Immunohistochemical expression of oestrogen and progesterone receptors during experimental acute and chronic murine Schistosomiasis mansoni

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ABSTRACT

Introduction: The responsibility of Schistosoma mansoni in female infertility is still controversial. This study was conducted to evaluate the effect of acute and chronic schistosomiasis mansoni infection on the endometrium using immunohistochemical analysis of uterine hormone receptor expression. Methods: Twenty-four nonpregnant swiss albino mice were divided into three groups: control, noninfected; acute; and chronic Schistosoma mansoni infection. Histological sections of uterine specimens were examined by light microscope with an image analyzing system to detect structural histological, estrogen receptor (ER) and progesterone receptor (PR) expression in the endometrium. Results: No secretory phase was detected in the endometrium in acute and chronic Schistosoma infection. Hormone receptor expression (ER and PR) showed statistically significant differences among the groups (p<0.05), with significant low ER hormone expression in chronic infection, compared to control proliferative, control secretory and acute infection cases, and statistically significant high PR expression in both acute and chronic infection cases compared to the control secretory cases (p<0.05). Conclusions: Schistosomiasis mansoni seems to have an important impact on the hormone expression of affected women. Further studies to explore the mechanism of such changes are recommended.

Key-words: Schistosoma mansoni, Endometrium, Hormone receptors, Oestrogen, Progesterone-immunohistochemistry.

INTRODUCTION

Schistosomiasis is one of the most serious parasitic diseases. More than 250 million people are infected with schistosomes in tropical or subtropical regions¹ and about 40 million women of child-bearing age are infected². The parasitic flukes have some unique biological features; complex life cycles, mechanisms to avoid host immune responses and an apparent reliance on host endocrine and immune signals to complete their development, maturation and egg production³. Despite the complex pathophysiological reactions during schistosomiasis, female genital infection is an almost entirely neglected disease entity that may give rise to considerable suffering among women in areas where schistosomiasis is prevalent. Several disorders in the female genital system due to both Schistosoma haematobium and mansoni have been reported, such as complications during pregnancy and related outcomes, menstrual disorders and other problems related to the female genital system. Several disorders in the female genital system due to both Schistosoma haematobium and Schistosoma mansoni have been reported, such as complications during pregnancy and related outcomes, menstrual disorders and other problems related to the female genital system. Although infertility is determined to be the result of Schistosoma infection⁴, the responsibility of schistosomiasis in female infertility is controversial.

Little is known regarding the hormonal changes associated with such infection in women. Parasites can alter hormone concentrations in their hosts, including estrogen and progesterone. Therefore, studies concerning host hormones responses to infection and the extent to which changes occur in endocrine-immune interactions following infection, mediated by the host or the parasite, are still recommended⁵.

Estrogen and progesterone receptors are members of the steroid receptor super family that binds the corresponding hormones that are expressed in the uterus, ovary, vagina and fallopian tubes in the female genital system. These receptors play a central role in women’s fertility cycles and...
were counterstained with Mayer’s haematoxylin and cover-slipped using a permanent mounting media. Estrogen and progesterone receptor expressions were evaluated as the percentage of positive nuclei in epithelial cells of the observed field in control, acute and chronic schistosomiasis specimens, observed at high magnification (400x). Cases were classified as ER and PR positive when more than 10% of cells exhibited positive nuclear epithelial staining. The mean of three fields were chosen from each slide, so as to best reflect the overall immunostaining of the hormone receptors contained on the entire slide. The two observers who examined the slides were blinded regarding the clinicopathological data.

**Statistical analysis**

Statistical analysis was performed by one way analysis of variance (ANOVA) followed by Turkey’s HSD (honestly significant difference) pair wise comparisons. The level of significance was set at p< 0.05 throughout the study. Current SPSS (version 13) statistical package was used in all statistical analysis. For each hormone receptor expression, Paired-Samples t test was used to detect significant differences between different groups.

