ACTA FACULTATIS
MEDICAE NAISSENSIS

UDC: 618.177:616.993:612.018(55)
DOI: 10.5937/afmnai39-31982

Original article

Running Title: Level of testosterone, DHEA and prolactin among *Toxoplasma gondii* infected and uninfected infertile couples

An Evaluation of the Level of Testosterone, DHEA and Prolactin among *Toxoplasma gondii* Infected and Uninfected Infertile Couples Attending to Fatima Al-Zahra Infertility Treatment Center, Babol, Northern Iran

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SUMMARY

Introduction/Aims: Toxoplasmosis modifies various hormones and cytokines in the infected hosts which may result in several disorders. This study was conducted to assess testosterone, DHEA, and prolactin concentration levels among *Toxoplasma gondii* infected and uninfected infertile couples.

Methods: Blood samples were collected and sera were separated. The sera were analyzed for the detection of anti-Toxoplasma (IgG& IgM) antibodies using commercial ELISA kits. The level of DHEA was measured by ELISA and the levels of testosterone and prolactin were evaluated by enzyme-linked fluorescent assay (ELFA, VIDAS).

Results: The overall seroprevalence of toxoplasmosis was 58.0% (218/376). Among women, 56.9% (107/188) and 6.5% (7/107) were positive for anti-T. gondii antibodies IgG and IgM, respectively. The IgG and IgM were detected in the sera of 111/188 (59.0%) and 9/111 (8.1%) in male subjects, respectively. A positive association was observed between T. gondii infection and the upper and lower ranges of the normal value of testosterone in males ($x^2 = 6.8$, $p = 0.033$) but not in females ($x^2 = 0.62$, $p = 0.99$). A positive correlation was seen between toxoplasmosis and the upper and lower ranges of the normal value of prolactin in females ($x^2 = 6.5$, $p = 0.039$) but not in male cases ($x^2 = 1.06$, $p = 0.59$).

Conclusion: Our results demonstrated no statistically significant differences between the level of sexual hormones between Toxoplasma-infected individuals and Toxoplasma-free subjects suffering from infertility. These findings suggest that further studies should be performed on infertile cases with large sample sizes in a case-control format.

Keywords: **dehydroepiandrosterone, prolactin, testosterone, toxoplasmosis**

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INTRODUCTION

Toxoplasma gondii, an ubiquitous obligate intracellular parasite, is the most common parasitic infection in humans and all warm-blooded animals (1). In humans, Toxoplasma infection occurs through the ingestion of tissue cysts via the undercooked meat of intermediate hosts and/or by the ingestion of water or food polluted by oocysts from the definitive hosts (2). This infection is usually self-limiting and asymptomatic among immunocompetent persons but it is life-threatening in immunosuppressed patients (3). However, during the acute infection phase, the rapidly multiplying form (tachyzoites) of the parasite invades an extensive variety of nucleated cells and disseminates in the host organs. Under the pressure of sufficient immune control, tachyzoites are transformed to dormant bradyzoites, which is enclosed in tissue cysts and establishes a life-lasting latent chronic infection (4).

Several studies have highlighted the fact that various cells including macrophages, dendritic cells, neutrophils, lymphocytes, and proinflammatory mediators (such as IL-12, IFN-γ and TNF-α) are involved to tightly control the infection course of *T. gondii* (4). On the other hand, the immune system cooperates closely with the nervous and endocrine systems using the same mediators and their receptors, which may be affected by invasion of the parasite (5, 6). There is some evidence that the parasite produces hormones and immunomodulating agents itself and also stimulates the immune cells of a host to produce hormones and cytokines in order to use them as growth factors to establish the infection (6, 7). Furthermore, experimental studies performed on laboratory animals have reported that *T. gondii* infection could be a cause of infertility (8, 9).

In humans, the relationship between sexual hormone level and toxoplasmosis in infertile subjects was observed in some published documents, which raises the concern about the importance of the aforementioned infection among infertile individuals (10 - 14). The aim of this study was to investigate the level of sexual hormones in Toxoplasma-seropositive and seronegative infertile subjects attending to the Fatima Al-Zahra infertility treatment center, Babol, northern Iran.

PATIENTS AND METHODS

Sample collection

In cross-sectional study format, the levels of testosterone, DHEA, prolactin, and *T. gondii* infection were evaluated in 376 infertile subjects who were attending to the Fatima Al-Zahra infertility treatment center, Babol, northern Iran. All participants who were included in our study were under the supervision of an obstetrician-gynecologist. Inclusion criteria for enrolment of cases were: (1) individuals suffering from infertility; (2) subjects without any structural problems that could cause infertility; and (3) those who agreed to contribute to the study.

Ethical aspects

The purpose of the current work was clarified for the participant and they gave their written and informed consent. This study was approved by the ethical committee of Babol University of Medical Sciences, Babol, Iran (MUBABOL.REC. 1395; grant number 9542434).

