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**Emergence behaviour of lesser horseshoe bats  
(*Rhinolophus hipposideros*):  
Intracolony variation in time and space  
(Carinthia and Salzburg, Austria)**

by

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**Räumliche und zeitliche Variabilität im Ausflugsverhalten Kleiner Hufeisennasen  
(*Rhinolophus hipposideros*)  
(Kärnten und Salzburg, Österreich)**

**Synopsis:** We studied variation of the nightly emergence behaviour of lesser horseshoe bats at two maternity colonies in the provinces of Carinthia and Salzburg, Austria. We focussed on the intracolony variation in space and time and conducted a case study in which we manipulated the flight paths of the bats by presenting a temporarily erected hedgerow, in order to optimise the emergence conditions. As its central hypothesis, this study considers the timing of the evening emergence to be a compromise between two conflicting demands: i) aerial insects show peak abundance around dusk, when most bats are still confined to their roosts and ii) predation risk decreases as light intensity decreases.

Emergence time was highly correlated with sunset, but also dependent on weather and reproductive state of the females. Increasing cloud cover resulted in progressively earlier emergences. Cloud cover also affected the relative use of two different emergence routes at one of the two roosts. Lactating females emerged earlier than pregnant ones and the earliest emergence could be noted for post lactating bats.

The erection of an artificial hedgerow at the roost Schwarzenbach affected the relative use of emergence routes (and the height of the flight path), with more individuals using the new path. We conclude that optimised emergence conditions might result in an earlier and safer emergence. This should positively influence the fitness of individual bats. For a threatened species like the lesser horseshoe bat optimising their emergence routes might be a helpful conservation measure that easily could be achieved. However, it has to be noted that bats will react with a delay to changed emergence conditions and time for adopting is necessary.

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## **1. Introduction:**

Though accounting only for a very short proportion of the daily time budget, the nightly emergence from the roost can have a critical impact on the fitness of individual bats (DUVERGÉ et al. 2000). Moreover, emerging bats have a higher predation risk compared to foraging or roosting bats, because of being conspicuous and predictable (GILLETTE & KIMBOURGH 1970, SPEAKMAN et al. 1999).

As its basic hypothesis, this study considers the timing of the evening emergence to be a compromise between two conflicting demands: i) aerial insects show peak abundance around dusk, when most bats are still confined to their roosts (RACEY & SWIFT 1985, RYDELL et al. 1996) and ii) decreasing predation risk as light intensity decreases (DUVERGÉ et al. 2000).

The emergence time can be modified by a number of factors: exogenous factors like insect availability (RYDELL et al. 1996), weather (MCANEY & FAIRLEY 1988), external light (REITER et al. in prep.) or endogenous factors like the reproductive state of the females and the nutritional state of the bats (DUVERGÉ et al. 2000).

The distance of the roost to foraging areas is an important factor explaining the distribution and selection of roosts (JENKINS et al. 1998, REITER 2004b). Tall vegetation like tree lines or hedgerows in the vicinity of the roosts reduces the predation risk, allowing an earlier emergence and thus increasing the foraging time of the bats (JENKINS et al. 1998).

Based on the hypothesis mentioned above, the aim of this study was to test the following predictions by means of studying the variation of the emergence behaviour at two colonies and by manipulating the flight paths by presenting a temporary and artificial hedgerow:

- a) The emergence time is overall related to the sunset time.
- b) The emergence time is modified by the reproductive state of the females and the ambient weather conditions.
- c) An optimised emergence route, i.e. a better connection of the roost to the next bush or tree, allows an earlier emergence.
- d) An additional hedgerow changes the proportion of bats using the different emergence route: more bats use the optimised flight path.
- e) The height above ground of flight path will increase with the presence of an additional hedgerow.

