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Evaluating invasion risk for freshwater fishes in South Africa



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Scan this QR code with your smart phone or mobile device to read online. **Background:** South Africa, as a signatory of the Convention on Biological Diversity, has an obligation to identify, prioritise and manage invasive species and their introduction pathways. However, this requires knowledge of the introduction pathways, factors influencing establishment success, invasive potential, current distributions and ecological impacts.

Objectives: To evaluate the Fish Invasiveness Screening Kit (FISK) to predict the invasion risk posed by fish species proposed for introduction into South Africa.

Method: FISK assessments were compiled for species whose invasion status in South Africa was known. A Receiver operating characteristic (ROC) analysis was conducted to calibrate the FISK for South Africa. The calibrated FISK was used to evaluate the risk that three species recently proposed for importation for aquaculture could become invasive in South Africa.

Results: A FISK score of 14 was identified as the threshold to delineate between species that could become invasive in South Africa and those that are unlikely to become invasive. Of the three species evaluated, *Silurus glanis* had a high risk of becoming invasive in South Africa, *Lates calcarifer* was likely to be invasive and *Oncorhynchus tshawytscha* was unlikely to be invasive in South Africa.

Conclusion: FISK was demonstrated to be a useful risk assessment tool to evaluate the invasion risk posed by species proposed for use in aquaculture. For the large number of fish imported for the pet trade, a rapid screening assessment to flag potentially high risk species was recommended prior to a full FISK assessment for flagged species.

Introduction

South Africa has a long history of vertebrate species importations (Picker & Griffiths 2017; van Rensburg et al. 2011). From the 1700s, freshwater fishes have been imported into the country (Ellender & Weyl 2014). While these introductions were legitimatised by the authorities at the time, they lacked appropriate consideration of the consequences of the ecological impacts on native biodiversity that followed. Some introduced fishes became invasive, detrimentally impacting native aquatic communities (Ellender & Weyl 2014). Currently, the presence of invasive alien fishes is considered the primary threat to most of South Africa's threatened endemic freshwater fishes (Tweddle et al. 2009).

Several publications have assessed the current knowledge of freshwater fish introductions in South Africa including introduction pathways (van Rensburg et al. 2011), failed and successful introductions (Ellender & Weyl 2014), the ecological cost and economic benefit of established introductions, especially conflict species (Ellender et al. 2014; Zengeya et al. 2017), and the management options for established introduced fishes (Woodford et al. 2017). Ellender and Weyl (2014) presented evidence for the introduction of 27 alien freshwater fishes into the wild in South Africa. Of these, 16 were evaluated as fully invasive (sensu Blackburn et al. [2011]; see Table 1). However, there are only sufficient data for five of these 16 invasive species to evaluate their ecological impact (sensu Blackburn et al. [2014]). Four species were evaluated as having 'major impacts' and one was evaluated as having had a 'massive impact' (see Table 1 for delineation of impact categories).

Ellender and Weyl (2014) identified that while enhancement of fisheries was one of the main pathways for early introductions, the ornamental fish trade and aquaculture are currently the most

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TABLE 1: List of freshwater fish species introduced into the water courses of South Africa including their invasion status [sensu Blackburn et al. (2011)] and invasion impact category [sensu Blackburn et al. (2014)] after Richardson et al. (2010) and Ellender and Weyl (2014).

Species	Introduction Ellender and Weyl (2014)		Establ	ishment	Impact		
			Blackburn	et al. (2011)	Blackburn et al. (2014)		
	Date of first introduction	Vector	No. of basins	Invasion status	Impact category	Impacts	
Carassius auratus (Linnaeus, 1758)	1726	ORN	4	E	DD	-	
Coptodon zillii (Gervais, 1848)	1959	AQU	0	F	NA	-	
Ctenopharyngodon idella (Valenciennes, 1844)	1967	BIO	2	E	DD	G	
Cyprinus carpio Linnaeus, 1758	1859	ANG	15	E	DD	-	
Gambusia affinis (Baird & Girard, 1853)	1936	BIO	8	E	DD	-	
Hypophthalmichthys molitrix (Valenciennes, 1844)	1975	AQU	1	D2	DD	-	
Lepomis macrochirus Rafinesque, 1819	1939	ANG	16	E	DD	-	
Micropterus dolomieu (Lacepède, 1802)	1937	ANG	12	E	MR	С, Р	
Micropterus floridanus (Lesueur, 1822)	1984	ANG	?	E	DD	-	
Micropterus punctulatus (Rafinesque, 1819)	1940	ANG	13	E	DD	С, Р	
Micropterus salmoides (Lacepède, 1802)	1928	ANG	18	E	MR	С, Р	
Oncorhynchus mykiss (Walbaum, 1792)	1897	ANG	16	E	MR	С, Р	
Oreochromis andersonii (Castelnau, 1861)	1982	AQU	0	F	NA	-	
Oreochromis aureus (Steindachner, 1864)	1910	AQU	0	F	DD	-	
Oreochromis niloticus (Linnaeus, 1758)	1955	AQU	2	D2	MA	С, Н	
Pangasius sanitwongsei Smith, 1931	2012	ORN	1	B3	DD	-	
Perca fluviatilis Linnaeus, 1758	1915	ANG	2	C3	DD	-	
Poecilia reticulata (Peters, 1859)	1912	ORN	3	E	DD	-	
Pterygoplichthys disjunctivus (Weber, 1991)	2000	ORN	1	D2	DD	С	
Salmo salar Linnaeus, 1758	1896	ANG	0	F	NA	-	
Salmo trutta Linnaeus, 1758	1892	ANG	14	E	MR	С	
Salvelinus fontinalis (Mitchill, 1815)	1890	ANG	0	F	NA	-	
Sarotherodon galilaeus (Linnaeus, 1758)	1959	AQU	0	F	NA	-	
Serranochromis robustus (Günther, 1864)	1960	ANG	0	F	NA	-	
Tinca tinca (Linnaeus, 1758)	1896	ANG	1	C3	DD	-	
Xiphophorus hellerii Heckel, 1848	1974	ORN	3	D2	DD	-	
Xiphophorus maculatus (Günther, 1866)	2006	ORN	1	B3	DD	-	

