

# On the origin of Bohemian cattle according to mitochondrial DNA

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**Abstract:** The genomic structure of extant cattle populations can contribute to the reconstruction of the history of particular breeds or their subpopulations. Genome-wide population resequencing of extant populations of Czech Red Pied (CRP) cattle, its conserved nucleus herd, and Czech Red (CR) cattle detected a T106C polymorphism in mitochondrial DNA shared by the conserved CRP herd with a geographical belt of Anatolian, Illyrian and Eastern Alpine breeds. On the other hand, this SNP is practically absent in the historical cattle breeds associated with Northern Germany, including German Black Pied cattle, Holstein-Friesian and German Red Mountain cattle (GRM). Correspondingly, this indicator SNP was absent in CR cattle, which, like the GRM, belongs to the group of mountain red cattle breeds. It seems that the precursory cattle population in Central Europe was influenced by the germplasm from the Balkans-related group of breeds, thanks to the political and commercial influence of the Northern Italian, Austrian and Hungarian regions. In addition, the presence of this polymorphism in some European aurochs (*Bos primigenius*) bone remains suggests possible introgression from local aurochs populations. Alternatively, the T106C presence in yaks and in neighbouring cattle breeds in Northern India, China and Korea raises the possibility of origin of this polymorphism from yak populations. The spread of the T106C mutation in alpine regions is consistent with the known role of this mtDNA region in adaptation to the reduced oxygen pressure.

**Keywords:** aurochs; Busha; Czech Red cattle; Czech Simmental; historical cattle

Cattle have played a decisive role in the economic development of Central Europe starting from the 6<sup>th</sup> millennium BC, and by at least the 4<sup>th</sup> millennium. They have also been used as traction animals (Sherratt 1983; Peske 1994; Kyselý 2012). The evo-

lution of the global cattle population can be broadly reconstructed by combining two approaches. The first one is derived from the population genetics of extant cattle populations since the extensive data on the current polymorphism enables restoring the

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starting structure backwards. On the other hand, archaeogeneticists concentrate on the recovery of the genetic information preserved in ancient DNA (aDNA) in archaeological bone finds. The archaeological findings can be interpreted in light of modern population genetics and *vice versa*.

Both approaches can solely rely on the mitochondrial DNA (mtDNA) structure. A reason for the preferential use of mtDNA in archaeogenetics is its abundance – about two orders of magnitude greater than that of chromosomal DNA, thus increasing the probability of its preservation. In cattle, the mtDNA variation is more conservative than the nuclear one due to the prevalence of bulls in long-distance trading. The role of mtDNA as a universal tool in tracking bovine history is documented by the key publication of a complete mitochondrial sequence of aurochs (*Bos primigenius*) based on an analysis of bone find CPC98 (humerus) from Carsington Pasture Cave in Derbyshire, England (Edwards et al. 2010).

On the other hand, advances in genomics now allow the use of whole genome data for the elucidation of domestic breed origin and evolution (Frantz et al. 2020). The publication of the cattle reference genome (Bovine Genome Sequencing and Analysis Consortium et al. 2009) was a milestone in the use of whole-genome sequencing (WGS) in cattle. If not prevented by commercial reasons or the restrictions of still unaccomplished projects, the breed-specific gene pools are available in public databases, e.g. on the server run by the University of Missouri (<http://bovinemine-v16.rnet.missouri.edu/bovinemine/begin.do>). In the case of archaeogenetics, the analogical sequencing of a complete genome of extinct aurochs (Park et al. 2015) also contributed to the fast advancement in this direction of research (Sinding and Gilbert 2016).

