Research Article / Araştırma Makalesi

Cervical Cytology Findings in Renal Transplant Patients and Comparison of These Findings with Normal Population

Böbrek Nakli Yapılmış Hastalarda Servikal Sitoloji Bulguları ve Bu Bulguların Normal Bireylerle Karşılaştırılması

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Abstract: Long-term immunosuppressive therapy increases the likelihood of renal transplant patients developing cervical cancer. We aimed to analyze the results of cervical cytology in renal transplant patients, compare these findings to those of the normal population, and determine the risk factors linked to the development of squamous intraepithelial lesions. Our analysis involved a retrospective study of hospital records from January 2000 to April 2018, encompassing 140 female renal transplant patients. A control group of 280 women with normal health status was selected and matched based on age and the cervical cytology procedure. The cervical cytology findings of the patients were re-evaluated using the Bethesda 2014 criteria. Of the total of 420 patients, 37 patients had epithelial cell abnormalities; including 32 (86.5%) patients in the renal transplant group and 5 (13.5%) control group ($p \leq 0.001$). Sixty-two patients had infections; including 44 (71%) patients in the renal transplant group and 18 (29%) control group ($p \leq 0.001$). We revealed that the development of squamous intraepithelial lesions was associated with factors such as having an immunologic disease as the primary renal disease, undergoing re-transplantation, and the presence of acute rejection ($p \leq 0.05$). Cervical cytology screening plays a crucial role in detecting preinvasive lesions. The incidence of epithelial cell abnormalities is significantly higher in renal transplant patients compared to the normal population. Regular cervical cytology follow-up is vital for the early detection and prevention of cervical cancer progression in transplant recipients.
Keywords: Cervical cytology, Immunosuppression, Pap smear, Renal transplantation

Özet: Uzun süreli immünsüpresif tedavi, böbrek nakli yapılan hastalarda servikal kanser gelişme olasılığını artırır. Bu çalışmada, böbrek nakli yapılan hastalarda servikal sitoloji sonuçlarını incelemeyi, bu bulguları normal popülasyonla karşılaştırmayı ve skuamöz intraepitelyal lezyon gelişimiyle ilişkili risk faktörlerini belirlemeyi amaçladık. Ocak 2000 ile Nisan 2018 tarihleri arasında, 140 kadın böbrek nakli alıcısının tıbbi kayıtlarını retrospektif olarak analiz edildi. Yaş ve servikal sitoloji prosedürü temel alınarak, normal sağlık durumuna sahip 280 kadından oluşan bir kontrol grubu seçildi ve eşleştirildi. Hastaların Pap smear bulguları, Bethesda 2014 kriterlerine göre yeniden değerlendirildi. Toplam 420 hastanın 37'sinde epitel hücre anormallikleri saptandı; bunların 32'si (%86,5) böbrek nakli grubunda ve 5'i (%13,5) kontrol grubundaydı ($p \leq 0,001$). Enfeksiyonların görüldüğü 62 hasta vardı; bunların 44'ü (%71) böbrek nakli grubunda ve 18'i (%29) kontrol grubundaydı ($p \leq 0,001$). Squamous intraepitelyal lezyon gelişiminin, hastanın primer böbrek hastalığının immünolojik bir hastalık olması, yeniden nakil olma ve akut rejeksiyon varlığı gibi faktörlerle ilişkili olduğunu saptandı ($p \leq 0,05$). Pap smear taraması, servikal preinvaziv lezyonların tespitinde önemli bir rol oynamaktadır. Epitel hücre anormalliklerinin görülme sıklığı, böbrek nakli alıcılarında normal popülasyona kıyasla belirgin şekilde daha yüksektir. Nakil alıcılarında servikal kanser ilerlemesinin erken tespiti ve önlenmesi için düzenli servikal sitoloji takibi hayati önem taşımaktadır.
Anahtar Kelimeler: Böbrek nakli, İmmunsupresyon, Pap smear, Servikal sitoloji,

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1. Introduction

Cervical cancer is one of the most common cancers in women worldwide (1). The precursor to cervical cancer is a high-grade squamous intraepithelial lesion (HSIL), typically associated with persistent human papillomavirus (HPV) infection. The majority of HPV infections naturally regress without causing cervical dysplasia. A robust immune response plays a critical role in effectively clearing HPV infections. However, long-term immunosuppressive therapy in renal transplant recipients hinders HPV clearance, leading to the progression of precancerous and cancerous lesions. Immunosuppressive drugs not only increase the likelihood of HPV infection but also contribute to DNA damage, impaired DNA repair, and reduced immune tolerance towards neoplastic cells (2,3).

