Are Short Chain Fatty Acids in Gut Microbiota Defensive Players for Inflammation and Atherosclerosis?

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Intestinal flora (microbiota) have recently attracted attention among lipid and carbohydrate metabolism researchers. Microbiota metabolize resistant starches and dietary fibers through fermentation and decomposition, and provide short chain fatty acids (SCFAs) to the host. The major SCFAs acetates, propionate and butyrate, have different production ratios and physiological activities. Several receptors for SCFAs have been identified as the G-protein coupled receptor 41/free fatty acid receptor 3 (GPR41/FFAR3), GPR43/FFAR2, GPR109A, and olfactory receptor 78, which are present in intestinal epithelial cells, immune cells, and adipocytes, despite their expression levels differing between tissues and cell types. Many studies have indicated that SCFAs exhibit a wide range of functions from immune regulation to metabolism in a variety of tissues and organs, and therefore have both a direct and indirect influence on our bodies. This review will focus on SCFAs, especially butyrate, and their effects on various inflammatory mechanisms including atherosclerosis. In the future, SCFAs may provide new insights into understanding the pathophysiology of chronic inflammation, metabolic disorders, and atherosclerosis, and we can expect the development of novel therapeutic strategies for these diseases.

Key words: Short chain fatty acids, Butyrate, Microbiota, Inflammation, Atherosclerosis

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Intestinal Bacterial Flora and Short Chain Fatty Acids

In accordance with systematic taxonomy, microbiota are organized by phylum, class, order, family, genus, and species. It has been difficult to culture microbiota, since most are obligate anaerobes, and thus many of their specific roles have remained unknown. However, recent developments in genetic analysis, including 16S ribosomal RNA sequencing and metagenomics analysis, have begun to reveal their function. The predominant microbiota bacteria that produce SCFAs are classified as Ruminococcaceae (cluster IV) and Eubacterium (cluster XIVa) in the order Clostridia, class Clostridia, and phylum Firmicutes. The predominant producers of SCFAs are shown in Table 1.

SCFAs account for 2-10% of the total energy consumption in humans, are the main energy source for large intestinal epithelial cells, and affect the production of mucins (mucus). In addition, SCFAs physiologically influence blood flow to the colon mucous membrane, the absorption of fluids and electrolytes, the autonomic nervous system, and the secretion of gut hormones. A considerable part of the beneficial effect of prebiotics (usually non-digestive fiber compounds) is thought to be due to SCFAs produced by intestinal microbes. Research has shown that the concentration of SCFAs is 70-140 mmol/L in the proximal colon and 20-70 mmol/L in the distal colon. In general, acetate is thought to be more prevalent, followed by proprionate and butyrate; however, assessing the ratios of SCFAs is extremely difficult since their production depends on the various types of fermentation substrates. Animal experiments have shown that the total amount of SCFAs may be approximately 400-600 mmol/day when 60 g/day of undigested carbohydrates reach the colon. In humans, however, since most studies on SCFAs have been conducted using fecal samples through absorp-

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**Table 1. SCFAs (Acetate, Propionate, Butyrate) production by microbiota in the Gut**

<table>
<thead>
<tr>
<th>SCFAs</th>
<th>Pathways/Reactions</th>
<th>Producers</th>
<th>References</th>
</tr>
</thead>
</table>

Citationed from Koh A, et al, Cell 165, 1332-45 (8)

spp., species; (A), acetate is the substrate for producing butyrate; (L), lactate is the substrate for producing butyrate
tion, the abovementioned estimates of intestinal concentrations may not reflect actual conditions, and many issues are yet to be elucidated. **Table 2** shows the concentrations of SCFAs in the feces of adult humans that have been previously reported\(^{19-33}\).

Nearly all SCFAs absorbed by the colon are thought to pass through the portal vein from the colon capillaries and reach the liver, though the concentrations of SCFAs in the human portal vein are broad-ranging. **Table 3** shows the results of the several studies that have reported on the concentrations of SCFAs in the portal vein\(^{4, 34-36}\).

