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Article in *Frontiers for Young Minds* · July 2024

DOI: 10.3389/frym.2024.1343838

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THE TINY BRAINS OF WASPS CAN LEARN AND REMEMBER INFORMATION

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**ON A BEAM
OF LIGHT**

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If you have a garden, you have probably seen many insects flying around looking for food. Despite having miniature brains, these small creatures can learn and memorize flower features, mainly colors and smells, which they associate with nectar and pollen (their food) provided by the flowers. Honeybees are not the only pollinators in nature—wasps also pollinate flowers, but less is known about them. We studied whether wasps could learn and memorize information. We developed a study to investigate whether wasps could learn to associate a flowery smell with sugary water. We found that female and male wasps have powerful learning and memory abilities, which are important for their daily social lives.

INSECTS' BRAINS HAVE SOPHISTICATED ABILITIES

If you look outside, you will find insects everywhere: a fly hovering around a basket of fruit, a bee landing on flowers, a beetle crawling on the ground, or even a wasp hovering over your barbecue. Have you ever thought about how these insects manage to find food in nature? Something you may not know is that, despite their relatively small brains, these tiny creatures are good at learning and memorizing information [1]. Their antennae and their compound eyes (Figure 1A) function kind of like our own noses and eyes. These organs are responsible for detecting odors and visual information, and the details are transported to their brains.

Figure 1

(A) A male wasp (white face) of the *Mischocyttarus cerberus* species. One compound eye and one antenna are labeled. These structures work in similar ways as our eyes and noses. (B) A nest of *M. cerberus*, with a female (yellow face) and a male (white face) indicated.

