

Mycosis Fungoides in Iraq: Novel Insights into Age Disparities and CD68 Expression as a Marker of Disease Progression

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Abstract: Background: In the context of Iraqi communities, mycosis fungoides (MF) is the main cutaneous T-cell lymphoma that is the most prevalent, but little information is available about it. One of the areas that is still being actively researched is the role that tumor-associated macrophages (TAMs), which are identified by their expression of CD68, play in the course of illness. **Objective:** is to define the demographic, clinical, and immunohistochemical characteristics of MF in an Iraqi cohort, with a particular focus on identifying relationships that may provide evidence regarding the pathogenesis and course of the illness. **Methods:** A retrospective review of 22 patients diagnosed with MF at a single dermatology center in Iraq (2020–2025). Data on age, sex, disease duration, clinical subtype, stage, and CD4/CD8/CD68 expression were analyzed using Mann-Whitney U tests, Kruskal-Wallis tests, Fisher's exact tests, and Spearman's correlation. **Results:** A retrospective review of 22 individuals diagnosed with MF at a single dermatological center in Iraq (2020–2025) was conducted as part of the methodology involved. Mann-Whitney U tests, Kruskal-Wallis tests, Fisher's exact tests, and Spearman's correlation were utilized in order to assess the data pertaining to age, gender, length of disease, clinical subtype, stage, and expression of CD4/CD8/CD68 and other related factors. As a result, the cohort consisted of 14 males and 8 females, with a gender ratio of 1.75:1. In this study, it was discovered that males presented at a considerably older age compared to females (49.2 ± 13.7 years versus 35.5 ± 13.1 years, $p = 0.023$). This is a novel discovery that has not been discussed in the literature before. There was a positive correlation between the length of the disease and the age at diagnosis ($\rho = 0.47$, $p = 0.028$), which indicates that older patients may have delayed presentation. In addition to lichenoid (18.2%) and pokilodermatous -erythrodermic (13.6%), the most common type of MF was classical, which accounted for 27.3% of all cases. The least was a female patient 15 years old who had hypopigmented MF. There were two significant associations that were found associated with CD68 expression: Strong CD68 positivity was significantly associated with tumor-stage disease (present in 2/2 tumor-stage patients compared to 3/17 patch/plaque patients; $p = 0.048$); and patients with strong CD68 expression had significantly longer disease duration than those with weak or negative expression (7.8 ± 3.1 years versus 4.2 ± 3.1 years, $p = 0.035$). The CD4+ phenotype was the most prevalent (84.2%). **Conclusion:** This work reveals unexpected findings in an Iraqi MF cohort, including a large age discrepancy between the sexes at the time of diagnosis, a link between age and the length of the disease, and, most importantly, dual associations of CD68 expression with both advanced stage and prolonged disease duration. These findings imply that tumor-associated macrophages have a role in the chronicity and progression of mycosis fungoides (MF), raising the possibility that CD68 could be used as a biomarker for risk classification. It is necessary to do more extensive research in order to verify these discoveries and investigate the consequences they have for prognosis.

Keywords: Mycosis Fungoides, CD68, Cutaneous T-Cell Lymphoma, Tumor-Associated Macrophages.

INTRODUCTION

Mycosis fungoides, also known as MF, is the most prevalent kind of primary cutaneous T-cell lymphoma, making up nearly fifty percent of all cutaneous lymphomas [1]. Although clinical variants such as folliculotropic, erythrodermic, hypopigmented, and lichenoid types are well-recognized [2-5], the characteristic course of the disease is often slow and

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gradual, progressing from patches to plaques and eventually tumors. When it comes to immunophenotype, the majority of patients of Mycosis fungoides (MF) display a mature CD3⁺ CD4⁺ CD8⁻ helper T-cell profile [6].

