Optimising anuran monitoring investigations in a tropical seasonal environment

Josephine D'urban Jackson

Zoology BSc

Subject Superviser: Dr Graeme Gillespie

Vocational Superviser: Dr Benoit Goossens

Danau Girang Field Centre (DGFC), Sabah, Borneo

Word count: 5810

Rhacophorus pardalis

Part A:

The year I spent at Danau Girang Field Centre, Borneo has been the most unforgettable and incredible experience I have ever had. I was privileged enough to assist on research focussing a diverse range of taxa including nocturnal and diurnal primates, reptiles, birds and of course my focal project on amphibians. Throughout the year I learnt new and further developed skills including camera trapping, radio tracking, mark recapture methods and behavioural observations. My knowledge of research techniques and tropical biodiversity has been vastly improved from my time at this research centre. Everyday was a learning experience as we were living within the forest, and we had facilities at the field centre that enabled us to expand our knowledge of the local fauna and flora.

During the year I also developed skills in science communication, a topic I am particularly interested in. We presented our research projects to visitors and all the field courses that came to the research centre and additionally displayed our practical skills with them in the field. This built up my confidence in public speaking and improved my skills in adjusting the explanations of scientific topics for a range of audiences with different abilities. Together (the PTYs) wrote, edited and produced a magazine to inform those that had an interest with DGFC of all the happenings each month including sections on projects occurring throughout Sabah and focal pages on animals of particular conservational interest.

I feel this year has also substantially developed my understanding of current worldwide conservation issues, most notably the oil palm industry and its' benefits as well as the well documented detrimental impacts to wildlife. Living in an area that has experienced severe modification through logging and forest conversion into oil palm plantations, put the issues I had learnt about previously, into a practical situation and has further encouraged me to become more involved with conservation, especially in South East Asia.

Having the opportunity to design and complete my own project has given me a broader and more realistic view of field research than I had had previously. This experience and meeting many biological researchers in the process has further encouraged me to extend my studies of zoology through post graduate research focussing on tropical biodiversity. I have gained a wide variety of skills, including learning a new language, and had the most incredible experience this year that I will remember for the rest of my life. I am so grateful to my supervisor who gave me this opportunity and to all staff and students at the field centre (and particular my research assistant, Samsir Laimun) and those who I met along the way that made my time at DGFC extend beyond all possible expectations.

Part B: Optimising anuran monitoring investigations in a tropical seasonal environment

Abstract

This study aimed to improve the efficiency of anuran monitoring and census methods by evaluating the optimal time of year to survey and the level of sampling effort required to sufficiently represent the focal amphibian community of a seasonal tropical environment. Over 11 months, visual and acoustic encounter surveys took place along transects within two distinct habitats, secondary degraded forest and oil palm plantations. Species estimates and accumulation curves were created to analyse the adequacy of sampling effort. The influence that environmental and methodological factors had on the number of species detected per survey were evaluated by a general linear model. Findings support previous studies, suggesting rainfall is the most influential factor determining the number of species detected throughout the year. Significantly fewer species were found within oil palm plantations compared with forest fragments. Plantation transects required a lower sampling effort to detect all species compared with forest transects. During the investigation, the discovery of two new calling behaviours and a potentially new species highlight the lack of knowledge in this region and indicate the need for further studies.

Introduction

Current detrimental changes to environmental conditions are causing catastrophic amphibian population losses worldwide (Stuart *et al.,* 2004). The threat to diversity within this group has in turn triggered an increase in development of amphibian monitoring studies (Heyer *et al.,* 1994). The majority of which have focussed on anurans (Gooch *et al.,* 2006). These studies provide essential information on species declines and the effects of conservation initiatives (Beebee & Griffiths, 2005). South East Asia is a region of high amphibian diversity (Frost, 2009, cited in: Rowley *et* al., 2010) yet additionally, experiences the highest rate of habitat loss in the world (Sodhi *et al.,* 2004; Sodhi & Brook, 2006). Despite documented intercontinental declines and extinctions of frogs and toads (Stuart *et al.,* 2004) combined with severe deforestation in South East Asia (Sodhi *et al.,* 2010), the amphibians that inhabit this region have been somewhat neglected in terms of population monitoring studies and conservation efforts (Rowley *et al.,* 2010).

"Designing a study is as much an art as a science" (Mackenzie, 2005). Crucial to all population monitoring investigations is the reliability of results to accurately represent their focal biological communities (de Solla *et al.,* 2005). In terms of conserving frogs and toads, focussing on large scale changes of anuran assemblages through presence/absence studies across a large range is more useful than detecting specific abundance changes in select populations (Beebee & Griffiths, 2005; Canessa *et al.,* 2012).

Key to the methodology of species richness and diversity studies in a number of taxa (Mackenzie & Royle, 2005; Garrard *et al.,* 2008; Watson *et al*., 2008) including amphibians and reptiles (Gibbons *et al.,* 1997; de Solla *et al.,* 2005) is the allocation of sampling effort. This has an important influence on the accuracy of results (de Solla *et al.,* 2005; Garrard *et al.,* 2008; Watson *et al.*, 2008). Sample-based species accumulation curves can accurately estimate the species richness of an area (Ugland *et al.,* 2003) and therefore indicate the adequacy of the allocated sampling effort (de Solla *et al.,*2005; Chiarucci *et al.,* 2008). Single visit surveys are not suitable for many anuran communities (Gomez-Rodrigues *et al.*, 2010) due to their occasionally low (Mazerolle *et al.,* 2007) and varying detectability between species (de Solla *et al.,* 2005) and within breeding seasons (Canessa *et al.,* 2012). However, if there are too many replicates with only a short time period in between each, some microhabitats of anurans will be destroyed giving the false appearance of species declines (Marsh

& Haywood, 2010). The occurrence of false absences as a result of insufficient sampling effort can easily diminish the integrity of the investigation (de Solla *et al.,* 2005; Mackenzie, 2005; Gomez-Rodrigues *et al.*, 2010), reduce statistical power (Fields *et al.,* 2005) and result in misdirected management and conservation decisions (Garrard *et al*., 2008; Canessa *et al.,* 2012).

False non-detection can also be caused by the seasonal variation of species present in an area (Canessa *et al.,* 2012). Population monitoring surveys often rely upon identifying species though acoustic and visual encounter surveys (de Solla *et al.,* 2005; Gooch *et al.,* 2006; Dorcas *et al.,* 2010). Anuran species inhabiting tropical environments have the highest detectability when they are most active, which is often during their breeding season (Bertoluci & Rodrigues, 2002). It is well researched that in seasonal tropical environments, anuran breeding activity is strongly dependant on exogenous factors such as ambient air temperature and most notably, rainfall (Aichinger, 1987; Donnelly &Guyer, 1994; Bertoluci & Rodrigues, 2002; Prado *et al.,* 2005). However, seasonality studies have so far been restricted to the Neotropics (Aichinger, 1987; Bandoni de Oliveira & Navas, 2004; Bertoluci & Rodrigues, 2002; Prado *et al.,* 2005). As suggested, previous studies have found a strong association with the rainy season of tropical environments and the maximum number of species breeding (Bertoluci & Rodrigues, 2002; Prado *et al.,*2005). However, several reproductive activity patterns can be recognized throughout the year between species due to differences in reproductive mode and behaviours (Duellman & Trueb, 1986; Bertoluci & Rodrigues, 2002; Prado *et al.,* 2005). The non synchronization between species breeding patterns is potentially an evolutionary consequence of competition avoidance via temporal segregation (Wells, 2007). Therefore, the efficiency of anuran monitoring programs in tropical environments is strongly determined by species breeding ecology and survey timing (Wells, 1977; Canessa *et al.,* 2012). Both of which should be taken into account when designing a study to account for seasonal biases of detection probability anurans (Bridges & Dorcas, 2000; de Solla *et al.,* 2005; Gomez-Rodrigues *et al.*, 2010; Canessa *et al.,* 2012).

One way to improve the efficiency of surveys by reducing the possibility of false absences is the use of statistical models. These models can predict the optimal survey timing and sampling effort required based on prior knowledge of the detection probabilities of the community (Fields *et al*., 2005; Canessa *et al.,* 2012). The use of statistical models can therefore optimise the design of surveys by maximising the probability of detection without over expenditure of time or budget (Garrard *et al*., 2008; Canessa *et al.,* 2012).

