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Review Article

Contributions to local herpetology in Great Britain – The Kent Reptile and Amphibian Group

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Wildlife conservation in Great Britain is supported by a host of voluntary bodies, some of which are devoted specifically to the island's herpetofauna. The administrative units of the three countries that comprise Great Britain (England, Scotland and Wales) are called 'counties' and of the 82 counties (this figure can vary according to how they are defined) 71 have Amphibian and Reptile Groups (ARGs). All are staffed by volunteers. These ARGs are affiliated to, and co-ordinated by, ARG UK (http://www.arguk.org/). The ARGs are grouped into regions that have their own annual meetings.

The ARGs all have similar conservation objectives but differ considerably in size and vary from being well established to nascent. One of the larger ARGs is based in the county of Kent in the south east corner of England. To give itself a more pronounceable acronym it swapped the letters of ARG around to become 'KRAG'; The Kent Reptile and Amphibian Group. The geographical identity of the Group seems only to have been confused once, this was when an enquirer sent a photo of a snake recently decapitated by her husband with a large axe. She asked for a species identification. The animal turned out to be a canebreak rattlesnake (*Crotalus horridus*) from Kent County, but in Texas not England!

This article describes the creation of KRAG, its functions and its contribution to local herpetology. All of Britian's widespread amphibian and reptile species live within the borders of Kent (Table 1).

At one time two of Britain's localised species, the sand lizard (*Lacerta agilis*) and natterjack toad (*Epidalea calamita*) also

Amphibia		
Anura	*Rana temporaria*	Common frog
	Bufo bufo	Common toad
Caudata	*Triturus cristatus*	Great crested newt
	Lissotriton vulgaris	Smooth newt
	Lissotriton helveticus	Palmate newt
Reptilia		
Lacertilia	*Anguis fragilis*	Slow worm
	Zootoca vivipara	Viviparous lizard
	Natrix natrix helvetica	Grass snake
	Vipera berus	Adder

Table 1. Herpetofauna native to Kent.

a. 1991 - 2006 b. 2007 to date c. Corporate member d. Conservation partner

Figure 1. The logos adopted by KRAG (a. & b.) and its specialised membership categories (c. & d.).

occurred naturally in Kent but were believed to be extirpated by the 1960s. They have recently been reintroduced at a few selected locations using specimens from elsewhere in England. Besides the native fauna there are a number of exotics in Kent that include, amongst others, the marsh frog (*Pelophilax ridibundus*), the wall lizard (*Podarcis muralis*) and the red-eared terrapin (*Trachemys scripta elegans*).

The evolution of KRAG

KRAG came to life following the efforts of founder members J.F.D Frazer and G.A.N. Davis. Deryk Frazer was a former president of the British Herpetological Society (from which he retired in 1981) and a very active Kent recorder, contributing over 600 records. In 1988, Frazer and Davis proposed that the Kent Wildlife Trust (formerly Kent Trust for Nature Conservation) needed a group dedicated to the conservation of herpetofauna and one was established with a logo showing a common toad and grass snake in front of a Kentish oast - an iconic Kentish farm building used for drying the hops used in beer making (Fig. 1a). The Group had the following aims:

1. Undertake survey work to map species distribution
2. Offer advice on planning applications that may affect species
3. Offer advice on general conservation measures
4. Raise awareness of amphibian and reptile conservation with local people
5. Mount 'rescues' of animals that may be in danger from development activities

Since its origins in 1988 the Group has had its ups and downs. The Kent Wildlife Trust had to re-establish KRAG in 1996 but since then it has gone from strength to strength as an active membership organisation. By 2007, the Group's future seemed assured and was celebrated by the adoption of a new logo. This retains the original elements but presents them in a new format (Fig. 1b). KRAG objectives have also evolved so that members no longer undertake development-based rescues (these are best handled by professional ecological consultants) and they have only limited time available to offer advice on planning applications (there are simply too many in Kent!), the remainder of those early objectives still remain very relevant. KRAG currently promotes reptile and amphibian conservation by:

1. Recording the distribution and monitoring the status of herpetofauna in the county using its own database
2. Providing general advice on reptiles and amphibians and their habitat management to relevant organisations and the general public, and
3. Raising awareness amongst the general public

However, these reduced aims do not reflect the breadth of activities actually undertaken by KRAG, as detailed below.

KRAG business is dealt with by an honorary Committee elected by its membership for a 3 year renewable term (current post holders and some contact details are shown in Annex 1). In 2012, it was decided that the Group would benefit for having a president to advise and support the Chair and offer other help on an *ad hoc* basis. The first president is Dr Lee Brady, county recorder for reptiles and amphibians, twice Chairman of the Kent Field Club and former Chairman of KRAG (2005 - 2010). The Group is governed by a constitution that is posted on the KRAG website (http://www.kentarg.org/KRAG-Information/View-

category).

KRAG offers various categories of membership. For many years the only category was Ordinary Member; this is still the mainstay of the Group and offered to individuals for an annual subscription (currently £7.50). In return members receive a biannual newsletter and invitations to field days and training events organised by the Group. There has been a steady growth in the number of Ordinary Members which for the first time exceeded 200 in 2012 (Fig. 2), with a 60:40 split between men and women. In 2006, the category of Corporate Member was created for ecological consultancies and other commercial bodies. These sign a pledge of 'good practice' and contribute an annual fee of £50. In return Corporate Members may display a special version of the KRAG logo (Fig. 1c) and, provided that they submit faunal records to the KRAG database (explained in more detail in the next section), they can request searches of the database free of charge. A third category of membership, Conservation Partner, was launched in 2010 for non-profit organisations whose conservation objectives are aligned with those of KRAG. This category of membership is free of any subscription, offers free database searches and allows the Partners to display their own version of the KRAG logo (Fig. 1d).

Recording for conservation

The centre piece to KRAG's work is a database of faunal records. The database holds over 35,000 records and these are being used as the basis to ecological appraisal of development activities, to plan and manage conservation projects and to designate important herpetofauna sites across the county; at last count there were

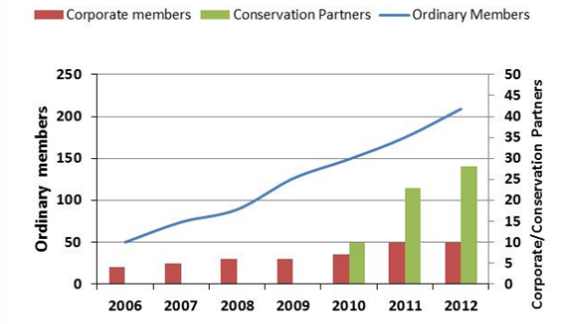

Figure 2. The development of the various categories of KRAG membership since 2006.

Figure 3. Conventional dot map showing the distribution of adders in Kent, generated from the KRAG database.

42 Key Amphibian Sites and 56 Key Reptile Sites.

The database receives records from diverse sources. There is a data sharing agreement with Kent and Medway Biological Recording Centre but many more are provided by KRAG members, other recorders and the general public, especially those participating in county events that are also attended by the KRAG stand (see below). A suitably configured iPad is available on the stand so that people can enter records based on their post codes. There is also a facility on the website that allows on-line submission of records using post codes or map references. These approaches result in a rapid accumulation of data but there is a bottleneck in the system as each record must be carefully validated before final entry into the database. Records of very common, easily identified species observed within their existing range are readily accepted while those that are more difficult to distinguish and/or are reported outside their normal range require further validation, usually by a site visit.

KRAG undertakes database search requests for those organisations and individuals needing access to this important information. There is a database search request form that can be completed on the website. Results come back with a list of species records, a Google Earth map to enable a quick review, and species range assessment score. The range assessment score classifies species presence using nearest neighbour analyses, with predictions summarised using the following categories:

• core range - nearest neighbour distance within which 75% of observations occur
• predicted range - nearest neighbour distance

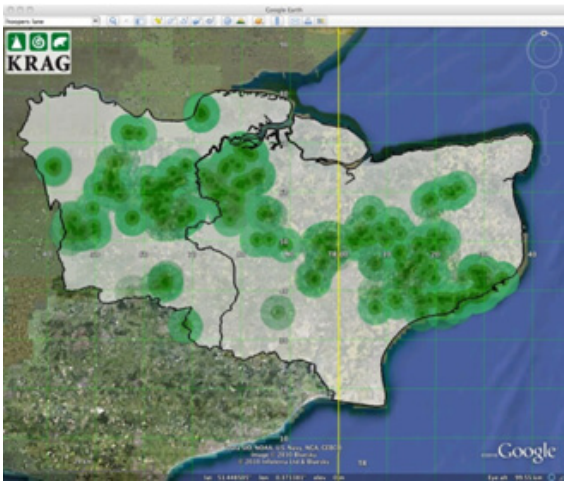

Figure 4. Range assessment map of adders in Kent, generated by the KRAG database (darker circles represent core range, lighter circles represent predicted range, lightest circles maximum expected range).

within which 95% of observations occur
• maximum expected range - distance from most isolated observation to nearest neighbour

This system is more informative than a conventional distribution map consisting of a series of dots such as the distribution of adders in Kent (Fig. 3), as such maps only hint at the true extent of a species' range.

The range map for adder appears instead as a more blurred picture indicating the probability of the species occurring in any particular locality (Fig. 4). The maximum expected range of a species relies on calculating distance to outliers. For species with isolated records (particularly where this is a result of under recording), these outliers can result in a significantly exaggerated range and so should be used with caution. Also, the likely presence of a species may be overstated when the site in question, although very close to an existing confirmed record, does not display any suitable habitat for the species in question.

In an attempt to provide better control of differences in recording effort, the range assessment score is modified by an assessment of the quality of available habitat (1 km square resolution) to generate a 'Likelihood of Presence Score'. Habitat quality includes several factors (e.g. broadleaf woodland cover) and for each species a landscape level Habitat Suitability Index is calculated from which are derived the following predictions of presence - Unlikely, Possible, Likely, High.

For users of the database search service the result is a table (Fig. 5) showing the Likelihood of Presence Score and the distance to closest record for each species.

Database searches are provided free of charge to Conservation Partners, Corporate Members who submit records to the database, and to organisations engaged in education or non-commercial conservation work. For others there is a modest charge which is used to cover KRAG's costs, with any excess used to support KRAG's conservation projects. In recent year there have been large numbers of database searches; 426 to date in 2013.

