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# Anti-Diabetic Activities of Fucosterol from *Pelvetia siliquosa*

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Fucosterol isolated from *Pelvetia siliquosa* was tested for its anti-diabetic activity *in vivo*. Fucosterol, when administered orally at 30 mg/kg in streptozotocin-induced diabetic rats, was caused a significant decrease in serum glucose concentrations, and exhibited an inhibition of sorbitol accumulations in the lenses. Fucosterol, when administered orally at 300 mg/kg in epinephrine-induced diabetic rats, was also caused an inhibition of blood glucose level and glycogen degradation. These results demonstrated that fucosterol is a main anti-diabetic principle from the marine algae *P. siliquosa*.

**Key words:** *Pelvetia siliquosa*, Fucaceae, Fucosterol, Anti-diabetic activity

## INTRODUCTION

Genus *Pelvetia* is a typical marine algae, and only four species of which have been known to be distributed worldwide. Among them, *P. siliquosa* has been reported to be peculiar to the Korean peninsula and self-grown on the craggy surfaces near to the seashores of the southern area (Yoon, 1995). It has traditionally been used as seasoned sea greens for religious services or as health food (Oh *et al.*, 1990). In previous papers, we reported the hepatoprotective and anti-diabetic effects of the ether fraction (Lee *et al.*, 2002) and the anti-oxidant activity of fucosterol (Lee *et al.*, 2003) from this plant.

In course of the evaluation of biological activities and its bioactive principle from this plant, anti-diabetic activity of fucosterol isolated from this plant was investigated.

## MATERIALS AND METHODS

### Material and chemicals

Fucosterol (Fig. 1) isolated from *Pelvetia siliquosa* as described previously (Lee *et al.*, 2003) was used for the experiment on anti-diabetic activities. Nicotinamide adenine dinucleotide (NAD), sorbitol dehydrogenase, glycine, citric acid, glucose, streptozotocin (STZ) and epinephrine (EP) were purchased from Sigma Chem. Co. (St. Louis, MO).

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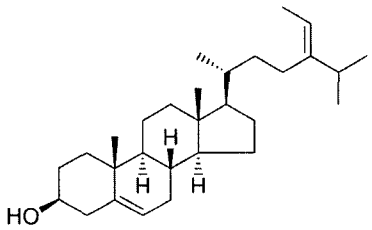


**Fig. 1.** Structure of fucosterol

All other chemicals and reagents were analytical grade and commercially available.

### Animals

All animal studies were carried out in the pathogen-free barrier zone at Seoul National University Hospital in accordance with the procedure outlined in the Guide for the Care and Use of Laboratory Animals. All animals were acclimatized for at least for one week, caged in groups of five or less and fed with a diet of animal chow and water *ad lib.* They were housed at 23±0.5°C at 10% humidity in a 12 h light-dark cycle.

### Preparation of STZ-induced diabetes and sample treatment *in vivo*

Diabetes mellitus was induced in male Sprague-Dawley rats by a single intravenous injection of STZ dissolved in phosphate buffered saline, acidified to pH 4.5 with 0.05 M citric acid (85 mg/kg). After the induction, fucosterol or epalrestat (ONO Co. Ltd.) were administered *via* an intragastric tube at a dose of 30 and 50 mg/kg, respectively,

at 4, 7 and 24 h. Three hours later after last administration, blood samples were collected, and lenses and sciatic nerves are removed. The plasma glucose levels were determined using a commercially available kit based on the glucose oxidase method (Trinder, 1969). The amount of glucosylated hemoglobin in the red blood cell (RBC) was measured using a Drabkins reagent kit 525 (Drakin and Ausin, 1932). The sorbitol contents in the rat lenses, sciatic nerves and RBC were determined enzymatically by a modification to the enzymatic assay of Clements *et al.* (1969).

### Preparation of EP-induced diabetes and sample treatment *in vivo*

Groups of 8 mice were administered i.p. with fucosterol dissolved in olive oil or vehicle alone. Four hours later, EP (0.6 mg/kg, i.p.) was administered and blood samples were withdrawn by decapitation 1 h post dosing for blood glucose determination. Serum glucose levels were measured using glucose oxidase method (Mariam *et al.*, 1996). Livers were removed for the measurement of glycogen content. To the liver, 30% KOH solution was added and the mixture was added to the mixture, which was then kept at 4°C overnight. The mixture was centrifuged at 3000×g for 15 min, fractionated twice with EtOH. The combined precipitate was dried in a desiccator, dissolved in water, and glucose content was measured using the anthrone-sulfuric acid method (Johnson and Fusaro, 1966).

### Statistical analysis

The statistical significance of the results was analyzed by Student's *t*-test for unpaired observations.

## RESULTS AND DISCUSSION

Anti-diabetic activity *in vivo* of fucosterol from *P. siliquosa* on was investigated. Effects of fucosterol on the glucose concentration as well as on the sorbitol contents in the lenses, RBC and sciatic nerves in STZ-induced diabetic rat were studied and the results were showed in Table I. In the diabetic control rats, the serum glucose levels were significantly elevated compared to those of the normal rats, and exhibited an inhibition of sorbitol accumulation in the lenses, but no inhibition in RBC and sciatic nerve.

