POPULATION GENETIC STRUCTURE OF AUTOCHTHONOUS BLACK SLAVONIAN PIG

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5 SUMMARY

6 As Black Slavonian pig was forgotten and replaced with commercial hybrid pigs, after mid 1950s population

- 7 declined drastically. Today's population was founded from 46 sows and 6 boars in 1996 while currently
- 8 consist of 1064 sows and 163 boars. Aim of this study was to evaluate the genetic structure and genetic
- 9 variability in autochthonous Black Slavonian pig breed using pedigree. The quality of the pedigree
- 10 information was evaluated by percentage of known ancestors, which was about 35.16%. Increase in pedigree
- 11 completeness over the years is evident. Number of inbred Black Slavonian animals was 1695. Average
- 12 inbreeding was 3.2%, with evident growth observed by year. Effective number of founders was 50.8 for males
- 13 and 51.1 for females. Effective number of ancestors was, 35.8 for male and 34.8 for female animals,
- 14 respectively. The proportion of the genes contributed to the reference population of males and females by the
- 15 most important ancestor was 7.1% and 9.1%. The first 15 ancestors for males and 12 for females explained
- around 50% variability in the gene pool. Average effective population size via number of parents for last 10
- 17 years of Black Slavonian pig is 189.2. In order to perform proper selection decisions, additional work should
- 18 be done to increase the quality of pedigree data which affects the reliability of estimated parameters and
- 19 genetic structure of the population.

20 Keywords: pedigree analysis, Black Slavonian pig, inbreeding, genetic variability, effective population size

21 INTRODUCTION

22 In pig breeding during the last decades, highly productive breeds developed mainly by breeding 23 companies have replaced the local breeds which are essential resources of genetic diversity (FAO, 24 2007). It is therefore crucial to preserve the genetic diversity of local autochthonous breeds for future breeding programs. In Croatia, the autochthonous Black Slavonian pig was created by earl Karl 25 Leopold Pffeifer in the second half of the 19th century. For this purpose of creating a new breed with 26 27 improved production performance, he crossed gilts of local breed Lasasta mangulica with Berkshire 28 boars. Later from the 1880s till the 1910, he was crossing the best gilts with boars of American breed Poland China to further improve performance. The result of this breeding program was successful as 29 Black Slavonian pig was the most widespread breed in Croatia till the middle of 20th century used for 30 31 both fat and meat production. However, later due to no breeding program established, breed was 32 replaced by modern hybrids so the population declined drastically and suffered a narrow bottleneck 33 especially in the mid-1990s.

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- In September of 1996, population consisted of 46 sows and 6 boars (Uremović, 2004) which were the
- 40 first sows and boars of this breed registered in Croatian Agricultural Agency. Since that time, the
- 41 pedigree was recorded.
- 42 Breeds as Black Slavonian pig usually have small population sizes which results in high inbreeding
- 43 coefficients often followed by reduced fitness. The high inbreeding coefficient should be reduced by
- 44 choosing the proper strategy (Meuwissen and Woolliams, 1994) considering the genetic structure and
- 45 genetic variability. Aim of this study was to evaluate genetic structure and genetic variability of Black
- 46 Slavonian pig with a use of pedigree information.

47 MATERIAL AND METHODS

- 48 The reference population contained all pigs born between 1995 and 2014. Entire pedigree of Black
- 49 Slavonian pig breed consisted of 3478 animals and was recorded by Croatian Agricultural Agency.
- 50 Current population of Black Slavonian pig consists of 1064 active sows and 163 boars registered
- 51 (Annual report 2014, Croatian Agricultural Agency).

