

## TYPES OF DYSPLASTIC LESIONS AND COMPLIANCE WITH FOLLOW UP IN NORTHEASTERN ROMANIA – A RETROSPECTIVE STUDY

I. L. Stoian<sup>1</sup>, C. Ilea<sup>1,3\*</sup>, Raluca Anca Balan<sup>1,2</sup>, Demetra Gabriela Socolov<sup>1,3</sup>

“Grigore T. Popa” University of Medicine and Pharmacy Iasi, Romania

Faculty of Medicine

1. Department of Mother and Child Medicine

2. “Elena Doamna” University Hospital Obstetrics-Gynecology, Iasi, Romania

3. “Cuza-Vodă” University Hospital Obstetrics-Gynecology, Iasi, Romania

\*Corresponding author. E-mail: ciprian.ilea@umfiiasi.ro

TYPES OF DYSPLASTIC LESIONS AND COMPLIANCE WITH FOLLOW UP IN NORTHEASTERN ROMANIA - A RETROSPECTIVE STUDY (Abstract): Cervical cancer is a worrisome condition as it correlates with a high mortality rate although being one of the most preventable cancer types. **Material and methods:** The medical data of all patients subjected to an excisional procedure between 2000 and 2020 were retrospectively analyzed. Thus, data regarding age, obstetric history, types of dysplastic lesions in the history, HPV infection status, colposcopic findings, identified dysplastic lesions, and the number of screening tests performed were analyzed. Data entry and analysis were performed using SPSS software version 20. **Results:** The study included 455 patients aged between 19 and 62 years old, mean age of 36 years and a standard deviation of 9.07. The analysis of the initial cytology results revealed that the most frequently identified dysplastic lesion was HSIL (High Grade Squamous Intraepithelial Lesion), closely followed by LSIL (Low-grade squamous intraepithelial lesion). The analysis of HPV DNA test recommendations highlighted the fact that in over 50% of cases this test was not performed. In the cases in which DNA HPV test was performed, strains 16 and 18 were most frequently identified. The colposcopic examination revealed that the lesions located within the transformation zone were the most numerous. Of the described lesions, the acetowhite epithelium was most often identified, either isolated or in combination with punctation, mosaic pattern of vascularization or leukoplakia. Conization and LLETZ (Large Loop Excision of the Transformation Zone) were the two excisional procedures used in 46.4% and 44% of the patients, respectively. Histopathological examination showed: 12.5% of the patients presented high-grade HSIL (CIN3) lesions, 47.5% low-grade CIN 1 lesions, and 7% were diagnosed with carcinoma in situ. In the post-intervention follow-up, a total of 202 patients (44.39%) showed up for the first and only 66 patients (14.5%) returned for the second post-intervention follow-up. **Conclusions:** The types of dysplastic lesions identified by cytology were found to be in percentages similar to those reported in the literature, with the exception of HSIL diagnosis, which was the most frequently identified. This high rate could explain the increased incidence of cervical cancer in our country. Conization and excision procedures represent a valuable method of conservative treatment, preserving at the same time patient's fertility and avoiding a radical intervention. Low compliance with follow-up among patients from the northeastern region of Romania and the increasing migration of the groups at high risk made us use these excisional procedures even in the case of low-grade lesions in an attempt to reduce the incidence of this disease. **Keywords:** CERVICAL DYSPLASIA, MONITORING, LLETZ, CERVICAL CANCER.

Globally, cervical cancer is the most commonly diagnosed cancer in 23 countries. In recent decades, incidence and mortality rates have decreased in most areas of the world. These declines are attributed to factors related either to the increase in the average socioeconomic status of the target population, or to a lower risk of persistent infection with high-risk HPV resulting from improved genital and sexual hygiene, low parity, and a decreasing prevalence of sexually transmitted diseases. Cervical cancer screening programs have greatly improved the prognosis of this disease after their implementation in many countries in Europe, Oceania and North America. In the absence of effective screening, as is the case in Eastern Europe and Central Asia, rapid increases in premature mortality from cervical cancer have been recorded in recent years (1). Cervical cancer is a worrisome condition because it correlates with a high mortality rate, although being one of the most preventable types of cancer (2). Regarding the measures that can be taken in order to decrease the prevalence, incidence, and mortality of cervical cancer, an optimal system of actions should include the long-term follow-up of both the processes and the results of the three basic activities: vaccination, screening and treatment - as outlined in the global strategy of the World Health Organization (WHO) to eliminate cervical cancer as a public health problem. However, these measures have not been implemented fairly and directly in all countries. As of May 2020, less than 30% of low-income countries have implemented national HPV vaccination programs, compared to over 80% in high-income countries (1). Unfortunately, the importance of early diagnosis and prevention of cervical cancer is still under-

estimated by the general population of Romania; however, as shown by the analyzed data, in the past 10 years young patients (aged 25-40 years) have demonstrated an increased interest in routine gynecological check-ups. So, in this context, we conducted a retrospective study that evaluates the types of cervical lesions detected in northeastern Romania over a 20-year period, compliance with follow-up, as well as the risk of cervical dysplasia recurrence among patients who underwent an excisional procedure.

