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**Schistosomicidal, antifibrotic and antioxidant effects of *Cucurbita pepo* L. seed oil and praziquantel combined treatment for *Schistosoma mansoni* infection in a mouse model**

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**Supplementary materials**

1. **Supplementary Table S1**
2. **Supplementary Table S2**
3. **Supplementary Figure S1**
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**Supplementary Table S1.** Effect of PSO with/without PZQ on male, female and coupled worms recovered from different studied groups.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  | Recovered worms | | | | Total reduction rate % |
| **Male** | **Female** | **Coupled** | **Total** |
| Infected control (GII) | 7.42 ± 0.71 | 3.5 ± 0.52 | 5.50 ± 0.53 | 16.42 ± 0.71 | - |
| PZQ-treated (GIII) | 0.39 ± 0.41\* | 0.3 ± 0.09\* | 0.4 ± 0.19\* | 1.1 ± 0.42\* | 93.3 |
| PSO-treated (GIV) | 0.52 ± 0.22\* | 0.25 ± 0.08\* | 0.72 ± 0.27\* | 2.0 ± 0.99\* # | 87.8 |
| PZQ + PSO treated (GV) | 0.13 ± 0.35 \* | 0.0 ± 0.0 \* | 0.0 ±0.0 \* # † | 0.13 ± 0.35 \* # † | 99.2 |
| *F-ratio* | 475.74 | 310.83 | 546.39 | 1066.68 |  |
| *P-value* | 0.000 | 0.000 | 0.000 | 0.000 |  |

Data are expressed as mean ± SD, (n = 8).

The percentage of reduction was calculated as follows:

Percent reduction = (value of infected controls – value of treated mice) / (value of infected controls) ×100

\* indicates significance (*P* < 0.05) vs. GII (infected control, IC), # indicates significance vs. GIII (PZQ treated, PZQ) and † indicates significance vs. GIV (PSO treated, PSO) one way ANOVA followed by Student-Newman-Kenls post-Hoc test.

**Supplementary Table S2.** Effect of PSO with/without PZQ on MAD, SOD in liver homogenate and serum liver enzymes activities among the studied groups.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  | MAD  (nmol/g) | SOD  (U/mg) | AST  (U/L) | ALT  (U/L) | ALP  (U/L) |
| Normal control (GI) | 24.16 ± 2.82 | 100 ± 0.24 | 71.1 ± 7.69 | 32.81 ± 2.23 | 107.7 ± 7.42 |
| Infected control (GII) | 45.11 ± 7.21 **‡** | 60.86 ± 0.36 **‡** | 371.8 ± 29.0 **‡** | 83.71 ± 5.15 **‡** | 191.65 ± 27.97 **‡** |
| PZQ-treated (GIII) | 30.59 ± 0.92 **‡ \*** | 79.91 ± 0.27 **‡** \* **†** | 193.3 ± 13.58 **‡** \* | 59.13 ± 9.58 **‡** \* | 153.47 ± 33.91 **‡** \* |
| PSO-treated (GIV) | 29.53 ± 0.77 **‡ \*** | 89.45 ± 0.79 **‡ \* #** | 168.07 ± 29.75 **‡ \*** | 62.81 ± 9.07 **‡** \* | 131.89 ± 33.9 \* |
| PZQ + PSO treated (GV) | 25.55 ± 2.81 **\* # †** | 99.82 ± 0.16 **\* # †** | 99.6 ±7.93 **‡ \* # †** | 42.8 ± 7.88 **‡ \* # †** | 109.95 ± 11.19 \* # |
| *F-ratio* | 40.12 | 11738.87 | 272.7 | 57.38 | 14.89 |
| *P-value* | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 |

Data are expressed as mean ± SD, (n = 8).

The percentage of reduction was calculated as follows:

Percent reduction = (value of infected controls – value of treated mice) / (value of infected controls) ×100

‡ indicates significance (*P* < 0.05) of the value vs. GI (normal control, NC), \* indicates significance vs. GII (infected control, IC), # indicates significance vs. GIII (PZQ treated, PZQ) and † indicates significance vs. GIV (PSO treated, PSO) by one way ANOVA followed by a post-Hoc test.

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**Supplementary Fig. S1.** Picro-Sirius red stained liver sections from *S. mansoni* infected mice groups. a) Infected control group, showing a granuloma (G) with condensed bright red PSR stained thick collagen fibres (Cf) surrounding a degenerated *S. mansoni* egg (DE). b) PZQ treated group, showing a granuloma with less condensed faint red collagen fibres and a degenerated egg. c) PSO treated group showing less defined granuloma around the ova (O) with greenish to red stained thin collagen fibres (arrows). d) Combined PSO-PZQ treated group showing a small granuloma with absence of the egg and scanty greenish stained thin collagen fibres (× 400).

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**Supplementary Fig. S2.** Angiogenesis in the liver sections from *S. mansoni* infected mice groups stained immunohistochemically with anti-CD34+. a) Normal control group, showing negative CD34+ reaction. b) Infected untreated group showing strong expression within and around the granulomas, as well as in the inter-granulomatous tissues which show proliferating blood vessels. c) PZQ treated group showing strong reaction around the granulomas with vascular proliferations. d) PSO treated group showing moderate reaction around the granuloma with also vascular proliferation. e) With PSO-PZQ combined treatment there was a moderate positive reaction with signs of restoration of the normal architecture with vascular proliferation, additionally, a small granuloma size were observed (×400).