ScienceDirect

Global Ecology and Conservation
Volume 54, October 2024, e03071

The use of thermal performance analysis to improve conservation management: The case of the invasive Red Sea swimming blue crab *Portunus segnis*

Guillaume Marchessaux [a] [b] [1], Raouia Ghanem [a] [c] [2], Amani Chaffai [d] [3], Wafa Rjiba Bahri [c] [4], Nicoletta Marsiglia [a], Vojsava Gjoni [a] [b] [5], Jamila Ben Souissi [c] [6], Gianluca Sarà [a] [b] [7]

Show more

Outline | Share | Cite

https://doi.org/10.1016/j.gecco.2024.e03071
Get rights and content

Abstract

The latest report from the Intergovernmental Platform on Biodiversity and Ecosystem Services (IPBES) expresses concern over the global proliferation of non-native species (NIS) driven by increasing temperatures. In 2023, the invasive blue crab *Portunus segnis* in the Mediterranean experienced a significant range expansion due to warmer waters in northern and western regions. Recognizing thermal tolerance as crucial for conservation, this study, using the blue crab as a model, emphasized the importance of considering thermal metabolic performance in conservation efforts. In this study, we experimentally measured the metabolic performance of *P. segnis* at different temperatures (from 7 to 42°C) using an oxygen consumption measurement system. The thermal performance curve (TPC) of *P. segnis* rose from 0.016 ± 0.011 $mgO_2\ h^{-1}\ gWW^{-1}$ (12°C) to 0.238 ± 0.011 $mgO_2\ h^{-1}\ gWW^{-1}$ (34°C), and decreased beyond. Organisms succumbed at 7°C and 42°C. The TPC obtained was compared with 24 TPC models and the O'Neill regression was the best fitting model determining the minimum critical thresholds (CT_{min}) at 11.33°C and CT_{max} at 41.13°C. Using the TPC curve equation, we applied the curve to satellite Mediterranean temperature data for the year 2022 and for the future (RCP 4.5 and 8.5) to determine the current and future monthly distribution of favorable thermal habitats for *P. segnis*. Results anticipated the future expansion of the species along the Spanish, French, Italian, and Greek coasts of the Aegean Sea, providing maps to prioritize high-risk invasion areas. Additionally, in the areas where the species in currently present, results predicted a long-term persistence of the species. Understanding species' responses to temperature changes appears crucial for anticipating and addressing the impending expansion of invasive species, underscoring the urgency of nature conservation measures.

Previous Next

Keywords

Thermal tolerance; Blue crab; Portunus segnis; Conservation measures; Biological invasion

1. Introduction

The latest report from the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services (IPBES) raises alarms about the global proliferation of Non-Indigenous Species (NIS) (Roy et al., 2023). Among the 37,000 recorded NIS, 3500 are Invasive Alien Species (IAS), contributing to 60% of global species extinctions, impacting biodiversity through ecosystem modifications, resource competition, and predation (Roy et al., 2023). Climate change exacerbates species distribution changes, indirectly facilitating IAS spread, often at the expense of indigenous species. Monitoring both indigenous and invasive species is crucial for understanding climate change impacts and implementing effective conservation measures (DeLong et al., 2018).

In 2023, a historic shift occurred in the invasion dynamics of the Red Sea swimming blue crab, *Portunus segnis* (Forskål, 1775), in the Mediterranean Sea. The unprecedented warming of Mediterranean waters facilitated its exponential expansion, with new sightings in the Gulf of Cadiz (Spain) and the Italian Adriatic coasts, surpassing its previous confinement to the southeastern Mediterranean (de Carvalho-Souza et al., 2023, Grati et al., 2023). Originating from the Indo-Pacific, *P. segnis*, an early Lessepsian invader, was first recorded in

Egypt in 1898 after the Suez Canal's construction. Since the 2000s, it has established across the south Mediterranean, posing threats to local biodiversity and artisanal fisheries (Marchessaux et al., 2023, Tureli and Yesilyurt, 2017). While its aggressive behavior negatively impacts native species and habitats, the challenge lies in devising effective conservation measures for such invasive organisms reaching new habitats (Blackburn et al., 2011). The potential expansion of *P. segnis* underscore the urgency of implementing robust conservation strategies to mitigate local biodiversity losses and safeguard ecosystem integrity.

Conservation is based on diverse methods, including habitat restoration, protected area management and proactive measures to combat invasive species (Rahel et al., 2008). However, new challenges posited by climate change need adaptation and refinement of the current accepted strategies (Ben Souissi et al., in press). For instance, creation of climate-resilient corridors may help species migrate to more suitable habitats, as well as the development of policies to prevent the further spread of invasive species. Ultimately, understanding the intricate relationship between climate change and the distribution of species now is becoming urgent for the effective preservation of biodiversity and the long-term health of our planet's ecosystems.

The increase in temperature is the main climate factor driving new invasions (Wallingford et al., 2020). Understanding the thermal tolerance of non-indigenous species (NIS) is crucial for predicting how, when, and where these species will colonize new habitats. Thermal tolerance is a critical trait in many species, particularly among ectothermic organisms, as it reflects their adaptation to local temperature conditions. Body temperature has a profound impact on metabolic rate, a crucial aspect for ectotherms that rely on external heat sources to regulate their internal temperature (Matzelle et al., 2015). Consequently, their internal temperature mirrors their environment, whether aquatic or terrestrial. This correlation implies that internal energy flows, such as assimilation and somatic maintenance (Kooijman and Kooijman, 2010), are controlled by body temperature, thus forming the basis of the thermal performance curve.

Organisms exhibit variations in their performance curves, including breadth, skewness, and optimal position (Angilletta and Angilletta, 2009, Rubalcaba et al., 2020, Sokolova et al., 2012). Performance curve analysis has proven essential in understanding the relationship between environmental factors like temperature and salinity and various fitness-related parameters such as growth efficiency, assimilation, fecundity, survivorship, and feeding rate (Angilletta and Angilletta, 2009, Sokolova et al., 2012). These curves (Fig. 1), which vary with body temperature, assume diverse shapes, often with right-positive skewness (indicating cold thermal preference), left-negative skewness (heat-tolerant species), or symmetry (generalists) (Angilletta and Angilletta, 2009, Sinclair et al., 2016) (Fig. 1). Compared to theoretical correlative models (e.g. Species Distribution Models) based on species distribution via their occurrence with potential bias in species presence data, the mechanistic measurement of metabolic performance via thermal tolerance involves measuring respiration rates across a wide range of temperatures, which may be outside the natural conditions where the species is observed, to determine the complete performance curve (Bosch-Belmar et al., 2022, Marchessaux et al., 2022). This method allows for the collection of valuable data to predict species' performance responses to higher temperatures beyond those currently observed, offering insights into potential colonization patterns under rising temperatures (Bosch-Belmar et al., 2022, Marchessaux et al., 2022).

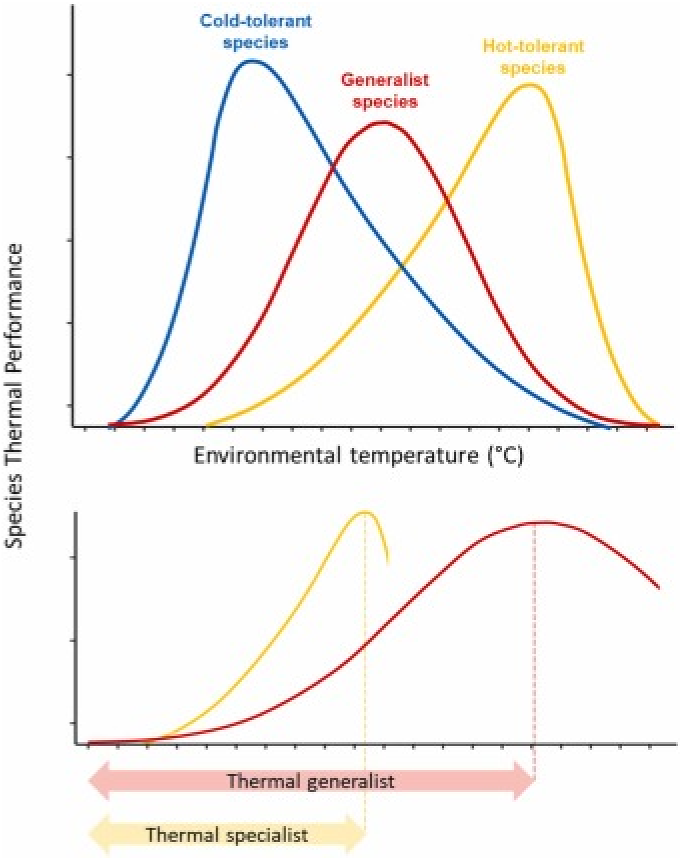

Fig. 1. Conceptual diagram presenting the different response scenarios of ectotherms cold-tolerant, hot-tolerant and generalist species to climate warming (on the top). Comparison of the different adaptive strategies of generalist and specialist (cold or hot tolerant) species to climate warming (on the bottom). Generalist species (red curve) have metabolic rates maintained over a wide thermal range, but have a limited capacity for adaptation and

acclimatization, leading to a reduction in adaptive potential. Specialist species (blue curve (for the cold tolerant) or orange curve (for hot tolerant)) have a narrower thermal niche, with a limited acclimatization response, and may be more sensitive to temperature changes. In contrast, tropical species (hot tolerant) can withstand modest increases in environmental temperatures.

