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Comparison of track and direct observation estimations for assessing abundance of the Eurasian otter, *Lutra lutra*

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Abstract. The population density of the Eurasian otter, as in many other carnivores, is difficult to estimate in the wild. Spraint (otter scat) counts are usually used as an indirect indicator of abundance but its reliability is poorly known. In this work two methods of estimation (direct observations, DO, and track sampling, MNT) are compared in Central Spain. A new approach is applied to correct inherent biases to track sampling. Furthermore, the influence of sample size on MNT estimations was tested. The results indicate a highly significant relationship between the estimations of abundance derived from DO and MNT methods, although MNT could underestimate the density of otters when it is under 0.01 otters/km. The application of the new track sampling method could result in a successful reduction or removal of the biases. On the basis of current knowledge, it is argued that both MNT and DO could provide a realistic picture of the otter populations and facilitate their estimation and monitoring with sufficient reliability.

Key words: central Spain, Mustelidae, population abundance, surveying methods

Introduction

The population size is a central parameter in ecology, but can be difficult to measure in the field. Therefore it is frequently replaced by estimations of density or even abundance. For carnivores in particular, these parameters are difficult to measure, due to their generally cryptic or nocturnal habits (Long et al. 2008). As a consequence, abundance estimations rely upon indirect surveys (e.g. sign counts), whose accuracy is doubtful (Long et al. 2008). The biological features of the otter, *Lutra lutra* (i.e. Kruuk 1995, 2006) make the species prone to decline and thus of general conservation interest.

Difficulty in direct observation and generally crepuscular or nocturnal activity has led to the development of the "Standard Otter Surveys" which consist in the search for spraints (otter faeces) in transects along the banks of water bodies (Mason & Macdonald 1986, Reuther et al. 2000, Chanin 2003). Both the number of positive sites and the abundance of spraints in a surveyed section have been considered to reveal the abundance of the otter population in an area (Mason & Macdonald 1986, Reuther et al. 2000, Chanin 2003). Nevertheless, mainly indirect evidence for such relationship exists and the validity of the method for population assessment is uncertain (Kruuk et al. 1986).

The recent rise of molecular scatology has built a more robust framework for the use of spraints in estimating population density (Chanin 2003, Ruiz-González et al. 2008, Hájková et al. 2009), but some of the biases inherent in spraint sampling remain, as well as high time and financial costs of genetic analysis.

Alternatively, tracks have been successfully used for evaluating the number of otters inhabiting an area, although the results depend strongly on the environmental conditions (Mercier & Fried 2004, Ottino & Giller 2004, Sulkava 2007, Hájková et al. 2009). Therefore, in northern areas of Europe, snow-tracking is widely used (Sulkava 2007, Hájková et al. 2009), whereas in southern areas tracks are sought in muddy and sandy soils (Ruiz-Olmo et al. 2001, Ottino & Giller 2004). The length of each track is measured and the number of individuals is estimated by the different track lengths (Ruiz-Olmo et al. 2001, Mercier

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& Fried 2004). Some authors have expressed their concern about this method because of several biases causing potential underestimations (Chanin 2003, Hájková et al. 2009). Furthermore, Ruiz-Olmo (1995) proposed the use of direct counts of otters based on their daylight activity in some Mediterranean areas. Only one field test has been reported regarding the combined effectiveness of these methods (Ruiz-Olmo et al. 2001). It demonstrated the efficiency of both techniques for estimating population, detecting practically all the individuals present in an area. The main aim of this work was to compare estimations of otter density based on track sampling and direct observations. Moreover, some improvements were made on the track sampling method.

