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Effect of Antibiotic Treatment of Chronic Endometritis on Pregnancy Rate in Patients with Unexplained Infertility in Suez Canal University Hospital

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Abstract:

Background: Chronic endometritis (CE) is an inflammatory disorder characterized by the presence of plasma cells in the endometrium's stroma. Aim: to improve the pregnancy rate in women with unexplained infertility after antibiotic treatment of chronic endometritis at Suez Canal University Hospital.

Patients and methods: This quasi-experimental research was performed on 27 women of childbearing period presenting at the outpatient clinic of the Department of Obstetrics and Gynecology diagnosed with unexplained infertility, primary or secondary, from November 2019 to September 2022. According to the positive diagnosis of chronic endometritis in the study population, we divided them into 3 groups: Group I: 11 patients with an initial diagnosis negative for histology and C&S; Group II: 12 patients with a diagnosis negative for histology and C&S after treatment; and Group III: 4 patients with a diagnosis positive for histology or C&S.

Results: Endometrial culture was positive in 12 females (75%) and negative in 4 (25%) in women with histological CE; the cure rate was 83% in cases with positive cultures and 50% in cases with negative cultures & sensitivity. In Group I, there were 4 spontaneous pregnancies (36.4%). In Group II, we observe 10 spontaneous pregnancies (83.3%). In Group III, 1 patient (25%) got pregnant naturally.

Conclusion: Chronic endometritis is commonly found in cases of unexplained infertility. The identification and management of chronic endometritis enhances the rate of spontaneous pregnancy in affected cases. Hysteroscopy, known for its great sensitivity, is an appropriate method for diagnosing CE in those suffering unexplained infertility.

Keywords: CE, Unexplained infertility, Hysteroscopy

1. Introduction

CE is a persistent inflammatory disorder characterized by the presence of plasma cells in the stroma of the endometrium. Chronic endometritis is a subtle condition that is frequently disregarded by medical professionals due to its limited symptomatology, which includes uterine hemorrhage, dyspareunia, pelvic discomfort, and leucorrhea [(1)].

The diagnosis of chronic endometritis is difficult. Clinical and ultrasound investigations lack specificity, while histological analysis can be misleading due to the frequent presence of leukocytes in the uterine lining, mostly prior to menstruation [(2)]. Fluid hysteroscopy can assist in identifying signs of CE by facilitating the identification of distinct indicators of CE, including micropolyps, stromal edoema, and localized or widespread hyperemia. Subsequently, the diagnosis was confirmed through histological investigation, which revealed the presence of one to five plasma cells per high-power field [(3)].

The changes in the endometrial milieu identified in CE may impede endometrial receptivity, potentially leading to female infertility [(4)]. However, previous investigations have consistently demonstrated that CE is very common in cases of infertility, particularly in cases of repeated IVF failures (RIF), with a prevalence rate as high as 57.55% [(5)].

Furthermore, various investigations have shown that antibiotic treatment can effectively restore the normal structure of the endometrium and enhance the rate at which babies successfully implant in cases of chronic endometritis. This suggests a causal relationship among CE and impaired receptivity of the endometrium [(6)]. While the associations among CE and RIF have been fully examined, there is currently a lack of sufficient research that has addressed the influence of CE on "unexplained infertility" (UI).

Couples with unexplained infertility have challenges in both their ability to conceive and the timing of their fertility. Upon reviewing studies on unexplained infertility, it was found that the average rate of successful conception per menstrual cycle was 1.8 percent in eleven nonrandomized research studies and 3.8 percent in six randomized trials [(7)].

If there is no recognized correctable defect, the treatment for unexplained infertility is typically empirical. Recommended therapy options involve intrauterine insemination (IUI), ovulation induction using oral or injectable medicines, a combination of IUI with assisted reproductive technologies (ART), and ovulation induction [(8)].

The aim of this work was to improve the rate of pregnancy in women with unexplained infertility after antibiotic treatment of chronic endometritis at Suez Canal University Hospital.

