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Activity patterns and behavior of European green toads (*Bufo viridis*) from an urban population compared to a natural habitat

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# Abstract

Animal tracking is an established tool for exploring the movement patterns of animals. Combined with information from accelerometers, it provides an even deeper insight into the behavior of animals. The method can be used to determine when and how often certain behaviors occur. Advancements in technology now enable these methods to be applied even to small animals, such as amphibians. In our study, we equipped European green toads (*Bufo viridis*) from an urban population in Vienna and from a rural population in the Neusiedler See - Seewinkel National Park with tracking packages. Despite the challenges of the urban habitat, the toads manage to survive in the city. This observation raises the question of whether and how they have adapted to the extreme conditions of the urban environment.

Using the accelerometer data, the VeDBA (Vectorial Dynamic Body Acceleration) was calculated in order to compare the activity levels between the populations. Subsequently, an algorithm was applied to classify different behaviors (resting, calling, vertical movement, horizontal movement, jumping, feeding). These behaviors were analyzed across the daily activity cycle and statistically compared between the urban and rural populations.

Toads in the urban habitat displayed significantly lower average activity levels. This difference was also reflected in the observed behaviors. Viennese toads showed an increased proportion of resting behavior, whereas all active behaviors were observed more frequently in the rural population. Particularly striking was the significantly higher frequency of calling among the animals in the National Park. These findings suggest potential behavioral adaptations to the urban environment. The reduced activity in urban toads may represent a strategy to save energy, given the usually limited availability of prey in cities. It could also be a reaction to minimize exposure to threats in the heavily human-dominated habitat. This study provides important insights into the ecology of green toads in urban habitats, which should be considered for developing conservation guidelines by offering e. g. suitable habitats to support the survival of the endangered species in cities.

# Zusammenfassung

Das Besondern von Tieren ist ein bewährtes Instrument, um ihre Bewegungsmuster besser zu verstehen. In Kombination mit Daten von Beschleunigungssensoren können noch tiefere Einblicke in das Verhalten der Tiere gewonnen werden. Damit lässt sich analysieren, wann und wie häufig bestimmte Verhaltensweisen auftreten. Durch den technologischen Fortschritt ist es mittlerweile möglich, diese Methoden auch bei kleinen Tieren wie Amphibien anzuwenden. In unserer Studie wurden Wechselkröten (*Bufo viridis*) einer Population aus einem Park in Wien und einer ländlichen Population im Nationalpark Neusiedler See - Seewinkel mit Tracking-Systemen ausgestattet. Trotz der Herausforderungen im urbanen Lebensraum gelingt es den Kröten, in der Stadt eine stabile Population aufrechtzuerhalten. Dies wirft die Frage auf, ob und inwiefern sie sich an die extremen Bedingungen der Stadt angepasst haben.

Auf Basis von Accelerometer-Daten wurde die VeDBA (Vectorial Dynamic Body Acceleration) der einzelnen Tiere berechnet, um die Aktivitätsniveaus zwischen den beiden Populationen zu vergleichen. Zusätzlich wurde ein Algorithmus angewendet, um verschiedene Verhaltensweisen wie Ruhen, Rufen, vertikale Bewegung, horizontale Bewegung, Springen und Fressen zu analysieren. Diese Verhaltensmuster wurden im Tagesverlauf dargestellt und statistisch zwischen der städtischen und der ländlichen Population verglichen.

Unsere Ergebnisse zeigen, dass die Kröten im städtischen Lebensraum durchschnittlich deutlich weniger aktiv sind. Diese geringere Aktivität spiegelt sich auch in den beobachteten Verhaltensweisen wider. So verbringen die Wiener Kröten mehr Zeit mit Ruhen, während in der ländlichen Population alle aktiven Verhaltensweisen, insbesondere das Rufen, häufiger auftreten.

Diese Ergebnisse deuten auf mögliche Anpassungen an den urbanen Lebensraum hin. Die reduzierte Aktivität könnte eine Strategie sein, da in der Stadt das Nahrungsangebot geringer ist. Ebenso könnte es sich um eine Anpassung handeln, um Gefahren im von Menschen dominierten Lebensraum zu minimieren. Unsere Studie liefert wertvolle Erkenntnisse über die Ökologie von Wechselkröten in urbanen Lebensräumen. Sie bietet eine wichtige Grundlage für die Entwicklung von Schutz- und Managementrichtlinien wie zum Beispiel das Angebot von passenden Habitaten, welche den Fortbestand dieser gefährdeten Art in Städten unterstützen können.

# 1. Introduction

Modern technological advancements have revolutionized the field of animal ecology, providing unprecedented opportunities to study animal behavior and movement patterns in the wild (Brown et al., 2013; Green II, 2023; Kays et al., 2015). The development of electronic tracking devices has enabled researchers to monitor the movement and behavior of animals without the need for direct observation (Brown et al., 2013). These technologies make it possible to collect vast amounts of movement data, resulting in more precise insights into animal ecology (Nathan et al., 2022). The technological approach has emerged as an invaluable tool for surpassing the constraints and biases associated with observation by humans, enabling a more detailed monitoring of animal movements (Trappes, 2023).

Since the 1950s, animal tracking has emerged as an important field of study, initially utilizing tools such as very high frequency (VHF) radio transmitters and more recently incorporating satellite-based systems such as the global positioning system (GPS) (Benson, 2016). Alongside location-tracking technologies, accelerometers have become increasingly important for studying behavior (Brown et al., 2013). Tri-axial accelerometers can record the acceleration in three axes, which allows the calculation of the Vectorial Dynamic Body Acceleration (VeDBA). The data can reveal detailed insights into movement patterns and provide valuable information about the behavioral ecology (Brandes et al., 2021; Painter et al., 2016; Williams et al., 2017). For instance, Studd et al. (2021) demonstrated that combining accelerometers with custom acoustic recorders enables researchers to monitor predator behavior, including feeding events and prey selection. These methods provide critical information about diet and daily feeding habits. Additionally, activity data can be linked to metabolic rate estimates, as shown in studies analyzing energy expenditure in cane toads (*Rhinella marina*) by Halsey and White (2010) and cormorants (*Phalacrocorax carbo*) by Wilson et al. (2006).

Technological progress has led to miniaturization in tracking transmitters (Ripperger et al., 2020). Smaller and lighter transmitters have made it feasible to equip species such as Monarch butterflies (*Danaus plexippus*) and bumble bees (*Bombus terrestris audax*) with tracking devices, expanding the scope of research in animal ecology (Green II, 2023; Woodgate et al., 2016). The development of light weight transmitters includes tracking packages consisting of e. g. a combination of temperatur-logger, VHF-transmitter and accelerometer as used in this study.

Understanding the movement and behavior of species is essential for effective conservation strategies (Reed, 2002). Habitat fragmentation is one of the major threats animals face in human dominated environments, it is predicted to cause on average

nine percent of terrestrial mammal species extinctions across all eco-regions (Kuipers et al., 2021). Studies utilizing tracking technology have shown to be vital in mitigating risks to wildlife caused by habitat fragmentation by setting conservation measures adapted to the specific animal activity, such as analyzing adequate locations for wind turbines, in order to prevent bird collisions (Liechti et al., 2013; Santos et al., 2022).

Amphibians are particularly vulnerable, with over 40% of species globally classified as threatened (Luedtke et al., 2023). Habitat loss and fragmentation, especially in areas altered by human activities, are primary drivers of this decline (Cushman, 2006). Amphibian reproduction often depends on access to water bodies for breeding and tadpole development (Sinsch, 1990). However, habitat fragmentation frequently obstructs migration routes between overwintering sites and reproduction sites (Matos et al., 2019). Consequently, many individuals encounter obstacles during migration, often resulting in roadkill incidents (Hels & Buchwald, 2001).

The challenges posed by habitat fragmentation are exacerbated in urban environments. Urbanization not only diminishes available habitat but also increases isolation among amphibian populations, which further reduces genetic diversity. This phenomenon is exemplified by studies on green toad (*Bufo viridis*) populations in Košice, where urbanization has had a pronounced impact on genetic patterns, highlighting the consequences of habitat fragmentation in urban settings (Vargová, Gužiová, et al., 2023). Similarly, the common toad (*Bufo bufo*) population in Kraków has witnessed a decline of genetic diversity due to ongoing habitat changes associated with urbanization (Budzik et al., 2013). Despite these challenges, urban habitats also present opportunities for green toads. Fish-free artificial ponds often serve as valuable breeding sites, and warmer city microclimates can accelerate tadpole development, providing some compensatory benefits (Mazgajska & Mazgajski, 2020).

However, little is known about the behavioral ecology of amphibians in urban habitats (Scheffers & Paszkowski, 2012). Tracking techniques can be a helpful tool in order to investigate spatial use and activity patterns of amphibians in cities. Radio-tracking of natterjack toads (*Epidalea calamita*) in a Parisian urban park revealed limited movements around breeding sites, emphasizing the importance of habitat connectivity in urban amphibian conservation (Husté et al., 2006).

A recent study has been conducted by Spießberger et al. (2023), who used a combination of VHF transmitters and accelerometers to study urban green toads. They found that urban green toads show peak activity before midnight and have reduced home ranges. Another tracking study with green toads has been performed by Indermaur et al. (2009), who came to the conclusion that prey density and habitat structure have a major influence on home ranges of *B. viridis*. Ott (2015) analyzed their migration behavior on the Baltic island Fehmarn, finding that the toads prefer coastal, agricultural and settlement areas.

In this study, I aim to expand upon Spießberger et al. (2023) preliminary investigation by examining the activity patterns and behavioral characteristics of green toads.



Specifically, I intend to compare a population in an urban environment with one in its natural habitat to identify potential adaptations to the artificial landscape and human activity.

One special feature of *B. viridis* is its tolerance to human disturbance as it appears in various European cities, such as Warsaw, Poland (Mazgajska and Mazgajski 2020), Rome, Italy (Formichetti et al. 2003) and Vienna, Austria (Formichetti et al., 2003; Landler et al., 2023; Mazgajska & Mazgajski, 2020). Natural distribution areas can be found at the dynamic river streams Tagliamento in Italy and Vjosa in Albania, but also in pannonic steppe landscapes like in the National Park Neusiedler See/Seewinkel (Amon, C., 2022; Frank et al., 2018; Indermaur et al., 2009). Green toads are ecologically versatile, as they are not restricted to specific breeding sites and can rapidly colonize new habitats as pioneer species (Stöck et al., 2008). In cities they tend to appear suddenly at new sites, which are often disturbed areas such as construction sites (Landler et al., 2023; Sistani et al., 2021).

In their natural habitats, *B. viridis* typically exhibit large home ranges and are known for their high degree of mobility (Indermaur et al., 2009; Stöck et al., 2008). Lehofer (2024) found that activity ranges in urban habitats are smaller, whether this also affects their behavior and activity patterns throughout the day is a question that still needs to be addressed.

I hypothesize that the activity patterns of night-active green toads in urban habitats have adapted to human presence, compared to those in rural environments. Specifically, I expect their behavior to be characterized by frequent resting during the day, coupled with increased activity at night when human activity is reduced. Additionally, I anticipate a general decrease in toad activity in urban areas due to restricted habitat space and extended periods of hiding. I also predict significant behavioral differences between urban and rural toad populations, with one potential adaptation being an increased frequency of resting.

To examine variations in behavior and activity patterns, I will analyze VeDBA (Vectorial Dynamic Body Acceleration) data and three-dimensional acceleration data collected from several male individuals in both habitat types using accelerometers equipped on the toads during their breeding season.

## 2. Material and Methods

### 2.1 Study species

The European green toad (*B. viridis*) is a medium-sized amphibian species belonging to the family Bufonidae. Its distribution ranges from Europe to western Asia (IUCN, 2024). The toads are characterized by a distinctive beige coloration adorned with dark green spots, a unique pattern that allows for individual recognition (Burgstaller et al., 2020). Males are typically smaller than females, less contrastingly colored and during breeding season they develop nuptial pads and call by producing a melodic trilling sound (Glandt, 2018). Green toads are thermophilic and rely on a substrate in which they can burrow or structures where they can hide during the day, as they are primarily nocturnal. For spawning, they require small, shallow bodies of water with minimal vegetation, where they can deposit spawning strings measuring up to four meters in length (Glandt, 2018). The breeding season starts with the migration of males to potential spawning habitats. It can last from March to August, and from September onward, they seek out their overwintering grounds (Glandt, 2018). Unlike many other anuran species, European green toads are relatively salt-tolerant with tadpoles developing in waters with up to 23‰ salinity (Gordon, 1962; Schmidt, A., & Loman, J., 2019).

*Bufo viridis* is listed in Appendix IV of the European Habitat and Species Directive and is classified as endangered in the Red List of Austria (Council Directive 92/43/EEC, 2013; Umweltbundesamt, 2023). Consequently, the species is strictly protected.

### 2.2 Tracking packages

We assembled tracking packages, each consisting of an accelerometer, a temperature logger, a magnetometer (Axy 5; Technosmart Europe srl, Rome, Italy), and a VHF transmitter (with a power output of 200  $\mu$ W; Plecotus Solutions GmbH, Freiburg, Germany). These components were connected to a single rechargeable 20 mAh lithium-ion battery. The tracking packages were designed and assembled to be small and lightweight by Plecotus Solutions GmbH. The packages measured 18 mm in length, 12 mm in width, and 10 mm in height and weighed between 2.5 and 2.9 g. The VHF transmitter was connected to a titanium antenna, varying in length from 12.5 to 15.5 cm (Fig. 1). Notably, while magnetometer data was recorded, it was not analyzed in this study.

Under full charge, the device operated for approximately 12 days with our specified data sample rate (10 Hz for accelerometer, 2 Hz for magnetometer,  $\pm 8 * 9.81 \text{ m/s}^2$ , 10bit resolution) and continuous recording (every 0.1 seconds). The frequencies used for the VHF transmitter ranged from 149.997 to 150.199. Accelerometer data was stored on the device and retrieved through a USB connection after each tracking period. To fasten the tracking device to the toads, we utilized silicon tubing with a diameter of 2 mm, securing it around the toads' waist (Fig. 1).

## **2.3 Data collection**

### **2.3.1 Data collection period**

The tracking was conducted between April 24th and June 20th, during the primary breeding season of European green toads. Data collection was carried out jointly by Nadine Lehofer and myself with initial guidance from Magdalena Spießberger. It took place over four tracking periods, alternating between the two study sites.

In the Seewinkel study site, eleven male toads were equipped with tracking packages, in total 50 days and 10 hours of activity data have been analyzed from the individuals (Table 1). Tracking took place from April 24th to May 1st and from May 23rd to May 31st. At the Rudolf-Bednar-Park study site in Vienna, seven male toads were tracked from May 15th to May 21st and from June 7th to June 20th, and a total of 29 days and 8 hours of activity data were analyzed (Table 1). Sunrise ranged between 5:49 and 4:54 am (UTC+2, April 24th to June 16th) and sunset from 7:58 to 8:58 pm (ZAMG, 2024).

### **2.3.2 Recorded data**

Prior to securing the tracking packages on the toads, we measured their snout-vent length (SVL) using a caliper gauge to an accuracy of 1 mm. The dorsal pattern of the toads was photographed, and their body mass was determined using a digital scale (USA Weigh Atlanta) with an accuracy of 0.1 g. Individuals with a body mass of at least 28 g were selected to ensure that the mass of the transmitters did not exceed 10% of the toads' body mass. This criterion, which has been applied in other tracking studies with amphibians, appears to be suitable (Indermaur et al., 2008). We opted to include only male specimens in our study due to the limited presence of females at the breeding sites, with the intention of minimizing any disruption to the population caused by our research.

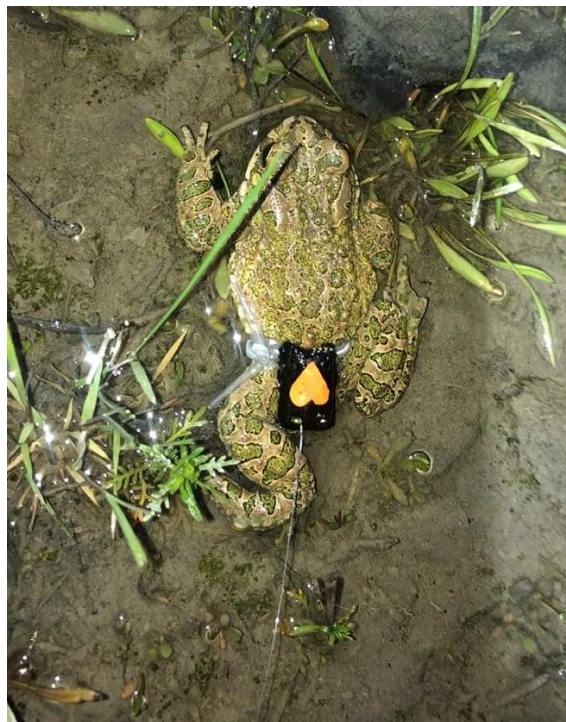
The transmitters were attached to the toads' waist using silicone tubing, secured tightly enough to prevent sliding over the hip bone, and sealed with knots. In cases where transmitters shifted to the toads' belly, they were readjusted to the back as soon as

noticed. Each toad equipped with a tracking package was closely observed until it exhibited calm and normal behavior. If any signs of discomfort due to the transmitter were observed, adjustments were made accordingly. Subsequently, the toads were released at the same location where they were found.

During tracking, toad locations were determined once during daytime (between 1 pm and 6 pm) and one to two times during nighttime (between 8 pm and 1 am). Toad locations were manually determined using VHF homing techniques, employing a handheld Yagi antenna (Lotek, operating within the 148–152 MHz frequency range) along with an RX98 receiver (from TVP Positioning AB, Lindesberg, Sweden). Final validation of toad presence relied on visual identification. Each field survey recorded the date, time, air temperature, and GPS coordinates each time a toad with transmitter was encountered.

After releasing the toads from the transmitters, we photographed and weighed them again. Additionally, we checked for bruises or skin damages caused by the transmitters.

Toads were named sequentially according to a system indicating the accelerometer number, site (SW = Seewinkel, BP = Rudolf-Bednar-Park), date of transmitter attachment, and individual number for that specific day, e. g. Axy2\_SW\_2023\_04\_24\_1.



**Fig. 1:** Toad Axy7\_SW\_2023\_04\_24\_2 equipped with tracking package Axy7, fastened with silicon tubing. Each tag has been given a specific symbol for ease of identification, in this case a heart for Axy7.

## 2.4 Study sites

#### 2.4.1 Kirchsee

The Kirchsee (47.758° N, 16.786° E) serves as a representative natural habitat for green toads in our study. It is one of approximately 59 soda pans within the Neusiedler See – Seewinkel National Park, located close to the village of Illmitz (Zimmermann-Timm & Teubner, 2021). The soda pans are shallow water bodies that heat up quickly under sunlight and periodically dry out. They are characterized by high soda concentrations and limited plant growth, which are suitable conditions for green toad to spawn. Due to ample precipitation in the spring of 2023, the soda pan was consistently filled with water during the data collection period, with its extent varying between 10 and 15 hectares of water surface. Access to the area is prohibited for people without special permission. The outer perimeter of the soda pan abuts the residential zone of Illmitz, with a moderately trafficked street located approximately 500 meters away from the soda pan's center. Natural predators observed in the area are grass snakes (*Natrix natrix*), hedgehog (*Erinaceus europaeus*) as well as birds such as white stork (*Ciconia ciconia*) and silver heron (*Ardea alba*) (pers. obs.).

#### 2.4.2 Rudolf Bednar Park

The second study site was the Rudolf-Bednar-Park (RBP, 48.226° N, 16.396° E) located in the second district of Vienna. It serves as an urban habitat for the European green toad. Surrounded by residential buildings and roads, the park is frequently visited by people, often accompanied by their dogs. It includes playgrounds, dog areas, flower beds, hedges and borders a school. The total area of the park is approximately 3.1 hectares. Within the park, there are 13 rectangular, metal water basins (Fig. 2). While one basin remains free from vegetation, the others are predominantly covered with reed patches. The basins measure approximately 2 meters in width and range from 8 to 12 meters in length, with a depth of around 10 to 15 centimeters. The pools were initially designed for aesthetic purposes; however, they are utilized by the green toad as spawning habitats. The toads face numerous dangers within the park. Observations have shown that crows prey on them, while humans, particularly children, may inadvertently harm them out of curiosity (Rienesl, 2021). Additionally, the traffic within the park can be a threat, with e-scooters and bicycles posing risks of accidental collisions with the toads. Moreover, in this human-dominated habitat, pets such as cats and dogs present significant dangers to the toad population.