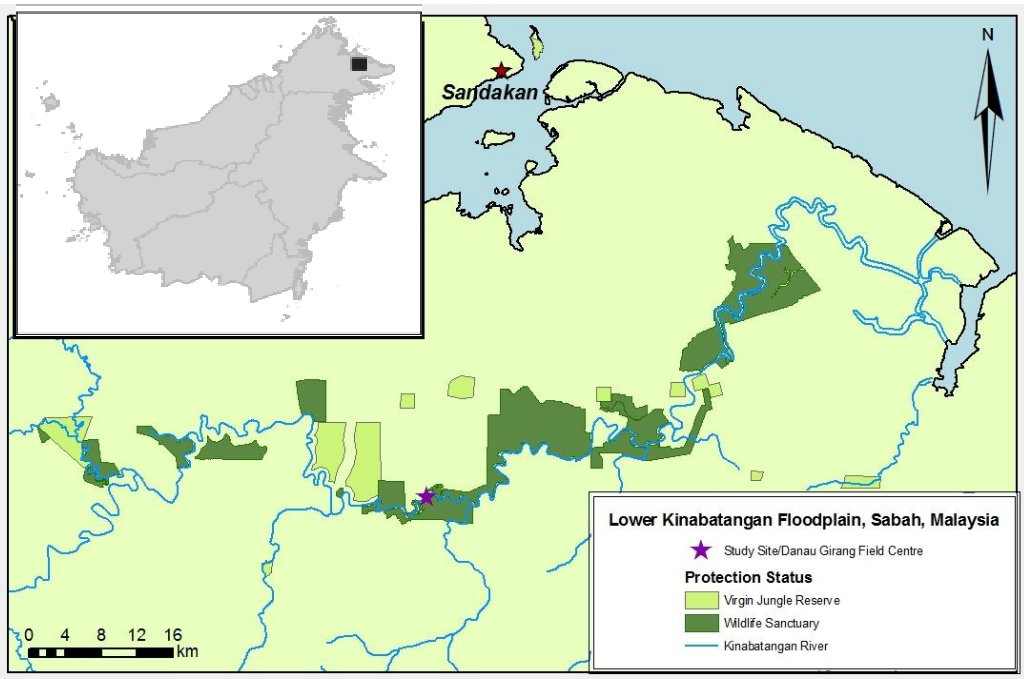

Figure 1. A map of the study location situated in Western Sabah, Borneo. The expanded section highlights the Lower Kinabatangan River and the surrounding protected areas where this investigation took place. Map by Danica Stark and adapted from Gillespie *et al. (*2012)

Despite the detrimental consequences of inaccurate surveys (de Solla *et al.,* 2005; Fields *et al.,* 2005 Mackenzie, 2005; Garrard *et al*., 2008; Gomez-Rodrigues *et al.*, 2010; Canessa *et al.*, 2012), studies concerning the efficiency of anuran monitoring programs in relation of sampling effort and seasonality are lacking (Peirce & Gutzwiller, 2004; Gomez-Rodrigues *et al.*, 2010) despite their growing need (Bridges & Dorcas, 2000; Garrard *et al.,* 2008). The paucity of amphibian monitoring is especially evident in the tropics (Stuart *et al.,* 2004). This region supports the majority of amphibian species (Vitt & Caldwell, 2001; Wells, 2007; Stuart *et al.,* 2004) yet suffers from widespread deforestation (Laurance, 1999),which is the greatest threat to amphibian diversity (Stuart *et al*., 2004; Sodhi *et al*., 2008; Gillespie *et al.,* 2012). The habitat preference for 59.1% of amphibian species, 48% of which are rapidly declining, is tropical lowlands (Stuart *et al.,* 2004). In South East Asia, during the past two decades, this habitat type has been severely exploited for conversion to oil palm plantations (Fitzherbert *et al.,* 2008).

Oil palm plantations are concentrated within lowland areas of Indonesia and Malaysia, together producing over 80% of the world's supply (Koh & Wilcove, 2007). By 2100, habitat loss through commercial logging and land conversion to oil palm (*Elaeis guineensis*) plantations has been predicted to cause a 13–85% reduction of biodiversity within South East Asia (Sodhi *et al.,* 2010). As a result of severe fragmentation and degradation of forest in Malaysian Borneo (McMorrow & Talip, 2001), several anuran species there are currently threatened (IUCN, 2012; Sodhi *et al.,* 2010). However, there have only been two in depth investigations into the effects of forest conversion to oil palm plantations on anuran abundance, composition and species richness (Scriven, 2011; Gillespie *et al.,* 2012). Both of which documented a significantly fewer species within oil palm plantations compared to secondary forest fragments (Scriven, 2011; Gillespie *et al.,* 2012). These studies support findings from studies from other taxa including birds (Aratrakorn *et al*., 2006), plants (Chung *et al.,* 2000), invertebrates (Falye *et al.,* 2010), lizards (Glor *et al.,*2001) and mammals (Maddox *et al.,* 2001). The most recent investigation into the impact of forest conversion on amphibian communities in Sabah, Borneo (Gillespie *et al.,* 2012) also found despite heavy degradation, forest patches surrounded by oil palm plantations have high value in terms of lowland amphibian conservation.

Despite the widespread amphibian extinctions and declines (IUCN, 2012) and yet the knowledge of persisting amphibian diversity (Gillespie *et al.,* 2012) within this extremely degraded region of Sabah (McMorrow & Talip, 2001), no amphibian monitoring system is currently in place here. The implementation of this would increase understanding of spatial patterns of species richness and community composition across different environments and land uses. By repeat surveying over

11 months in a seasonal tropical environment using a robust method, this investigation aims to find the optimum timing and number of surveys required to provide the most accurate estimation of species richness within secondary degraded forest fragments and oil palm plantations.

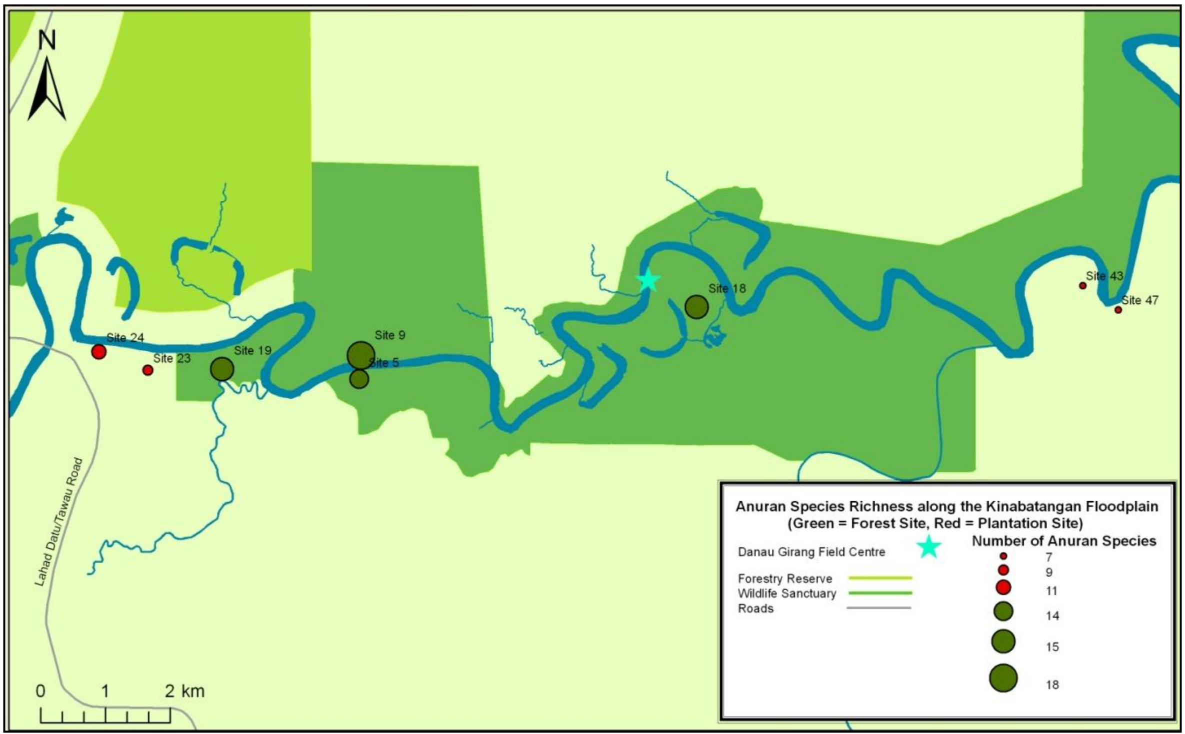

Figure 2. Map incorporating the location along the Lower Kinabatangan River of each transect monitored in this investigation and a visual representation of the total number of species found at each site. Site numbers were allocated by a previous study (Scriven, 2011). Map by Danica Stark.