The microenvironment of the tumor, and more specifically tumor-associated macrophages (TAMs), has emerged as a significant contributor to the advancement of lymphoma. This comes in addition to the malignant T-cell population. There is a correlation between higher TAM infiltration and bad outcomes in a variety of lymphomas, including cutaneous T-cell lymphoma [7]. CD68, which is a pan-macrophage marker, is used to identify TAMs in tissue sections. However, there is still a lack of information regarding the expression of CD68 in MF, and the relationship between this expression and clinical factors including the duration of the disease and its stage needs to be further investigated.

It is difficult to find epidemiological and clinicopathological data on MF from the Middle East, and there is no specialized series from Iraq that has thoroughly investigated the correlations between these factors. The purpose of this research is to define the demographic, clinical, and immunohistochemical characteristics of MF in an Iraqi cohort. The primary emphasis of this investigation is to uncover associations that may shed light on the pathophysiology of the disease and to identify potential biomarkers of further progression.

METHODS

Study Design and Setting

We carried out a retrospective review of all of the patients who were diagnosed with mycosis fungoides at the Dermatology Department of a tertiary care hospital in Iraq during the months of January 2020 and March 2025. Both clinical documents and histology data were consulted in order to identify the cases.

Data Collection

A number of variables were extracted for each individual patient, including the following: age at diagnosis (in years), gender, disease duration (in years, as reported by the patient from the onset of skin lesions to diagnosis), clinical subtype (classified according to published criteria (3) as classical, lichenoid, follicular, erythrodermic/tumor, hypopigmented, or classical with keratoderma), stage at presentation (patch, plaque, or tumor), and immunohistochemical results for CD4, CD8, and CD68. The immunohistochemical expression for CD4 and CD8 was recorded as "higher," "equal," "weak," "strong," "negative," or "not done" (according to the original pathology reports). For the purpose of analysis, the expression of CD68 was divided into two categories: "strong" and "weak/negative."

Immunohistochemistry

Immunohistochemical staining for CD68 (clone KP1, Dako/Agilent) was performed on formalin-fixed, paraffin-embedded tissue sections (3–5 μ m thick) mounted on coated slides. Sections were deparaffinized in xylene and rehydrated through graded alcohol. Heat-induced epitope retrieval was carried out using Target Retrieval Solution (pH 6.0) at 95–99°C for 20 minutes. The sections were subsequently incubated with the primary anti-CD68 antibody at a dilution of 1:50 for 30 minutes at room temperature. Immunodetection was performed using the Dako EnVision+ HRP system, with 3,3'-diaminobenzidine (DAB) as the chromogen. Slides were counterstained with Mayer's hematoxylin, dehydrated, and mounted. Human tonsil tissue was used as a positive control to confirm cytoplasmic staining of macrophages. Tumor-associated macrophages (TAMs) were evaluated in hotspot areas. CD68 expression was interpreted as weak/negative when tumor-associated macrophages showed light- yellow cytoplasmic staining, and strong/positive when dark- brown cytoplasmic staining was observed.

Statistical Analysis

The Shapiro-Wilk test was utilized to determine the normality of continuous data. The results of this test are reported as the mean plus or minus the standard deviation (SD). The presentation of categorical variables is done through the use of frequencies and percentages.

For statistical analysis, the following tests were carried out:

A Mann-Whitney test The U test is used to compare continuous variables between two groups that are independent of one another. For the purpose of comparing continuous variables across different groups, the Kruskal-Wallis test is utilized.

In order to investigate the relationships between categorical variables, Fisher's exact test is utilized. This is because the predicted cell counts are rather low.

For the purpose of evaluating the correlations between continuous variables, Spearman's rank correlation (ρ) is utilized.

When the p-value for both sides was less than 0.05, it was declared statistically significant. All of the analyses were carried out with SPSS version 26, which was developed by IBM Corporation in Armonk, New York.

Ethical Considerations

The institutional review board gave their assent to the findings of the study. The confidentiality of the patients was protected by anonymizing all of the data, and individual agreement was not required because the study was designed in a retrospective manner.

