

THE EUROPEAN WILDCAT IN THE POLLINO NATIONAL PARK: WORK IN PROGRESS

IL GATTO SELVATICO NEL PARCO NAZIONALE DEL POLLINO: LAVORI IN CORSO

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Abstract. In Pollino National Park (PNP) is being held a study whose aim is to define, on a genetic base, the conservation status of the European Wildcat *Felis silvestris silvestris* Schreber 1777, and to check if there could be found, in short or long-term, risk evidences for its survival: proximity/superimposition of its home range with anthropic activities such as contacts with domestic cats (cross-breeding) or passages across driveways (road casualties) are highly probable. The study was carried out through a gradual and integrated approach of camera-traps and the collection of non-invasive biological samples (hair collecting) at first, and later capturing in-vivo specimen to be submitted to health control and genetic analysis. To all the captured individuals, belonging to the subspecies *silvestris*, was applied a radio collar GPS-GSM to define their home range composition.

Riassunto. Nel Parco Nazionale del Pollino è in corso uno studio volto a definire su base genetica lo stato di conservazione del gatto selvatico europeo *Felis silvestris silvestris* Schreber 1777, e a verificare la presenza di fattori di rischio a breve e lungo termine per la sua sopravvivenza, come la vicinanza/sovrapposizione del suo home range con attività antropiche dove sono più probabili il contatto con i domestici (ibridazione) ed il passaggio su strade carrabili (investimenti). Il lavoro è stato svolto adottando un approccio graduale ed integrato: associando la tecnica del fototrappolaggio prima alla raccolta di campioni biologici non invasivi (cattura del pelo), poi alla cattura di esemplari in vivo da sottoporre a controllo sanitario e analisi genetica. Gli individui catturati, risultati poi tutti appartenenti alla sottospecie *silvestris*, sono stati muniti di radiocollare GPS-GSM per definire la composizione dei loro home range.

INTRODUCTION

Between 2012 and 2017, as part of a monitoring project of the park's carnivore communities carried out through the camera-trapping technique, in 42 capture sites was taken an amount of 8600 pictures of wild species, 2800 of which were Carnivores. More than a half of these capture sites (N = 22) proved the presence of the European Wildcat *Felis silvestris silvestris* Schreber 1777 or of cats whose coat did not have evidence of pattern clearly leading back to domestic phenotype (RAGNI & POSSENTI 1996). Based on these results referred to the area of camera-trapping surveys, a first and partial map of the felid presence in the Park was developed (fig. 1). The following step was to check these data on a genetic base.

STUDY AREA

Pollino National Park is an area of about 192.000 ha between Basilicata and Calabria on a mainly mountainous landscape, with two most important massifs of Pollino and Orsomarso, and a third one, isolated in the North-West, the Mount Alpi. The Northern reliefs softly decline along the river

valleys of Sinni and Mercure-Lao, while the Southern landscape is rough and uneven with the deep valleys of Argentino, Lao and Raganello. Its surface at almost 60% is covered with wooded habitat (110.000 ha), 70.000 ha of them, approximately, are broadleaves.

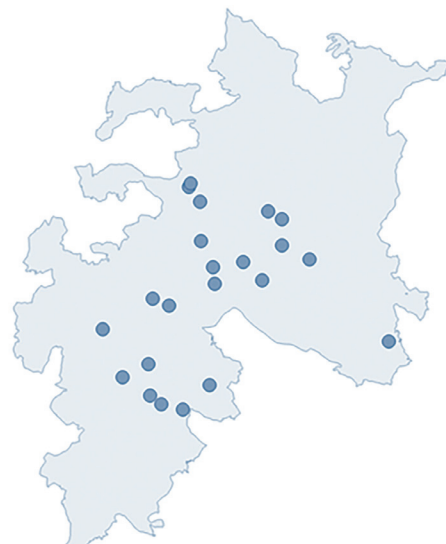


Fig. 1 - GSE presence sites of in the PNP (camera-trapping 2012-2017)

METHODS

Hair Traps

In the years 2017 and 2018, starting from the sites with more recent cat observations ($N = 9$), with the use of smelling spurs (70% hydroalcoholic extract of valerian) and food (pasta with sardines, fish and, above all, chicken), were tested three different devices for catching animal hair (Fig. 2)

