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RESEARCH ARTICLE

OPPORTUNISTIC OCCURRENCE, SIGNIFICANCE AND CONSERVATION IMPLICATIONS OF TRICHO-MORPHOMETRICS: LARGE WILD HERBIVORES OF CHANG CHENMO VALLEY, LADAKH, INDIA.

*Vinita Sharma^{1,3}, Mohd. Raza², Khursheed Ahmed², Parag Nigam³, Anjara Anjum Khan², Chandra Prakash Sharma³, Vipin Sharma³ and Surendra Prakash Goyal³.

1. Central University of Jammu, Department of Animal Sciences and Wildlife, Central University of Jammu, Rahya-Suchani (Bagla), District – Samba, Jammu – 181143, Jammu and Kashmir, India.
2. Centre for Mountain Wildlife Sciences, Faculty of Veterinary Sciences and Animal Husbandry, Sher-e-Kashmir University of Agricultural Sciences and Technology, Srinagar, Jammu and Kashmir, India-190006.
3. Wildlife Institute of India, Post Box # 18, Chandrabani, Dehradun, Uttarakhand, India-248001.

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Abstract

Tricho-morphometric studies plays significant role in understanding various aspects of mammalian biology and ecology that have direct conservation implications for species and associated habitats. In the present study, the Chang Chenmo Valley (CCV) region of Changthang Wildlife Sanctuary (CWLS) in the Western Himalayas, India was extensively surveyed during August–October, 2013 to document the occurrence of large wild herbivores. Both direct (sightings) and indirect (calls, faecal pellets, chew and hoof marks, hair, carcasses) evidences were recorded and geo-referenced accordingly. Resting sites, tracks and areas frequently visited by animals were intensively searched for the presence of opportunistically found naturally shed hair which were collected (n=43) for identification of species using tricho-morphometric analysis following established protocols. Five large wild herbivore species, namely the Tibetan Antelope (*Pantholops hodgsonii* Abel, 1826), the Blue Sheep (*Pseudois nayaur* Hodgson, 1833), the Tibetan Argali (*Ovis ammon hodgsonii* Blyth, 1841), the Wild Yak (*Bos mutus* Przewalski, 1883) and the Tibetan Wild Ass (*Equus kiang* Moorcroft, 1841) were directly sighted in the CCV region. Simultaneously multi-species assemblages in inhospitable terrain were also confirmed by tricho-morphometric analysis. Habitat conservation is the vital requirement for protecting the species and ecological processes of the entire ecosystem. Therefore, we suggest that hair be collected along with GPS locations when surveys are undertaken for direct evidence. Tricho-morphometric studies from opportunistically found hair provides additional help to understand species ecology, estimating relative abundance, occupancy and in establishing wildlife–habitat relationships which may help in the development of effective conservation policies for restoring unique habitats.

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Corresponding Author:- Vinita Sharma.

Address:- Central University of Jammu, Department of Animal Sciences and Wildlife, Central University of Jammu, Rahya-Suchani (Bagla), District – Samba, Jammu – 181143, Jammu and Kashmir, India.

