

РЕЗЮМЕТА НА НАУЧНИТЕ ТРУДОВЕ

НА ГЛ. АС. Д-Р СТАНИСЛАВА ПЕЙЧЕВА ПЕЕВА

Представени за участие в конкурс за заемане на академичната длъжност „Доцент“ по научна специалност „Специални отрасли (промишлен дивеч)“ област на Висше образование 6. Аграрни науки и ветеринарна медицина, професионално направление 6.3 Животновъдство,

1. Hisano M., E. Raichev, S. Peeva, D. Georgiev, H. Tsunoda, R. Masuda, Y. Kaneko, 2014. Notes on autumn-winter stomach contents of the stone marten (*Martes foina*) in the Balkan mountains, Central Bulgaria. *Zoonotes*, 56: 1-6.

Abstract. We studied autumn-winter food (November-February, 1997-2003) of the Stone Marten in the Balkan Mountains, Central Bulgaria. A total of 26 stomachs (18 of males and eight of females) were examined, of which contents was expressed as the number of occurrence and relative frequency of occurrence. The result clearly showed that rodents are primary prey for the Stone Marten. Birds and insects were the following categories consumed. The edible dormouse was detected as a prey for the first time in Bulgaria. Besides, wild ungulates and domestic animals were occasionally scavenged, while neither fruits nor artificial materials were detected in the marten stomachs. Thus, our study showed that the Stone Marten in the Balkan Mountains tended to be more carnivorous in winter.

2. Mizumachi K., Y. Nishita, N. Spassov, E. Raichev, S. Peeva, Y. Kaneko, R. Masuda, 2017. Molecular phylogenetic status of the Bulgarian marbled polecat (*Vormela peregusna*, *Mustelidae*, *Carnivora*), revealed by Y chromosomal genes and mitochondrial DNA sequences, *Biochemical Systematics and Ecology*, 70, pp. 99-107 **Q3**; **SJR= 0.31**; **IF=0.847**

Abstract. In this study, we investigated molecular phylogenetic status of the marbled polecat (*Vormela peregusna*) from Bulgaria, using sequences of two Y-chromosomal genes (SRY and ZFY). The phylogenetic tree inferred using combined sequences of both genes indicated that the marbled polecat was split from genera *Lutra*, *Neovison* and *Mustela* after genus *Martes* was diverged in family *Mustelidae*. In addition, we analyzed molecular phylogeography of the Bulgarian population of the marbled polecat, using cytochrome b and control region sequences of mitochondrial DNA (mtDNA). The phylogenetic tree of cytochrome b indicated that the haplotypes of the Bulgarian population comprised two haplogroups, which were the most ancestral clades. Additionally, the control region phylogeny showed that the haplotypes of Bulgaria formed two haplogroups: one was the most ancestral clade, and the other was the

derivative clade. One individual with the most ancestral cytochrome b clade had a control region haplotype of the derivative clade. Thus, this study revealed that the most ancestral lineages of the marbled polecat are included in the population of Bulgaria. The Bulgarian population could be a remnant lineage from a basal for the species, which in Pleistocene occupied a relatively large area related to the Balkan-Caucasian.

3. Raichev E., S. Peeva, R. Masuda, Y. Kaneko, H. Tsunoda, D. Georgiev, D. Georgiev, 2017. Sexual dimorphism in body parameters of the golden jackal *Canis aureus* L., 1758 (*Carnivora, Canidae*) in the Sarnena Sredna Gora Mountain and Thracian plain (Bulgaria). *Trakia Journal of Sciences*, № 2, pp. 135-140.

Abstract. The study was conducted in the area of the Sarnena Sredna Gora Mountain and the Thracian Plain in period 1996-2014. On a total of 262 golden jackals (*Canis aureus* L., 1758) (119 males and 143 females) thirteen somatometric parameters were measured. The comparison of the linear body parameters and the weights between males and females showed apparent sexual dimorphism in the jackals with a high level of reliability, with an exception of the length of the tail. The index of body compactness and the weight index were calculated and compared. The index of body compactness did not differ between sexes. The body weight (10,994.24 g for males and 9,776.02 g for females in average) showed clear sexual dimorphism – male-female ratio was 11.08%. Our findings indicated that the sexual size dimorphism in golden jackal was weaker and lower than those in red fox and wolf.

