Elena Ballesteros-Duperón*, Emilio Virgós, Marcos Moleóna, José M. Barea-Azcón and José M. Gil-Sánchez

How accurate are coat traits for discriminating wild and hybrid forms of Felis silvestris?

Abstract: Hybridisation between domestic cats, Felis catus, and wildcats, Felis silvestris, could lead to the genetic extinction of the latter; therefore, checking hybridisation rates in wild populations is of vital conservation importance. However, detecting hybridisation in the field is particularly challenging. Here, we aim to test the success of morphological-based procedures for discriminating wildcats from their hybrids and domestic cats, against genetic methods. We checked 17 putative Spanish wildcats by using two different classification systems based on coat patterns. None of the putative wildcats analysed in this study seemed to have an admixed genotype. Concordance between genetic and pelage approaches was almost total: only one coat classification produced mixed results with detection of one potential hybrid. Assignment was worse when performed in the field after a rapid examination of coat characters. We conclude that classification systems using coat traits could serve as surrogates of genetic approaches, but only after careful examination of those characters with more discriminatory power. Thus, the control of hybrid populations in the field as a management tool to preserve the genetic identity of wild forms is problematic if based on crude approaches or incomplete classification systems.

Keywords: genetic introgression; morphological discrimination; pelage characters; Spain; wildcat.

*Present address: Department of Applied Biology, University Miguel Hernández, Ctra. Beniel km 3.2, 03312 Orihuela, Alicante, Spain.
*aCorresponding author: Elena Ballesteros-Duperón, Environment and Water Agency of Andalusia, Regional Ministry of Environment of the Junta de Andalucía, C/Joaquina Eguaras, 10, E 18013 Granada, Spain, e-mail: eballesterosd@agenciamedioambienteyagua.es, eleduperon@hotmail.com

Emilio Virgós: Escet, Department of Biology and Geology, University King Juan Carlos, C/ Tulipan, s/n, E-28933 Madrid, Spain

Marcos Moleón: Environment and Water Agency of Andalusia, Regional Ministry of Environment of the Junta de Andalucía, C/Joaquina Eguaras, 10, E 18013 Granada, Spain; and Centre for African Ecology, School of Animal, Plant and Environmental Sciences, University of the Witwatersrand, Wits 2050, Johannesburg, South Africa

José M. Barea-Azcón and José M. Gil-Sánchez: Environment and Water Agency of Andalusia, Regional Ministry of Environment of the Junta de Andalucía, C/Joaquina Eguaras, 10, E 18013 Granada, Spain

Introduction

The wildcat (Felis silvestris Schreber, 1775) may be regarded as a single polytypic species with up to five wild subspecies, the European wildcat (F. s. silvestris), the African wildcat (F. s. lybica), the Asian wildcat (F. s. ornata), the sub-Saharan African wildcat (F. s. cafra) and the Chinese mountain cat (F. s. bieti; Driscoll et al. 2007). The species is distributed over a large geographical region (Nowell and Jackson 1996). In Europe, wildcats are threatened by a combination of habitat destruction, persecution, hybridisation with domestic cats (F. catus Linnaeus, 1758; Stahl and Artois 1991, Nowell and Jackson 1996, Lozano and Malo 2012) and reduction in prey availability (Lozano et al. 2003, 2007, Lozano and Malo 2012). These processes have led to their disappearance from large areas, with the resulting fragmentation of most of their populations (Stahl and Artois 1991, Lozano and Malo 2012). The decrease in geographical range and population size has prompted the inclusion of the species in the Annex IV of the European Directive on the conservation of natural habitats (Council Directive 92/43/EEC; EUROP 1992).

