

本维莫德调控 NRF2/ROS/NLRP3 通路减轻特应性皮炎

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摘要 **目的** 探讨本维莫德(BVM)治疗特应性皮炎(AD)的作用机制。**方法** 利用 TNF- α 和 IFN- γ 刺激 HaCaT 细胞,将细胞分为 NC 组、TNF- α /IFN- γ 组、TNF- α /IFN- γ 、BVM 组、TNF- α /IFN- γ 、BVM、ML385 组;DNCB 诱导 Balb/c 小鼠的 AD 模型,动物分为 CON 组、DNCB 组、DNCB + BVM 组、DNCB + 他克莫司(TAC)组,评估 BVM 效果,及其在抗氧化及焦亡调节中的作用。**结果** 与对照组相比,BVM 抑制了 TNF- α 和 IFN- γ 刺激的 HaCaT 细胞炎症反应,改善了 DNCB 诱导的 AD 小鼠模型皮损和炎症;同时,使核因子 E2 相关因子 2(NRF2)相关抗氧化应激蛋白表达显著增加,细胞活性氧(ROS)水平及焦亡蛋白表达显著降低。**结论** BVM 可通过激活 NRF2/ROS/NLRP3 通路抑制焦亡,从而减轻 AD 中的炎症反应。

关键词 本维莫德;特应性皮炎;焦亡;NLRP3;NRF2;ROS**中图分类号** R 758.23**文献标志码** A **文章编号** 1000-1492(2025)08-1490-09

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特应性皮炎(atopic dermatitis, AD)是常见的慢性炎症性皮肤病,已有研究^[1]表明焦亡与 AD 的发病存在密切关系。焦亡通常由炎症小体 NOD 样受体热蛋白结构域相关蛋白 3(NOD-like receptor family, pyrin domain-containing 3, NLRP3)触发并由 Gasdermin 蛋白家族成员执行,导致细胞内容物的释

放^[2]。活性氧(reactive oxygen species, ROS)被认为是 NLRP3 炎症小体激活的关键介质之一^[3]。炎症小体的激活和随后的焦亡导致促炎细胞因子释放,放大皮肤中的炎症反应并导致 AD 的发生发展^[4]。本维莫德(benvitimod, BVM)是一种小分子治疗药物,可激活核因子 E2 相关因子 2(nuclear factor erythroid-2-related factor 2, NRF2)通路,参与 ROS 的细胞防御^[5]。目前尚不清楚 BVM 是否能通过抑制 ROS 的生成干扰焦亡而减轻 AD。该研究通过细胞和动物实验探索 BVM 的抗氧化应激及抑制焦亡的作用。结果表明 BVM 可激活 NRF2/ROS/NLRP3 通路,抑制焦亡,从而减轻 AD 中的炎症反应。该研究为治

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eukaryotic expression recombinant plasmid pcDNA3.1(+)-human GIF-His tag was constructed, and the gastric intrinsic factor was purified by nickel column after expression in HEK293F cells. The purity and activity of purified gastric intrinsic factor were verified by SDS-PAGE, Western blot and indirect ELISA. **Results** Gastric factor contained 417 amino acids and was a hydrophilic acid stable protein. It was a secreted protein with Sec original signal peptide. pcDNA3.1(+)-human GIF-His tag recombinant plasmid was successfully constructed and soluble expression was obtained in HEK293F cell expression system. **Conclusion** The eukaryotic source of human gastric intrinsic factor is successfully prepared, and the bioinformatics results show that the protein is a hydrophilic acid stable secreted protein, laying a foundation for the subsequent use of this protein as an immunogen and protein calibrator to construct an immunoassay for gastric factor.

Key words gastric intrinsic factor; eukaryotic expression; bioinformatics; protein purification; activity identification; ELISA**Fund program** Major Science and Technology Program of Hainan Province (No. ZDKJ2021040)**Corresponding author** Wang Bo, E-mail: wangqugans@163.com

疗 AD 提供了新的思路,未来可通过抑制焦亡通路来治疗炎症性皮肤病。

1 材料与方法

1.1 材料

1.1.1 实验所用细胞 永生化人角质形成细胞 (HaCaT) 购自南京凯基生物科技发展有限公司。

1.1.2 实验试剂 NLRP3 (T5565)、Gasdermin D (GSDMD) (P30823)、半胱氨酸天冬氨酸特异性蛋白酶 1 (cysteiny aspartate specific proteinase-1, caspase-1) (M025280) (上海艾比玛特医药科技有限公司)、白细胞介素-18 (interleukin-18, IL-18) (10663-1-AP)、IL-1 β (WL00891)、NRF2 (80593-1-RR)、3-磷酸甘油醛脱氢酶 (glyceraldehyde-3-phosphate dehydrogenase, GAPDH) (60004-1-Ig)、山羊抗鼠 (SA00001-1)、山羊抗兔 (SA00001-2)、组蛋白 H3 (Histon-H3) (81984-2-RR) (武汉三鹰生物技术有限公司)、活性氧检测试剂盒 (15122020) (沈阳万类生物技术有限公司)、醌氧化还原酶 1 [quinoneoxidoreductase1 NAD(P)H, NQO1] (SC-32793)、血红素加氧酶-1 (heme oxygenase-1, HO-1) (SC-390991) (美国 Santa 公司)、细胞计数试剂盒-8 (cell counting kit-8, CCK-8) (521942) (合肥白鲨生物技术有限公司)、乳酸脱氢酶 (lactate dehydrogenase, LDH) 细胞毒性检测试剂盒 (C0016)、Calcein/PI 细胞活性与细胞毒性检测试剂盒 (C2015S) (上海碧云天生物技术有限公司)、组织切片活性氧检测试剂盒 (BB-470516) (上海贝博生物技术有限公司)。

