Movement and home range of green pit vipers (*Trimeresurus* spp.) in a rural landscape in north-east Thailand

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ABSTRACT - Rural communities and agricultural landscapes serve as important areas for biodiversity, yet study of snakes in these fragmented environments is severely lacking. Hospital records indicate that green pit vipers (*Trimeresurus* spp.) inflict the highest number of venomous snakebites of any snake group in the Nakhon Ratchasima, Pak Thong Chai, and Wang Nam Khieo rural regions of north-east Thailand, causing debilitating injuries and, subsequently, negative perceptions of these species. We utilised radio telemetry to assess male and female *Trimeresurus albolabris* (N = 1, N = 1) and *T. macrops* (N = 2, N = 7) movements and home ranges in rural portions of the the Sakaerat Biosphere Reserve in north-east Thailand from October 2015 through January 2017. Green pit vipers of both species were tracked for a mean of 97.6 ± 15 days (median = 88, range 35-190). They moved a mean distance of 26.3 ± 3.32 meters between locations (median = 25.11, range 13.2-50.4) and exhibited mean minimum convex polygon home ranges of 0.14 ± 0.043 hectares (median = 0.095, range 0.006-0.423). Big-eyed green pit vipers (*T. macrops*) differed in movement patterns and home range size by sex and fecundity, although not statistically so. Understanding green pit viper space use will aid in future conservation and snakebite mitigation efforts for this interesting but severely understudied group.

INTRODUCTION

Conversion of natural habitats to agriculture is among the major threats to biodiversity (Foley et al., 2005). Previous studies of terrestrial vertebrate fauna in fragmented habitats have primarily focused on birds and mammals, with far fewer studies of reptiles and amphibians conducted (MacNally & Brown, 2001). Snakes play an important role in natural systems (predators and prey) and can serve as crucial bioindicators for environmental health (Campbell & Campbell, 2001). Lack of information pertaining to reptile and amphibian natural history in human modified landscapes limits conservation efforts (Bury, 2006).

South-east Asia is a complex biodiversity hotspot which faces many anthropogenic threats (Hughes, 2017). However, this region remains under-represented in studies of faunal response to habitat loss and human modified landscapes (Trimble & Aarde, 2012). More than 142 species of amphibians and 218 species of reptiles have been described from Thailand (IUCN, 2014), yet this country has the smallest area of remnant forest cover in south-east Asia.

Green pit vipers (*Trimeresurus* spp.) are a diverse and complex vertebrate predator group in the Asian tropics (Orlov et al., 2002). At least 8 species are present in Thailand (Chanhome et al., 2011), with new species having been described as recently as 2011 (*T. phuketensis*, Sumontha et al., 2011). One species of Thai green pit viper, *T. kanburiensis*, is listed as Endangered due to its limited distribution and illegal harvest for the pet trade (IUCN, 2012). Four other species have been labeled Data Deficient (IUCN, 2016). Taxonomic confusion and nomenclature inconsistency is particularly prevalent within the *Trimeresurus* group, making assessment and conservation of these snakes difficult (David et al., 2001; David et al., 2011).

A snakebite is a devastating environmental and occupational health hazard prevalent in rural developing countries, particularly in the tropics (Warrell, 2010). White-lipped (*T. albolabris*) and big-eyed (*T. macrops*) green pit vipers previously accounted for 40% of total venomous snake bites throughout Thailand (Viravan et al., 1992) and 95 % for the metropolitan city, Bangkok (Meemano et al., 1987; Mahasandana & Jintakune, 1990). Green pit vipers are regularly encountered in our study area and account for the highest number of snakebites of any venomous snake group present in the region (compilation of local hospital records, unpublished data). Limited spatial study has been previously been conducted with green pit vipers in the core area (most protected zone) of our study area (Strine, 2015), herein, we present the first investigation of green pit viper (*Trimeresurus* spp.) natural history in non-natural habitats. Our study bridges the current knowledge gap for green pit viper ecology in rural habitats, which are places of high conservation priority and human safety concern for this group of snakes.

