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Movement and activity ranges of the European green toad (Bufotes viridis) in an urban area compared to a natural habitat

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Abstract

The expansion of towns and cities goes hand in hand with degradation of natural habitats, which results in decline of many species. In particular amphibians suffer badly from human induced environmental changes, partly due to their complex life cycle. There are a few species that manage to colonize secondary habitats in the heavily modified areas, such as the European green toad (*Bufotes viridis*). This makes it an interesting species for conducting research on effects of anthropogenic changes on wildlife. Our study attempts to answer the question how movement activity of *B. viridis* differs in a strongly anthropogenic environment compared to the natural habitat.

Using radiotelemetry, we showed that activity ranges in an urban park (Rudolf-Bednar-Park in Vienna - RBP) were significantly smaller than those in a natural habitat (soda pan in the national park Neusiedlersee/Seewinkel). We discovered previously unknown daytime hiding places and site fidelity was observed in both habitats. Also, it was witnessed that tracking packages mounted on the male's back did not prevent them from successfully mating.

The results of this study are an indication of potential adaption of spatial activity to different habitats (urban versus natural) in *B. viridis*, however further research is needed. The dataset generated in this study can be used as preliminary data to apply this approach to several sites along a human disturbance gradient for populations in Europe, which can contribute to collecting information for conservation measures.

Keywords: movement tracking, home range, urban habitat, natural habitat, *Bufotes viridis*, Bufonidae

Zusammenfassung

Die Ausdehnung der Städte ist eng mit der Zerstörung natürlicher Lebensräume verbunden, was zum Rückgang vieler Arten führt. Insbesondere Amphibien leiden sehr unter den vom Menschen verursachten Umweltveränderungen, unter anderem aufgrund ihres komplexen Lebenszyklus. Aber wenigen Arten gelingt es, sekundäre Lebensräume in den stark veränderten Gebieten zu besiedeln, wie z. B. der Wechselkröte (*Bufotes viridis*). Dies macht sie zu einer interessanten Art für die Erforschung von Auswirkungen anthropogener Veränderungen auf Wildtiere. Diese Studie versucht die Frage zu beantworten, wie sich die Bewegungsaktivität der Wechselkröte in einer stark anthropogen geprägten Umgebung im Vergleich zum natürlichen Lebensraum unterscheidet.
Unter Verwendung von Radiotelemetrie konnten wir zeigen, dass die Aktionsräume in einem städtischen Park (Rudolf-Bednar-Park in Wien - RBP) signifikant kleiner waren als die in einem natürlichen Habitat (Sodalacken im Nationalpark Neusiedlersee/Seewinkel). Weiters wurden bisher unbekannte Tagesverstecke entdeckt, sowie Standorttreue in beiden Lebensräumen festgestellt. Außerdem wurde nachgewiesen, dass die auf dem Rücken der Männchen angebrachten Tracking-Pakete sie nicht an der erfolgreichen Paarung hinderten.
Die Ergebnisse dieser Studie deuten auf eine mögliche Anpassung der räumlichen Aktivität von *B. viridis* an unterschiedliche Lebensräume (urban vs. natürlich) hin, jedoch sind weitere Untersuchungen erforderlich. Der bei dieser Studie entstandene Datensatz kann als Referenz für die Anwendung dieser Methode auf mehrere Standorte entlang eines Gradienten menschlicher Störungen für Populationen in Europa verwendet werden, was einen Beitrag beim Sammeln von Informationen für Erhaltungsmaßnahmen leisten kann.

Schlagwörter: Bewegungs-Tracking, Home Range, urbanes Habitat, natürliches Habitat, *Bufotes viridis*, Bufonidae

1. Introduction

Cities and agricultural areas are constantly growing at high speed and are occupying more and more space, leaving increasingly less room for wildlife (Blab, 1986; Hamer & McDonnell, 2008; Luedtke et al., 2023). What remains for free-living animals are usually only highly fragmented patches (Betts et al., 2019; Luedtke et al., 2023), which makes surviving and especially reproduction extremely difficult if not impossible (Keyghobadi, 2007; Wind, 1999). Amphibians are particularly affected by habitat loss and fragmentation, ~66 % of anurans and ~36% of caudates need to migrate to water pools for reproduction because of their aquatic larval development, which requires suitable and connected habitats for juvenile and adult animals, but this is often no longer fulfilled (Kovar et al., 2009; Lee et al., 2022; Liedtke et al., 2022). As an extensive global amphibian assessment confirms, the status of amphibian populations worldwide has deteriorated (92.9% of all amphibian species are IUCN red-listed) and they are the top threatened vertebrate class (40.7% of all amphibians are threatened) (Luedtke et al., 2023).

Some amphibians can survive in heavily altered environments like big cities, for example the common newt (*Lissotriton vulgaris,* formerly *Triturus vulgaris*) (Vershinin, 1996), the green toad (*B. viridis*) (Ensabella et al., 2003; Kaczmarski et al., 2019; Kühnel & Krone, 2003; Landler et al., 2023), and the common toad (*Bufo bufo*) (Mazgajska & Mazgajski, 2020). However, all environmental conditions must be suitable and often monitoring and compensatory measures are necessary to possibly maintain a stable population (Lee et al., 2022).

Species can be divided into three categories, according to how they deal with increasing urbanization: "Urban avoiders" are species that react most sensitive to urbanization and usually leave when the human population density becomes too high. "Urban adapters" can survive in cities and can even do well in certain areas of the city that are particularly suitable for them. "Urban exploiters" such as rock doves (*Columba livia domestica*), house sparrows (*Passer domesticus*), Norway rats (*Rattus norvegicus*), and house mice (*Mus musculus*) thrive in cities. They usually benefit from the increased food supply from humans, lower predation pressure and buildings used as breeding or resting structures, allowing their populations to grow to large densities (McKinney, 2006; Rogers et al., 2021). This categorization is mostly used for birds and mammals (McKinney, 2006) as they include the most common exploiter species, but it can also be applied for amphibians.

The effect of urban areas on amphibians was studied, for instance by Clark et al. (2008). They estimated egg masses of spotted salamanders (*Ambystoma maculatum*) and wood frogs (*Rana sylvatica*) to set up models that should predict the presence and abundance of the two species in the selected urban area. Among the most important variables in their set-up models were open areas, road length, and human population density, the latter had effects over very large spatial scales. As expected, there were differences between the two species. A species that again differs in its habitat requirements from the two species above is *B. viridis*, which avoids forests and dense structures (Landler et al., 2023). The green toads' presence correlates positively with dynamic areas such as construction sites, which is probably related to its life-history as a pioneer species and highlights the importance of preserving and creating open space in cities (Landler et al., 2023).

Radiotelemetry is a technology that can deliver movement information of free-roaming animals, as it remotely transmits positional information of the animal to a receiving device (Cooke et al., 2004; Hounslow et al., 2019). This method allows to gain knowledge about the animals' habitat use, home-range size, as well as mortality and migratory behavior (Martin et al., 2009). The behavior, amount of movement and spatial distribution in the city differs in many species from those in natural habitats (Mader, 1984; Prange et al., 2004; Sol et al., 2013 - behavioral aspect). Lehrer & Schooley (2010) studied home ranges and space use of adult woodchucks (*Marmota monax*) in urban areas with varying degrees of urbanization. The size of home ranges in the studied urban woodchucks was 90% smaller than the one of rural individuals. The home ranges of carnivores such as raccoons (*Procyon lotor*; Prange et al. 2004), skunks (*Mephitis mephitis*; Rosatte et al. 1991) and bobcats (*Lynx rufus*; Riley 2006) also became smaller with increasing degree of urbanization, often linked to food availability (Prange et al., 2004). There are not many studies dealing with the ecology of amphibians in urban environments and those that do exist often study communities (Scheffers & Paszkowski, 2012). But there are a few that focused on individual species: Groff et al. (2017) radio tracked wood frogs extensively at various sites in Maine (USA) to learn more about movement extensities in all habitats used by them from hibernation to breeding and foraging grounds. They found that movement patterns are different depending on the annual phase and the associated habitats. Telemetry can also be used to investigate the response of amphibians to habitat changes as it was done with wood frogs in a conservation area in Missouri by Rittenhouse & Semlitsch (2009). The habitat change was the clearing of an oak-hickory forest that had previously surrounded the spawning grounds. The natterjack toad (*Epidalea calamita*) is an amphibian species whose movement ecology has been quite well studied (Frei et al., 2016; Husté et al., 2006; Miaud et al., 2000; Sinsch et al., 2012). Husté et al. (2006) radio-tracked natterjack toads in an urban park in Paris with the goal of exploring the effects of landscape fragmentation. They additionally performed a translocation experiment with tagged toads.

The movement behavior of natterjack toads was also investigated using radioelemetry in a semiarid agriculturally used area in the North-East of Spain (Miaud et al., 2000). A study researching natterjack toads by Frei et al. (2016) combined radiotelemetry with population genetics to find out if the population in an agricultural area in Switzerland is genetically isolated and if so, whether improvements could be achieved with management measures. Radiotelemetry is well suited to complement genetic analyses, as these do not provide such high-resolution spatial details of movement and frequently used areas.

Combining radiotelemetry devices with accelerometry enables the assignment of movement patterns to behaviors in their habitat, with telemetry being used to detect and recapture the animals (Spießberger et al., 2023; Halsey & White, 2010). Halsey & White (2010) went a step further, they were the first that used accelerometers mounted on cane toads (*Rhinella marina*) to derive the approximate energy expenditure of distinct behaviors in free ranging animals. The energy expenditure was also approximated by Qasem et al. (2012) equipping various animal species (e.g. coypu (*Myocastor coypus*), larger hairy armadillo (*Chaetophractus villosus*), greylag goose (*Anser anser*) and magellanic penguin (*Spheniscus magellanicus*)) and humans with accelerometers while letting them run on a treadmill.

Tracking studies focusing on green toads have been performed sporadically (e.g. Indermaur et al., 2009; Ott, 2015; Spießberger et al., 2023). Ott (2015) equipped green toads (*Bufotes variabilis*) with radiotelemetry devices on the Baltic Island Fehmarn and Indermaur et al. (2009) mounted tracking devices on green toads (*B. viridis*) and spiny common toads (*Bufo bufo spinosus*) in Italy at the Tagliamento river. Both focused on the characterization of habitat use in the summer grounds and Ott (2015) additionally on emigration events from the spawning waters. In another tracking study, implanted temperature-sensitive transmitters were used to study behavioral strategies to regulate body temperature in two species (*B. virdis*, *E. calamita*) (Sinsch & Leskovar, 2011).

More studies are needed that focus on individual species and how they disperse, reproduce, move and survive in the city, as this information is essential for planning conservation measures (Scheffers & Paszkowski, 2012).

For addressing our research question, we use *B. viridis* as model species, because it still occurs in very natural habitats, e.g. dynamic flood plains at the Tagliamento River (Indermaur et al., 2008, 2009), the Vjosa in Albania (Frank et al., 2018) and at soda pans in the national park Neusiedlersee/Seewinkel (Amon, 2022), but also in various cities (e.g. Rome, Italy - Ensabella et al., 2003; Oradea, Romania - Kovács & Sas, 2010; Vienna, Austria - Landler et al., 2023). Their life-history as a pioneer species enables them to quickly colonize new habitats, and the green toad does not have special requirements for breeding pools (Glandt, 2015); both characteristics are advantageous for surviving in an anthropogenic habitat.

The aim of this study is to supplement knowledge about the activity and movement ranges of *B. viridis* in a human dominated and in a still very natural habitat. For this goal selected green toads were equipped with tracking packages.

Our hypothesis is that *B. viridis* shows different spatial behavior in urban habitats, in a way that activity ranges are smaller compared to more natural environments, because space in the city is more limited. Moving around also involves more risks in a crowded park with many dogs and people and when migrating over longer distances the danger of roadkill is omnipresent. We further hypothesize that the activity ranges are larger in the later spawning period compared to the beginning of the mating season, in both habitats. A conceivable explanation for this would be that increased activity is associated with warmer air temperatures, typically occurring in the later spawning period, which can also result in additional daytime activity of the toads (Ott, 2015).

As we have only analyzed the movement of individuals over short periods of time (several days) and the term home range is typically used for describing the area of movement throughout the year (Indermaur et al., 2009; Ofstad et al., 2016), we will refer to the areas which the green toads used daily for foraging, resting and reproduction as activity ranges.

