

ARTICLE ORIGINAL/ORIGINAL ARTICLE

**HUMAN PAPILLOMAVIRUS TESTING AS AN ADJUNCT TO CYTOLOGY
EVALUATION IN CERVICAL SPECIMENS OF SELECTED AND CONSECUTIVELY
SCREENED LEBANESE WOMEN : A PROSPECTIVE CLINICAL STUDY**

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ABSTRACT : Cervical cancer is the second leading cancer in women worldwide, with 85-100% of cervical cancers being HPV DNA positive. Our study aimed at evaluating the use of HPV DNA testing as an adjunct to the routine cervical cytology evaluation of Lebanese women. Cervical cytology evaluation and testing for human papillomavirus (HPV), as well as high-risk HPV were performed on two groups of women. One group consisted of 274 healthy women attending gynecology clinics for routine evaluation, while the other included 199 women selected based on a high-risk lifestyle and/or abnormal findings at cytology.

HPV and HR-HPV DNA were 4 and 10 times higher, and HPV was 4 to 5 times more frequent at all age groups in selected, compared to healthy women. HPV infection decreased in healthy women with age but did not decrease in selected women. HPV positive-normal cytology decreased with age in both groups. Although HR-HPV detection decreased with age in healthy women, it doubled in selected women. In addition, more severe cytology was associated almost uniquely with selected women. HR-HPV detection and advanced cytology lesions correlated well and were mutually predictive. The overall sensitivity, specificity, positive and negative predictive values of HPV testing for squamous intraepithelial lesions were (85.5%), (90.9%), (67.8%) and 96.5%, respectively.

Based on these findings, we recommend implementing HPV screening as an adjunct to routine cervical cytology in selected Lebanese women older than 30 years of age.

INTRODUCTION

Cervical cancer is the second leading cancer in women worldwide [1]. In Lebanon, official data on the prevalence of the disease do not exist, as a national tumor registry is not available. Genital human papillomavirus (HPV) causes nearly all cases of cervical cancer [2-3]. More than 70 HPV genotypes are currently known of which 15 types were associated with a high oncogenic risk and were classified as "high-risk HPV types"; these included types 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68, 73 and 82. Among those, HPV type 16 was the most common in all countries (Munoz et al. [4]).

Karam WG, Bedran F, Tohme RA, Moukarbel N, Abdallah I, Jurjus AR, Jurjus RA, Khairallah S, Aftimos G. Evaluation de la valeur du test de détection de l'ADN viral HPV comme un moyen adjuvant aux tests cytologiques de routine du col utérin chez les femmes libanaises : une étude clinique prospective. *J Méd Lib* 2005 ; 53 (3) : 132-138.

RÉSUMÉ : Par ordre de fréquence, le cancer du col utérin arrive en seconde position chez la femme dans le monde et dans 85 à 100% des cas est associé à la présence du virus HPV (human papillomavirus). Le but de notre étude est d'évaluer chez les femmes libanaises, la valeur du test de détection de l'ADN viral HPV comme un moyen adjuvant aux tests cytologiques de routine du col utérin. Nous avons évalué par cytologie et testé le virus HPV y compris le HPV génotype à haut risque chez deux groupes de femmes. Le premier groupe comprend 274 femmes saines se présentant pour un examen gynécologique de routine et le second groupe 199 sélectionnées suite à un mode de vie à haut risque et/ou à un résultat cytologique suspect.

L'ADN viral HPV et celui à haut risque oncogénique se sont avérés 4 et 10 fois plus fréquents respectivement chez les femmes du second groupe que chez les femmes saines. L'infection par le virus HPV diminue avec l'âge chez les femmes saines à la différence du second groupe qui ne montre pas la même évolution. Un résultat d'ADN positif et une cytologie normale diminue avec l'âge dans les deux groupes. Concernant le virus HPV à haut risque, malgré sa diminution avec l'âge chez les femmes saines, sa fréquence double chez les femmes sélectionnées qui présentent par ailleurs une cytologie plus sévère. Dans cette étude, une bonne corrélation a été obtenue entre la détection du virus HPV à haut risque et les lésions cytologiques avancées, ces deux paramètres étant interdépendants. La sensibilité globale, la spécificité et les valeurs prédictives positives et négatives du test HPV pour les lésions intraépithéliales malpighiennes sont de 85,5%, 90,0%, 67,8% et 96,5% respectivement.

