The Effect of Garlic on Some Parasitological Parameters and On Hepatic Tissue Reactions in Experimental Schistosomiasis Mansoni

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Abstract: This investigation was designed to appraise the antibilharzial efficacy of garlic. Adult male Swiss albino mice were allocated into two main groups; the first group was non-infected and killed after 6 weeks, while the second one was infected with S. mansoni cercariae and divided into two equal halves to be sacrificed at 6 and 12 weeks post-infection. Both, the non-infected and infected groups were divided into three subgroups: I. non-treated, II. treated with 50 mg/kg of garlic extract and III. treated with 100 mg/kg of garlic extract. Some parasitological parameters (including worm burden, tissue egg load and hepatic granulomas count and diameter) were measured. In addition, hepatic histological examinations were performed. In non-infected mice treated with garlic (50 mg/kg), the histological changes in the inspected organ were generally mild. Concurrently, the high dose of garlic (100 mg/kg) induced hepatocytic hypereosinophilia, vacuolation and necrosis, in addition to nuclear alterations. In infected non-treated mice, hepatic tissue manifested apparent features, including: vascular dilatation, bile ductule proliferation, periportal inflammation, granulomatous reactions and fibrosis, as well as Kupffer cell hypertrophy and pigmentation. Besides, liver cells suffered from cloudy swelling, vacuolation and necrosis. Treating infected mice with the low dose of garlic resulted in significant parasite eradication, considerable reduction in the number of ova/g tissue. In addition, a conspicuous suppression in granulomatous formation and diminutive histopathological changes were observed. Conversely, no major differences were detected in the parasitological or histological observations following the administration of 100 mg/kg of garlic after infection as compared to the non-treated infected groups. Therefore, we can conclude that garlic in low doses has a curative effect on S. mansoni infection and plays a role in ameliorating its pathological consequences, although at high doses the reversal of these effects is observed.

Key words: Schistosoma mansoni, garlic, mice, liver, egg, granuloma.

INTRODUCTION

Schistosomiasis is still one of the most prevalent epidemic diseases in Egypt and in other developing countries despite their great efforts undertaken for controlling it. Data indicate that schistosomiasis affects between 200 and 300 million people in 79 countries.

Treatment of schistosomiasis worldwide relies very heavily on praziquantel (PZQ). The effectiveness of this drug against schistosomes is well recognized but evidences are now accumulating that PZQ cannot prevent re-infection and may sometimes enhance it. Meanwhile, in endemic areas, repeated chemotherapy has resulted in the emergence of drug-resistant strains of schistosomes. The development of such resistance has drawn the attention of many researchers to alternative drugs of plant origin which may be helpful in the treatment of schistosomiasis.

Interest in the potential benefits of garlic has its origin in antiquity as it is one of the earliest documented examples of plants used for maintenance of health and treatment of several diseases. In recent times, the antihelminthic effect of garlic has been verified by many investigators. The only available literature on the schistosomicidal role of garlic was submitted by Zakhary and deals with the biochemical aspects of the issue. So, this study was carried out to confirm and assess the antischistosomal potency of garlic from other points of view, including its effect on some parasitological parameters and its efficacy upon hepatic tissue reactions.

MATERIALS AND METHODS

Host: A total number of 180 Swiss adult male albino mice (Mus musculus) of CD1 strain, weighing 18-22g, were purchased from the Schistosome Biological Supply Program (SBSP) unit, Theodore Bilharz Research Institute, Guiza, Egypt. The animals were
given access to water and standard diet *ad libitum* and were monitored daily for health status.

**Infection of Animals:** Mice of infected groups were exposed to 100±10 *S. mansoni* cercariae per mouse by the tail immersion method, modified by Oliver and Stirewalt.[46]

**Garlic Preparation, Dosage and Administration:** Fresh Egyptian garlic was purchased from local suppliers. Raw aqueous extract was prepared by homogenizing 100 g of peeled garlic cloves in 100 ml of distilled water. The garlic paste is squeezed out through a piece of cloth to obtain the extract. The doses 50 mg/kg body weight and 100 mg/kg b.wt of garlic were selected for this work. The tested doses were made by diluting the stock solution using distilled water and stored at -20°C until use. Garlic extract was given orally with an oesophageal tube, every other day for a month and half.

