

**CZECH UNIVERSITY OF LIFE SCIENCES PRAGUE**

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**The vocal repertoire of the captive sugar glider (*Petaurus breviceps*)**

**Master Thesis**

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## Declaration

„I hereby declare that this thesis, entitled Vocal repertoire of captive sugar glider is my own work and all the sources have been quoted and acknowledged by means of complete references.“

In Dalkeith, 23. 8. 2014

.....

Ondřej Sekera

## Acknowledgement

I would like to thank to my supervisor, Mgr. Irena Schneiderová, Ph.D. for helping me in every aspect of writing my Thesis. I also thank to all the sugar glider breeders, who allowed me to work with their animals and gain the data I needed, without their help my research would not be possible. I also would like to thank to all the teachers who taught me last two years, the knowledge I learned from them also helped me greatly while writing this Thesis. At last, I would like to thank to my family, without their never ending support I would not be able to finish my Masters education programme.

## Abstract

My Diploma Thesis is about the vocal repertoire of the sugar glider (*Petaurus breviceps*) in captivity. Sugar glider is a small nocturnal marsupial originally from Australia, where it lives in forest canopies. Because it is nice animal and can be easily kept in captivity, many people around the world keep it as a pet and it can be also seen in many zoos. Although some aspects of the sugar glider's lives have already been studied, there is only little known about their vocal communication. In my research, I observed four captive groups of sugar gliders and recorded their vocal communication at night, when they were active. I analysed those recordings, found out four singly produced vocalizations (bark, chatter, hiss and low hiss) and six combined vocalizations (bark-hiss, bark-low hiss, chatter-bark-hiss, chatter-bark-low hiss, chatter-bark and hiss-aggression). I also tried to estimate the behavioural context of those vocalizations according to my observations. I tried to compare my results about sugar glider's vocalizations with other research done on vocal repertoire of other gliders from family Petauridae. My research provided basic insight into captive sugar glider's vocal behaviour and helped in understanding of some aspects of proper sugar glider's keeping and breeding.

Key words: acoustic communication, bioacoustics, marsupials, sociality, vocal repertoire

## Souhrn

Má diplomová práce pojednává o hlasové komunikaci vakoveverky létavé (*Petaurus breviceps*) v péči člověka. Vakoveverky jsou malí noční vačnatci původem z Austrálie, kde obývají koruny stromů. Protože se jedná o atraktivní a relativně snadno chovatelné zvíře, mnoho lidí z celého světa si ho pořizuje jako domácího mazlíčka a vakoveverku je možné spatřit v expozicích mnoha zoologických zahrad. Přestože jsou vědecky zkoumány některé aspekty jejich života, jenom velmi málo se ví o jejich akustické komunikaci. Během mého výzkumu jsem pozoroval čtyři skupiny vakoveverek a nahrával jejich akustickou komunikaci během noci, kdy jsou aktivní. Získané nahrávky byly analyzovány a rozděleny do čtyř jednoduchých (bark, chatter, hiss and low hiss) a šesti kombinovaných kategorií vocalizací (bark-hiss, bark-low hiss, chatter-bark-hiss, chatter-bark-low hiss, chatter-bark and hiss-aggression). Také jsem se na základě pozorování snažil určit behaviorální kontext vokalizací. Pokusil jsem porovnat své výsledky s dosud zjištěnými fakty o vokalizacích ostatních druhů vakoveverek z čeledi Petauridae. Můj výzkum poskytl základní informace o vokálním repertoáru vakoveverky létavé a pomohl v porozumění některých aspektů správné péče o vakoveverky.

Klíčová slova: akustická komunikace, bioakustika, vačnatec, socialita, hlasový repertoár.

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# 1. Introduction

There has been great interest in mammal vocalization research. Many aspects of vocalizations has been studied, including for example behavioural context (e.g. Moody and Menzel, 1976; Aich *et al.*, 1990), acoustic structure (e.g. McConnell, 1990; Hauser, 1992) and their ontogeny (Elowson *et al.*, 1992; Schneiderová, 2014).

Sugar glider (*Petaurus breviceps*) is the smallest species within marsupial family Petauridae, which contains five species of small gliding possum and four non-gliding ones. Sugar gliders inhabit wide range of forested Australia, New Guinea and Tasmania. They are social animals, living in small family groups, males and females with offspring together in communal nests. Because sugar gliders are nocturnal and socially living mammals, vocal communication is expected to be more important for them than for example visual communication (Tyndale-Biscoe, 2005).

Regardless their nocturnal activity, sugar gliders became popular pets around the world and are present in many zoological gardens. There was some research done including sugar gliders biology, reproduction and behaviour, including brief and mostly onomatopoeic description of their vocal repertoire, but no more detailed study on acoustic structure of their vocalizations has been done yet (e.g. Smith, 1973; Kortner and Geiser, 2000; Shaw, 2004). Thus, the aim of my thesis was to describe the vocal repertoire of the sugar glider in captivity, exactly to determine how many different vocalization categories are present in vocal repertoire of this species, which are the basic physical characteristics of these vocalizations and in what behavioural context are they produced. This thesis should provide basic knowledge about sugar gliders vocal activity and its complexity for further research on this topic.

## **2. Aims of thesis**

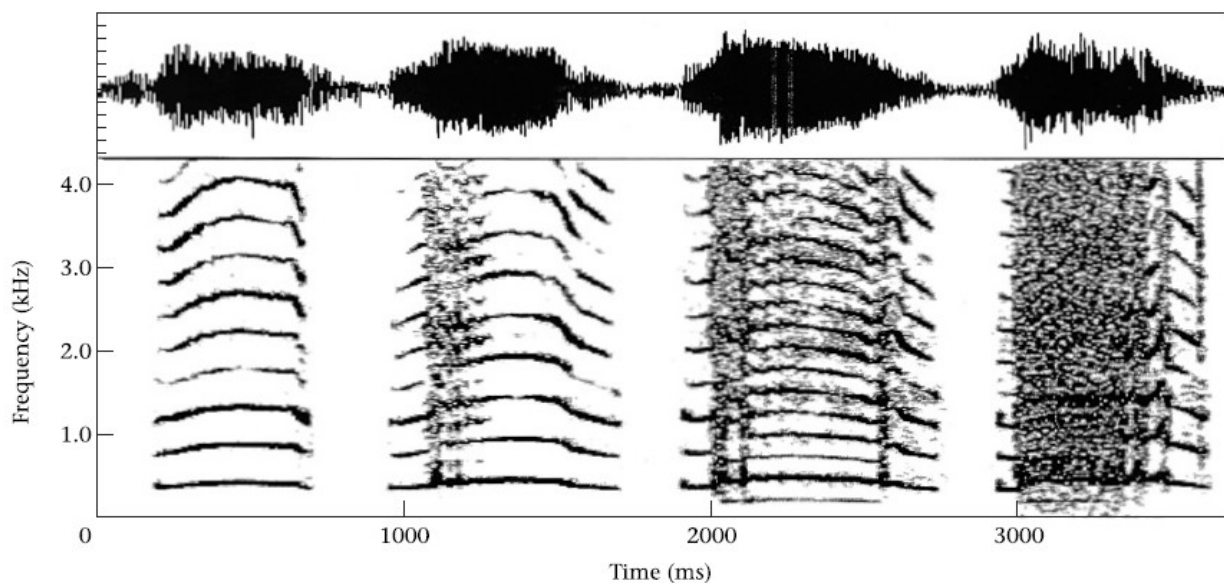
The proper knowledge of behaviour of certain species is one of the key factors (along with its diet) for successful breeding of that species. Although the sugar glider has become popular among hobby breeders and zoos all over the world, very little is known about its vocal communication, probably because of its night and secretive activity. Thus, the main aims of this thesis are to provide detailed description of its vocal repertoire, together with the description of behavioural contexts in which the described vocalizations are produced.