RESULTS

Demographic and Clinical Characteristics

There were a total of 22 patients that were included. The demographic and clinical characteristics of these individuals are summarized in Table 1. It was determined that there were 14 males (63.6% of the cohort) and 8 females (36.4% of the cohort), which resulted in a male-to-female ratio of 1.75:1. The mean age at diagnosis was 44.2 ± 14.9 years (range 15–72 years). Mean disease duration before diagnosis was 5.0 ± 3.4 years (range 1–11 years).

Table 1: Demographic, clinical, and immunohistochemical characteristics of 22 patients with mycosis fungoides

Characteristic	Value
Age (years), mean ± SD (range)	44.2 ± 14.9 (15–72)
Sex, n (%)	
Male	14 (63.6)
Female	8 (36.4)
Disease duration (years), mean ± SD (range)	5.0 ± 3.4 (1–11)
Clinical subtype, n (%)	
Classical (incl. classical erythematous)	6 (27.3)
Lichenoid	4 (18.2)
Poikilodermatous	3 (13.6)
Erythrodermic/tumor	3 (13.6)
Follicular	2 (9.1)
Classical + keratoderma	2 (9.1)
Hypopigmented	1 (4.5)
Not recorded	1 (4.5)
Stage (n=19), n (%)	
Patch	10 (52.6)
Plaque	7 (36.8)
Tumor	2 (10.5)
CD4 expression (n=19), n (%)	
Higher	16 (84.2)
Equal	3 (15.8)
CD8 expression (n=19), n (%)	
Weak	11 (57.9)
Equal	4 (21.1)
Higher	2 (10.5)
Equal high	1 (5.3)
Not done	1 (5.3)
CD68 expression (n=19), n (%)	
Weak	8 (44.4%)
Strong	5 (27.7%)
Negative	5 (27.7%)
Not done	1 (5.3)

Age and Sex: A Significant Disparity

Males presented at a significantly older age than females (49.2 ± 13.7 years vs. 35.5 ± 13.1 years; Mann-Whitney $U = 24.0$, $p = 0.023$) (Figure 1). This age disparity of nearly 14 years represents a novel finding in MF epidemiology. There was no significant difference in disease duration between males and females (5.1 ± 3.4 years vs. 4.9 ± 3.6 years; $p = 0.82$).

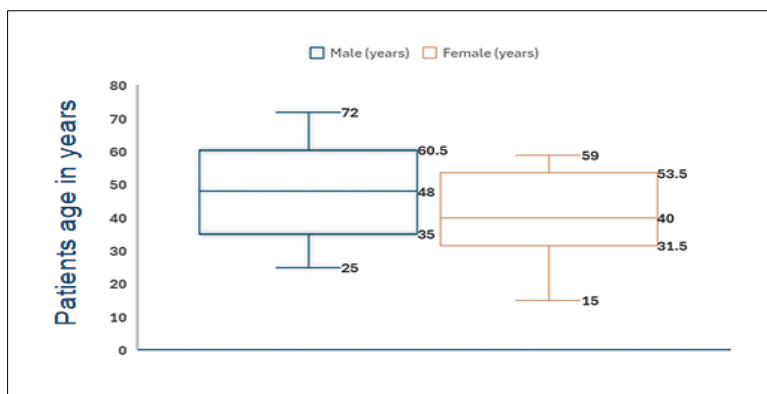


Figure 1: Dot plot comparing age at diagnosis between male (n=14) and female (n=8) patients with mycosis fungoides. Males were significantly older at diagnosis (49.2 vs. 35.5 years; Mann-Whitney U = 24.0, p = 0.023). Horizontal lines represent means.

Correlation between Age and Disease Duration

A moderate positive correlation was observed between age at diagnosis and disease duration (Spearman's $\rho = 0.47$, $p = 0.028$) (Figure 2), indicating that older patients tended to have a longer history of cutaneous lesions before receiving a diagnosis. The disease duration increases by about 0.11 years for each additional year of age at diagnosis.