2. Material and Methods

2.1 Study species

Bufotes viridis is listed in Annex IV of the European Habitat and Species Directive (Council Directive 92/43/EEC, 2013; Umweltbundesamt, 2019) and is strictly protected in the Vienna city area as defined by the Vienna Nature Conservation Act and the Vienna Nature Conservation Ordinance (Rienesl, 2021). It is a medium-sized toad, which can be identified by its large green spots on a lighter background; the pattern is individually different. The mating call of males is a melodic trilling similar to the chirping of the mole cricket (*Gryllotalpa gryllotalpa*) (Glandt, 2015; Nöllert & Nöllert, 1992). Besides the acoustic signals, a clear characteristic to determine the sex are the nuptial pads of the males, which are located on the inside of the first three fingers and can be very dark and rough during the mating season (Nöllert & Nöllert, 1992). Depending on the region, they start to migrate to the spawning waters in March to April and the breeding period lasts until July or even August. Green toads are prolonged breeders and have an extended spawning period. The female toads stay only briefly at the spawning ponds to deliver their spawn, while the males migrate to the spawning waters many nights and call there for several hours (Glandt, 2015). According to Gordon (1962) green toads can tolerate very saline environmental conditions as high as 19 permille at temperatures near 25° C. Successful development of green toad tadpoles in waters with 10 permille salinity is described by Glandt (2015). From October to March, green toads usually hide in burrows or bury themselves in the ground to be protected from frost (Stöck et al., 2008).

2.2 Study sites

2.2.1 Natural study site

The data for the natural habitat were collected at the Kirchsee in the national park Neusiedlersee/Seewinkel. The Kirchsee is a soda pan, but due to expansion of the drainage network for regulating the groundwater level the salt concentration has declined sharply and the ion composition has changed. Many soda pans are already destroyed and also the Kirchsee is called a "mutilated remnant" by Krachler et al. (2012). In most years it falls dry in spring, but data collection took place in an unusually wet spring (2023) with frequent rainfall, preventing it from falling dry until the end of data collection (end of May). The water level was around 25 cm in the deepest parts during data collection. The vegetation cover was sparse, which is an ideal condition for the green toad (Stöck et al., 2008). The area of the soda pans is located inside the national park, therefore the access for people is strongly limited.

2.2.2 Urban study site

The urban study site is a park named Rudolf-Bednar-Park (RBP), situated in the second district of Vienna. It spans approximately 31 000 m² and is surrounded by a residential area. Our study area included 13 rectangular water basins, flower beds, two dog areas, a playground, short cut lawns, a few hedges and private gardens. The water basins were created for decorative purposes but are used by the green toads as spawning water (Landler, 2021). They are ca. 2 m wide, 8 to 12 m long and around 10 cm deep. All but one include patches planted with reeds and a graveled ground. At the edge of each basin, there is at least one concrete slab, placed to serve as exit assistance for juvenile and adult green toads. In case the water level of the basins drops sharply, they are refilled.

2.3 Telemetry devices

The transmitter package consists of a VHF transmitter with a titanium antenna (200µW; Plecotus Solutions GmbH, Freiburg, Germany), an accelerometer, a magnetometer, a temperature sensor (Axy5; Technosmart, Europe srl, Rome, Italy), and a rechargeable 20 mAh lithium-ion battery. The device was customized by Plecotus Solutions to be extra small (18 mm length, 12 mm width, 10 mm height), lightweight (between 2.5 and 2.9 g) and waterproof. The antennas have a length of between 12.5 and 15.5 cm protruding at the back of the toad when attached (Fig.1).

The sampling frequency was 10 Hz for the accelerometer and 2 Hz for the magnetometer. The resolution has been set to 10 bits ranging from -8g to +8g. With these settings the device's battery has a lifetime of a maximum of 12 days, until recharging is necessary. The signal emitted by the VHF-transmitters was received with a handheld Yagi antenna (Lotek, 148-152 MHz) connected to a RX98 receiver (TVP Positioning AB, Lindesberg, Sweden). Accelerometer and magnetometer data were stored on the device and downloaded via USB connection after each recording period.

The recorded accelerometer data will be analyzed in another study and magnetometer data was recorded but will not be evaluated for this project.

Fig. 1: **Toad with tracking package**. Each device is marked with a symbol to enable identification (here: heart). The tracking package is attached with silicone tubing around the waist. The antenna is protruding beyond the toad's body.

2.4. Data collection

2.4.1 Equipping procedure

Toads were captured, the snout-vent-length (SVL) was measured with a vernier caliper (precision: 1 mm) and the dorsal pattern of the toads was photographed. The body mass was determined before equipping the toads with tracking packages and after removing them using a digital fine balance (USA Weigh Atlanta; precision: 0.1 g). We chose individuals with a body mass of at least 28 g so that the mass of the transmitters was not more than 10% of the body mass of the toads. This ratio has been described in other studies as being well applicable for amphibians (e.g. Altobelli et al., 2022; Indermaur et al., 2008). Sex was determined mainly by checking the presence of nuptial pads, which is a clear indication for males. Exclusively males were used, because females are underrepresented at the breeding waters, and we wanted to minimize disturbance of the population by this study. For each field survey date, time, air temperature and the exact position (GPS) of each toad were noted. The tracking packages were always attached by two people, one was holding the toad, while the second person attached the transmitter around the toad's waist with silicon tubings by binding a double knot (Fig.1). Then the toad was placed on the ground and observed for a few minutes to check if normal movement was possible. When this was the case, the tracking package was secured with a third knot and the toad was observed again until it showed normal behavior before being released as close as possible to the spot where it was found. If adjustments were necessary, they were performed, and the steps described above were repeated.

2.4.2. Data collection periods

Data were collected over a period of two months, alternately two times in Vienna and two times in Seewinkel, starting at the 24 April in Seewinkel and lasting until 24 June (in total 33 days of data collection). The general procedure was to locate the toads two times each day, once during daylight (between 2 and 5:30 pm) and once at night (starting between 9 and 10:30 pm) using a VHF-homing in procedure (White & Garrott, 1990). Locating refers to directly spotting the toad or being that close that a signal could be detected by using the cable connected to the receiver only (the antenna detached), which means that the toad is within ca. 1 m radius.

In total 19 male toads were successfully tracked (twelve at Kirchsee and seven at the RBP). At the first period at the Kirchsee six male toads were equipped with tracking packages for seven days. This was the initial plan for each tracking period, but successively more tracking packages stopped working properly and we could only equip four toads at the first period in RBP, three toads at the second period at the Kirchsee and two toads at the second period in RBP at once (Tab. 1). In order to obtain a large enough sample size, we performed two rounds at the second Kirchsee and RBP periods. Toads carried the tracking packages for three to seven days, then the devices were removed. For one toad at the second period in RBP the device could not be removed as planned, because it was hiding for a long time under a wooden terrace in a private garden, where we could not reach it. It could not be caught during the lifespan of the battery of the transmitter and was found after 14 days during additional searches (Tab. 1).

Tab. 1: Number of toads, that were equipped with tracking packages at each site with number of days they were tracked. In the second period of the two sites, two rounds were performed (RBP = Rudolf-Bednar-Park; x stands for times).

Date	Location	Nr. of Toads & Nr. of Days
24.04.23 – 01.05.23	Kirchsee	6 toads for 7 days
15.05.23 – 21.05.23	RBP	4 toads for 6 days
23.05.23 – 31.05.23	Kirchsee	2 x 3 toads for 4 days each
07.06.23 – 13.06.23/ 24.06.23	RBP	2 x 2 toads for 3 days & 1 toad for 14 days

2.5 Data analysis

Data were analyzed in the statistical software R (R Core Team, 2020) and QGIS (QGIS Development Team, 2023). Prior to calculating the activity ranges (Minimum Convex Polygons) in R using the package sp (Pebesma & Bivand, 2013) the geodetic datum of the coordinates needed to be transformed. This transformation from EPSG:3857-WGS84 to UTM N33 was performed in QGIS. A comparison of the earlier and later tracking periods was performed by splitting the data set according to the sites and time periods. Maximum and average distances covered by the toads between two localization events were determined by calculating path summaries between two localization spots, using the as.ltraj () function from the R-package adehabitatHR (Calenge, 2006). A Wilcoxon rank sum test was carried out for the above-mentioned comparisons in order to obtain a level of significance and box plots were created. In addition, the mean values of the distances traveled between two localization events were illustrated with the standard deviation for each toad. Toad Axy7_BP_2023_05_16_1 was tracked unintentionally two times. The location data collected in the two periods were combined to form one activity range for this individual. Further it was tested if body mass differed between the day of equipping the toads with the tracking unit and the day of releasing them after the tracking period using a pairwise Wilcoxon rank sum test.

A Spearman's rank correlation between air temperature and traveled distances between two localization events was computed for each site and plotted with a regression line and 95% confidence interval. The daily maximum and minimum temperature values were taken from AccuWeather (*AccuWeather - Österreich Wetter Monatlich*, 2024) and the calculated average for each day was used for the correlation.

All maps from Kirchsee (Google, n.d.) and Rudolf-Bednar-Park (Google, n.d.) were created using QGIS and Google Maps Satellite images as background, only for two zoomed-in inserts Open Street Map (Open Street Map, n.d.) was used. The paths were generated in QGIS with the points-to-path tool, which converts the layer with the data points to a line vector layer.

3. Results

In total a number of 19 toads were equipped with tracking packages and located twice a day using the homing-in method (White & Garrott, 1990). The data collected yielded an average activity range of 9 916.83 m² at the Kirchsee and 257.47 m² at the RBP (Fig. 2, Tab. 2).
The distances moved were quite variable between individual toads, especially in RBP, but also at the Kirchsee. The largest activity range at the Kirchsee was with 30 519.24 m² also the largest activity range recorded in this study. The largest activity range in RBP with 898.41m² was two decimal orders smaller than the largest activity range at the Kirchsee, although this toad was tracked unintentionally twice and therefore for a longer period. Minimum activity ranges were 728.53 m² at the Kirchsee and 16.74 m² in RBP (Tab. 2).
A comparison between all activity ranges (Minimum Convex Polygons) shows highly significant differences ($z = 3.3384$ $p = 0.0002$) in sizes between the Kirchsee and the RBP (Fig. 2).

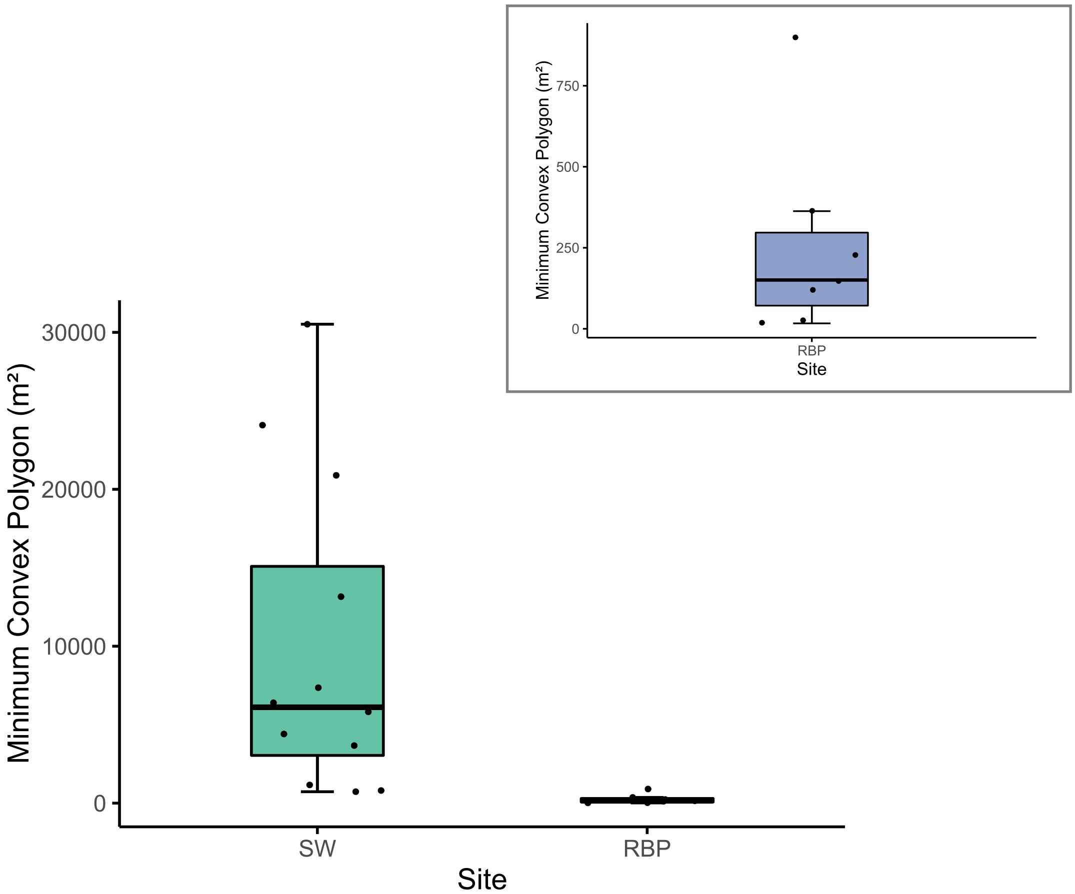

Fig. 2: Activity ranges (equals Minimum Convex Polygons) of all toads at the Kirchsee and at the Rudolf-Bednar-Park (RBP).
Top-right: Zoomed-in window with the activity ranges (Minimum Convex Polygons) of RBP. The outlier in the zoomed-in window with an activity range of almost 900 m² was caused by an unintentional double-tracking of one toad. The resulting data points of the two time periods were combined to form one activity range.

