Czech University of Life Sciences Prague

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Diploma Thesis

Wolf pack abundance - comparison of two approaches

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ZADÁNÍ DIPLOMOVÉ PRÁCE

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Cíle práce

Vlk obecný se v posledních letech objevuje ve středoevropské přírodě a již i u nás vytvořil plně funkční teritoria obhajovaná zformovanými smečkami. Moderní technika výrazně pomáhá ve studiu početnosti, aktivity a dalších projevů tohoto důležitého predátora. Rozvoj technologie fotopastí a její schopnosti jsou dnes značné, co prozatím není zcela usazené je způsob jejího nasazení. V základu je dnes používán tzv. ad hoc systém, kdy jsou fotopasti umísťovány na místa, kde byl evidován nějaký projev objektu zájmu. Tento způsob může mít svá úskalí, ta však jsou prozatím nedobře studována.

Metodika

Cílem práce bude vyhodnocení dvou přístupů fotomonitoringu, který proběhne na dvou známých výskytech vlka v severních Čechách (Krušné hory a České Švýcarsko).

Studium proběhne na základě následujících kroků:

- průběžný ad hoc fotomonitoring v oblastech s předpokládanou aktivitou smečky (Krušné hory a České Švýcarsko)
- intenzivní fotomonitoring s pravidelnou sítí fotopastí přes předpokládané ohnisko každé sledované smečky (základní parametry designu: 1000 fotodní na ploše cca 40 km2, dvě kampaně [jarní a podzimní] po dvě sezóny).

Doporučený rozsah práce

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vlk, fotomonitoring, homerange, abundance

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| Declaration |
|--|
| I declare that I have worked on my diploma thesis titled "Wolf pack abundance - comparison of two approaches" by myself and I have used only the sources mentioned at the end of the thesis. As the author of the diploma thesis, I declare that the thesis does not break copyrights of any their person. |
| In Prague on 29.6.2020 |
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| Ackn | nowledgement | |
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| | uld like to thank to my supervisor Aleš Vorel and all the members of the for their help, advise and support during my work on this thesis. | e OWAD |
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Wolf pack abundance - comparison of two approaches

Abstract

This master thesis compared two monitoring methods of wolf (*Canis Lupus*) packs with the use of camera traps. Camera traps were deployed in two different study areas in the north of the Czech Republic. Each of the method was repeated three times in both of the study areas. The first method – to which this thesis refers as the "Fishnet Coverage" - was based on covering random part of study area with camera traps placed in systematic square grid. The second method called the "Ad. Hoc" used lower number of camera traps with non-systematic placement but the sites of the camera traps were selected based on prior information acquired about the wolf pack activity.

The objective was to find out, how many of "wolf events – photos of wolves separated by an hour" are the methods able to capture. Prediction was that the "Ad. Hoc" method will perform better and thus produce more "wolf events". For each method different number of camera traps was deployed, therefore two indexes were used for possible comparison - "RAI – relative abundance index" and the second "naïve occupancy" index.

The "Ad. Hoc" method had overall better results in both of the study areas. But the when the methods were compared between the areas, results showed that, the "Fishnet Coverage" method was more suitable for one of the study areas and vice-versa.

Keywords: camera traps, monitoring methods, wolf, territory, population, pack size, "Fishnet Coverage", "Ad. Hoc"

Početnost vlčích smeček – porovnání dvou přístupů

Abstrakt

Tato diplomová práce porovnávala dvě metody monitoringu vlčích (Canis Lupus) smeček za pomocí foto-pastí. Foto-pasti byly rozmístěny ve dvou různých oblastech na severu České republiky, obě metody zde byly třikrát zopakovány. První metoda, která je v této práce označována jako "Fishnet Coverage" byla založena na pokrytí náhodně vybrané části monitorovaného území foto-pastmi seřazenými v rovnoměrné čtvercové síti. Druhá metoda nazývána "Ad. Hoc" využívala menší počet nerovnoměrně rozmístěných foto-pastí na předem vybraných místech dle dostupných informací o pohybu vlčí smečky.

Cílem práce, bylo zjistit, kolik vlčích událostí – fotek oddělených jednou hodinou jsou metody schopné zachytit. Předpokládalo se, že "Ad. Hoc" bude úspěšnější a zachytí víc vlčích událostí. Pro každou metodu bylo používáno rozdílné množství foto-pastí, a proto byly pro porovnání výsledků použity dva indexy: RAI – relative abundance index" a druhý "naïve occupancy" index.

Metoda "Ad. Hoc" měla celkově lepší výsledky v obou studovaných oblastech. Nicméně při porovnávání metod mezi oblastmi výsledky ukázaly, že "Fishnet Coverage" se lépe hodila pro jednu oblast a "Ad. Hoc" pro druhou.

Klíčová slova: foto-pasti, monitorovací metody, vlci, území, populace, velikost smečky, *Fishnet Coverage*", "Ad. Hoc"

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1. Introduction

Grey wolf (*Canis Lupus*) an apex predator with high ecological significance is making a comeback to central Europe. Successful monitoring is a key part in conservation efforts and mutual coexisting amongst this animal and humans.

Over the last decade, the Grey wolf (*Canis Lupus*) as the main representative of Europe's large carnivore species has formed stable populations in Germany and is now migrating over the borders into the Czech Republic. Wolves return is marked with polarized opinions on whether this elusive species has its place in the anthropogenic landscape that covers significant areas of central Europe. Due to the high mobility, widespread home range of wolf packs and central Europe's fragmented forest habitat, their localization is difficult and monitoring logically demanding. Therefore, precise population estimates are limited depending mostly on radio-telemetry.

The process of wolf monitoring may vary extensively according to diverse objectives appointed by different studies. Certain methods and principles are already known and accepted. However, it is essential to implement and approve standard methodological procedures amongst European nations as wolves do not respect national borders and can move within two or more states. One of the validated tools in wildlife monitoring is the use of the camera-traps. The main question is how to maximize its potential.

In this master thesis, two different approaches of camera-trap use were tested. The first was based on a network of quadrants placed in grid over the entire target area. Its principal was random selection is camera sites in systematic grid. The second method called "Ad Hoc" is based on placing the camera traps on non-randomly selected location with non-systematic spatial positioning. The principal functioning of the two methods is different, therefore they were compared to found out which one produces better results.

This thesis is supported and could only be elaborated in support of OWAD – the Wolf Project (Objective Wolf Acceptance in human-altered cross) and its team's assistance. The project is financed by the European Union from the European Regional Development Fund (ERDF). Its main goal, is to conduct continuous monitoring in the north of the county on the boarders with Germany and the part of the project should be sharing of the knowledge between both sides to standardize the monitoring methods and its evaluation systems. In the process the team efforts should lead to the gathering of data about wolves migrating from Germany and forming new packs in the Czech Republic. These packs should be monitored with the use of photo-cameras and the information obtained could potentially lead to collar telemetry.

Collected data also serve for molecular and food analysis. The project also aims to raise awareness via seminars with public, students and target groups such as hunter or farmers.

Because this thesis was one of the parts of the OWAD project, sometimes the word "we" is used as I could not to carry out all the tasks by myself. My part in the project was to provide all the maps and documentation necessary to create the designs for both of the two methods. Also, I was present in most of the camera installation or collection of camera traps so I could get the best inside into the workflow which is essential for the methods comparison.

The motivation for this thesis was to found out the best use of camera for increasing the potential of conservation efforts. Additionally, to determine the weaknesses of the methods and to discover diversity of their use.

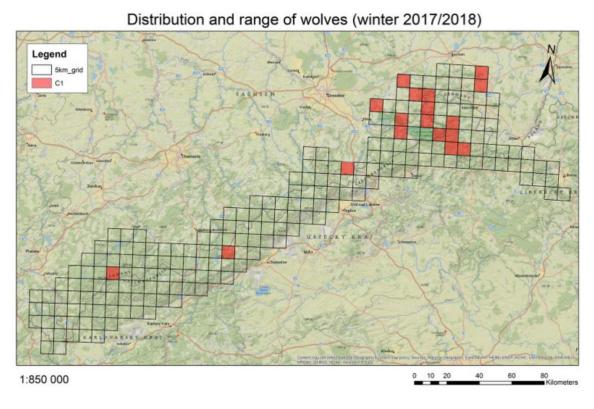


Figure 1: (Vorel, et al., 2017) / OWAD.cz

2. Objectives and Methodology

The main question this thesis wants to answer is, how many of "wolf events – photos of wolves separated by an hour" are the methods able to capture. Therefore, we tested which method will capture more wolf events over duration of approximately 1 month. Because one of the methods - the "Fishnet coverage" used more camera traps than the "Ad. Hoc" method two indexes were used for possible comparison of the results. The first index was "RAI – relative abundance index" and the second "naïve occupancy" index. The prediction – the alternative hypothesis - was that the "Ad. Hoc" method would perform better than the "Fishnet coverage" method in both study areas.

For the practical part, two target areas were selected and both methods were repeated tree times. For the first method - the "Fishnet coverage" - we started by setting grid of 47 camera traps in the Bohemian Switzerland National Park and after 30 days we moved into the Ore Mountains where the grid composed only from 40 camera traps as the area was smaller. We repeated this method 3 times - once during the winter and twice in the summer. For the "Ad. Hoc" method we used 10 camera traps. This method was done simultaneously in the area where the grid was not installed. We tried to install the camera traps in the exact same months for both of the methods but it was not practically possible as there was only limited number of camera traps for different purposes.

Both methods were compared by the two indexes in 5 different combinations. First all the data from "Fishnet coverage" method was compared to all the data from "Ad. Hoc" method without the dependence on the study area. Next the "Fishnet coverage" method from the Bohemian Switzerland National Park study area was compared to "Fishnet coverage" from the Ore mountains and the same comparison was done for the "Ad. Hoc" method. Lastly the results of the "Fishnet coverage" from the Bohemian Switzerland National Park were compared to the results from the Ore mountains and the same comparison was again done for the "Ad Hoc" method.

3. Literature Review

3.1 Background

Before the 17th century, wolf occurred in abundance throughout Europe's continent. But how modern human age proceeded and their settlements spread across wolf natural habitat, the large carnivore faced tough competition and was consecutively pushed out to very few areas. In Central and Northern Europe, wolves were basically eradicated on the beginning of the 20th century. In Bavaria, the last wolf was killed in 1847 and in the Czech Republic in 1874. One of the last wolf's refuges in Western Europe was France. In 1883, the population counted up around 1,386 wolves, but just four decades later in 1927, the last wolf was killed in France (Reinhardt et al. 2013). And around the 1960s, the wolf population in Europe was at an all-time low (Boitani 2000). However, in many Europe countries, there was a positive change of public attitudes towards large carnivores which resulted in higher interest in wildlife conservation efforts and the wolf population numbers became more stable. The main transformation started in the second half of the 20th century as the legal status of the wolf changed and the animal was put on partial or strict protection. For example, in Poland in the 20th-century wolf was listed as a pest, then it became a game animal and finally, it was marked as a protected species in 1998 (Reinhardt et al. 2013).

