

# Genetic structure and hybridization risk assessment for the wildcat (*Felis silvestris silvestris*) population in Croatia

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## INTRODUCTION

- European wildcat (*Felis silvestris silvestris* Schreber, 1777) is the least studied strictly protected wild carnivore in Croatia
- Genomic purity of European wildcat population is threatened due to frequent hybridization with domestic cats

## AIM

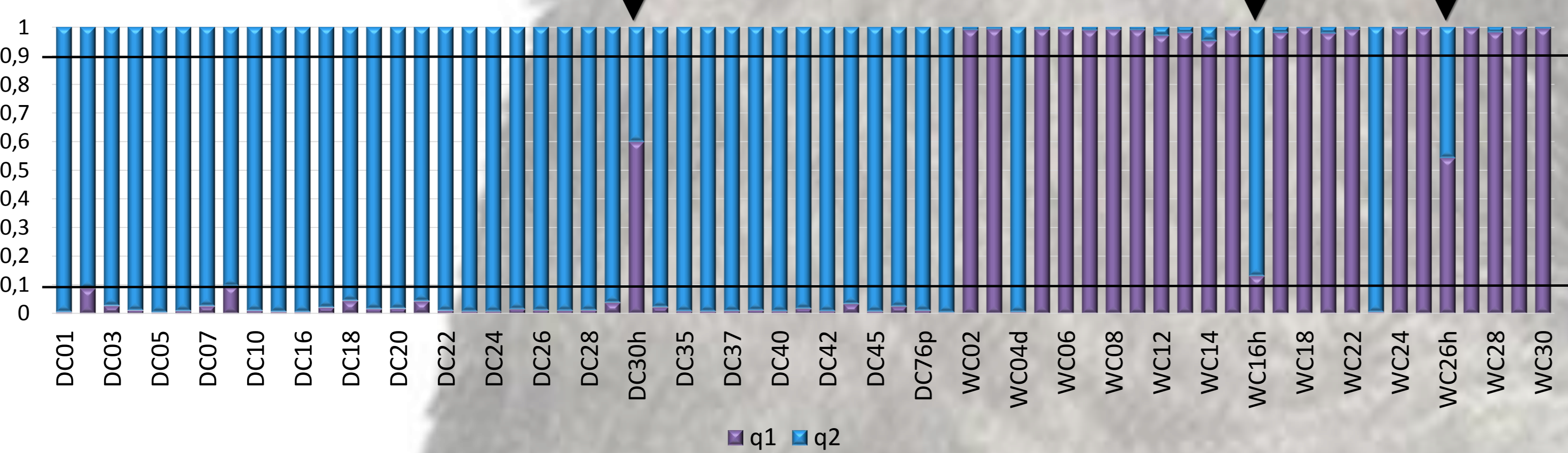
- To determine the level of hybridization between wild and domestic cats in Croatia by analyzing 12 microsatellite loci