Socio-demographic and clinical

Socio-demographic data including age, gender, residence and occupation were obtained for all cases using a questionnaire form. Clinical data was obtained for each subject, which included infertility history, abortion history, infertility period, hirsutism, polycystic ovary syndrome and hormone levels. Data was obtained from the subjects, physicians and medical examination records. Dehydroepiandrosterone sulfate (DHEA) (ng/ml), luteinizing hormone (LH) (MIU/ml), follicle-stimulating hormone (FSH) (MIU/ml), thyroid-stimulating hormone (TSH) (MIU /ml), and prolactin (ng/ml) were evaluated in serum samples using chemiluminescence. Testosterone level (nmol/l) was evaluated by ecl using serum samples.

Sample collection

Five milliliters of venous blood was taken from each subject and the serum was separated after

clot retraction by centrifugation at 2500 rpm for 5 min. The sera were collected in Eppendorf tubes and then transported in an icebox to Laboratory Sciences group, Faculty of Paramedical Sciences, Babol University of Medical Sciences, where they were kept at − 20° C until tested.

Detection of *T. gondii*

Firstly, the sera were evaluated for the presence of anti-*T. gondii* antibodies (IgG) using commercially available enzyme-linked immunosorbent assay (ELISA) (EUROIMMUN, Germany) based on the manufacturer's instructions. Secondly, all IgG-positive serum samples were assessed for IgM antibodies against Toxoplasma by an ELISA kit (EUROIMMUN, Germany). The optical density (OD) of IgG and IgM antibodies titers was read at 450 nm by an automatic ELISA reader. The results were interpreted according to the kit's guidelines as follow: negative samples with IgG titers of < 8 IU/ml; positive samples with IgG titer higher than 11 IU/ml; An IgG titer between 8 and 11 IU/ml indicates a borderline result. The sensitivity and specificity of the kits are 99.9% and 100%, respectively.

Evaluation of hormone levels

The ELISA method was performed to determine the level of dehydroepiandrosterone sulfate (DHEA-S) use (EUROIMMUN, Germany) based on the manufacturer's instructions. The normal level in females ranged from 0.03 to 5.88 μg/ml, whereas in males it ranged from 0.06 - 4.58 μg/mL. The level of testosterone, prolactin, luteinizing hormone (LH), follicle-stimulating hormone (FSH), thyroid-stimulating hormone (TSH) were measured by enzyme linked fluorescent assay (ELFA, VIDAS) in a clinical laboratory. The results were interpreted according to the kit's recommendations. The normal ranges were as follow: for testosterone, F: 0.2 - 0.95 ng/dl, M: 2.5 - 10 ng/dL; prolactin, 5- 35 ng/mL; LH, F: 18.4-61.1 mIU/mL, M: 0.7 - 7.4 mIU/mL; FSH, F: 8 - 22 mIU/mL, M: 1-14 mIU/mL; TSH, 0.5-6 mIU/mL.

Statistical analysis

To analyze the results, SPSS v. 22.0 software (SPSS Inc., Armonk, NY, USA) was used. Age values, hormones level and IgG titer were reported as median values and interquartile ranges (IQRs), which were compared by Mann–Whitney U test and $p < 0.05$ was considered statistically significant. The association between the features of the studied population and *T. gondii* infection was performed by χ^2 test and likelihood ratio. Odd ratios (OR) and 95% confidence intervals (CIs) were intended by logistic regression and $p < 0.05$ was considered statistically significant.

RESULTS

A total of 376 individuals (188 couples) participated in the present study. The overall seroprevalence of toxoplasmosis was 58.0% (218/376). Table 1 shows socio-demographic and clinical characteristics, and *Toxoplasma gondii* exposure among women and men attending to the Fatima Al-Zahra infertility treatment center. The demographical and clinical information for 10 couples were obtained as missing data. The mean age was 31.2 ± 6.1 years, with a range of 17 to 48 among females. Among female cases, 70.6% (125/177) and 29.4% (52/177) had primary and secondary infertility, respectively. Hirsutism was observed in 37.9% (67/177). Among the female group, 56.9% (107/188) and 6.5% (7/107) were positive for IgG and IgM anti-*T. gondii* antibodies, respectively. There was no statistically significant correlation between *T. gondii* and demographic and clinical data among the aforementioned women.

The mean age was 34.5 ± 6.0, ranging from 21 to 61 years in male participants. Anti-*T. gondii* antibodies (IgG) were detected in the sera of 111 (59.0%) male participants. Nine out of 111 (8.1%) IgG positive male subjects were positive for anti-*T. gondii* antibodies (IgM). Among male subjects, the highest prevalence rate of toxoplasmosis was observed in farmers (68.4%). An insignificant association was observed between *T. gondii* and demographic and clinical data among the abovementioned men (Table 1).

