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组蛋白脱乙酰酶3抑制通过调控Th17促进小鼠银屑病的发展*

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摘要 目的 探讨组蛋白脱乙酰酶3（histone deacetylase 3，HDAC3）对小鼠银屑病样炎症发生发展的影响及相关免疫机制。**方法** 选取6~8周龄健康C57BL/6小鼠，将小鼠随机分为对照组（Control），银屑病模型组（IMQ），HDAC3抑制剂RGFP966处理的银屑病模型组（IMQ+RGFP966），提前1 d对小鼠进行剃毛处理。待稳定1 d后，Control组涂抹等量的凡士林，IMQ组背部涂抹咪喹莫特（imiquimod，IMQ，62.5 mg/d），建立小鼠银屑病样炎症模型；IMQ+RGFP966组在银屑病模型基础上以高剂量HDAC3选择性抑制剂RGFP966（30 mg/kg）进行干预处理。各组持续处理5 d，观察记录背部皮肤银屑病样炎症症状（鳞屑、红斑、皮肤厚度）、体重变化及精神状态，并拍照存档。在小鼠解剖后，采用苏木精-伊红（hematoxylin-eosin，HE）染色检测RGFP966对银屑病模型小鼠皮肤组织层次结构的影响，并测量皮肤厚度。通过实时荧光定量PCR（reverse transcription real-time quantitative polymerase chain reaction，RT-qPCR）和蛋白质印迹法（Western blot，WB），分别检测皮肤组织中HDAC3的mRNA和蛋白质表达水平。采用流式细胞术检测各组小鼠外周血和淋巴结中性粒细胞、外周血$CD4^+$ T淋巴细胞和$CD8^+$ T淋巴细胞及外周血$CD4^+$ T淋巴细胞的白介素-17A（interleukin-17A，IL-17A）分泌水平，检测脾脏$CD4^+$ T淋巴细胞HDAC3、CC基序趋化因子受体（CC motif chemokine receptor，CCR）6、CCR8表达及IL-17A分泌水平。采用免疫组化检测皮肤HDAC3、IL-17A、白介素-10（interleukin-10，IL-10）水平。**结果** 与Control组相比，IMQ组小鼠展示出明显的银屑病样炎症，出现红斑、鳞屑及皮肤褶皱。RGFP966加重了银屑病样炎症症状，皮肤角化增多。银屑病面积与严重性指数（psoriasis area and severity index，PASI）皮肤症状评分，IMQ组高于Control，IMQ+RGFP966组高于IMQ组。测量各组皮肤厚度，IMQ+RGFP966>IMQ>Control。Control、IMQ、IMQ+RGFP966组的血液和淋巴结的中性粒细胞依次增多，血液$CD4^+$ T淋巴细胞和$CD8^+$ T淋巴细胞也呈相同趋势。在皮肤组织中，与Control组相比，模型组HDAC3的mRNA和蛋白质水平下降，RGFP966未能使HDAC3的mRNA和蛋白质表达进一步下降。HDAC3主要定位于细胞核内，与Control组相比，IMQ组的皮肤组织核内HDAC3减少，RGFP966使核内HDAC3进一步减少。与Control组和IMQ组相比，RGFP966处理使脾脏$CD4^+$和$CD8^+$ T细胞HDAC3表达下降。RGFP966处理增加了脾脏$CD4^+$ T细胞CCR6和CCR8的表达；同时，外周血和脾脏$CD4^+$ T淋巴细胞的IL-17A分泌显著升高。此外，与IMQ组相比，RGFP966减少了皮肤组织IL-10蛋白并上调了IL-17A表达。**结论** RGFP966通过抑制HDAC3，增加细胞因子IL-17A分泌和趋化因子CCR8、CCR6的表达，加重银屑病样炎症反应。

关键词 组蛋白去乙酰化酶3，IL-17，银屑病，CCR8
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银屑病是一种以特征性红斑鳞屑为主要表现的慢性炎症性皮肤病，影响全球约3%的人口[1]。虽然银屑病的病因至今尚不完全明确，但目前认为，遗传因素与环境因素（诸如创伤、感染、精神压力等）相互作用导致的免疫系统功能紊乱是其发生、发展的主要因素[2]。其中，异常分化和迁移的辅助性T细胞1（T helper cell，Th1）与Th17细胞亚群通过分泌肿瘤坏死因子α（tumor necrosis factor-α，TNF-α）、白介素（interleukin，IL）-17、IL-21、IL-22等因子，在促进银屑病皮肤炎症及角质形成

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细胞过度增殖方面发挥重要作用[3]。组蛋白脱乙酰酶3（histone deacetylase 3，HDAC3）属于一种表观遗传调控蛋白，主要通过去除染色质组蛋白带负电荷的乙酰基团抑制基因表达，进而影响多种组织细胞的发育分化[4]。在$CD4^+$ T细胞中，HDAC3敲除与Th17细胞特异转录因子RORγt的表达增加和IL-17的产生密切相关[5-6]。鉴于IL-17在银屑病发病机制中的核心地位，本研究进一步探讨了HDAC3对小鼠银屑病发生、发展的影响及其免疫学机制。