We describe the spatial ecology of 2 green pit viper species in 3 study sites in agricultural areas of Sakaerat Biosphere Reserve. We report preliminary (1) movement, (2) home range, and (3) home range overlap patterns of *T. albolabris* and *T. macrops* to supplement future habitat selection, thermoregulation, prey selection, and spatial study of green pit vipers in rural habitats.
The study area, Sakaerat Biosphere Reserve (SBR), is located in Nakhon Ratchasima Province, Thailand (14.44-14.55° N, 101.88-101.95° E). Approximately 80 km² of SBR is designated as a core area and set aside to preserve and maintain species diversity, genetic variation, and landscapes and ecosystems. The buffer and transition areas surround the core area and consist primarily of agricultural areas and settlements. Together they comprise approximately 360 km².

We obtained green pit vipers for radio telemetry by opportunistic searches and concurrent site occupancy surveys at three study sites (designated plantation, canal, and pond) within the transition and buffer areas of SBR (Fig. 1). All three sites contained ephemeral water sources. The plantation and canal sites contained creeks and were downstream from a local dam and pond. The pond site contained a small pond (0.1 ha). The plantation site bordered the core, buffer, and transition areas of the Sakaerat Biosphere Reserve and was less than 50 m south of a major highway (Highway 304). It was characterised by a rubber plantation with heterogeneously disturbed forest (HDF) and bamboo patches. The canal site was located in the transition zone of SBR and was downstream from a local dam and pond. The pond site was located in the transition zone of SBR and was bordered by dipterocarp forest in the core area of the biosphere reserve. Dry dipterocarp forest is a forest type endemic to south-east Asia and characterised by thick Vietnamosasa pusilla grass ground cover and dipterocarp trees including Shorea siamensis and Shorea obtusa (Lamotte et al., 1998). Dry Dipterocarp forest is fire adapted with man-made or natural fires occurring in the dry season (March and April), and effectively clearing ground cover.

We captured vipers during visual searches and surveys at night, and recorded morphometrics (snout-vent length-SVL and body mass) the following day using the acrylic tube method and isoflurane anesthesia as described by Wilkinson (2014). We further assessed body condition for T. macrops using a scaled mass index (SMI, Peig and Greene, 2009) with previous T. macrops captured at the Sakaerat Biosphere Reserve as a comparison population (Strine et al., 2015), and checked for gravidity by presence of vitellogenic follicles with light palpation while under anesthesia. We then surgically implanted radio transmitters (1.8 g, Holohil BD2-THX) into the coelomic cavities of 11 vipers (7 female and 2 male T. macrops, of which 5 females were gravid and 2 were not; 1 female and 1 male T. albolabris) following (Reinert & Cundall, 1982; Hardy & Greene, 2000). All vipers were returned to their location of capture within 72 hours of capture.

When radio tracking snakes we attempted to locate each snake once per day and once per night. The day being the usual inactive period (when they were most likely to be sheltering or resting) and the night being the active period (more frequently observed foraging and moving). Relocations of 5 m or greater from the last observation were recorded as a “move”. Distances between moves were calculated in the field with handheld GPS units (Garmin GSPMap64s and eTrex10) and confirmed later using ArcGIS 10.1. We calculated number of moves, mean distance moved, and mean daily displacement (MDD, number of days tracked divided by distance moved).

We used the adehabitat package in program R (version 3.1.2) to estimate home range, using fixed kernel (50 and 99 %) and minimum convex polygon (MCP, 100 %) methods. Accuracy of both methods has been questioned (Row & Blouin- Demers, 2006), but were utilised in this work for comparison to previous studies. The least-squares cross validation method was used to select the smoothing factor for core (50 %) and activity (99 %) area utilisation distributions (Tiebout & Cary, 1987). Home range asymptotes using the MCP method were estimated for individual vipers using the hrBootstrap function in the sp package in program R.

We assessed statistical assumptions of normality and heterogeneity for all variables using Shapiro- Wilk and Levene tests. We implemented a GLMM (generalised linear mixed-effect model) with Wald test (R packages lme4 and car) to determine if days or number of fixes were better predictor covariables for home range size using MCP as a response variable and species as a random effect. Our data displayed a Poisson distribution; days and
fixes were discrete variables, MCP was continuous. We compared movement patterns and home range size of *T. macrops* using two sample independent t-tests, activity area by males and females was log transformed to obtain a normal distribution. Number of moves and SVL of gravid and non-gravid females could not be adequately transformed (obtaining a normal distribution) and we used non-parametric Wilcoxon rank sum tests (using the wilcox.test function in R) with the understanding that sample size is a severe limiting factor. Male and female *T. macrops* were compared, as were gravid and non-gravid female *T. macrops*. We also present descriptive spatial data for the single female and male *T. albolabris*. Means are reported for morphometric and space use data with standard error and medians.