The concentrations of SCFAs in healthy adult human peripheral blood are estimated as 100 to 150 \(\mu\)mol/L for acetate, 4 to 5 \(\mu\)mol/L for propionate, and 1 to 3 \(\mu\)mol/L for butyrate, indicating that these concentrations in peripheral blood are vastly lower than in the intestinal tract\(^{4}\). In experiments using rats, a correlation has been found between the cecum content and portal or aortic serum concentrations of total SCFAs after feeding with highly fermentable fiber diets (pectin, guar gum, and fructo-oligosaccharides)\(^{37}\). Oral administration of tributyrin (a prodrug of butyrate) increased plasma butyrate concentrations in the portal vein to 2.4 mmol/L at 1 h and 0.7 mmol/L at 2.5 h\(^{38}\).

**Short Chain Fatty Acids and Their Receptors**

Several receptors for SCFAs have been found to be G-protein coupled receptors (GPR). Among these, GPR41 and 43 have been renamed as free fatty acid receptor (FFAR) 3 and FFAR2, respectively. GPR41/FFAR3 is distributed throughout the entire body with a high degree of expression in the intestinal tract, immune cells, and fatty tissues\(^{8, 39}\), and is thought to be associated with adiposity and energy homeostasis\(^{41}\).

GPR43/FFAR2 is expressed in intestinal tract epithelial cells and immune system cells, which suggests that it is related to cell chemotaxis and activation\(^{8, 39}\). Interestingly, GPR43/FFAR2 is also expressed within adipocytes in white adipose tissue, and experiments using GPR43/FFAR2\(^{-/-}\) mice have shown that GPR43/FFAR2 signaling with SCFAs may be effective in lipolysis control in adipocytes\(^{40}\).

GPR109A/hydroxycarboxylic acid receptor (HCA) 2, which is known to be a niacin receptor, has been identified as a receptor to butyrate as well as beta-hydroxybutyric acid, a ketone body\(^{43, 44}\). Its expression sites are intestinal tract epithelial cells, immune cells, and adipocytes\(^9\). GPR109A/HCA2 participates in homeostasis of regular T cells (Treg) in the colon and fat metabolism in adipose tissues\(^{45, 46}\). In addition, GPR109A/HCA2 signaling accelerates inflammation in hypertrophic adipose tissues\(^{40}\).

Recently, olfactory receptor (Olfr) 78, which is a member of the GPR family, has been reported to be a novel SCFA receptor\(^{47, 48}\). Olfr78 is thought to be associated with regulation of hormone secretion and blood pressure\(^{47, 48}\). **Table 4** shows the characteristics and physiological functions of SCFA receptors\(^8, 47, 48\).

**Short Chain Fatty Acids and T Cells**

SCFAs regulate T cell polarization and induction\(^{49}\). Propionate (at concentrations of 2.0 to 5.0 mmol/L) inhibits the proliferation of lymphocytes stimulated by mitogens\(^{50}\). Propionate (250 to 500 \(\mu\)mol/L) suppresses the Th1-type immune response in stimulated human peripheral blood mononuclear cells\(^{51}\). Butyrate (1.0 mmol/L) inhibits the proliferation of T lymphocytes, and more than 2.0 mmol/L of butyrate induces apoptosis in activated T lymphocytes, but not primary macrophages\(^{52}\). Trompet et al. have reported that there are differences in the intestinal *Firmicutes/Bacteroides* ratio (F/B ratio) and microbiota composition between high- and low-fiber diets in mice, and that a high-fiber diet increases blood concentrations of SCFAs (approximately 1.0 to 2.0 mmol/L) and attenuates allergic inflammation of the lungs\(^{53}\). The authors suggested that propionate is involved in bone marrow hematopoiesis and in the enhanced generation of macrophage and dendritic cell (DC) precursors and subsequent seeding of the lungs by DCs with high phagocytic capacity, but with an impaired ability to activate Th2 effector cells in the lung\(^{53}\). In addition, they suggested that these effects are induced via GPR41/FFAR3 but not GPR43/FFAR2\(^{59}\).

