US researchers have used statistical methods to unravel the detailed scent posts left by hyenas and demonstrated that at least in part, the markers are a product of more than one species.

Despite their perhaps dog-like appearance hyenas are rare carnivores more closely related to felines. There are just four species in this mammalian family found in Africa and Asia where their presence represents an important component of local ecosystems. They are not generally gregarious animals with the exception of the spotted hyena of Africa, *Crocuta crocuta*, colloquially known as the “laughing” hyena for its eerily human call. They are, however, more hunter than scavenger, regardless of the deceived wisdom regarding their behaviour and even the non-social species live in family groups and will congregate at a kill.

**Symbiotic scent**

The complex social behaviour of *C. crocuta* is driven by chemical communication, this much was well-known. Pheromones, glandular scent marking, urine and faeces all play a part in the status updates these fascinating creatures share in the olfactory equivalent of microblogging. Now, researchers from Michigan State University have used cheminformatics to sniff out the chemistry of hyena scent marking and revealed that, in part, the details of each marker are the product of symbiotic bacteria that co-exist with the hyena. MSU's Kevin Theis, Arvind Venkataraman, Jacquelyn Dycus, Keith Koonter, Emily Schmitt-Matzen, Aaron Wagner, Kay Holekamp and Thomas Schmidt provide more details in the *Proceedings of the National Academy of Sciences*.

“When hyenas leave paste deposits on grass, the sour-smelling signals relay reams of information for other animals to read,” explains lead author and post-doctoral researcher Theis. “Hyenas can leave a quick, detailed message and go. It's like a bulletin board of who's around and how they're doing.”

In work supported by the US National Science Foundation (NSF), the team demonstrated that the chemical fingerprint of the hyena scent pastes shows that the bacteria present are far more diverse than scientists had first thought. The bacteria are doing the yeoman’s job of sending these messages, the researchers suggest. “Scent posts are bulletin boards, pastes are business cards, and bacteria are the ink, shaped into letters and words that provide information about the paster to the boards’ visitors,” Theis explains. “Without the ink, there is potentially just a board of blank uninformative cards.”

**Glandular bacteria**

Theis working alongside MSU zoologist Holekamp and colleagues studied multiple groups of male and female *C. crocuta* and the related and striped hyena (*Hyaena hyaena*) in Kenya. Their cheminformatics on gas-chromatography/mass spectrometry data and bacterial surveying using next-generation sequencing gave them what they refer to as an unprecedented view of the diversity of microbes living symbiotically in the mammals’ scent glands revealing much greater diversity than earlier had suggested.

Importantly, the bacterial diversity is different between the hyena species studied and varies with sex and reproductive state among the spotted hyenas, the team reports. Moreover, the variation in scent gland bacterial communities was strongly
correlated with variation in the odour profiles of the scent glands, which corroborates the notion that the bacteria themselves are responsible for the variation in scent.

“There have been around fifteen prior studies pursuing this line of research,” Theis explains. “But they typically relied on culture-based methods, an approach in which many of the similarities and differences in bacterial communities can be lost. If we used those traditional methods, many of the key findings that are driving our research wouldn’t be detected at all.” In the current work, the team combined microbial surveys and complementary odour data from wild animals for the first time. Theis is now keen to get back to the natural habitat of the hyena so that he and his colleagues can investigate what impact their findings have on their observations of hyena behaviour. “Now I just need to get back into the field to test new predictions generated by this study,” Theis adds. “The next phase of this research will be to manipulate the bacterial communities in hyenas’ scent glands to test if their odours change in predictable ways.”

“The next steps are to first isolate specific bacteria from hyena pastes, give them appropriate growing conditions, and make sure they produce specific odorants as predicted by this study,” Theis told SpectroscopyNOW. “Secondly, to do scent discrimination experiments with hyenas in the field to test if they are indeed paying attention to the specific variation in odorants we observed,” he adds. “Thirdly, to manipulate hyenas’ paste bacterial communities to determine whether their odorant profiles change. Ultimately, I hope to demonstrate that the symbiotic hypothesis for animal communication is a widespread phenomenon in the animal kingdom, and that symbiotic microbes contribute to animals' behavioural phenotypes in many remarkable ways.”

Intriguingly, Theis is also now conducting related research in birds, in collaboration with MSU researcher Danielle Whittaker under the umbrella of MSU’s BEACON Center for the Study of Evolution in Action.

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