### 3. Literature review

#### 3.1 Production of vocalization in mammals

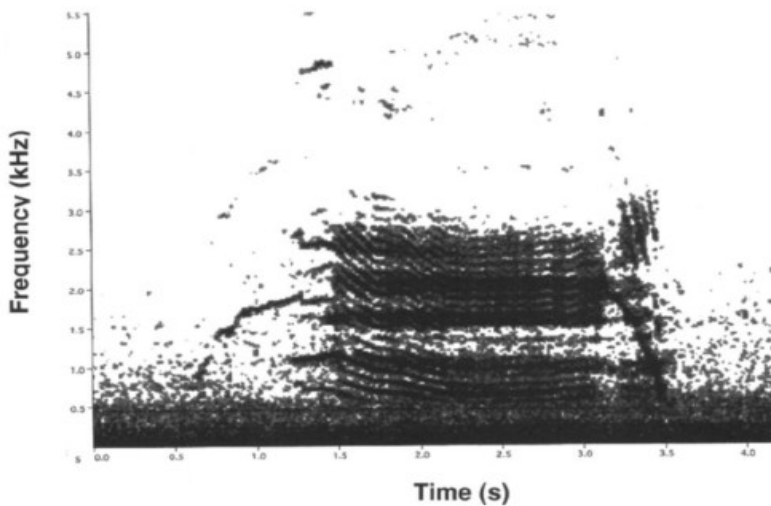
Production of vocalization is the conversion of air flow coming from lungs into acoustic energy. (Fitch and Houser, 2002) According to source – filter theory, vocal signals are produced in two stages. At first, glottal waves are produced in vocal folds (which is the source) and are modified, or filtered in supralaryngeal vocal tract (the filter) (Taylor and Reby, 2009). The opening and closing rate of vocal folds determines the fundamental frequency (F0) of vocalization. Thus, the fundamental frequency is determined primarily by the length and mass of vocal folds (the longer and heavier vocal folds vibrate at slower rate than smaller vocal folds) (Titze, 1994; Fitch, 1997). Many studies report presence of non-linear phenomena, such as bifurcations and deterministic chaos in source and filter components of mammalian vocalization (Taylor and Reby, 2009). This non-linear phenomena theory explains how complex and unpredictable vocalizations can occur in animals without complex neural control mechanisms. Because of this non-linear phenomena, rather simple neural commands can lead to highly complex and variable acoustic output. The non-linear phenomena may pose bulk of acoustic output from certain individuals and age classes of animals.



**Figure 1** Spectrograms of four vocalizations of adult Rhesus macaque (*Macaca mullata*). First vocalization is purely tonal, two following vocalizations contain sub-harmonics, while the last

vocalization shows presence of deterministic chaos (Fitch *et al.*, 2002).

The filtering process takes part in structures between vocal folds and the place where it radiates into environment, usually end of mouth and nose cavity. This structure, with all its cavities, is called vocal tract. This tract act as a set of bandpass filters, that selectively damper or enhance certain ranges of frequencies from vocal folds, according to its physical properties (Taylor and Reby, 2009). Those modified frequencies are called formants, which appears like spectral peaks (Fant, 1960; Titze 1994). In animals, the vocal tract is usually mostly rigid and its resonant properties are more static, thus predictable (Fitch, 1994).



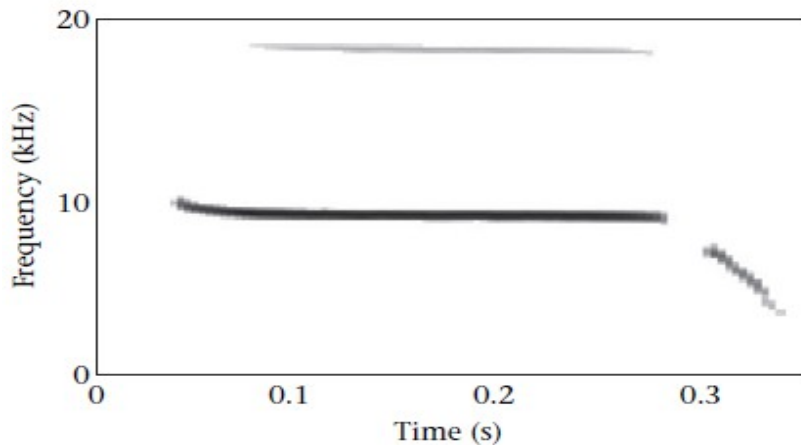
**Figure 2:** Spectrogram of aggressive call of Red deer (*Cervus elaphus*) with visible fundamental frequency and formants (Feighny *et al.*, 2006).

### **3.2 Acoustic structure of mammalian vocalizations**

Sounds, that animals emit are generally divided into three categories, based on their frequencies and audibility for human ear. Sounds, that human ear can hear, have their frequencies between 20 Hz up to 20 kHz (Pye and Langbauer, 1998). Sounds below 20 Hz are called infrasounds, and have been detected in communication of many animals, for example elephants (Payne *et al.*, 1986). Sounds above 20 kHz limit are called ultrasounds and are also vastly produced by many animals (rats, bats, cetaceans) for communication, orientation and as a tool to locate prey (Sales, 1972;

Samarra *et al.*, 2010).

As discussed in previous chapter, vocalizations can be also divided into tonal and noisy vocalizations according to their overall acoustic structure. Tonal vocalizations might be classified into vocalizations with constant fundamental frequency and vocalizations with modulated fundamental frequency (Fig. 3).



**Figure 3** Spectrogram of two vocalizations of Richardson ground squirrel (*Spermophilus richardsonii*). First vocalization has almost constant frequency, second vocalization with slightly modulated frequency (Sloan *et al.*, 2005).

There are several widely recognized theories about factors influencing acoustic structure of mammalian (Schleich and Bush, 2002). The effect of habitat influence on acoustic structure was proven for example by le Roux *et al.* (2002). Their study of two species of whistling rats, *Parotomys brandsii* and *P. littledalei*, shows that alarm vocalizations of *P. littledalei*, which inhabits denser habitats, are produced at lower frequencies than vocalizations of *P. brandsii*, which inhabits more opened habitats.

The change of acoustic structure with change of body mass was described by Morton (1977), who states that “frequency of an individual's voice negatively correlates with its body size.” Body size plays key role for individuals ability to fight and obtain resources. Because physical properties of vocal tract are more dependent on actual body size of individual than physical properties of larynx, spectral characteristics of formants are believed to be more honest about body size of individual

than characteristics of fundamental frequencies, especially on intraspecific level. Longer vocal tract produce lower and more closely spaced formants than shorter vocal tract. It was proven that certain animals respond to even slight changes in formant spacing frequencies, thus they are probably able to estimate a size of caller based on his formant characteristics (Charlton *et al.*, 2011).

Morton (1977) further proposed so called motivation – structure (MS) hypothesis, where he states that sounds produced by birds and mammals in hostile or aggressive behavioural context should be of low-frequency and wide-bandwidth, while sounds produced in fearful and appeasement behavioural context should be high frequency and tonal. Although considerable variation is presented within this hypothesis when tested, it is still considered to explain why some vocalization categories tend to be more tonal and some more wide-bandwidth (August and Anderson, 1987).

### **3.3 Behavioural context of mammalian vocalizations**

Animal vocalizations might be classified according their behavioural context. Widely used categories include territorial, advertisement, contact, mating, alarm, mobbing, distress, echolocation or food-associated vocalizations. Territorial calls are calls that serve the purpose of marking territory by signaling neighbours of sender where its territory lies (Goldingay and Possingham, 1995). Advertisement calls are calls emitted usually during courtship and its quality affect chance of individual to reproduce (Kime *et al.*, 1999). Contact calls are calls used during close contact of animals, for example during grooming (Snowdon and Cleveland, 1980). Similar the contact calls are sexual calls, but its use is limited to sexual behaviour of species (Reby and Charlton, 2012). Alarm calls are calls emitted in presence of danger and its function is to warn other individuals in the group, thus increasing their chance to survive whereas major function of mobbing calls is predator deterrence (Klump and Shalter, 1984). Distress calls are produced by captured prey, and they probably serve to distract predator in several ways (either attract other prey or other predators), which in consequences makes easier for captured prey to escape. Echolocation is special type of sound with high frequency, used to locate prey, obstacles and other individuals in condition with lower visibility, for example during night or under the water (Fenton and Bell, 1981). Food – associated vocalizations are calls produced in presence of food or during eating

(Hauser and Wrangham, 1987).

