

Research Article

Horizon scanning for potentially invasive non-native marine species to inform trans-boundary conservation management – Example of the northern Gulf of Mexico

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Abstract

Prevention of non-native species introductions and establishment is essential to avoid adverse impacts of invasive species in marine environments. To identify potential new invasive species and inform non-native species management options for the northern Gulf of Mexico (Alabama, Mississippi, Louisiana, Texas), 138 marine species were risk screened for current and future climate conditions using the Aquatic Species

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Invasiveness Screening Kit. Species were risk-ranked as low, medium, high, and very high risk based on separate (calibrated) thresholds for fishes, tunicates, and invertebrates. In the basic screening, 15 fishes, two tunicates, and 26 invertebrates were classified as high or very high risk under current climate conditions. Whereas, under future climate conditions, 16 fishes, three tunicates, and 33 invertebrates were classified as high or very high risk. Very high risk species included: California scorpionfish *Scorpaena guttata*, red scorpionfish *Scorpaena scrofa*, purple whelk *Rapana venosa*, and Santo Domingo false mussel *Mytilopsis sallei* under both current and future climates, with weedy scorpionfish *Rhinopias frondosa*, Papuan scorpionfish *Scorpaenopsis papuensis*, daggertooth pike conger *Muraenesox cinereus*, yellowfin scorpionfish *Scorpaenopsis neglecta*, tassled scorpionfish *Scorpaenopsis oxycephalus*, brush-clawed shore crab *Hemigrapsus takanoi*, honeycomb oyster *Hyotissa hyotis*, carinate rock shell *Indothais lacera*, and Asian green mussel *Perna viridis* under climate change conditions only. This study provides evidence to inform trans-boundary management plans across the five Gulf of Mexico states to prevent, detect, and respond rapidly to new species arrivals.

Key words: Alien species, Aquatic Species Invasiveness Screening Kit (AS-ISK), biodiversity, early detection, introduction vectors, risk analysis

Introduction

Impacts of invasive species are recognized as one of the major drivers of biodiversity loss globally (Molnar et al. 2008; Vilà et al. 2011; IPBES 2019). Indeed, the Convention on Biological Diversity (CBD; <https://www.cbd.int/>) named invasive species as one of the main drivers of biodiversity loss due to impacts on habitats, ecosystem services, and native species, leading to a decision (15/27) specific to Invasive Alien Species in 2022 (Secretariat of the CBD 2022, <https://www.cbd.int/meetings/COP-15>). As the rate of introduction and establishment of invasive species has been increasing substantially during recent centuries (Hulme et al. 2009; Seebens et al. 2017), the response of invasive species management schemes has increasingly included the development and implementation of control measures that attempt to reduce populations (Britton et al. 2010, 2011; Booy et al. 2017), diminish further spread, and mitigate harmful impacts (e.g. Coutts and Forrest 2007; Frazer et al. 2012; Morris 2012). However, as control and eradication techniques address species at the stage at which they are already established, these approaches typically incur high costs (Britton et al. 2011; Booy et al. 2017), are labor-intensive (Leung et al. 2002; Martins et al. 2006), can potentially decimate native flora and fauna populations (Coutts and Forrest 2007), and may not result in permanent eradication of the target species (Coutts and Forrest 2007; Parkes and Panetta 2009), which is particularly true for the marine environment where eradication is rarely an option (Sambrook et al. 2014; Lehtiniemi et al. 2015).

Therefore, invasive species management and associated conservation efforts are shifting from reactionary to preventative approaches (Sutherland and Woodroff 2009; Sutherland et al. 2018), and thus rely on non-native species risk analysis, which consists of risk identification (=risk screening), full risk assessment, and risk management and risk communication (Copp et al. 2005a; Baker et al. 2008). As such, interest in risk screening to predict potential future invaders is increasing (Kolar and Lodge 2001; Copp et al. 2005a; Sutherland and Woodroff 2009; Vander Zanden et al. 2010) as a means with which to inform national and trans-boundary conservation management strategies, including early detection and rapid response programs. As part of an early detection strategy, horizon-scanning

with a risk screening tool provides a means with which to identify and prioritize potential future invasive species (Kolar and Lodge 2001; Copp et al. 2009; Roy et al. 2015, 2019a; Gallardo et al. 2016) and thereby informs decision-makers about the allocation of resources to counteract those non-native species identified as carrying a high (or even moderate) risk of invasiveness, which will then be subject to a follow-up risk assessment. This involves detailed examination of the likelihood and magnitude of risk of species introduction, establishment, dispersal, and impacts (Copp et al. 2005a; Baker et al. 2008; Mumford et al. 2010).

Horizon scanning involves the systematic examination of ‘horizon species’, i.e. non-native species not currently in the risk assessment area but likely to enter it in the foreseeable future due to proximity or by way of human assistance (Vilizzi et al. 2022a). With regard to the aquatic environments, human assistance involves various introduction vectors such as ballast-water exchanges, the ornamental aquatics trade, and releases of pet fish to open waters by private citizens (Semmens et al. 2004; Copp et al. 2005c; Tidbury et al. 2021). Horizon species that are risk ranked as posing a high-risk of being invasive can then be prioritized for comprehensive (full) assessment of risks and subsequent management and risk communication (Copp et al. 2005a, 2016; Sutherland et al. 2011; Roy et al. 2014, 2015). The outcomes of a horizon-scanning exercise can serve to inform future decision making, public awareness, and policy development as well as help direct resources to detect and manage species of greatest concern (Roy et al. 2015; Tsiamis et al. 2020). Applications of horizon-scanning procedures have been carried out at national (e.g. Copp et al. 2009; Roy et al. 2014; Lucy et al. 2020), regional (e.g. Dodd et al. 2019, 2022), multi-national (e.g. Roy et al. 2015; Gallardo et al. 2016; Clarke et al. 2020), and global (e.g. Vilizzi et al. 2021, 2022a) scales. The main focus of horizon scanning is to identify species that could potentially exert adverse impacts on native biodiversity (Roy et al. 2014; Lucy et al. 2020), human health (Peyton et al. 2019), and ecosystem services (Roy et al. 2015), as well as impacts to habitats and species of conservation concern (Roy et al. 2014; Lucy et al. 2020; Tsiamis et al. 2020). These applications have encompassed freshwater (e.g. Tarkan et al. 2017), marine (e.g. Lyons et al. 2020a; Tsiamis et al. 2020; Tidbury et al. 2021), and terrestrial environments (e.g. Gordon et al. 2008; Kopecký et al. 2019), targeting either individual species (e.g. Moghaddas et al. 2020; Dodd et al. 2022), or a broad range of species and taxonomic groups (e.g. Roy et al. 2014; Peyton et al. 2019; Vilizzi et al. 2019, 2021).

Previous studies have recommended incorporating trans-boundary collaboration initiatives into conservation and natural resources management strategies in order to achieve large-scale management goals such as preservation of threatened and endangered species (Kark et al. 2015; Mason et al. 2020), mitigation of biodiversity loss (Kark et al. 2015; Liu et al. 2020; Mason et al. 2020), and protection of sensitive habitats and ecosystems (Kark et al. 2015). Management of invasive species has been identified as benefitting from trans-boundary collaboration because this method facilitates coordinated approaches to reduce the likelihood of a species introduced to one jurisdiction spreading to other neighboring areas (Sambrook et al. 2014; Graham et al. 2018; Roy et al. 2019a). While horizon scanning of existing and future non-native species has been used in Europe as part of non-native species management programs for over a decade (e.g. Copp et al. 2009; Caffrey et al. 2014; Roy et al. 2014, 2015; Gallardo et al. 2016; Matthews et al. 2017; Piria et al. 2017; Oficialdegui et al. 2023), the use of this approach elsewhere, such as Asia (e.g. Li et al. 2017; Clarke et al. 2020; Wei et al. 2021) and in the U.S. (e.g. Kolar and Lodge 2001; Mack et al. 2002; Lawson et al. 2015; Goldsmit et al. 2021) has been relatively limited. Recent adoption of horizon

scanning in the U.S. has led to numerous ongoing state, regional, and national efforts to initiate and facilitate multi-jurisdictional policy and decision-making (e.g. Tuckett et al. 2016; Hill et al. 2018). Recently, the importance of horizon scanning in the selection of non-native species has recently been emphasized in applications of Weed Risk Assessment (WRA) type decision-support tools to identify high-risk species (i.e. Vilizzi et al. 2022a); however, only 45% of the 78 screening studies they reviewed included horizon species (5.1% horizon species only, 39.7% both extant and horizon species, 51.3% existing species, and 3.8% without mention of species status).

In the marine environment, the spread of invasive species has been attributed largely to increased global connectivity through transport, shipping, and trade (Cohen and Carlton 1997; Keller et al. 2011; Seebens et al. 2013, 2017). The primary vectors for the rise in introductions are ballast water exchange (Carlton and Geller 1993; Ruiz et al. 1997; Gollasch 2008), hull fouling (Drake and Lodge 2007), aquaculture (Molnar et al. 2008), and releases of aquarium species (Chan et al. 2019), such as the Indo-Pacific red lionfish *Pterois volitans* (Linnaeus, 1758) in the western Atlantic (Semmens et al. 2004). In general, the incidence of the introduction and initial colonization by invasive species tends to be higher within major shipping ports and harbors relative to areas with fewer shipping activities (Molnar et al. 2008; O'Shaughnessy et al. 2020; Tidbury et al. 2016, 2021). Secondary spread of aquatic invasive species from these areas is then primarily via smaller mobile vectors, such as leisure and fishing craft (Anderson et al. 2014), movement by anglers (Hickley and Chare 2004; Copp et al. 2007; Smith et al. 2020), as contaminants of native aquatic species consignments (Copp et al. 2017), releases by the general public (Copp et al. 2005c, 2017), and in some cases through natural dispersal to neighboring ports or natural areas (Clarke Murray et al. 2012; Epstein and Smale 2018; Peters et al. 2019).

To achieve non-native species policy objectives, which increasingly include horizon scanning (e.g. Copp et al. 2009; Roy et al. 2014, 2015, 2019a, 2019b), the aim of the present work was to implement a risk screening study of potential future non-native species to identify those likely to pose a high risk of being invasive in the northern Gulf of Mexico over the next decade. To this end, a large number of marine species were screened using a widely employed WRA-type decision-support tool based on a comprehensive and multi-tiered horizon-scan (*sensu* Vilizzi et al. 2022a) that includes initial assessments of the potential of non-native aquatic species to cause ecological, economic, and/or social harm under both current and future climate conditions. The outcomes of the screening of horizon species for their risks of arrival, establishment, dispersal, and impacts under current and future climate conditions were used to identify the highest-risk species with a view to guide policy and management and focus conservation aims at a regional level towards development of comprehensive risk and economic analyses, appropriate prevention measures (e.g. regulation), and early detection and rapid response plans.

Methods

Risk assessment area

The risk assessment area consisted of the northern Gulf of Mexico, which is inclusive of coastal Alabama, Mississippi, Louisiana, and Texas (Fig. 1), with coastal and marine habitats out to the edge of the continental shelf considered. Notably, the Florida Gulf Coast was not included in the risk assessment area because horizon scanning for potentially invasive coastal and marine species has

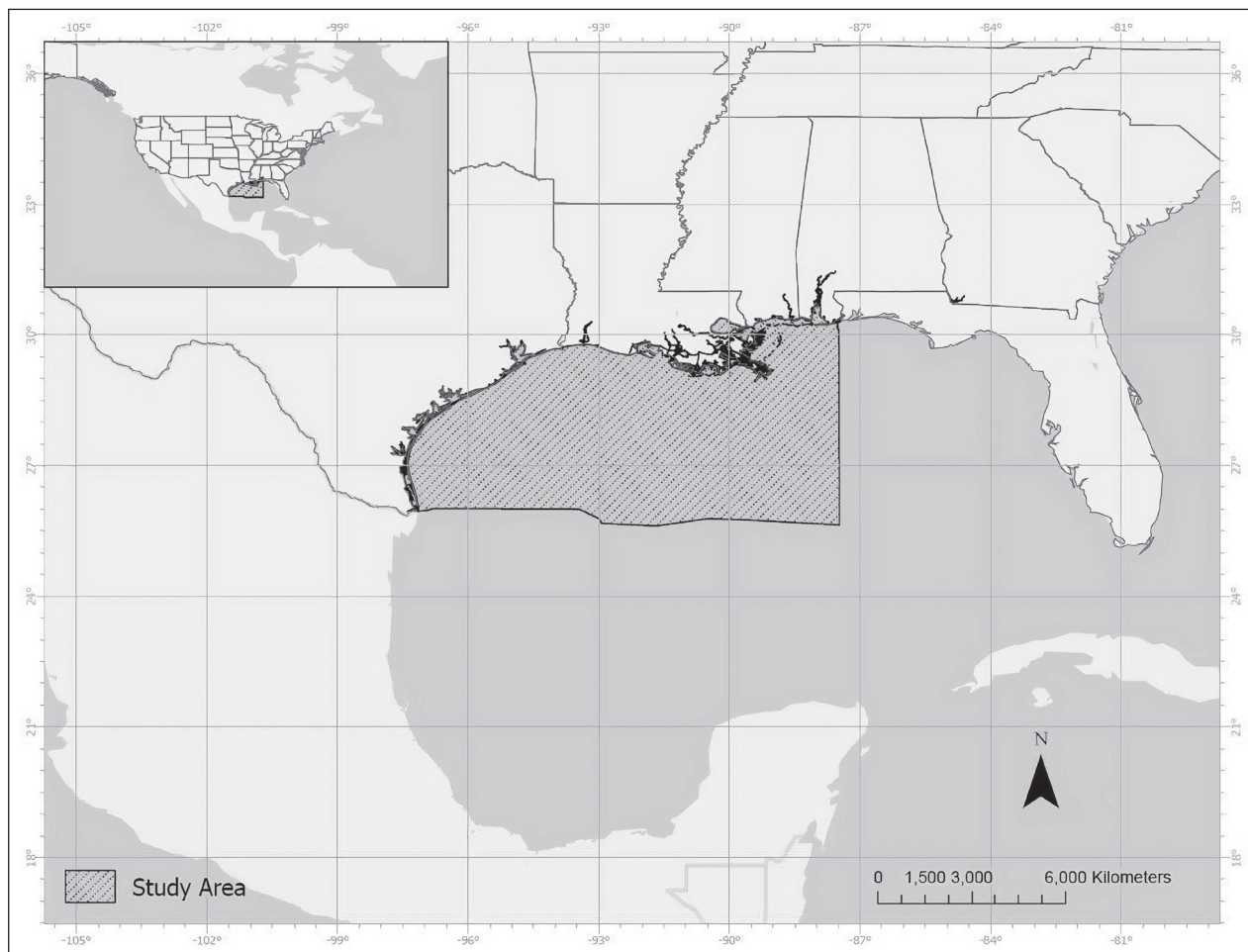


Figure 1. The risk assessment area: the northern Gulf of Mexico, which for the purpose of the present study included coastal Alabama, Mississippi, Louisiana, and Texas. Map lines delineate study areas and do not necessarily depict accepted national boundaries.

already been conducted for the state of Florida (Lieurance et al. in press; Kendig et al. 2022).

