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It is with great pleasure that I write this editorial to welcome you to the IJCBR. This journal provides a platform for publication of original and reviews research articles, short communications, letter to editor, thesis abstract, conference report, and case studies. These types of publication are directed at the interface of the fields of cancer and biomedical research.

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Mohamed L. Salem,

Editor in Chief

INTERNATIONAL JOURNAL OF CANCER AND BIOMEDICAL RESEARCH (IJCBR)

RESEARCH ARTICLE

Role of expression of p53 and Ki67 in the progression of Wilms tumor: Correlation with patients' survival

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ABSTRACT

Background: Wilms tumor (WT) is the most frequent renal tumor of childhood and is a highly responsive tumor to chemotherapy. P53 and ki67 are two of the most important markers that have been evaluated in many cancers. The role of p53 in the pathogenesis and progression of WT is only partly understood. Over the past decade, the importance of Ki-67 in the prognosis of breast cancer has been widely studied. On the other hand, fewer studies are available for this in WT. **Aim:** To study the possible role of p53 and ki67 in the progression and outcome of Wilms tumor cases. **Methods:** Immunohistochemical (IHC) staining for p53 and ki67 were done and correlated with clinicopathological data of Wilms tumor cases. **Results:** There was a positive correlation between p53 and ki67 expression and unfavorable histology, higher tumor stage, shorter disease-free survival (DFS) and overall survival (OS). **Conclusion:** Ki67 and p53 expression was positively correlated with aggressive behavior of Wilms tumor.

Keywords: Wilms tumor, anaplasia, p53, ki67.

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INTRODUCTION

Primary renal tumors account for 4–7% of all childhood cancers. In the Western world, Wilms tumor (WT) or nephroblastoma represents about 90% of these cases. Renal tumors are common in children aged less than 4 years but their relative frequency decreases in older age groups (Steliarova-Foucher et al., 2017). According to the cancer pathology registry, national cancer institute, Cairo University; nephroblastoma represents 31.03% of renal neoplasms (Mokhtar et al., 2016). Moreover, the incidence of Wilms tumor in Africa based on WHO data for children ages 0-14 y., reported an incidence of 6 per million in Egypt (Cunningham et al., 2020).

Treatment modalities of WT include surgery, chemotherapy and, for some patients, radiotherapy. Long-term cure rates have improved to >90% with the introduction of multimodal treatment (Brok et al., 2016). Tumor histology either favorable or unfavorable and tumor stage are the main determining factors of chemotherapy protocol of WT (Oostveen and Pritchard-Jones, 2019) . Despite the excellent prognosis for most children with WT, about 15% of patients will relapse, usually within 2 years of diagnosis (Pritchard-Jones et al., 2015). Furthermore, a proportion of patients will display severe early and late treatment-related complications, e.g. cardiotoxicity secondary to doxorubicin or radiotherapy-induced organ dysfunction, musculoskeletal abnormalities, infertility and secondary malignancies (Termuhlen et al., 2011). Therefore, the identification of ideal cost-effective prognostic markers that reduce these complications would be beneficial (Krishna et al., 2016).

Wilms tumor represents a genetically heterogeneous group, displaying a high degree of intra-tumoral heterogeneity (Malogolowkin et al., 2008). The tumor suppressor marker p53 and tumor proliferation marker (Ki67) are two of the most important markers that have been evaluated in many cancers. Ki67 is a nuclear

antigen associated with cell proliferation which is present throughout the cell cycle and is absent in resting cells (Berrebi, 2008).

The p53 tumor suppressor gene (Tp53) is located on chromosome 17p13.1. In a normal healthy cell, P53 behaves as a multifunctional transcription factor involved in the control of the cell cycle, programmed cell death, senescence, differentiation, deoxyribonucleic acid (DNA) replication and DNA repair. p53 prevents neoplastic transformation by three interlocking mechanisms: activation of temporary cell cycle arrest (quiescence), induction of permanent cell cycle arrest (senescence), or triggering of programmed cell death (apoptosis) (Wei et al., 2006).

