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Original article

Evaluating Immunohistochemical Expression of Junctional Adhesion Molecule - A (JAM- A) in Multiple Myeloma

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Abstract

Background: 1% of all cancers and about 10% of all haematologic malignancies are multiple myeloma (MM). In various human cancers, including multiple myeloma, abnormal expression or deregulation of JAM-A, which controls cell growth and differentiation, results in a more aggressive phenotype and poor prognosis. **Aim**: to evaluate the immunohistochemical expression of JAM-A in cases of multiple myeloma and correlate its expression with variable clinicopathological parameters to assess its possible diagnostic and prognostic role and hence therapeutic modalities. **Material and method:** This is a selected retrospective immunohistochemical study of JAM-A performed on 40 cases of MM. **Results**: There is a statistically significant direct correlation between JAM-A overexpression and older Age (P= .022), histological type (immature & plasmablastic)(P=.020), diffuse pattern of infiltration(P=.016), increased BMB cellularity(P=.017), higher ISS Stage (P=.033), advanced R- ISS Stage (P=.042), No Complete response(P=.008), more Resistance to therapy (P= .032), Karyotyping result (P=.019), short survival (P= .014). **Conclusion**: JAM-A overexpression might have an important role in proliferation, invasion, progression, poor outcome of multiple myeloma cases. So, JAM-A might have a promising value in treatment of MM.

Keywords: Immunohistochemical, Junctional Adhesion Molecule – A, Multiple myeloma.

Introduction

Being one of the most aggressive haematological malignancies, multiple myeloma (MM) is a huge, unsolvable health concern. The disease begins when cancerous plasma cells divide and multiply in the bone marrow [(1)].

According to GLOBOCAN 2020, MM comes in 22th rank among worldwide malignancies. In Egypt, the newly cases of MM represent 0.55% of all malignancies [(2)]

The "CRAB" criteria are a group of symptoms that are commonly observed in patients with multiple myeloma, a type of cancer that originates in the bone marrow. These symptoms include hyperCalcemia, Renal insufficiency, Anaemia, and lytic Bone lesions[(3)].

In nearly all instances, a bone marrow test should be conducted as it is an essential part of diagnosing plasma cell myeloma. Despite strong clinical, laboratory, and radiographic evidence, a bone marrow test is still necessary to confirm a diagnosis of myeloma. Results from bone marrow assays can help with prognosis, monitoring treatment efficacy, and spotting disease recurrence [(4)].

Smoldering myeloma (SMM) has a 50% progression risk and pre-neoplastic forms of monoclonal gammopathy of undetermined significance (MGUS) have a 5% risk of developing into multiple myeloma (MM) within 5 years [(5)].

Regarding pathological view, MM composed of 10% of clonal plasma cells. The pattern of infiltration of MM can be focal, interstitial, or diffuse, and their morphology can range from mature to immature, plasmablastic to anaplastic [(3)].

Malignant plasma cells can take on a variety of shapes and sizes, from those that are hardly noticeable from normal plasma cells to those that resemble undifferentiated blasts [(6)].

The degree of pleomorphism can change as the differentiation level changes. Nearly normal-looking plasma cells characterise well-differentiated myeloma, the most common type. There is nuclear and cytoplasmic atypia in moderately differentiated myeloma. Rarest of all myelomas, pleomorphic myelomas have almost no cytoplasm and looking very anaplastic with vesicular nuclei and prominent nucleoli [(7)].

Four primary categories will be used to classify the prognostic factors of MM: risk stratification (including MM staging, plasma cell labelling index, and cytogenetics); monitoring of response tools (including serum heavy/light chain assays, serum free of serum, and advanced imaging modalities); minimal residual disease monitoring methods; novel prognostic markers; and other prognostic factors concerning the host and tumour burden[(8)].

Over the past several years, our understanding of the prognostic markers in MM has made great strides. The development of risk-adaptive

treatment strategies is the end aim of creating prognostic models in MM [8].

Junctional adhesion molecule –A (JAM-A) is a type 1 transmembrane glycoprotein belong to the immunoglobulin superfamily [9]. JAM-A has been proved to have prognostic role in a number of tumors [10].

This study aims to evaluate the immunohistochemical expression of JAM-A in cases of multiple myeloma and correlate its expression with variable clinicopathological parameters to assess its possible diagnostic and prognostic role and hence therapeutic modalities.

Material and methods:

Study group:

This is a selected retrospective study including 40 cases of Multiple Myeloma. The material included archival formalin fixed paraffin embedded blocks were selected retrograde from 2018 till 2019. The patients were recruited from the Clinical Pathology Department-Faculty of Medicine-Benha University, National Cancer Institute, and Cairo University. The control cases were 7 cases who did not have the diseases, confirmed by other standard techniques [11].

Myelomas can be categorised into four stages: mature, intermediate, immature, and plasmablastic, all based on cytologic features [6].

Clinicopathological data were collected from the files of patients in form of age, sex, laboratory data, karyotyping, ISS staging systems [12], R-ISS staging system [13], complete response to therapy, resistance ,relapse and one year survival. The study was approved by the Research Ethical committee of Faculty of Medicine, Benha University, Egypt (MSC 15/9/2022).