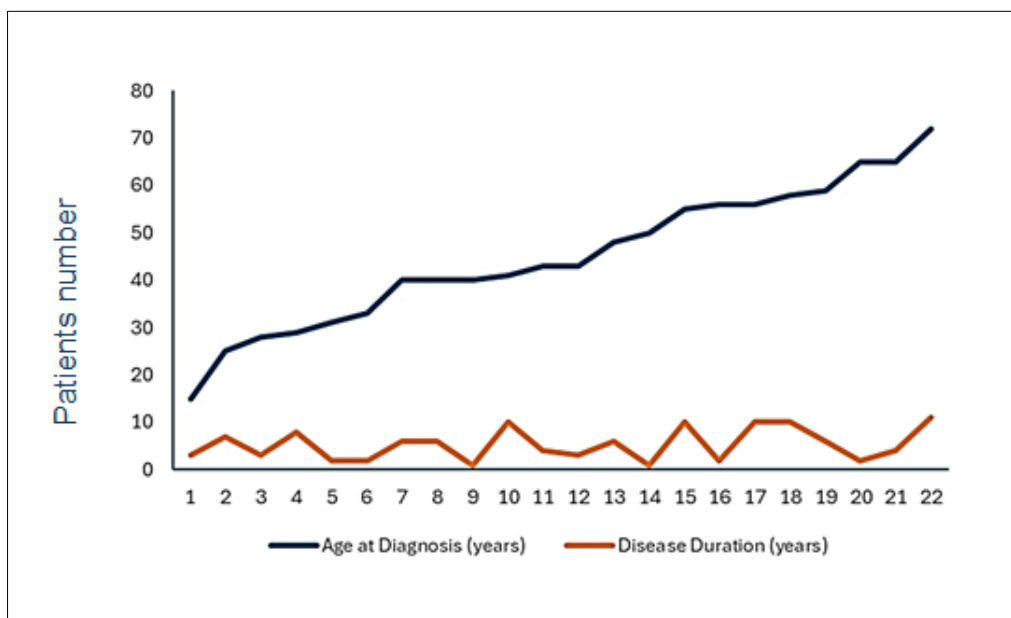


Figure 2: Scatter plot showing correlation between age at diagnosis and disease duration (Spearman's $\rho = 0.47$, $p = 0.028$). Dashed line represents linear regression trend. Older patients had longer disease duration before diagnosis.

Clinical Subtypes and Age/Duration Distribution

Classical MF was the most common subtype (27.3%), followed by lichenoid (18.2%), erythrodermic/tumor (13.6%), follicular (9.1%), classical with keratoderma (9.1%), and hypopigmented (4.5%). The hypopigmented MF case occurred in a 15-year-old female presenting with patch-stage disease, consistent with the known predilection of this variant for young, dark-skinned individuals [8].

Table 2 represents the mean age and disease duration for each clinical subtype. The hypopigmented variant occurred in the youngest patient, while erythrodermic/tumor and follicular variants occurred in older patients. Due to small subgroup sizes, differences were not statistically significant (Kruskal-Wallis test for age: $H = 9.2$, $df = 7$, $p = 0.24$; for disease duration: $H = 6.8$, $df = 7$, $p = 0.45$).

Table 2: Age and disease duration by clinical subtype

Subtype	n	Mean age (years) \pm SD	Mean duration (years) \pm SD
Classical	6	41.7 \pm 16.4	5.7 \pm 4.0
Lichenoid	4	38.5 \pm 4.9	3.3 \pm 2.1

Poikilodermatous	3	47.7 ± 8.5	5.7 ± 4.5
Erythrodermic/tumor	3	55.3 ± 12.7	7.0 ± 4.6
Follicular	2	56.0 ± 2.8	8.0 ± 2.8
Classical + keratoderma	2	42.0 ± 1.4	2.5 ± 0.7
Hypopigmented	1	15.0	3.0
Not recorded	1	43.0	4.0

Stage at presentation and subtype distribution

It was possible to determine the disease stage for 19 patients. Ten patients, or 52.6%, were diagnosed with the patch stage, followed by plaque (7 patients, or 36.8%), and then tumor (2 patients, or 10.5%). One of the patients who was diagnosed with "sezary/plaque" was categorized as having plaque for the purpose of analysis.

