



## Analysis of Keratin-Associated Protein-7 (KRTAP7) protein structure and function in Indian dromedary camel (*Camelus dromedarius*)

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### ABSTRACT

Studies on regulatory genes and proteins responsible for variation in hair quality attributes amongst Indian dromedary camel breeds are lacking. Keratin and keratin-associated proteins enable trichocytes, the hair-producing cells, to produce hair fibers. These proteins also specify the distinct molecular and structural properties of hair, fur, wool, and quills of the various animal species. The molecules of hair-keratin proteins are cross-linked by the keratin-associated proteins (*KRTAPs*), which are responsible for important properties of hair fibers, such as their thickness and curliness for its mechanical and physical characteristics. The amino acid sequence of KRTAP7 gene of Bikaneri, Kachchi, Mewari and Jaisalmeri camel breeds were subjected to bioinformatics analysis using various bioinformatics software for determining the biophysical properties, the two-dimensional hydrophobicity or hydrophilicity plots, subcellular localization of the KRTAP7 protein and the protein-protein interaction. Prediction of various post-translational modifications (PTMs) for the KRTAP7 protein was done with specific server tools based on the neural network model. Prediction of secondary and three-dimensional (3-D) structures of KRTAP7 protein was also done. All four breeds shared identical KRTAP7 gene sequence with 87 amino acids long KRTAP7 protein. The KRTAP7 protein amino acid sequence revealed 13 phosphorylation sites, S-S bonding patterns, and glycosylation sites besides high hydrophilicity, high proportion of specific amino acids, absence of signal peptides, and internal acetylation sites. The protein-protein association networks predicted both direct and indirect interactions with other regulatory genes and proteins essential for the process of keratin biosynthesis and hair characteristics.

**Keywords:** Camel Hair, Dromedary camel, Glycosylation sites, *Insilico*, KRTAP7 protein

The mammalian integumentary system represents some unique structural morphological features by having various adaptive appendages which evolved and got modified along with domestication and civilization patterns. The upper non-vascularized epidermal and underlying vascularized dermis layer of integumental tissues synthesize the keratinized fibrous products such as skin, hair, hoof and horn in livestock species. The skin and skin appendages *viz.* scales, feathers and hairs as well as varying coat thickness in different livestock species are vital for sound protective functioning (Chuong and Homberger 2003). The protective covering of hairs fiber is produced by skin cells embedded in hair follicles in mammals. The quality and quantity of these integumentary fibers are important for insulation,

visual appearance of the animal and for human use in cottage industry. Among two anatomical forms of these hair follicles the secondary hair follicles are commercially important in fiber/wool producing animals. The primary factor influencing both quantitative (follicle counts and density) and qualitative (staple length, medullation, and fineness) characteristics that determine the commercial aspect of these fibers is fibrous keratin and the proteins that are linked to it (Galbraith 2010). The fibrous keratin protein comprises the inner most medullary layer of hair, (Lee *et al.* 2022). The keratin cytoskeleton of hair fiber promotes hair growth and provide strength to already grown hairs in skin.

The quality of hair fibre is a quantitative trait having polygenic effect. The keratin and keratin associated proteins (*KRTAPs*) have a diversified family such as in human, it has 93 proteins classified in 26 sub-families (Litman and Stein 2023). The family of *KRTAPs* have two main groups in which one is a cysteine rich group while another is a high glycine-tyrosine group and play essential roles in the formation of rigid and resistant hair shafts. The expression and synthesis of *KRTAPs* family protein is under the regulation of respective gene(s). Different animal species

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*viz.* starfish, sea anemone, snails, fish, amphibians, reptiles and birds have some modification in integument, hence, the principal hair protein *KRTAPs* has a vital physiological role in both hair production and thermoregulation by some physiological changes.

Hair is a universally accepted and demanded animal product without any social taboo and has high shelf-life. In the cottage economy, camel hair is used to make useful items like rope, blankets, floor rugs, bags, mattresses, and other items that appeal to tourists (Khanna *et al.* 1990). The hair fiber obtained from camel have nearly equal worth as of any other wool producing species (sheep, angora rabbit *etc.*). Utility products made from camel hair are similar to wool in that they are strong, resilient, warm, and have low conductivity, when compared for 620 gram and 900 gm weights of camel hair and pure wool fabric, respectively (Khanna and Rai, 1991). Blending pure camel hair with other soft materials can enhance its quality and manufacturing capacity for efficient use.

