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Original Research

The Cervicovaginal Expression Level of Interleukin-6 Can be a Prediction Factor for Cervical Intraepithelial Neoplasias and Cervical Cancer: A Prospective Cohort Study

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Abstract

Background: The aim of this prospective study was to investigate the expression of interleukin-6 (IL-6) of cervicovaginal washings in cervical intraepithelial neoplasia (CIN) and cervical cancer, and to explore its value in predicting the treatment of CINs. **Methods**: Cervicovaginal washings were collected from of 200 patients from Tianjin First Central Hospital, who underwent colposcopy examination between March 2021 to March 2022. Those patients included 13 patients with cervical cancer, 33 patients with CIN3, 46 patients with CIN2, 76 patients with CIN1, and 32 with chronic cervicitis. IL-6 expressions of cervicovaginal washings were determined by enzyme-linked immuno-sorbent assay (ELISA), and other clinical records were obtained for analysis. **Results**: IL-6 expressions of the cervicovaginal washings in 5 groups of chronic cervicitis, CIN1, CIN2, CIN3, and cervical cancer group were 9.23 $\pm$ 11.30 pg/mL, 8.32 $\pm$ 12.18 pg/mL, 11.24 $\pm$ 24.61 pg/mL, 13.96 $\pm$ 23.19 pg/mL, and 71.53 $\pm$ 55.76 pg/mL, respectively. IL-6 expression in patients with cervical cancer were significantly higher than that in patients with chronic cervicitis and CIN1, 2, and 3 (p = 0.002, p = 0.003, p = 0.002, and p = 0.002, respectively). Particularly, IL-6 expression in CIN1, 2, 3, and cervical cancer is gradually increased which may be an indicator for CINs treatment. Besides, among 187 patients with high-risk human papillomavirus (HPV) infection, the IL-6 levels in HPV16/18 infection group and other high-risk HPV infection groups were 23.44 $\pm$ 43.33 ng/mL and 11.73 $\pm$ 23.37 ng/mL, which is statistically different (p = 0.043). **Conclusions**: IL-6 expression in cervicovaginal washings of CIN patients gradually increased suggesting that IL-6 may be involved in the whole process of cervical intraepithelial neoplasia developing into cervical cancer and can be a treatment biomarker for CINs.

Keywords: cervicovaginal washings; cervical intraepithelial neoplasia; interleukin-6; treatment predictor

1. Introduction

Cervical cancer is ranked the fourth in incidence and the third in mortality in women's malignancies worldwide [1]. There are more than one-quarter of a million deaths due to cervical cancer per year as the result of deficient treatments in many developing countries [2]. The vast majority of cervical cancers are a result of human papillomavirus (HPV) infection. Usually, most HPV infections are cleared naturally through humoral and cell-mediated immune responses. The persistence of HPV infection is currently considered as an independent factor for the development of cervical intraepithelial neoplasia (CIN) to cervical cancer. It is known that HPV infection possibly alters the percentages and ratios of immune cell sub-types in the local cervix. The expression of cytokines and growth factors in local cervix as prognostic and diagnostic factors are associated with the evolution of cervical lesions. CIN experiences CIN1, 2, and 3, and it usually takes about 10 years to develop into cervical cancer [3], during which effective intervention approaches can be applied to prevent its progress. In recent years, CIN and cervical cancer have more often been diagnosed in younger women ages (25–45 years old) [4,5]. Dunne *et al.* [6] reported that the incidence of CIN2 and CIN3 is happening in 1.5 per 100 women, with the peak prevalence age occurring in 25–35-year-old women in the USA. Meanwhile, with the increasing late childbearing, the incidence of patients undergoing conization before pregnancy is increasing [7]. Previous studies have suggested that conization is associated with a significantly increased risk of miscarriages in the second trimester [8], preterm birth [9], low birthweight, premature rupture of the membranes and perinatal mortality [10,11]. In recent years, researchers have found that CIN2 also has a natural regression rate of up to 66% [12]. Therefore, the treatment of CIN2 is becoming more and more conservative. For young patients of CIN2 with fertility requirements, it is important to make an appropriate treatment option to neither miss further diagnosis of cervical cancer, nor receive excessive treatments. Consequently, we herein aim to find a molecular

