

**Using species detectability to infer distribution, habitat use and absence of a cryptic species: the smooth snake (*Coronella austriaca*) in Saxon Transylvania**

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**Summary**

Ecologists often use binary data to estimate the habitat use, turnover and distribution of species and populations. The basic assumption in these studies is that the individuals are perfectly detected. However, only a few studies account for the detectability of the target organisms in various habitats. Not accounting for detection probability may bias the results to an unknown degree. The problem of detection probability is especially important in cryptic species since their presence may be easily overlooked by researchers (leading to false absences). In this paper we present the estimated habitat occupancy, detection probability and the minimum visits needed to infer the absence of species, using the smooth snake as model organism. This species is commonly present in the ecological literature on European snakes and is intensively inventoried in Romania. Our study was conducted in 2008, in seven landscapes (56 sites) where the presence of the smooth snake was previously proved. The detection probability varied from 0.09 to 0.37 whereas the minimum number of visits necessary to assert with 95% confidence that smooth snake is truly absent varied from 6 (stones) to 32 (orchards). Different implications of not accounting for detection probability (and associated statistics) are mentioned.

**Keywords:** detection probability, cryptic species, smooth snake, Romania.

## Introduction

The basic level of determining the habitat use and distribution of species and populations is to gather presence-absence data. It is not surprising therefore, that early mathematical models in spatial and community ecology, such as island biogeography (MacArthur and Wilson 1986) and metapopulation theory (Lewins 1970, Hanski 1998) were based on presence-absence data to estimate persistence, extinction and (re)colonization of species and populations. The issue of presence-absence data is present also in the relatively new landscape ecological approaches. In this approach, logistic regression (that is based on the binary data: presence-absence [1, 0]) is often used to identify the most important habitat and landscape parameters that positively or negatively influence habitat occupancy (Hartel et al. 2008). A key assumption in these approaches using presence-absence data is that, if locally present, populations and species are perfectly detected. In this assumption, non-detection is interpreted as absence, and the possibility of “false absences” (i.e. the situation when the species is present but goes undetected) is generally ignored. However, as recent studies show, the assumption of the perfect detectability of species is very rarely satisfied in nature. Although field ecologists are aware of the problem (Simberloff 1976, Preston 1979) and use repeated counts, favorable weather conditions and standardized routes to increase the probability of finding species in habitats, the detection probability is still relatively rarely quantified in ecological field studies (but see Bailey et al. 2004, Kéry 2002, Kéry and Schmid 2004, Pellet and Schmidt 2005, Schmidt 2004, 2005). If detection probability is not accounted for, the degree of accuracy and bias remain unknown. A recent study evaluated the factors affecting the habitat use of an amphibian using conventional logistic regression and accounting for the detection probability and demonstrated that not accounting for detection probability may under- or overestimate the importance of some factors (Mazerolle et al. 2005).

Another, currently less emphasized aspect of field studies dealing with the spatial distribution of organisms is the problem of “absences” (MacKenzie 2005a). If no individuals of a given species are found in a studied area or habitat, it is usually considered that the species is absent or extinct. What would be the implications of false absences? As Moilanen (2002) showed, false absences may strongly bias all models that deal with population and

metapopulation phenomena (extinction, colonization, dispersal range etc.). Moreover, the absence of certain species is important in environmental impact assessments. If some species of interest (for example a legally protected one) are absent from a certain area, the permissions for interventions (buildings, infrastructure etc.) in the natural environment are easily obtained. But if the species were falsely identified as absent, the consequences of these interventions may be catastrophic.

In this study we present an attempt to determine the habitat use, detection probability and the minimum number of surveys required to infer the absence of the smooth snake (*Coronella austriaca*) in a Transylvanian rural landscape. The biology and ecology of the smooth snake is well studied in Europe (see for example Spellerberg and Phelps 1977, Goddard 1984, Gent and Spellerberg 1993, Luiselli and Capizzi 1997, Reading 1997, Drobenkov 2000, Kéry 2002).