A-Histopathological Examination: Formalin-fixed and paraffin-embedded blocks were cut at 5 μm thickness and stained with hematoxylin and eosin. Two observers reviewed the microscopic sections from all the cases and were unaware of their diagnosis [14].

Inclusion criteria including the following

1- Egyptians.

2- Either sex is eligible.

3- Ages of patients are above 18 years old.

Exclusion criteria included the following

1- Less than 18 years old patients.

2- Localized plasma cell lesions without bone marrow involvement.

3- If the initial clinical information was absent.

4- Non-Egyptian patients.

5- History of chemotherapy.

Immunohistochemical studies:

Tissue sections measuring four microns were obtained from formalin-

fixed, paraffin-embedded tissue blocks on coated slides. A standard labelled streptavidin-biotin system (Biospes, China) was employed in accordance with the manufacturer's requirements. The antigen retrieval process was conducted using Protein A affinity purified as **detailed in Table (1).**

Immunoreaction to JAM-A was visualized by adding 0.02% diaminobenzine (DAB) as a chromogen. Negative control was achieved by omitting the primary antibody.

Interpretation of JAM- A expression:

JAM-A was considered positive in tumor cells as brownish homogenous membranous staining [(9)].

The intensities of JAM-A membranous staining were evaluated in terms of their extent and intensity. The final score was determined by multiplying the intensity and percentage scores. A case was classified as low expressed if it had a count of six or fewer, and as highly expressed if it had a count of six or more [(15)].

Kaplan-Meier curves were employed to represent survival data, and the log-rank test was employed to investigate its statistical significance.Twelve months was the median follow-up duration, with a range of six to eighteen months. The histological categories of plasmablastic and anaplastic origin were combined for statistical analysis.

Approval Code: MS 15-9-2022

Statistical analysis:

Results were analyzed using SPSS (SPSS Inc., Chicago, IL, USA) (version 22) statistical package for Microsoft Windows as follows: P value >0.05 is no significant (N), P<0.05 is significant, and P≤ 0.01 is highly significant.

Results

Clinicopathological results:

The age of 40 studied cases ranged from 22-74 years with the mean age was 53.53±SD10.046 years. Multiple myeloma cases included 70% of the studied patients < 60 years old while 30% were ≥60 years old

Out of 40 cases of multiple myeloma, 25% were mature type as shown in figure (1.A), 8% were immature type as shown in **figure (1.B)** and 7% were other types (plasmablastic and anaplastic types), as shown in **figure (1.C&D).**

Out of 40 cases of multiple myeloma, 45% were diffuse pattern as shown in figure (2.A), 42.5% were Interstatial pattern as shown in **figure (2.B)** and 12.5% were patchy pattern as shown in **figure (2.C).**

The characteristics of the patients and tumors are listed in Table (2).There was a significant statistical relation between histopathological type of MM and pattern of infiltration, BMB cellularity and ISS Stage as detailed in **Table (2).**

Immunohistochemical Results:

JAM-A expression in studied groups:

For MM cases, all cases were JAM-A positive, (47.5%) were with low JAM-A expression and (52.5%) were with high JAM-A expression.

Relation between JAM-A expression score and different clinicopathological parameters of studied MM cases:

There was a significant statistical correlation between JAM-A expression and age (p=0.022.), histopathological types (p=0.025) with 64% of mature type showing low JAM-A expression as shown in **Fig (3,A**), 62.5% of immature type showing high JAM-A expression as shown in **Fig (3,B**) and 71% of other types (plasmablastic & anaplastic) showing high JAM-A expression as shown in **Figure (3,C&D)** . There was also a significant statistical correlation between JAM-A expression and pattern of infiltration(p=0.016), BMB cellularity(p=0.017), ISS Stage(p=0.033), R- ISS Stage(p=0.042), Complete response(p=0.008), Resistance(p=0.032), Karyotyping result(p=0.019), one year survival(p=0.012) as shown in **Table (3) .**

Kaplin-Meier survival analysis:

Kaplin-Meier survival analysis showed that one-year overall survival rate in the group of patients with a low expression level of JAM-A was significantly longer than that for patients with a high level of JAM-A (p=.014*) as shown in **figure(4).**

Table (1): Data for using JAM-A antibody in studied cases:

Antibody	Type	Source	Dilution	Positive control	Incubation	Antigen retrival
JAM-A	monoclonal	Biospes, China	1:20	normal endometrium	at 4°Cover night	Protein A affinity purified

Table (2): Relation between histopathological types of MM and different parameters in studied cases:

