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ORIGINAL RESEARCH

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Human papillomavirus infections in low-grade squamous intraepithelial lesion in Peruvian pregnant woman: A cross-sectional cytology-based study

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Abstract

Background and Aims: Low-risk human papillomaviruses (HPV) are important in pregnant women because they are risk factors for *Condyloma acuminatum* and recurrent respiratory papillomatosis in the newborn. On the other hand, HPV may contribute to the development of preneoplastic lesions such as LSIL in pregnant women. We aimed to assess the frequency of HPV infections in low-grade squamous intraepithelial lesion (LSIL) in Peruvian pregnant women.

Methods: A cross-sectional study was conducted from 2011 to 2015 in 84 primiparous- and multiparous pregnant women (mean age: 27 ± 6.3 years). Pregnant women of 18–45 years attending gynecology outpatient department were included for the study. LSIL and HPV (nuclear irregularities, koilocytosis, and cytopathic effect) cellular alterations were reported using the Bethesda System guidelines.

Results: Sixty-four percent had cytological HPV infection (koilocytes and/or pathognomonic signs of infection) and more than a half of pregnant women had a previous Pap test. LSIL was more frequent in multiparous (increased by 12%, $p = 0.008$), in the second and third trimester (60.7%, $p = 0.002$), and between the ages of 18 and 30 (42 cases (50%), $p = 0.110$). This proportion was significatively increased in women with ≥2 sexual partners (39.3%), with an early onset of first intercourse, and from the Rímac Municipality (14.3%) ($p < 0.05$). The rate of pregnant women with HPV infection increased by 39% between 2011 and 2015 ($p = 0.001$).

Conclusion: Peruvian pregnant women with LSIL have a high frequency of HPV infections. These young pregnant women (≤30 years of age), with multiple pregnancy, ≥2 sexual partners, and early onset of sexual intercourse were mainly associated with the third trimester HPV infection and LSIL. To detect early lesions of the cervix, it is key to continue monitoring HPV infections with molecular techniques and screening in pregnant women.

KEYWORDS

cervical cancer, cytology, human papillomavirus, Peru, pregnant women, Squamous Intraepithelial Lesions

1 | INTRODUCTION

Cervical cancer (CC) is the most preventable cancer and causes serious public health problems worldwide. If left untreated, CC can affect millions of people, especially the low- and middle-income countries (LMIC).[1,2] Human papillomavirus (HPV) is the causative agent of CC and is a common sexually transmitted pathogen worldwide.[3]

High-risk HPV (HR-HPV) causes a major number of CCs. HPV 16 is the most prevalent viral genotype in all the stages of Squamous Intraepithelial Lesion (SIL), principally in squamous cancer cells and adenocarcinoma.[3,4] The prevalence of HPV infection in women with SIL is very high and it is unrelated to the presence or absence of koilocytic atypia at the microscopic level. In low-grade squamous intraepithelial lesion (LSIL), the presence of HPV varies from 59% to 71%.[5]

Among these cases, the prevalence of low-risk HPV (LR-HPV) was 0.8%, with HPV 6, 11, and 42 being the most prevalent.[6] It has been shown that LR-HPV is not directly related to CC, as only a minority of cases progress to the tumor.[7] This apparent low degree of malignancy may be due to the absence or low expression of the early 4 (E4) protein and/or the absence or partial integration of the DNA of LR-HPV.[8–10]

Furthermore, in LR-HPV, the expression of E6/E7 is less controlled and does not appear to be significantly mutagenic. The progression to *Condyloma acuminatum* leading to the development of the Buschke-Löwenstein tumor appears to depend on mutagenic factors and the microenvironment.[11] Often, HPV 6 and HPV 11 are the cause of recurrent respiratory papillomatosis (RRP), a common benign tumor located primarily in the larynx and occasionally affecting the trachea and lungs of newborns, causing chronic childhood dysphonia.[11,12]

The degree of immunosuppression naturally caused by pregnancy increases the prevalence and persistence of HPV infection.[13] Thus, the surveillance of this group of patients is relevant and the Papanicolaou test (Pap test) offers a window of opportunity for the screening and identification of SIL and HPV during pregnancy, principally in its early stages. Several studies showed that the LSIL with koilocytic atypia could change between 1.5% and 6.8% with different ratios of regression and progression of SIL, pointing to the need for detection and follow-up of SIL in all pregnant women and related complications such as RPP and vulgar wart and oral papilloma in newborns.[14–17]

In Peru, at the beginning of the second decade of the 21st century, there was no national agreement on the exclusive screening of pregnant women, therefore their evaluation by cytology and their referral to a specialized hospital in the event of suspicion or detection of cellular abnormalities was recommended.[18] It is strongly recommended that routine prenatal visits include cervical screening when necessary, as this is often the only opportunity for cervical screening for many women.[19] This article aims to evaluate the frequency of HPV infections in LSIL in pregnant women in a tertiary-care hospital in Lima, Peru.