Compounds acting as histone deacetylase (HDAC) inhibitors may be an effective treatment for inflammatory bowel disease and other pro-inflammatory cytokine-related diseases\(^{54}\). As SCFAs are widely known to have HDAC inhibitory activity, they may be involved in the expression of cytokines in T cells and the induction of Treg cells via inhibition of HDAC\(^{55}\). SCFAs (acetate 5-20 mmol/L, propionate 0.5-1.0 mmol/L) promote naïve CD4\(^+\) T cell polarization into Th1 and Th17 effector cells producing interleukin (IL)-17, interferon-\(\gamma\), and/or IL-10\(^{56}\). This effect is independent of GPR41 and GPR43, but directly dependent on the HDAC inhibitor activity and subsequent enhancement of mTOR-S6 kinase activity\(^{56}\). More than 1 mmol/L of butyrate induces Fas-mediated apoptosis of T cells by inhibiting HDAC 1 activity to induce Fas promoter hyperacetylation and Fas upregulation in T cells\(^{57}\).

When butyrate is supplied into the colons of T cell-dependent colitis mouse models, the number of
Table 2. Fecal concentration of individual SCFAs (Acetate, Propionate, Butyrate) by human adults

<table>
<thead>
<tr>
<th>Subjects (n); age:</th>
<th>Reported measure</th>
<th>Acetate</th>
<th>Propionate</th>
<th>Butyrate</th>
<th>Total SCFAs</th>
<th>Unit</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Healthy subjects</td>
<td>10; 21-34 years</td>
<td>Mean (SD)</td>
<td>218 (99)</td>
<td>72 (37)</td>
<td>58.7 (54.5)</td>
<td>378 (188)</td>
<td>µmol/g dry weight</td>
</tr>
<tr>
<td></td>
<td>20; 20-40 years</td>
<td>Mean (SEM)</td>
<td>320.3 (24.9)</td>
<td>97.3 (10.5)</td>
<td>93.8 (9.13)</td>
<td>511.4 (41.9)</td>
<td>µmol/g dry weight</td>
</tr>
<tr>
<td></td>
<td>13; 23-58 years</td>
<td>Median (IQR)</td>
<td>52.2</td>
<td>23.2 (13.6-37.3)</td>
<td>36.8 (5-128)</td>
<td>119.3 (64.5-197.0)</td>
<td>µmol/g wet weight</td>
</tr>
<tr>
<td></td>
<td>60; 18-24 years</td>
<td>Mean (SEM)</td>
<td>198.4 (14.2)</td>
<td>55.2 (4.7)</td>
<td>93.8 (9.13)</td>
<td>304.1</td>
<td>µmol/g dry weight</td>
</tr>
<tr>
<td></td>
<td>27; 18-55 years</td>
