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Assessing the health status of released, captive-bred giant pandas (*Ailuropoda melanoleuca*) through activity patterns

Liuyang HE[1,2,6], Qiang DAI[2]*, Zhisong YANG[1]*, Ke HE[1], Jing QING[1], Feng HUANG[3], Xiaodong GU[4]*, Xuyu YANG[4], Yan HUANG[5], Desheng LI[5], Hemin ZHANG[5] and Xiao ZHOU[5]

[1] *Key Laboratory of Southwest China Wildlife Resources Conservation (Ministry of Education), China West Normal University, 637002 Nanchong, China; e-mail: 517161929@qq.com, yangzhisong@126.com, heke0611@163.com, qingjing77@yeah.net*
[2] *Chengdu Institute of Biology, Chinese Academy of Sciences, 610041 Chengdu, China; e-mail: daiqiang@cib.ac.cn*
[3] *Liziping National Nature Reserve, 625400 Shimian, China; e-mail: 22259203@qq.com*
[4] *Sichuan Station of Wildlife Survey and Management, 610082 Chengdu, China; e-mail: 180793519@qq.com, 657525141@qq.com*
[5] *China Conservation and Research Center for the Giant Panda, 623006 Wolong, China; e-mail: 36641221@qq.com, 1050133153@qq.com, 2892959098@qq.com, 58339217@qq.com*
[6] *Gonggashan National Nature Reserve, 625400 Shimian, China*

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Abstract. Translocation is believed to be one of the important methods to protect small and isolated populations of giant pandas (*Ailuropoda melanoleuca*) from local extinction. Dynamic monitoring of the giant pandas' health statuses after release allows timely rescue measures or termination of the release procedure when necessary, thereby reducing the risk of death. However, it is difficult to diagnose the health of a giant panda after translocation by routine veterinary examinations. Based on data collected from GPS tracking collars, we analyzed the behaviours of six giant pandas after release. We compared the behavioural differences between the giant panda Xuexue (XU), which died due to disease, and four healthy giant pandas after release into the wild to identify a new method to assess the health statuses of giant pandas after release. Our results showed that daily activity levels of the healthy giant pandas were higher than XU after released into the wild; the healthy giant pandas were diurnal, but XU tended to be nocturnal. All healthy giant pandas and XU were not active in the crepuscular phase. These results suggest that daily activity levels and circadian rhythms can be used to assess the health status of giant pandas during the translocation project.

Key words: translocation, activity level, daily rhythm, behaviour indicator

Introduction

Translocation is an effective method for maintaining the long-term survival of species endangered by habitat fragmentation and population segregation (Tudge 1991). Although regarded as an important conservation method and extensively applied, many translocation efforts end in failure (Berger-Tal & Saltz 2014), especially for captive-bred individuals (Fischer & Lindenmayer 2000, Kellison et al. 2000, Mathews et al. 2005), whose behavioural skills associated with fitness in the wild are generally poor (Soulé 1987, Snyder et al. 1996, Rabin 2003). Due to stress during translocation and the inability of animals to adapt to the new environment in a timely manner, released individuals often have relatively high mortality rates during the early stages of translocation (Letty et al. 2000). Therefore, dynamic monitoring of the health status of individuals after the release allows timely rescue efforts or even the termination of the translocation procedure when necessary, which may reduce the risk of death of the released individuals.

Behavioural patterns, such as activity levels and rhythms, effectively reflect the health status of animals (Edmunds et al. 2018). Timely monitoring of the health status of individuals after release and taking corresponding rescue measures in the case of abnormalities are important means to reduce the risk of translocation failure (Mathews et al. 2006). However, it is difficult to assess animals' health statuses via routine veterinary examinations after their release into

* *Corresponding Author*

the wild, especially for species living in dense forests, such as giant pandas. Veterinarians often encounter challenges when performing even the most basic visual examinations on those animals. Moreover, frequent veterinary examinations may be stressful and increase the stress response of released animals, aggravating the risk of failure of the translocation program (Romero & Reed 2008, Leche et al. 2016).

