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Policy recommendations for the *Rana* trade towards the Republic of Korea

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- Ban the trade of non-native species for consumption as food or derived products and for personal use (i.e., as pets).
- Ban the trade of native species when they do not originate from within the nation (i.e., same genetically defined conservation unit).
- Tracking of potential established alien Rana populations.
- Eradication of potentially established alien Rana populations.

KEYWORDS

trade, Rana sp., alien species, Republic of Korea, northeast Asia, brown frog

1 Introduction

1.1 Species introduction

Wildlife trade is responsible for the introduction of numerous species into new environments globally (Souviron-Priego et al., 2018), including amphibians. Invasive species generally have negative impacts on local species through predation, competition (Mori et al., 2015; Sarashina and Yoshida, 2015) and other ecological interactions (Buckley and Catford, 2016), including carrying non-native pathogens (Bezerra-Santos et al., 2021). This is also the case in Ranidae species, such as *Pelophylax ridibundus* in the UK (Zeisset and

Beebee, 2003), and several species in the focal area of this study, including *Rana huanrenensis* that has been introduced to offshore islands in the Republic of Korea (Bae et al., 2022).

The amphibian trade towards the Republic of Korea includes live animals both for the pet trade (online and shops; Koo et al., 2020) and for human consumption (shops and markets; Othman et al., 2022). Trade principally originates from the People's Republic of China (hereafter China; 97% of weight) and the USA (0.94% of weight; Jo et al., 2022). Amphibian imports and sales have increased from 0.2 ton in 2002, to 11.8 tons in 2016, followed by a sharp increase over the last 5 years to reach 37.7 tons in 2021 (Korea Service; https://unipass.customs.go.kr/ets/index.do? Customs menuId=ETS_MNU_00000103). As a result, some amphibian species have been designated as Alert Alien Species (Notification of the Ministry of Environment, No. 2020-79). One of the most widely introduced amphibian species in the world, the American bullfrog (Lithobates catesbeianus; Luque et al., 2014) has become invasive in the Republic of Korea as a result of the trade for human consumption, starting in the 1970s (Groffen et al., 2019). The invasive population of American bullfrog in the Republic of Korea has resulted in a severe loss of aquatic biodiversity (Son et al., 2021), and numerous removal efforts have failed, indicating that eradication is extremely complicated and expensive to achieve (Oh and Hong, 2007; Groffen et al., 2019; Soto et al., 2022), although not impossible (e.g., Simberloff, 2003; Kahrs, 2006; Kamoroff et al., 2020). Therefore, the most efficient and cost-effective approach to prevent the establishment of species known to have the potential of becoming invasive is by prohibiting initial introduction (Othman et al., 2022).

The Republic of Korea imports live Brown frog (Rana spp.) individuals from China for human consumption (Othman et al., 2022). This trade is conducted under a legal umbrella as the species can be exported from China (http://www.gov.cn/zhengce/2020-12/ 27/content_5573532.htm; see acknowledgements for an English version), and species that are native to the Republic of Korea can be legally traded, including *R. uenoi*, *R. huanrenensis* and *R. coreana*. Two of these species, R. huanrenensis and R. coreana, also occur in China, and are present in the wildlife trade. The last species, R. uenoi, is endemic to the Korean Peninsula and Tsushima Island in Japan. Only described in 2014, R. uenoi has been split from the R. dybowskii species complex (Matsui, 2014). It was however previously treated as R. dybowskii, and was therefore assessed as occurring throughout the Korean Peninsula, northeast China, and the Primorsky region in Russia. However, the two clades are differentiated at the species level following multiple lines of evidence, and all individuals belonging to this species complex in the Republic of Korea are finally recognised as R. uenoi (Jeon et al., 2021). As a result, the legality of trade towards the Republic of Korea between 2014 and 2021 was a point of contention.