**Experimental Design:** The mice were allocated into two main groups: the first group was non-infected and killed after 6 weeks, while the second one was infected with *S. mansoni* and divided into 2 equal halves to be sacrificed at 6 and at 12 weeks post-infection. The non-infected and infected groups were divided into three subgroups: I. non-treated, II. treated with 50 mg/kg of garlic extract and III. treated with 100 mg/kg of garlic extract. Garlic was given orally every other day for 6 weeks, either from the first day of the experiment in mice killed after 6 weeks, or were given garlic from the beginning of the 7th week post-infection in mice killed after 12 weeks.

**Determination of the Percentage of Mouse's Body Weight Change:** The percentage of the weight change of mice during the experiment was calculated according to the following equation:

\[
\text{Weight change}\% = \frac{\text{The final body weight of observed mouse} - \text{The final body weight of normal mouse}}{\text{The final body weight of normal mouse}} \times 100
\]

**Perfusion and Worm Recovery:** Schistosome worms were recovered from the hepatic portal and mesenteric veins by perfusion technique described by Smithers and Terry.[45]

**Egg Count in Tissue:** The eggs in the liver and small intestine of infected mice were counted according to Cheever.[41]

**Histological Procedure:** After perfusion, small pieces from the liver of all groups were excised and fixed in aqueous Bouin’s solution for 24 hours. Liver sections, of 5 µm thickness, were stained using Mayer’s haematoxylin and 1% aqueous eosin.[36]

**Hepatic Granuloma Count and Diameter Measurement:** From each infected mouse, three hepatic paraffin sections were prepared and stained with haematoxylin and eosin. The sections were 5 µm thick and 250 µm distant from the preceding sections to avoid counting or measuring the same granuloma. The granuloma count was calculated as the number of granulomas in 5 successive fields using the low power of the light microscope. Measurements were done only for solitary granulomas containing a single egg in their centers using a calibrated ocular micrometer. The mean diameter (µm) of every granuloma was obtained by measuring two diameters of the lesion at right angles to each other.[46]. The mean granuloma diameter/group was calculated for about 50-100 lesions.

**Statistical Data Analysis:** The data obtained in the present work are represented as mean (X) ± Standard Error (SE). Statistical analysis of results was carried out using Student's t-test according to Bailey.[4].

**RESULTS AND DISCUSSION**

The percentages of the body weight change in infected non-treated mice and mice treated with the high dose of garlic (100 mg/kg b.wt) groups dropped significantly beyond normal values; but no marked changes in body weight were found in mice infected and treated with 50 mg/kg of garlic relative to the corresponding normal group (Table 1).

The perfusion of the hepatic portal and mesenteric veins of infected mice treated with garlic (100 mg/kg b.wt) revealed an insignificant reduction in the mean worm load by the 6th week post-infection. On the other hand, the mean worm count lowered significantly after treating infected mice with the low dose of garlic (50 mg/kg b.wt). Six weeks later, at the end of the 12th week of infection and after the application of the high dose of garlic, no noticeable changes in the count of the worms were found in comparison with the non-treated group. Meanwhile, a considerable decrease in the number of worms was recorded after the application of the low dose of garlic treatment. The results are shown in table 2.

The mean egg load in hepatic and ileal tissues of all infected groups revealed that at six weeks post infection and after treating infected mice with the high dose of garlic, there was no notable alteration in the number of eggs when compared to the corresponding infected control group. Conversely, mice treated with the low dose of garlic displayed a highly significant increase in the percentage of egg reduction, estimated by 95.1% in the liver and 96.4% in the small intestine.
Table 1: Effect of garlic on body weight (g) of non-infected mice and mice infected with *S. mansoni*.

<table>
<thead>
<tr>
<th>Groups of mice</th>
<th>6 weeks</th>
<th>Groups of mice</th>
<th>6 weeks</th>
<th>12 weeks</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-treated, non-infected Group</td>
<td>27.6±3.10</td>
<td>Non-treated, non-infected Group</td>
<td>27.6±3.10</td>
<td>34.5±2.41</td>
</tr>
<tr>
<td>Infected, non-treated Group</td>
<td></td>
<td>Infected, non-treated Group</td>
<td>19.1±0.55</td>
<td><em>(−30.8%)</em></td>
</tr>
<tr>
<td>Non-infected, treated with 50mg/kg garlic Group</td>
<td>27.2±3.93</td>
<td>Infected, treated with 50mg/kg garlic Group</td>
<td>25.2±3.56</td>
<td><em>(−8.7%)</em></td>
</tr>
<tr>
<td>Non-infected, treated with 100mg/kg garlic Group</td>
<td>20.9±0.52</td>
<td>Infected, treated with 100mg/kg garlic Group</td>
<td>17.1±2.67</td>
<td><em>(−24.3%)</em></td>
</tr>
</tbody>
</table>

* Values are expressed as means±SE.
* Numbers between parentheses indicate the percentage of change in comparison with the corresponding normal values.
* Asterisks indicate the significant difference of all groups versus normal group within the same column.