Introduction:-

Large wild mammalian herbivores occupy half of the earth's land surface. They are considered as ecological engineers (Owen-Smith, 1988; Dinerstein, 2003; Chane and Yirga, 2014) and play an important role in shaping the structure, diversity and functioning of most of terrestrial ecosystems (Owen-Smith, 1988; Gordon *et al.*, 2004). Although having a major impact on animals land use and habitats of conservation importance significantly, their very survival is threatened by natural and anthropogenic activities worldwide (Ripple *et al.*, 2015). Information on the distribution of species, diversity patterns and wildlife-habitat relationships is crucial for understanding the ecological and evolutionary determinant of spatial heterogeneity in biodiversity (Ricklefs and Schluter, 1993). The distribution of large herbivores is resource dependent (Olf *et al.*, 2002). Rangelands across the world are important for wildlife conservation (Bhatnagar *et al.*, 2007). The Tibetan Plateau (TP) is the highest and largest alpine grassland region in the world. It provides a unique habitat for many globally threatened wildlife species (Foggin, 2000). The TP is an important highland grazing ecosystem (Goldstein and Beall, 1990) supporting the intact assemblage of Pleistocene large wild herbivores (Schaller, 1998; Bhatnagar *et al.*, 2007). It is also one of the most fascinating regions for scientific research (Zeng *et al.*, 2015). Ladakh is the westernmost range of the TP in India. This area supports eight large wild herbivore species (Mallon, 1998; Bhatnagar *et al.* 2007; Namgail, 2009; Shawl and Takpa, 2009; Namgail *et al.*, 2010). A study of the literature clearly reveals that there is intense competition for resources between wild and domestic herbivores (Prins, 2000; Madhusudan, 2004). Over the last few decades, the survival of several Trans-Himalayan wild herbivores has been severely threatened. These species have undergone local extinctions and range reductions due to various natural and anthropogenic activities (Fox *et al.* 1991; Mishra *et al.* 2002; Bhatnagar *et al.* 2007). Habitat management practices provide aid to conserve, protect and restore the habitat of wild plants and animal species with especial reference to conservation reliant species worldwide. Determining the extent of occurrence (EOO) of conservation-reliant species is the foremost requirement in developing strategies for habitat management on a large scale. Various methods used for determining the EOO, including direct counts and indirect methods such as those involving faecal matter/pellets, have limitations in this inhospitable habitat in discriminating wild species from domestic counterparts. Hair collected using snares/traps has been widely used in EOO determination as well as in occupancy surveys of carnivore species (Long *et al.*, 2011). Naturally shed hair found near resting places of animals can also help to identify and discriminate species using laboratory-based methods. The microstructure of hair is a useful tool in identifying mammals and is increasingly gaining importance in many areas including forensic science, ecology, environmental toxicology, wildlife management, epidemiology, assessment of geographic mobility, archaeology and palaeontology (White 1993; Ashraf *et al.*, 1994; O'Connell and Hedges, 1999; Quadros and Monteiro, 2006).

The Chang Chenmo Valley (CCV) region, in the northern part of Changthang Wildlife Sanctuary (CWLS), along with the Daulet Beg Oldi (DBO) area of Karakoram (Nubra-Shyok) Wildlife Sanctuary (KNSWLS), in Ladakh, Jammu and Kashmir, forms the western-most range of the TP within the administrative boundaries of India. The CCV region was extensively surveyed for the large wild herbivore species of the TP ecosystem. The survey was based on direct and indirect evidence such as faecal matter, pellets and hair in the present study. Trichomorphometric characteristics (qualitative and quantitative) of five large wild herbivores encountered in the study area were used to identify species and determine the EOO and other conservation implications in CCV. Present and previous studies (Shawl *et al.*, 2011) justify the immediate development of a trans-boundary habitat management action plan that helps conserve the habitat of large wild herbivore species across the TP.

Materials and methods:-**Study area**

The study was carried out during August–October, 2013 in CCV (Fig. 1). The study area is a south-western extension of the TP (Alfred *et al.*, 2006; Shawl and Takpa, 2009), located on the border between India and China (Shawl *et al.*, 2011). Geographically, it is situated between 78°07'00" E to 79°00'00" E longitudes and 33°55'00" N to 34°39'00" N latitudes. The altitude ranges between 3700 and 5500 metres above the sea level (Sarkar *et al.*, 2008). The area is drained by the Chang Chenmo and Kurgrung rivers (Meinertzhagen, 1927; Shawl and Takpa, 2009; Shawl *et al.*, 2011). It bounded by the Silung Burma on the east, the Silung Yogma on the west, low hills on the north and hill ranges on the south (Shawl and Tapka, 2009; Shawl *et al.*, 2011) (Fig. 1). The area is characterized by steep, rugged mountains, narrow gorges and wide open valleys surrounded by open, rolling sandy mountains in the east and undulating rugged terrain in the north and south. The temperature ranges from 0° to 30° C in summer and from -10°C to -40°C in winter (Mishra and Humbert-Droz 1998). The vegetation of the region is characterized