### MATERIAL AND METHODS

To achieve this goal, we retrospectively analyzed (using the electronic archive and specific registers) the medical data of all patients subjected to an excisional procedure for dysplastic lesions of various degrees in the interval 2000-2020. The diagnosis and excisional procedure were carried out in a private clinic in Iasi town, being performed by gynecologists with over 10 years of experience in the field. All included patients gave consent for the use of personal information for health research and subsequent inclusion in the study. The data on appointments, consultations, results, and interventions in a population of 500 patients from the northeastern region of Romania, namely Suceava, Botosani, Iasi, Neamt, Vaslui, and Bacau counties, who presented themselves at the clinic between 2000-2020 were processed. Following the evaluation and application of the inclusion and exclusion criteria, a number of 45 patients were excluded from the study. Thus, data from 455 patients were analyzed, information related to age, obstetric antecedents, types of dysplastic lesions in antecedents, HPV infection status, colposcopic aspects, identified dysplas-

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tic lesions, and the number of follow-ups. The inclusion criteria were presence of cervical dysplasia or HPV infection, age 18-65 years, sexually active, non-pregnant, intact uterus, without previous treatment for cervical intraepithelial neoplasia. The exclusion criteria were withdrawal of consent, invasive genital cancer at diagnosis, pregnancy, previous cervical excisional procedures, acute bacterial or mycotic cervical-vaginitis, chronic diseases such as liver cirrhosis, kidney failure, cardiovascular diseases, drug addiction or psychiatric diseases. The main goal of the study was to evaluate the types of dysplastic lesions in the northeastern region of Romania, as well as patients' compliance with follow-up in order to identify a possible cause that could explain the increased incidence of cervical cancer among the population in this geographical region. Data entry and analysis were performed using software *SPSS version 20* using descriptive and inferential statistics. In order to maintain data confidentiality and protect the identity of the patients included in the study, the personal data were anonymized. The presented cytological findings are from cervicovaginal smears collected with a specific cervical brush with the help of which cells were taken from the level of the endo and exocervix and prepared mainly in ThinPrep liquid medium (70% of cases), but also from conventional smears, the staining method being the classic one, Papanicolaou. For reporting the results of cervical cytology, the 2014 Bethesda System was used. HPV detection and genotyping were performed by isolation from the collected cervical samples on ThinPrep medium by the gynecologist, later a polymerase chain reaction (PCR) with colorimetric detection and hybridization being used. All samples

were processed in RENAR accredited laboratories. Colposcopic and histological examinations were performed if a diagnosis of atypical squamous cells of undetermined significance (ASCUS) or high-grade atypical squamous cells (ASC-H), low-grade intraepithelial lesion (LSIL), high-grade intraepithelial lesion (HSIL) or carcinoma in situ was made. The colposcopic examination was performed with Video colposcope Full HD DCS 102 PRO by doctors certified in colposcopy and with more than 10 years of experience. During the colposcopic examination performed in the patients included in the study 3 parameters were considered to determine the need for biopsy. Criterion 1: colposcopic impression of lesion severity or lesion grade (G) with three degrees of severity: G1: colposcopic appearance of low-grade lesion, G2: colposcopic appearance of high-grade lesion, but without signs that could suggest invasion, G3 : colposcopic appearance of invasive lesion with signs of severity: atypical vessels, ulcerations, congestion, erosions, vegetative lesions; Criterion 2: the position of the inner line of the transformation zone (TZ) or the squamocolumnar junction line (SCJ): Type 1 (TZ1): The SCJ is fully visible and is located fully on the ectocervix; Type 2 (TZ2): The SCJ is fully visible and is located either fully or partially within the endocervical canal; Type 3 (TZ3): The SCJ is invisible being located within the endocervical canal; Criterion 3: atypical size or area of the transformation zone (Q): Q1: a single quadrant covered by the atypical transformation zone, Q2: two quadrants covered by the atypical transformation zone, Q3-Q4: three or four quadrants covered by the atypical zone transformation (3). Resection techniques included conization, Loop Electrosurgical Excision Proce-

dure (LEEP) or Large Loop Excision of Transformational Zone (LLETZ). The specimens were initially examined macroscopically and later the blocks were paraffin embedded and stained using the classic hematoxylin-eosin (HE) staining method.

This study was conducted in compliance with the principles of the Declaration of Helsinki and approved by the Research Ethics Committee of the Iasi “Grigore T. Popa” University of Medicine and Pharmacy, registration number 57/17.03.2021.

### RESULTS

The study included 455 patients who underwent excisional procedures for dysplastic lesions of various degrees. The study group was divided into 5 groups

depending on the type of dysplastic lesions, but also based on cytology findings according to the Bethesda classification: ASCUS (20.4%), ASCH (14.9%), LSIL (30.3%), HSIL (32.3%), CIS (2%) (tab. II). The study patients were aged 19 to 62 years, average age 36 years, standard deviation = 9.07. The highest percentage (38%) of patients presenting to the clinic for the assessment of dysplastic lesions were aged 31-40 years, and the lowest rate of presentation to the doctor, of only 9%, was recorded in patients aged 19-24 years. As to seeking medical advice, a first maximum peak was recorded in patients aged 27 and a second in those aged 34-35, subsequently the number of patients falling below 15% after the age of 44 (fig. 1).

