



Efficacy of TIAGIN® on post-operative reepithelialization in women subjected to cervical surgery (LEEP or laser cone biopsies): case-control study

Anna Daniela Iacobone¹, Eleonora Petra Preti¹, Ailyn Mariela Vidal Urbinati¹,
Eugenia Tomás Roldán²

¹ Preventive Gynaecology Unit, European Institute of Oncology, Milan

² CERISMAS Research Centre, Catholic University of Milan

ABSTRACT

Objective: to evaluate the efficacy of post-operative treatment with TIAGIN® vaginal softgel in the reepithelialisation of the cervix and the prevention of the most common post-operative complications.

Methods: a prospective and randomised case-control study in women who had undergone cervical surgery: TIAGIN® users for 15 days and non-users. All patients underwent a clinical examination at baseline and at 30 days. The post-treatment reepithelialisation process was evaluated by 2 colposcopists. A visual analogue questionnaire was completed in the second and fourth week after surgery to assess vaginal bleeding, leucorrhoea, abdominal pain and impact of pain on quality of life.

Results: 30 patients were enrolled: 12 as controls and 18 as cases. 5 LEEP (41.77%) and 7 laser cone biopsies (58.33%) were carried out among controls and 9 LEEP (50.00%) and 9 laser cone biopsies (50.00%) among cases. CIN2-3 was detected in more than 70% of patients. No statistically significant differences were observed in post-operative complications, with the exception of the rate of atypical leucorrhoea (66.67% in controls vs 16.67% in cases) ($p=0.011$). Complete reepithelialisation was seen in 66.6% of cases and in 8.33% of controls, respectively ($p=0.002$).

Conclusion: the post-operative use of TIAGIN® vaginal softgel leads to rapid cervical reepithelialisation.

Keywords: TIAGIN® vaginal softgel, cervical surgery, post-operative complications, reepithelialisation

SOMMARIO

Obiettivo: efficacia del trattamento post-operatorio di TIAGIN® softgel vaginali nella riepitelizzazione della cervice uterina e nella riduzione delle complicanze post-operatorie.

Metodi: studio prospettico randomizzato in pazienti sottoposti a LEEP/laserconizzazione: pazienti trattate con TIAGIN® softgel vaginali per 15 giorni e controlli non trattate. Le pazienti hanno eseguito visita ginecologica e colposcopia al tempo zero e a 30 giorni. Il grado di riepitelizzazione cervicale è stato valutato da 2 colposcopisti esperti. Un questionario visuo-analogico è stato somministrato dopo 2 e 4 settimane dall'intervento chirurgico per valutare entità del sanguinamento, leucorrea, intensità del dolore addominale ed impatto del dolore sulla qualità di vita.

Risultati: 30 pazienti arruolate di cui 12 nel gruppo controllo e 18 nei casi. Sono state eseguite: 5 LEEP (41.77%) e 7 laserconizzazioni (58.33%) nei controlli; 9 LEEP (50.00%) e 9 laserconizzazioni (50.00%) nei casi. 70% delle pazienti ha avuto un istologico CIN2/3. Nel gruppo dei controlli si è avuta una maggior leucorrea rispetto ai casi (66.67% vs 16.67%) ($p=0.011$), mentre la riepitelizzazione completa si è verificata nel 66.6% del gruppo dei casi rispetto all'8.33% dei controlli ($p=0.002$). Non sono state trovate altre differenze statisticamente significative nel tasso di complicanze post-operatorie.

Conclusioni: TIAGIN® softgel vaginali nel post-operatorio velocizza la riepitelizzazione della cervice.

INTRODUCTION

Wound healing is a multifaceted process comprising three phases: inflammation, proliferation and remodelling with the growth of the epithelium and connective tissue.⁽¹⁾

The production of cytokines deriving from the inflammatory processes influences the healing process, while certain immunological mediators and bacterial colonisation can delay this process⁽²⁾.

The early inflammation that occurs as a physiological response to the tissue damage can prevent an incipient infection, but prolonged inflammation can cause cell damage. In fact, the presence of pathogens produces toxins and enzymes that protract the inflammatory response and delay healing, with an increased risk of complications.⁽²⁻³⁾

The rapid repair of the wound bed is essential in order to avoid secondary bacterial infection and thus prevent complications, particularly after surgery.

