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Design, synthesis, anti-schistosomal activity and molecular docking of novel 8-hydroxyquinoline-5-sufonyl 1,4-diazepine derivatives

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ABSTRACT

Schistosomiasis remains one of the most prevalent parasitic infections and has significant public health consequences. Praziquantel (PZQ) is the only drug currently administrated to treat this disease. However, praziquantel-resistant parasites have been identified in endemic areas and can be generated in the laboratory. Therefore, it is essential to find new therapeutics. Herein we report a series of novel 8-hydroxyquinoline-5-sufonyl 1,4-diazepine derivatives, which were synthesized, characterized and tested as anti-schistosomal agents *in vitro*. Among all tested compounds, compounds **4a**, **5b**, and **7b** at different tested concentrations (50, 100, and 200 μg/mL) showed the highest schistosomicidal activity. Among those 3 compounds, compound **7b** was the most potent anti-schistosomal one. Moreover, all tested compound, at 50 μg/mL concentration, significantly reduced oviposition of adult worms *in vitro*. Furthermore, both compound **4a** and **7b**, as well as compound **6a**, completely diminished egg deposition. To clarify the possible mechanism by which novel 8-hydroxyquinoline-5-sufonyl 1,4-diazepine derivatives act as anti-schistosomal agents, molecular docking of all new compounds was carried out using Molsoft ICM pro 3.5-0a to investigate the binding affinity and binding mode to thioredoxin glutathione reductase enzyme (TGR), a potential drug target for anti-schistosomal agents. The docking results revealed moderate to high affinity of the new compounds towards TGR. Compound **7b** scored the highest binding energy (−101.13 kcal/mol) against TGR crystal structure forming eight hydrogen bonds with the amino acid residues at the binding site of the receptor. This result indicates that compound **7b** could exert its effect through inhibition of TGR, which is a vital enzyme for schistosome survival.

1. Introduction

Human schistosomiasis, or bilharzia, is considered one of the most significant and neglected tropical diseases in the world. This parasitic disease ranks second after malaria in terms of its public health importance [1]. Different approaches have been used to control schistosomiasis transmission including educational programs, mass chemotherapy, biological and chemical control for the snail intermediate host. However, despite these control programs, the disease still affects more than 200 million people worldwide and approximately 800 million people remain under infection risk [2]. Moreover, schistosomiasis continues to spread to new geographic areas due to environmental changes that result from the development of water resources, growth and migration of populations [3]. Praziquantel (PZQ, Fig. 1) is the only drug of choice for treatment of schistosomiasis. PZQ is administered to millions of people yearly [4,5]. They are rapidly re-infected and must be re-treated on an annual or semiannual basis. If praziquantel-resistant parasites develop, treatment for schistosomiasis will be in a crisis state [6,7]. Furthermore, the use of such drug for treatment is complicated due to the difficulties and expenses involved in a long-lasting maintenance of these programs [8]. These limitations, in combination with a considerable concern about the development of PZQ resistance, have motivated the scientific community to call for research and development of novel and inexpensive drugs against schistosomiasis [9–11]. A reliable alternative to PZQ does not exist at the moment. Oxamniquine (Fig. 1), the only other drug commercially available, is expensive and is active against only one of the main three schistosome species capable of infecting humans, *Schistosoma* (*S.*) *mansoni*. Artemisinins, although safe, are active only against the immature stages of the parasites. A treatment strategy that relies on just a single drug is at risk given the high

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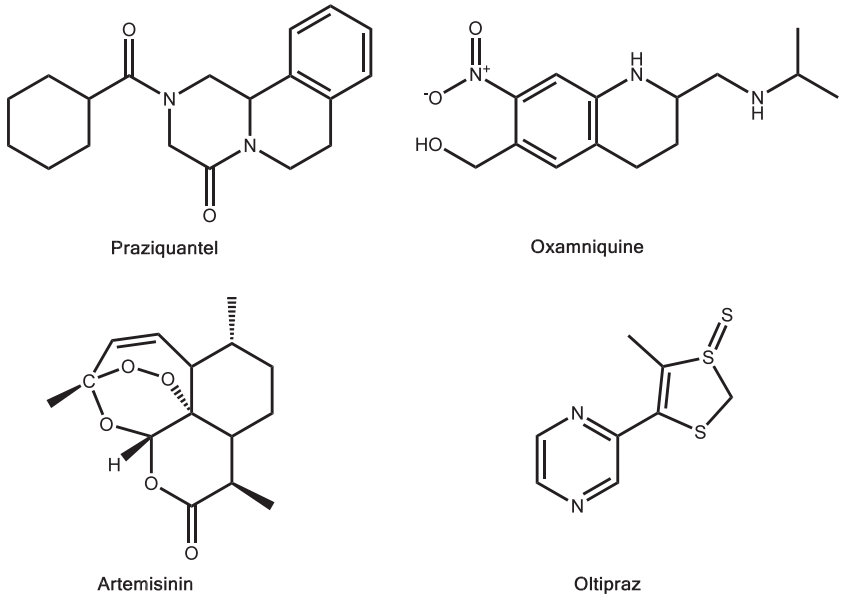


Fig. 1. Anti-schistosomal drugs.

probability that drug-resistant parasites will emerge [12,13]. Based on these findings, the identification of new and effective schistosomicidal compounds is in great demand.

8-Hydroxyquinoline derivatives are well known as powerful iron chelatores with antioxidant property. 8-Hydroxyquinoline derivatives are important constituents in a variety of pharmaceutically important compound classes. They have become of interest as a new class of potent HIV-1 integrase inhibitors [14] in modeling of the inhibition of retroviral integrases [15], protein tyrosine kinase inhibitors, anti-protozoal and retroviral co-infections [16], anti-HIV-1 agents [17], anti-leishmanial [18] and anti-filarial agents [19].