Table 2: Effect of garlic on worm burden in *S. mansoni*-infected mice

<table>
<thead>
<tr>
<th>Groups of mice</th>
<th>6 weeks post infection</th>
<th>12 weeks post infection</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infected, non-treated</td>
<td>11.2±2.7</td>
<td>5.9±1.5</td>
</tr>
<tr>
<td>Infected, treated with garlic (50mg/kg)</td>
<td>2.8±0.86</td>
<td>0±0</td>
</tr>
<tr>
<td>Infected, treated with garlic (100mg/kg)</td>
<td>5.6±2</td>
<td>1.7±0.9</td>
</tr>
</tbody>
</table>

Values are expressed as Means ± SEM. Numbers between parentheses indicate the percentage of change in comparison with the corresponding control. Asterisks indicate the significant difference versus the corresponding control; ** *P* ≤0.01, *** *P* ≤0.001.

Table 3: Effect of garlic on tissue egg load in mice infected with *S. mansoni*.

<table>
<thead>
<tr>
<th>Groups of mice</th>
<th>6 weeks post-infection</th>
<th>12 weeks post-infection</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infected, non-treated Group</td>
<td>1709±431.1</td>
<td>31019±7799</td>
</tr>
<tr>
<td>Infected, treated with garlic (50mg/kg)</td>
<td>84.4±26.9</td>
<td>5130±1406</td>
</tr>
<tr>
<td>Infected, treated with garlic (100mg/kg)</td>
<td>1281.4±123</td>
<td>2593±1898</td>
</tr>
</tbody>
</table>

Values are expressed as means ± SE. Numbers between parentheses indicate the percentage of reduction in comparison with the corresponding control infected values. Asterisks indicate the significant difference of all groups versus control infected group within the same column, ** *P* ≤0.01, *** *P* ≤0.001.

Table 4: Effect of garlic on the number and diameter of hepatic granulomas in mice infected with *S. mansoni*.

<table>
<thead>
<tr>
<th>Groups of mice</th>
<th>6 weeks post-infection</th>
<th>12 weeks post-infection</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infected, non-treated Group</td>
<td>13.6±0.93</td>
<td>235.1±93</td>
</tr>
<tr>
<td>Infected, treated with garlic (50mg/kg) Group</td>
<td>0.83±0.17</td>
<td>6±0.45</td>
</tr>
<tr>
<td>Infected, treated with garlic (100mg/kg) Group</td>
<td>10.2±0.49</td>
<td>229.7±9.1</td>
</tr>
</tbody>
</table>

Values are expressed as means ± SE. Numbers between parentheses indicate the percentage of reduction in comparison with the corresponding control infected values. Asterisks indicate the significant difference of all groups versus control infected group within the same column, *P* ≤0.05, ** *P* ≤0.01, *** *P* ≤0.001.
compared to the corresponding infected control group. Later, after other 6 weeks, no statistically significant difference was noticed in the egg count in both liver and intestine in garlic high dose-treated mice when compared to the control infected group. Once again, 50 mg/kg garlic application showed a highly significant reduction in the total number of eggs per gram of tissue as compared to that of the corresponding infected control group (Table 3).

In all treated mice, a significant reduction in the number of the hepatic granuloma was observed when compared to the corresponding untreated infected group values (Table 4).

It is clear, as shown from the same table, that the diameters of the developing liver granulomas were significantly suppressed after treating mice with garlic (with exception of mice treated with 100 mg/kg b. wt and sacrificed on the 6th week post-infection) and the significance was higher in groups treated with 50 mg/kg b. wt than those treated with 100mg/kg b. wt.

