Following the footsteps of the first rewilding European bison in southwest Romania

On the habitat use of the free bison
in the Țarcu Mountains of the Southern Carpathians

Internship report by Nick Huisman
MSc Biology 2016

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Internship report on WWF Romania's 'Magura Zimbrilor' project

MSc Biology
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Abstract

The European bison (*Bison bonasus*) is the largest wild herbivore in Europe that balanced on the brink of extinction almost a century ago. Nowadays, reintroduced bison herds are found across a limited number of suitable habitats in this Europe. The Carpathians provide a suitable habitat to establish new reintroduction sites across several Eastern European countries. In the Southern Carpathians WWF Romania has founded the Bison Hillock project, releasing a new group of bison into the Romanian wilderness every year. During the vegetative season of 2016 the habitat use of the first group of 20-odd bison that was released in this area was studied. The data was gathered from a GPS collar, camera traps, transects and indirect observations of bison-activity. Results from GPS data on 150 days in the wild show that the collard bison, called Romanita, spent most of her time (71,2%) in pastures. Other data showed that her average travelling speed was significantly higher during the day (167,8 m/h), compared to the night (66,8 m/h). Romanita spent all her time between 563 and 1000 meters altitude, preferring altitudes around 650m, 700-770m, and 860m. The camera traps filmed 47 bison individuals during 12 different moments, in both pasture and forest habitat. Other wildlife species that were captured on camera were Roe deer (*Capreolus capreolus*; 17 ind.), Wild boar (*Sus scrofa*; 20 ind.), Brown bear (*Ursus arctos*; 2 ind.), Fox (*Vulpes vulpes*; 2 ind.), Marten (*Martes martes*; 1 ind.), Red deer (*Cervus elaphus*; 15 ind.), and Greater mouse-eared bat (*Myotis myotis*; 1 ind.). Calculated bison densities based on the results of the transects, using the Faecal Accumulation Rate method, were comparable with bison densities in other reintroduction areas. Combining all GPS points from indirect observations and Romanita's locations, yielded a heatmap that indicates that 'hotspots' for bison-activity in the study area. The bison-activity was limited to an area of approximately 32 km^2, so it is believed that the bison group will be exploring the area more in the coming years.

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1 | Introduction

The European bison (*Bison bonasus*) is the largest living wild mammal in Europe, primarily found in the protected mountainous areas across several eastern European countries. Once, the bison was widespread and abundant in Central and Eastern Europe, but due to habitat destruction, deforestation and hunting the number of bison declined drastically during the last centuries and the last wild European bison was killed in 1927 in the Caucasus Mountains (Krasinska & Krasinski, 2002; Kuemmerle et al., 2016; Kuemmerle et al., 2012). The only remaining 54 bison, descendants of merely 12 individuals, were found in zoos and breeding enclosures (Krasinska & Krasinski, 2002; Perzanowski & Olech, 2007). Balancing on the brink of extinction, the bison were put through to an intense genetically based breeding and reintroduction program (Kuemmerle et al., 2011; Olech & Perzanowski, 2002). Now, almost a century later, the total number of bison is estimated around 3,100 individuals, of which nearly 2,000 roam free in the European wilderness (Kuemmerle et al., 2016; Perzanowski & Olech, 2007).

Historic observations describe the European bison as a forest inhabitant, but recently more evidence is showing that the bison is also largely dependent on grasslands and open meadows. It was found that the bison are actually mixed feeders, consuming both grass and browse, which can be found in forest-field habitats (Kuemmerle et al., 2010). It is now thought that the bison used the forest habitat as a refuge from human contact, when habitat degradation was increasing with human population growth (Bocherens et al., 2015; Krasinska & Krasinski, 2002). Allowing the bison to roam only in forest areas would restrict them from accessing their optimal habitat, classifying them as a refuge species that decreased in fitness and population density as a result of their suboptimal surroundings (Kerley et al., 2012). Looking at the habitat preferences of the bison that presently roam free in the Carpathians supports this theorem. Wołoszyn-Gałęza et al., (2016) studied the habitat preferences of the bison in the Polish Carpathians and found that bison prefer coniferous forest areas close to natural open grasslands, and stands with broken canopy during the vegetative season. However, other studies on the habitat of the Polish and Belarusian bison herds show that the bison were found mainly in forest habitats and did not prefer coniferous forest to mixed or deciduous forest. The amount of accessible water however is affecting the preference for a certain forest type (Daleszczyk, 2007; Kuemmerle et al., 2010). The rage of altitudes where bison prefer to stay, appears to be between approximately 400 to 1000 meters during the vegetative season (Schmitz et al., 2015; Wołoszyn-Gałęza et al., 2016). Taking this into consideration, I expect that the rewilding bison in the Southern Carpathians will be found around the edges of forest habitats, within relative close proximity of a stream or river up to 1000 meters altitude.

Studies on habitat use have used different non-invasive methods to gather wildlife monitoring data. Several studies on the habitat use of the European bison in the Carpathians have used radio-telemetry with GPS-locations as a method to determine bison occurrence (*e.g.,* Kowalczyk et al., 2013; Kuemmerle et al., 2010; Schmitz et al., 2015; Schneider et al., 2013; Wołoszyn-Gałęza et al., 2016). As GPS data was available on the rewilding bison, this method was included to determine the habitat use in the study area. For other studies, camera traps have proven to be a useful and non-invasive tool to study the presence and habitat use of different animal species (*e.g.,* Borchard & Wright, 2010; Bowkett et al., 2008; Long et al., 2008). The limitation of this method is the accuracy of the camera traps, dependent on the detection distance. This is the maximum distance at which the camera is triggered to take a photo or video. Depending on the type of animal, habitat cover, and time of the day detection distance may vary throughout the study period. As there were five camera traps available for this study, this method was also used to collect data on the habitat use of the bison. And a third way to monitor the presence of an animal species is by using an indirect observational technique, called pellet group counts (McClanahan, 1986; Neff, 1968). There are two methods, *e.g.*, the faecal standing crop (FSC) and faecal accumulation rate (FAR). Both methods use transects to count faecal pellets, where FSC uses defecation and decomposition rates to calculate the number of animals, while FAR replaces the decay rate with a specific time interval (Alves et al., 2013; Campbell et al., 2004; Torres et al., 2012). The defecation rate of bison in captivity was studied by Eycott et al., (2013), and results showed that during the vegetative season bison defecated 7.5-8 times during the day. As decay rate of dung is dependent on local habitat and environmental circumstances, a study is required to obtain this location specific information for the FSC method. Therefore, the FAR method is more widely used and was the third method to gather data on the habitat use of the bison for this study (Campbell et al., 2004). The fourth and final way in which data was collected, was by recording indirect observations of bison-activity. Although this method is not scientifically randomized, it can indicate areas where much activity has been found and therefore the habitat use might be higher.

This study is thus combining GPS locations from dominant bison, videos that confirm the presence/absence of bison in a certain habitat area, and identification of multiple types of indirect signs of bison-activity throughout the study area. This data is used to map which habitats are preferred by the first 20-odd bison that are roaming free in the Southern Carpathians.

2 | Methods

2.1 Study area

Studies on potential suitable habitats for reintroduction sites show that the Carpathian mountain range provides a suitable environment where a rewilding bison population can be established (Kuemmerle et al., 2010; Kuemmerle et al., 2016; Ziółkowska et al., 2012). The Carpathians are with approximately 210.000 km^2 Europe's largest mountain range. They cross eight Eastern European countries, *i.e.*, Austria, Czech Republic, Poland, Slovakia, Hungary, Ukraine, Romania and Serbia. During the first decade of the 21st century, six herds of free-roaming bison were established in the Carpathian mountains of Poland, Slovakia and Ukraine (Kuemmerle et al., 2010). Only recently Romania has founded reintroduction sites for the European bison, coordinated by Rewilding Europe and WWF Romania in collaboration with other local parties (Vlasakker, 2014). For my internship I joined the WWF bison rewilding project 'Magura Zimbrilor', the 'Bison Hillock' project in Romania.

The Bison Hillock project is situated in the south-western Caraş-Severin district of Romania (Figure 1). Stationed at the village of Fenes near the commune of Armeniş, the project in the Southern Carpathians is located at the edge of the protected Munții Țarcu Natura 2000 Site (EEA-Copenhagen, 2014). The Munții Țarcu, which can be translated to Țarcu Mountains, cover an area of approximately 58.600 ha between N 45°12' and 45°28' latitude and E 22°25' and E 23°37' longitude. It's altitudes vary roughly between 350 and 2100 meters above sea level and as a protected nature area, it hosts over 40 threatened floral and faunal species of which at least 28 are protected (EEA-Copenhagen, 2014; Kukula et al., 2003). The region of the Țarcu Mountains is composed out of two biogeographical regions, *e.g.*, continental and alpine regions. A more detailed map on land cover types shows that the study area holds the following categories: principally occupied land by agriculture, transitional woodland-shrub, broad-leaved forest, mixed forest, and coniferous forest (Appendix 1; EEA-Copenhagen, 2014; Munții Țarcu & Asociatia Altitudine, 2016).

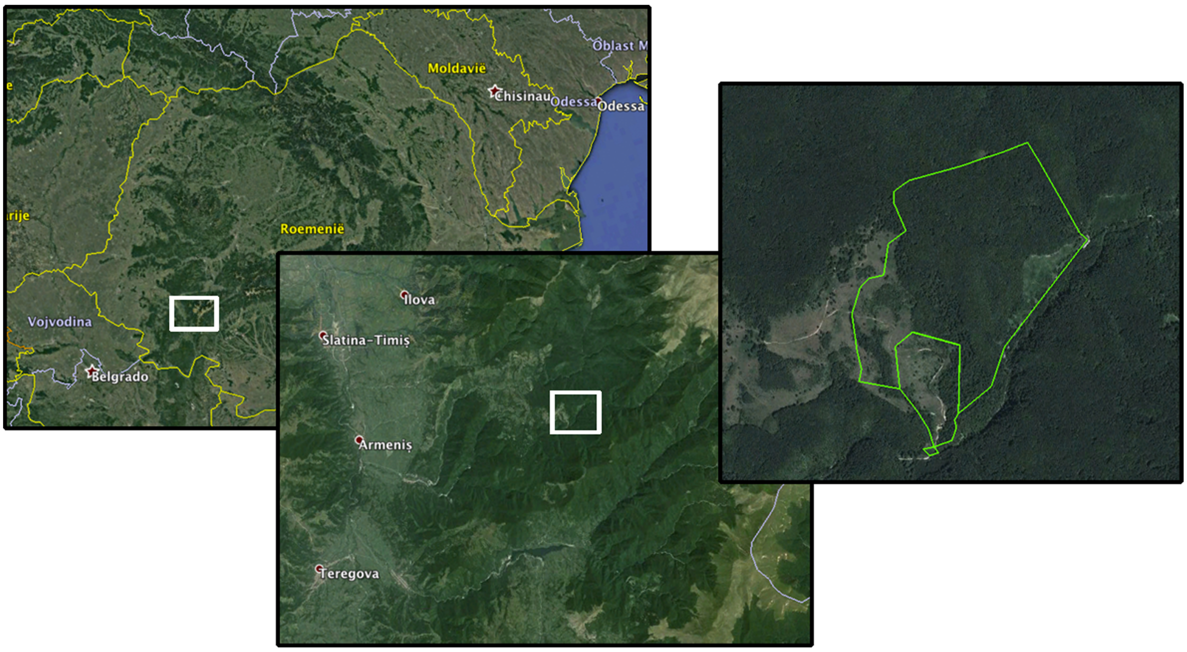

Figure 1 | The location of the Bison Hillock enclosure in the Caraș-Severin county, Romania.