One of the groups of indigenous breeds in Central Europe is the so-called red mountain cattle, which were originally spread in higher altitude regions through Germany, the Czech Republic, Poland and Slovakia (Ludwig et al. 2016). The persisting populations are protected under genetic resource conservation programmes; however, all have experienced significant bottlenecks in the past, as a logical consequence of the spread of commercially efficient modern breeds. Purposeful conservation efforts for the Czech Red cattle (CR), a member of this group, can be dated back to 1921. In 1987, the current population of the CR cattle was restored from

only one bull (with 50% admixture) and 14 cows. Accordingly, the diversity of the CR cattle in total only reaches the effective size of 11 individuals (Czernekova et al. 2006). The current herd of the CR cattle consists of approx. 200 animals which are included in the Program of Conservation and Utilisation of Farm Animal Genetic Resources in the Czech Republic (CUFAGR; <https://vuzv.cz/en/genetic-resources/>).

Another typical member of the red mountain cattle cluster, the German Red Mountain (GRM) cattle or Rotes Höhenvieh, alternatively called as Höhenrotvieh, Harzer Rotvieh or Harz Mountain cattle, was created by merging several local forms from different mountain regions due to the reduction in numbers (Ludwig et al. 2016).

Salers cattle, which are characteristic of the Auvergne mountain region in France, are also often assigned to this group of breeds. The origin of mountain red breeds is sometimes associated with the Celtic Shorthorn, a breed of the Celts inhabiting Central Europe in the Late Iron Age (e.g. Tornede 1948).

Another autochthonous breed of the region is the modern Czech Red Pied cattle (CRP). Formally, this breed was created by imports of genuine Simmental cattle during the 19<sup>th</sup> century. However, the genetic background of CRP might still contain a significant proportion of gene variants from the preceding local cattle breeds. The original type of this breed has been conserved in the nucleus herd of approximately 80 animals (CRP-genetic resources or CRP-GR) in the Czech program of genetic resources since 2010. The gene pool of the CRP nucleus herd reflects the state at the end of the nineties of the 20<sup>th</sup> century.

The early genetic analyses clearly demonstrated that European domestic cattle do not represent a local domestication event or even multiple events, but that the entire European cattle population is of Near Eastern origin (Troy et al. 2001; Edwards et al. 2007). The ongoing debate is on the influence of native European wild cattle, i.e. the aurochs of the subspecies *B. primigenius primigenius*. This animal had been present in Central Europe since the Pleistocene and the region served as the last refuge of the species until the last individual died in 1627 in Jaktorow near Warsaw, Poland (Rokosz 1995). It must be taken into account that domestic and wild cattle coexisted here for approximately 7 000 years, during which time crossbreeding could take place

(for the Czech lands see Kysely and Meduna 2009; Kysely and Hajek 2012). Consequently, a low genetic contribution of European aurochs is still present in recent cattle breeds.

Both the morphometric evidence and mtDNA data support this model. Aurochs trait introgression was first reported mainly for historical breeds in Italy (Achilli et al. 2009). Moreover, surviving cattle lineages with aurochs mtDNA were reported in a recent study, namely in the alpine Murbodner breed in Austria (Cubric-Curik et al. 2021).

Another line of evidence for cattle-aurochs interbreeding is provided by archaeogenetic data. Morphologically domestic cattle carrying aurochs mtDNA was reported for the Bronze Age site in northern Spain (Anderung et al. 2005). Similarly, an animal from the Swiss Neolithic site of Twann, morphologically corresponding to a domestic cow, was shown to have aurochs DNA (Schibler et al. 2014).

A reciprocal case comes from a large animal found in the Early Medieval context of Vysehrad (Prague, Czech Republic). Although resembling the aurochs, an archaeogenetic analysis revealed the T3 (domestic) haplogroup of mtDNA, so a domestic-wild crossbred status of this animal was discussed (Kysely and Hajek 2012).

In order to elucidate the relation between modern cattle populations and potentially also aurochs in the Czech lands, the extant populations of indigenous cattle breeds were subjected to the sequencing of mitochondrial DNA. The rare polymorphism T106C found in combination with other informative polymorphisms, 221insC and 1601delA, indicated a connection between indigenous breed populations and current cattle populations in the Illyrian belt and to the presumed ancestors of domestic cattle.

