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# The mate searching behavior of *Perlinella drymo* (NEWMAN) (Plecoptera: Perlidae) in relation to drumming on a branched system

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The mate searching behavior of a central Texas, USA population of the stonefly *Perlinella drymo* (NEWMAN) was studied experimentally on a tree branch substrate. The drumming duets of experimental pairs increased male searching activity, influenced their directional movements and significantly decreased the time required for them to find females. Search patterns showed that males could integrate information received from female answer signals, to correct wrong turns and reinforce correct turns toward her. This is the first study directly supporting the hypothesis that vibrational communication aids mate finding on the natural branched systems of woody vegetation encounter sites.

Keywords: Plecoptera, behavior, drumming, mate searching, communication

#### INTRODUCTION

Adults of the northern hemisphere stonefly group Systellognatha generally do not feed (HYNES, 1976); thus, their adult resource acquisition behavior is largely confined to mate finding. Mate finding in the suborder Arctoperlaria is presumably facilitated by species-specific drumming or other forms of intersexual vibrational communication (STEWART & MAKETON, 1 991), although no observations in nature or laboratory experiments on a natural, branched system have ever confirmed this. A study by ABBOTT & STEWART (1993) of *Pteronarcella badia* (HAGEN) showed that on a flat, experimental arena, males duetting with females located the female in less time than those of non-communicating pairs. However, the artificial nature of the flat arena may have influenced the search behavior of this species whose natural encounter site is on riparian shrubs (DEWALT & STEWART, 1995).

Drumming signals are known for many Arctoperlarian species, including those of *Perlinella drymo* (NEWMAN) (ZEIGLER & STEWART, 1977; STEWART & ZEI-GLER, 1984; STEWART & MAKETON, 1991), but their mate searching patterns, and specifically if or how vibrational communication is used for locating mates has remained virtually unstudied in stoneflies. STEWART (1994) proposed that the communication-search systems of Arctoperlaria will likely involve some combination of aggregation of males and females at specific encounter sites (PARKER, 1978), ranging search by males (BELL, 1990, 1991) accompanied by vibrational calling, then localized searching for females after duets with females have been established.

STEWART & ZEIGLER (1984) showed that *P. drymo* could communicate with their unique drumming duets for distances up to 8 m on a continuous wooden rod 5-9 mm in diameter. Pairs were unable to communicate over a 25-cm distance on a 4.2 kg rock. Our research reported here was conducted to determine: 1) the mate searching pattern of *P. drymo* on a natural, dried tree branch, that simulates the

branching of its natural tree encounter site, 2) if duetting facilitates mate finding on the branched system, and if so, 3) to what degree males can use duetting information to locate females.

#### MATERIALS AND METHODS

Nymphs of *P. drymo* were collected on February 24, 1994, from the Middle Fork of the Bosque River, McClennan Co., Texas, USA, ~1 mile north of the town of Crawford. They were returned to the Aquatic Entomology Laboratory at the University of North Texas and reared in a Frigid Units Living Stream<sup>TM</sup> at simulated stream temperatures. A representative tree branch 80 cm in length was collected from the same locality and mounted vertically on a wooden base for use in laboratory trials to simulate the natural encounter site of *P. drymo*.

Each experimental trial consisted of: (1) releasing a virgin female at the base of the branch and allowing her free ranging movement until she came to rest, (2) releasing a virgin male at the same starting point, and (3) videotaping each search sequence, including drumming, using a Sony CCD-FX510 camera for later analysis. Each trial was terminated after a successful find of a female by the male or after ten minutes. Eighteen trials were conducted with 7 virgin males and 8 virgin females. Experimental room temperatures were  $22 \pm 2^{\circ}$  C and fluorescent light intensity was about 85 fc.

Directional changes made by males while searching were categorized as either correct turns or incorrect turns. Correct turns consisted of the male traveling in a direction away from the female and reversing direction to travel in a direction toward the female or when he reached a bifurcation of the tree branch and traveled along the side leading to her. Incorrect turns consisted of the male traveling in a direction toward the female and reversing direction to travel away from the female or when he reached a bifurcation to travel away from the female or when he reached a bifurcation of the tree branch and traveled along the side not leading to her.

A chi-square test was used to test for differences in find time and success of finding between duetting and non-duetting pairs. A Yates correction was used to conservatively adjust the chi-square value because there were only two classes in each case. For comparing turns made by the male toward the female, Welch's approximate t-test was used instead of a parametric t-test because variances were not equal. The non-parametric Wilcoxon 2-sample test was used to compare distance traveled between males of duetting and non-duetting pairs and turns made by the male away from the female. In these instances, more than one assumption of parametric tests were not met.

#### RESULTS AND DISCUSSION

Fourteen of the trials involved duetting that was established within an average of 23.40 ( $\pm$  25.96 SD) seconds after release of the male, indicating that handling did not adversely affect the males and that they accepted the branch as a natural system for searching. After release, a female in each experimental trial typically moved up the tree branch and assumed a resting position. A typical search/communication pattern involved the male moving up the main stem immediately after release, and searching branches while intermittently calling the female who usually remained stationary on one of the upper branches. Duets were established after the female began to answer male calls, and this initiated a more localized search by the male. Figs 1-3 show three variations of this pattern.



Fig. 1. A *P. drymo* male search pattern covering 121 cm in 71 s that resulted in finding the female.

