Investigation of self-organisation and cryptogenesis ("budding") in intestinal organoids differentiated from porcine induced pluripotent stem cells

Methods:

- Differentiation of porcine induced pluripotent stem cells to intestinal organoids

Project description:
The pig serves as a popular intestinal model in biomedical research, as it is assumed that the human gastrointestinal tract is very similar to that of the pig. Nevertheless, there are differences that are difficult to clarify at the molecular level in animal experiments. Attempts to generate suitable permanent cell lines from the pig intestine have failed, with the exception of some small intestine lines. For some years now, induced pluripotent stem cells (iPSCs) can be reprogrammed from somatic adult cells. Human iPSCs (hiPSCs) can be differentiated into intestinal organoids specific to the intestinal segment. The advantage of the generation of intestinal organoids compared to classical cell culture is the intestinal segment specific formation of specialized epithelial cells (enterocytes, enteroendocrine cells, goblet cells, intestinal stem cells and in the small intestine also Paneth cells). Porcine iPSCs (piPCs) are also now available. In our group we have already succeeded in differentiating intestinal organoids from piPSCs. Gene expression data have been obtained during important differentiation steps in order to follow the progress of differentiation on a molecular level. In parallel, human intestinal organoids were generated using the same protocol. A typical course of gene expression of intestinal organoids could be traced. In comparison to human organoids, which showed a distinct cryptogenesis, indicating an organotypic arrangement of specialized epithelial cells, the porcine organoids remained in a spherical enterocyst form. It is known that different media must be used for the cultivation and differentiation of intestinal organoids from intestinal stem cells of different species. It is assumed that there are differences in the activation/inhibition of different signalling pathways, especially the Wnt signalling pathway. In this project, the influence of the so-called niche factors (Wnt3a, R-spondin and noggin), which maintain the stem cell niche in intestinal crypts, on cryptogenesis in porcine colonic organoids differentiated from piPSCs will be investigated. This should bring the existing porcine intestinal model closer to physiological states, positively influence the yield of emerging organoids and enable the long-term cultivation of these colon organoids. Finally, the intestinal organoids from pigs will be used in comparison to human intestinal organoids in order to study, for example, zoonotic intestinal diseases, which show different effects in humans and pigs, on a molecular level.

Funding:
Doctoral scholarship of the H. Wilhelm Schaumann Foundation

Project Lead:
Prof. Bettina Seeger, PhD (bettina.seeger@tiho-hannover.de; 0511-856-7602)
Institute for Food Toxicology, University of Veterinary Medicine Hannover, Foundation, Germany