## MATERIAL AND METHODS

### Samples from extant cattle populations

Thanks to the support of the breeding firm CHD Impuls (Bohdalec, Czech Republic) and the Association of Breeders of Czech Red Pied Cattle (Hradistko, Czech Republic), archived bull samples characterising full variability in the CRP breed were available. The current population was represented by a set of 164 bulls originating from the portfolio of CHD Impuls. Moreover, 35 blood samples of the

conserved nucleus herd of CRP (CRP-GR) were obtained from the archive of the CUFAGR program. Similarly, a set of 80 archived blood samples of the CR cattle were obtained from the same source.

### Resequencing of extant populations

**DNA isolation.** In the CRP commercial population, total DNA was prepared from cryopreserved insemination doses using affinity binding on paramagnetic particles with the MagSep Tissue Kit (Eppendorf, Hamburg, Germany). The archived blood samples were processed in the conserved genetic resources of CRP and CR cattle. One hundred microlitres of thawed blood were processed using silica membrane columns as part of the BloodPrep commercial kit (Life Technologies, Carlsbad, CA, USA). A normalised gDNA set containing 20 ng/ $\mu$ l DNA from each animal was prepared according to the concentrations determined spectrophotometrically and additionally purified with the AMPure XP magnetic bead procedure (Beckman Coulter, Brea, CA, USA).

**Pooling samples and WGS.** The conditions of the applied NGS methods have already been published elsewhere (Novak et al. 2019). Whole-genome sequencing was performed using HiSeq X Ten technology (Illumina, San Diego, CA, USA) in the Novogene UK Laboratory (London, UK). The obtained 60 $\times$  coverage was sufficient to detect polymorphisms above the 0.05 frequency at 95% efficiency.

**PoolSeq data processing and interpretation.** The read assemblies were built using the Geneious Mapper algorithm implemented in the Geneious program package (Biomatters, Auckland, New Zealand) and the UMD 3.1.1 bovine genome sequence ([https://www.ncbi.nlm.nih.gov/assembly/GCF\\_000003055.6/](https://www.ncbi.nlm.nih.gov/assembly/GCF_000003055.6/)) issued by the Centre for Bioinformatics and Computational Biology, University of Maryland.

## RESULTS

**Diversity in mtDNA of extant cattle in the Czech lands.** The alignment of the sequencing reads of the studied extant populations, i.e. of Czech Red Pied cattle, the conserved nucleus herd of this breed and of Czech Red cattle, to the reference genome sequence was accomplished in February 2018 and the preliminary interpretation was published by Novak et al. (2018, 2019). To illustrate this re-

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Table 1. Polymorphism detected in the pooled samples of three cattle populations using the HiSeq X-Ten sequencing

Position in the BRS mtDNA (V00654.1)	Change with respect to BRS	Frequency (%) in the population of			Polymorphism typical for <sup>1,2</sup>
		CRP	CRP-GR	CR	
106	T>C	–	16.90	–	recurrent mutation in the QT haplogroup, in the JQ437479 aurochs (Poland, from P), GU985279 aurochs (England, from P1), and in JS2
166	A>G	15.90	–	–	G to A at QT, at JQ437479 – Poland (from P) and GU985279 – England (from P1)
169	A>G	50.00	82.00	65.10	reversed at QT, present again in T3B and JS2 of P1A haplogroup
173	A>G	20.90	–	9.40	DQ124389 – Korea (ex P1a)
221	(C)6>(C)7	52.20	67.99	87.94	present in QT, Q, T, and P haplogroups (Polish and English aurochs JQ437479 and GU985279) except for some P1a lines of Japanese Shorthorn and the Korean aurochs; generally present in 43% of isolates reported by Cubric-Curik et al. (2021)
224	delC	43.20	–	0.20	–
300	G>A	16.70	8.10	8.20	P haplogroups
363	C>G	12.80	–	–	JQ437479 – Poland (from P) and GU985279 – England (from P1)
587	(C)5>(C)4	6.70	7.20	8.10	difference of all haplogroups from BRS NC_006853 (change after T3b)
1481	G>A	67.70	–	–	P haplogroups
1600	delA	80.00	57.50	49.00	generally present in 41% of isolates reported by Cubric-Curik et al. (2021)
2145	G>A	44.10	–	–	P haplogroups
2558	G>A	73.80	–	–	typical for QT, Q and P haplogroups
3188	TAT>CAC	19.40	–	–	only in JQ437479 aurochs – Poland (from the P haplogroup)
3238	T>C	27.80	–	–	Q + P haplogroups
3379	T>C	28.90	–	–	P haplogroups
3550	GT>AC	37.6→37.8	–	–	P haplogroups
3694	T>C	7.30	–	–	only in the JS9 and JS7 Korean cattle
5156	G>A	61.80	–	–	P haplogroups
5156	G>A	–	5.20	–	P haplogroups
5259	T>C	43.10	–	–	only in JS6 (the only member of the P1a haplogroup)
5500	GC>AT	43.4→43.8	–	–	in the QT, Q and P haplogroups
5743	TC>CT	14.00	–	–	PQT, P haplogroups
6451	T>C	5.20	–	–	in the JQ437479 aurochs – Poland (P haplogroup)
7604	T>C	25.30	–	–	reported for the JS1 Korean cattle only
7994	G>A	86.50	99.30	99.40	P haplogroups
10927	T>C	–	–	48.00	Q haplogroup only
10999	CCTA>TATT	43.2→43.9	–	–	in the PQT, Q, and P haplogroups

