High-risk human papillomavirus and risk of preeclampsia: a possible connection?

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Abstract

Objectives. *there is growing evidence of the high prevalence of human papillomavirus (HPV) in both spontaneous abortions and spontaneous preterm delivery; our purpose was to determine the correlation between high-risk (HR)-HPV cervical infection in early pregnancy and preeclampsia (PE).* ***Methods.*** *we conducted a retrospective case control study carried out in "Prof. Dr. Panait Sarbu" Hospital in Bucharest on a one-year period. Women who tested positive for HR-HPV at entry to prenatal care (n=108) were compared with those who were HR-HPV negative (n=216). We assessed the relationship between the presence of HR-HPV and preeclampsia (as defined by clinical guidelines).* ***Results.*** *One hundred and eight women with HR-HPV were matched with two hundred sixteen women HR-HPV negative. Patients carrying HR-HPV were younger (mean age 25.03 ± 3.42 years old vs. 27.08±3.14 years old, p< 0.001), more likely to be nulliparous, had lower body mass index (mean value 22.58± 3.01 Kg/m2 vs. 25.35± 3.19 Kg/m2, p<0.001) and more likely to develop PE (13% vs 3.7%, p= 0.001; adjusted odds ratio 5.30; 95% confidence interval, 2.03-13.84).* ***Conclusions.*** *we observed that HR-HPV infection was associated with an increased risk of developing preeclampsia. Further larger prospective studies are required to evaluate the mechanisms by which HR-HPV induces PE taking into account that this infection is nowadays preventable with vaccination.*
Keywords: *human papillomavirus, pregnancy, preeclampsia*

Introduction

Human papillomavirus (HPV) is the most common sexually transmitted infection among humans[1] and its relationship with cervical cancer is now well-established[2,3]. HPV infection causes virtually all cases of cervical intraephitelial neoplasia-3 and cervical cancer and approximately 40% to 50% of vaginal and vulvar cancers[4,5].

Although it has been clearly documented the sexually transmission of HPV, a number of studies have found that HPV infection may also be transmitted by nonsexual routes[6], taking into account that the virus has been detected in blood, reproductive and plancental cells, as well as in infants, in children and persons who have never had sexual intercourse. There is growing evidence of vertical transmission which, in theory, can occur by means of the following mechanisms: periconceptual transmission (during fertilization of an oocyte or immediately after fertilization), prenatal (during pregnancy), and perinatal (during or immediately after birth)[7].

The natural history of HPV infection is modified by physiological occurring changes in host immunity and hormone levels during pregnancy[8]; by inducing a temporary impaired cell-mediated immunity, the state of pregnancy often facilitates clinical expression of both recently acquired and long-term latent HPV infection[9].

Preeclampsia (PE) represents a pregnancy-specific syndrome that can affect virtually every organ system causing high maternal and fetal morbidity and mortality[10]. A number of mechanisms have been proposed to explain the cause of PE, leading to the idea that this complication appears to be a culmination of factors: abnormal trophoblastic invasion of uterine vessels, immunological maladaptative tolerance between maternal, paternal and foetal tissues, maternal maladaptation to cardiovascular or inflammatory changes of normal pregnancy, genetic factors[11].

In addition to the fact that HPV infections have been proved to be associated with cervical dysplasia and cancer, they also appear to play a role in abnormal pregnancy outcomes. It is well-known that HPV is a small, double stranded epitheliotropic virus typically infecting keratinocytes, but also possibly epithelial trophoblastic placental cells[12].

Clinical studies have shown that trophoblast cells express HPV receptors and are the preferential target for HPV in spontaneously aborted products of conception[13]. High-risk (HR)-HPV oncoproteins alter the main human trophoblastic functions by affecting its growth and adhesion and by increasing its migratory and invasive properties[14], results obtained by these studies suggesting that HR-HPV induces abnormal placental growth resulting in pregnancy wastage[15]. Consequently, failed trophoblast invasion in early pregnancy may lead to other adverse obstetric outcomes associated with placental dysfunction, as is the case for PE[16].

In the light of all the above, our purpose was to determine whether there is a connection between HR-HPV in early pregnancy and the risk of PE.

