Biogeography and hotspots of amphibian species of China: Implications to reserve selection and conservation

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Studies on species' biogeography, biodiversity are of remarkable significance in biogeographic regionalization and systematic conservation. In this article, we employed various numerical approaches to investigate distributions of amphibian species of China at different geographic scales. Distributional information of 224 amphibian species was first accumulated to build a database. The database was then analysed by (i) Geographical Information System for richness mapping; (ii) cluster analysis and parsimony analysis of endemicity for biogeographic regionalization and for the delimitation of areas with rich endemicity; (iii) complementarity analysis rank the quadrates for nature reserve selection and (iv) species potential distribution modelling for revealing species potential distribution boundaries. Our results indicated that most species were restricted to a latitudinal region between 27-30°N and about 31% of the species belonged to vulnerable threat category. Hotspots for species diversity were mainly detected in most areas of Sichuan and Yunnan. Seven zoogeographic regions and six endemic areas were identified. Compared with biogeographic regions of reptiles, mammals and plants, some regions were found overlapping among biogeographic regions of different species. Complementarity rarity-based analysis carried out 37 optimal grid cells that could represent the 152 selected species based on rarity values. The current potential distributions of 224 species based on Domain and GARP models had similarities as well as divergent aspects. Some shifts existed between the current and future distribution ranges based on Domain model. Our study represented the first attempt for a combined quantitative analysis for amphibian species distribution in China.

Keywords: Biogeography, biodiversity, parsimony analysis of endemicity, amphibians, geographical information system.

THE loss of biodiversity has become an issue of global concern. Amphibians, one of the most susceptible groups of animals, are representative of the loss of population and species¹. Since 1980s, there have been increasing reports about the decline and extinction of amphibian populations

worldwide^{2,3}. The World Conservation Union's (IUCN) 2001 Red List⁴ of Threatened Amphibians suggests that more than one-third of the species are threatened globally. It is reported that during 2004, about 168 amphibian species were extinct (www.geo.de). Several studies have revealed that⁵⁻⁷ amphibian groups show specific composition in any given geographical unit. Information on these rare species are highly useful in reconstructing biogeographical history of a region^{6,8,9}. In this respect, studies on amphibian species are significant¹⁰⁻¹². China is one of the richest areas in the world in terms of amphibian species (herein denoting toads and frogs) and is a confluence of two main zoogeographic realms, viz. Palaearctic Realm and Oriental Realm^{13,14}. More than 300 amphibian species have been recorded in China. Among them, many are endemic to China. One feature of these species is the narrow distributional range they have in China, including Taiwan province^{6,15}. Geographical Information System (GIS) is of important use in biogeography as it provides a visual representation of species distribution and habitat status¹⁶. Ecological niche modelling is being increasingly adopted to study the potential distribution of a species based on climatological data inputs. It is one of the best tools currently available for assessing the effects of climate changes on the distributions of species¹⁷. Associated mathematical techniques contain two categories¹⁸. Parsimony Analysis of Endemicity (PAE) is a technique to identify areas of endemism designated by species with convergent biogeographical history^{19,20}. The PAE uses the parsimony algorithm to obtain an area cladogram. It begins by compiling presence/absence information for taxa at sample localities, and then groups the localities into a cladogram²¹. The PAE has been used widely in many biogeographical studies in recent years^{22,23}. Multivariate analyses are popularly applied in biogeography and ecology. Herein we use cluster analysis for endemic amphibian regionalization at the level of provinces, which are geographical units. We calculate Euclidean distance with Ward's algorithm in cluster analysis, and the dendrogram used is showed here.

Perhaps our study is the first attempt for a combined analysis for amphibian species distribution in China. The PAE and cluster analysis procedures provide useful information for identifying areas of endemism/hotspots that may have been overlooked formerly. The GIS procedure and

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species projection modelling provide useful information for strategic conservation planning.

Data gathering and approaches for the analysis

We gathered information on 224 amphibian species to compile a database which was derived from an exhaustive review of earlier published literature and websites. On every species we recorded the following data: scientific name, common name, conservation status, distributional points and related coordinate. All the data were obtained from the following websites and literature: China Species Information Service (CSIS) (http://www.chinabiodiversity. com), Global amphibian assessment (http://www.globalamphi bians.org/), Amphibian Species in the World (http:// research.amnh.org/herpetology/amphibia/index.php); China Red Data Book of Endangered Animals – Amphibia and Reptilia²⁴, Identification Handbook of Herpetology in China²⁵, Systematic Searches of Herpetological Species of China²⁶, Acta Hebpetolocica Finica²⁷, An Illustrated Key to Chinese Amphibians²⁸, Atlas of Amphibians of China²⁹ and Rare and Economic Amphibians of China³⁰. All the data were listed in a spreadsheet running to over 2500 records.

