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Article

Fine-Scale Population Structure and Relatedness of Argali (*Ovis ammon*) in Kyrgyzstan Revealed by High-Density SNP Data

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Abstract

Argali (*Ovis ammon*), the largest wild sheep in Asia, are of high conservation concern and remain taxonomically and genetically debated across parts of their range. We investigated population structure, relatedness, and inbreeding within Argali sampled in Kyrgyzstan using the Illumina Ovine High-Density SNP array, with an emphasis on dense within-population sampling rather than range-wide comparisons. After quality control, 72 individuals and 135,242 markers were retained for analysis. Principal component analysis revealed subtle genetic variation within the sampled population, but no clustering consistent with discrete subspecies. In particular, we found no genomic support for separating *O. a. polii* and *O. a. karelini* within Kyrgyzstan, suggesting that they represent a single genetic unit in this region. Estimates of identity by descent indicated a high average relatedness (0.35), consistent with harem-based breeding systems typical of wild sheep, while individual inbreeding coefficients averaged near zero, with some evidence of moderate inbreeding in a subset of animals. Together, these results characterize fine-scale genetic structure and kinship within Tian Shan Argali and provide a regional genomic baseline for conservation planning in Kyrgyzstan. Our findings highlight the importance of maintaining connectivity within and among managed populations while acknowledging that broader inference will require sampling across the core Pamir range and other parts of the species' distribution.

Keywords: argali; *Ovis ammon*; conservation genomics; population structure; Kyrgyzstan; Tajikistan



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1. Introduction

Argali (*Ovis ammon* [1]), a species of Asian wild sheep, are among the most iconic mountain ungulates of Central Asia and a priority for conservation. The International Union for Conservation of Nature (IUCN) lists Argali as Near Threatened, citing habitat fragmentation, transboundary barriers, and hunting pressure as major threats [2]. Despite their ecological and cultural importance, Argali taxonomy has historically been debated, and recent genomic work suggests that several traditionally recognized subspecies may not represent distinct evolutionary lineages. Recent phylogenomic work in Caprinae highlights how subspecies distinctions are often not supported at the genomic level, with broad

implications for Argali taxonomy [3]. Hybridization with domestic sheep has also been documented across the range, complicating conservation and management boundaries [4].

Historically, subspecies designations were based on morphometrics, resulting in a confusing and often contradictory taxonomy. Geist [5] recognized seven subspecies, including *O. a. polii* and *O. a. karelini*, while also noting that morphological differences between them were so slight that they could reasonably be combined as a single subspecies. Earlier naturalists, including Lydekker [6,7], also concluded that no consistent differences existed between the two. Nonetheless, *O. a. polii*—often called the Marco Polo sheep—has persisted in the literature and in conservation management as a distinct form, valued for its exceptionally long horns and wide distribution across five Central Asian countries.

Genomic data now allow direct evaluation of these subspecies boundaries. A recent genome-wide SNP analysis by Dotsev et al. [8] found minimal differentiation between *O. a. polii* and *O. a. karelini* (pairwise $F_{st} = 0.083$), in contrast to much greater divergence among other Argali subspecies (e.g., *O. a. ammon* vs. *O. a. severtzovi*, $F_{st} = 0.421$). These findings support the view that Marco Polo Argali and Tian Shan Argali represent a single genetic unit rather than distinct evolutionary lineages. Unlike previous range-wide analyses like Dotsev et al. [8], which compared five Argali subspecies across Central and East Asia using both nuclear SNPs and mitochondrial genomes, our study provides a genome-wide SNP-based assessment using dense within-population sampling within Kyrgyzstan's Tian Shan region, and no morphological or geographical characterization was used in our dataset.

Recent genome-wide studies have challenged long-standing subspecies designations in Argali, suggesting that genetic differentiation among some proposed taxa is weaker than previously assumed. However, most existing work has emphasized broad geographic comparisons rather than dense sampling within individual regions. In contrast, the present study focuses on Argali sampled in Kyrgyzstan, using genome-wide SNP data to evaluate fine-scale genetic structure, relatedness, and inbreeding within a single regional population. The primary objective was to determine whether genetic evidence supports subdivision into distinct management units or whether Argali in this region function as a single population for conservation planning [9]. A secondary objective was to assess whether genomic data collected within Kyrgyzstan support the long-standing distinction between *Ovis ammon polii* [10], and *Ovis ammon karelini* [11], in this regional context.

This study is not intended as a comprehensive taxonomic reassessment of Argali across Central Asia. All samples were collected within Kyrgyzstan's portion of the Tian Shan, and individuals were not classified a priori by morphology or geography. Accordingly, inference is restricted to evaluating genetic structure, relatedness, and inbreeding within this regional population, with taxonomic interpretation limited to the sampled context.

2. Materials and Methods

2.1. Study Population

A total of 88 Argali (*Ovis ammon*) samples were collected in Kyrgyzstan; after quality control filtering, 72 individuals were retained and used in all downstream genomic analyses. Although our sampling includes areas historically associated with both *O. a. karelini* and the northern range edge of *O. a. polii*, it does not include individuals from the Pamir Plateau, which represents the ecological and geographical core of the Marco Polo sheep distribution. As such, our results should be interpreted as representing the Kyrgyzstan populations rather than the full taxonomic breadth of *O. a. polii*. Sample types included desiccated skin, hide punches, liquid blood, Whatman FTA blood cards (Cytiva, Marlborough, MA, USA), bone or horn, and tissue samples. Samples were obtained from Argali harvested under Kyrgyzstan's legal hunting program and from individuals found dead in the field from

unknown causes. DNA was extracted from each sample (see Supplementary Table S1 for metadata). Sampling locations are shown in Figure 1 by sample number for individuals included in the genomic analyses. No morphological or geographical classifications were placed on the data.

