



ORIGINAL PAPER

## Ezetimibe mitigates DNCB-induced mouse model of eczematous dermatitis

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### ABSTRACT

**Introduction and aim.** Atopic dermatitis (AD) is a life-long inflammatory dermatosis that features dry, erythematous skin. Ezetimibe is a lipid-lowering agent with enhanced anti-inflammatory and anti-oxidative capacities. This work attempted to evaluate the anti-eczematous action of topically administered ezetimibe in a mouse prototype of 1-chloro-2,4-dinitrobenzene (DNCB)-evoked AD. To our knowledge, this is the first study to investigate the topical use of ezetimibe in an experimental model of AD.

**Material and methods.** Thirty male Swiss albino mice were randomly allocated into five groups: healthy control, DNCB-induced model, vehicle, tacrolimus (0.1% ointment), and ezetimibe (2% ointment). Treatments were applied daily for 21 days. Clinical severity scores, total and differential leukocyte counts, histopathological changes, immunohistochemical expression of interleukin (IL)-4 and IL-13, and tissue levels of IgE, malondialdehyde (MDA), IL-17, IL-31, transforming growth factor- $\beta$  (TGF- $\beta$ ), and tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) were assessed.

**Results.** DNCB increased dermatitis severity (EASI score  $9.8 \pm 0.7$  vs.  $0.5 \pm 0.1$  in controls,  $p < 0.001$ ), total leukocytes ( $14.2 \pm 1.6 \times 10^3/\text{mL}$  vs.  $3.9 \pm 0.6 \times 10^3/\text{mL}$ ,  $p < 0.001$ ), and IgE ( $356 \pm 42 \text{ ng/mL}$  vs.  $92 \pm 15 \text{ ng/mL}$ ,  $p < 0.001$ ). Ezetimibe treatment significantly reduced EASI scores ( $2.1 \pm 0.4$ ,  $p < 0.01$  vs. DNCB), leukocytes ( $5.9 \pm 0.3 \times 10^3/\text{mL}$ ,  $p < 0.01$  vs. DNCB), IgE ( $128 \pm 18 \text{ ng/mL}$ ,  $p < 0.01$  vs. DNCB), and MDA ( $2.3 \pm 0.4 \text{ } \mu\text{mol/L}$  vs.  $5.9 \pm 0.7 \text{ } \mu\text{mol/L}$  in DNCB,  $p < 0.001$ ). Pro-inflammatory cytokines IL-4, IL-13, IL-17, IL-31, TGF- $\beta$ , and TNF- $\alpha$  were also markedly decreased (all  $p < 0.05$ ), with effects comparable to tacrolimus.

**Conclusion.** Topical ezetimibe significantly alleviated DNCB-induced AD-like lesions by reducing histopathological changes, leukocyte infiltration, IgE, oxidative stress, and key inflammatory cytokines. These findings support ezetimibe as a potential adjunctive topical therapy for immune-mediated dermatoses, warranting future clinical evaluation.

**Keywords.** atopic dermatitis, DNCB, eczema, ezetimibe, inflammatory dermatosis, immune-mediated skin diseases

### Introduction

Atopic dermatitis (AD) is a recurring and remitting auto-inflammatory skin disease evoked by the immune system that often begins during the early stages of life.

It is often known as eczematous dermatitis and is the most common skin condition that involves a rapid development of itchy patches on the skin.<sup>1-3</sup> Dermatoses is marked by atopic inflammation, lichen formation,

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itchiness, greater vulnerability to infection, and rashes, frequently followed by acute pruritus.<sup>1,4,5</sup> Inheritance variables, immune disruption, compromised skin barriers, and external stimuli are believed to be among the fundamental reasons.<sup>6</sup> AD collapses beneath the category of atopic ailments that encompass food intolerance, pulmonary asthma, rashes and hay fever, many of which are relevant pathologies associated with elevated immunoglobulin E (IgE) concentrations and diminished filaggrin concentrations.<sup>7,8</sup> Furthermore, AD can occur concurrently with other immune-driven dermatoses, such as psoriasis.<sup>9-13</sup> Furthermore, a possible correlation between ulcerative colitis and AD is shown in the literature study.<sup>14-17</sup> Meanwhile, an immune system mismatch in the Th1/Th2 reactions is a distinguishing prosperity of AD pathogenesis, with higher rates of released cytokines, including interleukin (IL)-4 and IL-13, which drive Th2 replication and B cell IgE liberation<sup>1</sup>. IgE value is a specified indicator of AD intensity, which is corroborated by the efficiency of IgE-focused therapy to reduce AD symptoms among sufferers.<sup>18-20</sup> Tumor necrosis factor (TNF)- $\alpha$  is a significant inflammatory cytokine that modulates immunological responses and cellular division.<sup>21-23</sup> It also facilitates the relocation of NF- $\kappa$ B into keratinocytes, which assists in transcriptional signaling and the liberation of cytokines, particularly IL-6 and IL-8.<sup>24-28</sup> Moreover, IL-17 is a pivotal target for treatment, since it fosters the secretion of TNF- $\alpha$ , exacerbating the effects of atopic inflammation.<sup>8</sup> IL-17 amplification elevates IL-6 and IL-1 $\beta$  concentrations in keratinocytes, which are associated with the pathogenesis of AD.<sup>2,5</sup> Additionally, transforming growth factor (TGF)- $\beta$  is an anti-inflammatory cytokine that facilitates angiogenesis while simultaneously promoting differentiation and influencing the expansion of keratinocytes; yet, excessive production of TGF- $\beta$ 1 in skin cells induces significant inflammation.<sup>29-31</sup> However, oxidative damage has a role in the etiology of AD. Environmental factors, such as contaminants, ultraviolet radiation, illness, and mental illness, may elevate reactive oxygen species (ROS) outputs, finally exceeding the protective capacity of the anti-oxidative barrier.<sup>32-36</sup>

Nevertheless, topical medicines, including calcineurin blockers, glucocorticoids, and, lastly, phosphodiesterase 4 (PDE4) antagonists such as crisaborole, are the bedrock of AD management.<sup>37-39</sup> Yet, repeated usage of these medicines might cause an array of unwanted effects. Thus, there is a pressing demand for the identification of innovative and secure methods for treating AD.<sup>40,41</sup> The approval of targeted biologics such as dupilumab, tralokinumab, nemolizumab, baricitinib, upadacitinib, and abrocitinib suggests subsequent best alternatives for controlling moderate to severe forms of AD.<sup>42,43</sup>

Ezetimibe is a lipid-diminishing medication that specifically hinders the cholesterol transporter Niemann-Pick C1-like protein 1 (NPC1L1) in the jejunum

brush borders, restricting the digestion of cholesterol in the stomach and bile systems. Ezetimibe was demonstrated to be safe and efficient in decreasing the contents of cholesterol regularly accessible to hepatocytes. As a result of diminished cholesterol supply, the liver raises LDL receptor generation and LDL removal from the circulation.<sup>44-47</sup> Ezetimibe was established to stop atherosclerotic events when combined with statin, making it useful in the treatment of dyslipidemia and related atherosclerotic complications.<sup>48,49</sup> Ezetimibe's impact on many inflammatory indicators was also studied. Ezetimibe was revealed to lower atherosclerotic plaques, limit macrophage accumulations, and prevent liberation of chemotactic cytokines like MCP-1 and TNF- $\alpha$ .<sup>50,51</sup> Other observation demonstrate that combining ezetimibe with simvastatin can effectively alleviate autoimmune alopecia totalis and alopecia universalis, making it a promising treatment option for resistant alopecia areata.<sup>52</sup> This combination also considerably lowered amounts of IL-1 $\beta$  and IL-18 in obese individuals.<sup>53</sup> Beyond that, this medicine exerted protective actions on IL-1 $\beta$ -exacerbated matrix collagen breakdown in murine chondrocytes by modifying NF- $\kappa$ B and Nrf2/HO-1 molecular parameters.<sup>54</sup> In parallel, ezetimibe enhanced endothelial functions and decreased inflammation indicators, disease progression, and aortic stiffening among individuals with rheumatoid arthritis.<sup>55</sup> In comparable vein, ezetimibe shows inhibitory impacts on acetic acid-exacerbated rat prototype of ulcerated colitis by downregulating inflammatory and oxidative indicators, particularly the Akt/NF- $\kappa$ B/STAT3/CXCL10 signaling network.<sup>56</sup>

## Aim

Previous studies suggest that ezetimibe possesses anti-inflammatory, antioxidant, and immunomodulatory properties; however, its anti-eczematous efficacy has not been investigated. This study aimed to evaluate, for the first time, the anti-atopic effects of topical ezetimibe in a DNCB-induced murine model of AD, providing novel preclinical evidence for its potential repositioning as a topical therapy in dermatological inflammatory disorders.

## Material and methods

### Drugs and chemicals

The suppliers of 1-chloro-2,4-dinitrobenzene (DNCB) was the Sigma Aldrich, Germany. Ezetimibe was purchased from Hangzhou Hyper Chemicals Limited. The 0.1% tacrolimus product was supplied by Cleveland Clinic, USA. The extra chemicals utilized in this investigation had been procured from several sources who met specific standards.

### Preparation of ezetimibe ointment

2.5 g of white wax (beeswax), 1.25 g of hard paraffin, 2.5 g of lanolin, polyethylene glycol, and a sufficient quan-

ity of soft paraffin were melted together in a saucepan on a hot plate at 70 °C.<sup>57</sup> 2g of ezetimibe powder was placed to the melted mixture while being continuously stirred.<sup>58,59</sup> The entire mixture was blended while transported on ice to produce 2% ezetimibe ointment.<sup>60-64</sup>

#### **Vehicle components formulation**

2.5 g of white wax, 1.25 g of hard paraffin, 2.5 g of lanolin, polyethylene glycol, and an adequate amount of white soft paraffin were combined and heated in a container on a heated surface at 70°C. The whole solution was constantly agitated and mixed.<sup>65,66</sup>

#### **Study design**

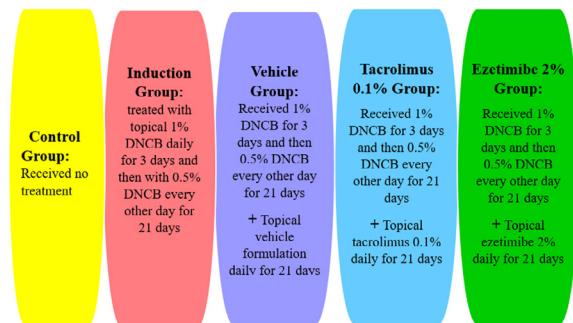
This work was licensed by the institutional review board (IRB) of the College of Medicine at Al-Nahrain University (approval no. UNCOMIRB20241037 on 14/10/2021). The experiments were carried out from November 2021 to July 2022. The trial consisted of thirty male Swiss albino-kind mice, with weight about 20 to 30 g and age about 8 to 9 weeks. Mice were placed in a sterile, isolated, and properly designated room with a humidity level of 49±2%. All investigations had been performed in conformity with rules of ethics for the handling of animals.