## MATERIALS AND METHODS

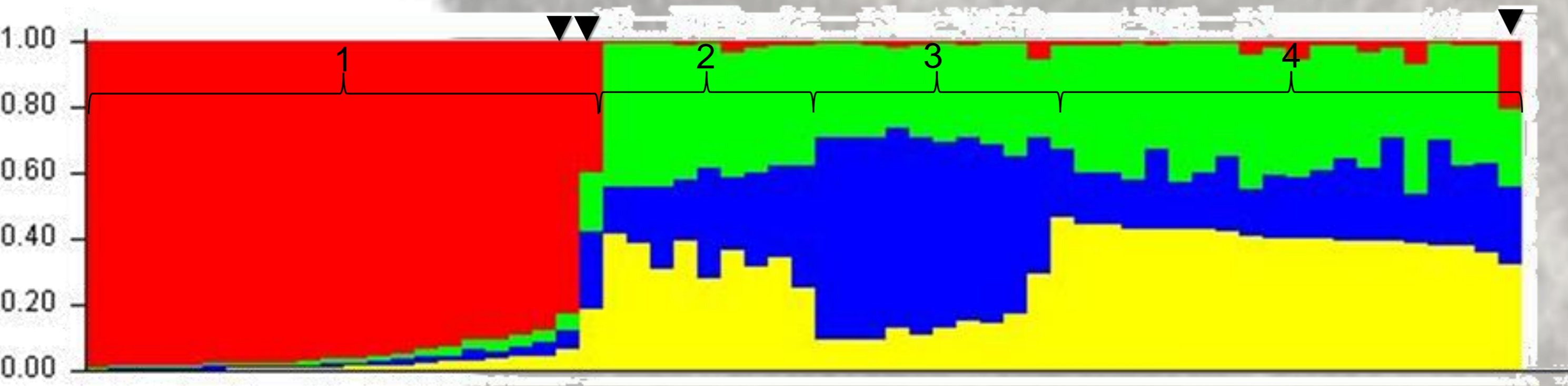
- 63 samples: muscle, blood and oral mucosa;-25 wild and 38 domestic cats were preliminary identified based on morphology. . The two subspecies cannot be always clearly determined based on external morphology, so genetic testing is necessary to provide for accurate population assignment.
- After DNA extraction, microsatellite loci were amplified using multiplex touchdown PCR and analyzed by capillary electrophoresis
- Population structure was assessed using Bayesian clustering implemented in STRUCTURE 2.3.4 and CLUMPAK, and the genetic variability of each population was analyzed using Arlequin 3.5.2.2., GenAEx 6.5 and Fstat 2.9.3.2.

## RESULTS

- Population assignment has identified 21 wildcats and 37 domestic cats (Fig. 1). Two individuals initially identified as wildcats proved to be domestic (cut-off value,  $q_i \geq 0.9$ ), and 3 domestic  $\times$  wildcat hybrids have been detected. Hybrid ratio in the sampled wildcats population is 8.70%, and 2.63% in domestic cats population, 4.92% of all samples, respectively. Misidentified and hybrid individuals were excluded from further population genetics analyses.



**Figure 1:** Bayesian clustering results (K=2). Probability of population assignment: q1 - wild cats, q2 - domestic cats. ▼ - hybrids ( $0.1 \leq q_i \leq 0.9$ )



**Figure 4:** Bayesian clustering (K=4). Four subpopulations detected: 1- wildcats; 2 – 4- domestic cats ▼ - hybrids are classified into the clusters with the most similarity

## CONCLUSIONS

Wildcats in Croatia represent a genetically distinct population with a moderate incidence of both hybridization with domestic cat and inbreeding (Figs. 1, 3, 4). Domestic cat population showed greater allelic variability but also larger inbreeding, as well as existence of distinct genetic subpopulations (Fig. 4). Like elsewhere in Europe, habitat loss presents a risk for cryptic extinction of wildcats.

