

Surviving cave bats: auditory and behavioural defences in the Australian noctuid moth, *Speiredonia spectans*

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SUMMARY

The Australian noctuid moth, *Speiredonia spectans* shares its subterranean day roosts (caves and abandoned mines) with insectivorous bats, some of which prey upon it. The capacity of this moth to survive is assumed to arise from its ability to listen for the bats' echolocation calls and take evasive action; however, the auditory characteristics of this moth or any tropically distributed Australian moth have never been examined. We investigated the ears of *S. spectans* and determined that they are among the most sensitive ever described for a noctuid moth. Using playbacks of cave-recorded bats, we determined that *S. spectans* is able to detect most of the calls of two co-habiting bats, *Rhinolophus megaphyllus* and *Miniopterus australis*, whose echolocation calls are dominated by frequencies ranging from 60 to 79 kHz. Video-recorded observations of this roost site show that *S. spectans* adjusts its flight activity to avoid bats but this defence may delay the normal emergence of the moths and leave some 'pinned down' in the roosts for the entire night. At a different day roost, we observed the auditory responses of one moth to the exceptionally high echolocation frequencies (150–160 kHz) of the bat *Hipposideros ater* and determined that *S. spectans* is unable to detect most of its calls. We suggest that this auditory constraint, in addition to the greater flight manoeuvrability of *H. ater*, renders *S. spectans* vulnerable to predation by this bat to the point of excluding the moth from day roosts where the bat occurs.

Key words: bat echolocation, moth ears, predator, prey, sensory ecology.

INTRODUCTION

Many moths possess ears that enable them to detect the echolocation calls of hunting insectivorous bats and avoid predation (Roeder, 1967). This now classic neuroethological story was originally set in the open skies where free-flying bats locate and attack their prey by what is known as aerial hawking. Further studies suggested that substrate-gleaning bats who find their prey by orienting towards the prey's incidental sounds (e.g. wing fanning) may be at an advantage in capturing eared moths owing to the reduced intensity as well as to the higher frequency of their calls (Fenton and Fullard, 1979; Faure et al., 1990; Faure et al., 1993). Roeder and Fenton introduced another potential arena for the arms-race between these two opponents, viz. subterranean roosts such as caves and mines in which bats and moths co-habit (Roeder and Fenton, 1973). In a North American mine, the noctuid moth, *Scoliopteryx libatrix* (Linnaeus) overwinters with hibernating bats [*Myotis* spp and *Eptesicus fuscus* (Beauvois)] and although it can hear the calls of these bats, it does not react behaviourally to them. This apparent anomaly was explained by the possibility that this moth inhibits its normal avoidance flight response to ultrasound to remain in the relative safety of the roosts, protected from abiotic (e.g. freezing) and biotic dangers (e.g. predation by birds). This study also reported that bats did not appear to eat the moths, perhaps because of the physical inability of aerially hawking bats to attack insects in the narrow confines of the mines.

Pavey and Burwell further developed this story of 'lambs laying down with lions' by describing the co-habitation of the Australian

Granny's Cloak noctuid moth, *Speiredonia spectans* Guenée, in subterranean day roosts with insectivorous bats, including the Eastern horseshoe bat, *Rhinolophus megaphyllus* Gray and the Little Bent-winged bat, *Miniopterus australis* Tomes (Pavey and Burwell, 2005). *S. spectans* lives in large numbers with these bats [Pavey and Burwell counted 1090 live moths in six sites (Pavey and Burwell, 2005)] but in contrast to Roeder and Fenton (Roeder and Fenton, 1973), they found moth wings on the floors of the roosts implying that co-habiting bats prey upon *S. spectans*. A report of cave-foraging in bats by Lacki and Ladeur described the Big-eared bat, *Corynorhinus rafinesquii* (Lesson) capturing moths in a cave in Kentucky, USA by both aerially hawking them and by gleaning them from the surface of the cave (Lacki and Ladeur, 2001). Pavey and Burwell suggested that in spite of the risk of predation, *S. spectans* co-habits with bats by being able to detect their echolocation calls and escaping to the walls of the day roosts (Pavey and Burwell, 2005). The echolocation calls of *R. megaphyllus* and *M. australis* contain frequencies of 60–70 kHz (Jones and Corben, 1993; Fenton et al., 1999; Reinhold et al., 2001), which although not frequency-matched (syntonic) are theoretically within the bandwidth of sensitivity of tropical moths (Fullard, 1988) and should be detectable by *S. spectans*. By contrast, subterranean day roosts occupied by the Dusky leaf-nosed-bat (*Hipposideros ater* Templeton) contained *S. spectans* wings but no live moths (Pavey and Burwell, 2005), suggesting that the very high frequency of this bat's echolocation calls [>150 kHz (Fenton, 1982; Crome and Richards, 1988)]

renders them less audible (allotonic) to the moths and allows this bat to prey more heavily on *S. spectans*.

The purpose of the present study was to examine the auditory capability of *S. spectans* to address the hypothesis proposed by Pavey and Burwell (Pavey and Burwell, 2005) that this moth can hear the echolocation calls of *R. megaphyllus* and *M. australis* but not those of *H. ater* in underground roosts. In addition, we examine the hypothesis that *S. spectans* avoids roost predation by adjusting its flight activity to minimize encounters with bats by testing the prediction that there will be an absence of flight overlap between the cave exits of the moths and the bats.