1.2 方法

1.2.1 细胞培养 将 HaCaT 细胞置于 10% 胎牛血清的 DMEM 培养基中 (美国 Gibco Life Sciences 公司), 在 37 °C 和 5% CO₂ 培养箱里培养。使用 10 ng/mL 的肿瘤坏死因子- α (tumor necrosis factor- α , TNF- α) 和干扰素- γ (interferon- γ , IFN- γ) (苏州近岸蛋白质科技股份有限公司) 刺激 HaCaT 细胞, 用二甲亚砜制备 100 mmol/L BVM 储备溶液 (广东中

昊药业有限公司) 观察其作用。

1.2.2 动物实验 使用 Balb/c 小鼠 (雌性, 18 ~ 22 g, 6 ~ 8 周, 24 只), 饲养在 SPF 级别环境中, 12 h 的光暗循环条件, 室温 20 ~ 25 °C, 湿度 50% ~ 60%, 自由进食、饮水, 适应性饲养 7 d 后开始实验。小鼠购自江苏集萃药康生物, 动物实验经安徽医科大学伦理委员会批准 (批号: LLSC20242243)。

将小鼠采用随机数字表法分为 CON 组、DNCB 组、DNCB + BVM 组、DNCB + 他克莫司 (tacrolimus, TAC) 组, 每组 6 只。DNCB 组于实验第 1、4、7 天使用 2% DNCB (20 μ L) 溶液均匀涂抹在小鼠耳部皮肤, 第 14 天开始, 每 2 d 用 0.5% DNCB (20 μ L) 溶液进行连续致敏刺激。DNCB + BVM/TAC 组用 0.5% DNCB 涂抹于耳部 4 h 后增加 1% BVM 乳膏 (广东中昊药业有限公司) 或 0.1% TAC 乳膏 (沈阳安斯泰来制药有限公司) 治疗。对照组给予溶液基质 (丙酮: 橄榄油 = 3: 1) 刺激, 时间频率和 DNCB 组相同。流程见图 1。处决所有小鼠后, 收集耳部组织保存于 4% 多聚甲醛或 -80 °C 冰箱中, 用于后期实验。

1.2.3 皮炎评分、耳朵厚度、脾脏指数的评估及组织病理观察 检测给药前后各组小鼠耳部皮损炎症情况^[6]。使用游标卡尺测量每只小鼠的双耳厚度, 测量耳廓同一区域 3 次, 取平均厚度。测量小鼠的体质量与脾脏质量。将组织制成石蜡包埋或新鲜冰冻组织切片。苏木精 - 伊红染色 (hematoxylin - eosin staining, H&E) 观察组织病理。甲苯胺蓝染色 (toluidine blue staining, TB) 检测肥大细胞。每张病理切片随机选择 3 个区域检测表皮和真皮厚度、肥大细胞数。免疫组织化学 (immunohistochemistry, IHC) 法检测 NLRP3、GSDMD、Caspase-1 (1: 100) 表达情况。使用活性氧试剂盒测量小鼠耳部新鲜冰冻组织中的 ROS 水平, 在正置显微镜下 (德国徕卡 DM6B) 观测 ROS 荧光强度。

1.2.4 细胞活力、ROS 及 LDH 检测 根据相关试剂盒说明书处理细胞, 通过多功能酶标仪 (PE

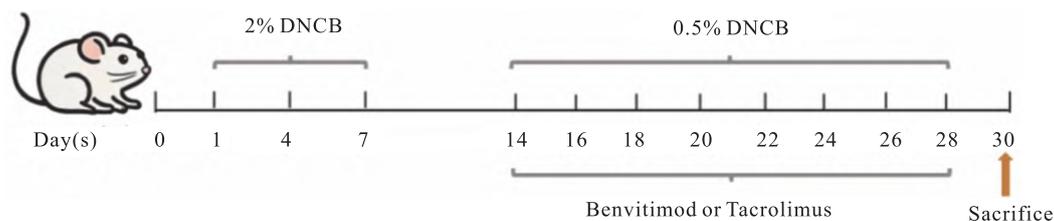


图 1 DNCB 诱导特应性皮炎小鼠流程图

Fig. 1 Flow chart of DNCB-induced atopic dermatitis mice

EnSpire)检测细胞存活率、ROS与LDH水平。

1.2.5 碘化丙啶(propidium iodide staining, PI)染色 根据试剂盒方案进行洗涤染色检测。通过全自动活细胞成像仪对各组染色的活细胞进行成像。

1.2.6 Western blot(WB)实验 使用RIPA缓冲液收集细胞或动物组织蛋白质,SDS-PAGE进一步分离并转移到硝酸纤维素膜,封闭后分别用一抗(1:1 000 NLRP3/GSDMD/Caspase-1/IL1 β /NRF2/NQO1/HO-1,1:3 000 IL-18,1:50 000 GAPDH/Histon-H3)在4℃下孵育过夜。之后将膜与二抗(1:10 000山羊抗兔或山羊抗鼠)在4℃下孵育1 h。最后进行条带检测。

1.2.7 实时荧光定量PCR(quantitative real-time PCR,qRT-PCR)实验 提取小鼠皮肤组织中的mRNA进行分析。NRF2引物序列:正向5'-TCCTG-GAGTAAGTCGAGAAGTGT-3',反向5'-GTTGAAAC-TGAGCGAAAAAGGC-3'。

1.3 统计学处理 采用*t*检验对两组进行比较,多组间的比较采用单因素方差分析。使用Graph Pad Prism 8.0进行数据分析。所有数据以 $\bar{x} \pm s$ 表示。 $P < 0.05$ 为差异有统计学意义。

