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ALUMINIUM-ADJUVANTED VACCINES – A REVIEW OF THE CURRENT STATE OF KNOWLEDGE

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ABSTRACT

Since decades aluminium formulations such as aluminium hydroxide and aluminium phosphate are widely used as adjuvants in vaccines for human use. They increase immune response induced by the vaccine antigens by mechanisms eg. a depot effect at the injection site, activation of the complement and stimulation of the macrophages.

Many studies, both case control ones and those performed *in vivo* on animal models, confirmed the safety of aluminium adjuvants even in vaccinated infants and children.

Although some of the aluminium-adjuvanted vaccines have certain limitations such as no Th1 reactivity and low stability at temperatures below 2ºC, its easy use, safety profile and low manufacturing costs confirm its suitability.

Key words*: vaccines, aluminium adjuvant, safety*

INTRODUCTION

Adjuvants are commonly used agents to augment the immune response induced with viral or bacterial inactivated vaccine antigens, bacterial toxoids or polysaccharides but not attenuated live viral ones.

Many studies have shown that adjuvant-containing vaccines are capable to efficiently increase and prolong the maintenance of antibody response comparing to the unadjuvanted equivalents (1). Adjuvants were proven to induce a repository or depot effect at the site of injection with slow releasing of the antigen which allow for targeting the antigen to antigen-presenting cells (APC), stabilize epitope conformation, stimulate the macrophages to induce retention and activation of lymphocytes and activate the complement (1, 2, 3). Their stimulatory properties are very practical as they allow to reduce the amount of antigen per human dose and the number of required doses in the vaccination schedule as well (4).

Recently, the concerns about safety of aluminium-adjuvanted vaccines have been frequently raised by media. It seems that aluminium attention have taken the place of thiomersal fear lowered lately by the competent international authorities statements and progressive elimination of thiomersal from most of the vaccines currently being in use.

Aluminium adjuvants were the first excipients that have been approved in the content of vaccines used in humans (5). By many decades they have successfully been used to enhance immune response to many vaccine antigens in order to improve the efficiency of vaccination. Aluminium hydroxide or aluminium phosphate have been the most common class of vaccine adjuvants, recognized as safe when used according to the recommended vaccination schedules (2). Nevertheless, its proven adjuvancity mechanisms are still not entirely understood (6). They are recognized as Th2 type response inducers, however with low potential to induce cell-mediated immunity or immunity to peptide antigens (3).

Firstly, aluminium adjuvants were used in the formulas of diphtheria, tetanus and pertussis vaccines and inactivated poliomyelitis vaccines and over time they have been introduced into newly developed vaccines such as hepatitis A and B and inactivated thick-borne encephalitis vaccines (2). Nowadays, most of the adjuvanted vaccines are adsorbed on aluminium hydroxide

and only some eg. meningococcal and pneumococcal conjugate ones are adsorbed on aluminium phosphate. The combination of these both is used rarely.

PHYSICAL AND CHEMICAL PROPERTIES OF ALUMINIUM ADJUVANTS

Aluminium adjuvants are often referred as "alum"-containing products, but this term should be rather avoided, as it refers to specific chemical compound, hydrated aluminium sulfate, which is not under scope of vaccine aluminium-containing adjuvant (7). Aluminium-containing vaccines are formulated by adsorption of a given antigen onto aluminium hydroxide or aluminium phosphate gels (8). Nevertheless, commonly used names of aluminium hydroxide or aluminium phosphate, do not exactly describe their structures. Aluminium hydroxide, as identified using X-rays crystallography, is a crystalline aluminium oxyhydroxide (AlO(OH)), and aluminium phosphate is an amorphous aluminium hydroxyphosphate $Al(OH)_x(PO_4)_y$ (9). They are prepared by exposing aqueous solution of aluminium ions under alkaline conditions in a well-defined and monitored chemical environment (2). An avidity of the association between adjuvant and antigen is affected by many factors, such as the form of aluminium salt, the physico-chemical properties of the antigen (including molecular weight), the mode of preparation of the antigen-adjuvant complex and pH of the chemical environment (10). The main difference between aluminium oxyhydroxide and aluminium hydroxyphosphate reffers to theirs point of zero charge (PZC) which is estimated at pH 11.0 and pH 4.0 – 5.5, respectively. PZC represents a pH value at which electrical charge density on a surface of a solid submerged in an electrolyte obtains value of zero. This feature decides on choice of the best adjuvant for a given vaccine antigen according the charge of the last one (6). Generally, an efficient adsorption of antigen depends on the pH value obtained between the isoelectric point (IEP) of the antigen and the PZC of the adjuvant, due to guarantee the opposite electrical charges and optimal levels of electrostatic attraction and adsorption (2). Thus, aluminium hydroxide at pH of 11.0 is preferable for adsorption of antigens with an acidic IEP and aluminium phosphate at pH 4.0 - 5.5 for antigens with alkaline IEP (11).

