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Molecular Characteristics of Bladder Tumor: Increased Gene Expression of MAGE-A6 and MAGE-A11 with Decreased MicroRNA-34a and MicroRNA-125b

Azin Aghamajidi[1], Zahra Ousati Ashtiani[2], Monireh Mohsenzadegan[3], Nader Tajik[1], Mahmood Ghafoori Yazdi[4], Laleh Sharifi[4], and Mohammad Reza Nowroozi[4]

[1] Department of Immunology, School of Medicine, Iran University of Medical Sciences, Tehran, Iran
[2] Department of Medical Genetics, School of Medicine, Tehran University of Medical Sciences, Tehran, Iran
[3] Department of Medical Laboratory Science, Faculty of Allied Medical Sciences, Iran University of Medical Sciences, Tehran, Iran
[4] Uro-Oncology Research Center, Tehran University of Medical Sciences, Tehran, Iran

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ABSTRACT

Bladder cancer is recognized as one of the top ten most common cancers worldwide. Activation of oncogenes, inactivation of tumor suppressor genes, and dysregulation of androgen signaling pathways are three major pathophysiological causes in the development of bladder tumors. Discovering potential biomarkers is required for the management and immunotherapy of bladder cancer. Melanoma-associated antigen (MAGE)-A6 and MAGE-A11 are two cancer-testis antigens that are potential coregulators of androgen receptors. MicroRNAs, especially miR-34a and miR-125b are two important tumor suppressors that play a critical role in regulating different signaling pathways and inhibiting tumor development.

Twenty-nine surgical tissue biopsies were collected from patients with no preoperative chemotherapy or radiotherapy (26 males and, 3 females, mean age±SD: 62.4±13.3 years). Seventeen adjacent uninvolved tissues with no abnormalities upon histological examination were considered normal controls (14 males and, 3 females, mean age±SD: 64.2±7.4 years) . Quantitative PCR was performed to evaluate the gene expression level of MAGE-A6, MAGE-A11, miR-34a, and miR-125b in bladder cancer biopsies.

MAGE-A6 and MAGE-A11 expressions were significantly increased in bladder tumors compared with normal tissues. However, the expression levels of miR-34a and miR-125b were significantly downregulated in bladder tumor tissues. Interestingly, the expression level of all these genes was significantly associated with tumor grade, pathological stage (pT), and muscular invasion.

MAGE-A6 and MAGE-A11 can be considered potential markers for the diagnosis and immunotherapy of bladder tumors. Furthermore, the modulation of miR-34a and miR-125b gene expression in association with increased MAGE-A6 and MAGE-A11 genes could open a new horizon in the improvement of bladder cancer.

Keywords: Bladder cancer; Immunotherapy; MAGE-A6; MAGE-A11; MicroRNA-34a; MicroRNA-125b

Corresponding Author: Monireh Mohsenzadegan, PhD; Department of Medical Laboratory Science, Faculty of Allied Medical Sciences, Iran University of Medical Sciences (IUMS), Tehran, Iran.

Tel: (+98 21) 8670 4681, Fax: (+98 21) 8862 2533, E-mail: monirehmohsenzadegan@gmail.com

INTRODUCTION

Bladder cancer is one of the most common malignancies of the urinary and reproductive system, which is a prevalent cause of cancer-related deaths worldwide.[1,2] The incidence of bladder cancer is increasing globally, especially in developed countries.[2] Moreover, the prevalence rate of bladder tumors is higher in men than in women.[3] According to the European Association of Urology (EAU), bladder cancer is mainly classified into two different types: non-muscle-invasive bladder cancer (NMIBC) and muscle-invasive bladder cancer (MIBC).[1] This malignancy is also divided into low- and high-grade tumors, which are the slow-growing and most aggressive types of bladder cancer, respectively.[4] It has been shown that molecular characteristics of bladder cancer are strongly associated with the activation of oncogenes and the inactivation of tumor suppressor genes including fibroblast growth factor receptor (FGFR), retinoblastoma (Rb), and p53 tumor suppressor gene.[5] On the other hand, the androgen receptor (AR) signaling pathway plays a fundamental role in the pathophysiology of bladder cancer.[6] Therefore, it is reasonable to perceive that men are more susceptible to developing bladder tumors than women.[7]

Recently, immunotherapy has emerged as the most effective approach to treating various types of cancers.[8] Cancer/testis antigens (CTAs) are potential immunotherapeutic targets with restricted expression in normal tissues.[9,10] In particular, the melanoma-associated antigen-A (MAGE-A) family are members of CTAs, which consist of highly conserved proteins. The MAGE-A family is frequently identified in most cancer types, including breast, prostate, and urothelial tumors.[10,11] Furthermore, MAGE-A expression is associated with clinical and pathological features of aggressive cancers.[12] Interestingly, MAGE-A6 and MAGE-A11 are considered potential coregulators of AR signaling that are coexpressed in AR-mediated human prostate cancer.[13]

