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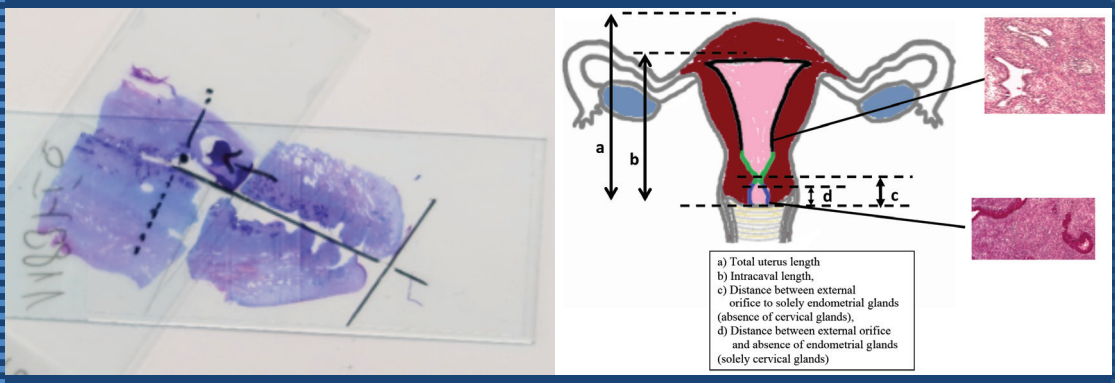

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Endometrial scratching for poor responders in IVF
Şenol Kalyoncu et al.; Ankara, Gaziantep, Turkey

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Aims and Scope

Journal of the Turkish-German Gynecological Association is the official, open access publication of the Turkish-German Gynecological Education and Research Foundation and Turkish-German Gynecological Association and is published quarterly on March, June, September and December. The publication language of the journal is English. Manuscripts are reviewed in accordance with "double-blind peer review" process for both reviewers and authors.

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Editorial

Dear Colleagues,

I am delighted to introduce the first issue of the "Journal of the Turkish-German Gynecological Association (J Turk Ger Gynecol Assoc)" in the publishing year of 2021. This first issue of year 2021 contains articles on subjects of obstetrics, oncology, gynecology and infertility.

As you know, telemedicine refers to the practice of caring for patients remotely when the doctor and patient are not physically present with each other. Modern technology has enabled doctors to consult patients by using video-conferencing tools. Here you will read a paper evaluating the feasibility of telephone interviews for patients in which non-urgent, conventional, post-operative check-up was suspended due to Coronavirus disease-2019 (COVID-19).

Catechol-O-methyltransferase (COMT), the product of the *COMT* gene, detoxifies the carcinogenic catechol estrogens. You will read an interesting paper examining the relationship between COMT Val158Met polymorphism and the risk of ovarian cancer.

Also you will get the occasion to read the consensus opinion and suggestions of "Turkish Society of Minimally Invasive Gynecological Oncology" for the preoperative evaluation and morcellation of fibroids, in line with the recent literature.

Dear esteemed readers,

As a result of the rising trend among academic journals, our journal will be published only online as of March 2021. The online version enables all readers to view, download and print the full-text of all published articles without any subscription or restrictions.

As part of our efforts to share scientific information with as many people as possible, we have added social media to Journal of the Turkish-German Gynecological Association's range of communication channels. Our journal's Twitter account began to actively share the journal's content as of November 2020. Keep in touch with us by following us on Twitter @JtggaOfficial.

I appreciate the work done by authors, reviewers and the editorial board, whose collaboration has resulted in production of the issues. On behalf of the editorial board, I wish you a happy and prosperous new year.

Best regards,

Prof. Cihat Ünlü, M.D.
Editor in Chief of *J Turk Ger Gynecol Assoc*
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The influence on resection line during supracervical hysterectomy: physiological extension of endometrial cells in the cervix uteri

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Abstract

Objective: A straight resection of corpus uteri using the sacrouterine ligament as landmark is a common method during supracervical hysterectomy. Subsequent spotting rates of up to 25% suggest the existence of residual endometrial glands in the remaining cervical tissue, casting doubt on the landmark qualities of the sacrouterine ligament. Fifty-one females who underwent total laparoscopic hysterectomy for benign diseases were investigated.

Material and Methods: Macroscopic uterine parameters were determined during operation. First appearance of endometrium cells, complete disappearance of endometrial cells in the cervix and others were measured microscopically with reference to the external cervical orifice. Associations were described using odds ratio with 95% confidence interval and p-value <0.05.

Results: The region of the cervix, in which exclusively cervical glands are found, is relatively small but varies considerably around the mean (mean, 23.3 mm, range, 10 to 35 mm). In this cohort in a remnant cervical stump of 23 mm length, endometrial glands would be found in 51%. There was no correlation between full cervical length and uterine parameters but smaller uteri tended to be associated with deeper endometrial penetration.

Conclusion: There is a discrepancy between common definition and histological findings concerning the cervix uteri. Our findings indicate that the sacral uterine ligament is not suitable as an anatomic landmark for the laparoscopic supracervical hysterectomy operation. Regarding the distribution pattern of endometrial glands in the isthmic zone, a deep conical excision seems to better prevent subsequent spotting than a straight resection with thermocoagulation of the remaining cervical canal. (J Turk Ger Gynecol Assoc 2021; 22: 1-7)

Keywords: Endometrial glands, cervical glands, uterine isthmus, supracervical hysterectomy, spotting

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Introduction

In northern Europe 41.5% of hysterectomies are performed as a supracervical hysterectomy (1). Patients often prefer this method as only the diseased part of the uterus is removed, for example the corpus uteri in the case of heavy bleeding, and the risk of intraoperative complications, such as lesion of the ureter or intraoperative blood loss, is reduced. In addition, when preserving the cervical tissues, the operation can be performed more easily and faster. A further benefit is that the remaining cervical structures may be useful for fixation in case of later prolapse.

One disadvantage of supracervical hysterectomy is the risk of persistent spotting. The uterine isthmus is considered to be the border between corpus and cervix (2,3). Surgeons often use the sacral uterine ligament as a macroscopic landmark of the uterine isthmus (Figure 1) (4,5). The excision of the corpus uteri is frequently performed as a straight resection preserving the ligament, followed by thermocoagulation of the remaining cervical channel (4,5). Endo-loops and slings simplify this procedure (6). To date, few data concerning the histological structure of the isthmus have been reported; however, the data available show that the isthmic region is more than just the border between cervix and corpus uteri (2,3,7-9). Endometrial cells are regularly found coexistent with cervical stroma in the uterine isthmus (2,3,10). Therefore, it is not unexpected that spotting rates of up to 25% have been reported after supracervical hysterectomy using a straight resection above the sacral uterine ligament (11). Forty percent of patients suffering from spotting after laparoscopic supracervical hysterectomy (LSH) report a reduced quality of life (12). Spotting can be reduced by a conical excision of the cervical canal, but is nevertheless reported to persist in 6% of cases (10,13).

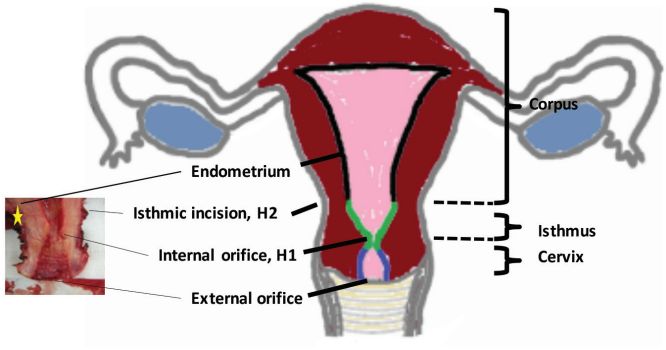

Figure 1. Anatomical subdivisions of uterus
The common anatomical doctrine is demonstrated schematically. The small picture on the left shows the lower uterine segment of one of our specimens. The yellow star indicates the sacrouterine ligament, used as the resection line in the case of laparoscopic supracervical hysterectomy

It is likely that the anatomical correlates are remnant endometrial glands. These glandular tissues will respond to female hormones and cyclic bleeding may be induced. If this hypothesis is true, the risk of later endometrial carcinoma cannot be excluded but appears to be very low. Only a few cases of new endometrial carcinoma in the remnant stump have been reported to date (14,15).

The aim was to determine the extension of the isthmic region and to investigate the extent of the explicit cervical part, as defined by the presence of cervical glands and the complete absence of endometrial glands. Additionally, we attempted to find a correlation between uterine length or weight and the tissue limits of the presence of endometrial glands.

Material and Methods

The uteri of 51 patients who underwent a complete hysterectomy were analysed in the Pathological Institute of Hagen Grevenbroich. Only patients with benign disease of the uterus were included in the study, including uterus myoma (n=13), adenomysosis (n=15), both myoma and adenomyosis (3) or other indications (n=20). Patients suffering from endometrial or cervical cancer were excluded.

Total uterus length and weight of the uterus were measured before further pathological investigations were undertaken. Intracaval length was reported, if measured intraoperatively (n=47).

The study was approved by the Institutional Ethical Committee University of Witten/Herdecke (approval number: 34/2011, date: 30.05.2011). Informed consent was obtained.

Histopathological reprocessing

A longitudinal median cut through the uterus was performed. The area of interest area of the cervix, that was the area between the external orifice and the area above the isthmical incision, was identified on one half of the uterus. Matched, longitudinal sections were obtained from the correlating area of the other uterus half.

These sections were prepared for further analysis (Figure 2a). If no endometrial glands were found in this first slide, the slide above was also analyzed (Figure 2b). The configuration at the interrupted areas was not always possible over the whole diameter of the cervix due sectioning artefact. The best configured parts were used for histological estimation of cervical and endometrial glands in these cases.

As a reference point for measurement, the perpendicular to the external orifice of cervix was used. Measurements were performed using an optical scale.

The distance between the external orifice of the cervical canal and the solely endometrial glands with absence of cervical glands was measured microscopically (Figure 3a, Table 1). The distance between the external orifice of the cervical canal

and the absence of endometrial glands was also determined (Figure 3a, Table 1). The length of the transition zone in which both type of glands (cervical and endometrial) were present, was derived from measurements (Table 1, Figure 3b).

Statistical analysis

Associations (length, intracaval length, weight of uterus vs. extension of endometrial cells) were described using odds ratios and 95% confidence intervals. P-values less than 0.05 were accepted as statistically significant. Statistical evaluation was performed with SPSS 18.0.2 (SPSS Inc., Chicago, IL, USA).

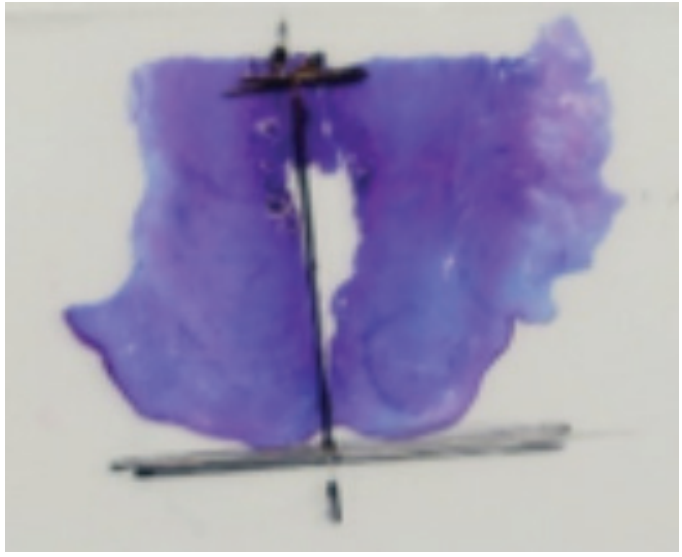

Figure 2a. Longitudinal section of uterine cervix (one slide needed)

A longitudinal section of uterine cervix is demonstrated. The first endometrial glands are present in this section. Thus, no second slide was needed. On the slide the cervical canal and reference line is marked for measurement

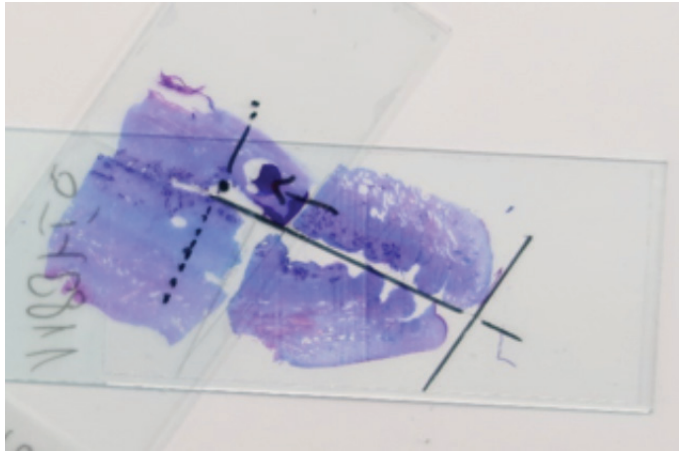

Figure 2b. Longitudinal section of uterine cervix (two slides needed)

In the first slide no endometrial glands were found. The second slide was positioned according to the line that marks cervical canal. The upper half was contiguous and could be used for measurement. The first endometrial cells were present more distally to the cervical canal than in Figure 2a

Results

The area of containing exclusively cervical glands was relatively small with a medium length of 23.3 mm (range, 10-35). In our study 17.6% of patients (9/51) had a cervix smaller than 20 mm (Figure 3a, b).

In this study the isthmic zone, characterized by the co-existence of both cervical and endometrial glands, had a medium depth of 2.8 mm (range, 0.5-8).

After supracervical hysterectomy the mean remaining cervical length was 2.3 cm. Measurement by ultrasound showed the cervical stump length to vary between 2.0 and 2.5 cm.

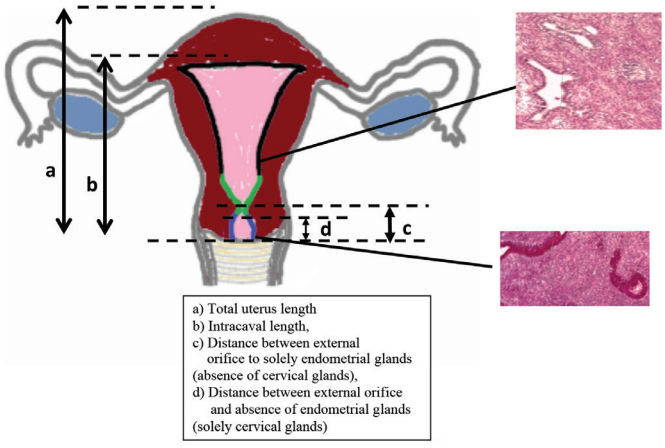

Figure 3a. Reference lines

Reference lines during the histological evaluation are demonstrated: (a) Total uterus length; (b) intracaval length; (c) distance between external orifice of the cervical channel and first exclusively endometrial gland area; (d) distance between external orifice of the cervical channel and area containing no endometrial glands

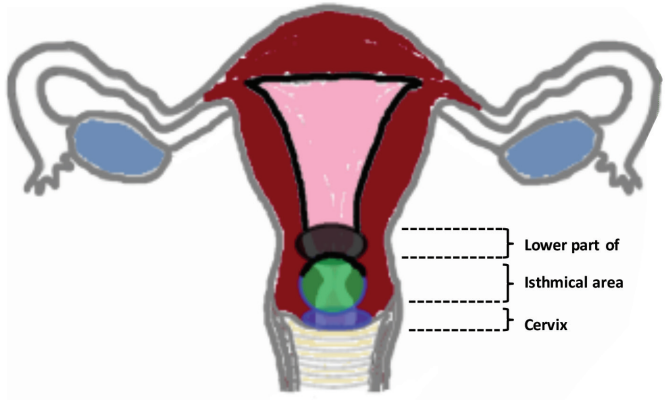

Figure 3b. Clinical zones of uterus

Schematic demonstration of the results of this study. Zones, rather than complex areas, were present in which specific cells were found

Table 1. Patient data and histological details

No	Age	Uterus length	Intracaval length	Weight	First EM gland	Beginning of EM	Transition zone
	Years	mm	mm	gr	mm	mm	mm
1	49	73	102	98	20	23	3
2	45	79	98	124	20	22	2
3	63	62	75	58	23	24	1
4	52	71	80	74	17	21	4
5	42	68	100	110	21	25	4
6	58	61	70	50	11	19	8
7	58	55	65	-	17	21	4
8	40	75	98	128	25	31	6
9	40	79	87	88	24	29	5
10	42	95	95	108	35	37	2
11	36	76	90	110	23	27	4
12	44	72	95	138	23	26	3
13	48	-	95	124	26.5	29	2.5
14	46	92	130	288	27	28	1
15	60	69	80	86	22	24	2
16	61	-	-	128	21	25	4
17	50	-	-	178	22.5	26	3.5
18	43	68	75	56	22	25	3
19	42	84	91	102	23.5	28	4.5
20	52	84	100	114	31.5	34	2.5
21	47	89	130	126	24	25	1
22	33	72	80	74	27.5	31	3.5
23	47	85	105	196	24.5	29	4.5
24	45	68	85	50	20.5	24	3.5
25	77	62	65	46	15.5	19	3.5
26	58	59	95	34	26	28	2
27	47	98	128	278	18.5	21	2.5
28	46	75	92	136	32	33	1
29	51	57	92	186	27	29	2
30	47	-	85	138	16	19	3
31	46	72	160	630	27	29	2
32	65	-	65	44	20.5	24	3.5
33	45	90	100	204	31	34	3
34	43	69	75	96	25.5	26	0.5
35	46	71	89	82	28	31	3
36	61	68	79	53	26.5	29	2.5
37	47	74	90	106	30	31	1
38	49	62	85	104	26.5	29	2.5
39	47	78	90	84	22	24	2
40	44	72	90	101	27.5	31	3.5
41	39	85	105	290	20.5	24	3.5
42	49	75	90	106	11.5	13	1.5
43	33	76	88	101	31	32	1

Table 1. Continued

No	Age	Uterus length	Intracaval length	Weight	First EM gland	Begin of EM	Transition zone
	Years	mm	mm	gr	mm	mm	mm
44	34	68	90	106	25.5	27	1.5
45	52	82	110	226	21	25	4
46	51	67	85	98	24.5	26	1.5
47	38	72	90	92	28	31	3
48	69	70	83	50	15	18	3
49	34	69	80	82	20	21	1
50	45	66	80	82	19	21	2
51	-	-	-	-	23	25	2
EM: Endometrial							

Endometrial glands were present in 51% of cases (Figure 3c). There was no correlation between the extension of endometrial glands and uterine length or weight. However; there was a tendency for small uteri to correlate with a deeper extension of endometrial glands into cervical tissues.

Discussion

The extent of the isthmic area is variable but tends to be small. In some cases, the areas containing solely cervical structures measured only 10 mm. It can be supposed that if the remaining cervical part following LSH measured on average 2.3 cm, endometrial glands would be found in 51% of cases. No correlation with other factors that could be estimated before the operation, such as uterine weight or length, was found.

Only a few studies have investigated the histophysiology of the uterine isthmus. In 1953, Frankl (2) complained of the ignorance of the necessity of a threefold division of the uterus into corpus, isthmus and cervix. He defined the isthmus as the "hollow space of the cranial part of the cervix" with a different histological structure, similar neither to corpus nor to cervix. In 1958, Ober described his findings of unexpected endometrial material after strict cervical surgery and proposed an area in which the characteristic cells of corpus and cervix coexist (3). He detailed these findings and characterised the uterine isthmus as an independent region with smaller and fewer endometrial glands and more compact stroma. Additionally, he defined lines, marking the borders between cervical part and isthmic part (inner cervical orifice, H1) and the isthmic part and the endometrial part (isthmic incision, H2, Figure 1). This threefold division is now part of common anatomic doctrine, but there is still a lack of knowledge concerning the definition, the extension and borders and especially the macroscopic landmarks of the transition zone. Hoogduin et al. (16) tried to identify the borders using immunohistochemical markers. These authors reported that all three parts of the uterus can be separately identified by immunohistochemical markers, and that the transition from cervical cells to the isthmic part is histologically abrupt.

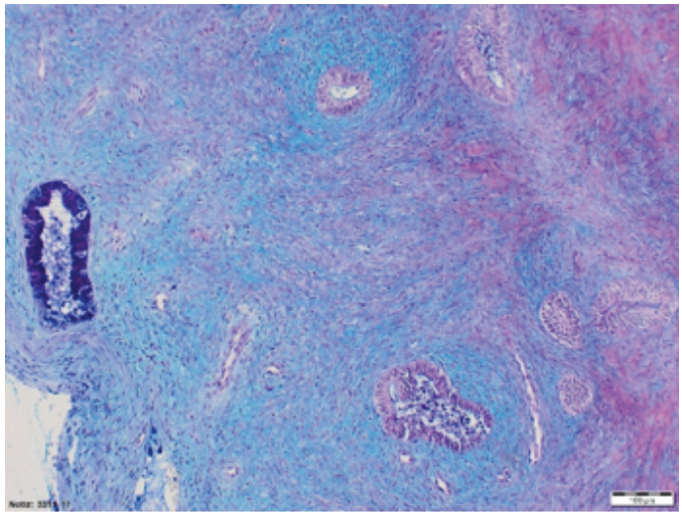

Figure 3c. Isthmic zone
Histological demonstration of the isthmus. Cervical glands (deep blue) and a few endometrial glands are coexistent

Few data concerning the risk of endometrial carcinoma following supracervical hysterectomy are available (14,15,17). This risk is generally considered to be unimportant. Most authors assume the implantation of endometrial cells during the removal process. Histologically, there is no consensus concerning the reaction of the isthmic endometrial glands to hormones (2,3,7). Our findings indicate that isthmic endometrial glands, at least in part, react to ovarian stimulation. It is very unlikely that endometrial carcinoma will occur after supracervical hysterectomy, but it cannot be completely ruled out.

In November 2014, the Food and Drug Administration (FDA) abandoned power morcellation for women having uterine fibrinoids due to the risk for spreading occult sarcoma. This rule was based on the finding that one of 458 operated patients suffered from occult leiomyosarcoma. A worldwide discussion concerning the sense of supracervical hysterectomy ensued and several surgeons returned to total hysterectomy. Meanwhile,

several studies showed that the risk for dissemination of occult malignant tissue during morcellation was much lower (one of 1,550 patients, 0.064%) compared to the risk acted on by the FDA (18-20). Most national societies developed guidelines to reduce the risk for intraoperative morcellation of sarcoma by giving detailed preoperative diagnostic rules. These developments have led to the widespread re-adoption of supracervical hysterectomy.

There are pros and cons that should be taken into account when considering the procedure to be undertaken; supracervical hysterectomy versus total hysterectomy. After total hysterectomy no more spotting will appear and if a complete removal, without further morcellation is possible, the risk of dissemination of occult sarcoma is very low. However, preparation is more complex, associated with a higher risk of bleeding and complications or postoperative infection and insufficient suture. In case of later descensus surgery, there is likely to be only low-quality vaginal tissue as fixation point.

In contrast, supracervical hysterectomy is associated with a lower complication rate and lower operation time, but there is a small (0.064%) risk of spreading sarcoma. New systems allow morcellation in a bag, which reduces the remaining risk. Finally, it should be mentioned that several studies could show that occult sarcoma found after morcellation are not associated with a worser outcome (19,20).

The preservation of the cervical stump has a positive effect on patients physical and psychological well-being, as women prefer the integrity of vagina and pelvic floor. Additionally, it offers a stable potential fixation area in case of later descensus surgery. The possibility of postoperative spotting rate should be discussed preoperatively. Spotting rates after a straight resection and coagulation of the remaining cervical canal vary between 6-25%. Banerjee et al. (21) analyzed spotting and infection rates over a six-month follow up after straight resection and coagulation versus conical excision and coagulation. The spotting rate after straight resection and bipolar coagulation was 21.6% (19/88) versus only 5.9% (5/85) after conical excision and coagulation. No difference concerning stump infection rate was found (straight resection 6.8% (6/88) vs 5.9% (5/85) for conical excision) (21).

Supracervical hysterectomy has not been performed optimally in cases of persistent spotting. This failure may be caused by the lack of a macroscopic landmark that indicates the correct resection area. This study has shown that there is a large variability concerning the extension and position of the uterine isthmus that makes it difficult to define a macroscopic landmark. In addition, our data suggests that the sacrouterine ligament is not a good landmark. In line with our findings, we refrain from defining surgical landmarks. Instead of landmarks we propose, in accordance with Schmidt et al. (13), a conical excision of the cervix but with the top of the conus positioned 10 mm away from the external orifice (Figure 4). In our opinion, this strategy better addresses the variance in isthmic area depth.

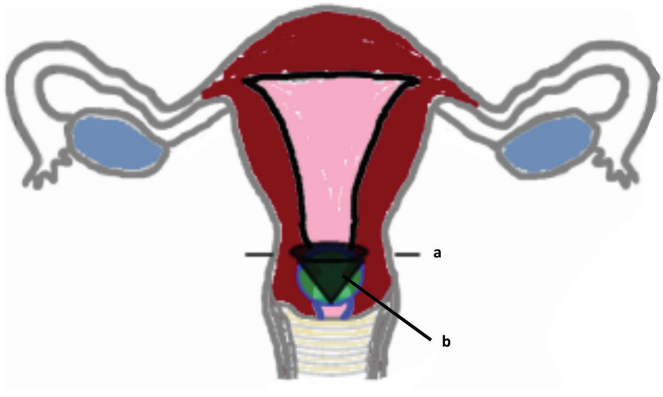

Figure 4. Schematic demonstration of conical excision
Former resection line (a) vs a proposed new resection zone (b): A conical excision of the lower uterine segment with preservation of only the external 10 mm of cervix is more likely to result in a resection of the complete isthmus

When conical deep cervical excision and minimising dissemination of sarcoma by precise diagnosis are practiced, we contend that LSH is still a very good method for removal of an uterus with benign disease.

Conclusion

The extension of the physiological cervix uteri, as defined histologically by the presence of exclusively cervical glands, is often small. The isthmic region, containing both endometrial glands and cervical glands, shows a large range of variability, which may vary further in individual patients with the female cyclus. Remnant cervical endometrial glands appear to be associated with a low risk for later endometrial cancer, but with a higher risk of spotting. Conical excision of the cervix may go some way to solving the problem of the remaining cervical endometrial glands.

Ethics Committee Approval: *The study was approved by the Institutional Ethical Committee University of Witten/Herdecke (approval number: 34/2011, date: 30.05.2011).*

Informed Consent: *Informed consent was obtained.*

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Author Contributions: *Surgical and Medical Practices: C.S., G.K.N.; Concept: C.S., M.B., E.W.; Design: C.S., M.B.; Data Collection or Processing: C.S., G.K.N., J.S.; Analysis or Interpretation: M.H., C.S., E.W.; Literature Search: E.W., C.S.; Writing: C.S., E.W.*

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References

1. Gimbel H, Settnes A, Tabor A. Hysterectomy on benign indication in Denmark 1988-1998. A register based trend analysis. Acta Obstet Gynecol Scand 2001; 80: 267-72.
2. Frankl O. On the physiology and pathology of the isthmus uteri. BJOG 1933; 40: 397-422.
3. Ober K, Schneppenheim P, Hamperl H, Kaufmann C. Die epithelgrenzen im bereich des isthmus uteri. Archiv für Gynäkologie 1958; 190: 346-83.
4. Bojahr B, Tchartchian G, Ohlinger R. Laparoscopic supracervical hysterectomy: a retrospective analysis of 1000 cases. JSLS 2009; 13: 129-34.
5. Kim DH, Lee ES, Park SD. A safer, simpler, classic intrafascial supracervical hysterectomy technique. JSLS 2005; 9: 159-62.
6. Hasson H. Cervical removal at hysterectomy for benign disease. Risks and benefits. J Reprod Med 1993; 38: 781-90.
7. Frick H, Leonhardt H, Starck D. Allgemeine Anatomie. Spezielle Anatomie I. vol 4 Thieme; 1992.
8. Netter FH. Atlas der Anatomie des Menschen. vol 2. Thienme; 1999.
9. Rauber A, Kopsch F. Anatomie des Menschen. 1987.
10. Banerjee C, Kaiser N, Hatzmann W, Reiss G, Schmitz J, Hellmich M, et al. Lower spotting rates after laparoscopic supracervical hysterectomy. Geburtshilfe Frauenheilkund 2010; 70: 798-802.
11. Ghomi A, Hantes J, Lotze EC. Incidence of cyclical bleeding after laparoscopic supracervical hysterectomy. J Minim Invasive Gynecol 2005; 12: 201-5.
12. Salfelder A, Lueken C, Bormann A, Gallinat CP, Moeller D, Busche W, et al. Scotland. Die suprazervikale hysterektomie in neuem licht. Frauenarzt 2003; 44: 1071-5.
13. Schmidt T, Eren Y, Breidenbach M, Fehr D, Volkmer A, Fleisch M, et al. Modifications of laparoscopic supracervical hysterectomy technique significantly reduce postoperative spotting. J Minim Invasive Gynecol 2011; 18: 81-4.
14. Patel S, Goodrich S. Endometrial adenocarcinoma following supracervical hysterectomy managed with radiation due to dabigatran etexilate. Mil Med 2014; 179: e351-3.
15. Turner T, Secord AA, Lowery WJ, Sfakianos G, Lee PS. Metastatic adenocarcinoma after laparoscopic supracervical hysterectomy with morcellation: A case report. Gynecol Oncol Case Rep 2013; 5: 19-21.
16. Hoogduin KJ, Hopman AN, Ramaekers FC, McCluggage WG, Smedts F. BCL2 and keratin 5 define the uterine-cervix-isthmus junction, a transition between endocervical and tubal-like epithelium. Int J Gynecol Pathol 2013; 32: 122-30.
17. Schneider A. Recurrence of unclassifiable uterine cancer after modified laparoscopic hysterectomy with morcellation. Am J Obstet Gynecol 1997; 177: 478-9.
18. Lieng M, Berner E, Busund B. Risk of morcellation of uterine leiomyosarcomas in laparoscopic supracervical hysterectomy and laparoscopic myomectomy, a retrospective trial including 4791 women. J Minim Invasive Gynecol 2015; 22: 410-4.
19. Siedhoff MT, Doll KM, Clarke-Pearson DL, Rutstein SE. Laparoscopic hysterectomy with morcellation vs abdominal hysterectomy for presumed fibroids: an updated decision analysis following the 2014 Food and Drug Administration safety communications. Am J Obstet Gynecol 2017; 216: 259.e1-6.
20. Bojahr B, De Wilde RL, Tchartchian G. Malignancy rate of 10,731 uteri morcellated during laparoscopic supracervical hysterectomy (LASH). Arch Gynecol Obstet 2015; 292: 665-72.
21. Banerjee C, Kaiser N, Hatzmann W, Reiss G, Schmitz J, Hellmich M, et al. Lower spotting rates after laparoscopic supracervical hysterectomy. Geburtshilfe Frauenheilkd 2010; 70: 798-802.

Telephone interview in urogynecology in the era of COVID-19 pandemic

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Abstract

Objective: During the Coronavirus disease-2019 (COVID-19) pandemic deferable access, including non-urgent outpatient visits, have been suspended. In our practice non-urgent routine visits for pelvic floor symptom assessment were discontinued and rescheduled, and telephone interview was performed. The aim was to evaluate patients' satisfaction for this alternative approach.

Material and Methods: Telephone interviews were conducted using a validated questionnaire to investigate pelvic floor symptoms. Patients were also asked if they had other symptoms or disorders, to identify patients who may need urgent hospital evaluation. At the end of the phone call, patients were asked to score their satisfaction with the telephone interview by grading their response to three questions from 0 (minimum) to 10 (maximum). The questions were: 1) "Was the telephone interview useful to check your state of health?"; 2) "Was the telephone interview an adequate healthcare tool in consideration of COVID-19 outbreak?"; 3) "Could the telephone interview replace the conventional visit?".

Results: Fifty-three patients were evaluated. All patients showed great satisfaction with telephone interview (Q1 median: 10, IQ range: 10-10) and were similarly positive in response to the second question (Q2 median: 10, IQ range: 10-10). Although fewer patients felt that telephone interview could replace conventional clinic visits most remained positive (Q3 median: 7; IQ range: 6-8).

Conclusion: This simple experience showed that phone interviews with validated questionnaires are appreciated and effective to safely perform interview of selected urogynecologic patients. (J Turk Ger Gynecol Assoc 2021; 22: 8-11)

Keywords: Telemedicine, prolapse surgery, anti-incontinence surgery, COVID-19, female pelvic medicine

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Introduction

Italy was one of the countries most severely affected by Coronavirus disease-2019 (COVID-19) in the World. Since hospitals present a high risk of infection - through COVID-19 patients and asymptomatic health workers - deferable access, including non-urgent clinical activities, have been suspended (1). Thus there was a need to develop alternative solutions, as the demand for healthcare does not disappear, to continue providing healthcare for the population. These solutions included mobile clinics and home-based care, thus reducing access to hospitals, exposure to gatherings of people and consumption of protective equipment (2). One solution is telephone interview that allows patients to remain in their own home. This can be suitable for the health-care of urological patients, as previous reports have shown it to be feasible and associated with good patients satisfaction (3). In female pelvic reconstructive surgery, telephone interview has been proposed for interview of patients undergoing midurethral tapes, as a screening tool to identify patients who need conventional clinical consultation (4,5). Recently, the Phone Study compared telephonic and clinic interview results after pelvic organ prolapse and anti-incontinence surgery in female patients and demonstrated the feasibility and reliability of a telemedical procedure (6). In the era of the COVID-19 pandemic, telephone interview may be even more

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appropriate, to limit the contagion. Therefore, we decided to perform telemedicine using a symptom related questionnaire for patients with non-urgent outpatient visits suspended due to COVID-19. We hypothesized that patients would appreciate this alternative approach to maintain care continuity. Specifically, we aimed to evaluate patients' satisfaction for telephone interview in terms of appropriateness and quality of healthcare service provided.

Material and Methods

This study was conducted in two university hospitals in Lombardy, the Italian region worst affected by COVID-19. Patients whose pelvic organ prolapse or anti incontinence postoperative clinical consultation was suspended due to COVID-19 lockdown, scheduled from March 16, 2020 to April 30, 2020, were involved. Interview visits were rescheduled, and in the meanwhile a telephone interview was performed. Telephone interviews were conducted through a modified version of the questionnaire validated by Balzarro et al. (6), investigating prolapse symptoms, urinary incontinence, sexual dysfunction, voiding difficulties, lower urinary tract symptoms and bowel dysfunction (Table 1). Patients were also asked if they had other symptoms or disorders, to identify patients who may need urgent hospital evaluation. At the end of the phone call, patients were asked to score their satisfaction with the telephone interview on a scale of 0 (minimum) to 10 (maximum). The patients were asked only three questions, which were: 1) "Was the telephone interview useful to check your state of health?"; 2) "Was the telephone interview an adequate healthcare tool in consideration of COVID-19 outbreak?"; 3) "Could the telephone interview replace the conventional visit?".

Statistical analysis

Data obtained during the telephone interview assessment were statistically analyzed with JMP 9.0 (SAS, Cary, NC, USA). Continuous data are presented as median and interquantile range for non-parametric variables and as mean ± standard deviation for parametric variables, while non-continuous data as number (percentage). This study was considered exempt from local ethical committee approval as it only involved standard clinical practices.

Results

In total 53 patients answered telephone calls. Mean patient age was 65.6±9.3 years. Surgical technique performed and duration of interview was very variable since it involved the activity of two different Institutions. In the cohort, prolapse repair surgery represented the main indication for interview (79.2%). All patients showed great satisfaction with telephone interview. Median scores for each question were: Q1 10; Q2 10; and Q3 7 (Figure 1). We also collected positive feedback about the supporting and reassuring effect of our calls. None of the patients described symptoms that required an urgent conventional evaluation.

Discussion

This study evaluated the feasibility of telephone interview for patients in which non-urgent, conventional, post-operative check-up was suspended due to COVID-19. All patients showed great satisfaction with telephone interview and considered telephone interviews as an adequate tool for replacement of routine hospital visits during COVID-19 lockdown. Moreover, most were positive about the possibility of telephone interview

Table 1. Question checklist

	Question	Not at all	Sometimes	Yes
1	Do you have sensation of bulging/protrusion from vagina?			
2	If bulging/protrusion is present does it bother you?			
3	Do you experience difficulty in emptying the bladder?			
4	Have you resumed your sexual life?			
5	Have you experienced dyspareunia?			
6	Do you experience urinary incontinence?			
7	Is your urinary incontinence in connection with physical efforts like laughing, coughing, sneezing, exercising or lifting something heavy?			
8	Is your urinary incontinence related to sudden, intense urge to urinate followed by an involuntary loss of urine?			
9	Do you need to urinate often?			
10	Do you need to urinate during the nighttime?			
11	Have you had bladder infection since last medical control?			
12	Do you experience difficulty in emtying the bowel?			
	Other to notify:			

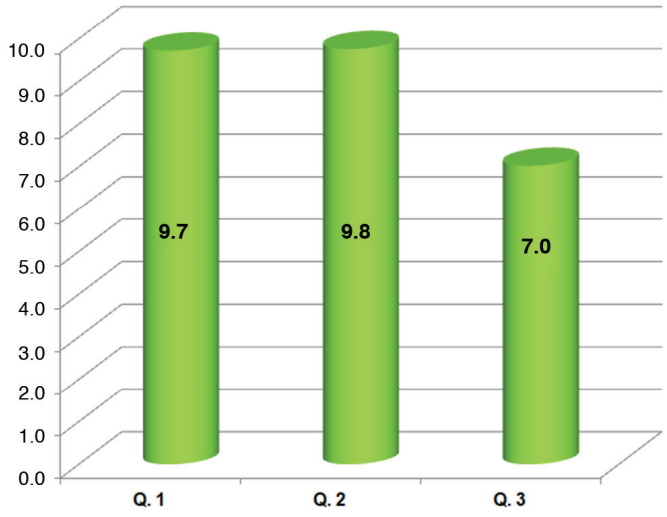

Figure 1. Mean satisfaction score with the telephone interview in a range from 0 (minimum) to 10 (maximum) with the following three questions: Q1) "was the telephone interview useful to check your state of health?"; Q2) "was the telephone interview an adequate healthcare tool in consideration of COVID-19 outbreak?"; Q3) "could the telephone interview replace the conventional visit?"
COVID-19: Coronavirus disease-2019

replacing conventional check-up. We also collected positive feedback about the supporting and reassuring effect of our calls.

The COVID-19 pandemic has significantly affected the way providers care for patients, and urogynecology is no exception. Despite no clear guidelines on the use of telemedicine in female pelvic medicine and reconstructive surgery, recently an effort was made to provide guidance regarding management of common outpatient urogynecology scenarios during the pandemic (7). While surgeons usually feel compelled to check postoperative patients, there is growing evidence that patient stratification based on perioperative risk and postoperative risk may decrease the total number of conventional visits to enhance physical distancing (6). Recently, a pre-COVID-19 pandemic randomized clinical trial showed that telephone interview after pelvic floor surgery resulted in non-inferior patient satisfaction, without differences in clinical outcomes or adverse events (8).

This study confirmed that telemedical interview may allow healthcare continuity and may be particularly appropriate in the era of the COVID-19 pandemic. Phone interviews using a validated questionnaire appeared to be appreciated and an effective tool to safely perform interview of patients with functional disorders. This approach involves several advantages. The first is the "forward triage", the capability to sort patients with urgent need for care from those who can safely postpone the conventional clinical examination. In this way it was possible to reduce community exposure for people with functional urologic disorders, which may potentially be looking for alternative health providers, such as inappropriate emergency room access or general practitioners consultation. This is extremely important, since these patients are often elderly and frail, and more prone to develop severe and life-threatening complications in case of COVID-19. Moreover, for functional disorders, telephone interview can often be adequate to address symptoms referred by patients, and set up an initial diagnostic or therapeutic path. At the same time, this procedure, acting as a screening tool, avoids neglecting serious conditions that require medical evaluation. One more aspect of the telephone interview is the capability to provide additional social support for patients living through the pandemic, who are lonely, anxious and fearful of abandonment. It was evident from our cohort that these emotions are widespread among patients compelled to stay home for days or weeks due to COVID-19 lockdown. The very high levels of satisfaction expressed by our patients can probably be explained in part by the human aspect of the conversation, besides clinical contents, that was particularly appreciated. Moreover, this telephonic approach reduced the consumption of protective equipment and clinicians' exposure to contagion. This is particularly relevant considering that large numbers of health care workers having to quarantine would impact the capacity of health institutions to face the current COVID-19 emergency. Lastly, this telehealth approach could be conducted by quarantined health workers, with both patient and clinician at home, greatly optimizing resources and permitting uninterrupted care of established patients (9).

Conclusion

Although this is a relatively small-scale pilot experience, we believe that the concept of telephone interview using validated clinical tools, such as checklists and questionnaires, could be potentially widespread to manage the arising limitations during the COVID-19 pandemic successfully. This is an efficient and low-resource solution that guarantees vulnerable patients' care while protecting both patients and health providers.

Ethics Committee Approval: *Since the study only involved standard clinical tools, ethics committee approval was not obtained. The study was conducted in accordance with the declaration of Helsinki.*

Informed Consent: *It wasn't obtained.*

Peer-review: *Externally peer-reviewed.*

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References

1. Ferreira R, McGrath M, Wang Y, Sener A, Siemens DR, Braga LH. How to prioritize urological surgeries during epidemics: Lessons learned from the Toronto SARS outbreak in 2003. Can Urol Assoc J 2020; 14: E159-60.
2. Patel HV, Srivastava A, Tabakin AL, Jang TL, Singer EA. The patient-urologist relationship in the COVID-19 era and beyond. Can Urol Assoc J 2020; 14: E271-3.
3. Witherspoon L, Fitzpatrick R, Patel P, Flannigan R, Roberts MT, Krakowsky Y, et al. Clinical pearls to managing men's health conditions during the COVID-19 pandemic. Can Urol Assoc J 2020; 14: E161-6.
4. Glavind K, Bjørk J. Follow-up of the tension-free vaginal tape operation in everyday practice. Acta Obstet Gynecol Scand 2013; 92: 349-51.
5. Jefferis H, Muriithi F, White B, Price N, Jackson S. Telephone follow-up after day case tension-free vaginal tape insertion. Int Urogynecol J 2016; 27: 787-90.
6. Balzarro M, Rubilotta E, Trabacchin N, Mancini V, Costantini E, Artibani W, et al. A prospective comparative study of the feasibility and reliability of telephone follow-up in female urology: the patient home office novel evaluation (PHONE) study. Urology 2020; 136: 82-7.
7. Grimes CL, Balk EM, Crisp CC, Antosh DD, Murphy M, Halder GE, et al. A guide for urogynecologic patient care utilizing telemedicine during the COVID-19 pandemic: review of existing evidence. Int Urogynecol J 2020; 31: 1063-89.
8. Thompson JC, Cichowski SB, Rogers RG, Qeadan F, Zambrano J, Wenzl C, et al. Outpatient visits versus telephone interviews for postoperative care: a randomized controlled trial. Int Urogynecol J 2019; 30: 1639-46.
9. Hollander JE, Carr BG. Virtually Perfect? Telemedicine for Covid-19. N Engl J Med 2020; 382: 1679-81.