2 | MATERIALS AND METHODS

2.1 | Study design, sampling, and inclusion criteria

This prospective cross-sectional study was conducted during 2011–2015 at Hospital Nacional Docente Madre Niño San Bartolomé (HONADOMANI SB) in Lima, Peru. The Institutional Review Board of the hospital approved the study (registry – N° 01220-OADI-UI-2017-OADI HONADOMANI-SB).

Pregnant women with a cytological diagnosis of LSIL (84 patients) who met the following inclusion criteria in the study were included: (1) Pregnant women who visited outpatient clinics from 2011 to 2015, (2) Pregnant women between the ages of 18 and 45, (3) Pregnant women in the obstetrics and gynecology clinic, and (4) Primiparous and multiparous pregnant women with complete medical records. Our exclusion criteria were women of other nationalities, and women with recent HIV treatment or diagnosis. The sample size was calculated using EPIDAT v4.1 (Xunta de Galicia), considering the sensitivity of 95%, heterogeneity of 50%, and a margin of error of 0.05. A sample size of 70 pregnant women was obtained (study population).

Cervical samples from all asymptomatic pregnant women included in the study were referred from the Gynecology Department outpatient consulting rooms, the High-Risk Obstetric Unit, and the Teenage Programs, after an informed consent form from each participant was previously approved and signed.

2.2 | Cytological procedure

Specimens were received and processed at the institution's Department of Pathology. Most of the hospital Pap tests come from primary care centers that work as an integrated gynecological care network, however, these samples are only reviewed in the hospital and some important epidemiological data are not available for all patients. Specimens were obtained according to national guidelines.[2,20] Conventional sampling was carried (with Ayre spatula and cytobrush) out in outpatient clinics by hospital health professionals. The immediate fixation of the samples was in 95% alcohol for 5 min. The samples were coded and sent to the Department of Pathology for analysis within 2 h (all slides shipped together) of sampling and specimen processing were carried out in accordance with the standardized organizational procedures (SOPs) established by the hospital.[21,22]

Processing included Papanicolaou staining and review first by cytotechnologists and then by pathologists. The exfoliative cytological processing followed a conventional method.[20,23] We used the Bethesda System (TSB) guide 2001 and 2014 for the report of cytological findings (mainly LSIL and HPV cellular alterations).[24,25] The staining and reading process presents important checkpoints for quality assurance following the SOP.[2,20] In this study, we follow the following operational definitions: HPV in cytology is defined by pathognomonic cytological changes such as koilocytosis, hyperkeratin

granules, or cytopathic effect. LSIL is defined by the presence of binucleation with nuclear features (i.e., enlargement, hyperchromasia, and contour irregularities), perinuclear cytoplasmic clearing (koilocytosis), and cytopathic effect.

2.3 | Data gathering and analysis

We used a structured record, which registered data from medical records of pregnant patients diagnosed with LSIL and followed the International Statistical Classification of Diseases and Related Health Problems with the code ICD-10 Diagnosis Code N87.0 (ICD-10-N87.0) to determine the presence or absence of HPV. We analyze clinical and demographic data such as the place of origin. Lima, the capital of Peru, is divided into five zones and borders the Region of Callao to the west. Roughly, 10,000 inhabitants live in Lima, and they are distributed mainly in the suburban of the capital.

Descriptive statistics were used to determine the frequency and distribution of values. Paired *t*-test and one-way analysis of variance were used for comparison between variables. Also, we used Kendall's τ-b test for the association of previous Pap test and LSIL results considering a confidence interval (CI) at 95%, and a *p*-value <0.05 as statistically significant for all tests. The data analysis was performed in the IBM SPSS v21.0 (Armonk) for Windows

3 | RESULTS

During the time of the study, we included 84 (0.2%) pregnant and the mean age of all patients was 27 ± 6.3 years (range: 18–45 years) (Table 1). Forty-one thousand six hundred and seventy-eight Pap tests were performed in pregnant women (6121 Pap tests in 2011, 8571 Pap tests in 2012, 7304 Pap tests in 2013, 9231 Pap tests in 2014, and 10,451 Pap tests in 2015). The total of pregnant women with LSIL is 0.2% (range: 0.18–0.23), and no significant difference was found between the years of study ($p = 0.752$).