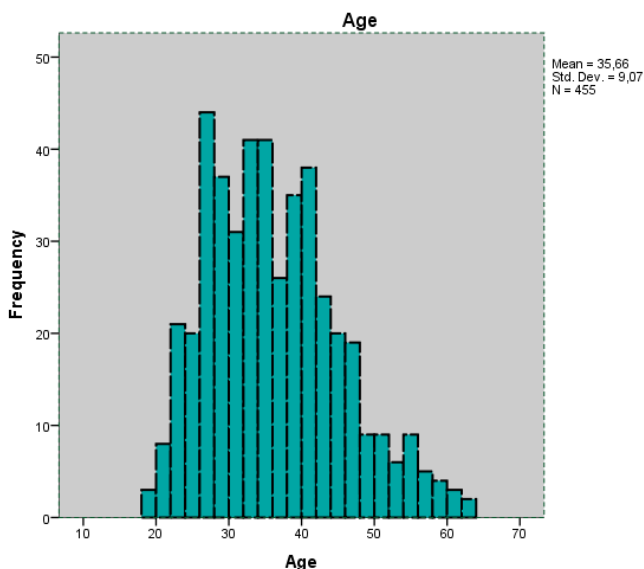


Fig. 1. Age-group frequency

Obstetric history included questions about the number of births and abortions, data that could influence the management of the identified cervical lesions. A maximum of 6 births per patient (with an aver-

age of 2 births per patient) with a standard deviation of 1.05, respectively 5 abortions per patient (with an average of 1 abortion per patient) with a standard deviation of 1.07 were recorded (tab. I). The Kruskal

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Wallis test was used to compare the abortion and birth rates by maternal age and found that the distribution of abortions

does not change with patients' age ( $p>0.05$ ), while obviously the number of births varied by age ( $p<0.05$ ).

TABLE I.  
**Obstetrical history of the study patients. Descriptive Statistics**

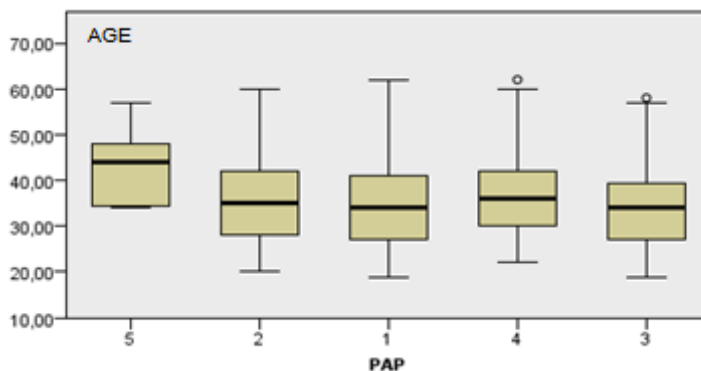
	N	Minimum	Maximum	Mean	Std. Deviation
Number of Births	455	0	6	1.23	1.053
Number of Abortions	455	0	5	0.72	1.079

The analysis of the initial cytodiagnosis revealed that the 455 patients presented the following pathological aspects: the most frequent diagnosis was HSIL, closely followed by LSIL. An initial cytological diagnosis of carcinoma in situ (CIS) was made in

2% of cases. The distribution of these lesions is detailed in table II. Age-distribution according to the type of lesions differed ( $p<0.05$ ), being higher in the case of carcinomas and high-grade lesions and lower in the case of low-grade lesions (fig. 2).

TABLE II.  
**Distribution of dysplastic lesions**

	Frequency	Percent	Valid Percent	Cumulative Percent
ASCH (2)	68	14.9	14.9	14.9
ASCUS (1)	93	20.4	20.4	35.4
CIS (5)	9	2.0	2.00	37.4
HSIL (4)	147	32.3	32.3	69.7
LSIL (3)	138	30.3	30.3	100.0
Total	455	100.0	100.0	



**Fig. 2.** Age distribution according to the type of dysplastic lesions

The analysis of the data on HPV DNA testing revealed ca over 50% of the study

women never had a HPV DNA test, and in the patients in which virus typing was done

the most frequently identified were strains 16 si 18, both as unique strain and in asso-

ciation with other high risk (HR) and low risk (LR) strains.