It has been reported that 8-hydroxyquinoline derivatives have anti-schistosomal activity [20,21]. In our earlier study [22], two novel 8-hydroxyqinoline-5-sulfonamido derivatives (**3**-amino-1-((8-hydroxyquinolin-5-yl)sulfonyl)-1H-pyrazol-5(4H)-one and1-((8-hydroxyquinolin-5-yl)sulfonyl)-3-methyl-1H-pyrazol-5(4H)-one) at 200 μg/mL concentration showed powerful anti-schistosomal activity against *S. mansoni* adult worms *in vitro*. However, at lower concentrations (50 and 100 μg/mL) both compounds showed a weak schistosomicidal activity in comparison with PZQ. The anti-schistosomal activity of such compounds was attributed to inhibition of thioredoxin glutathione reductase (TGR) enzyme. In *Schistosoma*, thiol redox homeostasis is completely dependent on the TGR enzyme, which directs the NADPH reducing equivalents to both GSH and thioredoxin [23]. This reliance on a single enzyme for both glutathione disulfide and thioredoxin reduction suggests that the parasite's redox systems are subject to a bottleneck dependence on TGR. Given the importance of cellular redox systems and the biochemical differences between the redox metabolism of *S. mansoni* and its human host, it was hypothesized that TGR could be an essential parasite protein and a potentially important drug target. The significance of *S. mansoni* TGR as a putative drug target was first demonstrated using an RNA interference approach, which killed 90% of treated parasites *in vitro* [24]. Moreover, treatment of *S. mansoni* with a TGR inhibitor, auranofin, resulted in 100% mortality [13]. It was demonstrated that TGR activity is inhibited by two schistosomicidal drugs used in the past to fight the infection, antimonyl potassium tartrate and oltipraz, suggesting that the enzyme is the main target of these compounds [24]. The dependence of *S. mansoni* on a single protein, TGR, for its protection from oxidative stress, makes it a promising drug target [25]. Therefore, TGR is an essential protein for the survival of *S. mansoni* and that it meets all the major criteria of an important target for anti-schistosomal chemotherapy development. Therefore the current study was conducted to synthesis a more potent anti-schistosomal 8-hydroxyquinoline-5-sufonyl 1,4-diazepine derivatives which may have a high binding energy with TGR.

2. Results and discussion

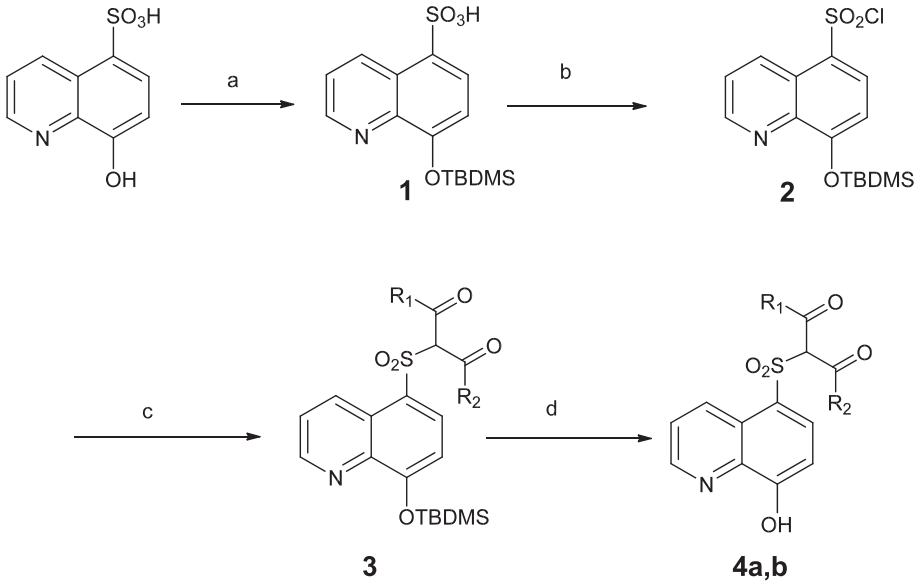
2.1. Chemistry

The study started with protection of 8-hydroxyquinoline-5-sulfonic acid with *tert*-butyl dimethyl silyl chloride (TBDMS) in presence of imidazole to afford the protected compound **1** in high yield. Compound **1** was then reacted with thionyl chloride in presence of anhydrous DMF to afford the corresponding sulfonyl chloride **2**. Compound 2 was then reacted with β-diketones namely acetyl acetone and 1,3-diphenylpropane-1,3-dione in presence of sodium methoxide which yielded corresponding protected substituted β-diketones **3a and b** (scheme 1).

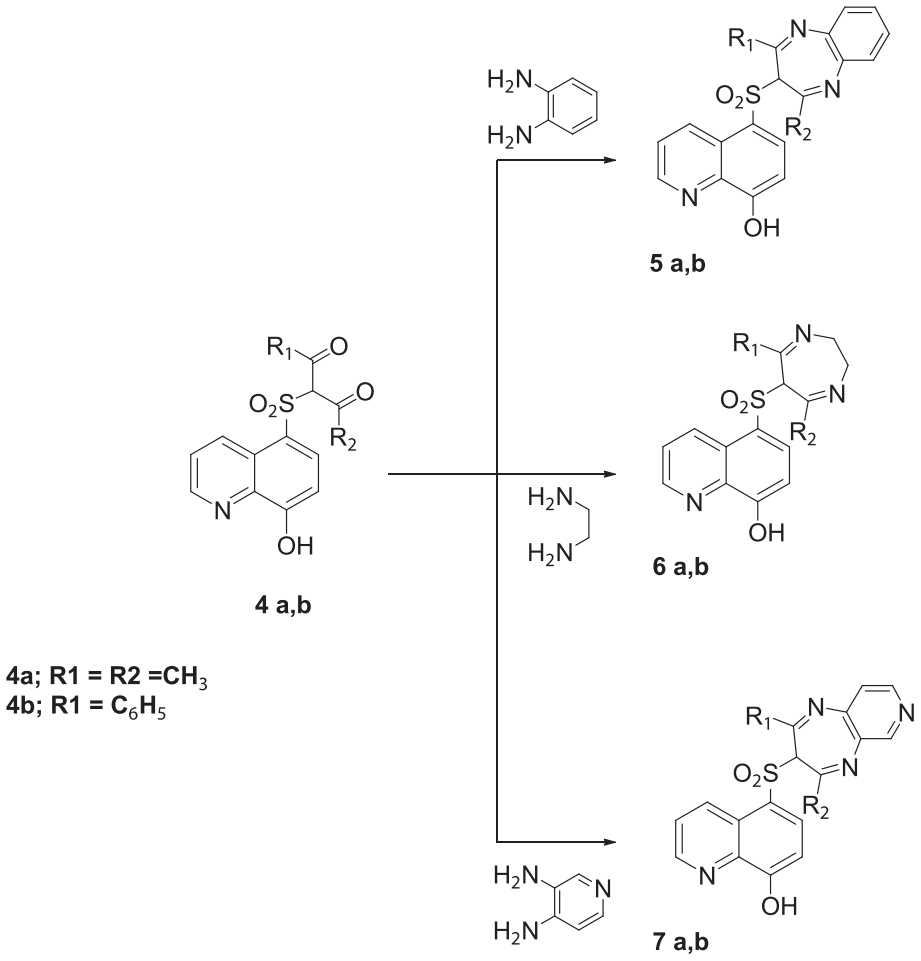
Compounds **3a and b** was then deprotected using 6N HCl to afford the free 8-hydroxyquinoline 5-sulfonyl-β-diketones **4a and b** in high yield. The structure of compounds **4a and b** was confirmed via ^{1}H NMR showing signals at δ 2.13 (s, 6H) dimethyl groups and δ 5.62 (s, 1H) SO_2–CH for **4a** and δ 6.43 (s, 1H) SO_2–CH for **4b**. Compounds **4a and b** were then condensed with diamines namely o-phenylendiamine, ethylenediamine and 3,4-diaminopyridine in ethyl acetate in presence of silica sulfuric acid (SSA) [26] (Scheme 2) to afford the corresponding 8-hydroxyquinoline-5-sufonyl 1,4-diazepine derivatives **5a and b**, **6a and b and 7a** and **b** respectively.

