THE BRITISH HERPETOLOGICAL SOCIETY

BULLETIN



No. 45 Autumn 1993

THE BRITISH HERPETOLOGICAL SOCIETY

c/o Zoological Society of London Regent's Park, London NW1 4RY

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The British Herpetological Society was founded in 1947 by a group of well-known naturalists, with the broad aim of catering for all interests in reptiles and amphibians. Four particular areas of activity have developed within the Society:

The Captive Breeding Committee is actively involved in promoting the captive breeding and responsible husbandry of reptiles and amphibians. It also and advises on aspects of national and international legislation affecting the keeping, breeding, farming and substainable utilisation of reptiles and amphibians. Special meetings are held and publications produced to fulfill these aims.

The Conservation Committee is actively engaged in field study, conservation management and political lobbying with a view to improving the status and future prospects of our native British species. It is the accepted authority on reptile and amphibian conservation in the UK, works in close collaboration with the Herpetological Conservation Trust and has an advisory role to Nature Conservancy Councils (the statutory government bodies). A number of nature reserves are owned or leased, and all Society Members are encouraged to become involved in habitat management.

The Education Committee promotes all aspects of the Society through the Media, schools, lectures, field trips and displays. It also runs the junior section of the Society – THE YOUNG HERPETOLOGISTS CLUB (YHC). YHC Members receive their own newsletter and, among other activities, are invited to participate in an annual "camp" arranged in an area of outstanding herpetological interest.

The Research Committee includes professional scientists within the ranks of the Society, organises scientific meetings on amphibian and reptile biology and promotes The Herpetological Journal, the Society's scientific publication.

Meetings

A number of meetings and events take place throughout the year, covering a wide range of interests.

Publications

The BHS Bulletin, Herpetological Journal and YHC Newsletter are all produced quarterly. There are in addition a number of specialised publications available to Members and produced by the various Committees, such as notes on the care of species in captivity, books and conservation leaflets.

Subscriptions

All adult subscriptions become due on the first day of January each year. Payment by Banker's Order is much preferred.

Ordinary Members Full Members Family Members	£20 £25 £30/£37.50	(Receive Bulletin only) (Receive Bulletin and Journal) (Without/with Journal) Family members with children also receive the YHC Newsletter
Student Members	£18	(Receive Bulletin and Journal)
Institutional rates YHC (Age 9-18):	£36	(Receive Bulletin and Journal)
Basic Membership	£5	(Receive YHC Newsletter)
Bulletin Membership	£10	(Receive Newsletter and Bulletin)
Group Membership	-	For Schools, Youth Groups etc. Contact Education Officer (Address on inside of back cover) for details)

Correspondence, Membership applications, subscription renewals and purchase orders for publications should be addressed to the Secretary (address as at page top) EXCEPT for YHC matters. YHC Membership and renewal details are available from the Education Officer (address on inside of back cover). PLEASE INCLUDE A STAMP-ADDRESSED ENVELOPE WHEN WRITING TO THE SOCIETY.

The Society does not, as a body, hold itself responsible for statements made or opinions expressed in the Bulletin; nor does the Editorial necessarily express the official opinion of the Society.

The Bulletin is edited and produced by

Simon Townson and Neill Clark.

Contributions and correspondence arising from the Bulletin should be sent to: Neill Clark, 15 Rivenhall End, Welwyn Garden City, Herts AL7 2PJ.

FRONT COVER

Male Pfeffer's Chameleon, *Chamaeleo pfefferi*, 1330 metres Nsoung Village, Mt. Manenguba, Cameroon. Photo by Chris Wild (see page 25).

BHS REMAINING MEETINGS FOR 1993

November 6thCAPTIVE BREEDING COMMITTEE OPEN MEETING
New Denham Community Centre, Uxbridge Road, New Denham.
2.30-6.30 p.m.December 4thRESEARCH COMMITTEE MEETING.

Birkbeck College, Malet Street, London WC1. 10.30-16.30.

BRITISH HERPETOLOGICAL SOCIETY MEETINGS FOR 1994

The following list provides dates and, in some cases, only preliminary details of meetings scheduled for 1994. Where information is currently incomplete, further details will follow in later Bulletins closer to the events in question.

February 5th	Herpetofauna Recorders' Meeting (HRM) 1994. (University of Manchester).
March 19th	 Annual General Meeting (Birkbeck College, London) Speakers will be: Rob Quest (Heathrow Customs): "Reptile Imports" (2) Dr Andy Smart (Bristol University): "Nesting success of green turtles at Kazanli, Turkey"
	Dr S. Hugh-Jones (Cambridge University): "Snakes, frogs, Indians and other herps in north-west Amazonia"
May 1st	Joint Conservation & Federation Committee meeting.
May 7th	Captive Breeding Committee Amphibian meeting (New Denham)
May 15th	"Leapers & Creepers" events, organised by Surrey Wildlife Trust, following by later (7-9pm) visist to Beam Brook*
July 2nd	Captive Breeding Committee Animal Husbandry workshop (New Denham)
October 15th	 Autumn General Meeting (Birkbeck College, London) Speakers will be: Chris Wild (Nottingham): "The montane chameleons of the Cameroons Highlands" (2) Dr Jim Foster (Durrell Institute of Conservation & Ecology): "Reptile conservation in south India" (3) Dr Angelo Lambiris (Essex): "Southern African amphibians"
November 5th	Captive Breeding Committee Captive Stock Sale (New Denham)

December 3rd Research Committee meeting (Birkbeck College, London)

* For those unfamiliar with Beam Brook, this is an old nursery site with a series of small ponds near the village of Newdigate (south of Dorking) in Surrey. Since 1905 it has been home to a variety of both native and introduced species of amphibians and reptiles; it is especially renowned for its colonies of edible frogs, Italian crested newts and alpine newts. BHS Members may either turn up at 7 pm at the Beam Brook site, or go there following attendance at the "Leapers & Creepers" session that runs through the day and should finish by 5 pm. There will no charge for entry, and Members will be allowed to examine and net the various ponds during the visit (but not to take away any animals caught).

For those going directly to Beam Brook, the nursery is situated in Partridge Lane, approximately 1 mile due east of Newdigate village (Map ref. TQ 216423). It is reached from Newdigate by taking the road leading out to the north-east, which after about a mile turns south-east and becomes Partridge Lane. Beam Brook is signed on the west side of the road.

BHS CONSERVATION COMMITTEE

Every July I visit a series of large compost heaps, consisting of sand and rotting seaweed that has been systematically cleared from a stretch of beach that is subjected to a lot of public pressure in the holiday season. This 'waste' is then dumped away from the beach and left to rot. The area is used by many Grass Snakes (*Natrix natrix*) for egg-laying, enabling the field herpetologist to observe and monitor the animals from year to year. The sandy component of the heaps makes it easy for the females to deposit their eggs and the warm, rotting seaweed provides a suitable temperature for incubation. Last year however, I found several adult animals that had been clubbed to death and no signs of egg laying in the heaps. Hopefully, some will return this year but the case serves to highlight the threats to all our British snakes in the form of persecution.

Current agricultural practices have also led to the loss of many grass snake breeding sites and as a conservation measure, the BHSCC are providing suitable egg-laying sites in our reserves. The BHSCC has just published a new Information Sheet giving guidelines for the making of grass snake egg-laying sites and we believe that if more egg-laying sites are provided, the species can greatly benefit.

The Information sheet has been compiled using the results of experiments in the UK, advice from gardening organisations on the creation of compost heaps and proven research from the Netherlands. The best time to start constructing a heap is in the Spring so that it will be ready for the egg-laying season in late June/early July.

If you would like a copy of this Information Sheet please send a SAE to:

BHSCC, 28, Old Fort Road, Shoreham-by-Sea, Sussex BN43 5RJ.

Remember the BHSCC also publishes leaflets on:

Surveying for Amphibians Save our Reptiles Garden Ponds as Amphibian Sanctuaries

Please write to the same address for further information enclosing a SAE.

Jan Clemons

NATTERJACK TOAD EXTINCTIONS: THE MALES GO FIRST TREVOR BEEBEE

434 Falmer Road, Woodingdean, Brighton BN2 6LG

Although extinction of plant and animal species is a subject of intense concern and public debate, few studies have been made about the process of extinction itself. This is perhaps not too surprising, bearing in mind the difficulties in finding everdiminishing numbers of a species in terminal decline. Natterjack toads have experienced many local extinctions in Britain over the past century, and despite current conservation efforts some of the smaller populations of this species still hover close to, and occasionally go over, the brink. One aspect of these minor tragedies seems to be that the sexes do not fare equally during these declines.