En fonction de nos résultats, nous recommandons de mettre en pratique le dépistage du virus HPV comme test adjuvant aux examens cytologiques de routine du col utérin chez les femmes libanaises dont l'âge est supérieur à 30 ans et présentant un mode de vie qualifié à haut risque.

Geographic variations in the prevalence of HPV have been reported. In a recent review, Munoz and coworkers [4] reported on 1,928 population-, hospital-, and clinic-based control women, of which 259 (13.4%) were HPV DNA positive. The reported country-specific distribution of HPV infections ranged from a high of 33.3% in Mali, to a low of 5.2% in Spain [4]. Frequencies of less than

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10% have been reported for Western Europe and the United States of America, while West-Africa, Costa Rica and Mexico had up to 20% infection rates [5-8]. Mroueh et al. [9] in a study on 1,026 Lebanese women attending a tertiary care hospital in Beirut reported an overall prevalence of 4.9% in their study group. The age-specific prevalence in this study increased with age and peaked at 60-69 years.

HPV prevalence values also vary with age, it is highest in young women and declines with increasing age [5-6, 10-12]. U-shaped age-dependent curves have also been reported by many, with the first arm of the U corresponding to a peak in the late teens and early 20's, following the onset of sexual activity, and the second arm corresponding to a peak that occurs in the late 30s to 50s [6, 12]. The first peak consists mainly of HPV positive women showing normal cervical cytology [13-14]. In fact, HPV positivity with normal cytology has been associated primarily with transient HPV infections in younger women in which multiple concurrent and sequential infections with different types of HPV are common. Most of these infections have a median duration of less than 12 months and are not associated with cytologic changes [13-15]. In contrast, the second peak consists mainly of women showing dysplastic cervical cytology, and HPV infection is uncommon in older women in the absence of morphologic abnormalities [14, 16].

Since the 1950's, the Papanicolaou (Pap) smear has been used as a screening method for cervical cancer. The Pap smear test, however, has significant limitations. Screening and interpreting Pap smears could be a highly subjective and tedious task, which limits the sensitivity of Pap smears to 70%-80% [17]. It is estimated that 65% to 75% of the women with equivocal Pap smears do not have cervical disease [2]. Women who receive an ASCUS result are often treated as if they have abnormal Pap smears. Guidelines developed by the American Society for Colposcopy and Cervical Pathology (ASCCP) consensus conference recommends that HPV DNA testing be used as a management option for women with a Pap test interpretation of ASCUS, and consequently, the HPV test is currently the main ancillary test to be used in conjunction with Pap smear [18].

The utility of HPV DNA testing as an adjunct to routine cervical cytology evaluation is currently being reassessed. This is supported by the finding that HPV detection in Pap-smear negative women is predictive of an increased risk of subsequent detection of cervical intraepithelial neoplasia (CIN) ; that the risk of progression of HPV-related lesions may be correlated with viral type ; that a repeat negative cytology and HPV test confers a negative predictive value of close to 100% [19] ; and that women, who are Pap smear- and HPV DNA-negative, for 2 years in a row, are at virtually no risk of developing cervical cancer or a high-grade precursor in the next 5 to 10 years (assuming no new sexual partners). Moreover, a study at Georgetown University Medical Center showed that adding HPV testing to life-

time Pap screening can help prevent 225 cases of invasive cervical cancer per 100,000 women and can decrease cervical cancer mortality by an additional 59% over the use of a Pap test alone [20].

This study aims at investigating human papillomavirus testing as an adjunct to cytology evaluation in cervical specimens of selected and consecutively screened Lebanese women.

PATIENTS AND METHODS

Study group

During the period from 07/2002 until 07/2004, 274 healthy non-selected women attending outpatient clinics in three general hospitals in Lebanon for routine gynecologic evaluation, and 199 Lebanese women, selected from various clinics and hospitals in Lebanon, were enrolled in the study. Exclusion criteria were pregnancy, previous hysterectomy or an immunosuppressed status. Healthy non-selected women were fully informed of the purpose of the investigation ; they filled out a questionnaire about demographic and behavioral information and provided a written consent form. Inclusion criteria for selected women were a clinical history suggestive of a high-risk lifestyle (number of sex partners in the last year ; age at first intercourse, previous STDs, etc.) and/or abnormal cytology findings.

