ANNALS OF GASTROENTEROLOGY 2009, 22(3):138-139

Editorial

Ethanol effects on mucin glycosylation: Another kick in the gut?

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In this issue, Grewal and Mahmood [1] report new findings concerning the effect of alcohol in the small bowel. They found altered composition of mucins in rats fed with ethanol for 4-8 weeks and they suggested that these alterations may be of pathological significance, since mucins are involved in protection and adhesion of microorganisms in intestinal lumen. This information comes to add another harmful weapon to the quiver of alcohol against the mucosal integrity and function of the gastrointestinal tract.

The effect of alcohol on the gastrointestinal tract

Alcohol has been known for a long time to interfere with the absorption of several nutrients, including vitamins, and to lead to mucosal damage of the upper small intestine, thereby contributing to the qualitative and quantitative malnutrition frequently observed in alcoholics. Acute alcohol exposure leads to mucosal damage that extends to loss of epithelium at the tips of the villi, haemorrhagic erosions and haemorrhage in the lamina propria.[2] In accordance with these findings are the results of a large, prospective case–control study that provided evidence that consumption of alcoholic beverages significantly increases the risk of major duodenal bleeding in non-predisposed individuals.[3] The mechanisms causing these pronounced structural changes include an increased influx of leukocytes leading to an enhanced release of noxious mediators, such as reactive oxygen species, leukotrienes and histamine, accompanied by changes in the mucosal microcirculation that leads to an enhanced transcapillary fluid filtration and subsequent rupture of the epithelial lining.[2]

Furthermore, marked qualitative and quantitative alterations of the jejunal microflora have been documented in recently drinking alcoholics. Studies in alcoholics using the H2-breath test after ingestion of glucose or lactulose have demonstrated increased prevalence of bacterial overgrowth. Bacterial overgrowth in the upper small intestine might contribute to mucosal damage and disturbed absorption in subjects chronically abusing alcohol. Furthermore, it might increase the production of bacterial toxins, especially endotoxins from Gram-negative bacteria which, in combination with the mucosal injury induced by alcohol, might contribute to an enhanced endotoxin translocation from the luminal side into the portal blood (Figure 1).[2]

Part of this phenomenon could be attributed to changes in gastrointestinal mobility caused by chronic alcohol consumption. Orocaecal transit time (OCTT), as assessed by the lactulose H2-breath test was significantly delayed in alcoholics in comparison to healthy controls or social drinkers.[4] Disturbances of the synthesis of smooth muscle contractile proteins in the small intestine, observed in rats receiving ethanol, might contribute to this altered motor function.[2]

In addition, significantly increased intestinal permeability in actively drinking alcoholics has been confirmed for macromolecules such as polyethylene glycol (PEG) 4.000 Mr and 10 000 Mr.[5] Interestingly, the enhanced intestinal permeability for these macromolecules had already been found in alcoholics without advanced stages of liver disease, a finding that strongly supports the assumption that the increased permeability of the gut mucosa is caused by ethanol itself and is not a consequence of advanced alcoholic liver disease. The hypothesis that the acute and chronic ingestion of large quantities of alcohol enhances

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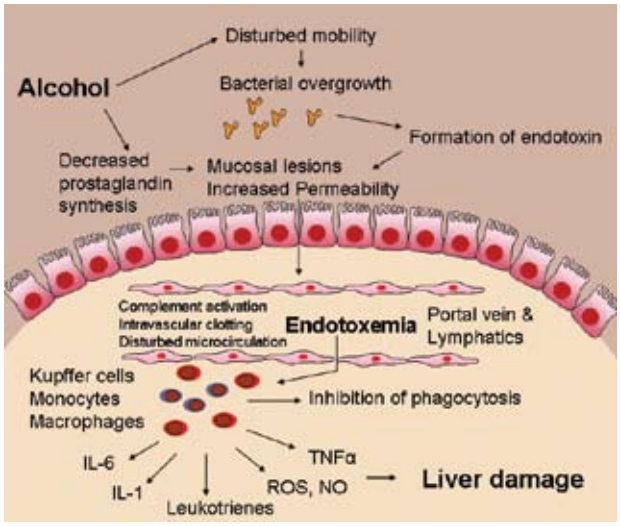

Figure 1. Common ethanol-related effects on the gut are (i) mucosal damage of the upper small intestine, thereby contributing to the qualitative and quantitative malnutrition frequently observed in alcoholics, (ii) alterations in the bacterial flora and (iii) increased gut permeability possibly resulting in endotoxemia and subsequent liver damage.