A recent study has demonstrated that the ranid species imported to the Republic of Korea do not only include legally traded species, but also some species that are not native to the nation but morphologically similar: *R. amurensis*, *R. chensinensis*, *R. dybowskii*, *R. kukunoris* and *R. taihangensis* (Othman et al., 2022; Shen et al., 2022). In addition, these species have not been explicitly listed as banned from the trade by the Ministry of Environment due to the absence of prior data on the potential for invasion, and fall under a broader legislation regarding the general trade of species. In addition to likely escapes from sellers, which could start the invasion process, communication with traders in the Republic of Korea revealed that at the end of the legal sale period, unsold individuals were released in nearby streams, reportedly for welfare reasons, but perhaps also because of costs or methods to keep them, or because of reduced market value. While individually these releases can seem ethical, they are biosecurity threats as the frogs are not scanned for pathogens. Both African Swine Fever and Avian influenza related pandemics are valuable examples of how quickly pathogens can spread.

Following the development of international trade, updated regulations guiding such exchanges are required, and sciencebased recommendations have the potential to lead the development of regulation to prevent the loss of biodiversity (Pullin et al., 2009). Thus, policy recommendations have the potential to help update national laws, especially in this context. For instance, a policy recommendation on the trade of invasive American bullfrogs towards the Republic of Korea (Borzée et al., 2020) was temporally coincidental with a regulatory update in the trade of amphibians (Korean Law Information Center, 2021). However, new discoveries such as the presence of non-native *Rana* species in the trade, calls for specific updates in regulations, resulting in this policy recommendation and its potential use for further legal adjustments in the trade of the genus.

1.2 Risks of invasion

The trade of amphibians and the resulting invasive species can pose two major ecological and biosecurity threats for the survival of native species. The first one being the impact on native species through ecological interactions, and the second being through pathogen dispersion (Kraus, 2015; Falaschi et al., 2020; Green et al., 2020). Both of these impacts have already been documented from the invasive American bullfrog (*L. catesbeianus*) in the Republic of Korea, impacting the ecology of native amphibians, reptiles and birds (reviewed in Groffen et al., 2019), and also increasing the pathogen loads on native species (Borzée et al., 2017).

Many Rana species have overlapping morphological and ecological traits, and are therefore more likely to occupy similar niches. For example, two species pairs, R. coreana plus R. amurensis, and R. dybowskii plus R. uenoi, have similar ecological requirements within pairs. The impact of the interactions between these species pairs is however unknown as the contact zones are in the Democratic People's Republic of Korea (Borzée et al., 2021). In addition, even within a single species, the ecological requirements of geographically distant clades can be significantly divergent (Stockman and Bond, 2007; Podnar et al., 2014), and the displacement of individuals, and the potentially resulting hybridisation, of the clades can result in the inadequate adaptation to the environment and a decrease in fitness (Parris, 2000) or resistance to pathogens (Parris, 2004). Therefore, the introduction of any of the non-native Rana species, and their establishment in the wild in the Republic of Korea could result in competition and hybridisation with the native species, driving the extirpation of the native Rana. The negative effect of displacements can also be within a single species, such as in R. huanrenensis, as the

species ranges from Liaoning in China in the north to the southern edge of the Korean Peninsula in the south, resulting in a latitudinal variation known to be linked to diverging ecological requirements (Andersen et al., 2022). The introduction of individuals from a northern latitude to the south, and any resultant hybridisation, would result in individuals less adequately adapted to the local environment and could even result in species losses. In addition, hybridisation can magnify the invasive capacity of a species (Coulter et al., 2020).

Two major pathogens that are transferred by Rana species include Ranavirus (family Iridoviridae; Kwon et al., 2017) and Batrachochytrium dendrobatidis (Bd; Bataille et al., 2013). Ranavirus is found in both captive and wild populations in the Republic of Korea, resulting in mortality events in both settings (Kwon et al., 2017; Park et al., 2017). Intriguingly, the mass mortality events in the wild occurred in the area where the non-native Rana species focal to this study had been released (although currently not known to have been involved), and even if no individual was intentionally released, all farms provide a substantial risk of individuals escaping over time. In addition, Ranavirus prevalence is known to be higher in invasive ranids in the Republic of Korea (Roh et al., 2022). While Bd is not known to have resulted in mass mortality events in the Republic of Korea, Rana species can be reservoirs for the pathogen (Bataille et al., 2013; Fong et al., 2015), and the introduction of non-native Bd strains could have deadly effects. Therefore, the introduction of Rana species across natural boundaries can have disastrous consequences for local populations, including local extirpation (e.g., Borzée et al., 2017).

