Male elephants woo females with precise chemistry

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The exact chemical blend of a pheromone emitted by older male elephants in musth influences both a female elephant's interest in mating and how other surrounding elephants behave, a new study has found.

The researchers at Oregon Health & Science University and the University of Auckland in New Zealand say the release of a specific proportion of two mirror images of the pheromone, frontalin, depends on whether the male elephant is mature enough and has reached a particular stage of musth, an annual period of sexual activity and increased aggression.

"This study reveals the precision and specificity of inter-animal signaling possible," co-author L.E.L. "Bets" Rasmussen, Ph.D., research professor of environmental and biomolecular systems, OHSU OGI School of Science & Engineering, said of the study published in the Dec. 22 edition of the journal *Nature*. "This is the first example, in mammals, of the use of this very precise signaling and ratio of enantiomers in signaling."

An enantiomer is one of a pair of chemical compounds whose molecular structures are mirror images of each other. Frontalin, a pheromone discharged during musth by male Asian elephants from a temporal gland located between the eye and ear, comes in two forms, each representing one half of the enantiomer pair and identified as either "plus" or "minus."

The OHSU and New Zealand team showed that the enantiomers of frontalin are released in the elephants in specific ratios that depend on the animal's age and stage of its musth. They also found that alterations in those ratios elicit different responses in not only female elephants, but also other male elephants near the male emitting the pheromone.

"We were certainly quite surprised" by the results, said the study's lead author, David R. Greenwood, Ph.D., associate professor in the School of Biological Sciences at the University of Auckland and scientist with the Molecular Olfaction Group at HortResearch in New Zealand.

Scientists have assumed that only one form of frontalin would be made by elephants, but by merely changing the ratios of frontalin's enantiomer composition, "you're actually changing the message signals, and you end up having a signal that is essentially different," Greenwood added. "It's not just a single message."

The research team analyzed secretion samples from six male elephants

and found that the pheromone is first detectable in the late teens, with the quantity secreted rising about 15-fold over a 25-year life span. Young males secreted significantly more of the "plus" frontalin than "minus," but the ratios became almost equal as the elephant matured, especially between ages 31 and 43.

Musth periods get longer as the males age, according to the study, and, importantly, the enantiomeric composition of the frontalin secreted is almost equal in the middle part of a musth episode, a period of prime signaling by male elephants. In contrast, at the end of the physiologically exhausting musth episode, the ratio becomes more varied so that at the end of musth, the ratio is askewed toward the minus enantiomer, allowing other males and females to detect the ending of musth.

The team then tested the effect the various enantiomer ratios had on the elephants. They examined ovulating or "follicular" females, and females that were either pregnant or in a non-reproductive "luteal" phase. They also tested young and old males. It found that low concentrations of frontalin, represented when the enantiomer ratio is more "plus" than "minus," was of mild interest to both young and old males, but when the ratio became balanced - equal amounts of plus and minus frontalin - males of all ages, as well as luteal-phase and pregnant females, were repulsed. Only ovulating females were attracted.

The study's results indicate that the ratio of frontalin enantiomers allows other elephants to distinguish both the maturity of male elephants in musth and the phase of musth.

The researchers hope to use the study's results to "tease apart the entire route of how pheromones move" from the source elephant to the recipient. They already have been studying the other end of that route - the proteins in the elephants' olfactory systems that receive the signals sent by the source.

"We're interested in where the frontalin is bound to the proteins in the various olfactory mucous milieus and transferred to the vomeronasal organ, where it's sensed as a pheromone," Greenwood said. "We're promoting the elephant as a good olfactory model because their olfactory systems are enormous and separated in space. This is combined with its easily observable and countable olfactory responses involving its trunk. We can't do this in any other mammalian species."

Rasmussen added: "All of these responses and resultant behaviors are measurable in time. Not milliseconds, but seconds. We have a repertoire of follow-up behavior, such as trumpeting, running away and circling, that also can be counted and scored. Such information is translatable at the basic level to other animals," including humans.

Source: Oregon Health & Science University