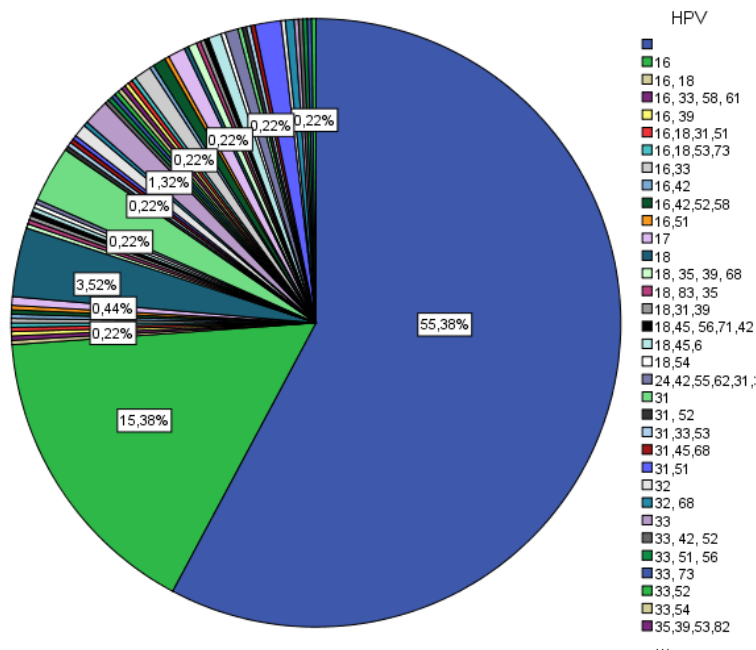


Fig. 3. Distribution of cases according to HPV genotyping results

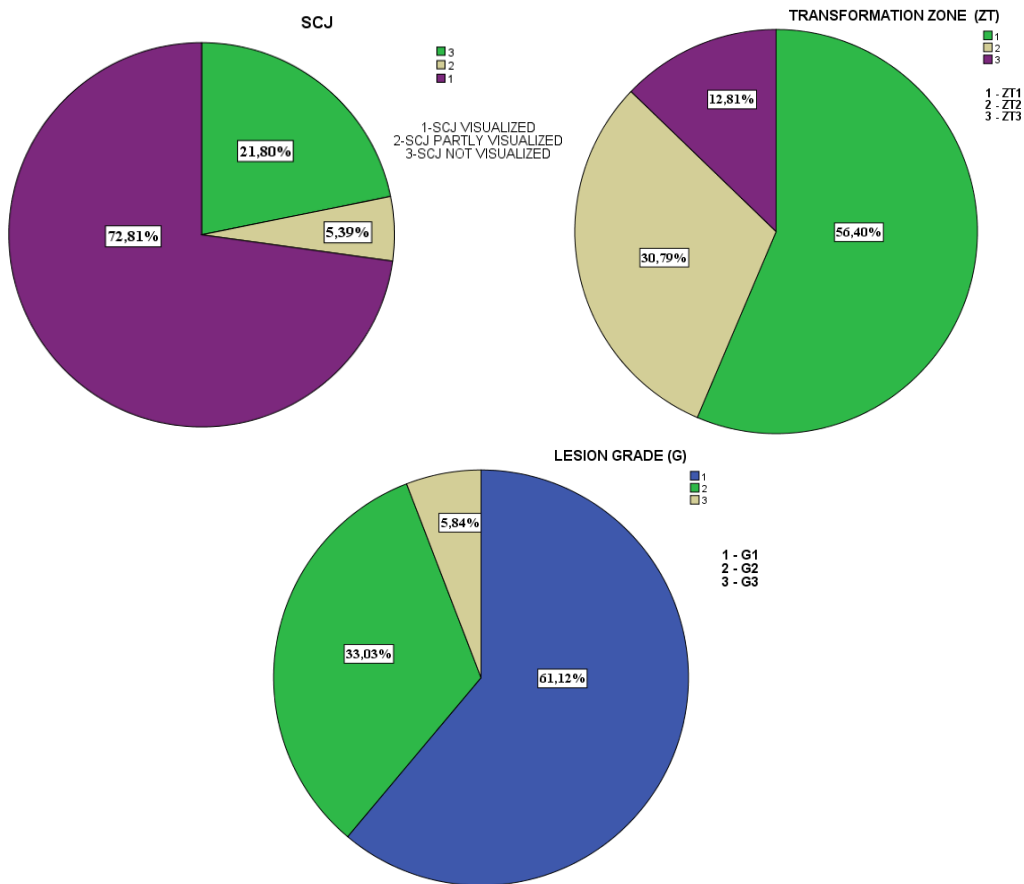
Distribution of HPV strains in the tested patients revealed that the dominant strain was HPV 16, with most of the test being performed in the 26-30 years age group. The prevalence of HR-HPV and HPV 16 strains was found in women aged 24-41 years, after that the infection rate started to decrease.

All 455 study participants underwent colposcopic examination. Colposcopy results showed that the SCJ was visible in 72.81% of cases (fig. 4). Later, during the colposcopic examination, the 3 parameters used to determine the need for biopsy were analyzed: lesion grade (G), squamocolumnar junction (SCJ) and surface of the atypical transformation zone (Q). The obtained results were: G1/low-grade lesions were identified in over 60% of cases (fig. 4); SCJ was fully visible in 56.4%, partially visible in 30.79%, and could not be identi-

fied in 12.81% of cases. The percentage of lesions located solely within the TZ was the highest, thus confirming the potential for missed diagnosis of neoplasias when they develop predominantly at this level.

