DOI:10.31557/APJCB.2026.11.2.499

RESEARCH ARTICLE

# Evaluation of Serum IL-40 and IL-41 as Integrated Immunometabolic Biomarkers in Iraqi women with Breast Cancer: A Case – Control Study

**Hussein Naeem Abd Ali, Hanaa Addai Ali**

Department of Chemistry, Faculty of Science, University of Kufa, Najaf, Iraq.

## Abstract

**Background:** Breast cancer (BC) is a complex malignancy characterized by the uncontrolled proliferation of the mammary epithelium. Emerging research highlights the pivotal role of novel cytokines in the tumor microenvironment. Specifically, the recently identified immunomodulators, Interleukin-40 (IL-40) and Interleukin-41 (IL-41), have been shown to orchestrate B-cell biology, chronic inflammation, and metabolic homeostasis. These cytokines represent critical mediators that influence the intricate pathological landscape and progression of various malignancies, including breast cancer. Objective: This study aims to quantify serum IL-40 and IL-41 levels in Iraqi women diagnosed with breast cancer and to evaluate their association with clinic-pathological features, disease staging and explore the potential of these cytokines as robust diagnostic and prognostic biomarkers within the clinical setting. **Materials and Methods:** This case-control study involved 180 Iraqi women, aged 45–60 years. The cohort consisted of 90 newly diagnosed breast cancer patients at various clinical stages and 90 age-matched healthy individuals who served as the control group. Venous blood samples were collected from all participants to obtain serum. Serum concentrations of IL-40 and IL-41 were quantified using commercially available enzyme-linked immunosorbent assay (ELISA) kits. Additionally, lipid profiles and liver functional enzymes were assessed via spectrophotometric analysis. **Result:** The study demonstrated that serum IL-40 and IL-41 levels were significantly elevated in breast cancer patients compared to healthy controls ($P < 0.001$), with high concentrations persisting across all clinical stages. Furthermore, patients exhibited a distinct pro-inflammatory and metabolic profile characterized by significant dyslipidemia (increased TC, TG, LDL-C, and VLDL-C), hepatic dysfunction (elevated ALT, AST, and ALP), and significantly reduced HDL-C levels ($P < 0.05$). A strong positive correlation was observed between IL-40 and IL-41 ($r = 0.90$; $P < 0.001$). Both cytokines showed significant positive correlations with all measured parameters, except for a negative correlation with HDL-C. ROC curve analysis revealed that IL-40 and IL-41 possess high diagnostic potential, with sensitivities of 92.0% and 74.5%, specificities of 88.5% and 70.0%, and Area Under the Curve (AUC) values of 0.892 and 0.738, respectively (at cut-off values of 107.63 pg/mL and 116.35 pg/mL). **Conclusion:** Serum levels of IL-40 and IL-41 are significantly elevated in breast cancer patients compared to healthy controls, demonstrating a robust association with disease stage and severity. These findings strongly suggest that the IL-40 and IL-41 axis serves as a promising panel of non-invasive biomarkers for the early diagnosis and prognostic monitoring of breast cancer.

**Keywords:** Breast Cancer- different stages- Immunometabolic- IL-40- IL-41- Iraqi population- ALT- AST- ALP- Lipid profiles

*Asian Pac J Cancer Biol*, **11 (2)**, 499-506 Submission Date: 01/17/2026 Acceptance Date: 03/19/2026

## Introduction

Breast cancer (BC) is a heterogeneous malignancy arising from the epithelial lining of the ducts or lobules. As the leading cause of global female cancer mortality, its pathogenesis involves critical genetic and epigenetic alterations in genes such as BRCA1/2 and TP53. These aberrations drive dysregulated proliferation and genomic instability, underlying the complex nature of the disease [1]. In Iraq, breast cancer accounts for approximately one-third of all female malignancies, a trend linked to shifting lifestyles, obesity, and enhanced diagnostic screening

**Corresponding Author:**
Dr. Hanaa Addai Ali
Department of Chemistry, Faculty of Science, University of Kufa, Najaf 54001 Iraq.
Email: hanaa.alsultani@uokufa.edu.iq

[2]. Molecularly, breast tumors are categorized into four primary subtypes Luminal A, Luminal B, HER2-enriched, and Triple-Negative (TNBC). Each subtype exhibits distinct biological behaviors and clinical trajectories, necessitating the identification of novel biomarkers to refine therapeutic response and prognostic accuracy [3]. The clinical spectrum of breast cancer encompasses acute manifestations, such as nipple discharge, cutaneous alterations, and axillary lymphadenopathy, alongside chronic sequelae in survivors. These long-term complications, including secondary lymphedema, chronic pain syndromes, and cancer-related fatigue, significantly impact the quality of life and necessitate comprehensive management strategies [4, 5]. Despite significant screening advancements, early detection remains clinically challenging, particularly in women with dense breast tissue. In such cases, the diagnostic sensitivity of mammography is often compromised, necessitating the use of magnetic resonance imaging (MRI) or contrast-enhanced techniques [6] .Given that the median age of onset coincides with postmenopausal years, understanding the interplay between hormonal decline, systemic inflammation, and metabolic dysregulation is essential [7].

Interleukin-40 (IL-40), encoded by C17orf99, is a recently identified cytokine predominantly secreted by activated B-cells, neutrophils, and T-cells under inflammatory and metabolic stress [8]. B-cells are the primary sources of IL-40; upon activation by IL-4 and CD40 ligand, they robustly secrete this cytokine. IL-40 is essential for antibody class switching and mucosal immunity, serving as a key mediator of the adaptive immune response within the tumor microenvironment [9].