#### **Animal preparation and sampling**

Mice had been randomly separated into 5 different categories of 6 mice each: a healthy untreated control group, an induction non-treated group, a vehicle group, an ezetimibe group, and a tacrolimus group. Following a 14-day acclimation period, mice underwent rigorous hair extraction from their back areas (about 4 cm<sup>2</sup>) with a precision shaving device. The residual hair strands were subsequently extracted with a depilatory lotion. To develop AD-mimicking skin irritation, DNCB was dispersed in an acetone-based olive oil mixture at a 3:1 proportion and watered down to 0.5 and 1% strengths. Mice developed a sensitivity with 200 µL of 1% DNCB applied to their back skin-folds for 3 straight days, whereas the healthy controls did not obtain any medications. Mice with AD-like symptoms were treated with 200 µL of 0.5% DNCB locally each other day for 21 days to prevent unexpected resurgence. The vehicle group was given a topical vehicle formulation daily for 21 days, the tacrolimus group obtained tacrolimus 0.1% ointment daily for 21 days, and the ezetimibe group administered ezetimibe 2% ointment product daily for 21 days as indicated in Figure 1. At the completion of the tests, all animals underwent the intraperitoneal analgesic medicines xylazine and ketamine (80 mg/kg). Upon achieving complete anesthesia, the animals were put to death by exsanguinations, which are the optimal means of harvesting and preserving.<sup>67-70</sup> Skin biopsies were subsequently processed to develop tissue homogenates for bio-indicator and histopathology analyses.<sup>71-74</sup>

#### **Measurement of total and differential WBC counts**

The complete blood counts involves the overall amount of leukocytes/white blood cells (WBC) alongside the individual percentages for each WBC subgroup, such as neutrophils, lymphocytes, monocytes, and eosinophils. Blood specimens were taken in EDTA-containers and scrutinized with a small 5-part hematologic tester, namely the Mindray BC-5000 version.<sup>75,76</sup> The methodology is based on triangular laser scattering, flow-cytometry, and biochemical dye processing.<sup>77-81</sup>



**Fig 1.** Tabular form of study groups

#### **Estimation of severity grading of AD**

The intensity of DNCB-evoked AD-mimicking cutaneous patches was estimated by 4 clinical manifestations: erythema, erosion/excoriation, epidermal thickenings, and lichenification, based on the Eczema Area and Degree Index (EASI) scoring technique.<sup>82</sup> The metrics were evaluated on a scale of 0 (none), 1 (mild), 2 (moderate), and 3 (severe) based on their severity, with total dermatitis levels determined by summing all analyzed data, achieving a highest possible score of 12.<sup>83</sup> The back skin-fold thickening was calculated using a vernier calibrator measuring instrument on the central line of the back skin.<sup>84-86</sup> Dermatitis severity was assessed by two independent raters. Dermatitis scoring and treatment administration were conducted in a blinded fashion to minimize observer bias. The experimenters were unaware of the treatment groups during scoring.

#### **Histopathological estimation**

The dorsal skins of mice were obtained from various categories and preserved in 10% neutralizing-buffer formaldehyde employing recognized guidelines.<sup>87-90</sup> Fixed paraffin was then applied to the skin samples. The paraffin-soaked tissues were sliced into slices and prepared for staining with hematoxylin and eosin. The derived specimens were viewed utilizing a light microscopy, and histological changes were documented.<sup>91-95</sup> Histological scoring relied on an amended edition of the scoring system described by Jeong et al., commonly used for DNCB-induced dermatitis models. Parameters encompassed skin thickness/acanthosis, hyperkeratosis, parakeratosis, erosion, inflammatory cell invasion, and

edema.<sup>96</sup> The grading mechanisms varied from 0, signifying no irregularity, to 1+, denoting mild irregularity; 2+, representing modest irregularity; and 3+, signifying substantial irregularities.

#### *Immunohistochemistry (IHC) examination*

The immunohistochemistry approach was used to assess the severity of AD lesions by determining the scores of IL-4 and IL-13. It is technique for detecting antigens or haptens in biological tissues. The immunohistochemical assessment in this study employs specific antibodies for identifying the protein product of gene expression in the tissues of the investigated animal groups. Following the application of the main antibody (rabbit monoclonal anti-IL-4 and anti-IL-13), the corresponding secondary antibody and a chromogen agent (3,30-diaminobenzidine) are then incorporated into paraffin-fixed segments of the skin tissues.<sup>31,97-99</sup> The skin portions have been treated with hematoxylin. The immunohistochemistry investigation is scored via the following method using semi-quantitative scoring (positive stained cells): "Score 0=no stain; score 1=25%; score 2=26–50%; score 3=51–75%; score 4=67–100%."<sup>30,100,101</sup>

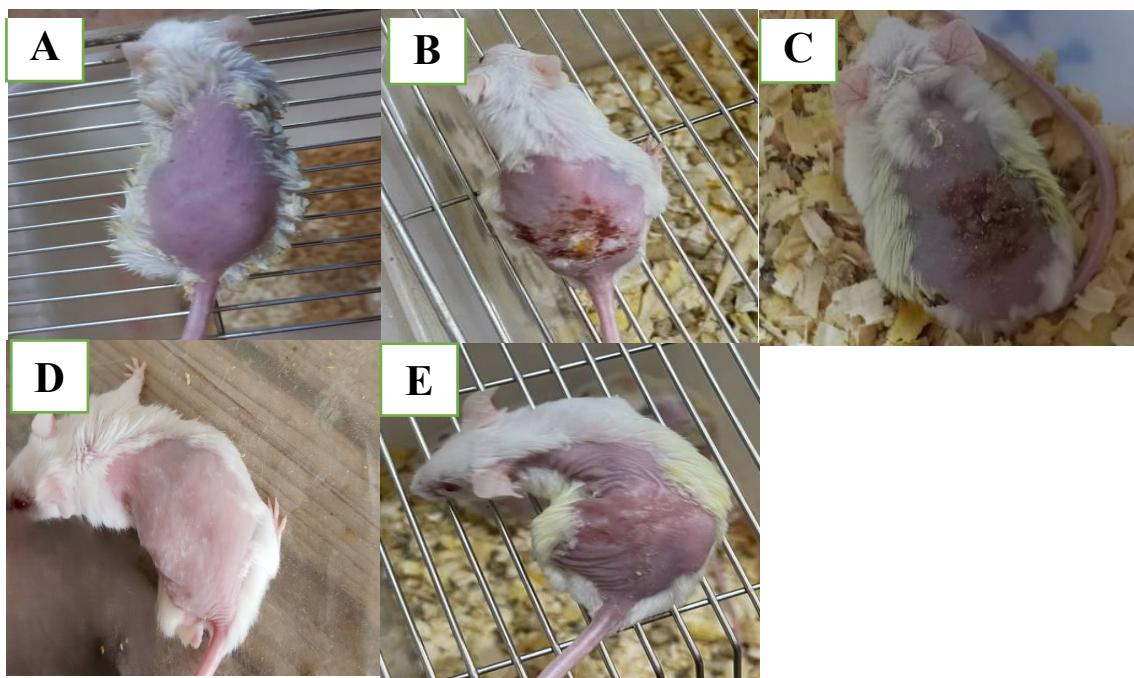
#### *Biochemical marker measurement*

Using sandwich enzyme-linked immunosorbent-assays (ELISA), the amounts of IgE, malondialdehyde (MDA), IL-17, IL-31, TGF- $\beta$ , and TNF- $\alpha$  in mouse skin tissues were determined by the supplier's rules. IgE Invitrogen: catalogue no.: 88-50460-88, sensitivity: 4 ng/mL; MDA Invitrogen: catalogue no.: EEA015, sensitivity: 1.13  $\mu$ mol/L; IL-17 Sino biological: catalogue no.:

KIT51065, sensitivity: 8 pg/mL; IL-31 Invitrogen: catalogue no.: BMS6030, sensitivity: 9.1 pg/mL; TGF- $\beta$  Invitrogen: catalogue no.: 88-8350-88, sensitivity: 8 pg/mL; TNF- $\alpha$  Sino biological: catalogue no.: KIT50349, sensitivity: 15.69 pg/mL. To ensure statistical validation, ELISA measurements were done in duplicate. Following a cleansing with washing buffer, the pores were treated with a biotin-based-specific antibody.<sup>102-106</sup> Upon extracting the disengaged biotin-linked antibody, streptavidin/horseradish peroxidases (HRP) were gently transmitted into the wells. Tetramethylbenzidine (TMB) substrate solution was added once all unbound components were eliminated. The amounts of various variables in every experimental group were determined spectro-photometrically by matching their optical density to regular curves. The HRP-combined antibodies and indicators switched to yellow upon the addition of the stop solution. The levels of various variables in each experimental group were determined spectro-photometrically by comparing their optical densities to standard curves. Following the addition of the stop solution, the HRP-conjugated antibodies and substrates produced a yellow color, which was measured using an absorbance microplate reader at a wavelength of 450 nm.<sup>107-109</sup>

#### *Statistical analysis*

The statistical evaluation was conducted using the program Excel 2013 and the SPSS statistical program edition 24 (IBM, Armonk, NY, USA). The numerical parameters are presented in terms of means with standard deviation ( $\pm$ SD), and the level of significance is estimated at a p value of less than 0.05. Means were contrasted



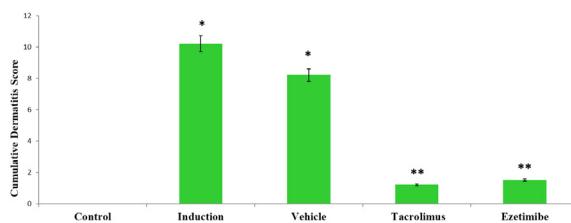
**Fig. 2.** Images displaying the degree of severity of eczematous lesions in study groups: A – control group, B – induction group, C – vehicle group, D – tacrolimus group, E – ezetimibe

with the t-test for independence and an assessment of variance (ANOVA). Pearson's correlation is adopted to determine linear relationships among the groups under study.<sup>110,111</sup>

## Results

### *Impact of researched agents on observational severity scores*

The induction and vehicle groups had significantly elevated observational severity levels than normal controls ( $p<0.05$ ). Also, there was no considerable variance among induction/model and vehicle groups with respect to observational dermatitis levels ( $p>0.05$ ). Nonetheless, the dermatitis severity ratings got significantly reduced in the tacrolimus and ezetimibe-treated groups when matched with the induction and vehicle groups ( $p<0.05$ ). No noteworthy variances were detected across the tacrolimus and ezetimibe groups with respect to observational dermatitis levels ( $p>0.05$ ) as observed in Figure 2 and Figure 3.



**Fig 3.** Impact of researched agents on the observational scores, data are illustrated as mean $\pm$ SD; \*denote considerable variation ( $p<0.05$ ) vs. control group; # denote considerable variation ( $p<0.05$ ) vs. induction and vehicle groups

**Table 1.** Impact of researched agents on total WBC, neutrophils, lymphocytes, monocytes, and eosinophils counts<sup>a</sup>

Variables ( $\times 10^3/\text{mL}$ )	Groups					p
	Control	Induction	Vehicle	Tacrolimus	Ezetimibe	
WBC	3916 $\pm$ 558	14180 $\pm$ 1856*	14120 $\pm$ 1625*	6640 $\pm$ 614#	5850 $\pm$ 290*	<0.05
Neutrophils	1465 $\pm$ 221	4934 $\pm$ 1434*	4910 $\pm$ 1522*	2880 $\pm$ 198*	2519 $\pm$ 250*	<0.05
Lymphocyte	2260 $\pm$ 638	5940 $\pm$ 1086*	5890 $\pm$ 982*	3220 $\pm$ 580*	2995 $\pm$ 442*	<0.05
Monocytes	81 $\pm$ 12	1325 $\pm$ 352*	1294 $\pm$ 416*	543 $\pm$ 118*	404 $\pm$ 44#	<0.05
Eosinophils	111 $\pm$ 28	1939 $\pm$ 512*	1888 $\pm$ 448*	725 $\pm$ 124*	577 $\pm$ 93#	<0.05

<sup>a</sup> data were expressed as mean $\pm$ SD, \* – denotes remarkable change ( $p<0.05$ ) contrasted to the control group, # – denotes remarkable change ( $p<0.05$ ) contrasted to the induction and vehicle groups

### *Effect of tested agents on total and differential WBC counts*

The percentages of total WBC, neutrophils, lymphocytes, monocytes, and eosinophils were substantially in-

creased in induction and vehicle groups as opposed to normal controls ( $p<0.05$ ). Besides, ezetimibe and tacrolimus groups show considerably diminished counts of total WBC, neutrophils, lymphocytes, monocytes, and eosinophils than induction and vehicle groups as indicated in Table 1.