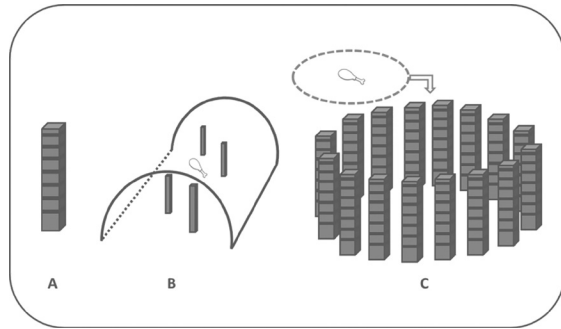


Fig. 2 - Devices for catching hair tested in the PNP in 2017-2018 – A: valerian hair trap, B: tunnel hair trap, C: wood air trap.

- A. *Valeriana hair trap*: wooden stake with velcro and 70% hydroalcoholic extract of valerian.
- B. *Tunnel hair trap*: tunnel cage with wooden stakes, velcro and food bait.
- C. *Wood hair trap*: open fence with velcro and central cage to protect food bait.

Camera-trapping and hair trap control, with substitution of SD card and restoration of food bait, was done on a weekly basis.

Live Trapping

In July 2018 the Minister of Environment granted to PNP the authorization to capture $n = 10$ samples of European Wildcat, as an exception to DPR 357/97, in order to monitor them through GPS/GSM for conservative purpose.

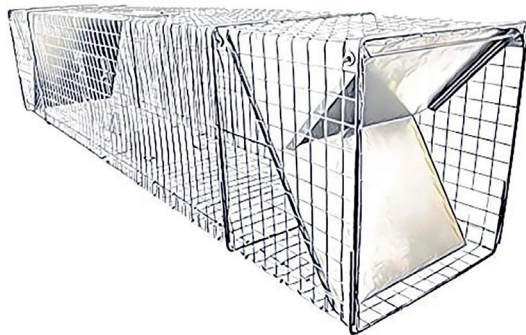


Fig. 3 - Double entry trap for small carnivores, 150 X 42 X 42, with tilting central platform and upper opening to release the animals.

Between January 2019 and February 2020, 15 capture stations in 10 different sites with tunnel traps 150 x 42 x 42 equipped with double entry tunnel traps 150 x 42 x 42 with tilting platform (fig. 3) associated to camera-traps were used (alternatively *Multipir 12*, *Cuddeback C1*, *IR plus BF 110*, *Scout Guard SG860C*). Counting on the strong territoriality of the species, it was decided to use, as an incentive, urine of domestic cat (either male or female), probably selective towards other carnivores.

The capture program, to be realized in autumn, winter and spring, avoiding the delivery and weaning period (May to September) was carried out by dividing the study area in two districts: Pollino and Orsomarso, managed by two different teams and sustained by a shared veterinarian. Once activated, the traps were controlled every day early in the morning, while the camera-traps were checked once a week or, exceptionally, to verify specific events (e.g., a closed but empty trap). The olfactory lures were changed at each control. In the event of capture, the general conditions of the individual (for example verifying the absence of wounds) were considered, then the cage was covered with a dark perforated cloth and the veterinarian was informed. At his arrival, the team had already executed a standard manipulation protocol, previously arranged:

1. The cats were moved from the capture cage to an enclosure cage with movable walls; here, by intramuscular injection, they were given anesthetic drugs. *The established anesthetic protocol of the first capture involved Tiletamina-Zolazepam 1mg/Kg + Medetomidina 0,05 mg/Kg + Atipamezolo (MAZZI 2008), while the others animals were given Ketamina 5 mg/Kg + Medetomidina 0,08 mg/Kg + Atipamezolo (WEST et al. 2014).*

2. When sedated, they were weighted and laid down in right lateral decubitus, covered their eyes to reduce visual stimulus and blocked their jaw with a lace to work safely. If their body temperature decreased, thermal towels, blankets and pocket hand warmers were used.

3. Their clinical conditions were evaluated, through teeth wearing sex and age were estimated, then biometric measurements were taken, the drawing-colour pattern observed and photographed and biological samples collected (a hematic sample with EDTA test-tube for genetic analysis, a hematic sample for serological analysis and a rectal swab and faeces for coprological analysis).

4. a satellite telemetry collar was applied (fig. 4).

5. If present, ectoparasites were collected and (for the two last captures) samples of hair from withers and abdomen for the cortisol dosage were taken.