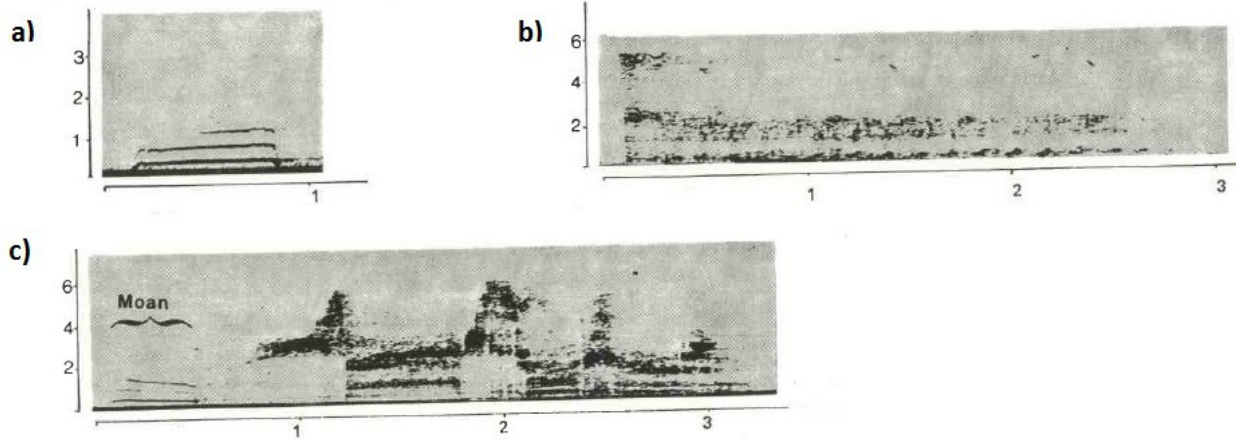
### **3.4 Vocalizations in marsupials**

Substantial qualitative evidence indicates more advanced level of sociality among this group, particularly in phalangerids and petaurids. Vocal communication of marsupials is varied and occur in number of social context (Kavanagh and Rohan-Jones, 1982).

During the research of eastern (*Macropus giganteus*) and western grey kangaroos (*Macropus fuliginosus*) behavioural repertoire (Coulson, 1996), several vocalizations were observed and their structure and behavioural context were studied. Those vocalizations included bucal clicking, produced by female when calling its offspring back to her pouch. Similar vocalization was produced by males when checking readiness of females to reproduce. Isolation vocalizations were produced by offspring when out of the pouch and searching for its mother. Those vocalizations were described as short, noisy and bleating, with three to four pulses and with most energy at 3 – 10 kHz in eastern grey kangaroo and without pulses in western grey kangaroo with most energy in 2 – 4 kHz in western grey kangaroo. There were two agonistic vocalizations: growl, produced by dominant individuals and cough, produced by subordinate individuals (Coulson, 1996).

There has been only brief description of sugar glider vocalization (e.g. Fleay, 1947; Russel, 1980; Kleinknecht, 1985; Kondratyeva *et al.*, 2006), however one relatively detailed study has been done by Kavanagh and Rohan-Jones (1982) on a closely relative species, the yellow-bellied glider (*Petaurus australis*). This study may give some insight on vocalization of the sugar glider, because of relatedness of those two species. In general, the sounds produced by the yellow-bellied glider can be characterized as short, frequently repeated units of wide frequency range with low band energy, with high intensity (loudness) and sharply sliding frequency shifts (cadence) (Fig.4), which proven to be carried best and on longest distances in the yellow-bellied's glider natural habitat. Kavanagh and Rohan-Jones (1982) observed and described six major calls in yellow-bellied glider: (1) *moan* is a low intensity, low frequency, monosyllabic call, used during take-off or landing upon another tree, might be a precursor of full call; (2) *gliding gurgle* – is generally a louder disyllabic call of higher frequency and longer duration than the moan, it is usually produced just after a take-off and lasts about half the flight; (3) *growl* – attenuated version of short call that is always low in

intensity and frequency with very short duration;(4) *full call* – noticeably greater in volume, duration and cadence than other vocalizations, consisted of at least two shrieks, which form highest frequency peaks; (5) *full call with beep* – call that begins with short component of high loudness and frequency and then continues as full call; (6) *short call* – similar to full call but with single shriek. None of these calls exceed 6.4 kHz, presence of ultrasonic sounds (over 20 kHz) was not studied, but was proven in other Petaurid species, the squirrel glider (*Petaurus norfolcensis*) by Kleinknecht (1985). Kavanagh and Rohan-Jones (1982) also state that during the research, some individuals vocalized more frequently than others. They observed that calls of one individual are frequently answered by calls of another glider, thus there is a theory that petaurids use acoustic calls to maintain contact during moving and foraging. Groups of animals were also observed to use the same routes and the same trees when approaching the nesting place. The onset of vocal communication among individuals followed the pattern of sunset, first sounds were mostly recorded 46 minutes after the sun went down. The vocal activity was uneven during the night, with the peak of vocalization taking time during early evening, shortly after leaving the den. Then the group communicates usually until middle of night, when vocal activity starts to decrease in call rates and around early morning there is almost no vocal activity at all. The vocal activity varies also during the year and it is clearly affected by season and weather. The yellow-bellied glider drops its vocal activity significantly during wet and cold months and increased during summer and autumn (Kavanagh and Rohan-Jones, 1982).



**Figure 4** Spectrograms of three different calls produced by the yellow- bellied glider: a) *moan*; b) *gliding gurgle* and c) *full call* with moan precursor (Kavanagh and Rohan-Jones, 1982)

Some description of Sugar glider vocalizations come from Fley (1947) and Russel (1980). They describe these vocalizations as “panting” or “conversational noises” in case of animals approached each other, but no recordings were made. Another description of Sugar glider vocalizations was made by Smith (1982), based on his observations: “The alarm call of nested young is interrupted hissing, that of adult is a repeated wok...wok...wok, somewhat like yapping of young terrier.” The anger call was described as turning over of high pitched starter motor. Both alarm call and anger call are full and loud, rapidly running down to faint grunts. Also, quiet hissing cries are sometimes given in the nest (Smith, 1982).

### **3.5 Biology of sugar glider**

Sugar glider is a small, arboreal, nocturnal marsupial that inhabits forest habitats of Australia, New Guinea and Tasmania. Sugar gliders consume mainly plant extrudates as a source of energy and insects living on those plants as a source of protein. They build their nests usually in hollow branches, spending most of their lives in tree canopies and only occasionally venture to the ground floor of forest (Tyndale-Biscoe, 2005). Sugar gliders are social animals, with tendency to stay together (Kleinknecht, 1985). In one communal nest, up to six males and females with offspring can be found. The dominant individual is always the male, and one or two dominant males perform most of social activities as odour distributing, territory patrolling and maintenance, chasing away members of different groups and mating (Smith, 1973). According to Tyndale-Biscoe (2005) in one, rarely two dominant males, are 20% heavier than other individuals in the group. The sex ration of sugar gliders is slightly in favour of females, because they enjoy the advantages of communal living and are not chased away when they turn to adults, as in case of young males. As many other nocturnal animals, sugar gliders have a complex chemical communication system (Smith, 1982). Kleinknecht (1985) states, that the distribution of odours among the group of sugar gliders by means of active and passive marking takes place mostly inside nest-box.

Sugar gliders shows strong family relationships. The behaviour of all animals depend on each other. The birth of a cub and its leaving the pouch plays very important role for the whole group. Kondratyeva *et al.* (2006) describes the introduction of animals and their sexual behaviour: The pair of newly introduced gliders quickly show social behaviour and begin to share same nest-box. Mouting and grooming precede sexual behaviour, which is consisted of androgenital grooming and male mounting. The mating takes place in nest-box, where the partners spent several hours. During following days, relationship between animals grows stable and pair formation is finished. The gestation period is only about 16 days long, such a short pregnancy being typical for the whole group of marsupials. No postnatal oestrus was observed. Pregnancy may be detected by state of pouch, which is pink with visible glands (it is not possible via female weight dynamics, because the weight of newborn is 0,19 g) (Kondratyeva *et al.*, 2006).

