

Efficacy and Long-term Outcomes of Repeated Large Loop Excision of the Transformation Zone of the Cervix

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OBJECTIVE: To evaluate the efficacy and long-term outcome of repeat large loop excision of the transformation zone in women with residual or recurrent cervical intraepithelial neoplasia.

METHODS: PALGA (the Dutch Pathology Registry), a database of deidentified cervical cytologic and histologic data, was used to examine women with cervical dysplasia who underwent two or more large loop excision of the transformation zone procedures between January 2005 and June 2015. We obtained cervical cytology and histology results. The main outcome was efficacy of repeated large loop excision of the transformation zone procedure in women with residual or recurrent cervical intraepithelial neoplasia. We also examined subsequent excisional procedures and hysterectomy.

RESULTS: We identified 499 women who had undergone two or more large loop excision of the transformation zone procedures. After their second procedure, 60.7% of women had a normal first cervical cytologic sample. The mean duration of follow-up was 68 months (0–163 months). Additional cervical excisional procedures were performed in 33.7% of women. Overall, 1.2% of women developed cervical cancer during follow-up. Moreover, 19.0% of women eventually underwent hysterectomy.

CONCLUSION: One third of the women who undergo two large loop excision of the transformation zone procedures require an additional excisional procedure or hysterectomy. Almost one fifth of these women eventually undergo hysterectomy.

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Cervical intraepithelial neoplasia (CIN) is the precursor of cervical cancer and most commonly develops in reproductive-aged women. It is classified into low-grade squamous intraepithelial lesions (CIN 1) and high-grade squamous intraepithelial lesions (HSIL, CIN 2–3).¹ High-grade CIN is a precursor of cervical cancer and, therefore, is usually treated with a large loop excision of the transformation zone.^{2,3} The aim of this procedure is to eliminate dysplastic cells in the cervical transformation zone.

After a first large loop excision of the transformation zone procedure, 5–27% of women have recurrent or residual CIN.^{2,4} Positive CIN margins, glandular, multiple quadrant disease and high-risk human papillomavirus (HPV) infection are all predictors of recurrent or residual cervical intraepithelial neoplasia after the large loop excision of the transformation zone procedure.^{5–8} Despite the fact that disease recurrence is not uncommon, national and

international guidelines offer no clear recommendations as to the preferred mode of treatment and follow-up of residual or recurrent cervical intraepithelial neoplasia. In most centers, a second excisional procedure is usually typically performed in case of recurrent high grade CIN.^{2,9}

Only one study, by Bowring et al,¹⁰ has reported the success rate of a repeated excisional procedure. The authors described findings in 53 women who underwent secondary cervical excisions and compared the efficacy of laser cone biopsy with large loop excision of the transformation zone procedure as second treatment. Fifty-nine percent of the women who underwent a second large loop excision of the transformation zone procedure had normal cytology in follow-up. To estimate the efficacy and long-term outcomes of repeat large loop excision of the transformation zone procedures, we performed an observational analysis of women with recurrent or residual CIN who underwent repeat cervical excisional procedures.

METHODS

We used data from PALGA (the Dutch Pathology Registry), a Netherlands nationwide network and registry of histopathology and cytopathology.¹¹ We extracted data for all women who had undergone two or more large loop excision of the transformation zone procedures due to CIN between January 2005 and June 2015. The study was approved by the Medical Ethics Review Board of the Erasmus University Medical Center (MEC 2015-443). There was neither patient nor public involvement in this study.

A search of the PALGA database was performed to identify women who had two histology results matching the criteria “cervix” AND “all neoplasms” AND “no malignancy” and “NOT biopsy” and “NOT curettage”. Two individual researchers separately reviewed the search results for possible inclusion. We included those women who had CIN, and excluded women who were either diagnosed with invasive carcinoma in the first or repeat large loop excision of the transformation zone procedure or those with adenocarcinoma in situ. Furthermore, to eliminate possible re-excisions because of positive margins, patients with an interval of less than 3 months between the first and second excisional procedures were excluded.