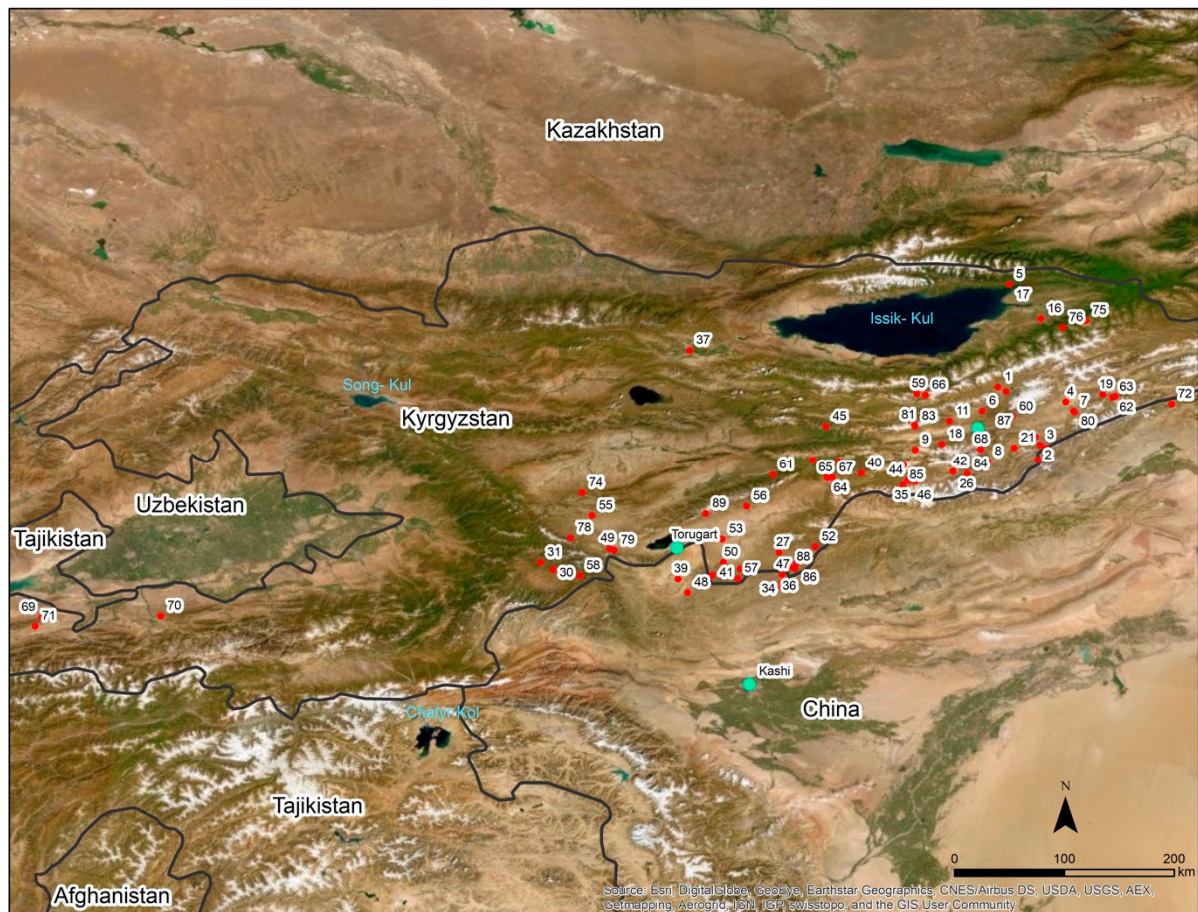


Figure 1. Geographic locations of samples of *Ovis ammon* collected in Kyrgyzstan. Sampling locations are shown by GPS coordinates and labeled by sample number for individuals included in the genomic analyses.

We extracted DNA using the Maxwell 16 LEV Blood DNA Kit (Promega, Madison, WI, USA) for blood and gene card samples, and the SEV Tissue Kit (Promega, Madison, WI, USA) for biopsy punches and tissue samples [12]. Horn and bone samples were processed following Harper et al. [13]. Briefly, a sterile drill bit was used to obtain approximately 20 mg of horn core or bone marrow tissue. The tissue was incubated in digestion buffer (QIAGEN, Hilden, Germany) with proteinase K (QIAGEN, Hilden, Germany) overnight, centrifuged at $12,000 \times g$ for 5 min, and then processed using the DNeasy Blood & Tissue Kit (QIAGEN, Hilden, Germany).

2.2. Genomic Dataset and Quality Control

Argali samples were genotyped using the Illumina High Density (HD) Ovine SNP array (Illumina, Inc., San Diego, CA, USA). All genotyped samples contained a minimum of 300 ng of DNA, a minimum concentration of 20 ng/ μ L, and a 260/280 nm absorbance ratio of at least 1.35. The HD Ovine array includes 606,006 single-nucleotide polymorphisms (SNPs) and was originally developed for domestic sheep (*Ovis aries* [1]), with an average density of one SNP per 4.28 kb. The development process also incorporated genetic material from five bighorn sheep (*Ovis canadensis* Shaw, 1804 [14]) and four Dall's sheep (*Ovis dalli*

Nelson, 1884 [15]) [16,17]. Advances in high-throughput sequencing across mountain ungulates [18,19] provide comparative baselines for assessing adaptive variation in Argali.

Three major groups of Eurasian wild sheep—mouflon, urial, and argali—have been proposed as ancestors of the domestic sheep. More recent genomic analyses suggest that mouflon were the primary ancestors, with repeated introgression of argali alleles, since hybrid offspring are viable and fertile [20,21]. Of the SNPs on the HD Ovine array, approximately 24,000 are informative in Rocky Mountain bighorn sheep (*O. canadensis*) [12,22–24]. Flesch et al. [22] also demonstrated that approximately 25 samples are sufficient to differentiate bighorn sheep populations.

Because the SNP array was developed for domestic sheep, ascertainment bias is expected when applied to Argali; this consideration informed our choice of quality-control thresholds and interpretation of downstream analyses [22].