Source: Vectors (after Ellender and Weyl 2014): BIO = Biological control of mosquitoes or macrophytes; ANG = angling; AQU = Aquaculture; ORN = ornamental (pets), Invasion state (after Blackburn et al. 2011): B3 = Individuals transported beyond limits of native range and directly released into novel environment; C1 = Individuals surviving in the wild (i.e. outside of captivity D2 = Self-sustaining; population in the wild, with individuals surviving and reproducing a significant distance from the original point of release; E = Fully invasive species, with individuals dispersing, surviving and reproducing at multiple sites across a greater or lesser spectrum of habitats and extent of occurrence; F = Failed introduction, Impact category (after Blackburn et al. 2014): MA = massive impacts cause at least local extinction of species, and irreversible changes in community composition; even if the alien species is removed the system does not recover its original state; MR = major impacts causing local or population extinction of at least one native species, but no changes to the structure of communities and the abiotic or biotic composition of ecosystems; MO = moderate impacts causing declines in the population densities of native species, but no declines in native population densities; ML = minimal impacts being unlikely to have caused deleterious impacts on the native biota or abiotic environment; DD = Data deficient when the best available evidence indicates that it has individuals existing in a wild state in a region beyond the boundary of its native geographic range, but either there is no reliable evidence that it has or had individuals existing in a wild state in a region beyond the boundary of its native geographic range.

Impacts: C = Competition; P = Predation; G = Grazing/herbivory; H = Hybridisation; D = Disease/parasites.

important pathways for new importations into South Africa. Proposals to import species for aquaculture are frequently received by the Department of Agriculture Forestry and Fisheries (DAFF), and there is a need to assess the risks posed should these species escape and become established, and subsequently invasive, in the wild. In addition, there is a steady stream of ornamental fishes being imported into the country and the potential invasion risk posed by these species also needs to be assessed. The current permitted list for alien ornamental fish species that may be imported into South Africa is over 1000 species and is highly likely to include species that pose significant risks to inland waters of South Africa if they are deliberately or accidentally released. Therefore, there is a need to develop a protocol to assess the invasion risk associated with proposed importations.

As a signatory of the Convention on Biological Diversity and its Strategic Plan for Biodiversity 2011–2020 (UNEP 2011), South Africa has an obligation to the international community Biodiversity 2011–2020, that is, to identify, prioritise and manage both alien and invasive species and their invasion pathways (UNEP 2011). The South African government has sought to fulfil this obligation through the promulgation of the *National Environmental Management: Biodiversity Act* (Republic of South Africa 2004) and its associated alien invasive species lists and regulations (Republic of South Africa 2014; Wilson et al. 2017). However, the resources available for managing established invasions in freshwater ecosystems are limited (Woodford et al. 2017) and there is a growing need to mitigate against potential future fish invasions.

to implement the Aichi Target 9 of the Strategic Plan for

Managing the risk posed by importing a species into a country requires that the economic benefits accrued by the species be weighed against its potential environmental impacts, that is, using a risk assessment framework. Four levels of risk assessment are currently used internationally: a full risk assessment, trait-based risk assessments, statistical assessments and rapid screening (Keller & Kumschick this issue). Full assessments are expensive and are usually conducted for a single species as they require a considerable time investment to review all the relevant literature available. Trait-based assessments use standard questions, scoring responses to evaluate the risk of the species introduction, for example, the Fish Invasiveness Screening Kit (FISK) (Copp, Garthwaite & Gozlan 2005b). Statistical approaches use statistical or machine learning algorithms to identify patterns in trait data that predict invasiveness or adverse impact, for example, Marchetti et al. (2004), Ribeiro et al. (2008) and Howeth et al. (2016). Rapid screening is a simple assessment that is usually based around just two species attributes: climate match and whether the species has a history of causing harm elsewhere in its introduced range. If a species has both a strong climate match and a history of impacts, it is designated as likely to cause harm in its introduced range, for example, the Rapid Screen developed by the United States Fish and Wildlife Service (Hoff 2014).