as dry alpine steppe (Champion and Seth, 1968). The area falls within Durbuk Block, which was earlier used by the nomadic pastoralists of Phobrang and adjacent areas. The area has representative components of the Ladakh Trans-Himalayan ecosystem, which is the most fragile and hardy ecosystem, having a unique, diverse and rare assemblage of high-altitude wildlife species (Fox *et al.*, 1991; Vinod and Sathyakumar, 1999; Rodgers *et al.*, 2000; Namgail, 2009; Shawl and Takpa, 2009). It is an important habitat for the endangered Tibetan antelope outside China (Shawl and Takpa, 2009; Shawl *et al.*, 2011). It also provides a habitat for the Tibetan argali, wild yak, kiang, blue sheep, Himalayan marmot, Tibetan woolly hare, red fox, Tibetan wolf and snow leopard (Meinertzhagen, 1927; Chundawat and Qureshi, 1999; Shawl and Takpa, 2009; Namgail *et al.*, 2010).

Collection of hair samples:-

Hair found opportunistically stuck on bushes and shed naturally near resting sites and track was collected during the survey. Hair was also collected from carcasses. Fine-tip forceps were used to collect the hair samples, which were transferred to transparent ziplock packets for analysis and geo-referenced accordingly.

Tricho-morphometric analysis:-

The characteristics of guard hairs are known to differ from one mammalian species to another (Brunner and Coman, 1974; Kondo, 2000). The morphological characteristics of the cuticle and medulla of a guard hair can be used to identify the species (Brunner and Coman, 1974; Kondo, 2000, Taru *et al.*, 2013). The tricho-morphometric approach was used to identify species and assess the presence and assemblage of species in the CCV (Table. 1). All the hair samples (n=43) were macroscopically screened for the presence of guard hairs. At least five guard hairs per sample were analyzed according to established protocols (Sahajpal *et al.*, 2009; Bahuguna *et al.*, 2010). The macroscopic and microscopic hair characteristics were studied to assess the biological origin (Sahajpal *et al.*, 2009; Bahuguna *et al.*, 2010). Both qualitative and quantitative (macroscopic and microscopic) hair characteristics (Table 2) of each hair were recorded. The colour, shape and texture of each guard hair were noted. The morphological characteristics (cuticular pattern, including the scale margin, scale distance and scale pattern, and medullary patterns) were observed and photo-micrographed using a comparison microscope (Leica DMR). The hair thickness and medulla thickness were determined. The medullary index was calculated using the following formula:

$$\text{Medullary Index (MI)} = \frac{\text{Hair Thickness (MT)}}{\text{Hair Thickness (HT)}}$$

The observed tricho-morphometric characteristics were compared with those of reference slides in the hair repository of the Wildlife Institute of India, Dehradun. The quantitative data were statistically analysed. A three-dimensional scatter plot between HT, MT and MI was generated using SPSS version 19.0 (IBM, 2010).

