产品说明书

26 版

基本信息

产品编号：	R10022
产品名称：	雷西莫特

产品简介：

一种作用于 TLR7 和 TLR8 免疫反应调节剂，EC50 分别为 0.26μm 和 6.4μm。

靶点：	TLR7	TLR8

体外研究：

Resiquimod (R-848) 诱导半抗原和过敏原特异性循环 T 细胞 (包括 TH2 效应子) 产生 IFN-γ，甚至丧失产生 IL-4 的能力。

Resiquimod (R848) 以剂量依赖性方式增强 PBL 增殖，并增加 BrdU 掺入试验中 BrdU 阳性细胞的数量。用 R848 处理的细胞表现出显著增加 (3.5 倍) 的荧光素酶 (NF-κB 活性的报告分子) 活性。

体内研究：

Resiquimod 可用于动物建模，构建大小鼠细胞因子释放模型、免疫介导的心脏组织损伤模型。Resiquimod (R-848) (50 μg/只，肌肉注射途径) 显著上调在 SPF 鸡中 IFN-α、IFN-β、IFN-γ、IL-1β、IL-4、iNOS 和 MHC-II 基因的表达。

Resiquimod 的药代动力学特性的特点是半衰期短，导致相关剂量下的 AUC 较低，同时 Cmax 较高。

使用方法（供参考）：

一：体外实验

DMSO：溶于 DMSO(10mg/mL 加热至 60 ° C)

浓度 \ 质量 / 溶液体积	1mg	5mg	10mg
1 mM	3.1809 mL	15.9043 mL	31.8086 mL
5 mM	0.6362 mL	3.1809 mL	6.3617 mL
10 mM	0.3181 mL	1.5904 mL	3.1809 mL

储备液的保存方式：-80° C, 1 year; -20° C, 6 months。

二：体内实验

1、请依序添加每种溶剂： 5% DMSO→40% PEG300→5% Tween-80→50% Saline

Solubility: ≥ 2.5 mg/mL (7.95 mM); 澄清溶液

2、请依序添加每种溶剂： 10% DMSO→40% PEG300→5% Tween-80→45% Saline

Solubility: ≥ 2.08 mg/mL (6.62 mM); 澄清溶液

此方案可获得 ≥ 2.08 mg/mL（饱和度未知）的澄清溶液。

以 1 mL 工作液为例，取 100 μL 20.8 mg/mL 的澄清 DMSO 储备液加到 400 μL PEG300 中，混合均匀；再向上述体系中加入 50 μL Tween-80，混合均匀；然后再继续加入 450 μL 生理盐水 定容至 1 mL。

3、请依序添加每种溶剂： 10% DMSO→90% (20% SBE-β-CD in Saline)

Solubility: ≥ 2.08 mg/mL (6.62 mM); 澄清溶液

此方案可获得 ≥ 2.08 mg/mL（饱和度未知）的澄清溶液。

以 1 mL 工作液为例，取 100 μL 20.8 mg/mL 的澄清 DMSO 储备液加到 900 μL 20% 的 SBE-β-CD 生理盐水水溶液 中，混合均匀。

4、请依序添加每种溶剂： 10% DMSO→90% Corn Oil

Solubility: ≥ 2.08 mg/mL (6.62 mM); 澄清溶液

此方案可获得 ≥ 2.08 mg/mL（饱和度未知）的澄清溶液，此方案实验周期在半个月以上的动物实验酌情使用。

以 1 mL 工作液为例，取 100 μL 20.8 mg/mL 的澄清 DMSO 储备液加到 900 μL 玉米油中，混合均匀。

注：工作液建议您现用现配，当天使用。

＜1mg/ml 表示微溶或不溶。

普西唐提供的所有化合物浓度为内部测试所得，实际溶液度可能与公布值有所偏差，属于正常的批间细微差异现象。

请根据产品在不同溶剂中的溶解度选择合适的溶剂配制储备液；一旦配成溶液，请分装保存，避免反复冻融造成的产品失效。

三、激酶实验

For luciferase assay,FG-9307 cells are transfected with the firefly NF-κB-specific luciferase reporter vector pNFκB-Met-Luc2.Transfection efficiency is monitored by co-transfection with the pSEAP2 control vector,which constitutively expresses the human secreted enhanced alkaline phosphatase (SEAP).Then the cells are treated with Resiquimod (R848,1μg/mL),CQ (10μM),CQ plus R848 or PBS and incubated at 22℃ for 24h.The culture medium of the transfectants is then analyzed for luciferase activity and SEAP activity using Luciferase Assay Kit and the Great EscAPe™ SEAP Chemiluminescence Detection Kit,respectively.The assay is performed three times.

四、细胞实验

For inhibition of lysosomal acidification,cells are incubated with 10μM CQ for 1h before Resiquimod (R848) treatment.After treatment,20μL of 5mg/mL MTT is added to the plate.The plate is incubated at 22℃ for 4h,and 200μL dimethyl sulfoxide is added to the plate to dissolve the reduced formazan.The plate is then read at 490nm with a microplate reader.To determine the effect of Myd88 inhibition on R848-induced cell proliferation,the Myd88 inhibitor Pepinh-MYD andthe control peptide Pepinh-Control are added to PBL at the concentration of 50μM,and the plate is incubated at 22℃ for 6h.After incubation,the cells are treated with R848 and subjected to MTT assay as above.To determine the effect of NF-κB inactivation on R848-induced cell proliferation,BAY-11-7082,an irreversible inhibitor of IκB-α phosphorylation,is added to the cells at the concentration of 1μM,and the plate is incubated at 22℃ for 1h.After incubation,the cells are treated with R848 and subjected to MTT assay as earlier.All experiments are performed three times.

五、动物实验

A total of 40 SPF chickens of two-week old are allotted to one of the following four experimental groups (n=10/group):Group A:PBS control;Group B:inactivated NDV vaccine;Group C:commercial oil adjuvanted inactivated NDV vaccine prepared from lentogenic strain and Group D:combination of inactivated NDV vaccine and R-848 (50μg/bird).Vaccine or PBS is administered by intramuscular route in the thigh muscle.A booster dose is given 14-day post immunization (d.p.i).Two weeks post-booster,experimental SPF birds are challenged with velogenic strain of NDV (105 ELD50 per bird) intramuscularly.Clinical signs and mortality are observed daily till 14 day post-challenge (d.p.c).Cloacal swabs (n=6/group) are collected from the birds on day 0,4,7 and 14 post-challenge and inoculated into 10-day old embryonated chicken eggs (n=3 eggs/sample) through intra-allantoic route.Three day post-inoculation, the allantoic fluid is checked for the NDV growth by spot

haemagglutination using 10% chicken RBC.

注意事项：

1、为了您的安全和健康，请穿实验服并戴一次性手套操作。
2、以上信息仅做参考交流之用。

储存：-20° C