2. Patients and methods

This quasi-experimental research was performed on 27 women of childbearing period presenting at the outpatient clinic of the Department of Obstetrics and Gynecology diagnosed with unexplained infertility, primary or secondary, from November 2019 to September 2022. According to the positive diagnosis of chronic endometritis in the study population, we divided them into 3 groups: Group I: 11 patients with an initial diagnosis negative for histology and

C&S; Group II: 12 patients with a diagnosis negative for histology and C&S after treatment; and Group III: 4 patients with a diagnosis positive for histology or C&S.

Inclusion criteria: from eighteen to forty years, presenting with 1ry or 2ry infertility following regular intercourse for a minimum of 1 year, history of infertility for more than 1 year, all participants fulfilled the diagnosis of unexplained infertility: The lack of any anomalies was observed during the tubal patency test, with at least one fallopian tube being patent. The regularity of ovulatory cycles was confirmed through the assessment of mid-luteal progesterone levels (>30 nmol/L) and folliculometry using trans-vaginal ultrasound. Additionally, the partner's semen analysis met the standards established by the World Health Organization and was found to be normal. [(9)]

Exclusion criteria: Diminished ovarian reserve is indicated by a follicle-stimulating hormone level above 10 mUI/mL on day three and an antral follicle count of five to seven or less. Additionally, a body mass index greater than thirty kilograms per square meter is also a factor, **as** is the uterine factor of infertility, as are uterine fibroids, polyps, scar tissue, radiation damage (from cancer treatment) or injuries, previous myomectomy, autoimmune diseases, thrombophilic conditions, the presence of co-morbidities such as ischemic heart disease, chronic hypertension, diabetes mellitus, and the refusal of participants.

Methods:

Microbiological analysis: The existence of microorganisms was assessed by plating the endometrial specimens into the appropriate agar medium, including five percent sheep blood Columbia Chocolate Agar, Agar Base, Mannitol Salt Agar, and Mac Conkey Agar (Bio Merieux, Rome, Italy), after they were Gram-negative stained in the laboratory. The plates were incubated in either air or five percent carbon dioxide for forty-eight hours. Bacteria were identified by employing published criteria (Dade International, Inc., Milan, Italy). Quantitative detection of genital mycoplasmas was achieved through immunoassay (Mycoplasma-IST, Biomerieux, and Rome, Italy). RT-PCR was implemented to evaluate Chlamydia. [**(10)**].

Histopathological analysis: The endometrial samples were preserved in neutral formalin, embedded within paraffin, and then subjected to histological examination using hematoxylin-eosin staining. The diagnosis of CE was established using criteria that have been previously outlined [(11)]. The following traits were observed: superficial stromal edema, heightened stromal density, and pleomorphic stromal inflammatory infiltration, primarily consisting of lymphocytes and plasma cells.

Antibiotic treatment: Ciprofloxacin, five hundred milligrams was commonly used as the initial treatment for a duration of ten days in most instances when Gram-negative bacteria were present. For Gram-positive bacteria, a prescription of Amoxicillin + Clavulanate, one gramme twice a day for ten days, was given [**(12)**]. Females with negative cultures were treated with a single dose of Ceftriaxone 250 milligrams IM, along with Doxycycline one hundred milligrams by mouth twice a day for fourteen days, and Metronidazole 500 milligram orally twice a day for fourteen days, as per the guidelines provided by the Centers for Disease Control [**(13)**]. If

symptoms of CE continued to be present during subsequent hysteroscopy, the treatment was repeated a maximum of 3 times.

Tools used in our study: Transvaginal ultrasound using a Mindray ultrasound machine, model DC-30, 240-270V, 50/60Hz, 630VA (Mindray, China). Patients were examined at the 9th, 11th, and 13th days of their cycle to evaluate ovulatory function and diagnose any uterine abnormalities. Prior to the operation, cases were directed to void their bladders. During the procedure, the transducer tip was coated with ultrasound coupling gel and put into the vaginal canal. A three millimeters Novak's curette, attached to a twenty milliliters syringe, was utilized to get an endometrial biopsy during the follicular phase of the following menstrual cycle. The goal of this procedure was to examine the tissue for histological analysis and to culture it to identify any potential infectious agents.

Technique of hysteroscope:

Anesthesia: consultation with the anesthesia team for all patients was done and they asked for CBC and PT for all patients, which was normal. Type of anesthesia: general anesthesia.