The role of p53 in the pathogenesis and progression of WT is only partly understood. The significance of p53 expression in WT is conflicting. Several studies have been carried out to determine the role of this protein expression as a prognostic factor in patients harboring WT. Some of these studies have confirmed the correlation of TP53 overexpression with anaplasia and prognosis in WT (Franken et al., 2013; Ooms et al., 2016); others revealed no correlation between p53 and Wilms tumor development (Krishna et al., 2016). Over the past decade, the importance of Ki67 prognosis of breast cancer has been widely studied as regard prognosis and treatment (Abubakar et al., 2019). On the other hand, with respect to WT, several studies are conducted (Menon et al., 2019).

PATIENTS AND METHODS

Patients

This is a retrospective study including 38 patients with Wilms tumor who were presenting to the pediatric oncology unit, Oncology Center-Mansoura University-OCMU, from January 2010 to July 2018. Formal consent was obtained from the children's guardians/ parents before data and sample collection. This study was approved by the Institutional Review Board (IRB code no: R/18.08.258).

The study groups

The International Society of Pediatric Oncology treatment policy of patients with WT was followed for our cases. This included preoperative chemotherapy then followed by surgery and further chemotherapy with or without radiotherapy, if necessary. Histologically, three risk groups are identified (a) low risk (cystic partially differentiated nephroblastoma), (b) intermediate-risk (regressive, epithelial, stromal, or mixed), and (c) high risk (blastemal or diffuse anaplasia). A diagnosis of anaplasia was made according to National Wilms' Tumor Study Group criteria (Beckwith, 1983).

Clinical data were obtained from the patient's clinical sheets including the patient's age, gender, tumor stage & tumor recurrence if present. Hematoxylin & Eosin (H&E) slides were reviewed to confirm the diagnosis and assess other histopathological parameters such as anaplasia and blastemal predominant histology by blinded review. Exclusion criteria include complete necrosis of the tumor following chemotherapy and incomplete clinical data.

Immunohistochemistry

Immunohistochemical staining of the formalin-fixed, paraffin-embedded tissue sections (4-µm thick) was performed following the technique of Hsu et al., 1981). Immunohistochemical staining was done of resection specimens. Epitope retrieval was performed with the use of heat-induced epitope retrieval technique using Cell Marque triology in conjunction with a pressure cooker. Each antibody was supplied in a concentrated form (Genemed; Biotechnologies, Mouse monoclonal anti-p53, clone BP-53-12; Mouse monoclonal anti-ki67, clone GM010) and diluted in the ratio of 1:100 in Phosphate-buffered saline. Then, they were incubated with UltraVision One AP Polymer for 30 min at room temperature. Then, subsequent ordinary steps were performed. Only cases with clear nuclear staining for p53 and ki67 were considered positive. ki67 was graded as low, moderate, and high. The expression of proliferation index Ki-67 is categorized into 3 groups: low (Ki-67 ≤15%), moderate (Ki-67:16-30%), and high (Ki-67 > 30%) according to the recommendations of the St Gallen International Consensus of Experts (Goldhirsch et al., 2011). Immunohistological upregulation of p53 was noted to be present if 20% or more of the nuclei stained positive (Krishna et al., 2016).

Statistical analysis

Data were analyzed using the Statistical Package of Social Science (SPSS) program for Windows (Standard version 21). The normality of data was first tested with a one-sample Kolmogorov-Smirnov test. Qualitative data were described using the number and percent. Association between categorical variables was tested using Chi-square test while Fischer exact test and Monte Carlo test were used when expected cell count less than 5.

Continuous variables were presented as mean ± standard deviation for parametric data and median for non-parametric data. The two groups were compared with the Student t-test for parametric data and Mann Whitney test for non-parametric. ANOVA test was used to compare more than 2 means, while Kruskal-Wallis test was used to compare more than 2 medians.

Cox regression analysis was used to predict the most significant determinants of predictors of mortality among significant variables on univariate analysis using the enter statistical technique. Survival analysis was tested by Kaplan-Meier and Log-rank test to determine the statistical significance of differences among curves. For all above mentioned statistical tests done, the threshold of significance is fixed at $\leq$ 5% level (p-value).

RESULTS

Study group

In total, 38 cases of Wilms tumor were identified from hospital records between January 2010 to July 2018. Two cases were excluded due to defective medical reports and another 2 cases were due to complete necrosis of the tumor after receiving chemotherapy.