MATERIALS AND METHODS

Bat/moth flight activity

This study was conducted in three abandoned mines in coastal Queensland, Australia (Fig. 1), two of which were located at Camp Mountain, west of Brisbane, in sub-tropical Queensland and contained the bats, *Rhinolophus megaphyllus* and *Miniopterus australis*. The other mine was at Bramston Beach, south of Cairns, in the wet tropics of north Queensland and at the time of our study, contained the bats *R. megaphyllus*, *H. ater* and *Hipposideros semoni* Matschie [*H. semoni* is a rare species (van Dyck and Strahan, 2008) whose 108 kHz calls are readily distinguishable from those of *H. ater*; however, this species was recorded only once in the Bramston Beach site and is, therefore, not considered further in this study]. Bats and moths were observed in the Camp Mountain sites by illuminating the mine shafts with a near infra-red (NIR) light source (Extreme CCTV Surveillance Systems, model EX12LED, Scottsdale, AZ, USA) at wavelengths of 850–940 nm, to which Lepidoptera are insensitive (Horridge, 1977), and videotaping them with a NIR-sensitive camera (Swann Communications Pty., Richmond, Victoria, Australia) as they flew in the mine.

The distribution of *S. spectans* was established by collating collection locality data from preserved specimens in the following Australian insect collections: Australian National Insect Collection, Canberra; Australian Museum, Sydney; Queensland Museum, Brisbane; Queensland Department of Primary Industries and Fisheries Insect Collection, Brisbane; University of Queensland Insect Collection, Brisbane and Northern Territory Museum and Art Gallery, Darwin and were supplemented by observational records (C.R.P. and M. Braby, unpublished observations). To determine

sympatry with *S. spectans*, distributions of Australian bats were taken from maps in Churchill (Churchill, 1998).

Bat echolocation

We characterized the acoustic environment of the mines with respect to bat echolocation by remotely recording echolocation calls of bats as they flew in the mines. We used an ultrasonic microphone (Avisoft condenser microphone type CM16; frequency response, 10–200 kHz) and digitizer (Avisoft UltrasoundGate 416, Avisoft Bioacoustics, Berlin, Germany; sampling rate, 16 bit, 500 kHz) to record the calls and then analyzed sequences of calls using BatSound Pro software (v. 3.20, Pettersson Elektronik AB, Uppsala, Sweden). Although we cannot ascertain the actual number of individuals used in our acoustic analyses, we believe that the massive exodus of the bats from the caves at night effectively rules out the possibility of pseudoreplication. Only one sequence from each file was selected for measurement and these sequences were chosen on the following criteria: (1) only sequences with calls with a high signal-to-noise ratio (i.e. oscilloscope signal from the bat was at least three times stronger than the background noise as displayed on a linear time-voltage window) were analyzed; (2) only calls that were not saturated were analyzed (Fenton et al., 2001); (3) only calls that did not overlap with other calls were analyzed. For each call in a sequence, we measured the peak frequency (PF) from the power spectrum (1024-point FFT), call duration, inter-pulse interval (IPI) and inter-onset interval (time from the onset of one call to the onset of the next call in the sequence) from the oscillogram, and minimum frequency of the frequency modulated (FM) tails for *R. megaphyllus* and *H. ater* from the spectrogram. Minimum frequency (which can be calculated by subtracting the bandwidth from peak frequency in Table 1) for *M. australis* was taken as the value –20 dB below peak frequency whereas for *R. megaphyllus* and *H. ater*, this was taken as the lowest frequency of the FM component of the call as measured from the BatSound Pro spectrogram (time/frequency plot, 512-point FFT, resolution=19.36 ms per plot). IPI was measured from the end of one call to the initiation of the next call, and duty cycle was calculated by dividing pulse duration by the inter-onset interval. Bandwidth was measured at ± 20 dB below peak frequency for *M. australis* whereas for *R. megaphyllus* and *H. ater*, bandwidth was calculated by subtracting the minimum frequency of the FM component from the frequency of the constant frequency (CF)

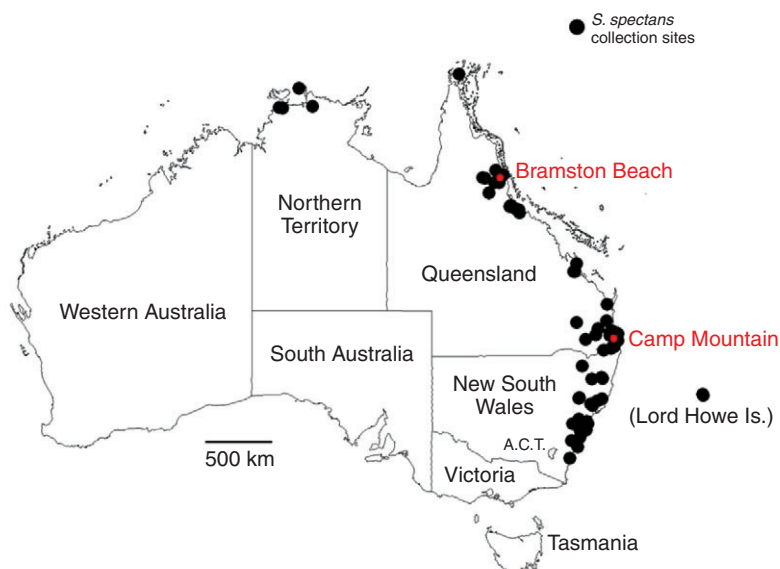


Fig. 1. Map of Australia showing locations of the study sites and the distribution of *Speiredonia spectans* based on specimen records from Australian insect collections and personal observations (C.R.P. and M. Braby, unpublished data).