The turning point of wolf's comeback from so called "refuges" back to central Europe was migration from western Poland to Germany. Wolves started to cross the borders to Germany around late 1990s and settled in the Saxony-Brandenburg region and the first reproduction was noticed in 2000 (Reinhardt et al. 2019). The increase in wolf numbers was remarkable and after the first successful reproduction a new pack was confirmed in next 5 years. In the 2005 there was and evidence about two wolf populations. Throughout monitoring in the 2017/2018 there was identified 73 packs, 30 pairs and 3 individual wolfs. Also wolf occupancy was confirmed in six federal states (Reinhardt and Jarausch 2016).

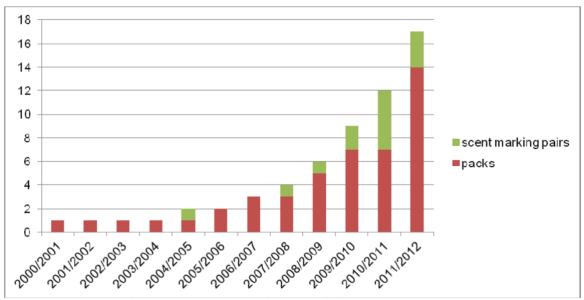


Fig. 3: Development of wolf population size in Germany. © LUPUS.

Figure 2: The figure shows the increase of wolf packs and pairs numbers in Germany from season 2000/2001 until 2011/2012. (Reinhardt, et al., 2013.

The extraordinary comeback of this predator to Germany is an example of how adaptable the animal can be. From 2000 to 2015 wolf population in Germany rose annually by 36% and the number still rises (Reinhardt et al. 2013). An important role in the return had MTAs (Military training areas) and research from 2019 suggests that MTAs functioned as stepping-stones between packs in the reoccupation process and even that these areas might had bigger impact than protected areas (Reinhardt, et al., 2019). A research published in Science in 2014 suggest that the wolf Europe's population is twice as bigger as the wolf population of the contiguous United States. That shows great adaptability of wolf to anthropogenic landscapes and partial independence on protected areas or remote wilderness. Moreover, when we take into account that the United States are almost double in size and have twice lower density of human population (Chapron, et al., 2014).

That being said, generally wolf population needs large forest habitat with low human activity and that is something what rural borderline areas in Czech countryside can offer. The border areas in the Czech Republic might seem as an ideal habitat for next migration into the western Europe as many are either under state protection or forest connected to other forest areas over the borders forming sufficient range of adequate habitat. Moreover, the Czech Republic position in the central Europe appears to be unique as it could be the meeting point between different wolf populations. The wolves coming from Germany form packs in the north of the country represent individuals from the Central European Lowlands population, in the east there is regular occurrence of the wolves migrating from Slovakia representing the

Carpathian population, and in the south, there is a possible arrival of an individuals from the Alpine population in Austria or Dinaric-Balkan population. As a result of the increasing population in Germany's Lusatia region, around 2012 the first wolf individual was registered near northern boarders at Šluknovko region. And by 2014 there was registered mating pack in the protected landscape area Kokořínsko. Next year reproduction was confirmed with the use of the camera trap monitoring. At the same time in 2012 at Šluknov Hook, there was an attack on two goats and zoologist from Bohemian Switzerland National Park assumed that signs of suffocation indicate potential wolf presence, but it was not confirmed. However, following car accident in Hohwald Germany, confirmed death of a wolf female in absolute vicinity to the Czech borders (Žák, 2015).

3.2 Current situation of Wolf population in Czech Republic

Wolves are spreading to the Czech Republic from 3 different foreign resources in 3 different parts of the country. In the Beskids mountain wolf occur continuously but due to the high road density and insufficient migrating corridors in the area the animals generally stay around the boarder with Slovakia. Recently wolf primarily occur in the north side of the country in Šluknovsk region, Lusatia Mountains, Ore Mountains and Broumovsko. In all of those areas wolf migrate from Germany and in 2018, minimum of 6 pack was confirmed by Nature Conservation Agency of the Czech Republic with cooperation of OWAD and Hnutí Duha. The pack are currently being monitored on both sides of the borders. The last place where wolf occur is in the Šumava National Park. It is assumed that 1 pack is currently living in this area but further information is needed for confirmation.

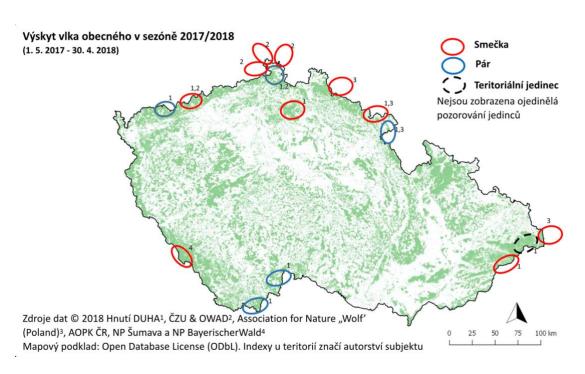


Figure 3: Wolf occurence in Czech Republic. (Hnutí Duha, 2018), (ČZU and OWAD, 2018) (Association for Nature "Wolf" (Poland) (NP Šumava and NP BayerscheWald)

3.3 Wolf populations

By a definition – a population is a group of organisms of the same species, living in a particular geographical area with a potential capability off interbreeding with each other. But if we take into consideration the widespread distribution of wolf packs inside of the European countries - how do we determine where on population ends and other begins? Basically, there are two concepts of how to design population maps. First, it could be done based only on distribution data or with the use of DNA analysis

In 2008, there were a recognized 10 wolf populations in Europe (Linnell et al. 2008). These 10 populations are North Western Iberian, Sierra Morena, Alpine, Italian Peninsula, Carpathian, Dinaric-Balkan, Baltic, Karelian, Scandinavian and Central European Lowlands (Boitani et al. 2015). Which is also an official information about wolf populations on the European Commission website. Linnell et al. (2008) categorized two of them – the Sierra Morena population and the population in Germany / western Poland, now known as Central European Lowlands population – as critically endangered in 2008. But since then, in the Central European Lowlands population, there has been a trend of increase of individuals contrary to the Sierra Morena population, where the decrease was noted, presumably because of the area's solitude. Different research by (Stronen et al. 2013) works with 4 population clusters – Italy, Dinaric-Balkan, Carpathians mountains, Ukrainian Steppe and Northcentral Europe ignoring

the Iberian population. It confirms genetic divergence between Italian population and wolves in the rest of the Europe suggested by (Lucchini, et al. 2004). Another research by (Hulva et al. 2018) found several different haplotypes in the European lowland, Baltic, Carpathian, Italian-Alpine and Dinaric Balkan populations dividing it into two groups. With haplotype W1 and W2 found mainly in Central European and Baltic populations with only few individuals with these haplotypes occurring in the Carpathian population. Haplotype W1 is also found in Scandinavian population and is the most frequent to be found in Central European lowlands. Whereas, haplotypes W14 and W6 were basically only identified in the Carpathian Mountains and few individuals in the Romania. Another two haplotypes were found in Dinaric-Balkan and Italian-Alpine populations. And W3 probably belonging to the Ukrainian population.

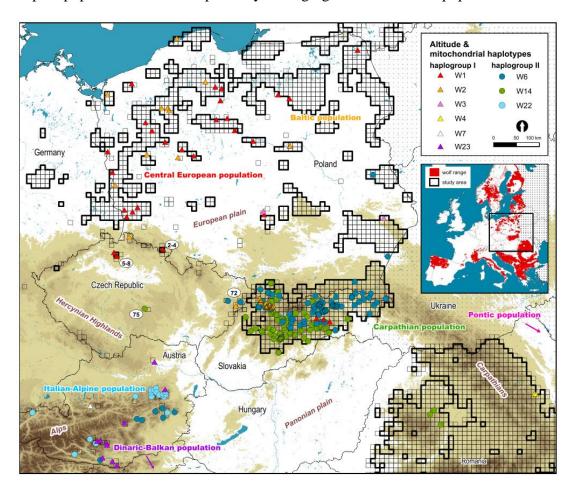


Figure 4: Map of haplotype distribution in Europe. (Hulva et al. 2018)

Results of genetic analyses of wolf DNA samples gathered from Poland in between 2004 and 2007, recognized two clusters and also separated the Carpathian wolf population from the European lowland population. This indicates that individuals repopulating western Poland come generally from the Scandinavia and Baltic region (Nowak and Myslajek 2008). This is supported by another research of wolf populations in Europe, by stating that the Carpathian

population indicates differentiation between wolf from Polish Carpathians and from northern lowlands. And even though the Carpathian samples were from Ukraine and Slovakia, individuals from the Polish side of the mountains were similar (Stronen et al. 2013). It is suggested that Carpathian population has reduced migration potential contrary to the Baltic or Central European lowland (Kutal et al. 2016). There is also clear clustering between Carpathian and Dinaric-Balkan population (Bakan et al. 2014) with morphological differences between skull size and shape (Milenković et al. 2010). But there is potential of gene flow between Eastern Carpathians in Romania and Pontic population in in the east (Hulva et al. 2018)

More examples of differences between populations could be found in many researches as it principally depends on what objectives the research wants to achieve and on exact area its focused on. For example in a study on genetic diversity of wolf population from Central Balkan which is location occupied by the Dinaric-Balkan population, the researchers found differences between haplotypes of wolves from several different countries and thus they divided the Dinaric-Balkan population into "eastern" for samples from Serbia and Makedonia and "western" for samples from Croatia and Bosna and Herzegovina (Djan et al. 2014). However it seems that the general scientific consensus is that we distinguish 10 separate wolf populations around the Europe (Chapron et al. 2014).

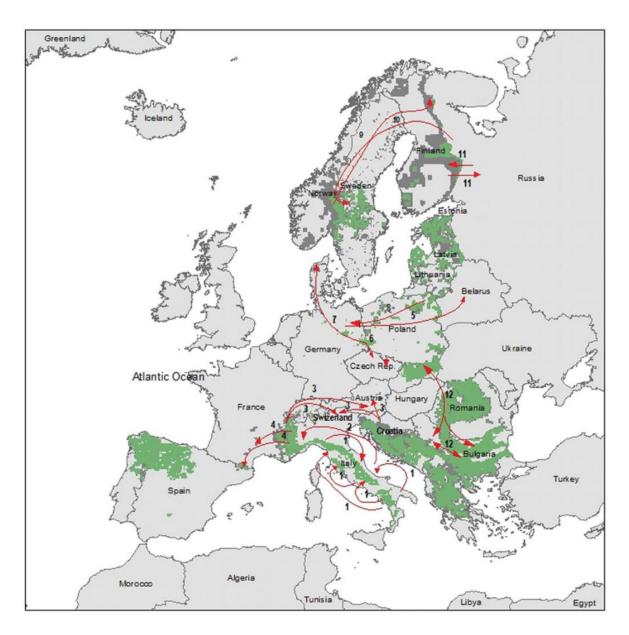


Figure 5 - Map of Wolf distribution in Europe. Red arrows indicates migration between and within populations. (Hindrikson et al. 2017)

Europe wolf population are generally transboundary and it was proven that wolves, dispersal abilities have large range of distances is long range. Therefore, wolf migration and gene flow between populations is essential to maintain its successful development and play a key part in similarities or difference in the populations.