Fortunately, the behavioural data of released animals can be continuously recorded with Global-positioning-system (GPS) tracking collars (Augustine & Derner 2013). Therefore, the behaviour of released animals can be expediently understood and be used as indicators to assess the status of organisms, and in turn evaluate and guide translocation programs (Berger-Tal et al. 2011, Caro & Sherman 2013, Greggor et al. 2016). A few studies have used this type of behavioural data to identify the reproductive statuses of animals in the wild (Friebe et al. 2013, Zhang et al. 2017, He et al. 2018). Application of a similar approach, monitoring the behavioural data of the released individuals for health monitoring without disturbing the released individuals, will lower the risk of failure of the translocation program. However, this behaviourally diagnostic method has not been extensively applied in translocation projects.

Translocation is an indispensable method in the conservation of giant pandas, especially for small populations (Zhang & Wei 2006). The giant panda, arguably the most popular flagship species in the world, had its IUCN Red List status changed from "Endangered" to "Vulnerable" (Swaisgood et al. 2016) because of its continuous increase in population size for the past 30 years. However, some local populations of giant pandas are small and isolated in heavily fragmented habitat patches, especially in the southern area of their range (Qing et al. 2016). Those small, isolated populations are vulnerable to extinction as a result of genetic erosion and a variety of demographic and environmental issues (Schonewald-Cox et al. 1983, Soulé 1987, Loeschcke et al. 1994). Translocation can increase the population size and genetic diversity of small, isolated populations, and thus reduce the risk of local extinction of giant pandas (Wei et al. 2015).

Behavioural reports of released giant pandas are very limited (but see He et al. 2018, Yang et al. 2018), mainly due to the small number of giant pandas have been released. Only 12 giant pandas have been released between 2006 and 2018. Nine giant pandas were released into the Liziping National Nature Reserve (LNNR), and the remaining three were released into the Longxi-Hongkou National Nature Researve in Sichuan Province, China. Behavioural diagnostic methods based on the activity data of GPS tracking collars is not only important for improving the translocation success rate of giant pandas, but also for translocation programs of other species. Therefore, this study analyzed the activity patterns of six captive-bred giant pandas at the early stages of the translocation process in the Xiaoxiangling Mountain Range during 2012-2016 to compare the daily activity rhythms and activity levels between surviving and dead giant pandas after the release. This allowed us to look for indicators to determine the health statuses of giant pandas after being released.

Study Area

The study area is located in the LNNR (Fig. 1) in the Xiaoxiangling Mountain Range, south-west of the distribution range of giant panda. The reserve covers an area of 47940 ha, 80 % of which is regarded as habitat suitable for giant pandas (Sichuan Forestry Department 2015). The reserve's altitude varies from 1330 to 4551 meters. The population of giant pandas in the reserve is small (22 native giant pandas) (Sichuan Forestry Department 2015) and heavily isolated (Qing et al. 2016).

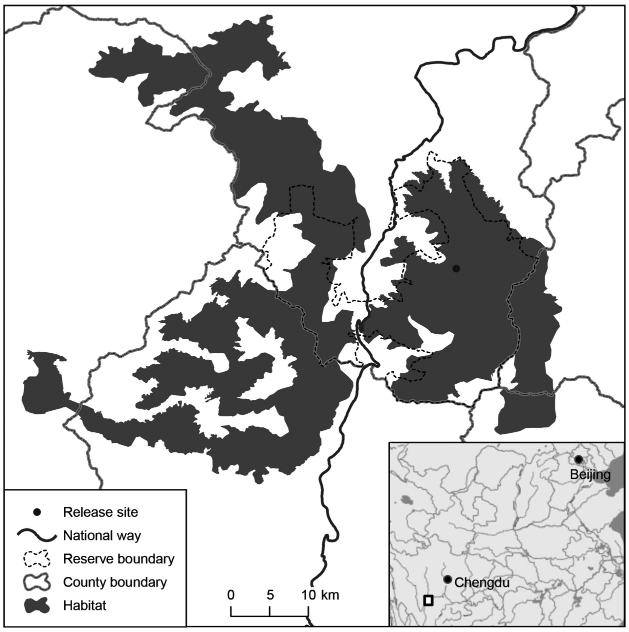

Fig. 1. Location of the Liziping National Nature Reserve.

The releasing enclosure, where the soft release projects were implemented, is located in the natural forest in the LNNR reserve. It fences 20 ha of natural forest, with altitudes ranging from 2050 to 2400 meters. The dominant vegetation cover in the enclosure is deciduous and evergreen trees (approximately 60 %), followed by evergreen conifer trees (approximately 36 %). One stream and two species of bamboo occur in the enclosure naturally, enabling giant pandas to drink and forage without any supplemental feeding.

