

Effect of Depolymerized Pyrodextrin on Human Intestinal Flora

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(Received June 13, 1996; Accepted for publication, September 25, 1996)

Abstract A new depolymerized pyrodextrin (DPD) was produced by hydrolyzing pyrodextrin, a material of indigestible dextrin, with hydrochloric acid and then heating. The effects of this substance was examined in an *in vitro* fermentation test of human intestinal bacteria and in a continuous ingestion test on healthy subjects. The fermentability of the indigestible portion of DPD by 106 bacterial strains from the human intestine was examined *in vitro*. The indigestible portion of DPD was fermented by most strains of genus *Bifidobacterium*, apart from *B. bifidum* and *B. animalis*. The indigestible portion of DPD-II (dextrose equivalent (DE); 33.0), which resulted from advanced acid hydrolysis, was fermented more than the indigestible portion of DPD-I (DE; 23.6). The indigestible portion of these DPDs was not fermented by *Clostridium perfringens*, *C. difficile*, *Escherichia coli*, and staphylococci. After 14 days of daily ingestion of 10 g of DPD-II by seven healthy males, the number of fecal bifidobacteria had increased significantly in comparison with the level before ingestion. However, the number of bacteroides, the predominant bacteria in the human intestine, did not change. These results suggest that DPD, a depolymerized pyrodextrin that was hydrolyzed without much change in the branched binding, may help to increase the number of bifidobacteria in the human intestinal flora.

Key words: depolymerized pyrodextrin; acid hydrolysis; *Bifidobacterium*; intestinal flora

The supply of fermentable carbohydrate is an important factor for the growth of bacteria in the human colon, while the balance of these intestinal bacterial flora is closely related to human health conditions. It has been reported that water-soluble dietary fibers and indigestible oligosaccharides are fermentable and effective for improving the intestinal environment (1, 4).

We previously developed an indigestible dextrin (water-soluble dietary fiber) with an average molecular weight of 1,600, this dextrin is produced by heat and enzyme treatment (9). This dextrin can only be fermented *in vitro* by bacteroides and

by a few species of *Bifidobacterium* (9). Bailey et al (2) reported that the rate of hydrolysis by α -1, 6 glucosidase of "*Lactobacillus bifidus*" did not vary according to the degree of glucose polymerization (DP) in dextran (DP values of 2–5, for an average molecular weight of 342–828). This rate was, however, smaller for DP values greater than 6, which suggests that the fermentation of indigestible dextrin would be increased by depolymerizing. Thus we developed a new depolymerized pyrodextrin (DPD) by hydrolyzing pyrodextrin, a material of indigestible dextrin, with hydrochloric acid and then heating (5). The average molecu-

lar weight of the indigestible portion was 900–520. We examined the fermentability of the indigestible portion of DPD *in vitro* and the fermentability of DPD using a continuous ingestion test on humans.

MATERIALS AND METHODS

1. *Preparation of the depolymerized pyrodextrin samples.* Hydrochloric acid (670 ppm) was added to corn starch, which was then roasted at 145°C for 30 minutes to produce pyrodextrin. The indigestible portion was determined to be 51.5% of the content by a revised method combining Prosky et al's method (11) and a high-performance liquid chromatographic method proposed by Ohkuma et al (10). The pH value of a 30% aqueous solution of this pyrodextrin was adjusted to 1.6–1.9 by adding 10% hydrochloric acid; the solution was then hydrolyzed by heating at 125°C for 20 minutes under a pressure of 1.25 kgf/cm². Next, the solution was decolorized with activated charcoal, desalted with ion-exchange resin and concentrated. We obtained two DPDs (DPD-I and DPD-II). In the two DPDs, the dextrose equivalent (DE) was 33.0 and 23.6, the indigestible portion 43.5% and 43.7%, and the average molecular weight of the indigestible portion 520 and 900, respectively (Table 1). The proportion of the binding site, analyzed by gas chromatography after methylation, is shown in Table 2. Straight binding was decreased, while branched binding was hardly affected by acid hydrolysis.

