Evaluation of the Istrian cattle population structure by pedigree analysis

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Abstract
Local cattle breeds are an important part of genetic diversity, but due to globalization and industrialization of production, they are often marginalized. Istrian cattle is a local breed that almost became extinct three decades ago. Active measures have been taken to protect the Istrian cattle and the population has increased, gaining a new economic importance. The objective of this study was to analyze the status and trends of Istrian cattle using pedigree information. The population of Istrian cattle has stabilized trends in the number of animals and herds, although it requires constant monitoring. The levels of relatedness and inbreeding have plateaued (at 4.00% and 3.14%, respectively) and bottleneck in the population has not been observed. The mean number of equivalent generations is relatively low (2.99) and further systematic monitoring is important.

Introduction
Genetic diversity is one of nine control variables in the planetary boundaries framework (Steffen *et al.*, 2015). The genetic diversity of cattle, the most important domestic species in terms of global production of food of animal origin, is gradually being depleted (FAO, 2015). Of the total number of breeds of mammalian species used for agriculture worldwide, cattle breeds account for 22.90% (FAO, 2019). Although the number of extinct cattle breeds is relatively low (12.8%), a large number of breeds are endangered (21.1%) and only 12.3% of cattle breeds are in "not endangered" status. For most cattle breeds, their endangered status is unknown (53.8%) (FAO, 2019).
Istrian cattle is one of the three Croatian local cattle breeds. The primary breeding area of Istrian cattle is the Istrian peninsula and the neighboring island, and in last ten years has been extended to karst area (Lika, Dalmatia). It belongs to the Podolian group of cattle breeds and is very similar to the Italian Podolian and Maremmana breeds (Maretto *et al.*, 2012). Until the early twenties of the 20th century, cattle production in Croatia was based on local cattle breeds. Due to the modernization of agricultural production, the use of mechanization and the introduction of allochthonous breeds, local breeds were marginalized, and their survival was in danger. In the nineties of the 20th century, the program of protection of Istrian cattle was launched and the initial population included 103 cows and eight bulls (Ramljak *et al.*, 2011). According to the Croatian Agency for Agriculture and Food (2021), the current population of Istrian cattle is 1,118 adult animals (65 bulls and 1,053 cows) housed in 174 breeding farms. Previous studies on Istrian cattle included phenotype analysis and genetic analysis (Ivanković *et al.*, 2007; Ramljak *et al.*, 2011; Maretto *et al.*, 2012), but pedigree analysis was not performed. Pedigree information is a useful source for conservation programs to maintain genetic variability and minimize inbreeding (Santana *et al.*, 2016), and to analyse the breeding status of population (Jarnecka *et al.*, 2021). Therefore, the objective of this study was to

describe the population structure and trends of the Istrian cattle population through pedigree analysis.

Materials & Methods

The database of Istrian cattle includes 7,399 animals born between 1971 and 2020, belonging to 174 herds located mainly on the Istrian peninsula. For each animal in the database, information such as the identification number, date of birth, sex, sire, dam, and herd were recorded. In this study, active (alive) animals of both sexes were selected as reference population (REF 01). Demographic and population parameters are presented for the last two decades, because Herdbook was closed in 2000. Pedigree analysis was performed using ENDOG v4.8 software (Gutiérrez and Goyache, 2005). Pedigree completeness was determined using the mean number of complete generations, mean number of maximum generations, and mean number of equivalent generations (Boichard *et al.*, 1997). Parameters related to inbreeding (mean inbreeding coefficient (*F*), mean average relatedness coefficient (*AR*), effective population size (*Ne*)), probability of gene origin (effective number of founders (f_e), effective number of ancestors (f_a), effective number of founder herds (f_h)), and generation interval (*GI*) were assessed. The effective population size was estimated via an individual increase in inbreeding per generation (Gutiérrez *et al.*, 2008). The average relatedness coefficients and individual inbreeding coefficients were calculated based on Gutiérrez and Goyache (2005). The generation intervals were calculated for the four paths: sire-son, sir-daughter, dam-son and dam-daughter.

