

spect to germinability and seedling growth<sup>6</sup>. In mango large seeds showed better performance than medium and small-sized seed nuts. Seed germination was higher as the seed weight increased in oilpalm<sup>7</sup>. The seed embryo possesses trichomes and during TZ staining only the plumle stained with unstained chlorophyllous cotyledon. Contribution of embryo to seed weight is negligible. This may be due to the barrier-like surface cells that could prevent the infusion of TZ solution. Further studies are needed for fruit weight ranging more than 1 g. Correlation studies revealed the positive nature of fruit size on seed size. Double-seeded fruits were superior in providing seeds of higher weight with no embryos.

1. Sivasubramaniam, K., Raja, K. and Geetha, R., In *Recalcitrant Seeds: Causes and Effects*, Satish Serial Publishing House, New Delhi, 2012, p. 7.
2. Hosomi, S. T., Santos, R. B., Custodio, C. C., Seato, P. T., Marks, T. R. and Machodoneto, N. B., *Seed Sci. Technol.*, 2011, **39**, 178–189.
3. Deswal, D. P. and Chand, U., *Seed Sci. Technol.*, 1997, **25**, 409–417.
4. Pina-Rodrigues, F. C., Figliolia, M. B. and Peixoto, M. C., In *Germinacao – do basico ao aplicado* [Germination – From the Basic to the Applied] (eds Ferreira, A. G. and Borghetti, F.), Artmed, Porto Alegre, 2004, pp. 283–297.
5. Enescu, V., In *Tree and Shrub Seed Handbook*, International Seed Testing Association, Zurich, Switzerland, 1991.

6. Berger, C. A. M., Cordoso, A. A., Dias, D. C. F. and Andsiqueira, Dc-de, *Rev. Bras. Fruticultura*, 1998, **20**, 272–282.
7. Myint, T., Chanprasert, W. and Srikul, S., *Seed Sci. Technol.*, 2010, **38**, 125–135.

Received 29 March 2012; revised accepted 23 August 2012

K. SIVASUBRAMANIAM\*  
K. SELVARANI

Department of Seed Science and  
Technology,  
Agricultural College and Research  
Institute,  
Madurai 625 104, India  
\*For correspondence.  
e-mail: seedmani@yahoo.com

## Wolves in Trans-Himalayas: 165 years of taxonomic confusion

Species recognition and systematics of canids has always been a subject of contention among the experts. Canid Action Plan, published by IUCN/SSC Canid Specialist Group in 2004, mentions that there is an existing argument among scientists regarding species number, which ranges from 34 to 38 species<sup>1</sup>. Many species and subspecies of wolves have been identified and reported from different parts of the world. Approximately 13 subspecies of Grey wolf (*Canis lupus*) are recognized, which may not be related so closely<sup>2</sup>. Presence of ecological, morphological and molecular continuum among the species and subspecies of canids makes it difficult to demarkate them. In recent years, discovery of a new wolf species from India has been reported, but the recognition and proper identification of this population as a different species is still ambiguous. This issue came into light with an article by Aggarwal *et al.*<sup>3</sup> showing that the wolf population found in the Indian region of Trans-Himalaya, earlier generally considered to be another population of Tibetan wolf (*Canis lupus chanco*) – a subspecies of the Grey wolf, could be an entirely different species or subspecies. Their results further showed that this population is the oldest lineage of the wolf-dog clan worldwide. Similar results were found in the case of the population from peninsular India, which was generally considered to be a population of

another subspecies of the Grey wolf, viz. *Canis lupus pallipes*. This population was also found suitable for upgradation as a separate species or subspecies of wolves. These results were based upon the analysis of mitochondrial DNA and rRNA samples from different populations of canids from all over the world.

