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DETECTION OF HUMAN PAPILLOMAVIRUS GENOTYPES IN CANCER CERVIX PATIENTS: DAMIETTA GOVERNORATE, EGYPT

By

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ABSTRACT

Background: Human papillomavirus (HPV) infections remain a significant problem worldwide particularly in underdeveloped countries. To develop cervical cancer, it is necessary to have a persistent infection with a high-risk or oncogenic HPV.

Objectives: This study aimed to detect the HPV and its possible genotypes in cervical samples from some Egyptian female patients, histopathologically diagnosed to have cancer cervix; to guide the introduction of prophylactic vaccines.

Patients and methods: Fifty women were recruited. Pap smears were taken and examined, followed by colposcopic examinations, pathologic examination, and HPV detection in cervical samples by polymerase chain reaction (PCR). Then HPV genotyping of positive samples was done by restriction fragment length polymorphism (RFLP).

Results: HPV-DNA was detected in 40 % of patients. HPV 16 was detected in 25 %, HPV 31 in 20 %, HPV 11in 15 %, HPV 6 in 10 %, and HPV 18 in 5% of cases. About 25% (5/20) cervical cancer specimens exhibited multiple infections by HPV genotypes 16and 18, which showed a higher risk for development of cancer cervix. Correlations of age, age-specific prevalence, residence, multiple marriages, parity, contraceptives, and its duration, diabetes mellitus were statistically significant .while correlations of occupation, level of education, age at marriage, duration of marriage, recurrent and chronic infections, smoking, and family history of cancers were insignificant.

Conclusions: The prevalence of HPV in cancer cervix was 40%.So, the association between HPV and the development of cancer cervix in the studied Egyptian women cannot be fully established. HPV infection was mostly (75%) in the form of single infections with HPV16, 18, 31, 11, and 6 genotypes. However, HPV infections in multiple forms, 16+18 genotypes were in 25% of positive cases.

Keywords: Human papillomavirus genotypes, cancer cervix, polymerase chain reaction.

INTRODUCTION

Human papillomavirus (HPV) represents the commonest sexually transmitted infections. Over 5 up to 80% of sexually active women are infected at some point in their lives and 10-20% develops persistent infection *(Einstein et al., 2009)*.

Cancer cervix represents the fourth most commonly diagnosed cancer and the fourth leading cause of cancer death in women worldwide. Its worldwide burden is massive, with over 570,000 new cases of cervical cancer diagnosed each year, and 311,000 deaths recorded *(Bray et al., 2018)*.

Egypt has a population of 30.55 million women ages 15 years and older who are at risk of developing cervical cancer. Recent estimates indicate that every year 969 women are diagnosed with cervical cancer and 631 die from the disease. Cervical cancer ranks as the 14th most frequent cancer among women in Egypt and the 11th most frequent cancer among women between 15 and 44 years of age. Data is not yet available on the HPV burden in the general population of Egypt. Although, in Northern Africa, the region Egypt belongs to, about 3.0% of ladies in the general population are assessed to have cervical HPV-16/18 infection at a given time, and 78.9% of invasive cervical cancers are attributed to HPVs 16 or 18 *(WHO/ICO, 2019)*.

Nowadays, the extent of cervical cancer and HPV infection can be reduced, and control strategies rely on HPV vaccination and early detection of benign or precancerous lesions *(Nyasenu et al., 2019)*. More than 200 types of human papillomavirus (HPV) have been recognized based on DNA sequence. Definite typing of HPV is relying on the site of epithelium infected and tissue tropism types are often classified as "cutaneous" or "mucosal" types *(Brianti et al., 2017)*.

Types of HPV are divided into major categories depending on their level of association with cervical intraepithelial neoplasia (CIN) and invasive squamous cell carcinoma. The "high-risk (oncogenic)" types are mainly 16 and 18, but also 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68, 73, and 82.HPV types 26, 53, and 66 have been considered as 'probably high-risk', and HPV types 6, 11, 40, 42, 43, 44, 54, 61, 70, 72, 81, and CP6108 are combined as the 'low-risk category *(Ehteram et al., 2019)*.

Infections of HPV are represented as subclinical, latent, or persistent infection of high-risk genotypes, chronic cervicitis with abnormal results of cytological examination, CIN, and cervical cancer. Infection with HPV is supposed by the presence of clinical lesions and via the results of cytology, histology, and colposcopy, all of which are subjective and often inaccurate *(Liao et al., 2019)*.