In following subchapters, I will summarize wolf populations with direct connectivity to the Central European Lowland population which is the subject of this study. Therefore, Scandinavian, Karelian, Iberian and Sierra Morena populations will not be part of following text.

Central European lowlands population

Central European lowlands population is a term used for a population expanding from Western Poland into the central Europe. Wolves were never totally exterminated in Poland but most wolves survived in eastern part of Poland which is more related to the Carpathian population. In the western Poland there were reports of only around 20 wolves living in small packs or solitary and without reproduction (Nowak and Mysłajek 2016). From year 2000 when first reproduction was confirmed in Germany their number increased rapidly and in 2015 wolf occurrence was documented in 16 federal states (Reinhardt et al. 2019). The estimates from 2014 suggested that around 43 wolves were present in Germany and around 100 wolves in Poland (Chapron et al. 2014).

The connectivity to other wolf populations is limited to the Baltic population and occasionally the Carpathian population. It is believed that road density limits the population's ability to spread further into western Germany (Hindrikson et al. 2017) which could be the reason why wolf migrate into the Czech Republic.

Carpathian population

Second biggest wolf population by size estimations occupies a region from southern Poland through Slovakia and Romania to Serbia where it connects with the Dinaric-Balkan population. The estimates are about 3000 individuals with most of the population located in Romania around 2000 individuals (Chapron et al. 2014). This population has significant importance as its location provides connection between wolves from eastern and western populations but also between the north and east (Hulva et al. 2018).

Italian population

The population from Apennine peninsula is believed to be affected by repeated bottleneck effect not only in last century but also during the last 2000-10000 years. But after relatively recent protection by law and enough of prey the Italian wolf now occupies large part of the Italy and migrates into the western Alps (Pilot et al. 2014). Recent estimates are around 600-800 individuals (Chapron et al. 2014).

Alpine population

The Alpine population contains around 160 individuals in 30 packs and covers a mountain area mainly in France and Italy but also in Austria and Switzerland (Chapron et al. 2014). The recolonization started by the very end of the 20th century in the western Alps after the last wolf in the province was killed around 1920 (Marucco, Avanzinelli, and Boitani 2012).

There are reports about gene flow between the Alpine population and Dinaric-Balkan population in recent years. A study from 2016 called "Long-distance dispersal connects Dinaric-Balkan and Alpine grey wolf (*Canis lupus*) population" it provides telemetric data from young male from the Balkan population who migrated to the Italian alps in 98 days covering more than 1200 km (Ražen et al. 2016).

3.4 Pack size estimates

Wolves form family groups controlled by a breeding pair with several generations of offspring. Beside wolf packs, wolves can live solitary or in pairs. Wolf packs are specific for their cooperation, social-bonding, high social cohesion and overall relationships of interspecific relations (Cafazzo et al. 2018). Deaths inside the pack can affect social stability and lead to dissolution or abandonment of home range. This is very important in fragmented landscape where car accident is frequent (Bassing et al. 2019). Only the breeding pair is allowed to reproduce which limits the potential of inbreeding. The gen flow inside the population is secured by dispersal of young wolves who leave the natal pack to find new territory and mates (Gula, Hausknecht, and Kuehn 2009). Therefore, wolf pack size fluctuates during the year with summer being the season with higher number of individuals. A study from Białowieża Forest in Poland, found average pack size ranging from 3 to 7 individuals in the winter and 4 to 8 individuals in the summer (Jerdrzejewski J. et al. 2007). Wolf packs in Yellowstone park usually consists of 9.8 wolves, but could grow up to 20 and 37 was the highest number of individuals recorded (Cassidy et al. 2015). However, in Europe the size of the pack infrequently surpasses 10 individuals (Duchamp et al. 2017). Naturally, the bigger size during the summer is due to the reproduction but as the year continues some pups may not survive and also some adults generally die during fall and winter (Todd K. Fuller, L. David Mech and A 2003). The pack size is important as it secures safety and improves packs ability to survive. But it is not a rule that the largest number of individuals is the best strategy. Large packs are the strongest, hence it provides successful outcome in conflicts with other packs. However, the most efficient number of individuals for hunting large ungulates is believed to be four wolves (MacNulty et al. 2012) and for the most efficient reproduction it is an eight pack members (Stahler et al. 2013) (Cassidy et al. 2015). To determine precise wolf pack size estimates, scientists generally use multiple integrated methods. Radio telemetry and direct aerial observations have the best potential but are often difficult to manage, are expensive, time-consuming and not effective in dense forest habitats (Roffler et al. 2019). The effectivity of non-genetic sampling (NGS) is still partially unexplored but could provide valid outcome. However collection of NGS depends on detailed knowledge of territory, packs movement and localization of frequently used trails and rendezvous sites (Stenglein et al. 2011). Another well know method for pack size estimates is elicited howling. Wolves tend to respond therefore it is possible to confirm the occurrence of the pack and actually determine how many "voices" were in present in the wolf chorus (Suter et al. 2017). This is a promising method but it may be problematic if wolves do not respond back and definitely not precise as all voices of all individuals cannot be found in the chorus or if large number of individuals are howling (Papin et al. 2019). The use of the camera traps is not well examined for this purpose. Estimating wolf pack size by camera traps is not frequently used because wolves as individuals are not easily recognizable (Mattioli et al. 2018) thus the implementation of classical capture-recapture model is difficult or impossible. Nevertheless, camera traps can provide photos/videos of the whole pack. Assuming that, the capture-recapture model could work while analyzing only captures of the whole pack. Moreover, installation of extensive camera trap grids over packs territory it can produce more valuable information about the pack movement and size. In any case, further examination is necessary to find the most efficient and precise method combination.

3.5 Home range size estimates

The size of the wolf territory depends on habitat quality and it can vary because of multiple reasons. It's believed that the size decreases with increasing prey availability and vice versa. In other words, the territory is as small as possible due to the energetic cost of its maintaining but can be as large as required (Kittle et al. 2015). Research suggests that vegetation type makes little difference as long as an abundance of prey is available. Wolf are considered habitat generalists. They can survive in mountain ranges, deserts, woodlands, swamps, tundra or prairies (Fuller, Mech 2003).

Wolf activity in the territory depends primarily on movement and behavior of their prey (Gurarie et al. 2011). Also wolf activity and therefore territory use changes throughout the seasons with shifts in their behavior. During denning season – in April and May the pack stays present in vicinity of the den site as it tries to protect the pups and only some individuals hunt and return with food (Roffler and Gregovich 2018). When the pups get older in the late summer the pack leaves the den and starts to roam their territory using rendezvous sites for gathering. This movement continues to the winter and in the spring young individuals might disperse out of the territory to find a new partner or pack (Lewis, White, and Murray 1997). Size of the territory changes depending on space limitation. Wolves generally live in well-defined

territories marked by scents, scats and howling. Scents present precise boarders of the territory and howling serves as short-term information about the packs current position. Territories may have small overlap with neighborhood packs. However, these overlaps usually create a "nowmans-lands" as wolves practically stay away from borders to dodge confrontations with rival packs (O'Neil, Beyer, and Bump 2019). Different study on the other side suggests that when wolves look for a new territory they may incline to prefer possible confrontation with other packs rather than settle for less favorable habitat (Harrington and Mech 1983). Overall, the size of the territory is shaped by multiple mechanisms of ecological factors. The negative correlation of home range size related to prey abundance is found in many studies but results are not consistent. Another factor is elevation, pack size, human density and prey selection. For example, wolves focusing on smaller prey generally use smaller home ranges than wolves who hunt large ungulates (Mattisson et al. 2013). Therefore, size of the wolf home range varies a lot depending on what is available in surrounding habitat. In Minnesota USA the estimates of territory controlled by one pack range from 125 to 310 km² (Lewis, White, and Murray 1997). According to (Mattisson et al. 2013) the smallest home range in Scandinavia exceed the size of 260 km² while an average home range in the rest of the Europe. Moreover, the biggest Scandinavia home range can approach the size of those in Yukon or Alaska. In Germany an average home range is estimated around 200 km₂ (Fechter and Storch 2014). In the Polish part of Bialowieza Primeval Forest, wolf home range were between 62km2 and 125 km2. During spring and summer wolves occupied smaller territory than in the autumn and winter (Jerdrzejewski J. et al. 2007). Also "floaters" – lone wolves generally occupy larger are than wolf packs (Mancinelli, Boitani, and Ciucci 2018).

Telemetry is perhaps the best tool for estimating wolf territory size as the movement of the collared animal can be constantly tracked. Therefore, the home range is designed within the area in which the individual travels. For precise estimates off annual home range it is needed at least 9 months with 5 or more locations per one month (Mattisson et al. 2013). The telemetry is often combined with other methods – NGS collection, observations and surveys for better results. The use of camera traps for home range size estimates is not generally used. But can be combined with other methods in the process. Although, if large grids of camera traps are installed it could provide valuable information about the boundaries of wolf territory.

3.6 Wolf monitoring

Wolf monitoring is a logistically difficult, time demanding and often a physically challenging task, due to the animal's high mobility, natural elusive behavior and vast home range.

At first, before designing a monitoring strategy, there is a need to determinate clear objectives that need to be achieved. Because for various objectives, there will be different monitoring methods appointed. Basically, two types of monitoring can be distinguished - active and passive. The term passive is used for the collection of data obtained from public or target groups, who may have relevant information about wolf activity in a certain area. The other one – active – refers to the actual fieldwork, when the team is in the area usually collecting genetic evidence, following tracks, placing photo-traps, etc. Because of the overlap of wolf populations over two or more countries, there is a strong need for standardization of wolf monitoring methods. The unification of the data evaluation from active monitoring is a key part of having credible information to compare within one country or between multiple countries.

For wolf data evaluation, there exists a method called SCALP (Status and Conservation of the Alpine Lynx Population) – at first design for evaluation Lynx (*Lynx Lynx*) data but eventually being also used for wolf monitoring purposes developed by Kora foundation.

Methods of monitoring can be generally divided into invasive and non-invasive. Majority of the monitoring methods is considered to be non-invasive. However, it could be argued that an elusive howling can affect the animal's behavior and similarly camera trapping with use of baits can lead to affected behavior. Although, generally a method is perceived as invasive only if there is a direct contact with the animal. The following chapters summarize commonly used methods for wolf monitoring.

3.6.1 Camera Trapping

The use of camera traps started around 1990 for tiger (*Panthera tigris*) abundance estimates. The purposes of camera traps vary through different ecological studies but is mostly used for population estimate, species richness and site occupancy. However most of the camera trap studies provide additional data about the target areas both related on non-related to the studying subject, that can be further used (O'Connell, Nichols, and Karanth 2011).