In this paper we evaluated whether the trait-based FISK assessment is a suitable tool to evaluate the invasion risk posed by fish species imported into South Africa. We selected the FISK because it has been widely used globally including Europe, Australia, North America and Asia (see Lawson et al. 2015; Mastitsky et al. 2010; Piria et al. 2016; Puntila et al. 2013; Tarkan et al. 2014; Vilizzi and Copp 2013 for examples). To calibrate FISK for South Africa, we applied it retrospectively to species that have been released into water courses in the country using the invasion status as determined by Ellender and Weyl (2014) as the outcome. We then applied the calibrated FISK to evaluate the invasion risks for three species that have recently been proposed for importation for aquaculture in South Africa.

Methods

The FISK evaluates a species' invasion risk based on a questionnaire comprising 49 questions in two categories, 'Biogeography and Historical' and 'Biology and Ecology', with three ('Domestication and Cultivation', 'Climate and Distribution' and whether it is 'Invasive Elsewhere') and five ('Undesirable Traits', 'Feeding Guild', 'Reproduction', 'Dispersal Mechanisms' and 'Persistence Attributes') subcategories, respectively (Copp 2013; Copp, Garthwaite & Gozlan 2005a; Copp et al. 2009). To evaluate the utility of FISK, the 27 alien fish species recorded as having been released into water courses in South Africa and their evaluated invasion status listed in Ellender and Weyl (2014) were split into species considered invasive (n = 16; classified D and E; sensu Blackburn et al. [2011]) and not invasive (species classified as B, C or F; n = 11; see Table 1). Six experts (B.R.E., M.E.A., S.M.M., R.J.W., O.L.F.W. and D.J.W.) independently completed FISK assessments based on published data and online resources such that each species was evaluated by at least three different experts. The whole country was used as the recipient area, which complicated the analyses because South Africa contains more than 10 Köppen-Geiger climate types (Peel, Finlayson & McMahon 2007).

Receiver operating characteristic (ROC) curves, a graphical technique plotting selectivity vs. specificity for visualising,

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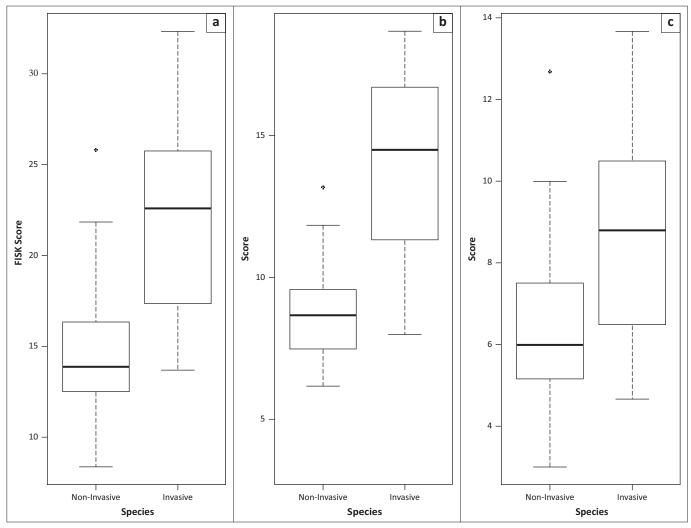
organising and selecting classifiers based on their performance (Fawcett 2006), were constructed to assess the predictive ability of FISK to identify potentially invasive fish species in South Africa. The area under the curve (AUC) is a measure of the accuracy of the calibration analysis. Typically, the AUC ranges between 0.5 (0% accurate, i.e. cannot discriminate between true positives and true negatives) and 1.0 (100% accurate) (Fawcett 2006). The closer the AUC is to 1.0, the better the ability of FISK to differentiate between invasive and non-invasive species. In addition, Youden's index (Youden 1950) was used to identify the threshold FISK score that maximises the probability of correct classification while minimising that of incorrect classification (sensu Copp et al. [2009]). The minimum and maximum FISK scores for each species were used to construct ROC curves to determine the thresholds for the 'medium' risk and 'high' risk categories, respectively. In addition, the threshold from the ROC curve for the average FISK score was found to discriminate between species that were invasive in South Africa and those that were not within the 'medium' risk category. Therefore, the 'medium' risk category was divided into 'upper medium' and 'lower medium' risk categories to distinguish between species with a higher invasion risk and those unlikely to become invasive. ROC analyses were conducted using the pROC package version 1.8 (Robin et al. 2011) for R 3.3.0 (R Development Core Team 2016).

The overall FISK score and the scores for its respective components and subcomponents were evaluated to determine whether they could be used as single-parameter surrogates for the full assessment. Shapiro–Wilks tests were used to determine whether the variables were normally distributed. Only three variables were not normally distributed: domesticated or cultivated; climate and distribution; and feeding guild. For normally distributed variables, the *t*-test was used to determine whether there was a difference between the two means. For non-normally distributed variables, the Mann–Whitney *U* test was used to determine whether there was a difference between the two medians. Box plots were used to visualise the outputs of the FISK assessments. All analyses were conducted using R 3.3.0 (R Development Core Team 2016).