After the birth, cubs migrate to the pouch by themselves, attach to the teat and stay in this position



for approximately 40 days. Birth usually stimulates female to build „nest“ inside the nest box, if proper material is present (Kondratyeva *et al.*, 2006). Cubs start leaving the pouch when they are about 70 days old. They are still blind, bare and with poor thermoregulation. Two weeks after leaving the pouch for the first time, juveniles leave the nest-box for the first time, already with eyes opened and furry, but only on the back of female or male. Later on, they start to explore the cage on their own and playing behaviour can be noticed, as catching other animals by fore feet and chasing each other. Forty to forty five days after leaving the pouch, young animals are fully independent. The weaning stops completely when cubs reached 110 to 120 days of age. Short after the end of lactation period, female cycle is restored and mating and new litter production takes place. Paternal behaviour sometimes occur, male may be building the nest, carry the juveniles on their backs and respond to their calls (Kondratyeva *et al.*, 2006).

### **3.6 Housing and feeding sugar glider**

Because the research was done on animals kept as a pets by hobby breeders, it is important to describe keeping and breeding of the sugar glider as the artificial conditions may influence its behaviour and thus the vocal behaviour as well (absence of possibility of gliding for example). Also, the description of sugar glider's breeding and sexual and parental behaviour is included to better understand the behavioural context of some vocalizations.

Kondratyeva *at al.* (2006) described used housing for sugar glider as vertical cages 80 long, 40 cm wide and 130 cm long. The interior of the cage should include tree branches for climbing, at least two nest-boxes, several food bowls. The water must be provided all the time. Fresh food, which must be provided and changed daily may include fruit, insects, cottage cheese, boiled eggs, honey and so on.

Some sugar glider breeders I worked with suggested that the cage should come with a floor grating to minimize the contact gliders have with old food and faeces in the drop pan. It is good to have a variety of washable fabric items hung throughout the cage that allow for sleeping, lounging, and landing areas. Another option is to use perches, shelves, branches, and nest-boxes. It is possible to cover the floor with plastic square mesh to give the animals more places to hold, rest, and limit their ability to reach through to the drop pan. To improve the sugar glider welfare, the cage might

be enriched with spinning wheel and other objects, that increase its foraging and nesting activity.

### 3.7 Ownership of the sugar glider

The ownership of sugar glider may be regulated by the legislation, as described below. According to UICN Red List database, sugar glider is in category LC least concern (Salas L. *et al.*, 2008). It is not cited in any CITES appendixes.

No law regulations on possession of sugar glider has been found in Europe, Africa and South America. According to USDA (2014), laws are specific to different countries. Sugar glider ownership may legal at the country, state or province level, but could be illegal at the city or neighbourhood level. In USA, possession of sugar glider is restricted by the law, breeders need licence from USDA (United States Department of Agriculture) and in some states, for example California, Hawaii, Alaska, Minnesota and some others, is possession forbidden completely. The same situation is in Canada. (USDA, 2014) In its native land, Australia, one must have the appropriate license. These vary from state to state and in fact, in some states one may not keep protected species at all. Even with the appropriate licence, wildlife may not be taken from the wild. (AGDA, 2013) In Czech Republic, possession of sugar glider is not limited, nor any permission is needed and it is bred by some zoological gardens and several hobby and market breeders.

**Figure 5:** Suitable cage for small group of sugar gliders (images.google.com)



## 4. Methods and materials

### 4.1 Study groups & animals

Because no breeders association of sugar gliders is present in the Czech Republic, the breeders were found on internet websites, usually when selling juveniles, or through friends and colleagues who knew some breeders. I was able to develop a long term cooperation with three breeders, and was allowed to work with four groups of sugar gliders (Table 1). The group composition was always one male and one female, but the age of individuals varied. I was able to recognize each individual, based on its size and differences in coloration. These characteristics were described to me by breeders before the beginning of my observation.

**Group 1 ( Praha ):** This group consisted of two individuals, a young male (cca 9 months old) and an old female (8 years old). They were placed in a convertible cage, 140 cm high, 90 cm long and 50 cm deep. The bottom of the cage was solid, removable, filled with scobs and covered with bars to prevent contact with fallen food and droppings. Inside the cage, several branches for climbing were also placed. Two cloth nests were placed inside, together with four hanging bowls for food, placed in different heights and one bell bottle for water. Food was provided once a day in the evening and the leftovers were removed the next morning. It consisted of fresh fruit, pellets for gliders and worms. The temperature during recording period was cca 25 degree Celsius.

**Group 2 ( Praha ):** This group consisted of two young non-sibling individuals, a male and a female, both 6 months old,. The cage, in which they were recorded, was about 120 cm high, 50 cm deep and 90 cm long, and its floor was covered with scobs. Compared to other cages, this one had just one wooden sleeping box hanged on the cage and no branches inside. The water and food were placed in bowls on the ground. Gliders were fed once a day in the evening, mostly with glider pellets and fruit, no worms were provided. The temperature during recording period was cca 28 degree Celsius.

**Group 3 ( Jihlava ) :** This group consisted of two young individuals, one castrated male (cca 8 months old) and one female ( cca 3 months old). During the recording period, the animals were placed into cage 160 cm high, 60 cm deep and 80 cm long. The cage was equipped with ropes, branches and several hanging nests on different places. Food was given into two bowls placed around the middle

part, the bell bottle was also placed inside the cage. The food was provided once a day and it consisted of fruit, vegetables, glider pellets and worms. The temperature during recordings was around 22 degree Celsius. The cage was placed right next to a cage with group 4, thus the animals had visual and acoustic contact with each other.

**Group 4 ( Jihlava ):** This group consisted of two animals, one male and one female, both approximately 3 years old. The animals were owned by the same breeder as the group 3, thus the cage size, its equipment, and feeding time, food composition and temperature were the same as in the group 3.

## **4.2 Data collection**

All data collection were made at evening and at night (from 7 pm to 5 am) because of nocturnal activity of sugar gliders. The audio recordings were collected either in MP3 or PCM – 24 format. The recorder was set up on following parameters: Recording channel stereo ; sample rate 44.1k; LED level – 38 dB; Auto track 10 minutes ; Silent skip off; Microphone attention – 24 dB; Low cut off ; Input jack stereo; skip back 3 seconds. The video recordings were collected in format MP4 format using Canon LEGRIA HF R48 camcorder. The light conditions for video recorder were created first by white 11W bulb, later red 11W bulb.

Each group was observed *ad libitum*, with different amount and duration of observations, according to the opportunities given by breeders. Because there were observations and data collections made in different conditions and at different breeders, two different methods were used.

The first method was **direct observation** and was applied to groups 1 and 2. During direct observation, each group was constantly directly observed and its vocalizations were recorded. Additional information were written into notebook, including identification number of recording, time of vocalization, individual that produced the vocalization, behaviour of individual that produced the vocalization, and behaviour of other individuals in the group. The group 1 was observed for six nights, from 10th and 20th June 2013. Total recording time for this group was 14 hours and 10 minutes. The group 2 was observed for six nights, from 5th to 16th June 2014. Total recording time for this group was 26 hours and 35 minutes.

The second method was **indirect observation**, and was applied to groups 3 and 4. Data were

collected with the help of an audio recorder and camcorder, set up in the way, so the recordings were collected automatically during the night. The same information as in the case of direct observation were therefore extracted from obtained recordings and were written into notebook. Data were then collected by watching the videos and extracting vocalizations from recordings. Additional informations were written into notebook the same way as in the case of direct observation.

**Table 1:** Description of observed groups

ID	Location	Composition	Recording method	Date	Number of recording days	Total time of recording
Group 1	Prague	1,1,0	Direct observation	10.6. - 20.6. 2013	6	14 hours, 10 minutes
Group 2	Prague	1,1,0	Direct observation	6.6. -15.6. 2014	6	26 hours, 35 minutes
Group 3	Jihlava	1,1,0	Indirect observation	22.2. - 4.3. 2014	6	24 hours, 15 minutes
Group 4	Jihlava	1,1,0	Indirect observation	22.2. - 4.3. 2014	6	24 hours, 15 minutes

## 4.3 Data analysis

### 4.3.1 Vocalization categories

Obtained recordings were visualized and analysed in Avisoft SASlab Pro software. Recorded vocalizations produced by sugar gliders were classified into four single vocalization categories, bark, hiss, chatter and low hiss, according to their overall acoustic structure. Each described category was further characterized according to its acoustic structure. Firstly, it was described whether the vocalization is tonal or noisy. Tonal vocalizations can be defined as those that have fundamental frequency ( $f_0$ ) and its harmonics ( $f_1$ ,  $f_2$ ,  $f_3$ , etc.) whereas noisy vocalizations lack such a structure. (Bradbury and Vehrencamp, 1998) Secondly, it was described whether the vocalization consists of one single unit or it was a short bout of two to six repeated units or a long bout of more than six repeated units.

