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Outcome of expectant management of cervical intraepithelial neoplasia grade 2 in women followed for 12 months☆

Michelle G. Discacciati [a,b], Carlos André S. de Souza [a,b], Maria Gabriela d'Otavianno [a,b], Liliana A.L. Ângelo-Andrade [a,d], Maria Cristina A. Westin [a,b], Silvia H. Rabelo-Santos [a,c], Luiz C. Zeferino [a,b,*]

[a] Woman's Hospital Prof Dr Jose Aristodemo Pinotti-CAISM, State University of Campinas (UNICAMP), Campinas, São Paulo, Brazil
[b] Departament of Obstetrics and Gynecology, School of Medical Sciences, State University of Campinas (UNICAMP), Campinas, São Paulo, Brazil
[c] School of Pharmacy, Federal University of Goiás, Goiânia, Goías, Brazil
[d] Departament of Pathology, State University of Campinas (UNICAMP), Campinas, São Paulo, Brazil

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ABSTRACT

Objective: To evaluate the outcome of CIN 2 diagnosed by colposcopy-directed biopsy in women followed without treatment for 12 months and to verify whether the regression and progression of this lesion are associated with the woman's age at diagnosis and age at first sexual intercourse.
Study design: Women diagnosed with CIN 2 by biopsy and with previous cervical smear showing LSIL were included in this cohort study and followed up for one year with cervical smear and colposcopy every three months. The rates of progression, persistence and regression of the CIN 2 were evaluated. The Kruskal–Wallis test was used to analyze the woman's age at diagnosis, age at first sexual intercourse and interval since the first sexual intercourse according to the CIN 2 outcome, assuming a significance level of 5%.
Results: At the end of 12 months of follow-up the CIN 2 regression rate was 74% (31/42), progression rate to CIN 3 was 24% (10/42) and in one case CIN 2 persisted (2%). Among women who had regression, this event was detected in the first six months of follow-up in 26 of the 31 cases. There was no statistically significant association between the evolution of CIN 2 and the woman's age at diagnosis, age at first sexual intercourse and interval since first sexual intercourse. Women whose lesions were restricted to one quadrant were more likely to have CIN 2 regression at three-month follow-up compared with women with a lesion extending to one or more quadrants (OR: 6.50; 95% CI: 1.20–35.23).
Conclusions: The results of this study indicate that the majority of CIN 2 diagnosed by biopsy in women with previous Pap smear showing LSIL will regress in 12 months and therefore an expectant approach could be considered in these cases, not only for young women. Nevertheless these findings are not conclusive, and larger studies are required in order to certify when it is safe to adopt expectant management for CIN 2.

1. Introduction

Cervical cancer and its precursor lesion, the cervical intraepithelial neoplasias (CINs) are induced by human papillomavirus (HPV). It is known that a significant percentage of CIN regresses spontaneously [1]. CIN 2 can show similar behavior to CIN 1 or CIN 3. Even when infected by high risk HPV, if there is predominance of the productive pattern, CIN 2 evolution would be similar to that of CIN 1. In these productive infections, cell proliferation and episomal maintenance in the lower epithelial layers are followed by genome amplification and the expression of capsid proteins. Conversely, some CIN 2 lesions and CIN 3 lesions, exhibit a dramatic topographical change in viral gene expression, which includes an increase in E6/E7 expression in atypical proliferating cells [2–4]. Since CIN 2 occurs more frequently in young women [5], a more conservative approach may be considered, similar to CIN 1.

In this way, the last consensus of the American Society for Colposcopy and Cervical Pathology (ASCCP) [6] recommends that adolescent and young women with CIN 2 confirmed by biopsy and satisfactory colposcopy can have, rather than immediate treatment, expectant management and follow-up every six months with colposcopy and a cervical smear for a period of 24 months. For

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* Corresponding author at: Rua Alexander Fleming, 101, Cidade Universitária Zeferino Vaz, 13083-881 Campinas, SP, Brazil. Tel.: +55 19 35219466; fax: +55 19 35219424.
E-mail addresses: zeferino@fcm.unicamp.br, zeferino@hc.unicamp.br (L.C. Zeferino).

