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Seroprevalence of *Toxoplasma gondii* and testosterone level in Palestinian butchers

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A cross-sectional study was conducted in 2024 to study the seroprevalence of *Toxoplasma gondii* (*T. gondii*) and male testosterone levels in Palestinian butchers in the West Bank. Sera were collected from 156 butchers and tested for anti-*T. gondii*-IgG and IgM using commercial immune assay and testosterone tests. Univariate and bivariate analyses were used to calculate crude and adjusted odds ratios. The seroprevalences were 62.8% for anti-*T-gondii*-IgG, 7.1% for anti-*T-gondii*-IgM, and 7.1% for both IgG and IgM. *T. gondii*-IgG seropositive individuals had a median testosterone level of 351 ng/dL, significantly lower than the 428.5 ng/dL observed in *T. gondii*-IgG seronegative participants ($p = 0.017$). *T. gondii*-IgG seropositivity was associated with raw meat consumption, exposure to soil, age, and years of practice with adjusted odd ratios of 3.85, 3.32, 1.10, and 1.27, respectively. However, low testosterone level was not significantly associated with *T. gondii* seropositivity ($p = 0.07$). This finding suggests that additional research is needed to understand the relationship between Toxoplasma infection and testosterone levels.

Keywords Toxoplasma, Testosterone, Infertility, West Bank-Palestine, Occupational hazard, *T. gondii*

Abbreviations

BMI	Body mass index
CI	Confidence interval
COI	Conflict of interest
IgG	Immunoglobulin G
IgM	Immunoglobulin M
IQR	Interquartile range
ng/dL	Nanograms per deciliter
OR	Odds ratio

Toxoplasmosis is an infectious disease caused by the protozoan *Toxoplasma gondii*. It can have severe consequences, especially for individuals with immunodeficiency[1]. Human infection can occur through exposure to infective stages or the consumption of infected, undercooked meat[2]. Toxoplasmosis poses a significant healthcare threat, impacting approximately one-third of the global population[3].

While infection with Toxoplasma is asymptomatic in healthy individuals, it can pose serious risks to pregnant women and their unborn babies[4]. In women, toxoplasmosis has been linked to an increased risk of infertility and pregnancy complications[5]. The parasite has been found in the reproductive organs and semen of males across various animal species, including humans[6,7]. A higher incidence of toxoplasmosis has also been observed in infertile men[8,9]. The findings of different studies have suggested that *T. gondii* infection can cause temporary impairment in hormones and other reproductive parameters of human and animal males[8–11]. In men, latent toxoplasmosis was found to affect specific semen parameters (sperm count and motility) but not sperm morphology or semen volume[12]. Tobacco smoking was found to exacerbate the negative impact of *T. gondii* infection on semen parameters[12].

Several studies have attempted to elucidate the relationship between *T. gondii* infection in males and testosterone levels. However, the link between infertility and *T. gondii* infection in males remains uncertain[13,14].

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T. gondii infection was found to cause imbalances in testosterone: estradiol ratios in a significant number of infertile men, which underscores the importance of conducting anti-*T. gondii*-IgG testing in individuals with abnormal ratios[15].

Previously, only one *T. gondii* seroprevalence study was carried out in Palestine. This study involved Palestinian women and revealed that 27.9% and 17.6% were positive for *T. gondii* IgG and IgM, respectively[4]. Seroprevalence of *T. gondii* in Palestinian males or butchers has never been studied before. This study examines the seroprevalence of *T. gondii* and testosterone levels in Palestinian men butchers in the West Bank. Also, it investigates the association between anti-*T. gondii* IgG and testosterone levels, eating raw meat, exposure to soil, contact with cats, smoking, infertility diagnosis, and body mass index (BMI) in butchers. In addition to investigating the association between low testosterone levels and BMI, location, smoking, and age.