Selection of an appropriate adjuvant is important for the expected level of immunogenicity and finally for the effectiveness of the vaccine. In case of DNA vaccines, the use of aluminium hydroxide as an adjuvant resulted in decreased immunogenicity, while application of the aluminium phosphate instead, effectively enhanced the immune response (12, 13).

Despite aluminum-containing adjuvants are used on so frequently, they reveal some limitations. Traditional aluminium-adsorbed vaccines are frost sensitive and thus not lyophilized (2). Exposure of the adjuvanted vaccines to freezing temperatures causes irreversible breakage of the lattice made up of bonds between the adsorbent and antigen, resulting in compromised immunogenicity and increasing of the risk of adverse local reactions (14).

SAFETY OF ALUMINIUM - ADJUVANTED VACCINES

Aluminium-containing adjuvants were proved for no evidence of risk of carcinogenicity or teratogenicity (15). As very high doses of aluminium can be toxic, safe aluminium compounds concentrations limits were clearly defined as 2 mg/kg per day. It should be emphasized, that exposure to aluminium content in vaccines is substantially lower than exposure originating from a diet (16), despite the fact that aluminium compounds in vaccines do not pass through the gastrointestinal tract, which is a significant barrier (17).

In Europe, the maximum acceptable amount of aluminium in vaccines administered to humans, in accordance with the requirements of the actual edition of the European Pharmacopoeia is 1.25 milligrams per human dose.

Aluminium compounds in some circumstances may however cause an allergic response. The most commonly observed adverse reactions related to aluminium-adjuvanted vaccines include painful and itchy nodules and redness at the injection site, however they are usually mild and short-lived (15).

Meta-analysis study on adverse events reported after immunization with aluminium-containing DTP vaccines administered to children, showed no evidence that aluminium salts in vaccine contents cause any serious and long-lasting adverse events (18). Up to date, only few cases of hypersensitivity reactions to aluminium such as dermatitis, either localized or systemic were described (19).

Recently, there have been data published indicating a possible link between exposure to aluminium and development of an Alzheimer's disease, however, this association still remains unproven as the estimated amount of aluminium absorbed by the body from the food is much higher than from vaccination (15).

Global Advisory Committee on Vaccine Safety (GACVS), which is a scientific advisory body of the World Health Organization in their report issued in June 2012 stated, that there are no scientific evidence of any harm related to aluminum-adjuvanted vaccines and similarly no link with autism (20 - 22). Moreover, GACVS pointed out that many incorrect assumptions on suspected associations of aluminium with neurological disease development coupled with the lack of reliable

data in ecological studies, as correlation of vaccine aluminium exposure and its outcomes on population averages, were not found or recognized as valid. Despite that, GACVS advised to continue clinical trials and epidemiological studies on monitoring and tracing evidence of aluminium safety (22).