Alternatively, microRNAs (miRs) are recognized as small non-coding RNAs that play a critical role in the post-transcriptional regulation of the target gene.[14] are considered oncogenes or tumor suppressors in different types of cancer.[15] In general, miR-34a and miR-125b are essential tumor suppressors that play a major role in inhibiting tumor development. Dysregulation of miR-34a and miR-125b is characterized in several human cancer tissues.[16,17] MAGE-A6 is a direct target for miR-34a, which can modulate the MAGE-A6 gene expression.[18] Also, miR-125b is identified as a negative coregulator of tumor suppressor genes which can upregulate the gene expression of miR-34a. It has also been identified that there is a cross-regulation between miR-34a and miR-125b, which exert inhibitory effects on IL-6 receptors as a direct target of miR-34a and signal transducer and activator of transcription (STAT) loop. The inhibition of this inflammatory pathway results in the upregulation of miR-34a expression,[16] In addition, miR-34a and miR-125b are negatively correlated with AR expression playing a fundamental role in AR signaling.[19] A comprehensive understanding of the molecular mechanisms involved in the development of bladder tumors could shed light on a more effective treatment for patients with bladder cancer. Hence, the objectives of this study were to investigate the gene expression of MAGE-A6, MAGE-A11 as well as miR-34a and miR-125b in patients with bladder cancer and normal subjects.

MATERIALS AND METHODS

Subjects and Tumor Samples

Surgical tissue biopsies with no preoperative chemotherapy or radiotherapy were collected between December 2020 and July 2021 from patients referred to the Uro-Oncology Research Center of Imam Khomeini Hospital Complex (IKHC) and Sina University Hospital, Tehran, Iran. Patients with autoimmune diseases or other types of cancer and those who had received any type of treatment, including immunosuppressive or immunomodulatory drugs, were excluded from the study. Twenty-nine patients who had not received any prior therapy were eligible for inclusion (26 males and 3 females; mean age±SD: 62.4±13.3 years) with a histologically confirmed diagnosis of NMIBC and MIBC (n=14 and n=15, respectively). Adjacent uninvolved tissues with no abnormalities upon histological examination of seventeen age- and sex-matched individuals were considered normal controls (14 men and 3 women, mean age±SD: 64.2±7.4 years) (p=0.42). Demographic and clinical characteristics of the patients are reported in Table 1. Medical documents and hematoxylin and eosin (H&E) stained slides were examined to compile clinical and pathological features, including age, gender, histological grade, pT stage, and muscular invasion. All tissue samples were collected during transurethral resection of a bladder tumor

(TURBT), and preserved in RNALater (Qiagen, Phoenix, AZ, USA). Written informed consent was obtained from each participant and the research proposal was confirmed by the Research Ethics Committee of Iran University of Medical Sciences (IR.IUMS.FMD.REC.1400.440).

RNA Extraction, Gene Expression, and MicroRNA Quantitation

Total RNA was extracted from each tissue sample using TRIzol (Sinaclon, Tehran, Iran) according to the manufacturer's protocol. The quantity and quality of extracted RNA were measured with an optical density 260/280 absorption ratio of >1.8 using a NanoDrop spectrophotometer (Thermo Fisher Scientific, Waltham, Massachusetts, USA) and agarose gel electrophoresis, respectively. The cDNA synthesis was performed from total RNA (1 µg) using the miScript Reverse Transcription Kit (Parstous, Tehran, Iran) with random hexamers in the thermal cycler according to the manufacturer's protocols to evaluate the mRNA expression of MAGE-A6 and MAGE-A11. The expression levels of the human MAGE-A6 and MAGE-A11 genes were measured using pairs of primers synthesized by Pishgam Biotech Co. (Tehran, Iran) (Table 2). Glyceraldehyde 3-phosphate dehydrogenase (GAPDH) was considered the housekeeping gene. Quantitative real-time PCR (qPCR) was carried out using qPCR SYBER Green Master Mix (BioFact, Daejeon, Korea) on Rotor-Gene Q (QIAGEN, Hilden, Germany) at 95°C for 15 minutes, followed by 40 cycles at 95°C for 15 seconds and 61.5°C for 25 seconds.

The specific forward and reverse primers of miR-34a and miR-125b were purchased from Pars Genome Co. (Tehran, Iran). Then the qPCR was carried out using the miScript SYBR Green PCR Kit (Pars Genome, Tehran, Iran) on Rotor-Gene Q (QIAGEN, Hilden, Germany) at 95 °C for 15 minutes followed by 40 cycles at 95°C for 30 seconds and 65°C for 30 seconds and 72°C for 30 seconds. The 5S ribosomal RNA (5S rRNA) was used as an internal control for normalization (Table 3). All measurements were performed in triplicate, threshold cycles (CT) were normalized, and relative expression was determined by the $2^{-\Delta\Delta Ct}$ formula.