The northern Gulf of Mexico region is a heavily perturbed system with a history of intense hurricanes, substantial flood events, and intensive industrial resource use. The northern Gulf of Mexico is one of the major petroleum-producing areas in the U.S. (> 1,800 oil and gas platforms) and represents one of the largest fisheries by volume of commercial landings in the U.S. (U.S. Department of Commerce 2019). Owing to the presence of large commercial fishing operations and crude oil import and export terminals, several of the U.S.'s major international shipping ports are in the northern Gulf of Mexico, including the Port of Mobile in Alabama, Port of Pascagoula in Mississippi, Port of New Orleans in Louisiana, and Ports of Corpus Christi and Houston in Texas. A combined freight tonnage of 527,536,964 passed through these ports in 2019 (Transport Topics 2020: www.ttnews.com/). The Port of Houston is the United States' number one port for imports and exports, with 69% of all U.S. Gulf Coast's container traffic passing through this port (Morton 2020). With human-induced activities linked to declines in biodiversity (Teofilova et al. 2012; Zeng et al. 2021) and increases in invasive species establishment and spread (Johnston et al. 2017), systems with impaired natural habitats such as the northern Gulf of Mexico may be particularly vulnerable to invasion by new species (Stachowicz et al. 2002). Moreover, infrastructure and operations to support human activities in the region (e.g. international shipping) provide pathways of

introduction and spread, with an increase in frequency of these pathways equating to an increase in species propagule pressure and, subsequently, establishment of new species (Lockwood et al. 2009; Tidbury et al. 2016, 2021).

Horizon scanning

A multi-step process was used to identify potential future (i.e. horizon) non-native species that have a high likelihood of arriving and establishing in the northern Gulf of Mexico. First, an initial list of marine species (i.e. species that spend at least part of their lives in fully marine conditions) with physiological tolerances that matched the conditions of the northern Gulf of Mexico was developed using several resources, which included the Centre for Agriculture and Bioscience International Invasive Species Compendium Horizon Scanning tool (www.cabi.org/HorizonScanningTool), a list of species imported into the continental United States from 2015 to 2019 (Law Enforcement Management Information System: U.S. Department of the Interior 2020), and invasive species databases such as the Ocean Biodiversity Information System (www.obis.org), the National Exotic Marine and Estuarine Species Information System (invasions.si.edu/nemesisnew/index.html), and FishBase (www.fishbase.org) or SeaLifeBase (www.sealifebase.org). Species on this list were then cross-checked with species in the USGS Non-indigenous Aquatic Species database (nas.er.usgs.gov) and Gulfbase (www.gulfbase.org) to determine whether they were already reported as established in the risk assessment area; if it was determined that they were already established, then they were removed from the list. AquaMaps (www.aquamaps.org; Kaschner et al. 2019), which is an online tool that produces computer-generated predicted global distribution maps for marine species, was then consulted to determine habitat suitability for each species. AquaMaps uses several criteria such as water depth, temperature, salinity, and primary production to determine habitat suitability. If the risk assessment area was determined to be unsuitable habitat for a species, then that species was removed from the list.

The resulting list included 1,303 non-native species (see Suppl. material 1) for pre-screening review with respect to their suitability for risk screening (i.e. those whose native habitat is similar to that of the northern Gulf of Mexico and whose physiological tolerances could allow establishment in the northern Gulf of Mexico if introduced). The pre-screening review involved categorization of the 1,303 species by major taxonomic group, with species distributed amongst eleven assessors, each specializing in one or more of the taxonomic groups. Using expert judgment, the assessors reviewed the likelihood of these species being introduced, becoming established, and causing ecological and/or socio-economic harm within the northern Gulf of Mexico over the next decade to help sub-select species for a full risk screening. To facilitate this pre-screening process and to ensure consistent review across taxonomic groups, assessors were provided with a short series of questions that helped determine the potential invasiveness of a species in a short amount of time and were based on attributes developed by Roy et al. (2014) (see Suppl. material 2). Species determined to be the most likely to be invasive from reviews were retained for formal risk screening of their potential invasiveness in the northern Gulf of Mexico.

Risk screening

From the initial list of 1,303 non-native species, 138 species were selected in the pre-screening step for risk screening: 32 bony fish (out of 232; 13.8%) (hereafter referred to as 'fishes'), zero elasmobranchs (253; 0%), six tunicates (20; 30%),

47 bivalves (61; 77%), one bryozoan (17; 5.9%), 19 cnidarians (486; 3.9%), 18 decapods (129; 14.0%), three echinoderms (32; 9.4%), eleven gastropods (72; 15.3%), and one stomatopod (1; 100%). These 138 species were screened by 13 assessors in total. Specifically, 102 species were screened by a single assessor, with eight assessors screening subsets of those species; and 36 species screened by two joint assessors (resulting in a consensus assessment), with three pairs of assessors screening subsets of those species. Upon completion of screening, each species' risk screening was subject to an independent review process in which responses, justification for responses, and scores were reviewed by an assessor other than the initial one ($n = 10$ reviewers), with reviews being recorded on a standardized review spreadsheet. Where there was disagreement in responses, the initial assessor and reviewer were consulted, and a consensus screening was achieved.

Risk screening was undertaken using the Aquatic Species Invasiveness Screening Kit (AS-ISK) v2.3.3 (Copp et al. 2016, 2021; www.cefas.co.uk/nns/tools/). This taxon-generic, decision-support tool, which complies with the 'minimum standards' for assessing non-native species (Roy et al. 2018), has also been accepted by policy (Florida Fish and Wildlife Conservation Commission and Florida Department of Agriculture and Consumer Services) for the screening of marine species, amphibians, and aquatic reptiles for the state of Florida (J.E. Hill, personal communication). The toolkit consists of 55 questions of which 49 comprise the Basic Risk Assessment (BRA) and six the Climate Change Assessment (CCA).

To achieve a valid screening, a standard protocol was followed: this is described in full in Vilizzi et al. (2022a). In brief, the assessor must provide for each question a response, confidence level, and justification (e.g. Copp et al. 2009; Vilizzi and Piria 2022), with two score outcomes (BRA and BRA+CCA). Scores < 1 suggest a 'low risk' of the species being or becoming invasive in the risk assessment area, whereas scores ≥ 1 indicate a 'medium risk' or a 'high risk'. The distinction between medium and high risk is defined using a calibrated threshold (Thr) that is obtained, whenever possible (see below), by Receiver Operating Characteristic (ROC) curve analysis (Vilizzi et al. 2022a, b). The discriminatory power of ROC analysis as defined by the Area Under the Curve (AUC) is measured as: $0.7 \leq \text{AUC} < 0.8 =$ acceptable, $0.8 \leq \text{AUC} < 0.9 =$ excellent, $0.9 \leq \text{AUC} =$ outstanding (Hosmer et al. 2013). In the present study, for the fishes and invertebrates classified as high risk, an additional 'very high risk' category was also distinguished using an *ad hoc* threshold such as first applied to the screening scores from aquatic species by Britton et al. (2011). Identification of very high-risk species may help prioritize allocation of resources for comprehensive risk assessment (Copp et al. 2005a, 2016; Vilizzi et al. 2022a).

The *a priori* categorization of species required for ROC curve analysis was implemented as per the standard protocol (Vilizzi et al. 2022a) (Table 1). This requires at least 15–20 species to achieve a successful, statistically robust calibrated threshold score with which to distinguish between medium-risk and high-risk species; these species must consist of both *a priori* non-invasive and invasive in a 'relatively balanced' proportion (Vilizzi et al. 2019, 2021). Unlike invertebrates, these requirements were not met for tunicates and fishes in the present study, so 'generalized' group-based thresholds from a global AS-ISK application (Vilizzi et al. 2021) were used for fishes (Thr = 12.75) and tunicates (Thr = 22.5). Implementation of ROC curve analysis followed the standard protocol (Vilizzi et al. 2022a), with true/false positive/negative outcome distinction not applied to medium-risk species because their further evaluation in a comprehensive risk assessment depends on policy/management priorities and/or the availability of financial resources. Fitting of the ROC curve for invertebrates was with pROC (Robin et al. 2011) for R x64 v4.0.5

Table 1. Taxa evaluated for their potential risk of invasiveness in the northern Gulf of Mexico. For each taxon, the *a priori* categorization into Invasive and Non-invasive is provided based on a the protocol described in Vilizzi et al. (2022a): (i) Database: FishBase (www.fishbase.org) for fishes; SeaLifeBase (www.sealifebase.org) for tunicates and invertebrates; (ii) Centre for Agriculture and Bioscience International Invasive Species Compendium (CABI: www.cabi.org/ISC) and Global Invasive Species Database (GISD: www.iucngisd.org); (iii) Invasive and Exotic Species of North America list (IESNA: www.invasive.org); (iv) Google Scholar literature search. N = no impact/threat; Y = impact/threat; ‘-’ = absent; n.e. = not evaluated (but present in database); n.a. = not applicable.

Taxon name	Common name	<i>A priori</i> categorization					Category
		D’base	CABI	GISD	IESNA	Scholar	
Fishes							
<i>Conger verreauxi</i>	southern conger eel	N	-	-	-	N	Non-invasive
<i>Dendrochirus barberi</i>	Hawaiian lionfish	-	-	-	-	N	Non-invasive
<i>Dendrochirus biocellatus</i>	twospot turkeyfish	-	-	-	-	N	Non-invasive
<i>Dendrochirus brachypterus</i>	dwarf lionfish	-	-	-	-	N	Non-invasive
<i>Dendrochirus zebra</i>	zebra turkeyfish	-	-	-	-	N	Non-invasive
<i>Fundulus heteroclitus heteroclitus</i>	mummichog	N	-	-	-	N	Non-invasive
<i>Heniochus diphreutes</i>	false moorish idol	N	-	n.e.	-	N	Non-invasive
<i>Lythrypnus dalli</i>	bluebanded goby	N	-	-	-	N	Non-invasive
<i>Muraenesox cinereus</i>	daggertooth pike conger	N	-	-	-	N	Non-invasive
<i>Parapterois heterura</i>	blackfoot firefish	-	-	-	-	N	Non-invasive
<i>Platax orbicularis</i>	orbicular batfish	N	-	-	N	N	Non-invasive
<i>Pterois antennata</i>	broadbarred firefish	-	-	-	-	N	Non-invasive
<i>Pterois cincta</i>	Red Sea lionfish	N	-	-	-	N	Non-invasive
<i>Pterois lunulata</i>	luna lionfish	-	-	-	-	N	Non-invasive
<i>Pterois mombasae</i>	frillkin turkeyfish	N	-	-	-	N	Non-invasive
<i>Pterois radiata</i>	radial firefish	-	-	-	-	N	Non-invasive
<i>Pterois russelii</i>	plaintail turkeyfish	-	-	-	-	N	Non-invasive
<i>Pterois sphex</i>	Hawaiian turkeyfish	-	-	-	-	N	Non-invasive
<i>Rhinopias eschmeyeri</i>	Eschmeyer’s scorpionfish	N	-	-	-	N	Non-invasive
<i>Rhinopias frondosa</i>	weedy scorpionfish	-	-	-	-	N	Non-invasive
<i>Scorpaena guttata</i>	California scorpionfish	-	-	-	-	N	Non-invasive
<i>Scorpaena mystes</i>	Pacific spotted scorpionfish	-	-	-	-	N	Non-invasive
<i>Scorpaena scrofa</i>	red scorpionfish	-	-	-	-	N	Non-invasive
<i>Scorpaenodes parvipinnis</i>	lowfin scorpionfish	-	-	-	-	N	Non-invasive
<i>Scorpaenopsis macrochir</i>	flasher scorpionfish	-	-	-	-	N	Non-invasive
<i>Scorpaenopsis neglecta</i>	yellowfin scorpionfish	-	-	-	-	N	Non-invasive
<i>Scorpaenopsis oxycephalus</i>	tassled scorpionfish	-	-	-	-	N	Non-invasive
<i>Scorpaenopsis papuensis</i>	Papuan scorpionfish	N	-	-	-	N	Non-invasive
<i>Scorpaenopsis vittapinna</i>	bandfin scorpionfish	N	-	-	-	N	Non-invasive
<i>Sebastapistes cyanostigma</i>	yellowspotted scorpionfish	-	-	-	-	N	Non-invasive
<i>Sebastapistes strongia</i>	brownbanded stingfish	-	-	-	-	N	Non-invasive
<i>Semicossyphus pulcher</i>	California sheephead	-	-	-	-	N	Non-invasive
Tunicates							
<i>Asterocarpa humilis</i>	compass sea squirt	-	-	-	-	N	Non-invasive
<i>Botrylloides violaceus</i>	purple colonial tunicate	N	-	Y	-	n.a.	Invasive
<i>Clavelina lepadiformis</i>	light-bulb ascidian	-	-	N	-	N	Non-invasive
<i>Corella eumyota</i>	orange-tipped sea squirt	-	-	Y	-	n.a.	Invasive
<i>Didemnum vexillum</i>	carpet sea squirt	-	-	Y	Y	n.a.	Invasive