Material and Methods

Subjects and data collection

Six giant pandas, named Taotao (TT), Zhangxiang (ZX), Xuexue (XU), Huajiao (HJ), Huayan (HY), and Zhangmeng (ZM), were released into the LNNR between 2012 and 2016. One male (TT) and five females were all captive-born in the China Conservation and Research Center for the Giant Pandas (CCRCGP). Those giant pandas received more than two years of pre-release training before they were released at about 2.5 years of age, which is the age wild giant pandas leave their mothers' home ranges and begin to live independently (Schaller et al. 1985). During the training, giant pandas meant to be released were cared for by their mothers in a semi-natural enclosure with abundant food and water since birth. No supplemental food or water was provided. In other words, the candidates were mainly trained by their mothers rather than by a human. All giant pandas were transported in a covered cage from CCRCGP to the release sites, which took about 4-5 hours' travel time.

Four giant pandas, one male (TT) and three females (HJ, HY and ZM), were hard released right after they reached the release site. The cage holding the giant pandas was placed at the release site, and the entrance was simply unblocked to let the giant pandas exit freely. TT and HY were released in 2012 and 2015, respectively, while HY and ZM were released at the same time in 2016. Two female giant pandas (ZX and XU) were soft released in 2013 and 2014, respectively. These giant pandas were transported directly into the releasing enclosure and kept for 28-53 days. No supplemental food or water was provided during those days; the pandas only had access to the naturally occurring bamboo and stream water. At the time of release, a door in the fence was opened and the giant pandas went out freely when they were released into the wild.

Before being transported to either the hard release site or the release enclosure, all giant pandas were chemically immobilized and fitted with GPS 7000MU collar (Lotek Wireless Inc, Newmarket, Ontario, Canada), which were lighter than 5 % of their body weight. The activity of each giant panda was recorded with a dual-axis activity (X axis: side-to-side and Y axis: up-down) sensor integrated into the collar. Counts of activity were recorded every five minutes for each axis from 0 (no activity) to 255 (highest activity).

Since this study focused on the activity levels and rhythms of the giant pandas in the early stages of the translocation process, data from the first 120 days after the translocation were used for analysis. Giant pandas TT, ZX, HJ, and ZM have been monitored for more than 120 day, and were considered to have normal health statuses after the translocation according > 1-year survival. However, giant panda XU died 34 days after release, so only 34 days of data were collected for this animal. An autopsy revealed no physical trauma on XU's body, and its death was possibly due to an infection caused by an opportunistic, pathogenic bacteria. XU was considered as a non-healthy individual in this study. In addition, since the GPS tracking collar worn by giant panda HY fell off on the 53^{th} day after release, the health status of subsequent dates was unknown, and therefore this animal was excluded from the healthy controls in this study.

Data Analysis

We selected three indicators, daily activity level, diurnal index, and crepuscular index, to reflect the activity patterns of giant pandas after released into the wild. We compared the differences between the healthy giant pandas (TT, ZX, HJ and ZM) and XU, and analyzed if these three indicators could be used to distinguish between non-healthy XU and the remaining, healthy giant pandas.

The sensor values of X- and Y-axes from the collars were summed at 5-min intervals, and then used to indicate the activity levels of the giant pandas, following the methods described by previous studies (Turner et al. 2002, Yamazaki et al. 2008). Daily activity level was defined as the averaged 5-min activity of every day, and is presented as mean ± SD. The activity was classified as daytime, nighttime, or twilight activity. Daytime was defined as the time from civil twilight at dawn to civil twilight at dusk, and nighttime was defined as the time from civil twilight in the evening to civil twilight in the morning of the next day. Twilight was defined as the time between nautical twilight and sunrise in the morning and between sunset and nautical twilight in the evening. To indicate the relative activity during daytime and twilight, the diurnal and crepuscular indices were calculated using equations 1 and 2, respectively (Ensing et al. 2014). The values of diurnal and crepuscular indices lie between −1 and 1. A positive value on the diurnal index indicates giant pandas are more active during the day, while a positive value on the crepuscular index indicates a preference for activity in the crepuscular phase.