Of the described lesions, acetowhite epithelium was the most frequently identified, either isolated or in combination with mosaic and punctate vascular patterns or leukoplakia. The mosaic pattern was also seen in 116 examinations and the punctate pattern in 119 examinations. It was found that Mosaic patterns were also seen in 116 examinations and the dotted pattern in 119 examinations. Most colposcopic examinations with pathological findings had lesions limited to 1 or 2 quadrants; in the group with lesions extended to 3 or 4 quadrants, 10 cases with endometrial or vaginal extension were identified.

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**Fig. 4.** Squamocolumnar junction (SCJ), Transformation zone, and Lesion grade

Excisional techniques consisted of 2 procedures performed separately or together: conization performed in 46.4% and LLETZ in 44% of the cases, in only 9 cases a colonoscopy with targeted biopsy being performed (tab. III).

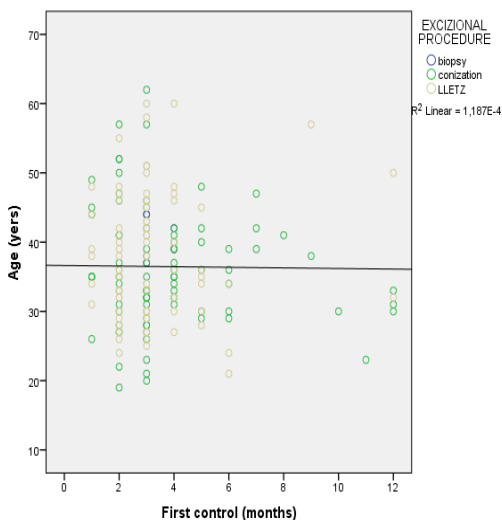
**TABLE III.**  
**Types of performed excisional procedures**

	Frequency	Percentage	Valid Percentage	Cumulative Percentage
	35	7.7	7.7	7.7
BIOPSY	9	2.0	2.0	9.7
CONIZATION	211	46.4	46.4	56.0
LLETZ	200	44.0	44.0	100.0
TOTAL	455	100.0	100.0	

The histopathological findings were: 12.5% of patients had high-grade HSIL (CIN3) lesions (combined or not with lower-grade lesions), 47.5% had a diagnosis of CIN1 (low-grade lesions), and 7% were diagnosed with carcinoma in situ.

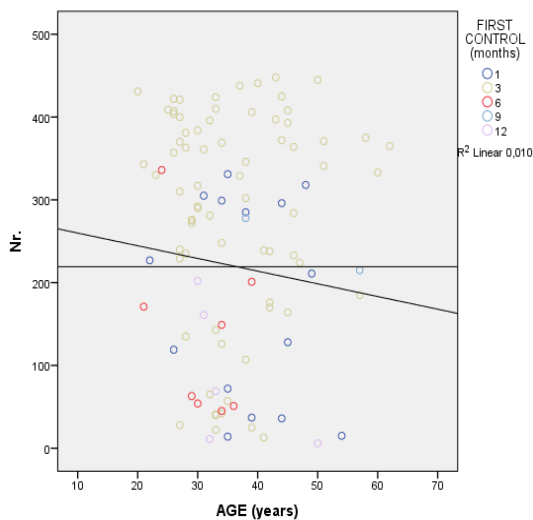
Regarding the post-intervention follow-up, a total of 202 patients (44.39%) showed up for the first follow-up. The first follow-up was performed at 1 to 12 months, on average being performed at 3 months with a standard deviation of 2.22. No direct correlation was found between patients' age and post-interventional follow-up, and the low compliance with follow-up did not prove to be related to patients' age, the correlation between the two variables being

very weak ( $R^2 = 0.01$  (fig. 6)). However, correlating age with the follow-up rate depending on the performed excisional procedure, most patients showed up for follow-up on average 3 months after the interventional procedure, with LLETZ being the most frequently performed. It is also found that the patients who showed up for the second follow-up were in the age range 26 -45 years old, therefore women of reproductive age. However, a statistically significant correlation between these variables could not be identified ( $R^2 = 1.18$ ). Thus, we can conclude that the type of performed procedure and compliance to follow-up are not age-dependent (fig. 5).



**Fig. 5.** Correlation between patients' age and compliance with follow-up depending on the excision procedure

Of the patients attending the first post-interventional follow-up, only 24 had an HPV test: 12 patients had a positive HPV test result and the remaining 12 negative results, demonstrating a low compliance



**Fig. 6.** Correlation between patients' age and time to the first post-interventional follow-up

with performing these investigations. A positive finding is that over 50% of the patients who attended the follow-up had a normal colposcopic appearance of the cervix or had a cytological diagnosis negative for



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intraepithelial or malignant lesions; only 6 cases were diagnosed at the cytological re-examination with ASCUS, in 7 cases high-risk lesions were identified (CIN3, HSIL), and in two other patients the colposcopic examination revealed atypical vascularization. As for low-grade lesions, 16 patients who showed up at the first follow-up received a diagnosis of LSIL, CIN1 or CIN2. A single case was diagnosed with CIS 2

months after the first intervention. As for the colposcopies performed in the patients attending the first follow-up, in 15 patients the TZ was visible in different degrees (TZ1, TZ2, TZ3), and on examination the aceto-white epithelium (AWE) had a clear outline in 4 patients or was classified G1 or G2 in 9 patients. In 16 cases a chronic inflammatory process and in other 3 patients an incomplete post-conization scarring were found.

