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MEDICINAL CHEMISTRY RESEARCH

ORIGINAL RESEARCH

Evaluating anti-*Toxoplasma gondii* activity of new serie of phenylsemicarbazone and phenylthiosemicarbazones in vitro

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Abstract While *Toxoplasma gondii* is able to infect and replicate within all eukaryotic cells, tachyzoites are the infective form of *T. gondii* that invades all eukaryotic cells leading to tissue rupture, the main features of toxoplasmosis. The present study evaluates the activity of (benzaldehyde)-4-phenyl-3-thiosemicarbazone and (benzaldehyde)-(4 or 1)-phenylsemicarbazone against intracellular *T. gondii*. The nine new compounds were incubated in infected Vero cells at concentrations of 0.01, 0.1, 0.5, and 1.0 mM and evaluated for three main effects: cytotoxicity, infection, and number of intracellular parasites. The cytotoxicity test showed a pattern by analyzing the substituent arylhydrazone, where trihydroxy Compounds **4**–**9** were cytotoxic at concentrations of 0.5 and 1.0 mM. The results highlight Compound **8**, which reduced the number of intracellular parasites by 82 % in a concentration of 0.01 mM and showed a LD50 of 0.3 mM in cell culture. These different biological actions are due to changes in the molecular structure and type of radical present in each compound. All compounds tested were more efficient than the control drug sulfadizine.

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Keywords Anti-*Toxoplasma gondii* · Thiosemicarbazones · Semicarbazones · Schiff base · Anti-parasitic

Introduction

Toxoplasma gondii, the agent of Toxoplasmosis disease, belongs to Apicomplexan Phylum which also includes others pathogens of medical and veterinary importance, such as the *Plasmodium, Cryptosporidium, Sarcocystis, Eimeria, Babesia*, and *Neospora* species (Levine *et al*., 1980). Toxoplasmosis affects millions of people worldwide and is the main cause of death in patients with AIDS (Luft and Remington, 1992). While chemotherapeutic treatment involves the combination of sulfonamides with mainly pyremithamine (or clindamycin) and folinic acid (Bosch-Driessen *et al*., 2002), there is a inefficient elimination of the intracellular parasites, up to 40 % of treated patients develop side effects such as the suppression of hematopoiesis, teratogenic effect during pregnancy, and severe skin reaction (James *et al*., 1996; Hill and Dubey, 2002; Dannemann *et al*., 1991; Wong and Remington, 1993). As consequence, the treatment must be changed or suspended. (Bosch-Driessen *et al*., 2002). *Toxoplasma* infects all nucleated cells of vertebrates, representing an attractive model for the study of cellular biology of the host cell-parasite interaction (Kim and Weiss, 2004). Therefore, it can offer a useful model for the treatment of other diseases such as Chagas disease and Leishmaniasis (Melo and Beiral, 2003).

Thiosemicarbazones and semicarbazones were shown to be useful as anti-parasitic, anti-bacterial, anti-microbial agents (Melo and Souza, 2000; Tenório *et al*., 2005; Carvalho and Melo, 2006). Considering this results, our

group developed new compounds belonging to the semicarbazone class to examine their anti-Toxoplasmic actions and to facilitate the development of more efficient drugs that do not cause severe side effects. The new analogues were synthesized from thiosemicarbazone and semicarbazone substituted at the arylhydrazone moiety and phenyl group at different positions as shown in Scheme 1.

Results and discussion

Chemistry

All compounds synthesized (**1**–**9**) were obtained through precipitation by cooling the reaction medium, showing solid form with colors ranging from yellow to brown and yielded an average of 65.41 %.

The compounds **3**, **6,** and **9** have 1-phenylsemicarbazide **15** as a starting material showed the lowest yield. This is because the amide group is less reactive than the 4-phenylsemicarbazide **14** and 4-phenyl-3-thiosemicarbazide **13** amine portion. The compounds were initially elucidated by infrared spectroscopy (IR) on a SHIMADZU, model IR AFFINITY, with the following major shifts observed in the IR spectra: 1602.91–1666.57 cm^{-1} due to C=N, which is the link that represents the formation of the Schiff base, altering the signals of the starting products, about 1,700 cm^{-1} due to C=O of aldehyde. The elucidation was confirmed by nuclear magnetic resonance (NMR) ^{1}H and ^{13}C on a Varian model Mercury VX 300, where the absence of the signal for the hydrogen on the carbonyl of aldehydes starting in 9.50–10.00 ppm was readily apparent. The confirmation was also made by the identification of Schiff base formation signaling CH=N, with 8.13 ppm in ^{1}H and 139 ppm in ^{13}C approximately for the compounds with phenyl at position 4 (**1**, **2**, **4**, **5**, **7**, **8**) and 5.84 ppm in ^{1}H and 160 ppm in ^{13}C for the compounds with phenyl in position 1 (**3**, **6**, **9**). In summary, all the synthesized compounds exhibited spectral data consistent with the structures.