Methods and Materials

Study Location

Surveys of anurans were conducted at predetermined sites situated at various intervals along the Lower Kinabatangan River floodplain, Eastern Sabah, Malaysia (5◦10’–5◦50’N; 117◦40’–118◦30’E)(Fig. 1). This region is characterised by an annual rainy season, typically lasting from November to March (Scott, 1989), during which the river swells and some areas become totally submerged (Hutton, 2004). Average monthly temperature ranges from 27°C to 29°C, humidity is 95-6% throughout the year and mean annual rainfall is between 2500-3500mm (Ancrenaz *et al*., 2004).

As a consequence of deforestation by commercial and illegal logging in the 50s and 90s and extensive land conversion to oil palm plantations, forest cover throughout the floodplain is degraded and highly fragmented (McMorrow & Talip, 2001). It was not until 2002, when the State Government selected ten remnant patches of secondary forest along the river to be protected, thus creating the Lower Kinabatangan Wildlife Sanctuary (LKWS)(Fig. 1 & 2).

Site Selection

Surveys took place at eight transects, 120 m in length and parallel to the river, which were originally established during a previous study (Scriven, 2011) to survey frogs in the region (Fig. 2). Four sites were located in LKWS forest fragments and four transects were set up within palm oil plantations bordering the river (Fig. 2). These sites were chosen from a selection of 50 transects on the basis of ease of access for repeated sampling (close proximity to Danau Girang Field Centre) whilst ensuring a high degree of dispersion of transects, either by distance or separated by the Kinabatangan, to maximise sampling independence. Each site was visited prior to sampling to ensure transects were clearly labelled every 15 m; when necessary the flagging tape indicating the distance, was replaced. During the investigation, if disturbances such as tree falls occurred, the survey would continue on the same transect, staying as close as possible to the original path.

Anuran Recordings

To build upon previous studies in this area (Scriven, 2011; Gillespie *et al.,* 2012), surveys were conducted using the same methods. Nocturnal visual encounter (VES) and acoustic surveys were combined with area constraint transect sampling (10 m either side) to record species richness of each site. In the original studies, VES was chosen as it provides a higher number of individuals and species per unit effort in comparison with other sampling methods (Pearman *et al.,* 1995; Bell & Donnelly, 2006). To ensure the highest probability of detection of species presence at each site and to minimize false absences, both acoustic surveys and VES were used, as some species are often well hidden (Mazarolle *et al.,*2007).

Each transect was visited once per month for 11 months, commencing in September 2011. The order of sites visited was kept constant to avoid disturbance effects and surveying was concentrated in the middle of each month to record seasonal differences. In total, across 58 nights, 88 VES and acoustic surveys were completed, totaling 88.28 hrs of sampling effort. The duration of forest surveys was on average 80.65 min ± SE 0.06 compared to plantation surveys that lasted on average 39.72 min ±SE 0.03.

To maximize species detection, sampling occurred during the highest activity period for anurans (6.30pm till 10.00pm, Graeme Gillespie University of Melbourne, personal communication). A maximum of two transects were walked per night by the same two recorders. Both observers used H7 LED lenser© head torches (170 lumens, 3 watt CREE) to search for anurans within the forest floor (leaf litter, underneath logs and within forest debris), understory, mid story and within the canopy. Where possible, species were identified in the field (Inger & Stuebing, 2005; Frogs of Borneo, 2012). If conditions were not suitable for this, individuals were brought to Danau Girang Field Centre in plastic sealed bags for further examination. Photographs were taken and the individuals were subsequently released where they were originally found the next night.

Temperature and time were recorded at the beginning and end of the transect as well as moon presence, cloud cover, rain and wind, which have been shown to influence anuran activity (Heyer *et al*., 1994; Canessa *et al.,* 2012). The presence/absence of each environmental factor was recorded at the beginning, middle and end of the transects. Upon completion of each survey the individual factor scores were aggregated. Daily rainfall and maximum and minimum temperatures readings were also recorded at a meteorological station at Danau Girang Field Centre.

Statistical analysis

Detection efficiency of each survey was calculated by dividing the number of species found in each individual survey by the total number of species found at the respective sampling site over the entire study (Peirce and Gutzwiller, 2004). This provided a proportion of species present and detected for each sampling event and enabled comparisons across all sites. Cumulative detection efficiencies were also calculated for sampling events in chronological order for each habitat type and then averaged again to provide mean results of both habitats combined. For example the detection efficiency results of repeat surveying three times, were calculated nine times for each site to detect seasonal differences (the first using data from September, October and November and the second from October, November and December and so on until July). This enables us to find the optimal survey effort and the start month with the highest detection efficiency to begin a study, therefore finding the best combination if these to gain accurate estimates of species across habitats. The mean detection efficiency was calculated for each allocation of sampling effort in each habitat and subsequently plotted on a line graph (Peirce & Gutzwiller, 2004). The difference between the detection efficiencies of plantation and forest transects for single surveys was tested for significance using a parametric two sampled T test (Minitab version 15).

To evaluate influence that exogenous factors (habitat type; census number; start time; survey duration; moon, cloud, rain and wind presence; start temperature; daily rainfall and monthly rainfall) had on the total number of species detected at each sampling event a general linear model of the poisson family was created (R, Version 2.12.0). Data was confirmed to be normal by testing the residuals, thus meeting statistical assumptions (Neter et al., 1989, cited in: Pierce & Gutzwiller, 2004). The affect all factors had on the number of species detected by only calling/visually or by both on each survey was also analysed by GLMs however after data transformations (natural log, log 10, square root, exponential) data could not be normalised and analysis was terminated. Two combined graphs were plotted of monthly rainfall and (1) the average number of species detected upon each survey for each habitat per month and (2) the total number of individual species detected each month.
Sample based species accumulation curves and two incidence based non parametric species richness estimation methods, Chao2 and Jackknife (Chao, 1984, 1987; Smith & van Belle, 1984; Colwell & Coddington, 1994), of each site were produced using

Table 1. Summary table of all species detected each month on transects within both habitats (F=forest; P= Plantation) and the mode of detection (V=visually; A= acoustic); * indicates non-detection. The total number of species found per month are reported for each mode of detection.

Species	**Habitat**	**S**	**O**	**N**	**D**	**J**	**F**	**M**	**A**	**M**	**J**	**J**
Dicroglossidae												
Fejervarya cancrivora	P	*	V	V	V	V	V	V	*	V	*	*
Fejervarya limnocharis	FP	V	V	A	V	V	V	V	V	V	V	V
Limnonectes finchi	FP	V	V	V	V	V	V	V	V	V	V	V
Limnonectes ingeri	F	V	V	*	*	V	V	V	V	V	V	V
Occidozyga laevis	FP	V	VA	V	V	VA	V	V	VA	V	V	V
Microhylidae												
Chaperina fusca	F	*	V	*	*	V	*	*	*	V	*	*
Kalophrynus pleurostigma	F	*	*	*	*	*	V	*	*	*	*	*
Kaloula baleata	FP	*	VA	A	*	A	*	*	*	*	*	*
Metaphrynella sundana	F	A	*	*	*	*	*	V	*	*	*	*
Microhyla borneensis	F	V	V	V	V	V	VA	VA	VA	V	V	V
Microhyla perpava	F	V	V	V	V	V	V	V	V	V	V	V
Ranidae												
Hylarana erythraea	P	VA	VA	VA	V	VA	VA	V	V	V	V	V
Hylarana glandulosa	FP	VA	VA	VA	VA	VA	VA	VA	VA	VA	VA	VA
Hylarana nicobariensis	FP	VA	VA	VA	VA	VA	VA	VA	VA	VA	V	V
Hylarana raniceps	FP	V	V	V	V	V	V	V	V	V	V	V
Rhacophoridae												
Polypedates colletti	F	*	A	*	A	*	*	*	*	V	*	*
Polypedates leucomystax	FP	VA	VA	VA	VA	VA	VA	VA	VA	VA	VA	V
Polypedates macrotis	FP	V	V	*	V	VA	*	*	VA	V	*	V
Polypedates otilophus	F	V	V	V	V	V	*	*	*	*	*	*
Rhacophorusappendiculatus	FP	VA	VA	VA	VA	VA	VA	VA	VA	VA	VA	V
Rhacophorus dulitensis	F	*	*	VA	A	*	VA	VA	*	*	V	*
Rhacophorus pardalis	F	V	V	V	V	V	V	V	V	*	V	A
Chiromantis sp.nov.	F	*	*	A	A	VA	A	A	A	*	*	*
Visual detection		10	11	8	11	10	9	10	7	12	11	12
Acoustic detection		1	1	3	3	1	1	1	1	0	0	1
Visual and acoustic detection		5	7	6	4	8	7	6	7	4	3	1
Total		**16**	**19**	**17**	**18**	**19**	**17**	**17**	**15**	**16**	**14**	**14**

