

ARE MATING FREQUENCIES RELATED TO ABSOLUTE ABUNDANCE IN *CENTROBOLUS* COOK, 1897?

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Abstract- Two species of *Centrobolus* were identified (*C. anulatus*, *C. inscriptus*) based on morphology and confirmed using Scanning Electron Microscopy (SEM) of gonopod structure. Two sets of measurements were made from field data: (1) absolute abundance, and (2) mating frequencies. Mating frequencies and absolute abundance were correlated late in the season on the ground and in the trees ($r=0.62$, Z score= 2.17 , $n=12$, $p=0.02$), and early in the season on the ground and in the trees ($r=0.68$, Z score= 2.46 , $n=12$, $p<0.01$). Mating frequencies and absolute abundance were correlated in the trees early and late in the season ($r=0.65$, Z score= 2.34 , $n=12$, $p<0.01$), and on the ground early and late in the season ($r=0.53$, Z score= 1.75 , $n=12$, $p=0.04$). Mating frequencies for *C. anulatus* were positively correlated with absolute abundances ($r=0.72$, Z score= 2.01 , $n=8$, $p=0.02$). Mating frequencies late on the ground and in the trees were related to absolute abundance in *C. anulatus* (Figure 6: $r=0.93$, Z score= 2.92 , $n=6$, $p<0.01$). Mating frequencies early and late on the ground were related to absolute abundance in *C. anulatus* ($r=0.87434522$, Z score= 2.34041316 , $n=6$, $p<0.01$). Mating frequencies for *C. inscriptus* were not correlated with absolute abundances ($r=0$, Z score= 0 , $n=8$, $p=0.50$).

I. INTRODUCTION

The red millipede genus *Centrobolus* is well known for studies on sexual size dimorphism (SSD) and displays prolonged copulation durations for pairs of individuals of the species [3-8]. *Centrobolus* is distributed in temperate southern Africa with northern limits on the east coast of southern Africa at -17° latitude South (S) and southern limits at -35° latitude S. It consists of taxonomically important species with 12 species considered threatened and includes nine vulnerable and three endangered species [23]. It occurs in all the forests of the coastal belt from the Cape Peninsula to Beira in Mocambique [22]. Spirobolida has two pairs of legs modified into gonopods on the eighth and ninth diplosegments [25]. In *Centrobolus* the coleopods are the anterior gonopods of leg-pair eight. They can be classed as paragonopods or peltogonopods because they are fused into a single plate-like

structure and play a subsidiary role as inseminating devices. In contrast, leg-pair nine is sperm-transferring [1]. The sternites (or stigma-carrying plates [26]) prevent lateral shifting (stabilizer) and stretch the vulva sac in a medial plane [3]. These worm-like millipedes have female-biased SSD [3-8, 11-18, 20]. From the results, correlations between absolute abundance and mating frequencies were checked for correlations.

II. MATERIALS AND METHODS

Millipedes were hand collected in coastal forest habitat at Mtunzini ($28^\circ 55' S$; $31^\circ 45' E$) during the summer season (1995-1996). Individual millipedes were identified as species and sexed based on the presence of gonopods in males and their absence in females. Individuals were counted as either on or above ground (>30 cm but <3 m above ground surface). The number of mating pairs was recorded. The total number of adults was used to estimate the relative abundance. Intercalary males were excluded from the counts. Two species of *Centrobolus* were identified based on morphology and confirmed using Scanning Electron Microscopy (SEM) of gonopod structure (*C. anulatus*, *C. inscriptus*). The gonopods were dissected from males of these two species and prepared for SEM. Specimens were fixed, first in 2.5% glutaraldehyde (pH 7.4 phosphate-buffered saline) at $4^\circ C$ for 24 hours, then in osmium tetroxide (2%). Dehydration through a graded alcohol series (50%, 60%, 70%, 80%, 90% to 100% ethanol) and critical point drying followed. Specimens were mounted on stubs and sputter coated with gold palladium. Gonopods were viewed under a Cambridge S200 SEM. SEM micrographs were examined and the individual

components of the gonopods were identified according to the available species descriptions. Two sets of measurements were made from the field data (1) absolute abundance and (2) mating frequencies. Absolute abundance and mating frequencies were correlated here using Pearson's Correlation Coefficient (<https://www.gigacalculator.com/calculators/correlation-coefficient-calculator.php>).