Of the 84 cases, we determined that 64.4% (54 patients) had cytological HPV infection (koilocyte and/or pathognomonic signs of infection). The rate of pregnant women with HPV infection increased by 39% between 2011 and 2015 ($p = 0.001$). The distribution of results with LSIL and HPV diagnosis per pregnant trimester is shown

TABLE 1 Frequency of LSIL with HPV positive or negative in pregnant after the cytological evaluation 2011–2015

		LSIL					
		HPV positive		HPV negative			
Year	Annual Pap tests[a]	18–30 years old	31–40 years old	18–30 years old	31–40 years old	41–45 years old	Total
2011	6121	6 (7.1)	1 (1.2)	2 (2.4)	2 (2.4)	1 (1.2)	12 (14.3)
2012	8571	6 (7.1)	4 (4.8)	5 (6)	-	-	15 (17.9)
2013	7304	11 (13.1)	1 (1.2)	2 (2.4)	2 (2.4)	-	16 (19)
2014	9231	10 (11.9)	1 (1.2)	5 (6)	1 (1.2)	-	17 (20.2)
2015	10,451	8 (9.5)	6 (7.1)	9 (10.7)	1 (1.2)	-	24 (28.6)
Total	16,572	41 (48.8)	13 (15.5)	23 (27.4)	6 (7.1)	1 (1.2)	84 (100)

Note: Data in *n* (%). $\chi^2 = 0.76$; *p*-value = 0.0152 (significant).

Abbreviations: HPV, human papillomavirus; LSIL, low-grade squamous intraepithelial lesion.

[a]Samples from other health centers have been excluded [see Seguro Social del Perú[18]], only hospital samples have been included.

TABLE 2 Distribution of pregnant women with LSIL differentiated by trimesters of pregnancy

	LSIL					
	HPV positive		HPV negative			
Trimesters	18–30 years old	31–40 years old	18–30 years old	31–40 years old	41–45 years old	Total
I Trimester	2 (2.4)	1 (1.2)	1 (1.2)	-	-	4 (4.8)
II Trimester	14 (16.7)	2 (2.4)	10 (11.9)	2 (2.4)	1 (1.2)	29 (34.5)
III Trimester	26 (31)	9 (10.7)	11 (13.1)	5 (6)	-	51 (60.7)
Total	42 (50)	13 (14.3)	22 (26.2)	7 (8.3)	1 (1)	84 (100)

Note: $\chi^2 = 0.51$; *p*-value = 0.003 (significant).

Abbreviations: HPV, human papillomavirus; LSIL, low-grade squamous intraepithelial lesion.

in Table 2, where we find different statistical significance between each trimester of pregnancy ($p = 0.003$). When we assessed frequency differentiated by the number of pregnancies (multiple and first pregnancy), LSIL increased by 12% in pregnancies with multiples compared with the first pregnancy. In this sense, LSIL is more frequent in multiparous than with one previous pregnancy ($p = 0.008$).

Table 3 shows the principal clinical risk components of this study. The age group where there was more HPV-positive in LSIL was 18–30 years with 42 cases (50%), in this, we observed the highest number of cases (8 cases, 9.5%) in pregnant women of 19 years. No statistical relationship was found between age and HPV-positive LSIL ($p = 0.110$). In both cases (HPV positive or not) of gestational status, 58.3% of pregnant women were multiparous (49 cases) nearly all with HPV-positive LSIL, although we did not find a significant relationship ($p = 0.896$). In the same way, 60.7% of pregnant women in the second and third trimesters presented HPV in LSIL, this showed a significant difference with patients with HPV-negative LSIL ($p = 0.002$).