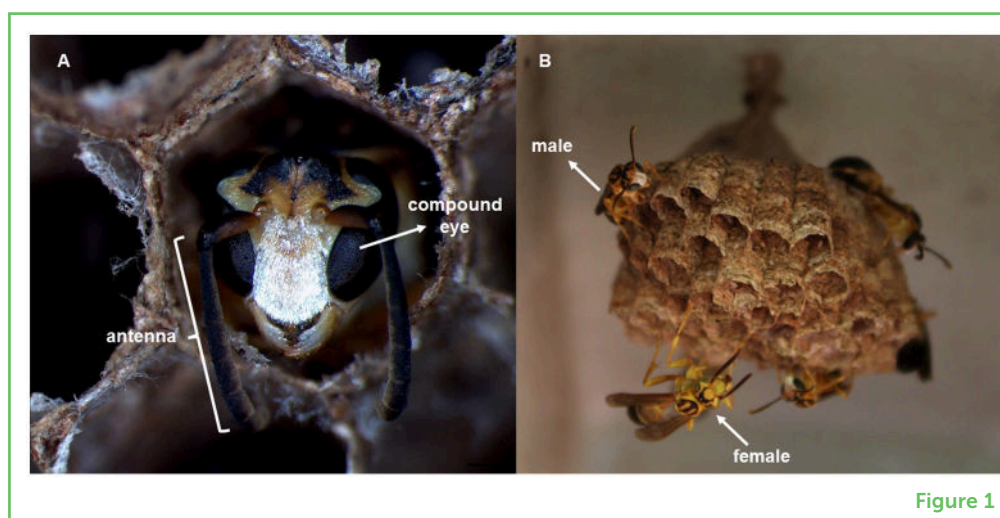


Figure 1

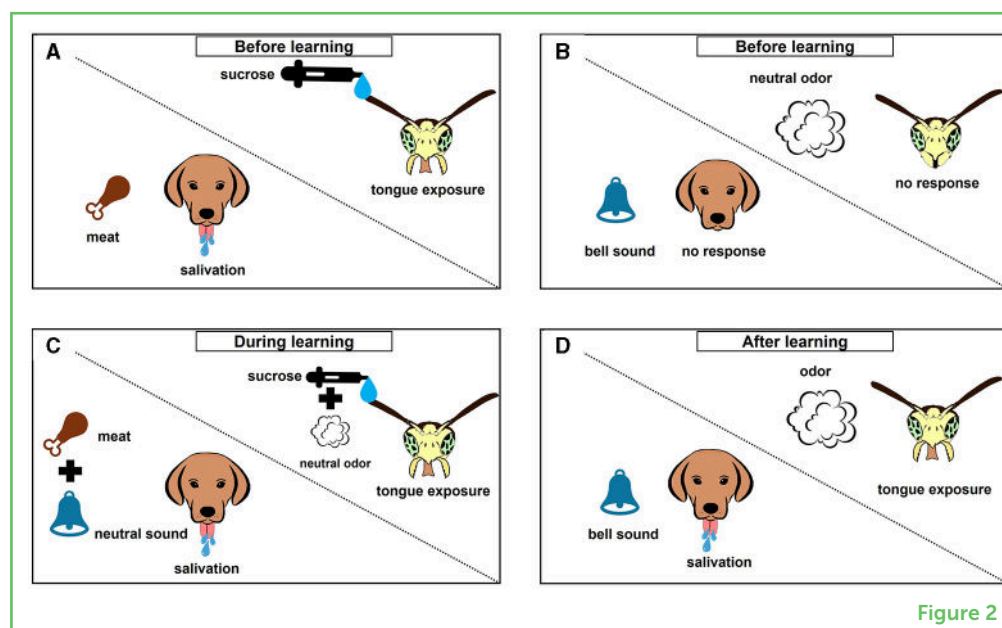
The learning and memory abilities of insects can be studied in the laboratory or in their natural environments. A simple way to study how animals learn, including insects, is by presenting them with the information we want them to learn in combination with something they like—a reward. Animals, like humans, are good at creating mental links between two things. For example, they can associate a new smell and a food they like eating. The approach we use to teach insects is inspired by the scientist Ivan Pavlov, who received the Nobel prize in 1904. Pavlov understood that dogs were good at creating a mental connection between neutral (not responsible for triggering a natural behavior) information like the sound of a bell and something they like to have for lunch, such as a piece of meat. Pavlov realized the reward could activate a natural behavior in the animals. For example, when dogs see a piece of meat (reward) they start salivating (natural behavior). On its own, the neutral information does not have the same effect as the reward—a bell does not normally cause dogs to salivate. However, we can cause the bell and the meat to have similar effects by using some tricks. When we present an animal with the neutral information and the reward *together* several times, after a while, both **stimuli** can induce the natural behavior (Figure 2) [2].

STIMULI

Signals that make an organism notice or react to something.

Figure 2

(A) Before the experiment, a dog salivates when it sees meat and a wasp exposes its tongue when its antennae contact sugar water. (B) Before learning, a dog does not salivate in response to a bell, and a wasp does not react to a flowery scent. (C) During the teaching phase, the dog hears the bell, followed immediately by the meat. The wasp experiences the scent followed immediately by the sugar water. This helps the animals learn the relationship between the two stimuli. (D) After being taught, the dog salivates when it hears the bell, and the wasp exposes its tongue in response to the scent.

**Figure 2**

But the story does not stop here! After an animal learns new information, scientists can also test whether the animal can keep memories of the new information. This is like the strategy your teachers might use after showing you a new concept at school. After learning something new, you probably take an exam so your teacher can check what you memorized. We decided to do this experiment with the wasp *Mischocyttarus cerberus* (*M. cerberus* for short; Figure 1B) [3].

UNDERSTANDING SOCIAL WASPS

Wasps are important in nature—they prey on certain crop pests and they also pollinate flowers [4]. The species we studied, *M. cerberus*, is a social species, which means that they live together in a shared nest and cooperate in their daily activities. In this society, individuals communicate with each other using chemical messages. Wasps touch each other frequently with their antennae, which allows them to smell and recognize each other. The bodies of social wasps are covered by a mix of chemical compounds, which carry information about their age, whether they are male or female, health status, the nest to which they belong, and their species. Most of these chemical compounds are **cuticular hydrocarbons** (CHCs), which are made of atoms of carbon and hydrogen. Once wasps touch each other, they detect the type of information provided by the CHCs.

We believe that noticing and learning odors is fundamental for wasps, not only when they are at their nests but also when they are outside searching for food. We decided to use the approach developed by Pavlov to understand if these tiny creatures can learn odors the way Pavlov's dogs learned the association between food and the bell.

