PROTECTIVE EFFECT OF HANWOO BEEF LOIN DIET ON DEXTRAN SULFATE SODIUM-INDUCED COLITIS ANIMAL MODLE

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I. INTRODUCTION

Inflammatory bowel disease (IBD) is characterized by recurrent chronic inflammation and mucosal tissue damage of the intestinal tract. Especially, 25–30% of patients with long-term conditions of colitis will develop colorectal cancer (CRC) [1]. In October 2015, the International Agency for Research on Cancer has classified the red meat as probably carcinogenic to humans (Group 2A) and it has been associated with CRC. However, some experimental studies showed opposition to that result [2, 3]. Relationship with consumption of red meat and development of CRC is not clear and continuous research is still needed. Therefore, this study aimed to evaluate effect of diet including Hanwoo beef loin, as a red meat, on dextran sulfate sodium (DSS)-induced ulcerative colitis in mice, as one of the main reason of CRC development

II. MATERIALS AND METHODS

Hanwoo beef loin (*M. longissimus*) was purchased from local market within 24 h post mortem (Chuncheon, Korea). Hanwoo beef loin was boiled in water bath up to 75°C of internal temperature. These were lyophilized and mixed with Standard rodent chow at concentration of 10 and 20% each. A total of 48 male C57BL/6J mice aged 4 weeks old were obtained from Daehan bio-link (Chungbuk, Korea). The mice were raised in conventional animal room. After 1 week adaptation, healthy mice were randomly allocated into 4 groups (n=12) as referred to Table 1. Hanwoo beef loin diet was supplemented for 42 days and 3% DSS was co-treated to DSS groups to induce colitis in mice for last one week. After sacrifice, the colon from cecum to rectum was removed and colon length was measured. The triglyceride (TG), total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), and low-density lipoprotein cholesterol (LDL-C) in serum were determined by enzymatic kits. For analyzing oxidative marker, colonic malondialdehyde (MDA) and glutathione (GSH) contents were measured by Colado *et al.* [4], and Ellman [5] method, respectively. The last one cm of colon was fixed and embedded in paraffin and stained with hematoxylin and eosin (H&E). All experiments were performed in triplicate and results are presented as the mean and standard error of the mean (SEM) with Tukey's tests to determine significance at $p < 0.05$.

III. RESULTS AND DISCUSSION

Typical symptoms of DSS-induced experimental colitis are reduction of body weight and feed intake, colon shortening, and histological alterations [6]. As colitis was induced, body weight in all groups except CON group was dropped about 20% at day 41 compared to day 35 ($p < 0.05$) (data not shown). However, mice feed with Hanwoo beef loin showed increased colon length compared to DSS group ($p < 0.05$) (Table 1). The TC and LDL-C levels of serum in DSS group were increased and HDL-C level was decreased compared to CON group ($p < 0.05$) (Table 1). Inflammation can increase cholesterol levels in serum by stimulating cholesterol synthesis, and decreasing lipoprotein clearance [7]. However, DSS+HL20 group alleviated lipid metabolism disorder caused by DSS-induced colitis via decreasing the level of TG, TC, and LDL-C in serum compared to DSS group ($p < 0.05$).

Table 1 Effects of Hanwoo beef loin diet on colon length and lipid profiles in serum of DSS treated mice

Treatment	Colon length (cm)	Lipid profiles (mg/dl)			
		TG	TC	HDL-C	LDL-C
CON[1)]	7.80^{A}	178.23^{AB}	109.86^{C}	63.78^{A}	10.43^{C}
DSS	4.52^{C}	198.88^{A}	134.77^{A}	37.58^{B}	57.41^{A}

DSS+HL10	6.27^{B}	145.52^{BC}	124.12^{B}	44.41^{B}	50.60^{AB}
DSS+HL20	6.52^{B}	135.54^{C}	114.59^{C}	40.67^{B}	46.82^{B}
SEM[2)]	0.178	8.396	2.049	1.799	2.510

$^{A-C}$ Means within a column with different superscript differ significantly at $p < 0.05$. [1)] CON, mice fed basal diet; DSS, mice fed basal diet and induced colitis; DSS+HL10, mice fed basal diet with 10% Hanwoo beef loin and induced colitis; DSS+HL20 mice fed basal diet with 20% Hanwoo beef loin and induced colitis. [2)] SEM, standard error of means.