Safety of HPV vaccination with aluminium-adjuvanted vaccine was also confirmed by GACVS statement released on 12 March 2014. After reviewing evidence on cases of macrophagic myofasciitis (MMF) – a rare muscle disease, characterized by microscopic lesions contained aluminium salts, primarily related to immunization with aluminium-adjuvanted vaccines, GACVS did not find any scientific evidence on relations of aluminium present in HPV vaccine and skin reactions occurring at the injection site (MMF) with any autoimmune syndrome (23). European Medicines Agency reviewed of registered HPV vaccines to further clarify aspects of their safety profile due to probability of occurrence of cause-effect link between administration of the vaccine against HPV and the presence of rare pain syndromes and dysfunction of the autonomic nervous system (24). The review concluded that based on evidence, there is no casual link between HPV vaccines and development of analysed syndromes ie. complex regional pain syndrome (CRPS) and postural orthostatic tachycardia syndrome (POTS) (25).

Several studies on aluminium pharmacokinetics have also been performed. Study on *in vivo* absorption of aluminium-containing vaccine adjuvants has been performed on rabbits using the ^{26}Al isotope as a tracer. Concentrations of aluminium in blood and urine of the animals were measured during the entire experiment. Based on the results, it was estimated that administration of a dose contained 0,85 mg of aluminium to adults, results in increasing of its concentration in plasma by approx. 0,04 ng/ml (about 0,8%) (26). According to the above presented data, the hypothesis that the amount of aluminium administered to the body via vaccination contributes significantly within the general exposure of humans to aluminium seems rather unlikely (2).

Safety of vaccines used according to the immunization program was confirmed by pharmacokinetic studies conducted by the US Food and Drug Administration (FDA), where the estimated risk for infants was found extremely low (17). These results finally updated the results of previously performed studies on aluminium toxicokinetics (27) where half-life of elimination of aluminium from the body was estimated approximately as 24 hours. Recently published studies provided data on benefits of the use of aluminium-containing vaccines far outweighing any theoretical concerns about the potential negative effects of aluminium on human health.

Analysis of the Immunization Schedule in Poland on the amount of vaccines doses obligatory given during the first year of life revealed that aluminum exposure is much lower than those originating from American Immunization Schedule. According to ACIP recommendations in 2011, maximal aluminium exposure in infants from vaccination schedule over the first year of life has related to 4.225 milligrams of Al^{3+}. Adoption of the same criteria to Immunization Schedule in Poland in 2015 (28), maximal aluminium exposure in infants from vaccination schedule over the first year of life was estimated as 2.850 milligrams of Al^{3+} (see tab. I).

It should be noticed however, that 1,25 mg per human dose as maximal allowable concentration of aluminium present in a vaccine, is far above a real value. The exact concentration per human dose in most vaccines is even two-three times lower. For example according to the Summary of Product Characteristics, DTP (IBSS BIOMED S.A.), whole-cell vaccine against diphtheria, tetanus and pertussis, contains not more than 0,7 milligrams of Al^{3+} per human dose and ENGERIX B (GSK Biologicals S.A.) or Euvax B (LG Life Sciences Poland Sp z o.o.) - vaccines against hepatitis B for infants and children contain 0,25 milligrams of Al^{3+} per human dose.

Table I. Aluminium exposure in infants over the first year of life based on Immunization Schedule in Poland in 2015.

Type of vaccine	Age of administration	Aluminium content (mg) per dose
Hep B	0	0.25
Hep B	2. month	0.25
DTP	2. month	0.7
DTP	3. – 4. month	0.7
DTP	5. – 6. month	0.7
HepB	7. month	0.25

* HepB – Hepatitis B vaccine
DTP – Diphtheria, tetanus and pertussis (whole cell) vaccine (adsorbed)

CONCLUSIONS

Aluminium adjuvants are widely used in vaccines for over six decades, and its both efficiency and safety show good and established profiles. Although they show some limitations such as no Th1 reactivity and stability in temperatures below 2ºC, its easy application, safe profile and low production costs are regarded as reasonable advantages, especially in vaccines used developing countries. Further studies on aluminium-based adjuvants in relation to the immune response and stability achieved by adsorbed antigens might influence the development of their new derivatives or alternatives.

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