<td>Mean (SEM)</td>
<td>35.8 (2.4)</td>
<td>11.4 (1.2)</td>
<td>10.0 (1.1)</td>
<td>61.1 (4.4)</td>
<td>µmol/g</td>
</tr>
<tr>
<td></td>
<td>12; 18-65 years</td>
<td>Mean (SD)</td>
<td>48</td>
<td>13.98</td>
<td>13.31</td>
<td>80.91</td>
<td>µmol/g</td>
</tr>
<tr>
<td></td>
<td>46; 31-66 years</td>
<td>Mean (95%CI)</td>
<td>44.7 females (39.7, 50.3)</td>
<td>12.3 females (10.7, 14.0)</td>
<td>11.7 females (9.8, 14.0)</td>
<td>69.5 females (61.3, 78.7)</td>
<td>µmol/g</td>
</tr>
<tr>
<td></td>
<td>36</td>
<td>Median (IQR)</td>
<td>43.7 (34.0-52.2)</td>
<td>13.1 (9.2-18.5)</td>
<td>8.8 (5.2-11.5)</td>
<td>91.8 (73.1-107.5)</td>
<td>µmol/g</td>
</tr>
<tr>
<td></td>
<td>20; 22-55 years</td>
<td>Mean (SEM)</td>
<td>42.13 (3.8)</td>
<td>11.5 (1.2)</td>
<td>11.28 (1.4)</td>
<td>67.3 (6.2)</td>
<td>µmol/g</td>
</tr>
<tr>
<td></td>
<td>8; 31-59 years</td>
<td>Mean (SD)</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>92.7 (33.9)</td>
<td>µmol/g</td>
</tr>
<tr>
<td></td>
<td>20; 23-28 years</td>
<td>Mean (SEM)</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>78.5 (6.4)</td>
<td>µmol/g</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>Mean (SD)</td>
<td>50.5 (12.6)</td>
<td>13.6 (5.2)</td>
<td>14.1 (7.6)</td>
<td>84.6 (22.9)</td>
<td>µmol/L</td>
</tr>
<tr>
<td>Obese subjects</td>
<td>20; 22-55 years</td>
<td>Mean (SEM)</td>
<td>47.2 (3.8)</td>
<td>13.6 (1.3)</td>
<td>14.7 (1.5)</td>
<td>78.8 (6.2)</td>
<td>µmol/g</td>
</tr>
<tr>
<td></td>
<td>91</td>
<td>Mean (SD)</td>
<td>58.5 (19.1)</td>
<td>17.6 (7.6)</td>
<td>18.3 (9.7)</td>
<td>102 (33.5)</td>
<td>µmol/L</td>
</tr>
<tr>
<td></td>
<td>32; 20-65 years</td>
<td>Mean (SEM)</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>34 (6)</td>
<td>mmol/24h</td>
</tr>
<tr>
<td></td>
<td>35 over weight</td>
<td>Mean (SD)</td>
<td>56.0 (18.2)</td>
<td>18.3 (7.9)</td>
<td>18.5 (10.1)</td>
<td>98.7 (33.9)</td>
<td>mmol/L</td>
</tr>
<tr>
<td></td>
<td>33 obese</td>
<td>Mean (SD)</td>
<td>59.8 (18.3)</td>
<td>19.3 (8.7)</td>
<td>18.1 (10.0)</td>
<td>103.9 (34.3)</td>
<td>mmol/L</td>
</tr>
</tbody>
</table>

