329

Mannooligosaccharides Blended Coffee Beverage Intake Increases the Fat Level in Feces

Toshio Kumao* and Shigeyoshi Fujii

 $Research\ and\ Development, A jinomoto\ General\ Foods, Inc., Sanken\ Bldg., 3-25-1, Hyakunin-cho, Shinjuku-ku, Tokyo\ 169-0073, Japan$

(Received March 17, 2006; Accepted March 25, 2006)

Liquid coffee and milk based liquid coffee, both containing 1.0 g of mannooligosaccharides (MOS) from coffee mannan, were administered to two groups of six subjects each. The subjects consumed one or the other of the beverage everyday for two weeks. The level of fat in their excrement was subsequently analyzed. In both liquid coffee (p < 0.05, respectively) and milk based liquid coffee (p < 0.05, respectively), the concentration of the beverage containing MOS intake showed a significant decrease in comparison with the placebo and interval or those who did not drink any coffee. The result suggests that the intake of MOS can increase the level of fat excreted from the body irrespective.

Key words —— coffee beverage, mannooligosaccharide, fat level, feces

INTRODUCTION

We succeeded in extracting mannooligaosaccharides (MOS) from coffee spent, an industrial waste.¹⁾ The previous studies confirmed that MOS has an intestinal conditioning effect by improving the intestinal microbiota.^{1–3)} Further investigations have been undertaken to explore the potential functionality of MOS. The liquid coffee beverage containing 3.0 g/day of MOS, assisted in reducing the areas of subcutaneous and visceral fat in somewhat overweight adults when consumed for 12 weeks.^{4,5)} In addition, rats fed diet containing MOS excreted more fat content in their stools than their counterparts fed regular diet free of MOS.⁶⁾ These results suggested that MOS was effective in lowering the level of body fat, the mechanism of which partly involves a greater concentration of fat in the excrement. This study was carried out to confirm this effect in humans. The liquid coffee beverage and the milk based coffee beverage containing MOS were administered to the subjects whose fecal samples were analyzed relative to the level of fat.

MATERIALS AND METHODS

Preparation of MOS and the Test Drinks —— The MOS were prepared according to the methods of Asano *et al.* Test drinks (the liquid coffee drink and milk based coffee drink) were prepared according to the methods of Kumao *et al.*⁷⁾ The amount of MOS present in the former and latter drinks was 1.0 g/280 ml and 1.0 g/300 ml, respectively. The placebo was prepared so that its composition would be identical to that of the drinks containing MOS except that digestive maltooligosaccharides were added instead of MOS.

Screening of the Subjects — After approval of the clinical test by Koei group Institutional Review Board, the subjects were provided with a full account of this experiment in advance, including the purposes and contents thereof. The subjects agreed to participate in this clinical test in writing. The test was performed based on the guidelines of the Helsinki Declaration, under the supervision of a qualified clinical trial physician. Twelve healthy female individuals were selected based on a questionnaire, whose responses showed an almost identical pattern in defecation (stool size and frequency).

Test Schedule and Sample Collection — The subjects were divided into two groups of six each and were administered different drinks accordingly (Average age: 44.0 ± 4.6 for the liquid coffee without milk; 38.7 ± 3.9 for the liquid coffee with milk). In addition, each group was subdivided into Groups A and B of three each. Group A was requested to consume the drinks containing MOS everyday for

^{*}To whom correspondence should be addressed: Research and Development, Ajinomoto General Foods, Inc., Sanken Bldg., 3–25–1, Hyakunin-cho, Shinjuku-ku, Tokyo 169–0073, Japan. Tel.: +81-3-5338-6741; Fax: +81-3-5338-6777; E-mail: Toshio_ Kumaoh@agf.co.jp

two weeks and then the placebo for two weeks after a two-week interval between these intake periods. Contrarily, Group B consumed the placebo first for two weeks, and then the MOS drinks for two weeks after the two-week interval between the periods. This experiment was designed to be a double-blind crossover study.

In each period of sample consumption, the subjects were asked to consume MOS or placebo drinks with a volume of 280 and 300 ml/day for those with and without milk, respectively. Thus, the amount of MOS administered was adjusted at 1.0 g/day for the groups administered the test drinks. There was no dietary control imposed on the subjects, except that they were instructed to refrain from taking medicine or consuming extremely high fat foods. Other instructions included avoiding binge eating or drinking or any other irregularities in their dietary habits. The subjects were also asked to briefly record their diets everyday in a questionnaire. Their fecal samples were collected for five days prior to the end of the experiment. The samples were immediately frozen for storage after collection.

