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Original Research

A Genome-Wide Association Analysis in Noriker Horses Identifies a SNP Associated With Roan Coat Color

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ABSTRACT

The roan coat color in horses is characterized by dispersed white hair and dark points. This phenotype segregates in a broad range of horse breeds, while the underlying genetic background is still unknown. Previous studies mapped the roan locus to the *KIT* gene on equine chromosome 3 (ECA3). However, this association could not be validated across different horse breeds. Performing a genome-wide association analysis (GWAS) in Noriker horses, we identified a single nucleotide polymorphism (SNP) (ECA3:g.79,543.439 A > G) in the intron 17 of the *KIT* gene. The *G* -allele of the top associated SNP was present in other roan horses, namely Quarter Horse, Murgese, Slovenian, and Belgian draught horse, while it was absent in a panel of 15 breeds, including 657 non-roan horses. In further 379 gray Lipizzan horses, eight animals exhibited a heterozygous genotype (*A/G*). Comparative whole-genome sequence analysis of the *KIT* region revealed two deletions in the downstream region (ECA3:79,533,217_79,533,224delTCGTCTTC; ECA3:79,533,282_79,533,285delTTCT) and a 3 bp deletion combined with 17 bp insertion in intron 20 of *KIT* (ECA3:79,588,128_79,588,130delinsTTATCTCTA-TAGTAGTT). Within the Noriker sample, these loci were in complete linkage disequilibrium (LD) with the identified top SNP. Based upon pedigree information and historical records, we were able to trace back the genetic origin of roan coat color to a baroque gene pool. Furthermore, our data suggest allelic heterogeneity and the existence of additional roan alleles in ponies and breeds related to the English

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Thoroughbred. In order to study the roan phenotype segregating in those breeds, further association and verification studies are required.

1. Introduction

The knowledge of coat color genetics in horses rapidly progressed within the last decades. One milestone was the detection of a single missense variant in the equine melanocortin-1-receptor gene (*MC1R*), causative for chestnut coat color by Marklund et al. [1]. With the identification of an 11-bp deletion in exon 2 of the agouti-signaling-protein *(ASIP)* gene associated with black coat color by Rieder et al. [2], the basic colors of horses could be described on a molecular level. Our knowledge regarding diluted colors was deepened by a series of publications by Mariat et al. (cream, *SLC45A2*/*MATP*) [3], Imsland et al. (dun, *TBX3*) [4], Cook et al. (champagne, *SLC36A1*) [5], and Brunberg et al. (silver, *PMEL*) [6]. In 2008 Pielberg et al. [7] successfully identified the causative variant for gray coat color, whereas for the high phenotypic variability of leopard spotting (*LP*) patterns up to date two genetic factors were identified: the causative variant for *LP* [8] and the modifier pattern1 (*PATN1*) [9]. The increasing scientific interest in the white patterns in horses started with the assignment of the Overo spotting pattern to *ENDRB* [10]. In on-going research, the *KIT* gene encoding the KIT proto-oncogene, receptor tyrosine kinase, was found to harbor numerous genetic variants, which are responsible for a wide range of white coat color patterns. To date, the patterns Sabino1 [11], Tobiano [12], Dominant White (up to 28 different alleles within *KIT*) were described by [13–23]. Splashed White patterns were found to be caused by variants in *MITF* or *PAX3* [17,24].

The causative genetic background for roan coat color, which segregates in a wide range of horse breeds, is still unknown. Marklund et al. [25] proposed *KIT* as a candidate gene for roan coat color and identified an association between several *KIT* variants and the roan coat color in Belgian draught horses. The proposed variants could not be verified in other horse breeds (Welsh, Shetland, and Gotland Pony), exhibiting a roan phenotype, which led the authors to conclude that the genetic background of roan coat color may be heterogeneous across breeds. Furthermore, Marklund et al. [25] confirmed the dominant mode of inheritance of the roan allele *Rn* and claimed the lethality of homozygous roan horses, which was already proposed by Hinz and VanVleck [26]. The hypothesis of the lethality in homozygous roan horses currently has been disputed by Sponenberg and Bellone [27], who postulate the existence of homozygous roan horses.