Other factors evaluated included the number of sexual partners, age at first intercourse, and miscarriage rates, all of which showed significant differences between pregnant women with and without HPV in LSIL ($p < 0.05$). Twenty-seven percent of pregnant women had a history of ≥1 miscarriage, but no significant association was found ($p = 0.460$). We also did not find a significant association between HPV-positive LSIL and the age of first intercourse ($p = 0.075$), nor with the number of sexual partners ($p = 0.921$). As for the latter, 59% had ≥2 sexual partners (range: 2–5 couples) in the last year before the assessment. In both groups of positive and negative HPV patients, the age of first intercourse was in the range of 16–19 years (47.6%), and in the range of patients aged 13–15 years, we observed a great difference among the patients who had HPV presence in LSIL ($p = 0.008$).

When the frequency of LSIL was assessed according to patient origin (distributed by the municipality), Lima downtown (37%) had the highest number of HPV patients (22 patients, 26.2%), followed by North Lima with 19 HPV-positive patients (32.1%) and 2 HPV-positive patients from other areas. In the Municipalities that make up northern Lima, six (7.1%) LSIL HPV-positive patients came from San Martin de Porres four (4.8%) from each Comas and Independencia, 3 (3.6%) from Puente Piedra, and 2 (2.4%) from Los Olivos. LSIL-HPV-positive patients in eastern Lima belonged to the Municipalities of San Juan de Lurigancho (three cases, 3.6%), La Molina (two cases, 2.4%), and Lurigancho (one case, 1.2%). In the downtown area of Lima, 12 (14.3%) LSIL HPV-positive patients were from Rímac, 9 (10.7%) from Cercado de Lima, and 1 (1.2%) from Santiago de Surco Municipalities. In Callao, only four (4.8%) patients were positive for LSIL HPV (Figures 1 and 2).

Finally, more than 50% of pregnant women had a previous Pap test, but we did not show a significant association with HPV-positive LSIL ($p = 0.416$). When we rescreening this Pap test, all the results issued by the reviewers were positive for LSIL (with or without HPV) ($p = 0.787$). Figure 3 shows the cytological characteristics of four LSIL patients' results (two with HPV-positive results).

TABLE 3 Baseline characteristics associated with frequency HPV infection in pregnant women ($N = 84$)

	LSIL			
	HPV positive	HPV negative	Total	*p*
Age				
18–30	42 (50)	22 (26.2)	64 (76.2)	0.001
31–40	12 (14.3)	7 (8.3)	19 (22.6)	
41–45	0 (0.0)	1 (1.2)		
Trimester of pregnancy				
I	3 (3.6)	1 (1.2)	4 (4.8)	0.002
II	16 (19)	13 (15.5)	29 (34.5)	
III	35 (41.7)	16 (19)	51 (60.7)	
Pregnant status				
Primiparous	23 (27.4)	12 (14.3)	35 (41.7)	0.057
Multiparous	31 (36.9)	18 (21.4)	49 (58.3)	
Result of the previous Pap test[a]				
Realized	30 (35.7)	17 (20.2)	47 (55.9)	0.458
Nonrealized	17 (20.2)	6 (7.1)	23 (27.4)	
Abortions				
Yes (≥1)	14 (16.7)	9 (10.7)	23 (27.4)	0.021
None	40 (47.6)	21 (25)	61 (72.6)	
Number of sexual partners[b]				
1	13 (15.5)	8 (9.5)	21 (25)	0.011
≥2	33 (39.3)	17 (20.2)	50 (59.5)	
Age of onset of sexual intercourse[b]				
13–15	12 (14.3)	1 (1.2)	13 (15.5)	0.008
16–19	23 (27.4)	17 (20.2)	40 (47.6)	
>20	11 (13.1)	7 (8.3)	18 (21.4)	

Note: Data in *n* (%).

Abbreviations: HPV, human papillomavirus; LSIL, low-grade squamous intraepithelial lesion.

[a]Seventy patients were evaluated.

[b]Seventy-one patients were evaluated.

4 | DISCUSSION

In this study of asymptomatic pregnant women with LSIL, more than half showed HPV-positive results in cytology screening. This proportion was increased in women with ≥2 sexual partners, with an early onset of first intercourse, and from the Lima downtown, mainly from Rímac Municipality (14.3%). In addition, a large

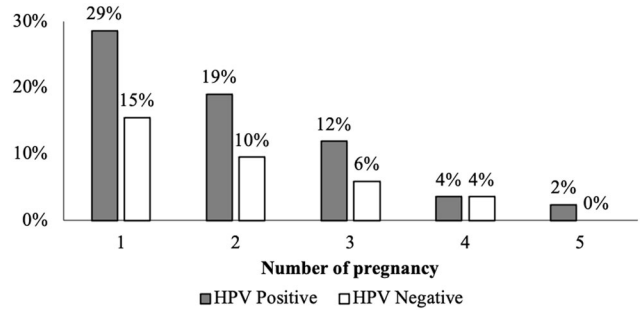

FIGURE 1 Percentage of number of pregnancies distributed by quant of born. Data in %.

proportion of these are young women in the third trimester and with multiple births.