Ecological studies on the smooth snake have not, to our knowledge, been carried out in Romania. The species is present in a large number of herpetofaunistic reports from different areas of this country. These studies consist of making lists of human settlements, and highlighting those localities where the different reptile species, including smooth snake were found. The results of the first large scale surveys regarding the herpetofauna of Transylvania (Ghira et al. 2002) summarize the results gathered for 1046 localities and mention the presence of smooth snake only in 212 of them (20.26%). The data collection in this study was made partly by collecting data from local people and partly by direct observations of the researchers involved. Although personal experience suggests that local people can recognize this species, the data collected does not account for search effort. Thus, it is very possible that the smooth snake was directly identified by researchers in an even smaller number of localities. The reports that followed this publication are numerous, and aim to “fill” areas previously uncovered in Transylvania or cover new areas in other regions of Romania. The percentage of localities where the smooth snake was identified in the field seems to be quite low even when studied at a local geographic scale: 10% (Covaciu-Marcov et al. 2006a), 9% (Strugariu et al. 2006), 6% (Covaciu-Markov et al. 2006b), 3% (Covaciu-Marcov 2005), 8% (Ghiurca et al. 2005), 34% (Strugariu et al. 2008) but see 50% in Sos (2007) who intensively surveyed a relatively small area and a smaller number of sites. False absences and detection probabilities were not considered in these reports.

Although previous herpetofaunistic reports used also multiple visits on each site, the number of visits per site was not presented nor controlled. An important detail that should be mentioned is that many of the previous herpetofaunistic surveys recognized that an important source of data (often used “crucial role” suggesting at least 50%) was dead specimens (killed by humans in various ways), and this also may bias their results.

The objectives of this study are:

- to estimate the proportion of site occupancy of the smooth snake,
- to determine the detection probability of this species,
- to estimate the minimum number of visits needed to conclude that the smooth snake is absent from different habitat types.

