# Saving the Amur Tiger and Amur Leopard

## **NEASPEC** Project Report

Transborder Movement of Amur Tigers and Amur Leopards Using Camera Trapping and Molecular Genetic Analysis





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# Introduction

Man-made national boundaries divide ecosystems and habitats with different management and governance structures, but living species freely cross the national borders for seasonal survival, reproductive successes and resilience to climate and other disturbances. Considering such transboundary nature of species and its habitats, an emphasis has been put on transboundary cooperation and conservation. As North-East Asia provides a complex mosaic of habitats for many critically endangered species, North-East Asian countries have established their own domestic protected areas including those along or across the international borders. However, with growing webs of communication and collaboration across the borders, strengthened cooperation among all stakeholders is required for better information sharing, coordinated action, and more efficient and effective transboundary cooperation. NEASPEC member States, in this context, adopted the NEASPEC Nature Conservation Strategy during its 12<sup>th</sup> Senior Officials Meeting in 2007, and identified six flagship species that are critically endangered and unique to the subregion.

The Amur tiger and Amur leopard, among the flagship species, are respectively designated as Endangered and Critically Endangered by IUCN Red List of Threatened Species. Population estimates reach over 500 for Amur tiger and over 60 for Amur leopard in the wild, with most of them live in Northeastern China and Far Eastern Russia, and a few individuals were assumed inhabit in the Democratic People's Republic of Korea (DPRK). As an apex predator residing at the top of a food chain, the Amur tiger and leopard play an important role for heathy ecosystem by balancing the number of wild species, and that they need large habitat area to search enough prey to feed themselves. Therefore, conservation of these two big cat species also includes sustainable management of ecosystem, such as preserving million square meters of the region's forests as well as other wild species. Reliable and precise scientific evidence underpinned by unified research methodologies has been identified as a key challenge for the range countries scaling up efforts on the conversation of endangered species and their habitats. To respond to this challenge, the joint study applied camera trapping and molecular genetic analysis of targeted species in transboundary areas of China and the Russian Federation to provide scientific assessment and consequently developed policy recommendations for better conservation plan and improved ecological corridor management among key member States in North-East Asia.

With the support of NEASPEC Secretariat, the project also reached the following breakthrough that: (1) DNA samples of the Amur tiger and leopard across the Russian Federation border to China were collected for the first time for joint molecular genetic analysis, and (2) Chinese and Russian experts shared data collected through camera trapping and carried out molecular genetic analysis applying the same methodologies that significantly enhanced the data reliability.

# Background

The Amur tiger and leopard are key species that constitute the subregion as a single ecological community, and NEASPEC member governments adopted the NEASPEC Nature Conservation Strategy at the 12th Senior Officials Meeting (SOM) in 2007 and identified six flagship species in North-East Asia, including three feline animals: Amur Tiger, Amur Leopard and Snow Leopard; and three migratory birds: White-naped Cranes, Hooded Cranes and Black-faced Spoonbills. For feline species, the Strategy recommends several priority actions including to: (1) support range countries to collaborate on the conservation of the flagship species facilitated by regular dialogue with international counterparts to coordinate actions on international and national corridors in order to secure safe movement of concerned species, (2) provide regular update on the conservation status, and (3) support capacity building among range countries on habitat management, population management, monitoring and research, law enforcement, environmental education and community development.

Under the NEASPEC project "Establishing Coordination Mechanisms for Nature Conservation in Transboundary Areas in North-East Asia in 2010-2020", two reports were developed regarding the "Review of Environmental, Socioeconomic and Institutional Conditions and Experiences in Multi-/Bilateral Cooperation on Nature Conservation for the Amur Tiger and Amur Leopard in Transboundary Areas in North-East Asia" and "A Cooperation Framework for the Conservation of the Amur Tiger and Amur Leopard in the Tumen River Area". The reports identified key issues and available monitoring methodologies for the conservation of the flagship species in the subregion. In 2013, SOM-18 considered and supported the proposal on species monitoring raised by the Russian Federation. Subsequently an Expert Group Meeting was held in 2014 to discuss the project on "Transborder Movement of Amur Tigers and Amur Leopards Using Camera Trapping and Molecular Genetic Analysis". Since then, the NEASPEC Secretariat and national focal points have implemented project activities including field research and laboratory studies, and held a review meeting in Harbin, China in 2015 to review interim outcomes and pending issues of the project. The project outcomes were presented to experts from China and the Russian Federation at a Workshop on Nature Conservation and Transboundary Cooperation organized in Beijing in 2016, and informed SOM-21 in Seoul in 2017.

# **Overview of the Amur tiger and Amur leopard**

The Amur tiger (Panthera tigris altaica) is a rare subspecies of tiger (P. tigris). Also known as the Siberian, Korean, Manchurian, or North China tiger, it is the largest of all animals in the feline family. Previously widely distributed throughout North-East Asia, Amur tiger is considered "Endangered" as most of the other tiger subspecies are in the International Union for the Conservation of Nature (IUCN) Red List of Threatened Species. Among less than 500 in the wild, approximately 95% of the entire Amur tiger population lives within the Russian Far-East



Distribution

as a single, unfragmented population in Primorsky and Southern Khabarovsk Krais. Although their population is not threatened to imminent extinction, the enlargement of human settlements, their habitat degradation and fragmentation, poaching and illegal trade of tiger parts and derivatives, particularly from 1993 to 2003, are of critical concerns for the destiny of the Amur tiger.

The Amur leopard (Panthera pardus orientalis) is also a rarest subspecies of the Felidae family. Historically it occupied a vast habitat area, including the northern region of the Korean Peninsula, eastern provinces of China and the southern region of the Sikhote-Alin Mountains (Primorsky Krai) of the Russian Federation. However, its habitat began to drastically shrink in the 20th Century, and no individual has been recorded in the Korean Peninsula since the mid-20th Century. The existence of the Amur leopard could not even be confirmed in the Sikhote-Alin Mountains and the northwestern region of Primorsky Krai since 1980s. A recent study showed that, from August 2012 to July 2014, 40 individuals were recorded in China and at most 70 in the Russian Federation<sup>1</sup>. Categorized as "Critically Endangered" in the IUCN Red List, the Amur leopard currently can only be found in Chinese provinces bordering the Russian Federation as well as the southwestern region of Primorsky Krai.

<sup>1</sup> Wang, Tianming, et al. "Amur tigers and leopards returning to China: direct evidence and a landscape conservation plan." Landscape Ecology 31.3(2016):491-503.



#### Amur Tiger (Panthera tigris altaica)

Class	Mammalia
Order	Carnivora
Family	Felidae
Genus	Panthera
Species	Panthera tigris Linnaeus, 1758
Subspecies	Panthera tigris altaica Temminck, 1844
IUCN Red List	Endangered
Population	about 540 (wild, estimated)



Amur Leopard (Panthera pardus orientalis)

Class	Mammalia
Order	Carnivora
Family	Felidae
Genus	Panthera
Species	Panthera pardus Linnaeus, 1758
Subspecies	Panthera pardus Linnaeus, 1758
IUCN Red List	Critically Endangered
Population	less than 60 (wild, estimated)

# **1. Distribution and population**

#### Amur tiger

The Amur tiger in China mostly stays in the northeastern forest area particularly close to Songhua River and Changbai Mountains. At the end of the 19<sup>th</sup> Century, the total number of Amur tigers was about 2,000-3,000 worldwide, and more than half of them lived in China. However, serious interference begun following the ban on forest exploitation eased after the collapse of the Qing Dynasty, and as a result the number of the Amur tiger fell down to about 500 by 1930s. The situation continuously worsened until recently following to the implementation of conservation measures and regulations to protect the Amur tiger and their habitats in China. More specifically, the Xiaoxinganling range was no longer the habitat for the Amur tiger in 1950-1960, and that the Amur tiger was only found in the mountain forest of Yanbian and Tonghua cities of Jilin province, and some parts of Mudan and Songhua river in Heilongjiang province in 1975-1976, shown in an investigation carried out by the Ministry of Forestry on Rare Wild Animals in China. The ensuing studies revealed that the Amur tiger distribution range compressed toward the east and north and became increasingly fragmented, and that Only 16-20 Amur tigers were reported in 2003 and 2004 in northeastern China, with 9-11 in Heilongjiang province in several isolated habitat patches. Figure 1 shows 779 occurrences of the Amur tiger by camera trapping in 41,200 km<sup>2</sup> of its habitat area in northeastern China during 1995-2015.



Over 95 per cent of the Amur tiger in the wild inhabit in the Russian Far-East now. From the early 19<sup>th</sup> Century to the late 1930s, intensive and unregulated hunting resulted in a huge decline in the total population and led to range fragmentation, including in its main habitat in the Sikhote-Alin area in 1940s.

The successful implementation of a blanket ban on the Amur tiger hunting introduced in 1947 stabilized its population and reversed the trend during mid-1960s and mid-1980s in all areas of south Amur River. The population recolonized in almost all of the suitable habitats within its former range and consolidated into one unified population in the Sikhote-Alin area. The population began to settle in the northern Sikhote-Alin where as a possible expansion in the early 1990s, and such recolonization over the larger part of its range remain stabilized since then. Censuses conducted respectively in 1996 and 2005 indicated that the population of the Amur tiger occurred in all forest areas within its habitat range and that the Sikhote-Alin area recovered and is no longer fragmented (Figure 2).

In terms of the monitoring methodology, the number of Amur tigers in the Russian Federation has been recorded in a special register following to the Methodological Recommendations for Conducting and Organization for the Amur Tiger Census in the Russian Federation as approved by the Ministry of Nature Resources in 2005, so as to ensure data credibility and reliability for long-term monitoring at federal and regional levels over in both selected areas and for full census every 10 years.

Following to the change of population and distributions observed in China and the Russian Federation, the habitat range of the Amur Tiger has extended to northwest since 2000, and currently spread from the southwestern Primorye into the neighboring provinces of Jilin and Heilongjiang provinces in China. According to researches focusing on the western and southwestern parts of the Amur tiger's range over the past years, the movement of the Amur tiger between the mountainous forests in China and the Russian Federation has become regular. The protection of the southwestern and western populations of the Amur tiger and wild ungulates (i.e. prey), along with their habitats, therefore requires special attention.

## Amur leopard

The Amur leopard was well distributed across what was known as the Ussuri Krai, nowadays a combined territory of Primorsky and Khabarovsk Krais in the Russian Federation prior to the active economic development in the 19<sup>th</sup> Century. Leopard sightings occurred, amongst others, on the left bank of the Amur River in the region of the Lesser Khingan Mountains. Outside the Russian Federation, the historical habitat for the Amur leopard includes Jilin and Heilongjiang provinces in northeastern China and the Korean Peninsula.

The population of the Amur leopard was substantially decreased from the beginning of the 20<sup>th</sup> century. Industrial development, its preys and habitat destructions led to the direct extinction of a portion of the leopard population along with habitat fragmentation. Expansion in agricultural development and population increasingly interrupted the access of the Amur leopard to the open habitat regions. As a result, the current number of its wild population is very small, estimated to be less than 60 in total. Most of them inhabit in the southwest of Primorsky Krai with its northern boundary at the Razdolnaya River, the only region where the Amur leopard can still be found in the Russian Federation, and in Hunchun Nature Reserve in Jilin province, its only remaining habitat in China. Despite no single leopard sighting in the Republic of Korea since 1969, there is a likelihood of a remaining leopard population in the DPRK within the largely inaccessible northern region of the country that borders with China (see the following figures of the distribution of the Amur leopard).



