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Title: Phylogenetic Tree analysis

1. Investigate the evolutionary relationships among bats, whales, and artiodactyls (even-toed ungulates) groups.
- a. Choose 3 to 5 species in each group. Mention each species, i. Name, ii. GenBank Accession Number, iii. Sequence Length & iv. Sequence.

1. Bat Species:

- ***Myotis lucifugus*** also known as **little brown bat**.

Accession: XR_002827941.1

150 bp linear ncRNA, snoRNA

>XR_002827941.1 PREDICTED: Myotis lucifugus small nucleolar RNA SNORA16B/SNORA16A family (LOC111824880),ncRNA

TGTGGCCCTTATCCAGGCTGCAGCTGGGGGGGATACAGGGTGAGGGGAAAGAAATTGTTTTTAAAAAGGA
GCCCTAGTGACAGTTTTCTTGATGGTCACCATCCGATTCTGTAAACAGATCTGCCACATTTTGGGAAAAGA
CAGTT

- ***Craseonycteris thonglongyai*** also known as **bumblebee bat**.

Accession: MK605583.1

399 bp linear DNA

>MK605583.1 Craseonycteris thonglongyai microsatellite Ct64 sequence

CTGGGATGGCTCAGTGGGTTGAGTGCTCACTTTGCAGCCCTGGGGACCAAGGATCGAATCCCAGCCCTCG
CCTGTGCAGACCCATATGCTGAACCTGGGGAGTGCCCCCAGTTTGGTTCTCACAGGGGTGAGACCAGGCCT
GGCCTAATTCCTGCCCTGTGCAGCCCTGCTCCACCTATGGAACAAGACTATGTGGAAAAATCAACAATGTGC
AACTATGAAGACTGAAGGCTGTTATGGGCTCTTCCCCTCTCTGTCTCTGTCTCTCTGTCTCTGTCTCTGT
CTCTCTCTCTCTCTCTCTCTCTCTTTGTCAAAAACAATAAAAACAAATCTTAAAAAAAAAAGAGGTTACT
GCAAAGATGAAATGAGGCAACGTATACAATGCATTT

- ***Desmodus rotundus*** also known as **common vampire bat**.

Accession: XR_002974494.2

188 bp linear ncRNA, snRNA

>XR_002974494.2 PREDICTED: Desmodus rotundus U2 spliceosomal RNA (LOC112301584), ncRNA
ATCGCTTCTCTGCCTTTTGGCTAAGATCAAATGTAGTATGTGTTCTTTGTTAATATCTGATACGTCCTCTATTC
GAGGACATTATATTAAGTGGATTTTGGAGCTGGGAGATGGAATAGGAGCTTGCTCCATCCACTG
CACACATCTACCCAGTATTGCAGTACTTCTGGGGATGGTGCCCCCTAC

- ***Artibeus jamaicensis*** also known as **Jamaican, common, or Mexican fruit bat**.

Accession: NW_026521193.1

507 bp linear DNA

>NW_026521193.1 *Artibeus jamaicensis* isolate MK-2021 breed Jamaican fruit bat unplaced genomic scaffold, CSHL_Jam_final contig_1001, whole genome shotgun sequence

```
AAGGAAAATTCAGGGGATTGAGGGAGGCAAACCTCACCACAGCTGACTTTGAGAAGGAGGGGAGTCCAAGG
CGAGAATGGGGGAGCCAGCAAGGAAACGGGGCCGAATTGTACCATCATAAGGAACTGAATTCGACAACC
AACCTGAGGGAGTCGGGAAGCAGATTCTCCCCAGGCCCGGGCAAGAACGTGATGCAGCTGACACCCTG
ATTCTCTGTCTTGTGAGACCTTGAACAGAGAGGGTCATGCTGTGGCAGGTCTCTTCACTTCCAGGATGG
GAGCGACGGGACTGGACGTCGGGGAGTCAGGTACGGAAGCTGGGACTCTCAAGCCCCTGTGACGCAGCT
GAGGAGCCAGCTACAGCTCGGAAGCTCTCAAATCCTGTGCCGCTGCCATAGCGACGGGACTGGACGCCGG
GGAGTCAGGTACGGAAGCCAGGACTCTCAAACCCCTGTGACGCAGCTGAGGAGCCAGCTACGGGAGCCG
GGAAGCTCTCAAATCCT
```

2. Whales:

- ***Balaenoptera physalus*** also known as **fin whale**.