Liver tissue is divided into lobules, the corners of which are occupied by the portal triads, each with a venule, an arteriole and a bile ductule. The lobules are made up of polyhedral hepatocytes organized as hepatic strands directed from the periphery of the lobule to a central vein at the core of each lobule and they anastamose freely forming labyrinthine and sponge-like structure. The space between these strands contains sinusoids which contain phagocytic cells known as von Kupffer cells (Fig. 1).

In comparison to the regular lobular structure of the liver or the hepatic strand organization, no apparent changes were observed in non-infected mice treated with the low dose of garlic (50 mg/kg). The tributaries of the hepatic artery and hepatic portal vein as well as bile ductules did not seem to have any obvious injuries. It was also noticed that Kupffer cells were hypertrophied and the cytoplasm of some liver cells appeared slightly vacuolated (Fig. 2). On the other hand, a general hepatic impairment, including necrosis and degeneration, was observed after the administration of 100 mg/kg of garlic to normal mice. Consequently, the degenerated and necrotic tissue induced remarkable changes in the normal organization of the hepatic strands. Centrilobular liver cells suffered to a great extent from hypereosinophilia, whereas periportal ones were extremely vacuolated and contained necrotic foci. Nuclei of these cells manifested distinct changes comprising pleomorphism and pyknosis (Fig. 3).
In non-treated mice, six weeks post-schistosomal infection, a high degree of disorganization in the hepatic lobular structure was observed, as no liver cords could be followed. This feature is presumably attributed to the emergence of granulomas (Fig. 4). Portal venules were dilated to some extent and their endothelial lining became detached at certain localities. Besides, most of the portal spaces were infiltrated with inflammatory cells. Additionally, bile ductules suffered from proliferation (Fig. 5). Schistosomal pigments derived from blood ingested by adult worms were deposited in the hypertrophied Kupffer cells (Fig. 6). Furthermore, hepatocytes were obviously altered. Some of them were homogenously stained indicating distinct signs of cloudy swelling. Other cells showed an advanced degree of injury symptomized by cytoplasmic vacuolation. Such vacuolations apparently represent hydropic degeneration. Additionally, a considerable number of these cells became distinctly necrotic. Nuclei of these injured liver cells became pleomorphic and pyknotic, as well as some of them appeared remarkably karyolyzed (Fig. 6).

After 12 weeks of infection, the previous symptoms of schistosomiasis were well pronounced in the examined specimens of infected non-treated mice. The liver tissue totally lost its ordinary configuration as a result of the presence of masses of fibrous granulomas (Fig. 7). The major hepatic pathological features observed at this stage of the disease were portal fibrosis, dilatation and diffuse inflammation, adding up the proliferation of the bile ductules (Fig. 8). Most parenchymal cells exhibited clear signs of

Fig. 4: Photomicrograph of liver section of *S. mansoni*-infected, non-treated mouse at the 6th week post infection showing the hepatic lobular distortion and multiformation of granulomas (G). Bar = 50 µm.

Fig. 5: Photomicrograph of liver section of *S. mansoni*-infected, non-treated mouse at the 6th week post infection showing the accumulation of inflammatory cells (asterisks) in the vicinity of the portal vessel (pv) which has a ruptured lining epithelium (arrow heads). Proliferated bile ductules (bd) are also noticed. Bar = 50 µm.

Fig. 6: High magnification of the mid-zonal region of liver section of *S. mansoni*-infected, non-treated mouse at the 6th week post infection showing noticeable disorganization of the parenchymatous tissue. Focal necrosis (ns) is observed among hepatocytes (H) manifesting clear signs of vacuolar degeneration. Nuclear pleomorphism, pyknosis and karyolysis (N) are common. Kupffer cells (K) are distinctly hypertrophied and some of them are recognized by the engulfed parasite pigments (arrow heads). Bar = 50 µm.

Fig. 7: Photomicrograph of liver section of non-treated mouse at the 12th week post infection showing plenty of granulomas (G) devastating the regular hepatic structural pattern. Bar = 50 µm.
hypereosinophilia and vacuolar degeneration. Besides, multifocal necrotic areas were evident especially at the margins of the granulomas. The nuclei of some hepatocytes displayed clear features of karyolysis and karyorrhexis. Other nuclei were markedly pyknotic to the extent that they could not show any of their structural details (Figs 8 and 9). The granulomatous size was rather larger than before and marked concentric fibrosis with many fibroblasts encircled the eggs that underwent distinct disintegration. The granuloma was surrounded by a cuff composed predominantly of lymphocytes, plasma cells, eosinophils and collagen fibers (Fig. 9).