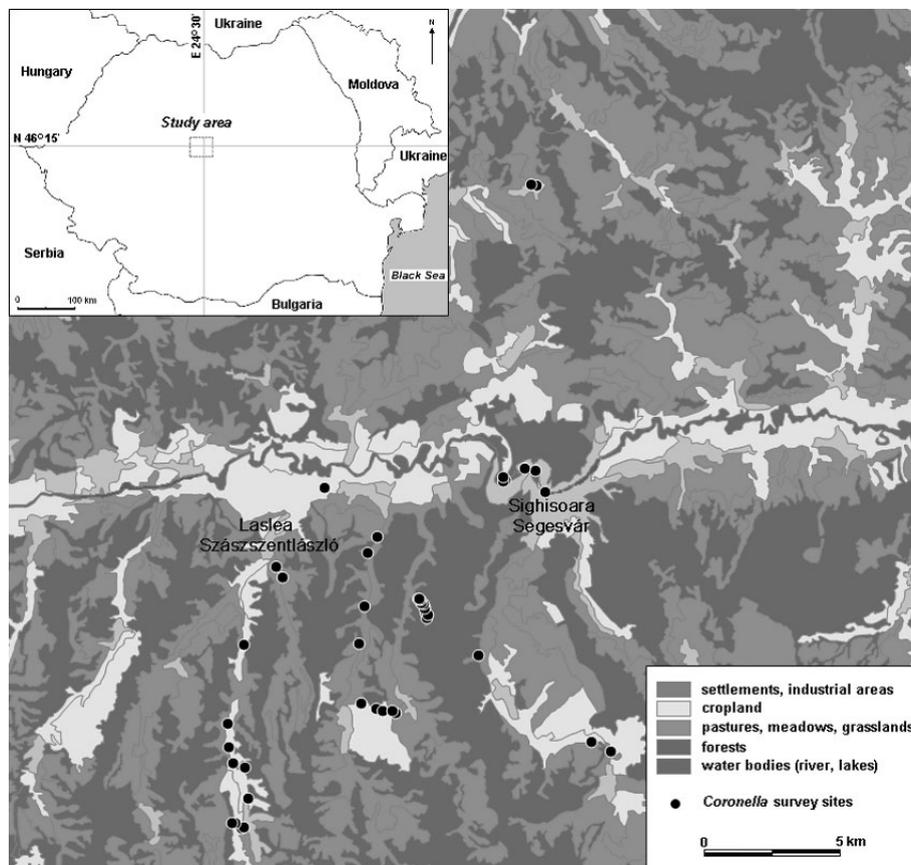
## **Material and methods**

### **Study area and data collection**

The study area is located in the middle section of the Târnava Mare Valley, Romania and is representative for the Continental biogeographic region (Figure 1). The landscape consists of a mixture of deciduous forests, grasslands and patchy use of arable lands; land uses that have been maintained from the Middle Ages to the present day. The climate is continental. The altitude range varies between 340–450 m above sea level.

Data on the presence of the smooth snake in the studied landscapes (see above) were gathered through intensive faunistic surveys carried out between 2000 and 2007 by ourselves and our colleagues. In 2008, we re-surveyed seven landscapes where the presence of smooth snake was previously confirmed: Şaeş Valley, Stejăreni Valley, Criş Valley, Floreşti Valley, Malâncrav Valley, an area west of Sighişoara (Şercheş area) and the Nadeş Table land (surroundings of the village Pipea). Overall 56 sites (transects) were visited on 3.28 (SD = 10.7) occasions in average (Min–Max = 1[for one site]-5) between May and the first part of September 2008. In some areas the site searches were finished when major impacts on habitats were noticed (i.e. the road construction in Stejăreni Valley). The area of the sites ranged from 10 m<sup>2</sup> to one hectare (all visually estimated by two people). All snake searches were made by the same person (TH). We consider that the visited sites were likely to include the territory of smooth snakes (the

movement range for this species varying from 2 to 130m (Spellerberg and Phelps 1977, Gent and Spellerberg 1993)). The habitats that were visited were grossly categorized as: grasslands, fallen wood (in the mixed deciduous forest edges), stony structures (abandoned walls, stones along the railways – all were artificially created), traditionally managed orchards, mixed forest-grassland ecotone), downy oak forests (in steep, xerophylic slopes) and drainage ditches. In each site visual searches were made, that lasted from 10 minutes to up to one hour, according to the site characteristics.



**Fig. 1.** The study area with the surveyed sites (black points)  
**1. ábra:** A kutatási terület és a kutatott pontok (feketével)

Because of time and financial constrains, the insides of the forests (where the forest canopy is closed to about 60%) were not surveyed in this study, although previous observations suggest that the smooth snake may be present in this landscape element.

### The model

The program PRESENCE ver. 2.0 (that implements the site occupancy models developed by MacKenzie et al. 2002) was used to estimate the detection probability ( $p$ ) and the proportion of sites occupied ( $\psi$ ). The essence of the site occupancy models developed by MacKenzie et al. (2002) is that they simultaneously estimate the site occupancy, and detectability (see also MacKenzie 2005 a,b). The model assumes that there are a number of sampled sites ( $N$ ),  $\psi$  is the probability that a specific site is occupied, and is constant across sites. The probability of detecting the species, given it is present at a site at the  $j$  th survey is  $p_j$ . Moreover, the probability of not detecting the species (given that it is present) is  $(1-p_j)$ . Assume that we have sampled three sites,  $S_1$ ,  $S_2$  and  $S_3$  and the species was detected in these visits in  $S_1$  and not detected in  $S_2$  whereas in the  $S_3$  the species was detected but there is a missing observation. The detection histories ( $H$ ) for these sites may be for example:  $HS_1 = 101$  (i.e. the species was detected in the first and third visit but not detected at the second one),  $HS_2 = 000$  (the species in question being never detected there in these three visits) and  $HS_3 = 1_0$  (the species was detected in the first survey, the second survey was not made and was absent in the third survey). The likelihood for site  $S_1$  would be:

$$Pr(H = 101) = \psi p_1(1-p_2)p_3$$

When assessing the likelihood for  $S_2$  two possibilities should be considered: (i): the species is present but not detected (i.e. false absence) and (ii) the species is absent.