2. *Fermentation of the depolymerized pyrodextrin by intestinal bacteria in vitro.* Both DPD-I and DPD-II were hydrolyzed with glucoamylase (Gluczyme NL-4; Amano Pharmaceutical Co., Japan) at 55°C and pH 4.5 for 24 hours; the remaining glucose was removed by passing through a chromatographic column (CCS-10-A; Hitachi, Japan). The two indigestible DPD portions were examined for their fermentability with the human intestinal-origin bacteria shown in Tables 3–5. Fructo-oligosaccharide (Meiologo-P, Meiji Seika Kaisha, Japan), a typically fermentable oligosaccharide, and glucose were used as positive controls.

After culturing these strains on BL agar (Nissui

Table 1. Basic properties of depolymerized pyrodextrin

Items	Depolymerized pyrodextrin	
	DPD-I	DPD-II
DE	23.6	33.0
Content (%) of indigestible portion	43.7	43.5
Average molecular weight	900	560

Table 2. Proportion of binding sites (%) in pyrodextrin and depolymerized pyrodextrin

Binding site	Pyrodextrin ¹⁾	Depolymerized pyrodextrin	
		DPD-I	DPD-II
Terminal	16.5	27.7	46.9
1 → 4	65.1	54.3	36.3
1 → 6	4.6	5.5	7.0
1 → 4, 1 → 6	6.6	5.9	4.2
1 → 3	3.2	3.5	3.3
1 → 3, 1 → 4	0.9	0.7	—
1 → 2, 1 → 4	1.6	1.3	0.9
Others	1.5	1.1	1.4

¹⁾ Pyrodextrin produced by adding hydrochloric acid to corn starch and roasting at 145°C for 30 min.

Pharmaceutical Co., Japan), the bacteria were incubated in GAM broth (Nissui Pharmaceutical Co., Japan) supplemented with Fieldes solution (6) at 37 °C for 24 hours under anaerobic conditions. The bacteria were again incubated on PYF semi-fluid agar (6) with 0.5% test carbohydrate at 37°C for 4 days under anaerobic conditions (7). The pH value of each culture medium was measured, and the degree of fermentation was evaluated according to the criteria given in Table 3.

3. *Fermentation of the depolymerized pyrodextrin samples during continuous ingestion in vivo.* DPD-II (10 g) was ingested by seven healthy adult male volunteers (mean age: 43.3 ± 5.0 years) once a day after breakfast (9 : 00 a.m.) for 14 days. Their feces were collected on the day before starting ingestion, on the 14th day of ingestion, and on the 14th day after stopping ingestion.

Bowel movement conditions and gastrointestinal symptoms were also examined by questionnaire at the time of fecal collection (Table 6). During the experiment, the consumption of any food

Table 3. Fermentability of depolymerized pyrodextrin by *Bacteroides*, *Bifidobacterium* and *Lactobacillus*

