CYTOLOGICAL DIAGNOSIS IN CERVICAL CANCER

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Abstract. Aim. The cytological test has multiple valences, allowing the early discovery and location of feminine genital cancer. **Material and methods**. In the period of time between 2001 and 2009, the study made within the Obstetrics and Gynecology Department of „Sf. Apostol Andrei” Emergency Hospital in Galaţi, revealed that from 415 cases with a changed PAP smear, the cytological diagnosis showed cancerous and pre-cancerous lesions in 53 patients (12.8%). We harvested cytological smears for most cases we studied and we even performed biopsies of the cervix where there was an indication to do that. The cytological smears were coloured using Giemsa technique or the method with hematoxylin-eosin (H-E). **Results.** The cancerous and pre-cancerous stage was identified in 12.8% of the cases we analyzed, as follows: L-SIL 1,9%; H-SIN 8,0%; CIN 2,9%. **Conclusion.** The cytological examination proved to have exceptional qualities as a screening method, becoming lately an accurate method for early detection of cervical cancer.

INTRODUCTION

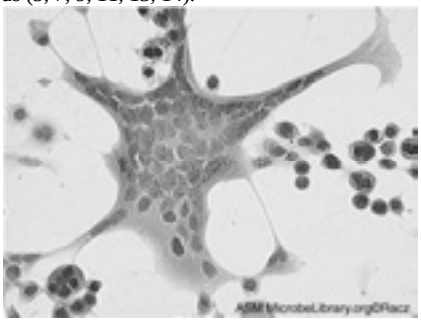
Cervical cancer is a very serious chronic disease with great importance from the social and medical point of view, that has a severe evolution when detected in advanced stages, being one of the most complex and difficult problems of human pathology.

The present concepts regarding carcinogenesis bring us to the idea that malignant tumours appear and develop as a consequence of certain events that weaken or overpass the immunologic competence of the body which allow in a critical moment the development of atypical cells. They can be induced by a virus, a physical or chemical factor or by products that suffer a spontaneous somatic mutation (3, 10, 12).

If the immune system were competent, the body would constantly destroy small groups of cancer cells if they were generated by the above mentioned agents. The defence mechanism is mainly cellular and belongs to immuno-competent „T” lymphocyte cells.

Here are the proofs for the immunologic response of the host when the tumour cells appear: spontaneous tumour regressions, as they were also demonstrated in the dynamic study of the invasive cervical neoplasias (ICN), proving the existence of tumour antigens and specific antibodies in the cancer patient and also on an experiment in which an animal was immunized, the rejection of the cancer cells grafts in healthy animals and the inducing of a tumour immunity. The other way round, the fact that tumours appear as a consequence of an immunologic deficit of the host is proven by the more frequent presence of malignant tumours in people with immunity diseases (for example HIV infection), in the cases where people received transplants - cases which were given radiation therapy and immunosuppressive medication for tolerating them, and also in elderly patients when their immune defence capacity is a lot reduced (6).

There are a lot of works that link different epidemiologic factors with dysplasia occurrence, their conversion into cancer *in situ* (CIS) and the latter into invasive cancer (IC). It gets clearer and clearer that in the *etiopathogenesis of the cervical neoplasia and of the invasive carcinoma, the key event is the sexual intercourse* that is the means for transmitting a carcinogenic agent, considered to be Herpes simplex virus (HVS-2) only two decades ago, and now certain types of papilloma-virus (5, 7, 9, 11, 13, 14).



Picture 1. Herpes simplex virus
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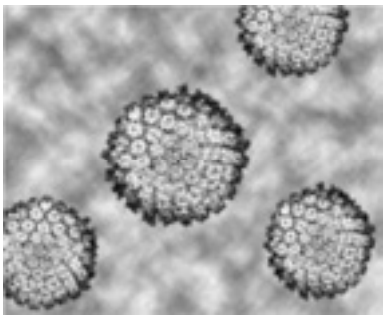

Picture 2. Papilloma virus
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The present research proves that the development of malignant tumours can be the consequence of some events that lower or overpass the immunologic competence of the body and which allow in a certain moment atypical cells to appear and multiply. These cells may be induced by a virus, a physical or chemical factor or produced by a spontaneous somatic mutation. When the immune system works properly the body is able to destroy small groups of cancer cells that are generated by cancerigenic agents (4, 14).

