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New 4-Amino-2-azabicyclo[3.2.2]nonane Derivatives and their Antiprotozoal Potencies

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Summary. Several 2-substituted 4-amino-2-azabicyclo [3.2.2]nonane derivatives were synthesized. The new compounds were investigated for their activities against the causative organism of East African sleeping sickness, *Trypanosoma b. rhodesiense*, and a protozoan parasite which causes Malaria tropica, *Plasmodium falciparum* K_1, a strain which is resistant to chloroquine and pyrimethamine. The results are compared to the activities of 2-unsubstituted 4-amino-2-azabicyclo[3.2.2]nonanes.

Keywords. Antiplasmodial activity; Antitrypanosomal activity; Amino alcohols; Alkylations; Structure-activity relationships.

Introduction

Recently we reported the synthesis and the antiprotozoal activities of some 4-amino-2-azabicyclo [3.2.2]nonanes [1]. They have been prepared from 4-aminobicyclo[2.2.2]octan-2-ones **1**, which were converted to 4-amino-2-azabicyclo[3.2.2]nonan-3-ones **2** *via* a *Beckmann* rearrangement. The latter were reduced giving 4-amino-2-azabicyclo[3.2.2]nonanes **3** (Scheme 1). Those compounds showed in an *in vitro* assay distinct antiplasmodial and antitrypanosomal activity and compound **3a** exhibited an inhibition of 35% of *P. berghei* in an *in vivo* test [1]. Because of these promising results, we subjected those compounds to further derivatizations to optimize their biological activities.

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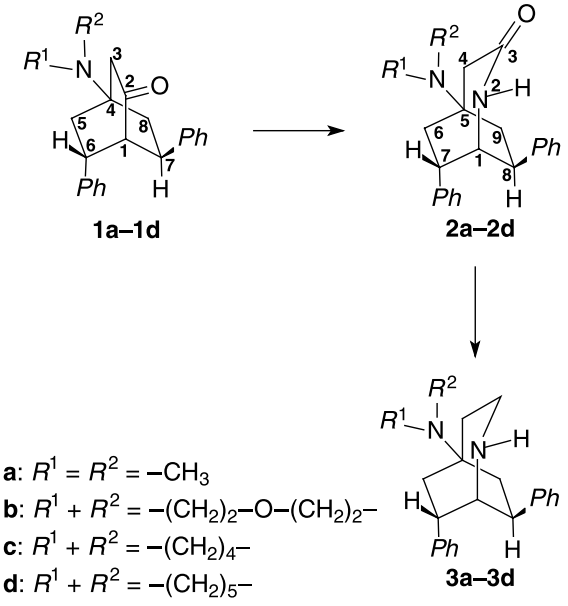
This paper reports the preparation of more hydrophilic compounds by alkylation of the nitrogen in ring position 2 with substituents bearing neutral, acidic, or basic groups. Those groups should be linked *via* a short alkyl chain. For that reason, we chose ethyl bromoacetate as reagent giving acetic acid esters, which were converted to the corresponding amino alcohols or to amino acids. Since sulfonyl derivatives of the 4-aminobicyclo[2.2.2]octane series have shown good antitrypanosomal activities [2] we prepared sulfonamides of the corresponding 4-amino-2-azabicyclo[3.2.2]nonanes.

Results and Discussion

Syntheses

N-Alkylation of 2-azabicyclo[3.2.2]nonanes gave with ethyl bromoacetate compounds **4**, which were reduced to the amino alcohols **5** in good yields using $LiAlH_4$. Compound **5d** was esterified with isonicotinic acid chloride to **6d**. The amino acid **7c** was accessible by the saponification of **4c**. The sulfonamides **8** and **9** were prepared from the reaction of diamines **3** with 4-tolylsulfonyl chloride and biphenyl-4-sulfonyl chloride (Scheme 2).

The structures of the new compounds were established using NMR spectroscopy: N-alkylation of **3** to compounds **4** caused a shift of 4.9 ppm for C-3 and a shift of 7.5 ppm for C-1 to lower field. The



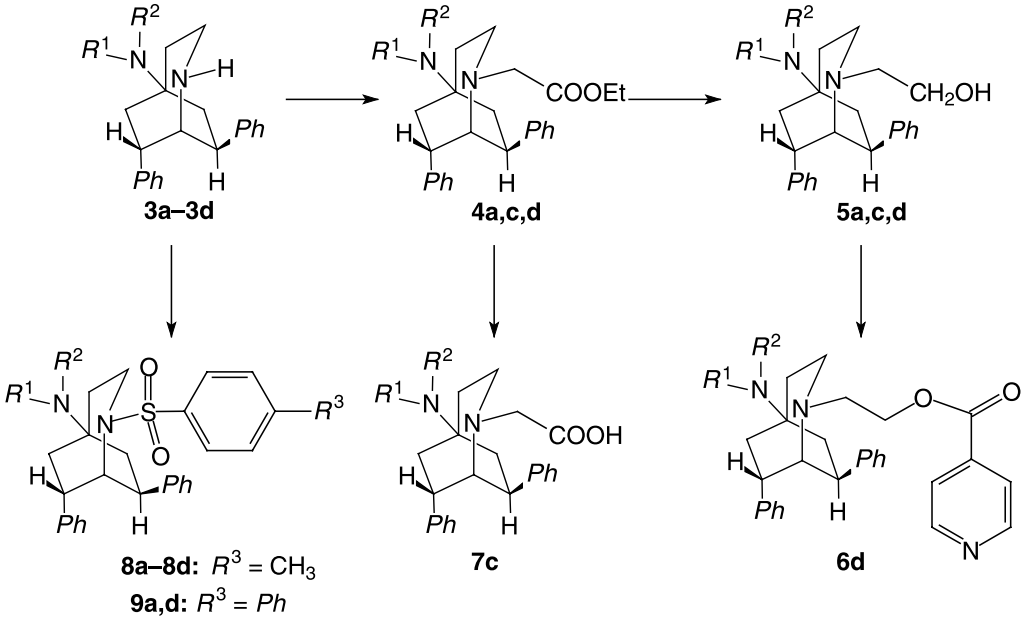
Scheme 1

alkylation of the ring nitrogen was revealed by long range couplings from C-1 and C-3 to the protons of the CH_2 group of the new substituent. The successful reduction of esters **4** to amino alcohols **5** was detected by the replacement of the carbonyl resonance at 172 ppm by a new signal at 59 ppm, which is typical for a CH_2OH group. Due to the formation of sulfonamides **8** and **9** the resonances of C-1 and C-3 were shifted 0.5–1.0 ppm to higher frequencies compared to **3**.