Basically, we can distinguish two types of camera traps depending on the triggering system. The first group called active, have infra-red beam and if something interrupts it the camera takes a photo. That is ideal when nothing can obstruct the beam as they hardly miss crossing animal. On the other side those cameras are prone to false captures caused by moving branches, leaves, shadows etc. The second group called passive, capture a photo whenever an object with temperature different than the environment crosses its field of view. Those camera traps do not incline to false captures but can more frequently miss a target when the temperature

of environment is close to the animal body. That can be caused by sunlight (Noss et al. 2013). Other research suggests that passive cameras are not 100% successful in detection. Tested by installing two passive cameras at the same site, final data are different (Rovero et al. 2013). Anyway, according to (Wearn, R. Oliver, Glover-Kapfer 2015) most camera traps on the market are passive. One of the most popular camera trapping use is the detection of elusive and endangered carnivores (Rovero et al. 2013). The use of camera traps is frequent for monitoring of lynx (*Lynx lynx*), jaguars (*Panthera onca*) or snow leopard (*Panthera uncia*). Those species have unique patterns of markings. That enables recognition of individuals, the use of capture-recapture model and hence precise population estimates (Janečka et al. 2011; Stergar and Slijepčević 2017; Noss et al. 2013). Recognition of individuals is something mostly impossible to practice in wolf monitoring unless the animal has extremely distinctive trait or skin disease.

Camera trap settings

The right settings of camera traps are the first step to achieve successful monitoring via this method and part of the process, which should be error-free as it is only a matter of careful and precise preparation. The major camera features that influence the result are trigger speed, flash type, detection zone, delay time — number of photos taken over time, sensitivity, image resolution, camera housing, and power autonomy. In many cases, the trigger speed is associated with a higher rate of successful rate of detection. Because if trigger speed is low, there is a chance that the camera trap will not capture all the passing animals. It will capture only part of the animal, or the image will be blurry. However, a wider detection zone could compensate for slow trigger speed (Fancourt, Sweaney, and Fletcher 2018). This study analyzed the importance of trigger speed by rabbit detection when used two different camera traps. One "Reconyx" with higher trigger speed and narrower detection zone and the second "Acorn" vice versa. The results showed that the wider detection zone had a better detection rate than the higher trigger speed (Fancourt, Sweaney, and Fletcher 2018).

Site selection and camera trap placement

For successfully capturing photos of wolves, there is a strong need to select the ideal and precise sites for the camera trap installation. Because without correct preparation and location scouting, the chances to succeed are low. Therefore, detail knowledge about the target area is essential.

If a particular area is targeted with signs of wolf presence, it is recommended by researchers to try contact local foresters, hunters, and citizens who can provide suggestions about specific locations, landscape features, and recently found carcasses or sightings in the area (Moskowitz and Huyett 2019). Although, in locations or countries where the public attitude

towards wolves is negative, which would not be uncommon, this may turn out problematic. Moskowitz and Huyett (2019), divided the placement of the camera traps into several groups. The first is called "General travel"; those are the camera traps where it is not used any bait to affect the animal. The second group called "Intra and interspecies communication," in this group, camera traps are placed near scats, scents, or artificial lures. The next group, called "Foraging behavior," are camera traps set near carcasses or other baits associated with prey. The last group, called "pup-rearing behavior," uses rendezvous sites such as dens, for camera placement. The use of attractants and lures can boost capture efficiency as it can draw the animal in front of the camera. There are many possibilities to choose from when selecting a bait, but generally, researchers use a mix of trapper's lures, imported food baits, artificial baits as Calvin Klein human sprays. Another option is to find the bait in the field, for example, carcasses discovered in the target area (Mills et al. 2019; Moskowitz and Huyett 2019). The reaction to attractants can vary, and wolves act accordingly to their previous experience with the specific lure. So, if the attractant is connected with a previous threat or negative experience, the animal can actually avoid the site. Most baits, when installed, do absorb human scent and cause a negative effect, but in areas where human activity is common, it is unlikely to cause any problem (Moskowitz and Huyett 2019).

Researchers frequently use camera traps placed in certain grids. Because, concrete methodology plays an important part in scientific studies, it is common to particular design systems repeatedly. According to (Wearn, R., Oliver, Kapfer-Glover 2017) there are 3 main designs to be distinguished. First a random selection of camera stations, the second is systematic random design and the thirds is clustered design.

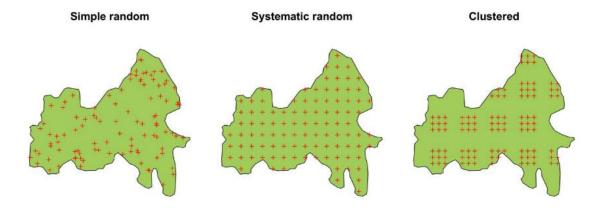


Figure 7-2. Basic sampling designs for camera-trapping. In general, systematic random sampling is preferred, but simple random or clustered designs are useful in some circumstances. Sampling designs for a hypothetical study area (in green) were generated using the "secr" package in R, by adapting code from Mike Meredith. This package has a number of useful functions for automatically creating sampling points.

Figure 6: Camera-Trapping grid designs. (Nichols et al. 2019)

Data management and analyzing

The installation and de-installation are only one part of the work process of camera trapping. The necessary part for analyzing your data is to process and organize all photos/videos collected. This may seem obvious but as stated in (Wearn, R., Oliver, Kapfer-Glover 2017) this step is a "significant bottleneck" in many studies and conservation efforts as not all scientists can afford manual processing by trained workers. This is serious setback because this results in wasting hard-obtained data instead of producing information. Therefore, it is vital to establish an effective system for ordering and organizing the data because camera-trapping generally produces hundreds or thousands of images. It is recommended to create and Excel file where all the information such as, unique identifier of camera trap, location with coordinates, site appearance, installation and pickup datetime, trap-days, camera model, number of captures, name of worker and so on (Noss et al. 2013). This allows simple orientation in the collected data which is essential especially for extensive camera trap surveys and longtime assignments. Multiple researchers recommend using multiple data forms (Athreya et al. 2014) or use of relational database (Microsoft Aces or SQL database) (Wearn, R., Oliver, Kapfer-Glover 2017) to achieve well-arrange overview. Camera Trap Metadata Standard (CTSM) is an example of system for storing and sharing camera trap data developed by multiple organizations. It collects and sorts information about the study design, the type of camera traps, location, species names in standardized way. That is important for comparing data between research, it may provide

3.6.2 Non-invasive genetic samples (NGS) - collection of evidence

The basics for collection of evidence are generally selecting certain transects in the target area and then repeated monitoring of these transects and collection of genetic samples from scats, urine, hair or dead prey. Transects are usually selected around trails, roads or shorelines as wolves use them for easier travel corridors (Marucco, Avanzinelli, and Boitani 2012; Kittle et al. 2015). The ideal season for collection of evidence is presumably the winter as there is much higher probability of detection of track in the snow but also urine and scats. But as noted by (I. Reinhardt et al. 2015) in some climate conditions as for example in central Europe, it is not possible to rely on snow tracking and winter monitoring in general but if the conditions are ideal, the opportunity should be used to maximize the output. However, selection of season for sample collection differs throughout the researches. The transect are generally walked on foot but it is no exception for vehicle being used (Moskowitz and Huyett 2019). I have not found any researches comparing effectivity of sample collection between walking and cycling or driving. But it is logical to assume that speed and restricted view on the road or trail limits the successful detection of scats etc. On the other hand, the effectivity of successful detection could be subject of further studying as it is possible to cover larger distances by vehicle and thus it could equalize the lower detection probability. For example, Llaneza, García, and López-Bao (2014) have mentioned vehicle use for monitoring purposes, but the speed was restricted to under 10 km/h.

3.6.3 Acoustic Detection

Wolves use howling for communication either for communication within the pack or with individuals and packs in the neighborhood. According to (Nowak et al. 2007), wolves howl most often from the center of their home territory and mainly to contact a separated member of the pack (43 %) or before the hunt (22 %) and after the pack aggregation.

The howling can be recorded with acoustic recorder and lately used for playback as wolves naturally tend to respond. Similarly, a human imitation of wolf howl can be used to identify the presence of the animal (Suter et al. 2017). Imitation of howling is also being used to confirm reproduction, but for this purpose, the method seems so far to be ineffective compared to other available methods (Reinhardt et al., 2015). However, this method is frequently used for pack number estimation, and it is confirmed that is can be used even for the identification of individuals (Llaneza et al. 2005). For pack number estimation, a method of a

visual examination of the wolf chorus – howling of a wolf pack – by spectrogram is used, and it has shown high effectiveness in identifying individuals. The study states in the results that with higher pack number raises the possibility of error as "phantom voices" or echoes. Another problem is that usually, only the first two or three wolves start to howl one after another, but the rest of the pack joins them together. However, for the chorus of two wolves, there was a 100 % success rate, and in a chorus of eight wolves, there was potential to recognize up to 6 individuals (Passilongo et al. 2015). So at least this method is quite reliable in predicting minimal pack size numbers. Moreover, acoustic detection combined with other methods as camera-traps can combine pretty well. Together they can validate pack size and also presence or absence of specific individuals who could be an advantage in the detection of poaching or migration, which are usually hard to confirm. Besides, the possibility of identifying a target individual is valuable, as it is almost impossible to do so with only use of camera-traps. The probability of wolf response to recorded howling is higher in summer and autumn due to the activity of the pups and presence of the pack in the home area (Harrington and Mech, 1982) (Llaneza et al. 2005); (Suter et al. 2017). Although, according to (Sadhukhan and Habib 2019) the howling intensity is higher in a pre-denning season as during the denning season wolf tends to be secretive. Another decisive factor is the range at which humans can hear the wolf howling. Multiple studies support the idea that the ideal distance is around 3 kilometers but also agree that, on some occasions, howling can be heard from 5 up to 16 kilometers (Harrington and Mech, 1982); (Llaneza et al. 2005). Undoubtedly whether and surrounding environment can affect how far the sound of howl travels. Rain, landscape relief, and snow can have a negative effect (Harrington & Mech, 1982). Because of the nocturnal activity of the animal night seems to be a better option for acoustic detection. In the study conducted by (Llaneza et al. 2005), most howling happened from 19:00 to 23:00. In the study by (Harrington and Mech, 1982), there is a recommendation to attempt to howl or play the playback several times as wolves do not necessarily answer to the first one but rather to the third one. Another study on subspecies of Wolf, the Indian Wolf (Canis Lupus Pallipes) shows that wolf response to playback of recorder howling if it was a chorus than solo howl (Sadhukhan and Habib 2019). The negatives of this method could be in confusion of wolf pack after hearing the stimulated howling as it could be recognized as invasive and change their behavior. Further, in areas when wolves are perceived negatively stimulated howling could evoke a negative response from the local human community (Llaneza et al. 2005). There is an option how to record and playback wolf howling without a momentous presence of a human via "howl-boxes" (Suter et al. 2017). Which supports the general idea that acoustic detection is not so financially or manually demanding as

other methods, but in a study conducted by (Llaneza et al. 2005) there is a recommendation for utilization of acoustic monitoring only in small areas as it would be ineffective or rather expensive in the greater ones.

