The genetic, physiologic and anatomic similarity to humans in combination with short reproduction times makes the mouse the most important model organism for studying the genetic regulation of developmental processes and the genesis of congenital diseases. The International Mouse Phenotyping Consortium (IMPC) plans to generate mouse strains carrying a deletion of each of the approximately 20,000 genes of the mouse genome. Individuals of the strains are phenotyped for gaining information of the function of the out-knocked gene. However, first results suggest that about one third of all knock out strains will produce homozygous offspring, which die during embryogenesis or in the perinatal period. Thus the “Deciphering Mouse Developmental Disorders” (DMDD) project was launched that aims at providing phenotype information of embryos of such strains. In this presentation we will present pilot data from the DMDD project obtained by employing the “High resolution episcopic microscopy” (HREM) method for phenotyping E14.5 embryos.

HREM is a post-mortem three-dimensional (3D) imaging technique based on digital images, which show physical sections through histologically embedded and sectioned specimen. We fixed a total of 92 knock out embryos in Bouins fixative, dehydrated them in ethanols, and embedded them in plastic resin dyed with eosin. Using a microtome the blocks were cut in 3 µm thick sections. During sectioning, images of the block surface were captured after each cut. For this a microscope equipped with a GFP-filter set and a digital camera was aligned with the photo-position of the microtome. Since the block comes to rest at this position after each cut, a series of aligned digital images, which consisted of 2,000 to 3,500 single images was created. This series was converted into a volume data set with a voxel size of 3 x 3 x 3 µm3. The quality of single HREM images nearly matches the quality of images captured from histological sections. In our pilot study, we developed a protocol, that is based almost solely on such sections. It enabled us to diagnose macroscopic malformations, as well as subtle, but potentially lethal tissue defects, which would escape their diagnosis with alternative 3D imaging techniques. Our results demonstrate that HREM is an optimal technique for phenotyping E14.5 mouse embryos with gene deletions.
Fig. 1: Figure: Sagittal virtual resection through HREM data of an E14.5 mouse embryo.