Taxon name	Common name	<i>A priori</i> categorization					Category
		D'base	CABI	GISD	IESNA	Scholar	
<i>Trididemnum solidum</i>	overgrowing mat tunicate	–	Y	–	–	n.a.	Invasive
Invertebrates							
<i>Acanthaster planci</i>	crown-of-thorns	–	Y	Y	–	n.a.	Invasive
<i>Acropora abrolhosensis</i>	–	–	–	–	–	N	Non-invasive
<i>Acropora acuminata</i>	–	–	–	–	–	N	Non-invasive
<i>Acropora grandis</i>	–	–	–	–	–	N	Non-invasive
<i>Acropora longicyathus</i>	–	–	–	–	–	N	Non-invasive
<i>Acropora robusta</i>	–	–	–	–	–	N	Non-invasive
<i>Anadara inaequalis</i>	inequivalve ark	–	–	n.e.	–	N	Non-invasive
<i>Anadara kagoshimensis</i>	–	–	–	–	–	Y	Invasive
<i>Anadara satowi</i>	Chinese blood clam	–	–	n.e.	–	N	Non-invasive
<i>Arcuatula senhousia</i>	Asian date mussel	–	–	Y	–	n.a.	Invasive
<i>Argopecten noronhensis</i>	–	–	–	–	–	N	Non-invasive
<i>Argopecten nucleus</i>	nucleus scallop	–	–	–	–	N	Non-invasive
<i>Argopecten ventricosus</i>	Pacific calico scallop	–	–	–	–	N	Non-invasive
<i>Atrina pectinata</i>	comb pen shell	–	–	–	–	N	Non-invasive
<i>Bankia destructa</i>	–	–	–	–	–	N	Non-invasive
<i>Bankia zeteki</i>	–	–	–	–	–	N	Non-invasive
<i>Batillaria attramentaria</i>	Japanese false cerith	–	Y	–	–	n.a.	Invasive
<i>Batissa violacea</i>	–	–	–	–	–	N	Non-invasive
<i>Brachidontes pharaonis</i>	–	–	–	–	–	Y	Invasive
<i>Calappa hepatica</i>	reef box crab	–	–	–	–	N	Non-invasive
<i>Calcinus laevimanus</i>	Hawaiian hermit	–	–	–	–	N	Non-invasive
<i>Camposcia retusa</i>	decorator crab	–	–	–	–	N	Non-invasive
<i>Celleporaria brunnea</i>	–	–	–	–	–	N	Non-invasive
<i>Cerithium columna</i>	–	–	–	–	–	N	Non-invasive
<i>Chama asperella</i>	jewel boxes	–	–	n.e.	–	N	Non-invasive
<i>Charybdis (Charybdis) hellerii</i>	spiny hands	–	Y	N	–	n.a.	Invasive
<i>Chyromorus bifasciata</i>	morus cerith	–	–	–	–	N	Non-invasive
<i>Crassostrea brasiliiana</i>	mangrove oyster	–	–	–	–	N	Non-invasive
<i>Crassostrea columbiensis</i>	Columbia black oyster	–	–	–	–	N	Non-invasive
<i>Crassostrea tulipa</i>	West African mangrove oyster	–	–	–	–	N	Non-invasive
<i>Crepidula onyx</i>	onyx slippersnail	–	–	n.e.	–	Y	Invasive
<i>Dardanus pedunculatus</i>	anemone hermit crab	–	–	–	–	N	Non-invasive
<i>Dendostrea sandwichensis</i>	Hawaiian oyster	–	–	–	–	N	Non-invasive
<i>Dipsastraea pallida</i>	–	–	–	–	–	N	Non-invasive
<i>Enoplometopus holthuisi</i>	bullseye reef lobster	–	–	–	–	N	Non-invasive
<i>Ensis lei</i>	–	–	–	–	–	N	Non-invasive
<i>Favites complanata</i>	larger star coral	–	–	–	–	N	Non-invasive
<i>Fragum fragum</i>	white strawberry cockle	–	–	–	–	N	Non-invasive
<i>Fulvia fragilis</i>	fragile cockle	–	–	n.e.	–	N	Non-invasive
<i>Gonodactylaceus falcatus</i>	Philippine mantis shrimp	N	–	–	–	Y	Invasive
<i>Hemigrapsus takanoi</i>	brush-clawed shore crab	–	–	Y	–	n.a.	Invasive
<i>Hiatula rosea</i>	–	N	–	–	–	N	Non-invasive
<i>Hippopus hippopus</i>	bear paw clam	N	–	–	–	N	Non-invasive

Taxon name	Common name	<i>A priori</i> categorization					Category
		D'base	CABI	GISD	IESNA	Scholar	
<i>Homophyllia australis</i>	–	–	–	–	–	N	Non-invasive
<i>Hyotissa hyotis</i>	honeycomb oyster	N	–	–	–	N	Non-invasive
<i>Ilyanassa obsoleta</i>	eastern mudsnail	–	Y	–	–	N	Invasive
<i>Indothais lacera</i>	carinate rock shell	N	–	–	–	N	Non-invasive
<i>Isopora palifera</i>	catch bowl coral	–	–	–	–	N	Non-invasive
<i>Laternula gracilis</i>	–	–	–	–	–	N	Non-invasive
<i>Limaria hians</i>	gaping file shell	–	–	–	–	N	Non-invasive
<i>Lopha cristagalli</i>	coxcomb oyster	–	–	n.e.	–	N	Non-invasive
<i>Lyrodus medilobatus</i>	–	–	–	–	–	N	Non-invasive
<i>Lysmata vittata</i>	Indian lined shrimp	–	–	–	–	Y	Invasive
<i>Madracis formosa</i>	eight-ray finger coral	–	–	–	–	N	Non-invasive
<i>Magallana gigas</i>	Pacific oyster	–	–	Y	–	n.a.	Invasive
<i>Montipora foliosa</i>	cabbage coral	–	–	–	–	N	Non-invasive
<i>Mytella strigata</i>	Guyana swamp mussel	N	–	–	–	Y	Invasive
<i>Mytilopsis adamsi</i>	–	–	–	–	–	N	Non-invasive
<i>Mytilopsis salei</i>	Santo Domingo false mussel	–	Y	Y	–	n.a.	Invasive
<i>Mytilus californianus</i>	California mussel	–	–	–	–	N	Non-invasive
<i>Mytilus edulis</i>	blue mussel	–	–	–	–	N	Non-invasive
<i>Nitidotellina valtonis</i>	–	N	–	–	–	N	Non-invasive
<i>Ophiothela mirabilis</i>	–	–	–	–	–	Y	Invasive
<i>Palaemon carinicauda</i>	oriental prawn	–	–	–	–	N	Non-invasive
<i>Panulirus regius</i>	royal spiny lobster	–	–	–	–	N	Non-invasive
<i>Panulirus versicolor</i>	painted spiny lobster	–	–	–	–	N	Non-invasive
<i>Paragoniastrea australensis</i>	lesser star coral	–	–	–	–	N	Non-invasive
<i>Pavona cactus</i>	–	–	–	–	–	N	Non-invasive
<i>Penaeus japonicus</i>	kuruma prawn	–	–	N	–	N	Non-invasive
<i>Penaeus stylirostris</i>	blue shrimp	–	–	–	–	N	Non-invasive
<i>Perna viridis</i>	Asian green mussel	–	Y	Y	Y	n.a.	Invasive
<i>Petrolisthes lamarckii</i>	–	–	–	–	–	N	Non-invasive
<i>Pinctada maxima</i>	silverlip pearl oyster	–	–	–	–	N	Non-invasive
<i>Plicatula plicata</i>	plicate kitten's paw	–	–	n.e.	–	N	Non-invasive
<i>Pocillopora damicornis</i>	cauliflower coral	–	–	–	–	N	Non-invasive
<i>Polinices albumen</i>	–	–	–	–	–	N	Non-invasive
<i>Porites cylindrica</i>	yellow finger coral	–	–	–	–	N	Non-invasive
<i>Portunus segnis</i>	–	–	–	N	–	Y	Invasive
<i>Portunus trituberculatus</i>	Gazami crab	N	–	–	–	N	Non-invasive
<i>Potamocorbula amurensis</i>	brackish-water corbula	–	Y	Y	–	n.a.	Invasive
<i>Pteria hirundo</i>	European wing oyster	–	–	n.e.	–	N	Non-invasive
<i>Rapana venosa</i>	purple whelk	N	Y	Y	Y	n.a.	Invasive
<i>Rhinoclavis kochi</i>	Koch's cerith	–	–	n.e.	–	N	Non-invasive
<i>Ruditapes philippinarum</i>	Manila clam	–	–	Y	–	n.a.	Invasive
<i>Saccostrea cucullata</i>	hooded oyster	–	–	–	–	N	Non-invasive
<i>Schizophrys aspera</i>	common decorator crab	–	–	n.e.	–	N	Non-invasive
<i>Septifer cumingii</i>	–	N	–	–	–	N	Non-invasive
<i>Seriatopora hystrix</i>	thin birdsnest coral	–	–	–	–	N	Non-invasive
<i>Spondylus spinosus</i>	spiny oyster	–	–	–	–	N	Non-invasive

Taxon name	Common name	<i>A priori</i> categorization					Category
		D'base	CABI	GISD	IESNA	Scholar	
<i>Stichodactyla gigantea</i>	gigantic sea anemone	–	–	–	–	N	Non-invasive
<i>Stiliger fuscovittatus</i>	brown-streak stiliger	–	–	–	–	N	Non-invasive
<i>Stylophora pistillata</i>	smooth cauliflower coral	–	–	–	–	N	Non-invasive
<i>Synalpheus africanus</i>	–	–	–	–	–	N	Non-invasive
<i>Tegillarca granosa</i>	granular ark	N	–	–	–	N	Non-invasive
<i>Theora lata</i>	–	–	–	–	–	N	Non-invasive
<i>Theora lubrica</i>	Asian semele	–	–	–	–	Y	Invasive
<i>Timoclea marica</i>	–	N	–	–	–	N	Non-invasive
<i>Toxopneustes pileolus</i>	flower urchin	N	–	–	–	N	Non-invasive
<i>Tubastraea tagusensis</i>	–	–	–	–	–	N	Non-invasive
<i>Urosalpinx cinerea</i>	Atlantic oyster drill	–	Y	Y	–	N	Invasive

(R Core Team 2023). Permutational ANOVA with normalization of the data was used to test for differences in confidence factor (CF: see Vilizzi et al. 2022a) between components (i.e., BRA and BRA+CCA); this used a Bray-Curtis dissimilarity measure, 9999 unrestricted permutations of the raw data, and statistical effects evaluated at $\alpha = 0.05$.

Results

A risk screening report was generated for each of the 138 species within this study (see Suppl. material 3). Regarding the confidence factor (CF), the mean CF_{Total} was 0.594 ± 0.006 , the mean CF_{BRA} 0.600 ± 0.006 , and the mean CF_{CCA} 0.544 ± 0.010 (hence, in all cases indicating medium confidence). The mean CF_{BRA} was higher than the mean CF_{CCA} ($F^{\#}_{1,274} = 23.83$, $P^{\#} < 0.001$; # = permutational value). Owing to differences in score minima and maxima of the BRA and BRA+CCA, the score intervals used to rank species, based on taxon-specific thresholds, were as follows: BRA: Low [–20, 1[; Medium [1, Threshold[; High [Threshold, 70]; BRA+CCA: Low [–32, 1[; Medium [1, Threshold[; High [Threshold, 82]. Note that threshold intervals are presented using the appropriate statistical use of interval brackets “]” and “[”, with the reverse bracket notation indicating an open interval.

