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Full Length Research Paper

Reproductive characterization of three species of Giant African land snails (GALs) in captivity

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Duration of reproductive activities (courtship latency, courtship and mating) were studied in three (3) species of Giant African land snails (GALs) namely, *Achatina fulica*, *Archachatina marginata* and *Achatina achatina* using mature snails gathered from rural Nigeria. The snails were housed singly for 90 days to acclimatize; shed already fertilized eggs from previous mating and develop strong desire to mate. Thereafter, monocultures of each species were formed namely, *A. fulica* x *A. fulica* (F x F); *A. marginata* x *A. marginata* (M x M) and *A. achatina* x *A. achatina* (A x A) for pre-mating trials. These were observed for one week and any two snails that showed consistent association for more than 10 min were isolated and kept together as potential mating pairs. Parameters measured were duration of courtship latency (DCL), duration of courtship (DC), duration of mating (DM), duration from mating to oviposition (DMO), egg weight (EW), egg length (EL) and weight of hatchlings (WH). Number of mated pairs and ovipositors per species were noted and mating propensity was calculated as the percentage of tested snails that mated in each species. Result shows significant (P ≤ 0.05) differences between snail species in all traits studied.

Key words: Land snails, courtship, copulatory duration, tentacular contact, monocultures.

INTRODUCTION

Gastropod reproductive behaviour is as varied as the animals themselves (Lind, 1976; Chase, 2007). Hermaphoditism is universal among pulmonate gastropods (Thompson and Cheney, 1996; Ejidike, 2002; Chase, 2007), almost so in opisthobranchs but rare in other taxa (especially marine snails) (Chase, 2007). Hermaphoditism refers to the presence of functional male and female reproductive systems in the same individual (Chase, 2007). In some species, individuals play specific (male or female) role during mating. In some (example, *Lymnaea stagnalis* and *Helix pomatia*), reproduction is reciprocal, that is, individuals play single roles per mating but roles can be switched in subsequent matings (Chase, 2007), while in some other species (example, *Cornu aspersus*), simultaneous reciprocal mating occur with both members of the pair acting simultaneously as male and female (Thompson and Cheney, 1996; Chase, 2007; Skelley et al., 2010). In this case, both animals receive sperm at every mating. In species that share roles, the smaller member of a mating pair often assume the male role (Angeloni et al., 2002; Dillen et al., 2010). These authors explained that this is because of the need for each snail to maximize reproductive fitness.

Snails find mates by chemical senses (olfaction and contact sensation) (Zeeck et al., 1988; Frey, et al., 1998; Horth, 2007). Terrestrial (land) snails locate potential mates by sensing cues in mucus trails then following the trails to find the source (Tekeichi et al., 2007). Contact chemo sensation, as well as tactile stimulation, is probably important for courtship interaction in all species (Weinberg et al., 1990; Saxena, 2009).

Reproduction is an essential aspect of the life cycle of every organism. For the edible land snails that use internal fertilization, courtship usually precedes mating. Snails depend on the stimuli delivered by courtship behaviour and copulation to initiate and co-ordinate reproductive function (Arnqvist and Nilsson, 2000; Neiman, 2004, 2006).

In the humid tropics, Giant African land snails in the wild lay eggs about two to three times per season (Cobbinah et al., 2008). Egg laying, hatching and development in the wild are thus seasonally influenced by

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Table 1. Percentage composition of feed fed to the experimental snails (24% CP).

Ingredient	Composition (%)
Maize	31.75
Soya bean meal	9.60
Fish meal	9.60
Groundnut cake	12.80
Vit. Premix	0.50
Bone meal	4.00
Wheat offal	31.75
Total	100.00

temperature, humidity and moisture content of the soil (Ebenso, 2006; Ejidike et al., 2002). In recent times, the volume of snail harvest from the wild has dwindled due to the impact of man's activities, such as deforestation, pesticide use, slash and burn agriculture and more intensive snail hunting (Raut and Barker, 2002). The demand for snail meat has also increased tremendously over the years so that at present, demand outstrips supply (Murphy, 2001, Ebenso, 2003, Paoletti, 2005). As a result of the economic opportunities offered by snail farming and marketing, there is a resurgence of interest in intensive culturing of edible land snails for domestic and foreign markets (Raut and Barker, 2002; Moyin-Jesu and Kemi, 2008; Cobbinah et al., 2008).