Associated factors with neonatal near miss in twin pregnancies in a public referral maternity unit in Brazil

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Abstract

Objective: The aim was to analyze the factors associated with neonatal near miss (NNM) in twin pregnancies in a public referral maternity unit in Brazil.

Material and Methods: This retrospective, cross-sectional study included 697 twin newborns. Cases of fetal and neonatal deaths were excluded. Neonates were divided into those meeting NNM criteria (5 min Apgar score <7, birth weight <1,500 g, gestational age at delivery <32 weeks, use of mechanical ventilation or congenital malformation, transfer before 28 days of life) and those who did not. In the bivariate analysis, the chi-square and Fisher's exact tests were used. Variables with a p-value ≤0.20 were subjected to the multiple analyses, which followed the Poisson regression model.

Results: The cohort consisted of 130 (18.7%) neonates meeting NNM criteria and 567 (81.3%) with no NNM criteria after multiple analyses, the following variables were associated with NNM: no previous pregnancy, prevalence ratio (PR): 1.38 [95% confidence interval (CI), 1.03-1.85]; >3 previous pregnancies, PR: 1.93 (95% CI, 1.38-2.69); premature rupture of membranes, PR: 1.50 (95% CI, 1.70-2.12); intrauterine growth restriction, PR: 2.28 (95% CI, 1.53-3.33); premature labor, PR: 1.63 (95% CI, 1.13-2.35); resuscitation in the delivery room, PR: 1.80 (95% CI, 1.24-2.62); and transfusion of blood products, PR: 4.44 (95% CI, 3.14-6.28).

Conclusion: The study findings indicate that having had 0 or >3 previous pregnancies, premature rupture of the membranes, intrauterine growth restriction, resuscitation in the delivery room, premature labor, and transfusion of blood products were associated with NNM in twin pregnancies. (J Turk Ger Gynecol Assoc 2021; 22: 12-21)

Keywords: Near miss healthcare, morbidity, twin pregnancy, perinatal care

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Introduction

The incidence of twin pregnancies is increasing worldwide (1), with a mean incidence of 13.1/1,000 live births (LB) (2). In three decades, in the USA, the birth rate of twins has risen 76%, attributed to the increase in the average maternal age and the emergence of new assisted reproduction technologies (1,3). In Brazil, between 2011-2014, twin births represented 1.13% of LB, with 0.98% in the state of Ceará (4). Studies have shown an association between twinning and a five minute (5 min) Apgar score <7, low birth weight, neonatal intensive care unit admission, and a consequent increase in neonatal morbidity and mortality rates (5-7).

The concept of neonatal near miss (NNM) is used for obstetrical events that almost resulted in the death of newborns from 0 to 28 days of life (8). However, globally accepted criteria for

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identifying cases have not yet been defined, which is a challenge for the identification and real estimate of its impact (9). Such criteria will depend on the production of evidence to enable the identification of really serious cases, the possibility of easy data collection in terms of clinical care and the applicability to different scenarios (10,11).

Research of various concepts of NNM showed associations with twin pregnancy (12-15). This term refers to cases of newborns who almost died as a result of some serious complications (11). The identification of these cases and application of this concept to the neonatal population is an important tool in the identification of deficiencies in the health services provided to the mother-baby dyad (8,16).

The choice of researching twin pregnancies, regardless of their classification by chorionicity, started from the need to identify and recognize the reality of twin deliveries that occurred in our institution, taking into account the absence of studies of this population. Considering the importance and scarcity of NNM studies, especially in twin pregnancies, this study aimed to analyze the factors associated with NNM in twin pregnancies in a public, tertiary care, referral maternity unit for high-risk pregnancies in Brazil.

Material and Methods

This retrospective cross-sectional study identified twins among all live newborns born at the maternity school between January 2016 and December 2018. NNM cases were those that met at least one of the criteria published by Da Silva et al. (13): birth weight <1,500 g, 5 min Apgar score <7, use of mechanical ventilation, gestational age at delivery <32 weeks, and presence of congenital malformations.

Exclusion criteria were: cases of early and late neonatal death; transfers (before 28 days of life); conjoined twins; abortion (gestational age <20 weeks, weight <500 g); delivery of the first twin outside the hospital environment; and patients for whom information was incomplete or missing from the medical records.

In the assessment of sociodemographic characteristics, pre-existing clinical conditions, prenatal care, complications during pregnancy, and childbirth, the pregnant women were the unit of analysis. They were classified as NNM (those for whom at least one twin met one or more NNM criteria) or non-NNM (those for whom neither twin met one or more NNM criteria). For variables related to the newborn's health conditions, the neonates were the unit of analysis. For each mother, there were one or two newborns since one could be excluded from the analysis due to fetal or neonatal death; the woman and her surviving newborn could still be included.

A form containing questions directed to the following study variables was constructed for data collection: maternal sociodemographic characteristics, obstetric clinical history, conditions related to pregnancy, NNM criteria, maternal outcome, conditions related to delivery, conditions related to the newborn, criteria for NNM and neonatal outcome. This instrument was reviewed before the data collection started, a pilot test was carried out in order to identify flaws and test its applicability. The information was extracted from medical records and/or other medical records as a declaration of LB.

The following near miss indicators were also calculated and adapted to the neonatal context by Pileggi et al. (8) and Pileggi-Castro et al. (17): NNM rate, severe neonatal outcome rate, early neonatal mortality index, NNM/neonatal death ratio, and early neonatal mortality rate.

The Federal University of Ceará Local Ethic Committee approved the study under the certificate of presentation for ethical appraisal (approval number: 04091418.7.0000.5050). Consent was also obtained when the participants signed the Term of Faithful Depositary prior to the data collection.

Statistical analysis

The data were analyzed using SPSS version 23.0 (SSP Inc., Chicago, IL, USA). For univariate analysis, chi-square and Fisher's exact tests were used, when appropriate. Missing information was not used in the significance calculation. Variables with values of $p \leq 0.20$ were tested again using multiple analyses and a Poisson regression model with robust variance, avoiding possible confounding variables. The prevalence ratio (PR) and 95% confidence intervals (CI) were calculated. The variables with values of $p<0.05$ on the multiple analyses were included in the final regression model.

Results

Between January 2016 and December 2018, a total of 14,870 births occurred at the surveyed institution and of these 904 (6%) were due to twin pregnancies. One hundred and 120 twins were excluded because they met the exclusion criteria. Of them, 87 (28 fetal deaths, 42 early neonatal deaths, and 17 late neonatal deaths) were excluded. In the population eligible for analysis, 567 (81.3%) live newborns met NNM criteria and 130 (18.7%) did not meet NNM criteria for a total of 697 twin studies.

Based on the proposed quality monitoring and neonatal care indicators (8), a NNM rate of 171.9/1,000 LB was obtained, an early neonatal mortality rate of 55.6/1,000 LB, index early neonatal mortality of 22.2%, severe neonatal outcome rate of 227.5/1,000 LB, and 2.2 cases of NNM for each neonatal death.

None of the variables related to maternal sociodemographic characteristics showed a statistically significant association with the NNM cases. Most of the women surveyed were 19-34 years of age (77.8%), were multiparous, and had no previous history

of abortion. Obesity (17%) was the most prevalent pre-existing condition, followed by chronic arterial hypertension (9.3%) and syphilis (2.7%). Among the clinical conditions studied, diabetes mellitus (p=0.034), kidney disease (p=0.019), and thyroid disease (p=0.013) were significantly different between the two groups (Table 1).

Table 1. Sociodemographic characteristics and pre-existing clinical conditions of mothers of twins considered neonatal near miss and non-neonatal near miss.

Variable	Neonatal near miss	Non-neonatal near miss	Total	p
	n (%)	n (%)	n (%)	
Marital status				
With partner	75 (77.3)	212 (79.1)	287 (78.6)	0.744[a]
With no partner	21 (21.7)	54 (20.2)	75 (20.6)	
Not registered	1 (1)	2 (0.7)	3 (0.8)	
Maternal education				
<8 years	28 (28.9)	88 (32.8)	116 (31.8)	0.433[a]
≥8 years	69 (71.1)	177 (66)	246 (67.4)	
Not registered	0 (0)	3 (1.2)	3 (0.8)	
Race				
White	6 (6.2)	9 (3.3)	15 (4.1)	0.258[b]
Black	0 (0)	5 (1.9)	5 (1.4)	
Mixed	89 (91.7)	249 (92.9)	338 (92.6)	
Not registered	2 (2.1)	5 (1.9)	7 (1.9)	
Maternal age				
≤18 years	3 (3.1)	24 (8.9)	27 (7.4)	0.168[a]
19-34 years	79 (81.4)	205 (76.5)	284 (77.8)	
≥35 years	15 (15.5)	39 (14.6)	54 (14.8)	
Parity				
0	36 (37.1)	90 (33.6)	126 (34.5)	0.142[a]
1-2	39 (40.2)	136 (50.7)	175 (48)	
≥3	22 (22.7)	42 (15.7)	64 (17.5)	
Previous vaginal delivery				
Yes	46 (47.4)	97 (36.2)	143 (39.2)	0.052[a]
No	51 (52.6)	171 (63.8)	222 (60.8)	
Number of previous cesarean sections				
1	14 (14.4)	57 (21.3)	71 (19.4)	0.151[a]
≥2	6 (6.2)	26 (9.7)	32 (8.8)	
None	77 (79.4)	185 (69)	262 (71.8)	
Number of previous abortions				
0	73 (75.3)	212 (79.1)	285 (78.1)	0.433[a]
≥1	24 (24.7)	56 (20.9)	80 (21.9)	
Nutritional status				
Low weight	1 (1)	7 (2.6)	8 (2.2)	0.339[b]
Eutrophic	18 (18.6)	75 (28)	93 (25.5)	
Overweight	21 (21.6)	69 (25.7)	90 (24.7)	
Obese	19 (19.6)	41 (15.3)	60 (16.4)	
Not registered	38 (39.2)	76 (28.4)	114 (31.2)	

Table 1. Continued

Variable	Neonatal near miss	Non-neonatal near miss	Total	p
	n (%)	n (%)	n (%)	
Chronic arterial hypertension				
Yes	9 (9.3)	25 (9.3)	34 (9.3)	0.988[a]
No	88 (90.7)	243 (90.7)	331 (90.7)	
Diabetes mellitus				
Yes	5 (5.2)	3 (1.1)	8 (2.2)	0.034[b]
No	92 (94.8)	265 (98.9)	357 (97.8)	
Kidney disease				
Yes	4 (4.1)	1 (0.4)	5 (1.4)	0.019[b]
No	93 (95.9)	267 (99.6)	360 (98.6)	
Heart disease				
Yes	1 (1)	7 (2.6)	8 (2.2)	0.687[b]
No	96 (99)	261 (97.4)	357 (97.8)	
Smoker				
Yes	3 (3.1)	6 (2.2)	9 (2.5)	0.705[b]
No	94 (96.9)	262 (97.8)	356 (97.5)	
Chronic respiratory disease				
Yes	2 (2.1)	3 (1.1)	5 (1.4)	0.612[b]
No	95 (97.9)	265 (98.9)	360 (98.6)	
HIV/AIDS				
Yes	0 (0)	3 (1.1)	3 (0.8)	0.568[b]
No	97 (100)	265 (98.9)	362 (99.2)	
Syphilis				
Yes	1 (1)	9 (3.4)	10 (2.7)	0.301[b]
No	96 (99)	259 (96.6)	355 (97.3)	
Obesity				
Yes	21 (21.6)	41 (15.3)	62 (17)	0.153[a]
No	76 (78.4)	227 (84.7)	303 (83)	
Thyroid disease				
Yes	6 (6.2)	3 (1.1)	9 (2.5)	0.013[b]
No	91 (93.8)	265 (98.9)	356 (97.5)	

[a]Chi-square test, [b]Fisher's exact test, HIV: Human immunodeficiency virus

Prenatal care, complications during pregnancy and childbirth data are shown in Table 2.

Preterm birth occurred in 69.4% and 66.9% of newborns weighed <2,500g. In addition to the variables used to identify cases of NNM, cesarean delivery, 1 min Apgar score >7, transfusion of blood products, resuscitation in the delivery room, and length of stay >28 days were also significantly associated with NNM (Table 3).

The variables used as a defining criterion for NNM outcome were removed from the multiple analyses. The variables number of previous pregnancies; premature rupture of membranes; intrauterine growth restriction; resuscitation in the delivery room; premature labor and transfusion of blood products, were associated with NNM and remained in the final model (Table 4).

Discussion

The assessment of severe neonatal morbidity is a new health indicator contributing to the identification of factors in the health system requiring remedial action, assessment of care quality, and guidance for decision-making by health managers and providers (11). These measures may contribute to the reduction of neonatal mortality rates in addition to allowing

Table 2. Characteristics of prenatal care, complications during pregnancy, and delivery in mothers of twins considered neonatal near miss versus non-neonatal near miss

Variable	Neonatal near miss	Non-neonatal near miss	Total	p
	n (%)	n (%)	n (%)	
Prenatal care				
Yes	96 (99)	266 (99.3)	362 (99.2)	1.000[b]
No	1 (1)	2 (0.7)	3 (0.8)	
Number of prenatal consultations				
≤6	45 (46.4)	61 (22.8)	106 (29.1)	<0.001[a]
>6	51 (52.6)	205 (76.5)	256 (70.1)	
Not registered	1 (1)	2 (0.7)	3 (0.8)	
Chorionicity				
Dichorionic/diamniotic	52 (53.6)	149 (55.6)	201 (55.1)	0.588[b]
Monochorionic-diamniotic	35 (36.1)	97 (36.2)	132 (36.1)	
Monochorionic-monoamniotic	6 (6.2)	10 (3.7)	16 (4.4)	
Not registered	4 (4.1)	12 (4.5)	16 (4.4)	
Pre-eclampsia/eclampsia				
Yes	23 (23.7)	69 (25.7)	92 (25.2)	0.692[a]
No	74 (76.3)	199 (74.3)	273 (74.8)	
Severe sepsis				
Yes	0 (0)	2 (0.7)	2 (0.5)	1.000[b]
No	97 (100)	266 (99.3)	363 (99.5)	
Premature labor				
Yes	54 (55.7)	84 (31.3)	138 (37.8)	<0.001[a]
No	43 (44.3)	184 (68.7)	227 (62.2)	
Amniotic fluid disorder				
Yes	7 (7.2)	8 (3)	15 (4.1)	0.080[b]
No	90 (92.8)	260 (97)	350 (95.9)	
Gestational diabetes				
Yes	7 (7.2)	28 (10.4)	35 (9.6)	0.354[a]
No	90 (92.8)	240 (89.6)	330 (90.4)	
Urinary tract infection				
Yes	46 (47.4)	112 (41.8)	158 (43.3)	0.337[a]
No	51 (52.6)	156 (58.2)	207 (56.7)	
Transvaginal bleeding				
Yes	11 (11.3)	38 (14.2)	49 (13.4)	0.482[a]
No	86 (88.7)	230 (85.8)	316 (86.6)	
Intensive care unit admission				
Yes	2 (2.1)	8 (3)	10 (2.7)	1.000[b]
No	95 (97.9)	260 (97)	355 (97.3)	
Premature rupture of membranes				
Yes	33 (34)	51 (19)	84 (23)	0.003[a]
No	64 (66)	217 (81)	281 (77)	
Fetal growth restriction				
Yes	26 (26.8)	24 (9)	50 (13.7)	<0.001[a]
No	71 (73.2)	244 (91)	315 (86.3)	

Table 2. Continued

Variable	Neonatal near miss	Non-neonatal near miss	Total	p
	n (%)	n (%)	n (%)	
Twin-to-twin transfusion syndrome				
Yes	7 (7.2)	2 (0.7)	9 (2.5)	0.002[b]
No	90 (92.8)	266 (99.3)	356 (97.5)	
Hemorrhagic syndrome in pregnancy				
Yes	2 (2.1)	4 (1.5)	6 (1.6)	0.658[b]
No	95 (97.9)	264 (98.5)	359 (98.4)	
[a]Chi-square test, [b]Fisher's exact test				

Table 3. Birth conditions, newborn health, and neonatal care among twins considered neonatal near miss and non-neonatal near miss

Variable	Neonatal near miss	Non-neonatal near miss	Total	p
	n (%)	n (%)	n (%)	
Gestational age at delivery				
20-27 weeks	21 (16.1)	0 (0)	21 (3)	<0.001[b]
28-31 weeks	40 (30.8)	0 (0)	40 (5.7)	
32-33 weeks	17 (13.1)	54 (9.5)	71 (10.2)	
34-36 weeks	41 (31.5)	311 (54.9)	352 (50.5)	
≥37 weeks	11 (8.5)	202 (35.6)	213 (30.6)	
Mode of delivery				
Vaginal	22 (16.9)	47 (8.3)	69 (9.9)	0.003[a]
Cesarean section	108 (83.1)	520 (91.7)	628 (90.1)	
Presentation				
Head	80 (61.5)	352 (62)	432 (62)	0.882[b]
Pelvic/podalic	47 (36.2)	195 (34.4)	242 (34.7)	
Transverse	3 (2.3)	19 (3.4)	22 (3.2)	
Not registered	0 (0)	1 (0.2)	1 (0.1)	
Birth order				
1st twin	68 (52.3)	286 (50.4)	354 (50.8)	0.701[a]
2nd twin	62 (47.7)	281 (49.6)	343 (49.2)	
Sex				
Male	74 (56.9)	294 (51.9)	368 (52.8)	0.296[a]
Female	56 (43.1)	273 (48.1)	329 (47.2)	
Birth weight (grams)				
≤500	1 (0.6)	0 (0)	1 (0.1)	<0.001[b]
501-1,000	23 (12.9)	0 (0)	23 (3.3)	
1,001-1,500	33 (18.5)	0 (0)	33 (4.7)	
1,501-2,500	102 (57.3)	308 (59.3)	410 (58.8)	
>2,500	19 (10.7)	211 (40.7)	230 (32.9)	
Neonatal size				
Appropriate for gestational age	95 (73.1)	441 (77.8)	536 (76.9)	0.354[b]
Small for gestational age	35 (26.9)	123 (21.7)	158 (22.7)	
Large for gestational age	0 (0)	3 (0.5)	3 (0.4)	

Table 3. Continued

Variable	Neonatal near miss	Non-neonatal near miss	Total	p
	n (%)	n (%)	n (%)	
Apgar score, 1 min				
<7	52 (40)	42 (7.4)	94 (13.5)	<0.001[a]
≥7	78 (60)	525 (92.6)	603 (86.5)	
Apgar score, 5 min				
<7	9 (6.9)	0 (0)	9 (1.3)	<0.001[b]
≥7	121 (93.1)	567 (100)	688 (98.7)	
Mechanical ventilation				
Yes	65 (50)	0 (0)	65 (9.3)	<0.001[a]
No	65 (50)	567 (100)	632 (90.7)	
Congenital malformation				
Yes	72 (55.4)	0 (0)	72 (10.3)	<0.001[a]
No	58 (44.6)	567 (100)	625 (89.7)	
Transfusion of blood products				
Yes	56 (43.1)	3 (0.5)	59 (8.5)	<0.001[a]
No	74 (56.9)	564 (99.5)	638 (91.5)	
Resuscitation in the delivery room				
Yes	62 (47.7)	59 (10.4)	121 (17.4)	<0.001[a]
No	68 (52.3)	506 (89.2)	574 (82.3)	
Not registered	0 (0)	2 (0.4)	2 (0.3)	
Upper airway aspiration				
Yes	90 (69.2)	391 (69)	481 (69)	0.952[a]
No	40 (30.8)	176 (31)	216 (31)	
Gastric aspiration				
Yes	47 (36.2)	224 (39.5)	271 (38.9)	0.479[a]
No	83 (63.8)	343 (60.5)	426 (61.1)	
Length of hospital stay				
0-7 days	15 (11.6)	383 (67.7)	398 (57.2)	<0.001[a]
8-28 days	32 (24.6)	161 (28.4)	193 (27.7)	
>28 days	83 (63.8)	22 (3.9)	105 (15.1)	

[a]Chi-square test, [b]Fisher's exact test

the calculation of ratio/rates between deaths and cases of near miss, better specifying health care indicators for the most severe cases (8,17).

According to the classification criteria applied (13), 2.2 cases of near miss were identified for each neonatal death, which was higher than the findings in earlier Brazilian studies that addressed the feasibility of the NNM concept (13,14). The scarcity of research of NNM in twins makes it difficult to compare these rates and rates within this specific population, yet our findings suggest that twins have worse outcomes for severe neonatal morbidity.

Variables, such as low maternal education level, race/skin color of mixed mothers, women without partners, and lower socioeconomic classes are widely debated since they show an association with increased neonatal morbidity and mortality (14,18). Although twin pregnancies are associated with Brazilian regions with the highest human development index, higher education level, and high maternal age (>35 years), this study showed no relationship between NNM and maternal socioeconomic and demographic conditions (4). In line with this finding, two studies in Brazilian maternity hospitals also reported no such association (15,19). It should be noted that

Table 4. Final model of factors associated with neonatal near miss in twin pregnancies

Variable	PR[a]	95% CI[b]	p[c]
Number of previous pregnancies			
0	1.38	1.03-1.85	0.032
1-2	1	-	-
≥3	1.93	1.38-2.69	<0.001
Premature rupture of membranes			
Yes	1.50	1.07-2.12	0.020
No	1	-	-
Intrauterine growth restriction			
Yes	2.28	1.56-3.33	<0.001
No	1	-	-
Premature labor			
Yes	1.63	1.13-2.35	0.008
No	1	-	-
Resuscitation in the delivery room			
Yes	1.80	1.24-2.62	0.002
No	1	-	-
Transfusion of blood products			
Yes	4.44	3.14-6.28	<0.001
No	1	-	-

[a]PR: Prevalence ratio, [b]CI: Confidence interval, [c]Values were estimated by Poisson multiple regression analysis

the surveyed population included only twin pregnancies born in a public maternity hospital.

Prematurity occurs in about 50% of Brazilian twin deliveries and is almost 5.0 times more prevalent when compared to singleton pregnancies, being up to 12 times higher in extremely preterm infants. Premature labor is also associated with a 5 min Apgar <7. In premature infants aged 32-36 weeks the risk of Apgar <7 at the 5th min is 2.5 and in newborns <32 weeks this risk may be 30 times greater, which may be related to adverse neonatal outcomes (4). The rate of prematurity (69.4%) was higher than that reported in previous Brazilian studies, a fact that can be attributed to the research being performed exclusively in a reference maternity hospital for high-risk pregnancies. Other studies also reported an association between NNM and preterm birth, 1 min Apgar score <7, premature rupture of membranes, and neonatal resuscitation, as was found in the current cohort (13,14).

The accuracy of the first minute Apgar score has been investigated as a diagnostic test or marker for the presence of asphyxia and indicates that less than half of newborns with low Apgar scores are asphyxiated, according to the gasometric criteria (20). The purpose of this study was not to analyze hypoxemic events, as the 5 min Apgar score <7 was used as an NNM defining criterion, as it is widely used as marker of neonatal morbidity.

In a Brazilian study, pregnant women with inadequate prenatal care were more susceptible to having spontaneous premature labors. The prenatal coverage in Brazil has advanced in the last 15 years. However, access failure, late start, and incomplete execution of procedures still occur. Data reflect gaps in assistance, in addition to the historical situation of regional and socioeconomic inequality that is present in the country (21,22). Lima et al. (19) found an association between <6 prenatal consultations and NNM, which resulted in a four times greater risk of NNM. In our findings, 99.2% of women attended at least one consultation and 70.1% attended >6 consultations, showing increased access, but this did not guarantee better care quality, especially in higher-risk pregnancies. Assessment of the number of consultations alone does not result in better assistance. Assessing the quality of the provided services was not the objective of this study.

This series revealed no relationship between birth order and NNM. Much has been discussed about the influence of birth order on worse neonatal outcomes, specifically that the second twin has worse perinatal outcomes (4). Other authors have demonstrated that if the birth conditions between the first

and second twins are uniform, the birth order will not influence perinatal outcomes (23).

Monochorionic pregnancies are generally associated with a higher risk of perinatal complications and perinatal morbidity and mortality compared to dichorionic pregnancies, since they have specific obstetric complications, such as twin-to-twin transfusion syndrome (TTTS), twin anemia-polycythemia sequence (TAPS), and twin reversed arterial perfusion (TRAP) sequence (6,24). Studies have shown that monochorionic twins have a higher incidence of prematurity, premature labor, olygohydramnios/polyhydramnios, intrauterine growth restriction, lower maternal age, use of mechanical ventilation, lower gestational age at delivery, low birth weight, and seven times greater chance of perinatal mortality (25-27). In this study, no significant association was observed between chorionicity and NNM, thus diverging from the results mentioned above. More recent studies have suggested a downward trend in perinatal morbidity and mortality in monochorionic pregnancies when there is an early diagnosis and intensive surveillance during prenatal care (28-32). The research institution in the present study, a maternity school, has a fetal medicine service, and the professionals who work there have expertise in handling high-risk pregnancies. We suggest that this may have resulted in their being no effect of chorionicity in our cohort.

Cesarean delivery plays an important role in reducing perinatal risks and, consequently, increasing newborn survival (30). Although mode of delivery did not appear in the explanatory model of factors associated with NNM, the significant association of NNM with vaginal delivery found here can be explained by the fact that twin pregnancies are associated with several maternal and fetal complications that require therapeutic cesarean section or a greater risk of neonatal death in cases of vaginal delivery (18,31).

Among the predictor variables that remained in the final model, blood product transfusion appeared with a PR of 4.44 (95% CI, 3.14-6.28) and a neonatal resuscitation PR of 1.80 (95% CI, 1.24-2.62). These two variables were also identified in a study that investigated the same outcome and made up some of the management criteria studied by the Latin American Center for Perinatology, because of their association with NNM (19). Efforts should be concentrated to avoid preventable complications of twin pregnancy, avoiding, for example, premature labor to achieve better neonatal outcomes since premature birth directly influences neonatal morbidity (32).

To verify the correlation between advanced maternal age and parity with NNM, Martinelli et al. (12) found an association between advanced maternal age and NNM in nulliparous women (odds ratio: 1.62; 95% CI, 1.05-2.50) and multiparous women (odds ratio: 1.51; 95% CI, 1.20-1.91) when women aged 20-29 years were compared. Although the research above included singleton and multiple pregnancies, the data corroborated the current findings.

The present study included a considerable sample and is a pilot in this institution, examining twin pregnancies with NNM outcomes. However, its limitations need to be recognized. Some complications related to monochorionic pregnancies (TTTS and TAPS) were not properly assessed and therefore were not presented here. The absence of a detailed description of ultrasound examinations hindered this analysis. There was no definition of chorionicity, even after macroscopic analysis of the placenta, in 4.4% of cases. In other situations, the lack of data in the medical records prevented the collection of some variables, such as body mass index in 31.2% of patients. These data can be extrapolated to aproximate the reality in many Brazilian and Latin American maternity hospitals: a tertiary-level institution that serves people of low socioeconomic level and does not feature highly complex technology such as equipment used to perform fetoscopy and laser therapy.

The results presented here show that it is necessary to target health policies, especially actions aimed at the socially vulnerable in the population and in conditions of high-risk pregnancies. There is also a need for studies that can compare NNM outcomes in twin versus single pregnancies, including research into management criteria recently listed by the Latin American Center for Perinatology to better investigate the association of these factors in twin pregnancies. This direct comparison was not made in this study. It is also necessary to reassess these results in the long term, especially after offering the best technological equipment. The use of protocols that identify neonatal morbidity criteria can help better guide twin care.

Conclusion

These findings allow us to understand that risk of NNM in twinning is associated with the number of previous pregnancies, premature labor, premature rupture of membranes, intrauterine growth restriction, need for resuscitation in the delivery room, and transfusion of blood products. In clinical practice, these results can assist with the implementation of protocols and measures to identify high-risk situations during obstetric and neonatal care and improve neonatal results. Future studies into the topic are essential, especially to better assess conditions related to chorionicity and outcomes between first and second twins.

Ethics Committee Approval: *The Federal University of Ceará Local Ethic Committee approved the study under the certificate of presentation for ethical appraisal (approval number: 04091418.7.0000.5050).*

Informed Consent: *Consent was also obtained when the participants signed the Term of Faithful Depositary prior to the data collection.*

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References

1. Hall JG. Twinning. Lancet 2003; 362: 735-43.
2. Smits J, Monden C. Twinning across the developing world. PLoS One 2011; 6: e25239.
3. Martin JA, Hamilton BE, Osterman MJK, Driscoll AK, Drake P. Births: final data for 2016. Natl Vital Stat Rep 2018; 67: 1-55.
4. Santana DS, Souza RT, Surita FG, Argenton JL, Silva CM, Cecatti JG. Twin pregnancy in Brazil: A profile analysis exploring population information from the national birth e-registry on live births. Biomed Res Int 2018; 2018: 9189648.
5. Santana DS, Surita FG, Cecatti JG. Multiple pregnancy: epidemiology and association with maternal and perinatal morbidity. Rev Bras Ginecol Obstet 2018; 40: 554-62.
6. Ferriman E, Stratton S, Stern V. Twin pregnancy. Obstet Gynaecol Reprod Med 2018; 28: 221-8.
7. Santana DS, Silveira C, Costa ML, Souza RT, Surita FG, Souza JP, et al. Perinatal outcomes in twin pregnancies complicated by maternal morbidity: evidence from the WHO multicountry survey on maternal and newborn health. BMC Pregnancy Childbirth 2018; 18: 449.
8. Pileggi C, Souza JP, Cecatti JG, Faúndes A. Neonatal near miss approach in the 2005 WHO global survey Brazil. J Pediatr (Rio J) 2010; 86: 21-6.
9. Kale PL, Mello-Jorge MHP de, Silva KS da, Fonseca SC. Neonatal near miss and mortality: factors associated with life-threatening conditions in newborns at six public maternity hospitals in Southeast Brazil. Cad Saude Publica 2017; 33: e00179115.
10. Avenant T. Neonatal near miss: a measure of the quality of obstetric care. Best Pract Res Clin Obstet Gynaecol 2009; 23: 369-74.
11. Say L. Neonatal near miss: a potentially useful approach to assess quality of newborn care. J Pediatr (Rio J) 2010; 86: 1-2.
12. Martinelli KG, Gama SGN, Almeida AHV, Pacheco VE, Neto ETS. Advanced maternal age and factors associated with neonatal near miss in nulliparous and multiparous women. Cad Saude Pública 2019; 35: 1-13.
13. Da Silva AAM, Leite ÁJM, Lamy ZC, Moreira MEL, Gurgel RQ, Cunha AJLA da, et al. Neonatal near miss in the birth in Brazil survey. Cad Saude Publica 2014; 30 Suppl 1: S1-10.
14. Brasil DRPA, Vilela MBR, França KEX, Sarinho SW. Neonatal morbidity near miss in tertiary hospitals in a capital of Northeast Brazil. Rev Paul Pediatr 2019; 37: 275-82.
15. Silva GA, Rosa KA, Saguier ESF, Henning E, Mucha F, Franco SC. A populational based study on the prevalence of neonatal near miss in a city located in the South of Brazil: prevalence and associated factors. Rev Bras Saúde Matern Infant 2017; 17: 159-67.
16. Santos JP, Pileggi-Castro C, Camelo JS, Silva AA, Duran P, Serruya SJ, et al. Neonatal near miss: A systematic review. BMC Pregnancy Childbirth 2015; 15: 320.
17. Pileggi-Castro C, Camelo JS, Perdoná GC, Mussi-Pinhata MM, Cecatti JG, Mori R, et al. Development of criteria for identifying neonatal near-miss cases: analysis of two WHO multicountry cross-sectional studies. BJOG 2014; 121 Suppl 1: 110-8.
18. Lansky S, Friche AA, Lima, Silva AAM, Campos D, Bittencourt SDA, et al. Birth in Brazil survey: neonatal mortality profile, and maternal and child care. Cad Saude Publica 2014; 30 Suppl 1: S192-207.
19. Lima THB, Katz L, Kassar SB, Amorim MM. Neonatal near miss determinants at a maternity hospital for high-risk pregnancy in Northeastern Brazil: a prospective study. BMC Pregnancy Childbirth 2018; 18: 401.
20. Pereira DN, Rocha VLL, Procianoy RS, Azeredo RCM, Kersting D, Cardozo A, et al. Evaluation of umbilical cord pH and its relationship with Apgar score in term newborn infants. J Pediatr (Rio J) 1996; 72: 139-42.
21. Viellas EF, Domingues RMSM, Dias MAB, da Gama SGN, Filha MMT, da Costa JV, et al. Prenatal care in Brazil. Cad Saude Publica 2014; 30 Suppl 1: S1-15.
22. Nunes JT, Gomes KRO, Rodrigues MTP, Mascarenhas MDM. Quality of prenatal care in Brazil: review of published papers from 2005 to 2015. Cad Saude Colet 2016; 24: 252-61.
23. Costa HLFF, Rocha ACO, Galvão AF, Souza JA, Rigaard ACO, Costa LOBF. Is the prognosis of the second twin worse? Rev Bras Ginecol Obstet 1998; 20: 261-4.
24. Cheong-See F, Schuit E, Arroyo-Manzano D, Khalil A, Barrett J, Joseph KS, et al. Prospective risk of stillbirth and neonatal complications in twin pregnancies: systematic review and meta-analysis. BMJ 2016; 354: i4353.
25. Manso P, Vaz A, Taborda A, Silva IS. Chorionicity and perinatal complications in twin pregnancy: a 10 years case series. Acta Med Port 2011; 24: 695-8.
26. Machado M, Teixeira EL, Ferreira LM, Rodrigues F, Henriques R, Afonso E. Perinatal outcome in relation to chorionicity in twin pregnancy. Acta Med Port 2017; 30: 12-6.
27. Machado RCA, Brizot ML, Miyadahira S, Francisco RPV, Krebs VLJ, Zugaib M. Intrauterine growth restriction in monochorionic-diamniotic twins. Rev Assoc Med Bras (1992) 2014; 60: 585-90.
28. Mackie FL, Morris RK, Kilby MD. The prediction, diagnosis and management of complications in monochorionic twin pregnancies: The OMMIT (Optimal Management of Monochorionic Twins) study. BMC Pregnancy Childbirth 2017; 17: 153.
29. Ozler S, Guler B. Twin pregnancy outcome of our clinic: A three-year follow up. Ann Med Res 2019; 26: 2892-8.
30. Giglio MRP, Lamounier JA, Neto OLM. Obstetric delivery and risk of neonatal mortality in Goiânia in 2000, Brazil. Rev Saude Publica 2005; 39: 350-7.
31. Cardoso PO, Alberti LR, Petroianu A. Neonatal and maternal morbidity related to the type of delivery. Cien Saude Colet 2010; 15: 427-35.
32. Dolgun ZN, Inan C, Altintas AS, Okten SB, Sayin NC. Preterm birth in twin pregnancies: Clinical outcomes and predictive parameters. Pak J Med Sci 2016; 32: 922-6.

Prevention of preterm delivery by cervical cerclage; a comparison of prophylactic and emergency procedures

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Abstract

Objective: Prophylactic or emergency type cervical cerclage procedures are being used for treatment of cervical insufficiency. The aim was to review and compare the outcomes of these cerclage types and identify factors affecting outcomes.

Material and Methods: Retrospective review of seventy-five patients in whom transvaginal cervical cerclage procedures were performed over a seven-year period in a tertiary referral center.

Results: Twenty seven of 75 (36%) patients were in the emergency cerclage group and 48 (64%) of them were in the prophylactic group. Mean body mass index (BMI), hospitalization time and gestational week at cerclage were significantly higher, whereas latency period was significantly shorter for the emergency group. Mean gestational ages at delivery were 35.6±4.5 and 33.6±5.9 weeks in the prophylactic and emergency groups, respectively (p=0.117). Delivery rates under 34th gestational week were 20.8% and 37.0% in the prophylactic and emergency groups, respectively (p=0.175). Birthweight, and delivery ≥34th gestational week was higher in the prophylactic group, whereas complication rate was higher in the emergency group, but these differences were not significant. High BMI was associated with more deliveries before 34-week in the prophylactic group. Pre-cerclage cervical length was shorter in patients who delivered before 34 gestational weeks at delivery.

Conclusion: Prophylactic and emergency cerclage procedures have comparable results regarding gestational week at delivery. High BMI and low pre-cerclage cervical length may have adverse effects on success of cerclage procedures. (J Turk Ger Gynecol Assoc 2021; 22: 22-8)

Keywords: Cervical insufficiency, cervical cerclage, preterm birth, neutrophil-lymphocyte ratio

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Introduction

Cervical insufficiency can be described as an inability of cervix uteri to retain the pregnancy in the absence of objective signs of labor, for example due to normal uterine contraction, especially in the second trimester. It has a particular clinical importance since preterm birth and prematurity-related risks are high in this group of patients. The incidence is reported to be around 1% in the general obstetric population, but this rate is 8% in women with second trimester pregnancy loss (1). The etiology of cervical insufficiency is not clear but risk factors include antecedent cervical surgeries such as conization, repeated dilatation and curretage, congenital uterine anomalies, in utero exposure to the synthetic estrogen, diethylstilbestrol and, possibly, the most important risk factor is a history of cervical insufficiency in previous pregnancies (2). Bed rest, activity restriction and vaginal pessaries are non-surgical treatment modalities for cervical insufficiency and the effectiveness of these modalities has been evaluated previously (3-5). Activity restriction was reported to be ineffective in one study (6). Moreover, a higher risk of preterm delivery has been reported in women advised to restrict activity. In singleton pregnancies diagnosed with short cervix, expectant management was compared with vaginal pessaries and pessaries were shown

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to be more effective at reducing delivery under 34 gestational weeks. However, in twin pregnancies, vaginal pessary was not superior to expectant management in preventing delivery under 34 gestational weeks in a contemporary publication. Due to a lack of consensus in identifying the optimal non-surgical treatment, these modalities are generally discouraged (2).

Cervical cerclage procedures can be performed transabdominally or transvaginally. Transabdominal approaches should be reserved for patients with cervical anatomical disturbances, such as trachelectomized patients, and also for patients with repetitive failure of transvaginal cerclage that resulted in pregnancy loss. McDonald- and Schirodkar-type transvaginal cervical cerclage are the best known and most widely performed and both are equally effective (7).

Indication for cerclage can be based on medical history or as a result of findings uncovered during physical examination often requiring emergency cerclage procedures. The American College of Obstetrician and Gynecologists (ACOG) define indications for prophylactic cerclage as painless cervical dilatation or a requirement for cervical cerclage in a prior pregnancy. ACOG guidelines recommend that indications for emergency cerclage include painless cervical dilatation in the second trimester and cervical length under 25 mm with a history of preterm birth before 34 gestational weeks in a prior pregnancy (2).

Success of these cerclage procedures in preventing preterm delivery may be affected by a range of clinical parameters and patient characteristics. The aim of this study was to analyze and compare outcomes of prophylactic and emergency cerclage performed in a tertiary referral center. A further aim was to delineate factors that can affect the efficiency of cervical cerclage which may include body mass index (BMI), pre-cerclage cervical length and neutrophil-lymphocyte ratio.

Material and Methods

Patients

Cervical cerclage procedures performed between January 2012 and February 2019 were reviewed retrospectively from hospital records. Pregnancy and labor information were obtained by telephone-based questioning, when hospital records were incomplete. ACOG recommendations on cerclage indications were taken as guidelines. Prophylactic or history based cerclage was applied between 11-14 gestational weeks, after first trimester screening tests, for patients with history of cervical insufficiency in previous pregnancy. Emergency cerclage was performed for patients with painless cervical dilatation in the second trimester and also for patients with preterm birth history and diagnosis of short cervix in the current pregnancy. Cervical cerclage procedures were not performed in the presence of regular uterine contractions, active vaginal bleeding, chorioamnionitis, fetal anomaly, rupture of membranes and dilated cervix beyond 3 cm.

Ethics

This study was approved by the Başkent University Faculty of Medicine Institutional Ethical Committee on 05/14/2019, with the approval number of KA19/168. Informed consent of patients was obtained before cervical cerclage procedures.

Interventions

Pre-cerclage cervical length was measured by transvaginal ultrasound (TVUS), with empty bladder. At least three measurements were taken and the mean value was calculated. The degree of cervical dilatation was also measured by TVUS, under sterile conditions. McDonald cervical cerclage was applied to all patients and Schirodkar type cerclage was not used in this study population. Sterilization was achieved by the application of povidone iodine to the vagina and cervix under sedoanalgesia. The anterior portion of the cervix was grasped with an oval clamp and sutured at the twelve, nine, six and three o'clock positions. Non-absorbable braided suture material was used for this (Cervix-set B. Braun Surgical S.A.). Prolapsed membranes were relocated by placing and inflating a pediatric Foley catheter into the cervical canal in those patients with a dilated cervix. Prophylactic antibiotic, intramuscular progesterone and indomethacine were given to all patients, postoperatively. Postoperative complications were defined as massive vaginal bleeding, chorioamnionitis and premature rupture of membranes. Hospitalization time was defined as the period from operation until discharge.

