CHLAMYDIA TRACHOMATIS AND CERVICAL INTRAEPITHELIAL NEOPLASIA

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Summary: 57 patients with viral condylomata of the uterine cervix and/or cervical intraepithelial neoplasia (CIN) were examined evaluating the presence of Chlamydia Trachomatis (Ct) cervical infection and serum IgG anti-Ct. The prevalence of Ct cervical infection was globally 7.22%, in CIN 3 group the rate was higher (16.67%). In addition, in this group the rate of IgG anti-Ct was significantly higher (66.67% vs 11.76%; $p = 0.01$) in comparison to the whole group. The role of the association between Human Papillomaviruses and Ct infection was finally discussed.

INTRODUCTION

Epidemiological data indicate that cervix carcinoma behaves like a sexually transmitted disease. The search for a transmissible agent, responsible for the neoplastic transformation has given disappointing results. The pathogenetic role of bacteria and protozoa has been excluded from previous studies and attention has been focused on the role of the viruses, whose mutagen potentiality is well known ([7]).

The mutagen activity of Herpes Simplex Virus (HSV) ([2]) and the presence of Rna ([3]) and Dna-HSV2 fragments integrated in the genoma of the neoplastic cells of cervical carcinoma ([4]), were the first inconstant observations on the presumed pathogenetic role of HSV2.

Successively the identification of Human Papillomaviruses (HPV) integrated in the genoma of epithelial cells in the majority of the cases of cervix carcinoma, with variable percentages in relation to the virus type ([5, 6, 7, 8]), has pointed out the possible causal role of the HPV-CIN association ([9]).

One possible hypothesis is that HPV may be a promoting factor of neoplastic transformation, after an HSV infection (initiating factor) with mutagen potentiality ([10]).

This view has been confirmed by other studies ([11, 12]) but the importance of HSV-HPV synergism was reduced by the variability of HSV2 infection in women with cervical cancer in different geographic areas ([11]).

For these reasons it has been suggested that other factors (chemical, physical, biological) apart from HSV could be responsible for neoplastic transformation through a mutagen event with a " hit and rund " mechanism ([13, 14]).

In this context the association between Chlamydia Trachomatis infection and cervical atypia could be important. We have evaluated the prevalence of past or present Chlamydia Trachomatis infection in a group of patients with atypical cervical lesions.

PATIENT AND METHODS

The population study comprised 57 outpatients attending the Diagnostic Center of Genital Neoplasia of the Department of Obstetrics and Gynecology of the University of Pavia.

Only patients with cytological and/or colposcopical lesions suggesting HPV infection or CIN were admitted to the study.

Each patient was investigated with detection of Chlamydia Trachomatis, titration of serum IgG anti-Ct and biopsy of the cervical lesion.

The direct immunofluorescent test (Micro-Track - Syva) was used to identify Chlamydia Trachomatis infection.

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Endocervical scrapings were fixed on apposite slides with acetone and air drying. Successively the specimens were stained with a reagent containing monoclonal murine antibodies (directed against external membrane proteins of elementary and reticulate bodies of CT) labelled with fluorescine isothiocyanate.

An indirect immuno-peroxidase test (Ipazime Chlamydia - Indirect Peroxidase Assay - Savyon diagnostic) was used for the detection and titration of IgG-anti-Ct.

Chlamydia Trachomatis infected cells (serotype L2) were used as antigens. The serum of the patient was absorbed with infected cells and the presence of IgG-anti-Ct was revealed by anti-IgG antibodies coniugated with radish peroxidase (a blue color precipitate in the wells on microscopic examination).

The biopsy specimens were stained with ematoxilin-eosin and evaluated for the presence of lesions suggesting HPV infection (cervical condylomas) [17, 18]. Cervical atypias were evaluated on the basis of CIN definition.

Goodness-of-fit chi-square between observed and expected frequencies was used for the statistical analysis of data.

RESULTS

The group we studied comprised 57 patients (mean age 32.17; range 20 to 45 years). In 50.88% (29/57) of cases Pap smear did not reveal dysplastic and/or viral cervical lesions, in 49.12% a koilocytotic or CIN picture was present (tab. 1). The positive predictive value (positive test and presence of disease) of Pap smear for koilocytosis and CIN was 100% and 43% respectively. The negative predictive value (negative test and absence of disease) was less than 10% for koilocytosis and 100% for CIN.

Table 1. – *Correlation between the results of target biopsies and Pap smears.*

	Biopsy					
	Negative	Condyloma	CIN 1	CIN 2	CIN 3	Tot.
Cytology						(%)
Negative	–	29	–	–	–	29 (50.88)
Koilocytosis	–	4	–	–	–	4 (7.02)
CIN 1	–	11	4	–	–	15 (26.31)
CIN 2	–	1	1	1	2	5 (8.77)
CIN 3	–	–	–	–	4	4 (7.02)
Total (%)	–	45 (78.95)	5 (8.77)	1 (1.75)	6 (10.53)	57

Table 2. – *Cervical Chlamydia Trachomatis infection and rate of IgG anti-Ct in relation to the results of the target biopsies.*

Biopsy	IgG Ct+ %	IgG Ct –	Ct+ %	Ct –	Tot.
Negative	–	–	–	–	–
Condyloma	5 (11.11)	40	3 (6.67)	42	45
CIN 1	1 (20.00)	4	–	5	5
CIN 2	–	1	–	1	1
CIN 3	4 (66.67)*	2	1 (16.67)	5	6
Total (%)	10 (17.54)	47 (82.46)	4 (7.22)	53 (92.98)	57

* $p = 0.01$

A histological diagnosis of cervical condylomas and CIN was obtained in 78.95% (45/57) of the cases. In the remaining 12 cases (21.15%) the diagnosis was CIN.

