

## **Genetic structure analysis of American Quarter Horses\***

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The study investigated inbreeding and relationship in American Quarter Horses (AQ) kept or bred on the Roleski Ranch stud farm (Stare Żukowice, Małopolska region). The data consisted of pedigrees of 76 AQ horses (40 stallions and 36 mares) born in 1993-2012. Coefficients of inbreeding ( $F_x$ ) and relationship ( $R_{xy}$ ) were calculated according to Tier. The effective number of founders ( $f_e$ ) and effective number of ancestors ( $f_a$ ) were calculated as well. Almost 91% of horses were found to be inbred – 90% of stallions and 91.67% of mares. In sex groups,  $F_x$  averaged 0.0132 and 0.0142 for all stallions and all mares, and 0.0146 and 0.0155 for inbred stallions and inbred mares. About 97.37% of animal pairs were found to be related. Over 99% of male pairs were related, with  $R_{xy}$  for all and inbred males averaging 0.0610 and 0.0641, respectively. In females, nearly 95% of pairs were related, but  $R_{xy}$  values were slightly lower, averaging 0.0590 and 0.0591 for all and related pairs, respectively. Among mixed male-female pairs 97.5% were related, with  $R_{xy}$  values averaging 0.0607 and 0.0622 for all and related pairs, respectively. The  $f_e$  was 121 and the  $f_a = 26$ . Half of the genetic pool was explained by contributions of only 10 ancestors, and 90% of alleles originated in 39 ancestors. Due to steady importation of stock AQ horses and careful mating plans, the inbreeding and relationship coefficients on the Roleski Ranch stud farm are relatively low.

**KEY WORDS:** inbreeding / relationship / founders / ancestors / American Quarter Horse

Maintaining genetic variation at a satisfactory level is particularly important in small populations, in which it is usually fairly difficult to avoid matings of related animals and an increase in homozygosity, which may result in the appearance of genetic defects. Monitoring of genetic variation in such populations is therefore a key problem for breeders [19, 29].

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Inbreeding and relationship coefficients and analysis of the contributions of ancestors and founders are usually useful tools for detecting short-term changes in the genetic structure of a population. The occurrence of a common ancestor or ancestors in the pedigree (going back more than 20 generations), resulting in inbreeding, does not have such far-reaching negative consequences as the occurrence of such an ancestor or ancestors up to 20 generations back. Thus the threshold number of generations in estimating inbreeding and relatedness is 20 [19, 29].

The American Quarter Horse is currently one of the most numerous breeds of horse in the world. According to data from the Annual Report of the American Quarter Horse Association (AQHA), at the end of 2013 there were 2,935,825 individuals registered in the studbook, of which over 2,533,000 were registered in the United States alone and over 36,000 in Germany. In contrast, in Poland only 650 animals of this breed have thus far been registered [2].

American Quarter horses began to appear in Poland relatively recently, at the turn of the 21st century. They were primarily brought in from Germany, and much less often from the United States, due to the distance and high costs. In 2005 the Polish Quarter Horse Association was founded, and was recognized by the AQHA in 2007. From that time there has been a gradual but steady increase in the popularity of western-style riding in Poland. Roleski Ranch is one of the first AQ horse breeding facilities in Poland (currently the largest), and it is the first and as yet the only one that holds western-style competitions recognized by the National Reining Horse Association in the USA [16].

The genetic structure of the American Quarter breed has been the subject of research in the USA [21, 30], while in Germany research has been conducted on horses of the American Paint breed [25], most of which possess AQ genes and are not counted among this breed only due to their pinto coat colour. However, the genetic structure of American Quarter horses in Poland has not yet been studied. The Roleski Ranch stud has the largest herd of American Quarter horses, which as breeding horses already have or may in the future have an impact on breeding of this breed in Poland. Therefore the aim of this study was to analyse the genetic structure of this herd.