Emerging evidence positions chronic inflammation as a driver of breast carcinogenesis, where dysregulated IL-40 signaling may facilitate tumor progression via immunosuppressive and pro-inflammatory pathways [10]. Elevated serum IL-40 levels have been documented in various malignancies and autoimmune disorders, suggesting its role as a pivotal mediator in the tumor microenvironment [11]. IL-40 may drive tumor progression by orchestrating immunosuppressive and pro-inflammatory pathways. Its expression is significantly upregulated under chronic inflammatory and metabolic stress, both of which are critical determinants in the clinical trajectory of breast cancer [12]. Among the novel cytokines recently implicated in oncological research, Interleukin-40 (IL-40) and Interleukin-41 (IL-41) emerge as pivotal mediators that potentially bridge the complex intersections of immune modulation, systemic metabolism, and tumor microenvironment dynamics [13, 14].

Similarly, Interleukin-41 (IL-41) also designated as Meteorin-like (Metrnl) functions as a pleiotropic cytokine synthesized by M2 macrophages, epithelial cells, and adipocytes. Secreted in response to tissue injury and metabolic stress, IL-41 acts as a molecular bridge linking oncogenesis with systemic metabolic and cardiovascular impairments [15]. The expression of IL-41 is dynamically modulated by hypoxia, pro-inflammatory cytokines, thermal stress, and tissue injury, thereby underscoring its pivotal role as a multifaceted, stress-responsive cytokine within the physiological and pathological landscape [16]. IL-41 exhibits distinct bifunctional regulatory roles: it promotes the secretion of anti-inflammatory cytokines, such as IL-10 and TGF-β, while simultaneously facilitating tissue remodeling, fibrosis, and angiogenesis. These physiological processes are frequently co-opted by malignant cells to consolidate the immunosuppressive landscape of the tumor microenvironment, thereby enhancing survival and immune evasion [17, 18].

## Materials and Methods

*Study Design and Participants*

This case-control study was conducted between May 2025 and October 2025 at the Oncology and Hematology Center in Najaf Al-Ashraf, Iraq. The study enrolled 180 female participants, aged 45–60 years, categorized into two groups: 90 newly diagnosed breast cancer (BC) patients and 90 age-matched healthy controls. The BC cohort was stratified according to TNM staging into Stage I (n=25), Stage II (n=25), Stage III (n=20), and Stage IV (n=20). Clinical diagnosis was rigorously confirmed through standardized mammography, ultrasonography, magnetic resonance imaging (MRI), and histological validation at the oncology center.

*Exclusion Criteria*

The study excluded Women who were pregnant, or presented with autoimmune disorders, microbial infections, or chronic conditions (e.g., hypertension, cardiovascular, renal, or thyroid diseases) were excluded. Additionally, those on hormonal medications or with a recent history of surgical intervention were omitted to ensure data accuracy.

*Data and Sample Collection*

Demographic and clinical parameters, including TNM staging and BMI was calculated as weight (kg)/height ($m^2$) (Keys et al., 1972). were documented via questionnaires and medical records. Prior to treatment, 5 mL of fasting blood was collected and centrifuged at 3000 rpm for 10 minutes; serum was then stored at –80°C for analysis.

*Immunological and Routine Assays*

Serum IL-40 and IL-41 levels were quantified using ELISA kits (Pars Biochem, China) following the manufacturer's protocol. Liver function markers (AST, ALT, and ALP) were measured spectrophotometrically using a Roche/Hitachi Cobas c analyzer. Lipid profiles, including TC, TG, and HDL-C, were determined via enzymatic colorimetric assays (Emclab, Germany), while LDL-C and VLDL-C were calculated using the Friedewald equation (Lewis, 1973).

*Statistical Analysis*

Data were analyzed using IBM SPSS Statistics 27. Normality was assessed via the Kolmogorov–Smirnov test. Continuous variables were expressed as mean ±

SD, with group comparisons performed using Student's t-test or one-way ANOVA. Correlations were evaluated using Pearson's coefficient (r). The diagnostic potential of biomarkers was determined by ROC curve analysis and Area Under the Curve (AUC) to identify optimal sensitivity and specificity. Statistical significance was defined as a two-tailed $p < 0.05$.

*Ethical Approval*

The study adhered to the Declaration of Helsinki and was approved by the Institutional Review Board (IRB) of the College of Medicine, University of Kufa (No. MEC 133; April 10, 2025). Written informed consent was obtained from all participants prior to enrollment.

## Results

The study included 90 patients with breast cancer and 90 apparently healthy controls. Patients were categorized into four groups based on disease stage: Stage I (n=25), Stage II (n=25), Stage III (n=20), and Stage IV (n=20). Compared to the control. Breast cancer (BC) patients exhibited significantly elevated serum levels of IL-40 (133.11 vs. 100.21 pg/mL; $p < 0.0001$) and IL-41 (136.08 vs. 106.84 pg/mL; $p < 0.0001$) compared to the control group. Furthermore, both cytokines demonstrated a significant progressive increase correlating with advancing disease stages (Stages I–IV). Laboratory analysis (Table 1) revealed significantly higher liver enzyme activities (ALT, AST, ALP) and lipid parameters (TC, TG, LDL-C, VLDL-C) in BC patients compared to controls, while HDL-C levels were significantly reduced. Stage-wise comparison (Table 2) showed that while most parameters remained stable across stages, a significant variation was observed between Stage I and Stage IV. Notably, ALP showed the most marked increase when comparing early stages (I and II) against Stage IV.