**Fig. 2: Eastern part of the Rudolf Bednar Park with water basins covered with reed patches.**  
(Photo taken by Hertha Hurnaus)

## 2.5 Data analysis

We analyzed the accelerometer data utilizing X Manager software from Technosmart Europe, Italy, converting it into the DDMT file format for subsequent analysis. Data extraction was conducted using the Daily Diary Multiple Trace (DDMT) software developed by Wildbyte Technologies in Swansea, UK. During data extraction, we excluded certain segments, specifically those recorded during the initial fitting of the transmitter onto the toad and its removal at the end. Additionally, instances where the tracking package shifted to the toad's belly were also excluded. These omissions were intentional to ensure greater accuracy in the data analysis process. All subsequent data processing was carried out using the R programming language. In an earlier pilot study, green toads were raised in terrariums at the University of Natural Resources and Life Sciences, Vienna. The adult toads were then placed in outdoor enclosures, equipped with tracking devices, and filmed. By linking the recorded behaviors to specific VeDBA values, it was possible to automatically identify behaviors of free ranging toads through analyzing VeDBA data. For the automatic recognition of behaviors, Lukas Landler developed an algorithm trained with the data from the toads kept in enclosures, accurately determining about 98% of captive toad's behaviors. The statistical analysis and the creation of plots was conducted in RStudio (Version 2023.09.0+463). First, the detected behaviors were grouped for data assessment; the grouped behaviors are resting, calling, jumping, feeding, vertical movement (VM) and

horizontal movement (HM). Pie charts were created with the function ggplot to illustrate behavior distribution by location. Histograms, created with the function barplot, depict the sequential behavior frequencies. The plots illustrate the plausibility of the behavioral analysis as the histograms show the frequency of transitions between behaviors. Since data is recorded every tenth of a second, the same behavior should often follow the one detected immediately before, therefore e. g. resting should be followed by resting in most of the cases. Additionally, the VeDBA values of all individuals were averaged over ten-minute intervals and compared with behavioral frequencies by plotting the curve of VeDBA alongside specific behaviors throughout the day in one graph. Mean values were calculated for each behavior of the individual animals in order to conduct the statistical analysis. Statistical differences were assessed using the Wilcoxon test.

The tools DeepL and ChatGPT have been used to proofread and translate individual sections of this thesis. The content was checked and edited independently to ensure that it met the academic requirements and my personal writing style.

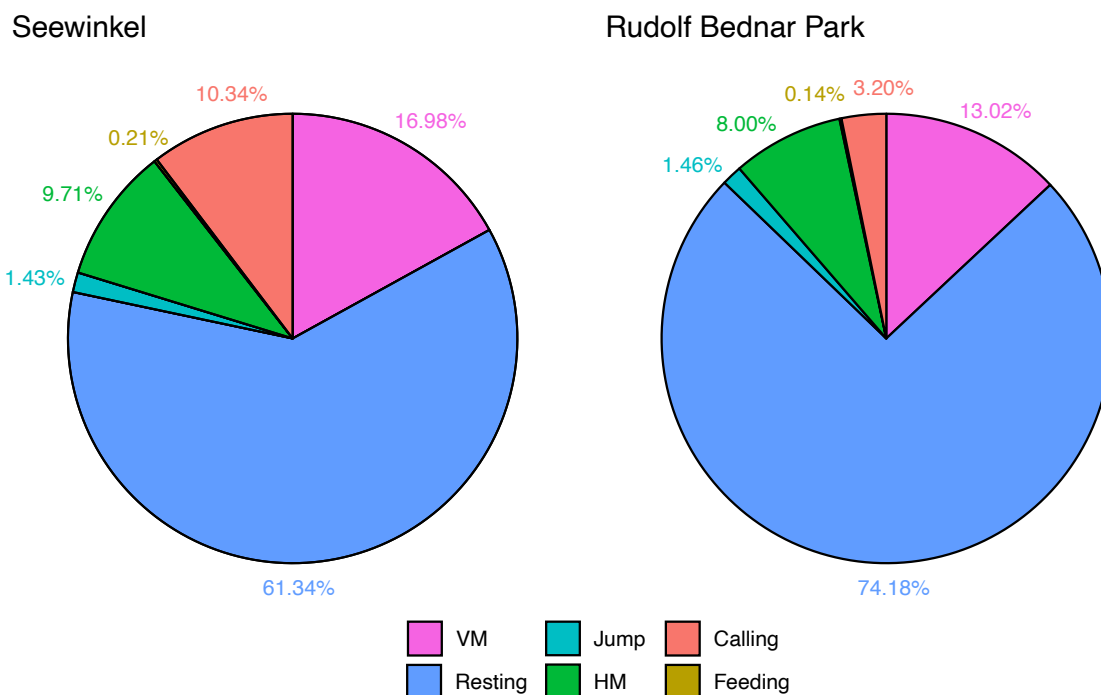
**Table 1: Toad IDs and the corresponding amount of analyzed activity data.**

| <b>ID</b>            | <b>Analyzed activity data</b> |
|----------------------|-------------------------------|
| Axy2_SW_2023_04_24_1 | 6 d, 21 h                     |
| Axy6_SW_2023_04_24_3 | 6 d, 22 h                     |
| Axy5_SW_2023_04_24_4 | 6 d                           |
| Axy1_SW_2023_04_24_5 | 6 d, 22 h                     |
| Axy4_SW_2023_04_25_2 | 4 d, 23 h                     |
| Axy2_BP_2023_05_15_5 | 5 d, 5 h                      |
| Axy6_BP_2023_05_15_6 | 3 d, 6 h                      |
| Axy5_BP_2023_05_15_7 | 4 d, 6 h                      |
| Axy2_SW_2023_05_23_1 | 3 d, 17 h                     |
| Axy6_SW_2023_05_23_2 | 3 d, 18 h                     |
| Axy5_SW_2023_05_23_3 | 2 d, 23 h                     |
| Axy2_SW_2023_05_27_1 | 3 d, 23 h                     |
| Axy5_SW_2023_05_27_2 | 2 d, 23 h                     |
| Axy6_SW_2023_05_27_3 | 1 d, 10 h                     |
| Axy6_BP_2023_06_07_3 | 2 d, 20 h                     |
| Axy5_BP_2023_06_07_4 | 3 d                           |
| Axy6_BP_2023_06_10_3 | 9 d, 20 h                     |
| Axy5_BP_2023_06_10_4 | 1 d, 23 h                     |

### 3. Results

#### 3.1 Comparison of behavior distribution between the urban and rural habitat

At both study sites the dominant behavior was resting, followed by vertical and horizontal movement and calling. Feeding and jumping behavior was rarely detected in the natural habitat as well as in the urban habitat. However, at the Rudolf Bednar Park, the proportion of resting is approximately 13% higher, resulting in a lower percentage of active behaviors overall. Particularly noteworthy is the strikingly lower proportion of calling, which is roughly three times lower compared to the natural habitat (Fig. 3).



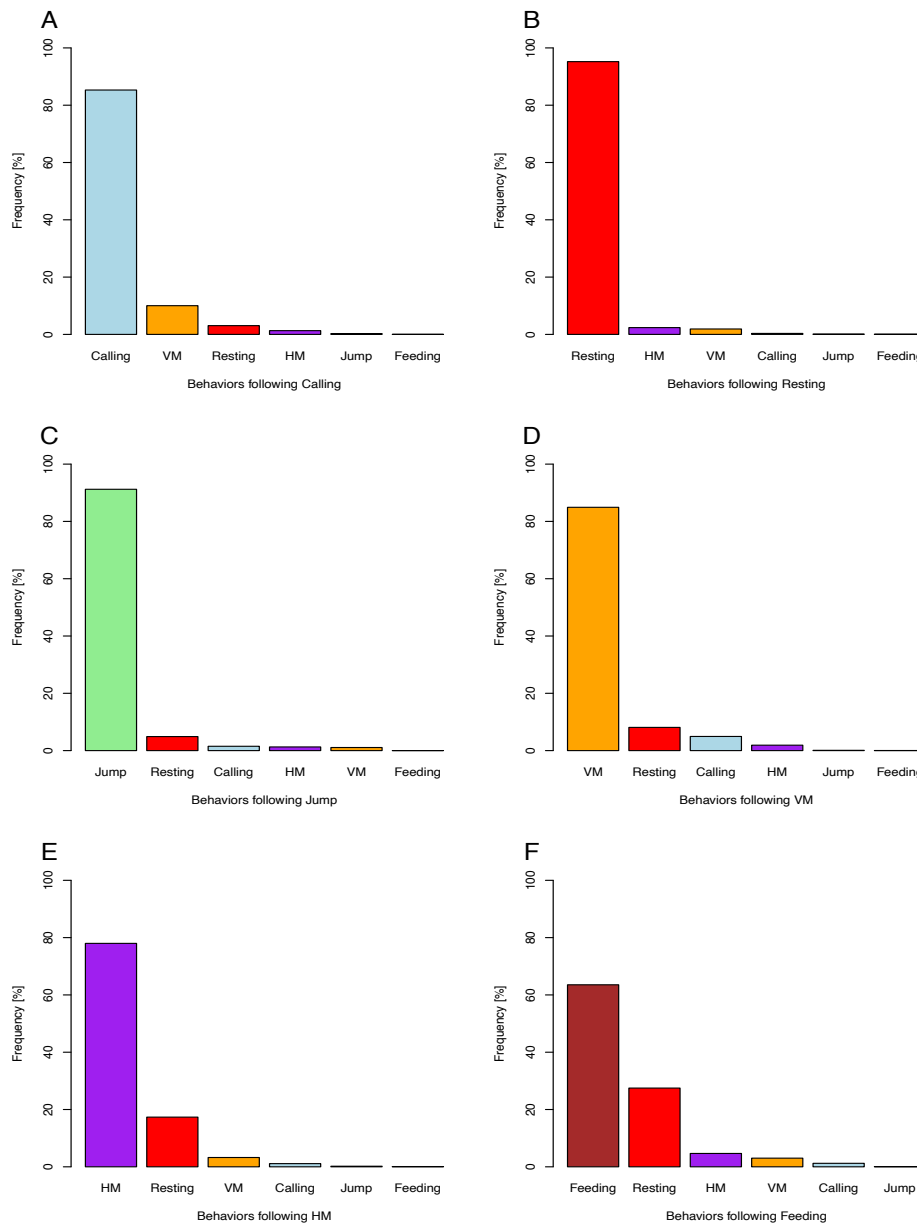
**Fig. 3:** Pie charts depicting the distribution of behaviors among green toad populations in Rudolf Bednar Park (7 individuals) and Seewinkel (11 individuals), illustrating behavior frequencies as percentages. Based on the analysis of all data collected from both sites. VM = Vertical movement, HM = Horizontal movement.



### 3.2 Sequential behavior frequencies

The specific behaviors show a high chronological consistency (e. g. jumping behavior is followed by jumping in about 90% of the cases, see Fig. 4). These data are crucial for testing the plausibility of behavior assignment to the data, which were determined at intervals of 0.1 s.

Resting shows the highest chronological consistency, with about 95% of instances followed by more resting. Feeding, on the other hand, shows the lowest consistency, with around 65% of feeding episodes followed by more feeding. Additionally, calling is succeeded by more calling approximately 85% of the time. Jumping behavior shows consistency in about 90% of instances, while vertical and horizontal movements follow each other with frequencies of approximately 85% and 80%, respectively (Fig. 4).



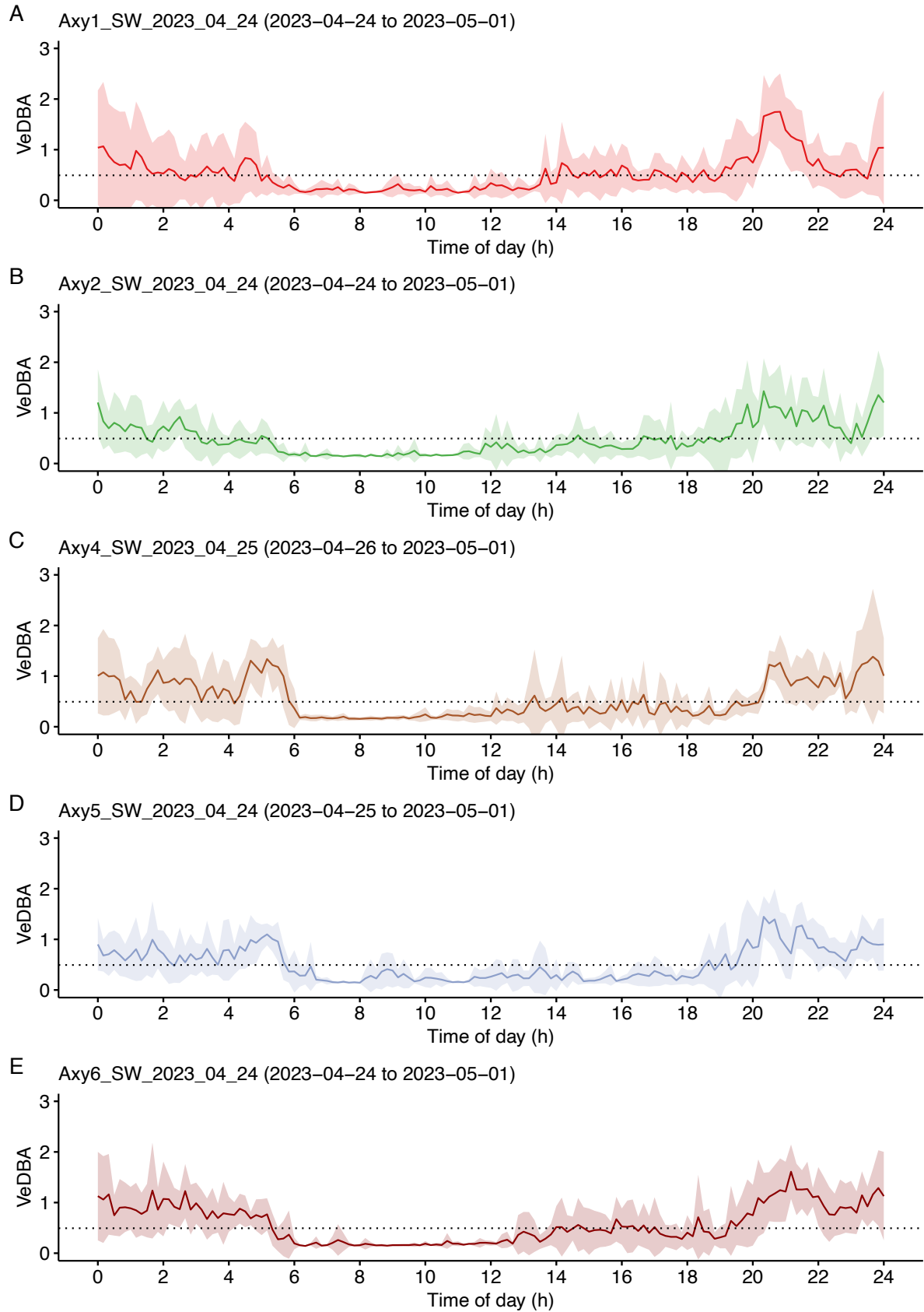
**Fig. 4: Frequencies of behaviors following to Calling (A), Resting (B), Jump (C), Vertical Movement (D), Horizontal Movement (E) and Feeding (F) as percentages.**

### 3.3 Activity patterns throughout the day

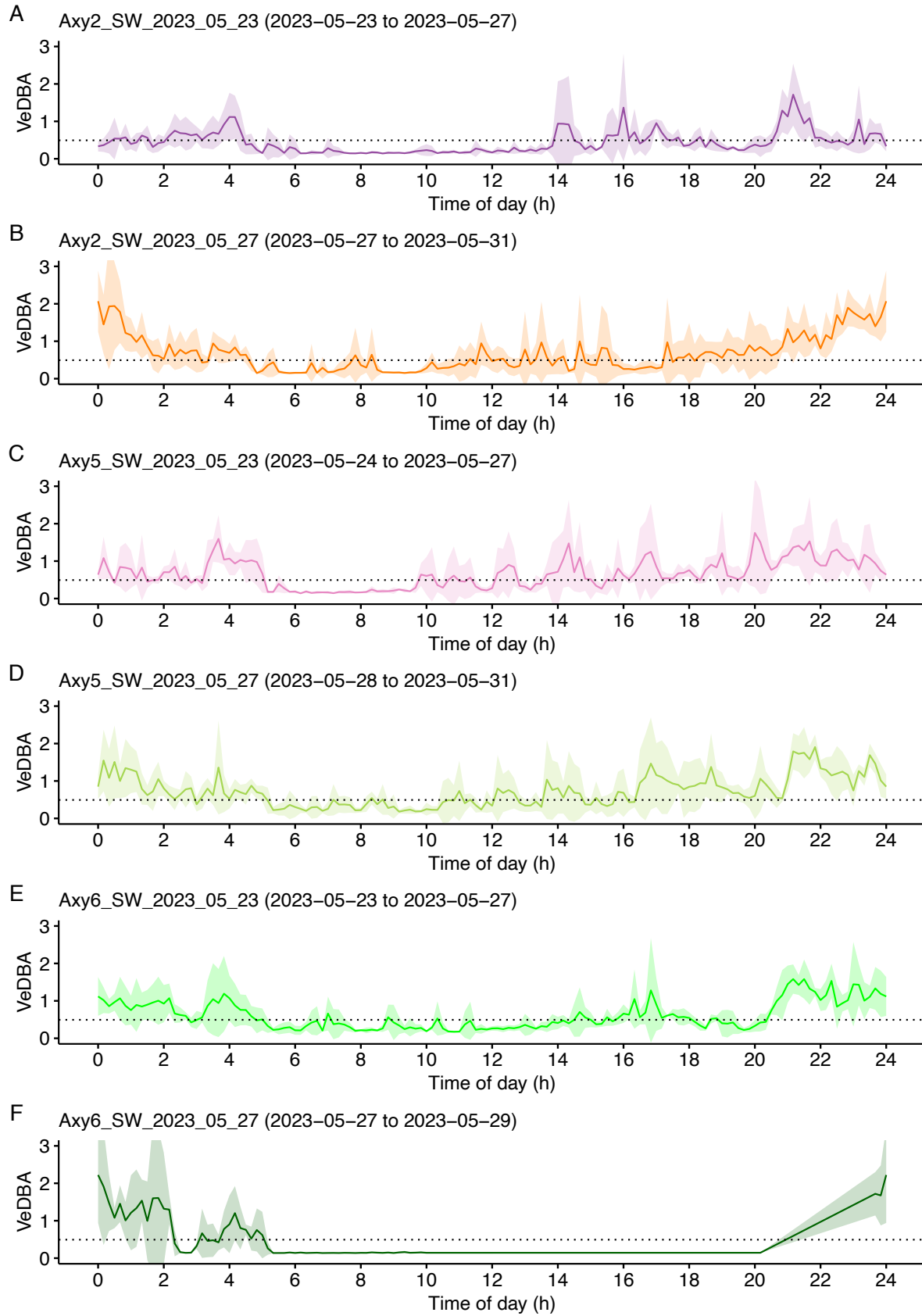
#### 3.3.1 Vectorial Dynamic Body Acceleration

The VeDBA is the sum of body acceleration in all three dimensions and represents the activity level of the toads. The average VeDBA across all 18 toads in the study was 0.49. The activity plots indicate that the individuals from both sites were typically more active during the night than during daytime, consistent with the nocturnal behavior characteristic of *B. viridis* (Fig. 5 to Fig. 8). However, it is striking that toads in the natural habitat had higher daytime activity levels than in the urban habitat. Particularly during the second tracking period in Seewinkel, activity levels during the afternoon were relatively high (Fig. 6). Daytime activity among individuals in Rudolf Bednar Park was generally low, but it is marked by occasional small fluctuations throughout the day (Fig. 7, Fig. 8). In the natural habitat activity peaks occurred mainly between 8 and 12 pm and decreased at about 6 am, toads were keeping a very low level of activity until about 12 am (Fig. 5, Fig. 6). The toads of the urban population showed high activity levels from approximately 8 pm to 6 am (Fig. 7, Fig. 8).