Figure modified from (Verberk et al., 2016).

Thermal tolerance is now recognized as a valuable tool for measuring conservation efforts (Sinclair et al., 2016, Verberk et al., 2016). Understanding how species respond to temperature changes can help conservationists assess the impact of climate change on ecosystems. This allows for estimating the detrimental repercussions on local biodiversity and the consequent effects on ecosystem functioning (*sensu* Bannar-Martin et al., 2018). Mapping an organism's thermal tolerance curve facilitates the establishment of niche-based mechanistic relationships, aiding in predicting potential habitats and emergence times within local biodiversity. This mapping tool based on species performance curves allows for the development of targeted strategies, yet to be appropriately designed, to protect and preserve biodiversity, as well as producing powerful tools for policymakers aimed at mitigating the effects of global warming. Essentially, this knowledge empowers us to make informed choices to safeguard our natural world in the face of environmental challenges.

Given the potential of this approach and the recent expansion of the invasive Red Sea swimming blue crab, *Portunus segnis*, this study presents how experimentally measured thermal tolerance can help produce predictive distribution maps to define its current and future distribution. This provides essential information to understand the invasion story of this impactful invasive species, enabling us to anticipate potential future expansion and improve conservation management at different spatial scales.

2. Materials and methods

2.1. Sampling and respirometry measurements

In September 2023, 100 organisms were collected in the Bizerte Lagoon, Tunisia (latitude: 37.230, longitude: 9.863), using artisanal gill nets. Crabs were acclimated to laboratory conditions (60 L aquarium, 26°C temperature, 28 psu salinity) for 48 hours, and divided into 16 groups of 6 individuals each, corresponding to the 16 experimental temperatures studied ranging from 7°C to 44°C. After 24 hours of acclimatization to experimental temperature (adjusted by 1°C per hour; Bosch-Belmar et al., 2022, Marchessaux et al., 2022, Prusina et al., 2014), organisms of each group (x 6) were placed in a respirometric chamber (volume: 2 300 mL; diameter: 15 cm) containing air-saturated filtered water (Whatman

GF/C, 0.45 µm) (Fig. 2). Two chambers without blue crab were used for controls to define the natural decrease in oxygen concentration in water only. The respirometric chambers (2 controls and 6 experimental) were distributed in temperature-controlled circulated stable water baths. The controlled chiller system and the thermostatic heater (Grant Optima TX150) were used to cool or heat the water and maintain the experimental temperatures (Fig. 2). Different organisms (6 per temperature) were used for each temperature treatment. In each respirometric chamber (controls and experimentals), water was mixed with magnetic stir bar placed in a plastic cage (6 cm diameter, 0.8 cm width) to avoid contact between the magnetic stir bar and the crab (Bosch-Belmar et al., 2022, Bosch-Belmar et al., 2021, Marchessaux et al., 2022). Oxygen consumption was measured in continuum (every second) for 1 hour using the Pyro Science Firesting O2 system (Fig. 2). At the end of the experiment, specimens' individual wet weight (WW) was measured to adjust respiration rates based on individual WW, to avoid the body mass effects (Bosch-Belmar et al., 2022, Marchessaux et al., 2022).

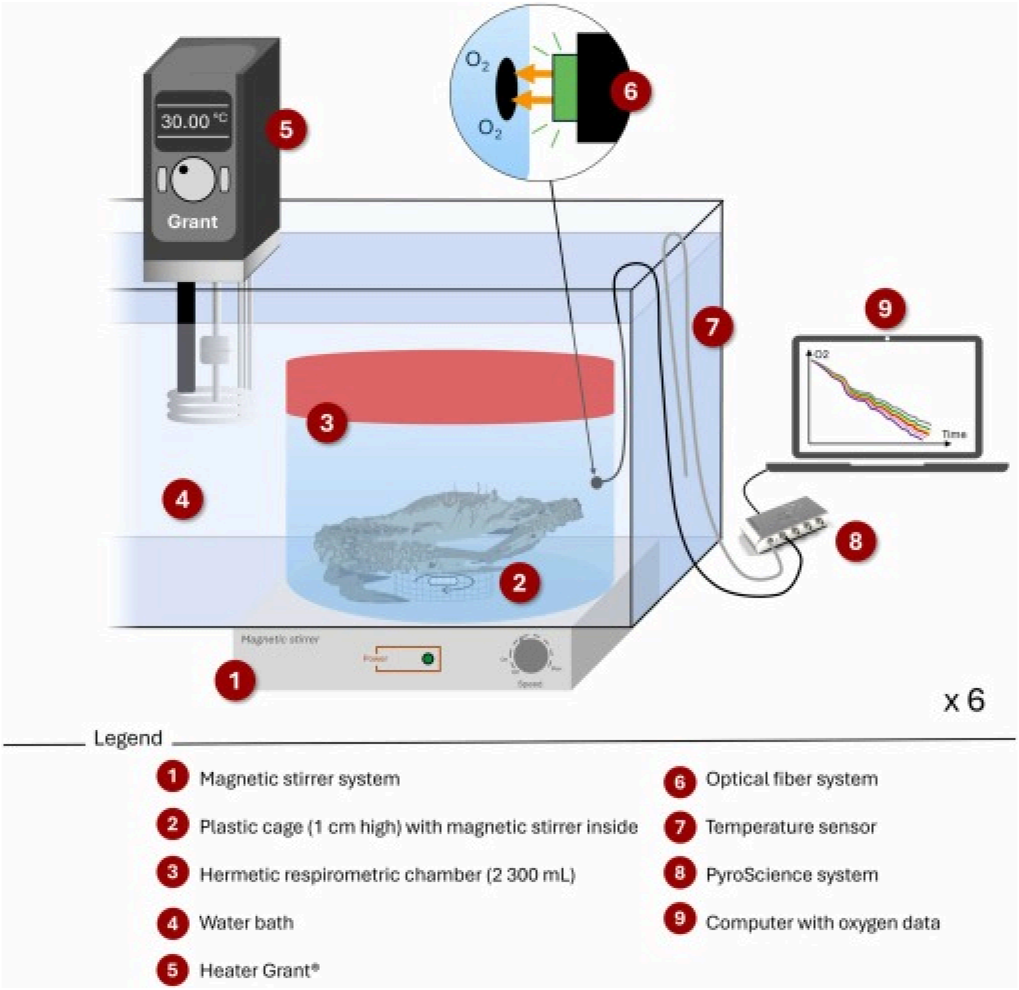

Fig. 2. Conceptual diagram presenting the experimental set-up used to measure the species thermal performance. The numbers correspond to the different elements used in the experiments (refer to the legend for more details). Each experimental setup consists of a water bath containing ~100 L of water heated using a Grant® heater to maintain a constant temperature. Below the water bath, four magnetic stirrer stations are positioned, on which the respiration chambers are immersed (1 control per water bath and 3 experimental chambers). Each respiration chamber (2 300 mL) contains filtered seawater and a plastic cage glued to the bottom enclosing a magnetic stirrer ensuring water homogenization within the respiration chamber and avoiding contact between the magnetic stirrer and the blue crab. A spot is attached to the glass wall inside the respiration chambers, allowing measurement of oxygen concentration using the optical probe, connected to the Pyro Science Firesting O2 system, positioned on the exterior wall of the respiration chamber. The respiration chambers are hermetically sealed before immersion. This diagram was performed by G. Marchessaux.

As respiration rate (RR) being a linear curve, for each experimental condition (control and experimental) at each temperature, a linear regression was performed, and the slope (a) of each curve (RR = a*Time + b), corresponding to an oxygen consumption rate per second, was extracted. The natural oxygen consumption decrease in control conditions was removed to the experimental data to define the real RR of each specimen. The RR was converted per hour and divided by the wet mass of each individual to obtain a rate in $mgO_2\ h^{-1}\ gWW^{-1}$.

2.2. Thermal performance curve model

Using R studio (version 2021.09.0), we employed the "rTPC" R package (Padfield and O’Sullivan, 2020) to define the “best” Thermal Performance Curve (TPC) model regression by comparing 22 non-linear least-squares models and selected the "best" model based on the lowest AIC (Akaike information criterion) scores (Bosch-Belmar et al., 2022, Marchessaux et al., 2022, Padfield and O’Sullivan, 2020) (Supplementary Table 1). We visualized the TPC, calculated 95 % prediction limits using bootstrapping (95 % confidence interval), and extracted parameters: maximum respiration rate (r_{max}), optimal temperature (T_{opt}), temperature coefficient (Q_{10}), and Critical Thermal maximum (CT_{max}), and minimum (CT_{min}) to assess heat species stress tolerance. The thermal safety margin was considered to gauge sensitivity to heat stress, especially relevant for tropical species (Desforges et al., 2023).