Besides, serology is unreliable and unable to distinguish past from current infection. Molecular diagnostic tests for HPV can augment screening for cervical cancer when used in conjunction with the Pap smear *(Donken et al., 2018)*. Colposcopy is a diagnostic method to detect CIN and cervical cancer, following an abnormal cytology so its major role is in guiding the diagnostic biopsy. HPV testing changes the colposcopic practice as HPV screening will increasingly

change the patient population referred to colposcopy *(Tidy et al., 2018)*.

There are several methods for the molecular diagnosis of HPV, varying from a conventional polymerase chain reaction (PCR) real-time PCR, hybrid capture. The PCR technique is still considered the "gold standard" for HPV diagnosis *(Coser et al., 2011)*. Use of nested PCR with degenerate primers has been described by multiple studies to be extremely sensitive tools of detecting a wide range of HPV types *(Erhart et al., 2016)*.

For the genotyping of HPV, the target products amplified by PCR are subjected to sequence analysis, RFLP analysis, and hybridization with type-specific probes, Reverse line blot assays have also been validated *(Clifford et al., 2016)*. Nested PCR plus RFLP, using MY09/11 primers with different restriction enzymes; were able to detect a broad range of HPV types *(Tsakogiannis et al., 2017)*.

In this study, we aimed to detect the HPV and its possible genotypes in cervical samples from some Egyptian female patients, histopathologically diagnosed to have cancer cervix attending the outpatients' clinics of obstetrics and gynecology department, Al-Azhar University Hospital, New Damietta faculty of medicine.

PATIENTS AND METHODS

During the period between January 2017 and August 2019, this study was conducted on 50 patients histopathologically diagnosed to have cancer cervix. These patients were referred from the outpatients' clinics of obstetrics and gynecology department, Al-Azhar University Hospital, New Damietta, for colposcopic examination of abnormal-looking cervices. And all patients provided written informed consent. The ethical research and review committee of the hospital approved the study protocol.

Inclusion criteria: All selected women married with suspicious cervix.

Exclusion criteria:

1. Pregnant woman, Postpartum within the last 6 months, Post-abortion and those with history of cervical surgery (cauterization, conization).

2. Vaginal bleeding at the time of examination.

3. History of blood clotting disorders or anticoagulant therapy.

4. Prior diagnosis of gynecological malignancy or hysterectomy .

Each patient was subjected to the following:

A. Full history taking .

B. Pap smear according to the Bethesda classification system 2015 *(Nayar and Wilbur, 2015)*.

C. Cervical swab as described by *Hernandez et al. (2018)*.

D. Colposcopy: as described by *Abdelbadiaa et al. (2016)*.

E. Colposcopic directed punch biopsy from the abnormal transformation zone of the cervix detected by acetic acid test and Schiller's iodine test was obtained. Each biopsy was kept in a10% formalin solution sent to the laboratory to be processed in paraffin blocks. From these paraffin blocks, slides were prepared for staining with hematoxylin and eosin for histopathological examination by a trained

pathologist, General Pathology Department, Faculty of Medicine (New Damietta), Al-Azhar University according to *Lagheden et al. (2016)*. Cervical swab of patients' who had proved to have cervical carcinoma by histopathological examination of colposcopic directed punch biopsies, were selected for PCR and restriction fragment length polymorphism *(Chen et al., 2015)*.

F. DNA Extraction carried out by DNA extraction kit "QiAamp DNA mini kit" (QIAGEN Inc., Valencia, CA, USA). To check the integrity of the DNA extracted from the specimens, a region of 268 base pairs of the cellular β-globin gene was amplified using primers GH20 and PC04. The PCR conditions were the same as described by *Camargo et al. (2011)*.

G. Polymerase chain reaction (PCR): Samples were analyzed through nested PCR using the degenerate consensus primers MY09and MY11, which amplify a region of 449 - 458 bp (depending on HPV type) of the highly conserved L1 gene *(Camargo et al., 2011)*.PCR reactions were carried out in a 50 ?l volume containing 25?l of Taq PCR master mix kit (Qiagen, Germany), 3 ?l extracted DNA, 2 ?l forward primer, 2 ?l reverse primer, and 18 ?l nuclease-free water. PCR reactions were performed in a thermal cycler (Biometra, Germany). Cycling condition was as follows: denaturation of DNA template at 95°C for 4minutes, followed by 35 cycles of 94°C for 30 seconds, 56°C for 30 seconds, and 72°C for 30 seconds, and a final extension step for 8 minutes *(Chen et al., 2015)*.