$$Pr(H = 000) = \psi (1 - p_1) (1 - p_2) (1 - p_3) + (1 - \psi)$$

The first part of the equation represent the probability of false absences and the second one the probability of the site being not occupied. For  $S_3$  that includes a missing observation, the likelihood would be:

$$Pr(H = 1_0) = \psi p_1(1-p_3)$$

After calculating the likelihoods for all  $N$  sites (i.e.  $N$  detection histories) the estimated site occupancy and the detection probabilities can be calculated as the products of all  $N$  (i.e. site) probabilities. The assumptions of this model (see also Schmidt 2005) are: (i) the sites remain occupied during the study period, no extinction, emigration or colonization happens, (ii) the detection probability is greater than zero and (iii) the detection of a species in a site is not influenced by the detection in other sites.

The detection probability ( $p$ ) can be used to estimate the minimum number of visits ( $N_{min}$ ) necessary to be certain with a specified degree of confidence a species is absent from a surveyed site. The degree of confidence ( $\alpha$ ) for this estimation can be set to 0.05 (95% confident) (Kéry 2002) or lower such as 0.01 (99% confident) (Reed 1996). Thus

$$N_{min} = \log(\alpha) / \log(1 - p).$$

The equation for  $N_{min}$  was solved for both 0.05 and 0.01 confidence intervals. To calculate the probability of not seeing a species ( $F$ ) after  $N$  visits we used the following equation:

$$F = (1-p)^N$$

$F$  was calculated for every habitat type that we considered in this study, for  $N = 3$  (i.e. the average number of visits per site) and 5 (i.e. the maximum number of visits per site). We chose this number of visits because it was the most appropriate for our possibilities and this number of visits per site is considered to be “acceptable” for detecting reptiles at national levels (see Kéry 2002). We have calculated the above parameters ( $\psi$ ,  $p$ ,  $N_{min}$  and  $F$ ) separately for every habitat type mentioned above

## Results

The smooth snake was detected in 48% of the sites (naïve estimate) where its presence was previously confirmed. The average number of detections per site was 0.60 (Min-Max = 0–3, SD = 0.8). The probability of species

presence across all sites ( $\psi$ ) was 0.99 (SE = 0.21) whereas the detection probability across all sites was 0.21. The values of  $\psi$  were well above the naïve estimate for the majority of habitats, with the exception of stony habitats and, to a lesser extent, downy oaks (Table 1). The estimates of  $\psi$  are probably biased in all cases except the stony habitats where  $p$  is  $\geq 0.3$ . The bias is especially high in cases when  $p$  is  $< 0.15$  (MacKenzie et al. 2002), such as the orchards in our case. The presence of the smooth snakes was not confirmed in this study in mixed forest-grassland ecotones and the drainage ditches along the dirt roads.

The density of individuals was low in all habitat types (no more than two individuals captured in stony habitats). The detection probability was largest in the anthropic habitats that contained structures made of stones (stone walls, stones along railways) (Table 1). The lowest detection probabilities were recorded for orchards ( $p = 0.09$ ) and grasslands ( $p = 0.18$ ) (Table 1). The detection probability was significantly positively related to the number of visits per site ( $\beta = 0.12$ ,  $t = 3.61$ ,  $p = 0.004$ ,  $R^2 = 54.24$ ).