Some vocalizations also regularly occurred in combination with other vocalizations, therefore additional ***combined vocalization categories*** formed by these frequently emitted combinations were defined, bark – hiss, bark – low hiss, chatter – bark – hiss, chatter – bark- low hiss, and chatter – bark. There was also one vocalization, recorded only in form of combination, defined as hiss – aggression.

### 4.3.2 One-zero sampling

One-zero sampling method was performed in order to find out how are the sugar gliders vocally active during the night. One – zero sampling is quick method for scoring activities on check – sheets. (Bateson and Martin, 1993). The recording is divided into short intervals. During each interval, it is recorded whether or not the behaviour pattern occurred, irrespective of how often, or for how long, the behavioural pattern occurred during the interval. The score obtained by one-zero sampling is expressed as a proportion of all sample intervals, during which the behavioural pattern occurred. It gives a single, dimensionless score. One-zero sampling allows to measure large amount of categories and provides relatively high inter- and intra-observer reliability. The disadvantage of one – zero sampling is that when wrong length of interval is chosen, the frequency of behavioural patterns might be biased both ways. The size of interval used in time sampling depends on how many categories were defined, as well as nature of behaviour. The shorter sample the more accurate relative frequency is obtained. However, the shorter time interval, the more difficult is to record several behavioural patterns at once. (Bateson and Martin, 1993) For analysing gliders vocal behaviour using one-zero sampling method, the continual recordings were divided into several five-minute intervals. This interval duration was chosen according to the fact that gliders do not vocalise very often together with the fact, that when they do, single vocalization can last a relatively long time. The whole recording was analysed. When specific vocalization was present, number 1 was scored, when specific vocalization was absent, number 0 was scored. This way, an approximate frequency of usage of every vocalization type was evaluated.

#### ***4.4 Measured Parameters definition***

**Duration of bout** is a parameter measured only for vocalizations produced in bouts and it indicates the duration of whole bout, from the beginning to the end. **Number of syllables** shows of how many syllables is the vocalization consisted. **Distance to maximum** is parameter, that shows the time within the vocalization, where the highest amplitude was reached. **Duration of syllable** states the duration from point where the syllable started to point where it is terminated. **Interval** is parameter, that indicates the duration between the beginning of one syllable to the beginning of a next one, in case that the vocalization is produced in bout. **Fundamental frequency  $f_0$**  indicates mean fundamental frequency of vocalization, and it is measurable only for tonal vocalizations. For wide-bandwidth vocalizations, **mean 50%** parameter was measured instead. This parameter indicates at which frequency the vocalizations energy reached 50% of its total energy. **Peak frequency** indicates frequency with maximal amplitude of vocalization. For **duration of bout**, **duration of syllable**, **distance to maximum** and the **interval**, the values were in seconds (s). For **fundamental frequency**, **mean 50%** and **peak frequency**, the values were in Hertz (Hz).

#### ***4.5 Measuring of physical parameters***

The minimum of four and maximum of 52 vocalizations was selected, depending on vocal category to measure the acoustic parameters. The number of measured parameters depended on vocalization category, because some of them was not possible to measure for some categories.

The vocalizations selected for measuring the parameters had to meet several conditions. Firstly, they needed to be classified as single vocalization category, that means that it should belong to bark, chatter, hiss or low hiss categories. The only exception was hiss – aggression, where it was impossible to find just “aggression” part, so this part was cut from its combination and measured separately. Secondly, only high quality vocalizations not overlapped with background noises with little or no background noises were chosen. All the parameters for single syllable were measured via specific software function.

For single vocalizations, number of syllables was always equal one, distance to maximum was measured together with duration of syllable, fundamental frequency or mean 50% and peak frequency.

For short bout vocalizations (two to six syllables), together with parameters measured in single vocalizations, duration of bout and interval were also measured in the way that each syllable was measured independently and then arithmetic average was taken from obtained values.

For long bout vocalizations (more than six syllables), the same parameters were measured as for short bout vocalizations. Because it would be very demanding to measure all the syllables, syllable with maximum amplitude was chosen with help of acoustic parameter distance to maximum. From this syllable, three forgoing and three subsequent syllables were chosen and acoustic parameters were measured from all these seven syllables. Arithmetic average was then taken as in case of short bout vocalizations.

## **4.6 Statistical analysis**

Results of descriptive statistics are given as  $\bar{x} \pm SD$ . Relative occurrence of empty intervals (intervals without vocalization) and active intervals (intervals with at least one vocalization) was calculated. Differences in vocal activity of the studied groups were tested using one-way ANOVA, where the relative occurrence of active intervals was the variable and group was the factor. For each group, mean number of single vocal categories and combined vocal categories was compared using a paired t-test. Parametrical tests could be used as the data did not departed from normality (Kolmogorov – Smirnov test) and homoscedasticity (Levene's test).



## 5. Results

### ***5.1 Structural variability of vocalizations emitted by sugar glider***

Vocalizations emitted by sugar gliders could be divided into four categories, based on their structural variability. These vocal categories were bark, hiss, low hiss and chatter. Spectrograms of these categories are shown in Figure 6 and their descriptive statistics are presented in Table 2.

#### **Bark vocalization (Fig.6 (a))**

Bark vocalization can be characterized as a tonal, rapid and loud, with average fundamental frequency  $803.67 \text{ Hz} \pm 596.45 \text{ Hz}$ , produced either as single syllable or in short or long bouts. Average bout duration was  $11.17 \pm 14.18 \text{ s}$  with syllable duration around  $0.21 \pm 0.23 \text{ s}$  and interval around  $1.1 \pm 0.34 \text{ s}$ . This vocalization was produced both by males and females among all studied groups. When producing bark vocalization, sugar glider were always out of nest-box, usually on some higher place, with its head slightly raised.

#### **Hiss vocalization (Fig. 6 (b))**

Hiss vocalization can be described as wide-bandwidth, short vocalization (average duration of syllable  $0.139 \pm 0.118 \text{ s}$ ), with highest average peak frequency of all measured vocal categories ( $7359.42 \pm 4545.15 \text{ Hz}$ ). It was produced either as single syllable or in short bouts, very often when both animals were inside nest-box together or when animals were approaching each other. During conflict between animals, this vocalization sometimes escalated into hiss-aggression vocalization, adding one or more aggression vocalization units between repeated hiss syllables.

#### **Low hiss vocalization (Fig.6(c))**

Low hiss vocalization is resembling hiss vocalization, also wide-bandwidth, also produced singly or in short bouts, but with longer average syllable duration ( $0.387 \pm 0.137 \text{ s}$ ) than hiss and also with lower average peak frequency ( $2866 \pm 4525.15 \text{ Hz}$ ). Behavioural context of this vocalization is similar to

hiss, most of low hissing was produced inside nest-box or during the contact of both animals.

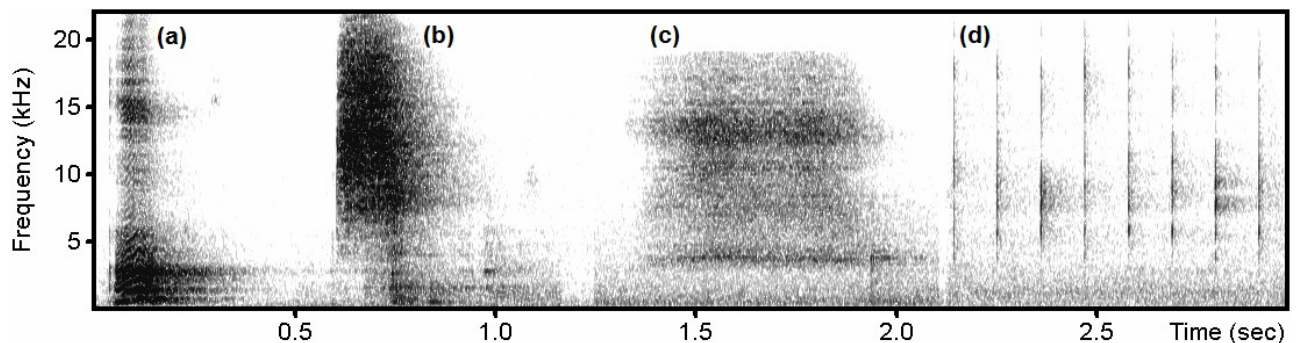
### Chatter vocalization (Fig. 6 (d))

Chatter vocalization can be described as low intensity, wide-bandwidth sound, produced only in bouts in very rapid succession, with short and very stable intervals of  $0,116 \pm 0,01$  s. Its average peak frequency reached  $4716,68 \pm 4129,47$  Hz, but was highly variable. Behavioural context of this vocalization is unknown, it was produced during variety of occasions, inside or outside nest-box alike. On some occasions, bark and hiss vocalizations were preceded or followed (or both) by chatter.