Major results of this study, however, were corroborated by similar studies almost at the same time<sup>2,4</sup>. The samples by Sharma *et al.*<sup>4</sup> were collected from much diverse sources and most of the canid populations of the world were represented. This study focused on marking the time of origin of the wolf lineages in India. It also applied the same genetic techniques using mitochondrial DNA and rRNA, and the species divergence was calculated based on fossil record estimates of the divergence time of the coyote and wolf lineages to calibrate sequence divergence rates for each gene. Results of this study show that samples of *C. lupus pallipes* (wolf from peninsular India, Iran, Iraq and parts of Arabia) and *C. lupus chanco* (wolf from Indian Trans-Himalaya, Tibet and Nepal) fall in three separate clades, viz. Indian *C. lupus pallipes* (eastern Pakistan and peninsular India), Himalayan clade of *C. lupus chanco* (Ladakh, Spiti, Tibet and Nepal) and wolf-dog clade (including *C. lupus chanco* from northwest Jammu and Kashmir, i.e. Gilgit and Baltistan). They, further, argue that Himalayan *C. lupus*

*chanco* is the most ancestral and diverged at 800,000 years ago, when the Himalayan region was going through a major geologic and climatic upheaval. Indian *Canis lupus pallipes* is altogether diverged from wolf-dog clade 400,000 years ago. These lineages are the oldest of all wolf lineages in the world, hence it is postulated that India could have been the centre of origin of wolf-dog clan. In this study, dogs were reported to be in close relation with the wolves from Europe and America, therefore, wolves of India might have not been used for domestication. Dogs have originated from multiple wolf ancestors and they started to diverge about 150,000 years ago<sup>5</sup>.

Although revising the status of wolves of the Himalayas as species or subspecies, distinct from the other populations, remains disputed, the Himalayan wolf (*C. lupus laniger*) was included in the agenda of Wolf Specialist Group of IUCN in 2005 (accessed from <http://wolf-specialistgroup.org/resolutions/>). Aggarwal *et al.*<sup>6</sup> published another paper improving on their previous paper. Moreover, they raised a few doubts on the results and methodology of Sharma *et al.*<sup>4</sup>. The new results again confirmed the distinctness of Himalayan and Indian wolves from the Trans-Himalaya and peninsular India respectively, as different species; and the lineage of the Himalayan wolf was confirmed to be the oldest. Following these

studies, CITES also included the Himalayan wolf and Indian wolf with the scientific names *Canis himalayensis* and *Canis indica* respectively as split from the Grey wolf (*Canis lupus*) in its Nomenclature Matters<sup>7</sup>.

Taxonomic confusion regarding the identification and recognition of wolves from the Trans-Himalayan region of India and parts of Tibet has persisted for the last 165 years. Hodgson<sup>8</sup> was the first to describe the Himalayan wolf as a distinct species, *Canis laniger*, noting its well-developed frontal sinuses, unusually elongated muzzle, distinct coloration and the woolliness of its under fur (cited in Sharma *et al.*<sup>4</sup>). Blanford<sup>9</sup> later combined *C. laniger* with *C. lupus* and elevated the Indian wolf to *C. pallipes* (Box 1). His views about the wolves of Baluchistan and Gilgit are consistent with the findings of Sharma *et al.*<sup>4</sup>. Much later, Pocock<sup>10</sup> described both taxa as subspecies of *C. lupus*, making *C. laniger* and *C. pallipes* parts of the more widely distributed *C. lupus chanco* and *C. lupus pallipes* respectively. These views were widely accepted until genetic analysis revealed otherwise and revived the discussion.

**Box 1.** Excerpt from Blanford's account of wolves from Trans-Himalaya<sup>9</sup>.

'The Sind, Baluchistan, and Gilgit animals appear undistinguishable from European wolves. The variety found in Tibet and Ladakh is, however, very pale-coloured, with woolly fur, and has generally been distinguished as *C. laniger*. I thought at one time that the dentition was different, the upper sectorial in *C. laniger* being generally shorter than the two upper true molars taken together, whilst the reverse was believed to be the case in *C. lupus* (P. A. S. B. 1877, p. 116); but Huxley in his paper already quoted (p. 279) has shown that the teeth of both European and Tibetan wolves vary in this respect, and the difference in the fur appears due to climate. The cranial distinctions mentioned by Blyth (J. A. S. B. xxiii, p. 733) are probably caused by age. The black Tibetan wolf, classed apart by some, is evidently a variety similar to the black European wolf that was called *Canis lycaon* by Schreber.'

