Therapeutic Potential of Coriander (Coriandrum sativum) Seeds Extract Treatment on Hematological and Biochemical Parameters in Healthy and Trichinella spiralis Infected Mice

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The smallest known human nematode parasite is Trichinella spiralis (T. spiralis). A parasitic zoonosis that can be found anywhere in the world is trichinellosis. When a host consumes raw meat contaminated with Trichinella larvae, the larvae mature into adults in the host's stomach in 5 to 6 days. The females subsequently give birth to additional larvae, some of which eventually make it to muscle cells after travelling through blood channels. These cells allow infective larvae to survive for years because, after two to three weeks, the larvae become infectious and the host cell transforms into a nurse cell. The current study used parasitological assessment to count adult worms and encysted larvae at 5 weeks after infection (5WPI) to determine the effect of coriander seeds extract (CSE) against encysted larvae further to hematological and biochemical tests. Current results revealed that; Trichinella spiralis experimentally infection (5WPI) induced significant elevation in white blood cells (WBCs) counts, neutrophil, monocyte, eosinophil percentage, alanine transaminase (ALT), aspartate transaminase (AST), alkaline phosphatase (ALP), urea, creatinine and a significant depletion hemoglobin (Hb), red blood cell (RBC) count, hematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), platelet count (PLT), lymphocytes percentage, albumin, total proteins as compared to control and CSE groups. Meanwhile, treatments of 5WPI with CSE (5WPI+CSE) revealed a significant reduction in the number of Trichinella spiralis encysted larvae and improvements in all hematological parameters, liver and kidney functions as compared to 5WPI. We can conclude that; CSE could be an alternative method for treatment against trichinellosis.

Keywords: Coriander seeds; kidney functions; Liver; Mice; Trichinella spiralis.

Trichinellosis is a parasitic zoonosis that is spread through ingesting raw or undercooked meat that has been contaminated with Trichinella spiralis larvae.1-3 It has three clinical phases: intestinal, migratory, and muscular. Pork and its products are the main sources of infection.4-6 According to Saad et al.7 and Abou Rayia et al.8, T. spiralis has the unusual capacity to change the infected muscle cell into a new kind of cell known as a nurse cell in the host body. Trichinellasag spiralis has been frequently used as an experimental model to determine the effects of numerous anthelmintic agents because it can develop into adult, migratory, and encysted stages in the same host and infects a wide range of mammalian hosts.9 According to Gottstein et al.10, mebendazole is the standard therapy and the main anthelmintic medication for the treatment of trichinellosis in the majority
of cautery and in KSA. However, according to Caner et al., they exhibit modest efficacy against encapsulated larvae, a high level of resistance, and restricted bioavailability. Medical plants create a wide range of chemical components that can treat a wide range of illnesses. The seeds of the fragrant, carminative, stomachic, and antispasmodic annual plant Coriandrum sativum L. (coriander, family Apiaceae) are used in medicine to treat abdominal symptoms such as dyspepsia, and gastralgia. To avoid stomach aches, they are also a component in laxative remedies. Thanks to the identification of certain polyphenolic and antioxidant components present in the coriander plant, it has been shown to play a substantial role in the aetiology of a number of disorders. Coriander is used in folk medicine to treat rheumatism and joint discomfort as well as intestinal parasites. Because of this, the current investigation looked at how therapy with coriander affected hematological and biochemical variables in mice with Trichinella spiralis infection as well as healthy mice.

MATERIALS AND METHODS

Making coriander seed extracts (CSE)

In accordance with Moustafa et al., coriander seeds (CS) were powdered, soaked in boiling water for 24 hours, extracted, and then stored at -30°C in the dark until use.

Animals and Ethical Considerations

A total of 40 male Swiss male albino mice were employed in this investigation, and they appeared to be in good health and free of parasites. 10 mice each were divided into 4 equal groups (Gps) of mice. The mice were bought in Giza, Egypt, from NRC. Prior to the trial, mice were kept at our faculty’s animal home for a week. They were kept in controlled lighting conditions with a conventional mouse feed and access to water at all times. Experimental design and animal groups

Gp1: Control group (Control), in which normal healthy non-infected mice.
Gp2: Coriandrum Gp (CSE) where mice treated orally (50 mg/kg body weight) daily for one week.
Gp3: Infected group (5WPI), in which mice were challenged with 300 larvae of Trichinella spiralis for 5 weeks.
Gp4: Post treated group (5WPI+CSE), in which mice were challenged with 300 larvae of Trichinella spiralis muscle larvae for 5 weeks then treated with CSE for 1 week post infection.

Sample collection

At the end of the experiment, overnight fasted rats will be anaesthetized with diethyl ether, dissection and blood will be collected in EDTA tubes for CBC determination and half of tubes were centrifuged at 3000 g for 20 min. plasma will be carefully separated, each of samples will label and kept at -20°C until parasitological, and biochemical analysis.