parameters	Histopathological type of MM				P value
	Total (n=40)	**Mature (n=25(%))**	**Immature (n=8(%))**	**Other types (n=7(%))**	
Age					**0.257**
<60	28	16(57)	6(21.5)	6(21.5)	
≥60	12	9(75)	2(16)	1(8)	
Sex					**0.337**
Male	26	18(70)	4(15)	4(15)	
Female	14	7(50)	4(29)	3(21)	
Pattern of infiltration					**0.042***
Patchy	5	5(100)	0(0)	0(0)	
Interstitial	17	12(71)	2(11)	3(18)	
diffuse	18	8(44.5)	6(33.5)	4(22)	
BMB cellularity					**0.020***
Normocellular	11	10(91)	1(9)	0(0)	
Hypercellular	29	15(52)	7(24)	7(24)	
Pathological fracture					**0.465**
Yes	16	11(69)	3(19)	2(12)	
No	24	14(58)	5(21)	5(21)	
Anemia					**0.698**
Yes	34	22(65)	6(17)	6(18)	
No	6	3(50)	2(33)	1(17)	
Calcium level					**0.688**
-Low	5	4(80)	0(0)	1(20)	
-Normal	32	18(56)	8(25)	6(19)	
-High	3	3(100)	0(0)	0(0)	
Creatinine level					**0.719**
-Normal	27	17(63)	6(22)	4(15)	
-High	13	8(62)	2(15)	3(23)	
Urea level					**0.258**
-Normal	14	10(72)	3(19)	1(7)	
-High	26	15(58)	5(21)	6(23)	
LDH level					**0.692**
-Normal	20	11(55)	6(30)	3(15)	
-High	20	14(70)	2(10)	4(20)	
B2microglobulin level					**1.000**
-Low	2	1(50)	1(50)	0(0)	
-Normal	17	15(88)	1(6)	1(6)	
-High	21	9(42.9)	6(28.6)	6(28.6)	
Albumin level					**0.564**
-Low	21	13(62)	3(14)	5(24)	
-Normal	19	12(64)	5(26)	2(10.5)	
ISS Stage					**0.014***
Stage 1	11	9(82)	2(18)	0(0)	
Stage 2	24	14(59)	6(25)	4(16)	
Stage 3	5	2(40)	0(0)	3(60)	
R- ISS Stage					**0.942**
Stage 1	12	8(67)	3(25)	1(8)	
Stage 2	21	11(52)	5(24)	5(24)	
Stage 3	7	6(86)	0(0)	1(14)	
Free light chain					**0.382**
No	27	17(63)	7(26)	3(11)	
Kappa	8	5(62.5)	1(12.5)	2(25)	
Lambda	5	3(60)	0(0)	2(40)	
Heavy chain					**0.484**
IG g	39	24(61.5)	8(20.5)	7(18)	
IG A	1	1(100)	0(0)	0(0)	
Complete response					**0.794**
Yes	23	15(65)	4(17)	4(17)	
No	17	10(59)	4(24)	3(17)	
Resistance					**0.178**
Yes	13	6(69)	4(31)	0(0)	
No	27	16(59)	4(15)	7(26)	
Relapse					**0.107**
Yes	19	10(52.5)	6(31.5)	3(16)	
No	21	15(71.5)	2(9.5)	4(19)	
Karyotyping					**0.264**
Hypodiploid	6	5(83)	0(0)	1(17)	
Diploid	17	12(70)	2(12)	3(18)	
Hyperdiploid	17	8(47)	6(35)	3(18)	
One year survival					**0.618**
dead	9	4(44.5)	2(22)	3(33.5)	
free	31	21(68)	6(19)	4(13)	

*Significant, BMB: bone marrow biopsy, ISS: international staging system. R-ISS: revised- international staging system

Table (3): Relation between JAM-A expression and different parameters in studied MM cases**.**

parameters	JAM-A score			P value
	Total (n=40)	**Low (n=19(%))**	**High (n=21(%))**	
Age (years)				**0.022***
<60	28	10(36)	18(64)	
≥60	12	9(75)	3(25)	
Sex				**0.822**
Male	26	12(46)	14(54)	
Female	14	7(50)	7(50)	
Histopathologhical types of MM				**0.025***
Mature				
Immature	25	16(64)	9(36)	
Other types	8	3(37.5)	5(62.5)	
	7	2(29)	5(71)	
Pattern of infiltration				**0.016***
Patchy	5	5(100)	0(0)	
Interstitial	17	8(47)	9(53)	
diffuse	18	6(33)	12(67)	
BMB cellularity				**0.017***
Normocellular	10	8(80)	2(20)	
Hypercellular	30	12(40)	18(60)	
Pathological fracture				**0.707**
Yes	16	7(44)	9(56)	
No	24	12(50)	12(50)	
Anemia				**0.464**
Yes	34	17(50)	17(50)	
No	6	2(33)	4(67)	
Calcium level				**0.511**
-Low	5	2(40)	3(60)	
-Normal	32	15(47)	17(53)	
-High	3	2(67)	1(33)	
Creatinine level				**0.909**
-Normal	27	13(48)	14(52)	
-High	13	6(46)	7(54)	
Urea level				**0.285**
-Normal	14	5(36)	9(64)	
-High	26	14(54)	12(46)	
LDH level				**0.355**
-Normal	20	8(40)	12(60)	
-High	20	11(55)	9(45)	
B2microglobulin level				**0.990**
-Low	2	0(0)	2(100)	
-Normal	17	10(59)	7(41)	
-High	21	9(43)	12(57)	
Albumin level				**0.988**
-Low	21	10(48)	11(52)	
-Normal	19	9(47)	10(53)	
ISS Stage				**0.033***
Stage 1	11	8(73)	3(27)	
Stage 2	24	10(42)	14(58)	
Stage 3	5	1(20)	4(80)	
R- ISS Stage				**0.042***
Stage 1	12	5(42)	7(58)	
Stage 2	21	7(33)	14(67)	
Stage 3	7	7(100)	0(0)	
Free light chain				**0.499**
No	27	13(48)	14(52)	
Kappa	8	5(62.5)	3(37.5)	
Lambda	5	1(20)	4(80)	
Heavy chain				**0.299**
IG g	39	18(46)	21(54)	
IG A	1	1(100)	0(0)	
Complete response				**0.008***
Yes	23	15(65)	8(35)	
No	17	4(23.5)	13(76.5)	
Resistance				**0.032***
Yes	13	3(23)	10(77)	
No	27	16(59)	11(41)	
Relapse				**0.209**
Yes	19	7(37)	12(63)	
No	21	12(57)	9(43)	
Karyotyping				**0.019***
Hypodiploid	6	3(50)	3058)	
Diploid	17	13(76.5)	4(23.5)	
Hyperdiploid	17	3(18)	14(82)	
one year survival				**0.012***
dead	9	1(11)	8(89)	
free	31	18(58)	13(42)	