Table 1. Echolocation parameters for *H. ater*, *R. megaphyllus* and *M. australis* recorded from free-flying bats in the mines

Species	Peak freq. (kHz)	Duration (ms)	IPI (ms)	Duty cycle (%)	Bandwidth (kHz)
<i>R. megaphyllus</i> (29 calls)	68.6±0.4	52.9±9.6	55.4±32.4	51.7±12.3	12.2±1.6
<i>M. australis</i> (35 calls)	66.4±1.8	3.4±0.7	62.9±5.9	5.6±1.9	32.1±10.6
<i>H. ater</i> (54 calls)	156.9±1.3	5.0±0.9	4.6±3.0	54.2±4.9	33.6±2.0

component. *R. megaphyllus* had an FM component at both the beginning and the end of each call, as opposed to the single FM sweep at the end of a call for *H. ater*. We, thus, used the first FM component when calculating the bandwidth for *R. megaphyllus* as this FM sweep was usually the longer of the two.

Moth auditory analyses

Moths were collected during the day from either the mines or from storm-water drains at the St Lucia campus of the University of Queensland and were used no more than 24 h later. Following decapitation and thoracic dissection, the action potentials of the A1 auditory receptor in the moths' tympanic nerve (IIN1b) (Nüesch, 1957) were recorded using a stainless steel hook electrode referenced to another in the moths' abdomen (Fullard et al., 2003). Neural responses were amplified (Grass Instruments P-15 Pre-amplifier, Astro-Med, West Warwick, RI, USA) and observed either on-line or stored in a laptop PC using digital acquisition boards (ADC 212/3, sampling rate=3 MHz; Pico Technology, St Neots, Cambridgeshire, UK or UltraSoundGate 416-200, 16 bit, sampling rate=250 kHz channel⁻¹) and oscilloscope-emulating software (PicoScope 5.10.7 or Recorder 2.9, respectively). Spike records were later analysed with a customized MATLAB (v. R2006b, The MathWorks Inc., Natick, MA, USA) application. In keeping with previous studies (Roeder, 1964; Fullard et al., 2003; Nabatiyan et al., 2003; Marsat and Pollack, 2006), we report spike periods rather than mean rates (e.g. spikes s⁻¹) as a direct measure of the activity of the auditory receptors and their likelihood to activate postsynaptic neural components (Hedwig, 2006).

Acoustic stimulation

Moth auditory preparations were exposed to pulsed synthetic sounds generated by a MATLAB application running on a separate PC laptop, amplified (Avisoft 70101) and broadcast from a speaker (ScanSpeak, Avisoft) mounted 30 cm from the moths. Intensities were recorded as voltages delivered to the speaker and then converted to peak equivalent sound pressure levels (dB peSPL) (r.m.s. re. 20 Pa) (Stapells et al., 1982) from equal-amplitude continual tones as previously measured with a Brüel & Kjær 4135 microphone and 2610 measuring amplifier (Brüel & Kjær; Nærum, Denmark). The entire system was calibrated before and after the study with a Brüel & Kjær 4228 pistonphone. Auditory threshold curves (audiograms) were derived using 20 ms sound pulses, 1 ms rise/fall times from 5 to 120 kHz delivered 2 s⁻¹ at randomly chosen 5 kHz intervals with A1 cell threshold determined as the stimulus intensity that evoked two receptor spikes per stimulus pulse.

Bat playbacks

Digital recordings of bat echolocation calls were made in the absence of human observers using the methods described above as the bats exited their day roosts in the Camp Mountain site. Of these recordings, two files approximately 1.5 s in duration that contained no saturated signals or overlapping bat calls were used as playbacks to seven auditory preparations of *S. spectans* using the same equipment as for the auditory measurements. One of these files

contained 23 calls of *R. megaphyllus* and the other contained five calls of *M. australis*. For each sequence, we set the highest amplitude of each bat call to 70 dB peSPL by matching its voltage to that of a continual 65 kHz tone (the frequency closest to that of the peak frequency for both bats for which our speaker was calibrated) of known intensity as generated using the set-up described above. We chose 70 dB as this intensity would minimize the number of occurrences of the moth's A2 receptor cell, which complicates the spike analysis. The echolocation sequence was played back five times to each auditory preparation and neural responses recorded using the UltraSoundGate 416-200 digitizer. From these playbacks, we isolated sections using BatSound Pro that contained calls of only one or the other of the two bat species and the moth's auditory nerve responses to those calls. We analyzed the auditory receptor responses off-line with a customized MATLAB application by measuring the number of A1 spikes per bat call and the percentage of calls in the playback sequence that evoked A1 spikes. We also applied Roeder's (Roeder, 1964) observation of 1.5–2.6 ms as the A1 spike period range that evoked evasive flight responses in North American noctuids and counted the percentage of spikes per bat call whose periods fell within this range. To check for possible auditory responses to electronic static from the speaker, the neural responses to the calls were compared with that of a 1.5 s playback of a blank file (i.e. one whose signal was reduced to zero) that broadcast only the output of the amplifier and speaker.