Anti-Toxoplasma gondii action

The host cells were initially infected for 24 h with tachyzoites of *T. gondii*. Thereafter the majority of cells showed proliferating parasites, thus confirming infection. At this point, the cultures were treated with the compounds.

Quantitative analyses were performed to observe the anti-*T. gondii* action of the drugs.

As expected, untreated cells had a large percentage of infection, while treated cells showed decrease rates where the parasites were eliminated (Table 1; Fig. 1). The cytotoxic effect including morphological alteration or cell death and intracellular parasites elimination was considered.

The compounds **1**, **2**, and **3** (with 2,6-dimethoxy-4-hydroxy substitution) showed reduced or no toxicity to host cells until 0.1 mM.

However, only the compound **1** exhibited powerful toxicity from 0.5 (50 %) to 1.0 mM whereupon it reached 100 % of cell lethality. Compounds **4**, **5**, and **6** (with trihydroxy), with 2,3,4-trihydroxy substituted and **7**, **8**, and **9** (with 2,4,5-trihydroxy substitution) showed powerful toxicity after 0.1 mM treatment where compounds reached 100 %.

There was no cytotoxic effect for any compound at 0.01 mM. Compound **6** exhibited the greatest reduction of infection, (50 %), but still demonstrated severe toxic effects at all other concentrations as well.

Although the infection parameters and the number of parasites (Table 2) were similar. In case of both the infection and number of intracellular parasites parameters, the quantification was done considering an infected cell all

Scheme 1 Synthetic Scheme

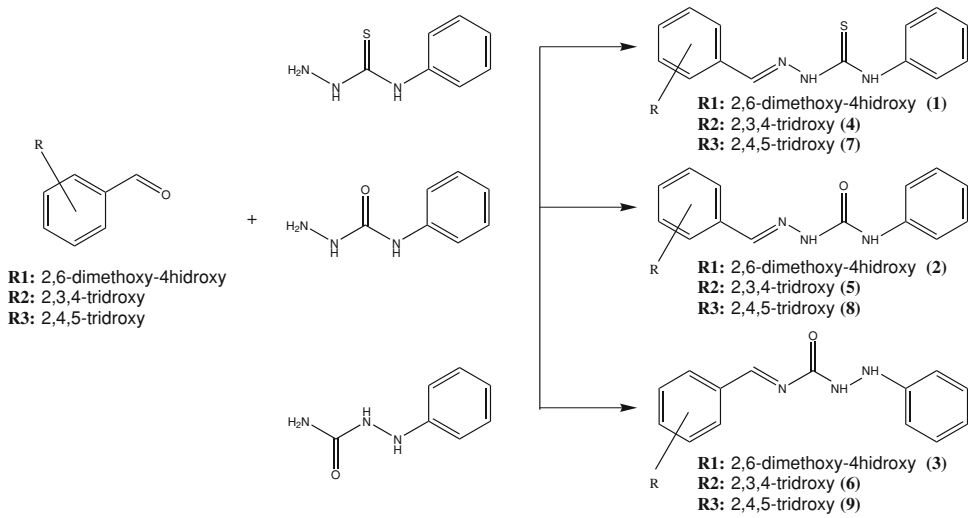

Table 1 Citotoxicity on Vero cells and LD50 values for cell culture and parasites of compounds (**1–9**)

