

Antioxidant and Anti-Cataract Effects of *Chlorella* on Rats with Streptozotocin-Induced Diabetes

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Summary The antioxidant activities of *Chlorella* in vitro and in vivo were investigated. *Chlorella* showed a strong antioxidant effect compared to various vegetables in a 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging assay. To evaluate the antioxidant and anti-cataract effects in vivo, a 7.3% *Chlorella* powder was fed to rats with streptozotocin-induced diabetes for 11 wk. At the end of the experiment, *Chlorella* had decreased the blood glycated hemoglobin (hemoglobin A_{1c}) and serum cholesterol levels significantly, however, it had not affected the serum glucose concentration. The serum lipid peroxide value (TBARS value) in the rats fed *Chlorella* was lower than that of the control rats. In the liver and kidney, *Chlorella* also reduced chemiluminescent intensities. In addition, it delayed the development of lens opacities. The lens lipid peroxide content of the rats fed *Chlorella* was lower than that of the control rats, however the differences were not significant. These results indicate that *Chlorella* has antioxidant activity and may be beneficial for the prevention of diabetic complications such as cataracts.

Key Words *Chlorella*, antioxidant activities, DPPH, diabetes, cataract

Oxidative stress, induced by reactive oxygen species, is thought to be an important factor leading to chronic diseases in mammals. In diabetes mellitus, hyperglycemia causes the nonenzymatic glycosylation of proteins through the Maillard reaction, in which reactive oxygen species are produced and oxidative stress may arise (1–3). Enhanced oxidative stress has been proposed to cause harmful damage to cells and tissue proteins through cross-linking, fragmentation, and lipid peroxidation. Nonenzymatic glycation and lipid peroxidation have been estimated to play an important role in the progression of diabetic complications such as atherosclerosis, neuropathy, renal disease, retinopathy, and cataracts.

Dietary antioxidants such as ascorbic acid, α -tocopherol and carotenoids can act as inhibitors of free radicals and lipid peroxidation (4–6). The consumption of fruits and vegetables, which are rich in antioxidant components, has been reported to increase plasma antioxidant capacity in humans (7). Thus, the active intake of dietary antioxidants may decrease oxidative stress in diabetes mellitus and be effective for the prevention of diabetic complications.

Chlorella powder is used as a functional food in Japan and is reported to have antihypertensive and hypocholesterolemic effects in humans and animals (8–11). *Chlorella* powder contains many dietary antioxidants such as lutein (2500 ppm), α -carotene (500 ppm), β -carotene (500 ppm), ascorbic acid (250 ppm), and α -

tocopherol (250 ppm). However, the antioxidant activity of *Chlorella* has not been fully clarified and there is no published study describing the antioxidant effect of *Chlorella* in rats with streptozotocin-induced diabetes (STZ rats).

The aim of this study was to determine the antioxidant activities of *Chlorella* both in vitro and in vivo. As antioxidant activity in vitro, the scavenging of the 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical was measured. To determine the antioxidant effect of *Chlorella* on diabetes, we investigated the effect of *Chlorella* on the glycated hemoglobin (hemoglobin A_{1c}) and lipid peroxide (TBARS) levels in the serum of STZ rats. Miyazawa and Kaneda have shown that the chemiluminescence of rat tissue homogenates was related to lipid peroxidation accompanied by the formation of singlet oxygen and free radicals (12). Thus, we measured the chemiluminescent intensities of kidney and liver homogenates to discuss the antioxidant activities of *Chlorella*. In addition, we also investigated the effect of *Chlorella* on the prevention of cataracts, a diabetic complication, in STZ rats.

MATERIALS AND METHODS

Determination of antioxidant activity by DPPH radical scavenging assay

Materials. Viable *Chlorella* (*Chlorella regularis*) cells were obtained from Nihon Chlorella Co., Ltd. (Tokyo, Japan). Vegetables, such as green sweet pepper, spinach, cabbage, and celery, were purchased in a local market in Tokyo, Japan. 1,1-Diphenyl-2-picrylhydrazyl (DPPH)

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