Facing the outside world

KRAG's interface with the outside world is through its website, biannual newsletter, the contribution of a display stand to local events around the county, its training courses, and more recently through the use of social media.

Website

No modern group can have a public profile without a website. KRAG's website has evolved over the years with the help of an external consultant on whose wind-powered server the site is hosted. The website offers information on the fauna, current events, membership options and facilities to pay subscriptions online. It also has sections for the submission of faunal records on-line and for requests for database searches. As searches are semi-automated and returned electronically as PDFs, the turn-around time is very rapid, quite often within 24 h.

Newsletter

The KRAG newsletter started life a few pages

Amphibians

	Likelihood of Presence Score	Dist (km)
Common Frog:	HIGH	1.14
Common Toad:	HIGH	2.02
Natterjack:	n/a	68.02
Smooth Newt:	Possible	2.52
Palmate Newt:	HIGH	2.52
Great Crested Newt:	Possible	4.57
Marsh Frog:	*unlikely*	*8.54*
Alpine Newt:	n/a	26.99

Amphibian survey effort in local area is considered to be relatively low. Results should be interpreted with caution.

ponds within 1 km: 9
distance to nearest pond (km): 0.24

Reptiles

	Likelihood of Presence Score	Dist (km)
Viviparous Lizard:	Possible	1.90
Slow-worm:	Possible	1.84
Sand Lizard:	*unlikely*	*69.35*
Grass Snake:	Possible	1.77
Adder:	HIGH	0.71
Smooth Snake:	n/a	n/a

Reptile survey effort in local area is considered to be below average. Results should be interpreted with caution.

Figure 5. Table of distances to closest records and likelihood of presence on a given site, achieved by a combination of nearest neighbour analysis tempered by habitat suitability.

of text with black and white illustrations published on an *ad hoc* basis. It has gradually become a substantial glossy document in full colour published in spring and autumn. Some editions focus on specific topics and pride is taken in some excellent photos taken by the membership, such as the toad crossing a road (Fig. 6). In keeping with sustainability concerns, the newsletter is now delivered electronically as a PDF file with just a few printed copies posted to members lacking internet connections and for display on the stand at events attended by KRAG. In an age that offers a constant stream of news, some people consider newsletters to be 'old hat' and a static form of communication, but they provide great snapshots over time, and many people like to receive something tangible for their subscription.

Display stand

The display stand (Fig. 7) is the workplace of a dedicated band of volunteers prepared to spend long days at events in Kent, such as the biannual Kent Garden Show, the Kent Anglers' Show, events organised by the Royal Society for the Protection of Birds, etc..

The volunteers talk to passers-by, encouraging their interest in amphibians and reptiles, and collect faunal records from them. The wealth of information available from the public is invaluable, and many people are proud of the herps that they have found in their gardens, yet really don't think that anyone is "interested in an ordinary frog". This is a valuable opportunity to introduce the public to the importance of faunal recording and explain how it supports local decision making.

The stand is graced by excellent photos of our fauna, however, there is an on-going debate about the pros and cons of displaying live animals; specifically slow worms (*Anguis fragilis*). It is accepted that there are substantial benefits in encouraging children to interact with live animals, to the extent that it is probably a major factor in recruiting wildlife enthusiasts for the future. But balanced against this is the concern that KRAG should neither encourage pet keeping of the native fauna nor subject animals to any undue stress. KRAG is currently preparing its own policy position on this while in the meantime there is a moratorium on having live animals on display.

Training courses

Of crucial importance to KRAG's aims of raising awareness and promoting recording are the training programmes and illustrated talks that are a regular part of the calendar of events run by the Group. Resources have been created that act as 'off the shelf' presentations to assist as many of the Group as possible to give talks and training sessions. Short talks and workshops are used as ways of collecting casual records from attendees as well as developing their interest in herpetofauna that later leads many to attend full-day courses. These longer courses are linked to KRAG's conservation projects and provide participants with the skills to actively record in their own localities (Fig. 8). Training is always offered free of charge to potential recorders and introductory courses typically include sessions on the identification and ecology of animals as well as recording techniques and how to identify suitable habitat. The training given as part of the Great Crested Newt Monitoring Project also provides trainees with the necessary skills to be made agents under KRAG's GCN licence from Natural England for torching, netting and egg searches (necessary as GCN enjoys full legal protection).


KRAG news

NEWSLETTER OF THE KENT REPTILE AND AMPHIBIAN GROUP

TOAD PATROL SPECIAL EDITION!

Figure 6. The biannual newsletter keeping the membership informed of KRAG's own news and with stories from elsewhere.

Figure 7. KRAG volunteers manning the stand at a an event to encourage wildlife recording.

Social Media

Assisted by technology and the rise of social media, KRAG's aims now go beyond its membership. At the time of writing, KRAG has over 300 members on its Facebook page, as well as 307 followers on Twitter with a count of 1043 tweets up to August 2013. These are great forums for debating current issues and give KRAG an insight into the priorities of those working outside of the Group.

Projects for Conservation

Great Crested Newt (GCN) project

Although GCN (*Triturus cristatus*) receives full legal protection, unless the geographical range and the density of populations around the county are known there is little that can be done to safeguard the habitat of these animals. This project is improving our understanding of great crested newts by surveying ponds. Data on population sizes at ponds has helped to improve our knowledge of newts metapopulations and, when identified, source ponds (those where newts breed successfully and disperse) and sink ponds (those where newts exist but do not breed in large numbers) are highlighted and relative emphasis can be given to their value for GCN. KRAG's objectives for this project include:

- To train volunteers in amphibian ecology and survey techniques
- Provide opportunities for volunteers to undertake amphibian survey work
- Gain a better understanding of GCN distribution in the county

Figure 8. Reptile recording training course are offered by KRAG, this one was held at Bedegbury Pinetum.

- Identify the most important amphibian sites, and
- To test the existing GCN habitat suitability model

Getting toads out of a hole project

Getting Toads Out of a Hole collects important data on the common toad (*Bufo bufo*), a newly designated biodiversity action plan priority species. This reflects the concern shared by many that the common toad is under threat. Habitat loss, both ponds and terrestrial habitat, as well as deaths on roads during migration are both thought to contribute to the decline of the species.

In recent years relatively little attention has been given to common toads, unlike GCN, and it is also felt that common frogs are also under-recorded. KRAG's objectives for the project include to:

- Identify more toad breeding sites
- Gain a better understanding of toad distribution
- Develop a habitat suitability model for toads, and
- Develop toad crossing in the county as a family-friendly activity

For some years toad crossings had faded from view, not least because it was felt that mortalities on roads were insignificant compared with the effects of habitat loss and predation. However, as toad populations appear to have continued to decline the relative importance of road kill has increased. To counter this problem KRAG established toad crossings (Fig. 9) in at

least seven locations across the county in the spring of 2013 with financial support for ARG UK and Kent County Council. The campaign was spearheaded by radio appeals, newspaper articles, Facebook and Twitter callouts so that in a short space of time 90 people had come forward to help, of which 60 attended training workshops. The volunteers saved several thousand toads and there are plans to increase the number of manned cross next spring.

Adders in Decline project

KRAG believes that although the common viper or adder (*Vipera berus*) is under-recorded, population declines are real. In order to address these concerns, KRAG launched the 'Adders in Decline' initiative in 2004. Several factors are believed to be responsible for apparent declines in this species, including habitat loss, fragmentation of remaining populations, unsympathetic management, direct persecution and public pressure.

The project aims to promote pro-active conservation of adder by:

• Recording the distribution of adder in Kent - in particular, through identifying Key Sites and important habitat components within each site (e.g. hibernacula).

• Monitoring important populations, including long-term studies to give indications of the impact of habitat management, and

• Raising awareness and publicizing apparent declines - by running reptile survey training events, publishing habitat management leaflets etc..

In 2009, KRAG became a partner in a wildlife conservation project funded by the European Commission (Interreg-LNA), using also matching funds solicited from the Esmée Fairbairn Foundation. This project promoted the exchange of experiences in adder conservation between Kent and northern France and resulted in the transfer of skills in monitoring and database management to the Conservatoire des Sites Naturels du Nord et Pas-de-Calais (Fig. 10). The final outputs were the joint development of a leaflet on land management for adders and a joint adder conservation conference at the SE Regional ARG meeting in November 2011. The one hundred plus participants at the meeting passed a motion unanimously stating that "the adder is in more urgent need of new conservation efforts than any other reptile or amphibian species in Britain" and this led to a new wave of interest in the press. The status of the species is still sufficiently uncertain in France that no specific statement could be made although the paucity of records suggests that the situation may not be much different from southern Britain.

The success of this joint venture has led to KRAG's involvement in a further project (Interreg-Liparis) that will commence in December 2013 together with the previous French partners. The project has two goals, the first to develop a generalised habitat assessment form that can be used to collect data of relevance to adder management. The second to raise public awareness and sympathy for this species by reworking the Amphibian and Reptile Conservation Trust's adder public engagement leaflet to create versions that are relevant for public engagement in Kent and Pas de Calais.

Dragon Garden project

KRAG has joined forces with Kent Wildlife Trust (KWT) to promote wildlife friendly gardening. As part of KWT's wildlife gardening award scheme KRAG introduced a "Dragons in Your Garden" award in 2010 for the most reptile and amphibian friendly garden in Kent. Entrants to the Dragons award are selected from among those that enter the wider gardening award scheme. Members of the KRAG committee make follow-up visits to score gardens according to a set of agreed criteria. The winners receive a plaque and some literature on herps. The winning gardens have been an inspiration shared with others through the KRAG newsletter and have highlight the fact that sympathetic gardening attracts species of interest even when there is limited time, money and space.

Conservation v. Development

There is a brisk demand from the public for advice on planning applications that appear to affect reptiles and amphibians. This is dealt with by the honorary Development Officer

Figure 9. Happy group of toad patrollers in spring 2013, collecting toads and carrying them across roads to reduce road kill.