As DSS induces infiltration of neutrophil, oxidative stress is increased in colon tissue. As a result, MDA is increased as products of lipid peroxidation, and GSH is decreased as the substrate of antioxidant enzyme [8]. As shown in Table 2, DSS+10HL and 20HL groups reduced the colonic MDA content and increased contents of colonic GSH compared to DSS group ($p < 0.05$). It is suggested that diet Hanwoo beef loin protected colonic mucosa by reducing the lipid oxidation. As protective effect of diet Hanwoo beef was also shown in histological exam, it appeared to lessen the severity of loss of epithelium and crypt, and infiltration of immune cells (Fig. 1).

Table 2 Effects of Hanwoo beef loin diet on colonic MDA and GSH contents in DSS treated mice

Treatment	MDA (nmol/mg protein)	GSH (nmol/mg protein)
CON[1)]	0.77^{B}	34.19^{A}
DSS	1.16^{A}	21.83^{B}
DSS+HL10	0.75^{B}	28.62^{A}
DSS+HL20	0.65^{B}	29.12^{A}
SEM[2)]	0.033	1.299

$^{A-B}$ Means within a column with different superscript differ significantly at $p < 0.05$. [1)] Refer to Table 1. [2)] SEM, standard error of means.

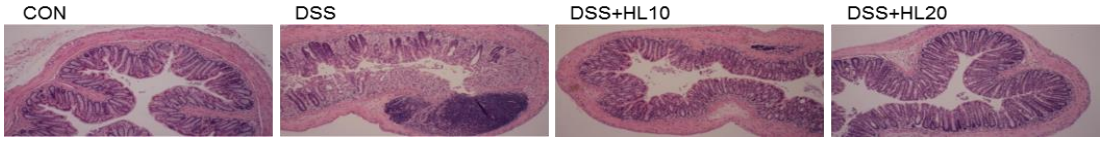

Figure 1. Representative microscopic view of the colon tissue (×100). CON, mice fed basal diet; DSS, mice fed basal diet and induced colitis; DSS+HL10, mice fed basal diet with 10% Hanwoo beef loin and induced colitis; DSS+HL20 mice fed basal diet with 20% Hanwoo beef loin and induced colitis.

IV. CONCLUSION

The Hanwoo beef loin diet had protective effect against DSS-induced colitis in mice. Its effect was via improving the lipid metabolism in serum, and reducing the oxidative stress in colon tissue. This study can suggest that there is no relation between red meat intake and development of colitis.

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REFERENCES

1. Macdonald, T. T. & Monteleone, G. (2005). Immunity, inflammation, and allergy in the gut. Science 307: 1920–1925.
2. Kettunen, H. L., Kettunen, A. S. & Rautonen, N. E. (2003). Intestinal immune responses in wild-type and Apcmin/+ mouse, a model for colon cancer. Cancer Research 63: 5136-5142.
3. Domingo, J. L. (2017). Concentrations of environmental organic contaminants in meat and meat products and human dietary exposure: A review. Food and Chemical Toxicology 107: 20-26.
4. Colado, M. I., O'shea, E., Granados, R., Misra, A. & Murray, T. K. (1997). Green AR. A study of the neurotoxic effect of MDMA (‘ecstasy’) on 5-HT neurones in the brains of mothers and neonates following administration of the drug during pregnancy. British Journal of Pharmacology 121: 827-833.
5. Ellman, G. L. (1959). Tissue sulfhydryl groups. Archives of Biochemistry and Biophysics 82: 70-77.
6. Solomon, L., Mansor, S., Mallon, P., Donnelly, E., Hoper, M., Loughrey, M., Kirk, S. & Gardiner, K. (2010). The dextran sulphate sodium (DSS) model of colitis: an overview. Comparative Clinical Pathology 19: 235-239.
7. Khovidhunkit, W., Kim, M. S., Memon, R. A., Shigenaga, J. K., Moser, A. H., Feingold, K. R. & Grunfeld, C. (2004). Effects of infection and inflammation on lipid and lipoprotein metabolism: mechanisms and consequences to the host. Journal of Lipid Research 45: 1169-1196.
8. Pandurangan, A. K., Saadatdoust, Z., Hamzah, H. & Ismail, A. (2015). Dietary cocoa protects against colitis-associated cancer by activating the Nrf2/Keap1 pathway. BioFactors 41: 1-14.