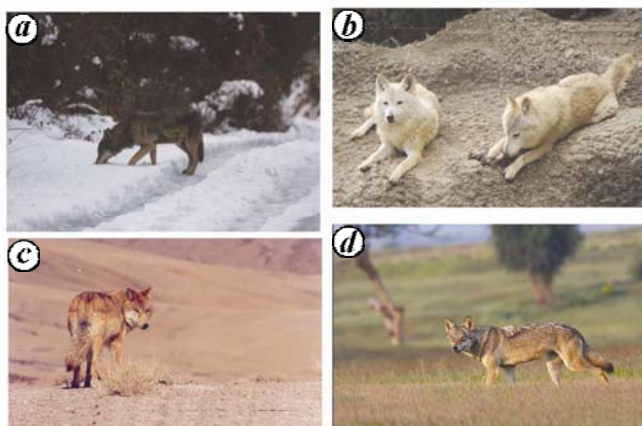
However, so far none of the studies has been able to sort out the problems related to the taxonomic identification of wolves of the Trans-Himalayan landscape of India. O'Brien<sup>11</sup> states that scientists, particularly biologists and geneticists, love to challenge established thinking and establish new paradigms. But in taxonomy, this dynamics is largely rhetorical and almost philosophical. Pocock<sup>10</sup> was probably influenced by intermediate pelaged individuals at areas of contact in northern Punjab and Kashmir, which caused the problem in recognizing the populations of wolves from India as distinct. With the advent of new molecular technology, it became easier to confirm the distinctness of a species at the molecular level. Though the new techniques of genetic identification of a species seem to be promising and helpful in many cases, these are still not error-proof to come up with clear results and, sometimes, add to the confusion. The same is the case with wolves in India.

The recent genetic studies have lacked in one or another aspect to provide a complete picture. Aggarwal *et al.*<sup>3,6</sup> have argued that the Himalayan wolf is present strictly in the Indian region of Ladakh and Spiti and differs from the Tibetan wolf. This raises another question: as these areas are part of the same landscape, what ecological or behavioural barriers could be facilitating such strict divergence, particularly when no striking morphological differences occur between the wolves from Tibet and Indian Trans-Himalaya? Sharma *et al.*<sup>4</sup> have shown the extent of the Himalayan wolf throughout the Himalayas and their findings suggest that the wolves from northwest Jammu and Kashmir, i.e. Gilgit and Baltistan, are part of another lineage. These have fallen in the clade of wolf-dog, suggesting that they could be part of the Grey wolf and diverged in recent times only. However, their failure to collect samples from the living population of this region makes it difficult to come to any conclusion. Our initial survey in Jammu and Kashmir revealed the presence of a population of wolf in Kashmir valley, which arguably had appeared in the valley quite recently, and their presence has become common. Sharma *et al.*<sup>4</sup> predict Kashmir as the area of potential contact of all three clads. However, none of the studies has collected samples from this population. Both the studies of Aggarwal *et al.*<sup>3,6</sup> and Sharma

*et al.*<sup>4</sup> have collected samples from the Indian zoo or museum specimen. Srivastav and Nigam<sup>12</sup> have recorded the family history of all the wolves of the Himalayas in Indian zoos. The records mention that only two female wolves were captured from the wild and rest were captive-bred. This suggests the lack of variety in the mitochondrial DNA samples of the zoo animals due to maternal heredity. In such a condition, any claim of collecting enough representative samples<sup>6</sup> cannot withstand in the absence of samples from the field. Zhang and Chen<sup>13</sup> state that the Tibetan wolf might be an archaic pedigree within the wolf subspecies. However the study defines *C. lupus laniger* as the Tibetan wolf distinct from *C. lupus chanco*, the Mongolian wolf.

