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Original Article

High Prevalence of Chronic Endometritis in Women Diagnosed With Hydrosalpinx Before In Vitro Fertilization Treatment

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ABSTRACT **Objective:** To compare the prevalence of chronic endometritis (CE) in women with hydrosalpinx undergoing *in vitro* fertilization (IVF), to a control group without hydrosalpinx.

Design: A bicentric historical prospective case-control study, between June 2017 and December 2021.

Setting: Angers and Montreal university hospitals.

Patient(s): In the Hydrosalpinx (H) group, we included all women undergoing IVF for various indications, and who were diagnosed with a hydrosalpinx before or during the cycle. In the control (C) group, we included women without hydrosalpinx, undergoing IVF for male factor infertility, or following bilateral tubal ligation.

Intervention(s): A laparoscopy was scheduled for the removal of the hydrosalpinx, and an endometrial biopsy was performed concomitantly to rule out CE. In the C group, an endometrial biopsy was performed in the clinic. CE diagnosis was confirmed using immunohistochemistry.

Measurements and Main Results: Our primary endpoint was the rate of positive biopsies for CE. Ninety-four patients were included, 62 in the H group and 32 in the C group. Mean age was 32.1 ± 5.1 years. The prevalence of CE was significantly higher in the H group compared to the C group (41.9% (26/62) vs 15.6% (5/32) (p = .01)). Multivariate analysis showed that the presence of hydrosalpinx was an independent risk factor of CE (aOR = 3.93 (1.31−11.81)), whether the hydrosalpinx was unilateral (aOR = 4.39 (1.32−14.61)) or bilateral (aOR = 3.52 (1.01−11.99)).

Conclusions: There is a significant increase in the prevalence of CE in women with hydrosalpinx undergoing IVF, whether the hydrosalpinx was unilateral or bilateral. Journal of Minimally Invasive Gynecology (2025) 00, 1−7.

Key words: Chronic endometritis; Hydrosalpinx; Infertility

Introduction

Chronic endometritis (CE) is suspected to have a negative impact on endometrial receptivity [1−5]. The diagnosis of CE is confirmed histologically, by the presence of plasma cells in the endometrial tissue [6]. The use of immunohistochemistry (IHC) has significantly improved the precision of CE diagnosis, through the use of a specific surface marker of plasma cells: CD-138, or syndecan-1 [7]. The probability of finding plasma cells in the endometrial tissue is significantly higher when the biopsy is performed in the follicular phase, when compared to a biopsy in the luteal phase [8].

The main cause of CE seems to be an intra-uterine bacterial infection [9] or an endometrial dysbiosis [10−11].

Prior Presentation: This work was presented in poster form at ESHRE annual meeting, Copenhagen, 2023.

IRB Approval: This study was approved by Montreal and Angers institutional review boards in 2017 (2017−46).

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Indeed, positive bacterial cultures were reported in 75% of endometrial samples taken from 388 women with CE (mainly *Streptococcus, Enterococcus faecalis, Escherichia coli* and *Mycoplasma/Ureaplasma*) [9]. Moreover, a significantly lower abundance of Lactobacillus crispatus, and a significantly higher abundance of 18 non-Lactobacillus taxa (Dialister, Gardnerella or Prevotella) were found in the endometrial cavity of 12 women with CE, when compared to women without CE [10]. On the other hand, almost 25% of CE seem to be linked to a "chronic inflammatory state of the endometrium" [10].

Hydrosalpinx is a dilation of the fallopian tube by a serous liquid, most commonly secondary to a gynecologic pelvic infection [12]. In the early 90's, several studies have confirmed the negative impact of hydrosalpinges on embryo implantation and pregnancy rates following in vitro fertilization (IVF) [13−14]. This is why a salpingectomy, or the proximal occlusion of a dilated fallopian tube, are indicated before any IVF treatment in women with hydrosalpinx [15]. Both have been shown to significantly improve live birth rates following IVF [15]. The flux of the hydrosalpinx fluid into the uterine cavity is suspected to have a negative impact on embryo development, and on endometrial receptivity [15−17]. Several studies have shown that the hydrosalpinx fluid is filled with microorganisms, tissue debris, lymphocytes and cytokines, which could all negatively impact embryo development and endometrial receptivity [15−17]. Other studies have shown that hydrosalpinges are associated with a dysregulation in the mRNA expression of proteins and cytokines involved in the inflammatory process (Homebox Protein A10, Interleukin-2, Tumor Necrosis Factor-α) [18−19]. However, the exact mechanism by which a hydrosalpinx alters the embryo implantation rates in IVF is yet to be fully understood.