In addition, FISK assessments were conducted for three species for which applications for importation for aquaculture have recently been received by DAFF (Chinook salmon *Oncorhynchus tshawytscha* [Walbaum, 1792], Barramundi *Lates calcarifer* [Bloch, 1790] and Wels catfish *Silurus glanis* Linnaeus, 1758), to evaluate the risk posed by their proposed importation and provide an example of the application of the FISK in the South African context.

Results

The FISK scores were significantly different between the invasive and non-invasive species (*t*-test, p = 0.002, Figure 1). Of the two categories of the FISK, a significant difference was found between invasive and non-invasive species for 'Biogeography and Historical' (*t*-test, p < 0.001, Figure 1), but not



Source: Average FISK scores, Fish Invasiveness Screening Kit

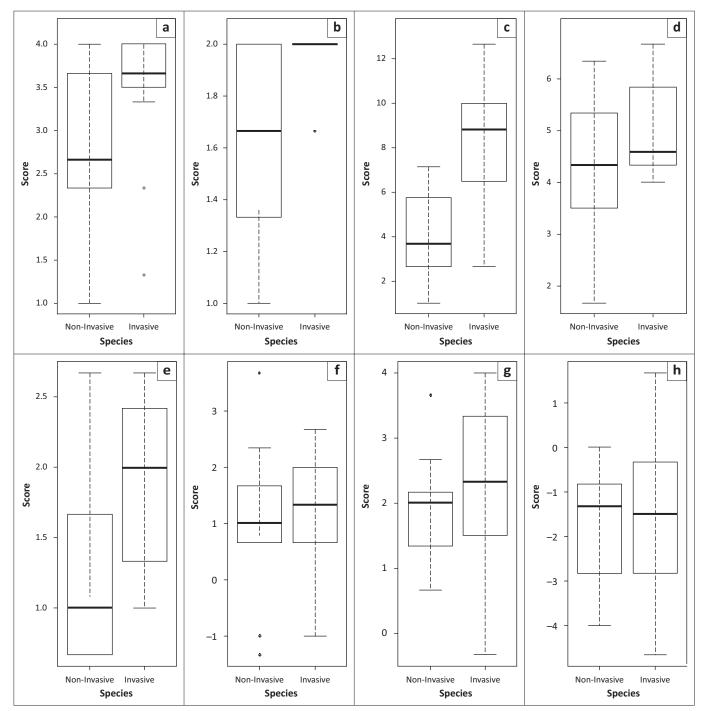
(a), FISK score; (b), Biogeography and History; (c), Biology and Ecology.

FIGURE 1: Box plots summarising the FISK score and its two components, 'Biogeography and History' and 'Biology and ecology' for the species introduced into South Africa.

for 'Biology and Ecology' (*t*-test, p = 0.06, Figure 1). Of the eight subcategories, 'Climate and Distribution' (Mann–Whitney U, p = 0.01) and 'Invasive Elsewhere' (*t*-test, p = 0.002) were significantly different between the invasive and non-invasive species (Figure 2). Both factors are included in the 'Biogeography and History' component of the FISK assessment. In addition, the 'Feeding guild' subcategory of the 'Biology and Ecology' component was also significantly different between the invasive and non-invasive and non-invasive species (Mann–Whitney U, p = 0.02; Figure 2).

The ROC curve resulted in an AUC well above 0.5 for the average (0.8409), maximum (0.8438) and minimum (0.8324) FISK scores (Figure 3). This indicated that FISK was able to discriminate reliably between invasive and non-invasive freshwater fish species in South Africa. The ROC assessment determined that the threshold for 'medium' risk was at a FISK value of 10.75, whereas that for 'high' risk was at 18.25. The threshold between 'upper medium' and 'lower medium' risk was 14.00. Of all the species assessed, 12 were classified as 'high' risk, two of which were classified non-invasive: redbelly tilapia *Coptodon zillii* (Gervais 1848) and Israeli tilapia *Oreochromis*

aureus (Steindachner 1864) (see Table 2). The remaining species were evaluated as posing 'medium' risk of becoming invasive, with the exception of the giant pangasius Pangasius sanitwongsei Smith, 1931, which was classified as having a 'low' risk of becoming invasive. Species in the 'medium' risk category that have become invasive in South Africa had 'upper medium' FISK scores, with the exception of Xiphophorus helleri Hackel, 1848, which was classified in the 'lower medium' risk category. Species classified as non-invasive in the 'upper medium' risk category were Oreochromis andersonii (Castelnau 1861), Tinca tinca (Linnaeus, 1758) and Serranochromis robustus (Günther, 1864). The ROC assessment was conducted to determine thresholds for the 'Biogeography and Historical' and 'Biology and Ecology' categories. The threshold scores for the Biogeography and Historical category were 5.5 for 'medium' risk and 9.75 for 'high' risk with the threshold between upper and lower 'medium' risk at 8.33. For Biology and Ecology, the thresholds were 6.5 for 'high' risk and 4.5 between upper and lower 'medium' risk. The 'medium' risk threshold for Biology and Ecology was negative infinity implying that there was no 'low' risk designation for this category.