2.2. Molecular docking study

In *S. mansoni*, thiol redox homeostasis is completely dependent on the enzyme thioredoxin-glutathione reductase (TGR), which directs the NADPH reducing equivalents to both GSH and thioredoxin [13,24,25]. Characterization of the parasite TGR revealed enzymatic properties that differed from those of mammalian TGR,



Scheme 1. Reagents and conditions: (a) TBDMS, imidazole, room temp., (b) thionyl chloride, DMF, reflux; (c) NaOMe, β-diketones, stirring, 50 °C; and (d) 6N HCl, reflux.



Scheme 2.

thioredoxin (TrxR) and glutathione reductase (GR) enzymes. TGR was also identified as a multifunctional oxidoreductase with a remarkably wide substrate specificity, capable of directly reducing peroxides, selenium-containing compounds, and several important low-molecular-weight antioxidants, as well as Trx, 5,5′-dithiobis 2-nitrobenzoic acid (DTNB), reduces glutathione disulfide (GSSG),

Table 1
The docking energy scores of compounds **4a–7b** with the amino acid residues forming hydrogen bonds in comparison with reference ligand praziquantel.

Cpd. no.	Docking score (kcal/mol)	No. of hydrogen bonds	Amino acid residues forming hydrogen bonds in A^0
Ligand (praziquantel)	−84.13	3	S117 hg – m M o2: 2.54 A R393 he – m M o1: 2.02 A R393 hh22 – m M o1: 1.83 A
4a	−93.36	4	T153 hg1 – m M o3: 2.74 A K227 hz1 – m M o1: 2.58 A K227 hz2 – m M o1: 2.14 A R260 hh11 – m M o2: 2.33 A
5a	−91.74	2	T153 hg1 – m M n2: 1.97 A R260 hh11 – m M o2: 2.49 A
5b	−88.85	5	G116 hn – m M o1: 2.42 A S117 hn – m M o1: 2.42 A Y138 hn – m M n2: 2.17 A T153 hn – m M n1: 2.38 A G115 o – m M h6: 2.66 A
6a	−86.71	3	T153 hn – m M n3: 2.78 A T153 hg1 – m M n3: 2.41 A E140 oe1 – m M h17: 2.33 A
6b	−99.01	5	E140 hn – m M n2: 2.75 A T153 hn – m M o1: 1.64 A T153 hg1 – m M o1: 1.15 A R260 hh11 – m M n2: 2.07 A T153 og1 – m M h6: 1.31 A
7a	−100.21	5	Y138 hn – m M o2: 2.18 A E140 hn – m M o3: 1.57 A G228 hn – m M n1: 2.04 A R260 hh11 – m M n3: 2.26 A G228 o – m M h6: 2.02 A
7b	−101.13	8	Y138 hn – m M o2: 2.45 A Y138 hn – m M n4: 2.15 A E140 hn – m M o3: 1.43 A K227 hz1 – m M n1: 2.38 A K227 hz1 – m M o1: 2.10 A K227 hz2 – m M n1: 2.46 A K227 hz2 – m M o1: 2.54 A G228 o – m M h6: 2.37 A

and glutathione–β-hydroxyethyl disulfide (GSH–HED) [24]. This dependence of *S. mansoni* on TGR for its protection from oxidative stress, makes it a promising drug target [25]. Based on these finding we decided to dock our new 8-hydroxyquinoline derivatives against TGR and compare it with the reference drug praziquantel. Molecular docking studies were carried out using Mol soft ICM 3.5-0a. The aim of the flexible docking calculations is the prediction of correct binding geometry for each binder. The scoring functions and hydrogen bonds formed with the surrounding amino acids of the receptor TGR are used to predict tested compounds binding modes. We evaluated the new compounds **4a**, **5b**, and **7b** (Table 1) through molecular modeling and docking techniques against TGR crystal structure which was downloaded from PDB website PDB id (2V6O).

Fig. 2 shows binding mode of the reference drug (Praziquantel), **4a**, **5b**, and **7b** into its binding site of TGR. Praziquantel "the reference drug" docking results to TGR reveals docking score of $\Delta G = -84.13$ kcal/mol and three hydrogen bonds to the active site with Ser-117 and Arg-393. on the other hand compound **4a** docking result against TGR shows docking score of −93.36 kcal/mol forming four hydrogen bonds with TGR binding site at Thr-153, Lys-227 and Arg-260. On the other hand compound **5b** docking results revealed a binding energy of −88.85 kcal/mol, forming five hydrogen bonds with binding site of TGR at Gly-115, Gly-116, Ser-117, Tyr-138 and Thr-153.

While compound **6a** docking result against TGR shows docking score of −86.71 kcal/mol forming three hydrogen bonds with TGR binding site at Glu-140 and Thr-153. Compound **7b** showed the highest binding energy among all tested compounds with docking score of −101.13 kcal/mol forming eight hydrogen bonds with TGR binding site at Tyr-138, Glu-140, Lys-227 and Gly-228.