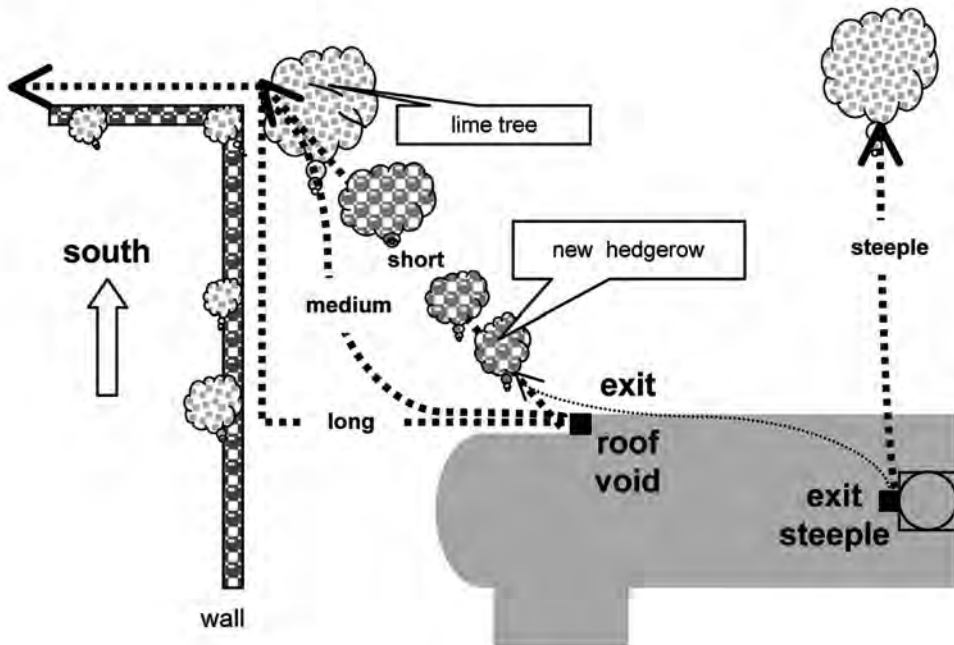
## 2. Material and methods:

### 2.1. Study colonies:

The study was conducted at two maternity colonies of lesser horseshoe bats (*Rhinolophus hipposideros*) situated in the Austrian provinces Carinthia and Salzburg: Tultschnig church near Klagenfurt (Carinthia, 13.14242°E, 46.655°N, 480 m a.s.l.), and Schwarzenbach church near Uttendorf (Salzburg, 12.592°E, 47.276°N, 778 m a.s.l.). In both buildings the bats roosted predominantly in the church tower, but occasionally the roof void was also used. Both roosts were not illuminated by external floodlight. The maximum numbers of adult and subadult bats counted were 52 in Tultschnig and 84 in Schwarzenbach, respectively.

At Tultschnig church two exits and flight paths (northern and southern) are used by the bats to leave the roost. On the southern side of the roof two windows are used as exit points and the bats fly subsequently along the roof. At the northern flight path the bats leave the roost through a small opening in the wall and fly along the wall afterwards. Thus, two observers are necessary to count the bats at the two different flight paths.

At Schwarzenbach church a hole in the roof void and a steeple window are used as exit holes. Four main flight paths exist: at 'roof void, long' the bats fly along the wall and from there they cross the open space of approximately 6 m to a wall covered with bushes, at 'roof void, short' the bats fly directly from the exit hole to the lime tree cross the open space (12 m), a small number of bats uses a flight path between the both flight paths, i.e. 'roof void, medium' and the bats from the steeple fly directly into a tree located in approximately 10 m distance (Fig. 1).



**Fig. 1:** Figure of the flight paths and experimental design at roost Schwarzenbach (not to scale). Flight paths are presented as dotted lines.

## 2.2. Data collection:

Field work at Tultschnig church was carried out from May 11<sup>th</sup> to October 3<sup>rd</sup> 2000, from April 4<sup>th</sup> to September 20<sup>th</sup> 2001, from March 16<sup>th</sup> to September 16<sup>th</sup> 2002, and from April 15<sup>th</sup> to October 1st 2003. Observations were made every 10-14 days, resulting in a total of 60 nights observed.

Field work at Schwarzenbach church was carried out daily from July 7<sup>th</sup> to July 14<sup>th</sup> 2002 resulting in a total of 8 nights observed.