The following data were collected for patients who met the inclusion criteria: date of first large loop excision of the transformation zone procedure; CIN classification of the first large loop excision of the transformation zone procedure; date of the second large loop excision of the transformation zone; CIN classification of the second large loop excision of the

transformation zone; patient’s age at the first and second procedures; date and results of the first cervical cytology after the second large loop excision of the transformation zone; and the date and result of the last cervical cytology in our follow-up period. If more than two large loop excision of the transformation zone procedures had been performed, the date and results of all subsequent large loop excision of the transformation zone procedures were extracted. Large loop excision of the transformation zone procedures without dysplasia were counted, to calculate how many invasive procedures had been performed after the second large loop excision of the transformation zone. The occurrences of hysterectomy as well as the histologic results were obtained. The end of follow-up was defined as the last cervical cytology or performance of hysterectomy. As this was a retrospective study, any missing data were recorded.

To enable international comparison, the cervical cytology results were converted from Pap classification to the Bethesda classification using the Dutch CISEO-A framework.¹² The main outcome was the first cervical cytology specimen approximately 6 months after the second procedure. A successful treatment was defined normal or negative cytologic results in the Bethesda classification. Additionally, we inventoried secondary procedures such as additional large loop excision of the transformation zone procedures or hysterectomy.

Descriptive statistics were applied to patient characteristics and the primary outcome measure. Pearson chi-square test was used for a subgroup analysis regarding the association between patient characteristics and risk for an additional procedure (large loop excision of the transformation zone or hysterectomy). The age of the patient at the time of the repeat large loop excision of the transformation zone procedure or hysterectomy was classified into two categories: 40 years and younger and older than 40 years. The time between the first two procedures was analyzed. Here, two different cut points were used: a period of 6 months and 12 months, respectively. We also estimate the risk difference with 95% CI.

We estimated the cumulative probability of performance of a third large loop excision of the transformation zone procedure or hysterectomy given a certain time by the Kaplan Meier estimator. CIs were obtained using the Greenwood method.

Additionally, we estimated the expected cumulative number of procedures (large loop excision of the transformation zone and hysterectomy) for an individual as a function of the time elapsed since the second large loop excision of the transformation zone (called the mean cumulative function) using the

nonparametric Nelson-Aalan estimator and the variance estimate of Lawless and Nadeau.¹³ This method is a generalization of the Kapan Meier method for recurrent events. Analysis was performed using R 4.0.3 and the Reda package 0.5.2. In all analyses, $P < .05$ was considered statistically significant. Statistical analysis was performed using SPSS 23.0 as well as R 4.0.3 and the Reda package 0.5.2.

RESULTS

In total, 499 women met the inclusion criteria and were selected for final analysis. Their baseline characteristics are shown in Table 1. The median age was 39 years (range 24–72 years) at the date of repeat large loop excision of the transformation zone. The mean time interval between the first and second procedure was 13 months (range 3–90 months). In the majority of patients (403/499 [80.8%], 95% CI 77.0–84.1%), CIN 3 was the histologic result in the second large loop excision of the transformation zone procedure. The first cervical cytology after the second procedure was performed after a median of 6 months (range 2–47 months), and the result was negative in 303 women (60.7%, 95% CI 56.3–65.0%) (Table 2).