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older women with CIN 2, both surgical excision and ablation of the lesion are acceptable treatment [6].

Thus the objective of this study was to evaluate the outcome of expectant management of CIN 2 diagnosed by colposcopy-directed biopsy in women followed by cervical smear and colposcopy every three months for 12 months. Considering that the clearance rate of HPV infection is higher in younger women, and the persistence of this infection appears to increase with age [7–10], and considering that the early onset of sexual intercourse is a co-factor of the risk for cervical cancer [11,12], we decided to evaluate the association of woman's age at diagnosis and age of first sexual intercourse with CIN 2 progression and regression rates after 12 months' follow-up.

2. Materials and methods

2.1. Study design and ethical aspects

A cohort study was carried out between January 2005 and December 2008 at the Womańs Hospital Prof Dr Jose Aristodemo Pinotti-CAISM, State University of Campinas (UNICAMP), São Paulo, Brazil. This study derived from a study previously conducted in our department to evaluate the management of CIN 1 and was approved by the Institutional Review Board (IRB) of the Faculty of Medical Sciences of UNICAMP. For the object of this study, the IRB of the medical center recommended that the selection criteria should consider only women with previous low grade squamous intraepithelial lesion (LSIL), but not high grade squamous intraepithelial lesion (HSIL). The selection of women followed strict criteria to ensure that none of them suffered adverse effects due to CIN 2 treatment delay.

2.2. Selection and follow-up of women

Between January 2005 and December 2008, 4732 women with a cervical smear showing LSIL, diagnosed according Bethesda System [13], were invited to participate in this study, through letters sent together with the reports of the cervical smear. Out of these women, 1584 agreed to participate and were admitted to the cervical pathology clinic (Fig. 1). All women had a clinical evaluation, and underwent to a second cervical smear, a colposcopic examination and biopsy of suspicious image. The cervix was divided into four quadrants. The extent of the lesion was recorded according to the number of involved quadrants on the cervix and biopsy guided by the worst-appearing.

Women were considered eligible for the study if they fulfilled the following criteria: (1) the second cervical smear showed atypical squamous cells of undetermined significance (ASC-US) or LSIL; (2) histological diagnosis of CIN 2 [14]; (3) lesion completely visualized by colposcopy and squamocolumnar junction totally visible; (4) not pregnant; (5) showing no evidence of any immunodeficiency diseases; (6) no history of previous neoplasms; and (7) having a fixed address and at least able to provide a fixed telephone number.

Fifty women who fulfilled these criteria agreed to participate in the study and signed the informed consent. These women were followed during one year with cervical smear and colposcopy every three months. Women who showed a worsening of the suspicious image at colposcopy in relation to the previous examination were submitted to biopsy. When the biopsy revealed CIN 2 or a less severe result, the woman was maintained in the follow-up plan. When the biopsy revealed CIN 3, immediate treatment by excision of the lesion was performed. At one year of follow-up, all the women who still showed cytological or colposcopy abnormalities were submitted to a complete diagnostic evaluation and were all treated by excision of the lesion for additional histological analysis.