Antiprotozoal Activities and Cytotoxicity

All new compounds **4**–**9** were screened for their activities against *Trypanosoma b. rhodesiense* and *Plasmodium falciparum* K_1 by a medium throughput screening at two concentrations (4.85 $\mu g/cm^3$ and 0.81 $\mu g/cm^3$). In addition, the IC_{50} values of selected compounds were determined. The cytotoxicity was measured using L-6 cells (Table 1).

All of the prepared N-alkylated derivatives **4**–**7** of bicyclononanes **3** were far less active against *P. falciparum* K_1 and *T. b. rhodesiense*. In contrast, most of the sulfonamides **8** and **9** showed quite good antiprotozoal activities. The most active compound against both tested parasites was **8a** ($IC_{50}(P.f.) = 0.43\ \mu M$, $IC_{50}(T.b.r.) = 0.76\ \mu M$). However, it is less active and more cytotoxic than its 2-unsubstituted analogue **3a**.



Scheme 2

Table 1. Activities of compounds **2–9** expressed as IC_{50} (μM)[a]

Compd.	*P. falciparum* K_1	*T. b. rhodesiense*	Cytotox. L6-cells
2a	1.40	37.97	>269.1
2b	>13.28	138.4	>239.1
2c	8.76	37.94	>249.7
2d	13.00	36.60	233.6
3a	0.28	0.60	108.8
3b	6.84	9.44	>206.7
3c	0.56	1.16	120.4
3d	0.64	6.57	89.74
4a	2.66	2.28	27.62
4c	nt	4.98	27.59
5a	7.54	10.36	98.75
8a	0.43	0.76	9.27
8b	4.75	9.11	71.89
8c	1.30	1.26	9.42
8d	1.26	2.65	10.50
9a	1.03	1.06	3.62
9d	0.85	1.45	7.33
chl	0.062		
sur		0.011	
mef			4.3

[a] Values represent the average of four determinations (two determinations of two independent experiments), *nt* not tested. *chl* Chloroquine, *mef* mefloquine, *sur* suramine

Conclusion

Several 2-alkylated 4-amino-2-azabicyclo[3.2.2]nonane derivatives were prepared. These compounds revealed *in vitro* decreased antiprotozoal activities compared to their unsubstituted analogues. If this loss of activity was generally due to N-alkylation or rather to the insertion of the polar groups will be determined by the preparation of the corresponding non-polar N-alkyl-2-azabicyclononanes.

The synthesized sulfonamides of 4-amino-2-azabicyclo[3.2.2]nonanes showed good antiplasmodial and antitrypanosomal activities, but they were more toxic than the parent 2-unsubstituted nonanes. If their carboxylic acid analogues have similar properties will be investigated in a future project.

Experimental

Melting points were obtained on a digital melting point apparatus Electrothermal IA 9200. IR spectra: infrared spectrometer system 2000 FT (Perkin Elmer). UV/VIS: Lambda 17 UV/VIS-spectrometer (Perkin Elmer). NMR spectra: Varian Inova 400 (300 K), 5 mm tubes, solvent resonance as internal standard. ^{1}H- and ^{13}C-resonances were assigned using ^{1}H,^{1}H- and ^{1}H,^{13}C-correlation spectra. ^{1}H- and ^{13}C-resonances are numbered as given in the formulae. MS, HR-MS: Kratos profile spectrometer 70 eV electron impact. Microanalyses: EA 1108 CHNS-O apparatus (Carlo Erba), Microanalytical Laboratory at the Institute of Physical Chemistry, Vienna; their values were in satisfactory agreement with the calculated ones. Materials: column-chromatography (CC): silica gel 60 (Merck 70–230 mesh, pore-diameter 60 Å); thin-layer chromatography (TLC): TLC plates (Merck, silica gel 60 F_{254} 0.2 mm, 200×200 mm^2); the substances were detected in UV light at 254 nm.

The preparation of bicyclooctanones **1a–1d**, bicyclononanones **2a–2d**, and bicyclononanes **3a–3d** has been reported [1, 3].

*Preparation of Ethyl-2-(2-azabicyclonon-2-yl) acetates **4a**, **4c**, and **4d***

Diamines **3a**, **3c**, and **3d** were dissolved in ethanol and cooled on an ice bath. Ethyl bromoacetate was added dropwise using a syringe. The solution was refluxed for 48 h at 100°C. Then H_2O was added and it was alkalized with 2 *M* NaOH until turbidity persisted. The mixture was extracted 3 times with ether. The combined organic layers were washed twice with H_2O and dried (Na_2SO_4). The solvent was evaporated *in vacuo* and the residue was further purified.

(7RS,8RS)-(±)-Ethyl-2-(5-dimethylamino-7,8-diphenyl-2-azabicyclo[3.2.2]non-2-yl) acetate (**4a**, $C_{26}H_{34}N_2O_2$)