Results

Demographic trends in the Istrian cattle population are shown in Figure 1. The number of cows has been constant1y increasing per year. According to Figure 1, the number of herds and number of new registered animals (offspring's included in the Herdbook per year) have increased from 2000 to 2011 and have only been roughly stable in the last decade.

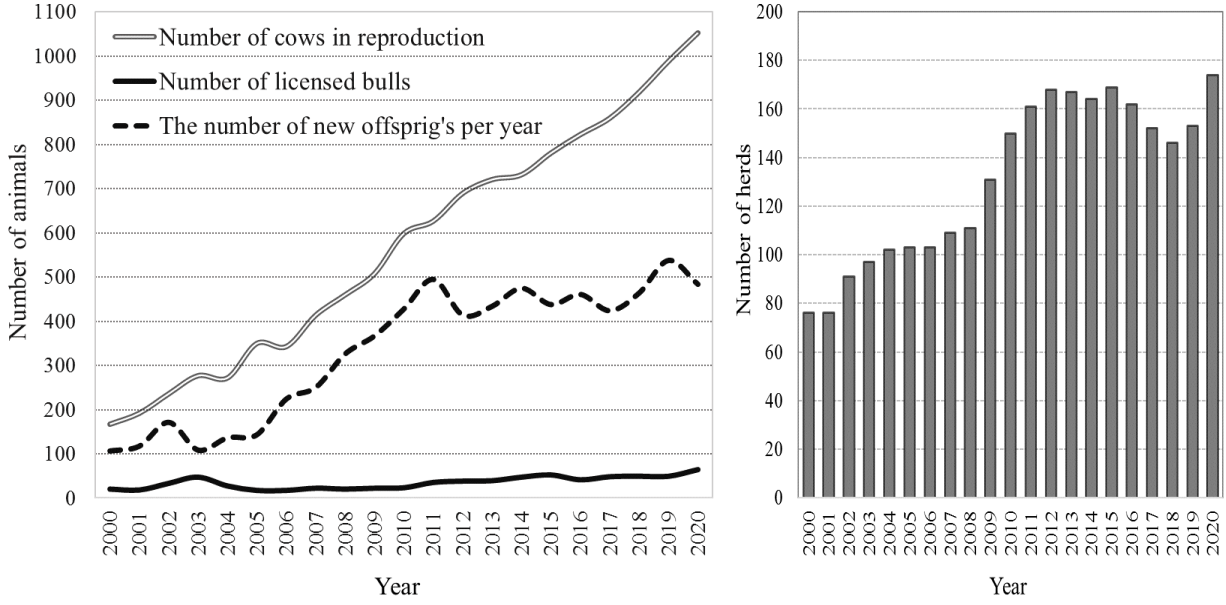

Figure 1. Population trends in the Istrian cattle population in the last two decades

An analysis of all pedigree entries revealed 6,756 animals for which both parents were known. The active living population consisted of 2,207 active animals and the effective

population size is 91.92 (Table 1). The number of founder animals was 235 and the mean number of equivalent complete generations was 2.99.

Table 1. Population size and genetic diversity indices of the Istrian cattle

Item	Total
Original dataset (TP; total population)	7,399
Reference population 1 (REF 01; live animals)	2,207
Mean inbreeding coefficient (*F*)	3.14
Mean Average Relatedness coefficient (*AR*)	4.00
Effective population size (N_e)	91.92
Mean number of complete generations (*MCG*)	1.79
Mean number of equivalent generations (*MEG*)	2.99
Mean number of maximum generations (*MMG*)	5.81
Number of founders contributing to reference population	235
Effective number of founders for the reference population (f_e)	36
Effective number of ancestors for the reference population (f_a)	35
Effective number of founder herds (f_h)	17.10
Effective number of founders/effective number of ancestors (f_e/f_a)	1.03

The ratio between effective number of founders and effective number of ancestors is 1.03, The average generation interval was 5.93 ± 3.65 years and ranged from 4.77 ± 3.07 (sire - son) to 6.69 ± 3.84 (dam - son). The average inbreeding coefficient in the REF 01 population was 3.14%, ranging from a minimum of 1.41% (2002) to a maximum of 3.19% (2018) (Figure 2). The *AR* in the population was 4.00%, ranging from 3.31% (2008) to 4.27% (2000).