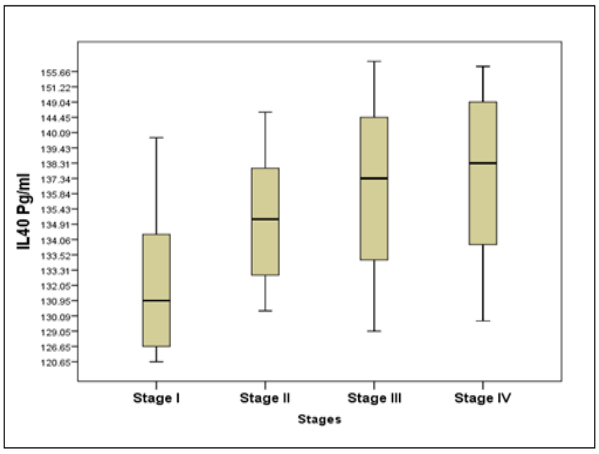


Figure 1. Comparison of Serum IL-40 Levels in BC Patients' stages

Correlation studies (Tables 3 and 4) identified a significant positive correlation between serum IL-40/IL-41 levels and elevated liver enzymes and pro-atherogenic lipids. Conversely, a significant negative correlation was observed with HDL-C, suggesting a link between cytokine dysregulation and metabolic stress. ROC curve analysis (Table 5) confirmed the diagnostic efficacy of both biomarkers. IL-40 demonstrated superior diagnostic accuracy with an AUC of 0.892 (95% CI: 0.829–0.955). At a cut-off of 107.63 pg/mL, IL-40 yielded 92.0% sensitivity and 88.5% specificity. In comparison, IL-41 showed moderate potential with an AUC of 0.738 (95% CI: 0.636–0.841), sensitivity of 74.5%, and specificity of 70.0% at a cut-off of 116.35 pg/mL (Figure 1-4).

## Discussion

This study provides the first evidence of significantly elevated serum IL-40 and IL-41 levels in Iraqi breast cancer patients, particularly in advanced stages (III and

Table 1. Clinical and Biochemical Characteristics of Breast Cancer Patients versus Healthy Controls

| Parameter | BC Group Mean ±SD | HC Group (Mean ± SD) | P-value |
|---|---|---|---|
| Number | 90 | 90 | -------- |
| Age (years) | 54.42 ± 5.39 | 53.57 ± 4.5 | 0.32 |
| BMI (kg/m²) | 28.70 ± 3.59 | 28.37 ± 3.77 | 0.664 |
| ALT (IU/L) | 36.68 ± 9.52 | 22.25 ± 4.75 | < 0.001 |
| AST (IU/L) | 30.40 ± 7.69 | 17.46 ± 4.36 | < 0.001 |
| ALP (IU/L) | 122.86 ± 29.10 | 88.42 ± 22.83 | < 0.001 |
| TG (mg/dL) | 146.14 ± 27.88 | 92.35 ± 18.43 | < 0.001 |
| TC (mg/dL) | 195.46 ± 38.33 | 154.91 ± 21.56 | < 0.001 |
| LDL-C (mg/dL) | 128.31 ± 37.95 | 94.61 ± 22.63 | < 0.001 |
| HDL-C (mg/dL) | 37.92 ± 5.39 | 42.27 ± 6.21 | < 0.001 |
| VLDL-C (mg/dL) | 32.21 ± 5.57 | 18.47 ± 3.68 | < 0.001 |
| IL-40(pg/mL) | 130.11 ± 14.39 | 100.21 ± 13.98 | < 0.001 |
| IL-41(pg/mL) | 141.08 ± 18.58 | 106.84± 12.85 | < 0.001 |

Data are presented as Mean ± SD (Standard Deviation). BC: Breast Cancer; HC: Healthy Controls; BMI: Body Mass Index; ALT: Alanine Aminotransferase; AST: Aspartate Aminotransferase; ALP: Alkaline Phosphatase; TC: Total Cholesterol; TG: Triglycerides; HDL-C: High-Density Lipoprotein Cholesterol; LDL-C: Low-Density Lipoprotein Cholesterol; VLDL-C: Very Low-Density Lipoprotein Cholesterol; IL-40: Interleukin-40; IL-41: Interleukin-41.