Table 1 to be continued

Position in the BRS mtDNA (V00654.1)	Change with respect to BRS	Frequency (%) in the population of			Polymorphism typical for <sup>1,2</sup>
		CRP	CRP-GR	CR	
11089	T>C	–	6.30	–	reported for Q only
12433	T>A	40.30	–	–	for Q, P haplogroups
12525	G>A	32.80	–	–	GU985279 – England (the only member of the P1 haplogroup)
13310	AT>CG	87.80	–	–	AT is characteristic for BRS V00654.1 compared to the BRS NC_006853
16058	C>T	–	–	20.00	in P haplogroups
16074	T>C	28.60	–	–	in P haplogroups
16119	T>C	–	–	47.10	reported for JS6 (ex P1a)
16122	T>C	12.10	–	–	P haplogroups
16231	C>T	16.70	–	–	P haplogroups
16231	C>T	–	–	22.00	in the PQT + P haplogroups
16255	T>C	15.10	–	–	occurs in non-T3, non-T3B haplogroups
16301	C>T	8.50	26.90	–	GU985279 – England (the only member of the P1 haplogroup)

<sup>1</sup>Sequences of the *Bos primigenius* finds are coded by DQ124389 for the aurochs from Korea (Achilli et al. 2008), GU985279 for the aurochs from England (Edwards et al. 2010), and JQ437479 for the aurochs from Poland (Zeyland et al. 2013);

<sup>2</sup>Animals coded by the JS letter series originate from the indigenous Japanese Shorthorn cattle as specified by Mannen et al. (2020)

CR = Czech Red cattle; CRP = Czech Red Pied cattle; CRP-GR = genetic resources subpopulation of Czech Red Pied cattle

sult, approximately 12.5 million single nucleotide polymorphisms distinguished the CR breed from the genomic reference sequence of the Hereford breed animal. However, it should be noted that the reference genome UMD\_3.1.1 was derived from an animal belonging to the Hereford breed, which is genetically quite distant from Central European historical breeds (Medugorac et al. 2009).

It should be taken into account that bovine mitochondrial DNA (reference NC\_006853 as a part of the UMD3.1.1 assembly of the bovine genome) is circular and comprises 16 338 bp. Nevertheless, the traditional reference sequence for the studies of mtDNA polymorphism in cattle is V006541, which is also used for the presentation of the found polymorphism below.

The variants differing from the traditional reference sequence of bovine mtDNA (V00654.1) are summarised in Table 1. For their classification, the diagnostic SNPs for the generally accepted mtDNA haplogroups (Mannen et al. 2020) are included, as well as known polymorphisms in the aurochs findings reported in literature (Achilli et al. 2008; Edwards et al. 2010; Zeyland et al. 2013).