The analysis conducted by Feline Research Center of Chinese State Forestry Administration (FRC-SFA) suggested the following features to be considered for more effective conservation of the Amur leopard:

- (1) prey presence facilitates population distribution;
- (2) population density of the Amur leopard is in inverse proportion to the Amur tiger occurrence; and
- (3) the largest habitat patch in China (Area 1 in Figure 5) to be connected with the current range in the Russian Federation.





# 2. Biological characteristics and reproduction

Tigers evolved in tropical Asia and gradually spread northwards. The Amur tiger, a separate subspecies, has settled in North-East Asia near the northern limit of the species' range where it might experience extreme environmental conditions. The Amur tiger is one of the largest subspecies of tiger, only the Bengal tiger second to it in terms of size and weight. A male Amur tiger can measure up to 220 cm in body length while a female varies in body length from 165 to 182 cm. The heaviest tiger recorded in the wild was 250 kg, but the average weight of adult males is between 165 and 180 kg, and females is about 120 kg. Compared to other tiger subspecies, the Amur tiger has dense and relatively long hair.

Meanwhile, it is hypothesized that the modern leopard species (*Panthera pardus*) appeared and developed in Africa about 470-825 thousand years ago and dispersed into Asia 170-300 thousand years ago. Among them, the species that spread into the Russian Far-East evolved to a new subspecies, the Amur leopard, which is one of the nine leopard subspecies. The Amur leopard is characterized by lightly-colored fur and very large black rosettes or spots over the surface of the entire body. The fur coat on the body and the tail can be up to 7 cm long and the winter fur is very long and thick which allows it to survive in the harsh climate. The Amur leopard has longer extremities than other subspecies which allows them to move through deep snow cover. Its body size ranges up to 136 cm in length for males and 112 cm for females, and adult males weigh up to 70 kg and a female up to 50 kg except rare cases.

The lifespan of the Amur tiger is usually no longer than 20 years and 12-15 years for the wild Amur leopard. The tiger can breed when it reaches 3.5 to 4 years old and pregnancy normally lasts 95 to 120 days. Cubs can be born during any season but mostly in summer. Meanwhile, sexual maturity of the Amur leopard is reached at 2.5 to 3 years after birth, and its gestation period is around 90 to 95 days. Female Amur leopard is also able to breed throughout the entire year, but mostly in spring. New born cubs of the Amur tiger and leopard remain in the den for the first 1 or 2 months and how well the female selects its den site often determines how safe the cubs and how successful the subsequent breeding will be. Dens tend to be located in steep cliff areas where it is highly inaccessible to humans and natural enemies. The family disperses when the cubs reach at average 18.8 months old (Amur tiger) and 16 months old (Amur leopard), and males usually move longer distances to explore new habitats. However, such dispersion is now limited due to a lack of appropriate habitats.

# 3. Habitat requirements

The Amur tiger prefers cedar pine, broadleaf and oak forest, and, to a lesser extent, broadleaf and riparian forests, while the Amur leopard inhabits in areas of mixed coniferous broadleaf forests which starts from the coastal area and extends up to 600 to 800 meters above sea level, particularly hill areas and lower-mountain terrains with abundant exposed cliffs. Tigers show relatively low biotope selectivity which brings a contiguous spatial distribution throughout highly variable forest mosaics, whereas leopards are quite conservative in their territory choice and tend to use the same paths, passages and dens. Therefore, any kinds of habitat-changing activities such as construction of roads or dwellings make substantial impact on habitats of the Amur leopard, and often lead to the loss of their inhabited territories.

Both the Amur tiger and the Amur leopard are extremely territorial and exhibit a solitary behavior within the area with their scent marking, except for the period when females nourish their cubs. Upon radio telemetry techniques, the average-sized territory for a male tiger is 1,380 km<sup>2</sup>, while that for a female is 400 km<sup>2</sup>; and the territory of an adult male leopard ranges up to 300 km<sup>2</sup> while the range for females is close to 100 km<sup>2</sup>. Moreover, the range of their territory could vary depending on a number of factors, such as population density of wild ungulates and the presence or absence of Amur tigers or leopards in the same region. It is important to note that the overlapping of several individual territories and that a territory of a male often includes female territories.

# 4. Diet and predatory behavior

Although the Amur tiger feeds on a wide range of species, it prefers ungulates whose size equal or exceed its own, such as wild boar, red deer, roe deer and sika deer. The proportion of these prey species in the tiger's diet depends on their population density. For instance, where there is no snow, the Himalayan and brown bear significantly contribute to the tiger's diet, so do badger and raccoon dog. In fact, the Amur tiger can easily switch its prey and has a great ability to survive in different habitats. The Amur leopard also has a long list of prey including all vertebrates inhabiting the same ecosystems. Its most important food source is ungulates, but the prey composition and diversity changes depending on factors such as habitat conditions and weather.

The Amur tiger requires between 50 and 70 large ungulates per year to satisfy its energy requirements, along with other smaller prey items depending on its geographical location. Adequate number of large ungulates within the tiger range is between 400 and 500 individuals, but unfortunately such density does not occur anywhere. The annual consumption of large ungulates by an adult Amur leopard is approximately 25 to 30 specimens, and similarly the rest of the nutrition intake for the Amur tiger is from smaller vertebrates such as dogs, small cattle or sika deer. According to the monitoring data, the number of all prey species has been declining due to degradation of natural habitats, climate change, and a shortage of natural prey. This can cause the big cats to enter human settlements for supplementing its diet by hunting livestock, which may lead to conflict between human and the Amur tiger and leopard, possibly, causing extermination of the species. Therefore, proactive measures to stabilize and significantly increase the number of prey population are required to meet the needs of and conservation for the concerned two species.

# 5. Big cats-human interaction

The Amur tiger and leopard can tolerate proximity to humans and are not that aggressive compared to other big cat species. As they enjoy secluded lifestyle and their habitats are mostly located in mountainous areas, big cats-human encounters rarely occur in the wild. The Amur tiger and leopard usually try to avoid eye contact and exit the area, not showing any signs of aggression. Nonetheless, potential threat does exist, as they become aggressive when chasing food or protecting cubs. So far, no case has been reported that the Amur leopard attacked human throughout the entire period of the settlement in the southern region of the Russian Far-East, but that 16 fatal cases by the Amur tiger have been recorded in the Russian Federation in the last 40 years and 19 cases of tiger attack causing 2 deaths and 12 injuries were recorded between 2001 and 2010.

The most common type of conflict happens when the Amur tiger and leopard attack pets or livestock near human dwellings or on pastures. Poaching and traffic collision are also key conflicts. As the population analysis of the Amur tiger and leopard indicated anthropogenic impact as the most influential cause for the population dynamics, proactive and precautionary measures to provide favorable natural conditions, proper guidance to residents on how to behave within the predator's range, and introduction of new hunting regulations are required to ensure a sustained and peaceful coexistence with minimized conflict and negative consequences.

## 6. Conservation efforts

To establish an effective conservation strategy for the survival of the Amur tiger and leopard in the wild, their entire population needs to be exhaustively studied in the all range countries, taking into account the threatening factors and the impact mechanisms. The following factors should be considered:

- (1) biotic factors, such as influences by other carnivores and changes in prey population;
- (2) abiotic factors, namely changes that occur in the ecosystem, most importantly the impact of climate change on the depth of snow cover; and
- (3) anthropogenic factors, namely human activities.

Among these three categories, anthropogenic factors show the strongest and direct impact on the concerned species from poaching, accidental/necessitated killing, mining, timber harvesting and building of infrastructures in the habitat. Furthermore, the indirect influences include human population increase, pollution and irrational use of wildlife resources.

In this regard, international and (sub-)regional collaboration is crucial to protect the big cat species, conserve their habitats, and create transboundary corridors by sharing information. One example of information sharing is the joint survey in Jilin Province of China which was conducted by experts from China, the Russian Federation and the United States of America. The scientific and systemic aspect of the joint study opened a new era of species monitoring on Changbai mountains. Since then, extensive international cooperation activities such as snow-tracking, technical training and camera monitoring have been conducted in Jilin and Heilongjiang Provinces. In 2002, a two-year study on the prey population dynamics trends of Wandashan Amur tiger was conducted with the subvention of American Tiger and Rhino Fund, and an expert working group on the Amur tiger and leopard was established between Jilin Province and Primorsky Krai in 2012 followed by annual exchanges. Table 1 shows key international agreements on the conservation and re-establishment of rare and endangered species including the Amur tiger and leopard.

Convention on Biological Diversity	CBD provides for the conservation of biological resources both <i>in situ</i> and <i>ex situ</i> , as well as for their sustainable use. • China: ratification on 5 January 1993 • Russian Federation: ratification 5 April 1995	
Convention on International Trade in rare and endangered species	<ul><li>CITES provides for strict regulation on the trades of rare species and their parts for commercial purposes.</li><li>The Amur tiger and leopard are listed on CITES Appendix 1, prohibiting all commercial trade.</li></ul>	
IUCN Red Data Book	<ul> <li>Amur tiger listed as Endangered (EN)</li> <li>Amur leopard listed as Critically Endangered (CR)</li> </ul>	

#### Table 1. International agreements on species conservation

In addition to the international and (sub-)regional efforts, China and the Russian Federation have established national regulations and laws, and nature reserves or national parks to protect the two species and their habitats.

#### China

Since 1980, China has established several nature reserves for two big cat species in northeastern China to protect their habitats and recover potential habitats, followed by related scientific research including monitoring in the wild population assessment, habitat evaluation, breeding and wildness training research for captive population. The Circular of the State Council on Banning the Trade of Rhinoceros Horns and Tiger Bones stipulated by the State Council of China in 1993 prohibits the import, export, selling, purchasing, transporting, carrying and sending by post of rhinoceros horn and tiger bones in China, and that in 2011 the State Forestry Administration issued a Wild Tiger Recovery Plan to place greater emphasis on the need of strengthening the network of nature reserves and local protection stations.

Following to the amendment of the Wild Animal Conversation Law of China in the past decade, the Amur tiger and leopard remain to be listed as the first-class national protected animals under the law authorized by the Standing Committee of the National People's Congress of China in 2016, and that the Amur tiger and leopard and their habitats are entitled to protection at all levels in the entire territory of China. Table 2 shows the nature reserves and scientific research on the Amur Tiger and Leopard in China over the past decades.

