The honey bee parasite *Varroa destructor* is a flesh-eater, not a blood-sucker

It is estimated that one-third of the food that you consume every day requires the pollination of a honey bee. But their health is threatened by the parasite *Varroa destructor,* which causes considerable damage to bees by feeding on them. For more than half a century, we believed these parasites consumed the blood of bees, similar to how ticks consume human blood. We show that these parasites consume fat body tissue and that blood ingested is minimal and, likely, incidental. The fat body is an insect organ that, like the mammalian liver, generates important proteins and detoxifies chemicals (like pesticides bees are exposed to). It has the added tasks of managing immune function, water balance and body temperature which is important for winter survival. These findings explain the failure of some previous attempts at developing effectively targeted *Varroa* management strategies. Our work provides a path forward for the development of new treatment methods for controlling *Varroa*.

To test our hypothesis, we utilized an array of different techniques and microscopes to directly assess the feeding behavior of the parasite. Our first hint was that *Varroa* are genetically similar to other parasites that feed on semisolid material rather than blood. Additionally, the structure of their mouths and digestive system lack the adaptations seen in other blood-suckers, and the excrement of *Varroa* is not consistent with a blood-based diet.

We suspected that the parasite fed on fat body when we analyzed the feeding location of the parasite on the bee. When we mapped the frequencies of feeding locations, we found that *Varroa* only fed on the underside of the abdomen of the bee, leaving their mouthparts in close proximity to the fat body. Such strong preference for a specific area is more consistent with feeding on a tissue specific to that area rather than blood which is distributed through the host's body.

To understand how the parasite is feeding, we examined the wound that it causes. To do this, we used a low-temperature scanning electron microscope. This microscope allows us to freeze the parasite on the bee mid-meal. We then removed the frozen parasite to examine the wound in the bee. The wounds were also examined by transmission electron microscopy, which confirmed our hypothesis revealing evidence of damage to the fat body. These methods further showed evidence that the mites digest portions of this organ in the bee before consuming it.

To further confirm the diet of the parasite, we used two dyes that would label either the blood or the fat body tissue. These dyes are fluorescent, which allows us to quantify how much of the dye is in a tissue using a fluorescent microscope. Both dyes were fed to bees simultaneously. We confirmed staining by dissecting the bees and imaging their digestive system, blood, and fat body tissue. As expected, the digestive system had a fluorescent signal from both dyes, the blood had signal from only the yellow, and the fat body had signal from only the red. We then allowed the parasite to feed on dyed bees, and examined the gut contents of the parasite. If the parasite had

been feeding on blood, we would expect to see yellow signal, and conversely, if the parasite had fed on the fat body tissue, we would expect red signal. Our results indicated that the gut of the parasite produced a red signal consistent with the signal from dyed honey bee fat body.

Finally, we examined how a blood or fat body diet would affect survival and reproductive fitness. We developed a test where the parasite would be raised in an artificial environment and be given a diet of blood, fat body tissue, different mixes of the two or nothing (as a control). We monitored the parasite on these diets for several days and checked for survival and egg-laying. The trend of our findings was that the more fat body tissue in the diet of the parasite, the higher the survival rate and the more eggs they produced.

Taken together, these findings disprove the long-held belief that *Varroa* consume blood. The location of the feeding site, the type of damage at the feeding site, the presence of fat body within the gut of the parasite and the relationship to fat body consumption and survival all suggest that the fat body tissue is consumed by the parasite. This fundamentally changes our understanding of this agriculturally significant parasite and has important implications the development of tools, both chemical and nonchemical, to manage this pest.