Application Note

Overcoming Bacterial Retention and Capacity Issues in Adjuvant Filtration

Adjuvants in vaccine formulations

The vaccine manufacturer's goal is to efficiently meet market demand for safe, efficacious vaccines that minimize patient side-effects and meet cost targets. One method that is increasingly employed to meet these objectives is the addition of an adjuvant to the vaccine formulation. The function of the adjuvant is to stimulate the immune response for the target antigen and increase the level of the immune response, thus improving the efficacy of the vaccine. Adjuvants can also help to induce broad immunity and allow for dose-sparing by decreasing the efficacious dose size, contributing to a more dependable vaccine supply by enabling a greater number of available doses per batch[1,2]. As a result of the advantages that adjuvants can provide to vaccine formulations, they are growing in use.

One particular trend that is driving increased use of adjuvants is the move away from vaccines based on whole viruses to vaccines based on peptides, proteins and subunits. This trend is driven by the need to increase patient safety and to reduce potential side-effects. However, as vaccines are pared down to improve safety, adjuvant use becomes necessary, in some cases, as a way to stimulate the immune system and produce the desired immune response[3].

Recently, adjuvants have received greater public attention because of their use in pandemic influenza vaccines. The benefits of increased immune response and smaller dose size requirements of the adjuvanted formulations are well-suited to the needs of a pandemic in which a large immuno-naïve population must be vaccinated quickly. In the 2008-2009 H1N1 pandemic, vaccines from Novartis and GlaxoSmithKline (GSK), which contained novel emulsion adjuvants, were broadly distributed in Europe, Asia, the Middle East, Canada and Mexico. However, formulations used in the US did not contain the adjuvant because these novel emulsions had not yet been approved for use by the FDA.

Despite the advantages that adjuvants can provide to vaccine formulations, some concerns around long term safety, necessity of adjuvants for vaccine efficacy and limited clinical data, have hampered the exponential growth of adjuvant use[4]. During the H1N1 pandemic, lack of an adjuvant in the US vaccine formulation sparked debate and heightened public awareness of the potential for adjuvant use[2,5].

However, as more data pours in substantiating claims of the benefits of adjuvant use, acceptance of adjuvanted vaccine formulations is growing. In late 2009, the FDA approved the first novel adjuvant for use in the US, a combination of alum and the microbial derivative monophosphoryl Lipid A, in GSK's Cervarix® vaccine for human papillomavirus[6].

Adjuvant use

In spite of recent public and media awareness around adjuvanted vaccines, the use of adjuvants in vaccine formulations is not a recent phenomenon. Adjuvants based on aluminum salts were first introduced in 1937 and have been used in selected vaccine formulations for many years. Currently, novel emulsion and lipid based adjuvant formulations are gaining approval for use in vaccine manufacturing. MF-59®, a novel emulsion adjuvant from Novartis, was approved for use in the Fluad influenza vaccine in 1997 and was introduced to the European market in 2000. Since then, Novartis has included MF-59 adjuvant in some formulations of its pandemic influenza vaccine and GlaxoSmithKline has also commercialized a pandemic influenza vaccine containing AS03, its novel emulsion.

1926
Alum introduced.

1936–1937
Adjuvant properties of Quil A and emulsions discovered.

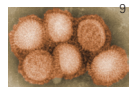
1920s | 1930s | 1940s | 1950s | 1960s | 1970s | 1980s | 1990s | 2000s

1997
Fluad® vaccine contains MF-59 emulsion adjuvant.

NOVARTIS

GlaxoSmithKline

2005
Fendrix® vaccine (EU) and Cervarix® vaccine (EU, US 2009) contain alum and AS04, a MPL adjuvant.



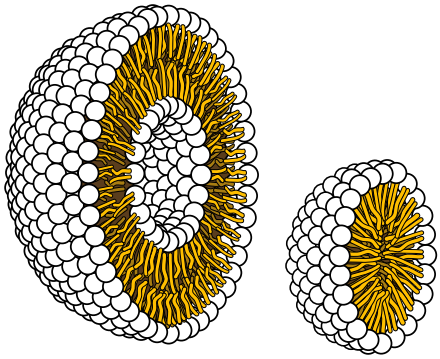
2008–2009
Pandemic flu vaccines contain MF-59 and AS03 emulsion adjuvant.