Clinical specimens

Specimens were collected with a wooden spatula by scraping the exocervix two to three times using circular motion. For the conventional Pap test, exfoliated cells were prepared as conventional smears, fixed with 70% ethanol and stained with an optimized Pap method. Cervical smears were evaluated by expert medical pathologists. Cytological diagnoses were made as per the Bethesda classification system. Namely, normal cytology, atypical squamous cells of undetermined significance (ASCUS), low-grade squamous intraepithelial lesions (LSIL), high-grade squamous intraepithelial lesions (HSIL) and carcinoma *in situ* (CIS).

HPV testing

Healthy women, were systematically tested while selected women were reflex tested for HPV following Pap smear evaluation. The exfoliated cells remaining on the spatula were then stored refrigerated in phosphate buffered saline (PBS) pH 7.4. DNA was subsequently extracted using the GFX blood genomic DNA extraction kit (Amersham Biosciences GmbH Wurzbachgasse Austria). Briefly, cervical cells were spun down for 5 min, extraction solution was then added and the lysate was applied to a silica spin-column. The column was then washed and the extracted DNA was eluted at 70 °C. DNA concentrations were determined spectrophotometrically at 260 nm.

Polymerase chain reaction (PCR) was performed following the methods of Mant et al. [21] and Fuginaga et

TABLE I
CHARACTERISTICS OF THE 274 HEALTHY
LEBANESE WOMEN

| CHARACTERISTIC | Number (%) |
|-----------------------------------|------------|
| AGE GROUP (years) | |
| 20-29 | 23 (8.4) |
| 30-39 | 114 (41.6) |
| 40-49 | 111 (40.5) |
| 50-59 | 24 (8.8) |
| 60-69 | 1 (0.35) |
| Mean age (years) ± SD | 39.4 ± 7.7 |
| Missing data | 1 (0.35) |
| SMOKING | |
| Nonsmoker | 130 (47.4) |
| Smoker | 76 (27.7) |
| Missing data | 68 (24.8) |
| MARITAL STATUS | |
| Married | 193 (70.4) |
| Single | 26 (9.5) |
| Divorced | 6 (2.2) |
| Missing data | 49 (17.9) |
| CONDOM USE | |
| Never | 141 (51.5) |
| Irregular | 49 (17.9) |
| Always | 15 (5.4) |
| Missing data | 69 (25.2) |
| ALCOHOLIC BEVERAGES | |
| Never | 44 (16.1) |
| Once a month | 87 (31.8) |
| Once a week | 59 (21.5) |
| Once a day | 14 (5.1) |
| Missing data | 70 (25.5) |
| EDUCATION | |
| University | 126 (46.0) |
| Technical | 41 (14.9) |
| School | 38 (13.9) |
| Missing data | 69 (25.2) |
| PREVIOUS PAP SMEAR | |
| Normal | 182 (80.9) |
| Abnormal | 23 (10.2) |
| Missing data | 69 (25.2) |
| AGE GROUP AT MARRIAGE | |
| 10-19 | 31 (11.3) |
| 20-29 | 140 (51.1) |
| 30-39 | 23 (8.4) |
| 40-49 | 2 (0.7) |
| Mean age at marriage (years) ± SD | 24.2 ± 4.7 |
| Missing data | 78 (28.5) |
| NUMBER OF LIVE BIRTHS | |
| None | 44 (16.1) |
| 1-2 | 118 (43.1) |
| 3-4 | 37 (13.5) |
| > 4 | 2 (0.7) |
| Missing data | 73 (26.6) |

al. [22]. Briefly, 1 µg of cellular DNA was denatured at 94 °C for 10 min, following which it was subjected to 35 cycles of denaturation at 94 °C, annealing at 55 °C and extension at 72 °C. The last cycle was followed by incubation at 72 °C for 10 minutes. The reaction mixture of 50 µl contained, 50 mM KCl, 10 mM Tris-HCl, pH 8.3, 800 µM dNTPs, 100 µg/ml gelatin and 2.5 units of Taq polymerase enzyme (ABgene Rochester, New York USA). Detection was performed using HPV L1-region consensus primers [21] and typing for high-risk HPV was done using high-risk e6/e7 ORF consensus primer combinations [22]. The typing method we used detects HPV types 16, 18, 31, 33, 52b and 58 [22]. Concurrent amplification of the CCR5 gene was performed as internal control, using primers described by Martinson et al. [23]. PCR products were separated by electrophoresis on 2% agarose containing ethidium bromide, in 0.5X Tris-borate EDTA running buffer, and visualized by trans-illumination with ultraviolet light. Digital photo-documentation was used to store collected data.