The clinical subtypes are broken down into their respective stages and given in Table 3. Of particular note is the fact that tumor-stage disease was only observed in one patient and in one patient who had erythrodermic/tumor variations.

Table 3: A distribution of clinical subgroups according to stage (n = 19)

Subtype	Patch	Plaque	Tumor	Total
Classical	3	1	0	4
Lichenoid	3	0	0	3
Poikilodermatous	0	2	1	3
Erythrodermic/tumor	1	1	1	3
Follicular	2	0	0	2
Classical + keratoderma	0	2	0	2
Hypopigmented	1	0	0	1
Not recorded	0	1	0	1
Total	10	7	2	19

Immunohistochemistry: CD4 and CD8 expression

There were 19 patients whose CD4 findings were available; out of these, 16 (84.2%) showed "higher" expression, and three (15.8%) showed "equal" expression, which confirmed that the CD4+ immunophenotype was the predominant one. There were no significant connections detected between the pattern of CD4 expression and parameters such as age, gender, length of disease, or stage (p > 0.05 for all of these factors).

There was a significant variation in the CD8 results, with 57.9% exhibiting "weak" expression. This is most likely due to the fact that different levels of reactive CD8+ T-cell infiltration were observed.

CD68 Expression: A marker of progression and chronicity.

CD68 results were available for 18 patients. For analysis, "strong" expression (5 patients, 27.7%) was compared against combined "weak/negative" (13 patients, 72.2%).

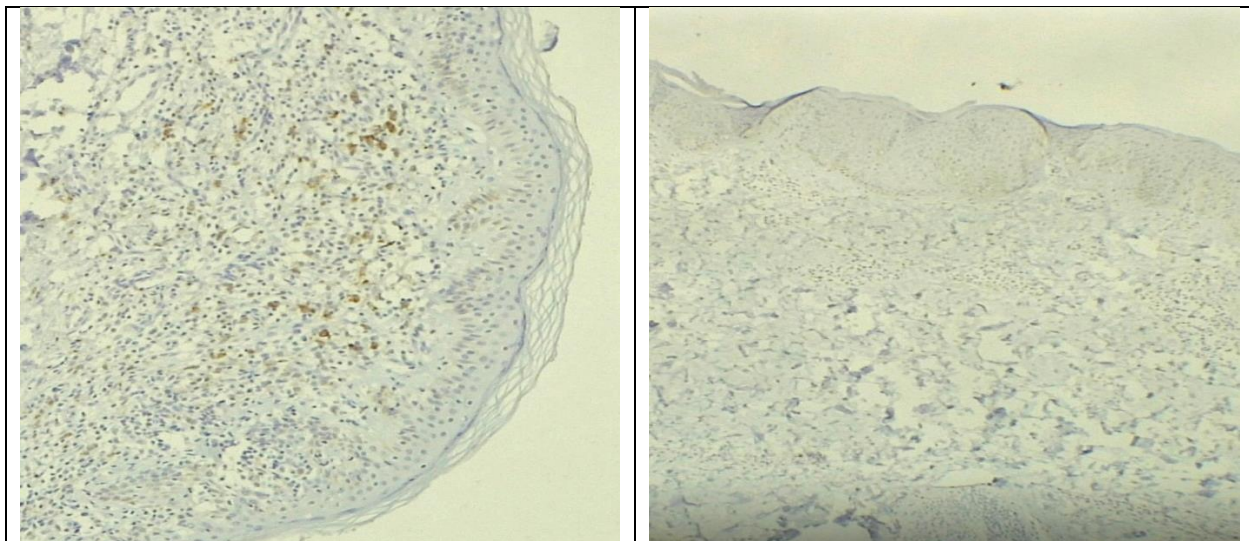


Figure 3: Immunohistochemical staining for CD68 in mycosis fungoides skin lesions. (A) Strong CD68 expression showing numerous positive tumor-associated macrophages (brown cytoplasmic staining) in a patient with tumor-stage disease and prolonged disease duration. (B) Weak/negative CD68 expression in a patient with patch-stage disease. (Original magnification ×200; counterstained with hematoxylin).