Our speaker was unable to reproduce the extremely high echolocation frequencies of *H. ater* without generating electronic noise that artifactually activated the moth's sensitive ear. This necessitated taking a portable auditory neural preparation (Faure et al., 1993) into the Bramston Beach day roost and allowing free-flying bats to stimulate the moth's ear as they exited. One moth was dissected and positioned near to the walls of the mine where *H. ater* preferred to fly and its auditory nerve was continuously monitored and recorded for 30 s every 15 min from 17:30 h until 19:00 h. All of the previously described neurophysiological recording methodology was used except that the USG digitizing board was set to an 8 bit, 500 kHz channel⁻¹ sampling mode to adequately capture the echolocation calls of the bat. The auditory A1 receptor responses (simultaneously recorded on a separate channel) to five different echolocation sequences (presumably of different bats because bats were not observed re-entering the mine once they had left) containing only *H. ater* calls (a mean of 26 calls sequence⁻¹) were then analyzed as described above.

RESULTS

Bat/moth flight activity

Bat and moth activity inside the roosts was recorded from 30 min before sunset (12 November 2007, 18:14 h) onwards for six nights during the Austral spring from the 7th to the 25th November 2007 for a total of 50 h. Of these six nights, four nights of continual recording from 30 min before sunset to 511 min after sunset for a total of 36 h were analysed for evidence of activity overlap between bats and moths. Activity was measured as the number of fly-bys (to a maximum of ten) of bats and moths on the video monitor per

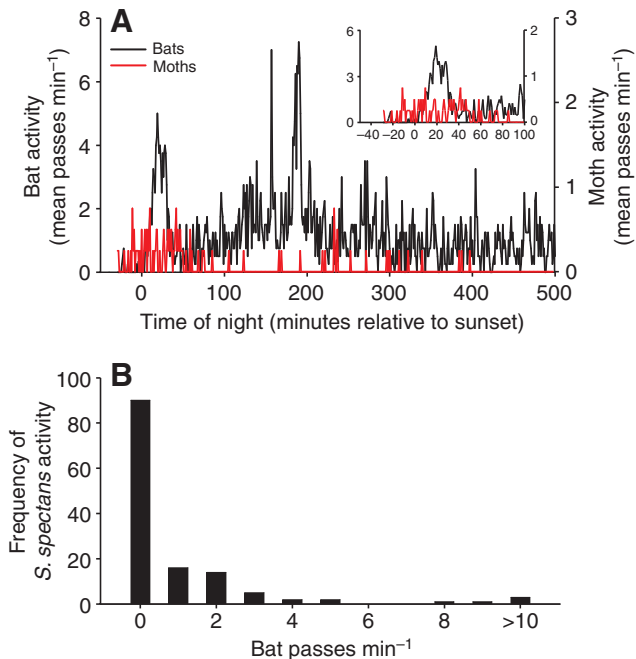


Fig. 2. (A) Activity patterns, in mean passes per minute ($N=4$ nights, total of 36 h), of bats and *S. spectans*, in an abandoned mine at Camp Mountain, Australia. Inset: expanded view of the initial portion of the evening flight activities. (B) The frequency of *S. spectans* flight episodes observed for each number of bat passes per minute.

one minute of observation time. We did not differentiate between exits and entrances of the bats or moths because our main interest was determining the degree of flight overlap between the two during the night. Fig. 2A illustrates the mean activity of both bats and moths over the four nights and indicates two bouts of bat activity, the first beginning at sunset and lasting for 50 min with another at approximately 200 min after sunset. Fig. 2A (inset) indicates a slight trend for moths to be active before and after bats began to fly but without an obvious separation of flight activity.

To more precisely test for activity overlap, we examined each 1 minute bin as a discrete event and showed that the presence of bats excludes that of moths. By analysing bat and moth fly-bys as binomial events, moth activity is significantly more likely to occur during periods of bat inactivity (Fisher's Exact test, $P<0.001$). Fig. 2B shows the frequency of 1 minute bins where moths are active for each number of bat passes ranging from 0 to >10 and indicates that it was more likely to observe a moth in flight when no bats were present. The single recording we made of an entire night showed moths returning to the roost once bat activity had completely subsided.

We observed no instances of predation by bats, either by aerially hawking or by gleaning in the 50 h of video recordings and saw only four in-flight interactions between bats and moths. In these instances, moths exhibited evasive flight manoeuvres in response to passing bats by either diving to the ground (one of four) or landing on the wall of the mine (three of four).