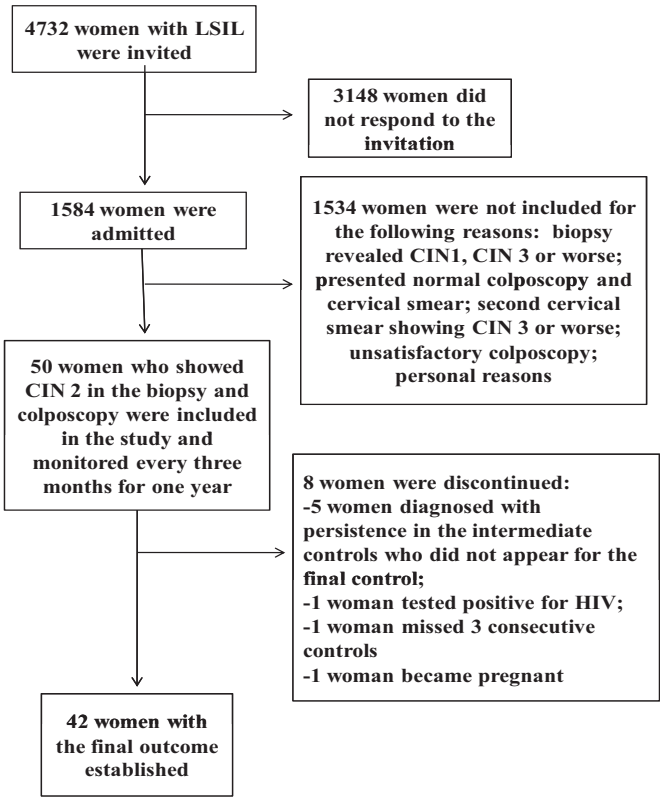

Fig. 1. Events related to the selection of the women.

2.3. Outcome of CIN 2 follow-up

The final outcome of CIN 2 was classified as regression, persistence or progression according to the following criteria:

- Progression: biopsy showing CIN 3 detected at any time during the follow-up. No lesion worse than CIN 3 was revealed.
- Persistence: biopsy showing CIN 1 or CIN 2 at the twelve-month follow-up.
- Complete regression: cervical smear, colposcopy and biopsies without neoplasia observed at any time during the follow-up and confirmed at the twelve-month follow-up.

At the third, sixth and ninth month follow-up, persistence was assigned for no changes of the colposcopy image and/or the cervical smear revealing ASC-US, LSIL or HSIL, specifically CIN 2 [15]. When the diagnosis of HSIL was classified as CIN 3, women underwent colposcopy-directed biopsy. A colposcopy-directed biopsy which showed CIN 1 and CIN 2 was also assigned as persistence. The women who showed CIN 3 in the biopsy were assigned as progression. The analysis included the data collected according to the follow-up plan. Data about the lesion outcome of the women who missed the twelve-month follow-up and returned later on were not included in the analysis. Women who showed progression to CIN 3 in the intermediate follow-ups were submitted to excision of the transformation zone (LLETZ) and it was considered the final outcome. The women who showed regression of the lesion in the intermediate months were followed up to the twelfth month, to ensure that the lesion actually regressed at the end of follow-up.

Given that the performance of cervical smears and colposcopy-directed biopsy may affect the probability of failure in the diagnosis CIN [16,17], the ideal method for evaluating the persistence and progression of CIN 2 should be to perform a LLETZ or cone biopsy, a more accurate diagnostic method.

Table 1
Cumulative frequency of the CIN 2 outcome for every three month follow-up.

		3 months	6 months	9 months	12 months
Progression	*n*	1	2	3	10
	%	2.6	4.9	7.5	24.0
	(95% CI)	(0.1–13.5)	(0.6–16.5)	(1.6–20.4)	(12.1–39.5)
Regression	*n*	15	26	28	31
	%	38.5	63.4	70.0	74.0
	(95% CI)	(23.4–55.4)	(46.9–77.9)	(53.5–83.4)	(58.0–86.1)
Persistence	*n*	23	13	9	1
	%	59.0	31.7	22.5	2.0
	(IC 95%)	(42.1–74.4)	(18.1–48.1)	(10.8–38.5)	(0.1–12.6)
Total	*n*	39	41	40	42
Unknown[a]	*n*	11	9	10	8

CI: confidence interval.
[a] Includes cases that were discontinued or missed follow-up.

Table 2
Woman's age at diagnosis, age at first sexual intercourse and interval since the first sexual intercourse for CIN 2 progression and regression.

	Progression			Regression			*p* value*
	n	Average (SD)	Median (min–max)	*n*	Average (SD)	Median (min–max)	*p** (Kruskall–Wallis)
Age at diagnosis	10	26.7 (8.7)	24.5 (19.0–43.0)	31	27.2 (7.7)	27.0 (17.0–47.0)	0.68
Age at first sexual intercourse	8	16.1 (1.9)	16.0 (13.0–18.0)	24	16.6 (2.4)	16.5 (13.0–21.0)	0.61
Interval since the first sexual intercourse	8	9.5 (6.2)	7.0 (3.0–22.0)	24	10.3 (7.9)	8.0 (1.0–30.0)	0.96

There was only one case of persistence of CIN 2 confirmed after 12 months, in a woman of 17 years of age.
* Kruskal–Wallis text assuming a significance level of 5% ($p < 0.05$).

