

# Interleukin 31 in vitiligo patients with and without pruritus

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## Abstract

**Introduction:** Vitiligo is a prevalent skin disorder characterized by the destruction of melanocytes, leading to depigmented patches across various areas of the body. Interleukin (IL)-31 has been implicated in the development of pruritus and skin inflammation, potentially contributing to cutaneous symptoms. This study measures IL-31 levels in vitiligo patients with and without pruritus, comparing them to healthy controls, and explores the relationship between IL-31 levels, disease activity, and other clinical factors to assess its potential role in the early diagnosis of vitiligo.

**Methods:** Ninety individuals were enrolled in the study and equally divided into three groups: vitiligo, vitiligo with pruritus, and healthy controls. The serum level of IL-31 was measured using the enzyme-linked immunosorbent assay (ELISA).

**Results:** Significant differences in IL-31 levels were observed across all groups. IL-31 levels were highest in vitiligo patients with pruritus, followed by those without pruritus, and lowest in healthy controls, with mean values and standard deviations of  $196 \pm 67.28$ ,  $152.10 \pm 74.39$ , and  $80.03 \pm 32.30$  pg/ml, respectively. In addition, IL-31 levels in serum showed significant differences in relation to disease activity in both vitiligo groups. Positive correlations were found between IL-31 levels and the Vitiligo Area Scoring Index (VASI) and Vitiligo Disease Activity (VIDA) in both patient groups, as well as between IL-31 levels and lesion extent in vitiligo patients without pruritus. In patients with pruritus, IL-31 levels also positively correlated with age and the 5-dimension itch scale score.

**Conclusions:** IL-31 may serve as a crucial marker and play a significant role in the early diagnosis of vitiligo in patients both with and without pruritus.

**Keywords:** vitiligo, IL-31, VASI, VIDA, itching

Received: 16 May 2024 | Returned for modification: 19 July 2024 | Accepted: 30 July 2024

## Introduction

Vitiligo is not only a cosmetic disorder but also a chronic autoimmune skin disease that affects almost 1% to 2% of the world population, resulting in psychological stress and diminished quality of life (1, 2). Vitiligo patients with or without the presence of irritated skin lesions can suffer from itch prior to the appearance of depigmented patches (3).

Previous studies have highlighted the critical role of interleukin (IL)-31 in various inflammatory skin diseases. Epidermal keratinocytes express IL-31 receptors, which are found in both tissue and serum of patients with pruritic inflammatory skin conditions (4–7). Due to its involvement in pruritic diseases, both IL-31 and its receptor have become therapeutic targets aimed at alleviating the negative impacts on life, sleep, and psychological wellbeing (5, 7).

This study measures the concentration of IL-31 in vitiligo patients with and without pruritus in comparison to healthy controls and explores its relationship with disease activity and other clinical parameters to assess its potential role in the early diagnosis of vitiligo.

## Methods

This study is a prospective case control study carried out at the outpatient clinic at the Department of Dermatology, STDs, and Andrology, Faculty of Medicine, Fayoum University Hospital, be-

tween January 2023 and August 2023. The study follows the rules of the 1975 Declaration of Helsinki, and all patients provided written consent at the beginning of the study. Ethical approval was obtained on December 11th, 2022, from the ethical committee of Faculty of Medicine at Fayoum University at its 101st session with approval number M633.

For calculating the sample size, the following equation was used with absolute error / precision of 3.2%, determining that 30 subjects for each group should be included in the study (8):

$$N = \frac{Z_{1-\frac{\alpha}{2}}^2 p (1-p)}{d^2} = 28.5$$

A total of 90 age- and sex-matched subjects were equally divided into three groups: 30 vitiligo patients without pruritus, 30 vitiligo patients with pruritus, and 30 healthy controls. All patients selected were adults and were categorized as non-segmental vitiligo patients with and without pruritus. A full medical history and clinical examination were taken from all subjects, including disease onset, course of the disease, duration of vitiligo, and past and family history of any chronic diseases/drugs/complications.