## REFERENCES

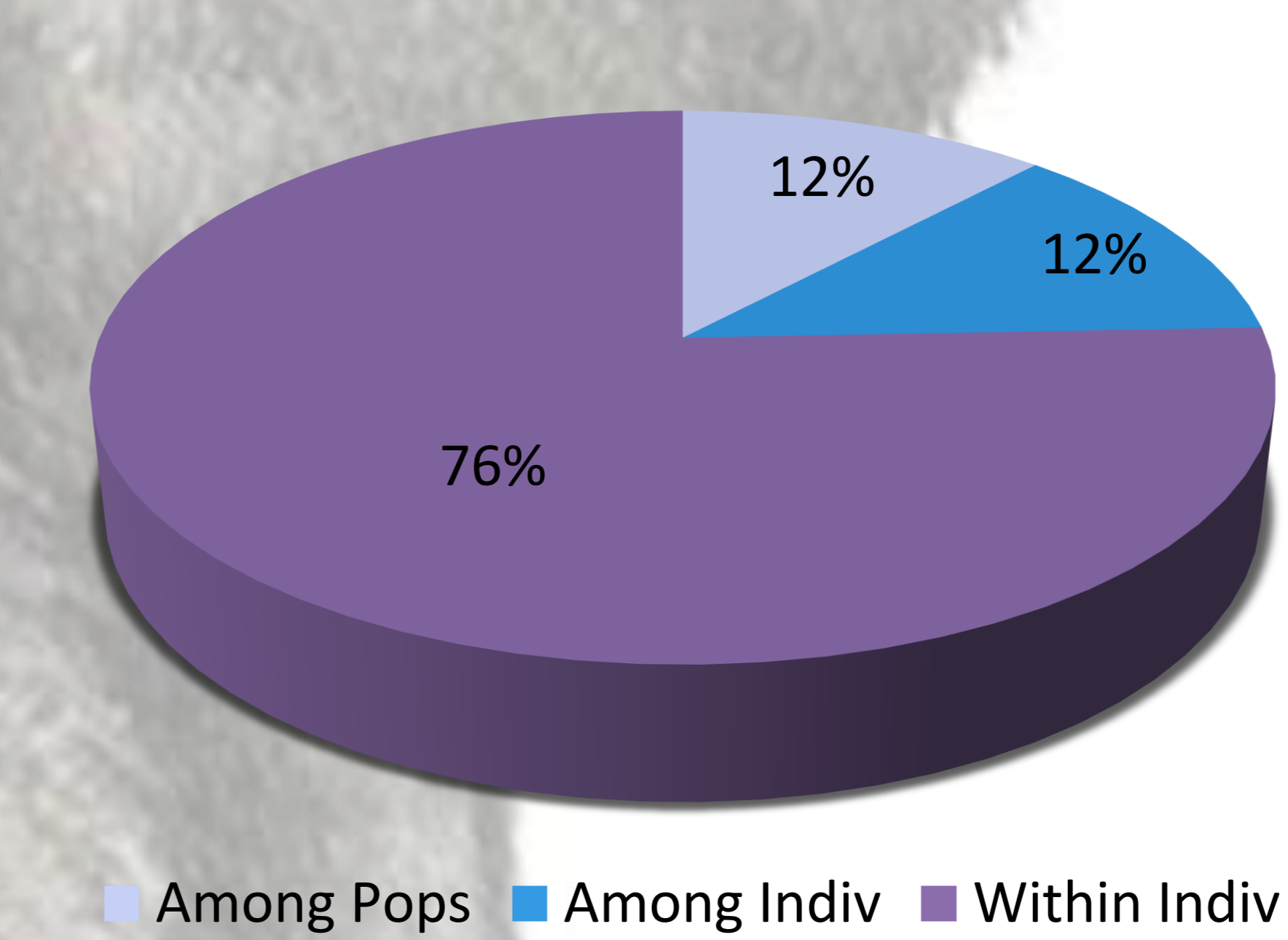
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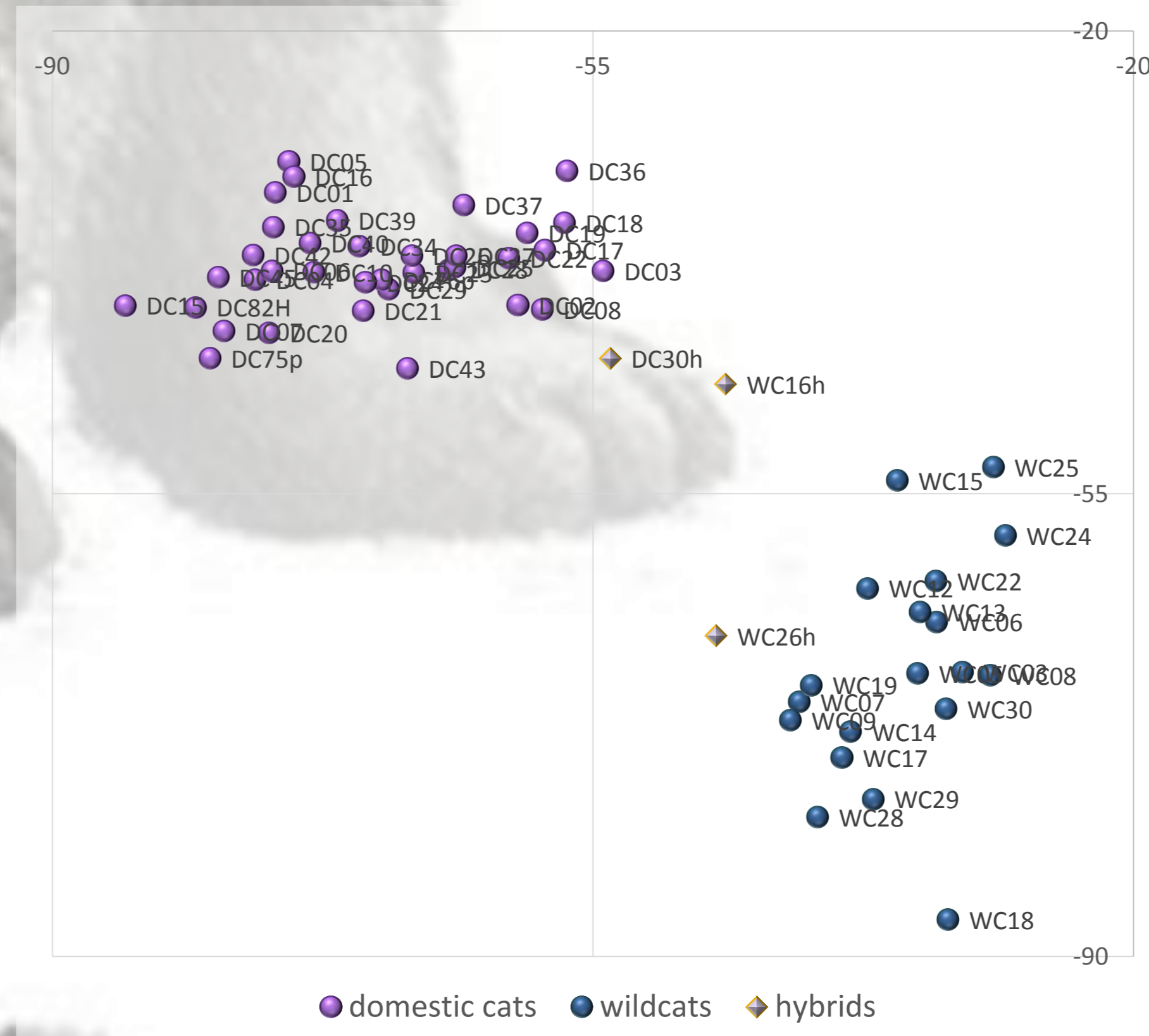
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- Pairwise diversity assesment between wildcat and domestic cat populations showed genetic distance index  $F_{ST}=0.12$ , inbreeding coefficient  $F_{IS}=0.141$ , overall inbreeding coefficient  $F_{IT}=0.244$ , Nei's genetic identity  $I=0.472$  and Nei's genetic distance  $-\log_e I=0.75$ . Molecular variance (AMOVA) data are given in Fig. 2. For each subspecies, genetic diversity data are given in Table 1.



**Figure 2:** AMOVA molecular variance analysis



**Table 1:** Genetic diversity data.  $N_a$  avg.no. of different alleles,  $N_p$  avg.no. of private alleles,  $H_o$ ,  $H_e$  observed and expected heterozygosity,  $F_{IS}$  inbreeding coefficient. Wildcats show more heterozygosity and less inbreeding.

population	domestic	wildcat
sample size	37	21
$N_a$	9.58	7.83
$N_p$	3.417	1.66
$H_o$	0.665	0.705
$H_e$	0.767	0.759
$F_{IS}$	0.146	0.096

**Figure 3:** PCoA analysis. Populations of wild cats, domestic cats and wild  $\times$  domestic hybrids clearly identified.