Cervical carcinoma develops through the gradual accumulation of epithelial abnormalities, and routine cervical screening enables the detection of precursor lesions of cervical cancer. The Papanicolaou (Pap) smear is the main screening method used to detect cervical preinvasive and invasive lesions (4). This screening method reduces the incidence and mortality of cervical cancer in both the normal population and renal transplant recipients.

This study aimed to assess cervical cytology findings in renal transplant patients, comparing them to the normal population using the Bethesda 2014 criteria and investigate risk factors associated with squamous intraepithelial lesions (SILs) in renal transplant patients.

2. Materials and Methods

A retrospective study was conducted on the hospital records of patients who underwent renal transplant surgery between January 2000 and April 2018 at Baskent University, Ankara Hospital. The inclusion criteria for this study mandated patients to have documented gynecologic follow-up for a minimum of one year following renal transplantation and to have undergone at least one cervical cytology sample after the transplantation. A total of 140 women who had cervical cytology performed after transplantation were included in the study. Furthermore, a control group of 280 women was selected using a propensity score matching program according to the technical procedure of cervical cytology and age to ensure comparability with the study group. The control individuals did not receive any immunosuppressive treatment. All patients were aged over 18 years. The Pap smears of all patients were re-evaluated based on the Bethesda 2014 criteria (4). Clinical and pathological data, including patient age, current and past immunosuppressive regimens, history of rejection episodes and treatments, and primary disease, were reviewed, and clinical follow-up findings were documented. The donor treatment and renal transplant surgeries followed standardized procedures. The study was conducted under the ethical guidelines outlined in the 1975 Declaration of Helsinki and approved by the Ethical Review Committee of the institute (KA23/60; Date 09.05.2023).

2.1.Statistical Analyses

Statistical analyses were performed using the Statistical Package for the Social Sciences (v.26.0; SPSS Inc., Chicago, IL, USA). Numerical variables were presented as mean ± standard deviation (SD). Analytical methods (Kolmogorov-Simirnov test) were used to determine whether the variables were normally distributed. Since normal distribution could not be obtained, non-parametric tests were performed. The Pearson chi-square or Fisher's exact test was used to compare the Qualitative variables and represented by numbers and percentages. The nonparametric Mann–Whitney test was used for the comparison of numerical data. The effects of clinicopathologic variables on the presence of epithelial cell abnormalities were assessed by univariate and multivariate

Cox proportional hazards regression models. For all tests, $p \leq 0.05$ was considered statistically significant.

3. Results

3.1.The Abnormal Cervical Cytology Findings in Renal Transplant Patients and the Normal Population, and Their Comparison

Figures 1 and 2 demonstrated representative microphotographs showing the epithelial abnormalities and the infectious agents.

Epithelial cell abnormalities were evaluated by analyzing Pap smear results from 140 women who underwent renal transplantation and 280 women in the control group. The mean age of patients in the renal transplant and control group was 40.37 ± 10.39 years and 40.36 ± 10.36 years (range, 20-66 years), respectively. As indicated in Table 1, out of the total 420 patients, 37 (8.8%) had epithelial cell abnormalities, with 32 (86.5%) in the renal transplant group and 5 (13.5%) in the control group ($p \leq 0.001$).

Among the patient population, seven (1.7%) individuals were diagnosed with atypical squamous cells of undetermined significance (ASC-US), with 5 (71.4%) being renal transplant group and 2 (28.6%) belonging to the control group ($p = 0.031$). Low-grade squamous intraepithelial lesion (LSIL) was detected in 25 patients (6%), including 22 (88%) renal transplant group and 3 (12%) individuals from the control group. HSIL was observed in 5 patients (1.2%), all of whom were renal transplant group, while none of the individuals in the control group showed HSIL. A statistically significant difference in epithelial cell abnormalities between the two groups was found when comparing the results to the control group ($p \leq 0.05$). Notably, no glandular cell abnormalities were detected.