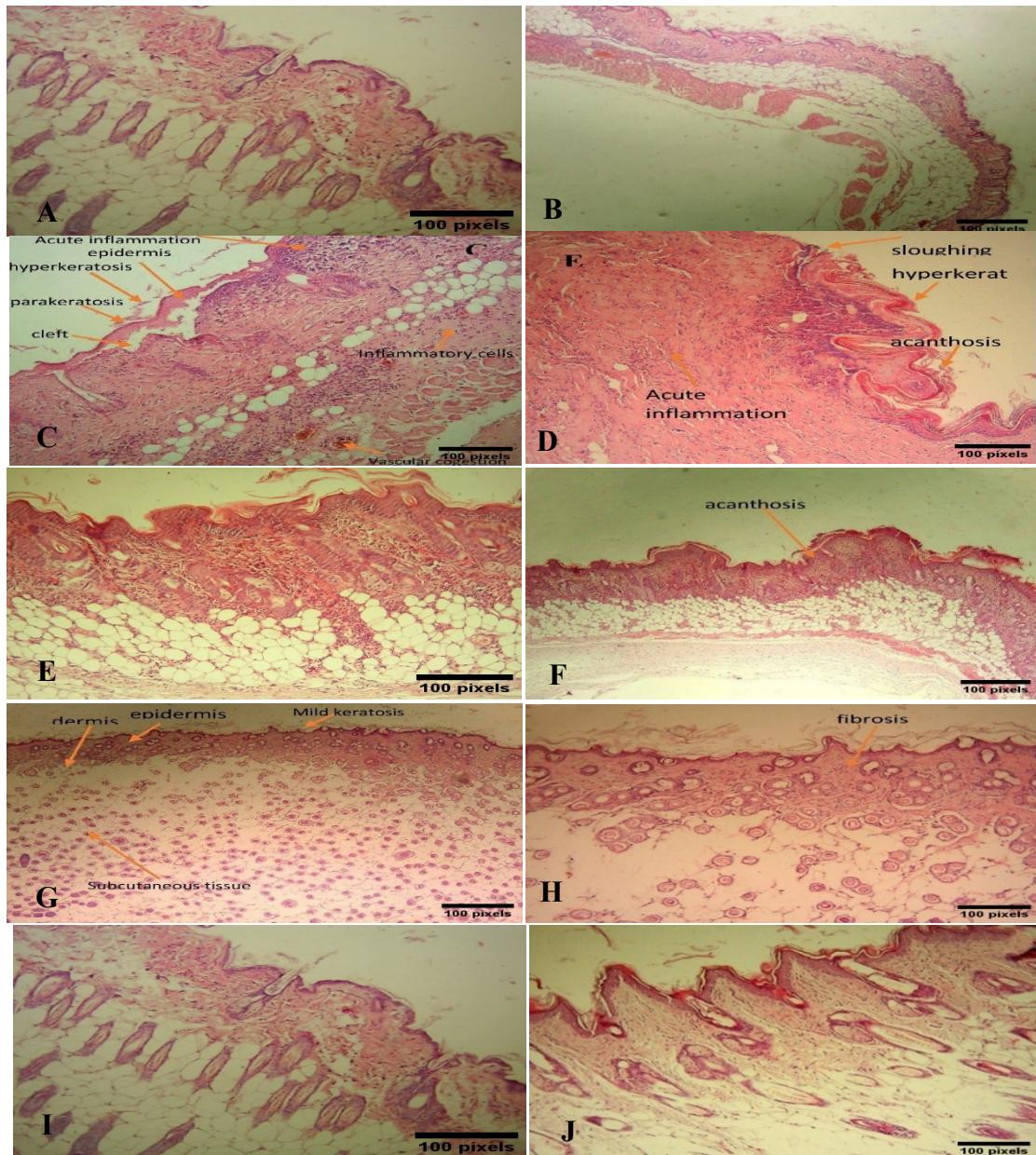
### *Effect of tested agents on skin histological abnormalities*

The healthy control group has regular skin microstructure; the keratin epidermal layers, dermal uppermost layer, sebaceous glands, and hair proliferating follicles all appear normal, as illustrated in Table 2 and Figure 4. However, the application of DNCB resulted in substantial histopathological changes in the induction group opposed to the healthy untreated controls ( $p<0.05$ ). Those skin changes were recognized by the breakdown of the skin barriers, the presence of clefts, marked hyperkeratosis, marked parakeratosis, increased acanthosis, profound erosion, remarkable edema, profound sloughing, and prominent inflammatory cell infiltration as represented in Table 2 and Figure 4. Except for erosion and extracellular edema, the vehicle group exhibited non-significant histopathological changes as matched to the induction group ( $p>0.05$ ), including increased epidermal thickenings/acanthosis, marked hyperkeratosis, marked parakeratosis, significant erosion, significant edema, and increased inflammatory cell penetration. The erosion and extracellular edema in the vehicle group were markedly alleviated compared to those seen in the induction. In opposition to the induction and vehicle groups, the tacrolimus 0.1% treated group showed substantially smaller histological aberrations, such as reduced cutaneous thickness, minor hyperkeratosis, minor parakeratosis, slight erosion, slight edema, and reduced inflammatory cell invasion as depicted in Table 2 and Figure 4. Additionally, the histopathological changes were markedly diminished in the ezetimibe group versus the induction group ( $p<0.05$ ), as demonstrated by decreased epidermal thickness, mild hyperkeratosis, mild parakeratosis, mild erosion and reduced inflammatory cell infiltration, as revealed in Table 2 and Figure 4.

**Table 2.** Effect of tested agents on histopathological scores<sup>a</sup>

Variables	Mean $\pm$ SD					
	Control	Induction	Vehicle	Tacrolimus	Ezetimibe	p
Acanthosis	0	3 $\pm$ 0.4*	3 $\pm$ 0.04*	1 $\pm$ 0.05#	1 $\pm$ 0.04#	<0.05
Hyperkeratosis	0	3 $\pm$ 0.06*	3 $\pm$ 0.08*	1 $\pm$ 0.2#	2 $\pm$ 0.04#	<0.05
Parakeratosis	0	3 $\pm$ 0.04*	3 $\pm$ 0.18*	0 $\pm$ 0.03#	0 $\pm$ 0.04#	<0.05
Erosion	0	3 $\pm$ 0.05*	1 $\pm$ 0.25*	0 $\pm$ 0.05#	0 $\pm$ 0.04#	<0.05
Inflammatory cell infiltration	0	3 $\pm$ 0.1*	3 $\pm$ 0.06*	1 $\pm$ 0.1#	1 $\pm$ 0.04#	<0.05

<sup>a</sup> data are illustrated as mean $\pm$ SD, \* – denote considerable variation ( $p<0.05$ ) vs. control group, # – denote considerable variation ( $p<0.05$ ) vs. induction and vehicle



**Fig. 4.** Effect of studied agents on mouse skin histological alterations: A and B: light microscopy cross-section of mouse skin histopathology for healthy controls (H&E stain=10x and 4x), C and D: light microscopy cross-section of mouse skin histopathology for induction/ model group (H&E stain=10x), E and F: light microscopy cross-section of mouse skin histopathology for vehicle group (H&E stain=10x and 4x), G and H: light microscopy cross-section of mouse skin histopathology for tacrolimus group (H&E stain=10x), I and J: light microscopy cross-section of mouse skin histopathology for ezetimibe group (H&E stain=10x)

groups

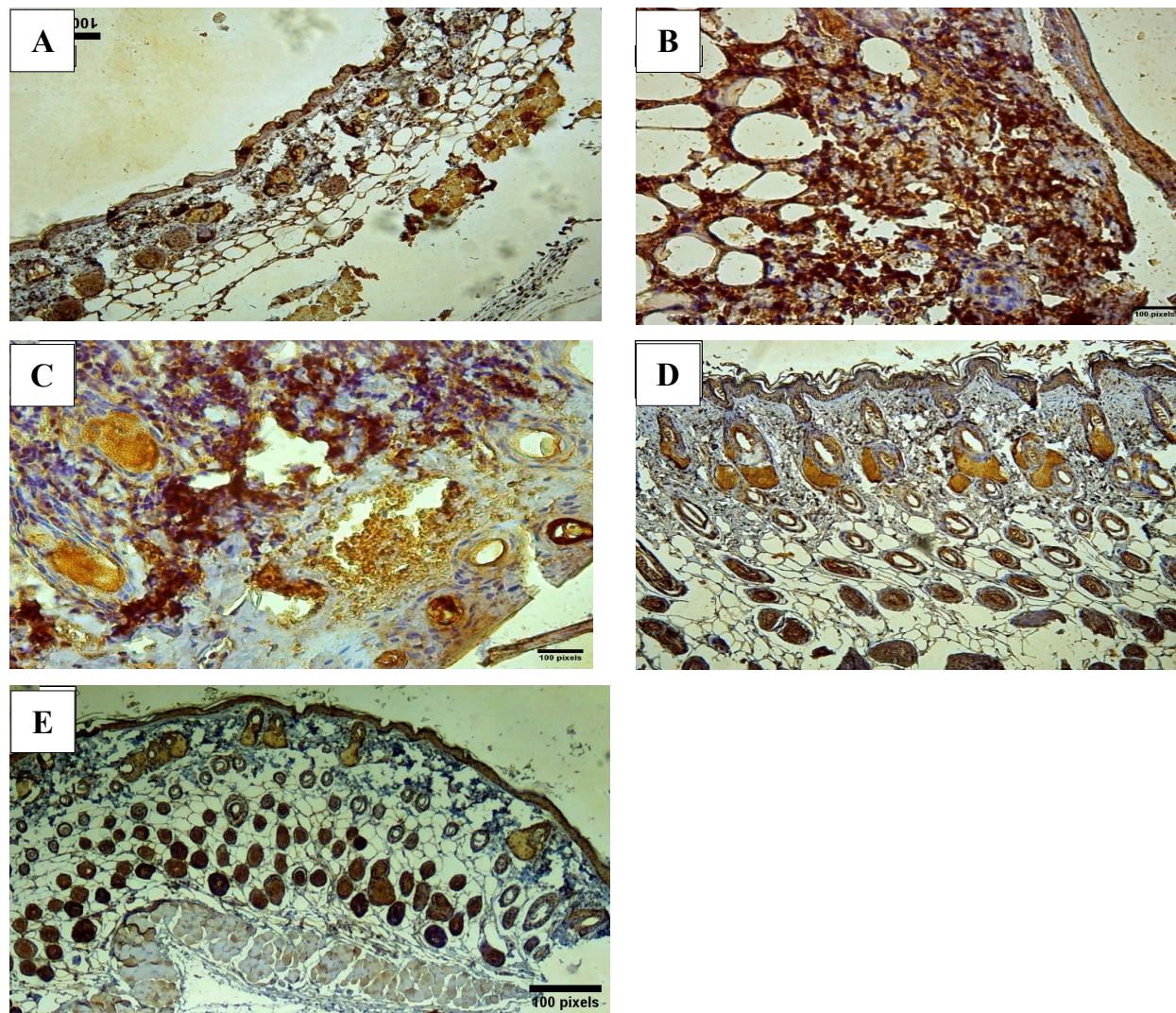
#### *Impact of tested agents on skin immunohistochemical levels of IL-4 and IL-13*

The immunohistochemistry investigation revealed considerable elevation of IL-4 and IL-13 scores among the model/induction and vehicle groups as opposed to the normal controls ( $p<0.05$ ), but there are no appreciable variations among the induction and vehicle groups ( $p>0.05$ ). The tacrolimus and ezetimibe-treated groups exhibited substantial diminution in IL-4 and IL-13 contents when opposed to the induction and vehicle

non-treated groups ( $p<0.05$ ). However, no substantial changes were noted across the tacrolimus and ezetimibe groups in terms of IL-4 and IL-13 scores ( $p>0.05$ ), as clarified in Figure 5 and Figure 6.