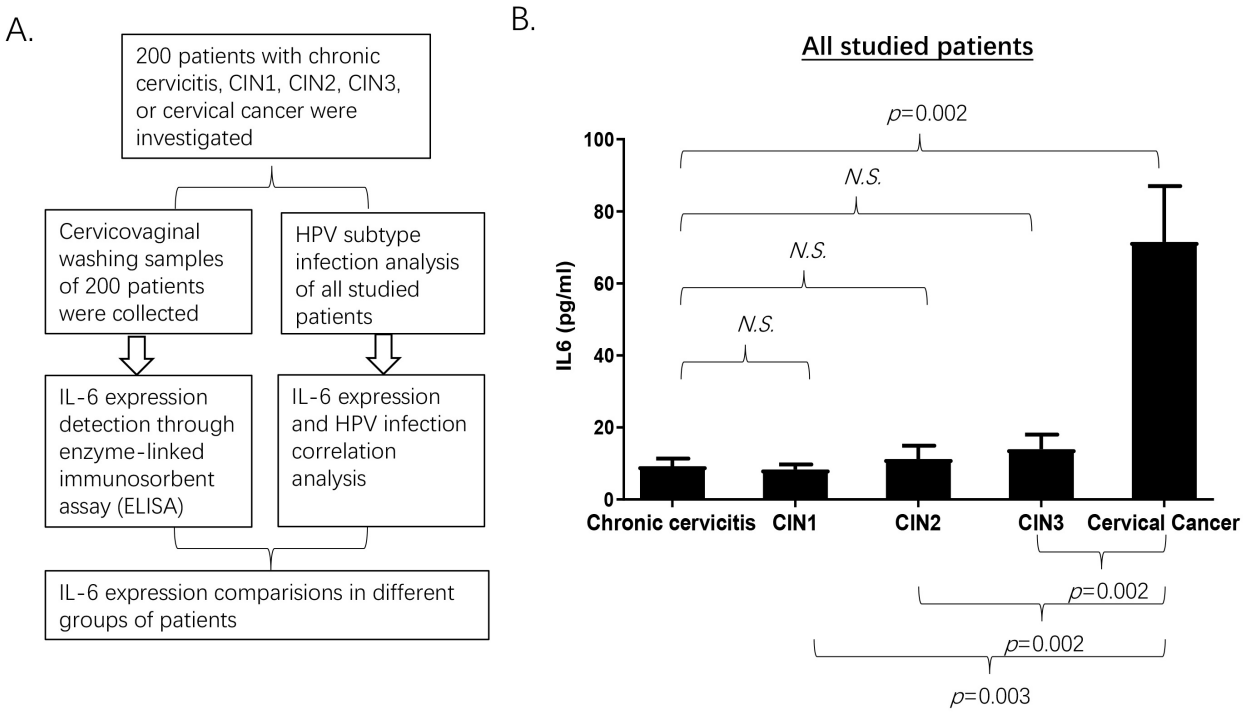


Fig. 1. Study outline and IL-6 expression comparisons between different groups of patients. (A) Outline of the present study. (B) IL-6 expression comparisons of in chronic cervicitis, CIN1, 2, 3, and cervical cancer. HPV, Human Papilloma Virus; CIN, cervical intraepithelial neoplasia; IL-6, interleukin-6; N.S., No significance.

biomarker that can be used to reasonably make the decision on which CIN2 patients can choose conservative treatment and which CIN2 patients can choose surgical treatment. Because of the link between IL-6 and oncogenic progression has been widely reported in a variety of malignancies, so in this present study, we measured the expression levels of interleukin-6 (IL-6) in cervicovaginal washing samples of 200 patients with CINs and cervical cancer. We aimed to analyze the expressions of IL-6 in patients with chronic cervicitis, different grades of CINs and cervical cancer to reveal its role in the diagnosis and treatment decision for patients with CINs.