2.3. Mapping the current and future thermal suitability

The TPC model was used to predict the potential distribution of *P. segnis* in current (year 2022) and future (2050) climate scenarios under different greenhouse gas emissions (RCP 4.5 (the optimistic one); and RCP 8.5 (the pessimistic one); outputs multiple CMIP5 models, https://cds.climate.copernicus.eu/ ↗). To convert the TPC into Thermal Habitat Suitability (THS) maps, respiration rates were scaled to the oxygen consumption at the predicted optimum temperature using the formula $THS = RR_t / RR_{Opt}$, where THS represents occurrence probability, RR_t the respiration rate at temperature t, and RR_{Opt} the respiration rate at the optimum temperature from the TPC model (Bosch-Belmar et al., 2022, Marchessaux et al., 2022). THS values (ranging from 0 to 1) were calculated for each 1 km^2 pixels, each month of the year up to a depth of 65 m in the Mediterranean Sea, the habitat range of *P. segnis* (Hosseini et al., 2014, Naderloo and Tuerkay, 2012, Rabaoui et al., 2015). Maps were generated using R Studio and formatted in QGIS (version 3.10.7-A Coruña).

Local population-level assessments were conducted by extracting THS probabilities from published *P. segnis* occurrence points (Castriota et al., 2022). To assess the species' persistence, we calculated the percentage change in THS between current and future scenarios (2050 RCP 4.5 and RCP 8.5) using the formula: $\% of\ change = (THS_{2050\ RCPi} - THS_{2022}) \times 100$, and presented as boxplots using the R Studio, "ggplot2" package (Wickham et al., 2016).

3. Results

Initially founded in the South-East Mediterranean (Tunisia, Sicily, Malta, Egypt to Turkey), *P. segnis* has expanded northward into Adriatic Sea (Italy) and southward into Gulf of Cadiz (Spain) since 2023 (Fig. 3A). In the Mediterranean, its thermal range spanned 9.9°C to 31.4°C (Fig. 3B). Winter temperatures (January-March) averaged 16.8 ± 1.9°C, ranging from 9.9°C to 20.4°C, and summer temperatures (July-September) averaged 28.3 ± 2.4°C, with a range of 19.1°C to 31.4°C.

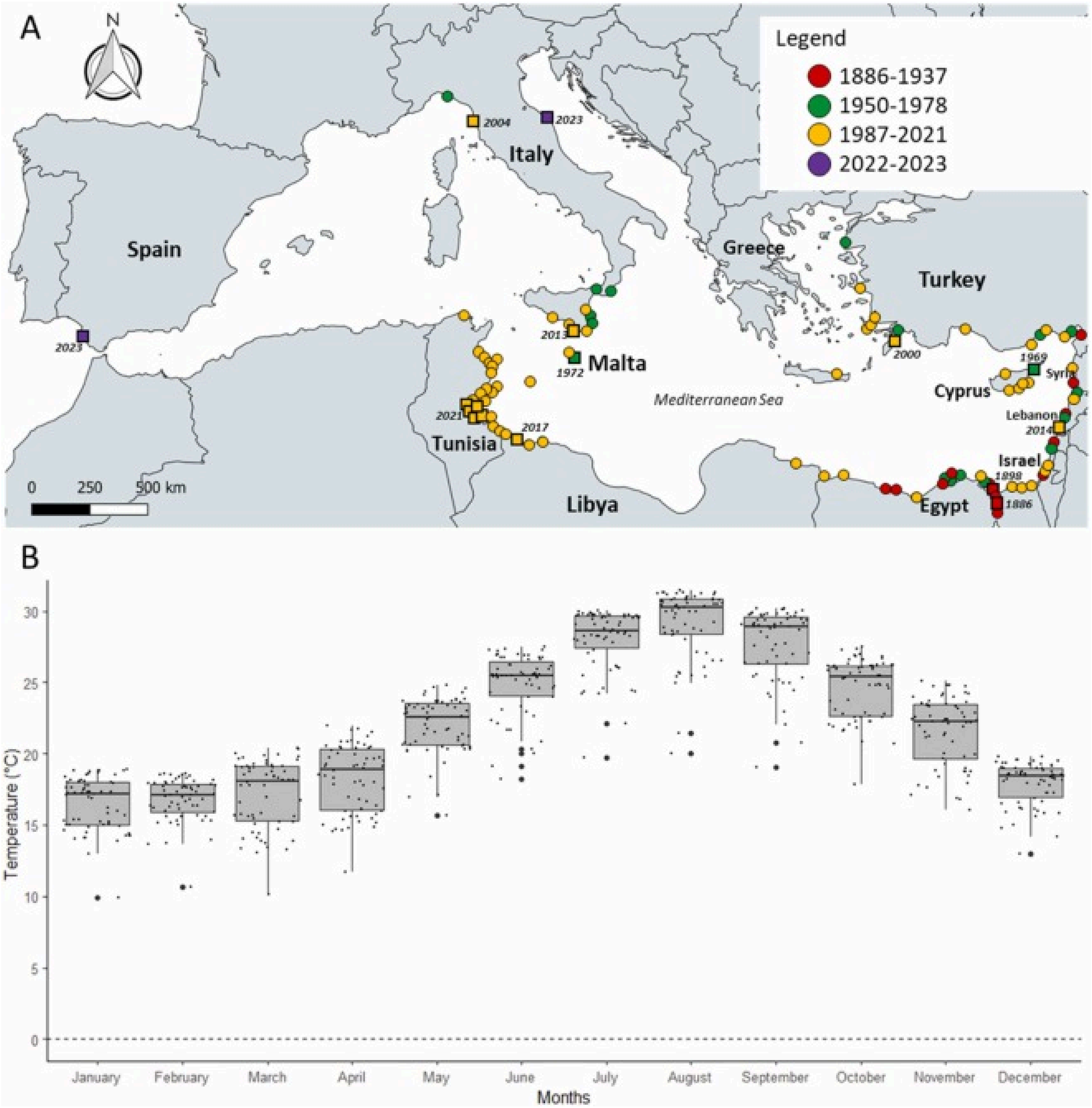
A
N
Legend
● 1886-1937
● 1950-1978
● 1987-2021
● 2022-2023
2004
2023
Italy
Spain
Greece
Turkey
2023
2013
Malta
1972
Mediterranean Sea
Cyprus
1969
Syria
Lebanon
2014
2000
2021
2017
Tunisia
Israel
1898
1886
Egypt
Libya
0 250 500 km
B
30
25
20
15
10
5
0
Temperature (°C)
January
February
March
April
May
June
July
August
September
October
November
December
Months

Fig. 3. (A) Current distribution of the Red Sea blue swimming crab *Portunus segnis* in the Mediterranean Sea, modified and updated from Castriota et al. (2022). The colored dots correspond to the invasion history of *P. segnis* defined by Castriota et al. (2022) and the squares represent the first report of the species for each country. New occurrence in 2023 from Adriatic Sea (Italy) was extracted from Grati et al. (2023), and for Spain from de de de Carvalho-Souza et al. (2023). (B) Boxplots showing the monthly thermal distribution of *P. segnis* in the Mediterranean Sea for the year 2022 extracted from the species occurrence points.

The thermal performance curve (TPC) of *P. segnis* rose from 0.016 ± 0.011 $mgO_2\ h^{-1}\ gWW^{-1}$ (12°C) to 0.238 ± 0.011 $mgO_2\ h^{-1}\ gWW^{-1}$ (34°C) (Fig. 4). Beyond 34°C, oxygen consumption decreased: 0.199 ± 0.015 $mgO_2\ h^{-1}\ gWW^{-1}$ (36°C); 0.019 ± 0.008 $mgO_2\ h^{-1}\ gWW^{-1}$ (40°C). Organisms succumbed at 7°C and 42°C (Fig. 3). The O’Neill regression was the "best" fitting model, with $r_{max} = 0.2337$ and $Q_{10} = 2.0563$ and Critical Thresholds: CT_{min} at 11.33°C and CT_{max} at 41.13°C. The TPC revealed a thermal optimum (T_{opt}) at 33.64°C (Fig. 4). The TPC regression was defined using the following equation:

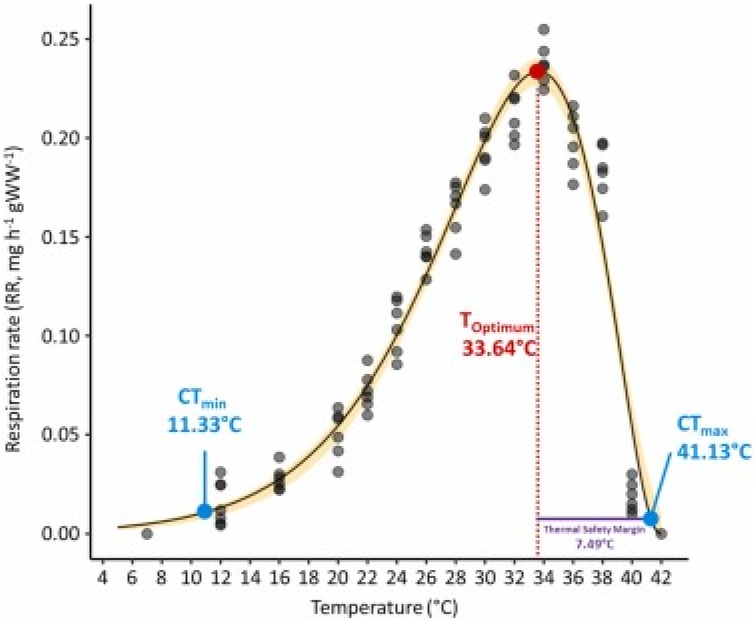

Fig. 4. Thermal Performance Curve (TPC) of the Red Sea swimming blue crab *Portunus segnis* according the O’Neill et al. (1972) model based on the respiration rates ($mgO_2\ h^{-1}\ gWW^{-1}$). The orange band represent the 95 % prediction limits. CT_{min}: critical threshold minimum, CT_{max}: critical threshold maximum, $T_{optimum}$: optimal temperature. The TPC was calculated following the O’Neill regression (O’Neill et al., 1972):.