The roosts were visited approximately 15 min before the expected emergence. The sessions were finished when no bats had left the roost for the last 15 min. Each emerging individual was visually registered. At Schwarzenbach church a bat detector (Pettersson D100, Pettersson Elektronik AB, Sweden) was used additionally.

At the roost Tultschnig each leaving bat was assigned to five-minute intervals, starting every full hour. Furthermore, the exact time of the first as well as the last leaving bat was recorded in the years 2001 to 2003 ( $n = 47$ ). At Schwarzenbach church the emergence activity was recorded on a micro-cassette recorder (Olympus, PearlCorder S701) and the recordings were subsequently analysed. Thus, every emergence event could be registered to an accuracy of  $\pm 1$  second.

The following variables describing emergence patterns were collected:

**Time of the median emerging bat (MET)** was calculated for the roost Tultschnig as follows: the five-minute interval of the median emerging bat was defined. This interval was then divided by the number of bats within this interval and thereafter multiplied with the number of bats until the median bat. The resulting value was summed with the starting time of the interval and registered to an accuracy of 30 seconds. The validity of this method was tested against the 1-second design, at three different colonies and three emerging counts respectively. The mean difference between the two methods was 30 sec (minimum 1 sec and maximum 41 sec) and no significant difference was measurable (Mann-Whitney-U-test:  $U = 39.0$ ,  $n = 18$ ,  $p = 0.931$ ; REITER 2002). Hence, this method was considered to be convenient for the purpose of the present study.

**Duration of emergence (minutes)** was measured as the time of the last emerging bat minus the time of the first emerging bat. Time of the first emerging bat (MET) was measured with an accuracy of 1 minute for the roost Tultschnig and 1 second for the roost Schwarzenbach. Time of the last emerging bat (MET) was registered at the roost Tultschnig only in the years 2001 to 2003, and in all cases at the roost Schwarzenbach.

**Relative emergence time (minutes after sunset)** was defined as the time of the median emerging bat minus the corresponding sunset time. Sunset time for the roosts were gathered from the database of the Astronomical Applications department of the U.S. Naval Observatory (Washington D.C., USA; [www.usno.navy.mil](http://www.usno.navy.mil))

The following weather conditions were recorded at the beginning of each emergence: Temperature: with accuracy to  $\pm 1^\circ\text{C}$ ; Clouds: clear, overcast, heavy clouds; Rain: no rain, drizzle, rain; Wind: calm, gentle wind, strong wind.

To reduce the number of variables and to account for cross-correlations we performed a principal component analysis on the weather data. A factor analysis was carried out on the four variables, and the first two factors explained 72 % of the variance contained in the original matrix (Tab. 1).

The highest variables loading on factor 1 where 'clouds' and 'rain' which loaded positively and the variable 'temperature' had a negative factor loading. Thus, factor 1 represented a gradient from cloudy and rainy nights to evenings with high temperatures. Factor 2 is characterised by a gradient of evenings with high temperatures and wind to evenings with heavy clouds (Tab. 1).

To define the reproductive state of the females we used data from a study about the reproductive biology of lesser horseshoe bats in Austria (REITER 2004a). From there we perceived the birth dates of all young lesser horseshoe bats in the roost Schwarzenbach. For Tultschnig we used the

**Tab. 1:** Rotated factor matrix of the principal component analysis of the weather data.

Variables	factor A	factor B
Wind	-0.06	0.087
Temperature	-0.19	0.72
Clouds	0.81	-0.29
Rain	0.91	0.11
Cumulative % of variance	42.0	72.0
Eigenvalue	1,7	1,2

phenology data of the roost Althofen (Carinthia), which is approximately 30 km apart from Tultschnig. As the phenology of reproduction is more or less synchronised locally we are confident, that this is a good approximation reflecting the phenology of reproduction at roost Tultschnig.

We arbitrarily defined the last three weeks of the pregnancy as 'late pregnancy' and, according to GAISLER (1966) and SCHOFIELD (1996), 75 days were used as the duration of the whole pregnancy.

The categories were defined as follows:

Early pregnancy –	begin = median birth date – 75 days end = median birth date – 21 days
Late pregnancy –	begin = median birth date – 20 days end = median birth date
Lactation –	begin = median birth date end = time of the median birth date + 28 days
Post lactation –	begin = time of the median birth date + 29 days

In 2002 and 2003 the presence or absence of potential predators, such as cats, owls or birds of prey was recorded at both roosts.

