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Original Paper

Genetic variability within the Polish population of red fox (*Vulpes vulpes*) – Preliminary results

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Red fox (*Vulpes vulpes*) represents family *Canidae* and is a very common predator in Poland. Foxes are present throughout all the country in a different geographical regions and habitats. The analyzed dataset consisted of 133 red foxes (*Vulpes vulpes*). There were 24 microsatellite sequences studied. The observed (H_O) and expected (H_S) heterozygosities were comparable within respective loci. H_O per locus ranged from 0.2135 to 0.9146 and H_S ranged from 0.2375 to 0.9711. The low genetic diversity of the population was found. There were four genetic subpopulations and one genetically distant individual found basing on genetic distances and PCoA analysis.

Keywords: genetic variability, *Vulpes vulpes*, microsatellites

1 Introduction

Red fox (*Vulpes vulpes*) represents family *Canidae* and is a very common predator in Poland. Foxes are present throughout all the country in a different geographical regions and habitats. The significant genetic differences between fox populations dependent on the place of occurrence have been found in many studies (Atterby et al 2015, Edwards et al. 2012, Mullins et al.2014, Oishi et al. 2011, Wandeler et al. 2003).

The aim of the study was to reveal the genetic variability of red foxes living in Poland.

2 Material and methods

Samples were collected from animals obtained from hunters and traffic accidents. Tissue samples for DNA extraction were taken from tongues of the 133 studied animals. The DNA was isolated in accordance to ARK Genomic self-protocol. There were 30 microsatellite markers described and localized in the dog genome chosen for the study. The multiplex PCR method was used to amplify markers sequences which were divided into four pools for analysis. Microsatellites were genotyped using automated sequencer 3730xl (Applied Biosystems). Obtained data were analysed using GeneMapper v. 4.0 (Applied Biosystems).

The population genetic analyses were performed using R Project (R Core Team 2015). The allele and genotype frequencies were calculated in pegas package (Paradis, 2010). The observed (H_O) and expected (H_S) heterozygosities and inbreeding coefficients (F_{IS}, F_{ST}) for each of the microsatellite markers were estimated using hierfstat package (Goudet and Jombart, 2015). Hardy – Weinberg Equilibrium tests were performed in pegas package. The inbreeding coefficients (F) of the individuals were estimated with poppr package (Kamvar et al., 2014, Kamvar et al., 2015). Genetic distances for Principal Coordinates Analysis (PCoA) were calculated using ape package (Paradis et al., 2004).

3 Results and discussion

There were twenty three polymorphic microsatellite loci (AHT137, FH2097, FH2613, FH2980, REN135K06, REN248F14, REN258F18, REN64E19, REN88H03, UOR4101, ZUBECA6, FH3241, FH3287, FH3713, FH3771, FH3775, FH3824, FH3970, FH4001, REN210I14, REN25E18, REN307J23, REN75M10, FH2295) analyzed. Locus REN248F14 was monomorphic and thus was excluded from the further analysis.

The observed (H_O) and the expected (H_S) heterozygosities were comparable within respective loci. H_O per locus ranged from 0.2135 for UOR4101 to 0.9146 for FH4001

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Table 1 Basic parameters of genetic variability estimated for studied *Vulpes vulpes*

	n	n_A	n_G	H_O	H_S	F_{ST}	F_{IS}	HWE p-value
AHT137	133	13	30	0.7778	0.8009	-0.0155	0.0288	0.201
FH2097	123	18	53	0.7238	0.9232	-0.0095	0.2160	0.000
FH2613	129	13	33	0.6470	0.6746	0.0238	0.0409	0.007
FH2980	127	20	83	0.8270	0.9117	0.0252	0.0928	0.074
REN135K06	126	8	20	0.6990	0.6648	0.0295	-0.0515	0.861
REN258F18	126	18	55	0.8182	0.8523	0.0239	0.0400	0.004
REN64E19	129	9	26	0.7730	0.8008	0.0299	0.0348	0.199
REN88H03	126	3	6	0.2405	0.2434	0.0380	0.0118	0.007
UOR4101	128	3	5	0.2135	0.2497	-0.0308	0.1450	0.337
ZUBECA6	127	54	118	0.8886	0.9711	0.0026	0.0849	0.000
FH3241	127	15	36	0.8092	0.8483	-0.0228	0.0460	0.267
FH3287	111	5	9	0.6979	0.6339	0.0765	-0.1010	0.000
FH3713	128	39	82	0.9143	0.9046	0.0276	-0.0106	0.433
FH3771	117	27	81	0.8333	0.8786	0.0640	0.0516	0.184
FH3775	125	4	6	0.5180	0.4517	0.1000	-0.1469	1.000
FH3824	130	48	112	0.8864	0.9367	0.0304	0.0537	0.007
FH3970	116	17	51	0.5727	0.8688	0.0200	0.3408	0.000
FH4001	126	60	111	0.9146	0.9611	0.0124	0.0484	0.001
REN210I14	123	7	21	0.3632	0.7631	0.0333	0.5241	0.000
REN25E18	130	2	3	0.2649	0.2375	-0.0022	-0.1153	0.474
REN307J23	127	12	46	0.7445	0.8406	0.0344	0.1143	0.000
REN75M10	121	13	46	0.7085	0.8785	0.0145	0.1936	0.000
FH2295	108	5	8	0.2992	0.3868	0.0089	0.2264	0.008
Average	-	18.96	45.26	0.6580	0.7253	0.0224	0.0812	-