1　材料与方法

1.1　小鼠

6~8周龄的雌性C57BL/6小鼠购自北京华阜康生物科技股份有限公司。小鼠在华北理工大学SPF级动物房饲养、繁殖，所有实验操作按华北理工大学实验动物管理委员会规定进行，伦理审查编号为SQ2022009。

1.2　咪喹莫特（imiquimod，IMQ）诱导银屑病模型

选取6~8周龄的雌性C57BL/6小鼠，随机分为对照组（Control）、银屑病模型组（IMQ）以及RGFP966处理组（IMQ+RGFP966），每组5只。小鼠均进行背部剃毛及脱毛处理，裸露面积约为2 cm×3 cm。对照组涂抹凡士林，银屑病模型组及RGFP966处理组均涂抹咪喹莫特（明欣利迪，中国），62.5 mg/d，连续5 d。处理组注射RGFP966（Ambeed，美国），每只30 mg/kg，模型组注射等量的二甲基亚砜（DMSO）。

1.3　小鼠银屑病评分

自模型建立起，对小鼠背部皮肤病损变化进行拍照记录，按银屑病面积与严重性指数（psoriasis area and severity index，PASI）评分，对小鼠背部皮肤的鳞屑、红斑、皮肤厚度并辅以体重变化及精神状态（表1），进行评分，总评分为3项评分之和。

Table 1　Psoriasis area and severity index standard

Symptom score	0	1	2	3	4
Thickness	Smooth skin with no wrinkles	Slight wrinkles appearing at the edges of the treated area	Slight wrinkles appearing across the entire treated area	Further deepening of wrinkles in the treated area	In addition to a score of 3, the mouse exhibits symptoms such as weight loss or poor condition
Scale	Smooth skin with no scales	Slight scaling appears on the skin of the treated area	The entire skin of the treated area is covered with scales	Further intensification of scaling in the treated area	In addition to a score of 3, the mouse exhibits symptoms such as weight loss or poor condition
Erythema	Smooth skin	Slight redness appears on the skin of the treated area	The entire skin of the treated area turns red	Further intensification of redness in the treated area	In addition to a score of 3, the mouse exhibits symptoms such as weight loss or poor condition

1.4　流式细胞术

分离小鼠外周血、脾脏、淋巴结，外周血使用纯水裂解红细胞8~10 s，脾脏及淋巴结经过研磨后使用氯化铵裂解红细胞，三者均使用含0.5%牛血清白蛋白（BSA）的PBS悬液清洗两次，制备成单细胞悬液。为检测细胞表面标志，采用流式抗体进行细胞表面染色（4℃，避光孵育30 min），染色后使用含0.5% BSA的PBS悬液清洗两次。为检测细胞因子的表达，将细胞悬液（2×10^6/孔）置于5% CO_2、37℃培养箱中，加入2 μl Leukocyte activation cocktail, with BD GolgiPlug™（BD Biosciences），在体外充分刺激4 h后，离心收集细胞，先加入1 μl CD4流式抗体4℃孵育30 min后加入1 ml染色缓冲液清洗，再加入100 μl Cytofix/Cytoperm固定/渗透溶液（BD Biosciences）固定和渗透细胞20 min后加入1 ul IL-17A流式抗体，孵育30 min后加入4%多聚甲醛固定。为检测不同组小鼠脾脏T淋巴细胞内HDAC3表达，先加入1 μl CD4和CD8抗体，4℃孵育30 min后加入1 ml流式缓冲液，离心清洗后Cytofix/Cytoperm固定/渗透溶液试剂盒固定和渗透细胞后，流式缓冲液清洗2次，加入1 μl HDAC3抗体，4℃孵育30 min，再度清洗后加入1 μl羊抗兔荧光标记抗体4℃孵育30 min。采用的流式抗体有：Anti-mouse-CD4-PE antibody（BD Biosciences）、Anti-mouse-CD8-PERCP antibody（BD Biosciences）、Anti-mouse-CCR8-PE antibody（Biolegend，美国）、Anti-mouse-CCR6-APC antibody（Biolegend）、Anti-mouse-IL-17A-FITC antibody（Biolegend）、Anti-mouse-CD11b-PERCP antibody（BD Biosciences）、Anti-mouse-ly6g-APC antibody（Invitrogen）、Anti-mouse-HDAC3 antibody（万类，中国）、Goat anti-

rabbit-IgG-FITC antibody（瑞帕特，中国）。采用贝克曼流式细胞仪收集标本，并用Flowjo v10.8进行数据分析。

1.5 实时荧光定量PCR（RT-qPCR）

称取50 mg皮肤组织，剪碎并研磨，使用TRIzol试剂法提取皮肤组织总RNA，并用NanoDrop 2000（Thermo Fisher Scientific，美国）检测RNA浓度及纯度。按照逆转录试剂盒说明书（聚合美，中国），取1 μg RNA逆转录为cDNA，取2 μl cDNA按照QPCR试剂盒（聚合美，中国）说明书构建20 μl PCR扩增体系：DNA 2 μl，SYBR预混液10 μl，引物各0.4 μl，双蒸水7.2 μl。通过SLAN-96S荧光定量PCR仪（宏石，中国）检测荧光信号，以$2^{-\Delta\Delta Ct}$法分析各组目的基因mRNA相对表达水平（表2）。PCR扩增程序：95℃ 5 min；95℃ 10 s，60℃ 30 s，40个循环；50℃ 30 s。