Table 1. Patient demographics and baseline characteristics

Classification	Total Sample
Group	
Patient	29 (63)
Gender	
Male	26 (89.5)
Female	3 (10.5)
Age	
(*Median age* in y*ears)*	
≤66	16 (55)
>66	13 (45)
Histological grade	
Low	11 (38)
High	18 (62)
Muscular invasion	
Involved	15 (51.7)
None	14 (48.3)
pT stage	
pTa	0
pT1	12 (41.4)
pT2	9 (31)
pT3	3 (10.3)
pT4	5 (17.3)

Table 2. Relative gene expression levels of MAGE-A6, MAGE-A11

Classification	Total Sample N (%)	Relative Expression of MAGE-A6 *	*p* value	Relative Expression of MAGE-A11 *	*p* value
Group					
Normal	17 (37)	1.75±0.28	≤0.0001	1.04±0.07	≤0.0001
Patient	29 (63)	3.77±0.27		8.69±1.46	
Age					
(Median age in years)			0.03		0.01
≤66	24 (52.2)	3.69±0.34		6.41±1.42	
>66	22 (47.8)	2.6±0.32		5.26±1.41	
Histological Grade					≤0.0001
Low	*11 (38)*	2.22±0.46	≤0.0001	1.56±0.11	
High	*18 (62)*	5.09±0.25		13.04±2.05	
Muscular Invasion			0.004		0.02
Involved	*15 (51.7)*	4.48±0.34		10.02±2.26	
None	*14 (48.3)*	3.01±0.39		7.26±1.84	
pT stage					
pTa	*0*				
pT1	*12 (41.4)*	2.44±0.32	≤0.0001	4.7±1.55	≤0.0001
pT2	*9 (31)*	4.35±0.44		4.9±1.07	
pT3	*3 (10.3)*	5.44±0.52		26.55±4.1	
pT4	*5 (17.3)*	4.59±0.89		6.07±1.13	

*Values are presented as mean±SD or number (%).

Statistical Analysis

Normal distribution of the data was examined by the Kolmogorov-Smirnov test. Mann-Whitney U test was used to compare gene expression in the patients and normal groups as well as gene expression with clinical and pathological outcomes in patients with bladder cancer. The correlation between data sets was evaluated by the Spearman correlation test. The association and correlation between gene expression and clinical and pathological characteristics were also analyzed using Pearson's chi-square, R test. Data are presented as Mean±SEM, and *p* values<0.05 were considered significant.

RESULTS

MAGE-A6 and MAGE-A11 Expressions were Significantly Increased in Bladder Tumors

Expression levels of MAGE-A6 and MAGE-A11 were evaluated in bladder cancer biopsies compared to normal tissues. The association between MAGE-A6 and MAGE-A11 expressions and clinical and pathological parameters of bladder cancer cases is reported in Table 1. The median age of the study population (patients and normal group) was 66 years (range 33–99). MAGE-A6 and MAGE-A11 relative expressions were increased in bladder tumors (Figures 1A and B) (p<0.0001).

Table 3. Relative gene expression levels of miR-34a and miR-125b.

Classification	Total Sample N (%)	Relative Expression of miR-34a *	*p* value	Relative Expression of miR-125b *	*p* value
Group					
Normal	17 (37)	1.1±0.29	0.001	0.87±0.03	≤0.0001
Patient	29 (63)	0.39±0.06		0.31±0.05	
Age					
Median age (Years)			0.4		0.7
≤66	24 (52.2)	5.74±2.7		0.5±0.06	
>66	22 (47.8)	1.12±0.27		0.53±0.05	
Histological Grade			≤0.0001		≤0.0001
Low	*11 (38)*	0.85±0.11		0.74±0.06	
High	*18 (62)*	0.1±0.01		0.05±0.005	
Muscle Invasion					
Involved	*15 (51.7)*	0.17±0.04	≤0.0001	0.11±0.03	≤0.0001
None	*14 (48.3)*	0.62±0.11		0.53±0.08	
pT Stage					
pTa	*0*				
pT1	*12 (41.4)*	0.7±0.12	≤0.0001	0.61±0.08	≤0.0001
pT2	*9 (31)*	0.23±0.06		0.16±0.05	
pT3	*3 (10.3)*	0.07±0.02		0.02±0.004	
pT4	*5 (17.3)*	0.11±0.04		0.05±0.01	

* Values are presented as mean±SD or number (%).

Furthermore, the relative expressions of both MAGE-A6 and MAGE-A11 were higher in high-grade tumors than in low-grade tissues (Figures 1C and D) (p<0.0001). Also, MAGE-A6 and MAGE-A11 expressions were significantly higher in MIBC patients vs. NMIBC (p=0.004 and p=0.020, respectively) (Table 1). There was a remarkable correlation between the mRNA expression of MAGE-A6 (p<0.03)and MAGE-A11with median age (p<0.01) (Table 1).

Combined Analysis of MAGE-A6/MAGE-A11 Expression

To classify MAGE-A6 and MAGE-A11 phenotypes in the patient group, the mean of gene expression was evaluated as a laboratory set-up cut-off point. Expression levels above the cut-off point were considered 'high expression'. Also, expression levels below the cut-off point were regarded as 'low expression'. Therefore, the expression of the MAGE-A6 and MAGE-A11 phenotypes was classified into 4 subgroups: MAGE-A6$^{Low\ (L)}$/MAGE-A11^{L} phenotype, 16 (55.2%);MAGE-A6$^{High\ (H)}$/MAGE-A11^{L}, 5 (17.2%); MAGE-A6^{L}/MAGE-A11^{H}, 0 (0%) and MAGE-A6^{H}/MAGE-A11^{H}, 8 (27.6%). Pearson's chi-square analysis was used to examine the correlation between the expression of MAGE-A6/MAGE-A11 phenotypes and clinical and pathological parameters. Along with a significant correlation of MAGE-A6 and MAGE-A11 with some clinical and pathological parameters described in Table 1, significant differences were also found between MAGE-A6/MAGE-A11 phenotypes with different histological grades (p<0.0001), pT stage (p<0.0001) and muscular invasion (p=0.003). These results showed a direct and positive relationship between the mentioned parameters and MAGE-A6/MAGE-A11 phenotypes. As the histological grade, pT stage, and muscular invasion increased, the expression of MAGE-A6/MAGE-A11 also increased.