Performing a genome-wide association analysis (GWAS) in Noriker horses, we identified an SNP (AX-103594067, ECA3:g.79,543,439A > G) in intron 17 of the *KIT* gene with the *G*-Allele associated with roan coat color [28]. Pedigree analysis and offspring ratios supported the homozygous state of *G/G* roan Noriker horses. Based on these results, a homozygous G/G roan Noriker stallion was identified as a reference animal for further whole-genome sequence analysis [29]. The aim of this study was to

Table 1
Description of the samples used for this study, including the data source for the top-SNP AX-103594067(ECA3:g.79,543,439A > G) and the three genetic variants A2 (3:79533214), A3 (3:79533281), B1 (3:79588127–29).

Non-roan horses	coat color	N	data source top-SNP	data source genetic variants
Akhal Teke	non-roan	36	SNP chip	
Appaloosa	non-roan	2		genotyping all 4 variants
Belgian draught horse	roan	1		genotyping all 4 variants
Bosnian Mountain Horse	non-roan	23	SNP chip	
Exmoor Pony	non-roan	256	SNP chip	
Franches Montagnes	non-roan	80	SNP chip	
German Sport horse	non-roan	1		genotyping all 4 variants
German Sport horse	roan	3		genotyping all 4 variants
Gidran	non-roan	20	SNP chip	
Lipizzan black/bay	non-roan	10	SNP chip	
Lipizzan, gray*	non-roan	379	SNP chip	
Murgese	non-roan	2		genotyping all 4 variants
Murgese	roan	2		genotyping all 4 variants
Noriker	non-roan	31		genotyping all 4 variants
Noriker	non-roan	7	SNP chip	genotyping all 4 variants
Noriker	non-roan	119	SNP chip	
Noriker	roan	26		genotyping all 4 variants
Noriker	roan	11	genotyping top-SNP	
Noriker	roan	14	SNP chip	
Posavina	non-roan	28	SNP chip	
Quarter Horse	non-roan	3		genotyping all 4 variants
Quarter Horse	non-roan	2		genotyping all 4 variants
Quarter Horse	roan	5		genotyping all 4 variants
Quarter Horse	roan	2	SNP chip	genotyping all 4 variants
Shagya Arabian	non-roan	33	SNP chip	
Shetland Pony	non-roan	2		genotyping all 4 variants
Shetland Pony	roan	1		genotyping all 4 variants
Shetland Pony	roan	2	SNP chip	genotyping all 4 variants
Slovenian draught horse	roan	3	SNP chip	genotyping all 4 variants
Trakehner	roan	1		genotyping all 4 variants
Trotter	non-roan	1		genotyping all 4 variants
All		1106		