Out of the 37 patients with epithelial cell abnormalities, only 10 (27.0%) underwent cervical biopsy following a Pap smear. All ten patients were renal transplant group. Among these patients, the preoperative diagnoses were ASC-US in 2 patients, LSIL in 3 patients, and HSIL in 5 patients. Among the two patients with ASC-US cytology, the histopathological findings revealed squamous metaplasia. Among the three patients with LSIL cytology, the histopathological diagnoses were LSIL in 2 patients and HSIL in 1 patient. Among the five patients with HSIL cytology, the histopathological diagnoses were HSIL in 4 patients and squamous cell carcinoma in 1 patient.

Table 1 demonstrates that out of the 420 patients, 62 (14.8%) had infections. Of these, 44 (71%) were in the renal transplant group, and 18 (29%) were in the control group ($p \leq 0.001$). Among the patient population, *Candida* infection was observed in 35 individuals (8.3%), with 28 (80%) of them belonging to the renal transplant group and 7 (20%) to the control group. *Trichomonas vaginalis* infection was detected in 3 (0.7%) patients from the renal transplant group. The incidence of *Candida* and *Trichomonas vaginalis* infection was significantly higher in the renal transplant group compared to the control group ($p \leq 0.001$, $p = 0.014$, respectively). *Herpes simplex* infection was found in only one (0.2%) patient in the renal transplant group, while *Actinomyces* infection was present in 2 individuals (0.5%) from the control group. There were no statistically significant differences between the renal transplant and control group regarding *Actinomyces* and *Herpes simplex* infections ($p = 0.316$, $p = 0.333$, respectively). Twenty-eight (6.7%) individuals had a shift in the vaginal flora suggestive of bacterial vaginosis. Of these, 12 (57.1%) patients were in the renal transplant group, and 9 (42.9%) individuals were in the control group. This difference was statistically significant ($p = 0.018$). *Chlamydia* microorganisms, reactive

changes due to intrauterine devices, and reactive changes due to radiation were not detected in either group.

Inflammatory reactive changes were observed in 50 (11.9%) individuals, including 27 (54.0%) in the renal transplant group and 23 (46.0%) in the control group. Atrophy was detected in 49 patients (11.7%), including 27 (55.1%) in the renal transplant group and 22 (44.9%) in the control group. These differences were statistically significant (p = 0.001, p = 0.001, respectively).

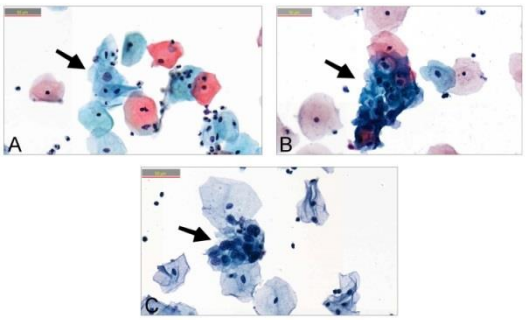

Figure 1. Representative microphotographs showing the epithelial abnormalities. A) Atypical Squamous Cells of Undetermined Significance (ASC-US). Nuclear enlargement and hyperchromasia are noted in a superficial cell. B) Low-Grade Squamous Intraepithelial Lesion (LSIL). Binucleation and koilocytosis are noted. C) High-Grade Squamous Intraepithelial Lesion (HSIL). Increased nuclear/cytoplasmic ratio and hyperchromasia are noted (Pap stain, original magnification ×400).