There is only limited study on the candidate genes targeting the hair production traits in camelid species to improve the commercial production and textile market of camel hair fiber unlike sheep wool industry. By using genotyping by sequencing (GBS), Calderon *et al.* (2021) annotated a group of SNPs at or close to potential genes for fiber quality and color (*KRT*, *KRTAP*, *ASIP*, *MC1R*, *TYRP1*) from samples of four farms in Huacaya alpacas. In addition, Allain and Renieri (2010) also concluded that a few keratin (*KRT*) and keratin-associated protein (*KRTAP*) genes are significant candidate genes for fleece and fiber quality. Yadav *et al.* 2024 reported *KRTAP7* gene sequence (OR243225.1) of four Indian dromedary camel. However, the structure and function of *KRTAP7* protein has not been reported for Indian Dromedary camel. Therefore, the goal of this study was *in-silico* analysis of *KRTAP7* protein in Indian dromedary camels to determine the structure and function of Keratin-Associated Protein-7 (*KRTAP7*) protein in four Indian dromedary camel (*Camelus dromedarius*) breeds.

## MATERIALS AND METHODS

*In-silico analysis of the KRTAP7 protein:* The amino acid sequence reported by Yadav *et al.* 2024 for the CDS of the *KRTAP7* gene was subjected to bioinformatics analysis using various bioinformatics software tools with default parameters. The biophysical properties of the *KRTAP7* protein were identified using the ProtParam server of ExPASy, SIB Bioinformatics resource portal (<https://web.expasy.org/cgi-bin/protparam/protparam>). The two-dimensional hydrophobicity or hydrophilicity plots were computed using the ProtScale tool of ExPASy, (<https://web.expasy.org/protscale/>). Subcellular localization of the *KRTAP7* protein was visualized using the DeepLoc 2.0 server (<https://services.healthtech.dtu.dk/services/DeepLoc-2.0/>). The protein-protein interaction of consensus translated amino acid sequence of the *KRTAP7* protein was predicted using the STRING database (<https://string-db.org/>).

Prediction of various post-translational modifications (PTMs) for the *KRTAP7* protein was done with specific server tools based on the neural network model. Generic and kinase-specific prediction of phosphorylation sites in the *KRTAP7* protein was done by using NetPhos-3.1 server (<https://services.healthtech.dtu.dk/services/NetPhos-3.1/>). The occurrence and position of signal peptide cleavage sites in respective deduced amino acid sequence of the *KRTAP7* protein was predicted using the SignalP 5.0 server (<https://services.healthtech.dtu.dk/services/SignalP-5.0/>). The internal lysine acetylation sites within the *KRTAP7* protein were predicted using GPS-PAIL 2.0 software (<http://pail.biocuckoo.org/download.php>).

*Prediction of secondary and three-dimensional (3-D) structures of KRTAP7 protein:* Protein structural features *viz.* accessible surface area (ASA), secondary structure, disorder, and phi/psi dihedral angles of amino acids in *KRTAP7* protein sequence were predicted by NetSurfP-3.0 webserver (<https://services.healthtech.dtu.dk/services/NetSurfP-3.0/>). Homology based secondary structure was predicted by applying SOPMA ([https://npsa-prabi.ibcp.fr/cgi-bin/npsa\\_automat.pl?page=/NPSA/npsa\\_sopma.html](https://npsa-prabi.ibcp.fr/cgi-bin/npsa_automat.pl?page=/NPSA/npsa_sopma.html)). Homology modelling was done by using a couple of software packages with their prescribed web instructions *viz.* MODELLER 10.4 (<https://salilab.org/modeller/10.4/release.html>), PHYRE2 (<http://www.sbg.bio.ic.ac.uk/~phyre2/html/page.cgi?id=index>) and SWISS-MODEL (<https://swissmodel.expasy.org/>). Predicted 3-D structures generated for the *KRTAP7* protein by various models were evaluated at different scales by using tools such as Errat, PROCHECK and VERIFY3D (<https://save.mbi.ucla.edu/>). The stereochemical quality and geometrical parameters (phi and psi rotations) were checked for the *KRTAP7* protein by generating the Ramachandran plot. The best generated model with the template model which passed various quality check filters using different computational tools was visualized by using UCSF Chimera (<http://www.cgl.ucsf.edu/chimera/>). A window version of RasMol software (RasWin) was used to observe different predicted protein models of *KRTAP7* protein.