Two significant associations emerged, as detailed in Table 4:

1. Association with Tumor-Stage Disease: Among the 2 tumor-stage patients, both (100%) showed strong CD68 positivity. In contrast, among 17 patients with patch/plaque stage, only 3 (17.6%) showed strong CD68 expression. This association between strong CD68 expression and tumor-stage disease was statistically significant (Fisher's exact test, $p = 0.048$).

2. Association with Longer Disease Duration: Patients with strong CD68 expression had a significantly longer disease duration (7.8 ± 3.1 years) compared to those with weak or negative CD68 expression (4.2 ± 3.1 years) (Mann-Whitney $U = 11.5$, $p = 0.035$) (Figure 4).

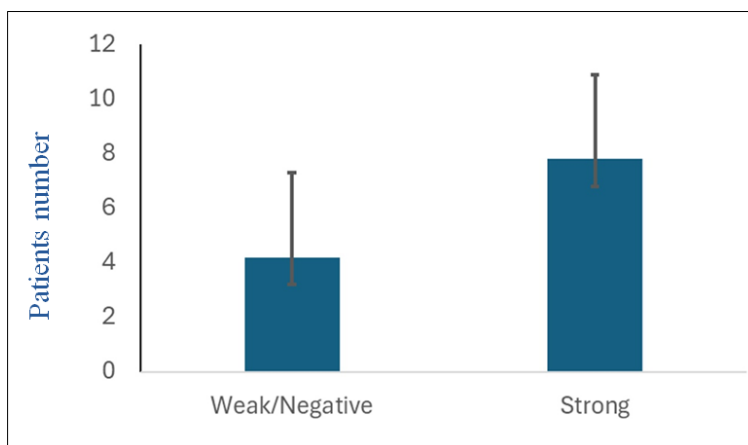


Figure 4: Bar chart comparing disease duration by CD68 expression status. Patients with strong CD68 expression (n=5) had significantly longer disease duration than those with weak/negative expression (n=13; 7.8 vs. 4.2 years; Mann-Whitney $U = 11.5$, $p = 0.035$). Bars represent mean \pm SD.

3. Association with Clinical Subtype: Strong CD68 positivity was observed in 2 of 3 erythrodermic/tumor patients (66.7%), and 1 of 6 classical patients (16.7%). Due to small numbers, this did not reach statistical significance ($p = 0.21$).

4. No Association with Age or Sex: There was no significant difference in age between patients with strong CD68 expression (mean 52.6 ± 14.4 years) and those with weak/negative expression (mean 41.8 ± 14.5 years; $p = 0.18$). Sex distribution was similar between groups ($p = 1.00$).

Table 4: Expression of CD68 according to the stage of the disease and clinical features

Characteristic	Strong CD68 (n=5)	Weak/Negative CD68 (n=13)	p-value
Stage			0.048
Patch/Plaque	3 (60.0%)	13 (100%)	
Tumour	2 (40.0%)	0 (0%)	
Mean disease duration (years)	7.8 ± 3.1	4.2 ± 3.1	0.035
Mean age (years)	52.6 ± 14.4	41.8 ± 14.5	0.18
Sex			1.00
Male	3 (60.0%)	8 (61.5%)	
Female	2 (40.0%)	5 (38.5%)	

DISCUSSION

This work is the first comprehensive clinicopathological examination of mycosis fungoides in an Iraqi cohort. Despite the relatively small sample size, it produces various novel findings that contribute to our advancement in our understanding of the disease. There are three primary observations that deserve to be discussed: 1. A significant age difference between the sexes at the time of diagnosis; 2. A positive correlation between age and the duration of the disease; and 3. A dual associations of CD68 expression with both tumor-stage disease and longer disease duration, which implicate tumor-associated macrophages in the progression and chronicity of the disease.

