

MAY SCHISTOSOMIASIS *HAEMATOBIMUM* AND *MANSONI* LEAD TO INFERTILITY IN THE HOST?

By

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Abstract

Schistosomiasis is worldwide water-borne infectious risky disease. Steroid hormones are synthesized from cholesterol in adrenal glands and gonads. They include corticosteroids, androgens, estrogens and progestogens. The present study evaluated the effect of *Schistosoma mansoni* and *S. haematobium* infections on the hamsters' fertility as indicated by the changes of steroid sex hormones levels. Eighteen *Mesocricetus auratus* clear laboratory bred hamsters (9 males & 9 females) were divided into 3 groups. G1: was infected with *S. mansoni* cercariae, G2: was infected with *S. haematobium* cercariae, and G3: was not infected (negative control). The results showed that testosterone level increased in all infected hamsters but markedly increased in those infected with schistosomiasis *haematobium* (7.52 & 0.78nmol/L) as compared to control (2.41 & 0.341nmol/L). Progesterone level decreased in all infected hamsters especially in *S. mansoni* and *S. haematobium* females (5.732 & 11.13nmol/L) in comparison to control (86.34 & 64.7nmol/L). Estradiol E2 level markedly decreased in female hamsters infected with either *S. mansoni* or *S. haematobium* (247.54 & 463.8pmol/L) as compared to control (1658pmol/L) but not detected in male hamsters.

Keywords: *Schistosoma* species, Hamsters, Testosterone, Estradiol, Progesterone, Infertility.

Introduction

Schistosomiasis is a major neglected tropical disease that affects more than 250 million people worldwide (Val *et al.*, 2017). Schistosomiasis *haematobium* and *mansoni* are serious public health problem in tropical and subtropical regions (Calasans *et al.*, 2018), particularly in rural agricultural areas with limited access to clean water (Oliveira *et al.*, 2019).

Neuroimmunoendocrine host-parasite interactions are very complex and involve hormones, cytokines, chemokines and neuropeptides which play important roles in these interactions. These systems are connected by a complex network in which the immune response of the host to the parasite is modulated by many hormones (Hernández-Bello *et al.*, 2010).

Habib *et al.* (2010) reported a relation between schistosomiasis and hormonal changes. Delmondes *et al.* (2014) found an association between schistosomiasis and infertility, which occurred due to formation of egg granuloma in reproductive organs. Woodall and Kramer (2018) reported that *S. haematobium*

infection in female may lead to infertility. Abdel-Naser *et al.* (2019) suggested that schistosomiasis may lead to male infertility due to hormonal imbalance, damage of testicular tissue or genital ducts obstruction. Ribeiro *et al.* (2019) concluded that basic hormonal mechanisms can lead to the discovery of entirely novel anthelmintic interventions against schistosomiasis.

The present study aimed to evaluate the effect of *Schistosoma mansoni* and *S. haematobium* infection on hamsters' fertility by studying the changes of steroid sex hormones levels.

Materials and Methods

Animals, parasites and infection: *Schistosoma mansoni* and *Schistosoma haematobium* was purchased from the Schistosome Biological Supply Centre (SBSC), Theodor Bilharz Research Institute (TBRI), Giza, Egypt. A total of 18 *Mesocricetus auratus* hamsters (9 males & 9 females) were divided in three groups each of three males and three females. G1: was infected with *S. mansoni* cercariae, G2: was infected with *S. haematobium* cercariae, and G3: was not

infected (negative control). Stool and urine examinations were used to prove the hamsters' infection with *S. haematobium* and *S. mansoni*.

Infection was done percutaneously with 350-400 *Schistosoma* cercariae/hamster. 11-12 weeks later, orbital sinus blood sample was collected under general anesthesia. Hamster was scuffed with thumb and forefinger of the non-dominant hand and skin around the eye was pulled taut. A capillary tube was inserted into eye medial canthus at 30 degree angle to nose. Slight thumb pressure was enough to puncture tissue, enter the plexus/sinus and blood was put into the capillary tube. Once the required blood volume was collected, the capillary tube was gently removed and wiped with sterile cotton. Bleeding if occurred was stopped by gentle finger pressure (Parasuraman *et al*, 2010).