Bacterial strain	B ¹⁾	G ²⁾	F ³⁾	DPD-I ⁴⁾	DPD-II ⁵⁾
<i>Bacteroides</i>					
<i>distasonis</i> GAI# 5462	—	+	±	±	±
<i>fragilis</i> GAI# 5524	—	+	++	+	+
<i>fragilis</i> GAI# 5562	—	++	++	+	+
<i>fragilis</i> CIFL N0058	—	++	++	+	+
<i>intermedius</i> CIFL N0074	—	+	±	±	+
<i>ovatus</i> ICM 5824	—	+	++	+	+
<i>ovatus</i> CIFL N0029	—	+	++	+	+
<i>thetaiotaomicon</i> GAI# 5628	—	+	++	+	+
<i>thetaiotaomicon</i> CIFL N010	—	+	++	+	++
<i>uniformis</i> GAI# 5466	—	+	++	+	+
<i>vulgatus</i> GAI# 5460	—	++	+	±	+
<i>vulgatus</i> CIFL N0107	—	++	++	±	+
<i>vulgatus</i> CIFL N0109	—	++	—	±	+
<i>Bifidobacterium</i>					
<i>adolescentis</i> CIFL N0035	—	+++	+++	—	+++
<i>adolescentis</i> CIFL N0037	—	+++	++	++	+++
<i>adolescentis</i> CIFL N0038	—	+++	++	++	+++
<i>adolescentis</i> CIFL N0046	—	+++	+++	++	+++
<i>adolescentis</i> CIFL N0077	—	+++	+++	±	++
<i>animalis</i> CIFL N0040	—	+++	+++	—	±
<i>bifidum</i> CIFL N0067	—	+++	—	—	—
<i>bifidum</i> CIFL N0089	—	+++	—	—	—
<i>breve</i> CIFL N0055	—	+++	++	±	+++
<i>breve</i> CIFL N0078	—	+++	+++	+	+++
<i>breve</i> CIFL N0088	—	+++	++	—	++
<i>breve</i> CIFL N0110	—	+++	+++	++	+++
<i>infantis</i> CIFL N0050	—	+++	++	—	+
<i>longum</i> CIFL N0036	—	+++	++	—	+
<i>longum</i> CIFL N0044	—	+++	++	±	++
<i>longum</i> CIFL N0051	—	+++	+++	—	++
<i>longum</i> CIFL N0052	—	+++	++	—	+
<i>longum</i> CIFL N0057	—	+++	++	—	++
<i>Lactobacillus</i>					
<i>acidophilus</i> CIFL A0038	—	+++	+++	—	+
<i>acidophilus</i> CIFL A0042	—	+++	+	—	++
<i>casei</i> CIFL A0037	—	+++	—	—	+
<i>casei</i> CIFL A0039	—	+++	—	—	++
<i>fermentum</i> CIFL A0066	—	+++	±	—	—
<i>fermentum</i> JCM 1173	—	+++	±	—	—
<i>gasseri</i> JCM 1131	—	+++	+++	—	++
<i>salivarius</i> CIFL A0041	—	+++	+++	—	—
<i>salivarius</i> CIFL A0043	—	+++	+++	—	—

¹⁾ B, blank; ²⁾ G, glucose; ³⁾ F, fructooligosaccharide; ⁴⁾ DPD-I, depolymerized pyrodextrin with DE 23.6;⁵⁾ DPD-II, depolymerized pyrodextrin with DE 33.0.

pH level	Degree of fermentation
6.0 ≤ pH	—
5.5 ≤ pH < 6.0	±
5.0 ≤ pH < 5.5	+
4.5 ≤ pH < 5.0	++
pH < 4.5	+++

Table 4. Fermentability of depolymerized pyrodextrin by *Clostridium*, *Eubacterium*, *Fusobacterium*, *Megamonas* and *Mitsuokella*

Bacterial strain	B ¹⁾	G ²⁾	F ³⁾	DPD-I ⁴⁾	DPD-II ⁵⁾
<i>Clostridium</i>					
<i>butyricum</i> GAI# 7503	—	+++	++	+	++
<i>butyricum</i> CIFL N0065	—	+++	++	+	++
<i>cadaveris</i> CIFL N0047	—	+	—	—	—
<i>clostridioforme</i> GAI# 5458	—	+	+	—	—
<i>clostridioforme</i> CIFL N0062	—	+	±	—	—
<i>difficile</i> GAI# 10038	—	+	—	—	—
<i>difficile</i> CIFL N0013	—	+	—	—	—
<i>difficile</i> GAI# 10042	—	±	—	—	—
<i>difficile</i> GAI# 10037	—	++	—	—	—
<i>hystlyticum</i> CIFL N0071	—	—	—	—	—
<i>innocuum</i> GAI# 5472	—	+++	±	—	—
<i>novyi</i> (Type A) GAI# 5614	—	++	—	—	—
<i>paraputrificum</i> CIFL N0061	—	+++	—	—	—
<i>paraputrificum</i> CIFL N0098	—	++	—	—	—
<i>perfringens</i> GAI# 5526	—	++	—	—	—
<i>perfringens</i> CIFL N0054	—	++	±	—	—
<i>perfringens</i> CIFL N0091	—	+	—	—	—
<i>perfringens</i> CIFL N0092	—	++	—	—	—
<i>perfringens</i> CIFL N0096	—	++	—	—	—
<i>ramosum</i> CIFL N0048	—	++	—	—	—
<i>ramosum</i> CIFL N0097	—	+++	±	—	++
<i>septicum</i> GAI# 7502	—	+	—	—	—
<i>tertium</i> GAI# 5618	—	++	—	±	±
<i>sordellii</i> GAI# 5612	—	+	—	—	—
<i>sporogenes</i> GAI# 5562	—	±	—	—	—
<i>Eubacterium</i>					
<i>aerofaciens</i> CIFL N0070	—	++	+	—	—
<i>aerofaciens</i> CIFL N0095	—	+++	±	—	—
<i>lentum</i> CIFL N0059	—	—	—	—	—
<i>limosum</i> CIFL N0068	—	+++	—	—	—
<i>limosum</i> CIFL N0104	—	+++	—	—	—
<i>nitritogenes</i> CIFL N0085	—	+++	—	—	—
<i>Fusobacterium</i>					
<i>mortiferum</i> GAI# 5442	—	+++	—	—	—
<i>varium</i> GAI# 5566	—	+	—	—	—
<i>varium</i> CIFL N0084	—	+	—	—	—
<i>Megamonas</i>					
<i>hypermegas</i> CIFL N0060	—	+++	++	—	+
<i>Mitsuokella</i>					
<i>multiacida</i> CIFL N0105	—	+++	+++	—	+