Silver-based products have historically been recommended in wound management because of their bactericidal properties. However, the frequent and long-term application of these topical products produced inflammation and cosmetic defects. What is more, the advent of antibiotic therapy considerably reduced the use of these medications. The arrival of nanotechnology has allowed to create silver nanoparticles, with improved solubility and surface properties and, consequently, fewer side effects.⁽⁴⁾

The innovative patented TIAB® formula creates a protective film on the vaginal mucosa that releases nanoparticles of ionic Ag (Ag+) stabilised with TiO₂ (TIAB® System). The silver nanoparticles have a larger surface area and this increases the release rate of the individual metal charges, which are the most soluble and biologically active part.⁽⁵⁾ They also possess multiple antibacterial mechanisms: they are oxidized and release a silver charge that damages the cellular membrane of the pathogenic microorganisms. The ions penetrate the cytoplasm, prevent the replication of the DNA, deactivate the respiratory enzymes and block cellular division until cell lysis occurs. They also generate free radicals, which have significant bactericidal properties.⁽⁶⁻⁸⁾

TIAGIN® contains a blend of TIAB® and hyaluronic acid.

During the inflammatory phase, low-molecular-weight hyaluronic acid accumulates in the wound bed, where its main function is to modulate the action of the inflammatory cells and the activity of fibroblasts, such as cell migration,

cytokine synthesis and microbe phagocytosis. High-molecular-weight hyaluronic acid, on the other hand, keeps the wound moist by absorbing water. During the second phase of tissue damage, hyaluronic acid acts on the fibrocyte differentiation mechanisms and the expression of macrophage metalloelastase. In brief, all the mechanisms involved in the tissue repair phase involve low-molecular-weight hyaluronic acid as the principal player, as it aids the rapid reconstruction of the healthy tissue.⁽⁹⁻¹⁰⁾

Various clinical studies have been carried out to evaluate the use of low-molecular-weight hyaluronic acid in the various fields of medicine, including gynaecology.⁽¹¹⁻¹²⁾

A cone biopsy is a surgical procedure used for the diagnosis and treatment of pre-cancerous lesions affecting the uterine cervix. Surgical techniques vary depending on the hospital and the experience of surgeons. A cold knife cone biopsy used to be the most common surgical procedure, but the most widespread technique today is LEEP (Loop Electrosurgical Excision Procedure), while specialist sites carry out laser cone biopsies.⁽¹³⁻¹⁴⁾ All these techniques remove part of the uterine cervix for histopathological analysis. They are relatively simple procedures and are usually performed under local anaesthetic.

In literature, there are very few studies about the immediate and/or subsequent complications associated with the various techniques described above. A recent meta-analysis, which compares LEEP/laser cone biopsy with cold knife cone biopsy estimates that adverse events affect around 1.1% of women, subdivided into: bleeding (RR 0.226-0.859), infection (RR 0.128-0.089), PID (RR 0.139-0.138) and minor bleeding (RR 0.363-2.450).⁽¹⁵⁾

On the contrary, a more considerable number of studies evaluated obstetric outcomes in women subjected to surgical treatment of the cervix. Cold knife cone biopsy would seem to be associated with a higher number of preterm births and/or PPROM, as laser cone biopsy, compared to LEEP procedures. The reason of these results is probably related to the quantity of tissue removed, that leads to an increased risk of bacterial infections, PPROM and preterm birth, rather than the technique itself.⁽¹⁶⁾

At present, there are no clinical studies evaluating the efficacy of capsules containing silver nanoparticles associated with hyaluronic acid (TIAGIN® vaginal capsules) in women who have just undergone cervical surgery. Hyaluronic acid has a positive effect on fibroblast proliferation.

Thanks to its negative charge, it is localised in the extracellular region, where a hydrophilic network develops and can carry nutrients and metabolites to the different cell types by means of a percolation mechanism. Moreover, the addition of Ag+ NPs fosters collagen formation and controls its spatial orientation. Therefore, it is possible to hypothesise that the scarring and antibacterial action of this combination (Ag + hyaluronic acid) can reduce post-surgical repair times and post-operative complications.

OBJECTIVE

The primary objective of our study was to evaluate the efficacy of post-operative treatment with TIAGIN® vaginal capsules in the reepithelialisation of the cervix and the prevention of the most common post-operative complications (abdominal pain, bleeding and atypical vaginal discharge).