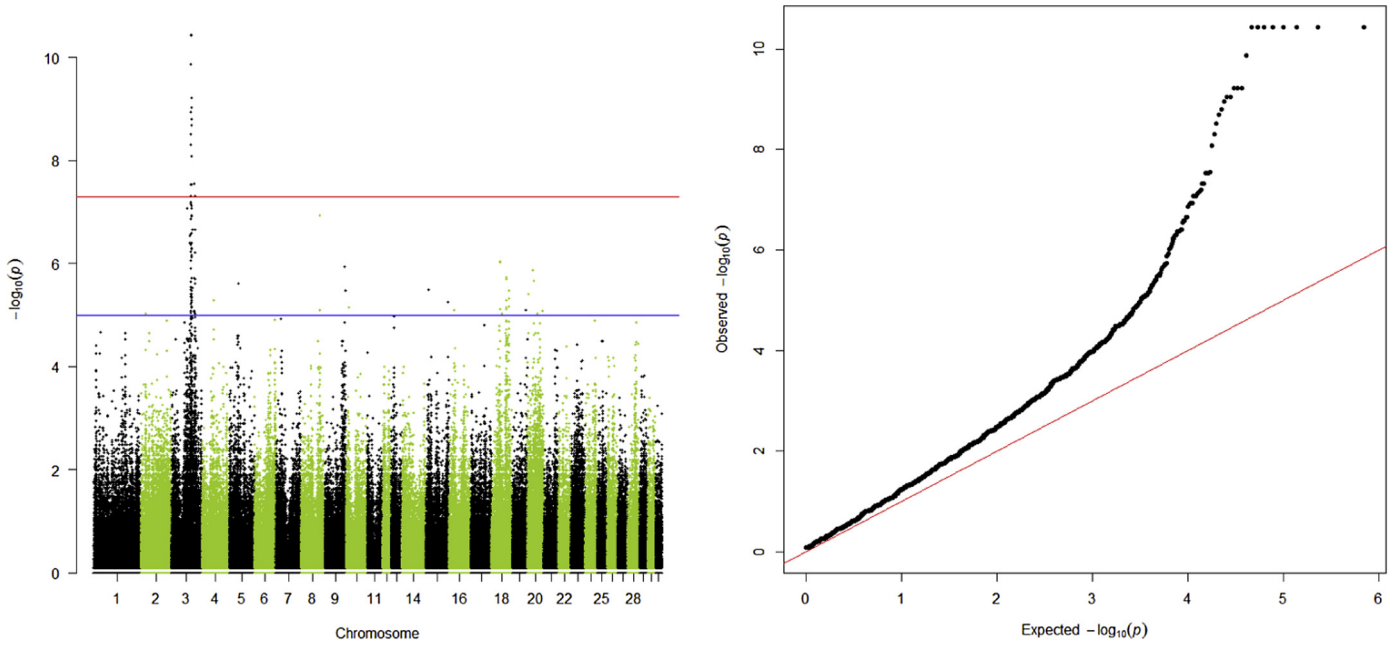

Fig. 1. Manhattan plot and Quantile-Quantile plot for the GWAS of roan versus black Noriker horses.

Table 2
Genotypes for the top-SNP AX-103594067(ECA3:g.79,543,439A > G) for 1035 non-roan/71 roan horses, derived from 670k SNP data and genotyping with KASP technology.

Non-roan horses	N	Genotype *A/A*	Genotype *A/G*	Genotype *G/G*
Appaloosa	2	2	0	0
Akhal Teke	36	36	0	0
Shagya Arabian	33	33	0	0
Franches Montagnes	80	80	0	0
Bosnian Mountain Horse	23	23	0	0
Posavina	28	28	0	0
Gidran	20	20	0	0
Lipizzan black/bay	10	10	0	0
Lipizzan, gray[a]	379	371	8	0
Quarter Horse	3	3	0	0
Quarter Horse	2	2	0	0
Noriker	31	31	0	0
Noriker	7	7	0	0
Noriker	119	119	0	0
Exmoor Pony	256	256	0	0
Shetland Pony	2	2	0	0
German Sport horse	1	1	0	0
Murgese	2	2	0	0
Trotter	1	1	0	0
All[b]	1035	1027	8	0
Roan horses				
Noriker	51	0	36	15
Quarter Horse	7	0	7	0
Slovenian draught horse	3	0	3	0
Murgese	2	0	2	0
Belgian draught horse	1	0	0	1
Shetland Pony	3	3	0	0
Trakehner	1	1	0	0
German Sport horse	3	3	0	0
All[b]	71	7	48	16

[a] for the 379 gray Lipizzan horses the underlying coat color was not known, but roan color is known to segregate in a specific mare family.
[b] Fisher's exact test for genotype association (A/A and G/-) with roan/non roan coat color for the entire sample revealed a *P*-value $P < .001$.

further explore the genome region around the associated top-SNP AX-103594067 and to better understand the underlying genetic background of roan coat color in horses.