Isolation and infection of Trichinella spiralis muscle larvae

T. spiralis muscle larvae were obtained from laboratory bred infected rats in parasitology unit, Faculty of Medicine, Tanta University. Larval preparation and extraction of inoculums were made after Dunn and Wright. Five weeks after infection (5WPI), 1% pepsin and 1% concentrated HCL are added to warm tap water to create an artificial gastric juice that is used to digest the muscles of infected mice. The mixture was incubated for 2 hours at 37°C while being continuously stirred by an electric stirrer. The digest was then filtered by sieve (50mesh/cm²), then by sieve (200mesh/cm²). After being collected, the larvae were rinsed in tap water two to three times before being suspended in a conical flask for 30 minutes to allow for sedimentation. Sediment larvae were counted microscopically while using a hemocytometer, and the supernatant fluid was discarded. Dead larvae were non-mobile and comma-shaped, whereas living larvae were coiled and motile. The Concentration of counted larvae in the fluid was adjusted to the appropriate dose for each rat, which is 0.25 ml of fluid containing 300 living larvae. Mice were starved for 12 hours before infection, then provided with 0.25ml of the infection orally by using a tuberculin syringe fitted with blunt, curved, 18-gauge needle to introduce infective larvae into mouse stomach.

Hematological studies

Complete blood picture (CBC) measurements were made in 2007 using a Nihon Kohden Corporation, Tokyo, Japan, automated hematology cell counter (serial number 11649, model Celltac, MEK-6410K). The following blood parameters were measured: mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin
concentration (MCHC), white blood cell count (WBC) and its differential, red blood cell (RBC) count, hemoglobin (Hb) level, hematocrit (HCT), and platelet count (PLT).

Liver and Kidney functions estimation

Reitman and Frankel’s 19 approach was used to measure the activities of ALT and AST in serum, whereas Belfield and Goldberge’s 20 method was used to measure the activity of ALP. Total protein concentration was calculated using Simonian’s method 21, whereas albumin concentration was calculated using Abd Eldaim et al.’s approach 22. According to Patton and Crouch (1977) urea and creatinine were estimated.

Statistical Analysis

Results were analysed using an adapted SPSS programme. Data are presented as mean ± standard error of mean (SEM). A one-way ANOVA followed by Dunnett’s test was used to analyse the differences between groups. Unpaired T-test was used to assess the level of statistical significance between the groups, with the threshold set at p<0.01.

RESULTS

Clinical signs

Only six out of twenty rats given an injection of *T. spiralis* larvae exhibited the peri-orbital edema, dyspnea, and respiratory issues that are indicative of trichinellosis during the infection period (Fig. 1). Figure 1 Photomicrograph revealed skeletal muscle fibers with marked fibrosis and marked inflammatory cellular infiltration with massive numbers of *T. spiralis* encysted encapsulated larvae. Table 1 revealed the mean number of *Trichinella spiralis* encysted larvae in mice diaphragms after 5 weeks of infections and in one week treatments with CSR after 5 week post infection with *Trichinella spiralis* (5WPI+CSE). 5WPI+CSE induced reduction in the number of *Trichinella spiralis* encysted larvae.

Table (2) revealed a significant decrease in the levels of Hb, RBCs count, HCT, MCV, MCH, PLT, lymphocytes percentage and significant decrease in WBCs counts, neutrophil, monocyte and eosinophil percentage when compared to 5WPI group (Table 2).

Fig. 1. Photomicrograph of skeletal muscle sections stained with Haematoxylin & Eosin revealed muscle fibers with marked fibrosis and marked inflammatory cellular infiltration (arrow heads) with massive numbers of *T. spiralis* encysted encapsulated larvae (arrows).
Table 3 showed a significant elevation in the activities of ALT, ALP, AST, urea, creatinine and a significant depletion in the levels of albumin, total proteins in 5WPI group as compared to control and CSE groups. Meanwhile, treatments of 5WPI with CSE (5WPI+CSE) revealed a significant depletion in the activities of ALT, ALP, AST, urea, creatinine and a significant elevation in the levels of albumin, total proteins as compared to 5WPI group.

**DISCUSSION**

It is still crucial to find novel ways to diagnose and manage zoonotic infections in developing nations, especially considering the possibility of human-animal contact there growing over time. With the ability to infect a wide range of mammals, including humans, *T. spiralis* is still regarded as one of the most dangerous and widely spread foodborne zoonotic nematodes. Drugs administered in trichinellosis patients include anthelmintics and steroids. Anthelmintics are the principal drugs for the treatment of Trichinellosis. A strong effort is currently being directed toward the development of an effective treatment against Trichinellosis.

