

Assessment of frequency of regression and progression of mild cervical neoplasia – LGSIL in women with positive high-risk HPV DNA test result

Ocena częstotliwości regresji oraz progresji śród nabłonkowej neoplazji szyjki macicy niskiego stopnia – LGSIL u kobiet z pozytywnym wynikiem testu molekularnego identyfikującego DNA wysoce onkogennych typów wirusa brodawczaka ludzkiego

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Abstract

Objectives: Assessment of frequency of regression and progression of mild cervical neoplasia in women positive for types of HPV DNA of high oncogenic potential.

Materials and methods: 111 women were studied. One-year-long observation of patients included cervical cytology, conducted every three months, and colposcopy, conducted every six months. After a period of 12 months all women were evaluated with colposcopy and directed biopsies of abnormal cervical tissue.

Results: This study confirms the significant effect of age on both regression and progression of low-grade cervical intraepithelial neoplasia.

Conclusions: In the age group below 26 years, complete regression of LGSIL occurs significantly more frequently than in older women. Whereas in the over 36 age group, progression to HGSIL occurred more frequently during 12 months of follow-up.

Key words: **cervical intraepithelial neoplasia / HGSIL / LGSIL / molecular test / progression / regression /**

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Streszczenie

Cel pracy: Ocena częstotliwości regresji oraz progresji śródnaślankowej neoplazji szyjki macicy niskiego stopnia – LGSIL u kobiet z pozytywnym wynikiem testumolekularnego identyfikującego DNA wysoce onkogennych typów wirusa brodawczaka ludzkiego.

Materiał i metoda: Cała grupa badana licząca 111 kobiet z rozpoznaniem histopatologicznym LGSIL została poddana rocznej obserwacji cytologicznej, wykonywanej co trzy miesiące oraz kolposkopowej, wykonywanej co 6 miesięcy. Po okresie 12 miesięcy u wszystkich pacjentek wykonano badanie kolposkopowe połączone z biopsją celowaną zmian szyjki macicy.

Wyniki: Zaobserwowano znaczący wpływ wieku na odsetek kobiet, u których dochodzi do regresji bądź progresji zmian o charakterze śródnaślankowej neoplazji szyjki macicy niskiego stopnia – LGSIL.

Wnioski: W grupie kobiet poniżej 26 roku życia całkowita regresja zmian typu LGSIL występuje znacznie częściej, a w populacji kobiet powyżej 36 roku życia w czasie rocznej obserwacji częściej dochodzi do progresji w kierunku zmian typu HGSIL.

Słowa kluczowe: **śródnaślankowa neoplazja szyjki macicy / HGSIL / LGSIL /
/ test molekularny / progresja / regresja /**

Introduction

Morbidity and mortality resulting from cervical cancer is a substantial social problem in Poland. With regard to registered annual incidence rate, it is the second most common malignant cancer in women after breast cancer. Physicians diagnose cervical cancer every minute, and every two minutes one woman dies as a result of this disease worldwide [1].

Pre-cancerous lesion of the cervix is cervical intraepithelial neoplasia (CIN). Low-grade squamous intraepithelial lesion (LGSIL) corresponds to CIN 1, and high-grade squamous intraepithelial lesion (HGSIL) histologically corresponds to CIN 2 and CIN 3. Diagnosis and treatment of CIN is an element of secondary prophylaxis and is a part of an integrated system, which purpose is to decrease morbidity and mortality resulting from cervical cancer [2].

Each year, physicians diagnose close to 1.2 million LGSIL, 300 thousand HGSIL, and 10 thousand cases of cervical cancer in the United States alone [3]. Each year, in the City of Portland, United States, 1.2 and 1.5 per 1000 women are diagnosed with LGSIL and HGSIL respectively. In the age group between 25 and 29 years, the diagnosis rate is highest and amounts to 8.1 per 1000 patients [4].

The role of persistent infection with human papillomavirus in the process of carcinogenesis of cervical cancer has been proven by multiple studies. In the year 1996, World Health Organization recognized the infection with oncogenic types, 16 and 18, of HPV as a factor of human carcinogenesis. Presence of DNA HPV was found in 99.7% of cervical cancer cases, which confirms the relationship between HPV infection and cervical intraepithelial neoplasia or cervical cancer [5,6,7].