**Table 1.** The percentage of habitat use (naïve estimate for 2008), the estimated site occupancy ( $\psi$ ), detection probability ( $p$ ), the minimum number of visits needed to conclude that the smooth snake is missing from a site ( $N_{min}$ ) and the probability of not finding smooth snake after 5 and 3 visits ( $F$ ).  $\alpha$  = degree of confidence of  $N_{min}$ .

**1. táblázat:** Az élőhelyhasználat százalékos aránya (naïv becslés, 2008), a becsült élőhely használata ( $\psi$ ), detektálási valószínűség ( $p$ ), a szükséges minimális számú kutatás ahhoz, hogy a rézsikló hiányát megbecsülhessük ( $N_{min}$ ) és annak valószínűsége, hogy ne találjuk meg a rézsiklót 5 illetve 3 vizsgálat után ( $F$ ).  $\alpha$  = az  $N_{min}$  konfidencia tartománya.

	Naïve estimate of habitat occupancy	$\Psi$ (SE)	$p$	$N_{min}$ ( $\alpha=0.05$ )	$N_{min}$ ( $\alpha=0.01$ )	$F$ (5 visits)	$F$ (3 visits)
Grassland	0.36	0.89 (0.45)	0.18	15	23	0.37	0.55
Fallen wood	0.4	0.6 (0.45)	0.25	10	16	0.23	0.42
Stony structures	0.92	1 (0.00)	0.37	6	10	0.09	0.25
Orchard	0.33	1 (0.00)	0.09	32	49	0.62	0.75
Downy oak	0.8	1 (0.00)	0.25	10	16	0.23	0.42

The minimum number of visits necessary to assert that the smooth snake is truly absent from the surveyed site ( $N_{min}$ ) with a confidence of 95% varies from 6 (stony structures) to 32 (orchards) (Table 1). The value of  $N_{min}$  with 99% confidence interval varies between 10 (stony structures) and 49 (orchards). The probabilities for not finding smooth snakes after 3 and 5 visits per site were generally high.

The detection of the smooth snake was not possible along some stony habitats (such are those along the railways near Sighișoara) from the second part of August – early September, due to frequent disturbance by humans and domestic animals (3–4 searches per site with no snake records). No dead specimens were found during these activities.

## Discussion

### $\psi$ , $p$ , $N_{min}$ and $F$

The results of this study can be summarized as follows: (i) the smooth snake is difficult to detect even at sites where its presence was previously documented (assuming no local extinction in 2008, the naïve estimation for this year is 48%). (ii) The estimated proportion of site occupancy ( $\psi$ ) was much higher (0.99) than the naïve estimation, suggesting that the smooth snake was probably overlooked in the majority of the studied sites in 2008. The high difference in the known and estimated site occupancy should be interpreted with caution because the possibility for bias in  $\psi$ , due to the sensitivity of the model to sampling occasions and  $p$  (especially in cases when  $p < 0.15$ ). (iii) the detection probability of the smooth snake varied between habitats, but the maximum values (in stony habitats) were still moderate to low (37%), (iv) the minimum number of searches needed to conclude with 95% confidence that the smooth snake is absent from a habitat varied from 6 to 32. These data suggest that the one to five visits per site (average = 3) that we used to infer the absence of the smooth snake were not enough in these landscapes.

Species and populations may go undetected due to many reasons including low local abundance and habitat occupancy, morphology and behavior of species that make it more cryptic, variations of the sampling ef-

fort, season when the studies are conducted, time of the day, observation bias, weather conditions and habitat complexity (Reed 1996, Boulinier et al. 1998, Kéry 2002, Schmidt 2004, 2005). Many studies demonstrate that the detection probability varies seasonally (among months) (Kéry 2002 for the smooth snake, Pellet and Schmidt 2005 for amphibians). The short survey period and the fact that only one person conducted the searches does not make it possible to account for various factors that may influence habitat occupancy and detection probability. However, in the long term, these will be evaluated and included as covariates in the constructed models. The efficiency of the observer is also important in determining the parameters that we used. Kéry (2002) estimated that an observer with an efficiency twice as his in detecting snakes would correctly identify only 46, 17 and 22% of the three population abundances.

