Appendix S1. Supplementary Methods

Animals

EDTA blood and hair root sampling was approved by the “Cantonal Committee for Animal Experiments” (Canton of Bern; permits BE77/13). Genomic DNA from blood was isolated using the Nucleon Bacc2 kit (GE Healthcare Life Sciences). The DNeasy Blood & Tissue Kit (Qiagen) was used for hair root samples.

Histological examination

A single wattle was taken after slaughtering and fixed in 4% buffered formalin. After processing the tissue was embedded in paraffin, sectioned at 4 µm and stained with hematoxylin and eosin (HE).

SNP genotyping and genome-wide association study

DNA of the 341 goats was genotyped with the Illumina goat SNP50 BeadChip containing 53,346 SNPs. Initially, we removed all individuals and markers with call rates <95%. We also excluded markers strongly deviating from Hardy-Weinberg equilibrium ($p = 0.0001$) and markers with a minor allele frequency of <5% using the Plink software (Purcell *et al.* 2007). Parentage verification was performed with the --mendel option of Plink. After these quality control steps 328 individuals and 48,991 SNPs remained for the subsequent analysis. We performed an allelic association study and analysed the data with a principal component model which corrects population stratification using the egscore function (Price *et al.* 2006) implemented the GenABEL package in the R environment (Aulchenko *et al.* 2007).

Genotyping

Primers for the amplification of three coding variants were designed with the software Primer3 (http://bioinfo.ut.ee/primer3-0.4.0/) after masking repetitive sequences with RepeatMasker software (http://www.repeatmasker.org).

PCR primers used for genotyping of three coding variants.					
CHIR 10 position	**Primer-ID**	**Forward**	**Reverse**	**Anneal. temp. (°C)**	**Product size (bp)**
25952169	***FMN1_EX2***	**TCATGGTCTTCTCTGCTTTGG**	**TTAGCAGCCTCGCAGACAC**	**60**	**303**
25973006	***FMN1_EX3***	**TCCTGTTGCTCTCCGTTCTG**	**TCCGATCCTCTACTGGGCTC**	**60**	**300**
26072689	***FMN1_EX7***	**AAGCCAAAATTCTACGCCAAC**	**TGGCTCCTTGATGTCATCTG**	**60**	**299**

PCR was performed with AmpliTaqGold360Mastermix (Life Technologies) and the obtained products were loaded on 1% agarose gels for visual inspection of band size. PCR products were subsequently directly sequenced by Sanger sequencing using BigDye Terminator Sequencing Kit 3.1 (Life Technologies) on an ABI 3730 (Life Technologies) after treatment with exonuclease I (New England Biolabs) and rAPid Alkaline Phosphatase (Roche). Sequence data were analysed with Sequencher 5.1 (GeneCodes).

Whole-genome sequencing

We prepared fragment libraries with 300 bp mean insert size for each family member and obtained 113.5 mio, 158.6 mio and 242.9 mio 101 bp paired-end reads per sample with an Illumina HiSeq2500. We assessed the quality of the reads using fastqc v. 0.10.1. The reads were mapped against the goat reference genome (CHIR 1.0; NCBI) with Bowtie2 v. 2.1.0 using the “sensitive” preset option in end-to-end mode and assuming minimum and maximum fragment length of 50 and 550 bp respectively. We marked possible PCR duplicates using MarkDuplicates from Picard tools v. 1.80. The estimated duplication level varied between 3.1% and 4.1% in the three samples. To correct misalignments around insertion/deletion sites, local realignment was performed by running IndelRealigner from the Genome Analysis Tool Kit (GATK) v. 2.6.4 on each sample separately (McKenna et al. 2010). Each base was covered by an across genome average of 14.51 reads in the daughter, 7.74 reads in the mother, and 10.73 reads in the father. GATK’s Unified Genotyper was used to call SNPs and indels simultaneously in the three individuals considering only

bases with a quality of at least 17. Low quality variants were removed by applying a hard filter using the summary statistics and thresholds recommended on the GATK website. The IGV software (www.broadinstitute.org/igv/) was used for visual inspection of sequence variants.

References

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