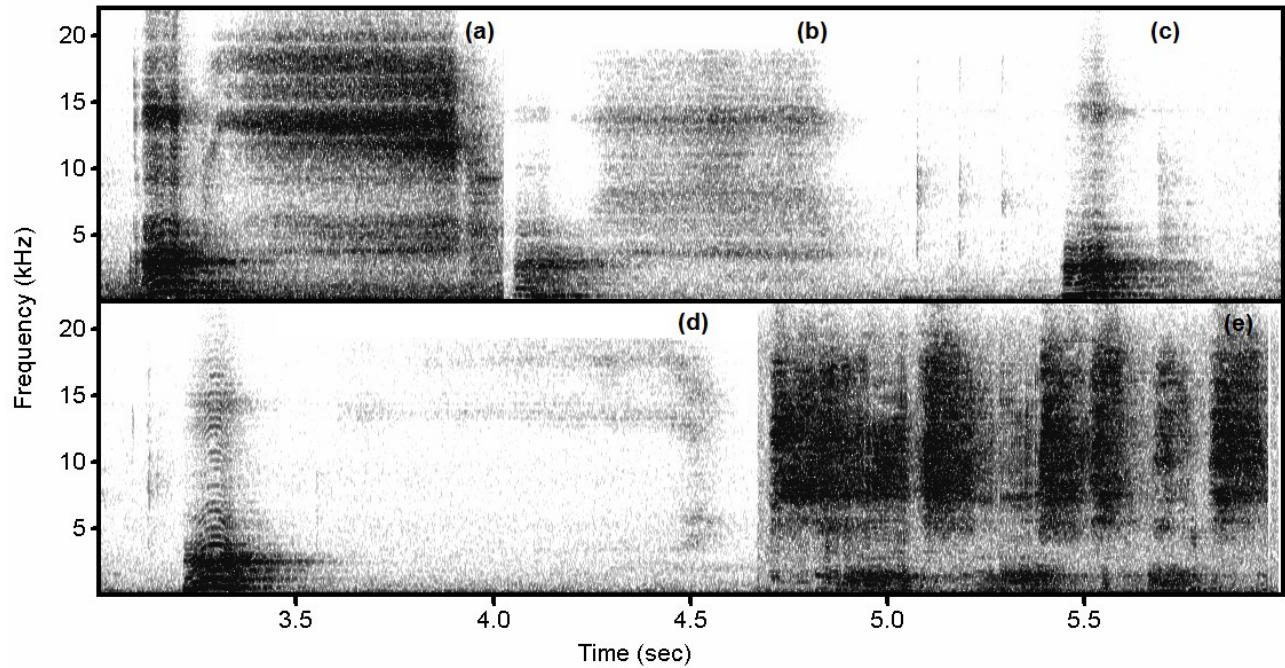
Apart from singly produced vocalizations, combined vocalizations were also emitted by sugar gliders. These vocalizations consisted of two or more different vocalization categories listed above (and aggression vocalization, described below), which immediately followed each other. For these combined vocalizations, no descriptive statistics were measured, except hiss-aggression, where the descriptive statistics were measured for aggression component.

### Hiss-aggression vocalization (Fig. 7 (e))

Hiss-aggression vocalization is combined wide-bandwidth vocalization, consisted of hiss vocalization and aggression component. Because this aggression component was overlapping with with hiss component, it was impossible to measure any of its properties apart from average distance to maximum ( $1,91 \pm 0,07$  s), average 50% quartil ( $7768,75 \pm 2121,2$  Hz) and average peak frequency ( $3150 \pm 4411,91$  Hz). The behavioural context of this vocalization is clearly agonistic, produced only during fight by both individuals at the same time.



**Figure 6:** Spectrograms of singly produced vocalizations: a) bark b) hiss c) low hiss d) chatter



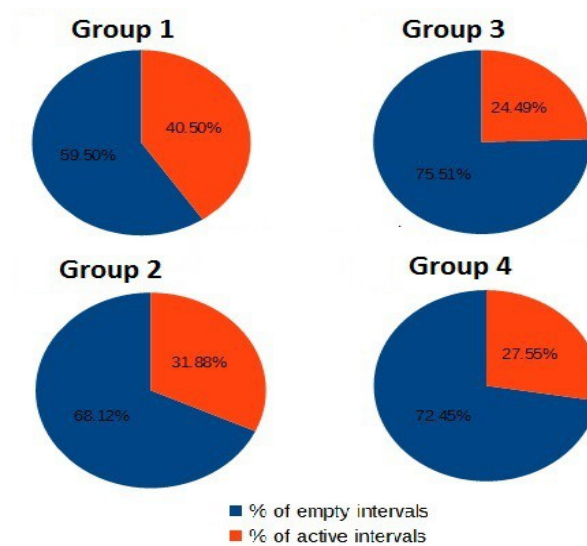
**Figure 7:** Spectrograms of combined vocalizations: a) bark hiss b) bark low hiss c) chatter bark d) chatter bark hiss and e) hiss-aggression

**Table 2:** Descriptive statistics of measured acoustic parameters for each single vocal category

Acoustic parameter	Vocalization									
	<i>n</i>	bark	<i>n</i>	hiss	<i>n</i>	low hiss	<i>n</i>	chatter	<i>n</i>	aggression
dur b (s)	16	11,17 ± 14,18	21	0,928 ± 0,616	4	1,65 ± 0,875	6	5,35 ± 2,62		not measured
dur s (s)	30	0,21 ± 0,23	52	0,139 ± 0,118	5	0,387 ± 0,137		not measured		not measured
int (s)	16	1,09 ± 0,34	21	0,428 ± 0,263	4	0,683 ± 0,124	9	0,116 ± 0,01		not measured
disttomax (s)	30	2,75 ± 5,87	52	0,1 ± 0,17	5	0,76 ± 0,726	9	1,91 ± 1,07	8	0,11 ± 0,07
mean f0 (Hz)	30	803,67 ± 596,45		not measured		not measured		not measured		not measured
50% quartil (Hz)		not measured	52	9270,67 ± 3093,49	5	5264,00 ± 2916,96	9	6692,22 ± 3208,78	8	7768,75 ± 2121,2
peak f (Hz)	30	1022,33 ± 924,65	52	7359,42 ± 4545,15	5	2866,00 ± 4525,15	9	4716,68 ± 4129,47	8	3150,00 ± 4411,91

