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The Relationship between Reproductive Hormones, MFG-E8, and Chlamydial Infection in Infertile Women

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ABSTRACT

Background and Objective: Chlamydia trachomatis (CT) is a common sexually transmitted infection that is often symptomless and can cause long-term complications, including infertility. The objective of this study is to evaluate the relationship between reproductive hormones, MFG-E8 (Milk Fat Globule-Epidermal Growth Factor 8), and chlamydial infection in infertile women.

Methods: This cross-sectional study was conducted in Al-Salama Hospital between November 2021 and February 2023 on 90 participants, 60 infertile patients and 30 fertile individuals as a control group. The participants were divided into four groups based on their chlamydial infection status and serum levels of CT Ig-G, MFG-E8, AMH (Anti-Müllerian Hormone), LH (Luteinizing Hormone), and FSH (Follicle-Stimulating Hormone), Prolactin, and Testosterone were measured and compared using the ELISA technique.

Findings: A notable difference in Ig-G levels was observed based on chlamydia trachomatis infection status of female participants. The infertile group demonstrated significantly higher Ig-G levels, with a mean of 17.22±6.43 in the presence of positive Chlamydial infection, compared to the other groups ($p<0.0001$). In addition, the mean level of MFG-8 in the fertile group with chlamydia positive, at 215.65±26.07 ng/ml, was significantly higher compared to other groups ($p<0.0001$). No statistically significant difference was observed in MFG-E8 level between fertile and infertile patients.

Conclusion: The results of the study revealed a significant link between chlamydia trachomatis infection and infertility. The fertile women with Chlamydia-positive cases exhibited elevated levels of MFG-8, suggesting its potential involvement in reproductive health. These findings contribute to our understanding of the complex relationship between Chlamydia trachomatis infection, reproductive hormones, and infertility.

Keywords: *AMH, MFG-E8, Chlamydia Trachomatis, Fertility Hormone, Infertility.*

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Introduction

Chlamydia trachomatis (CT) is a predominant sexually transmitted pathogen worldwide (1), with over 900,000 reported cases in the US in 2004 and an estimated higher number going unreported (1). Infection with CT can lead to critical consequences, including infertility, with approximately 45% of tubal infertility cases attributed to CT infection. Despite extensive research, the specific factors contributing to infertility caused by CT remain unknown due to limited experimental models and diagnostic tools. Therefore, large-scale screening programs are recommended for high-risk patients, relying on accurate and cost-effective laboratory tests and tools (2, 3).

Milk fat globule-EGF factor 8 protein (MFG-E8), also known as lactadherin, is a glycoprotein that exhibits notable anti-inflammatory properties. MFG-E8 plays a crucial role in the clearance of apoptotic cells, acting as an anti-inflammatory factor by suppressing the transcription of pro-inflammatory cytokines inhibition of neutrophil infiltration into inflamed tissues, indicating its significant role in inflammatory diseases (4-7). Additionally, recent studies have linked MFG-E8 to the process of implantation. It is expressed in the human endometrial tissue and up-regulated during the implantation period (3, 8).

The concentrations of reproductive hormones play a crucial role in female fertility. In clinical practice, the assessment of female fertility during the early follicular phase involves considering several factors, including age, anti-Müllerian hormone (AMH), follicle-stimulating hormone (FSH), luteinizing hormone (LH) levels, prolactin (PRL), and testosterone (T) levels, as well as the patency of the fallopian tubes (9). Nonetheless, the association between these fertility factors and Chlamydia trachomatis (CT) infection has been minimally explored in the existing research literature. Consequently, the objective of this study is to investigate the potential relationship between reproductive hormones and MFG-E8 in infertile patients with CT infection. We seek to explore the possible associations and understand the role of these hormones in chlamydia-related infertility.

Methods

This cross-sectional study was conducted at Al-Salama Private Hospital from November 2021 to February 2023. The study included 90 participants, 60 females who were infertile and 30 females who were fertile as controls. The study received approval from the Ethical Committee of the College of Medicine, Al-Iraqia University, Baghdad, Iraq (Approval No. FM.SA/159). Before they participated in the study, all participants provided written informed consent.

The participants were categorized into four groups based on their CT infection status. Group A included 33 women who were infertile and tested positive for Chlamydia. Group B included 27 infertile women who tested negative for Chlamydia. Group C included 15 fertile women who tested positive for Chlamydia, and Group D included 15 fertile women who tested negative for Chlamydia.