In 30 cm^3 ethanol 1.17 g **3a** (3.65 mmol) reacted with 610 mg ethyl bromoacetate (3.65 mmol) giving a residue which was dissolved in ethanol. The product crystallized upon stirring and dropwise addition of H_2O giving 830 mg (56%) **4a** as white crystals. Mp 110°C; ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.19$ (t, $J = 7.0$ Hz, CH_2CH_3), 1.87–1.98 (m, 4-H, 6-H), 2.13 (dd, $J = 13.0$, 10.6 Hz, 9-H), 2.20–2.30 (m, 6-H, 9-H), 2.31 (s, $N(CH_3)_2$), 2.88–2.93 (m, 1-H, 3-H), 3.03–3.10 (m, 8-H, CH_2COO), 3.27 (d, $J = 17.1$ Hz, CH_2COO), 3.33 (ddd, $J = 13.3$, 8.5, 5.5 Hz, 3-H), 3.89 (t, $J = 9.2$ Hz, 7-H), 4.08 (ddd, $J = 14.1$, 7.2, 2.2 Hz, CH_2CH_3), 7.14–7.45 (m, aromatic H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): $\delta =$ 14.19 (CH_3), 31.96 (C-4), 34.62 (C-6), 38.03 ($N(CH_3)_2$), 38.29 (C-8) 38.48 (C-9), 41.11 (C-7), 46.74 (C-3), 57.68 (C-5), 60.17 (CH_2CH_3), 60.30 (CH_2COO), 69.05 (C-1), 125.99, 127.67, 127.99, 128.43, 128.47, 144.90, 145.70 (aromatic C), 172.03 (COO) ppm; IR (KBr): $\bar{\nu} = 2932$, 2776, 1736, 1493, 1188, 1168, 1022, 760, 703 cm^{-1}; UV (CH_2Cl_2): λ (logε) = 233 (3.846) nm; MS (70 eV): m/z (%) = 406 (M^+), 361, 333, 257, 231, 198, 188, 158, 144, 125, 104, 91; HRMS (EI+): calcd. ($C_{26}H_{34}N_2O_2$): 406.26203; found: 406.25884.

(7RS,8RS)-(±)-Ethyl-2-(7,8-diphenyl-5-pyrrolidino-2-azabicyclo[3.2.2]non-2-yl) acetate (**4c**, $C_{28}H_{36}N_2O_2$)

In 35 cm^3 ethanol 1.44 g **3c** (4.16 mmol) reacted with 691 mg ethyl bromo acetate (4.14 mmol) giving a residue which was purified by CC using CH_2Cl_2:MeOH = 4:2 as eluent yielding 816 mg (45%) **4c** as yellowish resin. ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.84$ (t, $J = 7.2$ Hz, CH_3), 1.77 (br, s, $(CH_2)_2$), 1.94–2.09 (m, 4-H, 6-H), 2.11–2.26 (m, 6-H, 9-H), 2.70–2.82

(m, $N(CH_2)_2$), 2.89–2.95 (m, 1-H, 3-H), 3.03–3.11 (m, 8-H, CH_2COO), 3.27 (d, $J = 14.0$ Hz, CH_2COO), 3.36 (ddd, $J =$ 13.3, 8.9, 5.1 Hz, 3-H), 3.90 (t, $J = 9.4$ Hz, 7-H), 4.03–4.11 (m, CH_2CH_3), 7.14–7.46 (m, aromatic H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 14.20$ (CH_3), 23.69 ($(CH_2)_2$), 34.65 (C-4), 35.47 (C-6), 37.70 (C-9), 38.38 (C-8), 40.97 (C-7), 45.18 ($N(CH_2)_2$), 46.81 (C-3), 56.98 (C-5), 60.18, 60.36 (CH_2CH_3, CH_2COO), 69.15 (C-1), 125.98, 127.68, 127.99, 128.43, 128.47, 144.90, 145.80 (aromatic C), 172.04 (COO) ppm; IR (KBr): $\bar{\nu} = 3025$, 2932, 2871, 2805, 1737, 1600, 1493, 1452, 1369, 1315, 1266, 1188, 1119, 1030, 758, 746, 700 cm^{-1}; UV (CH_2Cl_2): λ ($\log\varepsilon$) = 233 (3.837) nm; HRMS (MALDI): calcd. ($C_{28}H_{36}N_2O_2Na^+$): 455.2674; found: 455.2635.

(7RS,8RS)-(±)-Ethyl-2-(7,8-diphenyl-5-piperidino-2-azabicyclo[3.2.2]non-2-yl) acetate (**4d**, $C_{29}H_{38}N_2O_2$)
In 40 cm^3 ethanol 1.69 g **3d** (4.69 mmol) reacted with 781 mg ethyl bromo acetate (4.68 mmol). The residue was purified by CC using CH_2Cl_2:MeOH = 4:1 as eluent giving 820 mg (39%) **4d** as yellowish resin. ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.18$ (t, $J = 7.2$ Hz, CH_3), 1.40–1.48 (m, CH_2), 1.54–1.64 (m, $2CH_2$), 1.87–1.95 (m, 4-H, 6-H), 2.12 (br, t, $J = 11.6$ Hz, 9-H), 2.22–2.35 (m, 6-H, 9-H), 2.60 (br, s, $N(CH_2)_2$), 2.92 (ddd, $J = 12.3$, 5.8, 5.8 Hz, 3-H), 2.98 (s, 1-H), 3.05–3.11 (m, 8-H, CH_2COO), 3.25–3.32 (m, 3-H, CH_2COO), 3.82 (t, $J =$ 9.4 Hz, 7-H), 4.04–4.10 (m, CH_2CH_3), 7.13–7.45 (m, aromatic H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 14.16$ (CH_3), 25.08 (CH_2), 26.93 ($2CH_2$), 32.87 (C-4), 34.56 (C-6), 38.63 (C-8), 38.80 (C-9), 41.49 (C-7), 46.23 ($N(CH_2)_2$), 47.11 (C-3), 58.21 (C-5), 60.13, 60.16 (CH_2CH_3, CH_2COO), 68.82 (C-1), 125.92, 125.94, 127.58, 127.99, 128.32, 128.35, 144.95, 145.72 (aromatic C), 171.97 (COO) ppm; IR (KBr): $\bar{\nu} =$ 2977, 2929, 2850, 2792, 1737, 1600, 1493, 1451, 1262, 1187, 1154, 1097, 1031, 758, 744, 700 cm^{-1}; UV (CH_2Cl_2): λ ($\log\varepsilon$) = 233 (3.812) nm; HRMS (MALDI): calcd. ($C_{29}H_{38}N_2O_2Na^+$): 469.2831; found: 469.2886.