Methodology

Study design and settings and population

A cross-sectional study was conducted in 2024 in the West Bank of Palestine. Blood samples were collected from male butchers in Nablus, Tubas, and Jenin governorates. Butchers of sheep, goats, and cattle were included in the study if they had been working as butchers for at least two consecutive weeks. They were excluded if they had been exposed to external hormonal supplements, had medical conditions that could affect testosterone level, have immunity-related issues, or had contraindications to blood sampling procedures. As there are no data on the prevalence of *T. gondii* seropositivity among males in Palestine, the prevalence of anti-*T. gondii* IgG seropositivity in females in Palestine, estimated at 27.9%, was used instead[16]. The targeted population in West Bank cities and villages was obtained from the municipalities in the studied governorates. There were 110 official butchers in Nablus city and camps, 100 in Jenin, and 50 in Tubas. The estimated sample size was 141 based on a probability of 0.279 and a precision of 0.05. A clustered sampling technique was used. Jenin city was inaccessible due to political conflict in Jenin governorates, and Jenin villages were visited instead.

Tools and operational definitions

This study utilized a face-to-face questionnaire to gather personal information, evaluate fertility status, and assess risk factors for *T. gondii* infection. The questionnaire was developed after an extensive literature review. A pilot study was then conducted to evaluate the questionnaire's validity, and subsequent revisions were made based on the results. The personal information collected included age, marital status, residency (urban and rural), educational level (up to high school, undergraduate or higher), years of working experience as a butcher, and smoking status, including cigarettes, waterpipe, and electrical cigarettes. Three smoking levels were studied: mild (1-10), moderate (11-20), and heavy smoking (more than 20), whereas waterpipe smoking was considered mild if less than once a week, moderate if one to six heads a week, and heavy if daily.

The World Health Organization (WHO) defines infertility as the inability to conceive after at least 12 months of regular, unprotected sexual intercourse[17]. Fertility status included infertility (yes, no), number of children, previous history of treatment for infertility, and comorbidities. Risk factors for *T. gondii* infection included eating uncooked meat or seafood (always, sometimes, never), having cats as pets (yes, no), and exposure to soil while working in gardens (always, sometimes, never).

Serological tests: Blood samples were collected from each participant to test for testosterone levels and anti-*T. gondii* IgM and IgG. These samples were analyzed at An-Najah National University Hospital laboratories and stored at 4 °C until analysis. The Elecsys Toxo IgG and IgM assays (Roche Diagnostics) were used to detect Toxoplasma *gondii* antibodies. For Toxo IgG, the results were reported quantitatively as IU/mL, with levels above 1 IU/mL indicating prior exposure or immunity to the parasite. The sensitivity and specificity of the IgG test ranges from 99.45 to 100% and 87.5–99.8%, respectively[18]. The cut-off index (COI) is used for IgM, with COI $\geq$ 1.0 considered reactive. It indicates a significant *Toxoplasma gondii* IgM antibody, suggesting a current or a recent infection, and COI values between 0.8 and $<$ 1.0 are considered equivocal. Repeating the test in 10–14 days may be helpful in the case of equivocal results. The sensitivity for the IgM test ranges from 95.3 to 98.8%, and the specificity ranges from 98.9 to 99.7%. Similarly, for *T. gondii* IgG levels, a result of less than 1 IU/mL is nonreactive, whereas a result of 1 to less than 3 IU/mL is equivocal. A result of 3 IU/mL or higher is considered positive for IgG antibody to Toxoplasma and indicates a current or past Toxoplasma infection[18,19]. Both assays are based on electrochemiluminescence immunoassay (ECLIA) technology and were run on the COBAS analyzer[20]. The laboratory verifies test results when initial findings are inconclusive or if there is a suspicion of an acute infection or positive IgM antibodies. Additionally, alternative tests are performed to assess various testing methods and compare their effectiveness with the Elecsys tests in this population.

Total testosterone levels (numerical continuous). Testosterone levels were measured using the Elecsys Testosterone II assay, with a reference range for males aged 19 to 49 years of 249–836 ng/dL (8.6–29.0 nmol/dL) and age 50 or older: 193–740 nmol/dL[21]. The analytical range of this assay is 0.025–15 ng/mL (0.087–52 nmol/L), with excellent correlation to gold-standard methods such as liquid chromatography-tandem mass spectrometry (LC-MS/MS).

Weight and height were obtained via tools and procedures with demonstrated accuracy and precision, as measured in previous research[22]. BMI was calculated by dividing weight in kilograms over the square of height in meters. The CDC[23] categories were as follows: underweight (less than 18.5), normal weight (18.5–24.9), overweight (25.0–29.9), obese (30.0–34.9), and morbidly obese (35.0 and above).