$$Respiration\ Rate = r_{\max} \times \left[\left(\frac{CT_{\max}-T}{CT_{\max}-T_{opt}}\right)\right]^{\left\{\frac{1}{400}\times[(Q_{10}-1)\times(CT_{\max}-T_{opt})]^2\times\left[1+\sqrt{\left\{1+\left[\frac{40}{\left[(Q_{10}-1)\times\left(CT_{\max}-T_{opt}\right)\right]}\right]\right\}}\right]^2\right\}} \times \exp\left\{\left[\frac{1}{400}\times[(Q_{10}-1)\times(CT_{\max}-T_{opt})]^2\times\left[1+\sqrt{\left\{1+\left[\frac{40}{\left[(Q_{10}-1)\times\left(CT_{\max}-T_{opt}\right)\right]}\right]\right\}}\right]^2\times\left[\frac{\left(T-T_{opt}\right)}{CT_{\max}-T_{opt}}\right]\right]\right\} \text{ with}$$

r_{max} = 0.2337 ± 0.0043 (p value < $2*10^{-16}$); CT_{max} = 41.13 ± 0.6305 (p value < $2*10^{-16}$); T_{opt} = 33.6406 ± 0.2001 (p value < $2*10^{-16}$); Q_{10} = 2.0563 ± 0.0478 (p value < $2*10^{-16}$). The TPC shows a typical stenotherm specialist tropical species dynamics.

The Thermal Habitat Suitability (THS) probability of *P. segnis*, based on its thermal performance, revealed a North-South division in Mediterranean Sea thermal habitats (Fig. 5). Throughout winter and early spring (January to May), THS were unfavorable (THS < 0.2) across the Mediterranean. However, in summer-autumn (June to October), particularly in the Gulf of Gabès, Tunisia, southern Mediterranean coasts became suitable (THS > 0.6). During summer (June to September), all southern Mediterranean coasts provided favorable thermal habitats. In the North, THS were favorable along Spanish, Tyrrhenian and Adriatic Italian coasts to Greek Aegean coasts. For both future scenarios, an increase of THS was observed earlier in the year in the North-Western Mediterranean (Fig. 6).

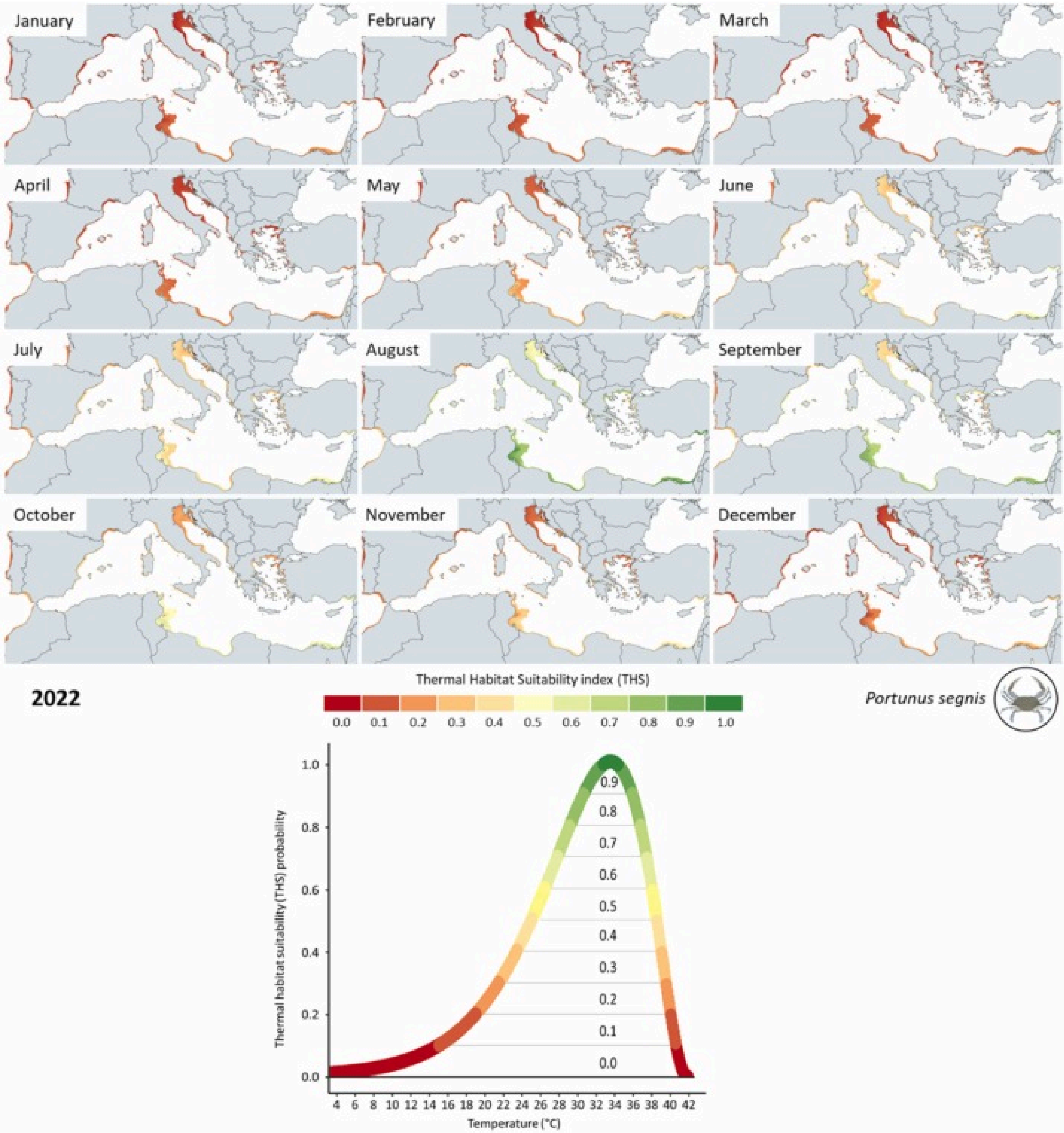

Fig. 5. Current predicted Thermal Habitat Suitability (THS) of *Portunus segnis* in the Mediterranean Sea based on the species metabolic performance. THS values at 0 = not favorable; and THS at 1 = favorable). Different colors (representing the species THS) would correspond to: optimum temperature range (THS > 0.8; green colored), lower and upper *pejus* ranges (0.5 < THS < 0.8; light green and yellow colored), *pessimum* (0.2 < THS < 0.5;

orange colored), and lethal ranges (THS < 0.2; red colored). The curve on the bottom represents the THS calculated using the TPC regression.