Clinicopathological criteria

The clinicopathological features of the studied patients are listed in Table 1. There was a female predominance (64.7%). Age at presentation ranged from 1 to 13 years with a median age of 3 years. Clinical presentations were variable, with abdominal mass being the most common (73.5%), followed by incidentally discovered (32.4%), hematuria (20.6%) and abdominal pain (20.6%). Distribution of the tumor stage was 38.2% for stages I and II, 61.8% for stages III and IV. About 60% of the cases were left-sided. Of the studied 34 cases, 15 (44.1%) cases were alive at the end of the current study , and 19 (55.9%) succumbed to the disease. As regard tumor characteristics, the tumor size mean was 10.67 cm. The predominant histology was the triphasic histology (97.1%) in the form of blastemal, epithelial, and mesenchymal elements (Figure 1d). There was predominance for favorable histology 65% (Figure 1d) as compared to 35% of the cases that showed anaplasia; either focal or diffuse (Figure 1a).

Immunohistochemical expression and correlation with patients' clinicopatholgical parameters

Regarding p53 immunoreactivity, about 60% of the studied cases showed a positive reaction (Fig. 1b), as compared to 40% with a negative reaction (Fig. 1e). When comparing p53 expression with histology, all tumors with unfavorable histology showed a positive reaction for p53**.** In respect to tumors with favorable histology, 68.2% showed negative reactions while only 31.8% of these tumors were positives ($p < 0.001$). Moreover, there was a positive correlation between p53 positivity and the presence of abdominal pain ($p=0.011$), higher tumor stage ($p=0.002$), and higher Ki67 immunoreactivity ($p < 0.001$). Moreover, there was a significant relation between positive p53 expression and decreased both overall survival and disease-free survival of our studied cases (Figure 2a,b).

Regarding ki67 immunoreactivity; 29.4% showed mild positive reaction for ki67, 35.3% showed moderate reaction (Fig. 1f) and the same percentage showed high reaction (Fig. 1c). Moreover, there was a positive correlation between increased ki67 positivity and higher tumor stage ($p=0.002$), higher risk group ($p=0.005$) and higher p53 immunopositivity ($p=0.014$). When comparing ki67 expression with histology, 66.7% of tumors with unfavorable histology showed high ki67 expression in comparison to 8.3 % that showed low expression. On the other hand, only 18.2% of tumors with favorable histology showed high expression with an equal percentage for low

and moderate expression (each represented 40.9%) (p=0.014). As noted in p53, there was a significant relationship between increased ki67 expression and decreased both overall survival and disease-free survival of our studied cases (Figure 2c,d).

Table 1. Descriptive data of the studied cases.

Variables	Total (n=34)
Age/ year; Median (Min-Max)	3.00 (1-13)
Sex, n (%) Male Female	 12 (35.3) 22 (64.7)
Clinical Presentation, n (%)	
Abdominal mass Hematuria Abdominal pain Accidental	25 (73.5) 7 (20.6) 7 (20.6) 11 (32.4)
Tumor stage, n (%) Stage 1 & 2 Stage 3 & 4	 13 (38.2) 21 (61.8)
Laterality, n (%) Left Right	 19 (55.9) 15 (44.1)
Tumor Size (cm); Mean ± SD	10.67±3.30
Histology, n (%) favorable unfavorable	 22(64.7) 12(35.3)
Histologic subtype, n (%) Triphasic Blastema only	 33(97.1) 1(2.9)
Risk group, n (%) low intermediate high	 1(2.9) 28(82.4) 5(14.7)
Recurrence, n (%)	7 (20.6)
Mortality, n (%) Died Survived	 19(55.9) 15(44.1)
P53, n (%) Negative Positive	 15(44.1) 19(55.9)
ki67, n (%) Mild Moderate high	 10 (29.4) 12(35.3) 12(35.3)

The relationships between the p53 and ki67 expression and clinicopathological data of the studied cases were represented in Table 2. By applying multivariate Cox regression analysis to test the prognostic yield of these prognostic factors; higher tumor stage (p=0.025), p53 positivity (p=0.001) and high ki67 positivity (p=0.007) were independent predictors of mortality (Table 3).