Table 2 shows the mean value, median values and interquartile ranges (IQRs) of the selected hormones, age and IgG titer among seropositive and seronegative female cases attending to the Fatima Al-Zahra infertility treatment center. The mean age among seropositive and seronegative female cases 32.1 ± 6.1 and 29.9 ± 5.9, respectively, indicated a statistically significant difference between the two groups ($p = 0.01$). The mean level of testosterone was 0.6 ± 0.5 in both seropositive and seronegative females. The mean amount of DHEA was lower in seropositive cases than non-infected subjects (2.3 ± 7.2 vs. 2.5 ± 16.3).

Table 1. *Socio-demographic data, clinical characteristics and Toxoplasma gondii exposure among women and men attending to the Fatima Al-Zahra infertility treatment center*

Variable		Total	Seropositive N (%)	Seronegative N (%)	Borderline N (%)	P-value
Female						
Age	< 27	53	26 (49.1)	27 (51.9)	0 (0.0)	0.08
	28 - 33	61	32 (52.5)	26 (42.6)	3 (4.9)	
	> 34	63	41 (65.1)	18 (28.6)	4 (6.3)	
Residency	Urban	87	48 (55.2)	35 (40.2)	4 (4.6)	0.91
	Rural	90	51 (56.7)	36 (40.3)	3 (3.4)	
Occupation	Work	25	10 (40.0)	13 (52.0)	2 (8.0)	0.17
	Homemaker	152	89 (58.6)	58 (38.2)	5 (3.3)	
PCOS*	No	127	75 (59.1)	47 (37.0)	5 (3.9)	0.35
Yes	50	24 (48.0)	24 (48.0)	2 (4.0)		
Abortion	Yes	34	19 (55.9)	14 (41.2)	1(2.9)	0.94
	No	143	80 (55.9)	57 (39.9)	6 (4.2)	
Type of Infertility	Primary	125	67 (53.6)	53 (42.4)	5 (4.0)	0.62
	Secondary	52	32 (61.5)	18 (56.3)	2 (3.8)	
Period	Irrigular	50	25 (50.0)	23 (46.0)	2 (4.0)	0.59
	Regular	127	74 (58.3)	48 (37.8)	5 (3.9)	
Male						
Age	< 31	60	38 (63.3)	19 (31.7)	3 (5.0)	0.65
	32 - 36	56	33 (58.9)	18 (32.1)	5 (8.9)	
	> 34	61	31 (50.8)	24 (39.3)	6 (9.8)	
Residency	Urban	91	44 (48.4)	35 (38.5)	6 (6.6)	0.12
	Rural	85	57 (67.6)	26 (30.6)	8 (9.4)	
Occupation	Selfemployee	80	48 (60)	26 (32.5)	6 (7.5)	0.06
	Administrative work	20	10 (50)	5 (25.0)	5 (25.0)	
	Farmer	19	13 (68.4)	6 (31.6)	0 (0.0)	
	Labourer	58	31 (53.4)	24 (41.4)	3 (5.2)	

Table 2. *The mean value, median values and interquartile ranges (IQRs) of some hormones, age and IgG titer among seropositive and seronegative female cases attending to the Fatima Al-Zahra infertility treatment center*

	Seropositive			Seronegative			
	Mean ± SD	95% CI*	Median& IQRs	Mean ± SD	95% CI	Median& IQRs	p-value
Testestrone	0.6 ± 0.5	0.51 - 0.71	0.46 (0.28-0.72)	0.6 ± 0.5	0.48 - 0.75	0.48 (0.30-0.75)	0.90
DHEA	2.3 ± 7.2	2.07 - 2.62	1.9 (1.3-3.1)	2.5 ± 16.3	2.2 - 2.75	1.6 (1.6-3.2)	0.19
Prolactin	25.1 ± 21.8	18.5 - 52.9	18 (12.1-29.5)	21.5 ± 15.8	17.61 - 25.46	11.1 (13.0-23.7)	0.55
LH	7.1 ± 8.9	5.2 - 8.9	3.8 (3.5-7.2)	6.6 ± 4.6	5.4 - 7.8	6.0.(3.4-9.2)	0.78
FSH	6.5 ± 2.89	5.83 - 7.06	4.0 (4.5-8.0)	6.1 ± 2.6	5.38 - 6.71	4.0 (3.8-8.2)	0.43
TSH	2.8 ± 2.2	2.34 - 3.28	2 (1.6-3.4)	2.5 ± 1.3	2.11 - 2.80	2.0 (1.5-3.1)	0.47
IgG titer	19.3 ± 6.76	18.0 - 20.5	10 (14.0-24.1)	3.3 ± 3.0	2.6 - 4.0	1.6 (1.9-3.4)	0.00
Age	32.1 ± 6.1	30.9 - 33.2	8.3 (28.0-36.0)	29.9 ± 5.9	28.5 - 31.3	8 (26.5-33.5)	0.01