Tab. 2: Activity ranges of all toads tracked at the Kirchsee (on the left) and at the Rudolf-Bednar-Park (RBP) (on the right). * toad was unintentionally tracked two times

ID - toads Kirchsee	Area (m²)	Round	ID - toads RBP	Area (m²)	Round
Axy1_SW_2023_04_24_5	**1 159.80**	round1	Axy2_BP_2023_05_15_5	**25.388**	round1
Axy2_SW_2023_04_24_1	**728.53**	round1	Axy5_BP_2023_05_15_7	**363.03**	round1
Axy4_SW_2023_04_25_2	**7 357.08**	round1	Axy6_BP_2023_05_15_6	**16.74**	round1
Axy5_SW_2023_04_24_4	**20 888.81**	round1	Axy7_BP_2023_05_16_1	**898.41 ***	round1
Axy6_SW_2023_04_24_3	**807.46**	round1	Axy5_BP_2023_06_10_4	**230.63**	round2
Axy7_SW_2023_04_24_2	**30 519.24**	round1	Axy6_BP_2023_06_07_3	**150.55**	round2
Axy2_SW_2023_05_23_1	**6 400.67**	round2	Axy6_BP_2023_06_10_3	**117.53**	round2
Axy2_SW_2023_05_27_1	**13 154.29**	round2			
Axy5_SW_2023_05_23_3	**24 088.39**	round2			
Axy5_SW_2023_05_27_2	**3 669.00**	round2			
Axy6_SW_2023_05_23_2	**4 409.75**	round2			
Axy6_SW_2023_05_27_3	**5 818.91**	round2			

The comparison of activity ranges between the two tracking periods at the Kirchsee (1st starting on 24 April and 2nd starting on 23 May) with each other revealed no significant difference ($z = 0.4003$, $p = 0.6991$) (Fig. 3). Also, analysis of possible differences between the two tracking periods at RBP (1st starting on 15 May and 2nd starting on 7 June) with each other showed no significant difference ($z = 0.1768$, $p = 1$) (Fig. 3).

All expect one activity range of the Kirchsee were larger than those at the RBP. The toad Axy5_SW_2023_05_23_3 (Fig. A 8) that traveled the longest distance (380.8 m) between two localization points was a different one than the toad with the largest activity range Axy7_SW_2023_04_24_2 (Fig. A 6) which only reached the third furthest distance 302.3 m (Fig. 4). Toad Axy5_SW_2023_05_23_3 (Fig. A 8) traveled also on average the longest distances between two localization events (144.3 m) (Fig. 5). The toad Axy7_SW_2023_04_24_2 (Fig. A 7) with the largest activity range moved an average of 71.9 m between two localization points (Fig. 5).

All but two toads had reduced body mass at the end of the tracking period compared to before. When comparing the body mass measurements using a pairwise Wilcoxon rank sum test the relationship is on the limit of being significant ($z = 1.9656$, $p = 0.0493$) (Fig. 6). One of the toads (Axy5_BP_2023_06_10_4) that gained weight was tracked in Mid-June and no longer participated in the mating activities (for localizations of this toad see Fig. A 18). The other toad (Axy6_BP_2023_06_07_3), whose body mass has increased was regularly migrating to the water basins (Fig. A 17).

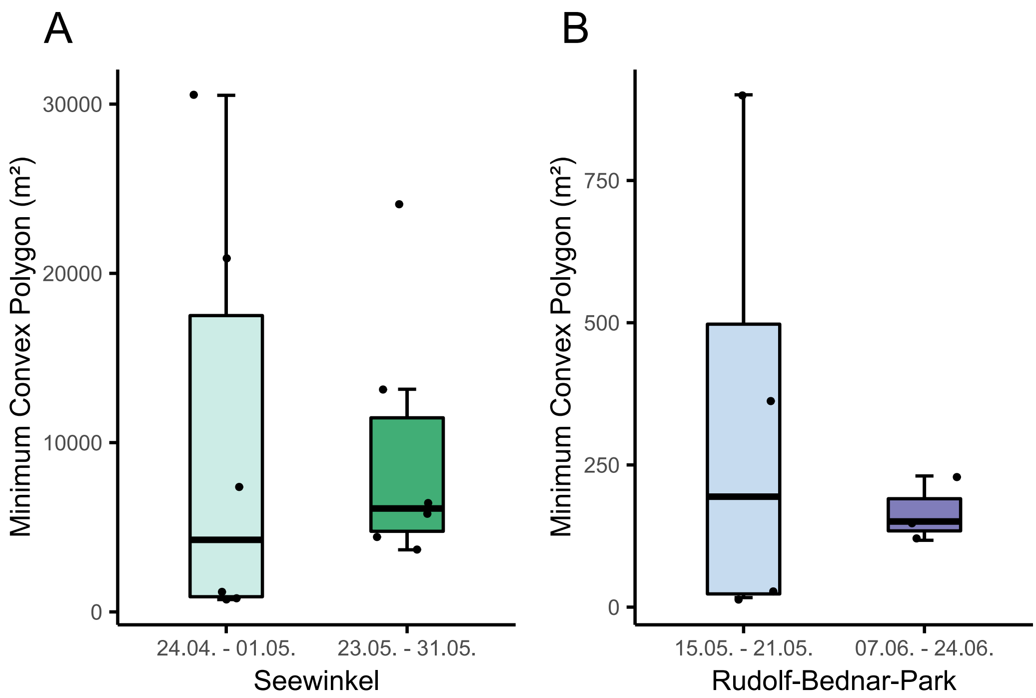

Fig. 3: Comparison of activity ranges between the two tracking periods (equals Minium Convex Polygons) of the twelve toads tracked at Seewinkel (Kirchsee) (A) and the seven toads tracked at the Rudolf-Bednar-Park (B).

Data set divided according to the two tracking periods: at the Kirchsee the 1st round started at the end of April and 2nd round started at the end of May with 1 month in between. At Rudolf-Bednar-Park the 1st round started in the middle of May and 2nd round started at the beginning of June, with three weeks in between. There is no statistically significant difference between the two rounds in both habitats.

In addition to the determination of activity ranges and trajectories, a Spearman rank correlation of the average tracking trajectories and the average temperature of that date was performed. All trajectories were calculated for each toad in both sites (one trajectory equals the distance between two localizations points). The correlation between distances traveled and temperature was positive and statistically significant for the site Kirchsee ($r = 0.5832$, $p = 0.0140$), but not for the RBP ($r = 0.3200$, $p = 0.2647$) (Fig. 7).

Overview maps with the paths from all tracked toads are presented in Fig. 8 (Kirchsee) and Fig. 9. (RBP). The maps with the individual paths of the two sites can be found in the Appendix (Fig. A 1 to Fig. A 19). In round one at the Kirchsee, there was a highly frequented spot, where many of the equipped toads were located most of the time (Fig. A 1, Fig. A 2, Fig. A 5). Only the individuals Axy4_SW_2023_04_25_2 and Axy5_SW_2023_04_24_4 (Fig. A 3, Fig. A 4) have moved over a larger area of the soda pan. Toad Axy7_SW_2023_04_24_2 was the only tagged toad of the round in April that moved to the westside of the soda pan (Fig. A 6).

During the second period at the Kirchsee, we equipped three toads from the north side of the soda pan from which two moved over a relatively large area (Fig. A 8, Fig. A 9) and three toads from the westside of the Kirchsee, which also covered a quite large area (Fig. A 10, Fig. A 11, Fig. A 12). During the tracking period in the second half of May, there was no longer such a spatially limited highly frequented spot.

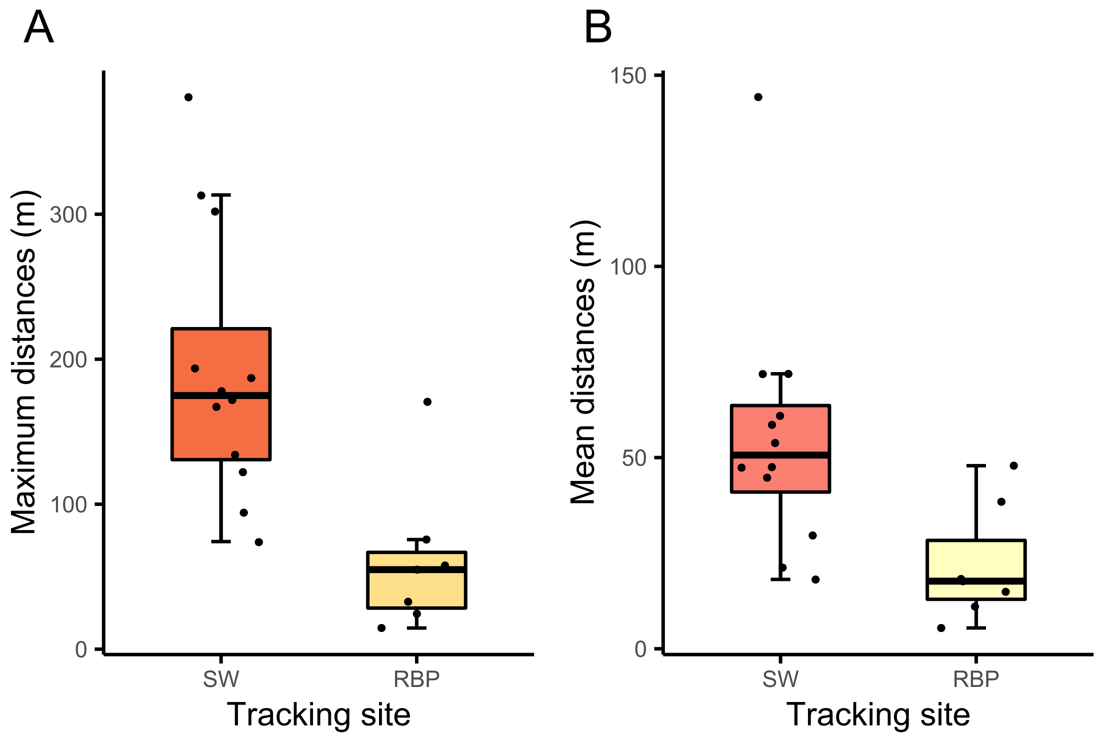

Fig. 4: Mean distances (A) and maximum distances (B) traveled between 2 localization events of all toads tracked in Seewinkel (Kirchsee) and at the Rudolf-Bednar-Park.

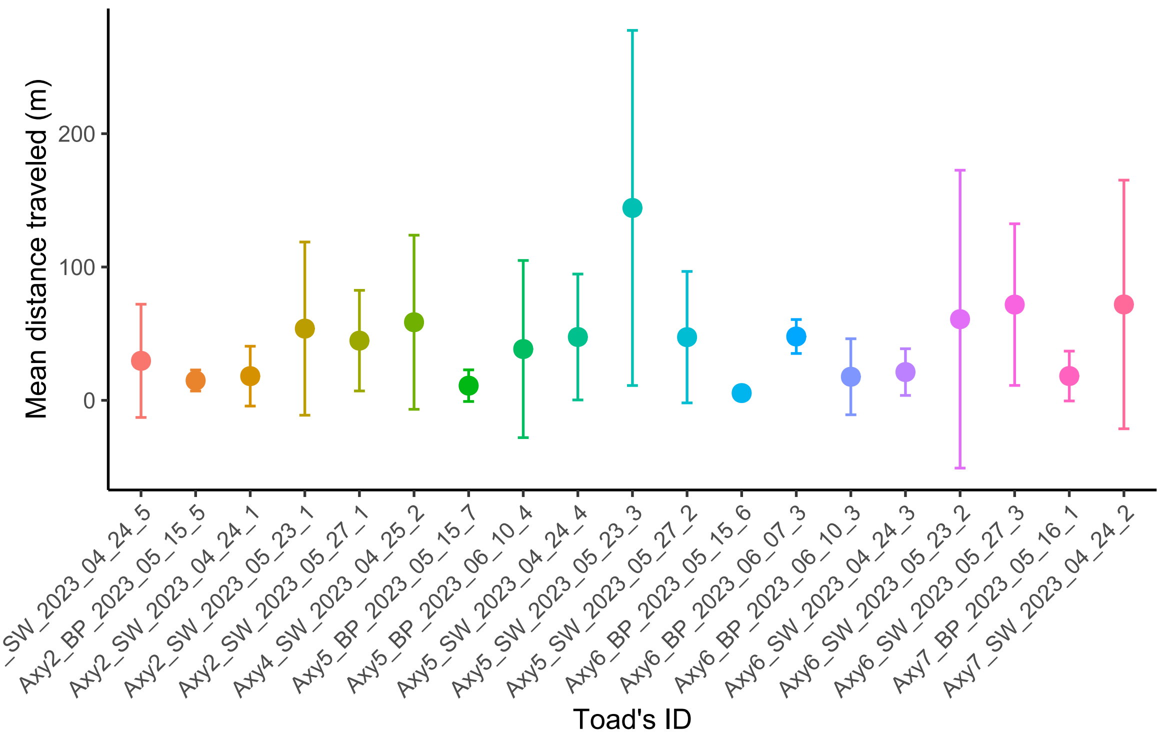

Fig. 5: Mean distances traveled between 2 localization events shown for each toad.