52 Pedigree analysis and genetic variability parameters

- 53 Ancestors which had both parents unknown were considered as founders, while in the case of one
- 54 known parent, that parent was considered to be a founder. The quality of the pedigree was evaluated
- 55 by the following parameters: percentage of known ancestors- expressed as percentage of known
- ancestors of the expected number in a generation; maximum generation traced back- expressed by
- 57 tracing back the pedigree for each individual and average pedigree completeness per year. The first
- measure of genetic variability calculated was the effective number of founders f_e (Lacy, 1989).
- 59 Secondly, effective number of ancestors f_a was calculated as the minimum number of ancestors
- 60 described by Boichard et al. (1997). Individual inbreeding coefficient (F) was used to show probability
- of two individuals having genes identical by descent and was calculated according to Wright (1931).
- 62 Average relatedness of individuals from a given pedigree was also analysed. Furthermore, effective
- 63 population size (N_e) per year was calculated according to Wright's (1931) method and denoted as N_{ew}
- 64 and by Falconer and Mackay (1996) denoted as NeF. Number of breeding males and females by year
- and generation interval is shown.

66 Software used

- 67 Preparation of pedigree file and statistical analyses were carried out using SAS software
- 68 (SAS/STATTM, 1999). Analysis were performed using CFC (Sargolzaei et al., 2006), PopRep 1.0
- 69 (Groeneveld et al., 2009) and PEDIG (Boichard et al., 1997) program packages.

70 RESULTS AND DISSCUSSION

- 71 Figure 1 shows the number of animals registered in Croatian Agricultural Agency per year starting
- from 1996. The figure clearly shows that the number of registered boars and sows was continuously
- 73 on the rise during the last 18 years.



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Figure 1. Number of animals registered in Croatian Agricultural Agency from 1996 till 2014

76 This positive trend could be explained by the several actions taken. In the early years, the important 77 actions were probably joining the FAO initiative for preserving the autochthonous pig breeds in 1999 78 and designing the breeding program in 2000, with the main aim to breed Black Slavonian pigs on a 79 small family farms. Later on, a small but important pig breeders associations were established that 80 further encouraged the breeding of this pig breed. Additionally, state government supported the 81 breeding of indigenous breeds by subsidizing production. Nowadays, producing the traditional high 82 quality products with more added values offers the opportunity for small local farmers and in that way 83 making these products more competitive on a global market. 84 Average pedigree completeness per year is shown in Table 1. The recording of the pedigree 85 information for Black Slavonian pig began in 1996 and there was a maximum of 14 generations traced 86 back, which was for the 2% of animals in the pedigree. Animals from the pedigree had 278 known 87 sires and 945 dams. By comparing the animals born in 1996 with the newer generations in 2014, pedigree completeness was increased as expected since the pedigree completeness provides 88 89 information of known ancestors within arbitrary generations. The highest completeness was in the last 90 recorded year where almost 70% of the pedigree was known for 6 generation back. However, still a lot 91 of effort should be done in order to provide higher quality of the pedigree data since the more

92 completed information is available only in last recorded years for a first 4 to 5 generations.

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Table 1. The average pedigree completeness (%) for 1 to 6 generations deep by year

	Year	Number	Complete	Complete	Complete	Complete	Complete	Complete
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	of animals	generation 1	generation 2	generation 3	generation 4	generation 5	generation 6
1995	2	0.0	0.0	0.0	0.0	0.0	0.0
1996	7	0.0	0.0	0.0	0.0	0.0	0.0
1997	14	0.0	0.0	0.0	0.0	0.0	0.0
1998	12	16.7	10.4	6.9	5.2	4.2	3.5
1999	16	31.2	15.6	10.4	7.8	6.2	5.2
2000	43	39.5	23.4	15.6	11.7	9.4	7.8
2001	59	54.2	30.6	20.5	15.4	12.3	10.2
2002	128	41.4	26.2	18.3	13.7	11.0	9.1
2003	256	62.5	42.3	29.3	22.0	17.6	14.6
2004	201	68.7	50.9	37.7	28.6	22.9	19.1
2005	213	92.0	68.0	49.9	38.1	30.5	25.4
2006	325	92.6	74.2	57.6	45.1	36.2	30.2
2007	234	94.9	84.0	68.7	54.7	44.3	36.9
2008	285	99.3	84.0	70.0	56.9	46.5	38.8
2009	306	92.8	88.6	79.2	66.8	55.6	46.6
2010	258	99.6	97.1	90.3	79.4	67.4	57.0
2011	286	94.4	92.4	87.3	79.3	68.9	58.8
2012	197	100.0	99.1	95.3	86.6	75.3	64.5
2013	292	100.0	99.1	96.0	89.0	79.1	68.6
2014*	18	100.0	100.0	95.2	89.0	79.9	69.6

94 * data included until 31.03.2014

95 Total number of founders for this population was 452 while the effective number of founders was 50.8

and 51.1 for males and females, respectively. The total number of founders is much higher than the

97 effective number of founders, probably because at the initial years of the recording all animals were

98 included. Similar results were obtained in the study of Crovetti et al., (2013) on Italian local breeds.

