The Risk of Human Papillomavirus Infection for Spontaneous Abortion, Spontaneous Preterm Birth, and Pregnancy Rate of Assisted Reproductive Technologies: A Systematic Review and Meta-Analysis

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Keywords

Human papillomavirus · Spontaneous abortion · Spontaneous preterm birth · Assisted reproductive technologies · Meta-analysis

Abstract

Aims: To estimate the impact of human papillomavirus (HPV) infection on spontaneous abortion, spontaneous preterm birth (sPTB), pregnancy rate of females undergoing assisted reproductive technologies (ART), and spontaneous abortion of ART pregnancy. **Methods:** PubMed, Medline, Embase, and the Cochrane Library were searched until December 16, 2016. The OR or relative risk (RR) with its corresponding 95% CI were selected as effect size. Subgroup analysis of HPV genotype infection (high-risk HPV [HR-HPV] or indiscriminate genotype) was performed. Statistical analysis was conducted using STATA 12.0. **Results:** Eighteen studies were included. Eight studies revealed no significant association between HPV infection and spontaneous abortion (OR 1.40, 95% CI 0.56–3.50). However, subgroup analysis showed indiscriminate genotype HPV infection increased the ratio of sponta-

neous abortion with OR of 2.24 (95% CI 1.37–3.65), while HR-HPV infection had no significant effect (OR 0.65, 95% CI 0.21–1.98). The results indicated that HR-HPV infection was a risk for sPTB with a pooled OR of 2.84 (95% CI 1.95–4.14). HPV infection was found to be independent of the ART-based clinical pregnancy rate (RR 1.04, 95% CI 0.64–1.70) and spontaneous abortion of ART pregnancy (RR 1.47, 95% CI 0.86–2.50). **Conclusions:** Indiscriminate HPV genotype infection can increase the risk of spontaneous abortion and HR-HPV infection was a risk factor for sPTB. However, there was not enough evidence to indicate the association between HPV infection and pregnancy rate of ART, and spontaneous abortion of ART pregnancy. Different genotypes of HPV infection may play a discrepant role in adverse pregnancy outcomes.

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Introduction

Human papillomavirus (HPV) is one of the most prevalent sexually transmitted viral infections in men and women worldwide [1]. The existence of more than 180

HPV genotypes has been reported [2]. According to the basis of oncogenic potential, HPV can be divided into 2 different groups-high-risk HPV (HR-HPV) and low-risk HPV (LR-HPV). Generally, the HR-HPV included HPV16, HPV18, HPV31, HPV33, HPV35, HPV39, HPV45, HPV51, HPV52, HPV56, HPV58, HPV59, HPV66, and HPV68 [3]. It is well established that infections with HR-HPV can lead to cervical cancer. Moreover, recent evidence has shown that HPV infection is associated with the risk of colorectal, head, and oropharyngeal carcinomas [4, 5].

To date, a succession of studies has been published to show the association between HPV infection and adverse pregnancy outcomes. A study reported that HPV was positive in 34 (24.4%) of 139 cervical samples from women who experienced spontaneous abortion. Additionally, the authors reported that 27.3% of HPV-positive women were found to have repetitive abortions, whereas this rate was only 17.4% in HPV-negative women [6]. A systematic review showed that the prevalence of HPV was significantly higher in the cervix of women who had spontaneous abortions than those who had normal full-term pregnancies (24.5 vs. 17.5%); this trend demonstrated a probable association between spontaneous abortion and the infection of HPV [7].