DISCUSSION

In this study, 34 cases of WT were studied. The male to female ratio was 1:1.8. This result is in discordance with most of the previous studies in which there was male predominance that ranged from 1.1 to 2 (table 4). However, other studies that reported female predominance like our study (Krishna et al., 2016). Age at presentation was wide-ranged from 1-13 years with median age of 3 years which is in concordance with some previous studies (Sredni et al., 2001; Mitchell et al., 2006; Chan et al., 2014). On the other hand, other studies reported lower mean age at presentation (Trehan et al., 2012; Sangkhathat et al., 2018) and others reported higher age (Anyanwu et al., 2015; Pribnow et al., 2017). High age at presentation may be attributed to the late age of presentation of the patient to the health care center, especially in less developed countries (Cunningham et al., 2020). The most common clinical presentation was abdominal mass (73.5%). This result was in concordance with other studies (Levitt et al., 2012; Illadea et al., 2018; Seminara et al., 2019).

About tumor laterality, all cases were unilateral with about 60% being left-sided. This contrasted with the study conducted by Seminara, et al. 2019 that found more prevalent right-sided tumors of their studied cases (57%) (Seminara et al., 2019). Right-sided predominance was also observed by Tang et al. (2019) (Tang et al., 2019). Moreover, there was predominance for higher tumor stages (Stage III and IV). These findings are like some studies (Sredni et al., 2001; Jadali et al., 2011; Salama and Kamel , 2011). However, patients of other studies were presented in earlier stages (Beniers et al.,2001; Skotnicka-Klonowicz et al., 2002; Franken et al., 2013; Krishna et al., 2016).

The high percentage of stages III and IV in the present study may be attributed to the lack of awareness by general practitioners of the probability of cancer and later presented with a more advanced stage in the tertiary hospital (Cunningham et al., 2020). Also, this late presentation may be the cause of the high mortality rate of our patients (about 60%). As regard to tumor characteristics, the tumor size mean was 10.67 cm.

Table 2. Relation between p53 and ki67 expression and clinicopathological data of the studied cases.

Variables	P53			Ki67			
	P53 positive (n=19)	P53 negative (n=15)	p-value	Mild (n=10)	Moderate (n=12)	Severe (n=12)	p-value
Age (year) Median (Min-Max)	3.00 (1-10)	3.00 (1-13)	0.609	3.00 (1-13)	3.00 (1-11)	3.00 (1-10)	0.842
Sex, n (%)			0.610				0.648
Male	6(31.6)	6(40)		4(40)	3(25)	5(41.7)	
Female	13(68.4)	9(60)		6(60)	9(75)	7(58.3)	
Clinical presentation, n (%)	13(68.4)	12(80)	0.447	9(90)	9(75)	7(58.3)	0.243
Abdominal mass							
Hematuria	6(31.6)	1(6.7)	0.104	1(10)	2(16.7)	4(33.3)	0.370
Abdominal pain	7(36.8)	0(00)	0.011*	4(40)	2(16.7)	1(8.3)	0.172
Accidental	7(36.8)	4(26.7)	0.529	4(40)	3(25)	4(33.3)	0.752
Tumor stage, n							
(%)	3(15.8)	10(66.7)	0.002*	7(70)	6(50)	0(0)	0.002*
Stage 1 & 2	16(84.2)	5(33.3)		3(30)	6(50)	12(100)	
Stage 3 & 4							
Laterality, n (%)							
Left	12(63.2)	7(46.7)	0.336	4(40)	6(50)	9(75)	0.226
Right	7(36.8)	8(53.3)		6(60)	6(50)	3(25)	
Tumor Size, Mean ± SD	10.94±3.56	10.33±3.03	0.599	10.60±3.27	9.83±3.04	11.58±3.62	0.444
Recurrence, n (%)	6 (31.6)	1(6.7)	0.104	0(0)	2(16.7)	5(41.7)	0.051
Histology, n (%)							0.014*
favorable	7(36.8)	15(100)	<0.001*	9(90)	9(75)	4(33.3)	
unfavorable	12(63.2)	0(0)		1(10)	3(25)	8(66.7)	
Histologic subtype, n (%)			1				1
Triphasic	18(94.7)	15(100)		9(90)	9(75)	4(33.3)	
Blastema only	1(5.3)	0(0)		1(10)	3(25)	8(66.7)	
Risk group, n (%)							
low							
intermediate	0(0)	1(6.7)	0.052	1(10)	0(0)	0(0)	0.005*
high	14(73.7)	14(93.3)		9(90)	12(100)	7(58.3)	
	5(26.3)	0(0)		0(0)	0(0)	5(41.7)	
ki67, n (%)	1(5.3)	9(60)	<0.001*	**p53**,n(%)	9(75)	4(33.3)	0.014*
Mild	6(31.6)	6(40)		Positive	3(25)	8(66.7)	
Moderate	12(63.2)	0(0)		9(90)			
High				Negative			
				1(10)			

Abbreviations: N, number; SD, standard deviation.