Both chronic endometritis and hydrosalpinx are associated with embryo implantation failures, via several pathophysiological mechanisms, including infection and inflammation. However, very few studies have looked at the potential association between these two entities [20−22], and these studies have suffered either from low inclusion numbers [20,22], or methodological biases [21], thus making it difficult to assert any conclusion. We believe it is essential to determine whether chronic endometritis is one of the mechanisms by which hydrosalpinges decrease embryo implantation rates, since it is very easily diagnosed with an endometrial biopsy and can be treated accordingly. Our hypothesis was that there is a pathological link between hydrosalpinx and CE that could be behind the embryo implantation failure. Therefore, the main objective of our study was to compare the prevalence of CE in women with hydrosalpinx undergoing IVF to a control group without hydrosalpinx. Our secondary objectives were to assess the impact of a unilateral or bilateral hydrosalpinx on the occurrence of CE, and to compare the live birth rate (defined as the birth of a viable baby >25 weeks gestational age) between women treated for CE and women without CE in the H group.

Material and Methods

Patients

We undertook a retrospective case-control study at the Angers and Montreal university hospital, between June 2017 December 2021.

The hydrosalpinx group (H group) included women undergoing IVF for various indications who were diagnosed with hydrosalpinx before or during cycles. The control group (C) included women undergoing IVF for male factor infertility.

In the H group, women were included retrospectively at the Montreal University Hospital, and at the Angers University Hospital. In the C group, women were included at Angers university Hospital.

All patients had a baseline infertility workup that included day 3 hormonal workup, transvaginal ultrasound for antral follicle count (AFC) and evaluation of the uterine cavity, and a hysterosalpingography or hysterosalpingofoamsonography. Hydrosalpinx was diagnosed, either by an hysterosalpingography, or transvaginal ultrasound. In the H group, whenever a hydrosalpinx (unilateral or bilateral) was diagnosed, surgery was scheduled for either salpingectomy or proximal tubal occlusion before IVF. Inclusion criteria in the group H were: (i) known history of infertility, (ii) indication for IVF management, (iii) decision to undergo surgery for hydrosalpinx, (iiii) endometrial biopsy during surgery with immunohistochemical analysis of endometrial tissue. Patient selection was random.

Women included in the C group were already included as the control group in another trial performed by the Angers University Hospital (NCT03690830), and all had endometrial biopsies. The inclusion criteria were: (i) women with no previous medical or surgical history related to infertility, (ii) women undergoing IVF for male factor infertility (oligoasthenospermia, azoospermia, anejaculation), (iii) Women undergoing IVF following bilateral tubal ligation, (iiii) women with normal workup prior to IVF.

We excluded from the study all patients who refused the endometrial biopsy or did not sign the consent form, patients with uninterpretable biopsies due to insufficient tissue, patients with a history of antibiotic treatment in the month preceding the biopsy, patients with uterine abnormalities, patients with unexplained uterine bleeding, and patients with a positive pregnancy test prior to surgery.