Recent evidence suggests that HPV infection may affect fertility and the clinical pregnancy rate of assisted reproductive technologies (ART). For instance, HPV infection can affect the parameters of sperm quality, and may play a crucial role in male fertility [8, 9]. However, there are controversial results whether HPV infection may reduce the clinical pregnancy and live birth rates in women undergoing ART [10–12]. Spandorfer et al. [10] reported that HPV-positive women had a decreased pregnancy rate (4 of 17, 23.5%) as compared with HPV-negative women (51 of 89, 57.3%; $p < 0.02$). Spandorfer et al. [10] reported that patients with HPV infection were less likely to become pregnant after undergoing in vitro fertilization (IVF), and clinical pregnancy rate was lower in the group of patients with HPV positive (HPV positive vs. negative: 23.5 vs. 51.7%). However, a prospective study including 362 patients indicated no significant difference in pregnancy rates between HPV-positive and HPV-negative women undergoing IVF treatment [12].

Considering the relationship of HPV infection and spontaneous abortion, spontaneous preterm birth (sPTB) and ART outcomes could have far-reaching implications in the medical care of pregnant and infertile women. Thus, we conducted this meta-analysis with a

summary of all published estimates to evaluate the effect of HPV infection in spontaneous abortion, sPTB, and clinical pregnancy rates of females undergoing ART.

Methods

This meta-analysis is conducted in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses guidelines [13].

Literature Search

We searched the PubMed, Medline, Embase, and Cochrane Library database from the earliest date available up until December 16, 2016. We used the following keywords separately and in combinations: “human papillomavirus,” “HPV,” “miscarriage,” “abortion,” “pregnancy,” “preterm birth,” “PROM,” “premature rupture of the membranes,” “in vitro fertilization,” “IVF,” “assisted reproductive technology,” “ART,” and “ICSI.” Manual searches of the reference lists of review articles and studies included in the final publication selection were also conducted.

Inclusion and Exclusion Criteria

Studies were screened for eligibility if they met the following criteria (1) reported spontaneous abortion, sPTB, and ART outcomes in patients with HPV infection; (2) reported corresponding effect estimates or sufficient data for their calculation; (3) cohort study, case-control study, or cross-sectional study design; and (4) HPV DNA in cervical or placental samples was detected to diagnose the infection by polymerase chain reaction or other methods. Studies were excluded for the following reasons: (1) if they were review articles, editorials, opinions, or case reports; (2) if they did not include effective control groups to calculate effect estimates; and (3) if cervical cytology was used as a surrogate for HPV infection.

Data Selection and Extraction

Citations were merged using Endnote (version X7) to facilitate management and data extraction. Two authors independently evaluated all retrieved articles by title, abstract, and full text according to the above inclusion criteria. Any disagreement was resolved by consensus. Data were independently extracted from each eligible study and included first author, publication year, country of origin, study design, HPV detection method, HPV genotype, number of HPV infected participants with or without spontaneous abortion, sPTB and pregnancy after ART, corresponding relative risk (RRs) or ORs, and 95% CI values. When data were reported from overlapping study samples (e.g., multiple publications from the same study), the most recent and comprehensive report was considered.

Quality Assessment

The quality of case control and cohort studies was assessed by using the Newcastle-Ottawa Scale [14, 15]. In this scale, studies are scored across 3 categories: selection of subjects, comparability of study groups, and assessment of outcome/exposure.

Fig. 1. Flow diagram of the studies identified in the meta-analysis.

The rating system was used to indicate the quality of a study, with a maximum score of 9. Studies were graded on an ordinal scoring scale, with higher scores representing studies of higher quality.

Statistical Analysis

The ORs or RRs with their corresponding 95% CIs were selected as the effect size to assess the risk of spontaneous abortion, sPTB, and pregnancy after ART in HPV-infected subjects. Cohort studies and case-control studies were included in the pooled results, while cross-sectional studies were included only in the sensitivity analysis. Inter-study heterogeneity was estimated by the I^2 statistic, and significant heterogeneity was defined as $I^2 \geq 50\%$. Pooled results and corresponding 95% CIs were calculated with a fixed effects model (Mantel and Haenszel method) when heterogeneity was not significant ($I^2 < 50\%$); otherwise, a random-effects model (DerSimonian and Laird method) was applied. Forest plots were constructed for visual display of pooled results if necessary. Subgroup analyses were performed according to study design (cohort study or case-control study) and HPV genotype infection pattern (HR-HPV alone or indiscriminate genotype [HR/LR-HPV]). A meta-regression analysis was also performed to explore inter-study heterogeneity. Moreover, sensitivity analyses were used to evaluate whether any single study dominated the results of the meta-analyses. Finally, publication bias was assessed by the visual inspection of funnel plots and Egger's linear regression [16]. Statistical analyses were conducted using STATA 12.0 (Stata Corp LP, College Station, TX, USA).