Citationed from Verbeke KA, et al., Nutr Res Rev. 28:42-66.26) Mean, the mean value; SD, standard deviation; SEM, standard error of the mean; IQR, interquartile range; 95%CI, 95% confidence limit; NR, not reported.
Treg cells increases in the colonic lamina propria, and bowel inflammation is attenuated.\(^{58}\) The ingestion of propionate increases Treg cells in intestinal mucosa in germ-free mice via GPR43/FFAR2.\(^{59}\) By inhibition of HDAC, butyrate and to a lesser extent propionate, but not acetate, promotes transcription of the FoxP3 gene, which is the transcription factor for Treg differentiation, and increases the expression of the FoxP3 gene.\(^{55}\) On the other hand, 1 mmol/L of butyrate causes the induction of Th17 cells and also exacerbates inflammation by the production of IL-23 in stimulated dendritic cells (DCs).\(^{60}\)

Thus, SCFAs, especially butyrate and propionate, play a complicated role in Treg differentiation and intestinal tract immune regulation.\(^{9, 53, 61, 62}\)

### Short Chain Fatty Acids and Neutrophils, Monocytes, and Macrophages

The chemotaxis of neutrophils is activated by inflammatory mediators [tumor necrosis factor (TNF)-α, IL-17, etc.] and chemokines [chemokine (C-X-C motif) (CXCL) 1, 8, etc.]. SCFAs (optimal concentrations for migration are 0.1-3.0 mmol/L) affect the chemotaxis and the viability of neutrophils.\(^{63}\) SCFAs (4.0-12 mmol/L propionate, 0.4-3.2 mmol/L butyrate) inhibit TNF-α production by neutrophils in the presence of lipopolysaccharide (LPS).\(^{64}\) The suppression of nuclear factor-kappa B (NF-κB) activity and the inhibition of HDAC are thought to be the underlying mechanisms.\(^{64, 65}\) On the other hand, neutrophils increase the production of IL-8, IL-6, and IL-1β at high concentrations (20 mmol/L) of SCFAs while lower concentrations (0.02-2.0 mmol/L) do not induce cytokine secretion. However, lower concentrations of SCFAs enhance TLR2-induced production of IL-8 and TNF-α production.\(^{66}\) SCFAs suppress large intestine inflammation in dextran sulfate sodium-induced colitis mice by the induction of apoptosis of neutrophils via GPR43/FFAR2\(^{67}\) and via HDAC inhibition.\(^{68}\) In addition, SCFAs produce and release reactive oxygen species (ROS) as well as nitric

### Table 3. Portal concentrations of individual SCFAs (Acetate, Propionate, Butyrate) by human

<table>
<thead>
<tr>
<th>Subjects</th>
<th>(n); age</th>
<th>Treatment</th>
<th>Reported measure</th>
<th>Acetate</th>
<th>Propionate</th>
<th>Butyrate</th>
<th>Unit</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Died suddenly</td>
<td>6; 16-89 years</td>
<td>Not fasting</td>
<td>Mean</td>
<td>258</td>
<td>88</td>
<td>29</td>
<td>μmol/L</td>
<td>Cummings JH, et al., Gut 1987 28, 1221-7.(^{46})</td>
</tr>
<tr>
<td>Surgical patients</td>
<td>5; -</td>
<td>Fasting</td>
<td>Mean</td>
<td>114</td>
<td>32</td>
<td>9</td>
<td>μmol/L</td>
<td>Dankert J, et al., Clin Chim Acta. 1981 110; 301-7.(^{34})</td>
</tr>
<tr>
<td>Surgical patients</td>
<td>28; 23-74 years</td>
<td>Fasting</td>
<td>Mean</td>
<td>128</td>
<td>34</td>
<td>18</td>
<td>μmol/L</td>
<td>Peters SG, et al., Gut. 1992 33; 1249-52.(^{55})</td>
</tr>
<tr>
<td>Surgical patients</td>
<td>10; 10 g Lactulose was injected into the caecum at surgery (Peak value)</td>
<td>Fasting</td>
<td>Mean</td>
<td>241</td>
<td>39</td>
<td>27</td>
<td>μmol/L</td>
<td>Peters SG, et al., Gut. 1992 33; 1249-52.(^{55})</td>
</tr>
<tr>
<td>Surgical patients</td>
<td>6; 6.7 g Lactulose was injected into the caecum at surgery (Peak value)</td>
<td>Fasting</td>
<td>Mean</td>
<td>166</td>
<td>31</td>
<td>22</td>
<td>μmol/L</td>
<td>Peters SG, et al., Gut. 1992 33; 1249-52.(^{55})</td>
</tr>
<tr>
<td>Surgical patients</td>
<td>7; 54-73 years</td>
<td>Fasting</td>
<td>Mean</td>
<td>236</td>
<td>18</td>
<td>26</td>
<td>μmol/L</td>
<td>van der Beek CM, et al., J Nutr. 2015 145: 2019-24.(^{36})</td>
</tr>
<tr>
<td>Surgical patients</td>
<td>7; 54-73 years</td>
<td>Fasting butyrate: 100 mmol/L; 60 mL enema at surgery (Peak value)</td>
<td>Mean</td>
<td>NR</td>
<td>NR</td>
<td>92</td>
<td>μmol/L</td>
<td>van der Beek CM, et al., J Nutr. 2015 145: 2019-24.(^{36})</td>
</tr>
</tbody>
</table>

Mean, the mean value; NR, not reported
oxide (NO) involving neutrophil bacteria phagocytosis. Thus, SCFAs have both suppressing and promoting functions in neutrophils.