Operative procedure:

The cervix was softened prior to operation by administering misoprostol vaginally, following the standard preoperative preparation protocol. The laparoscopic tower consists of the following components: a light source (Xenon, Germany), a TV monitor (Neovo, China), a hysteromatte (Karl Storz, Germany), an insufflator (Karl Storz, Germany), a video recorder (Sony, Japan), an endoscopy camera (Karl Storz, Germany), and an electrosurgical generator (The Valleylab Force, Germany). The case was placed in a lithotomy position and had general anesthesia with endotracheal intubation. The vulva was sterilized, and cloths were used to create a sterile field. Both legs were supported during the procedure. Urinary bladder emptying is achieved by inserting a metal catheter into the urethra. An examination conducted while the patient is under general anesthesia to assess the location of the uterus (either AVF or RVF) and a manual inspection of the adnexa. Holding the cervix using a multi-toothed volsullum. The examination of the uterine cavity involved a thorough assessment of the endometrial mucosa, preceded by a panoramic view of the cavity. The diagnosis of CE in all female participants was established by identifying polypoid endometrium, micro-polyps, stromal edema, and focal or diffuse hyperemia, as previously reported. [(3)].

Post-operative care:

The study observed participants in the hospital ward for up to 6 hours' post-surgery to check for any surgical or anesthesia-related complications, and no such issues were recorded.

Ethical considerations: The research protocol underwent revision and received approval from the Research Ethics Committee of the faculty of medicine at Suez Canal University. Explicit written consent was obtained in every case.

3. Results

Table 1 shows that the mean age of the research group was 29.89 ± 3.73 years, the mean partner age was 34.52 ± 3.93, and the mean BMI was 26.81 ± 1.80.

Table 2 shows that in 9 women (33.3%), hysteroscopy was negative for chronic endometritis; correspondingly, histology was negative. In contrast, 18 females (66.7%) were diagnosed with chronic endometritis at hysteroscopy (p value <0.001) and 16 (59.3%) at histology.

Table 3 shows that the culture of endometrial was positive in 12 females (75%) & negative in 4 (25%) in women with histological CE. The cure rate was 83% in cases with positive cultures and 50% in cases with negative cultures & sensitivity.

Table 4 shows a plethora of microorganisms, involving frequent bacteria (E. Faecalis, E. Coli), Mycoplasma, and Chlamydia, with no statistically significant difference in their prevalence.

Table 5 shows that in Group I, "patients with an initial diagnosis negative for histology and C&S," there were 4 spontaneous pregnancies (36.4%). In Group II, "Patients with a negative diagnosis of histology and C&S after treatment," we observe 10 spontaneous pregnancies (83.3%). In Group III, "patients with a diagnosis positive for histology or C&S" after treatment, 1 patient (25%) got pregnant naturally.

Table (1): Comparison among all groups of study corresponding to demographic data

	Total (n= 27)	**Group I (n = 11)**	**Group II (n = 12)**	**Group III (n = 4)**	**F**	**P**
Age						
Min. – Max.	23.0 – 36.0	23.0 – 36.0	24.0 – 33.0	27.0 – 35.0		
Mean ± SD.	29.89 ± 3.73	31.82 ± 3.84	28.08 ± 2.97	30.0 ± 3.56		
Median (IQR)	30.0 (27.50 – 33.0)	33.0 (30.0 – 34.5)	28.0 (25.0 – 30.0)	29.0 (27.5 – 32.5)	3.402*	0.049*
Sig. bet. grps.		**p_1=0.040*,p_2=0.641,p_3=0.604**				
Partner age						
Min. – Max.	28.0 – 40.0	28.0 – 40.0	28.0 – 36.0	33.0 – 40.0		
Mean ± SD.	34.52 ± 3.93	36.09 ± 3.96	32.25 ± 2.90	37.0 ± 3.56		
Median (IQR)	35.0 (30.5 – 37.5)	37.0 (35.0 – 39.0)	32.0 (30.0 – 35.0)	37.5 (34.0 – 40.0)	4.746*	0.018*
Sig. bet. grps.		**p_1=0.035*,p_2=0.895,p_3=0.064**				
BMI						
Min. – Max.	23.0 – 30.0	24.0 – 30.0	23.0 – 28.0	27.0 – 29.0		
Mean ± SD.	26.81 ± 1.80	27.36 ± 1.80	25.83 ± 1.53	28.25 ± 0.96		
Median (IQR)	27.0	28.0	26.0	28.5	4.549*	0.021*