## **Material and methods**

The material consisted of the pedigrees of 76 horses of the American Quarter (AQ) breed, including 40 stallions and 36 mares, born in the years 1993-2012. The group included breeding mares, stallions, and male and female young born at the stud. All AQ horses, except the youngest animals, have been registered with the American Quarter Horse Association. The initial four-generation pedigrees were expanded to include further generations of ancestors using the Quarter Horse Directory data base (<http://qhd.com>). In this manner pedigrees were obtained whose completeness, estimated according to the method of Caspell et al. [6], corresponded to information obtained from complete pedigrees ranging in length from 4.5 to 7.1 generations. For the horses analysed the completeness of pedigrees estimated in this manner reached the sixth generation (precisely 5.92 generations). The inbreeding coefficients ( $F_x$ ) and relationship coefficients ( $R_{xy}$ ), as well as the contributions of founders and ancestors, were estimated for the reference population of the 76 tested

animals. Calculations were made according to an algorithm proposed by Tier [27], with a recursive modification reducing computation time [9].

To assess the ongoing effects of breeding work conducted at the stud, the total and effective number of founders and ancestors was estimated and the founders and ancestors with the highest contributions in the reference population were identified. The effective number of founders ( $f_e$ ) and effective number of ancestors ( $f_a$ ) were estimated according to the method proposed by Lacy [17, 18] as modified by Boichard et al. [4, 5], according to the following formulas:

$$f_e = \frac{1}{\sum_{k=1}^f q_k^2}$$

where:

$f_e$  – effective number of founders

$f$  – number of founders

$q_k$  – probability of gene origin from the  $k$ -th founder

$$f_a = \frac{1}{\sum_{k=1}^f p_k^2}$$

where:

$f_a$  – effective number of ancestors

$f$  – number of ancestors

$p_k$  – marginal contribution of  $k$ -th ancestor

## Results and discussion

Among the 76 American Quarter horses analysed 69 individuals (90.79%) were inbred. This included 91.67% of males, while among females 90% had  $F_x$  coefficients  $> 0$  (Tab. 1).

Individual inbreeding coefficients of inbred animals ranged from about 0.04% to 7.8%, without exceeding the critical value of 12.5% [8, 29, 33, 34].

Fig. 1 presents changes in inbreeding over time among stallions and mares in the Role-ski Ranch herd and in the group of ancestors of this group of American Quarter horses. Part A (Fig. 1A) presents inbreeding of female and male ancestors of the herd born in different years, and part B (Fig. 1B) shows inbreeding of the group of horses broken down by sex in relation to their year of birth.

Analysis of chart A reveals that in the group of ancestors the level of inbreeding displayed a downward trend from a value of over 10% in horses born before 1970 to under 5% in the 1990s and at the beginning of the 21st century. This is particularly evident in the male ancestors. The reverse tendency was noted for the number of inbred ancestors. In the early periods (1960s and 1970s), from 0.3% to over 6.5% of ancestors were inbred during that

**Table 1**  
Mean inbreeding coefficient ( $F_x$ ) for American Quarter Horses from Roleski Ranch

Specification	Sex	
	males	females
Number of animals in active population	40	36
Number of ancestors of active population	466	715
Number of animals in pedigrees	506	751
Number of inbred animals	36	33
Mean $F_x$ – all animals	0.0132	0.0142
Mean $F_x$ – inbred animals	0.0146	0.0155
Maximum $F_x$ value	0.0593	0.0780

period, while after 1980 the number of inbred ancestors increased, accounting for over 30% of ancestors born in the 1980s, and over 67% in the 1990s.

In the active population (Fig. 1B), particularly in the most recent period, a slight increase in inbreeding can be seen in both mares and stallions, while the percentage of inbred animals in this group has increased considerably. Up to the year 2000 it was 50%, but in the subsequent five-year periods it surpassed 90%; it was nearly 92% in 2001-2005 and over 94% in 2006-2010. All individuals in the youngest group of AQ horses from Roleski Ranch were inbred.

Among the horses analysed as many as 97.37% were related, including 99.84% males and 95.13% females, while 97.5% of male x female pairs had non-zero relationship coefficients (Tab. 2).

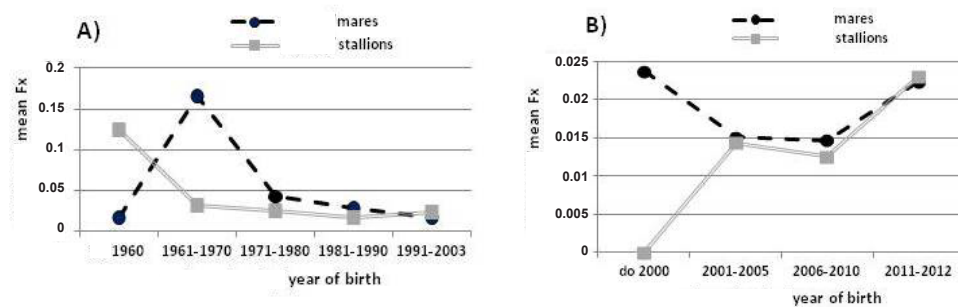


Fig.1. Mean inbreeding coefficients for mares and stallions from ancestor group (Fig. 1A) and from examined herd (Fig. 1B) over birth periods