n – number of genotyped animals, n_A – number of alleles, n_G – number of genotypes, H_O – observed heterozygosity, H_S – expected heterozygosity, F_{ST} – fixation index, F_{IS} – inbreeding coefficient

with an average value of 0.658. H_S ranged from 0.2375 for REN25E18 to 0.9711 for ZUBECA6 and over all loci amounted to 0.7253. Comparable values of F_{IS} were obtained for fox population in England (Atterby et al., 2015). Lower values of H_O and H_S (0.508–0.860 and 0.527–0.880 respectively) were obtained in the study of red fox populations from north-eastern part of Poland (Mullins et al. 2014). The fixation index F_{ST} did not exceed 10% (for FH3775) indicating a low genetic diversity of the population. Also the inbreeding coefficient F_{IS} achieved rather low values, not exceeding 52.41% (for REN210I14), with an average value of 8.12% (Table 1).

The estimated inbreeding coefficients F of the analyzed individuals were illustrated with the histogram (Figure 1) and did not exceed 45%. The F coefficient ranged from 10 to 15% for the largest group of individuals (47 red foxes). The genetic distances between pairs of individuals were illustrated with PCoA plot (Figure 2).

Four genetic subpopulations and one genetically distant individual were found.

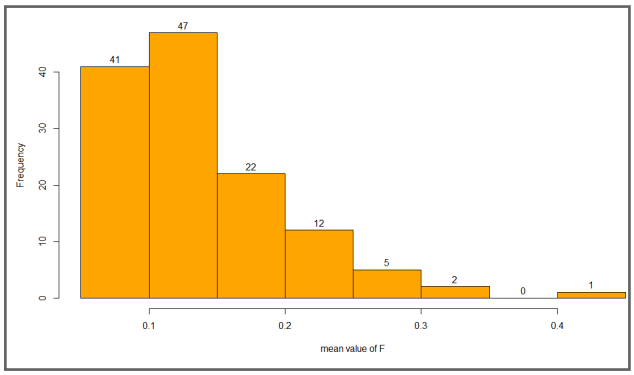

Figure 1 The histogram of the estimated inbreeding coefficients (F) of the studied individuals

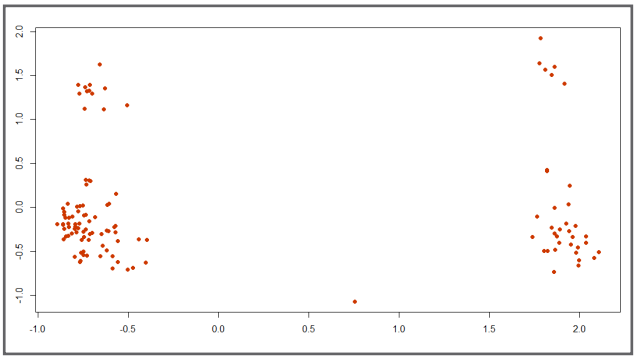

Figure 2 PCoA plot of genetic distances between pairs of individuals of studied population

4 Conclusions

The level of heterozygosity in analysed population was noticeable differential between microsatellite loci. Low genetic variability of the analyzed population confirms low level of diversity of other studied red fox populations in Poland and also in other European countries. The results showed, that fox population in Poland is not genetically uniform and at least four different subpopulations can be identified. The genetically distant subpopulations found with the PCoA method need further investigation.

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