Accession: AY822142.1

365 bp linear DNA

>AY822142.1 *Balaenoptera physalus* haplotype finF Y locus 2 genomic sequence

```
CTTTGGGTGGGAGGGGTGGGGCCGTACTGGGAAGGAGTTGAAAAGAGGGAGGAAAGAGAGAGGGGGGCTA
TCAAGTTTTTTTTCTAGCCAGATATGATTGAAAACTGAAAAGAGGCCCATCACCTCTAATTTTGTAG
ATTGTATTGTGACAGCTGTGCCACACCTTGCCATCTGATTCCCTCCTACAGAGCCCCAGGACCTATTTT
CCCCACTAAGTTGTTTGAATTCTACTGCCAACTTGAACCTCTGCCTTAGCATATTCTTTGAAAGTCAGTT
CCAGGGCTGCTGAGAACCCTTCCCCCACGCTACTGGCTTTATAATGAAGAGTGAGAGTGTGCCTCTGG
ACCTTCTCTCAAAA
```

- ***Orcinus orca*** also known as **killer whale**.

Accession: XR_007474996.1

145 bp linear ncRNA, scaRNA

>XR_007474996.1 PREDICTED: *Orcinus orca* small Cajal body-specific RNA 20 (LOC125963167), ncRNA

```
GGCCTCTCTCCCATGGGGCTGTGTGTGTCGGGGGGAGGGGACCTCTTCTCCTCAGCCCTTGGGCTGGAAG
CGCAGATCAGCCCTGGGGGATCCCTGTGCGGGCTGCCATGAGCTGGGCACCCCTGTTTCTACCTCAGA
CGTTT
```

- ***Eschrichtius robustus*** also known as **gray whale**.

Accession: AB195488.1

529 bp linear DNA

>AB195488.1 *Eschrichtius robustus* DNA, SINE flanking sequence IWA31 locus

```
GCTGTGGGCTACCTACAACACAGAGGGCAAACTAAGCAATGTAGTTTCAAGAGTAATCAAATTCACAAG
CTGCGGGCTTCCCTGGTGGCGCAGTGTTGAGAATCTGCCTGCCAATGCAGGGGACACGGGTTCGAGCCC
TGGTCTGGGAAGATCCACATGCCGCGGAGCGACTAGGCCCGTGAGCCACAATTGCTGAGCCTGCGCGTC
TGGAGCCTGTGCCCCGCAATAAGAGAGGGCCGCGACAGTGAGAGGCCCGCGCACCGCGATGAAGAGTGGCC
CCCACTTGCCGCAACTAGAGGAAGCCCTCGCACAGAAAAGAAGACCAACACAGCCAAAAATAAATAAAT
AAAATTTTAAAAATAAATAACTAAAAATAAATAAATTAATTAATAAAAAAAAAATTACAAAGCTGCTCCACT
AGAAGGTCAGAGTTCATAAACAGGAATATGCCAGTGTTATTATCTCAGCGGTATCAACAGGAAAGCAAA
CATAGAGGGAAAATATCTTATGTGCCACTATAGGGGC
```

- ***Eubalaena japonica*** also known as **North Pacific right whale**.

Accession: AB195498.1

282 bp linear DNA

>AB195498.1 Eubalaena japonica DNA, SINE flanking sequence Sei23 locus

```
GGATGGTCCCAGAGTCACATCTGAGGCACAGTTAACCAAAAGGTCATTGAATGTAGGACTTGAATCCT
TAAAGATGGTACAGGAAGAACAATATTAGATTTGGTGCTTGTGATCTCATTCTAGTACATCATGATGA
TGATGATGATGATGATGATGACGGTGATGATGATGAAGGGAGATGGTGTGTTTGTGTTGATTAAGAACAAG
AGCTTCAAATAATAGACCGATGTTTGAGTGCAGCGTTGCCTTTTATTGACTGAATGACCTTGGACAAG
TT
```

3. Artiodactyls:

- ***Sus domesticus*** also known as **Pig**.