Table 2 Primer sequences

Gene	Sequence (5'-3')
β-actin forward	CACTGTCGAGTCGCGTCC
β-actin reverse	TCATCCATGGCGAACTGGTG
HDAC3 forward	CCCTACTACAGTTCTTGCTTCC
HDAC3 reverse	CCTTGCCACTGTACTTCATCT

1.6 蛋白质印迹法（Western blot）

取20 mg皮肤组织，按照组织：RIPA裂解液：蛋白酶抑制剂=1 mg：10 μl：0.1 μl的比例加入RIPA裂解液（新赛美，中国）和蛋白酶抑制剂，将皮肤组织剪碎研磨，冰上裂解30 min，12 000 r/min离心10 min，取上清。使用BCA法测定蛋白质浓度，加入上样缓冲液，混合均匀煮沸10 min。取总蛋白质20 μg/孔，进行聚丙烯酰胺凝胶电泳（80 V电泳30 min，随后转为120 V电泳50 min）。电泳完成后，将蛋白质电转至聚偏二氟乙烯膜（polyvinylidene fluoride，PVDF）。PVDF用5%脱脂奶粉在摇床上以50 r/min轻摇2 h。将PVDF膜置于一抗用抗体稀释液（雅酶，中国）配制：GAPDH（ET1601-4，华安，中国；1∶10 000）、HDAC3（7G6C5，Cell Signaling Technology，美国；1∶1 000），4℃孵育过夜，用含Tween 20的Tris缓冲盐水（Tris buffered saline with Tween-20，TBST）清洗PVDF膜，并置于以1∶10 000比例稀释的辣根过氧化物酶标记的羊抗兔、羊抗鼠二抗（瑞帕特）中孵育2 h，清洗并进行显影。用Image Lab处理条带，测定蛋白质相对表达量。

1.7 苏木精-伊红（hematoxylin-eosin，HE）染色及免疫组化

解剖小鼠时，取小鼠皮肤组织浸泡于4%多聚甲醛溶液中过夜，流水冲洗多聚甲醛后使用脱水机进行脱水。用石蜡包埋组织并进行切片，对切片组织进行苏木精-伊红（hematoxylin-eosin，HE）染色，最后用中性树胶封片。免疫组化中，切片经柠檬酸钠进行抗原修复，并封闭内源性过氧化物酶和非特异性结合位点，向切片滴加一抗覆盖：HDAC3（1∶100，万类，中国）、IL-17A（1∶100，万类，中国）、IL-10（1∶50，万类，中国），于湿盒中4℃孵育过夜，清洗并在二抗（酶标羊抗小鼠/兔IgG聚合物；中杉金桥，中国）中37℃孵育1 h，滴加DAB染液至出现棕黄色染色信号，苏木染液染细胞核，用中性树胶封片。HE切片和免疫组化切片置于BX53全自动显微镜（Olympus，日本）上观察。皮肤厚度使用imageproplus6.0计算。

1.8 统计分析

数据分析采用GraphPad Prism 8、SPSS 27.0统计软件，以$P<0.05$为统计学显著性差异标准。统计图表采用GraphPad Prism 8绘制。

2 结　果

2.1 RGFP9666可增强咪喹莫特诱导的银屑病样皮肤炎症

相较于对照组，咪喹莫特处理显著加剧了模型组小鼠背部皮肤的红斑程度、鳞屑分布以及皮肤皱褶，表明小鼠银屑病模型的成功建立，且在第5天呈现出最为显著的病征表现。与银屑病模型组相比，RGFP966处理的银屑病小鼠皮肤红斑面积扩大、脱屑及皮肤皱褶现象也更为严重（图1a）。各组小鼠皮肤HE染色结果显示：对照组小鼠皮肤呈现出典型的薄表皮层与清晰的层次结构；模型组的皮肤则明显出现了角化过度及表皮增厚现象，而与银屑病模型组相比，RGFP966处理组小鼠皮肤角化过度及表皮增厚现象更加显著（图1b，c）；从咪喹莫特处理第2天开始，RGFP966处理组小鼠的银屑病皮损面积和严重程度指数（PASI）也显著高于对照组和银屑病模型组（图1d）。此外，与对照组和银屑病模型组相比，RGFP966处理组小鼠外周血、淋巴结中性粒细胞及$CD4^+$和$CD8^+$ T细胞所占比例也显著增加（图1e~g）。以上研究结果表明，HDAC3抑制剂处理可增强银屑病样皮肤症状和全身炎症反应。