For both patient groups—vitiligo and vitiligo with pruritus—dermatological examinations were applied, using the Vitiligo Area Scoring Index (VASI) (9) and Vitiligo Disease Activity (VIDA) (10), as well as the percentage of extension of lesions by using the rule of nines (11) and the Koebner phenomenon of the lesion (3). For patients with pruritus, the 5-dimension itch scale (5-D itch scale) was calculated (12).

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Exclusion criteria for the study included pregnant or lactating females; individuals with other inflammatory, autoimmune, or systemic diseases; those with renal or hepatic impairment; preexisting hyperlipidemia; and/or any other skin diseases. Participants that had received topical, systemic, or phototherapy treatments within the previous 2 weeks were also excluded.

For the quantitation of IL-31 in serum, an enzyme-linked immunosorbent assay (ELISA) kit was used, provided by Bioassay Technology Laboratory (catalog no. E3254Hu; Zhejiang, China). A 3 ml venous blood sample was drawn from each participant and placed in a plain tube. Serum was separated by centrifugation at  $4,000 \times g$  and stored at  $-80^\circ\text{C}$  until analysis.

### Data and statistical analysis

Data were collected and coded into Microsoft Access, and analysis was performed using the Statistical Package for the Social Sciences (SPSS) software version 17.0 on Windows 8.1 (SPSS Inc., Chicago, IL, USA). The Kolmogorov–Smirnov test was used to assess the normality of data distribution. For normally distributed parametric data, Student's *t*-test was employed to compare measures between two independent groups, and a one-way analysis of variance was used for comparing more than two independent groups. For non-parametric data, the Mann–Whitney *U* test was utilized to compare outcomes between two independent groups. The bivariate Pearson correlation test was applied to evaluate associations between different variables, with a two-tailed significance level used for testing.

**Table 1** | Descriptive and clinical data of patient groups.

Parameters	Vitiligo patients, <i>n</i> (%)	
	Without pruritus	With pruritus
Disease onset		
Sudden	6 (20.0%)	7 (23.3%)
Gradual	24 (80.0%)	23 (76.7%)
Course		
Stationary	3 (10.0%)	0 (0.0%)
Progressive	27 (90.0%)	30 (100.0%)
Koebner phenomenon		
None	21 (70.0%)	23 (76.7%)
Type 1	9 (30.0%)	7 (23.3%)
Number of lesions		
Single	2 (6.7%)	4 (13.3%)
Multiple	28 (93.3%)	26 (86.7%)
Duration of disease, years (median and range)	3.50 (0.08–25)	3 (0.16–15)
Extent of affected skin (median and range)	3.50 (0.5%–22.0%)	2 (1%–10.0%)
VASI score (median and range)	2.15 (0.45–10.8)	1.24 (0.23–10)
VIDA score (median and range)	3 (0–4)	4 (0–4)
5-D itch score (median and range)	–	10 (6–18)

VASI = Vitiligo Area and Severity Index, VIDA = Vitiligo Disease Activity Score, 5-D itch score = 5-dimension itch score. The chi-squared test was used.

**Table 2** | Relation between concentration level of interleukin-31 in serum with demographic and clinical data among vitiligo patients.

Parameters	Vitiligo without pruritus (pg/ml)	<i>p</i> -value	Vitiligo with pruritus (pg/ml)	<i>p</i> -value
Sex				
Female	158.23 ± 77.40	0.266	191.26 ± 67.89	0.930
Male	135.25 ± 67.19		204.18 ± 68.68	
Disease onset				
Sudden	89.83 ± 47.08	<b>0.019</b>	198.14 ± 81.32	0.340
Gradual	167.67 ± 72.33		195.35 ± 64.51	
Course				
Stationary	113.00 ± 105.69	0.351	–	–
Progressive	156.44 ± 71.54		196.00 ± 67.28	
Koebner phenomenon				
None	159.81 ± 80.08	0.068	194.09 ± 69.38	0.553
Type 1	134.11 ± 59.24		202.29 ± 64.59	
Number of lesions				
Single	159.50 ± 96.87	0.940	175.25 ± 77.48	0.622
Multiple	151.57 ± 74.78		199.19 ± 66.72	

Data are shown as mean and SD. Student's independent *t*-test was used. *P*-values in bold are statistically significant ( $p < 0.05$ ).