Bat echolocation

The number of files per species that we analyzed was: *R. megaphyllus*, four files; *M. australis*, five files; *H. ater*, four files. *H. ater* produced CF/FM calls of higher peak frequency and bandwidth than the FM/CF/FM calls emitted by *R. megaphyllus*;

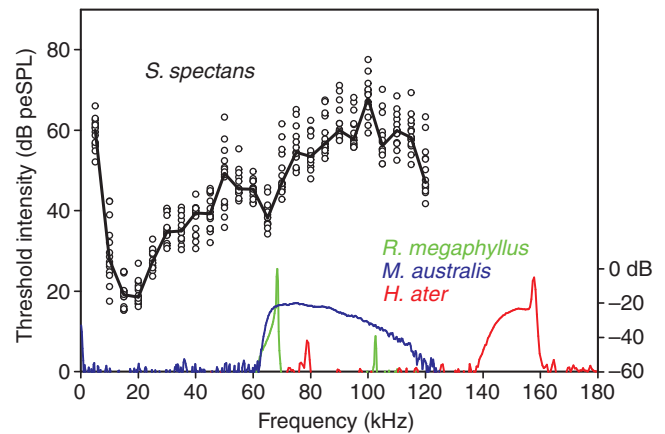


Fig. 3. Auditory sensitivity of *S. spectans* (median, black line; open circles, individuals, $N=14$). Superimposed are the frequency power spectra of the echolocation calls of three bats that day roost in the same subterranean sites as this moth.

however, the latter emitted calls of much higher call duration and IPI resulting in their duty cycles being similar (Table 1). The peak frequency of the FM calls of *M. australis* was similar to that of *R. megaphyllus* but the call duration of the former is much shorter and the IPI much longer giving *M. australis* a lower duty cycle. The broad bandwidth of the *H. ater* calls means that the FM sweep drops to a mean frequency of 123.3 ± 2.2 kHz.

Moth auditory analyses

Audiograms

Fig. 3 illustrates the auditory sensitivity of 14 individual *S. spectans* (10 females, four males; no obvious differences were observed between males or females so the genders were pooled). *S. spectans* exhibits its maximum sensitivity at a bandwidth of 15–40 kHz. At its best frequency (20 kHz), this moth possesses extraordinarily low thresholds of 20–30 dB, a trait that persists into the higher frequencies (>75 kHz) with thresholds of 40–60 dB. Subsequent to our study, the speakers used for the playbacks were tested for extraneous sub-harmonic noise that could have artifactually activated the moth ear. All energy outside of the test frequencies existed at intensities less than 40 dB from the peak of the test frequency, which leads us to believe that the threshold values in Fig. 3 are valid.

Comparing the median audiogram in Fig. 3 with the frequency spectra of typical echolocation calls that were recorded in the same mines where the moths day-roosted, we predicted that *S. spectans* should be able to detect the calls of *M. australis* and *R. megaphyllus* (in fact, there appears to be a specific increased sensitivity at the fundamental frequency of *R. megaphyllus*) but would be unlikely to hear the calls of *H. ater*, although this is uncertain as our speakers could not reproduce frequencies higher than 120 kHz.

Bat playbacks

Figs 4 and 5 illustrate the responses of *S. spectans*' A1 auditory receptor to the calls of *R. megaphyllus* and *M. australis*. The neural traces demonstrate that the ears of this moth responds to the calls of both bats, although the long calls of *R. megaphyllus* evoke a significantly greater number of A1 spikes per call than do the shorter calls of *M. australis* (Fig. 5A). In addition, the percentage of bat calls that evoked any A1 spikes in auditory preparations of *S. spectans* was significantly higher for *R. megaphyllus* than for *M.*

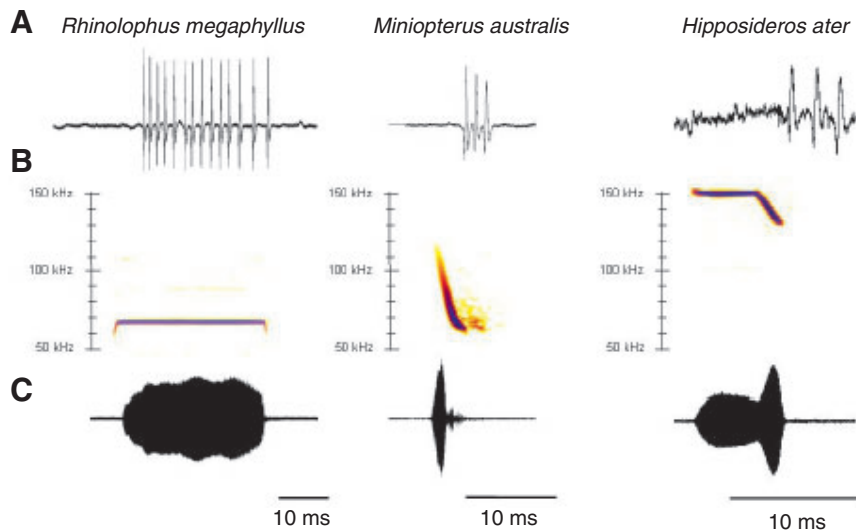


Fig. 4. *S. spectans*' maximum auditory nerve A1 receptor cell responses (A) to calls of the three bats (C) with their responses spectrograms (B).

australis (Fig. 5B). *R. megaphyllus* calls also elicited significantly more A1 spikes per call with periods that have been reported previously as evoking evasive flight (Roeder, 1964) (Fig. 5C).