Compound	Untreated	Treated (mM)				LD50 of cell culture	LD50 of Parasites
	Control	0.01	0.1	0.5	1.0		
1	583 ± 49	545 ± 37	697 ± 93	234 ± 28	0	0.7	0.2
2	661 ± 47	740 ± 56	755 ± 61	787 ± 53	837 ± 73	>1	0.5
3	551 ± 26	538 ± 32	497 ± 33	626 ± 43	635 ± 61	>1	0.6
4	492 ± 39	648 ± 50	748 ± 59	0	0	0.4	0.03
5	586 ± 36	627 ± 40	669 ± 41	0	0	0.3	0.2
6	527 ± 34	791 ± 56	0	0	0	0.07	0.02
7	624 ± 29	708 ± 37	695 ± 42	0	0	0.3	0.08
8	541 ± 15	742 ± 41	473 ± 23	0	0	0.3	0.006
9	601 ± 31	600 ± 30	597 ± 36	0	0	0.4	0.3
Sulfadiazine	420 ± 13	410 ± 13	515 ± 10	ND	500 ± 15	>10	6

ND not determined

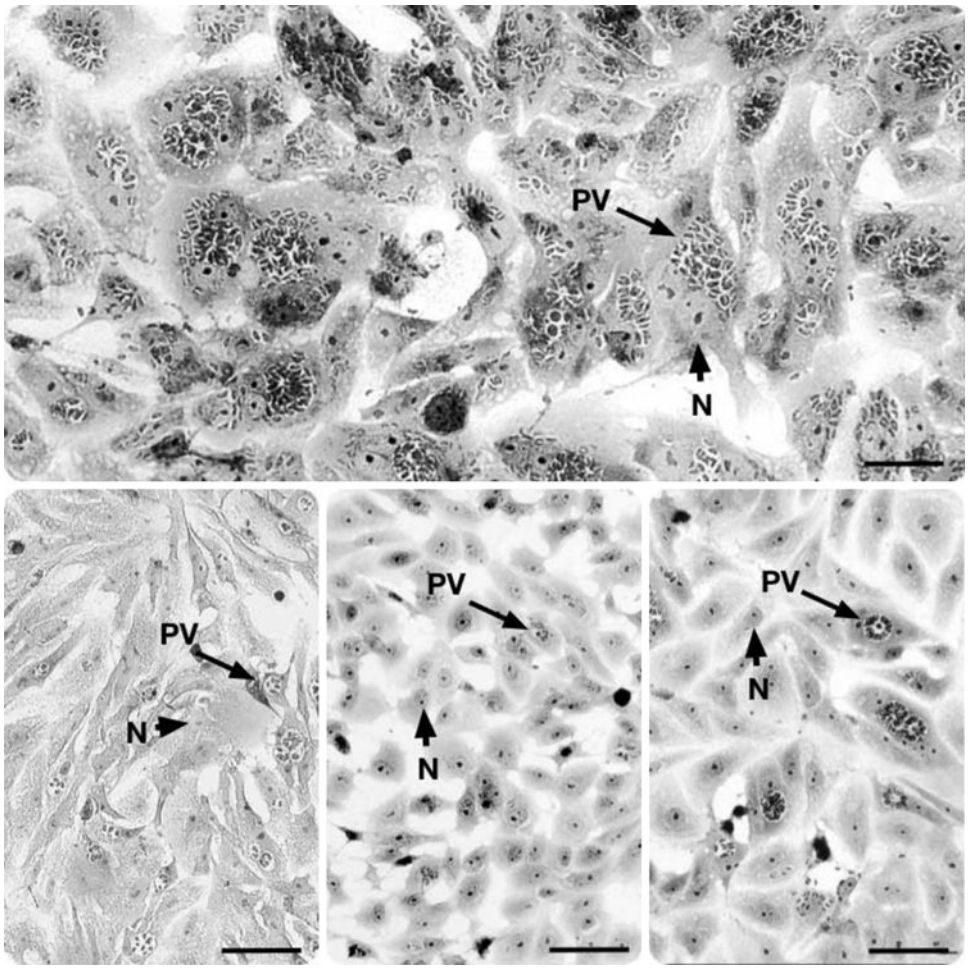

Fig. 1 Optical microscopy of biological test. *N* nuclei, *PV* parasitophorous vacuole

of them containing tachyzoites, and about the second parameter, only the parasites morphologically featured as "half-moon" were considered. The different effects between the compounds, considering the parameters above, are due to the efficiency of anti-parasitic action of each analogue as a result of it's molecular conformation.