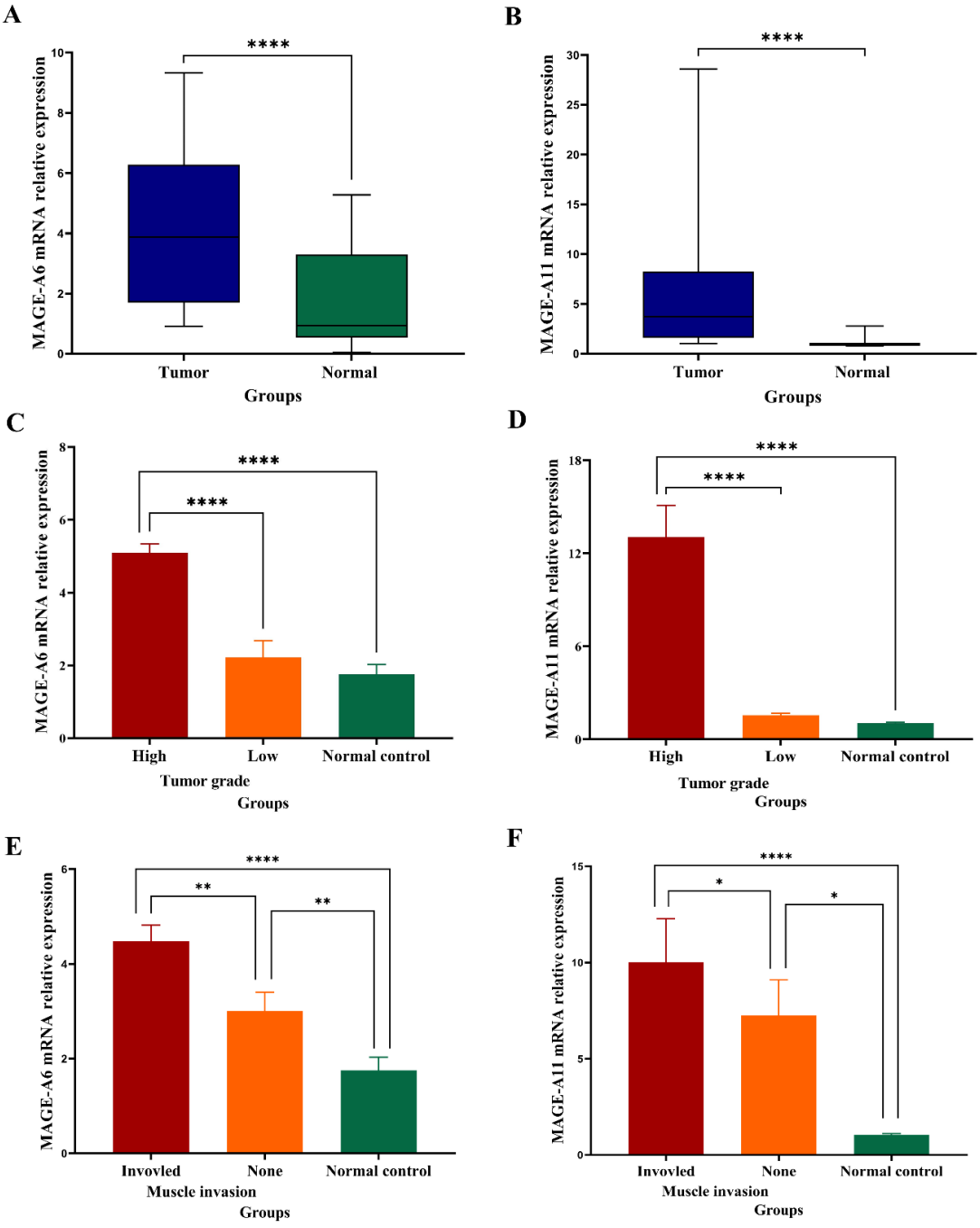

Figure 1. The gene expression levels of MAGE-A6 and MAGE-A11 in twenty-nine bladder cancer cases and seventeen non-tumoral bladder tissues. (A) The relative expression of MAGE-A6 and (B) MAGE-A11 are significantly higher in bladder cancer biopsies than in normal tissues. (C) The relative expression of MAGE-A6 and (D) MAGE-A11 are significantly higher in high-grade bladder cancer biopsies than in low-grade tumors and normal tissues. (E) The relative expression of MAGE-A6 and (F) MAGE-A11 are significantly higher in muscle-invasive bladder cancer biopsies compared to non-muscle-invasive tumors and normal tissues. Bar graphs represent the tumor tissues compared to control as assessed by Mann-Whitney U test. *p<0.05, **p<0.01, ***p<0.001, ****p<0.00001