Statistical analysis

Statistical analysis was performed using SPSS, version 17.0 (IBM Inc., Chicago, IL, USA). If continuous variables distributed normally, they are described as mean ± standard deviation ($p>0.05$ in Kolmogorov-Smirnov test or Shapiro-Wilk $n<30$) and if continuous variables did not distribute normally, they are described as median (range). Continuous variables were compared using Student's t test when normally distributed and using Mann-Whitney U test when did not distribute normally. Categorical variables were compared between groups by chi-square or Fisher's exact test. Values of $p<0.05$ were considered to indicate statistical significance.

Results

During the study period, a total of 89 cervical cerclage procedures were performed in this tertiary center. Twin pregnancies and pregnancies that ended before 21st week of gestation were excluded from the study (Figure 1). As a result, 75 patients with singleton pregnancies and diagnosis of cervical

insufficiency were included in the study. Twenty-seven of 75 patients were in the emergency cerclage group and 48 were in the prophylactic group. Mean BMI value, hospitalization time and gestational week of cerclage application were significantly higher in the emergency group compared to prophylactic group. Latency period, which is from cerclage week to delivery week was significantly shorter for the emergency cerclage patients (14.2±6.5 vs 21.7±4.8 weeks; p<0.001). Nonetheless, there were no statistically significant differences between the two groups regarding other clinical and demographic parameters (Table 1). The effect of BMI, preoperative cervical length and neutrophil-lymphocyte ratio on the week of delivery under and above 34 gestational weeks were evaluated. In the prophylactic cerclage group, patients who gave birth before 34 gestational weeks had significantly higher BMI values than the values of those giving birth after 34 gestational weeks (28.2±4.4 vs 25.0±4.2; p=0.04). There was a similar trend in the emergency cerclage group but the difference was not significant (31.8±10.6 vs 28.0±3.4; p=0.186) (Table 2). In the prophylactic group, mean pre-cerclage cervical length of patients who delivered before and after at 34 gestational weeks was not different; 30.9±5.3 mm vs 35.1±7.9 mm (p=0.117) respectively. In the emergency group these values were 9.6±6.3 mm vs 16.6±6.7 mm (p=0.136), respectively (Figure 2, 3).

In the emergency cerclage group, in patients with dilated cervix, the proportion giving birth before 34 weeks was 70%. This proportion dropped to 30% in the group with a diagnosis of short cervix only in the absence of cervical dialtation. However, the difference was still not significant (p=0.120; Table 3).

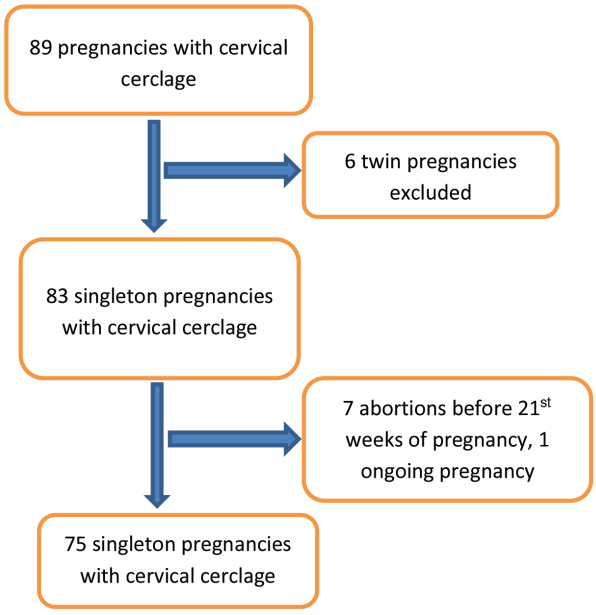

Figure 1. Flow diagram of the study

Discussion

The main determinant of success for a cervical cerclage procedure is the capability of preventing preterm birth and related adverse outcomes. In this cohort the prophylactic cerclage group had a higher mean gestational age, higher birth rate above 34 gestational weeks, higher mean gestational weight, and lower complication rate than the emergency

Table 1. Characteristics and perinatal outcomes of patients in emergency and prophylactic cervical cerclage patients

	Prophylactic	Emergency	p
Age (years)	29.9±4.4	31.7±4.4	0.104
Body mass index (kg/m^2)	25.7±4.4	29.4±7.1	0.006
Type of pregnancy spontaneous	75%, (n=36)	81.5%, (n=22)	0.438
Assisted reproductive techniques	25%, (n=12)	18.5%, (n=5)	
Mean hospitalization time (days)	1.02 days	5.1 days	0.003
Mean gestational week at cervical cerclage	13.9±1.7	19.4±3.2	0.000
Mean gestational week at birth	35.6±4.5	33.6±5.9	0.117
Mean time from cerclage to labor (in weeks)	21.7±4.8	14.2±6.5	0.000
Mean pre-cerclage neutrophile-lymphocyte ratio	3.5±0.8	3.8±1.2	0.196
Complication rate	2.1%, (n=1)	7.4%, (n=2)	0.368
Gestational week at delivery			
>34 weeks	20.8%, (n=10)	37%, (n=10)	0.175
≤34 weeks	79.2%, (n=38)	63%, (n=17)	
NICU need	37%, (n=10)	21.3%, (n=10)	0.178
Birth weights (grams)	2,842±957	2,475±1193	0.149

NICU: Neonatal intensive care unit

cerclage group. However, the mean gestational week at delivery did not differ between the two groups.

Gestational weeks completed and perinatal outcome have been compared previously in history-based and ultrasound-based cervical cerclage patients (8,9). Gluck et al. (10) compared the obstetric outcomes of patients admitted with cervical dilatation or shortened cervical length to history-indicated cerclage patients and gestational week at delivery and birthweights were similar for both groups. Liddiard et al. (11) also did not find any significant difference in gestational week, birthweight, live birth rate or requirement for neonatal intensive care unit (NICU) between emergency and prophylactic cerclage groups (11). In the same study, the complication rate in emergency cerclage patients was higher than in the prophylactic cerclage group, but it should be noted that approximately half of the patients in the emergency group had twin pregnancies and at least 3 cm cervical dilatation at admittance. In a recently published meta-analysis, which also included the Gluck et al. (10) and Liddiard et al. (11) studies, birth week and birth weights were found to be significantly lower and the risk of membrane rupture higher in the emergency cerclage group (12). In our cohort, age and pregnancy types were similar in both groups, but BMI values were significantly higher amongst emergency cerclage patients (p=0.006). Mean gestational week at delivery and mean birth weight tended to be higher in the prophylactic cerclage group, whereas complication rate and delivery under 34 gestational weeks tended to be greater in the emergency cerclage group.

Table 2. Pre-cerclage cervical length, BMI, and neutrophil-lymphocyte ratio by gestational age at delivery

	>34 weeks	≤34 weeks	p
Prophylactic cerclage group			
Mean pre-cerclage cervical length (mm)	30.9±5.3	35.1±7.9	0.117
Mean BMI (kg/m^2)	28.2±4.4	25.0±4.2	0.04
Neutrophil-lymphocyte ratio	3.5±0.9	3.2±0.7	0.342
Emergency cerclage group			
Mean pre-cerclage cervical length (mm)	9.6±6.3	16.6±6.7	0.136
Mean BMI (kg/m^2)	31.8±10.6	28.0±3.4	0.186
Neutrophil-lymphocyte ratio	4.0±1.4	3.5±0.9	0.411
BMI: Body mass index			

Table 3. Gestational week at delivery according to presence or absence of cervical dilatation, in emergency cerclage patients

Birth week	No dilatation	Dilatation	p
>34 weeks	30%	70%	0.120
≤34 weeks	64%	35%	-

Only hospitalization time was significantly different between the prophylactic and emergency groups.

In emergency cases, there is a process that has already started and is ongoing; short or dilated cervix was recognized as the sign of an impending threat of cervical insufficiency. In prophylactic cases, however, there is usually a well-known history of cervical insufficiency in a previous pregnancy, so both patient and physician are well-prepared for clinical situations and required treatment options in an on-going pregnancy. It is reasonable to assume that forestalling a process that has not yet started is clinically easier than forestalling one that has already started. The differences observed between the two groups in our study may be partially explained this way. Nevertheless, since differences were not significant regarding delivery week, birthweight, complication rate, and requirement for NICU

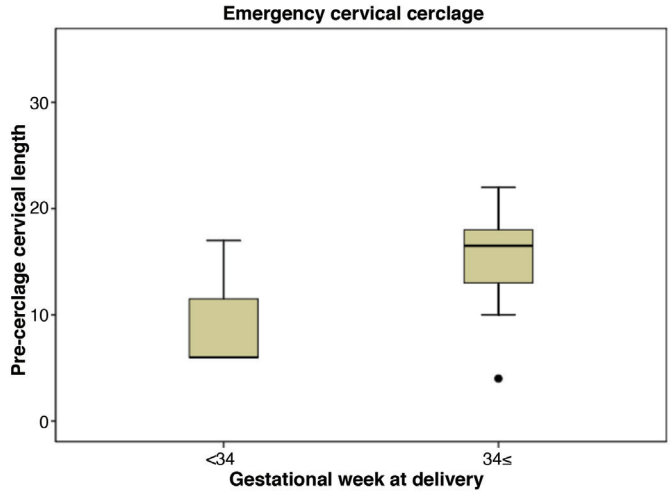

Figure 2. Pre-cerclage cervical length and gestational week at delivery in the emergency cerclage group

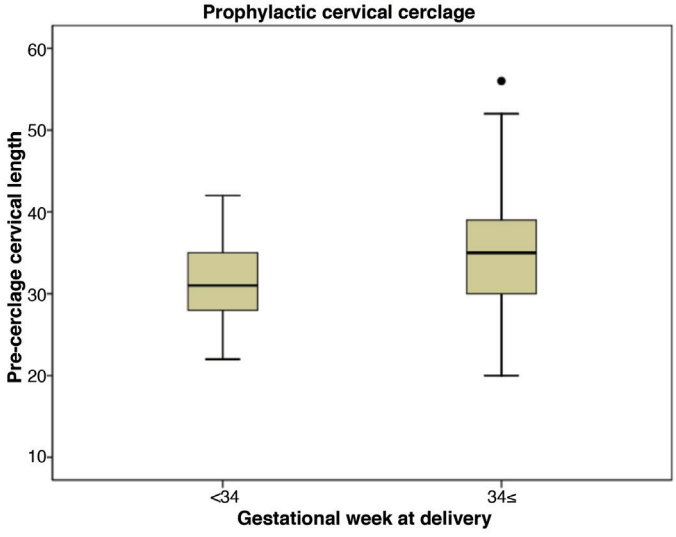

Figure 3. Pre-cerclage cervical length and gestational week at delivery in the prophylactic cerclage group

admission, we can conclude that outcomes of both cerclage types are similar and comparable. Latency period from cerclage to delivery was significantly higher in the prophylactic group, but gestational week at cerclage was higher in the emergency group, as expected. Since the mean cerclage week was significantly earlier (13.9 weeks) in the prophylactic group compared with 19.4 in the emergency group the longer latency period of the prophylactic group can be partially attributed to this difference.

A cervical cerclage procedure is recommended, with evidence level IA, for patients who had spontaneous preterm birth or had been diagnosed cervical insufficiency in previous pregnancies and have a cervical length under 25 mm in their current pregnancy (1). Berghella and Mackeen (13) published a meta-analysis including four randomized controlled trials and concluded that patients with a history of cervical insufficiency can be safely followed by serial TVUS cervical length measurement. This study concluded that cerclage procedures as a result of medical history may be unnecessary and may be reduced. A retrospective study of Brown et al. (8) found approximately 50% of patients with history did not require cerclage when followed by serial TVUS measurements. Moreover, the obstetrical and perinatal outcomes were similar between history-based and ultrasound-based cerclage groups. The main aim of serial cervical length measurements in patients with a history of cervical insufficiency is to reduce unnecessary cerclage procedures and related complications. In this study, ACOG's criteria were followed and cervical cerclage performed between 11-14 weeks of pregnancy in patients with a history of cervical insufficiency in a previous pregnancy. Cervical lengths were measured just before the procedure by TVUS. Mean cervical length of patients delivered at and after 34 gestational weeks did not differ from that of deliveries under 34 gestational week in the prophylactic cerclage group. There was also no difference in mean cervical length in the emergency cerclage group when comparing deliveries before and after 34 gestational weeks. As a result, an increase in pre-cerclage cervical length was associated with improvement of gestational week at delivery, although differences were not significant in this study. In patients requiring emergency cerclage, those who were admitted with cervical dilatation had a greater proportion within deliveries before 34 gestational weeks compared to the emergency cerclage patients without cervical dilatation, although this was again not significant. We suggest that the small sample sizes may have made our findings unreliable, otherwise it is highly probable that improvement of gestational week at delivery is directly proportional with pre-cerclage cervical length.

High BMI has been associated with various adverse pregnancy and obstetric outcomes. The effect of BMI on cerclage procedures has also been studied. Suhag et al. (14) investigated the effect of pre-pregnancy BMI on the success of history-indicated and ultrasound-indicated cerclage and reported no effect of BMI. In another retrospective observational study, no association was found between BMI and latency period (15). Interestingly, Schirodkar-type cerclage was reported to be superior to McDonald-type in obese patient groups in terms of better gestational week at delivery (16). One study showed an inverse proportion between BMI and gestational week at delivery in history-indicated cerclage patients (17). In the present study the mean BMI was significantly greater in the emergency cerclage group compared to the prophylactic cerclage patients. This difference may be due to the general adverse effect of higher BMI on obstetrical outcomes. There was a significant inverse relationship between gestational week at delivery and BMI in the prophylactic cerclage patients. A similar, but non-significant, trend was observed in the emergency cerclage patients. In general, high BMI values appeared to have a negative effect on cerclage efficiency. This should be taken into account when counseling patients, pre-procedurally.

The neutrophil-lymphocyte ratio is accepted as an indicator of the presence of pro-inflammatory processes. The prognostic value of this parameter has been investigated in chronic and acute inflammatory conditions and oncologic disease (18,19). The utility of neutrophil-lymphocyte ratio has been investigated in obstetric conditions including ovarian torsion and preeclampsia (20-21). Since delivery is a pro-inflammatory process, one can speculate that preterm delivery is also such a process and the neutrophil-lymphocyte ratio may be useful in predicting early delivery. A relationship has been reported between increase in neutrophil-lymphocyte ratio and delivery under 28 gestational weeks in patients with recurrent cervical cerclages (22). In our study, there was no difference in neutrophil-lymphocyte ratio in the emergency and prophylactic cerclage patients ($p=0.196$). In a subgroup analysis, neutrophil-lymphocyte ratio in emergency cerclage patients was higher, but not significantly so, in the group delivering after 34 gestational weeks. Similarly, in the history-indicated prophylactic cerclage patients, neutrophil-lymphopcyte ratio was non-significantly higher in patients who delivered at and after 34 gestational weeks. There appears to be a tend towards higher neutrophil-lymphocyte ratio in deliveries after 34 gestational weeks, but no significance was found and in addition, group sizes were small, so no reliable conclusion can be drawn.

Study Limitation

The main limitation of this study is its retrospective nature. Furthermore, results are robust, since strict criteria were

applied in order to identify patients who were candidates for prophylactic and emergency cerclage procedures. Also, number of included patients is not that insufficient when we consider the incidence of cervical incompetence but the small group, and in particular the small sub-group analyses make statistical comparisons less reliable.

Conclusion

Main indications for prophylactic cervical cerclage procedures are well defined in contemporary and evidence-based guidelines. The results of the present study indicated that emergency cerclage procedures may improve gestational week at delivery. Similarly; to prophylactic cerclage procedures, emergency cerclage procedures may be effective in preventing preterm and severely preterm deliveries. This result should not be interpreted as emergency cerclage being as effective as prophylactic cerclage. Emergency cerclage cannot substitute for a prophylactic procedure. Liberal use of cerclage may have adverse outcomes and risks, but when used with appropriate indication, the procedure improve obstetrical and perinatal outcomes. Pre-cerclage cervical length correlated with gestational week at delivery in both prophylactic and emergency cerclage groups. Cervical dilatation at admission may be a poor prognostic factor for preterm delivery in emergency cerclage patients. In all cerclage patients higher BMI values have a negative effect on gestational week at delivery. In addition, the prognostic value of neutrophil-lymphocyte ratio remains unclear. Larger randomized studies may illuminate the relationship between these factors and cerclage procedure outcomes.

Ethics Committee Approval: *This study was approved by the Başkent University Faculty of Medicine Institutional Ethical Committee on 05/14/2019, with the approval number of KA19/168.*

Informed Consent: *Informed consent of patients was obtained before cervical cerclage procedures.*

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References

1. Brown R, Gagnon R, Delisle MF, Maternal Fetal Medicine Committee. Cervical insufficiency and cervical cerclage. J Obstet Gynaecol Can 2013; 35: 1115-27.
2. ACOG practice bulletin no.142: Cerclage for the management of cervical insufficiency. Obstet Gynecol 2014; 123(2 Pt 1): 372-79.
3. Grobman W. 532: Short cervix and activity restriction. Am J Obstet Gynecol 2013; 208: S227-8.
4. Goya M, Pratcorona L, Merced C, Rodó C, Valle L, Romero A, et al. Cervical pessary in pregnant women with a short cervix (PECEP): an open-label randomised controlled trial. Lancet 2012; 379: 1800-6.
5. Nicolaides KH, Syngelaki A, Poon LC, Matallana CP, Plasencia W, Molina FS, et al. Cervical pessary placement for prevention of preterm birth in unselected twin pregnancies: a randomized controlled trial. Am J Obstet Gynecol 2016; 214: 3.e1-9.
6. Hoffman MC, McCarthy L, Heinrichs G, Hyer J, Carey JC. 818: Activity restriction does not slow the rate of cervical shortening in pregnancy. Am J Obstet Gynecol 2014; 210.
7. Odibo AO. Berghella V, To MS, Rust OA, Althuisius SM , Nicolaides KH. Shirodkar versus McDonald cerclage for the prevention of preterm birth in women with short cervical length. Am J Perinatol 2007; 24: 55-60.
8. Brown JA, Pearson AW, Veillon EW, Rust OA, Chauhan SP, Magann EF, et al. History- or ultrasound-based cerclage placement and adverse perinatal outcomes. J Reprod Med 2011; 56: 385-92.
9. Simcox R, Seed PT, Bennett P, Teoh TG, Poston L, Shennan AH. A randomized controlled trial of cervical scanning vs history to determine cerclage in women at high risk of preterm birth (CIRCLE trial). Am J Obstet Gynecol 2009; 200: 623.e1-6.
10. Gluck O, Mizrachi Y, Ginath S, Bar J, Sagiv R. Obstetrical outcomes of emergency compared with elective cervical cerclage. J Matern Fetal Neonatal Med 2017; 30: 1650-4.
11. Liddiard A, Bhattacharya S, Crichton L. Elective and emergency cervical cerclage and immediate pregnancy outcomes: a retrospective observational study. JRSM Short Rep 2011; 2: 91.
12. Chen Q, Chen G, Li N. Clinical effect of emergency cervical cerclage and elective cervical cerclage on pregnancy outcome in the cervical-incompetent pregnant women. Arch Gynecol Obstet 2018; 297: 401-7.
13. Berghella V, Mackeen AD. Cervical length screening with ultrasound-indicated cerclage compared with history-indicated cerclage for prevention of preterm birth: a meta-analysis. Obstet Gynecol 2011; 118: 148-55.
14. Suhag A, Seligman N, Giraldo-Isaza M, Berghella V. Pre-pregnancy body mass index (BMI) and cerclage success. J Matern Fetal Neonatal Med 2016; 29: 368-75.
15. Yalvac S, Esin S, Kocak O, Yirci B, Kandemir O. Effect of body mass index on latency periods after history-indicated cervical cerclage. Aust N Z J Obstet Gynaecol 2014; 54: 121-5.
16. Figueroa R, Crowell R, Martinez A, Morgan M, Wakefield D. McDonald versus Shirodkar cervical cerclage for the prevention of preterm birth: impact of body mass index. J Matern Fetal Neonatal Med 2019; 32: 3408-14.
17. Poggi SH, Vyas NA, Pezzullo JC, Landy HJ, Ghidini A. Does increasing body mass index affect cerclage efficacy? J Perinatol 2012; 32: 777-9.
18. Zhao Z, Zhao X, Lu J, Xue J, Liu P, Mao H. Prognostic roles of neutrophil to lymphocyte ratio and platelet to lymphocyte ratio

in ovarian cancer: a meta-analysis of retrospective studies. Arch Gynecol Obstet 2018; 297: 849-57.

19. Chen L, Wang X, Shu J, Xu S, Wu Q, Yu Y. Diagnostic value of serum D-dimer, CA125, and neutrophil-to-lymphocyte ratio in differentiating ovarian cancer and endometriosis. Int J Gynaecol Obstet 2019; 147: 212-8.
20. Yilmaz M, Cimilli G, Saritemur M, Demircan F, Isaoglu U, Kisaoglu A, Emet M. Diagnostic Accuracy of Neutrophil/Lymphocyte Ratio, Red Cell Distribution Width and Platelet Distribution Width in Ovarian Torsion. J Obstet Gynaecol 2016; 36: 218-22.
21. Gogoi P, Sinha P, Gupta B, Firmal P, Rajaram S. Neutrophil-to-lymphocyte ratio and platelet indices in pre-eclampsia. Int J Gynaecol Obstet 2019; 144: 16-20.
22. Song JE, Lee KY, Son GH. Prognostic significance of neutrophil-to-lymphocyte ratio for repeat cerclage in women with prolapsed membranes. Biomed Res Int 2018; 2018: 1507398.

Analyses of soluble endoglin and matrix metalloproteinase 14 using enzyme-linked immunosorbent assay in the diagnosis and assessment of severity of early- and late-onset pre-eclampsia

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Abstract

Objective: Abnormal trophoblastic invasion and impaired placentation have a crucial role in the etiopathogenesis of preeclampsia (PrE). Trophoblastic cells are involved in invading the maternal decidua and remodelling of the spiral arteries with matrix metalloproteinase-14 (MMP-14). MMP-14 cleavage of endoglin releases its extracellular region, the soluble form of endoglin (s-ENG), into the maternal circulation. In PrE, there is a relationship between endothelial dysfunction and s-ENG concentration. The aim was to determine and compare the serum levels of s-ENG and MMP-14 in different groups of PrE patients and healthy subjects.

Material and Methods: The study included 30 patients with late-onset preeclampsia (L-PrE) (group 1; gestational age ≥34 weeks), 33 patients with normal pregnancy (group 2; gestational age ≥34 weeks), 31 patients early-onset preeclampsia (E-PrE) (group 3; gestational age <34 weeks), and 31 patients with normal pregnancy (group 4; gestational age <34 weeks). s-ENG and MMP-14 concentrations measured using enzyme-linked immunosorbent assays were compared.

Results: In all groups, MMP-14 concentrations decreased with increasing gestational age. s-ENG concentrations were highest in the E-PrE group. In groups 1 and 3, 29 had mild PrE while 32 suffered severe PrE and s-ENG concentrations did not differ between mild and severe preeclampsia ($p=0.133$). However, there was a significant difference in MMP-14 concentration comparing mild with severe PrE (3.11 ± 0.61 vs 3.54 ± 1.00; $p=0.047$, respectively). There was no correlation between s-ENG and MMP-14 concentrations.

Conclusion: MMP-14 and s-ENG concentrations can be predictive biomarkers for the diagnosis of PrE. Maternal serum MMP-14 concentration may be a biomarker for determining the severity of PrE. (J Turk Ger Gynecol Assoc 2021; 22: 29-36)

Keywords: Endothelial dysfunction, hypertension, implantation, severe pre-eclampsia, trophoblast

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Introduction

Preeclampsia (PrE) remains an important obstetric syndrome and has a major effect on maternal/infant morbidity/mortality worldwide (1). The main features include protein leakage into urine and new-onset gestational hypertensive disease, or other signs/symptoms of PrE in the absence of proteinuria after 20 weeks' gestation. Although improvements have been reported in the diagnosis and treatment of PrE in previous studies, there are still many unanswered questions. In PrE, the reduced capacity of extravillous trophoblasts (EVTs) for invasion into the spiral arteries results in an inadequately perfused fetal-placental unit. When the embryo is implanted, EVTs invade the decidua and remodel spiral arteries getting as deeply as the inner third of the myometrium. Matrix metalloproteinases (MMPs) enable the infiltration of EVTs into the uterine wall. Decidual stromal cells produce high concentrations of MMPs, enhancing the invasiveness of the EVTs (2,3). Dysregulation of MMPs causes inadequate trophoblast invasion, inadequate uterine and spiral artery remodeling, which may result in the development PrE (4,5).

MMPs are calcium- and zinc-dependent proteases that break down different components of the extracellular matrix (ECM). MMPs are key to the mediation of apoptosis, cell proliferation, cell-cell adhesion, cell migration and invasion, and tissue remodeling (6). The presence of MMP-14 has been demonstrated in the membrane of trophoblast and vascular endothelial cells (7). In normal pregnancy, there is a notable increase in MMP-14 concentration in the last trimester versus the first trimester (8). In PrE, the abnormal release of vasoactive factors such as MMP-1 and MMP-14 occurs near the end of pregnancy, thereby contributing to the development of hypertension. MMP-1 and MMP-14 have also been investigated in other obstetric syndromes, such as premature rupture of membranes (PROM) and preterm labor. The role of MMPs has been investigated in a wide range of conditions including inflammation and malignant growth, as well as reproductive and neurologic disorders (2). Further, analysis of gene expression demonstrated that both MMP-14 and endoglin gene expression was increased in PrE (9).

Endoglin (CD105) is an integral, membrane-bound glycoprotein and one of its functions is as a co-receptor for transforming growth factor-beta. High endoglin concentrations on the syncytiotrophoblasts in patients with severe PrE have been reported using western blot analysis and immunohistochemistry staining. A soluble form of endoglin (s-ENG) has been demonstrated in human blood. s-ENG is released from endothelial tissues, phagocytes, syncytiotrophoblasts, and smooth muscle cells. The role and direct molecular mechanism of s-ENG is not clear; however, it is capable of reducing angiogenesis (10,11). MMP-14 cleavage of endoglin releases s-ENG into the maternal circulation. The rise in s-ENG concentration in PrE is proportional to the severity of the disease and reduces after delivery. The relationship between endothelial dysfunction following poor placentation and s-ENG has also been shown in PrE. Several studies have shown that MMP-14 has importance in the reduction of s-ENG concentrations leading to alleviation of the clinical manifestations of PrE (2,8).

There is currently no mechanism to predict PrE or confirm PrE diagnosis before clinical occurrence. Many studies have investigated MMP-14 in PrE. However, there are no studies measuring serum MMP-14 concentrations by enzyme-linked immunosorbent assay (ELISA) in PrE. ELISA is a less-invasive, easy, fast, and inexpensive method. The aim was to evaluate the values of maternal serum MMP-14 and s-ENG in patients with PrE compared to healthy pregnancies, and to investigate the possible role of these biomarkers in assessment of PrE severity.

Material and Methods

Patient selection

Our subjects were prospectively recruited from the Clinic of Obstetrics and Gynecology at Cengiz Gökçek Public Hospital, Gaziantep, Turkey, between January 2018 and December 2018. This study was conducted according to the Declaration of Helsinki, and the Institutional Ethical Review Board of Gaziantep University Faculty of Medicine approved the study (approval number: 2018/91). A total of 138 pregnant women were recruited to the study, out of which 13 were excluded on grounds of incomplete fetomaternal details, refusal to participate in the study, and an SGA fetus in the control group (Figure 1). The remaining 125 women were divided into groups as follows. Thirty women with late-onset (≥34 weeks of gestation) preeclampsia (L-PrE) formed group 1; 33 healthy women at ≥34 weeks gestation were recruited to group 2; 31 women with early-onset (<34 weeks gestation) preeclampsia (E-PrE) formed group 3; and 31 healthy women at <34 weeks pregnancy were recruited to group 4. Groups 2 and 4 were matched for maternal and gestational age with groups 1 and 3, respectively. The control groups comprised women with healthy pregnancies who presented to our hospital for routine obstetric examination. All subjects were informed about the study and each gave written consent. Gestational age assessment was based on last menstrual period or first-trimester ultrasonographic obstetric measurements.

The diagnosis of PrE was based on the presence of proteinuria (urinary excretion of protein ≥300 mg in a 24-hour urine specimen, or proteinuria ≥1+ in dipstick) and a maternal blood pressure of ≥140/90 mmHg (a mean of two blood

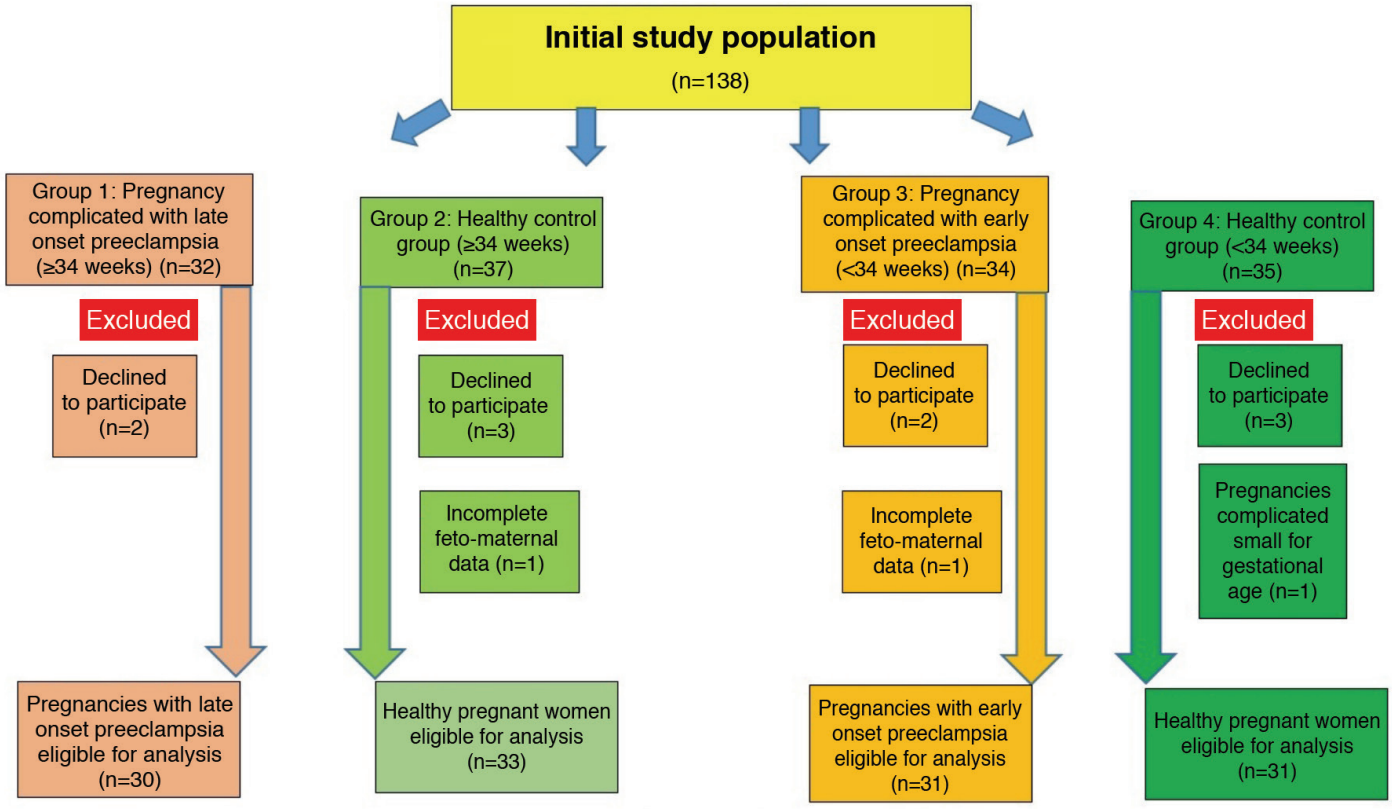

Figure 1. Flow chart of the pregnant women recruited in the study

pressure measurements obtained six hours apart), occurring after 20 weeks of gestation in a previously normotensive woman, as defined by the Committee on Terminology of the American College of Obstetricians and Gynecologists. Mild PrE was defined as a blood pressure between ≥140/90 mmHg and 160/110 mmHg (for systolic and/or diastolic measurement) and if values exceeded 160/110 mmHg it was accepted as severe, as previously described (12). Small for gestational age (SGA) newborns were accepted as birth weight <10th percentile for gestational age with Turkey's national nomogram as the reference for fetal growth (13). Maternal body mass index (BMI) was calculated in kg/m^2 using the standard formula. Exclusion criteria for both groups were: pregnant women with any systemic disease, such as chronic hypertension, diabetes mellitus, thyroid diseases, liver and kidney diseases; use of any kind of medication throughout pregnancy; use of any medication for PrE treatment at the time of first admission; history of pregnancies complicated by PROM or chorioamnionitis; fever at the time of first admission; fetal congenital abnormalities or genetic syndromes; smoking during pregnancy; multiple gestation; and active labor.

Each pregnant woman had obstetric ultrasound examination and fetal/maternal assessment, which were conducted by a single obstetrician-gynecologist specialist (AO). Obstetric anamneses were obtained from all pregnant women. Demographic data, such as age, gravidity, parity, BMI, and gestational age were recorded. Maternal venous blood samples were taken for measurement of s-ENG and MMP-14 concentrations after the diagnosis of PrE in the outpatient clinic. The samples were immediately centrifuged at 1500 g for 10 min, and serum samples were separated and stored at -80 °C until required for analysis. All patients with E-PrE were hospitalized. After hospitalization, a betamethasone injection (12 mg) was administered without delay. Pregnancy was terminated immediately in the event of urgent fetal/maternal situations such as severe PrE development or fetal distress. Otherwise, maternal blood pressure was measured at least every 4 hours during rest with the arm held at the level of the heart. Hypertension can persist for short intervals in patients with diastolic and/or systolic blood pressure ≥110 mmHg and ≥160 mmHg, respectively, to facilitate timely anti-hypertensive treatment. In cases of E-PrE, delivery should be delayed for at least 48 hours if maternal and fetal status permit. During this period, betamethasone injections for lung maturation (two doses of 12 mg at 24-hour intervals) were administered. All patients with L-PrE were also hospitalized and their pregnancies were terminated. Women with uncomplicated pregnancies were randomly selected at the same time as the

case selection was performed to serve as controls. The samples from the control groups were obtained during routine obstetric examinations in the last trimester of pregnancy. These women were then followed up until delivery. The four groups were compared in terms of maternal age, BMI, gravida, parity, week of gestation, systolic/diastolic blood pressure, full blood count, liver function tests including alanine aminotransferase and aspartate aminotransferase, blood urea nitrogen, creatinine, s-ENG, MMP-14 and total protein in spot urine sample, and infant weight at delivery.

Serum MMP-14 and s-ENG analysis

MMP-14 concentrations were assessed using a commercial ELISA kit, the Human MMP-14 ELISA Kit (Rel Assay Diagnostics, Gaziantep, Turkey), in accordance with the manufacturer's instructions. This ELISA kit (sensitivity range: 0.05 ng/mL; detection range: 0.1-30 ng/mL) is based on the principle of biotin double-antibody sandwich technique. The intra- and inter-assay variation coefficients were <8% and <10%, respectively. A commercial ELISA kit was also used for the assessment of s-ENG concentrations, specific for the detection of human s-ENG with high sensitivity and specificity (Rel Assay Diagnostics, Gaziantep, Turkey). The kit (sensitivity range: 0.23 ng/mL; detection range: 0.5-200 ng/mL) also used a sandwich-ELISA principle. The intra- and inter-assay variation coefficients were <8% and <10%, respectively.

Statistical analysis

Statistical analyses were performed using SPSS for Windows, version 25.0, software package (IBM Inc., Armonk, NY, USA), and p values <0.05 were accepted as statistically significant. Results were presented as mean ± standard deviation (SD) after investigating normality distribution by Shapiro-Wilk test. In the analyses of the variables, the Student's t-test was used for comparing two groups. ANOVA was used for the comparison of four groups. As a result of variance analysis, Tukey HSD test, one of the post-hoc tests, was used to determine the difference between the groups. In addition, the demographic data of the variables were investigated using frequency analyses.

Results

A total of 61 pregnant women with PrE were included. The control groups included 64 healthy pregnant women. The demographic data of the groups were compared (Table 1). There was no significant difference between maternal age, gravidity, and parity, but there was a significant difference between the groups for BMI, gestational age, systolic blood pressure, diastolic blood pressure, and birth weights ($p<0.05$). The laboratory results of the study and control groups are shown in Table 2. MMP-14 and s-ENG concentrations differed between the groups, as shown in Table 2 and Figure 2, 3. MMP-14 concentrations did not differ between group 1 and group 2, nor did they differ between group 3 and group 4. Thus, MMP-14 concentrations were the same in women with PrE and healthy pregnancies matched for gestational age but did differ between earlier and later pregnancies, that is between <34 weeks and ≥34 weeks gestation. In all pregnant women, as the gestational age increased, MMP-14 concentrations in maternal serum decreased.

The highest concentrations of s-ENG were found in the E-PrE group. No differences were detected between s-ENG and MMP-14 concentrations in mild (n=21) and severe PrE (n=9) in the L-PrE group ($p=0.829$, $p=0.210$, respectively). In addition, no differences were detected between s-ENG and MMP-14 concentrations in mild (n=8) and severe PrE (n=23) in the E-PrE group ($p=0.887$, $p=0.739$, respectively). When early and late PrE groups (group 1 and group 3) were compared, no differences were detected between s-ENG concentrations in mild (n=29) and severe PrE (n=32) ($p=0.133$). However, there was a significant difference in MMP-14 concentrations in pregnancies affected by mild or severe PrE (3.11±0.61 vs 3.54±1.00, $p=0.047$, respectively) (Table 3).

When the patients with (n=10) and without (n=115) SGA infants were compared, no differences were found in s-ENG and MMP-14 concentrations ($p=0.133$, $p=0.969$, respectively). When the patients with (n=15) and without (n=110) new-onset cerebral or visual disturbances were compared, there were again no differences in s-ENG and MMP-14 concentrations ($p=0.528$, $p=0.573$, respectively). When the patients with (n=8) and without (n=117) right upper quadrant or epigastric pain were compared, no difference was found in s-ENG and MMP-14 concentrations ($p=0.162$, $p=0.154$, respectively). When those who had first gravidity (31 patients) and gravidity >1 (94 patients) were compared, no differences were found between the s-ENG and MMP-14 concentrations ($p=0.855$, $p=0.364$, respectively). No difference was found in serum concentrations of s-ENG and MMP-14 when subjects were compared in terms of BMI [<30 kg/m^2 (n=56) and ≥30 kg/m^2 (n=69)] $p=0.373$ and $p=0.873$ for s-ENG and MMP-14, respectively or for maternal age [<35 years (n=100) and ≥35 years (n=25)] $p=0.167$ and $p=0.625$ for s-ENG and MMP-14 respectively. No statistically significant correlation was detected between s-ENG and MMP-14 concentrations ($p>0.05$).

Discussion

In the present study, maternal blood concentrations of s-ENG and MMP-14 were evaluated in order to examine the association between diagnoses of L-PrE/E-PrE, the severity of PrE, and these biomarkers. It was found that serum s-ENG and MMP-14 concentrations differed significantly between the

Table 1. The demographic data of the groups

Variables	L-PrE group 1 (n=30)	Control group 2 (n=33)	E-PrE group 3 (n=31)	Control group 4 (n=31)	p	Post-hoc comparisons	p
Age (years)	28.9±6.4	25.8±6.0	29.1±6.6	28.9±6.6	0.122	-	-
BMI (kg/m^2)	32.2±5.4^a	28.6±4.9^b	31.3±5.0^{ab}	29.5±4.7^{ab}	0.020	Group 1-group 2	0.026
						Group 1-group 3	0.473
						Group 1-group 4	0.137
						Group 2-group 3	0.147
						Group 2-group 4	0.915
						Group 3-group 4	0.463
Gestational age (weeks)	37.2±1.5^a	37.7±1.5^a	31.1±2.2^b	30.5±2.0^b	**0.001**	Group 1-group 2	0.753
						Group 1-group 3	0.001
						Group 1-group 4	0.001
						Group 2-group 3	0.001
						Group 2-group 4	0.001
						Group 3-group 4	0.555
Gravidity (n)	3.3±2.4	3.5±5.4	3.1±1.7	3.7±2.3	0.887		
Parity (n)	1.9±1.9	1.5±1.3	1.5±1.4	1.9±1.7	0.620		
Systolic pressure (mmHg)	160±18^b	105±10^c	173±18^a	102±10^c	**0.001**	Group 1-group 2	0.001
						Group 1-group 3	0.017
						Group 1-group 4	0.001
						Group 2-group 3	0.001
						Group 2-group 4	0.974
						Group 3-group 4	0.001
Diastolic pressure (mmHg)	103±10^b	65±6^c	110±11^a	65±7^c	**0.001**	Group 1-group 2	0.001
						Group 1-group 3	0.047
						Group 1-group 4	0.001
						Group 2-group3	0.001
						Group 2-group 4	0.979
						Group 3-group 4	0.001
Birth weight (grams)	2960±691^a	3249±483^a	1650±467^b	3172±346^a	**0.001**	Group 1-group 2	0.177
						Group 1-group 3	0.001
						Group 1-group 4	0.371
						Group 2-group 3	0.001
						Group 2-group 4	0.930
						Group 3-group 4	0.001

L-PrE group 1: Late-onset preeclampsia patient group, Control group 2: Late-onset preeclampsia control group, E-PrE group 3: Early-onset preeclampsia patient group, Control group 4: Early-onset preeclampsia control group, Age: Maternal age, BMI: Body mass index, Gestational age: Gestational age at the time of recruitment, Syst TA: Systolic blood pressure, Diast TA: Diastolic blood pressure, n: Number, SD: Standard deviation, $p<0.05$ indicates statistical significance, ^{a,b,c}: Different letters symbolize the difference between the groups

two PrE groups and their respective matched control groups. The concentrations of s-ENG were at the highest in the E-PrE group (group 3) in which severe PrE was present. MMP-14 concentrations, on the other hand, were found to be highest in severe PrE cases.