Table 3. *The mean value, median values and interquartile ranges (IQRs) of some hormones, age and IgG titer among seropositive and seronegative male cases attending to the Fatima Al-Zahra infertility treatment center*

	Seropositive			Seronegative			
	Mean ± SD	95% CI*	Median & IQRs	Mean ± SD	95% CI	Median& IQRs	**p-value**
Testestrone	0.6 ± 0.5	0.51 - 0.71	0.46 (0.28- 0.72)	0.6 ± 0.5	0.48 - 0.75	0.48 (0.30-0.75)	0.90
DHEA	2.3 ± 7.2	2.07 - 2.62	1.9 (1.3-3.1)	2.5 ± 16.3	2.2 - 2.75	1.6 (1.6-3.2)	0.19
Prolactin	25.1 ± 21.8	18.5 - 52.9	18 (12.1-29.5)	21.5 ± 15.8	17.61 - 25.46	11.1 (13.0- 23.7)	0.55
LH	7.1 ± 8.9	5.2 - 8.9	3.8 (3.5-7.2)	6.6 ± 4.6	5.4 - 7.8	6.0.(3.4-9.2)	0.78
FSH	6.5 ± 2.89	5.83 - 7.06	4.0 (4.5-8.0)	6.1 ± 2.6	5.38 - 6.71	4.0 (3.8-8.2)	0.43
TSH	2.8 ± 2.2	2.34 - 3.28	2 (1.6-3.4)	2.5 ± 1.3	2.11 - 2.80	2.0 (1.5-3.1)	0.47
IgG titer	19.3 ± 6.76	18.0 - 20.5	10 (14.0-24.1)	3.3 ± 3.0	2.6 - 4.0	1.6 (1.9-3.4)	0.00
Age	32.1 ± 6.1	30.9 - 33.2	8.3 (28.0-36.0)	29.9 ± 5.9	28.5 - 31.3	8 (26.5-33.5)	0.01

The mean level of prolactin, LH, FSH and TSH were higher among seropositive females compared with seronegative cases. There were no statistically significant differences of the mean of these hormones in *T. gondii* infected and non-infected infertile women.

The mean value, medianed values and interquartile ranges (IQRs) of the selected hormones, age and IgG titer among seropositive and seronegative male cases are shown in Table 3. The mean level of testosterone (5.7 ± 4.8 vs. 5.3 ± 5.2) and TSH (2.9 ± 4.8 vs. 2.1 ± 2.3) were higher among seropositive subjects compared with seronegative cases. The mean level of prolactin (15.7 ± 78.4 vs. 40.6 ± 106.9) and DHEA (2.7 ± 2.2 vs. 5.3 ± 17.6) were lower among seropositive males compared with seronegative cases. *T. gondii* infection had no statistically significant effect on the concentration of the studied sexual hormones.

A positive association was observed between *T. gondii* infection and the upper and lower limits of the normal value of testosterone in males ($p < 0.035$) but not in female cases ($p < 0.10$). Also, a positive correlation was seen between toxoplasmosis and the upper and lower levels of the normal range of prolactin in females ($p < 0.04$) but not in male cases ($p < 0.60$) (Figure 1). In women, the number of cases with the higher than normal value levels was equal in seropositive and seronegative cases (No. 1). Among men, 5 seronegative and 8 seropositive cases had DHEA level higher than normal values. The number of cases with the lower limit levels of the normal value of DHEA was zero in both male and females in seronegative and in seropositive cases.

Male factor infertility was diagnosed in 114 couples and female factor infertility was observed in 37 couples. Both female and male factors were seen in 15 couples, and the etiology was unknown for the others. The seroprevalence of toxoplasmosis among subjects with male factor infertility was 138/376 (36.7%) and 51/376 (13.6%) in cases with female factor infertility. The logistic regression analysis for the factors that influence Toxoplasma infection on infertility is shown in Table 4.