the transect data ('EstimateS'; Colwell, 2005). Fifty randomizations of data were completed to achieve species richness estimates. For Chao2 estimate, the bias corrected formula was used for all sites data apart from 'Site 19' for which the classic formula was used because the Chao's estimated incidence distribution (0.916) was larger than CV (0.5) (EstimateS, version Win820). Mao Tau mean estimations were used to create sample based rarefaction species accumulation curves with 95% confidence intervals and the actual species richness estimates were super imposed onto the graphs. The difference between the number of species detected and expected within forest sites compared with plantations sites was tested using parametric two sampled T tests (Minitab, version 15). A repeated measures two-way ANOVA was conducted, after a $\log_{10}$ transformation (Minitab, version 15), to evaluate the effect that habitat type and the number of censuses had on the difference between the observed and predicted species richness estimates.

Results

Species detected and habitat differences

In total, 23 species of frog, including one un-described species (*Chiromantis sp.nov.)*, belonging to four different families (Dicroglossidae, Microhylidae, Ranidae and Rhacophoridae) were detected during 88 surveys across two distinct habitats (Table 1; Fig. 2). Within the four secondary degraded forest transects, 21 species of anuran were identified, in comparison with 12 species on oil palm plantation transects. The observed species richness within forest transects was significantly higher than in plantations (T=5.42, DF=6, P=<0.001). This was also true when analysing the estimated total species richness (Chao2: T= 5.03, DF=6, P=<0.05; Jackknife: T=4.97; DF=6, P=<0.05)(Fig. 3 A-H). All species found within plantation sites, apart from *Fejervarya cancrivora* and *Hylarana erythraea* were also detected during forest surveys. The older plantations (Site 23 and 24, personal observation) had a higher species richness compared those with younger trees (Site 43 and 47)(Fig. 2).
Male calling was identified for 14 species, including *Microhyla borneensis*, for which the call was previously unknown (Inger & Stuebing, 2005) and second type of call from *Limnoncetes finchi* which is also undescribed (Inger & Stuebing, 2005). Several reproductive modes were seen including that of an undescribed, *Chiromantis* sp.nov. This species had not been found during surveys on the same transects from the previous study (Scriven, 2011). *Chiromantis* sp.nov was seen on transects only during the months of high rainfall (November until January, Table 1). However, calling

continued until April but due to their high location in the canopy, visualisation was not possible during the dryer months.

Table 2. Results of the GLM which was performed to evaluate factors that influenced the number of species found after each individual survey. Single term deletions were performed after each model, for each insignificant factor the values reported are those from the model before deletion.

Factor	DF	F value	P value	Significance
Habitat Type	1	51.60	2.50E-10	<0.001
Daily Rainfall	1	4.27	0.042	<0.05
Monthly Rainfall	1	29.64	5.07E-07	<0.001
Start time	1	2.14	0.16	>0.05
Moon presence	3	2.34	0.08	>0.05
N° of censuses	1	3.12	0.08	>0.05
Duration	1	1.89	0.17	>0.05
Start temp	11	0.95	0.50	>0.05
Site Number	6	0.88	0.51	>0.05
Rain presence	3	0.55	0.65	>0.05
Cloud presence	3	0.24	0.87	>0.05

Seasonality

A distinct pattern of seasonality of anuran detection was observed during the investigation (Fig. 4 A&B; Table 1). Seasonal rainfall in this region directly affected the number of individual species found per month in both habitat types (Table 1 & Fig. 4 A). Average monthly rainfall was 330ml (± SE 42) and monthly precipitation was 525ml at it's maximum in January, during which some low lying areas flooded (personal observation). Rainfall reduced to a minimum in June, when just 109ml was recorded. The cumulative number of species detected per survey for each month, peaked during December and January, the wettest two months of the investigation that together accounted for 29% of the total 11 month's rainfall (Fig. 4A).

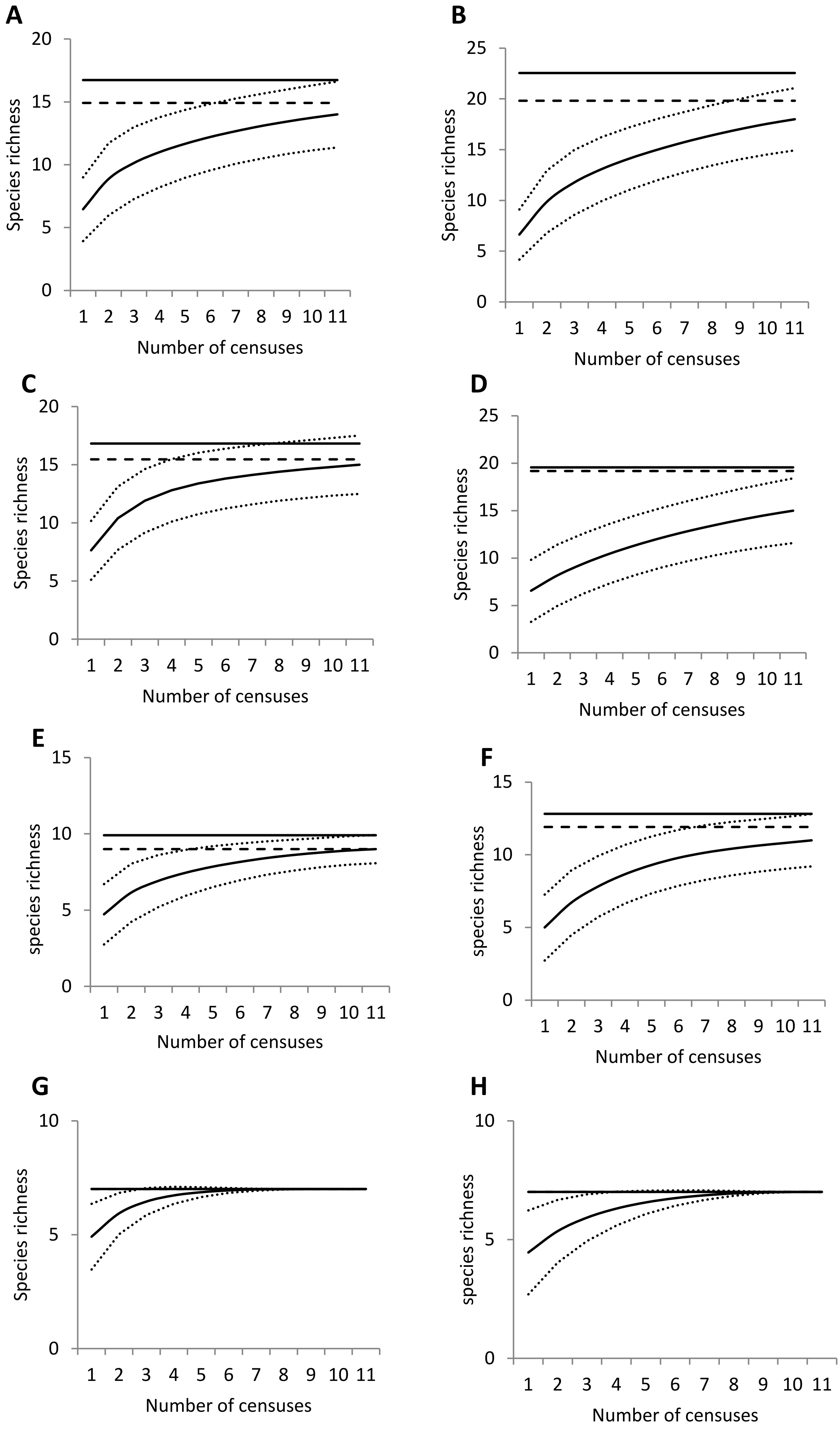
A
20
15
10
5
0
Species richness
1 2 3 4 5 6 7 8 9 10 11
Number of censuses
B
25
20
15
10
5
0
Species richness
1 2 3 4 5 6 7 8 9 10 11
Number of censuses
C
20
15
10
5
0
Species richness
1 2 3 4 5 6 7 8 9 10 11
Number of censuses
D
25
20
15
10
5
0
Species richness
1 2 3 4 5 6 7 8 9 10 11
Number of censuses
E
15
10
5
0
species richness
1 2 3 4 5 6 7 8 9 10 11
Number of censuses
F
15
10
5
0
species richness
1 2 3 4 5 6 7 8 9 10 11
Number of censuses
G
10
5
0
Species richness
1 2 3 4 5 6 7 8 9 10 11
Number of censuses
H
10
5
0
species richness
1 2 3 4 5 6 7 8 9 10 11
Number of censuses