At present, little is known about the reproductive behaviour of most Giant African land snails in captivity. This research was therefore undertaken to study the courtship and copulatory duration of three (3) species of Giant African land snails (GALs) in captivity to further characterize them and for improved reproductive performance in intensive holdings.

MATERIALS AND METHODS

This study was carried out in the Snailery Unit of the Department of Animal Science, University of Nigeria, Nsukka between February and August, 2008. A total of 216 adult Giant African land snails of unknown reproductive history gathered from rural Nigeria were used for the experiment. The snails were made up of (3) species namely, *Achatina fulica, Archachatina marginata and Achatina achatina.* On arrival, the snails were washed by spraying and housed in individual plastic baskets containing good mixture of garden soil. They were fed on a mixture of compounded feed (Table 1) and vegetable food materials (leaves of *Ipomea batata*, fruits, peels and leaves of *Carica papaya*, Musa species, etc).

The snails were isolated individually for 90 days, from arrival to adapt and shed eggs it might have carried from the wild as well as develop strong desire to mate (Baur and Baur, 1992; Dillen et al., 2008, 2010).

Mating arrangement

Pre-mating trials were initiated 90 days after the snails were isolated. Monocultures of each species (3 per species) made up of 24 snails each were observed for one week (7 days) for reproductive behaviour and to select pairs of snails for the mating trials. Two snails that showed close association for at least 10 min were isolated and housed together to form a potential mating pair. Twelve (12) potential mating pairs were established for *A. fulica,* 13 pairs for *A. marginata* and 11 pairs for *A. achatina.* Each pair's behaviour was observed continuously for two (2) weeks until the snails became unreceptive, achieved copulation or were separated at the end of 2 weeks.

Duration of reproductive activities was measured by means of a digital stop watch and the snails were observed from 6.00 pm to 7.00 am daily throughout the experiment.

Egg production and hatching

If mating occurred in a pair, they were allowed two (2) weeks to oviposit (Baur and Baur, 1992). If oviposition occurred, eggs were collected and transferred to another medium (hatchery) to develop and hatch.

Data collection and analysis

Data on reproductive behaviour included duration of courtship latency (duration of initial tentacular contact); duration of courtship (time period from oral contact to insertion of genital atria); duration of mating (time period from insertion of genital atria to separation of snails) and time between mating and oviposition. The duration of reproductive activity (DRA) per snail species was the sum of their respective time duration for courtship and mating (DRA = DC + DM). Egg weight, egg length and weight of hatchlings for each snail species were measured. Data were analyzed using the ANOVA option of SPSS (2001) computer programme to test for effect of species (genotype) on the parameters measured. The statistical model was:

$$X_{ij} = \mu + G_j + e_{ij}$$

Where, X_{ij} is the observation on the jth snail of the ith genotype; μ is the overall mean; G_i is the ith genotype (species) and e_{ij} is the residual.

Significant means were separated using the Duncan option of SPSS. Only data from trial pairs that proceeded from courtship to mating were included in the analysis.

RESULTS AND DISCUSSION

Table 2 presents the mating types, observed mating pairs, mating propensity and number of ovipositors per snail species. The table showed that twelve (12) pairs (24 snails) were tested for *A. fulica*, while only five (5) pairs (10 snails or 41.7%) mated. For *A. marginata*, three (3) pairs mating was observed out of thirteen (13) pairs (26 snails) tried, representing 23.1%, whereas for *A. achatina*, three (3) pairs mating was observed out of 11 pairs (22 snails) tested, representing approximately 27%. Number of ovipositors per snail species that mated were 4 (40%) for *A. fulica* and 2 (33%) each for *A. marginata* and *A. achatina*. The 41.7, 23.1 and 27% mating percentages (or mating propensities) obtained for *A. fulica*, *A. marginata* and *A. achatina*, respectively, are higher than the 10% reported for *A. fulica* by Tomiyama

Table 2. Mating types, observed mating pairs, mating propensity and number of ovipositors per snail species.