#### Table 2. List of nature reserves and scientific research on the Amur tiger and leopard in China

	Heilongjiang Qixing Lazi Nature Reserve (1980) – the first Amur tiger nature reserve of China
Nature reserves and national parks	Jilin Hunchun Nature Reserve (2001)
	Jilin Wangqing Nature Reserve (2011)
	Heilongjiang Laoyeling Amur Tigers Nature reserve (2011)
	Heilongjiang Wanda Mountain Amur Tiger Nature Reserve (2012)
	Amur Tiger and Leopard National Park (2016)
	Survey methodologies, statistics on nature reserves and their application by Forestry Department of Heilongjiang Province (1974-1975)
	Aerial and ground surveys to investigate the Amur tiger and leopard habitats in the Northeast China, the first time ever as a special investigation in China (1984-1986)
	Establishment of the Feline Research Center of Chinese State Forestry Administration for big cat conservation research at the national level (December 2011)
Scientific research	Project on "Individual Identification Technology of the snow footprint of Amur tiger" by Northeast Forestry University and WWF (2012)
	Study on artificial propagation technique of the Amur tiger to improve its fecundity in the Zhenzhou Song Harbin zoo
	Hengdaohezi Feline Breeding Center became as the world's largest artificial breeding population of the Amur tiger (establishment in 1986)
	Great progress on genealogy record of the Amur tiger by the Harbin Amur tiger park, which also trailed reintroduction of the species to the wild

One of the most noticeable progress took place recently in China is the Amur Tiger and Leopard National Park covering about 15,000 km<sup>2</sup> in Jilin and Heilongjiang provinces. Officially inaugurated in August 2017, the construction of the National Park is scheduled to be completed in 2020 aimed to providing stable habitats with rich environmental resources for Amur tiger and leopard.

# The Russian Federation

Most populations of the Amur tiger and leopard inhabit within the Russian territory, which made the country carry the main privilege and responsibility for the conservation of the concerned species in the wild. Several laws and by-laws regulating the conservation and use of animal species and their habitats have been developed in the past decades in the Russian Federation, for example:

- Federal Law on Specially Protected Natural Areas (14 March 1995, №. 33-F3)
- Federal Law on Animal Species (24 April 1995, №. 52-F3)
- Federal Law on Environmental Protection (10 January 2002, №. 7-F3)
- Forest Code of Russian Federation (8 November 2006, №. 200-FZ)
- Federal Law "On Hunting and Hunting Resources and Legal amendments in Federal Laws" (24 July 2009, №. 209-F3)
- Criminal Code of the Russian Federation (with changes made on 2 July 2013, №. 150-F3)
- Government decree, "Affirmation of the list of the most valuable wild fauna and aquatic biological resources that belong to species listed in the Red Data Book of the Russian Federation and protected by agreements" (See Article 226.1 and Article 258.1 of Criminal Law, 31 October 2013, № 978)

The Federal Law on Animal Species is particularly important for animal conservation. Covering animal species and their habitats, the Federal Law states that wild animal species within the Russian Federation are the property of the State and includes a list of measures to be carried out to conserve habitats of wild animals. Certain types of economic development and activities can be banned or restricted in areas necessary for the species to complete the life cycles of such as breeding, feeding, and resting. The Law has also identified the responsible agencies for animal conservation and habitat rehabilitation. Furthermore, it places an obligatory requirement to conduct environmental impact assessment prior to any development decisions that may affect animal species and their habitats. It also explicitly states that rare and endangered animal species should be listed in the Russian Red Data Book and the respective regional Red Data Books. Any use or trade of those listed animal species is only legally allowed when the required permission after environmental impact assessment is granted. The Amur tiger is listed as Category 2 subspecies and the Amur leopard as Category 1 subspecies in the Russian Red Data Book. However, the effectiveness of these generally well-developed systems is hindered by low efficiency in law enforcement and, in some areas, by deficiencies of legal instruments or regulations.

# **NEASPEC** project

The objective of the NEASPEC study on *the Transborder Movement of Amur Tigers and Amur Leopards Using Camera Trapping and Molecular Genetic Analysis* is to strengthen the scientific understanding on the Amur tiger and leopard and their habitat conditions, based on which to provide policy recommendations to policy-makers for more efficient conservation and improving current or establishing new transboundary ecological corridors. The project collected data from camera trapping and molecular genetic analysis of the concerned species, and shared data for joint analysis carried out by national focal points in China (Feline Research Center of the State Forestry Administration, hereinafter "FRC") and the Russian Federation (WWF-Russia, and "Land of the Leopard" National Park, hereinafter "LLNP"). The timeline below shows the key milestones of the project.



### Table 3. Institutional arrangement of the Project

Project co	mponents	Leading agency	Supporting agencies
		Land of the Leopard National Park (Russia)	
Camera trapping		Wildlife Research Institute of Heilongjiang Province (China)	Hunchun, Wangqing and Laoyeling National Nature Reserves (China)
Molecular ge	netic analysis	Feline Research Center (China)	
Sample collection Joint field study		Land of the Leopard (Russia)	Far Eastern Branch Russian Academy of Sciences
		FRC (China)	Hunchun and Wangqing National Nature Reserve
	DNA	IBSS (Russia)	
	extraction	FRC (China)	
	Final analysis and policy formulation WWF Russia		Russia

# 1. Camera trapping

Camera trapping is a very effective and non-invasive sampling method, especially for rare and elusive animals such as the Amur tiger and leopard. The range of issues that can be studied with camera trapping data includes abundance and density, sex and age structure of the population, spatial distribution, territorial use, juvenile dispersion and individual behavior. Therefore, as an effective survey methodology, camera trapping has become a routine procedure of nature conservation in China and the Russian Federation. In China, camera trapping has been carried out in nature reserves such as Hunchun and Wangqing in the last 10 years. While the survey scale applied in China was relatively limited, the Russian Federation has extensively conducted camera trapping since 2002 in Nezhinskoe Hunting Lease and the southwest of Primorsky Krai and has identified 17 individual Amur tigers and 41 individual Amur leopards.

Individual identification of the concerned species and its head count by camera trapping were estimated upon its unique pattern of spots and stripes on the skin, which are as unique ID features as human fingerprints (Figure 6).



### Figure 6. Unique Amur leopard pattern of spots

As the animal's pattern is asymmetrical on the left and right side of the body (Figure 7), two camera traps facing each other were installed to capture both sides of the species simultaneously.



#### Figure 7. Asymmetrical spot pattern on the left and right side of the same Amur leopard

During the project, FRC installed 634 camera traps in 317 points and LLNP installed 314 camera traps in 157 points respectively during 2013 and 2015 that in total covered approximately 4,740 km<sup>2</sup> to collect images (Figure 8), involving 21 researchers from China and 10 researchers from the Russian Federation.

Camera trapping carried out in the Russian Federation was supported through the Federal budget allocated for the LLNP, with further financial support from "Far Eastern Leopards", an autonomous non-profit organization, the Russian Geographical Society and NEASPEC.



#### Figure 8. Camera traps installed in China and the Russian Federation

After signing of the agreement on joint analysis, two representatives from FRC visited Vladivostok in 2016 and analyzed camera trapping data with experts from LLNP. FRC and LLNP provided at least one photo of left and/or right side of each Amur tiger and Amur leopard captured during the period. For individual identification by spots and line patterns, the following methodologies were applied:

- Manual method: a visual comparison of spots or line patterns from different photos by a specialist without using any software; and
- Extract-Compare software: a computer software that captures image of spots or lines from each animal's side and applies a standard algorithm to differentiate individuals by their patterns (see Figure 9)



#### Figure 9. Consistent individual identification of leopard specimens in Extract-Compare software

### Amur tiger

FRC provided 27 and LLNP provided 51 Amur tiger individuals' images respectively. Some images included both sides (left and right; LR, Annex 1), whereas others showed either the left side (L, Annex 1) or the right side (R, Annex 1). The following criteria were applied to calculate the number of individual Amur tiger and Amur leopard:

- (1) Adults and cubs were analyzed separately.
  - a. If an individual was captured as a cub and appeared later alone as an adult, the individual was considered as an adult.
  - b. If a cub was captured with its mother and never captured again as an adult, it was considered as one individual cub.
- (2) The minimal number of individuals was the sum of individuals captured from both sides (LR) and "bigger" number of L- or R-individuals, as some individuals were captured from only one side (left or right). The minimal number of captured adult individuals was separately calculated upon their sex.

Annex 1 represents the combined set of date from this joint analysis, where the detailed information on individual matches between FRC and LLNP IDs.

	Adult	Cub
FRC	19 LR; 3 L; 1 R (= 22 adults)	1 L; 3 R (= 4 cubs)
LLNP	37 LR; 5 L; 2 R (= 42 adults)	1 LR; 6 L (= 7 cubs)

#### Table 4. Summary of joint analysis on Amur tiger images

The joint analysis based upon the above calculation indicated that at least 45 adult tigers and 10 cubs (55 in total) were camera-captured in China and/or the Russian Federation in 2013 to 2015. Out of 45 adult tigers (20 females, 15 males and 10 unknown), 19 Amur tiger individuals were found in both countries, i.e. 8 females, 8 males and 3 unknowns crossed the border. In the meantime, 3 Amur tigers in China and 24 Amur tigers in the Russian Federation never crossed the border. Details are listed below in Table 5.

#### Table 5. Minimal number of adult Amur tiger individuals captured by camera traps in 2013-2015

	Total	Female	Male	Unknown
China and/or Russia	45	20	15	10
China	22	9	8	5
Russian Federation	42	18	15	9
Observed in both countries	19	8	8	3
China only	3	1	0	2
Russian Federation only	24	10	7	7

\*Note: The number does not represent the current number of the Amur tiger population in wild, as (a) the study area did not cover the entire habitat range of the species and (b) some individuals which were camera captured in 2013 or 2014 did not appear in 2015.

During the three-year cameral trapping investigation, 6 breeding females in total were captured, including one (CT5 or T10) in both countries (Table 6). Cubs will become a new member of Amur tiger population if it shows up as an adult again.

#### Table 6. Reproductive Indexes of Amur Tiger Captured by Camera Traps

	2013	2014	2015	Females with cubs
China	0	5	0	CT3, CT18, CT5(=T10)
Russian Federation	0	12	1	T1, T10(=CT5), T7, T21

## Amur leopard

24 individual Amur leopard images were provided by FRC and 95 were identified by LLNP respectively. The detailed information is indicated below in Table 7.

#### Table 7. Summary of joint analysis on Amur leopard images

	Adult	Cub
FRC	19 LR; 5 R or L (= 23 adults)	1 cub
LLNP	81 LR; 14 R or L (= 82 adults)	13 cubs

Following to the same methodologies and criteria applied in the Amur tiger analysis, 105 Amur leopard individuals, including 91 adults and 14 cubs, were distinguished in the joint study. A combined dataset from the joint analysis is presented in Annex 2 of the report, that also includes detailed information on individual matches between FRC and LLNP IDs.

The joint analysis reveals that at least 89 adult leopards were camera-captured in China and/or the Russian Federation in 2013-2015, including 41 females, 37 males and 11 unknowns. 15 out of 89 adult leopards were found in both countries, including 8 females and 7 males, and the other 8 Amur leopards camera captured in China and 66 in the Russian Federation were not found crossing the border. Table 8 summarized the details.