Detailed studies of relatively healthy natterjack populations give some clues about what is likely to happen when things go wrong. Life tables consistently indicate that female natterjacks live longer in the wild than do males, although the extent of this difference between the sexes differs substantially between populations. Females probably live longer mainly because they enjoy a safer lifestyle during the breeding season; mostly they make a single night-time visit to a pond each year, lay their spawn and rapidly retire to the relative safety of their summer burrows. By contrast, males stay around the ponds for weeks or months and many are known to perish at the hands, claws, beaks or teeth of predators during this vulnerable time. Even this may not be the whole of the story, and males may also pay a further penalty as a result of their innate male physiology; there is often a sharp decline in male numbers when they reach the age of 6-7 years in the wild, for reasons difficult to explain on the basis of predation. Although some males certainly can live longer than this in captivity, many do seem to keel over at roughly this age no matter how well they are kept.

The prediction of these observations is that in the final stages of extinction, small numbers of females may be the last survivors in sites where no males remain to mate with them. This prediction is compatible with what has been seen over the past few years at three localities, all in north-west England, where natterjacks have almost or completely disappeared. At one site in Lancashire, numbers of calling males declined during the 1980s following interference with the breeding site water regime and a single one was last seen in 1988. In 1990, however, two females (one dead, one alive) were located near the old breeding pond. Apart from this, and despite fairly intense monitoring, no other natterjacks have been seen at this locality in the intervening or subsequent years. At a second site, on the Merseyside coast, an isolated population declined through lack of available breeding ponds during the early 1980s and in 1987 only four large females were found in the vicinity of a new pond made specially for them. In this case, a single male from nearby on the coast was brought over and since then the population has begun to revive. Finally, at another even more isolated Merseyside site a steady decline left apparently only a single male calling by 1991. No natterjacks were seen in 1992, but this year (1993) one or two females did spawn in a new pond made specially for them. However, no males were seen or heard and none of the spawn developed. This site is not intensively monitored, so we cannot be sure about events here, but it maybe that the females simply deposited their spawn in the absence of male partners. This has been observed, albeit rarely, in captivity.

The difference in longevity between male and female natterjacks probably only averages out at a year or two in most populations, but it can be much more and in the extreme case of the single surviving native heathland population in southern England females live up to twice as long as males, a net difference of some 7-8 years. Clearly natterjack site managers should be aware of the sex bias during population declines, and be prepared to address the problem (perhaps by supplementation with males from adjacent populations, or suitable captive-bred stock) if the need arises. Evidently the last calling male does not mean the last natterjack, and prompt action may still save the day even in such apparently dire circumstances.

It would be interesting to know how typical natterjacks are with respect to this aspect of population dynamics. It seems to me quite likely that similar sex biases will occur in numerous other amphibian species, since males generally have the riskier lifestyles. Indeed, female left-overs may be the common immediate precursor to extinction in a variety of animal groups. Could this situation create conditions for one possible escape route from extinction, notably the evolution of parthenogenesis? Although rare, all-female populations do appear from time to time and include some wellstudied lizards in Europe and North America. Parthenogenesis seems to be one of Nature's side-shows, with little long-term prospects for the organisms which plump for it, but having no males around would certainly seem to provide one powerful force in this unenviable direction.

HIGH WATER POTENTIAL VERMICULITE AS AN INCUBATION MEDIUM FOR REPTILE EGGS

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ABSTRACT

Eggs with flexible and hard shells (N = 216) representing five lizard and seven snake species and subspecies, were incubated in ventilated incubation containers with a water reservoir and a saturated coarse grade vermiculite and water medium of 1: 4 (g/g). Hatching success was 95%. This was contrary to the widely used method of incubating reptile eggs in sealed containers with a vermiculite and water medium of 1: 1 (g/g), or less. The high hatching success was ascribed to the above factors as to well as the non disturbance of containers during incubation and the lack of fungal infections on fertile eggs. As a control, snake eggs (N = 42) were incubated under the same conditions using sealed containers and a fine grade vermiculite and water medium of 1: 1 (g/g). The control incubations resulted in a comparatively poor hatching success of 32%. The results and effects on the hatchability of reptile eggs in other studies where vermiculite with varying water potentials were used, are also discussed.

INTRODUCTION

The water content of an incubation medium may be expressed as either a ratio or a measurement. In the former, the ratio expresses the amount of water added to the incubation medium in either g/g or g/ml. In the latter, the water potential of the incubation medium is expressed as kiloPascal (kPa) (Tracy, 1980; Packard & Packard, 1987; Plummer & Snell, 1988). KiloPascal measurements are usually determined by the use of thermocouple hygrometry with a calibrated sample chamber (Wescor C-52) and a dewpoint microvoltmeter (Wescor HR-33T), or a water potential data system (Wescor HP-115) (Packard & Packard, 1987; Plummer & Snell, 1988). As water potential determination equipment is not always available, the vermiculite/ water ratio method is preferred by many breeders. An explanation as to the use of these two water determining measurements in text, is given under the Methods section.

The incubation methods of many researchers (Morgan, 1988; Boycott & Morgan, 1988; Deeming, 1989) have been based on that of Tryon (1975), who used a vermiculite and water medium of 1: 1 (g/g) (-160 kPa). Incubation mediums with a water potential of below -160 kPa (V/WR 1: 1) have been used extensively. Water potentials of -200 kPa (V/WR 1: 0,71) to -2000 kPa (V/WR 1: <0.1) were used by Plummer & Snell (1988); -300 kPa (V/WR 1: 0.43) to -900 kPa (V/WR 1: 0.17) by Packard, Taigen, Boardman, Packard & Tracy (1979); -200 kPa (V/WR 1: 0.71) to -450 kPa (V/WR 1: 0.32) by Packard, Packard & Boardman (1982); -150 kPa (V/WR 1: 1) to -950 kPa (V/WR 1: 0.15) by Packard & Packard (1987); and -825 kPa (V/WR 1: 0.18) and -1500 kPa (V/WR 1: 0.10) by Packard, Packard & Benigan (1991). Mattison (1991) recommended a water potential of -270 kPa (V/WR 1: 0.48) for husbandry use. Using sand in ventilated vials, Ferguson & Snell (1986) used a sand/water ratio

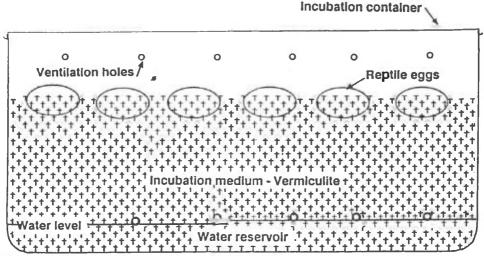


Fig.1. A diagrammatic presentation of a ventilated incubation container showing the water reservoir (not to scale).

of 1: 0.05 (<-1500 kPa). Despite the extensive use of drier incubation mediums, evidence in the literature strongly suggests that there may be distinct advantages in using wetter incubation mediums, the advantages of which will be discussed later in this paper. However, the use of incubation mediums with water potentials of above -160 kPa (V/WR 1: 1) appear to be relatively uncommon, although becoming more popular. Packard *et al.* (1979) used a water potential of -50 kPa (V/WR 1: 3.6); Packard *et. al.* (1982), -100 kPa (V/WR 1: 2.8); and Ford & Seigel (1989), and Seigel & Ford (1991), -70 kPa (V/WR 1: 3.1).

When using sealed incubation containers, researchers have found it necessary to open these repeatedly for ventilation purposes and sometimes the addition of water. Boycott & Morgan (1988) opened containers every two to three days. Anstandig (1984) weekly, and Deeming (1989) once or twice a week. Even when using commercially available incubators (Hova-Bator), it is recommended that the incubator be opened for occasional misting. Tracy (1980) also mentioned that the spread of fungal infections on incubating eggs may be a problem related to the use of sealed containers.

The choice of incubation mediums appears to be an individual choice, with a variety of mediums being utilized. Patterson (1987) suggested leaving soft-shelled eggs *in situ* and using wet paper towelling as an incubation medium. For the incubation of gecko eggs, Miller (1982, 1983) used either slightly damp sand, or peat and bark chips, with water at the bottom of the container. Mattison (1982) suggested the use of fine vermiculite, while Mattison (1991) gave sand, perlite, and sawdust as alternatives to vermiculite, and Ferguson & Snell (1986) used sand. Anstandig (1984) preferred wet, squeezed out vermiculite. However, in the majority of studies referred to in this investigation, vermiculite appears to be the most widely used incubation medium.