All Argali samples were shipped to Neogen (Lincoln, NE, USA) for genotyping. Raw genotypes by animal ID and an SNP mapping file aligned to the domestic sheep genome OAR 4.0 [25] were imported into Golden Helix SVS (Golden Helix, Inc., Bozeman, MT, USA; version 24.1) for quality control. First, we filtered samples using a minimum call-rate threshold of 0.75 and removed markers of unknown mapping or located on the sex chromosomes. SNPs were filtered in two stages. First, markers with a minor allele frequency (MAF) < 0.0001 were removed to eliminate monomorphic loci and reduce dataset size prior to downstream processing. Second, a conventional threshold of $\text{MAF} \geq 0.01$ was applied to remove rare alleles with limited power for population-level analyses. Next, we removed poorly performing SNPs by requiring a $\text{MAF} \geq 0.01$ and Hardy–Weinberg equilibrium $p \geq 0.00001$. We performed two separate filtering steps on MAF: our first filter removed SNPs that were fixed in our dataset, thereby reducing computational load for subsequent analysis.

Samples with $< 75\%$ genotyping success across SNPs were excluded. Linkage disequilibrium (LD) pruning was performed on the dataset used for PCA to remove non-independent SNPs, using a window size of 100, a window increment of 25, an LD threshold of $r \leq 0.99$, and the CHM computation method (Huisman et al., 2016) [26]. Linkage disequilibrium pruning for PCA was conducted using an $r \leq$ threshold of 0.99 to retain dense marker coverage for exploratory analyses of subtle structure and relatedness. We emphasize that alternative LD thresholds would alter the weighting of correlated loci but would not change the central conclusion of this study, namely the absence of robust evidence for discrete population subdivision within the sampled Kyrgyzstan Argali. We acknowledge that minimal LD pruning may increase the influence of correlated markers and kinship on PCA results; accordingly, PCA outputs were interpreted in conjunction with relatedness estimates rather than as definitive evidence of population subdivision. An outgroup of Rocky Mountain bighorn sheep was included in exploratory PCA analyses to evaluate the effect of scale on variance structure and to confirm that within-Argali variation was not driven by alignment or analytical artifacts. This resulted in 135,242 informative markers, substantially higher than the 65,158 informative SNPs previously reported in *Ovis ammon* [8].

The filtered dataset was used to conduct PCA for population structure, to perform identity-by-descent pairwise comparisons, and to estimate inbreeding coefficients for each individual using Golden Helix SVS. Raw genotypes and associated metadata are available on DRYAD [27].

2.3. Genomic Data Analysis

We used principal component analysis (PCA) to estimate population stratification. Population stratification is a strong indicator of genetic isolation or subpopulation structure

and has been used previously for similar analyses [12,22,28]. PCA can also be applied to correct for population stratification in association analyses [29]. We conducted PCA both with and without an outgroup of 12 Rocky Mountain bighorn sheep (*Ovis canadensis* Shaw, 1804 [14]) to determine whether subpopulations were present in our dataset. The distantly related outgroup from previous research was used to change the scale of PCA, as its genetic divergence dominates and makes the more subtle population variation visible in comparison. PCA was used as an unsupervised, exploratory tool to assess whether genetic structure emerged without a priori population assignments. Because all samples derive from a single regional population and include related individuals, PCA was not used to define discrete populations but to visualize patterns of variation for management interpretation.

Identity by Descent (IBD) was calculated as a measure of relatedness between individuals. IBD quantifies the proportion of alleles at any marker that are inherited from the same ancestral chromosome [30]. Identity by Descent calculation helps determine which animals are more closely related or have more recently shared a common ancestor. In small or endangered populations, it can indicate reductions in genetic diversity that can lead to impaired reproduction, susceptibility to disease, or reduced ability to adapt to environmental challenges.

Individual inbreeding coefficients were estimated in Golden Helix SVS using a method-of-moments estimator based on deviations between observed and expected heterozygosity across autosomal SNPs. These estimates are reported as individual-level inbreeding coefficients and used as proxies for Wright's within-subpopulation fixation index (F_{is}). Inbreeding coefficients range from -1.0 to $+1.0$, with negative values indicating outbreeding and positive values indicating inbreeding or higher relatedness than expected within the sampled subpopulation [26]. The inbreeding coefficient is the degree of genetic similarity between an individual and itself, or the proportion of its genetic material that comes from the same ancestors on both sides of the family tree. High inbreeding can lead to inbreeding depression, where animals are more likely to suffer from genetic disorders, lower fertility, and reduced survival rates. It can also be used to identify at-risk populations, monitor the impacts of habitat fragmentation, or population bottlenecks.

3. Results

3.1. Population Stratification or Subpopulation Identification by PCA

After quality control, 72 individuals and 135,242 autosomal SNPs were retained for downstream analysis. We used principal component analysis (PCA) to estimate population stratification and to identify potential patterns of variation (Figure 2). PC1 and PC2 explained 12.3% and 9.8% of total variance, respectively. The term 'EV' has been replaced with the percentage of variance explained by each principal component to improve clarity. To examine the genetic distance of potential subgroups, we also performed PCA with an outgroup consisting of 12 randomly selected Rocky Mountain bighorn sheep (*Ovis canadensis*) from previously published research [12]. The PCA indicated several small groups with allele frequency differences from the majority of samples. Several small clusters of individuals were observed along PC1 and PC2, including groups containing individuals sampled from the Kokshaal and Ashirak ridge regions (Figure 2). Because our dataset did not include Pamir samples, our ability to evaluate differentiation within the full *O. a. polii* lineage is limited. The patterns observed here pertain only to the Kyrgyzstan portion of the Tian Shan–Pamir interface. Because all individuals derive from a single regional population and include closely related individuals (see Section 3.2), PCA clustering likely reflects a combination of localized variation and familial relationships rather than discrete population subdivision. From a management perspective, retaining related individuals is informative

because kinship structure is an inherent component of demographic processes in small or socially structured wildlife populations.