Study Area

The study was carried out in the province of Salamanca (Central Spain) from the winter of 2006 to autumn 2009. Seven areas were selected in order to achieve a representative sample of the habitats (Fig. 1A). Each site was surveyed by no more than five people and estimations were independently repeated several times in five of these sites. The study areas were (a more complete description of the sites is presented in Table 1 and their locations in Fig. 1A):

1. Villagonzalo (the River Tormes): Lat: 40°54'0.43" N Lon: 5°28'48.03" W. This included only the River Tormes. Abundance was estimated only in the summer of 2009. Mean width of the stretch sampled was about 100 m.
2. Lagunas del Cristo (the River Yeltes): Lat: 40°39'57" N Lon: 6°75'4.74" W. These included the lakes of Cristo and La Cervera, a stretch of the River Yeltes and some clay pits near this river. Four samplings were carried out in this area, in the winter of 2006, autumns of 2007 and 2008 and summer of 2009. Mean width of the stretch sampled was about 150 m.
3. Riolobos area (the River Tormes and the River Guareña): Lat: 41°1'30.84" N Lon: 5°24'30.28" W. This included the Riolobos reservoir, the Aldearrubia reservoir and the wetlands near the River Merdero. Four samplings were carried out in this area, in the winter of 2006, autumn and winter of 2007 and winter of 2008. Mean width of the stretch sampled was about 200 m.
4. Navamuño (the River Cuerpo de Hombre): Lat: 40°21'40.54" N Lon: 5°45'36.48" W. This included only the Navamuño reservoir. Three samplings were carried out in this area, in the summer, autumn and winter of 2009. Mean width of the stretch sampled was about 200 m.
5. Juzbado (the River Tormes): Lat: 41°4'48.73" N Lon: 5°52'46.94" W. This included only the River Tormes. Three samplings were carried out in this area, in the winters of 2007 and 2008 and spring of 2009. Mean width of the stretch sampled was about 90 m.
6. Monleras (the River Tormes): Lat: 41°11'6.86" N Lon: 6°14'41.89" W. This included the Almendra reservoir (the River Tormes) and the Villar stream. Two samplings were carried out in this area, in the autumn of 2008 and spring of 2009. Mean width of the stretch sampled was about 70 m.
7. Ledesma (the River Tormes): Lat: 41°5'55.68" N Lon: 6°0'56.71" W. This included the River Tormes and the Cañedo stream. Two samplings were carried out in this area, in the winters of 2007 and 2008. Mean width of the stretch sampled was about 60 m.

Material and Methods

Direct observations

The basic design was in accordance with Ruiz-Olmo (1995). Basically, it consisted in monitoring the water body for otters for approximately one hour. This was performed by experienced observers at sunset and sunrise using binoculars and telescopes. At the study sites, observers were positioned in such a way that the entire surface of the water body could be monitored at the same time and without the overlapping of observation areas. In case of overlap only one of the observers monitored this area. Observations (vigils) were carried out from dawn until one hour later and from one hour before until dusk, thus increasing the probability of otters being active (Ruiz-Olmo 1995). To illustrate the procedure of the DO technique an example is presented in Fig. 1B.

Observers recorded the contacts with otters, the time of the observation and their behaviour. Taking into account the distance between successive contacts, the speed of the otters and the hour of the observation, the number of otters present in the area was estimated. This is expressed as the minimum number of different otters observed per kilometre sampled (DO).

Track sampling

The surface covered by direct observations was searched for fresh otter tracks by the same people. This was done just before the vigils (in the case that DO estimation was from a sunset vigil) or immediately after the vigil (in the case that DO was from sunrise). When a track was found, its length was measured from the pad to the claws (Ruiz-Olmo et al. 2001, Mercier & Fried 2004, Ottino & Giller 2004). The number of otters at the site was estimated from track length differences.

Only fresh tracks were used in this study, as the use of older ones may lead to overestimation (Ruiz-Olmo et al. 2001). The freshness of the tracks was assessed

Table 1. *Ecological and sampling attributes of the study sites. Estimations obtained from both DO and MNT as well as the length surveyed is also showed (see text).*