2 结果

2.1 BVM抑制HaCaT细胞焦亡 NLRP3的过度激活可诱导焦亡导致炎症细胞因子的大量释放,为探究BVM能否减轻HaCaT细胞中焦亡的程度,首先使用CCK-8试剂盒测定不同浓度的BVM溶液对HaCaT细胞活力的影响。结果显示20、40、60 $\mu\text{mol/L}$ 的BVM对HaCaT细胞无明显毒性,细胞活力均不低于90%(图2A)。随后,使用TNF- α 和IFN- γ 建立炎症细胞模型。刺激24 h后,细胞内ROS水平明显升高,在BVM的治疗作用下,细胞内ROS水平随BVM浓度的升高而降低($F = 54.86, P < 0.0001$) (图2B)。WB结果显示,随着BVM浓度的增加,焦亡蛋白NLRP3($F = 81.78, P < 0.0001$)、GSDMD-N($F = 47.28, P < 0.0001$)、Caspase-1($F = 73.51, P < 0.0001$)、IL-1 β ($F = 52.73, P < 0.0001$)和IL-18($F = 55.17, P < 0.0001$)表达水平逐渐降低,NRF2($F = 51.56, P < 0.01$)及下游抗氧化基因HO-1($F = 36.77, P < 0.01$)和NQO1($F = 38.54, P < 0.01$)的蛋白表达水平依次升高(图2C-E)。由于BVM在60 $\mu\text{mol/L}$ 治疗效果最好,后续实验选择此浓度。以上结果表明,BVM可抑制NLRP3炎症小体的激活进而抑制细胞发生焦亡。与此同时,BVM促进

NRF2移位至细胞核,并激活下游抗氧化基因HO-1和NQO1。

2.2 BVM通过激活NRF2通路抑制TNF- α 和IFN- γ 诱导的焦亡 探索NRF2与ROS以及焦亡的关系,加入NRF2抑制剂ML385(5 $\mu\text{mol/L}$)处理HaCaT细胞。结果显示,加入ML385处理后,ROS水平高于BVM组,这表明ML385的处理可消除BVM对ROS产生的抑制作用(图3A)。收集细胞上清液以测量LDH释放量。结果表明,ML385阻断了BVM对LDH的抑制作用($F = 59.25, P < 0.0001$) (图3B)。PI染色结果显示BVM可降低TNF- α 和IFN- γ 处理后的细胞死亡率,而ML385则阻断了BVM的这种作用(图3C)。WB结果显示,ML385可阻断BVM对NRF2($F = 29.73, P < 0.001$)、HO-1($F = 95.76, P < 0.0001$)、NQO1($F = 17.73, P = 0.0002$)蛋白的增强作用,同时抵消了BVM对NLRP3($F = 15.80, P < 0.001$)、GSDMD-N($F = 45.80, P < 0.001$)、Caspase-1($F = 38.76, P < 0.001$)、IL-1 β ($F = 49.58, P < 0.0001$)和IL-18($F = 51.81, P < 0.0001$)蛋白的抑制作用(图3D-F)。这些结果表明,BVM可通过激活NRF2通路抑制ROS的产生,同时抑制NLRP3炎症小体激活诱导的焦亡。

2.3 BVM对焦亡的抑制受ROS水平的调控 为验证ROS是否导致上述焦亡过程,向细胞中加入20 $\mu\text{mol/L}$ 的ROS激活剂(H_2O_2)和10 mmol/L 的ROS抑制剂(NAC)。加入 H_2O_2 后,NLRP3($F = 74.57, P < 0.0001$)、GSDMD-N($F = 68.76, P < 0.0001$)、Caspase-1($F = 38.48, P < 0.0001$)的表达水平较TNF-N/INF- γ 均升高(图4A、B)。而加入NAC后,NLRP3($F = 77.46, P < 0.0001$)、GSDMD-N($F = 42.31, P < 0.0001$)、Caspase-1($F = 74.03, P < 0.0001$)表达水平较TNF- α /IFN- γ 组有所下降(图4C、D)。因此,TNF- α 和IFN- γ 诱导的焦亡,可被 H_2O_2 进一步上调并被NAC下调。

2.4 BVM改善特应性皮炎小鼠症状 为探讨BVM在体内作用,本研究构建了DNCB诱导的AD模型小鼠(图1)。小鼠耳部皮肤出现明显红斑、部分糜烂、结痂伴有搔抓次数增多等表现,显示AD造模成功。病理染色结果显示,与DNCB组相比,DNCB+BVM/TAC组耳部表皮明显变薄($F = 16.87, P < 0.0001$)、肥大细胞浸润数明显减少,炎症减轻(图5A、B);脾脏大小、脾脏指数($F = 283.4, P < 0.0001$)以及皮炎评分($F = 41.94, P < 0.0001$)显著下降(图5C-E),显示了BVM和TAC具有相同