4. Peeva S., E. Raichev, D. Georgiev, A. Stefanov, 2017. Influence of elevated platform (wire-mesh or wooden) in the cage on domestic rabbit (*Oryctolagus cuniculus*) activity. *Agricultural science and technology*, Vol. 9, № 3, pp. 257-258, DOI: 10.15547/ast.2017.03.048

Abstract. Placing an elevated platform in the cage of rabbits is an option to increase its area. The aim of this study was to determine whether raised platform affected the overall activity of breeding rabbits and to evaluate the influence of used material (wire-mesh or wood). Two experimental groups of three male Californian rabbits at four months of age, equal body mass and condition were formed. Their activity was examined in three cage types: without platforms, with wire-mesh platforms and with wooden platforms using camera traps. Separation of cage volume by means of an elevated platform increased the activity of breeding rabbits. The rabbit activity was also influenced by the platform material. It was almost three times higher when the platform was made from wood than from wire-mesh.

5. Peeva S., 2017. On the sex and age structure of the Stone Marten (*Martes foina*) population from Sarnena Sredna Gora Mts. (Central Bulgaria). *ZooNotes*, 117: 1-3.

Abstract. The sex and age structure of the Stone Marten (*Martes foina* Erxleben, 1777) population from Sarnena Sredna Gora Mts. (Central Bulgaria) was studied. A total of 67 skulls were divided into three age groups using different methods. The population structure showed a male-biased sex ratio with adults predominating over yearlings.

6. Peeva S., E. Raichev, G. Zhelyazkov, 2017. Fish producer's attitude to the most common fish-eating birds in Central Bulgaria. *Ecologia Balkanica*, vol. 9, issue 2, 1-5

Abstract. In Bulgaria part of fish farming is through using extensive production technologies. Most of the dams used for fish production are located in the lowlands of the country and are the natural habitats of herons, cormorants and pelicans. Thus these birds are considered to be pests in extensive aqua production. To clarify whether in fact the owners and workers in fish farms obey the law with regard to fish-eating birds, an anonymous survey among 80 fish producers was conducted between January and August 2014. The positive and negative responses were expressed as a percentage. The economic factor determined the negative attitude of owners towards fish-eating birds. The lack of motivation for conservation of protected bird species was due to non-payment of compensations from the government.

7. Peeva S., A. Mikov, D. Georgiev, 2018. On the Arthropods in the Stone Marten's (*Martes foina*) diet in Central Bulgaria. *ZooNotes*, 121: 1-3.

Abstract. Fourteen newly found Arthropods taxa in the Stone Marten's diet in Central Bulgaria were reported. Five of them were defined to species level. The study was carried out on the base of collected fecal samples.

8. Georgiev D., E. Raichev, L. Dospataliev, S. Kalcheva, K. Georgieva, M. Ivanova, S. Peeva, 2018. Heavy metals concentration in organs of red foxes (*Vulpes vulpes* Linnaeus, 1758) and golden jackals (*Canis aureus* Linnaeus, 1758) inhabiting the Sarnena Sredna gora mountain in Bulgaria. *Bulgarian Journal of Agricultural Science*, 24 (supplement 1), 119-124 Q3; SJR=0.223

Abstract. The purpose of the study is to determine the concentrations of the heavy metals lead (Pb), cadmium (Cd), nickel (Ni), cobalt (Co), copper (Cu) and zinc (Zn) in the muscles, kidneys and liver samples taken from red foxes (n = 9) and jackals (n = 17) inhabiting the "Sarnena Sredna gora" mountain in Bulgaria and to compare the respective results. It was established that concentrations of heavy metals (mg/kg dry weight) in the muscles, kidneys and liver samples are

as follows: in the red foxes' muscles: Pb – 0.170, Cd – 0.089, Ni – 2.312, Co – 0.074, Cu – 2.565, Zn – 41.382; in the red foxes' kidneys: Pb – 0.761, Cd – 15.522, Ni – 2.834, Co – 2.033, Cu – 8.470, Zn – 30.157; in the red foxes' livers: Pb – 0.953, Cd – 0.628, Ni – 2.480, Co – 1.664, Cu – 15.121, Zn – 30.106; in the jackal's muscles: Pb – 1.126, Cd – 1.742, Ni – 7.416, Co – 1.798, Cu – 6.466, Zn – 66.804; in the jackal's kidneys: Pb – 8.419, Cd – 11.185, Ni – 7.710, Co – 5.039, Cu – 33.861, Zn – 61.576; in the jackal's livers: Pb – 8.879, Cd – 11.569, Ni – 7.342, Co – 4.963, Cu – 56.319, Zn – 63.618. The red fox (*Vulpes vulpes* Linnaeus, 1758) and the jackal (*Canis aureus* Linnaeus, 1758), could be used as an effective biomarker for the assessment of the environmental pollution in their natural habitats.