Among the identified negative factors, hybridisation with domestic cats has been claimed as one of the most important in many parts of the wildcat’s distribution (e.g., Stuart and Stuart 1991, Hubbard et al. 1992, Nowell and Jackson 1996, Mendelsohn 1999). However, the incidence of this threat is highly variable among regions (Parent 1974, Beaumont et al. 2001, Daniels et al. 2001, Randi et al. 2001, Pierpaoli et al. 2003, 2007, Lozano and Malo 2012). The decrease in geographical range and population size has prompted the inclusion of the species in the Annex IV of the European Directive on the conservation of natural habitats (Council Directive 92/43/EEC; EUROP 1992).
density. This situation highlights the conservation value of the Iberian wildcats within a global perspective (Lozano and Malo 2012). However, despite the low occurrence of hybridisation detected to date in these populations, checking hybridisation rates in Iberian regions is highly recommended because of their high conservation value.

Nevertheless, detecting hybridisation in the field is a considerable challenge for conservationists and wildlife managers, so that the provision of fast, reliable and low-cost tools to identify hybrids or domestic cats that may threaten the wildcat genotype should be a key ingredient of conservation programmes for the species. Some studies have identified several morphological or pelage characters allowing separation between wildcats and domestic cats (Ragni and Possenti 1996, Puzachenko 2002, Yamaguchi et al. 2004, Kitchener et al. 2005, Krüger et al. 2009). However, these studies have normally been undertaken in areas with fragmented and low-density populations, where hybridisation is common and taxonomic discrimination is an especially difficult task. Frequent hybridisation rates can produce a continuum in phenotypic variation that makes particularly difficult the separation of hybrids from pure genotypes using morphological features, at least in field situations (Ragni and Randi 1986, French et al. 1988, Balharry and Daniels 1998, Daniels et al. 1998, Yamaguchi et al. 2004, Krüger et al. 2009, Platz et al. 2011, but see Kitchener et al. 2005). The reliability of testing diagnostic characters between populations that show different levels of introgressive hybridisation is largely unknown.

In this article, we study the degree of hybridisation of wildcats in a Spanish area with a low human population density, with the main aim of comparing the success of morphological-based discriminating approaches already available. To achieve this goal, we explore the congruence between two different classification systems based on coat patterns (Ragni and Possenti 1996, Kitchener et al. 2005) to separate domestic cats, wildcats and their hybrids, with the microsatellite results recently obtained by Oliveira et al. (2008a) using Bayesian admixture procedures. We discuss the management implications of our results, considering the species conservation status at both national (Near Threatened; López-Martín et al. 2007) and international (Least Concern; Driscoll and Nowell 2010) levels.

Materials and methods

Specimens, locations and genetic identity

We studied 17 wild individuals showing typical colour and morphological patterns of wildcats or their hybrids with domestic cats. According to Ragni and Possenti (1996), this sample size is enough for detecting consistent patterns in phenotypic variation. All individuals were from Granada province (SE Spain; Figure 1). Data from 11 of these were recorded in Sierra Harana during a capture-recapture programme for an ecological study of the local wildcat population, where individuals were captured in box traps (see Barea-Azcón et al. 2007 for details). We also used data from five additional road-killed wildcats and another individual captured in another mountain range in Granada. The locations of the latter individuals were well spread across the province (Figure 1). On the basis of the intensive fieldwork in Sierra Harana, we estimated a wildcat winter population density of >0.18 ind/km² (own unpubl. data). All specimens were sampled in winter.

Individuals were recorded in the field as genuine wildcats or hybrids based on a fast visual examination (Corbett 1979), but without using any of the suggested methods to discriminate different types of cats (Ragni and Possenti 1996, Kitchener et al. 2005). We obtained a tissue sample from the ear of each cat. Tissues were preserved in 100% ethanol until DNA extraction in the laboratory. When examined genetically, all of them were classified as wildcats with very high membership probabilities (see Oliveira et al. 2008a for complete details on the genetic procedures and results). Although we cannot completely rule out some past undetected admixture, these authors found no evidences of recent introgression. In consequence, all the specimens in this study are considered as wildcats (i.e., *Felis silvestris silvestris*) in a genetic sense. Our assumption hereafter is that the genetic approaches used by Oliveira et al. (2008a) are good indicators of introgressive hybridisation, at least in more recent generations.