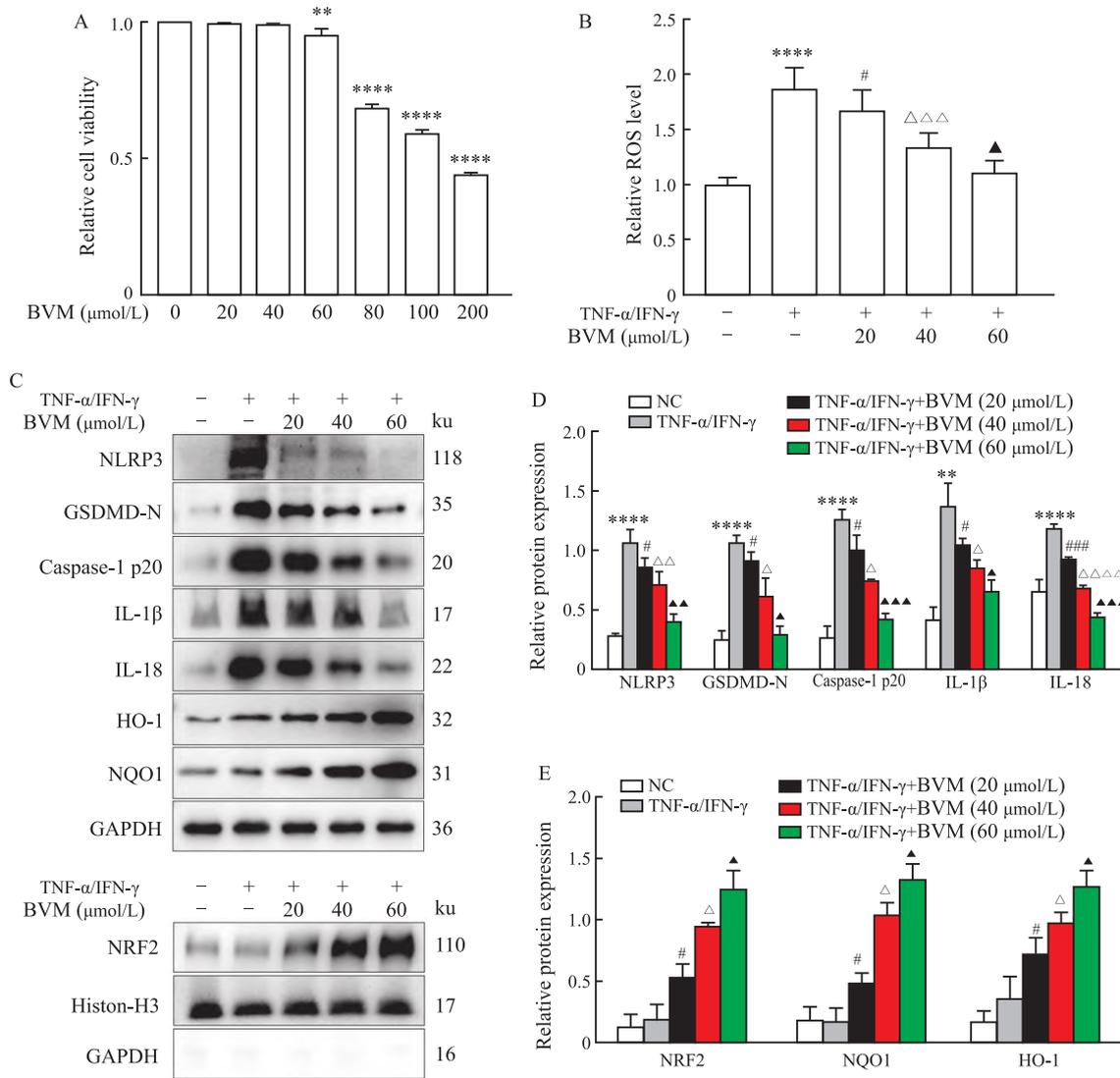


图2 BVM抑制HaCaT细胞焦亡

Fig. 2 BVM inhibited the pyroptosis of HaCaT cell

A: Cell viability was detected by CCK-8 in each group; B: ROS levels was detected by ROS Assay Kit in each group; C, D, E: WB analysis was used to measure the expression levels of NLRP3, GSDMD-N, Caspase-1 p20, IL-18, IL-1β and NRF2, HO-1, NQO1 protein expression in each group; ** $P < 0.01$, **** $P < 0.0001$ vs NC group; # $P < 0.05$, ### $P < 0.01$ vs TNF-α/IFN-γ group; △ $P < 0.05$, △△ $P < 0.001$, △△△ $P < 0.001$, △△△△ $P < 0.0001$ vs TNF-α/IFN-γ + BVM(20 μmol/L) group; ▲ $P < 0.05$, ▲▲ $P < 0.01$, ▲▲▲ $P < 0.001$ vs TNF-α/IFN-γ + BVM(40 μmol/L) group.

的效果。

2.5 BVM改善特应性皮炎小鼠ROS水平及焦亡程度 利用小鼠模型验证BVM能否改善ROS水平及焦亡程度。DNCB组ROS荧光强度与对照组相比明显升高,而DNCB+BVM/TAC组ROS荧光水平对比DNCB组则显著下降(图6A)。qRT-PCR结果显示,BVM显著提高NRF2 mRNA的表达($F = 258.3, P < 0.0001$)(图6B)。WB结果显示,对比DNCB组,DNCB+BVM/TAC组的NLRP3($F =$

$48.36, P < 0.0001$), GSDMD-N($F = 11.44, P < 0.01$), Caspase-1($F = 33.65, P < 0.0001$), IL-18($F = 17.45, P < 0.001$)和IL-1β($F = 22.08, P < 0.001$)蛋白水平明显下降,NRF2($F = 37.51, P < 0.0001$)、HO-1($F = 71.49, P < 0.0001$)、NQO1($F = 28.50, P < 0.0001$)蛋白水平明显上升(图6C-E)。IHC检测结果与WB结果一致(图6F)。以上结果表明,BVM可通过增强NRF2表达,降低ROS水平,抑制焦亡而改善AD(图7)。