2.3. Anti-schistosomal activity

2.3.1. Effect of 8-hydroxyquinoline derivatives on adult schistosomes survival

The survival of 56-day-old adult worms of *S. mansoni* was assessed *in vitro* by incubation with different concentrations of new 8-hydroxyquinoline derivatives. The effect of 8-hydroxyquinoline derivatives on the mortality rate of both male and female adult worms was analyzed with respect to the concentration and incubation time. Compounds **4b**, **5a**, **6a**, **6b**, and **7a** at 50, 100, and 200 μg/mL concentrations did not showed any schistosomicidal effect until 96 h post-incubation. However, only compounds **4b**, **5a**, and **6a** at 200 μg/mL concentration caused 50% death of both male and female worms after 120 h post-incubation. On the other hand, as depicted in Table 2, **4a** caused 100% death of all worms at a concentration of 100 and 200 μg/mL after 96 and 72 h, respectively. However, the 50 μg/mL concentration of **4a** caused only 50% and 40% death of male and female worms, respectively, after 120 h of incubation. Interestingly, both **5b** and **7b** at 50 and 100 μg/mL concentrations caused 100% death of all worms after 72 h and 48 h, respectively. Moreover, the 200 μg/mL concentration of **7b** resulted in 75% and 100% death of male and female worms, respectively, after 24 h of incubation.

Subsequently, the effect of new 8-hydroxyquinoline derivatives on the motor activity of the worms was examined, and a dose-dependent reduction of activity was observed in compounds **4a**,

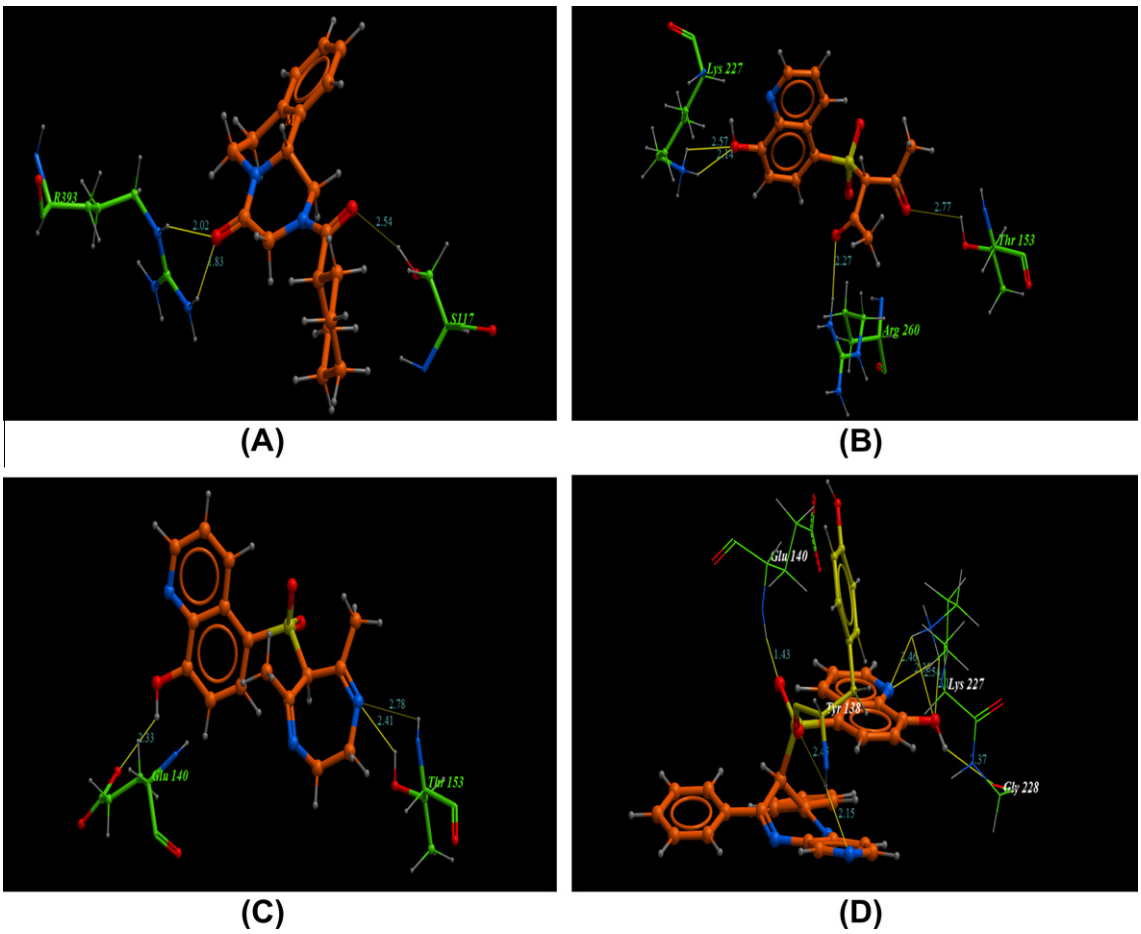


Fig. 2. Molecular docking and binding mode of tested compounds PZQ (reference drug), **4a**, **5b**, and **7b** to the active binding site of TGR receptor.

5b, and **7b** (Table 2). However, **4b**, **5a**, **6a**, **6b**, and **7a** did not showed any significant decrease in motor activity of adult parasites until 72 h post-incubation.

In addition to the mortality rate and changes in the motor capacity of *S. mansoni* adults, the results highlighted the effect of 8-hydroxyquinoline derivatives on the parasite's tegument. As shown in Table 2, only partial morphological alterations of the tegument occurred in a dose-dependent manner after incubation with compounds **4b** and **7b**. No extensive tegumental alterations were observed in all worms incubated with any compound. In contrast, 10 μg/mL PZQ group had tegumental alteration in all the worms and severe tegumental alteration were more pronounced in male than in female adults. Meanwhile, no tegumental changes in adult worms were observed in the negative control group.

From previously mentioned data, compounds **4a**, **5b**, and **7b** have a potent schistosomicidal activity, and compound **7b** is the most efficient one. The exact mechanism by which such compounds exert their *in vitro* schistosomicidal effect is not clear. However, there are two possible mechanisms; first: compounds **7b** has a high binding energy ($\Delta G = -101.33$ kcal/mol), and eight hydrogen bonds at the active binding site of TGR, as mentioned previously. Such binding could inhibit the activity of TGR, schistosome antioxidant enzyme, which in turn lead to inhibition of the antioxidant response of the parasite that can lead to parasite death. This assumption supported by the previous studies that showed silencing of TGR expression lead to death of *S. mansoni in vitro* within 4 days [24]. Moreover, treatment of *S. mansoni* with a TGR inhibitor resulted in 100% mortality of the parasite after 9 h of exposure *in vitro* [13]. This finding explains why compound **7b**, which has the highest binding energy with TGR, is more effective in killing of *S. mansoni* adult worms than other compounds. Second: it is well documented that 8-hydroxyquinoline is a potent chelator to various metals, especially iron [27,28]. Iron (Fe) is an important trace element found in nearly all organisms, and is used as a cofactor in many biological reactions. Schistosomes require Fe for development and maintain their life [29]. Lipid solubility of the neutral iron-8-hydroxyquinoline complex ensures penetration of epithelial cells and distribution within the cells [30]. It was shown that iron bound to 8-hydroxyquinoline causes oxidative damages (via free radical lipid peroxidation) in living cells [30,31] as well as DNA strand breakage in cultured cells [31]. The Fungicidal and bactericidal action of 8-hydroxyquinoline derivatives has been attributed in part to its ability to chelate essential trace metals [32]. Taken together, our new 8-hydroxyquinoline derivatives could exert its schistosomicidal effect through inhibition of TGR activity and Fe chelation.

