International Journal of Pharmacy
Journal Homepage: http://www.pharmascholars.com

Original Article **CODEN: IJPNL6**

Effect of cyclodextrin on postprandial blood glucose and triglycerides

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Received on: 06-08-2016; Revised on: 09-09-2016; Accepted on: 20-09-2016

ABSTRACT

The purpose of this study was to evaluate the potential inhibitory effect of α-cyclodextrin (αCD), consumed concurrently with a normal meal, on postprandial blood glucose increase and on triglyceride levels. Subjects were categorized into three groups: meal only (control group), αCD 5 g and meal (αCD group), and γCD 5 g and meal (γCD group). Ten subjects were studied using a crossover design. Compared to the control group, the αCD group showed an obvious blood glucose increase inhibitory effect at 120 minutes after a meal ($p < 0.05$). Furthermore, a significant difference was observed in insulin and triglyceride levels between these two groups. Based on our results, when αCD group was consumed concurrently, αCD demonstrated a postprandial blood glucose increase inhibitory effect, which may lead to the prevention of lifestyle-related diseases.

Key words: α-Cyclodextrin, Blood glucose increase inhibitory effect, Insulin, Triglycerides

INTRODUCTION

In recent years in developed countries, lifestyle-related diseases have become a major cause of mortality and this has been attributed to the increase in smoking, inadequate diet, and a lack of exercise [1]. One proposed method for the prevention of lifestyle-related diseases is glycemic control [2]. Glycemic control can be carried out by drug therapy, exercise therapy, or diet therapy. While exercise therapy and diet therapy are most successful prior to the development of lifestyle-related diseases, it is common to carry out drug therapy after disease development. Drug therapy has a strong immediate effect but is associated with side effects and concerns about patient compliance that may not result in ideal glycemic control. Exercise therapy has benefits in terms of health and stress release. In a study by Liu et al. where patients with type 2 diabetes were randomized to a conventional therapy group (daily drug therapy and diet control) or an intensive therapy group (a combination of aerobic exercise and strength training with conventional therapy), the intensive therapy group showed a significant improvement in fasting and postprandial glucose levels over that shown by the conventional therapy group [3]. However, the effects of exercise therapy do not last long after cessation of treatment and patients in poor physical condition find it difficult to undergo exercise therapy. However, diet therapy can be carried out easily regardless of physical condition by changing a patient's daily diet and eating healthy balanced meals, and a substantial improvement from the baseline health status can be expected.

Glycemic index (GI) is an index of postprandial blood glucose, proposed by Jenkins et al [4]. GI is the percentage of blood glucose increase after food intake, for a specific food, compared to the increased area under the curve (IAUC) for the same amount of

glucose. The GI score of food affects the glucose and insulin levels after meals. Based on the reduction of postprandial blood glucose response by low GI foods, they are known to improve glycemic control, insulin sensitivity, and blood lipid profiles [5]. Studies on the GI of many foods have been conducted. Sugiyama et al. reported to have significantly reduced the GI of rice by ingesting dairy products, such as milk and yogurt, with rice [6]. In addition, Taniguchi et al. were able to inhibit postprandial hyperglycemia and hyperinsulinemia in young healthy subjects by feeding them fermented soybeans with white rice [7]. However, excessive intake of fat may lead to obesity, lipid metabolism abnormalities, arteriosclerosis, and impaired glucose intolerance; therefore, it is important to limit the absorption of dietary fat. Globin proteolysate and tea catechin have been reported to suppress the increase of postprandial blood triglycerides [8,9].

Cyclodextrin (CD) is a cyclic polysaccharide composed of glucose units and has a number of formulation applications including inclusion, masking, and stability. For example, it is used for the formulation of the inclusion complex of itraconazole and hydroxypropyl β-cyclodextrin, to maintain the stability of limaprost and α-cyclodextrin formulation, and as a masking agent to hide the bitterness of active ingredient of drugs [10-12].

In addition, as a food additive, α-cyclodextrin (αCD) can function as dietary fiber. αCD has been reported to inhibit pancreatic amylase activity [13]. It is also resistant to hydrolysis by salivary and pancreatic amylases and is reported to be minimally digested in the small intestine [14]. αCD forms a stable complex with dietary fat that is resistant to lipolysis with lipase; therefore, it has been reported to decrease the absorption and bioavailability of fat, which is excreted unchanged in the feces [15]. Based on these characteristics, αCD is commercially available under the trade name FBCx® to help reduce body fat and manage weight [16].

