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Habitat utilization and release-site fidelity of translocated captive-bred giant pandas (*Ailuropoda melanoleuca*)

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Abstract. The behavioural adaptation (movement and habitat utilization) of translocated organisms to a new environment after their release is crucial in translocation programs because it may affect survival. Therefore, identifying the factors determining habitat selection by the relocated animals is essential to improving the planning and the outcome of such programs. Using the data from three relocated giant pandas (*Ailuropoda melanoleuca*), we studied the habitat utilization, release-site fidelity, and interaction with resident giant pandas. The results showed that the quality of habitat used by the relocated giant pandas was significantly higher than the average habitat quality of the research region, and was significantly lower than that used by resident giant pandas. This suggested that the released three giant pandas had habitat selection abilities. The three released giant pandas gradually moved away from their release sites and did not exhibit site fidelity. In the first six months, the giant pandas stayed within about 3 km of their release sites, where habitat was good quality but overlapped with the distribution of resident giant pandas. The overlap of location between released and resident giant pandas decreased after six months when the released giant pandas moved away from their release sites.

Key words: acclimation, interaction, movement, spread

Introduction

Conservation translocations, including reinforcement, reintroduction, and conservation introduction (IUCN 2013), have been widely utilized in protecting endangered species (Robert et al. 2015). It is believed that suitable habitat surrounding release sites can greatly enhance the survival rate of released wildlife (IUCN 1998, Hayward et al. 2007, Osborne & Seddon 2012). However, the released wildlife could still miss suitable habitat if they lack the ability to find or select proper habitat (Letty et al. 2007, Baling et al. 2016). Captive-bred animals, who have never

really experienced "wild" life, are more likely to fail in finding an appropriate habitat than wild-born individuals in a translocation project (Beck et al. 1994, Mathews et al. 2005, McPhee & Carlstead 2010, Harrington et al. 2013). Studies have showed that the quality of habitat used by captive-bred animals after release was generally lower than that of habitat used by wild individuals (Bellis et al. 2004, Swaisgood 2007, Sheean et al. 2012).

Many factors have effects on the habitat utilization of translocated animals (Kajiwara et al. 2016). Experience in a natal habitat might affect the released

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individuals' habitat preferences when they are translocated to unfamiliar habitats (Natal Habitat Preference Induction) (Davis & Stamps 2004, Stamps & Swaisgood 2007). The utilization of habitat by translocated animals could also be affected by the distribution of wild individuals due to intraspecific relationships (Mihoub et al. 2011, Richardson & Ewen 2016). In practice, the release sites are generally located in and surrounded by suitable habitat to increase the survival rate of released individuals (Cheyne 2006).

Release-site fidelity, the tendency to remain within the vicinity of the release site, could ensure the released individuals live in a suitable habitat (Roe et al. 2010, Yott et al. 2011). Many studies showed that released individuals with higher release-site fidelity has a higher survival rate than those with lower release-site fidelity (Tuberville et al. 2005, Terhune et al. 2010, Attum et al. 2013).

The giant panda (Ailuropoda melanoleuca) is a solitary animal that feeds on bamboo. Cubs leave their mothers around the age of 1.5 years and are active around the region of their mothers. At approximately 2.5 years of age, pandas will leave these regions. After being an adult, their home range becomes stable and the same region will be used for many continuous years (Zhang et al. 2014, Liu et al. 2015) or even for their entire life (Hu 2001). The size of the home range of giant panda is usually around 3-6 km² (Hu 2001) but there are age, gender, and seasonal differences in home range size (Song et al. 2006). Even though studies have shown that the size of the home range, activity range, and activity patterns of translocated giant panda individuals are similar to wild giant pandas (Gu et al. 2011, Yang et al. 2018), because of natal habitat preferences and unfamiliarity with the new environment, the movement and spread of translocated giant pandas may be affected.