¹⁾ B, blank; ²⁾ G, glucose; ³⁾ F, fructooligosaccharide; ⁴⁾ DPD-I, depolymerized pyrodextrin with DE 23.6;

⁵⁾ DPD-II, depolymerized pyrodextrin with DE 33.0.

pH level	Degree of fermentation
6.0 ≤ pH	—
5.5 ≤ pH < 6.0	±
5.0 ≤ pH < 5.5	+
4.5 ≤ pH < 5.0	++
pH < 4.5	+++

Table 5. Fermentability of depolymerized pyrodextrin by intestinal flora

Bacterial strain	B ¹⁾	G ²⁾	F ³⁾	DPD-I ⁴⁾	DPD-II ⁵⁾
<i>Peptostreptococcus</i>					
<i>magnus</i> GAI# 5528	-	-	-	-	-
<i>micros</i> GAI# 5540	-	-	-	-	-
<i>asaccharolytica</i> CIFL N0080	-	±	-	-	-
<i>prevotii</i> CIFL N0081	-	-	-	-	-
<i>Propionibacterium</i>					
<i>acnes</i> GAI# 5468	-	+++	-	-	-
<i>granulosum</i> CIFL N0083	-	++	-	-	-
<i>Veillonella</i>					
<i>parvula</i> GAI# 5602	-	-	-	-	-
<i>parvula</i> CIFL N0087	-	-	-	-	-
<i>Citrobacter</i>					
<i>diversus</i> CIFL A0016	-	+	-	-	-
<i>freundii</i> CIFL A0015	-	++	-	-	-
<i>Enterobacter</i>					
<i>cloacae</i> CIFL A0001	-	±	-	-	-
<i>Enterococcus</i>					
<i>faecalis</i> CIFL A0013	-	+++	-	±	+
<i>faecalis</i> CIFL A0033	-	+++	+++	-	±
<i>faecalis</i> CIFL A0035	-	+++	+++	±	++
<i>faecium</i> CIFL A0034	-	+++	++	-	±
<i>faecium</i> CIFL A0036	-	+++	+++	-	±
<i>Escherichia</i>					
<i>coli</i> CIFL A0008	-	++	-	-	-
<i>coli</i> CIFL A0044	-	+	-	-	-
<i>coli</i> CIFL A0045	-	++	-	-	-
<i>coli</i> CIFL A0046	-	+++	-	-	-
<i>coli</i> CIFL A0047	-	++	±	-	-
<i>Klebsiella</i>					
<i>pneumoniae</i> CIFL A0003	-	++	+	-	-
<i>pneumoniae</i> CIFL A0020	-	+	+	-	±
<i>Morganella</i>					
<i>morganii</i> CIFL A0023	-	+	-	-	-
<i>Proteus</i>					
<i>mirabilis</i> CIFL A0009	-	++	±	-	-
<i>vulgaris</i> CIFL A0011	-	++	±	-	-
<i>Serratia</i>					
<i>marcescens</i> CIFL A0007	-	++	±	-	-
<i>Staphylococcus</i>					
<i>aureus</i> CIFL A0012	-	+++	++	-	-
<i>epidermidis</i> CIFL A0018	-	++	+	-	-
<i>haemolyticus</i> CIFL A0014	-	++	++	-	-