Cross contamination was avoided by using pipette-tips fitted with aerosol barrier filters, and frequent decontamination of work surfaces with short ultra-violet light irradiation and diluted bleach. Carry-over contamination was prevented by physically separating the extraction, amplification and detection areas, and, by incorporating the dUTP, uracil-DNA-glycosylase system into our amplification mixes (Roche diagnostics GmbH Mannheim Germany).

Data analysis

The statistical software SPSS 12.0 was used for analysis. The Chi-square (χ²) test was used to determine the existence of statistically significant correlations between different variables. Exact methods when expected cell frequencies were less than 5 were applied and a p-value < 0.05 was considered statistically significant.

RESULTS

Cytology reports of the 274 consecutively screened, non-selected women were 89.8% (246) normal, 5.8% (16) ASCUS, 4% (11) LSIL and 0.4% (1) HSIL (Figure 1). HPV DNA was detected in 13.2% (36) of the patients of which 1.5% (4) were high-risk HPV (HR-HPV). HPV DNA was detected in 10.2% (25) of normal cytology, 31.3% (5) of ASCUS, 54.5% (6) of LSIL and in the only HSIL specimen. HR-HPV was detected uniquely in 27.3% (3) of LSIL and in the only HSIL specimen. The age ranged from 20 to 67 years, with a mean of 39.6 ± 7.7 years. The age distribution was as follows : 8.4% (23) 20-29, 41.6% (114) 30-39, 40.5% (111) 40-49, 8.8% (24) 50-59 and 0.35% (1) 60-69 years (Table I) ; 70.4% (193) were married ; 27.7% (76) were smokers, among those 2.6% had a HR-HPV DNA detected while none of those who denied smoking was infected with HR-HPV ; 11.2% (23) had an abnormal Pap test result reported previously.

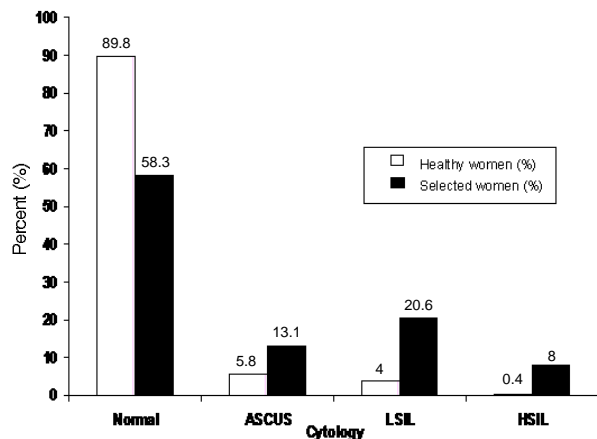


FIGURE 1
Cytology results in healthy and selected women.

HPV detection in this group of women was not associated with smoking habits (11.8% of smokers and 13.1% of non smokers were HPV positive), marital status (11.5% of singles and 15% of married were HPV positive), parity or use of male condoms. However, a statistically significant association was found between a previous abnormal Pap smear report and a current abnormal cytology result with HR-HPV DNA finding ($P < 0.05$) (Table II).

The prevalence of HPV in consecutively screened healthy women decreased with increasing age ; HPV DNA was detected in 21.7% (5), 14% (15), 13.5% (15), and 4.2% (1) of females in the (20-29), (30-39), (40-49), and (50-59 years) age categories respectively (Figure 2). This was paralleled by an age-associated decline in HPV positive specimens showing normal cytology, which decreased from 11.1% (2) to 10.5% (10) to 5.5% (5) and to 0% in the (20-29) (30-39) (40-49) (50-59) years age groups respectively. However, there was no statistically significant correlation between the overall HPV prevalence and age. HR-HPV was also more frequently detected among the younger age group with a frequency of 8.7% (2) for the 20-29 and a combined frequency of 0.9% (2) for both the 30-39 and 40-49 years age groups (Figure 3). HR-HPV was not detected in healthy non-selected women over the age of 50 years and the HSIL was from the 40-49 age group.

