes since the 1990s, with the support of the Ministry of Agriculture to diversify the local aquaculture industry (Karplus et al. 1995; Davidovich et al. 2021; http://www.fao.org/fishery/en/introsp/288). This tropical species seems not to be confined by the relatively cool conditions of the Israeli winter (Karplus et al. 1998).

In nature, the first individual of this species was caught in 2011 (Snovsky and Galil 2011) in the Sea of Galilee, an important source of drinking water in Israel, that also accommodates vast commercial and amateur fishery, as

Table 1. Summary of the confirmed crayfish introduction cases in Israel.

Species	Location name	Latitude	Longitude	Site description	First observation	Population status	Reference/Source of information
Cherax quadricarinatus	Sea of Galilee	32.789	35.547	Lentic; littoral, lake	January 2011	Not established	Svonsky and Galil 2011
Cherax quadricarinatus	Tel Saharon, Saharon Stream	32.449	35.545	Lotic; small, ecologically sensitive brook in a nature reserve	16 October 2012; 14 August 2017	Assumingly not established	Current study
Cherax quadricarinatus	Yarkon Stream	32.122	34.903	Lotic; large, disturbed natural stream	November 2015	Established	Current study
Cherax quadricarinatus	Amal Stream	32.500	35.463	Lotic; disturbed, canalised natural stream	20 June 2017; 5 June 2018	Assumingly not established	Current study
Cherax quadricarinatus	Rishon LeTsiyon (lake)	31.977	34.749	Lentic; artificial lake for flood control, sandy bottom	15 November 2022	Not established	Current study
Cherax quadricarinatus	Kfar Yona	32.316	34.932	Lentic; ornamental pond (likely the source)	July 2023	Not established	Current study
Procambarus clarkii	Hadera (quarry)	32.423	34.887	Lentic; ephemeral pond on artificial setup	11 March 2008	Eradicated	Wizen et al. 2008
Procambarus clarkii	Tel Aviv University	32.111	34.808	Lentic; small free flow pond (part of a constructed wetland)	November 2020	Established	Current study
Procambarus clarkii	Betset Stream	33.076	35.222	Lotic; natural mediterranean stream	10 October 2022	Established	Current study
Procambarus clarkii	Yeroham (reservoir)	30.989	34.897	Lentic; artificial lake for flood control, sandy bottom	August 2023	Not established	Current study
Procambarus virginalis	Guvrin Stream, Tsanen Pond	31.604	34.959	Intermittent stream, mainly in a dug pool with a constant supply of (sometimes only underground) water	2018 (social media); 21 December 2022	Established	Current study
Procambarus virginalis	Siah Stream, En Meshotetim	32.801	34.974	Spring-fed constructed basin (free flow) in an ephemeral stream course	August 2022	Established	Carneiro et al. 2023; current study
Procambarus virginalis	Nurit	32.543	35.355	Lentic; ornamental pond (likely the source)	July 2023	Not established	Current study
Unidentified	Shma'aya Pools, Dead Sea shore	31.139	35.377	Dug, spring-fed pool in open landscape. Drained today		Not established	Current study

well as recreational activity. Over a decade later, no additional individuals have been observed in the lake. We, therefore, believe that this individual does not represent an established population, until further information proves otherwise.

On the other hand, *C. quadricarinatus* established a sustainable population in the Yarkon Stream, the largest of the coastal streams in Israel, which runs through urban, agricultural, and natural landscapes. This population was first noticed in 2015 and has been regularly sampled and observed in the stream ever since. Despite the lack of standard tracking, professionals share the impression that the population extended down the stream, covering most