*Significant, BMB: bone marrow biopsy, ISS: international staging system, R-ISS: revised- international staging system, JAM-A: Junctional adhesion molecule –A

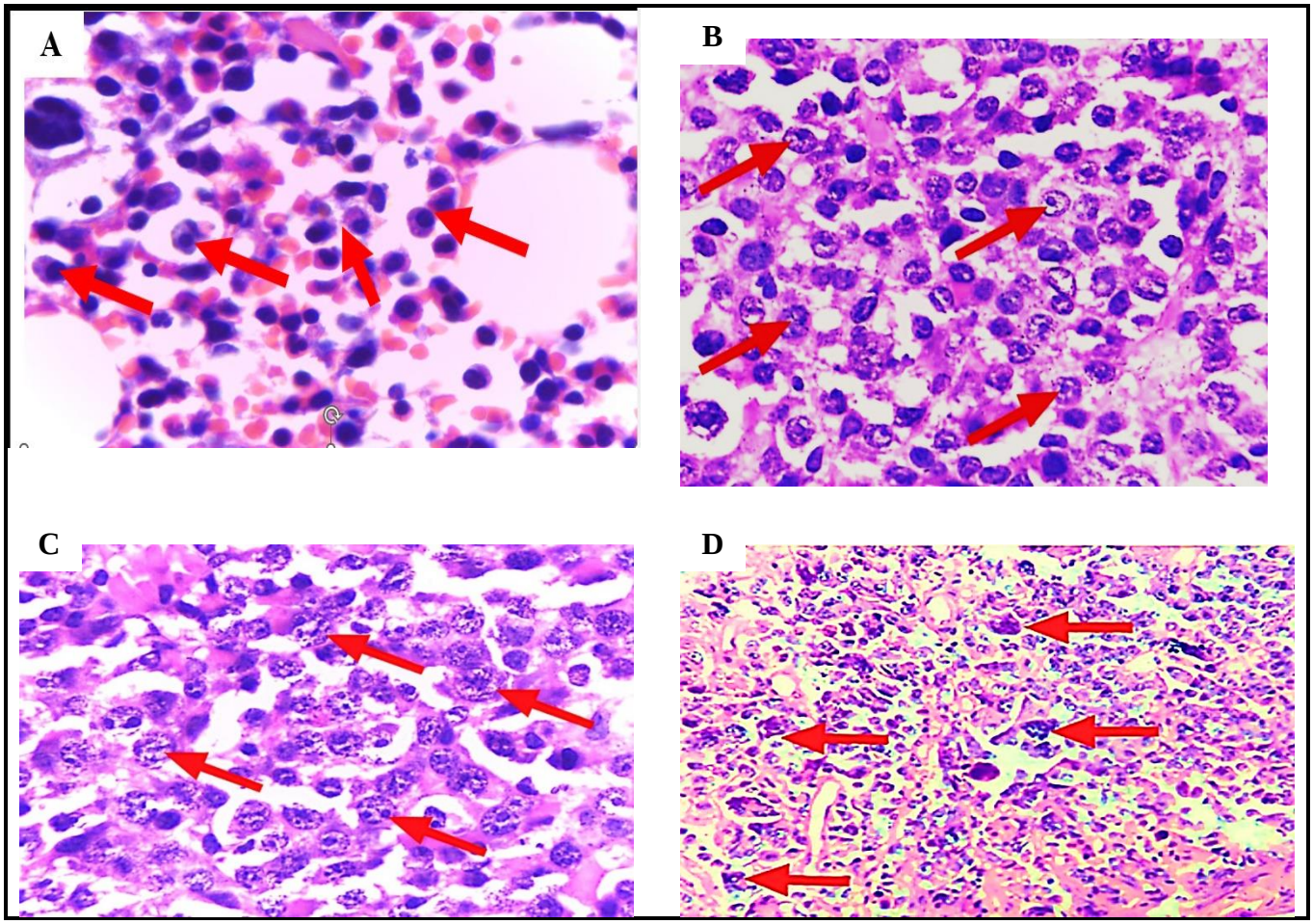

Figure (1): Histopathological types of multiple myeloma: (A): mature type , malignant plasma cells with eccentrically placed nuclei, mature chromatin and abundant basophilic cytoplasm (red arrow), H&E stain x1000. (B): immature type , malignant plasma cells with eccen-trically located large nucleus with one or several nu-cleoli, diffuse chromatin pattern, perinuclear clear area,and variable amount of blue cytoplasm (red arrow), H&E stain x1000. (C): plasmablastic type , malignant plasma cells with fine chromatin, increased nuclear size, large nucleoli and scant cytoplasm (red arrow), H&E stain x1000. (D): anaplastic type , malignant plasma cells with bizarre nuclei and purple bluish granular cytoplasm (red arrow), H&E stain x1000.