Spatial overlap was calculated for concurrently tracked vipers using utilisation distribution overlap index (UDOI, Fieberg & Kochanny, 2005) with the R package KernSmooth for fixed kernels and the intersect function in ArcGIS 10.1 for MCP overlap analysis. Overlap is presented as area (ha and %) for MCP and as the UDOI for kernels. Values from the UDOI range from 0 to 1 which suggests less overlap relative to uniform space use, 1 if both home ranges are uniformly distributed and have 100% overlap, and values > 1 indicate higher than normal overlap relative to uniform space use. We also present results from 2 relatively infrequently used indices for comparative purposes; volume of intersection index (VI, Seidel, 1992; Kernohan et al., 2001) and Bhattacharyya’s affinity (BA, Bhattacharyya, 1943), both of which range from 0 (no overlap) to 1 (identical home ranges). Advantages and biases for the different overlap indices are discussed in Fieberg & Kochanny (2005).

**RESULTS**

Green pit vipers, *T. albolabris* (TRAL; *N* = 1 male, 1 female) and *T. macrops* (TRMA; *N* = 2 male, 7 female) were radio tracked for a mean of 97.6 ± 14 days (median = 88) and 66.0 ± 12.3 fixes (median = 67.0, Tables 1-3, Fig. 2). Results of tests for assumptions for statistical analyses for *T. macrops* varied in normality and homogeneity (Table 4). Male (*N* = 2) and female (*N* = 7) *T. macrops* were tracked for a similar number of fixes (*t* = 0.28, df = 7, *P* = 0.79) and days (*t* = -0.30, df = 7, *P* = 0.77). Gravid (*N* = 5) and non-gravid (*N* = 2) female *T. macrops* were tracked for a similar number of fixes (*t* = -0.83, df = 5, *P* = 0.44) and for a similar number of days (*t* = -0.32, df = 5, *P* = 0.76). Number of days and fixes for all vipers exhibited similar effects on MCP size (*β* = -0.005 ± 0.003, *t* = -0.152, *P* = 0.879 and *β* = 0.0008 ± 0.004, *t* = 0.231, *P* = 0.817; respectively, with intercept *β* = 0.126 ± 0.114, *t* = 1.104); i.e. unless an asymptote is reached number of fixes and to a lesser extent days tracked should be positively correlated with MCP size. Tracked male *T. macrops* were significantly smaller than females based on SVL (*t* = 3.26, df = 7, *P* = 0.01) and almost significantly by body mass (*t* = 2.26, df = 7, *P* = 0.058). Gravid *T. macrops* were not significantly longer or heavier than non-gravid females (SVL: *W* = 2, *P* = 0.33; body mass; *t* = -0.91, df = 5, *P* = 0.40).

Vipers (both species collectively) relocated a mean of 8.18 ± 1 times (median = 9), moved 26.3 ± 3.32 m (median = 25.11) straight line distance per relocation, and displayed mean MDD straight line distance of 0.32 ± 0.06 m/day (median = 0.30). Number of moves was not significantly different between males and female *T. macrops* (*t* = -0.07, df = 7, *P* = 0.94); distance moved was not significantly different (*t* = -1.01, df = 7, *P* = 0.34); and MDD was not significantly different (*t* = -0.56, df = 1.04, *P* = 0.67).

**Table 1.** Summary of *T. albolabris* (TRAL) radiotracked, tracking site (plantation- PL, & pond- PO), tracking start and end date, sex, snout-vent length (SVL, mm), mass (g), standardised mass index (SMI), number of days tracked (days), number of fixes recorded (fixes), mean distance between moves (distance, m), mean daily displacement (MDD, m/day), minimum convex polygon (MCP, 100%, ha), and 50 and 99% fixed kernels (50 and 99 FK, ha, respectively).