Pelage characteristics

All cats were sexed and aged. The pelage characteristics of cats were assessed from dorsal, lateral, ventral, head and tail photographs (following the protocol outlined in Daniels et al. 2001). We used two different classification systems, the first based on Ragni and Possenti (1996) and the second on Kitchener et al. (2005). The pelage character scores of each individual were recorded on the basis of consensus criteria from independent and trained researchers (EB-D and EV).

The Ragni and Possenti (1996) system scores eight somatic regions where the pelage pattern was divided into several categories based on design, symmetry and colour. The system has been used to separate domestic, *lybica*, and *silvestris* types, but not their hybrids. We used a modification of this system with six of the original variables...
(gularis, occipitalis-cervicalis, lateralis, dorsalis, escapularis and caudalis), i.e., excluding those somatic regions that are most difficult to observe in field conditions (rhinarium and pinnae; see Ragni and Possenti 1996). For each variable, we scored each individual as either a wildcat (1) or domestic cat (2), and we computed a final average score for each individual. On the basis of this system, we used the average values to assign our 17 presumed wildcats as pertaining to typical wildcats or potential hybrids (average values scoring 1.5 or higher).

We also used the system proposed by Kitchener et al. (2005), which is based on 20 pelage characters and scoring discriminations: white on chin, stripes on cheek, dark spots underside, white on paw, white on flank, white on back, extent of dorsal line, shape of tail tip, colour of tail tip, distinctness of tail bands, alignment of tail bands, stripes on hind leg, bands encircling foreleg, tabby coat patterns, broken stripes on flanks and hindquarters, stripes on body, spots on flacks and hindquarters, stripes on napes, stripes on shoulders and colour on the back of ear. The character state for each measurement was recorded as 1, 2 or 3 following the observed range of morphological variation in each character. Although this recording was not associated to any specific cat type (i.e., intermediate scores are not by consequence hybrids), we assigned the value 1 to wildcats, the value 2 to hybrids and the value 3 to domestic cats for practical purposes (see Kitchener et al. 2005). For each individual, we obtained a total pelage score (hereafter, TPS) and a partial score (hereafter, 7PS) by summing the scores for each pelage character and for seven key characters only, respectively. Thus, TPS can range from 20 (“wildcat” extreme) to 60 (“domestic cat” extreme), whereas 7PS ranges from 7 (“wildcat” extreme) to 21 (“domestic cat” extreme). In the cat sample analysed by Kitchener et al. (2005), no presumed wildcat scored TPS > 26. However, the discriminatory power of TPS was limited, given that most presumed hybrids and some domestic cats also scored ≤ 26.

Instead, 7PS was proposed by these authors as diagnostic for the wildcat in their study area (Scotland): any cat with 7PS ≤ 9 and no scores of 3 for any of the seven characters should be regarded as a wildcat. Then, we compared for each individual the assignments obtained from each system and with those recorded by eye in the field.

The scores obtained from the two morphometric classification systems were used as data to perform cluster analyses and investigate the existence of any grouping pattern...
in the studied cat sample. In the case of the Kitchener et al. system, we used scores for the seven key characters (7PS) instead of all characters (TPS) owing to their higher discriminatory power; also, there were no missing scores regarding these key characters. Grouping was obtained using Euclidean distances and the unweighted pair group method with arithmetic mean (UPGMA). We compared the resultant groups with our by-eye field estimates and also compared the consistency of groups using the two classification systems. Cluster analyses were carried out using Statistica 6.0 software (StatSoft Inc. 2001).

Results

Sampling specimens

Our sample of presumed wildcats was divided into nine females (six adults and three subadults) and eight males (six adults and two subadults). Field identification indicated that WC6 (an adult female) and WC12 (a subadult male) could be hybrids because they had some anomalous coat traits such as irregular tail rings and white spots in some body areas. The remaining individuals were considered as showing pelage characteristics typical of genuine wildcats (see Figure 2 for a subsample of photographs).