MATERIALS AND METHODS

We carried out a prospective and randomised case-control study in women who had undergone cervical surgery in order to compare the reparatory phenomena and post-operative complications at 2 and 4 weeks after the operation between TIAGIN® vaginal capsule users and non-users.

All the women who underwent LEEP (Loop Electrosurgical Excision Procedure) or a laser cone biopsy at our institute from 11 April 2016 to 31 July 2016 were given the opportunity to take part in the study. The exclusion criteria were: pregnancy, congenital or acquired immune deficiency syndrome, immunosuppressant use, previous hysterectomy, and/or the use of other vaginal medications during the week prior to surgery.

The study was approved by our Institute Review Board and all the participating subjects signed an informed consent form.

The study protocol included two enrolment arms: a group of patients subjected to post-operative treatment with vaginal capsules containing TIAGIN® for 15 days and a group of patients who did not take any post-operative vaginal treatment. The randomisation procedure was carried out by personnel not involved in the clinical study, who also assigned the progressive participation numbers.

All patients underwent a clinical examination,

including a gynaecological visit and a speculum examination at baseline (at the time of surgery) and at 30 days. The clinical evaluations were carried out by gynaecological experts in cervical disease, certified by the Italian Society of Colposcopy and Cervicovaginal Pathology, who assigned a score between 1 and 3 to the post-treatment reepithelialisation process (1: ectocervix completely reepithelialised, 2: ectocervix covered in a thin epithelium with areas of immature metaplasia, 3: ectocervix with persistent areas of erosion, prone to bleeding), based on a subjective assessment.

All the enrolled women completed a baseline questionnaire regarding their socio-demographic characteristics and obstetric/gynaecological history and a questionnaire regarding the main post-operative complications in the second and fourth week after surgery. The complications questionnaire used a visual analogue scale as follows:

- √ extent of vaginal bleeding (slight-moderate-abundant)
- √ extent of leucorrhoea (0-10)
- √ intensity of abdominal pain (0-10)
- √ impact of pain on quality of life (1-5)

STATISTICAL ANALYSIS

Patients' main characteristics were expressed as absolute and percentage frequencies.

Post-operative complications and the clinical evaluation of the reepithelialisation process of cases and controls were grouped together as categorical variables and compared with Fisher's exact test. P values ≤ 0.05 were considered statistically significant.

All data were analysed using SAS software (version 9.3).

RESULTS

From April to July 2016, more than 150 cervical surgery procedures were carried out at our institute, all on an outpatient and day surgery basis. A total of 30 patients were enrolled: 12 in the control group and 18 in the case-study group. The low percentage of subjects signing up to the study can be attributed almost entirely to the need to attend a check-up visit 30 days after the operation, as many of our patients live over 100 km away from our hospital.

The main socio-demographic and clinical

characteristics of the study population are summarized in **Table 1** and are comparable in the two groups. The enrolled patients had a mean age of 39.26 ± 13.44 and were predominantly multiparous (56.67%).

As regards to methods of contraception, the use of birth control pills was prevalent in the control group (16.67%), while barrier contraception was predominant in the case group (44.44%).

Table 1.
Main socio-demographic and clinical characteristics of the enrolled population.

	TOTAL (N=30)	CONTROLS (N=12)	CASES (N=18)
	Mean (SD)	Mean (SD)	Mean (SD)
Age (years)	39.26 (13.44)	41.25 (3.54)	39.94 (14.14)
	n (%)	n (%)	n (%)
Nulliparous	13 (43.33)	6 (50.00)	7 (38.89)
Smoker	13 (43.33)	5 (41.66)	8 (44.44)
Oral contraceptives	3 (10.00)	2 (16.67)	1 (5.56)
Condom use	9 (30.00)	1 (8.33)	8 (44.44)
Vaginal infections in the last year	9 (30.00)	3 (25.00)	6 (33.33)
Cystitis in the last years	7 (23.00)	3 (25.00)	4 (22.22)
Operation type			
LEEP	14 (46.67)	5 (41.77)	9 (50.00)
Laser cone biopsy	16 (53.33)	7 (58.33)	9 (50.00)
Histology test			
Negative	3 (10.00)	1 (8.33)	2 (11.11)
CIN1	4 (13.00)	2 (16.67)	2 (11.11)
CIN 2-3	22 (73.33)	9 (75.00)	13 (72.22)
VAIN3	1 (3.33)	0 (0.00)	1 (5.56)

The surgical procedures carried out were as follows: 5 LEEP (41.77%) and 7 laser cone biopsies (58.33%) among controls and 9 LEEP (50.00%) and 9 laser cone biopsies (50.00%) among cases.