Since 2014 a new group of bison, originating from breeding centres and zoos across Europe, is brought to this area every year and released into a constructed 'bison enclosure' in the valley of Plopu. The bison enclosure encompasses three areas, which are connected by manual gates. The veterinarian enclosure is a small area where bison are fed during their first month and individuals can be quarantined if necessary. Connected to this area is the first 'acclimatization zone', a 14-hectare enclosure where the new bison can adapt to the new environment. The next and biggest enclosure is the 'rewilding zone' of 135 hectares that enables the bison to explore their surroundings and become familiar with different habitats like open grass fields and forest areas. Both acclimatization and rewilding zones are surrounded by an electric fence. After spending approximately two months in the bison enclosure, the gates are opened to let the bison roam free and explore the 20 km^2 Plopu valley and beyond towards to Țarcu Mountains. As I focussed my study on the habitat use of the free-roaming bison, the valley and surrounding hill slopes were my study area.

2.2 Experimental setup

The fieldwork for this study took place from June to September 2016, during the vegetative season. In the beginning of June, WWF Romania was granted permission to release a group of 20-odd European bison from the bison enclosure into the wilderness. These bison were released into the enclosure in 2014 en 2015, but were unfortunately not released earlier due to paperwork. To study their movements and habitat use, I used the different available methods to collect data. A Garmin Oregon 650 GPS device was used to navigate through the valley, pinpoint the randomized locations of the camera traps and transects and georeference all (in-)direct observations of bison-activity.

2.2.1 GPS collared data

Other studies on the bison habitat preference used 3-10 GPS collars. This project had access to only two collars that were used from 2015. A dominant female and male from the groups that were released into the bison enclosure in the second year of the project, named 'Romanita' and 'Birk' respectively, were collared in May 2015 with a GPS transmitter. The transmitter sends a signal every two hours, including the coordinates, a fix category, *e.g.*, 2D/3D/3D-V or 'No satellites', and a value for dilution of precision (DOP), measuring the accuracy of the data. Highest accuracy coincides with higher fix categories, as a 2D fix is created with only three satellites and a 3D(-V) fix with four or more satellites, and accuracy increases with lower DOP values. Therefore locations without satellites, or 2D and DOP>5 were excluded from further analysis (Frair et al., 2010; Lewis et al., 2007). Data collection on GPS coordinates was retrieved from the Lotek webservice (Lotek Wireless Inc., 2016). The coordinates were categorized in the different habitat types, in which they were found. Further, straight-line distances between consecutive GPS locations were calculated and averaged to travelling speed. Travelling speed (in meter/hour) was plotted for day and night time per month. To see if the bison had specific heights at which it prefers to reside, the altitude was also plotted over time. These heights were compared to the general altitude map of the surrounding area to find possible alternatives where the bison might go (Appendix 2).

2.2.2 Camera trapping

A total of five different camera traps (Moultrie M-880 Mini Game Camera) were randomly distributed throughout the Plopu valley (Appendix 3). An online coordinate randomizer was used to determine the locations for the camera traps. A suitable specific tree was chosen to set up the camera trap within a 20-meter radius from the randomized point, after agreement with the local landowners. As not every local supports the presence of camera traps, some cameras were slightly relocated to a near location that wouldn't create problems. Eventually two camera traps were deployed in a pasture-habitat, while the other three were deployed in a (open) forest-habitat. Videos were stored on 16GB Adata SD cards that were collected in three sets of approximately 21 days. Due to circumstances, one camera trap had to be used for another study, but the remaining four camera traps were functional during the entire study period. Camera trap data was used to determine if bison would use this specific area within the valley. Other wildlife species that were recorded, were noted down and the data was stored to be used for future wildlife monitoring studies in the project.

2.2.3 Transects

In five different locations throughout the Plopu valley I randomly distributed triangular transects (Appendix 3). The transects were constructed in a way that was based on the study of Torres et al., (2012) on the habitat use of red deer and roe deer. Since the study area was relatively small, the lengths of the triangular sides were set to 500m and 5m width (total surface $0{,}75E^{-2}$ km^2). Primarily the transects were done to georeference faecal samples. Also other indirect observations, *e.g.*, tracks and bark consumption, were georeferenced and used for later analysis on the bison-activity. According to the FAR method, each transect was visited twice to obtain the number of dung samples that were added during the study period. Using the following formula from The Deer Initiative, (2008), the number of bison per km^2 was calculated:

$$No. of\ bison\ per\ km^2 = \frac{no. of\ new\ dung\ samples\ per\ km^2}{no. of\ days\ between\ visits \times defecation\ rate}$$

To create more detailed information on the local habitat that the transect covers, a habitat mapping with a radius of 100 meters was done halfway on each side of the triangle. This method is based on the Common Bird Monitoring Scheme (Appendix 4; Szabó et al., 2016).

2.2.4 Other observations

During all the other walks that I did in the study area, I georeferenced signs of bison presence that were outside of the transects. Gathering this data provided more insight on the pathways that the bison used and areas where they could be found. This data was used as a layer of extra information to find 'hotspots' where a lot of bison-activity was found during the study.

2.3 Data analysis

Raw data from the GPS collar was exported from the Lotek webservice (Lotek Wireless Inc., 2016) to SPSS (IBM SPSS Statistics, version 24) for further analysis. The mapping was done in the open source Q-GIS software (GNU General Public Licence, 2.14.3-Essen). The percentage of GPS fixes was calculated to obtain information on the accuracy of the Lotek data. Coordinates were transferred from Lotek to Q-GIS and mapped accordingly. The numbers of coordinates within each habitat type were calculated in Q-GIS with the 'Points-in-Polygon' plugin. Pathways between consecutive points are created with a separate Q-GIS plugin to visualize the approximate pathways that were used to roam around and hotspots are identified. The distance covered between to consecutive data points was calculated and averaged for distance that the collared bison covered per hour. All the coordinates of the recorded dung samples, tracks and bark browse were added to the Q-GIS map and transformed into a heatmap to show the hotspots of bison-activity throughout the study area.

3 | Results

All the GPS datapoints were plotted over a map, which indicated the habitat types of the study area. The basis of this detailed land cover map was created by Angus Franz, a fellow student that joined the Bison Hillock project during July and August 2016. I modified and extended the level of detail of this map to my own needs for this study (Appendix 5).

3.1 GPS collar data

Up to the moment of writing, the GPS collar of Romanita is sending her location every two hours. The GPS collar on Birk however, unfortunately stopped sending signals after 131 days and 1552 GPS points. As Birk's GPS signal was lost before he was released into the wild, I only used the GPS data from Romanita for this study. During her stay in the bison enclosure, Romanita managed to escape and roam free for 44 days (from 17-03-'16 to 29-04-'16), before she voluntarily returned to the enclosure and was kept there after fixing the electric fence. The data points of these 44 days and the days after the official release on June 8th '16 to September 21th, making a total of 150 days, were used for the analysis. A total of 1800 points were collected from the Lotek webservice, of which 94,6% had a fix (Appendix 6). Data points without a fix, or 2D and DOP>5 were not used in further analyses. Romanita's habitat use is graphically displayed in a histogram, showing the percentage of data points within each habitat type (*e.g.,* woodland, pasture, or open forest; Figure 2).

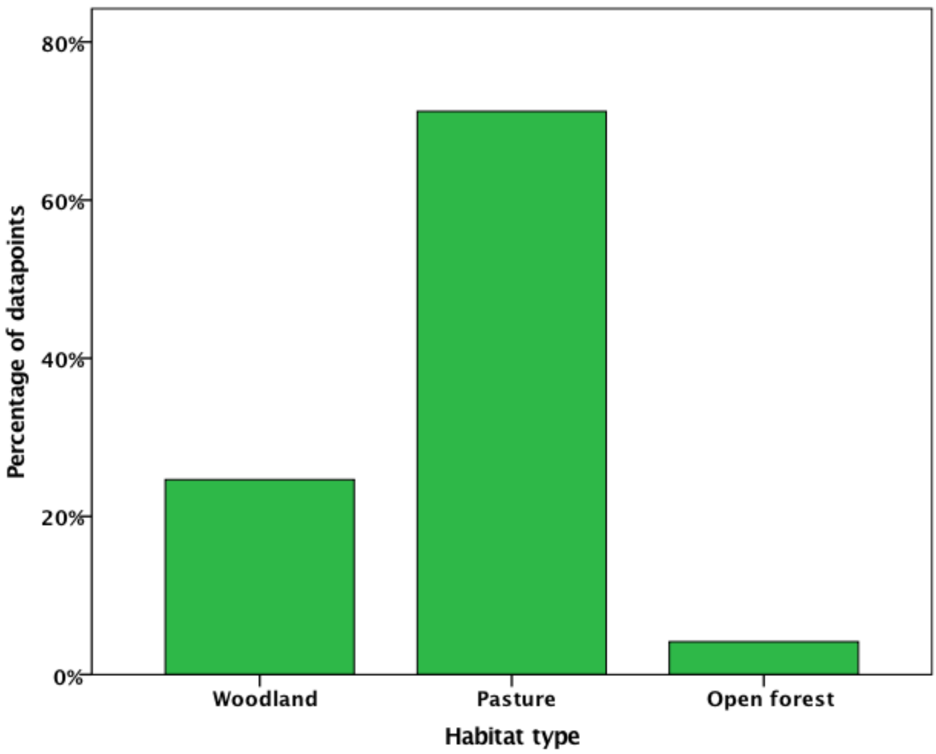

Figure 2 | Romanita's habitat use, displaying the percetage of data points in the woodland (24,6%), pasture (71,2%), or open forest (4,2%) habitat type.

All the GPS coordinates were mapped as a seperate layer in Q-GIS, and converted to a heatmap, to visualise the 'hotspots' where Romanita spent more time over the past months (Figure 3).