Tang et al. reported a predominance of cases with large tumor size > 7 cms, like our finding (Tang et al., 2019). Triphasic histology was the predominant histology (97.1%). About 35% showed anaplasia; either focal or diffuse. Percentages of unfavorable histology were variable among previous studies (Table 4).

A high percentage of anaplasia in our study may be due to the aggressive behavior of our cases that was presented also by a higher percentage of Stage III and IV. This may be due to the late presentation of the studied cases to our oncology center.

In respect to p53 immunostaining, about 56% of cases showed a positive reaction for p53. In the previous studies, p53 positivity among studied cases ranged from 8% to 60.3% (Table 4). Moreover, p53 immunoreactivity in the current study was significantly associated with the presence of abdominal pain, unfavorable tumor histology, higher tumor stage and higher ki67 immunostaining.

Table 3, Cox regression analysis of independent predictors of mortality

Independent predictor	β	P - value	HR (95%CI)
Tumor stage Stage 1 & 2 (r) Stage 3 & 4	1.727	0.025*	5.6 (1.2-25)
Recurrence Yes No (r)	-0.662	0.440	0.51 (0.09-2.8)
P53 Positive Negative (r)	3.06	0.001*	21.3 (3.6-74)
ki67 Mild (r) Moderate High	- 1.72 2.99	- 0.08 0.007*	1 5.6 (0.8-38) 20 (2.3-75)

CI: confidence interval; HR, Hazard ratio; r, reference.

Table 4. Previous studies that were examined the role of p53 IHC in Wilms tumor cases.

Ref.	NO of cases	Age	M:F ratio	Stage I, II (%)	Stage III, IV (%)	Unfavorable histology (%)	P53 positivity (%)	Correlation with unfavorable histology	Correlation with higher stage	Correlation with poor survival
Cheah et al., 1996	38	1m-6y	1.2:1	NA	NA	5.3	36.8	Yes	No	No
Lahoti et al., 1996	28	4-120m	2:1	50	50	50	60	Yes	Yes	Yes
Govender et al., 1998	93	4m-14ys (3.7)	1:1.1	43	57	7.5	8.6	Yes	Yes	Yes
Beniers et al., 2001	21	44(2-132)	1.1:1	66.7	33.3	19	33.3	Yes	borderline	Yes
Skotnicka-Klonowicz et al., 2001)	61	39m (2d-13ys)	1.1:1	63.9	36.1	18	37.7	Yes	Yes	Yes
Sredni et al., 2001	97	3ys (1m-21 ys)	1:1.1	47.4	52.6	7.2	13.4	No	Yes	Yes
D'Angelo et al., 2003	63	3(1-8)	NA	NA	NA	10	8	Yes	No	No
Salama and Kamel, 2011	63	NA	1:1	34.9	65.1	6.4	60.3	Yes	Yes	Yes
Jadali et al., 2011	44	36(4-96)	1.43:1	43.2	56.8	25	54.7	Yes	No	Yes
Franken et al., 2013	48	3.75 (3–9.1)	1.1:1	79.2	20.8	8.3	NA	Yes	Yes	Yes
Krishna et al., 2016.	31	3-8	1:1.8	90.3	0.7	29	NA	No	No	No

Abbreviations; M, month; NA, not available; ys, years

The positive relation between p53 expression and the presence of abdominal pain may be explained by that Increased p53 expression which is caused by mutation of Tp53 gene enables accumulating mutation (Rivlin et al., 2011). This in turn causes increased tumor aggressiveness, tumor size or I. ntratumoral hemorrhage that causes stretching of the renal capsule (Davidoff et al., 1998). This is may be a possible cause of pain. The positive relation between positive p53 expression and unfavorable tumor histology was observed by most of the reviewed previous studies except for (Sredni et al., 2001; Krishna et al., 2016). This finding is logical because Tp53 mutation allows the accumulation of other mutations that give the nuclei more atypical morphology and more frequent atypical mitoses.