3.6.4 Radio Telemetry

Radio telemetry is a method that provides the most accurate data. On the other hand, preparation and application are both complicated, so telemetry is generally used only for a small number of individuals. Mostly, for obtaining information about territory size, prey interactions, dispersal monitoring, habitat use, or mortality cause. Another important aspect of this method is that the capture and immobilization of the animal provide a detailed recording of morphological and genetic data, which would otherwise be impossible to get. GPS collaring used in Europe has brought some great success with monitoring of transboundary long-distance dispersals. Such events can provide essential information about connectivity among populations and also reveal migration corridors which animal use to move through the landscape. Additionally, that information can be later used by nature conservation organizations in strategical planning which might help in maintaining the functionality of these paths. This is extremely important in Europe fragmented landscape (Ciucci et al. 2009). The GPS collars can be programed to broadcast an actual position several times per day or just once in a week, depending on the objective. This is useful when following an activity patterns of predator and prey as, the collars can be programmed simultaneously and thus provide patterns of interaction (Eriksen et al. 2009)

Whereas the collection of evidence, surveys, or camera trapping are methods non-invasive, the telemetry with the use of GPS collars is perceived as highly invasive, and even though trapping can prove essential for research objectives, the public attitudes are often negative (Gese et al. 2019). The standard method for wolf capture is the use of foothold traps with the application of commercially-produced lures, canid urine, or food baits (Roffler, Gregovich, and Larson 2018). However, such a method can lead to a foot injury, which could affect the later animal movement, health, behavior, and thus the ability to survive (Gese et al. 2019). Therefore, reduction of capture-related injury and capture time should become a goal and standard for all researchers using these methods. There have been many modifications made to trapping hardware such as rubber padded traps, which significantly reduce the foot injuries, but on the other hand, rubber padded traps are more prone to escape of the animal (Frame, Paul F., Meier 2007). Additionally, more issues are to be found, for example, potential danger to domestic dogs or other animals and the risk of using foothold traps during the winter, which could lead to tissue freezing from reduction circulation of the restrained limb as stated in (Gese

et al. 2019). Overall the use of GPS collaring can produce essential data for a scientist; however, it is critical to be excellently prepared and provide a flawless risk assessment.

4. Practical Part

4.1 Study Areas

For this study, two target areas were selected in the north of the Czech Republic on the borders with Germany. The first study area was located in The Ore Mountains and the second one in the Bohemian Switzerland National Park (BSNP). Both areas had a somewhat similar size, but the BSNP area was slightly bigger. Both areas were specific by their terrain, habitat variety, remoteness, and touristic activity. In both areas, the same two methods with comparable parameters were used.

4.1.1 The Ore Mountains

The Ore Mountains study area had an approximate size of 40 km₂. The altitude ranged from 750 meters above sea level in lower parts up to 950 meters above sea level. In the lower elevation zones, the habitat was predominantly mixed forest, but it changed gradually to the coniferous forest with rising elevation. Apart from forests, large parts of the area were covered by wetlands. The area was also characterized by its relative remoteness, extended-lasting snow cover, and lower touristic activity. The high elevation areas with the wetlands were absolutely abandoned during the winter, which provides ideal conditions for wolf existence. Also, the combination of remoteness and absence status of the protected area resulted in much lower human activity compared to the Bohemian Switzerland National Park.

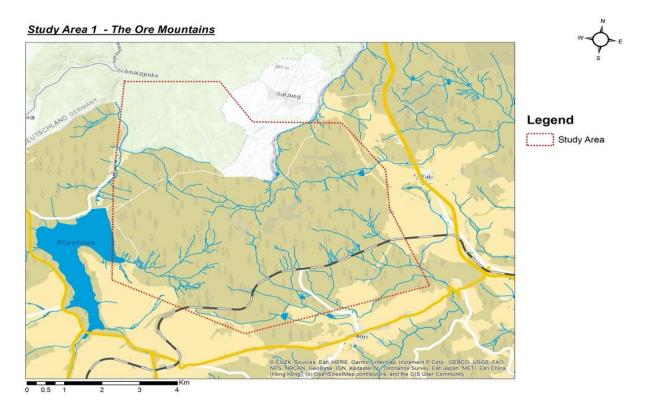


Figure 7: Study Area - The Ore Mountains

4.1.2 The Bohemian Switzerland National Park

The second study area was located inside the Bohemian Switzerland National Park. Presumably, state-protected areas should provide better habitat and overall beneficial conditions for wild animals, but because of high touristic activity, it may not prove right. The size of the area was 50km2, and habitat was mainly formed by coniferous forest. The elevation in this area was lower, with a range from around 350 to 450 meters above the sea level. This area was characteristic of massive rock formations spread over the whole area. The rock formations create valleys and gorges that are hard to reach and, therefore, practically abandoned, which provides excellent refuge for wild animals.

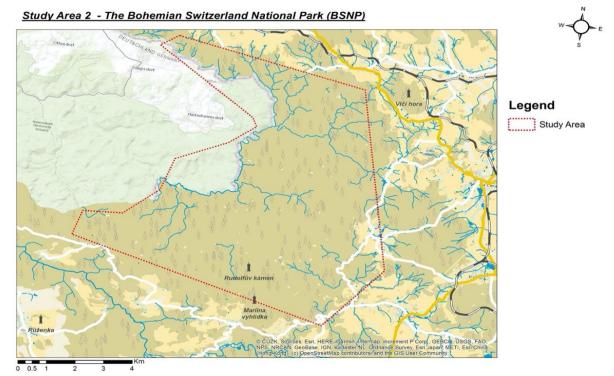


Figure 8: Study Area – The Bohemian Sitzerland National Park (BSNP)

4.2 Monitoring Methods – Ad. Hoc vs Fishnet Coverage

Two different methods were tested on both study areas in 3 repetitions. For both methods, mostly the "Spy Point FORCE-11D" were used but occasionally the "Spy Point Force Link Evo" for replacement. The first method, "Fishnet coverage," was based on a systematic random grid design and the second method, "Ad. Hoc" was based on the non-systematic placement of fewer camera traps on specifically selected sites. For the "Fishnet coverage," around 45 camera traps were used for an average of 1500 trap-nights and 10 camera traps for "Ad. Hoc" for an average of 310 trap-nights. The camera trap setting was: sensitivity – high, quality – high, delay – instant, multi-shot 3. With 24H time format, D/M/Year date format, and temperature in °C.

4.2.1 Fishnet Coverage method

A systematic grid was designed over the entire study areas and rotated to achieve the best overlay. Each point represented a camera trap site in each of the gird cells with an approximate density of 1,25km2. For the best site selection during the actual installation in the field, we added a 100m radius around each of the points and installed the camera traps within. For the Ore Mountains area, a total number of 40 camera traps were installed for each repetition. However, for the Bohemian Switzerland National Park 47 camera traps were set up in the first

campaign, 49 in the second repetition and 45 in the last one. More camera traps were used for this area to compensate for its bigger size and to keep equal density of camera trap sites.

Because the study areas were established only in forest habitat, the trees were selected for the camera trap placement. For the "Fishnet coverage" method, once the tree was selected, it was used for all of the three repetitions to keep the best possible integrity of the system.

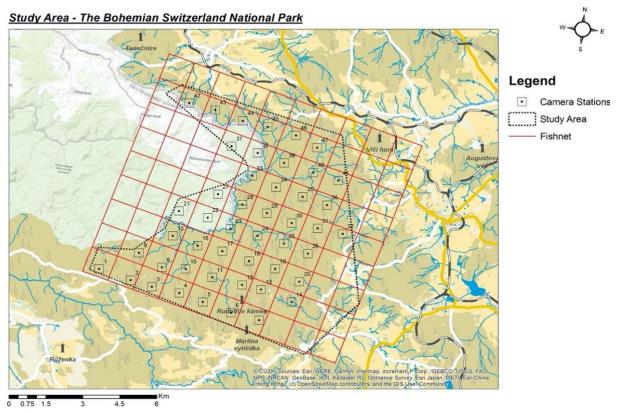


Figure 9: "Fishnet coverage method" - The Bohemian Switzerland National Park

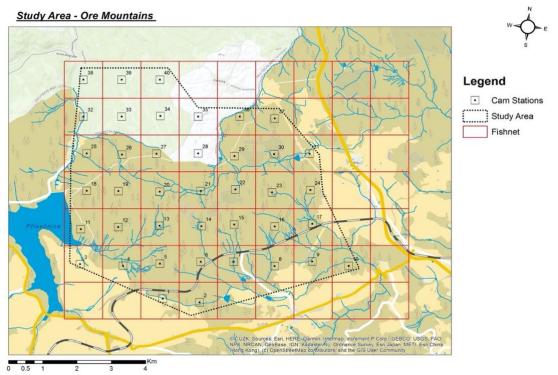


Figure 10 – "Fishnet Coverage" method - The Ore Mountains

4.2.2 Ad. Hoc method

The "Ad. Hoc" method is the exact opposite of the "Fishnet coverage". For this method, the camera trap sites are systematically selected based on information obtained about the area before the installation. Information gathered during regular monitoring such as the collected sample of evidence (scats, urine, etc.), information received from team members or photos captured by previous monitoring. The information obtained from the "Fishnet coverage" campaign was not used for designing of "Ad. Hoc" camera stations to achieve the best possible independence between the two methods. The camera stations in the "Ad. Hoc" designs mostly differed between the three different grid installations as more information was continuously collected, although some of the camera stations stayed on the exact same positions because of positive results depending on the actual situation. For example, the arrangement of the second and third designs in the Ore Mountains remained identical as it that the camera stations did not have expected results, but it was believed that they are well selected. Therefore, the same design was repeated to verify or deny the insufficient results.