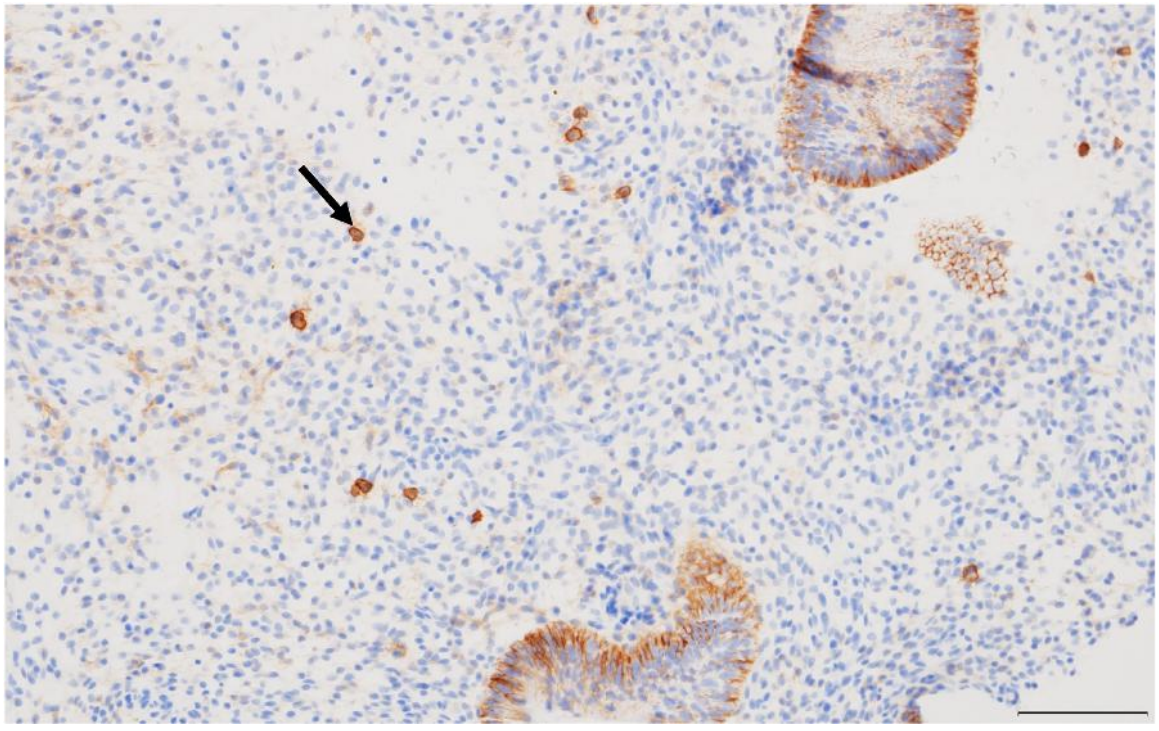
Recurrent pregnancy loss (RPL) and recurrent implantation failure (RIF) have both been linked with CE (3-5]. Therefore, we excluded from our study all patients with a history of RIF or RPL.

Procedure

In the H group, the laparoscopy and endometrial biopsy procedures were performed simultaneously. A vaginal speculum was placed, the cervix was gripped with *a Pozzi*

Fig. 1

Identification of endometrial plasma cells (brown staining) by immunostaining for syndecan-1 (CD-138) in endometrial stroma. In the diagnostic counting, the weaker stromal staining was not included by the pathologists, who only included isolated cells with intense and complete membrane stating in the connective tissue. The discrete staining in the connective tissue as well as endometrial glands staining was not included.



tenaculum and a pipelle (Pipelle de Cornier, CCD) was inserted under visual control into the uterine cavity.

The laparoscopy started with a full evaluation of the abdominopelvic cavity. Chromoperturbation was performed to verify the diagnosis and evaluate the contralateral tube. Salpingectomy was the first-choice treatment, but proximal tubal occlusion was performed for complicated cases with severe pelvic adhesions. All operations were carried out in the follicular phase of the menstrual cycle, and programmed as follows: in women with regular menstrual cycles, the date of the expected menses was calculated based on the mean cycle's length, and the surgery was scheduled between day 5 and 14 of the coming cycle. In women with irregular cycles, treatment with oral dydrogesterone was started between day 16 and 25 in order to regularize it and allow for withdrawal bleeding. On the first day of the withdrawal bleeding, which usually occurred a few days after stopping dydrogesterone, the patient called the physician's office and the surgery was programmed between day 5 and 14 of the coming cycle.