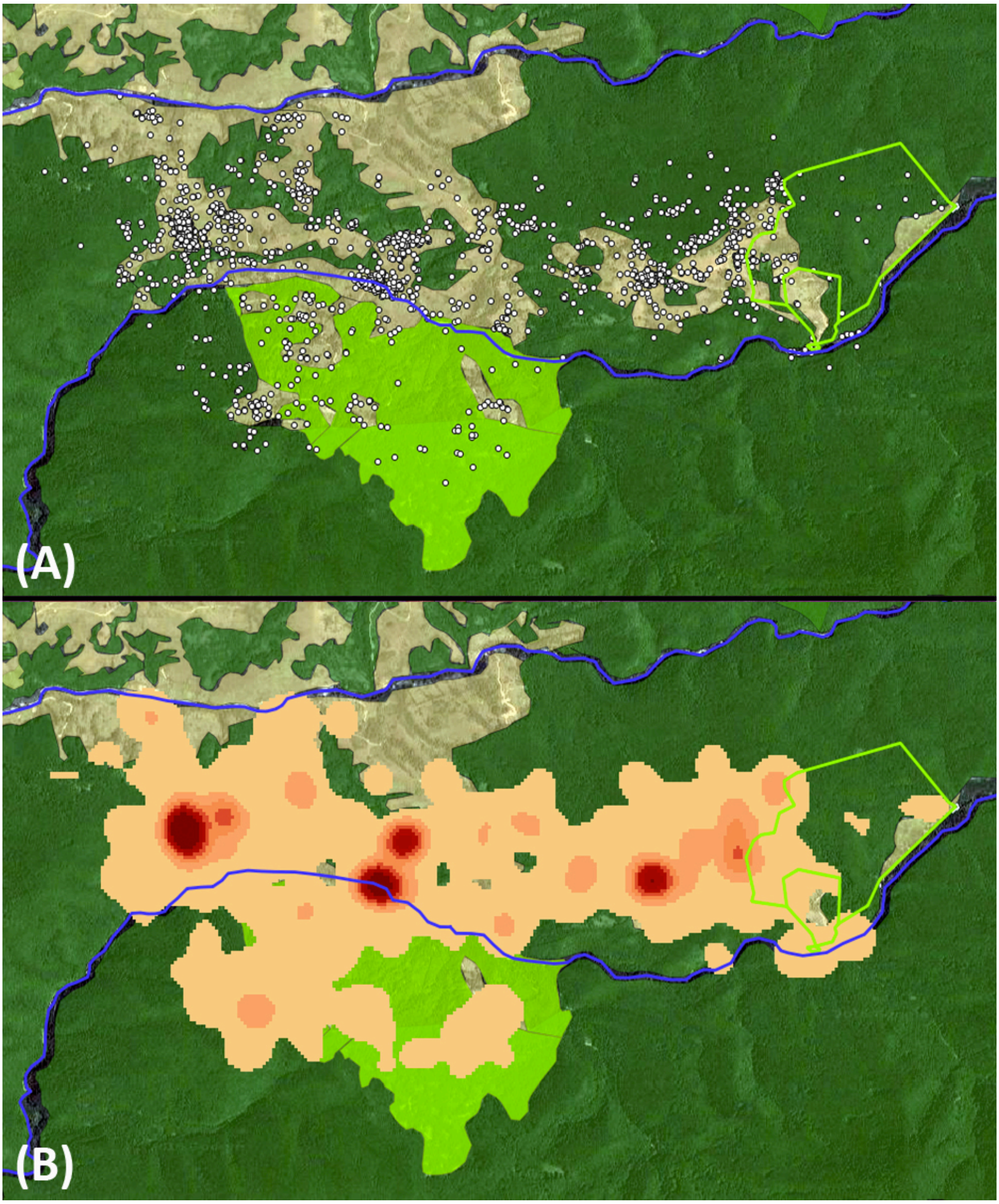

Figure 3 | (A) The coordinate points show that Romanita is found often in/around pastures. Converting these points to a heatmap (B) shows that Romanita's 'hotspots' are mainly in pasture habitats. Habitat coloration was done in Q-GIS, dark green: woodland; light green: open forest; yellow: pasture; blue line: road; green line: bison enclosure.

Romanita's movements were visualized by creating straight-line pathways in Q-GIS between to consecutive coordinates from the Lotek webservice (Figures 4). The hotspots from Figure 3(B) overlap with the clusters of lines, indicating that Romanita not only spends a lot of time there, but also visits the place often.

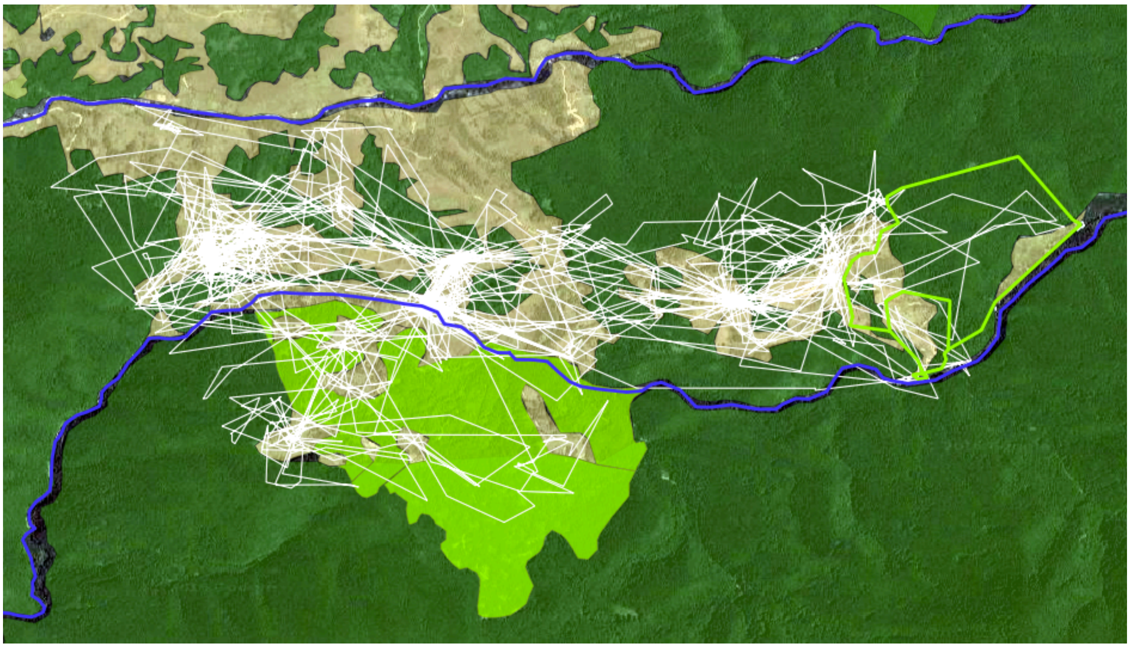

Figure 4 | Romanita's movements during her escape and after her release. Habitat coloration, see caption Figure 3.

The average travelling speed was calculated, based on the straight-line distances between consecutive GPS points (Figure 5). After transformation, the data showed a normal distribution and average speed (meters/hour), differed significantly during the day (167,8 m/h), compared to the night (66,8 m/h; t-test for eq. of means; t=13,6; df=290; $p<0.001$).

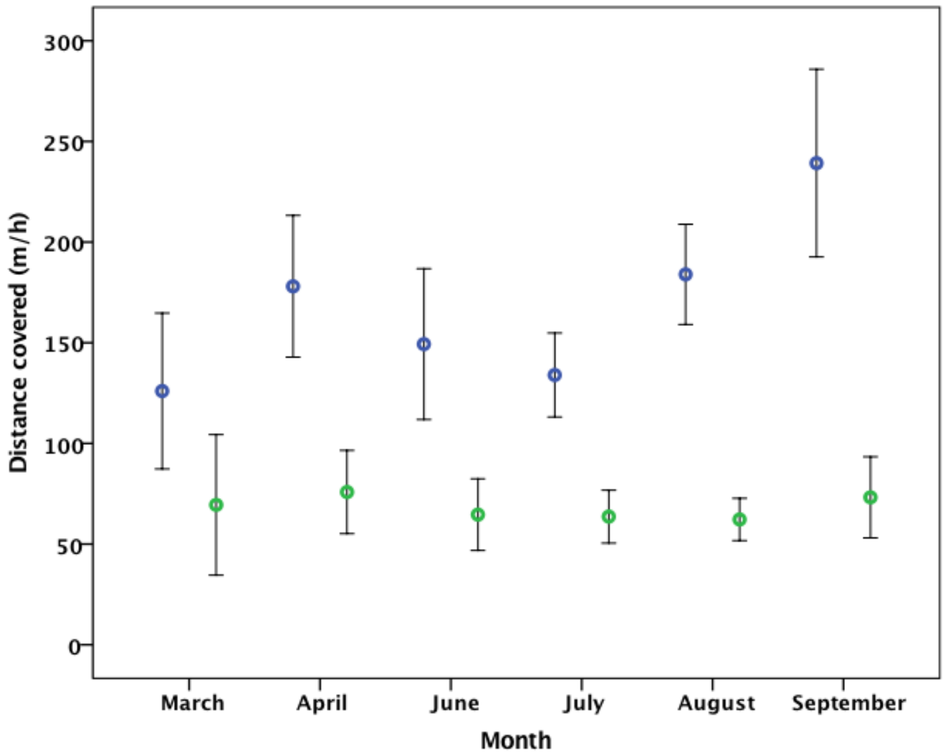

Figure 5 | The average travelling speed of Romanita (meters/hour) per month during the day (blue) and night (green) in the wild. Error bars indicate the 95% confidence interval. May is not taken into account, as it was spent in the enclosure.

A separate graph visualizes the altitude of the GPS points over time. Romanita spent all her time between approximately 1000 and 563 meters above sea level. The fitted regression line ($y=8{,}39*10^2 – 0{,}07x$) showed a relative low R^2 of 0,127 (Figure 6). There is thus no indication that Romanita is moving to lower elevations over time.

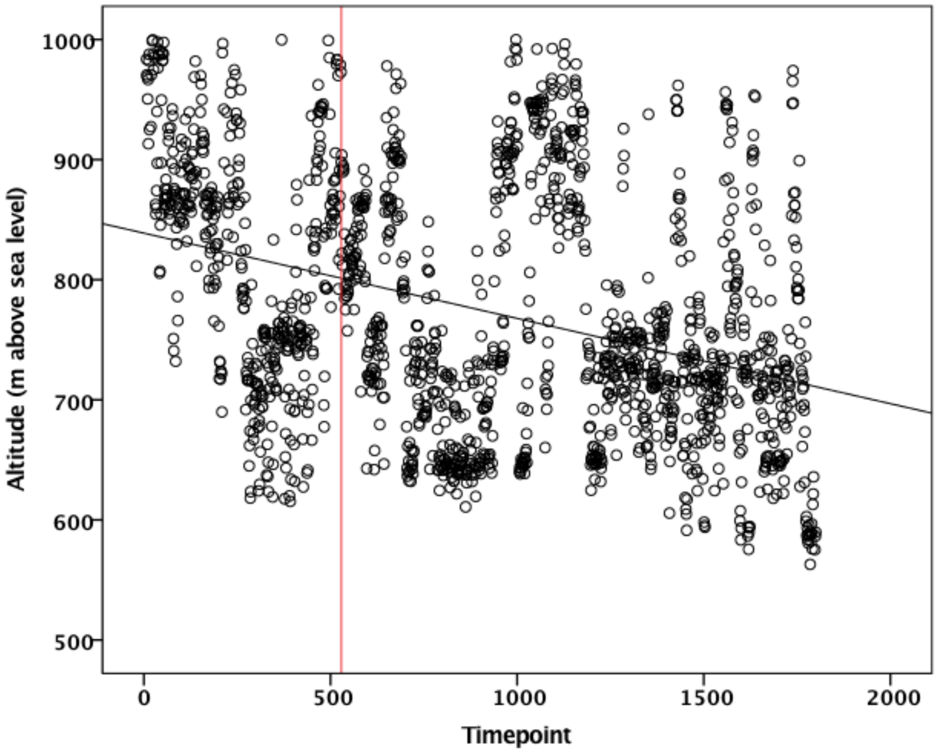

Figure 6 | The altitude of Romanita's locations during the break (prior to the red line) and after the release (after red line). Black trendline shows a relatively poor fit of R^2=0,127. Time points indicate the consecutive datapoints from Lotek webservice.