In the urban habitat at the RBP, we equipped four toads during the first round, which were in the water basins calling during the night and in hiding spots during the day, as expected. One day before the transmitters were due to be removed according to schedule, we found toad Axy5_BP_2023_05_15_7 dead and completely dried up next to the transmitter on the side of the footway (see Fig. A 14 for localizations of the toad). Another transmitter was lying a few meters away and the toad Axy6_BP_2023_05_15_6 was not detectable (see Fig. A 15 for localizations). Judging by the way the transmitter was detached and lying next to the toad, human intervention cannot be ruled out.

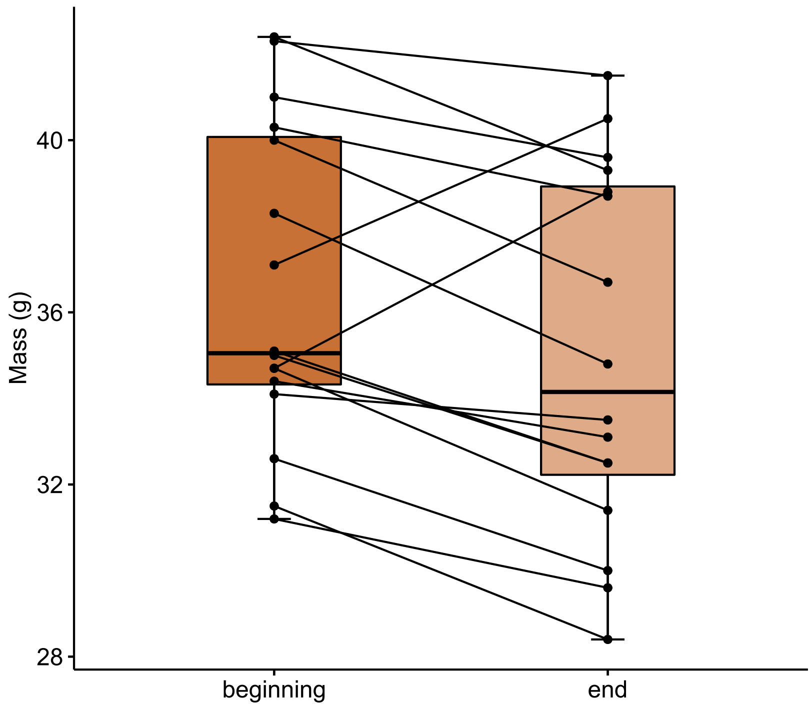

Fig. 6: Body mass of all toads measured before equipping the toads with transmitter packages and after removing them. Most of them did lose weight during the tracking period. Some toads could not be weighed after the tracking period, because they were in amplexus, these have not been included in the analysis (here: n=16).

The second round in the RBP was towards the end of the spawning season in the year 2023. The number of calling males had decreased clearly (pers. obs.) and two of our three tagged toads did not migrate to the spawning waters anymore in the nights after we had equipped them with a tracking package at a water basin. Toad Axy6_BP_2023_06_10_3 had to carry the transmitter for a longer period (14 days) because it was hiding under a private wooden terrace where we could not reach it.

We found that throughout the main breeding season, the male toads did not move far from the spawning waters during the day to look for hiding places. The toads crawled between vegetation in the damp transition zone between land and water body or they pressed themselves flat on the ground on land, so that they were hard to spot. Sometimes they burrow in a bit, or they stay in the water and hide between vegetation under water.

Six of the tagged toads were found in amplexus, some of them for several consecutive days with the same female, and spawning strings were recorded in both study areas.
We found that the green toads did not move far from the soda pan during the day. They tended to hide between vegetation in the still very damp transition area between land and water or on land, sometimes burrowing in or staying in the water hiding between the vegetation. In the RBP they also used spots very close to the breeding pools, such as drainage holes, flower beds or they crawled under a sculpture situated close to the water basins. One time a toad was also located in the water basin during the day, using it as a hiding spot and not for advertising.

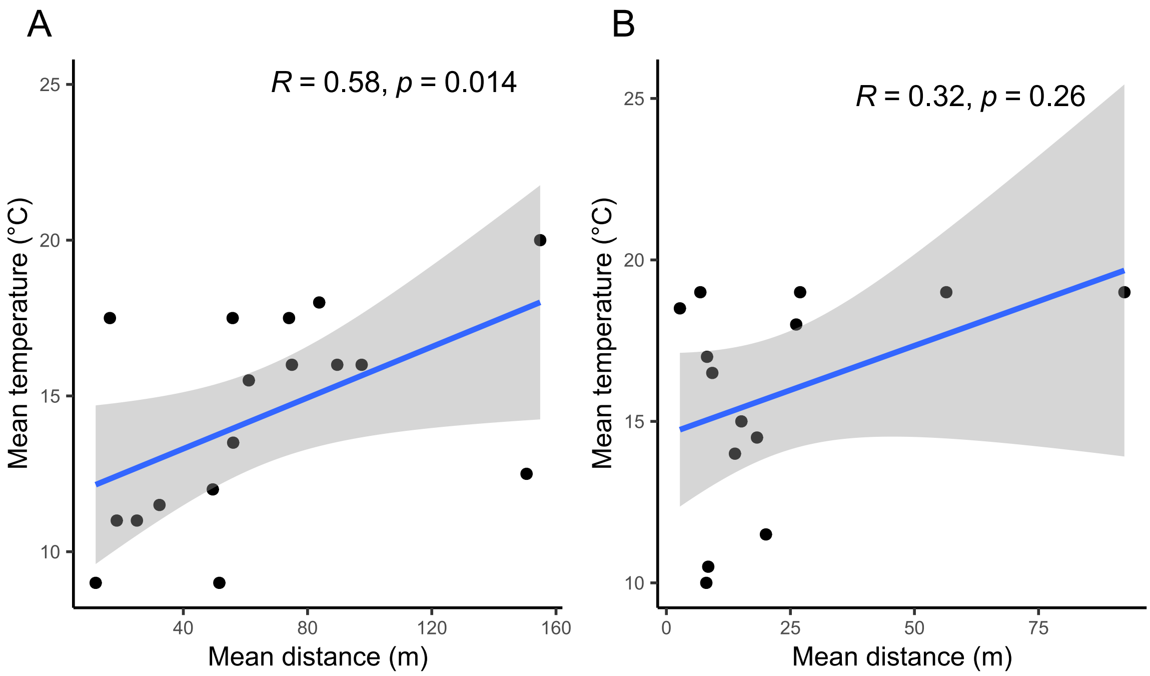

Fig. 7: Correlation of the average tracking trajectories between 2 localizations per toad at the Kirchsee (on the left) and at the Rudolf-Bednar-Park (on the right) and the average temperature per day. The relationship of the Kirchsee was statistically significant, but not the relationship at the Rudolf-Bednar-Park.

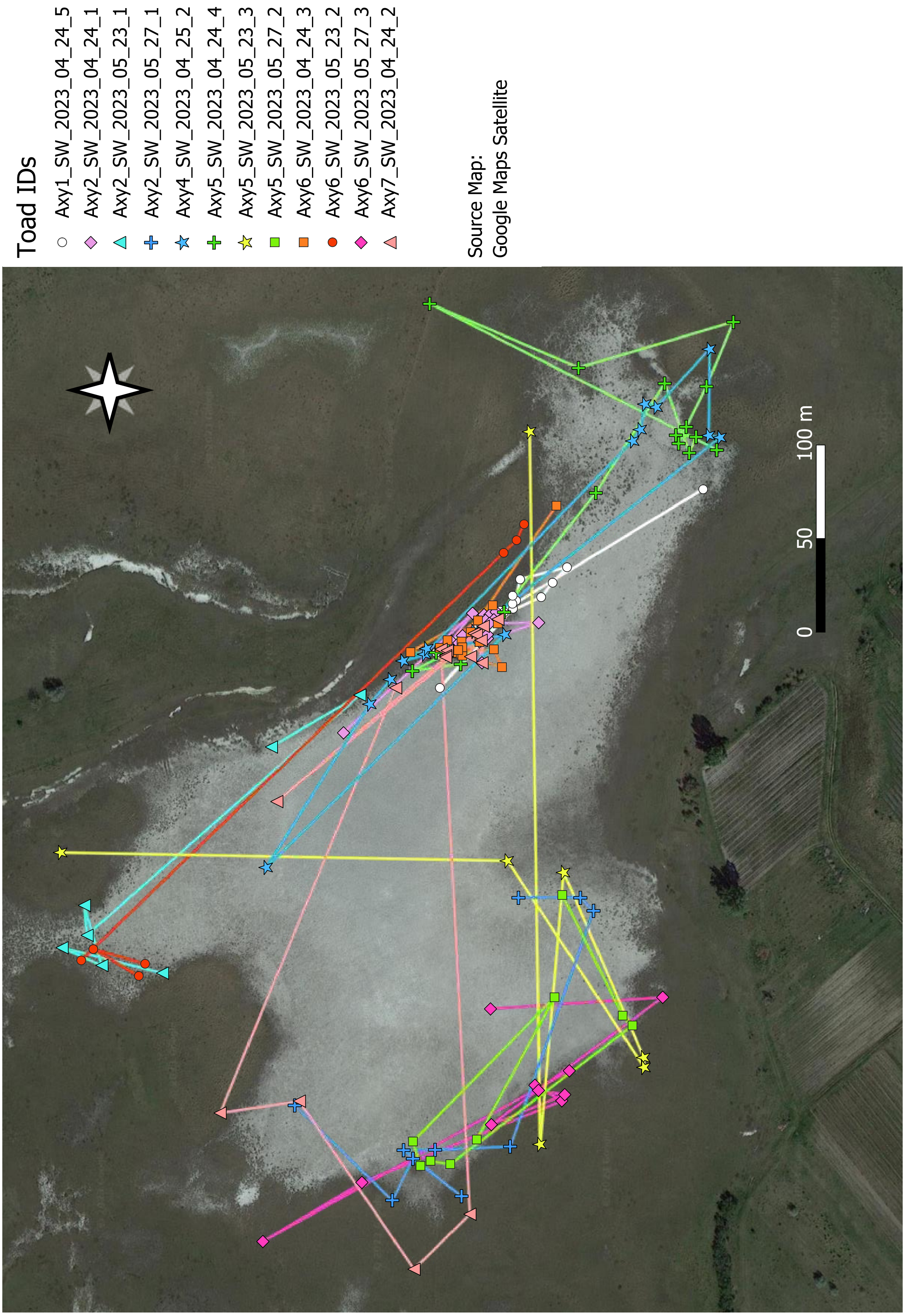

Fig. 8: Kirchsee

Recorded locations of all 12 individuals tracked at the Kirchsee, connected with lines. The purpose of this map is to give an impression of the position of the tracking paths in relation to each other. For individual paths, see the Appendix.

Toad IDs

Tracking_data_order
- Axy2_BP_2023_05_15_5
- Axy5_BP_2023_05_15_7
- Axy5_BP_2023_06_10_4
- Axy6_BP_2023_05_15_6
- Axy6_BP_2023_06_07_3
- Axy6_BP_2023_06_10_3
- Axy7_BP_2023_05_16_1

Source Map:
Google Maps Satellite

Fig. 9: Site Rudolf-Bednar-Park
Recorded locations of all 7 individuals tracked at the Rudolf-Bednar-Park, connected with lines. The purpose of this map is to give an impression of the position of the tracking paths in relation to each other. For individual paths, see the Appendix.

4. Discussion

The difference in the size of activity ranges in the Rudolf-Bednar-Park (RBP) and the natural habitat at Neusiedlersee/Seewinkel (Kirchsee) was very clear and significant. The largest activity range in RBP was with 898.41 m² two decimal orders smaller than the largest activity range at the Kirchsee with 30 519.24 m², although this toad in the RBP was an outlier due to an unintentional second tracking. One of the reasons for this incongruity in activity ranges is likely due to the limited space that can be used as habitat in the city (Kovács & Sas, 2010b; Palmer, 2003). Other conceivable reasons for smaller activity ranges are the many asphalted areas and the high frequency of park visitors mostly during the day, but some people also spend time at the RBP at night (pers. obs.).
There were also big individual differences at both habitats, the smallest activity range at the Kirchsee (728.53 m²) was even a bit smaller than the largest activity range at the RBP (although due to the longer tracking period). According to Calder (2001) body size has a major impact on the everyday life of an animal and its physiological properties, which probably also influences the different individual home range sizes.