	(25.50 – 28.0)	(26.5 – 28.5)	(25.0 – 27.0)	(27.5 – 29.0)		
Sig. bet. grps.		**p_1=0.075,p_2=0.613,p_3=0.038***				

SD: Standard deviation, IQR: Inter quartile range, pairwise comparison bet. each 2 groups were done using Post Hoc Test (Tukey), F: F for One-way ANOVA test,
p: p value for comparing between the three studied groups
p_1: p value for comparing between Group I and Group II
p_2: p value for comparing between Group I and Group III
p_3: p value for comparing between Group II and Group III, *: Statistically significant at $p \leq 0.05$

Table (2): Comparison among all groups of study corresponding to hysteroscopy:

	Total (n = 27)		**Group I (n = 11)**		**Group II (n = 12)**		**Group III (n = 4)**		X^2	**MCp**
	No.	**%**	**No.**	**%**	**No.**	**%**	**No.**	**%**		
Hysteroscopy										
Negative	9	33.3	9	81.8	0	0.0	0	0.0		
Positive	18	66.7	2	18.2	12	100.0	4	100.0	19.065*	<0.001*

X^2: Chi square test, MC: Monte Carlo,

Table (3): Distribution of all cases corresponding to histology and C&S in each group

		Total (n = 27)		**Group I (n = 11)**		**Group II (n = 12)**		**Group III (n = 4)**	
		No.	**%**	**No.**	**%**	**No.**	**%**	**No.**	**%**
Pre	**Histology**								
	Negative	11	40.7	11	100.0	0	0.0	0	0.0
	Positive	16	59.3	0	0.0	12	100.0	4	100.0
	C&S								
	Negative	15	55.6	11	100.0	3	25	1	25.0
	Positive	12	44.4	0	0.0	9	75	3	75.0
Post	**Histology**								
	Negative	12	44.4	0	0.0	12	100.0	0	0.0
	Positive	4	14.8	0	0.0	0	0.0	4	100.0
	Not done	11	40.7	11	100.0	0	0.0	0	0.0
	C&S								
	Negative	14	51.9	0	0.0	12	100.0	2	50.0

	Positive	2	7.4	0	0.0	0	0.0	2	50.0
	Not done	11	40.7	11	100.0	0	0.0	0	0.0

Table (4): Comparison among all groups of study corresponding to organism

	Total (n = 27)		**Group I (n = 11)**		**Group II (n = 12)**		**Group III (n = 4)**			
	No.	**%**	**No.**	**%**	**No.**	**%**	**No.**	**%**	**c2**	**MCp**
Organism										
Negative	15	55.6	11	100.0	3	25.0	1	25.0	15.931*	<0.001*
E. coli	4	14.8	0	0.0	3	25.0	1	25.0	3.477	0.223
Entrococeus fecalis	4	14.8	0	0.0	3	25.0	1	25.0	3.477	0.223
Chlamydia	2	7.4	0	0.0	2	16.7	0	0.0	2.053	0.628
Mycoplasma	2	7.4	0	0.0	1	8.3	1	25.0	2.689	0.279

Table (5): Comparison among all groups of study corresponding to pregnancy

	Total (n = 27)		**Group I (n = 11)**		**Group II (n = 12)**		**Group III (n = 4)**			
	No.	**%**	**No.**	**%**	**No.**	**%**	**No.**	**%**	**X^2**	**MCp**
Pregnancy										
Negative	12	44.4	7	63.6	2	16.7	3	75.0		
Positive	15	55.6	4	36.4	10	83.3	1	25.0	6740*	0.028*

4. Discussion

Around thirty percent of couples who are unable to conceive are diagnosed with unexplained infertility [(4)]. Hence, unexplained infertility seems to be either the most uncommon kind of normal fertility or it stems from an undetectable flaw in reproductive capacity that is not

identified through standard infertility assessments. Couples with unexplained infertility have challenges in both their ability to conceive and the timing of their fertility. Upon reviewing investigations on unexplained infertility, it was found that the average rate of successful conception per menstrual cycle was 1.8 percent in eleven nonrandomized investigations and 3.8 percent in six randomized research for the untreated control groups. [(4)].