# Table 8. Minimal number of adult Amur leopard individuals captured by camera traps in 2013-2015

	Total	Female	Male	Unknow sex	
China and/or Russia	89	89 41		11	
China	23	9	10	4	
<b>Russian Federation</b>	81	40	34	7	
Observed in both countries	15	8	7	0	
China only	8	1	3	4	
Russian Federation only	66	32	27	7	

\*Note: The number does not represent the current number of the Amur leopard population in wild, as (a) the study area did not cover the entire habitat range of the species and (a) some individuals which were camera captured in 2013 or 2014 did not appear in 2015.

10 breeding females were captured in both territories during the three-year investigation (see Table 9). At the end of 2013 and in the beginning of 2014, one female Amur leopard was captured with two cubs in China, and one of the cubs identified in China (leo9=leo28) was not captured in the Russian Federation. As for the parallel camera trapping in the Russian Federation, at least 9 cubs were born during the research in 2013 to 2015. According to the current research, 2 out of 9 cubs moved to China but follow-up study is required to understand how many cubs will become new members of the Amur leopard population.

	2013	2014	2015	Females with cubs
China	2	2	0	Leo 4
Russian Federation	6	9	7	Leo 1F, Leo 5F, Leo 7F, Leo 16F, Leo 23F, Leo 39F, Leo 55F, Leo 66F, Leo 89F

#### Table 9. Reproductive indexes of Amur leopard captured by camera traps

#### **Cross-border movement**

Analysis on cross-border movement of the Amur tiger and leopard was one of the key highlights of this project, as such findings and the implication on transboundary ecological border could be significant in the context of inter-governmental cooperation for the conservation of concerned species. To identify their cross-border movements, several parameters were applied including the number of encounters in each country, the number of boundary crossing, and the recorded maximum distance of movement from the border (see Table 10 and Table 11).

### Amur tiger

Analysis on the cross-border movements of the Amur tiger shows that some Amur tigers actively crossed the border, and it would be fair to assume that they could be resident individuals with home ranges in both China and the Russian Federation. For example,  $CT1(=T_7F)$ ,  $CT2(=T_3M)$ ,  $CT4(=T_11M)$ ,  $CT7(=T_4M)$  and  $CT16(=T_30F)$  crossed the border at least three times during 2013-2014.

Young individuals, particularly females with cubs, also actively crossed the border from the Russian Federation to China. Also, Amur tigers spotted in China tended to move from the border much further than those in the Russian Federation. For instance,  $CT5(=T_10F)$  was caught with 4 cubs in the Russian Federation in the beginning of 2014 and was captured again in China with only 2 cubs ( $CT21(=T10\_cub2)$  and  $CT24(=T10\_cub1)$ ) in the end of 2014.  $CT24(=T10\_cub1)$  then was caught later alone in China (39.7 km from the border) and  $CT5(=T\_10F)$  was appeared alone in China 2 times in 2015 (36.3 km from the border). In the meantime,  $CT1(=T\_7F)$  was captured with 4 cubs in the Russian Federation, but only one of the cubs  $CT26(=T1\_cub3)$  was caught in China as an adult (30.6 km from the border). In case of  $CT10(=T\_26M)$  which was caught in the Russian Federation in March 2014 for the first time was captured in China 259.3 km away from the border and did not return to the Russian Federation until 2015. Other 11 Amur tigers crossed the border only once, including 9 of them moved from the Russian Federation to China, and the other 2 vice versa.

LL-ID	FRC-ID	Number of encounters			% of en	counters	Times crossing	Max distance moved from the border (km)	
		China	Russia	total	China	Russia	the border	China	Russia
T_7F	CT1	21	14	35	60	40	10	5	7.9
T_3M	CT2	8	12	20	40	60	4	1.9	17
T_12F	СТЗ	5	5	10	50	50	2	1.6	2.5
T_11M	CT4	4	13	17	23.5	76.5	3	2.7	5.7
T_10F	CT5	3	2	5	60	40	1	36.3	4.4
T_4M	CT7	8	3	11	72.7	27.3	5	14.7	3.8
T_8F	CT8	2	13	15	13.3	86.7	2	1.9	4.4
T_26M	CT10	9	2	11	81.8	18.2	1	259.3	18.9
T_29M	CT11	2	1	3	66.7	33.3	1	0.9	2.3
T_31F	CT12	8	1	9	88.9	11.1	1	1.6	2.3
T_33M	CT13	8	11	19	42.1	57.9	1	1.6	16.2
T_32M	CT15	1	1	2	50	50	1	0.7	4.4
T_30F	CT16	18	6	24	75	25	3	2.9	3.9
T_9F	CT17	2	7	9	22.2	77.8	1	4.8	4.4
T_13F	CT18	1	8	9	11.1	88.9	2	0.3	4.1
T10_cub2	CT21	1	1	2	50.0	50.0	1	36.3	2.7
T_43Un	CT23	1	3	4	25.0	75.0	2	0.9	17
T10_cub1	CT24	2	1	3	66.7	33.3	1	39.7	2.7
T1_cub3	CT26	1	1	2	50.0	50.0	1	30.6	15.9
T_49Un	CT28	1	1	2	50.0	50.0	1	90.3	2.3

#### Table 10. Cross-border movements of Amur tiger individuals captured by camera traps

### Amur leopard

Similar to the Amur tiger, the Amur leopard was also identified by the joint study of actively crossing the border, and that some could be resident individuals having their habitats both in China and the Russian Federation. For example, Leo29M(=leo3) crossed the border 9 times during 2013-2014, and was captured three times more in the Russian Federation than in China, which indicates that the biggest part of its home range might be situated with the Russian Federation. Leo52M(=leo1) also actively roamed between the two countries. Following its regular movement captured in China since 2012, it appeared 10 times in the camera trapping in the Russian Federation and also found crossing the Russian border 10 times in 2014. Therefore, one can assume that Leo52M(=leo1) is a resident in China and visited the Russian Federation in 2014 probably to search for a female or try to enlarge his home range. Some Amur leopards such as Leo63F(= leo11) and Leo89F(= leo14) crossed the border only once, as captured in China in 2012-2013 then seen in the Russian Federation since 2014.

Another interesting story could be about two Amur leopards which were born in the Russian Federation and then moved across the border to China. One of them was a young male Leo81M(=leo29) who was captured once in the Russian Federation in August 2014 and once in China in April 2015 almost 30 km from the border. A female cub Leo9F(=leo10) was born and captured with her mother in 2013 within the Russian territory. It then was captured alone in the beginning of 2014 by the same camera trap. However, it moved to China in 2015 some 24 km away from the border and was spotted 6 times but never captured in the Russian Federation again during the study. Such migration shows the dispersion of young individuals from the Russian Federation to China, and further research is needed so to provide further evidence whether they become residents in China.





# Table 11. Cross-boundary movements of Amur leopard individuals captured by camera traps during 2013-2015

LL-ID	FRC-ID	Number of encounters			% of en	counters	Times crossing the	Max distance moved from the border (km)	
		China	Russia	total	China	Russia	border	China	Russia
Leo 22M	Leo 25	2	71	73	2.7	97.3	4	0.64	8.7
Leo 52M	Leo 1	44	19	63	69.8	30.2	10	37.6	4.9
Leo 25M	Leo 24	1	41	42	2.4	97.6	2	1.9	7.9
Leo 29M	Leo 3	10	31	41	24.4	75.6	9	36.3	5.7
Leo 7F	Leo 26	1	30	31	3.2	96.8	2	7.1	15.5
Leo 24M	Leo 21	1	27	28	3.6	96.4	2	0.6	7.1
Leo 91M	Leo 12	10	5	15	66.7	33.3	5	9.3	5.4
Leo 26F	Leo 17	2	13	15	13.3	86.7	1	0.46	3.5
Leo 9F <sup>1)</sup>	Leo 10	6	5	11	54.5	45.5	1	23.7	4.9
Leo 63F	Leo 11	3	7	10	30	70	1	0.46	6.2
Leo 89F	Leo 14	3	6	9	33.3	66.7	1	21.7	2.8
Leo 54F	Leo 22	1	7	8	12.5	87.5	2	2.2	6.2
Leo 49F	Leo 7	2	5	7	28.6	71.4	1	6.8	27.7
Leo 13F	Leo 27	1	5	6	16.7	83.3	2	0.9	14.2
Leo 81M <sup>2)</sup>	Leo 29	1	1	2	50	50	1	29.7	7

1) and 2) Captured as a cub in the Russian Federation

# 2. Molecular genetic analysis

With the fast development of biotechnology, molecular genetic analysis has become an innovative and widely applicable methodology to precisely identify an individual and its characteristics including history, geographic distribution, genetic diversity, population structure, family tree and even disorders by the methods of genome sequencing and microsatellite analysis. To analysis these characteristics, samples like tissues, blood or fecal were obtained by non-invasive methods. As many of the traits above are difficult if impossible to obtain by conventional survey methods, researchers begin to apply this modern technology to species of interest, yet the entire process from collecting non-invasive samples in the field to interpreting laboratory data without errors requires careful treatment with advanced technology and equipment. Fortunately, scientists in North-East Asia have continuously developed analytical methodologies, including the world's first genome mapping of the Amur tiger in 2013 by the Republic of Korea, which has enabled more accurate genetic diversity monitoring with higher accuracy.

Molecular genetic analysis is mainly conducted in two steps, namely sample collection from the field and laboratory analysis. For the NEASPEC project, FRC and LLNP provided non-invasive samples (scat) which were collected from the field during 2013 and 2015. DNA extraction from samples for laboratory test was respectively conducted by FRC for samples from China, and Institute of Biology and Soil Science (IBSS) of the Russian Academy of Science for samples collected from the Russian Federation. The overall analysis was conducted by a joint analysis team including Meng Wang and Yao Ning from China, and Marina Igorevna Chaika and Valentin Yurievich Guskov from the Russian Federation.

### Sample collection

Non-invasive samples were collected along the tracks where Amur tigers and leopards leave their traces during the winter. As a starting point to attain reliable outcomes, samples should be collected very carefully with Ziploc and sterile gloves to avoid any damages particularly on the surface of the sample, and to package individually with a marked card to record their geographic coordinates of collection site and encode. To guarantee the quality of DNA, packaged samples need to be stored in the ice box filled with ice bag immediately in the field and then preserved in the freezer (-80°C ) until DNA extraction in the laboratory.



# DNA analysis

#### DNA extraction and amplification

After delivering collected samples to the laboratory, the entire DNA of each sample was extracted by commercial toolkit (QIAamp DNA Stool Mini Kit, Qiagen). Researchers followed the kit instruction to carry out the procedures step by step.