These treatments have not proven their ability to fight and eliminate *Trichinella spiralis*

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**Table 1.** Changes in complete blood picture (CBC) parameters in different groups

<table>
<thead>
<tr>
<th></th>
<th>5WPI</th>
<th>5WPI+CSE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number mean ± SE</td>
<td>1849.3 ± 380.0</td>
<td>746.5 ± 129.1</td>
</tr>
<tr>
<td>Range</td>
<td>2250-1604</td>
<td>962-570</td>
</tr>
<tr>
<td>% of reduction</td>
<td>0 ± 0</td>
<td>59.7%</td>
</tr>
</tbody>
</table>

**Table 2.** Changes in complete blood picture (CBC) parameters in different groups.

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>CSE</th>
<th>5WPI</th>
<th>5WPI+CSE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hb (g/dl)</td>
<td>10.8±0.69</td>
<td>11.1±0.88</td>
<td>9.6±0.40</td>
<td>10.4±0.70</td>
</tr>
<tr>
<td>RBC (million/ul)</td>
<td>4.2±0.17</td>
<td>4.4±0.35</td>
<td>4.09±0.25</td>
<td>4.16±0.39</td>
</tr>
<tr>
<td>Hct %</td>
<td>35.6±2.20</td>
<td>36.3±2.07</td>
<td>31.7±1.98</td>
<td>34.3±1.33</td>
</tr>
<tr>
<td>MCV (fl)</td>
<td>84.2±6.04</td>
<td>83.0±5.39</td>
<td>77.5±4.55</td>
<td>82.5±4.05</td>
</tr>
<tr>
<td>MCH (pg)</td>
<td>25.5±1.60</td>
<td>25.2±0.91</td>
<td>23.5±1.14</td>
<td>4.74±0.33</td>
</tr>
<tr>
<td>MCHC (g/dl)</td>
<td>30.3±2.19</td>
<td>30.3±1.75</td>
<td>30.3±2.05</td>
<td>30.3±2.17</td>
</tr>
<tr>
<td>Platelets (10^9/ul)</td>
<td>735.0±11.88</td>
<td>741.0±11.5</td>
<td>559.0±9.35</td>
<td>618.0±10.23</td>
</tr>
<tr>
<td>WBC (10^9/ul)</td>
<td>6.2±0.44</td>
<td>6.1±0.42</td>
<td>11.45±1.16</td>
<td>8.9±0.60</td>
</tr>
<tr>
<td>Neutrophil %</td>
<td>29.0±1.06</td>
<td>30.6±1.82</td>
<td>36.0±1.75</td>
<td>33.9±2.08</td>
</tr>
<tr>
<td>Lymphocyte %</td>
<td>61.2±3.81</td>
<td>62.0±3.25</td>
<td>50.0±2.92</td>
<td>56.2±3.70</td>
</tr>
<tr>
<td>Monocyte %</td>
<td>6.3±0.51</td>
<td>5.0±0.40</td>
<td>8.0±0.55</td>
<td>5.2±0.64</td>
</tr>
<tr>
<td>Eosinophil %</td>
<td>3.5±0.28</td>
<td>2.4±0.17</td>
<td>6.0±0.49</td>
<td>4.7±0.29</td>
</tr>
</tbody>
</table>

*:* Significant difference from the control group at p < 0.05, #: Significant difference from the *Trichinella spiralis* infection (5WPI) group at p < 0.05.

**Table 3.** Changes in the liver and kidney functions in different groups

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>CSE</th>
<th>5WPI</th>
<th>5WPI+CSE</th>
</tr>
</thead>
<tbody>
<tr>
<td>ALT (U/I)</td>
<td>37.9±1.65</td>
<td>35.5±1.70</td>
<td>83.0±3.25</td>
<td>61.3±3.08</td>
</tr>
<tr>
<td>AST (U/I)</td>
<td>81.0±3.45</td>
<td>72.5±4.15</td>
<td>109.2±7.52</td>
<td>96.0±5.14</td>
</tr>
<tr>
<td>ALP (U/I)</td>
<td>102.5±6.75</td>
<td>93.9±5.42</td>
<td>125.0±8.76</td>
<td>116.2±8.51</td>
</tr>
<tr>
<td>Albumin (mg/dl)</td>
<td>4.11±0.39</td>
<td>4.25±0.28</td>
<td>3.05±0.31</td>
<td>4.74±0.33</td>
</tr>
<tr>
<td>Total protein (mg/dl)</td>
<td>5.86±0.42</td>
<td>6.11±0.48</td>
<td>3.85±0.29</td>
<td>4.54±0.35</td>
</tr>
<tr>
<td>Creatinine (mg/dl)</td>
<td>0.65±0.05</td>
<td>0.63±0.08</td>
<td>0.95±0.08</td>
<td>0.70±0.05</td>
</tr>
<tr>
<td>Urea (mg/dl)</td>
<td>31.9±2.14</td>
<td>28.5±2.65</td>
<td>43.5±2.31</td>
<td>33.0±2.83</td>
</tr>
</tbody>
</table>

*:* Significant difference from the control group at p < 0.05, #: Significant difference from the *Trichinella spiralis* infection (5WPI) group at p < 0.05.
effectively, so it was necessary to find new drug more effective for *T. spiralis* treatments. Consequently, the current research sought to the impact of coriander treatment as new treatments on hematological and biochemical parameters in healthy and *Trichinella spiralis* infected mice.