The database was then submitted to GIS for spatial analysis such that current distributional hotspots were identified and potential distribution of species in China were projected. Here we employed the general software Arc-View 3.3 and DIVA-GIS³¹ (downloaded free from the website: <u>http://www.diva-gis.org</u>). Most of the computational procedures we employed were based on these software.

We have adopted a method based on ecological niche modelling to detect significant hotspots for conservation planning. We used a batch of species for calculation: mapping the potential distribution as a whole with all the records. The digitized maps obtained could be interpreted as the suitable sites for all the species. At the meantime, we used CCM3 climatic modelling results to predict the shifts in distributional range of all the 224 species under further predicted climatic condition. (http://www.cgd.ucar.edu/cms/ccm3, http://eed.llnl.gov/cccm/hiresolu.html). We manipulated the projection procedure by adopting Domain model^{32,33} and Genetic Algorithm Rule-set Process model^{34,35}.

Software employed were DIVA-GIS and Desktop GARP³⁴ respectively. The default climatic variables inbuilt in these two software were used. In all 19 variables including annual mean temperature, isothermality, annual precipitation and precipitation seasonality were used in DIVA-GIS. In Desktop GARP, there were 14 variables, including topography, slope, etc. The maps produced are combined with Arcview 3.3. The WorldClim database (http://biogeo.berkeley.edu/worldclim/worldclim.htm) provided correlative environmental datasets. Base layers of China for our analyses were obtained from National Fundamental Geographic Information System (NFGIS) (http:// nfgis.nsdi.gov.cn).

Winclada^{*36} is used to perform the PAE at different geographical levels. First, we integrated the locations of our species into ranges of 24 provinces. Each species was used as one operational unit in the form of present/absent in each geographical unit. If a species was present in one particular unit, we tagged 1 in the spreadsheet, or else tagged. Thus we compiled a 24×224 matrix which included all information of species in all the provinces and a 232×152 matrix which included 152 species that have identified explicit point distribution records in all the 232 geographical quadrates. Then the two matrices were introduced for PAE and cluster analysis. Cluster analysis³⁷ was carried out by STATISTICA* 6.0.

We also performed complementarity analysis³⁸ for reserve selection using the 232×152 matrix, in order to identify grid cells with a defined size, which complemented each other in terms of species composition. The selection procedure was iterative³⁹. We selected minimum grid cells to represent all the 152 species concerned using Rebelo and Siegfried's rarity algorithm⁴⁰. The selection procedure had the following steps: We first assign each grid cell a rarity value according to the following formula:

Rarity = $\sum k/ai$

where k is the total number of unreserved quadrates and ai is the number of unreserved grid cells containing the ith species. The grid cell with the highest summed rarity value was nominally reserved and removed from the matrix, together with all the species present in that grid cell. The summed rarity value for each of the remaining grid cells was recalculated and the next highest quadrate was continued until all species were represented in the selected quadrates. If more than one site had the same rarity value, we choose the one with higher richness.

To minimize the effects exerted by the possible fact that the amphibian species located in the territory of China had not been widely surveyed and to obtain a more accurate result, we excluded some provinces. Figure 1 shows all the distributional points and quadrates we used in our study.

Results

Latitudinal distributional pattern and IUCN categories of species in China

The interrelationship between species richness and latitude shown in Figure 2 suggests that geographic area from 27 to 30°N lat. was the host range for amphibian species in China. Figure 3 shows the percentage of species of China falling into different threat categories of the IUCN.