In group C, the endometrial biopsy was performed in the clinic, using the same pipelle as the H group, during the follicular phase of the menstrual cycle.

Immunohistochemical staining was performed on each biopsy, by incubation with a 1:200 dilution of mouse monoclonal antibodies directed against syndecan-1, a specific marker of plasma cells (Agilent Technologies, France) for 20 minutes at room temperature. In both centers, all slides were stained with CD138, and the same anti-CD138 antibodies were used. The diagnosis of CE was considered positive if five or more plasma cells were observed in the endometrial tissue samples on 10 nonoverlapping high-power fields (Fig. 1).

Statistical Analysis

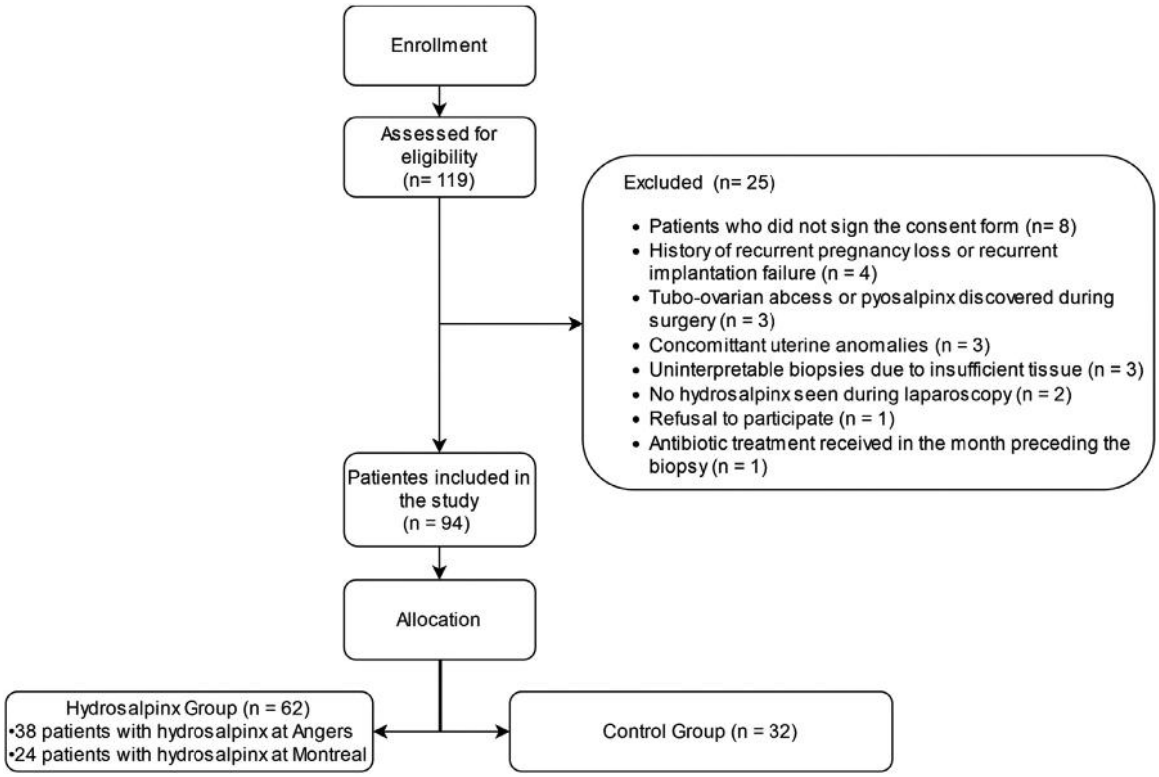
Our primary endpoint was the rate of positive biopsies for CE.

Our power calculations showed that a minimal sample size of 60 patients (30 in each group) was required to show a 30% difference between the C and H groups (10% vs 40%, respectively), with a 80% power, an α risk of 5%. Indeed, based on the available literature [10], we estimated the prevalence of CE to be 10% in a general population of women with infertility, such as those included in group C. On the other hand, taking into account that a hydrosalpinx could decrease the embryo implantation rate, we estimated a prevalence of CE of 40% in women with hydrosalpinx (H group), similar to that found in women with RIF [3].