III. RESULTS

Absolute abundance and mating frequencies were marginally related overall ($r=0.59094643$, Z score= 2.44859878 , $n=16$, $p=0.00717066$). Absolute abundance and mating frequencies were positively related late in the season on the ground and in the trees (Figure 1: $r=0.61858070$, Z score= 2.16810846 , $n=12$, $p=0.01507516$). Absolute abundance and mating frequencies were positively related early in the season on the ground and in the trees (Figure 2: $r=0.67510520$, Z score= 2.46019472 , $n=12$, $p=0.00694309$). Absolute abundance and mating frequencies were positively related early and late in the season in the trees (Figure 3: $r=0.65357112$, Z score= 2.34452249 , $n=12$, $p=0.00952571$). Absolute abundance and mating frequencies were positively related early and late in the season on the ground (Figure 4: $r=0.52588419$, Z score= 1.75331657 , $n=12$, $p=0.03977380$). Mating frequencies for *C. anulatus* were positively correlated with absolute abundances (Figure 5: $r=0.71652981$, Z score= 2.01352669 , $n=8$, $p=0.02202956$). Mating frequencies late on the ground and in the trees were related to absolute abundance in *C. anulatus* (Figure 6: $r=0.93341446$, Z score= 2.91725455 , $n=6$, $p=0.00176570$). Mating frequencies early and late on the ground were related to absolute abundance in *C. anulatus* (Figure 7: $r=0.87434522$, Z score= 2.34041316 , $n=6$, $p=0.00963119$). Mating frequencies for *C. inscriptus* were not correlated with absolute abundances in general ($r=0$, Z score= 0 , $n=8$, $p=0.50$).

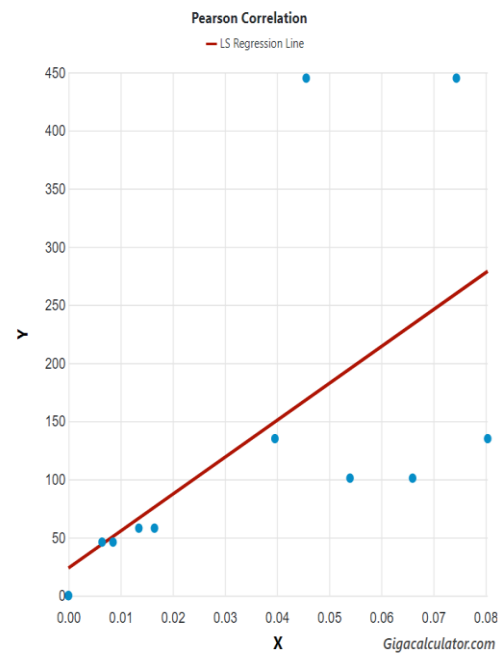


Figure 1. Relationship between absolute abundance (y) and mating frequencies (x) for *C. anulatus* and *C. inscriptus* late in the season on the ground and in the trees.

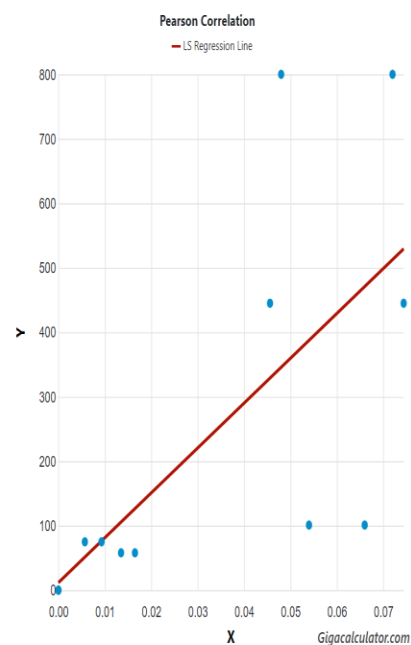


Figure 2. Relationship between absolute abundance (y) and mating frequencies (x) for *C. anulatus* and *C. inscriptus* early in the season on the ground and in the trees.