The primary aim of this investigation was to examine the hatching results of incubating reptile eggs in ventilated incubation containers with a reservoir of water and an initially saturated incubation medium of large grade vermiculite, and to compare these results to control incubations using fine grade vermiculite and water with a V/WR of 1: 1 in sealed containers. It was also deemed pertinent to examine the incubation results of other researchers who have used vermiculite and water in varying proportions.

MATERIALS AND METHODS

Plastic incubation containers with sealing lids, such as Tupperware (length (L) = 270 mm x width (W) = 270 mm x height (H) = 145 mm), or plastic ice cream containers (L = 315 mm x W = 210 mm x H = 105 mm, and L = 220 mm x W = 220 mm x W = 210 mm x H = 140 mm) were used for snake eggs, and similar, but smaller containers for lizard eggs. A series of 3 mm diameter holes (4 to 6 per side) were drilled around the bottom of the container, 5 mm up from the base, and another series of 3 mm holes (4 to 6 per side) were drilled around the top of the container, 10 mm down from the top edge (Fig. 1).

Tests were carried out to determine the saturation point of coarse (large) and fine (small) grade vermiculite, plus a small excess quantity of free water to form a reservoir' at the bottom of the container. Saturation points were determined by gradually adding measured quantities of water to the vermiculite, and mixing until, after being left to stand a short while, a small quantity of water accumulated at the bottom of the container. This determination resulted in a saturation V/WR for large and fine grade vermiculite of 1: 3,8 (g/g) (0,0 kPa) and 1: 3,2 (g/g) (0,0 kPa) respectively. The incubation medium used was granular, large grade Mandoval Vicafil vermiculite with a large grain size average of 8 mm x 6 mm x 6 mm (although smaller grain sizes were in abundance). For practical use, vermiculite was simply soaked in a bucket of water for 30 minutes before use.

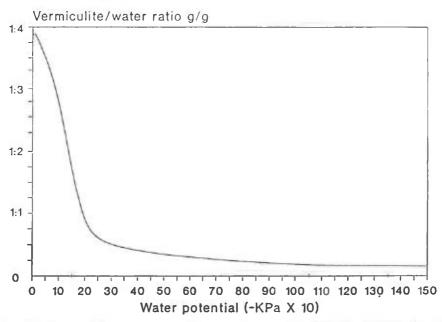
One-hundred and thirteen snake and lizard eggs, the latter consisting of both hard and soft-shelled eggs, were incubated by the above method between 1987 and 1990. A further 103 snake eggs were incubated between 1991 and 1992. As the number of eggs incubated during these two periods were similar, it was decided to regard them as separate samples. Eggs of species incubated during the 1987/90 period are marked *, eggs of species incubated during the 1991/92 period are marked **, and the eggs of species incubated during both periods are marked ***.

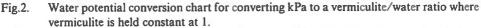
Sauria: Lygodactylus capensis capensis *, L. ocellatus *, Pachydactylus capensis capensis *, Mabuya quinquetaeniata margaritifer *, and Gerrhosaurus flavigulars *.

Serpentes: Dasypeltis scabra *, Pituophis melanoleucus sayi *, Elaphe obsoleta quadrivittata X E. o. obsoleta *, Lamprophis fuliginosus **, E. guttata guttata **, E. o. quadrivittata ***, E. taeniura freisii ***, and E. o. quadrivittata x E. o. obsoleta X E. o. quadrivittata E. o. obsoleta (F1) ***.

Incubation containers were filled with saturated vermiculite to a minimum depth of 6 cm, with maximum depth being determined by the upper level of vermiculite being from 0 cm to 1 cm below the level of the top row of holes. The latter always applied in this study. Soft-shelled eggs were loosely buried two-thirds into the medium and very lightly covered with a sprinkling of vermiculite, while hard-shelled eggs were buried one-third into the medium and left exposed. The lids were then firmly closed.

As no environmental control chambers were available for incubating the eggs, and in order to compensate for a summer ambient temperature variation of between 8°C and 38°C, a simple incubator was devised. This was constructed from a steel trunk (L = 850 mm x W = 470 mm x H = 340 mm), painted white, both inside and out, and lined with 12,5 mm polystyrene sheeting. A heater and wafer thermostat were fitted to the inside of the lid. Two (4 mm) ventilation holes were dilled just above the floor at each bottom corner of the trunk, and a 6 mm ventilation hole was drilled in the centre of the lid. The ventilation hole in the lid could be closed to





reduce air circulation when cooler conditions prevailed. In this manner incubation temperature was kept within a 25,5°C; to 29,5°C range. An open container of water was also placed inside the incubator.

For comparative purposes, an additional 42 snake eggs of Dasypeltis scabra, Elaphe obsoleta quadrivittata, and Pituophis melanoleucus sayi, from the 1987/90 period, were incubated using completely sealed containers, and incubation mediums of flaked and fine granular vermiculite. The mediums were mixed with water in a ratio of 1: 1 (g/g) (-160 kPa). During these incubations the containers were opened twice a week for aeration and sometimes the addition of water. These eggs were then incubated under exactly the same conditions as previously mentioned.

Both water determining values for vermiculite, kPa and V/WR, have been provided in order to accommodate both schools of use. Conversion values were extrapolated from the kPa to V/MR graph (Fig. 2). Because the capacity of vermiculite to hold water may not only vary with size, but also type (eg. flaked or granular); because different grades of vermiculite have been used by different researchers, with grade size often not being mentioned; and because grade size may vary from country to country; extrapolated comparative values should be considered as approximations.

RESULTS

Of the 113 eggs from the 1987/90, V/WR 1: 4 incubations, 22 eggs (19,5%) were presumed infecund due to there being no observable indication of embryonic development at the end of the incubation period. Of the remaining 91 fertile eggs, 86 eggs hatched successfully (94,5%) hatching success), while five eggs containing well developed embryos failed to hatch (5,5%) mortality).

Of the 103 eggs from the 1991/92, V/WR 1:4 incubations, seven eggs (6,8%) were presumed infecund due to there being no observable indication of embryonic development at the end of the incubation period. Of the remaining 96 fertile eggs,

91 eggs hatched successfully (94,8% hatching success), while five eggs containing well developed embryos failed to hatch (5,2% mortality). Further analysis revealed that in 60% of clutches (6) there was nil mortality, in 30% of clutches (3) there was one mortality, and in 10% of clutches (1) there were two mortalities.

Fungal infections remained confined to the infecund eggs during incubation, with no apparent infections spreading to, or developing on, fertile eggs. The spread of fungal infections to eggs containing dead embryos was rarely observed, and could be explained by the embryos only dying at a late stage of their development, with infections not having had time to become significantly established.

The incubations of eggs from the 1987/90 period with a V/WR of 1: 1, sealed containers, and fine vermiculite, resulted in the mortality rate being higher than the survival rate. Of the 42 eggs incubated, six eggs were presumed to be infecund due to there being no obvious indication of embryonic development at the end of the incubation period. Of the remaining 36 eggs, 13 eggs hatched successfully (36,1% hatching success), while 23 eggs contained developed embryos which failed to hatch (63,8% mortality). D. scabra recorded a 67% mortality, E. o. quadrivittata a 48% mortality, and P. melanoleucus sayi a 73% mortality.

DISCUSSION

There appeared to be numerous advantages in using a combination of coarse grade vermiculite, with either a V/WR of 1:4, or by simply saturating the vermiculite, and ventilated incubation containers. One apparent advantage was the minimal disturbance of containers during incubation. As the ventilation holes allowed for a certain amount of evaporation during incubation, it was necessary to keep the vermiculite moist without having to continuously open the containers. This was achieved by the lower row of holes providing a 5 mm deep reservoir of water at the bottom of the container. Owing to natural drainage, the upper lavers of vermiculite became less saturated after standing for a short period, and therefore the eggs incubated in the drained upper layers, free from any hygroscopic influence of the reservoir. An advantage of using initially saturated vermiculite was that water potential, as of water/vermiculite ratios and kPa, become irrelevant. The containers were usually only opened 10 to 14 days after the eggs were set, as this appeared to be about the time it took for infecund eggs to become obvious, and again in the middle of the incubation period, but never within the last two to three weeks. When the containers were opened, the water level in the reservoir was also carefully checked, and water added down the side of the container if necessary. The series of holes in the incubation containers, and incubator, also allowed for an exchange of air through the coarse vermiculite. Trials indicated that containers could be left undisturbed for up to four months in the incubator without any appreciable desiccation of the medium.