SCFAs also affect immunoregulation in monocytes and macrophages. In experiments using human monocytes, SCFAs (0.2-20 mmol/L) reduce the production of TNF-α and monocyte chemotactic protein-1 (MCP-1) under LPS stimulation and increase the production of prostaglandin E2 (PGE2). The suppression of NF-κB activity and inhibition of HDAC are thought to be the underlying mechanisms, as in the case of neutrophils. Butyrate increases the production of PGE2 by upregulating the expression of phospholipase A2 (PLA2) and cyclooxygenase-2 (COX2) in Kupffer cells (at butyrate concentrations of 0.5-10 mmol/L), in human peripheral blood mononuclear cells (1.0-2.0 mmol/L), and in a mouse macrophage cell line (0.2-1.0 mmol/L) (Fig. 1 (A)). In a human macrophage cell line, butyrate (1.0 mmol/L) increases the production and release of ROS when under LPS stimulation and increases caspase-1 expression and IL-1β production.

### Table 4. The characteristics and physiological functions of SCFA receptors

<table>
<thead>
<tr>
<th>G protein</th>
<th>Ligand</th>
<th>EC50</th>
<th>Expression</th>
<th>Physiological function</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPR41/FFAR3</td>
<td>Gi/o</td>
<td>C1-C5</td>
<td>12-274 μmol/L for C3</td>
<td>Colonic, colonic LP cells (mast cells), spleen, lymphnodes, bonemarrow, adipocytes, peripheral mononuclear cells, peripheral nervous system, etc.</td>
</tr>
<tr>
<td>GPR43/FFAR2</td>
<td>Gi/o, Gq11</td>
<td>C1-5</td>
<td>259-537 μmol/L</td>
<td>Colonic, colonic LP cells (mast cells, neutrophils, eosinophils, and Tregs), polymorphonuclear cells, adipocytes, skelatal muscle, etc.</td>
</tr>
<tr>
<td>GPR109A/HCA2</td>
<td>Gi/o, Gβγ</td>
<td>β-D-OHB &gt; Butyrate (C4)</td>
<td>0.7 mmol/L (mouse), 1.6 mmol/L (human) for C4</td>
<td>Apical membrane of colonic epithelium, macrophages, monocytes, DCs, neutrophils, adipocytes (white and brown), etc.</td>
</tr>
<tr>
<td>Olfr78</td>
<td>NR</td>
<td>Propionate(C3) &gt; acetate (C2)</td>
<td>920 μmol/L for C3, 2.35 mmol/L for C2</td>
<td>Nurons, enteroendocrine cells, colon (epithelial enteroendocrine cells), renal afferent arteriole, juxtaglomerular cells, smooth muscle cells (blood vessels)</td>
</tr>
</tbody>
</table>

Citationed from Koh A, et al., Cell. 165: 1332-45
Citationed from Fleischer J, et al., Cell Tissue Res. 361: 697-710
Citationed from Fleischer J, et al., Cell Tissue Res. 361: 697-710

NR, not reported; EC50, half maximal (50%) effective concentration; GPR, G-protein coupled receptor; FFAR, free fatty acid receptor; HCA, hydroxycarboxylic acid receptor; Olfr, olfactory receptor; LP, lamina propria; T reg, regulatory T cell; DC, dendritic cell; GLP-1, glucagon-like peptide; PYY, peptide YY; IBD, inflammatory bowel disease

**Short Chain Fatty Acids and Atherosclerosis**

Hazen *et al.* have revealed that gut microbial-derived metabolites trimethylamine (TMA) and trimethylamine N-oxide (TMAO) are proatherogenic in mice and humans. They have also shown that plasma TMAO concentrations can predict an enhanced risk of major adverse cardiac events in two independent cohorts.

Recently, Aguilar *et al.* demonstrated that a butyrate-supplemented chow diet suppresses atherosclerotic lesions in apoE knockout mice. They also observed that butyrate reduces chemotaxis protein-1 (CCL2/MCP-1), vascular cell adhesion molecule-1, and matrix metalloproteinase-2 production in the lesion site, resulting in a lower migration of macrophages and increased collagen deposition and plaque stability, and that peritoneal macrophages from butyrate-treated mice present lower ROS and NO release.