Methods

The present study is a retrospective case control study carried out in the Clinical Hospital of Obstetrics and Gynecology "Prof. Dr. Panait Sarbu", from Romania reviewing women who delivered in our clinic from June 2013

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to June 2014. We used the patients' medical records in our hospital; the exclusion criteria were: delivery prior to 24 weeks and unavailable HR-HPV deoxyribonucleic acid (DNA) testing at entry to prenatal care. This study was approved by the Committee of Ethics and Research in Humans of our institution. We matched 108 women who tested positive for HR-HPV at entry to prenatal care with 216 women who tested HR-HPV negative at entry to care of the index pregnancy. Subjects in the unexposed group were matched to those in the exposed group to within 2 days of the delivery date in a 2 to 1 ratio. We used for the reporting results of HR-HPV infection the Hybrid Capture II system (Digene Diagnostics Inc., Gaithersburg, MD, USA) which is an *in vitro* nucleic acid hybridization assay with signal amplification using microplate chemiluminescence for the qualitative detection of thirteen HR types of HPV DNA in cervical specimens. The HPV types detected by the assay were the HR-HPV 16, 18, 31, 33, 35, 39, 45, 51, 52,56, 58, 59, 68 (the test couldn't determinate the specific HPV type present)[17].

We collected the basic patients' characteristics: age, parity, body weight (in kg) and height (in meters), smoking status, history of PE, chronic hypertension, gestational age at delivery (in weeks). Body mass index (BMI) is defined as the individual's body weight (in kg) divides by the square of their height (in meters).

PE was defined according to the American Congress of Obstetricians and Gynecologists guidelines as the presence of either a systolic blood pressure ≥140mmHg and/or diastolic blood pressure ≥90 mmHg on 2 occasions at least 6 hours apart, along with proteinuria (either ≥1+ in dipstick test or ≥300 mg in an adequately collected, timed urine sample) after 20 weeks of gestation[11]. We assessed proteinuria by using 24 hour urine result; we did not use protein dipsticks in any of the patients in this study. The primary outcome measured was PE and the secondary outcomes were severity of PE, gestational age at onset and birth weight. Management of PE was determined according to the Romanian College of Obstetrics and Gynecology clinical guidelines which have been revised and approved in 2009 in conformity with WHO[18].

All patients delivered in our clinic and signed an informed consent at admission according to the World Medical Association Declaration of Helsinki regarding both delivery and anesthesia. We did not consider PE to be an absolute indication of caesarian section and the decision of the way of delivery was taken according to the obstetrical indication in each case[18].

Data obtained was reported as mean ± standard deviation for continuous variables and as a percentage for categorical variables. Clinical characteristics of the participants were compared using Student's t-test and the Chi-square test or Fisher's exact test for continuous and categorical variables, respectively. We have also used a multivariate logistic regression analysis to determine the relationship between PE and HR-HPV infection after adjusting for demographic and clinical variables significantly different between the two groups or known to be associated with PE (age, smoking status, nulliparity). The level of statistical significance was set at 5% ($p<0.05$) and all reported p- values were two-tailed. We used for the statistical analyses IMB SPSS Statistics 20.0.0.

Results

During the study period, 108 patients testing HR-HPV positive at entry to prenatal care were matched to 216 patients who tested negative for HR-HPV in their cervical specimens. The demographic and obstetric characteristics of the study participants are summarized in Table 1. There were statistically significant differences in age, parity,

Table 1 Characteristics of the study participants

Characteristic	HR-HPV*** positive (N= 108)	HR-HPV negative (N= 216)	p- value
Age, y	25.03± 3.42	27.088± 3.14	0.001
BMI*	22.58± 3.01	25.35± 3.19	0.001
Nulliparity(%)	33.3	22.2	0.03
Smoking(%)	22.2	12.5	0.02
Chronic hypertension(%)	2.8	4.2	0.39
History of PE**(%)	1.9	6	0.09
Twin pregnancy(%)	3.7	1.9	0.25