2. Material and Methods

2.1 Patients

200 patients aged from 18 to 50 years old at The First Center Hospital of Tianjin, between March 2020 and March 2021, were prospectively investigated (Fig. 1A). Cervicovaginal washing samples of these patients were collected during colposcopy examination in the outpatient clinic. The inclusion criteria of the study are as follows: (1) patients were with HPV infection or (and) cervical cytology abnormality, and been diagnosed as chronic cervicitis, CIN1, CIN2, CIN3, or cervical cancer; (2) no history of interferon treatment and the other antiviral treatments at the time of diagnosis; (3) informed consent for the study were collected from all studied patients. Exclusion criteria of the study were as follows: (1) history of physical cervical therapy, such as local laser or microwave; (2) previous history of cervical surgery, such as cold knife conization or loop electrosurgical excision procedure (LEEP); (3) patients with any immunodeficiency diseases, such as systematic lupus erythematosus (SLE) and autoimmune diseases; (4) patients diagnosed with other malignant tumors; (5) complicated with serious chronic diseases, such as heart, liver, kidney, and hematopoietic disorders; (6) patients were pregnant or breastfeeding. The median age of the studied patients was 40 years (ranging from 18 to 50). The investigated patients were composed of 16% of chronic cervicitis (n = 32), 38% of CIN1 (n = 76), 23% of CIN2 (n = 46), 16.5% of CIN3 (n = 33), and 6.5% of cervical cancer (n = 13). This study was approved by the medical ethical committee board of Tianjin First Central Hospital.

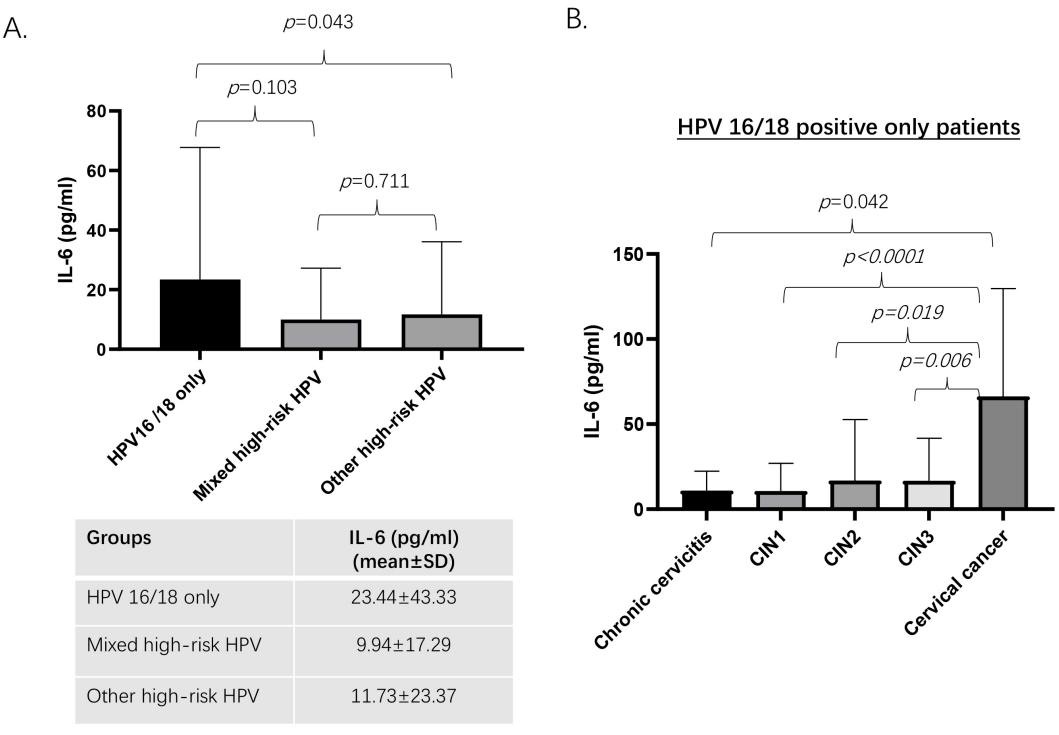
2.2 Sample Collection

Cervicovaginal washing samples of all studied individuals were collected. Briefly, patients were laid in a supine position in a gynecological examination table before performing colposcopy. Cervicovaginal washings were performed by flushing the cervix with 5 mL of 0.9% normal saline using a syringe (Catalog No.: DB-ZSQ-001, Shandong, China). Then, the fluid was collected and centrifuged for 5 min at 3000 rpm, room temperature, and the supernatants were collected and stored in aliquots at –20 °C until further assay.