Instead of drug therapy, αCD consumption with meals may help prevent the onset of lifestyle diseases and allow easy implementation of glycemic control and lipid control in the daily diet. Buckly et al. reported an inhibitory effect of αCD on the post-prandial blood glucose increase after consumption of cooked rice with water [17]. Furthermore Jarosz et al. reported that αCD has a postprandial lipid-lowering effect when used in the tablet form [18]. However, human studies have been limited in analysis of concurrent consumption of a normal meal and αCD and how it affects blood glucose and lipid profiles. The purpose of in this study was to verify the potential inhibitory effect of α-cyclodextrin (αCD), consumed concurrently with a normal meal, on postprandial blood glucose increase and on triglyceride levels.

MATERIALS AND METHODS

Subjects: Subjects were ten healthy adult volunteers who had not received drug treatment and no-smoking (5 male, 5 female). Subjects had an average age of 23 years (range, 21-25 years) and average BMI of 21.5 kg/m^2 (range, 17.4-27.8 kg/m^2)(Table.1). All subjects provided written informed consent, and the study protocol was approved by the ethics committee of Josai University (Sakado, Japan).

Table.1 Subjects characteristics

	Total (n=10)	Male (n=5)	Female (n=5)
Age (years)	22.9 ± 1.8	22.4 ± 2.0	23.4 ± 1.7
Height (cm)	164.0 ± 6.2	67.0 ± 4.2	159.0 ± 5.3
Weight (kg)	56.7 ± 9.8	60.2 ± 11.4	53.2 ± 7.5
BMI (kg/m^2)	21.3 ± 3.7	21.4 ± 3.9	21.1 ± 3.9

Data are presented as the mean ± standard deviation. BMI, body mass index.

Protocol: Three different meal conditions were tested: curry rice and 180 mL of mineral water (control group); curry rice, 180 mL of mineral water, and 5 g αCD (αCD group); and curry rice, 180 mL of mineral water, and 5 g γ-cyclodextrin (γCD) (γCD group)(Table.1). The curry and rice meal used was packaged cooked rice (Sato no Gohan Koshihikari ® 200 g: Sato Foods Industries Co., Ltd., Aichi, Japan) and curry (Ginza Curry ® 200 g Medium Hot: Meiji Co., Ltd., Tokyo, Japan). The αCD and γCD used were prepared by CycloChem Corporation (CycloChem Co., Ltd., Kobe, Japan). Each nutritional component is shown in Table. 1. In a study by Buckly et al. 10 subjects ingested rice with 2 g, 5 g or 10 g of αCD [17]. Although the blood glucose increase inhibitory effect of αCD was seen at 5 g and 10 g, subjects experienced side effects at the 10 g dosage. Therefore, 5 g was the selected dosage

for both αCD and γCD in this study. Three groups of tests on the same subjects (control group, αCD group, and γCD group) were carried out using a random crossover design. Subjects were prohibited from the ingestion of food and drink other than 500 mL of mineral water after the meal until the next meal. Curry rice was ingested within 5 minutes for all subjects followed by up to 200 mL of mineral water. Subjects collected blood 8 times: before the meal and 15, 30, 45, 60, 90, 120, and 180 minutes after ingestion.

Table 2. Nutritional composition of test meal (Ginza Curry)

Ingredient	Ginza Curry	Sato no Gohan
Total Calories (kcal)	223.0	294.0
Protein (g)	6.8	4.2
Fat (g)	13.5	0.0
Carbohydrate (g)	18.4	67.8
Sodium (g)	1.1	0.0