Age and Sex: A Previously Underappreciated Disparity

The fact that males appeared at a considerably older age than females (49.2 years versus 35.5 years, $p = 0.023$) is, to the best of our knowledge, a novel observation in the field of MF epidemiology. There has been no systematic

investigation or emphasis placed on the age differences between the sexes at the time of presentation, despite the fact that male predominance in MF has been well-established [1-9]. This gap could be attributed to a number of different explanations:

Possible biological causes include the fact that females in this demographic may experience an earlier beginning of MF, or that hormonal factors may play a role in the manifestation of the disease. The effects of estrogen on T-cell immunity are multifaceted, and it has the potential to influence how diseases manifest themselves.

Healthcare-seeking behavior: Within the Iraqi sociocultural context, it is possible that women of reproductive age have more regular contact with healthcare systems (for example, through antenatal care or child health checkups), which may result in the earlier diagnosis of skin lesions. In contrast, older men may put off getting medical assistance for skin issues for longer periods of time.

Exposures to the environment: In Iraq, the start of disease may be influenced by differences in occupational or environmental exposures between younger women and older men.

Reference bias: Because this was a study conducted at a single center, it is possible that referral patterns unintentionally chose for this age-sex distribution.

The validity of this finding needs to be established by larger-scale, population-based research, as well as through the investigation of its possible biological context.

Age-Duration Correlation: Delayed Diagnosis in Older Patients

Based on the somewhat positive connection between age and disease duration ($\rho = 0.47$, $p = 0.028$), it may be inferred that patients who are older had a longer history of skin lesions prior to being diagnosed. This may be a result of a delay in diagnosis in senior individuals, which may be caused by the attribution of skin changes to age, benign dermatoses, or comorbidities; decreased access to healthcare; or a lower clinical suspicion of lymphoma in older patients who have chronic dermatoses. As a result of this discovery, there is an increased need for increased awareness of MF among older populations and in places that provide care for the elderly.

Clinical Variants in the Iraqi Population

In the Iraqi population, clinical variations were observed.

There are many different clinical subgroups that have been detected, which lends credence to the idea that MF is a "great imitator" [5]. The rather high frequencies of lichenoid variants (18.2% and 13.6%, respectively) are remarkable and may be a reflection of genetic or environmental variables that are particular to this group; nonetheless, although the numbers are tiny, definitive conclusions cannot be drawn from them. Based on the findings shown in Table 3, it can be inferred that erythrodermic/tumor variations were the only ones to exhibit tumor-stage disease, indicating that these subtypes may have a higher prevalence of advancement.

The presence of hypopigmented MF in a female who was 15 years old is consistent with the well-known preference of this variant for young people with dark skin [8]. This finding highlights the significance of taking MF into consideration when making a differential diagnosis of hypopigmented patches in adolescents.

CD68 Expression: Linking Macrophages to MF Progression and Chronicity

The novel connections that were discovered in relation to CD68 expression, which is a marker of tumor-associated macrophages (TAMs), are the most important contributions that this study has made.

CD68 and illness at the stage of the tumor: There is compelling evidence that TAMs have a role in the course of disease, as demonstrated by the substantial relationship between strong CD68 expression and tumor-stage MF ($p = 0.048$), with both tumor-stage patients showing strong positive (Table 4). This fits with and extends the work that was done in the past by Sugaya *et al.*, [7], who revealed a link between CD163+ macrophages and the course of illness in cutaneous T-cell lymphoma. Our research indicates that CD68, which is a clinical marker that is more easily accessible, has the potential to function as an immunohistochemical indicator of advanced disease in a practical setting.

CD68 and the length of the disease: The unique discovery that individuals with robust CD68 expression had significantly longer disease duration (7.8 years vs to 4.2 years, $p = 0.035$) implies that there is a temporal dimension to the accumulation of TAM. This can be interpreted in two different ways: (1) cases with an inherently more indolent but prolonged course are more likely to accumulate TAMs over time, and these TAMs may eventually drive transformation to the advanced stage; or 2 cases with chronic inflammation that has been present for a long time promotes progressive recruitment and activation of macrophages, which in turn creates a microenvironment that is conducive to the progression

of the tumor. There is support for a paradigm in which persistent inflammation encourages the accumulation of TAM, which then accelerates the course of disease. This hypothesis is supported by the dual relationship of CD68 with both longer duration and advanced stage.