Fishes

All 32 fishes screened were categorized *a priori* as non-invasive (Table 1). Based on the BRA outcome scores (Table 2, Fig. 2A), 13 species (40.6%) were ranked as high risk and 19 (59.4%) as medium risk. Of these species, 13 were false positives. Based on the BRA+CCA outcome scores (Table 2, Fig. 2B), 14 species (43.8%) were ranked as high or very high risk and 18 (56.3%) as medium risk. Of these species, 14 were false positives. All high-risk species for the BRA were also classified as high risk after accounting for climate change predictions (cf. BRA+CCA), which resulted in the additional inclusion of the lowfin scorpionfish *Scorpaenodes parvipinnis* (Garrett, 1864) (medium risk for the BRA).

Based on an *ad hoc* very high-risk threshold ≥ 20 , the highest-scoring species were California scorpionfish *Scorpaena guttata* Girard, 1854 and red scorpionfish *Scorpaena scrofa* Linnaeus, 1758 for both the BRA and BRA+CCA, and daggertooth pike conger *Muraenesox cinereus* (Forsskal, 1775), weedy scorpionfish *Rhinopias frondosa* (Günther, 1892), yellowfin scorpionfish *Scorpaenopsis neglecta* Heckel, 1837, tassled scorpionfish

Table 2. Risk ranks for the taxa evaluated with the Aquatic Species Invasiveness Screening Kit for the northern Gulf of Mexico. For each taxon, the following information is provided: *a priori* categorization for invasiveness (N = non-invasive; Y = invasive; see Table 1), BRA (Basic Risk Assessment) and BRA+CCA (BRA + Climate Change Assessment) scores with corresponding risk ranks (L = Low; M = Medium; H = High; VH = Very high, based on an *ad hoc* threshold ≥ 20 for fishes and ≥ 40 for invertebrates; see text for details), classification (Class: FN = false negative; FP = false positive; TN = true negative; TP = true positive; ‘-’ = not applicable as medium-risk; see text for details), and difference (Delta) between BRA+CCA and BRA risk scores. CF = confidence factor, which is based on confidence levels (see text for details). Risk ranks are based on the following thresholds (Thr) for distinguishing between H-risk species and species of L or M risk: 12.75 for fishes, 22.5 for tunicates, 22.25 for invertebrates. In all cases, the threshold between L and M is ‘1’. Note also the differences in score minima and maxima for BRA (-20, 70) and BRA+CCA (-32, 82) due to differences in the total number of questions, i.e. 49 and 55, respectively.

Taxon name	<i>A priori</i>	BRA			BRA+CCA			Delta	CF		
		Score	Rank	Class	Score	Rank	Class		Total	BRA	CCA
Fishes											
<i>Conger verreauxi</i>	N	11.0	M	-	11.0	M	-	0	0.53	0.53	0.50
<i>Dendrochirus barberi</i>	N	6.0	M	-	6.0	M	-	0	0.55	0.56	0.50
<i>Dendrochirus biocellatus</i>	N	3.0	M	-	3.0	M	-	0	0.57	0.58	0.50
<i>Dendrochirus brachypterus</i>	N	12.0	M	-	12.0	M	-	0	0.61	0.63	0.50
<i>Dendrochirus zebra</i>	N	10.0	M	-	10.0	M	-	0	0.61	0.62	0.50
<i>Fundulus heteroclitus heteroclitus</i>	N	14.0	H	FP	14.0	H	FP	0	0.69	0.71	0.50
<i>Heniochus diphreutes</i>	N	7.0	M	-	9.0	M	-	2	0.70	0.71	0.54
<i>Lythrypnus dalli</i>	N	4.0	M	-	4.0	M	-	0	0.59	0.59	0.58
<i>Muraenesox cinereus</i>	N	15.0	H	FP	23.0	VH	FP	8	0.64	0.67	0.42
<i>Parapterois heterura</i>	N	4.0	M	-	4.0	M	-	0	0.47	0.46	0.50
<i>Platax orbicularis</i>	Y	15.0	H	TP	15.0	H	TP	0	0.65	0.68	0.33
<i>Pterois antennata</i>	N	9.0	M	-	9.0	M	-	0	0.59	0.60	0.50
<i>Pterois cincta</i>	N	5.0	M	-	5.0	M	-	0	0.55	0.56	0.50
<i>Pterois lunulata</i>	N	12.0	M	-	12.0	M	-	0	0.57	0.58	0.50
<i>Pterois mombasae</i>	N	7.0	M	-	7.0	M	-	0	0.57	0.58	0.50
<i>Pterois radiata</i>	N	5.0	M	-	5.0	M	-	0	0.60	0.61	0.50
<i>Pterois russelii</i>	N	19.0	H	FP	19.0	H	FP	0	0.56	0.57	0.50
<i>Pterois sphex</i>	N	5.0	M	-	5.0	M	-	0	0.60	0.61	0.50
<i>Rhinopias eschmeyeri</i>	N	16.0	H	FP	18.0	H	FP	2	0.53	0.54	0.50
<i>Rhinopias frondosa</i>	N	18.0	H	FP	26.0	VH	FP	8	0.60	0.61	0.54
<i>Scorpaena guttata</i>	N	22.0	VH	FP	30.0	VH	FP	8	0.70	0.72	0.50
<i>Scorpaena mystes</i>	N	13.0	H	FP	15.0	H	FP	2	0.50	0.51	0.46
<i>Scorpaena scrofa</i>	N	20.0	VH	FP	22.0	VH	FP	2	0.50	0.51	0.46
<i>Scorpaenodes parvipinnis</i>	N	9.0	M	-	13.0	H	FP	4	0.45	0.46	0.38
<i>Scorpaenopsis macrochir</i>	N	14.0	H	FP	16.0	H	FP	2	0.45	0.45	0.42
<i>Scorpaenopsis neglecta</i>	N	19.0	H	FP	21.0	VH	FP	2	0.46	0.46	0.42
<i>Scorpaenopsis oxycephalus</i>	N	19.0	H	FP	21.0	VH	FP	2	0.44	0.44	0.42
<i>Scorpaenopsis papuensis</i>	N	18.0	H	FP	22.0	VH	FP	4	0.45	0.44	0.50
<i>Scorpaenopsis vittapinna</i>	N	11.0	M	-	11.0	M	-	0	0.44	0.41	0.63
<i>Sebastapistes cyanostigma</i>	N	10.0	M	-	10.0	M	-	0	0.44	0.42	0.63
<i>Sebastapistes strongia</i>	N	11.0	M	-	11.0	M	-	0	0.46	0.44	0.63
<i>Semicossyphus pulcher</i>	N	10.0	M	-	12.0	M	-	2	0.56	0.60	0.25
Tunicates											
<i>Asterocarpa humilis</i>	N	11.5	M	-	13.5	M	-	2	0.64	0.65	0.54
<i>Botrylloides violaceus</i>	Y	34.0	H	TP	38.0	H	TP	4	0.59	0.60	0.50
<i>Clavelina lepadiformis</i>	N	14.0	M	-	14.0	M	-	0	0.60	0.62	0.46

Taxon name	A priori	BRA			BRA+CCA			Delta	CF		
		Score	Rank	Class	Score	Rank	Class		Total	BRA	CCA
<i>Corella eumyota</i>	Y	16.5	M	–	12.5	M	–	–4	0.59	0.60	0.50
<i>Didemnum vexillum</i>	Y	35.0	H	TP	41.0	H	TP	6	0.59	0.62	0.33
<i>Trididemnum solidum</i>	Y	23.0	H	TP	33.0	H	TP	10	0.55	0.57	0.38
Invertebrates											
<i>Acanthaster planci</i>	Y	9.0	M	–	11.0	M	–	2	0.69	0.69	0.63
<i>Acropora abrolhosensis</i>	N	9.0	M	–	11.0	M	–	2	0.55	0.56	0.54
<i>Acropora acuminata</i>	N	12.0	M	–	12.0	M	–	0	0.58	0.58	0.58
<i>Acropora grandis</i>	N	12.0	M	–	12.0	M	–	0	0.58	0.58	0.58
<i>Acropora longicyathus</i>	N	13.0	M	–	13.0	M	–	0	0.58	0.58	0.58
<i>Acropora robusta</i>	N	12.0	M	–	12.0	M	–	0	0.58	0.58	0.58
<i>Anadara inaequivalvis</i>	N	17.0	M	–	17.0	M	–	0	0.68	0.67	0.75
<i>Anadara kagoshimensis</i>	Y	11.0	M	–	–1.0	L	FN	–12	0.63	0.62	0.75
<i>Anadara satowi</i>	N	4.0	M	–	–2.0	L	TN	–6	0.65	0.64	0.75
<i>Arcuatula senhousia</i>	Y	22.5	H	TP	10.5	M	–	–12	0.54	0.52	0.75
<i>Argopecten noronhensis</i>	N	5.0	M	–	17.0	M	–	12	0.59	0.57	0.75
<i>Argopecten nucleus</i>	N	5.0	M	–	17.0	M	–	12	0.60	0.58	0.75
<i>Argopecten ventricosus</i>	N	9.0	M	–	21.0	M	–	12	0.57	0.58	0.50
<i>Atrina pectinata</i>	N	10.0	M	–	22.0	M	–	12	0.63	0.64	0.50
<i>Bankia destructa</i>	N	25.0	H	FP	35.0	H	FP	10	0.64	0.69	0.25
<i>Bankia zeteki</i>	N	24.0	H	FP	12.0	M	–	–12	0.56	0.60	0.25
<i>Batillaria atramentaria</i>	Y	28.0	H	TP	34.0	H	TP	6	0.62	0.63	0.50
<i>Batissa violacea</i>	N	9.0	M	–	21.0	M	–	12	0.41	0.43	0.25
<i>Brachidontes pharaonis</i>	Y	24.0	H	TP	36.0	H	TP	12	0.62	0.60	0.75
<i>Calappa hepatica</i>	N	13.0	M	–	21.0	M	–	8	0.56	0.57	0.46
<i>Calcinus laevimanus</i>	N	10.0	M	–	14.0	M	–	4	0.66	0.65	0.71
<i>Camposcia retusa</i>	N	12.0	M	–	18.0	M	–	6	0.54	0.55	0.46
<i>Celleporaria brunnea</i>	N	6.0	M	–	8.0	M	–	2	0.68	0.68	0.71
<i>Cerithium columna</i>	N	9.0	M	–	13.0	M	–	4	0.60	0.60	0.54
<i>Chama asperella</i>	N	15.0	M	–	15.0	M	–	0	0.57	0.61	0.25
<i>Charybdis (Charybdis) hellerii</i>	Y	10.0	M	–	10.0	M	–	0	0.62	0.64	0.50
<i>Clypeomorus bifasciata</i>	N	15.0	M	–	21.0	M	–	6	0.61	0.62	0.54
<i>Crassostrea brasiliiana</i>	N	26.0	H	FP	38.0	H	FP	12	0.72	0.71	0.75
<i>Crassostrea columbiensis</i>	N	29.0	H	FP	17.0	M	–	–12	0.63	0.64	0.50
<i>Crassostrea tulipa</i>	N	26.0	H	FP	38.0	H	FP	12	0.70	0.70	0.75
<i>Crepidula onyx</i>	Y	26.5	H	TP	26.5	H	TP	0	0.65	0.67	0.50
<i>Dardanus pedunculatus</i>	N	13.0	M	–	17.0	M	–	4	0.56	0.56	0.54
<i>Dendostrea sandwichensis</i>	N	16.0	M	–	28.0	H	FP	12	0.57	0.55	0.75
<i>Dipsastraea pallida</i>	N	14.0	M	–	14.0	M	–	0	0.58	0.58	0.58
<i>Enoplometopus holthuisi</i>	N	10.0	M	–	16.0	M	–	6	0.52	0.52	0.50
<i>Ensis lei</i>	N	9.5	M	–	3.5	M	–	–6	0.55	0.56	0.50
<i>Favites complanata</i>	N	12.0	M	–	12.0	M	–	0	0.58	0.58	0.58
<i>Fragum fragum</i>	N	4.0	M	–	16.0	M	–	12	0.52	0.52	0.50
<i>Fulvia fragilis</i>	N	–0.5	L	TN	–10.5	L	TN	–10	0.55	0.55	0.50
<i>Gonodactylaceus falcatus</i>	Y	28.0	H	TP	34.0	H	TP	6	0.63	0.64	0.54
<i>Hemigrapsus takanoi</i>	Y	34.0	H	TP	42.0	VH	TP	8	0.70	0.72	0.50
<i>Hiatula rosea</i>	N	–1.5	L	TN	10.5	M	–	12	0.56	0.57	0.50

