Reviewer’s report

Title: Small intestinal mucosa expression of putative chaperone fls485

Version: 1 Date: 8 July 2009

Reviewer: Mark Musch

Reviewer’s report:

The manuscript of Reinartz et al investigates the expression of a chaperone protein fls485 in intestinal epithelia. This gene product has received little attention, however, it possesses many domains that suggest it could be an important regulator of cell growth and or differentiation. An important facet of the studies is the generation of antibodies that allow assessment of protein expression. The authors determine that this gene product is expressed in intestinal epithelial cells in both crypt and villus enterocytes, but the expression is higher in the villus cells, consistent with a potential role in differentiation. Fls485 expression was also found in some cells of the lamina propria, supporting this its expression is not confined to epithelial cells. The investigators also demonstrate expression in a number of cultured intestinal epithelial cell lines. Fls485 is determined to be a cytosolic protein and the expression is decreased in celiac disease. The studies are potentially interesting and fls485 has the probability of being an important regulatory protein in the epithelial cells, however, at present the impact of the studies is limited since a functional role has not been established.

Major Compulsory Revisions

(1) The investigators have the opportunity to determine in fls485 may have a role in differentiation using an established cultured in vitro cell line, Caco2. This cell line differentiates post-confluence in culture. Expression of villin, cysteine-sensitive alkaline phosphatase, and sucrase increase after Caco2 cells come to contact inhibition of growth. While it may be presumptive to state that this is precisely representative of maturation of villus small intestinal enterocytes, it is a model that is well accepted. It is stated that this cell line expresses fls485 and the investigators have the expertise to determine if fls485 expression increases as these cells mature. This investigation should also include studies of the maturation to determine if one or more of the differentiated characteristics are regulated by fls485. The investigators could extend these studies using RNA silencing technologies to determine whether fls485 is necessary for maturation or simply expression parallels maturation of other markers.

(2) Another point that could be presented more clearly is the reason for investigations of celiac disease and not other intestinal pathologies. Fls485 expression might be anticipated to be up or downregulated by a number of conditions such as bacterial infections by Salmonella or viral infection by rotavirus. These two conditions may be difficult to obtain, however, through the
connection to the Pathology department, the investigators may be able to obtain sections of inflammatory bowel diseases, both pediatric and adult. It is appreciated that immunohistochemistry is a poorly quantitative technology however, to obtain fresh tissues for Western blotting and better quantitation would be excellent, but perhaps beyond the scope of the present studies. Another condition that might regulate fls485 expression is colon cancer where the importance of other chaperone proteins is being established as biomarkers as well as in the pathogenesis of the disease. Sections from a Pathology bank could well provide important information that allow further investigation of the roles of this protein.

Minor Essential Revisions

(1) For the studies of intestine, it would be good to also include a longitudinal study of fls485 expression. Sections of stomach, duodenum, jejunum, ileum, cecum, proximal and distal colon are readily obtained from animal models and perhaps the investigators have access to a pathology bank of preserved tissues. For these sections, it would be interesting and potentially important to establish which epithelial cells in each segment express fls485. By itself this survey is not sufficient for publication, but it would add to the initial investigations of this gene product.

Level of interest: An article of importance in its field

Quality of written English: Acceptable

Statistical review: Yes, and I have assessed the statistics in my report.

Declaration of competing interests:

I declare that I have no competing interests