It can be assumed that most of the animals in the CRP, CRP-GR and CR populations belong to the T3 haplogroup. This is suggested by the match of the corresponding markers at positions 169, 221, 587, 2 558, 13 310 and 16 255 (Achilli et al. 2009). Since they correspond to the reference sequence V00654.1, they are not always visible in Table 1, which summarises only the differences from this reference.

Surprisingly, all three studied populations also contained markers (Table 1) that were reported to be characteristic of the haplogroups Q and QT, which are the ancestors of the modern T3 haplogroup, and of the cluster of P haplogroups, which cover aurochs and sporadic breeds of cattle documenting intercrossing after the initial domestication (Mannen et al. 2020). This set comprising 33 minor markers was mostly concentrated in the production population of Czech Red Pied cattle (not shown). Their occurrence should be ascribed either to a wrong assignment of these markers to the Asian cattle populations or to a wider basis of diversity in the contemporary group of the Simmental breeds, which are bred on a large scale.

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The markers restricted to the historical populations of CRP-GR and CR represent more interest (Table 1). In both historical breeds, the markers A169G and T10927C (specific to Q haplogroup) are present. The CRP-GR specific markers comprise T106C, G5156A, T11089C (Q specific) and C16301T, while the CR harbours A1736G, T10927C (Q specific), C16058T, T16119C and C16231T. Notably, both historical populations bear markers described in aurochs to date, i.e. T106C and C16301T in CRP-GR and A173G in the CR cattle.

## DISCUSSION

The data on mitochondrial DNA from modern populations of indigenous cattle breeds in the Czech Republic can be interpreted in light of the knowledge of other extant breeds, archaeogenetic reports and wild cattle relatives to elucidate the origin and structure of historical cattle in the Czech lands.

**Implications derived from the sequencing of Czech indigenous breed populations.** The current DNA polymorphism in cattle populations can mainly be viewed in light of the geographical movement of animals. This might be a consequence of long-distance trade, and also a consequence of the migration of human populations bringing their livestock with them. The changes in livestock structure might also reflect military invasions (Upadhyay et al. 2017).

The significant occurrence of the polymorphism T106C in the CRP-GR subpopulation at 16.1% frequency enabled distinguishing the CRP-GR subpopulation from the contemporary production herd of the CRP cattle, as well as from the CR cattle. This links the historical basis of this breed to the area of historical breeds that are carriers of this SNP (Table 1), crossing Europe from the Southeast to the Central regions. It should be noted that this interpretation is possible due to the accomplished targeted resequencing of mtDNA in a range of extant cattle populations in Europe in a survey performed by Cubric-Curik et al. (2021) and Neumann et al. (2023). The 106C allele was reported for individual representatives of the European breeds included in the survey, where the traditional haplogroup and the number of nucleotides at the sites 221insC and 1600delA are indicated in brackets: Anatolian Black (T3, 6C, 6A), Katerini (Q, 6C, 7A),

Greek Shorthorn Pindos (Q, 6C, 7A; T3, 6C, 7A), Lekhiboj Busa (T3, 7C, 7A), Sharri Busha (T3, 7C, 8A), Croatian Busha (T1, 6C, 7A; 2 × T3, 6C, 6A; 3 × T3, 6C, 7A; T3, 7C, 6A; T3, 7C, 7A), Boskarin (T3, 6C, 7A), Murbodner (T3, 6C, 6A; T3, 6C, 7A), and Tux-Zillertal cattle (T3, 6C, 7A). Their regions of origin form a belt along the coast of the Illyrian region. This area starts from the original area of Anatolian Black cattle in Central Anatolia and includes the areas of conserved historical breeds of Southern Greece, Albania, Monte Negro and Croatia, and further the area of the Eastern Alps breeds Tux-Zillerthaler and Murbodner.