The main strength of our study is that it is the first cytological-based report of pregnant with LSIL related to the presence of HPV and others risk factors in a tertiary-care hospital in Lima, Peru. Another strength of the study was the quality workflow in the reading and diagnosis of cytological abnormalities (LSIL) in Peruvian pregnant women, as well as the standardized interpretation with the TSB.

A major limitation of this study was the size of the sample of pregnant women with cytology results for LSIL belonging to the hospital. The vast majority of patients analyzed by exfoliative cytology belong to health's micro-networks, which do not have a clinical history and data considered in this study available for analysis, also each health network manages its own healthcare systems.[26] Other limitations of the study are our inability to perform HPV genotyping testing to determine whether a pregnant woman has LR-HPV, HR-HPV infection, or both, as well as multiple infections, which results are important and often required as a tool for CC screening. Furthermore, it was not possible for cytological follow-up of the delivery of these patients to know the persistence, regression, or progression of LSIL. Likewise, we were unable to obtain previous Pap test results in pregnant women during prenatal control, which could provide valuable data on preneoplastic or neoplastic lesions and HPV infection.

According to our results, the LSIL's HPV infections were more frequently observed in young women (50%), than in adults (Table 1). Consistent with previously reported results, the highest number of HPV-positive LSIL cases was in pregnant women between the ages of 18 and 30.[16,26–28] Recent studies have shown that young women have the highest rates of HPV infection (≤25 years, range: 20–35 years), tapering to ≤5% (after age 55) due to low exposure, altered sexual behavior, and acquired immunity. Undoubtedly, the relationship between age and HPV infection has been established, although a synergistic effect of multiple infections has not been observed with the progression of high-risk SIL to CC, where single infections prevail.[29] After attending puberty, physiological migration of Squamocolumnar Junction towards the endocervix occurs, in this process, there is the replacement of the cylindrical epithelium by the stratified squamous epithelium forming the transition zone, where the risk of neoplastic transformation is greater than any other tissue exposed to cancer.[14,30]

Our results are consistent with previous studies,[15,17,31] albeit with divergence in the group of women who were assessed as less frequent than the general pregnant population. We determined a frequency of HPV-positive LSIL of 64.3% that agrees with the results of Alves de Souza et al.[32] which shows a frequency of 63% but disagrees with a previous study in Peruvian women where a frequency of 46.1% is indicated.[33]

This low frequency reported may be due to the fact that a general population of women belonging to different provinces of Peru was analyzed, with different sexual behavior and culture, and also the difference in years of collection of samples from patients. Likewise, our results coincide with the Latin American frequency (68.3%), the European frequency (67.8%), and the Asian frequency (67.1%) of HPV-positive in LSIL. Although the world (71.1%), African (59.1%), and from North America (80.1%) frequencies disagree with our findings, we believe that this may be due to the use of various diagnostic methods for the detection of HPV.[5]

Multiparity (≥3 births) is associated with an increased risk of HPV infection due to increased exposure to the virus, increased sexual intercourse, gestational immunosuppression, and possible cervical lesions or infections that may arise during childbirth altering the normal limits between the two epithelia in the transition zone.[34] As observed by other research groups, our results show that multiparous women with LSIL have a higher frequency of HPV infection.[35] Physiologically, this relationship can be explained because, during pregnancy, the transformed area of the cervix changes and becomes more extensive, leading to the origin of the precancerous lesions.[36] All these factors induced during pregnancy, mainly the immunosuppressive state and the exacerbation of hormone secretion at the end of pregnancy, may favor the proliferation of HPV and the progression of SIL, respectively. According to other studies,[37] we determined that LSIL and HPV infection was higher in pregnant women during the third trimester of pregnancy.

Other risk factors evaluated in this study were the abortion rate, the number of sexual partners, and the age of first intercourse (Table 3). Overall, a larger proportion of patients (72.6%) had a history of nonabortion. Our moderate frequency of LSIL pregnant women, history of previous miscarriage, and the presence of HPV are consistent with other molecular studies of the Mexican general population.[38] Although abortion causes multiple injuries to the cervix caused by the instrumentation used at the time of praxis, it is still unknown that it can be a risk factor for HPV infection.