CUTICULAR HYDROCARBONS

These are special substances on insects' bodies that protect them from losing water and help them recognize each other.

LINALOOL

It is one of the most common floral perfumes.

FORAGERS

Females of social insects responsible for leaving their colonies to search for food in nature.

QUEENS

Females of social insects responsible for producing laying eggs which become new individuals in the colony.

USING PAVLOV'S METHOD FOR WASPS

We developed an approach in which a wasp received an odor stimulation (the neutral information) followed by sugar water (sucrose; the reward) delivered to their antennae. This made the wasp stick out its tongue to drink the sugar water (Figure 2C). We wanted to teach the wasps that the odor was associated with food. We used a floral odor called **linalool** as the neutral information. After presenting both the food and the odor, we expected the wasps to open their mouths and stick out their tongues in response to the odor alone (Figure 2D).

To test if wasps learn odors based on the association of those odors with food, we used **foragers**, a group of wasps responsible for collecting food for the nest and divided them into two groups. One group, called group A, was presented with the odor and the reward together (with the odor a few seconds before the sugar water). The other group (group B) was presented with the reward and the neutral information separated by more time, so there was no association between these things.

We found that only group A wasps could make the connection between the smell and food. How do we know that? When wasps make the connection, they stick out their tongues in response to the odor, which is exactly what we examined in Figure 3A. More wasps that received the reward and neutral information together exposed their tongues across the six times we did the experiment. In the group B wasps, we saw no evidence of the wasps sticking out their tongues in response to the odor. Further, only the group A wasps could create a memory of the information we taught them.

DO ALL TYPES OF WASPS IN A NEST PERFORM SIMILARLY?

Wasp nests are composed by different groups of adult individuals (e.g. queens, workers and males). Queens are responsible for the reproduction in the colony, workers take care of all the colony activities and males do not contribute for the social environment (they stay around for a while and then they leave the nest to search for queens). Beyond foragers, we wondered whether different types of wasps in the nest could learn the same way, so we did an experiment that included **queens** (females) and males in our investigation. Since we already knew that pairing the sucrose solution and the floral odor was necessary for wasps to learn and memorize the association, in this experiment we only used the condition in which the food and odor were given together. We found that queens and males also learned the association between the food and the odor (Figure 3B), and they held that information in memory for at least 1 h after the training. The fact that multiple types of wasps in the nest can learn and remember associations tells us that learning and memory are present

Figure 3

(A) We presented the wasps with the odor and the sugar water together six times ("trial" on x-axis), to see if they would learn to stick out their tongues in response to just the floral odor. In the first trial, no wasps stuck out their tongues because it was the first time they were learning the task. From the second trial onwards, the wasps that received the odor and sugar water together remembered the association and got better at it! (B) Both queens and males could also learn in this way. The "n" corresponds to the number of wasps tested.

