

Study on the Active Ingredients and Mechanism of BioBran (MGN-3) in Inhibiting D-Galactosamine-Induced Liver Damage

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[Purpose] In rats, liver damage caused by the administration of D-galactosamine hydrochloride (D-GalN) shows a similar histopathological image to that of human viral hepatitis, and when the dose of D-GalN is increased, fulminant hepatitis-like pathology is known to be exhibited. In previous studies, BioBran (MGN-3) has shown an inhibitory effect against D-GalN-induced liver damage, and the effect tended to increase when it was hydrolyzed in dilute hydrochloric acid. It has been suggested that the active ingredient is absorbed from the intestine. A variety of cytokines are involved in the onset of liver damage. In our laboratory, we focused on the role of the cytokine IL-18 in the inhibition of D-GalN-induced liver damage. In this experiment, we investigated the active ingredient in MGN-3 and the involvement of IL-18 in the mechanism of MGN-3 in suppressing D-GalN-induced liver damage.

Chapter 1: [Method] In Experiment 1, four-week-old Wistar rats were injected intraperitoneally with a solution of D-GalN at 800 mg/kg body weight. One hour before the injections, a solution of MGN-3 hydrolyzed in 1NHCl at 100°C for 1 h, 2NHCl at 120°C for 1 h, or 2NHCl at 120°C for 3 h or unprocessed MGN-3 was injected intraperitoneally at 20 mg/kg body weight. Twenty-four hours after the D-GalN administration, serum transaminase (GOT and GPT) activity was measured. In experiment 2, MGN-3 was hydrolyzed in 1NHCl at 100°C for 1 h and passed through cation and anion exchange resins. Gel filtration was used to fractionate the molecules of low, medium, and high molecular weight. In the same way as in Experiment 1, 4-week-old Wistar rats were used, and before the GalN administration, the rats were given intraperitoneal injections. TOF-MS was used to analyze the composition of the low molecular weight fraction of MGN-3.

[Results] In Experiment 1, the activity of serum GOT and GPT, a marker of liver damage, showed significant inhibitory effects in the MGN-3 group and all three hydrolyzed MGN-3 groups compared to the control group. However, there was no significant difference among the three hydrolyzed MGN-3 groups. The active ingredient in MGN-3 showed an effect even after the hydrolysis treatment in 1NHCl at 100°C for 1 h. In Experiment 2, serum GOT and GPT activity showed significantly lower levels in the MGN-3 low molecular weight group. This suggests that the active ingredient in MGN-3 is in the low molecular weight fraction. The results of the TOF-MS suggest that the low molecular weight fraction includes monosaccharide and oligosaccharide.

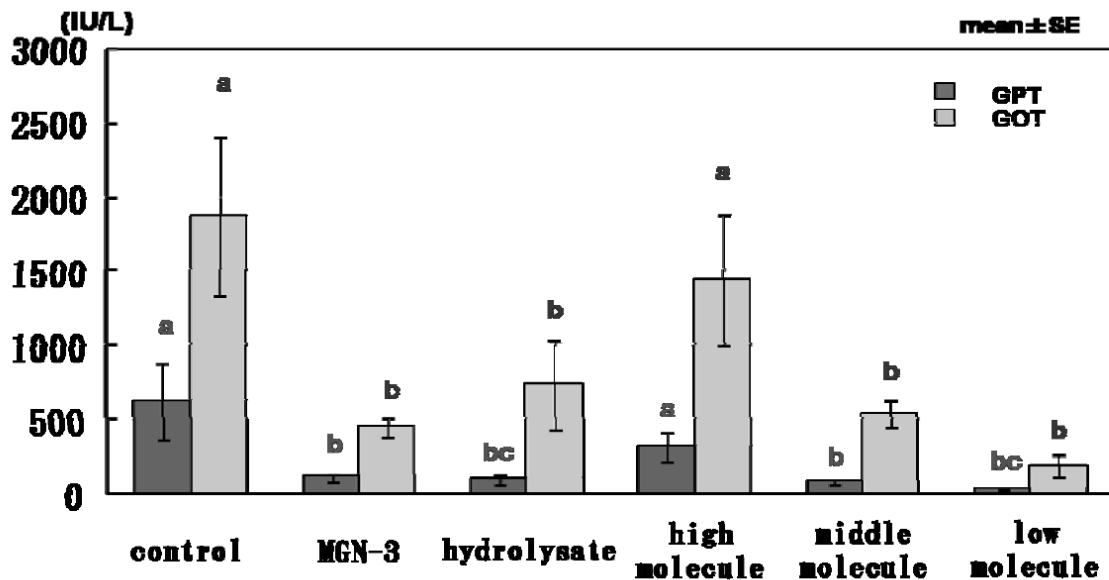
Chapter 2: [Method] To investigate the relation of IL-18 and caspase-1 to the mechanism of the D-GalN-induced liver damage, 4-week-old Wistar rats were injected with D-GalN solution in the same way as in the previous experiments. They were dissected 0, 2, 4, 6, and 8 hours later, and the expression level of cytokine IL-18 mRNA and caspase-1 activity in the liver was examined.

[Results] Four hours after the injection of D-GalN, the IL-18 mRNA expression level began to rise and reached its maximum after about 6 hours. Caspase began to rise after about 4 hours.

Chapter 3: [Method] We investigated the involvement of IL-18 in the inhibition of D-Ga1N-induced liver damage by the low molecular weight fraction of MGN-3. The experimental method was as previously explained, and the rats were dissected 8 and 24 hours after being injected with D-Ga1N solution. We examined the activity of the serum transaminases GOT and GPT and the expression level of cytokine IL-18 mRNA in the liver.

[Results] After 8 hours, the control group and the low molecular weight fraction group both had increased GOT and GPT activity. After 24 hours, the low molecular weight fraction group showed significant reduction compared with the control group. Eight hours after the D-GA1N injection, the levels of IL-18 mRNA expression in the low molecular weight fraction group had decreased significantly more than those of the control group. From these results, we infer that the reduction in IL-18 was partly responsible for the mechanism of the low molecular weight fraction of MGN-3 of suppressing D-Ga1N-induced liver damage.

Serum GOT and GPT activity



Differing letters indicate significant differences ($p < 0.05$)