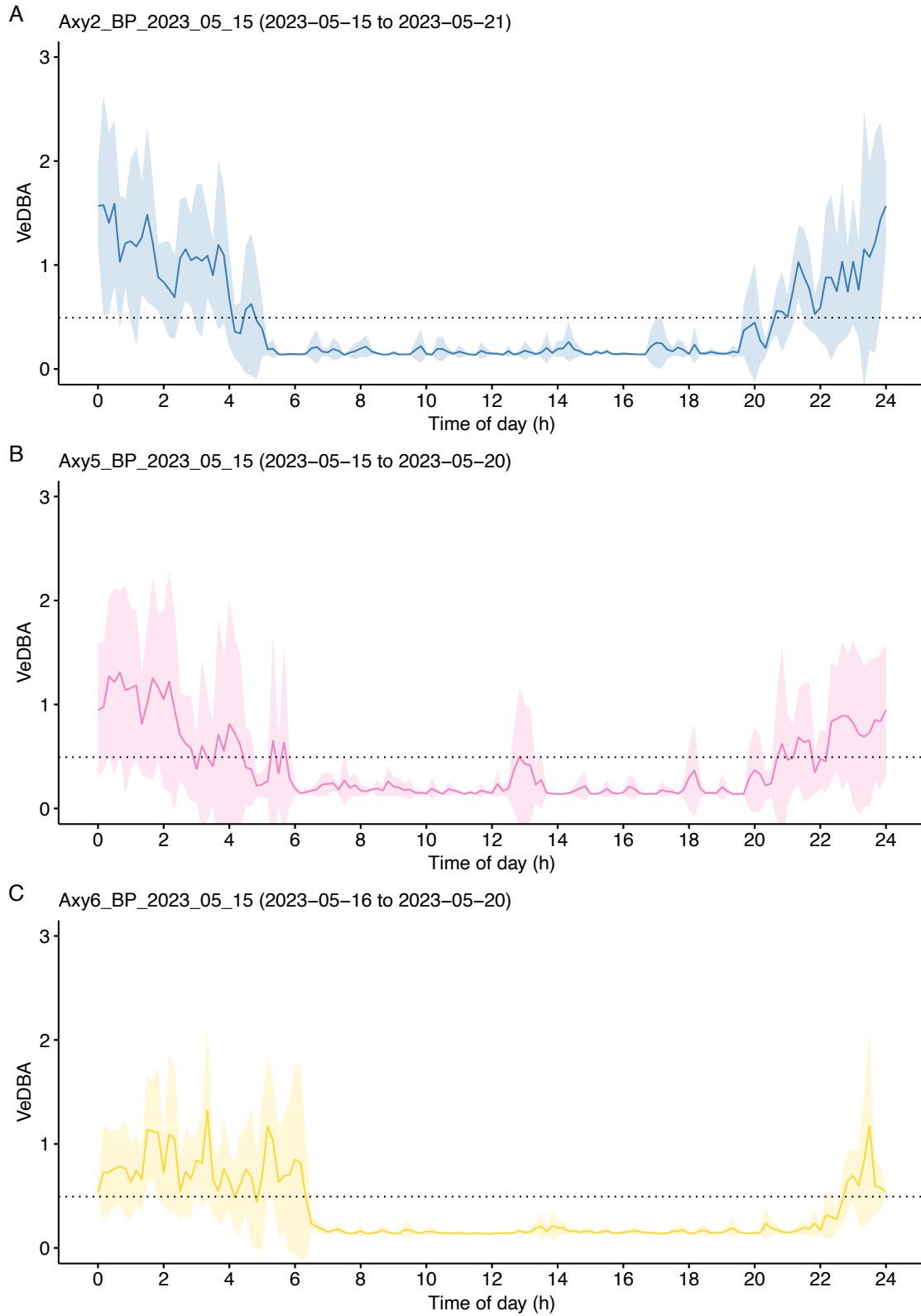
In the RBP, the trend of increasing activity levels in the early night remained consistent across both tracking periods. However, Axy6\_BP\_2023\_06\_10 stood out with consistently low activity levels throughout the day (Fig. 8). Overall, there were notable individual variations, particularly within the urban habitat. For instance, when comparing Axy6\_BP\_2023\_06\_10 to Axy6\_BP\_2023\_06\_07, the overall activity level of Axy6\_BP\_2023\_06\_10 was considerably lower. In contrast to the other toads in the urban population, Axy6\_BP\_2023\_06\_07 exhibited earlier activity, starting at approximately 6 pm (Fig. 8). When comparing the different tracking periods at the Seewinkel study site, it is remarkable that high activity levels were less concentrated during the night in the second tracking period. Activity was fairly evenly distributed throughout the day, whereas during the first period, it was more concentrated at night (Fig. 5, Fig. 6). The pattern of Axy6\_SW\_2023\_05\_27 (Fig. 6) was caused by a lack of data. From 10 am onwards, no reference data is available (Table 1).



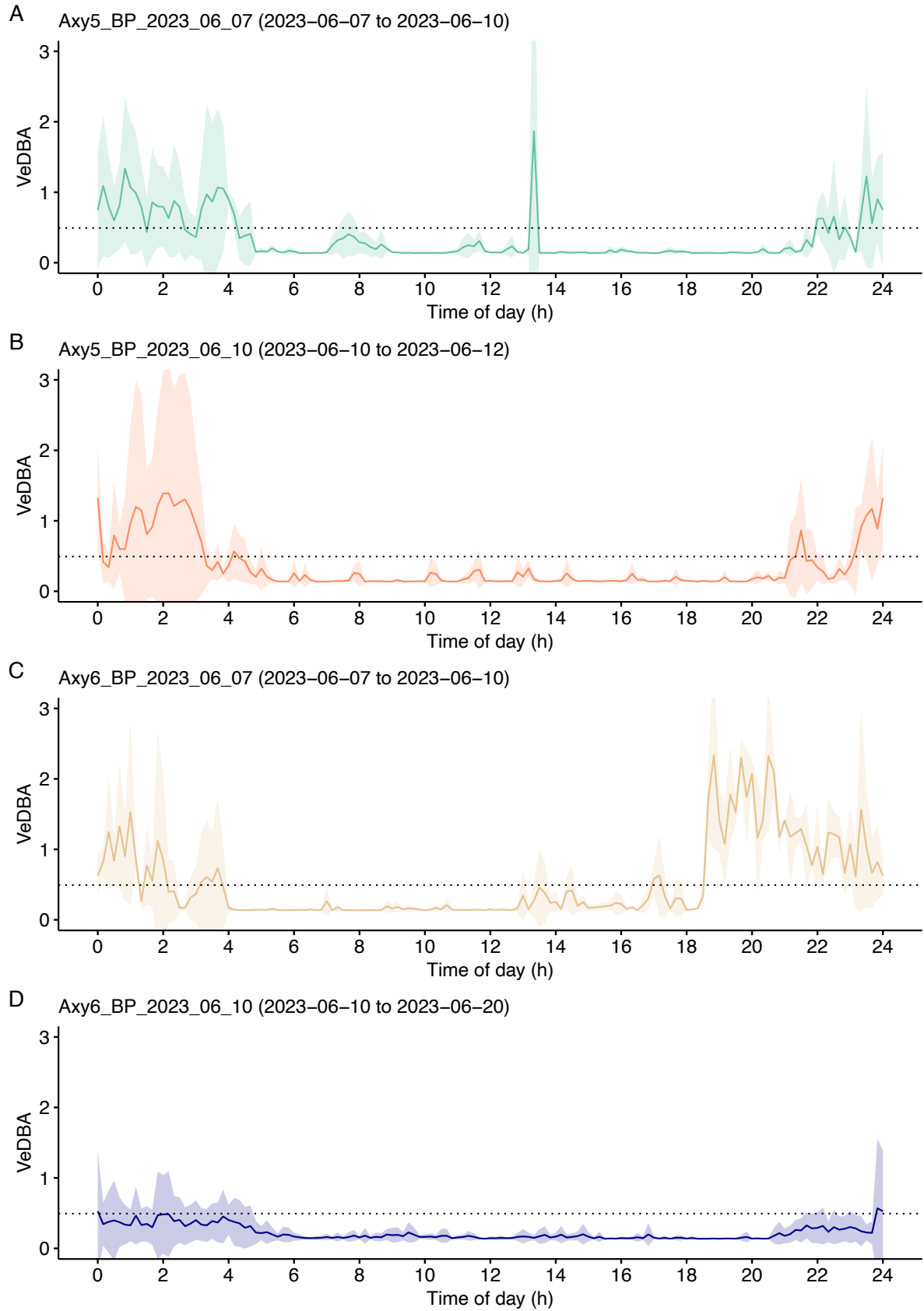
**Fig. 5: Activity patterns during the first tracking period in the Seewinkel.** Each plot (A-E) represents one individual (ID and tracking date range on the top). The colored line illustrates the average daily VeDBA, while the shaded area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



**Fig. 6: Activity patterns during the second tracking period in the Seewinkel.** Each plot (A-F) represents one individual (ID and tracking date range on the top). The colored line illustrates the average daily VeDBA, while the shaded area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



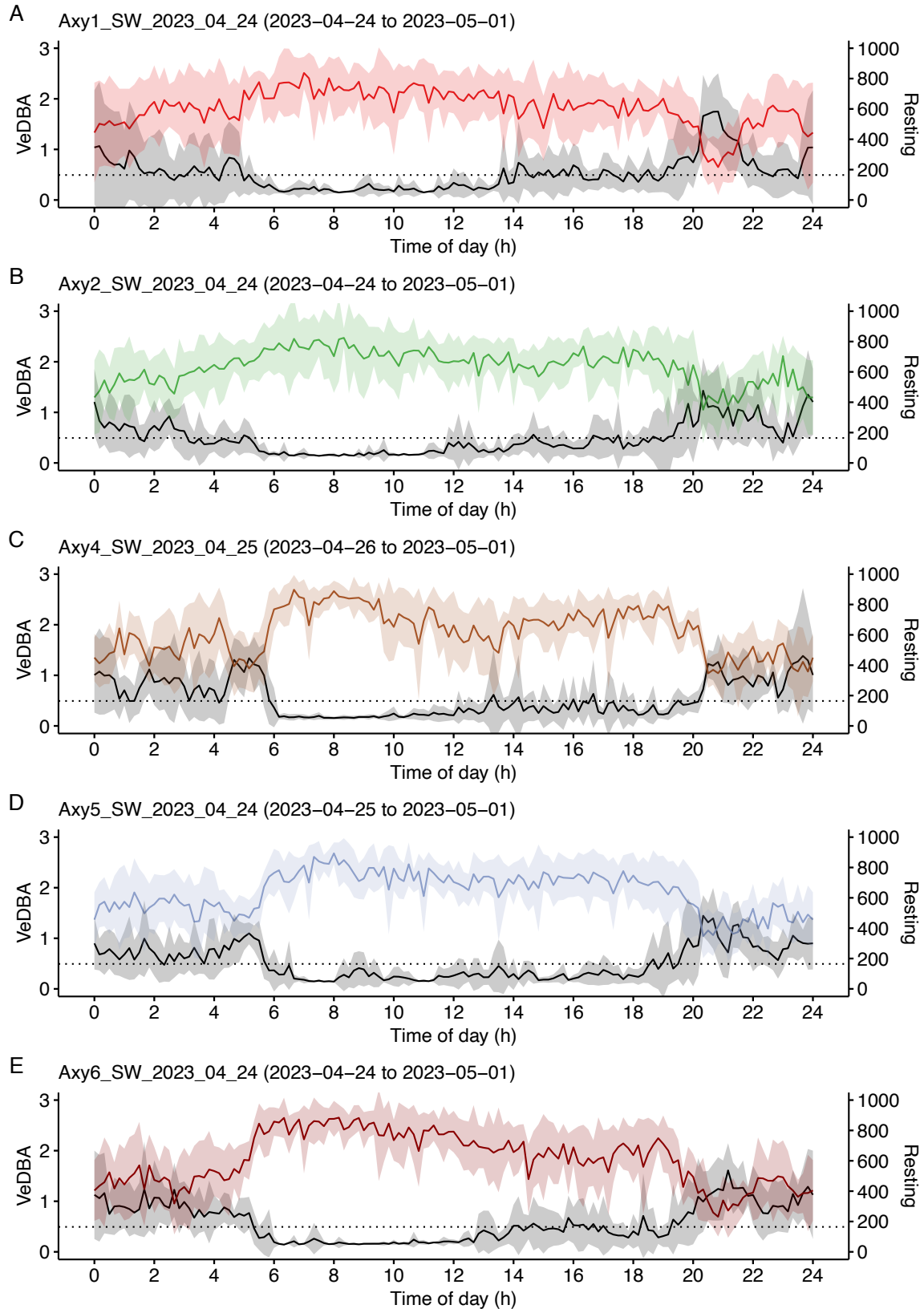
**Fig. 7: Activity patterns during the first tracking period in the RBP.** Each plot (A-C) represents one individual (ID and tracking date range on the top). The colored line illustrates the average daily VeDBA, while the shaded area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



**Fig. 8: Activity patterns during the second tracking period in the RBP.** Each plot (A-D) represents one individual (ID and tracking date range on the top). The colored line illustrates the average daily VeDBA, while the shaded area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

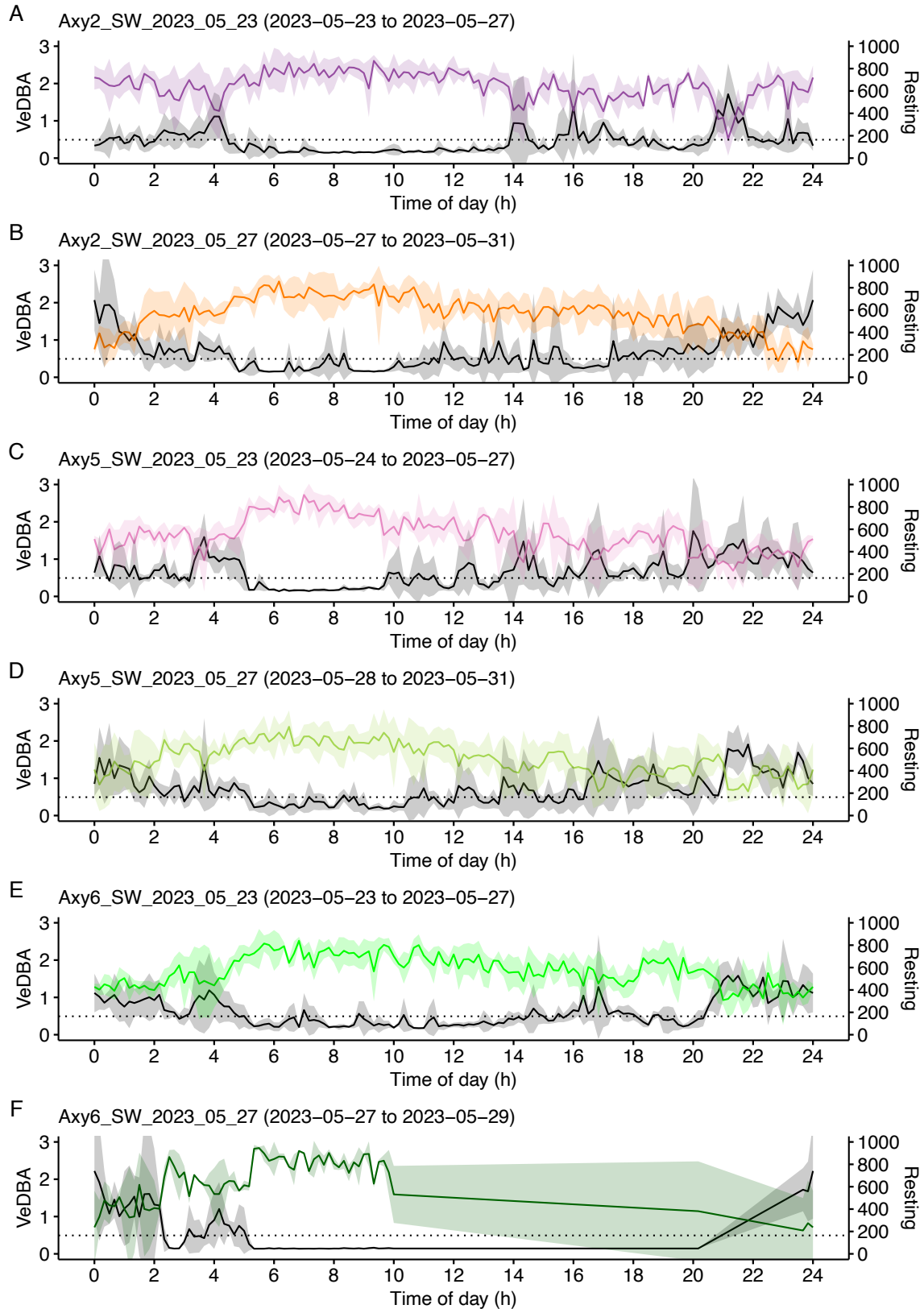
### 3.3.2 Resting

Resting behavior mirrored the overall activity levels at both study sites. Examples of this reversed pattern are observed in all toads, but one notable point is the activity peak in the individual Axy1\_SW\_2023\_04\_24 at around 9 pm, where resting sharply decreased while VeDBA increased (Fig. 9, Plot A). During the day, resting levels were high and decreased for most individuals at around 8 pm, reaching their peak between 6 and 10 am, indicating a consistent resting phase in the Seewinkel (Fig. 9, Fig. 10). In the Rudolf Bednar Park, resting levels were consistently high for an extended period during the day, spanning approximately 14 hours from 6 am to 8 pm (Fig. 11, Fig. 12). Notably, the resting level of toad Axy6\_BP\_2023\_06\_10 was comparatively high in general (Fig. 12).

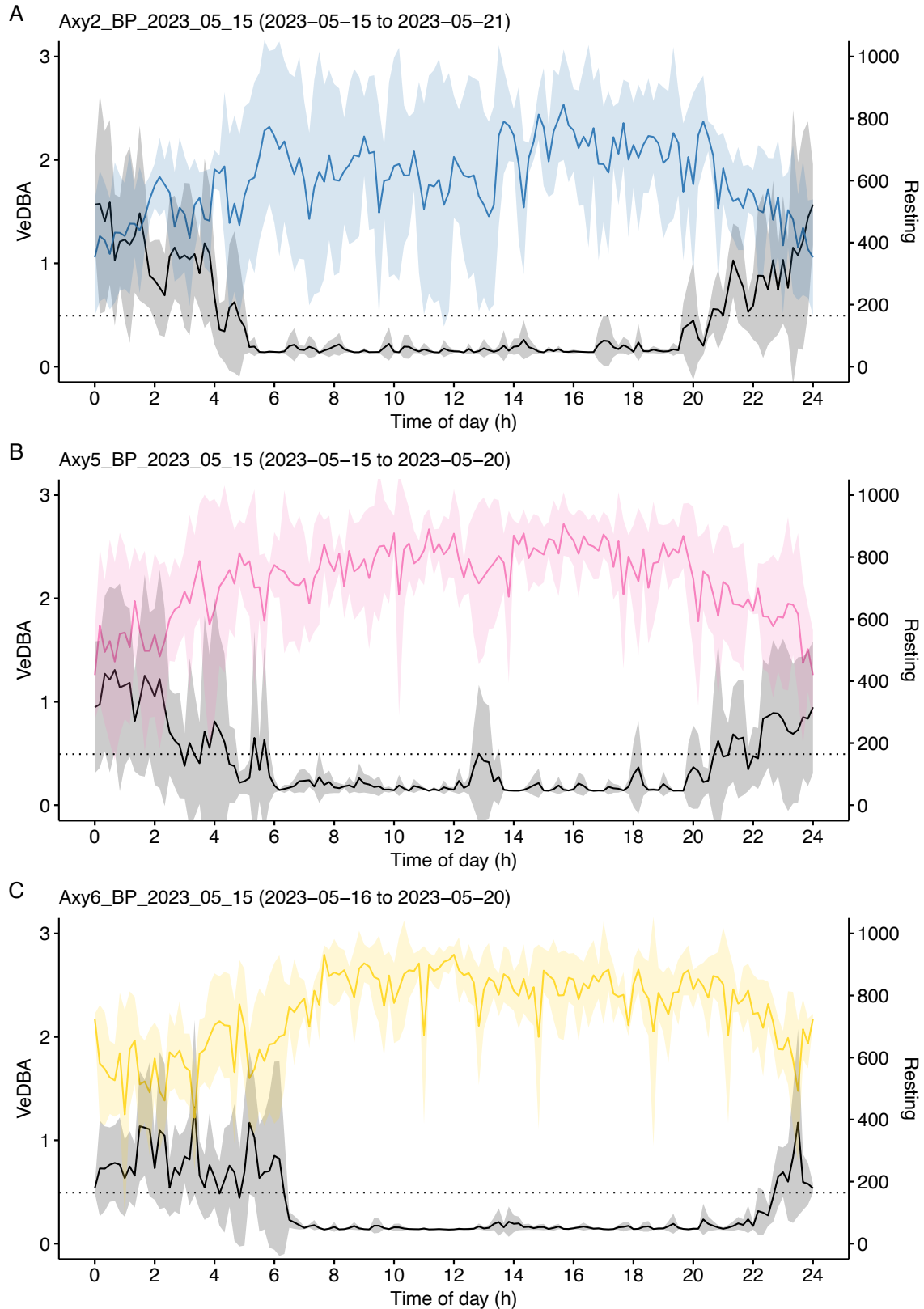


**Fig. 9: Resting and activity patterns during the first tracking period in the Seewinkel.** Each plot (A-E) shows one individual (ID and tracking date range on the top). The colored line represents the average daily resting frequency, which was calculated by averaging resting values, obtained by summing up each resting behavior detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of resting frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

