7 Abstract

Assembly of the tight junction is a key event in the differentiation of polarized epithelial cells. Claudins are transmembrane proteins and an integral part of the tight junction. Claudin-1 is a barrier protein, whose essential role in the sealing of the paracellular space has already been shown in native skin epithelium. Claudin-2 on the other hand plays a crucial role for the paracellular barrier because of its ability to form cation selective pores in the tight junction. Physiological and pathophysiological changes of the epithelial barrier are accompanied by changes in the expression of tight junction proteins.

This work deals with the examination of molecular mechanisms in the regulation of claudin-1 and claudin-2 gene expression. Genomic DNA containing promoter sequences of human claudin-1 and claudin-2 were functionally characterized by reportergene assay. Promoter activity of claudin-1 and claudin-2 was elevated in a murine epithelial cell line, which expresses Wnt-1. The secretory glycoprotein Wnt-1 is an extracellular activator of the Wnt signaling pathway, which regulates cell differentiation and proliferation. For LEF-1, a nuclear effector of the Wnt signaling pathway, a direct interaction with specific binding motives in the promoters of claudin-1 and claudin-2 was shown. The expression of LEF-1 and β-catenin resulted in an elevation of claudin-1 and claudin-2 promoter activity. Mutation of the LEF-1 binding sites led to a diminution of this activation. Another nuclear effector of the Wnt signaling pathway, TCF-4, showed also an elevation of claudin-1 and claudin-2 promoter activity. This suggests, that the Wnt signaling pathway regulates gene expression of the human tight junction proteins claudin-1 and claudin-2. Another important result was the verification of a functional crosstalk between Wnt signaling pathway and caudal related homeobox protein (Cdx) mediated transcriptional activation of claudin-2 promoter activity. The regulation of claudin-2 gene expression is therefore subjected to complex interactions of different signaling pathways.

The differential configuration of diverse epithelia with varying tight junction proteins is crucial for different paracellular barrier properties. The regulation of this differential expression takes place in the phases of cellular differentiation and polarisation. This work could demonstrate, that the Wnt signaling pathway and the Cdx mediated

transcriptional regulation, two important signaling pathways for differentiation and polarisation, are involved in the regulation of claudin gene expression.