Statistical analysis

The data were analyzed via IBM's Statistical Package for Social Sciences (SPSS) program for Windows, version 27 (IBM Corp., Armonk, NY, USA). Normality was assessed via the Kolmogorov‒Smirnov test. The descriptive statistics included the means, standard deviations (SDs) or medians, and interquartile ranges (IQRs) for continuous variables, whereas categorical variables are presented as frequencies and percentages. The Mann-Whitney test was used to analyze the variations in testosterone levels between participants who were infected and those who were not infected. Crude odds ratio (cOR) and adjusted OR (aOR) were calculated with a 95% confidence interval, and p-values less than 0.05 were considered statistically significant. In the adjusted binary regression, a p-value of less than 0.25 in the univariate test was considered for inclusion. cOR was adjusted using binary logistic regression. Two models were built. Model 1 examined the associations between anti-*T. gondii* IgG seropositivity (yes, no), contact with cats, raw meat consumption, and soil exposure. This model was adjusted for age and working years, and the interaction effect of age and years of working experience as butchers was also examined. Model 2 examined the associations between testosterone levels (low, normal) and anti-*T. gondii* IgG seropositivity (yes, no), adjusted for cigarette smoking (yes, no), waterpipe smoking (yes, no), years of working experience as butchers, and BMI as continuous variables.

Results

Two hundred and eight butchers were invited to participate in the study. Only 156 agreed to participate, with an estimated response rate of 75.0%. Most of the participants were from Nablus (85.9%) and lived in urban areas (60.9%). The median age of the participants was 32.5 years, with a considerable proportion being 40 to 65 years old (58.9%). Additionally, 73.7% of the participants had up to high school educational level, and 61.5% were married, with 91.7% of the married individuals having children. Furthermore, a significant portion of the participants were obese (68.8%) with a BMI of ≥ 25. The majority (78.2%) of the participants were tobacco smokers; smoking cigarettes (61.6%), waterpipe (24.4%), and e-cigarette (4.4%) (Table 1).

	Sociodemographic data	***n* (%)**
Educational level	Up to high school	115 (73.7)
	Diploma or bachelor's	41 (26.3)
Residency city	Nablus	134 (85.9)
	Tubas	8 (5.1)
	Jenin	14 (9.0)
Place of residency	Urban	95 (60.9)
	Rural	61 (39.1)
Marital status	Married	96 (61.5)
	Single	60 (38.5)
Having children	Yes	88 (56.4)
	No	8 (5.1)
	Not applicable (not married)	60 (38.5)
Age in years	< 40	64 (41.02)
	40–65	92 (58.97)
Body mass index (BMI)	≥ 25	107 (68.6)
	< 25	49 (31.4)
Cigarette smoking	Mild	9 (5.8)
	Moderate	33 (21.2)
	Heavy	54 (34.6)
	Nonsmoker	60 (38.5)
E-cigarette smoking	Mild	3 (1.9)
	Moderate	1 (0.6)
	Heavy	3 (1.9)
	Nonsmoker	149 (95.5)
Waterpipe smoking	Mild	13 (8.3)
	Moderate	4 (2.6)
	Heavy	21 (13.5)
	Nonsmoker	118 (75.6)
Tobacco smoking	Yes	122 (78.2)
	No	34 (21.8)
Variable	Minimum-maximum	Median [IQR]
Age	19–63	32.5 [21]
Body mass index (BMI)	17.28–41.98	27.1 [6.3]
Smoking duration in years	1–51	13 [17]
Number of children for married participants	0–11	3.5 [3]

Table 1. Sociodemographic, obesity, and background information of Palestinian butchers during the year 2024, $n = 156$.