To address some of these threats in compliance with the legal obligations established by the Convention on Biological Diversity (CBD), the Republic of Korea's Fourth National Biodiversity Strategy (2019-2023; Nature Conservation Bureau, 2018: https://www.cbd.int/doc/world/kr/kr-nbsap-v4-en.pdf) contains a number of action plans that aim at managing threats to biodiversity, among others, by establishing mechanisms to control human-mediated species introduction, strengthening policy responses and post-introduction control of invasive species. Biodiversity conservation through the protection of endangered and endemic species is also one of the targets established by the National Biodiversity Strategy of the Republic of Korea and other regulations with a special focus on strengthening research on and response to wildlife diseases, along with an improved wildlife rescue and care system.

2 Policy options and implications

Current laws in the Republic of Korea allow for the import and captive breeding of a number of *Rana* species such as *R. huanrenensis*, *R. dybowskii* and *R. coreana* as long as authorities have issued a permit. While the trade of native species is legal, further measures need to be implemented to avoid the introduction and establishment of non-native species with serious impacts for the native population of *Rana*. Recent amendments to the legislations will designate amphibians as aquatic organisms managed by the Ministry of Oceans and Fisheries as of 2023, and treated as fisheries livestock in this regard, and therefore subjected to the same regulation as fishery products (Ministry of Oceans and Fisheries

Ordinance N. 543, 2022.4. 29. Partial amendment). Trade bans have already been declared for certain species in the Republic of Korea, and the regulation can be built upon (Kang and Phipps, 2003).

3 Actionable recommendations

To ensure that new alien populations of *Rana* will not be established in the Republic of Korea, we recommend banning the trade of ecosystem disturbing non-native species or specimens of the native species that are from different genetically defined conservation units (following for instance Othman et al., 2022). "Trade" refers also to operations of import or export and includes notably the trade for human consumption and its derivatives and trade for personal use. To allow for this, authorities could conduct a risk assessment to decide whether alien *Rana* species are 'ecosystem disturbing species'. This measure is supported by the scientific literature referenced in this manuscript and by national legislation (Republic of Korea's Act N. 11257 of 2012 on the Conservation and Use of Biological Diversity Arts. 21 to 24 "The Biological Diversity Act").

This ban should also apply to domestic trade, with individuals of *R. uenoi* from Jeju Island not being traded on the mainland, as this population belongs to a different conservation unit (Jeon et al., 2021). This could be done by designating Jeju Island as a special protection district (Act N. 10977 of 2011 on Wildlife Protection and Management, Article 27).

One exception could be applied to this ban: when the trade of these species is not impacting the conservation of said species in exporting regions (*R. huanrenensis* and *R. coreana*), the trade should consist solely of dead specimens to ensure the absence of escapes, and release of pathogens.

Through trade, numerous individuals have been imported, released, or have escaped. As a result, the presence of established alien populations is not impossible. To avoid further disturbances and reducing the potential for economic costs associated with the establishment of an invasive population, the release of any alien specimen and its offspring originating from the past trade should also be banned as far as possible. While the banning measures are decided, we recommend authorities to use the competences granted in Art. 14.2 of the Biological Diversity Act and adopt emergency measures to prevent the risk of depletion or disappearance. In this sense, we recommend that such populations should be tracked, and controlled (captured and removed, or re-exported if the population of origin can be identified and the individuals can be certified to be free of pathogens), before it becomes too late and alien species are not manageable.

In addition, even though released and escaped frogs may not have established populations, they may have spread pathogens, infecting native populations. Thus, broad scale surveys for ranavirus and chytrid fungus (*B. dendrobatidis* and *Batrachochytrium salamandrivorans*) should be conducted around the establishments selling frogs traded from abroad, and a plan for the control of pathogens should be established.