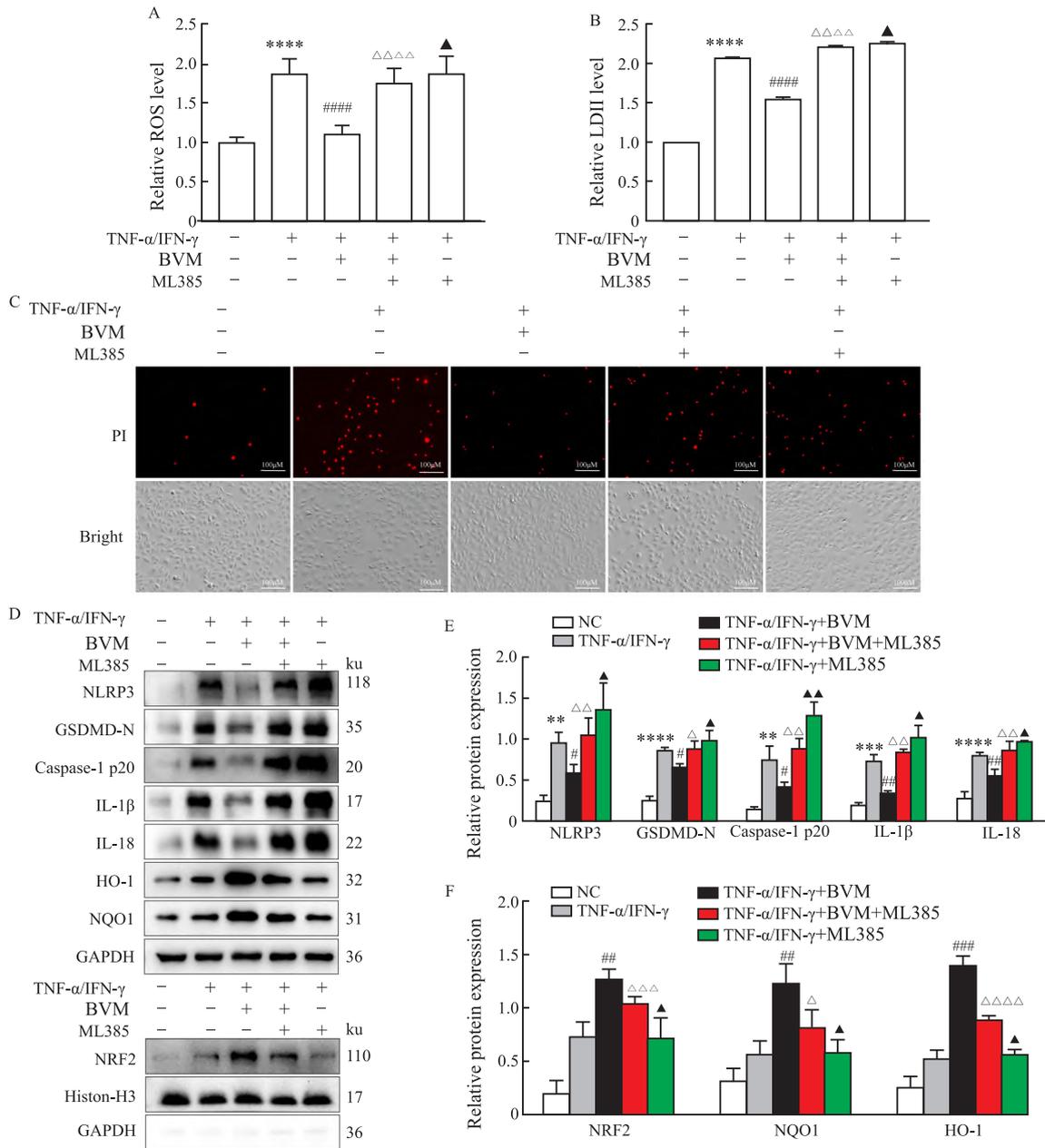


图3 BVM通过激活NRF2通路抑制TNF-α和IFN-γ诱导的焦亡

Fig.3 Benvitimod inhibited TNF-α/IFN-γ-induced pyroptosis via NRF2 activation

A: ROS Assay Kit was used to detect the level of ROS in cells in each group; B: Lactate dehydrogenase cytotoxicity detection kit was used detect LDH levels in cells in each group; C: PI staining ×100; D, E, F: WB analysis was used to measure the expression levels of NLRP3, GSDMD-N, Caspase-1 p20, IL-18, IL-1β and NRF2, HO-1, NQO1 proteins in each group; ** $P < 0.01$, *** $P < 0.001$, **** $P < 0.0001$ vs NC group; # $P < 0.05$, ## $P < 0.01$, ### $P < 0.001$, #### $P < 0.0001$ vs TNF-α/IFN-γ group; △ $P < 0.05$, △△ $P < 0.01$, △△△ $P < 0.001$, △△△△ $P < 0.0001$ vs TNF-α/IFN-γ BVM group; ▲ $P < 0.05$, ▲▲ $P < 0.01$ vs TNF-α/IFN-γ+BVM+ML385 group.

3 讨论

AD的发病机制涉及各种病理因素, NLRP3炎症小体的激活可能是其发展的关键触发因素^[7]。焦亡会加重AD等炎症性皮肤病^[8]。在AD患者皮损中检测出焦亡相关蛋白水平的升高^[9]。本研究