The histopathological analysis revealed the presence of CIN2-3 in more than 70% of patients of both groups.

The clinical evaluation of the post-surgery cervical reepithelialisation process at 30 days revealed a statistically significant difference between the two groups (**Figure 1**).

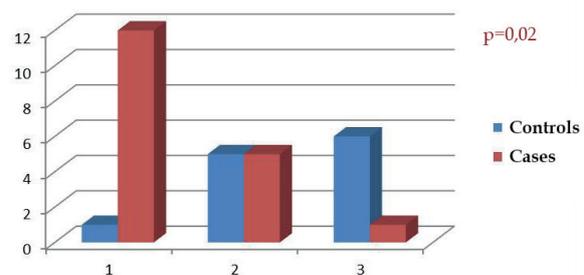


Fig. 1
Cervical reepithelialization score

In fact, in 66.6% (12/18) of the women who used vaginal capsules containing TIAGIN®, the examining gynaecologists assigned a score of 1, indicating complete reepithelialisation, which was only awarded in 8.33% (1/12) of the women who

did not use any local post-surgery treatment. In addition, 50% (6/12) of the control group showed areas of erosion prone to bleeding during the post-operative visit ($p=0.002$). (Figures 2.1 and 2.2).

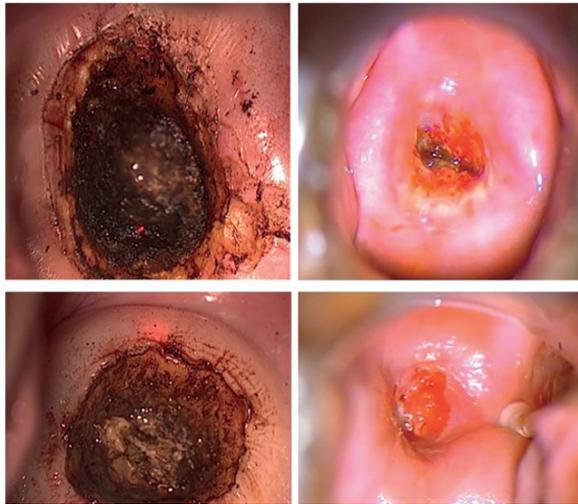


Fig. 2.1
Two control patients immediately after the operation and at the 30-day check-up



Fig. 2.2
Two cases patients immediately after the operation and at the 30-day check-up

As shown in Figure 3 no statistically significant differences were observed in post-operative complications between cases and controls, both at 2 and 4 weeks, with the exception of the rate of atypical leucorrhoea at 2 weeks, which was higher

in the control group (66.67%) compared to the case group (16.67%) ($p=0.011$).

Lastly, none of the patients reported severe abdominal pain or maximum impact of pain on quality of life.