Lastly, we recommend the establishment of an updated National Species List of native species that includes *R. uenoi* along with keys for species identification, which can be useful at borders and custom

TABLE 1 Identification key and comparison of 40 morphological characters between eight species of northeastern Asian brown frogs (genus *Rana*) recorded from the literatures and online databases of amphibians: AmphibiaWeb (https://amphibiaweb.org) and AmphibiaChina (https://www.amphibiachina.org/). The boxes coloured in grey and marked with symbol (+) indicate the presence of the character in the species and the colourless boxes marked with symbol (-) indicate the absence of the character in the species, OR, the absence of definitive information but matching with at least a few individuals of the species. The symbol (?) indicate the lack of information about the specific morphological characters and classifications of *Rana* into clades I, II and III in the table are adapted from the operational taxonomic units (OTU) based on the 16S rRNA phylogeny of Othman et al. (2022), wher *R. chensinensis* matches with OTU 7 and *R. taihangensis* matches with OTU 1 and 2, following Shen et al. (2022).

			with 010 7 and R. ta	Clade II		J					
		Cla	de l	Clad	e ll	Clade III					
Body part	Key morphological character	Rana coreana	Rana amurensis	Rana dybowskii		Rana chensinensis	Rana kukunoris	Rana huanrenensis	Rana taihangensis		Type of morphological character
Skin	Two dorsal stripes with tubercles within each	-	+	-	-	+	+	-	+	Song et al. (2006); Wang	Meristic
	black spot	-	Dorsal stripes Tubercles	-	-		T cm	-		et al. (2017); Shen et al. (2022)	
	Two dorsal stripes with visible black spots	-	-	+	+	+	-	-	+	Wang et al. (2017)	Meristic
	(somatime), without tubercles	-	-			-	-	-	-		
	Two dorsal stripes with smooth skin and black	+	-	-	-	-	-	+	-	Song et al. (2006);	Meristic
	spots, but without tuberele(s)		-	-	-	-	-	-	-	AmphilaChina (2022)	

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		Clade I		Clade II		Clade III					
Body part	Key morphological character	Rana coreana	Rana amurensis	Rana dybowskii		Rana chensinensis		Rana huanrenensis	Rana taihangensis		Type of morphologica character
	Two viable donslateral folds, with the presence of weakly marked tuberdes on the sides of donal region	-	-	-	-	-	-	-	-	Song et al. (2006); Matsui (2014)	Meritic
	Rough, Red-brown spots on flanks	-	·	-	-	-	-	-	-	Song et al. (2006)	Meristic
	Reddish body with presence of a few black spots between scapular and sacral region	-	-	-	•	-	-	-	-	Matsui (2014)	Meristic
	Reddish-brown dorsal head	-	-	-				-		Matsui (2014)	Meristic