9. Ito K., E. Raichev, S. Peeva, H. Tsunoda, Y. Kaneko, 2018. Fauna survey methods of rural area in central Bulgaria: comparison between camera traps and field signs. *J. Field Science*, 16: 23-30.

Abstract. There were no previous researches using camera traps in Bulgaria for determining regional mammal fauna, though researches using the techniques have been rapidly increased currently. We investigated regional mammal fauna using camera traps in a rural landscape in central Bulgaria. Moreover, we compared the result of the camera-trapping investigation to the checklist of regional mammal fauna investigated by animal signs (footprints, roars and/or direct observations) for past five years, to assess utility of the techniques in the region. We detected a total of ten mammal species (six Carnivora, two Cetartiodactyla, one Lagomorpha and Eulipotyphla species, respectively) at eight camera stations for 35 days between 10 June and 14 July in 2015 (total of 173 camera days). On the other hand, there was a total of 15 mammal species recorded in the regional checklist: thus, we found 67 % of the regional species by the camera traps, whereas we cannot detect five species, i.e., least weasel (*Mustela nivalis*), European polecat (*Mustela putorius*), Eurasian otter (*Lutra lutra*), European wildcat (*Felis silvestris silvestris*) and wild boar (*Sus scrofa*). Moreover, we noted daily activity patterns in three frequently-occurred carnivores (golden jackal, *Canis aureus*, European badger, *Meles meles* and stone marten, *Martes foina*) observed in the camera-trapping surveys. Finally, we discussed the methodological problems for camera traps in faunal researches.

10. Tsunoda H., K. Ito, S. Peeva, E. Raichev, Y. Kaneko, 2018. Spatial and temporal separation between the golden jackal and three sympatric carnivores in a human-modified landscape in central Bulgaria. *Zoology and Ecology*, doi: 10.1080/21658005.2018.1504406 Q4; SJR=0.24

Abstract. The range of the golden jackal (*Canis aureus*) in Europe has expanded from southern regions northward and westward, raising concerns of increased competitive interactions with other carnivores. In Europe, the jackal is most common in Bulgaria, where it co-occurs with

several other carnivore species. We investigated the spatial occurrence and daily activities of golden jackals and three smaller sympatric carnivores: the red fox (*Vulpes vulpes*), the European badger (*Meles meles*) and the stone marten (*Martes foina*). Using camera trapping in spring and summer in a human-modified landscape of central Bulgaria, we ascertained that red foxes were separated from jackals spatially, whereas badgers and martens were active at different times of the day. We suggest that differences in resource partitioning between jackals and the three smaller carnivore species were associated with a variation in resource use patterns (e.g., food or microhabitats). Our findings indicate that spatial/temporal separation allows smaller species to avoid direct confrontations and agonistic competitions with jackals, resulting in successful co-occurrence.

11. Peeva S., E. Raichev, N. Tsandev, 2018. Reliability of determining Stone Marten's (*Martes foina*) age by two different skull features. *Trakia Journal of Science*, №4, 344-347.

Abstract. Based on skulls from Central Bulgaria, the possibilities of aging stone martens were considered and compared. Two different skull morphological criteria methods were compared to more precise method – counting of annuli in dentin. Precise aging of stone martens was only possible by counting of annuli in dentin. It is not recommended using methods by development of sagittal crest and maxillary teeth attrition separately for aging stone martens. A combination of several methods is considered to be reliable.

12. Peeva S., 2018. On the activity of Edible dormouse (*Glis glis* Linnaeus, 1766) in the central part of Stara Planina Mts (Bulgaria). *Ecologia Balkanica*, 10(2): 255-258. Q4 SJR 2018= 0.13

Abstract. Both, the daily and seasonal activity of Edible dormouse (*Glis glis* Linnaeus, 1766) in the region of National Park “Bulgarka” (Stara Planina Mts, Bulgaria) were studied using camera traps. The results showed nonstop full night activity of this species in the region (1266 m a.s.l.). It became active in the beginning of May and minimized its movements above the ground in the end of October. Short decrease period of its activity was observed during August.