Hormone serum levels: Estradiol, testosterone and progesterone hormones levels in sera were measured using an electrochemiluminescence immunoassay (ECLIA) with Cobas e411 Analyser (Roche Diagnostics, USA), according to manufacturer instructions. ECLIA is a competitive test using two monoclonal antibodies. Endogenous sex hormones released from sample compete by adding hormone derivative labeled with a ruthenium complex for the binding sites on the biotinylated antibody. The reaction mixture was aspirated into the measuring cell

where the microparticles were magnetically captured onto the electrode surface. Unbound substances were removed with ProCell/ProCell M. Application of an electrode voltage induced chemiluminescent emission which was measured by a photomultiplier.

Statistical analysis: Data were coded, tabulated, and statistically analyzed using IBM SPSS statistics (Statistical Package for Social Sciences) software version 18.0, IBM Corp., Chicago, USA, 2009. Descriptive statistics were done for quantitative data as minimum and maximum of the range as well as M±SD (standard deviation). Inferential analyses were done using Shapiro-Wilk test for normality testing and ANOVA test with post hoc Tukey's test. The P value < 0.050 was significant.

Results

Testosterone level increased in infected male and female hamsters, markedly in *S. haematobium* infected males (7.22±0.27 nmol/L). Progesterone level decreased in all infected hamsters, markedly in *S. mansoni* and *S. haematobium* females (5.9±0.5 & 10.9±2.7nmol/L, respectively). Estradiol E2 level markedly decreased in females infected with both *S. mansoni* and *S. haematobium* (252.3±29.3 & 467.9±21.6 pmol/L). while in male hamsters, level was not detectable (18.4 pmol/L). Details were given in table (1) and figures (1, 2, 3, 4 & 5).

Table 1: Serum levels of sex hormones among infected and non-infected hamsters

Hormone	Sex	Group	M±SD	Range	p	HG
Testosterone (nmol/L)	Male	<i>S. mansoni</i>	5.35±0.99	4.23–6.12	* 0.001	a
		<i>S. haematobium</i>	7.22±0.27	6.98–7.52		B
		Non-infected	2.42±0.48	1.95–2.90		c
	Female	<i>S. mansoni</i>	0.50±0.26	0.27–0.79	0.507	A
		<i>S. haematobium</i>	0.69±0.23	0.43–0.87		A
		Non-infected	0.40±0.36	0.08–0.79		A
Progesterone (nmol/L)	Male	<i>S. mansoni</i>	41.8±10.0	31.7–51.6	* 0.002	a
		<i>S. haematobium</i>	66.6±6.3	59.5–71.7		B
		Non-infected	86.8±8.5	78.5–95.5		B
	Female	<i>S. mansoni</i>	5.9±0.5	5.4–6.4	* 0.001	A
		<i>S. haematobium</i>	10.9±2.7	8.1–13.5		A
		Non-infected	64.8±5.3	59.5–70.1		B
Estradiol (pmol/L)	Male	<i>S. mansoni</i>	<18.4		Could not be calculated	
		<i>S. haematobium</i>	<18.4			
		Non-infected	<18.4			
	Female	<i>S. mansoni</i>	252.3±29.3	225.7–283.7	* 0.004	a
		<i>S. haematobium</i>	467.9±21.6	448.6–491.2		a
		Non-infected	1902.3±682.1	1376.0–2673.0		b

ANOVA test with post hoc Tukey's test. *Significant. HG: Homogenous groups with same letter (a,b,c)

Discussion

Schistosomes infect more than 250 million people in 74 countries worldwide in the endemic areas of Africa, Eastern Asia, Central America, the Caribbean, South America, and the Middle East (Gryseels *et al*, 2006). Steroid hormones are synthesized from cholesterol in the adrenal glands and gonads. These include corticosteroids, androgens, estrogens and progestogens (Frye, 2009).

Many studies revealed that parasites can change the hormonal microenvironment to favor their growth and reproduction. Added to their capability to actively evade immune responses, parasites can also evolve structures similar to steroid hormone receptors that can bind to hormone metabolites produced by the host (Escobedo *et al*, 2005).