The medical history of the participants showed that 10.9% of butchers had reproductive conditions, 2.5% had endocrinological conditions, and 2.8% had other comorbidities. Additionally, 10.3% had a history of surgical procedures involving the reproductive tract, whereas 11.5% were diagnosed with primary infertility. Moreover, the participants had a testosterone median reading of 385 and IQR of 238. Most of the participants, 85.9% ($n = 134$), had normal total testosterone levels, and 14.1% ($n = 22$) had low total testosterone levels. (Table 2: A). Out of the studied butchers, 96 (61.6%) consumed raw meat, whereas 151 (96.8%) never consumed raw seafood. Additionally, 30 (19.2%) of the butchers had regular contact with cats, and 52 (32.7%) had soil exposure. Years of work experience ranged from 0.5 to 60 years, with a median [IQR] of 12 (19.8) years (Table 2: B). Ninety-eight participants (62.8%) tested positive for *T. gondii* IgG antibodies, with a median reading of 183 and IQR of 433.3, 7.1% tested positive for *T. gondii* IgM antibodies, with a median reading of 0.18 and IQR of 0.17, 3.8% had borderline *T. gondii* IgM levels, and 89.1% tested negative for *T. gondii* IgM antibodies. All participants who were seropositive for *T. gondii*-IgM had concurrent *T. gondii*-IgG seropositivity. The *T. gondii*-IgG seropositive participants ($n = 98$) had a median testosterone level and [IQR] of 351[239] ng/dL, which were significantly lower than those of the seronegative participants (428.5 [248] ng/dL; $p = 0.017$; Z score = −2.387) (Table 2: C).

The results of the univariate analysis for the relationship between anti-*T. gondii*-IgG seropositivity and other factors revealed that butchers exposed to soil (work in gardens) were 2.53 times more susceptible to be *T. gondii* seropositive (cOR = 2.53, $p = 0.021$). However, other tested variables were not statistically significant (p-values ≥ 0.05) (Table 3).

The binary logistic regression results for the risk factors associated with anti-*T. gondii*-IgG seropositivity (Model 1) revealed that participants who consumed raw meat were more likely to have anti-*T. gondii*-IgG seropositivity

		***n* (%)**
(A) Medical history of the participants		
Testicular and reproductive disorder	Undescended testicle	3 (1.9)
	Varicocele	14 (8.97)
Endocrinological disorders	Hypothyroidism	1 (0.6)
	Hyperthyroidism	1 (0.6)
	Unknown	2 (1.3)
Testosterone levels	Low	22 (14.1)
	Normal	134 (85.9)
Other chronic diseases	Diabetes	6 (3.8)
	Hypertension	12 (7.7)
	Asthma	5 (3.2)
	Gout	1 (0.6)
	Fatty liver disease	1 (0.6)
	Eczema	1 (0.6)
	Allergic rhinitis	1 (0.6)
History of reproductive tract surgeries	Varicocele removal	14 (8.97)
	Orchiopexy	2 (1.3)
Infertility diagnosis among the married	Yes	11 (7.1)
	No	145 (92.9)
(B) Factors related to *T. gondii* infection		
Raw meat consumption	Always	55 (35.3)
	Sometimes	41 (26.3)
	Never	60 (38.5)
Contact with cats	Yes	30 (19.2)
	No	126 (80.8)
Soil exposure	Always	8 (5.1)
	Sometimes	43 (27.6)
	Never	105 (67.3)
(C) Anti-*T. gondii*-IgG and IgM seropositivity		
T. gondii IgG	Reactive	98 (62.8)
	None reactive	58 (37.2)
T. gondii IgM	Reactive	11 (7.1)
	Border	6 (3.8)
	None reactive	139 (89.1)

Table 2. Descriptive statistics of (A) medical history of the participants, (B) factors related to Toxoplasma gondii infection, and (C) anti-*T. gondii*-IgG and IgM seropositivity in Palestinian butchers during the year 2024, $n = 156$.

Variable	Category	*T. gondii*-IgG seropositive *n* (%)				
		Yes	No	cOR	95% CI	P -value
Raw meat consumption	Yes	65 (67.7)	31 (32.3)	1.72	0.88–3.34	0.127
	No	33 (55.0)	27 (45)	1		
Soil exposure	Yes	39 (76.5)	12 (23.5)	2.53	0.19–5.38	0.021
	No	59 (56.2)	46 (43.8)	1		
Contact with cats	Yes	18 (60.0)	12 (40.0)	0.86	0.38–1.95	0.834
	No	80 (63.5)	46 (36.5)	1		
Smoking	Yes	76 (62.3)	46 (37.7)	0.90	0.41–1.99	0.844
	No	22 (64.7)	12 (35.3)	1		
Infertility diagnosis	Yes	10 (90.9)	1 (9.1)	3.71	0.45–30.62	0.281
	No	62 (72.9)	23 (27.1)	1		
BMI	Overweight	35 (59.3)	24 (40.7)	0.43	0.19–1.02	0.054
	Obese	37 (77.1)	11 (22.9)	1.29	0.60–2.77	0.514
	Normal weight	26 (53.1)	23 (46.9)	1		

Table 3. Univariate analysis results showing the crude odds ratios (cOR) of the associations between anti-*T. gondii*-IgG seropositivity in Palestinian butchers and related predictors during the year 2024, $n = 156$.