Plotting the altitudes in a histogram showed that Romanita had a few altitudes where she spent most of her time (*e.g.*, around 650m, 700-770m, and 860m; Figure 7).

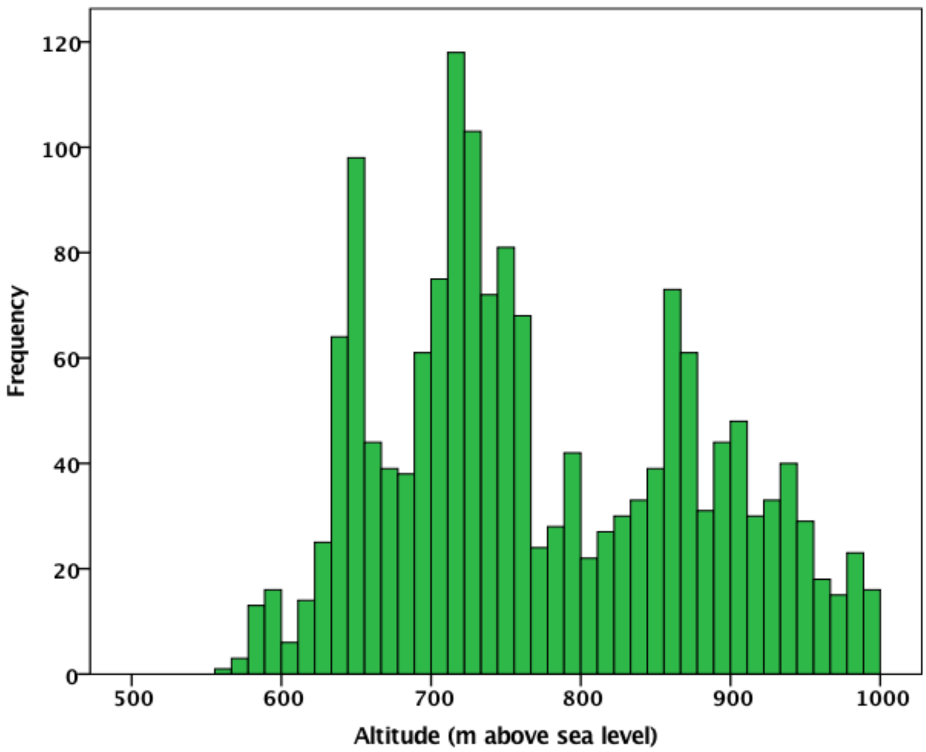

Figure 7 | The altitude frequency of Romanita's datapoints (mean=773,88; st.dev.=103,28; N=1.645). It is clear that Romanita has a few preferred altitudes where she likes to be. Other than that, she roams around between 563 and 1000 meters altitude.

3.2 Camera trapping

A total of 320 camera trap days were recorded. On average each SD card was changed after 24,6 days (min 16 days; max 37 days). The free bison (47 ind.) were recorded during 12 different moments on locations 1, 2 and 5, and remained absent on the other two cameras (Figure 8).

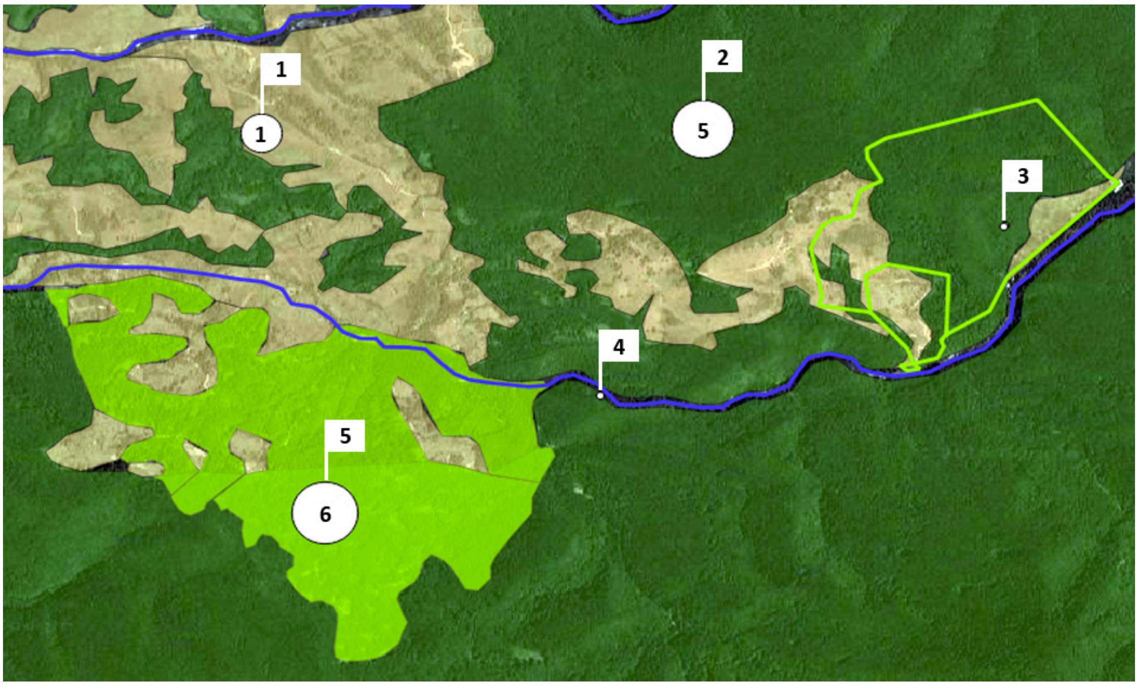

Figure 8| The number of times bison were captured on camera traps (numbered on flags) troughout the study area. Habitat coloration, see caption Figure 3.

Other wildlife species that were captured on film were Roe deer (*Capreolus capreolus*; 17 ind.), Wild boar (*Sus scrofa*; 20 ind.), Brown bear (*Ursus arctos*; 2 ind.), Fox (*Vulpes vulpes*; 2 ind.), Marten (*Martes martes*; 1 ind.), Red deer (*Cervus elaphus*; 15 ind.), and Greater mouse-eared bat (*Myotis myotis*; 1 ind.). Some wildlife species were filmed in both habitat types, while others were only filmed in one habitat type (Figure 9).

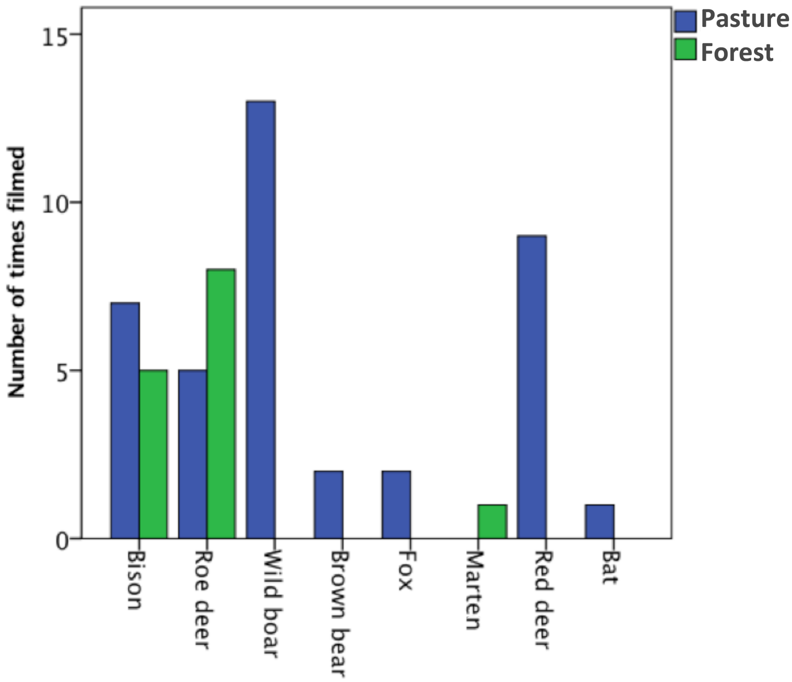

Figure 9| Occurance of wildlife species filmed with the camera traps in the specific habitat types (*e.g.*, forest or pasture). Bison and Roe deer were caught on camera in both, while the other species were only filmed in one habitat type.

3.3 Transects

The accumulation of bison dung samples that were found during the transects and the calculations according to the FAR-method show that the bison-density was highest in transect 3 (Figure 10; Appendix 6). There was no dung count in transect 1 and 2. The habiat cover indicates that transects 1 and 5 were in more open forest-fields, while transects 2,3, and 4 were in a (open) forest habitat (Figure 11).

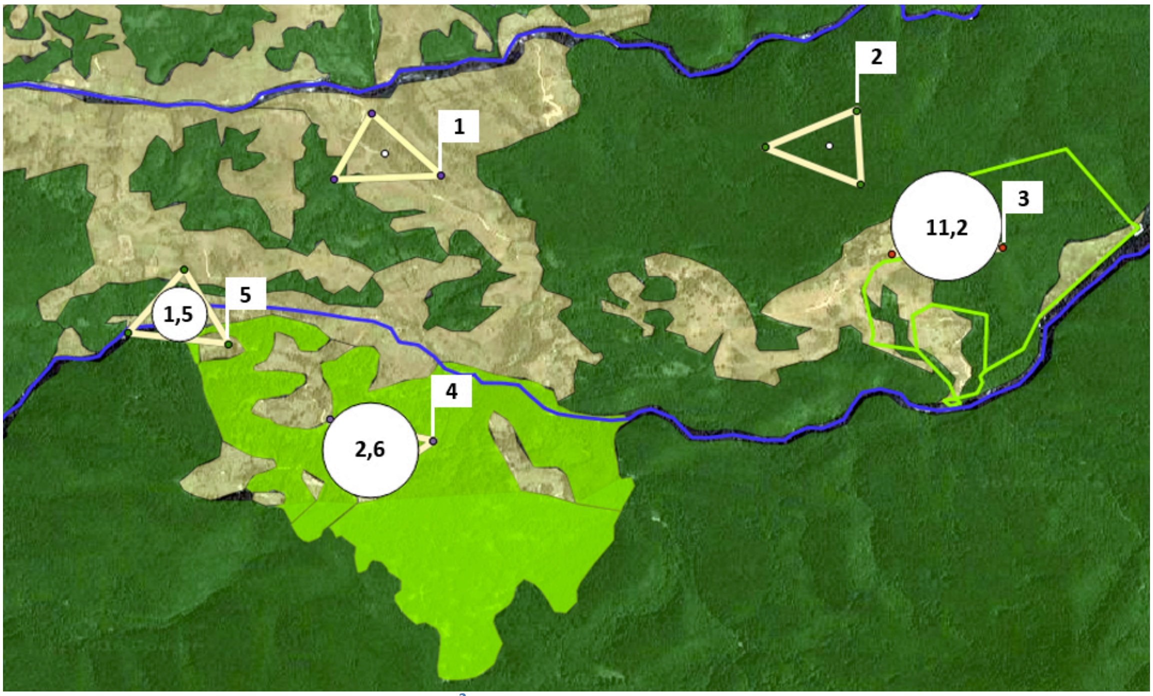

Figure 10 | Calculated bison-density (ind./km^2) with FAR-method shows that density was highest in transect 3. In transects 1 and 2 no dung samples were found. Habitat coloration, see caption Figure 3.