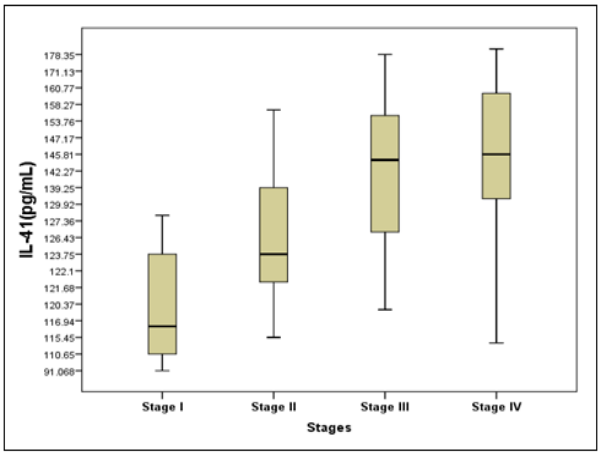


Figure 2. Comparison of Serum IL-41 Levels in BC Patients' stages

IV). These findings suggest a synergistic role within the tumor microenvironment, where IL-40 promotes a pro-inflammatory niche through B-cell dysregulation, and IL-41 acts as a pivotal immunometabolic regulator. The positive correlation between these cytokines and disease progression likely reflects metabolic reprogramming and immune evasion strategies. Furthermore, the observed elevation in patients aged ≥ 45 years aligns with age-associated immune dysregulation, positioning the IL-40/IL-41 axis as a promising biomarker for personalized immunometabolic therapies [19, 20]. Interleukin-40 (IL-40/C17orf99) and Interleukin-41 (IL-41/Metrnl) represent a novel class of cytokines, recently characterized in contrast to classical mediators such as IL-6 and TNF-α. Emerging evidence increasingly positions these molecules at the critical intersection of chronic inflammation, immune dysregulation, metabolic remodeling, and malignancy. Consequently, their involvement in breast cancer pathophysiology warrants focused investigation to elucidate their potential as diagnostic or therapeutic targets [21, 22]. In previous studies suggests a shared pathogenic axis between breast cancer, chronic inflammation, and autoimmune disorders. Analogous to IL-6 and IL-10 which are well established in promoting tumor growth IL-40 may exert similar tumor-supportive effects, reinforcing the concept that oncogenesis and autoimmunity converge through overlapping cytokine networks [23, 24].

Prior studies have shown IL-40 consistently exhibits pro-inflammatory characteristics and appears to increase with tumor progression, IL-40 has been linked to amplification of IL-6 and IL-17 signaling loops and to cardiovascular risk markers [25]. whereas IL-41 displays immunomodulatory properties and may decline in advanced or chronic inflammatory states, while the

Table 2. Comparison of Biochemical Parameters across Different Clinical Stages (I-IV) of Breast Cancer Patients.

| | | BC Patients Stages (n.= 90) (Mean ± SD) | | | | | |
|---|---|---|---|---|---|---|---|
| Variables | | Stage I | | | Stage II | Stage III | Stage IV |
| Number (%) | | 25 (27.8 %) | | | 25 (27.8 %) | 20 (22.2%) | 20 (22.2%) |
| Age (years) | | 51.75 ± 5.54 | | | 53.41 ± 4.66 | 55.20 ± 5.50 | 56.54± 5.85 |
| | p-value: | a) 0.001 | b) 0.001 | c) 0.002 | d) 0.001 | e) 0.001 | h) 0.001 |
| BMI (kg/m²) | | 29.12 ± 3.54 | | | 28.75 ± 3.25 | 28.39 ± 3.58 | 27.98 ± 3.89 |
| | p-value: | a) 0.261 | b) 0.167 | c) 0.110 | d) 0.277 | e) 0.119 | h) 0.197 |
| ALT(IU/L) | | 18.3 ± 2.11 | | | 22.13± 2.80 | 30.89 ± 3.68 | 39.27± 3.55 |
| | p-value: | a) 0.151 | b) 0.12 | c) 0.001 | d) 0.231 | e) 0.141 | h) 0.21 |
| AST (IU/L) | | 26.96 ± 3.33 | | | 31.19 ± 5.74 | 36.21 ± 7.39 | 43.14 ± 7.20 |
| | p-value: | a) 0. 194 | b) 0.123 | c) 0.001 | d) 0.094 | e) 0.11 | h) 0.15 |
| ALP (IU/L) | | 105.66 ± 8.21 | | | 115.58± 12.50 | 122.7 ±10.62 | 135.9 0± 14.46 |
| | p-value: | a) 0.19 | b) 0.09 | c) 0.001 | d) 0.061 | e) 0.001 | h) 0.11 |
| TG (mg/dL) | | 114.04 ± 16.01 | | | 125.01 ± 8.10 | 142.76± 9.25 | 168.84 ± 6.71 |
| | p-value: | a) 0.138 | b) 0.124 | c) 0.01 | d) 0.36 | e) 0.08 | h) 0.445 |
| TC (mg/dL) | | 173.64 ± 31.81 | | | 187.86 ± 10.92 | 201.16 ± 32.34 | 211.66 ± 34.40 |
| | p-value: | a) 0. 154 | b) 0.92 | c) 0.01 | d) 0.094 | e) 0.07 | h) 0.195 |
| HDL-C (mg/dL) | | 41.54 ± 3.01 | | | 40.27 ± 3.04 | 35.34 ± 4.10 | 34.49± 3.13 |
| | p-value: | a) 0.131 | b) 0.108 | c) 0.01 | d) 0.061 | e) 0.054 | h) 0.375 |
| LDL-C (mg/dL) | | 128.29± 30.46 | | | 133.58± 11.88 | 138.07 ± 31.41 | 146.79 ± 33.54 |
| | p-value: | a) 0.126 | b) 0.102 | c) 0.094 | d) 0.299 | e) 0.106 | h) 0.212 |
| VLDL-C (mg/dL) | | 28.80 ± 3.20 | | | 29.07 ± 2.62 | 32.77 ± 2.85 | 36.36 ± 2.34 |
| | p-value: | a) 0.238 | b) 0.144 | c) 0.01 | d) 0.121 | e) 0.085 | h) 0.545 |
| IL-40 (pg/mL) | | 118.09 ± 13.77 | | | 126.61± 14.31 | 134.64± 14.93 | 139.03± 16.16 |
| | p-value: | a) 0.001 | b) 0.001 | c) 0.001 | d) 0.001 | e) 0.001 | h) 0.001 |
| | | 124.47 ± 16.15 | | | 130.38 ± 16.39 | 137.39 ± 17.57 | 143.69 ± 18.72 |
| IL-41 (pg/mL) | p-value: | a) 0.001 | b) 0.001 | c) 0.001 | d) 0.001 | e) 0.001 | h) 0.001 |

Data are expressed as Mean ± SD. BC: Breast Cancer; Statistical comparisons were conducted using One-Way ANOVA and the Chi-squared ($\chi^2$) test. Significant differences ($p < 0.05$) between disease stages were identified via post-hoc analysis and are denoted by superscript letters: (a) Stage I vs. Stage II; (b) Stage I vs. Stage III; (c) Stage I vs. Stage IV; (d) Stage II vs. Stage III; (e) Stage II vs. Stage IV; (h) Stage III vs. Stage IV.