Accession: U07180.1

1,301 bp linear mRNA

>U07180.1 *Sus domesticus* lactate dehydrogenase-B (LDH-B) mRNA, complete cds

```
GCACGAGCTGCAGAGCAGACGCACGCTGCCGACTTGTCTCCTTCGGGCGACCCTTACAAGCCGTCTCA
AGAACTTTCACTCCTCTGCGCACAATGGCAACTCTTAAGGAAAACTGATTGCACCAGTTGCAGAAGAAG
AGACAACAATCCCAAACAATAAGATCACTGTAGTGGGTGTTGGACAAGTCGGTATGGCATGTGCCATCAG
CATTCTGGGAAAGTCTCTGACGGATGAGCTTGCTCTGGTGGATGTTTGAAGATAAACTCAAGGGAGAA
ATGATGGATCTGCAGCACGGAAGCTTGTTCTTCAGACACCCAAAATTGTGGCAGATAAAGATTACTCTG
TGACCGCCAATTCCAAGATTGTGGTGGTGACGGCAGGCGTTCGCCAGCAGGAGGGAGAGAGTCGTCTCAA
TCTGGTGCAGAGGAACGTCAATGTCTTCAAGTTCATCATCCCTCAGATTGTCAAGTACAGTCCTGACTGC
ATCATAATTGTGGTTTGAACCCAGTGGATATTCTCACATATGTTACCTGGAAGCTAAGTGGATTACCCA
AGCACCGTGTGATTGGAAGTGGATGTAACCTGGATTCTGCAAGGTTTCGCTATCTTATGGCTGAAAAGCT
TGGTGTTCATCCCAGCAGCTGCCACGGGTGGATTTTGGGAGAACATGGCGACTCAAGCGTGGCTGTGTGG
AGTGGGGTGAATGTGGCAGGTGTGTCTCTCCAGAACTGAATCCAGAGATGGGAACGGACAATGATAGTG
AAAATTGGAAAGAAGTGCATAAGATGGTGGTTGAAAGTGCCTACGAAGTCATCAAGCTGAAAGGCTACAC
CAACTGGGCAATTGGCTTAAGCGTGGCTGATCTTATTGAATCCATGTTGAAAAACCTATCCAGGATTAC
CCAGTGTCAACAATGGTGCAGGGCATGTATGGCATTGAGAATGAGGTCTTCTGAGCCTTCCATGTGTCC
TGAATGCTCGAGGACTAACCAGTGTTATCAACCAGAAGCTGAAGGATGATGAGGTTGCCAGCTCAAGAA
CAGTGCTGACACTCTGTGGGGCATCCAGAAGGATCTGAAGGACCTGTGACTCCCGGCTTCTAGGTTGTAG
TAACCTACAACTACAGTGTGATTTCCACAAGCCTTTAGTTTTATCCATGTATGTGGAGCACAGTTTG
CTTTTATCTTCTTAAATATGTGAATGTGGGCTCCAGAGTCAAAGCCCATGCTTAGTTTAATGCTTTCA
AGCAAGTCCTTGAACAAATAAAGTTAACTATCATAGTGTGA
```

- ***Lama pacos*** also known as **Alpaca**.

Accession: EU293064.1

122 bp linear mRNA

>EU293064.1 *Lama pacos* tyrosinase mRNA, partial cds

```
TCACTTACTGGGATAGCGGATGCCTCTCAAAGCAGCATGCACAATGCCTTGCACATCTTCATGAATGGAA
CGATGTCCCAGGTGCAGGGATCTGCCAACGATCCTATCTTCTTCTTCACCA
```

- ***Bos taurus*** also known as **Cattle**.