(Eq. 1)

$$I_{diurnal} = \frac{ACT_{day} - ACT_{night}}{ACT_{day} + ACT_{night}}$$

(Eq. 2)

$$I_{crepuscular} = \frac{ACT_{twilight} - ACT_{non\text{-}twilight}}{ACT_{twilight} + ACT_{non\text{-}twilight}}$$

Where $I_{diurnal}$ is the diurnal index, $I_{crepuscular}$ is the crepuscular index, ACT_{day} is the mean activity in the daytime, ACT_{night} is the mean activity in the nighttime, $ACT_{twilight}$ is the mean activity during twilight, and $ACT_{non\text{-}twilight}$ is the mean activity during the remainder of the day.

The averaged 5-min activities of every hour for every 10 days were calculated in order to assess the pattern of daily activity rhythm for each individual. To investigate whether the released giant pandas showed a crepuscular or a diurnal active pattern, we tested whether the diurnal index and crepuscular index significantly differed from 0 with a t-test at an interval of 10 days.

A receiver operating characteristic curve (ROC) was used to examine if daily activity level, diurnal index, and crepuscular index were suitable for distinguishing XU from healthy giant pandas. A ROC illustrates the diagnostic ability of a binary classifier system as its discrimination threshold is varied. ROC analyses has been used in medicine (Levy et al. 2006), radiology (Obuchowski 2003), biometrics (Toh et al. 2008), model performance assessment (Qing et al. 2016), and other areas for many decades. The area under the ROC curve (AUC) is a measure of how well a parameter can distinguish between two diagnostic groups (diseased/normal). An AUC > 0.9 indicates an excellent diagnostic performance, an AUC ranging 0.8-0.9 indicates a good performance, an AUC ranging 0.7-0.8 indicates a fair performance, and an AUC < 0.7 indicates a poor performance. The thresholds for an abnormal active pattern were determined by the parameter with the highest Youden Index, or equivalently, the highest Sensitivity + Specificity (Krzanowski & Hand 2009).

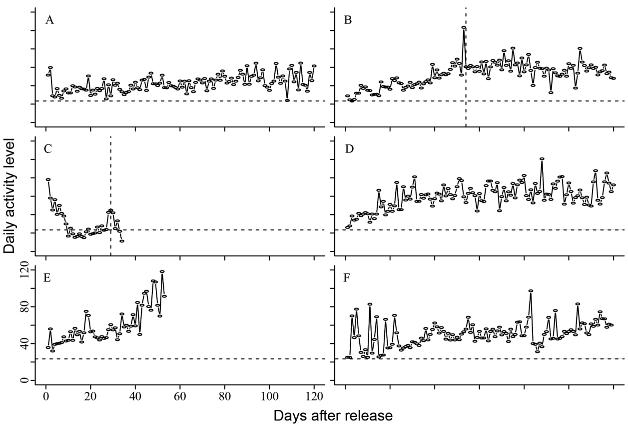

Fig. 2. The daily activity level of the giant pandas (A: TT, B: ZX, C: XU, D: HJ, E: HY, F: ZM). The open circles represent the daily activity level of the giant pandas. The dashed horizontal line shows the lowest daily activity level of living giant pandas after release, and the dashed vertical lines in panels C and D show the date when the giant panda left the enclosure.

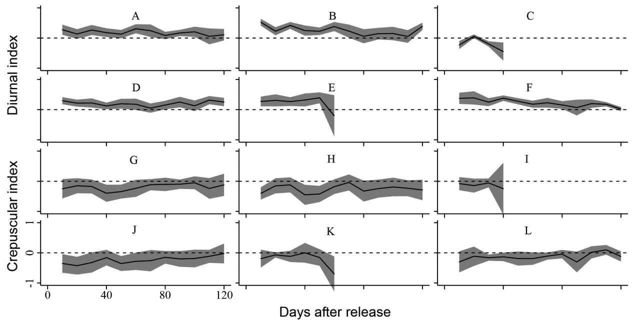

Fig. 3. Diurnal (A-F) and crepuscular indices (G-L) for HJ, XY, TT, XU, ZM, and ZX. The black lines represent the average index values. The grey areas represent the 95 % confidence interval, which can be interpreted as a one-sided t-test: when the 95 % confidence interval (grey area) does not encompass the 0 line (dotted), the activity pattern significantly deviates from an equal distribution ($P < 0.05$).

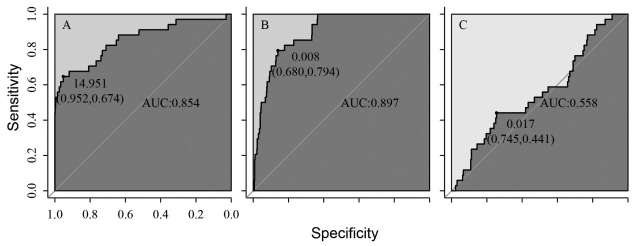

Fig. 4. The receiver operating characteristic curve (ROC) for daily activity level (A), diurnal index (B), and crepuscular index (C).