Indermaur et al. (2009) studied space use of *B. viridis* at a natural study site, the Tagliamento river in Italy and determined home ranges also with transmitters mounted on the back of the toads and their whole tracking package did not exceed 10% of the toads' body mass either. Therefore, the tracking method is quite comparable to ours, however, Indermaur et al. (2009) tagged the toads at a later date (from June until September) and their calculation of the home ranges was performed in a slightly different way. They calculated Fixed-Kernel home ranges with a smoothing factor and discarded the outer 5% of the data, as they no longer considered this to be part of the home range. Applying this method, they determined an average home range of 2 456 m², which would be smaller than the arithmetic mean of the activity ranges at the Kirchsee with 9 916.83 m². The home ranges of Indermaur et al. (2009) should still be comparable to ours and the differences in average home ranges is that clear that they can be considered as not of methodological origin, nonetheless it is important to consider the seasonal variation. What can explain the dissimilarity are differences in the habitat structures, food availability and predator density (Indermaur et al., 2009), as the soda pan Kirchsee and the Tagliamento river with many gravel banks certainly offer divergent conditions for *B. viridis*. Additionally, three green toads at the Kirchsee (Axy7_SW_2023_04_24_2, Axy5_SW_2023_05_23_3, Axy5_SW_2023_04_24_4) had outstanding large activity ranges, which certainly has amplified the difference of the mean values.

In the first round of our data acquisition at the Kirchsee, there was a highly frequented spot, where many of the equipped toads were located most of the time and from which they did not move far away, such as the individuals Axy1_SW_2023_04_24_5, Axy2_SW_2023_04_24_1 and Axy6_SW_2023_04_24_3. There were only a few individuals that moved over larger areas, while trying to attract females (e.g. Axy4_SW_2023_04_25_2 and Axy5_SW_2023_04_24_4). The advantage of staying in such a highly frequented spot can be on the one hand that the resulting chorus increases the probability of successfully attracting females and on the other hand reduces the probability of being eaten by predators, as the male toads are protected in the bulk. These hypotheses were examined on the example of the tungara frog (*Physalaemus pustulosus*) by Ryan et al. (1981) and proved to be true for the studied tungara frog population. A possible disadvantage of aggregating to a calling group would be that the whole group is discovered by predators at once, but with the tungara frogs the likelihood of being eaten by a predator was lower for the individual when it called in a chorus. What limits the comparability of tungara frogs with green toads is for example that the tungara frogs have quite different predators, such as the fringe-lipped bat (*Trachops cirrhosu*), the four-eyed opossum (*Philander opossum*) and the large crab (*Potamocarcinus richmondi*). Predators of adult *B. viridis* in Austria are mainly raptorial birds (e.g. carrion crows – *Corvus corone*) and grass snakes (*Natrix natrix*) (Rienesl, 2021). Chorusing behavior was also described in the natterjack toad, for instance by Arak (1983) and Tejedo (1993). Tejedo (1993) observed a tendency for *E. calamita* males from smaller groups to join larger groups, but this resulted in a higher mating rate with females only in one of three years. The number of females monitored was more dependent on environmental factors such as ambient temperature and rainfall.

During the second tracking period at the end of May, the green toads we studied showed a different movement behavior. They covered longer distances on average, which can probably be explained by the higher activity of the animals in general that we observed during this warmer phase. Although a correlation between temperature and distance traveled showed a positive relationship for the natural study site, there was no significant difference in the sizes of the activity ranges between the first and the warmer second round. This finding suggests that the toads were more active, but not in a larger space. The increased activity could also explain why there was no longer such a spatially limited highly frequented spot. With the small sample of six animals, we do not know the locations of all the toads in the soda pan, but it was ascertained that none of our sample toads visited the spot that was very popular one month earlier. The avoidance of breeding activity in areas where there are older tadpoles would be consistent with findings of *B. viridis* tadpoles growing slower when there is strong interspecific competition (Katzmann et al., 2003). Additionally, the tadpoles rarely show cannibalistic behavior, when there is a large accumulation of tadpoles (Kovács & Sas, 2009).

The body mass of our toads was slightly but significantly reduced after the tracking period. Indermaur et al. (2008) reported that their devices the green toads had to carry, which were comparable to ours, were very unlikely to have a negative effect on body mass. Also, for our study we assume that the devices had no effect on the energy consumption of the green toad, but that the mass reduction was due to the regular body mass loss during the mating season. This assumption is supported by the finding of Ott (2015), who determined a decrease in body mass too, in a capture-recapture study without tagging during the spawning period. Further, one of the two toads that gained weight during the tracking period in our study no longer took part in the exhausting mating activities. During the tracking period at the end of May, increased activity and calling was observed during the day as it was also noticed during a later spawning period at another location by Ott (2015).

None of our tracked male toads changed spawning waters (not counting the neighboring basins in RBP), individual toads even seemed to prefer certain areas within the Kirchsee and also in RBP only two toads (Axy5_BP_2023_05_15_7 and Axy7_BP_2023_05_16_1) were found in more than one basin. Adult green toads predominantly stay in one habitat, nevertheless they are described as pioneer species, but this trait appears to be more evident in young toads (Leskovar & Sinsch, 2005; Semlitsch, 2008).

Sinsch et al. (2012) stated that a complete picture of movement behavior and habitat use, which is necessary for conservation measures, can only be obtained by conducting a survey throughout all annual life-history phases of the animal. The scope of this study did not allow for a longer survey, but in the previous year toads were tagged after the main spawning season in the RBP by Spießberger et al. (2023), which together with this study covers a long period of time for which spatial information about the green toads is available.
If technically possible, longer periods during which an animal is tagged should be aimed for. Indermaur et al. (2009) claim that 25 localizations are necessary to calculate a solid home range. The fact that one toad we unintentionally tracked for two periods had a larger activity range when summarizing the localizations of both periods than when considering only one, shows that the green toads may use a larger area over a longer period and that we have not recorded their entire home range with a maximum of 14 localizations in a regular tracking period at the urban site and a maximum of 19 localizations at the natural site. To emphasize this, we used the term activity range for the movement ranges we measured.

The method of attaching the transmitters was not optimal despite long preliminary fitting attempts, as the transmitters and silicon tubes rubbed against the skin of the toads and sometimes left black chafes. Ott (2015) had even more severe problems, he had to remove the transmitter from one male toad ahead of schedule due to skin irritations and another toad died from injuries caused by the tagging.
In any case it is important to visit the animals regularly with this tracking method, especially in the first few days after mounting the tracking units, as they can slip to the belly-side (happened a few times with our toads), which makes the signal worse and is certainly not comfortable for the animals. But when they are regularly located, the transmitter can easily be brought back into the correct position by adjustment on-site.

The natural environment in the current study is an example of a habitat that is increasingly destroyed not by direct development but through extensive exploitation of the groundwater by humans in this area. This groundwater abstraction leads to increasingly silting of the highly sensitive soda pans in the national park Neusiedlersee/Seewinkel, some are already completely dry (Krachler et al., 2012). When the soda pans are dried out, the habitat is no longer suitable for green toads. Their search for compensatory spawning waters presumably leads them to more anthropogenic habitats, such as the garden ponds in the villages (Amon & Tobler, 2021). Stöck et al. (2008) states that the most common habitat for the green toad in Eastern Germany are gardens. The usage of a habitat in villages or cities by wild animals can thus also be motivated by the lack of natural habitats (Brand & Snodgrass, 2009).
An alternative reason for occurrence in an urban environment may also be that the city is expanding, and some green toad populations managed to find a habitat in the now much more urban area, like it was the case with Viennese green toads (Rienesl, 2021). Occupation of such habitats, however, is usually accompanied by a decline or disappearance of populations. For example Kühnel & Krone (2003) observed a strong decline of green toads after the completion of construction projects in Berlin. Disappearance of populations that still existed in 1992-1994 is also described by Mazgajska & Mazgajski (2020) in the city Warsaw (Poland) and Beutler &

Heckes (1991) reported that at their surveys in Munich in 1993 no other species showed such a clear decline in population numbers compared to the 1970s as the green toad.

The population in the RBP, from which we have equipped selected toads with tracking packages, seems to be at least stable, even if the physical condition (SMI-index) is on average lower compared to other populations (Landler et al., 2022). The water basins in RBP are not ideal for *B. viridis*, as they are ~10 cm deep and green toads choose if possible waters with a depth over 20 cm according to Hemmer & Kadel (1970). Most of the basins are also rich in reeds, but green toads prefer spawning waters with little vegetation (Glandt, 2015). However, there are also areas with no vegetation in the basins, which the toads choose for spawning. All in all, the water basins can be valued as suitable for green toads, as they have been used for at least 12 years (Csarmann, 2012). Although the condition of the water in the basins often appeared to be in a worrying state. Some of them are heavily polluted and covered in algae later in the year, also garbage and cigarette stubs were repeatedly found in the basins (pers. obs.). But enough tadpoles seem to be developing at least to maintain the population. As metamorphs and adults they then face a habitat with many people, dogs, crows and asphalted areas which probably leads to injuries and increased mortality (several dead green toads were found during data collection in the RBP, while none were found at the Kirchsee).

But the RBP provides specific hiding places that do not exist in natural habitats and which were detected for the first time using the radiotelemetry method, namly water drainage holes. We do not have enough information to judge the qualtiy of these hiding spots, but we noticed that when it rains heavily, the shafts fill up to the top with water, nervetheless they seem to be used frequently. For example toad Axy2_BP_2023_05_15_5 used exclusively a specific water drainage hole as day hiding spot and moved a few meters to the same water basin during every night we tracked it. Assigning green toads to a category of the adaper-exploiter hypothesis (Rogers et al., 2021), they should be categorized as adapters to urban habitats, according to what has been discussed so far, because they rather survive than thrive.
The Kirchsee on the contrary has relatively sparse vegetation and meets the criteria of a spawning ground for green toads as described in the literature (a.o. Stöck et al., 2008). It was directly observed how pairs span the spawning strings between blades of grass or similar.

In conclusion, the large difference in activity ranges of the green toads at the Kirchsee and at the RBP has many reasons and cannot be simply explained. Differences in the habitat structures, as the surrounding of the park area by buildings and the many asphalted surfaces, which both do not exist in the natural habitat, likely have an influence. The numerous factors that affect the size of the toads' activity ranges in two different habitats makes every population unique and comparing them very difficult (Indermaur et al., 2009). Nevertheless, data as those recorded in this study can help formulating better hypotheses when conducting tracking studies in various environments in Austria or even Europe and therefore answer research questions in a more targeted manner.

The dataset generated indicates a potential adaptation of green toads' spatial activity to different habitats (urban versus natural) and can be used as preliminary data to apply this approach to several sites along a human disturbance gradient for populations in Europe. Further, these findings can provide information for developing guidelines for management measures, as the movement behavior and spatial activity in habitats are an important information needed for management applications (Groff et al., 2017; Katzner & Arlettaz, 2020). To be able to provide suitable habitats for amphibians, creative solutions are needed now and will be needed even more in the future. A prerequisite for this solution is prior knowledge about the species of interest including expertise about movement behavior, which is only rarely applied for conservation measures yet and there are still barriers to overcome in the evaluation of tracking data for practical application (Katzner & Arlettaz, 2020), but it is certainly worth further exploring this avenue.