Moreover, cytologic diagnosis was not age-specific

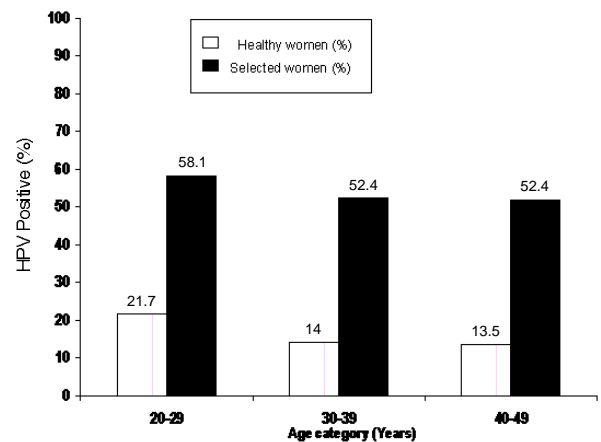


FIGURE 2.
Prevalence of HPV in healthy and selected women with age.

among the consecutively screened women. ASCUS was detected in 8.7% (2), 3.5% (4) and 9% (10) of the 20-29, 30-39 and 40-49 years age groups respectively ; however, none of those were HR-HPV. Furthermore, LSIL was detected in 8.7% (2), 3.5% (4) and 4.5% (5) of the 20-29, 30-39 and 40-49 years age groups, respectively. The correlation between cytology results and HPV status was highly statistically significant in this group between 30 to 49 years of age ($p < 0.0001$).

Cytology reports among the 199 selected women were 58.3% (116) normal, 13.1% (26) ASCUS, 20.6% (41) LSIL and 8% (16) HSIL (Figure 1) ; 51.8% (103) were positive for HPV, among those 14.6% (29) were HR-HPV. HPV DNA was detected in 11.1% (7) of normal cytology, 57.7% (15) of ASCUS, 85.4% (35) of LSIL and in 93.8% (15) of HSIL while HR-HPV was detected in 23.1% (6) of ASCUS, 19.5% (8) of LSIL and 68.8% (11) of HSIL. The presence and severity of cellular abnormalities were highly associated with the detection of HPV in selected patients ($p < 0.05$). The age range was 18-59 years with a mean of 34.8 ± 7.1 years. The age distribution was as follows: 21.6% (43) 18-29, 41.2% (82) 30-39, 26.1% (52) 40-49 and 1% (2) 50-59 years (Table III) ; 30.7% (61) of the selected patients had a previously abnormal Pap smear. A statistically significant association was found between a previous abnormal Pap smear report and a current abnormal cytology result with HPV DNA finding ($p < 0.001$).

TABLE II
STATISTICAL CORRELATIONS BETWEEN A PREVIOUS PAP RESULT WITH HPV GENOTYPE AND CURRENT CYTOLOGY IN HEALTHY LEBANESE WOMEN

| PREVIOUS PAP RESULT | HPV Genotype | | | <i>p</i> -value | Cytology | | | <i>p</i> -value |
|---------------------|--------------|-----------|----------|-----------------|-----------|-----------|-----------|-----------------|
| | HPV Negative | LR-HPV | HR-HPV | | Normal | ASCUS | LSIL | |
| NORMAL | 162 (79%) | 20 (9.7%) | | | 170 (83%) | 8 (3.9%) | 4 (1.95%) | |
| ABNORMAL | 18 (8.7%) | 4 (2%) | 1 (0.5%) | 0.042 | 17 (8.3%) | 4 (1.95%) | 2 (0.9%) | 0.008 |

DISCUSSION

As shown in the results, the overall general HPV prevalence was 4 times more frequent in selected women, and was 4 to 5 times more frequent in these women at all age groups. HPV infection decreased in healthy women with age, but not in selected women. HR-HPV was 10 times higher in selected women, and doubled in this group but decreased in healthy women with age. HSIL increased in selected women with age and was 8 to 40 times higher than in healthy women. HR-HPV was restricted to SIL in healthy women and, was detected in ASCUS and in SIL of selected women.