As previously described, the inability of our speakers to reproduce the calls of *H. ater* necessitated the field exposure of a moth auditory preparation to these bats as they exited their day roost. Fig. 4 illustrates the *S. spectans*' maximum A1 cell response to one of the calls of this bat and suggests that although the moth is completely deaf to the initial CF portion of the call, it can detect the lower frequencies contained in the FM portion. In spite of this detection ability, only 16% of the calls of *H. ater* recorded elicited any A1 activity compared with 98% and 66% for *R. megaphyllus* and *M. australis*, respectively (Fig. 5B). Although the single moth sample size for the *H. ater* exposure trials prohibits statistical comparisons, the few calls of *H. ater* that were detectable by *S. spectans* produced a surprisingly similar number of A1 spikes as those produced by *M. australis* (Fig. 5B) and with a similar percentage of periods (Fig. 5C) that meet Roeder's criterion of evoking evasive flight (Roeder, 1964).

DISCUSSION

Who is *S. spectans* listening for?

The ears of *S. spectans* are among the most sensitive ever analysed, exhibiting thresholds lower than those described in non-tropical Australian moths (Surlykke and Fullard, 1989; Fullard, 2006) and rival those of moths from other areas of high bat abundance and diversity [Côte d'Ivoire (Fenton and Fullard, 1979), South Africa (Fullard et al., 2008), Zimbabwe (Fullard and Thomas, 1981)]. By contrast, the ears of the most sensitive Canadian and Danish noctuids analysed possess thresholds of 30–40 dB at their best frequencies, which rapidly increase above 50 kHz becoming functionally deaf at 80–100 kHz (Surlykke et al., 1999). The sensitive ears of *S. spectans* probably arise from two conditions. First, it is a large species [mean forewing length 39.5 mm (36–42.1 mm, $N=20$) (C.J.B., personal observations)] and larger moths possess greater auditory sensitivity due to the fact that they provide more detectable echoes to searching bats (Surlykke et al., 1999). Second, *S. spectans* occurs in the wet–dry tropics of the Top End of the Northern Territory and the wet tropics of north Queensland south to southern New South Wales (Common, 1990) (Fig. 1) – areas that contain a rich diversity and abundance of echolocating bats (Crome and Richards, 1988; Law

and Chidel, 2002; Rhodes, 2002; Pavey et al., 2006). Fullard suggested that the broad tuning of moth ears is the evolutionary result of the need of these insects to detect the acoustic assemblage of all of the bats that hunt them, therefore, moths exposed to diverse bat communities typically found in the tropics will possess ears with greater sensitivity across more frequencies (Fullard, 1982; Fullard, 1988). Regardless of the total auditory sensitivity, it is the moth's audiogram peak sensitivity that identifies the frequencies [i.e. bats, assuming that bat-detection is the main use for these ears (Fullard, 1988)] from which the moth receives the greatest selection pressure. From this theory, bats that emit syntonetic echolocation frequencies (those matched to the moth's peak sensitivity) form the heaviest predation potential on moths compared with those that emit allotonic (frequency-mismatched) calls. Paradoxically, allotonic bats should be expected to consume more moths (i.e. present a greater selection pressure) as a result of their reduced detectability. The explanation to this apparent conundrum is the relative scarcity of allotonic bats, compared with the syntonetic predatory community, allowing them to exploit the sensory equilibrium that exists between moths and sympatric bats [i.e. they are 'cheaters' in an evolutionary stable acoustic relationship (Faure et al., 1993)]. To examine this, we created composite spectra of the peak frequencies of the calls emitted by the species of Australian bats sympatric with *S. spectans* for which acoustic data are available [Anabat files (D. J. Milne, unpublished data) and analysed with AnalookW (v. 3.5m, www.hoarybat.com)]. These spectra were weighted with the relative abundances (C.R.P. and D. J. Milne, unpublished data) of each bat species as estimated in the three locations that encompass the distribution of the moth: Top End Northern Territory, north Queensland (including the Bramston Beach study site) and south-east Queensland (including the Camp Mountain study site). Fig. 6 illustrates the three echolocation assemblages compared with the mean audiogram of *S. spectans*. These figures indicate that the heaviest predation potential on moths arises from bats that echolocate in a bandwidth of 20–60 kHz, predicting that common bats emitting these frequencies and preferring moths in their diets [e.g. *Chaerephon jobensis* (Miller), *Chalinolobus gouldii* (Gray) (Churchill, 1998)] form the greatest predatory threat for *S. spectans*. Fig. 6 further indicates that higher frequency echolocators such as *R. megaphyllus* and *M. australis* are less common hunters of *S. spectans* but still form a significant