Endocervical scrapings revealed Chlamydia Trachomatis infection in 4 patients (7.22%).

Serum IgG anti-Ct levels were significantly elevated (1:64) in 17.54% of patients (10/57) tab. 2). The prevalence of IgG anti-Ct was significantly ($p = 0.01$) higher in CIN 3 group in comparison to condilomata or CIN 1 group.

Figure 1 shows the distribution of histological lesions, serological data and endocervical C. Trachomatis identification in relation to the patient's age. Cervical intraepithelial neoplasia has a bimodal distribution and the prevalence of IgG anti-Ct is higher in the 31-35 year old group.

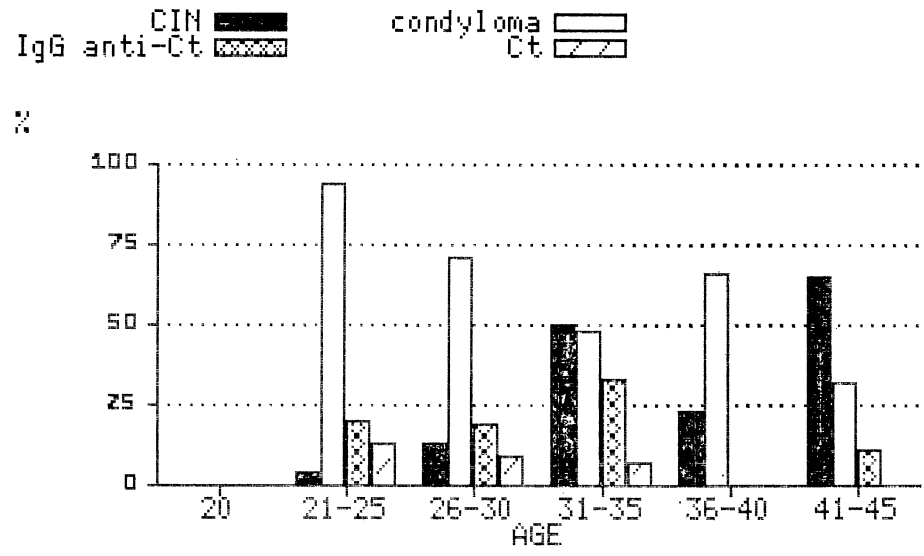

Fig. 1. — Age distribution of CIN, condylomata, Chlamydia Trachomatis cervical infection and serum IgG-anti-Ct.

DISCUSSION

Our data results indicate that cytology alone is an inadequate method in diagnostic screening of HPV lesions. The absence of koylocitosis in Pap smears does not exclude the presence of infection, however the reliability of cytology is higher in the diagnosis of CIN. For these reasons, colposcopy and target biopsies are necessary for the diagnosis of cervical condylomata. Colposcopy could not be considered as a screening procedure. It has been suggested that the Schiller test after cervical scraping may be an helpful method for the selection of the lesions to be colposcopically examined ([19]).

Cytology alone may over-estimate the severity of the lesions in presence of HPV infection (tab. 1).

In this population study the prevalence of Ct cervical infection (7.22%) is similar to the rate (6.12%) found among women attending our Sexually Transmitted Disease (STD) outpatients service ([20]), and reflects the high risk of STD of these women.

The significance of the higher rate (66.67%) of IgG anti-Ct in CIN 3 group is uncertain. In other studies ([14, 15, 21]) the prevalence of IgG anti-Ct was higher with respect to anti-HSV2 antibodies in patients with cervical atypia.

Epidemiologically Ct cervicitis is more frequent in the age group below 30 years; on the contrary the prevalence of IgG anti-Ct (previous infection) is greatest in the 31-35 year old group contemporarely to the higher rate of CIN.

This temporary relationship between Ct infection and CIN may be just a casual pattern due to the fact that Chlamydial cervicitis and cervical carcinoma precursors behave like sexually transmitted diseases. However, Ct infection is frequently coupled with HPV infection in the younger patients. Although long term effects of acute or persistent Ct infection are not known, its carcinogenetic or cocarcinogenetic role can not be excluded ([22]).

If a "hit and run" mechanism ([14]) has been postulated for the oncogenic potential of HSV and a synergistic interaction between HSV and HPV infection has been hypothesized ([10]), the same could be true for Chlamydial cervicitis, in this case only serology can attest to previous infection.

One objection to be raised to this hypothesis concerns the infection site; HPV is located in the transformation zone or in mature squamous epithelium, Chlamydia trachomatis is confined to the endocervical epithelium. Recently we have observed a contemporary HPV and Ct vaginal infection in a patient with a vaginal intraepithelial neoplasia (VAIN 2). This demonstrates that the pattern of endocervical Ct infection is not obligatory, raising further doubts and opening new perspectives for future studies.

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