2.3.2. Effect of 8-hydroxyquinoline derivatives on the reproductive fitness of S. mansoni

In order to evaluate the egg production by adult worms of *S. mansoni*, 8-hydroxyquinoline derivatives were tested at 50 μg/mL concentration. As shown in Fig. 3, worm pairs incubated with **5b**, **6a**, and **7b** did not produce any eggs until 72 h post-incubation. However, worm pairs incubated with **4a**, **4b**, **5a**, and **7a** showed about 50% significant reduction in oviposition after 24 h, 48, and 72 h of incubation in comparison with the negative control group. Unexpected, worm pairs incubated with **6b** showed significant

Table 2
In vitro effects of 8-hydroxyquinoline derivatives against 56-day-old adult *S. mansoni*.

Groups	Drug concentration	Incubation period (h)	Dead worms (%)		Motor activity reduction (%)				Worms with tegumental alteration (%)			
					Slight		Significant		Partial		Extensive	
			M	F	M	F	M	F	M	F	M	F
Control	RPMI 1640 + 1% DMSO	24	0	0	0	0	0	0	0	0	0	0
		48	0	0	0	0	0	0	0	0	0	0
		72	0	0	0	0	0	0	0	0	0	0
		96	0	0	0	0	0	0	0	0	0	0
		120	0	0	0	0	0	0	0	0	0	0
PZQ	10 μg/mL	24	100	100	0	0	100	100	20	35	80	65
4a	50 μg/mL	24	0	0	10	0	0	0	0	0	0	0
		48	0	0	20	10	0	0	0	0	0	0
		72	0	0	20	30	30	10	0	0	0	0
		96	25	0	25	30	50	40	0	0	0	0
		120	50	40	10	0	100	90	0	0	0	0
	100 μg/mL	24	0	0	20	10	30	20	0	0	0	0
		48	25	0	50	20	50	40	0	0	0	0
		72	50	40	0	20	50	80	0	0	0	0
		96	100	100	0	0	100	100	0	0	0	0
	200 μg/mL	24	0	0	50		0	50	100	0	0	0
		48	60	80	0	0	100	100	0	0	0	0
		72	100	100	0	0	100	100	0	0	0	0
5b	50 μg/mL	24	0	0	55	45	20	30	0	0	0	0
		48	40	45	0	0	100	100	0	0	0	0
		72	100	100	0	0	100	100	0	0	0	0
	100 μg/mL	24	0	0	30	10	70	90	10	10	0	0
		48	100	100	0	0	100	100	15	25	0	0
	200 μg/mL	24	0	0	0	0	100	100	10	15	0	0
		48	100	100	0	0	100	100	20	20	0	0
7b	50 μg/mL	24	10	10	40	55	20	45	0	0	0	0
		48	35	70	20	0	80	100	10	15	0	0
		72	100	100	0	0	100	100	20	20	0	0
	100 μg/mL	24	20	50	30	20	70	80	10	10	0	0
		48	100	100	0	0	100	100	20	20	0	0
	200 μg/mL	24	75	100	10	0	90	100	10	20	0	0
		48	100	100	0	0	100	100	20	20	0	0

Worm motor activity and survival of the parasites was monitored under a stereomicroscope. Tegumental alterations were monitored using an inverted microscope. The effects of the compounds on motor activity and tegumental alterations of 20 adult *S. mansoni* worms were assessed qualitatively from five separate experiments.

increase in oviposition after 48 h and 72 h of incubation in comparison with the negative control group.

Considering the strong lethal effect of 8-hydroxyquinoline derivatives on adult schistosomes, the *in vitro* oviposition was continually monitored to assess the sexual fitness of treated worms. From the aforementioned data, 50 μg/mL concentration of all compounds, except compound **6b**, showed a potent reduction in ova deposition. This effect could be attributed to chelation of Fe by 8-hydroxyquinoline derivatives. Schistosomes store abundant Fe in vitelline (eggshell-forming) cells of the female system. Schistosomes synthesize eggshells from among a range of precursor's proteins belonging to tyrosine-rich proteins, the P14, P19, and P48 proteins [33]. The eggshell is formed by enzymatic oxidation of the tyrosine residues to dihydroxyphenylalanine-quinones, which then bind lysine residues of adjacent proteins [34,35] to form a rigid structure. Fe plays an important role in cross-linking and stabilization of dihydroxyphenylalanine-rich proteins [29,36,37]. 8-hydroxyquinoline and its derivatives have been reported as potent Fe chelator [38]. Consequently, depravation of Fe leads to disruption of egg formation. However, mechanism by which compound **6b** increases oviposition is unclear and needs further study.

In conclusion, our results indicate that 8-hydroxyquinoline derivatives possess *in vitro* schistosomicidal activity against *S. mansoni* adult worms and may be novel broad-spectrum anti-schistosomal drug candidates. For future work, an *in vivo* study is required to evaluate schistosomicidal effect of compound **4a**, **5b,** and **7b** on *S. mansoni* adult worms and their induced pathology.