6. At the end the cats were put back in the

enclosure cage and given them the Atipamezolo as antidote and the cage was covered with a shading cloth to reduce visual stimulus. The cats were observed at a distance enough to reduce stress, and then released when completely awake.

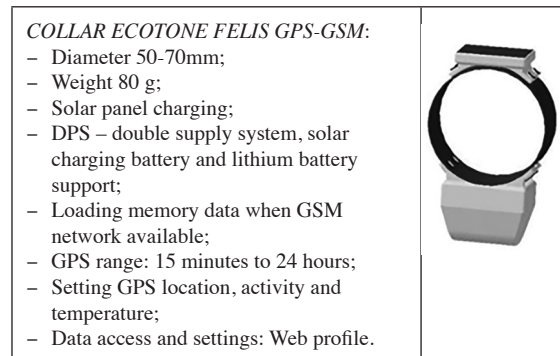


Fig. 4 - Ecotone satellite collar Felis GPS-GSM

RESULTS

Hair Trap

In the years 2017-2018, 730 pictures were taken in 9 different hair capture stations, 29 of them were cats (tab. 1) and only three hairs were picked up with *wood hair trap and food bait* (tab. 1).

Tab. 1 - picture catching with smelling spurs and feed in 2017-2018

Taxon	Capture with valerian	Capture with food bait
DOG	21	58
DOMESTIC CAT	0	2
WOLF	4	10
BADGER	17	26
FOX	100	270
SKUNK	0	3
MARTENS	0	15
BEECH MARTENS	19	156
WILDCAT	1	28
CARNIVORES	162	568

Live Trapping

During the first capture series, January to February 2019, due to heavy snowfalls, the trap nights were only 104. In the second series, carried out in two different periods, the first one from the 2nd to the 20th December 2019 and the second one from the 21st January to the 19th February 2020,

Tab. 2 - Trap Night January 2019 – February 2020

CAPTURE SITES	TRAP NIGHTS			TOT
	JANUARY-FEBRUARY 2019	DECEMBER 2019	JANUARY-FEBRUARY 2020	
POLLINO	97	60	61	218
GOREGHE	17	12	17	46
PRASTIO	18	0	0	18
SANTA ELENA	21	12	11	44
SANTA ELENA 2	11	0	0	11
VITO 3	15	0	0	15
MONTAGNA DI BASSO	15	12	11	38
CROCE PANTANA	0	12	11	23
VITO 1	0	12	11	23
ORSOMARSO	7	30	87	124
PIETRA CAMPANARA	0	6	18	24
TIMPONE FORNELLI	0	6	18	24
MARE PICCOLO	0	6	18	24
ROSSALE	0	6	18	24
FRASCINETO	7	0	0	7
NOVACCO	0	6	10	16
POVELLA	0	0	5	5
PNP	104	90	148	342

Tab. 3 - European wildcat specimens captured in the PNP between January 2019 and February 2020

Date	Capture site	Collar	Cat	Age	Weight
January 23, 2019	Santa Elena	FELT01	♂ VITO	2	4600
December 05, 2019	Vito 1	FELT02	♀ ROSARIA	4	2700
December 12, 2019	Montagna di Basso	FELT05	♀ MARIA	2	3150
January 31, 2020	Santa Elena	FELT03	♀ ERIKA	2	2700
February 19, 2020	Pietra Campanara	FELT04	♂ CIRO	6	2593

respectively 90 (60 Pollino and 30 Orsomarso) and 148 (61 Pollino and 87 Orsomarso) trap nights were realized.

In a total of 342 trap nights, three carnivore specimen were captured: beech marten N = 6, fox N = 8 and wildcat N = 5 (tab. 2 and 3)

Foxes and beech martens, after a quick control of their general health conditions, were released. Cats instead were subjected to the previously described manipulation protocol. Here below a summary of the examinations made and the proved circumstances in the different phases:

- ✓ **SEDATION:** in three out of five cases (*Rosaria*, *Erika*, *Ciro*) sedation was evident in 5-8 minutes from injection, with *onset* (stillness) in 10-15 minutes and *offset* (head lifting) in 40-45 minutes. Vito and Maria respectively were given one and two further doses of anesthetic to obtain a deeper sedation.
- ✓ **GENERAL CONDITION:** the captured samples were in good nourishing conditions, apart from *Ciro*, elder (5/6 year-old), thin and with dull coat. Maria, although she presented all young dental characters (reduced wear, crests on the canines and the shape of lower molar), had tartar on molars and pre-molars.
- ✓ **MANIPULATION:** each one took about 20-25 minutes for biometric measurement and coat pattern reliefs, blood sample and radio collar application.
- ✓ **BLOOD BIOLOGICAL SAMPLES:** each cat was taken of a blood sample for genetic analysis and was made a rectal swab, while only four of them were taken of a blood sample for serology. Faeces of three of them were found and picked up in their cages and the two last captured (*Erika* and *Ciro*) were taken some hairs from withers and abdomen.
- ✓ **RELEASE:** after being given the antidote, the cats gradually regained movement and coordination ability (8-12 minutes). They were observed

for about 15 minutes before being released in a suitable location close to the capture site.

The biometric measurements of the captured individuals (Fig. 5) were those reported in bibliography. All them were perfectly within the *range* described for the European wildcat (BOITANI *et al.* 2003). Noteworthy is the dimension of the first captured individual, a male of about 2 years old and 4,639 kg of weight. The coat drawing-colour pattern (Fig. 6) corresponded to the wild phenotype in all the samples that were captured (RAGNI & POSSENTI 1996).

**Fig. 5** - Biometrical reliefs

The genetic analysis carried out by ISPRA confirmed that all samples belong to the subspecies *silvestris*. The search for *Toxoplasma gondii* antibodies (ELISA method), gave positive results for all samples; the antigen of Feline Leukemia Virus (*FelV*) was only found in a sample, while none of them was positive to feline immunodeficiency and feline coronavirus (tab. 4).

The cat positive to *FelV* was Vito, a 2-year-old male, of 4,639 weight, with mucous membranes and lymph nodes in the norm, no breathing or heart problems observed, and no signs of other diseases.

On all captured specimen was deployed



Fig. 6 - Coat drawing-colour patterns: dorsal view

a GPS-GSM satellite collar (fig. 7) to allow the monitoring by the Park Conservative Area working team, through the *Ecotone* web server facilities. Below the first processing of the *home range* of three individuals is reported:

- ✓ Vito (FELT01) captured on January 23, 2019
- ✓ Rosaria (FELT02) captured on December 5, 2019
- ✓ Maria (FELT05) captured on December 12, 2019



Fig. 7 - Ecotone Felis GPS-GSM satellite collar

Tab. 4 - Serological and parasitological exams made by the IZS of Cosenza on samples of the captured wildcats.

	VITO Felt01	ROSARIA Felt02	MARIA Felt05	ERIKA Felt03	CIRO Felt04
Parasitological examination	Eimeria Oocysts sp. Ancylostoma eggs caninum e Toxocaracati.	Unsuitable sample	Negative to helminth eggs and oocyst coccidia	no faeces	no faeces
DNA research for Mycobacteriumavium subsp. Paratuberculosis through Real time PCR in faeces	Not observed	Not observed	Not observed	Not observed	Not observed
RNA Corona Virus Feline RNA research through Real Time RTPCR	Not observed	Not observed	Not observed	Not observed	Not observed
Salmonella research spp.(cultivation method)	absent	absent	absent		
DNA research for Giardia intestinalis and Cryptosporidium spp through Multiplex Real Time PCR				Not observed	
ELISA virological Leucemia Felina	positive	negative	no serum sample	negative	negative
ELISA direct Feline Immunodeficiency	negative	negative		negative	negative
Antibodies anti-toxoplasma gondii research through ELISA	positive	positive		positive	positive

Home range calculation

The *Kernel Density Estimation KDE* is used to calculate the animal species *home range* and gives a density estimation of land use. The result of KDE analysis is a *raster* map representing the most used part of land. In calculating KDE it is very important to choose the right *bandwidth or search radius*. There are different methods to estimate *home range*, starting from the available presence data: in this study the *Silverman* function (SILVERMAN 1986) in QGIS 3.14 environment was used.

Two density using levels were chosen: 95% (95° percentile) to define the *home range* and 50% (medium for density) to calculate the core area. The results of the analysis are referred, only for Vito, to the period between January 2019 and May 2020, and between January and May 2020 for Vito, Rosaria and Maria (Fig. 8, 9, 10 and 11).