Taking a biopsy without evidence of a lesion, however, would be unethical.

2.4. Statistical analysis

The rates of progression, regression and persistence of CIN 2, with a confidence interval of 95%, were calculated for every three-monthly consultation. The Kruskal–Wallis test was used to analyze the woman's age at diagnosis, age at first sexual intercourse and interval since the first sexual intercourse, according to the evolution of the CIN 2, assuming a significance level of 5%. The association between CIN 2 outcome and number of quadrants of the lesion at first colposcopic evaluation was measured by odds ratio (OR) with 95% confidence intervals (CIs).

3. Results

Eight (16%) of the 50 women followed in this study were not considered in the final evaluation of CIN 2 outcome because they discontinued their participation due to: diagnosis of persistence in the previous controls but failure to appear for the final consultation (five women), diagnosis of HIV confirmed during follow-up (one woman), three consecutive consultations missed (one woman), and positive pregnancy test during the first control consultation (one woman). All these eight women received appropriate management outside the study.

The average age of the 50 women included in the study was 26.5 years and the median was 25 years (min = 17; max = 47). The rates of progression, regression and persistence of CIN 2 at each follow-up consultation are shown in Table 1. At the end of the 12 months of monitoring, there was a 74% rate of regression (31/42), 24% of progression to CIN 3 (10/42) and only one case of persistence of CIN 2. No case of progression to invasive carcinoma was detected after 12 months of follow-up. Among women who had regression, this was detected in the first six months of follow-up in 26 of the 31 cases.

There was no statistically significant association between the woman's age at diagnosis, age at first sexual intercourse or the interval since the first sexual intercourse, with progression or regression of CIN 2 at the end of the 12-month period (Table 2).

Table 3
Association between CIN 2 outcome and number of quadrants of the lesion extension at first colposcopic evaluation.

	One quadrant	More than one quadrant	OR (95% CI)
Outcome	*n* (%)	*n* (%)	
3 months follow-up			
Regression	13 (52)	02 (14)	6.50 (1.20–35.23)
Persistence or progression	12 (48)	12 (86)	Reference
6 months follow-up			
Regression	20 (71)	06 (46)	2.92 (0.75–11.41)
Persistence or progression	08 (29)	07 (54)	Reference
9 months follow-up			
Regression	21(75)	07 (58)	2.14 (0.21–8.97)
Persistence or progression	07 (25)	05 (42)	Reference
12 months follow-up			
Regression	23 (79)	08 (67)	2.40 (0.46–12.72)
Persistence or progression	06 (21)	05(33)	Reference

OR: odds ratio; CI: confidence interval.
The cases that were discontinued or missed the follow-up were not included in the analysis.

Considering the extent of the initial lesion, one quadrant was involved in 34% of women and more than one quadrant in 66% of cases (data not shown). In one case, the colposcopist observed that the entire lesion has been removed by biopsy, and this case was not considered at final analysis. Women whose lesions were restricted to one quadrant were more likely to have CIN 2 regression at three-month follow-up when compared with women with a lesion extending to one or more quadrants (OR: 6.50; 95% CI: 1.20–35.23) (Table 3).

4. Comment

According to the results of this study, most CIN 2 regresses during the first six months of follow-up. At the end of 12 months, a rate of 74% of regression was observed, and 84% of these cases had regressed by the sixth month of follow-up.

Nasiell et al. [18] followed women with moderate dysplasia diagnosed in cervical smears and observed a regression rate of 54% at the end of 78 months. Although our study followed up women

with CIN 2 proven biopsy, the women had to have the screening cervical smear and the second cervical smear showing ASC-US or LSIL, cytological diagnoses less severe than in the Nasiell study. Longitudinal studies addressing the natural history of CIN are based on cytologic, histologic and colposcopic examinations, and used different nomenclatures and designs [1]. Guedes et al. [19] monitored women with CIN 2 diagnosed through colposcopy-directed biopsy in three-monthly follow-up over 12 months. They observed rates of 42% spontaneous regression. Unlike our study, Guedes' study included women with CIN 2 proven biopsy, regardless of whether the previous cervical smear showed ASCUS, LSIL or HSIL.