3. Experimental

3.1. Chemistry

All starting materials and reagents were purchased from Sigma–Aldrich, BDH and Fluka and used without further purification. All solvents were either of analytical grades or dried and distilled immediately prior to use. All of the reactions were performed using oven-dried glassware. Melting points were measured on a Stuart-SMP10 melting point apparatus and are uncorrected. TLC was performed using Merck precoated Silica gel 60 F254 aluminum sheets (20 × 20 cm, layer thickness 0.2 mm) and Merck precoated Silica gel aluminum sheets (20 × 20 cm, layer thickness 0.2 mm) and spots were visualized by UV (254 nm), $KMNO_4$ solution and/or charring with H_2SO_4–EtOH (5% v/v). All reaction products were stored refrigerated under 4 °C. The 1H NMR spectra were recorded (DMSO-d_6) with Varian 400-MR system, Chemical shifts are expressed in parts per million (ppm) and reported either relative to an internal tetramethyl-silane standard (TMS δ = 0.0) or relative to solvent peaks. High resolution mass spectra (HRMS) were

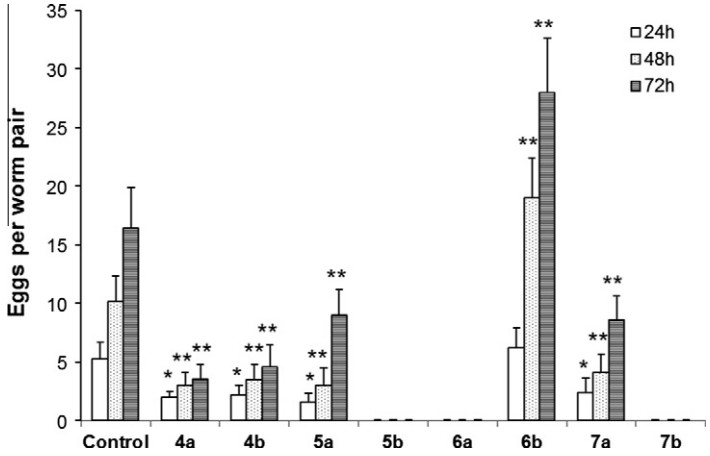


Fig. 3. *In vitro* effect of 8-hydroxyquinoline derivatives on *Schistosoma mansoni* oviposition. Adult worm couples were incubated with 50 μg/mL of either compound **4a**, **4b**, **5a**, **5b**, **6a**, **6b**, **7a**, **or 7b** and at the indicated time periods, the cumulative number of eggs per worm couple was assessed and scored using an inverted microscope. Values are means ± SD (bars) of 10 worm couples. $^{*}P < 0.05$ and $^{**}P < 0.001$ compared with control group (RPMI 1640 + 1% DMSO).

recorded on Bruker Maxis spectrometer. Elemental analyses were recorded with EuroVector EA3000 Elemental Analyzer.

3.1.1. 8-((tert-Butyldimethylsilyl)oxy)quinoline-5-sulfonic acid 1

A 100-mL flask was successively loaded with dry CH_2Cl_2 (50 mL), 8-hydroxy-5-sulfonic acid (7.2 g, 0.032 mol), imidazole (2.29 g, 0.034 mol, 1.05 equiv), and *tert*-butyldimethylsilyl chloride (5.31 g, 0.035 mol, 1.1 equiv). The solution was stirred at room temperature for 10 h, diluted with Et_2O (500 mL), washed with aqueous HCl (0.1 M, 50 mL), brine (100 mL), water (100 mL), dried over K_2CO_3 and evaporated under reduced pressure to afford 10.32 g (95%) of protected product as a colorless oil. ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.29 (s, 6H); δ 1.09 (s, 9H); 7.23 (d, 1H, J = 8.3); 7.66–7.73 (m, 2H); 7.94 (dd, 1H, J = 1.3, 5.3); 8.71 (dd, 1H, J = 1.3, 8.4); HRMS $(M)^+$ calcd for $C_{15}H_{21}NO_4SSi$ 339.0960. Found: 339.1034. Elemental analysis, calcd: C, 53.07; H, 6.24; N, 4.13; O, 18.85; S, 9.45. Found: C, 53.09; H, 6.25; N, 4.15; O, 18.87; S, 9.43.

3.1.2. 8-((tert-Butyldimethylsilyl)oxy)quinoline-5-sulfonyl chloride 2

In a 100 mL round-bottom flask fitted with magnetic stirrer, condenser and N_2 inlet were placed 8-((*tert*-butyldimethylsilyl)oxy)quinoline-5-sulfonic acid **1** (10.32 g, 0.03 mol), 60 mL of $SOCl_2$, and 1 mL of anhydrous DMF. The suspension was heated to reflux for 3 h, and complete dissolution was achieved after 30 min. After 1 h, product began to precipitate from the reaction mixture. After 3 h, the reaction mixture was cooled and transferred to a 250 mL flask before stripping off the $SOCl_2$. The remaining $SOCl_2$ was removed by successive azeotropic distillations with CH_2Cl_2 and toluene on a rotary evaporator. The crude sulfonyl chloride was used for next reaction without further purification.

3.2. General procedure for the preparation of 2-((8-((tert-butyldiphenylsilyl)oxy)quinolin-5-yl)sulfonyl)-1,3-dimehyl/1,3-diphenyl propane-1,3-dione 3a,b

Sodium methoxide (0.81 g, 0.015 mol) and β-diketones (0.015 mol) were placed in a dried round bottom flask and stirred for 1 h on a magnetic stirrer at 50 °C, after which a creamy mass was obtained. The 8-((*tert*-butyldimethylsilyl)oxy)quinoline-5-sulfonyl chloride 2 (5.37 g, 0.015 mmol) was taken in dry toluene (15 mL) and added drop by drop in above reaction mass. The reaction mixture was refluxed for 7 h at 100 °C with stirring. The progress of the reaction was monitored by TLC. After completion of the reaction, the mixture was cooled and toluene was removed under reduced pressure. The reaction mixture was extracted using chloroform and washed with water. The chloroform layer was dried using anhydrous sodium sulfate and distilled to yield the solid compound. The product was purified by column chromatography over silica gel using pet ether:ethyl acetate (1:2) as an eluent.

3.2.1. 2-((8-((tert-Butyldimethylsilyl)oxy)quinolin-5-yl)sulfonyl)-1,3-dimethylpropane-1,3-dione 3a

Colorless oil, yield 72%; ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.29 (s, 6H); δ 1.09 (s, 9H); δ 2.13 (s, 6H); 5.62 (s, 1H); 7.31–8.89 (m, 5H aromatic protons); HRMS $(M)^+$ calcd for $C_{20}H_{27}NO_5SSi$ 421.1379. Found: 421.1425. Elemental analysis, calcd: C, 56.98; H, 6.46; N, 3.32; O, 18.98; S, 7.61. Found: C, 56.95; H, 6.48; N, 3.31; O, 18.95; S, 7.65.