Figure 3. (A-H) Sample based rarefaction curves with 95% confidence intervals (dotted lines) generated for forest sites (A-D) and plantation sites (E-H) using transect data after 50 randomizations. Total species richness estimates from two non-parametric methods, Chao2 (straight dashed) and Jackknife (straight solid) are superimposed onto each graph.

There were more species detected during the months before and within the rainy season (Table 1; Fig. 4B). After the peak months, the number of individual species found decreases across all study sites, following the reduction in monthly rainfall levels (Fig. 4B).

The factors significantly influencing the number of species found on each survey were habitat type daily rainfall and most significantly, monthly rainfall (Table 2). However, the number of species detected was not significantly influenced by the start temperature of each survey (Table 2). During the study period, daily maximum and minimum temperatures did not fluctuate widely, staying between 23°C and 37°C (DGFC Weather Station, 2012). The other abiotic factors of moon, rain, wind and cloud presence did not influence the number of species (Table 2). The number of repeat surveys completed at each site did not significantly affect the total number of

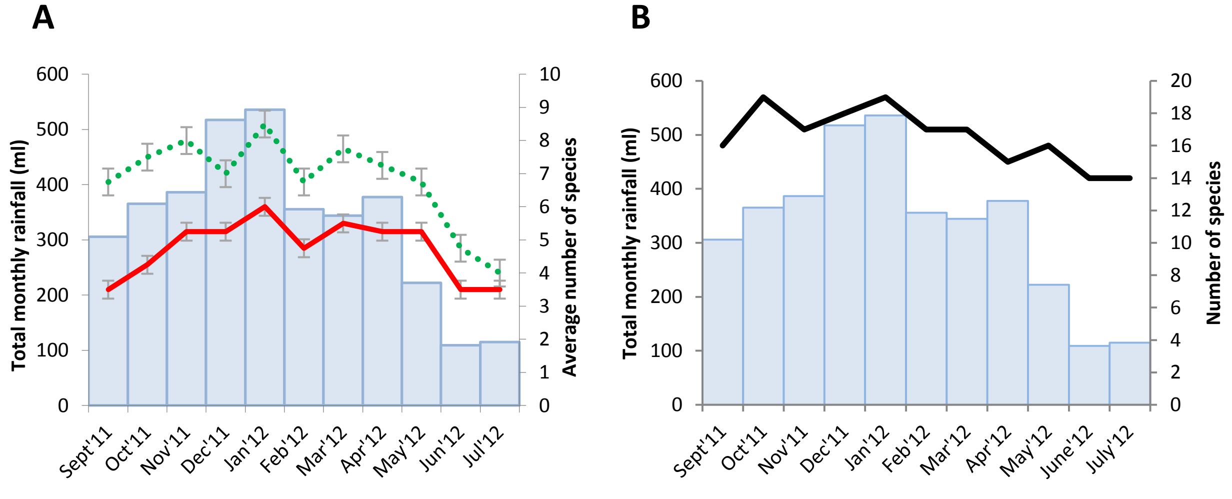

Figure 4, (A &B). (A) Average number of species detected each month in plantation (red solid line) and forest (green dotted line) per month with respect to rainfall (columns) with standard error bars. (B) The total number of individual species detected each month across both habitats with respect to monthly rainfall (columns). Data was collected from the study sites in the Lower Kinabatangan River, Sabah, Borneo from September 2011 – July 2012 species found either (Table 2).

In support of these results, there were several personal observations of explosive breeding and sightings of egg masses and individuals in amplexus occurring after heavy rains. Most notably, large calling aggregations of *Kaloula balaeta*, *Rhacophorus dulitensis* and *Chiromantis* sp. nov. only occurred after deep ground pools had formed. In contrast, other species such as *Rhacophorus appendiculatus* and *Hylarana glandulosa*, could be detected both acoustically and visually throughout the year (Table1).

Sampling effort

There was never 100% detection efficiency (DE) of species at any site on just one sampling event. The maximum and minimum DE from single surveys were higher in plantation transects (Max:86% Min:33%) than in forest transects (Max:80% Min: 11%). On average, the DE from single surveys was significantly higher (T=-4.12, DF=86, P=<0.001) in plantations (58% ±SE 0.02) compared with forest sites (44% ±SE 0.02). Plantation transects reached 100% in a minimum of two surveys, compared to at least five surveys required for forest transects (Table 3, A). To detect 90% of species in the area, forest transects required on average eight surveys compared with just five for plantation sites (Fig. 5). As expected, the rate of increase peaked for both habitats during the first five surveys but was slightly higher for forest sites (mean forest: 17% SE±6.7; mean plantation: 13% SE ±5) (Fig. 5). After this point, the percentage of species detected in forest transects continued to increase at a steady rate (mean: 3.5% SE ± 0.44), nearly three times faster than plantation detection efficiencies (mean: 1.3% SE ±0.33) (Fig 5.).

Species accumulation curves and Species richness estimates

The sample based rarefaction species accumulation curves for each site also illustrate the difference in the rate of change between habitats (Fig. 3 A-H). The two youngest plantation sites (43 & 47, Fig. 3, C & D) were the only transects to

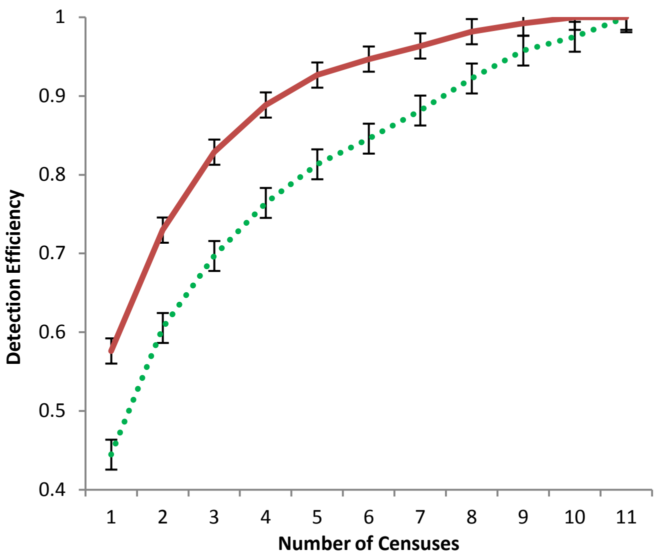

Figure 5. Average detection efficiency (proportion of anuran species detected) in relation to sampling effort (number of surveys) for both habitats, plantation (solid red) and forest (dotted green) with standard error bars. Graph is based on data from 88 surveys divided equally between the four sites in each habitat.

produce curves with clear asymptotes after 5 and 7 surveys respectively. The accumulation was more gradual and constant for forest transects and which show little evidence of asymptotion even after eleven censuses (Fig. 3 A-D). This continued increase of species accumulation in forest sites is further supported by the local estimations of species richness which predict across each site on average a maximum of four species are yet to be detected (Fig. 3, A-D). Whereas estimates predict on average no further species will be found within the plantation study sites (Fig. 3, E-H). However, there was no clear levelling of the curve for sites 23 and 24 (Fig.3, E&F) and therefore the predictions of actual species richness should be used with caution. The minimum number of censuses required to produce an accurate estimation of actual species richness was only found for sites 43 and 47 (Fig. 3, F & G). Only three surveys were required at sites 43 and 47 to produce a robust estimation using both Jackknife and Chao2 methods. Estimations from all forest sites and the two remaining plantation sites continued to increase with increasing census number. Therefore without asymptotion of the species accumulation curve, it would not be suitable to use predictions of actual species richness from the final survey as the true level of species richness to select a minimum sampling effort.