Qualitative variables were expressed as numbers and percentages, and compared using the Pearson chi-squared. Quantitative variables were expressed as means and

Fig. 2

Study flowchart.



standard deviations and compared using Student's t test. For the primary outcome, the relative risk between the group (H/C) and the presence of CE was calculated with its confidence interval. A logistic regression multivariate analysis taking into account potential confounding factors (age, body mass index, endometriosis, smoking) was performed. All analyses were performed using SPSS version 22.0 (New York, USA). A p-value $< .05$ was considered statistically significant.

Ethical Approval

The study was approved by the Montreal and Angers institutional review boards, and all patients signed an informed consent prior to inclusion.

Results

The study flow chart is shown in Fig. 2. In total, 94 patients were included, 62 in the H group and 32 in the C group.

Population Characteristics and Surgical Details

Patients' characteristics are in Table 1. All women in the H group (62/62) had a diagnosis of hydrosalpinx made either by vaginal ultrasound (69.4%) or hysterosalpingography (30.6%)

The characteristics of patients with CE (n = 32) were comparable to patients without CE (n-62) (Table 2) .

Intraoperative findings are listed in Table 3. 13 patients out of the 62 in the H group had endometriosis (20.6%). In this subgroup of patients, there were 3 cases of CE (23.1%). In the sub-group of patients with hydrosalpinx and without endometriosis (n = 49), there were 23 cases of CE (46.9%). There was no significant difference in the prevalence of CE between these two subgroups (p = .12).

Outcomes

The prevalence of CE was significantly higher in the H group compared to the C group (41.9% (26/62) vs 15.6% (5/32) (p = .01)). The prevalence of CE was comparable between the women included in Angers and those in Montreal (47% (18/38) vs 33% (8/24), p = .28, respectively). All patients with CE received antibiotic treatment.

The association between hydrosalpinx and CE was assessed in a univariate and multivariate analysis (Table 4).

Finally, in the H group, following surgery, the live birth rate (LBR) at 24 months was comparable between women with CE who were treated and women without CE (44% (11/25) vs 61% (22/36), p = .19)). It is worth noting that one patient with CE in the H group was lost to follow-up.

Table 1

Baseline characteristics of patients

	Overall (N = 94)	Group H (N = 62)	Group C (N = 32)	p-value
Age (years)	32.1 ± 5.1	32.4 ± 5.2	31.5 ± 4.8	.44
Body Mass Index (kg/m^2)	24.5 ± 4.5	24.0 ± 4.3	24.5 ± 4.7	.91
Primary Infertility	59 (62.0)	37 (8.7)	22 (68.7)	.38
Positive history of one pregnancy loss	15 (15.8)	10 (15.8)	5 (15.6)	1
History of ectopic pregnancy	9 (9.5)	9 (14.3)	0	.03
History of curettage	8 (8.4)	6 (9.5)	2 (6.2)	.71
Associated infertility factors:				
Endometriosis	13 (14)	13 (20.6)	0	< .01
Polycystic ovarian syndrome	12 (12.6)	12 (19.0)	0	< .01
Low ovarian reserve	10 (10.5)	10 (15.9)	0	.01
Myomas	2 (2.1)	2 (3.2)	0	.55
Polyps	1 (1.0)	1 (1.6)	0	1
Severe sperm abnormalities	35 (37.0)	5 (8.0)	30 (94.0)	< .01
History of tubal ligation	2 (2.0)	0	2 (6.0)	.23
History of tubal surgery	24 (25.2)	24 (38.1)	0	< .001
History of pelvic inflammatory disease	31 (32.6)	31 (49.2)	0	< .001
Antral follicle count	21.9 ± 12.8	21 ± 14	23.9 ± 9.9	.22
Baseline AMH (ng/ml)	3.62 ± 2.3	2.7 ± 1.7	3.42 ± 1.6	.32
Baseline FSH (mUI/ml)	7.5 ± 3.3	7.7 ± 5.7	7.2 ± 1.8	.52
Positive CE	31 (33.0)	26 (41.9)	5 (15.6)	.01

Data expressed as mean ± standard deviation or n (%).