Reduced Expression of miR-34a and miR-125b in Bladder Tumors

The expression levels of miR-34a and miR-125b were analyzed as regulators of MAGE-A6 and MAGE-A11. The association between miR-34a and miR-125b expressions and clinical and pathological parameters of bladder cancer patients is demonstrated in Table 1. The expression levels of miR-34a and miR-125b were significantly reduced in the patient group compared with healthy subjects (Figure 2A and B) (p=0.001 and p<0.0001, respectively). Interestingly, the relative expressions of both miR-34a and miR-125b were lower in high-grade tumors than in low-grade tissues (Figures 2C and D) (p<0.0001). Moreover, the expression levels of both miR-34a and miR-125b were significantly reduced in muscle-invasive bladder tumors compared with bladder tumors with no muscle invasion (p<0.0001) (Table 1). There is a significant difference between the relative expression of miR-34a and miR-125b in clinical pT stages of bladder tumors (both p values were <0.0001) (Table 1).

Combined Analysis of miR-34a/miR-125b Expression

To classify miR-34a and miR-125b phenotypes in the patient group, the mean of gene expression was evaluated as laboratory set-up or arbitrary cut-off point. Expressions above the cut-off point were considered 'high expressions'. Also, expressions below the cut-off point were defined as 'low expressions'. Therefore, the expression of miR-34a and miR-125b phenotypes were classified into 4 subgroups including miR-34a$^{Low\ (L)}$/miR-125b^{L} phenotype, n=18 (62.1%); miR-34a$^{High\ (H)}$/miR-125b^{L}, n=0 (0%); miR-34a^{L}/miR-125b^{H}, n=2 (6.9%); and miR-34a^{H}/miR-125b^{H}, n=9 (31%). Pearson's chi-square analysis was used to examine the correlation between the expression of miR-34a/miR-125b phenotypes and clinical and pathological parameters. Along with a significant correlation of miR-34a and miR-125b with some clinical and pathological parameters described in Table 1, significant differences were also found between miR-34a/miR-125b phenotypes with different histological grades (p<0.0001), pT stage (p<0.0001), and muscular invasion (p<0.0001). These results showed an inverse relationship between the mentioned parameters and miR-34a/miR-125b phenotypes. Simultaneously, as histological grade, pT stage, and muscular invasion increased, expression of miR-34a/miR-125b decreased.

Correlation between MAGE-A6, MAGE-A11, miR-34a, and miR-125b

Surprisingly, the obtained results indicate a positive correlation between the expression levels of MAGE-A6 and MAGE-A11 (r = 0.723, p<0.0001). Furthermore, the expression level of miR-34a was significantly correlated with miR-125b (r=0.491, p<0.0001). Interestingly, there was a negative correlation between miR-34a and MAGE-A6 (r=−0.374, p<0.0001) and MAGE-A11 (r = −0.506, p<0.0001). Also, there was a negative correlation between miR-125b and MAGE-A6 (r = −0.518, p<0.0001) and MAGE-A11 (r=−0.788, p<0.0001). There was no significant correlation between MAGE-A6, MAGE-A11, miR-34a, and miR-125b with age and gender (p>0.05).

TCGA Dataset Analysis

The Cancer Genome Atlas (TCGA) dataset analyses were performed to evaluate MAGE-A6 and MAGE-A11 expression in bladder cancer tumors and normal tissues. The results showed that the expressions of MAGE-A6 and MAGE-A11 genes were significantly upregulated in bladder tumors compared with normal tissues (Figure 3). Further TCGA dataset analyses revealed a moderate positive correlation (r=0.44, p<0.0001) between MAGE-A6 and MAGE-A11 expression levels (Figure 4).