Table 2 Effect of compounds (**1–9**) on cultures of Vero cells infected and intracelullar multiplication of *T. gondii*

Compound										
	Untreated	Treated (mM)				Untreated	Treated (mM)			
	Control	0.01	0.1	0.5	1.0	Control	0.01	0.1	0.5	1.0
1	75 ± 3	75 ± 4	35 ± 4	28 ± 1	0	2,759 ± 136	1,772 ± 86	1,029 ± 69	776 ± 76	0
2	77 ± 5	74 ± 4	69 ± 3	45 ± 2	28 ± 2	2,692 ± 98	2,402 ± 165	2,193 ± 167	1,278 ± 77	766 ± 65
3	80 ± 3	71 ± 4	78 ± 3	72 ± 3	37 ± 4	2,762 ± 127	2,225 ± 163	2,190 ± 87	1,945 ± 105	241 ± 28
4	65 ± 4	46 ± 2	37 ± 3	0	0	2,107 ± 152	1,310 ± 69	825 ± 65	0	0
5	79 ± 4	76 ± 3	49 ± 2	0	0	2,869 ± 163	2,460 ± 114	1,770 ± 68	0	0
6	78 ± 4	39 ± 2	0	0	0	2,754 ± 141	1,559 ± 115	0	0	0
7	78 ± 3	59 ± 2	42 ± 2	0	0	2,654 ± 106	1,966 ± 101	1,216 ± 101	0	0
8	72 ± 1	57 ± 2	28 ± 1	0	0	3,028 ± 234	528 ± 34	263 ± 18	0	0
9	77 ± 3	61 ± 2	37 ± 3	0	0	2,633 ± 114	1,761 ± 74	1,206 ± 77	0	0

Compound **3** induced the most drastic reduction in the number of parasites at 1.0 mM, followed by the compound **8** at 0.1 mM.

At lowest concentration, the derivative compounds of methoxy group did not show considerable results, and at 1.0 mM this was the only group which did not show cytotoxic effects. At 0.01 mM, the semicarbazones analogues were more effective while at 0.1 mM, the thiosemicarbazones showed better results.

Analyzing the position of the phenyl group, the radicals can be placed in the arylhydrazone position and the results of the isomers compared. No linearity was observed, as in each concentration different compounds stand out, as exemplified with compound **6** at 0.01 mM with the phenyl group at position 1 and the compound **8** at 0.1 mM with phenyl group at position 4.

Considering LD50, Compound **8** exhibited the best results because the concentration needed to reach parasite mortality is much lower than the lethal dose for the cell (Table 1), suggesting it can be a candidate for in vivo assays. Compound **8** performance is followed by Compound **4** and Compound **7**.

This study was performed out due to a need of new drugs that could improve toxoplasmosis treatment because current chemotherapy is extremely toxic. Anti-proliferative drugs are able to arrest the cell cycle inhibiting the synthesis of DNA, preventing their replications. The derivatives of thiosemicarbazones were chosen because of their wide range of pharmacological properties, including the anti-proliferative, anti-tumor (Chen *et al*., 2004), anti-viral (Kesel, 2011), anti-fungal (Qin *et al*., 2012), anti-bacterial (Turan-Zitouni *et al*., 2002), and anti-parasitic effects, such as anti-trypanosomal (Rodrigues *et al*., 2010) and anti-malarial (Oliveira *et al*., 2008). Their activities are related mainly with the inhibition of ribonucleotide reductase (Tenório *et al*., 2005). This class of compound is capable to bind heavy metal (Hutchinson, 1985), (Casas *et al*., 2000), (Chen *et al*., 2004) which changes their pharmacological activity (Hancock *et al*., 2011).

The nine new drugs proved more effective than the other derivatives for displaying a higher reduction rate at lower concentrations.

Experimental section

Chemistry

The nine new compounds (**1–9**) were synthesized through nucleophilic addition as described by (Aquino *et al*., 2008), were reacted 0.0029 mol of the semicarbazide compounds (**13**, **14**, and **15**) in a 10 mL solution EtOH/ H_2O (2:1), were added 0.0029 mol of benzaldehydes (**10**, **11**, and **12**) and 0.05 mL of acetic acid. The mixture was stirred under reflux for 1 h, then chilled in an ice bath. The precipitate was filtered under vacuum and washed with chilled ethanol.