Accession: NM_001080321.1

710 bp linear mRNA

>NM_001080321.1 *Bos taurus* shadow of prion protein (SPRN), mRNA

```
GGCACATCCCCGCTCCGCTCGGCGGCGCCGCGCCAGGTCTCCGCCACAGCCCCCTCCGCTCGGGCCGCG
```

GTCCTCGGAGCCGCCGCGTTCTGCCCCAGGCCCGCGGCTGGTTCTGCTCCAGGAACATGAATTGGGCGG
CCGCCGTGTGCTGGGCTCTGCTGCTAGCGGCCACCTTCCTCTGCGACGGCAGTGCAGCCAAGGGTGGCCG
CGGTGGGGCCCGTGGCAGCGCCGTGGCGGGCGCGGCCGCGAGGGTGCCTGTGAGGCCAGCGCCCCG
TACGCCGGCTCCTCAATGCGCGTGGCCGCGGGGGCGGCGGGGGCCGCGGGGGGCGAGCCGAGGCC
TGGCTGCGGGCTCGAGCTGGAGGAGGGCCGCGGGGGCGGCGGAGCTTGGCCCCGAGGATGCAGAAGACG
GAGCGCCCGGCAGCAACGGAACGGGGAGAGGCGTCTACAGCTACTGGGCGTGGACCTCAGGCACGGGGCC
CACAGGCCACAGGCACCTCTGCCACTGCTGGGCGGGGCCCTGGGCGCCCTGCGGCTGCTGCGGCCCTAGG
CTGGGCCGGGCGCATCAGGGCCAGAGCCGCCCCAGCCTGTGCCCTCCTCCAGGCCCGCGGCCCCGAT
CGCCCCCTCCCCTCCCCTGCCACAGCCAGGAGCCTAGAGCAGTGTGGGCAGCTCTGGCCCCCTCGGCCTGA
AGCTGTGACA

- ***Camelus bactrianus*** also known as **Bactrian camel**.