13. Zhelyazkov G., D. Georgiev, S. Peeva, S. Kalcheva, K. Georgieva, 2018. Chemical composition and levels of heavy metals in fish meat of the Cyprinidae family from Zhrebchevo Dam, Central Bulgaria. *Ecologia Balkanica*, 10(2): 133-140. Q4 SJR 2018= 0.13

Abstract. The aim of the study was to determine the chemical composition and for the first time – the content of cadmium (Cd), nickel (Ni), lead (Pb) and zinc (Zn) in the meat of bream

(*Abramis brama* Linnaeus, 1758), Prussian carp (*Carassius gibelio* Bloch, 1782), vimba (*Vimba vimba* Linnaeus, 1758) and chub (*Leuciscus cephalus* Linnaeus, 1758) in one of the most popular for angling dams in Central Bulgaria in order to evaluate the risk for human health. Chemical composition of fish from Zhrebchevo dam showed the highest moisture content in the meat of Prussian carp – $82.13 \pm 0.04\%$, and the lowest in that of vimba – $78.51 \pm 0.07\%$. The amount of protein and lipids was highest in vimba meat $18.41 \pm 0.23\%$ and $1.88 \pm 0.09\%$, and lowest in the Prussian carp - $16.49 \pm 0.01\%$ and $0.25 \pm 0.05\%$. The content of Cd, Ni and Pb in the meat of vimba was the lowest - 0.033 ± 0.001 mg.kg-1, 0.330 ± 0.001 mg.kg-1 and 0.135 ± 0.001 mg.kg-1 and the highest in bream, 0.045 ± 0.004 mg.kg-1, 0.457 ± 0.018 mg.kg-1 and 0.177 ± 0.013 mg.kg-1 respectively. The highest content of Zn was observed in the Prussian carp - 20.170 ± 0.266 mg.kg-1, and lowest in bream - 15.903 ± 0.593 mg.kg-1. The obtained values for the four studied heavy metals were below the maximum permissible level according to the European legislation.

14. Nishita Y., N. Spassov, S. Peeva, E. Raichev, Y. Kaneko, R. Masuda, 2019. Genetic diversity of MHC class II DRB alleles in the Marbled polecat, *Vormela peregusna*, in Bulgaria. *Ethology, Ecology and Evolution*, 31(1): 59-72 doi.org/10.1080/03949370.2018.1486887 Q,3; SJR= 0.47; IF=1.038

Abstract. The marbled polecat, *Vormela peregusna*, is a small marten-like mustelid distributed from southeastern Europe to western China. Since the individual numbers of this species have declined, it has been classified as a vulnerable (VU) species in the 2008 IUCN Red List. To better understand the adaptation of *V. peregusna*, we investigated diversity and selection in the major histocompatibility complex (MHC) class II DRB gene which is reported to be highly polymorphic in many other species. Among 10 *V. peregusna* individuals from Bulgaria, we detected 9 novel sequences of partial exon 2, including antigen-binding site (ABS) codons, as representatives of DRB allele (*Vope*-DRBs). Two of the alleles were detected in all individuals analysed, while the others showed limited geographical distributions. Our results provided evidence for positive selection on ABS codons and recombination break points, contributing to maintaining the diversity of *Vope*-DRBs. In a Bayesian phylogenetic tree, all *Vope*-DRBs grouped within the mustelid clade, with three of them in the basal group consisted of alleles from *Meles*, *Martes*, and *Vormela* but no *Mustela* alleles, and the others in the derived group consisted of alleles diverged relatively late in the various mustelid species. The *Vope*-DRBs showed trans-species polymorphism within the mustelid clade, suggesting that the DRB genes evolved under long-lasting balancing selection.

15. Tsunoda H., S. Peeva, E. Raichev, K. Ito, Y. Kaneko, 2019. Autumn dietary overlaps among three sympatric mesocarnivores in the central part of Stara Planina mountain, Bulgaria. *Mammal study*, 44(4): 1-7. <https://doi.org/10.3106/ms2018-0068> Q2; SJR 2018 = 0.28, IF = 0.581

Abstract. Food habits and dietary overlaps of the three sympatric mesocarnivores (golden jackal *Canis aureus*, red fox *Vulpes vulpes*, and stone marten *Martes foina*) in a mountain-forest region in central Bulgaria were investigated. These species showed high dietary overlaps, commonly consuming rodents and fruits through studied period. Moreover, their dietary overlaps were higher in November than in the earlier months, because rodents were a predominant prey for all carnivore species in this season. Spatiotemporal separations of smaller carnivores from larger competitor may enable their sympatry, when their food habits were similar.