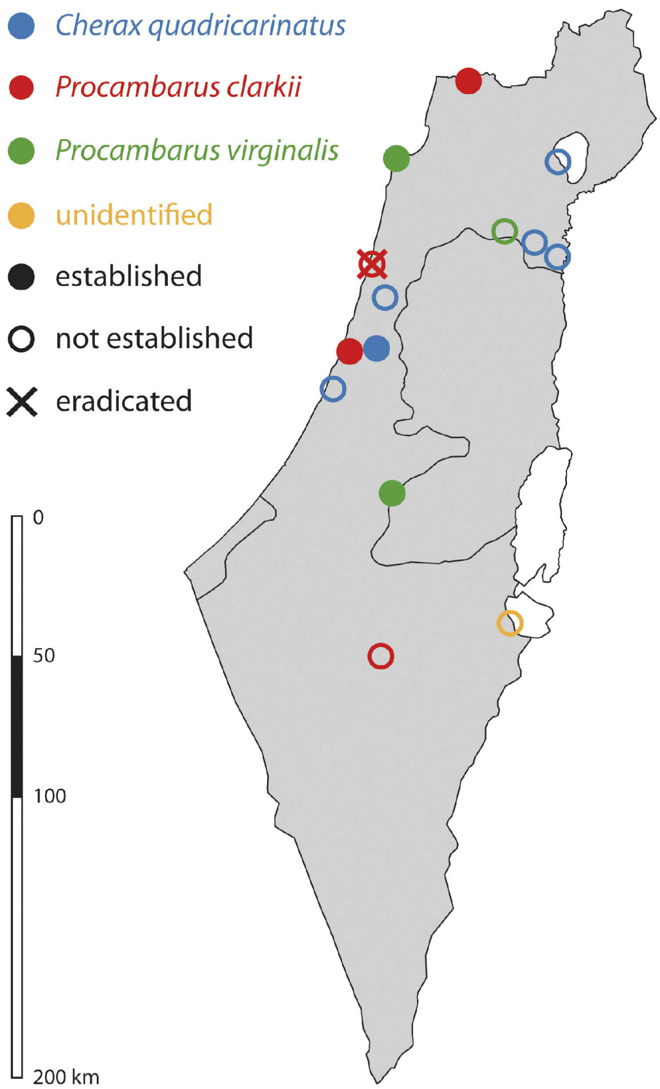

Figure 2. Map of Israel presenting all the reported cases, including (assuming) non-established, established, and eradicated crayfish populations.

of its upper, unpolluted section, and reached a plateau, probably maximising the benefit from local resources.

Cherax quadricarinatus specimens were also documented in a few other locations, a single or two individuals in each case. This includes two locations in Bet She'an Valley: Tel Saharon Nature Reserve and the Amal Stream. The latter site was recently surveyed once using flashlights at night (2022, ZY and NT *unpublished data*), and no crayfish were found. More recently, two specimens were also found (November 2022 and July 2023) by amateur fishermen in a large artificial reservoir used for flood regulation in the city of Rishon LeTsiyon (central district). Fishermen never reported the species to the site management again, and an attempt to find crayfish using electrofishing failed (2022, ZY, AS, and DM *unpublished data*). Finally, another individual was spotted, but not captured, in the rural settlement Kfar Yona (central coastal plain). Until further information is available, we consider this case an unestablished population.

