Supplemental Information

Genomic Adaptations and Evolutionary History of the Extinct Scimitar-Toothed Cat, *Homotherium latidens*

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Figure S1: Maximum likelihood phylogenetic tree constructed using a concatenated supermatrix of 29,216,712 bp. Related to Figure 1 and STAR methods phylogenetic analyses. Numbers at the nodes show bootstrap support values. Branch lengths show the average number of substitutions per site.
Figure S2. Phylogenetic analyses for tree topology discordances in the Felidae. Related to Figure 1 and STAR methods phylogenetic analyses. A) Species tree under the multispecies coalescent inferred from the maximum-likelihood gene trees. Branches have been numerically labelled for easy identification. B) Relative frequencies of the three possible bipartitions (possible arrangements of a quartet on an unrooted tree) induced by each internal branch of the estimated species tree. X axis numbers correspond to branch labels in A written in a quartet fashion based on the topology ([A,B],[C,D]). Dashed lines show the threshold value of ⅓, shown theoretically to be the minimum frequency for a true bipartition. Related to Figure 1 and STAR methods phylogenetic analyses.
Figure S3: Exome-wide heterozygosity estimates based single representatives for each species used in the current study. Related to Figure 3 and STAR methods genetic diversity. Variance was estimated by independently calculating the average heterozygosity in non-overlapping windows of 200kb of covered bases.
| Node               | Correlated-rates model | Independent-rates model | Low rate prior: Gamma(1,50) | High rate prior: Gamma(1,2) | Prior (no data) |
|-------------------|------------------------|-------------------------|-----------------------------|-----------------------------|----------------|
| Root              | 32.8                   | 32.3                    | 33.0                        | 32.8                        | 32.4           |
| Homotherium-sister| 22.5                   | 16.7                    | 22.6                        | 22.6                        | 30.3           |
| Pantherinae-Felinae| 14.1                   | 10.2                    | 14.1                        | 14.1                        | 28.3           |
| Neofelis-Panthera | 8.3                    | 5.5                     | 8.3                         | 8.3                         | 23.3           |
| Panther           | 5.4                    | 3.4                     | 5.4                         | 5.4                         | 17.9           |
| Caracal-sister    | 11.3                   | 8.3                     | 11.3                        | 11.3                        | 24.9           |
| Acinonyx-sister   | 9.7                    | 7.1                     | 9.6                         | 9.7                         | 21.6           |
| Lynx-sister       | 9.2                    | 6.7                     | 9.2                         | 9.2                         | 17.2           |
| Prionailurus-Felis| 8.2                    | 6.0                     | 8.1                         | 8.2                         | 12.7           |
| Felis             | 5.0                    | 3.8                     | 5.0                         | 5.0                         | 9.2            |

Table S1 - Ages of key nodes in the phylogeny, estimated under a range of settings.

Related to Figure 1 and STAR methods phylogenomic dating. The time-tree in Figure 1 is based on the correlated-rates model. *Correlated-rates model: Rates are assumed to be correlated between neighbouring branches in the phylogeny. Independent-rates model: Rates are assumed to be identically and independently distributed across branches in the phylogeny. Low rate prior: Gamma(1,50) prior for the rate, with a prior mean of 0.0002 substitutions/site/Myr. High rate prior: Gamma(1,2) prior for the rate, with a prior mean of 0.005 substitutions/site/Myr. Prior (no data): Analysis run without sequence data, such that the node ages are sampled from the joint prior.*
| GeneName       | OneRatio | FreeRatio | Biological role                                                                 | Functional role                  |
|---------------|----------|-----------|---------------------------------------------------------------------------------|----------------------------------|
| AGBL5         | 0.97893  | 3.7966    | vision sensitivity to diurnal light and circadian rhythms                       | vision                           |
| AK3           | 1        | 2.5265    | endurance                                                                      | mitochondria respiration         |
| B3GALNT2      | 1        | 4.501     | vision sensitivity to diurnal light and circadian rhythms                      | vision                           |
| C13orf30      | 1        | 2.3383    | unknown                                                                        | unknown                          |
| CAPNS2        | 1        | 2.8782    | vision sensitivity to diurnal light and circadian rhythms                      | vision                           |
| ECSCR         | 1        | 3.4687    | endurance                                                                      | angiogenesis                     |
| F5            | 0.0001   | 3.3227    | endurance                                                                      | circulatory system               |
| GCM1          | 1        | 2.0195    | reproduction                                                                   | placenta                         |
| GOLT1A        | 1        | 4.4857    | cell function                                                                  | Golgi                            |
| GPRC5A        | 1        | 2.7265    | immunity                                                                       | cancer                           |
| HMGB2         | 1        | 2.127     | immunity                                                                       | fecundity                        |
| IQCF5         | 0.64872  | 2.3567    | unknown                                                                        | unknown                          |
| ISCU          | 1        | 2.3704    | endurance                                                                      | mitochondria respiration         |
| LAGE3         | 1        | 2.449     | cell function                                                                  | apoptosis                         |
| MIS18A        | 1        | 3.7168    | cell function                                                                  | mitosis                          |
| MMP12         | 1        | 2.2541    | endurance                                                                      | respiratory/circulatory system   |
| NTF3          | 1        | 4.3076    | socialisation                                                                  | nervous system                   |
| OR11A1        | 0.83912  | 3.7452    | vision sensitivity to diurnal light and circadian rhythms                      | olfaction                        |
| Per1          | 0.02951  | 2.0949    | vision sensitivity to diurnal light and circadian rhythms                      | circadian clock                  |
| PGD           | 1        | 2.8531    | bone mineralization                                                            | bone mineralization              |
| Rplp1         | 0.0001   | 3.1993    | ribosome                                                                       | protein synthesis                |
| Rps13         | 1        | 2.4235    | ribosome                                                                       | protein synthesis                |
| SCTR          | 1        | 2.8874    | socialisation                                                                  | social behaviour                 |
| SDPR          | 0.98745  | 2.3358    | endurance                                                                      | angiogenesis                     |
| SFPQ          | 1        | 2.4583    | vision sensitivity to diurnal light and circadian rhythms                      | circadian clock                  |
| Gene     | P-value  | Free-ratio | Hypothesis                        | Function                  |
|----------|----------|------------|-----------------------------------|---------------------------|
| SLC1A7   | 2.1567   |            | vision sensitivity to diurnal light and circadian rhythms | vision                    |
| SPACA3   | 0.0001   | 3.0126     | reproduction                      | sperm                     |
| STAP1    | 0.99486  | 2.2366     | cell function                     | docking protein           |
| SURF1    | 1        | 2.1957     | endurance                         | mitochondria respiration   |
| TAF8     | 0.9784   | 2.9587     | endurance                         | adipogenesis               |
| TMEM45A  | 0.9962   | 4.899      | endurance                         | hypoxia                   |
| unknown  | 0.9995   | 2.1152     | unknown                           | unknown                   |

Table S3: 31 genes under positive selection in the *Homotherium* genome with high values (free-ratio > 2) detected using one-ratio/free-ratio models and their respective hypothetical biological and functional roles. Related to Figure 2 and STAR methods and tests of positive selection.