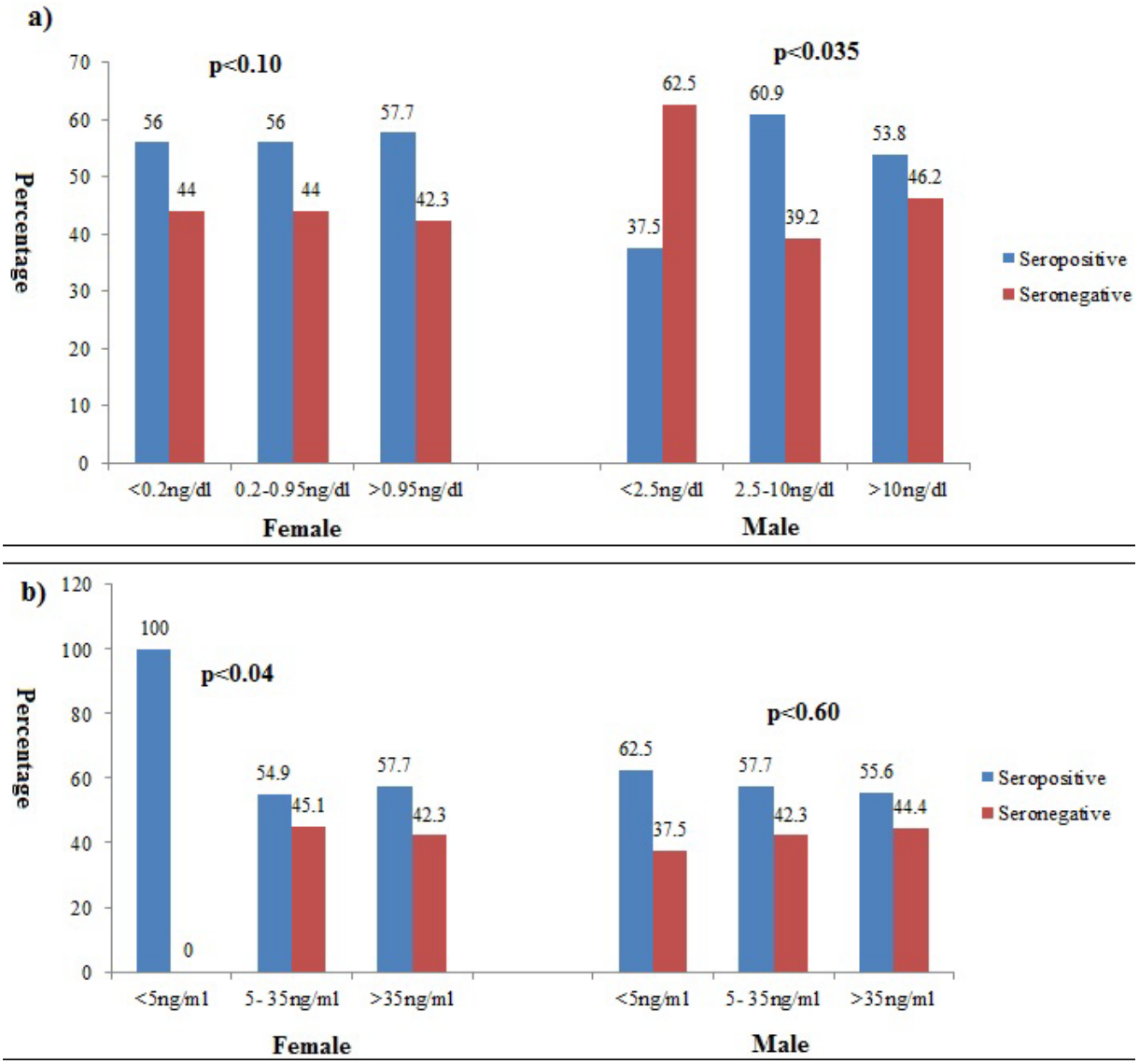

Figure 1. *The levels of a) testosterone and b) prolactin in seropositive and seronegative female and male subjects attending to the Fatima Al-Zahra infertility treatment center*

Table 4. *The logistic regression analysis for the factors that influence Toxoplasma infection in infertility*

Female					Male				
Single variable analysis	Multivariable analysis				Single variable analysis			Multivariable analysis	
Variables	OR (95% CI)	P-value	OR (95% CI)	P-value	**Variables**	OR (95% CI)	P-value	OR (95% CI)	P-value
Infertility factor					**Factor**				
Female	3.0 (0.90-	0.07	4.2 (0.90-19.28)	0.07	**Female**	1.3 (0.58-2.77)	0.56	0.93 (0.39-2.19)	0.86
Male	10.22)				**Male**				
Testestron					**Testestron**				
< 0.2	1.1 (0.36-3.36)	0.87	1.0 (0.29-3.40)	0.99	**< 2.5**	0.8 (0.14-4.53)	0.80	0.8 (0.13-5.04)	0.52
0.2 - .95	1.1 (0.47-2.64)	0.80	1.0 (0.38-2.68)	0.99	**2.5 - 10**	1.9 (0.56-6.12)	0.31	1.9(0.50-7.19)	0.51
> 0.95					**> 10**				
Prolactin					**Prolactin**				
< 5					**< 5**	2.8 (0.46-16.59)	0.27	3.6 (0.43-30.92)	0.23
5 – 35	1.1 (0.49-2.23)	0.90	0.59 (0.23-1.52)	0.27	**5 – 35**	1.7 (0.41-7.16)	0.47	2.1 (0.34-13.75)	0.42
> 35					**> 35**				
DHEA					**DHEA**				
< 1.19	1.2 (0.57-2.57)	0.63	1.76 (0.71-4.37)	0.22	**< 2.40**	2.0 (0.42-9.77)	0.38	1.8 (0.32-9.57)	0.52
1.19 - 8.97					**2.40 - 12.4**	2.4 (0.48-11.80)	0.29	1.8 (0.32-9.87)	0.51
> 8.97					**> 12.4**				
Residency					**Residency**			-	-
Urban	1.0 (0.54-1.81)	0.10	-	-	**Urban**	0.6 (0.29-1.16)	0.13		
Rural					**Rural**				
Age					**Age**			-	-
< 27	0.4 (0.18-0.83)	0.02	-	-	**< 31**	1.7 (0.74-3.95)	0.21		
27 – 33	0.5 (0.26-1.34)	0.10			**31 – 36**	1.4 (0.60-3.13)	0.45		
> 33					**> 36**				