Results

Description of Included Studies

The systematic literature searches identified 2070 potentially relevant studies. Most ineligible studies were excluded based on the information in the title or abstract. The selection process is shown in Figure 1. Eighteen studies [10–12, 17–31] were included in our analysis. The main characteristics of included studies are described in Table 1. Twelve studies [12, 19–22, 24–30] reported the impact of HPV infection on risk of spontaneous abortion. Five studies [17, 18, 21, 23, 31] reported the risk of sPTB and 7 studies [10–12, 19–22] reported the risk of clinical pregnancy rate of ART. Eight studies [12, 17, 18, 20, 21, 23, 27, 29] reported HR-HPV infection alone and the other 10 studies [10, 11, 19, 22, 24–26, 28, 30, 31] reported indiscriminate genotype infection (HR/LR-HPV).

Association between HPV Infection and Spontaneous Abortion

Twelve studies [12, 19–22, 24–30] including 5 cohort studies, 3 case-control studies, and 4 cross-sectional studies, with a total of 3,007 participants, reported the impact of HPV infection on spontaneous abortion. Pooled results

Table 1. Basic characteristics of the eligible studies

Author, reference, year	Country	Study design	HPV detection method	HPV genotype	HPV detection tissue	Spontaneous abortion	Spontaneous preterm birth	Pregnancy of ART	Time of abortion	NOS score
Conde-Ferraz et al. [24], 2013	Mexico	Case control	PCR	at least 3 HR-HPV and 2 LR-HPV: including 6, 11, 16, 18, 58	Cervical	Yes			Prior to 20 weeks	7
Hermanot et al. [25], 1997	USA	Cross-sectional	PCR	at least 3 HR-HPV and 1 LR-HPV: including 6, 11, 16, 18	Chorionic villi	Yes			First trimester	NA
Skoczynski et al. [26], 2011	Poland	Cross-sectional	PCR	at least 33 HPV: including 16, 18	Products of conception	Yes			Median 10 weeks	NA
Ticconi et al. [27], 2013	Italy	Case control	HC2 and PCR	13 HR-HPV: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68	Cervical	Yes			Prior to 20 weeks	7
Bennani et al. [28], 2012	Maroc	Cross-sectional	PCR	NA	Cervical	Yes			NA	NA
Joshi et al. [29], 2014	India	Cross-sectional	HC2 and TS-MPG	13 HR-HPV/pHR-HPV: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68	Cervical	Yes			NA	NA
Srinivas et al. [30], 2006	USA	Case control	PCR	2 HR-HPV and 2 LR-HPV: 6, 1, 16, 18	Placental tissues	Yes			Second trimester	6
Perino et al. [19], 2011 ^a	Italy	Prospective cohort	INNO-LiPA Genotyping system and nested PCR	15 HR-HPV, 7 LR-HPV and 6 UR-HPV: 6, 11, 16, 18, 26, 31, 33, 35, 39, 40, 43, 44, 45, 51, 52, 53, 54, 56, 58, 59, 66, 68, 69, 70, 71, 73, 74, 82	Cervical	Yes		Yes	NA	6
Tanaka et al. [20], 2000	Japan	Retrospective cohort		1 HR-HPV: 16	Cervical	Yes		Yes	NA	6
Yang et al. [21], 2013	China	Retrospective cohort	HC2	13 HR-HPV: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68	Cervical	Yes	Yes	Yes	not later than the 23 weeks	8
Oborna et al. [12] (2016)	Czech Republic	Prospective cohort	Roche Cobas 4800	14 HR-HPV: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68	Cervical	Yes		Yes	NA	6
Comar et al. [22], 2013	Italy	Retrospective cohort	TS-MPG	19 HR-HPV/pHR-HPV and 2 LR-HPV: 6, 11, 16, 18, 26, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 68a, 68b, 70, 73, 82	Cervical	Yes		Yes	NA	7
Spandorfer et al. [10], 2006 ^b	USA	Prospective cohort	HC2	13 HR-HPV and 5 LR-HPV: 6, 11, 16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 67, 68	Cervical			Yes		7
Depuydt et al. [11], 2015	Belgium	Prospective cohort	qPCR	13 HR-HPV and 5 LR-HPV: 6, 11, 16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 67, 68	Cervical			Yes		7