MATERIAL AND METHODS

In the period between 2001and 2009 the Department of Obstetrics and Gynecology of „Sf. Apostol Andrei" Emergency Hospital in Galati registered a number of 415 women with cervical neoplasia, 53 cases of which showed pre-cancerous and cancerous lesions after the cytological diagnosis.

We used both Babeş-Papanicoalou nomenclature and also TBS (The Bethesda System) when interpreting and diagnosing the cytological smears.

The activity of early detection of cervical cancer is not very well organized yet and that is why we can see that in today's practice most cases come to the doctor in relatively advanced stages that need complex and laborious interventions and usually have unsatisfying results.

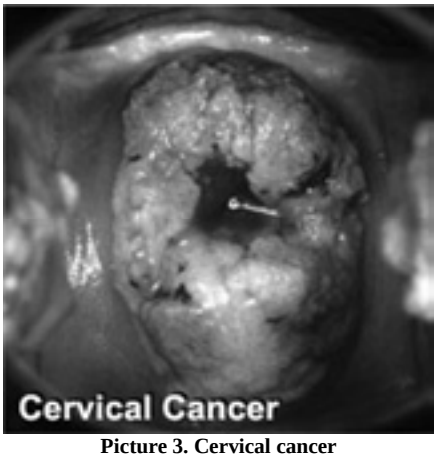
The vaginal cytological examination has reached such a high degree of accuracy in detecting cervical cancer. It represents a valuable method of detection due to the following qualities (1, 2, 13, 14):

- It is a simple, nontraumatic and easy to perform examination technique that is easily accepted by the woman who is being examined due to its non-invasive character;
- It allows the doctor to detect the disease in incipient stages (micro invasive carcinoma) and also the pre-cancerous lesions whose treatment and detection represent the primary prophylaxis of the disease, especially when associated with the colposcopic methods;
- It has a high degree of accuracy (sensitivity and specificity), that can reach 90-95%;
- It has an unlimited theoretical applicability, being able to basically cover the whole feminine population at risk, which confers it a great value and indicates it as a mass screening method;
- It can be repeated over a long period in a woman's life.

We used a great variety of studies that included colpo-microscopy, electronic microscopy, cultures of cells and autoradiography in order to establish the origin of an evolution mechanism of CIN.

CIN starts in the transformation area and spreads along the basal membrane, replacing the adjacent squamous and glandular epithelial cells. These methods showed that the neoplasic process begins in only one cell or in a very small group of cells.

The lesion expands by cloning, becomes less differentiated and extends into the endocervical canal. The election zone of the intraepithelial lesions is limited by their border with the native squamous epithelium of the exo-cervix, but proximally the lesions can extend variably in the endocervical canal and even in perimeter and the fallopian tubes and rarely in the peritoneal area. CIN appears twice as frequent on the anterior lip of the cervix as on the posterior lip and very rarely on the side. CIN is characterized by abnormal cellular proliferation and maturation and an atypical nucleus. Proliferation begins in the basal and parabasal layer with an increase in the number of parabasal immature cells that spread towards the intermediate and superficial layers.

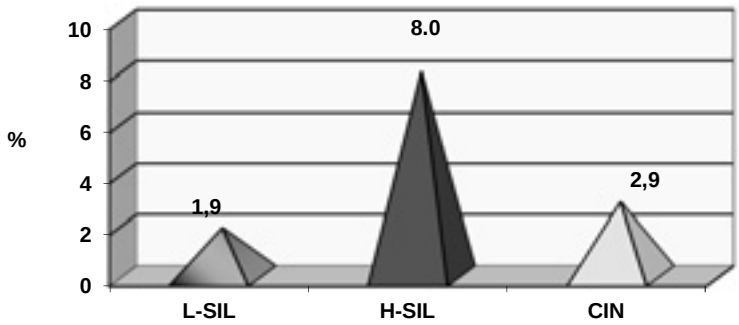


Picture 3. Cervical cancer
(source: http://embryology.med.unsw.edu.au/wwwhuman/MCycle/images/cervicalcancer.jpg)

Statistical processing. The data was expressed in a form that allows it to be included in certain categories; a variable will not be registered under more than one form; the data will be grouped on categories of variables. The data was centralized in EXCEL and SPSS data bases and processed with the appropriate statistical functions.