Table 1. Patients' clinical and demographic characteristics at baseline.

Characteristics	Patients (n = 200)	Range
Age (years)	40.54 ± 11.14	18–50
Weight (kg)	60.80 ± 10.72	39–130
Height (cm)	162.42 ± 5.39	150–176
Marriage status (yes/no)	163/37	N/A
Pregnancy and childbirth history (G >3/G ≤3)	159/41	N/A
Pregnancy and childbirth history (P ≥1/P = 0)	153/47	N/A
HPV infection (yes/no)	192/8	N/A
HPV infection (over 6 months or/not)	54/146	N/A
Delivery method (Vaginal/C-section)	56/128	N/A
Vaginitis (yes/no)	6/194	N/A
Disease stage		
Inflammation (n (%))	32 (16.00%)	N/A
CIN1 (n (%))	76 (38.00%)	N/A
CIN2 (n (%))	46 (23.00%)	N/A
CIN3 (n (%))	33 (16.50%)	N/A
Cervical Cancer (n, %)	13 (6.50%)	N/A

HPV, human papillomavirus; CIN, cervical intraepithelial neoplasia; N/A, not applicable.
G, gestation; P, parturition.



Groups	IL-6 (pg/ml) (mean±SD)
HPV 16/18 only	23.44±43.33
Mixed high-risk HPV	9.94±17.29
Other high-risk HPV	11.73±23.37

Fig. 2. Post Hoc analysis of IL-6 expression in patients with HPV sub-types infection. (A) IL-6 expression comparisons in patients with different types of HPV infection. (B) The expression of IL-6 in different groups of patients with high risk HPV16/18 infection only. HPV, Human Papilloma Virus; CIN, cervical intraepithelial neoplasia; SD, standard deviation.; High-Risk HPV sub-types: HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 67, 68, 73, 82; Low-Risk HPV sub-types: HPV6, 11, 40, 42, 43, 44, 54, 55, 57, 81.

2.3 Enzyme-Linked Immunosorbent Assay (ELISA)

Cervicovaginal washing samples of patients with chronic cervicitis, CINs, and cervical cancer were thawed at room temperature for analysis. IL-6 expression levels were measured by enzyme-linked immunosorbent assay (ELISA), using a Human IL-6 Pre-coated ELISA kit (1110603, Dakewe Biotech Co., Ltd., Shenzhen, Guangdong, China) according to the operation instructions. A

Table 2. Details of patients' HPV subtype infections.

Disease stage	High-risk HPV	Low-risk HPV	Mixed with high and low risk HPV	Negative
Inflammation (n (%)) (32 (16.00%))	30 (93.75%)	0 (0.00%)	0 (0.00%)	2 (6.25%)
CIN1 (n (%)) (76 (38.00%))	70 (92.11%)	0 (0.00%)	2 (2.63%)	4 (5.26%)
CIN2 (n (%)) (46 (23.00%))	42 (91.30%)	0 (0.00%)	3 (6.52%)	1 (2.18%)
CIN3 (n (%)) (33 (16.50%))	32 (96.97%)	0 (0.00%)	0 (0.00%)	1 (3.03%)
Cervical Cancer (n (%)) (13 (6.50%))	13 (100.00%)	0 (0.00%)	0 (0.00%)	0 (0.00%)
Total (200 cases)	187 (93.5%)	0 (0.00%)	5 (2.50%)	8 (4.00%)

HPV, human papillomavirus; CIN, cervical intraepithelial neoplasia;
High-Risk HPV subtypes: HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 67, 68, 73, 82;
Low-Risk HPV subtypes: HPV6, 11, 40, 42, 43, 44, 54, 55, 57, 81.

