

Incomplete Freund's adjuvant

Catalog # vac-ifa-10, vac-ifa-60 http://www.invivogen.com/ifa

For research use only. Not for use in humans.

Version # 17C09-MM

PRODUCT INFORMATION

Content:

IFA (Incomplete Freund's Adjuvant) is provided as a ready-to-use, clear, pale yellow liquid. IFA is available in two quantities:

- 10 ml: cat. code # vac-ifa-10
- 6 x 10 ml: cat. code # vac-ifa-60

Each 10 ml IFA vial contains 1.5 ml Mannide Monooleate and 8.5 ml Paraffin oil.

Storage and stability

- IFA is shipped at room temperature and should be stored at 4°C.
- IFA is stable for 2 years when properly stored. **DO NOT FREEZE.**

Quality control

- IFA is VacciGrade™ (preclinical grade). It is prepared under strict aseptic conditions and is tested for the presence of endotoxins. IFA is guaranteed sterile and its endotoxin level is <1 EU/ml.
- Adjuvanticity of IFA was evaluated by assessing the levels of total mouse IgGs (mIgGs) and the mIgG1 and mIgG2 isotypes after two consecutive subcutaneous injections of EndoFit™ Ovalbumin/IFA (1:1, v/v) in mice. Results were compared to mice which received the antigen alone.

DESCRIPTION

IFA (Incomplete Freund's adjuvant) is one of the most commonly used adjuvants in research. It is prepared from non-metabolizable oils (paraffin oil and mannide monooleate)¹. IFA does not contain killed *Mycobacterium tuberculosis* found in Complete Freund's Adjuvant (CFA) and is thus less inflammatory¹. IFA induces a predominantly Th2-biased response through the formation of a depot at the injection site and the stimulation of antibody producing plasma cells². It has been suggested that NOD2 modulates the adjuvant effects of IFA³. IFA is utilized to produce water-in-oil emulsions of antigens. It is routinely used for boosting immunizations subsequent to CFA. It can also be used for the initial immunization, particularly when a strong antigen is chosen or moderate antibody levels are sufficient.

Typical results obtained with IFA are shown in figure 1.

Lindblad EB., 2000. Freund's Adjuvants. In: Vaccine adjuvants: Preparation Methods and Research Protocols. Humana Press. Totowa, NJ.2. Petrovsky N. & Aguilar JC., 2004. Vaccine adjuvants: Current state and future trends. Immunol Cell Biol. 82(5): 488-96.
Moreira L0. et al., 2008. Modulation of adaptive immunity by different adjuvant-antigen combinations in mice lacking Nod2. Vaccine 26(46): 5808-13.

METHODS

Preparation of antigen-IFA emulsions

Antigens are preferentially diluted in saline or phosphate buffers. The amount of protein or conjugated peptide used for the primary immunization can be adjusted depending upon availability and immunogenicity of the antigen. For example, mice can be injected subcutaneously (s.c.) with 1 to 10 µg of endotoxin-free ovalbumin (cat. code #vac-pova).

- 1. Mix equal volumes of antigen and IFA using a 21 gauge needle and syringe. A white emulsion will form immediately.
- 2. Mix vigourously for several minutes to form an emulsion with maximum stability.

Note: A stable emulsion is essential to produce high titers of antibodies. 3. Vortex just before use.

The volume of injection depends of the site of administration. *Note: To avoid anaphylaxis, do not use adjuvants for intravenous injection.*

Recommended maximum volumes for injection of antigen/adjuvant mixtures per site of injection for laboratory animals. (Reference: Lindblad EB., 2000. Freund's Adjuvants. In: Vaccine adjuvants: Preparation Methods and Research Protocols. Humana Press. Totowa, NJ).

Species	Max. volume	Injection Site
Mice, hamsters	100 μl	subsutaneous (s.c.)
Mice, hamsters	50 μ1	intramuscular (i.m.)
Guinea pigs	200 μl	s.c. or i.m.
Rats	200 μl	s.c. or i.m.
Rabbits	250 μl	s.c. or i.m.

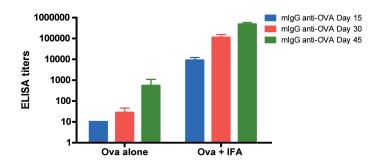


Figure 1. Anti-Ova mIgG levels at 15, 30 and 45 days after the initial immunization. Mice were immunized s.c at 0, 2 and 4 weeks with 1μg of EndoFit[™] Ovalbumin alone or 1 μg of EndoFit[™] Ovalbumin+ IFA (1:1, v/v) in a final volume of 100 μl. Serum anti-OVA mIgG was monitored by ELISA (coated with ovalbumin at 10 μg/ml in PBS).



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RELATED PRODUCTS

Product	Description	Catalog Code
Adjuvants		
AddaVax™	Squalene-Oil-in-water	vac-adx-10
Alhydrogel 2%	Aluminium hydroxide gel	vac-alu-250
Poly(I:C) VacciGrade™	TLR3 agonist	vac-pic
MPLA VacciGrade™	TLR4 agonist	vac-mpla
Flagellin FliC VacciGrade™	TLR5 agonist	vac-fla
Gardiquimod VacciGrade™	TLR7 agonist	vac-gdq
Imiquimod VacciGrade™	TLR7 agonist	vac-imq
R848 VacciGrade™	TLR7/8 agonist	vac-r848
ODN 1826 VacciGrade™	murine TLR9 agonist	vac-1826-1
ODN 2006 VacciGrade™	human TLR9 agonist	vac-2006-1
N-glycolyl-MDP VacciGrade™	NOD2 agonist	vac-gmdp
OVA Antigens		
EndoFit™ Ovalbumin	For in vivo use; endotoxin level <1EU/mg	vac-pova
Ovalbumin	For detection; Western, ELISA	vac-stova
Ova 257-264	For detection; ELISPOT	vac-sin
Ova 323-339	For detection; ELISPOT	vac-isq