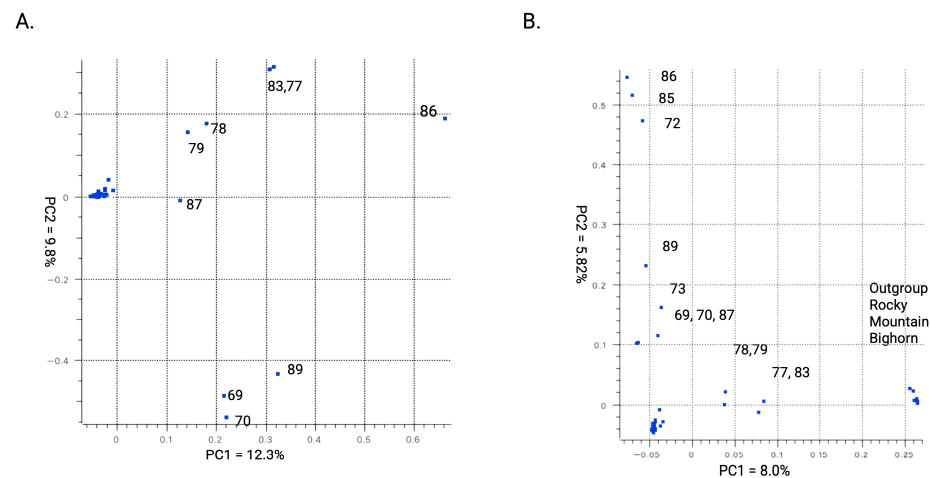


Figure 2. Principal component analysis (PCA) of *Ovis ammon* sampled in Kyrgyzstan. **(A)** PCA of 72 Argali individuals analyzed using 135,242 autosomal SNPs following quality control and linkage disequilibrium pruning ($r^2 = 0.99$). **(B)** PCA including an outgroup of Rocky Mountain bighorn sheep (*Ovis canadensis*) to illustrate the scale of genetic divergence relative to within-Argali variation [12]. Axes show the percentage of total genetic variance explained by each principal component (PC1 = 12.3%, PC2 = 9.8% for panel A; PC1 = 8.0%, PC2 = 5.82% for panel B). PCA was used as an exploratory tool to visualize genetic variation and was interpreted in conjunction with relatedness estimates rather than as evidence of discrete population subdivision.

3.2. Identity by Descent Estimation

Identity by descent (IBD) was estimated between all pairs of samples. Pairwise identity-by-descent estimates averaged 0.35 across all individuals (Figure 3). Values ranged from near zero to levels consistent with close kinship. Clusters observed in PCA corresponded to individuals with elevated pairwise relatedness in identity-by-descent analyses (Figure 3), indicating that kinship likely contributes to the observed structure.

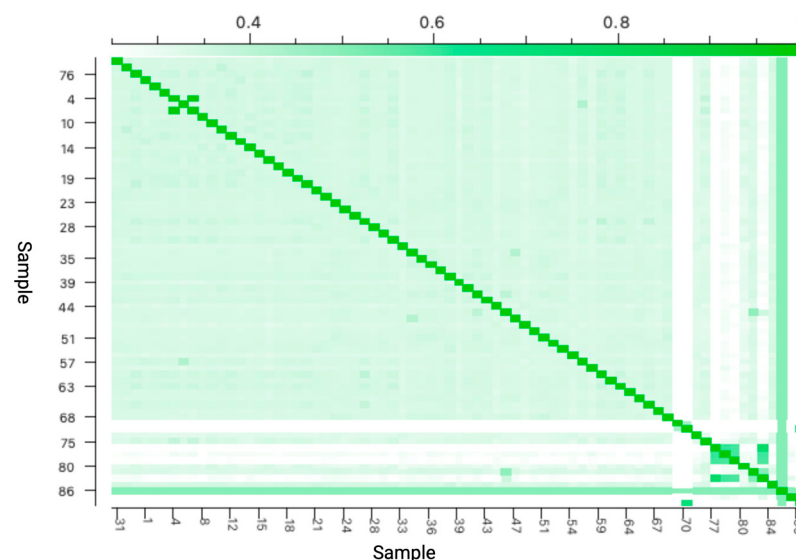


Figure 3. Heatmap of pairwise Identity-by-Descent (IBD) estimates among Argali sheep sampled in Kyrgyzstan. Darker colors indicate pairs of individuals that share a greater proportion of their genome through recent common ancestry, while lighter colors represent more distantly related pairs. Rows and columns correspond to individual sheep, arranged by sampling location, allowing visual comparison of relatedness patterns within and between regions.

3.3. Inbreeding Coefficient Estimation

We calculated inbreeding coefficients (F_{IS}), or Wright's within-subpopulation fixation index, for each sample. Individual inbreeding coefficients ranged from negative values to moderately positive values, with a mean of 0.03 and a median of 0.10 (Figure 4). The average observed heterozygosity was 20,654 markers or 15.54% with an average expected heterozygosity of 21,693 markers or 16.32% (Supplementary Table S1).

