Seminars in Cell & Developmental Biology 36 (2014) 166–176

Contents lists available at ScienceDirect

Seminars in Cell & Developmental Biology

journal homepage: www.elsevier.com/locate/semcdb

Review

Tight junction, selective permeability, and related diseases

Susanne M. Krug, Jörg D. Schulzke, Michael Fromm*

Institute of Clinical Physiology, Campus Benjamin Franklin, Charité – Universitätsmedizin Berlin, 12203 Berlin, Germany

ARTICLE INFO

Article history:
Available online 16 September 2014

Keywords:
Paracellular channel proteins
Claudin
TAMP
Inflammatory bowel diseases
Nonsyndromic deafness, FHHNC

ABSTRACT

The tight junction forms a barrier against unlimited paracellular passage but some of the tight junction proteins just do the opposite, they form extracellular channels zigzagging between lateral membranes of neighboring cells. All of these channel-forming proteins and even some of the barrier formers exhibit selectivity, which means that they prefer certain substances over others. All channel formers exhibit at least one of the three types of selectivity: for cations (claudin-2, -10b, -15), for anions (claudin-10a, -17) or for water (claudin-2). Also some, but not all, barrier-forming claudins are charge-selective (claudin-4, -8, -14). Moreover, occludin and tricellulin turned out to be relevant for barrier formation against macromolecule passage. Tight junction proteins are dysregulated or can be genetically defective in numerous diseases, which may lead to three effects: (i) impaired paracellular transport e.g. causing magnesium loss in the kidney, (ii) increased paracellular transport of solutes and water e.g. causing leak-flux diarrhea in the intestine, and (iii) increased permeability to large molecules e.g. unwanted intestinal pathogen uptake fueling inflammatory processes. This review gives an overview on the properties of tight junction proteins featuring selective permeability, and in this context explains how these proteins induce or aggravate diseases.

Contents

* Corresponding author. Tel.: +49 30 8445 2535.
E-mail address: michael.fromm@charite.de (M. Fromm).

http://dx.doi.org/10.1016/j.semcdb.2014.09.002
1084-9521/

1. Introduction

Epithelia form barriers against unlimited passage of solutes and water, but also regulate and allow distinct permeation across that barrier. On the one hand, such permeation sites are located within the cell membranes, forming a transcellular pathway via channels, carriers, and transporting ATPases. On the other hand, the paracellular pathway between the cells is sealed against uncontrolled passage by the tight junction (TJ). However, long before claudins and TAMPs (tight junction-associated Marvel proteins) were identified as constituents of the TJ it was demonstrated that the paracellular pathway of some, but not all, epithelia is permeable to small ions [30]. This in mind, the concept of "leaky" and "tight epithelia" was born [24]: in leaky epithelia the paracellular pathway is more ion-conductive than the transcellular one. In intestine and nephron, leaky epithelia are typically found in proximal segments. Tight epithelia behave the other way around and in intestine and nephron they are present in distal segments.

While many TJ proteins indeed have barrier-forming properties, there are also several claudins forming charge- and/or size-selective paracellular channels. These channels are not crossing membranes as transmembranal channels do, but are orientated parallel to the lateral membranes allowing permeation through the TJ. They are formed by the extracellular loops of TJ proteins interacting with extracellular loops of TJ proteins located in the opposing cell membrane.

Often there are uncertainties whether the conductive claudins should be named channels or pores. Simply said, both is correct: the pore is one part of a channel. A channel is the entity of a permeation site comprising (i) a pore, (ii) a narrow site that restricts access by size and shape (size selectivity), (iii) a site that favors passage by charge or charge density (charge selectivity), and (iv) a feature providing time-variant permeability changes (gating).

By definition, "selective for *x*" means that the permeability for *x* is higher than that for other substances or groups of substances. All channel-forming claudins exhibit at least one of the three types of selectivity: for cations (claudin-2, -10b, -15), for anions (claudin-10a, -17) or for water (claudin-2).

Charge selectivity cannot be determined from transepithelial resistance data but from dilution potential measurements. Here, charge selectivitiy is read out from the resulting ratio P_{Na}/P_{Cl}. $P_{Na} > P_{Cl}$ indicates cation selectivity and $P_{Na} < P_{Cl}$ indicates anion selectivity [42,138]. Ratio changes together with the calculated absolute permeabilities give information about the preference. Higher selectivity, as e.g. exclusively for Na^+ only can be found in some membrane channels like the epithelial sodium channel ENaC, but yet not for any TJ protein.