Figure 10. KRAG and Interreg partners of Conservatoire des Sites Naturels et du Pas de Calais on a field trip to familiarise them with adder sites in Kent.

who, since July 2010, has investigated and/or formally responded to over 40 planning applications within Kent and on occasion provided advice to the ARGs of other counties. In general, enquiries from the public are based around the hope that the discovery of protected species will lead to the rejection of a planning application. After reading the ecologists reports, submitted with planning applications, outcomes usually fall into one of two groups. In the first there are no obvious problems with the process, i.e. the ecologists report is satisfactory. The enquirer is told that although there may be genuine conservation concerns, the ecological report appears sound and that trying to stop the planning application by commenting on protected reptile and amphibian species it not likely to be effective given that the planning guidelines are being followed correctly. The second group of enquiries is far more interesting. These are where on reading an ecologists report serious flaws come to light. It is sad that professional ecologists make frequent errors, especially in following accepted procedures and guidelines and in applying to Natural England for licences to survey for protected species. These errors can easily pass through the system as busy planning officers often lack the knowledge to make critical assessments of ecologists' reports. On occasions either Natural England (NE) and/or wildlife crime officers of the Kent Police need to be informed that some action on their part is required although both institutions may be reluctant to act due to their own internal pressures.

Readers may feel that it is a sad reflection on the planning process that an unpaid voluntary group is effectively 'policing the system', but that is the reality of the situation in Kent. This may be a national problem and other ARGs may tell a similar story. Despite this, we do see examples of good work, where reptile and amphibian populations are 'saved' and the outcome is positive, but our dearest wish is for this to be the 'norm' and for standards to rise within the both the planning process and the ecological industry.

Aspirations

KRAG shows the typical strengths and weaknesses of a voluntary group; members are dedicated and self-motivating but their availability revolves around the demands of the day job and family. It is often hard to quantify what the Committee do, but this article has shown the very broad range of activities on which KRAG is engaged. The dedication shown by Committee members, past and present, reflects the ambition and determination of those wishing to make a difference in their local area, one step at a time.

KRAG enjoys a committed following and although 200 is an encouraging size for our membership there is certainly room to recruit more. This would bring increased revenues. It would also enlarge the pool of potentially active members which is important because the Committee frequently has to prioritise among the opportunities available in Kent based on optimising limited human resources.

Some of the things that KRAG would like to achieve over the coming years include:

• Increased influence with local decision makers

• Increased availability of records to aid those looking to make informed decisions
• Development of policies and strategies for KRAG itself, including a policy on acquiring its own wildlife reserves in the future
• Creating more family friendly activities, such as Toad Crossings, and finding more and better ways to exploit these for conservation awareness
• Encouraging the creation of a new national award for conservation organisations that have achieved significant success with herpetofauna (i.e. a national scale equivalent of the Kent 'Dragon Garden' award), and
• Lending help and support to other ARGs, especially the smaller emerging ones.

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Newsletter	Jason Steel	
Alien species	Paul Lambourne	
Toad crossings	Amy Wright	

Annex 1. KRAG Honorary Committee

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Research Article

Differences in basking site selection between the sympatric snakes *Vipera berus* and *Natrix natrix*

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ABSTRACT - The contributions of habitat selection to survival are of great importance to all species but especially in terrestrial, diurnal basking ectotherms. Despite a well-documented influence on thermoregulation efficiency, factors that influence basking, secondary to temperature are often overlooked. To assess the importance of secondary factor importance, basking site selection of *Vipera berus* and *Natrix natrix* was comparatively observed. Their sympatric distribution provided the opportunity to determine whether they select similar basking sites. Significant differences in basking site distance from cover were revealed; *V. berus* demonstrated preference for close-to-cover basking sites, *N. natrix* selected more open basking areas. No significant difference in surface temperature was found at the basking sites of either species', nor was there significant correlation shown between temperature and basking site DFC. Similarly habitat structure showed no significant impact on basking site selection. Body size correlation with site selection was however found to be inconclusive. Investigation into predator diversity and pressure, as well as prey habitat use are recommended for future research as this may offer explanation for the observations that are currently unexplained.

INTRODUCTION

Behaviours surrounding habitat selection in animals are of great interest in ecology (e.g. Eskew, Willson & Winne et al., 2009) as they represent the principal factors in the evolution of behavioural characteristics (e.g. Huey et al., 2003). Influences over resource availability and predation risk (e.g. Brown, 1999) are key factors in survival particularly in the case of terrestrial, diurnal basking reptiles. An added dependence on basking for thermoregulation creates supplementary selection pressures, placing basking habitat selection at the centre of survival (see for example Stevenson, 1985; Blouin-Demers & Weatherhead, 2002).

The selection of optimal basking habitat is therefore crucial for basking reptiles such as snakes. The value of a basking site is primarily determined by its obtainable thermal energy; higher quality, warmer sites offering increased basking efficiency (Huey, 1991a; Row & Blouin-Demers, 2006; Diaz, 1997). Mosaic basking is an alternative strategy utilised by some basking reptiles (Bauwens et al., 1990) including *Vipera berus* (Palmer, 2011) as a predator avoidance strategy. Basking in such a way can impact basking efficiency however, as lower quality/more covered basking sites though safer will ultimately require longer basking sessions (see Cooper, 1997; Row & Blouin-Demers, 2006), costing hunting time (Avery, Bedford & Newcombe, 1982).

Despite a well-documented influence over basking behaviour and duration, "secondary" factors e.g. predation pressures, resource availability or body size (see Dunham, Grant & Overall, 1989; Webb & Whiting, 2005), are less frequently reported. To assess such secondary influences, basking site selection of *V. berus* and *Natrix natrix* was observed and compared. Both of these species occupy the same habitat and this enabled observations of basking site selection that gave insight into whether each species will select basking sites, and exploit their thermal environment in similar ways.

MATERIALS AND METHODS

Data were collected across Hatfield Moor, Doncaster, South Yorkshire, UK, along two separate 3 km transects; woodland border habitat "Triangle Woods", and pathway habitat "Green Mile". Observations were made in August through to September 2011, between 0800 h and 1300 h; attempts later in the day proving impractical due to an increased alertness of the animals. Similarly, preliminary investigations showed little basking activity before 0800 h, and absence of snake activity on days of heavy rainfall, most likely due to low temperatures.

Basking site distance from cover (DFC in cm) was measured by running a ten metre tape measure at ground level, from the edge of a snakes coiled body, to the nearest point of cover. This was considered the best method of measuring DFC as snakes being long and thin animals, have no obvious centre. For the purpose of this investigation, cover was regarded as a patch of vegetation, or other matter, that would provide adequate obstruction for predator evasion.

Injury during capture was considered as too great a risk for gravid female snakes, and subsequently body size was estimated to the nearest five centimetres, as well as body temperature (T_b) and basking site ground temperature (T_g) being measured using an infra-red thermometer gun (FLUKE 68 ir thermometer, Fluke Corporation, Washington, USA).

Statistical Analysis

The Mann-Whitney U test was selected to determine significance between interspecific DFC median values, data sets from both species being found to be non-normal by a Shapiro-Wilk normality test (see Results). Additionally Pearson correlation coefficients were used to identify trend significance between between body length and temperature with DFC data, with r^2 values depicting trend direction.

RESULTS

A total of 126 snakes were observed throughout this investigation; 45 *N. natrix*, 81 *V. berus*.

Interspecific Distance from Cover (DFC) Comparison: Triangle Woods

Triangle woods DFC data for *V. berus* and *N. natrix* were non-normal (Shapiro-Wilk $p < 0.001$ for both species). Basking site DFC values differed significantly between species (Mann-Whitney U-test = 142.500, $p = 0.011$); *V. berus* median DFC = 11, range = 490 cm, *N. natrix* median DFC = 50.50 cm, range = 419 cm (Fig 1.).

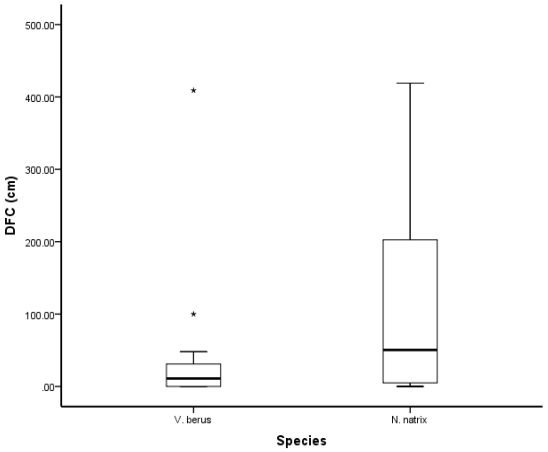

Figure 1. Differences in species median DFC values, full DFC range, and interquartile ranges of measurements taken in Triangle Woods habitat.

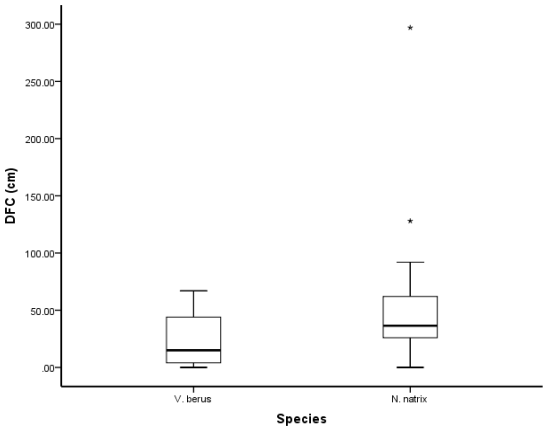

Figure 2. Differences in species median DFC values, full DFC range, and interquartile ranges of measurements taken in Green Mile habitat.

Interspecific Distance from Cover Comparison: Green Mile

Green Mile data samples were similarly non-normal (Shapiro-Wilk, $p < 0.002$ for both species). Basking site DFC values differed significantly between species (Mann-Whiteny U-test = 313.00 $p = 0.003$), *V. berus* median DFC = 15.00 cm, range 67 cm, *N. natrix* median DFC = 35. 00 cm, range = 297 (Fig 2.).

Temperature and Distance from Cover

Basking site ground temperatures (T_g), demonstrated no significant correlation for either habitat or species when plotted against DFC (Triangle Woods: *V. berus* $r^2 = 0.083$, $p = 0.182$; *N. natrix* $r^2 = 0.098$, $p = 0.179$; Green Mile: *V. berus* $r^2 = 0.009$, $p = 0.557$, *N. natrix* $r^2 = 0.022$, $p = 0.472$). No significant correlation between basking site DFC and snake body temperature (T_b) was found for either habitat or species (Triangle Wood: *V. berus* $r^2 = 0.002$, $p = 0.150$, *N. natrix* $r^2 = 0.002$, $p = 0.842$; Green Mile: *V. berus* $r^2 = 0.024$, $p = 0.378$ *N. natrix* $r^2 = 0.032$, $p = 0.194$).