the translocation of macromolecules through the intestinal mucosa is supported by the observation of a transient endotoxaemia following acute alcohol consumption in healthy volunteers and in alcoholics with fatty liver [5]. Direct evidence of enhanced translocation of endotoxin from the gut lumen into the portal blood has been achieved in experiments using the Tsukamoto-French model in rats.[6]

The intermittent endotoxaemia stimulates Kupffer cells and other macrophages in the liver, thereby enhancing the release of reactive oxygen species and proinflammatory mediators such as tumour necrosis factor-a (TNF-a), interleukin-1 (IL-1), IL-6, reactive oxygen species and nitric oxide (NO).[7] We have previously demonstrated a systemic increase in NO, possibly as a result of iNOS up-regulation after a single administration of 80g of ethanol in healthy volunteers[8] Chronic overproduction of such mediators following chronic alcohol abuse may induce the influx and activation of neutrophil leukocytes, endothelial lesions, increased permeability of sinusoids, disturbed microcirculation and other injurious events that finally lead to severe forms of liver injury and other organ damage (Figure 1).[2, 7]

The role of mucins

In this issue, Grewal and Mahmood [1] explore a less known effect of alcohol on the quantity and composition of the small bowel mucins. Mucins form a thick adherent and unstirred layer of mucus gel of median thickness of 180 micron in stomach in humans, which lines the gastrointestinal tract and acts firstly as a lubricant and secondly as a protective physical barrier between the mucosal surface and the luminal contents. The mucins are huge molecules, typically with a molecular mass of the order of 1–20 x 10^6 Daltons. They consist of a central protein backbone with large numbers of attached oligosaccharides. Common to all mucin protein cores that have so far been identified are tandem repeat sequences (VNTR). These are sequences of amino acids which are repeated and contain a high proportion of serine and threonine, the attachment sites for O-linked oligosaccharides. Glycosylation accounts for up to 60%–80% of the mass of the molecule, and is responsible for many of the properties of mucins.[9]

Since the first experimental evidence for the mucus bicarbonate barrier was reported nearly three decades ago by Allen and coworkers, mucins have become firmly established as a key component of the gastroduodenal mucosal protective mechanisms.[10] The secretion of HCO_3^- into a stable, adherent mucus gel layer creates a pH gradient from acid in the lumen to near neutral at the epithelial surface in the stomach and proximal duodenum and provides the first line of mucosal defence against luminal acid. This mucus gel layer is also the major physical barrier that prevents the back-diffusion of luminal pepsin thus preventing the proteolytic digestion of the underlying epithelium. Despite its well established physiological role in the proximal gastrointestinal tract reviewed by Allen and Flemstrom, the role of mucin in small and large bowel has not been adequately evaluated.[11]

The effect of alcohol on small bowel mucins

In this issue of the Annals of Gastroenterology, Grewal and Mahmood [1] show that chronic alcohol feeding increases mucin production in the small intestine of rats. The observed effect remains questionable as it has not been validated by the demonstration of an increase of the thickness of the small bowel mucus layer in situ using either conventional or intravital microscopy. The increased thickness of the mucin bands demonstrated by SDS-PAGE electrophoresis could be due to alcohol-induced qualitive changes that allow more mucin to be detached from the bowel wall in the case of alcohol-treated animals. Alternatively, alcohol might cause a true increase in mucin production by intestinal goblet cells but whether this is a compensatory response to alcohol toxicity or a direct result of the alcohol effect on goblet cells remains unknown.

The undisputable fact demonstrated by the authors is the qualitive changes in mucin composition induced by

alcohol. Specifically, alcohol seemed to reverse the fucose/sialic acid molar ratio of oligosaccharide residues from a predominance of fucose to a predominance of sialic acid. This was attributed alcohol-induced changes in sialyltransferase and fucosyltranferase activities at the intestinal brush borders. Furthermore, analysis of mucins isolated from intestinal lumen of control and ethanol fed animals for alkaline phosphatase and sucrase revealed the enzyme activities were augmented in ethanol fed animals compared to controls. It is tempting to hypothesize that such changes may have an effect on the viscoelastic properties of the gels formed and influence interaction of mucins with intraluminal bacteria. Although the authors have not pursued this hypothesis and have not demonstrated an effect of these qualitive changes on intestinal permeability or bacterial translocation there seem to be some relevant experimental data that associate similar qualitive changes in colonic mucins with the development of inflammatory bowel diseases.

