

How healthy the Indian leopards are?

Carnivores face a continuous threat for their survival from pathogens, indirectly resulting from anthropogenic factors such as human population expansion, habitat loss or its alteration and climate change leading to their endangerment in the wild. These effects are also manifested as a distortion of historically stable host-pathogen interaction and introduction of more virulent pathogens responsible for epizootic events. Many felid species like Tanzania lion, Florida panther and African cheetah are facing a threat from reduced genetic diversity which is reflected by decreased survival and fitness-related features such as inferior reproductions, higher mortality rates and increased vulnerability to opportunistic infectious diseases, ultimately bringing the population towards the brink of extinction.

Major Histocompatibility Complex (MHC) is a multi-genic protein complex in vertebrates which interacts with antigens (e.g. viruses, cancer infected cells, bacteria, protozoa, arthropods etc.) and trigger adaptive immune response during any disease outbreak. MHC polymorphism is important for the host to possess genetic variations for handling a huge range of emerging pathogens and for the health and viability of the population to overcome a disease outbreak. MHC is therefore one of the desirable markers to study the patterns of adaptive genetic variations related to disease resistance in host natural populations.

Leopard, a member of the cat family (genus: *Panthera*), are found highly adaptable to habitats ranging from desert to rainforest and hence, have the largest geographic distribution among the wild cats. Anthropogenic pressures caused heavily fragmented and isolated leopard populations, which ultimately placed most leopard subspecies as 'Near Threatened' and few others, as 'Critically Endangered' by The International Union for the Conservation of Nature (IUCN). The current estimated population size of Indian leopards (*Panthera pardus fusca*) is in the range of 12,000–14,000 individuals in wild, and 502 individuals in captivity which are continuously exposed to a wide range of pathogens as being highly adaptable to different habitats.

A study on captive Asiatic lions earlier, revealed a high prevalence of feline parvovirus (FPV) and canine distemper virus (CDV), a major cause of mortality in captive felid populations indicated the importance of proper care management practices and suggested conservation measures to be taken in wild and in captive bred animals. The dearth of information on the genetic structure of MHC regions in the Indian leopard was therefore, necessary to understand the evolutionary predisposition of these elusive, near threatened and widely distributed species in India to cope with the pathogenic exposure in their vicinity. Further, growing numbers of incidents of human-leopard conflicts in India lead to repeated exposures to novel pathogens specific to human populated areas hitherto unexposed to leopards and is suggestive of the pertinence of studying their immuno-genetic structure. Estimating the level of MHC polymorphism has direct implications for the conservation of species.

Our study imparted an understanding of molecular characterization of MHC-I and MHC-II DRB loci

to estimate nucleotide and allelic differences in MHC region among Indian leopards. The better understanding of the MHC-I and MHC-II loci should help researchers to comprehend immune gene diversity in Indian leopards and other related species. We also aimed to examine the genetic variation at MHC loci in wild as well as captive individuals, which will help to find out if habitat fragmentation has resulted in significant changes at these two genes.

The wild group of individuals showed a significantly higher diversity/polymorphism in their DNA sequences (both, nucleotide and amino acids) when compared to the captive group of individuals. This indicates better health and survivability against pathogenic attacks among wild individuals. Besides, the reason behind reduced polymorphism in captive animals could be inbreeding that comes with isolation in captivity, or growth in a restricted environment, both of which result in a loss of genetic variation. Also, the variations found among wild and captive animals might be the micro-evolutionary forces such as pathogen mediated selection that might be acting on two different groups of animals as they have been bred in two different climatic/environmental conditions for a substantial period of time. The peptide binding sites (PBS) in MHC regions which are responsible for recognizing a diverse array of antigens, showed a higher degree of polymorphism in comparison to non-PBS regions in both captive and wild group of animals. Signs of positive selection were found acting on PBS sites of MHC-II DRB region responsible for the maintenance of variations. Since, MHC diversity is often linked with the quality of immunological health; the results from the current study fill the gap of knowledge on disease predisposition among wild and captive Indian leopards.

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