#### *Effect of tested agents on skin tissue levels of IgE and MDA*

The outcomes uncovered that the induction/model group exhibited markedly greater mean skin amounts of IgE and MDA as matched to the healthy untreated control group ( $p<0.05$ ). No notable variations were seen



**Fig. 5.** Impact of studied agents on skin immunohistochemistry scores of IL-4: groups: (scale bar=100 mm), A: microscopic mouse cutaneous slice from the control group illustrating the immunohistochemical levels of IL-4, B: microscopic mouse cutaneous slice from the induction/model group illustrating the immunohistochemical levels of IL-4, C: microscopic mouse skin slice from the vehicle group illustrating the immunohistochemical levels of IL-4, D: microscopic mouse cutaneous slice of the tacrolimus group illustrating the immunohistochemical levels of IL-4, E: microscopic mouse cutaneous slice of the ezetimibe group illustrating the immunohistochemical levels of IL-4

in any of the parameters between the induction and vehicle groups. However, the tacrolimus and ezetimibe groups had dramatically lower levels of IgE and MDA relative to the induction and vehicle groups ( $p<0.05$ ). Both the tacrolimus and ezetimibe groups did not vary considerably from one another on any of the parameters ( $p>0.05$ ), as shown in Figure 7.

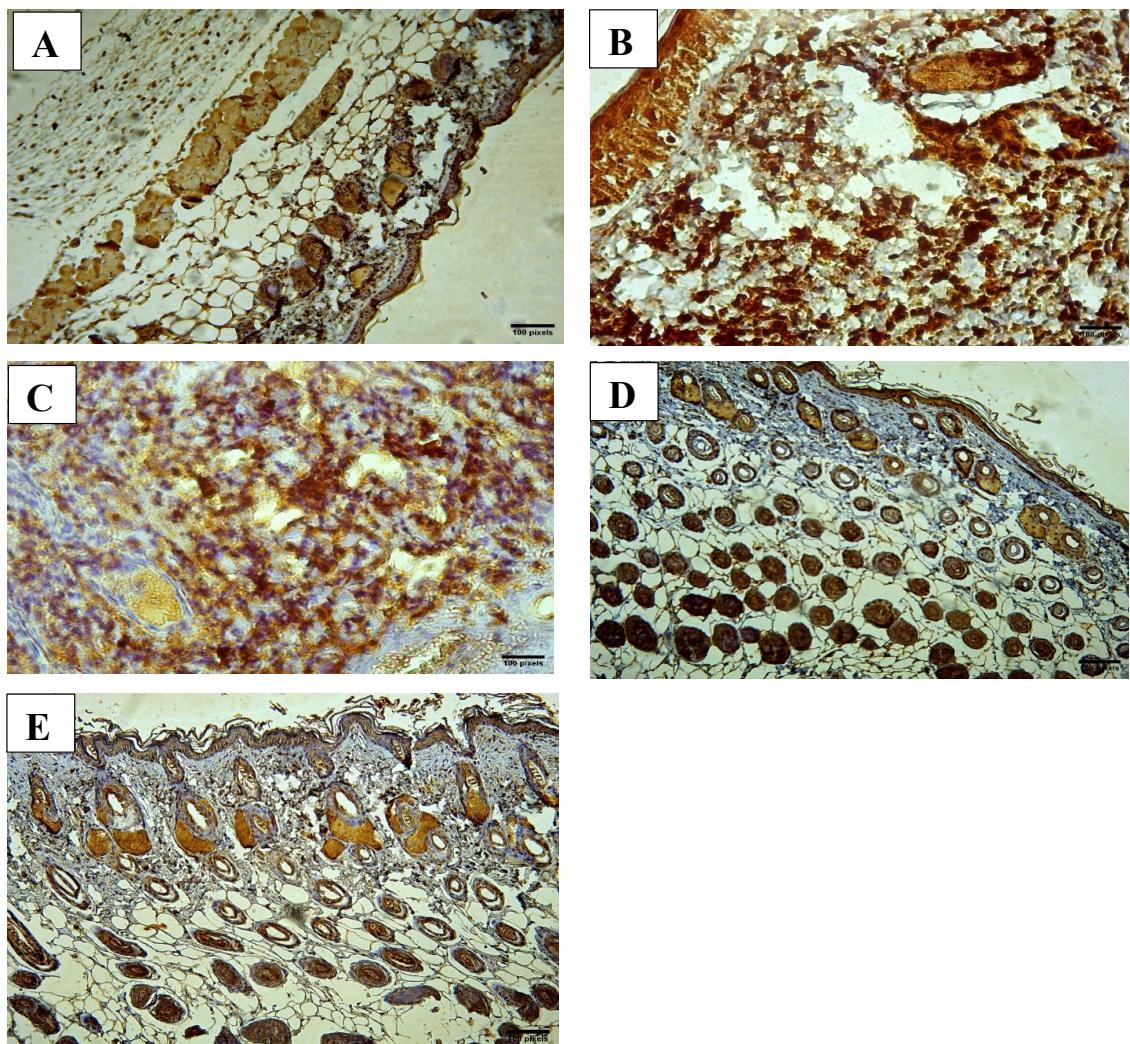
#### **Effect of tested agents on skin tissue levels of IL-17, IL-31, TGF- $\beta$ , and TNF- $\alpha$**

The results demonstrated that the induction group had markedly elevated mean epidermal levels of IL-17, IL-31, TGF- $\beta$ , and TNF- $\alpha$  in relation to the healthy controls ( $p<0.05$ ). No remarkable variances occurred in all metrics across the model/induction and vehicle groups. Still, the tacrolimus and ezetimibe groups exhibited ex-

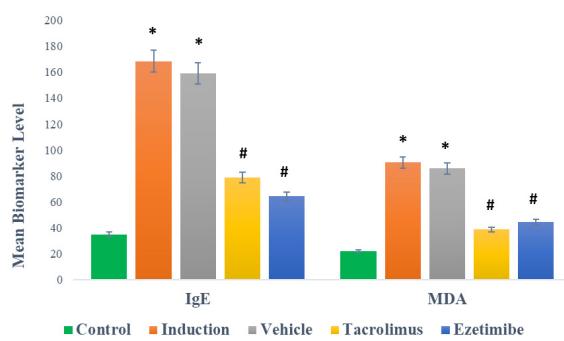
tensively mitigated amounts of IL-17, IL-31, TGF- $\beta$ , and TNF- $\alpha$  relative to the induction/model and vehicle groups ( $p<0.05$ ). The tacrolimus and ezetimibe groups exhibited no significant distinctions across all variables ( $p>0.05$ ), as explained in Figure 8.

#### **Discussion**

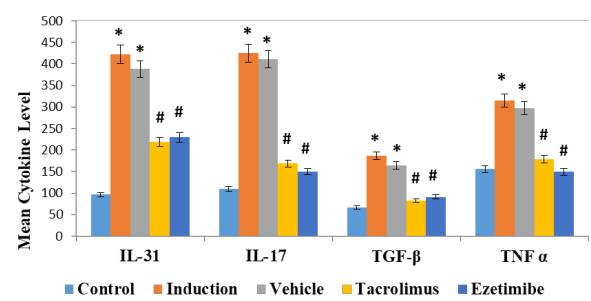
The novelty of this work lies in demonstrating for the first time that ezetimibe, beyond its systemic lipid-lowering action, exerts strong topical anti-inflammatory and antioxidant effects in a murine model of AD. The mouse prototype of DNCB-aggravated atopic dermatitis was extensively utilized to assess innovative therapies and compounds. The research designs and methodologies are markedly varied, revealing distinct patterns of atopic dermatitis that manifest after sensitization and



**Fig. 6.** Impact of studied agents on skin immunohistochemistry scores of IL-13: groups: (scale bar=100 mm), A: microscopic mouse cutaneous slice from the control group illustrating the immunohistochemical levels of IL-13, B: microscopic mouse cutaneous slice of the induction/model group illustrating the immunohistochemical levels of IL-13, C: microscopic mouse cutaneous slice of the vehicle group illustrating the immunohistochemical levels of IL-13, D: microscopic mouse cutaneous slice of the tacrolimus group illustrating the immunohistochemical levels of IL-13, E: microscopic mouse cutaneous slice of the ezetimibe group illustrating the immunohistochemical levels of IL-13



**Fig. 7.** Impact of tested agents on IgE and MDA (the data expressed as mean $\pm$ standard deviation; \* –implies a statistically marked difference ( $p<0.05$ ) when contrasted to the normal controls, while # – implies a marked variation ( $p<0.05$ ) when contrasted to the model/induction and vehicle groups



**Fig. 8.** Effects of tested agents on IL-17, IL-31, TGF- $\beta$ , and TNF- $\alpha$ , the data expressed in the form of mean $\pm$ standard deviation, \* – implies a statistically remarkable difference ( $p<0.05$ ) when contrasted to the control group, while # – implies a remarkable variation ( $p<0.05$ ) when contrasted to the model/induction and vehicle groups, IL – interleukin, TGF- $\beta$  – transforming growth factor- $\beta$ ; TNF- $\alpha$  – tumor necrosis factor- $\alpha$

frequent exposure to DNCB on the back skin.<sup>112</sup> In this work, the disease was induced with 1% DNCB in the sensitization phase and successive administrations of 0.5% DNCB in the challenge stage, resulting in a mild phenotype of eczematous dermatitis.<sup>83</sup> The DNCB-treated skin exhibited a moderate extrinsic subacute atopic dermatitis lesion on day 12 and a mild extrinsic subacute to prolonged phenotype and endotype on day 22, characterized by a predominant Th2 response. Both timepoints demonstrate hallmark features linked with atopic dermatitis, including dorsal skin thickening, hyperkeratosis, parakeratosis, elevated TH1 and TH2 cytokine levels, and alterations in barrier proteins. Elevated mast cell influx in the epidermis and higher plasma IgE levels signify a type I allergic reaction.<sup>83,113</sup>

In this study, administering DNCB to the murine dorsal regions resulted in evident indications of desquamation, erythema, and blisters, along with a notable rise in eczema severity indices. Furthermore, the aggregate populations of WBCs, neutrophils, lymphocytes, monocytes, and eosinophils were strongly elevated, and immunohistochemical testing indicated a large increment in IL-4 and IL-13 values. DNCB further drastically boosts the amounts of IgE, MDA, IL-17, IL-31, TGF- $\beta$ , and TNF- $\alpha$ , while histological examination reveals pathological features such as hyperkeratotic changes, dermal thickness, and infiltrating lymphocytes, therefore confirming the AD model. Topical treatment is a popular method of ameliorating clinical manifestations, although it might trigger epidermal thinning and hypersensitivity.<sup>114–116</sup> Tacrolimus is an effective prolonged topical therapy for AD that acts by reducing phosphatase activity in the calcineurin pathway, resulting in decreased T-lymphocyte activation and inflammatory cytokine production. It also improves skin barrier function by stimulating the development and maturation of skin cell.<sup>117–119</sup> Prior research has demonstrated that the anti-atopic benefits of tacrolimus entail the suppression of total and differential leukocytes, IgE activity, oxidative measures notably MDA, and Th1- and Th2-related inflammatory cytokines, notably IL-4, IL-13, IL-17, IL-31, TNF- $\alpha$ , and TGF- $\beta$ , as well as various molecular signaling cascades.<sup>120–124</sup>

Topical tacrolimus 0.1% yielded an immediate decrement in overall dermatitis scores, inflammation, and pruritus.<sup>117</sup> The most frequent undesirable actions of tacrolimus entail burning irritations at the sites of administration.<sup>125–127</sup> Consequently, developing innovative, efficacious medications for AD is essential.