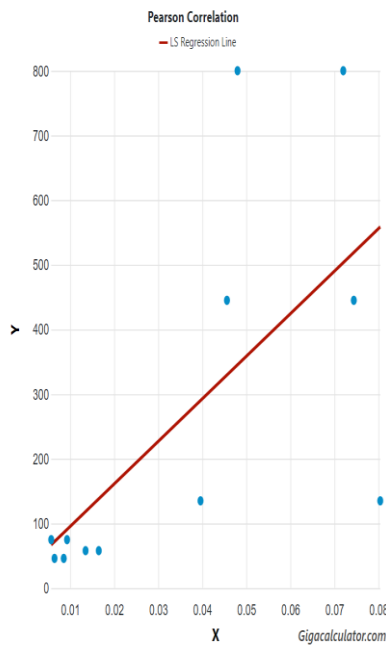


Figure 3. Relationship between absolute abundance (y) and mating frequencies (x) for *C. anulatus* and *C. inscriptus* early and late in the season in the trees.

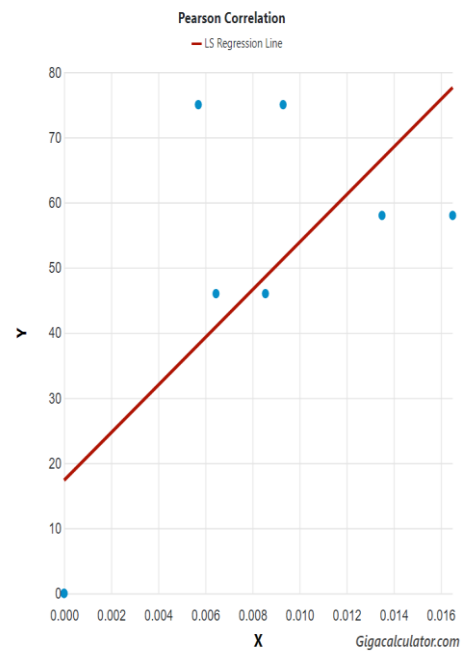


Figure 5. Relationship between absolute abundance (Y) and mating frequencies (x) for *C. anulatus*.

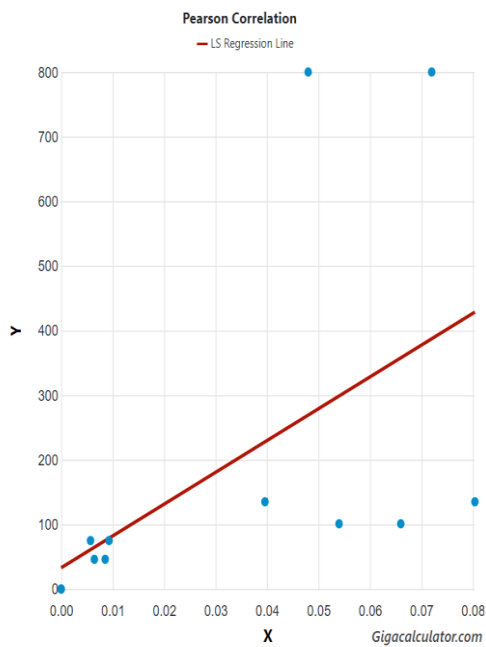


Figure 4. Relationship between absolute abundance (y) and mating frequencies (x) for *C. anulatus* and *C. inscriptus* early and late in the season on the ground.