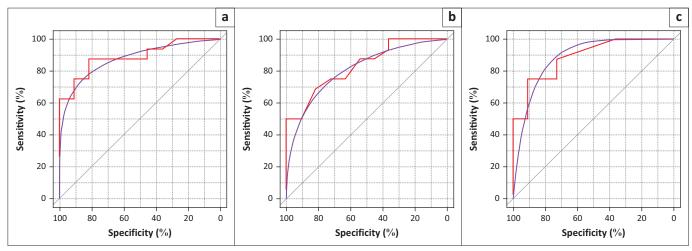
Source: Average FISK component scores, Fish Invasiveness Screening Kit

(a), Domestication and Cultivation; (b), Climate and Distribution; (c), Invasive elsewhere; (d), Undersirable traits; (e), Feeding guild; (f), Reproduction; (g), Dispersal mechanisms; (h), Persistence atributes.

FIGURE 2: Box plots summarising the eight subcategories contributing to the 'Biogeography and History' and 'Biology and Ecology' components of the FISK assessment for the species introduced into South Africa.

Finally, three species under consideration for potential aquaculture ventures were evaluated using the calibrated FISK: *L. calcarifer, O. tshawytscha* and *S. glanis. Silurus glanis* was evaluated to be a 'high' risk species, *L. calcarifer* an 'upper medium' risk species and *O. tshawytscha* a 'lower medium' risk species (Table 3). *Silurus glanis* was also evaluated to be a 'high' risk species for both 'Biogeography and History' and 'Biology and Ecology' categories. *Lates calcarifer* scored a 'lower medium' risk for the 'Biogeography and History' category but was classified a 'high' risk

species for the 'Biology and Ecology' category. It is thus likely that *L. calcarifer* would at least establish populations in South Africa if released into the wild. *Oncorhynchus tshawytscha* scored an 'upper medium' risk for the 'Biogeography and History' category but was classified 'lower medium' risk for the 'Biology and Ecology' category. Therefore, it is expected that *O. tshawytscha* would share the fate of Atlantic salmon *Salmo salar* (Linnaeus, 1758) and brook trout *Salvelinus fontinalis* (Mitchill, 1815) and fail to establish in South Africa.



Source: ROC curves of FISK scores, Fish Invasiveness Screening Kit

(a) Fisk average; (b) Fisk max; (c) Fisk min

FIGURE 3: Receiver operating characteristic curves (actual and smoothed) for the FISK assessment for the invasion potential of species introduced into South Africa.

TABLE 2: Results of the FISK assessment of freshwater fish introduced into South Africa where the outcome of the introduction is known presenting the scores and risk categories for the FISK assessment and its Biogeography and History (B & H) and Biology and Ecology (B & E) categories. All FISK scores are the mean of three independent assessor scores. Medium risk is depicted as Upper Medium (UM) and Lower Medium (LM) as described in the text.

Species	FISK score	FISK risk	B & H score	B & H risk	B & E score	B & E risk
Carassius auratus (Linnaeus, 1758)	25.17	High	14.5	High	10.7	High
Coptodon zillii (Gervais, 1848)	21.83	High	11.8	High	10.0	High
Ctenopharyngodon idella (Valenciennes, 1844)	22.00	High	12.3	High	9.7	High
Cyprinus carpio Linnaeus, 1758	32.33	High	18.7	High	13.7	High
Gambusia affinis (Baird & Girard, 1853)	25.00	High	15.7	High	9.3	High
Hypophthalmichthys molitrix (Valenciennes, 1844)	27.67	High	17.3	High	10.3	High
Lepomis macrochirus Rafinesque, 1819	18.00	High	10.7	High	7.3	High
Micropterus dolomieu (Lacepède, 1802)	23.17	High	14.8	High	8.3	High
Micropterus floridanus (Lesueur, 1822)	21.67	High	13.3	High	8.3	High
Micropterus punctulatus (Rafinesque, 1819)	15.00	UM	10.0	High	5.0	UM
Micropterus salmoides (Lacepède, 1802)	24.50	High	15.3	High	9.3	High
Oncorhynchus mykiss (Walbaum, 1792)	21.83	High	14.5	High	7.3	High
Oreochromis andersonii (Castelnau, 1861)	16.67	UM	8.7	UM	8.0	High
Oreochromis aureus (Steindachner, 1864)	25.50	High	12.8	High	12.7	High
Oreochromis niloticus (Linnaeus, 1758)	26.33	High	15.7	High	10.7	High
Pangasius sanitwongsei Smith, 1931	8.33	Low	3.3	Low	5.0	UM
Perca fluviatilis Linnaeus, 1758	13.00	LM	7.0	LM	6.0	UM
Poecilia reticulata (Peters, 1859)	14.17	UM	8.5	UM	5.7	UM
Pterygoplichthys disjunctivus (Weber, 1991)	29.00	High	15.3	High	13.7	High
Salmo salar Linnaeus, 1758	12.33	LM	8.0	LM	4.3	LM
Salmo trutta Linnaeus, 1758	16.67	UM	12.0	High	4.7	UM
Salvelinus fontinalis (Mitchill, 1815)	12.67	LM	9.7	UM	3.0	LM
Sarotherodon galilaeus (Linnaeus, 1758)	12.17	LM	6.2	LM	6.0	UM
Serranochromis robustus (Günther, 1864)	14.83	UM	9.5	UM	5.3	UM
Tinca tinca (Linnaeus, 1758)	16.00	UM	9.0	UM	7.0	High
Kiphophorus hellerii Heckel, 1848	13.67	LM	8.0	LM	5.7	UM
Xiphophorus maculatus (Günther, 1866)	13.83	LM	8.2	LM	5.7	UM