In the current study, ezetimibe 2% ointment demonstrated a suppressive impact on DNCB-evoked AD-mimicking cutaneous erosions in mice, as evidenced by substantially reduced amounts of inflammatory and oxidative biomarkers, histological alterations, and the dermatitis severity scores. Additionally, a con-

siderable decrement in the numbers of total leukocytes, lymphocytes, monocytes, eosinophils, and neutrophils was observed in comparison to the induction group. These outcomes were similar to those of Suchy et al. who discovered that ezetimibe has direct inhibitory effects on the activation pathways of immune cells, including monocytes and macrophages.<sup>128</sup> Another research indicated that ezetimibe administration yielded in a dose-related decline in both the overall proportion of CD3+CD4+ T cells and the number of CD3+/CD4+/CD45RO T memory lymphocytes.<sup>129</sup> Aside from the effect on immune cells, ezetimibe additionally offers other benefits, such as lowering the production of C-reactive protein, an influential inflammatory biomarker.<sup>130</sup> Likewise, topical ezetimibe experienced anti-inflammatory properties, reducing ear edema by 64%.<sup>131</sup> supporting current research that suggests it may be applied to treat inflammatory skin conditions. These favorable effects of ezetimibe could be explained by its ability to down-regulate inflammatory mediators, suppress macrophage activation, and regulate the NF- $\kappa$ B, a chain accountable for producing various inflammation-related cytokines. In this context, ezetimibe treatment caused I $\kappa$ B decomposition and thereby suppressed NF- $\kappa$ B translation via the mitogen-activated protein kinases (MAPKs) mechanism. This evidence showed that there might be an opportunity of using ezetimibe to treat and prevent inflammatory disorders.<sup>132</sup> Another proof implies that ezetimibe might exhibit anti-inflammatory features alongside its lipid-lowering impacts, as NF- $\kappa$ B stimulation is influenced by chemotactic cytokines and substances involved in intracellular defense.<sup>53</sup> Moreover, ezetimibe alleviated clinical manifestations of ankylosing spondylitis in animals by suppressing Th17 differentiating-associated genes like IL-23R and IL-1R and modifying the Th17/Treg cell harmony resulting in a distinct anti-inflammatory impact irrespective of circulatory lipid lowering. The researchers found that ezetimibe dramatically boosted the overall count of Treg cells while decreasing the proportion of Th17 and Th1 lymphocytes. Ezetimibe also markedly decreased IFN- $\gamma$ , TNF- $\alpha$ , IL-1 $\beta$ , IL-6, and IL-17 concentrations. As a result, ezetimibe modulates T-cell proliferation and pro-inflammatory cytokine release by immune system cells.<sup>133</sup> Similarly, ezetimibe inhibited lymphocytic production of TNF- $\alpha$ , IFN- $\gamma$ , and IL-2 in hypercholesterolemic individuals in a lipid-unrelated way.<sup>134</sup> Subsequent studies concluded that ezetimibe successfully managed the rat model of ulcerative colitis by alleviating the output of pro-inflammatory measures like TNF- $\alpha$ , IL-1 $\beta$ , and NF- $\kappa$ B alongside oxidative measures like MDA and MPO in colon homogenate.<sup>135–137</sup> Likewise, literatures showed that ezetimibe is effective in treating of alopecia, an immune-related disease featured by T-lymphocyte stimulation, accumulation mast cells and pro-

duction TNF- $\alpha$ .<sup>138-140</sup> Alopecia sufferers are more likely to acquire other autoimmune disorders, like atopic dermatitis, vitiligo, and skin cancer.<sup>141</sup>

Meanwhile, this work found that ezetimibe remarkably reduced the immunohistochemical expressions of IL-4 and IL-13 in the back cutaneous folds. This outcome concurs with prior research, which indicated that ezetimibe/simvastatin combination contributed to decreased immunohistochemistry expression of chemotactic cytokines among alopecia areata patients.<sup>141</sup> Additional observation revealed that ezetimibe possesses immunomodulatory impacts on antigenic presentation, lymphocytic trafficking, and regulatory T cell function.<sup>142</sup> Ezetimibe was also documented to mitigate inflammatory variables like CRP, monocyte chemoattractant protein-1, IL-1 $\beta$ , IL-6, TNF- $\alpha$ , and pro-oxidant markers.<sup>143-145</sup> In macrophage tests, ezetimibe reduced NF- $\kappa$ B expression and blocked the NLRP3 inflammasome-IL-1 $\beta$  route, resulting in anti-inflammatory activity.<sup>146</sup> Ezetimibe can also attenuate inflammation and oxidative injury induced by caspase-1/IL-1 $\beta$  via the AMP-stimulated protein kinase/NF-E2-relating factor 2 (Nrf2)/thioredoxin-interacting protein (TXNIP) system.<sup>147</sup> In addition, this medication decreases monocyte chemoattractant protein 1-induced recruitment.<sup>148,149</sup> All these outcomes support our hypothesis that ezetimibe could be of an important value in attenuating experimentally-induced model of AD.

### Study limitations

Nonetheless, the present investigation encompassed some limitations. Actually, our data exclusively confirmed the inhibitory effect of ezetimibe on a murine male prototype of DNCB-aggravated AD, neglecting the influence of gender-related fluctuations on the experimental results and failing to apply human participants due to the disparities between rodents and humans. Consequently, the effectiveness and security of ezetimibe in the management of AD ought to be assessed in people. Moreover, the further in-depth mechanism by which ezetimibe confers preventive impacts in mice afflicted with AD needs to be elucidated. The issue pertains to implications on other immune-related elements such as the proliferation of systemic immune receptors (sCD25, sCD30), the atopy patch tests, and the filaggrin genes. Further investigation is necessary to assess the efficacy of the combined use of ezetimibe and tacrolimus.

### Conclusion

Topical ezetimibe significantly attenuated DNCB-induced eczematous dermatitis in mice by reducing histopathological alterations, suppressing pro-inflammatory cytokines, and lowering total and differential leukocyte counts. These results provide the first evidence support-

ing ezetimibe as a potential adjunctive therapy for immune-mediated and inflammatory dermatoses. The findings also highlight a novel therapeutic avenue for drug repurposing of ezetimibe in dermatology, particularly in AD.

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### Declarations

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#### Author contributions

Conceptualization, A.M.S. and A.R.A.; Methodology, H.R-S.; Software, W.H.A.; Validation, A.R.A., H.R-S. and A.J.H.A.; Formal Analysis, A.M.S.; Investigation, W.H.A.; Resources, A.M.S.; Writing – Original Draft Preparation, H.R-S., A.M.S. and A.R.A.; Writing – Review & Editing, H.R-S.; Visualization, A.R.A., W.H.A. and A.J.H.A; Supervision, A.R.A. and H.R-S.; Project Administration, A.J.H.A.; Funding Acquisition, A.M.S.

#### Conflicts of interest

The authors declare no conflicts of interest.

#### Data availability

The corresponding author may obtain data validating the findings of this research on an adequate request.

#### Ethics approval

The research proposal has been authorized by the Institutional Review Board (IRB) of AL Nahrain University's College of Medicine (authorization number: 2 on October 14, 2021).

### References

1. Sroka-Tomaszewska J, Trzeciak M. Molecular Mechanisms of Atopic Dermatitis Pathogenesis. *Int J Mol Sci.* 2021;22(8):4130. doi: 10.3390/ijms22084130
2. Hassan ZY, Hassan TY, AbuRaghif AR. Evaluation the Effect of Phytosterol Fraction of Chenopodium Murale-in Comparison with Tacrolimus on Mice Induced Atopic Dermatitis. *Iraqi Journal of Pharmaceutical Sciences* 2023;32(1):84-91. doi: 10.31351/vol32iss1pp84-91
3. Aman S, Nadeem M, Mahmood K, Ghafoor MB. Pattern of skin diseases among patients attending a tertiary care hospital in Lahore, Pakistan. *Journal of Taibah University Medical Sciences.* 2017/10/01/ 2017;12(5):392-396. doi: 10.1016/j.jtumed.2017.04.007
4. Baidya A, Mabalirajan U. Pathogenesis and management of atopic dermatitis: insights into epidermal barrier dysfunction and immune mechanisms. *Explor Asthma Aller-*

gy. 2025;3:100973. doi:10.37349/eaa.2025.100973

5. Hassan Z, Hassan T, Abu-Raghif A. Evaluation the Effectiveness of Phenolic Compound of *Salvia frigida* on Induced Atopic Dermatitis in Experimental Mice. *Iraqi Journal of Pharmaceutical Sciences*. 2022;31(1):154-166. doi: 10.31351/vol31iss1pp154-166
6. Lyons JJ, Milner JD, Stone KD. Atopic dermatitis in children: clinical features, pathophysiology, and treatment. *Immunol Allergy Clin*. 2015;35(1):161-183.
7. Moosbrugger-Martinz V, Leprince C, Méchin MC, et al. Revisiting the Roles of Filaggrin in Atopic Dermatitis. *Int J Mol Sci*. 2022;23(10):5318. doi:10.3390/ijms23105318
8. Hameed MF, Kadhim EJ, Abu-Raghif AR. Effect of Topical Artemisinin from *Artemisia Annua* in Comparison with Tacrolimus on Induced Atopic Dermatitis in Mice. *Journal of Global Pharma Technology*. 2020;12(2):447-453.
9. Griffiths CE, van de Kerkhof P, Czarnecka-Operacz M. Psoriasis and Atopic Dermatitis. *Dermatol Ther (Heidelberg)*. 2017;7(1):31-41. doi: 10.1007/s13555-016-0167-9
10. Khafaji AWM, Al-Zubaidy AAK, Farhood IG, Salman HR. Ameliorative effects of topical ramelteon on imiquimod-induced psoriasisform inflammation in mice. *Naunyn Schmiedebergs Arch Pharmacol*. 2024;397(8):6231-6248. doi: 10.1007/s00210-024-03017-7
11. Ridha-Salman H, Shihab EM, Hasan HK, Abbas AH, Khorsheed SM, Ayad Fakhri S. Mitigative Effects of Topical Norfloxacin on an Imiquimod-Induced Murine Model of Psoriasis. *ACS Pharmacol Transl Sci*. 2024;7(9):1-16. doi: 10.1021/acspctsci.4c00152
12. Ali BF, Abu-Raghif AR, Ridha-Salman H, Al-Athari AJH. Vildagliptin topical ointment: an effective treatment for imiquimod-induced psoriasis in mice. *J Mol Histol*. 2025;56(3):143. doi: 10.1007/s10735-025-10416-4
13. Abbas AH, Abbood MS, Ridha-Salman H, Fakhri SA, Abbas ZH, Al-Athari AJH. Suppressive effect of topical moxifloxacin on imiquimod-induced model of psoriasis in mice. *Naunyn Schmiedebergs Arch Pharmacol*. 2025. doi: 10.1007/s00210-025-04317-2
14. Niwa Y, Sumi H, Akamatsu H. An association between ulcerative colitis and atopic dermatitis, diseases of impaired superficial barriers. *J Invest Dermatol*. 2004;123(5): 999-1000.
15. Manna MJ, Abu-raghif A, Muhsin HY. The effect of Niclosamide in acetic acid induce colitis: an experimental study. *Prensa médica argentina*. 2019;309-316.
16. Oubaid EN, Abu-Raghif A, Al-Sudani IM. Ibudilast ameliorates experimentally induced colitis in rats via down-regulation of proinflammatory cytokines and myeloperoxidase enzyme activity. *Pharmacia*. 2023;70(1):187-195. doi: 10.3897/pharmacia.70.e98715
17. Dawood JO, Abu-Raghif A. Labetalol Ameliorates Experimental Colitis in Rat Possibly Through its Effect on Proinflammatory Mediators and Oxidative Stress. Article. *Clinical Laboratory*. 2024;70(2):353-362. doi: 10.7754/ Clin.Lab.2023.230659
18. Zink A, Gensbaur A, Zirbs M, et al. Targeting IgE in Severe Atopic Dermatitis with a Combination of Immunoabsorption and Omalizumab. *Acta Derm Venereol*. 2016;96(1):72-76. doi: 10.2340/00015555-2165
19. Kashiwakura J-i, Otani IM, Kawakami T. Monomeric IgE and mast cell development, survival and function. *Mast Cell Biology: Contemporary and Emerging Topics*. 2011;29-46.
20. Habbas AH, Abu-Raghif AR, Ridha-Salman H, Hussein MN. Therapeutic effect of bosentan on 2, 4-dinitrochlorobenzene (DNCB)-induced atopic dermatitis mouse model. *Arch Dermatol Res* 2025;317(1):436. doi: 10.1007/s00403-025-03955-z
21. Raheem AKR, Abu-Raghif AR, Abbas AH, Ridha-Salman H, Oubaid EN. Quercetin mitigates sepsis-induced renal injury via inhibiting inflammatory and oxidative pathways in mice. *J Mol Histol*. 2025;56(3):184. doi: 10.1007/s10735-025-10442-2
22. Raheem AK, Abu-Raghif AR, Abd-alakhwa SZ. Irbesartan Attenuates Sepsis-Induced Renal Injury In Mice Models. *J Pharm Negat Results*. 2022;662-669. doi: 10.47750/pnr.2022.13.%20S05.104
23. Abdul-Majeed ZM, Al-atrakji MQYMA, Ridha-Salman H. Cranberry extract attenuates indomethacin-induced gastrulcer in rats via its potential atioxidant and anti-inflammatory effects. *J Mol Histol* 2025;56(4):206. doi: 10.1007/s10735-025-10438-y
24. Shareef SM, Khaleel RA, Hameed ZE, Alsaraf KM. The protective effect of zingiber officinale l. Extract on kidney tissues and blood factors of kidney functions after the damage caused by azathioprine. Article. *Science-Rise: Pharmaceutical Science*. 2021;32(4):78-86. doi: 10.15587/2519-4852.2021.239434
25. Salman HR, Alzubaidy AA, Abbas AH, Mohammad HA. Attenuated effects of topical vinpocetine in an imiquimod-induced mouse model of psoriasis. *J Taibah Univ Med Sci*. 2024;19(1):35-53. doi: 10.1016/j.jtumed.2023.09.002
26. Dawood JO, Abu-Raghif A. Moxifloxacin ameliorates clinical disease activity and histopathological changes of acetic acid-induced colitis model in rat possibly through its effect on proinflammatory mediators and oxidative stress. Article. *Adv Life Sci*. 2023;10(S1):106-112. doi: 10.62940/als.v10i0.2135
27. Asal HA, Diajil AR, Al-Asady FM. Salivary Interleukin-6 Level in Iraqi Patients with Oral Lichen Planus receiving Platelet-Rich Plasma Injections. Article. *Al-Kindy College Med J*. 2023;19(2):44-49. doi: 10.47723/kcmj.v19i2.934
28. Asal HA, Diajil AR, Al-Asady FM. Effects of Intraleisional Platelets-Rich Plasma Injections on Oral Lichen Planus Lesions and Salivary Interleukin-8. Article. *Med J Babylon*. 2023;20(3):457-462. doi: 10.4103/MJBL.MJBL\_359\_22
29. Salman HR, Al-Zubaidy AA, Abbas AH, Zigam QA. The

