

## The Hypoglycemic Effect of Adlay Diet is not Significant when the Amount of Total Fiber Consumption is Controlled

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### ABSTRACT

The purpose of this study was to investigate the potential hypoglycemic effect of adlay diets when total fiber consumption was controlled in streptozotocin-induced diabetic rats. Forty eight rats were fed for 3 weeks with either control diets or experimental diets : raw milled adlay (RMA), raw whole adlay(RWA), steamed milled adlay(SMA) or roasted milled adlay(OMA). The composition of the AIN-76 diet was modified to ensure the same composition of protein, carbohydrate, fat, and fiber between the control diet and experimental diets. The concentrations of glucose, insulin, glycogen, and protein in plasma, liver, or skeletal muscle were compared. Compared to diabetic control rats, plasma postprandial glucose levels tended to be decreased in RMA, RWA, SMA, and OMA rats until the 2nd week, but no difference was shown at the 3rd week. There was no significant difference in insulin levels among those groups. After glucose loading, the plasma glucose level of SMA was lower than that of diabetic control rats throughout 2 hrs. Liver glycogen was lower than control values in RMA and RWA rats and not different in SMA and OMA rats. The muscle protein level of RMA, RWA, SMA, and OMA rats tended to be lower than in diabetic control rats. There was no significant difference in muscle glycogen among groups. These results suggest that the hypoglycemic effect of an adlay diet is not significant when the amount of total fiber consumption is controlled. (*Korean J Nutrition* 30(9) : 1055~1060, 1997)

**KEY WORDS** : hypoglycemic effect · adlay diet · glucose tolerance.

### Introduction

Over the years, adlay(*Coix Lachryma-Jobi*) has been used as a food source for humans and livestock. It has also been utilized as a diuretic, stomach medicine, analgesic, antispasmodic and hypoglycemic agent in oriental folk medicine. There is no scientific verification of it having any medicinal properties. Recently, physiologically active substances have been isolated that possess antiphlogistic and antitumor promoting activity<sup>1-3</sup>. However, no active hypoglycem-

ic constituent other than dietary fiber has been isolated from adlay<sup>4</sup>. Studies in both humans and animals generally support, the view that dietary fiber lowers postprandial hyperglycemia and insulin response in individuals with and without diabetes<sup>5-7</sup>. Because the dietary fiber content of adlay is not higher or even lower than other grains like barley, millet, or sorghum<sup>8,9</sup> and because adlay is not an economically useful grain, it is important to determine whether an adlay diet shows hypoglycemic activity when total dietary fiber intake is controlled. Glycemic response generally depends on digestibility, which is affected by the source of starch, protein, lipid, dietary fiber, and antinutrients<sup>10</sup>. The cooking method is an espe-

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cially important determinant of postprandial metabolic responses<sup>11</sup>). Thus, as a part of our continuing studies on biological activities of natural products, we have investigated the potential hypoglycemic activities of adlay cooked by different methods when the total fiber consumption was controlled.

## Materials and Methods

### 1. Animals and diets

A total of 48 male Sprague-Dawley rats (Samyang Exp. Animals Co.) of 160–180g were divided into 6 groups of 8 rats with similar body weights: normal-control group, diabetic-control group, diabetic-raw milled adlay group, diabetic-raw whole grain adlay group, diabetic-steamed milled adlay group, and diabetic-roasted milled adlay group.

Diabetes was induced by streptozotocin injection into the tail vein (45mg/kg body weight), which affects the  $\beta$ -cell of pancreas specifically<sup>12</sup>). Animals received 50% adlay diet or control diet for 3 weeks. Control diet was a vitamin-free casein-based semisynthetic diet that met AIN-76 recommendation<sup>13)14)</sup>. The nutritional composition of 50% adlay diet was manipulated to get a composition similar to the control diet (Table 1). All adlay was dried and powdered after cooking. Thus, all experimental diets contained 20% protein, 5% fat, 65% carbohydrates, 5% fiber, and 3850kcal/kg by weight. Prior to initiating experimental diets, rats were given ad libitum access to the control diet for 1 week to adapt to the diet and feeding schedule and to bring all the rats to a similar metabolic status.

