The human brain is the most complex and remarkable organ of the body. It contains over a 100 billion nerve cells that have the unique capacity to communicate with each other through highly specialized contact spots, called synapses. Each neuron can form on average 10.000 – 100.000 synaptic contacts, which in the whole brain results in 10^{15} synapses. This is a thousand times more than there are stars in our entire galaxy. This enormous degree of connectivity is what makes the brain so exceptional and at the same time - the task of understanding it, so challenging. Moreover, this huge network is not fixed. On the contrary, it undergoes changes all the time as we learn new things and acquire new memories. As you are reading this, new connections are forming and strengthening in your brain to store the information contained in this text.

How much you will remember after reading this or any other text? It depends, among others, on whether it has any emotional value for you: whether it makes you feel good or bad. Learning what actions or events lead to positive or negative outcomes has a special function for a living organism. How to avoid pain and danger, where to find tasty food – these things are essential for our well-being or survival. Although we know a lot about brain structures engaged in these types of learning, many things are still unclear. How are these structures connected to each other? Which of them controls which, which one stores the memory and which one distinguished the positive from the negative? This is the sort of questions that we will try to answer in our research.

How can you see memory formation after it happened? Modern biology offers us amazing tools for that. For example, we know that a specific protein – c-fos – always appears after a neuron is activated by a new stimulus. In our institute we grew rats that have one tiny genetic modification: in these rats a gene encoding fluorescent protein is inserted into genome. This protein is driven by the same promoter as c-fos, so whenever c-fos is produced, a fluorescent protein is produced as well and we can see it under a microscope. If these animals learn something new – for example, that they will get food if they press a lever – some structures in their brains will produce both the c-fos protein and the additional fluorescent protein. This will show us in which structures took part in formation of the new memory.
Next, to find the connections between these structures we will use another tool developed by neurobiologists – viral neuronal tracing. Viral tracing relies on viruses that are modified in such a way that they are completely harmless to the animals. The only thing they do is produce fluorescent proteins that fill up the whole cell making it visible among all other cells in the brain. This way we can see which structures are connected to each other and in which order: which one sends and which receives the signals.

Single fluorescent neurons are way too small to see them with a naked eye or magnetic resonance. We need to use an optical microscope. Of course, looking at the brain under a microscope is not so easy: the brain is not transparent! Because of this we need to extract the brain from the skull and make it transparent using some chemicals. Afterwards we can cut it and using a fluorescence microscope we can simply scan the whole volume point by point, like we do it now with thin slices. However, if we want to see all the fine details, scanning of one brain point by point can take weeks or months, which is very unpractical. Because of this we build a special setup, called light-sheet microscope. In this microscope the laser beam that excites the fluorescence is formed in such a way that it illuminates the sample not point by point, but plane by plane: hence the name “light sheet”. Now, to investigate the entire three-dimensional volume, we only need to scan in one direction and not three, like in a conventional microscope. Right now, only a few labs in the world have such a device.

We believe that when we are finished, the process of formation of positive and negative memories in the brain will be a bit less of a mystery. In the meantime, we hope that some positive memories stayed in your brain after reading this.