This study included women who did not have diabetes or cardiovascular disease and were not undergoing hormonal therapy. Both infertile patients and control groups provided fasting venous blood samples collected under sterile conditions. Serum was extracted from 1-2cc of blood and the CT Ig-G, MFG-E8, as well as reproductive hormone levels including AMH, LH, FSH, Prolactin (PRL), and Testosterone (T) were measured using the ELISA technique. The Cut-off of Chlamydia trachomatis anti-IgG conjugate is 10 DU. The positive results are determined if the result is greater than ten percent of cut-off, and negative when a result value is less than ten percent of cut-off.

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Data analysis method: The data obtained from the ELISA tests underwent analysis using the SPSS software version 27 for measuring mean and SD. The t-test and ANOVA were employed to examine the potential correlation between groups. Pearson Correlation Coefficient were estimated for all groups
Statistical significance was defined as a p-value<0.05, indicating a significant correlation. A p-value<0.001 was considered statistically significant, indicating a robust and highly reliable correlation between the variables.

Results

The study encompassed 90 participants with a mean age of 29.88±0.49 years, a mean weight of 72.70±3.65 kg, and a mean height of 162.2±1.3cm. No statistically significant variations were observed in the demographic characteristics of female participants, as shown in Table 1. The infertile group exhibited higher mean Ig-G (DU) levels of 17.22±6.43 for positive Chlamydial infection and 8.06±0.97 for negative Chlamydial infection. In contrast, the fertile group had lower mean Ig-G (DU) levels of 15.79±4.31 for positive Chlamydial infection and 7.87±1.07 for negative Chlamydial infection. These findings demonstrate significant disparities in Chlamydial infection status between the infertile and fertile groups (p<0.0001).

The mean level of MFG-8 among infertile patients with positive Chlamydia infection were measured at 192.79±52.16 ng/ml, while infertile patients without Chlamydia infection had levels of 90.24±44.16 ng/ml. In comparison, the fertile group exhibited higher mean Ig-G levels, with 215.65±26.07 ng/ml in Chlamydia-positive cases and 107.83±39.17 ng/ml in Chlamydia-negative individuals. These results indicate significant differences in Chlamydia infection status between infertile and fertile groups (p<0.0001), as shown in Table 2.

Table1. Demographic Characteristics and Chlamydial Infection status of Female Participants:

Demographic characteristics	Infertile group (n=60)		Fertile group (n=30)		p-value
	Chlamydial Positive (n=33)	Chlamydial Negative (n=27)	Chlamydial Positive (n=15)	Chlamydial Negative (n=15)	
Age (years)	29±6.24	28±6.38	32±7.99	29±6.18	0.250
Weight (kg)	73±12.61	73±11.18	71±10.24	73±13.47	0.917
Height (cm)	162±5.65	161±5.79	162±3.84	161±3.33	0.772
Ig-G (DU)	17.22±6.43	8.06±0.97	15.79±4.31	7.87±1.07	0.001

Table 2. Associations between Reproductive Hormones and MFG-E8 Levels in infertile patients with Chlamydial Infection (IgG) status

Reproductive Hormones and MFG-E8 Levels	Infertile group (n=60)		Fertile group (n=30)		p-value
	Chlamydial Positive (n=33)	Chlamydial Negative (n=27)	Chlamydial Positive (n=15)	Chlamydial Negative (n=15)	
MFG (ng/ml)	192.79±52.16	90.24±44.22	215.65±26.07	107.83±39.17	0.0001
AMH (ng/ml)	3.39±2.58	3.86±3.76	4.75±10	4.90±0.002	0.194
FSH (µg/ml)	10.16±6.68	9.67±6.83	6.87±1.71	6.10±1.56	0.0533
LH (µg/ml)	6.45±3.89	8.18±9.33	6.78±1.83	5.44±2.02	0.4770
Testosterone (ng/ml)	0.29±0.18	0.31±0.33	0.19±0.16	0.33±0.22	0.431
PRL (µg/ml)	16.50±9.21	16.39±11.22	11.79±4.60	9.46±3.62	0.026

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Moreover, the study found no significant variations in the mean levels of AMH, FSH, LH, and Testosterone hormones when considering the Chlamydia infection status across all study groups. However, there was a notable difference in PRL levels among all study groups, with a p-value of 0.026.

Table 3 revealed no statistically significant differences in MFG-E8 levels (p=0.297) between fertile and infertile patients. However, significant disparities were observed in FSH (p=0.006) and prolactin (p=0.003) levels when comparing fertile individuals with those experiencing infertility. Conversely, no significant variations were detected in serum LH and testosterone levels between the two groups.

Table 3. Associations between Reproductive Hormones and MFG-E8 Levels regarding the Fertility Status of the Study Groups.