Accession: NW_011509604.1

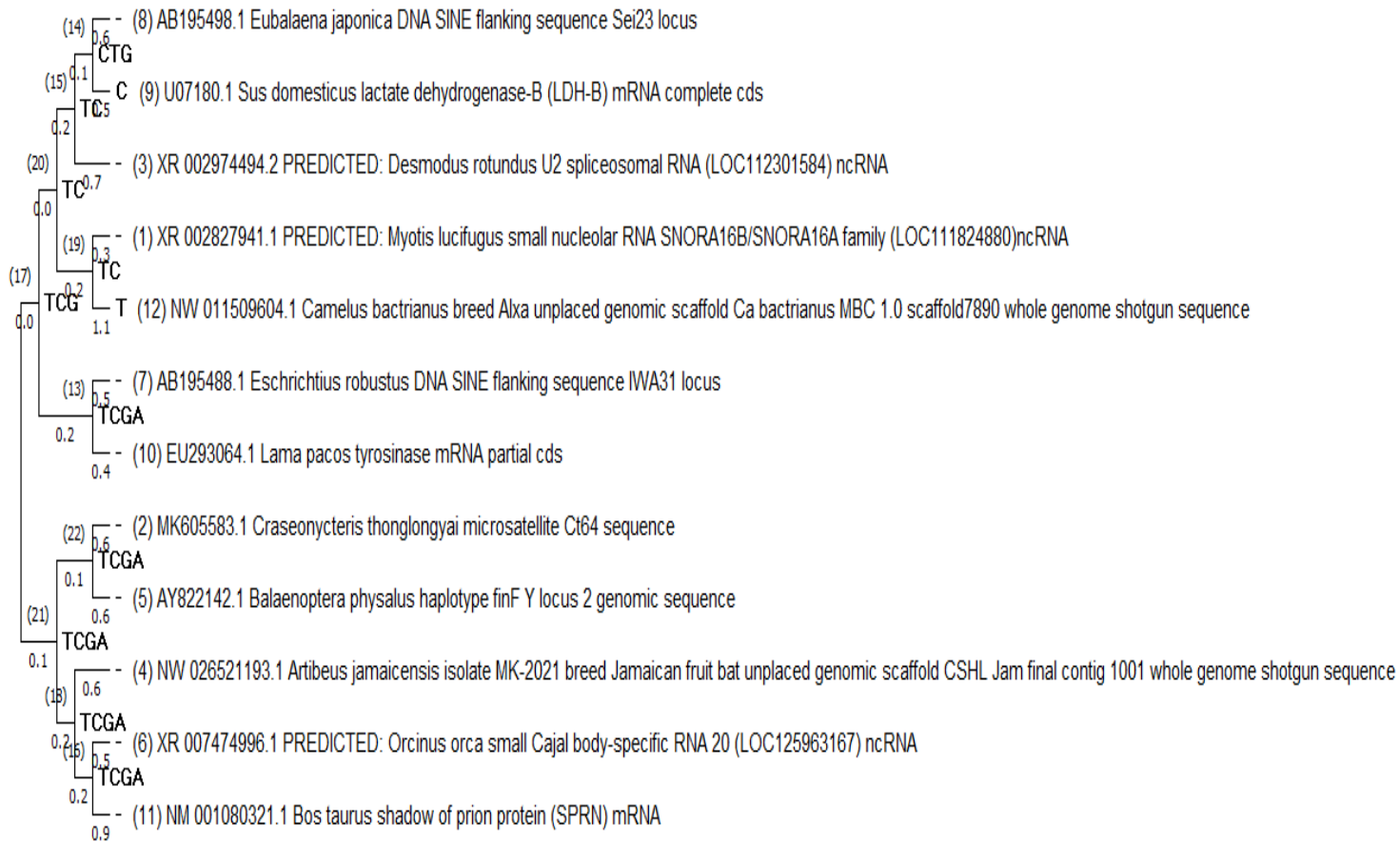
967 bp linear DNA

>NW_011509604.1 Camelus bactrianus breed Alxa unplaced genomic scaffold,
Ca_bactrianus_MBC_1.0 scaffold7890, whole genome shotgun sequence

TCCACCCTCAAACTTCCTTTTAGTGTTTTTTTTTAATTAATCATTGTTACCATATTTTCCTTGGTTGT
CATGCTTTCTCTGGTTTTCTTGAATCCTTGGAACGTATTTATCACAGCTGCTGTGAAGGTGGTGTCCGCC
AGACCCAGCACCCGGCCCCCTCAGAGTCAGTTTCTGTTCACTGACTGTCTTCTGAGGACGGCTCACACTT
GCCAGTGTTTTGTGTAGCCATAATTTTTATTGAAAACCTGGACATTTAAGATTGTGTTGTGTCATGTG
GACTCTGACCTTTATTTTTTTTTTAATTTGTAATGTTGCTACAATTCCTTGACTGTTTTGTAACATGCC
TGGAATGACTCTTTGAAATCTCCCTACCAGTGTACATAGCTACTGATGTCACAGCTTCACTTTGCTATT
TTCAATTTCTGTTTTCAAGCCTGATTTTTAGGATTTGCTCCGTATCTGCTTGGTGTAGCACCCAAGCCTGC
TGAATTTTCCCTTCTGTAATGGATCTCTGCGGGGATAGGGCAGGGAGGGCGTGGAAATCAGTGTCTGG
CAGTTTTCTGTGTGTGTACCCCACTCTCCCACTTTCTGAGTTCATGTGCCCTGGGTACCCAGGCTG
CCATGATGTCCTCACTTATCTTCTCTGCTCTCTGACTTTCAGGATCTTCTGTTAGATCTCTGGCTAG
TTCTTAGGTCTGGACCCTCCTAGTCCCACCACTCGGGCCAGCAGAGAGGCAGGCTCTCCCGTGTGTTCTC
CGTCGCCCTCAGCGCCCTCTGACTCCCAGGATCTCTGCCAGGCACTGCCCTGCTCTGCACCAGCGAGCA
CCGAGGAGGCAGGGCCCCCTCCTGGTCGGGCTCTGCTCCAGCTCGTGTAGACCAGCCAAGGCGGAAACA
CCCCAGGCTTCCCCATCCCTTCCCGAATGCAGCAGTTTTTTCTCAGTTTGTGTTT

b. Construct a phylogenetic tree using sequence data for the chosen species.