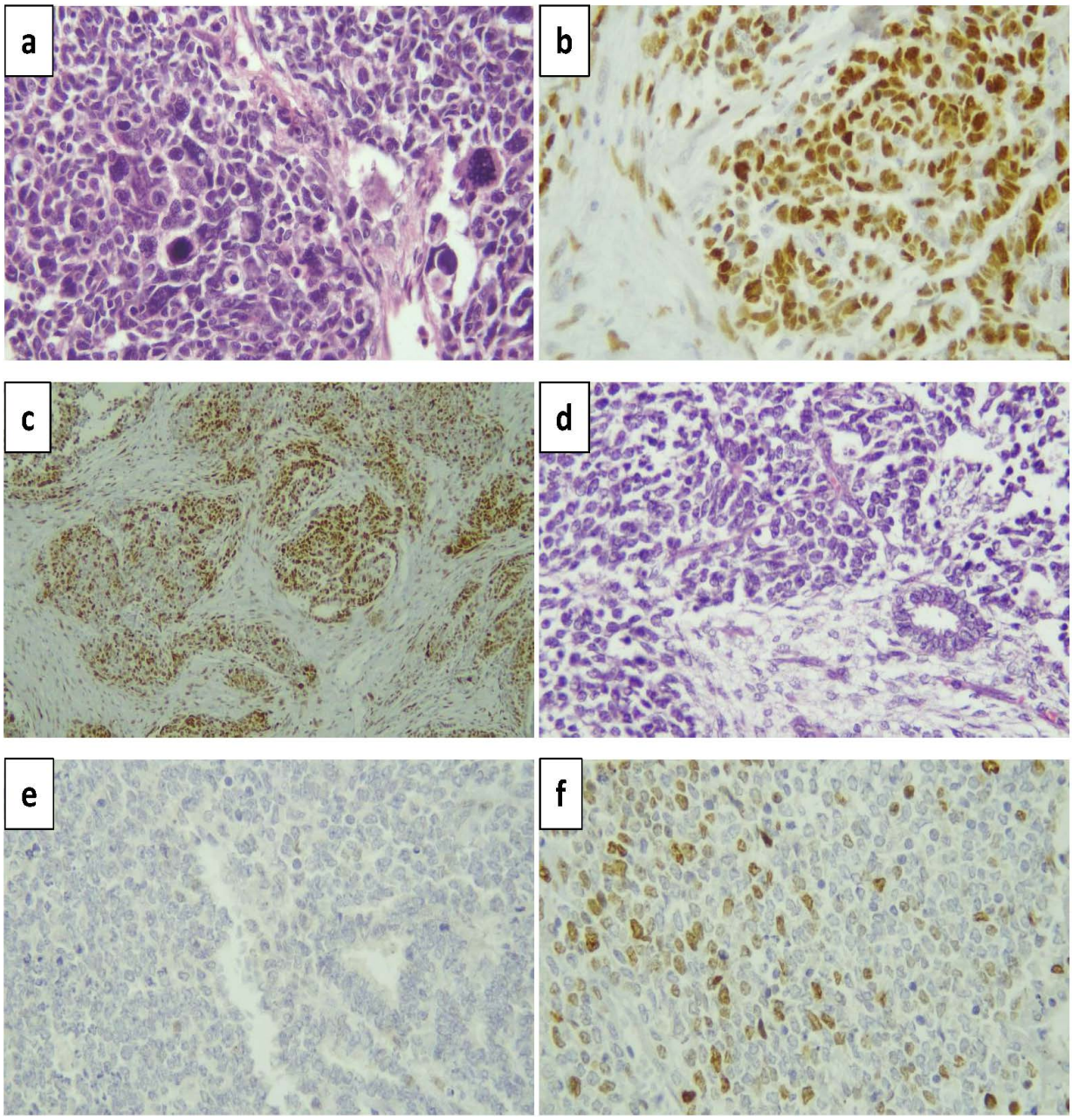

Figure 1. Immunohistochemical data (a) A case of Wilms tumor with unfavorable histology (diffuse anaplasia) that shows an increase in the greatest nuclear dimension at least three times that of adjacent nuclei of the same cell type, nuclear hyperchromasia and abnormal multipolar mitotic figures (H&E x400). (b) A case of Wilms tumor with unfavorable histology (diffuse anaplasia) that shows an immunohistochemical nuclear positivity for p53 (p53 x400). (c) A case of Wilms tumor with unfavorable histology (diffuse anaplasia) that shows a high immunohistochemical nuclear positivity for ki67 (ki67 x200). (d) A case of Wilms tumor with favorable histology that shows a favorable histology formed of blastemal, epithelial and mesenchymal elements (triphasic histology) (H&E x400). (e) A case of Wilms tumor with favorable histology that shows an immunohistochemical negativity for p53 (p53 x400). (f) A case of Wilms tumor with favorable histology that shows a moderate positive nuclear reaction for ki67 (ki67 x400).