TABLE IV.  
**Diagnostic findings at the first follow-up**

<b>Diagnosis at first follow-up</b>	<b>Number of patients</b>	<b>Percentage</b>
<b>CIS</b>	1	0.49%
<b>HSIL</b>	7	3.46%
<b>LSIL</b>	16	7.92%
<b>ASCUS</b>	6	2.97%
<b>Inflammatory aspects</b>	16	7.92%
<b>NILM</b>	123	60.89%
<b>Colposcopic visualization ZT</b>	15	7.42%
<b>Colposcopic aspect EAA G1/G2</b>	13	6.43%
<b>Colposcopic atypical vascularization</b>	2	0.99%
<b>Other diseases</b>	3	1.48%

Only 66 (14.5%) patients showed up for the second post-interventional follow-up. Cytological examination revealed: 7 patients presented ASCUS features and 4 patients LSIL type lesions, 19 patients had a cytodiagnosis negative for intraepithelial lesions or malignancy (NILM), 14 patients had normal colonoscopic appearance, and 4 patients presented inflammatory aspects (tab. V). Other pathological findings were suggestive of various diagnoses, such as Naboth cysts (1 case), genital mycosis (1 case), TZ ectopy or TZ2 (5 cases), or bacillary flora (1 case). None of the followed-up patients developed CIS or invasive carcinoma.

There was only one case with HSIL (followed up at 2- and 4-months post intervention) which at the 7-month follow up was reclassified as LSIL.

Regarding the correlation between age and post-interventional follow-up, no direct correlation was found, low compliance with follow-up being independent of the patients' age. However, the fact that most patients showed up for follow-up 3 months after the interventional procedure, predominantly LLETZ, deserves mention. And so is the fact that the patients who did not miss the follow-up appointments were aged between 26 and 45 years, that is the reproductive years.

TABLE V.  
**Diagnostic findings at the first follow-up**

DIAGNOSIS AT FOLLOW-UP 2	NUMBER OF PATIENTS	PERCENTAGE
ASCUS+LSIL	11	16.6%
Inflammatory aspects	4	6.06%
NILM	19	28.78%
Normal colposcopic findings	14	21.21%
Other diseases	18	27.27%

## DISCUSSION

The study population included patients who required an excisional procedure, so the types of dysplastic lesions identified by cytology tests were in different percentages compared to the data in the literature on this topic. Thus, ASCUS-type lesions were identified in 20.4% of our patients compared to 4-5% in the literature. In our study LSIL lesions had an incidence of 30.3%, similar to that of 25-30% reported in the literature on this topic, while for HSIL lesions the incidence in our study was 32.3% versus 8% in the literature. As to carcinomas, the percentages are similar, 2% in our study vs 1% in the literature. These differences can also be explained by the profile of the study population, but at the same time they can raise a question mark regarding the increased incidence of cancer in the northeastern region of our country. The high percentage of severe lesions could be a predictive factor for the high rate of cervical cancer in our country.

Most initial HPV infections are believed to be cleared by the immune system. However, there are people in whom the HPV virus persists in a latent state in cells of the cervix, the latter becoming active through replication mechanisms, leading to dysplasia at a given time in the future. HPV induces pathological changes in the cervix

due to the effects of viral proteins expressed by the virus. These changes include basal cell proliferation, nuclear multiplication with hypernucleosis, koilocytosis and abnormal mitoses (4).

The natural history of HPV infection demonstrates that it is often transient in young women, most CIN I and II cases regressing within 2 years of primary infection (5,6). The age range in the population analyzed in our study was 19 - 62 years, average age 36 years, thus most of them socially active young women. We believe that analyzing if patients saw a doctor or general practitioner and the classification of cervical diseases by age group due to future different obstetric implications are an interesting addition. Regarding the first age group in our study, patients under 24 years of age, they accounted for only 9% of all study patients and did not require HPV testing because it is a group at high risk for infection but also for spontaneous remission within 2 years mainly due to individual immunological mechanisms. However, at least those women who are already sexually active should be screened for cervical cancer by cytology.

The age group 25-40 years accounted for 64% of the patients diagnosed with cervical dysplasia included in the study. If to these we add the age group under 24

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years we get a total of 73%, meaning that three quarters of the women who require consultations for dysplasia are of childbearing age, the obstetric prognosis being important and essential for this age category. Thus, in addition to preventing cervical cancer, preserving the obstetrical future of these patients has to be another main concern. These observations are in agreement with the data regarding the obstetric history of these patients, the statistical analysis showing that the number of births differs according to age, women aged between 19 and 40 years having more babies than those over 40. So, in women over 40 years, the obstetric prognosis is no longer a priority, the main concern being the prevention of cervical cancer. Also, we found that these women seek medical help less than the younger ones, accounting for only 27% of the examined patients.