All PCRs were carried out under conditions that minimize sample cross-contamination. Also, a negative control and positive control were added to each run. Then, the amplified DNA was analyzed in a horizontal 2% agarose. The gel was carefully removed and was observed and photographed over the UV transilluminator. The positive specimens; showing PCR amplicon of the expected size 450 bp, were undergone genotyping by RFLP, while negative samples had to processed by double nested PCR *(Tawe et al ., 2018)*.

H. Double-nested PCR analysis: All the HPV negative samples from nested PCR were then subjected to another cycle of PCR amplification using GP5/6 primers' pair. PCR reactions were carried out in a 50 ?l volume containing 25?l of Taq PCR master mix kit (Qiagen, Germany), 3 ?l extracted DNA, 2 ?l forward primer, 2 ?l reverse primer, and 18 ?l nuclease-free water. PCR reactions were performed in a thermal cycler (Biometra, Germany). Cycling condition was as follows: denaturation of DNA template at 95°C for 4minutes, followed by 35 cycles of 94°C for 30 seconds, 40°C for 30 seconds, and 72°C for 30 seconds, and a final extension step for 8 minutes. Positive and negative controls were added. Then, the amplified DNA was analyzed in a horizontal 2% agarose. The gel was carefully removed and was viewed and photographed over the U.V. transilluminator. The positive specimens; showing PCR amplicon of the expected size 150bp, were undergone genotyping by RFLP (Figure: 4) *(Tawe et al., 2018)*.

I. Restriction Fragment Length Polymorphism: HPV-positive cases were typed by RFLP analysis. Each restriction reaction was processed separately in a final volume of 50 ?l, using 10 ?l of

product of nested and double nested PCR, 5 ?l of 10X recommended restriction buffer, 34 ?l nuclease-free water and 10 units (1?l) of the following restriction endonucleases: DdeI (Qbiogene, USA), RsaI (Fermentas, Canada), PstI (Qbiogene, USA) and HaeIII (Fermentas, Canada), according to the manufacturers' instructions. Reactions took place at 37°C for 1.5 hours. Digested products were electrophoretically separated on 3 % agarose gels, in the presence of 50bp DNA molecular weight marker (Invitrogen, Carlsbad, CA, USA) *(Tawe et al., 2018)*.

Statistical Analysis:

Data were collected, revised and entered using the statistical package SPSS version 22. The collected data were tabulated and analyzed with the suitable statistical methods using mean value ± standard deviation, t-test, z-test, and chi-square test. P-value of <0.05 was considered statistically significant.

RESULTS

This study included fifty patients with ages ranging from 30 to 63 years and a mean of 42 ± 8.3years. Nested PCR was positive in 20 (40%) cases and 30 (60%) cases were negative. After double nested PCR, there were 26/30 patients (86.7%) still negative, while4/30 patients (13.3%) were positive. Different HPV genotypes were detected in the current study.

Table (1): Different HPV genotypes detected by nested PCR plus RFLP

HPV genotype of nested PCR	SCC (19)		AD (1)	
	n	%	n	%
High risk 16 (5)	5	100	0	0
18 (1)	1	100	0	0
31 (4)	4	100	0	0
16+18 (4)	3	75	1	25
Low risk 6 (3)	3	100	0	0
11 (3)	3	100	0	0

Double nested PCR		Studied patients (n = 30)	
		n	%
HPV- DNA	Negative	26	86.7
	Positive	4	13.3
HPV Genotype	(8)	2	50
	(11)	2	50

A statistically significant relations between the results of nested PCR and the age, the age-specific prevalence of HPV and the residence of the studied women. No statistically significant differences between the results of nested PCR and occupation or educational level of the studied patients (Table 2).