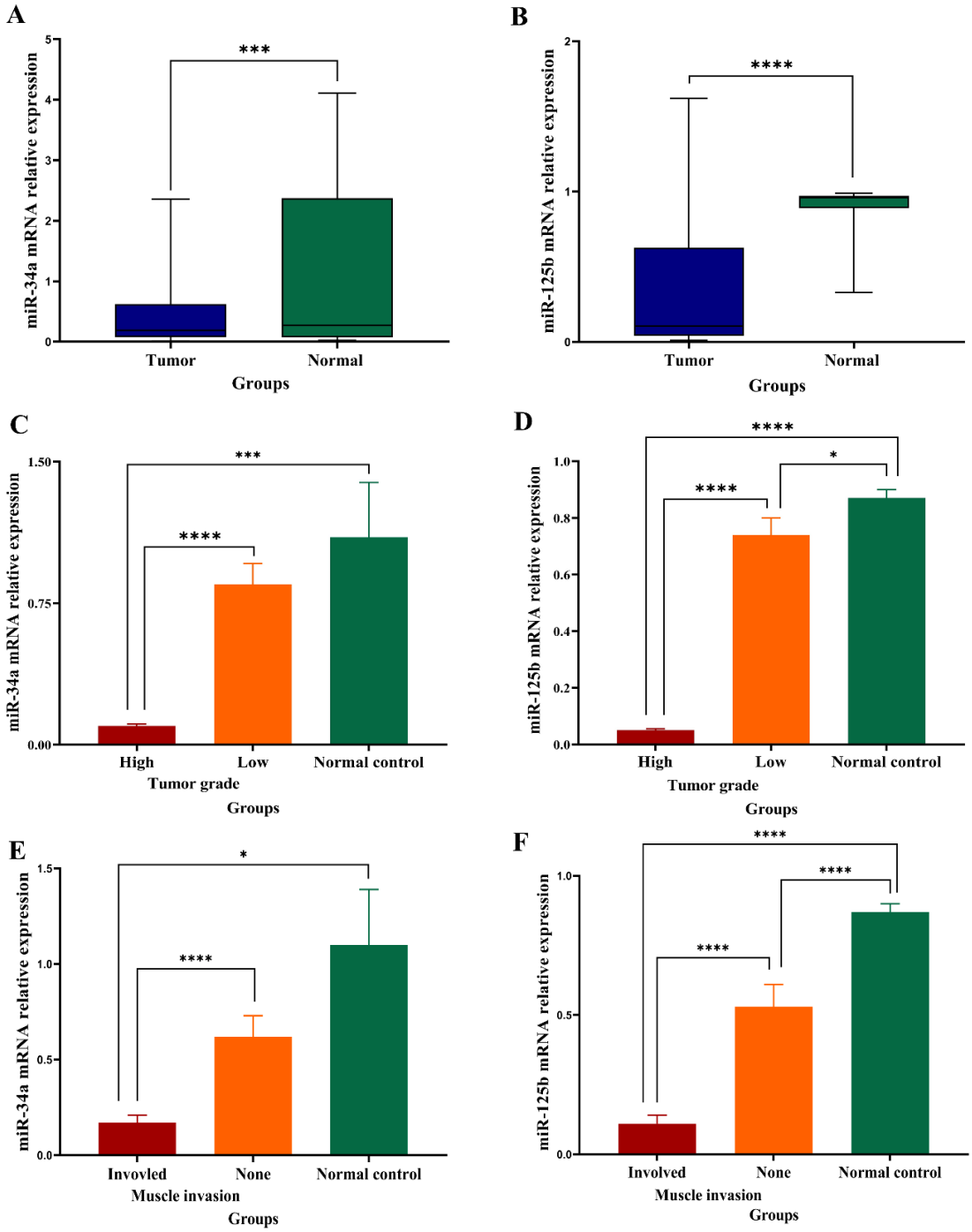

Figure 2. The gene expression level of miR-34a and miR-125b in twenty-nine bladder cancer cases and seventeen non-tumoral bladder tissues. (A) The relative expression of miR-34a and (B) miR-125b are significantly lower in bladder cancer biopsies than in normal tissues. (C) The relative expression of miR-34a and (D) miR-125b are significantly reduced in high-grade bladder cancer biopsies than in low-grade and normal tissues (E) The relative expression of miR-34a and (F) miR-125b are significantly lower in muscle-invasive bladder cancer biopsies compared to non-muscle-invasive and normal tissues. (Bar graphs represent the tumor tissues compared to controls as assessed by Mann-Whitney U test. *p<0.05, **p<0.01, ***p<0.001, ****p<0.00001

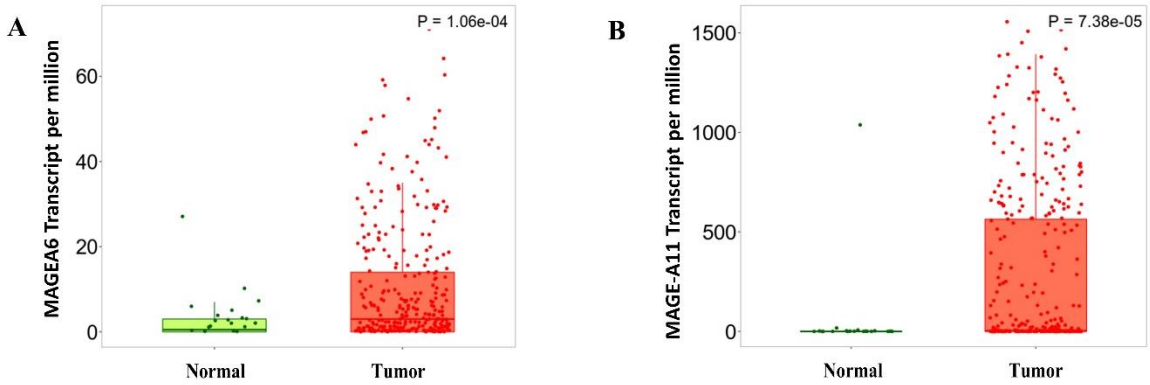

Figure 3. Relative expression of MAGE-A6 and MAGE-A11 in bladder 412 tumors and normal tissues based on the data provided by TCGA. The expression of MAGE-A6 and MAGE-A11 mRNA is significantly higher in tumors than in normal tissues. Transcript per million is used as normalization method for RNA-seq indicated the number of genes expressed.