Compound ***(1)****: 2.6-dimethoxy-4-hidroxybenzaldehyde-4-phenyl-3-thiosemicarbazone*

Solid with yellow color, and 76.61 % of yield; MP: 127.5 °C; IR (υ cm^{-1} KBr): 1602.91 (C=N); ^{1}H NMR (DMSO−d6, 300 MHz/ppm): δ 11.59 (s, 1H, NH), 10.04 (s, 1H, NH–Ar), 9.62 (s, 1H, OH), 8.40 (s, 1H, CH=N), 7.74−7.72 (d, 2H, J = 6.0 Hz, Ar–H), 7.39−7.33 (t, 2H, J = 9.0, 6.0 Hz, Ar H), 7.18−7.13 (t, 1H, J = 9.0, 6.0 Hz, Ar–H), 6.15 (s, 1H, Ar–H), 3.79 (s, 6H, OCH_3), ^{13}C NMR (DMSO−d6, 75.4 MHz/ppm):δ 174.04 (C=S), 161.07 (Car−OH), 160.36 (Car−OCH_3), 138.78 (CH=N), 138.72

(Car–NH), 128.31 (Car), 124.37 (Car), 122.57 (Car) 102.01 (Car), 92.32 (Car), 55.79 (CH_3).

*Compound **(2)**: 2.6-dimethoxy-4-hidroxybenzaldehyde-4-phenylsemicarbazone*

Solid with yellow color, and 69.72 % of yield; MP: 186.4 °C; IR (υ cm^{-1} KBr): 1655.00 (C=N); ^{1}H NMR (DMSO−d6, 300 MHz/ppm): δ 10.36 (s, 1H, OH), 9.90 (s, 1H, NH), 8.49 (s, 1H, NH–Ar), 8.13 (s, 1H, CH=N), 7.53−7.50 (d, 2H, $J = 9.0$ Hz, Ar–H), 7.32−7.27 (t, 2H, $J = 9.0$, 6.0 Hz, Ar–H), 7.02−6.97 (t, 1H, $J = 6.0$, 9.0 Hz, Ar–H), 6.15 (s, 1H, Ar–H), 3.80 (s, 6H, OCH_3), ^{13}C NMR (DMSO−d6, 75.4 MHz/ppm): δ 160.37 (Car−OCH_3), 159.87 (Car−OH), 152.94 (C=O), 138.98 (CH=N), 136.26 (Car−NH), 128.90 (Car), 122.17 (Car), 118.31 (Car), 102.40 (Car), 92.23 (Car), 58.81 (CH_3).

*Compound **(3)**: 2.6-dimethoxy-4-hidroxybenzaldehyde-1-phenylsemicarbazone*

Solid with yellow color, and 52.63 % of yield; MP: 152.9 °C; IR (υ cm^{-1} KBr): 1664.64 (C=N); ^{1}H NMR (DMSO−d6, 300 MHz/ppm): δ 7.65 (s, 1H, NH), 7.49 (s, 1H, NH–Ar), 7.17−7.12 (t, 2H, $J = 9.0$, 6.0 Hz, Ar–H), 6.72−6.70 (d, 2H, $J = 6.0$ Hz, Ar–H), 6.09 (s, 2H, Ar–H), 5.84 (s, 1H, CH=N), 3.76 (CH_3), ^{13}C NMR (DMSO−d6, 75.4 MHz/ppm): δ 185.12 (C=O), 164.88 (Car−CH_3), 163.58 (Car−OCH_3), 159.98 (CH=N), 149.44 (Car−NH), 128.56 (Car), 118.49 (Car), 112.09 (Car), 106.94 (Car), 91.82 (Car), 55.64 (CH_3).

*Compound **(4)**: 2.3.4-trihidroxybenzaldehyde-4-phenyl-3-thiosemicarbazone*

Solid with yellow color, and 67.36 % of yield; MP: 206.2 °C; IR (υ cm^{-1} KBr): 1631.85 (C=N); ^{1}H NMR (DMSO−d6, 300 MHz/ppm): δ 10.50 (s, 1H, NH), 9.86 (s, 1H, NH−Ar), 9.81 (s, 1H, OH), 8.34 (s, 1H, CH=N), 7.59−7.57 (d, 2H, $J = 6.0$ Hz, Ar–H), 7.35−7.30 (t, 2H, $J = 6.0$, 9.0 Hz, Ar–H), 7.17−7.13 (t, 1H, $J = 6.0$, 9.0 Hz, Ar–H), 7.09−7.06 (d, 1H, $J = 9.0$ Hz, Ar–H), 6.93−6.90 (d, 1H, $J = 9.0$ Hz, Ar–H), ^{13}C NMR (DMSO−d6, 75.4 MHz/ppm): δ 175.24 (C=S), 150.07 (C−OH), 148.39 (C−OH), 146.63 (C–OH), 139.16 (CH=N), 132.70 (Car–NH), 129.24 (Car), 127.90 (Car), 124.97 (Car), 111.12 (Car), 110.66 (Car), 107.72 (Car).

