# Experimental Studies on the Preventive Effect of Garlic Extract against Infection with Influenza Virus \*

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\*Below is a quick translation via Google Translator. Full Study PDF In Original Japanese, including Table Data I, II, III, IV, V & VI is available here: <a href="https://www.jstage.jst.go.jp/article/kansenshogakuzasshi1970/47/9/47\_9\_321/\_pdf/-char/en">https://www.jstage.jst.go.jp/article/kansenshogakuzasshi1970/47/9/47\_9\_321/\_pdf/-char/en</a>

For many viral diseases, effective prophylaxis has not been discovered, so vaccine prevention is important; however, influenza vaccines are still present. Because influenza virus is susceptible to antigenic variation and the antigenicity of its strains is constantly changing, vaccines have been discussed. It takes time to select the strain to be used for clotting, it is difficult to prevent the outflow at an early stage, and the secretor of influenza local immunity is the secretory IgA antibody in the airway mucosa. It is not a circulating antibody in serum.1) Local administration of vaccine is considered to be effective for efficiently inducing the production of secretory IgA antibodies in the respiratory tract mucosa. Therefore, it is not commonly used because it has low strain specificity and is easy to administer. Therefore, garlic has been used empirically in the East for a long time and its effect has been recognized, but for the purpose of verifying the effectiveness of garlic. I report because I tried an experiment.

#### **Experimental Materials and Methods**

1. Male and female mice of ddYS strain and weighing 129  $\pm$  19 or 209 were used.

2. Vaccine and attacking virus influenza vaccine (Hitaken) Mice) were inoculated subcutaneously with 0.1 ml of the mouse only once.

3.Garlic extract and administration method Vitamin B1, B12 and liver excretion were extracted to a final concentration of 10m9 / ml, 1 $\gamma$  / ml, and garlic extracted with lowconcentration ethanol. The extract added to 50 mg / ml was used as a garlic extract. The mice were given 0.2 ml or 0.002 ml daily via mouth 15 days before vaccination.

4.Measurement of virus infection inhibitory factor in serum After vaccination, on days 3, 7, 10, and 14 days, 10 mice from each experimental group were used. Blood was collected and the serum was pooled. The experimental procedures such as virus volume and serum were performed according to a standard method. In the case of sozavirus, the mice were dropped intranasally into 209 mice, and in the case of Japanese encephalitis virus, they were dropped intranasally as in 129 mice, followed by observation for 3 weeks.

5.Measurement method for neutralized antibody The same method as in (4) was used to fractionate globulin by adding an equal amount of ammonium sulphate sulphate solution to the test serum. Measurements were made in the same way.

### Experimental results

# **1. Influence on the prevention of infection of influenza vaccine and garlic extract**

The experimental mice were set up as shown in Table 1. The infectious titer of the same amount of virus was measured on the mouse of each experimental group, and the logarithm of the prevention was calculated. The logarithm of the mouse experimental group in which only the vaccine was inoculated was calculated as The difference was 1.0 for females and 1.2 for males, and no significant difference was observed between the experimental group in which 0.2 ml of garlic extract was administered daily and vaccinated. The protective logarithm of mouse was 2.3 in females and 2.4 in males, which was about 20 times higher than that of mice in the single-inbred group of vaccines, indicating an infection control effect. 0.002 ml of extracted liquid per day. The mice in the experimental group that received the vaccine and received the vaccine exhibited the same protective logarithm as the mice in the group that received the vaccine alone, while the mice in the experimental group that received only the garlic extract were the same. However, in the group to which 0.2 ml was administered one by one, the logarithm of protection was 2.1 in females and 2.0 in males, indicating the same degree of protective effect as when vaccine was used in combination. The protective log of mice with the 0.002 ml extract alone was 0.6 in females and 0.5 in males, lower than the protective effect of mice inoculated with vaccine alone.

### 2. Japanese encephalitis Vaccine and garlic extract

The effect of the solution on the protection against infection was set up in the same experimental group as in the influenza experiment, and the logarithm of the infection was calculated.As shown in Table II, the logarithm of the mouse in the group inoculated with vaccine alone was No difference between male and female was observed at 1.0, and garlic extract was used together However, there was no difference from the case of vaccine alone, and the log of infection protection was 0.6-0.7 in the mouse treated with garlic extract alone, and the protective effect was greater than that in the case of influenza. Is low.