T able 3. Correlation Analysis between Serum IL-40 Levels and Clinical/Biochemical Parameters in the Breast Cancer Patient Group.

| Parameters | r | p-value |
|---|---|---|
| Age (years) | 0.156 | 0.1418 (NS) |
| BMI (kg/m$^2$) | 0.128 | 0.225 (NS) |
| ALT (IU/L) | 0.626** | 0.001 |
| AST (IU/L) | 0.660** | 0.001 |
| ALP (IU/L) | 0.548** | 0.001 |
| TG (mg/dL) | 0.681** | 0.001 |
| TC (mg/dL) | 0.532** | 0.001 |
| HDL-C (mg/dL) | –0.481** | 0.001 |
| LDL-C (mg/dL) | 0.507** | 0.001 |
| VLDLC (mg/dL) | 0.681** | 0.01 |
| IL-41 (pg/mL) | 0.520** | 0.001 |

r: Pearson correlation coefficient

T able 4. Correlation Analysis between Serum IL-41 Levels and Clinical/Biochemical Parameters in the Breast Cancer Patient Group.

| Parameters | r | p-value |
|---|---|---|
| Age (years) | 0.148** | 0.162 |
| BMI (kg/m$^2$) | 0.001 | 0.991 |
| ALT (IU/L) | 0.631** | 0.001 |
| AST (IU/L) | 0.585** | 0.001 |
| ALP (IU/L) | 0.44** | 0.001 |
| TG (mg/dL) | 0.629** | 0.001 |
| TC (mg/dL) | 0.544** | 0.001 |
| HDL-C (mg/dL) | –0.450** | 0.001 |
| LDL-C (mg/dL) | 0.525** | 0.001 |
| VLDLC (mg/dL) | 0.629** | 0.01 |
| IL-40 (pg/mL) | 0.520** | 0.001 |

r: Pearson correlation coefficient

precise anti- or pro-tumorigenic roles of IL-41 in vascular biology and defined [26].

Extensive evidence from autoimmune diseases supports a direct association between IL-40 and B-cell hyperactivity [27]. In systemic lupus erythematosus (SLE), elevated IL-40 levels correlate with disease activity indices (such as SLEDAI), renal involvement, and autoantibody production, reinforcing the hypothesis that IL-40-driven B-cell dysregulation may contribute to the immune imbalance observed in breast cancer [28] .

Interleukin-40 (IL-40) elevation precedes or parallels inflammation and correlates strongly with disease severity in multiple disorders. In rheumatoid arthritis, heightened IL-40 levels are observed even in at-risk individuals, while in SLE, IL-40 demonstrates high diagnostic sensitivity and specificity for active disease. These findings support its potential utility as a biomarker reflecting inflammatory burden [29, 30]. Interleukin-40 (IL-40) may influence hepatic acute-phase responses and fibrotic pathways, potentially explaining the associations with liver enzyme abnormalities (e.g., ALT and AST) observed in various autoimmune and inflammatory disorders [31]. Its elevation in ankylosing spondylitis, thyroid autoimmunity, and systemic sclerosis further supports a systemic role in immune–metabolic dysregulation, a phenomenon that may be mirrored during tumor progression [32, 33] .

Experimental data confirm that IL-40 is significantly upregulated in breast cancer cells compared with normal mammary epithelium. Functional studies demonstrate that IL-40 enhances proliferation, motility, and immune evasion, reinforcing its role as a prognostic biomarker and a potential therapeutic target linked to the inflammatory tumor microenvironment. These findings are in agreement with a previous study by Hameed & Abed [34].

Although less studied, IL-41 is increasingly recognized as a mediator of inflammatory and metabolic pathology. Previous studies by Duan et al. [35] and Namitokov et al. [36] found that elevated IL-41 levels are associated with vascular calcification, atherogenesis, and adverse lipid profiles, including inverse correlations with HDL-cholesterol, suggesting its participation in immunometabolic risk amplification [35, 36]. Lipid metabolism is essential for membrane synthesis, signaling, and energy storage processes frequently dysregulated in cancer. In the present study shown the interaction between IL-40, IL-41, and lipid metabolism may yield clinically relevant biomarkers for early detection, prognosis, and therapeutic stratification in breast cancer

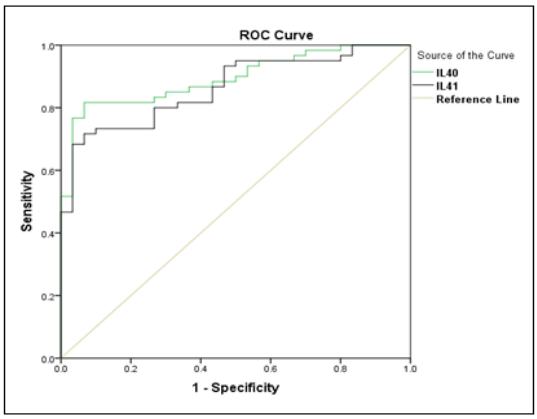


Figure 3. Receiver Operating Characteristic Curve for Breast Cancer Patients vs. Healthy Control.