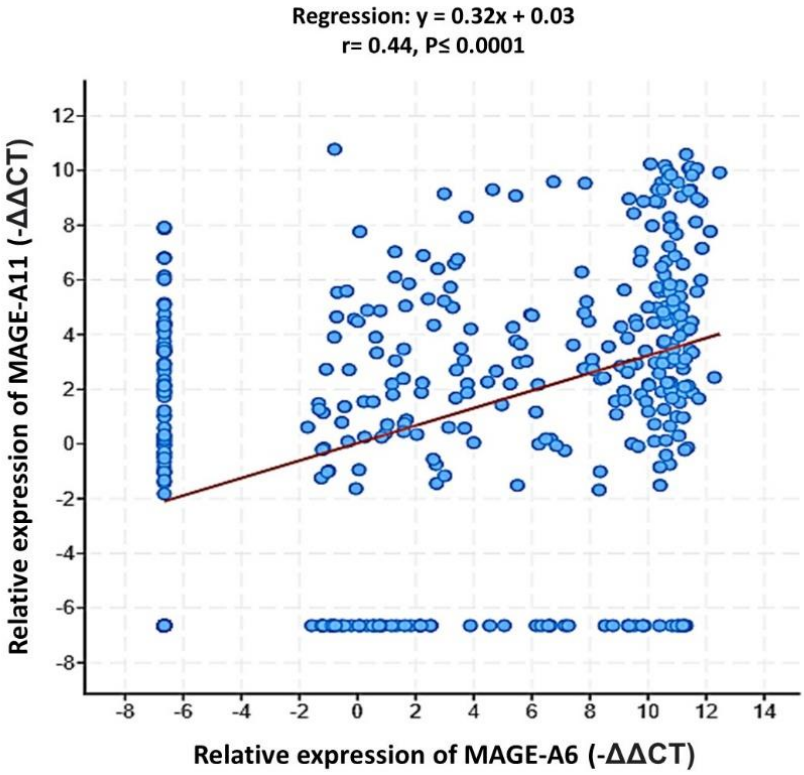

Figure 4. A significant correlation between the expression of MAGE-A6 and MAGE-A11 genes in 412 bladder tumors based on the data provided by The Cancer Genome Atlas (TCGA). There is a positive correlation between the mRNA levels of MAGE-A6 and MAGE-A11. Each dot indicates an individual sample.

DISCUSSION

Bladder cancer is the most common malignancy of the urinary system, which is steadily increasing worldwide[20] Immunotherapy has been used as a practical approach in the treatment of bladder cancer; however, discovering a potential therapeutic target remains a subject of debate. Hence, understanding the molecular mechanisms involved in the pathogenesis of bladder cancer could provide new insights into cancer treatment. This study investigated the expression levels of MAGE-A6, MAGE-A11, miR-34a, and miR-125b genes as well as their association with clinical and pathological characteristics in patients with bladder cancer.

P53 mutation and AR-dependent mechanisms play an essential role in the development and progression of bladder cancer.[21] It has been demonstrated that AR is highly expressed in bladder tumors, which is related to the high prevalence of bladder cancer in men (22). On the other hand, the MAGE-A family consists of 13 highly immunogenic proteins upregulated in various cancers, including urothelial carcinoma and breast cancer.[23,24] Recent evidence has suggested that MAGE-A proteins could inhibit the function of p53.[9,25] Among the MAGE-A family, MAGE-A11, which is a coregulator of AR, enhances androgen signaling in prostate cancer.[26,27] The functional interaction between MAGE-A6 and MAGE-A11 promotes AR activity in prostate cancer.[13] In this study, MAGE-A6 and MAGE-A11 were highly expressed in bladder tumors compared with normal tissues. These findings are consistent with the data provided by TCGA. Also, there is a significant difference between the expression levels of MAGE-A6 and MAGE-A11 in high- and low-grade tumors. Furthermore, the data revealed a significant association of MAGE-A6 and MAGE-A11 mRNA expressions with muscle invasion, pT stage, and aggressive pathological parameters. These findings are consistent with our previous MAGE-A6 and MAGE-A11 protein expression study using an immunohistochemical approach in patients with bladder cancer. The immunohistochemical analysis was similar to the quantitative real-time PCR results in having a strong expression in the bladder cancer tissues and a positive association with aggressive pathological parameters (data not published). Therefore, it is reasonable to presume that the increased levels of MAGE-A6 and MAGE-A11 may associate with a poor prognosis in patients with bladder cancer. Besides, due to the important role of AR signaling in the development of bladder cancer and the complex formation of MAGE-A6/MAGE-A11 in the activation of AR,[13] the upregulation of MAGE-A6 and MAGE-A11 in bladder tumor tissues possibly promotes AR signaling and mediates bladder tumor progression.