In our analysis of risk factors, >50% of pregnant women had ≥2 sexual partners in the past year. There is a relationship between the risk of HPV infection and the number of sexual partners, this relationship is most due to the likelihood of exposure to HPV as reported by various authors in their results of association of both factors in population studies of the region.[39,40]

Likewise, the early onset of sexual intercourse also involves multiple sexual partners, which infers an increased risk of HPV

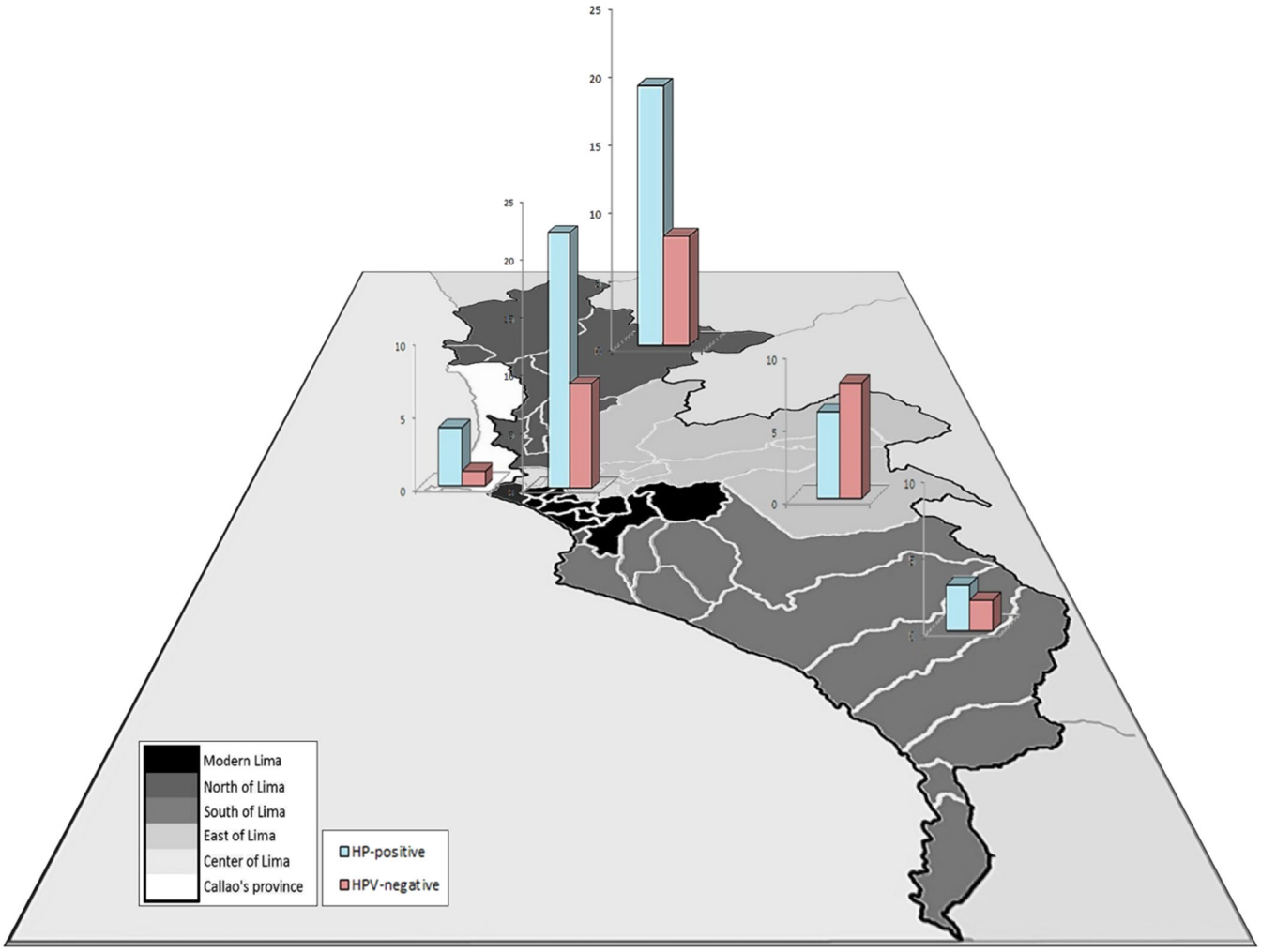

FIGURE 2 Distribution of patients with LSIL according to district of origin in Lima and in the Constitutional Region of Callao, Peru. In the North of Lima, there were 19 (22.6%) patients with human papillomavirus (HPV) and eight without HPV (9.5%). In South of Lima, there were 3 (3.6%) and 2 (2.4%) patients with HPV and without HPV, respectively. From the Lima Center, there were 22 (26.2%) patients with HPV and 9 (10.7) patients with HPV-negative. In the East of Lima, 6 (7.1%) patients with HPV-positive and 8 (9.5%) with HPV-negative were observed. In Callao region, there were four (4.8%) patients with HPV and one (1.2%) HPV-negative. Data (*n*). Map created by Jeel Moya-Salazar© in ArcGIS 10.8 (Environmental Systems Research Institute).

infection. During youth, cervical tissues are more susceptible to the action of carcinogens, when the age of first intercourse is ≤17 years there is a greater risk of producing intraepithelial lesions than when the first coitus is at 21 years of age. Significantly, we show more pregnant women ≤19 years with HPV-positive LSIL than pregnant women without this infection, these results agree with the results of Krüger-Kjær et al.