Table 1. (continued)

Author, reference, year	Country	Study design	HPV detection method	HPV genotype	HPV detection tissue	Spontaneous abortion	Spontaneous preterm birth	Pregnancy of ART	Time of abortion	NOS score
Zuo et al. [17], 2011	USA	Retrospective cohort	HC2	13 HR-HPV: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68	Cervical	Yes	Yes			6
Cho et al. [18], 2013	Korea	Cross-sectional	HC2	13 HR-HPV: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68	Cervical	Yes	Yes			NA
McDonnold et al. [23], 2014	USA	Retrospective cohort	NA	13 HR-HPV: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68	Cervical	Yes	Yes			6
Gomez et al. [31], 2008	USA	Case control	PCR	2 HR-HPV and 2 LR-HPV: 6, 11, 16, 18	Placental tissues	Yes	Yes			7

^a The author excluded the participants infected with any of human immunodeficiency virus, chlamydia trachomatis, neisseria gonorrhoeae, herpes simplex, or treponema pallidum; ^b the author excluded the participants infected with any of human immunodeficiency virus, chlamydia, gonorrhea, or syphilis; HPV, human papillomavirus; HR-HPV, high risk HPV; LR-HPV, low risk HPV; pHR-HPV, probable high risk HPV; PCR, polymerase chain reaction; qPCR, quantitative polymerase chain reaction; HC2, Hybrid Capture 2 Modular System; TS-MPG, type-specific E7 PCR bead-based multiplex genotyping assay; NOS, Newcastle-Ottawa Scale; NA, not available.

from 8 cohort/case-control studies suggested that infection with HPV probably increased the ratio of spontaneous abortion by 1.40 (95% CI 0.56–3.50; $I^2 = 79.4\%$; Fig. 2). The pooled ORs of cohort [12, 19–22] and case-control studies [24, 27, 30] were 1.47 (95% CI 0.86–2.50; $I^2 = 8.3\%$) and 1.02 (95% CI 0.21–5.02; $I^2 = 92.4\%$) respectively (Table 2). When only HR-HPV infection was considered, the pooled OR from 3 cohort studies and one case-control study involving a total of 456 participants was 0.65 (95% CI 0.21–1.98, $I^2 = 64.7\%$). The results of HR-HPV infection alone and all HPV genotypes' infection indicated that there was no significant association between HPV infection and spontaneous abortion. However, when removed from the HR-HPV infection studies, the pooled results based on 4 cohort/case control studies indicated that indiscriminate genotype HPV infection increased the ratio of spontaneous abortion with OR of 2.24 (95% CI 1.37–3.65; $I^2 = 0.0\%$; Table 2).