Individual Body Length and DFC

Length showed a non-significant correlation with DFC in both species ($r^2 = 0.011$, $p = 0.512$) in Triangle Woods habitat. Samples from Green Mile also showed no significant trend for *N. natrix* ($r^2 = 0.007$, $p = 0.678$,) however *V. berus* demonstrated a significant trend in DFC and body length ($r^2 = 0.128$, $p = 0.026$).

DISCUSSION

The results have revealed a significant difference in basking site DFC selection between these snakes. Although neither species was observed to mosaic bask, *V. berus* has demonstrated a strong preference for close-to-cover basking sites. *N. natrix* has alternatively been shown to exploit more open basking habitats, particularly in the Triangle Woods area. Absence of correlation between DFC and temperature (T_b or T_g) suggests no thermoregulatory influence to site DFC selection. Similarly, absence of competitive displacement (Reitz & Trumble 2002) between *N. natrix* and *V. berus* described by Luiselli (2006) suggests this also is an unlikely cause for the difference in species basking site selections. Data collected regarding body length and DFC selection remains inconclusive, showing conflicting results between both sites and within species groups.

The foraging lifestyle of *N. natrix* (Meister et al., 2010) may result in less detailed habitat familiarity than in the more sedentary *V. berus* (see Chelazzi & Calzolai, 1986). Predator impact on basking site selection has not been assessed here but may be involved. For instance, it is perhaps surprising that *V. berus* is the species seen to opt for safer basking areas, given that they are aposematic and venomous (see Wüster et al., 2004) but increased mortalities from basking in more exposed locations have been found in other vipers, for instance *V. aspis* (e.g. Bonnet & Naulleau, 1996; Naulleau et al., 1997; Meek, 2013). The latter perhaps suggests that the survivorship costs associated with higher DFC basking in vipers (see Carrascal et al., 1992; Burger, 1998; Cooper, 2003; Reading & Jofre, 2009) are lower for *N. natrix*, for example, they may be less easily detected by predators, or are outweighed by key benefits from basking in more exposed locations. The observations here have highlighted a potential gap in knowledge of basking site selection of *V. berus* and *N. natrix*, as well as the importance of secondary factors in basking site selection in terrestrial reptiles. Further research is now needed to more accurately identify the key factors involved.

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Research Article

Frog Communities in Fire-Disturbed Forests of the Peruvian Amazon

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ABSTRACT - Amphibian declines are a pressing global concern. The lowland rainforests of the south-eastern Amazon harbour exceptionally high amphibian diversity, but also face a range of threats including habitat modification caused by forest fires. In this study, we sampled amphibians in areas of forest in Madre de Dios, Peru, that were affected by anthropogenic fires following severe drought in 2005. Two forest types, bamboo and terra-firme, were assessed. Forty-two anuran species were recorded in 22 survey nights. Amphibian diversity and abundance were not significantly different in burned areas of either forest type, and amphibian community composition did not change significantly between burned and unburned forests within any forest type, while bamboo forest was found to support a distinctly different amphibian assemblage to terra-firme forest. Our results suggest that further sampling over wider spatial and temporal scales to encompass a greater range of fire impacts could consolidate insights into the effects of fire on anuran communities in this region, and help to highlight the conservation value of these disturbed forests. These preliminary results are novel and enhance our understanding of how tropical forest fires may affect amphibian communities. The data also highlight the conservation value of forests affected by a fire event, as they harboured large numbers of anuran species known in the region. This is of particular interest for those species that have so far only been recorded outside of protected areas in Madre de Dios, such as *Ranitomeya cf. ventrimaculata* and *Osteocephalus buckleyi.*

INTRODUCTION

Amphibian declines are a pressing global problem, with neotropical species declining particularly rapidly (Stuart et al., 2004). Habitat disturbance, pollution and disease are considered major drivers of these declines (Beebee & Griffiths, 2005; IUCN, 2005; Wake & Vredenburg, 2008), with habitat loss and degradation thought to be affecting almost 4000 amphibian species worldwide (IUCN et al., 2008). Therefore, improving understanding of how human perturbation of tropical forests affects amphibian communities has been identified as a priority by the IUCN Amphibian Conservation Action Plan (IUCN, 2005).

As an area of extremely high biological diversity, the south-western Amazon was listed as one of the original 25 global 'biodiversity hotspots' (Myers et al., 2000). In the Peruvian department of Madre de Dios that lies at the south-western edge of the Amazon Basin, this diversity includes an exceptionally high number of amphibian and reptile species. While the department represents only 1% of the Amazon

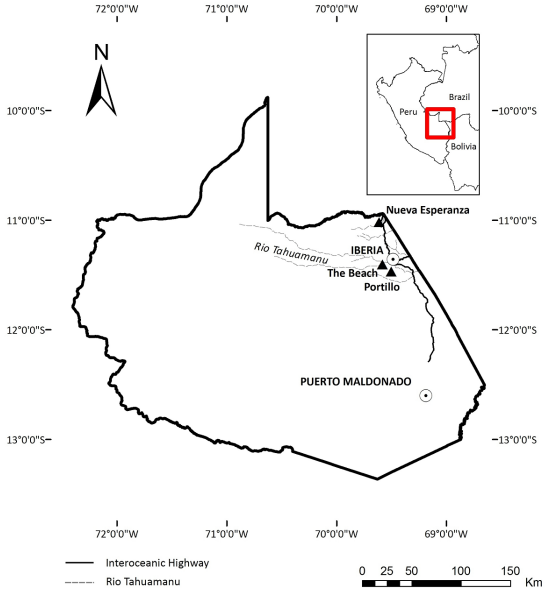

Figure 1. Study area in context of tri-national border (inset) and study sites in relation to Inter-Oceanic Highway (shown on map as completed in 2009).

Basin land area, it supports an estimated 30% of the known Amazonian herpetofauna, including 114 amphibian species (Duellman, 2005; von May et al., 2008).

The forests of Madre de Dios, which covered 94% of its area in 2011, are under threat of loss through clearance for settlements and agriculture (Oliveira et al., 2007) and artisanal gold mining, during which large areas of pristine primary forest are cleared and mercury is released into waterways and sediments (Swenson et al., 2011). Forest degradation also affects large areas, for example through selective logging and fire. Habitat disturbance by fire is thought to have a particularly strong effect on tropical forest biodiversity (Barlow & Silveira, 2009). It is currently emerging as an increasingly prevalent threat to moist tropical lowland rainforests (Cochrane, 2003; Barlow & Peres, 2008).

In closed-canopy rainforest, fire results in a dramatic alteration of forest structure within 1-3 years with widespread mortality of large trees, upper canopy thinning and increases in bamboo stems and pioneer tree species (Barlow & Peres, 2008). These changes are expected to affect amphibian communities via alterations to habitat structure, microclimate and vegetation species composition (Crump, 1994; Urbina-Cardona et al., 2006).

In 2005 a severe drought affected the whole

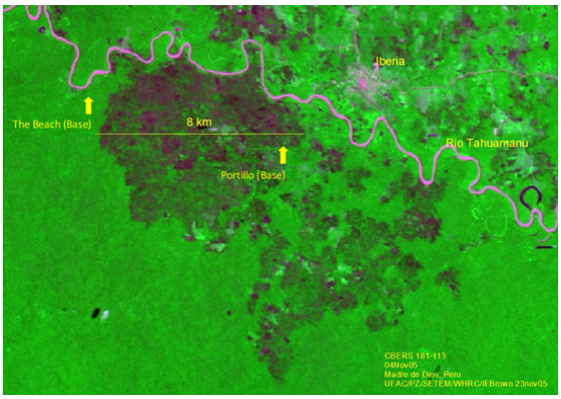

Figure 2. China-Brazil Earth Resources Satellite Program (CBERS) image of burned forest, showing dark purple against green primary forest in colour reproduction. The Rio Tahuamanu and urban areas show pink.

of Amazonia (Marengo et al., 2008; Zeng et al., 2008). In Madre de Dios, this was characterised by a virtual absence of rainfall for several months (Brown et al., 2006). This coincided with the building of a new highway in the south-western Amazon, where the increase in human activity (Perz et al., 2011; Southworth et al., 2011) led to accidental transitions of fires from settled areas into the unusually dry adjacent primary rainforests (Brown et al., 2006). This stimulated unprecedented forest burning across the region, affecting more than 467,000 ha of forest and open areas in Acre (Vasconcelos & Brown, 2007) and at least 10,000 ha of forest in Madre de Dios (Brown et al., 2006).

Relatively few studies have previously assessed the consequences of fire for amphibian communities in moist lowland tropical forests. Fredericksen & Fredericksen (2002) studied a tropical forest landscape in Bolivia disturbed by both fire and logging, and chiefly focussed on the impact of logging gaps, finding only four amphibian species. Other studies in South America directly assessing the impact of fire have focussed on open areas surrounded by forest (Papp & Papp, 2000), restinga forests along the Atlantic Coast (Rocha & Ariani, 2008) or savannah forests where fire is a natural part of the ecosystem (Cano & Leynaud, 2009).

The aim of this study was therefore to make a preliminary assessment of anuran species richness, abundance and community composition in forests affected by fire during the 2005 drought, and to compare these data to

Forest type	Study site name	Burn status	Number of transects
Bamboo	The Beach	Burned	5
	The Beach	Unburned	5
Terra-firme	Nueva Esperanza	Burned	2
	Nueva Esperanza	Unburned	4
	Portillo	Burned	4
	Portillo	Unburned	2

Table 1. Transects surveyed in each forest type, showing burn status and location of transects. Two localities were sampled in terra-firme forest, the results of which were pooled in the analysis (Nueva Esperanza and Portillo). One locality was sampled in bamboo forest (The Beach).

nearby non-impacted forests.

METHODS

Site description

The study sites lie close to the Inter-Oceanic Highway in Madre de Dios, south-eastern Peru (Figure 1), paving of which began in 2006 (Southworth et al., 2011). Significant migration of people to the area in advance of the road paving led to some of the greatest extent of fire damage occurring here in 2005, associated with settlement activity (Perz et al., 2011; Southworth et al., 2011).