Clinical repercussions include It is possible that the expression of CD68 could operate as a beneficial biomarker for risk classification if it is proven in larger investigations. Patients who have a high level of CD68 expression, particularly those who have had the condition for a long time, may benefit from more active early intervention or tighter monitoring in order to prevent the progression of the disease to a more advanced stage. 66.7% of erythrodermic/tumor and variations have strong CD68 expression, which shows that these subtypes may be particularly connected with a macrophage-rich milieu. This is because these subtypes have a higher prevalence of strong CD68 expression.

Comparison with Regional Data

Our demographic findings are, for the most part, in agreement with series from nations that are geographically close by. In a Turkish series of 66 patients with myocardial infarction, the mean age was 46.8 years, and the majority of the patients were male (58%) [10]. Similar findings were reported by an Iranian sample of 38 patients, which included a mean age of 47 years and a male predominance [11]. On the other hand, neither of the studies assessed CD68 expression in relation to disease duration or reported age variations based on gender, which highlights the novel contributions that the current study makes.

Limitations

The number of the cases is limited because the disease is a rare disease despite the study is conducted at a leading national referral hospital in Iraq. This creates limitations in terms of statistical power and generalizability in this cohort, need to be validated. Second, the retrospective design may result in biases in the selection of participants and the sources of information. Third, the immunohistochemistry data for some patients were lacking the necessary information. The clinical staging was based on the records that were accessible, and it is possible that it did not adhere to the normative norms. Fifth, because there is a lack of follow-up data, it is impossible to determine whether or not the expression of CD68 can accurately predict survival outcomes. In the sixth place, the expression of CD68 was evaluated using a semi-quantitative approach; quantitative methods may provide more accurate relationship analysis. Because this was a study conducted at a single center, it is possible that the findings cannot be generalized to the total population of Iraq.

In spite of these limitations, the statistically significant relationships that were revealed, in particular the unexpected findings concerning CD68, offer a solid basis for the development of hypotheses and for the conduct of further study.

CONCLUSION

In this first comprehensive clinicopathological investigation of mycosis fungoides in an Iraqi cohort, various unexpected insights have been obtained, including the following:

Males present roughly 14 years older than females when they are diagnosed, indicating that there is a considerable age gap between the sexes at the time of diagnosis. Due to the fact that this discovery has not been highlighted in the previous research, it is necessary to do research on the biological and sociocultural factors that influence it [2]. The patients' ages at the time of diagnosis had a positive correlation with the length of the condition, which suggests that older patients may have diagnostic delay and highlights the necessity of heightened clinical vigilance in this population.

According to Table 4, the expression of CD68, which is a marker of tumor-associated macrophages, has dual and substantial relationships with both the stage of the disease and the length of time the disease has been present. This research suggests that TAMs have a role in the course and chronicity of myocardial infarction (MI), and it also suggests that CD68 immunohistochemistry could be used as a practical biomarker for identifying patients who are at risk for advanced illness [5]. The spectrum of clinical variants in this Iraqi community, which includes relatively high rates of lichenoid and MF as well as a case of hypopigmented MF in an adolescent, contributes to the expansion of the recorded global diversity of MF presentations.

Despite the fact that these findings are preliminary, they present hypotheses that can be tested for further research: What is the relationship between CD68 expression and progression-free survival? Can anti-macrophage medicines be used to modify the progression of the disease in MF? Are there biological differences in the pathophysiology of diseases or sociocultural variables that influence access to healthcare that are responsible for the age-sex disparity?

There is an immediate need for larger, prospective multicenter studies that include standardized data collection, quantitative immunohistochemistry, and long-term follow-up in order to validate these observations and determine whether or not CD68 expression should be incorporated into routine prognostic assessment of patients who have mycosis fungoides.

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