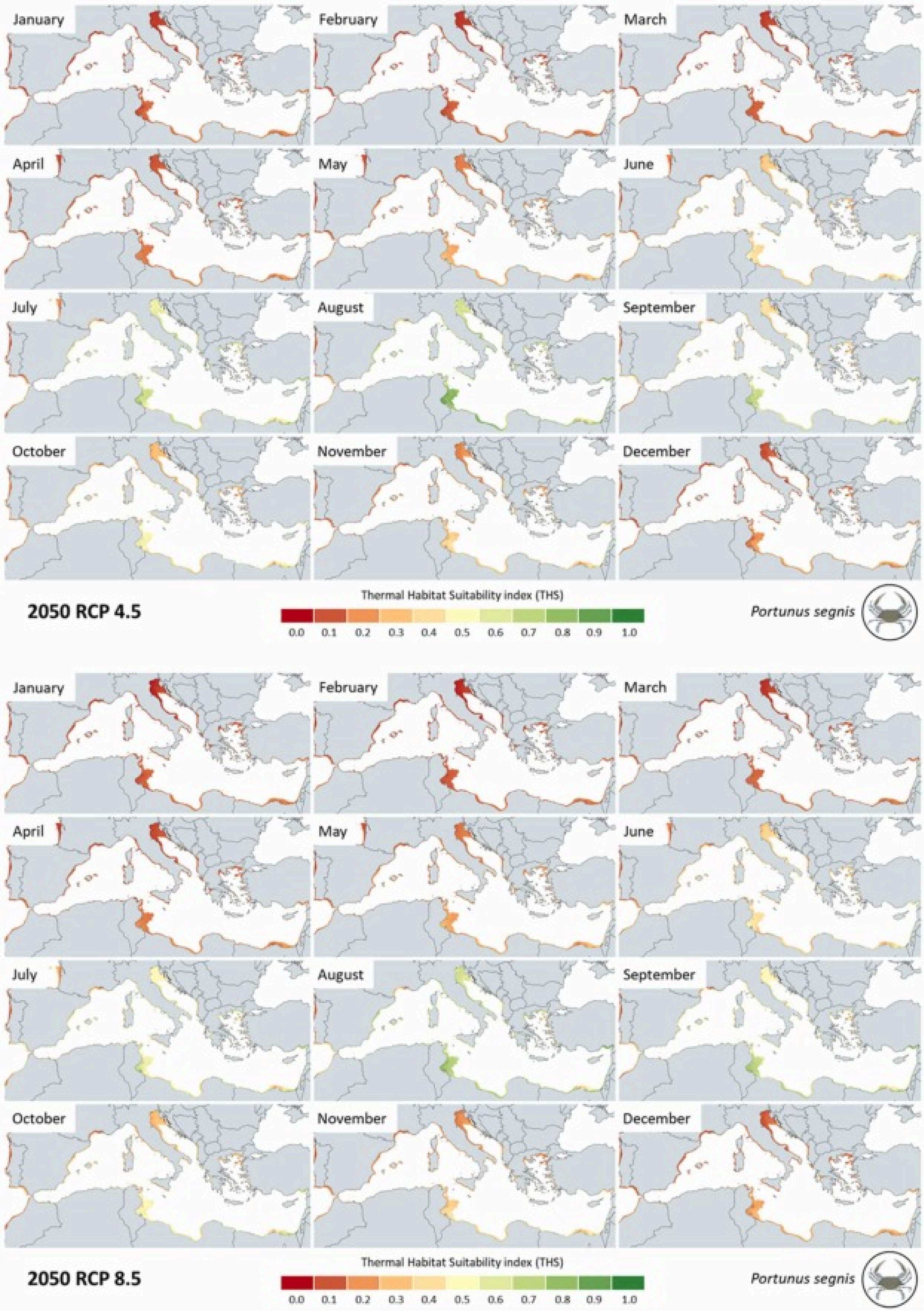
January
February
March
April
May
June
July
August
September
October
November
December
2050 RCP 4.5
Thermal Habitat Suitability index (THS)
0.0
0.1
0.2
0.3
0.4
0.5
0.6
0.7
0.8
0.9
1.0
Portunus segnis
January
February
March
April
May
June
July
August
September
October
November
December
2050 RCP 8.5
Thermal Habitat Suitability index (THS)
0.0
0.1
0.2
0.3
0.4
0.5
0.6
0.7
0.8
0.9
1.0
Portunus segnis

Fig. 6. Predicted future 2050 RCP 4.5 (on the top) and 2050 RCP 8.5 (on the bottom). RCP: Representative Concentration Pathway. Thermal Habitat Suitability (THS) of *Portunus segnis* in the Mediterranean Sea based on the species metabolic performance. THS values at 0 = not favorable; and THS at 1 = favorable). Different colors (representing the species THS) would correspond to: optimum temperature range (THS > 0.8; green colored), lower and upper *pejus* ranges (0.5 < THS < 0.8; light green and yellow colored), *pessimum* (0.2 < THS < 0.5; orange colored), and lethal ranges (THS < 0.2; red colored).

The change in Thermal Habitat Suitability (THS) probability between the 2050 RCP 4.5 scenario and the situation in 2022 varied across countries (Fig. 7A). In the Eastern Mediterranean (Israel, Egypt, Lebanon, Syria), THS was negative, indicating a decrease: −5.2 ± 6.4 %, −4.5 ± 8.9 %, −3.4 ± 5.2 %, −2.6 ± 6.0 %, respectively. Tunisia and Cyprus showed stability (THS ~ 1) with minor changes: −0.7 ± 9.7 %, 0.4 ± 7.6 %, respectively. Increases in THS were observed in Greece (+1.0 ± 6.2 %), Malta (+4.5 ± 7.4 %), and Italy (4.9 ± 8.3 %). At the Mediterranean Sea level, 2050 RCP 4.5 showed −0.7 ± 8.9 %, contrasting with +0.1 ± 8.7 % for the 2050 RCP 8.5 scenario (Fig. 7B). In 2050 RCP 8.5, Cyprus (-4.4 ± 9.5 %), Italy (-3.9 ± 5.7 %), and Malta (-3.1 ± 5.6 %) decreased in THS, while Syria, Greece, Turkey, Lebanon, Tunisia stabilized (-0.9–0.7 %). Egypt, Libya, and Israel had positive THS changes for this scenario (Fig. 6B).

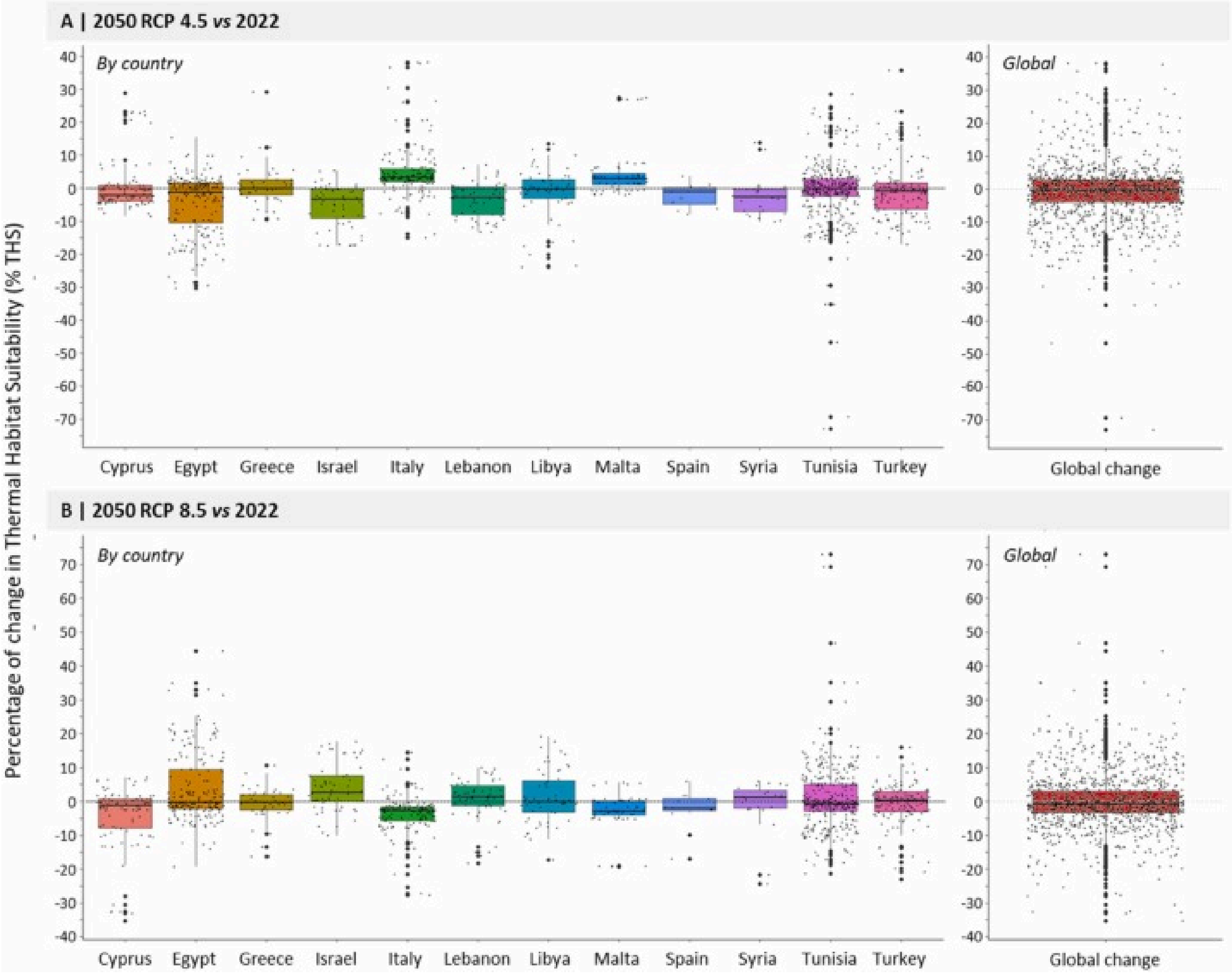

Fig. 7. Percentage of change (%) in Thermal Habitat Suitability (THS) probability in countries where *Portunus segnis* is currently established, (A) change between 2050 RCP 4.5 and 2022; (B) change between 2050 RCP 8.5 and 2022.