Four major forest types found across the south-western Amazon were present: terra-firme forest, floodplain forest, bamboo forest and palm swamp (Pitman et al., 1999). These forest types are strong predictors of amphibian assemblages, with a high proportion of the regional amphibian diversity attributable to a high beta-diversity across the forest types (von May et al., 2010). The overall amphibian species pool of Madre de Dios is well-documented through a number of long-term studies, and species lists are available for a number of localities in nearby protected areas (Doan & Arriaga, 2002; Duellman, 2005; von May et al., 2010).

Meteorological records from the Los Amigos Biological Station (CICRA) approximately 140 km from the study site indicate that annual precipitation ranges between 2700 and 3000 mm, with a mean annual temperature between 21˚C and 26˚C (von May et al., 2010). A distinct period of reduced rainfall with slightly cooler temperatures occurs between June and September. This dry season is also characterised by periodic cold weather events driven by southerly winds from Patagonia known as friajes. During these events, windspeeds are elevated and air temperatures can drop as low as 5°C at night.

Study sites

Images captured by the China-Brazil Earth Resources Satellites (CBERS) in 2005 were used to identify areas of burned forest (Figure 2) and to select three study sites located outside protected areas. One study site was known to have been well established bamboo forest before the fires, with the other two study sites located within closed-canopy terra-firme forest.

Terra-firme forests are characterised by high (>30 m) closed canopies with many emergent trees and high tree species diversity. In contrast, bamboo forests are dominated by *Guadua sarcocarpa* and *G. weberbaueri*, rising to ~25 m, with reduced tree species diversity and fewer high-canopy trees (Griscom & Ashton, 2006; Griscom et al., 2007). These forests form where disturbance events in the past opened the canopy, allowing invasion of bamboo and formation of a self-perpetuating cycle of tree damage and bamboo regrowth (Griscom & Ashton, 2003; Griscom & Ashton, 2006).

Sites were chosen near the boundary between large continuous burned areas and unburned areas in order to allow pairing of sampling sites with minimal micro-geographic variation. The first terra-firme site 'Nueva Esperanza' (11° 1'13.07"S, 69°36'38.72"W), was located approximately 4 km from the highway. The second terra-firme site, 'Portillo' (11°28'6.60"S, 69°30'2.66"W), was located 6 km south of the

	Bamboo		Terra Firme		
Species	**Burn**	**Unburn**	**Burn**	**Unburn**	**Sampling method**
Allobates conspicuus	0	0	0	1	O
Ameerega hahneli	1	6	1	0	T
Ameerega petersi	0	1	0	0	T
Ameerega trivittata	0	0	4	2	Both
Chiasmocleis bassleri	0	0	1	1	Both
Chiasmocleis ventrimaculata	1	0	0	0	T
Teratohyla midas	0	1	0	0	O
Dendropsophus acreanus	0	0	0	1	O
Dendropsophus allenorum	1	0	0	0	T
Dendropsophus koechlini	1	2	0	0	T
Dendropsophus leali	0	2	0	0	T
Dendropsophus leucophyllatus	0	1	0	1	T
Dendropsophus parviceps	0	5	1	3	T
Dendropsophus schubarti	2	3	0	0	T
Dendropsophus xapuriensis	0	0	0	1	T
Elachistocleis bicolor	6	1	1	0	Both
Eleutherodactylus sp.	1	0	0	0	T
Engystomops petersi	0	0	0	1	O
Hamptophryne boliviana	0	9	0	0	T
Hypsiboas fasciatus	31	37	3	1	Both
Hypsiboas lanciformis	0	0	1	0	O
Hypsiboas sp.	0	1	0	0	T
Leptodactylus andreae	4	0	0	10	Both
Leptodactylus cf. petersii	0	1	0	0	T
Leptodactylus lineatus	0	0	2	0	T
Leptodactylus rhodonotus	0	1	0	0	O
Leptodactylus sp.	1	0	0	0	T
Oreobates quixensis	0	0	1	0	T
Osteocephalus cabrerai	0	0	0	1	T
Osteocephalus leprieurii	0	0	3	1	T
Osteocephalus sp.	0	0	4	2	Both
Phyllomedusa palliata	1	2	0	1	T
Pristimantis peruvianus	1	3	6	17	Both
Ranitomeya cf ventrimaculata	0	0	0	3	T
Rhinella margaritifera	0	5	3	0	T
Scarthyla cf. goinorum	1	0	0	0	O
Scinax garbei	3	2	0	0	T
Scinax ictericus	16	15	1	1	Both
Scinax pedromedinae	0	1	0	0	T
Scinax ruber	1	1	0	0	T
Trachycephalus venulosus	1	0	0	0	O

Previous page: Table 2. Anuran species list, showing number of individuals recorded in each forest type. Sampling methods: O = opportunistic, T = transect, Both = individuals found both on transects and opportunistically.

town Iberia on the Inter-Oceanic Highway (5km south of the Tahuamanu River). The bamboo-dominated site 'The Beach' (11°24'14.14"S, 69°34'50.79"W) was situated next to the Tahuamanu River 9 km east of Iberia (Figure 1).

Study plots

We set up randomly oriented 250 m transects at 100 m intervals along a main path at all sites. In the bamboo forest site, five transects were cut in burned areas and five in unburned areas. At the two terra-firme forest sites, two transects were cut in burned areas and four in unburned at Nueva Esperanza, and four in burned and two in unburned at Portillo. Data from these two terra-firme sites were combined in the analysis to give six transects in burned forest and six transects in unburned forest (Table 1).

Transects were cut just wide enough to allow passage through the undergrowth and were left undisturbed for two days after cutting to allow any disturbed amphibians to return to the area.

Sampling

Each transect was surveyed once using a Visual Encounter Survey (VES) between 18:00 h and 00:00 h according to the protocol described by Doan (2003). Sampling was completed over 22 nights in July and August 2009. Night time VES were chosen because comparative studies have found this method to be best for detecting inter-site variation in amphibian species richness (Pearman et al., 1995), encompassing unique species (Doan, 2003) and finding the greatest amphibian abundance and species richness in short-term studies (Rödel & Ernst, 2004; Donnelly et al., 2005). Two to four surveyors completed each transect, all with comparable surveying experience. The length of time spent searching each transect was recorded, allowing expression of abundance as a product of search effort. It cannot be assumed that the abundances recorded represent actual abundance and abundance is therefore defined as 'estimated activity abundance' (Pearman et al., 1995). Amphibians that were encountered opportunistically, for example on the way to

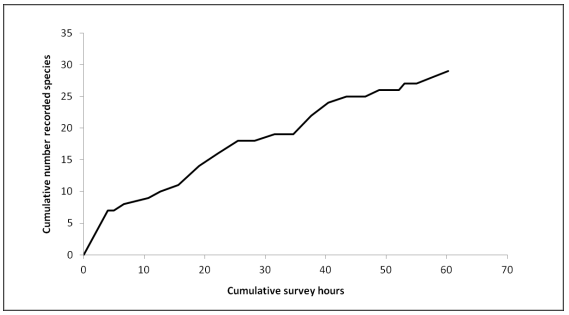

Figure 3. Species accumulation curve, showing the number of cumulative species recorded with cumulative survey hours. The curve shows no sign of stabilisation.

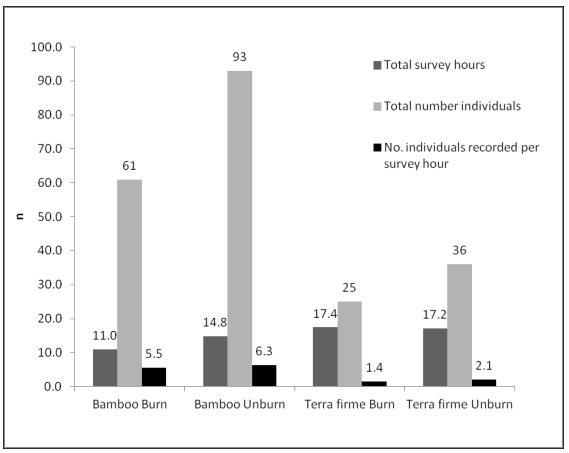

Figure 4. Anuran abundance. Three bars represent: number of survey hours in each forest type, number of individuals encountered in each forest type (sum of all surveys), and the number of individuals recorded by survey hour.

transects, were also identified to add to the overall species list, but they were not included in any statistical analysis.

Individuals were either identified immediately or retained until the following morning, when they were photographed and identified using a number of published field guides (Bartlett & Bartlett, 2003; Knell et al., 2004; Duellman, 2005; von May et al., 2007; Barbosa de Souza, 2009) before being released. No voucher specimens were taken.

Tree density was recorded along the same 250 m transects during daylight hours after the completion of amphibian surveys. All living and standing dead trees >10 cm DBH were recorded within 5 m on either side of transects. Sapling stem density and bamboo data were

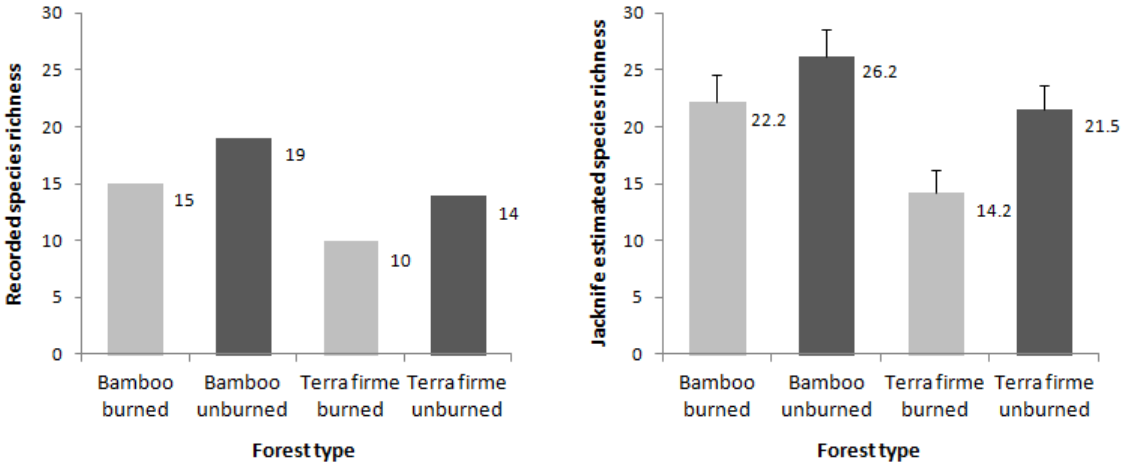

Figure 5. Anuran species richness. Recorded species richness is shown in the left panel, and Jacknife estimated species richness in the right panel, for each forest type. Error bar on Jacknife estimated species richness represents the calculated standard deviation.

collected within two 50 m x 1 m plots at the start and end of each transect. Within each plot, all tree saplings with <10 cm DBH (or diameter <10 cm immediately below branching if between 1 m and 1.3 m high) and height ≥1 m were recorded, with bamboo stems within this size class recorded separately.