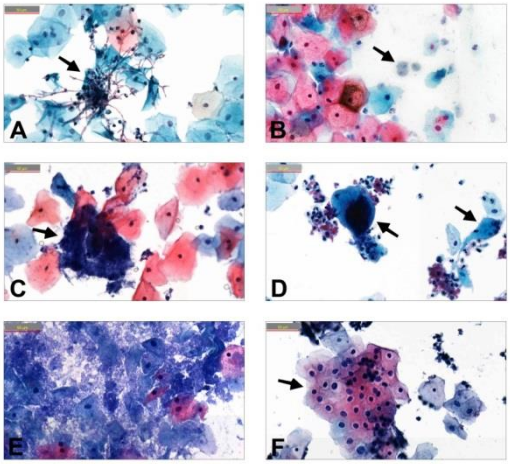

Figure 2. Representative microphotographs showing the infectious agents. A) *Candida albicans*. Fungal organisms with pseudohyphae are noted between squamous cells. B) *Trichomonas vaginalis*. Pear-shaped organisms with eccentrically located nuclei and eosinophilic cytoplasmic granules are shown. C) *Actinomyces.* Tangled clumps of filamentous organisms, often with acute angle branching, are shown as "cotton ball" clusters. D) *Herpes simplex.* A typical multinucleated cell showing the "ground-glass" appearance of the nuclei. E) Bacterial vaginosis. Shift in flora suggestive of bacterial vaginosis. Individual squamous cells are covered by a layer of coccobacilli noted as "clue cells". F) Reactive changes. Reactive squamous epithelial cells display small perinuclear halos, mild nuclear enlargement, and prominent nucleoli without any significant chromatin abnormalities (Pap stain, original magnification ×400)

Table 1. Cervical cytology findings of renal transplantation patients and normal population, and their comparison

	N (%)	Renal Transplantation Group	Control Group	*p*
	420 (100%)	140 (33.3%)	280 (66.7%)	
Age (mean ± SD) (years)		40.4 ± 10.38	40.4 ± 10.38	
Technical procedure				
Conventional cytology	272 (64.8)	77 (28.3%)	195 (71.7%)	
Liquid-based cytology	148 (35.2%)	63 (42.6%)	85 (57.4%)	
Epithelial cell abnormalities				
Absent	383 (91.2%)	108 (28.2%)	275 (71.8%)	≤ 0.001*
Present	37 (8.8%)	32 (86.5%)	5 (13.5%)	
ASC-US	7 (1.7%)	5 (71.4%)	2 (28.6%)	0.031*
LSIL	25 (6%)	22 (88.0%)	3 (12.0%)	≤ 0.001*
HSIL	5 (1.2%)	5 (100%)	0 (0%)	0.001*
Infections				
Absent	358 (85.2%)	96 (26.8%)	262 (73.2%)	≤ 0.001*
Present	62 (14.8%)	44 (71%)	18 (29%)	
Organisms				
Candida	35 (8.3%)	28 (80%)	7 (20%)	≤ 0.001*
Trichomonas Vaginalis	3 (0.7%)	3 (100%)	0 (0%)	0.014*
Actinomyces	2 (0.5%)	0 (0%)	2 (100%)	0.316
Herpes Simplex	1 (0.2%)	1 (100%)	0 (0%)	0.333
Bacterial vaginosis	22 (5.2%)	12 (57.1%)	9 (42.9%)	0.018*
Nonneoplastic findings				
Reactive changes	50 (11.9%)	27 (54.0%)	23 (46.0%)	0.001*
Atrophy	49 (11.7%)	27 (55.1%)	22 (44.9%)	0.001*

ASC-US indicates Atypical Squamous Cells of Undetermined Significance; LSIL, Low-Grade Squamous Intraepithelial Lesion; HSIL, High-Grade Squamous Intraepithelial Lesion
**Statistically significant*

3.2.The Association Between Clinicopathologic Features and Squamous Intraepithelial Lesion Development in Renal Transplant Patients

The ASC-US lesion was excluded and included in the absent group for the SIL in this section. Table 2 shows that out of 140 renal transplant patients, 22 (15.7%) were diagnosed with LSIL, and 5 (3.6%) had HSIL. The average number of cervical cytology tests performed was 4.5 ± 3.6 (range; 2-16). The mean age for patients with SIL and without SIL was 35.5 ± 8.8 years (range, 26-51) and 36.8 ± 9.0 years (range, 18-59), respectively. The two groups had no significant differences (p = 0.107). The average interval between renal transplantation and the Pap smear test for patients with SIL and without SIL was 92.98 ± 74.88 months (range, 12-301 months) and 92.66 ± 64.42 months (range, 12-329 months), respectively. The two groups had no significant differences (p = 0.814).

