

## **Abstract**

The most important adjuvant used in human vaccines is alum adjuvant. Alum stimulates humoral immunity strongly. But alum is not suitable for stimulating cellular immunity. The immune system adopts different strategies to deal with various pathogens, both intracellular and extracellular, and strengthens one of the arms of the immune system, cellular and/or humoral immunity. By knowing and using different types of adjuvants, the immune system can be pushed in a certain direction. Propranolol is one of the inhibitors of beta-adrenergic receptors, which is used as an adjuvant in vaccination. These receptors exist on a wide range of immune system cells. This substance alone and in combination with other substances can show different adjuvant properties. Liposomes are lipid compounds that have received much attention in pharmaceuticals and vaccines in recent years. This combination can preserve the pathogenic agent or antigenic fragments at the injection site and plays a significant role on immune cells to stimulate cellular immunity. In this study, *Salmonella typhimurium* was selected as an example of pathogenic bacteria inside the cell and the effect of the mentioned adjuvants was investigated to check the level of stimulation of the immune system. Heat-killed *Salmonella typhimurium* with alum adjuvant, liposome and propranolol were injected into BALB/C mice. After completing the vaccination course, to check the effectiveness of each adjuvant, the spleen and serum of mice were isolated to check the vital capacity of spleen lymphocytes, the amount of IL-4, IFN- $\gamma$ , and the ratio of IgG2a to IgG1. The increase in delayed sensitivity and the rate of survival and checking the bacterial load in the spleen were also investigated in each group. The highest survival rate in the challenge with live bacteria is related to the combined group of adjuvants of alum, propranol and glycyrrhizic acid liposome. The next teahouse was related to the group receiving alum and liposome. The simultaneous combination of these three adjuvants induces the production of a higher level of IgG2a/IgG1. Also, the amount of IFN- $\gamma$  production in this group was significantly higher than other groups. The group receiving propranol was in the second place and the lowest level was related to the control group. In order to promote Th2 responses, the Ngroup receiving alum along with killed bacteria had the highest induction of IL-4 production. After that, the group receiving liposome and alum and the group receiving liposome and propranol had the highest amount. The proliferation of splenic lymphocytes in the group receiving all three adjuvants was significantly higher than the other groups, and after that, the group receiving propranol had the highest induction of splenic lymphocyte proliferation. The stimulating effects of alum, propranol and liposome glycyrrhizic acid are much wider than the adjuvant effects of each of them alone or compared to the combined groups of two adjuvants. Alum alone promotes humoral immune responses, but does not support cellular immune responses. Glycyrrhizic acid liposomes alone are not efficient in inducing cellular immune responses, but together with adjuvant alum and propranol, they induce high protective power and strong immune responses against intracellular bacteria.

## **Keywords:**

alum, propranol, salmonella typhimurium, glycyrrhizic acid liposome