99 As these parameters were calculated using the formula of Lacy (1989) which considers only the

100 founders contribution, information on more recent ancestors could be more relevant in expressing

101 genetic diversity. Therefore, calculating the effective number of ancestors f_a using the formula of

102 Boichard et al., (1997) was used to overcome this drawback and cases if the bottlenecks in the

103 pedigree were present. Obtained f_a values were lower than effective number of founders, 35.8 for male

and 34.8 for female animals, respectively. Moreover, those values were higher than the values of

105 Italian (Crovetti et al., 2013) and French (Maignel et al., 2001) local breeds what could be the result of

106 shallow pedigree in our situation compared to theirs.

107 The proportion of the genes contributed to the reference population of males and females by the most

108 important ancestor is 7.1 % and 9.1 %. The first 15 ancestors for males and 12 for females explained

around 50 % variability in gene pool. The average age of parents indicates that animals reproduce

110 young and stay relatively short in the reproduction (3.1 years was the mean age of reproducing sires

111 and 2.7 years for dams).





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Figure 2. Average inbreeding (F) per year

114 Mean relatedness coefficient was 0.049. Number of inbred Black Slavonian animals is 1695. The

mean coefficient of inbreeding for the population (inbred and non-inbred animals) was very high, 3.20 115 % (with maximum value of 40.75 %). Mean inbreeding coefficient for inbred animals was estimated

117 to 6.56%. As the pedigree was getting more complete by years, it is easier to spot inbreeding.

118 Therefore, highest inbreeding was present in the years 2011 and 2013. Average inbreeding by year is

119 shown in Figure 2.

120 The calculated values of inbreeding were higher than the values from the studies of Crovetti et al.,

121 (2013) and Maignel et al., (2001) on similar type of local breeds, but were lower than the values

calculated for the Bisaro local breed (Fernandes et al., 2010). Nevertheless, planned and carefully 122

organized breeding is required in order to prevent high inbreeding and loss of genetic variability. 123

Effective population size (N_e) per year is presented in Table 2. With regard to certain conditions like 124

population substructure or pedigree completeness, different methods for calculating Ne should be 125

126 considered. In this study, inbreeding New calculated by Wright (1931) was 132 in the year 2014.

However, as the inbreeding was present in this study, it can be used to calculate more realistic 127

128 effective population size of a population. When inbreeding rate was accounted, NeF (Falconer and

129 Mackay, 1996) dropped significantly. On the other hand, this method is affected by the long term

effects of selection and it is also very sensitive to incomplete pedigree information (Boichard et al., 130

- 131 1997).
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Table 2. Effective population size by year via number of parents

			Nu	nber		
Year	Animals	Sires	Dams	Parents	New	Ne_F
1995	2	1	1	2	1	-
1996	9	3	1	4	2	-
1997	23	3	1	4	2	-
1998	33	6	3	9	6	-
1999	42	10	8	18	12	-
2000	71	16	22	38	26	142
2001	118	21	43	64	40	111

2002	230	33	83	116	66	121
2003	443	53	147	200	109	278
2004	585	69	184	253	141	124
2005	670	76	223	299	159	120
2006	739	89	253	342	184	69
2007	772	93	265	358	193	37
2008	844	106	283	389	216	25
2009	825	102	271	373	208	20
2010	849	101	296	397	211	25
2011	850	102	308	410	215	26
2012	741	92	281	373	194	62
2013	775	83	285	368	180	92
2014	507	62	197	259	132	-

¹³³ 134 New: Effective population size calculated by Wright (1931)

Ne_F: Effective population size calculated by Falconer and Mackay (1996)

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136 There are few recommendations for minimum N_e in domestic animals. Food and Agriculture

137 Organization of the United Nations (FAO, 2000) suggested the N_e values higher than 50 which is

138 considered as critical. Meuwissen and Woolliams (1994) suggested that the values of N_e should range

139 from 31 to 250 to prevent a decline in fitness. Values in this study are higher, however, they should be

140 taken with caution as they depend on the quality of the pedigree data.