A particular disadvantage of using fine granular or flaked vermiculite was that it tended to compact around the eggs during incubation, with flaked vermiculite sticking in layers to the eggs. This situation was aggravated by increases in the water portion of the medium and may have well contributed to the suffocation of the eggs by inhibiting the exchange of gasses between the egg and the atmosphere. It was concluded that fine granular and flaked vermiculite were unsuitable as incubation mediums. When using the aforementioned incubation mediums as comparisons to other studies, it should be noted that the difference between wet and dry will be relative to the moisture range being used, or compared to. In this investigation, and for convenience, a V/WR of 1: 1 (-160 kPa) has been taken as an arbitrary division between wet and dry incubation mediums.

The temperature fluctuation in the incubator was felt to the advantageous, in that when the eggs of environmentally sex determined (ESD) species were incubated, the temperature variation possibly allowed for a more 50/50 related sex ratio to develop. This latter statement may appear somewhat incongruous when referring to snakes, as the literature indicates that there is little or no ESD bias in snakes (Bull, 1980). However, a clutch of *E. o quadrivittata* x *E. o. obsoleta* X *E. o. quadrivittata* x *E. o. obsoleta* (F1) eggs which were excluded from the incubator and allowed to reach 38°C, produced 100% females, 50% of which had chronically reduced eyes of only 10% normal size, while incubated clutches produced near to 50:50 males and females.

From the advanced developmental stages of dead embryos in the eggs, it was judged that the majority of embryo mortality occurred within the few weeks prior to hatching, and therefore could possibly have been related to the disturbance of the containers. It was also found over the numerous incubations that embryos of certain species appeared to react more adversely to the disturbance factor than others. From the V/WR 1:1 mortalities, it was noted that *P. melanoleucus sayi* eggs had a 73% mortality, *D. scabra* a 67% mortality, and *E. o. quadrivittata* a 48% mortality, all of which were incubated under the same conditions. This was contrary to the results of the undisturbed incubations where no particular pattern emerged.

Hatchlings were left in the incubation containers until their first slough, only after which were they removed to rearing containers.

Numerous studies have indicated that there is a strong relationship, particularly in soft-shelled eggs, between hatchability, size of hatchlings, and the water potential of the incubation medium (Tracy, 1980; Packard & Packard, 1987; Deeming & Ferguson, 1991; Packard *et al.* 1991; Christina, Lawrence & Snell, 1991). From these studies it would appear that there may be defininte advantages arising from the use of wetter mediums. Packard *et al.* (1982, 1991) noted that the size of hatchling turtles was positively correlated with the net water-exchange experienced by the incubating eggs, with a wetter medium resulting in a higher hatching success and larger hatchlings, sometimes after a longer incubation period. As with this study, Packard *et al.* (1991) also found a correlation between hatching success and water potential, with incubations on the wettest medium (-150 kPa; V/WR = 1: 1.13) producing a hatching success of 87.9%, on the intermediate medium (-825 kPa; V/WR = 1: 0.18) 51.4%, and on the driest medium (-1500 kPa; V/WR = 1:0.01) 25.7%. Galapagos iguanas, *Conolophus subcristatus*, from wet nests have been also been reported as having a higher survival rate than hatchlings from dryer nests (Christian *et al.*, 1991).

Eggs of the Cuban iguana, Cyclura nubila, showed a negative correlation between the percentage yolk and the percentage fat in respect to the whole hatching mass, although in this instance water potential in natural nests appeared to have no effect on hatchling size (Christain *et al.*, 1991). It was also noted that the more negative the nest water potential, the greater the retarding effect was on the conversion of yolk to fat, and consequentially the accrual of fat and the depletion of yolk was highest in hatchlings from wetter nests (Christian *et al.*, 1991). Although yolk was found to be an energy source utilized for growth, activity, and maintenance after hatching, the conversion of yolk to fat prior to hatching rendered fat as a longterm storage media which persisted in neonates for longer periods than absorbed yolk (Christian *et al.*, 1991). Water limitations during incubation may therefore result in the introduction of severe survival limitations for *C. nubila*, with negative ecological and energetic implications. Converseley, in species such as the Green iguana, *Iguana iguana*, and the Green sea turtle, *Chelonia mydas* yolk was found to be the quantitively greater storage medium with post-hatching energy being largely expended on activity (Christian et al., 1991). Snapping turtles, Chelydra serpentina, have also been found to absorb energy reserves from yolk more rapidly when incubated in wetter environments (Packard et al, 1982). Therefore, the conversion of yolk to fat may be dependent on whether energy expenditure in hatchlings is to be utilized for activity, as in *I. iguana*, or for growth maintenance, as in *C. nubila*. In these examples it would appear irrelevant as to whether yolk or fat is the greater storage medium, as life history strategies would appear to dictate that the most important energy source for a particular species will be greater when eggs are incubated on a wetter medium.

When the snake, *Opheodrys aestivus*, was given a choice of hydric environments in which to deposit eggs, preference was given to the wetter mediums of -200 kPa (V/WR = 1: 0.71) and -300 kPa (V/WR = 1: 0.45) (Plummer & Snell, 1988). Although these water potential may appear low when compared to this investigation, they were the highest presented to the snakes, and were also relatively high when considering that the driest environment presented was -2000 kPa (V/WR = 1: 0.01).

It was suggested by Tracy (1980) that fungi invaded viable Sceloporus undulatus eggs, and eventually killed the embroyonic lizards, rather than invading eggs containing dead embryos. The highest mortality rate of 72% was recorded by Tracy (1980) on the wettest medium of -200 kPa (V/WR = 1: 0.71), and it was speculated that this was either due to the initial high influx of water into the eggs resulting in an increased invasion of microbes, or the suffocation of the embryo due to an envionment in which the transport of respiratory gases was inhibited because the pore space between the vermiculite particles was more severely occluded by water. With the exception of the latter statement, this was contrary to all observations during this investigation where a considerably wetter medium (6 X) was used and fungal infections were primarily attributable to the intertility of the egg, and secondly, to the death of the embryo during development. It has been noted that the albumen of certain reptile eggs, such as those of Testudo horsfieldii, contain strong antibacterial properties which act against a wide spectrum of micro-organisms (Movchan & Gabaeva, 1967), and these properties may well be common in other reptile eggs as well. Broad & Fuller (1974) also found evidence of a variety of substances in the albumen of bird eggs, which either inhibit or kill bacteria and fungi. It would therefore appear that the high influx of water and microbes as a cause of fungal infections can largely be discounted. More feasible alternatives for the high mortality recorded by Tracy (1980) would be the use of fine or flaked grade vermiculite which would have resulted in the suffocation of the eggs, and the disturbance of the containers, which were opened at least twice a week. It is of interest to note, that with all other factors being equal, Tracy's (1980) second highest mortality (50%) was recorded on the driest medium of -590 kPa (V/WR = 1: 0.27) which would make the use of fine grade vermiculite and the disturbance factor common denominators.

There however does not appear to be a relationship between mortality, hatchling mass, and water potential of the incubation medium in hard shelled turtle eggs (Packard *et al.*, 1979). Packard *et al.*, (1979) noted that the hard-shelled eggs of the Softshell turtle, *Trionyx spiniferus*, incubated on substrata lower than -50 kPa (V/WR = 1: 1.36) resulted in a decrease in mass throughout incubation. Although the eggs of lizards which lay hard-shelled eggs (eg. *Pachydactylus* and *Lygodactylus*) are environmentally adapted to incubate in the surroundings in which they are laid, observations during this investigation showed that when removed to a drier environment, they may rapidly dehydrate. Under these circumstances it is therefore

necessary to incubate the eggs, and the above described method of using a high water potential incubation medium appeared to have no adverse affect on either the eggs, the hatching success, or the hatchlings.

The success of incubating reptile eggs may also be dependent on factors such as the composition of the egg shell, which in turn may be influenced by variables such as diet, health, geographic location, seasonality, habitat, climate and microenvironment. Initial clutch masses, clutch size, and relative clutch masses, which may be ascribed to factors such as geographic variation, may well be the result of pheotypic plasticity. Factors such as prey availability and diet have been found to influence phenotypic plasticity in reptile eggs (Seigel & Ford, 1991). However, Seigel & Ford (1991) found that although clutch mass and egg width were significatly higher in higher energy intake groups, as in the superior availability of prey and diet, there was no difference between egg mass, egg length, hatchling mass, and hatchling snoutvent length for either the high or low energy intake groups. The effects of factors influencing phenotypic plasticity on fertility and hatchability do not appear to have been researched in depth.