## RESULTS AND DISCUSSION

*Bio-physicochemical analysis of protein sequences:* The entire coding region of camel *KRTAP7* protein was compared using ProtParam with *Camelus dromedaries*, *Camelus bactrianus*, *Camelus ferus*, *Vicugna pacos*, *Homo sapiens*, *Mus musculus*, *Equus caballus*, *Sus scrofa*, *Bubalus bubalis*, *Bos indicus*, *Capra hircus* and *Ovis aries* (Table 1). The stop codons (UGA and UAG) encoding rare 21<sup>st</sup> and 22<sup>nd</sup> amino acids Selenocysteine (Sec) and pyrrolysine (Pyl) as well as Gln (Q), Glu (E) and Lys (K) amino acids were absent in *KRTAP7* protein in all considered mammalian species. The camel *KRTAP7* protein has variations in composition of essential amino acids (large percentage: cysteine, leucine, tyrosine, arginine and glycine; small percentage: aspartic acid, isoleucine and methionine; absent: histidine, lysine, glutamine and

Table 1. Description of the *KRTAP7* gene sequence from different mammalian species

S. No.	Species	Reference acc no.	Chr no.	Accession no. (Protein)	Gene (bp)	CDS (bp)	Protein (Amino acids)
1	Camelus dromedarius	NC_044511.1	1	KAB1284088.1	732	264	87
2	Camelus bactrianus	NW_011514130.1	Un	XP_010954900.1	730	264	87
3	Camelus ferus	NC_045696.1	1	EQB78523.1	729	264	87
4	Vicugna pacos	NW_021964153.1	1	XP_006216075.1	734	264	87
5	Homo sapiens	NC_000021.9	21q22.11	NP_853637.2	721	264	87
6	Mus musculus	NC_000082.7	16; 16 C3.3	NP_082047.1	621	264	87
7	Equus caballus	NC_009169	26	XP_014591977.1	739	264	87
8	Sus scrofa	NC_010455.5	13	XP_003358967.2	597	258	85
9	Bubalus bubalis	NC_059157.1	1	XP_006053907.3	740	264	87
10	Bos taurus	KJ551549.1	1	AHZ89844.1	285	264	87
11	Capra hircus	NC_030808	1	QES86378.1	707	258	85
12	Ovis aries	NC_056054	1	QPP12018.1	676	258	85

acc. no.= Accession number; Chr no.= Chromosome number; Un= Unknown; bp=base pairs; CDS= Coding sequence

glutamic acid). Amino acid frequencies (%) and numbers revealed numbers and percentage of some important proteins *viz.* Aspartic acid (Asp), Cysteine (Cys), Glycine (Gly), Histidine (His) and Isoleucine (Ile) were different in camelids for *KRTAP7* protein. The high frequency of RGD (Arginine-Glycine-Aspartic acid) motifs in camel *KRTAP7* protein are responsible for cell adhesion sequences in the water-soluble keratin (Aboushwareb *et al.* 2009). The cysteine provide the antioxidant property to cure many skin ailments and provide mechanical strength to the hair fibres due to higher level of disulfide (S-S) group cleavage in the black hair compared to the white hair (Piste 2013). Histidine and isoleucine amino acids play a role in strength and proper growth of hair-fibre.

The important physicochemical properties of *KRTAP7* protein for mammalian species and Indian dromedary

camel are compared in Table 2 and 3, respectively. The molecular mass of protein in analysis was ranged between 9078.06-9515.63 daltons, isoelectric point 8.57-8.91 pI with 10-21 phosphorylation sites and 3-5 and 0-1 positive (Arg + Lys) and negatively (Asp + Glu) charged residues, respectively. The GRAVY values were between -0.264 to 0.007 in the considered species while it was -0.118 for camel *KRTAP7* protein (Table 2 and 3).