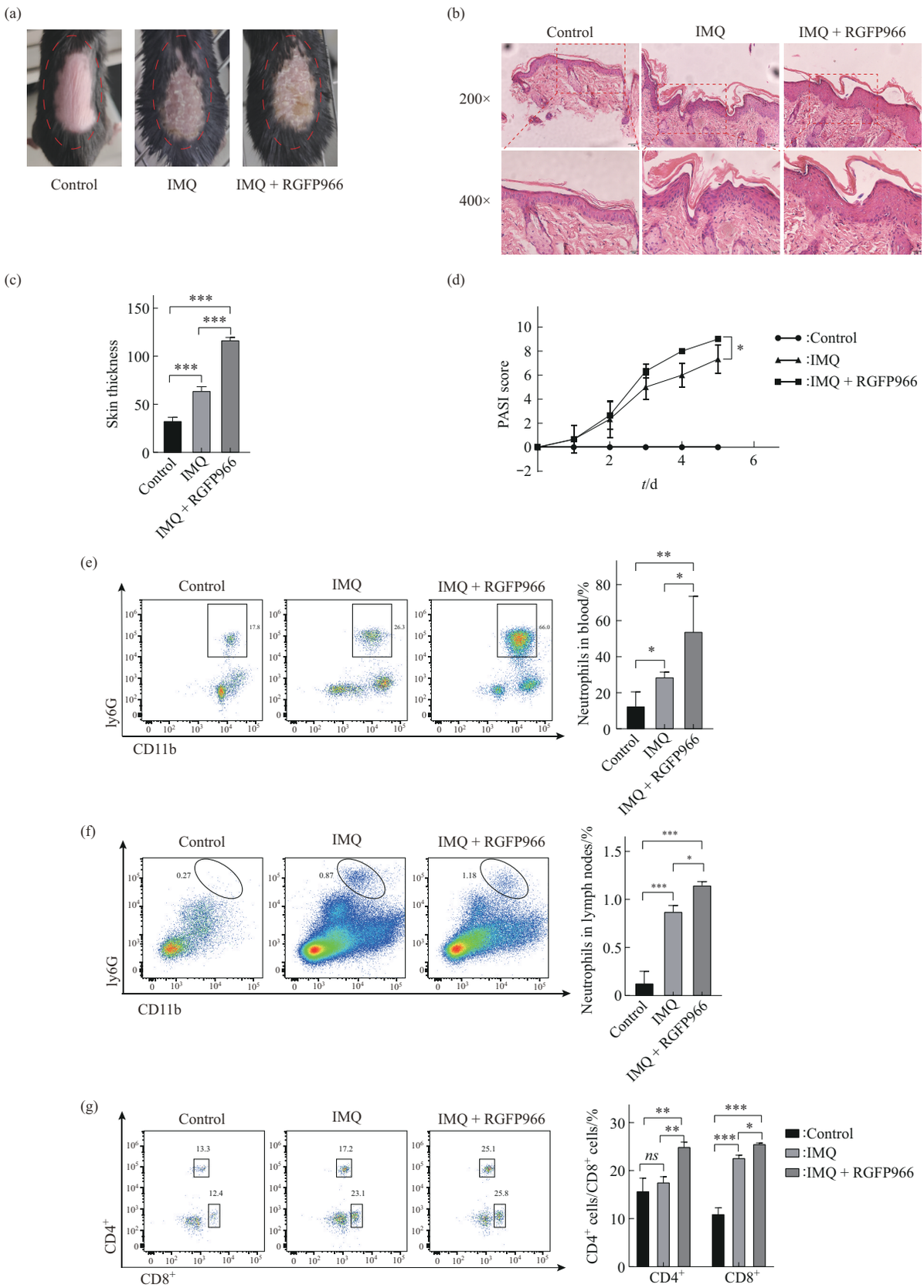

Fig. 1　Enhancement of imiquimod−induced psoriatic skin inflammation by RGFP9666

(a) Symptoms of skin on the back of mice. (b) Histopathological changes observed by HE staining. (c) Skin thickness measured by HE staining. (d) Daily PASI score for the symptoms of psoriasis. (e, f) The proportion of neutrophils in blood (e) and lymph node (f). (g) The proportion of $CD4^+$ and $CD8^+$ T cells in blood. *ns*, no significance, **P*<0.05, ***P*<0.01, ****P*<0.001.

2.2 RGFP966抑制脾脏和皮肤的HDAC3表达

RT-qPCR结果表明，相较于对照组，小鼠皮肤模型组中HDAC3的mRNA表达水平显著降低，而在这一基础上，RGFP966的引入并未进一步改变皮肤的HDAC3表达（图2a）。Western blot结果显示，银屑病模型有效降低了小鼠皮肤中HDAC3蛋白的表达水平，而RGFP966的抑制也未有统计学意义（图2b）。免疫组化结果显示，HDAC3主要定位于细胞核内，且在模型组小鼠的细胞核和细胞质内，HDAC3的表达相较于对照组有显著降低，而RGFP966的干预则进一步降低了细胞核内HDAC3的表达水平（图2c）。已有研究和本研究都发现，咪喹莫特诱导的模型小鼠不仅皮肤完整性受损，还常伴有脾肿大现象。而在这种情况下，RGFP966的干预能够显著降低脾脏中$CD4^+$和$CD8^+$T淋巴细胞内HDAC3的表达比例和表达水平，这一发现提示，RGFP966可能通过影响免疫细胞的活化与分化过程，进而加剧银屑病样病变的发展（图2d，e）。

