Journal of Bacteriology and Virology 2015. Vol. 45, No. 4 p.394 – 396
http://dx.doi.org/10.4167/jbv.2015.45.4.394

Letter to the Editor

High-fat-diet-modulated Gut Microbiota Promotes Intestinal Carcinogenesis

Irshad Ali and Young-Sang Koh*

Department of Microbiology and Immunology, School of Medicine and Brain Korea 21 PLUS Program, and Institute of Medical Science, Jeju National University, Jeju, Korea

Gut microbiota play a critical role in the development of intestinal cancer. Dietary changes cause dysbiosis of gut microbiota that mediates production of dietary factors triggering intestinal cancer. Genetic and dietary factors work in different combinatorial ways in initiation and progression of intestinal cancer, one of which is changes in gut microbiota. Recently, it has been found that high-fat-diet promote intestinal tumorigenesis in a genetically susceptible K-ras^{G12Dint} mice without induction of obesity. High-fat-diet along with oncogene activation dampened paneth-cell mediated immunity and thus shift bacterial communities in such a way that promotes intestinal cancer.

Key Words: Gut microbiota, Intestinal cancer, High-fat-diet, K-ras^{G12Dint}, Paneth cell

In Nature on 23rd October 2014, Schulz *et al*. reported that high-fat-diet-mediated dysbiosis promotes intestinal carcinogenesis independently of obesity (1). The gastrointestinal tract persistently provides shelter to large number of commensal (2). Alteration in the composition of gut microbiota has been associated with the development of intestinal cancer (3). In different human studies, high consumption of red meat and dietary fat have been implicated as risk factor for the development of colon cancer, which has been associated with dietary induced alteration in composition and metabolic activities of intestinal microflora (3). Inflammation causes the induction and expansion of intestinal microbiota including *E. coli* by decreasing protective mucin layer (4). The altered microbiota drives enhanced proliferation of epithelial cells which leads to increase tumorigenesis (3). Gut microbiota through fermentation of dietary fibre produces short chain fatty acids like butyrate, propionate and acetate, which promote colonic health. Butyrate acts as energy source, mediator of anti-inflammatory and antitumorigenic effect. It has been found that butyrate producing bacteria are significantly decreased in colon of ulcerative colitis and colon cancer patients (5). Genetic and nutritional risk factor operates in distinct combinatorial pattern in the development of intestinal carcinoma. Mutation in oncogenes or tumour suppressor genes and high consumption of diet containing more fats and less fibres, vitamin D_3, calcium and methyl donors increase susceptibility to intestinal carcinogenesis (6).

Ras family proteins including (K-Ras4A, K-Ras4B, H-Ras and N-Ras) are the member of small G proteins super-

Received: October 22, 2015/ Revised: October 25, 2015/ Accepted: October 26, 2015
*Corresponding author: Young-Sang Koh. Department of Microbiology and Immunology, Jeju National University School of Medicine, 102 Jejudaehakno, Jeju 63243, Korea.
Phone: +82-64-754-3851, Fax: +82-64-702-2687, e-mail: yskoh7@jejunu.ac.kr
**This study was supported by a grant from the National R&D Program for Cancer Control, Ministry of Health and Welfare, Republic of Korea (1120340).

family. They work as GDP/GTP-regulated switches to transmit extracellular signals that influence cell proliferation, apoptosis and modelling of actin cytoskeleton. In these proteins mutation at amino acids 12, 13 or 61 lock the enzyme in GTP-bound, activated form (7). Persistent Ras-ERK signal activation is linked with various cancers (8). Here, we summarize the results of Schulz *et al*. which demonstrate that high-fat-diet (HFD)-mediated dysbiosis promotes intestinal carcinogenesis independently of obesity (1).