Data analysis

Comparisons of species richness between sites do not incorporate key information about species abundances or sample completeness patterns. Therefore, species diversity indices taking into account both species richness and relative abundances or estimators of true species richness are useful. Due to the low frequency of many species in the data, the use of diversity indices was difficult, but the data did allow meaningful calculations of non-parametric species richness estimators. These indicators approximate the total species richness of the site, had sampling continued until no additional species were found, based on the number of singletons and doubletons in the sample. The Jackknife estimator (Jack 1) was selected and calculated using EstimateS 8.2 (Colwell, 2009). Species accumulation curves were also created to assess sample completeness.

RESULTS

Over the 22 sampling nights, 254 anuran specimens comprising 33 species were sampled on the transects. A further 39 individuals comprising 17 species were caught opportunistically, including nine additional species that were not recorded during VES on transects (Table 2). Fourteen species were only found in unburned forests, while nine were only recorded in burned forest. A species accumulation curve for all anurans collected on transects does not show any levelling off, indicating that sampling was incomplete (Figure 3).

Significant reductions in large tree density were recorded in burned areas of terra-firme forest (Mann Whitney tests: Portillo w = 10, p = 0.01; Nueva Esperanza w = 6, p = 0.05), while there was no significant difference in bamboo forest (Mann Whitney test w = 12.5, p = 0.07). The density of standing dead trees was higher in burned areas at all sites.

The total number of recorded anuran individuals (burned and unburned data) was higher in bamboo forest than in terra-firme forest (Mann-Whitney w = 79, p = <0.001) (Figure 4). Within both forest types, more individuals were recorded in unburned areas than burned areas (Figure 4), but these differences were not statistically significant (bamboo: Mann-Whitney w = 27, p = 0.92; terra-firme: Mann-Whitney w = 32.5, p = 0.31).

Recorded anuran species richness was higher in unburned areas of both forest types (Figure 5), but again, differences were non-significant

Figure 6. Anuran families recorded during sampling with representative species: a) Bufonidae – the true toads b) Dendrobatidae – poison dart frogs c) Strabomantidae – diverse group, all species thought to be direct-developing d) Hylidae – tree frogs and allies e) Leptodactylidae and f) Microhylidae – large and diverse group containing many different guilds.

(bamboo Mann-Whitney w = 25, p = 0.25; terra-firme Mann-Whitney w = 29, p = 0.40). No correlation could be found between recorded or estimated anuran species richness and any of the measured vegetation variables. In relation to anuran family spectra, the vast majority of individuals sampled in bamboo forest were from the Hylidae (75%), while in terra-firme forest, Hylidae comprised 38%, while members of other families like Strabomantidae (22%), Leptodactylidae (17%), Dendrobatidae (16%) and Bufonidae (5%) were also strongly present

(Table 2). These families contain species with strongly varying life history characteristics (Figure 6).

Pristimantis peruvianus (Strabomantidae) was found in higher numbers in terra-firme forest (n = 15) compared to bamboo forest (n = 4), while the two Hylidae species *Hypsiboas fasciatus* (n = 61 in bamboo forest, n = 4 in terra-firme) and *Scinax ictericus* (n = 26 in bamboo forest, n = 1 in terra-firme) were much more abundant in the bamboo forest. These two species represented a substantial proportion of the total specimens in the bamboo forest and contributed considerably to the difference in species composition between the forest types (Table 2).

DISCUSSION

While burned areas of both bamboo and terra-firme forest appeared to support lower anuran species richness and abundance, these differences proved not statistically significant. This may be due to the short sampling period, as species accumulation curves and Jacknife estimations of the true species richness suggested that sampling was incomplete in both forest types. Long term datasets in the region have yielded up to 67 anuran species at a single site (Duellman, 2005), while Doan and Arriaga (2002) recorded 76 anuran species across five sites with 23 months. In context, our species list of 38 named species and 42 species overall recorded in such a short sampling period nonetheless highlights the exceptional anuran diversity at our study sites even after disturbance events such as the 2005 fires.

All of the recorded species are known to be tolerant to some degree of habitat modification, and even in unburned areas, no primary forest specialist species were recorded. This is a likely indication that our sampling sites lay within transition zones or edge areas, rather than within core areas of burned and unburned forest sites. Additionally, some highly generalistic species were recorded, like *Elachistocleis bicolor*, a species exclusively found in bamboo forests and more often in burned areas. This species has been recorded in savannah habitats subjected to various burning and grazing regimens in northeast Argentina (Cano & Leynaud, 2009) and survives in substantially disturbed and modified habitats. It may therefore act as an indicator for habitat modification. Only one direct developing species (*Pristimantis peruvianus*) was recorded, occurring in notably higher numbers in terra-firme forest. All other recorded species have aquatic larval stages.

Overall, our data suggest that to fully assess the impact of forest fire on amphibian communities, particularly on primary forest specialists, more extensive sampling over a wider spatial scale between unburned and burned areas stretching over a large number of survey nights is required.

The finding that bamboo forest supported a different assemblage of anuran species with a higher species richness and abundance than terra-firme forest is in contrast to the findings of von May et al. (2010), who found no difference in community composition or species diversity between these forest types. This may be due to differing water availability between the two forest types during this study. While all sites were criss-crossed by small streams and close to large rivers, large patches of *Heliconia* sp. at the bamboo site suggested particularly high levels of soil moisture (Griscom et al., 2007). Increases in diversity and abundance of amphibians are expected close to water bodies due to enhanced numbers of species that rely on them for breeding (Rojas-Ahumada & Menin, 2010), but general water availability becomes particularly important for some anurans in the dry season (Suazo-Ortuño et al., 2008; Urbina-Cardona et al., 2006) when the present study took place.

Supporting this was the dominance of Hylidae at the bamboo forest site. Members of this predominantly arboreal family often only breed in temporary ponds (Bartlett & Bartlett, 2003), making them particularly reliant on areas with higher water availability.

While the short sampling period and the incompleteness of sampling means that these results must be considered as preliminary, the findings are still important for our understanding of how tropical forest fires may affect amphibian communities. The results support previous work (von May et al., 2010) in highlighting the importance of identifying distinct forest types when conducting amphibian studies in this region. Our data also highlight the conservation value of forests degraded by a burn event and of associated edge areas, as even in this short

study, a large proportion of anuran species known to the region were recorded. This is of particular interest for those species that have so far only been recorded outside of protected areas in Madre de Dios, such as *Ranitomeya cf ventrimaculata* and *Osteocephalus buckleyi* (von May et al., 2008). In the eastern Amazon, it has been reported that 38% of burned forests were subsequently deforested (Alencar et al., 2011) suggesting that the risk of further degradation or removal of burned forest may be high. Taking measures to prevent further degradation of the burned areas might therefore be more important for the preservation of anuran communities in this region than previously thought.

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Short Note
Male-male ritual combat in *Spilotes pullatus* (Serpentes: Colubrinae)

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INTRODUCTION

Some males snakes present morphological changes during reproduction in the breeding season and also display social interactions, such as male-male ritual combat. Ritual combat involves the interaction between two males, apparently in competition for mating dominance (Carpenter, 1977). Generally, the ritual consists of bodily contact between males. They exert pressure, either by pushing or twisting one another, with the goal being to physically dominate an opponent by forcing their head toward the ground (Carpenter, 1977).

Combat between males has been described for several snake taxa; Boidae, Elapidae, Viperidae and Colubridae (Carpenter, 1977; Shine, 1978, 1994; Almeida-Santos & Marques, 2002). Different taxa exhibit different behaviour during a ritual. During combat colubrids tend to assume a horizontal posture with trunk and tail regions entwined and some oblique or slight vertical elevation of the head (Carpenter, 1977). Combat is usually observed during the breeding season (Capula & Luiselli, 1997; Schuett, 1997; Schuett et al., 2001) and factors such as body size of the male and previous combat experience are important for victory in combat and mating success (Schuett, 1997).

The genus *Spilotes* (Wagler, 1830) (Colubridae, Colubrinae) comprises one species, *Spilotes pullatus*. It is a widely distributed snake, inhabiting areas of Central and South America from Tamaulipas in Mexico to Paraguay, and across the extreme northeast of Argentina (Savage, 2002). It is a diurnal, terrestrial and arboreal (Vanzolini et al., 1980; Sazima & Haddad, 1992; Marques, 1998; Boos, 2001; Marques & Sazima, 2004; Pontes & Rocha 2008).

METHODS

The description of ritual combat by *S. pullatus* herein was based on footage and detailed notes from four observations. The four instances detailed combat in situ in Brazil. Ritual combat #1 and #2 were observed in the southeast region, on Cardoso Island in the municipality of Cananéia, state of São Paulo (25° 7'S 47° 57'W) in early spring. Ritual combat #3 and #4 were observed in the central-west region of Brazil in late spring and early summer respectively. Combat #3 was filmed on the left bank of Sucuriú River (19º 08' 30"S 52º 58' 03"W), Paraiso Farm, municipality of Costa Rica, state of Mato Grosso do Sul and combat #4 was filmed at Vagafogo Farm (15º 50' 14.62"S 48º 58' 33.35"W), municipality of Pirenópolis, state of Goias (Table 1).

RESULTS

Description of the ritual

Males of *S. pullatus* kept their bodies partially entwined during combat (Fig. 1). The intensity at which the bodies became entwined varied. Specimens tried to keep the cranial portion of the body in the most upright position, trying to raise their heads as far as possible above the ground (Fig. 1a and 1d). The male that was disadvantaged in rituals retreated its head, loosened its body from the entwined posture,

Case	Season Date	N specimens	Combat	Copulation behaviour	Duration	Place
#1	Austral spring 12 Oct 2000	3	X	X	-	Cardoso Island-SP
#2	Austral spring Oct 2001	3	X	X	-	Cardoso Island-SP
#3	Austral spring 13 Dec 2007	2	X*	-	+ 1 hr	Costa Rica-MS
#4	Austral summer 26 Dec 2012	2	X	-	+ 1 hr	Pirenópolis-GO

Table 1. Male-male ritual combat in *S. pullatus* observed in situ. (x) observed; (-) not observed; (*) hemipenial exposition.