Effect of fucosterol on EP-induced diabetic mice was tested and the results were indicated in Fig. 2. EP produced a highly significant hyperglycemic response by approximately 300% increase in the blood glucose level

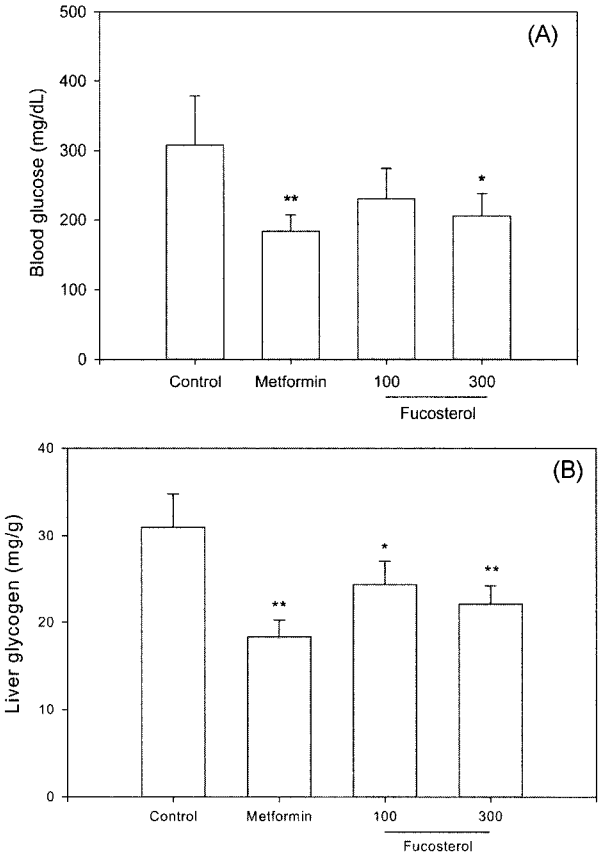


**Fig. 2.** Effect of fucosterol on the blood glucose level (A) and the glycogen content (B) of EP-induced hyperglycemic mice. Mice were administered i.p. with fucosterol (100 and 300 mg/kg) and metformin (100 mg/kg) dissolved in olive oil. Four hours later, EP (0.6 mg/kg, i.p.) was administered and one hour later, blood glucose level and glycogen content were determined. Each value represents the mean±S.E.M. (n = 8). Significantly different from the control: *p < 0.05, **p < 0.01.

**Table I.** Effect of fucosterol on the serum glucose concentration and the sorbitol accumulation of STZ-induced diabetic rats (n = 8)

| Treatments | Serum glucose (g/L) | Sorbitol accumulation | | |
|---|---|---|---|---|
| | | Lens (nmol/mg dry wt.) | RBC (nmol/g Hb) | Sciatic nerve (nmol/mg dry wt.) |
| Control | 474.20±15.15 | 1.83±0.14 | 278.34±23.62 | 0.98±0.08 |
| Epalrestat | 413.75±20.60* | 1.35±0.14* | 89.84±7.03** | 0.91±0.06 |
| Fucosterol | 404.00±21.24** | 1.42±0.08* | 242.98±13.29 | 1.03±0.05 |

Fucosterol or epalrestat were administered orally at a dose of 30 and 50 mg/kg/day, respectively.
Significantly different from the control: *p < 0.05, **p < 0.01.

compared with that of the normal control. Fucosterol at a dose of 100 and 300 mg/kg was shown to cause a reduction, respectively, 25 and 33% in hyperglycemic effect induced by EP. Lowered the liver glycogen level of the control mice compared to the normal animals, but some significant reversal of its levels was observed by treatment of fucosterol. Fucosterol at a dose of 100 and 300 mg/kg was shown to cause a decrease of 23 and 29%, respectively, in glycogen degradation of rat liver.

The EP-induced diabetic rat may be explained in terms of changes in the effects of catecholamines i.e., enhanced glycogenolytic effects of EP, which is principally responsible for the acute hyperglycemia produced by this hormone (Kreisman *et al.*, 2000). The present results demonstrated that EP caused a marked elevation of blood glucose level as well as a significant reduction in the liver glycogen levels in the control animals. We can postulate that a significant decrease in the blood glucose and a significant reversal of liver glycogen contents in fucosterol-treated group in the present study occurred due to the inhibition of glycogen break down in the liver. The inhibition of glyconeogenesis, the increase in the utilization of peripheral glucose or the direct inhibition of insulin release in the liver might be another reason for the hypoglycemic effects of fucosterol.

These results demonstrated that fucosterol is a main anti-diabetic principle from the marine algae *P. siliquosa*. Further research is required to study its mechanism at the molecular level.

## ACKNOWLEDGEMENT

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