2. Material and Methods

2.1. Genome-wide SNP (Single Nucleotide Polymorphism) Data and Genome-wide Association Analysis

From previous studies [30–33] 670k SNP data for a total of 1012 horses was available, including the following breeds: 140 Noriker, 36 Akhal Teke, 33 Shagya Arabian, 80 Franches Montagnes, 23 Bosnian Mountain Horse, 28 Posavina, 20 Gidran, 389 Lipizzan, two Quarter horses, two Shetland Ponies, three Slovenian draught horses, and 256 Exmoor Ponies (Table 1).

The GWAS was performed, including 40 Noriker horses using a case-control design (case-group: 14 blue roan Noriker horses, control-group: 26 black Noriker horses) and associations were corrected for multiple testing using the Fisher's exact test. SNP extraction and GWAS were performed using the software package PLINK v.1.7 [34]. Quality control (QC) and SNP filtering were applied on the Noriker data set using a minor allelic frequency (MAF) threshold of <0.01. After QC, a total of genome-wide 464,880 SNPs were included in the GWAS.

For the verification of genotype and haplotype distribution of roan/non-roan horses of the entire multibreed data set, we applied Fisher's exact test. Statistical analyses and graphical representations were performed using the R-platform (www.r-project.com).

2.2. Genotyping

For further verification of our top SNP (AX-103594067) and further putative variants, we genotyped 97 horses (roan/non-roan), including the following breeds: Noriker (26/38), Quarter horse (7/5), Appaloosa (0/2), Murgese (2/2), Belgian draught horse (1/0), Slovenian draught horse (3/0), Shetland Pony (3/2), Trakehner (1/0), Trotter (0/1), Warmblood (3/1) (Table 1). In addition, we genotyped 11 archived roan Noriker horses only for the top SNP (AX-

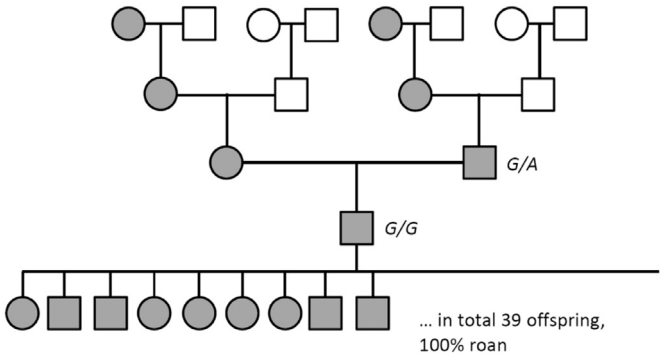

Fig. 2. Pedigree information of the roan stallion NO180 given as an example for the segregation of roan coat color in Noriker horses. Stallions are illustrated by squares, mares by circles; roan phenotype is represented by gray color, non-roan phenotype by white color. On the bottom NO180, homozygous *G/G* at the top SNP AX-103594067 (ECA3:g.79,543,439A > G), and his 100% roan offspring, including 39 foals are shown.

103594067). Genomic DNA was isolated from hair roots using nexttecTM Tissue & Cells Kit, following the manufacturer's protocol. For genotyping, competitive allele-specific PCR SNP genotyping assays (KASP) were used. LGC KASP assays were designed to genotype five variants (Table 3) using the LGC service (http://www.lgcgroup.com). KASP screening was performed as described by the supplier on a CFX96 Touch Real-Time PCR Detection System.

2.3. Pedigree- and ROH Analysis

The genotype plausibility (checking sire and dam coat color, offspring ratios) of 51 roan Noriker horses were examined, including the pedigree information of 55.567 horses from the Noriker studbook [35]. Furthermore, we performed a ROH analysis for eight roan Noriker horses homozygous *G* for the top-SNP AX-103594067 with an overlapping window approach as implemented in PLINK v1.7 [34] based on the following settings: minimum SNP density of ROH segments was set to one SNP per 50 kb with a maximum gap length of 50 kb; final segments were called runs of homozygosity (ROH) if the minimum length of the homozygous segment was greater than 125 kb and comprised more than 20 homozygous SNPs; one heterozygote and one missing genotype were permitted within each segment.