From the findings of this investigation and other similar studies, it would appear that wetter incubation mediums may have some distinct advantages over drier mediums. It is also indicated from the prevailing evidence that fine grade of flaked vermiculite may result in the suffocation of the eggs, and that the disturbance of the containers during the latter stages of incubation may also be a cause of death in developing embryos. The described method of incubating reptile eggs provided a successful incubating environment for a wide range of reptile eggs, and would appear to go some way in helping to eliminate certain adverse factors apparent in other incubating methods, thereby ensuring a higher hatching success.

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DYSTOCIA (EGG-BINDING) IN REPTILES

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INTRODUCTION

One of the primary aims of herpetology is captive breeding, and certainly the pinnacle of success for any amateur herpetologist must be the successful breeding of his or her animals. Responsible captive breeding is to be applauded in these times of widespread population depletion. Serious reproductive disorders are, fortunately, uncommon; the two main breeding problems encountered being failure to mate successfully and a failure to produce viable eggs or young. Difficulty in producing eggs or young at the end of the gestation period is called dystocia, and it is most prominent in the oviparous (egg laying) species. Dystocia can lead to the death both of the unborn young and of the female and therefore must be treated as an emergency with professional veterinary advice sought as soon as possible.

DYSTOCIA

The phenomenon of dystocia is not uncommon in the polyparturient domestic animals, the dog, cat and pig. In these species dystocia can be caused by an obstruction in the genital tract or by uterine inertia (weak uterine contractions). Obstructive dystocia is often seen when only a few, large offspring are present and resolution usually requires surgery if the obstruction cannot be overcome manually. Dystocia due to uterine inertia can be due to a host of factors but medical treatment with the parenteral administration of oxytocin and calcium will often cause expulsion of the offspring by increasing the strength and rate of uterine contractions.

Dystocia in reptiles can also be obstructive or muscular in aetiology. If, after careful examination, the oviduct and cloaca are free and there seems to be no obstruction, an intramuscular injection of oxytocin (2 units per 100g body weight) will often result in egg laying or livebirth. The molecular structure of the oxytocin is well conserved throughout the animal kingdom and therefore the use of mammalian oxytocin does not seem to pose problems in reptiles. Priming the uterus with calcium prior to oxytocin administration is neither necessary or recommended in reptiles (Frye, 1991). In cases where the cloaca or oviduct is damaged or an obstruction is feared, medical treatment is contraindicated because rupture of the oviduct and consequent peritonitis may occur after the administration of oxytocin. In these cases surgery is the only course of action.

Dystocia occurs with some regularity in captive reptile populations but it is not a random event, indeed there are several well documented pre-disposing factors that increase the likelihood of egg-binding (Frye, 1991; Ross and Marzec, 1990).

- 1. An incorrect breeding programme may fail adequately to stimulate the reproductive system of the female. Improper cooling, improper diurnal temperature variation and abnormal photoperiod may be involved.
- 2. Suitable nest sites are important for many species and failure to provide a suitable nesting place may delay or prevent egg laying leading to dystocia. In general; many species of chelonia and terrestrial lizards dig holes and deposit their eggs in a warm, moist substrate, terrapins prefer a fairly dry sandy land area while snakes select the seclusion of a humid hide box. These are generalisations and individual species may have different preferences from the expected norm.
- 3. Maintaining large numbers of reptiles in a confined vivarium can result in competition for the limited egg laying sites. This is an unfortunate but increasing trend that can result in one dominant female preventing others from accessing the laying site, thereby resulting in a delay in egg deposition and possible egg-binding in the sub-ordinate females.
- 4. Stress caused by disturbance of the environment such as failure to provide seclusion or the constant interference by the owner, is unfortunately too commonly seen.
- 5. The transport of pregnant females can result in stress and consequently dystocia.
- 6. An imbalance of calcium and phosphorous may prevent proper egg calcification or muscular contraction of the oviduct that is essential for the expulsion of eggs. Therefore, the provision of sufficient vitamins and minerals in the diet is important.
- 7. Concurrent infection of the female's reproductive tract, especially the oviduct, can also complicate egg laying or livebirth.
- 8. Inbreeding using closely related individuals may cause genetic homogeneity and inbreeding depression by the expression of deleterious or lethal genes, thereby increasing foetal death, egg infertility and dystocia (Ross and Marzec, 1990).

CASE REPORT – DYSTOCIA IN AN AFRICAN ROCK PYTHON

A female African rock python (Python sebae), measuring approximately 3m in length, was presented to the Royal Veterinary College because of dystocia. The female had previously been kept with a male but no purposeful attempt at breeding had been attempted: no changes in the diurnal temperature viariation or photoperiod had been employed. The female had become restless and laid 12 eggs before ceasing to lay the remainder of her clutch. A partial prolapse of cloacal tissue or oviduct through the cloacal opening also became evident.

On clincial examination, the prolapsed cloacal tissue had become necrotic and devitalised and several eggs were palpable within the lower third of the body. From the female's aggressive demeanour and perpetual restlessness within the vivarium it appeared that she was in some discomfort. The administration of oxytocin was contraindicated in this case as the damage to the cloaca presented an obstruction to normal laying. Indeed, stimulation of the oviduct by oxytocin, if attempted, might have caused rupture of the oviduct and consequently peritonitis and death. It was therefore decided that surgery was the only course of action, and the snake was promptly admitted for coeliotomy (Frye, 1991; Lawton and Stoakes, 1992).

DIAGNOSIS

Confirmation that the snake was indeed suffering from egg-binding was achieved by radiography. To facilitate handling, and as a preparation for surgery, the snake was heavily sedated using an intramuscular injection of ketamine hydrochloride. The radiographs clearly depicted many retained eggs within the final third of the snake, and therefore the snake was immediately prepared for surgery.

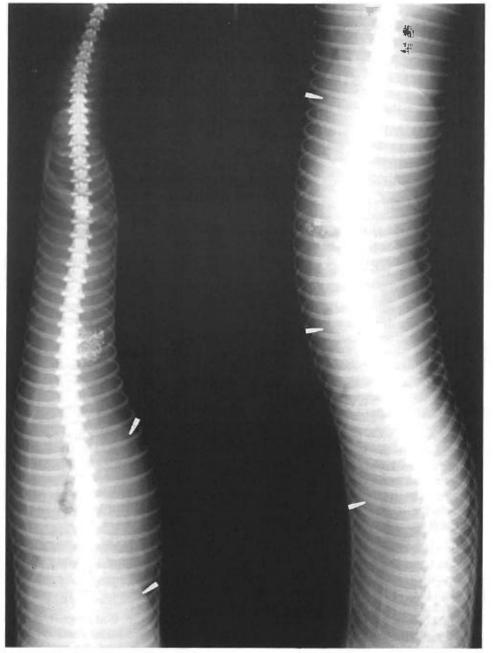


Plate 1: This radiograph was taken prior to surgery. The presence of the retained eggs (indicated by the arrows) clearly confirmed the diagnosis of dystocia in this Rock Python (*Python sebae*).

SURGERY

After transportation to the sterile surgical theatre, the reptile was placed on a heating pad and intubated. Intubation involves passing a endotracheal tube down into the trachea so that gaseous anaesthetic and oxygen can be delivered directly into the lung. Since there are no standard endotracheal tubes for snakes a shortened dog urinary chatheter was employed.

Anaesthesia was maintained with a supply of oxygen and isoflurane. The snake's heart rate was monitored by electrocardiography, while the depth of anaesthesia was judged on the basis of tongue and ventral muscle reflexes. Many reptiles tend to stop breathing during surgery and therefore it was necessary to artificially ventilate during the two hour operation.

An initial longitudinal incision was made along the ventral aspect of the body cranial to the cloaca. Careful dissection through the skin, connective tissues and musculature revealed the oviduct which was then incised and several eggs were removed. The large number of retained eggs required a second incision cranially and the removal of several more eggs. In total, 12 infertile eggs were removed. The thin-walled oviduct, muscles and skin were sutured, and the damaged cloaca and prolapse were resected, sutured and cleaned. The anaesthetic agent was withdrawn but the oxygen supply was maintained until the snake was breathing voluntarily.

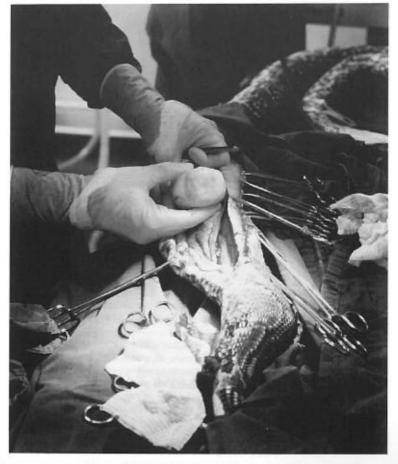


Plate 2: A large ventral midline incision was made in the lower third of the snake and several eggs were removed. A second incision had to be made more cranially in order to remove all the eggs.