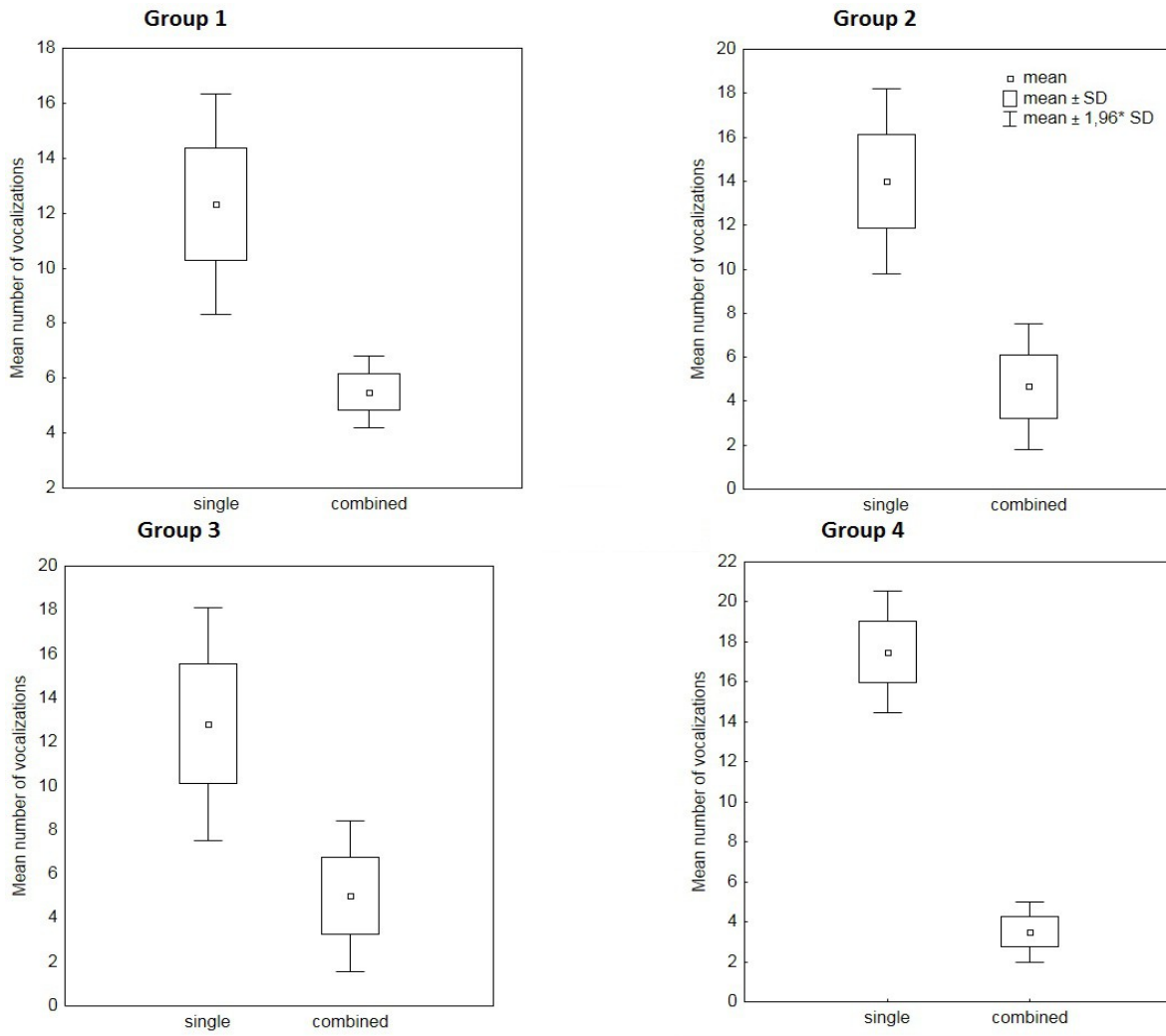
## 5.2 Vocal activity of sugar gliders

In Group 1, there were 168 intervals recorded, and 100 (59,5%) were empty, whereas 68 (40,5%) contained at least one vocalization category. In Group 2, there were 320 intervals recorded, 218 (68,12%) of them were empty, whereas 102 (31,88%) contained at least one vocalization category. In Group 3, there were 294 intervals recorded, 222 (75,51%) of them were empty, whereas 72 (24,49%) contained at least one vocalization category. In Group 4, there were 294 intervals recorded, 213 (72,45%) of them were empty, whereas 81 (27,55%) contained at least one vocalization category. There was no difference in vocal activity in studied sugar glider groups (one-way ANOVA:  $F_{(3,20)} = 2,66$ ;  $p = 0,08$ ).



**Figure 8:** The relative occurrence of empty intervals and active intervals, i.e. intervals containing at least one vocal category, in each of studied groups.

All of studied groups were producing both singly and combined vocalizations. However, certain vocal categories may be missing in some groups. There was no difference in use of combined and singly produced vocalizations, all groups produced fairly less combined vocalizations than singly produced ones (group 1:  $t_{(5)} = 2,86$ ,  $p < 0,05$ ; group 2:  $t_{(5)} = 4,23$ ,  $p < 0,05$ ; group 3:  $t_{(5)} = 4,00$ ,  $p < 0,05$ ; group 4:  $t_{(5)} = 6,89$ ,  $p < 0,05$ ) (Figure 1.2).



**Figure 9:** Mean numbers of single and combined vocalizations for each of studied groups

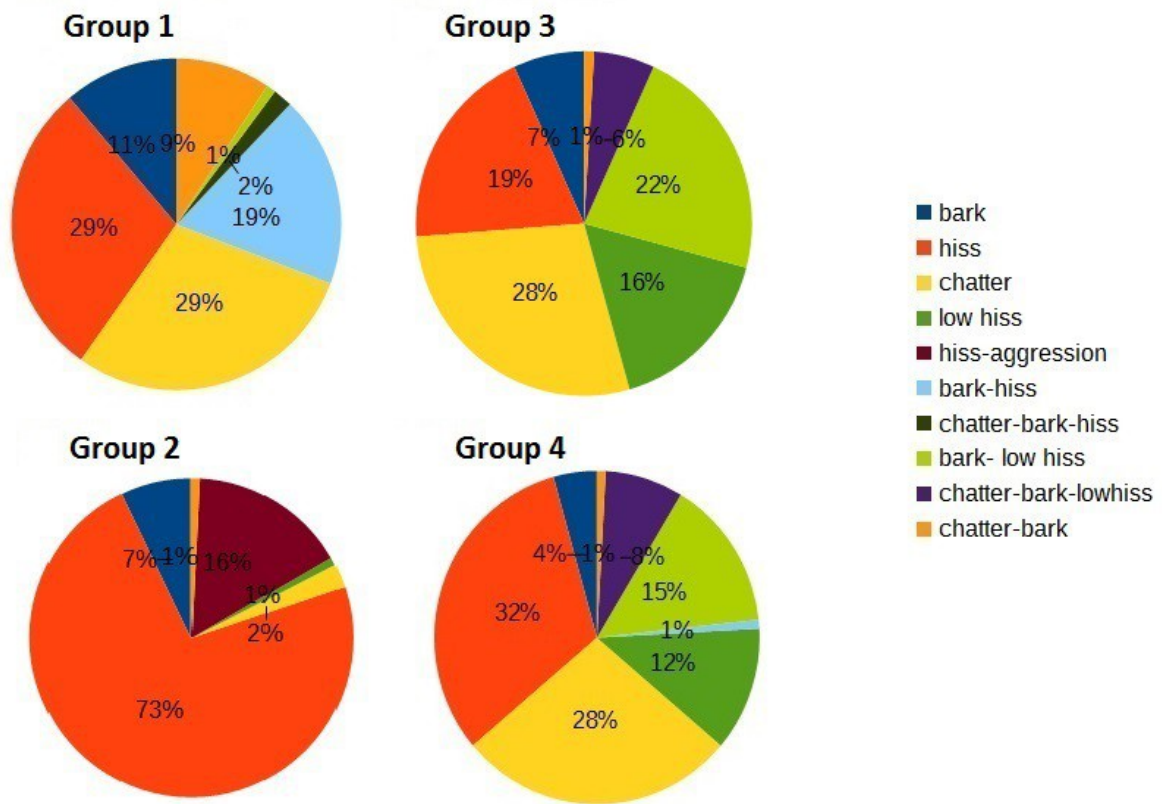
Total number of 107 occurrences of vocal categories was noted in Group 1. Bark was noted 12 times (11.2%), hiss was noted 31 times (29.0%), chatter was noted 31 times (29.0%), chatter – bark was noted 10 (9.3%), chatter- bark- hiss was noted twice (1.9%) and bark- low hiss was noted once (0.9) Other vocal categories were not noted in this group.

Total number of 126 occurrences of vocal categories was noted in Group 2. Bark was noted 9 times

(7.14%), hiss was noted 92 times (73.2%), chatter was noted 3 times (2.38%), low hiss was noted once (0.79%), hiss aggression was noted 20 times (15.87%), chatter – bark was noted once (0.79%). Other vocal categories were not noted in this group.

Total number of 103 occurrences of vocal categories was noted in Group 3. Bark was noted 7 times (6.8%), hiss was noted 20 times (19.42%), chatter was noted 29 times (28.16%), low hiss was noted 17 times (16.5%), chatter – bark was noted once (0.97%) bark – low hiss was noted 23 times (22.33%), chatter – bark – low hiss was noted 6 times (5.83%) Other vocal categories were not noted by this group.