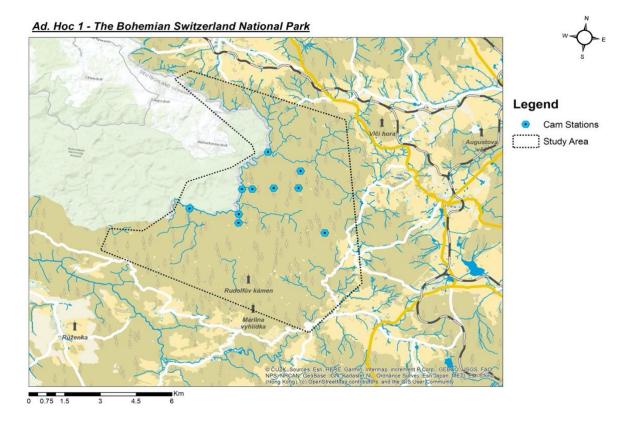


Figure 11: 1st "Ad. Hoc" method - The Bohemian Switzerland National Park

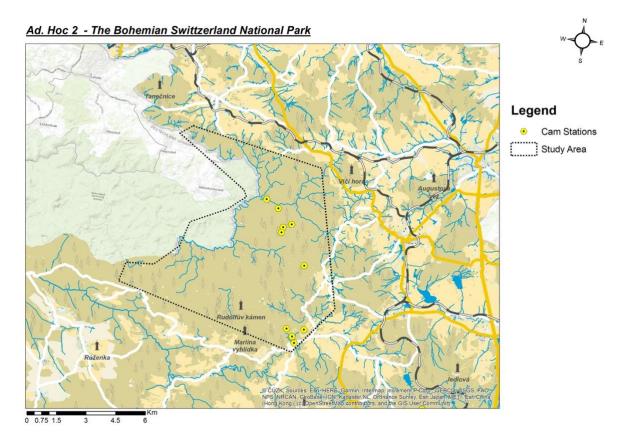


Figure 12: 2nd "Ad. Hoc" method - The Bohemian Switzerland National Park

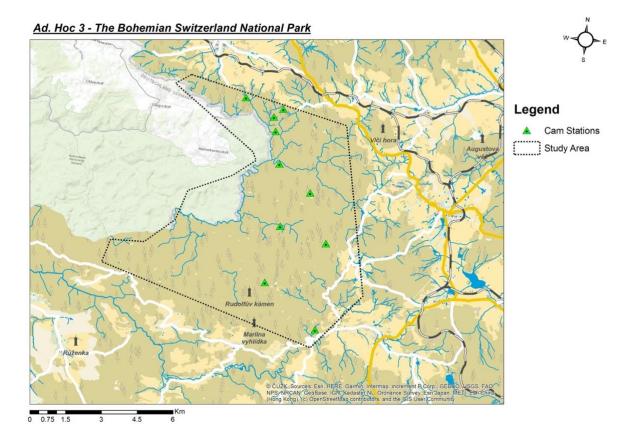


Figure 13: 3rd "Ad. Hoc" method - The Bohemian Switzerland National Park

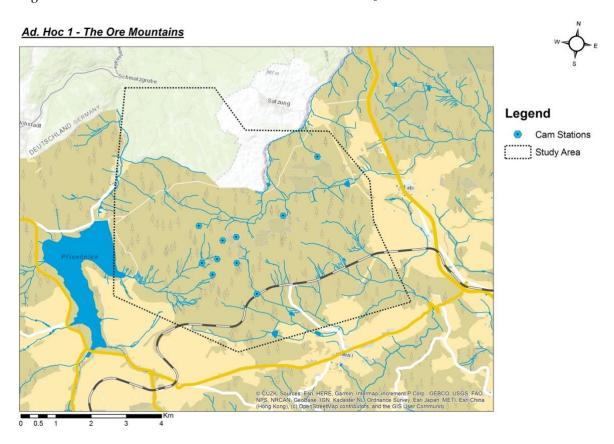


Figure 14: 1st "Ad. Hoc" method - The Ore Mountains

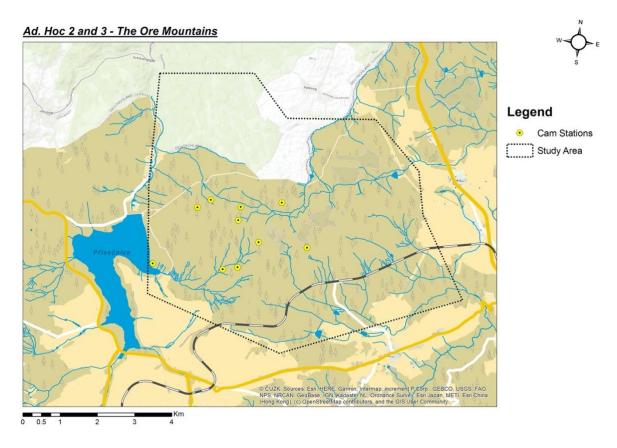


Figure 15: 2nd and 3rd "Ad. Hoc" method - The Ore Mountains

4.2.3 Data management

The study areas were covered by the grid, and each grid cell had a specific number. Each time when a camera trap was installed into a grid cell, it was given an identification number for more fluent orientation. Simultaneously excel worksheets were used to keep track of all the camera stations installed with the IDs being the primal factor for the storing system. In the excel worksheet, multiple information was saved. The first was the ID, second name of the camera trap, date of installation, date of collection, coordinates, grid cell number, initials of staff that set up the camera trap, number of trap-nights, cell for identification if the camera trap captured wolf and additional note.

| No | Trap/point ▼ | date of instalation | date of collection | X-coordinate 🔻 | Y-coordinate 🔻 | sector 🔻 | staff ▼ | trapdays 🔻 | wolf 🖪 | note | ~ |
|----|--------------|---------------------|--------------------|----------------|----------------|----------|----------------|------------|--------|-------|---|
| 1 | ? | 27/12/2017 | 10/02/2018 | 50,747449 | 14,947198 | 82 | JH | 45 | Yes | NP_ČŠ | |
| 2 | W14 | 23/12/2017 | 28/01/2018 | 50,8312 | 14,5119667 | 65 | AV | 36 | | NP_ČŠ | |
| 3 | W15 | 23/12/2017 | 28/01/2018 | 50,8319167 | 14,5114667 | 65 | AV | 36 | | NP_ČŠ | |
| 4 | W1 | 20/12/2017 | 04/04/2018 | 51,049307 | 14,298261 | 12 | LZ | 105 | Yes | NP_ČŠ | |
| 5 | W2 | 20/12/2017 | 26/01/2018 | 51,044515 | 14,295981 | 12 | LZ | 37 | | NP_ČŠ | |
| 6 | W3 | 20/12/2017 | 04/04/2018 | 51,032268 | 14,299659 | 12 | LZ | 105 | | NP_ČŠ | |
| 7 | W4 | 22/12/2017 | 30/03/2018 | 50,918774 | 14,421445 | 39 | LZ | 98 | | NP_ČŠ | |
| 8 | W5 | 23/12/2017 | 23/03/2018 | 50,948749 | 14,348542 | 29 | LZ | 90 | Yes | NP_ČŠ | |

Figure 16: Preview of OWAD's Excel worksheets for metadata organization

After the data were downloaded from SD cards inside the camera traps into the notebooks, it was stored in folders with the camera trapped ID and backed up on web storage. In the next step, the data trained workers downloaded the data and categorized it into the following subfolders: birds, mammals, people, and wolves. If no wolf photo was captured, the subfolder was renamed to "No_Wolves" so there would be no need to open the folder and check if photos are there or not. After all the camera stations were categorized, they were renamed by its date of installation, date of collection, and ID, as the following example - 2018_07_04-2018_09_24_136_R.

Data categorized and renamed were then stored on "My Cloud" where folders with grid cell numbers and the camera stations were put into those folders as multiple camera stations could be inside one grid cell.

4.2.4 Data analysing

Because the number of camera stations and trap-nights differed, "RAI – relative abundance index" and "naïve occupancy" were used to compare the data. "RAI" and "naïve occupancy" were counted for each repetition of both methods for the two study areas. The following formulas were used. Wolf event was identified as two different photos, separated by 60 minutes.

$$RAI = n$$
. of events/total n. of trap-nights * 100

N. occupancy = n. of camera traps that captured /total n. of camera stations

To determine which method had better overall results, they were compared in 5 different combinations by both "RAI" and "naïve occupancy". First, all data from "Ad. Hoc" and "Fishnet coverage" were compared, not depending on the study area. Next "Ad. Hoc" and "Fishnet coverage" data were compared for The Bohemian Switzerland National Park study area, and the same comparison was made for The Ore Mountains study area. Additionally, both "Fishnet coverage" and "Ad. Hoc" methods were compared between the two areas to find out if one of the methods functioned significantly better in one study area compared to the other.

To test

the normality of the data sets, the "Shapiro-Wilk normality test" was used. For the "RAI" dataset, the p-value = 0.05162 and therefor the dataset was tested by the unpaired Wilcoxon test for non-parametric data. In the dataset for "naïve occupancy," the p-value = 0.2925 and therefore unpaired T-test for parametric data was used.

5. Results

For the "Fishnet" method in the Ore Mountains 40 camera stations were used for each repetition with an average of 1440 trap-nights and it captured total of 31 wolf events. For the "Fishnet" Method in the Bohemian Switzerland 47, 49 and 45 camera stations were used with average of 1568 trap-nights and it captured total of 14 wolf events.

Fishnet - The Ore Mountains

| Study Area - repetition | Cam. Stations | Trap-Nights | Wolf Events | <u>RAI</u> | Naïve occupancy |
|-------------------------|---------------|-------------|-------------|------------|-----------------|
| Ore Mountains 1 | 40 | 1455 | 7 | 0,48 | 0,08 |
| Ore Mountains 2 | 40 | 1412 | 9 | 0,64 | 0,18 |
| Ore Mountains 3 | 40 | 1453 | 15 | 1,03 | 0,21 |

Fishnet - The Bohemian Switzerland National Park

| Study Area - repetition | Cam. Stations | Trap-Nights | Wolf Events | <u>RAI</u> | Naïve occupancy |
|-------------------------|---------------|-------------|-------------|------------|-----------------|
| BSNP 1 | 47 | 1321 | 3 | 0,23 | 0,07 |
| BSNP 2 | 49 | 2018 | 7 | 0,35 | 0,12 |
| BSNP 3 | 45 | 1367 | 4 | 0,29 | 0,08 |

Figure 17 - The Fishnet method data for both study areas

For the "Ad. Hoc" method in the Ore mountains 10 camera stations were used for each of the repetitions with an average of 333 trap-nights and it captured total of 12 wolf events. For the "Ad. Hoc" method in the Bohemian Switzerland also 10 camera stations were used for all of the repetitions with an average of 296 trap-nights and it captured total of 15 wolf events.

Ad. Hoc - The Ore Mountains

| Study Area - repetition | Cam. Stations | Trap-Nights | Wolf Events | <u>RAI</u> | Naïve occupancy |
|-------------------------|---------------|-------------|-------------|------------|-----------------|
| Ore Mountains 1 | 10 | 340 | 6 | 1,76 | 0,3 |
| Ore Mountains 2 | 10 | 340 | 4 | 1,17 | 0,2 |
| Ore Mountains 3 | 10 | 320 | 2 | 0,62 | 0,2 |

Ad. Hoc - The Ore Mountains

| Study Area - repetition | Cam. Stations | Trap-Nights | Wolf Events | <u>RAI</u> | Naïve occupancy |
|-------------------------|---------------|-------------|-------------|------------|-----------------|
| BSNP 1 | 10 | 310 | 2 | 0,64 | 0,2 |
| BSNP 2 | 10 | 260 | 7 | 2,69 | 0,4 |
| BSNP 3 | 10 | 320 | 6 | 1,87 | 0,3 |

Figure 18 - The Ad. Hoc Method data for both study areas

Results showed that "Ad. Hoc" method has overall higher "RAI" values, although "Fishnet Coverage" captured more events, which was caused by a higher number of camera stations and trap-nights. When "naïve occupancy" data were compared, similar results were achieved - the "Ad. Hoc" method had higher overall values. The third "Fishnet coverage" repetition in the Ore Mountains had the highest "RAI" and "naïve occupancy" values from all "Fishnet coverage" repetitions. The biggest values with "Ad Hoc" method was achieved during second repetition in The Bohemian Switzerland. However, when data were tested, multiple p-values did not show enough significance between the data, so the null hypothesis – that both methods would provide similar results could not be rejected.