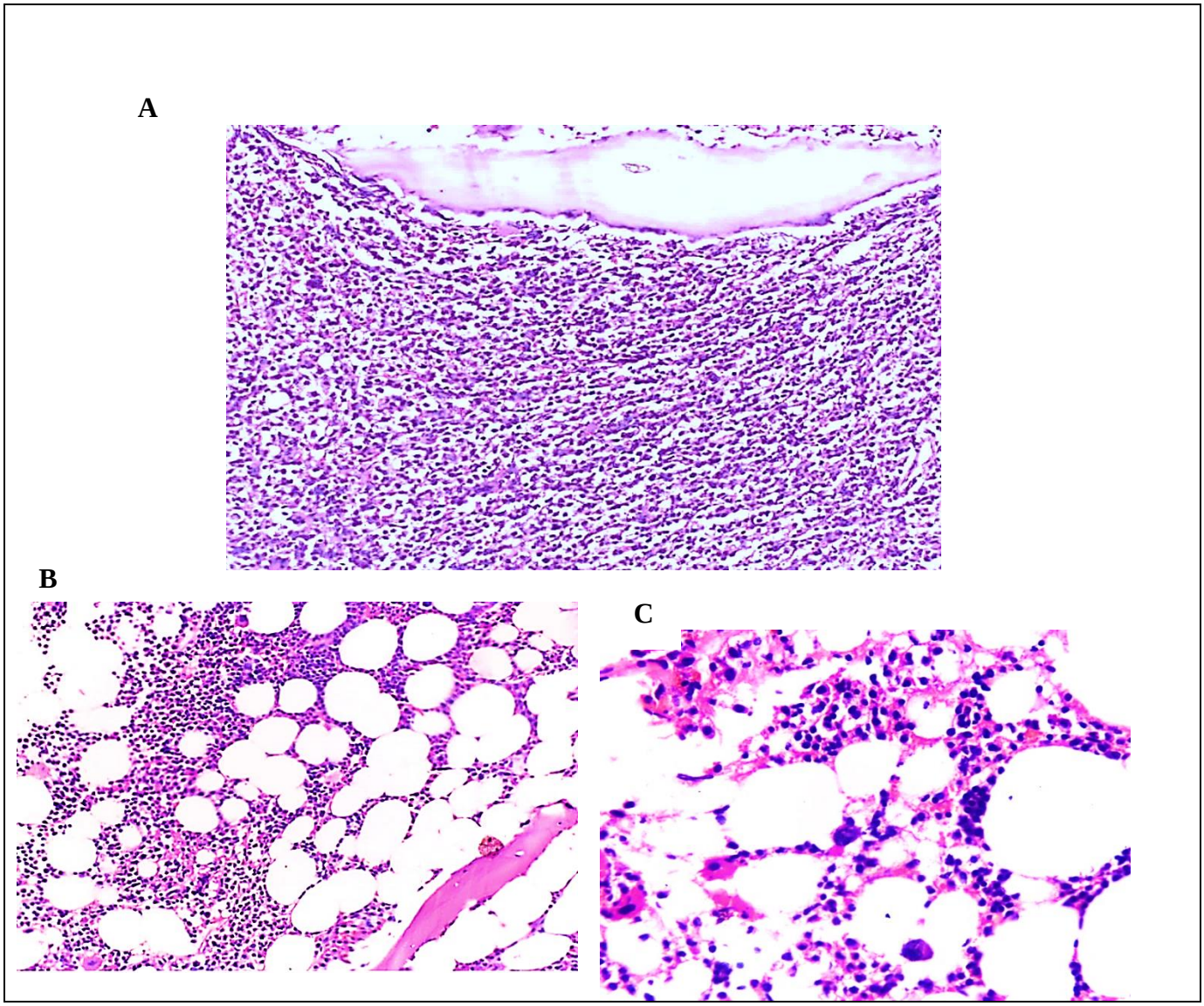

Figure (2): pattern of infiltration of multiple myeloma: (A) diffuse pattern , (B): Interstatial pattern . (C): patchy pattern, (H&E stain x200).