Taxon name	<i>A priori</i>	BRA			BRA+CCA			Delta	CF		
		Score	Rank	Class	Score	Rank	Class		Total	BRA	CCA
<i>Hippopus hippopus</i>	N	8.0	M	–	20.0	M	–	12	0.56	0.57	0.50
<i>Homophyllia australis</i>	N	12.0	M	–	12.0	M	–	0	0.57	0.57	0.58
<i>Hytissa hyotis</i>	N	33.0	H	FP	45.0	VH	FP	12	0.59	0.60	0.50
<i>Ilyanassa obsoleta</i>	Y	37.0	H	TP	35.0	H	TP	–2	0.68	0.70	0.46
<i>Indothais lacera</i>	N	37.0	H	FP	43.0	VH	FP	6	0.57	0.58	0.50
<i>Isopora palifera</i>	N	12.0	M	–	12.0	M	–	0	0.58	0.58	0.58
<i>Laternula gracilis</i>	N	1.5	M	–	–10.5	L	TN	–12	0.55	0.56	0.50
<i>Limaria hians</i>	N	9.0	M	–	21.0	M	–	12	0.53	0.54	0.50
<i>Lopha cristagalli</i>	N	5.0	M	–	17.0	M	–	12	0.53	0.53	0.50
<i>Lyrodus medilobatus</i>	N	17.0	M	–	29.0	H	FP	12	0.66	0.65	0.75
<i>Lysmata vittata</i>	Y	25.0	H	TP	31.0	H	TP	6	0.64	0.65	0.54
<i>Madracis formosa</i>	N	10.0	M	–	10.0	M	–	0	0.58	0.58	0.58
<i>Magallana gigas</i>	Y	37.0	H	TP	25.0	H	TP	–12	0.67	0.70	0.46
<i>Montipora foliosa</i>	N	13.0	M	–	17.0	M	–	4	0.58	0.58	0.58
<i>Mytella strigata</i>	Y	27.5	H	TP	39.5	H	TP	12	0.68	0.67	0.75
<i>Mytilopsis adamsi</i>	N	18.0	M	–	30.0	H	FP	12	0.61	0.63	0.50
<i>Mytilopsis sallei</i>	Y	40.0	VH	TP	52.0	VH	TP	12	0.69	0.71	0.50
<i>Mytilus californianus</i>	N	13.0	M	–	1.0	M	–	–12	0.68	0.67	0.75
<i>Mytilus edulis</i>	N	18.5	M	–	6.5	M	–	–12	0.70	0.70	0.75
<i>Nitidotellina valtonis</i>	N	–1.0	L	TN	11.0	M	–	12	0.63	0.62	0.75
<i>Ophiothela mirabilis</i>	Y	10.5	M	–	12.5	M	–	2	0.66	0.67	0.63
<i>Palaemon carinicauda</i>	N	9.0	M	–	9.0	M	–	0	0.61	0.62	0.50
<i>Panulirus regius</i>	N	22.0	M	–	28.0	H	FP	6	0.64	0.65	0.54
<i>Panulirus versicolor</i>	N	17.0	M	–	23.0	H	FP	6	0.65	0.66	0.58
<i>Paragoniastrea australensis</i>	N	12.0	M	–	12.0	M	–	0	0.58	0.58	0.58
<i>Pavona cactus</i>	N	10.0	M	–	10.0	M	–	0	0.58	0.58	0.58
<i>Penaeus japonicus</i>	N	22.5	H	FP	26.5	H	FP	4	0.66	0.67	0.54
<i>Penaeus stylirostris</i>	N	15.5	M	–	19.5	M	–	4	0.60	0.62	0.50
<i>Perna viridis</i>	Y	37.0	H	TP	49.0	VH	TP	12	0.68	0.70	0.50
<i>Petrolisthes lamarckii</i>	N	13.0	M	–	13.0	M	–	0	0.60	0.61	0.50
<i>Pinctada maxima</i>	N	17.0	M	–	29.0	H	FP	12	0.68	0.70	0.50
<i>Plicatula plicata</i>	N	22.0	M	–	34.0	H	FP	12	0.56	0.57	0.50
<i>Pocillopora damicornis</i>	N	14.0	M	–	14.0	M	–	0	0.58	0.58	0.58
<i>Polinices albumen</i>	N	9.0	M	–	15.0	M	–	6	0.62	0.64	0.50
<i>Porites cylindrica</i>	N	14.0	M	–	14.0	M	–	0	0.58	0.58	0.58
<i>Portunus segnis</i>	Y	26.0	H	TP	32.0	H	TP	6	0.59	0.60	0.50
<i>Portunus trituberculatus</i>	N	17.0	M	–	17.0	M	–	0	0.62	0.63	0.50
<i>Potamocorbula amurensis</i>	Y	27.0	H	TP	21.0	M	–	–6	0.73	0.77	0.46
<i>Pteria hirundo</i>	N	25.0	H	FP	25.0	H	FP	0	0.63	0.64	0.50
<i>Rapana venosa</i>	Y	51.0	VH	TP	61.0	VH	TP	10	0.76	0.77	0.71
<i>Rhinoclavis kochi</i>	N	10.5	M	–	16.5	M	–	6	0.58	0.59	0.54
<i>Ruditapes philippinarum</i>	Y	17.0	M	–	29.0	H	TP	12	0.65	0.64	0.75
<i>Saccostrea cucullata</i>	N	27.0	H	FP	39.0	H	FP	12	0.63	0.61	0.75
<i>Schizophrys aspera</i>	N	11.0	M	–	13.0	M	–	2	0.57	0.58	0.50
<i>Septifer cumingii</i>	N	27.0	H	FP	39.0	H	FP	12	0.62	0.61	0.75
<i>Seriatopora hystrix</i>	N	10.0	M	–	10.0	M	–	0	0.58	0.58	0.58

Taxon name	<i>A priori</i>	BRA			BRA+CCA			Delta	CF		
		Score	Rank	Class	Score	Rank	Class		Total	BRA	CCA
<i>Spondylus spinosus</i>	N	17.0	M	–	29.0	H	FP	12	0.61	0.59	0.75
<i>Stichodactyla gigantea</i>	N	14.0	M	–	14.0	M	–	0	0.57	0.57	0.58
<i>Stiliger fuscovittatus</i>	N	7.0	M	–	9.0	M	–	2	0.61	0.62	0.50
<i>Stylophora pistillata</i>	N	14.0	M	–	14.0	M	–	0	0.58	0.58	0.63
<i>Synalpheus africanus</i>	N	10.0	M	–	18.0	M	–	8	0.62	0.63	0.50
<i>Tegillarca granosa</i>	N	21.0	M	–	33.0	H	FP	12	0.64	0.62	0.75
<i>Theora lata</i>	N	11.0	M	–	23.0	H	FP	12	0.51	0.52	0.50
<i>Theora lubrica</i>	Y	16.5	M	–	4.5	M	–	-12	0.52	0.52	0.50
<i>Timoclea marica</i>	N	5.0	M	–	17.0	M	–	12	0.55	0.56	0.50
<i>Toxopneustes pileolus</i>	N	1.0	M	–	3.0	M	–	2	0.65	0.66	0.63
<i>Tubastraea tagusensis</i>	N	26.0	H	FP	26.0	H	FP	0	0.60	0.60	0.58
<i>Urosalpinx cinerea</i>	Y	31.0	H	TP	31.0	H	TP	0	0.69	0.71	0.50

Scorpaenopsis oxycephalus (Bleeker, 1849), and Papuan scorpionfish *Scorpaenopsis papuensis* (Cuvier, 1829) for the BRA+CCA only (Fig. 2A, B). The CCA resulted in an increase in the BRA score for 13 species and in no change for the remaining 19 (Table 2).

For the high-risk species, invasiveness was mostly attributed to their ability to exploit resources and their undesirable traits, which may increase persistence, and moderately positively influenced by their ability to disperse. There was very little influence on their potential invasiveness from domestication, climate/distribution/introduction risk, invasiveness elsewhere, and reproduction, and no influence from tolerance attributes. Invasiveness was moderately influenced by climate change (Fig. 3A).

Tunicates

Of the six tunicates screened, two were categorized *a priori* as non-invasive and four as invasive (Table 1). Based on the BRA outcome scores (Table 2, Fig. 2C), three species (50.0%) were ranked as high risk (i.e. purple colonial tunicate *Botrylloides violaceus* Oka, 1927, carpet sea squirt *Didemnum vexillum* Kott, 2002, and overgrowing mat tunicate *Trididemnum solidum* (Van Name, 1902)) and three species (50.0%) as medium risk. All three high-risk species were *a priori* invasive hence true positives. Of the three medium-risk species, two were *a priori* non-invasive and one was *a priori* invasive. Based on the BRA+CCA outcome scores (Table 2, Fig. 2D), the same risk ranks were obtained as for the BRA. The CCA resulted in an increase in the BRA score for four species, no change for one, and a decrease for one (Table 2).

High-risk scores for the tunicates were mostly driven by their history of invasion elsewhere and their undesirable traits making them more persistent, and moderately by their ability to tolerate a wide range of environmental conditions (e.g. salinity, flow rates) (Fig. 3B). The highest risk scores for tunicates were generally lower than the highest invertebrate scores; this was largely due to lack of domestication and cultivation of tunicates as well as lower resource exploitation by tunicates compared to shellfishes and marine snails.

Invertebrates

Of the 100 invertebrates screened, 78 were categorized *a priori* as non-invasive and 22 as invasive (Table 1). The large sample size and proportion of *a priori* non-invasive and invasive species allowed for risk assessment area-specific calibration using

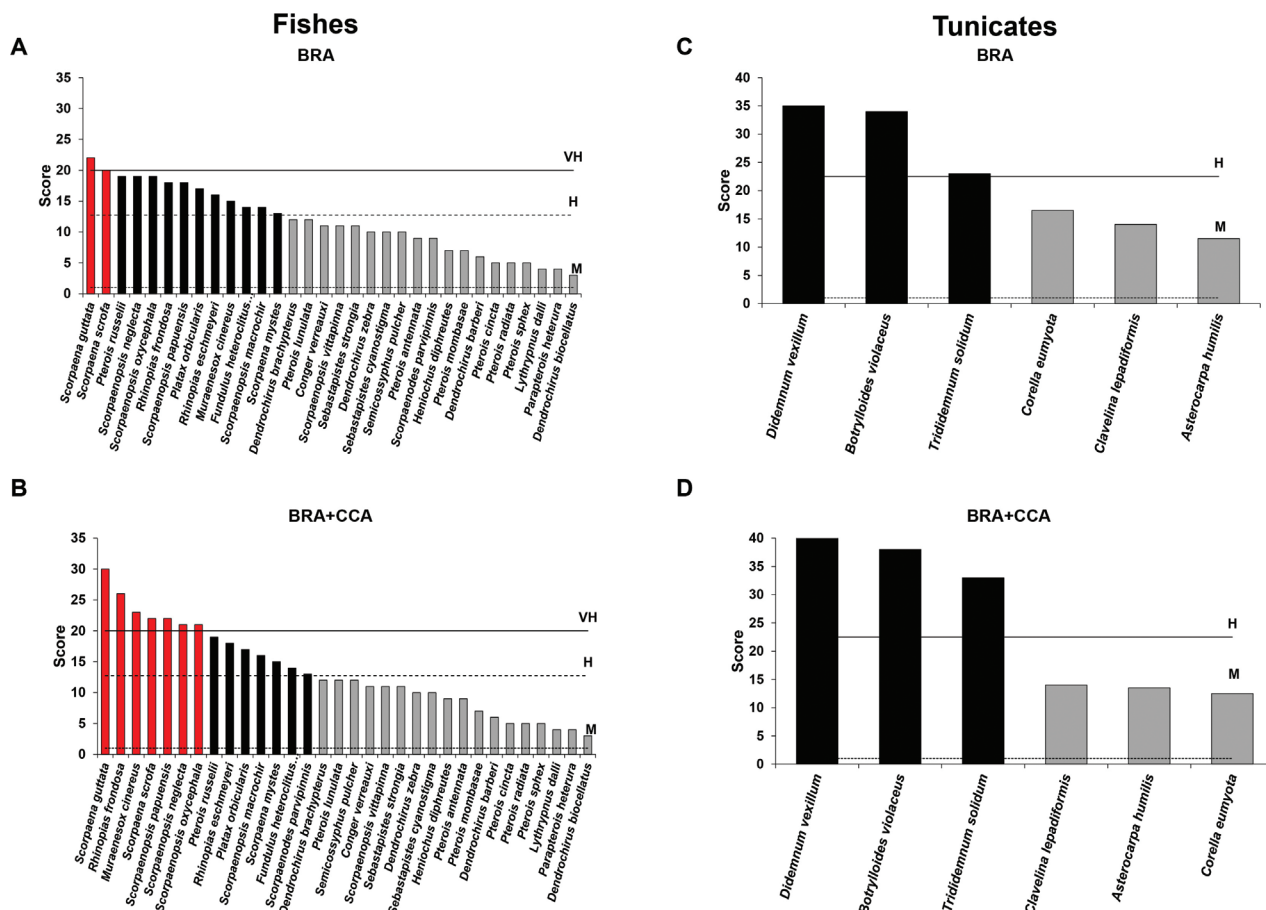


Figure 2. Aquatic Species Invasiveness Screening Kit (AS-ISK) outcome scores: (A) Basic Risk Assessment (BRA) scores for fishes; (B) BRA+CCA (Climate Change Assessment) scores for fishes; (C) BRA scores for tunicates; (D) BRA+CCA scores for tunicates. Red bars = very high-risk species; Black bars = high-risk species; Gray bars = medium-risk species. Solid line = very high-risk (VH) threshold; Hatched line = high-risk (H) threshold; Dotted line = medium-risk (M) threshold (thresholds as per Table 2).