**Table 1.** Composition of diet(%)

	Milled adlay		Whole
	Control	Raw, Roasted, Steamed	grain adlay Raw
Casein(vitamin-free)	20.00	12.65	12.81
DL-Methionine	0.30	0.30	0.30
Sucrose	50.00	24.96	29.00
AIN. Vitamin Mixture	1.00	1.00	1.00
AIN. Mineral Mixture	3.50	3.25	2.43
Cellulose	5.00	3.65	3.07
Corn oil	5.00	3.99	1.19
Choline bitartrate	0.20	0.20	0.20
Corn starch	15.00	–	–
Adlay	–	50.00	50.00

### 2. Sample collection

Blood samples were obtained weekly in heparinized tubes from eye veins of fed rats. After 3 weeks of feeding on the respective diets, the oral glucose tolerance test (GTT) was performed after an overnight fast. For GTT, blood glucose level was checked at 0, 30, 60, and 120 minutes after glucose solution (1g/kg BW) was dosed intragastrically using an animal feeding needle. At the end of week 3, animals were anesthetized with ether and sacrificed by decapitation. Immediately following decapitation, blood was collected in heparinized tubes and centrifuged to separate the plasma. Organs were rapidly blotted dry and weighed. Plasma and tissues were stored at  $-70^{\circ}\text{C}$  until analyzed.

### 3. Analysis

All animals were weighed weekly and their food intake was measured daily. Plasma glucose was analyzed with a commercial kit based on enzymatic methods (Youngdong Pharmaceutical Co., Korea). Plasma insulin was analyzed with a commercial kit using the radio immunoassay method immediately after decapitation. Protein was analyzed with a commercial kit based on the Biuret method (Youngdong Pharmaceutical Co., Korea).

Liver and muscle glycogen was measured by a colorimetric procedure<sup>15</sup>). For statistical analysis, all data was first evaluated by analysis of variance. For those F values that were significant, the least significant difference test was performed. A P value  $< 0.05$  was considered to be statistically significant.

## Results

The diabetic status of the rats was evaluated using body weight and feed efficiency ratio (FER) as an indirect, longterm measure and plasma glucose and insulin level as a direct measure. At week 3, the total body weight gain and FER of diabetic rats were significantly lower than those of normal rats (Table 2). Among diabetic rats, there were no differences between diabetic controls and diabetic-adlay groups, regardless of different cooking methods, in body weight and FER.

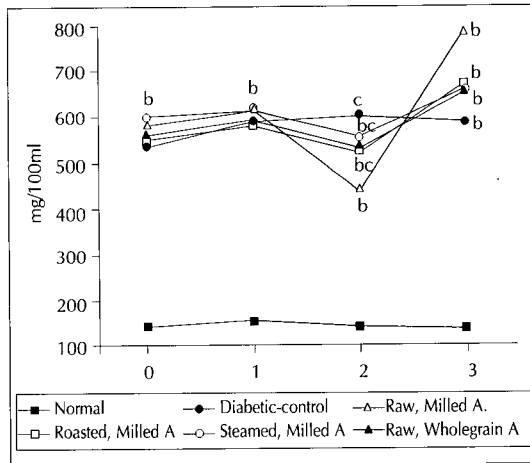
Compared to the normal control group, the di-

**Table 2.** The effect of adlay diet on body weight gain(g) and feed efficiency ratio(FER)<sup>1)2)</sup>

	1st week	2nd week	3rd week	FER <sup>*</sup>
Normal	50.14 ± 13.27	32.57 ± 9.59	8.71 ± 8.77	0.270 ± 0.04 <sup>a2)</sup>
Diabetic-control	17.40 ± 14.70	16.20 ± 18.29	- 2.60 ± 12.41	0.076 ± 0.08 <sup>b</sup>
- Raw milled adlay	22.00 ± 9.97	9.86 ± 15.69	- 11.43 ± 13.10	0.029 ± 0.06 <sup>b</sup>
- Roasted milled adlay	12.67 ± 19.67	18.22 ± 19.85	- 10.44 ± 12.67	0.047 ± 0.10 <sup>b</sup>
- Steamed milled adlay	18.00 ± 9.67	7.38 ± 12.09	- 12.63 ± 10.27	0.018 ± 0.06 <sup>b</sup>
- Raw whole grain adlay	7.56 ± 15.26	12.89 ± 11.20	- 19.67 ± 6.18	0.006 ± 0.03 <sup>b</sup>

1) Values are mean ± SEM, n=8

2) Within a given column, values with different superscripts are significantly different(p < .05)



**Fig. 1.** Effect of adlay diet on the change in plasma glucose level in fed rats (within a given column, values with different superscripts are significantly different(p < .05)).

abetic group showed higher plasma glucose throughout the study(Fig. 1). When compared to diabetic control rats, the plasma glucose level tended to be lower in rats fed raw, roasted, and steamed adlay until the 2nd week. However, the difference was not statistically significant due to large standard deviation, and no difference was shown in those groups at week 3.