Infection with HPV is mainly transmitted via sexual contact. More than 80% of sexually active women and men were, are, or will be infected with HPV in their lifetime [8]. In women below 25 years of age most infections regress in the course of 12 to 18 months. Persistence of infection, for a period of more than 24 months, is usually associated with viruses of high oncogenic potential. The incidence of infection with HPV decreases with age. However, risk of infection transformation to chronic persistent infection increases with age of the infected individual [9].

Objectives

Assessment of frequency of regression and progression of mild cervical neoplasia in women positive for types of HPV DNA of high oncogenic potential.

Materials and methods

One hundred eleven women diagnosed with low-grade cervical intraepithelial neoplasia were studied from 2008 to 2011 at the Laboratory of Cervical Pathophysiology, Gynecology and Obstetrics Clinical Hospital in Poznań. The mean age of patients at diagnosis was 29.2 years, median age was 27 years. The patient age range was 17 to 60 years. Patients were observed for a period of one year. Study design included cytology every three months and colposcopy every six months. After 12 months all patients had colposcopy, directed biopsies and liquid based cytology for molecular testing of high oncogenic HPV DNA. Every patient has given a written consent to the tests.

Cytological Investigations: Cells were sampled from ectocervix and endocervix using cervex-brush. Prepared slides were examined under optic microscope, Axiolab (Zeiss), using 200 and 400 times magnification. The specimens were classified according to The Bethesda System. The results were recorded by cytotechnician, supervised by a cytotechnician with ten years of experience. Every tenth normal specimen with respect to cyto-oncologic changes was subjected to a repeat evaluation conducted by a more experienced cytotechnician. All specimens, classified as abnormal with respect to cyto-oncologic changes, were subjected to a repeat evaluation by a pathologist. Such an evaluation schematic of cytologic specimens is consistent with principles applied in mass prevention of cervical cancer programs used worldwide.

Virological Investigations: The test material was obtained, using a cervex-brush, from the external os of the cervix and vaginal wall. The obtained specimen was placed into a liquid-based medium, ThinPrep PreserCyt Solution (Cytoc Corporation), and stored at temperature of +2 to +8°C. The specimens were further treated in Laboratory of Pathophysiology of Uterine Cervix.

In the study, molecular test PCR - Amplicor HPV Roche Diagnostics, which identifies the presence of one or more of the 13 oncogenic types of human papillomavirus, was employed.

Amplicor HPV test is a quality test. It serves to identify high-risk HPV DNA of the following genotypes: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59 and 68, in vitro. Positive test in molecular test PCR - Amplicor HPV Roche Diagnostics confirms the presence of DNA of at least one of the 13 oncogenic types of human papillomavirus in the collected specimens.

Simultaneously, isolation and amplification of β -globin gene provides expert analysis of the number of cells and inhibition reactions in every tested specimen. Amplicor HPV Roche Diagnostics is the only available test that is PCR based technique with a certificate of reproducibility, CE marking, in in vitro medical diagnostics.

Colposcopic Investigations: In the study, stereoscopic colposcope (Olympus OCS-500) was used. Three to five percent acetic acid and Schiller's test were used to stain the cervix during colposcopy evaluation. Colposcopic images were evaluated based on eight point Reid's Colposcopic Index (RCI), evaluating lesion margin, acetowhite epithelium after staining with acetic acid, iodine-negative areas, and atypical vessels. Abnormal result was defined as three or higher out of eight RCI.

Histopathologic Investigations: All patients had colposcopy-directed biopsies of abnormal cervical tissue and endocervical curettage. Indication for a histopathological verification was primarily an abnormal pap smear. All histopathologic specimens were subjected to an evaluation in the Laboratory of Pathomorphology at the Gynecology and Obstetrics Clinical Hospital in Poznan.

Statistical Investigation: Statistical analysis included 111 patients with diagnosis of LGSIL and positive molecular test that identified DNA types of human papillomavirus of high oncogenic potential. Calculations, as well as graphic analysis, of results were conducted using Microsoft Office program - Excel 2010.

Results

Out of 111 patients participating in this study, 38 (34%) patients showed lesion regression confirmed by histopathological findings after 12 months of follow-up. In the case of 45 (41%) patients, initially diagnosed with LGSIL, abnormalities persisted at the same level. In the remaining 28 (25%) patients progression of lesion to HGSIL was observed. (Figure 1). It is worth noting that, none of the patients developed invasive cervical cancer.

In the age group below 26 years, histopathology confirmed lesion regression was observed in 21 (51%) patients after one year. In 12 (29%) patients, diagnosed with CIN 1, the degree of intraepithelial neoplasia persisted at the same level. In eight (20%) other patients under 26 years, cervical neoplasia progressed to HGSIL. (Figure 2).