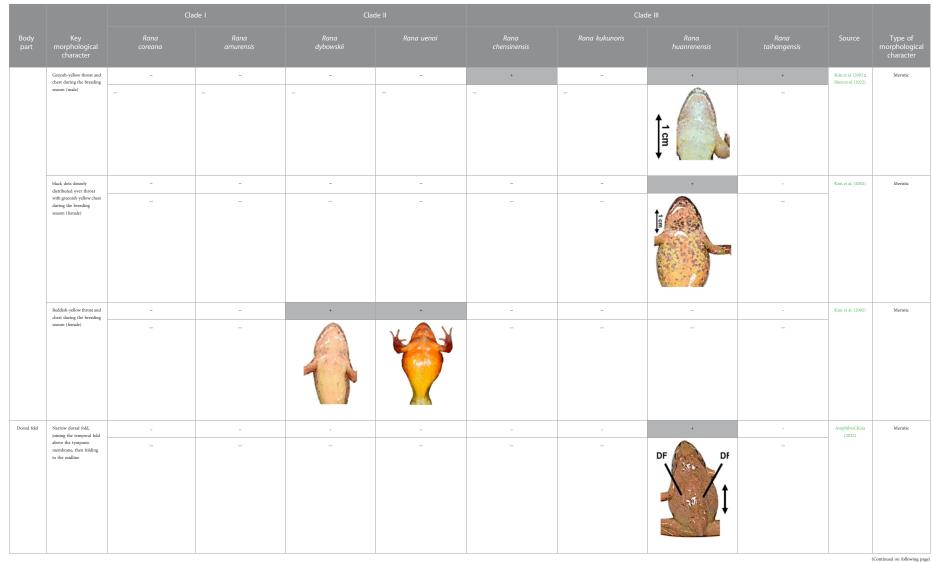
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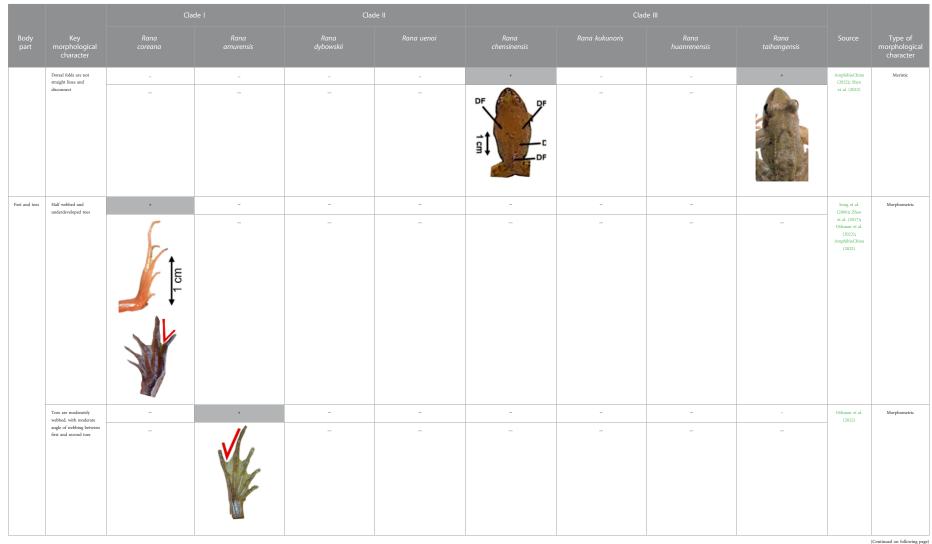
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			de I		de II		Clad	le III			
Body part	Key morphological character	Rana coreana	Rana amurensis	Rana dybowskii		Rana chensinensis	Rana kukunoris	Rana huanrenensis	Rana taihangensis		Type of morphological character
	Tubercles on ventral side of thigh	·		-	-	·				Song et al. (2006); Amphibiat.China (2022); Shen et al. (2022)	Meriatic
	Smooth ventral skin			•						Matsui (2014)	Meriatic
	Smooth ventral skin, and milky-white throat and chest during the breeding season (male)	-	-			-	-	-	-	Kim et al. (2002)	Meriatic

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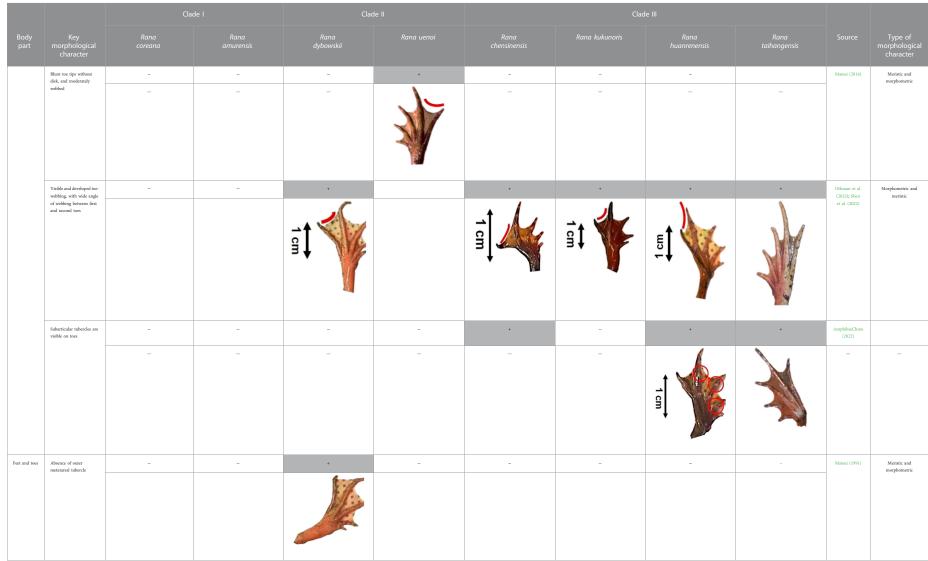