Table 1. Clinical and Demographic Characteristics of the Cohort (N=499)

Variable	n (%)
Interval between 1st and 2nd LLETZ procedure (mo)	
3–6	113 (22.6)
6–12	227 (45.5)
More than 12	159 (31.9)
Age at 2nd LLETZ procedure (y) [median (range)]	39 (24–72)
Cervical cytology before 2nd LLETZ procedure	
Inadequate	1 (0.2)
Negative	4 (0.8)
ASC-US	15 (3.0)
LSIL	57 (11.4)
HSIL	407 (81.4)
Missing	15 (3.0)
HPV status before 2nd LLETZ procedure	
Negative	2 (0.4)
Positive	71 (14.2)
Missing	426 (85.4)
Histologic result of 2nd LLETZ procedure	
CIN 1	44 (8.8)
CIN 2	43 (8.6)
CIN 3	403 (80.8)
Unknown grade of dysplasia	9 (1.8)

LLETZ, large loop excision of the transformation zone; ASC-US, atypical squamous cells of undetermined significance; LSIL, low-grade squamous intraepithelial lesions; HSIL, high grade squamous intraepithelial lesion; HPV, human papillomavirus; CIN, cervical intraepithelial neoplasia.

Table 2. First Cytology Results After Repeat Large Loop Excision of the Transformation Zone (N=499)

Variable	n (%)
Cervical cytology	
Inadequate	2 (0.4)
Negative	303 (60.7)
ASC-US	51 (10.2)
LSIL	27 (5.4)
HSIL	81 (16.2)
SCC	1 (0.2)
Missing	34 (6.8)
HPV status	
Positive	37 (7.4)
Negative	36 (7.2)
Missing	426 (85.4)

ASC-US, atypical squamous cells of undetermined significance; LSIL, low-grade squamous intraepithelial lesion; HSIL, high grade squamous intraepithelial lesion; SCC, squamous cell carcinoma; HPV, human papillomavirus.

The mean duration of follow-up was 68 months (range 0–163 months). In total, 168 women (33.7%) had 214 additional surgical procedures after the second large loop excision of the transformation zone procedure. Overall, 111 (66.1%) of women who underwent an additional procedure had HSIL. The group of 57 women who did not have HSIL but underwent an additional large loop excision of the transformation zone included women with low-grade squamous intraepithelial lesions, dysplasia of unknown grade, no dysplasia, or invasive carcinoma. An additional large loop excision of the transformation zone procedure was performed in 94 (18.8%) women, and 95 women (19.0%) underwent hysterectomy, 21 after an additional large loop excision of the transformation zone procedure (Table 3). Six women (1.2%, 95% CI 0.4–2.2%) were diagnosed with cervical cancer, including stage I tumors in five women and unclassified in one.

In Figure 1, the expected number of additional procedures is plotted against the follow-up time. The majority of the estimated additional procedures were performed in the first 72 months of follow-up. The curve then flattens. At 96 months, the estimated cumulative rate of additional procedures is 0.47 (95% CI 0.40–0.54). The red line represents the first additional procedure (third large loop excision of the transformation zone or hysterectomy), plotted against time. This line represents the women who had a repeat procedure after the two large loop excision of the transformation zone procedures. This shows that there is a 33% (95% CI 29–37%) probability of an additional procedure after 6 years (72 months).

Table 3. Histologic Results of Procedures Performed After the Second Large Loop Excision of the Transformation Zone

Histologic Result	LLETZ 3	LLETZ 4	LLETZ 5	LLETZ 6	Hysterectomy
CIN 1	10/94 (10.6)	1/19 (5.3)	0/4 (0.0)	0/2 (0.0)	8/95 (8.4)
CIN 2	9/94 (9.6)	2/19 (10.5)	1/4 (25)	0/2 (0.0)	10/95 (10.5)
CIN 3	50/94 (53.2)	9/19 (47.4)	2/4 (50)	2/2 (100)	53/95 (55.8)
Unknown grade of dysplasia	1/94 (1.1)	1/19 (5.3)	0/4 (0.0)	0/2 (0.0)	3/95 (3.2)
No dysplasia	21/94 (22.3)	5/19 (26.3)	1/4 (25)	0/2 (0.0)	19/95 (20.0)
Invasive carcinoma	3/94 (3.2)	1/19 (5.3)	0/4 (0.0)	0/2 (0.0)	2/95 (2.1)
Total	94/499 (18.8)	19/499 (3.8)	4/499 (0.8)	2/499 (0.4)	95/499 (19.0)

LLETZ, large loop excision of the transformation zone; CIN, cervical intraepithelial neoplasia. Data are n/N (%).