DISCUSSION

A positive correlation between latent toxoplasmosis and infertility has been reported in several studies (11, 13, 14). In the current study, the levels of various hormones were measured among *T. gondii* seropositive and seronegative infertile cases. The present results showed that the level of testosterone in infected males was higher than in non-infected cases but it was equal among infected and Toxoplasma-free females. In regard to testosterone levels, our results were not similar to Zouei et al. (14) which revealed that the mean concentration of serum testosterone was significantly greater in men and women infected by *T. gondii* (12). It is also in disagreement with the results obtained by Mahbodfar et al. (15) who observed a significant difference in testosterone levels among infected and non-infected individuals. However, in regards to DHEA and prolactin levels, the current results are in line with the aforementioned study (15). However, that study compared the mean levels of the abovementioned hormones among infected and non-infected subjects without considering gender. In addition to increased testosterone levels in infected males, the current work found that the frequency of cases with higher than normal values of testosterone in Toxoplasma-infected men and women was higher than in Toxoplasma-free subjects (Figure 1a). High concentrations of testosterone are known to have immunosuppressive (16) or immunomodulatory effects (17), and therefore the increased level of this hormone in humans could be explained by a positive link between testosterone and dopamine which could increase in response to local inflammatory processes in the infected brain (18, 19). Our results demonstrated that the frequency of cases with lower than normal values of testosterone in Toxoplasma-infected men was lower than in Toxoplasma-free subjects (37.5% vs. 62.5%); however, in women, the frequency of cases with lower than normal values of testosterone in infected cases was greater than in non-infected indi-

viduals (60% vs. 40%) (Figure 1a). This finding was in line with a study performed by Flegr et al. who showed that Toxoplasma-infected men had a higher concentration of testosterone, while women had a lower concentration of the hormone compared to the control group (20).

In regard to prolactin levels, the current work showed that infected females tend to have a higher concentration but infected males have lower amounts than non-infected cases. Also, the frequency of cases with upper limits of the normal range of prolactin in Toxoplasma-infected women and men were higher than in Toxoplasma-free subjects (female: 69.2% vs. 30.8%; male: 53.8% vs. 46.2%). In other words, hyperprolactinaemia was observed in infected individuals. These findings were in disagreement with other studies indicating that lower prolactin levels were observed in such women (21). We have no explanation for the increased concentration of this hormone in infected subjects. However, several researchers have reported that prolactin has a positive effect on anti-microbial immunity, which is mainly related to its ability to enhance the response mediated by the Th1-profiled lymphocytes (6, 21, 22). On the other hand, a recent study reported that prolactin could have an inhibitory effect on exacerbated immune responses to infections (23).

Furthermore, the findings obtained here showed that the amounts of the other studied hormones in seropositive and seronegative infertile males and females were different. For example, Toxoplasma-infected males had a higher amount of DHEA levels compared with non-infected individuals, which was reversed in females. Also, seropositive males had lower levels of LH and FSH, which is supported by a study that demonstrates that the concentration of these hormones were significantly lower in the serum of male patients during acute toxoplasmosis than in controls (24). The present results showed that both Toxoplasma-infected men and women tend to have a higher concentration of TSH. Nevertheless, statistically significant differences were not observed for the mean levels of serum testosterone, prolactin, DHEA, LH, FSH and TSH hormone concentration in *T. gondii* seropositive and seronegative male and female cases (Tables 2 and 3). These results are supported by other published documents studies on humans (11).

In respect to the infertility factor, an elevated risk of Toxoplasma infection was observed in women with female factor infertility compared to men with male factor infertility (OR = 3.0; 95% CI: 0.90 - 10.22, p = 0.07) (Table 4).