Schulz *et al*. used well characterized serrated hyperplasia mice model having oncogenic expression of K-ras in intestinal epithelium and fed them HFD for 22 weeks started at the age of 3 weeks. They found that only 33% of age-matched $Kras^{G12Dint}$ mice on normal diet exhibited murine serrated hyperplasia, while 60% of HFD-fed $K\text{-}ras^{G12Dint}$ mice led to further tumour progression ranged from murine serrated adenoma, with low grade dysplasia and high grade dysplasia, to invasive carcinoma. This tumour progression pattern closely recapitulates human's carcinogenic sequence. Furthermore it was found that diet induce obesity was impaired in $K\text{-}ras^{G12Dint}$ mice during cancer progression, evident from high insulin sensitivity and lower lipid accumulation in liver of these animals. These results show that diet induced effects were culprit for promoting serrated intestinal tumorigenesis (1).

Inflammation triggers the induction and expansion of intestinal microbiota including *E. coli* that has positive correlation with colitis associated cancer (4). In the duodenal samples from $K\text{-}ras^{G12Dint}$ mice, they unexpectedly found significant decrease in the expression of genes having role in antigen recognition and immune response, which signify the oncogene mediated down regulation of host immunity. Paneth cell in response to bacterial exposure produce cryptdins that have antimicrobial activity and thereby shape the gut microbiota. This function of paneth-cell was sufficiently down regulated in $K\text{-}ras^{G12Dint}$ mice irrespective of tumour incidence. Mucin, a major component of mucus layer covering the intestinal epithelium was significantly down regulated after intake of HFD. In correlation with gene expression data from duodenal samples, in lamina propria and Peyer's patches the MHC II expression in $CD11c^{+}$ and $CD11b^{+}$ cell population was remarkably decreased in $K\text{-}ras^{G12Dint}$ mice irrespective of diet. These data demonstrate that reduced cryptdins expression by paneth cell, could contribute to the susceptibility of $K\text{-}ras^{G12Dint}$ mice to dampened immunity (1).

Alteration in the composition of gut microbiota has been associated with the development of intestinal cancer. The altered microbiota drive enhanced proliferation of epithelial cells which lead to increase tumorigenesis (3). To check whether increased tumour incidence were mediated by HFD-induced alteration in composition of microbiota, the 16S ribosomal RNA gene sequencing of amplicons generated from small intestine and colonic faecal DNA was conducted. As compare to normal diet HFD altered the intestinal microbiota community diversity. Higher abundance of Enterobacteriaceae, Lactobacillaceae, Helicobacteraceae, Peptostreptococcaceae and Clostridiaceae and lower abundance of Bifidobacteriaceae, Porphyromonadaceae and Alcaligenaceae in HFD-fed $K\text{-}ras^{G12Dint}$ mice was found as compare to $Kras^{G12Dint}$ mice provided with normal diet. Although no tumours were found in colon, still the colonic tissue from $K\text{-}ras^{G12Dint}$ mice fed with HFD showed high abundance of Enterobacteriaceae, Porphyromonadaceae, Desulfovibrionaceae, Rikenellaceae, Lachnospiraceae, Ruminococcaceae, Coriobacteriaceae and Deferribacteraceae and lower abundance of Peptostreptococcaceae, Bifidobacteriaceae, Roseburia and Butyricicoccus as compare to those $K\text{-}Ras^{G12Dint}$ mice fed with normal diet. So it was concluded that HFD mediated major changes in composition of intestinal microbiota of $K\text{-}ras^{G12Dint}$ mice bearing tumours in their gut (1).

Butyrate, a short chain fatty acid is the fermentation product of dietary fibre by bacteria. During ulcerative colitis and colon cancer progression, the abundance of butyrate producing bacteria and potential of butyrate production are significantly reduced (5). HFD-mediated tumour progression was associated with remarkable decrease in concentration of butyrate, propionate and acetate in stool samples. To confirm whether HFD-associated tumour progression could be stopped by oral administration of butyrate to the mice. Although butyrate treatment only slightly increase faecal butyrate level, but significantly decrease the tumour incidence.