Morphological appearance of the wolves from different parts of India (Figure 1) shows certain dissimilarities. Skulls of the two males from Chumar, Ladakh were measured by Allen<sup>14</sup> (234 and 236 mm), which are the largest for wolves in India, but smaller compared to North American wolves, which can measure up to 290 mm (refs 14 and 15). *C. lupus pallipes* has the smallest skull length, measuring maximum up to 220 mm. Zygomatic widths of the skull of wolves from Ladakh (129 and 128 mm) were also comparatively larger than those of peninsular wolves from India (90.2–109 mm). Upper cheek teeth, i.e. canine to last molar of two wolves from Ladakh measured 105 and 98.4 mm, which is larger compared to those of peninsular wolves and Arabian wolves (93.6–97 mm and 81.3–93 mm respectively)<sup>14–16</sup>. The wolf from peninsular India appears smaller in size and more brownish in colour, whereas wolves from the Himalayan regions are large and whitish. Peninsular wolf weighs 25 kg on an average, which may be the lowest among all wolves, whereas wolves from the Himalayan region weigh about 35 kg, similar to Tibetan wolves<sup>17,18</sup>. Wolves from Ladakh, Spiti, Sikkim and the zoo wolves (all the wild caught animals were captured from Spiti valley) appear to have similar morphological features, falling in the same continuous landscape. The genetic relatedness of wolves in Kashmir valley, which appears much greyish in colour, is yet to be determined.

Wolves in the Himalayas, despite their abstruse status as distinct species or subspecies, serve an important role in the ecology of Trans-Himalaya, holding



**Figure 1.** Wolves from **a**, Kashmir valley, North-west Himalayan region of India (courtesy: Mir M. Mansoor); **b**, Sikkim Zoo – Captive-bred individuals, wild individuals were captured from Spiti, Himachal Pradesh, Trans-Himalayan Landscape (courtesy: Pankaj Kumar); **c**, Leh–Ladakh, Trans-Himalayan Landscape (courtesy: Y. V. Bhatnagar); **d**, Peninsular India, central Indian Landscape (courtesy: A. Patil).

the status of the top predator along with the snow leopard. Wolves in India are protected by law under Schedule I of the Wildlife Protection Act 1972. However, the wolves from the Himalayas are one of the least protected large mammals and also the least studied mammals of the country. The only study on their abundance, so far, concludes with the presence of only 350 animals in the wild<sup>19</sup>. The fact revealed through the genetic studies, that they are the oldest lineage of the wolves in the world, adds to their importance with respect to conservation. The Wildlife Institute of India, Dehradun started a project on ecology and conservation of wolves in the Himalayas in 2010 to fill this information gap and formulate conservation measures for these mammals of high altitude. The initial survey was conducted to study the level and pattern of human–wolf conflict in their distribution area and mark their distribution range in the Himalayas, with identification of conservation priority areas for them. Studies show that these wolves are the top predators of livestock accounting for 60% of the total livestock loss because of predation, followed by the snow leopard (38%)<sup>20</sup>. Agriculture is limited in arduous living conditions in the Trans-Himalayan region and livelihood of local people is mainly dependent on small livestock. This landscape serves as a grazing ground for nomadic and local herders and is economically important to these groups. Moreover, low productivity in these areas constricts the population of wild prey population and brings the wolves into conflict with humans. This results in retaliatory killing of

the wolves, which is one of the biggest threats to them.

The confusion regarding identification of wolves in the Himalayas and recognition of their taxonomic status needs to be resolved to provide a stronger conservation impetus. Thus, genetic samples from all the areas in the Himalayas where the wolves are present are needed to be collected to get a good representation for reliable results of genetic relatedness among the different wolves. Ecological and behavioural studies of their populations in the Himalayas are equally important and will be vital to its conservation. It has been 165 years since the wolf of the Himalayas was first described and it still has remained a taxonomic confusion. Thus, recognizing a taxa properly is essential in order to prevent the extinction of any species without recognition.