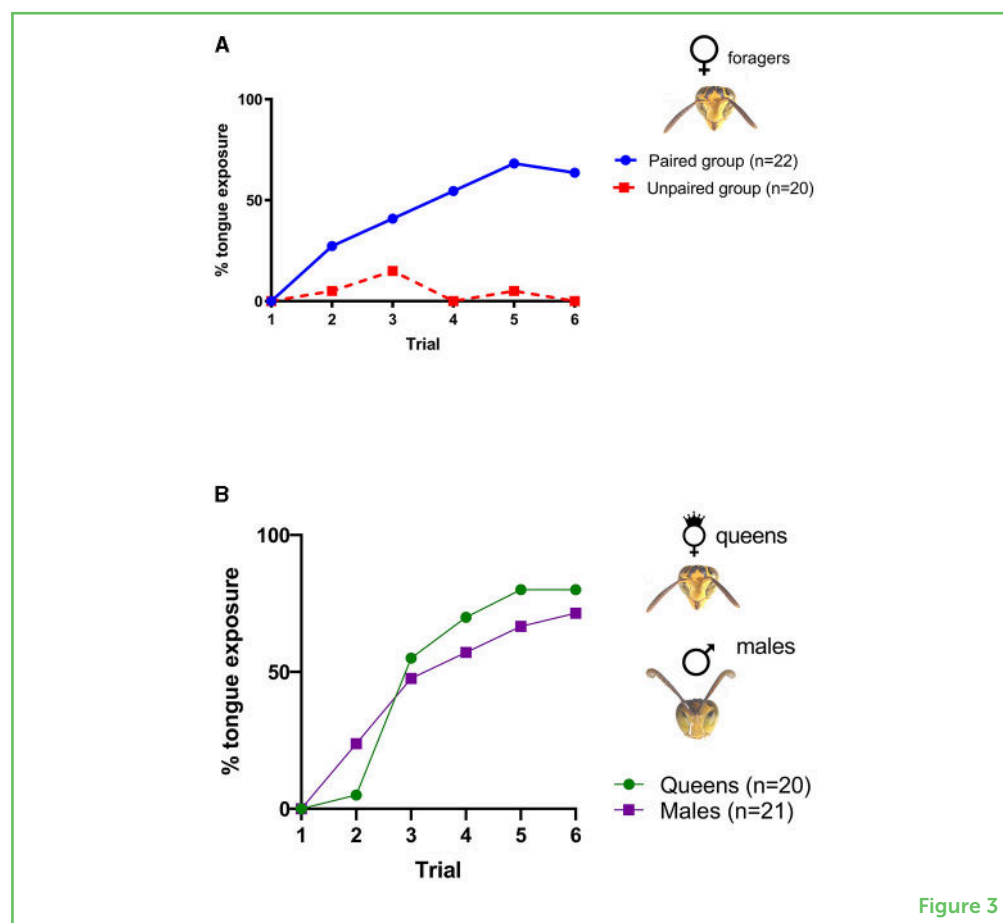


Figure 3

in this species and do not differ according to the role developed by different individuals.

LEARNING AND MEMORY IN A NATURAL CONTEXT

The ability to learn and memorize odors is important when wasps search for food. A wasp landing on a flower may learn the connection between a floral fragrance and **nectar** reward. When wasps experience both floral fragrance and the nectar at the same time, they learn to come back to the same flower in a second trip. Plus, since wasps can recognize each other *via* chemical messages, their ability to learn and remember might possibly help them to remember who does or does not belong to their nests, which may prevent their nests from being occupied by strangers.

CONCLUSION: TINY BRAINS, IMPORTANT FUNCTIONS

Although wasps have tiny brains, they have fascinating learning and memory abilities. In this article, we explained how different groups of wasps can learn *via* by associating neutral information (an odor) with something they like to eat (a sugary reward). They achieve this in the

NECTAR

This is a sweet liquid produce by flowers that attract insects and contribute for promoting pollination services.

same way described by Ivan Pavlov when he studied his famous dogs, which learned to salivate to the sound of a bell predicting food. In our experiment, the wasps learned to extend their tongues in response to an odor that predicted sugar water. Learning and memory abilities are present in both female and male wasps, and can help them in multiple aspects of their lives, from foraging for food to social interactions. Our results call attention to the fact that complex brain abilities, including learning and memory, are not limited to big brains but can also be found in tiny brains. Even though humans and insects are different, they share a common ancestor. By studying how insects learn, we can better understand our own brains and the complex activities they allow us to do every day.

ACKNOWLEDGMENTS

This study was financially supported by Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Finance Code 001 and grant 2018/22461–3 São Paulo Research Foundation (FAPESP) to RS. Funding was provided from Bilateral grant FWO-FAPESP to CO and FN (process numbers: 2018/10996–0 and 2021/05598–8 FAPESP and FWO: GOF8319N, FWO: GOF6622N), Research Foundation Flanders to CO (postdoctoral fellowship FWO-12V6318N and research grant FWO-1513219N), and Conselho Nacional de Desenvolvimento Científico e Tecnológico to FN (307702/2018–9). MG was supported by the Institut Universitaire de France and Sorbonne University. We would like to acknowledge the agencies that gave us the necessary resources to allow the study to be performed. RS currently holds a Fyssen Postdoctoral Fellowship (Fyssen Fondation, France).

