CL401 VacciGrade™

Dual TLR2 & TLR7-based adjuvant

Catalog # vac-401-5

http://www.invivogen.com/cl401-vaccigrade

For research use only. Not for use in humans.

Version # 17C24-MM

PRODUCT INFORMATION

Content:

• 5 mg of CL401 VacciGrade™

• 10 ml sterile endotoxin-free physiological water (NaCl 0.9%)

Storage:

- CL401 VacciGrade™ is shipped at room temperature. Store lyophilized product at -20 °C. Lyophilized product is stable for 1 year at -20 °C.

- Upon resuspension, store at 4 °C. Resuspended product is stable for 6 months at 4 °C. Do not store resuspended product in plastic tubes.

Quality control

CL401 VacciGrade™ is a preclinical grade preparation of CL401. It is prepared under strict aseptic conditions and is tested for the presence of endotoxins. CL401 VacciGrade™ is guaranteed sterile and its endotoxin level is <5 EU/mg.

BACKGROUND

InvivoGen has developed a series of novel molecules designed to induce potent immune responses through the combined activation of several pattern recognition receptors (PRRs) that trigger different innate immune signaling pathways. These molecules are agonists for TLR2, TLR7 or both. Agonists that activate TLR2 are derived from the well-established TLR2 ligand, Pam2CSK4, and those recognized by TLR7 are derived from the 8-hydroxyadenine derivative CL264, a TLR7 agonist recently developed by InvivoGen.

TLR2 and TLR7 are two PRRs with distinct characteristics. TLR2 is a cell surface receptor expressed by many cell types, while TLR7 is an endosomal receptor expressed predominantly in plasmacytoid dendritic cells (pDC) and to a lesser extent in B cells. TLR2 signaling triggers the NF-κB pathway and the production of pro-inflammatory cytokines, such as TNF-α, whereas TLR7 signaling induces mainly the IRF pathway and the production of IFN-α.

DESCRIPTION

CL401 is a small lipophilic molecule comprising an 8-hydroxyadenine compound conjugated with a Pam2C group. This bipartite structure confers to CL401 the ability to efficiently stimulate both TLR7 and TLR2, respectively. *In vivo* tumor studies have demonstrated intratumoral injection of CL401 leads to a significant antitumor activity (data in InvivoGen Insight Spring 2013). When tested as an adjuvant in mice, CL401 exhibits a Th1-dominated immune response (see figures 1 & 2, overleaf).

CHEMICAL PROPERTIES

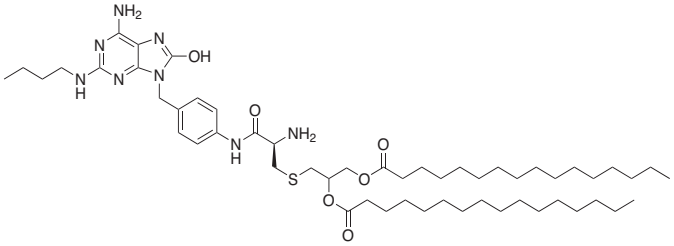
Synonym: S-(2,3-bis(palmitoyloxy)-(2RS)propyl)-(R)-cysteinyl 4-((6-amino-2(butyl amino)-8-hydroxy-9H-purin-9-yl)methyl) aniline

Formula: $C_{54}H_{92}N_8O_4S$

Molecular weight: 981 g/mol

Solubility: DMSO (20 mg/ml)

Structure:



METHODS

Working Concentration: 20-50 µg/mouse

Preparation of sterile stock solution (1 mg/ml)

1. Resuspend CL401 VacciGrade™ by adding 5 ml of DMSO (not provided) to 5 mg of product and vortexing until completely dissolved.
2. Store at 4 °C. Do not store resuspended product in plastic tubes.
3. Further dilutions can be prepared using sterile aqueous solutions for injection, such as the sterile endotoxin-free physiological water (provided).