Table (2): Clinical parameters of the patients and their relation to PCR detection of HPV

Parameters \ Groups		Positive (n =20)		Negative (n = 30)		P-value
Age (years)	**Mean ±SD**	**47.8±9.8**		**39.6±5.1**		**0.001 S**
Age categories	**variables**	**n**	**%**	**n**	**%**	**0.001**
	30 – 39y	**3**	**15**	**14**	**47**	
	> 39 – 49y	**9**	**45**	**16**	**53**	
	> 49 – 59y	**4**	**20**	**0**	**0**	
	> 59 - 63y	**4**	**20**	**0**	**0**	
Occupation	**Housewife**	**5**	**25**	**5**	**17**	**0.47**
	Employee	**15**	**75**	**25**	**83**	
Education level	**Illiterate**	**3**	**15**	**2**	**7**	**0.4**
	Read / write	**5**	**25**	**3**	**10**	
	Elementary	**1**	**5**	**4**	**13**	
	High school	**10**	**50**	**20**	**67**	
	University	**1**	**5**	**1**	**3**	
Residence	**Urban**	**5**	**20**	**15**	**50**	**0.077**
	Rural	**15**	**75**	**15**	**50**	

A statistically significant relations between the results of nested PCR and marital status and the number of marriages of the studied patients. No statistically significant differences between the results of nested PCR, and age of marriage or duration of marriage of the studied women. Also, there were statistically significant relations between the results of nested PCR and parity of studied patients. Moreover, a statistically significant relation between the results of nested PCR and contraceptives. A highly statistically significant occurred differences between the results of nested PCR and duration of contraception (Table 3).

Table (3): Correlation of HPV detection by nested PCR and the gynecological and reproductive history risk factors of the studied women

Parameters / Risk factors		Positive (n =20)		Negative (n = 30)		P Value
		n	%	n	%	
Marital status	Married/husband has 1 wife	15	75	26	87	0.006
	Married/husband has more than 1 wife	5	25	0	0	
	Divorced/widow	0	0	4	13	
Age at marriage	<20 years	10	50	10	33	0.339
	20 - 30 years	9	45	16	54	
	>30 years	1	5	4	13	
Duration of marriage	< 5 years	1	5	5	17	0.068
	5 - 10 years	9	45	5	17	
	>10 years	10	50	20	66	
Number of marriages	One	10	50	25	87	0.011
	≥ two	10	50	5	13	
Parity	Single parity	0	0	9	30	0.007
	Multiple parity	20	100	21	70	
Contraception	None	1	5	4	13.3	0.005
	IUD	6	30	4	13.3	
	O.C	12	60	8	26.7	
	Progesterone	1	5	14	46.7	
Duration of contraception	0	1	5	4	13.3	0.005
	<2 years	1	5	14	46.7	
	>2-4 years	6	30	4	13.3	
	> 4 years	12	60	8	26.7	

A statistically significant relations between the results of nested PCR and DM and genital warts. No statistically significant differences were recorded between results of nested PCR and recurrent infections, chronic infections, passive smoking or family history of cancers in the studied patients (Table 4).

Table (4): Correlation between medical and family risk factors of the 50 studied patients according to the results of nested PCR

Parameters / Risk factors		Positive n=20		Negative n=30		P-value
		n	%	n	%	
Medical history	Recurrent infections	6	30	9	30	1.0
	Chronic infections	7	35	8	27	0.39
	DM	7	35	3	10	0.03
	Genital warts	10	50	5	17	0.011
Passive smoking	No	3	15	2	7	0.335
	Yes	17	85	28	93	
Family history	Family history of cancer cervix	1	5	4	13	0.673
	Family history of other cancers	1	5	2	7	

DISCUSSION

Cancer Cervix remains a significant problem worldwide, particularly in underdeveloped countries. Prevention of cancer cervix can be made even better. Significant modifications of practice are imminent, motivated by improved understanding of HPV natural history and cervical carcinogenesis *(Thabet et al., 2014)*.

The present study included 50 women who had cancer cervix, classified as the following: 23 cases were squamous cell carcinoma (SCC) and 27 cases were adenocarcinoma (AD). HPV-DNA was detected in 20 cases (40%), were distributed as the following: 19 cases (95%) were SCC and 1 case (5%) was AD. this result disagrees with those reported by *Abdelbadiaa et al. (2016),* and *EL-Moselhy et al. (2017)*. The mean patient age as shown in table1 was 47.8±9.8 years. A similar age incidence was reported by *Thabet et al. (2014)*.

The association of HPV with cancer cervix in the current study was only 20/50 (40%). These results were not in accordance with many results *Abdel Azim et al. (2011)* (93.3%) *Bhatla et al. (2013)* (99.7%) and *Girgis et al. (2015)* (98%) reported that HPV is the main cause for developing of cancer cervix. The discrepancy may be explained by different sociodemographic criteria of our patients and the presence of other risk factors as smoking, hormones, and infections rather than HPV.