The ANOVA performed to show the interactions between observed versus predicted species richness and the habitat type and number of censuses, indicated both habitat and the number of censuses had a significant affect on the estimates of

species richness (habitat type: F=76.85, DF=1, P=<0.001; number of censuses: F=9.19, DF= 10, P=<0.001).

Sampling effort and seasonality combined

By combining the mean cumulative detection efficiencies per month for each habitat, we can see the start month and number of repeat surveys required to achieve a desired detection efficiency depending on the requirements of the monitoring program at hand (Table 3, A-C). To illustrate this, a minimum DE requirement of 80% (0.80) will be used. Table 3 (A) shows that on average to detect minimum of 80% of the anurans on plantation transects, just two surveys must be completed, beginning in December, resulting in on average 90% (±SE 6) detection. The table shows if the number of surveys are increased, the optimal start time changes to earlier in the year to includes the months before the peak rainy season. This pattern is also evident for the detection efficiencies of forest sites (Table 3, B). However to achieve 80% detection of anurans, on average at least three surveys must be completed beginning in November. Again, by increasing the number of surveys, the optimal start month is pushed back to include the months leading up to and including the rainy season. By combining the results of both habitats together (Table 3, C) to give the maximum detection of anurans with a minimal sampling effort, surveys must include the peak rainfall months and preferably commence before it.

Discussion

Results from this investigation support those of previous studies from the Lower Kinabatangan River which found significantly lower species richness of anurans in oil palm plantations compared to within secondary degraded forest fragments (Scriven, 2011; Gillespie *et al.,* 2012). The reduced species richness is most likely due to unsuitable habitat conditions within plantations (Scriven, 2011; Vitt & Caldwell, 2001) for most forest species, especially those with specific feeding (Scriven, 2011; Gillespie *et al.,* 2012; Suazo-Ortuño *et al*., 2008) and breeding (Greenberg, 2001; Suazo-Ortuño *et al*., 2008) requirements (Scriven, 2011). In contrast, the surrounding forest fragments, despite being at varying levels of degradation as a result of illegal and commercial logging (McMorrow & Talip, 2001) have high conservation value for local anuran communities (Gillespie *et al.,* 2012). As a result of the global decline of amphibians, the accurate monitoring of areas that provide havens for anuran populations is urgently needed (Beebee & Griffiths, 2005; Canessa *et al.,* 2012), especially within South East Asia (Rowley *et al.,* 2010). This investigation aimed to provide important information to incorporate into the future planning anuran monitoring programs in tropical environments with seasonal rainfall.

Our findings support previous studies from other seasonal tropical environments (Aichinger, 1987; Donnelly & Guyer, 1994; Bertoluci & Rodrigues, 2002; Prado *et al.,* 2005), suggesting rainfall is the most significant abiotic factor influencing the number of species detected during sampling events. The detection of anuran species is strongly influenced their breeding behaviour (Bertoluci & Rodrigues, 2002) which, for many species is heavily influenced by the availability of suitable aquatic sites (Aichinger, 1987). A combination of high monthly rainfall and heavy rain on the day of a survey results in higher species detection via either acoustic or visual identification. The reduced detection of anurans during the drier months also supports the theory that rainfall is the most important extrinsic factor influencing the reproductive behaviour of tropical anurans (Prado *et al.,* 2005). However, a variety of reproductive patterns including explosive and prolonged breeding (Wells, 1977) were witnessed throughout the year, supporting findings from previous studies in the Neotropics (Duellman & Trueb, 1986; Bertoluci & Rodrigues, 2002; Prado *et al.,* 2005) which show that not all species only breed during the wettest months. This temporal segregation could be a form of interspecific competition avoidance (Bridges & Dorcas, 2000) or a difference in breeding habitat requirements (Bandoni de Oliveira & Navas, 2004). Therefore if monitoring of areas in this location is restricted only to the months of high rainfall, several species may be left undetected, thus resulting in false absences that will diminish the integrity of the study (de Solla *et al.,* 2005; Mackenzie, 2005; Gomez-Rodrigues *et al.*, 2010). This further illustrates that prior knowledge of the ecology of species inhabiting a region should be taken into account if particular species are targeted for population monitoring (Canessa *et al*., 2012).

If the study is restricted to a small time period, the most optimal months to survey in would be those with high rainfall. However, this may cause logistical and health and safety complications associated with flooding that could result in a risk of reduced sampling effort and therefore weakening statistical power of the results (Pierce & Gutzwiller, 2004). Furthermore, if ground saturation occurs, the detection of litter dwelling species such as *M. borneensis* and *L. finchi* could be compromised (personal observation). Despite having a distinct rainy season in this region of Borneo, there are inter-annual variations in the wettest months (Scriven, 2011). Abnormal fluctuations in rainfall could have a direct effect on the detection of anurans which is known to vary between years (Prado *et al.,* 2005; Bridges & Dorcas, 2000). However, a longer term study would be required to confirm the extent of yearly variation of rainfall and the effect it has on anuran detectability in this region.

Other exogenous factors that have previously been found to influence anuran breeding activity (Heyer *et al*., 1994; Canessa *et al.,* 2012) and therefore detection, such as the presence of wind, cloud, moon and rain did not have significant effect on the number of species found. This suggests the inclusion of these factors is un-necessary during monitoring programs, however, further investigation is required because the effects could have been too low to detect statistically. In addition, there was also no significant effect of survey duration and start time, suggesting short term variations of individual surveys did not influence the number of species found which contrasts with previous findings (Pierce & Gutzwiller, 2004; Gooch *et al.,* 2006; Canessa *et al.,*2012). The fact that fluctuations in air temperature in this region did not significantly influence reproductive behaviour either contrasts with previous studies from more distinctly seasonal environments (Aichinger, 1987; Bertoluci & Rodrigues, 2002; Prado *et al.,* 2005) where cooler temperatures are known to inhibit breeding. However, results from this study are supported by data from similarly temperature stable locations where rainfall has a more dominant effect (Donnelly &Guyer, 1994; Bandoni de Oliveira & Navas, 2004). Although, the lack of influence of this factor may be a result of too narrow a temperature range in this region for it to significantly influence anuran behaviour.

In support of de Solla *et al. (*2005), these findings suggest single surveys of anurans in both plantations and forest transects do not provide accurate representations of species richness, even when taking place during peak detection months. The significantly lower species richness within oil palm plantations and the faster levelling of species accumulation curves when compared with forest transects, suggests fewer surveys are required to sufficiently obtain reliable estimations of species richness from these environments. The many irrigation ditches which were flooded for the majority of the year (personal observation) in oil palm plantations may provide sufficient all year round breeding environments for the species present in these locations thus resulting in high detection efficiencies with minimal sampling effort throughout the year. To gain a reliable representation of anuran communities inhabiting oil palm plantations at this location results suggest no more than three surveys taking place during and before the rainy period would be required.

Contrastingly within forest transects, a higher level of sampling effort is needed to provide an accurate total of species present. Despite the fact significantly more species are present in forest transects, a previous study found no significant difference in abundance of anurans across both habitats (Scriven, 2011). Therefore the detection probability of all species in forest sites is potentially lower than that of plantation transects, thus more surveys may be required to reduce the possibilities of false absences. Habitat characteristics of forest transects, such as dense vegetation made both the acoustic and visual identification of species more difficult. This may be an additional explanation for the significantly lower detection efficiencies during single surveys of forest sites compared with the openness of plantations. However, the duration of forest surveys was extended to account for the increase in complexity of this habitat. Despite this, survey duration was not found to significantly influence the number of species found per survey, contrasting with findings from Peirce and Gutwiller (2004).