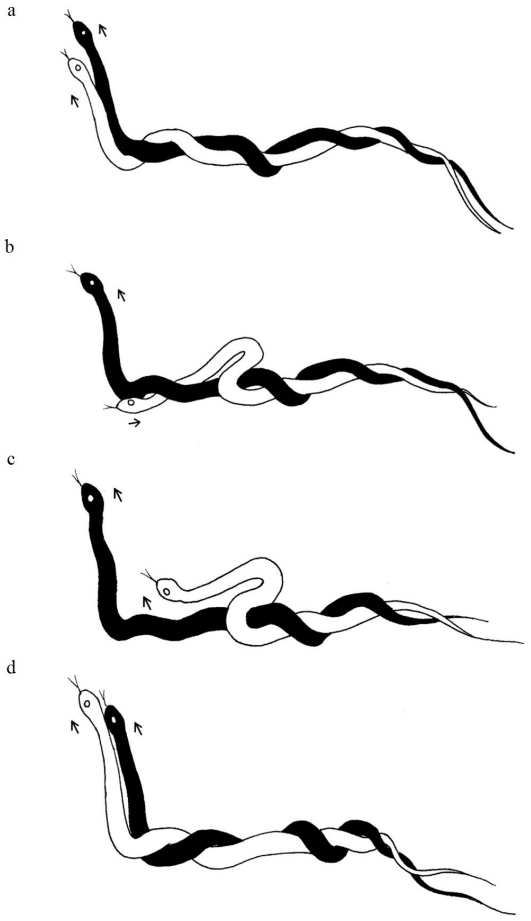

Figure 1. Male-male ritual combat in *S. pullatus*. A. The males try to keep their heads as high as possible. B. The male that is at a disadvantage (white) retreats its head and loosens a portion of the body twining. C. After gaining free space with the body, the male that retreated (white) places its head up again. D. At this time, this male (white) is able to keep its head higher than the opponent (black). The arrows indicate the direction of the movement performed by the individuals.

and placed its body back to a vertical position by raising its head above that of its opponent (Fig. 1b and 1c). Throughout each combat ritual, the snakes also used their tails to restrain their opponent by preventing forward locomotion. Bites were not observed during rituals.

Observations of male combat

Case #1: One *S. pullatus* was observed near the curb of a sidewalk while a second individual approached. The two paired their heads and a third individual approached while the other two were exhibiting courting behaviour. One of the snakes (probably a male) interrupted the courtship behaviour to pursue the third individual, who fled. The "pursuer" returned to courtship and the third snake, which had been chased away, approached the courting pair again. It was pursued a second time and finally chased away by the "pursuer". The pair remained copulated and later were observed mating on a lemon tree.

Case #2: Three individuals were observed together, two keeping their heads raised on display and later in combat. After a short period two of the snakes engaged in copulation. The third individual was no longer near the site.

Case #3: This episode occurred on the banks of a river. Males were entwined and repeatedly tried to keep the cranial part of their body raised above their opponent's. During certain moments, the tail was used in an attempt to immobilize the opponent and, on some occasions, it was possible to observe a brief exposure of the hemipenis of one individual. Combat occurred in low-lying vegetation and partly among

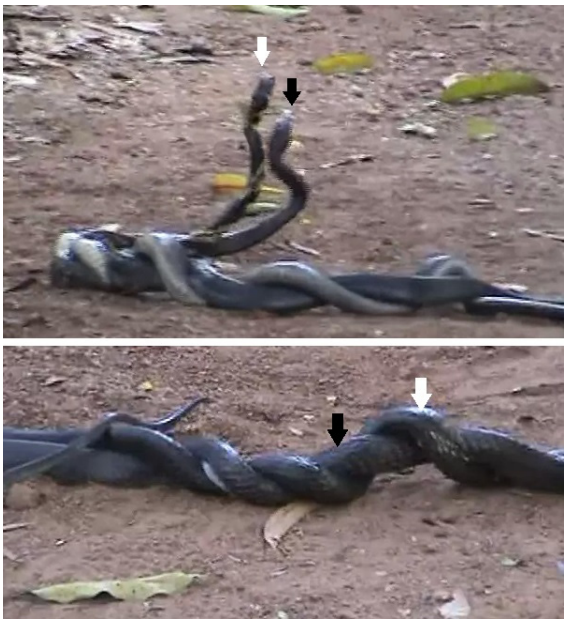

Figure 2. Two males in combat. Note that the smaller male (white arrow) tries to immobilize the larger male (black arrow) keeping his body intertwined with its opponent.

terrestrial substrate. During combat the snakes fell off branches. The beginning of the ritual was not observed and it lasted for approximately one hour.

Case #4: On a dirt road two males were sighted already commencing combat. The snakes kept their bodies entwined throughout combat, although it was noticeable that the smaller male intertwined its body more than the larger male in an attempt to immobilize its opponent (Fig. 2). After a period approximately one hour the larger male gave up and retreated to a tree nearby. The smaller male then chased the losing individual away.

DISCUSSION

Reproductive behaviour in snakes involves courtship, copulation, and in some species, mate guarding or surveillance that can lead to combat between males (Carpenter, 1977; Almeida-Santos et al., 1999). Of the approximately 2700 known species of snakes, combat has been recorded for only 6% of species (Schuett et al., 2001).

Combat ritual among colubrids was described by Carpenter (1977) and reported to be horizontal, with the snakes rarely raising the head and neck regions off the ground. The only previous exceptions were species such as *Elaphe longissima* and *Ptyas mucosa* that raise the cranial part of the body to try to force the opponent's head toward the ground. Almeida-Santos & Marques (2002) reported that *Chironius* did not fit this general pattern for colubrids since the species displayed a combat ritual where males tended to maintain the cranial portion of the body vertical. This posture of lifting the cranial portion of the body is more typical of Viperines. *S. pullatus* also fits the pattern demonstrated by *Chironius* and both genera are semi-arboreal. This suggests that semi-arboreal colubrids may engage in combat ritual in a similar manner to viperines and not adopt a standard horizontal position as other colubrids do. Although there is a similarity in the position of the combat ritual of *Spilotes* with that of vipers, it appears different because male vipers often try to push the opponent's head against the ground (Carpenter, 1977). This suggestes that a possible goal of the behaviour is to restrain, through physical force, the head of the rival. For *S. pullatus* the goal of the combat ritual seems to be to keep the head higher than that of the opponent, possibly to boast that it is the larger individual. Schuett (1997) demonstrated that male size is important for success in combat and influences mate selection by females. The fourth ritual (case #4) showed clearly that larger males are not always more dominant than smaller ones and it is possible that a male with more energy may have the advantage during combat. Combatant snakes that did not become fatigued in our observations were able to remain upright and win over weaker rivals.

Combat in *S. pullatus* lasted approximately one hour. Despite the long period of the ritual, bites or any application of physical damage to an opponent was not observed, and therefore the ritual may only result in an energy expenditure cost for the participating snakes. In one case (case #1), the males were not observed commencing a duel but the encounter did not result in combat. Instead, the dominant male chased the subordinate male away suggesting that the cost of energy expenditure, or perhaps even previous combat experience, may serve as a deciding factor for males during combat rituals. In two of the reported cases (case #1 and #2), three animals were present, two of which participated in the ritual and the third (female) observed to copulate with the likely winner of

the combat. It is possible that proximity of this nearby female may have incited combat ritual. Combat behaviour in snakes typically occurs during the mating season and is mostly related to copulation (Capula & Luiselli, 1997; Schuett et al., 2001; Almeida-Santos & Marques, 2002).

In both interactions of *Spilotes* in the southeast region on Cardoso Island, Sao Paulo, combat occurred during early spring, which is when the final period of copulation for *Spilotes* species in the southern hemisphere occurs (Muniz-Da-Silva, 2012). However, combats between male *Spilotes*, observed in the central-west region, occurred in late spring and early summer, which does not correspond to the mating season of the species. During these events, there were no females present. This may suggests that male-male combat rituals for *S. pullatus* may serve other purposes besides access to females such as dominance or territory dispute. Dominance and territory has also been suggested as a cause of combat for *Chironius bicarinatus* (Marques et al., 2009)

In one instance (case #3) exposure of the hemipenis of one of the snakes was observed during combat. Almeida-Santos et al. (1998) also observed this behaviour in *Micrurus frontalis*. Behavioural analysis of this phenomenon is not easily explained and would require further observations to decipher exact reason for its function.

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Natural History Notes

CALOTES VERSICOLOR (Asian garden lizard): ALBINISM. Albinism has been observed in all vertebrate groups. It occurs through a reduction or absence of melanin pigment, due to genetic mutations, diet, age, disease or injury (Sage, 1962; Owen & Shimmings, 1992; Summers, 2009). Albinism has been reported in lizards of the Anguidae, Sphaerodactylidae, Varanidae and Lacertidae (Robert et al., 1965; Delaugerre, 1981; Eidenmullser, 2003; Spadola & DiToro, 2007) but there have been no previous reports on albinism in Agamid lizards.

This note reports albinism in a laboratory-born hatchling of the Asian garden lizard *Calotes versicolor* (Agamidae) in Dharwad, Karnataka, India. The lizard's body, skin and eyes appeared pink. The neonate hatched on 29th August 2012 from laboratory-laid eggs (n = 13) of a female collected from the surrounding areas of Dharwad city (15° 17' N and 75° 3' E) during the second week of June 2012. Figure 1A & B show the albino hatchling alongside a normally-coloured sibling (Fig. 1C). The snout-vent length of the newly-hatched lizard was 24.53 mm; tail length 50.87 mm; weight 469 mg. The sex was recorded as male by extrusion of the hemipenes as described by Harlow (1996). The hatchling survived for only 12 days and died of unknown causes.