3. Influenza vaccine and garlic extract: the elimination of viral infection inhibitory factors in serum

[See Tables I and II in Japanese PDF (untranslated) https://www.jstage.jst.go.jp/article/kansenshogakuzasshi1970/47/9/47\_ 9\_321/\_pdf/-char/en ]

When vaccine was inoculated into garlic extract-administered mice, the protective effect was greater in the case of influenza than in the case of mice vaccinated alone, whereas in the case of Japanese encephalitis, Inoculation of vaccinated mice with vaccinated extract did not show a difference in the protective effect against vaccinated mice compared to mice vaccinated alone. When vaccines were inoculated into mice treated with 0.2 ml or 0.002 ml, daily serum was collected from mice inoculated with vaccine alone, and the activity of whole blood or globulin fractions to prevent infection. As shown in Table III, the virus infection inhibitory activity that appeared in the sera of mice vaccinated alone with vaccine increased four-fold on day 7 after vaccine inoculation, as shown in Table III. 14 days later, a 50-fold increase in activity was observed. ml administration to mouse. When the vaccine was administered, a high infection-inhibitory activity was detected after 3 days, but the globulin fraction was extracted from the serum of the mouse in each experimental group and the infection-inhibitory activity was measured. Then, as shown in Table IV, only activity comparable to that of vaccine-inoculated mice was observed, and in mice inoculated with vaccine alone, whole serum and globulin were observed. Significant between the infectioninhibiting activities of the fractions.

[See Tables III, IV, and V in Japanese PDF (untranslated) - <u>https://www.jstage.jst.go.jp/article/kansenshogakuzasshi1970/47/9/47 9 321/</u> \_pdf/-char/en ]

Based on the above results, administration of garlic extract to mice did not enhance the production of neutralizing antibodies by vaccine stimulation, and showed that globulin fractionation did not increase. In the case of Japanese encephalitis, vaccine is applied to the mouse to which garlic extract was administered, as shown in Tables V and VI. No significant difference was observed between the whole sera and the globulin fraction in the case of inoculation and vaccination alone.

By administering an appropriate amount of the extracted garlic extract to mice, the protective effect against influenza virus can be enhanced. It is not due to the enhancement of antibody production against the virus, but to the appearance of an infection-inhibiting factor in serum, and the nature and biological specificity of this infection-inhibiting factor is I have to repeat my research. Substances that appear in the serum, such as circulating antibodies, but are difficult to translocate to the respiratory tract mucosa, where infection with influenza virus occurs, cannot exert infectionprotective ability3). It is considered that the infection inhibitor can migrate to the mucosal surface and, unlike interferon, the infection inhibitor acts directly on influenza virus particles to transmit the infection. However, Japanese encephalitis virus is insensitive to this infection inhibitor. As for specificity, we are currently conducting experiments, but show an inhibitory effect on A2 / B viruses as well as A0 / PR8 strains. Because the antigenicity of epidemic strains is constantly changing, it is difficult to prevent such viruses by vaccines. It may be effective in stopping the epidemic of flu.

## Conclusion

<u>Oral administration of garlic extract to mice was effective in</u> <u>protecting against influenza virus</u>, but was not observed in Japanese encephalitis virus. Infection-inhibiting activity was observed in the serum of the mouse to which the effluent was administered; it was present in places other than the globulin fraction in the serum; and garlic extract enhanced the production of neutralizing antibodies At the end, I would like to thank Dr. Kumagai and Mr. Kunio Ito, who assisted in the experiment.

### SUMMARY

Preventive effect of garlic extract added with vitamine B1, B12, and liver extracts against influenza infection in mice (ddYS) per nasally inoculated with influenza virus AO/PR 8 strain was studied. <u>The</u> <u>experiment revealed that the garlic extract</u>, contrary to Japanese encephalitis case, <u>further hightened the preventive effect of</u> <u>influenza vaccines and in the case of using alone it was as effective</u> <u>as vaccines</u>.

In order to analyze that augmented preventive activity against influenza infection, the activity of globulin fraction extracted from the mice was measured. But, no increase has been seen in the activity in that fraction. Then, it is presumed that it is not neutralization antibody but other preventive factor or factors that was prompted to increase.