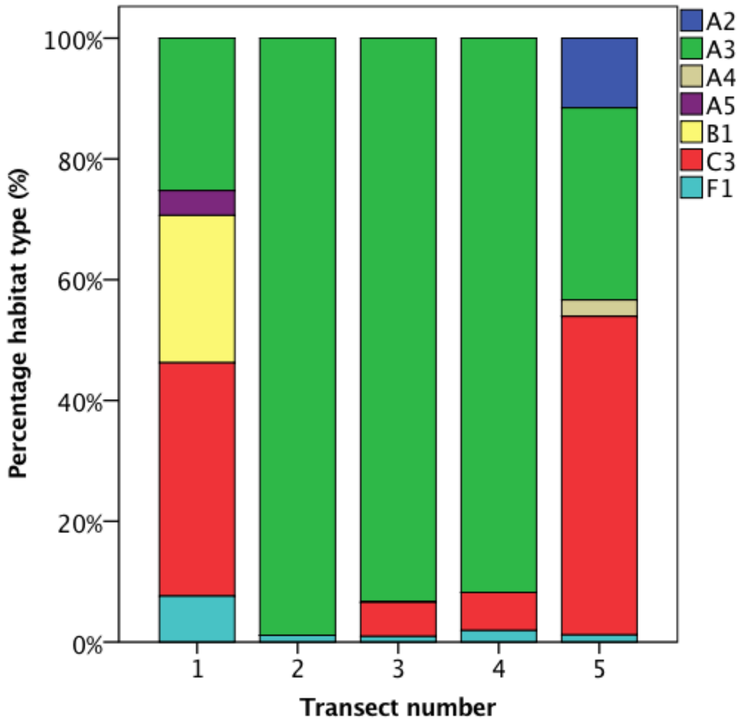

Figure 11 | Habitat covers along transects: A2= Coniferous forest; A3= Mixed forest; A4= Riparian forest; A5= Alignment of trees; B1= Forest in regeneration; C3= Pasture with isolated trees; F1= Stream (width <3m).

3.4 Other indirect observations

Signs of bison-activity that were collected during hikes through the study area, were added as a seperate layer to the Q-GIS map and converted to a heatmap to visualise the bison-activity 'hotspots' (Figure 12).

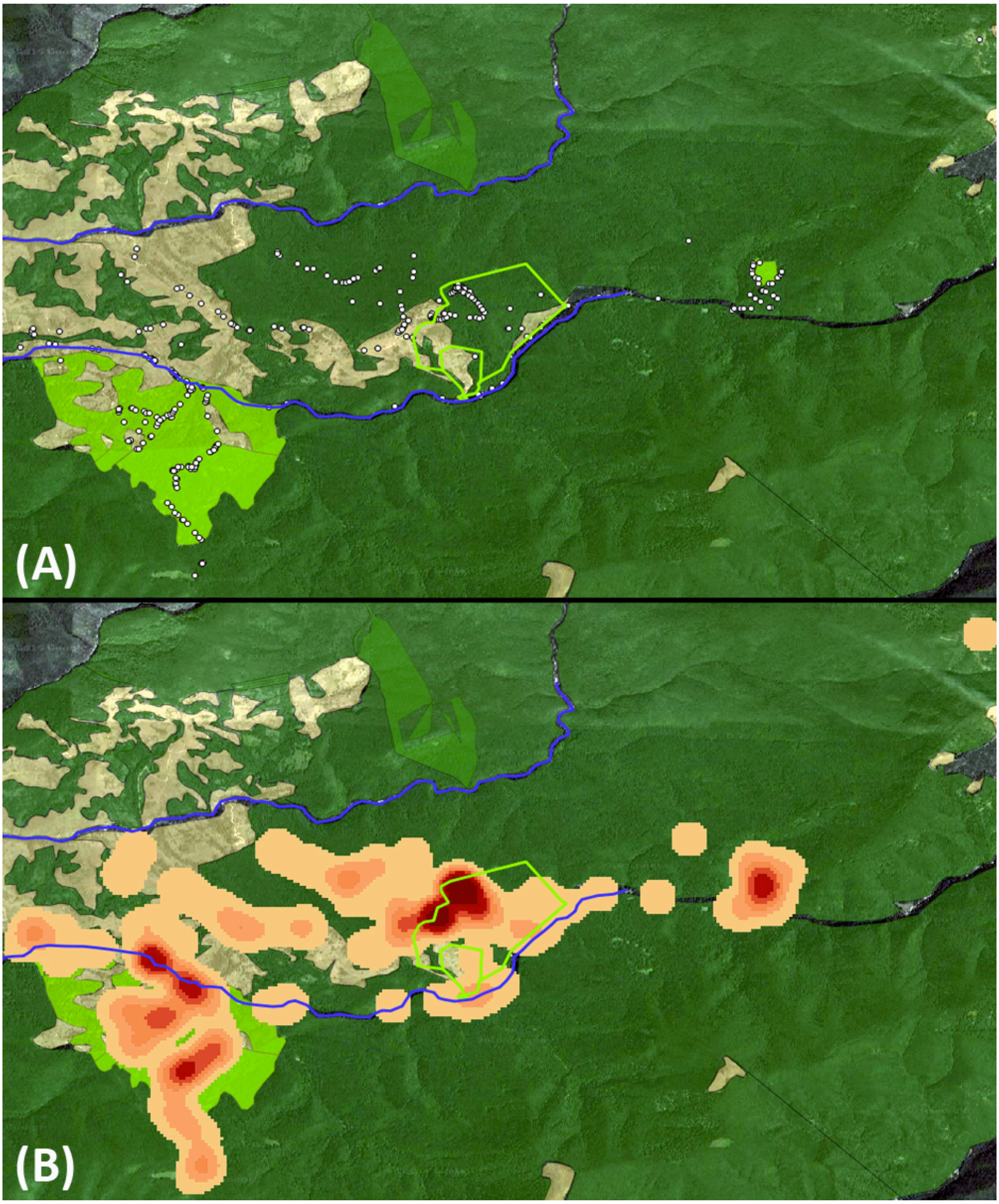

Figure 12 | A. The coordinate points of all the indirect observations (*e.g.,* dung, tracks, and bark browse) show certain locations where a lot of bison-activity was found. Converting these GPS-points to a heatmap (B.) shows the 'hotspots' where many indirect obervations can be found. Habitat coloration, see caption Figure 3.

Combining all the GPS points from Romanita with all the personal indirect observations of bison, gives an overview of all the bison-activity that was located throughout the study area. Converting this to another heatmap, displays the overall hotspots where most of the bison-acitvity took place during the past months, since the bison roamed free (Figure 13).

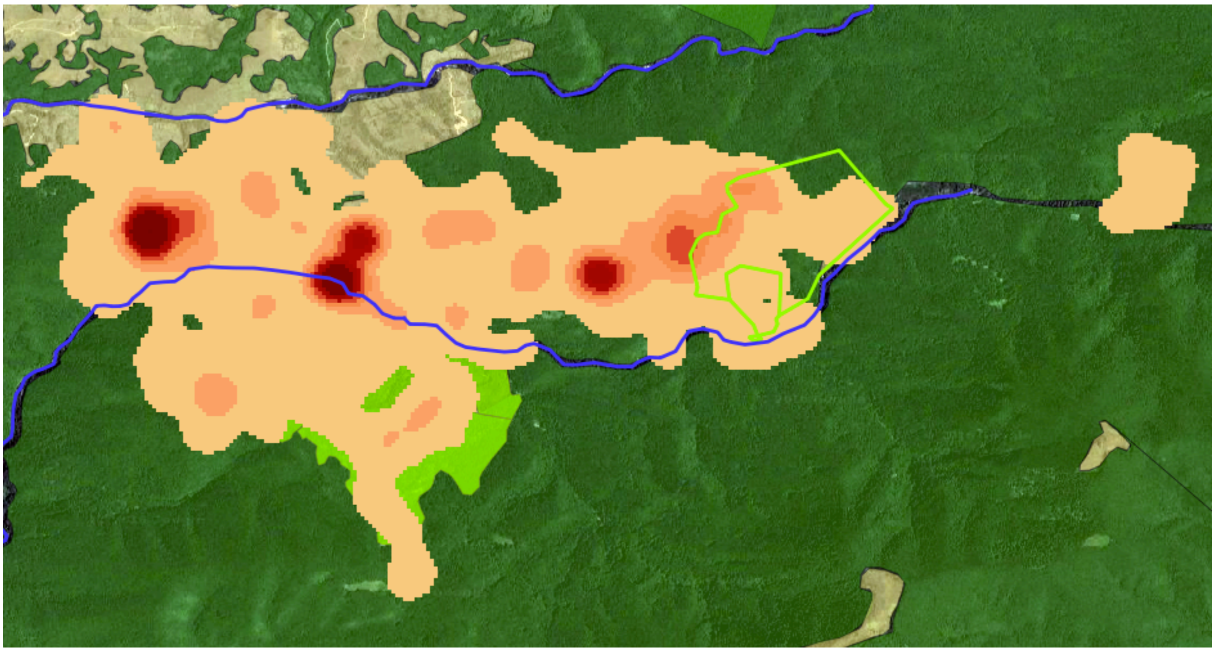

Figure 13 | Heatmap of all the GPS points that were collected during this study (*e.g.*, Romanita's locations, dung samples, tracks, and bark browse), indicating the 'hotspots' of bison-activity in the study area. Habitat coloration, see caption Figure 3.

4 | Discussion

Looking at the final result of the habitat use map, it is clear that there are a few hotspots where a lot of bison activity is found. High densities of dung samples and other indirect observations were found in several forest-field areas. From Romanita's GPS data and the habitat use histogram we can see that she likes to spend a lot of time in pastures (*i.e.*, meadows and agricultural fields). This is in contrast with previous studies that found that bison spend most of their time in the forest, like Daleszczyk, (2007). That study area in the Polish Carpathians however, is for 96% comprised of forest. Therefore it is not very remarkable that the bison spent most time in forest habitats. The fact that Romanita and her group of approximately seven bison (seen multiple times on the camera traps) are often found in close proximity of agricultural farms is not unique. A study on bison crop predation in Poland showed that between 2000 and 2010 nearly two tons of compensation was paid to local farmers for crop damages. The majority of damages were in close proximity of woodlands, and occurred during the winter season. These bison consumed cereals (61%), hay (20%) and rape (13%) the most (Hofman-Kamińska & Kowalczyk, 2012). The farms that were targeted by Romania during this vegetative season had mostly hay and several types of vegetables (pers. obs.). To prevent further damage in the future, temporarily measures are taken, like building electrical fences around the farms. Other options like triggered noise disturbance, scent barriers ('chilli fences') or guarding dogs could be tested in order to find the most effective and non-invasive solution. An explanation for this behaviour, which is not seen in all the bison, is that Romanita was a Romanian 'aged' female bison. Born in 2010, Romanita was fed hay in the Vama Buzăului breeding centre for five years, before she was released (Raczynski, 2015). Being accustomed to this, it is not unlikely that she went on a search for hay during her escape and after the release. A future recommendation would be to allow only younger bison to the rewilding project, of course while taking their genetic background into consideration, or bison that are more used to grazing and browsing.