*Preparation of Amino Ethanols **5a**, **5c**, and **5d***
Compounds **4a**, **4c**, and **4d** were dissolved in dry ether and $LiAlH_4$ was added in portions under stirring and cooling on an ice bath. After 1 h the mixture was refluxed over night at 50°C. The reaction was quenched cautiously by dropwise addition of 2 *M* NaOH under cooling on an ice bath. The mixture was extracted 5 times with ether. The combined organic layers were washed twice with H_2O, dried (Na_2SO_4), and filtered. The solvent was evaporated *in vacuo* giving amino ethanols **5a**, **5c**, and **5d** as colourless resins.

(7RS,8RS)-(±)-2-(5-Dimethylamino-7,8-diphenyl-2-azabicyclo[3.2.2]non-2-yl)-ethanol (**5a**, $C_{24}H_{32}N_2O$)
A solution of 500 mg **4a** (1.23 mmol) in 20 cm^3 of dry ether was treated with 186 mg $LiAlH_4$ (4.90 mmol) giving 436 mg (98%) **5a** as a colourless resin. The dihydrochloride was prepared by treatment of a solution of the diamine in CH_2Cl_2 with excess HCl (2 *M* solution in diethyl ether) and subsequent evaporation of the solvents *in vacuo*. The residue crystallized from ethyl acetate. Mp (HCl): 200°C; ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.91$–1.94 (m, 4-H), 1.98 (dd, $J =$ 13.9, 7.9 Hz, 6-H), 2.12 (br, t, $J = 12.5$ Hz, 9-H), 2.26–2.39 (m, 6-H, 9-H), 2.34 (s, $N(CH_3)_2$), 2.45 (ddd, $J = 12.9$, 5.7, 3.8 Hz, NCH_2), 2.55 (ddd, $J = 12.7$, 6.9, 3.8 Hz, NCH_2), 2.73 (d, $J = 3.4$ Hz, 1-H), 2.76–2.82 (m, 3-H), 2.95–3.05 (m, 3-H, CH_2OH), 3.19–3.24 (m, CH_2OH), 3.29 (ddd, $J = 11.1$, 7.3, 3.8 Hz, 8-H), 3.44 (br, t, $J = 8.8$ Hz, 7-H), 7.17–7.38 (m, aromatic H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 30.90$ (C-4), 34.99 (C-6), 35.97 (C-9), 37.98 (C-8, $N(CH_3)_2$), 40.39 (C-7), 48.30 (C-3), 57.38 (C-5), 58.81 (CH_2OH), 58.99 (NCH_2), 69.34 (C-1), 126.21, 126.27, 127.45, 128.20, 128.40, 128.66, 144.29, 145.67 (aromatic C) ppm; IR (KBr): $\bar{\nu} = 3430$, 2932, 2823, 2782, 1631, 1600, 1494, 1450, 1156, 1040, 757, 743, 699 cm^{-1}; UV (CH_2Cl_2): λ ($\log\varepsilon$) = 232 (3.843) nm; MS (70 eV): m/z (%) = 364 (M^+), 319, 215, 188, 176, 158, 130, 115, 104; HRMS (EI+): calcd. ($C_{24}H_{32}N_2O$): 364.25146; found: 364.24999.

(7RS,8RS)-(±)-2-(7,8-Diphenyl-5-pyrrolidino-2-azabicyclo[3.2.2]non-2-yl)-ethanol (**5c**, $C_{26}H_{34}N_2O$)
A solution of 816 mg **4c** (1.89 mmol) in 30 cm^3 dry ether was treated with 286 mg $LiAlH_4$ (7.53 mmol) giving 593 mg (81%) **5c** as a colourless resin. ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.78$ (br, s, $(CH_2)_2$), 1.98–2.01 (m, 4-H), 2.09 (dd, $J = 13.7$, 7.9 Hz, 6-H), 2.19–2.29 (m, 6-H, 9-H), 2.33 (ddd, $J = 13.7$, 7.3, 2.6 Hz, 9-H), 2.45 (ddd, $J = 13.1$, 5.6, 3.8 Hz, NCH_2), 2.55 (ddd, $J = 13.1$, 7.2, 3.7 Hz, NCH_2), 2.73–2.84 (m, 1-H, 3-H, $N(CH_2)_2$), 2.95–3.05 (m, 3-H, CH_2OH), 3.17–3.25 (m, CH_2OH), 3.31 (ddd, $J = 11.1$, 7.2, 3.8 Hz, 8-H), 3.44 (br, t, $J = 8.9$ Hz, 7-H), 7.17–7.38 (m, aromatic H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 23.61$ ($(CH_2)_2$), 32.89 (C-4), 35.68 (C-6), 36.30 (C-9), 37.94 (C-8), 40.16 (C-7), 45.11 ($N(CH_2)_2$), 48.40 (C-3), 56.51 (C-5), 58.82, 59.00 (CH_2OH, NCH_2), 69.54 (C-1), 126.16, 126.23, 127.51, 128.17, 128.44, 128.63, 144.36, 145.83 (aromatic C) ppm; IR (KBr): $\bar{\nu} = 2932$, 2872, 2816, 1600, 1493, 1449, 1156, 1105, 1051, 746, 700 cm^{-1}; UV (CH_2Cl_2): λ ($\log\varepsilon$) = 232 (3.836) nm; HRMS (MALDI): calcd. ($C_{26}H_{35}N_2O^+$): 391.2749; found: 391.2780.