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	Key morphological character	Clade I		Clade II		Clade III					
Body part		Rana coreana	Rana amurensis	Rana dybowskii		Rana chensinensis	Rana kukunoris	Rana huanrenensis	Rana taihangensis		Type of morphological character
	Webbing of second, third and fourth toes are just	_	_	-	-	+	_	-	-	AmphibiaChina (2022); Shen	Meristic and Morphometric
	slightly exceeding the line connecting the second joints of each toe									et al. (2022)	
	Presence of four or less than four dark bars on	-	-	+	+	-	-	-	?	Matsui (1991)	Meristic
	uha	-	-	Dark bars		_	_	_	-		
	Female tibiotarsal articulation of adpressed limb reaching the point between anterior corner of eye and nostril	-	-	-	+		_	-	2	Matsui (2014)	Morphometric
	Male tibiotarsal articulation reaching the point between nostril and tip of snout	-	-	-	+	-	-	-	?		
	Female tibiotarsal articulation reaching the anterior corner of eye	-	-	·	-	-	-	-	ę		
	Male tibiotarsal articulation joint reaching the point between anterior corner of eye and nostril	-	-	+	-	-	-		?		
	Male tibiotarsal articulation joint reaching the tip of the snout	-	-	+	-	+	-	-	?	Matsui (1991); Wang et al. (2017)	Morphometric
	Female tibiotarsal articulation joint is not reaching nostril	_	-	-	-	+	-	-	ş	Matsui et al. (1993); Wang et al. (2017)	Morphometric



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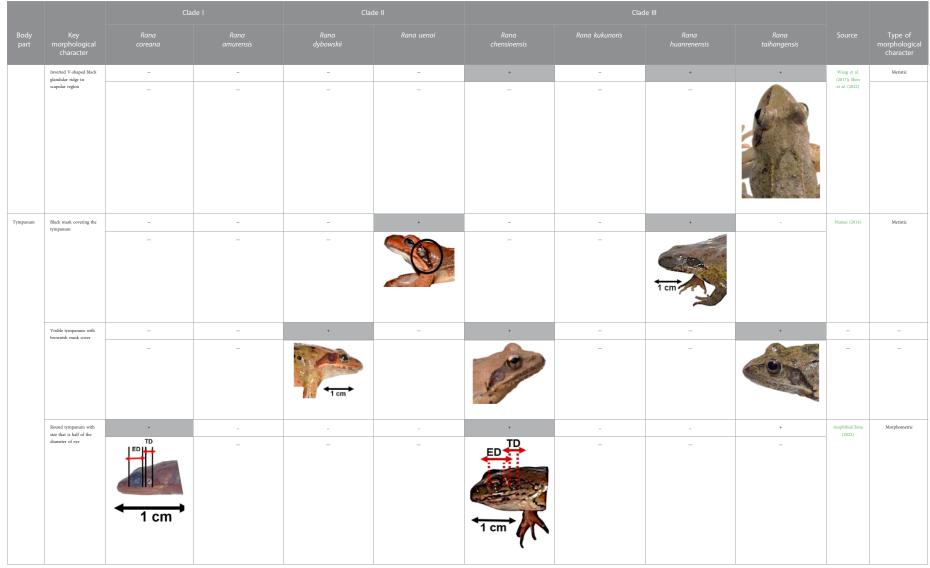
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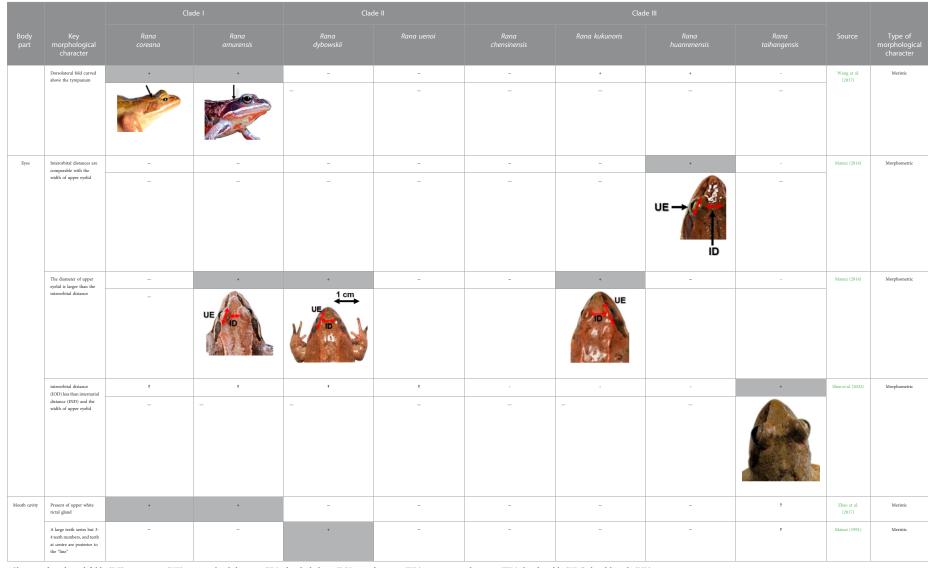
		Clade I		Clade II		Clade III					
Body part	Key morphological character	Rana coreana	Rana amurensis	Rana dybowskii		Rana chensinensis	Rana kukunoris	Rana huanrenensis	Rana taihangensis		Type of morphological character
	Blunt and round finger's end, but with thin distal	+	-	-	-	-	-	-	-	AmphibiaChina (2022)	Meristic
	phalanx	P	-	_	-	-	_	-	-		
	Lengthy hind limb (sum of thigh, tibia tarsus and		_	-	+	-	+	-	-	Leung et al. (2021); Shen	Morphometric
	or ungo, door area and foot length)	_	_	_	î at	-		-	_	(001), 5000 et al. (2022)	
Head	A continuous white line	+	+			-	_	-	-	AmphibiaWeb	Meristic
	along the upper lip	White line		-	-	-	-	-	-	(2022)	
	Flat and slender head (head width = head length)	+	+	-	-	-	-	-	-	AmphibiaChina (2022)	Meristic
	length)	+		_	-	-	_	-	-		