### 2.3. Artificial hedgerow – experimental design:

To test the predictions mentioned above we erected an experimental temporal hedgerow at Schwarzenbach church (Salzburg) on July 11<sup>th</sup> 2002, i.e. on day 5 of the 9 day observation period. It was erected at the flight path 'roof void, short' (Fig. 1). Hence, this flight path was transformed into the shortest path along vegetation which was formerly the flight path 'roof void, long'. Therefore this flight path was assumed to be used by the bats as the main flight path after some time. The proportion of lesser horseshoe bats using the different emergence routes at the roost Schwarzenbach had been studied in the previous years, e.g. 2000-2001 (REITER 2002).

The artificial hedgerow was created by means of three big plastic puckets filled with water, stones and branches of various deciduous trees and bushes (*Prunus avium*, *Salix* spp., *Alnus incana*, *Corylus avellana*). They resulted in a single hedgerow line with a maximum height of approximately four meters and a minimum height of approximately one meter. Branches of the hedgerow reached the wall of the church as well as a lime tree where all flight paths from the roof void meet (Fig. 1). The length of the flight path 'roof void, short' (i.e. = the hedgerow line) was 12 meter from the wall of the church to the lime tree.

Two observers recorded the flight path of every individual bat, the emergence variables defined above and, additionally, the following parameters:

Light intensity (lux): measured with a luxmeter (Profifix®, Gossen) vertical against the sky.

Minimum height of flight (centimeter): it was measured at the flight path 'roof void, short' with an erected pole, which was marked at intervals of 25 cm. The minimum height of flight was then related to the nearest marking.

## 2.4. Clustering behaviour:

To test for clustering in the emergence behaviour, we used the program CLUSTAN (J.R. SPEAKMAN, Department of Zoology, University of Aberdeen, UK), and analysed the data of 8 nights at the roost Schwarzenbach. The recorded tapes were transcribed with second accuracy, using a stop watch and the program TIMER (J.R. SPEAKMAN, Department of Zoology, University of Aberdeen, UK). The software compares the observed distribution of intervals between two consecutive individual emergence events with the distribution expected for random emergence and tested the significance of this difference using Chi<sup>2</sup> and G-test (see SPEAKMAN et al. 1992, SPEAKMAN 1993). As the number of emerging bats ranged only from 62-84 individuals, we did not correct for the number of emerging bats as described by SPEAKMAN et al. (1999).

Results are presented as box-and-whisker plots, whereby the line across the box indicates the median and the box the interquartile range (i.e. 50% of the data). The whiskers extend to the highest and lowest value, excluding outliers and extremes. Outliers between 1.5 box lengths and 3 box lengths from the end of the box are indicated by a circle. Extremes are more than 3 box lengths from the end of the box and indicated by an asterisk.

## 3. Results:

### 3.1. Variation in the timing of the emergence:

Lesser horseshoe bats leave the roost in Tultschnig on average 26 min after sunset ( $\pm$  6.1 STD, minimum = 12 min, maximum = 38 min,  $n = 47$ ; time of the median emerging bat), whilst the mean emergence time at Schwarzenbach was 33 min after sunset ( $\pm$  6.4 STD, minimum = 22, maximum = 40 min,  $n = 9$ ). The timing of the nightly emergence in Tultschnig was overall correlated with sunset time (Pearson's correlation coefficient for the roost Tultschnig:  $r = 0.98$ ,  $n = 47$ ,  $p < 0.001$ , Fig. 2).