## Results

All individuals included in the study were matched for age and sex. According to the demographic data, the results showed no statistically significant difference in age across the three groups, with mean ages and standard deviations of  $40.50 \pm 11.63$  years for vitiligo patients,  $36.17 \pm 14.52$  years for vitiligo patients with pruritus, and  $38.13 \pm 13.10$  years for healthy controls ( $p > 0.05$ ). Table 1 provides a full description of the demographic and clinical data for the patient groups.

Upon evaluating patients for the Koebner phenomenon, it was observed that the affected lesions varied among vitiligo patients, with the trunk and hands being the most commonly affected areas, followed by the limbs. Among patients with pruritus, the trunk and upper limbs were the most frequently affected sites, followed by the hands (either right, left, or both).

The serum concentration of IL-31 was assessed across all patient groups. The results revealed that IL-31 levels were generally higher in vitiligo patients compared to healthy controls. Specifically, a statistically significant difference was found between the vitiligo patients and controls, with mean IL-31 levels of  $152.10 \pm 74.39$  pg/ml and  $80.03 \pm 32.30$  pg/ml, respectively ( $p = 0.0001$ ). In addition, there was a highly significant difference between the vitiligo with pruritus group and controls, with mean IL-31 levels of  $196 \pm 67.28$  pg/ml ( $p < 0.0001$ ). A significant difference was also noted between the two vitiligo groups in terms of IL-31 levels ( $p = 0.020$ ; Fig. 1a).