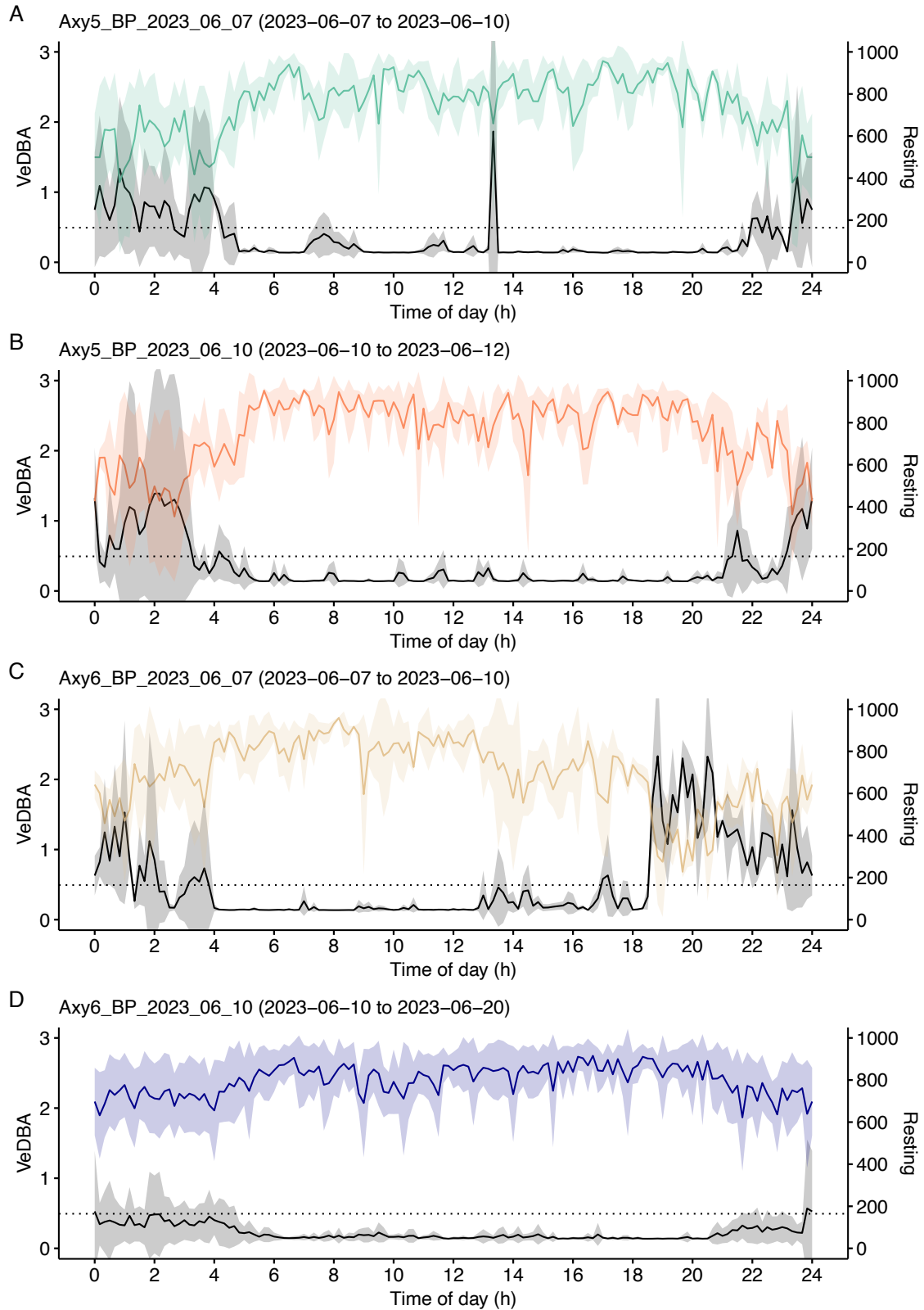




**Fig. 10: Resting and activity patterns during the second tracking period in the Seewinkel.** Each plot (A-F) shows one individual (ID and tracking date range on the top). The colored line represents the average daily resting frequency, which was calculated by averaging resting values, obtained by summing up each resting behavior detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of resting frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



**Fig. 11: Resting and activity patterns during the first tracking period in the RBP.** Each plot (A-C) shows one individual (ID and tracking date range on the top). The colored line represents the average daily resting frequency, which was calculated by averaging resting values, obtained by summing up each resting behavior detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of resting frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

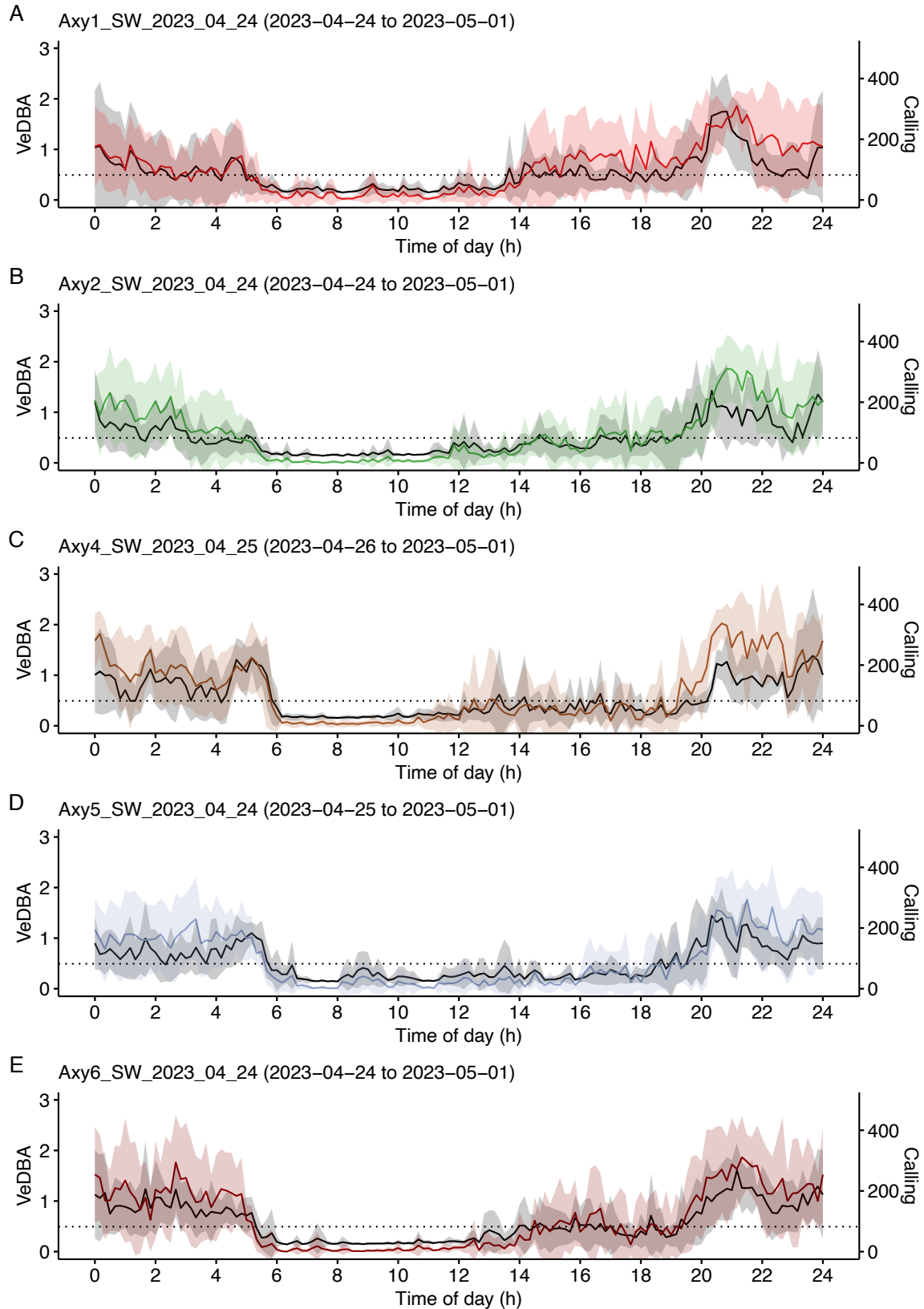


**Fig. 12: Resting and activity patterns during the second tracking period in the RBP.** Each plot (A-D) shows one individual (ID and tracking date range on the top). The colored line represents the average daily resting frequency, which was calculated by averaging resting values, obtained by summing up each resting behavior detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of resting frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

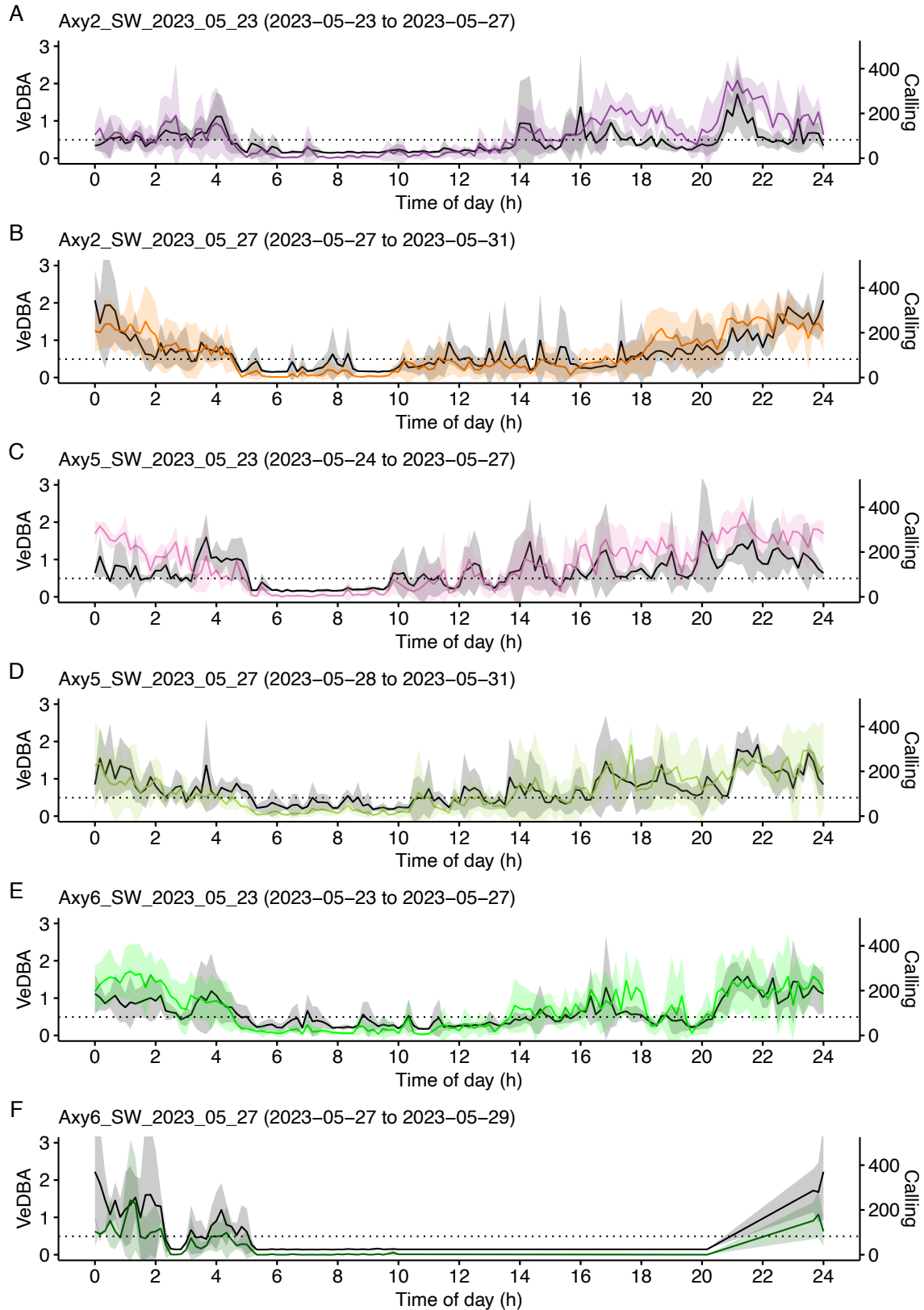
### 3.3.3 Calling

Calling patterns roughly mirrored the activity levels throughout the day, with higher frequencies typically observed during nighttime, often peaking concurrently with overall activity (Fig. 13 - Fig. 16).

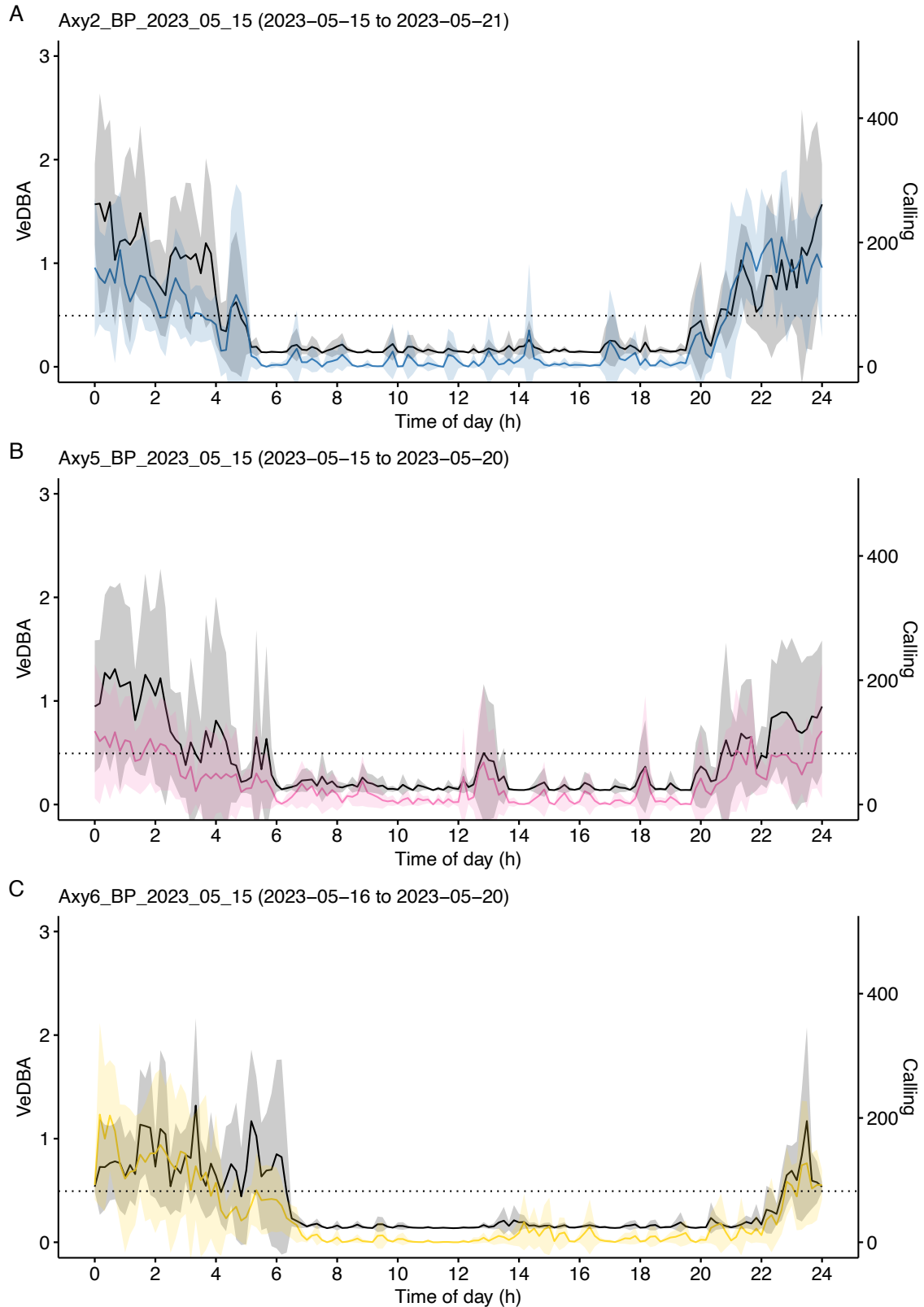
When comparing the first and second tracking periods in the Seewinkel study site, it is noteworthy that during the second period, calling activity lacked clear concentration at specific times. While there was a pronounced increase in calling around 8 pm during the first tracking period, calling activity in the second period fluctuates between midday and 5 am, without a distinct peak (Fig. 13, Fig. 14). Moreover, a comparison between the study sites revealed that calling transitioned relatively quickly from low to high levels from day to nighttime in the RBP (Fig. 15, Fig. 16). In contrast, in the Seewinkel, calling also occurred frequently during the day, particularly in the afternoon (Fig. 13, Fig. 14).



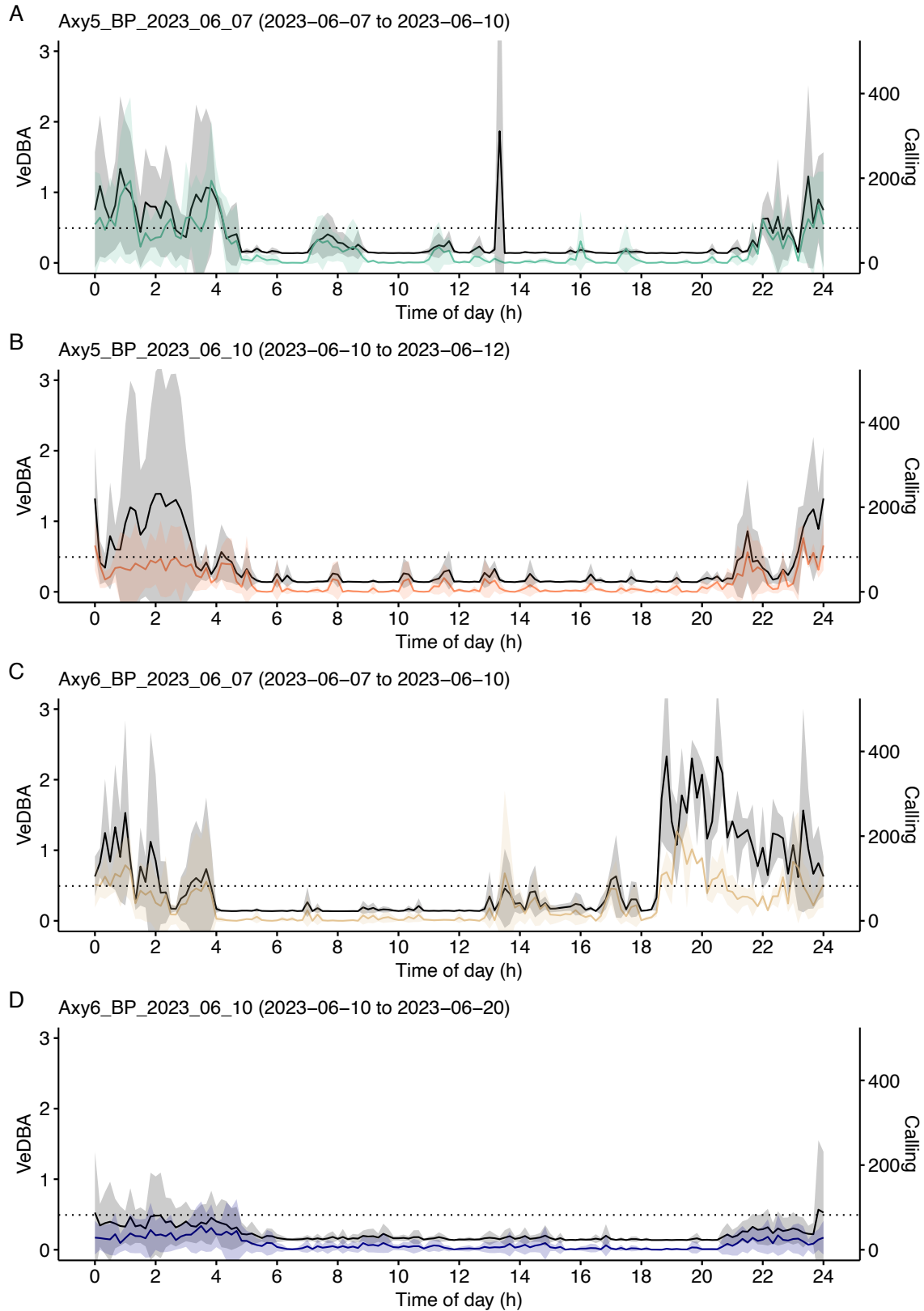
**Fig. 13: Calling and activity patterns during the first tracking period in the Seewinkel.** Each plot (A-E) shows one individual (ID and tracking date range on the top). The colored line represents the average daily calling frequency, which was calculated by averaging calling values, obtained by summing up each calling behavior detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of resting frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



**Fig. 14: Calling and activity patterns during the second tracking period in the Seewinkel.** Each plot (A-F) shows one individual (ID and tracking date range on the top). The colored line represents the average daily calling frequency, which was calculated by averaging calling values, obtained by summing up each calling behavior detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of resting frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



**Fig. 15: Calling and activity patterns during the first tracking period in the RBP.** Each plot (A-C) shows one individual (ID and tracking date range on the top). The colored line represents the average daily calling frequency, which was calculated by averaging calling values, obtained by summing up each calling behavior detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of resting frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