**Table 2**  
Mean relationship coefficients ( $R_{XY}$ ) for American Quarter Horses

Specification	Pairs		
	males	females	mixed*
Total number of pairs of animals	630	780	1440
Number of related pairs of animals	629	742	1404
Mean $R_{XY}$ – all pairs of individuals	0.0590	0.0610	0.0607
Mean $R_{XY}$ – related pairs of individuals	0.0591	0.0641	0.0622
Maximum $R_{XY}$ value	0.5268	0.5371	0.5409

\*Male x female pairs

The total and effective numbers of founders in the herd of AQ horses at Roleski Ranch were 484 and 121, respectively, while the total and effective numbers of ancestors were 60 and 26. Half of the variation in the genetic pool of the reference population is explained by the contributions of only 10 ancestors, but the combined contributions of 39 ancestors were needed to explain 90% of its genetic pool (Tab. 3).

In the population analysed there were 17 founders whose contributions of alleles in the reference population of AQ horses exceeded 1%; this was 3.5% of all founders, and the sum of their contributions accounted for about 30% of the genetic pool of the population. The combined contributions of the 23 main ancestors (38.3% of all ancestors) explained about 79% of the genetic variation in the population of horses at Roleski Ranch.

Among the founders with the highest contributions of alleles in the population, the top two were the mare JABALINA (72570) and the stallion ZANTANON (81745), both with nearly a 3% contribution of alleles; these were the parents of the stallion KING (234), born in 1932 and present as the ancestor with 14th highest contribution. The group of founders also included two mares related to the stallion KING—his granddaughter POCO LENA (30475), with the eighth highest contribution, who was the daughter of the stallion POCO BUENO (Fig. 4), immediately followed by the mare SAN SUE DARKS (14898), the granddaughter of the stallion ZANTANON and daughter of another of his sons, SAN SIEMON.

The stallion WIMPY (1), born in 1937, placed 10th among the group of founders, with a contribution slightly over 1%. This was the first individual entered into the studbook of the American Quarter Horse breed.

Figures 2, 3 and 4 present the relationships between the ancestors with the highest contributions of alleles in the analysed group of American Quarter horses and the relationships between some of the ancestors and the founders.

Figure 2 shows the stallion DOC BAR (76136), born in 1956, whose contribution to the gene pool of ancestors was 8.5%, placing him second, and three other stallions related to

**Table 3**

Total and effective number of founders ( $f_e$ ) and ancestors ( $f_a$ ) for 76 horses from the Roleski Ranch reference population

Parameter	Value
Number of animals in the reference population	76
Number of animals in pedigrees	1257
Mean number of generations traced	9.9
Total number of founders	484
Effective number of founders ( $f_e$ )	121
Total number of ancestors	60
Effective number of ancestors ( $f_a$ )	26
Number of founders explaining 50% of the gene pool	50
Number of founders explaining 90% of the gene pool	264
Number of ancestors explaining 50% of the gene pool	10
Number of ancestors explaining 90% of the gene pool	39

him through the thoroughbred stallion THREE BARS, one of the progenitors of modern AQ horses. It also gives the names of stallions and mares descended from DOC BAR who had the largest contributions of alleles as ancestors of the analysed population. This group included the stallion LEAN WITH HOLLYWOOD (4317664), who placed first among the ancestors of the population, with an allele contribution of over 10.5%.

Figure 3 shows the stallion WIMPY, who was among the founders with the highest gene contributions in the population, together with his descendants of both sexes, who are among the population's most important ancestors.

Figure 4 shows the genealogical relationships between the stallion KING and his descendants of both sexes who were in the group of ancestors with the greatest contributions in the population of AQ horses.

Tunnel et al. [30] noted a high percentage of alleles of the thoroughbred stallion THREE BARS and AQ stallions such as KING, DOC BAR, ZANTANON and WIMPY among offspring winning awards in various categories of western-style competitions. The same stallions were also among the ancestors or founders with the greatest contributions of genes in the Roleski Ranch horses.