Habitat fragmentation is the most important threat to the sustainable survival of the giant panda (Pan 1995, Qing 2016). The wild giant panda is distributed in twenty-four habitat patches along six mountain ranges in western China (State Forestry Administration 2015, Swaisgood et al. 2018). In order to reinforce the small populations of giant pandas in wild, the Chinese government has initiated the release of giant pandas into the wild from 2005 onwards. Up until the present, 12 giant pandas (Table S1) have been translocated, of which two were rescued from the wild, and the other 10 were captive-bred pandas that underwent wild adaptation training with their mothers. Currently, there are eight remaining individuals that survived more

than a year; the survival of one panda was unable to be determined as the collar was lost, one panda died, and two recently released individuals have survived more than five weeks.

Although a few previous researches have been performed on the home ranges, activity rhythm, activity range (Gu et al. 2011, Yang et al. 2018), and reproductive behaviours (He et al. 2018) to examine the adaptation process after translocation, the habitat utilization of translocated giant pandas in the early stage after release is still unknown. An analysis of the habitat utilization status of released giant pandas can aid in our understanding of the adaptability of released individuals towards the field environment and improve wild adaptation training and release methods. In this study, we investigated the habitat utilization of translocated captive-bred giant pandas and their spatial interaction with resident giant pandas. This study could provide feedback on the release project for future improvement.

Material and Methods

Study area

The study was conducted in the Liziping National Nature Reserve (LZP) (102°10'33"-102°29'07" E, 28°51'02"-29°08'42" N, area: 478.85 km², elevation: 1330-4550 m a.s.l.) of Xiaoxiangling Mountains, which is located in southwestern China (Fig. 1). The population of giant pandas in Xiaoxiangling Mountains is the smallest giant panda population, whose extinction risk is high due to genetic diversity loss and stochastic fluctuation (Zhu et al. 2010a, b).

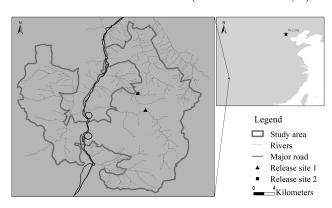


Fig. 1. Study site.

Translocation and monitoring

Eight captive-bred individuals were released at LZP (Table S1). However, one individual, Xuexue, died four weeks after release due to illness. The GPS collar monitoring data of the individuals Zhangmeng, Huayan, Baxi, and Yingxue were less than half a

year and were thus not included in this analysis. In this study, the three individuals, Taotao (TT), Huajiao (HJ), and Zhangxiang (ZX), were all from the China Conservation and Research Center for the giant panda. Before the release of these individuals, they underwent two years of wild adaptation training in nature environment and the training method was led by the cubs' mothers. Of these individuals, the male giant panda, TT (two years old), and female giant panda, HJ (two years old) were directly released at LZP in October 2012 and November 2015, respectively. This release site located in area that is accessible by roads in the nature reserve and the habitat quality is similar to their wild adaptation training area. In addition, the density of wild individuals in this area is relatively small. The female giant panda ZX (two years old) underwent 53 days of adaptation in the wild adaptation area (0.2 km²) in LZP and walked out of the area by herself in November 2013.

After the giant panda was released, a GPS collar was used for tracking and monitoring. The activity site was recorded every two hours. The collar data was downloaded once a week. TT, ZX, and HY were monitored continuously for 30, 28, and 11 months, respectively. The monitoring data for ZX are the data after it had left the wild adaptation area. As we only have three released individuals that were released at different timepoints and sites, the sample size is small and cannot represent all released individuals. Therefore, we consider each released individual as a group for analysis.