(7RS,8RS)-(±)-2-(7,8-Diphenyl-5-piperidino-2-azabicyclo[3.2.2]non-2-yl)-ethanol (**5d**, $C_{27}H_{36}N_2O$)
A solution of 820 mg **4d** (1.84 mmol) in 30 cm^3 of dry ether was treated with 277 mg $LiAlH_4$ (7.3 mmol) giving 545 mg (73%) of pure **5d** as a colourless resin. 150 mg were purified for analytical purposes by CC over silica gel using CH_2Cl_2 as eluent. ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.42$–1.48 (m, CH_2), 1.57–1.63 (m, $2CH_2$), 1.91–1.97 (m, 4-H, 6-H), 2.11 (br, t, $J = 12.5$ Hz, 9-H), 2.29 (ddd, $J = 12.8$, 9.7, 2.4 Hz, 6-H), 2.37 (ddd, $J = 13.5$, 7.8, 2.6 Hz, 9-H), 2.46 (ddd, $J = 13.0$, 5.8, 3.9 Hz, NCH_2), 2.55 (ddd, $J = 13.0$, 6.8, 4.1 Hz, NCH_2), 2.62 (br, s, $N(CH_2)_2$), 2.76–2.82 (m, 1-H, 3-H), 2.99 (ddd, $J = 12.6$, 6.0, 6.0 Hz, 3-H), 3.07 (ddd, $J = 10.6$, 6.5, 3.8 Hz, CH_2OH), 3.20–3.25 (m, CH_2OH), 3.29 (ddd, $J = 11.3$, 8.0, 3.6 Hz, 8-H), 3.41 (br, t, $J = 9.1$ Hz, 7-H), 7.17–7.37 (m, aromatic H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 25.05$ (CH_2), 26.87 ($2CH_2$), 32.31 (C-4), 35.02 (C-6), 35.42 (C-9), 38.51 (C-8), 41.23 (C-7), 46.22 ($N(CH_2)_2$), 48.31 (C-3), 57.97

(C-5), 58.70, 58.83 (CH_2OH, NCH_2), 69.28 (C-1), 126.18, 126.23, 127.38, 128.21, 128.27, 128.63, 144.28, 145.78 (aromatic C) ppm; IR (KBr): $\bar{\nu} = 2930$, 2852, 2807, 1600, 1493, 1449, 1156, 1096, 1055, 1032, 757, 699 cm^{-1}; UV (CH_2Cl_2): λ ($\log\varepsilon$) = 232 (3.855) nm; HRMS (MALDI): calcd. ($C_{27}H_{36}N_2ONa^+$): 427.2725; found: 427.2748.

(7RS,8RS)-(±)-2-(7,8-Diphenyl-5-piperidino-2-azabicyclo [3.2.2]non-2-yl)-ethyl isonicotinate (**6d**, $C_{33}H_{42}Cl_3N_3O_2$)
A solution of 326 mg (2.30 mmol) isonicotinic acid chloride in 2 cm^3 CH_2Cl_2 was added dropwise to an ice-cooled solution of 450 mg **5d** (1.11 mmol) and 13.7 mg (0.112 mmol) 4-*DMAP* in 8 cm^3 CH_2Cl_2. The mixture was stirred under an Ar atmosphere for 2 h at room temperature and then cautiously shaken 4 times with H_2O, 3 times with 2 *M* NaOH, and again with H_2O until the aqueous layer was neutral. The organic layer was dried (Na_2SO_4), filtered, and the solvent was evaporated *in vacuo*. The residue was purified over basic Al_2O_3 using CH_2Cl_2 as eluent giving 290 mg (51%) **6d** as yellowish resin. For analytical purposes the trihydrochloride was prepared by treatment of a solution of **6d** in CH_2Cl_2 with excess of HCl (2 *M* solution in diethyl ether). The solvents were evaporated *in vacuo* and the residue recrystallized once from ethanol/ethyl acetate and finally from ethanol. Mp 219°C; ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.40$–1.48 (m, CH_2), 1.54–1.65 (m, $2CH_2$), 1.85–1.98 (m, 4-H, 6-H), 2.11 (br, t, $J = 12.0$ Hz, 9-H), 2.22–2.32 (m, 6-H, 9-H), 2.54–2.65 (m, $N(CH_2)_2$), 2.74–2.84 (m, NCH_2), 2.97–3.12 (m, 1-H, 3-H), 3.21 (ddd, $J = 10.8$, 8.5, 2.2 Hz, 8-H), 3.43 (br, t, $J = 9.2$ Hz, 7-H), 4.00–4.13 (m, CH_2OH), 7.08–7.35 (m, aromatic H), 7.66 (d, $J = 5.5$ Hz, heteroaromatic H), 8.70 (d, $J = 5.5$ Hz, heteroaromatic H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 25.06$ (CH_2), 26.87 ($2CH_2$), 32.13 (C-4), 35.54 (C-6), 36.81 (C-9), 39.08 (C-8), 42.08 (C-7), 46.24 ($N(CH_2)_2$), 48.46 (C-3), 55.96 (NCH_2), 58.04 (C-5), 64.35 (CH_2OH), 68.93 (C-1), 122.76, 125.98, 126.16, 127.26, 127.90, 128.40, 128.62, 150.46 (aromatic C), 137.37, 144.49, 145.78 (aromatic C_q), 164.86 (COO) ppm; IR (KBr): $\bar{\nu} = 3422$, 2951, 2653, 2533, 2363, 1742, 1637, 1602, 1498, 1451, 1283, 1130, 1002, 753, 703, 678 cm^{-1}; UV (CH_3OH): λ ($\log\varepsilon$) = 207 (4.378) nm; HRMS (MALDI): calcd. ($C_{33}H_{39}N_3O_2Na^+$): 532.2940; found: 532.2963.

(7RS,8RS)-(±)-2-(7,8-Diphenyl-5-pyrrolidino-2-azabicyclo[3.2.2]non-2-yl) acetic acid dihydrochloride (**7c**, $C_{26}H_{34}Cl_2N_2O_2$)
A suspension of 0.5 g **4c** (1.16 mmol) in 100 cm^3 HCl_{conc} was heated in a 250 cm^3 round bottomed flask which was connected with a distillation apparatus. Most of the generated ethanol was distilled off on an oil bath at 160°C within 4–5 h. Then the reaction mixture was refluxed over night and the solvent evaporated *in vacuo*. The residue was dissolved in the minimum amount of ethanol and this solution was dropped into acetone giving a white precipitate which was sucked off and dried under reduced pressure yielding 380 mg (69%) **7c**. Mp (HCl, decomp.): 233–235°C; ^{1}H NMR (400 MHz, CD_3OD): $\delta = 2.11$–2.25 (m, $(CH_2)_2$), 2.54–2.67 (m, 4-H, 6-H, 9-H), 2.77 (ddd, $J = 16.2$, 9.7, 7.2 Hz, 4-H), 2.98 (br, d, $J = 17.4$ Hz, CH_2COO), 3.16 (dd, $J = 13.5$, 9.4 Hz, 9H), 3.46–3.52 (m, $N(CH_2)_2$), 3.66–3.75 (m, 3-H, $N(CH_2)_2$), 3.92 (br, d, $J = 17.4$ Hz, CH_2COO), 4.03 (br, t, $J = 9.4$ Hz, 7-H, 8-H), 4.17 (d, $J = 2.7$ Hz, 1-H), 7.40–7.73 (m, aromatic H) ppm; ^{13}C NMR (100 MHz, CD_3OD): $\delta = 25.06$ ($(CH_2)_2$), 29.76 (C-9), 30.31 (C-4), 35.29 (C-6), 37.38 (C-8), 43.63 (C-7), 49.94, 50.20, 50.68 (C-3, $N(CH_2)_2$), 57.08 (CH_2COO) 64.82 (C-5), 72.08 (C-1), 128.77, 128.87, 129.60, 129.89, 130.76, 131.29, 138.44, 141.61 (aromatic C), 167.99 (COO) ppm; IR (KBr): $\bar{\nu} = 3417$, 2968, 2853, 2589, 2493, 1736, 1497, 1452, 1428, 1414, 1208, 759, 707 cm^{-1}; UV (CH_3OH): λ ($\log\varepsilon$) = 207 (4.176) nm.