5.1 Relative abundance index – "RAI"

When comparing the methods without the significance of study areas, the alternative hypothesis was that the "Ad. Hoc" method would have significantly higher values than the "Fishnet coverage" method.

RAI - CLUMPED DATA

RAI - Fishnet 0,48 0,64 1,03 0,23 0,35 0,29 RAI - Ad. Hoc 1,76 1,17 0,62 0,64 2,69 1,87

Figure 19 - RAI clumped data

When all "RAI - Ad. Hoc" data was compared to all "RAI - Fishnet coverage" data; results were expected to show higher values for the "Ad. Hoc" method. Tested by non-parametric Wilcoxon test with p-value = 0.02472 the null hypothesis could be rejected.

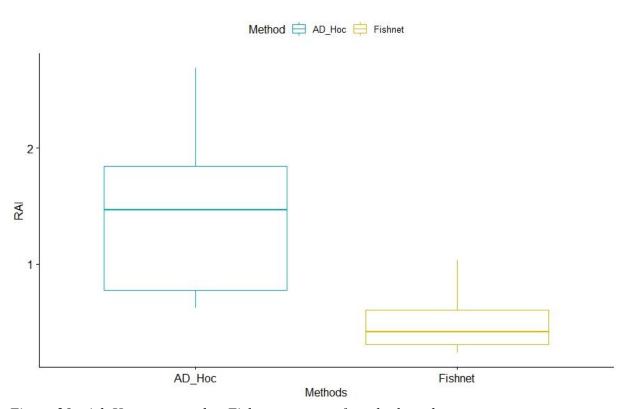


Figure 20 - Ad. Hoc compared to Fishnet coverage from both study areas

When both methods were compared in The Bohemian Switzerland National Park study area, the "RAI" values of "Ad. Hoc" method were 5.9 times higher than the values of "Fishnet" method. With overall "RAI" values for "Ad. Hoc" = 5.2 and overall values of "Fishnet" = 0.87. Nevertheless, when tested by non-parametric Wilcoxon test, the p-value = 0.1, and therefore the null hypothesis could not be rejected.

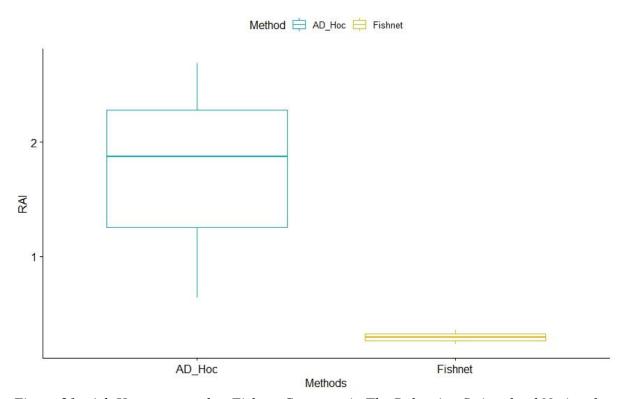


Figure 21 - Ad. Hoc compared to Fishnet Coverage in The Bohemian Switzerland National Park

When the same comparison was tested for the Ore Mountains study area, the same alternative hypothesis was expected. The differences were not so extensive, as in the Bohemian Switzerland National Park because the "Fishnet coverage" method had overall better results in the Ore Mountains. However, when tested by non-parametric Wilcoxon test, the p-value = 0.4, and therefore the null hypothesis could not be rejected.

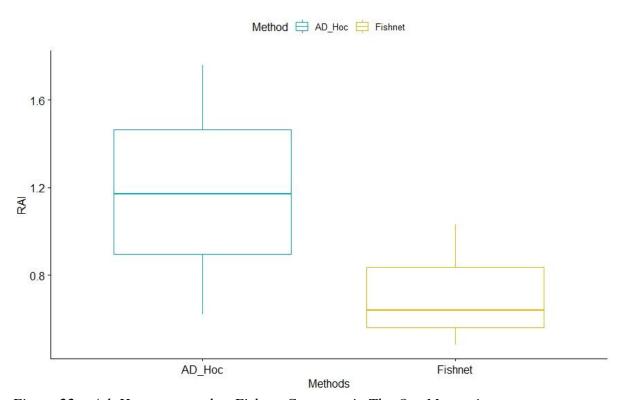


Figure 22 - Ad. Hoc compared to Fishnet Coverage in The Ore Mountains

The "Ad. Hoc" data from the Bohemian Switzerland National Park were compared to "Ad. Hoc" data from the Ore Mountains, to find out if there was a significant difference between the two study areas when the same methods were compared. The "Ad. Hoc" method performed better in the Bohemian Switzerland National Park, but p-value = 0.4, so it was concluded that data do not show any significant difference. The same process was done for the "Fishnet coverage". The results of this method were compared between the two areas. "RAI" values for "Fishnet coverage" were higher in the Ore mountains, but the non-parametric Wilcoxon test showed p-value = 0.1, and therefore the null hypothesis could not be rejected.

"Ad. Hoc" method had better results in the Bohemian Switzerland National Park. With "RAI" values of 2.69, 1.87, 0.64 in the BSNP and the "RAI" values of 1.76, 1.17, 0.62 in the

Ore Mountains. Contrary to the "Fishnet coverage," which produced better results in the Ore Mountains.

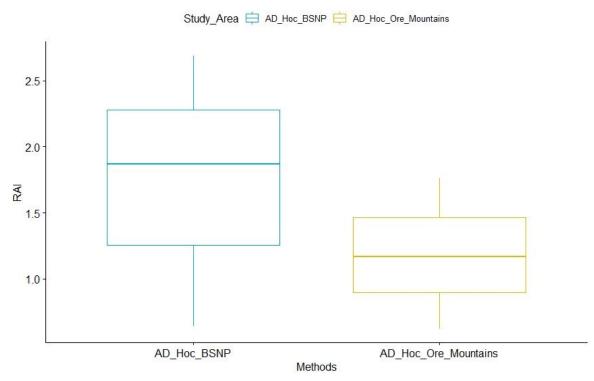


Figure 23 - Comparison of Ad. Hoc between the two study areas

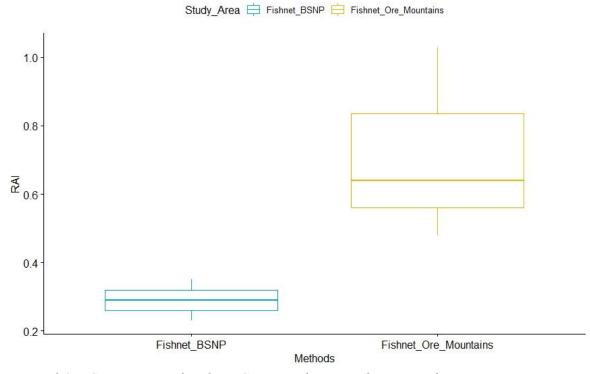


Figure 24 - Comparison of Fishnet Coverage between the two study areas

5.2 Naïve occupancy

The same comparison as for the "RAI" data was also made for the "naïve occupancy" data. Because "Ad. Hoc" values were higher than the "Fishnet coverage" values; the same alternative hypothesis was expected.

When all "naïve occupancy - Ad. Hoc" data was compared to all "naïve occupancy - Fishnet coverage" data, results confirmed higher values of the "Ad. Hoc" method. When tested by T-test for parametric data, the p-value = 0.006747 the null hypothesis could be rejected.

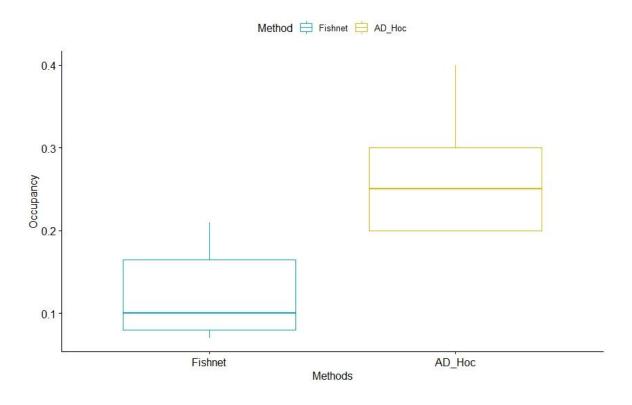


Figure 25 - Ad. Hoc from both study areas compared to Fishnet coverage from both study areas

When both methods were compared in The Bohemian Switzerland National Park study area, the "naïve occupancy" values of "Ad. Hoc" methods were 3.3 times higher. With overall "naïve occupancy" values for "Ad. Hoc" = 0.9 and overall values of "Fishnet" = 0.27. The results of parametric T-test showed the p-value = 0.5962, and therefore the null hypothesis could not be rejected as data did not show significant difference.

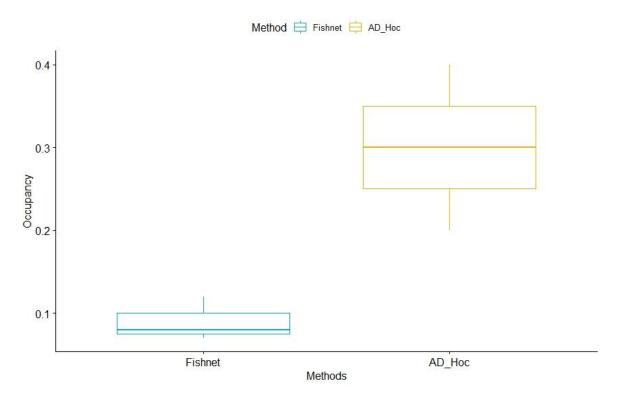


Figure 26 - Fishnet compared to Ad. Hoc in BSNP

When the same comparison was tested for the Ore Mountains study area, the same alternative hypothesis was expected. The prediction was that the "Ad. Hoc" method would perform better than the "Fishnet coverage" method in both study areas. The differences were not so extensive, like in the Bohemian Switzerland National Park because the "Fishnet coverage" method provided better results in the Ore Mountains for both "RAI" and "naïve occupancy". However, when tested by parametric T-test, the p-value = 0.2129, and therefore the null hypothesis could not be rejected.

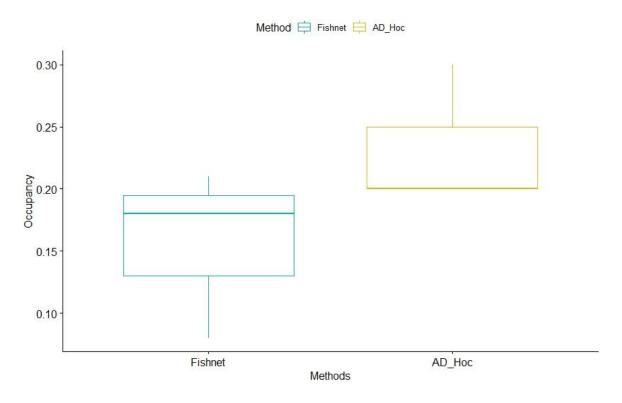


Figure 27 - Fishnet compared to Ad. Hoc in The Ore Mountains

The "Ad. Hoc" data from the Bohemian Switzerland National Park were compared to "Ad. Hoc" data from the Ore Mountains, to find out if there was a significant difference between the two studies when the same methods were compared. The "Ad. Hoc" performed better in the Bohemian Switzerland National Park, but p-value = 0.2259, so it was concluded that data don't show any significant difference.