In 2015 another student from the Wageningen University, Tiago Miguel, did a study on this project. At that stage the bison were not free however. GPS data of six weeks where Romanita was spending her time in the enclosure was analysed. Results on average travelling speed within and outside the enclosure were comparable. It seems thus that the bison enclosure is large enough to provide enough space for up to 20 bison (the maximum number during Tiago's stay; Miguel, 2015). But both results are a minimum distance that is covered. As previously mentioned, results are based on straight-line distances. It is however nearly impossible to walk in a straight line from point A to B in this wilderness area. More likely, as large herbivores normally do, the bison roam around with random movements, e.g., the Brownian motion (Venter et al., 2015). The distance that is covered by Romanita is thus underestimated according to this theory.

The altitudes between which Romanita spent here time was approximately 550-1000 meters above sea level. The histograms shows preferred altitudes around 650m, 700-770m, and 860m that are, not surprisingly, matching the altitudes of the hotspots in Figure 3B. The range of altitudes is at higher elevations than reported by Schmitz et al., (2015), but this is many because their study area was located between 450 and 700 meters above sea level. Romanita's data was similar with Wołoszyn-Gałęza et al., (2016), as their bison were found between 548 and 927 meters above sea level. Indirect observations that were done during hikes were also found in higher elevations above 1000 meters. The highest indirect observation was even at 1335 meters above sea level. These tracks were far out of the study area, up into the Țarcu Mountains. It is most likely that a few bison went exploring in this (north-eastern) direction, and will hopefully continue to do so (Appendix 2). In the coming years, I expect that the bison will not explore into even higher elevations above 1300 meters. This mainly because the forest fragmentation and transition to alpine grasslands in the Țarcu Mountains starts between 1200 and 1300 meters altitude. As the bison like to hang around the forest habitats, they will stay below of close to these altitudes.

The camera traps were used primarily to see whether the bison would be roaming in the area where the randomized camera traps were located. Interestingly camera trap 2, in the forest, caught bison walking past multiple times, while transect 2 showed no bison density in a neighbouring area of the same forest. Camera trap 3, placed in the enclosure during the time when the gates were open, showed that the bison did not return to that specific forest area inside the enclosure once they were set free, but Romanita's GPS coordinates show that she did pay a short visit after her release. Upon the release of the newly arrived group into the rewilding zone, this camera had to be used elsewhere, so a confirmation if the bison would actually go there when the gates are closed is missing. Camera trap 4 was placed on a rather unstable hill slope. Therefore, and taking into account that bison preferably avoid steep areas, it was expected that no bison were found on this camera trap (Vlasakker, 2014). The data that was collected on other wildlife species was insufficient to draw reliable conclusions, but provides data for a future wildlife monitoring study that can be done in the area.

The calculated bison density per km^2, based on the FAR and defecation rates, showed no sign of bison presence in transect 1 and 2, while the other transect outside the enclosure showed little bison presence. The results however, may not be very precise. The main cause for this is the rough terrain of the study area. As it is almost impossible to walk the exact same pathway twice trough the wilderness, even if you are following GPS directions, dung samples may have been found on the second track, while they were also present during the first track. This gives an overestimation of the accumulation of dung samples over time. Still, given the fact that is it very difficult, I tried my best to walk in straight lines to minimize the chance on this error. A good example is the result for bison density in transect 3 (e.g., 11,2 ind./km^2), as the bison density is actually known. In August, the newly arrived 10 bison were released into the rewilding zone. The actual density of bison in the 135ha rewilding zone would thus be 7,4 ind./km^2. This shows a slight overestimation that must be taken into consideration. The differences in bison density between the transects outside the bison enclosure, although approximately at the same altitude, is most likely also be influenced by the presence of shepherds. The local shepherds often have a pack of 6-8 Romanian shepherd dogs with them. From personal observation I can say that these dogs are very aggressive and are said to attack even bears and wolfs if they come to close to their sheep. In places where shepherds thus often roam (*i.e.*, near transect 1), around these pastures bison density is expected to be lower. Also wildlife predators might influence their movements, like the wolf and lynx. The wolf has been caught on camera several times in this area, and the lynx was caught for the fist time during my stay. Wounded or weakened bison may well become their prey, thus bison might avoid confrontation with these animals. Comparing the mean bison density of the transects outside the enclosure (*e.g.*, 1,0 ind./km^2) with the Polish bison herd, it seems that the densities are actually quite similar as the bison density in the Białowieza Forest is 0,7 ind./km^2, (Kowalczyk et al., 2011). The density is still twice as high as recommended by bison specialists though (e.g., 0,5 ind./km^2; Flint et al., 2002).

Although home range was not studied, I was able to make a prediction on the size of the home range according to a linear relationship between the Ln-bodyweight and Ln-home range size (Venter et al., 2015). Using average weight from Krasinska & Krasinski, (2002), the free roaming bison weighs 634 kg (males) and 424 kg (females), the home range would be between approximately 4.000-6.000 ha, or 40 to 60 km^2. Comparing these numbers with literature on home range of the free bison in the Polish Carpathians and Germany, 66.40 and 42,5 km^2 respectively, match these calculations (Daleszczyk, 2007; Schmitz et al., 2015). Looking at the area where the first 20 bison are holding up at this moment, approximately 32 km^2 is presently used by the bison as far as we know. It is therefore expected that the bison will be moving away from the bison enclosure, most likely towards the north-eastern direction, as there is less human interference higher up in the mountains. Most likely the bison will stay within close proximity of forest habitats, primarily between 500 and 1300 meter altitudes (Appendix 2).

In order to monitor the future behaviour and movements of the free-roaming bison in the Southern Carpathians, one of the goals for this project should be to increase the numbers of GPS collars on the bison. As this project works on a small budget, it is going to be a challenge, but since it is clear that the bison will disperse in smaller groups and maybe different directions the GPS collars are the most efficient method to keep track of their whereabouts. Another more labour-intensive recommendation would be to organize more exploration hikes into the higher elevated areas of Mt. Țarcu. Covering more area could help to map indirect observations of bison on higher altitudes and directions. Assisting in this could be the deployment of camera traps at predefined locations at higher altitudes. Locations should be close to forest-field habitats, preferably with an old path or trail, and in relatively close proximity of water. This data can also be used in the future wildlife monitoring study. My recommendation regarding the entire project should be a light shift in the focus. At this moment, a lot of energy and time goes into making sure that the bison are heaving a good time (feeding them and visually checking the body conditions). Focussing more on the actual 'rewilding' would benefit the project more on the longer term, but might result in losing more bison during the rewilding process.

Getting the bison to avoid human contact and settlements is a place to start with. However, the human-wildlife conflict is a problem that will always occur, as long as bison and human share the same areas. For example, even after decades of bison presence, locals in Lithuania preferred that the bison were at least 10 km away from their pastures (Balčiauskas & Kazlauskas, 2014). This will probably happen in this area too, as bison will damage human properties by consuming products or damaging properties in other ways. Another conflict is the communal interest in tree species. Logging occurs in the area, mainly focussed on beech (*Fagus sylvatica*) and fir (*Abies alba*), two tree species that bison often target for debarking. Especially during the winter, the free bison will tend towards browsing, gnawing and debarking these trees in lower elevated areas due to the snow cover. As Baraniewicz & Perzanowski, (2015) showed, bison could benefit from remains of logging sites (where debarking takes place to a certain extent) and reduce the potential conflict where bison damage and/or kill valuable trees for human logging activities.

Another thing that needs more attention is the use of the different zones in the bison enclosure. Since the first groups overstayed their initial time the young trees have been dealing with high browsing pressure over the last three years. Future conservation effort must also be directed towards giving the vegetation in the bison enclosure enough time to recover. What may be considered is the option of clearing a part of the older forest in the enclosure, to provide a patch for bushes and seedlings to grow over the years. This would create more space for seedlings to grow, which bison like to browse on. Once the bison are free, browsing pressure on the trees will be released, and bison will be browsing on other trees.

In conclusion, as Kuemmerle et al., (2011) mentioned, the reintroduction of the European bison is the most cost-effective approach to establish a viable population across the Carpathians, which have proven to be a suitable mountain range with suitable habitats to sustain a metapopulation. From the perspective of the Bison Hillock, it has a long way to go, but there is enough reason to be optimistic and progress towards a successful development of a Țarcu Mountain bison population.

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Appendix 1 | Land cover map Natura 2000 Network

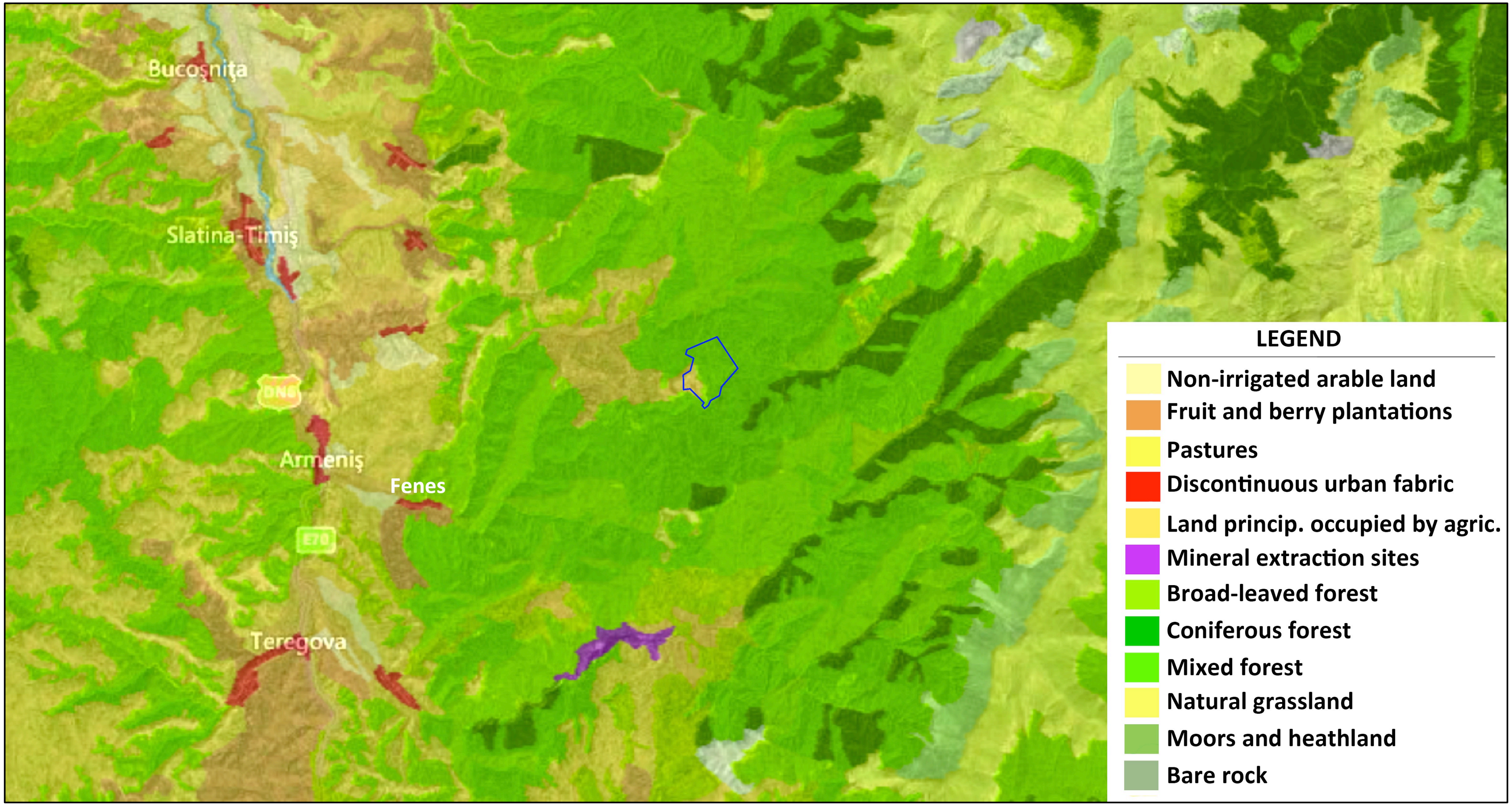


Figure 14 | General land cover map from the Natura 2000 Network, blue lining indicating the bison enclosure (based on EEA-Copenhagen, 2014).