Generally, miRs play a fundamental role in the post-transcriptional regulation of target gene expression.[28] Mainly miR-34a is regulated through the tumor suppressor p53 and suppresses the expression of MAGE-A6 by direct binding to the 3′ UTR of its mRNA.[29] Our results showed that miR-34a is significantly decreased in bladder tumors. Particularly, a reverse relationship was found between MAGE-A6 and MAGE-A11 expressions with miR-34a expression. These findings indicate that the reduced levels of miR-34a may affect MAGE-A6 and MAGE-A11 expression levels in bladder cancer patients. However, more studies are required to confirm this association in patients with bladder cancer. According to a study by Misso et al., miR-125b upregulates miR-34a gene expression.[16] and can directly target the p53 gene, which inhibits the tumor suppressor signaling pathway.[16] In this study, miR-125b expression was strongly reduced in bladder tumor tissues. It also decreased significantly in high-grade tumors compared to low-grade tumors. The results showed a significant positive correlation between miR-34a and miR-125b gene expression. Therefore, it can be perceived that a reduced level of miR-34a and miR-125b possibly influence p53, eventually resulting in tumor development. Significantly, miR-34a and miR-125b expressions were inversely correlated with the MAGE-A6 and MAGE-A11 gene expressions. These findings indicate that miR-34a and miR-125b may affect the expression of MAGE-A6 and MAGE-A11 in bladder cancer such that the suppression of miR-34a and miR-125b results in the increased expression of MAGE-A6 and MAGE-A11 (Figure 5).

Taken together, MAGE-A6 and MAGE-A11 can be considered as potential markers for the diagnosis, prognosis, and immunotherapy of patients with bladder cancer. The modulation of miR-34a and miR-125b gene expression in association with increased MAGE-A6 and MAGE-A11 genes could open a new horizon in improving bladder cancer.

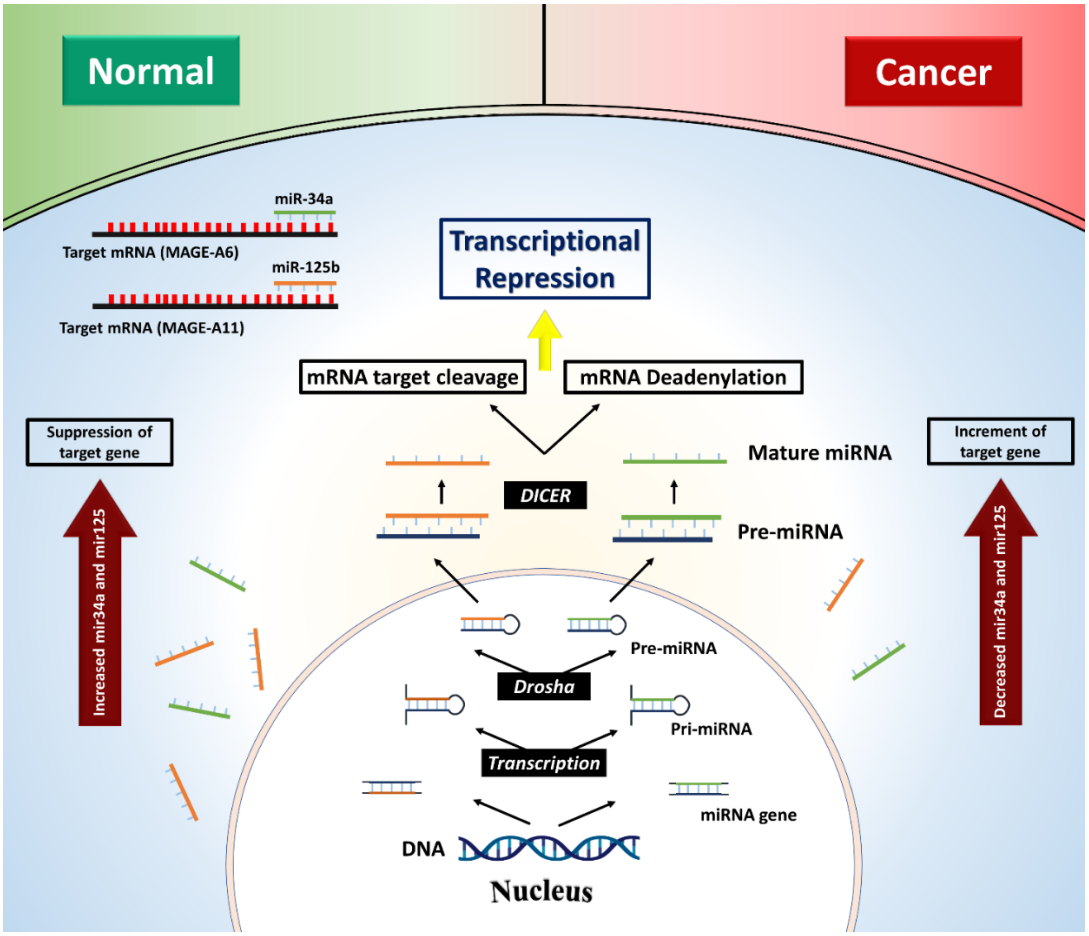

Figure 5. Target gene regulation by miR-34a and miR-125b. MicroRNA (miRNA) is processed by RNA polymerases II or III, leading to the production of the primary miRNA transcript (pri-miRNA). Then, Drosha processes the cleavage of the pri-miRNA in the nucleus. Pre-miRNA is exported from the nucleus, which could mediate target mRNA cleavage or mRNA deadenylation and transcriptional repression. Downregulation of miRNA, especially mir-34a and miR-125b may increase MAGE-A6 and MAGE-A11, which promote tumor development and cancer.

Data Availability

The data used to support the findings of this study are available from the corresponding author upon request.

STATEMENT OF ETHICS

This paper reflects the original work in a truthful and complete manner which is not currently being considered for publication elsewhere.

FUNDING

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CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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