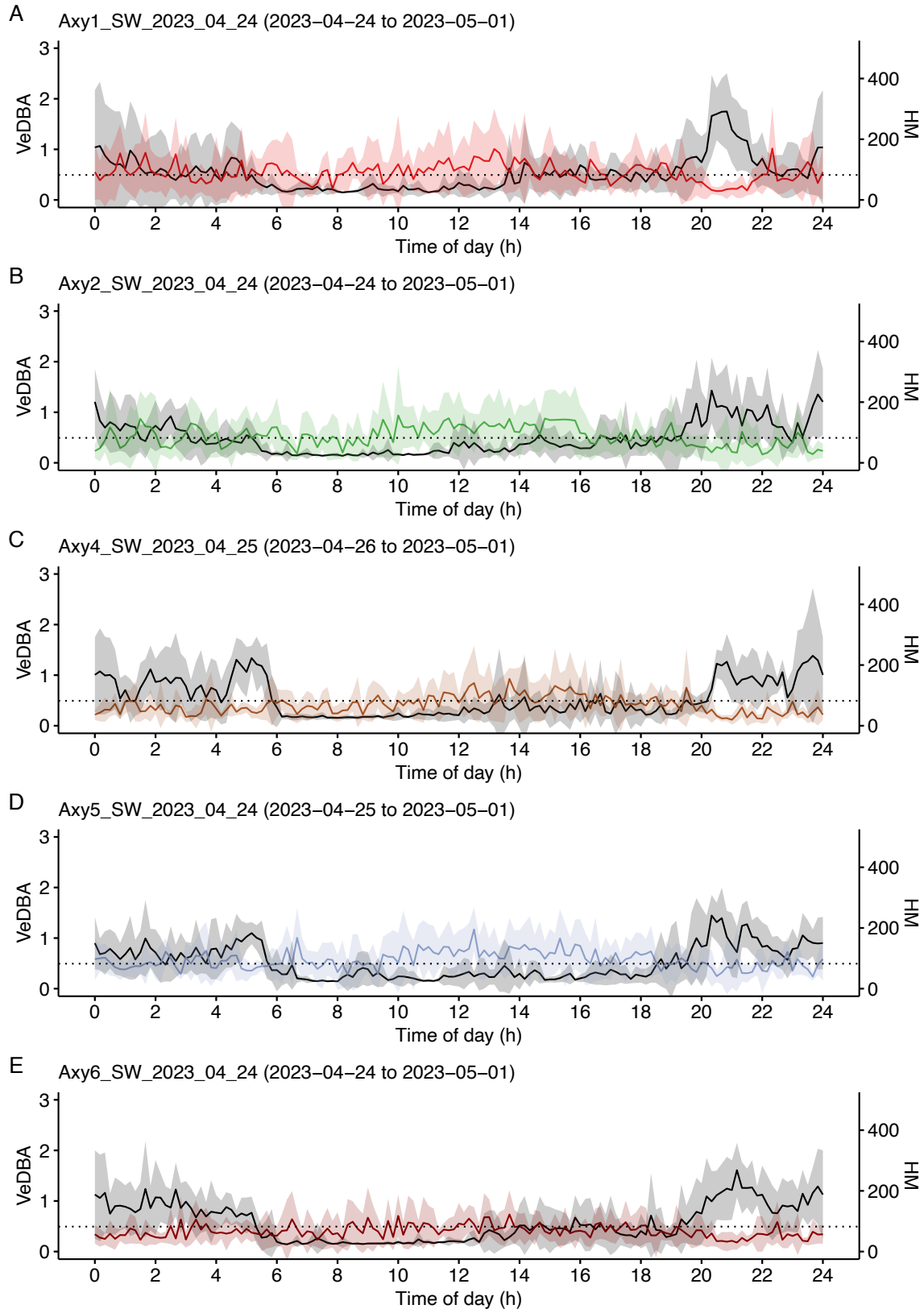
**Fig. 16: Calling and activity patterns during the second tracking period in the RBP.**

Each plot (A-D) shows one individual (ID and tracking date range on the top). The colored line represents the average daily calling frequency, which was calculated by averaging calling values, obtained by summing up each calling behavior detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of resting frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

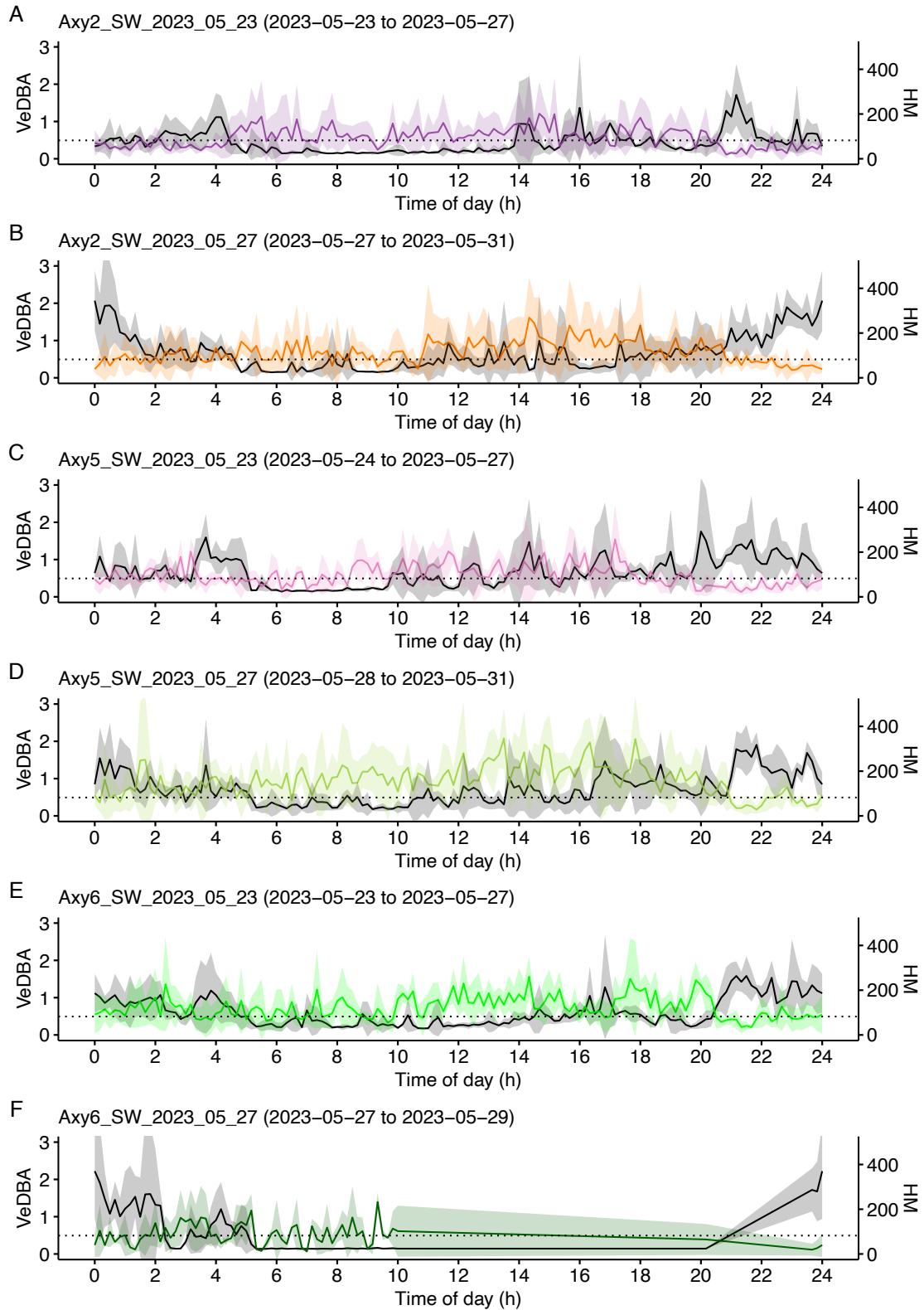


#### 3.3.4 Horizontal Movement

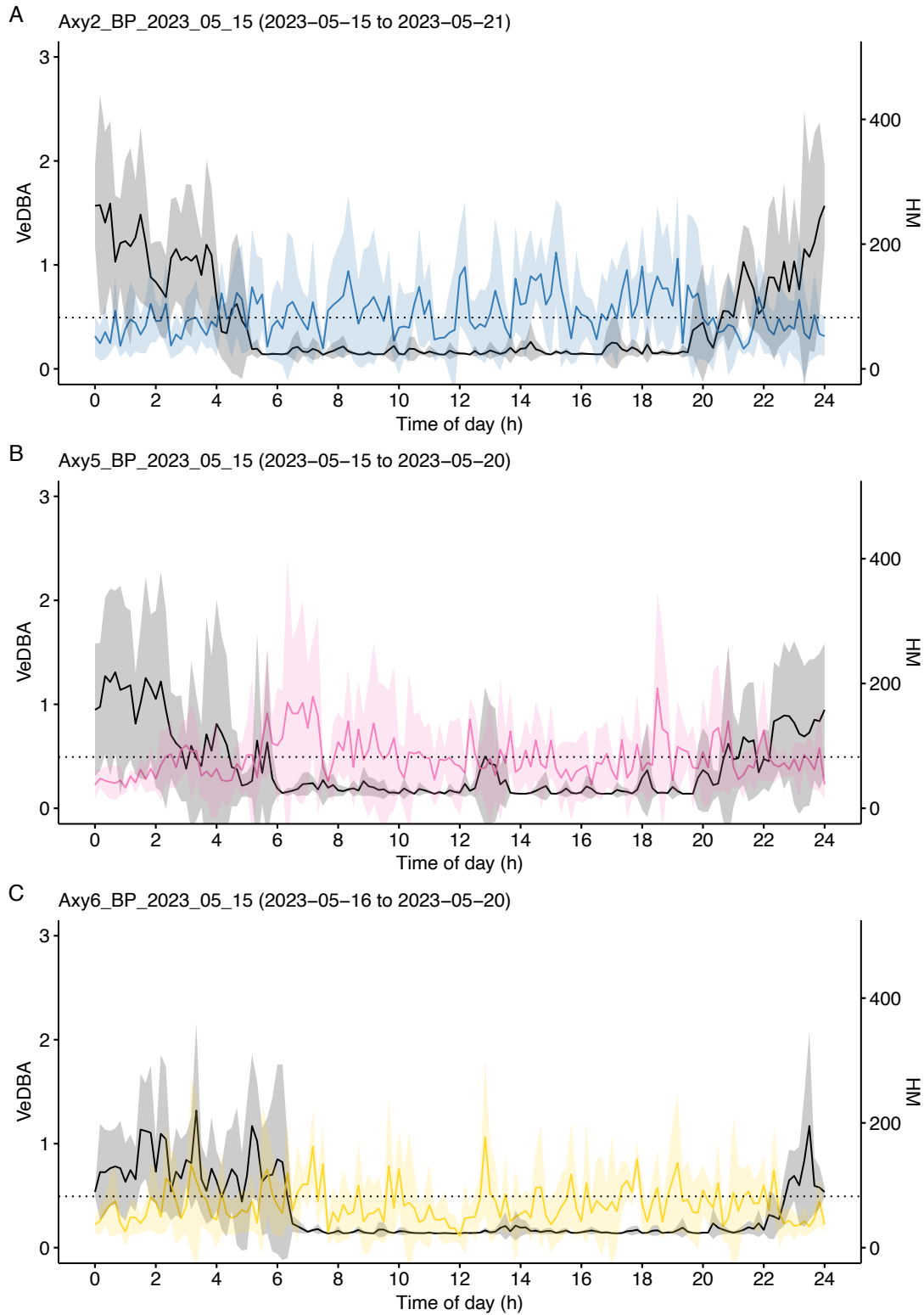
Horizontal movement did not exhibit a clear correlation with the activity patterns observed at both study sites (Fig. 17 - Fig. 20). Instead, it fluctuated throughout the day without any discernible peaks. Interestingly, levels of horizontal movement were slightly higher during the daytime compared to nighttime, despite overall lower activity levels during the day. The frequency of horizontal movement throughout the day remained consistent across different tracking periods and study sites, with no notable variations observed when comparing them (Fig. 17 - Fig. 20).



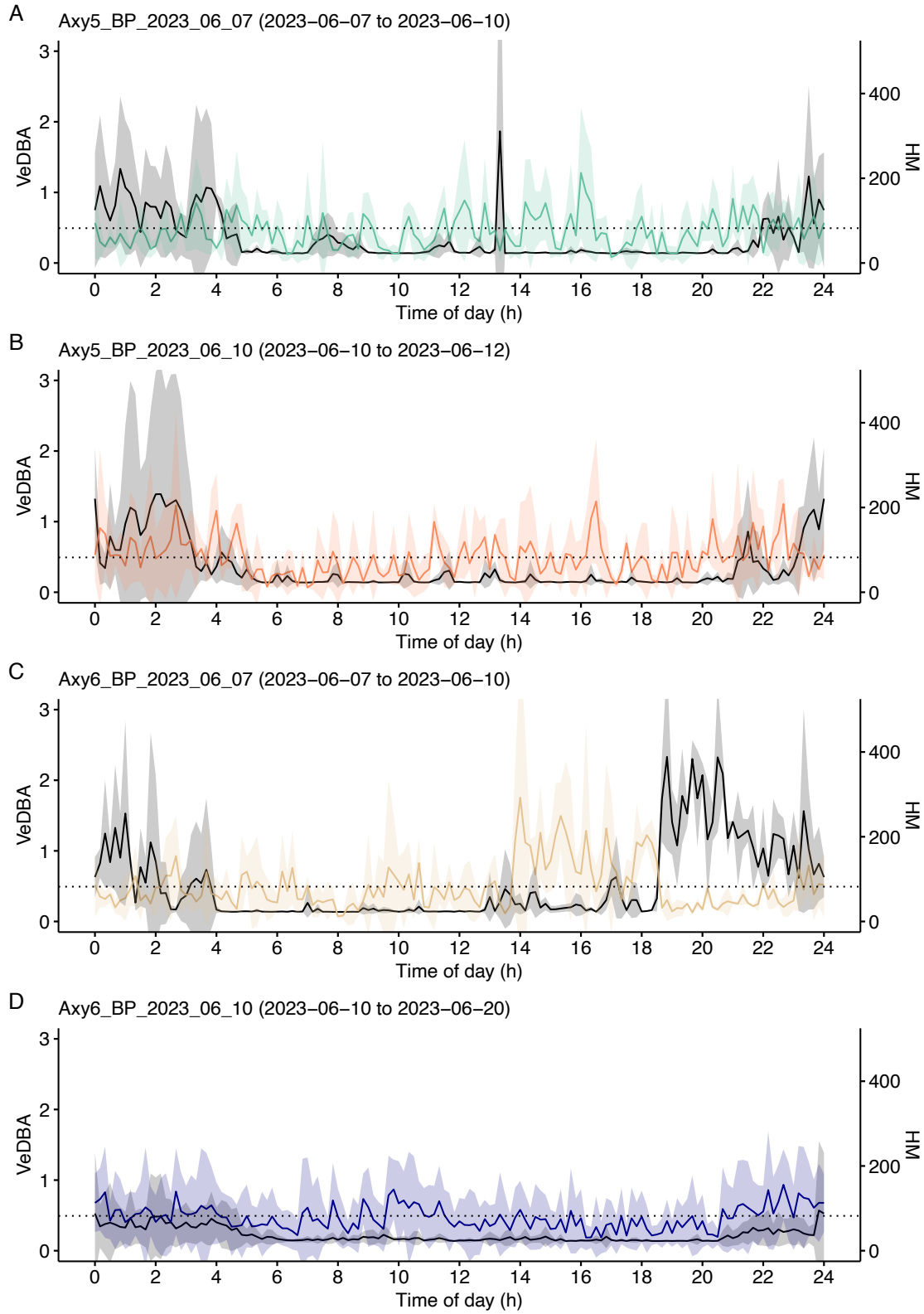
**Fig. 17: Horizontal movement and activity patterns during the first tracking period in the Seewinkel.** Each plot (A-E) shows one individual (ID and tracking date range on the top). The colored line represents the average daily horizontal movement (HM), which was calculated by averaging HM values, obtained by summing up each HM detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of HM frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



**Fig. 18: Horizontal movement and activity patterns during the second tracking period in the Seewinkel.** Each plot (A-F) shows one individual (ID and tracking date range on the top). The colored line represents the average daily horizontal movement (HM), which was calculated by averaging HM values, obtained by summing up each HM detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of HM frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



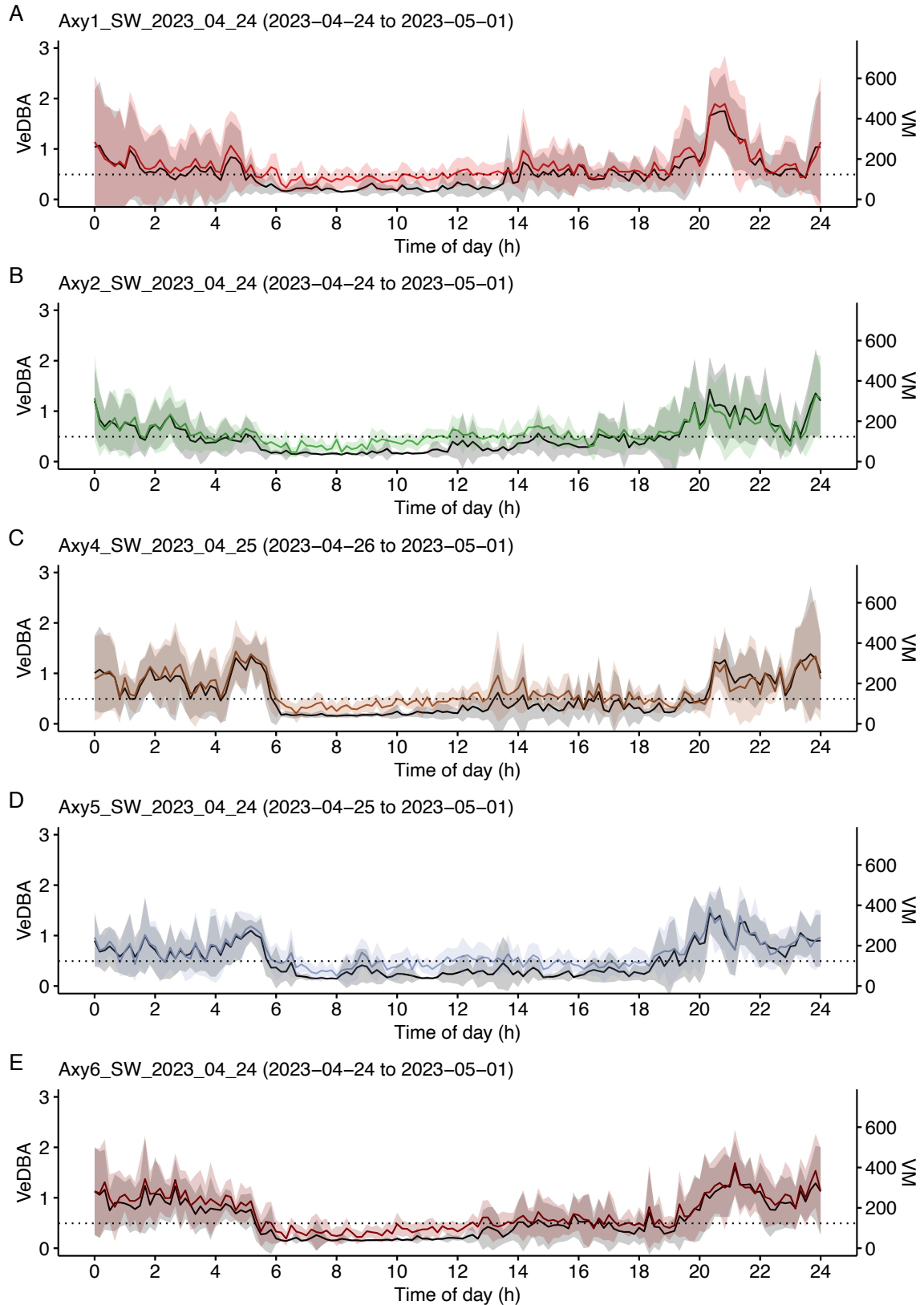
**Fig. 19: Horizontal movement and activity patterns during the first tracking period in the RBP.** Each plot (A-C) shows one individual (ID and tracking date range on the top). The colored line represents the average daily horizontal movement (HM), which was calculated by averaging HM values, obtained by summing up each HM detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of HM frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