The hydrophobicity profile of *KRTAP7* protein indicated a greater number of peaks in negative regions *i.e.*, below "0" value in graph plotted between position of amino acid and hydropath score justify the hydrophilicity of protein with -0.118 GRAVY value. The molecular mass and isoelectric point of Indian dromedary camel were valued in range of other mammalian species (Table 3). The primary structure analysis *viz.* negative GRAVY value, instability

Table 2. Physicochemical properties of *KRTAP7* protein in different mammalian species

Species	Formula	Molecular mass (Daltons)	pI	GRAVY	Asp+Glu	Lys+Arg	Phosphorylation sites
Camelus dromedarius	C <sub>417</sub> H <sub>587</sub> N <sub>109</sub> O <sub>120</sub> S <sub>9</sub>	9335.45	8.68	-0.118	1	5	13
Camelus bactrianus	C <sub>420</sub> H <sub>589</sub> N <sub>111</sub> O <sub>120</sub> S <sub>8</sub>	9369.45	8.75	-0.184	1	5	14
Camelus ferus	C <sub>420</sub> H <sub>589</sub> N <sub>111</sub> O <sub>120</sub> S <sub>8</sub>	9369.45	8.75	-0.184	1	5	14
Vicugna pacos	C <sub>416</sub> H <sub>587</sub> N <sub>109</sub> O <sub>117</sub> S <sub>9</sub>	9275.44	8.89	0.007	0	5	10
Homo sapiens	C <sub>421</sub> H <sub>582</sub> N <sub>106</sub> O <sub>121</sub> S <sub>7</sub>	9288.31	8.57	-0.120	0	3	14
Mus musculus	C <sub>424</sub> H <sub>591</sub> N <sub>111</sub> O <sub>122</sub> S <sub>10</sub>	9515.63	8.67	-0.264	0	4	15
Equus caballus	C <sub>416</sub> H <sub>581</sub> N <sub>105</sub> O <sub>121</sub> S <sub>5</sub>	9149.12	8.78	-0.062	0	3	20
Sus scrofa	C <sub>412</sub> H <sub>579</sub> N <sub>105</sub> O <sub>117</sub> S <sub>10</sub>	9195.36	8.74	-0.042	0	4	18
Bubalus bubalis	C <sub>430</sub> H <sub>581</sub> N <sub>107</sub> O <sub>121</sub> S <sub>7</sub>	9409.41	8.57	-0.160	0	3	16
Bos taurus	C <sub>424</sub> H <sub>582</sub> N <sub>108</sub> O <sub>121</sub> S <sub>8</sub>	9384.42	8.73	-0.178	0	4	18
Capra hircus	C <sub>415</sub> H <sub>569</sub> N <sub>105</sub> O <sub>118</sub> S <sub>6</sub>	9109.08	8.91	-0.146	0	4	20
Ovis aries	C <sub>415</sub> H <sub>568</sub> N <sub>104</sub> O <sub>117</sub> S <sub>6</sub>	9078.06	8.91	-0.125	0	4	21

Asp+Glu: negatively charged residues; Lys + Arg: positively charged residues; pI: Isoelectric point

Table 3. The physico-chemical properties of the *KRTAP7* protein in Indian dromedary camel

S.N.	Parameters	Value
1	Molecular weight (Dalton)	9335.45
2	Isoelectric point (pI)	8.68
3	Total number of negatively charged residues (Asp + Glu)	1
4	Total number of positively charged residues (Arg + Lys)	5
5	Formula	$C_{417}H_{587}N_{109}O_{120}S_9$
6	Number of atoms	1242
7	EC range (in units of $M^{-1} cm^{-1}$ , at 280 nm measured in water)	18910-19410
8	EC Abs 0.1% (=1 g/l) range	2.026-2.079
9	Estimated half-life (mammalian reticulocytes, in vitro)	30 hours
10	Instability index (II)	29.02
11	Aliphatic index (AI)	35.86
12	GRAVY value	-0.118

Asp+Glu: negatively charged residues; Lys + Arg: positively charged residues; pI: Isoelectric point; EC: Extinction coefficients

index (II; <40), low aliphatic index (AI; 35.86) and half-life (30 hours) in Indian dromedary camel indicates that the *KRTAP7* protein is a non-polar, stable, low thermal tolerant and with high hydrophilic residues (Wang *et al.* 2022) (Table 3). Hydrophilicity in a polypeptide chain determines its interaction with water and low thermal stability of camel *KRTAP7* protein. In accordance of the present work, the keratin family members in Yak were non-polar (GRAVY<0) and stable (II; <40) while the other physiochemical properties *viz.* molecular weight (34262.85 Da to 64015.55 Da), EC (14,450 to 52385), AI (57.77 to 91.92) and isoelectric points (4.25 to 8.65 pI) were in contrast to our study (Bao *et al.* 2022).