Table 3. HPV subtype comparisons of patients with CINs and cervical cancer.

Disease stage	High-risk HPV	Mixed with high- and low-risk HPV	Negative	*p*-value
CIN1 (n)	70	2	4	0.4216
CIN2 (n)	42	3	1	
CIN2 (n)	42	3	1	0.3208
CIN3 (n)	32	0	1	
CIN1 (n)	70	2	4	0.5552
CIN3 (n)	32	0	1	
CIN1 (n)	70	2	4	0.5768
Cervical Cancer (n)	13	0	0	
CIN2 (n)	42	3	1	0.5454
Cervical Cancer (n)	13	0	0	
CIN3 (n)	32	0	1	0.7861
Cervical Cancer (n)	13	0	0	

HPV, human papillomavirus; CIN, cervical intraepithelial neoplasia;
High-Risk HPV subtypes: HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 67, 68, 73, 82;
Low-Risk HPV subtypes: HPV6, 11, 40, 42, 43, 44, 54, 55, 57, 81.

standard curve was made according to the manufacturer's instructions, and samples concentrations were calculated through ELISA reader (SpectraMax 190, Sunnyvale, CA, USA).

2.4 Statistical Analysis

Statistical analysis was performed using GraphPad 8.0 (GraphPad Software, Inc., San Diego, CA, USA) for Windows. Data were analyzed using *t*-test and Chi-square test for group comparisons. $p < 0.05$ was considered to be significantly different.

3. Results

3.1 Patients' Clinical and Demographic Characteristics

Basic clinical profiles of studied patients, including age, weight, height, marriage status, pregnancy, childbirth history, HPV infection, delivery method, pathology, HPV infection, and proportion of disease stages are shown in Table 1. The age range of studied patients was 18–50 years. There were 13 (6.50%, 13/200) patients with invasive cervical cancer, 33 (16.50%, 33/200) patients with CIN3, 46 (23%, 46/200) patients with CIN2, 76 (38%, 76/200) patients with CIN1, and 32 (16%, 32/200) control subjects with chronic cervicitis. Among the 200 patients, 192 patients were HPV-positive, from which187 presented high-risk HPV infection and 5 were high- and low-risk HPV-mixed infection, and lastly, 8 patients were HPV-negative.

3.2 Vaginal IL-6 Expression Is Significantly Higher in Patients with Cervical Cancer by Comparison with Patients with CINs and Chronic Cervicitis

The expression levels of IL-6 in the vaginal lavage gradually increased in patients with CIN1, 2, 3, and cervical cancer. IL-6 expression in patients with cervical cancer was significantly higher than that in patients with CINs ($p = 0.003$, $p = 0.002$ and $p = 0.002$, respectively) (Fig. 1B), which indicated that there may be more IL-6 secretion in tumor tissues than CINs samples, and the molecular mechanisms of this phenomenon is needed to be further studied. These results also suggest that cervical cancer cells and cervical cells in CINs patients may carry different proliferation and differentiation activities, which need to be further investigated. There was no significant difference in IL-6 expression between different grades of CIN groups.

Table 4. Detailed information of the infection of HPV16/18 and other high-risk HPV types in different groups of patients.

Disease stage (n (%))	HPV16/18 positive only (n (%))	Other high-risk HPV types (n (%))	HPV16/18 mixed with other high-risk HPV types (n (%))
Chronic cervicitis (30 (16.04%))	8 (26.67%)	18 (60.00%)	4 (13.33%)
CIN1 (70 (37.43%))	24 (34.29%)	35 (50.00%)	11 (15.71%)
CIN2 (42 (22.46%))	19 (45.24%)	14 (33.33%)	9 (21.43%)
CIN3 (32 (17.11%))	20 (62.50%)	8 (25.00%)	4 (12.50%)
Cervical Cancer (13 (6.95%))	7 (53.85%)	5 (38.46%)	1 (7.69%)
Total (187 cases)	78 (41.71%)	80 (42.78%)	29 (15.51%)

HPV, human papillomavirus; CIN, cervical intraepithelial neoplasia;
High-Risk HPV subtypes: HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 67, 68, 73, 82.