Species accumulation curves and the estimations of total species richness in forest sites both suggest even after 11 surveys on the same transect, throughout the year, not all species present in the area were detected. At least four or fives surveys during peak activity months are required to achieve a high level of detection. The fact that species richness remains a function of sampling effort in the majority of sites throughout the investigation, suggests additional surveillance is required to provide an accurate and reliable estimation of total species richness. This was further confirmed by personal observations of species in forest fragments found outside of surveying time or transect area. Future investigations could use the findings of this study and apply them to different methods of detection to evaluate their efficiency in these environments.

The results of this study contrast with previous findings that two or three repeat surveys of a site are sufficient (Field & Tyre *et al.,*2005) but is in support of results de Solla *et al.*(2005) who found that even after removing species that are difficult to detect, three surveys did not adequately represent the focal anuran community. This has implications for the design of future survey and census programs. However, the findings do suggest that statistical models of species richness can provide accurate representations of anuran communities without over expenditure of time or budget (Garrard *et al*., 2008; Canessa *et al.,* 2012) in some habitats. Problems may arise during statistical analysis when comparing two distinct habitats if the sampling effort of one is lower than the other. Therefore, to overcome this problem and provide important information to incorporate into the methodologies of future anuran monitoring programs in this region I have we have combined both the seasonality data with information on sampling effort. Results of which suggest, to yield high detection efficiencies with minimal effort, surveys should ideally commence before the rainy season and continue throughout the period of high rainfall. For example, if the sampling effort allocated is two surveys per habitat type, the sampling should take place during December and January. If the number of surveys is increased, it is more beneficial to include the months before peak rainfall rather than after it despite similar total rainfall during the months either side of the highest precipitation period. This display of high anuran activity before and during the wettest months is supported by findings from Prado *et al.* (2005).

The new species and behaviours witnessed during this investigation confirm the lack of knowledge of amphibians in this region (Gillespie *et al.,* 2012). Despite being heavily altered over recent years (Mcmorrow & Talip ,2001), this study highlights the potential for new discoveries to be made within forest fragments of the Lower Kinabatangan Wildlife Sanctuary and the surrounding area. Further surveys and an inventory of amphibians in this region is crucial to demonstrate areas that support high levels of biodiversity that should designated for wildlife conservation (Beebee & Griffiths, 2005). Results of this investigation can be incorporated into the future planning of anuran investigations in seasonal lowland locations to enhance their efficiency. By combining the knowledge of seasonal breeding patterns with the designation of an appropriate sampling effort, an accurate and reliable estimation of species richness can be produced. In addition, the use of statistical models to estimate total species richness and reduce the possibility of false absences would give further significance to the results.

Acknowledgements

I would firstly like to thank Dr. Benoit Goossens my supervisor, who gave me this incredible opportunity for which I am so grateful for. I would also like to thank my core supervisor Dr. Graeme Gillespie for his help with initially designing the project and throughout the year for his advice and thorough knowledge of the subject. I would also like to thank PhD students Danica Stark and Luke Evans for their help with creating maps and statistical analysis. I could not have completed this project if it wasn’t for my fantastic research assistant Samsir Laimun who accompanied me on every survey and demonstrated incredible forest skills and knowledge of local amphibians. I would also like to thank all the staff and students at Danau Girang Field Centre, including my fellow PTY students Alice Miles and Becky Lawrence who also assisted me during surveys.

References

Aichinger, M. (1987). Annual activity patterns of anurans in a seasonal neotropical environment. *Oecologia*, **71**: 583–592.

Ancrenaz, M., Goossens, B., Gimenez, O., Sawang, A. & Lackman-Ancrenaz, I. (2004). Determination of ape distribution and population size using ground and aerial surveys: a case study with orang-utans in lower Kinabatangan, Sabah, Malaysia. *Animal Conservation*. **7**: 375-385

Aratrakorn, S., Thunhikorn, S., Donald, P.F. (2006). Changes in bird communities following conversion of lowland forest to oil palm and rubber plantations in southern Thailand. *Bird Conservation International*. **16**: 71–82.

Bandoni de Oliveira, F., & Navas, C. A. (2004). Plant Selection and Seasonal Patterns of Vocal Activity in Two Populations of the Bromeligen Treefrog Scinax perpusillus Plant Selection and Seasonal Patterns of Vocal Activity in. *Journal of Herpetology*, **38***:* 331–339.

Beebee, T. J. C., & Griffiths, R. A. (2005). The amphibian decline crisis: A watershed for conservation biology? *Biological Conservation*, **125:** 271–285.

Bell, K.E. & Donnelly, M.A., (2006). Influence of forest fragmentation on community structure of frogs and lizards in northeastern Costa Rica. *Conservation Biology*. **20**: 1750–1760

Bertoluci, J., & Rodrigues, M. T. (2002). Seasonal patterns of breeding activity of Atlantic Rainforest anurans at Boracéia , Southeastern Brazil. *Amphibia-Reptilia*, **23:**161–167.

Bridges, A. S., & Dorcas, M. E. (2000). Temporal Variation in Anuran Calling Behavior : Implications for Surveys and Monitoring Programs. *Copeia*, **2**:587–592.

Canessa, S., Heard, G. W., Parris, K. M., & McCarthy, M. A. (2012). Integrating variability in detection probabilities when designing wildlife surveys: a case study of amphibians from south-eastern Australia. *Biodiversity and Conservation*, **21:** 729–744.

Chao, A. (1984). Non-parametric estimation of the number of classes in a population. *Scandinavian Journal of Statistics* **11**: 265-270.

Chao, A. (1987). Estimating the population size for capture-recapture data with unequal catchability. *Biometrics* **43:** 783-791.

Colwell, R. K. (2005). EstimateS: Statistical estimation of species richness and shared species from samples. Version 8.2. User's Guide and application published at: http://purl.oclc.org/estimates

Colwell, R.K. & Coddington, J.A. (1994). Estimating terrestrial biodiversity through extrapolation. *Philosophical Transactions of the Royal Society (Series B)* **345**: 101–118.

Chung, A.Y.C., Eggleton, P., Speight, M. R., Hammnd, P. M. & Chey, V. K., (2000). The diversity of beetle assemblages in different habitat types in Sabah. *Malaysian Bulletin of Entomological Research*. **90**: 475–496

de Solla, S. R., Shirose, L. J., Fernie, K. J., Barrett, G. C., Brousseau, C. S., & Bishop, C. a. (2005). Effect of sampling effort and species detectability on volunteer based anuran monitoring programs. *Biological Conservation*, **121:** 585–594.

Donnelly, M. A., & Guyer, C. (1994). Patterns of reproduction and habitat use in an assemblage of Neotropical hylid frogs. *Oecologia*, **98:** 291–302.

Dorcas, M. E., Price, S. J., Walls, S. C., & Barichivich, W. J. (2010). Auditory monitoring of anuran populations. In: K. Dodd (ed.). *Conservation and Ecology of Amphibians. pp.* 281-298. Oxford University press, Oxford, UK.

Duellman, W. E. & Trueb, L. (1986). *Biology of Amphibians.* McGraw-Hill Publishing Company, New York.

Fayle, T. M., Turner, E. C., Snaddon, V.K., Chey, A.Y.C., Chung, P., Eggleton, W.A. & Foster, W.A. (2010). Oil palm expansion into rain forest greatly reduces ant biodiversity in canopy, epiphytes and leaf-litter. *Basic and Applied Ecology*. **11**: 337–345

Field, S. A., Tyre, A. J., Thorn, K. H., O'Connor, P. J., & Possingham, H. P. (2005). Improving the efficiency of wildlife monitoring by estimating detectability: a case study of foxes (*Vulpes vulpes*) on the Eyre Peninsula, South Australia. *Wildlife Research*. **32**: 253- 258.

Fitzherbert, E. B., Struebig, M. J., Morel, A., Danielsen, F., Brühl, C. A, Donald, P. F., & Phalan, B. (2008). How will oil palm expansion affect biodiversity? *Trends in ecology & evolution*. **23:** 538–45.

Frogs of Borneo (2012) See: www.frogs ofborneo.org (date last accessed 1th July 2012).