The contact between the placenta and the decidua ensures metabolic exchange between the fetus and the mother. Trophoblast cells of the embryo initially attach to the uterine epithelium. Trophoblasts differentiate into EVTs, which degrade the uterine epithelium basement membrane and ECM, then migrate into the decidual stroma. EVTs are characterized by their invasiveness, ensuring sufficient contact with the maternal circulation. Unsurprisingly, this invasion stage is under strict biological control and restricted to the decidua and the proximal third of the myometrium in healthy pregnancies. When this invasive process is dysregulated fetal

Table 2. The laboratory parameters of the four groups

Variables	Group 1 (n=30)	Group 2 (n=33)	Group 3 (n=31)	Group 4 (n=31)	p	Post-hoc comparisons	p
Hemoglobin (g/dL)	12.0±1.4[a]	10.9±1.3[b]	11.9±1.3[a]	11.4±1.2[ab]	0.004	-	-
Hematocrit (%)	36±3[a]	33±3[c]	35±3[ab]	33±2[bc]	**0.001**	-	-
Platelets (x10^3/μL)	240±84	235±82	200±83	236±51	0.152	-	-
WBC (μL/mL)	10.9±2.46	10.5±2.6	10.4±3.0	9.9±2.8	0.587	-	-
BUN (mg/dL)	8.9±3.0[ab]	7.3±2.5[bc]	10.4±3.9[a]	6.4±2.1[c]	**0.001**	-	-
Creatinine (mg/dL)	0.56±0.11[b]	0.47±0.09[c]	0.66±0.17[a]	0.46±0.08[c]	**0.001**	-	-
ALT (IU/L)	20±30	10±4	22±30	11±4	0.054	-	-
AST (U/L)	22±17	16±3	31±35	14±3	**0.005**	-	-
Proteinuria (positivity on dipstick)	2.67±1.1[a]	0±0[b]	3.0±1.2[a]	0.0±0.2[b]	**0.001**	-	-
s-ENG (ng/mL)	17.24±1.73[a]	18.49±2.01[ab]	22.64±12.98[b]	18.21±4.48[ab]	**0.015**	Group 1-group 2	0.892
						Group 1-group 3	**0.016**
						Group 1-group 4	0.086
						Group 2-group 3	0.999
						Group 2-group 4	0.064
						Group 3-group 4	0.463
MMP-14 (ng/mL)	2.83±0.31[a]	2.93±0.43[a]	3.82±0.94[b]	3.81±1.26[b]	**0.001**	Group 1-group 2	0.957
						Group 1-group 3	**0.001**
						Group 1-group 4	**0.001**
						Group 2-group 3	**0.001**
						Group 2-group 4	**0.001**
						Group 3-group 4	1.00

Group 1: Late-onset preeclampsia ≥34 weeks of gestation, Group 2: Healthy pregnancy ≥34 weeks of gestation, Group 3: Early-onset preeclampsia <34 weeks gestation, Group 4: Healthy pregnancy <34 weeks of gestation, WBC: White blood cells, BUN: Blood urea nitrogen, ALT: Alanine aminotransferase, AST: Aspartate aminotransferase, Proteinuria: Spot urine proteinuria by dipstick test, s-ENG: Soluble endoglin, MMP-14: Matrix metalloproteinase-14, n: number, SD: Standard deviation, p<0.05 indicates statistical significance, [a,b,c]: Different letters symbolize the difference between the groups

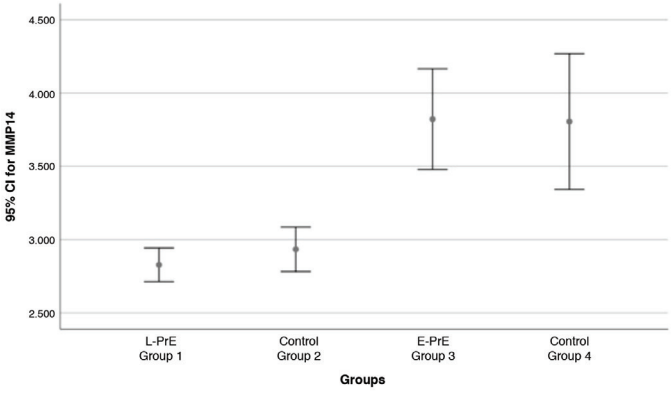

Figure 2. Error plot showing the mean value of matrix metalloproteinase-14 in the groups

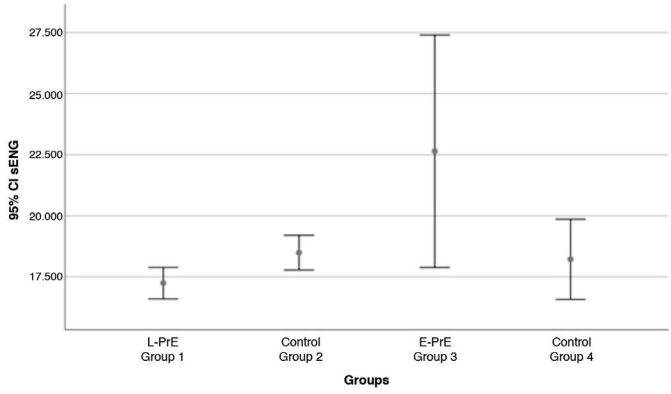

Figure 3. Error plot showing the mean value of soluble endoglin in the groups

growth restriction and PrE may result. Invasive processes are generally assisted by the expression and activity of MMPs (8). EVTs also reach and remodel the spiral arteries, transforming them into low resistance vessels, a process necessary to allow an adequate blood supply to the fetus. It has been shown that this process is regulated by MMP-14 and MMP-15, mediated by endothelin-1 in a low oxygen environment (2-3% O_2) (14). These pathologic changes constitute a process that starts in the first trimester, proceeds through a normal pregnancy period, and are finally reflected in an excessive clinical manifestation in the last trimester. These events consist of pathogenic factors that are produced in the placenta as a response to hypoxia, mixing

Table 3. Clinical characteristics and biomarkers levels in the preeclampsia groups, group 1 (late PrE) and group 3 (early PrE). Biomarker data is given as mean ± SD

Variables		Biomarkers			
Group 1 and 3	**Subgroups**	**s-ENG (ng/mL)**	**p**	**MMP-14 (ng/mL)**	**p**
Nausea, vomiting and epigastric symptoms	Absent (n=117)	18.37±3.96	0.162	3.30±0.92	0.154
	Present (n=8)	30.65±22.19		3.93±1.09	
Persistent cerebral symptoms	Absent (n=110)	19.00±7.19	0.528	3.33±0.91	0.573
	Present (n=15)	20.26±7.30		3.47±1.21	
Preeclampsia	Mild (n=29)	18.61±5.88	0.291	3.11±0.61	0.047
	Severe (n=32)	21.24±12.06		3.54±1.00	
Small for gestational age	No (n=115)	18.87±6.56	0.133	3.35±0.94	0.969
	Yes (n=10)	22.43±12.38		3.33±1.04	

s-ENG: Soluble endoglin, MMP-14: Matrix metalloproteinase-14, n: number, SD: standard deviation, $p<0.05$ indicates statistical significance

with the maternal blood and causing endothelial dysfunction. Despite the presence of abnormally high concentrations of excreted molecules, inflammatory factors, autoimmunity or anti-angiogenic processes are far more important (15).

Several authors have demonstrated that the ECM and MMP-14 were vital regulators of angiogenesis (16,17). Placental MMPs may influence spiral artery remodeling in implantation. In PrE, MMPs have roles in prompting vasoconstriction, changes in vascular reactivity, and pathological damage to the endothelium. Thus, there has been an increase in interest in the pathological effect of the imbalance of angiogenic and/or anti-angiogenic factors in PrE, including endoglin/s-ENG and MMP-14. The close association between the location and expression levels of MMP-14 and the same features relating to endoglin has attracted much interest. MMPs have become important biomarkers for the identification of women with an elevated risk of developing PrE and also important biologic targets for treating this syndrome (1,7,18). The process of investigating MMP-14 tissue level and expression postpartum is complex, laborious and expensive. The aim of this study was to evaluate MMP-14 through direct measurements in maternal serum, a minimally invasive process, which can be repeated throughout pregnancy. The drawback of serum measurements of MMP-14 are that these will not reflect tissue specific expression patterns in normal or PrE pregnancy, because of the significant membrane-binding properties of MMP-14 (19).

Zhang et al. (7) found that s-ENG concentrations were high in patients with severe PrE, and showed that this event was triggered by MMP-14, leading to the development of severe PrE. In contrast, Levine et al. (9) suggested that there were no important differences in s-ENG concentrations between women with mild PrE and severe PrE, reporting no correlation between s-ENG concentrations and the severity of PrE. However, it was also shown that there was an evident increase in s-ENG concentration, starting 2-3 months before the onset of PrE. They also reported that this increase was greater in E-PrE than in L-PrE (9,20). Sezer et al. (4) conducted a study in a PrE group comprising patients with E-PrE and L-PrE, and found that s-ENG concentrations in PrE were high in the maternal circulation and also umbilical cord blood. However, there was no difference between both preeclamptic groups regarding s-ENG concentration of the maternal circulation (4). Zafer et al. (11) compared s-ENG concentrations in maternal serum and amniotic fluid and reported that they were incompatible. All these findings suggest that all compartments have independent dynamics in terms of s-ENG. Moreover, it is possible that MMP-14 may have independent dynamics, because of the close biological association with s-ENG. When all patients with severe PrE were analyzed, MMP-14 concentrations were found to be higher although s-ENG was not different. Interestingly, this might suggest that MMP-14 serum concentrations in PrE may be more significant than s-ENG. Although s-ENG and MMP-14 concentrations showed a similar pattern, the relationship between the two showed no statistically significant correlation. This may be because of the membrane-binding properties of MMP-14.

Study Limitation

We acknowledge strengths and limitations of the study. The most important limitation of our study was the small number of participants. We do not have the pre-pregnancy BMI of all participants. s-ENG and MMP-14 concentrations could have been examined in different compartments, such as the placenta, umbilical cord serum, and amniotic fluid. The other limitation was that there are no uterine artery Doppler studies in this study. The strengths of the study were that none of the patients had any treatment for PrE and only participants who were not in active labor were selected for the study. Another strength was the comparison of MMP-14 serum concentrations with s-ENG, which has a major role in angiogenesis and endothelial cell function.

Conclusion

Larger basic and clinical studies are necessary to assess whether MMPs have an important role in the pathophysiology of PrE. In addition, meta-analyses should be performed to investigate the predictive value of these enzymes as biomarkers or therapeutic targets. Finally, in view of the literature data, we believe that testing s-ENG is worthy of further investigation for the early diagnosis of PrE. In addition, it appears reasonable to investigate serum MMP-14 concentrations in predicting patients who will proceed to a severe form of PrE.

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Ethics Committee Approval: *This study was conducted according to the Declaration of Helsinki, and the Institutional Ethical Review Board of Gaziantep University Faculty of Medicine approved the study (approval number: 91, date: 06.06.2018).*

Informed Consent: *All subjects were informed about the study and each gave written consent.*

Peer-review: *Externally peer-reviewed.*

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References

1. Schmella MJ, Roberts JM, Conley YP, Ren D, Storvold GL, Ingles SA, et al. Endoglin pathway genetic variation in preeclampsia: a validation study in Norwegian and Latina cohorts. Pregnancy Hypertens 2018; 12: 144-9.
2. Espino Y Sosa S, Flores-Pliego A, Espejel-Nunez A, Medina-Bastidas D, Vadillo-Ortega F, Zaga-Clavellina V, et al. New insights into the role of matrix metalloproteinases in preeclampsia. Int J Mol Sci 2017; 18: 1448.
3. Pollheimer J, Fock V, Knofler M. Review: the ADAM metalloproteinases - novel regulators of trophoblast invasion? Placenta 2014; 35 Suppl: S57-63.
4. Sezer SD, Kucuk M, Yenisey C, Yuksel H, Odabasi AR, Turkmen MK, et al. Comparison of angiogenic and anti-angiogenic factors in maternal and umbilical cord blood in early- and late-onset pre-eclampsia. Gynecol Endocrinol 2012; 28: 628-32.
5. Kasurinen A, Tervahartiala T, Laitinen A, Kokkola A, Sorsa T, Bockelman C, et al. High serum MMP-14 predicts worse survival in gastric cancer. PloS One 2018; 13: e0208800.
6. Almalki SG, Llamas Valle Y, Agrawal DK. MMP-2 and MMP-14 silencing inhibits VEGFR2 cleavage and induces the differentiation of porcine adipose-derived mesenchymal stem cells to endothelial cells. Stem Cells Transl Med 2017; 6: 1385-98.
7. Zhang XH, Zhang HY, Lu S, Jiang LL, Wu J, Yang YL, et al. MMP-14 aggravates onset of severe preeclampsia by mediating soluble endoglin release. Eur Rev Med Pharmacol Sci 2018; 22: 1209-15.
8. Anacker J, Segerer SE, Hagemann C, Feix S, Kapp M, Bausch R, et al. Human decidua and invasive trophoblasts are rich sources of nearly all human matrix metalloproteinases. Mol Hum Reprod 2011; 17: 637-52.
9. Levine RJ, Lam C, Qian C, Yu KF, Maynard SE, Sachs BP, et al. Soluble endoglin and other circulating antiangiogenic factors in preeclampsia. N Engl J Med 2006; 355: 992-1005.
10. Hawinkels LJ, Kuiper P, Wiercinska E, Verspaget HW, Liu Z, Pardali E, et al. Matrix metalloproteinase-14 (MT1-MMP)-mediated endoglin shedding inhibits tumor angiogenesis. Cancer Res 2010; 70: 4141-50.
11. Zafer E, Demircan Sezer S, Nergiz Avcioglu S, Atakul T, Kurt Omurlu I, Yuksel H. Correlation between maternal serum-amniotic fluid anti-angiogenic factors and uterine artery Doppler indices. J Matern Fetal Neonatal Med 2017; 30: 2653-7.
12. ACOG practice bulletin no.202: gestational hypertension and preeclampsia. Obstet Gynecol 2019; 133: 1.
13. Topcu HO, Guzel AI, Ozgu E, Yildiz Y, Erkaya S, Uygur D. Birth weight for gestational age: a reference study in a tertiary referral hospital in the middle region of Turkey. JCMA 2014; 77: 578-82.
14. Majali-Martinez A, Velicky P, Pollheimer J, Knofler M, Yung HW, Burton GJ, et al. Endothelin-1 down-regulates matrix metalloproteinase 14 and 15 expression in human first trimester trophoblasts via endothelin receptor type B. Hum Reprod 2017; 32: 46-54.
15. Hypertension in pregnancy. Report of the American College of Obstetricians and Gynecologists' Task Force on hypertension in pregnancy. Obstet Gynecol 2013; 122: 1122-31.
16. Abu El-Asrar AM, Mohammad G, Allegaert E, Ahmad A, Siddiquei MM, Alam K, et al. Matrix metalloproteinase-14 is a biomarker of angiogenic activity in proliferative diabetic retinopathy. Mol Vis 2018; 24: 394-406.
17. Eddy AC, Bidwell GL 3rd, George EM. Pro-angiogenic therapeutics for preeclampsia. Biol Sex Differ 2018; 9: 36.
18. Kaitu'u-Lino TJ, Tuohey L, Ye L, Palmer K, Skubisz M, Tong S. MT-MMPs in pre-eclamptic placenta: relationship to soluble endoglin production. Placenta 2013; 34: 168-73.
19. Sitras V, Paulssen RH, Gronaas H, Leirvik J, Hanssen TA, Vartun A, et al. Differential placental gene expression in severe preeclampsia. Placenta 2009; 30: 424-33.
20. Rana S, Karumanchi SA, Levine RJ, Venkatesha S, Rauh-Hain JA, Tamez H, et al. Sequential changes in antiangiogenic factors in early pregnancy and risk of developing preeclampsia. Hypertension 2007; 50: 137-42.

Factors affecting parametrial involvement in cervical cancer patients with tumor size ≤4 cm and selection of low-risk patient group

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Abstract

Objective: The primary aim of this study was to evaluate the factors affecting parametrial involvement in cervical cancer patients with tumor size ≤4 cm and selection of the low-risk patient group based on long-term oncologic outcomes.

Material and Methods: Cervical cancer patients operated in the gynecologic oncology division between 2007 and 2013 were retrospectively evaluated. One-hundred and sixty-eight patients with tumor size ≤4 cm were identified. Of these, 159 (86.8%) underwent radical hysterectomy plus pelvic-para-aortic lymphadenectomy and nine (13.2%) underwent fertility-sparing surgery [radical trachelectomy (n=7); large conization (n=2)]. Factors affecting parametrial invasion, including lymphovascular space invasion (LVSI), deep stromal invasion (DSI), lymph node metastases, and tumor size, were evaluated. Statistical analyses were performed using SPSS 23.0 (IBM Corp., Armonk, NY, USA).

Results: Median age was 49.5 years and median tumor size was 2.5 cm (0.45-4 cm). In both univariate and multivariate analyses, the risk of parametrial involvement was increased with LVSI with a hazard ratio (HR) of 3.45 [95% confidence interval (CI): 1.1-10.8] and DSI with a HR of 4.1 (95% CI: 1.18-14.8), while tumor size of ≤2 cm was only significant in univariate analyses. Furthermore, 26 early-stage patients were identified with low-risk factors and they had no parametrial involvement, lymph node metastases, recurrence, or death from disease over 77 months.

Conclusion: Parametrial involvement in low-risk cervical cancer is very rare and less radical procedures may be safe in these patients. (J Turk Ger Gynecol Assoc 2021; 22: 37-41)

Keywords: Low-risk cervical cancer, parametrial invasion, less radical surgery

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Introduction

The incidence of cervical cancer and cancer-related deaths have decreased in the last decades due to widespread screening methods (1). Today, in most centers, radical surgery, such as radical hysterectomy or radical trachelectomy, are performed in cases of stage 1a2 and 1b1 cervical cancer as standard care. These radical surgeries may cause sympathetic and parasympathetic nerve damage in the hypogastric plexus and lead to urinary, sexual, and colorectal dysfunction (2). In 2017 Querleu et al. (3) published an updated version of the classification of radical hysterectomy and defined dorsal, ventral and lateral parametrium and how radical parametrectomy can harm branches of the hypogastric and splanchic nerves.

In stage 1a2 cancers, the risk of recurrence after surgery is 3-5% and 5-year survival is 96-100% (4,5). Some authors suggest that conservative treatment can be applied for these tumors due to the low rate of parametrial and lymphatic involvement (2,4,6-8).

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Studies have shown that as the extent of stromal invasion increases, lymph node and parametrial involvement rates increase (9,10). Likewise, lymphovascular space invasion (LVSI) and increased tumor size have been associated with parametrial and lymph node involvement. According to these findings, low-risk factors are a tumor size of less than 2 cm, deep stromal invasion (DSI) of less than 50%, and no LVSI (11). In a review published by Ramirez et al. (7) parametrial involvement was reported to be 0.4-0.6% in low-risk patients in retrospective studies. Researchers have focused on the applicability of less radical surgeries, such as simple hysterectomy, simple trachelectomy and wide conization, in these patients (12-17).

The primary aim of this study was to identify the factors affecting parametrial involvement in cervical cancer patients with tumor size of ≤4 cm. The secondary aim was to evaluate oncologic outcomes of patients with low-risk factors.

Material and Methods

This retrospective cohort study was carried out in Ankara, Turkey. The records of cervical cancer patients operated on in the department of gynecology and obstetrics and division of gynecologic oncology between 2007 and 2013 were evaluated. Accordingly, 268 patients were identified, and 68 patients with tumor size of >4 cm were excluded. Also excluded were 20 patients who received neoadjuvant chemotherapy before surgery and 12 patients with microinvasive carcinoma. Rare histologies, such as neuroendocrine tumors were also excluded. Figo stages were 1a2-1b1, according to 2009 system, and some patients were upstaged to stage III according to the revised FIGO 2018 staging system (18). Finally, 168 patients with tumor size of ≤4 cm according to pathology report and who had received no treatment before surgery were enrolled in the study. Tumor histologies were squamous cell, adenocarcinoma and adeno-squamous cell cancer.

Of the enrolled patients, 159 (86.8%) underwent radical hysterectomy (Piver type C1 C2) plus pelvic para-aortic lymphadenectomy and nine (13.2%) underwent fertility-sparing surgery [radical abdominal trachelectomy (n=7); large conization (n=2)]. Lymphadenectomy defined as removal of at least 10 lymph nodes from each pelvic region and five from the para-aortic region.

All of the cases were discussed by experienced gyneco-pathologists. Measurement of tumor diameters were made macroscopically. Tumors were measured in millimeters in three dimensions. One of the three dimensions was the measurement on a single slide in which the extent of the invasion was the greatest. The depth of invasion was measured from the basement membrane of the epithelium that the tumor was considered to have arisen from to the deepest point of invasion. Features that helped in the recognition of LVSI were as follows: 1) a tumor nest within a space associated with other vascular structures; 2) the presence of an endothelial lining, adherence of the tumor cell group to the side of the space; 3) the contour of the intravascular component matching the contour of the vessel; and 4) the presence of adherent fibrin. Immunohistochemistry was not performed routinely but if necessary, immunohistochemical demonstration of an endothelial cell lining was undertaken. For this the D2-40 antibody, specific for lymphatic endothelium, or CD31 and CD34 antibodies, which recognize both the lymphatic and blood vascular endothelium, were used.

All patients were discussed in the tumor board and adjuvant treatment policies were determined according to post-operative risk factors as defined in international guidelines (19). Every patients was evaluated every three months in the first two years, every six months for the next three years and annually thereafter for five years. Bimanual vaginal examination were done at each visit, a pap test and thoraco-abdominal computed tomography were performed annually. Recurrences were recorded either by imaging modalities and via fine needle biopsy, if needed, before treatment.

This study was approved by the Başkent University Faculty of Medicine Institutional Review Board (approval number: KA13/252). Due to the retrospective design, informed consent from involved patients was waived.

Statistical analysis

SPSS 23.0 (IBM Corp., Armonk, NY, USA) was used to perform all statistical analyses. Continuous variables were expressed as medians and ranges, and binary variables were reported as counts and percentages. Pearson's chi-square test, Fisher's exact test, and the Mann-Whitney U test were used in univariate analysis. The Cox proportional hazards regression model was used to obtain hazard ratios (HRs) and 95% confidence intervals (CIs). All values of $p<0.05$ were considered statistically significant.

Results

The median (range) age of the patients was 49.5 (29-80) years and median follow-up time was 77.6 (11-142) months. Median tumor size was 2.5 cm (0.45-4 cm). Sixty-two patients had tumor size of ≤2 cm (36.9%) and 57 (33.9%) patients had stromal invasion of ≤50% of the cervical stroma. Thirty-one (18.5%) of the patients had parametrial involvement and 49 (29.8%) had lymph node metastases. Patients' characteristic and percentages of adjuvant treatments are given in Table 1.

In univariate analyses, LVSI, tumor size, DSI, and lymph node metastases were found to have significant effects on parametrial involvement (Table 2). In multivariate analyses,

positive LVSI increased the risk of parametrial involvement with an HR of 3.45 (95% CI: 1.1-10.8), while DSI increased it with an HR of 4.1 (95% CI: 1.18-14.8). Lymph node metastases also increased the risk of parametrial involvement with an HR of 3.2 (95% CI: 1.3-7.5).

Of the sixty-two patients with a tumor size ≤2 cm, 28 (45%) of them had positive LVSI while 18 (29%) had DSI.

In subgroup analyses 26 patients were found to be in the low-risk group according to pathology reports (negative LVSI, depth of cervical stromal invasion of ≤50%, and tumor size of ≤2 cm) (Table 3). In this low-risk group, no patients had parametrial involvement or lymph node metastases. Median follow-up time was 77 months and none of these patients received adjuvant treatment, none experienced recurrence and none died during the follow-up period.

Table 1. Characteristics of patients

Median follow-up time (months)	77.6 (11-142)
Median patient age (years)	49.5 (29-80)
Histology, n (%)	
Squamous	132 (78.6)
Adenocarcinoma	23 (13.7)
Adenosquamous	13 (7.7)
Median tumor size (cm)	2.5 (0.45-4.0)
Median depth of stromal invasion (mm)	12 (0.4-30)
DSI, n (%)	
>50 %	111 (66.1)
≤50 %	57 (33.9)
Tumor size ≤2 cm, n (%)	62 (36.9)
LVSI, n (%)	
Positive	105 (62.5)
Negative	63 (37.5)
Parametrial involvement, n (%)	
Positive	31 (18.5)
Negative	137 (81.5)
Type of Surgery, n (%)	
Radical	159 (86.8)
Fertility sparing	9 (13.2)
Median number of resected lymph nodes	38 (22-88)
Pelvic lymph nodes, n (%)	
Positive	49 (29.8)
Negative	119 (70.2)
Para-aortic lymph nodes, n (%)	
Positive	13 (7.8)
Negative	155 (92.2)
Adjuvant Treatment, n (%)	
NAT	95 (56.5)
RT	21 (12.5)
CRT	48 (28.5)
Chemo	4 (2.5)
DSI: Deep stromal invasion, LVSI: Lympho-vascular space invasion, NAT: No adjuvant treatment, RT: Radiotherapy, CRT: Chemo-radiotherapy, Chemo: Chemoterapy	

Discussion

In recent years, gynecologic oncologic surgeons have focused on performing less radical surgeries for low-risk patients. Therefore, in this study, we evaluated the factors affecting parametrial involvement in cervical cancer patients with low-risk factors. First, positive LVSI, DSI, and lymph node metastases increased the risk of parametrial involvement in all patients. Second, in subgroup analyses, there were no parametrial involvements or lymph node metastases in any patients in the low-risk patient group and no recurrence or death was observed in the 77 months follow-up of these patients, who did not receive any adjuvant therapy.

One of the important points to be considered while planning the treatment of cervical cancer is determination of the risk group of the patient. Therefore, studies have investigated various

Table 2. Univariate and multivariate analyses of factors affecting parametrial involvement

	Univariate analyses		**Multivariate analyses**		
Factor	**Parametrial involvement**	**p**	**HR**	**(95% CI) Lower-upper**	**p**
LVSI					
Positive	27/105 (25.7%)	0.001	3.45	1.1-10.8	0.034
Negative	4/63 (6.3%)				
Tumor size					
≤2 cm	7/62 (11%)	0.008	1.16	0.3-3.4	0.76
>2 cm	24/106 (22%)				
DSI					
≤50%	3/57 (5.2%)	0.001	4.1	1.18-14.8	0.03
>50%	28/111 (25.2%)				
Age in years					
>49.5	20/84	0.072	1.1	0.8-1.2	0.09
≤49.5	11/84				
Lymph node Metastases	18/49 (36%)	0.001	3.2	1.3-7.5	0.008
Histology	Squamous 22/130 (16%)	0.4	-	-	-
	Adeno 5/22 (22%)				
	ASQ 4/13 (30%)				
DSI: Deep stromal invasion, LVSI: Lympho-vascular space invasion, ASQ: Adenosquamous, CI: Confidence interval					

Table 3. Parametrial involvement rates of patient with tumor size ≤2 cm according to LVSI and DSI

DSI			Parametrial involvement		Total (n)	Percentage %
			Negative (n)	Positive (n)		
≤%50	LVSI	Negative	26	0	26	0
		Positive	13	3	16	23.07
	Total		39	3	42	7.14
>%50	LVSI	Negative	6	0	6	0
		Positive	8	4	12	33.3
	Total		14	4	18	22.2
DSI: Deep stromal invasion, LVSI: Lympho-vascular space invasion						

factors in order to correctly classify patients. In the study of Covens et al. (20), which included 842 stage 1A1-1B1 patients who underwent radical surgery, there was a parametrial involvement rate of 0.6% in 536 patients with tumor size of less than 2 cm, DSI of <10 mm, and negative lymph nodes. In another study, advanced age, poor histology, large tumor size, DSI, LVSI, vaginal involvement, and pelvic or para-aortic lymph node metastasis were demonstrated as factors predicting the high-risk group (13). When subgroup analysis was performed, the same authors found that for negative LVSI and tumor size of ≤2 cm, the parametrial involvement rate in patients without lymph node metastasis was 0.4% (13). In our study, similar results were obtained, and in subgroup analyses, there ws no parametrial involvement or lymph node metastases in any patient in the low-risk group and no recurrence or death observed in the 6-year follow-up of these patients. All of the preceding retrospective studies and our data show that the parametrial involvement rate in patients within the low-risk group is less than 1%. This should be kept in mind so that more conservative surgeries may be safe and viable treatment options for patient groups with these predictive factors. In our cohort the percentage of parametrial involvement in patients with a tumor size less than 2 cm was 11% and this number is relatively high compared to earlier studies. The main reason for this inconsistency seems to be the high rates of positive LVSI and DSI in this sub-group.

In the literature, there are many studies concerning the oncologic outcome in patients with early-stage cervical cancer who underwent less radical surgeries, such as simple trachelectomy, conization, or simple hysterectomy (21-23). In these studies, the oncologic results were satisfactory and, according to these outcomes, gynecologic oncologic surgeons and patients are still expected to shift towards less radical surgery, although many centers continue to perform radical surgeries in this patient group. The most important reason for this is the lack of convincing evidence to the contrary in this area. Although the results of our study contribute to and confirm the previous reports, and we recommend less radical surgery, further prospective studies are still required. Our results should be further supported with the results of ongoing prospective studies, including the SHAPE and CONCERV studies, which are investigating this topic (24-25).

Study Limitation

The present study has some limitations. The first and main limitation is the retrospective design; as a result, the possibility of selection bias could not be completely excluded. Another limitation is that, although comparable to similar studies in the literature, the number of patients analyzed in the subgroups was relatively low. This particular study shows post-operative risk factors for parametrial involvement but these risk factors should be combined with pre-operative findings, such as magnetic resonance imaging and biopsy histopathological reports.

Conclusion

At present, the available data suggest that patients with low-risk, early-stage cervical cancer maybe good candidates for conservative surgery so that conization, simple trachelectomy, or simple hysterectomy and pelvic lymphadenectomy may be good options for these patients. For this reason, all patients with cervical cancer should be examined in detail with pre-operative pelvic examination and imaging methods, and less radical surgery options should be considered in patients with low-risk factors following careful assessment.

Ethics Committee Approval: *This study was approved by the Başkent University Faculty of Medicine Institutional Review Board (approval number: KA13/252).*

Informed Consent: *Due to the retrospective design, informed consent from involved patients was waived.*

Peer-review: *Externally and internally peer-reviewed.*

Author Contributions: *Surgical and Medical Practices: A.A., H.A., N.H.R.; Concept: A.A., H.A., E.G., N.H.R.; Design: A.A.,*

Y.A.T.; Data Collection or Processing: İ.K., M.T.; Analysis or Interpretation: H.A., E.G.; Literature Search: İ.K., M.T.; Writing: H.A., A.A., Y.A.T.

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References

1. Parkin DM, Bray F, Ferlay J, Pisani P. Global cancer statistics, 2002. CA Cancer J Clin 2005; 55: 74-108.
2. Plante M, Gregoire J, Renaud MC, Roy M. The vaginal radical trachelectomy: an update of a series of 125 cases and 106 pregnancies. Gynecol Oncol 2011; 121: 290-7.
3. Querleu D, Cibula D, Abu-Rustum NR. 2017 Update on the Querleu-Morrow Classification of Radical Hysterectomy. Ann Surg Oncol 2017; 24: 3406-12.
4. Creasman WT, Zaino RJ, Major FJ, DiSaia PJ, Hatch KD, Homesley HD. Early invasive carcinoma of the cervix (3 to 5 mm invasion): risk factors and prognosis. A gynecologic oncology group study. Am J Obstet Gynecol 1998; 178(1 Pt 1): 62-5.
5. Quinn MA, Benedet JL, Odicino F, Maisonneuve P, Beller U, Creasman WT, et al. Carcinoma of the cervix uteri. FIGO 26th annual report on the results of treatment in gynecological cancer. Int J Gynaecol Obstet 2006; 95 Suppl 1: S43-103.
6. Gadducci A, Sartori E, Maggino T, Landoni F, Zola P, Cosio S, et al. The clinical outcome of patients with stage Ia1 and Ia2 squamous cell carcinoma of the uterine cervix: a cooperation task force (CTF) study. Eur J Gynaecol Oncol 2003; 24: 513-6.
7. Ramirez PT, Pareja R, Rendon GJ, Millan C, Frumovitz M, Schmeler KM. Management of low-risk early-stage cervical cancer: should conization, simple trachelectomy, or simple hysterectomy replace radical surgery as the new standard of care? Gynecol Oncol 2014; 132: 254-9.
8. Ayhan A, Tohma YA, Sahin H, Kocaman E, Tunc M, Haberal AN. Oncological and obstetric outcomes after fertility-sparing radical abdominal trachelectomy for early stage cervical cancer: a tertiary centre's 10 years' experience. J Obstet Gynaecol 2019; 39: 248-52.
9. Potish RA, Twiggs LB, Okagaki T, Prem KA, Adcock LL. Therapeutic implications of the natural history of advanced cervical cancer as defined by pretreatment surgical staging. Cancer 1985; 56: 956-60.
10. Roman LD, Felix JC, Muderspach LI, Agahjanian A, Qian D, Morrow CP. Risk of residual invasive disease in women with microinvasive squamous cancer in a conization specimen. Obstet Gynecol 1997; 90: 759-64.
11. Kinney WK, Hodge DO, Egorshin EV, Ballard DJ, Podratz KC. Identification of a low-risk subset of patients with stage IB invasive squamous cancer of the cervix possibly suited to less radical surgical treatment. Gynecol Oncol 1995; 57: 3-6.
12. Naik R, Cross P, Nayar A, Mayadevi S, Lopes A, Godfrey K, et al. Conservative surgical management of small-volume stage IB1 cervical cancer. BJOG 2007; 114: 958-63.
13. Wright JD, Grigsby PW, Brooks R, Powell MA, Gibb RK, Gao F, et al. Utility of parametrectomy for early stage cervical cancer treated with radical hysterectomy. Cancer 2007; 110: 1281-6.
14. Frumovitz M, Sun CC, Schmeler KM, Deavers MT, Reis RD, Levenback CF, et al. Parametrial involvement in radical hysterectomy specimens for women with early-stage cervical cancer. Obstet Gynecol 2009; 114: 93-9.
15. Fagotti A, Gagliardi ML, Moruzzi C, Carone V, Scambia G, Fanfani F. Excisional cone as fertility-sparing treatment in early-stage cervical cancer. Fertil Steril 2011; 95: 1109-12.
16. Palaia I, Musella A, Bellati F, Marchetti C, Di Donato V, Perniola G, et al. Simple extrafascial trachelectomy and pelvic bilateral lymphadenectomy in early stage cervical cancer. Gynecol Oncol 2012; 126: 78-81.
17. Raju SK, Papadopoulos AJ, Montalto SA, Coutts M, Culora G, Kodampur M, et al. Fertility-sparing surgery for early cervical cancer-approach to less radical surgery. Int J Gynecol Cancer 2012; 22: 311-7.
18. Bhatla N, Berek JS, Cuello Fredes M, Denny LA, Grenman S, Karunaratne K, et al. Revised FIGO staging for carcinoma of the cervix uteri. Int J Gynaecol Obstet 2019; 145: 129-35.
19. Dostalek L, Avall-Lundqvist E, Creutzberg CL, Kurdiani D, Ponce J, Dostalkova I, et al. ESGO survey on current practice in the management of cervical cancer. Int J Gynecol Cancer 2018; 28: 1226-31.
20. Covens A, Rosen B, Murphy J, Laframboise S, DePetrillo AD, Lickrish G, et al. How important is removal of the parametrium at surgery for carcinoma of the cervix? Gynecol Oncol 2002; 84: 145-9.
21. Pluta M, Rob L, Charvat M, Chmel R, Halaska M Jr, Skapa P, et al. Less radical surgery than radical hysterectomy in early stage cervical cancer: a pilot study. Gynecol Oncol 2009; 113: 181-4.
22. Maneo A, Sideri M, Scambia G, Boveri S, Dell'anna T, Villa M, et al. Simple conization and lymphadenectomy for the conservative treatment of stage IB1 cervical cancer. An Italian experience. Gynecol Oncol 2011; 123: 557-60.
23. Biliatis I, Kucukmetin A, Patel A, Ratnavelu N, Cross P, Chattopadhyay S, et al. Small volume stage 1B1 cervical cancer: Is radical surgery still necessary? Gynecol Oncol 2012; 126: 73-7.
24. Plante M. A randomized trial comparing radical hysterectomy and pelvic node dissection vs simple hysterectomy and pelvic node dissection in patients with low risk early stage cervical cancer. NCIC Clinical Trials Group. NCT016589309.ClinicalTrials.gov The SHAPE Trial; 2013.
25. Schmeler K, Pareja R, Lopez A, Fregnani JH, Lopes A, Perrotta M, et al. 23 Concerv: a prospective trial of conservative surgery for low-risk early stage cervical cancer. Int J Gynecol Cancer 2019; 29: A14-5.

Investigation of *Catechol-O-methyltransferase (COMT)* gene Val158Met polymorphism in ovarian cancer

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Abstract

Objective: Catechol-O-methyltransferase (COMT), the product of the *COMT* gene, detoxifies the carcinogenic catechol estrogens. The aim of the present study was to examine the relationship between *COMT Val158Met* polymorphism and the risk of ovarian cancer.

Material and Methods: The study groups consist of 94 individuals as a patients group with ovarian cancer (n=47) and control group (n=47). The allele and genotype frequencies were determined according to Hardy-Weinberg equilibrium (HWE). The allele and genotype frequencies. determined according to HWE. Genetic analysis were performed by real-time-polymerase chain reaction instrument, and the statistical analysis were performed by SPSS program.

Results: Although no significant relationship was obtained among groups (p=0.413) regarding *COMT* gene Val158Met polymorphism, the genotype frequencies for COMT Val158Met (rs4860) polymorphism in groups was homozygote wild type GG genotype 25.5%, heterozygote GA genotype 46.8%, homozygote mutant AA genotype 27.7%.

Conclusion: This study is the first to investigate the relationship between ovarian cancer and the *Val158Met* polymorphism in the *COMT* gene in a Turkish population. No statistically significant relationship was identified among genotypes belonging to the patient and control groups although sample sizes were relatively small and the analysis should be repeated in a larger cohort. (J Turk Ger Gynecol Assoc 2021; 22: 42-6)

Keywords: *COMT Val158Met* polymorphism, catechol estrogen, ovarian cancer

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Introduction

Ovarian cancer is the seventh leading cause of death and is the eighth most common cancer among women. It has the highest mortality rate among all gynecological cancers. The prognosis of ovarian cancer is poor, especially when the disease is diagnosed at an advanced stage (1). There are many risk factors, which change the genetic predisposition to ovarian cancer, including consumption of alcohol, obesity, aging and family history. In addition, oxidative stress, inflammation, angiogenesis, and apoptosis may alter the progression of carcinoma. Thus, controlling these factors play a crucial role in the prevention of many cancers, including ovarian carcinoma (2).

The *Catechol-O-methyltransferase (COMT)* gene is located on chromosome 22q11.2. The *COMT* gene includes six exons, of which exon 1 and 2 are noncoding (3). *COMT* gene products are expressed in many tissues, such as bone marrow, brain, bladder, heart, kidney, liver, lung, and ovary (4). The enzyme product, COMT participates in DNA repair mechanisms (5). Many polymorphisms have been identified in the coding region of the *COMT* gene including at codon 158 (G→A), codon 72 (G→T) and codon 62 (C→T). At codon 158, valine may be

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replaced by methionine and this alteration in the *COMT* gene lead to a decrease in the function of the protein product (6,7).

Catechols can be of endogenous or exogenous origin (8). Some evidence indicates that reactive catechol metabolites from estrogens, the catechol estrogens, have a reactive effect which may contribute to a predisposition to cancer. The COMT enzyme Catalyzes O-methylation, which has a role in inactivation of catechol estrogens (9). COMT catalyzes the transfer of the methyl group in the coenzyme SAM to the hydroxyl group in the catechols. In the *COMT* gene, the *Val158Met* polymorphism results in a decrease in the activity of this enzyme, resulting in accumulation of carcinogenic catechol estrogens (10).

The aim of the present study was to examine the relationship of the *COMT Val158Met* polymorphism to disease by comparing polymorphism expression in patients with ovarian cancer and healthy controls.

Material and Methods

The present study investigated COMT polymorphism frequency in 94 individuals, including patients with ovarian cancer and an equal number of healthy controls. The patients (n=47) were attending the Yeditepe University Faculty of Medicine, Department of Obstetrics and Gynecology, İstanbul, Turkey. The age-matched control group (n=47) consisted of healthy females who visited the hospital for gynecologic evaluation as part of routine check up. All procedures performed in studies involving human participants were in accordance with the ethical standards of the 1975 Declaration of Helsinki guidelines and its later amendments. The study protocol was approved by Yeditepe University Faculty of Medicine Ethics Committee (approval number: 915, date: 25.10.2018). Informed consent was obtained from all participants, prior to enrollment in the study.

Genomic DNA isolation from blood

Blood samples of participants were taken into EDTA tubes to a volume of 5 mL. DNA isolation was performed by the iPrep DNA Extraction Robot (Invitrogen, Carlsbad, California, USA). DNA purity and concentration was determined by NanoDrop (Invitrogen, Carlsbad, California, USA).

Genotyping analysis

The allele and genotype frequencies determined according to Hardy-Weinberg equilibrium. Analysis of *COMT* gene variants performed by using the 7500 fast-real-time-polymerase chain reaction (RT-PCR) instrument (Applied Biosystems, Foster City California, USA). *COMT Val158Met (rs4860)* gene variations determined by using TaqMan Genotyping Assay probe (Applied Biosystems, Foster City, CA, USA). COMT Val158Met (rs4860) polymorphism determined by Forward 5'-GGA GCT GGG GGC CTA CTG TG-3' and Reverse 5'-GCC CTT TTT CCA GGT CTG ACA-3' primers. Reaction mixture and conditions were used due to manufacturer instructions. The conditions for the RT-PCR reaction were; holding stage for 10 minutes at 95 °C and 40 repeating cycles with denaturation stage for 15 seconds at 92 °C and connecting/elongation stage for 1 minute at 60 °C.