As regards the HPV genotypes ,the present study showed that the commonest HPV genotypes associated with cancer cervix were HPV 16; 5/20 (25%) ,HPV 31; 4/20 (20%) , mostly in the form of multiple infections with HPV 16+18;5/20 (25%) and HPV 6 and 11 separately each one represent (3/20); 15% while HPV18 represented once 1/20 (5%).which agrees with *Shaltout et al. (2014)* and *Thabet et al. (2014)*. The present study results are not concordant with those reported by *Al-Awadhi et al. (2011), Bansal et al. (2014)*

and *Kamel et al. (2018)*. The interpretation of different genotypes elsewhere was hampered by variation in HPV testing methodology, sensitivities of different methods used for HPV detection *(Haghshenas et al., 2013)*. In this study, single HPV types were observed more frequently (75%) than multiple types of infection (25%). Our findings are in line with *Chen et al. (2015)* and *Iwasaki et al. (2014)*.

HPV 16 and 18 (25%) co-infection and HPV 16 (25%) are the most presentation for HPV association with cervical cancer in our study, which may be due to synergistic action of multiple genotypes infection *(Monsonego et al., 2012)*.

Table 5 shows that HPV-DNA detection was prevalent among women between 40 and 49-year-old (45%). While, (15%) of the patients at the age group30-39 year, had HPV infections. This result agreed with the results reported by *Oliveira et al. (2013), Vaccarella et al. (2013)* and *EL-Moselhy et al. (2016)* who detected more HPVs in older patients. The reason for the higher prevalence of HPV in older patients could be due to poor or the lack of effective cervical cancer screening method and deficiency of using the most sensitive HPV detection methods e.g. molecular tools *(Richter and Dreyer, 2013)*.

As regards the residence, there is a significant relation between the residence and HPV infection i.e. almost patients who showed positive PCR are residents of rural areas. This result is concordant with studies done by *Irimie et al. (2011), Thabet et al. (2014)* and *EL-Moselhy et al. (2017)*, who showed that the populations from rural areas facing huge problems of poverty and a more difficult access to healthcare services.

Multi-marriages revealed a statistically significant association between HPV prevalence and the marital status in studied patients. This finding is consistent with different studies showed that the polygamous marriage was significantly associated with HPV prevalence *(Hammouda et al., 2011, Khodakarami et al., 2012* and *Thabet et al.,2014)*.

As regards the parity, statistically significant relation between HPV prevalence and parity of studied patients. These results are concordant with the results published worldwide, *Irimie et al. (2011), Demir et al. (2012)* and *El-Moselhy et al. (2017)*. On the contrary *Khodakarami et al. (2012), Sarma et al. (2013)* and *Thabet et al. (2014)* stated that women positive HPV-DNA, and the multiparity ≥4 insignificant. The difference in geographical region may be an element of such variation.

As regards contraception, the present study shows a statistically significant relation between HPV infections and contraception and a highly statistically significant relation between HPV infections and duration of contraception.

These results are coincident with *Irimie et al. (2011), Thabet et al. (2014)* and *El-Moselhy et al. (2016)*. Cervical squamous epithelium contains estrogen receptors that respond to chronic estrogen administration. Persistent proliferation stimulated by long-term use of contraceptives could initiate progression to invasive cancer *(Thabet et al., 2014)*.

On the other hand, some studies have shown no association between oral

contraceptive pills and the risk of HPV infection *Schmeink et al. (2010)*, and *Demir et al. (2012)*. The difference in association across studies suggests that oral contraceptives are exerting their influence on a very narrow window in the HPV natural history either at the point of HPV acquisition or at the point of the establishment of a persistent infection prior to the development of high-grade precancerous lesions or cervical cancer *(Marks et al., 2011)*.

Concerning the other risk factors associated with HPV infections, no significant differences were found between the patients regarding recurrent infections and chronic infections. There is a statistically significant relation between HPV infections and diabetes melitues and genital warts. The study results are in line with those reported by *Jalil et al. (2015)*, and *Fuchs et al. (2016)*. These are not concordant with the results reported by *Mohammad et al. (2017)*.