4. Discussion

Identifying thermal habitats for the Red Sea swimming blue crab *Portunus segnis* is vital for biodiversity conservation. Its invasive success in the Mediterranean is explained from its physiological plasticity and ability to adapt to global temperatures. Our study pioneered the revelation of *Portunus segnis* thermal tolerance through metabolic data, disclosing a thermal optimum at 33.64°C, CT_{min} at 11.33°C, and CT_{max} at 41.13°C, and contributing to understanding its distribution dynamics in response to temperature changes (Taheri et al., 2021). Limited existing data on this species heightened the novelty of our findings. Previous heart rate-focused research on *P. segnis* aligned with CTs founded in our study; CT_{min} at 5°C and CT_{max} at 40°C (Zainal and Noorani, 2019). Similar thermal patterns in another Portunidae, *Portunus pelagicus,* from the same native region as *P. segnis,* further validated our results (Azra et al., 2018). The TPC mirrored *P. segnis'* native temperature range, 10°C in winter to 38°C in summer, in the Red Sea and Indo-Pacific coasts (Zainal and Noorani, 2019). In the expanding Mediterranean Sea, our identified thermal range was 9.9°C to 31.4°C (Taheri et al., 2021), confirming the link between species occurrence and environmental suitability (Weber et al., 2017). Experimental thermal data presented in our study, aligned with observed occurrence density around the Mediterranean Sea (Castriota et al., 2022), indicated a positive temperature effect on *P. segnis* metabolism. Understanding temperature's influence on metabolism was vital for anticipating the species colonization of new areas, especially amid expected future temperature changes. Suitability maps extended beyond current records, aligning with the invasive potential of IAS, where heat tolerance played a crucial role (DeLong et al., 2018) providing a competitive edge, potentially causing ecological imbalances. This strengthened our confidence in anticipated implications and underscored the likely expansion of *P. segnis* to the highest Mediterranean Sea latitudes in the near future.

The TPC of *P. segnis* aligned with thermophilic species, exhibiting gradual metabolism increase up to the optimum temperature and abrupt decline thereafter (Verberk et al., 2016). In contrast, the other invasive Portunidae *Callinectes sapidus*, spreading in the Mediterranean Sea, displayed a typical TPC of temperate ectotherms (Marchessaux et al., 2022). *P. segnis*, limited to warmer regions in the eastern Mediterranean, is less thermo-tolerant, being a thermal specialist (stenothermal, Verberk et al., 2016), while *C. sapidus is* a generalist. However, *P. segnis* may outperform *C. sapidus* at higher temperatures. This distribution reflects the eastern Mediterranean's proximity to *P. segnis* native thermal habitat, while *C. sapidus*, being a generalist, tolerates a broader temperature range. The question arises as to which of the two invasive Portunidae species will prevail under climate change. *C. sapidus* may have an advantage in the Mediterranean Sea, global warming could have adverse effects on tropical ectothermic species, favoring temperate ectotherms (DeLong et al., 2018, Vasseur et al., 2014). We partially replied to this question because heat tolerance studies can reveal the adaptive strategies of invasive species, shedding light on their ability to cope with environmental stressors (DeLong et al., 2018). This knowledge is valuable for designing effective strategies to control and manage invasive populations.

Invasive species proliferation poses a significant threat to biodiversity (Bax et al., 2003, Costello et al., 2015). Emphasizing the role of heat tolerance is crucial in interpreting the mapped data. Aligning maps with heat tolerance provides insights into invasive species distribution, enhancing analysis precision and ecological understanding (Azra et al., 2018, Bates et al., 2013, Iftikar et al., 2010, Kelley, 2014). Our study, complementing previous investigations on rising temperatures' impact on blue crabs in the Mediterranean, enriches our understanding of how environmental changes influence invasive species. Thermal performance serves as a simple functional trait for identifying *P. segnis* suitable habitats. This knowledge contributes to biodiversity management, utilizing trait-based information to enhance conservation and ecosystem response forecasting to climate change (Anton et al., 2010, Truchy et al., 2015). Incorporating species functional traits into management tools and distribution models is crucial to inform nature conservation management strategies. Understanding the thermal suitability of IAS significantly enhances conservation efforts. This knowledge is crucial for developing effective Early Detection and Rapid Response (EDRR) systems, a critical strategy for managing IAS with early and cost-effective interventions (Reaser et al., 2020). Thermal suitability data integrates into predictive models to forecast IAS spread, aiding proactive measures and risk mapping strategies through Geographical Information Systems (GIS) (McMahon et al., 2021). Recent observations of *P. segnis* in Spain (de Carvalho-Souza et al., 2023) and in the Italian Adriatic Sea (Grati et al., 2023) are confirmed by predictions of suitability thermal habitats predicted in our study in these areas. Given that *P. segnis* began to invade the Mediterranean after the construction of the Suez Canal, and the fact that the species can go unnoticed in certain areas (low densities, areas little frequented by human uses (e.g. professional and recreational fishing, swimming, etc.)), it could be that *P. segnis* is already present in some of the areas where its expansion is predicted by our study. Also, although THS probabilities may increase due to the rise in temperature predicted for the future, *P. segnis* is still found today in areas tolerable for inhabiting these regions (e.g. particularly in the southern Mediterranean). In addition to temperature, its expansion could also be conditioned by other factors such as food availability, habitat suitability, the presence of predators, trophic and habitat competition with the Atlantic blue swimming crab *Callinectes sapidus*, etc. Our study therefore gives an indication of future trends in the temperature-based expansion of *P. segnis*, but further information on its ecology is yet to be explored.

The use of functional traits of an invasive species is essential for improving management tools for ecosystem conservation (Anton et al., 2010, Truchy et al., 2015). In addition to the temperature tolerance presented here in our study, it is necessary to determine the species' response to other environmental parameters (such as salinity, for example) and the species' ecology (e.g., diet, trophic relationships between native/invasive prey-predator, habitat use, reproduction, etc.). All these biotic and abiotic parameters influence the ecological niche of the species and therefore its potential expansion. Creating baseline layers based on species' functional traits using mechanistic approaches and integrating them into traditional management tools (such as correlative species distribution models) (Bosch-Belmar et al., 2022, Bosch-Belmar et al., 2021, Gamliel et al., 2020, Talluto et al., 2016), can generate more feasible and ecologically informed management measures, thereby increasing our ability to predict ecosystem responses to climate change (Mangano et al., 2019).

In the context of climate change, identifying the upper thermal limits of IAS is crucial for understanding climate-induced shifts in their thermal suitability. This foresight refines predictions of distribution changes, aiding adaptive conservation strategies. Knowledge of thermal limits is also valuable for selecting effective biocontrol agents and guiding community-based monitoring efforts through citizen science campaigns (Reaser et al., 2020). This information informs policy decisions, such as establishing quarantine zones or regulating the movement of goods between regions with differing thermal profiles (Venette et al., 2021). Given limited conservation resources, understanding where invasive species are likely to thrive allows for efficient allocation. To apply thermal information effectively, innovative solutions, including new technological approaches, are essential for advancing IAS management in a changing world. Thanks to this approach, we can give some recommendations for managers and decision-makers in two contexts:

(1) **The species is already present in the national territory**: Decision-makers can therefore use the results obtained in our study to implement measures to control the species on a national scale in areas where the thermal habitat is favorable. This concerns countries where it has been determined that in the future (e.g. 2050) the species will extend its range (refer to the Fig. 6) or maintain its population (Fig. 7). As an example, Tunisia is one of the main countries that has been harvesting and marketing *P. segnis* since 2014, with a view to controlling its populations nationwide. Thus, our results can also serve as evidence that the species will be maintained over the long term in Tunisia, where fisheries should continue in an attempt to control populations.

(2) **The species is not yet present in the national territory**: in this case, prediction maps for current (Fig. 5) and future scenarios (Fig. 6) enable decision-makers to determine the areas where the species could expand. This concerns Northern Mediterranean countries (for example Spain, France, Italy). Using predictive maps, decision-makers could, for example, set up early species detection programs (EDRR) using citizen science, or regular monitoring of potential risk areas. Monitoring and anticipating the

introduction of species on a national scale also requires precise, anticipatory regulations. France has already anticipated the potential arrival of *P. segnis*: on March 2, 2023, the French government issued a ministerial decree (n° TREL2138499A) classifying the blue crab *P. segnis* as a level 2 species, i.e. "*prohibited from introduction into the national territory, possession, transport, peddling, use, exchange, offering for sale, sale or purchase*". The species is therefore under surveillance on the French Mediterranean coast, and specific monitoring is currently underway to anticipate its possible arrival.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

This study is part of the BLEU-ADAPT project supported by Interreg Italy-Tunisia Program 2014–2020, Ref. n° IS_3.1_021. Guillaume Marchessaux is supported by the Project “National Biodiversity Future Center - NBFC”; National Recovery and Resilience Plan (NRRP), Mission 4 Component 2 Investment 1.4 - Call for tender No. 3138 of 16 December 2021, rectified by Decree n.3175 of 18 December 2021 of Italian Ministry of University and Research funded by the European Union – NextGenerationEU. We would like to thank all the fishers from the Bizerte Lagoon for their help in sampling and for their availability to collaborate in this study. The authors would like to thank the editor and the two reviewers for their comments and suggestions.

Appendix A. Supplementary material

Download: Download Word document (46KB)

Supplementary material

Recommended articles

Data Availability

Data will be made available on request.