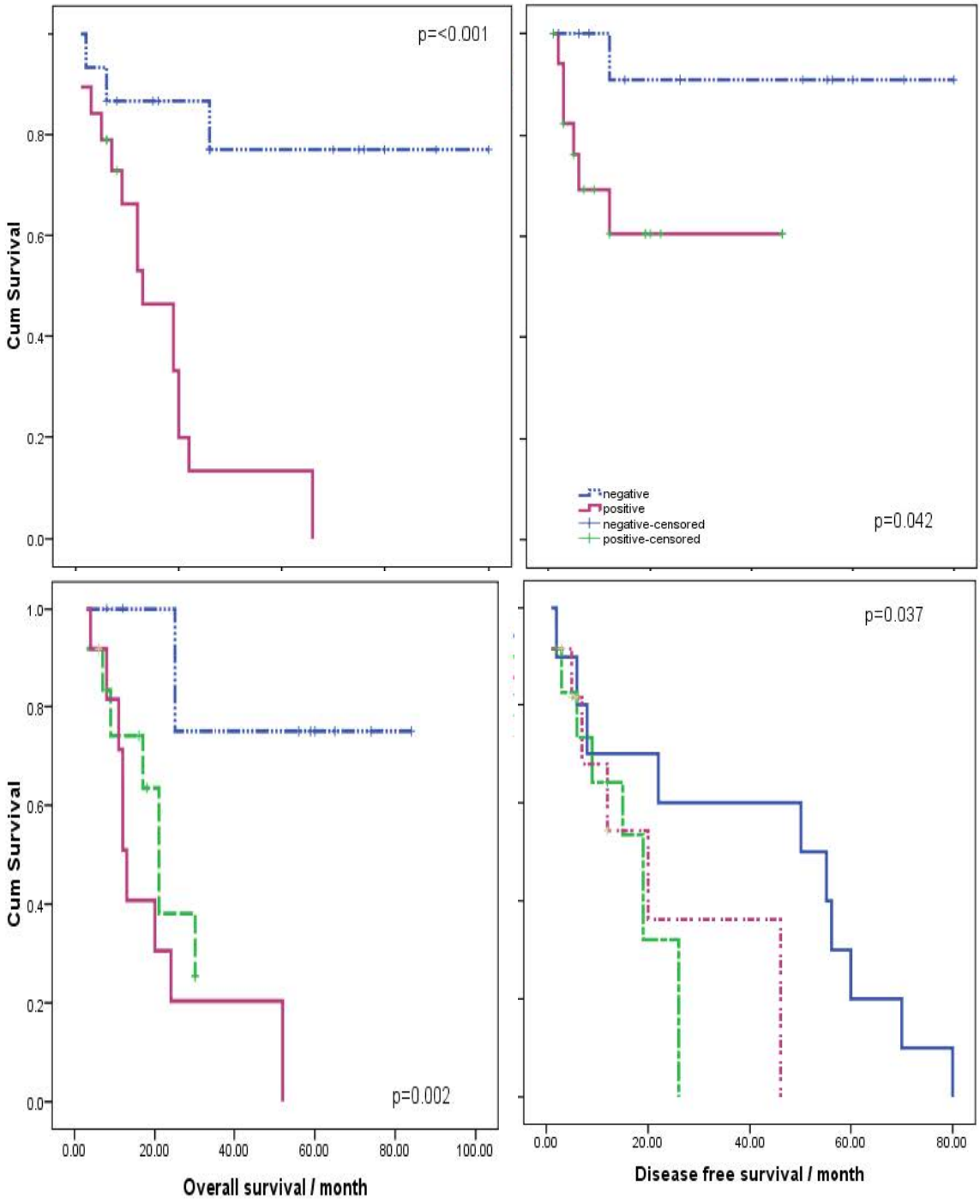

Figure 2. (a) Kaplan-Meier survival curve for overall survival among patients with Wilms tumor stratified by the positive or negative expression of p53. (b) Kaplan-Meier survival curve for disease-free survival among patients with Wilms tumor stratified by the positive or negative expression of p53. (c) Kaplan-Meier survival curve for overall survival among patients with Wilms tumor stratified by low, moderate and high expression of ki67. (d) Kaplan-Meier survival curve for disease-free survival among patients with Wilms tumor stratified by low, moderate and high expression of ki67.

This is because p53 protein in a normal cell is the gate keeper of the cell cycle that prevent mutation or induce apoptosis via caspase activation (Rivlin et al., 2011). Also, a positive relation was observed between positive p53 expression and higher tumor stage was observed by previous studies (Lahoti et al., 1996; Govender et al., 1998; Skotnicka-Klonowicz et al., 2002) and not observed by others (Cheah et al., 1996; D'Angelo et al., 2003; Krishna et al., 2016). This difference may be due to the different sample sizes among different studies. Also, there was a positive correlation between p53 positivity and shorter disease-free survival and overall survival. So, by proving this association; target therapy may be beneficial to improve the survival of Wilms tumor cases. Like relation with tumor stage, there was a discrepancy about the relation between p53 expression and patient survival.

Most of the previous studies regarding the p53 immunoreactivity in WT are shown in (table 4). This variation in different studies could be attributed to a) different antibodies and staining techniques used, b) different thresholds for calling a sample p53 positive, c) different degrees of anaplasia in the samples or selection of areas with known anaplasia to be stained. It is also positive that p53 is upregulated in areas of tumor hypoxia or necrosis.

Over the past decade, the importance of Ki-67 in the prognosis of breast cancer has been widely studied and established, however fewer studies and literature are available in the context of renal cancer which has an increasing incidence (Beckwith, 1983). Regarding ki67 immunoreactivity in the current study, 29.4% shows a mild reaction, 35.3% shows moderate reaction and the same percentage shows a severe reaction.