No known motifs and no protein family membership was predicted using PROSITE and EBI-Inter pro scan tool respectively. Biosequence analysis revealed that the camel *KRTAP7* protein was a member of *KRTAP* type 7 family which is present in interfilamentous matrix of hair cortex. A total of 70 significant query matches from different species were identified for targeted *KRTAP7* protein. The camel *KRTAP7* protein have one low complexity region (12aa: GYSSLGYSFGGS) and two domains (SCOP d1dlja1: 17aa-SNIGNLGCYGGGFCRP; SCOP d1b78a: 9aa-SNIGNLGCYGGGFCRP). The taxonomic lineage was established for the camel *KRTAP7* sequence with other organism(s) in the absence of definite domains from database. The camel *KRTAP7* protein belongs to a highly conserved protein family which shows similarity with the keratin associated proteins in different mammalian species due to its emergence by divergence before the mammalian radiation in late evolution of taxonomic strata. The

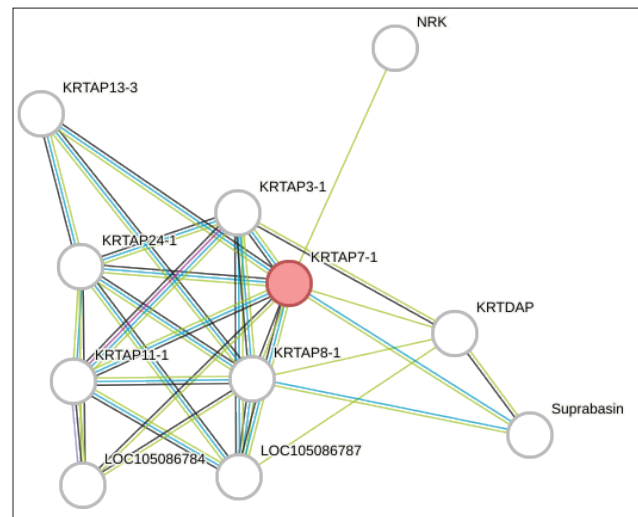


Fig. 1 The protein interaction network analysis for the *KRTAP7* protein of Indian dromedary camel.

abundance of cross-linking disulphide due to high-sulphur content of cysteine residue and high-glycine-tyrosine containing keratins protein is an important intermediate filament of interfilamentous matrix which is essential for the rigidity and resistant of hair-fibre.

STRING database was used to identify the interacting proteins network with closely related and available database of camel *KRTAP7* proteins (Fig. 1). The interacting network of *KRTAP7* protein higher interaction scores for various proteins such as *KRTAP24-1* (0.929), *KRTAP11-1* (0.929), *KRTAP8-1* (0.914), *LOC105086784* (0.859), *LOC105086787* (0.848), *KRTAP3-1* (0.670), *KRTDAP* (0.606), *NRK* (0.597), *KRTAP13-3* (0.488) and Suprabasin (0.467). The proteins are directly or indirectly related with keratin protein biosynthesis process for hair-fibre.

Among the predicted proteins from interacting network some are directly involved in keratin synthesis process (*KRTAP24-1*, *KRTAP11-1*, *KRTAP8-1* and *KRTAP13-3* proteins) while others have indirect role (*LOC105086784*, *LOC105086787*, *KRTDAP*, *NRK* and Suprabasin proteins) for hair characteristics (Fig. 1). The *KRTAP* gene repertoire which express keratin genes and its associated proteins (*KRTs* and *KAP*) are vital for hair characteristics such as physical-mechanical properties (strength, resistance and stiffness), relative composition, interactions and quantitative fibre traits (fibre diameter, medullation%, staple length) in mammalian species (Anello *et al.* 2022; Pérez-Cabal *et al.* 2010). The *KRTDAP* gene helps in differentiation and maintenance of keratin producing cells (keratinocyte) of stratified epithelia (Su *et al.* 2021). The *NRK* and Suprabasin (*SBSN*) are tumor-suppressor genes, also act as a component of the cornified envelope of stratified epithelial cells (Pribyl *et al.* 2021; Denda *et al.* 2019).

*Subcellular localization and post-translational modifications (PTM) of KRTAP7 protein:* The *KRTAP7* is a soluble and extracellular located protein (Table 4).