[40] which show three times more (74.4%) of LSIL with HPV in women with a sexual debut at ≤17 years compared with those at onset ≥18 years. In addition, more studies should be performed since we assume that the vast majority of these patients have their first intercourse with older couples who have had multiple sexual partners, which would represent a higher risk of infection by HPV and other sexually transmitted infections.

Due to its high throughput, the WHO 2021 guidelines on CCs recommend that HPV testing be implemented globally. However, because not all countries have fully operational HPV molecular testing, the guideline recommends continued cytology testing.[41] In this study in asymptomatic pregnant women, the Pap test has made it possible to describe LSIL and the presence of HPV at the same time. This benefit of cytology is important because, in a setting where there is no full access to HPV testing, cytology can guide the follow-up of patients with cervical abnormalities. Thus, cervical exfoliative cytology is one of the indispensable tools for CC screening in LMICs. Unfortunately, there are limitations that hinder the success of programs for the prevention and control of CC.[42] We showed in this study that 27.4% of pregnant women with LSIL had no previous Pap test, which attempts against prenatal control and secondary prevention of this population group, which is subject to several factors that can condition the progression of SIL to CC.

Given the associations between age at diagnosis and aggressive clinical behavior reported in the US National Registry of RRP, Danish, and Colombian studies, the assessment of HPV in pregnant women is