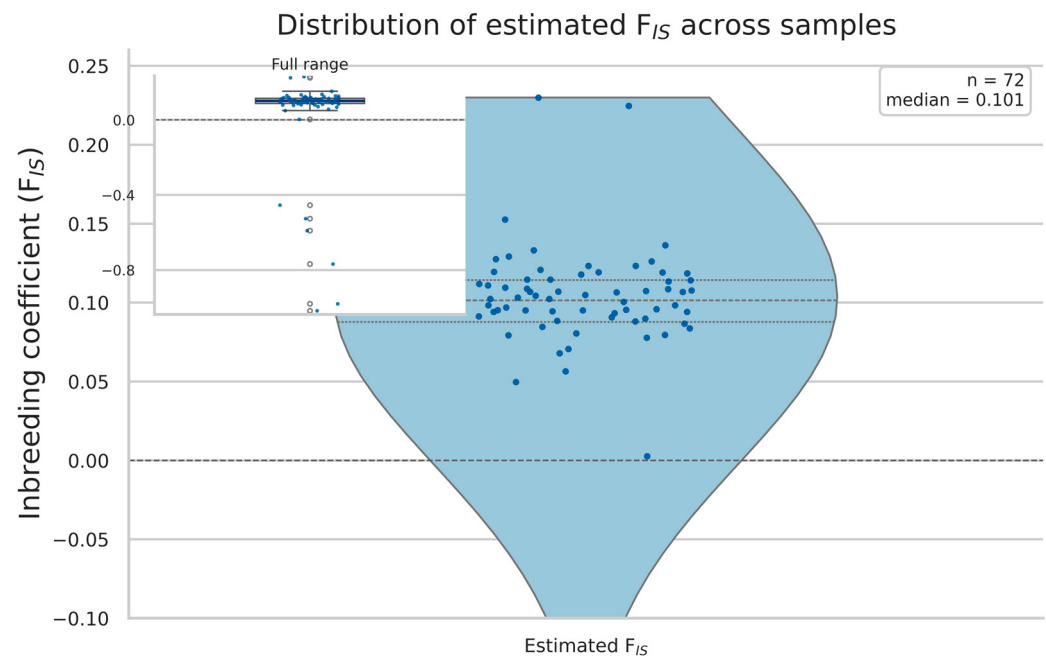


Figure 4. Distribution of individual inbreeding coefficients F_{IS} for *Ovis ammon* sampled in Kyrgyzstan. The violin plot depicts the density of individual F_{IS} estimates, with points representing single animals and the internal line indicating the median. The dashed horizontal line denotes $F_{IS} = 0$ (Hardy–Weinberg expectation). Most individuals exhibit modest positive F_{IS} values, indicating slightly reduced heterozygosity relative to random mating, while a small number of individuals show strongly negative values consistent with excess heterozygosity. The inset displays the full range of F_{IS} values to highlight rare extreme observations.

4. Discussion

Our genomic analysis of Argali from Kyrgyzstan provides new insight into within-region genetic variation, relatedness, and inbreeding in this iconic mountain ungulate. Importantly, this study is designed as a regional assessment based on dense within-population sampling in Kyrgyzstan's Tian Shan, rather than a range-wide evaluation of Argali across Central Asia. Accordingly, conclusions about taxonomy and connectivity are restricted to the sampled Kyrgyzstan context, and broader inference will require additional sampling from the Pamir Plateau and other portions of the species' distribution [8,31].

Principal component analysis (PCA) revealed subtle patterns of genetic variation among individuals, including several small clusters. Because our dataset represents a single regional population and includes close relatives, these clusters are most parsimoniously interpreted as reflecting a combination of localized demographic structure and kinship rather than discrete population subdivision. Such effects are well documented in wild mammal populations with family-based social structure and limited dispersal [32]. In addition, our PCA retained a dense marker set with minimal LD pruning ($r^2 = 0.99$), which can increase the influence of correlated loci and amplify family structure; therefore, PCA is best viewed here as an exploratory visualization tool interpreted alongside relatedness estimates rather than as definitive evidence of population subdivision [26,32].

Taxonomic confusion has long challenged Argali conservation [5–7], with the status of *O. a. polii* and *O. a. karelini* particularly debated. Our results, consistent with Dotsev et al. [8], suggest minimal differentiation among putative subspecies within the sampled Kyrgyzstan population and do not support subdivision into distinct genetic units based on subspecies labels in this regional context. However, these findings do not constitute a comprehensive taxonomic reassessment across the full range of either form [8].

The elevated average relatedness observed in this study (mean IBD = 0.35) indicates substantial sharing of recent ancestry among individuals. This level of relatedness is consistent with herd-forming ungulates that exhibit dominant male or harem-based mating systems, where reproductive skew can elevate within-population kinship [26]. Such social structure can also contribute to small clusters in ordination analyses without implying long-term demographic isolation [26].

Despite elevated relatedness, mean individual inbreeding coefficients were near zero, indicating that close-kin matings are not pervasive across the sampled Kyrgyzstan population. At the same time, the presence of individuals with moderate inbreeding coefficients suggests that localized inbreeding may occur under certain demographic or landscape conditions, particularly if connectivity among subareas is reduced. This combination of high relatedness and generally low inbreeding is consistent with socially structured populations that retain genetic diversity but may be vulnerable to future fragmentation [21,26].

Our ability to evaluate differentiation within the full distribution of Marco Polo sheep is limited by sampling geography. All individuals analyzed here were collected within Kyrgyzstan's portion of the Tian Shan, and the dataset does not include animals from the Pamir Plateau, which represents the ecological and geographic core of *O. a. polii* [31,33]. Consequently, our findings characterize Argali genomic structure within Kyrgyzstan and should not be interpreted as capturing the full genetic breadth of Marco Polo sheep across Tajikistan, Afghanistan, and western China. Because individuals were not classified a priori by morphology and Pamir populations were not sampled, *O. a. polii* may be under-represented or absent from this dataset; therefore, taxonomic interpretation is necessarily limited to the sampled populations [8,31].

Our SNP-based analysis complements earlier genetic studies in Mongolia, notably Feng et al. [34], who used mitochondrial control region sequences and nuclear microsatellites to identify geographically structured lineages and conservation units. While our SNP data similarly reveal within-region genetic variation, direct comparison with these studies is constrained by differences in sampling scale and marker systems [34].

The availability of a high-quality reference genome for *Ovis ammon polii* [35] provides an important foundation for interpreting genomic studies. However, because the SNP array used here was designed for domestic sheep (*Ovis aries*), ascertainment bias is expected when applied to Argali and may influence allele frequency distributions and the representation of lineage-specific variants [22,23,35]. Accordingly, our results are best interpreted as a genome-wide SNP-based baseline for Kyrgyzstan populations rather than a comprehensive survey of adaptive or diagnostic variation [22,35].