Out of the total 140 renal transplant patients, four patients (2.9%) underwent re-transplantation at a mean interval of 175.50 ± 70.14 months (range: 110-242 months) after their initial transplantation. Among these four re-transplanted patients, all of them (100%) were diagnosed with SIL. Additionally, SIL was found in 23 (16.9%) out of the 136 patients who had undergone transplantation only once. There was a significant association between SIL and re-transplantation (p = 0.001).

Among the patients, 35 (25%) had immunologic diseases such as glomerulonephritis, familial Mediterranean fever, and systemic lupus erythematosus, while 105 (75%) had non-immunologic diseases including vesicoureteral reflux, nephrolithiasis, pre-eclampsia, hypertension, and diabetes mellitus. Out of the 27 patients with SIL, 18 (66.7%) had immunologic diseases, while 9 (33.3%) had non-immunologic diseases. The incidence of SIL was significantly higher in patients with a

primary immunologic disease compared to those without ($p \leq 0.001$).

The majority of patients (91.4%) were treated with a regimen consisting of calcineurin inhibitors, specifically tacrolimus or cyclosporine-A, in combination with steroids. A smaller proportion of patients (8.6%) received sirolimus or mycophenolate mofetil in combination with steroids. Among the 27 patients with SIL, 25 (92.6%) were treated with a calcineurin inhibitors regimen, while 2 (7.4%) received other medications. However, there was no significant association between SIL and the immunosuppressive regimen used ($p = 0.810$). Among the patients with SIL, 14 (51.9%) had experienced acute rejection, while 13 (48.1%) had not. The incidence of SIL was higher in patients with acute rejection compared to those without ($p = 0.015$).

Out of the 140 patients, 28 (20%) had reached menopause. Among the 27 patients with SIL, 4 (14.8%) were menopausal, and 23 (85.2%) were not. However, there was no significant association between SIL and menopause status ($p = 0.453$).

Regarding cytologic findings, out of the 140 renal transplant patients, 44 (31.4%) had infections, including 28 (63.6%) with *Candida* infection, 3 (6.8%) with *Trichomonas vaginalis* infection, 12 (27.3%) with bacterial vaginosis, and 1 (2.3%) with *Herpes simplex* infection. No patients had *Actinomyces* infections. There was no significant association found between SIL and infections ($p = 0.493$).

Atrophy was detected in 27 (19.3%) patients. Among the 27 patients with SIL, 26 (96.3%) did not have atrophy, while only 1 (3.7%) had atrophy. The incidence of SIL was higher in patients without atrophy compared to those with atrophy ($p = 0.022$).

Table 2. Association between clinicopathologic features and squamous intraepithelial lesion in renal transplantation patients

Variables		**Squamous intraepithelial lesion**		*p*
	N (%)	**Present**	**Absent**	
	140 (100%)	27 (19.3%)	113 (80.7 %)	
Age (Mean ± SD) (years)		35.5 ± 8.8	36.8 ± 9.0	0.107
The mean interval time after transplantation (Mean ± SD) (months)		92.98 ± 74.88	92.66 ± 64.42	0.814
Re-transplantation				
Absent	136 (97.1%)	23 (85.2%)	113 (100%)	0.001*
Present	4 (2.9%)	4 (14.8%)	0 (0%)	
The primary disease				
Immunologic diseases	35 (25.0%)	18 (66.7%)	17 (15%)	≤ 0.001*
Non-immunologic diseases	105 (75.0%)	9 (33.3%)	96 (85%)	
Immunosuppressive Regimen				
Calcineurin inhibitors	128 (91.4%)	25 (92.6%)	103 (91.2%)	0.810
Others	12 (8.6%)	2 (7.4%)	10 (8.8%)	
Acute rejection				
Absent	95 (67.9%)	13 (48.1%)	82 (72.6%)	0.015*
Present	45 (32.1%)	14 (51.9%)	31 (27.4%)	
Menopause status				
Absent	112 (80.0%)	23 (85.2%)	89 (78.8%)	0.453
Present	28 (20.0%)	4 (14.8%)	24 (21.2%)	
Cytologic Findings				
Infection				
Absent	96 (68.6%)	20 (74.1%)	76 (67.3%)	0.493