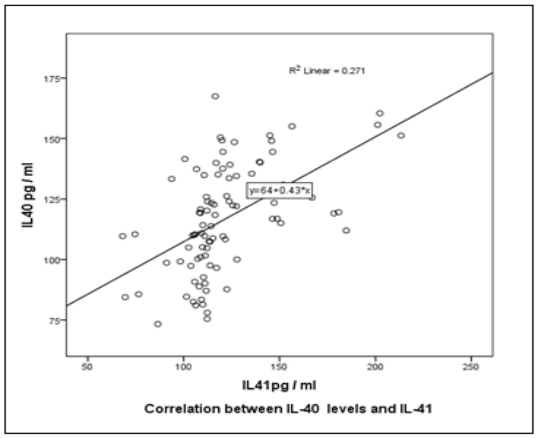


Figure 4. Correlation between IL-40 levels and IL-41 for study groups

Table 5. Receiver Operating Characteristic (ROC) Curve Analysis for IL-40 and IL-41 in Discriminating Breast Cancer Patients from Healthy Controls.

| Variables | Interleukin-40 (pg/mL) | Interleukin-41(pg/mL) |
|---|---|---|
| Cut-off value | 107.63 | 116.35 |
| Specificity | 88.50% | 70.00% |
| Sensitivity | 92.00% | 74.50% |
| Area Under Curve (AUC) | 0.954 | 0.861 |
| Confidence interval 95% | 0.829-0.955 | 0.636- 0.841 |
| P-value | $p < 0.001$ | $p < 0.01$ |

this results agreement with previous studies [37, 38]. A study by Bridgewood et al. [39] illustrated that higher IL-41 levels correlate with disease activity and cartilage degradation in ankylosing spondylitis and psoriatic arthritis. These findings support the role of IL-41 as a marker of inflammatory severity and highlight its potential as a therapeutic target in immune-mediated diseases [39]. Another study by Cen et al. [40] demonstrated that IL-41 elevation is linked to disease exacerbation in chronic obstructive pulmonary disease, myasthenia gravis, and Kawasaki disease, as well as parasitic infections such as hydatid cyst disease. This further underscores its role as a general marker of inflammatory intensity and disease progression [40]. Direct measurements of IL-41 in breast cancer remain scarce. However, the consistent elevation of other inflammatory cytokines in breast malignancies strongly suggests a broader inflammatory milieu in which IL-41 may play an unrecognized role, thereby justifying targeted investigation [41].

*Limitations and Future Directions*

Despite the significant findings, this study is limited by its specific age range (45–60) and localized Iraqi population, which may affect the generalizability of the results. Furthermore, the case-control design precludes establishing definitive causal relationships. To address these limitations, future research should prioritize multi-center longitudinal studies to monitor IL-40 and IL-41 fluctuations across diverse populations and during various treatment stages, such as chemotherapy and radiotherapy. Such studies are essential to establish standardized clinical cutoffs and validate the prognostic utility of the IL-40/IL-41 axis. Additionally, mechanistic and tissue-based investigations are warranted to explore these cytokines as potential therapeutic targets and integrate them into personalized immunometabolic screening protocols for breast cancer.

In conclusion, the present study demonstrates a significant elevation in serum IL-40 and IL-41 levels among Iraqi women with breast cancer ($p < 0.0001$), identifying this dual-marker panel as a robust indicator of disease presence and progression. Mechanistically, the elevation of IL-40 may be attributed to the chronic inflammatory state and B-cell activation within the tumor microenvironment, where it likely functions as a pro-tumorigenic factor. Concurrently, the upregulation of IL-41 reflects a convergence of immune-metabolic dysregulation and systemic disturbances, such as hepatic dysfunction, which correlate significantly with advanced disease stages. By integrating this immunometabolic signature into clinical practice, the IL-40/IL-41 axis offers a viable strategy for improved disease stratification and the development of targeted, personalized therapeutic interventions.

## Acknowledgments

We would also like to express my deep appreciation to the medical staff and patients of breast cancer of the National Oncology Hospital and Blood Diseases in Najaf Governorate and private laboratories for providing them with the necessary facilities during this study.

*Conflict of Interest*

Author declares no conflict of interest.

## References

1. Nunes MKdS, Lobo LT, Paiva AS, Luna RCP. Influence of fetal programming on carcinogenesis: Pathophysiological and epigenetic perspectives. OBSERVATÓRIO DE LA ECONOMÍA LATINOAMERICANA. 2025;23(6). https://doi.org/10.55905/OELV23N6-060.
2. Hassan F. Risk factors associated with breast cancer among women in iraq: A case-control study. Asian Pacific Journal of Cancer Prevention : APJCP. 2025;26(5). https://doi.org/10.31557/APJCP.2025.26.5.1701.
3. Aponte-Rueda M, Gómez-González F, Merck B. Breast cancer heterogeneity in latin america: A scoping review of clinical-pathological characteristics, molecular subtypes, and survival. World Journal of Surgery. 2025;. https://doi.org/10.1002/WJS.70096.
4. Arko D, Takac I. Inquiry and computer program onko-online: 25 years of clinical registry for breast cancer at the university medical centre maribor. Slovenia | WwwRadioloncolCom Radiol Oncol. 2019;53(3):348–56. https://doi.org/10.2478/raon-2019-0043.
5. Álvarez-Salvago F, Medina-Luque J, Pujol-Fuentes C, Atienzar-Aroca S, Molina-García C, Gutiérrez-García P. Relationship of different levels of physical activity on the health status of long-term breast cancer survivors: A cross-sectional study. Disability and Rehabilitation. 2025;. https://doi.org/10.1080/09638288.2025.2489063;WGROUP:STRING:PUBLICATION.
6. Cerdas M, Farhat J, Elshafie S, Mariyam F, James L, Qureshi A. Midland Metropolitan University Hospital, Smethwick, GBR 11. Jinnah Postgraduate Medical Centre. 2025;. https://doi.org/10.7759/cureus.82762.
7. Higginbotham E, Bhimla A, Batulan Z. Advancing research on chronic conditions in women. Advancing Research on Chronic Conditions in Women.2024:1–562. https://doi.