Several studies investigated the relationship between the immunohistochemical expression of ki67 and clinicopathological parameters of Wilms tumor cases. Several studies reported that ki67 positivity is associated with higher tumor stage: like the results of the present study (Juszkiewicz, 1997; Das et al., 2012; Krishna et al., 2016). Moreover, our study proved a positive relation between increased ki67 positivity and shorter disease-free survival and overall survival which is reported also by other studies (Ghanem et al., 2005; Diniz et al., 2011). However, Jurić et al. reported that Ki-67 is a relevant marker for assessing the proliferative activity and tumor cell dynamics of nephroblastoma, but it may not be a good clinical prognostic marker (Jurić et al., 2010). By applying multivariate Cox regression analysis to test the prognostic yield of our detected prognostic factors; higher tumor stage (p=0.025), p53 positivity (p=0.001) and high ki67 positivity (p=0.007) were independent predictors of mortality (table 3). The tumor stage was reported to be an independent prognostic factor by many studies (Sonn et al., 2008; Davidoff, 2009; Tang et al., 2019).

CONCLUSION

Ki67 proliferative marker and p53 mutated protein has an important role in Wilms tumor prognosis.

Study limitations: small sample size is one of the present study limitations. The other limitation was the lack of genetic testing for p53 gene mutation due to financial limitation.

Future prospective or recommendations include doing more studies regarding this point of research on a larger sample size including multiple health care centers in Egypt. The aim will be to study the possible role of mutant p53 protein in the progression of Wilms tumor. If this role has been proved, targeting this mutant protein may improve the prognosis of Wilms tumor cases that are resistant to conventional chemotherapy. The same is for ki67.

Abbreviations

DFS	Disease free survival
DNA	deoxyribonucleic acid
H&E	Hematoxylin & Eosin
IHC	Immunohistochemical
OS	Overall survival
Tp53	p53 tumor suppressor gene
WT	Wilms tumor

CONFLICT OF INTEREST

All authors declared no conflicts of interest.

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