References

Angilletta and Angilletta, 2009 M.J. Angilletta, Michael James Angilletta
Thermal adaptation: a theoretical and empirical synthesis
Oxf. Univ. Press, 289 (2009)
Google Scholar ↗

Anton et al., 2010 C. Anton, J. Young, P.A. Harrison, M. Musche, G. Bela, C.K. Feld, R. Harrington, J.R. Haslett, G. Pataki, M.D. Rounsevell
Research needs for incorporating the ecosystem service approach into EU biodiversity conservation policy
Biodivers. Conserv., 19 (2010), pp. 2979-2994
Crossref ↗ View in Scopus ↗ Google Scholar ↗

Azra et al., 2018 M.N. Azra, J.-C. Chen, M. Ikhwanuddin, A.B. Abol-Munafi
Thermal tolerance and locomotor activity of blue swimmer crab *Portunus pelagicus* instar reared at different temperatures
J. Therm. Biol., 74 (2018), pp. 234-240
View PDF View article View in Scopus ↗ Google Scholar ↗

Bannar-Martin et al., 2018 K.H. Bannar-Martin, C.T. Kremer, S.K.M. Ernest, M.A. Leibold, H. Auge, J. Chase, S.A.J. Declerck, N. Eisenhauer, S. Harpole, H. Hillebrand, F. Isbell, T. Koffel, S. Larsen, A. Narwani, J.S. Petermann, C. Roscher, J.S. Cabral, S.R. Supp
Integrating community assembly and biodiversity to better understand ecosystem function: the Community Assembly and the Functioning of Ecosystems (CAFE) approach
Ecol. Lett., 21 (2018), pp. 167-180, 10.1111/ele.12895 ↗
View in Scopus ↗ Google Scholar ↗

Bates et al., 2013 A.E. Bates, C.M. McKelvie, C.J. Sorte, S.A. Morley, N.A. Jones, J.A. Mondon, T.J. Bird, G. Quinn
Geographical range, heat tolerance and invasion success in aquatic species
Proc. R. Soc. B Biol. Sci., 280 (2013), p. 20131958
Crossref ↗ View in Scopus ↗ Google Scholar ↗

Bax et al., 2003 N. Bax, A. Williamson, M. Aguero, E. Gonzalez, W. Geeves
Marine invasive alien species: a threat to global biodiversity
Mar. Policy, 27 (2003), pp. 313-323
View PDF View article View in Scopus ↗ Google Scholar ↗

Ben Souissi et al., in press Ben Souissi, J., Ghanem, R., Rjiba, W., Ounifi-Ben Amor, K., Neloufi, F., Ben Hafsia, I., Alaya, S., in press. Managing the explosion of the blue swimming crab Portunus segnis (Forskål, 1775) in Tunisia, in: Compilation and Analysis of Fisheries Management Responses to Aquatic Non Indigenous Species (Aquatic NIS) in the Context of Climate Change.

Google Scholar ↗

Blackburn et al., 2011 T.M. Blackburn, P. Pyšek, S. Bacher, J.T. Carlton, R.P. Duncan, V. Jarošík, J.R. Wilson, D.M. Richardson

A proposed unified framework for biological invasions

Trends Ecol. Evol., 26 (2011), pp. 333-339

View PDF View article View in Scopus ↗ Google Scholar ↗

Bosch-Belmar et al., 2021 M. Bosch-Belmar, C. Giommi, G. Milisenda, A. Abbruzzo, G. Sarà

Integrating functional traits into correlative species distribution models to investigate the vulnerability of marine human activities to climate change

Sci. Total Environ. (2021), Article 149351

View PDF View article View in Scopus ↗ Google Scholar ↗

Bosch-Belmar et al., 2022 M. Bosch-Belmar, S. Piraino, G. Sarà

Predictive Metabolic Suitability Maps for the Thermophilic Invasive Hydroid *Pennaria disticha* Under Future Warming Mediterranean Sea Scenarios

Front. Mar. Sci., 9 (2022), pp. 1-10, 10.3389/fmars.2022.810555 ↗

Google Scholar ↗

de Carvalho-Souza et al., 2023 G.F. de Carvalho-Souza, J.A. Cuesta, D. Arana, C. Lobato, E. González-Ortegón

Westward range expansion of the blue swimmer crab *Portunus segnis* (Forskål, 1775)(Crustacea, Decapoda, Portunidae) into Atlantic European waters

BioInvasions Rec., 12 (2023)

Google Scholar ↗

Castriota et al., 2022 L. Castriota, M. Falautano, T. Maggio, P. Perzia

The Blue Swimming Crab *Portunus segnis* in the Mediterranean Sea: Invasion Paths, Impacts and Management Measures

Biology, 11 (2022), p. 1473

Crossref ↗ View in Scopus ↗ Google Scholar ↗

Costello et al., 2015 M.J. Costello, S. Claus, S. Dekeyzer, L. Vandepitte, E.O. Tuama, D. Lear, H. Tyler-Walters

Biological and ecological traits of marine species

PeerJ, 3 (2015), Article e1201

Crossref ↗ Google Scholar ↗

DeLong et al., 2018 J.P. DeLong, G. Bachman, J.P. Gibert, T.M. Luhring, K.L. Montooth, A. Neyer, B. Reed

Habitat, latitude and body mass influence the temperature dependence of metabolic rate

Biol. Lett., 14 (2018), p. 20180442, 10.1098/rsbl.2018.0442 ↗

View in Scopus ↗ Google Scholar ↗

Desforges et al., 2023 J.E. Desforges, K. Birnie-Gauvin, F. Jutfelt, K.M. Gilmour, E.J. Eliason, T.L. Dressler, D.J. McKenzie, A.E. Bates, M.J. Lawrence, N. Fangue, S.J. Cooke

The ecological relevance of critical thermal maxima methodology for fishes

J. Fish. Biol., 102 (2023), pp. 1000-1016, 10.1111/jfb.15368 ↗

View in Scopus ↗ Google Scholar ↗

Gamliel et al., 2020 I. Gamliel, Y. Buba, T. Guy-Haim, T. Garval, D. Willette, G. Rilov, J. Belmaker

Incorporating physiology into species distribution models moderates the projected impact of warming on selected Mediterranean marine species

Ecography, 43 (2020), pp. 1090-1106

Crossref ↗ View in Scopus ↗ Google Scholar ↗

Grati et al., 2023 F. Grati, C. Froglia, J.B. Souissi, L. Bolognini, E. Azzurro

The blue swimming crab *Portunus segnis* (Forskål, 1775) reaches the Adriatic Sea: A distant and disjointed occurrence

BioInvasions Rec., 12 (2023)

Google Scholar ↗

Hosseini et al., 2014 M. Hosseini, J. Pazooki, M. Safaei

Size at maturity, sex ratio and variant morphometrics of blue swimming crab *Portunus segnis* (Forskal, 1775) from Boushehr coast (Persian Gulf)

J. Mar. Sci. Res. Dev., 4 (2014), p. 149

Crossref ↗ Google Scholar ↗

Iftikar et al., 2010 F.I. Iftikar, J. MacDonald, A.J. Hickey

Thermal limits of portunid crab heart mitochondria: Could more thermo-stable mitochondria advantage invasive species?

J. Exp. Mar. Biol. Ecol., 395 (2010), pp. 232-239

View PDF View article View in Scopus ↗ Google Scholar ↗

Kelley, 2014 A.L. Kelley

The role thermal physiology plays in species invasion

Conserv. Physiol., 2 (2014)

Google Scholar ↗

Kooijman and Kooijman, 2010 B. Kooijman, S. Kooijman

Dynamic energy budget theory for metabolic organisation

Cambridge university press (2010)

Google Scholar ↗

Mangano et al., 2019 M.C. Mangano, F. Ape, S. Mirto

The role of two non-indigenous serpulid tube worms in shaping artificial hard substrata communities: case study of a fish farm in the central Mediterranean Sea

Aquac. Environ. Interact., 11 (2019), pp. 41-51

Crossref ↗ View in Scopus ↗ Google Scholar ↗

Marchessaux et al., 2022 G. Marchessaux, M. Bosch-Belmar, L. Cilenti, N. Lago, M.C. Mangano, N. Marsiglia, G. Sarà

The invasive blue crab *Callinectes sapidus* thermal response: predicting metabolic suitability maps under future warming Mediterranean scenarios

Front. Mar. Sci., 9 (2022), Article 1055404

View in Scopus ↗ Google Scholar ↗

Marchessaux et al., 2023 G. Marchessaux, M.C. Mangano, S. Bizzarri, C. M'Rabet, E. Principato, N. Lago, D. Veyssiere, M. Garrido, S.B. Scyphers, G. Sarà

Invasive blue crabs and small-scale fisheries in the Mediterranean sea: local ecological knowledge, impacts and future management

Mar. Policy, 148 (2023), Article 105461

View PDF View article View in Scopus ↗ Google Scholar ↗

Matzelle et al., 2015 A.J. Matzelle, G. Sarà, V. Montalto, M. Zippay, G.C. Trussell, B. Helmuth

A bioenergetics framework for integrating the effects of multiple stressors: opening a 'black box'in climate change research