**RESULTS**

Sections of control mice uterus showed either the proliferative or the secretory phases of endometrium. The proliferative endometrium was relatively thin, consisting of stratum basalis and stratum functionalis (stratum spongiosum and stratum compactum). The glandular epithelium exhibited basally located nuclei and prominent nucleoli, while the stroma was highly cellular and devoid of collagen fibers. Some sections showed the convoluted tubular glands of the late proliferative phase with the pseudo stratified appearance. The secretory endometrium showed irregular configuration of the glands that were lined by tall columnar epithelium with basal vacuolation. The glands of the late secretory phase appeared tortuous, coiled and contained copious thick glycogen secretion. The stroma appeared highly vascular with the interstitial fluid accumulated between the cells (Figure 1, A and B).

After acute and chronic *S. mansoni* infection, although no granulomas were observed, the endometrium showed only the late proliferative phase with the highly cellular stroma and the convoluted tubules that showed no secretion in their lumina (Figure 1, C and D). Nuclear epithelial expression for ERs showed negative reaction in all the groups studied, whereas PR expression was negative only in the control secretory group and positive in all the other groups (Figure 2, 3).

Hormone receptor expression (ER and PR) showed statistically significant differences among the groups (p< 0.05) (Table 1). Statistically significant low ER hormone expression occurred in chronic infection cases compared to that of control

![Figure 1](image1.png)

FIGURE 1 - Photographs of histopathological sections from the uterine of control and infected groups: A) Proliferative endometrium with highly cellular stroma, some convoluted tubular glands of the late proliferative phase can be observed, B) The secretory endometrium with highly vascular stroma and coiled irregular glands that contain glycogen secretion, C) Acute and D) Chronic *Schistosoma* infection showing the late proliferative endometrium with the highly cellular stroma and the nonsecretory convoluted tubules (HE, 200x).
### TABLE 1 - Hormonal receptor expression* in control, acute, and chronic schistosomiasis using anti-estrogen (ER) and anti-progesterone (PR) antibodies.

<table>
<thead>
<tr>
<th></th>
<th>Control proliferative (n=6)</th>
<th>Control secretory (n=6)</th>
<th>Acute schistosomiasis (n=8)</th>
<th>Chronic schistosomiasis (n=8)</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>ER receptors</td>
<td>3.98 ± 0.28</td>
<td>4.37 ± 0.34</td>
<td>4.32 ± 0.56</td>
<td>0.09 ± 0.05</td>
<td>0.00</td>
</tr>
<tr>
<td>PR receptors</td>
<td>84.95 ± 2.84</td>
<td>7.89 ± 0.44</td>
<td>83.61 ± 1.51</td>
<td>84.63 ± 2.44</td>
<td>0.00</td>
</tr>
</tbody>
</table>

*Values are presented as means ± standard error of mean. **ER:** estrogen receptor, **PR:** progesterone receptor.
proliferative, control secretory and acute infection cases (p=0.000). No statistically significant differences were observed between acute infection and any of the control cases (Table 2). In contrast, statistically significant higher PR expression occurred in both acute and chronic infection cases compared to the control secretory cases (p=0.000) (Table 2).

### Table 2 - Statistical analysis of hormonal receptor expression in control, acute, and chronic schistosomiasis using Paired-Samples t test.

<table>
<thead>
<tr>
<th>Pairs</th>
<th>ER (t-test)</th>
<th>PR (t-test)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control proliferative - acute infection</td>
<td>0.817</td>
<td>0.765</td>
</tr>
<tr>
<td>Control proliferative - chronic infection</td>
<td>0.000*</td>
<td>0.361</td>
</tr>
<tr>
<td>Acute infection - chronic infection</td>
<td>0.000*</td>
<td>0.715</td>
</tr>
<tr>
<td>Control secretory - acute infection</td>
<td>0.718</td>
<td>0.000*</td>
</tr>
<tr>
<td>Control secretory - chronic infection</td>
<td>0.000*</td>
<td>0.000*</td>
</tr>
</tbody>
</table>

*significant at p<0.05

ER: estrogen receptor, PR: progesterone receptor.