**Table 2.** Summary of male *T. macrops* (TRMA) radiotracked, tracking site (canal- CA, plantation- PL), tracking start and end date, snout-vent length (SVL, mm), mass (g), standardised mass index (SMI, using Strine et al., 2015 as comparison population), number of days tracked (days), number of fixes recorded (fixes), number of moves (moves), mean distance between moves (distance, m), mean daily displacement (MDD, m/day), minimum convex polygon (MCP, 100%, ha), and 50 and 99% fixed kernels (50 and 99 FK, ha, respectively) with total mean and standard error (SE).
### Table 3.
Summary of female *T. macrops* (TRMA) radiotracked, tracking site (canal- CA, plantation- PL), tracking start and end date, whether gravid or not, snout-vent length (SVL, mm), mass (g), standardized mass index (SMI, using Strine et al., 2015 as comparison population), number of days tracked (days), number of fixes recorded (fixes), number of moves (moves), mean distance between moves (distance, m), mean daily displacement (MDD, m/days), minimum convex polygon (MCP, 100%, ha), and 50 and 99% fixed kernels (50 and 99 FK, ha, respectively) with means and standard error (SE).

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<th>End</th>
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<th>Morphometrics</th>
<th>Tracking</th>
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<th>Home range</th>
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**Gravid Mean**:
- SVL: 583.4 ± 81.5
- Mass: 82.23 ± 10.3
- SMI: 98 ± 5.61
- Days: 82 ± 25
- Fixes: 6 ± 2
- Distance: 24.42 ± 4.78
- MDD: 22.9 ± 4.78
- MCP: 17.74 ± 5.14
- FK50: 2.1116 ± 1.091
- FK99: 1.736 ± 0.936

**Not Gravid Mean**:
- SVL: 554 ± 62.9
- Mass: 75.71 ± 17.74
- SMI: 80 ± 80
- Days: 40 ± 40
- Fixes: 23 ± 13
- Distance: 47.8 ± 11.0
- MDD: 22.9 ± 11.0
- MCP: 20.5 ± 5.14
- FK50: 2.033 ± 0.963
- FK99: 1.695 ± 0.963

**Both Mean**:
- SVL: 575 ± 62.9
- Mass: 80.37 ± 17.74
- SMI: 94 ± 80
- Days: 67 ± 67
- Fixes: 29 ± 29
- Distance: 47.8 ± 20.24
- MDD: 22.9 ± 12.11
- MCP: 17.74 ± 5.14
- FK50: 2.1116 ± 2.033
- FK99: 1.736 ± 1.695

### Table 4.
Summary for tests of normality (Shapiro-Wilk) and homogeneity (Levene) of data for *T. macrops*. * indicates normal, ** indicates homogenous.

<table>
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<th>Homogeneity</th>
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Similarly, non-gravid female *T. macrops* did not move significantly more frequently than gravid females (*W* = 10, *P* = 0.07). Distance moved by gravid and non-gravid female *T. macrops* was also similar (*t* = -0.18, *df* = 5, *P* = 0.87) as was MDD (*t* = -0.16, *df* = 5, *P* = 0.88).

Mean MCP home range size for all individuals was 0.14 ± 0.043 ha (median = 0.095), and 50% kernel and 99% kernel were 0.10 ± 0.026 ha (median = 0.084) and 0.69 ± 0.187 ha (median = 0.554), respectively. Male MCP home ranges were not significantly larger than females (*t* = -0.56, *df* = 1.04, *P* = 0.67). Similarly, core area (50% kernel) did not differ significantly between males and females (*t* = -0.44, *df* = 1.05, *P* = 0.73), nor did activity area (99%, *t* = 0.23, *df* = 1.05, *P* = 0.85). Non-gravid female MCP home range size was not significantly larger than gravid female (*t* = 0.87, *df* = 1.42, *P* = 0.51). Core area (50% kernel) were similar (*t* = 0.59, *df* = 5, *P* = 0.58), as were activity area (99%, *t* = 0.75, *df* = 5, *P* = 0.49).