On the other hand, Kasahara *et al.* demonstrated that the lack of microbiota in apoE-deficient mice...
causes a significant reduction in atherosclerotic lesion formation in spite of a significant increase in plasma and hepatic cholesterol concentrations, and suggested that this might be associated with the attenuation of LPS-mediated inflammatory responses. They also found that gut microbiota can regulate cholesterol homeostasis via bile acid metabolism under hypercholesterolemia. Ryan demonstrated shifts in the composition of the gut microbiome in apoE-deficient mice fed high fat/cholesterol in conjunction with...
plant sterol esters or oat β-glucan, and increased concentrations of cecum acetate or butyrate, and found that these feedings attenuated the microbial production of TMA and reduced serum cholesterol concentrations.

In accordance with experimental data, recent clinical work has shown that coronary artery disease is linked with an alternation of gut microbiota, and that gut microbiota may be a diagnostic marker of morbidity from coronary artery disease. Emoto et al. reported that the incidence of coronary artery disease is related to a decreased prevalence of the phylum Bacteroidetes and increased F/B ratio in the intestinal tract.

Therefore, it is possible that the use of SCFAs, prebiotics, or probiotics (live microbiota) to improve the gut microbiota environment can allow SCFAs to prevent metabolic disorders and prevent ASCVD.

**Short Chain Fatty Acids and Visceral Adipose Tissues**

Adipose tissues not only store energy through the accumulation of triglycerides, but secrete various adipokines that affect metabolism throughout the body. In hypertrophic adipose tissues, the invasion of macrophages activates the secretion of free fatty acids (FFAs), TNF-α, IL-6, MCP-1, plasminogen activator inhibitor-1, and other pro-inflammatory cytokines and chemokines, which leads to metabolic abnormalities, including insulin resistance, resulting in the promotion of atherogenesis. These interactions between adipocyte-derived FFAs and macrophage-derived adipokines represent a vicious cycle.

Using a co-culture system with adipocytes and macrophages, we found increased production of TNF-α, IL-6, MCP-1, and the release of free glycerol and FFAs into the medium. Butyrate (0.1-1.0 mmol/L) significantly reduces these effects. Butyrate inhibits the phosphorylation of mitogen-activated protein kinases and NF-κB activity in co-cultured macrophages and suppresses lipolysis caused by the suppression of adipose triglyceride lipase expression and hormone-sensitive lipase phosphorylation in adipocytes. In these co-culture conditions, butyrate increases the production of PGE2, and approximately 40% of the suppressive effect of butyrate on lipolysis depends on the PGE2-mediated pathway (Fig. 1 (A-C)).

Intraperitoneal administration of butyrate (1.0 g/kg) in db/db mice suppresses obesity-induced inflammation and the expression of IL-1, IL-6, and TNF-α mRNA in epididymal, subcutaneous adipose tissues by inhibiting the NOD-like receptor 3 inflammation signaling pathway. In experiments using explants of human omental and subcutaneous adipose tissues, propionate (3 mmol/L) suppresses the production of resistin, which is an adipocyte-derived adipokine and pro-inflammatory cytokine.

In hypertrophic visceral adipose tissues, a decrease of the CD4 (+) Treg and an increase of the CD153+PD-1+CD44hiCD4+ T cell population ratio have been observed. This change in T cell populations may induce macrophages into adipose tissues as well as decreases in the M2 macrophage population ratio, and an increase in the M1 macrophage population ratio. However, it has not been clarified whether SCFAs are directly associated with changes in the M1/M2 macrophage ratio or in T cell differentiation and population in adipose tissues. This issue therefore requires further study. Finally, mast cells may be involved in the inflammation of visceral adipose tissues prior to macrophage invasion. Nevertheless, the effect of SCFAs on mast cells remains unknown.