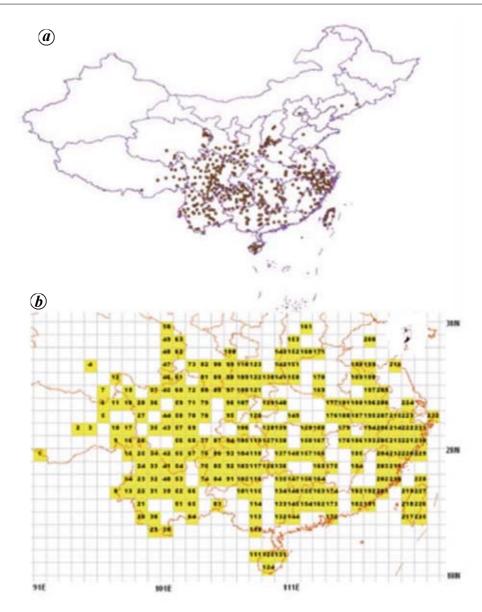


Figure 1. a, All the amphibian species distributional points used in our study and recorded in our database. b, Quadrate geographical units used in our study. In all there are 232 grid cells whose labels were marked.

About 31% of the all species (n = 69) was in the vulnerable category; five species, viz. *Rana wuchuanensis*, *R. minima*, *Scutiger maculatus*, *Oreolalax liangbeiensis* and *Leptolalax ventripunctatus* were in the critically endangered category.

Map of amphibian species hotspots based on richness

Figure 4 depicts the amphibian species richness map. We used the $1^{\circ} \times 1^{\circ}$ grid highlighted in different colours to show the richness. The hotspots (high richness values) could be found in the middle and south of Sichuan Province. The narrow range from north of Guangxi to the south

of Hunan could also be identified. Other regions, such as most of the territory of Yunnan, Fujian, Taiwan, Hainan, Guizhou, Hubei and a narrow area in Xizang also show high level of richness to some extent.

Cluster analysis and PAE

The dendrogram of cluster analysis based on Ward's algorithm is shown in Figure 5. The similarity matrix was calculated based on Euclidean distance. Sichuan and Yunnan were first clustered into a group for their significant abundant species. Hubei, Shananxi and Gansu were classified as another group. Most southeastern and northeast provinces were divided into two groups respectively. The PAE was performed using heuristic search to identify the best trees. At the level of province geographic units, 24×224 matrix (24 geographic units and 224 species) carried out 68 best trees. The length of all trees is 309, consistency index is 0.5955, homoplasy index is 0.4045,

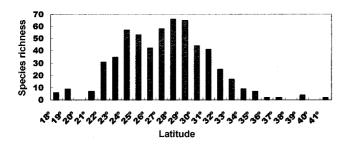


Figure 2. Interrelationship between latitude and species richness. *X*-axis represents the latitude; herein we use the equal latitude interval, for example, 18° means the latitude interval between 18 and 19° , including 19° . *Y*-axis represents the number of species involving in the related interval.

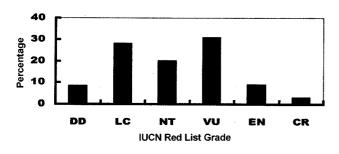


Figure 3. IUCN red list grade composition of amphibian species in China. DD, Date Deficient; LC, Least Concern; NT, Near Threaten; VU, Vulnerable; EN, Endangered; CR, Critically Endangered. Largest percentage of species (n = 69 amounts to 31%) were in the vulnerable category.

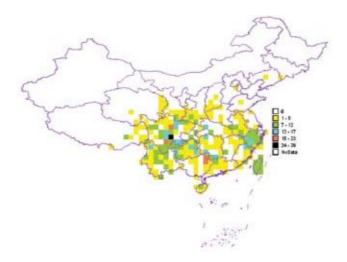


Figure 4. Amphibian species richness map in China. The colours, light to heavy, represent the richness from the minimum to maximum value, while white colour represents no data or no species detected currently. Sichuan, Yunnan, Guizhou, the range from north of Guangxi to middle of Hunan, east areas along sea coast areas (including Zhejian, Fujian, Taiwan) are the locations holding high richness values.

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and retention index is 0.4703. By operating strict consensus of 68 trees, we obtained the strict consensus cladogram shown in Figure 6 *a*. At the level of $1^{\circ} \times 1^{\circ}$ geographical quadrates, 232×152 matrix carried out more than 1000 best trees and the calculation procedure sometimes flowed out.

We used the restriction that 1000 trees hold at each step, and the strict consensus cladogram carried out by Winclada* (Figure 6 *b*) and the 15 groups marked therein have been updated into the geographical map (Figure 5 *c*). We denoted a group as an endemicity region which involves more than three clade branches²².