2.3 RGFP966通过抑制HDAC3上调CC基序趋化因子受体8（CC motif chemokine receptor 8，CCR8）、CCR6、IL-17A的表达

实验结果显示，与模型组小鼠相比，接受RGFP966干预处理的小鼠脾脏$CD4^+$T淋巴细胞中，$CCR8^+$细胞、$CCR6^+$细胞以及Th17细胞的比例均呈现出显著的上升趋势，外周血Th17细胞也呈上升的趋势（图3a~d）。本研究还通过免疫组化技术对皮肤组织进行分析，结果显示，IL-17A与IL-10在皮肤组织细胞中呈现弥漫性分布，主要定位于细胞质内，RGFP966的处理显著地上调了IL-17A的表达水平，而相应地，IL-10的表达水平则出现了下调（图3e，f），这一发现揭示了RGFP966可能对皮肤局部免疫微环境的影响。

3 讨 论

近年来，有关HDAC3在银屑病发生与发展中的作用已开展了一些研究。Thatikonda团队[7]与Qian等[8]的研究发现，荜拔酰胺（piperlongumine，PPL）及酸奶可通过遏制巨噬细胞HDAC3的核转位，缓解银屑病样皮肤炎症；Choudhary等[9-10]发现抑制HDAC3可增加角质形成细胞水通道蛋白3（aquaporin 3，AQP3）的表达，提示抑制HDAC3可用于AQP3表达降低相关疾病如银屑病的治疗。然而，这些研究在揭示HDAC3在银屑病中的作用时仍存在一定的局限性，特别是缺乏在动物模型中特异性抑制HDAC3的直接证据，且主要聚焦于巨噬细胞，忽视了银屑病中更为关键的Th17细胞的作用。因此，在动物实验中特异性地抑制HDAC3，以验证其在银屑病病理进程中的确切作用，显得尤为重要。

值得注意的是，先前的研究已表明，HDAC3的缺失会扰乱Th17细胞与Treg细胞之间的平衡，促使初始T细胞向Th17细胞分化，进而引发强烈的自身免疫反应与炎症[5]。同时，课题组的前期工作也发现，高剂量RGFP966能够提高外周T细胞中Th17细胞的比例[11]。基于这些发现，本研究在制备小鼠银屑病模型的同时采用高剂量HDAC3特异抑制剂RGFP966处理，以观察其对疾病进展的影响。研究结果显示，RGFP966处理显著加剧了银屑病模型小鼠的皮肤炎症反应，加速了病情的发生和发展。这一现象与IL-23-IL-17A轴的激活密切相关，HDAC3抑制导致$CD4^+$ T细胞中Th17细胞比例上升，进而提高了皮肤组织中IL-17A的表达水平，加速了银屑病的进展。

CCR6作为一种在淋巴及非淋巴组织中均有表达的跨膜G蛋白偶联受体，其异常表达与多种炎症性疾病紧密相关，包括银屑病、多发性硬化症和类风湿性关节炎等[12-14]。CCR6被其特异性配体趋化因子配体20（C-C motif chemokine ligand 20，CCL20）激活，促进CCR6淋巴细胞迁移到炎症部位[15-18]。CCL20和CCR6的表达在银屑病等慢性炎症患者的上皮组织中上调[19]。CCR6在Th17细胞中的高表达使其成为Th17细胞迁移的重要驱动力，并与CD4共同作为Th17细胞的主要表面标记[20-22]。本研究进一步证实，HDAC3抑制显著增加了T淋巴细胞中$CCR6^+$T细胞的比例，与IL-17A水平的升高一致，进一步证实了HDAC3在调控Th17细胞分化与功能中的重要作用。

CCR8作为另一种跨膜G蛋白偶联受体，其表达广泛分布于不同效应性T淋巴细胞亚群及皮肤驻留的T细胞中，是T细胞归巢至皮肤的重要分子标记，主要配体为CCL1[23-26]。趋化因子CCR8已在皮肤病如特应性皮炎、银屑病中得到了一定的研究[27-28]。本研究对CCR8表达的检测发现：HDAC3抑制后脾脏T细胞中CCR8的表达明显增加。这一结果提示，HDAC3抑制可能通过上调CCR8的表达，调控外周T淋巴细胞的皮肤归巢，进而促进银屑病的发生与发展。