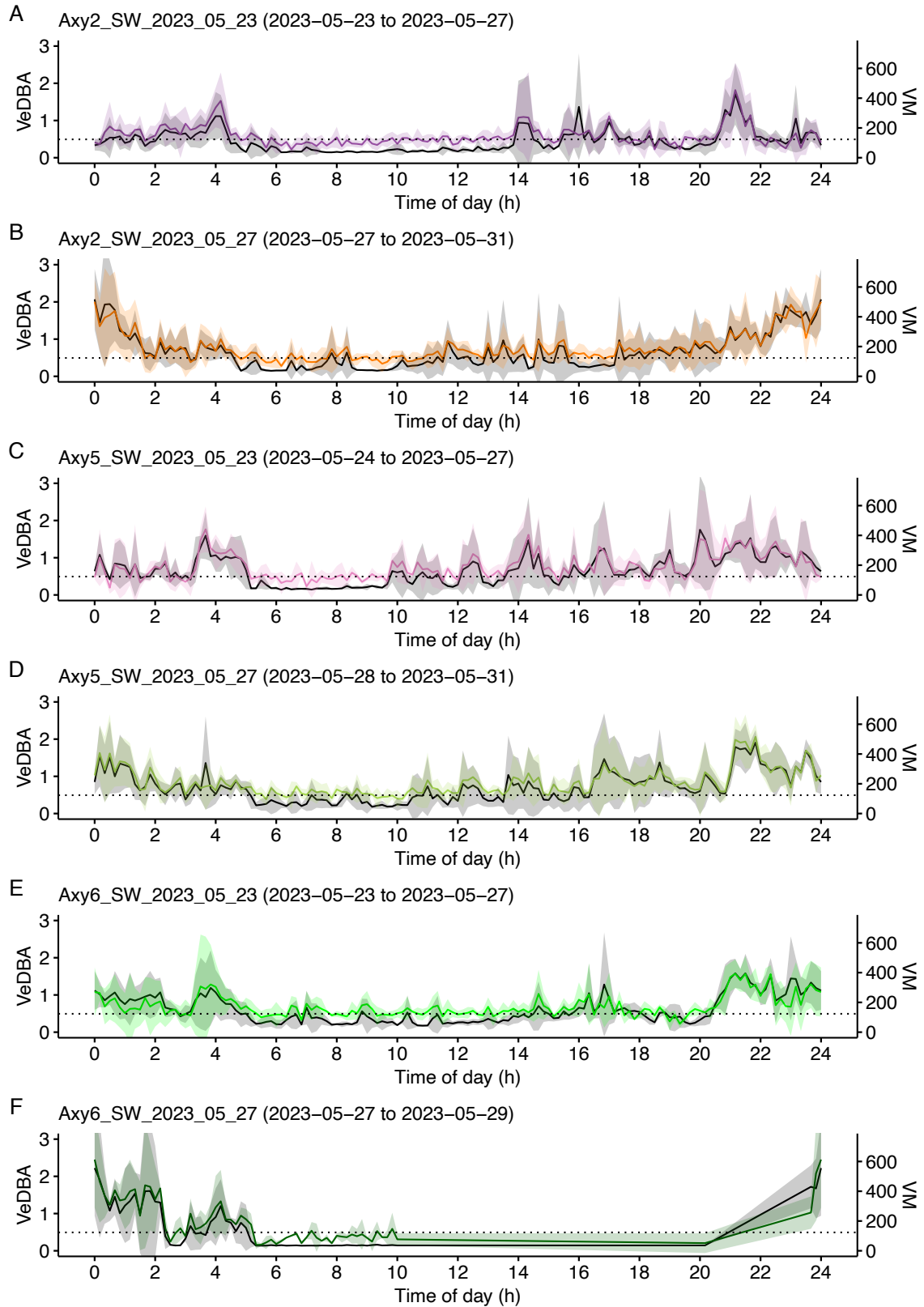
**Fig. 20: Horizontal movement and activity patterns during the second tracking period in the RBP.** Each plot (A-D) shows one individual (ID and tracking date range on the top). The colored line represents the average daily horizontal movement (HM), which was calculated by averaging HM values, obtained by summing up each HM detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of HM frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

### 3.3.5 Vertical Movement

The pattern of vertical movement throughout the day closely reflected activity levels (Fig. 21 - Fig. 24). At night, the frequency of vertical movement followed activity patterns, while during the day, vertical movement levels were slightly higher in proportion to general activity levels. A notable example illustrating this trend was observed in Axy2\_BP\_2023\_05\_15 (Fig. 23, Plot A). This variation was consistent across both the Seewinkel and the Rudolf Bednar Park at all tracking periods. Consequently, peaks in vertical movement occurred simultaneously with the highest activity levels, which predominantly happened at night, while lower levels of vertical movement occurred during the day.

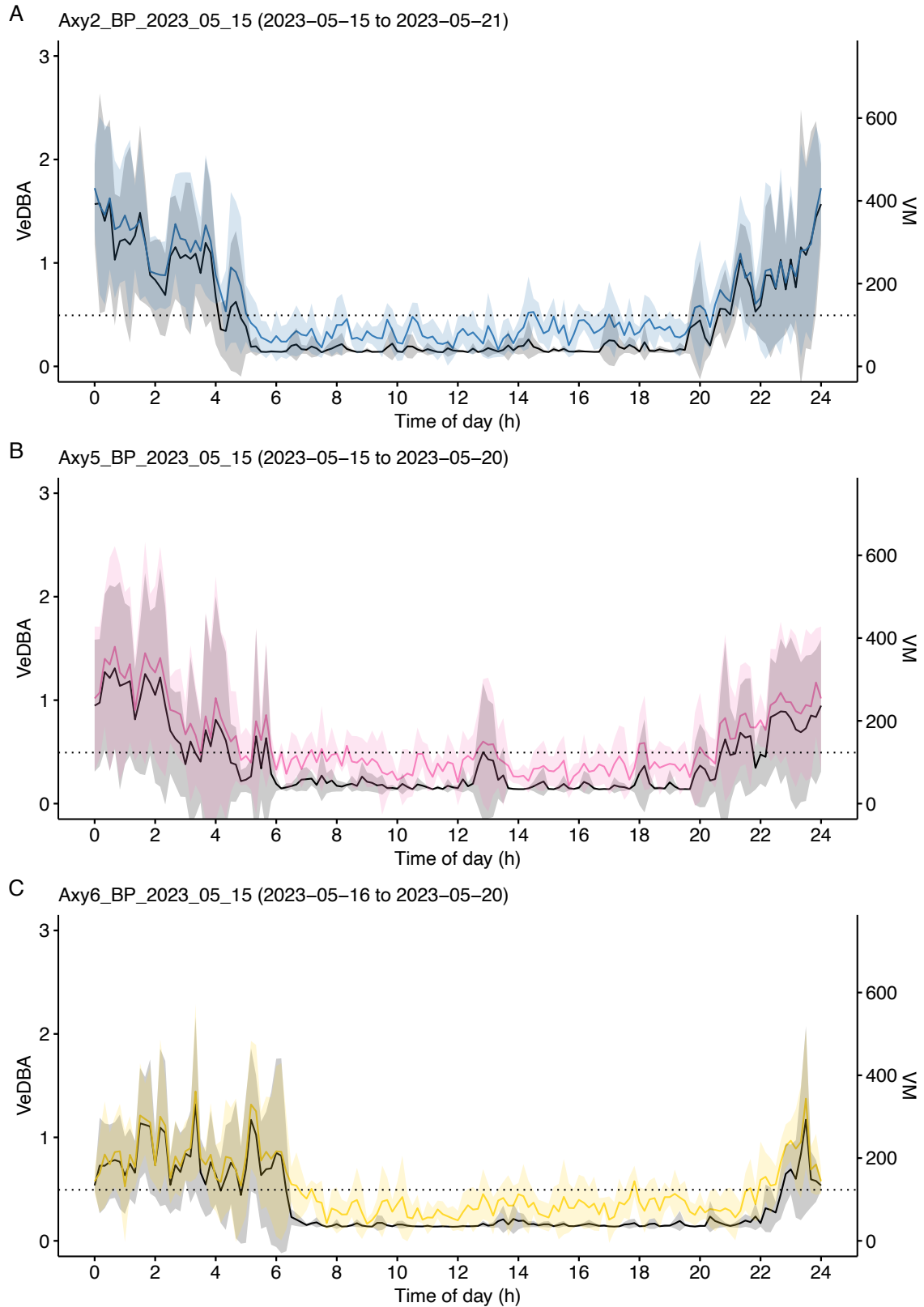


**Fig. 21: Vertical movement and activity patterns during the first tracking period in the Seewinkel.** Each plot (A-E) shows one individual (ID and tracking date range on the top). The colored line represents the average daily vertical movement (VM), which was calculated by averaging VM values, obtained by summing up each VM detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of VM frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

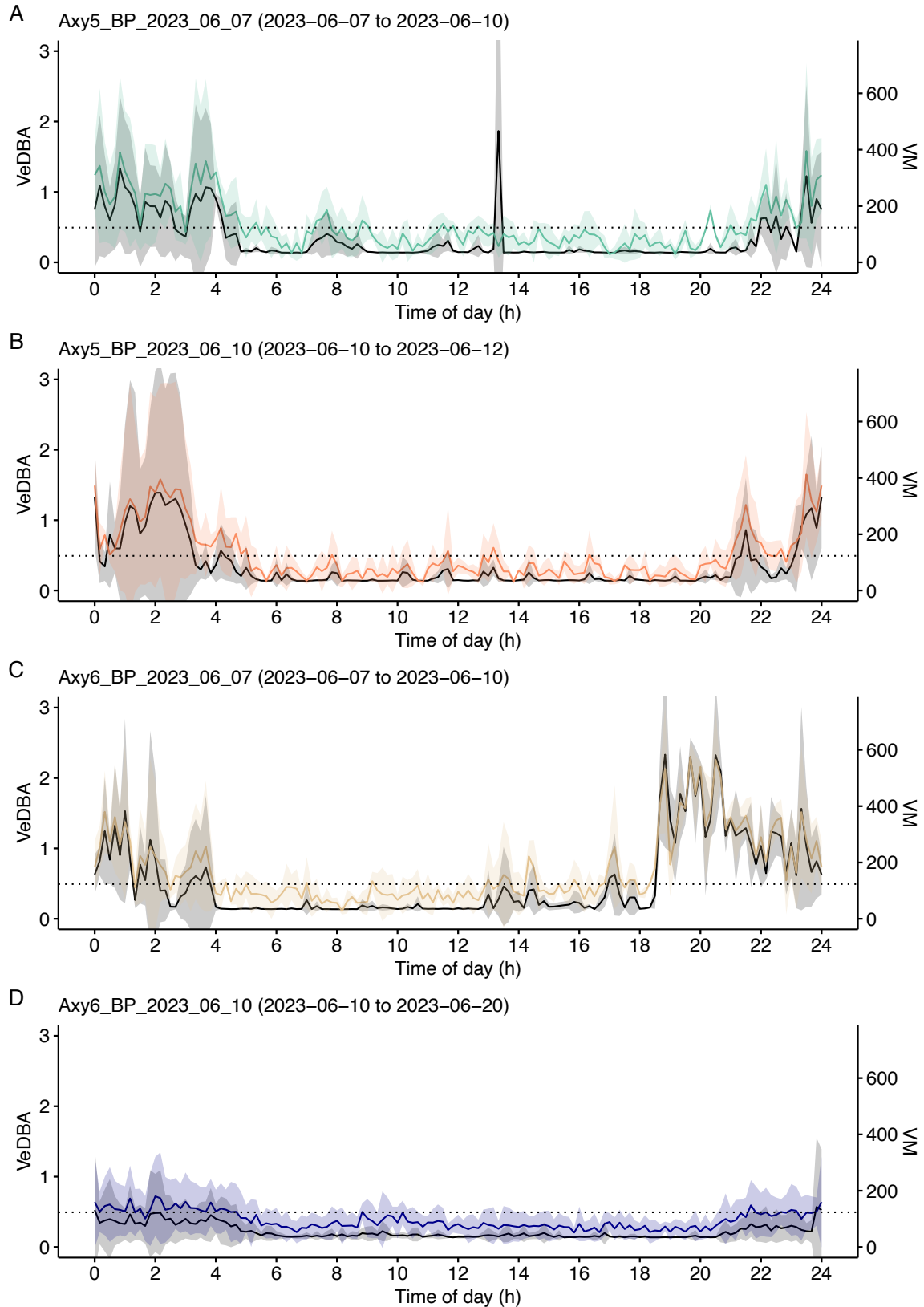


**Fig. 22: Vertical movement and activity patterns during the second tracking period in the Seewinkel.** Each plot (A-F) shows one individual (ID and tracking date range on the top). The colored line represents the average daily vertical movement (VM), which was calculated by averaging VM values, obtained by summing up each VM detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of HM frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).





**Fig. 23: Vertical movement and activity patterns during the first tracking period in the RBP.** Each plot (A-C) shows one individual (ID and tracking date range on the top). The colored line represents the average daily vertical movement (VM), which was calculated by averaging VM values, obtained by summing up each VM detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of HM frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



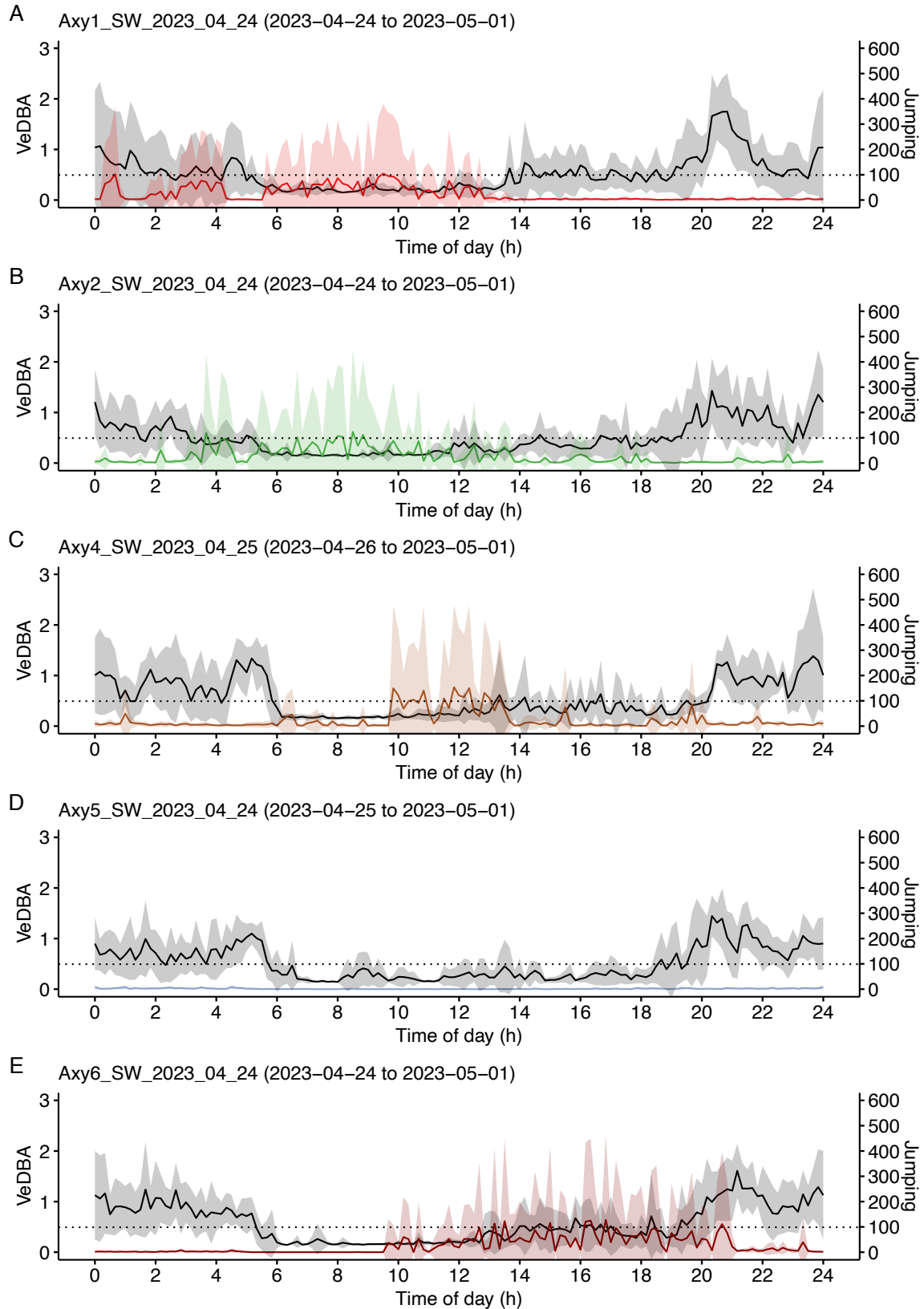
**Fig. 24: Vertical movement and activity patterns during the second tracking period in the RBP.** Each plot (A-D) shows one individual (ID and tracking date range on the top). The colored line represents the average daily vertical movement (VM), which was calculated by averaging VM values, obtained by summing up each VM detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of HM frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

### 3.3.6 Jumping

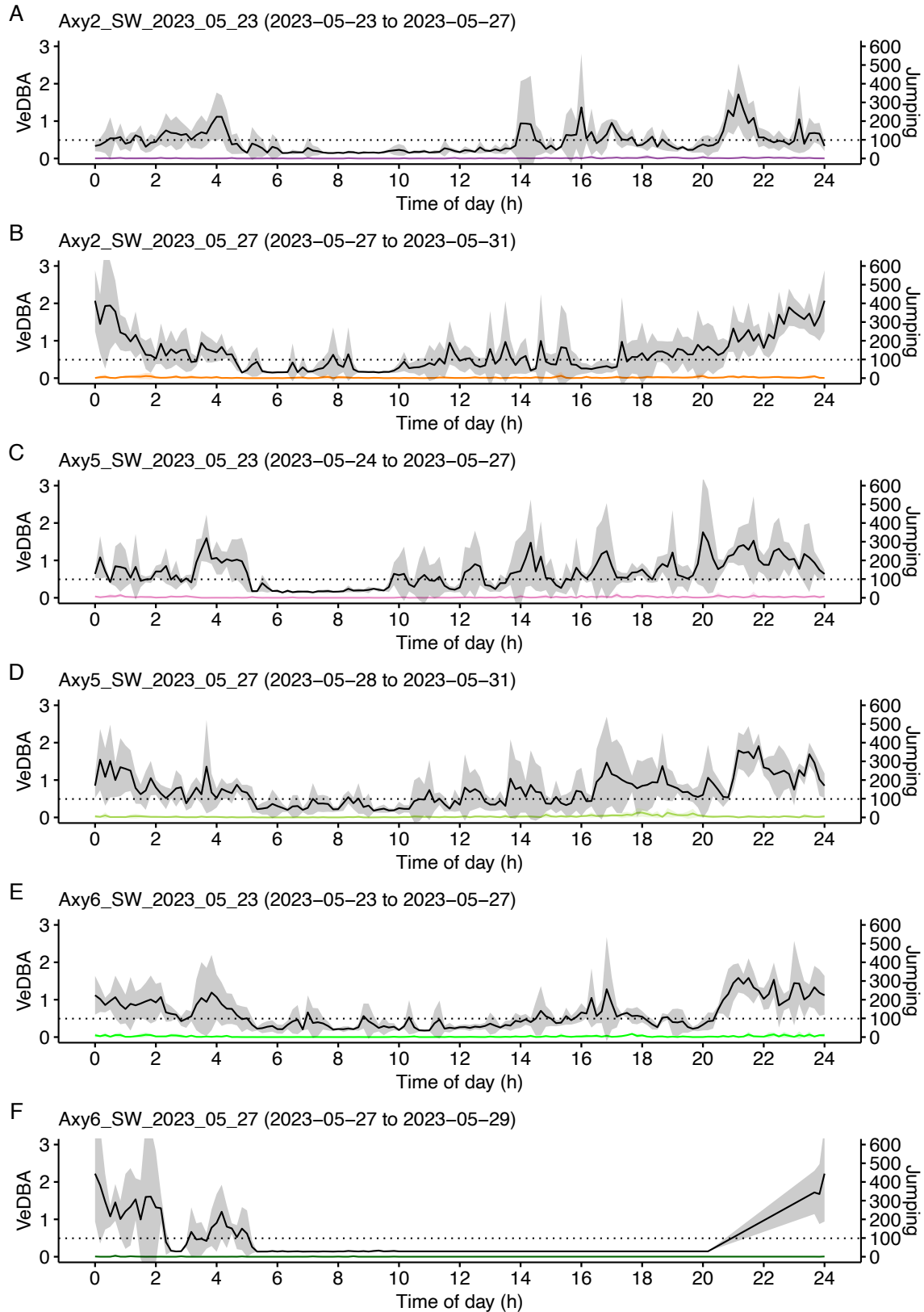
Jumping frequency did not correlate with activity patterns and was highly individual-dependent. While most toads showed no jumping events, some individuals exhibited high jumping frequencies at various times of the day (Fig. 25 - Fig. 28).

In the Rudolf Bednar Park, only one toad displayed a high jumping frequency distributed throughout the daytime, contrasting with activity levels, which were high during the day and low during the night (Fig. 27).