The parsimony analysis produced consistent assemblages of areas. Nevertheless, the relationships among some clades in our study either in 24 provinces or 232 quadrates were not resolved. One reason for this was that some geographical units had too less records to manipulate using the computer. The consequences from the PAE were different from the cluster analysis. In the dendrogram of cluster analysis, Hubei, Gansu and Shananxi were clustered into a group, while in the cladogram of the PAE at province level, Hubei was a branch of the geographical group, including Hubei, Zhejiang, Fujian, Anhui, etc. Despite the differences, they have similar aspects such as the group involving Hunan, Guangxi, Guizhou can be obtained in both methods. Comparing the two approaches, a PAE cladogram supplied more useful information and could indicate historical sequences of biotic divergence and isolation^{19,20,41}

Biogeographical regionalization for amphibian species of China

Based on cluster analysis and PAE, biogeographic divisions for amphibian species in China can be divided into seven regions (Figure 7).

A, Southwest region: Yunnan, Sichuan, East Xizang. These regions located in the altiplano of lower altitude

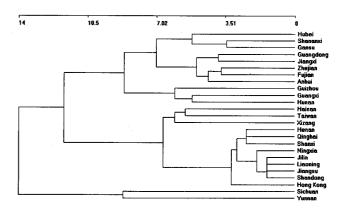


Figure 5. Dendrogram of amphibian species at the level of province geographical units, the cluster algorithm is Ward's algorithm based on Euclidean distance.

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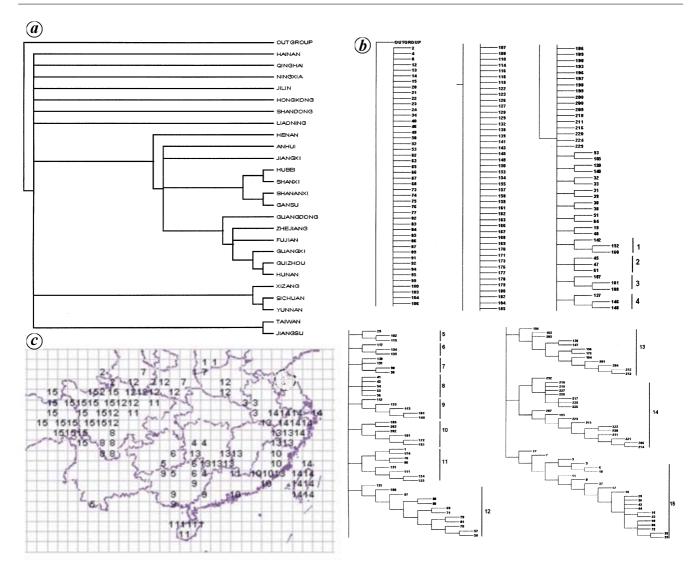


Figure 6. *a*, Strict consensus cladogram of PAE at the level of province geographical units. The tree is 309 steps long, consistency index (Ci) is 0.5955, and retention index (Ri) 0.4703. *b*, Strict consensus cladogram of PAE at the level of $1^{\circ} \times 1^{\circ}$ geographical quadrates. The tree is 804 steps long, Ri = 42, Ci = 18. *c*, Endemic regions grouped by Winclada* heuristic search. Labels marked in the map denote the groups showed in the cladogram of Winclada*.

are characterized by various climatic patterns (including North Torrid Zone, South Sub Torrid Zone, South Temperate Zone, etc.), topographic types and rich precipitation⁴², all of which help gestate the diversified endemic amphibian species. In addition, this region also retains a number of virgin forests, as is regarded as one of the world's species' hotspots. This region can be divided into two sub-regions: (A1), Qinghai–Xizang Plateau sub-region (includes Xizang) and (A2), Yunnan–Guizhou Plateau sub-region (includes Yunnan and Sichuan).

B, Southeast region: Guangdong, Anhui, Jiangsu, Zhejiang, Fujian, Hong Kong and Jiangxi. The features of this region include abundant precipitation and affluence in water resource.

C, Northeast region: Ningxia, Jilin, Liaoning, Heilongjian, Shandong. The region covers most areas of the north

China, and congruent with the region based on mammals¹³, is poor for amphibian species habitat.

D, Middle to South region: Guangxi, Guizhou, Hunan. The species shared among the provinces include *Rana* schmackeri, R. adenopleura, R. margaretae, R. lungshengensis, Polypedates omeimontis, Pachytriton labiatus, Paa shini, etc.

E, Middle to Northwest region: Hubei, Gansu, Shananxi. The climatic conditions of this region are affected by the Qinling Mountains, which stretch from east to west in Shananxi.