中, BVM可以通过激活NRF2/ROS/NLRP3调节轴, 显著减少AD细胞模型与动物模型中焦亡相关蛋白的产生。

在AD患者中, 氧化应激是评估皮肤炎症中重要指标。过量ROS会诱发高氧化应激, 加重AD患者皮肤中的氧化损伤和膜脂质过氧化, 导致细胞死

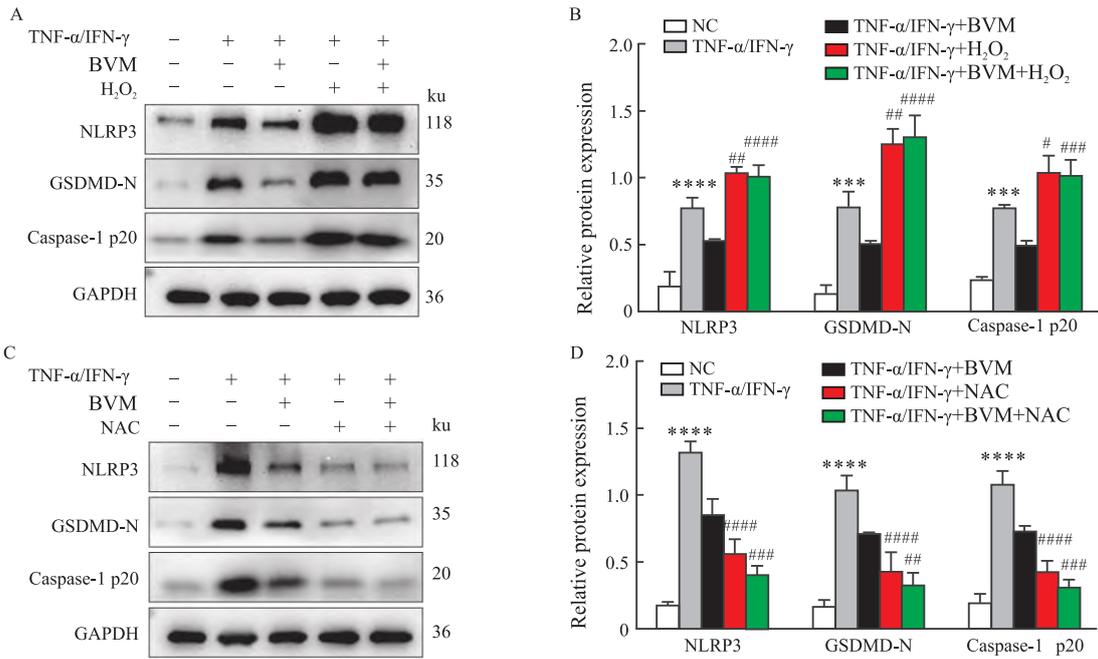


图4 BVM对凋亡的抑制受ROS水平的调控

Fig.4 The inhibition of pyroptosis by BVM was regulated by ROS levels

A, B: After the addition of H₂O₂, the expression levels of NLRP3, GSDMD-N, and caspase-1 p20 protein were analyzed by WB; C, D: After the addition of NAC, the expression levels of NLRP3, GSDMD-N, and caspase-1 p20 protein were analyzed by WB; *** P < 0.001, **** P < 0.0001 vs NC group; # P < 0.05, ## P < 0.01, ### P < 0.001, #### P < 0.0001 vs TNF- α /IFN- γ + BVM group.

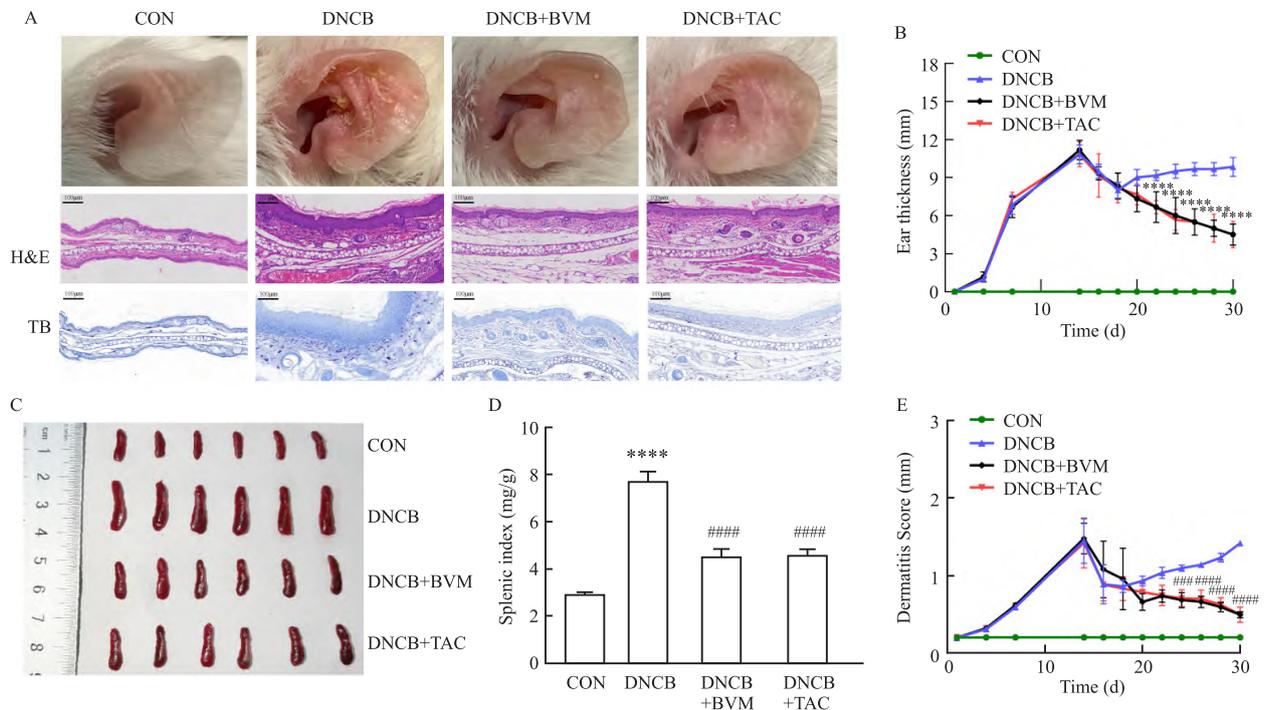


图5 BVM改善特异性皮炎小鼠症状

Fig.5 Benvitimid improved symptoms in atopic dermatitis mice

A: Morphological and histological changes in mice in each group, HE staining $\times 20$, TB staining $\times 20$; B: Ear tissue thickness; C: Spleen size; D: Spleen index; E: Dermatitis score; **** P < 0.0001 vs CON group; ### P < 0.001, #### P < 0.0001 vs DNCB group.