Phylogenetic tree:



c. Analyze the tree to identify key evolutionary events and relationships in about 350 words.

Analyzing a phylogenetic tree involves several key steps and considerations to understand the evolutionary relationships it represents.

- **Nodes** represent common ancestors, branches represent evolutionary relationships, and the length of branches may indicate time or genetic distance. Common ancestor of all the organisms included in the tree.
- The point where **branches** split represents a common ancestor, and the length of the branches may suggest the amount of evolutionary change.
- **Clades** are formed by branches that are connected by a single node. A monophyletic group includes all the descendants of a common ancestor.
- **Outgroups** help root the tree and provide a reference point for understanding the evolutionary relationships within the main clade

Branching Pattern:

The branching pattern in the tree is bifurcating, meaning each internal node splits into two branches. This indicates a dichotomous evolutionary history, where a single ancestral species diverges into two descendant species.

Rooting:

The tree appears to be rooted, meaning it has a designated ancestral node at the bottom. A rooted tree allows for inferences about the direction of evolutionary change.

Branch Length:

The branch lengths in the tree seem to be proportional to the amount of evolutionary change that has occurred along that branch. Longer branches suggest greater evolutionary divergence between the species at the tips of the branches.

Bootstrap Values:

These values represent the likelihood that a particular grouping of species in the tree is due to common ancestry rather than chance. Here's what the presence or absence of bootstrap values can tell us:

- High bootstrap values (usually above 70%) indicate strong support for the branching order represented by that branch. This suggests that the grouping is likely due to shared ancestry, rather than random chance.
- Low bootstrap values (less than 50%) indicate weak support for the branching order. This means there is uncertainty about the evolutionary relationships between the species grouped by that branch.

In the present case: only *Eschrichtius robustus* and *Lama pacos* shows the Bootstrap value more than 50% and all the other species shows Bootstrap value below 50%.

Evolutionary Models:

The specific evolutionary model used to construct the tree is not mentioned in the image. Evolutionary models incorporate assumptions about how DNA sequences change over time.

1. Bats (*Myotis lucifugus*, *Craseonycteris thonglongyai*, *Desmodus rotundus*, *Artibeus jamaicensis*)

- Models : HKY85, GTR
- Reasoning: Bat genomes might show different nucleotide frequencies and substitution rates, making the HKY85 or GTR models appropriate.

2. Whales (*Balaenoptera physalus*, *Orcinus orca*, *Eschrichtius robustus*, *Eubalaena japonica*)

- Models : GTR, TN93
- Reasoning: Whale mitochondrial genomes can exhibit complex substitution patterns, and these models can handle varying rates and frequencies.

3. Artiodactyls (*Sus domesticus*, *Lama pacos*, *Bos taurus*)

- Models : K2P, HKY85
- Reasoning: Artiodactyls often exhibit distinct transition/transversion biases, making K2P or HKY85 models suitable.

Comparative Analysis:

The tree facilitates comparative analysis by allowing researchers to compare the evolutionary histories of different species. By looking at the branching pattern and branch lengths, researchers can infer which species are more closely related and how long ago they diverged from a common ancestor.

Biogeography:

Bats

- *Myotis lucifugus* (Little Brown Bat): Widespread in North America, from Alaska to central Mexico; inhabits forests and human structures.
- *Craseonycteris thonglongyai* (Bumblebee Bat): Endemic to Thailand and Myanmar; found in limestone caves near rivers.
- *Desmodus rotundus* (Common Vampire Bat): Central and South America; roosts in caves and hollow trees, blood-feeding.

- *Artibeus jamaicensis* (Jamaican Fruit Bat): Caribbean, Central and northern South America; tropical forests and plantations.

