

Romancing the firefly: New insights into what goes on when the lights go off

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This is a male *Photinus* firefly that failed to successfully transfer a spermatophore -- or nuptial gift -- during mating. The spermatophore is trailing from male reproductive tract. While a female's initial assessment of potential mates is based on males' luminescent flashes, research by Tufts University biologists reveals that once a pair makes contact, flashes no longer matter. Instead, it's those males that have larger nuptial gifts to give that win out with higher reproductive success. Credit: Tufts University

The twinkling of fireflies heralds summer romance for these magical insects. While courting on-the-wing, male fireflies attract females' attention with bioluminescent flashes.

But new research from <u>biologists</u> at Tufts University's School of Arts and Sciences, published online in <u>Proceedings of the Royal Society B</u>:



Biological Sciences, reveals that, after the lights go out, female fireflies prefer substance over flash. They seem to choose mates able to give them the largest "nuptial gift" (a high protein sperm package that helps females produce more eggs) without regard to flashes. Those generous males are also more likely to succeed in becoming the fathers of the next firefly generation.

Previous work on *Photinus* fireflies shows that females are very picky during the on-wing stage of <u>courtship</u>. These females will only flash a response toward select males that light up with especially attractive courtship flashes. After a lengthy back-and-forth exchange, the flashing stops, the lights go out, and firefly pairs spend the night together.

A night of firefly romance also includes gifts, called spermatophores, that each male donates to his sweetheart. But the next night these females are likely to mate again with a different male.

After a female has mated with several males, the big evolutionary question becomes: which male gets to pass along his genes to the next generation of firefly babies?

"Lots of people don't realize that <u>sexual selection</u> is happening not only before mating, but also during and even after mating," said Professor of Biology Sara Lewis, an expert on the evolutionary process of sexual selection and senior author on the paper. "Focusing on what happens after contact, we wanted to examine how much a male's success -- in both mating and fathering offspring -- depended on his flashes or on his nuptial gift offering."

Lewis and coauthor Adam South, who studied in the Lewis lab and recently received his Ph.D. from Tufts' Graduate School of Arts and Sciences, set up an experiment using infrared video and paternity testing based on firefly DNA to determine what makes certain males more



successful after the lights go out.

To get the female fireflies in the mood, the researchers relied on LED lights programmed to make two kinds of flashes. Some females saw only artificial male flashes determined by previous research to be highly attractive to females; others saw unattractive flashes.

Male fireflies were also split into two groups: virgins whose nuptial gifts were large since the males had never mated and males whose spermatophores were smaller because they had mated the previous night. After several minutes of courtship flashing, males and females were put together in pairs, and the Tufts biologists videotaped their close-up courtship behaviors under infrared illumination. Because they take place under cover of darkness, many of these behaviors had never before been observed.

Analysis of hours of firefly video revealed that once a female was in close quarters with a male, she was much more likely to mate with males that had larger nuptial gifts to offer, as determined by the researchers' later examination. The females didn't seem to care what kind of male flashes they had seen.

"We were surprised to discover that attractive flashes only seem to benefit males during the early stages of firefly courtship," said South. "Initially, flashes are important. Female fireflies preferentially respond to males based on temporal flash characteristics. Once males make physical contact, however, females switch to an alternative cue -- one that's related to male nuptial gift size. What makes this especially intriguing is that females have no way to directly evaluate gift size, since it's created and transferred internally."

Furthermore, when females mated sequentially with two different males, paternity testing of their offspring revealed yet another benefit for big



gift-givers. Males that gave larger nuptial gifts fathered more of their mate's offspring compared with rival males. South and Lewis say that's probably because larger gifts contain more sperm.

So not unlike human romance, love remains a mystery among <u>fireflies</u> and first impressions are only part of the story. While a female's initial assessment of potential mates is based on males' luminescent flashes, the Tufts research shows that once a pair makes contact, sexy flashes no longer matter. Instead, it's those males that have larger nuptial gifts to give that win out with higher reproductive success. <u>Males</u> with limited resources may face a trade-off between investing either in sexy flashes or in costly gifts.

More information: A. South and S.M. Lewis, Determinants of reproductive success across sequential episodes of sexual selection in a firefly, *Proc. R. Soc. B* rspb20120370; published ahead of print April 25, 2012, doi:10.1098/rspb.2012.0370 1471-2954

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