Extracted DNA samples were further analyzed to identify species and individuals by using specific primers for target species. In the winter of 2015, the joint analysis team visited Suo-Lin Luo's laboratory at Peking University of China to discuss on the microsatellites to be applied. As a result, researchers selected 8 microsatellite loci for the Amur tiger and 9 microsatellites loci for the leopard that had been presented in previous studies, including:

Specific primers for Amur tiger:

Pta-CbF (5'-TTTGGCTCCTTACTAGGGGTG-3')

Pta-CbR (5'-CCGTAAACAATAGCACAATCCCGATA-3')

• Specific primers for Amur leopard:

Ppo-CbF (5'-GTAAATTATGGCTGAATTATCCGG-3')

Ppo-CbR (5'-CATAACCGTGAACAATAATACGAC-3')

The microsatellite loci were amplified three times by Polymerase Chain Reaction (PCR) which was set up in a 20  $\mu$  system, containing 1 × PCR buffer, 50 mM of Tris-HCl (Ph 8.0), 25 mM of KCl, 0.1 mM of EDTA, 1 mM of dTT, 0.4 mM each of 4 dNTP (TOYOBO), 0.2  $\mu$ M each of forward and reverse primer, 0.4 U units of KOD FX Neo DNA polymerase (TOYOBO) and ~10 ng of total DNA. PCR amplification was performed on a Model 9700 Thermocycler (Perkin-Elmer) with the following procedure: (a) 1 cycle of denaturation at 94°C for 2 minutes, (b) 35 cycles at 94 °C for 15 seconds, (c) annealing at 60 °C for 30 seconds and 68 °C for 30 seconds, and (d) 1 more cycle at 68 °C for 20 minutes. Amplified DNA samples were further tested by 0.8% agarose gel electrophoresis (Amur tiger: 271 bp; and Amur leopard: 156 bp).



#### Individual identification

Multiplex PCR was conducted using the Qiagen Multiplex PCR Plus Kit (Qiagen) in a 10  $\mu$ l reaction system containing 5  $\mu$ l of 2×Multiplex PCR Mastermix, 1  $\mu$ l of 10×primer mix, and 10 ng of DNA for individual identification. PCR then was performed with Model 9700 Thermocycler (Perkin-Elmer) under the following conditions: (a) 1 cycle of denaturation at 95 °C for 5 minutes, (b) 35 cycles of denaturation and annealing at 95 °C for 30 seconds, 60 °C for 90 seconds and then 72 °C for 3 minutes, and (c) a final extension at 68 °C for 30 minutes. The size of microsatellite fragments was estimated by ABI3030 genetic analyzer with Liz 500 size standard as well as Gene Mapper V4.0 program. The sample only can be categorized as "homozygous" when the same peak repeats at least for 3 times and as "heterozygous" repeated at least twice.

# Figure 11. Multiplex PCR, a widely using molecular biology technique for amplification of multiple targets in a single PCR experiment



#### Sex identification

This exercise was performed following a similar approach with X and Y chromosome fragments from collected samples. The size of X and Y chromosome fragments were 205 bp and 156 bp, respectively. The following primers, specifically designed to both Amur tigers and Amur leopards, were applied. PCR operation carried out the same as the one for species identification mentioned above, except for the annealing temperature at 56  $^{\circ}$ C

• For X chromosome:

ZFX-PR(5'-TACCGAGCGATATAGCTCCAG-3')

ZFX-PF(5'-GTGTTCCTACGTTAAGCTATTG-3')

• For Y chromosome:

DBY7-PF(5'-CTCATGAAGCCCTATTTTTGGTTG-3')

DBY7-PR(5'-ACGGCGTCCGTATCTTCCA-3')



### Amur tiger

FRC collected 103 suspected feces of Amur tigers during 2013-2015, and LLNP collected 78 in 2015. By species identification, researchers found 93 and 65 positive samples for Amur tigers, respectively, and all of them went through amplification process using 8 microsatellite loci. Except those samples showed a low amplification rate, 22 samples for each side were successful amplified. Individual and sex identification was further conducted for only amplified samples, and 15 and 9 individual Amur tigers were identified, respectively. For Chinese side, 9 out of 15 individuals were detected once and the other 6 tigers were captured 2~3 times. Among 9 Amur tigers identified in the Russian Federation, 4 detected once and the other 5 for 2~4 times.

# Table 12. Minimal number of Amur tiger individuals captured by DNA analysis in China (2013-2015) and the Russian Federation (2015)

	Collected samples	Positive samples	Selected genotypes	Identified individuals	Gender
China	103	93	22	15	5 Female; 9 Male; 1 unknown
Russian Federation	78	65	18	9	5 Female; 4 Male

## Amur leopard

FRC collected 104 suspected feces of Amur leopards during 2013-2015, and LLNP collected 57 in 2015. Same methodologies were applied for the collected samples, and Table 13 represents the results.

# Table 13. Minimal number of Amur leopard individuals captured by DNA analysis in China (2013-2015) and the Russian Federation (2015)

	Collected samples	Positive samples	Selected genotypes	ldentified individuals	Gender
China	104	78	15	10	1 Female; 6 Male; 3 unknown
Russian Federation	86		23	17	2 Female; 13 Male

Among 10 individuals identified in China, 9 Amur leopards were detected once, and the other 1 for 6 times; and 13 out of 17 leopards identified in the Russian Federation were detected once, but others were detected for 2~3 times.

### **Cross-border movement**

Unfortunately, only one individual Amur tiger and one individual Amur leopard were detected in both China and the Russian Federation. Table 14 and Table 15 below summarize the results of the joint molecular genetic analysis.

# Table 14. Amur tigers and Amur leopards identified by molecular genetic analysis in China(only for positive samples, including 93 for Amur tigers and 78 for Amur leopards)

No.	Sample ID	Species	Individual ID	No. of crossborder movement	No.	Sample ID	Species	Individual ID	No. of crossborder movement
1	1	tiger			87	112	tiger		
2	2	tiger	1		88	113	tiger		
3	3	tiger			89	115	tiger		
4	4	tiger	2		90	116	tiger		
5	5	tiger			91	118	tiger		
6	6	tiger			92	121	leopard		
7	7	tiger			93	122	leopard		
8	8	tiger	3		94	124	leopard	7	
9	9	tiger			95	125	leopard		
10	10	tiger	4		96	126	leopard		
11	11	tiger	3		97	127	leopard		
12	12	tiger			98	150	tiger		
13	13	tiger			99	151	tiger		
14	14	tiger			100	152	tiger	7	
15	15	tiger			101	153	tiger		
16	16	tiger			102	154	tiger	8	
17	20	leopard			103	155	tiger	8	
18	21	leopard			104	156	tiger		
19	24	leopard			105	157	tiger		
20	28	leopard			106	158	tiger		
21	30	leopard	1		107	159	tiger	9	
22	31	tiger	5	1	108	161	leopard		
23	33	leopard			109	162	leopard		
24	34	leopard			110	163	leopard		
25	35	leopard			111	164	leopard		
26	36	leopard	2		112	165	leopard		
27	37	tiger			113	166	leopard		
28	38	tiger			114	167	leopard		
29	39	tiger			115	168	leopard		
30	40	tiger			116	169	leopard		
31	41	tiger			117	171	tiger		
32	42	tiger			118	172	tiger	10	
33	43	tiger			119	173	tiger	10	
34	44	tiger			120	174	tiger		
35	45	tiger			121	175	tiger		
36	46	tiger			122	176	tiger		
37	49	tiger			123	177	tiger		
38	50	tiger			124	178	tiger		
39	51	tiger	6		125	179	tiger		
40	52	tiger			126	180	tiger		
41	53	tiger			127	181	tiger		
42	54	tiger			128	182	leopard		
43	55	tiger			129	183	tiger	2	
44	56	tiger			130	185	tiger		

No.	Sample ID	Species	Individual ID	No. of crossborder movement	No.	Sample ID	Species	Individual ID	No. of crossborder movement
45	57	tiger			131	186	tiger		
46	58	tiger			132	187	tiger		
47	59	tiger			133	189	tiger		
48	60	tiger			134	190	leopard		
49	61	tiger			135	191	leopard		
50	62	tiger			136	192	leopard		
51	63	leopard	3		137	193	leopard		
52	64	leopard			138	195	leopard		
53	65	leopard			139	196	leopard		
54	66	leopard			140	197	leopard		
55	67	leopard			141	198	tiger		
56	68	leopard			142	199	leopard		
57	69	leopard			143	200	leopard		
58	70	leopard			144	201	tiger		
59	71	leopard			145	202	tiger		
60	73	leopard			146	203	tiger		
61	74	leopard	3		147	204	tiger	11	
62	75	leopard			148	205	tiger	11	
63	76	leopard			149	206	tiger		
64	77	leopard			150	207	tiger		
65	79	leopard			151	208	tiger		
66	80	leopard			152	209	tiger	12	
67	81	leopard			153	210	tiger		
68	82	leopard			154	211	tiger		
69	83	leopard			155	214	leopard	8	1
70	84	leopard			156	215	leopard	9	
71	85	leopard			157	216	leopard		
72	87	leopard			158	217	leopard		
73	88	leopard	4		159	218	leopard	10	
74	89	leopard			160	219	leopard		
75	90	leopard			161	221	leopard		
76	94	leopard	5		162	223	leopard		
77	96	leopard			163	224	leopard		
78	97	leopard			164	225	tiger		
79	98	leopard	3		165	226	tiger	13	
80	99	leopard	3		166	227	tiger		
81	101	leopard	3		167	228	tiger	14	
82	102	leopard			168	231	tiger	15	
83	103	tiger			169	232	tiger	15	
84	106	leopard			170	233	tiger	15	
85	107	leopard	3		171	234	tiger		
86	108	leopard	6						

# Table 15. Amur tigers and Amur leopards identified by molecular genetic analysis in the Russian Federation

(only for positive samples, including 65 for Amur tigers and 56 for Amur leopards)

No.	Sample ID	Species	Individual ID	No. of cross- border movement	No.	Sample ID	Species	Individual ID	No. of cross- border movement
1	862	leopard	1		62	924	tiger		
2	863	leopard	2		63	931	tiger		
3	864	leopard	3		64	933	tiger		
4	865	leopard			65	936	tiger	3	1
5	866	leopard	2		66	952	tiger		
6	867	leopard	2		67	953	tiger		
7	873	leopard	4		68	954	leopard		
8	880	leopard	5		69	955	tiger	1	
9	881	tiger			70	956	tiger	6	
10	884	leopard	6		71	959	tiger		
11	885	tiger			72	961	tiger		
12	887	leopard	7		73	962	tiger		
13	893	leopard	6		74	966	tiger		
14	894	leopard			75	969	tiger	7	
15	896	leopard	8		76	972	tiger	8	
16	907	leopard			77	973	tiger		
17	908	leopard	5		78	974	tiger	9	
18	909	leopard			79	975	tiger		
19	911	leopard	9		80	976	tiger		
20	914	leopard			81	980	tiger		
21	917	leopard			82	982	tiger		
22	919	leopard	6		83	984	tiger		
23	921	leopard			84	1000	tiger		
24	922	leopard			85	1019	tiger	2	
25	923	leopard	10		86	1021	tiger		
26	928	leopard	9		87	1023	tiger		
27	930	leopard			88	1025	tiger	2	
28	940	leopard			89	1029	tiger		
29	983	leopard	11		90	1034	tiger		
30	986	leopard			91	1037	tiger		
31	989	leopard	12		92	1048	tiger		
32	990	leopard	13		93	899	leopard		
33	995	leopard			94	901	tiger		
34	996	leopard			95	906	tiger		
35	997	leopard			96	910	tiger		
36	998	leopard			97	912	leopard		
37	1004	leopard			98	913	leopard		
38	1005	leopard			99	932	tiger	1	
39	1009	leopard			100	934	tiger		
40	1010	leopard	14		101	935	tiger	3	1
41	1012	leopard			102	937	tiger		
42	1015	leopard			103	938	tiger		
43	1016	leopard			104	939	tiger		
No.	Sample ID	Species	Individual ID	No. of cross- border movement	No.	Sample ID	Species	Individual ID	No. of cross- border movement
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44	1022	leopard			105	943	leopard		
45	1027	leopard	15		106	944	tiger	4	
46	1028	leopard	16	1	107	946	tiger		
47	1032	leopard			108	947	tiger	3	1
48	1040	leopard			109	948	tiger	5	
49	1050	leopard			110	957	tiger		
50	870	tiger			111	958	tiger		
51	871	tiger			112	967	tiger	3	1
52	875	tiger			113	1001	tiger	5	
53	876	tiger			114	1008	tiger	4	
54	877	tiger			115	1024	tiger		
55	888	tiger			116	1043	leopard		
56	892	tiger			117	1045	tiger		
57	900	tiger			118	1046	leopard		
58	903	tiger			119	1047	leopard		
59	904	tiger	1		120	1052	tiger		
60	918	tiger	2		121	1053	leopard	17	
61	920	tiger							