Petersen et al. [21], in a later study on the American Quarter breed, also name the stallion DOC BAR as the ancestor having the greatest allele contribution in the genotypes of

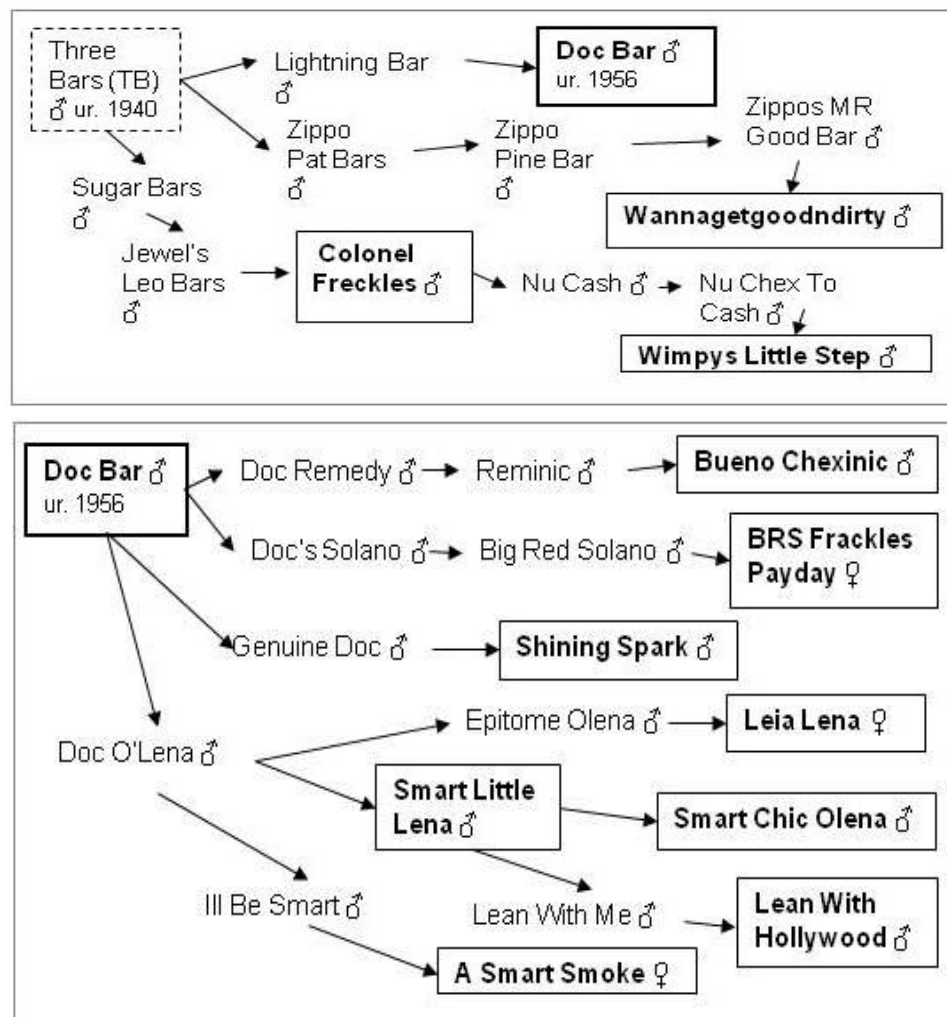


Fig. 2. Genealogical relations between the most influential ancestors (names are in bold and enclosed in boxes)

horses starting in the categories of reining, cutting (separating a cow from the herd) and working cow (work with cattle). The authors also name his son DOC O'LENA and grandson SMART LITTLE LENA (1565822) as belonging to the group of ancestors with the greatest allele contributions in horses starting in similar categories of western-style competitions. The latter also appeared as one of the ancestors with the greatest contributions in the present study, with a contribution of over 1% to the population.