The same process was done for the "Fishnet coverage". The results of the methods were compared between the two areas. "Naïve occupancy" values for "Fishnet coverage" were higher in the Ore mountains, but the test showed p-value = 0.3868, and therefore the null hypothesis could not be rejected.

However, this comparison showed that the "Ad. Hoc" method had better results in the Bohemian Switzerland National Park contrary to the "Fishnet coverage", which produced better results in the Ore Mountains. Values of "naïve occupancy" of the "Ad. Hoc" in the Bohemian Switzerland national Park = 0.3, 0.2, 0.2 and values of "naïve occupancy" of the "Fishnet coverage" in the Ore Mountains = 0.08, 0.18, 0.21.

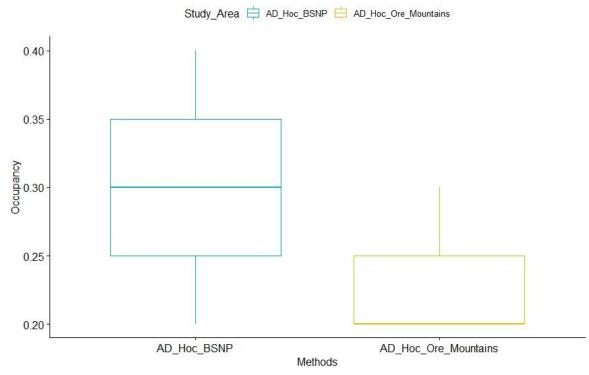


Figure 28 - Comparison of Ad. Hoc between the two study areas

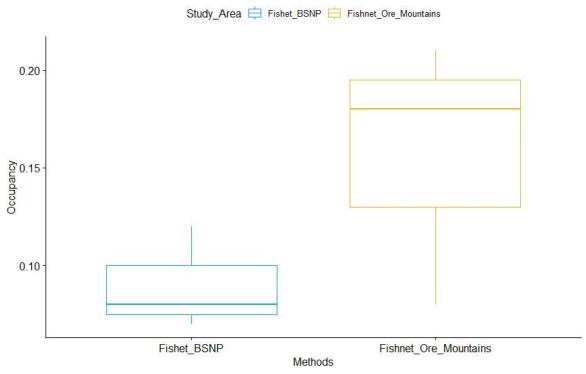


Figure 29 - Comparison of Fishnet coverage between the two study areas

6. Discussion

As the wolf population is rising in Central Europe, efficient and precise monitoring is important. Therefore, the selection of the right method will determine the success of the research. Based on the results and experience from this thesis, suggestions for maximizing potential of camera trapping can be made.

If the objectives of the research would be focused on constant monitoring of the wolf pack about which there is enough prior information, the "Ad. Hoc" method seems to be a more suitable choice as it provides better results with lower time and energy effort. Although, for success of this method sufficient knowledge about the target area and the wolf pack activity is essential. Therefore, when there is no prior information – for example if potential new wolf territory is discovered, it makes more sense to use the "Fishnet coverage" method and place the grid over the whole area. This process should provide more complex information about the area and about the wolf activity and could have better practical use in future conservation efforts. Additionally, the "Fishnet coverage" produces further information about the behavior of the wild animals and about the area itself, which could be helpful in the area management. For example, the data from The Bohemian Switzerland National Park could be used by the park service for a better understanding of local processes and the implementation of appropriate measures. However, the installation of camera trap grids can be and usually is a physically challenging job depending mostly on a number of camera traps and area terrain. For example, a study in Bhutan camera placement in a steep landscape with an elevation around 3000 m, defined the installation as "labor-intensive" and time-consuming (Sangay, Rajaratnam, and Vernes 2014). Interesting results were shown by the comparison of the two methods between the areas. It is shown that the "Fishnet coverage" method had better results in the Ore Mountains than in Bohemian Switzerland. That was probably caused by better positioning of the grid in the Ore Mountains study area as it is believed that the grid overlapped the core territory of the pack. On the other side, in the Bohemian Switzerland the "Fishnet" grid was placed in between 3 existing wolf territories, but in the area where none of the packs had its core.

Furthermore, every tool or equipment has its advantages and disadvantages. We faced several technical issues with the correct placement, settings of the camera traps and data analyzing.

Camera traps are usually placed with its field of view on rarely used roads, dirt roads, game trails, shorelines, river-streams, or wildlife crossing structures. However, some additional factors can be found. One such important factor is the camera field of view; for example, when installing the camera trap near the forest road, it is beneficial to situate it on a crossroad as

chances for capture are presumably higher.

From our experience whenever the wolf crossed the detection zone, the camera trap was able to capture it just once, despite the setting of the trigger delay set on the instant. Nevertheless, that was not the rule for each of the camera stations as they did not have the same distance from the targeted site. In other words, those who had a narrower view would need higher trigger speed.

Regarding the advantages of the camera trap use. The primary advantage is its relative independent function, although some maintenance such as battery change, memory capacity control so routine checks are necessary. However, from our experience, battery life and memory capacity with modern technology are sufficient enough; hence camera traps can function more than two months in the field, and maintenance checks can be done when collecting data. Other advantages are the possibility of long-term monitoring with continuous observation, non-invasive use with strictly standardized and repeatable potential. The replicability of the methods is crucial as different teams can use the exact methods and expect almost exact results, which is harder to accomplish when humans are used as the detectors (Wearn, Oliver, Glover-Kapfer 2015).

Finaly, the educational aspect of nature conservation is more and more perceived as essential as raising awareness can help to accomplish the achievements, and in some cases, the engagement of residents can be beneficial. Camera traps often produce fascinating photos and a unique view into the secret lives of wild animals, which can be used as a tool to connect with local communities (Wearn, Oliver, Glover-Kapfer 2015).

One of the most significant disadvantages is the malfunction or theft, because the researcher is left out without the data. If the malfunction is not result of wrong settings as low trigger sensitivity, wrong trigger delay or leaving the camera trap in the field without turning it on, there is not much that could be done about the malfunction, but it is essential to mark the camera trap so it would not be used again. While in case of theft, there are multiple measurements one can take to lower the theft probability. We, for example, used wires with locks to decrease the chance of the camera traps being stolen, and additionally, we marked the camera traps with text stickers explaining the purpose. Another frequently used option to avoid the theft of camera traps is higher camera trap placement. However, the exact height can be decisive. A study by (Meek, Ballard, and Falzon 2016) compared the detection efficiency of two specific heights for camera traps. They placed camera traps in 0.9 meters and 3 meters height; the result showed a decline in detection rate in the higher placed camera traps. Meaning,

that it is possible that researchers in an attempt to save their camera traps may unknowingly sacrifice some of their data. We, on the other side, intentionally placed camera traps higher in the later period of the research as we noticed that the flash in the camera traps disturbs the animals during a night. Moreover, from our experience, the high placement of the camera traps does not guarantee its safety. And as mentioned in (Rovero et al. 2013) detailed study comparing multiple camera traps, there is a statement that no camera trap remains absolutely unnoticed by animals as even most professional "no glow" flashes are noticed or heard by animals. Lastly, another significant drawback can be the financial aspect. The first investment can be a problem for many scientists as the cost of camera traps moves around 300-500€. For an extensive study, the needed number of camera traps can come close to 50, which is a significant investment. Although, the cost of operational labor as installation or maintenance checks are relatively low the later data analyzing and image storing can also be costly (Wearn, R. Oliver, Glover-Kapfer 2015). The methodological standards for camera trap data analyzing and image storing are still being developed. Moreover, from our experience keeping the correct schemes for organization and precise workflow is challenging but essential.

Firstly, when the memory cards from camera traps are downloaded into computer it is crucial to properly label a folder for each camera trap. It is useful to back up the data once more before formatting the memory card. The formatting of the memory card should be neglected because it prevents chaos in future use. Managing your metadata – information about work you do on your actual data is another important and frequent instruction. Because in such time consuming and multi operational task it is hard to keep track on everything that has been done, particularly if more people involved. Therefore, documenting every step that has been done – creating metadata - when managing photos/videos is key in keeping clear order in the database (Wearn, R., Oliver, Kapfer-Glover 2017). There are many specifically-designed camera trap software for more effortless management of the data. There are multiple choices for managing camera trap images, managing databases or metadata. For example "MapView", "DeskTeam", "CameraBase" and many others (Athreya et al. 2014). Although, they may be costly, hard to understand especially for the whole team and therefore no particular software was adopted by camera trapping specialists. Also for extensive camera trapping projects it is uneasy to identify one software package that would cover everything and it is not uncommon that teams design their own system or software (Wearn, Oliver, Kapfer-Glover 2017).

7. Conclusion

The "RAI" and "naïve occupancy" results showed the ability of "Ad. Hoc" method to capture more wolf events. Furthermore, the "Ad. Hoc" method uses a lower number of camera stations, and thus, the installation and photo management are less time demanding. However, it should be noted that the successful selection of prosperous camera stations requires knowledge about the targeted area and information about wolf activity. The time and effort to successfully create an effective "Ad. Hoc" design was not included in testing and could influence the final effectivity of the examined methods as the "Fishnet coverage" grid can be designed with no information about the area and still achieve positive results. Moreover, the advantage of the "Fishnet coverage" is the complexity of spatial information it provides about the area. The method not only displays areas where wolves are active, but it will also show areas which wolves do not use, and that information can be used in following changes of the design or for other monitoring activities whereas the "Ad. Hoc" method will only provide information about the areas where wolves were active.

The main downside of the "Fishnet coverage" method is a large number of needed camera traps and relatively high time and energy effort. Essentially, the combination of the two methods could create a robust and complex monitoring system for wolf monitoring in the future. The suggestion is to use both methods for discovering and later monitoring of new wolf territories and wolf packs. When information about new territory is obtained, the "Fishnet coverage" method should be used to verify the presence of wolves and if proven right, the data could be used to create a robust "Ad. Hoc" design, which would then be used for constant monitoring of the wolf pack. Moreover, if system like this would be implemented and unified by more scientist the final data would be easily comparable and verifiable. However, it should be definitely noted that none of the methods will work without parallel field monitoring and collection of evidence for genetics analyzes. Furthermore, field monitoring provides essential information for the creation of "Ad. Hoc" design and cannot be easily replaced.

In the end, it should be stated that, for the design of the "Ad. Hoc" method only information from field monitoring and other "Ad. Hoc" photos were used. However, because the "Fishnet coverage" method was placed in the same areas, it provided useful information about the areas. Therefore, two different teams would be needed to separate the two methods fully and to achieve the best integrity for the testing.

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