1. Sillero-Zubiri, C., Hoffman, M. and Macdonald, D. W., Report, IUCN – The World Conservation Union, 2004.
2. Jhala, Y. V. and Sharma, D. K., *Int. Wolf*, 2004, **14**, 15–16.
3. Aggarwal, R. K., Ramadevi, J. and Singh, L., *Genome Biol.*, 2003, **4**, P6.
4. Sharma, D. K., Maldonado, R. E., Jhala, Y. V. and Fleischer, R. C., *Proc. R. Soc. London: Ser. B (Suppl. 3)*, 2004, **271**, S1–S4.
5. Vila, C. *et al.*, *Science*, 1997, **276**, 1687–1689.
6. Aggarwal, R. K., Kivisild, T., Ramadevi, J. and Singh, L., *J. Zool. Syst. Evol. Res.*, 2007, **45**, 163–172.
7. CITES Nomenclatural Matters, Twenty-fourth Meeting of the Animals Committee Geneva, Switzerland, 20–24 April 2009.
8. Hodgson, B. H., *Calcutta J. Nat. Hist.*, 1847, **7**, 469–477.

9. Blanford, W. T., *The Fauna of British India, Including Ceylon and Burma, Mammalia*, Taylor and Francis, 1888, pp. 135–140.
10. Pocock, R. I., *The Fauna of British India, Including Ceylon and Burma, Mammalia*, Taylor and Francis, 1941, vol. II.
11. O'Brien, S. J., In *Tears of the Cheetah: And Other Tales from the Genetics Frontier* (eds O'Brien, S. J. and Mayr, E.), Thomas Dunne Books, 2003.
12. Srivastav, A. and Nigam, P., *National Pedigree Book of Tibetan Wolf (Canis lupus chanco)*, Wildlife Institute of India, Dehradun, 2009.
13. Zhang, H. and Chen, L., *Mol. Biol. Rep.*, 2009, **38**, 1651–1660.
14. Allen, G. M., *Proc. Acad. Natl. Sci. Philadelphia*, 1938, **90**, 261–294.
15. Elbroch, M., *Animal Skulls: A Guide to North American Species*, Stackpole Books, 2006, pp. 375–377.
16. Harrison, D. L., Some comparative features of skulls of Wolves (*Canis lupus* Linn.) and Pariah Dogs (*Canis familiaris* Linn.) from the Arabian Peninsula and neighboring lands. *Bonner Zoologische, Beirtrage*, 1973, pp. 185–191.
17. Kaczensky, P., Enkhsaikhan, N., Ganbaatar, O., Walzer, C. and The, C., *Wildl. Biol.*, 2008, **14**, 444–456.
18. Goldman, A. E. A., *J. Mammal.*, 1937, **18**, 37–45.
19. Fox, J. L. and Chundawat, R. S., In *Ecology and Conservation of Wolves in a Changing World*, Proceedings of the Second North American Symposium on Wolves, Alberta, Canada, 1995, pp. 95–104.
20. Namgail, T., Fox, J. L. and Bhatnagar, Y. V., *Environ. Manage.*, 2007, **39**, 490–496.

**ACKNOWLEDGEMENTS.** We thank the Director and Dean, Wildlife Institute of India, Dehradun for permission to undertake this project and for their constant guidance. We also thank the MBZ Species Conservation Fund for funding the initial part of the study; all those who shared their photographs of wolves from Himalayan and Trans-Himalayan landscape and Dr Jhala for his comments on an earlier draft.

Received 27 July 2012; accepted 3 September 2012

SHIVAM SHROTRIYA  
SALVADOR LYNGDOH  
BILAL HABIB\*

Department of Animal Ecology and  
Conservation Biology,  
Wildlife Institute of India,  
Dehradun 248 001, India  
\*For correspondence.  
e-mail: bh@wii.gov.in