#### Key implications of molecular genetic analysis

Upon completing the joint analysis under the NEASPEC project, it seems that molecular genetic analysis is less efficient method compared to camera trapping analysis in terms of identifying individuals and their cross-border movement given it shows a smaller number of identified individuals, which could be affected by the following factors:

- (1) The quality of samples collected falls short for analysis on DNA extraction and amplification. Among the total sample size, only 23.6% of Amur tiger samples collected from China and 27.6% from the Russian Federation were successfully amplified. The case for Amur leopard was slightly better that 19.2% samples collected from China and 41.1% of Russia were amplified.
- (2) The data analysis chose to apply strict principles to ensure accuracy. Non-polymorphic samples and those showed low amplification rate were excluded for analysis. In other words, only those samples successfully amplified the entire locus were used for data analysis, and that only when all genetic information or only one mismatch was identical were the samples categorized as "same individual".

In addition, as geographic coordinates were not recorded when collecting samples from the field in the Russian Federation, the analysis on the spatial distribution of two species across the border of China and the Russian Federation could not be carried out.

### **Conclusion and ways forward**

According to the results of the joint analysis carried out under the NEASPEC project, the research team concluded on the following findings with suggestions on future collaboration.

- (1) Comparing the two research methodologies applied in the project, camera trapping could be a better monitoring methodology than molecular genetic analysis for understanding the cross-border movement of the Amur tiger and leopard. 42% of Amur tigers and 17% of Amur leopards captured by camera trapping in this study showed transborder movement, while molecular genetic analysis found only 1 Amur tiger and 1 Amur leopard crossing the border between China and the Russian Federation.
- (2) Although one of the goals of molecular genetic analysis was to estimate genetic diversity and cross-border movement of two species, it was not feasible to achieve that goal due to insufficient number of samples for lab analysis. For more detailed genetic studies, quality and expanded sample size should be collected in the field and shared among researchers to enable future studies on the biological features of the concerned species, so to support discussions and policy development on transboundary ecological corridors.
- (3) Detailed geographic coordinates of all collected samples need to be recorded and shared in order to indicate clearer cross-border movement of the target species and to identify the specific corridors across the Sino-Russian border.
- (4) Sino-Russian border fence or human disturbance between Changbai Mountains and the Land of Leopard National Park do not appear to be a serious obstacle for the movement of the Amur tiger and leopard. It is possible that Amur tigers and leopards may have used broken border fences as transborder corridors. Further investigation is needed on this.
- (5) High density of Amur tigers and leopards live in the core areas of the Sino-Russian border, and they frequently cross the border. Thus, well-developed joint Sino-Russian monitoring activities should continue for efficient conservation of the concerned species. It is strongly recommended to diffuse such dense population out of the core area.
- (6) More specifically for Amur tigers, it is urgent to avoid population collapse caused by disruptions such as disease outbreak particularly among smaller groups, and further research is needed with the attempt to connect or distribute between small population in Changbai Mountain and the big population in Sikhote-Alin.
- (7) The conservation of Amur leopards should be enhanced as its population is very small, and that more emphasis should be placed on expanding its habitat where possible so to avoid the territory competition with Amur tigers.

#### Suggestions

The conservation of the Amur tiger and Amur Leopard can only be assured by implementing a set of activities targeting the animal itself, its habitat and its food sources. These activities must consider the unique biological features of the subspecies' boreal existence as well as the lessons learnt from the past years. There are two main tasks necessary for conserving their populations:

- (1) To remove the causes of the decline in population number, and simultaneously
- (2) To minimize the negative impacts that lead to the contraction and degradation of their habitats.

Although the Russian Federation presently carries the main responsibility for the conservation of the Amur tiger and Amur leopard in the wild, the future of these two subspecies largely depends on the status and condition of populations and its habitat in neighboring countries, specifically in China and the Democratic People's Republic of Korea. Small populations of these big cats in border areas of China are apparently supplemented by individuals who cross over from the Russian Federation, and that their appearances in the northern parts of DPRK has also been recorded. Without joining the efforts among these neighboring countries, it will not be possible to assess the level of habitat degradation, to unleash the potential for restoring the natural range of Amur tigers and leopard, and to determine the size for their entire populations that could be sustained in the wild.

Further cross-border collaboration at the subregional level will help foster the exchange of information that drives research and enhance the capability of the region to conserve not only the subspecies but also the entire big cat species in all the range countries. Therefore, following areas for cooperation have been identified at both regional and global level:

- (1) Establishment of international transboundary protected areas for the conservation of the Amur tiger and Amur leopard.
- (2) Establishment of transboundary ecological corridors to stabilize or connect habitats of two species in range countries, for instance through international corridors connecting China, DPRK and the Russian Federation.
- (3) Joint activities such as (a) joint habitat assessment on human disturbance, (b) joint research on long-term human-Amur tiger/leopard conflicts to provide further understanding on adopting conservation measures and managing nature reserves and national parks more effectively, (c) joint monitoring to assess the entire population dynamics, and (d) joint research to establish a national Amur tiger and leopard park.
- (4) Coordination of research programs and technological cooperation among big cat experts from different countries, focusing on the development of a joint methodology for monitoring Amur tigers and leopards that can enable greater level of data compatibility and comparability and eventually strengthen the research capacity in the range countries and beyond.

- (5) Coordination on activities to stop illegal trades of products derived from illicit hunting of Amur tigers and leopards, especially on the collaboration between China and the Russian Federation. More specifically the customs services in the neighboring provinces of China and the Russian Federation should work together and exchange information on the cross-border movement of illegal animal products, as well as for the respective local institutions to exchange information on illegal international trading routes in both countries.
- (6) Participation in the Global Tiger Initiative (GTI)<sup>2</sup>, a platform for international collaboration launched by the World Bank in 2008, to coordinate planning activities in tiger conservation that requires targeted efforts among all tiger range countries.
- (7) Continuation of collaboration in the management of captive Amur tigers and leopards' populations within the European Endangered Species Programme (EEP), the European Association of Zoos and Aquaria (EAZA), and the North American Tiger Species Survival Plan (SSP) of the Association of Zoos and Aquariums (AZA).

<sup>2</sup> The main objectives of GTI are (1) to increase the effectiveness of conservation activities through the exchange of experience and information, (2) to improve the enforcement of conservation law through exchanging experience and international cooperation in combating the illegal cross-border trade in products derived from rare and endangered animal species, (3) to decrease the demand for tiger products by *inter alia* conducting public awareness campaigns amongst consumers in those countries where tiger products are being used in traditional medicine and where there is also a demand for tiger skins, (4) to raise the effectiveness of tiger habitat protection, (5) to develop incentives for supporting tiger conservation at the local level, and (6) to develop and improve innovative mechanisms for funding tiger conservation activities, e.g. developing mechanisms for joint funding of conservation projects by using carbon credits to compensate for carbon retention or by paying for environmental services.

### ANNEX

# Annex 1. Joint catalog of Amur tiger individuals captured by camera traps in China and the Russian Federation (2013-2015)

No.	ID-LL*	ID-FRC*	Sex	Captured in 2013	Captured in 2014	Captured in 2015	Side	Adult/Cub	The year when an individual was captured as a cub	Notes
1	T_1	-	F	Y	Y	Y	LR	adult		
2	T_2	-	F	Y	Y	Y	LR	adult		
3	T_3	CT 2	м	Y	Y	Y	LR	adult		
4	T_4	CT 7	м	Y	N	N	LR	adult		
5	T_5	-	м	Y	Y	N	LR	adult	died in 2014	
6	T_7	CT 1	F	Y	Y	Y	LR	adult		
7	T_8	CT 8	F	Y	Y	Y	LR	adult		
8	T_9	CT 17	F	Y	Y	N	LR	adult		
9	T_10	CT 5	F	N	Y	N	LR	adult		
10	T_11	CT 4	м	Y	Y	Y	LR	adult		
11	T_12	CT 3	F	Y	Y	N	LR	adult		
12	T_13	CT 18	F	Y	Y	N	LR	adult		
13	T_14	-	F	N	N	Y	R	adult		
14	T_16	-	м	Y	Y	Y	LR	adult		
15	T_17	-	F	Y	Ν	N	LR	adult		
16	T_18	-	м	Y	Y	Y	LR	adult		
17	T_19	-	UN	Y	N	Y	LR	adult		
18	T_20	-	F	Y	N	Ν	LR	adult		
19	T_21	-	F	Ν	Y	Y	LR	adult		
20	T_22	-	UN	Ν	Ν	Y	LR	cub	cub in 2015	cub of T_21F
21	T_23	-	F	N	Y	Y	LR	adult		
22	T_25	-	F	N	Y	Y	LR	adult		
23	T_26	CT 10	м	Ν	Y	N	LR	adult		
24	T_27	-	М	N	Y	Y	LR	adult		
25	T_29	CT 11	м	Ν	Y	N	LR	adult	cub in 2014	cub of T_7F
26	T_30	CT 16	F	Ν	Y	Y	LR	adult	cub in 2014	cub of T_7F
27	T_31	CT 12	F	Ν	Y	Y	LR	adult	cub in 2014	cub of T_7F
28	T_32	CT 15	М	Ν	Y	Y	LR	adult		
29	T_33	CT 13	М	Y	Y	Y	LR	adult		
30	T_34	-	UN	Ν	Y	N	L	adult		
31	T_35	-	М	Ν	Y	Ν	LR	adult		
32	T_36	-	UN	Ν	N	Y	LR	adult		
33	T_37	-	F	Ν	N	Y	LR	adult		
34	T_38	-	М	Ν	N	Y	LR	adult		
35	T_39	-	М	Ν	N	Y	LR	adult		
36	T_40	-	F	Ν	N	Y	LR	adult		
37	T_41	-	F	Ν	Ν	Y	LR	adult		