F, Hainan Island: This is the second largest island in China, after Taiwan. It has monsoon tropical climate with annual average temperature of 22–26°C and annual rainfall of 1500–2000 mm. Its topography is low on four sides and high at the centre. Because of the geographical isolation from the mainland, it holds a good many amphibian species most of which are endemic. For example, *Rana* former s

fragilis, Philautus menglaensis, R. hainanensis, Pelophryne scalptus, Amolops hainanensis, A. torrentis, etc.

G, Taiwan Island: Taiwan straddles the tropical and subtropical zones where the tropics of cancer and the north tropic cross, and has warm summer and mild winter. The climate is moderated by the warm water of the Kuroshio Current. There are also several native species. For example, *Hynobius sonani*, *Micryletta steinegeri*, *Rhacophorus taipeianus*, *Rana longicrus*, *R. swinhoana*, *Buergeria robusta*, *Rhacophorus arvalis*, *Rhacophorus moltrecht*, etc.

Areas of endemism for amphibian species of China

Endemism could be distinguished from species richness using an appropriate index and mapping of such indices can detect centres of endemism⁴³. Endemic regions are shown in Figure 6 c by integrating the PAE result at the 1° lat. $\times 1^{\circ}$ long. quadrate level. The following six regions could be identified as endemic: Taiwan Island; Hainan Island; Southwest region: Sichuan and Yunnan; East region: Fujian, Zhejiang and Anhui; North region: Henan, Hubei, Shanxi and South region: Guizhou, Hunan and Guangxi. Some of these regions had been identified in previous work^{6,13}. The most endemic regions were: Southwest of China, the richest area for amphibian species; and Hainan and Taiwan Islands. Long periods of separation experienced by endemic species inhabiting these two islands resulted in their high endemic values. The narrow range from Guangxi to Hunan, Eastern Xizang also could be

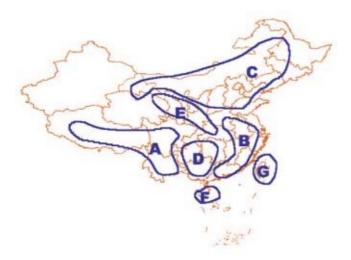


Figure 7. Regionalization of amphibian species in China based on cluster analysis and PAE. A, Southwest Region: Yunnan, Sichuan, Xizang; B, Southeast Region: Guangdong, Anhui, Jiangsu, Zhejian, Fujian, Hong Kong, Jiangxi; C, Northeast Region: Ningxia, Jinlin, Liaoning, Heilongjian, Shangdong; D, Middle to East Region: Guangxi, Guizhou, Hunan; E, Middle to Northwest Region: Hubei, Gansu, Shananxi; F, Hainan Island and G, Taiwan Island.

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recognized as endemic areas, but had been overlooked in former studies^{6,13,15}.

Reserve selection by complementarity rarity-based algorithm

Figure 8 shows results of the complementarity analysis. Complementarity analysis investigates the minimum areas for nature reserve selection by protecting maximum number of species. Based on the rarity selection algorithm, we obtained 37 optimal grid cells for representing 152 selected species, occupying 16% of all the 232 quadrates.

Potential distribution ranges based on different models

Figure 9 *a* shows the current potential distribution for all the 224 species, Figure 9 *b* shows their potential distribution map under future predicted climate using CCM3 model. Figure 9 *c* shows the current potential distribution map for all the 224 species based on GARP model. A prevalent feature shared among the three maps was that the most suitable areas were located south and east of China, including Taiwan and Hainan Islands. Northern and western regions were not potential areas for survival of the species. Especially in Figure 9 *c* under GARP model, most areas of Qinghai, Inner-Mongolia and Xinjiang are devoid of amphibian species.

Comparing the two digitized maps based on Domain model we could predict that the suitable ranges would decrease in future climatic conditions, especially in the western and northern regions, where potentially endemic species had almost disappeared. The most suitable regions would decline sharply, such as East Xizang. Based



Figure 8. Map of thirty-seven optimal priority areas derived from complementarity rarity-based selection algorithm.