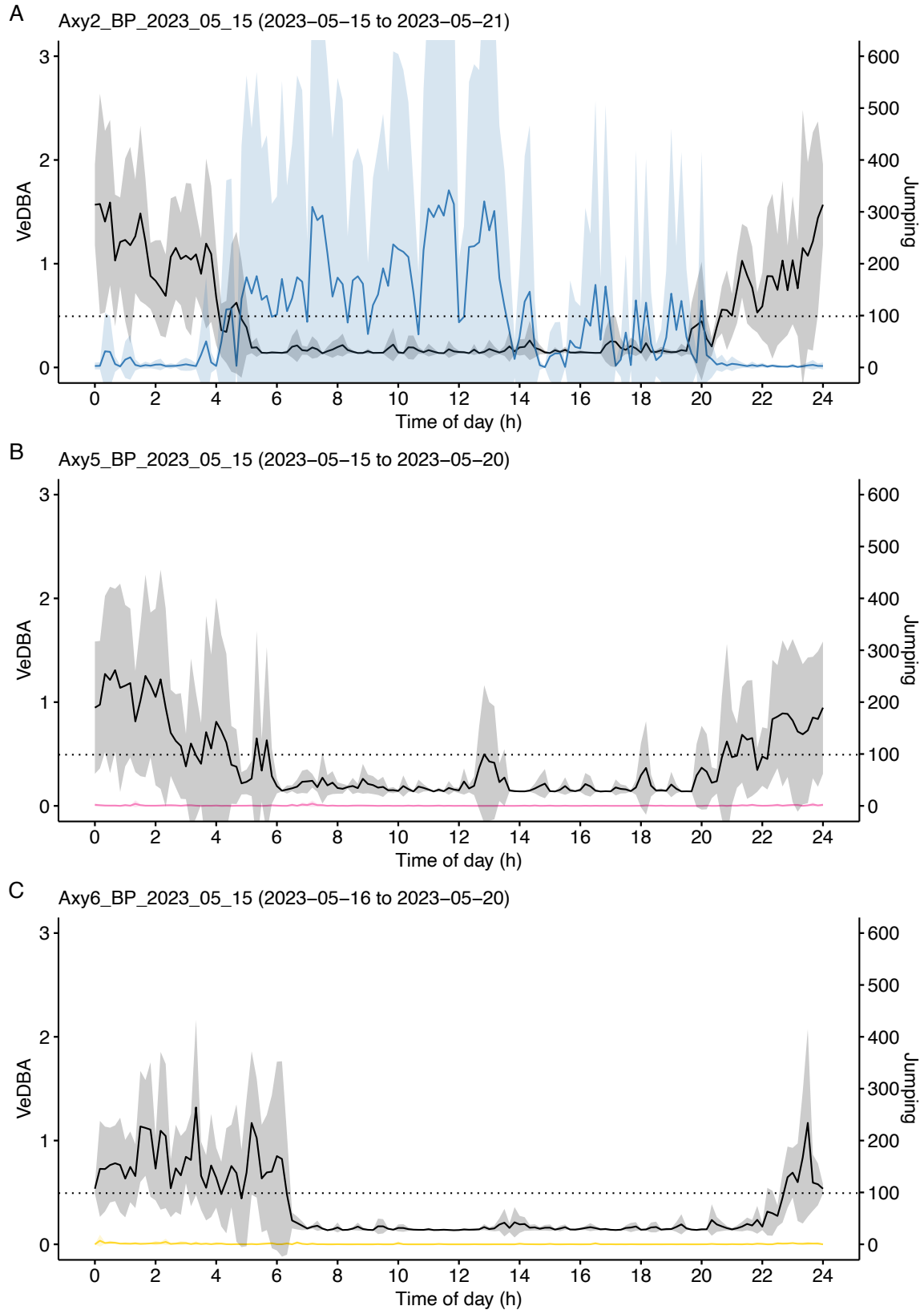
Notably, during the first tracking period in the Seewinkel, most toads demonstrated jumping behavior, whereas during the second period, no jumping was detected (Fig. 25, Fig. 26). The observed jumping behavior concentrated at specific times of the day but varied among individuals. For example, Axy4\_SW\_2023\_04\_25 and Axy6\_SW\_2023\_04\_24 exhibited jumping between 10 am and 9 pm, while Axy1\_SW\_2023\_04\_24 and Axy2\_SW\_2023\_04\_24 showed increased jumping events between approximately midnight and 2 pm (Fig. 25).



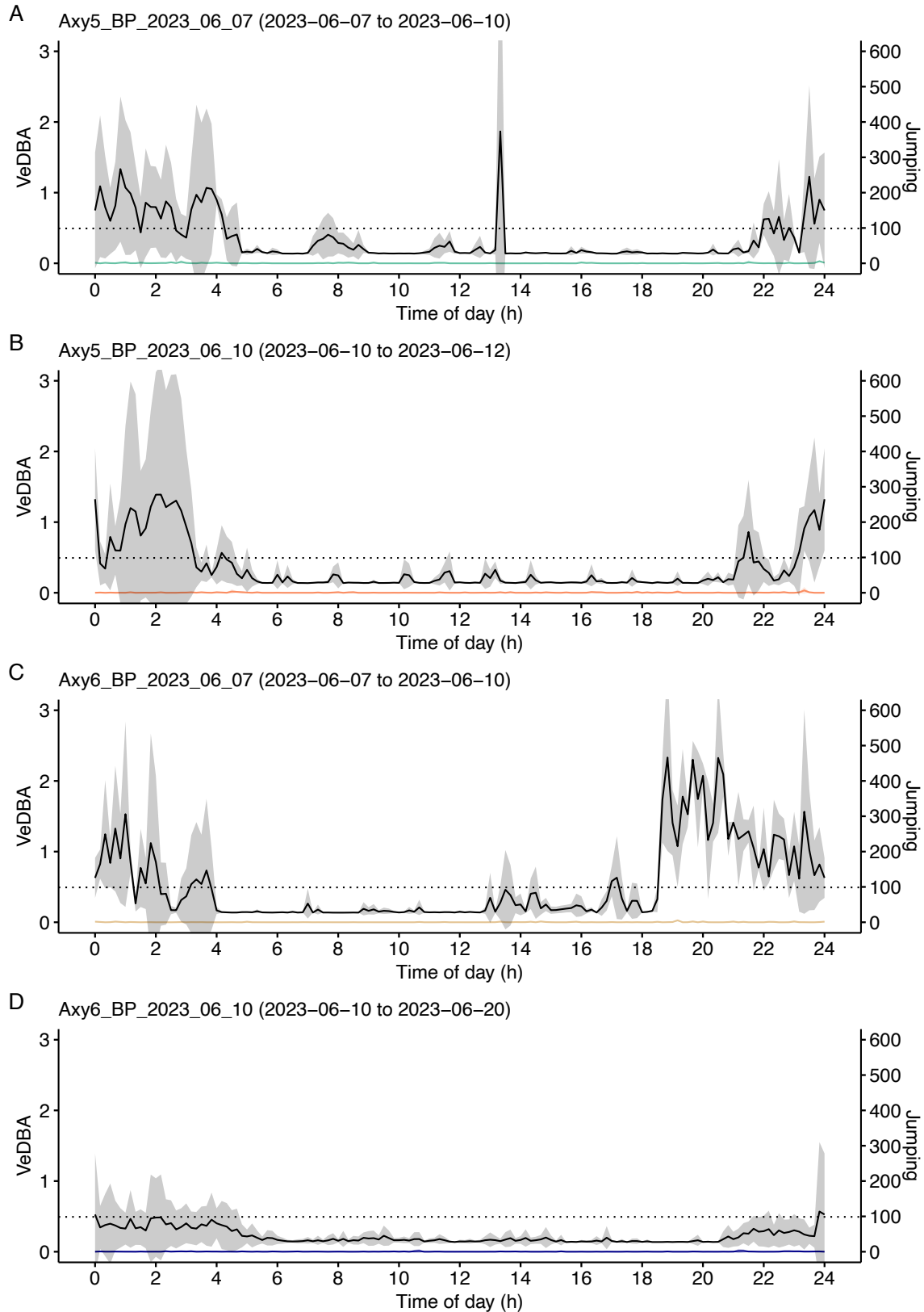
**Fig. 25: Jumping and activity patterns during the first tracking period in the Seewinkel.** Each plot (A-E) shows one individual (ID and tracking date range on the top). The colored line represents the average daily jumping frequency, which was calculated by averaging jumping values, obtained by summing up each jumping detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of jumping frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



**Fig. 26: Jumping and activity patterns during the second tracking period in the Seewinkel.** Each plot (A-F) shows one individual (ID and tracking date range on the top). The colored line represents the average daily jumping frequency, which was calculated by averaging jumping values, obtained by summing up each jumping detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of jumping frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



**Fig. 27: Jumping and activity patterns during the first tracking period in the RBP.** Each plot (A-C) shows one individual (ID and tracking date range on the top). The colored line represents the average daily jumping frequency, which was calculated by averaging jumping values, obtained by summing up each jumping detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of jumping frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

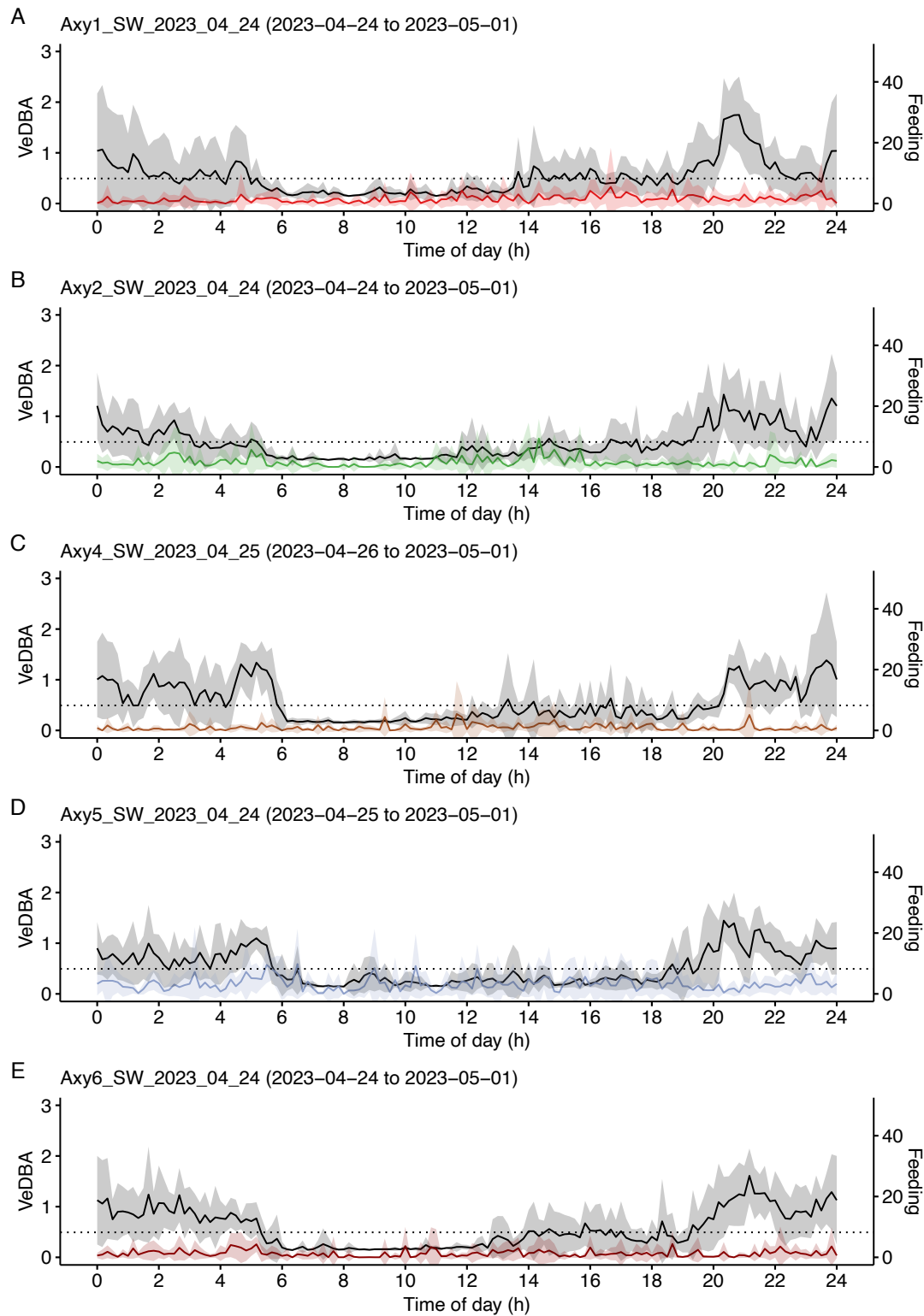


**Fig. 28: Jumping and activity patterns during the second tracking period in the RBP.** Each plot (A-D) shows one individual (ID and tracking date range on the top). The colored line represents the average daily jumping frequency, which was calculated by averaging jumping values, obtained by summing up each jumping detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of jumping frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

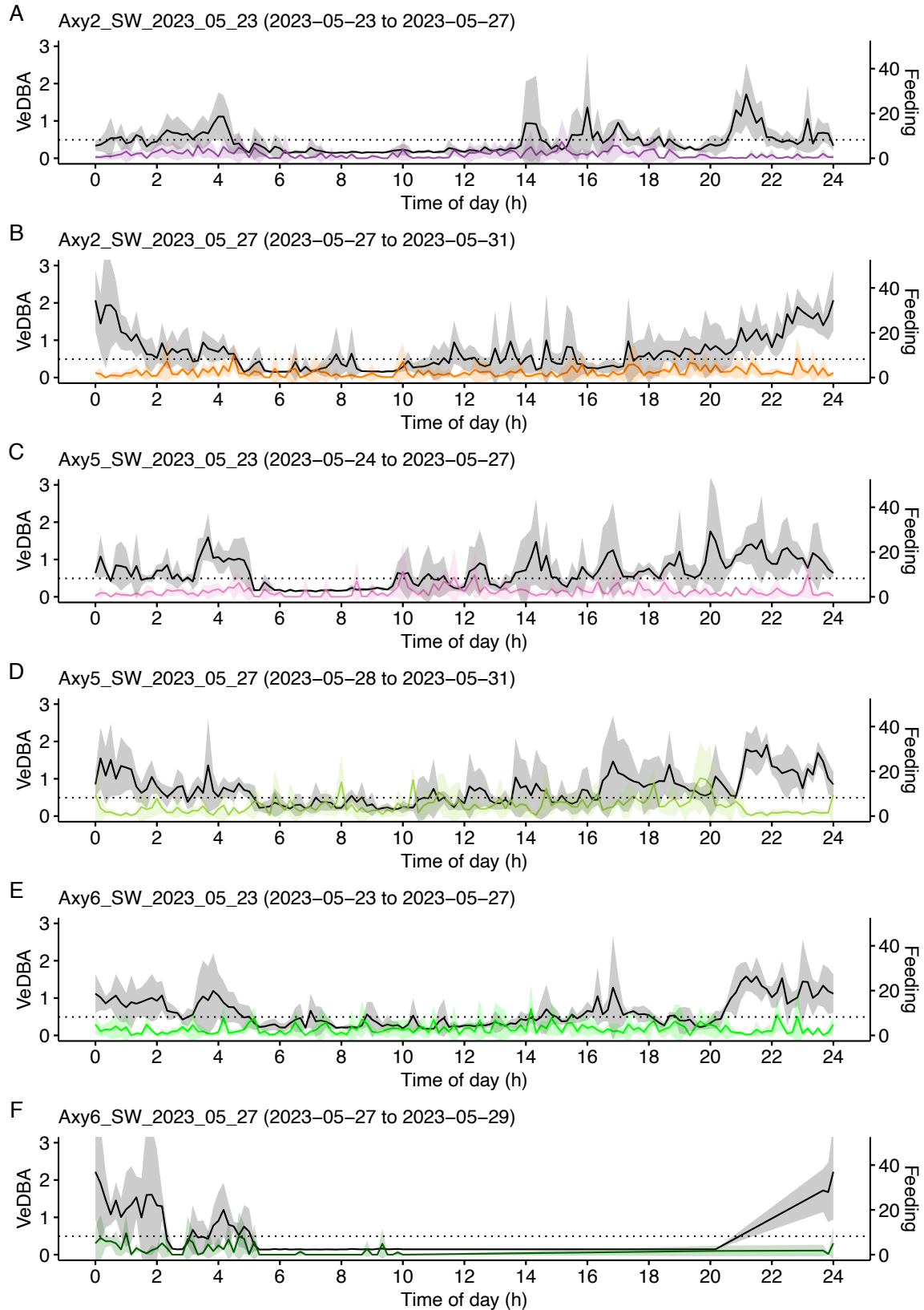
### 3.3.7 Feeding

Feeding activity showed a consistent distribution throughout the day at both study sites, irrespective of VeDBA levels. Overall, feeding frequency remained consistently low across all individuals and time points (Fig. 29 - Fig. 32).