No.	ID-LL*	ID-FRC*	Sex	Captured in 2013	Captured in 2014	Captured in 2015	Side	Adult/Cub	The year when an individual was captured as a cub	Notes
38	T_42	-	UN	Ν	Ν	Y	LR	adult		
39	T_43	CT 23	М	Ν	Ν	Y	LR	adult		
40	T_44	-	UN	N	Ν	Y	R	adult		
41	T_47	-	UN	N	Ν	Y	L	adult		
42	T_48	-	UN	N	N	Y	L	adult	cub in 2014	
43	T_49	CT 28	UN	N	Ν	Y	LR	adult		
44	cub1 of T_1F	-	UN	Ν	Y	Ν	L	cub	cub in 2014	cub of T_1F
45	cub2 of T_1F	-	UN	Ν	Y	N	L	cub	cub in 2014	cub of T_1F
46	cub3 of T_1F	CT 26	UN	Ν	Y	Y	L	adult	cub in 2014	cub of T_1F
47	cub4 of T_1F	-	UN	Ν	Y	Y	L	cub	cub in 2014	cub of T_1F
48	cub1 of T_10F	CT 24	UN	Ν	Y	Y	L	adult	cub in 2014	cub of T_10F
49	cub2 of T_10F	CT 21	UN	Ν	Y	Y	L	cub	cub in 2014	cub of T_10F
50	cub3 of T_10F	-	UN	Ν	Y	Y	L	cub	cub in 2014	cub of T_10F
51	cub4 of T_10F	-	UN	Ν	Y	Y	L	cub	cub in 2014	cub of T_10F
52	-	CT6	UN	Ν	Y	Y	R	cub	cub in 2014	cub of CT 18F
53	-	CT9	UN	Ν	Y	Y	LR	adult		
54	-	CT14	UN	Ν	Y	Y	R	cub	cub in 2014	cub of CT 18F
55	-	CT19	UN	N	Y	Y	R	cub	cub in 2014	cub of CT 3F
56	-	CT22	F	N	N	Y	LR	adult		
57	-	CT25	UN	Y	Ν	N	R	adult		
58	-	CT27	UN	Ν	N	Y	L	adult		

ID-LL - unique number of tiger individual in the database of Land of the Leopard

ID-FRC - unique number of tiger individual in the database of Feline Research Center

F – Female / M – Male / UN – Unknown / Y – yes / N – no / L – left / R – right

## Annex 2. Joint catalog of Amur leopard individuals captured by camera traps in China and the Russian Federation (2013-2015)

No.	ID-LL*	ID- FRC*	Sex	Captured in 2013	Captured in 2014	Captured in 2015	Side	Adult/ Cub	The year when an individual was captured as a cub	Notes
1	Leo 52M	Leo 1	М	Y	Y	Y	LR	adult		
2	-	Leo 2	М	Y	Y	N	LR	adult		
3	Leo 29M	Leo 3	М	Y	Y	N	LR	adult		
4	-	Leo 4	F	Y	Y	N	LR	adult		
5	-	Leo 6	UN	Y	N	N	R	adult		
6	Leo 49F	Leo 7	F	N	Y	N	LR	adult		
7	-	Leo 8	UN	Y	Y	N	LR	cub	cub in 2013 and 2014	cub of Leo 4F
8	-	Leo 9 = leo18	UN	Y	Y	Y	LR	adult	cub in 2013 and 2014	cub of Leo 4F
9	Leo 9F	Leo 10	F	Y	Y	N	LR	adult	cub in 2013	cub of Leo 7F
10	Leo 63F	Leo 11	F	Y	Y	Y	LR	adult		
11	Leo 91M	Leo 12	м	Y	Y	Y	LR	adult		
12	Leo 89F	Leo 14	F	Y	Y	Y	LR	adult		
13	-	Leo 15	м	N	N	N	L	adult		Excluded from the analysis 2013-2015
14	-	Leo 16 = leo 19	м	Y	N	N	R	adult		
15	Leo 26F	Leo 17	F	Y	Y	Y	LR	adult		
16	-	Leo 20	UN	N	N	Y	R	adult		
17	Leo 24M	Leo 21	М	Y	Y	N	LR	adult		
18	Leo 54F	Leo 22	F	Y	Y	N	LR	adult		
19	-	Leo 23	М	N	N	Y	R	adult		
20	Leo 25M	Leo 24	м	Y	Y	Y	LR	adult		
21	Leo 22M	Leo 25	м	Y	Y	Y	LR	adult		
22	Leo 7F	Leo 26	F	Y	Y	Y	LR	adult		
23	Leo 13F	Leo 27	F	Y	Y	Y	LR	adult		
24	-	Leo 28	UN	N	N	Y	R	adult		
25	Leo 81Un	Leo 29	F	N	Y	Y	LR	adult	cub in 2014	Mother unknown
26	Leo 1F	-	F	Y	Y	N	LR	adult		
27	Leo 2M	-	м	Y	Y	Ν	LR	cub	cub in 2013, 2014	cub of Leo 1F
28	Leo 3M	-	М	Y	Y	N	LR	adult	cub in 2013, 2014	cub of Leo 1F
29	Leo 4F	-	F	Y	Y	Y	LR	adult	cub in 2013, 2014	cub of Leo 1F
30	Leo 5F	-	F	Y	Y	Y	LR	adult		
31	Leo 6Un	-	UN	Y	N	N	LR	cub	cub in 2013	cub of Leo 5F
32	Leo 8F	-	F	Y	Y	Y	LR	adult	cub in 2013	cub of Leo 7F
33	Leo 10M	-	М	Y	Y	Y	LR	adult		
34	Leo 11M	-	м	Y	Y	Y	LR	adult		Died by car accident in Oct 2015
35	Leo 12M	-	М	Y	Y	Y	LR	adult		
36	Leo 14M	-	М	Y	Y	Y	LR	adult		
37	Leo 15M	-	М	Y	Y	Y	LR	adult		
38	Leo 16F	-	F	Y	Y	Y	LR	adult		
39	Leo 17F	-	F	Y	Y	Ν	LR	adult		

No.	ID-LL*	ID- FRC*	Sex	Captured in 2013	Captured in 2014	Captured in 2015	Side	Adult/ Cub	The year when an individual was captured as a cub	Notes
40	Leo 18M	-	М	Y	Y	N	LR	adult		
41	Leo 19M	-	м	Y	Y	N	LR	adult		
42	Leo 20M	-	м	Y	Y	N	LR	adult		
43	Leo 21Un	-	UN	Y	Y	Y	LR	adult		
44	Leo 23F	-	F	Y	Y	Y	LR	adult		
45	Leo 27F	-	F	Y	Y	Y	LR	adult		
46	Leo 28M	-	М	Y	Y	N	LR	adult		
47	Leo 30M	-	М	Y	Y	Y	LR	adult		
48	Leo 31F	-	F	Y	Y	Ν	LR	adult		
49	Leo 32M	-	М	Y	Y	Y	LR	adult		
50	Leo 33F	-	F	Y	Y	Ν	LR	adult		
51	Leo 34M	-	М	Y	Y	Y	LR	adult		
52	Leo 35M	-	М	Y	Y	Y	LR	adult		
53	Leo 36M	-	М	Y	Y	Y	LR	adult		
54	Leo 37F	-	F	Y	Y	N	LR	adult		
55	Leo 38F	-	F	Y	Y	Y	LR	adult		
56	Leo 39F	-	F	Y	Y	Y	LR	adult		
57	Leo 40M	-	М	Y	N	N	R	adult		
58	Leo 41F	-	F	Y	Y	Y	LR	adult		
59	Leo 42M	-	м	Y	Y	Y	LR	adult		
60	Leo 43M	-	М	Y	Y	N	LR	adult		
61	Leo 44F	-	F	N	Y	Y	LR	adult		
62	Leo 45F	-	F	Y	Y	Y	LR	adult		
63	Leo 46M	-	м	N	Y	Y	LR	adult		
64	Leo 48F	-	F	N	Y	N	LR	adult		
65	Leo 50F	-	F	Y	Y	Y	LR	adult		
66	Leo 55F	-	F	N	Ŷ	Y	LR	adult		
67	Leo 56F	_	F	Y	Y	Y	LR	adult		
68	Leo 57Un	-	UN	N	Y	N	LR	adult		
69	Leo 58F	-	F	N	Y	Y	LR	adult		
70	Leo 59M	-	M	N	Y	Y	LR	adult		
71	Leo 64M	-	M	N	Y	Y	LR	adult		
72	Leo 65M	-	M	N	Y	Y	LR	adult		
73	Leo 66F	-	F	N	Y	N	LR	adult		
74	Leo 67Un	-	UN	N	Y	N	R	cub	cub in 2014	cub of Leo 66F
75	Leo 68M	-	м	N	Y	N	LR	adult		
76	Leo 69F	-	F	Y	Y	Y	LR	adult		
77	Leo 70F	-	F	Y	N	N	LR	adult		
78	Leo 72Un	-	UN	N	Y	N	LR	adult		
79	Leo 73F	-	F	Y	N	Y	LR	adult		
80	Leo 74F	-	F	Y	Y	Y	LR	adult		
81	Leo 75F	-	F	Y	Y	Y	LR	adult		
82	Leo 76M	-	M	N	Y	Y	LR	adult		

No.	ID-LL*	ID- FRC*	Sex	Captured in 2013	Captured in 2014	Captured in 2015	Side	Adult/ Cub	The year when an individual was captured as a cub	Notes
83	Leo 77M	-	М	N	Y	N	R	adult		
84	Leo 78Un	-	UN	N	Y	N	L	adult		
85	Leo 79F	-	F	N	Y	Y	LR	adult		
86	Leo 80M	-	М	N	Y	Y	LR	adult		
87	Leo 82Un	-	UN	N	Y	Y	LR	adult		
88	Leo 84Un	-	UN	N	Y	N	LR	cub	cub in 2014	cub of Leo 55F
89	Leo 85Un	-	UN	N	Y	N	L	cub	cub in 2014	cub of Leo 55F
90	Leo 87M	-	М	N	Y	N	LR	adult		
91	Leo 88Un	-	UN	N	Y	Y	LR	adult		
92	cub 2 ofLeo 89F	-	UN	Ν	Y	N	R	cub	cub in 2014	cub of Leo 89F
93	Leo 90Un	-	UN	N	N	Y	LR	adult		
94	Leo 92F	-	F	N	N	Y	LR	adult	cub in 2014	cub 1 of Leo 89F
95	Leo 93F	-	F	N	N	Y	R	adult		
96	Leo 94F	-	F	N	N	Y	LR	adult		
97	Leo 96Un	-	UN	N	N	Y	L	adult		
98	Leo 97Un	-	UN	N	N	Y	R	adult		
99	cub 1 ofLeo 39F	-	UN	N	N	Y	LR	cub	cub in 2015	cub of Leo 39F
100	cub 2 ofLeo 39F	-	UN	N	N	Y	LR	cub	cub in 2015	cub of Leo 39F
101	cub 1 ofLeo 16F	-	UN	N	N	Y	L	cub	cub in 2015	cub of Leo 16F
102	cub 2 ofLeo 16F	-	UN	N	N	Y	L	cub	cub in 2015	cub of Leo 16F
103	cub 3 ofLeo 16F	-	UN	Ν	N	Y	L	cub	cub in 2015	cub of Leo 16F
104	cub 1 ofLeo 23F	-	UN	N	N	Y	L	cub	cub in 2015	cub of Leo 23F
105	cub 2 ofLeo 23F	-	UN	N	N	Y	L	cub	cub in 2015	cub of Leo 23F