Thus, TJ protein channels formers and also barrier formers exhibit substrate-specific transmissive properties. Therefore, the term "permeability" is incomplete without relying to the analyzed substance(s) for which the TJ protein is transmissive.

It is still unknown, how exactly claudin-based paracellular channels are built and it is not even resolved how many claudins assemble to form one channel within the tight junction meshwork. Several specific effects on paracellular selectivity based on the 27 mammalian claudins, their splice variants and on TAMPs [97], have been described so far.

TJ proteins are dysregulated or can be genetically defective in numerous diseases of epithelial organs. Various TJ proteins are involved in tumor formation and progression. However, as there is so far no clear relation between tumors and claudin selectivity, a detailed discussion of this aspect is beyond the scope of the present review.

In summary, TJ protein-related diseases may lead to three general effects:

(i) Reduced paracellular transport of solutes. As an example, in familial hypomagnesemia, hypercalciuria, and nephrocalcinosis (FHHNC), a genetic defect of claudin-16 or -19 indirectly impairs the reabsorption of Mg^{2+} and Ca^{2+} in the kidney (see Section 2.4) [111].
(ii) Increased paracellular transport of solutes and water. This takes place e.g. in intestinal inflammatory diseases, where claudin-2 is upregulated (see Sections 2.1 and 4.1). As a consequence, ions and water diffuse from blood to lumen and result in a distinct form of diarrhea, called leak-flux diarrhea [104].
(iii) Increased permeability to large molecules. Also typical for inflammatory bowel diseases is the development of significant uptake of luminal pathogens (e.g. food antigens and bacterial lipopolysaccharides) which may initiate the immune response and cause or maintain the inflammatory process [2,19].

Regarding (ii) and (iii), the TJ dysfunction would represent both reason and consequence of the disease.

2. Cation-selective channel formers

2.1. Claudin-2

Claudin-2 was the first TJ protein for which a decrease instead of an increase of the transepithelial resistance (TER) was shown [35]. For this, claudin-2 was introduced into a renal cell line, MDCK I, which is genuinely devoid of claudin-2 and develops TER values as high as 10 kΩ cm^2. After transfection with claudin-2-cDNA TER reached only 150–500 Ω cm^2.

A crucial step was then to find out for which solutes claudin-2-containing cells are permeable. It turned out that claudin-2 mediates permeability only for small cations like Na^+ and K^+, but not for anions and for molecules larger than 180 Da. From this it was concluded that claudin-2 forms cation-selective channels in TJs [4].

In order to characterize the molecular basis of that cation selectivity, electrostatic interaction sites were identified. Charge selectivity was mediated by the electrostatic interaction of partially dehydrated permeating cations with a negatively charged site within the pore that is formed by the side chain carboxyl group of aspartate-65 [138]. For further details of that analysis, see Section 4.1.

Claudin-2 proved to be of great clinical impact. Its abundance increases in various diseases such as Crohn's disease, ulcerative colitis, and celiac disease.

In Crohn's disease or after stimulation of epithelial cells with the pro-inflammatory cytokine TNFα, claudin-2 is found elevated in the intestine while the barrier formers claudin-5 and claudin-8 were distributed off the TJ domain into a basolateral membrane compartment and into subapical vesicular compartments (most likely endosomes). In result, the permeability for cations was increased, pointing to a driving mechanism of leak flux diarrhea [139]. Freeze fracture electron microscopy revealed a decreased number of horizontal TJ strands, a condensed strand meshwork, and the appearance of strand discontinuities. These discontinuities were interpreted as a possible pathway for the uptake of macromolecules (see Fig. 3, right) [139].

In ulcerative colitis, human colon biopsies exhibited a ten-fold increase of claudin-2 and in the colon cell line HT-29/B6 the proinflammatory cytokine IL-13 was identified as an important effector cytokine which impairs epithelial barrier function by three processes, increased epithelial apoptosis, increased claudin-2 expression, and reduced epithelial restitution velocity [44].

In celiac disease, duodenal biopsies revealed increased expression of claudin-2 and -15 as well as a reduction in claudin-3, -5 and