HPV positive-normal cytology was detected at a combined frequency of 10.1% (48/473) in our study groups. Others have detected HPV by amplified and non-amplified DNA detection techniques in up to 30% of cervical samples showing normal cytology [5, 7, 26-27]. In fact, the critical factor in determining the progression of HPV-infected normal-appearing cervical cells to a more severe cytology is the persistence of the infection. In this context, nononcogenic HPV infections clearance has been shown to be twice as fast as infections with oncogenic HPV types [28, 13]. The frequency of HPV positive specimens having normal cytology decreased dramatically with age in both groups studied, this trend has also been reported by others and reflects the acquisition of HPV near the onset of sexual activity followed by the resolution of infection [8, 10-12]. Overall, 25% (38/152) of HPV positive samples in our study had normal cytology, none of which were HR-HPV, while Mroueh et al. [9] reported 93.5% (29/31) in cytology matching cases all of which were HPV16.

A decline in overall HPV infection was seen in the group of healthy women and not in selected women with age. Although, the frequency of HPV-positive normal cytology smears decreased, this decrease was offset by a reciprocal increase in HR-HPV, resulting in no net change in overall HPV infection with age in this group. Mroueh et al. [9] reported an increase in HPV infections with age that peaked at 69 years. A similar trend was described in studies of West-African women and of women in Costa Rica and Mexico [5-7]. We did not observe this second peak, as most of our study population was younger than 49 years. This restricted age distribution in our study was due to the fact that HPV positive post-menopausal women were treated by hysterectomy, and were not followed thereafter for HPV infections. Although both our study and that by Mroueh et al. reported a doubling of HR-HPV with age, we observed this rise in the 40-49 years age group of selected women, while theirs was in the 60-69 years age group. Their overall HPV prevalence was about 50% lower than ours (4.9%).

Most studies report HR-HPV to be the predominant HPV type (mainly HPV16) in normal and abnormal cervical cytology [4, 6, 27, 29]. Our results do not agree

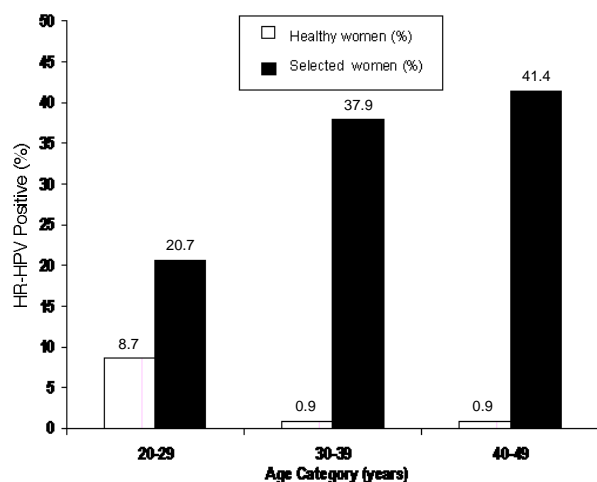


FIGURE 3

Prevalence of HR-HPV in healthy and selected women with age.

The prevalence of HPV in the group of selected women did not significantly decrease with advancing age and was as follows : 58.1% (25) 18-29, 52.4% (43) 30-39 and 51.9% (27) 40-49 years (Figure 2). In contrast, HPV positive specimens showing normal cytology decreased from 20% (3) at (20-29), to 14.8% (4) at (30-39), to 0% at (40-49) years of age and a contiguous and reciprocal rise of HR-HPV with age was seen in this group of selected women 20.7% (6), 37.9% (11) and 41.4% (12) in the 20-29, 30-39 and 40-49 age groups respectively (Figure 3). A parallel age-associated increase in the distribution of HSIL ranging from 3.2% (1) in the 20-29, to 11.9% (7) in the 30-39 and 16.3% (7) in the 40-49 years age categories was also noted in this group. The correlation between HPV type and cytology in this group was statistically significant between the age of 20 and 49 ($p < 0.05$ for the 20-29 age category and $p < 0.0001$ for the 30-49 age category).