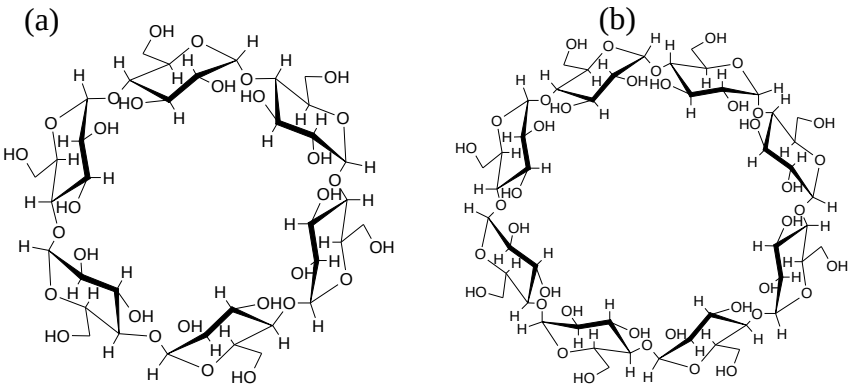

Fig. 1. Chemical Structures of (a) αCD and (b) γCD.

Blood glucose, serum insulin, and blood triglyceride concentrations: Blood was collected from the fingertip using a lancet for self-collection. Blood sugar levels were measured by the subjects by using a blood glucose meter (Glutest Neo Alpha®; Sanwa Kagaku Kenkyusho Co., Ltd., Aichi, Japan). Two hundred microliters of blood was collected in a capillary tube (Hemantlon-L; Minatomedical Co., Ltd., Tokyo, Japan), and centrifuged to obtain plasma. Plasma (50 μL)was stored immediately at -75 ˚C, and plasma insulin and plasma triglyceride levels were measured using an insulin kit (YK060 Insulin ELISA kit®; Yanahira Institute Inc., Shizuoka, Japan) and triglyceride kit (Triglyceride E- Test Wako; Wako Pure Chemical Industries, Ltd., Osaka, Japan), respectively. Insulin and triglyceride levels were measured using a SpectraMax 190 Microplate Reader (Molecular Devices Japan, LLC., Tokyo, Japan).

Statistical analysis: Blood glucose, insulin, and triglyceride levels recorded for the subjects following the consumption of the test meal were analyzed as the Δblood glucose level, Δinsulin level, and Δtriglyceride level compared to the baseline level before the meal. The increase in the area under the curve of blood glucose, insulin, and triglyceride levels was calculated by the respective trapezoidal method. For each test meal, we obtained Δblood glucose, Δinsulin, and Δtriglyceride levels, and these parameters were analyzed using Tukey's test with significance set at $p < 0.05$. Group differences were evaluated with a repeated-measures analysis of variance (ANOVA). All statistical analyses were performed with R (The R Foundation for Statistical Computing). More precisely, it is a modified version

of R commander designed to add statistical functions frequently used in biostatistics.

RESULTS

All 10 subjects completed the study. During the study period, none of the subjects complained of health problems. In addition, none of the subjects noted any differences in the taste of the study diet.

Blood glucose levels: Changes in blood glucose levels in each group are shown in Fig. 2. Blood glucose levels in the control group increased immediately after meals and peaked 30 min after meals. Blood glucose levels in the αCD group increased for up to 45 min after a meal but were consistently lower than levels in the control group from 90 to 180 min after a meal. Blood glucose levels in the γCD group increased for up to 45 min after a meal, and peak blood glucose levels in that group were higher than peak levels in the other two groups. Blood glucose levels in the αCD group 120 min after meals were lower than levels in the control group or the γCD group. Blood glucose levels in the αCD group differed significantly from levels in the control group ($p<0.05$).

The cumulative IAUC for glucose in each group is shown in Fig. 3. Significant differences in the cumulative IAUC for glucose were not noted in any of the three groups. The cumulative IAUC for glucose was smaller in the αCD group at the measured times than the cumulative IAUC for glucose in the control group or the γCD group. There were few differences between the cumulative IAUC for glucose in the control group and that in the γCD group.

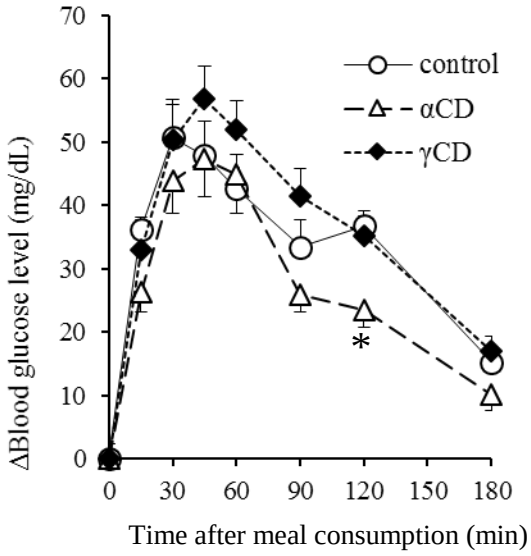

Fig.2 Data are means, with their standard errors represented by vertical bars (respectively n=10). *P < 0.05 vs control (Tukey's test).