The calculated HR (*KDE 95%*) are as follows: Vito 3770 *ha* (3.611 *ha* in total), Rosaria 871 *ha*, Maria 376 *ha*. About the Core Areas (*KDE 50%*): Vito 1.380 *ha* (1.187 *ha* in total), Rosaria 483 *ha*, Maria 305 *ha*.

The overlapping area of the total HR (Kernel 95% JAN-MAY 2020) for Vito and Rosaria is 4,8%

for Vito's HR and 20,7% for Rosaria's HR (Fig. 12). The overlapping area of the total HR (Kernel 95% JAN-MAY 2020) for Vito and Maria is 8,7% for Vito's HR and 87,2 for Maria's HR.

So, between January and May 2020 Vito's HR includes the 20,7% of Rosaria's HR, the minimum distance between them was 800 mt and it was registered on March, 23. In the same period Vito's HR includes the 87,2% of Maria's HR and the minimum distance between them was 196 mt and it was registered on February, 24 (Fig. 13).

From the environmental point of view, the prevailing types of land cover, taken from *Corine Land Cover 2012 level IV*, are the forests of deciduous mesophilic oaks and beeches, which are either for Vito and Rosaria the 59-60% of the total (Fig. 14 and 15).

Both animals also stand in rural areas formerly occupied by riparian forest, where can still be found remains of natural vegetation along ditches and rows; these were often used by the male to move and rest.

For the other female (Maria) the forests represent 100% of her HR (Fig. 16).

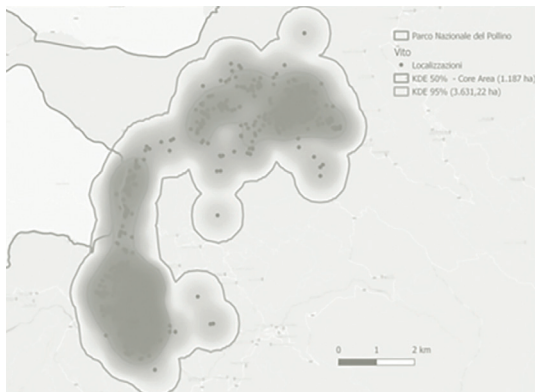


Fig. 8 - Home range (KDE 95%) and Core area (KDE 50%) for Vito from January 23 to, 2019 to May 31, 2020

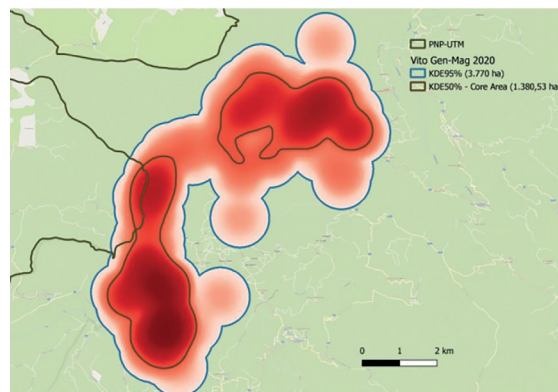


Fig. 9 - Home range (KDE 95%) and Core area (KDE 50%) for Vito January to May 2020

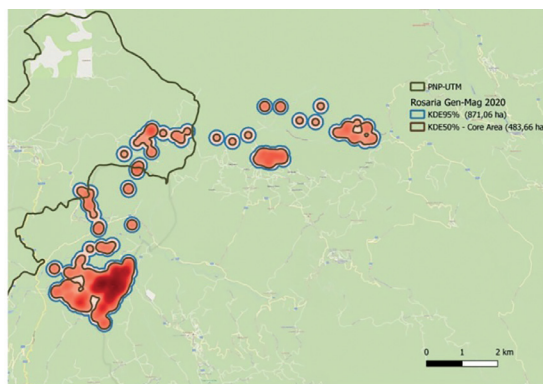


Fig. 10 - Home range (KDE 95%) and Core area (KDE 50%) for Rosaria January to May 2020

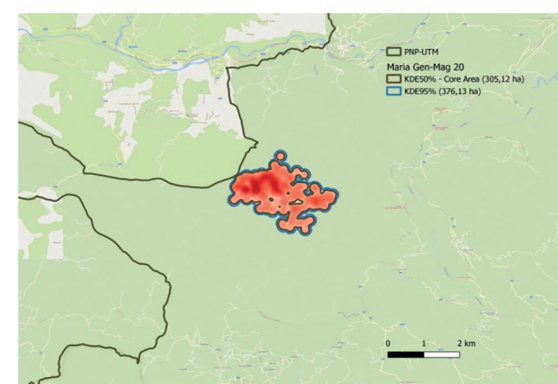


Fig. 11 - Home range (KDE 95%) and Core area (KDE 50%) for Maria January to May 2020