¹⁾ B, blank; ²⁾ G, glucose; ³⁾ F, fructooligosaccharide; ⁴⁾ DPD-I, depolymerized pyrodextrin with DE 23.6;

⁵⁾ DPD-II, depolymerized pyrodextrin with DE 33.0.

pH level	Degree of fermentation
6.0 ≤ pH	-
5.5 ≤ pH < 6.0	±
5.0 ≤ pH < 5.5	+
4.5 ≤ pH < 5.0	++
pH < 4.5	+++

Table 6. Scores of bowel, stomach and intestinal conditions in questionnaire

Bowel conditions
Evacuation frequency
4: more than 2 times per day
3: 1 time per day
2: 1 time per 2 days
1: 1 time per 3 days or irregular
Stool volume
4: large
3: moderate
2: small
1: very small or none
Stool state
4: soft
3: rather soft
2: normal
1: diarrhea or hard
Conditions after evacuation
4: refreshed
3: normal
2: feeling of residual soft stool
1: feeling of residual hard stool
Stomach and intestinal conditions
a. desire to evacuate but could not evacuate and felt gripping pain
b. rumbling bowel
c. abdominal inflation
d. gas in the bowel
e. nausea

that might influence the intestinal flora (*e.g.*, dairy products) and the use of medicines (*e.g.*, antibiotics) were prohibited. This human study was approved by the board of ethics at Matsutani Chemical Industry Company, Ltd. (approval no. 95-01).

The collected feces were examined for intestinal flora according to the method of Mitsuoka et al (6) (Table 7); the number of bacteria per gram of feces being expressed in logarithmic values. The percentage (%) of *Bacteroides* and *Bifidobacterium* in the total bacterial count in the feces was also calculated.

4. *Statistical analysis.* The results of the experiments are expressed as means \pm standard error (SEM). The Student's *t*-test (change in fecal flora and change of percentage of main intestinal bacteria with continuous ingestion test) and Friedman's test (change of bowel movement with

Table 7. Culture media and incubation methods (6)

Medium	Main micro-organisms enumerated	Incubation method
EG agar	predominantly anaerobes	anaerobic
BL agar	predominantly anaerobes	anaerobic
TS agar	predominantly aerobes	aerobic
BS agar	<i>Bifidobacterium</i>	anaerobic
ES agar	<i>Eubacterium</i>	anaerobic
NBGT agar	Bacteroidaceae	anaerobic
NN agar	<i>Clostridium perfringens</i>	anaerobic
VS agar	<i>Veillonella</i>	anaerobic
LBS agar	<i>Lactobacillus</i>	anaerobic
DHL agar	Enterobacteriaceae	aerobic
TATAC agar	<i>Streptococcus</i>	aerobic
PEES agar	<i>Staphylococcus</i>	aerobic
P agar	Yeast	aerobic

continuous ingestion test) were used to evaluate the differences between groups; these differences were considered significant for *p* values less than 5%.

RESULTS

1. Fermentation of the Depolymerized Pyrodextrin by Intestinal Bacteria in Vitro

Table 3-5 shows the fermentability of the indigestible portion of the DPDs by 106 stock strains of human intestinal bacteria. The indigestible portion was fermented by many strains of genus *Bifidobacterium*, with DPD-II (DE 33.0) particularly being fermented by *Bifidobacterium* spp. excluding *B. bifidum* and *B. animalis*. The degree of fermentation of the indigestible portion of DPD-II was higher than that of DPD-I (DE 23.6). The degree of fermentation of these indigestible portions by bacteroides was comparable or slightly inferior to that of glucose. Both indigestible portions were fermented by *Clostridium butyricum* and *C. ramosum*, but not by *C. perfringens* or *C. difficile*. The indigestible portion of DPD-II was fermented by enterococci, *Lactobacillus acidophilus*, *L. casei*, and *L. gasseri*. Both indigestible portions of DPDs were not fermented *E. coli*, or staphylococci.