The whole-genome sequence of one blue roan (black base color) Noriker stallion (NO180), verified as homozygous for the roan factor [28], was available from [29]. The variant callings from NO180 (published in [29]) in the ROH region of homozygous Noriker horses, spanning the *KIT* gene (ECA3:79,472,667–79,694,802), were comparatively analyzed with the sequences published for 87 horses from different breeds [29] (4 Akhal-Teke, 4 American Paint Horse, 1 American Standardbred, 2 Arabian, 1 Polish Warmblood, 3 German Warmblood, 29 Franches-Montagnes, 4 Haflinger, 2 Hannoveraner, 8 Holsteiner, 2 German Riding Pony, 1 Morgan Horse, 4 Icelandic, 1 Dutch Warmblood, 3 Oldenburger, 3 Quarter Horse, 4 Swiss Warmblood, 3 Shetland Pony, 1 Thoroughbred, 3 Trakehner, 2 UK Warmblood, 2 Welsh Pony). For the majority of these horses (84), information on coat color phenotypes was provided for the present study (Suppl. 1). Among the 84 horses with color phenotypes and whole-genome information [29], three Quarter horses were roan. However, although the roan coat color is known in a variety of horse breeds, the overall incidence of roan horses in the sample set of the 88 horses with whole-genome information [29] remained low. Accordingly, we filtered the variant calls published in [29] for positions on ECA3:79,472,667–79,694,802 that are homozygous in NO180 and called at maximum in five additional horses.

Finally, a comparison analysis between the variants identified by Marklund et al. [25] in the Belgian draught horse and the variants inferred in this study was conducted.

3. Results

The GWAS in 40 Noriker horses revealed eight SNPs on ECA3 that were significantly associated with roan coat color. These variants were located in the *KIT* genic region (Fig. 1). The top-SNP (AX-103594067, ECA3:g.79,543,439A > G,) was either heterozygous or homozygous for the alternate allele in 64 roan horses (51 Noriker, three Slovenian draught horses, seven Quarter horses, two Murgese horses, and one Belgian draught horse), whereas all non-roan horses were homozygous for the wildtype-allele (*A/A*) (Table 2). One-third of the roan Noriker horses (n = 15) and one Belgian draught horse were homozygous for the roan associated *G* allele (Table 2). The *G* allele was not present in three roan Shetland Ponies, one roan Trakehner, and three roan German Sport horses. In Table 2, the genotype distribution for the top-SNP AX-103594067 of 1036 non-roan horses and 71 roan horses is illustrated. Among 379 gray Lipizzan horses, eight heterozygous *A/G* carriers could be detected (Table 2).

Pedigree analyses of roan Noriker horses were in concordance with the genotyping results and supported a homozygous state and the dominant mode of inheritance. Eight of the investigated roan Noriker horses with homozygous state *G/G* of the top SNP AX-103594067 had 72 documented offspring, whereas 70 horses were roan, one colt was leopard-spotted, and one foal was tobiano. In Fig. 2 the pedigree information of the sequenced Noriker stallion (NO180) is presented. Based on 670K SNP chip data from eight roan Noriker horses homozygous *G* for the top-SNP AX-103594067, we identified a 222 kb homozygous region from ECA3:79,472.667–79,694.802 containing the entire *KIT* gene. The comparative sequence analysis of the region ECA3:79,472,667–79,694,802 in 88 horses [29], resulted in a total of 1552 variants. Assuming that the roan factor should be present in a homozygous state in animal NO180, 314 variants remained, from which 88 variants were private for this animal. Applying a filter that allows five animals to share these variants, 115 candidate variants were retained. Finally, we selected two regions (A and B) harboring structural variants as putative candidate loci for roan coat color. In the 3′-flanking region of *KIT* (region A), three deletions were identified and in intron 20 of the *KIT* gene (Region B), a 3 bp

Table 3
Location, reference allele (REF) and alternate allele (ALT) for the three deletions found in region A (A1, A2, A3), the top-SNP AX-103594067 from the GWAS (SNV) and the insertion in region B (B1) associated with roan coat color.

