



## Original Article

Propolis Extract vs. Alum as Adjuvants for *E. Coli* Antigens in RatsMaysaa Abdulridha Jumaah<sup>1</sup>, Ali Abbood Issa Al-iedani<sup>1</sup><sup>1</sup>Department of Microbiology, College of Veterinary Medicine, University of Basrah, Basrah, Iraq.Received 2 August 2025, revised 22 October 2025;  
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**ABSTRACT**

Vaccines are vital tools in the fight against infectious illnesses. The choice of an appropriate adjuvant is one of the most crucial steps in vaccine development. Although aluminum salts are still the most widely used adjuvant, numerous novel compounds with improved safety and adjuvant properties have been the focus of the current study. The focus of this research is the alcoholic extract of local propolis as a natural vaccine adjuvant used with two types of Gram-negative antigens in rats, one with crude and another with killed bacterial antigens, compared with alum hydroxide adjuvants with the same antigens. The study's findings indicate that there was an increase in the total leukocyte count in all groups compared with the control group (all results are statistically significant ( $P < 0.05$ )), whereas, the result of G5 which consist of propolis adjuvant vaccine with the whole killed bacteria (PRKE) was not significant, and a notable increase in phagocytosis rates and respiratory burst assessment in the two types of adjuvants compared with the control group ( $P < 0.0001$ ). Concerning adaptive immune responses propolis, as a natural adjuvant, was able to stimulate humoral immune responses. However, the alum adjuvant was more potent in this respect, and the difference with the control was statistically significant ( $p < 0.001$ ). In conclusion, using the propolis vaccine adjuvant was efficient in the induction of responses against Gram-negative antigens.

<https://creativecommons.org/licenses/by-nd/4.0/>**Keywords:** Adjuvant, Propolis, Aluminum Salts, Interferon Gamma.**1 Introduction**

The most successful medical intervention to date in lowering the mortality and morbidity rates from infectious illnesses has been vaccination [1]. It saves millions of lives yearly [2]. Around the world, vaccines have been used to avoid much more fatal diseases in both humans and animals [3]. Worldwide vaccination initiatives have eliminated smallpox and nearly eliminated polio [4]. Due to their aging immune systems, older people may respond less well to vaccinations; nonetheless, adjuvant formulations or other methods of boosting immune responses may be necessary [5].

Adjuvants are substances in high molecular weight bound to an antigen with low molecular weight (Hapten Ag) that boost the immune system's response to an antigen. In the vaccination process, combining adjuvants with Ag increases the immune response and boosts their effectiveness. The Latin adjuvant, which means "to help, to assist," is where the word "adjuvant" originates [6].

Over 100 adjuvant preparations have been documented; however, a large number of these adjuvants are rarely employed because of their costly, difficult-to-prepare, or harmful effects [7].

Adjuvants for vaccinations are still mostly made of aluminum salts [8] or Freund's adjuvant oil emulsions, which are designed specifically for use with veterinary vaccines [9]. Despite being developed about a century ago, aluminum-based adjuvants remain the most widely utilized since they are included in at least 146 licensed vaccines for the prevention of illness [10]. However, they can also result in unfavorable side effects, including minimally induced cell-mediated immunity, long-lasting inflammatory reactions of the injection site's local tissues, and immunity that can trigger unwanted, uninvited immunoglobulin E (IgE)-mediated responses [10]. For these reasons, the scientific community should concentrate on developing a variety of adjuvants, as there isn't a single adjuvant that works for all situations, as this could cause problems with stability, toxicity, or production costs [11].

Therefore, creating novel immunological adjuvants is very crucial. Natural medicine is now the main focus of medication development, and the study of natural goods has become increasingly popular. Because propolis can generate an extended

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