Data sources

The distribution of resident giant pandas was determined by feces collected in study areas. A total of 627 giant panda feces samples were collected in the research area during the Forth National Giant Panda Survey (NGPS4) in 2012 and following continuous monitor programs for translocated giant pandas from 2012 to 2017. In all 494 of them were identified and came from 21 wild giant pandas, according to their average length of bamboo stem fragments in feces (Hu 2001), distance to the translocated giant pandas (State Forestry Administration 2006, Sichuan Provincial Forestry Department 2015), and microsatellite DNA analysis (Zhan et al. 2006, Chen et al. 2016). These identified feces did not include any that came from the translocated individuals.

Three categories of environmental factors (physical environmental factors, biological environmental factors and human activity) were generally considered to have important contribution to habitat quality for a giant panda (Wei 1996, Zhang & Hu 2000). The habitat suitability in LZP, therefore, was evaluated using following environmental factors: 1) Terrain, including elevation, slope, curvature, topography position index, aspect and solar radiation index. The elevation was obtained from the scientific database of the Chinese Academy of Sciences (http://www.gscloud.cn) with a resolution of 30 × 30 m Digital Elevation Model (DEM). Other terrain data were derived from the DEM data. 2) Land cover: data was obtained from the Second National Forest Inventory and revised by the NGPS4 dataset. The factors of human disturbance (residential land, roads and farm land) were included in the dataset of land cover.

Data analysis

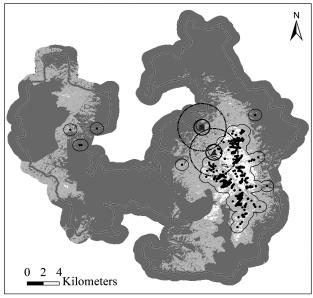
Habitat suitability in the nature reserve and 2 km buffer zones surrounding it was evaluated with Maximum Entropy Modeling (MaxEnt model) (Phillips & Dudík 2008). The records of giant panda from NGPS4 were used as presence data. We used 10-folding crossvalidation to assess the predictive performance of the model. The resulting MaxEnt model generates a Habitat Suitability Index (HSI) between zero and one, and predicts giant panda habitat suitability across the whole reserve region. Model performance was assessed using the Area Under Curve (AUC) method (Fielding & Bell 1997) and True Skill Statistics (TSS) (Allouche et al. 2006). For AUC values, a value closer to one indicates greater predictive ability (Fielding & Bell 1997). A TSS value greater than 0.70 is considered as excellent (Allouche et al. 2006).

We established 100-m buffer zones around the activity sites of released individuals (HJ: 1-11 months; TT: 1-24 months; ZX: 1-24 months) with the feces of resident giant pandas as the center and calculated the mean HSI in each buffer region. We randomly generated 1000 spots in the nature reserve and calculated the mean HSI within 100 m around the site as markers for background habitat quality. In order to compare the differences between two release sites, we calculated the quantity of resident giant panda track points, number of individuals, and mean HSI within 1 km and 3 km of the two release sites.

We used an analysis of variance (ANOVA) to compare the HSI of around the activity sites for TT, HJ, and ZX at 1-6 months, 7-12 months, 13-18 months, and 19-24 months after release in order to examine whether there are changes in the post-release habitats of released individuals with time. As data at 1-6 and 7-11 months after release was only available for HJ, we employed a t-test to compare whether there were significant

Table 1. The quantity of wild giant panda track points, number of individuals, and mean HSI within 1 and 3 km of the two release sites.

	Site 1		Site 2	
	1 km buffer zones	3 km buffer zones	1 km buffer zones	3 km buffer zones
Resident individuals	1	1	1	1
Track points	6	107	1	1
Mean HSI	0.4264	0.4213	0.1075	0.1703



- Study area Track-points
 - Track-points of resident individuals
- 736-m buffer zones surrounding track-points
- Release site 2 3-km buffer zones surrounding site

Fig. 2. The Habitat Suitability Index (HSI) for giant pandas in Liziping National Nature Reserve and the 2 km buffer zones surrounding it. From dark to light changes represent the quality classification of habitat, which is lower than the background average, between the average value of background and resident individuals, and higher than the average value of resident individuals.

differences in HSI of the habitats in these two time periods. Tukey's multiple comparison adjustment was used for pairwise comparisons of the HSI of habitats in different time periods for the other individuals.