Table 2

Baseline characteristics of patients with or without chronic endometritis

	Overall (N = 94)	Group CE- (N = 63)	Group CE+ (N = 31)	p-value
Age (years)	32.1 ± 5.1	31.6 ± 4.6	33.6 ± 5.6	.07
Body Mass Index (kg/m^2)	24.5 ± 4.5	24.4 ± 4.5	24.6 ± 4.5	.91
Primary Infertility	59 (62.0)	43 (68.3)	16 (51.6)	.12
Positive history of one pregnancy loss	15 (15.8)	9 (14.3)	6 (19.4)	.52
History of ectopic pregnancy	9 (9.5)	7 (11.1)	2 (6.5)	.71
History of curettage	8 (8.4)	3 (4.8)	5 (16.1)	.11
Associated infertility factors:				
Endometriosis	13 (14)	10 (15.9)	3 (9.7)	.53
Polycystic ovarian syndrome	12 (12.6)	8 (12.7)	4 (12.9)	.98
Low ovarian reserve	10 (10.5)	8 (12.7)	2 (6.5)	.36
Myomas	2 (2.1)	2 (3.2)	0	1
Polyps	1 (1.0)	0	1 (3.2)	.33
Severe sperm abnormalities	35 (37.0)	26 (41.3)	9 (29)	.25
History of tubal ligation	2 (2.0)	2 (3.2)	0	1
History of tubal surgery	24 (25.2)	14 (22.2)	10 (32.3)	.29
History of pelvic inflammatory disease	31 (32.6)	18 (28.6)	13 (41.9)	.20
Antral follicle count	21.9 ± 12.8	21.5 ± 10.8	22.3 ± 16.2	.84
Baseline AMH (ng/ml)	3.6 ± 2.3	3.3 ± 2	3.3 ± 2.8	.77
Baseline FSH (mUI/ml)	7.5 ± 3.3	7 ± 2.2	8.1 ± 4.1	.49

Data expressed as mean ± standard deviation or n (%).

Discussion

Our study found a high prevalence (42%) of CE in women with hydrosalpinx undergoing IVF, as well as a correlation between hydrosalpinx and CE (aOR=3.93 (1.31−11.81)).

A recent monocentric prospective cohort study looked at the association between CE and tubal infertility [20]. They included 100 women who had a hysteroscopy with endometrial biopsy and a laparoscopy. Among the 100 included, only 9 had a hydrosalpinx and 13 had CE. Among the nine

Table 3

Surgical details

	Groupe H (n = 62)
Intraoperative findings	
Unilateral hydrosalpinx	34 (54.8)
Bilateral hydrosalpinx	25 (40.3)
Tubes not visualized (dense pelvic adhesions)	3 (4.9)
Dye hydrotubation	
Unilateral permeability	40 (64.5)
Bilateral permeability	0 (0)
No permeability	22 (35.5)
Operative treatment	
Unilateral salpingectomy	33 (53.3)
Bilateral salpingectomy	24 (38.7)
Unilateral salpingectomy and contralateral neosalpingostomy	2 (3.2)
Unilateral Filshie clips	1 (1.6)
Bilateral Filshie clips	2 (3.2)
Concomitant hysteroscopies	46 (74.2)
Concomitant endometrial biopsies	62 (100)

Data expressed as n (%).

women with hydrosalpinx, two (22%) had CE and seven (88%) did not (p = .6) [20].

Another larger retrospective study looking at the association between CE and hydrosalpinx was published in 2022 [21]. All included women had a laparoscopy and a hysteroscopy with an endometrial biopsy for the diagnosis of CE. 624 women with hydrosalpinx were included in group A, and compared to 789 women without hydrosalpinx in group B. The prevalence of CE was 21% in group A compared to 14% in group B (p < .0001) [21].