Ecological and sampling attributes		Riolobos	Villagonzalo	Cristo's lakes	Navamuño	Ledesma	Monleras	Juzbado
Altitude (masl)		830	800	873	1280	780	796	790
Trophic category		Hypertrophic	Mesotrophic	Eutrophic	Oligotrophic	Eutrophic	Eutrophic	Mesotrophic
Morphologic characteristics[1]		Entrenched in moderate gradients, unstable channels	Low gradient, meandering rivers	Braided and anastomosed rivers	Mountain reservoir	Low gradient, meandering rivers	Entrenched on moderate gradients, unstable channels	Low gradient, meandering rivers
Bed morphology[1]		Cohesive clay	Bars and islands	Bedrock and cohesive clay	Clay	Bars and islands	Cohesive clay	Bars and islands
Riparian vegetation[1]		Aquatic plant formations or without vegetation (muddy and sandy shores)	Woody vegetation communities Bank shrub and herbaceous units Aquatic plant formations	Woody vegetation communities Bank shrub and herbaceous units Aquatic plant formations	Without vegetation (muddy and sandy shores)	Woody vegetation communities Bank shrub and herbaceous units Aquatic plant formations	Bank shrub and herbaceous units Aquatic plant formations	Woody vegetation communities Bank shrub and herbaceous units Aquatic plant formations
Length of the sampled section in kilometers (sampled both in DO and in MNT)		10 / 11 / 15 /15	10	11 / 11 / 10.5 / 9	11 / 11 / 9	14 / 10	14	6 / 7
Population estimation (otters/km)	DO	0.20 / 0.18 / 0.20 / 0.20	0.1	0.18 / 0.09 / 0.1 / 0.05	0.27 / 0.18 / 0.22	0.14 / 0.1	0.14	0.3 / 0.28
	MNT	0.21 / 0.18 / 0.2 / 0.2	0.1	0.18 / 0.1 / 0.13 / 0.05	0.27 / 0.18 / 0.22	0.14 / 0.1	0.14	0.33 / 0.33

[1] Classified following the guidelines for Spanish rivers proposed by González del Tanago & García de Jalón (2004).

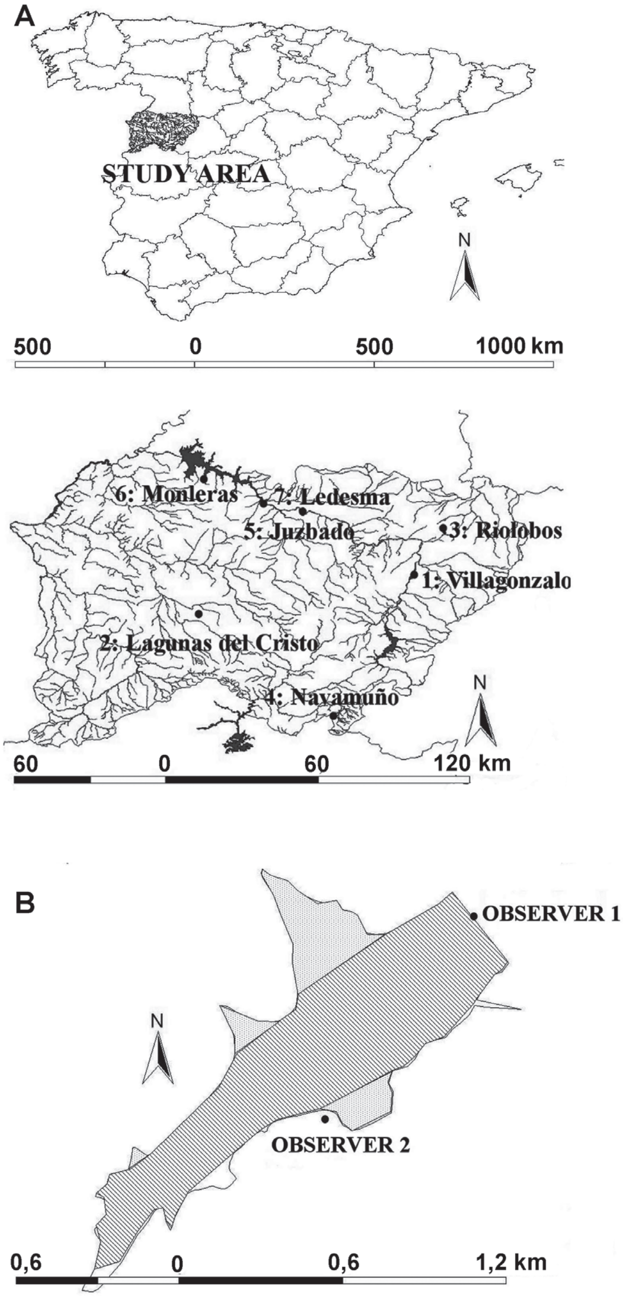

Fig. 1. *A) Geographical location of the study areas in central Spain. B) Example of the design of DO method employed to estimate otter populations in the Navamuño reservoir. Two observers (O1 and O2) monitored at the same time the lined (O1) and the dotted surface (O2).*