The ordinary liver lobular architecture was relatively restored after the application of 50 mg/kg of garlic to mice infected and killed 6 weeks post-infection (Fig. 10). Granulomas of this group were few and smaller than those of the corresponding infected non-treated group. Hepatocytes, even in the vicinity of granulomas, were unharmed. Sinusoids contained leukocytes; however, Kupffer cells were neither hypertrophied nor loaded with pigments (Fig. 11). In mice sacrificed 12 weeks post-infection and treated with 50 mg/kg of garlic, the liver lacked its familiar
Fig. 12: Photomicrograph a liver section of infected mouse treated with the low dose of garlic and sacrificed at the 12th week post infection showing a reduced number of cellular granulomas (G) in comparison to that of the corresponding control infected group. Bar = 50 µm.

Fig. 13: High magnification of the centrilobular area in a liver section of infected mouse treated with the low dose of garlic and sacrificed at the 12th week post infection revealing intact and well organized hepatic strands radiating from a central vein (cv). Prominent Kupffer cells (K) and infiltrative cells are seen in the associated sinusoids (s). Hepatocytes possess slightly enlarged nuclei (N). G, a part of a granuloma. Bar = 50 µm.

Fig. 14: Photomicrograph a liver section of infected mouse treated with the high dose of garlic and sacrificed at the 6th week post infection showing several cellular granulomas (G) among disorganized hepatic lobules. Bar = 50 µm.

Fig. 15: A part of a magnified granuloma (G) in a liver section of infected mouse treated with the high dose of garlic and sacrificed at the 6th week post infection showing a degenerated bilharzial egg surrounded with inflammatory cells (asterisks). Hepatocytes (H) are vacuolated and necrotic, with karyolytic nuclei (N). Kupffer cells are enlarged and loaded with pigments (arrow heads). Bar = 50 µm.

regular architecture despite the presence of small number of little cellular granulomas (Fig. 12). Blood vessels became distended but their linings were yet intact to a large extent. These vessels were surrounded by mild lympho-plasmacytic infiltration. Hepatic strands were well organized and central veins appeared normal. Prominent Kupffer cells and infiltrative cells were seen in the associated sinusoids. Hepatocytes were not greatly changed but their nuclei were rather larger than normal (Fig. 13).

In mice treated with 100 mg/kg of garlic and sacrificed 6 weeks post-infection, the hepatic tissue showed apparent degenerative alterations (Fig. 14). The normal architecture was lost by the appearance of numerous granulomas in response to infection. The endothelial lining of most portal venules was damaged and the venules themselves were deformed and inflamed. It was clearly observed that liver cells were vacuolated and many necrotic foci were seen especially near the granulomas. Some of these cells possessed karyolyzed nuclei. Kupffer cells appeared hypertrophied and loaded with pigment (Fig. 15). In 12-week-infected mice treated with the high dose of garlic (100 mg/kg), the standard architecture of liver lobules was no longer preserved as numerous large granulomas invaded the parenchymatous tissue and most of the portal spaces were greatly dilated (Fig. 16). The endothelia of the portal vein tributaries were damaged at several localities coupled with high perivascular inflammation. Kupffer cells were very hypertrophied and loaded with
Nevertheless, the current results are in consistence with 30 mg/kg/day of garlic extract for a week.

In this regard, Nakagawa et al.[44] mentioned that raw garlic juice at a dose of 5 ml/kg resulted in death of rats due to stomach injury and the surviving rats exhibited swelling of the liver cells and reduction in erythrocyte count with various morphological changes after 3 and 8 days. Joseph et al.[31] studied the effect of 500mg/kg of aqueous garlic extract and garlic oil on liver of rats and found focal areas of hepatocytic necrosis and inflammatory cell infiltration. In addition, nuclear pyknosis and Kupffer cell hyperplasia were observed. Another study by Augusti[8] showed that prolong feeding of rats with large quantities of uncooked garlic resulted in anemia, weight loss and failure to grow due to lysis of red blood cells. Also, Hemmaid and Rahmy[29] reported that administrating garlic powder (equivalent to 70 mg/kg of raw garlic) to rats for 10 days induced several pathological changes represented by severe hepatocytic vacuolation, necrosis, nuclear abnormalities and Kupffer cell hypertrophy. Moreover, Soliman[55] studied the effect of 100 mg/kg/day of garlic for 21 days on rats and mentioned that most hepatocytes appeared vacuolated with pyknotic nuclei, while the others showed deeply eosinophilic homogenous cytoplasm.