ameliorative effects of topical gemifloxacin alone or in combination with clobetasol propionate on imiquimod-induced model of psoriasis in mice. *Naunyn Schmiedebergs Arch Pharmacol.* 2024;397(1):599-616. doi: 10.1007/s00210-023-02629-9

30. AL-Bairmani RJ, Kadhim HM. Evaluation of Anti-aging Effects of Gemfibrozil on D-galactose induced Aging Mouse Model. *Int J Drug Del Tech.* 2023;13(3):1011-1016. doi: 10.25258/ijddt.13.3.40

31. Abdulhameed OA, Kadhim HM. Exploring the Role of Pleurotus Ostreatus as an Ointment Formulation in Inducing Wound Healing in Mice Skin. Article. *J Pharm Bi allied Sci.* 2024;16:S243-S246. doi: 10.4103/jpbs.jpbs\_480\_23

32. Alhussien ZA, Mossa HAL, Aboot MS. The Effect of L-carnitine on Apoptotic Markers (Annexin V and Clusterrin) in Polycystic Ovarian Syndrome Women undergoing ICSI. Article. *nt J Drug Del Tech.* 2022;12(4):1682-1686. doi: 10.25258/ijddt.12.4.33

33. Khaleel BJ, Ridha-Salman H, Kadhim HM, Hassan OM, Kubba A, Sahib HB. Inhibitory effects of carbohydrazide indole derivative on micro-blood vessel growth using ex vivo, in vivo, and in vitro assays. *In Vitro Cell Dev Biol Anim.* 2025. doi: 10.1007/s11626-025-01019-0

34. Abu-Raghif AR, Ali NM, Farhood IG, Hameed MF, Sahib HB. Evaluation of a standardized extract of Ginkgo biloba in vitiligo remedy. Article. *Asian J Pharm Clin Res.* 2013;6(5):127-130.

35. Al-Asady FM, Omran TZ, Aboot FM. Humoral Immunity Role in Diagnosis of COVID-19 among People Visited to a Tertiary Care Hospital in Hilla City. Article. *Med J Babylon.* 2023;20(3):497-502. doi: 10.4103/MJBL.MJBL\_124\_23

36. Khaleel BJ, Ridha-Salman H, Kadhim HM, Hassan OM, Kubba A, Sahib HB. Anti-angiogenic and anti-oxidant effects of 2-NTI indole derivative vs. suramin in ex vivo, in vivo, and in vitro studies. *Cytotechnology.* 2025;77(1):38. doi: 10.1007/s10616-024-00701-7

37. Bieber T. Atopic dermatitis: an expanding therapeutic pipeline for a complex disease. *Nat Rev Drug Discov.* 2022;21(1):21-40. doi: 10.1038/s41573-021-00266-6

38. Puterman A, Green R. Topical and systemic pharmacological treatment of atopic dermatitis. *S African Med J.* 2014;104(10):716-719.

39. Alsatari ES, AlSheyab N, D'Sa JL, et al. Effects of argan spinosa oil in the treatment of diaper dermatitis in infants and toddlers: A quasi-experimental study. *J Taibah Univ Med Sci.* 2023;18(6):1288-1298. doi: 10.1016/j.jtumed.2023.05.008

40. Pandaleke TA, Handono K, Widasmara D, Susanti H. The immunomodulatory activity of Orthosiphon aristatus against atopic dermatitis: Evidence-based on network pharmacology and molecular simulations. *J Taibah Univ Med Sci.* 2024;19(1):164-174. doi: 10.1016/j.jtumed.2023.10.005

41. Souto EB, Dias-Ferreira J, Oliveira J, et al. Trends in atop dermatitis-from standard pharmacotherapy to novel drug delivery systems. *Int J Mol Sci.* 2019;20(22):5659. doi: 10.3390/ijms20225659

42. Davis DMR, Drucker AM, Alikhan A, et al. Guidelines of care for the management of atopic dermatitis in adults with phototherapy and systemic therapies. *J Am Acad Dermatol.* 2024;90(2):e43-e56. doi: 10.1016/j.jaad.2023.08.102

43. Kamata M, Tada Y. Optimal Use of Jak Inhibitors and Biologics for Atopic Dermatitis on the Basis of the Current Evidence. *JID Innov.* 2023;3(3):100195. doi: 10.1016/j.jxjidi.2023.100195

44. Arsenault BJ, Perrot N, Puri R. Therapeutic Agents Targeting Cardiometabolic Risk for Preventing and Treating Atherosclerotic Cardiovascular Diseases. *Clin Pharmacol Ther.* 2018;104(2):257-268. doi: 10.1002/cpt.1110

45. Niedzielski M, Broncel M, Gorzelak-Pabiś P, Woźniak E. New possible pharmacological targets for statins and ezetimibe. *Biomed Pharmacother.* 2020;129:110388. doi: 10.1016/j.bioph.2020.110388

46. Pandor A, Ara R, Tumur I, et al. Ezetimibe monotherapy for cholesterol lowering in 2,722 people: systematic review and meta-analysis of randomized controlled trials. *J Int Medi.* 2009;265(5):568-580.

47. Phan BAP, Dayspring TD, Toth PP. Ezetimibe therapy: mechanism of action and clinical update. *Vascular health and risk management.* 2012;415-427.

48. Cannon CP, Blazing MA, Giuglano RP, et al. Ezetimibe Added to Statin Therapy after Acute Coronary Syndromes. *N Engl J Med.* 2015;372(25):2387-97. doi: 10.1056/NEJMoa1410489

49. Tobaru T, Seki A, Asano R, Sumiyoshi T, Hagiwara N. Lipid-lowering and anti-inflammatory effect of ezetimibe in hyperlipidemic patients with coronary artery disease. *Heart Vessels.* Jan 2013;28(1):39-45. doi: 10.1007/s00380-012-0243-8

50. Cho Y, Kim RH, Park H, Wang HJ, Lee H, Kang ES. Effect of Ezetimibe on Glucose Metabolism and Inflammatory Markers in Adipose Tissue. *Biomedicines.* 2020;8(11):0512. doi: 10.3390/biomedicines8110512

51. Tie C, Gao K, Zhang N, et al. Ezetimibe Attenuates Atherosclerosis Associated with Lipid Reduction and Inflammation Inhibition. *PLoS One.* 2015;10(11):e0142430. doi: 10.1371/journal.pone.0142430

52. Choi JW, Suh DW, Lew BL, Sim WY. Simvastatin/Ezetimibe Therapy for Recalcitrant Alopecia Areata: An Open Prospective Study of 14 Patients. *Ann Dermatol.* 2017;29(6):755-760. doi: 10.5021/ad.2017.29.6.755

53. Ghanim H, Green K, Abuaysheh S, et al. Ezetimibe and simvastatin combination inhibits and reverses the pro-inflammatory and pro-atherogenic effects of cream in obese patients. *Atherosclerosis.* 2017;263:278-286. doi: 10.1016/j.atherosclerosis.2017.06.010

54. Weng Q, Hu T, Shen X, Han J, Zhang Y, Luo J. Ezetimibe Prevents IL-1 $\beta$ -induced Inflammatory Reaction in

Mouse Chondrocytes via Modulating NF- $\kappa$ B and Nrf2/HO-1 Signaling Crosstalk. *Curr Pharm Biotechnol*. 2022;23(14):1772-1780. doi: 10.2174/138920102366220104141521

55. Mäki-Petäjä KM, Booth AD, Hall FC, et al. Ezetimibe and Simvastatin Reduce Inflammation, Disease Activity, and Aortic Stiffness and Improve Endothelial Function in Rheumatoid Arthritis. *JACC*. 2007;50(9):852-858. doi: 10.1016/j.jacc.2007.04.076

56. Mostafa RE, Abdel-Rahman RF. Ezetimibe alleviates acetic acid-induced ulcerative colitis in rats: targeting the Akt/NF- $\kappa$ B/STAT3/CXCL10 signaling axis. *J Pharm Pharmacol*. 2023;75(4):533-543. doi: 10.1093/jpp/rgad013

57. Hon KL, Kung JSC, Ng WGG, Leung TF. Emollient treatment of atopic dermatitis: latest evidence and clinical considerations. *Drugs Context*. 2018;7:212530. doi: 10.7573/dic.212530

58. Drais HK. Design Hybrid Nanogel of Prednisolone for Topical Application, Preparation, Characterization, In-vitro and Ex-vivo Evaluation: Hybrid Nanogel of Prednisolone. *Iranian Journal of Pharmaceutical Sciences*. 2023;19(3):194-207.

59. Drais HK, Ihmedee FH, El-Din MKS, Naser NA. Investigation of biological membrane permeability of nanoemulsion, nanostructured lipid carriers and lipid polymer hybrid nanocarriers based nanogel as transdermal carvedilol delivery system. *Res J Pharm Technol*. 2024;17(10):4947-4952.

60. Ahmed NH, Al-Zubaidy AAK, Qasim BJ. Effect of Topical Dipyridamole Gel in Comparison with Clobetasol on induced Psoriasis in Mice. *Int J Drug Del Tech*. 2021;2(11):524-529. doi: 10.25258/ijddt.11.2.51

61. Almudaris SA, Gatea FK. Effects of topical Ivermectin on imiquimod-induced Psoriasis in mouse model – Novel findings. *Pharmacia*. 2024;71:1-14. doi: pharmacia.71.e114753

62. Hassan RF, Kadhim HM. Comparative Effects of Phenolic Extract As an Ointment Dosage Form in Inducing Wound Healing in Mice and  $\beta$ -sitosterol in Experimentally Induced Acute Wound Healing in Mice. *J Phar Neg Res*. 2022;13(3):194-203. doi: 10.47750/pnr.2022.13.03.031

63. Kadhim H, Gatea F, Raghif AA, Ali K. Role of Topical Ritodrine Hydrochloride in Experimentally Induced Hypertrophic Scar in Rabbits. *Iraqi J Pharm Sci*. 2022;31(2):260-270. doi: 10.31351/vol31iss2pp260-270

64. Mohammed SS, Kadhim HM, AL-Sudani IM, Musatafa WW. Anti-inflammatory Effects of Topically Applied Azilsartan in a Mouse Model of Imiquimod-induced Psoriasis. *Int J Drug Deliv Technol*. 2022;3(12):1249-1255. doi: 10.25258/ijddt.12.3.53

65. Rajalakshmi G, Damodharan N, Chaudhari V, Pogal J. Formulation and evaluation of clotrimazole and ichthammol ointment. *Int J Pharma Bio Sciences*. 2010;1(4):7-16.