Fig. 2. A *P. drymo* male search pattern covering 110 cm in 57 s that resulted in finding the female. Fig. 3. A *P. drymo* male search pattern covering 142 cm in 63 s that resulted in finding the female

Generally, males found the female within the ten minute limit in 14 of the trials, and were unsuccessful in four trials. The male located the female and attempted copulation in 12 of the 14 trials involving duetting. Duetting was never established in only four of the 18 trials, indicating a strong tendency toward vibrational communication, yet the male found the female in two of these. Table 1 gives a summary of the trials.

Duet establishment stimulated males to search more intensely; those of duetting pairs traveled significantly farther than males of non-duetting pairs (Wilcoxon 2-Sample Test with Continuity Correction of 0.5, Z = -1.96, p = 0.05) (fig. 4). This supports the idea that the male gains information indicating that a receptive female is within communication distance, and this stimulates further and more vigorous localized searching. This is energetically important to adult *P. drymo* males, who presumably do not feed as adults, but live on fixed energy stores obtained as aquatic nymphs.

Time required for males to find females was then examined. Males of duetting pairs required significantly less time to locate a female than males in non-duetting pairs ( $X^2$  with Yates correction,  $X^2 = 45.88$ , p<<0.001) and male find time was highly contingent upon duetting (Contingency Coefficient = 0.44). Reducing find time would be important in the ephemeral (5-6 days) adult lives of these stoneflies where time is an important constraint.

Trial	# of duets	# of incorrect turns	# of correct turns	Distance traveled (cm)	Time elapsed (sec)	Find ?
1	0	0	2	58.5	60	Yes
2	50	22	24	387.5	592	Yes
3	17	4	5	110.0	57	Yes
4	60	20	23	604.0	600	No
5	57	6	7	203.0	300	Yes
6	N/A	3	6	121.0	70	Yes
7	35	41	21	463.5	300	Yes
8	7	0	1	72.5	28	Yes
. 9	51	15	15	377.7	334	Yes
10	5	2	4	144.9	64	Yes
11	0	2	1	90.6	600	No
12	0	8	4	145.4	72	Yes
13	0	4	1	38.0	600	No
14	64	17	32	498.7	420	Yes
15	1	2	1	101.5	32	Yes
16	19	6	5	172.5	120	Yes
17	4	3	2	142.4	63	Yes
18	17	3	1	70.0	118	Yes

Table 1. Summary of mate searching trials for P. drymo pairs on a branched system

Males of duetting pairs did not locate the female significantly more often than males in non-duetting pairs in this experimental setting ( $X^2$  with Yates correction,  $X^2 = 0.51$ , 0.50>p>0.25). The small numbers of non-duetting pairs and the small size of the branch used in these trials may have biased our results against showing a difference in frequency of finds. The small branch allowed even a random search by the two nonduetting males to find the female. The much larger size of trees and shrubs in riparian vegetation would render a random search much less efficient and presumably lower chances of a male accidentally finding a female.

During duetting the male presumably receives information carried in the female's answer. If he is able to integrate and utilize this information it would influence his search pattern and result in greater efficiency in locating her. We compared the number of turns toward and away from the female made by the males in duetting pairs to those in random searches, to determine if males could integrate and utilize information contained within the female answer. Males of duetting pairs made significantly more turns toward the female than did males of non-duetting pairs (Welch's Approximate T-Test, T = 3.00, p = 0.01), but males of duetting pairs did not make significantly fewer turns away from the female than males of non-duetting pairs (Wilcoxon 2-Sample Test with Continuity Correction of 0.5, Z = -1.01, p = 0.31).

These results would be expected on a branching system due to the nature of the substrate-borne signals. Without using any form of communication the male is making a random search. His searching of branches and sub-branches results in a certain number of turns away from or toward the female over a fixed time or until the female is found. When duetting, the female is giving the male directional infor-



mation about her location as an answer to each male call (i.e., the vibrational signals come from behind or in front of him). His response results in a general directional movement toward her. Males never called at branch bifurcations. When a male therefore reaches a fork he randomly travels up one side or the other. If he first travels up the one that does not lead to the female, the further answer signals come from behind him and the turn is an incorrect one. At that point, he turns around and travels back to the bifurcation and takes the other side resulting in a significant increase in correct turns toward the female. Conversely, if a male takes the branch leading to the female, then further duetting provides reinforcing information coming from in front of him indicating that he is traveling toward her. This pattern is repeated at each bifurcation, until he finds the female. These observations indicate that female answers provide correctional and reinforcing information to the male search. Repeated duetting results in the male not searching entire branches of the main stem but shorter distances on branches not containing the female until correctional information from the female signal is received and integrated (figs. 1-3). We surmise that future selection pressures may lead to the more efficacious behavior of a male calling immediately at each bifurcation, where a female answer would provide the correct directional information every time, and reduce required branch traveling distance.

In conclusion, this study provides strong support for the hypothesis that drumming in *P. drymo* and probably other Arctoperlarian stoneflies is a mechanism for locating mates. Duetting stimulates a male to vigorously continue a localized search and provides the male with directional information on a branched system that he can integrate to make corrections in direction and to decrease time and energy expended to locate a female. This study also indicates that *P. drymo* is well adapted to locating mates on bifurcating branches using vibrational signals. Additionally, due to the limitations on directional travel of percussion-produced vibrational signals through naturally branched wood substrates and directional choices a stonefly responding to those signals can make, the drum search system of *P. drymo* may represent a model for species that utilize bush or tree encounter sites.

#### ACKNOWLEDGEMENTS

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