Moreover, this contradictory effect of 100 mg/kg of garlic compared to that of the low dose could be due to the action of some garlic components, such as diallyl sulfide and diallyl disulfide. Such components are known to induce cytotoxic activity in case of over dose administration[8,26,29,5].

Schistosomiasis causes various adverse effects. One of these effects is growth retardation which could be due to the histopathological lesions which cause disturbance in many metabolic activities. Livers of infected animals suffered from disturbed lobular organization. The great majority of the hepatocytes manifested vacuolation, cloudy swelling and necrosis. Kupffer cells were also evidenced in the present material to be markedly hypertrophied and loaded with pigment. In addition, bile ductular hypertrophy and perivascular inflammation were very common in liver sections of infected, non-treated mice. With the chronicity of infection, some hepatic cells embodied a number of large, clear vacuoles and fibrosis began to affect the granulos and portal spaces. This hepatic tissue destruction results in the inability of

Discussion: In non-infected mice treated with garlic (50 mg/kg body weight) the body weight gain was more or less similar to the normal value. Besides, no histopathological lesions were encountered in the examined hepatic tissues except for insignificant hepatocytic vacuolations and Kupffer cell hypertrophy. In this regard, Nakagawa et al.[44] reported no histological changes in the liver of mice given 30 mg/kg/day of garlic extract for a week. Nevertheless, the current results are in consistence with those of Alnaqeeb et al.[5] and Soliman[55] who reported the presence of mild vacuolations in liver cells after treating rats with 50 mg/kg/day of raw garlic extract for 3 weeks.

The reduction in the body weight of non-infected mice treated with 100 mg/kg b. wt of garlic could be attributed to the deteriorated physiological status as a result of the induced histopathology in these treated mice. The present work revealed that liver sections of mice treated with this garlic dose displayed hepatocytic hypereosinophilia, vacuolization and necrosis accompanied with nuclear alterations. Such hepatic lesions usually result in abnormalities in the main functions of the liver[57]. In this respect, Nakagawa et al.[44] mentioned that raw garlic juice at a dose of 5 ml/kg resulted in death of rats due to stomach injury and the surviving rats exhibited swelling of the liver cells and reduction in erythrocyte count with various morphological changes after 3 and 8 days. Joseph et al.[31] studied the effect of 500mg/kg of aqueous garlic extract and garlic oil on liver of rats and found focal areas of hepatocytic necrosis and inflammatory cell infiltration. In addition, nuclear pyknosis and Kupffer cell hyperplasia were observed. Another study by Augusti[8] showed that prolong feeding of rats with large quantities of uncooked garlic resulted in anemia, weight loss and failure to grow due to lysis of red blood cells. Also, Hemmaid and Rahmy[29] reported that administrating garlic powder (equivalent to 70 mg/kg of raw garlic) to rats for 10 days induced several pathological changes represented by severe hepatocytic vacuolation, necrosis, nuclear abnormalities and Kupffer cell hypertrophy. Moreover, Soliman[55] studied the effect of 100 mg/kg/day of garlic for 21 days on rats and mentioned that most hepatocytes appeared vacuolated with pyknotic nuclei, while the others showed deeply eosinophilic homogenous cytoplasm.

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Liver to metabolize proteins and fats\textsuperscript{[37]}, or to utilize glucose and store glycogen\textsuperscript{[61,23,17,62]}. Collectively, these consequences lead to anemia and loss of body weight.