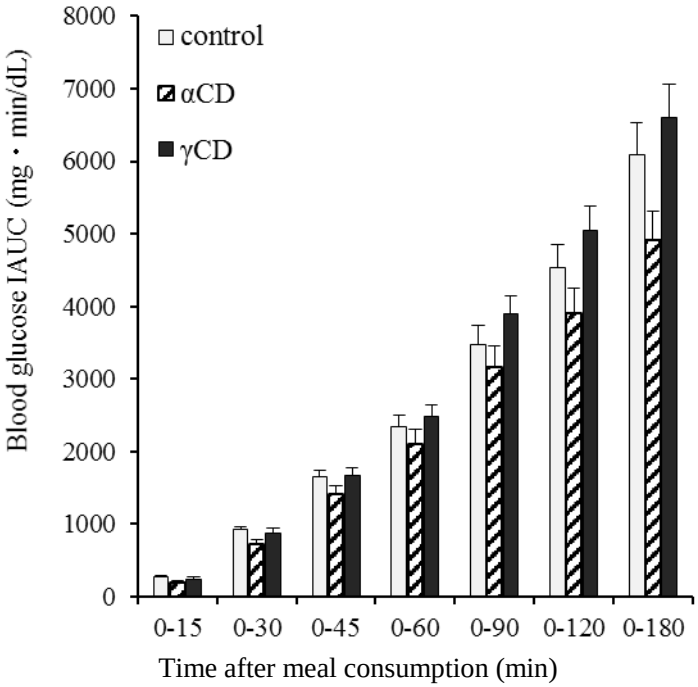

Fig.3 Data are means, with their standard errors represented by vertical bars (respectively n=10).
No significance (Tukey's test).

Serum insulin levels: Changes in serum insulin levels in each group are shown in Fig. 4. Elevated serum insulin levels were evident in each group 30 min after meals. Significant differences in serum insulin levels were not noted in any of the three groups. Plasma insulin levels in the control group 120 min after meals tended to be moderately higher than levels in the other two groups. Substantial differences between the three groups in terms of peak plasma insulin levels were not noted. The cumulative IAUC for insulin in each group is shown in Fig. 5. An increase in the IAUC over time was noted in the control group, the αCD group, and the γCD group. Significant differences in the cumulative IAUC for insulin were not noted in any of the three groups. An increase in the IAUC for insulin was noted in the αCD group 180 min after meals, and this increase tended to be greater than that in the other two groups.

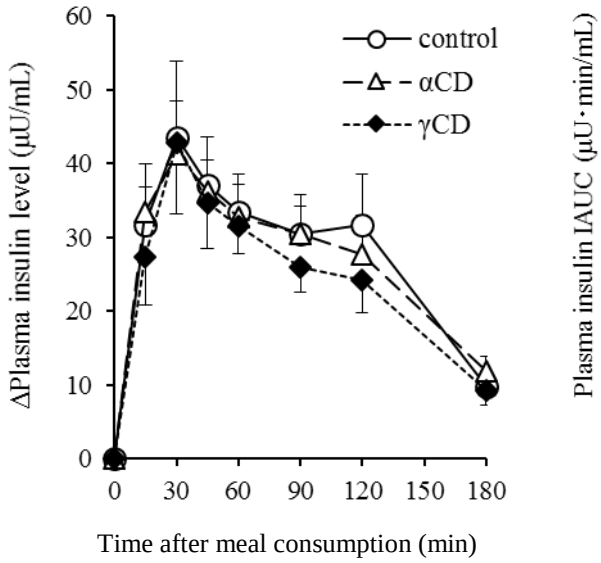

Fig.4 Data are means, with their standard errors represented by vertical bars (respectively n=10). No significance (Tukey's test).