In our study, women were selected based on the cervical smear showing LSIL and the biopsy revealing CIN 2. These criteria might have selected CIN 2 with characteristics of productive infection, according to Snijders et al.'s model [2], and this fact could explain the high rate of regression observed in our study. Castle et al. [20], analyzing data on an ASCUS/LSIL Triage Study (ALTS) – that is, women selected based on an ASCUS or LSIL cervical smear – also considered that CIN 2 detected under these conditions could have a greater possibility to regress. These authors observed that women with biopsy-diagnosed CIN 2 following HSIL cytology at enrollment (women enrolled into ALTS had cytology repeated at the enrollment visit) were more apt to have an underlying CIN 3 than those with less severe enrollment cytology, and hypothesized that CIN 2 following HSIL cytology may be on average less regressive than that observed for CIN 2 diagnosed in the ALTS population.

The rate of progression to CIN 3 was 24%, of which 70% was detected at the 12-month follow-up. This finding may be because all women with suspicious colposcopy were subject to LLETZ at the final consultation. In our study, it is possible that some CIN 3 cases were already present at previous follow-ups but were not detected in the cytological and colposcopy evaluation, which suggests that they were borderline lesions to CIN 2. It should also be considered that the colposcopy-directed biopsy is not 100% sensitive regarding the detection of more severe lesions present on the uterine cervix [16]. Thus, it could be that some of the CIN 3 cases detected in the twelfth month were already present when the patient was admitted. The sensitivity of biopsy is greater when two or more biopsies are carried out [16,17]. In this study only the location where the image was considered more suspect was subject to a biopsy, and two locations only when the lesion was very extensive.

Although the HPV regression rate could be higher in young women [9,10], in our study the age of the women who showed CIN 2 regression was similar to the age of the women with CIN 2 progression. In this context and in agreement with these results, Rodriguez et al. [9] found that, in women with recent infections by HPV, the rate of progression to CIN 2 or a more severe lesion was not greater in women of 34 years of age or older. It should be considered that one sample of 50 cases has a low statistical power and, therefore, it is not possible to demonstrate statistically an actual, but discrete, association between age and regression or progression of CIN 2. On the other hand, if this association was strong, it would be demonstrated even with a sample of low statistical power.

The age at first sexual intercourse and the interval since first sexual intercourse did not differ for women with regressive or progressive CIN 2, even though the former is considered to be a co-factor in the risk of developing CIN and carcinoma of the uterine cervix [11,12]. Similarly, Moscicki et al. [21], monitoring women with LSIL, also showed no association between lesion regression and factors related to sexual behavior.

There is a possibility that biopsy may influence the regression of CIN, since it is suggested that biopsy of a small lesion can remove it entirely or precipitate an inflammatory response that will be curative [1,22]. According to the results of this study, a CIN 2 lesion size no larger than one quadrant presented higher regression rate at three-month follow-up (OR = 6.5, IC 95%: 1.5–35.2), suggesting complete disappearance of a small lesion after a diagnostic procedure at inclusion consultation. These results do not allow us to assert that CIN 2 involving one quadrant has a higher regression rate until the third month of follow-up, but we can affirm that CIN 2 involving one quadrant and undergoing biopsy has a higher regression rate. Chenoy et al. [22] indicated that directed punch biopsy trauma does not have a significant effect on the immediate natural history of CIN and did not find statistically significant differences in lesion size whether biopsy was employed or not. Obviously, all CIN 2 must be diagnosed by biopsy; if the biopsy can induce regression of this lesion; this would be a benefit for all women.

The results of this study indicate that the majority of CIN 2 diagnosed by biopsy in women with previous Pap smear showing LSIL will regress in 12 months and, therefore, an expectant approach could be considered in these cases, not only for young women as the ASCCP [6] recommends. Nevertheless, these findings are not conclusive, but suggest that larger studies are required in order to certify when it is safe to adopt expectant management for CIN 2.

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