As regards smoking, there is no statistically significant relation between HPV infections and smoking. the present study results are matched with those recorded by *Abdel Azim et al. (2011)*. The study results are not concordant with those reported by *Thabet et al. (2014)* and *EL-Moselhy et al. (2016)*. Such variation might be due to cultural traditions and type of population.

As regards family history of cancer there is no significant relation between HPV infections and family history of cervical cancer or other cancers. This result is in line with *El-Moselhy et al. (2017)* and is not matched with *El-Moselhy et al. (2016)*. This controversy may be due to differences in the studied groups.

CONCLUSION

The prevalence of HPV in malignant cervical lesions was 40%, this means the association between HPV and the development of cancer cervix in Egyptian women cannot be fully established. HPV infection mostly 80%) in the form of single infections with HPV 16, 18, 31, 11, and 6 genotypes. However, HPV infections in multiple form, 16+18 genotypes in 25% of positive cases.

RECOMMENDATIONS

Further multi-central randomized studies are recommended to clarify the prevalence of HPV in premalignant and malignant cervical lesions in Egyptian women to determine the benefits of HPV vaccination in Egypt.

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الكشف عن الأنماط الجينية لفيروس الوَرَمِ الحُلَيمِيُّ البَشَرِيّ في مريضا ت أورام عنق الرحم بمحافظة دمياط، مصر

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خلفيــة البحــث: تبقــى عــدوى فيــروس الــورم الحليمــي البشــري مشــكلة كبيــرة فــي جميــع أنحــاء العــالم وخاصــة فــي البلــدان الناميــة. مــن الضــروري وجــود عــدوى دائمــة بفيروس الورم الحليمي البشري عالي الخطورة. لحدوث سرطان عنق الرحم.

الهــدف مــن البحــث: الكشــف عــن فيــروس الــورم الحليمــي البشــري وأنماطــه المحتملــة فــي عينــات عنــق الــرحم مــن بعــض المرضــى المصــريين، الــذين تــم تشخيصــهم باثولوجيا على أنهم مصابون بسرطان عنق الرحم.

المريضــات وطــرق البحــث: أجريــت هــذه الدراســة علــى 50 ســيدة أخــذت مســحات عنــق الــرحم وتــم فحصــها خلويــا وباثولوجيــا والكشــف عــن فيــروس الــورم الحليمــي البشــرى بواســطة تفاعــل البلمــره المتسلســل (PCR)، ثــم التنمــيط الجينــي للعينــات الإيجابيــة بواسطة تعدد طول جزء الاشكال المقيد (RFLP).

النتــائج: تــم اســتخلاص الحمــض النــووي DNA لفيــروس الــورم الحليمــي البشــري فــي 40 ٪ مــن المرضــى. تــم الكشــف عــن الــنمط الجينــي 16 فــي 25 ٪ والــنمط الجينــي 31 فــي 20 ٪ والــنمط الجينــي 11 فــي 15٪ (20/3) والــنمط الجينــي 6 فــي 10 ٪ والــنمط الجينــي 18 فــي 5 ٪ مــن الحــالات. 25 ٪ مــن عينــات ســرطان عنــق الــرحم أظهــرت عــدوى متعــددة بالأنمــاط الجينيــة 16و18والتــى أظهــرت مخــاطر عليــا لحــدوث ســرطان عنــق الــرحم. كانــت الارتباطــات بــين العمــر، والانتشــار فــي فئــة عمريــة محــددة، والإقامــة، والــزواج المتعــدد، وكثــرة الإنجــاب، ووســائل منــع الحمــل ومــدة اســتخدامها وداء الســكري كبيــرة. وفــي الوقــت نفســه، كانــت الارتباطــات بــين

الوظيفة، ومستوى التعليم، وسن الزواج، ومدة الزواج، والالتهابات المتكررة والمزمنة، والتدخين، وتاريخ السرطانات بالعائلات غير مهمة.

الاستنتاج: تشير الدراسة الحالية إلى أن معدل انتشار فيروس الورم الحليمي البشري في سرطان عنق الرحم كان 40 ٪، وهذا يعني أن الارتباط بين فيروس الورم الحليمي البشري وحدوث سرطان عنق الرحم في النساء المصريات لا يمكن أن يكون كاملا وكان 75% من عدوى فيروس الورم الحليمي البشري في شكل منفرد بالأنماط الجينية 16و18و31و11و6.وكان 25% من عدوى فيروس الورم الحليمي البشري في أشكال متعددة بالأنماط الجينية، 16 و18.