Morphological discrimination based on the Ragni and Possenti system

We obtained four mean scores of unequivocal wildcats (value of index equal to 1), whereas most of the sample was composed of values between 1.17 and 1.33, except for WC11, which showed a mean score of 1.5 (intermediate between wildcats and domestic cats; Table 1). It is interesting to note that gularis, dorsalis and caudalis variables did not show any evidence of domestic traits.

Figure 2 Examples of photographs from some of the individual cats captured in Sierra Harana (Granada province). Dorsal and lateral views of WC1 (typical wildcat for all pelage characters; first line) and WC11 (considered hybrid according to the Ragni and Possenti classification system but wildcat according to the Kitchener et al. system; second line); in the third line, dorsal view and tail detail of WC6 (considered a hybrid in the first field estimation but typical wildcat after applying the two pelage classification systems; see text for further details).
Table 1  Character scores of individual cats (WC) to wildcat (1) or domestic phenotype (2) for the different somatic regions based on Ragni and Possenti (1996) classification system.

<table>
<thead>
<tr>
<th>Individual</th>
<th>Gularis</th>
<th>Occipitalis</th>
<th>Escapularis</th>
<th>Dorsalis</th>
<th>Lateralis</th>
<th>Caudalis</th>
<th>Mean assignment</th>
</tr>
</thead>
<tbody>
<tr>
<td>WC1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>WC2</td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1.33</td>
</tr>
<tr>
<td>WC3</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>WC4</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1.33</td>
</tr>
<tr>
<td>WC5</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1.17</td>
</tr>
<tr>
<td>WC6</td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1.33</td>
</tr>
<tr>
<td>WC7</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1.17</td>
</tr>
<tr>
<td>WC8</td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1.33</td>
</tr>
<tr>
<td>WC9</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1.17</td>
</tr>
<tr>
<td>WC10</td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1.33</td>
</tr>
<tr>
<td>WC11</td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1.50</td>
</tr>
<tr>
<td>WC12</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>WC13</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1.33</td>
</tr>
<tr>
<td>WC14</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1.17</td>
</tr>
<tr>
<td>WC15</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>WC16</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1.17</td>
</tr>
<tr>
<td>WC17</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1.33</td>
</tr>
</tbody>
</table>

In contrast, for the variables occipitalis and escapularis, we found nine individuals as showing domestic cat traits, and for lateralis we found four individuals with domestic cat traits. The cat WC11, the only individual that showed domestic traits for these three variables, was identified as a typical wildcat in the field. In contrast, WC12 was scored as a typical wildcat (value equal to one) despite its identification as a hybrid in the field (Table 1).

The UPGMA tree generated with these variables showed four groups (using a distance criterion of 1.2; Figure 3A). The individual WC17 was the most differentiated specimen with a mean score of 1.33. Group two included cats with mean scores of 1.33 (WC4 and WC13) and 1.5 (WC11). Group three was composed of individuals with mean scores of 1.17 (WC9, WC14 and WC16) and 1.33 (WC2, WC6, WC8 and WC10). The last group included all cats scored with values of 1 (typical wildcats: WC1, WC3, WC12 and WC15), although also individuals with some domestic traits in some variables (mean scores of 1.17: WC5 and WC7; Table 1).

**Morphological discrimination based on the Kitchener et al. system**

In general, TPS values were <26, except for individuals WC6 and WC11, and perhaps WC10. This suggests some departure, at least of these specimens, from the potential wildcat phenotype. Nevertheless, all cats met the diagnostic criteria to be unequivocally considered as wildcats, namely TPS values ≤9 and no scores of 3 for any of the seven key characters (Table 2). This result contrasts with the assignment of WC11 as hybrid based on the Ragni and Possenti system, as well as with the identification of WC6 and WC12 as possible hybrids during field recording. Twelve cats (70.5% of the total sample) showed values of 1 (genuine wildcat) for all the seven key pelage characters. From the 20 analysed variables, 11 showed that all cats were typical wildcats: white on chin, dark spots underside, white on paw, white on flank, white on back, extent of dorsal line, colour of tail tip, spots on flacks and hindquarters, stripes on napes, stripes on shoulders and colour on the back of ear (Table 2). Typical domestic traits were only found in stripes on cheek (WC11), stripes on hind leg (WC11), bands encircling foreleg (WC6, WC9, WC10 and WC11), tabby coat patterns (WC6) and stripes on body (WC11). Values indicating traits of potential hybrids were found in the shape of tail tip (WC10 and WC12), distinctness of tail bands (WC6 and WC11), alignment of tail bands (WC6 and WC11), tabby coat patterns (WC11 and WC13) and broken stripes on flanks and hindquarters (WC6, WC9, WC10 and WC11). The tabby coat patterns and stripes on body variables could not be adequately quantified in some individuals because of unsatisfactory photograph quality (see Table 2).