Home range overlap between individual vipers was minimal (Table 5). Only 2 sets of MCP home ranges overlapped; minimal overlap (0.041 ha, 14.7%) was suggested at the plantation site with the male *T. albolabris* (TRAL016) and a male *T. macrops* (TRMA222), but a significant proportion of the two female *T. macrops* at...
the canal site (TRMA270 and 273) overlapped (0.094 ha, 78.9 %). Core area (50 %) home ranges did not overlap for any vipers. Activity area (99 %) overlap was minimal for UDOI (range 0-0.124). Interestingly, more sets of vipers (4) overlapped using the activity area method than MCP. Both BA (range 0-0.326) and VI (range 0-0.17) methods similarly suggested minimal activity area home overlap between individuals. All indices (UDOI, BA, and VI) suggested the highest set of home range overlap to be the pair of males at the Plantation site (TRAL016 and TRMA222).

### DISCUSSION

Both species of green pit vipers in our study displayed extremely limited movement and small home ranges (mean MCP size 0.14 ha), which were comparable to the smallest viper in the world (Bitis schneideri, mean 0.10 ha for females, Maritz & Alexander, 2012). High levels of activity and movement can increase predation risk of an organism (Gerritsen & Strickler, 1977), which could be a significant factor for vipers in rural communities. Habitat availability could also potentially affect space use, and reduction of natural features such as trees and hedges in non-natural environments may in turn influence arboreal green pit viper home range and movement.

Small home range and limited movement suggest both MCP and kernel methods to be imperfect estimators for green pit viper home range. The MCP method does not take into account movement (or lack thereof) and may include large areas of unused space (Nilsen et al., 2008), while kernels may overestimate overall home range size (Row & Blouin-Demers, 2006). Limited movement also potentially brings issues of autocorrelation between points (Laver & Kelly, 2011); we sought to limit temporal biological autocorrelation by taking data at different activity periods (night and day). Further study is needed to quantify both statistical and biological independence for fixes in snakes exhibiting limited movement and small home range size. Home range asymptotes were achieved for just over half of the study vipers, an issue traditionally attributed to limited movement combined with low tracking duration (Laver & Kelly, 2011). We were able to track vipers for a mean of 98 days (transmitter life being < 1 – 4 months), so limited movement (mean 8 moves, 26 m per move) likely influenced home range asymptotes more than tracking duration.

Adult male and female snakes face different challenges which may be reflected in behaviour, movement, and space use (Madsen, 1987; Shine, 2003). Drawing general conclusions between sex and movement and home range size of male and female snakes is difficult, although males are generally more active and exhibit larger home ranges than females (Macartney et al., 1988). Members of the genus *Trimeresurus* are sexually dimorphic (Orlov et al., 2002; Chanhome et al., 2011; Strine et al., 2015; Devan-Song et al., 2017), which likely also influences movement and space use between the sexes. The male *T. albolabris* and male *T. macrops* had higher MDD than females of their respective species (comparison of medians), although not statistically significantly so. Male *T. macrops* also exhibited larger home ranges (medians, not statistically significant) than female *T. macrops*, but Interestingly the female *T. albolabris* exhibited larger home range than the male *T. albolabris*. This may be explained at least in part by the longer duration of tracking of the female. Standardising tracking duration for all vipers would be ideal, although would have drastically reduced our already scarce data (number of vipers or days). All of the males were tracked outside of the previously described breeding seasons for their respective species (September to November, Chanhome et al., 2011), which could be one explanation as to why their home ranges were not significantly larger than females. It could also be due to small sample size. However, previous study of Armenian vipers (*Montivipera raddei*) in an agricultural setting also had comparable male and female home range sizes, which were attributed to habitat use in the matrix of fields and natural environments (Ettling et al., 2013).

<table>
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<th>MCP Overlap (ha)</th>
<th>MCP Overlap (%)</th>
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Gravid female big-eye green pit viper movement patterns were observed to be similar statistically to non-gravid females in our study, although median movement and distance was higher for non-gravid females. Most previous studies have found gravid snakes to move less frequently and smaller distances than non-gravid female snakes (Johnson, 2000). Gravidity can pose significant locomotor challenges for snakes, which can be reflected in movement and spatial patterns (Seigel et al., 1987). Further understanding of the benefits and mitigation of costs of similar space use by gravid and non-gravid female *T. macrops* is required.