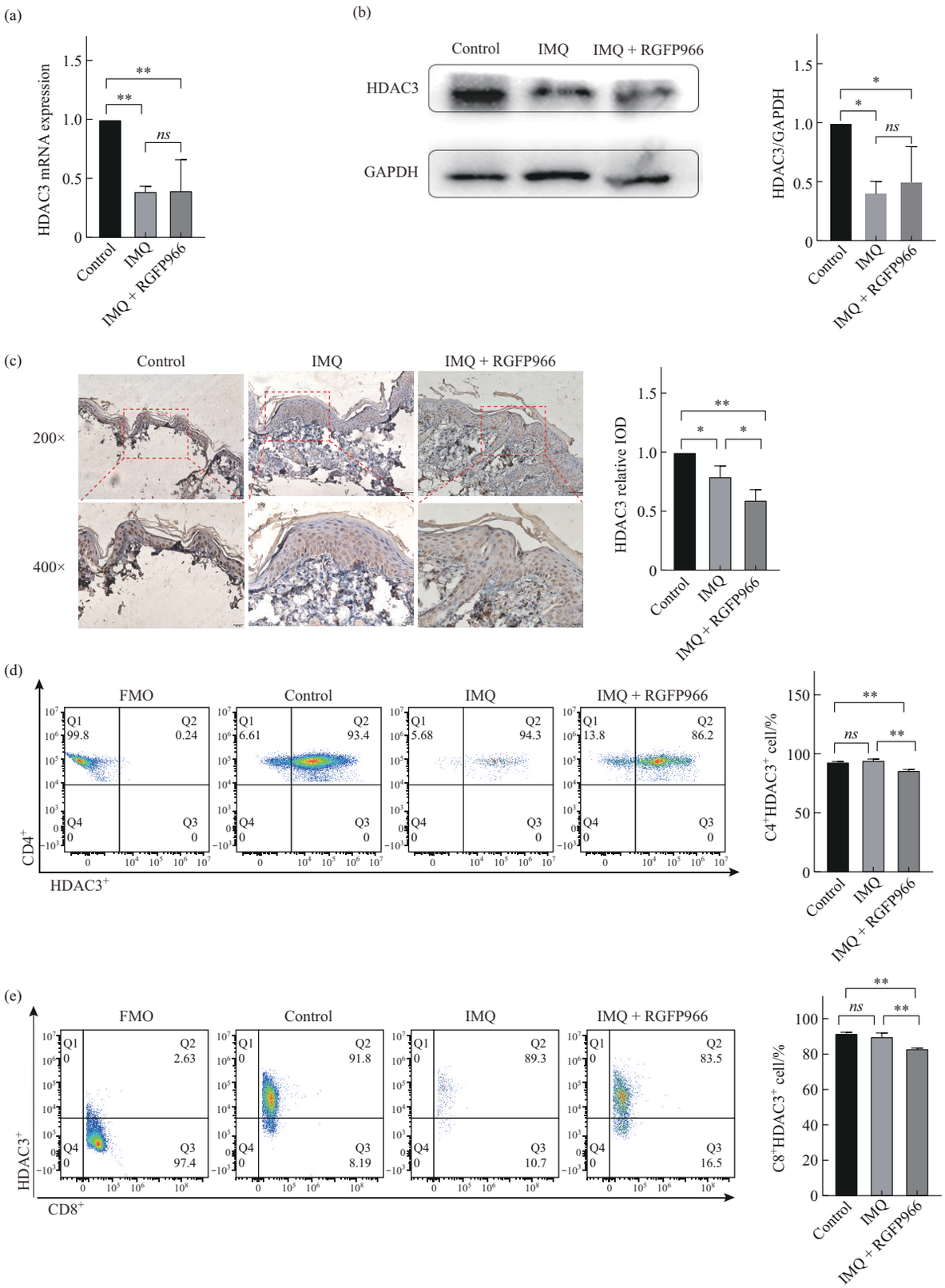

Fig. 2　Inhibition of HDAC3 expression in mice by RGFP966

(a) HDAC3 mRNA expression in mice skin. (b) HDAC3 protein expression in mice skin. (c) HDAC3 expression in mouse skin detected by immunohistochemistry. (d, e) The proportion of HDAC3 positive cell in $CD4^+$ (d) and $CD8^+$ (e) T cells in spleen. *ns*, no significance, **P*<0.05, ***P*<0.01. FMO: fluorescence minus one. IOD: integrated optiondensity.