org/10.17226/27757
8. Dabbagh-Gorjani F. A comprehensive review on the role of interleukin-40 as a biomarker for diagnosing inflammatory diseases. Autoimmune Diseases.2024a;2024(1):3968767. https://doi.org/10.1155/2024/3968767.
9. Navrátilová A, Andrés Cerezo L, Hulejová H, Bečvář V, Tomčík M, Komarc M. Il-40: A new b cell-associated cytokine up-regulated in rheumatoid arthritis decreases following the rituximab therapy and correlates with disease activity, autoantibodies, and netosis. Frontiers in Immunology.12(745523). https://doi.org/10.3389/FIMMU.2021.745523/BIBTEX.
10. Panda V, Mishra B, Mahapatra S, Swain B, Malhotra D, Saha S. Molecular insights on signaling cascades in breast cancer: A comprehensive review. Cancers. 2025;17(2). https://doi.org/10.3390/CANCERS17020234.
11. Dabbagh-Gorjani F. A comprehensive review on the role of interleukin-40 as a biomarker for diagnosing inflammatory diseases. Autoimmune Diseases.2024b;2024(1):3968767. https://doi.org/10.1155/2024/3968767.
12. Askarzadeh M, Ataollahi M, Shokrolahi Z, Atashzar M. Interleukin 40, a novel cytokine. Medical Immunology (Russia. 2024;26(6):1177–82. https://doi.org/10.15789/1563-0625-IAN-2892.
13. Navrátilová A, Bečvár V, Hulejová H, Tomčík M, Štolová L, Mann H. New pro-inflammatory cytokine il-40 is produced by activated neutrophils and plays a role in the early stages of seropositive rheumatoid arthritis. RMD Open. 2023;9(2). https://doi.org/10.1136/RMDOPEN-2022-002894.
14. Lu W, Li Z, Shi S, Meng S, Pang J, Li Y. Elevated serum interleukin-41 levels as a potential biomarker in ankylosing spondylitis. Future Science OA. 2025;11(1). https://doi.org/10.1080/20565623.2025.2545732.
15. Velikova T, Bakopoulou K, Gulinac M, Manova E, Valkov H, Miteva D. Emerging therapeutic and inflammation biomarkers: The role of meteorin-like (metrnl) and follistatin-like 1 (fstl1) in inflammatory diseases. International Journal of Molecular Sciences. 2025;26:26–19. https://doi.org/10.3390/IJMS26199711.
16. Zhang S, Huang G, Li M, Mao Y, Li Y. Interleukin-41 as a biomarker of the anti-inflammatory response associated with hyperuricemia. Immunology Letters. 2023;264:25–30. https://doi.org/10.1016/j.imlet.2023.11.003.
17. Huang D, Liu X, Gao X, Choi C, Giglio G, Farah L. Meteorin-like protein/metrnl/interleukin-41 ameliorates atopic dermatitis-like inflammation. Allergy: European Journal of Allergy and Clinical Immunology. 2024;80(2). https://doi.org/10.1111/ALL.16150.
18. Dent S, Guha A, Moore H, Makari D, McCaleb R, Arias I. Cardiac-star: Prevalence of cardiovascular comorbidities in patients with hr + /her2 − metastatic breast cancer. Cardio-Oncology. 2025;11(1). https://doi.org/10.1186/S40959-025-00305-W.
19. Navrátilová A, Andrés Cerezo L, Hulejová H, Bečvář V, Tomčík M, Komarc M. Il-40: A new b cell-associated cytokine up-regulated in rheumatoid arthritis decreases following the rituximab therapy and correlates with disease activity, autoantibodies, and netosis. Frontiers in Immunology.12(745523). https://doi.org/10.3389/FIMMU.2021.745523/FULL
20. Ababil RY, Alaubydi Mouruj A. Estimation of the level of serum il-40 and il-41 in chronic myeloid leukemia of iraqi patients. Cuestiones de fisioterapia. 2024;53(3):1379. Https://openurl.Ebsco.Com/contentitem/gcd:186654453?Sid=ebsco:Plink:Crawler&id=ebsco:Gcd:186654453.
21. Dabbagh-Gorjani F. A Comprehensive Review on the Role of Interleukin-40 as a Biomarker for Diagnosing Inflammatory Diseases. Mageed R. Autoimmune Diseases. 2024 03 01;2024:1-8. https://doi.org/10.1155/2024/3968767
22. Freedman P, Schock B, O'Reilly S. The Novel Cytokine Interleukin-41/Meteorin-like Is Reduced in Diffuse Systemic Sclerosis. Cells. 2024 07 17;13(14):1205. https://doi.org/10.3390/cells13141205
23. anikawa T, Wilke CM, Kryczek I, Chen GY, Kao J, Núñez G, et al. Data from interleukin-10 ablation promotes tumor development, growth, and metastasis. 2023.
24. HWGd S, BC B, MM P, GLd V, BdCA A, FLA F. Expression profile of il-2, il-6, il-10, and tnf-α in breast tumors. International Journal of Molecular Sciences. 2025;26(16). https://doi.org/10.3390/IJMS26167841.
25. Manager K, Hussain A, MJALH S, May Mohammed A. Diagnostic value of interleukin-40 and interleukin-41 for diabetic kidney disease and diabetes. Karbala J M. 2025;18(1). https://doi.org/10.70863/karbalajm.v18i1.3850.
26. Navrátilová A, Kodet O, Alquicer Barrera G, Mocová K, Prokopcova A, Becvar V. Ab1160 il-40 elevation in systemic sclerosis: Unravelling a link to disease activity, gastrointestinal involvement, immune regulation and fibrotic processes. Annals of the Rheumatic Diseases. 2024;83:1913–4. https://doi.org/10.1136/ANNRHEUMDIS-2024-EULAR.1213.
27. Rizzo C, Barbera L, Lo Pizzo M, Mohammadnezhad L, Lentini V, Donzella D. Pos0101 Potential Involvement Of Il-40 And Il-40 Producing Cells In Systemic Lupus Erythematosus And Lupus Associated Nephritis. Annals of the Rheumatic Diseases. 2022;81(273). https://doi.org/10.1136/ANNRHEUMDIS-2022-EULAR.4803.
28. Wang W, Zhao J, Wu S, Fu J, Zhang Y, Peng W. Serum il-40 increases in patients with rheumatoid arthritis and correlates with some clinical characteristics and comorbidities. Scientific Reports.2024a;14(1):28945. https://doi.org/10.1038/s41598-024-80104-y.
29. Wang W, Zhao J, Wu S, Fu J, Zhang Y, Peng W. Serum il-40 increases in patients with rheumatoid arthritis and correlates with some clinical characteristics and comorbidities. Scientific Reports.2024b;14(1). https://doi.org/10.1038/S41598-024-80104-Y.
30. Navrátilová A, Oreská S, Wünsch H, Mocová K, Kodet O, Rybar M. Serum il-40 is elevated in systemic sclerosis and is linked to disease activity, gastrointestinal involvement, immune regulation and fibrotic processes. Arthritis Research and Therapy. 2025;27(1). https://doi.org/10.1186/s13075-025-03570-3.
31. Bouare N, Delwaide J. Interleukin-mediated therapies in liver diseases and comorbidity effects. World Journal of Hepatology. 2024;16(7). https://doi.org/10.4254/WJH.V16.I7.980.
32. Barberis M, López A. Metabolic imbalance driving immune cell phenotype switching in autoimmune disorders: Tipping the balance of t- and b-cell interactions. Clinical and Translational Medicine. 2024;14(3). https://doi.org/10.1002/CTM2.1626.
33. Jia K, Shao Y, Tan T, Liu X, Zhang Q, Zhao N. Serum interleukin-40 increases in anti-achr antibody-positive myasthenia gravis and correlates with disease activity. Frontiers in Neurology. 2025;16. https://doi.org/10.3389/FNEUR.2025.1680530.
34. Hameed S, Abed R. The elevated serum interleukin-40 levels and gene expression as novel biomarkers in iraqi women with breast cancer: Il-40 as a biomarker in iraqi bc. Archives of Breast Cancer. 2025;12(4):448–56. https://doi.org/10.32768/ABC.5801274963-802.
35. Duan Y, Li H, Luo D, Jiang J, Liu B, Li G. Serum il-41 might be a biomarker for ivig resistance and coronary artery lesions