*Compound **(5)**: 2.3.4-trihidroxybenzaldehyde-4-phenylsemicarbazone*

Solid with yellow color, and 60.51 % of yield; MP: 185.2 °C; IR (υ cm^{-1} KBr): 1639.56 (C=N); ^{1}H NMR (DMSO−d6, 300 MHz/ppm): δ 10.30 (s, 1H, OH), 9.60 (s, 1H, NH), 9.31 (s, 1H, OH), 8.76 (s, 1H, OH), 8.33 (s, 1H, NH–Ar), 8.13 (s, 1H, CH=N), 7.60−7.57 (d, 2H, $J = 9.0$ Hz, Ar–H), 7.30−7.25 (t, 2H, $J = 6.0$, 9.0 Hz, Ar–H), 7.08−7.05 (d, 1H, Ar–H), 7.01−6.96 (t, 1H, $J = 6.0$, 9.0 Hz, Ar–H), 6.39−6.36 (d, 1H, $J = 9.0$ Hz, Ar–H), ^{13}C NMR (DMSO−d6, 75.4 MHz/ppm): δ 152.69 (C=O), 147.75 (C−OH), 146.16 (Car−OH), 141.64 (Car−OH), 139.24 (CH=N), 132.68 (Car−NH), 128.38 (Car), 122.07 (Car), 119.24 (Car), 118.54 (Car), 112.41 (Car), 107.52 (Car).

*Compound **(6)**: 2.3.4-trihidroxybenzaldehyde-1-phenylsemicarbazone*

Solid with yellow color, and 33.76 % of yield; MP: 137.2 °C; IR (υ cm^{-1} KBr): 1660.78 (C=N); ^{1}H NMR (DMSO−d6, 300 MHz/ppm): δ 7.66 (s, 1H, NH), 7.49 (s, 1H, NH−Ar), 7.18−7.12 (t, 2H, $J = 9.0$, 6.0 Hz, Ar–H), 7.11−7.08 (d, 1H, $J = 9.0$ Hz, Ar–H), 6.73.11−6.71 (d, 1H, $J = 6.0$ Hz, Ar–H), 6.50−6.47 (d, 1H, $J = 6.0$ Hz, Ar–H), 5.85 (s, 1H, CH=N), ^{13}C NMR (DMSO−d6, 75.4 MHz/ppm): δ 193.14 (C=O), 160.01 (CH=N), 153.25 (Car−OH), 150.89 (Car−OH), 149.44 (Car−OH), 132.16 (Car−NH), 128.57 (Car), 123.84 (Car), 118.52(Car), 115.41 (Car), 112.10 (Car), 108.35 (Car).

*Compound **(7)**: 2.4.5-trihidroxybenzaldehyde-4-phenyl-3-thiosemicarbazone*

Solid with yellow color, and 63.38 % of yield; MP: 127.7 °C; IR (υ cm^{-1} KBr): 1633.76 (C=N); ^{1}H NMR (DMSO−d6, 300 MHz/ppm): δ 11.47 (s, 1H, NH), 9.87 (s, 1H, NH−Ar), 9.81 (s, 1H, OH), 8.34 (s, 1H, CH=N), 7.62−7.59 (d, 2H, $J = 9.0$ Hz, Ar–H), 7.37−7.32 (t, 2H, $J = 9.0$, 6.0 Hz, Ar–H), 7.29 (s, 1H, Ar–H), 7.19−7.14 (t, 1H, J=6.0, 9.0 Hz, Ar–H), 6.98 (s, 1H, Ar–H), ^{13}C NMR (DMSO−d6, 75.4 MHz/ppm): δ 174.84 (C=S), 151.49 (C−OH), 149.80 (C−OH), 142.09 (C−OH), 139.16 (CH=N), 138.64 (Car−NH), 129.73 (Car), 128.40 (Car), 125.34 (Car), 113.32 (Car), 111.05 (Car), 103.75 (Car).