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Appendix

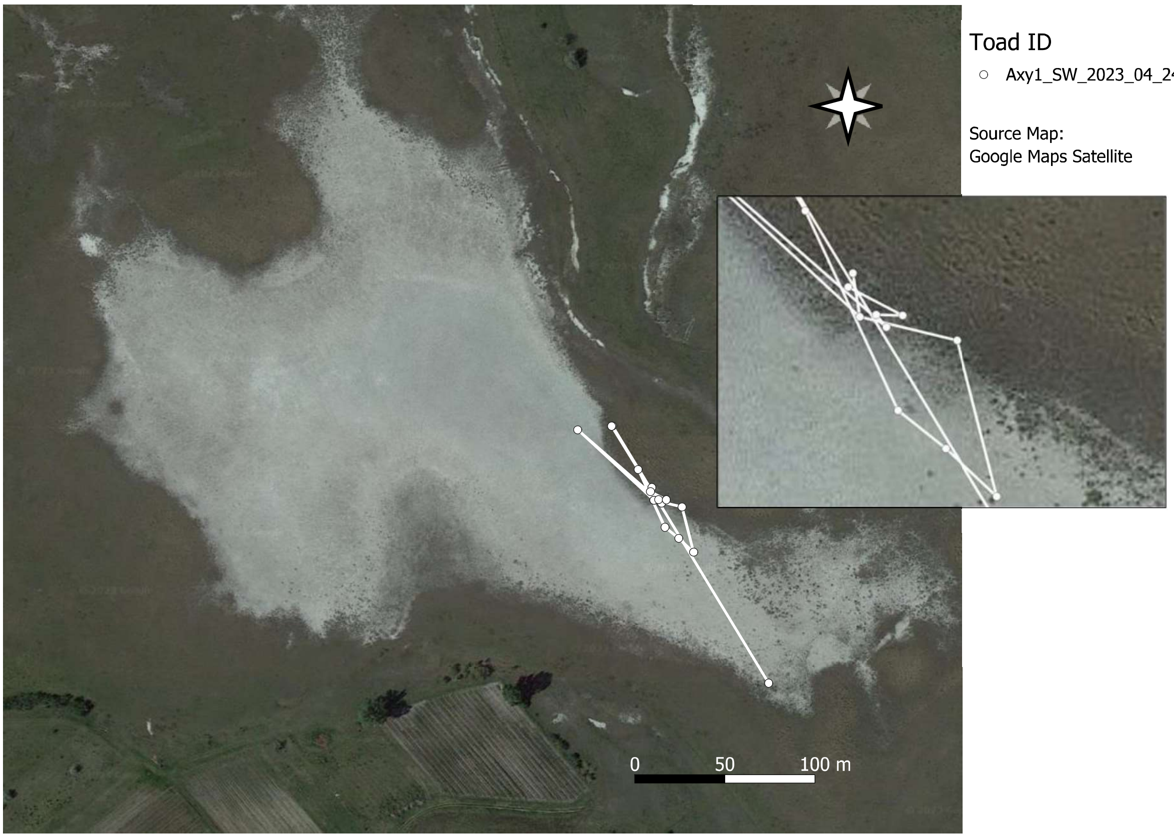

Fig. A 1: Site Kirchsee: Round 1
Recorded locations of individual Axy1_SW_2023_04_24_5, connected with lines.

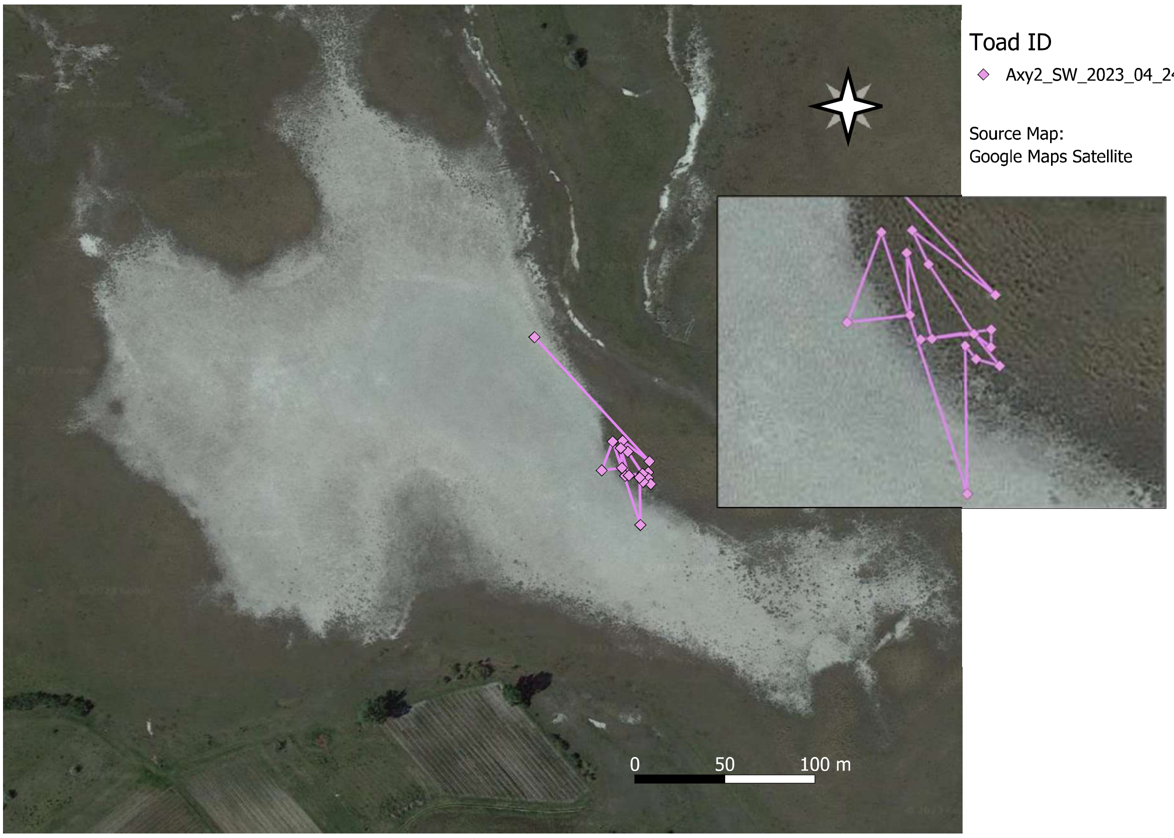

Fig. A 2: Site Kirchsee: Round 1
Recorded locations of individual Axy2_SW_2023_04_24_1, connected with lines.

Fig. A 3: Site Kirchsee: Round 1
Recorded locations of individual Axy4_SW_2023_04_25_2, connected with lines.

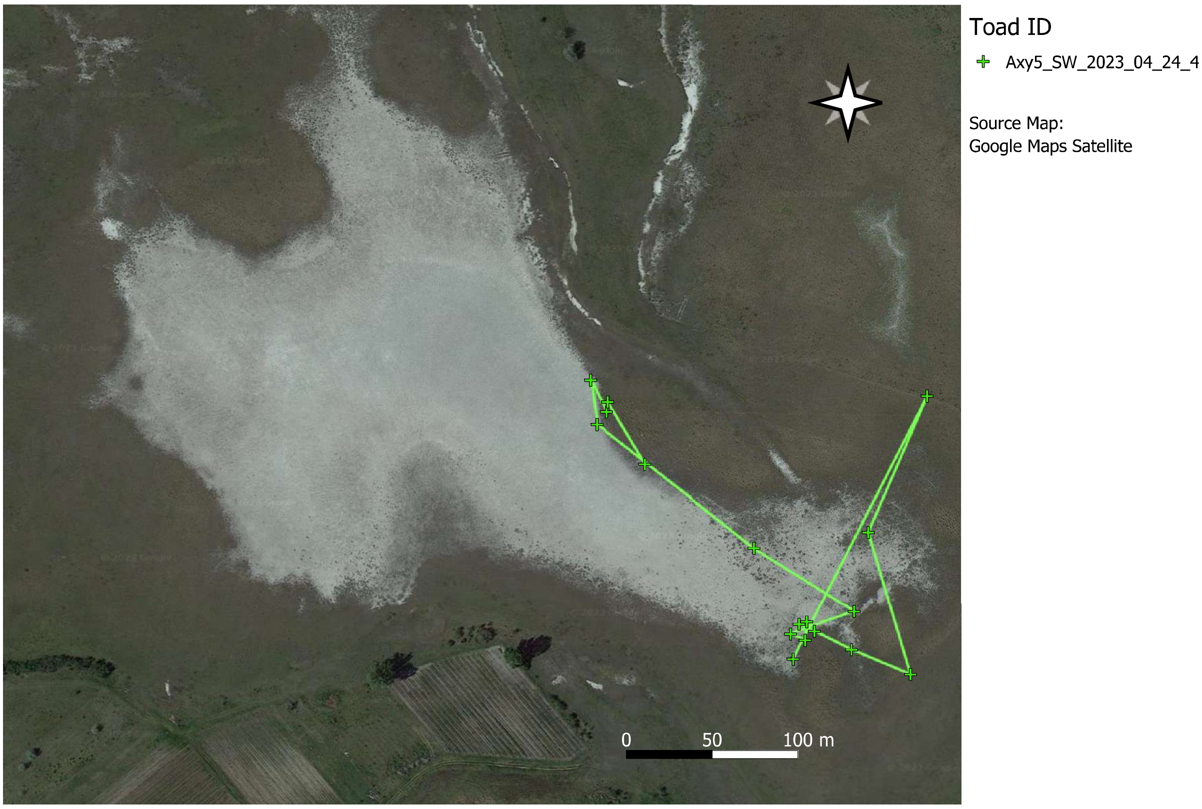

Fig. A 4: Site Kirchsee: Round 1
Recorded locations of individual Axy5_SW_2023_04_24_4, connected with lines.

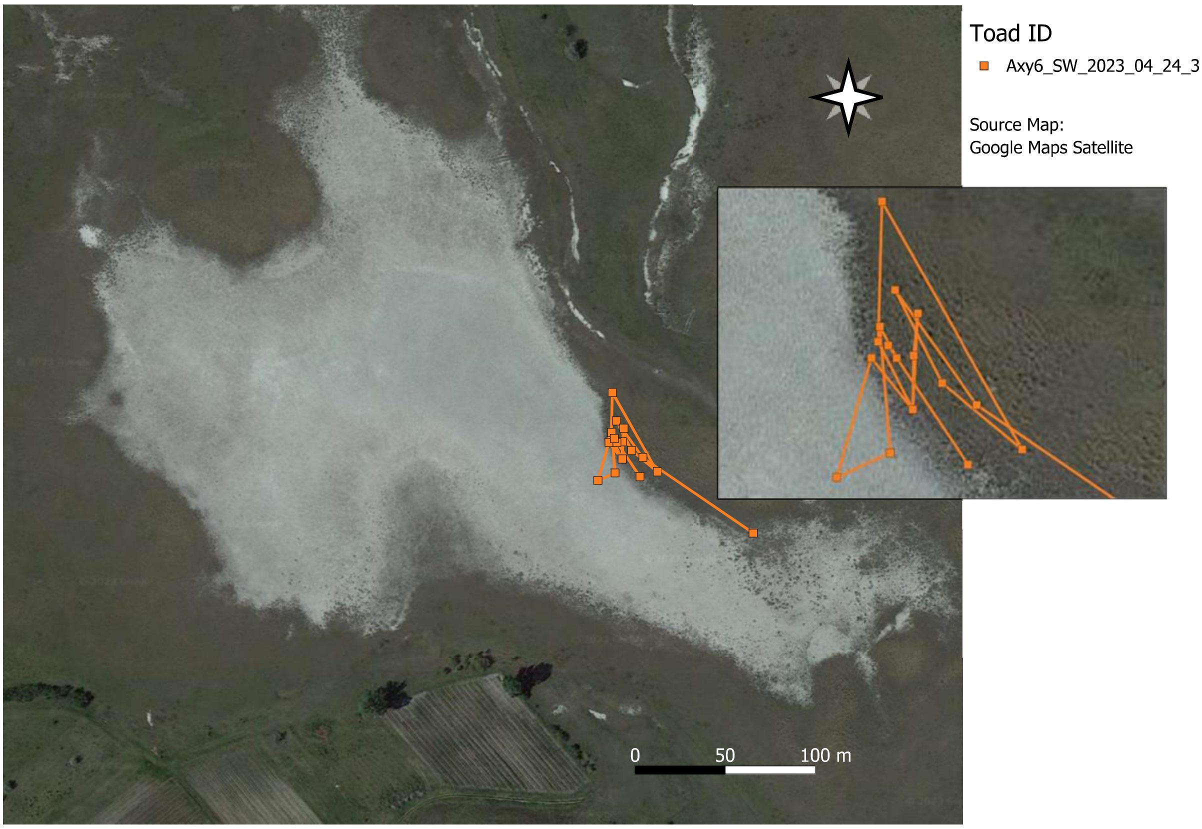

Fig. A 5: Site Kirchsee: Round 1
Recorded locations of individual Axy6_SW_2023_04_24_3, connected with lines.