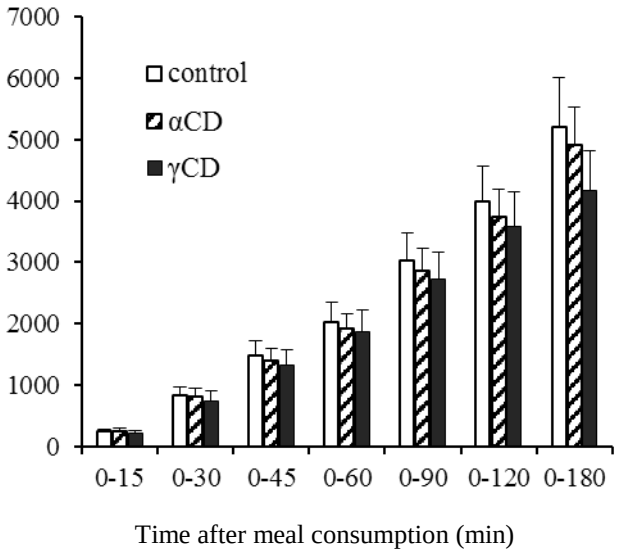

Fig.5 Data are means, with their standard errors represented by vertical bars (respectively n=10). No significance (Tukey's test).

Blood triglyceride levels: Changes in blood triglyceride levels in each group are shown in Fig. 6. Significant differences in blood triglyceride levels were not noted in any of the three groups. Blood triglyceride levels in the γCD group from 60 to 120 min after a meal tended to be lower than levels in the other two groups. Blood triglyceride levels in the γCD group tended to be markedly elevated 180 min after meals. Similar trends were evident in the control group and in the αCD group.

The cumulative IAUC for triglycerides in each group is shown in Fig. 7. An increase in the IAUC over time was noted in the control group, the αCD group, and the γCD group. Significant differences in the cumulative IAUC for triglycerides were not noted in any of the three groups. The IAUC for triglycerides tended to be greater in the control group 180 min after meals than that in the other two groups.

DISCUSSION

In the current study, young, healthy subjects consumed a normal diet along with 5 g of αCD. Results indicated that αCD inhibited an increase in postprandial blood glucose levels. Results also indicated that αCD significantly inhibited an increase in blood glucose 120 min after meals. Buckley et al. added αCD to boiled white rice and they found that this diet inhibited a postprandial increase in blood glucose [17]. When Buckley et al. added 5 g of αCD, they noted a statistically significant difference in the inhibition of a postprandial increase in blood glucose. In the current study, subjects consumed a normal diet along with 5 g of αCD, and results were similar to those reported by Buckley et al.

In addition, Gentilcore et al. reported that αCD slowed gastric emptying and that it delayed intestinal carbohydrate absorption [19]. A drop in blood glucose levels 120 min after meals is presumably the result of delayed intestinal carbohydrate absorption as a result of gastric emptying slowed by αCD and its inhibition of the hydrolysis of complex carbohydrates in the small intestine [19].

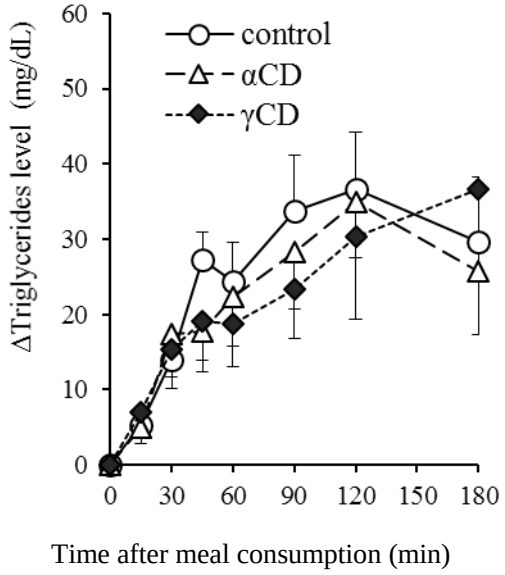

Fig.6 Data are means, with their standard errors represented by vertical bars (respectively n=10). No significance (Tukey's test).