The risk of HPV infection in a population increases sharply after the first intercourse. Cohort studies show that after a rapid increase in young women, the incidence of cervical HPV infection decreases with age. On one hand, this decrease is due to fewer sex partners and, on the other hand, to the specific immunity developed by some patients. Of the 203 patients assessed for HPV DNA in our study, 41.38% tested positive for HPV 16, 10.84% tested positive for HPV 18, and 32.02% tested positive for other HPV HR strains than the 2 above mentioned. Thus, the percentage of tests in which high-risk strains were detected exceeded 80%. Comparing the different age groups with positive HPV genotyping an upward trend can be observed, with a peak in high-risk viral DNA detection in patients up to 30 years of age, and a decrease after the age of 40. Fifty-one of the positive cases were up to 30 years old, the

result being mainly influenced by the increased rate of infections with high-risk strains at young ages, but also possibly due to an increased level of information of this age group or a particularly favorable socio-economic status. The age-related distribution of HPV is also supported by other studies such as that by Aro *et al.* who evaluated the distribution of HPV types in a cohort of 1,279 women with cervical dysplasia (7) and found that the distribution was distinctly polarized by age. So, in terms of incidence, HPV 16/18 proved to be more common in young women. Another study reported comparable results, HPV16/18-related cervical dysplasia being found in 64%, 58%, and 35% of women aged <30, 30–44, and ≥45, respectively(8). The results are somewhat similar to those obtained in our study.

In our study, excisional interventions were performed in 47.5% of the patients with CIN1 dysplasia, which is low-grade dysplasia, and in 25.3% of the patients with CIN2 dysplasia. This therapeutic approach was based on the following considerations: they were performed when the lesions did not regress spontaneously, in low grade lesions but worrisome for the examiner because they were extended, and also in patients who did not want or did not have the possibility (for various reasons) to comply with the specific follow-up essential to adopt a “wait and see” attitude, as in the case of those from a disadvantaged socio-economic environment. Added to these is the particular group of patients who migrate to other countries for a longer period of time, who do not benefit from medical insurance, and who request a quick solution.

It would have been interesting to study how the patients with cervical anomalies

were followed-up before making the decision to perform excisional interventions, knowing that a high percentage of the dysplastic lesions, especially the low-grade ones, are likely to regress spontaneously. Unfortunately, the vast majority of women included in the study saw a doctor because of a more or less specific symptomatology, rather than for a screening assessment. In these conditions, it is not surprising that the most common therapeutic attitude was conization or LLETZ. It is obvious that from the point of view of specific management, the actions taken, and the approach used in these cases are not in accordance with the international guidelines regarding the management of the cases in which small asymptomatic lesions are discovered.

A discrepancy can be noted between the compliance with follow-up of the patients in the northeastern region of Romania included in the study and that of patients with cervical dysplasia from other countries where a follow-up appointment is scheduled every 6 months in the first 2 years after diagnosis and annually thereafter, and includes pap smear, colposcopy, and colposcopic guided biopsy, of course if clinically indicated. As a rule, HPV testing is performed at the first examination after conization in patients with documented HPV infections. Persistence of HPV infection is defined as HPV identification at the first specific examination after conization (9). In this study, we failed to establish a correlation between the persistence of HPV infection and the degree of cervical lesions, since only 24 patients underwent HPV testing after the excisional procedure.

As to the follow-ups after excisional procedures conducted in the first 3 months after the intervention, it was found that a total of 202 patients showed up for the first

post-procedural follow-up, which accounted for over 50% of all examinations. Sixty-six of these patients showed up for a second follow-up, of which 43 patients in the first 6 months after the intervention. Age-related differences in the low compliance with follow-up were noticed, this aspect not only influencing the visits to a doctor, but also the subsequent follow-up. Although most initial infections in young women are generally eradicated, there are patients in whom the HPV virus persists in the cervical cells in a latent state, the latter becoming active through replication mechanisms leading to dysplasia at a certain time in the future (4). In young women, most CIN 1 and CIN 2 cases regress within 2 years, the treatment and excision being recommended only when CIN2 persists for over 2 years on biopsy according to the guidelines. Regarding the CIN3 lesions, a small percentage will regress, but most of them will progress to cervical cancer. Current recommendations support that CIN3 in young women under 24 years to be managed as is adult women, excisional procedures being recommended (10). Although these new recommendations are medically reasonable, it is important to consider the issue of compliance. Unfortunately, a substantial number of patients is lost to follow-up as shown by our study, where less than half of the patients who underwent excisional procedures showed up for follow-up. Delayed follow-up may result in a delayed initiation of the specific treatment. Data in the literature demonstrate low compliance at 2-year follow-up even after an excisional procedure such as a LEEP (11, 12). Failure to comply with the follow-up protocol was associated with young age, lower education level, unmarried status, inadequate health insurance, better overall health, low grade

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cervical lesions, as well as the lack of understanding the severity of their condition (13). Women with a low socioeconomic status and those from minority communities were identified as less likely to accept cervical cytology as a screening method. Furthermore, compliance with follow-up in these cases is much lower after an abnormal result. The absence of monitoring can substantially contribute to an increase in the incidence of invasive cervical carcinoma, this increased incidence already being reported among poor and minority women (14, 15). For all these reasons we believe that delaying additional testing for more than 12 months in patients with abnormal cytology may contribute to a delay in diagnosis and management.