*Compound (**8**): 2.4.5-trihidroxybenzaldehyde-4-phenylsemicarbazone*

Solid with brown color, and 81.66 % of yield; MP: 162.0 °C; IR (υ cm^{-1} KBr): 1666.57 (C=N); ^{1}H NMR (DMSO−d6, 300 MHz/ppm): δ 10.24 (s, 1H, OH), 9.55 (s, 1H, NH), 9.35 (s, 1H, OH), 9.29 (s, 1H, OH), 8.67 (s, 1H, NH–Ar), 8.09 (s, 1H, CH=N), 7.90 (s, 1H, Ar–H), 7.29−7.23 (t, 2H, $J = 9.0$, 9.0 Hz, Ar–H), 7.14 (s, 1H, Ar–H), 7.00−6.95 (t, 1H, $J = 9.0$, 6.0 Hz, Ar–H), 6.34−6.32 (d, 1H, $J = 6.0$ Hz, Ar–H), ^{13}C NMR (DMSO−d6,

75.4 MHz/ppm): δ 153.60 (C=O), 151.02 (C−OH), 149.16 (Car−OH), 140.94 (Car−OH), 139.24 (CH=N), 129.28 (Car−NH), 129.18 (Car), 122.87 (Car), 120.02 (Car), 119.20 (Car), 113.73 (Car), 111.57 (Car).

*Compound (**9**): 2.4.5-trihidroxybenzaldehyde-1-phenylsemicarbazone*

Solid with brown color, and 54.91 % of yield; MP: 137.5 °C; IR (υ cm^{-1} KBr): 1620.27 (C=N); ^{1}H NMR (DMSO−d6, 300 MHz/ppm): δ 7.68 (s, 1H, NH), 7.50 (s, 1H, NH−Ar), 7.18−7.12 (t, 2H, $J = 9.0$, 6.0 Hz, Ar–H), 6.98 (s, 1H, Ar–H), 6.73−6.71 (d, 1H, $J = 6.0$ Hz, Ar–H), 6.36 (s, 1H, Ar–H), 5.87 (s, 1H, CH=N), ^{13}C NMR (DMSO−d6, 75.4 MHz/ppm): δ 190.36 (C=O), 160.10 (CH=N), 156.57 (Car−OH), 154.60 (Car−OH), 149.44 (Car−OH), 139.06 (Car−NH), 128.60 (Car), 118.58 (Car), 114.49 (Car), 112.14(Car), 103.00 (Car).

Biological assays

Vero cells (kidney fibroblasts of the African monkey) were plated with DMEM 1152 Sigma supplemented with 5 % of serum fetal bovine on 24-well tissue plates containing a sterile round coverslip and maintained at 37 °C , 24 h. Next, the cells were infected with tachyzoites of *T. gondii*, RH strain (5:1) at 37 °C, for other 24 h. The infected culture was incubated with the compounds at concentrations 0.01, 0.1, 0.5, and 1 mM during 24 h and then, processed for light microscopy.

The concentrations were selected within range of 0.01–1.0 mM, in accord with the preliminary experiments and previous studies carried out by (Carvalho *et al*., 2010).

All compounds were dissolved in DMSO in final concentration of 1.5 % v/v, that was not toxic for both host cell and parasites.

After treatment, the culture was washed three times with Phosphate Buffer Solution (PBS) and fixed for 1 h using 4 % paraformaldehyde. It was then stained using GIEMSA (1:9) diluted in distilled water for 6 h at room temperature.

The cell quantification was done by observing random fields of eight samples and at least, 300 cells were counted of each one using optical microscopy AXIOPLAN Zeis Germany in each concentration with all compounds. The number of infected and uninfected cells and intracellular parasites were determined. The cytotoxic effect was considered by observing morphological alterations on cells and number of control cells compared with treated cells.

Conclusion

(Benzaldehyde)-4-phenyl-3-thiosemicarbazone and (benzaldehyde)-(4 or 1)-pheny-3-semicarbazone, were synthesized and characterized based on their physical, analytical, and spectral data.

The new synthesized analogues had their anti-*T. gondii* activity analyzed in vitro. All compounds showed activity in all concentrations tested, reducing the percentage of infected cell and number of intracellular parasites, mainly at 0.01 and 1.0 mM, respectively. These news compounds were more effective than the reference drugs Sulfadiazine.

Changing on molecular conformations and modifications on the ring at arylhydrazone moiety altered the activity of the drugs. These results indicate that the new class of drugs representing an interesting model for development of new medicines against toxoplasmosis and others parasitic diseases.

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