Results

After the release, the daily activities of giant pandas TT, ZX, HJ, and HY began at relatively low levels at the earliest stage and gradually increased over time. The daily activities of giant panda ZM greatly varied in the first 20 days after the release, but subsequently and gradually increased over time. In contrast, giant panda XU was highly active immediately after release, and this activity level was rapidly declined to a minimal level (Fig. 2).

After translocation, the average daily activity levels of TT, ZX, HJ, ZM, and HY were 21.23 ± 4.41 (n = 120), 26.08 ± 6.61 (n = 120), 30.00 ± 6.74 (n = 120), 25.23 ± 6.61 (n = 120), and 30.11 ± 10.00 (n = 53), respectively. XU died 34 days after translocation. Even though XU was highly active in the first several days, the overall average daily activity of XU was the lowest at 15.08 ± 7.58 (n = 34), due to her low activities in the days that followed. The daily activity level of XU was significantly lower than the other individuals (t-test TT: df = 152, t = −6.009, $P < 0.01$; ZX: df = 152, t = −8.282, $P < 0.01$; HJ: df = 152, t = −11.076, $P < 0.01$; HY: df = 85, t = −7.486, $P < 0.01$, ZM: df = 152, t = −7.632, $P < 0.01$).

Individuals TT, ZX, HJ, HY, and ZM showed similar patterns in their daily rhythm of activity, which were characterized by one or two peaks during the daytime and one in the night (Fig. S1). One simple t-test on the diurnal index revealed that TT, ZX, HJ, HY, and ZM are generally more active in the daytime than in the nighttime, while XU was just the opposite (Fig. 3). The negative crepuscular indices indicated all the giant pandas, including XU, were inclined to follow non-twilight activity patterns (Fig. 3). XU was no less active in the dawn twilight than in daytime or nighttime, and her negative crepuscular indices were due to the inactivity in dusk twilight (Fig. S1).

The threshold for abnormal active pattern was determined by a ROC (Fig. 4). The point of maximum sensitivity and specificity occurred when the daily activity level was 14.951. Diagnostic performance of averaged activity level as determined by the AUC was 0.854, which indicated a good accuracy for a diagnostic test. Thus, an averaged activity level < 14.951 identified XU as different from other released giant pandas. Thresholds and AUC scores for diurnal index and crepuscular index were determined in a similar manner from ROC analysis. The threshold for diurnal index was 0.008, with an AUC of 0.897, which indicated a good diagnostic performance. However, the AUC for crepuscular index was 0.558, which indicated a poor diagnostic performance.

Discussion

Animals' behaviours respond to changes in health status (Clubb & Mason 2003, Maple 2007). Behaviour has always been recognized as a crucial indicator of health in domestic animals (Fraser & Broom 1997) and zoo animals (Hill & Broom 2009). However, studies assessing the health status of wild animals through their behaviours are very limited. This study proposes a method for health assessment of giant pandas after being release into the wild. According to the results, the daily activity levels and diurnal indices of giant pandas were able to distinguish individual XU, the giant panda which died after release, from the other, healthy giant pandas. Behavioural identification of the released animals based on the activity data collected from the GPS tracking collars helped us to understand the health of individual animals after translocation and to adjust the translocation strategy in a timely manner. This may have a great impact on improving the success rate of translocations.

The dead individual, XU, had a significantly lower activity level after release than the other individuals and its circadian rhythm was also different from the others. Unhealthy animals usually behaves abnormally (Edmunds et al. 2018), but we still cannot rule out the possibility that the abnormal behaviours make XU more vulnerable to sickness and not the other way around. In general, the establishment of an anomaly index requires a large number of normal and abnormal data points. However, in this study, XU is the only dead giant panda with activity data after the translocation. Therefore, application of XU's data as the standards for the establishment of abnormal cases (daily activity level: < 14.951, diurnal index: < 0.008) still provided a valuable reference for future translocation programs. It is important to note that the causes of death in individuals after translocation may be diverse (Moreno et al. 1996, Larkin et al. 2003), and therefore the abnormal behaviours before death may also be diverse. Inactivity is common for animals with poor health (Montgomery 1953, Edmunds et al. 2018). However, agitation is also common among diseased animals (Hill & Broom 2009). In addition, the abnormal behavioural patterns of animal dying of hunger (Islam et al. 2008, Herzog et al. 2009) may be different from the abnormal behavioural patterns of animal which die of disease. Hence, accumulation of more data is needed to refine this series of diagnostic criteria.

