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GINSENOSIDERB1 PREVENTING ANAPHYLACTIC DEATH IN MOUSE

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Abstract

Repeated immunization induces strong stress to kill mice. Ginsenosides have adaptogenic activities to cope with animal stresses. Mice treated with a ginsenoside may escape from the anaphylactic death. In the present study, effects of ginseng extract and ginsenosideRb1 on the death was examined. Mice were repeatedly given immunization stress. Fifty or 60 percent of them were dead at the third immunization. All mice treated with ginseng extract at the third immunization escaped from the anaphylactic death. GinsenosideRb1 dose-dependently prevented the anaphylactic death. These suggest that ginsenosideRb1 coped with the strong stress induced by repeated immunization. The present findings help to elucidate stress-coping mechanism of animals.

Key words : immunization, stress, anaphylactic death, ginsenosideRb1, adaptogenic activity

Introduction

To maintain adaptation, living organisms produce endogenous substances for coping with stresses. Some botanical substances, neither proteins nor peptides and known as adaptogens, reduce stress reaction in animals[1,2]. Ginsenosides, contained in Korean red ginseng roots, are well-known adaptogens. On the other hand, repeated immunization induces severe stress to kill mice. Mice given the stress will produce adaptogenic substances to escape from the death. In fact, it was reported that the surviving mice produced a humoral lipid preventing the anaphylactic death[3]. The author supposes that administration of a ginsenoside may prevent anaphylactic death from mice. In the present study, the author sets an immunization system to assess anaphylactic death in mice, examines adaptogenic effect of ginseng extract, and investigates the effect of ginsenosideRb1.

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Materials and Methods

1. Animal

Male 6 weeks-old DDY mice were purchased from SLC Co. (Hamamatsu, Japan) for use in the present study. This study was conditioned in accordance with animal research regulations at Akita University School of Medicine. All experiments were performed under permission of Animal Experiment Ethics Committee (a-1-2824).

2. Assessment of anaphylactic death

Mice were intraperitoneally injected with 1 mg/kg of ovalbumin (OVA, Grade V, Sigma-Aldrich Co., St. Louis, MO, USA) dissolved in physiological saline (PS). Ten days after this, they were given second immunization with the same OVA concentration. Seven days after the second immunization, they were injected with OVA solution 1 mg/kg and samples, or PS as a control. Anaphy-

lactic death of the mice was assessed at 1 h after the last immunization.

3. Statistical analysis

Fisher's exact test was used. A $p<0.05$ was considered a significant difference.

4. Effect of ginseng extract

① Ginseng extract

Ginseng extract was purchased from Tsumura Co. Ltd. (Tokyo, Japan), and was prepared to 100 mg/ml with water.

② Effect preventing anaphylactic death

Forty mice were twice immunized as described above. Seven days after the second immunization, 10 mice were intraperitoneally injected with OVA solution 1 mg/kg and 100, 20 or 4 μl of the ginseng extract solution, or 100 μl PS as a control. At one h after the third immunization, anaphylactic death was assessed.

5. Effect of ginsenosideRb1

① GinsenosideRb1

GinsenosideRb1 was purchased from Cayman Chemical Co. Ltd. (Ann Arbor, MI, USA), and was prepared to 1 mg/ml with water.

② Effect preventing anaphylactic death

Forty mice were twice immunized. Seven days after the second immunization, 10 of them were treated with OVA solution 1 mg/kg and 4, 1 or 0.2 mg/kg of the ginsenosideRb1 solution, or 100 μl PS as a control. At one h after the treatment, anaphylactic death was assessed.

Results

1. Effect of ginseng extract

All mice treated with 100 μl of the ginseng extract escaped from the anaphylactic death. With 20 and 4 μl treatment, 2 and 4 mice did not. Five of 10 mice treated with PS, as the negative control, died (Table 1).

2. Effect of ginsenosideRb1

All mice treated with ginsenosideRb1 4 mg/kg escaped from the anaphylactic death. With 1 and 0.2 mg/kg, 4 and 6 mice did not. Six of 10 mice treated with PS died (Table 2).

Table 1. Effect of ginseng extract (100 mg/ml) on anaphylactic death

Dead (/Total 10)			
Ginseng extract (μl)			The control
100	20	4	PS 100 μl
0*	2	4	5

*$P<0.05$ compared with the control (Fisher's exact test).
PS : physiological saline

Table 2. Effect of ginsenosideRb1 on anaphylactic death

Dead (/Total 10)			
GinsenosideRb1 (mg/kg)			The control
4	1	0.2	PS 100 μl
0*	4	6	6

*P<0.05 compared with the control (Fisher's exact test).
PS : physiological saline

Discussion

These findings indicated that mice subjected to immunization stress had lower anaphylactic death rates when treated with ginseng extract. Furthermore, ginsenosideRb1, an adaptogen contained in the extract, had a role to prevent the anaphylactic death.

It is reported that ginsenosidesRg3, Rf and Rh2 inhibit passive cutaneous anaphylactic reaction in mice[4]. On the other hand, ginsenosideRb1 protects hippocampal neurons from the ischemic damage[5], and improves spatial learning and memory in hippocampal sub-regions in rats[6]. These suggest that anaphylactic death might closely relate to hippocampal dysfunction in mice. Mice escaping from the anaphylactic death would produce internal adaptogenic substance maintaining the hippocampal function. However, mechanism producing the substance is not known and, similarly, affecting mechanism of ginsenosideRb1 is not yet well understood.

Overall, these findings suggest that animals would have an adaptation system, strengthened by ginsenosideRb1, coping with severe stress inducing the death.

Competing Interests

The author declares that he has no competing interests.

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