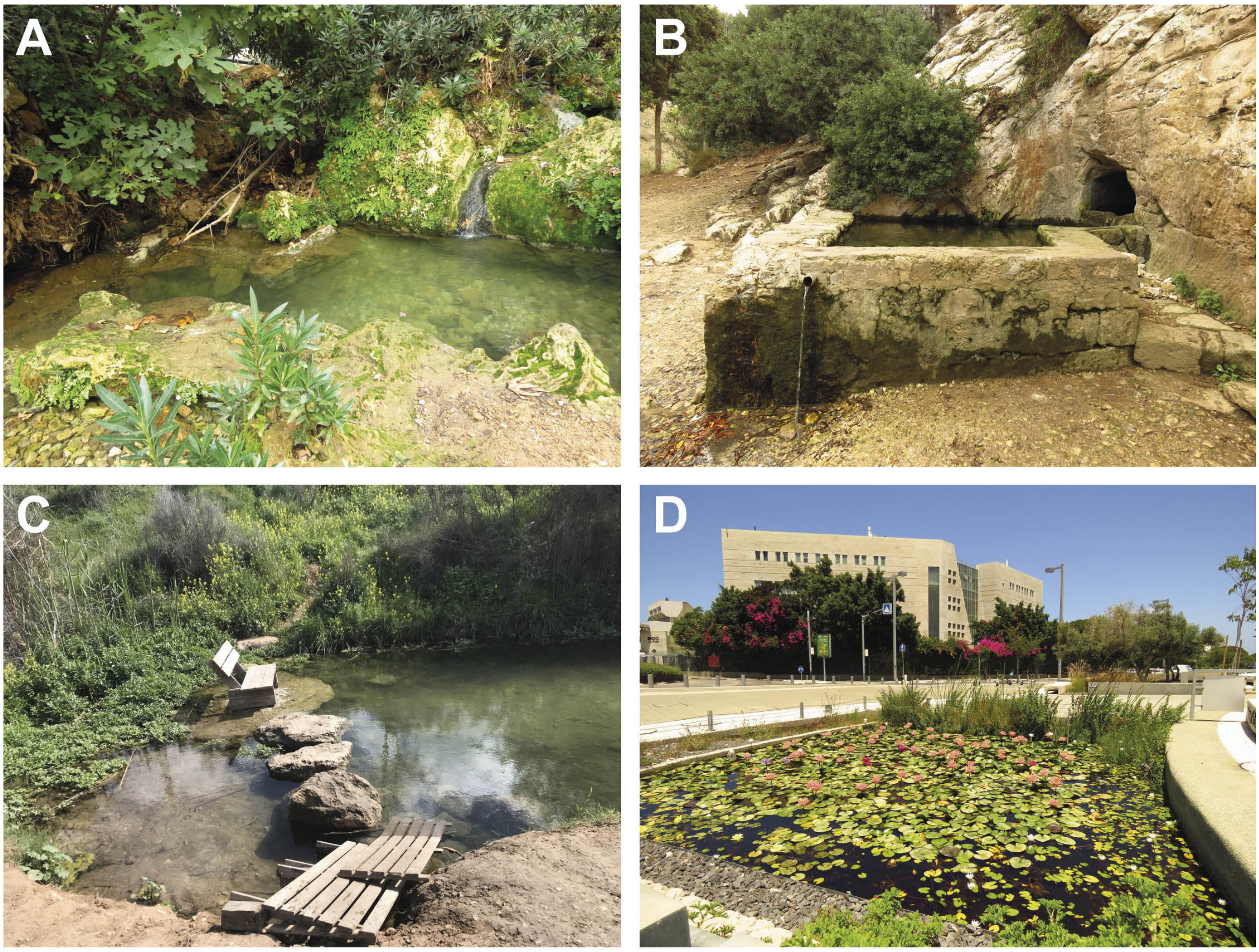

Figure 3. Representation of the various invaded habitats in Israel: A – Betset Stream, a natural, recovered mediterranean stream; B – Meshotetim Spring in Siah Stream, a spring feeding a restricted constructed basin with low complexity; C – a dug pond in Guvrin Stream, which remains dry in the dry season; D – an artificial constructed wetland at Tel Aviv University Campus. Photos: Zohar Yanai (A, B, D); Oren Kolodny (C).