The activity levels of the healthy, released giant pandas in this study were slightly lower than that of wild giant pandas in the first month. The average activity level of the wild giant panda was approximately 24 (Zhang et al. 2015), while the initial average activity level of the healthy giant pandas was 20 in this study. Given the different type of GPS collar used between Zhang et al. (2015) and the current study, we should be cautious about the comparison between the two studies. The initial low activity observed in the released giant pandas implied activity suppression by potential stress; subsequently, the activity of the released giant pandas increased, suggesting there may be an initial exploration behaviour in the individual animals. Exploration, coupled with high activity, is common in animals in a novel environment (Renner 1990, Pinter-Wollman 2009). Exploration can provide animals with critical information about food, refuge, and predators (Renner 1990, Inglis 2000, Russell et al. 2010). The increase in activity after the initial days indicated a gradual recovery from the potential stress and acclimation to the novel environment.

The activity rhythm is critical for the health of animals, a disorder of which can result in metabolism or immunity dysfunction (Bechtold et al. 2010). In this study, the released individuals that survived after release all showed a continuous diurnal activity pattern. Similar diurnal activity patterns were observed in wild giant pandas in the studies carried out in the Qionglai Mountain Range (Hu 2001, Zhang et al. 2015, Zhang et al. 2017) and the Qinling Mountain Range (Pan et al. 2014). In this study, the activity levels of the released giant pandas in the crepuscular phase were lower than the activity levels in the remaining phase. Hu et al. (1985) suggested that the wild giant pandas in Wolong were inclined to be active in the crepuscular phase, especially at dusk, while Zhang et al. (2015) found that most of the wild giant pandas had relatively low activity levels at dusk. In addition, Zhang et al. (2015) defined the crepuscular phase as two hours around sunrise and two hours around sunset, while Hu et al. (1985) did not clearly define the crepuscular phase in their study. Nevertheless, according to the description in the results, we infer the definition of crepuscular phase of Hu's et al. (1985) study is similar to that of Zhang et al. (2015). The definition of crepuscular phase of these two studies is different from our current study, which may affect the preference analysis of giant pandas in the crepuscular phase.

Berger-Tal et al. (2019) suggested using behavioural indicators to monitor the status of wildlife and guide conservation efforts. This study screened two behavioural indicators to assess the health status of individual giant pandas after translocation. According to the results of this study, giant panda XU, which died after release, showed behavioural abnormalities in the release enclosure. This translocation failure might have been avoided with a timely suspension of the translocation procedure for this animal upon observing its abnormal behaviours. It should be noted that the sample size of this study is small so the effectiveness of the method cannot be fully evaluated. More translocation attempts and relevant studies will help improve the current method and potentially find even more behavioural indicators.

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Literature

Augustine D.J. & Derner J.D. 2013: Assessing herbivore foraging behavior with GPS collars in a semiarid grassland. *Sensors 13: 3711–3723.*

Bechtold D.A., Gibbs J.E. & Loudon A.S. 2010: Circadian dysfunction in disease. *Trends Pharmacol. Sci. 31: 191–198.*

Berger-Tal O., Greggor A.L., Macura B. et al. 2019: Systematic reviews and maps as tools for applying behavioral ecology to management and policy. *Behav. Ecol. 30: 1–8.*

Berger-Tal O. & Saltz D. 2014: Using the movement patterns of reintroduced animals to improve reintroduction success. *Curr. Zool. 60: 515–526.*

Berger-Tal O., Polak T., Oron A. et al. 2011: Integrating animal behavior and conservation biology: a conceptual framework. *Behav. Ecol. 22: 236–239.*

Caro T. & Sherman P.W. 2013: Eighteen reasons animal behaviourists avoid involvement in conservation. *Anim. Behav. 85: 305–312.*

Clubb R. & Mason G. 2003: Animal welfare: captivity effects on wide-ranging carnivores. *Nature 425: 473–474.*