3.2.2. 2-((8-((tert-Butyldimethylsilyl)oxy)quinolin-5-yl)sulfonyl)-1,3-diphenylpropane-1,3-dione 3b

Colorless oil, yield 62%;^{1}H NMR (400 MHz, DMSO-d_6) δ 0.29 (s, 6H); δ 1.09 (s, 9H); δ 6.43 (s, 1H); δ 7.31–8.89 (m, 15H, aromatic protons); HRMS $(M)^+$ calcd for $C_{30}H_{31}NO_5SSi$ 545.1692. Found: 545.1632. Elemental analysis, calcd: C, 66.03; H, 5.73; N, 2.57; O, 14.66; S, 5.88. Found: C, 65.98; H, 5.76; N, 2.58; O, 14.62; S, 5.89.

3.3. General procedure for the preparation of 2-((8-hydroxyquinolin-5-yl)sulfonyl)-1,3-dimethyl/1,3-diphenylpropane-1,3-dione 4a,b

The *tert*-butyl ester **3a and b** (0.01 mol) was refluxed in 6N HCl (20 mL) under nitrogen until TLC analysis of the solution showed complete deprotection (ca. 5 h). The hydrolyzed mixture was cooled, diluted with 1 M HCl (25 mL), extracted with EtOAc (2 × 50 mL), and concentrated to dryness under reduced pressure to afford the deprotected products **4a** and **b**.

3.3.1. 3-((8-Hydroxyquinolin-5-yl)sulfonyl)pentane-2,4-dione 4a

M.p. 198 °C, yield 59%; ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.12 (s, 6H); δ 5.62 (s, 1H); δ 7.31–8.89 (m, 5H aromatic); δ 8.34 (s, 1H, broad, OH); HRMS $(M)^+$ calcd for $C_{14}H_{13}NO_5S$ 307.0514. Found: 307.0872. Elemental analysis, calcd: C, 54.71; H, 4.26; N, 4.56; O, 26.03; S, 10.43. Found: C, 54.74; H, 4.23; N, 4.57; O, 26.01; S, 10.45.

3.3.2. 2-((8-Hydroxyquinolin-5-yl)sulfonyl)-1,3-diphenylpropane-1,3-dione 4b

M.p. 203 °C, yield 65%; ^{1}H NMR (400 MHz, DMSO-d_6) δ 6.43 (s, 1H); δ 7.31–8.89 (m, 15H, aromatic protons); δ 8.34(s, 1H, broad, OH); HRMS $(M)^+$ calcd for $C_{24}H_{17}NO_5S$ 431.0827. Found: 431.1037. Elemental analysis, calcd: C, 66.81; H, 3.97; N, 3.25; O, 18.54; S, 7.43. Found: C, 66.83; H, 3.95; N, 3.28; O, 18.51; S, 7.43.

3.4. General procedure for the preparation of 1H-1,4-diazepine 5a-b, 3H-1,5-benzodiazepine 6a-b and 3H-pyrido[3,4-b][1,4]diazepine derivatives

An equimolar ratio of β-diketones (10 mmol) 3a–d, and EDA/o-PDA/3,4-DAP (10.0 mmol) in ethyl acetate (50 mL) in the presence of silica sulfuric acid (10.0 mmol) was stirred 50 °C for 2 h. The progress of reaction was monitored by TLC using 7:2:1 (benzene:ethanol:ammonia) upper layer as mobile phase. Upon completion of reaction, the mixture was extracted with ethyl acetate (2 × 25 mL) and the solvent was removed. The crude product was washed with dry ether and recrystallized from petroleum ether:ethyl acetate (1:1). The product was purified by column chromatography over silica gel using pet ether:ethyl acetate (40:60) as an eluent.

3.4.1. 5-((2,4-Dimethyl-3H-benzo[b][1,4]diazepin-3-yl)sulfonyl)quinolin-8-ol 5a

M.p. 143 °C, yield 78%; ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.13 (s, 6H); δ 5.62 (s, 1H); δ 7.31–8.89 (m, 9H, aromatic protons); δ 8.34 (s, 1H, broad, OH); ^{13}C NMR (DMSO) δ 24.8, 61.8, 110.8, 117, 124.6, 126.4, 127.1, 130.3, 133.2, 138.3, 142.3, 143.3, 161.11, 162.1; HRMS $(M)^+$ calcd For $C_{20}H_{17}N_3O_3S$ 379.0991. Found: 379.1045. Elemental analysis, calcd: C, 63.31; H, 4.52; N, 11.07; O, 12.65; S, 8.45. Found: C, 63.28; H, 4.54; N, 11.08; O, 12.63; S, 8.47.

3.4.2. 5-((2,4-Diphenyl-3H-benzo[b][1,4]diazepin-3-yl)sulfonyl)quinolin-8-ol 5b

M.p. 187 °C, yield 64%; ^{1}H NMR (400 MHz, DMSO-d_6) δ 6.43 (s, 1H); δ 7.31–8.89 (m, 19H, aromatic protons); δ 8.34 (s, 1H, broad, OH); ^{13}C NMR (DMSO) δ 68.2, 110.0, 120.1, 124.6, 126.1, 127.8, 129.1, 129.5, 130.2, 130.9, 132.9, 133.2, 133.9, 135.5, 142.7, 143.3, 161.1, 162.6; HRMS $(M)^+$ calcd for $C_{30}H_{21}N_3O_3S$ 503.1304. Found: 503.1296. Elemental analysis, calcd: C, 71.55; H, 4.20; N, 8.34; O, 9.53; S, 6.37. Found: C, 71.58; H, 4.18; N, 8.35; O, 9.50; S, 6.39.

3.4.3. 5-((5,7-Dimethyl-3,6-dihydro-2H-1,4-diazepin-6-yl)sulfonyl)quinolin-8-ol 6a

M.p. 132 °C, yield 81%; ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.13 (s, 6H); 3.61 (m, 4H, N–CH2–CH2–N); δ 5.23 (s, 1H); δ 7.31–8.89 (m, 5H, quinolone aromatic Protons); ^{13}C NMR (DMSO) δ 23.5, 47.7, 61.12, 110.8, 120.6, 126.5, 127.8, 130.3, 138.3, 141.6, 143.3, 160.5, 161.1; HRMS $(M)^+$ calcd for $C_{16}H_{17}N_3O_3S$ 331.0991. Found: 331.1094. Elemental analysis, calcd: C, 57.99; H, 5.17; N, 12.68; O, 14.48; S, 9.68. Found: C, 58.02; H, 5.15; N, 12.69; O, 14.43; S, 9.71.