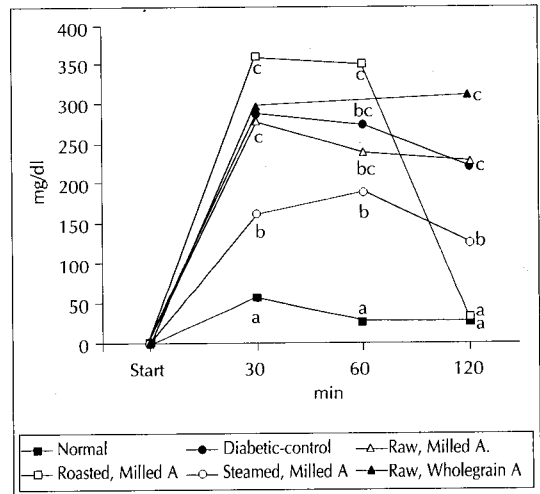
Table 3 shows the level of plasma glucose and insulin in rats fed on adlay diets for 3 weeks. The plasma glucose level in adlay groups was not different from the diabetic control from regardless of cooking method. The plasma level of insulin tended to be even lower in steamed adlay and raw whole grain adlay groups, although this difference was not statistically significant. Fig. 2 shows glucose tolerance curves after oral glucose loading in rats after 3 weeks' experimental diet. Compared to normal control group, abnormal blood glucose tolerance was shown in all diabetic animals. While the glucose tolerances in

**Table 3.** The effect of 3 weeks' adlay diet on levels of plasma glucose and insulin in fed rats<sup>1)2)</sup>

	Glucose (mg/100ml)	Insulin (μ/ml)
Normal	142.28 ± 36.61 <sup>b1)</sup>	9.53 ± 5.24 <sup>a</sup>
Diabetic		
- control	591.99 ± 286.55 <sup>a</sup>	6.40 ± 2.72 <sup>a</sup>
- Raw milled adlay	783.31 ± 148.60 <sup>a</sup>	6.10 ± 3.18 <sup>a</sup>
- Roasted milled	676.34 ± 218.35 <sup>a</sup>	6.81 ± 5.84 <sup>a</sup>
- Steamed milled	662.33 ± 219.67 <sup>a</sup>	4.06 ± 1.31 <sup>a</sup>
- Raw whole grain	653.53 ± 273.66 <sup>a</sup>	4.05 ± 3.08 <sup>a</sup>

1) Values are mean ± SEM, n=8

2) Within a given column, values with different superscripts are significantly different(p < .05)



**Fig. 2.** Effect of adlay diet on plasma glucose increment after glucose loading(1g/kg BW)(within a given column, values with different superscripts are significantly different(p < .05)).

raw milled adlay and raw whole grain adlay groups were not different from that of diabetic control group, the steamed adlay group showed lower blood glucose levels throughout the test period. The blood glucose level returned to the level of pre-glucose loading after 2 hrs in the roasted adlay group, al-

**Table 4.** The effect of 3 weeks' adlay diet on levels of glycogen and protein in liver and muscle in fed rats<sup>1)2)</sup>

	Glycogen( $\mu\text{g/g}$ )		Protein( $\text{mg/g}$ )	
	Liver	Muscle	Liver	Muscle
Normal	88834 $\pm$ 27157 <sup>a</sup>	2050 $\pm$ 1713 <sup>a</sup>	474 $\pm$ 251 <sup>a</sup>	357 $\pm$ 75 <sup>a</sup>
Diabetic – control	44333 $\pm$ 16538 <sup>b</sup>	834 $\pm$ 407 <sup>a</sup>	399 $\pm$ 119 <sup>a</sup>	350 $\pm$ 282 <sup>a</sup>
– Raw, milled adlay	31431 $\pm$ 16466 <sup>cd</sup>	1642 $\pm$ 613 <sup>a</sup>	390 $\pm$ 190 <sup>a</sup>	153 $\pm$ 38 <sup>b</sup>
– Roasted, milled adlay	34944 $\pm$ 14141 <sup>bcd</sup>	1511 $\pm$ 944 <sup>a</sup>	330 $\pm$ 251 <sup>a</sup>	208 $\pm$ 162 <sup>ab</sup>
– Steamed, milled adlay	48884 $\pm$ 17704 <sup>b</sup>	3064 $\pm$ 1172 <sup>a</sup>	285 $\pm$ 81 <sup>a</sup>	184 $\pm$ 55 <sup>b</sup>
– Raw, whole grain adlay	19053 $\pm$ 6702 <sup>d</sup>	1715 $\pm$ 1374 <sup>a</sup>	343 $\pm$ 96 <sup>a</sup>	238 $\pm$ 185 <sup>ab</sup>

1) Values are mean  $\pm$  SEM, n=8

2) Within a given column, values with different superscripts are significantly different ( $p < .05$ )

though the peak level of blood glucose was not different from that of the diabetic control group.