**BMI=body mass index; **PE=preeclampsia; ***HR-HPV=high-risk human papillomavirus*

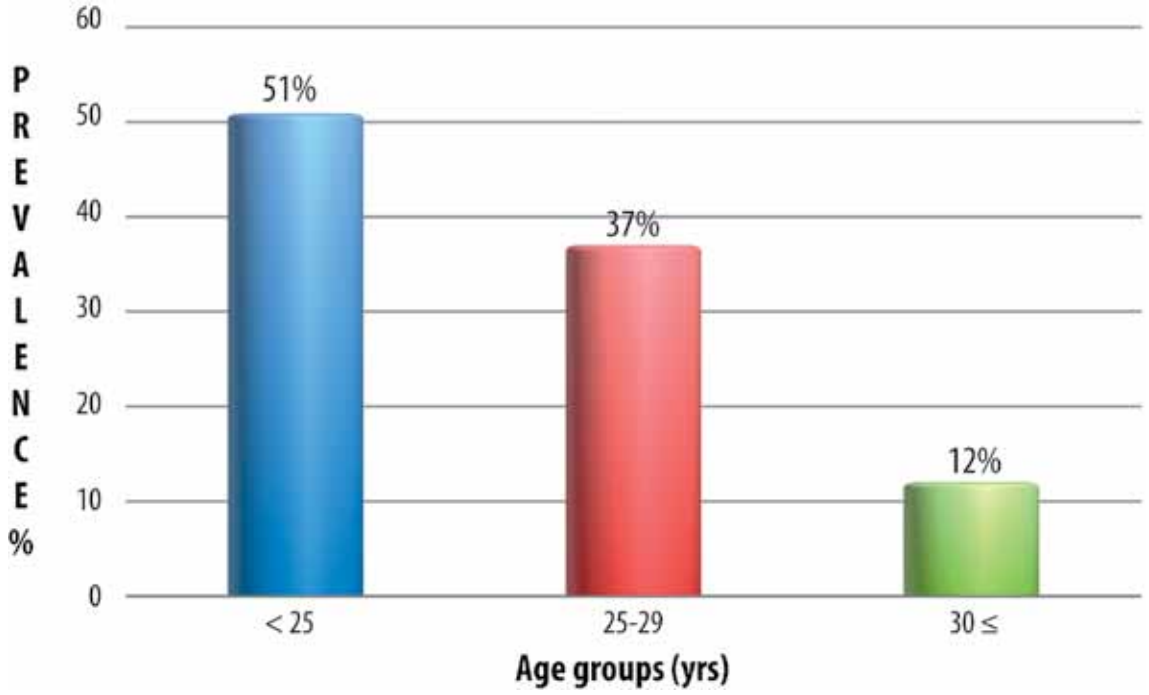

Figure 1. ***Positioning of volume caliper to measure MPI***

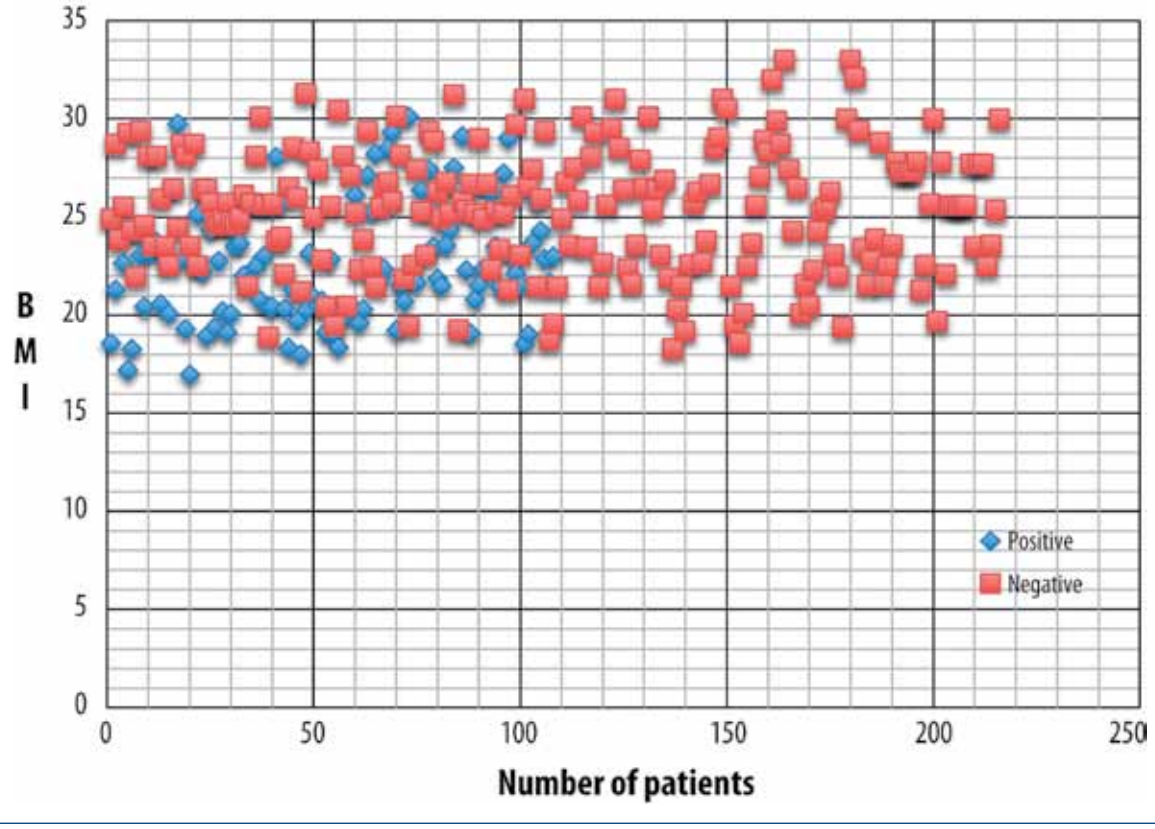

Figure 2. ***BMI distribution according to HR-HPV status***

body BMI and smoking status between the exposed and unexposed groups: women in the HR-HPV group were younger (mean age 25.03±3.42 years old vs. 27.08±3.14 years old, p<0.001) with 55.5% of the women from the exposed group having ages below 25 years old (Figure 1), more likely to be nulliparous (33.3% vs. 22.2%, p<0.03), had lower BMI (mean value 22.58±3.01 vs. 25.35±3.19, p<0.001; Figure 2) and more likely to be smokers (22.2% vs. 12.5%, p<0.02).

There was no difference regarding the history of PE between the two groups (1.9% vs. 6%, p<0.09). Therefore, no statistically significant difference was obtained for the presence of chronic hypertension (2.8% vs. 4.2%, p<0.39).

Table 2 shows the pregnancy outcomes of the two groups of the study. We observed that the distributions of the delivery mode (vaginal birth vs. caesarean section) did not vary substantially between the exposed and unexposed groups (caesarean section rate of 53.7% vs. 57.87%, p<0.4), nor the ones regarding the gestational age at delivery (mean ages 38.31±1.35 weeks vs. 38.53±1.28 weeks, p<0.15). Women with HR-HPV were more likely to develop PE as defined by the clinical guidelines in comparison to the HR-HPV negative group (13% vs. 