Reproductive Hormones and MFG-E8	Infertile group (n=60) Mean±SD	Fertile group (n=30) Mean±SD	p-value
MFG (ng/ml)	147.60±70.57	163.48±63.68	0.297
FSH (µg/ml)	9.94±6.70	6.50±1.66	0.006
LH (µg/ml)	7.21±6.82	6.13±2	0.393
PRL (µg/ml)	16.46±10.05	10.66±4.26	0.003
Testosterone (ng/ml)	0.29±0.26	0.26±0.21	0.540

Discussion

The findings of the present study reveal that the infertile group exhibited higher mean Ig-G (DU) levels, regardless of Chlamydial infection status, in comparison to the fertile group. These findings highlight significant differences in Chlamydial infection status between the infertile and fertile groups, with a statistically significant result (p<0.0001). Moreover, the present finding is consistent with a previous study, demonstrating a heightened incidence of CT infection among women experiencing infertility, suggesting that CT infection could be a potential risk factor for the development of infertility (10). Conversely, divergent findings from alternative studies suggest a lower prevalence of CT infection among women with infertility (4, 11, 12). These inconsistencies could be attributed to the variations in study settings, including study duration, socioeconomic status, sample size, and diagnostic methods employed. Furthermore, our study findings reveal a noteworthy disparity in MFG-E8 levels between fertile and infertile individuals based on their Chlamydia infection status. Specifically, within the Chlamydia-positive group, the fertility group displayed higher mean Ig-G levels compared to the Chlamydia-negative group. These findings suggest that MFG-E8 may be involved in the pathogenesis of Chlamydia-related infertility. However, another study revealed that women with CT infection exhibit reduced levels of MFG-E8 compared to those without CT infection. These results suggest that chlamydial conditions might influence both ovarian reserve and the overall health of the reproductive tract (13).

MFG-E8 is involved in various biological processes, including cell signaling and modulation of immune responses, and tissue repair (14, 15). Chlamydial infection is known to induce inflammation in the reproductive tract, leading to cellular damage and tissue remodeling (15-18). This significant difference in MFG-E8 levels suggests that CT infection may influence the regulation of MFG-E8. Limited research has explored the potential link between MFG-E8 and reproductive hormones in infertile women with CT infection.

Furthermore, the study revealed no statistically significant differences in MFG-E8 levels between fertile and infertile patients. However, significant disparities were observed in FSH and prolactin levels regarding their fertility status, suggesting a potential association between these hormones and fertility outcomes. This

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finding aligns with previous research, which suggests that elevated PRL levels in infertile women can contribute to female infertility (19). Another study highlighted the role of sex hormones, FSH, LH, estradiol, progesterone, and PRL not only in the susceptibility of chlamydia infection but also in the progression of reproductive tissue complications (20, 21). Conversely, another study reported no notable discrepancy in prolactin (PRL) levels between females who had infertility with Chlamydia trachomatis (CT) infection and the control subjects (12, 22). These conflicting results may be attributed to various factors, including differences in study populations, methodologies, and confounding variables.

In conclusion, the study revealed a significant link between CT infection and infertility, evident from higher Ig-G levels in the infertile group with positive CT infection. Moreover, the fertile group with Chlamydia-positive cases exhibited elevated levels of MFG-8, suggesting its potential involvement in reproductive health. However, no notable differences in MFG-E8 levels were observed between fertile and infertile patients. These findings contribute to our understanding of the intricate relationship between CT infection, reproductive hormones, and infertility, emphasizing the need for further research to explore underlying mechanisms and implications.

Conflict of interest: The authors stated that they have no conflicts of interest to disclose.

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References

1.Liu L, Li C, Sun X, Liu J, Zheng H, Yang B, et al. Chlamydia infection, PID, and infertility: further evidence from a case-control study in China. BMC Womens Health. 2022;22(1):294.

2.Rashidi BH, Chamani-Tabriz L, Haghollahi F, Jeddi-Tehrani M, Naghizadeh MM, Shariat M, et al. Effects of Chlamydia trachomatis Infection on Fertility; A Case-Control Study. J Reprod Infertil. 2013;14(2):67-72.

3.Koca Ö. The Laboratory Diagnosis of Chlamydia Infections. In: Sarier M, editor. Chlamydia - Secret Enemy From Past to Present [Internet]. Infectious Diseases. IntechOpen; 2023. p.3. Available from: http://dx.doi.org/10.5772/intechopen.110464

4.Albus E, Sinningen K, Winzer M, Thiele S, Baschant U, Hannemann A, et al. Milk Fat Globule-Epidermal Growth Factor 8 (MFG-E8) Is a Novel Anti-inflammatory Factor in Rheumatoid Arthritis in Mice and Humans. J Bone Miner Res. 2016;31(3):596-605.