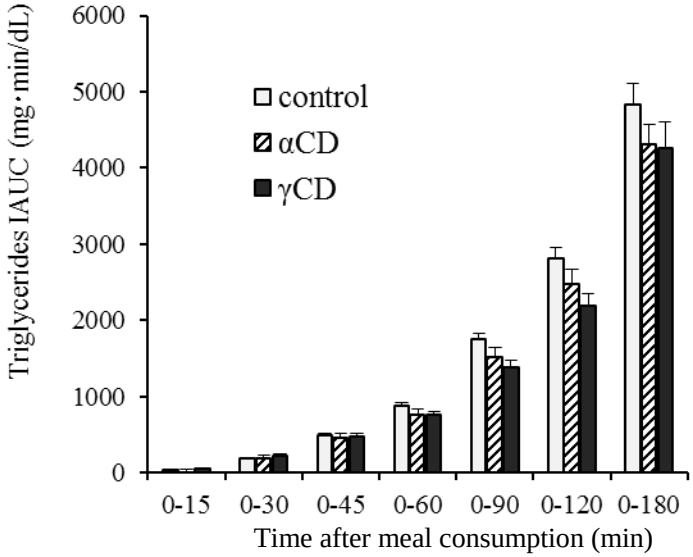

Fig.7 Data are means, with their standard errors represented by vertical bars (respectively n=10). No significance (Tukey's test).

In the current study, an increase in the IAUC for serum insulin tended to be greater in the αCD group 180 min after meals than in the other two groups. This is because amalyase is inhibited by αCD. Complex carbohydrates were broken down and insulin was secreted more slowly in the αCD group than in the other two groups. Buckley et al. noted no significant differences between the amount of αCD added to a diet and the IAUC for insulin [17]. αCD has a function of both a water-soluble dietary fiber and an insoluble dietary fiber. This changes liposolubility substances, such as saturated fatty acid and lipid peroxide, to an insoluble dietary fiber by the inclusion complex form into the CD cave, although αCD is water solubility. Being excreted as facilities has contributed the Inclusion complex. As an interesting thing, the conventional dietary fiber is also having completely different character as property of αCD. Moreover, it is shown that αCD has amylase inhibitory activity which is amylolysis enzyme [20]. Thus Hyperglycemic inhibitory effect by αCD absorption speculated an amylase obstruction action as well as discharge movement restraint in the stomach to have also contributed.

Furune et al. added αCD to simulated intestinal fluid to study cholesterol dissolution in micelles containing lecithin and bile salts [21]. It reported that αCD and lecithin formed complexes and that αCD decreased the micellar solubility of lipids. In the current study, serum triglyceride levels tended to be higher in the αCD group 180 min after meal than levels in the other two groups, so αCD may bind with lecithin in bile salt micelles and inhibit the uptake of lipids by bile salt micelles, thus reducing lipid absorption. Jarosz et al. had healthy adults consume a high-fat diet and 2 tablets containing 1 g of αCD [18]. Jarosz et al. reported that subjects who consumed the tablets containing αCD had lower triglyceride levels 1 hr and 3 hrs after a meal than did controls consuming a placebo [18]. In the current study, significant differences in triglyceride levels were not noted in the three groups. This is presumably because of two factors. One is the amount of fat in the diet: the diet in the study by Jarosz et al. contained 26 g of fat while the diet in the current study contained 13.5 g of fat. The second factor is the substantial difference in triglyceride levels in individual subjects. Triglyceride levels in the γCD group from 60 to 120 min after a meal tended to be lower than levels in the other two groups and triglyceride levels tended to rise

markedly in the γCD group 180 min after meals. These two findings probably warrant an examination of the effect of γCD on triglyceride levels. Muraki et al. reported that the risk of developing type 2 diabetes decreased with greater consumption of raw, fresh fruit but that it increased with greater consumption of fruit juice [22]. However, continuing to consume raw, fresh fruit daily is difficult. If αCD inhibits a postprandial increase in blood glucose, then αCD could be consumed daily simply by consuming αCD powder prior to a meal. The current results suggested that αCD may help to control blood glucose and thus help to prevent lifestyle-related diseases.

Acknowledgments: The authors wish to sincerely thank all of the subjects for their participation in this study.

Conflict of Interest: αCD and γCD that were used in the current study are sold by Cyclochem Co., Ltd. Two of the co-authors of this paper, Keiji Terao and Daisuke Nakata, are employees of Cyclochem.

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