Association between HPV Infection and sPTB

Five studies [12, 17, 18, 21, 23], including 3 cohort studies, one case-control study, and one cross-sectional study, with a total of 1,719 participants, reported the impact of cervical HPV infection on the risk of sPTB. All 5 studies studied HR-HPV infection. The pooled results of cohort/case-control studies indicated that HR-HPV infection was a risk factor for sPTB, with a pooled OR of 2.84 (95% CI 1.95–4.14; $I^2 = 23.5\%$; Fig. 3). Meanwhile, the pooled result of cohort studies showed a similar RR of 2.37 (95% CI 1.68–3.35; $I^2 = 0.0\%$; Table 2).

Association between HPV Infection and Clinical Pregnancy Rate of ART, Spontaneous Abortion of ART Pregnancy

Seven cohort studies [10–12, 19–22], including 4 prospective cohort studies and 3 retrospective cohort studies, with a total of 1,390 participants, reported the impact of HPV infection on the risk of clinical pregnancy rate of ART. The pooled result of all 7 cohort studies indicated no significant association between HPV infection and clinical pregnancy rate of ART, with a pooled RR of 1.04 (95% CI 0.64–1.70; $I^2 = 74.3\%$; Fig. 4). Similar results were obtained upon stratified analyses of prospective cohort studies [10–12, 19] and retrospective cohort studies [20–22], with RRs being 1.30 (95% CI 0.61–2.80; $I^2 = 82.5\%$) and 0.82 (95% CI 0.37–1.81; $I^2 = 69.1\%$) respectively (Table 2). Of the 7 studies, 3 studies [12, 20, 21] reported infection with HR-HPV alone and the other 4 studies [10, 11, 19, 22] reported mixed-type infection with HR/LR-HPV, with pooled RRs of 1.66 (95% CI 0.29–9.63; $I^2 = 90.7\%$) and 0.85 (95% CI 0.66–1.09; $I^2 = 20.2\%$) respectively (Table 2). In addition,

Fig. 2. Forest plot of the association between human papillomavirus infection and risk of spontaneous abortion.

Table 2. Subgroup analysis of association between HPV infection and spontaneous abortion, spontaneous preterm birth and pregnancy rate of ART

	Study included	Number of participants	OR/RR	95% CI	Heterogeneity (I^2), %
<i>Spontaneous abortion</i>					
Study design					
Cohort/case control study	8	1,167	1.40	0.56–3.50	79.4
Cohort study	5	266	1.47	0.86–2.50	12.9
Case control study	3	901	1.02	0.21–5.02	92.4
Cohort/case control and Cross-sectional study	12	3,007	1.45	0.84–2.52	77.4
Genotype of HPV infection (only cohort and case control study included)					
Only HR-HPV included	4	711	0.65	0.21–1.98	64.7
Only HR/LR-HPV included	4	456	2.24	1.37–3.65	0.0
<i>Spontaneous preterm birth</i>					
Study design					
Cohort/case control study	4	1,408	2.84	1.95–4.14	23.5
Cohort study	3	1,348	2.37	1.68–3.35	0.0
Case control study	1	60	6.00	1.17–30.73	NA
<i>Pregnancy rate of ART</i>					
Study design					
Prospective/retrospective cohort study	7	1,390	1.04	0.64–1.70	74.3
Prospective cohort study	4	1,018	1.30	0.61–2.80	82.5
Retrospective cohort study	3	372	0.82	0.37–1.81	69.1
Genotype of HPV infection					
Only HR-HPV included	3	649	1.66	0.29–9.63	90.7
Only HR/LR-HPV included	4	741	0.85	0.66–1.09	20.2

HPV, human papillomavirus; HR-HPV, high risk HPV; LR-HPV, low risk HPV; NA, not available.

Fig. 3. Forest plot of the association between human papillomavirus infection and risk of spontaneous preterm birth.

Fig. 4. Forest plot of the association between human papillomavirus infection and risk of pregnancy rate of assisted reproductive technologies.

5 studies [12, 19–22] reported the risk of HPV infection on spontaneous abortion of ART pregnancy, with the pooled RR being 1.47 (95% CI 0.86–2.50; $I^2 = 12.9\%$).