Our findings add to the growing body of evidence that the long-debated subspecies distinction between *O. a. polii* (Marco Polo Argali) and *O. a. karelini* is weakly supported at the genomic level within the sampled Tian Shan populations [8]. Historical taxonomic treatments based on morphometrics recognized these forms as distinct [5–7], although even early authors noted that diagnostic differences were inconsistent. Taken together with previous genomic studies, our results support the interpretation that these forms may represent a single subspecific lineage in parts of the Tian Shan; however, robust evaluation of range-wide taxonomic boundaries will require dense sampling from the Pamir core and adjacent regions [8,25,29].

From a conservation and management perspective, the absence of clearly delineated genetic units within the sampled Kyrgyzstan population suggests that Argali in this region function largely as a single regional population. This supports management strategies that emphasize maintaining connectivity and demographic stability rather than subdividing populations based on weak genetic signals [19,36]. Nevertheless, individuals with elevated inbreeding coefficients highlight the potential risk of genetic erosion if movement among subareas is further restricted [26,37].

Genetic differentiation among proposed Argali subspecies appears weak relative to within-population variation, consistent with patterns observed in other Caprinae [3]. At the same time, introgression from domestic sheep has been documented across the range, posing additional management challenges [4]. Mountain ungulates worldwide face increasing isolation due to land-use change and border infrastructure [19,38], underscoring the importance of coordinated conservation strategies [36]. Because Kyrgyzstan lies within a broader transboundary landscape, maintaining permeability across key movement areas may help sustain gene flow and reduce the likelihood of localized inbreeding [31,36].

Recent spatial modeling by Zhuo et al. [31] demonstrated that border fencing can severely restrict potential movement corridors for Marco Polo sheep across the Pamir Plateau. Although our sampling does not include the Pamir core, these findings provide important context for interpreting how physical barriers could influence connectivity in adjacent mountainous regions, including those near Kyrgyzstan [31]. Together with our regional genomic baseline, this supports conservation strategies that prioritize landscape connectivity where feasible [31,36].

From a conservation perspective, these findings reinforce long-standing calls for international cooperation in Argali management [33]. Within the limits of Kyrgyzstan-based sampling, our results provide genomic evidence supporting regional management frameworks that emphasize connectivity, monitoring, and demographic resilience [33,36].

The primary limitation of this study is the absence of samples from the Pamir Plateau, the ecological and geographic core of Marco Polo sheep. Future work incorporating dense sampling across the Pamir and additional portions of the Tian Shan—ideally using whole-genome sequencing and spatially explicit analyses—will be essential for distinguishing kinship-driven patterns from broader demographic structure and for fully evaluating subspecies boundaries and connectivity across Central Asia [31,35].

5. Conclusions

This study provides the first high-resolution genomic assessment of Argali populations in Kyrgyzstan. By emphasizing dense within-population sampling rather than range-wide comparisons, our analysis was designed to evaluate whether genetic evidence supports subdivision into distinct management units or indicates that Argali in this region function as a single regional population.

Across all analyses, we found no genomic support for separating individuals historically attributed to *O. a. polii* and *O. a. karelini* within the sampled Argali population from Kyrgyzstan, recognizing that populations from the Pamir Plateau and other portions of the species' range were not included. Principal component analysis revealed only subtle patterns of genetic variation, which are best interpreted in the context of elevated kinship and localized demographic structure rather than discrete population subdivision. These findings suggest that, within Kyrgyzstan, Argali are more appropriately managed as a single regional population rather than as multiple genetically distinct units.

High average relatedness (mean IBD = 0.35) reflects the social structure typical of wild sheep, where dominant male or harem-based mating systems can elevate kinship among individuals. Despite this elevated relatedness, individual inbreeding coefficients were

generally low, indicating that widespread inbreeding is not currently a defining feature of the sampled population. However, the presence of individuals with moderate inbreeding coefficients highlights the potential vulnerability of localized groups if connectivity is further reduced.

From a conservation perspective, these results emphasize the importance of maintaining demographic and genetic connectivity within Kyrgyzstan's Argali populations. Management strategies that prioritize landscape permeability, movement corridors, and coordinated oversight are likely to be more effective than approaches based on subdividing populations according to weak or unsupported genetic signals. Given Kyrgyzstan's position within a broader transboundary landscape, such strategies may also contribute to sustaining gene flow in adjacent regions where Argali populations interface across political boundaries.

The conclusions of this study are necessarily constrained by sampling geography. All individuals analyzed were collected within Kyrgyzstan, and populations from the Pamir Plateau—the ecological and geographic core of Marco Polo sheep—were not represented. Accordingly, our findings should be interpreted as a regional genomic baseline rather than a comprehensive evaluation of Argali taxonomy or connectivity across Central Asia.

Future research incorporating dense sampling from the Pamir Plateau and additional portions of the Tian Shan, ideally combined with whole-genome sequencing and spatially explicit analyses, will be essential to fully resolve subspecies boundaries and to distinguish kinship-driven structure from broader demographic patterns. Together, such efforts will strengthen evidence-based conservation planning for Argali across their range.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/d18030194/s1>, Table S1: Calculated inbreeding coefficient, observed and expected homozygosity, observed and expected heterozygosity for argali sheep samples collected in Kyrgyzstan.

Author Contributions: Conceptualization, J.M.T., A.D. and M.R.F.; methodology, J.M.T. and M.R.F.; software, J.M.T.; sample collection and oversight, A.D. and M.R.F.; resources, M.R.F.; data curation, J.M.T.; writing—original draft preparation, J.M.T. and M.R.F.; writing—review and editing, J.M.T., A.D. and M.R.F.; visualization, J.M.T.; funding acquisition, M.R.F. All authors have read and agreed to the published version of the manuscript.

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Institutional Review Board Statement: The samples for this study were collected from harvested animals and found carcasses and did not require approval from our Institutional Animal Care and Use Committee. Samples were imported under USDS/APHIS/VS import permit number 127106 (Approved November 2015 and renewed December 2017) and handled in a BSL-2 laboratory.