An important limitation of the current study was a small sample size for the detection of a significant difference in the level of sexual hormones between the seropositive and seronegative cases.

CONCLUSION

Our results demonstrated no statistically significant differences between the level of sexual hormones between Toxoplasma-infected individuals and Toxoplasma-free subjects suffering from infertility. It also revealed that female factor infertility may be related to latent toxoplasmosis. These findings suggest that further studies should be performed on infertile cases with large sample sizes in a case-control format.

Authors' contributions

Narges Kalantari developed the study, performed data analsysis and edited the manuscript. Masomeh Bayani developed the study. Sedigheh Esmailzadeh performed medical examination and all participants were under her supervision. Salman Ghaffari drafted the manuscript. Faeze Ghofrani collected data. Taraneh Ghaffari collected data, performed experiments and edited the manuscript. All authors read and approved the final manuscript.

References

1. Liassides M, Christodoulou V, Moschandreas J, et al.Toxoplasmosis in female high school students, pregnant women and ruminants in Cyprus. Trans R Soc Trop Med Hyg 2016; 110(6): 359-66. https://doi.org/10.1093/trstmh/trw038

2. Mirza Alizadeh A, Jazaeri S, Shemshadi B, et al. A review on inactivation methods of *Toxoplasma gondii* in foods. Pathog Glob Health 2018; 112(6): 306-19. https://doi.org/10.1080/20477724.2018.1514137

3. Wang ZD, Liu HH, Ma ZX, et al. *Toxoplasma gondii* infection in immunocompromised patients: a systematic review and meta-analysis. Front Microbiol. 2017; 8: 389. https://doi.org/10.3389/fmicb.2017.00389

4. Miller CM, Boulter NR, Ikin RJ, et al. The immunobiology of the innate response to *Toxoplasma gondii*. Int J Parasitol 2009; 39(1): 23-39. https://doi.org/10.1016/j.ijpara.2008.08.002

5. de la Luz Galván-Ramírez M, De Arellano AR, Rodríguez-Pérez LR, et al. Hormonal modulation of *Toxoplasma gondii* infection: Regulation of hormonal receptors and cytokine production in THP-1 cells. Exp Parasitol. 2019; 204. https://doi.org/10.1016/j.exppara.2019.107721

6. Dzitko K, Dziadek B, Gatkowska J, et al. *Toxoplasma gondii* binds sheep prolactin. Exp Parasitol 2013; 134(2): 216-9. https://doi.org/10.1016/j.exppara.2013.02.010

7. Ouaissi A. Regulatory cells and immunosuppressive cytokines: parasite-derived factors induce immune polarization. BioMed Res Int 2007; 4:94971. https://doi.org/10.1155/2007/94971

8. Terpsidis KI, Papazahariadou MG, et al. *Toxoplasma gondii*: reproductive parameters in experimentally infected male rats. Exp Parasitol 2009; 121(3): 238-41. https://doi.org/10.1016/j.exppara.2008.11.006

9. Dvorakova-Hortova K, Sidlova A, Ded L, et al. *Toxoplasma gondii* decreases the reproductive fitness in mice. PloS one 2014;9(6):e96770. https://doi.org/10.1371/journal.pone.0096770

10. Shiadeh MN, Niyyati M, Fallahi S, et al. Human parasitic protozoan infection to infertility: a systematic review. Parasitol Res 2016;115(2):469-77. https://doi.org/10.1007/s00436-015-4827-y

11. Fezaa JH, Gati MA-AR, Al-Aboody BA. Study the relationship between some sex hormones and toxoplasmosis among infertile men patients in Thi-qar province. Journal of Education for Pure Science 2019; 9(2): 151-7. https://doi.org/10.32792/utq.jceps.09.02.16

12. Zakai HA, Bisharah MA. Is *Toxoplasma gondii* IgG seropositivity a predisposing factor for infertility? J Adv Lab Res Biol 2014; 6(2): 43-5.

13. Li S, Cui L, Zhao J, et al. Seroprevalence of *Toxoplasma gondii* infection in female sterility patients in China. J Parasitol 2011;97(3):529-30. https://doi.org/10.1645/GE-2680.1

14. Zouei N, Shojaee S, Mohebali M, Keshavarz H. The association of latent toxoplasmosis and level of serum testosterone in humans. BMC Res Notes 2018; 11(1): 365. https://doi.org/10.1186/s13104-018-3468-5

15. Mahbodfar HR, Yousefi Razin E, Saki J, et al. Study of latent *Toxoplasma gondii* role in level of testosterone, DHEA, cortisol and prolactin hormones of young persons. Asian J Epidemiol 2015; 8(3): 64-71. https://doi.org/10.3923/aje.2015.64.71

16. Yao G, Liang J, Han X, et al. In vivo modulation of the circulating lymphocyte subsets and monocytes by androgen. Int Immunopharmacol 2003; 3(13-14): 1853-60. https://doi.org/10.1016/j.intimp.2003.09.002