The mean age of the study group was 29.89 ± 3.73 years, the mean partner age was 34.52 ± 3.93, and the mean BMI was 26.81 ± 1.80. In the present investigation, hysteroscopy was negative for chronic endometritis in nine females (33.3 percent), and as a result, histology was negative. Conversely, eighteen females (66.7 percent) were identified with chronic endometritis during hysteroscopy (p value<0.001), as well as sixteen females (59.3 percent) who were additionally diagnosed with chronic endometritis during histology.

This finding agrees with that reported by **E. Cicinelli Matteo et al.** [(14)] who reported a high prevalence of histological CE (55.7%) among cases of unexplained infertility.

Regarding microbiological findings, our study reported that endometrial culture was positive in 12 females (75%) & negative in 4 (25%) in women with histological CE.

Tersoglio AE, Salatino DR, et al. [(15)] reported that culture and sensitivity were positive in 40 % and negative in 60 % of patients with CE and repeated implantation failure.

Our investigation found that antibiotics resulted in the clearance of CE (confirmed by negative hysteroscopy, histology, & cultures) in ten out of twelve cases (eighty-three percent) with positive cultures. However, in the other two cases, CE persisted after antibiotic treatment. In four cases, hysteroscopy & histology confirmed a positive diagnosis, whereas cultures didn't. As a result, we administered a multidrug treatment, which successfully cured the condition in two out of the two females treated (fifty percent). In the remaining 2-woman, CE was persistent. Of 16 positive cases for histology, in 3 cases (18.7%), signs of chronic endometritis were negative following a single therapy cycle, in 4 cases (25%), 2 treatment cycles, and in 5 cases (31.3%), three treatment cycles.

This discovery is consistent with **E. Cicinelli Matteo et al.** [(14)] reported that the cure rate was 82.3% in patients with CE with positive culture and sensitivity after treatment with a specific antibiotic according to culture & sensitivity. In patients with CE with negative culture and sensitivity, the cure rate was low (52.6%) after treatment with empiric broad-spectrum antibiotic therapy.

Our study discovered a plethora of microorganisms, involving common bacteria (E. coli, E. faecalis), Mycoplasma, and Chlamydia, with no statistically significant difference in their prevalence.

This conclusion is consistent with the research conducted by Cicinelli, **A. Ballini, et al.** **[(16)]**, which also reported a high prevalence of common bacteria in cases with CE. This is in line with the fact that bacterial vaginosis is highly prevalent & the understanding that bacteria can climb and colonize the uterine cavity.

In Group 1, "patients with an initial diagnosis negative for histology and C&S," there were 4 spontaneous pregnancies (36.4%). In Group 2, "Patients with a negative diagnosis of histology and C&S after treatment," we observe 10 spontaneous pregnancies (83.3%). In Group 3, "patients with a diagnosis positive for histology or C&S" after ttt, 1 patient (25%) got pregnant naturally.

These results are consistent with **E. Cicinelli Matteo et al.** [14] reported that spontaneous pregnancy was 76.3% in patients with chronic endometritis after resolution.

Cicinelli E, Matteo M, Tinelli R et al. [5] found that females in the first group, who were treated for CE, had a significantly greater pregnancy rate compared to females in the second group, who had persistent CE (65.2% vs. 33.0%, P = 0.039, respectively) when undergoing IVF therapy.

The main weakness of the current research is the limited number of cases (27 patients). So, we need to conduct a large multi-centre study to confirm these results.

5. Conclusion

Chronic endometritis is commonly found in cases of unexplained infertility. The identification and management of chronic endometritis enhances the rate of spontaneous pregnancy in affected cases. Hysteroscopy, known for its great sensitivity, is an appropriate method for diagnosing CE in those suffering unexplained infertility.

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