Edmunds D.R., Albeke S.E., Grogan R.G. et al. 2018: Chronic wasting disease influences activity and behavior in white-tailed deer. *J. Wildlife Manage. 82: 138–154.*

Ensing E.P., Ciuti S., de Wijs F.A. et al. 2014: GPS based daily activity patterns in European red deer and North American elk (*Cervus elaphus*): indication for a weak circadian clock in ungulates. *PLOS ONE 9: e106997.*

Fischer J. & Lindenmayer D.B. 2000: An assessment of the published results of animal relocations. *Biol. Conserv. 96: 1–11.*

Fraser A.F. & Broom D.M. 1997: Farm animal behaviour and welfare. *CAB International, Wallingford.*

Friebe A., Zedrosser A. & Swenson J.E. 2013: Detection of pregnancy in a hibernator based on activity data. *Eur. J. Wildlife Res. 59: 731–741.*

Greggor A.L., Berger-Tal O., Blumstein D.T. et al. 2016: Research priorities from animal behaviour for maximising conservation progress. *Trends Ecol. Evol. 31: 953–964.*

He K., Qing J., Zhang Z. et al. 2018: Assessing the reproductive status of a breeding, translocated female giant panda using data from GPS collar. *Folia Zool. 67: 40–46.*

Herzog C.J., Czeh B., Corbach S. et al. 2009: Chronic social instability stress in female rats: a potential animal model for female depression. *Neuroscience 159: 982–992.*

Hill S.P. & Broom D.M. 2009: Measuring zoo animal welfare: theory and practice. *Zoo Biol. 28: 531–544.*

Hu J. 2001: Research on the giant panda. *Shanghai Publishing House of Science and Technology, Shanghai.*