The effect of adlay diets on glycogen and protein levels in liver and muscles of fed rats is shown in Table 4. Compared to the normal group, the liver glycogen level of diabetic rats was significantly lower. Among diabetic rats, the liver glycogen level of the raw milled adlay group and the raw whole grain adlay group was lower than that of the diabetic control group. The liver glycogen levels of roasted milled adlay and steamed milled adlay were not significantly different from that of the diabetic control group. There was no significant difference in muscle glycogen among experimental groups due to a large standard deviation. The protein level of diabetic control rats was not different from that of normal rats in liver and muscle. However, regardless of different cooking methods, the muscle protein levels of adlay groups were lower than that of either diabetic control rats or normal control rats. However, there was no difference in liver protein among experimental groups.

## Discussion

At various points during the study, higher blood glucose levels, lower growth rates, and lower FER were shown in diabetic animals. Thus, these animals were considered to be severely diabetic throughout the study. Because all animals consumed diets with similar nutritional composition including fiber, the results of this study is from the effect of the adlay itself rather than the difference in nutritional composition between adlay and control diet. Compared to diabetic control rats, all adlay diet groups, showed some tendency of decreased plasma glucose until the

2nd week on experimental diets, but this tendency could not be found at the 3rd week. Thus, it is not possible to suggest that adlay diet might have some hypoglycemic effects.

Plasma insulin levels also tended to be lower in the steamed adlay group. This data suggests that there may be a slight difference between steamed adlay and raw milled adlay in their ability to influence insulin secretion or metabolism. However, the lower insulin concentration observed in the steamed adlay group, as compared with the raw milled adlay group, was not of sufficient magnitude to draw any conclusion. During 2 hrs after glucose loading, the steamed group showed a lower peak level and stayed at lower levels throughout the glucose tolerance test period. It is reported that the glycemic index of food depends on the difference in the proportion of starch present as amylose, with varieties that contain a higher proportion of amylose having a slower rate of digestion and producing lower glycemic response<sup>16</sup>. This amylolytic digestion is also considerably slower in food containing viscous fibers<sup>17)18</sup>. The viscosity of adlay starch was greater than that of other grains<sup>20</sup>. The water-soluble major polysaccharides from adlay were water soluble glucans, to which are attached glucosyl side chains in a similar way to amylopectin<sup>21</sup>. Because of these factor, the lowering of blood glucose seen in the steamed adlay group might be the result of different digestion rates due to viscosity. Thus, despite the many uncertainties regarding mode of action, these results suggest that steamed adlay could have potential for reducing postprandial hyperglycemia in clinical settings. The reason why the increment pattern of roasted adlay group after glucose loading was different from that of other groups is still in question and more work has to be done.

Insulin has been shown to reduce the rate of protein degradation resulting in net protein retention<sup>22)</sup> and a reduced rate of protein degradation could occur with a resulting decrease in protein oxidation. Thus, the insufficiency of insulin due to diabetes induced increase the protein degradation and release to the blood for the source of energy and gluconeogenesis. However, the protein level of plasma, liver and muscle of diabetic rats in this study was not different from those of normal rats. Furthermore, the protein level of muscle and liver in adlay group were even lower than that of control. Thus, 3 weeks period of diabetes might not be sufficient to draw the protein degradation.

In conclusion, the hypoglycemic effect of adlay diet is not significant when total fiber consumption is controlled. It has been reported that a heat-stable trypsin inhibitor was found in adlay that has activity stable against pepsin digestion<sup>23)24)</sup>. Therefore, adlay diets may lead to increased body protein degradation, one of the chronic complications of diabetes. Besides, adlay is not a palatable or economic grain. These results suggest that it is better to find other high fiber grains other than adlay for controlling hyperglycemia because taste and price are still the most influential factors in food selection, although nutrition is also important.

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