Plate 3: Seven of the retained eggs. All the eggs that were removed were infertile as determined by *post-mortem* examination.

Potentiated sulphonamide was given by intramsucular injection to provide antibiotic cover post-operatively and an adhesive bandage was applied to the wound to provide some degree of protection. The snake started regaining her righting reflexes and consciousness almost immediately after the gaseous anaesthetic was stopped, and became active and pugnacious before being returned to her hospital vivarium.

The snake returned home and made a full and uneventful recovery.

CONCLUSIONS

Within the herpetological hobby as a whole there seems to be the view that veterinary surgeons are unwilling or unable to offer the same level of service for reptiles as they do for our more domesticated animals. We hope that this short report of a relatively simple operation will lay these reservations to rest and persuade all amateur herpetologists to seek professional veterinary advice when disease and illness threaten their reptiles.

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FURTHER OBSERVATIONS ON SPAWNING PARSLEY FROGS SIMON HARTLEY 8 Harnorlen Road, Peverell, Plymouth, Devon, PL2 3NU

INTRODUCTION

Regular readers of the Bulletin may recall my article in the Spring 1990 issue (No. 31), in which I described successful captive spawnings of the Parsley Frog, *Pelodytes punctatus*, during 1988 and 1989. As a result of breeding this species since then, a number of noteworthy additions can now be made to those earlier observations. 1990 proved to be by far the most productive and interesting of subsequent years, so this will be covered in detail, with reference to the others being made only where necessary. In view of the considerable depth given to describing the various stages of my spawning method in the above mentioned article, excessive repetition will be avoided in the course of the following, so I would refer those requiring greater detail here to that earlier publication.

1989 METHOD SUCCESSFULLY REPEATED

For 1990, the main task was initially to discover whether the Parsley Frog breeding method arrived at in 1989 was a reliable one. As then, pairs of frogs were accommodated in 14" x 8" x 8" indoor aquaria which were furnished with an island built up from tiles and rocks from which clumps of grass hung into the water to a depth of 5-6" (12-15cm). The tanks were kept at room temperature, 50-68°F(10-19°C), and once attempted/actual amplexus was observed, frequent $\frac{1}{2} - \frac{1}{4}$ water changes were made. Frogs came into condition as previously recounted, (Hartley, 1990), and spawnings were duly obtained from all but one of five mature females available. Spawning temperatures were between 54 and 65°F (12-18°C), mostly in the upper 50s, and only once was strong sunlight available, these features conforming to the pattern noted the previous year. With varying degrees of success, this was also the case subsequently, the only noteable exception being one very late spawning obtained in mid-May (17th) last year (1992).

The experiment to discover if significantly more green individuals could be produced by raising tadpoles on green algae than on other foods was, however, inconclusive, with no real difference between each group, although those fed mainly on boiled lettuce, which had not been used previously, have been found both in 1990 and since to grow faster than ones raised on fish flakes. This year, 1993, a greater number of green froglets have so far emerged from tadpoles housed in a brightly lit outdoor shed than those kept indoors under less well lit conditions most of which have been brown, but again the difference is unlikely to be significant.

EARLY SPAWNINGS & INFERTILITY

Very early spawnings were one of the novel features of the 1990 season. At least two of the males were observed to have fully-developed nuptial pads, and most of the females appeared to be ripe with eggs, at the beginning of January, a month earlier than was the case in '89, though no typical breeding calls had yet been heared. On 2nd January a male and two females were moved to a breeding aquarium, and amplexus was observed several times over the next 6 days. No further progress having be made, another female was introduced. 7 days later the least co-operative female was removed, but the pattern of periodic amplexus continued until the male stayed with the same female from 22nd to 26th, when breeding calls were heard from another male in the terrestrial vivarium. This one was added to the breeding tank, and both went into amplexus intermittently over the next week. On 2nd February, the pair introduced a month earlier finally spawned (producing c.700 eggs) at 56°F (c.13°C), and the other pair followed suit on 8th, (c.1,000 eggs) at 57-60°F (13-16°C). These spawnings were 5-6 weeks earlier than any of my own, or so far as I am aware, those of anyone else, in 1989.

However, the negative side of this apparent success was that within 2-3 days it was clear that each batch of eggs was totally infertile – something which had never occurred in the previous two years. Conditions were identical to those under which spawn had been laid and hatched in '89, (Hartley, 1990), so this did not appear to be the problem, and the frogs themselves were perfectly healthy, otherwise they would not have spawned at all. Was there a possibility that one or both sexes were not quite in full breeding condition after all, and had been stimulated to do so too early?

A possible answer was provided at the end of the month, when, using two males discovered by Howard Turner in his unheated greenhouse, and two of the non-spawning females, further spawnings were obtained on 27th (c.800 eggs) at $57^{\circ}F$ ($13^{\circ}C$), and 28th (c.50 eggs) at $57-60^{\circ}F$ ($13-16^{\circ}C$). These proved to be fertile, and developed normally, perhaps supporting the theory that the earlier individuals had not been in peak condition, or alternatively that the new males were more fertile than the others because they had come from a more natural environment. The certainty was that they were still 13 and 14 days respectively in advance of the first spawnings of 1989. However, the only subsequent February batch was produced in 1991 (on 27th), when approximately 60% of the eggs were fertile.

For reasons involving either insufficient ocnditioning of the adult frogs, or some other factor related to their captive environment, spawings in which both the older and maturing young breeders showed an increasing rate of infertility, irrespective of when the eggs were laid, in 1991, and almost total infertility in 1992. This trend has also been noted by Howard Turner and Charles Snell, both of whose *Pelodytes* are maintained in different captive set-ups to my own. Any suggestions from readers as to the possible causes of the infertility would be very welcome.

SECOND SPAWNINGS BY FEMALES

Finally to the most interesting, unexpected and perhaps the most significant element of the 1990 breeding season. As with all other anurans, males of this species will mate with as many females as they can find throughout the course of the spawning period. However, there is to my knowledge no record of female Parsley Frogs producing more than a single large batch of eggs during that time, and even the earliest spawners in 1989 had ever looked like doing anything else.

By the beginning of April 1990, the females which had spawned in early February had put on sufficient weight to suggest that they might again be carrying eggs. All the males were still in breeding condition, so I decided to set them up to see if anyting would happen. One of the males involved with the first spawnings, and one used in those at the end of February, were placed with the two females above on 4th. The usual behaviour pattern followed, with the result that spawning indeed took place on 6th (c.590 eggs), at 54°F (14°C), then 13th (c.870 eggs) at 56°F (13°C), and finally a second batch was produced by the female which first spawned on 28th February (c.350 eggs) at 60-65°F (21-25°C), on the latter occasion using the other male which bred at the beginning of the season. In each case, there was a gap of 63, 64 and 47 days respectively between the first and second spawnings. The vast majority of each batch of eggs hatched normally, despite the involvement of those frogs whose spawnings were infertile. Perhaps this suggests that incomplete development of sperm and/or eggs was indeed to blame for the previous failures that year.

The above clearly shows that Parsleys are well capable of producing more than one batch of eggs in the course of the same breeding season, and if given sufficient time, might also do so in the wild state. According to Salvador (1985), they breed between November and March in southern Iberia, which is quite long enough for this to occur if conditions remain suitable, but he makes no specific reference to females spawning more than once. As the last of the 'second spawners' proved, it is also possible for two fertile batches to be produced. At least the 7 weeks required by that individual may be necessary for egg stocks to be replaced sufficiently to breed again, which partly explains why there were no second batches in '89. It should be noted that all three females received the same amount of food in that period as they had previously, so a significant increase in feeding is not necessary to bring them back into condition. The approximate number of eggs in the second batches were certainly not vastly inferior to those in the first ones. Note also that of the four females successfully spawned in 1990, only one failed to produce a second batch, further suggesting that the second spawnings were not just a freakish event.

In subsequent years, however, despite numerous attempts, none of my female *Pelodytes* have produced a second batch of eggs, largely because they refused to breed as early as in either 1990 or '91, thus (probably) leaving insufficient time to do so before the breeding males went out of condition, and in 1992 only one female spawned at all. This may of course be connected with the infertility problem, but it may also be that a combination of exceptionally good conditioning and some other factor/ factors were responsible for the successes of '90.

SUMMARY

To conclude, the main features of the 1990 Parsley Frog breeding season can be outlined as follows.