Furthermore, in a study on German horses of the Paint breed [25], the vast majority of which have some American Quarter alleles, the stallions DOC BAR and KING appear

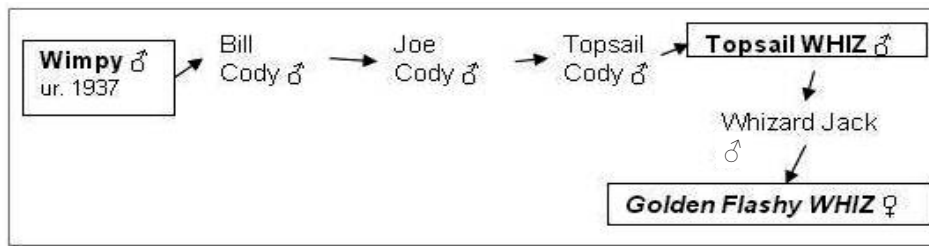


Fig. 3. Genealogical relations between the founder and its progeny – most influential ancestors (names are in bold and enclosed in boxes)

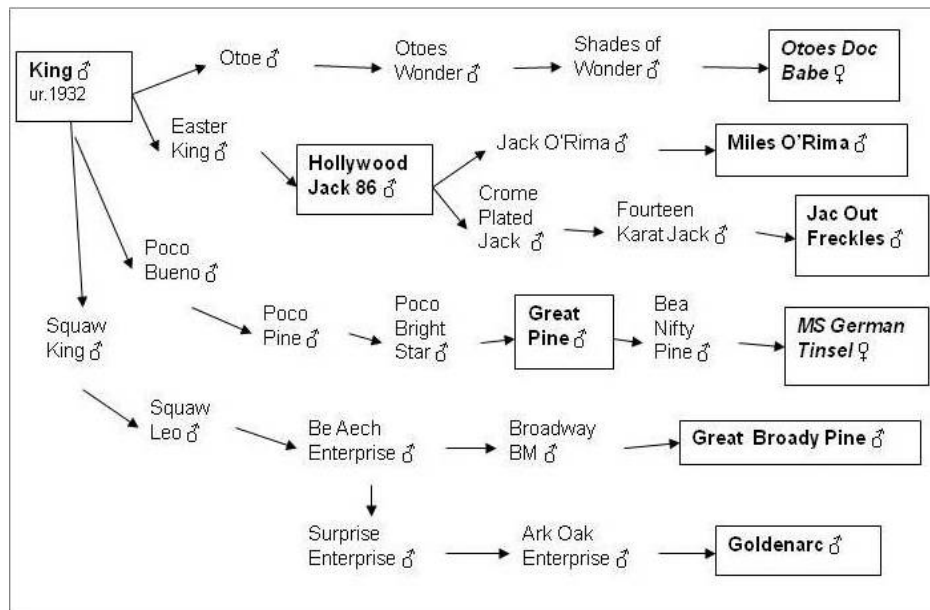


Fig. 4. Genealogical relations between the most influential ancestors (names are in bold and enclosed in boxes)

among the ancestors with the largest allele contributions, as well as the thoroughbred stallion THREE BARS with the top contribution.

Zechner et al. [35], in a study on Lipizzan horses, found that the diversity of ancestors in pedigrees is not always reflected in genetic diversity, which was partially confirmed in the AQ horses in the present study, in which the ancestors and founders with the greatest contributions were often more or less closely related (Figs. 2, 3, 4).



Table 4 presents results obtained by various authors on inbreeding and relatedness for various breeds of horse, while Table 5 gives the effective numbers of founders and ancestors obtained in studies of various breeds by different authors.

