

Questions to Prof. Wieland B. Huttner (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden) about the publication:

Michael Heide, Christiane Haffner, Ayako Murayama, Yoko Kurotaki, Haruka Shinohara, Hideyuki Okano, Erika Sasaki and Wieland B. Huttner: “Human-specific ARHGAP11B increases size and folding of primate neocortex in the fetal marmoset”, published by Science, via First Release, 18th June 2020.

Why did you generate transgenic monkeys in Japan for subsequent analysis?

Providing evidence that the human-specific gene *ARHGAP11B* can actually enlarge the developing neocortex of primates requires *ARHGAP11B* to be expressed in the fetal neocortex of non-human primates. Two non-human primate species, the rhesus monkey and the common marmoset, are appropriate for such studies. Our Japanese colleagues with whom we have collaborated are pioneers in the generation of transgenic common marmosets worldwide and therefore, the generation of the *ARHGAP11B*-transgenic monkeys for our study took place in Japan.

How are the animal welfare standards there?

The animal welfare standards in Japan are just as high as in Germany and are also laid down in a corresponding law.

Are there plans to study animals with this gene modification in later phases of life to investigate the effects on behavior?

Such experiments are currently neither planned nor in progress.

Why was self-restriction to fetal stages of development done and why was this ethically necessary from the point of view of the scientists involved?

Since a possible influence of *ARHGAP11B* on the behavior of the common marmosets could not be foreseen, a self-restriction to the fetal developmental stage was made. In addition, we are experts in the analysis of the neural cell architecture of the developing brain, but we have no expertise in the analysis of monkey behavior.

What would you have expected if the transgenic common marmoset had further developed into adult animals?

One cannot predict that.

Do you expect other scientists to move on now because they know that ARHGAP11B is important for neocortex development?

That, too, cannot be predicted. In any case, such experiments would need to be endorsed by a positive vote of the relevant ethics committee. Only then they may be carried out.

What ethical problems would you personally see in such a case?

From my personal point of view, I consider it ethically problematic to establish a transgenic monkey line with a human-specific gene, where this gene is passed on from generation to generation via the germ line. If postnatal or adult common marmosets, which express the human-specific gene *ARHGAP11B* in the neocortex, are to be studied for their behavior, such research should be restricted to single, repeatedly generated transgenic monkey individuals

rather than creating a transgenic monkey line. In our case we did not only limit our investigations to repeatedly newly generated transgenic common marmoset individuals. We also deliberately limited the analyses to the fetal stage to not get into a situation that could potentially be problematic from an ethical point of view.

What ethical requirements had to be met at the start of the project? What review processes were carried out at the CIEA and Keio University?

I would like to refer here to the Methods section of our publication: *“All animal experiments were approved by the Institutional Animal Care and Use Committee of the Central Institute for Experimental Animals (CIEA: 17029A, 18031A) and were performed in accordance with CIEA standard guidelines which are in accordance with the guidelines for the “proper conduct of animal experiments” determined by the Science Council of Japan.”*

Would such experiments be possible or allowed in Germany?

Yes, in principle such experiments could be carried out here as well, for example, at the German Primate Centre (DPZ) in Göttingen, provided that the necessary official permits have been obtained. However, at the time when we wanted to start our investigation, the relevant know-how at the DPZ was not yet available. That is why we have chosen to collaborate with Japan and in Japan.