The reproductive state of the females as well as weather conditions had a significant influence on the relative emergence time of lesser horseshoe bats (ANCOVA for the roost Tultschnig: reproduction:  $F_{3,39} = 7.1$ ,  $p < 0.001$ ; factor 1 of the weather data:  $F_{1,39} = 20.3$ ,  $p < 0.001$ ; factor 2 of the weather data:  $F_{1,39} = 11.1$ ,  $p = 0.002$ ). Emergence was comparatively earlier at the onset of pregnancy and was latest during late pregnancy (Fig. 3). Lactating females left the roost again earlier and the earliest emergence was noted during the time of post lactating (Fig. 3). Emergence was furthermore earlier on nights with heavy clouds and rain compared to nights with high temperatures and strong wind (Spearman Rank Correlation of median emergence and factor 1 of the PCA:  $r_s = -0.56$ ,  $n = 40$ ,  $p < 0.001$  and factor 2 of the PCA:  $r_s = 0.40$ ,  $n = 40$ ,  $p = 0.012$ ).

At the roost Schwarzenbach we tested 4 days post-manipulation versus 4 days pre-manipulation. Bats tended to emerge on average 6 min earlier with the artificial hedgerow present (Tab. 2). No difference could be found for the light level and the duration of the emergence (Tab. 2).

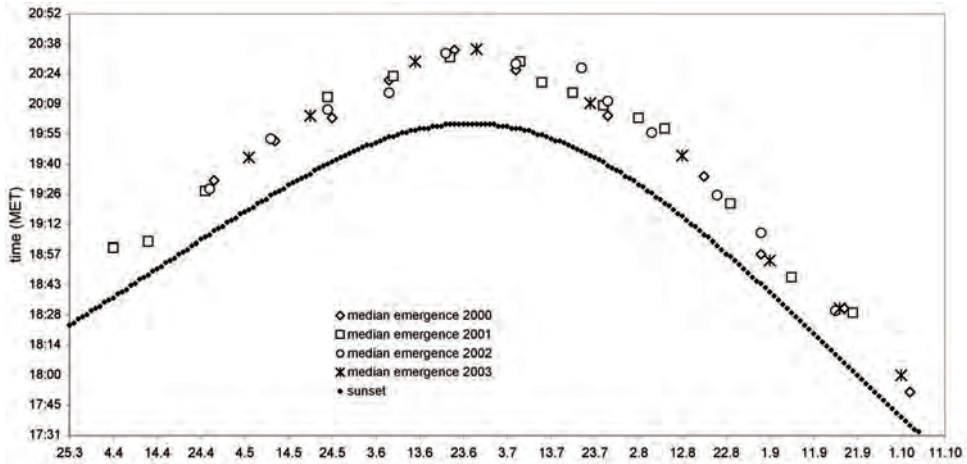


Fig. 2: Emergence of the median bat in relation to sunset at the roost Tultschnig.

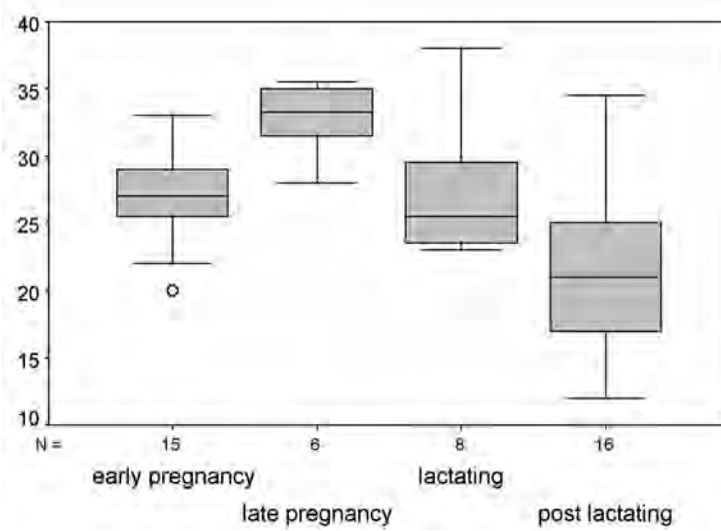


Fig. 3: Median emergence time (relative to sunset) and reproductive state of the females at roost Tultschnig. Note that the state of post lactating includes juveniles.