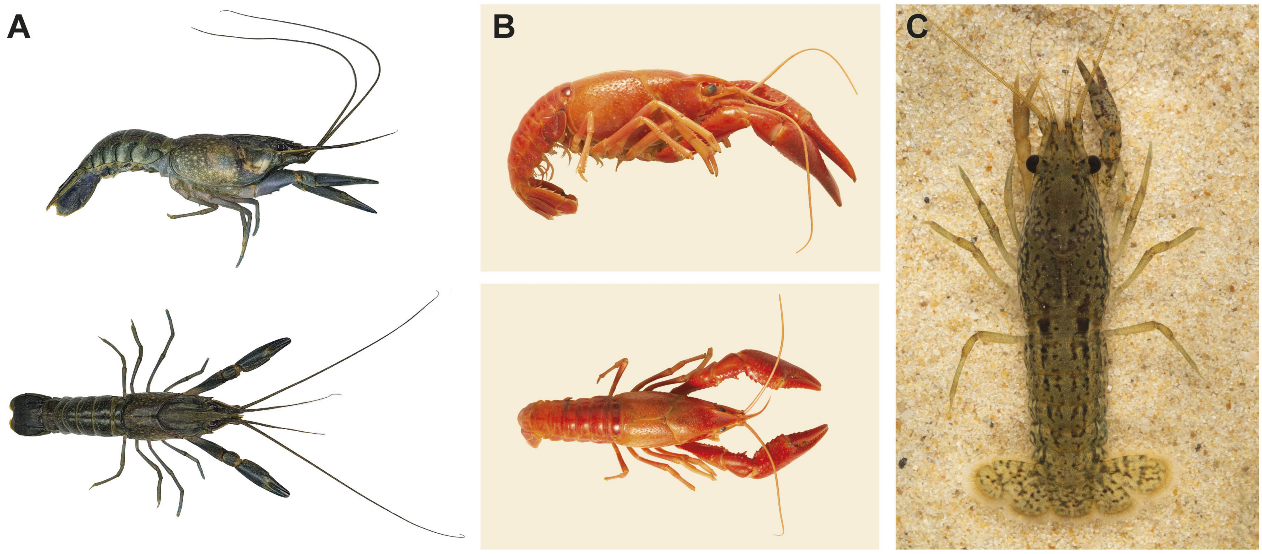

Figure 4. Individuals of the invasive crayfish species in Israel: A – *Cherax quadricarinatus* (in captivity); B – *Procambarus clarkii* (Betset Stream population, lateral and dorsal view); C – *Procambarus virginalis* (Siah Stream population). Photos: Khristina Ermak (A), Oz Rittner (B, C).

Procambarus clarkii

Figure 4B

The most devastating invasive crayfish species globally is *P. clarkii*, originally from Mexico and southern USA, and currently spread across Europe, Africa, eastern Asia, and other regions (Gherardi 2006; US Fish and Wildlife Service 2011; Souty-Grosset et al. 2016). It was the first crayfish to be discovered in Israel.

The first *P. clarkii* population in Israel was found in 2008 in a deserted quarry that is seasonally flooded near the city of Hadera (Wizen et al. 2008). Crayfish were collected in the pond intensively for three years, using manual harvesting and traps. Whilst this pond did not accommodate any sensitive natural elements, there are a few ecologically valuable seasonal ponds in very close vicinity. All the experts involved agreed that determined action was needed as the natural ponds faced concrete threat. The quarry pond was destroyed by covering it with vast amounts of dirt, and the crayfish population was completely eradicated.

Since 2020, a small population of *P. clarkii* is known from an artificial pond at Tel Aviv University Campus (Figure 3D). This pool illustrates a constructed wetland used for effluent treatment and is rich with aquatic vegetation and shelters for crayfish. For a while, the pond crayfish population was monitored regularly by manual collection and baited traps, and an eradication attempt was carried out using biological control agents (A. Gasith *personal communication*). No crayfish are documented on the site since this last attempt; however, not enough time has passed and for the moment there is still no guarantee that the eradication was successful.

A stable population was recently discovered in Betset Stream in northern Israel (Figure 3A). This is a mediterranean stream found within a nature reserve, that suffered a severe decline in water availability and held no water at all for almost two decades. The Nature and Parks Authority and the Water Authority have been investing resources and efforts to support the weakened natural springs and resume the water supply to the stream. Consequently, since 2019 it has been running again, aiming to regain the stream's permanent flow in the future. Its fauna, flora, and entire ecosystem are being gradually recovered. The *P. clarkii* population was discovered in Betset Stream in October 2022. Repeated nocturnal monitoring operations have been carried out ever since, confirming that this population is stable, although rather small and restricted to a limited section of the stream for the moment. This population poses the greatest conservation challenge of all cases, due to the complexity of the site and since any eradication measure may endanger the delicate restoration programme. Sadly, it is obvious that the ecosystem will never fully recover whilst accommodating an aggressive invasive crayfish.

A single specimen was recently reported from an artificial reservoir in southern Israel, most likely representing another episodic observation rather than a population.

Procambarus virginalis

Figure 4C

Recently reported from a captive population, this crayfish is capable of parthenogenetic reproduction (Vogt et al. 2004), making its eradication extremely challenging. It is derived from the slough crayfish *P. fallax*, to which it is very similar morphologically and genetically. Integrating some strong evidence, it was described as an independent species (Lyko 2017). The homogeneous sequences obtained in this analysis confirm the parthenogenetic reproduction mode in this species.

The species' first documentation from natural habitats in Israel comes from social media in 2018 in a dug pool in the course of Guvrin Stream (Figure 3C). Whilst this is an intermittent stream that dries out for most of the year, the pool maintains some volume of water and rarely dries. Recent visits (December 2022–May 2023) proved the population was still viable, with many dozens of individuals found in the water or the humid conditions under nearby rocks. This finding is in accordance with published features of the species, such as burrowing capacities and drought resistance (Kouba et al. 2016; Tönges et al. 2021).