This is consistent with the previously reported relatedness of the Balkan breeds, namely, the various subgroups of Busha cattle, with a group of eastern Alpine breeds, such as Tyrolean Grauvieh, Original Braunvieh of Murnau-Werdenfelser cluster (Medugorac et al. 2009). There is one exception: the Murbodner breed was found to be surprisingly related to the geographically distant Franken Gelbvieh. This fact can be ascribed to the efforts to improve the milk yield of the Murbodner breed after the Second World War through crossbreeding with the more commercially exploited Franken Gelbvieh (Medugorac et al. 2009).

The general occurrence of T106C in the Illyrian group of breeds contrasts to the absence of T106C in the more northwestern local breeds, like GRM cattle (T3 haplogroup) (Ludwig et al. 2016) and German Black-Pied (DSN) (Neumann et al. 2023). Consistently, in the Holstein breed derived from the DSN breed, T106C appeared only sporadically, at the total frequency of 1.5% in the background of T3 haplotypes (2 × T3, 6C, 6A; T3, 6C, 7A; T3, 7C, 7A) (Cubric-Curik et al. 2021).

As indicated in Table 1, T106C has also been reported for Korean cattle in the framework of the P haplogroup (Mannen et al. 2020), and in some extinct aurochs, namely JQ437479 (Poland, P haplogroup) and GU985279 (England, P1 haplogroup), and also in yak (Liu et al. 2023). T106C has also been reported in some Northern Indian breeds and some breeds from the north of China (Liu et al. 2023).

The occurrence of this mtDNA marker only in the conserved subpopulation of the Czech Red Pied breed should obviously be ascribed to the timely inclusion of the nucleus herd into the CUFAGR program. The CRP might originate from the introgressive crossing of the original local cattle breed

with the imported highly productive Simmental breed. This fact suggests that a part of the starting genetic information was contained in the gene pool of the CRP population until recently.

However, T106C is also absent in the German Black Pied (DSN) cattle, an endangered historical breed (Neumann et al. 2023). Its initial farming dates back to the 18<sup>th</sup> century in the North Sea region of Germany and the Netherlands (as Dutch Friesian). This breed also gave rise to the modern Holstein breed. Consequently, the potential spread of cattle maternal genotypes along the Elbe River valley from Northern Germany to the Bohemian Basin and *vice versa* is not supported. Consistently, almost no presence of the SNP was found in Holstein cattle from Northern Germany (below 1.5% according to Cubric-Curik et al. 2021). No T106C has been reported for the autochthonous GRM cattle (Ludwig et al. 2016). The combination of T106C with the neighbouring markers 221insC and 1600delA, which includes the T106C marker in the CRP-GR population, appears in the Holstein population at a frequency below 0.5%. Consistently, we could not detect it in the historical breed of Czech Red cattle, which is considered to be also a member of the group of mountain red breeds.

It should be taken into account that the genetic background of CRP cattle (of the Simmental group) has been affected by intercrossing with the modern Holstein-Friesian population (Citek et al. 2006; Czernekova et al. 2006). Nevertheless, since the cows in this improvement hybridisation mostly belonged to the CRP breed, the effect on the mtDNA structure was far less pronounced than the effect on chromosomal genes, reported in the cited works.

**Potential historical factors leading to the observed polymorphism.** Consequently, the distribution of T106C in the Illyrian belt of breeds suggests that the persistence of the marker in the extant CRP-GR population might be assigned to the political and commercial influence of the North-Italian and Austrian regions in the Czech lands during the Middle Ages.

The cattle spread into Europe via the Aegean Sea around the mid-7<sup>th</sup> millennium BCE (Scheu et al. 2015). Further spread followed two routes as part of the so called Neolithic Package. One of the routes ran along the Mediterranean coast, while the other branch advanced via the Balkans and Danube valley. A newly reported T6 haplogroup is shared by the present-day Rhodope Shorthorn cattle on one

side and the local archaeological samples on the other. This fact confirms the antiquity of Balkan shorthorn cattle (Hristov et al. 2018). In the region of Bohemia (the upper Elbe basin), domestic cattle have been well documented since the beginning of the Neolithic, i.e. since about 7700 BP onwards (Peske 1994; Kysely 2012).