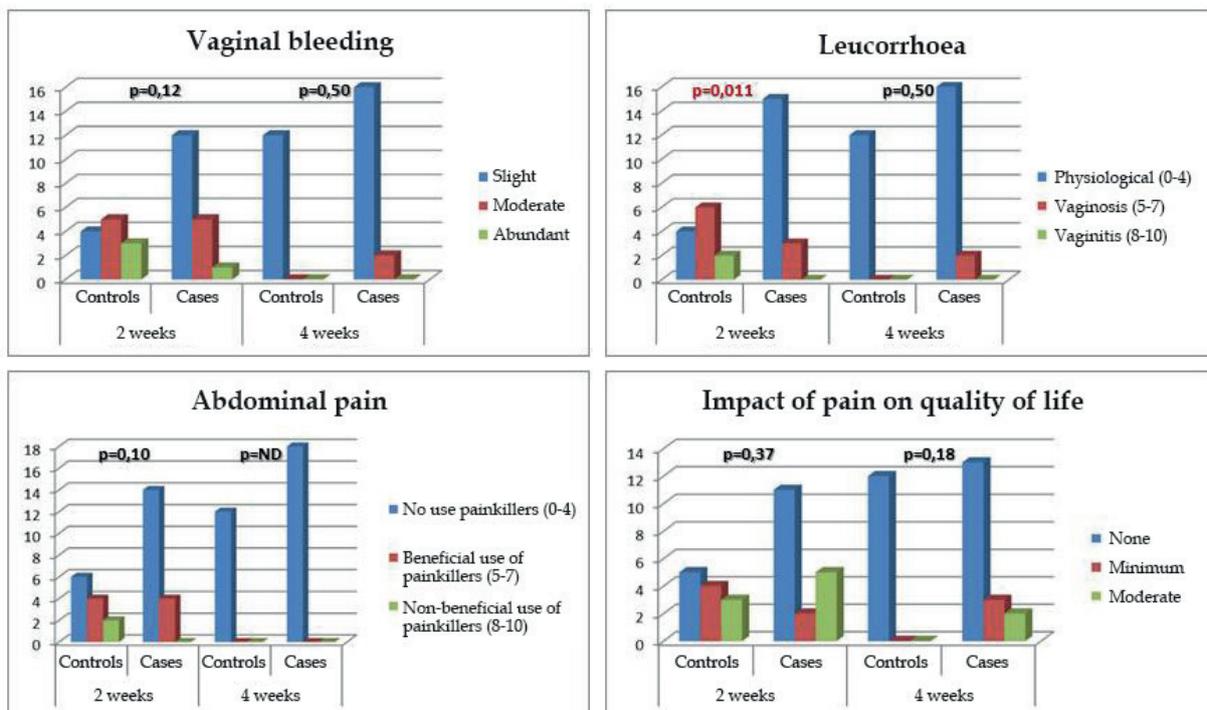


Fig. 3
Post-operative complications

CONCLUSIONS

In the light of recent literature, there are no studies about the use of silver nanoparticles in combination with hyaluronic acid to reduce the risk of complications after cervical surgery.

Many methods have been evaluated for the prevention of severe bleeding after cone biopsies. Doyle reports the application of a Monsel solution post-treatment,⁽¹⁷⁾ Kim K. describes a retrospective study for the use of a fibrin sealant on the wound bed post-LEEP [18] and Kim JJ. describes the preventive use of Tachosil®.⁽¹⁹⁾ All these authors showed that these agents do not lead to a statistically significant difference in the prevention of bleeding, except Kim K., which demonstrates a significant reduction of severe bleeding ($p=0.033$, OR - 0.328, 95% CI 0.117-0.917), despite a consistent impact on costs.⁽¹⁸⁾

Indeed, the most recent guidelines do not advise any specific therapeutic agent for the prevention of severe bleeding.

Our study reflects the results of previous studies regarding the risk of slight or severe bleeding, with no statistically significant difference between TIAGIN® vaginal capsule users and non-users. However, the number of patients was limited and there were no episodes of severe bleeding that required hospitalisation and/or cervical haemostasis, even in the cases of cone biopsies with a cone length of > 2 cm.

For what concerns compliance of patients, none of the case-subjects stopped treatment due to problems associated with the use of the capsules

(increased bleeding, blood loss, discomfort or other), so confirming the good tolerability of the capsules and their easy application.

The most statistically significant finding is related to the clinical evaluation, which demonstrates good and rapid reepithelialisation in the group of women treated with TIAGIN® capsules. This result could have a considerable impact on the risk of infection and chronic inflammation. We should not forget that, by reducing the length of the uterine cervix, cervical surgery can increase the risk of infection and preterm birth. Previous literature already showed that the higher risk of preterm birth is a consequence of modifications of the local vaginal flora in association with other co-factors.

The rapid repair process in TIAGIN® vaginal capsule users means that the uterine cervix is shortly exposed to chronic inflammation and associated cytotoxic factors. Thereafter, the bactericidal effect of silver acts as an adjuvant in this process, restricting bacterial colonisation. Although not statistically significant in terms of the immediate complications associated with cervical surgery, all these factors entail an improved healing of the uterine cervix and a possible benefit for subsequent obstetric outcomes.

A long-term follow-up and more clinical studies evaluating obstetric outcomes are certainly mandatory to verify whether this kind of support treatment during the post-surgery phase can lead to a long-term improvement of quality of life.

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