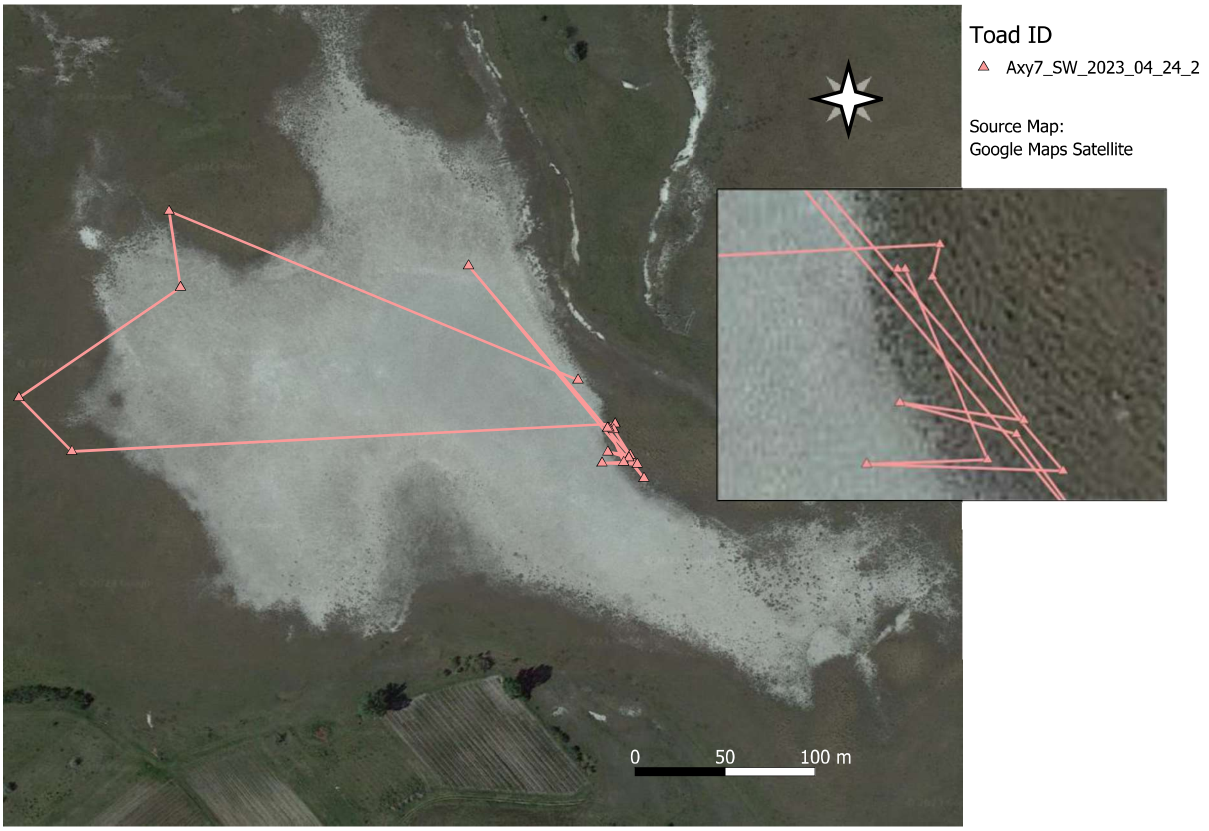

Fig. A 6: Site Kirchsee: Round 1
Recorded locations of individual Axy7_SW_2023_04_24_2, connected with lines.

Fig. A 7: Site Kirchsee: Round 2 (1st Run)
Recorded locations of individual Axy2_SW_2023_05_23_1, connected with lines.

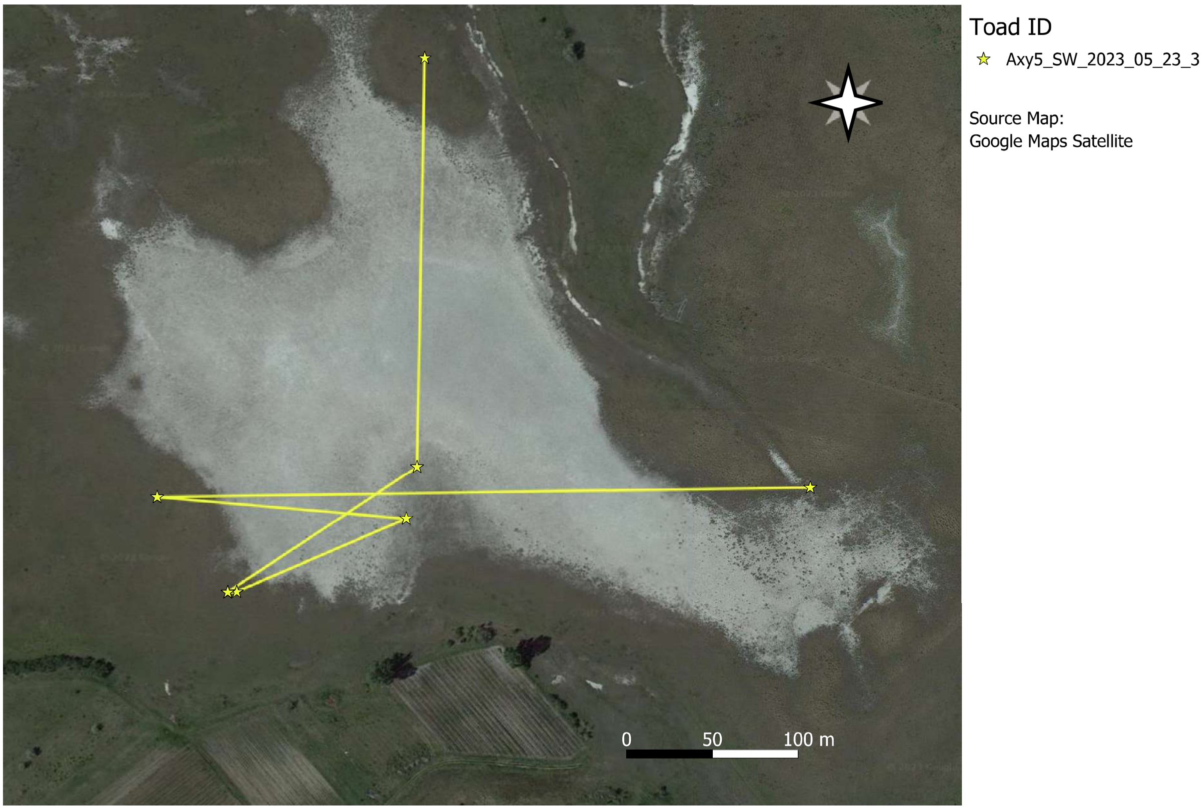

Fig. A 8: Site Kirchsee: Round 2 (1st Run)
Recorded locations of individual Axy5_SW_2023_05_23_3, connected with lines.

Fig. A 9: Site Kirchsee: Round 2 (1st Run)
Recorded locations of individual Axy6_SW_2023_05_23_2, connected with lines. The water level was higher during data acquisition; therefore the 3 southern points were in the water zone at this time.

Fig. A 10: Site Kirchsee: Round 2 (2nd Run)
Recorded locations of individual Axy2_SW_2023_05_27_1, connected with lines.

Fig. A 11: Site Kirchsee: Round 2 (2nd Run)
Recorded locations of individual Axy5_SW_2023_05_27_2, connected with lines.

Fig. A 12: Site Kirchsee: Round 2 (2nd Run)
Recorded locations of individual Axy6_SW_2023_05_27_3, connected with lines.

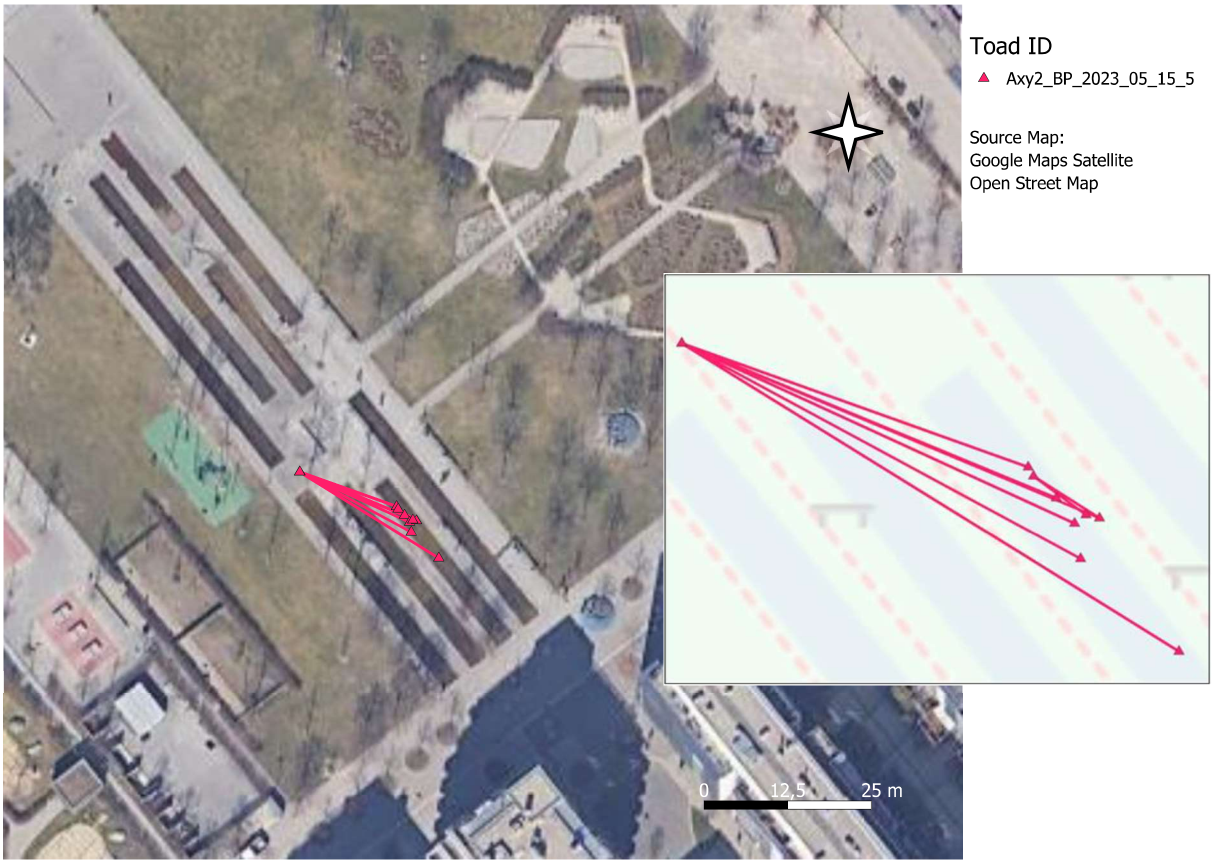

Fig. A 13: Site Rudolf-Bednar-Park: Round 1
Recorded locations of individual Axy2_BP_2023_05_15_5, connected with lines.

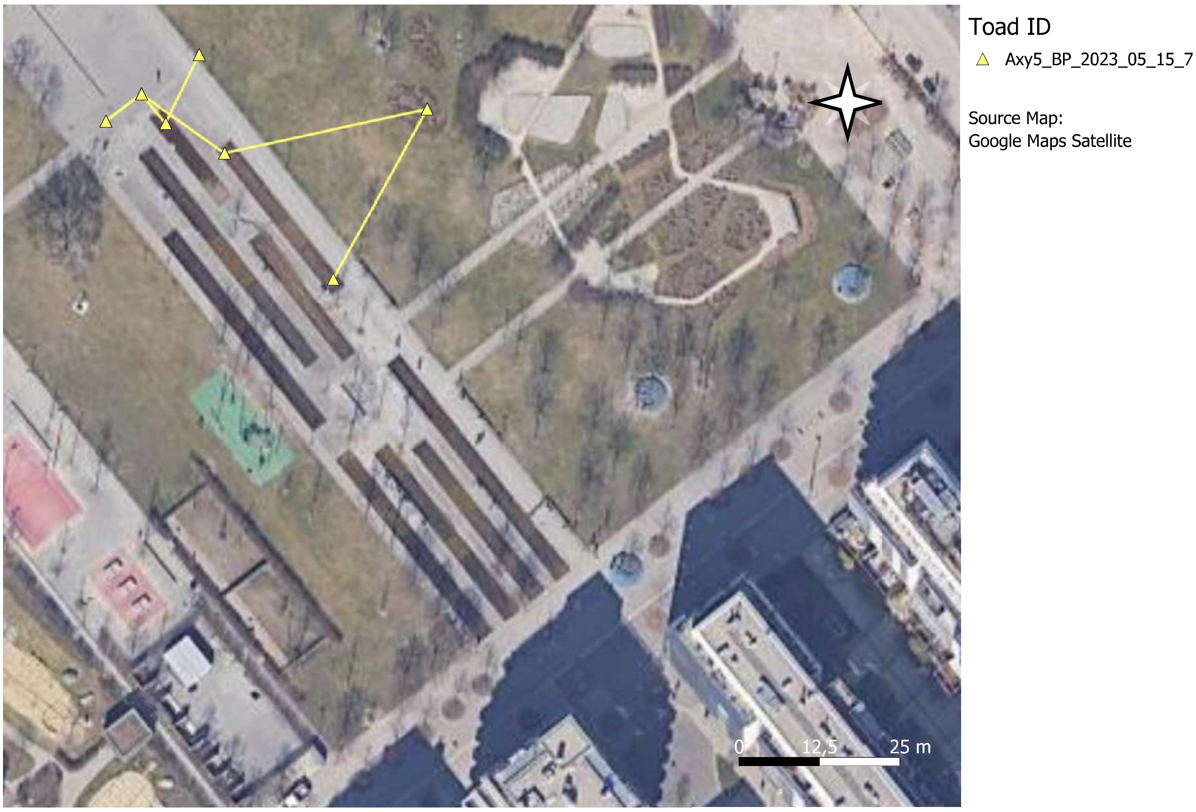

Fig. A 14: Site Rudolf-Bednar-Park: Round 1
Recorded locations of individual Axy5_BP_2023_05_15_7, connected with lines.