66. Drais HK. Development and Evaluation Essential Oils Nanoemulgel as Human Skin Sanitizer Using Novel Method. *Tur J Phar Sci*. 2024;21(5):456.

67. Abed-Mansoor A, Abu-Raghif AR. Attenuated effects of rivastigmine in induced cytokine storm in mice. *Journal of Emergency Medicine, Trauma & Acute Care*. 2022;2022(3):12. doi: 10.5339/jemtac.2022.ismc.12

68. Yahiya YI, Hadi NR, Abu Raghif A, Al Habooby NGS. Protective effect of IAXO-102 on renal ischemia-reperfusion injury in rats. *J Med Life*. Apr 2023;16(4):623-630. doi: 10.25122/jml-2022-0280

69. Ali KH, Al-Jawad FH, Kadhim HM. The possible hepatoprotective effects of combination of an oral krill oil and silymarin against carbon tetrachloride (Ccl4)-induced liver fibrosis/ injury in white albino rats: Histopathological, and biochemical studies. *Int J Drug Del Tech*. 2021;11(3):827-833. doi: 10.25258/ijddt.11.3.29

70. Al-Asady FM, Al-Saray DA. Impacts administration of rifampicin on sperm DNA integrity and male reproductive system parameters in rats. Article. *Res J Pharm Technol* 2021;14(9):4897-4902. doi: 10.52711/0974-360X.2021.00851

71. Oubaid EN, Abu-Raghif AR, Al-Sudani IM. Phytochemical Screening and Antioxidant Activity of Uncaria tomentosa Extract: In Vitro: and: In Vivo: Studies. *Med J Babylon*. 2023;20(1):136-142.

72. Abbas AH, Abbas ZH, Ridha-Salman H, Jabar HE, Abd AH. The attenuated effects of Topical Empagliflozin on Imiquimod-induced Model of Psoriasis in Mice *J Trop Life Sci*. 2024;14(3):459-468. doi: 10.11594/jtls.14.03.03

73. Khorsheed SM, Abu-Raghif A, Ridha-Salman H. Alleviative Effects of Combined Topical Melatonin and Rutin on Imiquimod-Induced Psoriasis Mouse Model. 10.3897/pharmacia.71.e128832. *Pharmacia*. 2024;71:1-13. doi: 10.3897/pharmacia.71.e128832

74. Luty RS. The Effect of Moringa oleifera on High Fat Diet and Streptozotocin Induced Diabetic Rats: The Antihyperglycemic Effect of Moringa oleifera. *Iran J Phar Sci*. 2021;17(2):11-24.

75. Luty RS, Al-Zubaidy AA, Malik AS, Ridha-Salman H, Abbas AH. Protective effect of orientin on diabetic nephropathy in rat models of high-fat diet and streptozotocin-induced diabetes. *Naunyn-schmiedeberg's Arch Pharm*. 2025. doi: 10.1007/s00210-025-03949-8

76. Muhammad SM, Al-Anbari LA, Abood MS. Evaluating Endometrial Thickness at Antagonist Starting Day as an Adjuvant Criterion to Decide GnRH Initiation in Flexible Antagonist Protocols. Article. *HIV Nursing*. 2022;22(2):2073-2081. doi: 10.31838/hiv22.02.395

77. Thongsahuan S, Fonghui L, Kaewfai S, Srinoun K. Precision and accuracy of the Mindray BC-5000Vet hematology analyzer for canine and feline blood. *Vet Clin Pathol*. 2020;49(2):207-216. doi: 10.1111/vcp.12861

78. Sharquie KE, Turki KM, Abu-Raghif AR, Al-Geboori HG. Oxidative stress in patients with premature hair grayness. *Saudi Med J*. 2005;26(8):1310-1311.

79. Kadhim HM. Antiinflammatory and antihyperlipidemic effect of adjuvant cinnamon in type 2 diabetic patients.

*Int J Pharm Sci Rev Res.* 2016;41:88-98.

- 80. Mekkey sm, Abu raghifar, Alkafaji ha-R, Hadi nr. The anti-Parkinson effects of Liraglutide in rat model of Rotenone induced Parkinsonism. *Int J Pha Res.* 2020;12(2):3695-3706. doi: 10.31838/ijpr/2020.SP2.449
- 81. AbdulAemah MA, Hussein GM, Hussein NY. Association of Insulin-Like Growth Factor-1 Gene Single Nucleotide Polymorphism rs10860860 with Type 2 Diabetes. Article. *Iran J War Pub Health.* 2023;15(3):305-310. doi: 10.58209/ijwph.15.3.305
- 82. Hanifin J, Thurston M, Omoto M, et al. The eczema area and severity index (EASI): assessment of reliability in atopic dermatitis. *Exp Derm.* 2001;10(1):11-18.
- 83. Riedl R, Kühn A, Hupfer Y, et al. Characterization of Different Inflammatory Skin Conditions in a Mouse Model of DNCB-Induced Atopic Dermatitis. *Inflammation.* 2024;47(2):771-788. doi: 10.1007/s10753-023-01943-x
- 84. Hasan AM, Gatea FK. Topical apigenin as a promising therapeutic agent for psoriasis: Evaluating efficacy alone and in combination with clobetasol in an imiquimod-induced model of psoriasis in mice. *ACTA Pharmaceutica Scientia.* 2024;62(3).
- 85. Mansur HJ, Gatea FK. Effects of topical pentoxifylline on induced thermal burn in mic. Article. *IJDPR.* 2021;12(3):299-305. doi:10.25258/ijpq.12.3.26
- 86. Abu-Raghif AR, Mahdi ZA. In vivo Study of the Effect of 5 and 10% Nebivolol Cream on Hair Growth in Mice Models. *Int J Drug Del Technol.* 2022;12(3):1148-1155.
- 87. Voss GT, Oliveira RL, de Souza JF, et al. Therapeutic and technological potential of 7-chloro-4-phenylselanyl quinoline for the treatment of atopic dermatitis-like skin lesions in mice. *Mater Sci Mater Eng: C.* 2018;84:90-98. doi: 10.1016/j.msec.2017.11.026
- 88. Aal-Aaboda M, Abu Raghif A, Hadi N. Renoprotective Potential of the Ultra-Pure Lipopolysaccharide from Rhodobacter Sphaeroides on Acutely Injured Kidneys in an Animal Model. *Arch Razi Ins.* 2021;76(6):1755-1764.
- 89. Naji ME, Gatea FK, Ali KA, Raghif ARA. Effect of *Convolvulus arvensis* Ethanolic Extract on Testosterone-induced Alopecia in Mice. *INT J D DEL TECH.* 2022;12(3):1070-1075. doi: 10.25258/ijddt.12.3.24
- 90. Hassan RF, Kadhim HM. Exploring the role of phenolic extract as an ointment dosage form in inducing wound healing in mice. *J Pharm Negat Results.* 2022;13(3):186-193. doi: 10.47750/pnr.2022.13.03.030
- 91. Raheem AKK, Abu-Raghif AR, Zigam QA. Cilostazol Protects Against Sepsis-Induced Kidney Impairment in a Mice Model. *J Me Chem Sci.* 2023;6(5):1193-1203. doi: 10.26655/jmchemsci.2023.5.25
- 92. Ghazy DN, Abu-Raghif AR. Effects of Apremilast on Induced Hypertrophic Scar of Rabbits. *Arch Razi Inst.* 2021;76(6):1803-1813. doi: 10.22092/ARI.2021.356195.1800
- 93. Ridha-Salman H, Al-Zubaidy AA, Abbas AH, Hassan DM, Malik SA. The alleviative effects of canagliflozin on imiquimod-induced mouse model of psoriasis-like inflammation. *Naunyn-schmiedeberg's Arch Pharm.* 2024;398:1-21. doi: 10.1007/s00210-024-03406-y
- 94. Kadhim HM, Al-Mosawi AM. Effects of Emodin and Salvinolic Acid on Carbon Tetrachloride (CCl<sub>4</sub>)-induced Lung Fibrosis in Mice Model. *I INT J D DEL TECH.* 2021;11(4):1269-1274. doi: 10.25258/ijddt.11.4.25
- 95. Abbas A, Abd A, Salman H, Abbas Z. The Attenuated Effects of Cimifugin on Imiquimod-Induced Model of Psoriasis in Mice. *Latin American J Pha.* 2023;42(special issue):362-369.
- 96. Jeong H, Shin JY, Kim M-J, Na J, Ju B-G. Activation of Aryl Hydrocarbon Receptor Negatively Regulates Thymic Stromal Lymphopoietin Gene Expression via Protein Kinase C $\delta$ -p300-NF- $\kappa$ B Pathway in Keratinocytes under Inflammatory Conditions. *J Inve Derm.* 2019;139(5):1098-1109. doi: 10.1016/j.jid.2018.11.012
- 97. Yahiya YI, Hadi NR, Abu Raghif A, Qassam H, Al Ha-booby NGS. Role of Iberin as an anti-apoptotic agent on renal ischemia-reperfusion injury in rats. *J Med Life.* 2023;16(6):915-919. doi: 10.25122/jml-2022-0281
- 98. Shihab EM, Kadhim HM. The Impact of Carvedilol on Organ Index, Inflammatory Mediators, Oxidative Stress Parameters and Skin Markers in D-Galactose-Induced Aging Mice. *Int J Drug Del Tech.* 2023;13(3):1017-1023. doi: 10.25258/ijddt.13.3.41
- 99. Ali KH, Al-Jawad FH, Kadhim HM. The possible hepatoprotective effects of "krill oil and silymarin against carbon tetrachloride (CCl<sub>4</sub>)-induced rats model of liver fibrosis: In vivo study". *Research J Phar Tech.* 2021;14(11):5953-5958. doi: 10.52711/0974-360X.2021.01034
- 100. Attarbashee RK, Mammdoh JK, Al-Kazzaz SG. The protective effect of bosentan on methotrexate-induced oral mucositis in rats. *Iraqi J Vet Sci.* 2023;37(3):629-635. doi: 10.33899/IJVS.2023.135827.2536
- 101. Swayeh N, Kadhim H. Effects of methanol extract of *Corchorus olitorius* cultivated in Iraq on high fat diet plus streptozotocin-induced type ii diabetes in rats. *Int J Drug Deliv Technol.* 2022;12(2):754-759. doi: 10.25258/ijddt.12.2.51
- 102. Attarbashee RK, Hamodat HF, Mammdoh JK, Ridha-Salman H. The Possible effect of Bosentan on the methotrexate-induced salivary gland changes in male rats: histological and Immunohistochemical study. *Toxicology Research.* 2025;14(1):tfaf007. doi: 10.1093/toxres/tfaf007
- 103. Thammer MR, Sahib HB, Ridha-Salman H. Skin Healing Potential of Bioactive Components From *Lycoperdon lividum* Mushroom Versus  $\beta$ -Sitosterol in Rat Model of Burn Wounds. *Microscopy Research and Technique.* 2025;88:1-53. doi: 10.1002/JEMT.24864
- 104. Luty RS, Abbas SF, Haji SA, Ridha-Salman H. Hepatoprotective effect of catechin on rat model of acetaminophen-induced liver injury. *Comp Clin Pathol.* 2025. doi: 10.1007/s00580-025-03682-x
- 105. Hussein GM, AbdulAemah MA, Aboob AJ, Albeagy MD,

Hussein NY. Evaluation of serum galactin-3 concentration in patients with heart failure. Article. *J Bio Res*. 2023;15:238-243.