We detected three main groups of cats based on UPGMA cluster analysis (based on a distance value of 1.2; Figure 3B). The first group included the most
differentiated cats, i.e., WC6 and WC11. The second group was composed by WC10 and WC12. The third group was formed by all remaining cats. WC6 and WC11 showed 7PS equal to 9, which corresponds to wildcats with anomalous traits for two characters. These cats also showed the highest TPS values. Group two was formed of cats with 7PS scores of 8 and 9. Group three included all most genuine wildcats (7PS=7) plus one individual showing an anomalous trait for one character (7PS=8).

Figure 3 UPGMA cluster of the individual cats (WC) based on the scores obtained using the Ragni and Possenti (1996; all pelage characters included, panel A) and Kitchener et al. (2005; seven key pelage characters, 7PS, included, panel B) classification systems.
hybrid records, depending on the classification method that some biases are possible, with an exaggeration of traits may be used as a surrogate for genetic markers, but domestic cats. This result points out that morphological data, we may conclude that this anomalous individual was a relatively rare specimen placed at the intermediate classification based on previous classification systems also indicated that all individuals were clearly wildcats despite a large variability in coat pattern, colouration, or presence of some anomalous marks or traits (see Tables 2 and 3). The only exception was a cat that, in accordance with the classification system of Ragni and Possenti (1996), could be a hybrid. However, on the basis of microsatellite data, we may conclude that this anomalous individual was a wildcat from the genetic point of view, although it was a relatively rare specimen placed at the intermediate morpho-space between wildcats and their hybrids with domestic cats. This result points out that morphological traits may be used as a surrogate for genetic markers, but that some biases are possible, with an exaggeration of hybrid records, depending on the classification method used. This point contrasts with previous studies where admixture analysis allowed the identification of cryptic hybrids in Portugal, Italy or Bulgaria (Pierpaoli et al. 2003).

On the basis of our results, it is possible to suggest that researchers or managers using certain readily available and easy-to-use classification systems may detect more hybrids or rare individuals than when using microsatellite profiles (see also Eichholzer 2010, Nussberger 2012). Krüger et al. (2009) described hybrids with almost “pure” wildcat characters, and wildcats with more intermediate characters. The experience may be even worse when classification is performed in the field based on a limited set of morphological characters. The presence of a hybrid based on crude approaches or incomplete classification systems, which may cause problems in the potential control of hybrid populations.

**Discussion**

**Genetic introgression and concordance between morphological classification systems**

The analysis of admixture from microsatellites, performed by Oliveira et al. (2008a), indicated a lack of domestic alleles in our sample of wildcats, which appear to be of a clear “pure” genotype. Interestingly, morphological discrimination based on previous classification systems also indicated that all individuals were clear wildcats despite a large variability in coat pattern, colouration, or presence of some anomalous marks or traits (see Tables 2 and 3). The only exception was a cat that, in accordance with the classification system of Ragni and Possenti (1996), could be a hybrid. However, on the basis of microsatellite data, we may conclude that this anomalous individual was a wildcat from the genetic point of view, although it was a relatively rare specimen placed at the intermediate morpho-space between wildcats and their hybrids with domestic cats. This result points out that morphological traits may be used as a surrogate for genetic markers, but that some biases are possible, with an exaggeration of hybrid records, depending on the classification method used. This point contrasts with previous studies where admixture analysis allowed the identification of cryptic hybrids in Portugal, Italy or Bulgaria (Pierpaoli et al. 2003).