in kawasaki disease. International Immunopharmacology. 2023;122. https://doi.org/10.1016/j.intimp.2023.110600.
36. Namitokov A, Karabakhtsieva K, Malyarevskaya O. Inflammatory and lipid biomarkers in early atherosclerosis: A comprehensive analysis. Life. 2024;14(10). https://doi.org/10.3390/LIFE14101310.
37. Fernández L, Cedrón M, Molina A. Alterations of lipid metabolism in cancer: Implications in prognosis and treatment. Frontiers in Oncology. 2020;10(577420). https://doi.org/10.3389/FONC.2020.577420.
38. Chatterjee T, Beffert E, Liefwalker D. Fueling the fire–a pan-cancer analysis of myc-regulated lipid metabolism. Frontiers in Cell and Developmental Biology. 2025;13(1669544). https://doi.org/10.3389/FCELL.2025.1669544.
39. Bridgewood C, Russell T, Weedon H, Baboolal T, Watad A, Sharif K. The novel cytokine metrnl/il-41 is elevated in psoriatic arthritis synovium and inducible from both entheseal and synovial fibroblasts. Clinical Immunology. 2019;208. https://doi.org/10.1016/j.clim.2019.108253.
40. Cen T, Huang M, Li M, Jin J, Ding Q, Lv D. Increased serum il 41 associated with acute exacerbation of chronic obstructive pulmonary disease. Experimental and Therapeutic Medicine. 2024;28(2). https://doi.org/10.3892/ETM.2024.12601.
41. Abd AAl, muslem hade I, ghuraibawi ahraa AZAG, ghazi sadeq Z, majid alkhaiat M. Association between serum interleukin-41 levels and breast cancer status: A case-control study in iraqi women. Iranian journal of breast diseases. 2025;18(3):85–99.