Statistical analysis

The data obtained from genotyping was evaluated using chi-square and Fisher's exact tests using SPSS, version 25.0 (IBM Inc., Armonk, NY, USA). Student's t-test was used to analyze numeric values. A $p<0.05$ were considered statistically significant.

Results

As a result of the demographic data analysis, body mass index (BMI), body surface area and fasting blood glucose values in ovarian cancer patients were found to be significantly higher than the control group. There was no difference between the patients and controls regarding mean age ($p=0.154$). While significantly differences were deteceted in diabetes ($p<0.001$), and smoking ($p<0.001$) and these risk factors were found to be statistically meaninful among groups. Also menopausal status (premenopausal and postmenopausal), pregnancy status (number of pregnancies ≤1 or number of pregnancies >1) and parity (number of births ≤1 or number of births >1) are statistically meaningful. In the patients with ovarian cancer, the rate of postmenopausal women (80.9%) was higher than the control group (40.4%). The controls whose number of births were less than or equal to one (number of births ≤1) (54.4%) were found to be statistically higher than the patient group (26.7%). The rate of postmenopausal women (80.9%) in the patients with ovarian cancer was higher than the control group. While the rate of pregnancy status (number of pregnancies ≤1) (54.4%) in the control group was higher than the patient group. (Table 1).

In the present study, in the patients with ovarian cancer, the distribution of premenopause (80%) was analyzed to be high in comparison with postmenopause state (20%). The treatment parameters were resulted that 72.1% of the patients received adjuvant chemotherapy and 27.9% of the patients did not receive adjuvant chemotherapy. While 36.4% of the patients received neoadjuvant chemotherapy and 63.6% of the patients did not receive neoadjuvant chemotherapy. 47.5% of the patients underwent debulking surgery. Also it was observed that the rates of being pregnant more than once (77.8%) and a number of births (>1) were high (73.3%) in the patients with ovarian cancer. The ratio of patients at stage 3 (42.9%) were found to be high as compared to stage 1 (23.8%), stage 2 (23.8%) and stage 4 (14.3%). When we evaluated the tumors in terms of cell types, epithelial tumors were found at a high rate (92.3%) in comparison to the other types. Serous epithelial

tumors from epithelial tumor types were calculated in a ratio of 56.4%. The ratio of sex-cord stromal tumors (5.3%) was found to be higher than germ cell tumors (2.6%).

The allele and genotype frequencies for *COMT Val158Met (rs4860)* polymorphism in groups are shown in Table 2. There was no significant difference between the proportions of patients and healthy controls in terms of polymorphism frequency (p=0.413). However, there was a tendency for patients to express the G-allele (56.4%) compared to the controls (48.9%) (p=0.389). Accordingly, there was also no difference in the expression of the A-allele between patients and controls (p=0.301).

Discussion

Several molecular signaling pathways as hormone signaling, apoptosis, angiogenesis and oxidative stress, play crucial roles in ovarian cancer such progression of ovarian cancer (11). When the individuals with ovarian cancer are at a late-stage, 70% of patients are able to be diagnose. On the other hand, symptoms of ovarian cancer are not clear, and survival rate is almost 90% for 5-years during the first stages (12).

It is considered that COMT enzyme plays a significant role in estrogen metabolism based on many different studies (13-16). Ovarian cancer development is influenced by various risk factors such as BMI, age, tumor histology, family history of patients with ovarian cancer and smoking. Therefore, there are many factors that create a risk for ovarian cancer. In a nutshell, it is not sufficient to diagnose individuals simply according to carrying the Val158Met polymorphism in the *COMT* gene (14).

Goodman et al. (15) reported that COMT Val158Met polymorphism was not related to ovarian cancer risk due to a limited sample size. That study contained 108 cases and 106 controls from the German population. The ratio of heterozygote

Table 1. Demographic data related to the patients with ovarian cancer and healthy controls

		Control group (n=47) %	Patient group (n=47) %	p
Body mass index, $\overline{X}$ ± SD (kg/m^2)		23.04±3.62	29.31±5.37	<0.0001* (S)
Age, $\overline{X}$ ± SD (years)		51.11±12.86	54.87±12.55	0.154 (NS)
Body surface area, $\overline{X}$ ± SD (m^2)		1.66±0.14	1.77±0.15	<0.0001* (S)
Fasting blood glucose $\overline{X}$ ± SD (mg/dL)		86.45±7.60	108.52±33.28	<0.0001* (S)
Smoking	Yes (%)	(n=26) 55.3%	(n=37) 82.2%	<0.0001* (S)
	No (%)	(n=21) 44.7%	(n=1) 17.8%	
Menopause	Postmenopause (%)	(n=19) 40.4%	(n=38) 80.9%	<0.0001* (S)
	Premenopause (%)	(n=28) 59.6%	(n=9) 19.1%	
History of diabetes	Yes (%)	(n=0) 0%	(n=12) 26.7%	<0.0001* (S)
	No (%)	(n=47) 100%	(n=33) 73.3%	
Parity	≤1	(n=27) 54.4%	(n=12) 26.7%	<0.0001* (S)
	>1	(n=20) 42.6%	(n=33) 73.3%	
Pregnant state	≤1	(n=27) 54.4%	(n=10) 22.2%	<0.0001* (S)
	>1	(n=20) 42.6%	(n=35) 77.8%	

Method for statistical analysis: The difference between the groups was analyzed by the chi-square test and independent sample Student's t-test.
n: Number, $\overline{X}$ ± SD: Mean value ± standard deviation, *(S) Significant, NS: Non-significant

Table 2. The genotype and allele distributions for the *COMT* gene between the patient and control groups

	Control group (n=47)	Patient group (n=47)	p	OR	CI (95%)
COMT genotype			0.413		
GG	25.5% (n=12)	37.8% (n=17)	0.216 (NS)	1.77	0.727-4.314
GA	46.8% (n=22)	42.2% (n=19)	0.658 (NS)	0.83	0.364-1.892
AA	27.7% (n=13)	20% (n=9)	0.547 (NS)	0.747	0.289-1.932
Allele count					
G	(48.9%) 46	(56.4%) 53	0.389 (NS)	1.529	0.579-4.037
A	(51.1%) 48	(39.4%) 37	0.301 (NS)	0.630	0.262-1.516

Method for statistical analysis: The difference between the groups was analyzed by the chi-square test.
n: Number of sample, NS: Non-significant (p>0.05), COMT: Catechol-O-methyltransferase, OR: Odds ratio

genotype carriers (50%) with ovarian cancer was found to be high in comparison with homozygote wild and variant type genotype carriers (25%, 25%) in the cases. There was no evidence that COMT Val158Met polymorphism increases the risk of ovarian cancer (p=0.73), and Goodman et al. (15) implied that advance studies are required to explain different combinations of polymorphisms in estrogen metabolizing enzymes.

A multigenic model constructed by eleven gene variations including the COMT Val158Met polymorphism, was performed by Delort et al. (16). Although there was no significant difference, they suggested that heterozygote COMT Val158Met polymophism was one of high-risk genotype. Heterozygote COMT Val158Met genotype could have possible effect on ovarian cancer by reducing activity of phase II enzyme that decrease the elimanition of carcinogens (16). Pan and Liao (17) investigated that COMT might be as a biomarker, which can be important factor in suppressing tumor development and treatment of cancer . In estrogen metabolism, COMT prevents DNA damage, therefore, it is called the gate-keeper (17). In light of these developments, present study will contribute to understanding molecular mechanisms for ovarian cancer.

There are two meta-analyses perfomed in order to determine the role of COMT Val158Met polymorphism in ovarian cancer susceptibility and both of them could not find any associations (8,18). But it should not be forgotten that meta-analysis data could be tested in clinical studies by recruiting homogeneous patients and controls. Moreover, in meta-analysis there are population bias caused by homogeneity, and they did not implicate any data for Turkish population. Thus, our study aim to provide reliable data about the role of COMT Val158Met polymorphism in ovarian cancer due to these uncertain results.

Conclusion

As *COMT* polymorphisms affect enzyme capacity, the *COMT* gene has a significant role in estrogen metabolism and thus on hormone dependent gynecological cancers. Although, there were no statistically significant differences between the patient and control groups in the present study this may have been due to the small sample sizes. Therefore associations between *COMT* gene variations and ovarian cancer risk should be investigated in larger sample sized studies in a Turkish population.

Ethics Committee Approval: *The study protocol was approved by Yeditepe University Faculty of Medicine Ethics Committee (approval number: 915, date: 25.10.2018).*

Informed Consent: *Informed consent was obtained from all participants, prior to enrollment in the study.*

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References

1. Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin 2018; 68: 394-424.
2. Doubeni CA, Doubeni AR, Myers AE. Diagnosis and management of ovarian cancer. Am Fam Physician 2016; 93: 937-44.
3. Grossman MH, Emanuel BS, Budarf ML. Chromosomal mapping of the human catechol-O-methyltransferase gene to 22q11.1-q11.2. Genomics 1992; 12: 822-5.
4. COMT catechol-O-methyltransferase [Homo sapiens (human)] https://www.ncbi.nlm.nih.gov/gene/1312#gene-expression (Access date 18.04.2019)
5. Zhou Q, Wang Y, Chen A, Tao Y, Song H, Li W, et al. Association between the COMT Val158Met polymorphism and risk of cancer: evidence from 99 case-control studies. Onco Targets Ther 2015; 8: 2791-803.
6. Lachman HM, Papolos DF, Saito T, Yu YM, Szumlanski CL, Weinshilboum RM. Human catechol-O-methyltransferase pharmacogenetics: description of a functional polymorphism and its potential application to neuropsychiatric disorders. Pharmacogenetics 1996; 6: 243-50.
7. Saito S, Iida A, Sekine A, Miura Y, Sakamoto T, Ogawa C, et al. Identification of 197 genetic variations in six human methyltransferase genes in the Japanese population. J Hum Genet 2001; 46: 529-37.
8. Zou LW, Xu XJ, Liu T, Wang HY, Fan WJ, Wang XH, et al. No association between COMT Val158Met polymorphism and prostate cancer risk: A meta-analysis. Genet Test Mol Biomarkers 2013; 17: 78-84.
9. Lavigne JA, Helzlsouer KJ, Huang HY, Strickland PT, Bell DA, Selmin O, et al. An association between the allele coding for a low activity variant of catechol-O-methyltransferase and the risk for breast cancer. Cancer Res 1997; 57: 5493-7.
10. Guldberg HC, Marsden CA. Catechol-O-methyl transferase: pharmacological aspects and physiological role. Pharmacol Rev 1975; 27: 135-206.
11. Grayson K, Gregory E, Khan G, Guinn BA. Urine biomarkers for the early detection of ovarian cancer-are we there yet? Biomark Cancer 2019; 11: 1179299X19830977.
12. Dochez V, Caillon H, Vaucel E, Dimet J, Winer N, Ducarme G. Biomarkers and algorithms for diagnosis of ovarian cancer: CA125, HE4, RMI and ROMA, a review. J Ovarian Res 2019; 12: 28.
13. Lin H, Pizer ES, Morin PJ. A frequent deletion polymorphism on chromosome 22q13 identified by representational difference analysis of ovarian cancer. Genomics 2000: 69; 391-4.

14. Goodman JE, Lavigne JA, Hengstler JG, Tanner B, Helzlsouer KJ, Yager JD. Catechol-O-methyltransferase polymorphism is not associated with ovarian cancer risk. Cancer Epidemiol Biomarkers Prev 2000: 9; 1373-6.
15. Goodman MT, McDuffie K, Kolonel LN, Terada K, Donlon TA, Wilkens LR, et al. Case-control study of ovarian cancer and polymorphisms in genes involved in catecholestrogen formation and metabolism. Cancer Epidemiol Biomarkers Prev 2001; 10: 209-16.
16. Delort L, Chalabi N, Satih S, Rabiau N, Kwiatkowski F, Bignon YJ, et al. Association Between Genetic Polymorphisms and Ovarian Cancer Risk. Anticancer Res 2008; 28: 3079-82.
17. Pan W, Liao H. Correlations between the COMT gene rs4680 polymorphism and susceptibility to ovarian cancer. Genet Mol Res 2015; 14: 16813-8.
18. Liu JX, Luo RC, Li R, Li X, Guo YW, Ding DP, et al. Lack of associations of the COMT Val158Met polymorphism with risk of endometrial and ovarian cancer: a pooled analysis of case-control studies. Asian Pac J Cancer Prev 2014; 15: 6181-6.

Endometrial scratching for poor responders based on the Bologna criteria in ICSI fresh embryo transfer cycles: a preliminary retrospective cohort study

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Abstract

Objective: This study evaluated the effect of endometrial injury on pregnancy outcomes in patients with a poor ovarian response (POR), based on the Bologna criteria, who underwent intracytoplasmic sperm injection (ICSI) cycles.

Material and Methods: Sixty-eight patients were enrolled in this retrospective cohort study. All patients in the endometrial scratching group (group 1, n=32) and control group (group 2, n=36) underwent office hysteroscopy in the early follicular phase of the cycle before controlled ovarian stimulation. Group 1 also underwent endometrial scratching. The main outcome measure was the ongoing pregnancy rate.

Results: The study groups had similar baseline demographics, including age, body mass index, duration of infertility, number of ICSI cycles, and hormone levels. However, the antral follicle count was significantly higher in group 1 than in group 2 (4.2±1.9 vs 3.3±1.8; $p<0.05$). There were no significant group differences in ovarian stimulation characteristics (ovarian stimulation time, trigger day endometrial thickness, number of metaphase II oocytes), number of embryos transferred, or the ratio of embryo transfer on days 3 and 5. Moreover, there were no significant differences between groups 1 and 2 in the rates of chemical pregnancy (25% vs 19.4%), clinical pregnancy (15.6% vs 11.1%) or ongoing pregnancy (9.4% vs 8.3%) ($p>0.05$ for all).

Conclusion: Endometrial scratching did not improve pregnancy outcomes for patients meeting the Bologna criteria for a POR to ICSI cycles using fresh embryo transfer and the GnRH antagonist protocol. (J Turk Ger Gynecol Assoc 2021; 22: 47-52)

Keywords: Endometrial scratching, Bologna criteria, poor ovarian response, ICSI, GnRH antagonist protocol

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Introduction

The world's first in-vitro fertilization (IVF) baby was born in 1978. Since then, according to the International Committee for Monitoring Artificial Reproductive Technology, approximately 8 million babies have been born worldwide by means of IVF or other advanced infertility treatment methods. However, pregnancy rates in Europe appear to have stabilized, at approximately 36%, for both IVF and intracytoplasmic sperm injection (ICSI) treatment cycles (1).

The number of oocytes retrieved during an IVF cycle (ideally about 15) is directly related to the success thereof (2). A poor ovarian response (POR), in which controlled ovarian stimulation (COS) during IVF/ICSI cycles yields a limited number of oocytes, has an average prevalence of 6% to 35% (3,4). POR is frustrating for both patients and clinicians, as it is related to low pregnancy rates in IVF and high cancellation rates during COS (5). Thus, strategies to increase pregnancy rates in patients with POR are important in IVF/ICSI cycles.

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Embryo implantation is one of the most important steps in the IVF cycle. Implantation failure may be caused by low endometrial receptivity, embryo problems, or abnormalities of both the endometrium and embryo. As reported previously, about 66% of implantation failures are caused by decreased endometrial receptivity (6). Thus, ensuring a receptive endometrium could increase pregnancy and delivery rates for artificial reproductive technology (ART). Various techniques have been used to ensure a receptive endometrium, including management of intracavitary abnormalities by hysteroscopy, treatment of thin endometrium, immunotherapy, and adjuvant treatments for women with POR (7,8).

Endometrial scratching, which is also called endometrial biopsy, endometrial injury, and endometrial trauma, has been offered by clinicians as a means of increasing endometrial receptivity, by an as yet unknown mechanism (9). Endometrial scratching can be done with endometrial biopsy instruments during the early follicular or luteal phase of the preceding IVF treatment cycle. The first study demonstrating a substantial increase in pregnancy rates after endometrial scratching was followed by many additional studies and reviews including various populations, although these have had conflicting results (10-15). However, no study has directly addressed the effect of endometrial scratching on patients with POR.

The objective of this preliminary trial was to assess the role of endometrial scratching in patients with a POR undergoing ICSI cycles with fresh embryo transfer.

Material and Methods

This study included 68 women seen at a private IVF Center in Koru Hospital, Ankara, Turkey. Approval was obtained from the Koru Hospital Ethical Committee before the study commenced (approval number: 81, date: 04/11/2019). The study population was enrolled between January 1, 2017 and November 1, 2019. The Declaration of Helsinki was followed.

All participants were undergoing their first or repeated ICSI fresh embryo transfer and met the following inclusion criteria: fulfilment of the Bologna criteria for POR (16); aged 20-42 years; body mass index (BMI) 20-30 kg/m^2; and a normal uterine cavity based on office hysteroscopy or hysterosalpingography. Patients were excluded if they had uterine anomalies, endocrine disorders, ovarian cysts, hydrosalpinx, or severe male factor infertility (e.g. aspermia, azoospermia), or if they had undergone uterine surgery in the last 3 months, or IVF cycles for preimplantation genetic diagnosis. Patients with genetic abnormalities, and those who had undergone cryo-thawed embryo transfer, were also excluded.

A detailed history, including age, previous treatments, and duration of infertility, was taken from all patients. A gynecologic examination consisting of a bimanual pelvic examination plus transvaginal ultrasonography (TVUSG) was conducted to check for structural abnormalities of the pelvis and ovaries. On days 2-5 of the menstrual cycle, gonadotropic hormone and estradiol concentrations were assessed.

All patients underwent office hysteroscopy during the early follicular phase of the menstrual cycle, i.e., immediately preceding the planned IVF cycle. A rigid 30°, 5.5 mm hysteroscope was used to perform hysteroscopy without anesthesia (Karl Storz Endoscopy, Tuttlingen, Germany). Serum physiological solution was used to distend the uterine cavity during hysteroscopy. Group 1, but not group 2, also underwent endometrial scratching during hysteroscopy; scissors were used to create mechanical tissue damage in local areas of the fundus, and in posterior and anterior regions of the endometrium.

COS began on day 2 or 3 of the menstrual cycle. Subcutaneous injections of recombinant follicle-stimulating hormone (FSH) (Gonal-F; Serono, Rome, Italy) or highly purified human menopausal gonadotropin (Menopur, Ferring, Sweden or Merional; IBSA, Collina d'Oro, Switzerland) were used for ovarian stimulation. The initial dose for each patient was based on the predicted ovarian response, and varied from 300 to 450 IU. On day 5 of COS, the flexible GnRH antagonist treatment protocol was implemented to prevent premature luteinizing hormone (LH) surge (Cetrotide; Merck Sharp and Dohme Ltd., Athens, Greece). Hormones, including FSH, estradiol, and thyroid-stimulating hormone, were measured before stimulation. LH, estradiol, and progesterone were also measured on the day of human chorionic gonadotropin (hCG) administration.

When two or more follicles with a diameter of at least 17 mm were detected, ovulation was triggered by a subcutaneous injection of 250 μgr r-hCG (Ovitrelle; Serono). Oocyte retrieval was carried out 35.5-36 hours after the r-hCG injection using TVUSG. On day 3 or day 5 after oocyte retrieval, depending on oocyte development, a maximum of two good-quality embryos were transferred into the uterine cavity using a Wallace semirigid catheter (Cooper Surgical, Malov, Denmark) under abdominal ultrasonography guidance.

Luteal support was provided until the 12th week of gestation using Crinone gel (8% progesterone Serono). A pregnancy test was conducted approximately 2 weeks after embryo transfer. Clinical pregnancy was defined as any intrauterine gestational sac with a fetal heartbeat at 4 weeks after the first pregnancy test. The existence of at least one live fetus at the 12th week of gestation was considered an ongoing pregnancy, which was the primary outcome measure.

Statistical analysis

Statistical analysis was carried out using SPSS software (version 23.0; SPSS Inc., Chicago, IL, USA). Continuous

variables are expressed as mean ± standard deviation or median (minimum-maximum). Categorical variables are expressed as number and percentage (%). The Kolmogorov-Smirnov test was used to check the distribution of the data. The Independent Samples t-test and Mann-Whitney U test were used to compare continuous variables. Pearson's chi-squared test or Fisher's exact test was used to compare categorical variables. A two-tailed p-value of <0.05 was considered significant.

Results

A total of 68 patients were included in the present study. The endometrial scratching group (group 1) and control group (group 2) included 32 and 36 women, respectively. The groups were well-balanced in terms of baseline demographic data, including age, BMI, length of infertility, number of IVF cycles, and hormone levels (Table 1). However, the antral follicle count was significantly higher in group 1 compared to group 2 (4.2±1.9 vs 3.3±1.8; p<0.05).

Table 2 shows the COS, ICSI, and pregnancy outcomes. Groups 1 and 2 did not differ in duration of COS, trigger day endometrial thickness, or number of metaphase II oocytes retrieved. In addition, the number of embryos transferred, and the ratio of day 3 to day 5 embryo transfers, were similar between the two groups. Moreover, regarding pregnancy outcomes, groups 1 and 2 had similar rates of chemical pregnancy (25% and 19.4%, respectively), clinical pregnancy (15.6% and 11.1%) and ongoing pregnancy (9.4% and 8.3%).

Discussion

This retrospective cohort trial investigated the impact of endometrial scratching on the pregnancy outcomes of women meeting the Bologna criteria for POR, who were undergoing ICSI fresh embryo transfer cycles using the GnRH antagonist protocol. The results showed that endometrial scratching did not increase rates of clinical or ongoing pregnancy.

Poor responders constitute a major challenge for ART. These patients are more likely to show poor oocyte retrieval and less favorable pregnancy outcomes. A previous review reported that 47 studies used 41 different definitions of POR (17). To overcome this challenge, ESHRE used the Bologna criteria to clearly define POR (16).

Endometrial injury may be useful in patients undergoing ART cycles, to improve endometrial receptivity and the chance of pregnancy. Although endometrial injury has been studied for over a decade, the biological mechanism by which it increases the chance of pregnancy is not clear. However, one putative mechanism is stimulation of the production of cytokines and growth factors, which are essential for endometrial receptivity and embryo implantation after endometrial scratching and subsequent healing (18).

Endometrial injury to improve the outcomes of IVF/ICSI cycles has attracted increasing attention since Granot et al. (19) conducted the first two studies in 2000 and 2003 (20). In the first study, repeated endometrial biopsies were performed to evaluate the endometrium of 12 infertile women with several unsuccessful cycles of IVF treatment. The authors noted that 11 of these women became pregnant during the first IVF cycle after endometrial biopsy (19). In the second study, a quasi-

Table 1. Baseline demographic, clinical, and laboratory characteristics of groups 1 and 2

Parameter	Group 1 (n=32)	Group 2 (n=36)	p
Age (years)	34.1±4.0	33.2±4.0	0.361[a]
	35.0 (32.0; 37.0)	34.0 (30.0; 36.5)	
Body mass index (kg/m^2)	24.7±3.4	25.4±3.8	0.754[a]
	25.3 (20.9; 27.6)	25.0 (22.5; 27.4)	
Duration of infertility (years)	7.2±3.8	5.3±2.4	0.045[a,*]
	6.5 (4.0; 9.5)	5.0 (3.0; 7.0)	
Number of cycles (n)	1.6±0.8	1.6±0.8	0.854[a]
	1.0 (1.0; 2.0)	1.0 (1.0; 2.0)	
Antral follicle count (n)	4.2±1.9	3.3±1.8	0.035[a]
	4.0 (2.0; 6.0)	3.0 (2.0; 4.0)	
Baseline FSH (IU/L)	10.2±3.0	11.8±3.7	0.123[a]
	10.1 (8.2; 11.6)	10.6 (9.0;15.1)	
Baseline estradiol (pg/mL)	50.5±19.0	52.1±19.4	0.736[b]
	47.5 (37.0; 63.0)	55.0 (36.5; 66.0)	

Data are presented as mean ± standard deviation, median (interquartile range), or number (percentage).
FSH: Follicle stimulating hormone, [a]: Mann-Whitney U test, [b]: Independent samples t-test, *Statistically significant difference

Table 2. Controlled ovarian stimulation and pregnancy outcomes of groups 1 and 2

Parameter	Group 1 (n=32)	Group 2 (n=36)	p
Stimulation period (days)	8.3±1.5	7.8±1.5	0.146[a]
	8.0 (7.0; 9.0)	8.0 (7.0; 8.0)	
Trigger day endometrial thickness (mm)	8.8±1.7	9.1±1.9	0.397[b]
	8.7 (7.2; 9.9)	8.8 (7.6; 10.0)	
MII oocytes (n)	2.4±1.3	2.6±1.4	0.622[a]
	2.0 (1.0; 3.0)	2.0 (1.5; 4.0)	
2PN embryos (n)	1.8±1.0	1.9±1.0	0.486[a]
	1.5 (1.0; 2.0)	2.0 (1.0; 2.0)	
Embryos transferred (n)	1.3±0.5	1.3±0.4	0.772[a]
	1.0 (1.0; 2.0)	1.0 (1.0; 1.5)	
Embryo transfer day, n (%)			0.725[c]
3	27 (84.4%)	32 (88.9%)	-
5	5 (15.6%)	4 (11.1%)	-
Pregnancy outcomes, n (%)			
Beta hCG positive	8 (25%)	7 (19.4%)	0.581[d]
Clinical pregnancy	5 (15.6%)	4 (11.1)	0.725[c]
Ongoing pregnancy	3 (9.4%)	3 (8.3%)	1.000[c]

Data are presented as mean ± standard deviation, median (interquartile range) or number (percentage).
hCG: Human chorionic gonadotropin, PN: Pronucleus, M: Metaphase, [a]: Mann-Whitney U test, [b]: Independent Samples t-test, [c]: Fisher's exact test, [d]: Pearson's chi-squared test

randomized prospective trial, the same authors reported that endometrial injury doubled the conception rate during IVF treatment (20).

Numerous subsequent studies have reported inconsistent results, including positive (21-23), negative (24,25), and neutral (26,27) effects of endometrial scratching in women undergoing IVF. Many reviews have also been published over the past 15 years. However, the conflicting results of randomized prospective studies are reflected in the lack of consensus in the interpretation thereof. Reviews articles have variously concluded that (i) endometrial scratching during ART cycles improves pregnancy outcomes (11,14,23); (ii) endometrial scratching during ART cycles does not improve pregnancy outcomes (12,13); and (iii) the evidence is insufficient for definitive conclusions to be drawn (23,28).

Previous trials of endometrial scratching during IVF cycles have included women with normal ovarian response (28), unselected populations (24-27), women undergoing their first IVF cycle (22), and women with one or more failed cycles (23,29,30). The present study is the first to recruit a subgroup of patients meeting the Bologna criteria for POR and undergoing an ICSI cycle with fresh embryo transfer. Our patients had similar pregnancy outcomes regardless of whether they underwent endometrial scratching during office hysteroscopy.

In most previous studies, endometrial scratching was done by endometrial aspiration using pipelle biopsy (22-26). Our study was consistent with previous ones in that scratching was completed during diagnostic hysteroscopy (28,29). The optimal time to induce endometrial injury is not clear; some propose that it should be performed during the luteal phase (22,24,25,28,29) while others suggest that it should be completed during the follicular phase (as in our study) so that there is more time for the effect of injury to become apparent (31). A previous head-to-head comparison found that proliferative-phase endometrial scratching conferred no advantage over endometrial injury during the luteal phase (32).

Study Limitation

The retrospective nature of the present study is an important limitation. Another potential limitation is that endometrial scratching was performed during office hysteroscopy. The effect of hysteroscopy on endometrial receptivity could be similar to that of endometrial injury. However, we believe that office hysteroscopy alone is insufficient for endometrial injury.

Conclusion

The present study is the first to demonstrate that endometrial scratching does not improve pregnancy outcomes in patients meeting the Bologna criteria for POR, and undergoing ICSI cycles using fresh embryos and the GnRH antagonist protocol. Further randomized prospective trials using alternative endometrial

scratching techniques and COS protocols, and larger samples drawn from this subgroup of patients, are needed.

Ethics Committee Approval: *Approval was obtained from the Koru Hospital Ethical Committee before the study commenced (approval number: 81, date: 04/11/2019).*

Informed Consent: *Retrospective study.*

Peer-review: *Externally and internally peer-reviewed.*

Author Contributions: *Surgical and Medical Practices: Ş.K., A.Y., M.D.; Concept: Ş.K., A.Y., M.D.; Design: Ş.K., A.Y., M.D.; Data Collection or Processing: Ş.K., A.Y., M.D.; Analysis or Interpretation: Ş.K., A.Y., M.D.; Literature Search: Ş.K., A.Y., M.D.; Writing: Ş.K., A.Y., M.D.*

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References

1. European Society of Human Reproduction and Embryology. More than 8 million babies born from IVF since the world's first in 1978: European IVF pregnancy rates now steady at around 36 percent, according to ESHRE monitoring. ScienceDaily, 2018.
2. Sunkara SK, Rittenberg V, Raine-Fenning N, Bhattacharya S, Zamora J, Association between the number of eggs and live birth in IVF treatment: an analysis of 400 135 treatment cycles. Hum Reprod 2011; 26: 1768-74.
3. Patrizio P, Vaiarelli A, Levi Setti PE, Tobler KJ, Shoham G, Leong M, et al. How to define, diagnose and treat poor responders? Responses from a worldwide survey of IVF clinics. Reprod Biomed Online 2015; 30: 581-92.
4. Oudendijk JF, Yarde F, Eijkemans MJ, Broekmans FJ, Broer SL. The poor responder in IVF: is the prognosis always poor?: a systematic review. Hum Reprod Update 2012; 18: 1-11.
5. Ubaldi F, Vaiarelli A, D'Anna R, Rienzi L. Management of poor responders in IVF: is there anything new? Biomed Res Int 2014; 2014: 352098.
6. Simón C, Moreno C, Remohí J, Pellicer A. Molecular interactions between embryo and uterus in the adhesion phase of human implantation. Hum Reprod 1998; 13(Suppl 3): 219-32 discussion 233-16.
7. Margalioth EJ, Ben-Chetrit A, Gal M, Eldar-Geva T. Investigation and treatment of repeated implantation failure following IVF-ET. Hum Reprod 2006; 21: 3036-43.
8. Zhang Y, Zhang C, Shu J, Guo J, Chang HM, Leung PCK. Adjuvant treatment strategies in ovarian stimulation for poor responders undergoing IVF: a systematic review and network meta-analysis. Hum Reprod Update 2020; 26: 247-63.
9. Lensen S, Sadler L, Farquhar C. Endometrial scratching for subfertility: everyone's doing it. Hum Reprod 2016; 31: 1241-4.
10. Ko JK, Ng EH. Scratching and IVF: any role? Curr Opin Obstet Gynecol 2016; 28: 178-83.
11. Vitagliano A, Di Spiezio Sardo A, Saccone G, Valenti G, Sapia F, Kamath MS, et al. Endometrial scratch injury for women with one or more previous failed embryo transfers: a systematic review and meta-analysis of randomized controlled trials. Fertil Steril 2018; 110: 687-702.e2.
12. Vitagliano A, Andrisani A, Alviggi C, Vitale SG, Valenti G, Sapia F, et al. Endometrial scratching for infertile women undergoing a first embryo transfer: a systematic review and meta-analysis of published and unpublished data from randomized controlled trials. Fertil Steril 2019; 111: 734-746.e2.
13. Farquhar C. Endometrial scratching: how much evidence do you need to stop offering this to women having in vitro fertilization? Fertil Steril 2019; 111: 1092-3.
14. Lensen S, Venetis C, Ng EHY, Young SL, Vitagliano A, Macklon NS, et al. Should we stop offering endometrial scratching prior to in vitro fertilization? Fertil Steril 2019; 111: 1094-101.
15. van Hoogenhuijze NE, Kasius JC, Broekmans FJM, Bosteels J, Torrance HL. Endometrial scratching prior to IVF; does it help and for whom? A systematic review and meta-analysis. Hum Reprod Open 2019; 2019: hoy025.
16. Ferraretti AP, La Marca A, Fauser BC, Tarlatzis B, Nargund G, Gianaroli L; ESHRE working group on Poor Ovarian Response Definition. ESHRE consensus on the definition of 'poor response' to ovarian stimulation for in vitro fertilization: the Bologna criteria. Hum Reprod 2011; 26: 1616-24.
17. Polyzos NP, Devroey P. A systematic review of randomized trials for the treatment of poor ovarian responders: is there any light at the end of the tunnel? Fertil Steril 2011; 96: 1058-61.e7.
18. Gnainsky Y, Granot I, Aldo PB, Barash A, Or Y, Schechtman E, et al. Local injury of the endometrium induces an inflammatory response that promotes successful implantation. Fertil Steril 2010; 94: 2030-6.
19. Granot I, Dekel N, Bechor E, Segal I, Fieldust S, Barash A. Temporal analysis of connexin43 protein and gene expression throughout the menstrual cycle in human endometrium. Fertil Steril 2000; 73: 381-6.
20. Barash A, Dekel N, Fieldust S, Segal I, Schechtman E, Granot I. Local injury to the endometrium doubles the incidence of successful pregnancies in patients undergoing in vitro fertilization. Fertil Steril 2003; 79: 1317-22.
21. Tumanyan A, Gemilyan M, Hambartsoumian E. Single and double endometrial scratching (ES) in infertile women with strict criteria of recurrent implantation failure (RIF). Gynecol Endocrinol 2019; 35(Suppl): 11-4.
22. Maged AM, Rashwan H, AbdelAziz S, Ramadan W, Mostafa WAI, Metwally AA, et al. Randomized controlled trial of the effect of endometrial injury on implantation and clinical pregnancy rates during the first ICSI cycle. Int J Gynecol Obstet 2018; 140: 211-6.
23. Bar G, Harlev A, Alfayumi-Zeadna S, Zeadna A, Bord I, Har-Vardi I, et al. Recurrent implantation failure: which patients benefit from endometrial scratching prior to IVF? Arch Gynecol Obstet 2020; 301: 817-22.
24. Eskew AM, Reschke LD, Woolfolk C, Schulte MB, Boots CE, Broughton DE, et al. Effect of endometrial mechanical stimulation in an unselected population undergoing in vitro fertilization: futility analysis of a double-blind randomized controlled trial. J Assist Reprod Genet 2019; 36: 299-305.
25. Frantz S, Parinaud J, Kret M, Rocher-Escriva G, Papaxanthos-Roche A, Creuxz H, et al. Decrease in pregnancy rate after endometrial scratch in women undergoing a first or second in vitro fertilization. A multicenter randomized controlled trial. Hum Reprod 2019; 34: 92-9.
26. Lensen S, Osavlyuk D, Armstrong S, Stadelmann C, Hennes A, Napier E, et al. A Randomized Trial of Endometrial Scratching before In Vitro Fertilization. N Engl J Med 2019; 380: 325-34.

27. Pluddemann A, Onakpoya I. Endometrial scratching before in vitro fertilisation does not increase live birth rate. BMJ Evid Based Med 2020; 25: 38-9.
28. Kumbak B, Sahin L, Ozkan S, Atilgan R. Impact of luteal phase hysteroscopy and concurrent endometrial biopsy on subsequent IVF cycle outcome. Arch Gynecol Obstet 2014; 290: 369-74.
29. Seval MM, Şükür YE, Özmen B, Kan Ö, Sönmezer M, Berker B, et al. Does adding endometrial scratching to diagnostic hysteroscopy improve pregnancy rates in women with recurrent in-vitro fertilization failure? Gynecol Endocrinol 2016; 32: 957-60.
30. Günther V, von Otte S, Maass N, Alkatout I. Endometrial "Scratching" An update and overview of current research. J Turk Ger Gynecol Assoc 2020; 21: 124-9.
31. Shohayeb A, El-Khayat W. Does a single endometrial biopsy regimen (S-EBR) improve ICSI outcome in patients with repeated implantation failure? A randomised controlled trial. Eur J Obstet Gynecol Reprod Biol 2012; 164: 176-9.
32. Liu W, Tal R, Chao H, Liu M, Liu Y. Effect of local endometrial injury in proliferative vs. luteal phase on IVF outcomes in unselected subfertile women undergoing in vitro fertilization. Reprod Biol Endocrinol 2017; 15: 75.

This article will be credited by TMA within the scope of CME/CPD.

Morcellation in gynecology: short review and suggestions from Turkish Society of Minimally Invasive Gynecologic Oncology

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Abstract

Morcellation allows the removal of a large uterus and fibroids through small incisions with minimally invasive surgery. It helps to prevent the complications associated with large incisions in both hysterectomy and myomectomy operations. Currently, there is much debate regarding the use of power morcellation in laparoscopic hysterectomy and myomectomy, mainly due to the risk of peritoneal dissemination of undiagnosed uterine sarcomas. Unfortunately, there is no valid pre-operative diagnostic method that can differentiate sarcomas from myomas, and the currently available scientific literature regarding morcellation is insufficient. As the Turkish Society of Minimally Invasive Gynecological Oncology, we present our consensus opinion and suggestions for the preoperative evaluation and morcellation of fibroids, in line with the recent literature. (J Turk Ger Gynecol Assoc 2021; 22: 53-7)

Keywords: Myomectomy, hysterectomy, morcellation, laparoscopic surgery, vaginal surgery

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Introduction

Morcellation is performed for reducing the size of a uterus or myoma, to ease extraction of tissues from the abdominal cavity. The history of procedure goes back to the 19th century. The first applications were mechanically made after vaginal surgery to reduce the size of the tissue (1). In this way, vaginal surgery can be also performed in when a large uterus is present, which previously generally required open surgery. In subsequent years, minimally invasive surgery started to replace most open and vaginal procedures and, as a result, the need for a new way to extract huge uteruses and myomas from smaller incisions arose.

In 1976, a laparoscopic manual morcellator, which can work through 15 mm and 10 mm incisions, was produced (2). The technical properties of subsequent equipment have improved and devices have been replaced by electromechanical morcellators which has also reduced the time required for tissue extraction (3). By 1993, the use of a morcellator with more advanced features was approved by the Food and Drug Administration (FDA) (3).

A morcellator is used when performing hysterectomy or myomectomy for large uteruses during minimally invasive surgery to avoid open surgery-related morbidities. On the other hand, if morcellation is performed in the presence of uterine malignancy, especially uterine sarcoma, which usually cannot be diagnosed preoperatively, this may cause the disease to upstage and have a negative effect on the prognosis (4,5).

In 2013, a patient was diagnosed with leiomyosarcoma after total hysterectomy, which was performed with a minimally

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invasive approach for a presumed benign uterine fibroid and in the later staging surgery, intraperitoneal spread was observed. After this case was published, a debate started about morcellator usage and in 2014 the FDA discouraged the use of laparoscopic power morcellation during hysterectomy or myomectomy for uterine fibroids (6). In November 2014, the FDA updated its recommendations and specified contraindications for morcellation (7):

1. Morcellators are contraindicated for removal of uterine tissue containing suspected fibroids in patients who are peri- or post-menopausal or are candidates for en-bloc tissue removal through the vagina or mini-laparotomy incision.

2. Morcellators are contraindicated in patients with uterine fibroids suspicious for malignancy.

However, the scientific basis of this advice was not clear and definition of perimenopause was not explained. Despite the FDA's clear advice against morcellation, some national societies have not made a strict recommendation to prohibit morcellation (8-12).

The Turkish Minimally Invasive Gynecologic Oncology Society formed a working group on this subject and prepared suggestions in the light of current literature, which will guide both surgeons and patients.

Uterine sarcoma types and occult sarcoma risk in presumed myoma

Endometrial adenocarcinoma constitutes nearly 95% of all uterine malignant tumors (13). Mostly, diagnosis is obtained through pre-operative endometrial sampling and it is rarely diagnosed incidentally after hysterectomy. However, pre-operative diagnosis of uterine sarcomas, which make up 5% of uterine tumors, is not possible most of the time (13). These patients are at highest risk from inappropriate morcellation.

Leiomyosarcoma, endometrial stromal sarcoma and rhabdomyosarcoma are most common types of uterine sarcomas. Unfortunately, the exact rate of post-operative sarcoma diagnosis is not known in patients who are presumed to have benign fibroids pre-operatively. Since this is a rare situation, most of the relevant studies are retrospective and contain much bias. In a report of the FDA, the incidence of all sarcomas and leiomyosarcomas were reported as 1/350 and 1/458, respectively (6,7).

However, it is also seen that this rate varies according to the method of studies. In a meta-analysis of 133 studies (14), occult leiomyosarcoma risk was calculated as 1/1960 when both retrospective and prospective studies were included, but this rate dropped to 1/8300 when only prospective studies were considered. In studies investigating the incidence of sarcoma in patients who underwent morcellation during myomectomy or hysterectomy for presumed benign disorders, the highest rate was reported as 0.6% (15). Recently, two studies from Turkey reported the incidence of occult uterine sarcomas (16,17). Topdagi Yilmaz et al. (16) reported the incidence of unexpected uterine sarcoma in patients who underwent hysterectomy for benign indications as 0.6% (7/1050). In addition, Yorgancı et al. (17) investigated the rate of occult uterine sarcoma in 18,604 women who underwent hysterectomy or myomectomy with a pre-operative diagnosis of uterine leiomyoma and occult uterine sarcoma incidence was 0.3% (56/18604).

Possible adverse effects of morcellation: sarcoma and benign conditions

Morcellation can be performed manually, either using scissors or scalpel, or power morcellation can be performed using electromechanical devices. It can be performed during minimally invasive surgery or vaginal surgery. The procedure can be performed un-contained, contained (in bag) or using a mini-laparotomic incision.

After hysterotomy, regardless of morcellation, malignant cells, if present, may spread to the peritoneal cavity. During morcellation, the specimen is divided into smaller pieces in the peritoneal cavity and, irrespective of the malignancy potential, some problems may arise including spread of tissues into the peritoneal cavity, incomplete removal of tissue fragments, and microscopic residues becoming peritoneal implants. Thus, an increased incidence of benign peritoneal diseases, including parasitic leiomyoma, endometriosis and extensive intraperitoneal leiomyomatosis, have also been reported after morcellation (18). It should be kept in mind that morcellation significantly increases the risk of these benign sequelae compared to the risk of spreading malignancies.