TABLE III
CHARACTERISTICS OF THE 199 SELECTED
LEBANESE WOMEN

| CHARACTERISTIC | Number (%) |
|---------------------------|----------------|
| AGE GROUP (years) | |
| 18-29 | 43 (21.6) |
| 30-39 | 82 (41.2) |
| 40-49 | 52 (26.1) |
| 50-59 | 2 (1.0) |
| Mean age (years) \pm SD | 34.8 \pm 7.1 |
| Missing data | 20 (10.1) |
| PREVIOUS PAP SMEAR | |
| Normal | 99 (649.7) |
| Abnormal | 61 (30.7) |
| Missing data | 39 (19.6) |

with others that HPV16 is the predominant HPV genotype in cytology normal specimens; we have not detected HR-HPV in our normal cytology specimens. This difference may reflect different methodologies used in genotyping. For example, Mroueh et al. used real-time PCR and a fluorogenic probe to genotype their HPV samples, this method is more sensitive than ours. In addition, the lower sensitivity of the assay we used may be due to the use of consensus primers to detect HR-HPV genotypes-16, -18, -31, -33, -52b, and -58. The forward and reverse primers have three and two nucleotide mismatches with their respective binding sites in the e6/e7 ORF of HPV16. Despite of the lower sensitivity of the method we used, HR-HPV detection and severe cytology correlated well in our study, and, were mutually predictive (overall, 0% of normal 14.3% (6/42) of ASCUS, 21.1% (11/52) of LSIL and 70.6% (12/17) of HSIL were HR-HPV positive – $p < 0.001$) (Figure 4). Furthermore, the overall sensitivity, specificity, positive and negative predictive values of HPV testing for squamous intraepithelial lesions in our study were, 85.5%, 90.9%, 67.8% and 96.5%, respectively. In contrast, of 31 HPV16 positive samples reported by Mroueh et al., only 2 (6.6%) had abnormal cytology. In addition, it is well documented that the majority of HPV16 infections with normal cytology are only transient and will not progress to more severe lesions. In fact, Van Duin et al. [30] have demonstrated that women with normal cervical smears had lower HPV16 viral loads and that, a sustained or increased viral load is predictive of viral persistence and progression to CIN II/III. Therefore, in a clinical setting, the genotyping method we have used provided a better correlation with the severity of cytological lesions, and may be a better predictor of progression, as HPV16 genotype becomes detectable with rising HPV load. Nevertheless, the more sensitive method used by Mroueh et al. is preferred when determining the prevalence of various HPV genotypes in a specific population for future HPV vaccination strategies.

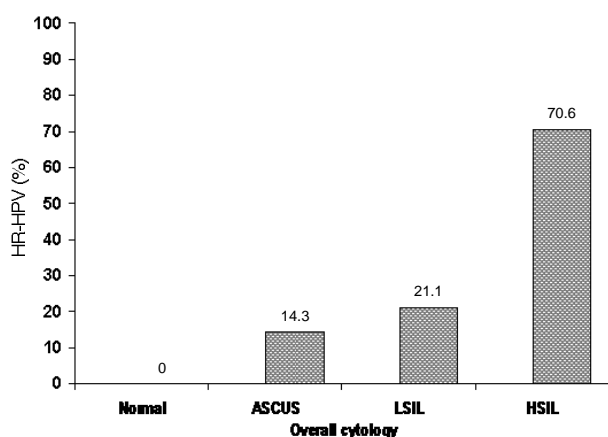


FIGURE 4
Overall distribution of HR-HPV by cytology.

Risk factors strongly associated with HPV transmission include among others, having multiple sex partners and early age at first intercourse [24]. Since genital HPV infection is sexually-transmitted, geographic, as well as regional variations in the distribution of HPV exist. In addition, reported HPV incidences are influenced by the methodology used to collect data, among these, the sensitivity of the method used is perhaps decisive [25]. Our study and that of Mroueh et al. investigated different sections of the Lebanese society, and used dissimilar methodologies to detect and type HPV. More comprehensive investigations encompassing all sections of the Lebanese society should therefore be undertaken before any systematic HPV screening is implemented in Lebanon. We conclude that since HPV prevalence and abnormal cytology results were significantly increased in the group of selected women, we therefore recommend implementing HPV screening as an adjunct to cervical cytology to such selected groups. Moreover, based on our findings, and in the absence of any other indications, we recommend performing this screening in selected Lebanese women older than 30 years of age. Finally, standardization of clinical evaluation of patients and a consensus on HPV detection methodologies to be used in HPV detection and typing will undoubtedly contribute positively to the effective management of cervical HPV infections in Lebanon.

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