AlumVax Phosphate 2%

Vaccine Adjuvant

Product information.

AlumVax Phosphate is wet gel (colloidal) of aluminum phosphate 2%, provided as a ready-to-use suspension. It is sterilized and aseptically filled. AlumVax phosphate is an amorphous aluminum hydroxyphosphate which is negatively charged at physiological pH (pI=5–7), suitable for adsorption of positively charged or neutral, alkaline proteins.
AlumVax Phosphate is available in two quantities:
#AP0050: 50 mL and #AP0250: 250 mL.

Storage and stability.

Shipping and storage: Room Temperature. Product is stable for 6 months. DO NOT FREEZE (ice crystal may impair the product).

Chemical properties.

Formulation: $AlPO_4$, Aluminium phosphate gel
Appearance: White gelatinous precipitate

Description.

Aluminum salts are the most common adjuvant used in approved prophylactic vaccines because of their excellent safety profile and ability to enhance protective humoral immune response. Since more than 80 years, it has been observed that aluminium compounds act by a depot effect and also by direct activation of the immune cells. Adsorption or entrapment of antigens in aggregates through hydrophobic and electrostatic interactions favors a high local antigen concentration and improved uptake by antigen presenting cells (APC).
Alum Phosphate stimulates Th2 response through the release of Th2-associated cytokines (IL4, IL-5, IL-13…) and Th2-associated antibodies (IgG1 & IgE). It increases Ag-specific $CD4^+$ T Cell proliferation and promotes NALP3 inflammasome activation and caspase 1-mediated release of IL-1 and IL-18.
NOTE: Alum is frequently used as an alternative to Freund's adjuvants, as it is less hazardous and less likely to cause tissue necrosis at the injection site.

Results.

Results below present the effect of Aluminum phosphate adjuvant on immune system response:

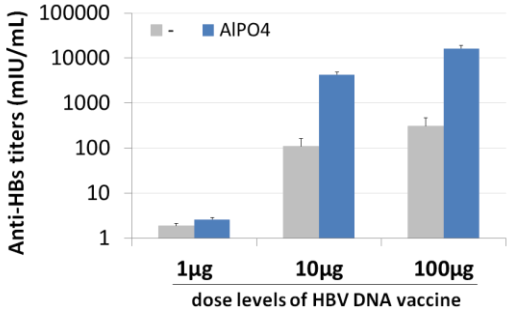

Figure 1. Adjuvant effect of AlPO4 for HBV DNA vaccines. Aluminum phosphate had a powerful adjuvant effect for Hepatis B (HBV) DNA vaccines in mice (adapted from Wang S. *et al.*, Vaccines. 2000; 18:1227-35).

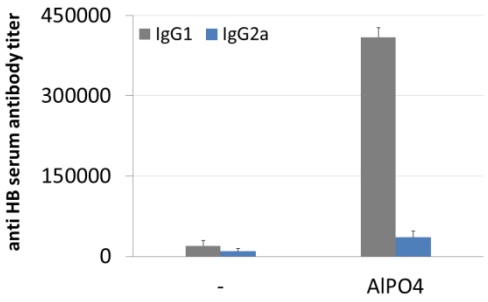

Figure 2. Efficient delivery of protein antigen. BALB/C mice were vaccinated i.m. with recombinant Hepatitis B surface antigen with or without AlPO4. 4 weeks sera were tested for anti-HBsAg antibodies by ELISA (adapted from Kwissa M. *et al.*, J Mol Med. 2003; 81:502-510.

Method/protocol.

Recommendations before starting:
The inoculum should be free of extraneous microbial contamination; filtration of the antigen before mixing with the adjuvant is recommended.
Preparation of Aluminum Phosphate-immunogens mix is easier than other adjuvants as it does not require laborious emulsification.

1. Ensure a complete re-suspension of AlumVax Phosphate adjuvant by vigorous shaking of the vial before use.
2. Dilute antigen mixture in saline buffer or phosphate buffer for a final immunogen concentration of 10-100 µg/100 µL.
3. Mix AlumVax phosphate adjuvant with an equal volume of antigen solution for a 1:1 ratio:
   a. Add AlumVax phosphate dropwise with constant mixing to the immunogen solution.
   b. Pipet up and down several times to ensure correct absorption of antigen by alum adjuvant and incubate 5 to 10 minutes.

NOTE: Ratio can be optimized from 1:1 (100µL adjuvant per 100µL antigen) to 1:9 (100µL adjuvant per 900µL antigen)

4. Inject into the animal according to the table below; the volume depends on the site of injection. Typical routes of administration include subcutaneous (SC), intramuscular (IM), intradermal (ID) or intraperitoneal (IP).

Species	Max vol/site	Primary injection	Subsequent Injection(s)
Mice, hamsters	0.1 mL	SC	SC
Mice, hamsters	0.05 mL	IM$^{\Delta}$	IM$^{\Delta}$
Mice	0.5 mL	IP$^{\times}$	SC, IM$^{\Delta}$
Guinea pigs, rats	0.2 mL	SC, IM$^{\Delta}$	SC, IM$^{\Delta}$
Rabbits	0.25 mL	SC, IM	SC, IM
Rabbits	0.025 mL	ID	SC, IM
Sheep, goats, donkeys, pigs Chickens	0.5 mL	SC, IM	

$^{\Delta}$ Not recommended in general, in particular not for viscous adjuvants
$^{\times}$ Not recommended for pAb production

Table 1: Maximum volumes for injection of immunogen/adjuvant mixtures per site of injection for different animal species (Adapted from Leenars MPPA, Hendriksen CFM *et al.*, 1999)

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