ROC analysis to identify a threshold with which to distinguish between medium-risk and high-risk species. The ROC curve resulted in an AUC of 0.8185 (0.7074–0.9296 95% CI), hence with excellent discriminatory power, and thus providing a threshold of 22.25.

Based on the BRA outcome scores (Table 2, Fig. 4A, 5A), 28 species (28.0%) were ranked as high risk, 69 (69.0%) as medium risk, and three (3.0%) as low risk. Amongst the 78 *a priori* non-invasive species, 12 were false positives and three true negatives, and amongst the 22 *a priori* invasive species, 16 were true positives. Of the 69 medium-risk species, 63 were *a priori* non-invasive and six invasive. Based on the BRA+CCA outcome scores (Table 2, Fig. 4B, 5B), 35 species (35.0%) were ranked as high risk, 61 (61.0%) as medium risk, and four (4.0%) as low risk. Amongst the *a priori* non-invasive species, 20 were false positives and three true negatives. Amongst the *a priori* invasive species, 15 were true positives and one a false negative: *Anadara kagoshimensis* (Tokunaga, 1906). Of the 61 medium-risk species, 55 were *a priori* non-invasive and six invasive.

Based on an *ad hoc* very high-risk threshold ≥ 40 , the highest-scoring species were Santo Domingo false mussel *Mytilopsis sallei* (Récluz, 1849) and purple whelk *Rapana venosa* (Valenciennes, 1846) for both the BRA and BRA+CCA, and honeycomb oyster *Hyotissa hyotis* (Linnaeus, 1758), carinate rock shell *Indothais lacera* (Born, 1778), brush-clawed shore crab *Hemigrapsus takanoi* Asakura & Watanabe, 2005 and, Asian green mussel *Perna viridis* (Linnaeus, 1758) for the BRA+CCA

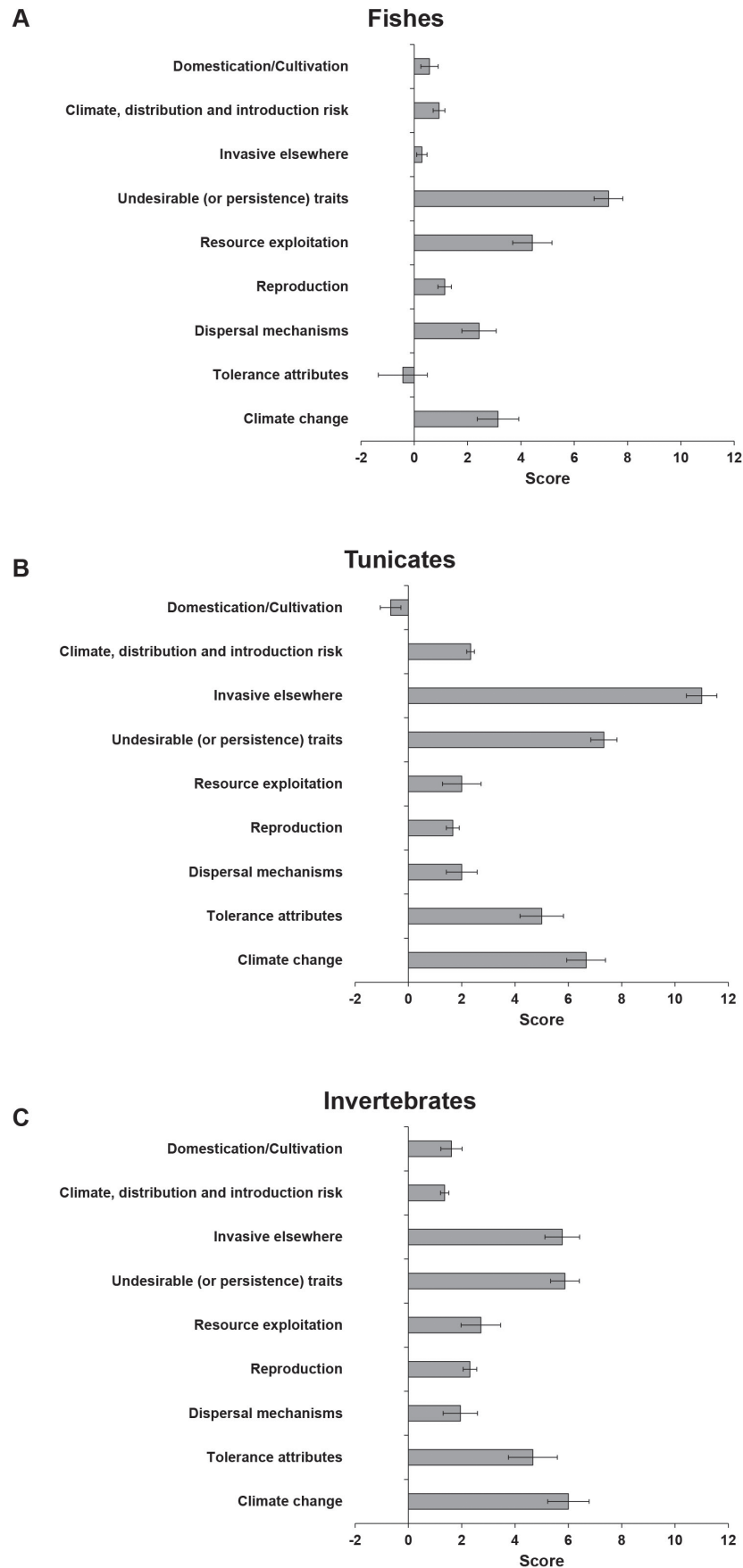


Figure 3. Mean (\pm SE) score partitioning for the AS-ISK scoring categories used to evaluate the invasiveness potential of the species ranked as high and very high risk (see Table 2): **(A)** Fishes ($n = 14$); **(B)** Tunicates ($n = 3$); **(C)** Invertebrates ($n = 35$).

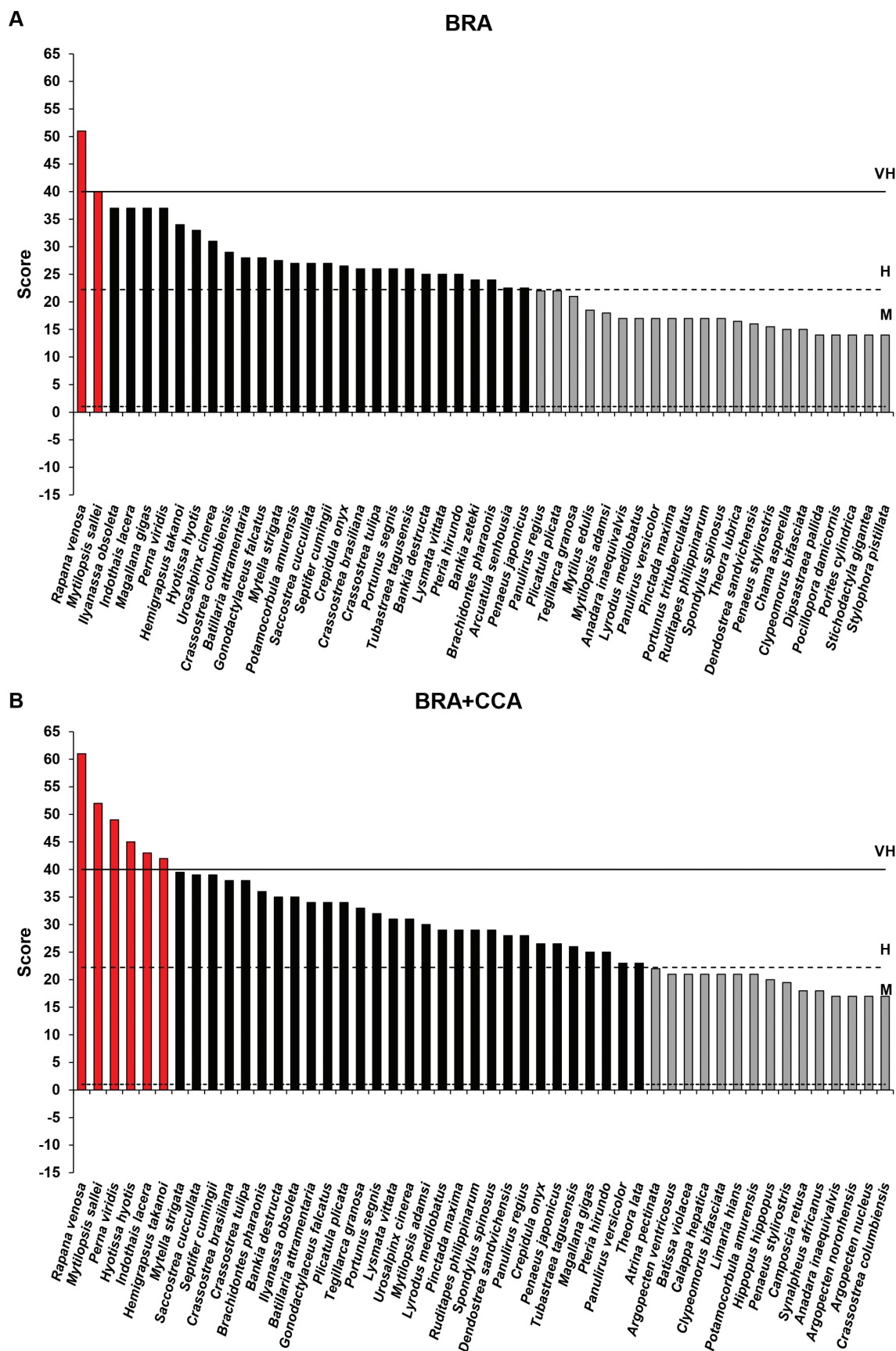


Figure 4. Outcome scores for the first 50 invertebrates (sorted by decreasing score) screened with the AS-ISK: (A) BRA scores; (B) BRA+CCA scores. Red bars = very high-risk species; Black bars = high-risk species; Gray bars = medium-risk species. Solid line = very high-risk (VH) threshold; Hatched line = high-risk (H) threshold; Dotted line = medium-risk (M) threshold (thresholds as per Table 2).

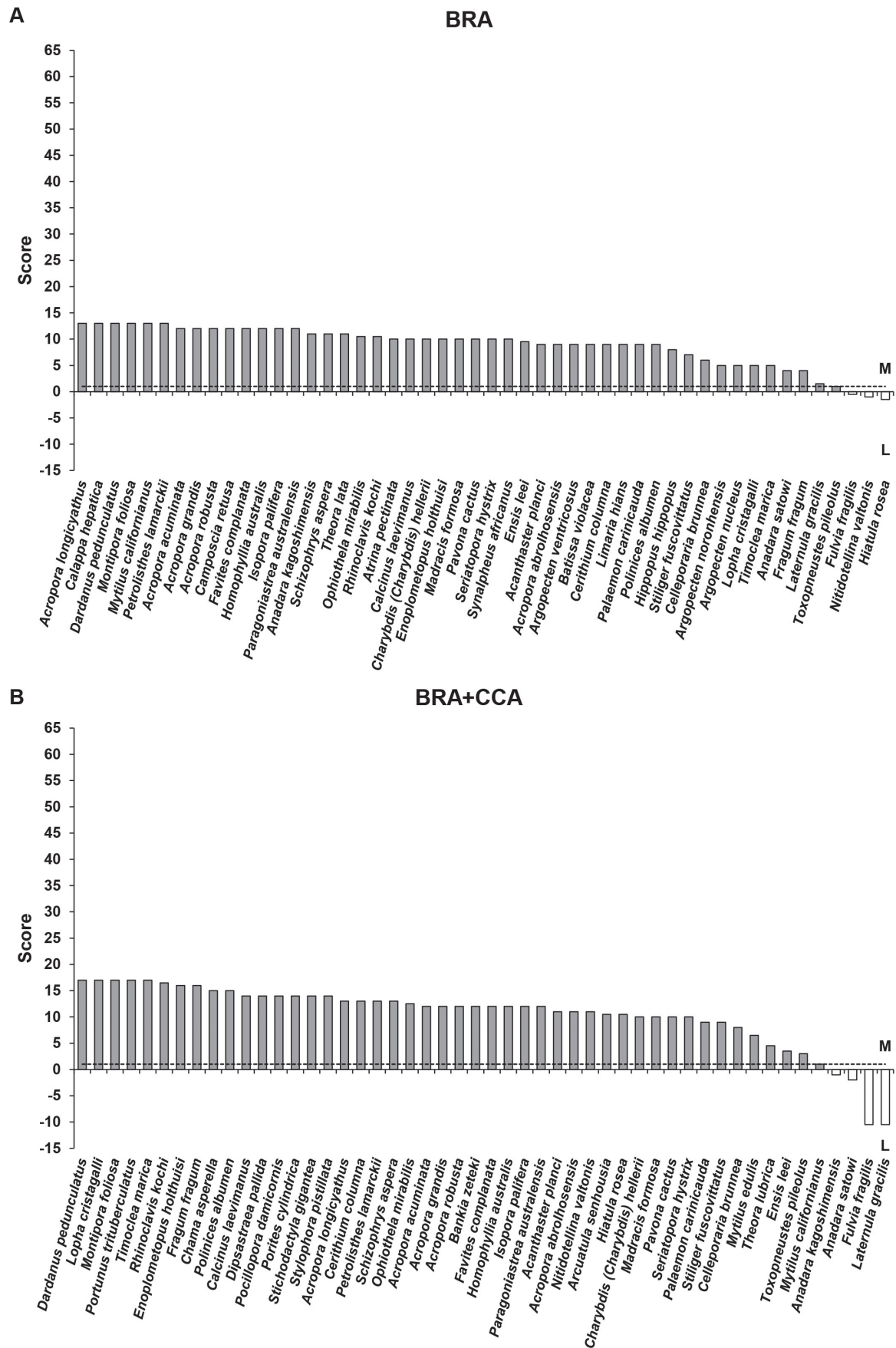


Figure 5. Outcome scores for the last 50 invertebrates (sorted by decreasing score) screened with the AS-ISK: (A) BRA scores; (B) BRA+CCA scores. Gray bars = medium-risk species; White bars = low-risk species. Dotted line = medium-risk (M) threshold (as per Table 2).

only (Fig. 4A, B). The CCA resulted in an increase in the BRA score for 60 species, in no change for 26, and in a decrease for the remaining 14 (Table 2).