Measurement of the Fecal Fat Concentration —— The amount of fat per one gram of feces was calculated to determine the level of fecal fat, according to the methods of van de Kamer *et al.*⁸⁾

Statistical Analysis — The data obtained were expressed as a mean \pm the SEM. A paired Student's *t*-test was employed to compare the data between the sample groups. The level of statistical significance was set at p < 0.05.

RESULTS AND DISCUSSION

The questionnaire confirmed regularity in the subjects' diets throughout the test periods. No harmful effect was observed in the subjects in relation to this test, and this experiment was completed safely without any complications.

Figure 1 shows the changes in the level of fecal fat due to the intake of the liquid coffee drinks containing MOS. The level of fecal fat was significantly higher during the period when the MOS drink was being consumed than the period of placebo drink intake and the interval (p < 0.05, respectively). On the other hand, there was no statistically significant difference in the data between the periods of placebo drink intake and interval. Similarly, the data showed a significantly higher level of fecal fat when the milk based coffee drinks containing MOS were



Fig. 1. Effect of Liquid Coffee Containing MOS on the Fat Level in Feces

Values expressed as the mean \pm SEM of mg/g feces. *Statistically significant from placebo intake (p < 0.05), †Statistically significant from interval (p < 0.05).



Fig. 2. Effect of Milk Based Liquid Coffee Containing MOS on the Fat Level in Feces

Values expressed as the mean \pm SEM of mg/g feces. *Statistically significant from placebo intake (p < 0.05), \dagger Statistically significant from interval (p < 0.05).

being consumed than the periods of placebo intake and interval (Fig. 2) (p < 0.05, respectively). No statistically significant difference was observed between the period of placebo drink intake and the interval with regard to the milk based coffee drink containing MOS (Fig. 2).

It has been reported that the administration of MOS causes propionic acids to increase in the appendix⁹⁾ and that the acids are involved in the inhibition of fat synthesis in the liver.¹⁰⁾ Other studies revealed that MOS can prevent a fatty liver⁶⁾ and that MOS inhibits lipid absorption during meals.¹¹⁾ According to these study, the ability of MOS in lowering the level of body fat seems to involve a plurality of mechanisms. Our studies confirmed that the group of rats fed a diet containing MOS excreted feces with a significantly higher concentration of fat.⁶⁾ In this study, this effect was examined in human subjects by administering the level of fat

excreted in their feces.

According to the previous clinical studies, the daily intake of 3.0 g of MOS was necessary to lower the level of body fat.^{4,5)} However, our studies showed a smaller quantity of MOS, or 1.0 g per day, heightened the level of fat in the excrements. Kumao et al. reported that MOS, when ingested with high fat diets, deterred a surge in the serum triglyceride level and inhibited the absorption of lipids in the intestines.¹¹⁾ Since the intake of oligosaccharides is known to shorten diet retention in the digestive tract,¹²⁾ they are assumed to play more than a small part in the inhibition of lipid absorption. Based on these findings, even an amount less than 3.0 g of MOS per day was potentially effective in reducing the level of lipid absorption. However, as mentioned above, this effect appears to involve complex factors. Further investigations are necessary to endorse 1.0 g of MOS as the criterion to achieving other beneficial effects of MOS.

Previous experiments have focused on the application of MOS in food products. Accordingly, various types of food products have been used in these experiments. This is particularly notable in the case of the development of food products having the intestine conditioning effect of MOS.^{13,14)} In this study, two types of products were administered to the subjects. In both cases, the groups administered the MOS-added drinks showed a significantly high level of fecal fat. This result suggests that dairy products, when mixed with the MOS-added liquid coffee drinks, do not affect the body fat lowering mechanism of MOS, or at the very least only partially.

This study confirmed that the intake of MOS results in a higher level of fecal fat in the excrement. Umemura *et al.* reported that the intake of MOS also increases the volume of feces.¹⁵⁾ Therefore, it is possible that not only the level of fecal fat but also the volume of fat excreted are likely to increase as the result of the intake of MOS. To prove this, clinical tests with a focus on details of fat input and output balance will be necessary. Further investigations are awaited in this regard.