Tab. 2: Variables describing the emergence patterns before and after the erection of the artificial hedgerow (Values  $\pm$  STD).

	median emergence min after sunset)	mean duration of emergence (min)	mean light level (lux)
pre-manipulation	36.3 $\pm$ 5.5	33.0 $\pm$ 4.8	4.6 $\pm$ 0.9
post-manipulation	30.0 $\pm$ 3.6	34.0 $\pm$ 3.1	4.7 $\pm$ 0.9
Mann-Whitney-U-test	U = 2.0, p = 0.11	U = 4.0, p = 0.63	U = 8.0, p = 1.0



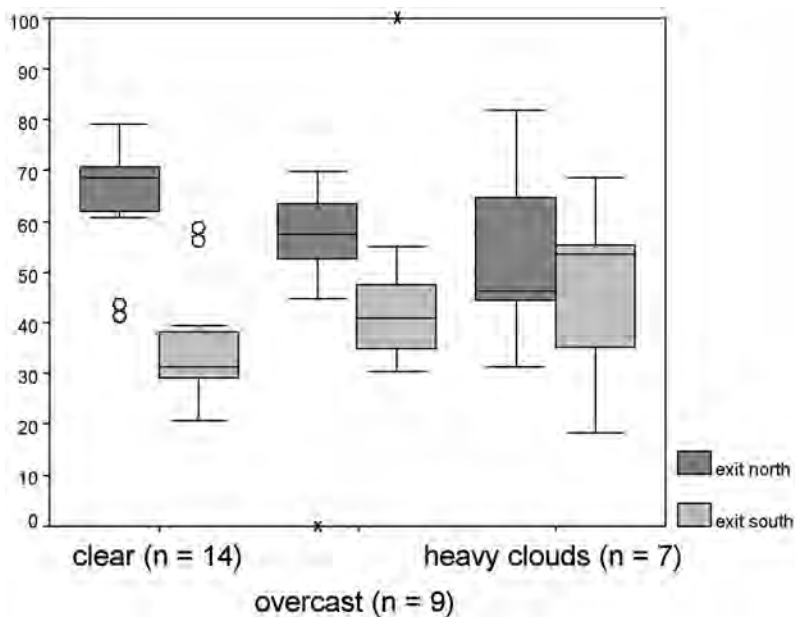
### 3.2. Clustering in the emergence pattern:

Only in two out of eight analysed nights the emergence pattern at the roost Schwarzenbach was significantly clustered. On both evenings with a clustered emergence, cats and on one evening even a hobby (*Falco subbuteo*) was present. In contrast, cats were only present once during the 6 evenings without a clustered emergence pattern.

### 3.3. Variation in the use of exit points and emergence routes:

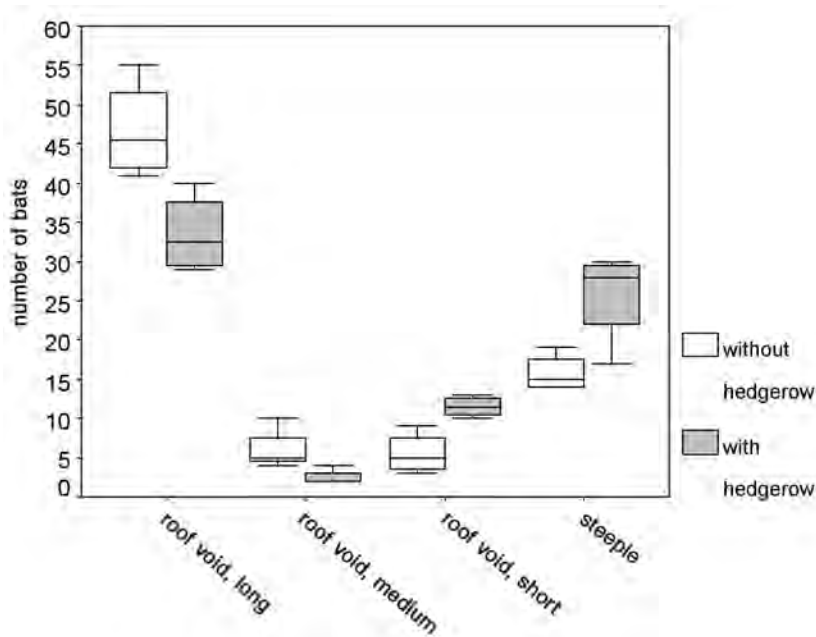
For the roost Tultschnig the relative use of the two exits depended on the coverage with clouds (ANCOVA for the difference between the number at the northern exit vs. southern exit: clouds:  $F_{2,30} = 5.1$ ,  $p = 0.012$ ; reproductive state:  $F_{3,30} = 1.7$ ,  $p = 0.19$ ; total number:  $F_{1,30} = 4.6$ ,  $p = 0.039$ ; Fig. 4). On clear nights the bats used mainly the northern exit point, whilst in nights with heavy clouds, more bats used the southern exit point (Fig. 4). The number of bats using the different emergence routes at the roost Schwarzenbach differed without and with the hedgerow present (Pearson  $\chi^2 = 33.2$ ,  $df = 3$ ,  $p < 0.001$ ; Fig. 5). More bats were noted at the emergence routes 'roof void, short' and 'steeple', whilst the use of the routes 'roof void, long' and 'roof void, medium' was less frequent after the erection of the hedgerow.