3.4.4. 5-((5,7-Diphenyl-3,6-dihydro-2H-1,4-diazepin-6-yl)sulfonyl)quinolin-8-ol 6b

M.p. 171 °C, yield 74%; ^{1}H NMR (400 MHz, DMSO-d6) δ 3.68 (m, 4H, N–CH_2–CH_2–N); δ 6.25 (s, 1H); δ 7.31–8.89 (m, 15H, aromatic protons); ^{13}C NMR (DMSO) δ 50.7, 67.4, 110.0, 121.9, 126.4, 128.5, 128.7, 129.1, 130.1, 131.3, 133.3, 135.4, 142.0, 143.3, 156.9, 161.1; HRMS $(M)^+$ calcd for $C_{26}H_{21}N_3O_3S$ 455.1304. Found: 455.1267. Elemental analysis, calcd: elemental analysis, calcd: C, 68.55; H, 4.65; N, 9.22; O, 10.54; S, 7.04. Found: C, 68.52; H, 4.68; N, 9.19; O, 10.57; S, 7.04.

3.4.5. 5-((2,4-Dimethyl-3H-pyrido[3,4-b][1,4]diazepin-3-yl)sulfonyl)quinolin-8-ol 7a

M.p. 153 °C, yield 61%; ^{1}H NMR (400 MHz, DMSO-d6) δ 2.22 (s, 3H); δ 2.25 (s, 3H); δ 4.98 (s, 1H); δ 7.31–8.89 (m, 8H, aromatic protons); ^{13}C NMR (DMSO) δ 24.8, 61.8, 110.8, 117.0, 118.6, 126.4, 127.1, 130.0, 135.9, 138.3, 142.2, 143.3, 143.8, 144.9, 155.0, 161.1, 161.4, 162.1; HRMS $(M)^+$ calcd for $C_{19}H_{16}N_4O_3S$ 380.0943. Found: 380.1034; Elemental analysis, calcd: C, 59.99; H, 4.24; N, 14.73; O, 12.62; S, 8.43. Found: C, 60.01; H, 4.19; N, 14.74; O, 12.65; S, 8.41.

3.4.6. 5-((2,4-Diphenyl-3H-pyrido[3,4-b][1,4]diazepin-3-yl)sulfonyl)quinolin-8-ol 7b

M.p. 217 °C, yield 58%; ^{1}H NMR (400 MHz, DMSO-d_6) δ 6.14 (s, 1H); δ 7.31–8.89 (m, 18H, aromatic protons); ^{13}C NMR (DMSO) δ 68.1, 110.0, 119.3, 120.1, 126.4, 127.8, 129.1, 129.5, 130.2, 133.9, 133.6, 134.2, 135.3, 136.2, 142.7, 143.3, 143.9, 145.6, 155.3, 161.1, 162.7; HRMS $(M)^+$ calcd for $C_{29}H_{20}N_4O_3S$ 504.1256. Found: 504.1202. Elemental analysis, calcd: C, 69.03; H, 4.00; N, 11.10; O, 9.51; S, 6.36. Found: C, 69.01; H, 4.01; N, 11.11; O, 9.53; S, 6.34.

4. Molecular docking studies

All docking studies were performed using 'Internal Coordinate Mechanics (Molsoft ICM 3.5-0a).

4.1. Preparation of small molecule

Novel 8-hydroxyquinoline-5-(1,4-diazepin-6-yl)sulfonyl derivatives were synthesized and tested as anti-schistosomal agents and compiled using ChemDraw. 3D structures were constructed using Chem 3D ultra 12.0 software [Molecular Modeling and Analysis; Cambridge Soft Corporation, USA (2010)], and then they were energetically minimized by using MOPAC (semi-empirical quantum mechanics), Jop Type with 100 iterations and minimum RMS gradient of 0.01, and saved as MDL MolFile (*.mol).

4.2. Generation of ligand and enzyme structures

The crystal structure of target enzyme thioredoxin-glutathione reductase TGR (2V6O) is was retrieved from the Protein Data Bank (http://www.rcsb.org/pdb/welcome.do). All bound waters ligands and cofactors were removed from the enzyme.

4.3. Docking using Molsoft ICM 3.4-8C program

1. Convert our PDB file into an ICM object: This conversion involves addition of hydrogen bonds, assignment of atoms types, and charges from the residue templates.
2. To perform ICM small molecule docking:
   (a) Setup docking project:
      (1) Set project name
      (2) Setup the receptor
      (3) Review and adjust binding site
      (4) Make receptor maps

   (b) Start docking simulation:
3. Display the result: ICM stochastic global optimization algorithm attempts to find the global minimum of the energy function that include five grid potentials describing interaction of the flexible ligand with the receptor and internal conformational energy of the ligand, during this process a stack of alternative low energy conformations is saved (Table 1).

All inhibitors were compared according to the best binding free energy (minimum) obtained among all the run.

5. Anti-schistosomal activity

5.1. In vitro studies with S. mansoni

The John Bruce Egyptian strain of *S. mansoni* was maintained by passage through *Biomphalaria alexandrina* snails and MF1 mice [39]. After 8 weeks, *S. mansoni* adult worms (male and female) were recovered under aseptic conditions from mice previously infected with 100 cercariae by perfusion of the liver and mesenteric veins [40]. The worms were washed in Roswell Park Memorial Institute (RPMI) 1640 medium (Invitrogen), kept at pH 7.5 with HEPES 20 mM, and supplemented with penicillin (100 UI/mL), streptomycin (100 μg/mL), and 10% heat-inactivated fetal calf serum. After washing, one pair of adult worms was transferred to each well of a 24-well Falcon plates containing 2 mL of the same medium and incubated at 37 °C in a humid atmosphere containing 5% CO_2 prior to use. At 24 h after incubation, 8-hydroxyquinoline derivatives were dissolved in 1% DMSO and used at concentrations of 50, 100, and 200 μg/mL. The control worms were assayed in RPMI 1640 medium with 1% DMSO as a negative control group and in 10 μg/mL PZQ as a positive control group. The experiment was carried out in quadruplicate and repeated at least five times. Worm motor activity, tegumental alterations, mortality rate, and egg output (oviposition) were monitored on daily basis for 5 days using an inverted microscope and a stereomicroscope (Nikon) [41,42].

6. Statistical analysis

The statistical tests were performed with the SPSS (version 16) software. Significant differences were determined by one-way analysis of variance (ANOVA) and applying Tukey's test for multiple comparisons with a level of significance set at $P < 0.05$.

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