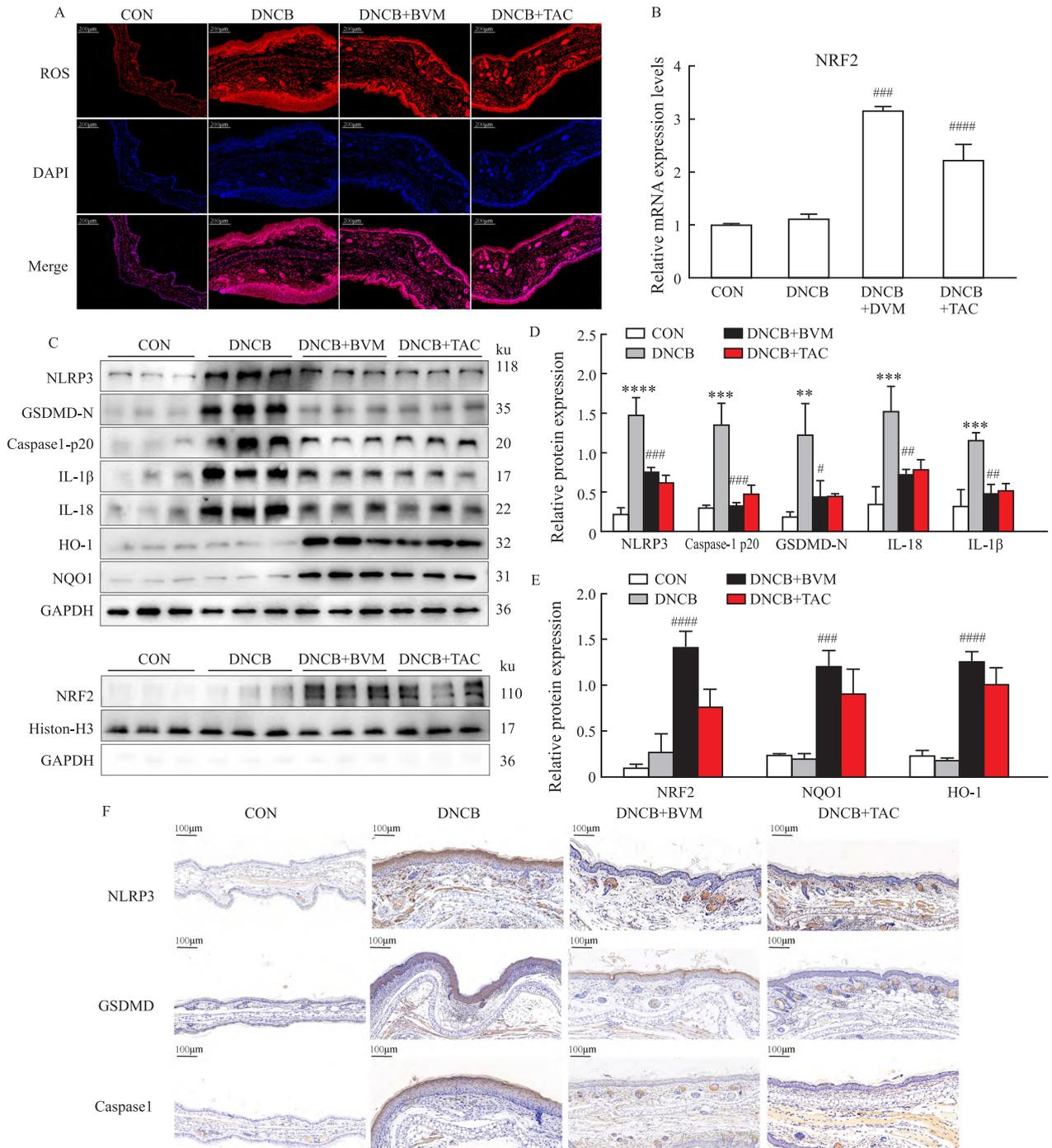


图6 BVM改善特异性皮炎小鼠ROS水平及焦亡程度

Fig.6 BVM improved the ROS levels and pyroptosis in atopic dermatitis mice

A: ROS fluorescence staining of mouse ear tissues $\times 10$; B: mRNA expression level of NRF2 in mouse ear tissues of each group; C, D, E: WB analysis was used to measure the expression levels NLRP3, GSDMD-N, Caspase-1 p20, IL-18, IL-1 β and NRF2, HO-1, NQO1 protein expression levels in each group; F: IHC detection of NLRP3, GSDMD, Caspase-1 expression levels in mouse ear tissues $\times 20$; * $P < 0.01$, ** $P < 0.001$, *** $P < 0.0001$ vs CON group; # $P < 0.05$, ## $P < 0.01$, ### $P < 0.001$, #### $P < 0.0001$ vs DNCB group.