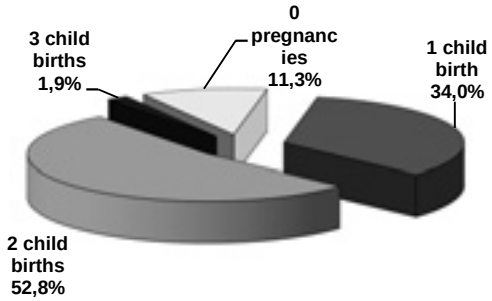
RESULTS AND DISCUSSION

We harvested cytological smears from the vaginal secretion for 53,10% of the patients and we did not perform the same action for the rest of 46,89% because they have already been diagnosed by biopsy or other services or the smear could not be interpreted because of hemorrhage, infection or necrosis.



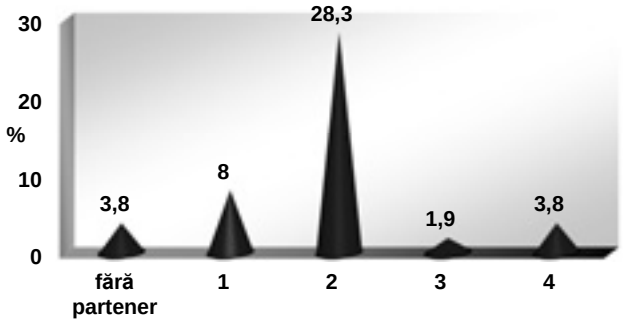
Picture 4. Distribution of cytologically confirmed neoplasias depending on the lesion types

The association of HVS-2 infection with CIN and IC was more frequent if the virotic infection occurred during pregnancy (88,7%).



Picture 5. Correlation of virotic infection with pregnancy

34% of the patients with cytologically confirmed cervical neoplasia had multiple sexual partners.



Picture 6. Distribution of cases depending on the number of sexual partners

Virotic cervicitis, especially the ones with genital herpes (HVS-2) and papilloma viruses, are especially incriminated in the genesis of cervical neoplasias. After lots of research the HPV was discovered to be involved in CIN and IC etiology (7, 9).

35 types of HPV were described for the human, which were classified after the sequence of the nucleotides in the viral DNA by using recombining methods (Southern Blot Hybridisation). The types 16, 18, 31 and 35 are oncogene. About 1-2% of the women who are sexually active have a cervical infection with (7, 14).

Branca (1995) links the degree of risk of HPV cervicitis with HIV association, 47% of the women who were HIV positive also had HPV lesions that were found out during the colposcopic and/or cytological examination, and 40% of them had CIN I, CIN II and CIN III, 37% of the cases with CIN I and CIN II progressed rapidly towards CIN III. For the HIV negative cases only 23% had HPV lesions, and 26% of them were associated with CIN I, CIN II or CIN III; only one case had a fast evolution (after a year) from CIN I to CIN III.

Remmink (1995) monitors for 3-4 months a lot of women by performing cervical cytology, colposcopy and HPV tests. The cases with progressive lesions of CIN were HPV positive all the time, and the biopsy confirmed CIN III stage. The author appreciates that HPV effect on the progression of CIN lesions is also influenced by other risk factors (age, the number of sexual partners, smoking etc.).

The present stage of research creates premises for separating the CIN cases with an irreversible potential of evolving towards malignancy from the ones with a benign evolution. The infections with oncogene types would necessitate a more aggressive treatment with the purpose of blocking the progression towards CN types with a high malignancy (3, 4).

The cervicitis with genital herpes virus (HVS 2) was incriminated as a risk factor for producing dysplasias and cervical cancers.

The serologic screening shows that the incidence of HVS-2 infection is 10% for the women who are apparently healthy and 20% for those with various gynecologic diseases. The percentage of positive results for cervical cultures is 1-2%. The incidence varies between 5% for the young women under 19 who are sexually active and 27% for women over 23 years old. The incidence becomes 10 times higher if the number of sexual partners is 4 or even more (9).