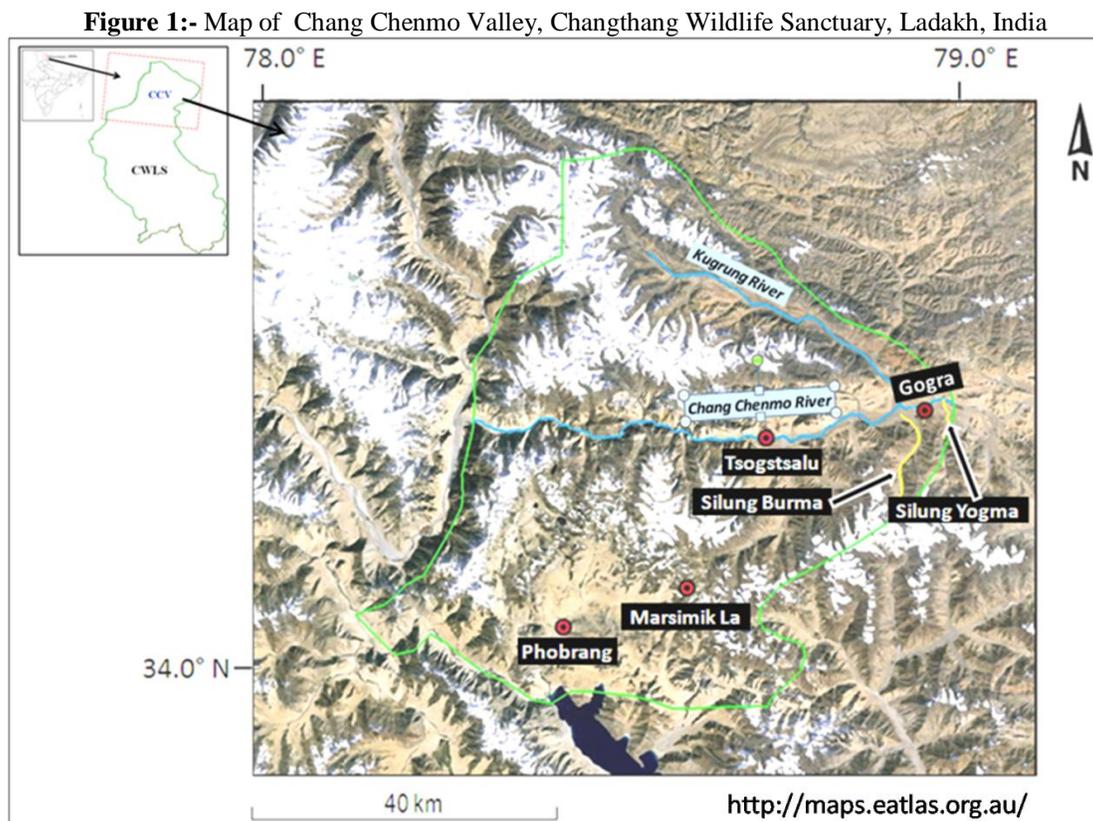
Results and discussion:-

A total of five large wild herbivore species were sighted in the CCV during the study period. Four of these belong to the order Artiodactyla and one belongs to the order Perissodactyla (Table 1). All five species are conservation-reliant and significant components of the TP ecosystem. The occurrence of these species reveals the conservation importance of the area. The status of a landscape and its ecosystem can be determined by evaluating the distribution pattern of the large herbivores of the area. Guard hairs could be obtained from 39 samples of the 43 hair samples that were collected from CCV. Thus, the success rate of getting dorsal guard hairs for tricho-morphometric analysis from opportunistically found hair samples was 90.7%. The relative frequency of occurrence of hairs of large wild herbivore species in randomly collected hair samples (n=39) from the CCV region was *Equus kiang* (74.4%) > *Pantholops hodgsonii* (10.3%) > *Ovis ammon hodgsonii* (7.7%) > *Pseudois nayaur* (5.1%) > *Bos mutus* (2.6%) (Table 2). Details of the qualitative and quantitative hair characteristics are provided in Table 3. Photo-micrographs of the cuticular scale casts and medullary patterns are shown in Figure 2.

High-altitude mammals have characteristic wavy and kinky hair along with peculiar brittleness (Sahajpal, 2009). Hair of *P. hodgsonii*, *P. nayaur*, *O. a. hodgsonii* and has the characteristics kinks, while these are absent in the hair of *B. mutus* and *E. kiang* (Table 3). The cuticle scale margins are crenate in *B. mutus* and smooth in the other four species (Table 3). The cuticle scales were close in *O. a. hodgsonii*, *B. mutus* and *E. kiang*, while they were far apart in *P. hodgsonii* and *P. nayaur* (Table 3). The value of HT was greatest in *O. a. hodgsonii*, followed by *P. nayaur*, *E. kiang*, *P. hodgsonii*. It was least in *B. mutus*. The value of MT was greatest in hairs of *O. a. hodgsonii*, followed by *P. nayaur*, *E. kiang* and *P. hodgsonii*, being least in *B. mutus*. The highest value of MI of guard hairs was observed in *P. hodgsonii* and *O. a. hodgsonii*, followed by *E. kiang*, and the lowest value was in *B. mutus*. The three-

dimensional scatter plot generated between HT, MT and MI also shows a distinct demarcation between the hair characteristics of the five large wild herbivore species found study area (Fig. 3).