Heterogeneity Analysis

The inter-study heterogeneity in the analysis of impact of HPV infection on spontaneous abortion and pregnancy rate of ART was explored by meta-regression analysis. The following four variables were included in the meta-regression analysis: (1) study design (cohort study vs. case-control study); (2) HPV genotype (HR-HPV infection alone vs. HR/LR-HPV infection); (3) HPV detection method (Hybrid Capture 2 Modular System vs. others);

and (4) number of participants (≥ 100 vs. < 100). The HPV genotype ($p = 0.043$) and HPV detection method ($p = 0.016$) had significant influence on heterogeneity of association between HPV infection and spontaneous abortion. HPV detection method ($p = 0.035$) had significant influence on heterogeneity of association between HPV infection and pregnancy rate of ART.

Sensitivity Analysis

We performed a sensitivity analysis to evaluate whether any single study dominated the results of the meta-analyses by sequentially removing individual eligible studies. We observed that no individual study altered the overall

significance of the ORs/RRs in the analyses of the impact of HPV infection on spontaneous abortion, sPTB, and outcome of ART. Additionally, we added 4 cross-sectional studies [25, 26, 28, 29] to the analysis of association between HPV infection and spontaneous abortion. The summary result of cohort, case-control, and cross-sectional studies was similar to the previous summary result of cohort/case-control studies (1.45 [95% CI 0.84–2.52; $I^2 = 77.4\%$] vs. 1.40 [95% CI 0.56–3.50; $I^2 = 79.4\%$]; Table 2).

Published Bias

The funnel plots for publication bias were symmetrical and Egger's test indicated no significant asymmetry was found in any of the analyses.

Discussion

To the best of our knowledge, this is the first systematic literature review and meta-analysis on the published evidence of the impact of HPV infection on spontaneous abortion, clinical pregnancy rate of female undergoing ART, and spontaneous abortion of ART pregnancy. Our results indicated that there was no significant association between HPV infection and spontaneous abortion (OR 1.40; 95% CI 0.56–3.50), clinical pregnancy rate of ART (RR 1.04; 95% CI 0.64–1.70), and spontaneous abortion of ART pregnancy (RR 1.47; 95% CI 0.86–2.50). While, indiscriminate genotype HPV infection increased the ratio of spontaneous abortion with pooled OR of 2.24 (95% CI 1.37–3.65), HPV infection could increase the risk of sPTB with pooled OR of 2.84 (95% CI 1.95–4.14).

Spontaneous abortion has been reportedly associated with several factors such as intrauterine infection, uterine malformations, fetal and placental anomalies, genetic factors, and environmental exposures [32, 33]. Srinivas et al. [30] reported that the infection rates for all cytomegalovirus, HPV, or adeno-associated virus-2 groups and for any bacteria of Group B *Streptococcus*, *Ureoplasma urealyticum*, or *Mycoplasma hominis* were 44 and 19%, respectively, in the control group, whereas the infection rates for any viruses and for any bacteria were 79 and 27%, respectively, in the spontaneous case group. Although the authors did not report the co-infection rate for HPV and other viruses or bacteria, the results indicated that co-infection for HPV and other intrauterine infection were not rare in pregnant women. In the included studies for analysis of spontaneous abortion in our study, only Perino et al. [19] stated that all study participants tested negative for HIV, *Chlamydia trachoma-*

tis, *Neisseria gonorrhoeae*, herpes simplex, or *Treponema pallidum*. Additionally, the authors reported a significant association between HPV infection and spontaneous abortion (OR 4.19, 95% CI 1.14–15.45). As most original results included in our study lacked adjustment, some other intrauterine infection, genetic factors, and environmental exposures may have had confounding effects on the association between HPV infection and spontaneous abortion. Additional prospective studies that exclude the co-infection of HPV and other intrauterine infection in participants could be more accurate to uncover the association between HPV infection and adverse pregnancy outcomes.