**Concentration level of IL-31 in serum in different characteristics among patient groups**

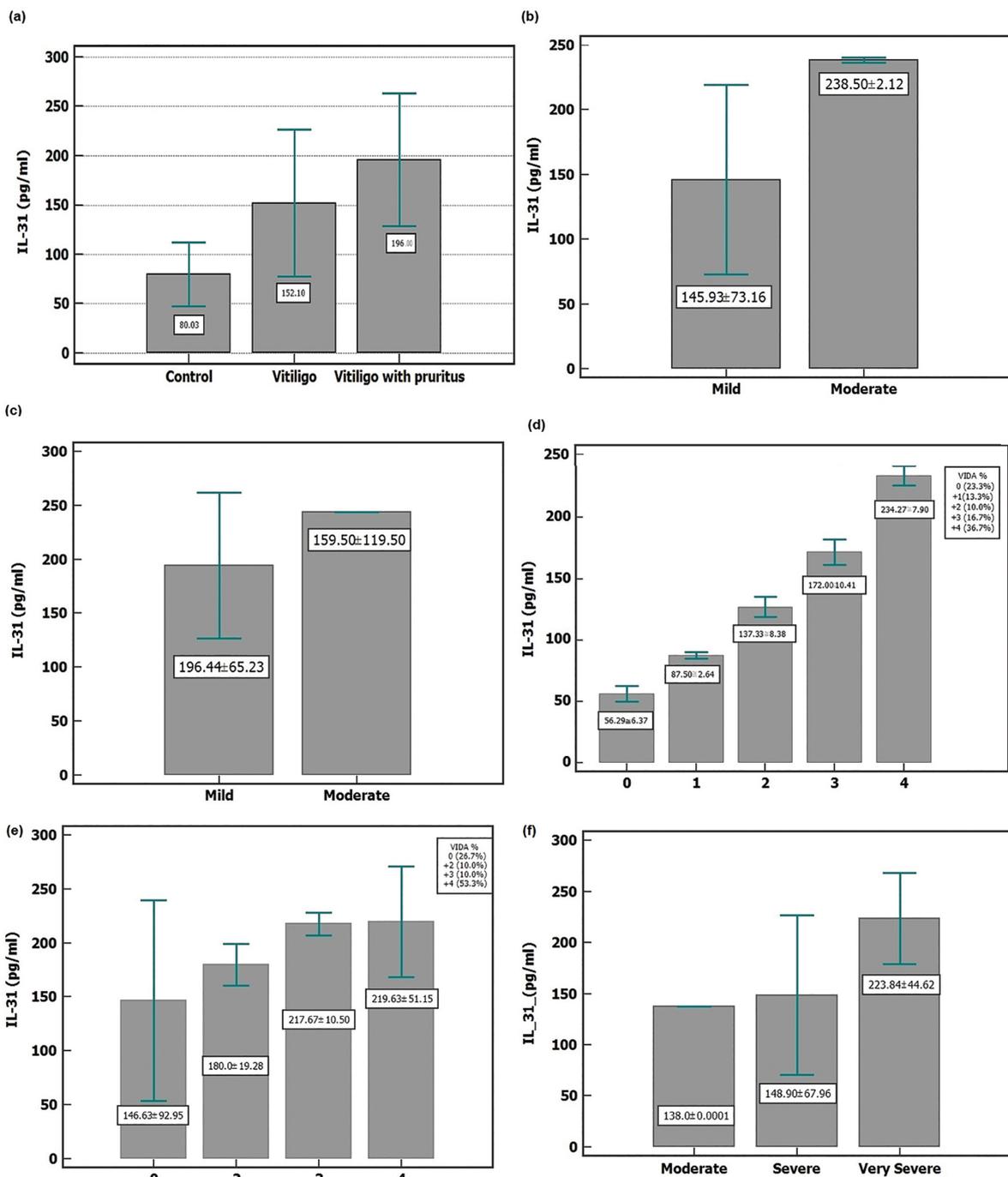
This study evaluated and compared serum IL-31 levels with various clinical parameters among the patient groups. The findings indicated that IL-31 levels tend to be higher in vitiligo patients with a gradual onset of the disease compared to those with a sudden onset, showing a significant difference ( $p = 0.019$ ). However, no significant differences were observed between IL-31 levels and other dermatological assessments in either vitiligo patients or vitiligo patients with pruritus (Table 2).

The patients were further classified based on disease severity using the VASI. In the vitiligo group, 93.3% of patients were classified as having mild disease, and 6.7% were moderate. Among

patients with pruritus, 96.7% were mild, and 3.3% were moderate. When evaluating IL-31 levels in relation to disease severity, the results showed that IL-31 levels were significantly higher in moderate cases compared to mild cases in the vitiligo group ( $p = 0.006$ ; Fig. 1b). Conversely, in patients with pruritus, mild cases had higher IL-31 levels than moderate cases, although this difference was not statistically significant ( $p = 0.234$ ; Fig. 1c).

Regarding the VIDA score, there was a statistically significant difference in IL-31 levels across different activity levels (0, +1, +2, +3, +4) in both patient groups. IL-31 levels tended to increase progressively with greater disease activity (Figs. 1d, 1e).

For the itching score, patients were categorized into three sub-groups: moderate (3.3%), severe (33.3%), and very severe (63.4%). IL-31 levels tended to rise with increasing pruritus severity, with



**Figure 1** | Concentration level of interleukin (IL)-31 among the groups studied: a) level of IL-31 among vitiligo, vitiligo with pruritus, and control groups, b) level of IL-31 among vitiligo patients based on disease severity, c) level of IL-31 among vitiligo patients with pruritus based on disease severity, d) level of IL-31 among vitiligo patients based on Vitiligo Disease Activity (VIDA), e) level of IL-31 among vitiligo patients with pruritus based on VIDA, f) level of IL-31 among vitiligo patients with pruritus based on the 5-dimension itch scale score.

a significant difference observed between the severe and very severe groups ( $p = 0.016$ ; Fig. 1f).