On the basis of our results, it is possible to suggest that researchers or managers using certain readily available and easy-to-use classification systems may detect more hybrids or rare individuals than when using microsatellite profiles (see also Eichholzer 2010, Nussberger 2012). Krüger et al. (2009) described hybrids with almost “pure” wildcat characters, and wildcats with more intermediate characters. The experience may be even worse when classification is performed in the field based on a limited set of morphological characters. The presence of a hybrid based on crude approaches or incomplete classification systems, which may cause problems in the potential control of hybrid populations.
Table 3  Summary table with all the results obtained for each individual cat (WC) from the two pelage systems and microsatellite analyses.

<table>
<thead>
<tr>
<th>Individual</th>
<th>Genetics</th>
<th>Pelage classification system</th>
</tr>
</thead>
<tbody>
<tr>
<td>WC1</td>
<td>Wildcat</td>
<td>1</td>
</tr>
<tr>
<td>WC2</td>
<td>Wildcat</td>
<td>1.33</td>
</tr>
<tr>
<td>WC3</td>
<td>Wildcat</td>
<td>1</td>
</tr>
<tr>
<td>WC4</td>
<td>Wildcat</td>
<td>1.33</td>
</tr>
<tr>
<td>WC5</td>
<td>Wildcat</td>
<td>1.17</td>
</tr>
<tr>
<td>WC6</td>
<td>Wildcat</td>
<td>1.33</td>
</tr>
<tr>
<td>WC7</td>
<td>Wildcat</td>
<td>1.17</td>
</tr>
<tr>
<td>WC8</td>
<td>Wildcat</td>
<td>1.33</td>
</tr>
<tr>
<td>WC9</td>
<td>Wildcat</td>
<td>1.17</td>
</tr>
<tr>
<td>WC10</td>
<td>Wildcat</td>
<td>1.33</td>
</tr>
<tr>
<td>WC11</td>
<td>Wildcat</td>
<td>1.50</td>
</tr>
<tr>
<td>WC12</td>
<td>Wildcat</td>
<td>1</td>
</tr>
<tr>
<td>WC13</td>
<td>Wildcat</td>
<td>1.33</td>
</tr>
<tr>
<td>WC14</td>
<td>Wildcat</td>
<td>1.17</td>
</tr>
<tr>
<td>WC15</td>
<td>Wildcat</td>
<td>1</td>
</tr>
<tr>
<td>WC16</td>
<td>Wildcat</td>
<td>1.17</td>
</tr>
<tr>
<td>WC17</td>
<td>Wildcat</td>
<td>1.33</td>
</tr>
</tbody>
</table>

The Ragni and Possenti scoring ranges between 1 (wildcat) and 2 (domestic cat). The Kitchener et al. scoring (7PS) ranges between 7 (“wildcat” extreme) and 21 (“domestic cat” extreme); according to these authors, scores ≤ 9 are diagnostic of wildcats.

Results from this study indicate that some traits are better than others in identifying wildcats based on their consistency in identifying all cats of our populations. On the basis of the Ragni and Possenti (1996) system, those traits using gularis, dorsalis and caudalis are better than those based on occipitalis, escapularis or lateralis. In the case of the Kitchener et al. (2005) morphological system, agreement with the genetic results was total. Accordingly, this system appears to provide better morphological discrimination of different cat types. The best traits of this system are the presence of white on chin, dark spots underside, white on paws, white on flank, white on back, extent of dorsal line, colour of tail tip, spots on flanks and hindquarters, stripes on napes, stripes on shoulders and colour on the back of ear. Stripes on napes and on shoulders were also highlighted by Krüger et al. (2009) as diagnostic characters of wildcats in their German sample, along with tail bands. However, tail bands were relatively variable in our sample, as found by Ragni and Possenti (1996) with Italian wildcats.