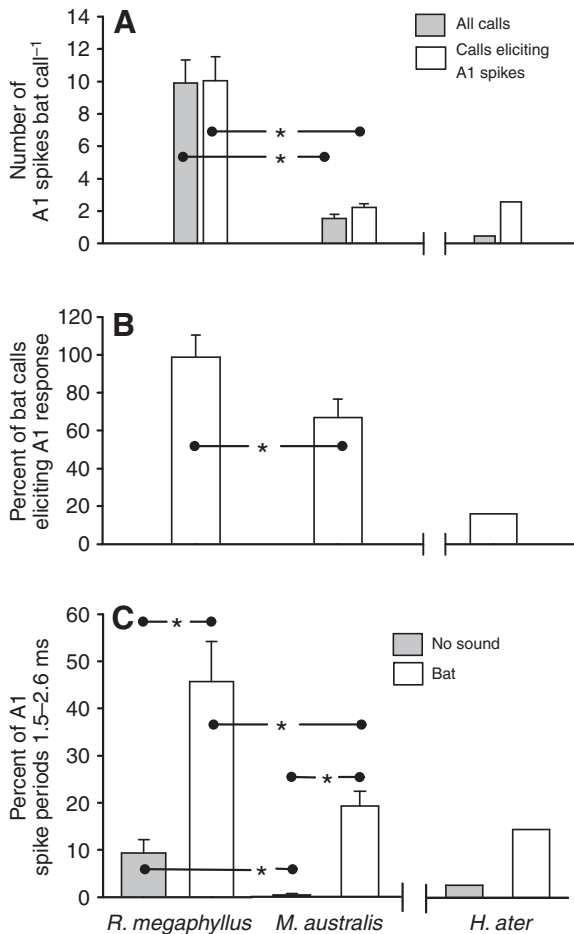


Fig. 5. (A) The mean (+ one standard error) numbers of *S. spectans* ($N=7$) A1 receptor spikes that were evoked to the calls of the three bats. (B) The percent (+ one standard error) of bat calls that exhibited any A1 spikes. (C) The mean percent (+ one standard error) of A1 spikes that possessed periods less than 2.6 ms compared with an equal amount of time with no sound. Significant differences ($P<0.05$, paired t -tests) indicated by asterisks, statistical comparisons were not done with the single moth response to the calls of *H. ater*.

predation potential [*R. megaphyllus* feeds extensively on moths over a wide geographical range (Pavey and Burwell, 2004)]. Finally, Fig. 6 indicates that the allotonic calls of uncommon bats such as *H. ater* fall considerably outside of the sensitivity of *S. spectans*, and predicts that while these bats will take large numbers of moths in their diets [a prediction supported by data for *H. ater* at Bramston Beach where 91.7% of prey items were noctuid moths (Pavey and Burwell, 1998)] they are, nevertheless, rare predators of moths across the entire geographical range of *S. spectans* and, therefore, impart little selective force on its auditory design. Faced with the need to detect the greater number of bats echolocating at lower frequencies, the moth's high frequency insensitivity constitutes a sensory constraint that cannot be overcome by its limited auditory hardware. The general similarity of the echolocation assemblages among the three locations also predicts that the ears of *S. spectans* will not exhibit auditory signs of local adaptation. To verify this prediction would require testing moths from the three locations sympatric with the local community of bats.

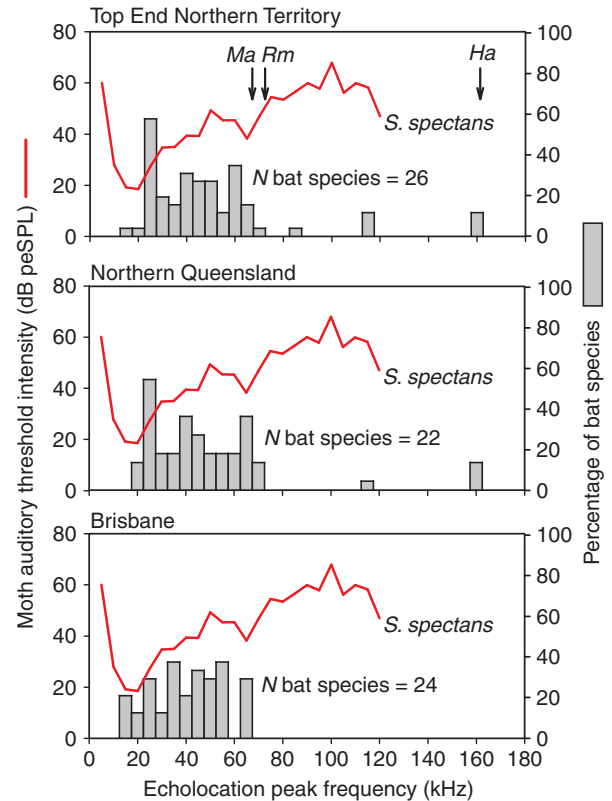


Fig. 6. Comparison of the auditory sensitivity of *S. spectans* with the echolocation assemblages of sympatric species of insectivorous bats (for which data exist) in three areas of the moth's distribution following a weighting of the echolocation frequencies by the relative commonness of each bat species. The peak frequencies of the three bats examined in the present study are indicated by arrows (Ma, *Miniopterus australis*; Rm, *Rhinolophus megaphyllus*; Ha, *Hipposideros ater*).

Why does *S. spectans* co-habit with *R. megaphyllus* and *M. australis*?