**Correlation between serum level of IL-31 and descriptive/clinical data among study groups**

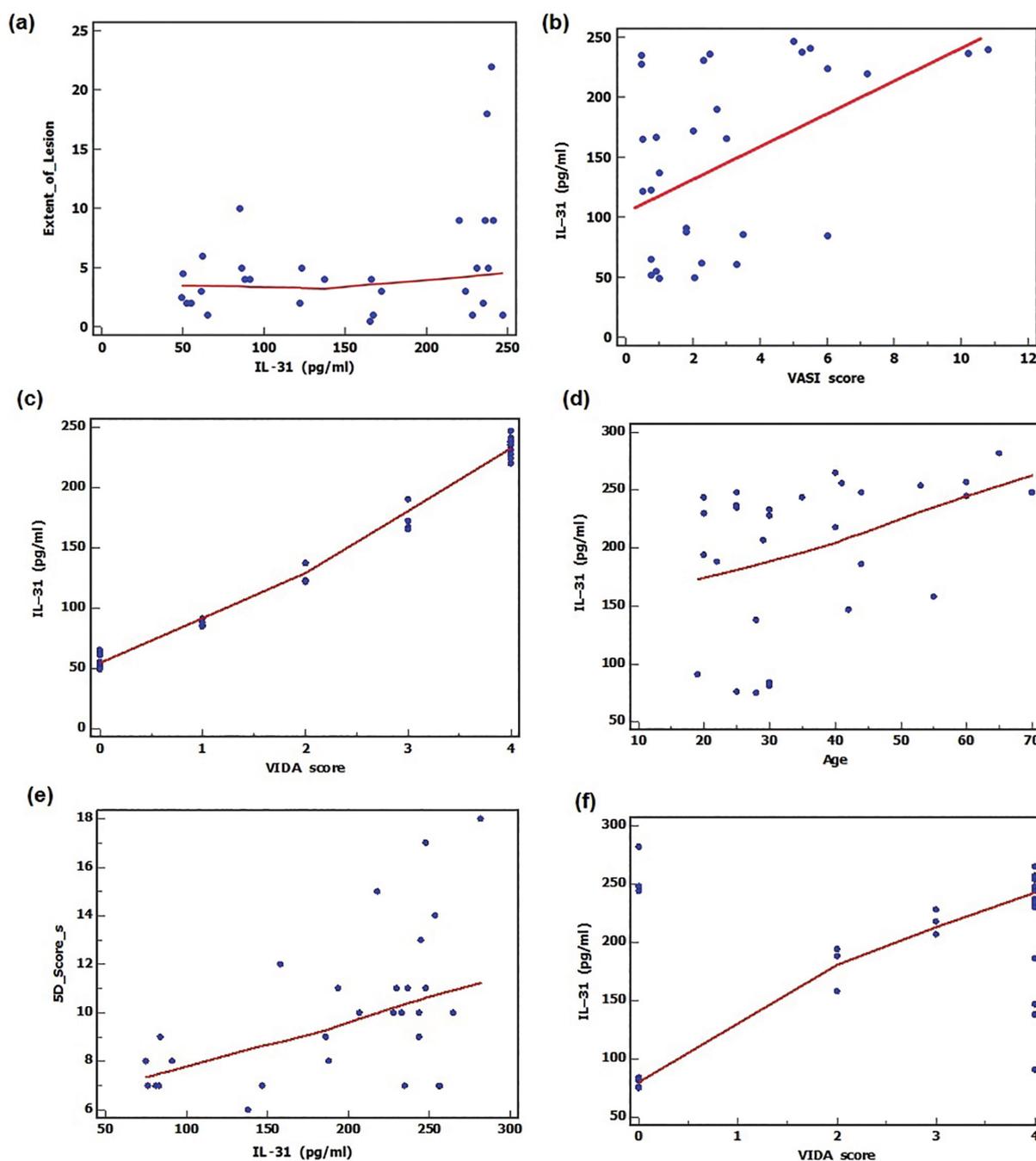
For vitiligo patients, the results indicated a positive correlation between IL-31 levels and the extent of the lesions ( $r = 0.357$ ,  $p = 0.05$ ; Fig. 2a), the VASI score ( $r = 0.4734$ ,  $p = 0.0082$ ; Fig. 2b), and the VIDA score ( $r = 0.9893$ ,  $p < 0.0001$ ; Fig. 2c). However, there was no significant correlation between IL-31 levels and either age or disease duration.