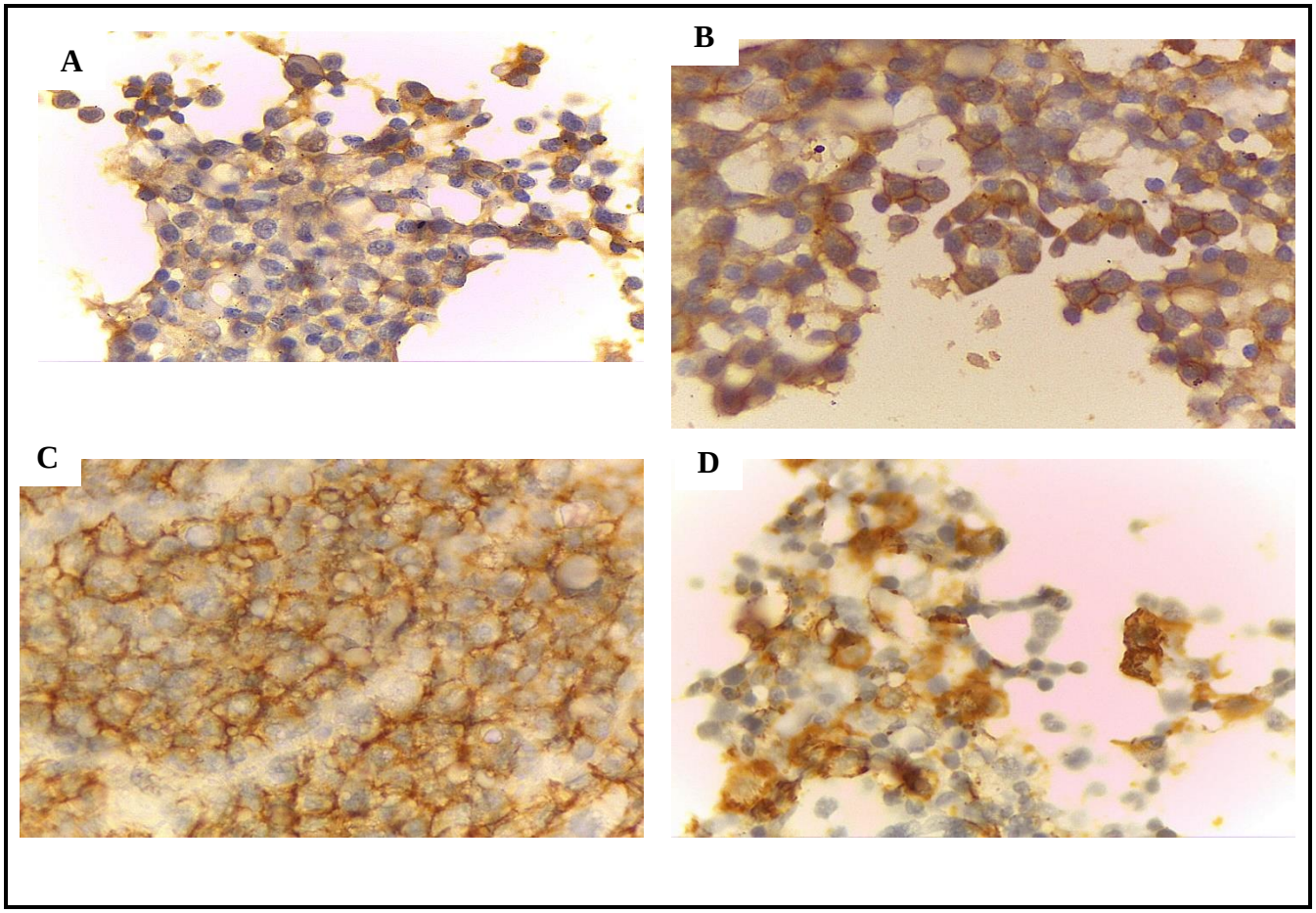

Figure (3): Immunohistochemical expression of JAM-A in studied cases: (A): MM, Mature type, showing low membranous expression for JAM_A with score 4. (B): MM, immature type, higher strong membranous expression for JAM_A with score 12. (C): MM, Plasmablastic type, diffuse strong membranous staining for JAM_A with score 12. (D): MM, Anaplastic type, diffuse strong membranous staining for JAM_A with score 9 (IHC X 1000).

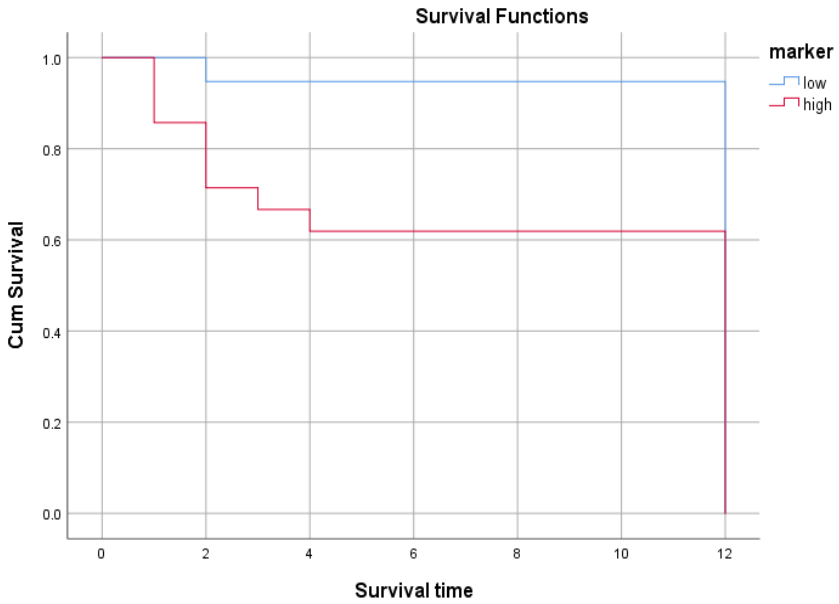

Figure (4): Relation between JAM-A expression and one year survival rate in studied MM cases.