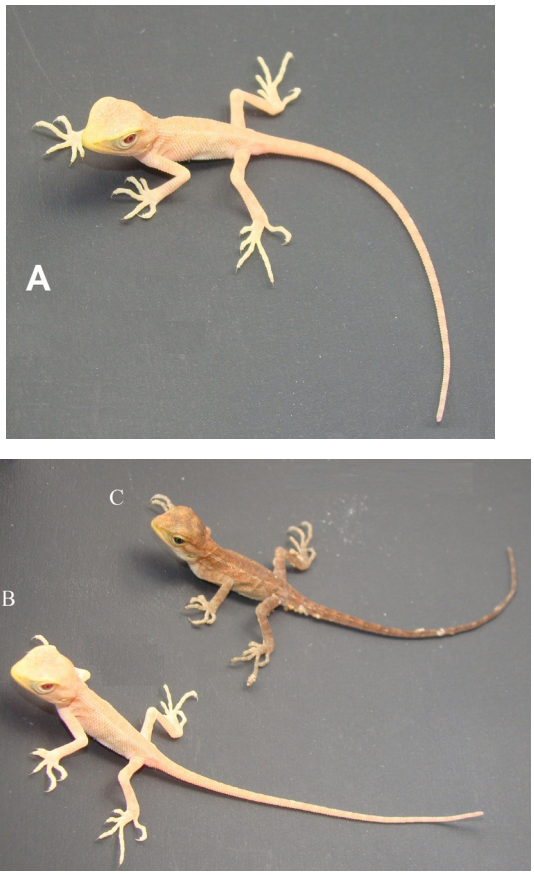

Figure 1. Albinism in hatchling of Asian garden lizard *C. versicolor*: (A) albino hatchling; (B) albino hatchling in comparison with (C) normal hatchling from the same clutch.

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DERMOPHIS GLANDULOSUS (Glandular Caecilian) DIET. As with most caecilians we know little about the biology of *Dermophis glandulosus*, other than its taxonomic status and its distribution from Costa Rica to northern Colombia (Savage & Wake 2001). This species seems to be one of the more rare caecilians for this region with just 10 specimens deposited in the collection of the Museum of Zoology at the University of Costa Rica. Herein we describe the stomach contents of a specimen found dead in the field on the 9 October 2005 near Rio Járis, Canton de Mora, Costa Rica (9.9043 N, 84.3012W; 550 m asl; in tropical humid forest).

The specimen was in good condition when found, however, cause of death could not be determined. Upon examination of its stomach contents we discovered a well-developed egg of an *Anolis* sp. It was identified as an *Anolis* egg as the embryo had well developed lamellae on all of its fingers (Losos, 2009); nevertheless it was impossible to identify the species as the body scales were not yet fully developed. The embryo presented 14 lamellae under the forth toe, which is common for most medium sized anoles in the region (Savage, 2002). The most abundant species in this area are *A. cupreus* and *A. intermedius*. Because *Anolis* eggs are commonly laid above ground or just under a rock or log (Losos, 2009), this record suggests this species could be actively foraging on the forest floor and not just underground as it is thought of for most caecilians.

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CORONELLA AUSTRIACA (smooth snake): PHOTOGRAPHIC RECORD OF AUTUMN COPULATION. The smooth snake, *Coronella austriaca*, is one of three species of snake native to the UK. Its range is restricted to lowland heathland sites in southern England, and it is both secretive and rare (Reading, 2004a). In the UK, the smooth snake is often associated with mature dry heathland, though it will also take advantage of adjacent habitats including wet heath, scrub and grassland (Gent, 1988; Beebee & Griffiths, 2000). It is believed that, after emerging from hibernation in April, mating will generally take place in May (Spellerberg & Phelps, 1977; Frazer, 1983; Reading, 2012). The adult females subsequently give birth to live young between late August and October (Smith, 1951; Goddard & Spellerberg, 1980; Frazer, 1983; Braithwaite et al., 1989).

The smooth snake's mating, copulation, and parturition behaviour has, however, been witnessed so infrequently that the species' reproductive ecology is still not fully understood. Recorded copulations in the wild in the UK are rare. The information gained from those that have been recorded may indicate a more protracted and complex mating behaviour than previously thought. For example, Spellerberg & Phelps (1977) observed copulations across an extended mating period from May to early June, Braithwaite et al. (1989) record a mating in August, and Bull (2010) records a mating in September. Some females have been found to be gravid during the spring, suggesting that mating could have taken place late in the previous year (Spellerberg & Phelps, 1977; Braithwaite et al., 1989). This has also been witnessed by Atkins (2011).

On 27 September 2013 at 15:40, copulation between two adult smooth snakes was observed and photographed (see Figs. 1 and 2).

The snakes were found during fieldwork trialling a new proposed 'Standardised Reptile Survey Methodology' (Limburn & Woodley, 2014), which involves a photographic mark-recapture study of smooth snakes. The survey site is a heathland reserve unit within the Morden Bog and Hyde Heath SSSI near Wareham, Dorset, which is managed by Amphibian and Reptile Conservation (ARC). Fieldwork was carried out under Natural England licence to ARC.

Figure 1. Copulating smooth snakes. Photograph by Stuart Handyside.

Figure 2. Close-up view of copulating smooth snakes. Photograph by Stuart Handyside.

The snakes were found under a corrugated metal refuge on a north-east facing slope. The sloping habitat consisted of dry heath leading to low lying wet heath; the dry heath primarily consisting of ling (*Calluna vulgaris*), bell heather (*Erica cinerea*) and dwarf gorse (*Ulex minor*). The vegetation under the refuge consisted of compressed dead *C. vulgaris*. Both snakes were calm when handled and remained joined at the cloaca allowing photographs and the recording of identification details. The female snake had a total length of 492 mm and snout-vent length (SVL) of 413 mm. The male

snake had a slightly longer total length (520 mm) with a SVL of 405 mm. Sexual dimorphism in the species is indicated by longer tail length in the males, due to allometric growth. Measurements of SVL applied to calculations in Reading (2004a) suggest the age of both snakes to be between 6 and 7 years.

Weather conditions at the time of the observation were dry, with a light wind and cloud cover of 67%. Environmental data were recorded using a digital thermometer and hygrometer. The air temperature was 21ºC, with relative humidity of 65%. These were average conditions for September, based on environmental data from the nearest weather station, covering the period 2000-2013 inclusive. The mean spring temperature for 2013 was 0.9 ºC cooler than average, whilst the mean summer temperature was 0.5 ºC warmer.

Three neonate smooth snakes were also found under the same refuge as the copulating adults. Although it is unknown if the female found in copulation had given birth to these juveniles, we consider it possible. Our observation may therefore suggest that mating in smooth snakes can occur shortly, or immediately after, the female has given birth. Pernetta (2009) suggests that high levels of rainfall and lower average temperatures can reduce basking opportunities for gravid smooth snakes, resulting in a longer gestation period. If our observed smooth snake gave birth late in the season due to the unfavourable weather conditions recorded for spring 2013, this could have been a reason for the female's willingness to mate so late in the year. Reading (2004b) describes the smooth snake as being 'prudent opportunistic breeders' compared to the observations of Strugariu (2007), who suggests the species could be a 'capital breeder' utilizing long-term sperm storage.

We aim to further investigate these possibilities by relocating and monitoring the female during spring 2014, to determine if the copulation has resulted in successful fertilization and to deduce the probable length of her gestation. The individual markings (head patterns) of each snake were photographed during the observation, enabling re-identification during future surveys of the site. Further investigation could also be carried out to compare temperature and other environmental data with the dates of known copulations and births.

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PHYSALAEMUS CENTRALIS (Central Weeping Frog): VOCALISATIONS AND CONSERVATION. Playback of vocalisations is a commonly used technique in field ornithology (Boscolo et al., 2006), with multiple applications in ethology (Mc Gregor & Falls, 1984), species detection, resolution of taxonomic issues (Whittaker, 2002), censusing (Ratcliffe et al., 1998) and assessments of intra- and interspecific interactions (e.g. Stouffer, 1997). However, despite the vocal nature and territoriality of many Anurans, use of the technique in herpetological studies has been limited and frequently restricted to more specialised studies of vocalizations and their associated behaviours (e.g. Wells, 1988). Whereas playback is habitually used even by non-professional birdwatchers as a means of visualising secretive or hard to see species, few herpetologists commonly employ the technique, preferring instead more invasive and frequently frustrating active search methods. Many anurans do react strongly to playback, and in some cases the technique may even have advantages over active searching such as assisting in the location of small species, those that call from burrows or concealed locations and even in tracing the source of unknown calls.

At Reserva Natural Laguna Blanca (RNLB), Departamento San Pedro, Paraguay on 15 December 2009 the author heard a loud chorus of frog calls from dense grassy vegetation at the edge of the eponymously named lake. The lake lies in an area of transition from Cerrado to Atlantic Forest, and takes its name from the white sand substrate, giving it the aspect of a "white lagoon" from the air. The throaty and tireless "RONK RONK …" call was strongly reminiscent of the song of the Spot-billed Toucanet *Selenidera maculirostris* (Aves: Ramphastidae). Despite considerable active searching with a flashlight the numerous singing individuals could not be located. However, playback of a recording of the toucanet (Straneck, 1990) close to a singing frog in thick grass had the remarkable result of bringing it immediately into the open from where it could be captured and identified as *Physalaemus centralis*.

The Central Weeping Frog *Physalaemus centralis* Bokermann 1962 is widespread in the Pantanal and Cerrado eco-regions of eastern Bolivia, central Brazil and northeastern Paraguay. However, though it is not uncommon throughout much of its Brazilian range, the species has until recently been little recorded in Paraguay (Brusquetti & Lavilla, 2006), being known only from a small number of specimens from Departamentos Amambay and Canindeyú. This led to its classification as Vulnerable B2b(iii) at the national level during the last conservation assessment of Paraguayan amphibians (Motte et al., 2009). However, subsequent to the initial finding, the species has proved to be one of the most abundant Anuran species at RNLB (Smith et al., 2012), and considering its abundance in its Brazilian range, downgrading of the Paraguayan national conservation status to least concern seems warranted.

The issues of chronic under-sampling of amphibians in Paraguay, the reliance on rapid ecological assessments for distribution data and the effects of disproportionate field effort in certain favoured areas have already been raised (Smith et al., 2012). The example here further illustrates the benefits of employing a variety of different field techniques when monitoring amphibians, and the wider use of playback techniques, especially when confronted with unfamiliar calls produced by reclusive species, is one simple way in which techniques can be diversified and results potentially improved.

Thanks to the hard work of all the Para La Tierra (www.paralatierra.org) herpetology volunteers at Reserva Natural Laguna Blanca, who have made the long term protection of this small private reserve a national conservation priority through inventory work, establishing it as the most biodiverse protected area for reptiles and amphibians in the country.

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