Table 4. Probability of cell localization of *KRTAP7* protein

Localization	Probability
Cytoplasm	0.2631
Nucleus	0.2428
Extracellular	0.7069
Cell membrane	0.1494
Mitochondrion	0.2164
Plastid	0.0796
Endoplasmic reticulum	0.0458
Lysosome/Vacuole	0.1301
Golgi apparatus	0.0529
Peroxisome	0.0246

The amino acid residues are not varying in location and exposed outside the cell membrane for *KRTAP7* protein. A total of 10-21 phosphorylation sites were predicted in *KRTAP7* protein of considered mammalian species. The phosphorylation sites (13) in camel *KRTAP7* protein were lower which involved phosphorylation of 4-serines, 5-threonines and 4-tyrosine residues. In addition, Arginine/Lysine signal peptide cleavage and propeptide cleavage sites were absent in amino acid sequence of camel as well as in other proteins in analysis. The internal acetylation sites were absent for *KRTAP7* protein of all considered species at high, medium and low threshold. The 8 sulphur containing cysteine residues (positions: 6, 7, 16, 32, 44, 46, 66 and 73) indicates high probability of four disulphide bond (S-S) patterns (between pairs: 6-32, 7-46, 16-44 and 66-73) in primary sequence of camel *KRTAP7* protein (Fig. 2). One O-glycosylation site was detected between 35-40 amino acid sequence in camel *KRTAP7* protein.

Both PTMs activities presence (phosphorylation, S-S bonding patterns and glycosylation sites) and absence (internal acetylation, signal peptide cleavage) have significance to modify the phenotypes and biological processes of cells. The PTMs affect the regulatory and functional characteristics of *KRTAP7* protein with its localization, stability, charges and interactions with other biomolecules. The glycosylation and phosphorylation are most important PTMs which play vital role in conformation and activity of camel *KRTAP7* protein by affecting filament

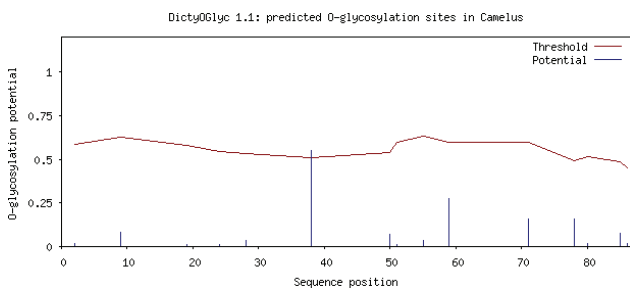


Fig. 2 Predicted O-glycosylation site for *KRTAP7* protein in Indian dromedary camel.

Table 5. Helix propensity of prediction of secondary structure of *KRTAP7* protein in Indian dromedary camel

Structural arrangement	Number (%)
Alpha helix (Hh):	2 (2.30%)
3 <sub>10</sub> helix (Gg):	0 (0.00%)
Pi helix (Ii):	0 (0.00%)
Beta bridge (Bb):	0 (0.00%)
Extended strand (Ee):	20 (22.99%)
Beta turn (Tt):	16 (18.39%)
Bend region (Ss):	0 (0.00%)
Random coil (Cc):	49 (56.32%)
Ambiguous states (?):	0 (0.00%)
Other states:	0 (0.00%)

solubility and regulating keratin binding with other proteins (Ramazi and Zahiri 2021).

*Prediction of secondary structures of proteins:* The absolute surface accessibility (ASA) and relative surface accessibility (RSA) value ranged between 0.103 to 0.803 and 17.27 to 211.19 respectively for camel *KRTAP7* protein. The secondary structure of *KRTAP7* protein was dominated by the random coil (56.32%) followed by extended strands (22.99%), beta-turn (18.39%), alpha helix (2.30%) and absence of other structures like Pi helix, Beta Bridge, Beta region, ambiguous states, etc. (Table 5).

*Prediction of three-dimensional (3-D) structures of proteins:* The final 3-D structure of camel *KRTAP7* protein was predicted with appropriate template by homology modelling (Table 6).

*Ramachandran plot and superimposition of three-dimensional (3-D) protein structure:* The predicted 3-D structure of camel *KRTAP7* protein was validated based on different specificities for all generated models using different structure analysis and verification approaches (Table 7).