2. Fermentation of the Depolymerized Pyrodextrin Samples during Continuous Ingestion in Vivo

The changes in the intestinal flora counts (log number of colony forming unit/g of feces) and in

Table 8. Change in fecal flora with ingestion of depolymerized dextrin

Organism	Before ingestion	During ingestion	After ingestion
Total	10.7 ± 0.1	10.8 ± 0.1	10.8 ± 0.1
Bacteroidaceae	10.5 ± 0.1	10.5 ± 0.1	10.4 ± 0.1
<i>Eubacterium</i>	10.1 ± 0.1	10.2 ± 0.1	10.1 ± 0
<i>Bifidobacterium</i>	9.8 ± 0.2	10.1 ± 0.1*	9.9 ± 0.1
Peptococcaceae	9.4 ± 0.2	9.6 ± 0.2	9.6 ± 0.2
<i>Streptococcus</i>	6.0 ± 0.7	6.8 ± 0.5	6.1 ± 0.6
Enterobacteriaceae	7.1 ± 0.3	7.3 ± 0.3	7.8 ± 0.3
<i>Lactobacillus</i>	6.5 ± 0.8 (86)	6.0 ± 1.1 (86)	5.0 ± 0.7
<i>Veillonella</i>	6.3 ± 0.5 (86)	6.5 ± 0.4 (71)	6.6 ± 0.6 (86)
<i>Clostridium perfringens</i>	3.1 ± 0.6 (43)	3.2 ± 0.8 (43)	3.3 ± 0.4 (86)
<i>Clostridium</i> (others)	9.1 ± 0.3 (57)	9.8 ± 0.1 (57)	9.6 ± 0.1 (43)
<i>Staphylococcus</i>	4.8 ± 0.5 (29)	2.3 (14)	2.3 (14)
Yeasts	3.4 ± 0.2 (29)	2.8 ± 0.5 (29)	3.7 ± 0.8 (57)

Values are mean ± SEM of log number of bacteria/g of wet feces (frequency of occurrence) in 7 volunteers. Statistically significant at * $p < 0.05$ levels when compared with the values before ingestion.

Table 9. Change in the percentage of main intestinal bacteria with ingestion of depolymerized pyrodextrin

Organism	Before ingestion	During ingestion	After ingestion
<i>Bifidobacterium</i>	13.4 ± 2.2	19.8 ± 2.6*	15.2 ± 1.4
Bacteroidaceae	54.9 ± 2.9	44.5 ± 2.4*	49.3 ± 2.5
Others	31.7 ± 2.4	35.6 ± 3.4	35.5 ± 3.4

Values are mean ± SEM of percentage to the number of total intestinal bacteria in 7 volunteers. Statistically significant at * $p < 0.05$ levels when compared with the values before ingestion.

Table 10. Change of bowel movement with ingestion of depolymerized pyrodextrin

Items	Before ingestion	During ingestion	After ingestion
Evacuation frequency ¹⁾	3.29 ± 0.18	3.71 ± 0.18	3.29 ± 0.18
Stool volume ²⁾	3.00 ± 0.00	3.14 ± 0.14	2.86 ± 0.14
Stool state ³⁾	3.14 ± 0.34	3.00 ± 0.31	3.00 ± 0.31
Conditions after evacuation ⁴⁾	3.14 ± 0.14	3.57 ± 0.20	3.14 ± 0.14

¹⁾⁻⁴⁾ See the scores for each condition in Table 6. Values are mean ± SEM of 7 volunteers.

the percentages (%) of intestinal flora are shown in Tables 8 and 9, respectively. The number of bifidobacteria increased significantly after 14 days of daily ingestion of DPD-II, while the number of bacteroides did not change throughout the experiment; the percentage of bacteroides decreased significantly after the ingestion of DPD-II.