Long-term survival is not favorable in patients with leiomyosarcoma (19). Besides, there are publications supporting the idea that morcellation can worsen the stage and negatively affect survival in the presence of malignancy (5,20,21). In a study evaluating the effect of morcellation on survival, in the "no morcellation" group, 1-year mortality rate was 5.3% and in the morcellation group this rate was 18.2% (20). Patients who were diagnosed with stage 1 sarcoma or smooth muscle tumors of uncertain malignant potential during initial surgery were operated after a median of 33 days (22) and widespread peritoneal disease was found in 28% and 25% of the patients, respectively (22). Although the studies are retrospective, it was found that hysterotomy affects survival negatively compared to intact hysterectomy. In morcellated sarcoma cases, the risk of abdominopelvic spread increased significantly (44% vs 12.9%) and survival decreased significantly compared to those in whom morcellation was not performed (5). In another case series, the 1-year mortality rate was found to be significantly higher in the morcellation group (20). Result

of a meta-analysis also supported increased risk of recurrence and death (21).

After morcellation, integrity of the specimen is damaged and this can cause both difficulty in pathological examination and it may also adversely affect the diagnosis and staging procedures (23).

In-bag morcellation

In order to prevent or reduce the adverse effects of intraperitoneal uncontained morcellation, morcellation in a closed peritoneal space has been suggested as a possible solution. The most popular method is in-bag morcellation but its potential for avoiding harmful effects and superiority over other morcellation strategies is unconfirmed and needs to be studied further. In addition, there is no consensus opinion from interested societies that in-bag morcellation will prevent morcellation related complications.

In studies evaluating a limited number of patients, tissue or dye leakage or spreading out of the bag were observed in 9-33% of the cases when morcellation was performed in the bag (24,25). Some of the leaks, perhaps, represent microscopic spread. However, there is no data yet on whether this will affect survival in case of malignancy.

Preoperative sarcoma diagnosis

Risk factors include age, history of pelvic irradiation, tamoxifen usage, genetic syndromes (i.e. hereditary leiomyomatosis and renal cell cancer mutation, Lynch syndrome) and history of retinoblastoma in childhood and have been shown to increase the risk of sarcoma (9). If the lesion shows rapid growth in a 3-month period (exact clinical and radiological criteria have not been determined), and especially if there is a lesion greater than 8 cm in the menopausal period, or lesions with central necrosis, heterogeneous appearance, non-calcified cystic degeneration and irregular high blood supply may arouse suspicion of sarcoma (9). However, none of these criteria is effective enough to establish a definitive preoperative diagnosis (26).

Preoperative endometrial biopsy

Although it is an effective diagnostic method in the diagnosis of endometrial pathologies, the effectiveness of endometrial biopsy in the diagnosis of uterine sarcomas is low. It was shown that endometrial biopsy identified only 36% of leiomyosarcomas in submucous lesions (27). In the diagnosis of endometrial stromal sarcoma, the sensitivity was 33% (28). Localization of lesions can vary significantly, and therefore endometrial biopsy is not considered as a useful preoperative diagnostic test in these lesions. Besides, in asymptomatic women, no benefit has been shown. However, endometrial biopsy can help clinicians in patients with preoperative abnormal uterine bleeding.

Imaging methods

Ultrasonography is the first and most frequently used radiological method but differential diagnosis between leiomyoma and sarcoma cannot be made always (29,30). In color doppler, atypical vessel pattern, low resistance index and high systolic velocity is observed in sarcomas (31). However, depending on variables, such as location of the lesion, menopausal status of the patient, and size of the lesion, ultrasonographic features of sarcoma and leiomyomas may overlap and are not distinctive in the majority of cases.

Magnetic resonance imaging (MRI) may show more diagnostic accuracy in differentiation of leiomyoma and sarcoma (13,32). Features such as necrosis, rapid growth, intense contrast enhancement, and restriction on diffusion-weighted imaging can ease the diagnosis and help to differentiate sarcomas from leiomyomas. However, specificity and positive predictive value are low (32). If diffusion-weighted images and contrast imaging are used, discrimination can increase (33). However, despite studies that reported diagnostic efficacy as 88% with these methods (34), some studies have reported imprecision in the successful distinction of fibroid and sarcoma (33). Therefore, the role of MRI should be evaluated in further studies involving more patients. Also, MRI is not recommended for routine use in all lesions and should be used after ultrasonography, in the presence of clinical suspicion (13).

Computed tomography and a positron emission tomography scan are not helpful to discriminate leiomyoma and sarcoma, and they should not be used pre-operatively, solely for this purpose.

Biochemical markers

It is thought that elevated levels of lactate dehydrogenase (LDH) may serve as an indicator of necrosis in the tumoral tissue and invasion into the intravascular area in the presence of sarcoma. Studies have shown that increased levels of LDH are significantly more frequent in the sarcoma group than in the leiomyoma group (35). In one study, the diagnostic success rate of a combination of LDH and MRI was reported to be 100% (32). Success in evaluations with LDH subtypes was also reported, investigating LDH isozyme type 1 and 3 (32,35). In a study using receiver operating characteristic curve analysis for the prediction of sarcoma in the pre-operative period, the optimum cut-off value for LDH was 279.0 U/L (36). However, further studies are needed to confirm the utility of LDH in differential diagnosis.

Intraoperative management

Some characteristics of uterine lesions raise suspicion for sarcoma intraoperatively. These are: no clear mass borders like leiomyoma; no bulging during uterine incision; soft, homogenous, yellowish appearance; and increased tissue fragility. However, these features may also present in patients with degenerative myoma or after use of pre-operative hormonal treatment. In advanced stages, sarcomas may lead to overgrowth and local invasion to adjacent organs (e.g bladder, rectum). Intraoperative frozen-section analysis does not have much efficacy and diagnostic accuracy was reported as only 11-38% (37).

Postoperative management

A uniform clinical management plan for patients with morcellated uterine sarcoma does not exist. Several authors have advised completion of surgery with hysterectomy in case of myomectomy and the abdominal cavity can be evaluated for the presence of metastatic implants (38). Also, patients with late surgical (>30 days) re-exploration had a higher mortality rate (39).

Opinions and suggestions

Since uterine sarcomas are rare and most of the available data are based on retrospective studies, it is difficult to provide certain and conclusive suggestions. The following opinions and suggestions are presented in line with the available data. The following statements can be potentially modified or altered as per new evidence.

1. There is no method that can definitively differentiate sarcomas pre-operatively in patients who are going to be operated with a preliminary diagnosis of uterine myoma.
2. Uterine sarcomas usually occur in women of advanced age, but there is no exact age limit. Especially in patients aged >35 years who are being considered for morcellation, it is recommended that the risk factors should be investigated, that the patient should be examined with advanced imaging methods in case of suspicion, and the necessary precautions should be taken to prevent peritoneal contamination in case of intraoperative suspicion.
3. Ultrasonography is the recommended first-line imaging method. Routine MRI is not recommended for every pre-operative patient and should be performed when malignancy is suspected. A pre-operative endometrial biopsy may only be useful in patients with abnormal uterine bleeding. Its effectiveness in diagnosing sarcomas is very poor.
4. Survival outcomes are worse in uterine sarcomas, even in the early stages, compared to endometrial malignancies. The morcellation of sarcomas can result in disease progression and worsen survival outcomes compared to non-morcellation.
5. The peritoneal seeding resulting from morcellation increases the incidence of benign sequelae. These sequelae account for the vast majority of morcellation-related morbidities and should not be ignored.
6. Although it is assumed that morcellation with tissue containment may be protective against negative outcomes, there is not enough evidence regarding the preventive efficacy of this method. Further studies are needed to establish conclusive data.
7. Patients should be informed in detail regarding the advantages of minimally invasive surgery and the risks of morcellation. In patients in whom malignancy is suspected, morcellation should be avoided (or not performed at all), regardless of the patient's consent.
8. In patients who will be operated with a preliminary diagnosis of uterine myoma, intact removal of the uterus may be primarily considered, depending on the patients' fertility preferences and age.
9. Studies should be designed to determine the efficacy of pre-operative diagnostic methods and the preventive potential of contained morcellation techniques. These studies should aim for the inclusion of as many centers as possible due to the low prevalence of the disease.

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References

1. Langenbeck CJM. Nosology and Therapy of Surgical Illnesses. Gottingen, Germany: 1845.
2. Semm K. Pelviscopy and Hysteroscopy: Color Atlas and Textbook. New York: Schattauer; 1976.
3. Mettler L, Semm K, Lehmann-Willenbrock L, Shah A, Shah P, Sharma R. Comparative evaluation of classical intrafascial-supracervical hysterectomy (CISH) with transuterine mucosal resection as performed by pelviscopy and laparotomy-our first 200 cases. Surg Endosc 1995; 9: 418-23.
4. Perri T, Korach J, Sadetzki S, Oberman B, Fridman E, Ben-Baruch G. Uterine leiomyosarcoma: does the primary surgical procedure matter? Int J Gynecol Cancer 2009; 19: 257-60.
5. Park JY, Park SK, Kim DY, Kim JH, Kim YM, Kim YT, et al. The impact of tumor morcellation during surgery on the prognosis of patients with apparently early uterine leiomyosarcoma. Gynecol Oncol 2011; 122: 255-9.
6. US Food and Drug Administration. Laparoscopic Uterine Power Morcellation in Hysterectomy and Myomectomy: FDA Safety

Communication, c.2104 Apr 17 (cited 2017 July 6) Available from: https://www.fda.gov/MedicalDevices/Safety/AlertsandNotices
7. FDA UPDATED laparoscopic uterine power morcellation in hysterectomy and myomectomy: FDA safety communication FDA, 24 (2014, November) Retrieved from: http://www.fda.gov/MedicalDevices/Safety/AlertsandNotices/ucm424443.htm [13.09.15]
8. Halaska MJ, Haidopoulos D, Guyon F, Morice P, Zapardiel I, Kesic V, et al. European society of gynecological oncology statement on fibroid and uterine morcellation. Int J Gynecol Cancer 2017; 27: 189-92.
9. Brölmann H, Tanos V, Grimbizis G, Ind T, Philips K, van den Bosch T, et al. Options on fibroid morcellation: a literature review. Gynecol Surg 2015; 12: 3-15.
10. AAGL Advancing Minimally Invasive Gynecology Worldwide. AAGL practice report: morcellation during uterine tissue extraction. J Minim Invasive Gynecol 2014; 21: 517-30.
11. Beckmann MW, Juhasz-Böss I, Denschlag D, Gaß P, Dimpfl T, Harter P, et al. Surgical methods for the treatment of uterine fibroids - risk of uterine sarcoma and problems of morcellation: position paper of the DGGG. Geburtshilfe Frauenheilkd 2015; 75: 148-64.
12. Statement of the Society of Gynecologic Oncology to the Food and Drug Administration's Obstetrics and Gynecology Medical Devices Advisory Committee Concerning Safety of Laparoscopic Power Morcellation. SGO,2014, July. [13.09.15] Available from: https://www.sgo.org/wp-content/uploads/2014/04/SGO-Testimony-to-FDA-on-Power-Morcellation-FINAL.pdf
13. Rousseau M, Morel A, Dechoux S, Bouet PE, Catala L, Lefebvre Lacoeuille C, et al. Can the risks associated with uterine sarcoma morcellation really be prevented? Overview of the role of uterine morcellation in 2018. J Gynecol Obstet Hum Reprod 2018; 47: 341-9.
14. Pritts EA, Vanness DJ, Berek JS, Parker W, Feinberg R, Feinberg J, et al. The prevalence of occult leiomyosarcoma at surgery for presumed uterine fibroids: a meta-analysis. Gynecol Surg 2015; 12: 165-77.
15. Tan-Kim J, Hartzell KA, Reinsch CS, O'Day CH, Kennedy JS, Menefee SA, et al. Uterine sarcomas and parasitic myomas after laparoscopic hysterectomy with power morcellation. Am J Obstet Gynecol 2015; 212: 594.e1-594.e10.
16. Topdagi Yilmaz EP, Cimilli Senocak GN, Topdagi YE, Aynaoglu Yildiz G, Kumtepe Y. Incidence of occult malignancies identified during hysterectomies performed for benign indications. J Gynecol Obstet Hum Reprod 2020; 49: 101620.
17. Yorgancı A, Meydanlı MM, Kadıoğlu N, Taşkın S, Kayıkçıoğlu F, Altın D, et al. Incidence and outcome of occult uterine sarcoma: a multi-centre study of 18604 operations performed for presumed uterine leiomyoma. J Gynecol Obstet Hum Reprod 2020; 49: 101631.
18. Cohen A, Tulandi T. Long-term sequelae of unconfined morcellation during laparoscopic gynecological surgery. Maturitas 2017; 97: 1-5.
19. Tasci T, Karalok A, Taskin S, Ureyen I, Kimyon G, Tulek F, et al. Does lymphadenectomy improve survival in uterine leiomyosarcoma? Int J Gynecol Cancer 2015; 25: 1031-6.
20. Raine-Bennett T, Tucker LY, Zaritsky E, Littell RD, Palen T, Neugebauer R, et al. Occult uterine sarcoma and leiomyosarcoma: incidence of and survival associated with morcellation. Obstet Gynecol 2016; 127: 29-39.
21. Bogani G, Cliby WA, Aletti GD. Impact of morcellation on survival outcomes of patients with unexpected uterine leiomyosarcoma: a systematic review and meta-analysis. Gynecol Oncol 2015; 137: 167-72.
22. Oduyebo T, Rauh-Hain AJ, Meserve EE, Seidman MA, Hinchcliff E, George S, et al. The value of re-exploration in patients with inadvertently morcellated uterine sarcoma. Gynecol Oncol 2014; 132: 360-5.
23. Rivard C, Salhadar A, Kenton K. New challenges in detecting, grading, and staging endometrial cancer after uterine morcellation. J Minim Invasive Gynecol 2012; 19: 313-6.
24. Cohen SL, Morris SN, Brown DN, Greenberg JA, Walsh BW, Gargiulo AR, et al. Contained tissue extraction using power morcellation: prospective evaluation of leakage parameters. Am J Obstet Gynecol 2016; 214: 257.e1-257.e6.
25. Solima E, Scagnelli G, Austoni V, Natale A, Bertulessi C, Busacca M, et al. Vaginal uterine morcellation within a specimen containment system: a study of bag integrity. J Minim Invasive Gynecol 2015; 22: 1244-6.
26. Parker WH, Fu YS, Berek JS. Uterine sarcoma in patients operated on for presumed leiomyoma and rapidly growing leiomyoma. Obstet Gynecol 1994; 83: 414-8.
27. Schwartz LB, Diamond MP, Schwartz PE. Leiomyosarcomas: clinical presentation. Am J Obstet Gynecol 1993; 168 (1 Pt 1): 180-3.
28. Bansal N, Herzog TJ, Burke W, Cohen CJ, Wright JD. The utility of preoperative endometrial sampling for the detection of uterine sarcomas. Gynecol Oncol 2008; 110: 43-8.
29. Aviram R, Ochshorn Y, Markovitch O, Fishman A, Cohen I, Altaras MM, et al. Uterine sarcomas versus leiomyomas: gray-scale and Doppler sonographic findings. J Clin Ultrasound 2005; 33: 10-3.
30. Kurjak A, Kupesic S, Shalan H, Jukic S, Kosuta D, Ilijas M. Uterine sarcoma: a report of 10 cases studied by transvaginal color and pulsed Doppler sonography. Gynecol Oncol 1995; 59: 342-6.
31. Bonneau C, Thomassin-Naggara I, Dechoux S, Cortez A, Darai É, Rouzier R. Value of ultrasonography and magnetic resonance imaging for the characterization of uterine mesenchymal tumors. Acta Obstet Gynecol Scand 2014; 93: 261-8.
32. Goto A, Takeuchi S, Sugimura K, Maruo T. Usefulness of Gd-DTPA contrast-enhanced dynamic MRI and serum determination of LDH and its isozymes in the differential diagnosis of leiomyosarcoma from degenerated leiomyoma of the uterus. Int J Gynecol Cancer 2002; 12: 354-61.
33. Thomassin-Naggara I, Dechoux S, Bonneau C, Morel A, Rouzier R, Carette MF, et al. How to differentiate benign from malignant myometrial tumours using MR imaging. Eur Radiol 2013; 23: 2306-14.
34. Lin G, Yang LY, Huang YT, Ng KK, Ng SH, Ueng SH, et al. Comparison of the diagnostic accuracy of contrast-enhanced MRI and diffusion-weighted MRI in the differentiation between uterine leiomyosarcoma / smooth muscle tumor with uncertain malignant potential and benign leiomyoma. J Magn Reson Imaging 2016; 43: 333-42.
35. Di Cello A, Borelli M, Marra ML, Franzon M, D'Alessandro P, Di Carlo C, et al. A more accurate method to interpret lactate dehydrogenase (LDH) isoenzymes' results in patients with uterine masses. Eur J Obstet Gynecol Reprod Biol 2019; 236: 143-7.
36. Nagai T, Takai Y, Akahori T, Ishida H, Hanaoka T, Uotani T, et al. Novel uterine sarcoma preoperative diagnosis score predicts the need for surgery in patients presenting with a uterine mass. Springerplus 2014; 3: 678.
37. Lynam S, Young L, Morozov V, Rao G, Roque DM. Risk, risk reduction and management of occult malignancy diagnosed after uterine morcellation: a commentary. Womens Health (Lond) 2015; 11: 929-44.
38. Ricci S, Stone RL, Fader AN. Uterine leiomyosarcoma: epidemiology, contemporary treatment strategies and the impact of uterine morcellation. Gynecol Oncol 2017; 145: 208-16.
39. Graebe K, Garcia-Soto A, Aziz M, Valarezo V, Heller PB, Tchabo N, et al. Incidental power morcellation of malignancy: a retrospective cohort study. Gynecol Oncol 2015; 136: 274-7.

“Hey Siri! Perform a type 3 hysterectomy. Please watch out for the ureter!” What is autonomous surgery and what are the latest developments?

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Abstract

As a result of major advances in deep learning algorithms and computer processing power, there have been important developments in the fields of medicine and robotics. Although fully autonomous surgery systems where human impact will be minimized are still a long way off, systems with partial autonomy have gradually entered clinical use. In this review, articles on autonomous surgery classified and summarized, with the aim of informing the reader about questions such as “What is autonomic surgery?” and in which areas studies are progressing. (J Turk Ger Gynecol Assoc 2021; 22: 58-70)

Keywords: Autonomous surgery, robotic surgery, machine learning, skill learning, skill analysis

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Introduction

“Come on! Self-driving automobiles are okay, but autonomous surgery? Impossible!”

“Sooner or later, this dream will come true too, I'm sure ...”

“Oh God! Are we going to be unemployed?”

In the face of revolutionary technological developments, people react more or less like the above examples. Weaving workers in England at the dawn of the industrial revolution thought they would be unemployed after the invention of the moving shuttle, which is the key component of automatic looms. Subsequently they committed acts of violence in the form of breaking automatic looms and burning factories, known as the “Luddite Movement”. Despite these impulsive outbreaks, technological development continued and, as a result, a new balance was established between machine and human, and this inter-relationship has continued to develop to the present day. Looking from this perspective, we can think that weaving loom workers and future surgeons may not be too different.

When it comes to changing habits in the face of new products, I always think of our world-famous photographer, “Ara Güler”, who was once quite resistant to digital technology. The following exchange illustrates his attitude to the advances in photographic technology:

- fotograf.net: What do you use as a camera?
- Ara Güler: I can even take pictures with a Singer sewing machine.
- fotograf.net: Do you have a digital camera?
- Ara Güler: I do. I haven't even put my hands on it yet ...

However, years after this interview, according to his assistant, Fatih Aslan, one day he saw a digital camera in the hands of a photojournalist and looked at what he shot: “Awesome! Does this machine give such a result? I want the same right now, Fatih!” From then on until his death, he carried a digital camera with

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him, besides his famous Leica with a 35 mm Summicron lens.

All kinds of changes are painful processes, and no professional group is pleased with its waning importance or the idea of no longer being socially needed...human nature. However, we know historically that professions have changed, disappeared, new professions have emerged and a new equilibrium has been reached each time. Thanks to the autopilot and computer-aided flight assistance systems that have been used in aircraft for a long time and are now considered indispensable, airline transportation has become the most reliable form of travel today. Similarly, worker robots used in Amazon's warehouses have distinct advantages such as high performance, 24-hour working and low error rates compared to human workers. For now, the best approach, taking into account the examples we have given above, is to look at the revolutionary developments in terms of "social benefit" rather than developing resistance. It is also important for interested parties - the surgeons - not to be left out of the process at the development stage.

Autonomous surgery has great similarities with autonomous driving, both in terms of development stages and definitions. Although autonomous surgery seems to be less popular and overshadowed by studies on autonomous driving, it is actually an area where investments and studies have been increasingly made in recent years. When we look at the companies and funds investing in this area, it is also surprising to see that the vast majority of them are the same companies and funds trying to develop autonomous driving. Apart from academic and state-sponsored studies, many international technology companies are also involved in autonomous surgery research. Highlights include Intuitive Surgical, Google, Verb Surgical (a joint venture of Google and Johnson & Johnson), Siemens, Toyota Research Institute, Autodesk, Honda, Intel, Comcast, Hewlett Packard, PhotoNeo, NVidia, National Science Foundation, and National Robotics Initiative.

For this review, a search was performed in PubMed and Institute of Electrical and Electronics Engineers databases with the keywords "surgery, robotics, surgical and medical robotics, skill learning, skill analysis, learning to perceive, machine learning, deep learning" and the articles were classified and summarized in order to give the reader information about questions such as what autonomic surgery actually is, what are the current developments and which areas are progressing.

1. Definitions and concepts

"Autonomous" means to be independent and capable of self decision-making. However, due to the ambiguity in the definitions and the lack of a standard, we encounter inappropriate use of the word in many cases. For example; the master-slave systems, which are now used in many surgical centers and are actually a high-tech motion repeater manipulator, have been mis-named as "robots". This robot terminology, which is actually completely wrong, is unfortunately now in place. Therefore, it would be appropriate to create an autonomous terminology and classification to be used in surgery as soon as possible, in order to prevent a similar situation from occurring for autonomic surgery. The norms determining the degree of autonomy are not yet available for surgical systems, but the classification being used in the automotive industry can be helpful to give an idea. It is likely that future standards for autonomic surgery will be very similar.

The Society of Automotive Engineers standards are as follows:

Level 0: Full control by the human driver; just warnings provided by the machine.

Level 1: Hands are on the steering wheel with full control taken over immediately (hands-on).

Level 2: Hands are not on the steering wheel, but with full control taken over immediately (hands-off).

Level 3: Driving does not have to be monitored, driver may concentrate on anything else, but may take the control at any time (eyes-off).

Level 4: The driver can leave his seat and even sleep (mind-off).

Level 5: Fully autonomous driving, no human contribution required (driverless robotic taxis).

Only Level 0 to 2 systems (driver-assistance) are legally allowed in Europe today.

We do not know exactly how autonomous systems will achieve the maturity and reliability to enter clinical use and into which surgical systems autonomic features will be integrated (robotic laparoscopic devices, endoscopic robots, microbots, bio-inspired robots, etc). Classification according to the developmental order of the post-traditional surgical systems can give an idea about this issue (1):

1st generation (Stereotaxic robots):

- PUMA 200 and Neuromate were developed for use in stereotaxic brain biopsies and tumor excisions as the first examples of this group. Subsequently, systems named SCARA robot, ROBODOC and AcroBot were developed and used for various stereotaxic procedures.
- These systems could not perform multiple consecutive procedural steps and needed the control of a human surgeon after each step. They were more suitable systems for use in tissues and areas with sharp boundaries (such as orthopedics) using mathematical/mechanical strategies. The definition of master/slave appeared during this period.

2nd generation (Endoscopic robots):

- Mainly developed for use in soft tissue surgery and in difficult-to-reach surgical fields.

- The first sample, PROBOT (Imperial College, London) was developed to be used in the excision of pre-defined prostate tissue volumes.

In 1998 The Zeus (Computer Motion, Goleta, CA, USA) and in 2000 da Vinci (Intuitive Surgical Inc, Mountain View, CA, USA) emerged as two of the most famous robotic systems to date. The Zeus was used in Canada for the first beating-heart coronary surgery and in 2001 in the first trans-Atlantic telerobotic operation. In 2003, Intuitive Surgical bought The Zeus company, making da Vinci the only commercial product in this field. Within a relatively short period of time, many disciplines of surgery besides gynecology found a common ground and wide spectrum of applications with the da Vinci system. The limit/range of indications for gynecological operations performed with the da Vinci system also gradually expanded (2).

- Although da Vinci dominates the market as leader, a small number of other second generation robotic systems continue to be offered to the market. Among these, Raven (University of Washington) has made significant progress and has come to the fore as a programmable, modular system with a certain degree of autonomy, having surgical arms with 6-Degrees of Freedom (DOF). Another notable system, DLR MicroSurge (German Aerospace Center), has the advantage of using a common platform with Raven and sharing the same console, allowing surgeons to share teleoperative experiences.

3rd generation (Bio-inspired robots):

- In the development process of minimally invasive surgery, single-port laparoscopy and natural orifice transluminal endoscopic surgery (NOTES) platforms have been developed claiming fewer intervention ports and less scarring. The third generation surgical robots logged in, therefore, with snake-like systems having high articulation numbers to be used in NOTES.

- We can classify existing bio-inspired systems under three headings:

1. Flexible systems working with tendons:

- i-SNAKE (Imperial College, London), CardioArm.

2. Catheter navigation systems:

- Developed for percutaneous cardiovascular interventions. It is divided into two:

- **Mechanical:** Amigo (Catheter Robotics Inc.), Magellan (Hansen Medical Inc.).

- **Electromagnetic:** Niobe (Stereotaxis Inc.), CGCI (Magnetecs Inc.).

These are the robotic systems being guided by changing magnetic fields. Newer systems operating with regular magnetic resonance imaging (MRI) systems (keeping the investment costs at an appropriate level) are also under development.

3. Concentric tube devices:

- Since catheter robots have problems in transferring power to the end region as the length increases, concentric tube systems have been developed in order to overcome this problem. They are used especially in cardiovascular, neurovascular and respiratory system interventions, since they have very suitable structures for tubular organ groups. These systems are also suitable for fetal imaging and surgery during pregnancy due to the advantage of having very small footprint and needle size entry.

4th generation (Microbots):

- These are systems that have been dreamed about since the science fiction movie “Fantastic Journey”, released in 1966.

- They comprise systems ranging from nanometer scale to a few millimeters in size.

- A popular example is the capsule endoscope (PillCam) system that moves passively through the digestive tract by peristaltic organ movement and wirelessly transmits image and data.

- New generation microbots will soon be put into clinical use. Studies continue on improvements in higher image quality, movement, location, telemetry, power, diagnosis and tissue manipulation. Biological systems are closely imitated to achieve these improvements. For example; insect, fish, snake, bacteria and parasite (flagella) movement patterns are being studied for locomotion. There are also highly promising studies on microbots in which locomotion is provided externally by the electromagnetic field of MRI systems (Metin Sitti et al.).

5th generation (systems capable of autonomous decision making):

- Over time, we can expect that some of the surgical methods mentioned above in each of the technological generations will become more prominent, and some will decrease in popularity. Although current studies are mainly conducted on endoscopic robots, autonomous decision-making capability in the future will be able to be integrated into one or more systems that we cannot predict today.

The acceleration of autonomous surgery studies has been made possible by the development of deep learning algorithms. Artificial intelligence (AI) applications in medicine have often started in the form of enhancing human performance with computer systems, but are gradually evolving into systems that make more independent decisions. Successful examples include algorithms that reduce the error rates of pathologists in the diagnosis of cancer-positive lymph nodes from 3.4% to 0.5%, and help breast radiologists and breast surgeons to identify the high-risk group and reduce lumpectomy rates by 30%. While these applications are at the lower end of the spectrum, autonomous systems are located at the advanced stages of the spectrum of AI in medicine.

2. Current developments

When defining autonomic surgery, it is necessary to mention the complex relationship of elements including anatomy, surgical instruments and surgical techniques and also a plethora of subgroups within each element. If we use an analogy with autonomous driving so as to facilitate understanding; anatomy can be compared to the highway and all other environmental factors (buildings, trees, garbage bins, pedestrians, other vehicles, etc), the surgical instrument to the automobile, and surgical technique to the driving techniques and styles. In this analogy, the asymmetrical nature of both anatomy and highway components is immediately apparent. It is the anatomy (highway) component that causes the main difficulties in studies related to autonomous surgery and is responsible for the fact that the fully autonomous surgical systems will be delayed for a long time, even beyond our estimates. While surgical studies in real-time anatomical models are difficult enough, the in vivo situation is even more complex. Additional disadvantages include anatomical variations, respiratory movements, different tissue characteristics, similar textural properties of different organs, internal movements of tissue (positional changes caused by peristalsis), and deformation. For this reason, studies on autonomic surgery, which are still in the early stages of development, have focused and progressed almost entirely on surgical instruments, technical components and performing some predetermined limited subgroup tasks, such as suturing, knot tying, palpation and debridement.

The autonomous robotic systems, which are either at the project stage or under development are as follows (3):

1. The University of California Santa Cruz Surgical Robotics Platform (RAVEN);

- It is a system that uses stereo imaging.
- It has low degree motion planning and ability to detect grasper positions.
- It can perform multilateral debridement of phantom tissue.

2. Da Vinci Research Kit (DVRK);

- It can perform viscoelastic phantom tissue debridement.
- It can perform circular cutting in tissues with variable viscoelasticity.
- It has the ability to dissect in cryogel phantoms with ultrasound guidance.

3. RAVEN-II;

- It has a stereo tracking and near-infrared wavelength (NIR) imaging system.
- It recognizes pseudoneuroblastoma, creates a surgical plan and can perform semi-autonomous surgical ablation.

4. Smart Tissue Anastomosis Robot (STAR), John Hopkins University;

- It is the first system that has made significant progress in solving the problems of tissue movement, deformation and tissue similarity in soft tissue surgery.
- A 7-DOF arm is used, utilizes NIR imaging and has deep learning software for motion planning.
- Suture passing, knot tying and anastomosis features are quite improved. It has recently shown great success in semi-autonomous intestinal anastomosis. It can perform autonomic intestinal anastomosis in ex-vivo and in-vivo animal models, exceeding the performance of human surgeons.
- It can make straight cutting and tumor extraction in experimental animal tissue.

5. Bone drilling in ear surgeries that require precision such as cochlear implant placement.

6. Needle navigation in lung biopsies (Vanderbilt, University of North Carolina).

7. Brain tumor resection (Brain Tool Lab, Duke University);

- Using image data obtained from tomography, ultrasound and MRI systems, the robot arm precisely and accurately takes the appropriate position proper for the tumor tissue.
- The route of the surgical laser incision, control of the firing energy, safe firing and safe tissue ablation are provided using an autonomous algorithm in a closed-loop feedback.

None of the systems listed above are currently Food and Drug Administration (FDA) approved. For information purposes, the FDA can approve any medical device in two ways: Premarket approval (PMA) and 510 (k) permission. PMA covers the permits for completely new products, is costly (costs over $30 million per license) and requires a lengthy process for approval. The 510 (k) authorization includes approvals for improved versions of products that have already received PMA or similar products, is a much lower cost operation (costs less than $200,000) and can be completed in a relatively short time. FDA has not approved any medical device with the definition of a surgical robot to date. The da Vinci robotic system, which is commonly referred to as a robot with a misidentification, but is actually a master-slave system based on the principle of motion repetition, received FDA approval for the equivalent of “non-robotic laparoscopic equipment” in 2000 [via 510 (k)]. However, PMA approval will be required for autonomous surgery systems in the future. Recently an autonomous anesthesia device, named Sedasys by Johnson&Johnson, which can adjust the dose of sedation drug administered intravenously by monitoring parameters such as the patient's breathing, heart rhythm, and saturation, has passed FDA approval (with PMA) (4).

If we think of autonomic surgery as a big and complex problem, trying to solve it by dividing it into its small components is the right method to follow and studies continue in this way. In the near future, we will be able to find new generation robotic

systems, which will facilitate the surgeon's work during the operation, and which will have added some equipment with limited autonomy including the ability to perform certain tasks such as needle grasping at the right angle, suturing, knot tying, surgical ablation, and tumor resection with systems with varying degrees of autonomy.

We can summarize the main areas where the studies are concentrated as follows:

1. Identifying surgical instruments separately (segmentation) and real-time tracking of their movements and paths,
2. Grasping the surgical needle from the right angle and point,
3. Suturing,
4. Knot tying,
5. Biopsy needle guidance and precise bone drilling applications,
6. Real-time measurement of tissue flexibility and deformation, adaptive tissue cutting procedures,
7. Palpation,
8. Ablation, resection, debridement,
9. Evaluation of surgical skills and surgical training,
10. Computer-aided anatomy.

2.1. Identifying surgical instruments separately (segmentation) and real-time tracking of their movements and paths

One of the most critical issues for autonomic surgery is segmentation, which is the ability to identify the instruments in the surgical console separately, both from the surrounding tissues and from each other. In simpler terms, segmentation is the process of the computer program to understand which pixels belong to the living tissue, which pixels belong to the surgical instrument and the region of the instrument while evaluating the surgical area as a picture. This segmentation process is the most critical step in the realization of most autonomous tasks such as suturing, needle catching, knot tying, incision, ablation, and debridement. Real-time monitoring of segmentation and segmented structures are not easy processes due to reasons such as shadowing, specular reflections during surgery, fogging of the camera and lens, visual occlusion with blood and clots, and dynamic and complex changes of the surgical tissue area (5).

Successful segmentation studies have become possible as a result of developments in high-resolution, new generation cameras and deep learning algorithm architectures (U-Net, TernasusNet, LinkNet) (Figure 1).

The next step, after successful segmentation, is the ability to track the instruments in real-time. Real-time instrument tracking is getting faster and more precise, thanks to highly powerful computer processors and deep learning algorithms that successfully determine spatial location. The success rate can be increased further by linking the instruments to be used to machine learning systems and pre-educating them before the operation (6).

A secondary benefit and usage area of autonomous surgical instrument tracking is that the camera can take an autonomous position by tracking the instrument without the need for an assistant (cameraman robot). Some of the advantages of autonomous camera control compared to human assistants can be: cameraman robots do not get distracted, get tired, their stability does not decrease, they can follow better and more precise positions, and since there will be one less person in the operation team, it is ensured that the remaining team has more movement area in the operating room. In recent years, many cameraman robot systems have been introduced and put into use (EndoAssist, Aesop, EvoLap, etc). There are also systems that have human-machine interface differences (foot pedal, eye/head movements or voice control), whose common purpose is to take the task of holding the camera by relaxing the operator's hand. However, these systems do not have instrument segmentation, motion tracking and autonomy capabilities. A laparoscopic system capable of autonomous monitoring has recently been commercially available (AutoLap, Medical Surgery Technologies, Yokneam, Israel). However, there is no published article about the sensitivity, success or deficiencies of this system yet (7).

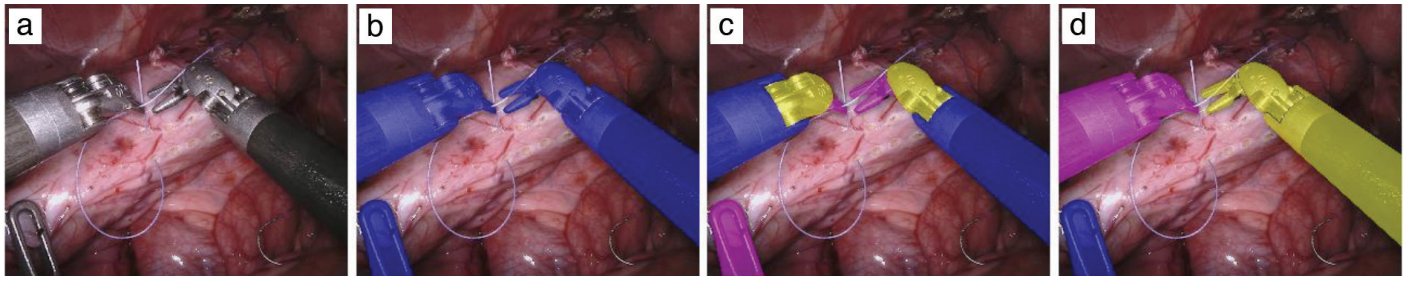

Figure 1. An image section taken during robotic surgery: a) Original image, b) binary segmentation (instruments blue, tissue red), c) multi-fold segmentation (three separate regions of the instrument can be identified: body, articulation joint, grasper tip), d) multiclass segmentation (each instrument can be defined separately) (5)

2.2. Grasping the surgical needle from the right angle and point

Intelligent robotic surgical assistant systems, which are the next stage of master/slave systems and capable of performing some low-level surgical tasks autonomously, are on the horizon. The first of these surgical tasks is to grasp the needle autonomously. In this way, the problem of not grasping the surgical needle at the appropriate point/angle and the prolongation of the surgery time as a result of multiple regrasping attempts can be avoided. Three parameters are considered in surgical needle grasping: Manipulation, dexterity and torque metrics. Each of these parameters is important for a successful needle grasping (8).

Studies on robot gripping/grasping were first developed with the aim of facilitating the daily life activities of disabled people with motor deficits, long before being co-opted into autonomic surgery. While the first developed systems work with the help of comprehension databases, prepared with previously 3-D modeled objects (glass, fork, ball, etc), with the advancements in deep learning algorithms, successful grasping/capturing operations can be accomplished with machine learning today without the need for any preformed database (9).

Studies on grasping the surgical needle proceed on similar principles. The critical steps are the segmentation and movement planning of the tail, body and tip parts of the needle (10,11) (Figure 2, 3).

Various visual-based algorithms are used in robot capture studies. However, these algorithms are usually not suitable for surgery as autonomous robotic capture is largely unpredictable. In such cases, the surgeon's choice among multiple capture points and different capture scenarios has emerged as an acceptable method. More successful systems can be developed after the data pool created by these choices in a cloud system is evaluated with deep learning (9).

2.3. Suturing

One of the most time consuming procedures in robotic, minimally-invasive surgery is suturing, due to the lack of haptic (touch sensitive) feedback, limited range of motion, and narrow-angle field of view (12). After determining the basic elements of suturing (needle entry/exit points and depth of transition), autonomous needle capturing/grasping, suturing and knot tying will shorten the duration of the surgery, as well as causing less tiredness for the surgeon, less distraction and healthy planning of the next surgical stage. We will soon see some limited tasks added to the new generation of robotic surgical equipment, which we can call "semi-autonomous" or "restricted-authorized autonomy" (12) (Figure 4).

There are three components required for a successful suturing process: needle, surgical thread and tissue. These three components should be defined separately by segmentation

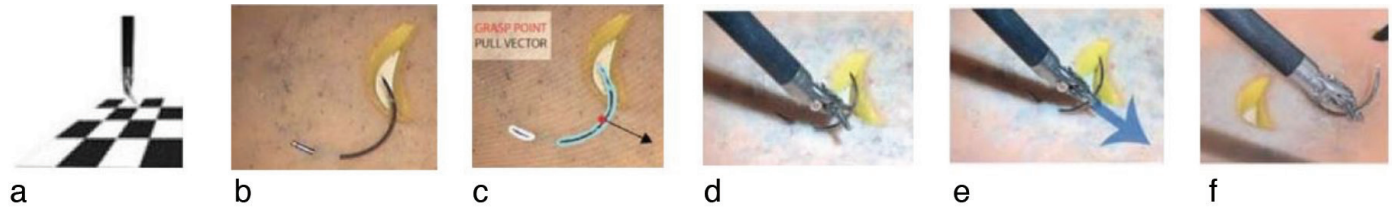

Figure 2. Steps in the process of grasping a surgical needle stuck into a tissue phantom at an appropriate angle and extracting it from the tissue by calculating the tensile force vector. a) Calibration, b) snapshot image creation, c) planning the segmentation and extraction phase, d) approaching phase, e) withdrawal phase, f) complete removal of the needle from the tissue (10)

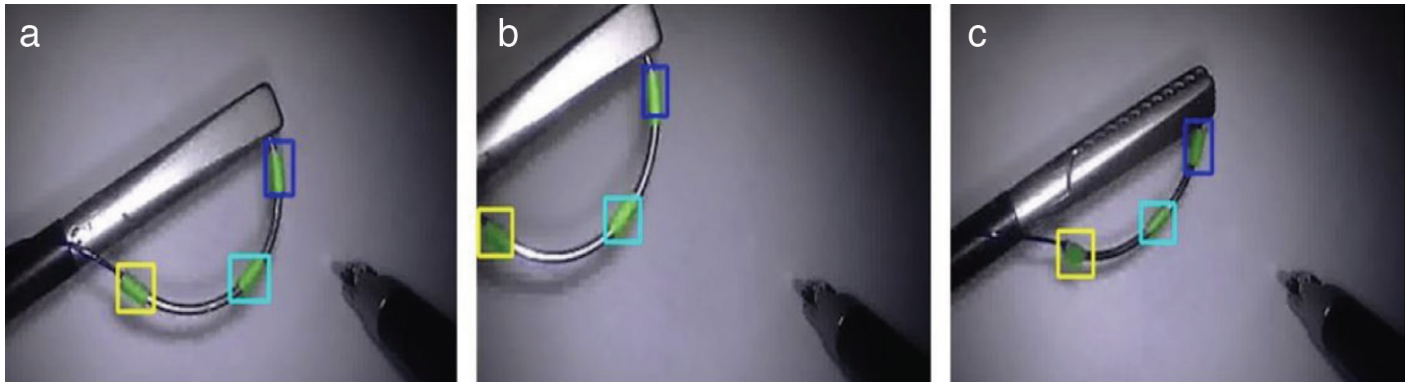

Figure 3. The segmentation regions (tail, body and tip) determined by the tracking algorithm that adapts to changing positions autonomously. a) Ideal position, b) position at the border of the endoscope's visual field, c) the position at which the needle angle changes in the spatial plane. Segmentation of the needle was successful in all three cases (11)

and their real-time movements should be followed in terms of both each individual component and the inter-relationship with the other two components. We have covered segmentation, and the grasping of the surgical needle in the previous section. Identifying the thread along its entire visible length in the surgical area and following it in real-time is essential for both suturing and the next step, knotting. Various methods have been proposed to identify and follow-up the suture material. Some studies have reported success with RGB plus imaging, which provides a high level of depth information, but RGB plus is not usually available in systems used today. In recent studies, stereo imaging, which is already widely used in existing systems and is starting to become standard, has been used and successful results have been obtained. Non-Uniform Rational B-Spline, a modeling algorithm for non-linear curves, can perform real-time suture thread identification and tracking by analyzing the stereo image pair used in existing systems (13) (Figure 5).