HPV infection is common in the general population, including normal pregnant women. In normal full-term pregnancy, prevalence of HPV was reported to vary between 2.2 and 75% in the cervical tissue, with a summary estimate of 17.5% [7]. In placental and aborted tissues, the infection rate was slightly lower, with a summary estimate of 8.3% (95% CI 7.6–9.1). Correspondingly, the prevalence of HPV was significantly higher in the cervix and placental and aborted tissue of women who underwent spontaneous abortions [7]. Although the prevalence of HPV was significantly different in normal pregnancies and the spontaneous abortion group, our results based on 5 cohort studies and 3 case-control studies indicated that there was no significant association between HPV infection and spontaneous abortion (OR 1.40; 95% CI 0.56–3.50). When the included studies reported HR-HPV infection alone, the pooled OR was 0.65 (95% CI 0.21–1.98). However, the pooled OR of 4 cohort/case-control studies [19, 22, 24, 30] reported that the HR/LR-HPV infection indicated that HPV infection was a risk factor for spontaneous abortion with an OR of 2.24 (95% CI 1.37–3.65). As the high- or LR-HPV genotype was classified according to the influence of different neoplasms, the risk of these HPV genotypes for adverse pregnancy outcomes had not been well investigated. Those studies that only detected HR-HPV infection may reflect a decreased rate of HPV infection in the spontaneous abortion group. Further research is still required to clarify whether certain HR-HPV or LR-HPV genotypes increase miscarriage risk.

A previous meta-analysis reported that HPV infection was a risk factor for preterm birth, with an OR of 2.12 (95% CI 1.51–2.98) [34]. We observed that 2 of its included studies [31, 35] recruited a component of HPV non-infection but subjects with abnormal cervical cytology in the HPV infection group; this might influence the real association between HPV infection and preterm birth. After eliminating the disputable studies, our results indi-

cated that HR-HPV infection remained a risk for sPTB with a pooled OR of 2.84 (95% CI 1.95–4.14).

Sexually transmitted infections such as *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, and HPV may affect fertility or reproductive outcomes [36, 37]. Our meta-analysis results showed that there was no significant association between HPV infection and pregnancy rate of ART, with a pooled RR of 1.04 (95% CI 0.64–1.70). Meanwhile, the pooled results of 4 prospective cohort studies [10–12, 19] suggested that HPV infection may increase the risk of spontaneous abortion of ART pregnancy, but the RR was not statistically significant (RR 1.30; 95% CI 0.61–2.80). We observed that of 7 cohort studies [10–12, 19–22] involving a total of 1,390 participants that were included in the summary analysis, only 210 subjects were positive for HPV infection. Therefore, the results of association between HPV infection and ART outcome in this study should be interpreted with caution. In a prospective study [19], the authors detected HPV infection in both women and their male partners among couples who underwent ART cycles. The result showed that couples experienced an increased risk of pregnancy loss when HPV was positive in the male partner, as compared to noninfected patients (66.7 vs. 15%, $p < 0.01$); this indicated that HPV infection in the male partner could play a role that cannot be neglected in pregnancy loss among couples who are undergoing ART. Prospective cohort studies with larger sample sizes are required to clarify whether HPV infection in women and their male partners could alter the ART outcome.