(Continued on following page)

	Key morphological character	Clade I		Clade II		Clade III					
Body part		Rana coreana	Rana amurensis	Rana dybowskii		Rana chensinensis	Rana kukunoris	Rana huanrenensis	Rana taihangensis		Type of morphological character
	Flat, wide and short head (head width > head	-	-	+	-	-	+	+		AmphibiaChina (2022)	Morphometric
	(ength)	_	-	1 cm	_	_	en e		_		
	Flat, wide and lengthy head (head length > head	?	ż	?	ş	+	?	?	?	Matsui et al. (1993);	Morphometric
	width) and sometimes elongated but with stretched head								-	AmphibaChina (2022)	
	Visible internal vocal sacs (male)	-	-	+	+	-	-	-	-	Kim et al. (2002); Matsui (2014)	Meristic
	Lack of vocal sacs (male)	+	+	-	-	-	-	+	-	Kim et al. (2002)	Meristic



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*Glossary: dorsolateral folds (DF), upper eye (UE), inter-orbital distance (ID), distal phalanx (DP); eye diameter (ED); tympanum diameter (TD), head width (HW), head length (HL). = = = = = = = =

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offices, and especially for the Korea Customs Service, which is in charge of controlling the trade (see identification key in Table 1).

4 Conclusion

Several non-native Rana species have been traded towards the Republic of Korea over the past decades. The trade was conducted legally. However, trade followed regulations that failed to incorporate current advances in taxonomy. Although the establishment of alien Rana populations in the Republic of Korea has not been confirmed, factors which could potentially result in such establishments have been brought together. It is therefore important to monitor Rana populations with molecular tools around establishments selling frogs to clarify their native status, as well as to monitor the presence of pathogens. In addition, to avoid the establishment of such alien populations and associated pathogens, it is important to ban the trade of non-native species entirely for human consumption. If the trade of native species has to be maintained, it should be limited to processed products, linked to verifiable data informing on the origin of the animals and results of disease screening, so that the risk of establishment of alien populations is nullified. The species of principal focus are R. dybowskii, R. amurensis, R. chensinensis, R. taihangensis and R. kukunoris, although further analyses to provide a clearer definition of taxonomy and conservation units are also needed.

Author contributions

The ideas were developed, and the manuscript drafted by AB, MR, NB and SO. All other authors provided constructive feedback and revised the manuscript.

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Conflict of interest

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