Variable	Category	aOR (95%CI)	*P* Value
Contact with cats	Yes	0.68 (0.24–1.93)	0.469
	No	1	
Raw meat consumption	Yes	3.85 (1.48–10.0)	0.006
	No	1	
Soil exposure	Yes	3.32 (1.272–8.671)	0.014
	No	1	
Age in years		1.1 (1.02–1.18)	0.011
Years of working experience as a butcher		1.27 (1.05–1.5)	0.014

Table 4. Bivariate logistic regression analysis results showing adjusted odds ratios for some risk factors associated with anti- *T. gondii* -IgG seropositivity (model 1), and related predictors, in Palestinian butchers during the year 2024, $n = 156$.

(aOR = 3.85, 95% CI: 1.48–10.0, $p = 0.006$). Moreover, participants with soil exposure were more likely to have anti- *T. gondii*-IgG seropositivity than those without (aOR = 3.32, 95% CI: 1.27–8.67, $p = 0.014$). Additionally, older participants were more likely to have anti-*T. gondii*-IgG seropositivity than younger participants were (aOR = 1.1, 95% CI: 1.02–1.8, $p = 0.011$). Furthermore, the more years of experience the participants had working as butchers, the more likely they were to have anti-*T. gondii*-IgG seropositivity (aOR = 1.272, 95% CI: 1.05–1.5, $p = 0.014$). No interaction between age and other variables, years of experience, raw meat consumption, exposure to soil, and contact with cats, were found ($p \geq 0.05$) (Table 4).

The univariate analysis of the associations between low testosterone level in Palestinian butchers and some related predictors are summarized in Table 5. Participants seropositive to anti-*T. gondii* IgG were associated with increasing odds of lower testosterone levels, though not significant ($p > 0.05$).

The binary logistic regression results (Table 6) demonstrated the associations between low testosterone level and BMI with aOR of (1.17, 95% CI: 1.04–1.23, $p = 0.006$).

Discussion

In this study, anti-*T. gondii* IgG seropositivity was observed in 62.8% of studied butchers, reflecting high exposure to the parasite, whereas 7.1% were seropositive for both IgG and IgM, indicating current infection.

Several studies have tried to assess the seroprevalence of *T. gondii* infection among different populations and occupations, consistently finding that *T. gondii* is most prevalent among butchers. In a study comparing subgroups of participants with different characteristics, butchers had the highest percentage of *T. gondii* infection compared with other subgroups (50%)[20]. However, another study revealed no relationship between *T. gondii* seropositivity and being a butcher[21]. The study results indicated that the seroprevalence of *T. gondii* infection among butchers in Palestine was high (62.8%). The lack of previous local studies has limited our ability to compare our results with other demographic groups. However, in a previous study targeting pregnant women in Palestine, the estimated prevalence of *T. gondii* infection was 27.9%[16]. This higher seroprevalence among Palestinian butchers suggests a possible negative impact of occupational exposure to *T. gondii* infection. The seroprevalence of *T.* gondii