Frost,D. R. (2009) Amphibian species of the world: an online reference, version 5.3. New York, NY: American Museum of Natural History. See research.amnh.org/herpetology/ amphibia/index.html

Garrard, G. E., Bekessy, S. A., McCarthy, M. A., & Wintle, B. A. (2008). When have we looked hard enough? A novel method for setting minimum survey effort protocols for flora surveys. *Austral Ecology*. **33***:* 986–998.

Gibbons, J.W., Burke, V.J., Lovich, J.E., Semlitsch, R.D., Tuberville, T.D., Brodie, J.R., Greene, J.L., Niewiarowski, P.H., Whiteman, H.H., Scott, D.E., Pechman, J.H.K., Harrison, C.R., Bennett, S.H., Krenz, J.D., Mills, M.S., Buhlmann, K.A., Lee, J.R., Seigel, R.A., Tucker, A.D., Mills, T.M., Lamb, T., Dorcas, M.E., Congdon, J.D., Smith, M.H., Nelson, D.H., Dietsch, M.B., Hanlin, H.G., Ott, J.A. & Karapatakis, D.J. (1997) Perceptions of species abundance, distribution, and diversity: lessons from four decades of sampling on a government-managed reserve. *Environmental Management* **21**: 259–268.

Gillespie, G. R., Ahmad, E., Elahan, B., Evans, A., Ancrenaz, M., Goossens, B., & Scroggie, M. P. (2012). Conservation of amphibians in Borneo: Relative value of secondary tropical forest and non-forest habitats. *Biological Conservation*, **152:** 136–144.

Gooch, M., Dorcas, M., Price, S., & Heupel, A. (2006). The effects of survey protocol on detection probabilities and site occupancy estimates of summer breeding anurans. *Applied Herpetology*, **3:** 129–142.

Glor, R.E., Flecker, A.S., Benard, M.F. & Power, A.G. (2001). Lizard diversity and agricultural disturbance in a Caribbean forest landscape. *Biodiversity Conservation*. **10**: 711–723

Gómez-Rodríguez, C., Guisan, A., Díaz-Paniagua, C., & Bustamante, J. (2010). Application of detection probabilities to the design of amphibian monitoring programs in temporary ponds, A*nnales Zoologici Fennici,* **47**: 306–322.

Greenberg, C. H. (2001). Response of reptile and amphibian communities to canopy gaps created by wind disturbance in the southern Appalachians. Forest Ecology and Management **148:** 135–144

Heyer, W.R., Donelley, M.A., McDiarmid, R.W., Hayek, L.C. & Foster, M.S. (1994). Measuring and monitoring biological diversity: standard methods for amphibians. Smithsonian Institution Press, Washington, D.C

Hutton, W. (2004) *Kinabatangan: Sabah Colour Guide.* Natural History Publications (Borneo), Malaysia.

Inger, R. F. & Stuebing, R.B. (2005). *A Field Guild to the frogs of Borneo. Second Edition*. Natural History Publications. Borneo.

IUCN (2012). IUCN Red List of Threatened Species, version 2012.1. See www.iucnredlist.org (date last accessed 7th July 2012).

Koh, L.P. & Wilcove, D.S. (2007) Cashing in palm oil for conservation. *Nature* **448**: 993–994

Mackenzie, D. I., Nichols, J. D., Lachman, G. B., Droege, S., Royle, J. A., & Langtimm, C. A. (2002). Estimating site occupancy rates when detection probabilities are less that one. *Ecology*, **83:** 2248–2255.

Mackenzie, D. I., & Royle, J. A. (2005). Designing occupancy studies: general advice and allocating survey effort. *Journal of Applied Ecology*, **42:** 1105–1114.

Mazerolle, M.J., Bailey, L.L., Kendall, W.L., Royle, A.J., Converse, S.J. & Nichols, J.D. (2007) Making great leaps forward: accounting for detectability in herpetological field studies. *Journal of Herpetology* **41:**672–689

McMorrow, J.& Talip, M.A., (2001). Decline of forest area in Sabah, Malaysia: relationship to state policies, land code and land capability. *Global Environmental Change* **11:** 217–230.

Maddox, T. M., Gemita,E., Wijamukti, S. & Selampassy, A. (2007). The conservation of tigers and other wildlife in oil palm plantations, Jambi Province, Sumatra, Indonesia. *Zoological Society of London. Conservation Report* **7**: 1-62

Marsh, D.M. & Haywood, L.M.B, (2010) Area-based surveys, In: *Amphibian Ecology and Conservation,* ed. Dodd, C.K. *et al.* pp. 247-262. Oxford University Press, New York

Mazerolle, M.J., Bailey, L.L., Kendall, W.L., Royle, A.J., Converse, S.J. & Nichols, J.D. (2007) Making great leaps forward: accounting for detectability in herpetological field studies. *Journal of Herpetology,* **41:** 672–689

Pearman, P.B., Velasco, A.M. & López, A. (1995). Tropical amphibian monitoring: a comparison of methods for detecting inter-site variation in species composition. *Herpetalogica*. **51**: 325-337.

Pierce, B. A., & Gutzwiller, K. J. (2004). Auditory Sampling of Frogs: Detection Efficiency in Relation to Survey Duration. *Journal of Herpetology*, **38:** 495–500.

Prado, C. Haddad, C. & Uetanabaro, M. (2005). Breeding activity patterns, reproductive modes, and habitat use by anurans (Amphibia) in a seasonal environment in the Pantanal, Brazil. *Amphibia-Reptilia*, **26:** 211–221.

Rowley, J., Brown, R., Bain, R., Kusrini, M., Inger, R., Stuart, B., Wogan, G., Thy, N., Chan-ard, T., Trung, C.T., Diesmos, A., Iskandar, D.T., Lau, M., Ming, L.T., Makchai, S., Truong, N.Q. & Phimmachak, S., (2010). Impending conservation crisis for Southeast Asian amphibians. *Biology Letters,* **6:** 336–338.

Scott, D. A. (ed). (1989). *A Directory of Asian Wetlands,* IUCN, Gland, Switzerland.

Scriven, S. (2011) Biodiversity Conservation in Oil Palm Plantations and Fragmented Secondary Forests: A Study of Bornean Anurans. Unpublished MRes Thesis, Cardiff University.

Smith, E.P. & van Belle, G. 1984. Nonparametric estimation of species richness. *Biometrics* **40 :** 119-129.

Sodhi, N.S. & Brook, B.W. (2006). *Southeast Asian Biodiversity in Crisis*. Cambridge (United Kingdom): Cambridge University Press.

Sodhi, N.S., Koh, L.P., Brook, B.W. & Ng, P.K.L. (2004). Southeast Asian biodiversity: an impending disaster. *Trends in Ecology and Evolution*. **19**: 654-660.

Sodhi, N. S., Posa, M. R. C., Lee, T. M., Bickford, D., Koh, L. P., & Brook, B. W. (2010). The state and conservation of Southeast Asian biodiversity. *Biodiversity and Conservation*, **19:** 317–328.

Stuart, S.N., Chanson, J.S., Cox, N.A., Young, B.E., Rodrigues, A.S.L., Fischman, D.L., & Waller, R.W. (2004). Status and trends of amphibian declines and extinctions worldwide. *Science*. **306**: 1783-1786.

Suazo-Ortuño, I., Alvarado-Díaz, J. and Martínez-Ramos, M. (2008). Effects of conversion of dry tropical forest to agricultural mosaic on herpetofaunal assemblages*. Conservation Biology.* **22**: 362–374.

Ugland, K. I., Gray, J. S., & Ellingsen, K. E. (2007). The species-accumulation curve and estimation of species richness. *Journal of Animal Ecology*, **72:** 888–897.

Vitt, L.J., & Caldwell, J.P., 2001. The effects of logging on reptiles and amphibians of tropical forests. In: Fimbel, R.A., Grajal, A., Robinson, J. (eds.), *The Cutting Edge: Conserving Wildlife in Logged Tropical Forests*. Columbia University Press, New York, pp. 239–259.

Watson, C. A., Weckerly, F. W., Hatfield, J. S., Farquhar, C. C., & Williamson, P. S. (2008). Presence-nonpresence surveys of golden-cheeked warblers: detection, occupancy and survey effort. *Animal Conservation*, **11:** 484–492

Wells K. D. (1977). The social behaviour of anuran amphibians. *Animal Behaviour* **25**:666–693.

Wells, K. D. (2007). *The ecology and behaviour of amphibians*. The University of Chicago Press.