Total number of 116 occurrences of vocal categories was noted in Group 4. Bark was noted 5 times (4.31%), hiss was noted 37 times (31.90%), chatter was noted 32 times (27.59%), low hiss was noted 14 times (12.7%), bark – hiss was noted once (0.86%) , chatter – bark was noted also once (0.86%) bark – low hiss was noted 17 times (14.66%), chatter – bark – low hiss was noted 9 times (7.76%) Other vocal categories were not noted in this group.



**Figure 10:** The relative occurrence in vocalization categories in all studied groups.

## 6. Discussion

This Thesis describes structural variability of vocalizations emitted by four pairs of sugar glider kept in captivity. Living conditions in captivity are different from conditions in wildlife. Thus, it is possible that some vocal categories, that normally occur among wild populations may be missing in vocal repertoire of captive sugar gliders. This might include vocalizations accompanying gliding (Kavanagh and Rohan-Jones, 1982) or presence of predator (alarm calls). Normal composition of a group of sugar gliders in wild includes more individuals than just two, so this fact may also lead to more limited vocal repertoire observed in my Thesis. No offspring were present in any of the studied groups, so vocalizations produced by parents towards offspring and vice versa are missing, too.

During this Thesis, I was not directly testing dependence of vocalization onset on light conditions. However, my observations corresponds with findings of Kavanagh and Rohan-Jones (1982) on other species from Petauridae family, yellow-bellied glider. They states that the onset of vocal activity is predictable and changes with changes of daylight, and that yellow-bellied gliders began their vocal activity approximately 46 minutes after sunset. During my observations, I noticed that the onset of vocal activity of studied sugar gliders began within an hour after the light conditions changed from light to dark. More research would needed to be done to prove the dependence of onset of vocal activity on light conditions.

Four single vocal categories (bark, hiss, low hiss and chatter) were identified as well as six combined vocal categories (hiss-aggression, chatter-bark, bark-hiss, bark-low hiss, chatter-bark-hiss and chatter-bark-low hiss). The vocal diversity generally corresponds with diversity of the yellow-bellied glider vocal repertoire, where six major vocal categories were described. Combinations of vocalizations were also observed in yellow-bellied glider. The yellow-bellied glider's full call, described by Kavanagh and Rohan-Jones (1982), may be homologous to sugar glider's bark in terms of structure and behavioural context, but some major differences were found. Both vocalizations can be characterized as loud and rapid, produced when animals are stationary on a tree, but full call of yellow-bellied glider has at least two shrieks (consisted of two same syllables), while bark was produced also as a single syllable. In the case of the single syllable bark, there might be similarity with another call described in the yellow-bellied glider, the short call. Full call, short call and bark were produced in combinations. Short call was combined with gurgles, full call also with moan or beep (Kavanagh and Rohan-Jones,1982). Bark



vocalization was produced with chatter, hiss, low hiss or their combination. The main difference between yellow-bellied glider's and sugar glider's vocalizations in term of behavioural context is that agonistic vocalizations, described in sugar glider's vocal repertoire in this Thesis and also by Fleay (1947) are probably missing from yellow-bellied glider's vocal repertoire (Kavanagh and Rohan-Jones, 1982). In spite of the fact that I was not able to observe and record the full vocal repertoire of sugar glider, it seems that somehow limited vocal repertoire (in comparison to other groups of mammals) and use of combined vocalizations may be typical for Petauridae family vocal repertoire, as Kavanagh and Rohan-Jones (1982) also described relatively small number of vocalizations in other representative of this taxa.

The diversity of vocal repertoire of the sugar glider seems to be limited, when compared to eutherian small mammals, especially small, socially living nocturnal species. More than 15 different vocalization categories have been described in vocal repertoire of some socially living rodents (Yosida *et al.*, 2007). 17 different vocalizations were identified in vocal repertoire of insectivore Asian house shrew (*Suncus murinus*) (Schneiderová, 2014), 18 different vocalizations were identified in vocal repertoire of adult Senegal bushbaby (*Galago senegalensis senegalensis*) (Zimmermann, 1985).

To clearly determine the proper behavioural context of vocal categories has been complicated due to the fact that many vocalizations were produced inside nest-box, where no observation of behaviour could be done. Still, some conclusions about behavioural context of described vocal categories can be drawn. Hiss vocalization was produced during agonistic behaviour of lesser intensity, for example when one animal was approaching the other. Hiss vocalization was commonly recorded when both animals occupied the same nest-box. This vocalization might graduated into hiss-aggression, usually when animals began to fight with each other. In spite of unknown behavioural context of other vocalizations, I do not suggest that any vocalization apart from the ones that contain hiss or low hiss have agonistic behavioural context. Hiss-aggression was detected only in Group 2, where the only one nest-box was provided for sugar gliders. Vocalizations, that accompanied agonistic behaviour composed over 90% of vocalizations, while in other groups, where were at least two nest-boxes provided, it made maximally around 50% of all vocalizations. These findings suggest that it is important for welfare of sugar gliders in captivity to provide adequate number of nest-boxes, so animals are allowed to avoid each other.

There were two very similar vocal categories produced by sugar gliders, hiss and low hiss vocalizations. They could be distinguished from each other by their audible quality and their descriptive statistics differed also to some extent. However, because of their high variability, the fact that they sounded very similar (hiss as a “sss” and low hiss as “shshsh”) and were produced in similar behavioural context it is possible that low hiss is just a variation of hiss, caused by unknown factor (emotions or age of individual producing them, for example). To determine the true relation of these vocalizations, more research, including more groups of sugar glider would be needed.

All described vocalizations showed high variability, as indicated by high standard deviation. This variability may arise from several factors including age, individuality, sex, behaviour, emotinal state ect. More research is needed to reveal the causes of this variability and thus better understand vocal communication of the sugar glider.

## 7. Conclusion

I studied the vocal communication of the sugar glider kept in captivity. I found out that they emit singly produced vocalizations and also combined vocalizations. Their vocal repertoire composed of ten distinguishable vocal categories, four of them are classified as singly produced (bark, hiss, low hiss and chatter) another six of them were combined vocalizations (bark-hiss, chatter-bark, bark-low hiss, chatter-bark-hiss, chatter-bark-low hiss, hiss-aggression). I measured the relative occurrence of group of combined and singly produced vocalizations, using data obtained by one-zero sampling, and found out that the singly produced vocalizations are emitted at relatively high occurrence than combined vocalizations. I also measured the relative occurrence of different vocalizations for every group.

During my research I was recording four pairs of sugar glider, observing them during the recording and taking notes on their behaviour. All the recordings and observations were done continuously during night, when sugar gliders were vocally active. The recordings were then analysed, searched for the vocalizations and these vocalizations were classified into categories according to their auditory characteristics and acoustic structure. Average values of chosen acoustic parameters and their standard deviations were then measured for singly produced vocalizations and for one specific component of one combined vocalization (aggression component). The relative occurrence of all identified vocalizations were measured using one-zero sampling method for each group.

As written above, I managed to identify four singly vocalizations, which were described in terms of chosen acoustic parameters acoustic structure and, where it was possible, in terms of behavioural context. I also identified six combinations of these four vocalizations and one vocalization with component, which was never used as singly produced vocalization (hiss-aggression vocalization). I found there is a great variability in each vocal category, but the reason for this fact is unknown and additional research needs to be done to make some conclusions about this variability. I also found sugar gliders tend to produce relatively more vocalizations, that are clearly connected with agonistic behaviour, when not enough shelters are provided in enclosure. When comparing my results about the sugar glider vocalizations with known information about other species from family Petauridae, I concluded that somehow limited vocal repertoire and use of combined vocalizations might be characteristic for description of vocal behaviour of gliders, but more research both on the sugar glider and other gliders from Petauridae family needs to be done to prove this conclusion.

My Thesis successfully provided basic informations about captive sugar glider's vocal repertoire. However, there are still many aspects of vocal behaviour of sugar glider that needs to be explained. I suggest additional research on groups with more individuals, especially where offspring are present, to describe vocalizations produced by parents to offspring and vice versa. To identify the behavioural context of all identified vocalizations, research should include observing sugar glider's activity. To describe full vocal repertoire of sugar glider, intensive research on wild populations would be needed. This way, presence or absence of alarm, distress and other potential vocal categories can be proven. I also suggest research about presence of the ultrasonic vocalizations in the sugar glider vocal repertoire.

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