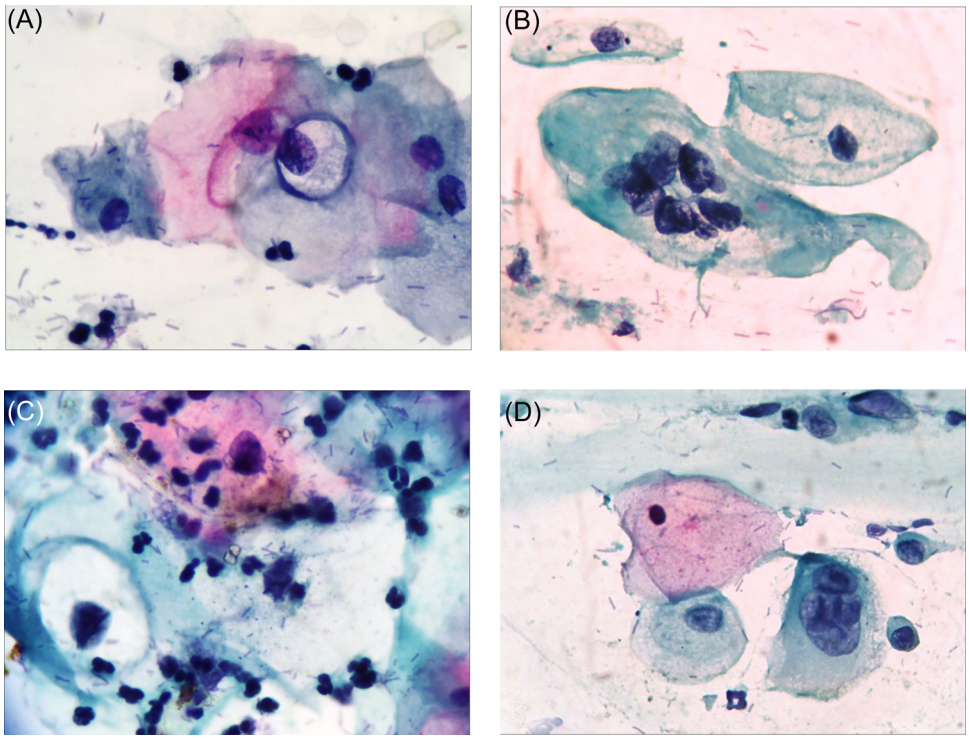

FIGURE 3 Human papillomavirus (HPV) infection in low-grade squamous intraepithelial lesion (LSIL) pregnant woman. (A–C) HPV-positive in LSIL (cases 33, 12, and 1, respectively); (D) HPV-negative in LSIL (case 22). (A, C) koilocytes; (B) pathognomonic signs of HPV infection (multinucleation and perinuclear halo); (D) clean background, nucleus/cytoplasm ratio slightly altered, and binucleation; (C) eosinophilic and basophilic koilocytes. All slides in 40×, Modified Pap stain (Eco-Pap).[21] Photographs by Victor Rojas-Zumaran.

most important for RRP.[43–45] Although it is a benign disease that normally involves only the larynx, the RRP has an unpredictable clinical course, tends to recur, extends to the rest of the aerodigestive tract, and can undergo a malignant transformation from 3% to 7% of cases.[13,46]

Transmission of HPV 6 or HPV 11 in juvenile-onset RRP is most likely to occur in mother-to-child transmission while the fetus is passing through an infected birth canal.[47] RRP is a rare pediatric pathology but difficult to manage and may compromise the patient's life. In children, the most common oral mucosal epithelial tumor manifestations are the vulgar wart and oral papilloma, however, studies in the pediatric population are scarce and require, ideally, prospective studies to obtain representative samples, typifying the lesions in children, which are small and vulnerable population, and where preventative maneuvers can be made (adequate maternal anamnesis to define the route of delivery and decrease the risk).

Finally, there is no pathognomonic image of pregnancy without clinical data, so the dichotomous conjunction of these makes it possible to make an accurate diagnosis. The cytological image of normal pregnancy has characteristics similar to the second phase of the menstrual cycle, with navicular cells, most of the grouped intermediate cells, döderlein bacilli, and background; any change in these cell populations necessarily indicates a sign of alarm over abortion, distress, or fetal death, or SIL.[48] All our cytological evaluations of pregnant women with LSIL should continue with molecular studies of HPV genotyping to better understand the actions of prevention and clinical management (this is a necessity in Latin America and in other LMICs).[49] Despite the large amount of HR-HPV progressing to CC, LR-HPV can also progress to SIL to CC, since in HPV 6 and HPV 11 the E7 protein also has an affinity, although reduced by Retinoblastoma protein.[50]

5 | CONCLUSION

In Peruvian pregnant women with LSIL have a high frequency of HPV infection that is close to the regional and global averages for the female population. These young pregnant women (≤30 years of age), with multiple pregnancy, ≥2 sexual partners, and early onset of sexual intercourse (≤19 years of age) were mainly associated with the third trimester HPV infection and LSIL. This cytology-based study is the first report on HPV-positive in LSIL in Peru that establishes the need to use molecular tests for HPV in this risk group and in the general population.

AUTHOR CONTRIBUTIONS

Jeel Moya-Salazar: Conceptualization; data curation; investigation; methodology; writing–original draft; writing–review & editing. **Víctor Rojas-Zumaran**: Conceptualization; investigation; methodology; writing–original draft; writing–review & editing. **Omar Bravo L.**: Data curation; formal analysis; methodology; writing–original draft; writing–review & editing. **Gonzalo Moscoso**: Conceptualization; data

curation; project administration; writing–original draft; writing–review & editing. **Hans Contreras-Pulache**: Data curation; investigation; supervision; writing–original draft; writing–review & editing.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

Jeel Moya-Salazar confirms that she had full access to all of the data in this study and takes complete responsibility for the integrity of the data and the accuracy of the data analysis.

TRANSPARENCY STATEMENT

The lead author Jeel Moya-Salazar affirms that this manuscript is an honest, accurate, and transparent account of the study being reported; that no important aspects of the study have been omitted; and that any discrepancies from the study as planned (and, if relevant, registered) have been explained.

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