Our tricho-morphometric studies confirm the presence of five large wild herbivore species, namely the Tibetan antelope (*P. hodgsonii*), the blue sheep (*P. nayaur*), the Tibetan argali (*O. a. hodgsonii*), the wild yak (*B. mutus*) and the Tibetan wild ass (*E. kiang*) in the study area. The distinct characteristics allow discrimination of these five species. It is evident from the present study that tricho-morphometry play a significant role in identifying the biological origin of the species and indirectly confirm the presence of species in an area. The results of the present study also support the findings of previous studies (Mallon, 1998; Bhatnagar *et al.* 2007; Namgail, 2009; Shawl and Takpa, 2009; Namgail *et al.*, 2010). Although some studies have been conducted previously (Mallon, 1998; Bhatnagar *et al.* 2007; Namgail, 2009; Shawl and Takpa, 2009; Namgail *et al.*, 2010) to assess the assemblage of species, this was the first time an attempt was made using the tricho-morphometric approach.



The present study highlights the significance of tricho-morphometry in identifying species from opportunistically found hair samples and assessing the biological diversity of an area. The present study confirms the presence of the species identified and supports the determination of the worldwide distribution range of these threatened species in the TP. Therefore, we suggest hair be collected along with GPS locations during surveys undertaken for direct evidence in inhospitable habitats for subsequent use in estimating the relative abundance and occupancy. The tricho-morphometry of the mountain ungulates that has been described may also be very useful in studies undertaken to determine predator-prey relationships.

Figure 2:- Hair characteristics (cuticular and medullary) of five large wild herbivores of Chang Chenmo Valley, Changthang Wildlife Sanctuary, Ladakh, India