The first example of successful suturing in living tissue is STAR, developed by John Hopkins University. It consists of a needle system activated by a modified Endo360 attached to a 7-DOF arm. Using NIRF and multispectral vision systems, it can perform the segmentation process in real-time. In its first results, published in 2014, it was reported that STAR performed intestinal anastomosis with acceptable accuracy (<0.5 mm) and deviation rates (0.2 mm). Subsequent studies continued on both 3-D models and live tissue, and successful preliminary results were published (14) (Figure 6).

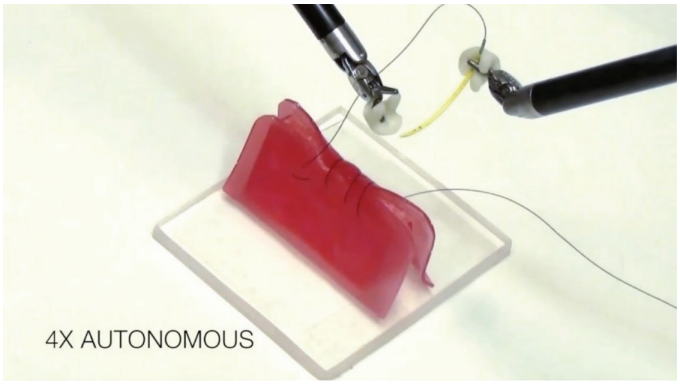

Figure 4. Fully autonomous suturing (needle grasping, needle transfer between instruments, suturing and knot tying) performed on a gel phantom after determining the starting point, distance between sutures and tissue depth (12)

2.4. Knotting

Surgical knotting is a sequential procedure by rotating the suture thread held with instrument A around instrument B, then grasping the free suture tip with instrument B, and tightening the knot by pulling both instruments in opposite directions, as shown in Figure 7. This sequential task, which seems quite simple, is one of the reasons for the prolongation of the operation time in minimally invasive surgery. Difficulties during knotting are related to folding of the suture thread, narrow working area, limited depth perception, inability to feel the correct pulling force to be applied and insufficient experience. Studies on this subject can be divided roughly into two groups: new instrument designs that facilitate knotting and autonomous knotting algorithms using already existing instruments. The issue of new instrument designs, which are not widespread and far from developing a standard yet, will not be addressed here (15) (Figure 7).

Most of the studies were done with stereo cameras and two or three arms having at least 3-DOF, with standard grasper tips. Autonomous knot tying begins with the determination of the spatial planes of the needle entry and exit points and the step of removing the needle from the tissue and pulling the suture. Surgical arms, which are placed in opposite positions close to the needle entry and exit sites, are ready for the next step, ring formation. Often the distal end of the suture is left at a length of 25-35 mm. Two different methods can be used to wrap the suture on the opposite arm to form a ring: Spiral ring creation (slower) or rolling-arc method (faster). The knot is completed

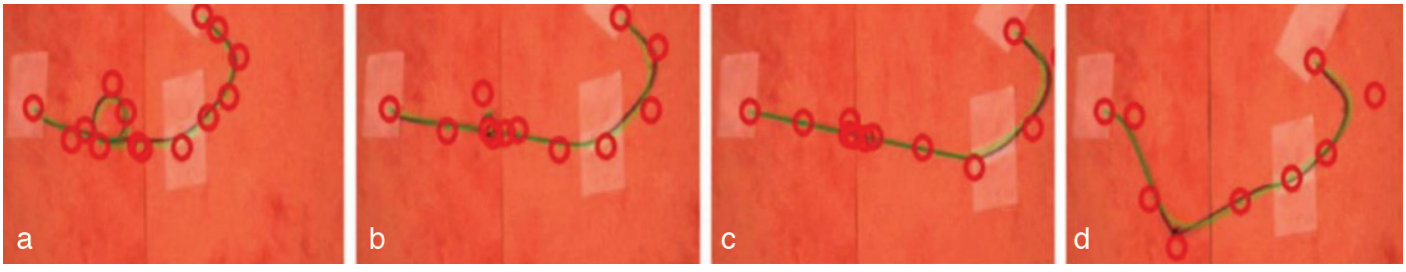

Figure 5. Identification of the suture thread along the entire length by segmentation and additionally determining the control points for motion tracking (tracking the dynamic relationship of the changing points in respect to each other is especially important during the knot tying phase). a) Multiple control points along the suture length, b) as a knot is tightened, the control points get closer and closer, c) after sufficient knot tightening, the knot with multiple points is redefined after a while, d) as a single control point (13)

by grasping the distal end and pulling it mutually, after forming a suitable ring (15).

There are some studies on the use of skill transfer from humans which is a new learning method, mostly used in the field of rehabilitation robotics. Together with successful studies on this skill transfer with machine learning methods, which learns by analyzing the human surgeon's movements in detail, a faster development of autonomic surgery may occur (16).

2.5. Biopsy needle guidance and precise bone drilling applications

Semi-autonomous biopsy needle guidance studies find use in three main areas: ablation, biopsy and brachytherapy. The common goal here can be summarized as minimizing tissue damage with increased sensitivity and smart maneuvers. Neuro-interventional procedures and spinal surgical procedures that require precise positioning and orientation, working with similar principles, are also clinical uses for these robotic systems. Clinical applications include ultrasound, MRI or luminous coherent tomography-guided interventions, cochlear implantation, motion compensation and orthopedic/neurological/radiosurgery robots. "MiniAture Robot for Surgical" procedures, Mazor Surgical Technologies, Israel, which provides instrument positioning and orientation in spinal surgery using a 6-DOF arm, SpineAssist robot used in spinal fusion surgery and precision percutaneous pedicle screwing, NeuroArm used under MRI guidance (University of Calgary, Canada) are examples of these systems (17-21).

2.6. Real-time measurement of tissue flexibility and deformation, adaptive tissue cutting procedures

In surgery, almost all tissues except bone structures are deformable. Among the studies on deformable objects, studies of surgical cutting procedures in virtual environment have been especially prominent in recent years. Deformation calculations are used not only in cutting processes, but also in stereotaxic biopsies and suturing, in which the needle should pass through the correct line and exit from the correct point (Figure 8).

In order to solve the problem of tissue flexibility and deformation, we should consider three consecutive steps:

1. Modeling the textures with different material properties,
2. Analysis of traction and counter-traction responses,
3. Updating the new geometry and topology of the changed model after each cutting process.

In order for the tissue procedures to be suitable for real-time surgery, they must have 30 Hz refresh rates visually and 1,000 Hz as haptic feedback. In other words, the three steps mentioned above should be calculated over and over again each time and these calculations should be repeated at least 30 times per second in order to create a real-time perception. For this purpose, very high processing power is required. Studies often were done on mesh sheets, first in 2-dimensional models, then 3-dimensional models using multi-layered mesh. However, methods and hybrid models that do not use mesh have been recently studied. These are methods commonly referred to as point-based approaches. The precision and accuracy rates

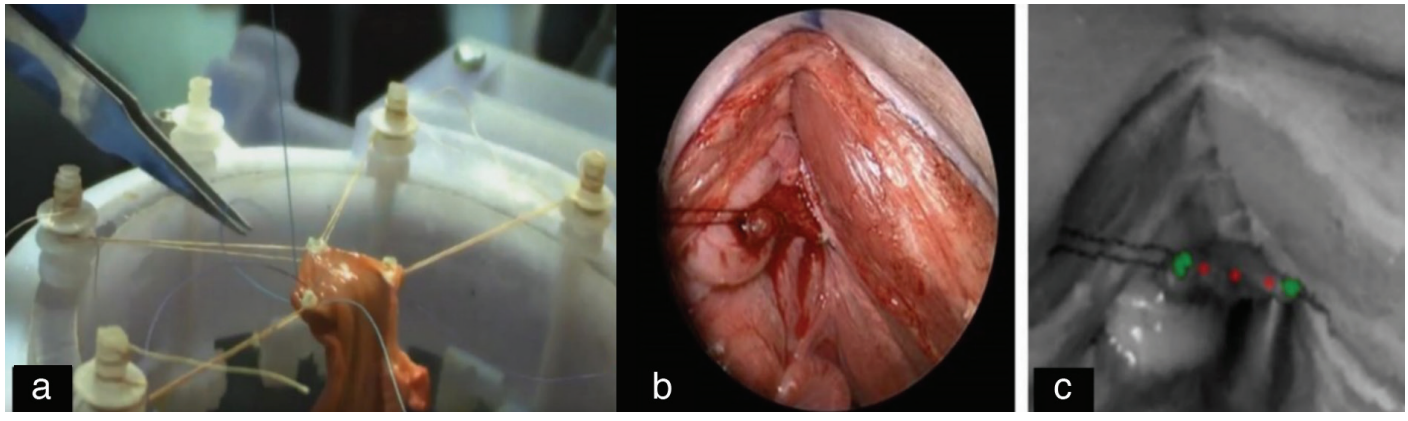

Figure 6. STAR procedures; a) intestinal anastomosis in ex-vivo pig tissue, b) in-vivo vaginal cuff (RGB camera view) after hysterectomy of pig, c) plenoptic camera view with the markings of the beginning, end and suture transition points (vaginal cuff closure was completed in about five minutes) (14)

STAR: Smart Tissue Anastomosis Robot

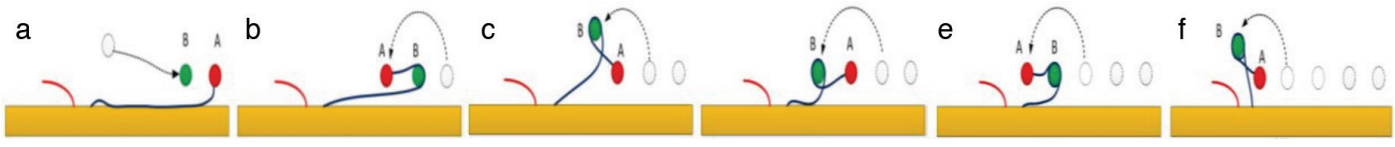

Figure 7. Trajectory of rolling arc looping (15)

are significantly higher in point-based approaches than mesh models, especially in models with large deformations, but require proportionately higher processor power (22).

2.7. Palpation

Palpation is one of the main components of the physical examination. It is defined as the feeling and differentiation of parts with different degrees of hardness from the normal tissue, such as lymph nodes, nodules, and masses. Such lesions are the most common signs and symptoms encountered in the diagnosis of some diseases. Today, many robotic systems have force and position feedback features to different degrees and sensitivities. There are experimental systems used in the diagnosis of breast tumors, thyroid nodules and for determining the precise location of kidney stones during laparoscopy (17).

2.8. Ablation, resection, debridement

We mentioned in previous sections projects designed to perform surgical tasks such as ablation, resection and debridement with certain degrees of autonomy. RAVEN, which can perform multilateral debridement in phantom tissue, and DVRK, which can determine circular targets in viscoelastic tissue phantom, perform circular cutting and ultrasound-guided resection in cryogel phantom are the leading projects in this field. The drawbacks of these projects are that they have not been tested in animal tissues and they are limited to single-pass mechanical cutting. The problems of tissue motion, deformation and structural similarity in living tissues could not be solved in either of the projects. STAR is the first soft tissue surgery system that has found satisfactory answers to these problems. Using plenoptic and NIR imaging, the STAR platform can overcome the deformation problem by calculating metric co-ordinates in living tissue when marked with biocompatible markers. In a study using STAR and electrocautery, it was reported that electrical power, cutting speed and cutting depth were autonomously and precisely adjusted. Successful resection with STAR was also reported in the same study when the margin of resection of tumor tissue stored in pig living tissue was marked with biomarkers (21).

2.9. Evaluation of surgical skills and surgical training

"I climb the steepest mountains, jump over the high cliffs, dive deep into cold waters ..."

Everyone is a very good surgeon! - from the rookie assistant who has just completed his first month, to our seniors who are at retirement age. The lack of a system that objectively measures surgical skills is a large deficit here. The accepted standard today in the evaluation of surgical skills is peer review of either live surgery or video images during traditional surgery or retrospectively by a peer or more senior surgeon. However, there are question marks about the objectivity and accuracy/sensitivity issues that depend on human evaluators. The first studies to overcome this problem have been tried with surgical scenarios built on virtual reality systems. However, although it is indisputable that virtual surgery systems are very useful in education and gaining some skills, there are opinions that they cannot be assessed with the same criteria when it comes to surgical skill measurement.

A new field of the objective measurement of surgical skills was developed during studies of deep learning in medicine. Developing autonomous skills and teaching/transferring surgical experiences to deep learning systems have shown promising results. Since it is possible to convert and analyze video recording and quantitative motion data into descriptive mathematical models in robotic surgery, these data can also be used for surgical training and extraction of automatic performance metrics (OPM). JIGSAWS (skill assessment set developed by John Hopkins University), a robotic surgery database open to everyone, is a system based on scoring of three parameters: Suturing, needle threading and knot tying. The JIGSAWS database was created by assessing these three parameters in a total of eight surgeons with different experience levels. Many parameters such as the spatial position (x, y, z), tracking of the surgical instrument tip, its rotation, linear velocities, angular velocities and the angle of the gripper tip are

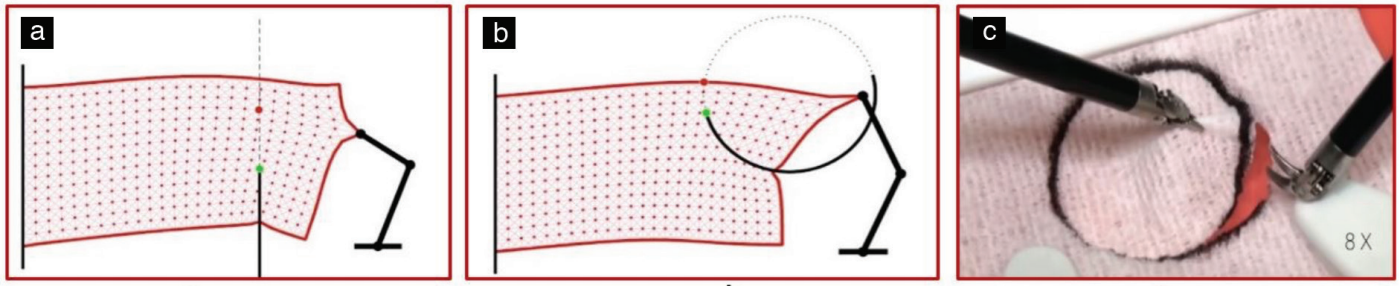

Figure 8. Autonomously calculated traction and counter-traction to overcome the problem of significant tissue deformation. a) Directing the biopsy needle tip towards the target, b) adjusting the needle exit point, c) completing the circular cutting process with high precision (21)

recorded as kinematic motion data in the JIGSAWS database (23) (Figure 9).

Studies in the field of measuring surgical skills can be divided into two main categories:

1. Descriptive statistical analysis: Skill analysis is performed using movement time, length of the path taken, motion pollution, curvature, semantic signs, instrument orientation, and vectoral force evaluations. However, very intensive manual engineering effort is required to properly arrange such optimal skill metrics. Another problem encountered here is the open-ended discussion about what is the best skill.

2. Predictive model-based methods: This is based on the principle of predicting skill measurement from the motion database. In itself it is divided into two parts; a) Descriptive and b) generative (machine learning). Recent studies have focused on the latter, the generative predictive model-based method. This model works on the basis of the analysis of OPM data (action data such as instrument and endoscopic camera motion tracking, energy modality usage times, etc) collected in live surgery with the da Vinci System's recorder (dVLogger; Intuitive Surgical Inc) with machine learning algorithms. The data obtained here are extremely extensive and often contain details that can be missed by human observers.

One of the important differences between machine learning and traditional methods is the need for a reference for comparisons in traditional analysis. However, even among expert surgeons, there is often no consensus on what "good surgery" should be used as a reference and standard. There is no need for such a reference system in machine learning systems. Here, instead of a pre-built model, there are surgical templates shaped and learned with data itself. When the clinical outcomes of the patients are also added to the performance metrics in a machine-learning algorithm, such as intraoperative complications, postoperative complaints/findings, and length of hospital stay, a more objective evaluation can be obtained (24).

If we examine a system based on the machine learning principle as an example, the subject might be a little clearer. In this example, the basic setup of a machine learning system for robotic radical prostatectomy operation is given. By using only the determined OPMs, the length of stay of the patient can be predicted with an accuracy rate of 87.2%. When some extra data concerning the patient are added (age, body mass index, prostate specific antigen level, prostate volume), the accuracy rate increased up to 88.5%. There is an ongoing study to determine the effectiveness of this model in predicting oncological and functional outcomes in the future and this will be possible after enough clinical data have been obtained over time. In this study, some different and unpredicted results were also obtained, which even experienced surgeons may not be aware of. For example; while the view that a balanced use of both hands is ideal for good surgery is widely accepted, this study found that master-level surgeons use their dominant hands more than novice surgeons (25) (Figure 10).

2.10. Computer-aided anatomy

Autonomic surgery will only be possible after the development of computer-assisted anatomy. Anatomy still remains a difficult problem to model digitally, not only because of the spatial volumes of highly complex physical structures but also because of their functional relationships with each other. Pioneering studies on this subject started with the studies of matching topographic anatomical information with radiological images by radiologists. Thanks to the developed algorithms, the location and segmentation of complex organs such as the pancreas, has recently become possible. There are studies in digital anatomy-related diagnoses and treatment assistance, radiotherapy planning, surgical simulation and estimation of tissue damage severity. In parallel with the widespread use of tomography and MRI, the number of such studies is also increasing (a study showing that studies involving multi-organ analyzes increased parallel to the number of tomography/MRI tests in the UK between 1995 and 2013).

The developments in this field will accelerate with the development and widespread use of 3-D modalities in medical imaging, the introduction of low artifact and high-resolution

Figure 9. a) Suturing, b) needle passing, c) knotting tasks (parameters evaluated and reported by JIGSAWS) (22)

systems, and algorithms that make anatomically-consistent single organ and system segmentations.

This review will not go into details about the anatomical models, which is a large subject in its own right. Among these, 3-D MRI and probability calculations using deep learning algorithms and the generation of temporal and spatial data from 2-D MR and tomography sections stand out as promising methods, especially in multi-organ analyses (26). When anatomical studies are performed by dividing the body into regions, the most difficult region to analyze is the abdominal area. The reasons for this include the existence of more complex relationships between organs, peristaltic movements of the digestive system, respiratory movements, and changing anatomical relationships depending on factors such as age, disease and edema. Since deep-learning systems can process high-density data regarding abdominal anatomy more easily with increased processor speeds, there had been a significant increase in the number of publications in this area. When we look at the distribution of the studies by anatomical regions, the frequency of publications are as follows: the abdomen 24%, chest 8%, vertebrae 7%, pelvic organs (bladder, rectum, prostate, vagina) 7%, hand 3%, hip joint 3%, knee 2%, and elbow 1%. There are two freely accessible anatomical image databases (Tomography, MR, positron emission tomography) to be used in digital anatomy studies: National Institute of Health Cancer Imaging Archive and UK (United Kingdom) Biobank Imaging Study. It is an important advantage that these databases include normal anatomy as well as non-normal anatomy (cancerous organs, displacement of the remaining organs after surgery, etc) (18,27).

3. Ethical and legal issues

Although technical developments related to autonomic surgery are still in the very early stages, disproportionately intense ethical discussions are reported in the literature. Why is AI in surgery, which is still in its infancy, heavily discussed ethically and legally? We are not in a hurry, right? Actually, we are. Before the systems are put into use, ethical discussions should be completed and legal regulations should be started. While longer time is needed for the development of fully autonomous surgical systems, hybrid systems are on the horizon. A road map study was carried out in Geneva in 2006 by European Robotics Research Network and attention was drawn to these issues (28).

Currently, three areas where ethical debates are particularly heated are nuclear physics, genetic engineering and autonomous robotic systems. The first scientific symposium on robotics was held in 2004 and the World Robot Declaration, defining the basic features that a robotic system should have, was published. Accordingly, a robot;

1. should work on the basis of partnership with people,
2. should help people physically and psychosocially,
3. should contribute to the realization of a safe and peaceful society.

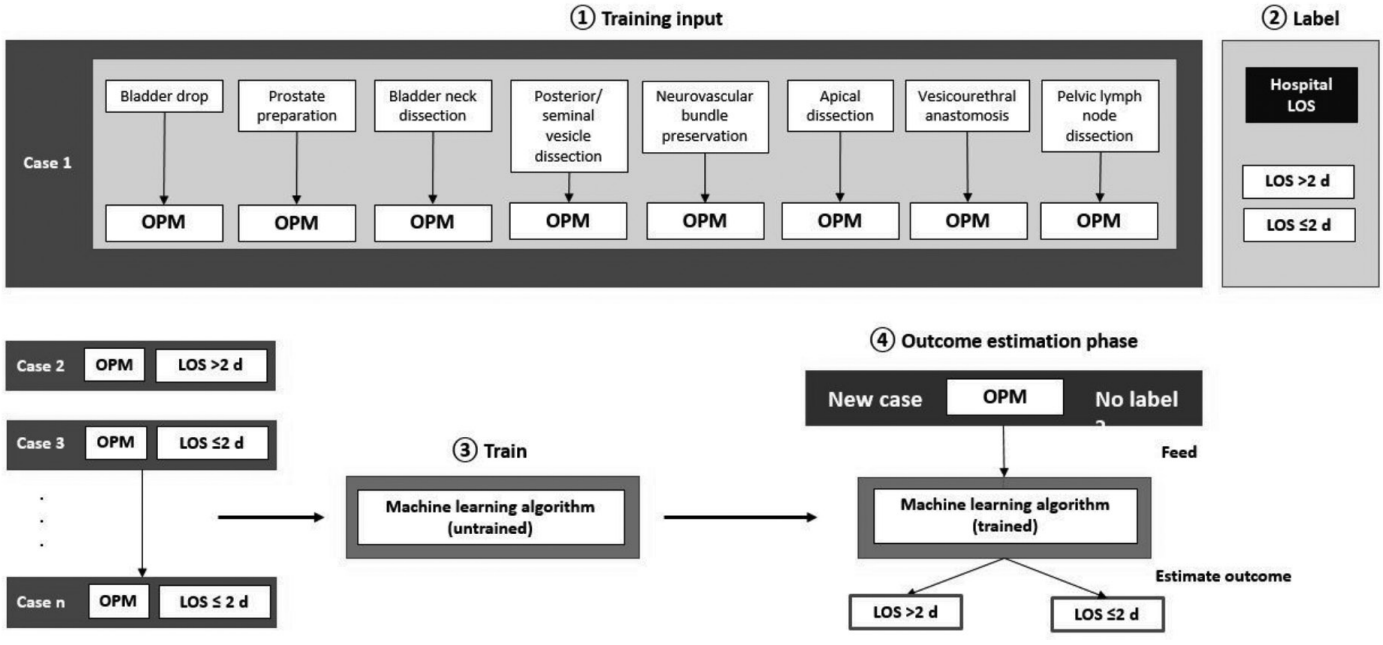

Figure 10. A machine learning model in which surgical performance in the surgical treatment of prostate cancer is measured with OPM and its effects on hospital stay are predicted (24)

OPM: Automated performance metrics

The vast majority of the publications regarding the ethical issues of AI and autonomous systems are related to medicine (e.g. surgical robots), civilian vehicles (e.g. autonomous cars) and military technologies (e.g. armed drones, targeting systems). Discussions and recommendations share similarities for all three areas.

The main discussion in the aforementioned systems is within the framework of "responsibility" (29). Responsibility has three elements:

1. Accountability: The ability to explain each transaction made. In the series of operations consisting of three parts, which are input, internal state (deep learning algorithm) and output, there is often no full transparency, especially concerning algorithms, for reasons such as the "black box" architecture specific to deep learning systems, and also non-disclosure of source codes because of trade secrets and copyright laws.

2. Liability: Who is primarily responsible for the system - manufacturer, operator or maintenance service provider?

3. Culpability: Who is guilty and should be punished in the event of a malfunction, such as an interruption of telecommunication during telesurgery? Again the candidates would be the manufacturer, operator or maintenance service provider.

As emphasized in Matthias' (30) article and the European Parliament Resolution of 16 February 2017 on the rules of civil laws in robotics, there is great unpredictability in adaptive autonomous robots that may cause damage and the traditional laws of culpability would be unsuitable in such situations. One of the suggested solutions for liability is similar to that in autonomous cars, the surgeon sitting in the same room where the system operates, both as an observer and as a safety provider who can take full control any time. Another option would be to use a restricted system that only helps and assists the surgeon in routine operations, without giving full autonomy. Instead of "automatic machine learning" (uninterpretable, internal functioning, black box architecture), which is one of the discussion topics, "interactive machine learning" stands out as a more acceptable system in autonomous surgery. In the interactive machine learning system, the process of how the end-result is achieved can be monitored transparently, contributing also to the professional development of surgeons.

4. Justification of why a discovery is needed should be presented to the society for all inventions and developments with huge potential that will directly affect the society and will enter all our lives irreversibly. While the justification is fairly easy and understandable in some cases (such as laparoscopic vs conventional surgery), in some cases it can be really difficult (there is still an intense debate about the need and consequences of armed drones used in the military field). The situation concerning autonomic surgery is also more complicated than expected. Why should autonomic surgery be needed when there are human surgeons? While the debate continues about the need for autonomous systems for routine surgical practice, it is easier to justify why it is needed in some selected cases. Autonomous surgery can be truly life-saving in war zones, large infection outbreaks, space research stations and prolonged space flights. The intubation and resuscitation of seriously ill patients infected with "Severe acute respiratory syndrome" in 2002-2003 and during the ongoing Coronavirus disease-2019 pandemic are procedures that carry great risks for healthcare professionals. Robotic systems with deep learning algorithms that will facilitate teleoperative procedures (e.g. intubation) can be very useful in such cases.

Another issue is the problem of cybersecurity. In 2017 Bonaci et al. (31) published a case of hacking the control of a robotic system during a teleoperation by infiltrating the UK National Health System network. The issue was opened up to discussion and emphasized that security certificates should be redefined and strengthened, in much the same way as in military systems (31).

It is also useful to briefly mention a situation that is currently under discussion for telesurgery (and the same discussion will be held for autonomous systems in the future) called the Tort Law. If a lower performance operation is performed by a human surgeon while there is a possibility of performing a high performance operation with telesurgery or advanced autonomic surgery system by a more experienced surgeon, can the human surgeon be held responsible and blamed for this? It is highly controversial, but it will certainly raise the bar for surgical skills that surgeons must have.

The discussion on ethicolegal regulations and more should be completed before the development of a perfect autonomous surgical system that is completely safe and exceeds human performance.

Conclusion

Autonomous surgery will happen sooner or later, although it may seem far away at present. First of all, we have to accept this. On the other hand, we need to do something in order not to adopt the position of the weaving workers during the Industrial Revolution. While there are dizzying speeds in technology, the relatively slow progress in autonomic surgery is actually an important chance for surgeons to adapt to and influence this process. Advances in AI and deep learning algorithms in non-surgical disciplines are quite advanced, especially in branches such as radiology, pathology, histology, embryology, and dermatology, which are predominantly image-based. AI systems that can evaluate mammography at the expert level, make serious progress in the diagnosis of diabetic retinopathy,

and select the embryo with the highest chance of pregnancy in IVF cycles are rapidly coming into daily use. Most of our colleagues working in these disciplines either remained completely passive during the development phase of these systems or are still unaware of the advanced development stage the process has reached.

It would be an appropriate approach for surgeons to take active roles in the development process and trigger innovative initiatives, rather than passively waiting for the developed technology to be presented to them. The point that should be emphasized again and again is that the basic criterion underlying every development related to AI/deep learning and autonomic surgery in medicine is "social benefit".

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References

1. Ashrafian H, Clancy O, Grover V, Darzi A. The evolution of robotic surgery: surgical and anaesthetic aspects. Br J Anaesth 2017; 119(Suppl 1): i72-84.
2. Takmaz Ö, Gündoğan S, Özbaşlı E, Karabük E, Naki M, Köse F, et al. Laparoscopic assisted robotic myomectomy of a huge myoma; Does robotic surgery change the borders in minimally invasive gynecology? J Turk Ger Gynecol Assoc 2019; 20: 211-2.
3. Camarillo DB, Krummel TM, Salisbury JK Jr. Robotic technology in surgery: past, present, and future. Am J Surg 2004; 188(4A Suppl): 2S-15S.
4. Britton D. Autonomous surgical robots. Life Sciences Innovation: Law 321, 2016. Available from: https://ssrn.com/abstract=2977943
5. Shvets AA, Rakhlin A, Kalinin AA, Iglovikov VI. Automatic Instrument Segmentation in Robot-Assisted Surgery using Deep Learning. 2018 17th IEEE International Conference on Machine Learning and Applications (ICMLA) Orlando, FL; 2018: 624-8.
6. Zhao Z, Chen Z, Voros S, Cheng X. Real-time tracking of surgical instruments based on spatio-temporal context and deep learning. Comput Assist Surg (Abingdon) 2019; 24(Suppl 1): 20-9.
7. Khoiy KA, Mirbagheri A, Farahmand F. Automatic tracking of laparoscopic instruments for autonomous control of a cameraman robot. Minim Invasive Ther Allied Technol 2016; 25: 121-8.
8. Liu T, Çavuşoğlu MC. Optimal needle grasp selection for automatic execution of suturing tasks in robotic minimally invasive surgery. IEEE Int Conf Robot Autom 2015; 2015: 2894-900.
9. Jain S, Argall B. Grasp detection for assistive robotic manipulation. IEEE Int Conf Robot Autom 2016; 2016: 2015-21.
10. Sundaresan P, Thananjeyan B, Chiu J, Fer D, Goldberg K. Automated extraction of surgical needles from tissue phantoms. 2019 IEEE 15th International Conference on Automation Science and Engineering (CASE) Vancouver, BC, Canada; 2019: 170-7.
11. D'Ettorre C, Dwyer G, Du X, Chadebecq F, Vasconcelos F, de Momi E, et al. Automated pick-up of suturing needles for robotic surgical assistance. IEEE Intl Conf Robot Autom 2018; 1370-7.
12. Sen S, Garg A, Gealy DV, McKinley S, Jen Y, Goldberg K. Automating Multiple-Throw Multilateral Surgical Suturing with a Mechanical Needle Guide and Sequential Convex Optimization. 2016 IEEE International Conference on Robotics and Automation (ICRA) 2016; DOI: 10.1109/ICRA.2016.7487611
13. Jackson RC, Yuan R, Chow DL, Newman W, Çavuşoğlu MC. Automatic initialization and dynamic tracking of surgical suture threads. IEEE Int Conf Robot Autom 2015; 2015: 4710-6.
14. Leonard S, Wu KL, Kim Y, Krieger A, Kim PC. Smart tissue anastomosis robot (STAR): a vision-guided robotics system for laparoscopic suturing. IEEE Trans Biomed Eng 2014; 61: 1305-17.
15. Chow D, Jackson RC, Çavuşoğlu MC, Newman W. A novel vision guided knot-tying method for autonomous robotic surgery. 2014 IEEE International Conference on Automation Science and Engineering (CASE) Taipei; 2014: 504-8.
16. Knoll A, Mayer H, Staub C, Bauernschmitt R. Selective automation and skill transfer in medical robotics: a demonstration on surgical knot-tying. Int J Med Robot 2012; 8: 384-97.
17. Najarian S, Fallahnezhad M, Afshari E. Advances in medical robotic systems with specific applications in surgery--a review. J Med Eng Technol 2011; 35: 19-33.
18. Moustris GP, Hiridis SC, Deliparaschos KM, Konstantinidis KM. Evolution of autonomous and semi-autonomous robotic surgical systems: a review of the literature. Int J Med Robot 2011; 7: 375-92.
19. Majewicz A, Wedlick TR, Reed KB, Okamura AM. Evaluation of robotic needle steering in ex vivo tissue. IEEE Int Conf Robot Autom 2010; 2010: 2068-73.
20. Moreira P, Patil S, Alterovitz R, Misra S. Needle steering in biological tissue using ultrasound-based online curvature estimation. IEEE Int Conf Robot Autom 2014; 2014: 4368-73.
21. Opfermann JD, Leonard S, Decker RS, Uebele NA, Bayne CE, Joshi AS, et al. Semi-autonomous electrosurgery for tumor resection using a multi-degree of freedom electrosurgical tool and visual servoing. Rep U S 2017; 2017: 3653-9.
22. Wang M, Ma Y. A review of virtual cutting methods and technology in deformable objects. Int J Med Robot 2018; 14: e1923.
23. Wang Z, Majewicz Fey A. Deep learning with convolutional neural network for objective skill evaluation in robot-assisted surgery. Int J Comput Assist Radiol Surg 2018; 13: 1959-70.
24. Zia A, Essa I. Automated surgical skill assessment in RMIS training. Int J Comput Assist Radiol Surg 2018; 13: 731-9.
25. Hung AJ, Chen J, Gill IS. Automated performance metrics and machine learning algorithms to measure surgeon performance and anticipate clinical outcomes in robotic surgery. JAMA Surg 2018; 153: 770-1.
26. Cerrolaza JJ, Picazo ML, Humbert L, Sato Y, Rueckert D, Ballester MÁG, et al. Computational anatomy for multi-organ analysis in medical imaging: A review. Med Image Anal 2019; 56: 44-67.
27. Lin B, Sun Y, Qian X, Goldgof D, Gitlin R, You Y. Video-based 3D reconstruction, laparoscope localization and deformation recovery for abdominal minimally invasive surgery: a survey. Int J Med Robot 2016; 12: 158-78.
28. Veruggio G. EURON Roboethics Roadmap. EURON Roboethics Atelier, Genoa, Feb 20-March 3, 2006. Available from: https://www3.nd.edu/~rbarger/ethics-roadmap.pdf (July 1, 2006).
29. O'Sullivan S, Nevejans N, Allen C, Blyth A, Leonard S, Pagallo U, et al. Legal, regulatory, and ethical frameworks for development of standards in artificial intelligence (AI) and autonomous robotic surgery. Int J Med Robot 2019; 15: e1968.
30. Matthias A. The responsibility gap: ascribing responsibility for the actions of learning automata. Ethics Inf Technol 2004; 6: 175-83.
31. Bonaci T, Yan J, Herron J, Chizeck HJ. Experimental analysis of cyber security attacks on teleoperated surgical robotics. In preparation to the ACM Transaction on Cyber-Physical Systems 2017.

What is your diagnosis?

A 29-year-old, 20-month pregnant woman was admitted to our center, since she had a history of hydrocephalus and anencephaly in her previous pregnancy. There was no history of taking folic acid tablets before pregnancy, but she took them in the first trimester of her current pregnancy. A detailed obstetric history was taken and it was determined that the patient had a consanguineous marriage with a second cousin. She had a history of three previous pregnancies. Two of them resulted in the birth of healthy babies and one was terminated because of hydrocephalus and anencephaly.

Fetal ultrasonographic findings were: a hyperechogenic, cystic structure, measuring 28x31 mm in the right lobe of the liver, hyperechogenic and polycystic left kidney, cystic hygroma, pulmonary hypoplasia and single umblical artery. On magnetic resonance imaging, there were no cerebellar vermis and an absent corpus callosum (Figure 1). Informed consent was obtained and amniocentesis was performed. The patient decided to terminate the pregnancy without waiting for the result of amniocentesis, so the pregnancy was terminated and the fetus was sent for an autopsy examination.

The male fetus weighed 476 g. The crown rump length, chest circumference and abdominal circumference were measured as 18 cm, 19 cm, 21 cm, respectively. It was seen that cerebellar vermis and corpus callosum were absent. The fetus had bulging eyes, broad nose, depressed nasal bridge, folded ears, large mouth with a protruding tongue, long philtrum, and small chin. An increase in his nuchal fold thickness was found with the help of an external examination (Figure 2).

On dissection and internal examination: cardiac defect and diaphragmatic hernia were not observed. A serous cystic structure with a size of 28x31 mm was observed in the right lobe of the liver (Figure 3). The left kidney parenchyma could not be observed. Pulmonary hypoplasia was observed (total lung weight was 21 grams). Amniocentesis reported a normal karyotype.

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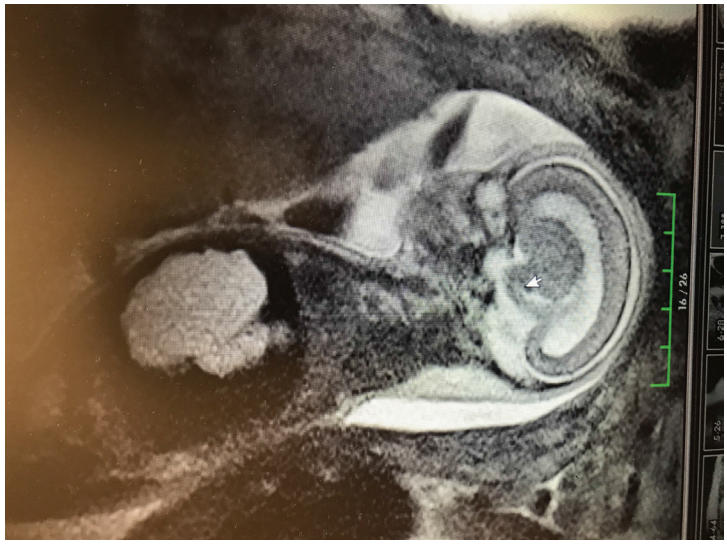

Figure 1. Magnetic resonance imaging of agenesis of corpus callosum

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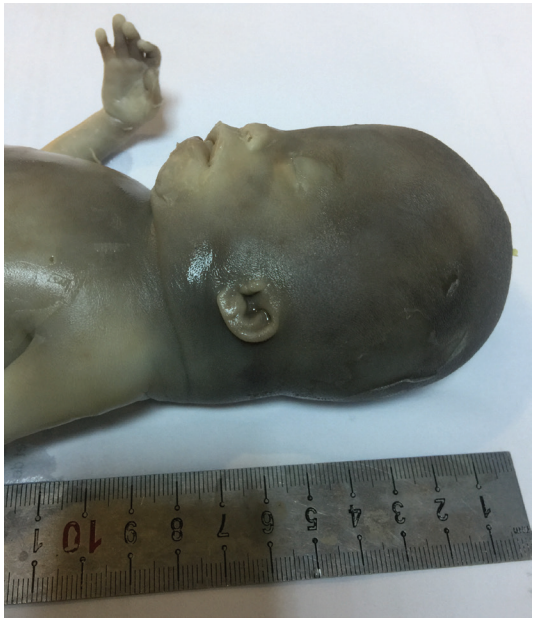

Figure 2. Bulging eyes, broad nose, depressed nasal bridge, folded ears, large mouth with a protruding tongue, long philtrum, small chin and increase in nuchal fold thickness

Figure 3. Hyperechogenic cystic structure measuring 28x31 mm in the right lobe of the liver

Answer

Fryns syndrome is a rare, autosomal recessive disorder with multiple congenital anomalies and has a prevalence of about 0.7 per 10,000 births (1). It is characterized by diaphragmatic defects, typical face, distal digital or nail hypoplasia, pulmonary hypoplasia and some associated anomalies which may include polyhydramnios, cloudy corneas and/or microphthalmia, orofacial clefting, renal dysplasia/renal cortical cysts, and/or malformations including the cardiovascular system, gastrointestinal system, brain or the genitals. It is also closely related with consanguineous marriage. The diagnosis is based on clinical findings and is made with the presence of three criteria (2,3). Regarding genetic analysis, fetal karyotypes of cases are usually normal, so it is important to diagnose the syndrome at autopsy. Fryns syndrome, known as the most common autosomal recessive disorder causing congenital diaphragmatic hernia (CDH), is responsible for 4-10% of all patients with CDH. Diaphragmatic defects are found in almost all cases with Fryns syndrome (4). A limited number of cases with Fryns syndrome without diaphragmatic hernia have been identified in the literature (5).

The purpose of this autopsy case report is to show that Fryns syndrome can be diagnosed without CDH and to show its association with rare abnormalities, such as cystic hygroma, agenesis of the corpus callosum and the cerebellar vermis.

Fryns syndrome is one of the most common syndromes associated with CDH and is reported in up to 10% of patients with CDH. Despite the fact that there wasn't any diaphragmatic defect, other findings of our patient were similar to the rest of the typical diagnostic findings, with the exception of cystic hygroma, and some other defects not previously described. Several chromosomal abnormalities show symptoms similar to Fryns syndrome. Therefore, the diagnosis of Fryns syndrome can only be made if the karyotype is normal and the diagnosis is confirmed by autopsy. In this case, autopsy findings were very useful in making a differential diagnosis. Based on the ultrasonographic findings, the first idea was chromosomal anomaly. However, amniocentesis was reported as normal. We did not recommend non-invasive prenatal testing as an alternative to amniocentesis as non-invasive prenatal testing is not suitable for genetic evaluation of ultrasound anomalies (6). Simpson-Golabi-Behmel syndrome, an X-linked overgrowth syndrome resulting from deletions or mutations in the *GPC3* gene, and conditions with hypoplasia or absence of the distal phalanges such as DOOR syndrome (deafness, onychodystrophy, osteodystrophy, and mental retardation), Schinzel-Giedion syndrome, and Rudiger syndrome should be considered in differential diagnosis. However, heterozygous de novo mutation in the *SETBP1* gene is characteristic of Schinzel-Giedion syndrome and none of the patients with Rudiger syndrome has diaphragmatic hernia. The diagnosis of Fryns syndrome without diaphragmatic hernia is quite difficult. Although this patient is diagnosed as Fryns syndrome, it cannot clearly be differentiated from Rudiger syndrome and Schinzel-Giedion syndrome. Therefore, these diseases can be called Fryns-like syndromes (7). In the presence of cystic hygroma,

other signs of Fryns syndrome should be carefully monitored, since the risk of recurrence is 25 percent per pregnancy and should be kept in mind during subsequent pregnancies.