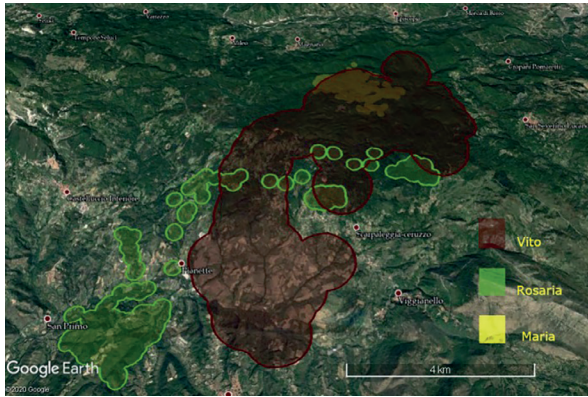


Fig. 12 - Spatial overlapping FELT01 (Vito) - FELT02 (Rosaria) and FELT 05 (Maria) January to May 2020

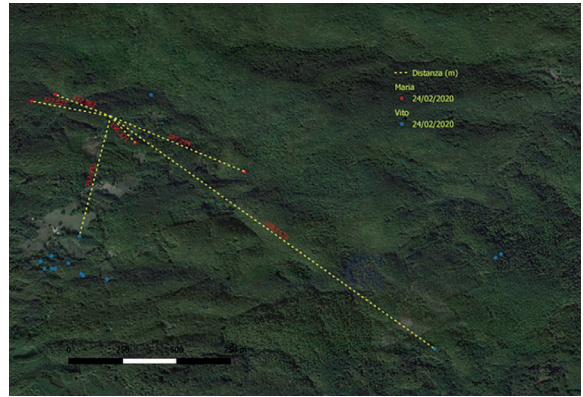


Fig. 13 - Minimum distance between FELT01 (Vito) and FELT05 (Maria) February, 24 2020

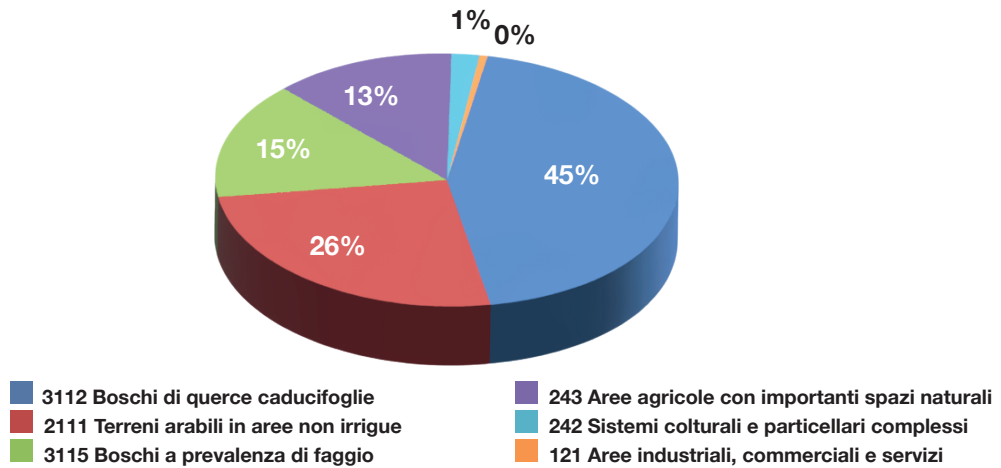


Fig. 14 - KDE environmental characteristics 95% FELT01 (Vito) - Land use categories *Corine Land Cover 2012 IV level*