Our study shows that there are no differences regarding patients' compliance with follow-up regardless of disease severity or the performance of an excisional procedure. This lack of compliance with follow-up in the analyzed population may be explained by their low socioeconomic status and increased population exodus. According to the National Institute of Statistics, "Romania continues to be a country of emigration, the phenomenon of emigration being the second main cause of the declining population. The international migration balance of Romania in 2021 was negative" (16). The number of Romanians living abroad is around 5.7 million according to the data of the Ministry of Foreign Affairs, accounting for almost 30% of the country's population (17). This problem explains the sometimes slightly more aggressive therapeutic approach compared to international guidelines. The low compliance with follow-up and the fact that some patients showed up for the first follow-up after the interventional procedure at 6 or 12 months

did not allow a relevant correlation of cytodiagnosis in dynamics. Based on the results of the first follow-up after the excisional procedure we can report that 11 patients presented low-grade lesions (ASCUS+LSIL) and HPV HR infection, the group being too small for a statistically significant result. It can be noted that after a longer time interval, over 12 months, no residual high-grade lesions were observed. The literature supports the fact that the persistence of high-risk HPV infection (both HPV16/18 or other HPV HR strains) was correlated with an increased risk of recurrence of CIN 2-3 cervical lesions. Persistence of HPV infection is associated with recurrence, much more than margin status after excisional procedure, being one of the main factors predictable of relapse. This is also supported by Zhang *et al.* (18) who analyzed the data of over 500 patients who underwent conization and observed that positive margins after the excisional cause a 6-fold increase in the risk of recurrence, while the persistence of HPV HR infection cause to a 20-fold increase in the risk of recurrent disease. Considering these results, it is imperative to implement co-testing both for the detection of cervical lesions and for their monitoring (9).

A possible solution could be an organized screening and a much more effective vaccination campaign, which could increase patients' compliance with health services, being able to address this condition before the onset of clinical symptoms and thus minimizing the interventional procedures. In this respect, better ways to inform the population and the mobilization of the target population, the involvement of several health professionals, mobile screening units facilitating access to healthcare services, treatment optimization and fol-

low-up systematization, as well as the establishment of a national registration and tracking register are needed (19). Measures to inform and educate the population about this disease also necessary. Increasing the degree of awareness of this disease severity would increase compliance with follow-up. Primary prevention by vaccination against HPV virus was initiated in 2008 but was eventually abandoned because of parental refusal of vaccines. This opposition is found even in medical circles and even in cancer services. For convincing the parents about the value and safety of vaccination it is still essential that professionals to send a clear message and inspire through the power of example. Secondary prevention through a systematic screening program was initially implemented in 2012 and abandoned subsequently; a new campaign with mobile units was resumed in 2020, the target population being women aged of 25 to 65 years, which is approximately 4.2 million women in Romania. The large sample size is the main strength of the present study, and the most important limitation was the inclusion of a large group of patients who were not tested for HPV. Although being a limitation it reflects the current practice in a “real life” setting. Low compliance with follow-up also reflects a current practice, being a favorable factor for the increased incidence of cervical cancer in our country.

## CONCLUSIONS

Although the etiopathogenesis of cervical cancer is strongly associated with HPV infection, not all lesions will undergo malignant transformation as cases of spontaneous remission were demonstrated under adequate monitoring. Cervical cancer is the neoplasia with the most effective preven-

tive methods including vaccination and primary or secondary screening, thus making possible the early detection of precancerous lesions. The types of dysplastic lesions identified by cytological examination are in similar percentages with those reported in the literature, except for the diagnosis of HSIL which is the most frequently identified. This increased rate could be the basis for the increased incidence of cervical cancer in our country. Conization and excisional procedures represent a valuable method of conservative treatment while preserving the patient’s fertility and avoiding a radical intervention.

The low compliance with follow-up among the patients in this region of the Romania and the increased population migration are two particular elements that lead to the use of excisional procedures even in the case of low-grade lesions in an attempt to reduce the incidence of cervical cancer. This low compliance to follow-up and the absence of vaccination can justify the fact that Romania ranks first in Europe in terms of the incidence of cervical cancer. The risk of recurrence identified after excisional procedures was low, being correlated with the persistence of HPV infection. Improving the organized screening programs and a more effective vaccination campaign could increase patients’ compliance with public health services, being able to deal with this condition before symptoms arise thus minimizing the interventional procedures and progression to cervical cancer.

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**Types of dysplastic lesions and compliance with follow up in Northeastern Romania  
– a retrospective study**

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