Insights into the worm

The roundworm *C. elegans* is transparent and tiny. In the wild, it lives in compost. In the laboratory, it might help to replace elaborate animal experiments.

If you hold the Petri dish against the light, you can see it. Just. Tiny, whitish commas. Wispy fluff, a little longer than a millimetre. But this dust is alive. On closer inspection, we can see what is wriggling around: roundworms. Nematodes. They belong to the species *Caenorhabditis elegans*, *C. elegans* for short. In the laboratories at the German Centre for the Protection of Laboratory Animals in Berlin-Marienfelde they are being researched to find further alternative tests to conventional animal experiments. The centre is part of the BfR.

"The cool thing about the worms is that they are completely transparent," says project leader Dr. Silvia Vogl. A doctoral student puts a few worms under the microscope. Looking at the creatures twitching under the lens, you can immediately see what Silvia Vogl means. The elegant worm is fascinating. Its entire anatomy is visible, as if under an X-ray screen. The animal essentially consists of an intestinal tube that moves through its small world, such as compost heaps or decaying leaves, slurping bacteria. Its elongated body is filled with eggs at various stages of maturation. They are lined up like a string of pearls. The worm has 300 offspring in its three-week life span.

The principle of self-fertilisation

C. elegans conveniently self-fertilises its eggs. The worm is a hermaphrodite, a male-female hybrid. There are also pure males, but they are rare and make up only 0.2 percent of the population. It is actually "more an accident when they occur," says Dr. Vogl with a smile. They are still important because sexual reproduction "refreshes" the worm's genetic material and protects it from demise. Lungs, kidneys, liver, heart, eyes? Nothing. *C. elegans* doesn't need any of that. It has exactly 959 cells (the male 1031) and each one of these cells followed a predetermined development plan. Transparent, frugal, harmless, rapidly reproducing, inexpensive to keep, good to study – there are many reasons why the roundworm became the most widely researched multicellular organism. And also the first whose complete genome was deciphered in 1998.

And there's more. The worm casts a spell over scientists and never lets them go. "*C. elegans* researchers make up an international community," says Dr. Vogl. They meet every two years for the international conference. Almost all information needed for breeding is available online free of charge in the "WormBook". Innovative solutions to some worm problems are often published in the "Worm Breeder's Gazette". There is a bit of a wink, the research, however, is to be taken very seriously: to date, six scientists have received Nobel Prizes for their work with *C. elegans*.

The road to becoming the "model organism"

One of the Nobel laureates was British biologist Sidney Brenner. At the beginning of the 1960s, Brenner was looking for the simplest possible multicellular organism to study its development down to the smallest detail. He found what he was looking for in *C. elegans*. He began to study the animal in December 1963. Since then,

The roundworm can help to investigate the effects of chemicals

developmental biology, genetics, neuroscience and cell research have learned from and with *C. elegans*. The worm became a "model organism".

The well-connected and diverse scene of *C. elegans* research makes it easy for Silvia Vogl and her colleague Dr. Paul Wittkowski to work with the exact variant of the animal that they need for their questions. The approach of using roundworms to investigate the effect of potentially toxic substances is still new. Toxicology, the "science of poisons" – has so far found it somewhat difficult to accept *C. elegans* as a test animal.

Understanding exactly how a toxic substance works

Toxicologists traditionally work with laboratory animals like rats and mice, which, like humans, are mammals. This procedure is often required by law. But this area is changing. Experiments with cell cultures ("in vitro") or computer calculations ("in silico") complement conventional animal experiments ("in vivo"). Today we want to understand in detail how certain substances affect the organism, the cell or even individual genes. In addition, testing mixtures of substances, such as plant protection products, hormonally active substances or small amounts of toxic substances, poses a particular challenge. And finally, there is the 3Rs principle, which must be adhered to in science.

It describes the goal of reducing the number of animals in experiments ("reduce"), reducing suffering in experiments ("refine") or completely replacing animal experiments ("replace").

Enter *C. elegans*. It does not have all the organs that distinguish mammals. But the worm has far more in common with humans than those outside of the discipline might expect. The nematode has nerve cells, an (albeit simple) digestive tract, muscles and a reproductive system, as well as hormones and behaviour controlled by simple sensory stimuli. "The animal can even learn," says Silvia Vogl. "For example, to favour or avoid certain paths in a labyrinthine environment."

In addition, the genetic make-up of *C. elegans* has many similarities with that of mammals (such as humans). This concerns, for example, genes that are responsible for metabolism, communication between cells and detoxification. A short span between generations and rapid reproduction also help to make experiments faster and simpler than in conventional animal experiments. As a very small organism, *C. elegans* fills a gap between testing cells and experiments with vertebrates.

Fast test, lots of information

The BfR has been working with the roundworm for several years already. Paul Wittkowski developed an automated four-day test for *C. elegans*. This makes it possible to test many chemicals and mixtures of chemicals quickly and comprehensively. Using five different azole fungicides (substances that kill off fungi) as an example, Wittkowski studied how potentially toxic substances influenced growth and fertility of the worm. At the same time, he was able to determine whether residues of the azole fungicides had accumulated in the animal and which genes the substances had activated. Some reactions corresponded to those of mammals. In addition, the working group is testing chemical substances that have hormone-like effects in the body as an undesirable side effect.

In the future, the scientific community hopes that *C. elegans* will help to determine the mechanisms of action of chemicals. This approach may also reduce suffering because certain tests may no longer be necessary in conventional animal experiments. However, Silvia Vogl rules out that "exactly one worm experiment will replace one other animal experiment that is subject to approval". It is a bit more complicated than that. It is, however, conceivable that combining *C. elegans* tests with in vitro and in silico methods will work out. In any case, the worm has a future. ◘