The P2 model comparatively passes the maximum validation parameters which was visualized through red, yellow and white coloured Ramachandran plot indicating most favoured (59.5%), additional allowed (32.4%) and disallowed (8.1%) regions respectively in camel *KRTAP7* protein (Fig. 3 A). The phi and psi

Table 6. Template specification criterion for the *KRTAP7* protein in Indian dromedary camel

Parameters Software	Template	Template specification criterion
SWISS-MODEL	Alpha Fold G3U062_LOXAF in <i>Loxodonta africana</i> (African elephant)	Query coverage, GMQE score and % identity criterion
Phyre2	c1pdiQ: short tail fibres into t4 baseplate	Alignment coverage, percent identity and confidence percent

Table 7. Validation properties of different homology models for predicted 3D-structure of the *KRTAP7* protein in Indian dromedary camel

Model (SAVESv6.0)	Errat value (Overall Quality Factor)	Residues in most favoured regions (%)	Residues in additional allowed regions (%)	Residues in generously allowed regions (%)	Residues in disallowed regions (%)	Residues with average 3D-1D score $\geq 0.1$ (%)
Swiss-Model	-	34.5	44.8	20.7	0.0	-
Pyre2	33.34	59.5	32.4	0.0	8.1	66.07

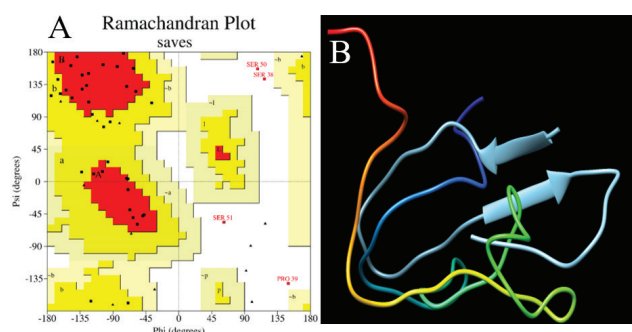


Fig. 3 A. Ramachandran plot generated from best 3-D structure Phyre2 (P3) of *KRTAP7* protein in Indian dromedary camel. B. Superimposition of final *KRTAP7* protein structure of Indian camel using UCSF Chimera.

torsion angles of amino acids and top left quadrant of the Ramachandran plot confirm the stereo chemical properties and helix propensity of camel *KRTAP7* protein. The P2 model was superimposed with highly similar protein model (d1nezg\_12 model) to confirm accuracy of 3-D structure of protein in Indian dromedary camel (Fig. 3 B).

The secondary structures revealed camel *KRTAP7* protein is a non-globular (<30% alpha helix) protein which lack specific stabilizing interactions and having all random conformations for protein folding and binding regions due to higher proportion of the random coils. The amino acids proportion with high level of Gly, Asn and Asp and lower level of Ile, Val and Leu indicates the possible presence of the random coils between two beta strands (Khrustalev *et al.* 2013). The 3-D structure of camel *KRTAP7* protein revealed that the protein is a twisted  $\beta$ -sheet with small segments of  $\alpha$ -helix and the predicted model has better stereochemical properties because only four residues (Ser38, Ser50, Ser51 and Pro39) were present in disallowed regions.

We hypothesize that the non-globular camel *KRTAP* protein provide plasticity to hair fibre by lower molecular weight and high sulphur containing amino acids but unlikely to play a candidate role in quantitative fibre traits such as fibre diameter and medullation%. This could explain the pseudogene effect of camel *KRTAP7* protein for considered hair quality characteristics. The hair-fibre traits might be linked to other candidate gene(s), pathway(s), or network(s). Consequently *KRTAP7*, alone might not be considered as a key determinant of hair quality traits

in Indian dromedary camel. However, the other gene from the family of keratin gene and associated protein (*KRTs* and *KAPs*) needs to be explored for the hair characteristic trait(s).

This maiden report describes Indian dromedary camel *KRTAP7* protein structure and function. The analysis of the *KRTAP7* protein using *in-silico* tools revealed physico biochemical properties, post-translational modifications and protein-protein interactions of this gene in Indian dromedary camel. The predicted amino acid composition and important post-transcriptional modifications of the camel *KRTAP7* protein have been known for their effect on keratin biosynthesis mechanism and are associated with phenotypic variations in hair fibre traits in camel. The secondary and 3D structures of the *KRTAP7* protein revealed important functionally active sites for mutations and protein-protein interactions. The putative interacting protein networks unveiled in the study indicated the role of pleiotropic genes in polymorphic association and/or epistatic interaction for varying hair-fibre quality traits in Indian dromedary camel breeds.

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