TECHNICAL SUPPORT

InvivoGen USA (Toll-Free): 888-457-5873

InvivoGen USA (International): +1 (858) 457-5873

InvivoGen Europe: +33 (0) 5-62-71-69-39

InvivoGen Hong Kong: +852 3-622-34-80

E-mail: info@invivogen.com

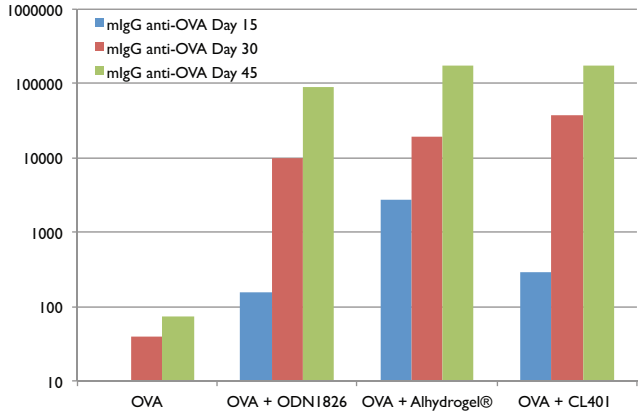


Figure 1. Anti-OVA mIgG levels at 15, 30 and 45 days after the initial immunization in different groups. Mice were immunized s.c at 0, 2 and 3 weeks with 1 µg of EndoFit™ Ovalbumin alone, 1 µg of EndoFit™ Ovalbumin/ODN182 VacciGrade™ (10 µg/ml), 1 µg of EndoFit™ Ovalbumin/Alhydrogel® adjuvant 2% (1:1, v/v), or 1 µg of EndoFit™ Ovalbumin/CL401 VacciGrade™ (20 µg/ml), in a final volume of 100 µl. Serum anti-OVA total mIgG was monitored by ELISA (coated with ovalbumin at 10 µg/ml in PBS).

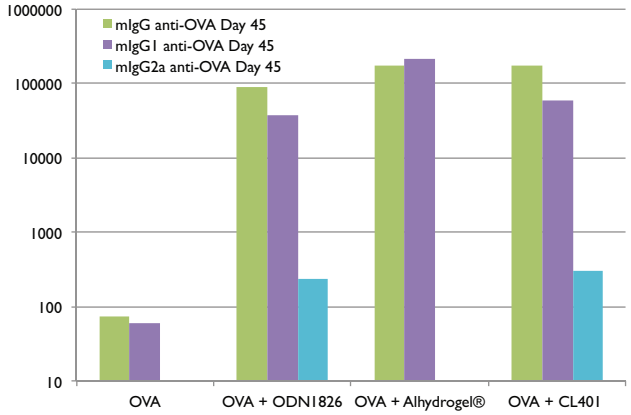


Figure 2. Anti-OVA mIgG levels 45 days after the initial immunization in different groups. Mice were immunized s.c at 0, 2 and 3 weeks with 1 µg of EndoFit™ Ovalbumin alone, 1 µg of EndoFit™ Ovalbumin/ODN182 VacciGrade™ (10 µg/ml), 1 µg of EndoFit™ Ovalbumin/Alhydrogel® adjuvant 2% (1:1, v/v), or 1 µg of EndoFit™ Ovalbumin/CL401 VacciGrade™ (20 µg/ml), in a final volume of 100 µl. Serum anti-OVA total mIgG, anti-OVA mIgG1 and anti-OVA mIgG2a were monitored by ELISA (coated with ovalbumin at 10 µg/ml in PBS).

Alhydrogel® is a trademark which belongs to Brenntag Biosector A/S and which is registered in a large number of countries and regions worldwide.

RELATED PRODUCTS

Product	Description	Catalog Code
AddaVax™	Squalene-Oil-in-water	vac-adx-10
Adilipoline (CL413) VacciGrade™	TLR2 & TLR7 agonist	vac-c413
Alhydrogel® 2%	Aluminium hydroxide gel	vac-alu-250
Flagellin FliC VacciGrade™	TLR5 agonist	vac-fla
Gardiquimod VacciGrade™	TLR7 agonist	vac-gdq
IFA	Incomplete Freund's adjuvant	vac-ifa-10
Imiquimod VacciGrade™	TLR7 agonist	vac-imq
MPLA VacciGrade™	TLR4 agonist	vac-mpla
MPLAs (synthetic) VacciGrade™	TLR4 agonist	vac-mpls
N-glycolyl-MDP VacciGrade™	NOD2 agonist	vac-gmdp
ODN 1585 VacciGrade™	Murine TLR9 agonist	vac-1585-1
ODN 1826 VacciGrade™	Murine TLR9 agonist	vac-1826-1
ODN 2006 VacciGrade™	Human TLR9 agonist	vac-2006-1
ODN 2395 VacciGrade™	Human/murine TLR9 agonist	vac-2395-1
Pam3CSK4 VacciGrade™	TLR2 agonist	vac-pms
Poly(I:C) VacciGrade™	TLR3 agonist	vac-pic
Quil-A® adjuvant	Saponin vaccine adjuvant	vac-quil
R848 VacciGrade™	TLR7/8 agonist	vac-r848
TDB VacciGrade™	Mincle agonist	vac-tdb
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

TECHNICAL SUPPORT
InvivoGen USA (Toll-Free): 888-457-5873
InvivoGen USA (International): +1 (858) 457-5873
InvivoGen Europe: +33 (0) 5-62-71-69-39
InvivoGen Hong Kong: +852 3-622-34-80
E-mail: info@invivogen.com