16. Peeva S., 2019. Sexual size dimorphism in Stone marten (*Martes foina*, Erxl. 1777) from Sarnena Sredna gora Mts (Bulgaria). *Trakia journal of sciences*, 17(4): 318-322.

Abstract. Sexual dimorphism in body size measurements of 65 stone martens was investigated in winter periods (10.12.- 01.03) 2013 to 2017. There was found sexual dimorphism in body size of Stone Marten from Central Bulgaria: males were larger than females. The tail length (with or without hairs) did not depend on sex. The sexual dimorphism in the most variable linear parameters of the species varied from 2.76% to 10.63%. Male stone martens were 23.37% heavier than females.

17. Mizumachi K., N. Spassov, D. Kostov, E. Raichev, S. Peeva, D. Hirata, Y. Nishita, Y. Kaneko, R. Masuda, 2020. Mitochondrial haplogrouping of the ancient brown bears (*Ursus arctos*) in Bulgaria, revealed by the APLP method. *Mammal Research*, 65(2): 413-421. IF=1.29 Q1 SJR 2019= 0.63

Abstract. In order to investigate the detailed zoogeographical history of brown bears (*Ursus arctos*) in Bulgaria and their relationships with populations in neighboring regions of Europe and Asia, the amplified product length polymorphism (APLP) method for the mitochondrial DNA (mtDNA) haplogrouping was applied to ancient bone remains. The results showed that haplogroups for 12 of 31 samples (38.7%) were successfully classified using the APLP method, although partial sequences of the mtDNA control region were determined with PCR product-direct sequencing for only 6 samples (19.4%). Even among 25 samples, of which nucleotide sequences could not be determined, the APLP method successfully classified haplogroups of 6 samples (6/25, 24.0%), indicating an advantage of the APLP method. In Bulgaria, although both the Balkan/Italian lineage of mtDNA (clade 1b) and the Eastern European lineage (clade 3a1) have been identified from modern bears as reported in other studies, all bear remains examined in the present study had APLP haplogroup W. The mtDNA phylogenetic analysis showed that

the ancient Bulgarian brown bears had clade 1b. This indicates that clade 1b was originally distributed in Bulgaria, whereas those with clade 3a1 could have entered from Romania to Bulgaria. In addition, the APLP and phylogenetic analysis of recent skin samples from Turkey showed that they have mtDNA of the Middle Eastern/Turkish lineage (referred to clade 7a). Therefore, Bulgaria on the Balkan Peninsula might have been located between two distribution borders: one is between clades 1b and 3a1, and the other is between clades 1b and 7.

18. Tsunoda H. C. Newman, S. Peeva, E. Raichev, C. Buesching, Y. Kaneko, 2020. Spatio-temporal partitioning facilitates mesocarnivore sympatry in the Stara Planina Mountains, Bulgaria. *Zoology*, 141(2020)125801. IF=1.77 Q1 SJR 2019= 0.87

Abstract. The top trophic level in many terrestrial food webs is typically occupied by mammalian carnivores (Order Carnivora) that broadly affect and shape ecosystems through trophic cascades. Their inter-specific interactions can further complicate effects on community dynamics as a consequence of intra-guild competition. The capacity for competitive mammalian carnivores to segregate their hunting and activity regimes is in major part a function of their similarity, in terms of body-size and dietary niche; termed the ‘niche variation hypothesis’. In this study, we used camera-trapping to investigate intra-guild interactions and spatio-temporal partitioning among five mesocarnivores, the golden jackal (*Canis aureus*), European badger (*Meles meles*), red fox (*Vulpes vulpes*), European wildcat (*Felis sylvestris*) and stone marten (*Martes foina*), in the Stara Planina Mountains, Bulgaria. We collected a total of 444 images of golden jackals, 236 images of European badgers, 200 images of red foxes, 171 images of stone martens, and 145 images of European wildcats, from 6612 camera-days across fifteen camera trapping stations. With respect to body size, the three smaller species (fox, wildcat and marten) were active in different time periods than the two larger competitors (jackal and badger) through both the warm and cold season. The more similar the trophic niche between species pairs (particularly relating to rodent consumption), the greater the spatio-temporal partitioning we observed within the pair; however, this adapted to seasonal dietary shifts. In conclusion, spatial and temporal (fine-scale and seasonal) niche partitioning appeared to reduce encounter probabilities and competition and may act to facilitate sympatric coexistence among this regional mesocarnivore guild.