30% of infected rats with *Trichinella spiralis* showed clinical signs as peri-orbital edema, dyspnea, and respiratory problems without changes in body temperature during the infection time. Current results agree with Ribicich et al. 24 who reported that; only two of nine pigs inoculated with *T. spiralis* larvae showed clinical signs consistent with trichinellosis. Treatments of 5WPI with CSE for one week showed a 59.6% reduction in number of larvae encysted in diaphragms of infected rats. Our findings are consistent with those of Abu El Ezz 25 and Soliman et al. 26, who found that untreated rats' diaphragms had a large number of migratory larvae. Both in vitro and in vivo studies have shown that biological components found in CSE, including as polyphenols, tocopherols, and sterols, have potent anti-parasitic activities. 27,28

Current results showed that; infected rats with *Trichinella spiralis* for 5WPI induced significant decrease in the levels of Hb, RBC count, HCT, MCV, MCH, platelet count (PLT), lymphocytes percent and significant increase in WBCs, neutrophil, monocytes, eosinophil percent and the treatments with CSE improved these parameters. Low MCV and MCH means your hemoglobin production is less than normal. As a result, the number of healthy red blood cells also decreases, leading to anemia. Mean corpuscular volume and mean corpuscular hemoglobin are lower than the healthy range, it may indicate iron-deficiency anemia and microcytosis. Our findings are consistent with those of Ribicich et al. 24, who discovered that pigs implanted with 500 and 5000 larvae between 1 and 6 weeks after contracting *Trichinella spiralis* had lower haemoglobin values and higher white blood cell counts. Our results in the line of Sugane et al. 29 who reported that *T. canis* infection was induced elevation in WBCs count and eosinophil percent. Eosinophilia and infection severity are correlated. Additionally with Oto et al. 30, who investigated *T. canis*-caused eosinophilic meningo-encephalo-myelitis. On the first day after infection, a rise in eosinophils and basophils directly denotes the initiation of a primary-allergic reaction in the body. *T. spiralis* create a significant allergic reaction when they enter the body. 31 Allergy symptoms are brought on by the histamines that basophil cells produce. It is feasible to check the blood’s neutrophil count in addition to the clinical signs of invasion during *T. spiralis* infection and its early identification. This study shows that during the 5-week experiment, this marker is raised when compared to the control. According to Ovington and Behn 32, the presence of neutrophils and monocytes in the infiltrates of enclosing nurse cells indicates that an isolated rise in neutrophils is a sign of capsule development.

Our findings showed that, in comparison to control (healthy mice who were not infected with *T. spiralis*), *T. spiralis* infection (5WPI) caused a substantial increase in blood AST, ALT, ALP, urea, and creatinine levels and a significant decrease in serum total proteins and albumin. According to Gamble et al. 33 and Nada et al. 34, who confirmed that elevated AST and ALT is pointing to hepatic damage, while increased urea and creatinine is indicative of a kidney disease, these changes may be attributed to liver and kidney damages induced during larval migration. According to Saggu et al. 35, ALP is an enzyme that serves as a marker for the plasma membrane. Any of the two anomalies (an increase in normal levels or a decrease in normal levels) may result from damage to the biological membrane. This shows potential injury to the plasma membrane of the experimental rat tissues, and this conclusion is consistent with that of Adeyemi et al.36. Current study agreed with Mikhail 37 who find that *T. spiralis* infection induced in ALT and AST. Additionally, this investigation supported the findings of Basyoni and El-Sabah 38 and Soliman et al. 26 who reported that; After *T. spiralis* infection, levels of total proteins and albumin decreased. The decrease in total proteins and albumin levels may be caused by the migratory larvae damaging the liver parenchyma or by the metabolic byproducts of the parasites harming the liver.

**CONCLUSION**

*Trichinella spiralis* experimentally infection (5WPI) induced significant changes in hematological parameters, liver and kidney functions and the treatments of 5WPI with CSE...
(5WPI+CSE) induced a significant reduction in the number of *Trichinella spiralis* encysted larvae and improvements in all hematological parameters, liver and kidney functions.

**ACKNOWLEDGEMENT**

None.

**Conflict of Interest**

The authors declare no competing interests.

**Funding Sources**

There is no funding Sources.

**Data availability**

All the data and material were available. The data of this article are included within the article and its additional files.

**REFERENCES**

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