In the age group between 26 and 35, histopathology confirmed lesion regression was observed in 17 (32%) patients after one year of observation. In 23 (43%) patients, initially diagnosed with LGSIL, the degree of intraepithelial neoplasia persisted at the same level. In the remaining 13 (25%) patients in this age group, lesion progression to HGSIL was observed. (Figure 3).

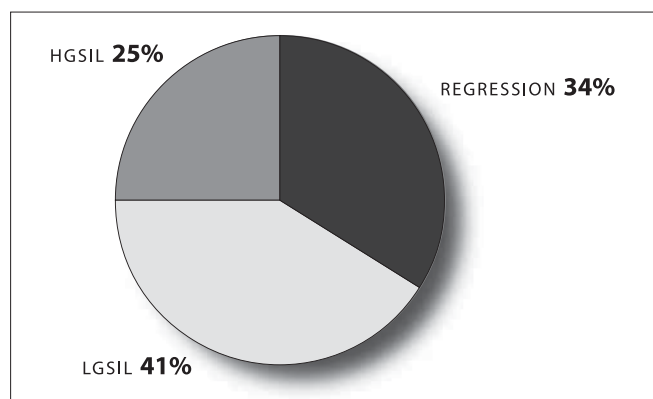


Figure 1. Assessment of frequency of regression and progression of mild cervical intraepithelial neoplasia – LGSIL in HPV DNA HR positive women.

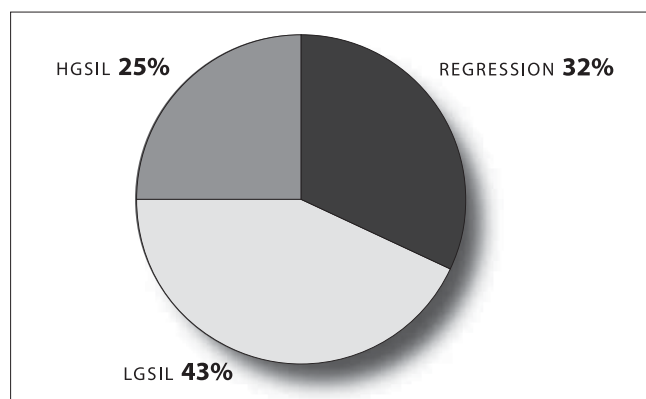


Figure 3. Assessment of frequency of regression and progression of mild cervical intraepithelial neoplasia – LGSIL in HPV DNA HR positive women between the age of 26 and 35.

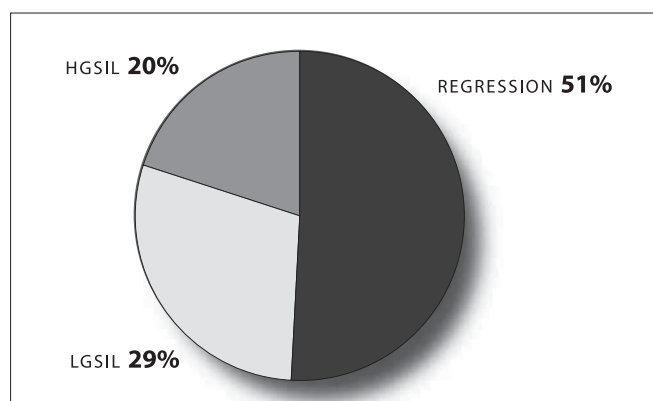


Figure 2. Assessment of frequency of regression and progression of mild cervical intraepithelial neoplasia – LGSIL in HPV DNA HR positive women under the age of 26.

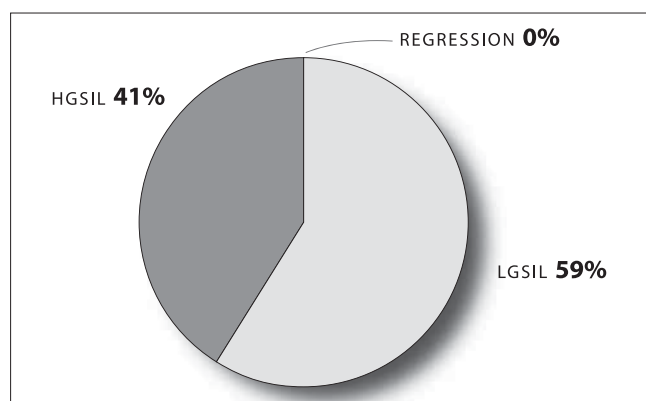


Figure 4. Assessment of frequency of regression and progression of mild cervical intraepithelial neoplasia – LGSIL in HPV DNA HR positive women over the age of 36.