Snakes are not known to exhibit territoriality and individuals from the same population often display widely overlapping home ranges (see review by Gregory et al., 1987; Weatherhead and Hoysak, 1988; Secor, 1994). Small home range size, and thus limited chance of encounter, may explain the limited overlap we observed between radio tracked snakes. Estimation of density via mark-recapture with concurrent radiotelemetry study could better clarify encounter rate between individual vipers, as our small sample may not be representative.

Tracking males outside of the breeding season may explain limited home range overlap of males with females. Interestingly, the highest UDOI home range overlap observed in our study was with two male green pit vipers of different species. The largest MCP home range overlap, however, was by two female *T. macrops* (TRMA270 and 273). This large overlap could be due to a general lack of intraspecific sexual antagonism observed between female snakes (Shine, 1994) or different use of resources as one of the vipers was gravid and the other was not (Shine, 1979; Macartney et al., 1988).

Caution is expressed regarding extrapolating our preliminary findings to other populations or species, which may exhibit different patterns in rural communities and other non-natural environments. Multiple previous natural history studies of large bodied vipers in rural habitats have observed both indirect and intentional killing of study animals by humans (Bonnet et al., 1993; Durbian, 2006; Wittenberg, 2012). However, we did not observe human caused mortality to the snakes in our study, which we attribute primarily to the cryptic and perceived inoffensive nature of green pit vipers. Two of the *T. macrops* in our study spent extensive time (TRMA221 and 273, > 2 weeks) less than 10 m from human habitations. The homeowners were tolerant of the vipers’ presence when informed by researchers so long as the study animals did not directly enter living quarters. While study vipers were observed in plantations, they did not fully enter agricultural fields (although they did utilize edges) which likely reduced chance of mortality by certain agricultural practices (tilling, harvesting, etc.) which have been observed for other snakes in the SBR (Kneirem et al., 2017). Limited movement and small home range size may facilitate green pit viper persistence and resilience in our study area, although habitat selection investigation is needed for future conservation measures.

Previous studies have suggested that while well intentioned, translocation can have disastrous results for “nuisance” snakes moved outside of their home ranges. Irregular movement patterns, increased home range sizes, and decreased survival rates have been recorded for translocated snakes (Fitch & Hampton, 1971; Nowak et al., 2002; Barve et al., 2013). Translocation is generally perceived by the public to be a humane strategy because it does not result in the immediate death of the individual (Riedl et al., 2008), however, serious consideration is required in particular for relocation of snakes which exhibit small home ranges. Previous study of *T. albolabris*...
in Hong Kong by Devan-Song (2014) found a 100% male and 71% female mortality rate for individuals translocated outside of their home ranges. Green pit vipers are commonly translocated in rural north-east Thailand when encountered in human habitats, and our findings of very small home ranges of *T. albolabris* and *T. macrops* may present significant management implications. Future studies of the effects of short and long distance (within and outside of home range, respectively) translocation on green pit viper health and survival are needed.

While both study species are listed as Least Concern by the IUCN (IUCN, 2012), *T. macrops* were encountered relatively infrequently (9.1 surveyor hours to find one at all study locations, total 228.7 surveyor hours surveyed during study period) and only the 2 *T. albolabris* were observed at the study sites during the entire study period. Overall, there exists a severe limitation in small bodied snakes as track-able individuals are quite rare given mean body masses often fall well below the minimum size for radio transmitter implantation. Mean body mass for *T. macrops* at the SBR is very close to this accepted minimum, particularly males (Strine et al., 2015). Ethical constraints of number of transmitter implantations due to short battery life must be considered also. Although radiotelemetry can prove challenging for study of small bodied snakes, there are many aspects of green pit viper natural history which still require investigation and can build upon our preliminary work.

Thermoregulation and prey availability have previously been suggested to influence snake space and habitat use in rural environments (Durner & Gates, 1993; Shine & Fitzgerald, 1996; Wisler et al., 2008). Future study of thermoregulation, habitat selection, and prey selection of green pit vipers in rural habitats is required. How vipers utilise non-natural prey and habitat features are subjects which still require investigation and can build upon our preliminary work.

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