Whales

- *Balaenoptera physalus* (Fin Whale): Cosmopolitan; deep offshore waters and migratory between polar and tropical regions.
- *Orcinus orca* (Killer Whale): Global distribution; versatile habitats from coastal waters to open oceans.
- *Eschrichtius robustus* (Gray Whale): North Pacific; coastal waters, migrates between Baja California and the Arctic.
- *Eubalaena japonica* (North Pacific Right Whale): Bering Sea and Gulf of Alaska; coastal and shelf waters, critically endangered.

Artiodactyls

- *Sus domesticus* (Domestic Pig): Global distribution due to domestication; adaptable to various environments.
- *Lama pacos* (Alpaca): Andes of South America; high-altitude grasslands and plateaus.
- *Bos taurus* (Domestic Cattle): Worldwide distribution due to domestication; inhabits diverse environments.

The deepest divergence in the tree separates the bats from the whales and artiodactyls. This suggests that the lineage leading to bats diverged from the lineage leading to whales and artiodactyls earlier than any other divergence event in the tree.

Within the artiodactyl clade, the alpaca and cow are more closely related to each other than they are to the pig or camel. This suggests that the lineage leading to the alpaca and cow diverged from the lineage leading to the pig and camel after the lineage leading to artiodactyls diverged from the lineages leading to bats and whales.

These species have diverse evolutionary histories but may share common ancestors at various points in evolutionary time. For example, bats and whales both belong to the larger group of mammals, but they have distinct evolutionary paths and adaptations based on their respective habitats and lifestyles. Similarly, domesticated animals like pigs, alpacas, cattle, and Bactrian camels have been selectively bred by humans over millennia for specific purposes, leading to their domestication and unique traits.

- d. Compare and contrast the evolutionary histories of the groups in about 250 words.**

The evolutionary histories of the groups in the image can be compared and contrasted based on their relative positions on the phylogenetic tree. Here's a breakdown:

Similarities:

- All the species likely originated from a single ancestral organism that lived millions of years ago.
- The branching pattern suggests that all groups underwent periods of divergence from a common ancestor, accumulating genetic mutations over time.

Differences:

- The deepest divergence separates the bats from the whales and artiodactyls. This suggests the lineage leading to bats diverged earlier than the lineages leading to whales and artiodactyls.
- Within the artiodactyl clade, the alpaca and cow are more closely related to each other than they are to the pig or camel. This indicates an independent divergence within artiodactyls after they diverged from the whale and bat lineages.
- The whales diverged from each other more recently than any of the artiodactyl species diverged from each other.

Overall, the tree depicts a complex evolutionary history with multiple divergence events. The bats represent the earliest divergence, followed by the artiodactyls, and then the whales. There were also subsequent diversification events within each of the major clades.

e. Discuss the implications of your findings for our understanding of mammalian evolution in about 150 words.

The findings from the phylogenetic tree are limited to the specific species analyzed, but they do offer some insights into mammalian evolution:

Early bat divergence: The tree suggests bats diverged from the lineage leading to whales and artiodactyls earlier than any other branching event. This supports the idea that bats possess unique characteristics that evolved separately from other mammals.

Artiodactyl diversification: The tree shows the alpaca and cow are more closely related than to the pig or camel. This indicates an independent diversification event within artiodactyls after they diverged from the whale and bat lineages.

The Phylogenetic Tree diverse evolutionary paths taken by mammals in response to different ecological niches and pressures. Bats demonstrate convergent evolution in traits like echolocation, highlighting adaptive radiation in nocturnal environments. Whales exemplify the

profound morphological and physiological changes necessary for a transition from terrestrial to aquatic life, revealing the extent of evolutionary plasticity in response to marine habitats. Artiodactyls, especially domesticated species like *Sus domesticus*, *Bos taurus*, *Lama pacos*, and *Camelus bactrianus*, illustrate the significant impact of human intervention on evolutionary trajectories. These findings underscore the importance of environmental pressures, ecological niches, and human activities in shaping mammalian evolution. The diverse evolutionary strategies observed across these groups enhance our understanding of how mammals adapt to varied and changing environments, contributing to the broader narrative of mammalian biodiversity and evolutionary resilience.