3.3 HPV Subtype Infection Analysis of Studied Patients and Comparisons of IL-6 Expression for Patients with Different HPV Subtype Infection

Of the 200 patients, 187 patients were with high-risk HPV infection, 5 patients were infected with mixed high- and low-risk HPV, there is no patients infected with low-risk HPV, and 8 patients were HPV-negative for infection (Table 2). Furthermore, there was no significant difference between CINs and cervical cancer patients based on above HPV grouping (Table 3). As HPV16/18 infection was reported as the most relevant high-risk factor for cervical cancer, we thus analyzed HPV16/18 infection features in different groups of patients (Table 4). Among the 187 high-risk HPV infection patients, 78 patients (41.71%) were infected with HPV16/18 only, 80 patients (42.78%) were infected with other high-risk HPV types, and 29 patients (15.51%) were infected with mixed HPV16/18 and other high-risk HPV types (Table 4). Interestingly, most patients with CIN2, 3, and cervical cancer were infected with HPV16/18 only, and patients with CIN1 and chronic cervicitis were mostly infected with other high-risk HPV types only (Table 5). There is significant difference in HPV subtypes infection when comparing patients with CIN3 with patients with chronic cervicitis and CIN1 (p = 0.012 and p = 0.024, respectively, Table 5). These results were consistent with the fact that HPV16/18 infection is more often to develop into high grade squamous intraepithelial lesion (HSIL) and cervical cancer.

Moreover, we further analyzed the IL-6 expression based on different types of HPV infection. We revealed that the IL-6 expression in HPV16/18 only infection group was significantly higher than that in other high-risk HPV groups (23.44 $\pm$ 43.33 pg/mL and 11.73 $\pm$ 23.37 pg/mL, respectively, p = 0.043) (Fig. 2A, Table 6). Among the 78 patients with HPV16/18 infection only, we found that IL-6 expression gradually increased in chronic cervicitis, CIN1, 2, 3, and cervical cancer (Fig. 2B, Table 6, **Supplementary Fig. 1**). This result indicated that IL-6 expression may be associated with HPV16/18 infection and its molecular mechanisms are still open questions.

4. Discussion

The treatment of patients with CIN2 is still controversial. Those patients were previously often being advised to receive cervical conization treatment right after diagnosis. However, conization treatment usually increases the obstetrical complications, especially preterm birth when these patients get pregnant. Particularly, there is an ascending premature birth incidence with the deepening of the excision depth and the increasing times of the conization treatments. A systematic review and meta-analysis revealed that cervical treatment significantly increased the risk of overall (<37 weeks; 10.7% *vs*. 5.4%), severe (<32–34 weeks; 3.5% *vs*. 1.4%), and extreme (<28–30 weeks; 1.0% *vs*. 0.3%) preterm birth by comparing with patients without cervical treatment [9]. The risk (<37 weeks) was higher for cold knife conization (3.53, 2.05 to 6.05), followed by excision not otherwise specified (1.70, 1.17 to 2.46), large loop excision of the transformation zone (1.60, 1.22 to 2.08) [9]. Another study shows that among 1808 women who underwent large loop excision of the transformation zone (LLETZ) treatment, a total of 321 women had a pregnancy, 9.1% delivered at <37 weeks of gestation, and 14.6% miscarried at <24 weeks of gestation [13].

Further, a recent meta-analysis showed a high regression rate of 66% for women with CIN2 without treatment but only follow-ups [12]. The current incidence rate of CIN2 is still relatively common, and recent clinical guidelines show that conservative and individualized management are suggested for up to 2 years for women younger than 25 years old [14]. However, there are a group of patients with CIN2 who are over 25 years old and are nulliparous. It requires a reasonable approach to make a treatment option for this group of patients to not miss the treatment opportunities and without increasing the risk of adverse obstetric outcomes.