Conservation and management implications

Wildcats in remote areas with low human population densities in most of the Iberian Peninsula are probably scarcely introgressed (Oliveira et al. 2008a, see also Fernández et al. 1992) and so are of high conservation value for the species globally (Lozano et al. 2003). The conservation of these environments is crucial when considering increasing human pressure on large areas of Europe, and several authors have argued for the urgent preservation of these areas to conserve a pool of “pure” wildcats at a continental scale (Daniels et al. 1998, 2001, Lozano and Malo 2012). Because genetic introgression may not be so important in most of the wildcat populations of the Iberian Peninsula and other healthy populations (e.g., Pierpaoli et al. 2003, Lecis et al. 2006), conservation measures here need to be focused on other threats. Several studies in Spain have indicated the negative role that illegal persecution, which has eliminated wildcats from a number of localities (Virgós and Travaini 2005), can produce on wildcat conservation. Furthermore, the loss of forest patches in agricultural areas may cause the regional extirpation of the species in some areas (Virgós et al. 2002). Recently, it has been shown that depletion of the species’ main prey (rabbits and rodents) as a consequence of epizootics and the overabundance of some wild ungulates in hunting lands can reduce the abundance of wildcats (Lozano et al. 2007). All these negative impacts need to be mitigated or eliminated to improve the conservation of wildcats in the south-western part of their European range.

Nevertheless, although introgression with domestic cats appears to be of minor relevance in these areas, there is still a need to develop a standard monitoring programme for the presence of hybrids. Ideally, management of wild-living cat populations requires detailed genetic analyses of individuals that preclude any immediate action in the field, for example, when individuals are captured. Strikingly, however, we found that external examination of coat patterns could serve as a good surrogate of genetic markers, provided that careful attention is paid on key characters (see the Kitchener et al. 2005 classification method based on 7PS scores). Furthermore, eradication of domestic cats in the field seems not to be a necessary or urgent tool, at least in areas with large populations of wildcats, unless other threats reduce population densities leading to higher hybridisation probabilities. In addition, this can also lead to the elimination of pure wildcats, which may look like hybrids if not checked properly. Therefore, this action should be re-evaluated in situations where wildcat population density could be lower and hybridisation may indeed pose a true threat for the species. In this sense, we encourage further comparisons between genetic approaches and discrimination methods based on coat traits in other populations that show different or wider range of introgression levels and/
or phenotypic variability. However, until this information is available to the managers, we suggest that the money and people devoted to domestic cat eradication would be better directed to genetic and population monitoring and to eliminate other major threats to the species: illegal predator control, habitat destruction and depletion of main prey.

Acknowledgments: This work has been supported by the project “Carnivore mammals in the Granada province: distribution, status and management,” conducted by the Regional Ministry of Environment of the Junta de Andalucía. We are very grateful to M. Chirosa for sharing his knowledge and providing support; to R. López, J.F. Sánchez-Clemot and J. Bellido for their assistance during the field work; and the people of the Endangered Species Recovery Centre “El Blanqueo” (Regional Ministry of Environment of the Junta de Andalucía) for providing some wildcat samples. We also sincerely thank R. Oliveira (CTM-LNIV, Centro de Testagem Molecular, Laboratório Nacional de Investigação Veterinária, and CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, Universidade do Porto), E. Randi (Laboratory of Genetics, ISPRA, and INFS, Istituto Nazionale per la Fauna Selvatica) and co-workers for analysing microsatellites of our cat samples, and for allowing us to access raw data on cat genotypes. J. Muddeman kindly improved the English, and four anonymous referees provided interesting comments on earlier drafts of the manuscript. MM benefitted from a postdoctoral fellowship from the Spanish Ministry of Education (Plan Nacional de I+D+i 2008–2011) during the final stage of this article. All work conforms to the legal requirements of the regional government (Regional Ministry of Environment of the Junta de Andalucía, Spain).

Received February 13, 2013; accepted February 11, 2014; previously published online March 7, 2014

References


Lecis, R., M. Pierpaoli, S. Birò, L. Szemethy, B. Ragni, F. Vercillo and E. Randi. 2006. Bayesian analyses of admixture in wild and