3.7%, p<0.001), with an odds ratio of 3.87 ;95% confidence interval, 1.57-9.54). We confirmed the strength of this association by using the logistic regression model after adjusting for age, nulliparity, smoking status (adjusted odd ration of 5.3; 95% confidence interval, 2.03-13.84).

Although there was no difference in the gestational age at onset of PE (37.00 weeks [35.5, 37.2] vs. 36.65 weeks [36, 37.1], p<0.9), we observed a tendency of women in the exposed group to develop severe PE compared with the unexposed group (6.5% vs. 1.4%, p<0.01). Regarding birth weight, in our study we obtained lower values in the HR-HPV positive group in comparison to the HR-HPV negative group (3245.5±291.54 g vs. 3380.38±317.43 g, p<0.001).

Discussion

In the present study, we sought to determine the association between the presence of HR-HPV at entry to prenatal care and the development of PE. By matching 108 patients who tested positive for HR-HPV with 216 patients HR-HPV negative, we observed that there is a statistically significant association between the infection with HR-HPV in early pregnancy and the risk of PE. This association was still significant after adjusting for factors that may affect predisposition to PE, as age, nulliparity and smoking status. Interestingly, although studies showed that excessive weight gain among obese patients increases overall risk of pregnancy-related hypertension[19], the patients in our exposed group had lower BMI values than the patients in the unexposed group.