Mating type (type)	Potential mating pair (number)	Observed mating pair (number)	Mating propensity (%)	Ovipositor (number)	Ovipositor as percentage of number mated
F x F	12	5	41.7[a]	4[a]	40[a]
M x M	13	3	23.1[b]	2[b]	33[b]
A x A	11	3	27.0[b]	2[b]	33[b]

[a,b]: Means on the same column with different superscripts are significantly (P≤0.05) different; F X F = *A. fulica* pairs; M X M = *A. marginata* pairs; A X A = *A. achatina* pairs. Mating propensity = ratio of snails that mated to total number of snails that were tested in each species.

(1994) probably as a result of isolating the snails for 90 days prior to the experiment. Dillen et al. (2010) reported that isolating snails prior to mating increased their propensity to mate. The significantly higher mating propensity which is an indirect measure of mating frequency (Baur and Baur, 1992) and higher number of ovipositors observed for *A. fulica* as compared to the other species (41.7 vs 23.1% and 41.7 vs 27.0%, and 40 vs 33%, respectively) indicate that *A. fulica* has higher sexual reproductive potential than the other species tested. Tomiyama and Miyashita (1992) reported similar conclusion from review of reproductive data on *Achatina* and *Archachatina* species. *A. fulica* is generally regarded as the most invasive and hardy of all species of achatinides (Raut and Barker, 2002; Skelley et al., 2010). The high reproductive capacity of *A. fulica* is an adaptation to its invasive nature, enabling it to survive in diverse environments (Raut and Barker, 2002).

Table 3 shows the duration of various reproductive activities for the snail species. The table shows that duration of courtship latency (DCL) was 68.4 ± 3.6 min on the average for *A. fulica* which was significantly (P ≤ 0.01) lower than those of *A. marginata* (92.6 ± 2.8 min) and *A. achatina* (85.4 ± 2.4 min). For courtship duration, *A. fulica* averaged 195.5 ± 5.7 min, while *A. marginata* and *A. achatina* averaged 240.4 ± 9.2 and 210.7 ± 6.4 min, respectively. Duration of mating in *A. fulica* was also comparatively shorter at 278.6 ± 6.1 min as compared to 305.8 ± 10.0 min for *A. marginata* and 368.8 ± 9.3 min for *A. achatina*. Duration of reproductive activities (DRA) (mean time from courtship to end of mating) was correspondingly shorter for *A. fulica* (474.1 min or 7.9 h). Reports of duration of reproductive activities in the Giant African land snails are scarce in literature. The few reports available are, however, in good agreement with the values reported in the present study. Specifically, Raut and Ghose (1984) reported that duration of copulation in *A. fulica* is typically 6 to 8 h with range of 1 to 24 h, while Hodasi (1979) reported copulation time of 12 h in *A. achatina*. Tomiyama (1994) observed duration of courtship in *A. fulica* of less than 5 min and average copulation time of 4.6 h (range of 1.5 to 7.5 h) under field situation. Other reports (Wikipedia, 2007; Chase, 2007) showed that duration of courtship was 30 min and the actual transfer of gametes (duration of copulation) was several hours under field situation. These reports indicate very wide variation in the duration of these reproductive behaviours, being highly species and environmentally dependent (Kakline and Bacon, 1961; Raut and Barker, 2002). Table 3 also shows that the snails laid their first set of eggs between 9 to 16 days post copulation which agrees with the generally reported range of 8 to 20 days post mating (Petsnails, 2009).