Most HPV infections are transient, and persistent HPV infection is the main cause of CINs and cervical cancer. It has been reported that 90% of high-risk HPV infections and 75% of most low-grade intraepithelial lesions can be eliminated naturally [15,16]. HPV elimination is induced by innate immune responses, and the immune system plays an important role in this process [17,18]. Presentation of HPV

Table 5. Comparison of the infection of HPV16/18 and other high-risk HPV types of different groups of patients.

Disease stage	HPV16/18 positive only (n)	Other high-risk HPV types (n)	16/18 mixed with other high-risk HPV types (n)	*p*-value
Chronic cervicitis	8	18	4	0.6516
CIN1	24	35	11	
Chronic cervicitis	8	18	4	0.0803
CIN2	19	14	9	
Chronic cervicitis	8	18	4	0.0115
CIN3	20	8	4	
Chronic cervicitis	8	18	4	0.2281
Cervical Cancer	7	5	1	
CIN1	24	35	11	0.2269
CIN2	19	14	9	
CIN2	19	14	9	0.3206
CIN3	20	8	4	
CIN1	24	35	11	0.0240
CIN3	20	8	4	
CIN1	24	35	11	0.3840
Cervical Cancer	7	5	1	
CIN2	19	14	9	0.5322
Cervical Cancer	7	5	1	
CIN3	20	8	4	0.6416
Cervical Cancer	7	5	1	

HPV, human papillomavirus; CIN, cervical intraepithelial neoplasia;
High-Risk HPV subtypes: HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 67, 68, 73, 82.

Table 6. Comparison of IL-6 expression in different groups of patients with different high-risk HPV subtype infection.

Disease stage	HPV16/18 positive only (n)	Vaginal IL-6 expression level (ng/mL, mean ± SD)	Other high-risk HPV types only (n)	Vaginal IL-6 expression level (ng/mL, mean ± SD)	HPV16/18 mixed with other high-risk HPV types (n)	Vaginal IL-6 expression level (ng/mL, mean ± SD)
Chronic cervicitis	8	10.89 ± 11.47	18	8.21 ± 9.58	4	14.4 ± 19.4
CIN1	24	10.7 ± 16.3	35	6.7 ± 10.4	11	7.9 ± 9.5
CIN2	19	16.8 ± 35.9	14	22.2 ± 66.0	9	3.8 ± 5.4
CIN3	20	16.7 ± 25.0	8	12.4 ± 25.8	4	6.7 ± 6.2
Cervical Cancer	7	66.3 ± 63.3	5	75.8 ± 56.5	1	86.6

HPV, human papillomavirus; CIN, cervical intraepithelial neoplasia; SD, standard deviation.
High-Risk HPV subtypes: HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 67, 68, 73, 82.

antigens by antigen presenting cells (APCs) induces adaptive immunity through the actions of cluster of differentiation 4 (CD4+) and cluster of differentiation 4 (CD8+ T) cells [19]. According to their phenotypes, they are divided into T helper 1 (Th1) and Th2 cells. It has been observed that HPV infection altered the Th1/Th2 balance, differentiated Th0 cells into Th2 cells, resulting in relative dominance of Th2 cells [20,21]. It has also been shown that there are reduced IL-2 (i.e., by Th1) production, enhanced IL-4 (by Th2), and IL-10 (by Th2) production in CIN patients [19]. Hence, more cytokines are produced by Th2 cells, such as IL-4, IL-5, IL-6, IL-8, and IL-10. Furthermore, IL-6 can promote the production of vascular endothelial growth factor and promote the occurrence of cancer. The link between IL-6 and oncogenic progression has been widely reported in a variety of malignancies, such as cervical cancer [22,23], head and neck cancer [24], hepatocellular carcinoma [25], and gastric cancer [26]. IL-6 acts on target cells via a specific IL-6 receptor (IL-6R), and induces the dimerization of a second receptor subunit-gp130. IL-6 binds to the IL-6 receptor and activates the signal transducer and activator of transcription 3 (STAT3) pathway, which leads to epithelial-mesenchymal plasticity (EMP) in cervical cancer [27].