This reduction in tumour incidence was associated with a marked increase in the abundance of Porphyromonadace and Bifidobacteriaceae and sharp reduction in abundance of Helicobacteraceae in the small intestine. Remarkably, butyrate supplementation partially blocked the branching, serration and proliferation in K-ras^{G12Dint} mice. Similarly the HFD-induced reduction in *Muc2* expression level and dampened immunity was partially restored to the extent of normal diet group with butyrate treatment. So they concluded that butyrate protects against intestinal tumorigenesis at least partially by changing the composition of gut microbiota and regulation of K-ras signalling (1).

To confirm that diet induced dysbiosis is associated with intestinal tumorigenesis, fresh faecal transfer experiment was conducted and shown that disease was transmitted to healthy K-ras^{G12Dint} mice on normal diet when colonized with fresh faecal sample from tumour bearing HFD-fed K-ras^{G12int} donors. The K-ras^{G12Dint} mice on normal diet receiving stool from HFD-fed K-ras^{G12Dint} mice showed high abundance of Helicobacteraceae, Lactobacillaceae and Clostridiales, showing the transfer of HFD-shaped microbiota to the normal diet group. Similarly, mice on normal diet showed lower MUC2 expression and dampened immunity when colonized with fresh faecal sample from HFD-fed K-ras^{G12Dint} mice. Antibiotic treatment abolished tumour formation in HFD-fed K-ras^{G12Dint} mice (1).

Schulz *et al*. showed that HFD-shaped dysbiotic gut microbiota aggravated oncogene driven intestinal carcinogenesis independently of obesity. This dysbiotic microbiota was transmissible to oncogenic susceptible mice on normal diet through colonization with fresh stool from K-ras^{G12Dint} mice on HFD. Butyrate abrogated tumour formation and dysbiosis and also restored the dampened immunity. This study showed that diet induced cancer is based on distinct shift in microbial communities independent of obesity. Thus, modulating the microbiota of an individual through personalized diet intervention might help to promote health, particularly in those peoples who are at high risk because of high fat intake and genetic susceptibility.

REFERENCES

1) Schulz MD, Atay C, Heringer J, Romrig FK, Schwitalla S, Aydin B, *et al*. High-fat-diet-mediated dysbiosis promotes intestinal carcinogenesis independently of obesity. Nature 2014;514:508-12.
2) Lee SH, Hu LL, Gonzalez-Navajas J, Seo GS, Shen C, Brick J, *et al*. ERK activation drives intestinal tumorigenesis in Apc$^{(min/+)}$ mice. Nat Med 2010;16:665-70.
3) Hu B, Elinav E, Huber S, Strowig T, Hao L, Hafemann A, *et al*. Microbiota-induced activation of epithelial IL-6 signaling links inflammasome driven inflammation with transmissible cancer. Proc Natl Acad Sci U S A 2013; 110:9862-7.
4) Arthur JC, Perez-Chanona E, Mühlbauer M, Tomkovich S, Uronis JM, Fan TJ, *et al*. Intestinal inflammation targets cancer-inducing activity of the microbiota. Science 2012;338:120-3.
5) Singh N, Gurav A, Sivaprakasam S, Brady E, Padia R, Shi H, *et al*. Activation of Gpr109a, receptor for niacin and the commensal metabolite butyrate, suppresses colonic inflammation and carcinogenesis. Immunity 2014;40:128-39.
6) Wang D, Peregrina K, Dhima E, Lin EY, Mariadason JM, Augenlicht LH. Paneth cell marker expression in intestinal villi and colon crypts characterizes dietary induced risk for mouse sporadic intestinal cancer. Proc Natl Acad Sci U S A 2011;108:10272-7.
7) Haigis KM, Kendall KR, Wang Y, Cheung A, Haigis MC, Glickman JN, *et al*. Differential effects of oncogenic K-Ras and N-Ras on proliferation, differentiation and tumor progression in the colon. Nat Genet 2008; 40:600-8.
8) Manzoor Z, Koo JE, Koh YS. Mitogen-activated protein kinases signaling in inflammation-related carcinogenesis. J Bacteriol Virol 2014;44:297-304.