Data Availability Statement: Metadata and genotype files can be found at: <https://doi.org/10.5061/dryad.76hdr7t7> (accessed on 15 March 2026).

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Conflicts of Interest: The authors declare no conflicts of interest.

References

1. Linnaeus, C. *Systema Naturae per Regna Tria Naturae*, 10th ed.; Laurentii Salvii: Stockholm, Sweden, 1758; p. 70. Available online: <https://archive.org/details/systemanaturae01linnuoft> (accessed on 16 March 2026).
2. Reading, R.; Michel, S.; Amgalanbaatar, S. The IUCN Red List of Threatened Species 2020: *Ovis ammon*. Available online: <https://www.iucnredlist.org/species/15733/22146397> (accessed on 16 March 2026).
3. Ropiquet, A.; Hassanin, A. Molecular phylogeny of caprines (Bovidae, Antilopinae): The question of their origin and diversification during the Miocene. *J. Zool. Syst. Evol. Res.* **2005**, *43*, 49–60. [CrossRef]
4. Li, X.; Han, B.; Liu, D.; Wang, S.; Wang, L.; Pei, Q.; Zhang, Z.; Zhao, J.; Huang, B.; Zhang, F.; et al. Whole-genome resequencing to investigate the genetic diversity and mechanisms of plateau adaptation in Tibetan sheep. *J. Anim. Sci. Biotechnol.* **2024**, *15*, 164. [CrossRef]
5. Geist, V. On the taxonomy of giant sheep (*Ovis ammon* Linnaeus, 1766). *Can. J. Zool.* **1991**, *69*, 706–723. [CrossRef]
6. Lydekker, R. *Horns and Hoofs; Or Chapters on Hoofed Animals*; Horace Cox: London, UK, 1893.
7. Lydekker, R. *Wild Oxen, Sheep and Goats of All Lands, Living and Extinct*; Rowland Ward: London, UK, 1898.
8. Dotsev, A.; Koshkina, O.; Kharzinova, V.; Deniskova, T.; Reyer, H.; Kunz, E.; Mészáros, G.; Shakhin, A.; Petrov, S.; Medvedev, D.; et al. Genome-Wide Insights into Intraspecific Taxonomy and Genetic Diversity of Argali (*Ovis ammon*). *Diversity* **2023**, *15*, 627. [CrossRef]
9. Jiufeng, J. Molecular Approaches for Conservation of Endangered Giant Argali Sheep (*Ovis ammon*) and Dwarf Blue Sheep (*Pseudois nayaur schaeferi*) in Asia. Ph.D. Thesis, State University of New York at Buffalo, Buffalo, NY, USA, 2000.
10. Blyth, E. On the species of the genus *Ovis*. *Ann. Mag. Nat. Hist.* **1841**, *6*, 195. Available online: <https://archive.org/details/annalsmagazineof6184unse> (accessed on 16 March 2026). [CrossRef]
11. Severtzov, N.A. Original description of *Ovis karelini*. *Turkestanskies Zhivotnye* (in Russian). 1873, p. 154. Available online: <https://treatment.plazi.org/GgServer/xhtml/03F50713993DFF87064AF6A5FE7DF485> (accessed on 16 March 2026).
12. Flesch, E.P.; Garrott, R.A.; Rotella, J.J.; Thomson, J.M.; White, P.J.; Cross, P. Evaluating wildlife translocations using genomics: A bighorn sheep case study. *Ecol. Evol.* **2020**, *10*, 13687–13704. [CrossRef]
13. Harper, C.K.; Vermaak, E.; Labuschagne, K.; Kotzé, A.; De Wet, J.I.; Guthrie, A.J.; Kotzé, A. Extraction of nuclear DNA from rhinoceros horn and characterization of DNA profiling systems for white (*Ceratotherium simum*) and black (*Diceros bicornis*) rhinoceros. *Forensic Sci. Int. Genet.* **2013**, *7*, 428–433. [CrossRef]
14. Shaw, G. The Canadian Sheep, *Ovis canadensis* (Pl. 610). *The Naturalist's Miscellany* **1804**, *15*, 610. Available online: <https://archive.org/download/biostor-263584/biostor-263584.pdf> (accessed on 16 March 2026). [CrossRef]
15. Nelson, E.W. *Ovis dalli*. In *United States National Museum. Proceedings*; Superintendent of Government Documents: Washington, DC, USA, 1884; Volume 7. Available online: https://archive.org/details/sim_united-states-national-museum-proceedings_1884_7 (accessed on 16 March 2026).
16. Kijas, J.W.; Townley, D.; Dalrymple, B.P.; Heaton, M.P.; Maddox, J.F.; McGrath, A.; Wilson, P.; Ingersoll, R.G.; McCulloch, R.; McWilliam, S.; et al. A genome-wide survey of SNP variation reveals the genetic structure of sheep breeds. *PLoS ONE* **2009**, *4*, e4668. [CrossRef] [PubMed]
17. Kijas, J.W.; Hadfield, T.; Naval-Sanchez, M.; Ciani, E. Linkage disequilibrium over short physical distances measured in sheep using a high-density SNP chip. *Anim. Genet.* **2014**, *45*, 754–757. [CrossRef] [PubMed]
18. Martchenko, D.; Shafer, A.B.A. Contrasting whole-genome and reduced representation sequencing for population demographic and adaptive inference: An alpine mammal case study. *Heredity* **2023**, *131*, 273–281. [CrossRef]
19. Wu, H.; He, W.; Zhao, K.; Wang, X.; Zhang, Q.; Zeng, Y.; Li, M.; Liu, Z.; Yang, J.; Ren, J.; et al. Population genomics of mountain ungulates reveals conservation priorities under climate and land-use change. *Mol. Ecol.* **2023**, *32*, 3650–3667. [CrossRef]
20. Hiendleder, S.; Kaupe, B.; Wassmuth, R.; Janke, A. Molecular analysis of wild and domestic sheep questions current nomenclature and provides evidence for domestication from two different subspecies. *Proc. R. Soc. B Biol. Sci.* **2002**, *269*, 893–904. [CrossRef]
21. Li, X.; He, S.-G.; Li, W.-R.; Luo, L.-Y.; Yan, Z.; Mo, D.-X.; Wan, X.; Lv, F.-H.; Yang, J.; Xu, Y.-X.; et al. Genomic analyses of wild argali, domestic sheep, and their hybrids provide insights into chromosome evolution, phenotypic variation, and germplasm innovation. *Genome Res.* **2022**, *32*, 1669–1688. [CrossRef]
22. Flesch, E.P.; Rotella, J.J.; Thomson, J.M.; Graves, T.A.; Garrott, R.A. Evaluating sample size to estimate genetic management metrics in the genomics era. *Mol. Ecol. Resour.* **2018**, *18*, 1072–1084. [CrossRef]
23. Kohn, M.H.; Murphy, W.J.; Ostrander, E.A.; Wayne, R.K. Genomics and conservation genetics. *Trends Ecol. Evol.* **2006**, *21*, 629–637. [CrossRef] [PubMed]
24. Miller, J.M.; Poissant, J.; Kijas, J.W.; Coltman, D.W.; International Sheep Genomics Consortium. Consistent divergence times and allele sharing measured from cross-species application of SNP chips developed for three domestic species. *Mol. Ecol. Resour.* **2012**, *12*, 1150–1160. [CrossRef]
25. Archibald, A.L.; Cockett, N.E.; Dalrymple, B.P.; Faraut, T.; Kijas, J.W.; Maddox, J.F.; McEwan, J.C.; Hutton Oddy, V.; Raadsma, H.W.; Wade, C.; et al. The sheep genome reference sequence: A work in progress. *Anim. Genet.* **2010**, *41*, 449–453. [CrossRef]