Variant	Position on EquCab3	REF	ALT (NO180)
A1	ECA3:79,531,997_79,532,011	TTCCATGATTAATTA	—
A2	ECA3:79,533,217_79,533,224	CTCGTCTT	—
A3	ECA3:79,533,282_79,533,285	TTCT	—
SNV	ECA3:79,543,439	A	G
B1	ECA3:79,588.127–29	ACA	TTATCTCTATAGTAGTT

Table 4
Genotyping results of the four genotyped variants and respective haplotypes (homozygous for reference allele (Hom.REF), homozygous for alternate allele (hom.ALT) or heterozygous (Het.), roan horses without the roan haplotype, are marked with*.

Non-roan horses	n	Haplotype	A2 (3:79533214)	A3 (3:79533281)	SNP (3:79543439)	B1 (3:79588127–29)
Noriker	38	00-0-0/00-0-0	Hom.REF	Hom.REF	Hom.REF	Hom.REF
Quarter Horse	5	00-0-0/00-0-0	Hom.REF	Hom.REF	Hom.REF	Hom.REF
Appaloosa	2	00-0-0/00-0-0	Hom.REF	Hom.REF	Hom.REF	Hom.REF
Murgese	2	00-0-0/00-0-0	Hom.REF	Hom.REF	Hom.REF	Hom.REF
Trotter	1	00-0-0/00-0-0	Hom.REF	Hom.REF	Hom.REF	Hom.REF
German Sport horse	1	00-0-0/00-0-0	Hom.REF	Hom.REF	Hom.REF	Hom.REF
Shetland Pony	2	00-0-0/00-0-0	Hom.REF	Hom.REF	Hom.REF	Hom.REF
All[a]	51					
Roan horses						
Noriker	18	11-1-1/00-0-0	Het.	Het.	Het.	Het.
Noriker	8	11-1-1/11-1-1	Hom.ALT	Hom.ALT	Hom.ALT	Hom.ALT
Quarter Horse	7	11-11/00-0-0	Het.	Het.	Het.	Het.
Slov.Coldblood	3	11-1-1/00-0-0	Het.	Het.	Het.	Het.
Belgian Draught	1	11-1-1/11-1-1	Hom.ALT	Hom.ALT	Hom.ALT	Hom.ALT
Murgese	2	11-1-1/00-0-0	Het.	Het.	Het.	Het.
Shetland Pony*	3	00-0-0/00-0-0	Hom.REF	Hom.REF	Hom.REF	Hom.REF
Trakehner*	1	00-0-0/00-0-0	Hom.REF	Hom.REF	Hom.REF	Hom.REF
German Sport horse*	3	00-0-0/00-0-0	Hom.REF	Hom.REF	Hom.REF	Hom.REF
All[a]	46					

[a] Fisher's exact test for haplotype association (Hom.REF and Hom.ALT/Het.) with roan/non-roan coat color for the entire sample revealed a *P*-value $P < .001$.

deletion combined with 17 bp insertion was detected (Table 3). All five variants were homozygous for the alternate allele (ALT) in the Noriker stallion NO180. LGC KASP assays were designed for the genotyping of the associated top SNP AX-103594067 from the 670K SNP Chip, the three deletions located downstream of the *KIT* gene (A1-A3) and the 3 bp deletion combined with 17 bp insertion in intron 20 of *KIT* (B1). The genotyping results revealed that the deletion A1 was not roan specific, and was, therefore, excluded from further analyses.

The remaining two deletions (A2 and A3) and the 3 bp deletion combined with 17 bp insertion (B1) were solely detected in roan horses for the breeds Noriker, Quarter Horse, Murgese, Slovenian and Belgian draught horse. These three loci were in complete LD with the top SNP AX-103594067 at ECA3g:79,543,439A > G and the resulting roan-associated haplotype was assigned 11-1-1 (variants phased, REF = 0, ALT = 1) (Table 4., Fig. 3). This roan haplotype was not present in three roan German Sport Horses and one roan Trakehner and three roan Shetland Ponies. All these animals exhibited the same haplotype (00-0-0) as non-roan horses.