The detection probability of the smooth snake varied according to population sizes of this species in Switzerland (Kéry 2002). The  $p$  of smooth snake in that study for small (maximum 1 individual), medium (2–3 individuals) and large ( $\geq 4$  individuals) populations were 0.09, 0.45 and 0.56 respectively, the largest  $p$  values being found for cliff faces. In our study area, we have never captured more than three individuals on a site in one trip in 2008, suggesting that according to Kéry's (2002) classification, the population abundances in our site were at low to medium (stony habitats) levels. The range of the values of  $p$  in our study area (from 0.09 to 0.37) also suggests that the local densities of the smooth snake are generally low. Recent studies suggest that the detection probability covaries negatively with local probability of extinction of a species (Alpizar-Jara et al. 2004, MacKenzie 2005b). This evidence also infers the potential vulnerability of the smooth snakes as a result of their low population abundance. Although Kéry (2002) found no variation in detection probability of smooth snakes between years (probably because the densities did not change), one of us (TH) observed that the density of smooth snakes can vary between years, probably affecting detection probability. Moreover, a low level of detection probability (assumingly due to the generally low activity level) can also be caused by local disturbances of human origin, suggesting that short term human impacts should also be considered. The  $N_{min}$  (95% confidence) in the Kéry study ranged between 4,5, 34 visits for small, medium and large populations. In our case, the range of variation was from 6 to 32.

The values of  $p$  and  $N_{min}$  differed highly between habitat types. Habitats with dense ground vegetation (grasslands, orchards) had low detection probabilities (most probably due to low population densities, complex habitat structure or an interaction between these two) and require increased effort in order to obtain confidence (with 95%) that the smooth snake is absent. We should consider that the grasslands may contain microhabitats such as stinging nettle patches or eroded areas (especially on steep slopes) that may attract snakes or, being less complex, may allow a better identification of them. For example, study sites with dense grasslands had a lower detection probability (0.09) than the eroded slopes 0.12 or nettle patches (0.16) (Hartel *unpublished*). In our study, the detection probability and snake density was highest in human made structures such as abandoned stone walls, and stone heaps. These habitat structures are not naturally present in our area. Reading (1997) showed in a recapture study that 96% of the smooth snakes were found beneath artificial refuges, the open areas being less used. Moreover, the snake density increased with the refuge number. He concluded that in order to maximize the detection of smooth snakes on heathland sites artificial refugia should be used.

Considering the fact that at regional level the grassland cover (grasslands, pastures) is proportionally large (36.22%) whereas those of orchards relatively low (3.48%), the potential of grasslands in maintaining the smooth snake populations in this area should not be ignored. Although the detection probability is low in grasslands (the structural complexity of grasslands may also be a reason for this [Spellerberg and Phelps 1977]) we believe that the smooth snake has a wide distribution in these habitats and more effort should be allocated to detect it.

The smooth snake was not found in drainage ditches and forest-grassland ecotones during this study. Forest-grassland ecotones are certainly suitable for smooth snakes (as this snake is frequently observed by chance in these habitats (TH *personal observations*)). However, their structural complexity probably does not allow the identification of this snake using the sampling effort we used. The absence of the snakes from ditches (structurally less complex habitats than the ecotones) may be explained by the possibility that only dispersing (not territorial) individuals were identified here in the past. Moreover, even if included in a smooth snake territory, it may be that the ditches do not represent the “core” part of the snake’s habitat.