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References

1. Nirmaladevi M, Kurup S, Ajitha EV. Fryns syndrome in monozygotic twins: a case report with review of literature. Int J Morphol 2012; 30: 56-60.
2. Lin AE, Pober BR, Mullen MP, Slavotínek AM. Cardiovascular malformations in fryns syndrome: is there apathogenic role for neural crest cells? Am J Med Genel A 2005; 139: 186-93.
3. Slavotinek A. Fryns syndrome. GeneReviews® [Internet]. 2007.
4. Dillon E, Renwick M. Antenatal detection of congenital diaphragmatic hernias: the Northern region experience. Clin Radiol 1993; 48: 264-7.
5. Alessandri L, Brayer C, Attali T, Samperiz S, Tiran-Rajaofera I, Ramful D, et al. Fryns syndrome without diaphragmatic hernia. Report on a new case and review of the literature. Genet Couns 2005; 16: 363-70.
6. Beulen L, Faas BHW, Feenstra I, Van Vugt JMG, Bekker MN. Clinical utility of non-invasive prenatal testing in pregnancies with ultrasound anomalies. Ultrasound Obstet Gynecol 2017; 49: 721-8.
7. Alessandri JL, Cuillier F, Malan V, Brayer C, Grondard M, Jacquemot-Dekkak L, et al. Fryns syndrome without diaphragmatic hernia, DOOR Syndrome or fryns-like syndrome? Report on patients from Indian Ocean Islands. Am J Med Genet A 2014; 164A: 648-54.

Elective peripartum hysterectomy for placenta accreta: is it always a severe maternal morbidity?

To the Editor,

The American College of Obstetricians and Gynaecologists (ACOG) and the Society of Fetal Medicine (SFM) have published a document saying that records of all women needing more than 4 units of blood transfusion or intensive care unit (ICU) admission must be reviewed to identify women with severe maternal morbidity (1).

To date, there is no clear consensus as to what conditions should be termed severe maternal morbidity. ACOG and SFM have given an example list of diagnoses and complications constituting severe maternal morbidity, wherein all women undergoing emergency or unplanned peripartum hysterectomy are assumed to have severe morbidity, while planned peripartum hysterectomies for cancers are not considered to cause severe morbidity. All cases of hysterectomy for placenta accreta have been classified as severe morbidity irrespective of the number of blood transfusions or ICU admission (1).

The government of India's operational guidelines for the "maternal near-miss" state that any woman who requires emergency hysterectomy for controlling blood loss is to be labeled as a near-miss (2). World Health Organization classifies hysterectomy due to uterine infection or hemorrhage as a critical intervention, labeling the woman as a near-miss (3). However, in all these guidelines, a planned peripartum hysterectomy done for a pre-operatively diagnosed placenta accreta where the woman remains hemodynamically stable does not find an appropriate mention.

Placenta accreta spectrum (PAS), formerly known as morbidly adherent placenta, refers to the range of pathologic adherence of the placenta, including placenta increta, placenta percreta, and placenta accreta. This is definitely a potentially life-threatening condition with an increasing incidence all over the world, secondary to the rising caesarean section rate. The recommended management is a planned caesarean hysterectomy before term, where no attempt must be made to separate the placenta from the uterus. Though the exact timing of scheduled delivery remains controversial, with ACOG recommending planned surgery at 34 to 36 weeks, Royal College of Obstetrics and Gynecology at 35 to 36+6 weeks and SFM at 34 to 37 weeks, the basic principle is to balance the benefit of fetal lung maturity with a risk of excessive haemorrhage when the surgery is performed in an emergency setting (4-6).

Optimal pre-operative preparation, adoption of a multidisciplinary approach and minimizing blood loss intraoperatively are the most critical steps in the management of PAS. Many surgical approaches have been suggested over time with variable benefits. One such approach is separating the bladder up to the cervico-vaginal junction prior to uterine incision so that the aberrant blood vessels traversing between uterus and bladder are ligated and divided. We have reported significantly reduced blood loss with this technique, minimizing the need for massive blood transfusion. None of the women out of 12 cases performed over 17 months required ICU stay or had bladder or ureteric injury (7).

When performed in tertiary centers with all necessary preparation by a multidisciplinary expert team, this surgery may have significantly reduced morbidity. Thus the present recommendation of considering all peripartum hysterectomy performed for placenta accreta as a near miss may not always be appropriate. In our opinion peripartum hysterectomy performed for placenta accreta should be further classified in order to properly categorize the cases which actually are near misses.

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References

1. Obstetric Care Consensus No. 5: Severe maternal morbidity: screening and review. Obstet Gynecol 2016; 128: e54-60.
2. Mishra CK. Maternal Near Miss Review Operational Guidelines. December 2014. Annexure 2: 35-40.
3. Say L, Souza JP, Pattinson RC, WHO working group on Maternal Mortality and Morbidity classifications. Maternal near miss- towards a standard tool for monitoring quality of maternal health care. Best Pract Res Clin Obstet Gynaecol 2009; 23: 287-96.
4. Obstetric Care Consensus No. 7: Placenta Accreta Spectrum. American College of Obstetrics and Gynaecologists; Society for Maternal-Fetal Medicine. Obstet Gynecol 2018; 132: e259-75.
5. Jauniaux ERM, Alfirevic Z, Bhide AG, Belfort MA, Burton GJ, Collins SL, et al. Placenta Praevia and Placenta Accreta: Diagnosis and Management. Green-top Guideline No. 27a. BJOG 2019; 126: e1-48.
6. Society for Maternal-Fetal Medicine (SMFM), Gyamfi-Bannerman C. Society for Maternal- Fetal Medicine (SMFM) consult series #44: Management of bleeding in the late preterm period. Am J Obstet Gynecol 2018; 218: B2-8.
7. Saha PK, Bagga R, Kalra JK, Arora A, Singla R, Suri V, et al. An alternate surgical approach to reduce haemorrhage and complications during caesarean hysterectomy for adherent placenta. Eur J Obstet Gynecol Reprod Biol 2018; 228: 215-20.

LESS hysterectomy through a bluntly created 11 mm incision

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Abstract

In the field of minimally invasive surgery, there is a constant drive to devise and execute the most minimally invasive surgeries possible. By the very nature of laparoscopy and robotic surgery, what one can accomplish with several ports of a given size will invariably be studied and attempted with fewer ports and with ports of smaller sizes. After researching the literature, we were not able to find any single port hysterectomies performed through a port size of smaller than 15 mm. We were able to perform, described here, a technique for performing laparoscopic hysterectomy through a single port of only 11 mm in diameter. We illustrate the technique in the accompanying video and believe the technique to be safe and reproducible.

Keywords: Hysterectomy, single port, LESS, laparoendoscopic single site surgery, robotic hysterectomy, laparoscopic hysterectomy, laparoscopy

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Introduction

Unlike other specialties which are defined by the general field of medicine they pertain to, "minimally invasive surgery" itself can be understood as a challenge to its practitioners, its very name encouraging them to pursue a more minimally invasive approach. The specific issue we sought to address here was attempting the most minimally invasive, single-port hysterectomy ever performed, while still performing meaningful laparoscopic visualization of the abdomen and with the expectation to be able to realistically operate in the abdomen from a laparoscopic approach. This meant that we specifically did not wish to perform a procedure that one could consider to be a laparoscopy then followed by vaginal hysterectomy, and desired meaningful laparoscopic access to deal with issues such as adhesions, mobilization of the bladder flap, or performing a bilateral salpingo-oophorectomy without significant vaginal assistance. After researching the literature, we were not able to find any single port hysterectomies performed through a port size of smaller than 15 mm (1). The authors also wished to exclude cases with no significant pathology or adhesive disease, as the purpose of describing the technique is to show that the technique can be used in many challenging situation, not to demonstrate the technique can be successful on the easiest of hysterectomies. All authors strongly contend that hysterectomies that can be performed through a completely vaginal technique should be, and that a vaginal hysterectomy, or zero port hysterectomy, is superior to laparoscopic hysterectomy, if it can be accomplished (2).

This Manuscript has been reviewed by the institutional IRB board at Marchand Institute and was found to be exempt from IRB review (July 2018).

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Multiple authors have documented the feasibility of single incision laparoscopic hysterectomy (3). Many authors have commented that the idea, although novel, does not significantly improve intra-operative pain, recovery or surgical cosmesis (4). The most commonly used system is a robotic assisted single port system. All systems, to the knowledge of the authors, require incisions greater than 15 mm in the umbilicus (5,6). We examined different single port systems and combined available instrumentation to create a feasible, repeatable technique for performing a laparoscopic single site hysterectomy using only an 11 mm umbilical incision that is created with a blunt laparoscopic trochar. We have explained the technique in a video for reproducibility.

Objective

We devised a technique for laparoscopic single port hysterectomy based on the concept that a bluntly created incision would be less likely to herniate than a sharply created incision. Therefore, after creating the initial skin incision with an 11-blade scalpel, (Figure 1) rather than perform an open dissection that would result in a large incision and a much larger fascial footprint, we then place an 1 mm blunt laparoscopic trochar (Figure 2) into the incision after insufflating with a veress needle. The multiport device is then loaded into its introducer, (Figure 3) and is inserted into the abdominal cavity after removing the 11 mm port from the umbilical incision (Figure 4). The multiport device can then be installed and actively utilized to perform the hysterectomy through only an 11 mm incision (Figure 5). Following this, the uterine pedicles are divided with a bipolar power coagulation and division device, and the circumferential colpotomy is made with a monopolar cautery set to 30 watts of coagulating current with a laparoscopic hook extender. The vaginal cuff is sewn from the vaginal approach and ovaries and tubes are removed after removal of the uterus. The patient returned foru weeks post-operatively and no scars were visible (Figure 6). We believe this technique to be significantly different from any

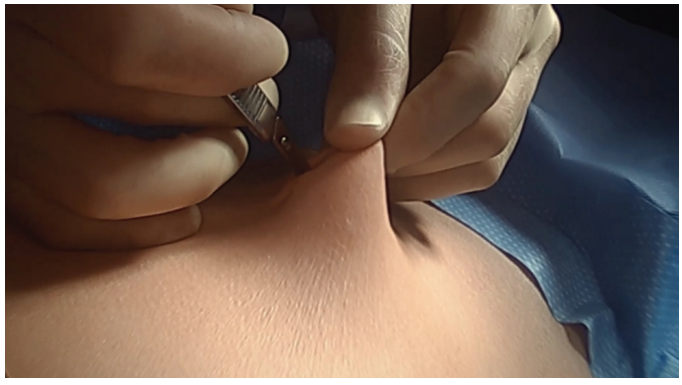

Figure 1. An 11 mm incision is made at the bottom of the patient's umbilicus with an 11-blade scalpel

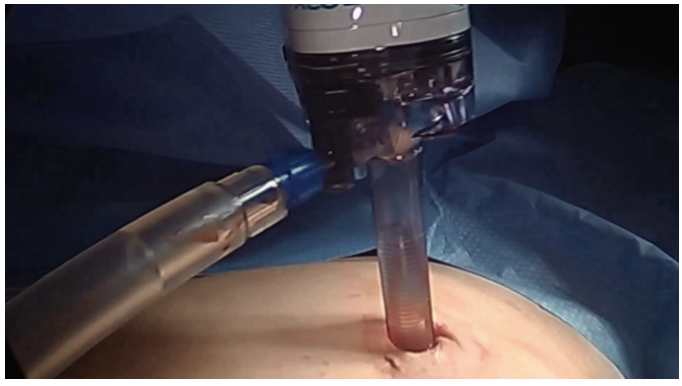

Figure 2. A blunt 11 mm laparoscopic trochar is utilized to make the entry into the abdominal cavity, in order to avoid a sharp dissection into the abdomen which would result in a larger fascial footprint

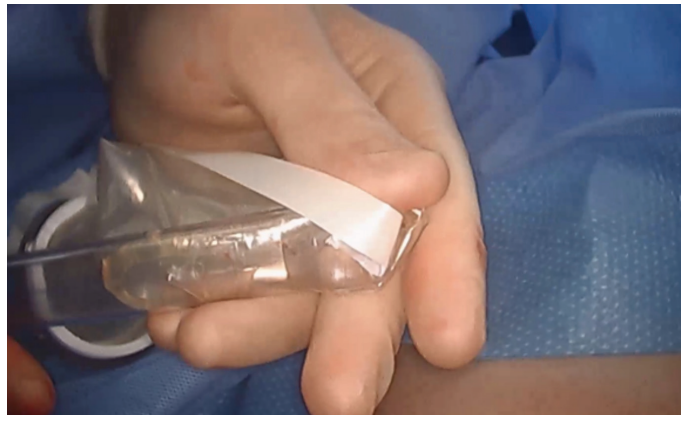

Figure 3. A multiport device is then loaded into the introducer, for insertion into the abdomen

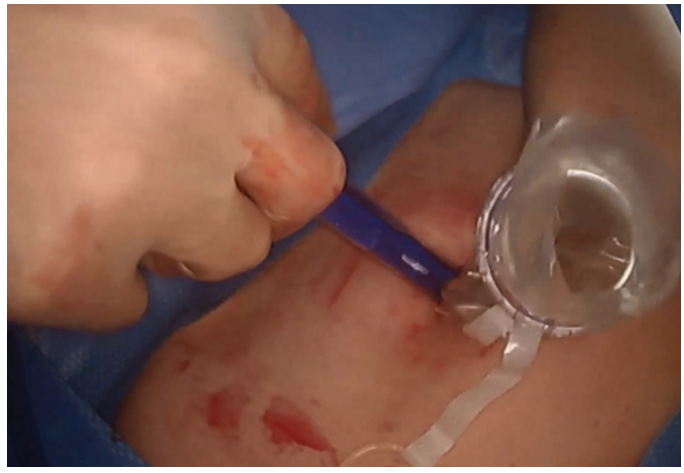

Figure 4. The multiport device is inserted through the abdominal incision after withdrawing the 11 mm blunt trochar. This ensures the incision width will not exceed 11 mm and has been created by blunt entry, which further decreases the chance of postoperative hernia

previously described techniques because of the usage of an 11 mm blunt trochar to create the umbilical incision. This creates a reproducible footprint in the fascia that should be identical and reproducible, regardless of circumstances. By keeping the incision small and created bluntly we believe the risk of postoperative herniation has been minimized (Figure 7).

Design

A narrated video demonstration of the surgical procedure (Canadian Task Force Classification III). We developed a novel method for performing laparoscopic hysterectomy through a single 11 mm incision that was created with a blunt trochar. The most novel aspects of our procedure involve the placement of a multiport manipulator device through a small, 11 mm incision created by an 11 mm blunt trochar. It is our belief that the small size of this blunt trochar likely makes fascial closure unnecessary, although it is still recommended by the authors.

Interventions

A 32-year-old woman with endometriosis, adenomyosis and chronic pelvic pain with recurrent ovarian cysts presented for laparoscopic hysterectomy with bilateral salpingo-oophorectomy. The patient had previously tried more conservative surgeries and medical treatments, including a six-month course of luprolide acetate and multiple surgeries for fulgaration of endometriosis. The patient completed her desired childbearing and requested definite treatment. The patient had a history of prior bilateral salpingectomy and one prior cesarean section. The patient had confirmed endometriosis at previous laparoscopic exploration, and was suspected to suffer from adenomyosis, based on cyclic pain and pain that seemed to originate from the uterus with gentle palpation with the vaginal ultrasound probe. Patient was extensively counseled to the risks of bilateral salpingo-oophorectomy and offered more conservative surgical options including hysterectomy without bilateral salpingo-oophorectomy. The patient refused more conservative treatments, citing her fear of the necessity of future surgeries for endometriosis or ovarian cysts, the desire for definitive treatment of endometriosis, as well her fear of ovarian cancer in the future, despite there being no family history. Patient politely refused BRCA testing, citing that it would not influence her decision for bilateral salpingo-oophorectomy. The total operative time was 38 minutes, and the estimated blood loss was 100 cc. The patient was discharged 18 hours after surgery and the recovery was uneventful. The final pathology report showed endometriosis and adenomyosis.

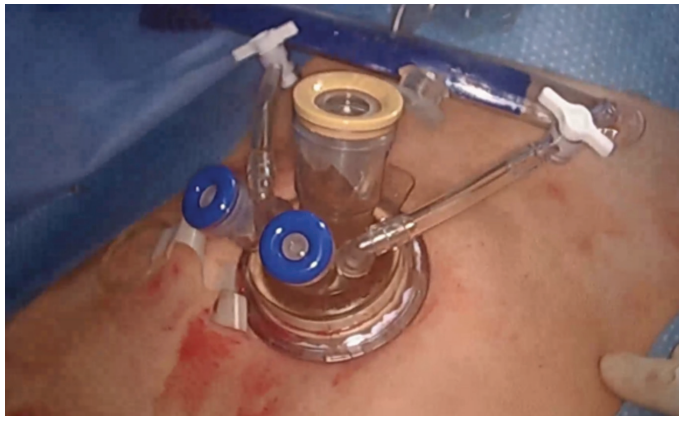

Figure 5. The multipart device is in place and the laparoscopic hysterectomy can proceed with one or two instruments in addition to the 5 mm laparoscope

Figure 6. The patient's abdomen at a visit four weeks after surgery. No scars are visible

Conclusion

Our described technique is a feasible, reproducible procedure for hysterectomy and may improve cosmesis and postoperative pain over traditional laparoscopic and single port techniques.

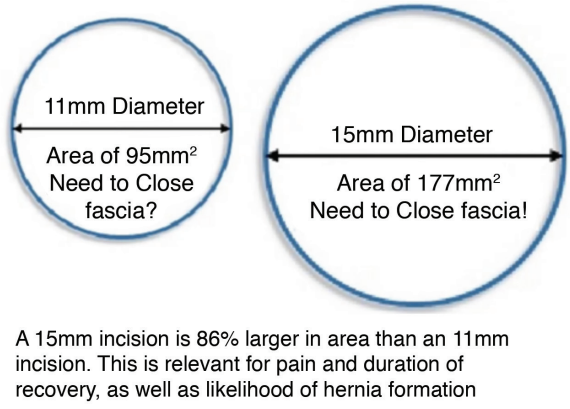

Figure 7. Secondary to the fact that laparoscopic incisions are stretched into a circular shape by the penetrating instrumentation, even a small decrease in the size of a fascial incision will greatly decrease the area of the opening that can pass through that incision. This figure compares the large jump from an area of 95 mm² to 177 mm² when increasing the umbilical incision by only 3 mm

Acknowledgement: *Dr. Marchand would like to acknowledge the time and effort of the students, residents, clinical researchers and fellows at the institute who contributed to producing this video.*

Video 1.

https://www.doi.org/10.4274/jtgga.galenos.2020.2020.0028.video1

Informed Consent: *Patient gave written consent for usage of video prior to and after procedure.*

Conflict of Interest: *No conflict of interest was declared by the authors.*

Financial Disclosure: *The authors declared that this study received no financial support.*

References

1. Langebrekke A, Qvigstad E. Total laparoscopic hysterectomy with single-port access without vaginal surgery. J Minim Invasive Gynecol 2009; 16: 609-11.
2. Garry R, Fountain J, Mason SU, Hawe J, Napp V, Abbott J, et al. The evaluate study: two parallel randomised trials, one comparing laparoscopic with abdominal hysterectomy, the other comparing laparoscopic with vaginal hysterectomy. BMJ 2004; 328: 129.
3. Behnia-Willison F, Foroughinia L, Sina M, McChesney P. Single-incision laparoscopic surgery (SILS) in gynaecology: feasibility and operative outcomes. Aust N Z J Obstet Gynaecol 2012; 52: 366-70.
4. Song T, Kim ML, Jung YW, Yoon BS, Joo WD, Seong SJ. Laparoendoscopic single-site versus conventional laparoscopic gynecologic surgery: a meta-analysis of randomized controlled trials. Am J Obstet Gynecol 2013; 209: 317.e1-9.
5. Demir RH, Marchand GJ. SELS: Laparoscopic oophorectomy, bagging and vaginal morcellation. J Minim Invasive Gynecol 2011; 18: S114.
6. Cela V, Marrucci E, Angioni S, Freschi L. Robot-assisted laparoscopic single-site hysterectomy: our experience and multicentric comparison with single-port laparoscopy. Minerva Ginecol 2018. doi: 10.23736/S0026-478104197-7.

Robot-assisted laparoscopic myomectomy for FIGO type II sub-mucosal leiomyoma without endometrial injury for a patient with history of miscarriage

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Abstract

To introduce a technique for robot-assisted laparoscopic myomectomy for FIGO type II sub-mucosal leiomyoma with >50% myometrial extension, without endometrial injury. A narrated video demonstration of our technique has been provided. Our patient was a 35-year-old, gravida 1, para 0 woman with secondary infertility. She had been married for three years. She complained of heavy menstrual bleeding and severe dysmenorrhea with a pain score of 10 on visual analogue scale (VAS). Surgery was done after thorough counseling and an informed consent was obtained. Institutional Review Board number: KC17OESI0375, approval date: 21.09.2018. Several steps can be taken to help prevent endometrial injury, and these include: (1) proper preoperative imaging to plan surgery; (2) use of intraoperative ultrasound to determine best location of incision; (3) use of a "cold cut" technique with monopolar curved scissors without energy to avoid obscuring the border between the leiomyoma and the endometrium; (4) careful millimeter by millimeter dissection; (5) use of diluted indigo carmine to aid delineation of the endometrial cavity during dissection. The patient had a normal post-operative course. On follow-up her VAS pain score was 0. Transvaginal ultrasound repeated four months postoperatively showed normalization of uterine anatomy and endometrial contour. Robot-assisted laparoscopic myomectomy may be an option to preserve fertility and minimize endometrial injury. This surgical method allows complete removal of large sub-mucosal leiomyomas in one session with exact suturing.

Keywords: Endometrium, surgical procedures, robotic, fibroid uterus

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Introduction

Uterine leiomyomas are the most common benign gynecologic tumors, with an estimated 70-80% of reproductive-aged women having leiomyomas (1). Most patients are asymptomatic but around 30% to 40% present with symptoms. Symptoms depend on location, size and number of leiomyomas (2) and include heavy menstrual bleeding, pain, pressure symptoms, infertility and recurrent pregnancy loss (3).

Around 5-10% of patients with infertility are found to have leiomyomas, with 1-2.4% having them as the only finding. Patients with sub-mucous myomas were found to have a lower clinical pregnancy rate, implantation rate, and ongoing pregnancy and live birth rate, with a significantly higher spontaneous abortion rate (4). Casini et al. (5) found that pregnancy rates were higher after myomectomy in women with sub-mucosal leiomyomas compared with expectant management.

In this video (Video 1) we present our technique for management of type II leiomyoma, in a 35-year old woman with a history of infertility. She had a history of one first trimester miscarriage and was complaining of heavy menstrual bleeding and severe dysmenorrhea. Management options were discussed, and it was decided to opt for robot-assisted laparoscopic removal as, in her case, hysteroscopic removal may have resulted in a significant destruction of the

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enodometrial surface. Robot-assisted laparoscopic removal can also be beneficial when the leiomyoma is large and may need more than one operation for complete removal. A robot-assisted technique is also a good option when the distance between the leiomyoma and the serosa is small, in order to avoid the risk of perforation.

A clinical aim was to keep the endometrium intact and this was achieved through several steps, starting with proper preoperative imaging (Figure 1) and planning. In our opinion it is also helpful to use a transvaginal ultrasound to determine the best location for the incision. With the use of "cold cut" technique, using a monopolar curved scissors without energy, and by careful millimeter-by-millimeter dissection, removal of the leiomyoma was accomplished with minimal damage to the adjacent myometrium. Diluted indigo carmine can aid in dissection as it delineates the cavity and it can also aid in suturing the endometrium, in case of injury (Figure 2).

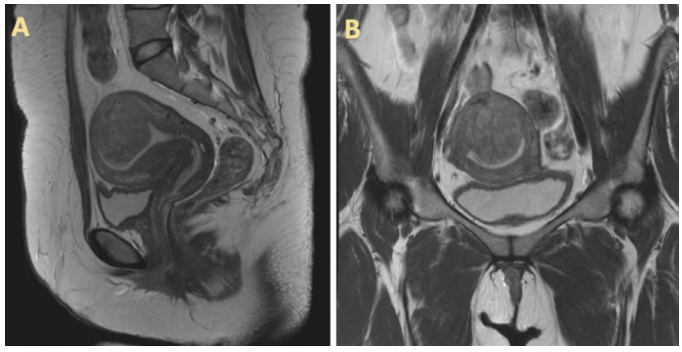

Figure 1. Pelvic magnetic resonance imaging (MRI). (A) Sagittal and (B) coronal pelvic MRI showed a sub-mucosal, posterior fundal uterine leiomyoma measuring 4x4x4.5 cm

After removal of the leiomyoma, the defect was closed in two layers and the leiomyoma was then removed via contained morcellation. The patient was followed up with an ultrasound four months after surgery, which showed normalization of the uterine anatomy and endometrial contour (Figure 3).

Robot-assisted laparoscopic myomectomy may be an option to preserve fertility and minimize endometrial injury. This surgical method allows complete removal of large sub-mucosal leiomyomas in one session with exact suturing.

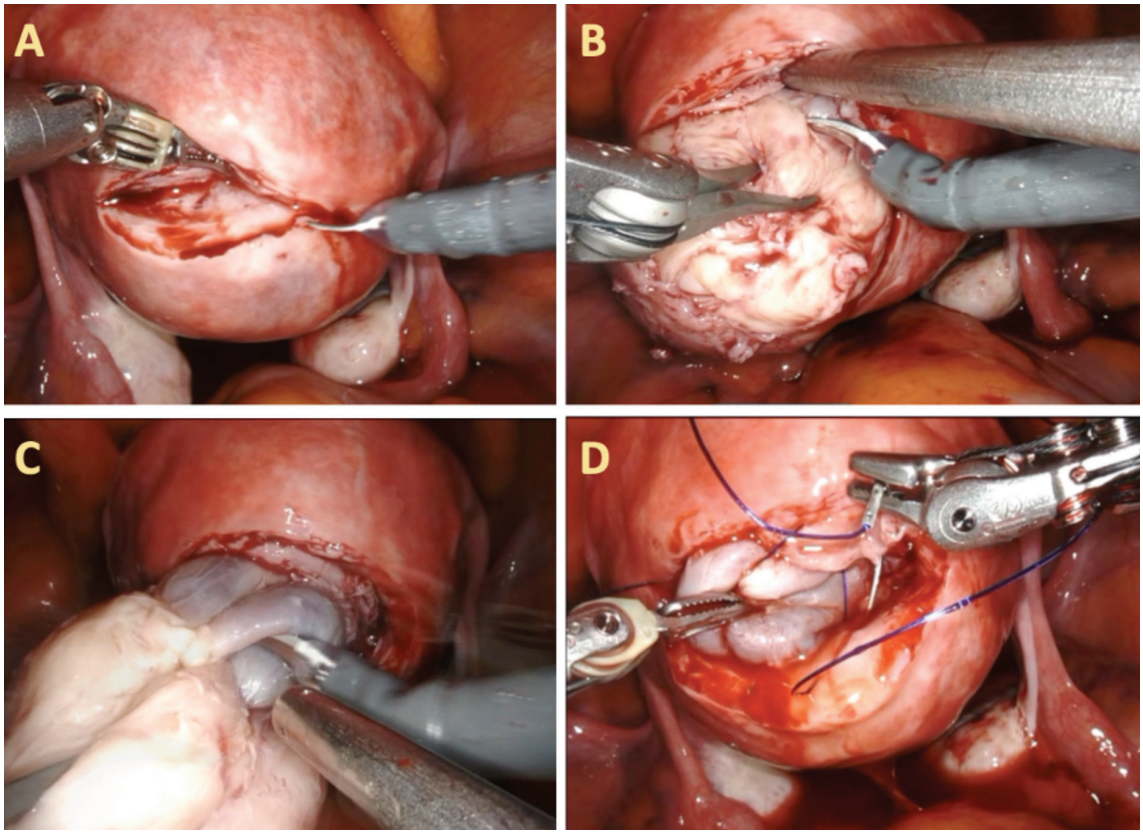

Figure 2. Surgical procedure. Monopolar curved scissors without electrocauterization (also called "cold-cut"), was used to make a transverse incision along the uterine wall overlying the leiomyoma (A). A tenaculum forceps was then used to provide traction while sharp and mechanical dissection was continued (B). To ascertain endometrial integrity and to aid in dissection a diluted indigo carmine solution was injected into the endometrial cavity through the RUMI uterine manipulator (C). After enucleation of the leiomyoma, the defect was sutured in two layers (D)

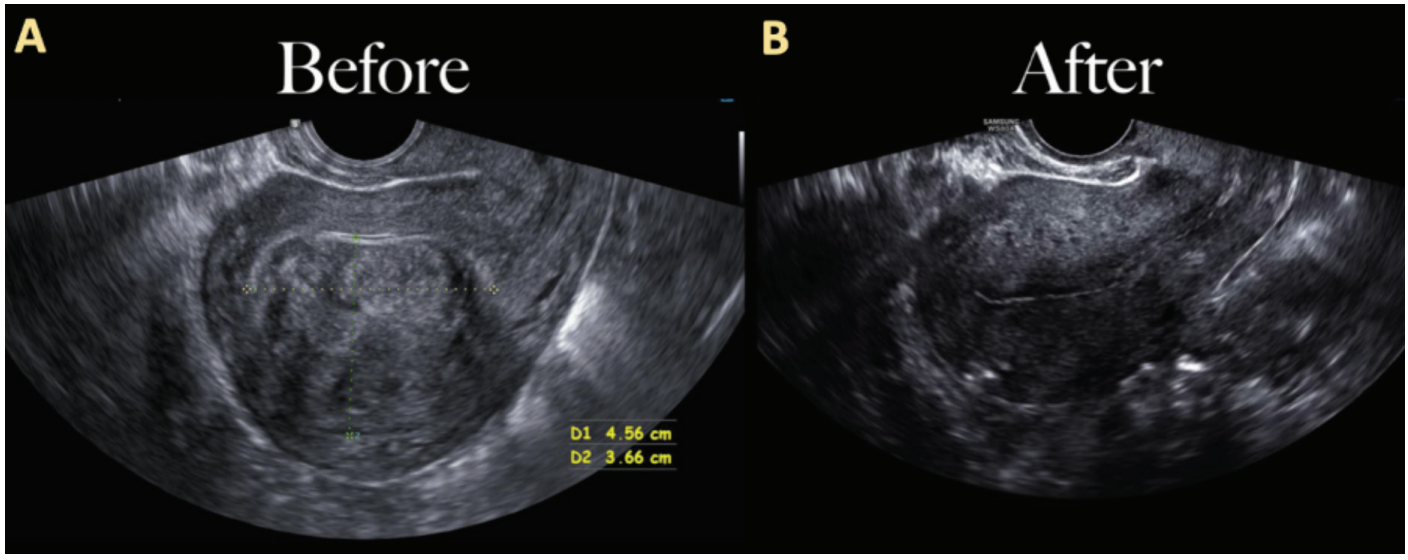

Figure 3. Transvaginal ultrasound of the uterus. Follow up transvaginal ultrasound (B) done 4 months postoperatively showed normalization of uterine anatomy and endometrial contour, compared to the preoperative ultrasound (A)

Video 1.

https://www.doi.org/10.4274/jtgga.galenos.2020.2020.0139.video1

Conflict of Interest: *No conflict of interest was declared by the authors.*

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References

1. Gingold JA, Gueye NA, Falcone T. Minimally invasive approaches to myoma management. J Minim Invasive Gynecol 2018; 25: 237-50.
2. Flyckt R, Coyne K, Falcone T. Minimally invasive myomectomy. Clin Obstet Gynecol 2017; 60: 252-72.
3. Closon F, Tulandi T. Uterine myomata: organ-preserving surgery. Best Pract Res Clin Obstet Gynaecol 2016; 35: 30-6.
4. Cook H, Ezzati M, Segars JH, McCarthy K. The impact of uterine leiomyomas on reproductive outcomes. Minerva Ginecol 2010; 62: 225-36.
5. Casini ML, Rossi F, Agostini R, Unfer V. Effects of the position of fibroids on fertility. Gynecol Endocrinol 2006; 22: 106-9.

Essure® removal in 10 steps

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Abstract

Many women request Essure® removal because of possible side effects related to the device itself. Laparoscopic Essure® removal in symptomatic women may be associated with improvement in quality of life. We aim to describe the surgical technique in ten steps in the accompanying video as the standardization of the laparoscopic Essure® removal procedure could help to diminish the risk of fractures of the device with this easy and safe 10-step procedure.

Keywords: Essure, surgical technique, salpingectomy, laparoscopy

Received: 06 September, 2020 **Accepted:** 18 November, 2020

Introduction

Several studies have demonstrated an improvement of symptomatology and quality of life after removal of the Essure® device in symptomatic patients (1,2). The pathophysiology of adverse effects related to the device may be explained by the release of heavy metals from a possible corrosion of the implant (3). Therefore, because there is a risk of fracture in up to 30% of cases (2), the implant should be removed completely and safely (4). Our aim was to give a step-by-step description of an easy surgical technique with a demonstrative video.

Surgery technique

This video clearly described the laparoscopic technique in 10 steps (Video 1): 1) pelvis exploration; 2) peritoneal cytology, for two reasons a) heavy metal analysis b) usually done in our department during prophylactic and opportunistic salpingectomy because of the potential tubal pathway for ovarian carcinogenesis (3,5); 3) longitudinal incision over the proximal fallopian tube towards the uterine horn (Figure 1); 4) circumferential incision around the interstitial tubal portion; 5) circumferential incision on the 2/3 anterior portion of the fallopian tube (Figure 2); 6) horizontal incision of the tube under the proximal rectangular end of the microinsert; 7) hemostasis of the uterine horn; 8) Essure® removal under visual control; 9) Inspection and dissection of the Essure® device on a surgical drape (Figure 3); 10) bilateral salpingectomy and other associated procedures, peritoneal washing and prevention of postsurgical adhesions. As compared with laparoscopic myomectomy, the small incision in the myometrium to

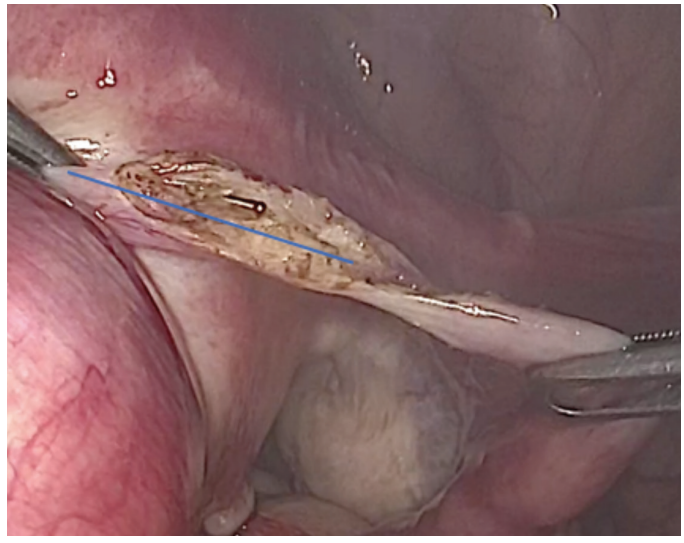

Figure 1. Longitudinal incision over the proximal fallopian tube towards the uterine horn

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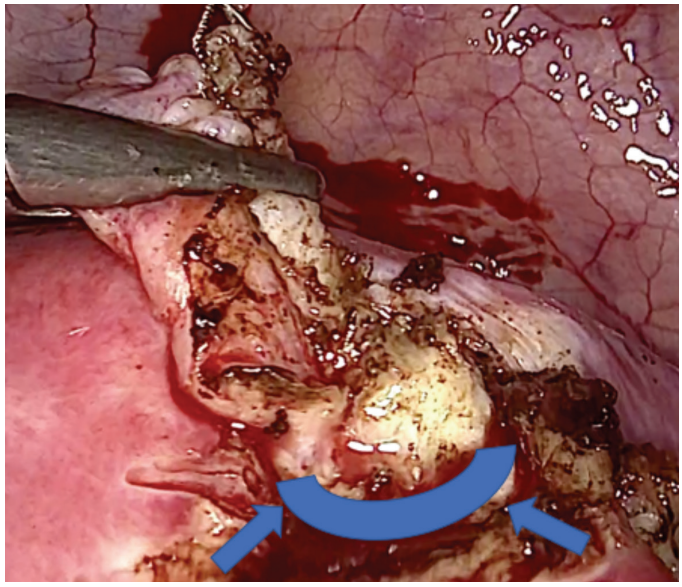

Figure 2. Circumferential incision on the 2/3 anterior portion of the fallopian tube

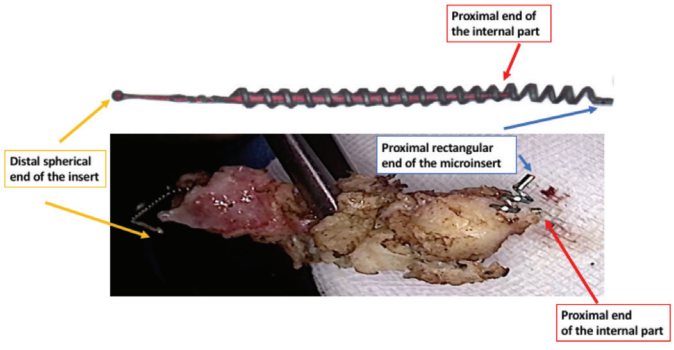

Figure 3. Inspection of the complete implant Essure®

perform this mini-cornuectomy should theoretically limit the risk of uterine rupture, if the patient wished to conceive via in vitro fertilization later. However further studies are required to confirm this retention of fertility (6).

Conclusion

Since improvement of quality of life has been demonstrated after laparoscopic Essure® removal in symptomatic women the standardization of the removal procedure could help to diminish the risk of fractures of the device.

Video 1.

https://www.doi.org/10.4274/jtgga.galenos.2020.2020.0159.video1

Conflict of Interest: *No conflict of interest was declared by the authors.*

Financial Disclosure: *The authors declared that this study received no financial support.*

References

1. Maassen LW, van Gastel DM, Haveman I, Bongers MY, Veersema S. Removal of essure sterilization devices: a retrospective cohort study in the Netherlands. J Minim Invasive Gynecol 2019; 26: 1056-62.
2. Chene G, Cerruto E, Moret S, Lebail-Carval K, Chabert P, Mellier G, et al. Quality of life after laparoscopic removal of essure® sterilization devices. Eur J Obstet Gynecol Reprod Biol X 2019; 3: 100054.
3. Parant F, Bost M, Nohuz E, Prost J, Cerruto E, Moret S, et al. Potential release of toxic metal elements from essure® device in symptomatic patients: first results of the French Ablimco cohort. Eur J Obstet Gynecol Reprod Biol 2020; 252: 434-8.
4. Chene G, Lamblin G, Mezzetta L, Lablanche O, Cerruto E, Nohuz E. How I do… easily laparoscopic residual essure® resection without hysterectomy? Gynecol Obstet Fertil Senol 2020; 48: 768-71.
5. Chene G, Rahimi K, Mes-Masson AM, Provencher D. Surgical implications of the potential new tubal pathway for ovarian carcinogenesis. J Minim Invasive Gynecol 2013; 20: 153-9.
6. Parker WH, Einarsson J, Istre O, Dubuisson JB. Risk factors for uterine rupture after laparoscopic myomectomy. J Minim Invasive Gynecol 2010; 17: 551-4.

CONGRESS CALENDER

INTERNATIONAL MEETINGS

(for detailed International Meeting please go website:
http://www.medical.theconferencewebsite.com/conferences/obstetrics-and-gynaecology)

March 12-17, 2021	**Obstetrics and Gynecology 2021, United States (VIRTUAL)**
March 25-27, 2021	**3rd World Congress on Maternal Fetal Neonatal Medicine, Venice, Italy**
April 20-23, 2021	**19th World Congress of the Academy of Human Reproduction, Jerusalem, Israel**
April 22-25, 2021	**Japan Society of Obstetrics and Gynaecology 73rd Annual Congress, Nigata, Japan**
May 05-08, 2021	**European Menopause and Andropause Society Conference, Florence, Italy**
May 12-16, 2021	**53rd Society for Obstetric Anesthesia and Perinatology Annual Meeting, New Orleans, United States**
June 04-06, 2021	**Twins Congress, Beijing, China**
June 23-26, 2021	**International Urogynecological Association 46th Annual Meeting, Singapore**
June 24-26, 2021	**VII. MIPS Annual Meeting, Athens, Greece**
June 27-30, 2021	**European Society of Human Reproduction and Embryology (ESHRE) virtual 37th Annual Meeting (VIRTUAL)**
July 06-09, 2021	**Society for Reproductive Investigation 68th Annual Meeting, Boston, United States**
July 14-17, 2021	**XXVII European Congress of Perinatal Medicine (ECPM), Lisbon, Portugal**
August 30-Sep 02, 2021	**International Gynecologic Cancer Society (IGCS) Meeting, Rome, Italy**
October 16-20, 2021	**American Society for Reproductive Medicine (ASRM) 77th Annual Meeting, Baltimore, Maryland, United States**
November 14-18, 2021	**50th American Association of Gynecologic Laparoscopists (AAGL) Global Congress on Minimally Invasive Gynecologic Surgery (MIGS), Austin, Texas, United States**

CONGRESS CALENDER

NATIONAL MEETINGS

(for detailed International Meeting please go website:
http://www.kongre2020.com)

April 01-04, 2021	**CİSED 5. Ulusal Cinsel Sağlık Kongresi, Antalya**
May 19, 2021	**TAJEV Master Class (Online)**
September 10-12, 2021	**Her Yönüyle Histerektomi Kongresi, İzmir**
September 22-26, 2021	**3. Obstetrik ve Jinekoloji Tartışmalı Konular Kongresi, Muğla**
November 10-14, 2021	**9. Üreme Sağlığı ve İnfertilite Kongresi - TSRM, Antalya**
December 03-05, 2021	**Çukurova Kadın Doğum Günleri 2021, Adana**