Present	44 (31.4%)	7 (25.9%)	37 (32.7%)	
Atrophy				
Absent	113 (80.7%)	26 (96.3%)	87 (77.0%)	0.022*
Present	27 (19.3%)	1 (3.7%)	26 (23.0%)	

LSIL indicates Low-Grade Squamous Intraepithelial Lesion; HSIL, High-Grade Squamous Intraepithelial Lesion
**Statistically significant*

3.3.The Assessment of the Risk Factors for Squamous Intraepithelial Lesion Development in Renal Transplant Patients

The multivariate Cox regression analyses revealed that primary renal disease and cervical atrophy were significantly associated with the presence of SIL in the cervical cytology ($p \leq 0.001$, $p = 0.036$, respectively). The univariate Cox regression analysis showed that renal disease was significantly associated with the presence of SIL in the cervical cytology ($p \leq 0.001$) (Table 3).

Table 3. Prognostic significance of clinicopathologic features on the presence of squamous intraepithelial lesion (multivariate and univariate Cox stepwise regression analysis)

Multivariate Cox Regression Analysis				**Univariate Cox Regression Analysis**			
Variable	Hazard ratio	95% Cl	*p*-value	Variable	Hazard ratio	95% Cl	*p*-value
Re-transplantation (absent vs. present)	0.516	0.135-1.973	0.334	Re-transplantation (absent vs. present)	0.501	0.161-1.566	0.235
Primary renal disease (immunologic vs.non-immunologic)	8.744	3.671-20.883	≤0.001*	Primary renal disease (immunologic vs. non-immunologic)	7.031	3.050-16.206	≤0.001*
Immunosuppressive regimen (calcineurin inhibitors vs. others)	0.519	0.108-2.484	0.412	Immunosuppressive regimen (calcineurin inhibitors vs. others)	0.812	0.186-3.543	0.781
Acute rejection (absent vs. present)	0.971	0.371-2.543	0.952	Acute rejection (absent vs. present)	0.580	0.270-1.243	0.161
Infection (absent vs. present)	1.394	0.499-3.893	0.526	Infection (absent vs. present)	1.845	0.773-4.404	0.168
Atrophy (absent vs. present)	8.665	1.154-65.076	0.036*	Atrophy (absent vs. present)	6.645	0.891-49.563	0.065

Cl indicates Confidence interval.
**Statistically significant*

4.Discussion

Organ transplant recipients have a threefold increase in the incidence of in situ cancer compared to the normal population. Studies have shown that organ transplant recipients have a 1% incidence of invasive cervical cancer and a 3.3% incidence of cervical cancer in situ (5). Routine cervical screening plays a crucial role in detecting precursor lesions of cervical cancer. The Pap test is a convenient, affordable, and highly accurate screening method with highsensitivity and specificity. This cytologic screening test is recommended for all women, including those who have undergone transplantation. Existing guidelines for cervical cancer screening primarily emphasize the age range of 21 to 65 for women (6), but they often lack specific recommendations for immunocompromised women, including transplant patients. This gap in guidelines highlights the need for further research and consensus on appropriate screening protocols tailored to the unique needs of immunocompromised individuals.

The pathogenesis of cervical carcinoma involves the progressive accumulation of epithelial abnormalities. Consistent with previous research, the current study revealed a statistically significant increase in the risk of

epithelial cell abnormalities among renal transplant patients compared to the normal population. These findings are consistent with a study conducted by Paternoster et al., which examined 151 transplant patients and found a higher incidence of HSIL and LSIL among this population when compared to the normal population (7). Similarly, Origoni et al. conducted a study that also showed significant differences between the groups, particularly in LSIL cytology (8).

Furthermore, our study presented a case of a 47-year-old female patient diagnosed with cervical squamous cell carcinoma after HSIL cytology four months following renal transplantation. This case highlights the importance of regular cervical cancer screening and prompt follow-up in transplant recipients to detect and address any abnormal cytological findings or potential malignancies at an early stage.