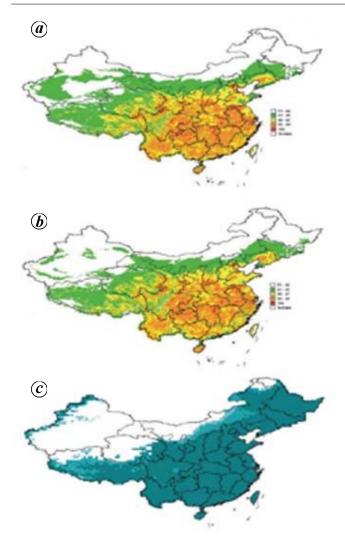


Figure 9. *a*, Current potential distribution map of all 224 amphibian species in China based on Domain model. Colours from green to red represent the suitability degrees of areas from the minimum to maximum value, while white colour represents no data or not suitable areas. *b*, Future potential distribution map of all 224 amphibian species in China based on Domain model under predicted climatic condition using CCM3 model. Colours from green to red represent suitability degrees of areas from the minimum to maximum value, while white colour represents no data or not suitable areas. *c*, Current potential distribution map of all 224 amphibian species in China based on GARP model. It is a combination of 18 layers carried out by Desktop Garp. All the layers are merged by Arcview 3.3. Colours from light to heavy represent suitability degrees of areas from the minimum to maximum value, while white colour represents no data or not suitable areas.

on the results from ecological niche model, we found that suitable ranges of many species would decrease in future though they occupy large regions currently.

Discussion and conclusion

Amphibian species need more attention

Amphibian species are one of the most sensitive groups of animals to climatic changes. It is feared that anthropogenic influence on global climate has increased the rate of extinction of amphibians worldwide. Further, infectious diseases are now recognized as a major cause of decline in populations of amphibians⁴⁴. Many documented data suggest that climate change has indirect influence on the initiation of breeding activities of a few amphibians species.

Regardless of the role that climate change may have played in the past and in the current decline of amphibian populations, future change in global climate will certainly pose challenges for the surviving amphibian populations. It is important to consider the trends in global climate change while planning recovery efforts for endangered sepcies⁴⁵. Understanding distributional range and habitat of amphibian species is necessary for further studies seeking ways to reduce the rates of population decline. Some species exhibited narrow distributional ranges, even just one geographical point.

For instance, Bufo wolongensis only survives in Wolong Nature Reserve, Sichuan; Hyla zhaopingensis only in Zhaoping county, Guangxi; Oreolalax nanjiangensis only in Nanjiang county, Sichuan; Scutiger muliensis only in Muli, Sichuan; Xenophrys daweimontis only in Daweishan Nature Reserve, Hunan; Cynops wolterstorffi only in Kunming City, Yunnan; Theloderma kwangsiense only in Dayaoshan Nature Reserve, Guangxi. These species need immediate attention. Contrarily, some species have wide distributional ranges, and often can be found in many provinces. For instance, Bufo andrewsi has been documented in Yunnan, Sichuan, Hubei, Guangxi, Gansu, Guizhou, Shananxi, Zhejiang, and Qinghai. Rana quadranus is distributed in Shanxi, Shananxi, Hubei, Gansu, Yunnan, Sichuan, Anhui, Henan, and Hunan. However, many of them have suffered from habitat fragmentation and population declines in the recent years²⁴.

Biological invasion has penetrated into the amphibian populations. *Rana catesbeiana* is native to north of America and was introduced into China for its economic value. It is believed that its capacity to survive and its higher reproductive abilities have led to potential decrease in the population sizes of native species²⁴.

Comparison among biogeographic regions of reptiles, mammals, plants and amphibians

The zoogeographic regions of endemic amphibian species include seven regions involving most humid areas of China. We have compared our results with those of other publications and literature about the zoogeographic regionalization of China, and have found some overlap among different animal zoogeographic regions. For instance, regions A, F, G have also been identified by other workers^{6,13,15}. These results may reveal that the species distribution is closely correlated with the environmental and climatic factors.

In our study, East Xizang has significant diversity values where many endemic species occur, and is identified as one hotspot, which is congruent with the observations of Chen *et al.*⁴⁶. They recommended that the Qinghai– Xizang plateau be considered as an independent zoogeographic division equivalent to Palaeoarctic realm and Oriental realm. They elucidated their viewpoint by studying several endemic fishes occurring in the plateau. Based on our analysis of the diversified amphibian species, we support their recommendation.

As shown in Figure 4, most amphibian species are in the southwest and southeast regions of China, which have prolific precipitation and water resources. However, there are no endemic species records in the northwest of China. In other words, distribution range of amphibian species is concentrated in the southwest and southern regions of China. Such distribution reflects a species dispersal in China, i.e. plants/animal species have high diversity values in the South and East and low values in the North and West^{6,47}.