ORIGINAL SOURCE ARTICLE

da Silva, R. C., Aguiar, J. M. R. B. V., Oi, C. A., Batista, J. E., Giurfa, M., and do Nascimento, F. S. 2023. Sex and lifestyle dictate learning performance in a neotropical wasp. *Iscience* 26:106469. doi: 10.1016/j.isci.2023.106469

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SUBMITTED: 24 November 2023; **ACCEPTED:** 02 July 2024;
PUBLISHED ONLINE: 17 July 2024.

EDITOR: Mubarak H. Syed, University of New Mexico, United States

SCIENCE MENTORS: Hengameh M. Taraz and Jill Dolata

CITATION: da Silva RC, Aguiar JM RBV, Oi CA, Batista JE, do Nascimento FS and Giurfa M (2024) The Tiny Brains of Wasps Can Learn and Remember Information. *Front. Young Minds* 12:1343838. doi: 10.3389/frym.2024.1343838

CONFLICT OF INTEREST: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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YOUNG REVIEWERS

ANIA, AGE: 15

My name is Ania. I am a high school student who loves to learn and is interested in a wide range of topics, including nature, physics, language, and theater. I love to read and write, and I have goals of working in editing or publishing as an adult. In my free time, I swim, bike, and spend time with my cats.

ON A BEAM OF LIGHT, AGES: 15–16

On a Beam of Light is a group of inquisitive, kind, and science-enthusiast young reviewers who meet to discuss new scientific findings and STEM related topics. We are writers, musicians, artists, and makers who love discovering the mysteries of the world.

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Rafael works with wasps and bees and is doing a postdoc at Sorbonne Université, in Paris, France. He was born in Brazil and, during his Ph.D., he went to Belgium and the United States to do research internships. In Belgium, he studied several wasp species, including yellowjackets and hornets. In the United States, he worked on zombie wasps. He likes insects, ice-cream, karaoke, going to the cinema, traveling, and spending time with friends. *rksilva2812@usp.br; rkswasp@gmail.com





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João received his doctorate in ecology from the University of Campinas, Brazil, and his doctorate in neurosciences from University Paul Sabatier—Toulouse III, France, both in 2019. Currently he is a researcher at the University of São Paulo, Brazil, where he trains bees, to investigate how they choose and memorize flowers during pollination. Outside of research, João loves cooking and long-distance running!



CINTIA AKEMI OI

Cintia works with wasps as a postdoc at University College London. She was born and raised in Brazil and did her Ph.D. at the KU Leuven (Belgium). There, she studied the evolution of queen pheromones and chemical communication in several wasp species. She loves her job and loves to combine fun and work. She collects wasps in amazing places including Brazil (the Amazon), New Zealand (invasive wasps), and the heathlands of the UK. Cintia enjoys insects, cuddling cats and dogs, traveling, cycling, and hanging out with her friends.



JAQUELINE ETERNA BATISTA

Jaqueline is studying the reproductive behavior and brains of two species of social bees and has a foot in the wasp world, thanks to living with her vespologist friends. In her Ph.D. in Brazil, she is studying the stingless bee known as Mandaguari. In her internship in Belgium, she worked on the stinging bumblebee known as Mamangava. Jaqueline is engaged in designing educational activities using bees as the central theme. As a Black young woman scientist, she will keep doing science and inspiring other people about the bee's world.



FABIO SANTOS DO NASCIMENTO

Fabio is a professor at the Universidade de São Paulo, Ribeirão Preto, Brazil. He is a teacher of animal behavior and he works on ants, bees, and wasps. During his master's degree, he worked on nocturnal wasps and during his Ph.D., he lived for a while in Rio Branco (Acre state – Brazil) to work with Amazon wasps.



MARTIN GIURFA

Martin is a French-Argentinean professor of neurosciences at Sorbonne University in Paris. He did his Ph.D. in Argentina and then moved to Berlin, Germany, where he spent 11 years until moving to Toulouse, France, where he founded a research institute dedicated to studying animal cognition. In 2023, he moved to Paris to become the director of the Biology Institute of Sorbonne University. Martin loves insects, neurons and brains, and is specialized in the topic of learning and memory. Outside the lab, he plays lead guitar in a rock band and loves to play in concerts.