Am. Malacol. Bull., 33 (2015), pp. 150-160

Crossref ↗ Google Scholar ↗

McMahon et al., 2021 D.E. McMahon, A.K. Urza, J.L. Brown, C. Phelan, J.C. Chambers
Modelling species distributions and environmental suitability highlights risk of plant invasions in western United States
Divers. Distrib., 27 (2021), pp. 710-728, 10.1111/ddi.13232 ↗
View in Scopus ↗ Google Scholar ↗

Naderloo and Tuerkay, 2012 R. Naderloo, M. Tuerkay
Decapod crustaceans of the littoral and shallow sublittoral Iranian coast of the Persian Gulf: faunistics, biodiversity and zoogeography
Zootaxa, 3374 (2012), pp. 1-67
View in Scopus ↗ Google Scholar ↗

O’Neill et al., 1972 R.V. O’Neill, R.A. Goldstein, H.H. Shugart, J.B. Mankin, R.S. Booth
Terrestrial ecosystem energy model, eastern deciduous forest biome
Int Biol. Program Memo. Rep. (1972)
72–19
Google Scholar ↗

Padfield and O’Sullivan, 2020 Padfield, D., O’Sullivan, H., 2020. rTPC: Functions for fitting thermal performance curves.
Google Scholar ↗

Prusina et al., 2014 I. Prusina, G. Sarà, M. De Pirro, Y.-W. Dong, G.-D. Han, B. Glamuzina, G.A. Williams
Variations in physiological responses to thermal stress in congeneric limpets in the Mediterranean Sea
J. Exp. Mar. Biol. Ecol., 456 (2014), pp. 34-40
View PDF View article View in Scopus ↗ Google Scholar ↗

Rabaoui et al., 2015 L. Rabaoui, M. Arculeo, L. Mansour, S. Tlig-Zouari
Occurrence of the lessepsian species *Portunus segnis* (Crustacea: Decapoda) in the Gulf of Gabes (Tunisia): first record and new information on its biology and ecology
Cah. Biol. Mar., 56 (2015), pp. 169-175
View in Scopus ↗ Google Scholar ↗

Rahel et al., 2008 F.J. Rahel, B. Bierwagen, Y. Taniguchi

Managing aquatic species of conservation concern in the face of climate change and invasive species

Conserv. Biol., 22 (2008), pp. 551-561, 10.1111/j.1523-1739.2008.00953.x ↗

View in Scopus ↗ Google Scholar ↗

Reaser et al., 2020 J.K. Reaser, S.W. Burgiel, J. Kirkey, K.A. Brantley, S.D. Veatch, J. Burgos-Rodríguez

The early detection of and rapid response (EDRR) to invasive species: a conceptual framework and federal capacities assessment

Biol. Invasions, 22 (2020), pp. 1-19

Crossref ↗ Google Scholar ↗

Roy et al., 2023 Roy, H.E., Pauchard, A., Stoett, P., Renard Truong, T., Bacher, S., Galil, B.S., Hulme, P.E., Ikeda, T., Sankaran, K.V., McGeoch, M.A., 2023. IPBES Invasive Alien Species Assessment: Summary for Policymakers.

Google Scholar ↗

Rubalcaba et al., 2020 J.G. Rubalcaba, W.C.E.P. Verberk, A.J. Hendriks, B. Saris, H.A. Woods

Oxygen limitation may affect the temperature and size dependence of metabolism in aquatic ectotherms

Proc. Natl. Acad. Sci., 117 (2020), pp. 31963-31968, 10.1073/pnas.2003292117 ↗

View in Scopus ↗ Google Scholar ↗

Sinclair et al., 2016 B.J. Sinclair, K.E. Marshall, M.A. Sewell, D.L. Levesque, C.S. Willett, S. Slotsbo, Y. Dong, C.D.G. Harley, D.J. Marshall, B.S. Helmuth, R.B. Huey

Can we predict ectotherm responses to climate change using thermal performance curves and body temperatures?

Ecol. Lett., 19 (2016), pp. 1372-1385, 10.1111/ele.12686 ↗

View in Scopus ↗ Google Scholar ↗

Sokolova et al., 2012 I.M. Sokolova, M. Frederich, R. Bagwe, G. Lannig, A.A. Sukhotin

Energy homeostasis as an integrative tool for assessing limits of environmental stress tolerance in aquatic invertebrates

Mar. Environ. Res., 79 (2012), pp. 1-15

View PDF View article View in Scopus ↗ Google Scholar ↗

Taheri et al., 2021 S. Taheri, B. Naimi, C. Rahbek, M.B. Araújo

Improvements in reports of species redistribution under climate change are required

Sci. Adv., 7 (2021), Article eabe1110

View in Scopus ↗ Google Scholar ↗

Talluto et al., 2016 M.V. Talluto, I. Boulangeat, A. Ameztegui, I. Aubin, D. Berteaux, A. Butler, F. Doyon, C.R. Drever, M.-J. Fortin, T. Franceschini

Cross-scale integration of knowledge for predicting species ranges: a metamodelling framework

Glob. Ecol. Biogeogr., 25 (2016), pp. 238-249

Crossref ↗ View in Scopus ↗ Google Scholar ↗

Truchy et al., 2015 A. Truchy, D.G. Angeler, R.A. Sponseller, R.K. Johnson, B.G. McKie

Linking biodiversity, ecosystem functioning and services, and ecological resilience: towards an integrative framework for improved management, in

Advances in Ecological Research, Elsevier (2015), pp. 55-96

View PDF View article View in Scopus ↗ Google Scholar ↗

Tureli and Yesilyurt, 2017 C. Tureli, I.N. Yesilyurt

Reproductive biology of blue swimming crab, *Portunus segnis* (Forskal, 1775) in Yumurtalık Cove, Northeastern Mediterranean, Turkey

Mediterr. Mar. Sci., 18 (2017), pp. 424-432

Crossref ↗ View in Scopus ↗ Google Scholar ↗

Vasseur et al., 2014 D.A. Vasseur, J.P. DeLong, B. Gilbert, H.S. Greig, C.D.G. Harley, K.S. McCann, V. Savage, T.D. Tunney, M.I. O'Connor

Increased temperature variation poses a greater risk to species than climate warming

Proc. R. Soc. B Biol. Sci., 281 (2014), p. 20132612, 10.1098/rspb.2013.2612 ↗

View in Scopus ↗ Google Scholar ↗

Venette et al., 2021 R.C. Venette, D.R. Gordon, J. Juzwik, F.H. Koch, A.M. Liebhold, R.K. Peterson, S.E. Sing, D. Yemshanov

Early intervention strategies for invasive species management: connections between risk assessment, prevention efforts, eradication, and other rapid responses

Invasive Species . Rangel. U. S. Compr. Sci. Synth. U. S. . Sect. (2021), pp. 111-131

Crossref ↗ View in Scopus ↗ Google Scholar ↗

Verberk et al., 2016 W.C.E.P. Verberk, F. Bartolini, D.J. Marshall, H. Pörtner, J.S. Terblanche, C.R. White, F. Giomi

Can respiratory physiology predict thermal niches?

Ann. N. Y. Acad. Sci., 1365 (2016), pp. 73-88, 10.1111/nyas.12876 ↗

View in Scopus ↗ Google Scholar ↗

Wallingford et al., 2020 P.D. Wallingford, T.L. Morelli, J.M. Allen, E.M. Beaury, D.M. Blumenthal, B.A. Bradley, J.S. Dukes, R. Early, E.J. Fusco, D.E. Goldberg
Adjusting the lens of invasion biology to focus on the impacts of climate-driven range shifts
Nat. Clim. Change, 10 (2020), pp. 398-405
Crossref ↗ View in Scopus ↗ Google Scholar ↗

Weber et al., 2017 M.M. Weber, R.D. Stevens, J.A.F. Diniz-Filho, C.E.V. Grelle
Is there a correlation between abundance and environmental suitability derived from ecological niche modelling? A meta-analysis.
Ecography, 40 (2017), pp. 817-828
Crossref ↗ View in Scopus ↗ Google Scholar ↗

Wickham et al., 2016 H. Wickham, W. Chang, M.H. Wickham
Package 'ggplot2.' Create Elegant Data Vis. Using Gramm
Graph. Version, 2 (2016), pp. 1-189
View in Scopus ↗ Google Scholar ↗

Zainal and Noorani, 2019 K. Zainal, A. Noorani
Temperature dependence of the heart rates in the blue swimming crab *Portunus segnis* (Forskal, 1775)
Arab. J. Sci. Eng., 44 (2019), pp. 6259-6265, 10.1007/s13369-019-03826-6 ↗
View in Scopus ↗ Google Scholar ↗

Cited by (1)

Heat hardening improves thermal tolerance in abalone, without the trade-offs associated with chronic heat exposure

2024, Journal of Thermal Biology

Show abstract

[1] ORCID 0000–0001-5557–2274

[2] ORCID 0000–0001-6947–582X

[3] ORCID 0000–0001-8590–4719

[4] ORCID 0000–0001-8416–8342

[5] ORCID 0000–0003-1740–6093

[6] ORCID 0000–0003-1761–4204

[7] ORCID 0000–0002-7658–5274