Invertebrate invasiveness was largely attributed to history of invasion elsewhere as well as undesirable traits, and moderately influenced by tolerance attributes; however, there was very little influence from climate/distribution/introduction risk. Invasiveness potential of the high-risk invertebrates was substantially exacerbated by climate change (Fig. 3C).

Discussion

The present study represents the largest horizon-scanning of marine species thus far implemented in the risk screening of non-native species with WRA-type decision support tools. This screening of potential marine invasive species in the northern Gulf of Mexico has identified a total of four very high-risk and 40 high-risk species under current conditions (cf. BRA). When predicted future climate conditions were taken into consideration (BRA+CCA), 13 species were classed as very high-risk and 39 species as high-risk. The high proportion of positive delta values (i.e. the difference between BRA and BRA+CCA) for fishes, tunicates, and invertebrates (Table 2) indicates that climate change will enhance the chances of introduction, establishment, and dispersal in the northern Gulf of Mexico, with all very high-risk species classified as such due to an elevation of their BRA scores following the climate change assessment. This, in combination with numerous known pathways of introduction to the northern Gulf of Mexico such as importation of ornamental marine organisms, exchange of ballast water, presence of aquaculture activities, and support of international vessels, may further impact an already highly perturbed waterbody.

Highest-risk species

Invasiveness of the highest-scoring fish species was mostly attributed to their undesirable traits and resource exploitation. Notably, six of the seven very high-risk fish species are from the Family Scorpaenidae, all of which share many similarities with two invasive scorpaenid lionfishes: *Pterois volitans* and devil firefish *Pterois miles* (Bennett, 1828). These invasive traits include their biological attributes (i.e. extremely cryptic camouflage, stationary ambush/suction predator), depth occupation (i.e. remains on or near the sea floor or other substrata), and habitats. Scorpaenid fishes are popular in the aquarium trade, and like lionfishes, some species (e.g. *Rhinopias frondosa*) are likely to be released from aquaria to open waters (Courtenay and Stauffer 1990; Dill and Cordone 1997; Copp et al. 2005c), especially if they become 'a nuisance' (e.g. displaying aggressive and predatory behaviors: Duggan et al. 2006). Fishes within the family Scorpaenidae can have social and ecological impacts because they possess hollow spines on their dorsal fins, which inject a painful venom when stepped on or pressed into the skin (Halstead et al. 1955)—this poses a risk to human health and limits predation by native fishes (Anton et al. 2014). If established in the northern Gulf of Mexico in high densities, then the highly predatory feeding behavior, including prey naiveté of Scorpaenidae (*sensu* Anton et al. 2014), may lead to declines in native reef fishes and forage species like that seen in the lionfish invasion. *Muraenesox cinereus* was the only very high-risk fish species that is not within the Scorpaenidae, but it is known to exploit resources by opportunistic predation on a wide variety of small-bodied fishes and crustaceans (McCosker et al. 2021).

Many fishes categorized as *a priori* non-invasive, particularly within the Scorpaenidae, were subsequently ranked as high and very high risk in this study. As much of the available information on the biology of these species is only in the form of aquarium and hobbyist websites, these species may have been extremely difficult to classify appropriately prior to risk screenings being conducted. Moreover, the northern Gulf of Mexico has experienced the invasion of the con-familial Indo-Pacific lionfishes *P. volitans* and *P. miles* over the past decades—these invasions are viewed as amongst the most destructive experienced in the Caribbean and along the U.S. Atlantic coast (NOAA 2022). Lionfish populations in these areas have expanded at astonishing speeds, with noticeable impacts on native coral-reef fishes, the wider coral reef communities, and commercial and recreational fisheries (Lesser and Slattery 2011; Albins 2013; NOAA 2022). The Flower Garden Banks National Marine Sanctuary, a protected coral reef system in the northwestern Gulf of Mexico, has experienced a rapid invasion of lionfish (Johnston et al. 2016). There is growing concern that lionfishes will cause detrimental ecological impacts to the area (Belter et al. 2020). As such, scientists have noted the threat posed by lionfishes and fishes in the wider Scorpaenidae family (e.g. Lyons et al. 2019). Furthermore, many fish species screened in the present study may have scored high due to the risk assessment area's proximity to the subtropical and tropical conditions in Florida and the Caribbean Sea, respectively, and the known invasions documented in these areas (e.g. Debrot et al. 2011; Tidbury et al. 2021).

For the tunicates and invertebrates, high screening scores in the present study were largely attributed to their history of invasion elsewhere in the world, driven by their ability to tolerate a wide range of environmental conditions—this being underpinned by their biological characteristics facilitating establishment and dispersal. The highest-scoring tunicate, *Botrylloides violaceus*, originates from the western Pacific but has invaded marine waters worldwide, including the northeast Pacific, northeast and northwest Atlantic, Mediterranean, Adriatic, Black and North Seas, and Australian waters (Zaniolo et al. 1998; Pederson et al. 2005; Bock et al. 2011; Seebens et al. 2017). Invasion success of *B. violaceus* has been attributed to tolerance of a wide range of environmental conditions including temperature, salinity, and nutrients (Carman et al. 2007), as well as its ability to colonize a diversity of substrata and discourage settlement of other organisms due to rapid growth and potentially acidic tunic (Pisut and Pawlik 2002).

Amongst screened invertebrates, the highest scoring species *Rapana venosa*, which predated on ecologically and economically important bivalves such as mussels, oysters, clams, and scallops, has severely affected bivalve fisheries in the Black Sea (Zolotarev 1996). *Rapana venosa* is projected to negatively affect populations of the hard clam *Mercenaria mercenaria* (Linnaeus, 1758) in Chesapeake Bay, where it was estimated that a *R. venosa* population of just 1,000 individuals could reduce the annual harvest of *M. mercenaria* by 0.3–0.9% per year (Savini et al. 2002). In the northern Gulf of Mexico, *R. venosa* could impact the commercially important fisheries for eastern oyster *Crassostrea virginica* (Gmelin, 1791). The impact, however, may be less pronounced for *C. virginica* compared to *M. mercenaria*, as many oyster beds are found in salinities at or near the lower tolerance limits of *R. venosa* (Mann and Harding 2000). *Rapana venosa* also feeds on the carrion of mussels, oysters, fishes, and crabs (Zolotarev 1996) and thus may benefit in areas of heavy recreational fishing, which are common along the northern Gulf of Mexico coast. *Rapana venosa* may also compete with native predatory gastropod species of the genera *Busycon*, *Busycotypus*, *Neverita*, *Stramonita*, as well as other predators of bivalve mollusks, such as croakers and rays (Lercari and Bergamino 2011). Also, the invasion of Chesapeake Bay by *R. venosa* has changed mollusk shell resources for hermit crabs in a manner

that favors striped hermit crab *Clibanarius vittatus* (Bosc, 1801) over other species of hermit crab (Harding and Mann 1999). In addition, *R. venosa* may affect juvenile sea turtles in the northern Gulf of Mexico, such as the endangered Kemp's ridley sea turtle *Lepidochelys kempii* Garman, 1880 by fouling their carapaces as much as 20% of their body weight, thus weighing them down substantially (Lezama et al. 2013). However, that initial study reported the intriguing result that fitness (i.e. weight relative to carapace length) was greater in heavily biofouled than in 'clean' green turtle *Chelonia mydas* (Linnaeus, 1758) of the study area (Lezama et al. 2013).

Introduction vectors and dispersal pathways

Ballast water exchange is recognized as one of the primary introduction vectors of marine non-native species (Carlton and Geller 1993; Ruiz et al. 1997; Gollasch 2008), with this vector being mentioned for several of the very high-risk species from this study (e.g. *Hemigrapsus takanoi*, *Rapana venosa*, *Scorpaena scrofa*; see Suppl. material 3). Owing to the heavy presence of global ship traffic to support oil and gas operations, commercial fisheries, and other large industries, there is an increased risk of invasive species being introduced to the northern Gulf of Mexico in ballast waters taken onboard in other regions of the world. Previous studies have determined that species of diatom and dinoflagellate (Steichen and Quigg 2015), coral (Sammarco et al. 2010), and shrimp (Fuller et al. 2014) were all introduced to waters around shipping ports along the U.S. Gulf Coast. Most of the very high-risk species identified in the present study exhibit a pelagic stage during their life cycles and are known to produce a large number of offspring within a short time-frame. For example, an individual *R. venosa* can produce 182,000–1,302,000 eggs/year (Chung et al. 2013), with larvae that remain planktonic for 24 to 42 days (Harding 2006). Similarly, *Hemigrapsus* spp. are known to produce \approx 56,000 eggs per brood 5–6 times per year (Fukui 1988), with larvae being planktonic for up to one month before entering the juvenile stage (EOEEA 2012). These planktonic larval stages are well within the temporal window of opportunity for trans-oceanic transport in ballast water from distant locations (Kerckhof et al. 2006).

A common vector of introduction for fouling marine taxa is hull fouling of ships and mobile marine structures (e.g. mobile oil platforms) (Gollasch 2002; Godwin 2003; Wanless et al. 2010), with this vector being highlighted for several biofoulers in this study. The high-risk tunicate, *Didemnum vexillum*, is a colonial species native to Japan but invasive globally, including coastal areas of New Zealand, Europe, UK, and the east and west coasts of the U.S. *Didemnum vexillum* has a non-feeding tadpole larval stage that is present in the water column for < 24 hours (Olson 1983; Holland 2016). As such, transport of *D. vexillum* in ballast water or its spread via natural dispersion over long distances is unlikely. Rather, *D. vexillum* introductions have largely been as a hull foulant (Pederson et al. 2005), whereby adult colonies are able to remain attached because they create very little drag for the relatively slow-moving vessels (Clarke Murray et al. 2012). Non-native species can also be transported by way of associations with biofouling communities (i.e. smaller organisms living amongst interstitial crevices created by fouling organisms). For example, the screening of *Hemigrapsus takanoi* identified potential spread through juveniles or adults living amongst hull fouling communities (Suppl. material 3)—this introduction vector has previously been documented for grapsid crab *Hemigrapsus penicillatus* (Gollasch 1998), which was later re-identified as *H. takanoi* (as described in Wood et al. 2005). This phenomenon of 'hitchhiking invasives' also includes non-native marine organisms attached to, even colonizing, floating marine debris (Barry et al. 2023), which is an emerging research area.

Over the past few centuries, global movement of fishes and shellfishes has contributed substantially to the translocation of associated marine species, with many introductions being traced back to specific transport events (Carlton 1989; Minchin 2007). In the present study, the screening of *Rapana venosa* identified translocation with shellfishes as a potential vector for its introduction into the northern Gulf of Mexico, as it has been known to spread via transport of egg masses within marine aquaculture and live seafood (Mann and Harding 2000; ICES 2004; Harding and Mann 2005). Moreover, the U.S. is the world's leading importer of ornamental marine organisms, representing half of all marine ornamental trade (Rhyne et al. 2012). Most of the high-risk fishes screened in the present study include species that are popular in the aquarium trade (i.e. Scorpaenidae). As such, there is a high risk of introduction to the risk assessment area through this pathway via unintentional escape during transport or intentional release of unwanted fishes by owners or breeders (Semmens et al. 2004; Chan et al. 2019).

Although new species introductions and secondary spread are often attributed to human-mediated vectors, warmer waters under future climate change conditions are expected to enhance the natural dispersal and migration of invasive species from established populations (Dukes and Mooney 1999; Mieszkowska et al. 2006; Dodd et al. 2022). This is of particular concern for the northern Gulf of Mexico, which due to its proximity to subtropical and tropical conditions in Florida and the Caribbean Sea, respectively, is already at risk of invasions by naturally dispersing warmwater invasive species that are already present in neighboring areas. This pattern has previously been observed in other locations with some of the species screened for the present study. For example, the tropical *Perna viridis* arrived in Florida in the late 1990s and early 2000s from only two distinct introduction events (one in Tampa Bay in the Gulf of Mexico and the other in the northeast part of the state along the Atlantic Ocean), but subtropical conditions in these locations allowed for rapid natural spread south from both populations (Benson et al. 2001; Baker et al. 2007). This indicates that further spread from these populations into the northern Gulf of Mexico is likely. Although the invasive Indo-Pacific lionfishes, *Pterois volitans* and *Pterois miles*, were not screened in the present study, they have been screened for Grenada and St Vincent and the Grenadines, where only *P. volitans* is known to be present but both were ranked as 'high risk' under both current and future climate conditions (Tidbury et al. 2021). The expansion of lionfishes following their initial report for coastal waters of North Carolina in the 1990s has been rapid (Schofield 2010). Lionfishes are now considered ubiquitous throughout the Caribbean and Western Atlantic (Goodbody-Gringley et al. 2019), being pervasive from Rhode Island south to Belize and expanding west across the northern Gulf of Mexico (Campbell et al. 2022). Continued warming of Gulf of Mexico waters can facilitate the natural spread of other invasive warmwater taxa introduced adjacent to or within the risk assessment area.