More recently, in a small study including 55 women, Zou *et al.* [22] reported a correlation between the presence of hydrosalpinx and CE. In a group of women with unilateral hydrosalpinx (n=10), 8 women had CE and 2 did not (aOR 7.84, CI95% 1.28−48.09). In the group of women with bilateral hydrosalpinx (n=10), 9 had CE and one did not (aOR 9.45, CI95% 1.04−86.15).

Based on our results, we can establish that there is an association between hydrosalpinx and CE. We hypothesize that the chronic inflammation and refluxed fluid from the hydrosalpinx into the endometrial cavity may initiate the endometrial inflammation, leading to CE, which in turn could worsen the hydrosalpinx. We therefore believe that screening for CE and treating it should be part of the management of patients with hydrosalpinx undergoing IVF. Finally, even though our study included only patients undergoing IVF, and based on studies associating CE with infertility, we believe the same management could be offered for all infertile patients in whom a hydrosalpinx is diagnosed during the initial work-up, and not only those undergoing IVF. However, the pathological link between hydrosalpinx and CE is far from being fully understood, and requires further analysis with different approaches. For instance, if we believe CE to be the consequence of hydrosalpinx − the chronic inflammation caused by the continuous fluid spilling from the tube to the uterine cavity − it would be interesting to investigate whether CE persists or resolves spontaneously, without any antibiotic treatment, 2 or 3 months following hydrosalpinx surgery, which completely stops the endometrial exposure to the hydrosalpinx fluid. On the other hand, if we believe CE to be the cause of hydrosalpinx − ascending inflammation from the uterine cavity to the tubes- it would be interesting to assess the cure rate of CE, and the total disappearance of hydrosalpinx following antibiotic therapy, without any surgical intervention. Such a hypothesis would be difficult to test in a study in the context of infertility and IVF, given the significant reduction in IVF outcomes in women with hydrosalpinx [13−15].

The main limitations of our study are (i) the retrospective inclusion of the cohort; (ii) the confidence interval for the odds ratio and adjusted odds ratio are wide. With a hundred patients, the confidence interval for a binary factor (hydrosalpinx yes/no) is relatively imprecise and several hundred patients would be needed to obtain better precision. In general, for a binary factor, the p-value predominates; (iii) the fact that in 16 cases (25.8%) there were no hysteroscopy associated with biopsy which could have increased the detection rate of endometritis. Indeed, studies have demonstrated that there would be benefit in performing targeted biopsies under hysteroscopic control to increase the detection rate of chronic endometritis [23−24]. Nevertheless, there is no consensus on the optimal technique for diagnosing chronic endometritis, especially as there is no consensus on the histological definition. As we

Table 4

Association between chronic endometritis and unilateral or bilateral hydrosalpinx

Outcome (Model)	Global population H group (N=62)		Unilateral Hydrosalpinx (N=35)		Bilateral Hydrosalpinx (N=27)	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Chronic endometritis Univariate analysis	4.05 (1.38−11.89)	.011	4.26 (1.32−13.73)	.015	3.81 (1.14−12.74)	.030
Chronic endometritis Multivariate analysis (adjusted OR)	3.93 (1.31−11.81)	.015	4.39 (1.32−14.61)	.016	3.52 (1.01−11.99)	.041

OR, odds ratio; aOR, adjusted odds ration; CI, confidence interval.

have showed in a previous study [4], hysteroscopy alone has a low sensitivity of 40% and a specificity of 80% for the diagnosis of CE. A following study confirmed these results, showing a sensitivity of 59%, a specificity of 70%, and a diagnostic accuracy of 67% [25].

The strengths of our study are the bicentric design, and the inclusion of a large sample size despite using restrictive inclusion criteria for an already infrequent pathology, even in women with infertility undergoing IVF.

Conclusion

Our study showed a high prevalence of CE in patients diagnosed with hydrosalpinx, whether unilateral or bilateral. Larger studies are needed to confirm our findings, and to further elucidate the correlation between the hydrosalpinx fluid and endometrial bacteria and the need for a tailored treatment.

Declaration of competing interest

The authors declare no conflict of interest related to the present study.

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