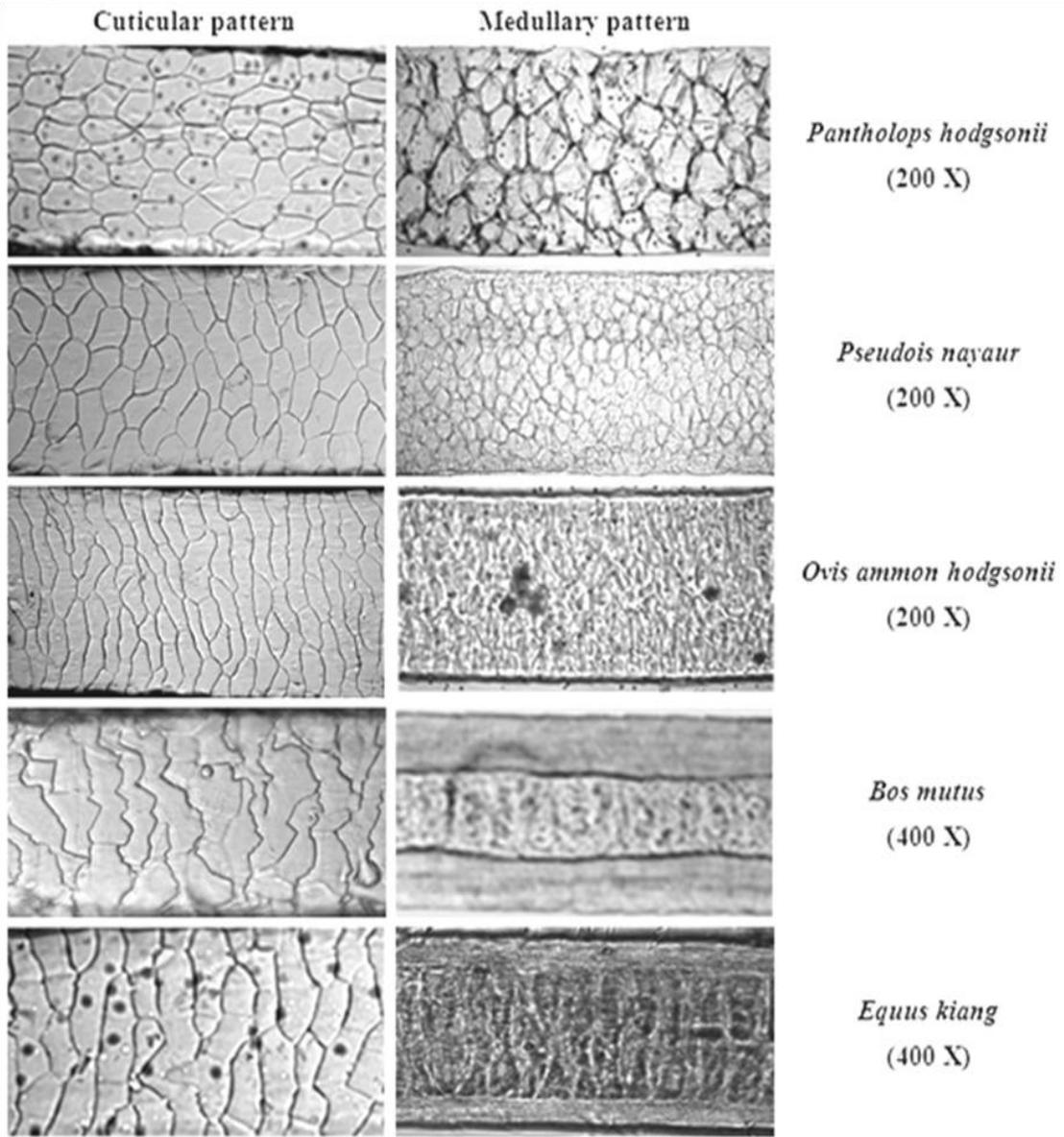


Figure 3:- A three-dimensional scatter-plot showing the clear distinction between values of three hair parameters (HT, MT and MI) of the five large wild herbivore species of Chang Chenmo Valley, Changthang Wildlife Sanctuary, Ladakh, India.

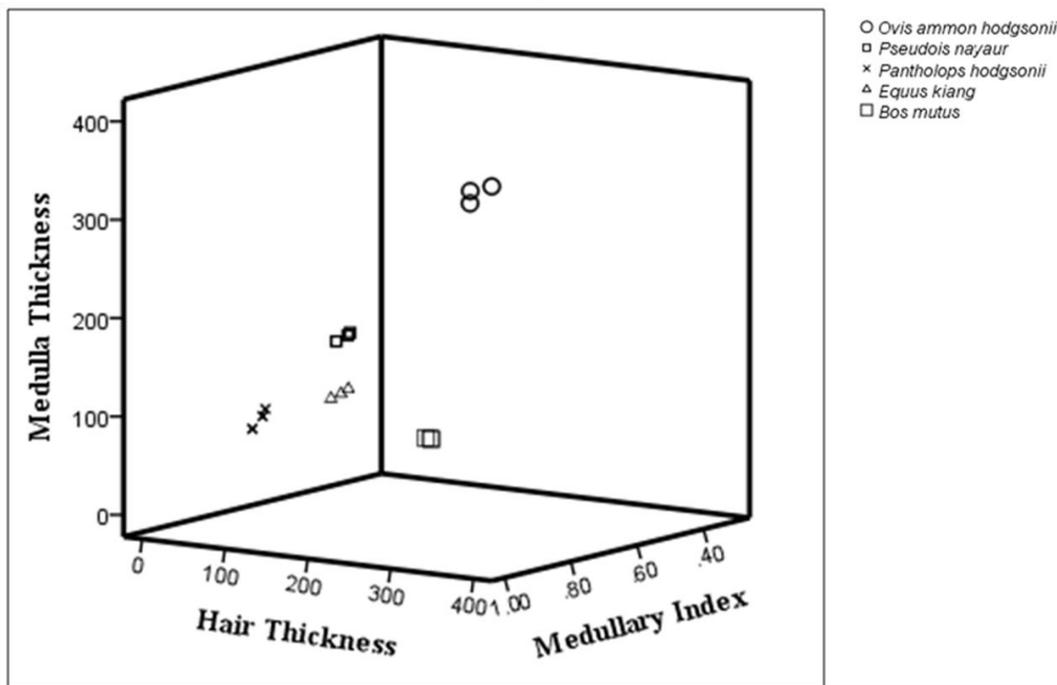


Table 1:- List of large wild herbivores of Chang Chenmo Valley, Changthang Wildlife Sanctuary, Ladakh, with their conservation status