The comparison of the identified variants with the regions associated with roan coat color by Marklund et al. [25] showed a close proximity, especially for the SNP AX-103594067 (Table 5), and two roan associated sites of Marklund et al. [25] (ECA3:79,545,912G > A in site SSCP, ECA3:79,540,501G > A in site TaqI) were confirmed in the homozygous stallion NO 180 (Table 5).

4. Discussion

For a long time, the homozygous state of a dominant roan coat color allele was supposed to be lethal in utero according to

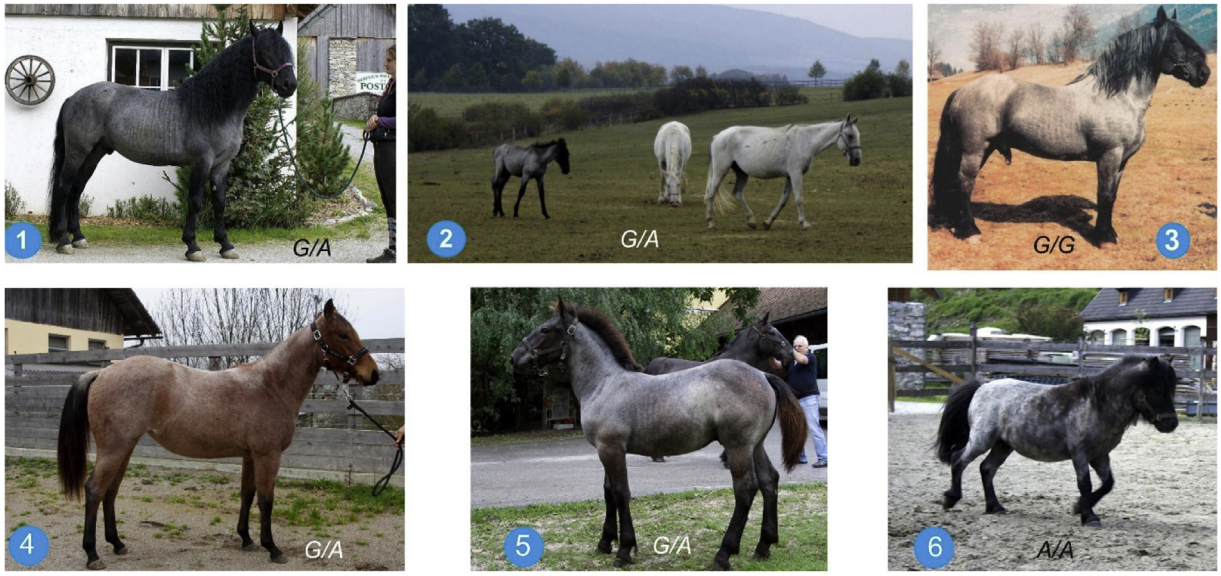

Fig. 3. Roan color phenotype associated with different roan haplotypes: (1) Murgese stallion (haplotype: 11-1-1/00-0-0), (2) Lipizzan colt before greying (haplotype: 11-1-1/00-0-0), (3) Noriker stallion (haplotype: 11-1-1/11-1-1), (4) Quarter Horse (haplotype: 11-1-1/00-0-0) and (5) Slovenian draught horses (haplotype: 11-1-1/00-0-0). The Shetland Pony in (6) is not carrying the reported roan haplotype (haplotype: 00-0-0/00-0-0) (images by Grilz-Seger, Mesaric, archive Druml).

Table 5
Distance between the top SNP AX-103594067 and the sites associated with roan coat color by Marklund et al. (1999).