106. Abdulbari A, Ali N, Abu-Raghif A, Matloob N, Ridha-Salman H. Impact of Azithromycin on Specific Biochemical Markers and Sebum Composition in Acne Vulgaris Patients. *Arch Dermatol Res*. 2025;317(1):798. doi: 10.1007/s00403-025-04299-4

107. Ali KA, Abu-Raghif AR, Ridha-Salman H. Evaluation of common topical therapeutic agents of plane warts. *Arch Dermatol Res*. 2025;317(1):246. doi: 10.1007/s00403-024-03789-1

108. Sraibit Abbad M, Al-Jawad FH, Anoze AA, Ali Jawad M, Qasim BJ. Antiatherosclerotic effect of L-thyroxine and verapamil combination on thoracic aorta of hyperlipidemic rabbits. Article. *Int J Pharm Sci Rev Res*. 2014;27(1):254-260.

109. Abdullah MA, Abdulhamed WA, Abood MS. Effects of Vaginal Sildenafil Citrate on Ovarian Blood Flow and Endometrial Thickness in Infertile Women Undergoing Intra-uterine Insemination. Article. *Int J Drug Del Tech*. 2021;11(4):1450-1453. doi: 10.25258/ijddt.11.4.56

110. Abhaya Indrayan RKM. Medical Biostatistics. 4th Edition ed. Chapman & Hall/CRC; 2017:1032

111. Hasan HK, AbdElrahman MM, Aldallal AAR, et al. Assessing Pharmacy Students' Knowledge and Awareness Regarding the Rational Utilization of Antibiotics and the Issue of Antimicrobial Resistance in Iraq. *Al-Mustaqlab J Pharm Med Sci*. 2024;2(1):2. doi: 10.62846/3006-5909.1006

112. Riedl R, Kühn A, Rietz D, et al. Establishment and Characterization of Mild Atopic Dermatitis in the DNCB-Induced Mouse Model. *Int J Mol Sci*. 2023;24(15):12325. doi:10.3390/ijms241512325

113. Min G-Y, Kim E-Y, Hong S, et al. *Lycopus lucidus*Turcz ameliorates DNCB-induced atopic dermatitis in BALB/c mice. *Mol Med Rep*. 2021;24(6):827. doi: 10.3892/mmr.2021.12467

114. C. Williams H, Chalmers J. Prevention of Atopic Dermatitis. *Acta Dermato-Venereologica*. 2020;100(12):380-388. doi: 10.2340/00015555-3516

115. Zhang L, Zhang H, Niu X, et al. Liangxue-Qushi-Zhiyang Decoction Ameliorates DNCB-Induced Atopic Dermatitis in Mice through the MAPK Signaling Pathway Based on Network Pharmacology. *ACS Omega*. 2024;9(16):17931-17944. doi: 10.1021/acsomega.3c09218

116. Zhao W, Jiang H, Gu Y, et al. Fangji Dihuang formulation ameliorated DNCB-induced atopic dermatitis-like skin lesions by IL-17 signaling pathway: integrating network analysis and experimental validation. Original Research. *Front Pharmacol*. 2023;14:1220945. doi: 10.3389/fphar.2023.1220945

117. Umar BU, Rahman S, Dutta S, et al. Management of Atopic Dermatitis: The Role of Tacrolimus. *Cureus*. 2022; 14(8):e28130. doi: 10.7759/cureus.28130

118. Kraaijveld R, Li Y, Yan L, et al. Inhibition of T Helper Cell Differentiation by Tacrolimus or Sirolimus Results in Reduced B-Cell Activation: Effects on T Follicular Helper Cells. *Transplant Proc*. 2019;51(10):3463-3473. doi: 10.1016/j.transproceed.2019.08.039

119. Pascual JC, Fleisher AB. Tacrolimus ointment (Protopic) for atopic dermatitis. *Skin Therapy Lett*. 2004;9(9):1-5.

120. Chen S, Liao P, Xi L, et al. The Therapeutic Effect of Tacrolimus in a Mouse Psoriatic Model is Associated with the Induction of Myeloid-derived Suppressor Cells. *Rheumatol Immunol Res*. 2022;3(4):190-197. doi: 10.2478/rir-2022-0034

121. He H, Gao X, Wang X, et al. Comparison of anti-atopic dermatitis activities between DHMEQ and tacrolimus ointments in mouse model without stratum corneum. *Int Immunopharmacol*. 2019;71:43-51. doi: 10.1016/j.intimp.2019.03.015

122. Chang K-T, Lin HY-H, Kuo C-H, Hung C-H. Tacrolimus suppresses atopic dermatitis-associated cytokines and chemokines in monocytes. *J Microbiol Immunol Infect*. 2016;49(3):409-416. doi: 10.1016/j.jmii.2014.07.006

123. Quist SR, Wiswedel I, Doering I, Quist J, Gollnick HP. Effects of Topical Tacrolimus and Polyunsaturated Fatty Acids on In Vivo Release of Eicosanoids in Atopic Dermatitis During Dermal Microdialysis. *Acta Dermato-Venereologica*. 2016;96(7):905-909. doi: 10.2340/00015555-2383

124. Ku JM, Hong SH, Kim SR, et al. The prevention of 2,4-dinitrochlorobenzene-induced inflammation in atopic dermatitis-like skin lesions in BALB/c mice by Jawoon-go. *BMC Complement Altern Med*. 2018;18(1):215. doi: 10.1186/s12906-018-2280-z

125. Pinto LM, Chiricozzi A, Calabrese L, Mannino M, Peris K. Novel Therapeutic Strategies in the Topical Treatment of Atopic Dermatitis. *Pharmaceutics*. 2022;14(12):2767.

126. Pereira U, Boulais N, Lebonvallet N, Pennec J-P, Dorange G, Misery L. Mechanisms of the sensory effects of tacrolimus on the skin. *Br J Dermatol*. 2010;163(1):70-77. doi: 10.1111/j.1365-2133.2010.09757.x

127. Chaudhari ND, Talaniker HV, Gupta S, Gupta A, Deshmukh P, Rizvi A. Topical tacrolimus: A boon to dermatology. *Int J Pharm Biomed Sci*. 2012;3(4):188-192.

128. Suchy D, Łabuzek K, Machnik G, Okopień B. The influence of ezetimibe on classical and alternative activation pathways of monocytes/macrophages isolated from patients with hypercholesterolemia. *Naunyn-schmiedeberg's Arch Pharm*. 2014;387:733-742.

129. Shaw SM, Najam O, Khan U, Yonan N, Williams SG, Filides JE. Ezetimibe and atorvastatin both immunoregulate CD4+ T cells from cardiac transplant recipients in vitro. *Transplant immunology*. 2009;21(3):179-182.

130. Suchy D, Łabuzek K, Pierzchała O, Okopień B. RP-HPLC-UV determination of ezetimibe in serum: method development, validation and application to patients chronically receiving the drug. *Acta Chromatographica*. 2013;25(3):483-502.

131. Bracht L, Caparroz-Assef SM, Magon TFDs, Ritter AMV,

Cuman RKN, Bersani-Amado CA. Topical anti-inflammatory effect of hypocholesterolaemic drugs. *J Pharm Pharmacol.* 2011;63(7):971-975. doi: 10.1111/j.2042-7158.2011.01302.x

132. Qin L, Yang YB, Yang YX, et al. Anti-inflammatory activity of ezetimibe by regulating NF- $\kappa$ B/MAPK pathway in THP-1 macrophages. *Pharmacology.* 2014;93(1-2):69-75. doi: 10.1159/000357953

133. Moon J, Lee S-Y, Na HS, et al. Ezetimibe ameliorates clinical symptoms in a mouse model of ankylosing spondylitis associated with suppression of Th17 differentiation. Original Research. *Front Immunol.* 2022;13. doi: 10.3389/fimmu.2022.922531

134. Krysiak R, Zmuda W, Okopien B. The effect of ezetimibe, administered alone or in combination with simvastatin, on lymphocyte cytokine release in patients with elevated cholesterol levels. *J Intern Med.* 2012;271(1):32-42. doi: 10.1111/j.1365-2796.2011.02394.x

135. Jaafar F, Abu-Raghif A. Ezetimibe Ameliorates Clinical Parameters, Oxidative Stress, and Adhesion Molecules in Experimentally Induced Colitis in Male Rat Models. *Open Access Med Physiol.* 2023;10(4):103-110. doi: 10.24412/2500-2295-2023-4-103-110

136. Jaafar FR, Abu-Raghif A. Comparative treatment of Sulfasalazine+ Ezetimibe combination and Sulfasalazine in a rat model with induced colitis. *J Medi Life.* 2023;16(8):1165.

137. Jaafar FR, Abu-Raghif AR. The Effects of Sulfasalazine and Ezetimibe on Proinflammatory Cytokines in Male Rat with Induced Colitis: A Comparative Study. Article. *Medi J Babylon.* 2024;21(3):681-685. doi: 10.4103/MJBL.MJBL\_393\_23

138. Lattouf C, Jimenez JJ, Tosti A, et al. Treatment of alopecia areata with simvastatin/ezetimibe. *JAAD.* 2015;72(2):359-361. doi: 10.1016/j.jaad.2014.11.006

139. Cervantes J, Jimenez JJ, DelCanto GM, Tosti A. Treatment of alopecia areata with simvastatin/ezetimibe. *Elsevier.* 2018;S25-S31.

140. Loi C, Starace M, Piraccini BM. Alopecia areata (AA) and treatment with simvastatin/ezetimibe: experience of 20 patients. *JAAD.* 2016;74(5):e99-e100.

141. Wang ECE, Christiano AM. The Changing Landscape of Alopecia Areata: The Translational Landscape. *Advances in Therapy.* 2017;34(7):1586-1593. doi: 10.1007/s12325-017-0540-9

142. Moreira FT, Ramos SC, Monteiro AM, et al. Effects of two lipid lowering therapies on immune responses in hyperlipidemic subjects. *Life Sciences.* 2014;98(2):83-87. doi: 10.1016/j.lfs.2014.01.001

143. Fraunberger P, Gröne E, Gröne H-J, Drexel H, Walli AK. Ezetimibe reduces cholesterol content and NF- $\kappa$ B activation in liver but not in intestinal tissue in guinea pigs. *J Inflamm.* 2017;14:1-11.

144. Yagi S, Akaike M, Aihara K-i, et al. Ezetimibe ameliorates metabolic disorders and microalbuminuria in patients with hypercholesterolemia. *J Atheroscler Thromb.* 2010;17(2):173-180. doi: 10.5551/jat.2378

145. Krysiak R, Okopien B. The effect of ezetimibe and simvastatin on monocyte cytokine release in patients with isolated hypercholesterolemia. *J Cardiovasc Pharm.* 2011;57(4):505-512. doi: 10.1097/FJC.0b013e318211703b

146. Kim SH, Kim G, Han DH, et al. Ezetimibe ameliorates steatohepatitis via AMP activated protein kinase-TFEB-mediated activation of autophagy and NLRP3 inflammasome inhibition. *Autophagy.* 2017;13(10):1767-1781. doi: 10.1080/15548627.2017.1356977

147. Yu J, Wang W-n, Matei N, et al. Ezetimibe attenuates oxidative stress and neuroinflammation via the AMPK/Nrf2/TXNIP pathway after MCAO in rats. *Oxid Med Cell Longev.* 2020;2020(1):4717258. doi: 10.1155/2020/4717258

148. Gómez-Garre D, Muñoz-Pacheco P, González-Rubio M, Aragoncillo P, Granados R, Fernández-Cruz A. Ezetimibe reduces plaque inflammation in a rabbit model of atherosclerosis and inhibits monocyte migration in addition to its lipid-lowering effect. *Br J Pharmacol.* 2009;156(8):1218-1227. doi: 10.1111/j.1476-5381.2008.00091.x

149. Luo L, Guo Y, Chen L, Zhu J, Li C. Crosstalk between cholesterol metabolism and psoriatic inflammation. *Rev. Fron Immunol.* 2023;14:1124786. doi:10.3389/fimmu.2023.1124786