Josey (quoted by Rădulescu, 1995) finds frequent associations of HVS-2 infection with HPV or trichomonas, gonococcus, mycoplasma or chlamydia. The same author (Josey) used a serologic method of identifying the HVS-2 infection and proved the presence of this infection in 56% of the

women with dysplasia, 70% of those with CIS (carcinoma *in situ*) and 83% of the women with IC (invasive carcinoma) when compared to a witness lot where the infection was found only in 24% of the cases.

CONCLUSIONS

Cervical cancer is a very serious chronic disease which has a great importance from the medical and social point of view with a severe evolution when it is detected in advanced stages, being one of the most complex and difficult problems of feminine pathology.

The cytological examination represented a „revolution” for the process of early detection and diagnosis of cervical cancer.

The cancerous and pre-cancerous stage was identified in 12,8% of the cases we analyzed, as follows: L-SIL 1,9%; H-SIN 8,0%; CIN 2,9%.

The association of HVS-2 infection with CIN and IC was more frequent of the virotic infection if it occurred during pregnancy, suggesting the intervention of an eventual hormonal factor or of the immuno-depression associated with pregnancy.

The existence of multiple sexual partners was considered an associated risk factor.

REFERENCES

Austin RM. 1997. Managing risk in gynecologic cytology: reactive and unsatisfactory smears. *Cancer*, 81(3): 137-138.

Bailie R. 1996. An Economic appraisal of a mobile cervical cytology screening service. *South African Medical Journal*, 86 (9 Suppl): 1179-84.

Chen CA, Liu CY, Chou HH, et al. 2006. The distribution and differential risks of human papillomavirus genotypes in cervical preinvasive lesions: A Taiwan Cooperative Oncologic Group Study, *Int J Gynecol Cancer*. 16(5): 1801-1808.

Crauciuc E, Pricop Fl, Masheh O. 1999. The value of cytological examinations in diagnosing the cervical cancer. *The British Journal of Family Planning (Ed. Lb. Rom.)*, 4(1): 3-5.

Epstein E, Jamei B, Lindqvist P. 2006. High risk of cervical pathology among women with postmenopausal bleeding and endometrium: long-term follow-up results, *Acta Obstet Gynecol Scand, Department Of Obstetrics And Gynaecology, Lund University Hospital, Lund, Sweden*, 85(11): 1368-1374.

Gilks Cb, Young Rh, Gersell Dj, Clement P. 1997. Large cell carcinoma of the uterine cervix: a clinicopathologic study of 12 cases. *American Journal of Surgical Pathology*, 21(8): 905-914.

Hall S, Lorincz A, Shah F, et al. 1996. Human papillomavirus DNA detection in cervical specimens by hybrid capture: correlation with cytological and histological diagnoses of squamous intraepithelial lesions of the cervix. *Gynecologic Oncology*, 62(2), 353-359.

Herbert A. 2006. Liquid-based versus conventional cervical cytology, *The Lancet* - Vol. 368, Issue 9530, pg. 118.

Janerich DT, Hadjimichael O. 1995. The screening histories of women with invasive cervical cancer. *Connecticut. American Journal Of Public Health*, 86(6): 791-4.

Katz IT, Wright AA. 2006. Preventing cervical cancer in the developing world, *N Engl J Med* 2006; 354: 1110.

Lyng H, Brovig RS, Svendsrud DH, et al. 2006. Gene expressions and copy numbers associated with metastatic phenotypes of uterine cervical cancer, *BMC Genomics*. 7: 268.

Mitchell H, Hocking J, Saville M. 2006. Temporal characteristics of laboratory screening errors in cervical cytology. *Acta Cytol*. 50(5): 492-8, Victorian Cytology Service, Carlton South.

Moss Sue, Alastair Gray, Rosa Legood, et al: Liquid based cytology/human papillomavirus cervical pilot studies group - effect of testing for human papillomavirus as a triage during screening for cervical cancer: observational before and after study, BMJ, Jan 2006;

Wied GL, Keebler M: Compendium on diagnostic cytology. Techniques for the collection and preparation of cytological specimens from the female reproductive tract. 7th Ed. Tutorials of Cytology, Chicago, Illinois, 1992, 35-39.

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