Specifically, altered biochemical properties of colonic mucin such as decreased of the oligosaccharide chain length, reduced sulphation and increased sialylation have been observed in IBD patients.[9] Sulphation and sialylation are important biochemical determinants as they play a part in the resistance of mucins to bacterial degradation. Furthermore, it has been shown that there is increased mucin sulphatase activity in ulcerative colitis that mirrors disease activity.[12] Additionally, an association has been reported between rare alleles of the MUC3 gene, one of the nine genes MUC genes which code for the protein cores of mucins, and ulcerative colitis.[13] In Crohn's disease, a reduction in MUC3, MUC4, and MUC5B levels has been observed in both diseased and normal tissue compared to controls, which implies an early or primary mucosal defect related to disease pathogenesis[14] The strongest evidence in favour of a pathogenetic role of mucins in IBD comes from a recent study in mice lacking core 3 í1,3-N-acetylglucosaminyltransferase (C3GnT), an enzyme predicted to be important in the synthesis of core 3–derived O-glycans. C3GnT-deficient mice displayed a discrete, colon-specific reduction in Muc2 protein and increased permeability of the intestinal barrier. These mice were found to be highly susceptible to experimental triggers of colitis and colorectal adenocarcinoma.[15]

The present study by Grewal and Mahmood [1] describe similar changes in small bowel mucins induced in rats following alcohol consumption. Despite the relative lack of information concerning the physiological role of small intestinal mucins the observed effects of alcohol on mucin composition and production indicates the complexity and variety of insults and injuries of the alcohol on the gastrointestinal tract.

REFERENCES

1. Grewal RK, Mahmood A. Ethanol induced changes in glycosylation of mucins in rat intestine. intestine. Annals of Gastroenterology, 2009;22:178-183.
2. Bode C, Bode JC. Effect of alcohol consumption on the gut. Best Pract Res Clin Gastroenterol 2003;17:575-592.
3. Kelly JP, Kaufman DW, Koff RS, Laszlo A, Wiholm BE, Shapiro S. Alcohol consumption and the risk of major upper gastrointestinal bleeding. Am J Gastroenterol 1995;90:1058-1064.
4. Addolorato G, Capristo E, Gasbarrini G, Stefanini GF. Depression, alcohol abuse and orocaecal transit time. Gut 1997;41:417-418.
5. Parlesak A, Schafer C, Schutz T, Bode JC, Bode C. Increased intestinal permeability to macromolecules and endotoxemia in patients with chronic alcohol abuse in different stages of alcohol-induced liver disease. J Hepatol 2000;32:742-747.
6. Mathurin P, Deng QG, Keshavarzian A, Choudhary S, Holmes EW, Tsukamoto H. Exacerbation of alcoholic liver injury by enteral endotoxin in rats. Hepatology 2000;32:1008-1017.
7. Kolios G, Valatas V, Kouroumalis E. Role of Kupffer cells in the pathogenesis of liver disease. World J Gastroenterol 2006;12:7413-7420.
8. Oekonomaki E, Notas G, Mouzas IA, Valatas V, Skordilis P, Xidakis C, Kouroumalis EA. Binge drinking and nitric oxide metabolites in chronic liver disease. Alcohol Alcohol 2004;39:106-109.
9. Shirazi T, Longman RJ, Corfield AP, Probert CS. Mucins and inflammatory bowel disease. Postgrad Med J 2000;76:473-478.
10. Allen A. Structure of gastrointestinal mucus glycoproteins and the viscous and gel-forming properties of mucus. Br Med Bull 1978;34:28-33.
11. Allen A, Flemstrom G. Gastroduodenal mucus bicarbonate barrier: protection against acid and pepsin. Am J Physiol Cell Physiol 2005;288:C1-19.
12. Tsai HH, Dwarakanath AD, Hart CA, Milton JD, Rhodes JM. Increased faecal mucin sulphatase activity in ulcerative colitis: a potential target for treatment. Gut 1995;36:570-576.
13. Kyo K, Parkes M, Takei Y, Nishimori H, Vyas P, Satsangi J, Simmons J, Nagawa H, Baba S, Jewell D, Muto T, Lathrop GM, Nakamura Y. Association of ulcerative colitis with rare VNTR alleles of the human intestinal mucin gene, MUC3. Hum Mol Genet 1999;8:307-311.
14. Buisine MP, Desreumaux P, Debailleul V, Gambiez L, Geboes K, Ectors N, Delescaut MP, Degand P, Aubert JP, Colombel JF, Porchet N. Abnormalities in mucin gene expression in Crohn's disease. Inflamm Bowel Dis 1999;5:24-32.
15. An G, Wei B, Xia B, McDaniel JM, Ju T, Cummings RD, Braun J, Xia L. Increased susceptibility to colitis and colorectal tumors in mice lacking core 3-derived O-glycans. J Exp Med 2007;204:1417-1429.