In the over 36 age group none of the female patients' lesions regressed, which was confirmed by histopathological tests. In the case of 10 (59%) patients, initially diagnosed with LGSIL, the degree of intraepithelial neoplasia persisted at the same level. In seven (41%) of the patients in this age group lesion progression to HGSIL was observed. (Figure 4).

Comparisons between the three age groups show that the percentage of patients with either regression or progression of cervical intraepithelial neoplasia is significantly influenced by age. In the youngest patients, histopathology confirmed regression of lesions was most prevalent – it was observed in over 50% of patients after 12 months of follow-up. In the age group between 26 and 35 histopathology confirmed regression was observed in over 30% of patients after 12 months of observation. On the other hand, in the older age group, seven (over 40%) of the patients' lesions progressed to HGSIL. (Figure 5).

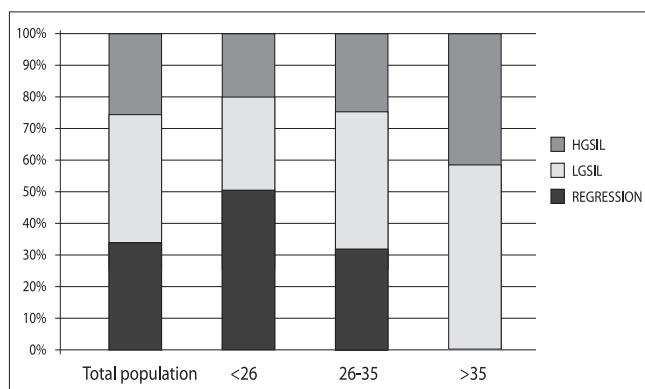


Figure 5. Comparison of frequency of regression and progression of mild cervical intraepithelial neoplasia – LGSIL in HPV DNA HR positive women in the entire study population, as well as in specific age groups.

Discussion

Persistent viral infection precedes in time the development of cervical cancer. It takes about four to five years from initial infection to development of advanced cervical intraepithelial neoplasia. Development of cancer generally takes nine to 15 years from initial HPV infection [10].

Observation approach and “wait and see” strategy for a period of 12 months results from the fact that 80% of lesions, such as LGSIL, are a direct consequence of incidental infection with HPV and along with spontaneous regression of HPV infection, lesions undergo spontaneous regression. Such an approach is particularly indicated in nulliparous women under the age of 30. In some research centers frequency of spontaneous regression of LGSIL was estimated to be almost 90% [11, 12]. Indications for observation of lesions characteristic of LGSIL results from the fact that only 15% of lesions of this type progress to HGSIL [13].

Pretorius et al. found that after 24 months of observation of LGSIL, only 47 out of 2490 patients had lesion progression to HGSIL and cervical cancer, which constituted 1.9% of the entire studied population. In the above referenced study, authors also observed increase in the frequency of occurrence of progression of LGSIL with increasing age of the women studied [14].

In teenagers and young women planning future pregnancies, with a diagnosis of LGSIL, Polish Gynecologic Society

recommends pap smears once a year and in the case of an abnormal pap smear result, colposcopy investigation and possibly colposcopy-directed biopsies [13].

In Ostor's publication the frequency of regression of LGSIL was estimated to reach 60%, in 10% progression to HGSIL occurred, and in 30% cervical intraepithelial neoplasia persisted at the same level [15].

In our study, in the group of 111 women with histopathological diagnosis of LGSIL, we observed significant influence of age on the percentage of women, in whom either progression or regression of low-grade cervical intraepithelial neoplasia occurred. In the age group below 26 years of age, after 12-month observation we found that 50% of LGSIL regressed, which was confirmed by histopathologic investigation. In the age group between 26 and 35 years, regression occurred in 30% of evaluated women, whereas in the age group over 36 years, 40% of women showed progression to HGSIL. This analysis confirms the importance of intensive cytological and colposcopic follow-up of low-grade cervical intraepithelial neoplasia for a period of 12 months.

Conclusions

In the age group below 26 years, complete regression of lesion of cervical intraepithelial neoplasia occurs significantly more frequently than in older women.

Whereas in the over 36 age group, progression to CIN 2 and CIN 3 occurs more frequently than in younger women during 12 months of follow-up.

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