Table 4 presents the egg weight, egg length and weight of hatchlings for the snail species. The table shows that mean maiden egg weight, egg length and weight of hatchling differed significantly (P ≤ 0.05) among the three species with *A. marginata* eggs being significantly (P ≤ 0.05) heavier and longer than those of *A. fulica* and *A. achatina*. *A. fulica*, though a heavier species lays smaller yellowish eggs while *A. marginata* lays larger chalky white eggs (Raut and Barker, 2002). Comparative assessment of weight of hatchlings for the three species showed significant differences (P ≤ 0.05) with hatchlings of *A. marginata* being heavier (2.46 ± 0.04 g) than those of *A. achatina* (1.62 ± 0.01 g) and *A. fulica* (1.26 ± 0.02 g). For all species, weight of hatchlings accounted for ≥60% of the egg weight which is 61.14% for *A. fulica*; 65.25% for *A. marginata* and 60.45% for *A. achatina*. Ejidike et al. (2002) reported mean maiden egg weight and egg length of 3.2 g and 2.1 cm, respectively, for *A. marginata* and corresponding maiden mean weight of hatchlings of 2.3 g which substantially concur with our report for *A. marginata*. Overall mean calculated from data presented by Ejidike et al. (2002) on all eggs produced (maiden and residual) over a month was, however, higher for egg weight at 4.53 g but remained essentially the same for egg length at 2.30 cm and weight of hatchlings at 2.52 g. Omole et al. (2006) found overall mean egg weight of 4.76 ± 0.91 g and corresponding mean weight of hatchings of 3.88 ± 0.26 (81% of egg weight) in *A. marginata* fed varying levels of supplementary calcium. The lesser values reported in this study was because the first clutch of eggs (maiden eggs) was evaluated. Egg weight and weights of hatchlings are expected to increase with age and with subsequent clutches as suggested by Ejidike et al. (2002).

The phenotypic correlation coefficient for egg weight, egg length and weight of hatchlings showed highly significant (P ≤ 0.001) positive correlation between pairs of these traits (range, r_p = 0.829 to 0.952), which indicates that all three traits are phenotypically and

Table 3. Mean ± S. E. for duration of reproductive activities for different species.

Mating type	Courted/mated snail (pair)	DCL (min)	DC (min)	DM (min)	DRA (min)	DMO (day)
F x F	5	68.4±3.6[a]	195.5±5.7[a]	278.6±6.1[a]	474.1[a]	9±0.6[a]
M x M	3	92.6±2.8[b]	240.4±9.2[b]	305.8±10.0[b]	546.2[b]	11±1.4[a]
A x A	3	85.4±2.4[b]	210.7±6.4[b]	368.9±9.3[c]	578.8[b]	16±2.2[b]

[a,b,c] = Means on the same column with different superscripts are significantly (P ≤ 0.05) different. DCL = Duration of courtship latency; DC = duration of courtship; DM = duration of mating; DRA = duration of reproductive activities (DC + DM); DMO = duration from end of mating to oviposition.

Table 4. Mean ± S. E. for egg parameters and weight of hatchlings for different species of giant African land snails.

Mated type	Egg number	EW (g)	EL (cm)	WH (g)
F x F	240	2.05±0.02[b]	1.70±0.01[a]	1.26±0.02[a]
M x M	98	3.77±0.03[a]	2.29±0.04[b]	2.46±0.04[b]
A x A	120	2.68±0.01[b]	1.46±0.01[a]	1.62±0.01[a]

[a,b]: Means on the same column with different superscripts are significantly (P ≤ 0.05) different. F x F = *A. fulica* pairs; M x M = *A. marginata* pairs; A x A = *A. achatina* pairs. EW = egg weight, EL = egg length; WH = weight of hatchling.

positively associated to a reasonable extent.

Conclusion

This study examines the duration of reproductive activities, egg parameters and weight of hatchlings in three giant African land snails. From the results presented, it is concluded that snails differ in their time spent in pre-mating and mating activities (courtship latency, courtship and copulation). While such reproductive traits as mating propensity and number of ovipositors were higher for *A. fulica*, *A. marginata* had the highest egg parameter values and weight of hatchlings. The possibility of hybridization in these two species should be explored to show their individual genetic potentials in intensive snail production.

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