Source: Average of FISK score by species, Fish Invasiveness Screening Kit, risk level as determined from ROC analysis

TABLE 3: Results of the FISK assessment of freshwater fish species proposed for importation into South Africa for the establishment of aquaculture operations presenting the scores and risk categories for the FISK assessment and its Biogeography and History (B & H) and Biology and Ecology (B & E) categories. All FISK scores are the mean of three independent assessor scores. Medium risk is depicted as Upper Medium (UM) and Lower Medium (LM) as described in the text.

Species	FISK score	FISK risk	B & H score	B & H risk	B & E score	B & E risk
Lates calcarifer (Bloch, 1790)	14.3	UM	5.7	LM	8.7	High
Oncorhynchus tshawytscha (Walbaum, 1792)	12.5	LM	8.5	UM	4.0	LM
Silurus glanis Linnaeus, 1758	21.3	High	10.3	High	11.0	High

Source: Average of FISK score by species, Fish Invasiveness Screening Kit, risk level as determined from ROC analysis of species where the invasion status was known.

The evaluations of the respective assessors were reviewed to determine whether there was evidence of bias between the assessors. Overall, the average FISK score for the species assessed was 19.07. The average FISK score for the species evaluated by four of the assessors was within 10% of this value. Two assessors, A3 and A4, had average FISK scores about 30%

away from the overall average score, A3 30% above the average and A4 30% below the average (Table 4). The respective scores assigned by the assessors per species are presented in Figure 4a. The range in FISK scores varied between 0.5 and 25 (Figure 4b), similar to the ranges found by Copp et al. (2009) for Europe.

Discussion

The retrospective FISK assessment conducted using species where the invasion status [sensu Blackburn et al. (2011)] were known was found to be a good predictor of whether a species would become invasive in South Africa. Only two of the species identified by FISK as having a high risk of becoming invasive have so far failed to establish: C. zillii and O. aureus. The reason for these failures could be because of a lack of introduction effort as a result of more efficient species being introduced for aquatic vegetation control, for example, grass carp Ctenopharyngodon idella (Valenciennes, 1844) or because they were similar to species indigenous to the region, for example, Mozambique tilapia Oreochromis mossambicus (Peters, 1852). However, there is anecdotal evidence that O. aureus has persisted since the 1960s in isolated farm dams near Stellenbosch in the Western Cape but has not been spread from these dams (ND Impson Cape Nature, personal communication). This indicates that this species is climate matched to a part of South Africa and could yet become invasive there if spread from these locations.

A number of species that are invasive in South Africa were classified as 'upper medium' risk including the bluegill sunfish *Lepomis macrochirus* Rafinesque, 1819, brown trout *Salmo trutta*

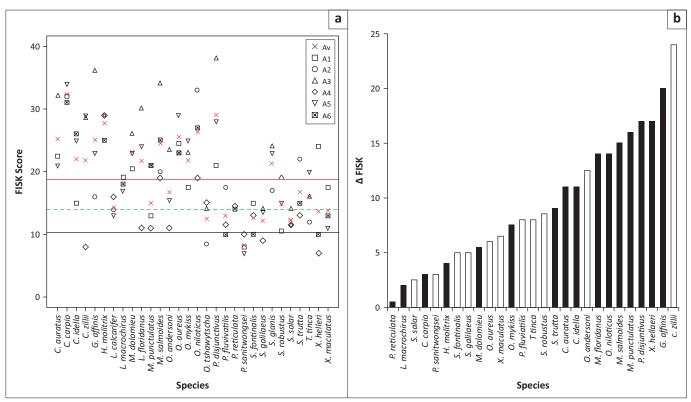
Linnaeus, 1758 and spotted bass *Micropterus punctulatus* Rafinesque, 1819. Other authors, for example, Copp et al. (2009) and (Tarkan et al. 2014), consider that the 'medium' risk classification implies that the species will not be invasive. However, the results clearly indicate that the 'upper medium' risk classification includes species that are invasive in South Africa. Therefore, greater care should be applied in evaluating 'medium' risk species and a division of 'medium' risk into an upper and lower risk levels is recommended. The thresholds for the high- and medium risk species was lower than those found in studies in Europe, the Balkans and Turkey (Copp et al. 2009; Simonovic et al. 2013; Tarkan et al. 2014), similar to those for the Murray-Darling Basin in Australia (Vilizzi & Copp 2013), but higher than those for Florida (Lawson et al. 2015).