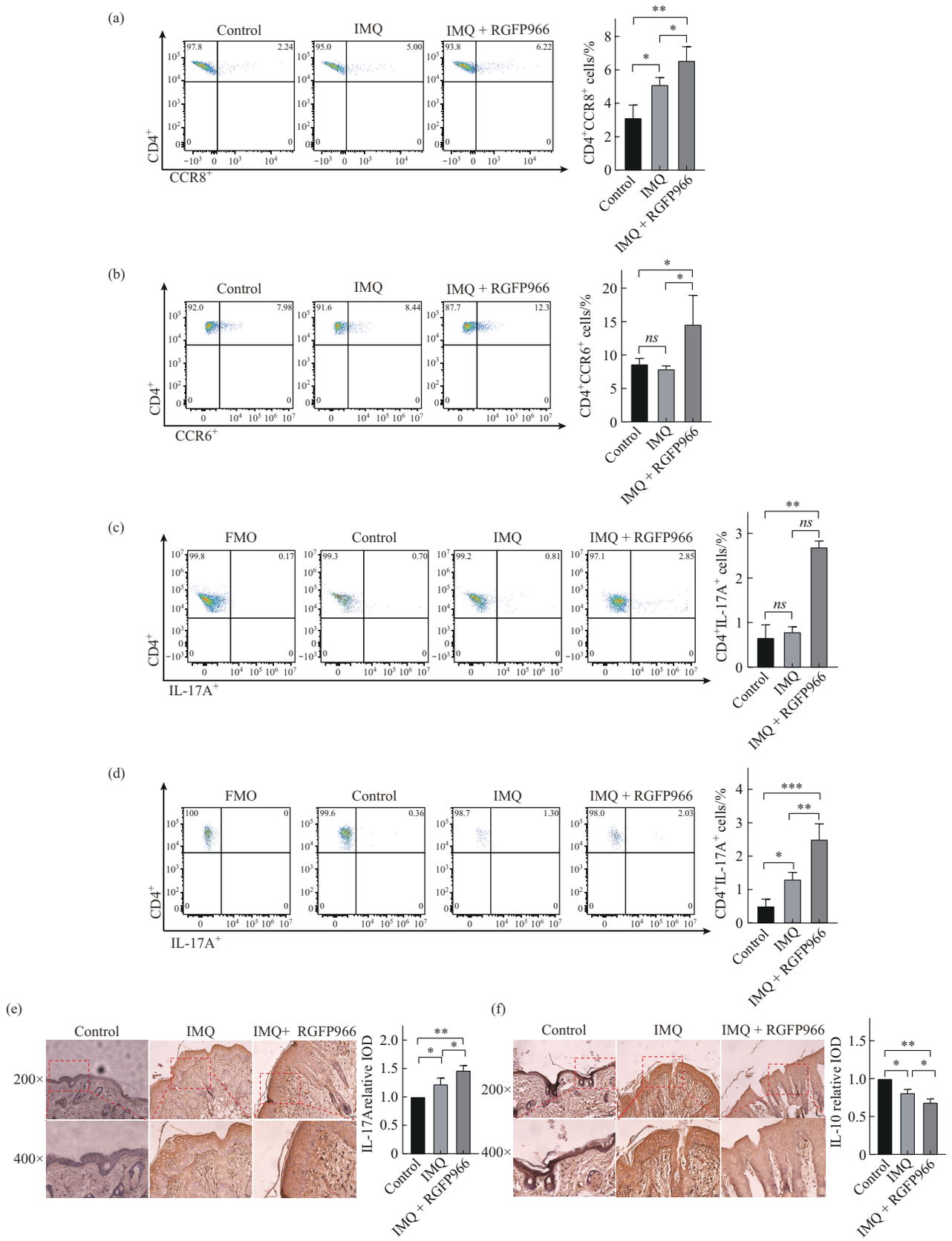

Fig. 3　Changes in related immune molecules caused by the effect of RGFP966

(a−c) The proportion of $CCR8^+$ (a), $CCR6^+$ (b), and $IL\text{-}17A^+$ (c) cells in $CD4^+$ T cells in spleen. (d) The proportion of $IL\text{-}17A^+$ cells in $CD4^+$ T cells in blood. (e, f) IL-17A (e) and IL-10 (f) expression in mouse skin detected by immunohistochemistry. *ns*, no significance, **P*<0.05, ***P*<0.01, ****P*<0.001. FMO: fluorescence minus one. IOD: integrated optiondensity.

因此，HDAC3抑制可能通过调控T淋巴细胞分化和迁移，特别是增强Th17细胞的数量与功能，以及增加CCR6与CCR8的表达，来加重银屑病的病理进程。然而，HDAC3对T淋巴细胞中CCR6、CCR8和IL-17A的调控机制尚不清楚，需要在后续实验中进一步研究。

4 结　论

本研究发现HDAC3的抑制显著上调了IL-17A和CCR6、CCR8的表达水平，这两个分子在调控T细胞分化与迁移过程中扮演着至关重要的角色。通过这一调控机制，HDAC3的抑制不仅促进了Th17细胞的分化与增殖，还增强了T细胞向皮肤组织的迁移能力，从而加剧了银屑病的病理进程。研究结果为深入理解银屑病的免疫病理机制及寻找银屑病的潜在治疗靶点提供了新的线索。

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Inhibition of HDAC3 Promotes Psoriasis Development in Mice Through Regulating Th17*

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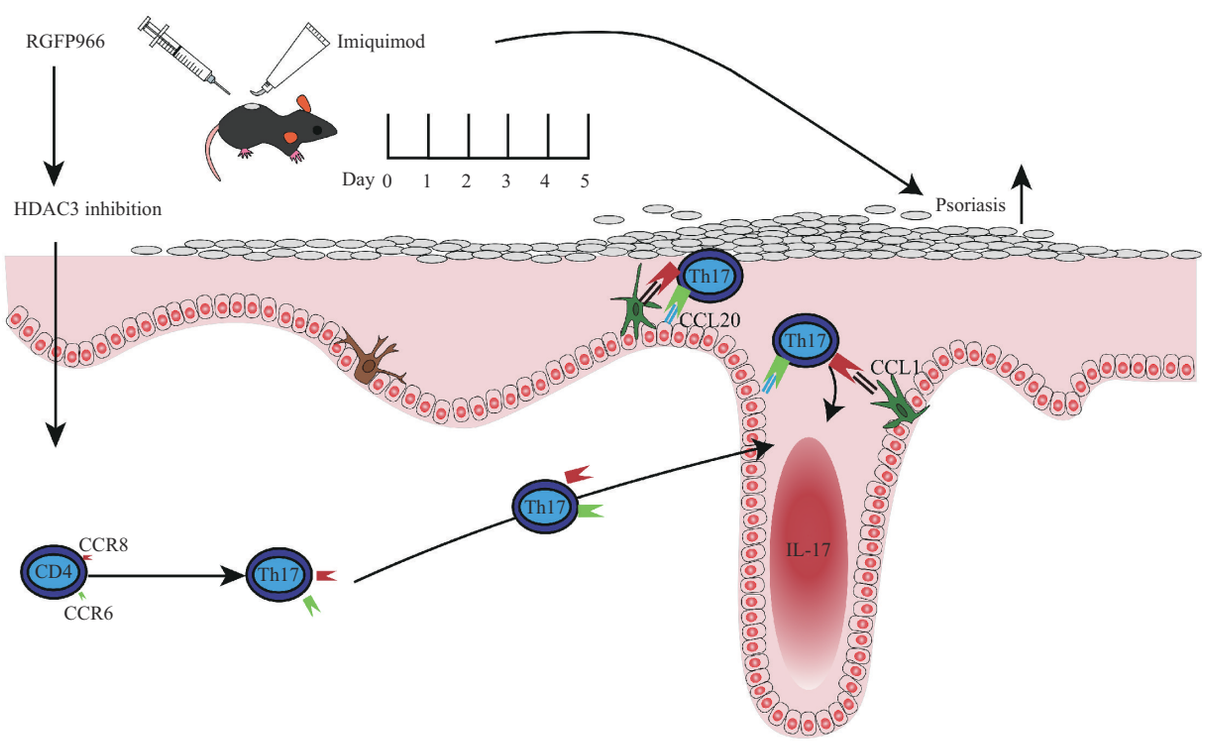
Graphical abstract