### DISCUSSION

Schistosomiasis is a serious and highly prevalent helminthic infection related to water contact and poverty and affects approximately 250 million people living in tropical and subtropical areas. The number of women with schistosomiasis-related signs and symptoms is likely to be high and could easily involve millions of cases. Numerous published case reports have presented clinical features, often described as unusual, of genital schistosomiasis in female patients. Yet, it is surprising how sparse and unsatisfactory the literature is on this subject. Incidental schistosomiasis of various lesions has been cited in the literature due to both Shistosoma mansoni and haematobium; however, insufficient data is available regarding the clinical picture of genital schistosomiasis. Systematic research concerning comorbidities caused by genital involvement during the course of Schistosoma infection is currently sketchy and research on the possible impact of schistosomiasis on different genital problems is still unsatisfactory.

The genital system is usually affected by urinary schistosomiasis caused by haematobium species, with complications including ectopic pregnancies or infertility. Concurrently, genital problems due to S. mansoni have also been reported in the form of atrophy of the corpus luteum cells and nuclear alterations of the interstitial cells in the ovaries of mice. Farah et al. reported S. mansoni to be a contributing factor in diminished fecundity among women in endemic regions. Numerous authors have emphasized the relation between parasitic and hormonal status in female mice, concluding that not only can host hormones affect responses to infection, but parasites can have pronounced effects on hormone signaling within the host. Moreover, after studying endocrine-immune interactions during parasitic infection, Klein et al. confirmed the importance of the relation between the endocrine system and parasitic infection. The lack of data concerning hormonal changes associated with Schistosoma infection prompted our group to study the effect of S. mansoni on the hormonal profile of endometrium using immunohistochemical technique.

In the present study, although no direct evidence of S. mansoni infection was detected (no granulomas were observed), alterations in uterine hormone receptor expression were observed. Hormone receptors in the endometrial epithelial cells showed statistically significant higher PR expression in both acute and chronic infection cases compared to the control secretory cases. This corroborated the histopathological results of the present study, where no secretory glands were observed in the endometrium in either acute or chronic cases, and reflected the expected low progesterone level in both cases.

Regarding ER expression, chronic cases showed statistically significant low ER hormone expression compared to that of both control proliferative and secretory cases, whereas no significant changes occurred in ER expression in acute infection cases compared to any of the control cases. This also coincides with the detection of late proliferative endometrium, which was observed in this study, and reflects the expected high estrogen level in chronic infection cases. These results explore the negative impact of S. mansoni on the female genital system, even in the absence of direct lesional infectious stages. Such observations might explain the infertility that has been attributed to Schistosomiasis infection by some authors.

Hormonal alteration is also recorded in males during S. mansoni infection, since it suppresses testosterone production in male mice. Alterations in hormone receptors in the present study might be due to the toxic effect of such hemoperasites on female hormones. These results are in agreement with those obtained by Klein et al., who reported increased concentration of serum oestradiol and progesterone levels and serious complications in genital system during hemoparasite Plasmodium infection. To our knowledge, no data exists concerning the findings described in the present work and this study seems to be the first report regarding uterine estrogen and progesterone hormone receptors in cases of Schistosoma mansoni infection.

From the data obtained, it is clear that genital infection by schistosomiasis worsens the disease burden of women in the child bearing period. Pathological consequences can be damaging for the affected women. A lack of clinical awareness of genital schistosomiasis can lead to misdiagnosis and therefore false and ineffective therapy. Therefore, future studies concerning the effect of different species of schistosomes are recommended to current understanding and to explore the possible mechanisms of such changes during both acute and chronic infection.

### CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

### REFERENCES