Appendix 2 | Altitude map of study area

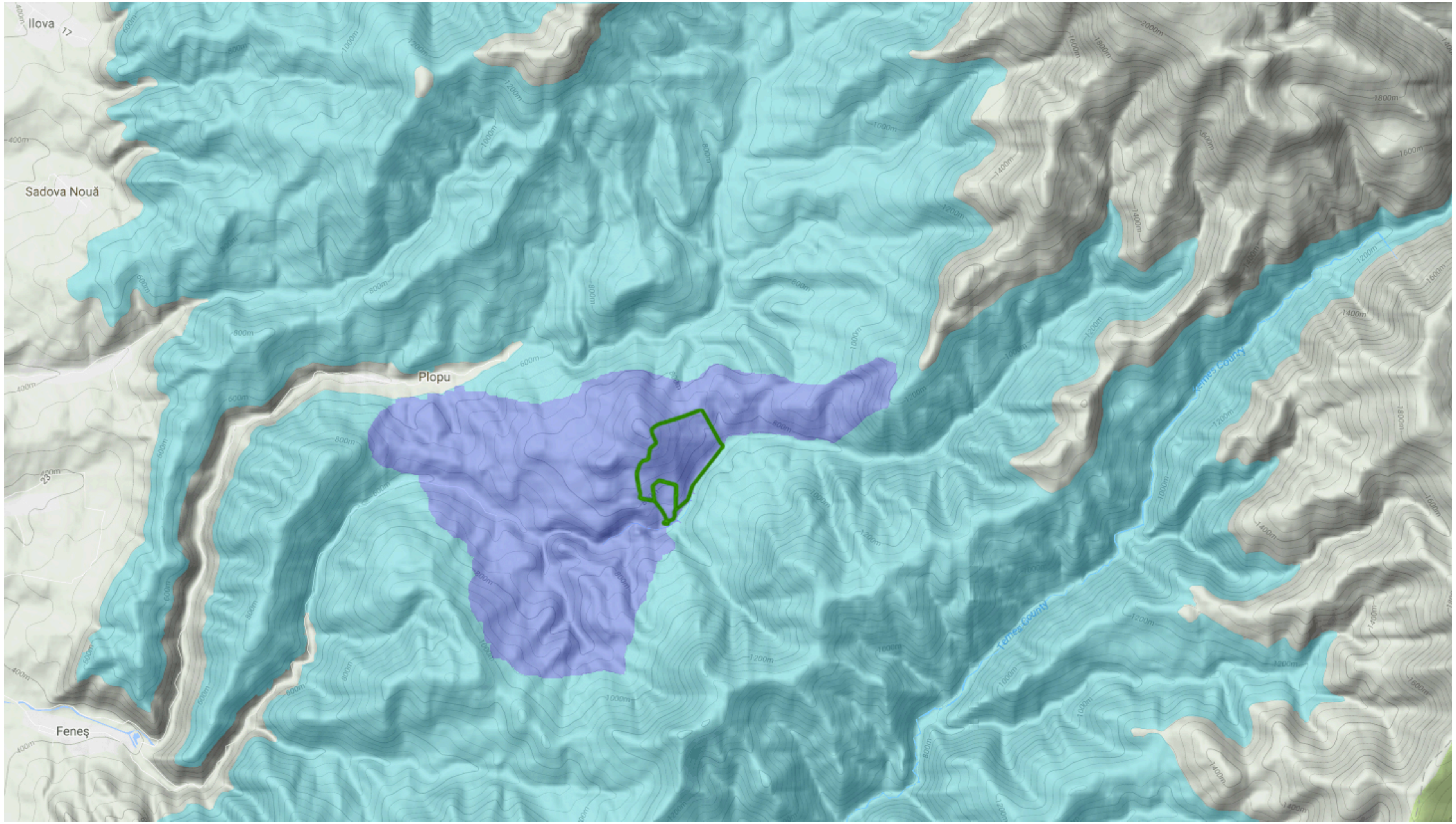


Figure 15 | Altitude map of study area and surroundings, dark blue polygon displays present bison-activity range, light blue polygon displays possible bison area between 550-1300 m altitude. Green lining indicates bion enclosure (source: Lotek Wireless Inc., 2016).

Appendix 3 | Study area setup

The positions of camera traps and transects were randomized in a way that different habitat types were covered for this study (from Google Earth; Google Inc., 2015).

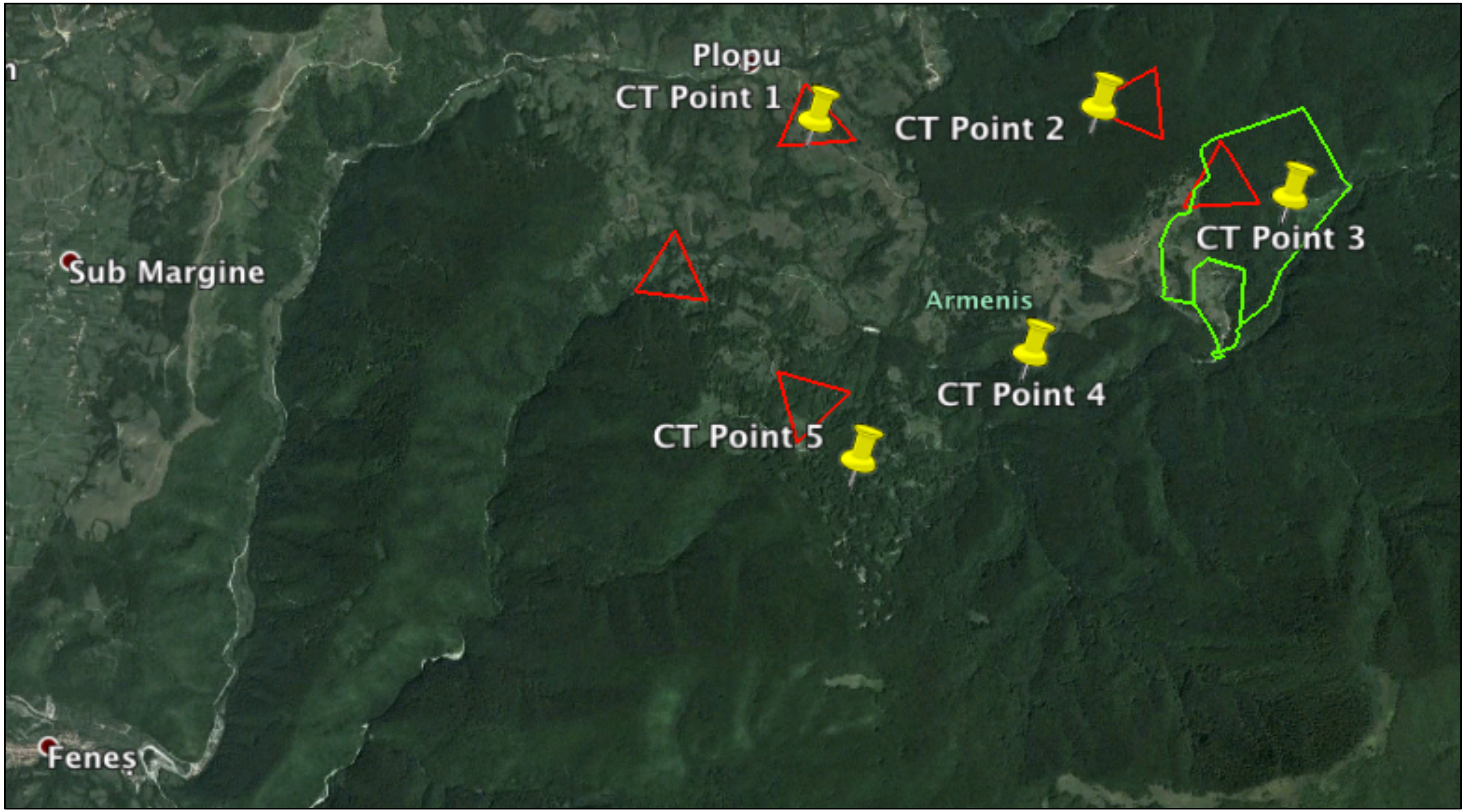


Figure 16 | Map of study area displays location of camera traps (CT) and transects (red triangles). Green lines indicate bison enclosure (from Google Earth).