It has been shown that the single-nucleotide polymorphisms of IL-6 might be associated with high cervical cancer risk. Shaswati M *et al.*'s [28] study suggests that polymorphism rs1800795 and rs1800797 of the IL-6 gene play a significant role in cervical cancer susceptibility in Bangladeshi Women. Vitkauskaite A *et al.* [22] also examined the significant association of the C allele and CC genotype of IL-6 1800795 gene with cervical cancer in the Lithuanian population. Accordingly, genotype CC of IL-6 rs1800795 has a significant association with HPV infection [22]. Furthermore, the serum IL-6 levels in the cervical cancer group were found significantly higher than that

in the control group, and high serum IL-6 levels are associated with adverse prognosis in patients with cervical cancer and could be a prognosis indicator for cervical cancer patients [23]. Another two studies evaluated the association between cervical cancer prognosis and IL-6 expression in tumor tissue of cervical cancer patients [29,30]. They both showed a better prognosis in cervical cancer patients with lower IL-6 expression [29,30]. This fully demonstrates the correlation between IL-6 and cervical cancer, and is also consistent with the results of the present study. Meanwhile, it has found that the concentration of IL-6 in the cervical region is higher than that in the serum [31]. Therefore, we speculated that studying IL-6 in the cervical region is more sensitive and specific than that in the peripheral blood.

As early as in 1998, Tjiong *et al*. [32] showed that IL-6 expression is locally increased in cervicovaginal secretions in patients with cervical carcinoma compared with patients with CINs and healthy controls. The cytosolic IL-6 levels in malignant and non-malignant cervical tissue had also been examined and it was reported that cytosolic IL-6 expression in malignant samples was significantly higher than that of non-malignant samples [33]. Moreover, Naik *et al*. [34] observed increased levels of IL-6 in cervicovaginal secretions in patients with pre-malignant and malignant cervical lesions. Besides, the cervical levels of IL-6 expression in the patients with CIN were higher than that of the healthy control group [35]. However, the expression levels of IL-6 in different grade of CINs have not been reported. In the present study, we first measured the expression levels of IL-6 in the cervicovaginal washing of patients with CIN1, 2, 3, cervical cancer, and chronic cervicitis. We revealed that the expression levels of cervicovaginal IL-6 in different grades of CIN to cervical cancer have a gradual uptrend. This result suggests that IL-6 may be involve in the whole process of cervical intraepithelial neoplasia developing into cervical cancer. Interestingly, we found that IL-6 was significantly higher in HPV16/18 group than in other high-risk HPV types groups, which coincides with HPV16/18 having the strongest effect on cervical cancer.

5. Conclusions

We propose that the expression levels of cervicovaginal IL-6 can be measured as a reference biomarker for the treatment of CIN2, which is that in patients with CIN2 that presents high concentration of cervicovaginal IL-6 should be actively treated by surgery. On the contrary, the low expression levels of vaginal IL-6 suggests that the degree of CIN may be reduced and the risk of disease progression may be relatively low, which can be followed up conservatively. Although there was no significant difference between CIN groups of different grades, which may be due to the small size of patients, and larger population studies are needed to confirm these results in the future.

Availability of Data and Materials

Raw data can be provided by the corresponding authors upon reasonable requests.

Author Contributions

XL, FL, LZhao and QQ designed the research study. JL, YG, LZhang, MZ and MJ collected specimens. XZ provided help and advice on the ELISA experiments. XL and FL analyzed and explained the data, drafted the manuscript. FL and YW conducted data statistical analysis and critically revised manuscripts containing important knowledge content. QQ supervised the entire process. All authors contributed to editorial changes in the manuscript. All authors read and approved the final manuscript. All authors have participated sufficiently in the work and agreed to be accountable for all aspects of the work.

Ethics Approval and Consent to Participate

All subjects gave their informed consent for inclusion before they participated in the study. The study was conducted in accordance with the Declaration of Helsinki, and the protocol was approved by the Ethics Committee of Tianjin First Central Hospital (approval number: 2023DZX24).

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Conflict of Interest

The authors declare no conflict of interest.

Supplementary Material

Supplementary material associated with this article can be found, in the online version, at https://doi.org/10.31083/j.ceog5011239.

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