S. spectans exists by the hundreds in subterranean roosts occupied by *R. megaphyllus* and *M. australis* and should be a valuable prey for these bats, a prediction supported by the observations of Pavey and Burwell of wings left on the floors of day roosts (Pavey and Burwell, 2005). The question is therefore, how in the face of predation from these bats does *S. spectans* defend itself when sharing its day roosts? Edmunds defines primary and secondary defences as those that operate before and after, respectively, a predator is aware of its intended prey (Edmunds, 1974). We suggest that *S. spectans* employs both types of defence to allow it to exploit the relative safety of subterranean roosts from diurnal predators such as birds. Regarding primary defence, Soutar and Fullard (Soutar and Fullard, 2004) examined how earless moths could protect themselves against bats, such as by flying less (Roeder, 1974). For *S. spectans*, the simplest defence could be that they do not fly when the bats are active in their day roosts and this is borne out by our observations that the activity of *S. spectans* within the roosts is greater in the absence of bats. Although it is possible that other factors (e.g. metabolic condition) may contribute to the evening flight patterns of these moths, we believe that the most likely reason that moths do not fly when bats do, is that they are acoustically aware of the bats and remain perched.

The possession of sensitive ears should allow *S. spectans*, when inside the roosts, to employ its secondary defence of detecting the bats' echolocation calls and thereby either remaining perched when a bat flies by or by avoiding the bat if the moth is already in flight. *S. spectans* characteristically rests with its wings elevated from the surface of the cave wall thus exposing its ears and increasing its ability to hear approaching bats. Pavey and Burwell reported that *S. spectans* flying in mines responded to the attacks of bats, as well as to the ultrasonic sounds of dog whistles, by immediately lighting upon the walls (Pavey and Burwell, 2005). They, as we, did not observe any incidence of bats landing on the walls to capture moths (i.e. gleaning) and it may be that *R. megaphyllus* and *M. australis* infrequently, if ever, glean their prey, thus allowing the moth enough protection to successfully co-habit with these bats. Alternatively, *R. megaphyllus*, like other rhinolophids, may require that perched prey be moving their wings (Siemers and Ivanova, 2004) to localize them; hearing this bat would allow *S. spectans* the opportunity to remain still. The rarity of observed captures of *S. spectans* by bats while in the roosts suggests that the majority of the wings on the floors of the roosts originate from captures outside of the mines and that little predation takes place during the day or the evening as the moths begin to exit. The sensitivity of its ears, therefore, allows *S. spectans* to monitor the presence of bats in flight and remain motionless on the cave walls until there is a break in bat activity. Being acoustically 'pinned down' by bat echolocation calls may confer an immediate survival advantage to these moths but may delay the evening emergence of moths and leave some trapped inside the cave for the entire night. These predictions could be tested by comparing the exit activity of *S. spectans* in day roosts with and without co-habiting bats.

Why does *S. spectans* not co-habit with *H. ater*?

Pavey and Burwell suggested that *H. ater* uses the allotonic nature of its calls as an acoustic counter-manoeuve to increase its foraging success on *S. spectans* to the point of excluding them from subterranean roosts (Pavey and Burwell, 2005). If *S. spectans* could adequately hear *H. ater* we would expect to find some moths co-habiting with this bat but this is not the case (Pavey and Burwell, 2005). Although the extraordinary sensitivity of *S. spectans* allows it to detect the FM portion of some of *H. ater*'s calls, the low percentage of calls that the moth did detect compared with those of *R. megaphyllus* and *M. australis*, suggests that this bat is functionally inaudible to the moth or only detectable at very short distances. We suggest that the chance of *S. spectans* escaping *H. ater* would depend on the distance at which the moth first hears the bat and whether or not *H. ater* can glean. Some hipposiderid bats, including Australian species, are able to glean prey from surfaces (Bell and Fenton, 1984; Pavey and Burwell, 2000) and *H. ater* may share this trait. A gleaning *H. ater* would be able to take a moth that, unable to hear the bat until late in its attack sequence, had landed on the wall and was still moving its wings [hipposiderid bats require movement to detect their prey (Link et al., 1986)]. Alternatively, as mentioned above, a perched moth vibrating its wings in preparation for flight might detect a bat homing in on it but not in time to stop moving and deny the bat its localization cue.

What if *H. ater* does not glean (Pavey and Burwell, 2000)? Allotonic echolocation in combination with other factors that influence a bat's dietary composition [e.g. flight ability, prey preference (Jacobs et al., 2008)] may explain a non-gleaning *H. ater*'s success at capturing *S. spectans*. *H. ater* is a small bat with

a relatively high wing-aspect ratio (Crome and Richards, 1988), which should allow it to better negotiate the physically and acoustically cluttered confines of its day roosts. The percentage of moths in the diet of individuals in a population of the African hipposiderid, *Hipposideros ruber*, was positively correlated with aspect ratio and wingspan (Jones et al., 1993) suggesting that bats with agile flight may be better suited to capture moths. A non-gleaning *H. ater* could, therefore, combine flight manoeuvrability with allotonic calls to enhance its chances of catching flying *S. spectans* in the day roosts. Differences in flight manoeuvrability may also explain why *H. ater* includes more moths in its diet than the larger *Hipposideros cervinus* (Gould) (CF: 145 kHz) despite both using allotonic echolocation frequencies (Pavey and Burwell, 2000). In this respect, the moth-preference of *H. ater* is of special interest as this bat may actually restrict the cave and mine dwelling distributions of *S. spectans* in northern Australian locations where *H. ater* and similar bats are abundant.

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