Variable	Category	Testosterone level				
		Low	Normal	cOR	95% CI	P -value
Anti-*T*-IgG positivity	Yes	18 (18.4)	80 (81.6)	3.0	0.97–9.47	0.075
	No	4 (6.9)	54 (93.1)	1		
Anti-*T.*-IgM positivity	Yes	3 (27.3)	8 (72.7)	1.88	0.15–23.4	0.625
	Border	1 (16.7)	5 (83.3)	2.52	0.61–10.39	0.201
	Not	18 (12.9)	121 (87.1)	1		
Educational level	Up to high school	18 (15.7)	97 (84.3)	1.72	0.55–5.41	0.44
	Undergraduate or higher	4 (9.8)	37 (90.2)	1		
Body Mass Index	Overweight	7 (11.9)	52 (88.1)	0.40	0.1–1.13	0.083
	Obese	12 (25.0)	36 (75.0)	2.06	0.50–8.45	0.314
	Normal Weight	3 (6.1)	46 (93.9)	1		
Location	City	10 (10.5)	85 (89.5)	0.43	0.19–1.19	0.156
	Rural	12 (19.7)	49 (80.3)	1		
Cigarette smoking	Mild and moderate	4 (9.5)	38 (90.5)	1.51	0.54–4.22	0.434
	Heavy	7 (13.0)	47 (87.0)	2.13	0.63–7.23	0.224
	No	49 (81.7)	11 (18.3)	1		
Waterpipe smoking	Mild and moderate	4 (23.5)	13 (76.5)	1.14	0.30–4.36	0.843
	Heavy	3 (14.3)	18 (85.7)	054	0.10–2.844	0.469
	No	15 (12.7)	103 (87.3)	1		
Tobacco smoking	Yes	16 (13.1)	106 (86.9)	0.70	0.25- 1.96	0.502
	No	6 (17.6)	28 (82.4)	1		
Age in years	< 40	12 (12.4)	85 (87.6)	0.69	0.28–1.72	0.426
	40–65	10 (16.9)	49 (83.1)	1		

Table 5. Univariate analysis results of the association between low testosterone levels in Palestinian butchers and some related predictors during the year 2024, $n = 156$.

Variable	Category	aOR (95%CI)	*P*-value
Anti-*T. gondii*-IgG seropositivity	Yes	3.18 (0.91–11.1)	0.07
	No	1	
Cigarette smoking	Yes	1.49 (0.54–4.06)	0.442
	No	1	
Waterpipe smoking	Yes	0.6 (0.2–1.80)	0.359
	No	1	
Body mass index		1.17 (1.04–1.23)	0.006
Years of working experience as a butcher		1.02 (0.98–1.06)	0.344

Table 6. Binary logistic regression results for the associations between having low testosterone levels (model 2) and anti-T. gondii -IgG seropositivity, smoking, body mass index (BMI), and working years in Palestinian butchers during the year 2024 and related predictors, $n = 156$.

infection among butchers in the West Bank was greater than those in other Middle Eastern countries, including Egypt (33.3–52.4%)[23,24] and Iran (46.1%), compared with the control group (31.4%)[22] and in Iraq (28%)[25].

Consuming undercooked meat or seafood contaminated with Toxoplasma, handling food, and not washing hands can lead to Toxoplasma infection. Similarly, knives, utensils, cutting boards, and other foods in contact with raw contaminated meat or seafood can lead to Toxoplasma infection[26]. The results of this study agree with previous studies reporting that the consumption of raw meat substantially increased the likelihood of becoming infected regardless of the quantity consumed by different individuals[27]. Moreover, exposure to soil had similar results, with an increased seroprevalence rate in those who dealt more frequently with soil, whether for gardening or other reasons. In contrast to expectations, contact with cats showed no association with anti-*T. gondii* IgG seropositivity. This could reflect the nature of the exposure; for example, cats fed cooked food are less likely to carry *T. gondii* than those fed raw meat, thereby posing a lower transmission risk to humans[23].

The study revealed that the more years spent practicing butchery, the more likely the butcher becomes infected, likely due to prolonged occupational exposure to infected raw meats. To minimize occupational exposure to infected/contaminated raw meats, butchers should consider using gloves and aprons, practicing good hand hygiene, and ensuring proper and regular sanitization of work surfaces and equipment. Moreover, many Palestinian butchers taste raw meat to assess its quality. This practice should be replaced with alternative, safe, standardized procedures. Health authorities should initiate awareness campaigns to educate butchers on the risks associated with improper handling of raw meat.