ID	Study	position	Distance to top-SNP	NO180	REF	ALT
CfoI	Marklund et al. (1999)	ECA3:79,538,738	4701 bp	Hom.REF	C	T
TaqI	Marklund et al. (1999)/this study	ECA3:79,540,501	2938 bp	Hom.ALT	G	A
SNV	this study	ECA3:79,543,439		Hom.ALT	A	G
SSCP	Marklund et al. (1999)/this study	ECA3:79,545,912	2473 bp	Hom.ALT	G	A

offspring ratios [25,26]. Sponenberg and Bellone [27] proposed the existence of living homozygous Rn/Rn roan horses, which was confirmed in our study.

Marklund et al. (1999) [25] reported an incomplete association between roan phenotype and genotype information in 33 roan and 92 nonroan horses, mapped to the *KIT* gene. With the investigation of additional breeds (Welsh, Shetland, and Gotland Pony), the level of association declined, which led the authors to assume genetic heterogeneity. Our study recapitulates these findings as the genetic association for roan coat color was not complete. However, we found a complete association between a roan haplotype in Noriker and further draught horses (Belgian and Slovenian draught horse), as well as Murgese, and seven Quarter horses of our dataset. The identified roan haplotype was not present in three roan Shetland Ponies, three German Warmblood horses, and one Trakehner. The three roan Quarter horses published in [29] did also not exhibit the roan haplotype. Our four sampled roan German Sport horses and Trakehner horses all descended from the same stallion and might carry a very rare roan allele. The existence of breed-specific segregation of coat color alleles is well known for Sabino (*SB1*) and chestnut (*MC1R*) [11,36]. The *SB1* allele associated with the Sabino phenotype was firstly detected in the Tennessee Walking horse [11], and it was further documented in numerous breeds, including American Miniature, Paint Horse, Azteca, Missouri Foxtrotter, Shetland Pony, and Spanish Mustang. Brooks et al. [11] further demonstrated that within breeds with the classical Sabino phenotype like Shire Horse or Clydesdale, the SB1 allele did not segregate, a result that was also confirmed by Reissmann et al. [37]. Further allelic heterogeneity at the KIT locus was reported for Dominant White, where several breeds or even family-specific mutations are responsible for depigmented phenotypes in horses [38].

The breed-specific segregation of roan associated alleles may be explained by the population history of the breeds. Roan coat color represented a major breeding objective in Old-Italian and Old-Spanish horse populations before 1800 [39,40]. The breeds Lipizzan, Murgese, Noriker, Slovenian draught horse (mainly derived from Noriker horses), Belgian draught horse, and Quarter Horse, are directly connected to this baroque gene pool or founder populations are proven by either pedigree or historical records.

For the draught horse breeds in this sample, a direct connection to the baroque gene pool can be found in imperial stud farms established in original breeding areas of respective breeds: for example, in the clerical stud farm Rif near Salzburg, which interacted with local breeding (horse in Noriker-like type), roan horses were already recorded in the year 1652 [41]. A similar gene-flow between a baroque gene pool and local working horse populations of later on Belgian draught horse type like horses and introgression of roan can be assumed from the imperial stud farm Alost, founded by the Austrian empire in Belgium in 1770 [42].

In our sample, Shetland Ponies and breeds derived from English Thoroughbred were characterized by the absence of the roan associated *G*-allele and haplotype, and thus, the roan phenotype in these breeds may be traced back to another founder population. Introgression of single dominant inherited traits was mainly conducted using single breeding animals, especially stallions. Y-chromosomal studies [43] revealed that Northern European Pony breeds and the Thoroughbred cluster show distinct population history and genetic distances to the core clusters, which mainly evolved from a unique group, including the descendant breeds from a prior baroque gene pool.

5. Conclusion

Our results suggest the SNP AX-103594067 (ECA3g:79,543,439A > G) in the *KIT* gene as a genetic marker for roan coat color in Noriker horses and as a putative marker in draught horse breeds, Lipizzans and Murgese. Our data supports the previously postulated allelic heterogeneity in roan horses, as the marker was not identified in roan Shetland ponies and roan Thoroughbred-related breeds. To validate the marker and the associated roan haplotype in other breeds than Noriker, we suggest further investigation within a bigger panel comprising additional multibreed samples.

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Supplementary data

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