A denser population has been found, at least since November 2021, in En Meshotetim, a small spring in Siah Stream in the city of Haifa (Carneiro et al. 2023). This spring derives from the rocky walls of Mount Karmel and constantly feeds a small water body carved within the rock, from which the water flows directly into a small, constructed basin, commonly used for bathing (Figure 3B). Few to tens of individuals are collected in every visit in the spring, including gravid females. A few individuals were recorded outside the basin, proving that escape from the aquatic medium to its humid environment is possible. It seems that the dry conditions further away, or perhaps pure chance, have prevented the crayfish from spreading and populating Siah Stream so far. However, the threat, especially from such a parthenogenetic species, remains.

Procambarus virginalis was also collected in the rural settlement of Nurit (the Gilboa Rigde, northern Israel), where it was observed walking around on pavements at nighttime and collected (four mature females and many larvae) in a nearby ornamental pond. The pond has recently been dried, and the site is monitored to assess the stability of this population.

Unidentified crayfish

An additional report of crayfish occurrence, supported by photographed evidence, arrives from a semi-natural pool near the Dead Sea, impacted by heavy industrial activity. Unfortunately, the quality of the photograph did not enable the morphological identification of the species, but it is evident that a dense population existed in the pool. No crayfish were ever collected at the site, and the pool was drained out since the first record. We currently have no reliable way to identify this species.

Identifying the crayfish that currently occur in Israel

We provide an identification key to the crayfish currently known from Israel, to assist ecologists and nature managers in future encounters. Ecologists and practitioners may benefit from this key, especially given the fact that crayfish are not usually familiar in the country. The key is based mostly on Holdich (2009), and also on Lyko (2017). Potential users must bear in mind that these are non-native species and that crayfish, as a group, are highly invasive; this implies that a random crayfish in the field may be included in the presented key but may also be a novel non-native species. Furthermore, many crayfish species present wide morphological variability, and as a rule of thumb combining molecular identification (based on COI sequences) is preferred over solely morphological identification.

An identification key to the crayfish species that are currently known to be found in open water systems in Israel (see Figure 4)

1. Long, developed rostrum, margins extending far onto carapace, acumen (apex) bordered with spines. Postorbital ridges long, reach half-way down the carapace, forming lateral keels. Inner margin of propodus (palm) longer than dactylus (finger). Adult males with red patch on outer margin of claw. Carpal spur absent. Annulus ventralis (sperm storage organ) absent. First pleon segment without pleopods in both sexes .... Parastacidae; *Cherax quaricarinatus*

–. Short, triangular rostrum, acumen (apex) not bordered with spines. Postorbital ridges short and shallow. Inner margin of propodus (palm) shorter or similar in length to dactylus (finger). Males without red patch on outer margin of claws. Carpal spur present. Annulus ventralis (sperm storage organ) present between the last two pairs of pereopods in females. First pleon segment with developed (males) or thread-like (females) pleopods ....Cambaridae; 2

2. Both males and females exist. Colour usually red, reddish-brown or reddish-orange, rare colouration exists but not marbled; juveniles olive-green to brown. Narrow, almost closed, areola. Chelae dorsally covered in tubercles, and with sinuous propodus. Prominent carpal spur............. .... *Procambarus clarkii*

–. Only females exist. Marbled pattern. Wider areola. Chelae with few tubercles and straighter. Carpal spur less prominent, not found in juveniles.. *Procambarus virginalis*

Discussion

Non-native crayfish species can now be found in a variety of local freshwater habitats, as part of what appears to be a series of independent introductions with tremendous potential to affect local freshwater biodiversity and ecological stability.