It should be distinguished whether the currently observed pattern of the T106C polymorphism can be ascribed to this ancient primary process of the cattle husbandry spread or whether it is a consequence of secondary migrations and long-distance trade in the Middle Ages. Intense trade with cattle in the Czech lands must be assumed with respect to Austria, due to the proximity, good state of commercial trails and the periods of common central ruling, e.g. by Ottokar II (1261–1278). Additional testing of the available archaeological bone finds from the region could provide a more precise picture.

**Relations to the aurochs and yaks.** The critical SNP T106C in mtDNA suggests not only the putative relations between the medieval and extant cattle populations in the Czech lands, but also the links to wild cattle, aurochs. The origin of T106C in cattle can be associated with intercrossing with local aurochs populations in Europe, occurring after the initial spread from the Near East centre of domestication. The presence of the informative mtDNA SNP T106C has been reported for the CPC98 aurochs sample from Derbyshire (Edwards et al. 2010).

On the other hand, the presence of this polymorphism in yaks suggests intercrossing with yaks at the early stages of cattle evolution in Central Asia (Liu et al. 2023). Direct evidence based on ancient mtDNA from archaeological finds goes back as far as 2 500 years ago (Chen et al. 2023). This hypothesis is supported by sharing the polymorphism of yaks in the neighbouring extant cattle populations, including the breeds from Northern India (Liu et al. 2023). In China, T106C was reported to be present in historical breeds close to the area of wild yaks, which are known to intercross with domestic cattle (Wang et al. 2016). The reports of the T106C presence in historical Japanese breeds (Mannen et al. 2020) can be explained by the Chinese origin of these breeds.

The above findings support the origin of this mutation in the populations of yaks in the Tibet region. The selective advantage of the T106C

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polymorphism and subsequent fixation in cattle populations can be due to the presumed adaptation to mountain conditions. A parallel evolution in this mtDNA region (the RS2 region) has been demonstrated in a number of species of the tribe Caprini that underwent adaptation to higher altitudes (Hassanin et al. 2009).

**Position of Czech Red cattle.** The traditional opinion on cattle evolution in the Czech lands ascribed to the Czech Red cattle the role of the local cattle breed that resulted from the historical adaptation process (Citek et al. 1997). If this is the case, the gene pool of this cattle breed would be a unique source of information about the history of the cattle population in the Elbe and Morava river basins.

However, the current population of this breed has not been confirmed as a carrier of the indicator mtDNA variant T106C. Interpreting this result, it should be taken into account that the CR population underwent at least two bottlenecks; the first one in the early 1920s, when the breed was saved by Frantisek Bilek, head of the research station preceding the current Institute of Animal Science, and in the mid-eighties of the 20<sup>th</sup> century (Citek et al. 1997; Czernekova et al. 2006). Moreover, the CR breed is generally known for containing admixtures from a number of breeds used for its recovery, modernisation and for the improvement of economic traits. The breeds forming the admixture include Polish Red, Angler, GRM cattle, Czech Simmental cattle, Ayrshire, Red Holstein and Piedmont cattle (Citek et al. 2006; Czernekova et al. 2006). Nevertheless, the T106C absence does not support the historical continuity of this breed to the local medieval cattle, nor to the morphologically corresponding Balkan breeds like Busha or Cika, but it rather indicates the relatedness to the northwestern group of mountain breeds like the GRM cattle (Ludwig et al. 2016). The existence of the assumed group of breeds with the original area spanning from the Balkans to Central Europe is partly supported by the fact that most of the local breeds from Bohemia to the Baltic countries and even Ukraine belong to the Y chromosome Y1 haplogroup (Edwards et al. 2011).

## CONCLUSION

The resequencing of the mtDNA of indigenous cattle breeds of the Czech lands indicated the influence of the Illyrian breed belt on the assumed

historical populations, mostly based on the T106C polymorphism, but this evidence was only partially preserved in the current CRP cattle, subsequently crossbred with the imported Simmental breed. This model is expected to be supported by independent processing of archaeogenetic findings from the past, ideally the medieval period.

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## Conflict of interest

The authors declare no conflict of interest.

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