19. Raichev E., Peeva S., Kirilov K., Kaneko Y., Tsunoda H. 2020. Autumn-winter Dietary Adaptability of the Golden Jackal *Canis aureus* L., 1758(Mammalia: Carnivora) with Respect to Type and Intensity of Human Activities in Three Areas of Central Bulgaria. *Acta Zoologica Bulgarica*, 72(3): 413-420 IF=0.278 Q4 SJR 2019= 0.21

Abstract: Food preferences of golden jackal *Canis aureus* Linnaeus, 1758 from three areas with different anthropogenic impact in Central Bulgaria during the autumn and winter seasons of 2016–2018 were investigated. A total of 170 stomachs were analyzed. In the mountainous area (Stara Planina Mts.), wild ungulates predominated in the jackal’s diet, followed by rodents, trash

and plant material (mainly fruits). Domestic mammals predominated in the jackal's diet in the hilly and semi-mountainous area (Sarnena Sredna Gora Mts.), followed by carcasses of wild carnivores and fruits. In the agricultural lowland area (Thracian Plain), rodents occurred in the jackal's diet most frequently, followed by plant materials (fruits and cereals), domestic mammals and fishes. Considering the narrower jackal's food niche in Upper Thracian Plain and in Stara Planina Mts. compared to that in Sredna Gora Mts., it can be concluded that rodents were favored by the jackal in the lowlands, and hunter-killed ungulates in the mountainous area. In the semi mountainous area, the species did not take advantage of a particular food category, demonstrating feeding generalization. Delivering easily accessible and high energy food in both anthropogenic environments and natural habitats makes human influence widespread.

20. Kirilov K., S. Peeva, 2021. On the Golden Jackal's (*Canis aureus* Linnaeus, 1758) distribution during snow period in the central part of Stara Planina Mts, Bulgaria. *Ecologia Balkanica*, 13(1): 77-84. producers conflict" in South-eastern Bulgaria. *Trakia Journal of Science* 1: 35-37.


Abstract. The aim of the present study was to determine the frequency of occurrence of the Golden jackal according to snow cover changes in different elevations and slope exposure in the central parts of Stara Planina Mts, Bulgaria. To achieve the purpose, 15 camera traps were used, at both, south and north slopes of the mountain on 5 different elevations. The data revealed a significant difference in the number of jackals detected between the southern and the northern slopes as well as at different elevation of the Central Stara Planina Mts. The largest number of jackals was found at the lowest level (up to 600 m a.s.l.). With elevation increasing, the number of detected individuals decreased. The jackal presence at 1000 - 1200 m a.s.l. in both studied areas was sporadic, and over 1200 m a.s.l. the species was not detected. The Golden jackal from the region of Central Stara Planina Mts demonstrated clear preferences to the southern slopes up to 800 m a.s.l.

21. Ishikawa K., R. Doneva, E. Raichev, S. Peeva, V. Doichev, Y. Amaiike, Y. Nishita, Y. Kaneko, R. Masuda, 2021. Population genetic structure and diversity of the East Balkan Swine (*Sus scrofa*) in Bulgaria, revealed by mitochondrial DNA and microsatellite analyses. *Animal Science Journal*, 92:e13630. Q1 SJR 2020= 0.61 IF=1.749

Abstract. The East Balkan Swine (EBS) is the only indigenous pig breed in Bulgaria. We analyzed the mitochondrial DNA (mtDNA) control region and 21 microsatellite loci for 198 individuals from 11 farms in Bulgaria. Obtained 11 mtDNA haplotypes including three novel ones were grouped to two major clades, European clade E1 (146/198 individuals, 73.7%) and Asian clade A (52/198, 26.3%). The mixture of the two clades may have resulted from historical crossbreeding between the European and Asian pig breeds. Clade A was frequent in southeastern Bulgaria (Burgas Province), but less frequent or absent in northeastern Bulgaria (Varna and Shumen Provinces). The distribution of Europe- and Asia-specific haplotypes relative to EBS farm locations could be attributed to regional differences of breeding systems (e.g., crossbreeding with imported commercial pigs). A microsatellite analysis showed high heterozygosities for all the EBS farms, and negative inbreeding coefficients presumably due to

crossing with commercial pigs or wild boars and/or efforts to reduce inbreeding by farmers. Bayesian clustering analyses showed that all farm populations are genetically well distinguishable from one another. Although diversity has been maintained by the efforts of farmers and a breeding association, the effective population size remains small, and conservation efforts should be continued.

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