Discussion

Plasma cell disorders - including multiple myeloma (MM) - are experiencing an increase in prevalence worldwide [(16)]. It presents increasing interest due to its uprising frequency both in young and elderly patients [(17)]. It represents approximately 1% of all cancers and approximately 10% of hematological malignancies [(18)].

Drug resistance is one of the major challenges in the treatment of MM, so the incurable nature of MM persists despite the advent of new and innovative therapeutic approaches [(11)].

The aberrant expression or deregulation of JAM-A in various types of human malignancies, such as multiple myeloma, results in a more aggressive phenotype with a poor prognosis, while it regulates cell proliferation and differentiation [(19, 20)].

The current study revealed a statistically significant relation between JAM-A expression and age of studied MM cases (p =.022). This was compatible with previous studies on MM [(21)].

This disagreed with previous studies [(21, 22)] reported that JAM-A was insignificantly correlated with age of studied cases. This could be explained by difference in genetic and environmental factors between their studies and our study.

However; there was no statistically significant correlation between JAM-A overexpression and sex of studied MM cases (p =.822). This agreed with previous studies [(19, 22 and 23)] on non-small cell lung cancer and MM.

Another statistically significant positive relation was reached, between JAM-A overexpression and diffuse pattern of bone marrow infiltration (p =.016). This agreed with a study on MM [(21 & 24)] who reported that JAM-A overexpression was correlated with high bone marrow plasma cell infiltration which whenever increased, the pattern of infiltration progress from local patterns to diffuse pattern. The study they conducted

demonstrated that in vitro JAM-A inhibition impaired MM migration, colony formation, chemotaxis, proliferation, and viability. Tumour progression in a murine xenograft MM model was inhibited by in vivo treatment with an anti-JAM-A monoclonal antibody (αJAM-A moAb).

Also, a previous study on MM [22] reported that diffuse pattern of infiltration associated with higher ISS stage and JAM-A overexpression was correlated with higher stages. So, this may be suggest a correlation between pattern of infiltration and JAM-A overexpression.

A statistically significant positive relation was reached, between JAM-A overexpression and higher BMB cellularity (p =.017). This was compatible with previous study on MM which reported that high JAM-A expression was associated with high plasma cell percentage in bone marrow biopsy in multiple myeloma cases that might be associated with high BMB cellularity [11].

A previous study [22] also reported that high BMB cellularity associated with higher ISS stage and JAM-A overexpression was correlated with higher stages

A statistically significant positive relation was reached, between JAM-A overexpression and poorly differentiated or pleomorphic histopathological type of MM (p =.025), as 62.5% of immature, 71% of plasmablastic & anaplastic showing JAM-A overexpression**.** The double-immunofluorescence results could indicate that JAM-A expression may diminish as the tumour cells become more differentiated, which is consistent with a previous study on MM. **(24)** that reported that JAM-A intensity was significantly higher in grade IV tumours than in grade II tumours of glioma.

This may be attributable to the fact that JAM-A has been identified as an adhesion factor that affects the tumorigenic potential of brain tumor-initiating cells (BTICs), which have the capacity to self-renew and generate new tumours. It was reported in a subsequent study that JAM-A overexpression could promote self-renewal[24].

The receptor/ligand binding affinities of both membrane-bound and soluble JAM-A are high, and they can form homophilic and heterophilic interactions with proteins such as AFDN, tight junction protein-1 (TJP1), calcium/calmodulin-dependent serine protein kinase (CASK), and lymphocyte function associated antigen 1 (LFA-1). Signaling pathways in the downstream region of JAM-A are initiated by interactions such as these, which regulate the survival, growth, angiogenesis, and dissemination of tumour cells[19].

F11R/JAM-A function in the progression of cancer is not only linked to the regulation of cell migration, but also to an influence on apoptosis, epithelial-to-mesenchymal transition (EMT), cancer stem cell maintenance (self-renewal and pro-survival factor), and cell proliferation through the actions of various signaling pathways [25].

Similarly**,** previous studies [22, 26] on MM reported that immature cell morphology is correlated with higher ISS stages and also JAM-A overexpression is correlated with higher ISS stages. So, this suggested a correlation between the previous included histopathological parameters and JAM-A overexpression.

A statistically insignificant correlation was found between JAM-A overexpression and Pathological fracture

(p =.707). A previous study was in a line with our result [19, 24].

In contrast, previous studies [11 , 20 & 27] reported that whenever the volume of MM tumor increased , increased cytokine secretion, increased RANKL and IL-6 causing bone destruction and pathological fracture; all this with JAM-A overexpression.

There was statistically insignificant correlation between JAM-A overexpression with anemia (p =.464), calcium level (p =.511), creatinine (p =.909), urea level (p =.285), LDH level (p =.355) and B2 microglobulin level (p =.990), This was in line with a previous study on MM [11].