26. Huisman, J.; Kruuk, L.E.B.; Ellis, P.A.; Clutton-Brock, T.; Pemberton, J.M. Inbreeding depression across the lifespan in a wild mammal population. *Proc. Natl. Acad. Sci. USA* **2016**, *113*, 3585–3590. [CrossRef] [PubMed]
27. Thomson, J.M.; Davletbakov, A.; Frisina, M.R. Data associated with: Fine-Scale Population Structure and Relatedness of Argali (*Ovis ammon*) in Kyrgyzstan Revealed by High-Density SNP Data. Dryad, Dataset. 2026. Available online: <https://doi.org/10.5061/dryad.76hdr7t7> (accessed on 16 March 2026).
28. Flesch, E.P.; Rotella, J.J.; Thomson, J.M.; Newby, J.; Cross, P.; Garrott, R.A. Average kinship within bighorn sheep populations is associated with connectivity, augmentation, and bottlenecks. *Ecosphere* **2022**, *13*, e3964. [CrossRef]
29. Price, A.L.; Patterson, N.J.; Plenge, R.M.; Weinblatt, M.E.; Shadick, N.A.; Reich, D. Principal components analysis corrects for stratification in genome-wide association studies. *Nat. Genet.* **2006**, *38*, 904–909. [CrossRef]
30. Knief, U.; Hemmrich-Stanisak, G.; Wittig, M.; Franke, A.; Kerick, M.; Timmermann, B.; Bens, M.; Boltze, J.; Platzer, M.; Kempnaers, B.; et al. Quantifying realized inbreeding in wild and captive animal populations. *Heredity* **2015**, *114*, 397–403. [CrossRef]
31. Zhuo, Y.; Weckworth, B.V.; Sharma, R.; Zhang, H.; Yao, J.; Qiao, M.; Xu, W. Border fences reduce potential for transboundary migration of Marco Polo sheep (*Ovis ammon polii*) in the Pamir Plateau. *Sci. Total Environ.* **2024**, *912*, 169298. [CrossRef]
32. Städele, V.; Vigilant, L. Strategies for determining kinship in wild populations using genetic data. *Ecol. Evol.* **2016**, *6*, 6107–6120. [CrossRef] [PubMed]
33. Schaller, G.B.; Kang, A. Status of Marco Polo sheep (*Ovis ammon polii*) in China and adjacent countries: Conservation of a vulnerable subspecies. *Oryx* **2008**, *42*, 100–106. [CrossRef]
34. Feng, J.; Frisina, M.R.; Webster, M.S.; Ulziimaa, G. Genetic differentiation of Argali sheep (*Ovis ammon*) in Mongolia revealed by mitochondrial control region and nuclear microsatellites analyses. *J. Bombay Nat. Hist. Soc.* **2009**, *106*, 38–44.
35. Yang, Y.; Ma, Y.; Liu, B.; Xie, X.; Yu, H.; Wang, G.; Yang, J.; Chen, S.; Wang, X.; Zhang, X.; et al. Draft genome of the Marco Polo sheep (*Ovis ammon polii*). *GigaScience* **2017**, *6*, 1–7. [CrossRef]
36. Shackleton, D.M. (Ed.) *Wild Sheep and Goats and Their Relatives: Status Survey and Conservation Action Plan for Caprinae*; IUCN: Gland, Switzerland; Cambridge, UK, 1997. Available online: <https://portals.iucn.org/library/efiles/documents/1997-006.pdf> (accessed on 16 March 2026).
37. de Cara, M.A.R.; Villanueva, B.; Toro, M.A.; Fernández, J. Using genomic tools to maintain diversity and fitness in conservation programmes. *Mol. Ecol.* **2013**, *22*, 6091–6099. [CrossRef] [PubMed]
38. Kauffman, M.J.; Cagnacci, F.; Chamaillé-Jammes, S.; Hebblewhite, M.; Hopcraft, J.G.C.; Merkle, J.A.; Mueller, T.; Mysterud, A.; Peters, W.; Roettger, C.; et al. Mapping out a future for ungulate migrations. *Science* **2021**, *372*, 566–569. [CrossRef]

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