*Preparation of Sulfonamides **8a–8d**, **9a**, and **9d***
Compounds **3a–3d** were dissolved in dry CH_2Cl_2, 4-*DMAP* and toluene-4-sulfonyl chloride or biphenyl-4-sulfonyl chloride were added. The solution was refluxed over night at 50°C, cooled to room temperature, diluted with CH_2Cl_2, and extracted 5 times with 2 *M* NaOH. The organic layer was washed 3 times with H_2O, dried (Na_2SO_4), filtered, and the solvent was evaporated *in vacuo*. The residue was extracted 3 times with hot *n*-heptane, the solvent was removed *in vacuo*, and the residue further purified.

(7RS,8RS)-(±)-5-Dimethylamino-7,8-diphenyl-2-(toluene-4-sulfonyl)-2-azabicyclo[3.2.2]nonane (**8a**, $C_{29}H_{34}N_2O_2S$)
In 10 cm^3 CH_2Cl_2 400 mg **3a** (1.25 mmol), 746 mg (2.5 mmol) toluene-4-sulfonyl chloride, and 305 mg (2.5 mmol) 4-*DMAP* gave a residue, which was purified by means of CC, using CH_2Cl_2:CH_3OH = 8:2 as eluent, giving 300 mg (51%) pure **8a**. Mp (*n*-heptane): 177°C; ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.91$–2.02 (m, 4-H, 6-H), 2.13 (dd, $J = 13.1$, 11.2 Hz, 9-H), 2.23 (ddd, $J = 13.8$, 8.8, 2.3 Hz, 9-H), 2.28–2.34 (m, 6-H, $N(CH_3)_2$), 2.36 (s, Ar-CH_3), 3.30–3.37 (m, 3-H, 7-H, 8-H), 3.80 (ddd, $J = 14.0$, 4.6, 4.4 Hz, 3-H), 4.28 (d, $J = 3.1$ Hz, 1-H), 7.06–7.42 (m, aromatic H) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 21.39$ (Ar-CH_3), 30.45 (C-4), 34.32 (C-9), 35.76 (C-6), 37.92 ($N(CH_3)_2$), 38.19 (C-8), 42.19 (C-3), 45.78 (C-7), 57.66 (C-5), 62.42 (C-1), 126.41, 126.78, 126.83, 127.51, 127.73, 128.36, 128.73, 129.39, 137.55, 141.78, 142.55, 143.42 (aromatic C) ppm; IR (KBr): $\bar{\nu} = 2946$, 2782, 1598, 1495, 1450, 1336, 1301, 1286, 1163, 1089, 1047, 960, 863, 814, 751, 702, 670 cm^{-1}; UV (CH_2Cl_2): λ ($\log\varepsilon$) = 235 (3.918) nm; MS (70 eV): m/z = 474 (M^+), 410, 319, 276, 237, 215, 186, 174, 144, 104, 91, 70; HRMS (EI+): calcd. ($C_{29}H_{34}N_2O_2S$): 474.23410; found: 474.23866.

(7RS,8RS)-(±)-5-Morpholino-7,8-diphenyl-2-(toluene-4-sulfonyl)-2-azabicyclo[3.2.2]nonane (**8b**, $C_{31}H_{36}N_2O_3S$)
In 10 cm^3 CH_2Cl_2 394 mg **3b** (1.1 mmol), 650 mg (3.4 mmol) toluene-4-sulfonyl chloride, and 265 mg (2.2 mmol) 4-*DMAP* gave a residue, which was purified by means of CC, using CH_2Cl_2:CH_3OH = 8:2 as eluent, giving 329 mg (58%) pure **8b** as a colourless resin. ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.88$ (br, t, $J = 11.8$, 6-H), 1.93–2.29 (m, 4-H, 6-H, 9-H), 2.36 (s, Ar-CH_3), 2.57–2.67 (m, $N(CH_2)_2$), 3.29–3.36 (m, 3-H, 7-H, 8-H), 3.71–3.72 (m, $O(CH_2)_2$), 3.82 (ddd, $J = 13.9$, 4.4, 3.9 Hz,

3-H), 4.31 (d, $J = 2.4$ Hz, 1-H), 7.06–7.42 (m, aromatic H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 21.40$ (Ar-CH_3), 31.63 (C-4), 34.27 (C-9), 35.59 (C-6), 38.27 (C-8), 42.20 (C-3), 45.65 (N(CH_2)$_2$), 45.72 (C-7), 57.63 (C-5), 62.28 (C-1), 67.54 (O(CH_2)$_2$), 126.45, 126.84, 127.44, 127.67, 128.40, 128.73, 129.40, 137.58, 141.73, 142.59, 143.42 (aromatic C) ppm; IR (KBr): $\bar{\nu} = 2954$, 2852, 1599, 1496, 1450, 1336, 1303, 1290, 1156, 1117, 1093, 1032, 926, 747, 699, 676, 668 cm^{-1}; UV (CH_2Cl_2): λ (logε) = 237 (3.930) nm; MS (70 eV): $m/z = 516$ (M^+), 362, 318, 257, 218, 171, 156, 145, 127, 104, 91, 77, 44; HRMS (EI+): calcd. ($C_{31}H_{36}N_2O_3S$): 516.24467; found: 516.23931.