REFERENCES

 Asano, I., Nakamura, Y., Hoshino, H., Aoki, K., Fujii, S., Imura, N. and Iino, H. (2001) Use of mannooligosaccharides from coffee mannan by intestinal bacteria. *Nippon Nogeikagaku Kaishi*, 75, 1077-1083.

- Asano, I., Hamaguchi, K., Fujii, S. and Iino H. (2003) In vitro digestibility and fermentation of mannooligosaccharides from coffee mannan. *Food Sci. Technol. Res.*, 9, 62–66.
- Asano, I., Umemura, M., Fujii, S., Hoshino, H. and Iino, H. (2004) Effects of mannooligosaccharides from coffee mannan on fecal microflora and defecation in healthy volunteers. *Food Sci. Technol. Res.*, **10**, 93–97.
- 4) Asano, I., Fujii, S., Ozaki, K., Takehara, I., Yano, Y. and Fukuhara, I. (2005) Effects of "Coffee Beverage" containing mannooligosaccharides from coffee on human abdominal fat by long term ingestion. *Jap. J. Food Engineering*, 6, 133–141.
- 5) Asano, I., Fujii, S., Kaneko, M., Takehara, I. and Fukuhara, I. (2006) Investigation of mannooligosaccharides blended coffee beverage on abdominal fat reduction in humans. *Jap. J. Med. Pharm. Sci.*, 55, 93–103.
- 6) Kumao, T., Fujii, S., Ozaki, K. and Takao, I. (2005) Effects of diets containing mannooligosaccharides from coffee mannan on fat in blood serum and liver fat in (ZUC)-*fa*/*fa* rats. *Jap. J. Med. Pharm. Sci.*, 54, 505–509.
- 7) Kumao, T., Fujii, S., Ozaki, K., Mutoh, K., Nakamuro, K., Matsushima, T. and Iino, H. (2005) Effect of "Liquid coffee beverage" containing mannooligosaccharides from coffee mannan on defecation and fecal microbiota in healthy volunteers. *Health Sci.*, **21**, 69–76.
- van de Kamer, J. H., ten Bokkel Huinink, H. and Weyers, H. A. (1949) Rapid method for the determination of fat in feces. *J. Biol. Chem.*, **177**, 347– 355.
- Fujii, S., Asano, I. and Kumao, T. (2006) Effects of high cholesterol diets containing mannooligosaccharides from coffee mannan on cecal microbiota, fat level in blood serum and liver in rats. *Health Sci.*, 22, 121–127.
- 10) Daubiol, C., Rousseau, N., Demeure, R., Gallez, B., Taper, H., Declerck, B. and Delzenne, N. (2002) Dietary fructans, but not cellulose, decrease triglyceride accumulation in the liver of obese Zucker *fa/fa* rats. *J. Nutr.*, **132**, 967–973.
- Kumao, T., Fujii, S., Ozaki, K. and Takao, I. (2005) Effects of diets with mannooligosaccharides from coffee mannan on fat in blood serum in healthy volunteers. *Jap. J. Food Engineering*, 6, 301–304.
- 12) Tokunaga, T., Oku, T. and Hosoya, N. (1986) Influence of chronic intake new sweetener fructooligosaccharides (Neosuger) on growth and gastrointestinal function of the rat. J. Nutr. Sci. Vitaminol. (Tokyo), 32, 111–121.

- 13) Umemura, M., Fujii, S., Asano, I., Hoshino, H. and Iino, H. (2004) Effect of coffee mix drink containing mannooligosaccharides from coffee mannan on defecation and fecal microbiota in healthy volunteers. *Food Sci. Technol. Res.*, **10**, 195–198.
- 14) Kumao, T., Fujii, S., Ozaki, K., Mutoh, K., Nakamuro, K., Matsushima, T. and Iino, H. (2005) Effect of liquid coffee containing mannooligosac-

charides from coffee mannan on defecation and fecal microbiota in healthy volunteers. *Health Sci.*, **21**, 137–146.

15) Umemura, M., Fujii, S., Asano, I., Hoshino, H. and Iino H. (2004) Effect of small dose of mannooligosaccharides from coffee mannan on defecating conditions and fecal microflora. *Food Sci. Technol. Res.*, **10**, 174–179.