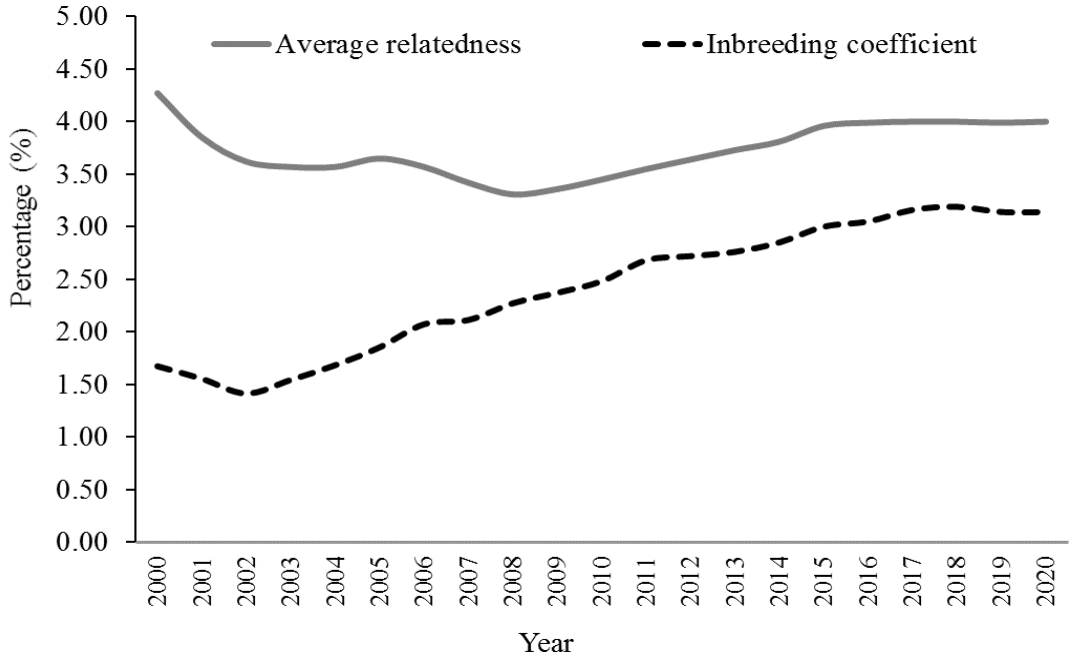

Figure 2. Level of inbreeding coefficient and average relatedness (in %) in the Istrian cattle from 2000 to 2020

Discussion

The Istrian cattle population is subject to a systematic protection program, and an economic revitalization program has been implemented in the last decade. Considering the number of

animals, the population trend is positive, as well as the increase in the average size of the herd from 2.5 adult animals (in 2000) to 6.4 (in 2020). The sex ratio in the population (cows/bulls) increased from 8.0 (in 2000) to 16.2 (in 2020). In the last ten years, the growth of the inbreeding level has slowed down, while in the last five years it has stagnated around 3.14%. The level of inbreeding observed in Istrian cattle is not alarming, but requires constant monitoring. We need to be careful in monitoring the level of inbreeding because Boichard *et al.* (1997) pointed out that the *F* trend has some limitations, especially when pedigree completeness is low. It should consider the fact that pedigree completeness is still low in the Istrian cattle population (*MCG*=1.79). The relationships between N_e, f_e and f_a provide information about the occurrence of bottleneck in a population. For example, a f_e/f_a ratio close to 1 suggests that bottlenecks have not occurred in the population (Boichard *et al.*, 1997). A high f_e/f_a ratio indicates disproportionate use of breeding animals (Stephens and Splan, 2013). The f_e/f_a ratio in Istrian cattle is currently 1.03, indicating that there is no obvious bottleneck in the population. We assume that this is a consequence of the higher presence of natural mating compared to artificial insemination in practice.

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