The height of flight path at the 'roof void, short' increased significantly with the hedgerow present (Mann-Whitney-U-test: 152.0,  $n = 66$ ,  $p < 0.001$ ), though the mean minimum height increased only from 0.7 m to 1.1 m.



**Fig. 4:** Proportion of bats emerging at the northern exit in relation to the southern exit at different degrees of cloudiness at roost Tultschnig.





**Fig. 5:** Proportion of bats using the different flight paths at roost Schwarzenbach before and with the presence of the artificial hedgerow (4 days before vs. 4 days after manipulation).

#### 4. Discussion:

##### 4.1. Variation of the emergence patterns in time:

The results of our study demonstrated that the nightly emergence of lesser horseshoe bats depends on a couple of factors affecting the exact timing of the emergence. Furthermore the results were in good accordance with the underlying hypothesis and other studies.

Sunset had the strongest influence on the emergence time (see as well ERKERT 1982), but additional factors modify the exact timing.

The lactating females in our study left the roost earlier than pregnant females. This has already been shown for *Eptesicus nilssonii* (DUVERGÉ et al. 2000). As lactation is very energy demanding (SPEAKMAN & RACEY 1987), lactating females could try to exploit the higher insect abundance at dusk despite the higher risks (DUVERGÉ et al. 2000). Moreover, pregnant females have much higher 'wing loadings' (NORBERG & RAYNER 1987), hence their agility and manoeuvrability is reduced (HUGHES & RAYNER 1991), resulting in a higher predation risk. This would predict a later emergence of pregnant females, as we found in our study.

After the time of lactation, lesser horseshoe bats leave the roosts earlier than during other reproductive states. A reason for this might be a poor nutritional condition of the females after lactation and therefore the need to leave the roosts earlier to exploit the higher insect abundance (see DUVERGÉ et al. 2000). On the other hand at that time of the year, bats born this year with their still inadequate flight performance are included in the samples and they should leave the roost comparatively later to reduce the predation risk.

The bats in this study reacted immediately to the artificial hedgerow in terms of space, but only a trend for an earlier emergence was recorded at the roost Schwarzenbach due to the presence of the hedgerow. Probably, lesser horseshoe bats need more time to adapt their behaviour to the new emergence conditions. However, it seems likely that optimised emergence conditions will result in an earlier and safer emergence. Subsequently this should have a positive influence on the fitness of individual bats (DUVERGÉ et al. 2000).

In contrast to other studies (e.g., SPEAKMAN et al. 1999, PETRZELKOVA & ZUKAL 2001) clustering in the emergence pattern was a rare event in the present study. Despite the long debate that has been going on about the functional significance of this behaviour, a clear answer is yet to be given. However, according to the anti-predator behaviour hypothesis later emerging species should display lower degrees of clustering because of the decreasing predation risk with decreasing light levels. Since the lesser horseshoe bat has a similar emergence time as *Pipistrellus pipistrellus* and *Eptesicus serotinus* (JONES & RYDELL 1994), both species with a high level of clustering (e.g. SPEAKMAN et al. 1999, PETRZELKOVA & ZUKAL 2001), there must be other factors influencing the degree of clustering in the different species.