(7RS,8RS)-(±)-7,8-Diphenyl-5-pyrrolidino-2-(toluene-4-sulfonyl)-2-azabicyclo[3.2.2]nonane (**8c**, $C_{31}H_{36}N_2O_2S$)
In 10 cm^3 CH_2Cl_2 392 mg **3c** (1.1 mmol), 675 mg (3.5 mmol) toluene-4-sulfonyl chloride, and 276 mg (2.3 mmol) 4-*DMAP* gave a residue, which was purified by means of CC, using CH_2Cl_2:CH_3OH = 8:2 as eluent, giving 393 mg (69%) pure **8c** as a colourless resin. ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.77$ (br, s, 2CH_2), 2.03–2.09 (m, 4-H, 6-H), 2.21–2.30 (m, 6-H, 9-H), 2.35 (s, Ar-CH_3), 2.66–2.78 (m, N(CH_2)$_2$), 3.32–3.40 (m, 3-H, 7-H, 8-H), 3.80 (ddd, $J = 13.7$, 4.7, 4.5 Hz, 3-H), 4.27 (d, $J = 3.0$ Hz, 1-H), 7.05–7.43 (m, aromatic H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 21.36$ (Ar-CH_3), 23.53 (2CH_2), 32.05 (C-4), 34.76 (C-9), 36.54 (C-6), 38.04 (C-8), 42.21 (C-3), 45.21 (N(CH_2)$_2$), 45.71 (C-7), 56.61 (C-5), 62.60 (C-1), 126.32, 126.70, 126.80, 127.53, 127.72, 128.31, 128.66, 129.35, 137.54, 141.80, 142.48, 143.56 (aromatic C) ppm; IR (KBr): $\bar{\nu} = 2956$, 2874, 1599, 1495, 1449, 1336, 1304, 1289, 1156, 1091, 1037, 864, 747, 699, 669 cm^{-1}; UV (CH_2Cl_2): λ (logε) = 231 (3.941) nm; MS (70 eV): $m/z = 500$ 500 (M^+), 345, 302, 255, 241, 212, 184, 156, 130, 104, 91; HRMS (EI+): calcd. ($C_{31}H_{36}N_2O_2S$): 500.24975; found: 500.24630.

(7RS,8RS)-(±)-7,8-Diphenyl-5-piperidino-2-(toluene-4-sulfonyl)-2-azabicyclo[3.2.2]nonane (**8d**, $C_{32}H_{38}N_2O_2S$)
In 10 cm^3 CH_2Cl_2 400 mg **3d** (1.1 mmol), 664 mg (3.5 mmol) toluene-4-sulfonyl chloride,c and 272 mg (2.2 mmol) 4-*DMAP* gave a residue, which was purified by means of CC, using CH_2Cl_2:CH_3OH = 8:2 as eluent, giving 403 mg (70%) pure **8d** as a colourless resin. ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.46$ (br, s, CH_2), 1.61 (br, s, 2CH_2), 1.80–2.34 (m, 4-H, 6-H, 9-H), 2.36 (s, Ar-CH_3), 2.50–2.65 (m, N(CH_2)$_2$), 3.26–3.35 (m, 3-H, 7-H, 8-H), 3.84 (ddd, $J = 13.9$, 4.3, 4.1 Hz, 3-H), 4.30 (d, $J = 3.1$ Hz, 1-H), 7.05–7.42 (m, aromatic H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 21.40$ (Ar-CH_3), 24.79 (CH_2), 26.56 (2CH_2), 31.65 (C-4), 34.20 (C-9), 35.68 (C-6), 38.49 (C-8), 42.34 (C-3), 45.95 (C-7), 46.31 (N(CH_2)$_2$), 58.21 (C-5), 62.23 (C-1), 126.39, 126.73, 126.78, 127.46, 127.65, 128.35, 128.68, 129.39, 137.60, 141.80, 142.53, 143.46 (aromatic C) ppm; IR (KBr): $\bar{\nu} = 2930$, 1599, 1495, 1450, 1336, 1304, 1286, 1157, 1091, 1034, 971, 922, 866, 813, 747, 699, 672 cm^{-1}; UV (CH_2Cl_2): λ (logε) = 232 (3.928) nm; MS (70 eV): $m/z = 514$ (M^+), 359, 316, 275, 255, 226, 171, 125, 104, 91; HRMS (EI+): calcd. ($C_{32}H_{38}N_2O_2S$): 514.26540; found: 514.26820.

(7RS,8RS)-(±)-2-(Biphenyl-4-sulfonyl)-5-dimethylamino-7,8-diphenyl-2-azabicyclo[3.2.2]nonane (**9a**, $C_{34}H_{36}N_2O_2S$)
In 10 cm^3 CH_2Cl_2 450 mg **3a** (1.4 mmol), 710 mg (2.8 mmol) biphenyl-4-sulfonyl chloride, and 342 mg (2.8 mmol) 4-*DMAP* gave a residue, which was purified by means of CC, using CH_2Cl_2:CH_3OH = 9:1 as eluent, giving 350 mg (47%) pure **9a** as a colourless resin. ^{1}H NMR (400 MHz, $CDCl_3$): $\delta =$ 1.92–2.03 (m, 4-H, 6-H), 2.11 (dd, $J = 13.4$, 11.1 Hz, 9-H), 2.25 (ddd, $J = 13.7$, 11.1, 2.3 Hz, 9-H), 2.31 (br, s, N(CH_3)$_2$), 2.34 (ddd, $J = 13.4$, 11.1, 2.3 Hz, 6-H), 3.33–3.42 (m, 3-H, 7-H, 8-H), 3.95 (ddd, $J = 13.7$, 4.6, 4.3 Hz, 3-H), 4.30 (d, $J = 3.3$ Hz, 1-H), 7.08–7.57 (m, aromatic H) ppm; ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 30.43$ (C-4), 33.99 (C-9), 35.89 (C-6), 37.95 (N(CH_3)$_2$), 38.37 (C-8), 42.36 (C-3), 45.29 (C-7), 57.54 (C-5), 62.84 (C-1), 126.48, 126.83, 127.17, 127.19, 127.38, 127.51, 127.70, 128.29, 128.40, 128.77, 128.98, 139.17, 139.43, 141.67, 143.43, 144.63 (aromatic C) ppm; IR (KBr): $\bar{\nu} = 2947$, 1596, 1496, 1480, 1449, 1336, 1156, 1092, 1046, 971, 866, 763, 746, 698, 675 cm^{-1}; UV (CH_2Cl_2): λ (logε) = 269 (4.347) nm; MS (70 eV): $m/z = 536$ (M^+), 320, 275, 186, 154, 128, 104, 91, 69, 51; HRMS (EI+): calcd. ($C_{34}H_{36}N_2O_2S$): 536.24975; found: 536.25112.