Hu J., Schaller G.B. & Pan W. 1985: The giant panda of Wolong. *Sichuan Science and Technology Press, Chengdu.*
Inglis I.R. 2000: The central role of uncertainty reduction in determining behaviour. *Behaviour 137: 1567–1599.*
Islam Z.M., Ismail K. & Boug A. 2008: Re-introduction of the red-necked ostrich, *Struthio camelus camelus*, in Mahazat as-Sayd Protected Area in central Saudi Arabia. *Zool. Middle East 44: 31–40.*
Kellison G.T., Eggleston D.B. & Burke J.S. 2000: Comparative behaviour and survival of hatchery-reared versus wild summer flounder (*Paralichthys dentatus*). *Can. J. Fish. Aquat. Sci. 57: 1870–1877.*
Krzanowski W.J. & Hand D.J. 2009: ROC curves for continuous data. *Chapman & Hall, New York.*
Larkin J.L., Alexy K.J., Bolin D.C. et al. 2003: Meningeal worm in a reintroduced elk population in Kentucky. *J. Wildl. Dis. 39: 588–592.*
Leche A., Cortez M.V., Costa N.S.D. et al. 2016: Stress response assessment during translocation of captive-bred greater rheas into the wild. *J. Ornithol. 157: 599–607.*
Letty J., Marchandeau S., Clobert J. & Aubineau J. 2000: Improving translocation success: an experimental study of anti-stress treatment and release method for wild rabbits. *Anim. Conserv. 3: 211–219.*
Levy W.C., Mozaffarian D., Linker D.T. et al. 2006: The Seattle heart failure model: prediction of survival in heart failure. *Circulation 113: 1424–1433.*
Loeschcke V., Tomiuk J. & Jain S. 1994: Conservation genetics. *Birkhäuser Verlag, Basel.*
Maple T.L. 2007: Toward a science of welfare for animals in the Zoo. *J. Appl. Anim. Welf. Sci. 10: 63–70.*
Mathews F., Moro D., Strachan R. et al. 2006: Health surveillance in wildlife reintroductions. *Biol. Conserv. 131: 338–347.*
Mathews F., Orros M., McLaren G. et al. 2005: Keeping fit on the ark: assessing the suitability of captive-bred animals for release. *Biol. Conserv. 121: 569–577.*
Montgomery K.C. 1953: The effect of the hunger and thirst drives upon exploratory behavior. *J. Comp. Physiol. Psychol. 46: 315–319.*
Moreno S., Villafuerte R. & Delibes M. 1996: Cover is safe during the day but dangerous at night: the use of vegetation by European wild rabbits. *Can. J. Zool. 74: 1656–1660.*
Obuchowski N.A. 2003: Receiver operating characteristic curves and their use in radiology. *Radiology 229: 3–8.*
Pan W., Lü Z., Zhu X. et al. 2014: A chance for lasting survival: ecology and behavior of wild giant pandas. *Smithsonian Institution Scholarly Press, Washington, D.C.*
Pinter-Wollman N. 2009: Spatial behaviour of translocated African elephants (*Loxodonta africana*) in a novel environment: using behaviour to inform conservation actions. *Behaviour 146: 1171–1192.*
Qing J., Yang Z., He K. et al. 2016: The minimum area requirements (MAR) for giant panda: an empirical study. *Sci. Rep. 6: 37715.*
Rabin L.A. 2003: Maintaining behavioural diversity in captivity for conservation: natural behaviour management. *Anim. Welfare 12: 85–94.*
Renner M.J. 1990: Neglected aspects of exploratory and investigatory behavior. *Psychobiology 18: 16–22.*
Romero L.M. & Reed J.M. 2008: Repeatability of baseline corticosterone concentrations. *Gen. Comp. Endocrinol. 156: 27–33.*
Russell J.C., McMorland A.J.C. & MacKay J.W.B. 2010: Exploratory behaviour of colonizing rats in novel environments. *Anim. Behav. 79: 159–164.*
Schaller G.B., Hu J., Pan W. et al. 1985: The giant pandas of Wolong. *University of Chicago Press, Chicago.*
Schonewald-Cox C.S., Chambers S.M., MacBryde B. & Thomas L. 1983: Genetics and conservation: a reference for managing wild animal and plant populations. *Benjamin-Cummings, London.*
Sichuan Forestry Department 2015: The pandas of Sichuan: the 4th survey report on giant panda in Sichuan Province. *Sichuan Science and Technology Press, Chengdu.*
Snyder N.F.R., Derrickson S.R., Beissinger S.R. et al. 1996: Limitations of captive breeding in endangered species recovery. *Conserv. Biol. 10: 338–348.*
Soulé M.E. 1987: Viable populations for conservation. *Cambridge University Press, Cambridge.*
Swaisgood R.R., Wang D. & Wei F. 2016: *Ailuropoda melanoleuca*. The IUCN Red List of Threatened Species 2016: e.T712A102080907. *Downloaded on 15 March 2017, http://dx.doi.org/10.2305/IUCN.UK.2016-2.RLTS.T712A45033386.en*
Toh K., Kim J. & Lee S. 2008: Maximizing area under ROC curve for biometric scores fusion. *Pattern Recogn. 41: 3373–3392.*
Tudge C. 1991: Last animals at the Zoo. *Hutchison Radius Press, London.*
Turner J.W., Tolson P. & Hamad N. 2002: Remote assessment of stress in white rhinoceros (*Ceratotherium simum*) and black rhinoceros (*Diceros bicornis*) by measurement of adrenal steroids in feces. *J. Zoo Wildl. Med. 33: 214–221.*
Wei F., Swaisgood R.R., Hu Y. et al. 2015: Progress in the ecology and conservation of giant pandas. *Conserv. Biol. 29: 1497–1507.*
Yamazaki K., Kozakai C., Kasai S. et al. 2008: A preliminary evaluation of activity-sensing GPS collars for estimating daily activity patterns of Japanese black bears. *Ursus 19: 154–161.*
Yang Z., Gu X., Nie Y. et al. 2018: Reintroduction of the giant panda into the wild: a good start suggests a bright future. *Biol. Conserv. 217: 181–186.*
Zhang J., Hull V., Huang J. et al. 2015: Activity patterns of the giant panda (*Ailuropoda melanoleuca*). *J. Mammal. 96: 1116–1127.*
Zhang J., Hull V., Ouyang Z. et al. 2017: Modeling activity patterns of wildlife using time-series analysis. *Ecol. Evol. 7: 2575–2584.*
Zhang Z. & Wei F. 2006: Giant panda *ex situ* conservation theory and practice. *China Science Publishing & Media Ltd., Beijing.*

Supplementary online material

Fig. S1. Activity profiles of the released giant pandas at an interval of ten days (A-L). Dark green areas represent activity, and light green areas represent standard deviation. Dotted lines indicate start of nautical twilight (blue), start of civil twilight (green), sunrise (red), sunset (red), end of civil twilight (green) and end of nautical twilight (blue) from left to right (https://www.ivb.cz/wp-content/uploads/FZ-vol.-68-2-2019-He-et-al.-Fig._S1-1.jpg).