CONCLUSION 141

142 Although population of Black Slavonian pig showed positive trends in growth during last 20 years, it

- 143 is still very vulnerable. Pedigree for the Black Slavonian pig population showed to be more complete
- 144 by each year. Inbreeding coefficient of 3.2 % for entire population is not alarming itself, but

145 completeness of the pedigree should be taken into account as well as information that inbreeding is

- growing with the pedigree completeness. Effective population size presented here has to be considered 146
- 147 with caution in regard to inbreeding coefficient. Future breeding strategies should rely more on the
- 148 pedigree to avoid inbreeding.

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151 **LITERATURE**

- 1. Boichard, D., Maignel, L., Verrier, E. (1997): The value of using probabilities of gene origin 152 153 to measure genetic variability in a population. Genet Sel. Evol. vol 29, pp. 5-23.
- 2. Crovetti, Alessandro (2013): Pedigree analysis of Cinta Senese and Mora Romagnola breeds 154 155 breeds. Acta Agriculturae Slovenica, suppl. 4, pp. 41-44.
- 3. Falconer, D. S. and Mackay, T. F. C. (1996): Introduction to Quantitative Genetics. Longman, 156 157 Essex, U.K., 4th edition.
- 158 4. FAO (2000): Secondary guidelines for development of national farm animal genetic resources 159 management plans: Management of small populations at risk. UN Food and Agric. Org. 160 http://www.fao.org/AG/AGAInfo/resources/en/pubs_gen.html

161	5.	FAO (2007): The State of the World's Animal Genetic Resources for Food and Agriculture.
162		UN Food and Agric. Org. http://www.fao.org/docrep/010/a1250e/a1250e00.htm.
163	6.	Fernandes, S.D.: Malovrh, S.; Kovac, M.; Cadavez, V. (2010): Study of genetic diversity of
164		Bisaro pigs breed by pedigree analysis. Lucrări Științifice. SeriaZootehnie.53:15. pp. 326-330
165	7.	Groeneveld, E., Westhuizen, B., Maiwashe, A., Voordewind, F., Ferraz, J. B. S. (2009):
166		Population Management. POPREP: A Generic Report for Genetics and Molecular Research
167	8.	Croatian Agricultural Agency, Annual reports (1996-2014.)
168	9.	Janssens, S., J. Depuydt, S. Serlet, and W. Vandepitte (2005): Genetic variability in pigs
169		assessed by pedigree analysis: The case of Belgian Landrace NN and Pietrain in Flanders.
170		Page 98 in Proc. 56th Eur. Assoc. Anim. Prod., Uppsala, Sweden.
171	10.	Lacy, R. (1989): Analysis of founder representation in pedigrees: Founder equivalents and
172		founder genome equivalents. Zoo Biology Volume 8, Issue 2, pages 111-123.
173	11.	Maignel, L., Labroue, F. (2001): Analyse de la variabilité génétique des races porcines
174		collectives et des races locales en conservation à partir de l'information généalogique.
175		Journées dela Recherche Porcine. 33: 111–117
176	12.	Meuwissen, T.H.E., Woolliams, J.A. (1994): Effective sizes of livestock populations to
177		prevent a decline in fitness. Theoretical and Applied Genetics.89: 1019-1026
178	13.	Sargolzaei, M., H. Iwaisaki and J.J. Colleau. (2006): CFC: A tool for monitoring genetic
179		diversity. Proc. 8th World Congr. Genet. Appl. Livest. Prod., 27-28. Belo Horizonte, Brazil.
180	14.	Uremović, Marija (2004): Crna slavonska pashmina svinja, Hrvatska izvorna pasmina.
181		Vukovar: Vukovarsko-srijemska županija, 2004.
182	15.	Wright, S. (1931): Evolution in mendelian populations. Genetics 16, 97-159.