(7RS,8RS)-(±)-2-(Biphenyl-4-sulfonyl)-7,8-diphenyl-5-piperidino-2-azabicyclo[3.2.2]nonane (**9d**, $C_{37}H_{40}N_2O_2S$)
In 10 cm^3 of CH_2Cl_2 450 mg **3d** (1.25 mmol), 632 mg (2.5 mmol) biphenyl-4-sulfonyl chloride, and 305 mg (2.5 mmol) 4-*DMAP* gave a residue, which was purified by means of CC, using CH_2Cl_2:CH_3OH = 9:1 as eluent, giving 270 mg (37%) pure **9d** as a colourless resin. ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 1.46$ (br, s, CH_2), 1.60 (br, s, 2CH_2), 1.85–2.20 (m, 4-H, 6-H, 9-H), 2.25 (br, dd, $J = 12.9$, 9.7 Hz, 9-H), 2.33 (br, t, $J = 11.0$ Hz, 6-H), 2.46–2.70 (m, N(CH_2)$_2$), 3.31–3.40 (m, 3-H, 7-H, 8-H), 3.97 (ddd, $J = 13.9$, 4.2, 4.0 Hz, 3-H), 4.32 (d, $J = 3.0$ Hz, 1-H), 7.08–7.57 (m, aromatic H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 24.90$ (CH_2), 26.66 (2CH_2), 31.68 (C-4), 33.91 (C-9), 35.80 (C-6), 38.70 (C-8), 42.53 (C-3), 46.32 (N(CH_2)$_2$), 46.46 (C-7), 58.08 (C-5), 62.67 (C-1), 126.45, 126.77, 127.16, 127.37, 127.45, 127.65, 128.27, 128.39, 128.72, 128.98, 139.28, 139.44, 141.74, 143.52, 144.58 (aromatic C) ppm; IR (KBr): $\bar{\nu} = 2930$, 1597, 1496, 1480, 1449, 1337, 1158, 1093, 1035, 971, 921, 866, 762, 745, 697, 675 cm^{-1}; UV (CH_2Cl_2): λ (logε) = 269 (4.345) nm; MS (70 eV): $m/z = 576$ (M^+), 493, 360, 315, 275, 202, 186, 170, 154, 128, 104, 91, 69, 51; HRMS (EI+): calcd. ($C_{37}H_{40}N_2O_2S$): 576.28105; found: 576.27586.

Antiprotozoal Tests, Cytotoxicity

Plasmodium falciparum

Antiplasmodial activity was examined using the K1 strain of *P. falciparum* (resistant to chloroquine and pyrimethamine). Viability is determined by the incorporation of [^{3}H]-hypoxanthine into living protozoal cells by a modification of a reported assay [4]. Infected human red blood cells in RPMI 1640 medium with 5% Albumax were exposed to serial drug dilutions ranging from 5 to 0.078 μg/cm^3 in microtiter plates. After 48 h of incubation at 37°C in a reduced oxygen atmo-

sphere, 0.5 μCi ^{3}H-hypoxanthine were added to each well. Cultures were incubated for a further 24 h before they were harvested onto glass-fiber filters and washed with distilled H_2O. The radioactivity was counted using a Betaplate^{TM} liquid scintillation counter (Wallac, Zurich, Switzerland). The results were recorded as counts per minute (CPM) per well at each drug concentration and expressed as percentage of the untreated controls. From the sigmoidal inhibition curves IC_{50} values were calculated. Assays were run in duplicate and repeated once. Chloroquine was used as the standard.

Trypanosoma b. rhodesiense, Cytotoxicity

Minimum Essential Medium (50 mm^3) supplemented according to a known procedure [5] with 2-mercaptoethanol and 15% heat-inactivated horse serum was added to each well of a 96-well microtiter plate. Serial drug dilutions were prepared covering a range from 90 to 0.123 $\mu g/cm^3$. Then 10^4 bloodstream forms of *Trypanosoma b. rhodesiense* STIB 900 in 50 mm^3 were added to each well and the plate incubated at 37°C under a 5% CO_2 atmosphere for 72 h. 10 mm^3 Alamar Blue (containing 12.5 mg resazurin dissolved in 1 dm^3 distilled H_2O) were then added to each well and incubation continued for a further 2–4 h. The Alamar blue dye is an indicator of cellular growth and/or viability. The blue, non fluorescent, oxidized form becomes pink and fluorescent upon reduction by living cells. The plate was then read in a Spectramax Gemini XS microplate fluorometer (Molecular Devices Cooperation, Sunnyvale, CA, USA) using an excitation wavelength of 536 nm and emission wavelength of 588 nm [6]. Fluorescence development was measured and expressed as percentage of the control. Data were transferred into the graphic programme Softmax Pro (Molecular Devices) which calculated IC_{50} values. Suramine served as the standard. Cytotoxicity was assessed using the same assay and rat skeletal myoblasts (L-6 cells) with mefloquine as standard.

Medium Throughput Screening

In the medium throughput screening (MTS) the same methods were used in principle as for the IC_{50} value determination. In the MTS only two drug concentrations were tested instead of a serial drug dilution. The results were expressed as percentage of growth inhibition.

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