based on the wetness of the footprints, if these are in newly deposited sand or mud (heavily dependent of whether the previous day has had any rainfall), and given that track sampling was even done just after or before direct observations. So, we are confident that we only take into account fresh footprints.

Track sampling has some biases for the estimation (Chanin 2003, Mercier & Fried 2004, Long et al. 2008, Hájková et al. 2009). Track length depends on the substrate (longer tracks in mud than sand), differences in the measurements due to different observers, the sample size (number of tracks found), which depend on the availability of good substrates for track printing and the distance covered searching for tracks.

Due to these potential drawbacks a new approach for a more accurate use of this method was developed in this work, while the existence of the rest of the biases was tested after the application of the new procedure.

Each otter track found was photographed with a known size scale with a digital camera (8 megapixels). Each track picture was measured to the nearest millimetre with ImageJ free software (http://rsbweb.nih.gov/ij/). This procedure allows the elimination of the errors of different observers.

The first potential bias factor described here was tested. In the Riolobos reservoir, whose banks are both sandy and muddy and without vegetation, eleven trails (groups of tracks of the same otter) were found to continue both in the sand and in the mud. Forepaw prints were photographed and measured, as described above, in the two substrates. The tracks in the sand were 0.92 ± 0.01 (mean ± SD) times smaller than those in the mud and this factor was used to correct the substrate-based differences. Length in sand seems to be more representative of the real size of the tracks.

The number of otters present in the area was estimated after measuring forepaw length in ImageJ and applying the 0.92 correction factor. Individuals were differentiated by the corrected length of their forepaw. The length criterion in Ottino & Giller (2004) was employed to estimate the minimum number of individuals per km sampled (MNT): < 5.0 cm (cub/young), 6.0–7.0 (adult female), > 7.0 cm (adult male). See Ruiz-Olmo et al. (2001) and Ottino & Giller (2004) for further details.

The search for tracks was performed by trained and experienced people, while the computer-processing was performed by one person PG-D in order to eliminate inter-personal differences.

Statistical analysis

The existence of a positive association between DO and MNT numbers was assayed with a non-parametric Spearman's rank test. A Least Squares Linear Regression (LSR) was used to discover whether there was a threshold of population abundance that remains undetected when using track sampling but detected by direct observations. The influence of the number of tracks found during the surveys on the final population estimations was analysed using a Spearman's rank test. Equally, the effects of the length of the stretch searched for footprints on the density values obtained were tested in the same way. Statistical calculations were performed with MYSTAT 12.0 and G-Stat statistical packages.

Results

Seventeen otter abundance estimations were available (n = 17) both for MNT and DO methods. Similar population estimates were obtained with DO (median ± SD: 0.14 ± 0.07 otters/km, minimum = 0.05, maximum = 0.33, n = 17) and MNT (0.14 ± 0.08 otters/km, minimum = 0.05, maximum = 0.30, n = 17). This fact is also demonstrated by the high significant positive correlation between the two abundance estimators (Spearman's rank, $R_S = 0.98$, *t*-test, $t = 19.77$, $p = 0.000$, n = 17; Fig. 2). Results of the abundance estimations coming from the fieldwork in each locality are in Table 1, and their relationship is shown in Fig. 2.

Least Squares Regression analysis for DO over MNT results in an intercept value of 0.01 (DO = 0.01 + 0.88 MNT; see also trends in Fig. 2), meaning that abundances below 0.01 otters/km are detected by DO, but not by the MNT method. The slope of the LSR indicates a similar parallel increase in the DO and MNT otter numbers.