**Short Chain Fatty Acids and Metabolic Abnormality**

Several reports have shown that obesity is associated with changes in the relative abundance of the two dominant bacterial phyla, Firmicutes and Bacteroidetes. Increased F/B ratios are observed in the guts of obese adults. Recent cohort studies in Chinese and European women have shown that regardless of the difference in race and eating habits, type 2 diabetes patients are characterized by a moderate degree of gut microbial decrease in the abundance of some universal butyrate-producing bacteria.

Dietary fibers promote metabolic benefits on body weight and glucose control, and several studies have demonstrated the impact of an SCFA-enriched diet, establishing a direct causal relationship between fiber fermentation and improved metabolism in humans. Lin et al. have shown that butyrate, propionate, and acetate protect against diet-induced obesity and insulin resistance and that butyrate and propionate, but not acetate, induce gut hormones and reduce food intake. De Vadder et al. have demonstrated that propionate and butyrate activate intestinal gluconeogenesis via complementary mechanisms. Increased production of acetate by an altered gut microbiota in rodents leads to activation of the parasympathetic nervous system, which promotes increased glucose-stimulated insulin secretion, increased ghrelin secretion, hyperphagia, obesity, and related sequelae. Oral supplementation of SCFAs may thus improve impaired glucose metabolism in humans.

Dietary acetic acid reduces serum cholesterol and...
triglycerides (TG) in rats fed a cholesterol-rich diet\textsuperscript{102}. Propionate inhibits fatty acid synthesis and to a lesser extent cholesterol synthesis, while butyrate is a potent activator of both synthetic pathways in rat isolated liver cells\textsuperscript{103}. A high propionate diet reduces hepatic gene and protein expression of lipogenic enzymes leading to reduced hepatic TG concentrations in high-fat fed mice\textsuperscript{104}. Although it has been reported that a high-cholesterol diet does not alter gut microbiota composition in mice\textsuperscript{105}, further examination is needed to reveal the association between SCFAs and lipid metabolism. Interestingly, Tarini et al. demonstrated in healthy adult humans that supplementation with the fermentable dietary fiber inulin significantly increased postprandial serum acetate, propionate, and butyrate concentrations at 4-6 h, and significantly decreased postprandial serum FFAs concentrations at 4 h. They also showed that inulin significantly increased plasma glucagon-like peptide-1 concentrations at 30 min, and reduced ghrelin at 4.5 h and 6 h\textsuperscript{106}. Thus, compositional changes of microbiota might influence adiposity and glucose and lipid metabolism by regulating food intake.

**Conclusion and Perspectives**

SCFAs may suppress inflammation by reducing migration and proliferation of immune cells, reducing many types of cytokines, and inducing apoptosis. Thus, SCFAs are thought to have anti-inflammatory effects. However, marked changes of SCFAs concentrations in blood or various tissues are thought to cause disorders related to immunological and metabolic imbalances. Thus, gut bacteria exert both beneficial and harmful effects\textsuperscript{107}. Therefore, it may be important to estimate the appropriate concentrations of SCFAs to maintain a normal metabolism and immune system for the prevention and treatment of diseases using diet and SCFAs. Recently, Bergeron et al. demonstrated that a lower carbohydrate diet (39-40% energy) high in resistant starches was associated with higher plasma TMAO levels in spite of reduced postprandial insulin and glucose responses, while there was no difference in TMAO affected by resistant starches when carbohydrate intake was high (51-53% energy)\textsuperscript{108}. It may be necessary to develop food patterns or medications to reduce plasma TMAO concentrations and maintain appropriate concentrations of SCFAs.

Although there have been only a small number of studies thus far, in the future SCFAs may provide new insights into the pathophysiology of inflammatory diseases, and we can expect the development of novel therapeutic strategies against chronic inflammation, metabolic disorders, and atherosclerosis.

**Conflict of Interest**

All authors declare that they have no conflict of interest.

**Sources of Funding**

Some studies in this review were supported by a Grant-in-Aid for Scientific Research (C), JSPS KAKENHI grant number 26350166.

**Author Contributions**

All authors contributed equally to the preparation of the manuscript and approved the final manuscript.

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