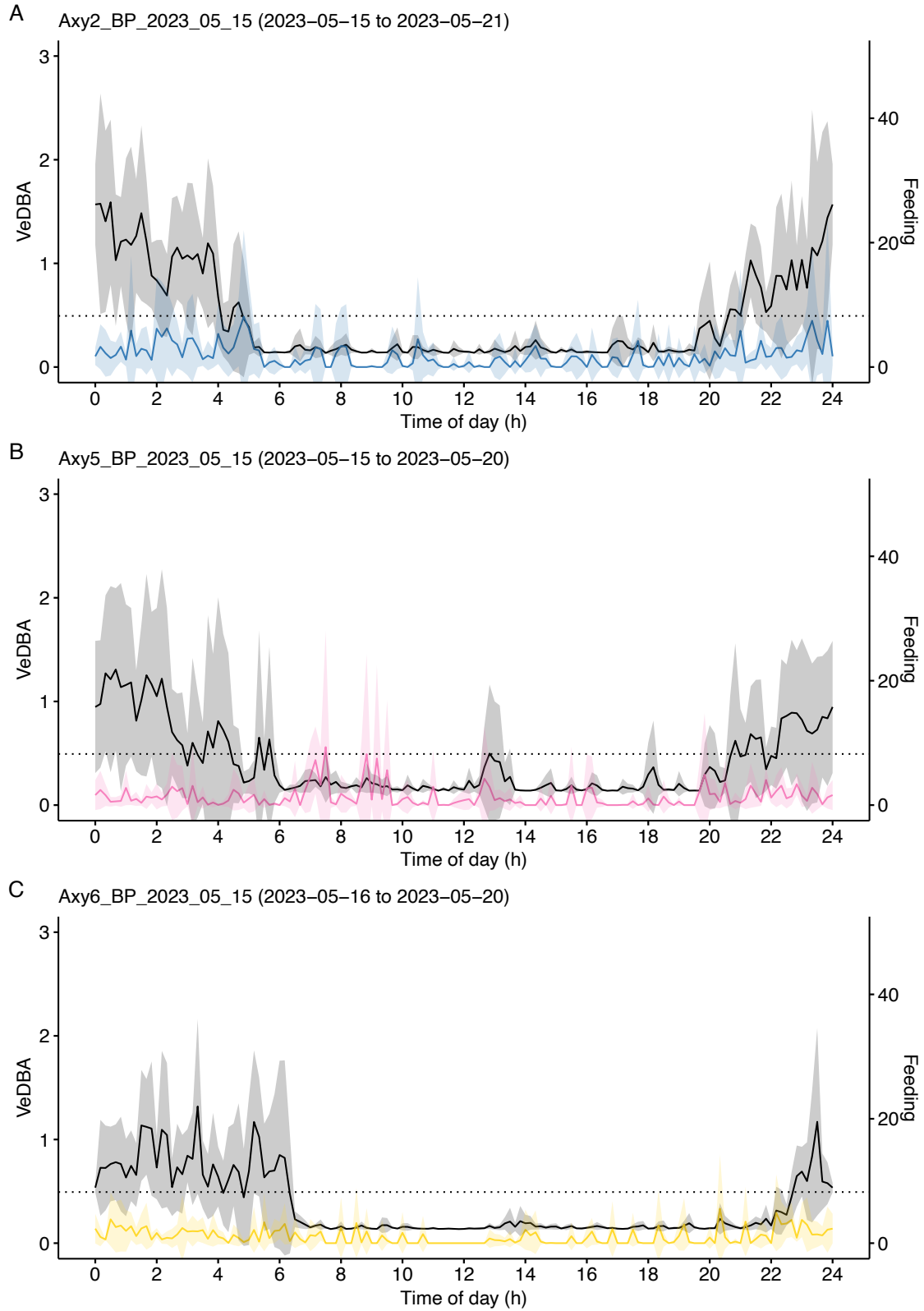




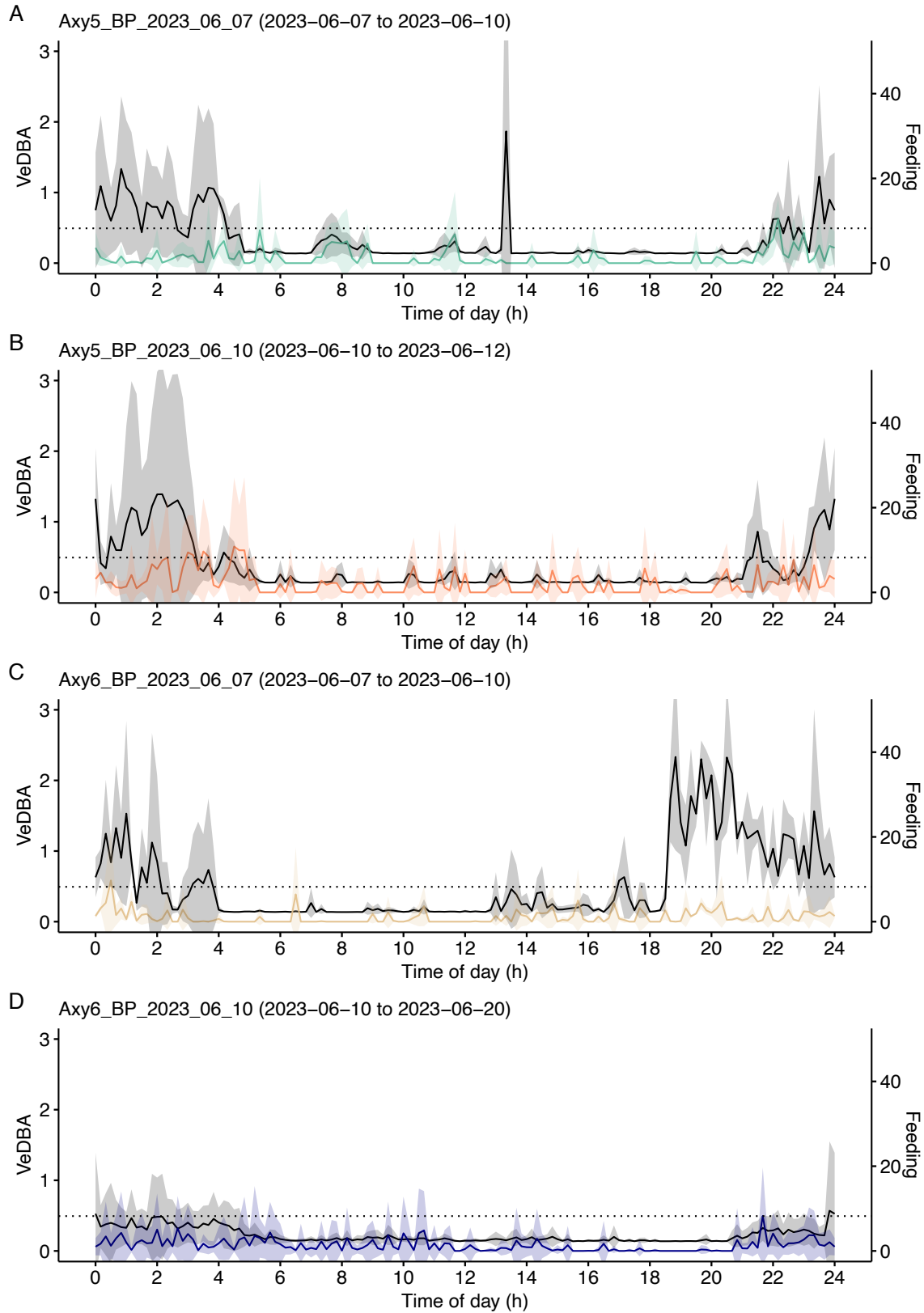
**Fig. 29: Feeding and activity patterns during the first tracking period in the Seewinkel.** Each plot (A-E) shows one individual (ID and tracking date range on the top). The colored line represents the average daily feeding frequency, which was calculated by averaging feeding values, obtained by summing up each feeding detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of feeding frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



**Fig. 30: Feeding and activity patterns during the second tracking period in the Seewinkel.** Each plot (A-F) shows one individual (ID and tracking date range on the top). The colored line represents the average daily feeding frequency, which was calculated by averaging feeding values, obtained by summing up each feeding detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of feeding frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).



**Fig. 31: Feeding and activity patterns during the first tracking period in the RBP.** Each plot (A-C) shows one individual (ID and tracking date range on the top). The colored line represents the average daily feeding frequency, which was calculated by averaging feeding values, obtained by summing up each feeding detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of feeding frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

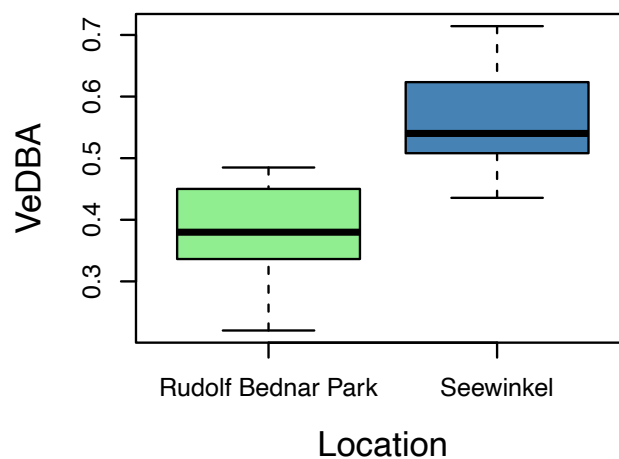


**Fig. 32: Feeding and activity patterns during the second tracking period in the RBP.**

Each plot (A-D) shows one individual (ID and tracking date range on the top). The colored line represents the average daily feeding frequency, which was calculated by averaging feeding values, obtained by summing up each feeding detected in 10-minute time sequences. The colored shaded area shows the standard deviation (SD) of feeding frequency. The black line illustrates the average daily VeDBA, while the grey area represents the standard deviation (SD). The dotted line depicts the overall mean VeDBA, computed across all 18 toads in the study (0.49).

### 3.5 Statistical analysis

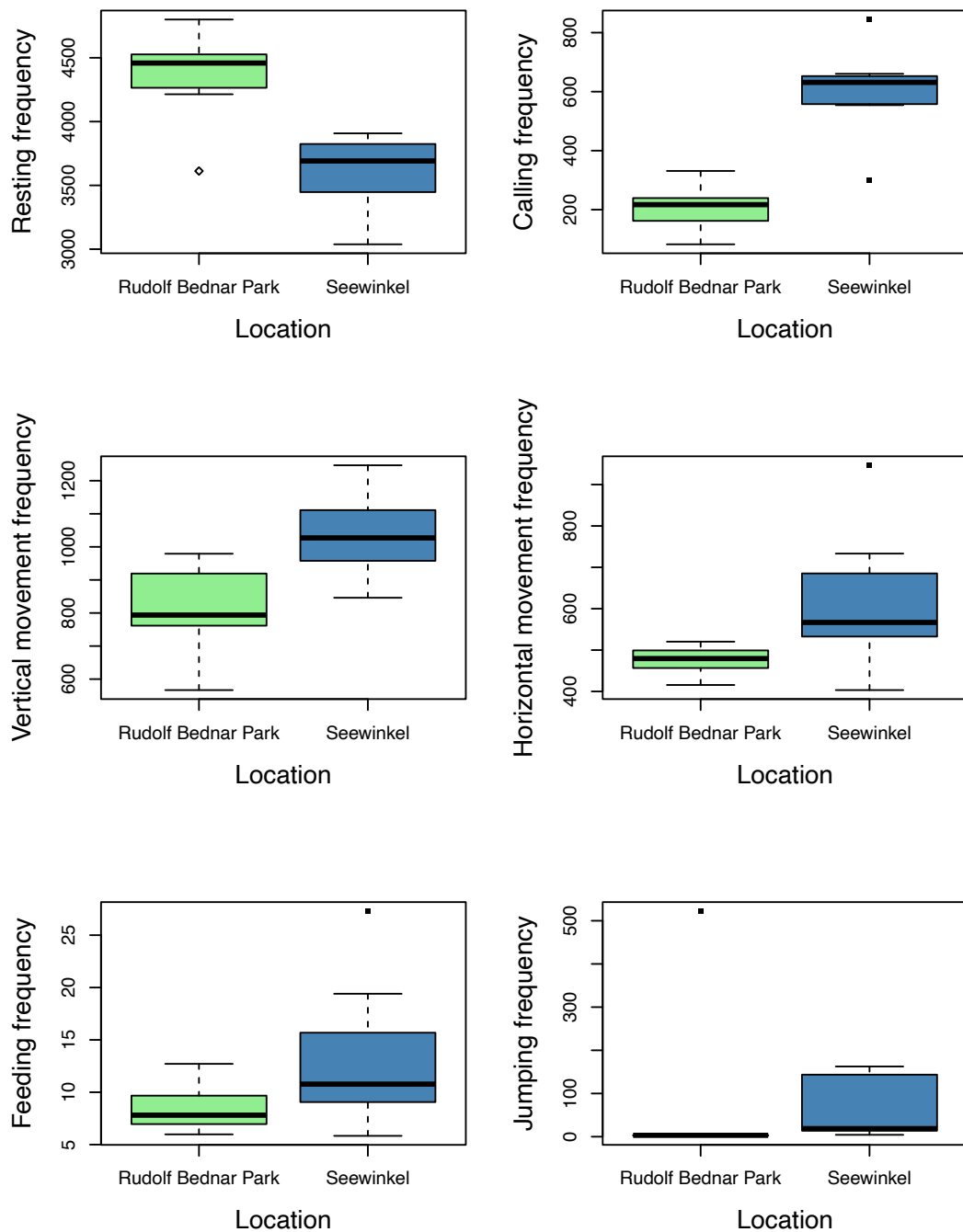
Comparison of the VeDBA values between the urban and natural populations revealed differences in activity levels. Boxplots illustrating the VeDBA distributions showed a median VeDBA of approximately 0.38 for the Rudolf Bednar Park population and 0.54 for the Seewinkel population (Fig. 33). Statistical analysis using the Wilcoxon rank sum test confirmed significant differences between the two populations ( $W = 4$ ,  $p = 0.002$ ). The test indicated a significant shift of VeDBA values, with the natural population exhibiting higher VeDBA levels compared to the urban population. The estimated difference in location was -0.174, with a 95% confidence interval ranging from -0.285 to -0.082.



**Fig. 33: Boxplots comparing the VeDBA (Vectorial Dynamic Body Acceleration) of the urban and natural population, calculated by determining means for each individual.**

Resting behavior exhibited a notable and highly significant distinction between the two populations ( $W = 70$ ,  $p = 0.004333$ ). The median resting time was higher in the urban population, indicating longer periods of rest compared to the natural habitat. Similarly, calling behavior also showed significant variations ( $W = 1$ ,  $p = 0.0006832$ ), with the natural population having higher levels of calling. Jumping behavior differed significantly between the urban and natural population ( $W = 11$ ,  $p = 0.01275$ ), as well as horizontal movement ( $W = 13$ ,  $p = 0.02092$ ). The natural population displayed higher levels of horizontal movement compared to the urban habitat. Vertical movement also exhibited highly significant differences ( $W = 8$ ,  $p = 0.00574$ ), with the natural population showing higher levels of vertical movement. Jumping behavior differed significantly between the two populations ( $W = 11$ ,  $p = 0.01275$ ). Toads in the natural habitat displayed higher jumping frequencies compared to their urban counterparts. It's worth noting that there was a strong outlier for jumping behavior in the Rudolf Bednar Park (Fig. 34), with one individual exhibiting much higher jumping frequencies than the

others in the habitat (Axy2\_BP\_2023\_05\_15, Fig. 27). Although not statistically significant, there was a trend towards differences in feeding behavior ( $W = 20$ ,  $p = 0.09384$ ), with slightly higher feeding frequencies observed in the natural population. These findings underscore distinct behavioral patterns between urban and natural habitats among green toads, suggesting potential environmental influences on behavior.



**Fig. 34: Boxplots of the behaviors resting, calling, vertical and horizontal movement, feeding and jumping at the study sites Rudolf Bednar Park and Seewinkel, calculated by determining means for each individual.**

## 4. Discussion

The present study aimed to investigate the activity patterns and behavior of green toads (*B. viridis*) in urban (Rudolf Bednar Park) and natural (Seewinkel) habitats using accelerometer data. Our results indicate significant differences between the two habitats.

A key finding of this study is the significantly lower activity level of green toads in the urban habitat, with a median VeDBA of 0.38 for the RBP population, compared to 0.54 for the Seewinkel population. Having a closer look at the daily activity patterns, the VeDBA levels of all toads are increased during nighttime. However, the contrast of low daytime activity to increased nighttime activity is considerably stronger in the urban population. Resting patterns follow this trend the other way around, indicating a consistent resting phase of the urban toads during the day, resulting in an overall altered proportion of resting behavior (about 13 % more) compared to the toads in the natural habitat. The increased overall activity during nighttime can be explained by the nocturnal ecology of green toads (Glandt, 2018). One reason for the reduced activity in the urban habitat is likely that home ranges of green toads in the RBP are significantly smaller than in their natural habitat (Lehofer, 2024; Spießberger et al., 2023). Furthermore, during the day the Rudolf-Bednar-Park is highly frequented by humans, resulting in strong disturbance for the amphibians (pers. obs.). By restricting their activity to nighttime in the urban habitat, the toads reduce the risks associated with the presence of humans. Shifting or restricting activity to the nighttime in urban habitats has been observed in other species, such as coyotes (*Canis latrans*) and bobcats (*Lynx rufus*) in urban areas of California (Ditchkoff et al., 2006; Dunagan et al., 2019; Riley et al., 2003). On the other hand, the surprisingly high activity during the day of the rural population could be caused by the time of data collection. Tracking was conducted during the breeding season of green toads. Perhaps high competition between the male toads and strong reproduction pressure lead to an altered activity. For example, while data collection we were able to observe several breeding events during the day at Seewinkel study site (Fig. 35), while toads in the RBP spend daytime in their hiding spots (Lehofer, 2024). However, there is no clear evidence, further research would be needed to test this hypothesis.

The reduced overall activity level of the RBP toads could represent an adaptation to the urban environment. Limiting activity could lead to a reduced probability of road mortality, which is one of the most significant threats to amphibian populations worldwide (Hels & Buchwald, 2001). A general reduction in activity could also help to save energy, which might be beneficial for toads in urban areas where prey is typically less abundant (Fenoglio et al., 2020).



Fig. 35: Green toads mating in a soda pan at Seewinkel study site.

Average VeDBA (0.49) across all individuals was about 18 times higher than in the study of Spießberger et al. (2023), where the average VeDBA was 0.027. Two major differences between the studies might explain this deviation. First, Spießberger et al. (2023) included only individuals from the RBP study site, which, as our findings suggest, appear to be less active than toads in natural habitats. Second, their data collection took place after the breeding season, when overall activity was lower due to the absence of daily migrations between hiding spots and spawning ponds.

In the natural habitat, calling activity of the toads was significantly higher. The calling behavior is similar to the activity pattern, peaking particularly during the night. For RBP toads, the calling primarily occurred at night, which may be influenced by their tendency to rest in hiding spots during the day (Lehofer, 2024). It could also serve as a predator avoidance strategy, because during daylight there are many dogs and crows in the park area, which are potential predators for the toads. A study by Gomez (2024) found that the calling frequency of the strawberry poison dart frog (*Oophaga pumilio*) decreased in the presence of predators, supporting the idea that increased predator presence could lead to a reduction in calling behavior to avoid being detected.

Less calling in the urban habitat could also be an adaptation to save energy. McCauley et al. (2000) found that resource availability is crucial for the calling intensity of anurans and prey is usually limited in urban environments (Fenoglio et al., 2020; Landler et al., 2022). The results of this study contradict the findings of Vargová et al. (2023), who came to the conclusion that the urban environment has no effect on the calling behavior of green toads. In contrast to our study, recordings only took place during the evening hours, whereas our data revealed that calling in the natural environment also took



place in the afternoon. Therefore, probably parts of the calling events were not recorded in the rural habitat by Vargová et al. (2023).

The Seewinkel population displays higher levels of horizontal and vertical movement than the Viennese toads, which is not surprising as the urban individuals have smaller home ranges and tend to be less active in general (Lehofer, 2024). Considering the course of horizontal movement throughout the day there is a noticeably high level of HM during daylight hours compared to the activity pattern. This could be caused by stationary movements, which were identified as HM by the algorithm. During data collection we were able to observe that green toads often turn around while sitting on one spot. Another behavior that could have been identified as HM could be movements to dig themselves in the ground. Furthermore, high HM levels might also be due to toads remaining in water during the day and being rocked back and forth by water movement, e. g. under rainy or windy conditions. Subsequently to this study it would be interesting to compare high events of HM with environmental conditions and to examine whether these movements also occur while a toad remains in the water.

Vertical movement levels align with the activity curve and display most probably movements in the water and crossing obstacles. The green toads we tracked often hid by diving to the ground in open water bodies. Moreover, RBP toads had to climb the edges of the water basins used as spawning ponds each night they migrated for breeding, consequently moving in vertical direction.

Jumping and feeding behavior also differ between the two habitats, being higher in the Seewinkel area. This might also be caused by a higher general activity of the rural toads. Peaks of jumping and feeding behavior are distributed very unevenly depending on daytime and the specific toad. Both do not follow the course of activity. Jumping seems to be very individually dependent, e. g. Axy2\_BP\_2023\_05\_15 displayed high levels of jumping frequency, while Axy6\_BP\_2023\_05\_15 showed no jumping behavior at all. This could be explained by individual behavioral differences within the populations. Jreidini and Green (2024) radio-tracked 13 Fowler's Toads (*Anaxyrus fowleri*) of one population and analyzed their behavior. They found out that the frogs exhibit strong individual variation in the frequency of certain behaviors and how long behaviors continue.

Feeding behavior seems to occur randomly throughout the day. One possible reason for this could be a lack of training data available to the algorithm for detecting this behavior. Few feeding events were observed in the captive toads, which may have limited the accuracy of the algorithm. This limitation is evident in the histogram plots of sequential behavior frequencies, which show low plausibility for feeding data. In contrast, less complex behaviors such as resting exhibit higher plausibility.

During data collection some complications occurred. A major obstacle was the limited lifespan of the transmitters, only one device was still working at the end of the data collection. In some cases, the transmitters slipped onto the animals' bellies, which

required the data to be removed from the analysis until the position was corrected. In the case of Axy6\_SW\_2023\_05\_27 after the exclusion of a period with a slipped tracking device, there was not enough data available to create an activity curve throughout the day. As a result, the line is linear from 10 am to 12 pm. Tracking in the RBP involved some challenges. One transmitter was probably removed by a park visitor, as it was lying on a bench with the silicon tubing opened. In another incident, a toad was found dead and dried out during the daily site check, with the transmitter lying next to it (Axy5\_BP\_2023\_05\_15). At the same time another transmitter was discovered a few meters away, but without any sign of the corresponding toad (Axy6\_BP\_2023\_05\_15). The cause might also have been human intervention. Moreover, dark spots were visible on the skin of some animals after removal of the transmitters, which were caused by irritation from the devices. In another tracking study of green toads, problems also occurred due to injuries, leading Ott (2015) to remove the transmitters prematurely to prevent further damage.

The study provides new insights into the life of green toads, building upon Spießberger et al. (2023) by analyzing their behavior and comparing an urban population to toads in a rather natural environment. The study shows reduced activity levels and behavioral frequencies in urban habitats, suggesting possible adaptations to urban conditions. Future studies comparing the genetics of RBP green toads with natural populations could further deepen our understanding of their adaptation mechanisms.

The data collected provides an important contribution to the understanding of movement patterns in amphibians. The use of accelerometers has rarely been applied to amphibians (Halsey et al., 2009; Spießberger et al., 2023). For green toads, this study represents a first approach that provides insights into behavior over the course of the day. Even though accuracy of the algorithm used in this study is already high, further development and training of the algorithm with additional behavioral data could enable the recognition of more complex behaviors. Smaller transmitters and a longer battery life would make it possible to collect data over a longer period of time and analyze behaviors in different ecological phases of the year.

The accelerometer data can be a valuable tool to assess differences among populations, as demonstrated in this study, but it could also be used to analyze individual differences and calculate the energetic needs of a species or population, thereby making it possible to determine the resource requirements for the specific animals. Halsey and White (2010) analyzed accelerometer data of the in Australia invasive species *Bufo marinus* to maximize control efficiency. The detailed insights in the movement ecology of animals should also be applied to set guidelines for conservation. Data from tracking studies can be used to better understand animal behavior and environmental changes, for example by monitoring disturbances or analyzing energy requirements (A. D. M. Wilson et al., 2015). The data collected in this study contribute to a better understanding of the ecology of green toads and could play

an important role in evaluating potential habitats. Green toads often inhabit disturbed environments such as construction sites, which they frequently lose when these areas are developed (Landler et al., 2023). A deeper understanding of their ecological requirements is important for designing urban landscapes to be suitable habitats. Green toads can survive in highly human-influenced environments, as demonstrated by the RBP population and many other urban populations (Mazgajska & Mazgajski, 2020; Sistani et al., 2021; Vargová, Balogová, et al., 2023). Conservation-oriented urban planning should incorporate these insights to develop targeted measures that preserve and create suitable habitats.

Although the data from tracking systems offer many possibilities, up to now they are rarely applied in conservation strategies and translated into concrete measures (Katzner & Arlettaz, 2020; A. D. M. Wilson et al., 2015). The combination of tracking and accelerometer data offers untapped potential that, if effectively utilized, could improve both the understanding and protection of amphibian species, especially considering the ongoing global biodiversity crisis.

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