In the pruritus group, the bivariate Pearson correlation analysis revealed positive correlations between IL-31 levels and several

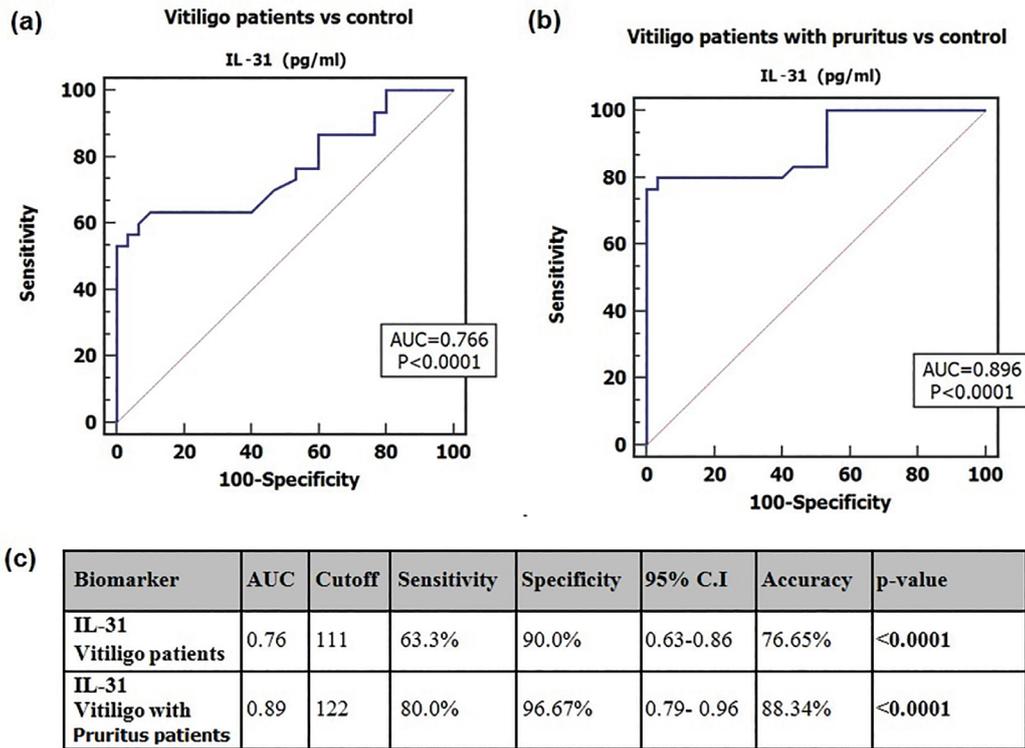
factors. Specifically, IL-31 levels were positively correlated with age ( $r = 0.398$ ,  $p = 0.0292$ ; Fig. 2d), the 5-D itch score ( $r = 0.5291$ ,  $p = 0.0026$ ; Fig. 2e), and the VIDA score ( $r = 0.4767$ ,  $p = 0.0077$ ; Fig. 2f).

**Sensitivity and specificity of IL-31 concentration level in serum among patient groups**

IL-31 levels in serum samples from both vitiligo patients and individuals with pruritus were analyzed to evaluate their potential as a diagnostic marker for vitiligo. The diagnostic accuracy of IL-31 was assessed using receiver operating characteristic (ROC) curve analysis (Fig. 3).