The relationship between seropositivity to Toxoplasma and testosterone level is controversial and may be influenced by different parasite strains[28,29]. Understanding total testosterone level changes associated with Toxoplasma infection could improve our understanding of acquired infertility, as shown in a previous study[12]. The results of the present study revealed that the mean total testosterone level was significantly lower in individuals infected with *T. gondii* than in noninfected individuals. Nevertheless, both means lie within the normal range of total testosterone levels. Although disparities in total testosterone levels among seropositive and seronegative individuals fall within the normal reference range, subclinical alterations may still occur without overt manifestations; for example, lower testosterone levels could result in lower-than-expected hematocrit levels, even if they do not cause frank anemia[30]. Additionally, decreased testosterone may contribute to dyslipidemia, resulting in below-reference semen parameters and negatively impacting bone mineral density[31–33]. Although the univariate and adjusted analyses revealed no statistically significant associations between reactive IgG and low testosterone levels ($p = 0.055$ and $p = 0.07$, respectively), the results still hold clinical significance. The result may have practical importance or real-world implications despite failing to reach statistical significance at the conventional level of 0.05. Males infected with *T. gondii* were found to have noticeably lower semen concentrations[12]. Moreover, the concentration of progressively and nonprogressively motile sperm is negatively impacted in infected individuals[12]. These results highlight the need to include toxoplasmosis testing in infertility evaluations, especially for those facing unexplained infertility challenges.

To control infections among butchers in the West Bank, regulations in abattoirs and fresh meat shops must be strengthened, emphasizing hygiene and safety. Comprehensive training for workers on proper handling and sanitation is crucial. Monitoring animal health before slaughter and reviewing existing legislation will enhance infection control. Public awareness campaigns in collaboration with health authorities will further ensure a safer meat supply chain for the community. While the study revealed a high prevalence of *T. gondii* infection and its potential association with infertility in this high-risk group, more comprehensive investigations are necessary to establish a conclusive link. We recommend conducting longitudinal studies on the relationship between Toxoplasma infection and testosterone levels across diverse populations.

Limitations and strengths

This study has several limitations. One limitation could be establishing a direct causal relationship between anti-*T. gondii*-IgG seropositivity and testosterone levels among butchers, as various confounding factors, such as lifestyle, diet, and other health conditions, could influence testosterone levels. Additionally, the study's cross-sectional design may limit the ability to draw definitive conclusions about the temporal relationship between anti-*T. gondii*-IgG seropositivity and testosterone levels. The testosterone assay shows minimal interference from substances like bilirubin, hemoglobin, and biotin, ensuring reliable results[34,35]. Despite these limitations, the strength of this study lies in its focus on a specific high-risk group, the butchers, and its investigation of the relationship between anti-*T. gondii*-IgG seropositivity and testosterone levels within this population. The study indicated that the prevalence of *T. gondii* infection in the West Bank is greater than that in other Middle East countries.

Conclusions

This study revealed a high seroprevalence of *T. gondii* infection in individuals occupationally exposed to raw livestock meat and viscera in abattoirs and slaughterhouses, indicating that butchers were at high risk of *T. gondii* infection in Palestine. Additionally, raw meat consumption and soil exposure were significant risk factors for anti-*T. gondii*-IgG seropositivity. Although the current study did not find a statistically significant association between reactive IgG and low testosterone levels, the observed link is worthy of further investigation. Furthermore, there is a need to increase awareness regarding safe handling and the negative health impacts of the consumption of raw meat products to minimize the risk of zoonoses.

Recommendations

Based on these findings, further research on *T. gondii* infection in the general population and other specific demographic groups in Palestine would provide important information about the prevalence and potential risk factors associated with the infection. Further investigations should be conducted to assess the relationship between *T. gondii* infection and infertility.

Data availability

All data is provided within the manuscript file.

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Author contributions

B.D. conceptualized and designed the study, supervised the data collection and analysis, and critically revised the manuscript for important intellectual content. B. E., B. A., A. S., and M. D. contributed to the data collection, analysis, and interpretation, and drafted the manuscript. L. K. and S. B. contributed to the study design, data interpretation, and critical revision of the manuscript. All authors approved the final version of the manuscript and agreed to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

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Declarations

Competing interests
The authors declare no competing interests.

Ethics approval and consent to participate
Ethical approval was obtained from the Institutional Review Board (IRB) at An-Najah National University in Palestine (ref: Med.Dec.2023/31). The study followed the ethical standards of the Declaration of Helsinki. The participants were assured that all the data would be confidential and available only to the researcher.

Informed consent
Informed consent was obtained from each participant prior to participation.

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