Pet shops are a major source for non-native aquatic species in Israel, many of which have already escaped to nature (e.g. Roll et al. 2008, 2009; Mienis et al. 2016; Yanai et al. 2017), including crustaceans (Levitt-Barmats et al. 2019). Whilst we cannot prove it for certain, the source for most of the reported populations is highly likely deliberate release by hobbyists, similar to other countries (Chucholl 2013; Souty-Grosset et al. 2016; Patoka et al. 2018; Haubrock et al. 2021). Some crayfish species (e.g., *C. quadricarinatus*) have been legally imported to Israel in the past, but all species are currently illegal for import, owning, and trade in Israel: the few crayfish species that used to be legal have recently (since 2016) been removed from the list of approved aquatic organisms (Fisheries Regulations 1937). Nevertheless, they are still common amongst aquarium shops and private aquaria. The updated regulations are hardly communicated to the public, and an actual change is yet to be seen in aquarium shops (unpublished data). In addition, observations of common aquarium fishes at the same sites (e.g. goldfish *Carassius* sp. in Siah Stream, suckermouth catfish *Hypostomus plecostomus* in Betset Stream) suggest that aquarium contents have been spilled there. The pace of these recent releases perhaps reflects a social trend that is expected to increase. Such acts are fed by misunderstanding of ecological concepts and must be addressed by educational activities and designated information campaigns (Patoka et al. 2018).

An exception is Amal and Saharon Streams, which are relatively close to aquacultural ponds in Bet She’an Valley. Following attempts to breed *C. quadricarinatus* in those ponds, it is likely that escapees found a path to adjacent watercourses (as has happened elsewhere; Haubrock et al. 2021). Ornamental garden ponds may serve as mediators for future invasions as well (Patoka et al. 2016). For the moment, it appears that this mode of introduction is more limited in Israel, although attention must be kept especially around ornamental ponds that are already known to inhabit potentially invasive populations (e.g. in Tel Aviv University, Figure 3D).

With profound ecological impact on mediterranean wetlands (Geiger et al. 2005; Gherardi 2006), invasive crayfish pose a great threat to the aquatic biodiversity in Israel. The pace of new records and geographical distribution is alarming (Figures 1, 2). Four populations are currently suspected to be established in natural habitats is Israel, as well as one in an artificial urban pond. We do not rule out the possibility that other episodic observations will turn out to be established populations; we also consider that additional introduction cases may already exist and will be found sooner or later. Early detection is a key factor for successful eradication of invasive species (Reaser et al. 2020). Invasive crayfish have tremendous impact on ecosystems around the world (e.g. Rodríguez et al. 2005; Gherardi 2006; US Fish and Wildlife Service 2011; Souty-Grosset et al. 2016; Madzivanzira et al. 2022), and it is too early to predict their concrete impact on Israeli ecosystems, that have evolved without crayfish components.

Now that the information about crayfish invasions in Israel is clear and available, it is time to gain better understanding of the circumstances of these introductions, and phrase nationally relevant tools for prevention and coping. Further genetic investigation of the population structure and diversity of the introduced species can provide insights on the introduction origins and the status of invasion. In addition, the environmental impact on the recipient ecosystems should be investigated. A joint taskforce, combining ecologists from the academia, Nature and Parks Authority, and other relevant authorities, is examining effective eradication techniques, with special attention to the unique characteristics of each site.

Authors' contribution

Research conceptualisation: ZY, TGH, OK, YLB, AS, NT, DM; sample design and methodology: ZY, TGH, OK, AM, ARM, AS, NT, DM; investigation and data collection: ZY, TGH, OK, AM, ARM, AS, NT, DM; data analysis and interpretation: ZY, TGH, YLB, ARM, AS, NT, DM; writing – original draft: ZY, TGH, OK, YLB, AS, NT, DM; writing – review and editing: ZY.

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Ethics and permits

Ethics was not required. Collection within nature reserves was either done by, or together with, ZY (under permits 43135 and 43407) or DM or AM (Nature and Parks Authority employees).

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Supplementary material

The following supplementary material is available for this article:
Figure S1. A maximum-likelihood tree of *Cherax quadricarinatus* and an outgroup taxa.
Figure S2. A maximum-likelihood tree of *Procambarus clarkii* and outgroup taxa.
Figure S3. A maximum-likelihood tree of *Procambarus virginalis* and outgroup taxa.
Table S1. COI Sequences obtained for the present study.
Table S2. *Cherax quadricarinatus* specimens and outgroup species included in the phylogenetic analysis.
Table S3. *Procambarus clarkii* specimens and outgroup included in the phylogenetic analysis.
Table S4. *Procambarus virginalis* specimens and outgroup included in the phylogenetic analysis.

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