Immunocompromised renal transplant recipients undergoing long-term immunosuppressive therapy face a significant risk for the presence of precancerous lesions and the development of cervical cancer (2,3,9,10). The use of immunosuppressive medications in these individuals can weaken the immune system's ability to detect and eliminate abnormal cervical cells, thereby increasing their susceptibility to HPV infection and the progression of cervical abnormalities (2,3,8). The use of calcineurin inhibitors, such as cyclosporine and tacrolimus, in long-term immunosuppressive therapy for renal transplant patients has been associated with an increased risk of carcinogenesis. This may be attributed to their potential to induce the production of cytokines that regulate factors, which play a role in cell growth and differentiation. Furthermore, these medications have been implicated in promoting metastasis, the spread of cancer cells, and angiogenesis that supports tumor growth. Consequently, calcineurin inhibitors have the potential to contribute to the development of precancerous lesions and cervical cancer in renal transplant patients. In the literature, the relationship between calcineurin inhibitors and skin cancer has been shown in renal transplant patients (11,12). We did not indicate this relationship in our series. The current study revealed that the presence of re-transplantation and acute rejection were associated with the presence of SILs in the Pap test. Besides that, most of the patients with SIL were found to have the immunologic disease as primary kidney disease. In both the multivariate and univariate Cox regression analyses, we identified that the immunologic disease as the primary renal disease was an independent risk factor for the development of SIL in renal transplantation patients. Indeed, this result is not surprising, as patients with an immunologic disease often receive immunosuppressive therapy even before undergoing transplantation.

Renal organ transplant patients frequently experience infections with HPV, which can increase the risk of developing cervical cancer (13). High-risk oncogenic types of HPV, such as HPV-16, are more commonly found in HSIL compared to LSIL, while non-oncogenic HPV types are often observed in LSIL cases. The studies have demonstrated an accelerated progression from ASC-US to LSIL or HSIL and from LSIL to HSIL or carcinoma in women infected with oncogenic types of HPV (14). Furthermore, the research conducted by Moscicki et al. indicated that the rate of regression in LSIL was solely associated with the HPV status at the current visit, while no significant association was found between LSIL regression and HPV status at baseline in their univariate analysis (15). In the study conducted by Paternoster et al. involving HPV testing, they observed a significant association between high-risk HPV infections and CIN lesions. However, no significant association was found between low-risk HPV infections and CIN lesions (7). The HPV vaccine prior to transplantation and conducting regular HPV testing are highly recommended for individuals with renal transplantation. They provide a crucial layer of protection against HPV infection, which can lead to cervical cancer.

Immunosuppression, as a notable consequence of immunosuppressive therapy, weakens the immune system and consequently increases susceptibility to

infections. The compromised immune response makes individuals more prone to acquiring various infections in the cervical region due to the decreased ability to fight off pathogens effectively. Consistent with the findings from previous study conducted in 2015 among solid organ transplant patients (kidney and liver) (16), the current study also demonstrated a higher incidence of cervical infections in renal transplant patients compared to the normal population. We revealed a significantly higher incidence of *Candida* and *Trichomonas vaginalis* infections among renal transplant patients compared to the normal population. However, our study did not reveal any significant differences between the two groups in terms of *Actinomyces* and *Herpes simplex virus* infections.

Our Pap smear cytology findings showed a change in vaginal flora that indicates bacterial vaginosis. Our study revealed notable disparities in bacterial vaginosis occurrence between the two groups. Long-term use of immunosuppressive drugs disrupts the estrous cycle and leads to decreased mucus production in the squamous epithelium, thinning, and eventually atrophy of the cervical epithelium. We observed significant differences in atrophy between the two groups, likely associated with immunosuppression due to transplantation.

Our study has several limitations. Firstly, we were unable to determine the HPV status of the transplanted women. Secondly, we did not collect data on factors such as oral contraceptive use, smoking status, and specific dosing of immunosuppressive therapy. However, we ensured that the transplanted women and control individuals were matched for the technical procedure of cervical cytology and age. The control individuals did not receive any immunosuppressive treatment.