Theory and observation suggest that local rarity and wide distribution (habitat use) can be one of the many forms of the spatial organization of natural populations. These populations are also characterized by low growth rate and low ratio of extinction to colonization rates (Hanski 1991). These characteristics of population dynamics make these organisms extremely sensitive to habitat loss and fragmentation. Various studies have reported relatively low abundances, small home ranges, sedentary lifestyle, high juvenile mortality, and late maturation of females for smooth snakes (Spellerberg and Phelps 1977, Goddard 1984, Gent and Spellerberg 1993, Reading 1997, Drobenkov 2000). Spellerberg and Phelps (1977) reported after conducting a three year intensive study on the smooth snake: "*Compared to the adder and the grass snake, smooth snakes were not easy to locate in the field. It was not unusual to find only one snake in eight hours of searching and the best record was a total of eight snakes caught by two observers in three hours*". It seems that the low local density and its wide distribution range is a normal feature of its population dynamics in some landscapes, and to understand its population ecology, large scale approaches should be adopted (landscape approaches).

### **Implications for study design of cryptic organisms**

What are the implications of this study? First, it highlights some interesting aspects of considering the detection probability in animal distribution studies. We should recognize that organisms with wide distributions but low local density require other approaches for population delimitation, habitat use and conservation than more locally and regionally abundant and conspicuous species. The distribution and habitat use of the smooth snake may be (and we think that in Romania it actually is) seriously underestimated if the detection probabilities (and the associated statistics) are not made. Secondly, and from a more practical point of view, estimating parameters such as  $p$ ,  $N_{min}$  and  $F$  for various habitats, may help researchers to better estimate the absence and / or extinction of cryptic species. Now, in the era of intense agricultural and infrastructural development, the environmental impact assessments are of crucial importance in lowering the human impact on natural / seminatural environments. An important part of these assessments is linked to species presences and absences. Researchers should be aware that a larger effort should be invested to be sure that the species

absences are real and not false. Third, as the extent of land conversion (especially the open seminatural areas tend to be converted in agricultural and urban areas) is increasing, the role of artificial structures as refuges may be increased in the modified landscapes. However, their efficiency in long term may be lowered if other measurements, such as assuring the connectivity between them and decreasing other anthropogenic pressures (especially direct killing by humans and domestic animals) are not made.

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**A detektálási valószínűség használata egy kriptikus faj: a rézsikló (*Coronella austriaca*) elterjedésének, élőhelyhasználatának és hiányának kimutatásában az erdélyi szász vidéken**

**Összefoglaló**

Az ökológusok gyakran használnak bináris változókat a fajok és populációk élőhelyhasználatának és a bennük végbemenő egyedcserének (turnover) becsléséhez. Ezen kutatások alapfeltételezése az, hogy az egyedek tökéletesen detektálhatók, kimutathatók. A szervezetek detektálásának változását a különböző élőhelyek szerint csak kis számú kutatásban veszik figyelembe, ennek elhanyagolása viszont téves eredményekhez vezethet. Az detekció figyelembe vétele különösen fontos a kriptikus fajoknál, mert ezeket a kutató könnyen szem elől tévesztheti, és ennek következményeként hiányzóként könyvelhet el egy fajt, holott az jelen van. Dolgozatunkban a rézsikló élőhelyhasználatát mutatjuk be, valamint a faj detektálhatóságát és a szükséges minimális kijárások számát annak érdekében, hogy megállapíthassuk, a faj valóban hiányzik egy adott élőhelyből. A rézsikló Európa-szerte kutatott faj, Romániában is intenzíven leltározzák. Kutatásainkat 2008-ban végeztük hét különböző tájhasználati egységben (56 élőhelyen), ahol a rézsikló jelenléte az előző évek során már jelezve volt. A detektálási valószínűség 0,09 és 0,37 között változott, míg a faj egyedeinek hiányát 95% pontossággal kimutató minimális terepkutatások száma 6 (köves élőhelyek – vasúti töltések, omladozó falak stb.) és 32 (gyümölcsösök) között változott. Dolgozatunkban megemlítjük a detektálási valószínűség elhanyagolásának lehetséges veszélyeit is.