Regarding the methodological deviations in the MNT method involving the sample size, there is no significant association between the length surveyed and the abundance estimation (Spearman's rank, $R_S = -0.11$, *t*-test, $t = -0.43$, $p = 0.68$, n = 17) nor between the number of tracks measured along a river stretch and the abundance estimation (Spearman's rank, $R_S = 0.41$, *t*-test, $t = 1.75$, $p = 0.10$, n = 17).

Discussion

The use of different techniques for monitoring otter populations is an important issue of debate. Spraint-based methods have been demonstrated to include numerous deviations, so it is interesting to test other potential methods and their reliability. MNT has been argued to be a useful complementary technique by some authors (Ruiz-Olmo et al. 2001, Mercier & Fried 2004, Sulkava 2007).

Direct observation is among the best methods for estimating the abundance of otters (Kruuk 1995, 2006, Ruiz-Olmo 1995), but depends upon the existence of a diurnal activity pattern. In the Mediterranean area the snow cover is generally scarce impeding a wide use of snow-tracking and thus depends upon the use of mud or sand substrates. Problems with the use of tracks as an estimator of population size are common (Chanin 2003). We tested a quick and easy method for correcting these biases, taking a photo of each track, processing it with image software and applying a correction factor considering the substrate where the track was found. The results indicate no relationship of the MNT to the sample size, supporting the idea that the biases could be eliminated with this approach. Despite no relationship with the length of the river stretch sampled, according to Ruiz-Olmo (1995), it seems that lengths of at least 10 km are recommendable in order to obtain reliable data.

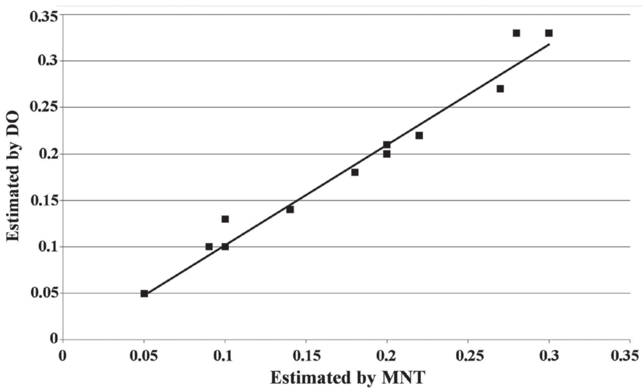

Fig. 2. Relationship between estimated abundance (otters/km) as assessed by DO and MNT. The Least Squares Regression line is also shown. LSR: DO = 0.01 + 0.88 MNT).

The studies carried out in populations of known size (Ruiz-Olmo et al. 2001) indicate that nearly all the populations can be appropriately counted using the DO method, although Chanin (2003) expresses some concern about the validity of the results, but without providing any evidence against this method or the MNT. It should be noted that Ruiz-Olmo et al. (2001) found that visual estimations could be a problematic method whenever abundance values were < 0.1 otters/km. This seems not to be our case, as sometimes we get numbers under such value (Table 1), albeit most of our estimations are above 0.1 otters/km (Table 1). For instance, this suggests that DO perform better than previously demonstrated.

The number of otters per kilometre sampled (0.14 ± 0.07 otters/km for DO and 0.14 ± 0.08 otters/km for MNT) is in accordance with the density estimations made in other areas of Europe (Ruiz-Olmo 1995, Kruuk 1995, 2006, Sulkava 2007, Ruiz-González et al. 2008, Hájková et al. 2009).

The highly significant relationship of the abundance estimated through DO and the MNT methods is in accordance with the positive association reported by Ruiz-Olmo et al. (2001) in north-eastern Spain. Moreover, the relationship indicates that the estimations do not vary under a range of environmental conditions.

Similarly, Ruiz-González et al. (2008) found that DO, MNT and molecular scatology at the same site result in similar densities. Assuming that "molecular scatology" (Chanin 2003) and the DO method (Ruiz-Olmo et al. 2001) yield close approximations to the real situation, the MNT method could be a cost-effective way to obtain reliable estimations.

The evidence suggests that at least the minimum number of otters inhabiting an area could be evaluated using DO and MNT method. Nonetheless it would be advisable to check the validity of these methods in different areas and habitats.

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