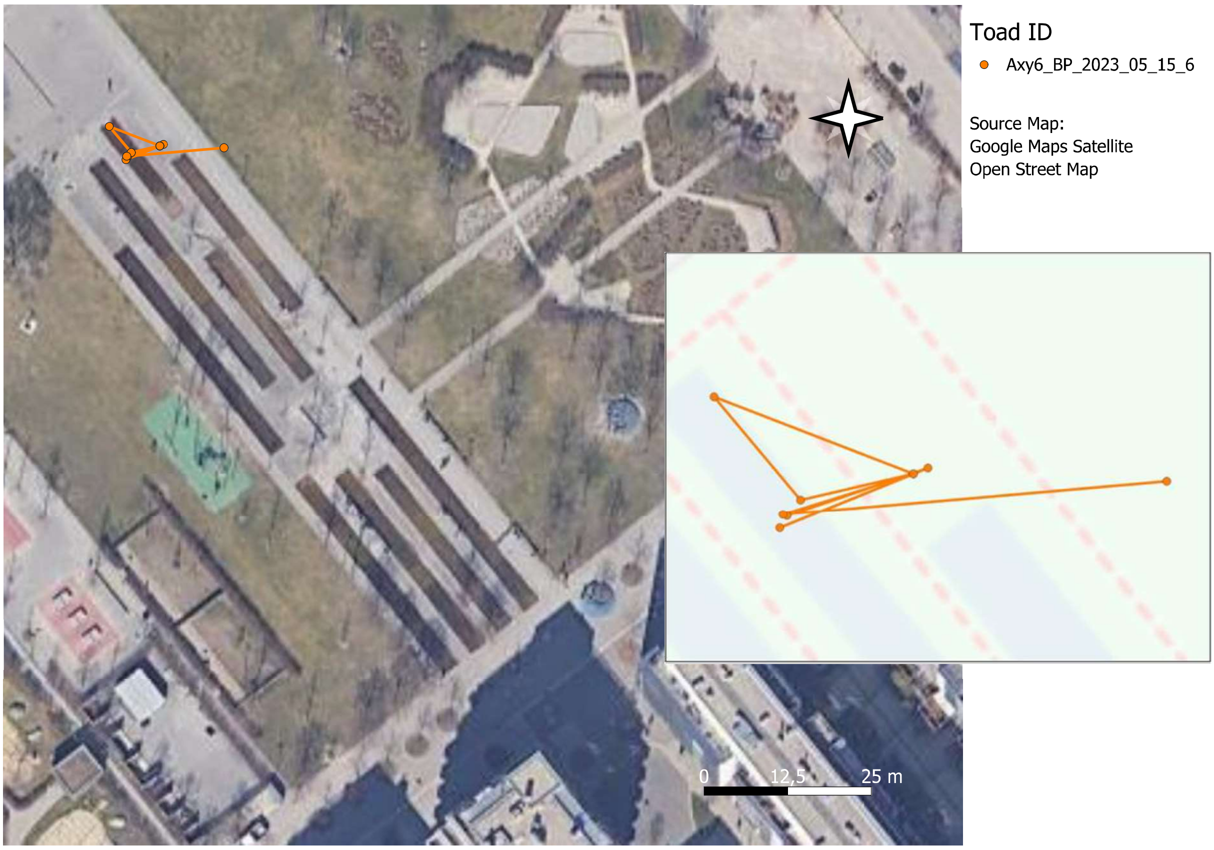

Fig. A 15: Site Rudolf-Bednar-Park: Round 1
Recorded locations of individual Axy6_BP_2023_05_15_6, connected with lines.

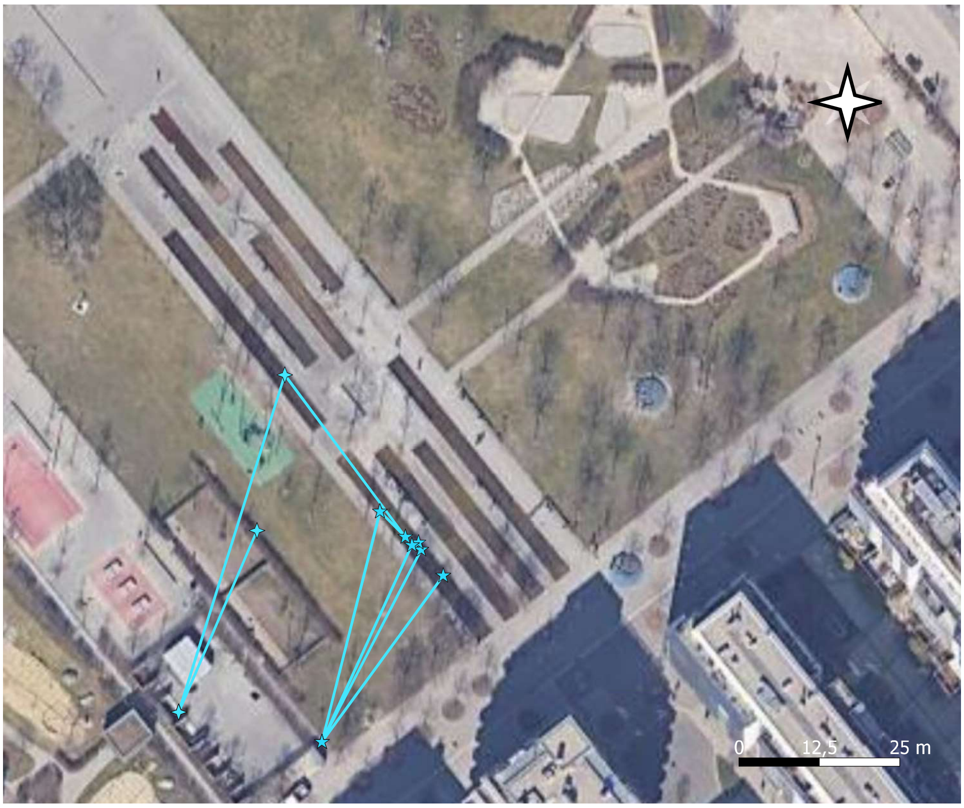

Fig. A 16: Site Rudolf-Bednar-Park: Round 1
Recorded locations of individual Axy7_BP_2023_05_16_1, connected with lines. This individual was tracked unintentionally twice, and the recorded activity ranges have been joint.

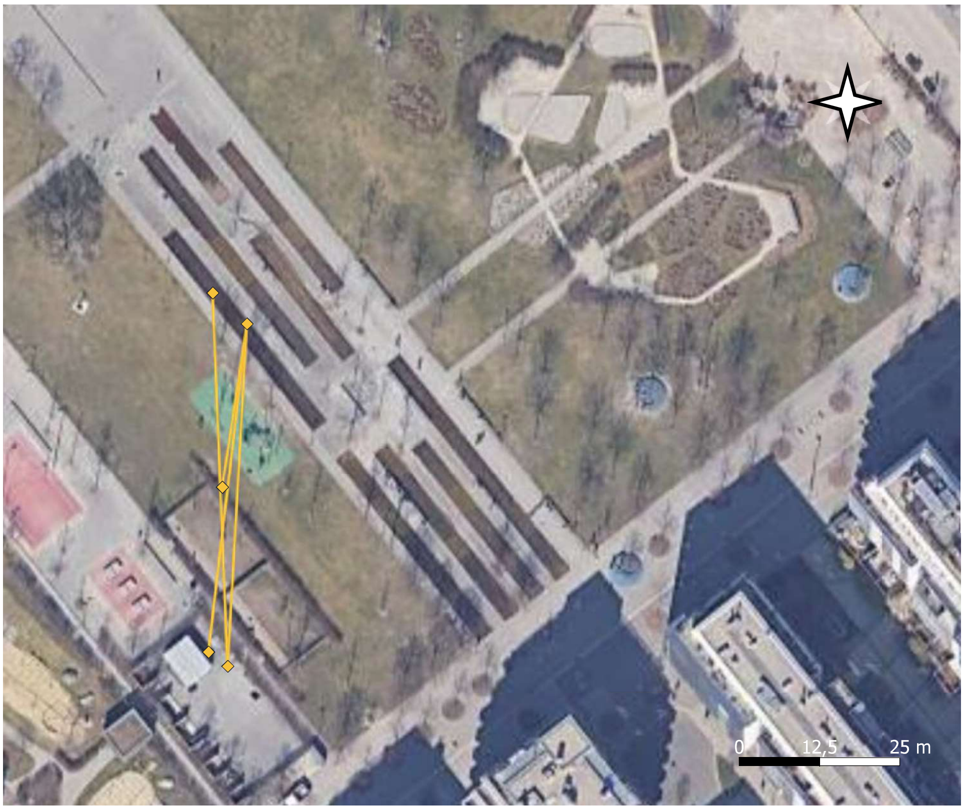

Fig. A 17: Site Rudolf-Bednar-Park: Round 2 (1st Run)
Recorded locations of individual Axy6_BP_2023_06_07_3, connected with lines.

Fig. A 18: Site Rudolf-Bednar-Park: Round 2 (2nd Run)
Recorded locations of individual Axy5_BP_2023_06_10_4, connected with lines.

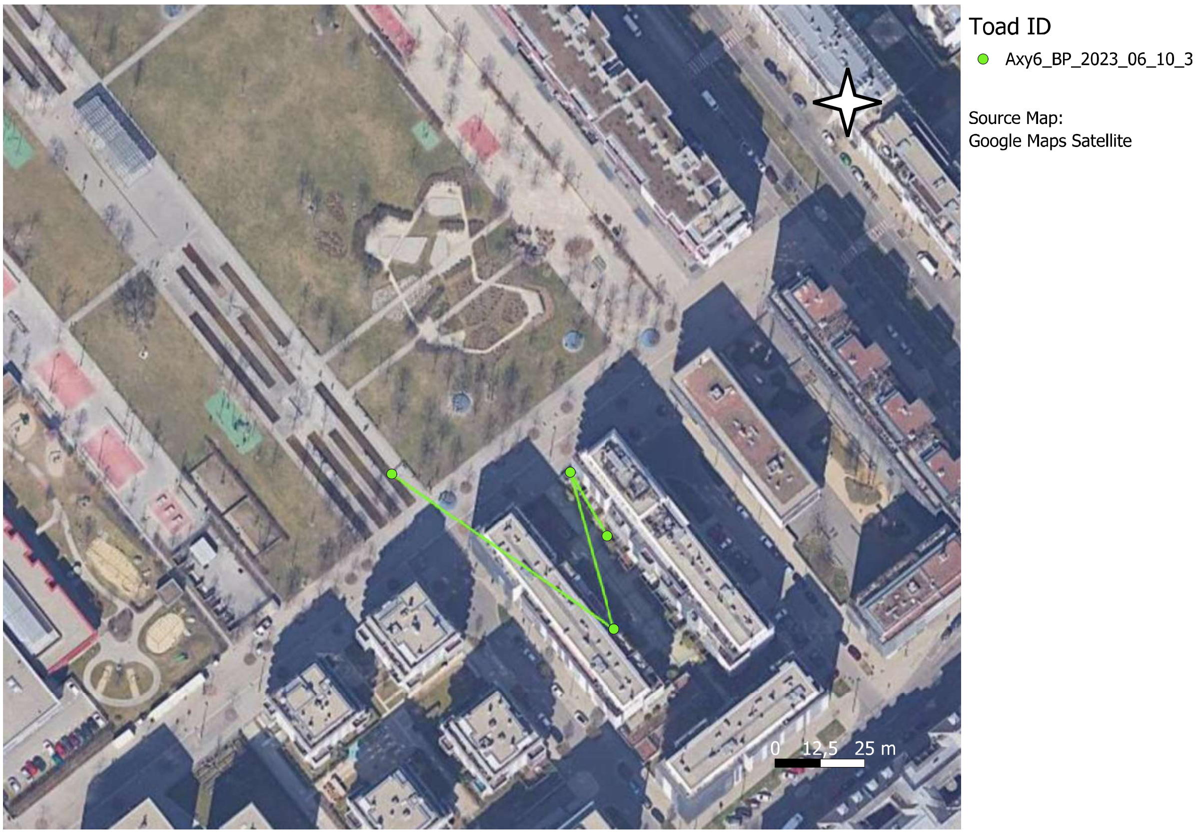

Fig. A 19: Site Rudolf-Bednar-Park: Round 2 (2nd Run)
Recorded locations of individual Axy6_BP_2023_06_10_3, connected with lines.