The finding that 'Climate and Distribution', 'Invasive Elsewhere' and 'Trophic Group' were the most important factors determining establishment success supports the findings of previous studies, for example, Moyle and Marchetti (2006) and García-Berthou (2007), and may provide

TABLE 4: Summary of the assessor estimates for the FISK assessment of freshwater fish for South Africa (Av represents the average FISK scores for that species and A1 to A6 represent the 6 independent assessors). Accuracy values represent the percentage of an individual assessor's scores that fell into the same class as the Average FISK score.

Assessor	Av	A1	A2	A3	A4	A5	A6
Average FISK score	19.07	18.27	18.23	24.80	13.21	20.84	19.14
Accuracy in predicting average class (%)	-	60	55	73	70	89	93

Source: Average FISK assessor scores, Fish Invasiveness Screening Kit



Source: Assessor FISK scores, Fish Invasiveness Screening Kit, range in FISK scores by species

FIGURE 4: Summary of the FISK scores by species: (a) plot of the respective FISK scores given by the assessors by species (Av represents the average of the assessor scores for that species and A1 to A6 represent the scores given by the 6 independent assessors) and (b) the range in the FISK scores per species (black bars represent invasive species and white bars non-invasive species). Red line in 4a represents medium/high risk threshold value for South Africa, green dashed line represents lower medium/ upper medium threshold and black line represents low/medium threshold.

a template for a rapid risk assessment approach that could be used in South Africa. Although fisheries scientists are usually accurate in matching the climate between source and recipient regions (Bomford, Barry & Lawrence 2010), *S. salar* and *S. fontinalis* were rather optimistically introduced for angling in the late 19th century (de Moor & Bruton 1988) but failed to establish because of poor climate matching. Because climate matching is one of the important categories for determining whether a species will become invasive, the level of climate matching between the source and recipient areas should be conducted with greater accuracy, for example, using the Australian Department of Agriculture's CLIMATCH utility (Australian Department of Agriculture 2010), rather than on the Köppen-Geiger climate regions used in the FISK.

Calibration of the FISK for South Africa provided an opportunity to conduct risk assessments for three species proposed for importation for aquaculture, S. glanis, L. calcarifer and O. tshawytscha. Of these, S. glanis was classified as a high risk species that could become invasive in South Africa. This species is the third largest freshwater fish and an internationally renowned angling species that is likely to be spread by anglers in South Africa should it become released in the wild, as has happened in Spain (Clavero & García-Berthou 2006) and in South Africa with extra-limital introductions of African sharptooth catfish Clarias gariepinus (Burchell, 1822) (Weyl et al. 2016). The illegal movement of fish between water bodies can seriously compromise both recreational fisheries and conservation programmes (Gozlan et al. 2010; Johnson, Arlinghaus & Martinez 2009) and steps to manage the secondary spread of fish species within South Africa need to be established (e.g. Vander Zanden & Olden 2008). The FISK assessment classified L. calcarifer as 'upper medium' risk, suggesting it is likely to be invasive in South Africa, whereas O. tshawytscha was classified 'lower medium' risk and less likely to be invasive. The results of the FISK assessment for these species indicate that the importation of S. glanis into South Africa is not recommended because of the high risk that the species would become invasive. Proposed aquaculture ventures for L. calcarifer should be treated cautiously as there is an 'upper medium' risk that this species could become invasive in South Africa. Overall, these assessments demonstrated that FISK is an accurate, time-efficient and defensible risk assessment tool to evaluate the invasive potential of fish species proposed for importation.

While the vectors and history of introductions into the wild are fairly well documented for fishery, aquaculture and biological control species (see Ellender & Weyl 2014), the magnitude of fish introductions into natural water courses via the ornamental fish trade has never been formally evaluated in South Africa. The extent of ornamental fish releases is frequently underestimated globally (Welcomme 1992) and because of the widespread dispersal of ornamental fish to homes and businesses, unwanted pets can potentially be released into all freshwater habitats (Padilla & Williams 2004). Ornamental fish enter natural waterways through the dumping of unwanted pets, escape from garden ponds or breeding farms (e.g. during floods) and the ritualistic release of species during religious practices (Copp, Wesley & Vilizzi 2005c; Duggan, Rixon & MacIsaac 2006; Padilla & Williams 2004). Healthy ornamental fish are most commonly released when owners tire of them, or the fish become too large, aggressive, expensive to maintain or prolific for their aquaria (Duggan et al. 2006; Gertzen, Familliar & Leung 2008). In the United Kingdom, for example, ornamental fish releases were positively related to human population density, the ornamental trade (density of pet shops) and human access routes (Copp et al. 2005c; Copp, Vilizzi & Gozlan 2010).