#### **4.2. Variation of the emergence patterns in space:**

The changing use of alternative emergence routes at the roost Tultschnig with different levels of cloudiness could be interpreted as an anti-predator behaviour. At higher light levels (i.e. clear evenings) the bats used preferably the smaller, northern exit point with a safer emergence route into the next tree. However, at evenings with heavy clouds and hence lower light levels bats are able to use the bigger southern exit point, which has a more exposed emergence path into the vegetation.

Lesser horseshoe bats select exit points and emergence routes with low light levels along these routes (DUVERGÉ et al. 2000). However, the location and distance to the next patches of woodland might be also of importance for their selection (see REITER 2004b) and a combination of these factors is very likely.

The presence of the artificial hedgerow influenced the emergence behaviour of the lesser horseshoe bats as indicated by the shift in the use of the different emergence routes. This change is particularly significant since in a previous study we found that the different flight paths at this particular roost in Schwarzenbach were used in very constant proportions (REITER 2002). In detail we noticed a higher number of bats flying along the artificial

hedgerow accepting the hedgerow as a safer and shorter emergence path compared to the situation without the hedgerow present. On the other hand we also recorded more bats at the emergence route from the steeple. This might be interpreted as an avoidance of the new situation. However we expect increasingly more bats flying along the optimised route after the bats get used to the new conditions.

Although the artificial hedgerow started from the wall of the church, the height of the hedgerow didn't match the height of the exit point. Based on the behaviour of this species and the fact that the ultrasounds emitted are very high (AHLÉN 1988), the bats might have problems in recognising the new hedgerow. It is therefore likely that more bats would have used the new hedgerow if the vegetation was better connected to the exit point.

We conclude that optimised emergence conditions are likely to result in an earlier and safer emergence. This should positively influence the fitness of individual bats. For a 'threatened' species like the lesser horseshoe bat optimising their emergence routes might be a helpful conservation measure that could be achieved easily.

## 5. Zusammenfassung:

Wir untersuchten die Variabilität im abendlichen Ausflugsverhalten Kleiner Hufeisennasen (*Rhinolophus hipposideros*) an zwei Wochenstubenquartieren in den Bundesländern Kärnten und Salzburg (Österreich). Der Schwerpunkt lag auf Intra-Kolonie-Variabilität in Raum und Zeit. Zudem wurde ein Experiment durchgeführt, bei dem der Ausflugsweg durch die Errichtung einer temporären Hecke verändert wurde. Die Hecke sollte den Ausflugsweg optimieren. Die zentrale Hypothese der Studie ist, dass das abendliche Ausflugsverhalten von Fledermäusen einen Kompromiss darstellt: fliegende Insekten erreichen in der Dämmerung höchste Dichten, wenn sich die meisten Fledermäuse noch in den Quartieren befinden, das Prädationsrisiko sinkt hingegen mit fallenden Lichtintensitäten.

Die Ausflugszeit korrelierte stark mit dem Sonnenuntergang, hing aber zudem von der Witterung und dem Reproduktionszustand der Weibchen ab. Zunehmende Wolkenbedeckung resultierte in einem früheren Ausflug. Die Wolkenbedeckung beeinflusste aber auch die Nutzung zweier alternativer Ausflugsrouten an einem der beiden Quartiere. Säugende Weibchen flogen früher aus als trächtige und der früheste Ausflug konnte für post-laktierende Weibchen registriert werden.

Die Errichtung einer künstlichen Hecke in Schwarzenbach beeinflusste die Nutzung der unterschiedlichen Ausflugsrouten sowie die Flughöhe. Nach Errichtung der Hecke nutzten mehr Fledermäuse diese neue kürzere Flugroute sowie einen alternativen Ausflug, während die normale Ausflugsroute weniger stark benutzt wurde.

Optimierte Ausflugsbedingungen können in einem früheren und sichereren Ausflug resultieren, was einen positiven Einfluss auf die individuelle Fitness der Fledermäuse haben kann. Für eine gefährdete Art wie die Kleine Hufeisennase ist die Optimierung der

Ausflugsrouten eine praktikable und verhältnismäßig einfach durchzuführende Artenschutzmaßnahme.

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