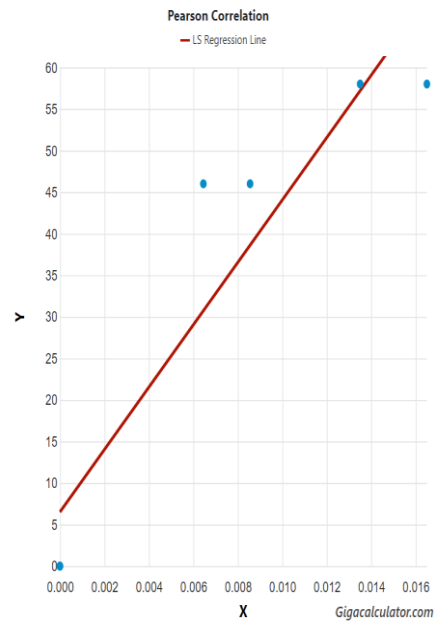


Figure 6. Relationship between absolute abundance (Y) and mating frequencies (x) for *C. anulatus* late on the ground and in the trees.

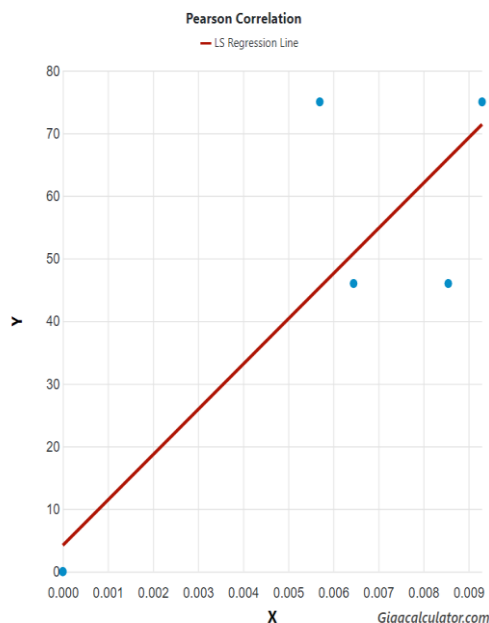


Figure 7. Relationship between absolute abundance (Y) and mating frequencies (x) for *C. anulatus* early and late on the ground.

IV. DISCUSSION

The absolute abundance and mating frequencies were estimated in two *Centrobolus* species. A direct relationship between two factors (absolute abundance and mating frequencies) in the millipedes is compared which certainly supports the relationship in four ways, i. e. early, late, on the ground, and in the trees. A relationship between these behavioral factors is present across two species suggesting adaptation to lifetime reproductive success. *C. inscriptus* had the mating frequencies independent of absolute abundance while *C. anulatus* had lower mating frequencies and a lower abundance. In other words, there were four positive relationships between absolute abundance and mating frequencies early, late, on the ground, and in the trees, probably all attributed to *C. anulatus*. Mating frequencies late on the ground and in the trees were related to absolute abundance and mating frequencies early and late on the ground were related to absolute abundance in *C. anulatus*. The *C. anulatus* - *C. inscriptus* species pair represents an example of "homotypy", "nondeceitful homotypy" and "arithmetic homotypy" [24].

V. CONCLUSION

New relationships between absolute abundance and mating frequencies among the *Centrobolus* millipedes support the function of this behavior as adaptive toward mate competition and assuring paternity among increased mating frequencies with absolute abundance. Frequency-dependent and frequency-independent selection were demonstrated in two species.

APPENDIX.

Male and female mating frequencies (early, and late in a season, on the ground, and in the trees), in two species of *Centrobolus*.

- 0, 0 (*C. anulatus*).
- 0, 0 (*C. anulatus*).
- 0.0165, 58 (*C. anulatus*).
- 0.0135, 58 (*C. anulatus*).
- 0.066, 101 (*C. inscriptus*).
- 0.054, 101 (*C. inscriptus*).
- 0.0744, 445 (*C. inscriptus*).
- 0.0456, 445 (*C. inscriptus*).
- 0.0093, 75 (*C. anulatus*).
- 0.0057, 75 (*C. anulatus*).
- 0.072, 800 (*C. inscriptus*).
- 0.048, 800 (*C. inscriptus*).
- 0.00855, 46 (*C. anulatus*).
- 0.00645, 46 (*C. anulatus*).
- 0.0396, 135 (*C. inscriptus*).
- 0.0804, 135 (*C. inscriptus*).

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