17. Trumble BC, Blackwell AD, Stieglitz J, et al. Associations between male testosterone and immune function in a pathogenically stressed forager-horticultural population. Am J Phys Anthropol 2016; 161(3): 494-505. https://doi.org/10.1002/ajpa.23054

18. Dominguez JM, Hull EM. Dopamine, the medial preoptic area, and male sexual behavior. Physiol Behav 2005; 86(3): 356-68. https://doi.org/10.1016/j.physbeh.2005.08.006

19. Flegr J. Effects of Toxoplasma on human behavior. Schizophr Bull. 2007; 33(3): 757-60. https://doi.org/10.1093/schbul/sbl074

20. Flegr J, Lindova J, Kodym P. Sex-dependent toxoplasmosis-associated differences in testosterone concentration in humans. Parasitology 2008;135(4):427-31. https://doi.org/10.1017/S0031182007004064

21. Dzitko K, Malicki S, Komorowski J. Effect of hyperprolactinaemia on *Toxoplasma gondii* prevalence in humans. Parasitol Res2008;102(4):723-9. https://doi.org/10.1007/s00436-007-0824-0

22. Paredes M, Gonzalez K, Figueroa J, et al. Immunomodulatory effect of prolactin on Atlantic salmon (Salmo salar) macrophage function. Fish Physiol Biochem 2013; 39(5): 1215-21. https://doi.org/10.1007/s10695-013-9777-7

23. Olmos-Ortiz A, Déciga-García M, Preciado-Martínez E, et al. Prolactin decreases LPS-induced inflammatory cytokines by inhibiting TLR-4/NFκB signaling in the human placenta. Mol Hum Reprod 2019; 25(10): 660-7. https://doi.org/10.1093/molehr/gaz038

24. Oktenli C, Doganci L, Ozgurtas T, et al. Transient hypogonadotrophic hypogonadism in males with acute toxoplasmosis: suppressive effect of interleukin-1β on the secretion of GnRH. Human Reprod. 2004; 19(4): 859-66. https://doi.org/10.1093/humrep/deh161

Article info

Received: Aril 24,, 2021
Revised: September 6, 2021
Accepted: January 4, 2022

Procena nivoa testosterona, dehidroepiandrosterona i prolaktina kod parova sa sterilitetom inficiranih i neinficiranih parazitom *Toxoplasma gondii,* koji su posećivali Centar za lečenje steriliteta Fatma Al-Zahra u Babolu, u Iranu

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SAŽETAK

Uvod/Cilj. Toksoplazmoza modifikuje različite hormone i citokine kod inficiranog domaćina, što može da dovede do različitih poremećaja. Cilj ove studije bila je procena nivoa koncentracija testosterona, dehidroepiandrosterona i prolaktina kod parova sa sterilitetom, inficiranih i neinficiranih parazitom *Toxoplasma gondii*.

Metode. Uzeti su uzorci krvi, nakon čega su serumi odvojeni i analizarini na prisustvo anti-Toxoplasma (IgG i IgM) antitela primenom ELISA testa. Nivo dehidroepiandrosterona izmeren je primenom ELISA testa, dok su nivoi testosterona i prolaktina određeni enzimskim fluorescentnim testom (ELFA, VIDAS).

Rezultati. Ukupna seroprevalencija toksoplazmoze bila je 58,0% (218/376). Kod žena je na prisustvo IgG *Toxoplasma gondii* antiteta bilo pozitivno 56,9% (107/188), a na IgM 6,5% (7/107). IgG antitela detektovana su kod 111/188 (59,0%) muškaraca, a IgM antitela kod 9/111 (8,1%) muškaraca. Uočena je pozitivna veza između infekcije izazvane parazitom *Toxoplasma gondii* i gornjih i donjih opsega normalnih vrednosti testosterona kod muškaraca (x^2 = 6,8, p = 0.033), ali ne i kod žena (x^2 = 0,62, p = 0,99). Utvrđena je pozitivna korelacija između toksoplazmoze i gornjih i donjih opsega normalne vrednosti prolaktina kod žena (x^2 = 6,5, p = 0,039) ali ne i kod muškaraca (x^2 = 1,06, p = 0,59).

Zaključak. Naši rezultati nisu ukazali na stistički značajne razlike između nivoa seksualnih hormona između osoba sa sterilitetom inficiranih i neinficiranih parazitom *Toxoplasma gondii*. Ovi nalazi ukazuju na to da je neophodno sprovesti dalja istraživanja sa parovima sa sterilitetom, ali sa većim brojem ispitanika i u formatu studije slučaja/kontrole.

Ključne reči: **dehidroepiandrosteron, prolactin, testosterone, toksoplazmoza**