亡,从而加重AD^[10]。ROS的产生是NLRP3炎症小体激活的常见上游机制,抑制细胞中ROS水平可以抑制NLRP3炎症小体的激活^[11]。

三期临床实验^[12]显示BVM能有效治疗AD。

BVM可激活NRF2抑制ROS水平,但对于焦亡的影响并无研究。本研究显示,BVM的治疗减轻了AD小鼠的皮炎症状和组织病理学损伤,BVM可通过抑制焦亡通路治疗AD。通过DNCB诱导小鼠,TNF- α

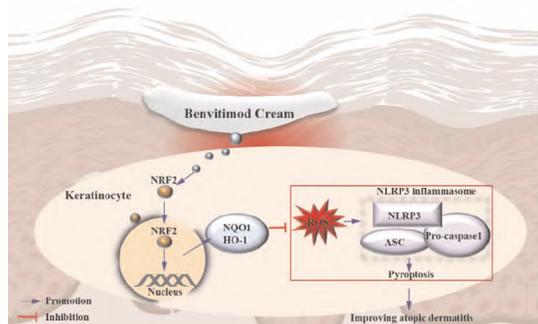


图7 BVM 调控 NRF2/ROS/NLRP3 通路减轻特应性皮炎机制图
Fig. 7 The schematic drawing of the mechanisms of Beenvitimid attenuating atopic dermatitis by modulating the NRF2/ROS/NLRP3 pathway (Created with bioRender.com, with permission)

和 IFN- γ 诱导 HaCaT 细胞,建立 AD 小鼠模型和细胞模型,再使用 BVM 治疗。本实验再现了 AD 小鼠皮肤典型的病理变化,并且有效降低了细胞以及 AD 模型小鼠的焦亡程度。

BVM 能激活 NRF2 抗氧化信号通路,该通路在细胞对 ROS 和亲电应激的防御中起重要作用^[13]。在 TNF- α 和 IFN- γ 诱导的 HaCaT 炎症细胞中,BVM 通过上调 NRF2 抗氧化应激通路,减少 ROS 的产生来抑制 NLRP3、GSDMD、Caspase-1 的激活及 IL-1 β 、IL-18 炎症因子的释放。ROS 激活剂 H₂O₂ 增强 ROS 对 NLRP3 炎性小体的促进作用,ROS 抑制剂 NAC 则逆转此作用,这表明 ROS 对 NLRP3 炎症小体的激活有直接作用。动物实验中,BVM 减轻 DNCB 诱导的 AD 模型小鼠的皮炎症状,抑制小鼠体内 ROS 及焦亡相关蛋白的产生。此外,BVM 减少脾脏指数提示其可能系统性地抑制 Th2 细胞活化,但本研究未检测 Th2 相关细胞因子(如 IL-4、IL-13),需后续实验补充。以上观察结果支持了 BVM 能通过调节 NRF2/ROS/NLRP3 轴减少 AD 中的炎症损伤和焦亡的结论。此研究为抑制焦亡途径可治疗 AD 这一结论提供了进一步的支持。

本研究有一定的局限性。一是研究范围仅限于焦亡经典途径在 AD 中的作用,并且未检测焦亡特征性事件(如细胞膜孔形成或 ATP 释放);二是需通过 NRF2 基因敲除动物模型或 ChIP 实验验证其直接靶点。

综上所述,BVM 通过上调 NRF2 的表达激活抗氧化应激通路,减少 ROS 的产生,有效抑制 AD 中的焦亡程度,从而起到治疗 AD 的作用,揭示了 BVM 可通过 NRF2/ROS/NLRP3 轴减轻 AD 中的炎

症损伤和焦亡。

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ured by CBA method. Hematoxylin and eosin (HE) staining was employed to examine pathological alterations in the spleen. The expression of PU.1 and IL-9 in spleen tissue was detected using immunohistochemistry. Additionally, the expression level of PU.1 protein in the spleen tissue was ascertained through Western blot analysis. **Results** The administration of DB2313 significantly ameliorated spleen lesions in MRL/lpr mice and decreased the levels of anti-ds-DNA, ANA, TNF- α , IL-6, and IFN- γ . It also reduced the proportion of total T cells, TFH cells, Th17 cells, and Th9 cells in the mouse spleen, while increasing the proportion of Treg cells. Furthermore, it lowered the level of PU.1 protein in the spleen. Immunohistochemistry results demonstrated that DB2313 treatment significantly diminished the expression of PU.1 and IL-9 in spleen tissue. **Conclusion** The PU.1 inhibitor DB2313 can improve spleen lesions in MRL/lpr mice and slow the progression of the disease, and its mechanism is related to the regulation of immune cell functions.

Key words systemic lupus erythematosus; PU.1 inhibitor; TACI-Ig; PU.1; MRL/lpr mice; immune function

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Benvitimod attenuates atopic dermatitis by regulating the NRF2/ROS/NLRP3 signaling pathway

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Abstract Objective To investigate the mechanism of action of benvitimod (BVM) in the treatment of atopic dermatitis (AD). **Methods** HaCaT cells were stimulated by TNF- α and IFN- γ , and the cells were grouped into NC group, TNF- α /IFN- γ group, TNF- α /IFN- γ BVM group, and TNF- α /IFN- γ BVM ML385 group. The AD model of DNCB-induced Balb/c mice was divided into CON group, DNCB group, DNCB + BVM group, and DNCB + TAC group. The efficacy of BVM and its roles in antioxidant and pyroptosis regulation were evaluated. **Results** Compared with the control group, BVM inhibited the inflammatory response of HaCaT cells stimulated by TNF- α and IFN- γ , and ameliorated the skin lesions and inflammation in the DNCB-induced AD mouse model; at the same time, it significantly increased the expression of nuclearfactorerythroid-2-relatedfactor2 (NRF2)-related anti-oxidative stress proteins, and significantly reduced the expression of cellular reactive oxygen species (ROS) levels and pyroptosis proteins. At the same time, the levels of NRF2-related anti-oxidative stress proteins significantly increased, and the levels of ROS and pyroptosis proteins significantly decreased. **Conclusion** BVM activates the NRF2/ROS/NLRP3 pathway to inhibit pyroptosis, thereby reducing the inflammatory response in AD.

Key words benvitimod; atopic dermatitis; pyroptosis; NLRP3; NRF2; ROS

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