In conclusion, our study highlights the higher susceptibility of renal transplant patients to develop cervical precancerous lesions and cervical cancer as a result of long-term immunosuppressive treatment. We observed a significant increase in the incidence of LSIL and HSIL compared to the normal population. Consequently, we recommend that Pap test screening and HPV vaccination be conducted prior to renal transplant procedures. Moreover, there is a clear necessity for a distinct cervical cancer screening program tailored specifically for immunocompromised women. Regular Pap tests and HPV tests should be performed at appropriate intervals to detect and prevent the development of precancerous lesions and cervical cancer.

REFERENCES

1. Sung H, Ferlay J, Siegel RL, et al. Global Cancer Statistics 2020: Globocan Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. CA Cancer J Clin. 2021;71(3):209-49.
2. Park ST, Song MJ, Park JS, et al. Incidence and clinicopathologic behavior of uterine cervical carcinoma in renal transplant recipients. World J Surg Oncol. 2011;9:72.
3. Courtney AE, Leonard N, O'Neill CJ, et al. The uptake of cervical cancer screening by renal transplant recipients. Nephrol Dial Transplant. 2009;24(2):647-52.
4. Wilbur DC, Nayar R. Bethesda 2014: improving on a paradigm shift. Cytopathology. 2015;26(6):339-42.
5. Madeleine MM, Finch JL, Lynch CF, et al. HPV-related cancers after solid organ transplantation in the United States. Am J Transplant. 2013;13(12):3202-9.
6. Fontham ETH, Wolf AMD, Church TR, et al. Cervical cancer screening for individuals at average risk: 2020 guideline update from the American Cancer Society. CA Cancer J Clin. 2020;70(5):321-46.
7. Paternoster DM, Cester M, Resente C, et al. Human papilloma virus infection and cervical intraepithelial neoplasia in transplanted patients. Transplant Proc. 2008;40(6):1877-80.
8. Origoni M, Stefani C, Dell'Antonio G, et al. Cervical human papillomavirus in transplanted Italian women: a long-term prospective follow-up study. J Clin Virol. 2011; 51(4):250-4.
9. Marschalek J, Helmy S, Schmidt A, et al. Prevalence of genital dysplasia after kidney transplantation--a retrospective, non-interventional study from two centers. Acta Obstet Gynecol Scand. 2015;94(8):891-7.

10. Bilgi A, Gökulu ŞG, İlgen O, et al. Cervical dysplasia after renal transplantation: A retrospective cohort study. Turk J Obstet Gynecol. 2021;18(1), 7–14.
11. Murray SL, Daly FE, O'Kelly P, et al. The impact of switching to mTOR inhibitor-based immunosuppression on long-term non-melanoma skin cancer incidence and renal function in kidney and liver transplant recipients. Ren Fail. 2020;42(1):607-12.
12. Smith A, Niu W, Desai A. The Effect of Conversion from a Calcineurin Inhibitor to Sirolimus on Skin Cancer Reduction in Post-renal Transplantation Patients. Cureus. 2017;9(8):e1564.
13. Chin-Hong PV. Human Papillomavirus in Kidney Transplant Recipients. Semin Nephrol. 2016;36(5):397-404.
14. Schlecht NF, Platt RW, Duarte-Franco E, et al. Human papillomavirus infection and time to progression and regression of cervical intraepithelial neoplasia. J Natl Cancer Inst. 2003;95(17): 1336-43.
15. Moscicki AB, Shiboski S, Hills NK, et al. Regression of low-grade squamous intra-epithelial lesions in young women. Lancet. 2004; 364(9446):1678-83.
16. Ok Atılgan A, Tepeoğlu M, Haberal AN, et al. Papanicolaou smear findings in solid-organ transplant recipients compared with normal subjects according to the Bethesda 2001 system. Exp Clin Transplant. 2015;13 Suppl 1:219-22.

Ethics

Ethics Committee Approval: The study was approved by Baskent University Noninterventional Clinical Research Ethical Committee (Decision no: KA23/60, Date: 09.05.2023).
Informed Consent: The authors declared that it was not considered necessary to get consent from the patients because the study was a retrospective data analysis.
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