In a subgroup analysis of the association between patient characteristics and the risk for an additional procedure, older age was a risk factor for hysterectomy. Hysterectomy was performed in 37 of 292 (12.7%) women aged 40 years or younger compared with 58 of 207 (28.0%) women aged older than 40 years (risk difference -15.3%, CI -22.6 to 8.1%) ($P<.001$). An additional large loop excision of the transformation zone procedure was performed in 63 of 292 (21.6%) women aged 40 years or younger compared with 31 of 207 (14.9%) women aged older than 40 years (risk difference 6.6%, CI -0.2% to 13%) ($P=.06$).

We analyzed the risk of an additional large loop excision of the transformation zone procedure or hysterectomy based on the duration of the interval between the first and second large loop excision of the transformation zone procedures. The length of the interval between the two initial large loop excision of the transformation zone procedures was not related to the risk of an additional large loop excision of the transformation zone procedure or hysterectomy. In 21 of the 113 patients (18.6%) with an interval of less than 6 months between the first two large loop excision of the transformation zone procedures, an additional large loop excision of the transformation zone had been performed, compared with 73 of the 317 patients (18.9%) with an interval longer than 6 months (risk difference -0.3%, CI -8.5% to 7.8%) ($P=.94$). We repeated the same analysis for an interval of 12 months between the first two large loop excision of the transformation zone procedures. Repeat excision was performed in 61 of 340 (17.9%) patients with an interval of 12 months or less compared with 33 of 159 (20.8%) patients with an interval of greater than 12 months (risk difference -2.8%, CI -10.3% to 4.7%) ($P=.45$).

Sixty-three of 340 women (18.5%) underwent hysterectomy when the interval between the two initial procedures was less than 12 months, compared with 32 of 159 women (20.1%) with a longer interval (risk difference -1.6%, CI -9.1% to 5.9%) ($P=.67$). Similarly, 26 of 113 (23%) women with an interval of 6 months or less between the two large loop excision of the transformation zone procedures underwent hysterectomy, compared with 69 of 386 (17.9%) women with an interval of greater than 6 months (risk difference 5.1% CI -3.5% to 13.8%) ($P=.22$).

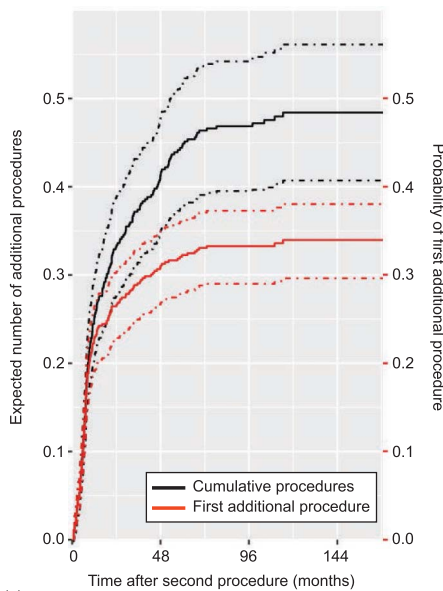


Fig. 1. Procedures performed in follow-up. van de Sande. Efficacy of a Repeated Excisional Procedure of the Cervix. *Obstet Gynecol* 2022.

DISCUSSION

We conducted a large nationwide study that examined the efficacy of repeat large loop excision of the transformation zone procedure and found that 60.7% of the

women had a negative result for the first cervical cytologic test after their second procedure. One third of the women required an additional procedure after the second procedure. Almost one fifth of the women eventually underwent hysterectomy.