5.Hua X, Li B, Yu F, Zhao W, Tan Y, Li X, et al. Protective Effect of MFG-E8 on Necroptosis-Induced Intestinal Inflammation and Enteroendocrine Cell Function in Diabetes. Nutrients. 2022;14(3):604.

6.Gao YY, Tao T, Wu D, Zhuang Z, Lu Y, Wu LY, et al. MFG-E8 attenuates inflammation in subarachnoid hemorrhage by driving microglial M2 polarization. Exp Neurol. 2021;336:113532.

7.Yang WL, Sharma A, Zhang F, Matsuo S, Wang Z, Wang H, et al. Milk fat globule epidermal growth factor-factor 8-derived peptide attenuates organ injury and improves survival in sepsis. Crit Care. 2015;19:375.

8.Schmitz CR, Oehninger S, Genro VK, Chandra N, Lattanzio F, Yu L, et al. Alterations in expression of endometrial milk fat globule-EGF factor 8 (MFG-E8) and leukemia inhibitory factor (LIF) in patients with infertility and endometriosis. JBRA Assist Reprod. 2017;21(4):313-20.

9.Xu J, Bian G, Zheng M, Lu G, Chan WY, Li W, et al. Fertility factors affect the vaginal microbiome in women of reproductive age. Am J Reprod Immunol. 2020;83(4):e13220.

10.Kaushic C, Zhou F, Murdin AD, Wira CR. Effects of estradiol and progesterone on susceptibility and early immune responses to Chlamydia trachomatis infection in the female reproductive tract. Infect Immun. 2000;68(7):4207-16.

11.Fanchin R, Schonäuer LM, Righini C, Guibourdenche J, Frydman R, Taieb J. Serum anti-Müllerian hormone is more strongly related to ovarian follicular status than serum inhibin B, estradiol, FSH and LH on day 3. Hum Reprod. 2003;18(2):323-7.

12.Cortes E, Montero B, Yuguero O. Chlamydia trachomatis Infection Could Be a Risk Factor for Infertility in Women: A Prospective Descriptive Study. Cureus. 2022;14(10):e30697.

13.Agrawal T, Vats V, Wallace PK, Salhan S, Mittal A. Role of cervical dendritic cell subsets, co-stimulatory molecules, cytokine secretion profile and beta-estradiol in development of sequalae to Chlamydia trachomatis infection. Reprod Biol Endocrinol. 2008;6:46.

14.Salman B, Abdul Kareem W, Khadhim SR. Study the correlation between chlamydial infection and some novel biomarkers in fertile women. J Glob Pharma Technol. 2017;12(9):322-7.

15.Liu Y, Yang X, Guo C, Nie P, Liu Y, Ma J. Essential role of MFG-E8 for phagocytic properties of microglial cells. PLoS One. 2013;8(2):e55754.

16.Yi YS. Functional Role of Milk Fat Globule-Epidermal Growth Factor VIII in Macrophage-Mediated Inflammatory Responses and Inflammatory/Autoimmune Diseases. Mediators Inflamm. 2016;2016:5628486.

17.Darville T, Hiltke TJ. Pathogenesis of genital tract disease due to Chlamydia trachomatis. J Infect Dis. 2010;201(Suppl 2):S114-25.

18.Bryan ER, Kim J, Beagley KW, Carey AJ. Testicular inflammation and infertility: Could chlamydial infections be contributing?. Am J Reprod Immunol. 2020;84(3):e13286.

[DOI: 10.22088/jbums.26.1.49]

19.Özlem Özdemir-Karabağ A. Chlamydia: The Female Reproductive System and Infertility. In: Sarier M, editor. Chlamydia - Secret Enemy From Past to Present [Internet]. Infectious Diseases. IntechOpen; 2023. p.3-4. Available from: http://dx.doi.org/10.5772/intechopen.111756

20.Al-Hilali BMI, Al-Samarrai MKO, Al-Bdri AHA. Measuring some hormonal Levels of infertile women in Samarra city - Iraq. IOP Conf Ser Mater Sci Eng. 2018;454(1):012119.

21.Luján A, Fili S, Damiani MT. Female Infertility Associated to Chlamydia trachomatis Infection. In: Darwish AM, editor. Genital Infections and Infertility [Internet]. IntechOpen; 2016. p.142. Available from: http://dx.doi.org/10.5772/62462

22.Ositadinma IM, Ejenawome DOP, Ezekiel E, Chukwudi IL, Martina NA. Follicle Stimulating Hormone, Luteinizing Hormone, Prolactin, Progesterone and Testosterone Level in Chlamydia Trachomatis Infected Subject. IOSR J Pharm Biol Sci. 2016;11(2):71-4.

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