Trans-boundary conservation management implications

The identification of potential high-risk non-native species through horizon scanning is becoming an essential component of government invasive species management strategies (Roy et al. 2015), with trans-boundary collaboration being recognized as vital for regional and large-scale initiatives (Graham et al. 2019; Otto and Brunson 2021). For instance, the European Union (EU) Regulation 1143/2014 on the control and management of alien invasive species (https://ec.europa.eu/environment/nature/invasivealien/index_en.htm) established an EU-wide strategy aimed at preventing and mitigating the adverse impacts of invasive species and

focuses on a target list of high-risk species, which were chosen based on a systematic horizon scan (Roy et al. 2015). Heavily influenced by risk analysis developments in the UK (Baker et al. 2008; Mumford et al. 2010; Booy et al. 2011), the EU regulatory structure, species list, and associated guidance are intended to allow Member States to focus prevention, early detection, and rapid response efforts on the species of highest concern. Concerns have been raised, however, that this piece of legislation has been less than successful in addressing horizon species and recent invaders (Lehtiniemi et al. 2016; Kleitou et al. 2021). Regardless, there already exists a large body of literature in peer-reviewed international journals on risk screening and horizon scanning of non-native species in Europe, both within and across countries (Copp et al. 2009; Booy et al. 2017; Dodd et al. 2022).

By contrast, the delayed adoption of regulation and horizon scanning in the U.S. and Canada relative to Europe (Copp et al. 2005b) especially at larger, trans-boundary scales, has resulted in most published reports for the marine environment being at the local or state levels with little evidence of interstate or larger regional assessments in the published literature (but see Lyons et al. 2020a). Already a heavily disturbed system, the northern Gulf of Mexico is being subjected to an expanding human population and the associated resource exploitation and habitat modification and loss. Therefore, the prevention of detrimental invasive species, which could exacerbate current pressures, is a potential focus for resource management in the region. Such a process could involve regional, cross-border collaboration among states as well as direction and support at the federal level in order to develop and implement effective strategic plans.

The non-native species screened in the present study have not yet been considered for U.S. ‘injurious wildlife’ listing, which is a designation under the Lacey Act (18 U.S.C. 42) for species that are injurious to the interests of human beings, agriculture, horticulture, forestry, wildlife, or wildlife resources of the United States. The resulting list of high- and very high-risk species (‘watchlist’) from this study will be provided to the U.S. Fish and Wildlife Service for further review and consideration for ‘injurious wildlife’ listing. Owing to lack of ‘injurious wildlife’ designation, there is a lack of higher-level federal prohibition on importation of watchlist species identified in the present study; however, interstate transport of any invasive species that is in violation of state laws (i.e. species prohibited by states) is a violation of the wildlife trafficking provisions of the Lacey Act. Thus, species identified as high or very high risk in the present study may necessitate prevention action implemented at the state level. However, differences among states in regulatory authority and impetus and support for exercising that authority could result in a patchwork of prevention measures that may undermine effective prevention at a regional scale. This may be the case for the northern Gulf of Mexico, where species introduced in one state where regulations are lacking could subsequently spread to other neighboring areas. Regional coordination can help to provide effective prevention strategies (e.g. through the Gulf and South Atlantic Regional Panel on Aquatic Nuisance Species) for the development of a strategic plan for preventing introductions of high- and very high-risk species across the northern Gulf of Mexico region through a combination of public awareness and regulatory approaches with a focus on addressing jurisdictional limitations (see Hill et al. 2018).

To inform the development of such a strategic plan, it may be necessary to conduct an evaluation of relevant regulatory authorities across northern Gulf of Mexico states as well as an assessment of the prevalence and economic importance of each high- and very high-risk species in the aquarium trade and other barriers to implementation. Horizon scan watchlists provide the opportunity to think critically and strategically about the live marine organisms currently imported into a

jurisdiction. Species that can enter a jurisdiction through trade, and are deemed to be high-risk, may need to be evaluated to determine whether their continued importation is justified, based on the potential risk the species may pose to the local ecology and economy. Such assessments can aid in the development of effective and feasible approaches for each species to strike a balance between effective prevention and minimization of negative economic impacts of regulation. Engagement with the aquatics trade to encourage the implementation of voluntary prevention measures could draw upon the information provided by the risk screenings in the present study. Furthermore, working directly with industry can provide the opportunity to reduce sales of species on the watchlist or provide customers with information on invasiveness and dangers of release. For example, in the UK, legislation and obligatory licensing to keep or release regulated non-native fishes proved to be the effective means of eliminating potential future invaders from the aquatics trade (Copp et al. 2005a; see also Chan et al. 2019; Li et al. 2021).

The watchlist composed of high- and very high-risk species from the present study can also raise awareness of species that could enter a jurisdiction within the northern Gulf of Mexico. This provides the opportunity to work across state (and indeed national) boundaries to make natural resource managers, wildlife conservation organizations, researchers, and the public mindful of the risk of these species, which may facilitate early detection of new introductions. For example, in the UK, marine conservation organizations that host beach cleaning events distribute leaflets describing potential invasive species. Furthermore, watchlists can help decrease the lag time between a new sighting of a non-native species and management decisions (Branquart et al. 2009; Debrot et al. 2011). As such, the watchlist created from the present study can be used to help inform early detection and rapid response plans and related strategic planning at both state and regional levels.

Identifying potential geographic entry points, also known as 'hotspots' (Tidbury et al. 2016, 2021), and the associated introduction vectors and pathways of a species on the watchlist, in combination with habitat suitability mapping (e.g. Dodd et al. 2022), can inform decisions in the early detection and rapid response process. These analytical approaches can focus efforts on optimal habitats where species are known to arrive, survive, and reproduce (Tidbury et al. 2016; Cook et al. 2019; Lyons et al. 2020b). Mapping hotspots and suitable habitat can inform adjustments to existing monitoring programs and facilitate targeted pathway management plans to enhance likelihood of detection by sampling in target locations or focusing on key vectors and pathways (Tidbury et al. 2021). Furthermore, identifying these geographic entry points and pathways can help to raise public awareness and focus citizen science monitoring programs (Morrisseau and Voyer 2014), thereby increasing the temporal and spatial breadth of monitoring efforts.

In conclusion, the present study has identified high- and very high-risk horizon species of relevance to the northern Gulf of Mexico's marine environment for the current decade, thus providing a list of potential future invaders for consideration by resource managers. Given the popularity of certain imported species and varieties, changes with trends in pathways, increases in propagule pressure, emergence of new information in the literature, and that novel species are invading new regions, horizon scans could be refined by repeating these scans at regular intervals (e.g. 5–10 years). The resulting watchlist will be used for consideration for 'injurious wildlife' listing, with the associated species risk-screening reports providing baseline information with which to conduct comprehensive risk assessments for the highest-risk species. The lack of available information for several species, which resulted in an *a priori* classification of 'non-invasive', particularly ornamental fish species, emerged as a potential limitation for predicting impacts to the risk assessment area,

and thus, may be a potential barrier for future scans when informing downstream management decisions.

In combination with other horizon scanning studies carried out in adjacent regions (e.g. Lieurance et al. in press) and with multi-state level coordination, the present study could facilitate a joined-up, regional approach across the northern Gulf of Mexico to address harmful marine invasive species before they arrive. In the same manner that horizon scanning applications elsewhere have endeavored to identify potential future invaders from neighboring countries to inform cross-border cooperation in non-native species management (e.g. Glamuzina et al. 2017; Dodd et al. 2019, 2022; Wei et al. 2021; Mumladze et al. 2022), the results of the present study are likely to prove useful at an international level, specifically for neighboring countries and Caribbean islands (e.g. Tidbury et al. 2021).

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Dedication

This manuscript is dedicated to Professors Joseph W. Goy and Gordon H. Copp. Joe was truly inspirational in the field of biological systematics. His contribution to the knowledge of decapod crustaceans was invaluable. Gordon was not only a hugely influential scientist, mentor and friend, but also a notable biologist who made significant contributions to the fields of aquatic ecology and invasive species as well as the management of freshwater and marine ecosystems. Professors Goy and Copp will be sorely missed.

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Authors' contribution

KAO: research conceptualization, sample design and methodology, investigation and data collection, writing – original draft, writing – review and editing; LV: research conceptualization, sample design and methodology, investigation and data collection, data analysis and interpretation; writing – original draft, writing – review and editing; WD: research conceptualization, sample design and methodology, investigation and data collection, writing – original draft, writing – review and editing; MEM: research conceptualization, sample design and methodology, investigation and data collection, writing – original draft, writing – review and editing; HB: investigation and data collection, writing – original draft, writing – review and editing; LH: research conceptualization, investigation and data collection, writing – original draft, writing – review and editing; SG: investigation and data collection, writing – review and editing; PS: investigation and data collection, writing – review and editing; SK: investigation and data collection, writing

– review and editing, SP: investigation and data collection, writing – review and editing; JD: investigation and data collection, writing – original draft, writing – review and editing; MRM: investigation and data collection, writing – original draft, writing – review and editing; MN: investigation and data collection, writing – review and editing; AF: investigation and data collection, writing – review and editing; TJJ: investigation and data collection, writing – review and editing; JP: investigation and data collection, writing – original draft, writing – review and editing; LB: investigation and data collection, writing – review and editing; WB: investigation and data collection, writing – review and editing; MW: investigation and data collection, writing – review and editing; DR: investigation and data collection, writing – review and editing; JAL: investigation and data collection, writing – review and editing; ER: investigation and data collection, writing – review and editing; JB: investigation and data collection, writing – review and editing; JG: investigation and data collection, writing – review and editing; AMO: investigation and data collection, writing – review and editing; ALEY: investigation and data collection, writing – review and editing; GHC: research conceptualization, sample design and methodology, investigation and data collection, data analysis and interpretation; writing – original draft, writing – review and editing.

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Supplementary material 1

Initial list of species

Authors: Kathryn A. O'Shaughnessy, Lorenzo Vilizzi, Wesley Daniel, Monica E. McGarrity, Hanna Bauer, Leslie Hartman, Stephen Geiger, Paul Sammarco, Steve Kolian, Scott Porter, Jessica Dutton, Matthew R. McClure, Michael Norberg, Alex Fogg, Timothy J. Lyons, Justin Procopio, Lauren Bantista, Wayne Bennett, Mary Wicksten, David Reeves, Julie Lively, Elizabeth Robinson, Jorge Brenner, Joseph Goy, Ashley Morgan-Olvera, Anna L. E. Yunnice, Gordon H. Copp

Data type: xlsx

Explanation note: Includes the initial list of species with physiological tolerances/ranges that matched that of the northern Gulf of Mexico. Species are presented by major taxonomic groups.

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Link: <https://doi.org/10.3391/ai.2023.18.4.114182.suppl1>

Supplementary material 2

Initial list questions

Authors: Kathryn A. O'Shaughnessy, Lorenzo Vilizzi, Wesley Daniel, Monica E. McGarrity, Hanna Bauer, Leslie Hartman, Stephen Geiger, Paul Sammarco, Steve Kolian, Scott Porter, Jessica Dutton, Matthew R. McClure, Michael Norberg, Alex Fogg, Timothy J. Lyons, Justin Procopio, Lauren Bantista, Wayne Bennett, Mary Wicksten, David Reeves, Julie Lively, Elizabeth Robinson, Jorge Brenner, Joseph Goy, Ashley Morgan-Olvera, Anna L. E. Yunnice, Gordon H. Copp

Data type: docx

Explanation note: Preliminary rapid risk assessment consisting of a short series of questions that helped determine the potential invasiveness of a species.

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Link: <https://doi.org/10.3391/ai.2023.18.4.114182.suppl2>

Supplementary material 3

Risk screening reports

Authors: Kathryn A. O'Shaughnessy, Lorenzo Vilizzi, Wesley Daniel, Monica E. McGarrity, Hanna Bauer, Leslie Hartman, Stephen Geiger, Paul Sammarco, Steve Kolian, Scott Porter, Jessica Dutton, Matthew R. McClure, Michael Norberg, Alex Fogg, Timothy J. Lyons, Justin Procopio, Lauren Bantista, Wayne Bennett, Mary Wicksten, David Reeves, Julie Lively, Elizabeth Robinson, Jorge Brenner, Joseph Goy, Ashley Morgan-Olvera, Anna L. E. Yunnice, Gordon H. Copp

Data type: xlsx

Explanation note: Contains species report outputs that were produced upon completion of each AS-ISK risk screening, and are organized by major taxonomic group: Fishes, Tunicates, Invertebrates.

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Link: <https://doi.org/10.3391/ai.2023.18.4.114182.suppl3>