Appendix 4 | Transect habitat type field form

Date: __ __ month __ __ day

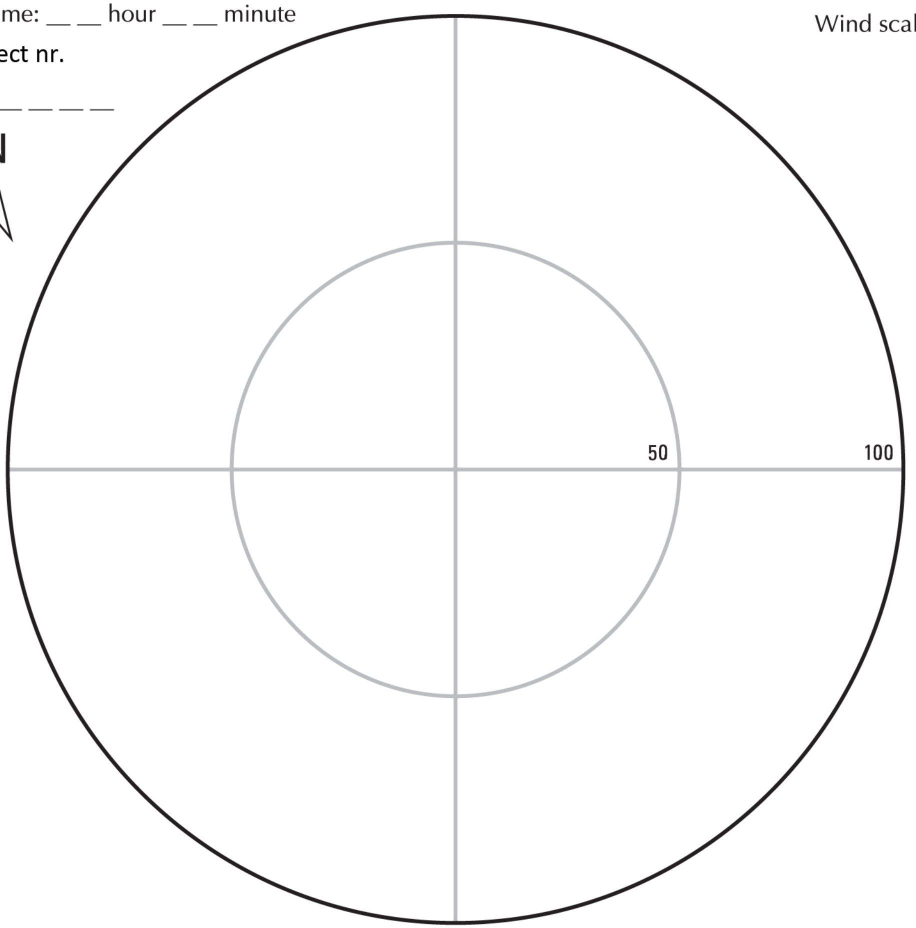
Start time: __ __ hour __ __ minute

Transect nr.

__ __ __ __ __ __

Number of observation point: ___ ___

Wind scale: ___



HABITAT TYPES

A. Forest
1. Broad-leaved
2. Conifer
3. Mixed
4. Riparian forest
5. Alignment (line) of trees

B. Bush
(height less than 5 m)
1. Forest in regeneration
2. Forest plantation
3. Riparian bush-belt

C. Meadow, pasture
1. Open pasture
2. Pasture with bushes
3. Pasture with isolated trees
4. Wet meadow
5. Hayfield

G. Areas without or scarce vegetation
1. Sand dunes
2. Rocks

D. Agricultural land
1. Annual crops (cereals, maize, rape, sunflower), intensive, >1 ha
2. Mosaic of annual crops, in small parcels (<1 ha)
3. Vegetables
4. Vineyard
5. Orchard

E. Human (anthropogenic)
1. Urban
2. Rural
3. Parks
4. Industrial areas, mining

F. Water
1. Stream (width <3 m)
2. River (width> 3 m)
3. Natural lake
4. Fishpond
5. Water reservoir
6. Sea
7. Reed-bed

The habitat types should be easy to assign. First we have to identify the main habitat type (A, B ... G) then if possible the sub-category (1, 2 ...). The size and shape of the habitat should be noted in the 100 meter radius circle, together with the habitat code (e.g. F8, D1) — see the example on the inside cover.

BEAUFORT-SCALE FOR WIND SPEED ESTIMATION

0 – calm (smoke rises vertically, 0.0 to 0.5 m / sec.)
1 – light air (smoke drifts and leaves rustle, 0,6 - 1,7 m / sec.)
2 – light breeze (wind felt on face, 1.8 to 3.3 m / sec.)
3 – gentle breeze (leaves move, flags extended, 3.4 to 5.2 m / sec.)
4 – moderate breeze (dust and small branches move, 5.3 to 7.4 m / sec.)
5 – fresh breeze (large branches move, wires whistle 7.5 to 9.8 m / sec.)

The Beaufort-scale has values up to 12, but we do not mention them, because above the value 3 bird counting is not recommended, especially in forests, where they are identified primarily based on the song.

Figure 17 | Halfway each triangular side of the transect, this form was filled in to map the habitat types and cover. The form is based on the fieldwork form from the Common Bird Monitoring Scheme.

Appendix 5 | Habitat types in the study area

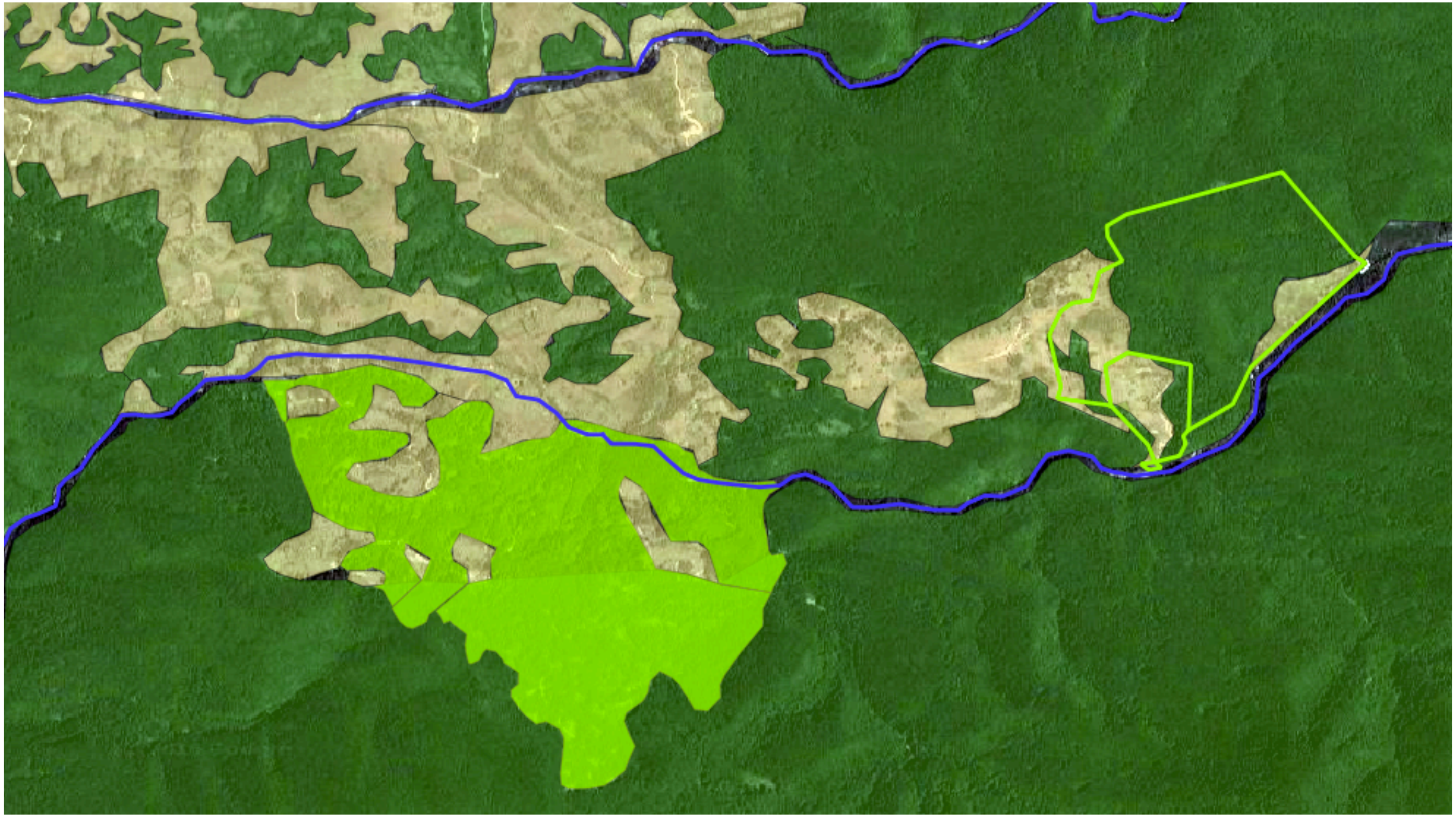


Figure 18 | The main habitat types that are found in the study area are woodland (dark green), open forest (light green), and pasture (yellow),. Dark blue lines indicate roads (gravel), green lining the bison enclosure. Mapping was done in Q-GIS by Angus Franz and myself.

Appendix 6 | Calculations

A total of 1800 data points were retrieved from the Lotek webservice page, during the first 150 days that Romanita was outside the bison enclosure. The accuracy of the data set is visualized (Figure 19), and other calculations are shown (Table 1 & 2).

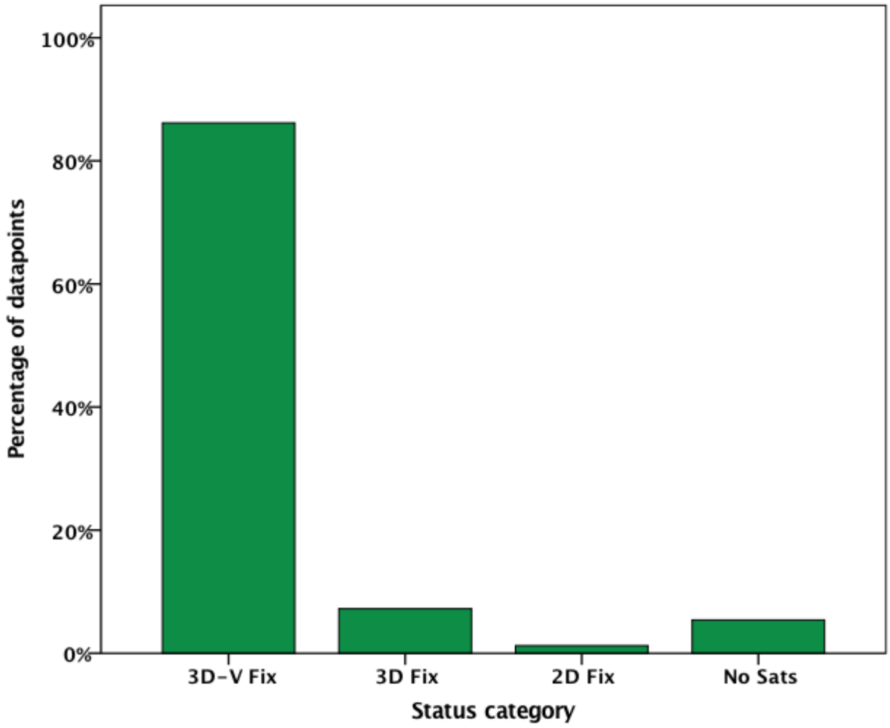


Figure 19 | Categorized datapoints show that most datapoints (94,6%) were fixed. The majority was 3D(-V) fix, indicating signal from 4 satellites or more.

Table 1 | Detailed information on the average travelling speed of Romantia (m/h) at the t-test.

Time	Mean	N	Std. Deviation
Day	167,8252	146	85,16117
Night	66,7753	146	42,22213
Total	117,3002	292	84,04515
T-test for equality of means			
t	df	Sig. (2-tailed)	
13,557	290	0,000	

Table 2 | Calculations of the bison density (number of bison/km^2), based on the dung-counts from the transects by using the FAR-method.

Transect	Round 1	Round 2	Date round 1	Date round 2	Day interval	Nr bison/km2
1	0	0	29 jun '16	09 sep '16	72	0,0
2	1	0	13 aug '16	12 sep '16	30	0,0
3	25	39	19 jul '16	15 sep '16	58	11,2
4	1	9	07 jul '16	02 sep '16	57	2,6
5	1	5	13 jul '16	07 sep '16	56	1,5