- (1) The spawning method found to be successful in 1989 proved to be effective. The only difference in '90 was that spawnings took place at slightly lower temperatures, and none occurred at anywhere near the high 70s°F(20s°C) suggested as the upper limit in my previous article (Hartley, 1990). Spawnings in following years were produced under the same conditions, so it can be concluded that temperatures in the mid-upper 60s°F(c.20°C) are sufficient.
- (2) Successful (i.e. fertile) spawnings can be obtained earlier in the year than previously described, although the evidence does suggest the need for animals to be in absolute peak condition.
- (3) Parsleys are obviously able to produce a second batch of eggs during a spawning period of two-two and a half months, whatever the most crucial factor involved here, but this is still a discovery of considerable potential value both to those wishing to breed them and also those simply interested in obtaining additional

knowledge of their behaviour. Apart from not having been recorded from other captive collections, this seems to be another area in which sufficient information on the reproductive activity of wild populations is lacking, and would certainly be worth further investigation.

Since that very successful year, there has been a gradual decline in the fertility of spawn from both older and younger frogs, and also an increasing reluctance on the part of females especially to spawn. Fresh blood has been added to the stock over this period, so inbreeding can probably be ruled out as a main contributary cause. Experimental alternations in general husbandry will possibly prove to be the answer.

ACKNOWLEDGEMENTS

My thanks are due to Howard Turner for the use of his male frogs in 1990, tadpoles in 1993, and for advice etc. throughout, and to Charles Snell for information provided.

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REVIEW

THE HERPETOFAUNA WORKERS GUIDE 1993

Compiled by Andrew Arnon and Catherine Becket on behalf of Herpetofauna Conservation International Ltd. ISBN-0-9521106-0-1

Copies of this guide are available free of charge from HCI Ltd, Triton House, Bramfield, Halesworth, Suffolk IP19 9AE, enclosing an A5 size SAE.

This guide consists of a directory of information and resources for anyone interested in the conservation of amphibians and reptiles and their habitats in Britain. Part funded by grant-aid from the Department of the Environment, the directory is based on the format of the Bat Worker's Guide.

The guide contains a list of local Amphibian and Reptile Groups, with the names and addresses of regional representatives. Also information on how to set up your own local group and how the Herpetofauna Groups of Britain network help and advise is outlined.

There are also sections on all the major voluntary organisations involved in herpetofauna conservation and the Statutory bodies, such as English Nature, Countryside Commission for Wales, Scottish Natural Heritage, with regional contacts.

Wildlife Legislation in the UK is explained in terms of reptile and amphibian conservation issues and includes details of the common scoring system for the selection of amphibian asemblage SSSI's and procedures, such as licensing, for working with herpetofauna. A county by county list of toad crossing sites and information on helping toads across roads is also included.

Information on current conservation projects and the organisations involved, with sources of grant aid is listed, together with a list of references on general issues, identification, species distribution, survey and practical management.

The first edition of the Herpetofauna Worker's Guide is a handy reference for anyone interested in the conservation of our native reptiles and amphibians. The regional and national contact lists provide the opportunity to get involved in various projects and a resource such as this guide has been long overdue.

Jan Clemons

NOTES ON THE REDISCOVERY AND CONGENERIC ASSOCIATIONS OF THE PFEFFER'S CHAMELEON CHAMAELEO PFEFFERI (TORNIER, 1900) (SAURIA: CHAMAELEONIDAE) WITH A BRIEF DESCRIPTION OF THE HITHERTO UNKNOWN FEMALE OF THE SPECIES

CHRIS WILD

38 Main Street, Normanton-On-Soar, Loughborough, Leics, LE12 5HB, U.K.

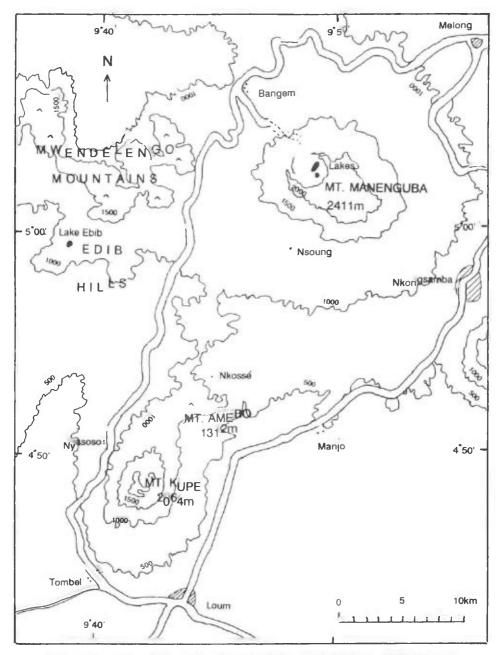
INTRODUCTION

In 1899 Von Gustav Tornier, a German taxonomist, wrote; "More and more very strangely formed chameleons from tropical Africa are becoming known and this chameleon is amongst them". The specimen in question, a male, was collected by M. Conradt from the Kupe Mountain forest situated on the former East/West Cameroon border. Tornier named the new species Chamaeleo pfefferi in honour of Dr. Pfeffer of the Hamburg Natural History Museum. Since that time the Pfeffer's Chameleon remained elusive to the many herpetologists who have collected reptiles from the relatively small (21km²) forest of Mt. Kupe. Occasional references to the species have been made, invariably based on Tornier's original description (1900), or from observations of the type specimen itself.

During recent field studies I had the pleasure of collecting three specimens of this little known species from the Kupe and Manenguba mountains of western Cameroon including two examples of the hitherto unknown female of the species. The first specimen, a gravid female collected in May 1990, was discovered two metres from the ground basking in direct sunlight on secondary growth near a fallen tree in primary forest. The site was located at 1320m on the western slopes of Mt. Kupe above Nyasoso (grid ref: 4^{0} 50'N, 9^{0} 40'E). This specimen remained unidentified until in April 1992 I returned to the neighbouring Mt. Manenguba where a male and a second female, also gravid, were collected from village farms at Nsoung situated at 1330m on southern slopes of the mountain (grid ref: 4^{0} 59'N, 0^{0} 49'E). The male was recovered from four metres up on the outer foliage of a Cola tree (Cola nucifera). Data on the perching habit of the second female was not recorded as it was found by a resident of Nsoung village.

NOTE ON THE TYPE LOCALITY

The type locality given by Tornier 'Nyassosso, Nkossogebirge, Kamerun', is worthy of some discussion as it is confusing to both cartographers and herpetologists alike. An extensive review of maps of the region has not revealed any mountain by the name of 'Nkosso' or a second Cameroon village by the name of Nyasoso (=Nyassoso) (grid ref: 4° 50' N, 9° 40' E). The US Gazeeter (Cameroon) lists a mountain southeast of Bamenda by the name 'Nkossa' (grid ref: 5° 48' N, 10° 20' E), but this peak is an improbable location for the Type locality as there is no record of a village called 'Nyassosso' in the area. Perret (1957) however, referred to the type locality as: "Chamaeleo pfefferi Tornier (Zool. Anz. 23: 21-23. 1990), dont le type et unique exemplaire récolté provient de Nyassôssô, mont Nkossoo, partie sud du massif du Manenguba, à la frontière des deux Cameroons." This description closely corresponds with a 1312m peak formerly known as Mount Amebo rising from the 1000m intermontane ridge 2km or so north of Kupe (grid ref: 4° 52' N, 9° 46' E; map ref: Carte de L' A.E.F. et du Cameroun AU. 200,000e. 1958, Douala, FEUILLE NB-32-IV), on the north-eastern flanks of which lies the village of 'Nkossé' (= Nkosso?) situated at 800m (grid ref: 4° 52' N, 9° 47' E). Since modern maps no longer include Amebo or Nkosso as names for distinct peaks and both Nyasoso and Nkossé lie in the premontane zone of Mt. Kupe, it is therefore reasonable to regard the type locality as follows: 'Nyasoso, Mount Kupe, Cameroon'.



MAP OF THE KUPE AND MANENGUBA MOUNTAINS, CAMEROON.

BRIEF DESCRIPTION OF THE FEMALE HABITUS

SPECIMEN: BMNH 1992, 279.

LOCALITY: Nsoung 1330m, Mount Manenguba, Littoral Province, Cameroon.