The bowel movement score did not change

throughout the experiment (Table 10). From the results of the questionnaire about stomach and intestinal conditions, no clinical problems such as diarrhea were apparent in any subject under these conditions. One subject complained of abdominal inflation on the first day of ingestion, but this symptom disappeared after 3 days without any medical care. Two subjects complained of gas in

the bowel, although this was not clinically serious.

DISCUSSION

We developed a new depolymerized pyrodextrin (DPD) that was hydrolyzed without much change in the branched binding and with hardly any decrease in the indigestible portion content. The fermentability of DPD by human intestinal bacteria was studied, and bifidobacteria fermented the indigestible portion of the DPD samples. Compared with the indigestible portion of DPD-I, the indigestible portion of DPD-II, which was more acid hydrolyzed, was fermented by a greater number of bifidobacteria species tested and underwent a higher degree of fermentation in the *in vitro* fermentability test. The degree of fermentation of the indigestible portion of the DPDs by bacteroides was the same or lower than that of fructooligosaccharides and glucose, although there was almost no difference in the degree of fermentation between the indigestible portions of DPD-I and -II. The indigestible portions of these DPDs were not fermented by *E. coli*, or clostridia, except for *Clostridium butyricum*. These results suggest that, as depolymerization was advanced by acid hydrolysis, utilization by *Bifidobacterium* increased.

Water-soluble dietary fibers such as pectin and guar gum are known to be fermentable saccharides. Although their molecular weights are greater than 50,000, the indigestible portion of DPDs I and II is 900 and 520, respectively, which is slightly longer than oligosaccharides. DPD-II (average molecular weight of indigestible portion = 520) was fermented better than DPD-I (average molecular weight of indigestible portion = 900). Thus, we think these DPDs are fermented better than pectin and guar gum like oligosaccharides.

The indigestible portion of DPD-II was fermented by some strains of enterococci and lactobacilli. *L. casei* has been reported, as well as an increasing number of intestinal bifidobacteria (15), to reduce urinary indican, phenol and *p*-cresol (16), and to facilitate immunopotential (17). Fructooligosaccharide and the indigestible portion of DPD-I were not fermented by *L. casei*. However, the indigestible portion of DPD-II was fermented

by *L. casei*: the difference in this fermentability possibly being one of the characteristics of DPD-II used in this experiment.

The number of fecal bifidobacteria at 14 days after the ingestion of 10 g/day of DPD-II by healthy adults was significantly higher than that before ingestion. However, the number of *Bacteroides* did not change throughout the experiment (Table 8). Consequently, the percentage of bifidobacteria to the number of total intestinal bacteria increased from 13.4% to 19.8%, and the percentage of bacteroidaceae decreased significantly from 54.9% to 44.5% (Table 9). These results suggest that DPD-II selectively enhanced the proliferation of bifidobacteria.

The bowel movement condition of the subjects was not affected by the daily ingestion of DPD-II. This was probably due to all the subjects being healthy males who already had good bowel movement before the ingestion, so that there was no room for improvement, or the ingestion period might have been too short to elicit any change. Although bifidobacteria are known to suppress the production of intestinal putrefaction products by suppressing the growth of putrefying bacteria (3), we think that DPD-II has the potential to maintain a good intestinal environment by increasing the number of bifidobacteria. We intend to examine continuous ingestion by various subjects under different conditions again in the future.

Some oligosaccharides of fructooligosaccharide (3), galactooligosaccharide (14), lactulose (12), raffinose (13), etc., are well-known as energy sources for bifidobacteria. These are all oligosaccharides with a degree of polymerization (DP) between 2 and 5, and are mainly used as sweetening agents because of their sweetness. Although DPD is a corn syrup, it has useful and different physical properties from oligosaccharides such as its low sweetness, high freezing point, high viscosity and low crystallization (8). We consider that DPD can be used in various foods as a corn syrup to selectively enhance the growth of *Bifidobacterium*.

Acknowledgment. We are deeply grateful to the

Intestinal Flora Laboratory of Calpis Food Industry Co. Ltd. for performing the fermentation test.

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