Although a second excisional procedure is often advised in case of disease recurrence, guidelines do not provide clear recommendations for the management of residual or recurrent cervical intraepithelial neoplasia, and little is known about the success rate after a second procedure.^{2,9} The success rate of 60.7% that we found is comparable with that found by Bowring et al,¹⁰ the only previous study of the efficacy of repeat large loop excision of the transformation zone procedure. In that study, in which only 19 women underwent such a procedure, 10 of 17 women (59%) had negative cytologic results after a secondary large loop excision of the transformation zone procedure.

Our study included only women who had tested positive for CIN before each procedure. Requiring an interval between procedures of 3 months or greater was aimed at excluding procedures performed for positive margins from the first procedure. Dutch guidelines do not routinely advise a repeated procedure after a large loop excision of the transformation zone with positive margins.⁹ This advice varies internationally, however. Moss et al¹⁴ reported that incomplete excision or positive margins with the first large loop excision of the transformation zone procedure was one of the most common indications for a second large loop excision of the transformation zone procedure. However, even after excluding possible re-excisions because of positive margins, our success rate is comparable with that of Bowring et al.

Six women included in our study developed an invasive carcinoma during follow-up. This suggests that these women are at high risk for cervical cancer. Additionally, the histologic results of the majority of women who had undergone repeat procedures indicated a high-grade CIN lesion and justifies repeated procedures.

The PALGA database that we used does not provide information about the type of excisional procedure. We assumed that most of the procedures were large loop excision of the transformation zone procedures, because this is the most common surgical option in the Netherlands for CIN. However, some of the procedures could have been cold-knife cone biopsies. Bowring et al¹⁰ demonstrated a trend towards a higher success rate for laser cone biopsies than a large loop excision of the transformation zone procedure as a secondary treatment. Because fertility and obstetric complications are concerns in women of childbearing age, the risk of cone biopsy should be weighed against the risk of recurrence of CIN.

Another treatment modality, imiquimod, has been investigated in women with residual or recurrent cervical intraepithelial neoplasia¹⁵ and was associated with a success rate of 61%. Imiquimod has been shown to be effective in the treatment of primary CIN lesions¹⁶ and may have fewer effects on future pregnancies. For women who do not desire future fertility, hysterectomy may be a reasonable alternative to a large loop excision of the transformation zone procedure to prevent cervical cancer, despite the higher risk of complications. In our study, women 40 years of age and older had a 28% probability of undergoing hysterectomy after the second procedure. Overall, a high proportion of women underwent an additional procedure after the second large loop excision of the transformation zone procedure. The fact that most of the additional procedures were performed within 2 years after the second procedure affirms the currently recommended practice to screen these women frequently for at least 2 years after a large loop excision of the transformation zone procedure.⁹ Determination of high-risk HPV or biomarkers may help to identify women at risk for residual or recurrent cervical intraepithelial neoplasia.¹⁷

The main strengths of our study are the large sample size, the long follow-up period, and the use of data from a reliable national database. Additionally, our study yields useful information about the efficacy and risk of additional procedures which may aid in patient counseling.

Inherent to any retrospective analysis, missing data constituted a major limitation of this study. All data were de-identified, and, as such, some clinical and demographic characteristics of the cohort were unknown. We lacked data on tobacco use, use of medications, medical history, and performance of any cervical procedures. Lastly, HPV status was largely unknown. High-risk HPV DNA detection has been the primary cervical cancer screening modality in the Netherlands since 2017 and is included in follow-up.¹⁸ The majority of our study period was before 2017, when HPV detection was not routinely performed.

The findings from our study provide strong support for the assumption that the success rate of a second large loop excision of the transformation zone procedure is lower than a first procedure. Moreover, women who have undergone multiple excisional procedures are at high risk for additional excisional procedures and hysterectomy. Nonexcisional therapy should be further explored for women who wish to conceive in the future. For women who have no wish to conceive, hysterectomy may be considered as a treatment option.

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