DESCRIPTION: A small-medium sized chameleon, the female being only marginally smaller than the male. Casque moderately elevated (19%). Parietal crest slightly indicated posteriorly. Lateral and orbital crests prominent with denticulate outline. Temporal crest present and partially fused with lateral crest. Gular crest prominent, with three (2nd, 3rd and 4th) large, narrow spike-like conical scales preceeded by one, and followed by two, somewhat smaller projections, ending below angle of the mouth. Posterior to forelimbs ventral crest absent. Dorsal crest absent but dorsum moderately keeled to form distinct ridge-like formation. Tail marginally shorter than body length (snout-vent).

SQUAMATION: HEAD; dorsal/parietal area comprised of polygonal and tubercular scales. Scia dentata present. Auditory area heterogeneous with granular and tubercular scales. BODY; heterogeneous, granular, irregular in shape and size except for the largest tubercular, plate like scales which are distributed at equidistant intervals and diagonally aligned across the dorso-lateral aspect of the body and limbs in conjunction with lateral interstitial veining sloping downwards posteriorly, further transversed by vertical and diagonal veining.

MEASUREMENTS:	Total length	152mm
	Head length (snout-casque apex)	24mm
	Body length (snout-vent)	80mm
	Tail length	72mm
	Mouth length (angle-snout)	15mm
	Casque apex – nostril	20mm
	Casque apex – angle of mouth	16mm
	Casque apex – occiput	3mm
	Casque elevation	19%

FEMALE LIVERY: scalation various shades of green offset by blood red interstitial skin, lateral and temporal crests red, gular crest pale green, whitish transverse dorsolateral band mid-body sloping posteriorly, paler ventrally, claws brownish, horizontal eyestripe indicated. When roosting, scalation very bright yellow vividly offset by interstitial blood red veining.

MALE LIVERY: scalation generally dull to bright green/turquoise offset by prominent blood red interstitial veining. In courtship colours more vivid – posterior aspect of body, hindlimbs and tail bright yellowish green, anterior aspect of body, upper forelimbs and head bright turquoise. Horizontal eyestripe present, whitish transverse dorsolateral band mid-body sloping posteriorly.

Both sexes of the species may be readily diagnosed from other Cameroonian congenerics as it is the only regional form known to possess a very pronounced gular crest in the absence of a ventral crest posterior to the forelimbs.

CONGENERIC ASSOCIATIONS

It is well known that the Biafran Mountains of Cameroon, Nigeria and Equatorial Guinea have proven to be a fertile environment for speciation and endemism in many floral and faunal groups (Kingdon, 1990). In West Africa inner divergence within the genus *Chamaeleo* is nowhere expressed more highly than in the montane forests of Mts. Kupe, Manenguba and adjacent Bakossi Highlands where at least two taxa, *C. quadricornis quadricornis* and *C. pfefferi*, are thought to be endemic.



Plate 1. Male Pfeffer's Chameleon, *Chamaeleo pfefferi*, in courtship livery. 1330 metres Nsoung village, Mt. Manenguba, Cameroon.



Plate 2. Gravid female Pfeffer's Chameleon, Chamaeleo pfefferi, 1320 metres submontane forest, western slopes of Mt. Kupe, Cameroon.



Plate 3. Female Cameroon Mountain Chameleon, *Chamaeleo montium*, 1060 metres, riparian vegetation of Mbwe River, northwest of Bangem, Mwenzekong Mts. (= Bakossi Highlands).



Plate 4. Male Wiedersheim's Chameleon, Chamaeleo wiedersheimi, in courtship livery, 1800 metres, shores of Lac Femme, Mt.Manenguba, Cameroon.

Mt. Manenguba is a largely deforested mountain due to the annual slash and burn practice of the resident Bororo herdsmen. Consequently only remnant gallery forest remains within the mountain's great caldera along with a few square kilometres of montane forest on the upper peaks and on the south-eastern slopes above Nsoung village. At Nsoung and on local farmbush, *C. pfefferi* was found to occur sympatrically with numerous Four-Horned Chameleons, *C. quadricornis quadricornis*, a high elevation montane forest/savannah species, and the equally locally abundant Wiedersheim's Chamaeleon, *C. wiedersheimi*, a high elevation montane savannah/ grassland species which has invaded the Nsoung farmbush from the grassland of the mountain's caldera at 1800m. The Mountain Chameleon, *C. montium*, a premontane and lower submontane forest species was found parapatrically to the three aforementioned congenerics just below Nsoung at 1070m and in the forest/ farm mosaic throughout the 16km Kupe-Manenguba intermontane ridge which lies chiefly above 1000m.

Within the closed canopy forest of Mt. Kupe, C. pfefferi was only found sympatrically with the euryzonal Western Pygmy Chameleon, Rhampholeon spectrum, a common and widespread semi-arboreal species of the Western Equatorial Forests. In addition, C.q. quadricornis has recently been recorded from the Kupe mountain forest (1550m C. Bowden pers. comm. and 1940m, C.J. Wild unpubl. data) although an undescribed four-horned chameleon from Essossong 1060m, which lies in the northern pre/submontane forest of the mountain, was previously known from the Hamburg Zoological Museum (Böhme and Klaver, 1981).

C. pfefferi, a remnant paleo-endemic is closely allied to C. montium, an expansive neo-endemic, to which it succeeds in the strata of vertical distribution of montane chameleons found on Mt. Kupe and Manenguba. The comparatively well known C. montium occupies the intermontane hill forest, premontane and lower submontane zones from 550-1170m in this region, although it has been found as high as 1250m in the elfin thicket of the summit and ridge crests near Lake Edib in the Bakossi Mountains (pers. obs.). Furthermore, the regional topography suggests that both C. pfefferi and C. quadricornis probably occur in the adjacent Mwendelengo and Mwenzekong Mts. of the Bakossi Highlands which are connected to Mt. Manenguba by intermontane plateau-like ridges rising in excess of 1300m and 1200m respectively.

Like most montane chameleons, *C. pfefferi* displays a remarkable array of ornamentation and livery (such 'ornamentation' in the Biafran chameleons being; horns, rostral appendages, dorsal, gular and ventral crests, sails, squamation and livery) which generally is far greater than that exhibited by their lowland congenerics. These adaptations are thought to be due to the high species density of the chameleons occurring on these mountains which has led to the manifestation of such bizarre appearances presumably to play the role of courtship isolating mechanisms as discussed by Rand (1967).

In reference to East African montane chameleons, Rand (1967) suggested that such ornamentation served the function of species and partner recognition and that this was analogous to the bright feathers and songs of birds, to the calls of frogs, and to the strikingly coloured gular fans in iguanid lizards such as *Anolis*. Rand's hypothesis being derived not only from the fact that these chameleons exhibit strong sexual dimorphism, but also because no two sympatric taxa share the same assemblage of ornamentation or livery – a situation also common to the six endemic montane chameleon species of Biafra. One such character unique to the male *C. pfefferi* is that of the greatly enlarged canthus rostrales which meet above the nostrils to bridge the two short diverging horns in a laterally flattened and concave formation.

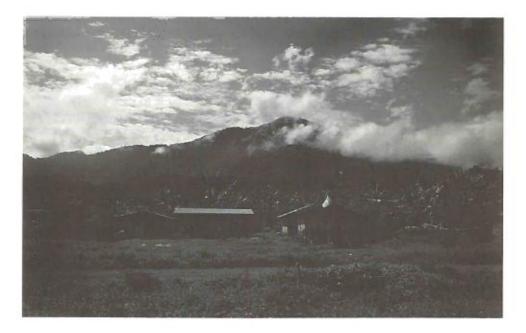


Plate 5. Foreground: Nyasoso village and farms, 820 metres, habitat for C. montium, C. cristatus, and Rhampholeon spectrum.

Background: Mt. Kupe Forest, 2064 metres, Cameroon, habitat for C. montium, C. pfefferi, C. quadricornis, and Rhampholeon spectrum.



Plate 6. Lac Femme, 1800 metres, Mt. Manenguba, Cameroon. Ungrazed patches of grass/ scrub are habitat for C. *wiedersheimi*. Remnant montane and gallery forest habitat for C. *q. quadricornis*. In his study of Malagasy chameleons, Parcher (1974) concluded that the ability of chromatic change served to communicate, noticeably in courtship, between individuals and taxa rather than the popular notion of the phenomenon being that of cryptic disguise. However the pygmy chameleons of the genus *Rhampholeon* have a more limited ability to change colour and are obviously pro-cryptic, (Cott, 1957).

The chameleons of the Cameroon Highlands are not yet fully known. Numerous mountains of high elevation remain where the reptile fauna is to date poorly known. In a region where so many allopatric populations exist in isolated mountain refuges with marked changes in environmental conditions separating them, there is strong biogeographical evidence to suggest that new taxa from this genus are still to be found in these mountains.

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