Adjuvants in recently approved therapeutics have taken a variety of forms, such as oil-in-water emulsions, liposomes and microbial derivatives to address indications such as influenza, Hepatitis A and B, and human papilloma virus. In addition, water-in-oil emulsions, saponins such as QS-21, polymeric microparticles and oligonucleotides (ex. CpG) are currently being used in clinical trials for malaria, cancer, Human Immunodeficiency Virus (HIV), tuberculosis and Hepatitis C vaccines[2].

Adjuvant Types		Indications
Mineral salts (alum)	Approved	Influenza
Oil-in-water emulsions		Hepatitis A
Liposomes		Hepatitis B
Microbial derivatives (ex. MPL)		Human Papilloma Virus
Water-in-oil emulsions	In trials	Malaria
Saponins (ex. QS-21)		Cancer
Polymeric microparticles		HIV
Oligonucleotides (ex. CpG)		Tuberculosis
		Hepatitis C

Processing challenges

While novel adjuvants based on emulsions or liposomes are enabling vaccine manufacturers to meet their goals of efficacy, safety and availability, they pose some processing challenges.



Liposome[10] Micelle[10]

Emulsions and liposomes are suspensions of small particles made up of surfactant or lipid particles. If the interior contains an oil phase, the particle is called a micelle and the suspension is referred to as an emulsion. If the lipid or surfactant molecules form a double layer with an aqueous interior, the particles are called liposomes, which are used both as adjuvants and as drug delivery vehicles.

One particular area for which liposomes have gained attention is cancer therapeutics because of their ability to target cancer cells and to protect healthy cells from toxic cancer therapies[11,12].

Sterile filtration is commonly used to ensure sterility of these temperature-labile compounds. However, these adjuvant formulations can be especially challenging to sterile filtration since the typical particle size of micelles and liposomes (~80–180 nm) is similar to the pore size of a sterilizing-grade filter and only slightly smaller than the bacteria which the filter needs to retain, making the separation difficult. In addition, these streams can have high particle concentrations. The combination of small particle sizes and high particle concentration makes these streams very plugging for sterilizing-grade filters. Filterability is further compromised because emulsions can be more viscous than typical aqueous streams due to friction between the two aqueous phases[13], which further limits flow through the filter membrane. This combination of factors makes these streams more difficult to validate than a typical aqueous based stream and necessitates a balance of operational factors to achieve the optimal combination of filter capacity and retention performance.

Points to consider

We have studied the impact of stream properties, operating conditions and filter types on capacity and retention in these challenging streams and offer the following recommendations:

1. Validation of these streams can be challenging. Find a lab that understands your process constraints, operating conditions and stream properties that influence filtration and can help you design a process which optimizes filtration performance and meets your process objectives.
2. Execute **early** scoping and screening validation studies to identify any processing limitations.
3. In designing capacity tests and retention validations, start by understanding your process variables and simulating worst-case conditions to ensure the process meets your efficiency and sterility requirements.
4. Evaluate the impact of test duration, temperature, pressure/flowrate, and upstream processing steps on filter capacity and retention performance to identify any process constraints relative to these variables.
5. Based on the results of screening and evaluation studies, assess the impact of the adjuvant stream on the bacteria and its interaction with the membrane. As appropriate, evaluate any changes to the stream and the filtration process that may occur over time.
6. Choose a sterilizing-grade filter that does not alter the stream and provides robust, reliable retention while optimizing throughput of these challenging streams.

Conclusion

The ability to sterile filter the proper adjuvant should not be a limiting factor in vaccine development, and it need not be a limiting factor if you perform filter bacterial retention screening studies as part of your filter selection and sizing trials. The knowledge gained from these studies can improve process design and ensure robust performance for throughput and bacterial retention.

For Further Information

Access® Services Validation Sciences Laboratories can help ensure the successful sterile filtration of your adjuvant containing formulations, by providing you with bacterial retention screening studies as part of your filter selection and qualification process. Retention screening studies combined with filter sizing and capacity data will provide you with the critical information that is required to select the optimal filtration design for your unique product formulation.

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