Effects of transit time on bifidobacteria correlation patterns

The described work has been performed during a two-month stay at the University of Reading (UK) under the supervision of Dr. Anne McCartney. The initial aim of the visit was to address to what extent gut transit time can explain the observations of correlations between bifidobacteria species observed previously (Avershina et al., 2012). This project was planned as a part of a bigger study which addresses the factors contributing to the species correlations observed in a large healthy population of mothers and their children. However, due to time constraints, we decided to test the differences in substrate preferences between selected isolates instead of transit time effects.

Eighteen stool samples from newborns, 4-month-olds, 1- and 2-year-olds and their mothers during pregnancy, were selected from a large study cohort. Dilutions (10^{-1} to 10^{-7}) of selected stool samples were cultured on bifidobacteria-selective medium. In case of positive growth, around ten individual colonies per sample were picked, ensuring that each colony-type was represented by at least one colony. In total, 128 isolates were subcultured from the stool samples. Subsequently, DNA analysis confirmed that 93 of these isolates belonged to the genus *Bifidobacterium*. We subsequently tested the growth of eight of the bifidobacterial isolates on various carbon-source substrates. Isolates were selected based on the DGGE profiles of 16S rRNA gene, and they belonged to a) two mother and child pairs; b) 3-days-old child and c) child at 4 months and at 2 years. The following sugars were used for the experiment: a) inulin; b) Vivinal® GOS; c) HMO powder; d) glucose; and e) lactose. As a result, we established growth curves and carbon-source preferences of these isolates. All tested isolates showed strong growth on HMO and GOS, whereas only one isolate (from a pregnant woman) grew well on inulin. Isolates from a newborn and 4-month-olds seemed to be more effective in utilizing HMO than those from 2-year-old and pregnant women. Interestingly, the isolate from a 3-day-old baby was the least flexible in substrate utilization, exhibiting growth only when HMO or glucose was used.

In the future, we plan to continue collaboration with Dr. McCartney's lab with regards to characterization of obtained bifidobacterial isolates. Our lab will be focusing on the molecular aspects of the study, whereas Dr. McCartney's lab will be responsible for cultivation work. As mentioned before, the performed work is part of a bigger study addressing the mechanism behind the described correlations, and it will result in a publication within a PhD study. The

proposed title for the prospective publication is ‘Effects of oxygen tolerance, diet preferences and gut transit time on succession of bifidobacteria in a population’.