In South Africa, importations associated with the pet trade have resulted in the establishment of wild populations of koi carp *Cyprinus carpio* (Linnaeus, 1758), goldfish *Carassius auratus* (Linnaeus, 1758), the vermiculated sailfin *Pterygoplichthys disjunctivus* (Weber, 1991,) guppy *Poecilia reticulata* (Peters, 1859), swordtail *X. helleri* and *platy Xiphophorus maculatus* (Günther, 1866). Research has demonstrated that *P. disjunctivus* has established in the wild (Jones et al. 2013) and has the potential of competition for basal resources (Hill et al. 2015), although impacts on the recipient aquatic community are yet to be evaluated. Our assessment classified this species as having a high risk of becoming invasive and there is a distinct possibility that other ornamental fish could pose an equivalent risk.

There is no easy way to evaluate or mitigate against the risk of future invasions into South African waters emanating through the ornamental fish trade, where thousands of fish species are traded globally. McDowall (2004) highlighted that difficulties with the vast number of species traded include (1) poor taxonomic and/or ecological data for the species; (2) challenging identification because of similarities between species that is compounded by inadequate and/or difficult to access descriptions (particularly for juveniles which lack the diagnostic characters needed for identification); (3) undescribed species from the speciose American, Asian and African faunas; and (4) multiple origins of imported fish from both the wild and aquaculture facilities result in hybrids and/or specially selected colour varieties. Van der Walt et al. (in press) conducted a DNA barcoding study in 2012 of 187 ornamental fish species from pet stores in Gauteng, Cape Town and Durban finding poor alignment between the trade names and the species names, mismatches between the trade name and the species name and about a third of the species being unidentifiable. Until 2014, ornamental fish imports into South Africa were controlled using a 'Permitted List' containing > 1400 taxa. A 'Prohibited List' of alien fish species whose import is prohibited was implemented and has been developed for NEM:BA (Republic of South Africa 2014). To date, neither the former Permitted nor the NEM:BA Prohibited species lists have been formally evaluated through any form of risk assessment. Therefore, a formal evaluation, including an invasion risk assessment, for the commonly traded and permitted aquarium fish species is urgently required. Public awareness and education of ornamental fish hobbyists, via

pet shops and websites (e.g. SA Pet Traders Association), is required to strongly discourage the release of aquarium fishes into inland waters. This is especially relevant because the evaluation made by Ellender and Weyl (2014) regarding the introductions into the wild demonstrated a continuous trickle of introductions and establishment of species that are distributed via the pet trade, highlighting the need to formalise the disposal of unwanted pets at a national level. Because of the nature of the data required for a FISK assessment, we recommend that an initial screening of the prohibited species list (and the whitelist) be conducted using a rapid screening method, for example, Hoff (2014), to flag potential medium- and high risk species, which can then be assessed using the FISK.

Conclusion

The current legislation on invasive species in South Africa, NEM:BA (Republic of South Africa 2004) and its associated regulations and notices (Republic of South Africa 2014), recognises the conflicting conservation and economic interests associated with alien species, including invasive fishes, and makes provision for their utilisation through a permitting system that allows possession, sale and release into the wild of selected species subject to certain conditions. Although there is little doubt among conservation practitioners that alien fishes require management, it is recognised that, at a country level, there is significant economic benefit derived from the use of alien fishes in fisheries and aquaculture (see Ellender et al. 2014; Woodford et al. 2016). The intention of national legislation is to prevent the secondary spread of invasive alien fishes into areas where they could establish, which has been shown to be significant for even those established species (Ellender et al. 2014), for example, rainbow trout Onchorhynchus mykiss (Walbaum 1792).

This analysis of FISK has demonstrated its utility for the transparent and equitable assessment of species proposed for importation into South Africa for the establishment of aquaculture or fisheries. Given that there are multiple climatic regions within the country from subtropical to cool temperate, it could be argued that a FISK should be performed for each Köppen-Geiger zone within South Africa. However, control of the secondary spread after a species has established in the wild is unlikely and every water body in the country could become a target for illegal fish releases. Therefore, implementation of a broad-based assessment that covers all potential release sites and climate types may be more useful for mitigation against the arrival of future invaders. Should finer resolution assessments be required, a climate match tool, such as CLIMATCH (Australian Department of Agriculture 2010), could be used as a screening tool and to improve the predictive power of the FISK. For the large number of fish being imported for the pet trade, we recommend a rapid screening assessment be conducted for species on the Permitted List, with a full FISK assessment conducted on those flagged as potentially high risk species thereafter.

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Competing interests

The authors declare that they have no financial or personal relationship(s) that may have inappropriately influenced them in writing this article.

Authors' contributions

B.R.E. and O.L.F.W. conceptualised the study. B.R.E., M.E.A., S.M.M., R.J.W., O.L.F.W. and D.J.W. compiled the FISK risk assessments. S.M.M. conducted the statistical analysis and compiled the results. O.L.F.W., S.M.M. and D.J.W. led the writing of the manuscript, with all authors contributing to the text.

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