| S.No. | English Name | Scientific Name | Conservation status | | |
|-------|---------------------------|---|---------------------|----------|-----------|
| | | | IUCN | CITES | WPA, 1972 |
| 1. | Tibetan Antelope or Chiru | <i>Pantholops hodgsonii</i> (Abel, 1826) | EN | App.-I | Sch.-I |
| 2. | Blue Sheep or Bharal | <i>Pseudois nayaur</i> (Hodgson, 1833) | LC | App.-III | Sch.-I |
| 3. | Tibetan Argali | <i>Ovis ammon hodgsonii</i> (Blyth, 1841) | NT | App.-I | Sch.-I |
| 4. | Wild Yak | <i>Bos mutus</i> (Przewalski, 1883) | VU | App.-I | Sch.-I |
| 5. | Tibetan Wild Ass | <i>Equus kiang</i> (Moorcroft, 1841) | LC | App.-II | Sch.-I |

Table 2:- Relative frequency of occurrence of hair of large wild herbivore species in randomly collected hair samples (n=39) during survey in the Chang Chenmo Valley, Ladakh

| Species | % occurrence of species |
|-----------------------|-------------------------|
| <i>P. hodgsonii</i> | 10.3 |
| <i>P. nayaur</i> | 5.1 |
| <i>O.a. hodgsonii</i> | 7.7 |
| <i>B. mutus</i> | 2.6 |
| <i>E. kiang</i> | 74.4 |

‡ IUCN - International Union for Conservation of Nature and Natural Resources; CITES - Convention on International Trade in Endangered Species of Wild Flora and Fauna; WPA – Wildlife (Protection) Act, 1972; EN – Endangered; LC - Least Concern; NT - Near Threatened; VU – Vulnerable; App. – Appendix; Sch.-Schedule

Table 3:- Tricho-morphometrics of large wild herbivore species encountered in the Chang Chenmo Valley region, Changthang Wildlife Sanctuary, Ladakh

| | Species | | | | |
|---|---|------------------------|-----------------------------|---------------------------|------------------------|
| | <i>Pantholops hodgsonii</i> | <i>Pseudois nayaur</i> | <i>Ovis ammon hodgsonii</i> | <i>Bos mutus</i> | <i>Equus kiang</i> |
| (a) Qualitative hair characteristics | | | | | |
| Macroscopic | | | | | |
| Hair colour | Light tan | White | Greyish white | Black | Brown and muddy white |
| Hair shape | Short and wavy with undulations and kinks | Long with kinks | Long with kinks | Long and straight | Long and straight |
| Hair texture | Soft | Soft and smooth | Rough | Rough | Soft |
| Microscopic | | | | | |
| Hair cuticle | | | | | |
| Scale margin | Smooth | Smooth | Smooth | Crenate | Smooth |
| Scale distance | Distant | Distant | Near | Near | Near |
| Scale pattern | Regular mosaic | Regular wave | Regular wave | Irregular wave | Regular wave |
| Hair medulla | | | | | |
| Medullary pattern | Wide cellular lattice | Wide cellular lattice | Wide medullary lattice | Narrow and simple medulla | Wide medullary lattice |
| (b) Quantitative hair characteristics (in μm) (mean \pm SE) | | | | | |
| HT | 109.670 \pm 6.741 | 208.00 \pm 3.606 | 374.33 \pm 6.741 | 90.33 \pm 1.202 | 149.67 \pm 4.333 |
| MT | 105.000 \pm 7.000 | 197.00 \pm 2.646 | 360.33 \pm 5.783 | 36.00 \pm 1.155 | 122.00 \pm 2.887 |
| MI | 0.960 \pm 0.005 | 0.95 \pm 0.004 | 0.96 \pm 0.008 | 0.40 \pm 0.008 | 0.82 \pm 0.004 |

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[§] Legend: HT – Hair Thickness, MT – Medulla Thickness, MI – Medullary Index

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