In this study, we are aiming to investigate the effect of schistosomiasis *mansoni* and *haematobium* on the fertility of the host by studying the changes of the levels of steroid sex hormones in infected male and female hamsters.

Testosterone is the primary male sex hormone that plays a key role in the development of male reproductive tissues such as testes and prostate (Mooradian *et al*, 1987). The results of the present work showed increased testosterone level in all infected hamsters (male and female) in comparison to non-infected ones. This agreed with Abdallah *et al*, (1994) who detected an increase in serum levels of testosterone in both *S. mansoni* infected males and females. Oliveira *et al*. (2019) reported increase of testosterone level in male hamsters infected with *S. mansoni*, while it decreased in females. These results are in contrast with those obtained by He *et al*, (2000) who found that infection with *S. japonicum* led to a decrease in testosterone levels in the serum of male mouse, and Marzouki and Amin, (1997) who found that schistosomiasis *mansoni* causes a decrease in testosterone level, while serum levels of gonadotropins (FSH, LH) showed no significant changes. Besides, Ashby *et al*, (2012) reported that in-

creased testosterone levels could be the cause of infertility associated with *S. mansoni* and *haematobium* infections.

Progesterone is an endogenous steroid and progestogen sex hormone involved in the menstrual cycle, pregnancy, and embryogenesis of humans and other species (Patel *et al*, 2015).

The results of the present work also showed that progesterone level decreased in all infected hamsters but markedly decreased in female hamsters infected with both *Schistosoma mansoni* and *haematobium* (5.732 & 11.13 nmol/L) in comparison to non-infected hamsters (86.34 and 64.7 nmol/L). This agreed with Oliveira *et al*, (2019) who reported that *S. mansoni* infection caused a decrease of progesterone levels in hamsters. But, Abdallah *et al*, (1994) detected an increase in progesterone level in both *S. mansoni* infected males and females.

To diagnose fertility, progesterone level is measured to assess ovulation and luteal phase (Guillaume *et al*, 1987). Progesterone has a role in regulation of sperm functions such as capacitation and motility. If sperms lose their acrosomes before binding to oocyte, they become unable to bind to zona pellucida and thereby, to fertilize (Shah *et al*, 2003). Hence, the reduction of progesterone levels shown by the present study could be a cause of infertility associated *S. mansoni* and *S. haematobium* infection, both in females and males.

Ovary is the organ responsible for the production of estrogens (the most active of which is 17 β -estradiol). Estrogens are responsible for maturation of vagina and uterus and have a role in ovarian function. Therefore, estrogens play an important role in female fertility (Ryan, 1982).

The present results showed that estradiol E2 level markedly decreased in female hamsters infected with *Schistosoma mansoni* and *S. haematobium* (247.54 & 463.8 pmol/L) in comparison to non-infected female hamsters (1658pmol/L). This agreed with Wang *et al*, (2001) who found that schistosome infection

may suppress estradiol and progesterone secretion, decrease the rate of pregnancy and it may also increase the complications and mortality during the gestation periods. Santos *et al.*, (2014) revealed in their study that infection with *S. haematobium* may lead to infertility in females. Oliveira *et al.* (2019) detected a significant decrease in estradiol in hamsters infected with *S. mansoni*.

Conclusion

Schistosomiasis *mansoni* and *haematobium* may lead to infertility in male and female hosts by affecting levels of steroid sex hormones; testosterone, progesterone and estrogen. Studying physiological and molecular mechanisms involved in complex hormonal host-parasite relationship are ongoing and will be published in due time elsewhere

Ethical consideration: The study was approved by the Research Ethics Committee, Faculty of Medicine, Ain Shams University, FWA 00006444. Animals were dealt with according to the national regulations for the Animal Ethics rules, Ain-Shams University.

Declarations: Authors declared that they neither have any interest nor received fund.

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Explanation of figures

Fig. 1: Serum testosterone among male groups

Fig. 2: Serum testosterone among female groups

Fig. 3: Serum progesterone among male groups

Fig. 4: Serum progesterone among female groups

Fig. 5: Serum estradiol among female groups