But studies on MM [20, 27] showed that JAM-A overexpression is associated with hypercalcemia. This discrepancy may be due to 80% of cases were normal calcium level.

This disagreed with a forementioned study on MM[11] reported that there was a significant correlation with b2 microglobulin, this can be explained by difference in number of cases with elevated B2 microglobulin associated with high JAM-A overexpression.

In this current study, however 52% of studied MM cases with low albumin level showing JAM-A overexpression, the correlation was statistically insignificant (p =.988), this was in a line with previous studies [28, 29] reporting that the albumin level in MM reflect tumor cell burden, which is mainly associated with cytokine induced impairment of albumin synthesis and excessive degradation.

This statistically insignificant correlation might be due to low number of studied cases and different follow up periods.

In the present study, a statistically significant positive relation was found between JAM-A expression with ISS stages (p =.033) & R-ISS staging of MM (p =.042). This was compatible with other studies [19, 21 and 24] reporting that high JAM-A expression is related to higher ISS stage in MM. In addition, previous studies founded that high JAM-A expression was correlated to higher ISS stage of diffuse large B cell lymphoma and epithelial ovarian cancer [30, 31].

The critical function of JAM-A in the bone marrow microenvironment, which contributes to tumour progression and metastasis, was identified in a study on MM. This role is mediated by the regulation of intracellular signaling cascades that are responsible for tumorigenesis and metastasis. This may be due to the vital function of JAM-A in the promotion of tumour progression, which results in tumour invasion and metastasis [25].

This disagreed with previous study [11] who reported that there was insignificant relation between ISS stage of MM and JAM-A expression which was explained by difference in number of cases with high JAM-A expression in higher stages (II-III) between both studies.

In this work, JAM-A overexpression was insignificantly related to free light chain (p =.499) and heavy chain (p =.299) in studied MM cases.

Inversely with this finding, other results carried out by studies [19, 22, 24 and 32] on MM reported that lambda light chain restriction was higher in high-risk patients of MM which had higher ISS stage. This can be explained by most of our cases showing no free light chain (67.5%).

This was compatible with previous studies on MM reported that no dramatic

difference in heavy chain types when made comparison between high and low risk groups [22, 32].

In this work, there was inverse correlation between JAM-A overexpression and the complete response of MM cases (p =.008) as 76.5% of JAM-A overexpression has no complete response. In agreement with this finding, a study on diffuse large B cell lymphoma reported that Patients with high JAM-A expression tended to have low complete remission rate [30].

Another statistically significant correlation was reached, between JAM-A overexpression and resistance to treatment (p =.032) as 77% of resistant cases was JAM-A overexpression. This was in alignment with on MM [11, 33].

MM cells acquire resistance to anti-cancer medications through two interrelated mechanisms: functional and physical interactions with the BM microenvironment. The initial step is the production of soluble factors, such as IL-6, by bone marrow stromal cells (BMSCs). These factors activate downstream signal transduction pathways, resulting in drug resistance. Secondly, they augment the expression of numerous molecules, including cell cycle inhibitors and anti-apoptotic members of the Bcl-2 family, in myeloma cells as a result of direct adhesion[11].

Also, a study on MM founded that there was a significant correlation between JAM-A overexpression and IL-6 [11], so this suggested a correlation between JAM-A overexpression n and drug resistance.

In this work, although 63% of relapsed MM cases was JAM-A overexpression but had no significant correlation (p =.209). Studies on MM revealed that relapse is more common with high JAM-A expression as this overexpression mostly associated with resistance to treatment which a cause to relapse as reported in other studies on MM [21, 24 and 33].

Also, a study on the glioblatoma reporting that JAM-A expression was significantly higher in recurrent glioblatomas (GBMs) than primary GBMs ($p < 0.001$) [24].

The current study reached a statistically significant relation between JAM-A overexpression and karyotyping result (p =.019). This was in accordance with studies that indicated that 70.8% of genetic high-risk patients had membrane JAM-A levels above the median on the day of biopsy. In the genetic intermediate risk group, this percentage decreased to 46.2%, and among the standard genetic risk patients, it decreased to 34% [11, 21 and 24].

In this work, JAM-A overexpression was significantly related to short one year survival in studied MM cases (p =.012). Also, Kaplan-Meier survival analysis, found that patients with high expression of membrane JAM-A had a significant lower survival than patients with low expression (p =.014). In accordance with this finding, other studies [19, 21, 22, 24 and 34] It was reported that the median progression-free survival of subjects with higher membrane JAM-A expression levels was significantly different from that of those with lower expression. This discrepancy was determined to be statistically significant (P=0.0022). Furthermore, additional research has demonstrated that the overexpression of JAM-A is linked to a reduced survival rate in breast cancer, lung cancer, and ovarian cancer [31, 35 and 36].

In contrast, previous studies found that low JAM-A expression is associated with poor outcomes and short overall

survival in pancreatic cancer, gastric cancer and endometrial cancer [37, 38 and 39]. The prognostic value of JAM-A expression in cancers may be organ specific.

Conclusion

JAM-A overexpression might have an important role in proliferation, invasion, progression, poor outcome and resistance to treatment in patient of multiple myeloma. So, JAM-A might have a major targeting and promising value in treatment of MM.

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