In order to test whether the released individuals possess habitat selection capabilities, we used one-way ANOVAs to compare the quality and HSI of habitats of released individuals with the HSI of habitats of resident individuals. To test the interaction on habitat between released giant pandas and resident ones, we assessed the overlap between location of released giant pandas and ranges occupied by resident ones. Hu et al. (1985) suggested that overlapping among the home-range of giant pandas are common, and the non-overlapping habitat used by giant pandas

are only about 1.7 km², even if the size of the home range is range from 3-6 km². Therefore, we regarded the areas within a 736 m radius from each location of resident giant panda as the areas used by the residents. We generally believe that this area does not contain the home range of released pandas due to home range of wild adult giant panda becomes stable and the same region will be used for many continuous years (Zhang et al. 2014, Liu et al. 2015).

To test the release site fidelity, we compared the distances of every released giant pandas to their release sites among months after release, respectively, using a one-way ANOVA. Tukey's multiple comparison adjustment was used for pairwise comparison. All analyses were carried out in the statistical computing environment R (R Core Team 2018).

Results

For the MaxEnt model, the AUC values of the training sets and validation sets were 0.9935 and 0.9887 respectively, and the TSS values was 0.7716. These results indicated that the MaxEnt model had high levels of predictive performance.

The average HSI of habitat used by HJ, TT, ZX, resident giant pandas, and background habitat were 0.4782, 0.3928, 0.2045, 0.5783, and 0.1594, respectively, in the study area (Fig. 2). The quantity of resident giant panda track points, number of individuals, and average HSI of habitat within 1 km and 3 km of the two release sites were difference (Table 1, Fig. 2).

The HSI of the habitats used in different time periods showed significant differences for all three released individuals after release (one-way ANOVA; HJ: $F_{1,6696} = 6460.9, P < 0.01$; TT: $F_{3,11391} = 640.5, P < 0.01$; ZX: $F_{3,8771} = 460.8, P < 0.01$). A pairwise comparison showed that the HSI of the habitats used by HJ at 7-11 months was significantly lower than at 1-6 months (t-test; $t_{5151} = 59.2$; P < 0.01) (Fig. 3a). The HSI of the habitat used by TT in the four time periods after release first decreased before increasing (Fig. 3b). The HSI of the habitat used by ZX in the four time periods after release first increased before decreasing (Fig. 3c).

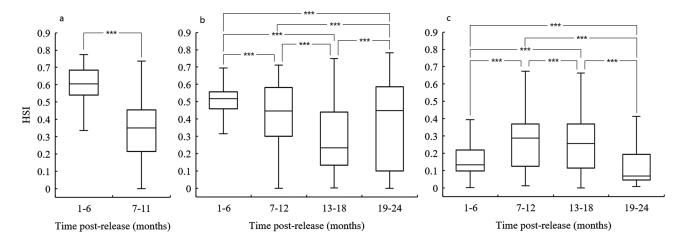


Fig. 3. a), b) and c) are the Habitat Suitability Index (HSI) of habitat used by three translocated giant pandas HJ, TT and ZX during different stage over the time post-release, respectively. In each box plot, the median is represented by a line in box, the box represents the upper and lower quartiles, while the whiskers represent 95 % of the data. *** = P value less than 0.001.