It was reported that there are 2 different pathways in the natural history of HPV infections, namely, the infectious virion producing pathway and the clonal transforming pathway [38]. The infectious virion producing pathway can lead to subfertility or early abortion and it is infectious. The clonal transforming pathway can lead to cancer but is noninfectious. The 2 pathways of HPV infection can occur simultaneously at a single location (e.g., cervix) in patients; however, it is in separate cells at different physical locations within the epidermis or mucosa [39–41]. Oncogenic (HR) HPV types induce more rapid cell division arrest than LR-HPV or intermediate HR-HPV types. It takes the embryo longer to die after LR-HPV infection, which makes it possible to measure the spontaneous abortion [38]. When HR-HPV virions (e.g., HPV 16) infect the embryo, the cell division should arrest over 2 cell stage. HR-HPV (e.g., HPV 53) once transferred to the embryo will first replicate each time cell division occurs. Because of the less oncogenic character of LR-HPV, the embryo will not die from division arrest but only weeks later when the dividing cells (with LR-HPV

inside) differentiate into syncytiotrophoblast. Accumulation of HPV virions in the trophoblasts that are responsible for maintaining placental contact with maternal tissue and through which nutrient exchange occurs prompted the hypothesis that HPV-infected trophoblasts may have altered characteristics that may lead to a compromised gestation [38]. These may explain why when including LR-HPV in our analysis, the result indicated a significant association between HPV infection and spontaneous abortion with OR of 2.24 (95% CI 1.37–3.65), while when only HR-HPV was included, no significant association was found (OR 0.65, 95% CI 0.21–1.98). In order to assess the impact of the HPV infection for fertility, it is necessary to identify the origin of the detected HPV DNA (the infectious virion-producing pathway or the clonal-transforming pathway). Unfortunately, the HPV DNA tests used in the 18 studies included in our meta-analysis could not identify the origin of the detected HPV DNA. Further researches, which separate analysis the HR-HPV or LR-HPV and identify the origin of HPV, are still required to investigate the impact of HPV infection on adverse pregnancy outcomes.

Since 2006, 3 vaccines (bivalent, quadrivalent, and 9-valent) against HPV have been available. HPV vaccination programs have been implemented in many countries worldwide to prevent the high mortality from cervical cancer and to lower the incidence of dysplasia and genital warts [42]. Considering the high prevalence of HPV infection in spontaneous abortion, preterm birth, and male and female infertility, it remains uncertain whether such vaccination programs will improve the pregnancy outcome and infertility, and further investigation is required.

Some limitations of this meta-analysis should be addressed. First, there was significant heterogeneity among the included studies. Different participant inclusion and exclusion criteria, HPV-detection methods, and participants' ethnicities could have contributed to the heterogeneity. Although we performed subgroup analysis and meta-regression, the results did not identify all the heterogeneity among studies. Second, the genotypes of HPV detection were varied in the included studies; some of the studies only detected types 16 and 18, or 13 HR-HPV genotypes, whereas other studies reported both HR-HPV and LR-HPV infection. Because the influences of HR-HPV and LR-HPV infection on adverse pregnancy outcomes and infertility have not been well expounded, the varied HPV genotypes in our included studies could have had some as yet unidentified influence on the pooled results. Third, HPV infection may play diverse risk in different stages of pregnancy. We did not distinguish the

risk of HPV infection for spontaneous abortion in first, second, or third trimesters, since the time of abortion was reported inadequately in original studies. Fourth, since most studies included in our analysis did not report other adverse pregnancy outcomes risk factors, such as age of mother, ethnic origin, psychological state of the mother, smoking, alcohol consumption and a number of infections (malaria, brucellosis, human immunodeficiency virus, influenza virus and so on), we could only calculate unadjusted pooled RRs/ORs. Hence, the unadjusted pooled RRs/ORs results in our study should be interpreted with caution.

Conclusion

Indiscriminate HPV genotype infection can increase the risk of spontaneous abortion and HR-HPV infection was a risk factor for sPTB. However, there was not enough evidence to indicate the association between HPV infec-

tion and pregnancy rate of ART, and spontaneous abortion of ART pregnancy. Different genotypes of HPV infection (HR-HPV or LR-HPV) may play varied roles in adverse pregnancy outcomes; hence, further epidemiologic research is required to clarify whether HPV infections indeed increase adverse pregnancy outcomes.

Ethical Standards

This article does not contain any studies with human participants.

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