The correlation between HR-HPV infection and adverse pregnancy outcomes has been the subject of other recent studies[20,21]. Mc Donnold et al. showed in their paper that women whose Papanicolaou smear at entry to prenatal care was read as either low grade squamous intraepithelial lesions, high grade squamous intraepithelial lesions or atypical squamous cells of undetermined significance with HPV DNA positive for HR types were more likely to develop PE compared to women who had 2 normal pap smears at entry to care (10.19% vs. 4.94%, p=0.004), results that are comparable to the ones obtained in our study. We used in our study HPV typing results as criteria for the exposed/nonexposed group as opposed to Mc Donnold's study in the effort to rule out the possibility of false-positive and false-negative results of HR-HPV infection based on pap smear interpretations. On the other hand, we did not assess in our study other adverse pregnancy outcomes correlated with HR-HPV infection, such as preterm delivery or premature rupture of the membranes (PROM). In their paper, Cho et al. observed that HR-HPV infection has a high prevalence in pregnant women (14.1% of their study participants), it is more common in women below the age of 25 (which is similar to our observations) and that PROM was more common in patients with HR-HPV infection[19]. Still, they did not find a correlation between HR-HPV infection and preterm delivery, PE or gestational diabetes mellitus. Similarly, HPV was detected more frequently in placentas from spontaneous preterm deliveries than in placentas from controls, as shown by Gomez et al. in their study[22].

The results obtained in our study could be explained on a molecular basis by the growing evidence of the possible transplacental transmission of HPV. Recently, it has been revealed that HPV-16 is able to replicate its DNA de novo and produce progeny in placental trophoblasts cultures[14]. HPV-16 early proteins enhance trophoblastic growth and intensify the malignant phenotype by impairing cell adhesion (decreasing E-cadherin expression) leading to increased cellular motile and invasive properties[14,15]. This is a surprising report taking into account that HPV was previously believed to be an exclusively keratinocyte/skin-specific virus[23]. In addition, HPV oncogenic component genes E6 and E7 were proved to have serious effects upon the cellular behavior of the trophoblasts including cell survival, endometrial binding, proliferation, differentiation, and immortalization[23]. This is the reason for which You et al. suggested in their study that these alterations in the trophoblast/placental physiology may be responsible for the increased prevalence of HPV infection in spontaneously aborted products of conception compared to elective abortions[13,23,24]. By inducing cell death (rates of apoptosis being 3- to-6 fold greater in transfection cells than in non-transfection cells), HPV infection of extravillous trophoblasts may cause placental dysfunction and, thus, explain its association with various adverse pregnancy outcomes[22,25].

Being a transient infection[7,26], it is possible that early HR-HPV infection in pregnancy could modify placental vascular architecture and thus result in PE, but then become undetectable in the placenta at the time of delivery. McDonnold et al. discussed also the hypothesis of HR-HPV infection acting in PE in a similar manner to the one found in atherosclerotic cardiovascular disease: inflammation, hypercoagulability, or vascular alterations[20].

Nevertheless, our findings have several limitations.

Table 2 Obstetric outcomes of study participants

Characteristic	HR-HPV** positive (N= 108)	HR-HPV negative (N= 216)	p- value
Gestational age at delivery, wks	38.31± 1.35	38.53± 1.28	0.15
Birthweight, g	3245.5± 291.54	3380.38± 317.43	0.003
PE* (%)	13	3.7	0.001
Gestational age at onset of PE, wks	37 (35.5, 37.2)	36.65 (36, 37.1)	0.91
Severe PE (%)	6.5	1.4	0.01
Caesarean delivery (%)	53.7	57.87	0.4

**PE=preeclampsia; **HR-HPV=high-risk human papillomavirus.*

First, we did not demonstrate a relationship of causality, but only showed an association between two factors. Secondly, our study is retrospective and the number of patients with PE in each group was lower than expected, taking into account that testing for HR-HPV types is still not a common practice for patients delivering in our institution. Most of our patients were diagnosed with PE before delivery; of the exposed and unexposed groups, 42.82% and 50% respectively of patients with PE presented before 37 weeks of gestation, were admitted to hospital and had the proteinuria confirmation by 24 hour urine results. We did not use urine dipstick to diagnose proteinuria in none of the cases in this study and we did not include intrapartum blood pressure values, nor systolic and diastolic blood pressure at entry to care. Thirdly, due to the retrospective nature of our study we lacked information regarding sexual behavior (number of sexual partners, use of condoms and other active sexually transmitted infections) which can act as or increase the risk factors for HR-HPV infection during pregnancy. This is a potential confounder that ought to be included in potential future studies.

Conclusions

We observed a statistically significant correlation between HR-HPV infection early in pregnancy and PE. Further prospective studies or larger cohorts are needed to confirm our findings and also to evaluate the mechanisms by which this association takes place; if this is to happen, then HPV vaccination may prove to have additional health benefits by possibly participating in the prevention of various adverse pregnancy outcomes, including PE. ■

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