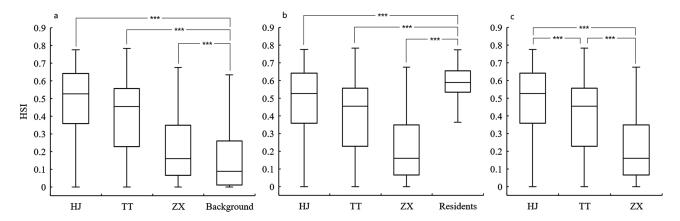


Fig. 4. a) Comparison of Habitat Suitability Index (HSI) of the habitats used by the three individuals and background HSI; b) comparison of HSI of the habitats used by the three individuals and the HSI of habitats used by wild pandas; c) comparison of HSI of the habitats used by the three individuals. In each box plot, the median is represented by a line in box, the box represents the upper and lower quartiles, while the whiskers represent 95 % of the data. *** = P less than 0.001.

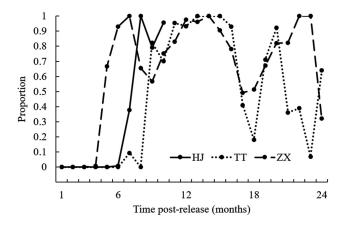


Fig. 5. The averaged proportion of the activity points of the three translocated giant pandas outside the areas used by resident individuals over the time post-release (months).

The HSI of the habitats used in different time periods by the three released individuals showed significant differences with the background values of the study

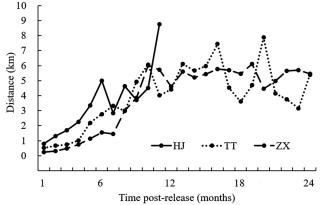


Fig. 6. Mean distance (km) of the three translocated giant pandas moved away from their release site over the time post-release (months).

site and the HSI of habitats used by resident individuals (one-way ANOVA; HJ: $F_{2,24123} = 13541.8$, P < 0.01; TT: $F_{2,35320} = 18318.1$, P < 0.01; ZX: $F_{2,32700} = 27428.9$, P < 0.01). Multiple comparison showed that the HSI

of the habitats used by the three individuals were all significantly higher than the background values but significantly lower than the HSI of habitats used by resident individuals at the same time (Fig. 4a, b). The HSI of the habitats used by the three individuals show significant differences (one-way ANOVA; $F_{2,26865} = 4651.4$, P < 0.01). The HSI of habitats used by ZX released at site 2 was significantly lower than the HSIs of the habitats used by TT and HJ released at site 1 (Fig. 4c).

The released giant pandas stayed in the areas used by resident giant panda in the first several months after release. They then moved out for several months, before then moving in and out of that area alternately (Fig. 5). In the entire study site, 23.45 km² (13.8 %) of regional habitats had HSIs higher than the mean HSI of habitats used by resident individuals, of which 19.26 km² (82.1 %) were located within 763 m ranges of distribution sites of resident individuals (Fig. 2). This means that almost all high-quality habitats were occupied by resident individuals.

The monthly mean distance to the release site ranged from 1.04 to 6.56 km. The mean distances of the activity sites to the release site in different months in the three released individuals showed significant differences (one-way ANOVA; HJ: $F_{10,6687} = 2952.1$, P < 0.01; TT: $F_{23,11371} = 1905.6$, P < 0.01; ZX: $F_{23,8750} = 2072.4$, P < 0.01). Overall, multiple comparisons showed that the distances from the activity site to the release sites of the three individuals in the first six months after release gradually increased, but were mainly within the 3 km range (Fig. 6, Tables S2-S4). At 10 months after release, the distance between the activity site and the release site for TT and ZX was stabilized around 6 km.

Discussion

Our study found that the overall quality level of the habitats selected by the three released individuals was higher than the background value, showing that the three released giant pandas have the ability to select habitats. The ability to select suitable habitats in released individuals is key to success in release programs (Kleiman 1989, Stamps & Swaisgood 2007, Armstrong & Seddon 2008). In addition, the selection must have essential habitat resources to achieve threshold values for survival support and successful reproduction, which is regarded as the first step of release success (Stamps & Swaisgood 2007, Le Gouar et al. 2012). Zhou et al. (2008) carried out a study on releasing giant pandas in the wild adaptation training site and found that captive-bred giant pandas are able to select suitable habitats.