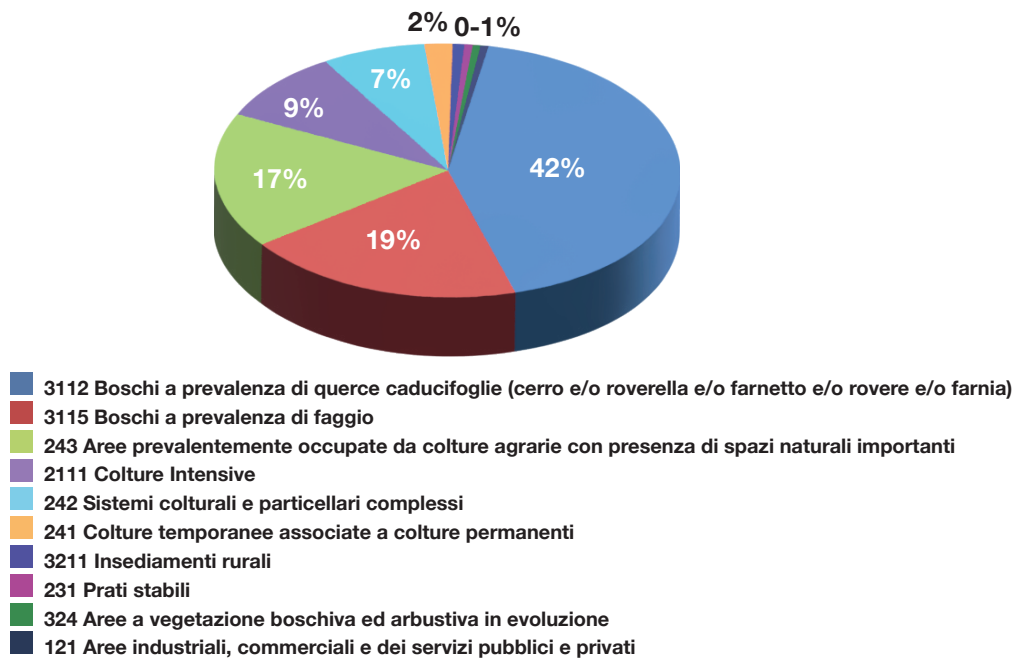


Fig. 15 - KDE environmental characteristics 95% FELT02 (Rosaria) - Land use categories *Corine Land Cover 2012 IV level*



Fig. 1 - KDE environmental characteristics 95% FELT05 (Maria) - Land use categories *Corine Land Cover 2012 IV level*

DISCUSSION

The trials on the techniques for collecting hair suggested that the most efficient capture device was the *wood hair trap* used with food bait. On the contrary, valerian as an attractant resulted inefficient. On a total of 29 wildcat observations, only one was made with the use of this incentive, and the picture shows an absolutely casual passage. More generally, from a costs/benefits point of view, the hair capture showed not to be very convenient.

Instead, the traditional capture *in-vivo* with cassette traps and smelling incentive (urine of male or female domestic cat) was very efficient, with a Capture Index of 0,015 in 342 trap nights, which seems to be very encouraging if compared with the 0,029 obtained in 1995 in the Maremma Regional Park for 700 trap nights, with food incentive (live breeding quails).

The use of the enclosure cage with movable wall allowed to immobilize the cats very quickly, to give them drugs safely, and to reduce the stress exposition time. The capture protocol proved to be always very efficient, in fact no wildcat had problems resulting from narcosis or manipulation (BIZZARRI *et al.*, 2010).

During the first weeks following the release of Vito (FELT01) there was a lower number of localizations (fixes) than expected. It was probably due to the extensive vegetation cover of the areas frequented by the wildcat, that prevented a suitable

power supply to the battery photovoltaic charging process.

In particular situations the battery charge level reaches a critical point: the GPS receiver needs a quite high power and the poor satellite signal coverage in ravine and hiding places quickly strains the small devices batteries.

Following these observations, the Ecotone Company made some changes to other satellite collars that were not in use; by modifying the software implementation in the managing of the “motion” mode the waste of energy in its search for the GPS signal was reduced, stopping the search for satellites from the receiver when animals activity is scarce or completely absent.

However, in spite of such changes, the satellite collars gave a lower number of fixes than expected.

A first elaboration of the home ranges, only related to three samples and to a given period, suggests that the broadleaved forest is the predominant habitat (Vito and Rosaria) or the unique one (Maria), but also a regular use of rural areas for the first two animals (about 40% of total HR), which sometimes move towards isolated houses, using parts of riparian forest as a shelter or passageway. Between January and May 2020 the two females HR have wide spatial overlapping as the male, but they are not mutual. In particular it is very interesting the space/time overlapping between Vito and Maria in the reproductive period (February 24, 2020) with a minimum distance of 196 mt.

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