As seen in current and future potential distribution maps of species, we can conclude that the areas occupied by amphibian species will decrease in future. The suitable ranges for species habitation will shift gradually to South and East China, where there is sufficient food and suitable climate. Our observations are in agreement with those on mammals¹⁴.

Hotspots and nature reserve selection for amphibian protection

Hotspots are considered as an easy way to protect biodiversity. Fifty-four hotspots in the world have been described (http://www.biodiversityhotspots.org/xp/Hotspots). Southwest China (mainly indicates Yunnan, Sichuan, Xizang) is recommended as one of them. At a downscaling level, based on complementarity analysis and hotspots, we recommend the following regions as most ones for amphibian biodiversity protection. They are Hainan Island, Taiwan Island, most areas of Sichuan and Yunnan, East Xizang, range from North Guangxi to Middle Hunan, as well as the transitional zone of Anhui, Zhejiang and Fujian. Some of these areas are also hotspots for other vertebrate species and plants⁴⁶. Yunnan and Sichuan are areas that have maximum number of endemic plant species in the world⁴⁷.

Nature reserves are one of the important practical approaches for biodiversity conservation, especially for the scope they offer for *in situ* conservation of natural ecosystems and wild life species⁴⁸.

During recent years, the Chinese Government has attached much importance to the protection of biodiversity, About 799 nature reserves and over 200 centres have been established for the introduction and propagation of animals and plants (<u>http://www.brim.ac.cn/books/cntrysdy_ cn/index.html</u>). There are a number of nature reserves that cover most of the amphibian species hotspots/ endemism areas. For example, in the narrow range from Guangxi to Hunan we find the Dayaoshan Nature Reserve, Huaping Nature Reserve, Manshan Nature Reserve, etc.

Biogeography and biodiversity studies integrating multiple approaches provide new insights

Decline in amphibian populations in relation to global climate change during the past two decades has become a hot topic in science. We have used two models to predict the potential species distribution under the current climate and that predicted under future climatic conditions⁴⁹. The results indicate that the habitat regions for amphibian species would shrink and it is likely that the more suitable regions in the future would shift southwards.

Cluster analysis was employed for zoogeographical delimitation of amphibian species in China⁵⁰. Combining with the consequences of the PAE, seven regions are delimited by cluster analysis at province geographical level. They are southwestern region, northeastern region, middle to northwest region, middle to south region, southeast region, Hainan Island and Taiwan Island. The PAE is used to classify areas of endemism at different levels, but some classified groups are still unresolved. The problem may be solved by adding more distributional records and new statistical techniques⁵¹. The groups classified by PAE at the $1^{\circ} \times 1^{\circ}$ quadrate geographic unit level could be regarded as amphibian species endemism areas.

Mapping species distribution has been broadly studied^{52–54}. As seen in our study, it focuses on current species habitat regions, and attempts to predict their potential distribution. Integrating to GIS, it becomes a powerful tool for biodiversity conservation planning and biological control of exotic species. Our study emphasizes mapping species distribution to interpret the spatial distribution of endemic amphibian species. The distributional maps indicate the biodiversity of these regions in China.

Complementarity analysis³⁹ is a rational nature reserve selection procedure that has been widely used since it can protect maximum amount of species by selecting minimum areas. We employed the rarity-based algorithm to obtain 37 complementary areas for protecting the concerned 152 amphibian species.

Our zoogeographic regionalization, endemism centre delineation and priority area selection are all based on cluster analysis, PAE, biodiversity mapping and complementarity analysis. Combining various methods to investigate biodiversity and biogeography is a trend nowadays^{55,56}. Our systematic investigation on species distribution range by employing multiple analytical techniques has seldom been seen in other publications from China^{6,13}.

Future studies on vertebrate species in China

Future studies should enhance the analytical tools and approaches to evaluate vertebrate species distribution and their

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related information, especially their habitats and physiological conditions in more detailed ways. All the amphibian species located in China, both endemic and non-endemic, will be taken into account. As global climate changes have severely affected species compositions, we will also take other vertebrate species into account in future studies. We have gathered a lot of information on distribution and habitat status of the endemic avian species, endemic mammals and endemic reptiles in China. A database containing all the information about endemic vertebrate species of China will be built up in the future.

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