**Figure 2 |** Correlation between interleukin (IL)-31 and descriptive/clinical data among patient groups: a) correlation between the concentration level of IL-31 and the extent of the lesion for vitiligo patients, b) correlation between the Vitiligo Area Scoring Index (VASI) score and IL-31 for vitiligo patients, c) correlation between the Vitiligo Disease Activity (VIDA) score and IL-31 for vitiligo patients, d) correlation between age and IL-31 for vitiligo patients with pruritus, e) correlation between 5-D itch score and IL-31 for vitiligo patients with pruritus, f) correlation between the VIDA score and IL-31 for vitiligo patients with pruritus.



**Figure 3** | Diagnostic and prognostic performances of interleukin (IL)-31 for vitiligo patients with and without pruritus groups: a) receiver operating characteristic (ROC) curve analysis for IL-31 in vitiligo patients, b) ROC curve analysis for IL-31 in vitiligo patients with pruritus, c) sensitivity and specificity analysis for IL-31 among patient groups.

AUS = area under curve, C.I. = confidence interval.

**Discussion**

Vitiligo is a chronic autoimmune hypopigmentary skin disorder that affects 1% to 2% of the global population, with a prevalence rate of 1.2% in Egypt (13, 14). It is primarily characterized by milky white patches with sharply demarcated margins, and it is generally considered an asymptomatic dermatosis. However, pruritus has occasionally been reported in vitiligo patients, with a prevalence of around 20%. Notably, pruritus is associated with active vitiligo in 78.1% of patients (15).

Various studies have demonstrated that IL-31 plays a significant role in inducing pruritus in several chronic skin disorders, such as atopic dermatitis (16–18), contact dermatitis (19), and psoriasis (18). However, to the best of our knowledge, no studies have yet investigated its role in vitiligo.

Therefore, this study measured IL-31 levels in vitiligo patients and explored its relationship with disease activity and severity.

This study found that age and sex were not significantly associated with either vitiligo without or vitiligo with pruritus. The findings are consistent with previous studies, in which authors evaluated the characteristics and prevalence of itching in vitiligo patients and reported that age was not a significant factor (3, 20, 21).

Although females were predominant in this study, no statistically significant difference was found between the groups studied. This may be due to the social stigma associated with the condition and the fact that women are more likely to notice changes in their appearance and seek medical attention sooner than men. Like our findings, most studies on vitiligo have reported a higher prevalence in females, but without a statistically significant difference between males and females (2, 22). However, contrary to our results, some studies in the literature did not demonstrate a gender predilection (21, 23). In addition, other studies have reported that

males were more commonly affected than females (24).

Our findings revealed that IL-31 levels were elevated in vitiligo patients, with a more pronounced increase observed in those with pruritus compared to healthy controls.

Regarding disease severity, we found that IL-31 levels were higher in moderate vitiligo patients compared to those with mild disease. Interestingly, in patients with pruritus, IL-31 levels were higher in mild cases than in moderate ones. In addition, a significant difference was observed in relation to 5-D itch scores, with IL-31 levels increasing as pruritus severity escalated. These results suggest that IL-31 may serve as a valuable marker for assessing disease activity in vitiligo.

The results also revealed positive correlations between IL-31 levels and VASI scores, VIDA scores, and itch intensity as measured by the 5-D itch score. These findings suggest the potential clinical relevance of elevated IL-31 levels in vitiligo patients.

To our knowledge, this is the first study to explore the role of IL-31 in vitiligo patients, both with and without pruritus. While IL-31's involvement has not been previously examined, other interleukins such as IL-17, IL-4, IL-6, and IL-22 have been studied in the context of vitiligo (25, 26).

IL-31 plays a crucial role in skin inflammation, tissue homeostasis, immune defense, and pruritus (27). Prior research has already highlighted its involvement in these processes. This study's findings align with earlier research and provide new insights into IL-31's role in vitiligo, whether pruritus is present or not (26–28).

**Conclusions**

IL-31 may serve as an important marker for the early diagnosis of vitiligo, regardless of the presence of pruritus. Beyond its diagnostic potential, IL-31 could also be a valuable indicator for assessing the severity and activity of the disease.

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