The quality of the habitats used by the three subadult giant pandas were all lower than those used by resident giant pandas. This means that the habitat preferences of released individuals are different from resident individuals but may also mean that exclusion by resident individuals causes them to be unable to enter high-quality habitats. Studies have shown that subadult solitary animals are often excluded to the edges of high-quality habitats due to poor competitiveness (Done & Heatwole 1977, Lovegrove & Veitch 1994, Moehrenschlager & Macdonald 2003). The three giant pandas in this study were all subadults and most of the high-quality habitats in the study site were occupied by resident individuals. During the early stages of release, these pandas were active around resident individuals. However, they later left these areas and the quality of the habitats used also decreased gradually. The studies of Hu (2001) and Song et al. (2006) also showed that high-quality habitats were usually occupied by adults, and subadults could only use areas with poorer habitat quality.

The three individuals did not show loyalty to the release sites. This is evident because they gradually moved further away from the release site after they were released. There are three possibilities for this: searching for better habitat, exploratory behaviour, or intra-species competition. After release, HJ and TT left relatively good habitats for poorer habitats. During the early stage of release, the habitat quality occupied by ZX was not high. During 7-12 months, ZX left for a better habitat. However, it subsequently left that site and moved to an area that had poor habitat quality. This shows that habitat choice cannot explain why released giant pandas did not show loyalty. If released individuals leave the release site purely for exploration and not due to intra-species competition, we can expect that the activities range of released individuals and the distribution of surrounding resident giant pandas to show no association. However, the results showed that the three individuals gradually left the activity area of resident giant pandas. This indicates that the lack of loyalty to the release site in the three released giant pandas in this study is mainly due to competition.

Studies have shown that the habitat selection is dependent on factors such as the quality of habitat at the release site, the number of individuals being introduced (Hodder & Bullock 1997), distribution of conspecific residents (Stamps 1991), and natal experience (Stamps et al. 2009). In this study, site 1 is located at the main activity region of resident giant pandas while site 2 is located outside this region. The habitat quality at site 1 is higher than site 2 and the number of track points of resident giant pandas

is also higher than at site 2. The source and training methods for all released individuals were the same. The quality of the habitats used by pandas released in site 1 is higher than that in site 2. Therefore, we believed that the habitat quality of the release site may affect the choice and utilization of habitats by released individuals after release.

A more ideal habitat selection study would be to experiment releasing pandas in high quality vs. low quality habitats and monitor their movements, and that could help us obtain that knowledge. More questions need to be answered before offering a comprehensive and safe set of guidelines for captive-bred giant panda translocation projects. However, the current number of released giant pandas is very small and only a minority of those individuals have complete post-release GPS collar datasets. With regards to the selection of release sites, we are still unclear on whether in situ adaptation is required before release. However, such trials are almost strategically impossible, due to low tolerance for failure in the release program of an endangered

flagship species such as the giant panda. It is still difficult to balance the risk and potential knowledge gained. In this case, results from other wildlife release projects will be valuable references for giant panda relocation projects. We believe experience will be gained during the practice of releasing other captive-bred species. A good example is the study on translocation of grizzly bears by Milligan et al. (2018). Those experiences could be used for reference when translocating giant pandas in the future. Therefore, we suggest carrying out more captive-bred wildlife releasing projects on other endangered species besides flagship species such as the giant panda in China.

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Supplementary online material

- Table S1. Information of giant pandas released.
- Table S2. Pairwise comparison of the monthly mean distance from individual TT's activity sites to the release site.
- Table S3. Pairwise comparison of the monthly mean distance from individual HJ's activity sites to the release site.
- **Table S4.** Pairwise comparison of the monthly mean distance from individual ZX's activity sites to the release site (https://www.ivb.cz/wp-content/uploads/FZ-vol.-68-2-2019-Zhu-et-al.-Tables-S1-S4.pdf).