Hirschsprung disease - genetics and development

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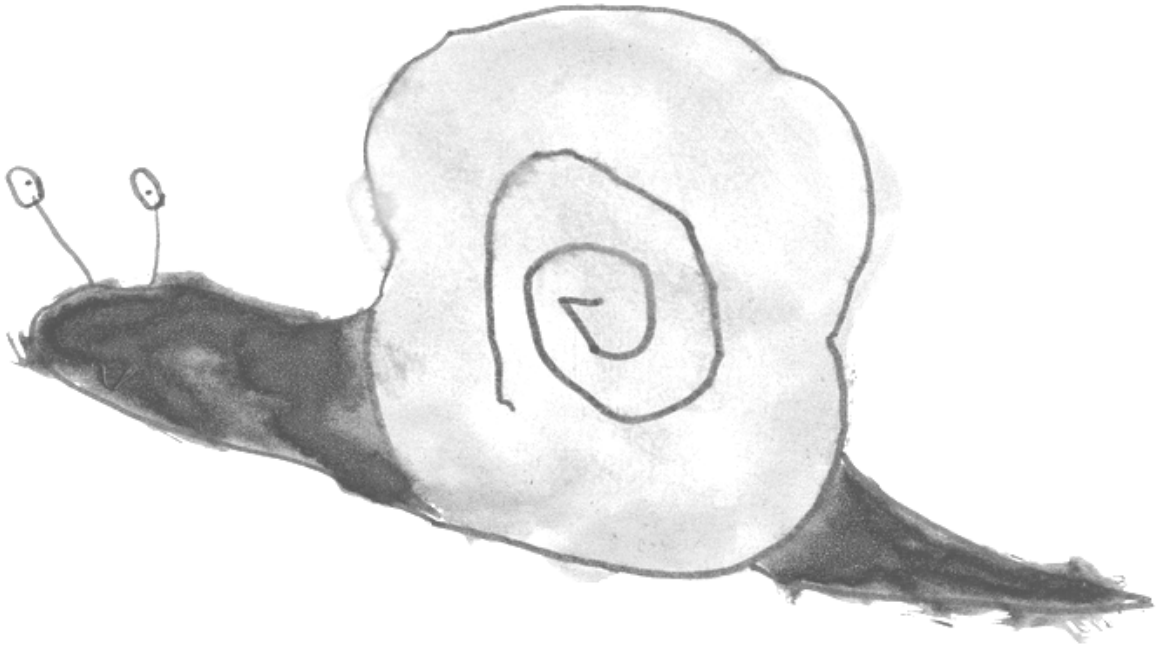
Scope

Hirschsprung disease (HSCR) is a complex genetic disorder that is characterized by the lack of the Enteric Nervous System (ENS) in the myenteric and submucosal plexuses of the gastrointestinal tract. HSCR is a neurocristopathy and can result from aberrant migration, proliferation or differentiation of vagal neural crest cells. The length of the affected intestine classifies HSCR. The most common form of the disease (~80% of patients) is called short-segment HSCR (S-HSCR), where aganglionosis does not extend beyond the sigmoid colon. Approximately 20% of the patients is diagnosed with long-segment HSCR (L-HSCR), in which aganglionosis extents proximal to the sigmoid. The population incidence is generally assumed to be 1/5000 live births, though it varies among patients of distinct ethnic origin. A sex bias is also present in this disorder as approximately three times more males are affected. The pattern of inheritance for L-HSCR is autosomal dominant with low penetrance whereas the inheritance of the much more common S-HSCR is believed to be multifactorial. The major gene in HSCR is *RET*. Nevertheless, mutations in the coding sequence of *RET* are hardly being found in the main classes of patients.

The work included in this thesis had two main goals. Firstly, to elucidate further the role of *RET* in those cases where no coding sequence mutations were found. We focused on the most common group of patients, namely those with S-HSCR who lack a disease history in the family. We found, that the *RET* gene is involved in nearly all HSCR cases and that the majority of mutations are not lying within the coding regions, but in the regulatory sequences of the *RET* locus.

Secondly, baring in mind that HSCR is genetically complex disease, we attempted to find new genes and loci involved in HSCR development and more generally genes encoding proteins,

which contribute to ENS development. In a collaborative effort with the Rotterdam group a new HSCR susceptibility locus was identified in a multigenerational HSCR family. Further, ENS precursors cells, isolated from mouse embryos' intestines were studied and expression profiling was performed. Genes encoding proteins belonging to the RET pathway or genes encoding proteins affected by RET signaling in these cells, were identified. By this approach we hope to get a better insight in ENS development, and HSCR susceptibility.



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Abstract

Hirschsprung disease (HSCR) is a developmental disorder characterized by the absence of enteric neurons in myenteric and submucosal plexuses in the distal parts of gastrointestinal tract. Any failure occurring on the level of migration, differentiation, proliferation or interaction with surrounding tissue can lead to a HSCR phenotype. Since the precursors of the enteric neurons are a sub-population of cells derived from the neural crest that migrate in rostro-caudal direction in the gut HSCR is considered a neurocristopathy.

The approximate incidence of HSCR is 1/5000 for the world population. In 90% of the cases HSCR occurs in a single patient in a family (simplex) and in 70% HSCR occurs as an isolated disease trait. In the remaining 30% cases HSCR is accompanied by other congenital anomalies, which in part are due to chance, can be a consequence of a chromosomal abnormality associated with HSCR or are due to a mutation(s) giving rise to a more complex (syndromic) phenotype.

HSCR is considered a genetic disease, it is caused by genetic alterations. Until now mutations in 10 genes and allele sharing with 4 loci have been identified. as a risk factors of developing HSCR. Of the genes identified *RET* proved to be the major genetic risk factor. In the common simplex cases, however, no more than 15% of patients possess *RET* coding mutations, but most patients do show haplotype sharing at the *RET* locus. Mutations in the other 9 genes are rare and mostly found in the syndromic HSCR cases. Judging from the low, sex-dependent penetrance of HSCR, its variable expression and multiple susceptibility loci discovered in linkage and association studies, isolated HSCR should be considered as an oligogenic disorder. We review

the most recent findings in HSCR genetics as well as the developmental conditions of the disorder, underlining the importance of all processes that precursors of enteric neurons need to undergo before successfully colonizing the gut. We discuss the signaling pathways involved and their spatial and temporal cross talks that enable the development of enteric innervation. This review links results from genetic studies and developmental knowledge and considers research strategies that take into account their interplay.