**Table 4**Inbreeding ( $F_x$ ) and relationship ( $R_{xy}$ ) coefficients in different Equidae breeds

Breed	$F_x$	$R_{xy}$	Source
American Quarter Horses	0.0020-0.0520	0.0016-0.0640	Tunnel et al., 1983 [30]
	0.0090-0.0370	0.0180-0.0550	Petersen et al., 2014 [21]
Paint Horses	0.0035-0.0049	–	Siderits et al., 2013 [25]
Arab horses	0.000-0.0980	0.0020-0.1160	Cervantes et al., 2008 [7]
	0.0306-0.0531	0.0379-0.0751	Głażewska and Jezierski, 2004 [11]
	0.0293-0.0536	–	Głażewska, 2004 [10]
Friesian horses	0.1450-0.1580	–	Sevinga et al., 2004 [24]
Catalonian Donkeys	0.0073-0.0722	0.0139-0.0661	Gutiérrez et al., 2005 [12]
Hanoverian horses	0.0119-0.0133	–	Hamann and Distl, 2008 [13]
Mallorquí horses	0.0250-0.0470	0.1010-0.1120	Álvarez et al., 2010 [1]
Norwegian Fjord horse breed	0.0160-0.0320	–	Bhatnagar et al., 2011 [3]
Belgian Draught horses	0.0290-0.0330	–	Janssens et al., 2010 [15]
Norwegian Døle and Nordland/Lyngen horses	0.1175-0.1277	–	Olsen et al., 2010 [20]
Trakehner horses	0.000-0.0120	0.000-0.0150	Teegen et al., 2009 [26]
Asturcón ponies	0.0410-0.0520	0.0890-0.0970	Royo et al., 2007 [23]
Andalusian horses	0.0806-0.1301	0.1197-0.1400	Valera et al., 2005 [31]
Lusitano horses	0.0992-0.1134	0.1164-0.1354	Vicente et al., 2012 [32]
Lipizzan horses	0.086-0.1440	0.1160-0.1610	Zechner et al., 2002 [35]
Slovak horse breeds	0.0267-0.0626	0.0308-0.0934	Pjontek et al., 2012 [22]
Franches-Montagnes horse breed	0.0478-0.0521	0.1135-0.1205	Hasler et al., 2011 [14]
Native Danish horse breeds	0.0300-0.0600	–	Tirstrup et al., 2008 [28]

**Table 5**  
Effective number of founders ( $f_e$ ) and ancestors ( $f_a$ ) in different Equidae breeds

Rasa – Breed	$f_e$	$f_a$	Source
Paint Horses	560.8-963	186.1-214.8	Siderits et al., 2013 [25]
Arab horses	38.6-39.5	13-19	Cervantes et al., 2008 [7]
Catalonian Donkeys	70,6	27	Gutiérrez et al., 2005 [12]
Hanoverian horses	244.9	77.73	Hamann and Distl, 2008 [13]
Mallorqui horses	11-13	11-12	Álvarez et al., 2010 [1]
Norwegian Fjord horse breed	96	30	Bhatnagar et al., 2011 [3]
Belgian Draught horses	84-85	42.7-42.9	Janssens et al., 2010 [15]
Norwegian Døle and Nordland/Lyngen horses	14-48	7-12	Olsen et al., 2010 [20]
Asturcón ponies	17.8-19.5	12-13	Royo et al., 2007 [23]
Andalusian horses	39.6	16.5	Valera et al., 2005 [31]
Lusitano horses	27.5	11.7	Vicente et al., 2012 [32]
Lipizzan horses	39.3-55.8	12.5-18.8	Zechner et al., 2002 [35]
Slovak horse breeds	26-160	16-32	Pjontek et al., 2012 [22]
Franches-Montagnes horse breed	63	22	Hasler et al., 2011 [14]

Comparison of the results obtained in the present study on inbreeding and relatedness with literature data for the same and other breeds of horse reveals that the  $F_x$  values estimated in the present study are similar or in some cases much lower than those obtained by most of the authors cited in Table 4. In particular, comparison with the results obtained by American authors [21, 30] for the same breed is clearly favourable to the herd studied here, as both the inbreeding and relationship coefficients are lower for the horses from Roleski Ranch.

The values for the effective number of founders ( $f_e$ ) and effective number of ancestors ( $f_a$ ) obtained in the present study are also within the range of values given by other authors for various breeds of horse (Tab. 5) and thus far do not show substantial narrowing of the gene pool, as indicated by some authors who studied small horse populations of various breeds [1, 7, 12, 13, 26, 28, 32]. Petersen et al. [21], however, found that while the genetic variation of AQ horses in the United States is satisfactory, due to increasing specialization of breeding of these horses for particular western sport disciplines, recently a certain gene-

tic division of the entire population into smaller subpopulations has appeared, which was less common in the 1980s [30].

To sum up, the mean inbreeding and relationship coefficients were low despite the high percentage share of inbred and related horses in the herd. On the other hand, the  $f_e$  and  $f_a$  values obtained for the American Quarter horses from Roleski Ranch were relatively high in relation to the number of horses analysed. This indicates that the state of the gene pool of the herd is satisfactory and that well thought-out measures have been taken by breeders in purchasing sires and selecting them to be paired with broodmares. The presence of the stallion DOC BAR and his descendants in the pedigrees of the Roleski Ranch horses confirms that the breeding conducted in the herd is consistent with the western-style sport profile promoted by the facility, i.e. reining competitions.

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