ID-LL – unique number of leopard individual in the database of Land of the Leopard

ID-FRC - unique number of leopard individual in the database of Feline Research Center

F – Female / M – Male / UN – Unknown / Y – yes / N – no / L – left / R – right

Sample ID	Individual ID (*an individual crossed the border)	Time for collection	Located for collection	Person for collection
2	1	2012.1.14	Madida, Hunchun	Zhu Jiang, Sheng Ledong, Li Min
4	2	2012.4.14	Huangsongdianzi Hunchun	Lang Jianmin, Gao Wenbin, Xue Yangang
8	3	2012.12.18	Sanguan, Hunchun	Lang Jianmin, Gu Jiayin, Xue Yangang
10	4	2013.1	Wulindong, Dongfanghong	Gao Kejiang
11	3	2013.1.22	Naozhigou, Hunchun	
31	5*	2013.5.24	Linghoushan, Songlinmiao	Lang Jianmin, Xue Yangang
51	6	2014.1.8	Hulubie, Hunchun	Xue yangang, Gu jiayin, Li zhilin, Zhang xue
152	7	2015.1.26	Madida, Hunchun	
154	8	2015.1.29	Hunchun	
155	8	2015.1.29	Madida, Hunchun	
159	9	2015.2.8	Huangnihe	
172	10	2013.10.19	Xibeigou	
173	10	2013.12.6	Quliugou, Madida	
183	2	2011.12.18	Northwest of Madida	
204	11	2015.12.24	Heping Muling	Gu Jiayin
205	11	2015.12.18	Huapi Frost farm, Tianqiaoling Administration of Forestry	
209	12	2015.12.22	Dahuanggou	Wu Guoqing
226	13	2016.3.1	Heshangouli	Yang Eryan
228	14	2016.2.26	Heshangouli	Yang Eryan
231	15	2016.3.2	Shichang	
232	15	2015.11.11	Malugou	
233	15	2016.1.17	Yongpinggou	
233	15	2016.1.17	Yongpinggou	

### Annex 3. Amur tigers identified by molecular genetic analysis in China

Sample ID	Individual ID (*an individual crossed the border)	Time for collection	Located for collection	People for collection
30	1	2013.5.8	Lanjia, Wangqing	Jiang Guangshun, Li Zhilin, Zhang Hongjun
36	2	2013.8.21	Lanjia, Wangqing	Li Qi, Cao Zhixin
63	3	2013.9.26	Lanjia, Wangqing	Cao Zhixin
74	3	2013.11.9	Lanjia, Wangqing	Cao Zhixin
88	4	2014.3.30	Lanjia, Wangqing	Cao Zhixin
94	5	2014.3.31	Lanjia, Wangqing	Cao Zhixin
98	3	2014.4.20	Lanjia, Wangqing	Cao Zhixin
99	3	2014.4.18	Lanjia, Wangqing	Cao Zhixin, Li Qi
101	3	2014.4.19	Lanjia, Wangqing	Cao Zhixin, Mu Yanjun
107	3	2012.12.13	Lanjia, Wangqing	
108	6	2012.12.13	Lanjia, Wangqing	
124	7	2014.3.3	Lanjia, Wangqing	Cao Zhixin
214	8*	2015.11.04	Hunchun	Cao Zhixin
215	9	2015.11.04	52 Linban	Cao Zhixin
218	10	2015.11.04	Hunchun	Cao Zhixin

### Annex 4. Amur leopards identified by molecular genetic analysis in China

Tube	Sample ID	Individual ID	Species	DNA concentration	Date of extraction
1	862	1	PPO	17.18	02.07.2015
2	863	2	PPO	22.9	17.06.2015
3	864	3	PPO	20.43	01.07.2015
4	865		PPO	23.08	15.06.2015
5	866	2	PPO	25.8	25.06.2015
6	867	2	PPO	21.21	25.06.2015
7	873	4	PPO	22.78	25.06.2015
8	880	5	PPO	37.02	30.06.2015
9	881		PPO	46.92	30.06.2015
10	884	6	PPO	136.68	22.06.2015
11	885		PTA	29.8	22.06.2015
12	887	7	PPO	25.27	11.06.2015
13	893	6	PPO	75.81	15.06.2015
14	894		PPO	34.55	26.06.2015
15	896	8	PPO	52.44	30.06.2015
16	907		PPO	15.57	02.07.2015
17	908	5	PPO	28.49	09.06.2015
18	909		PPO	41.4	30.06.2015
19	911	9	PPO	22.57	30.06.2015
20	914		PPO	23.56	19.06.2015
21	917		PPO	103.04	15.06.2015
22	919	6	PPO	37.43	25.06.2015
23	921		PPO	21.82	18.06.2015
24	922		PPO	42.85	16.06.2015
25	923	10	PPO	39.85	01.07.2015
26	928	9	PPO	28.98	17.06.2015
27	930		PPO	41.23	01.07.2015
28	940		PPO	15.81	18.06.2015
29	942		NEITHER	61.1	02.07.2015
30	964		NEITHER	138.33	22.06.2015
31	983	11	PPO	45.02	11.06.2015
32	986		PPO	22.97	24.06.2015
33	987		NEITHER	93.13	17.06.2015
34	989	12	PPO	40.48	11.06.2015
35	990	13	PPO	69.42	02.07.2015
36	995		PPO	45.1	15.06.2015
37	996		PPO	20.72	30.06.2015
38	997		PPO	23.07	16.06.2015
39	998		PPO	36.65	26.06.2015
40	999		NEITHER	53.48	02.07.2015
41	1002		NEITHER	159.05	16.06.2015
42	1004		PPO	25.55	01.07.2015
43	1005		PPO	87.08	08.06.2015
44	1009		PPO	35.89	15.06.2015
45	1010	14	PPO	59.05	25.06.2015
46	1012		PPO	25.49	25.06.2015

### Annex 5. Amur tigers and Amur leopards identified by molecular genetic analysis in Russia

Tube	Sample ID	Individual ID	Species	DNA concentration	Date of extraction
47	1013		NEITHER	67.37	26.06.2015
48	1014		NEITHER	69.45	23.06.2015
49	1015		PPO	16.78	02.07.2015
50	1016		PPO	58.37	09.06.2015
51	1022		PPO	51.5	09.06.2015
52	1026		NEITHER	169.29	30.06.2015
53	1027	15	PPO	67.33	19.06.2015
54	1028	16*	PPO	43.31	01.07.2015
55	1032		PPO	60.17	15.06.2015
56	1040		PPO	30.25	02.07.2015
57	1050		PPO	54.02	02.07.2015
58	861		NEITHER	159.11	15.06.2015
59	870		PTA	49.12	02.07.2015
60	871		PTA	54.25	30.06.2015
61	875		PTA	64.73	30.06.2015
62	876		PTA	20.73	17.06.2015
63	877		PTA	17.39	30.06.2015
64	888		PTA	31.41	18.06.2015
65	890		NEITHER	133.21	30.06.2015
66	892		PTA	23.69	24.06.2015
67	900		PTA	224.2	15.06.2015
68	903		PTA	23.22	26.06.2015
69	904	1	PTA	38.14	01.07.2015
70	918	2	PTA	47.5	25.06.2015
71	920		PTA	123.62	30.06.2015
72	924		PTA	36.8	15.06.2015
73	931		PTA	38.06	26.06.2015
74	933		PTA	356.24	15.06.2015
75	936	3*	PTA	23.54	22.06.2015
76	945		NEITHER	127.65	25.06.2015
77	952		PTA	29.19	23.06.2015
78	953		PTA	15.55	15.06.2015
79	954		PPO	139.95	24.06.2015
80	955	1	PTA	313.63	15.06.2015
81	956	6	PTA	80.65	09.06.2015
82	959		PTA	20.96	02.07.2015
83	961		PTA	38.17	22.06.2015
84	962		PTA	58.94	23.06.2015
85	966		PTA	17.93	22.06.2015
86	969	7	PTA	34.13	25.06.2015
87	972	8	PTA	25.38	24.06.2015
88	973		PTA	15.58	30.06.2015
89	974	9	PTA	45.85	24.06.2015
90	975	-	PTA	18.88	25.06.2015
91	976		PTA	104.68	15.06.2015
92	980		PTA	15.43	19.06.2015
93	982		PTA	19.48	24.06.2015
94	982		PTA	32.81	01.07.2015

Tube	Sample ID	Individual ID	Species	DNA concentration	Date of extraction
95	985		NEITHER	222.45	26.06.2015
96	993		NEITHER	190.38	24.06.2015
97	1000		ΡΤΑ	26.99	18.06.2015
98	1019	2	PTA	35.82	22.06.2015
99	1021		PTA	21.1	09.06.2015
100	1023		PTA	32.36	25.06.2015
101	1025	2	PTA	15.51	15.06.2015
102	1029		PTA	32.96	25.06.2015
103	1034		PTA	62.04	17.06.2015
104	1037		ΡΤΑ	36.96	01.07.2015
105	1048		ΡΤΑ	16.78	22.06.2015
106	1051		NEITHER	72.6	30.06.2015
107	899		PPO	38.85	30.06.2015
108	901		PTA	18.93	19.06.2015
109	906		PTA	24.43	09.06.2015
110	910		PTA	32.39	24.06.2015
111	912		PPO	40.81	24.06.2015
112	913		PPO	202.91	09.06.2015
113	932	1	PTA	27.4	19.06.2015
114	934		PTA	59.58	26.06.2015
115	935	3*	PTA	19.85	17.06.2015
116	937		PTA	75.47	18.06.2015
117	938		PTA	26.04	30.06.2015
118	939		PTA	24.7	02.07.2015
119	943		PPO	21.81	19.06.2015
120	944	4	PTA	38.84	18.06.2015
121	946		PTA	26.72	02.07.2015
122	947	3*	PTA	22.85	25.06.2015
123	948	5	PTA	15.26	16.06.2015
124	957		PTA	29.58	02.07.2015
125	958		PTA	89.11	22.06.2015
126	967	3*	PTA	28.01	23.06.2015
127	1001	5	PTA	48.89	15.06.2015
128	1008	4	PTA	16.75	11.06.2015
129	1024		PTA	50.7	26.06.2015
130	1043		PPO	37.86	18.06.2015
131	1045		PTA	15.23	02.07.2015
132	1046		PPO	50.25	02.07.2015
133	1047		РРО	28.3	17.06.2015
134	1052		PTA	23.17	16.06.2015
135	1053	17	PPO	27.66	22.06.2015

1. \* represented for common samples between China and the Russian Federation.

2. PTA represents the Amur tiger, and PTO represents the Amur leopard.







## North-East Asian Subregional Programme for Environmental Cooperation (NEASPEC)

NEASPEC is a comprehensive intergovernmental cooperation framework, established in 1993 by six member states, namely, China, Democratic People's Republic of Korea, Japan, Mongolia, Republic of Korea and the Russian Federation. Senior Officials Meeting (SOM) is held annually as the governing body and principal vehicle for the evolution of NEASPEC. UNESCAP-ENEA Office functions as the NEASPEC Secretariat.

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