Abstract **Objective** To investigate the influence of histone deacetylase 3 (HDAC3) on the occurrence, development of psoriasis-like inflammation in mice, and the relative immune mechanisms. **Methods** Healthy C57BL/6 mice aged 6−8 weeks were selected and randomly divided into 3 groups: control group (Control),

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psoriasis model group (IMQ), and HDAC3 inhibitor RGFP966-treated psoriasis model group (IMQ+RGFP966). One day prior to the experiment, the back hair of the mice was shaved. After a one-day stabilization period, the mice in Control group was treated with an equal amount of vaseline, while the mice in IMQ group was treated with imiquimod (62.5 mg/d) applied topically on the back to establish a psoriasis-like inflammation model. The mice in IMQ+RGFP966 group received intervention with a high dose of the HDAC3-selective inhibitor RGFP966 (30 mg/kg) based on the psoriasis-like model. All groups were treated continuously for 5 d, during which psoriasis-like inflammation symptoms (scaling, erythema, skin thickness), body weight, and mental status were observed and recorded, with photographs taken for documentation. After euthanasia, hematoxylin-eosin (HE) staining was used to assess the effect of RGFP966 on the skin tissue structure of the mice, and skin thickness was measured. The mRNA and protein expression levels of HDAC3 in skin tissues were detected using reverse transcription real-time quantitative polymerase chain reaction (RT-qPCR) and Western blot (WB), respectively. Flow cytometry was employed to analyze neutrophils in peripheral blood and lymph nodes, $CD4^+$ T lymphocytes, $CD8^+$ T lymphocytes in peripheral blood, and IL-17A secretion by peripheral blood $CD4^+$ T lymphocytes. Additionally, spleen $CD4^+$ T lymphocyte expression of HDAC3, CCR6, CCR8, and IL-17A secretion levels were analyzed. Immunohistochemistry was used to detect the localization and expression levels of HDAC3, IL-17A, and IL-10 in skin tissues. **Results** Compared with the Control group, the IMQ group exhibited significant psoriasis-like inflammation, characterized by erythema, scaling, and skin wrinkling. Compared with the IMQ group, RGFP966 exacerbated psoriasis-like inflammatory symptoms, leading to increased hyperkeratosis. The psoriasis area and severity index (PASI) skin symptom scores were higher in the IMQ group than those in the Control group, and the scores were further elevated in the IMQ+RGFP966 group compared to the IMQ group. Skin thickness measurements showed a trend of IMQ+RGFP966>IMQ>Control. The numbers of neutrophils in the blood and lymph nodes increased sequentially in the Control, IMQ, and IMQ+RGFP966 groups, with a similar trend observed for $CD4^+$ and $CD8^+$ T lymphocytes in the blood. In skin tissues, compared with the Control group, the mRNA and protein levels of HDAC3 decreased in the IMQ group, but RGFP966 did not further reduce these expressions. HDAC3 was primarily located in the nucleus. Compared with the Control group, the nuclear HDAC3 content decreased in the skin tissues of the IMQ group, and RGFP966 further reduced nuclear HDAC3. Compared with the Control and IMQ groups, RGFP966 treatment decreased HDAC3 expression in splenic $CD4^+$ and $CD8^+$ T cells. RGFP966 treatment increased the expression of CCR6 and CCR8 in splenic $CD4^+$ T cells and enhanced IL-17A secretion by peripheral blood and splenic $CD4^+$ T lymphocytes. Additionally, compared with the IMQ group, RGFP966 reduced IL-10 protein levels and upregulated IL-17A expression in skin tissues. **Conclusion** RGFP966 exacerbates psoriatic-like inflammatory responses by inhibiting HDAC3, increasing the secretion of the cytokine IL-17A, and upregulating the expression of chemokines CCR8 and CCR6.

Key words histone deacetylase 3, IL-17, psoriasis, CCR8
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