Incidentally, the present hepatic histopathological observations confirm those recorded by the preceding workers\textsuperscript{[10,7,5,9,19,20,33,25,35,28,14,64]}. Parastologically, the worm recovery in the present work was higher at the 6th weeks than at the 12th weeks post-infection in the infected (treated and non-treated) groups. This result is in line with that of Soliman et al.\textsuperscript{[54]} who found that, in both the Beni Suef and Guiza strains of Schistosoma mansoni, the worm recovery increase started from the 3rd week, reached the maximum at the 6th-8th week and then declined gradually until the 12th week. This could be due to the fact that on the 6th week post-infection all schistosomes complete their migration and maturation\textsuperscript{[41]} and after egg deposition large numbers of the adult worms begin to die\textsuperscript{[39]}. Considering hundreds of eggs laid by each worm pair every day, the increase of the egg count with the chronicity of the infection was expected. Egg load in the small bowel was higher than that detected in the liver of infected mice (either control or treated). In this respect, Saoud\textsuperscript{[40]} infected mice with three different strains of \textit{S. mansoni} and always found the greatest number of eggs in the small intestine, followed by the liver and then by the large intestine. Studies by Cheever\textsuperscript{[12]}, Tiboldi\textsuperscript{[60]}, Gerges et al.\textsuperscript{[24]} and Mahmoud et al.\textsuperscript{[97]} showed the same results.

Viable eggs lodged in tissues induced granulomatous reactions consisting chiefly of eosinophils, many lymphocytes, macrophages, peripheral plasma cells and few fibroblasts. The latter became predominant with striking concentric fibrosis seen in the central and intermediate areas of the old granulomas. At this stage, the granuloma diameter was somewhat larger, most likely because of the peripheral cuffing composed mainly of lymphocytes and, to a lesser extent, of plasma cells, macrophages and eosinophils. These findings were similar to those obtained by Schiller and Haese\textsuperscript{[50]}; Riad\textsuperscript{[67]} and Doughty and Phillips\textsuperscript{[15]} in their light microscopic study and by Stenger et al.\textsuperscript{[56]}, Bogitsh\textsuperscript{[7]}, Smith\textsuperscript{[51]}, Epstein et al.\textsuperscript{[21]}, Mansy\textsuperscript{[38]} and Mohamed and Fares\textsuperscript{[40]} using electron microscopy.

In the current study, treating infected mice with garlic in a dose of 50 mg/kg caused an apparent body weight gain and a significant eradication of worms if compared to the corresponding infected control group. This may be perhaps because of the role of garlic as an immune enhancer. Likewise, there was an extensive reduction in the tissue egg count and hepatic granuloma in the mice of these groups relative to the corresponding infected control groups. According to El-Gowhary et al.\textsuperscript{[18]}, several factors may be put forward to explain such reduction: the low worm load, the diminished fecundity of worm pairs and the increased rate of egg excretion as a result of egg death. Histological examination revealed a notable suppression in the granulomatous diameter and a remarkable histological recovery. The commonly encountered hepatic alterations were periportal inflammation, sinusoidal infiltration and Kupffer cell activation. The reduced parasite recovery and, so, the decreased egg production interpret these findings as Cheever\textsuperscript{[11]} stated that hepatic fibrosis in infected mice is related to egg numbers, i.e. mice with heavier infection have more total hepatic fibrosis. Also, in accordance to Chesney et al.\textsuperscript{[13]}, who described the infiltration of circulating fibroblasts into granulomas and speculated that these cells may be important for attracting lymphocytes as well as forming collagen, the fibrinolytic effect of garlic may possibly explain the reduction in the diameter and cellularity of the granuloma of this group.

Subsequent to the higher garlic dose application (100 mg/kg b.wt), most results (parasitological and histological) were insignificantly different comparable to those of the analogous infected control groups. This could be explained on the basis of the induced-hepatocytic damage in livers of non-infected mice treated with this high dose of garlic. Such injury might affect the synthesis of many antioxidant enzymes, detoxifying agents and immunoglobulins produced by the liver, which, doubtless, leads to a drop in the immunity making it a hard task to challenge infection to the same extent the low dose does.

Garlic (100 mg/kg b.wt) administration was marked to bring about several vascular alterations in the injected hepatic section. The hepatocytes were obviously vacuolated and necrotic; at the same time their nuclei acquired certain pyknotic and, to a lesser magnitude, karyolytic signs. Hypertrophy and pigmentation of Kupffer cells were also observed. Nonetheless, no publications were devoted to the influence of garlic on the structural characteristics of the organs of schistosomiced animals to compare the present results with.

In conclusion: garlic in low doses has a therapeutic effect on established \textit{S. mansoni} infection and plays a role in ameliorating its pathological consequences, although at high doses of garlic the reversal of these effects is observed. Meanwhile, the present findings suggest the need for initiating more studies to understand the principal reasons for the antibilharzial effect of garlic and to adjust its dose for optimal
benefit and prevention from its overdose undesirable side effects.

REFERENCES


