In vitro antimalarial activity of inhibitors of human Rac1: a possible new way to fight Plasmodium?

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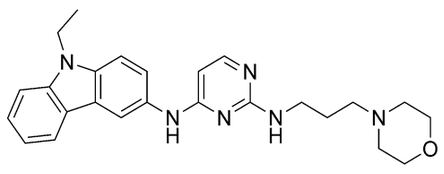
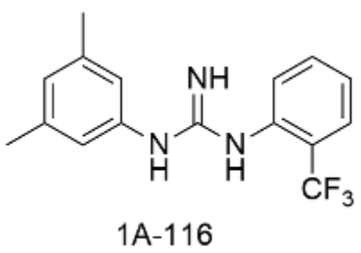
INTRODUCTION

Plasmodium parasites developed resistance for all the classes of known antimalarial drugs. Thus, the search for antimalarial drugs with novel mechanisms of action is compelling. The **human GTPase Rac1** plays a role in parasite invasion of the host cells in many parasites. Also in *Plasmodium falciparum,* it was suggested an involvement of Rac1 both during the invasion process and parasite intracellular development. **The aim of the present work was to test a series of Rac1 inhibitors for potential antimalarial activity.**

MATERIALS AND METHODS





- ANTIMALARIAL ACTIVITY
  - pLDH assay on *P. falciparum* asexual parasites (D10, W2)
  - Luciferase assay on *P. falciparum* gametocytes (3D7elo1CBG99 strain)
  - Association assays on *P. falciparum* asexual parasites (pLDH assay)
- PARASITE INVASION → FACS analysis of fluorescent labelled parasites
- PARASITE INTRAERYTHROCYTIC DEVELOPMENT → Microscopic observation of Giemsa stained smears
- CITOTOXICITY → MTT assay (OD550/650) on human microvascular endothelial cells HMEC-1

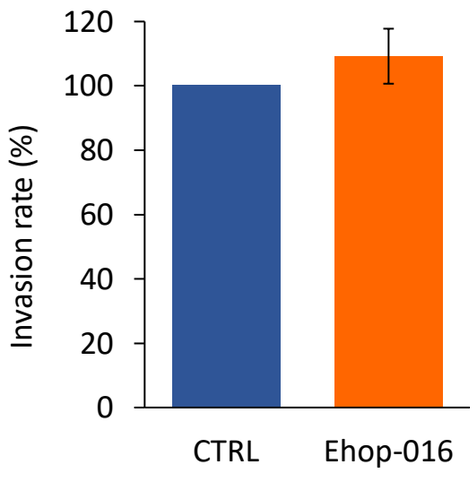
RESULTS

Fourteen commercially available* or newly synthetized Rac1 inhibitors were tested for their potential antimalarial activity. Among these, three inhibitors had an IC_{50} lower than 1 µM, only two were inactive (IC_{50}> 50 µM), whereas the remaining showed intermediate IC50s. **EHop-016 was the most effective compound**

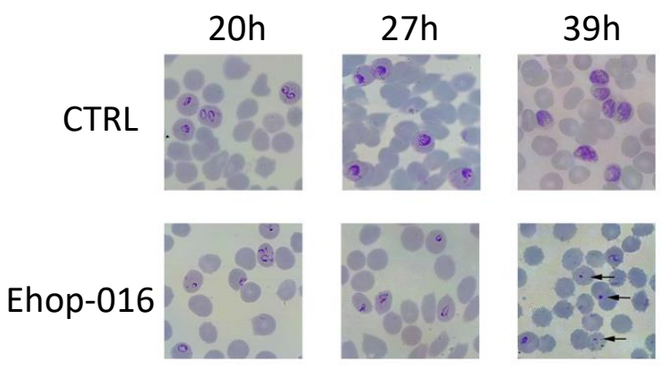
Antimalarial activity and citotoxicity of six selected Rac1 inhibitors

	D10 IC_{50} (µM)	W2 IC_{50} (µM)	3D7 immature gametocytes IC_{50} (µM)	3D7 mature gametocytes IC_{50} (µM)	HMEC-1 IC_{50} (µM)	SI
EHT-1864 *	3.88 ± 0.74	7.75 ± 1.82	>50.00	32.59 ± 0.92	48.47 ± 1.23	12.5
1A116 *	3.21 ± 1.17	6.89 ± 1.41	78.13 ± 17.68	56.64 ± 21.34	131.50 ± 37.26	41.1
EHop-016 *	**0.14 ± 0.02**	**0.32 ± 0.03**	**0.74 ± 0.20**	**4.65 ± 0.52**	**5.22 ± 1.07**	**37.8**
Aza-1 *	0.25 ± 0.03	0.76 ± 0.27	1.25 ± 0.48	7.23 ± 2.12	12.32 ± 6.11	48.9
Compound 3	0.66 ± 0.05	0.52 ± 0.02	2.26 ± 0.22	21.85 ± 2.71	10.59 ± 1.19	16.1
Compound 3'	2.76 ± 0.12	1.24 ± 0.13	2.13 ± 0.43	26.58 ± 3.70	16.00 ± 0.44	5.8
MB			0.03 ± 0.01	0.12 ± 0.03		
Chloroquine	0.02 ± 0.003	0.42 ± 0.08				

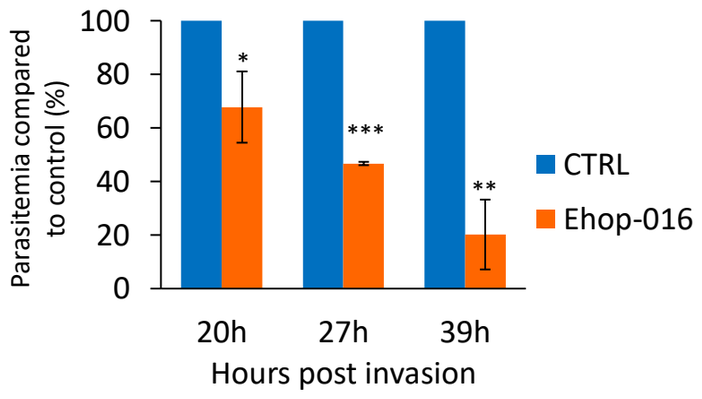
IC_{50}= concentration of drugs inducing 50% of growth inhibition; SI= Selectivity Index, calculated as the ratio between IC_{50} against HMEC-1 and IC_{50} against asexual parasites of the D10 strain



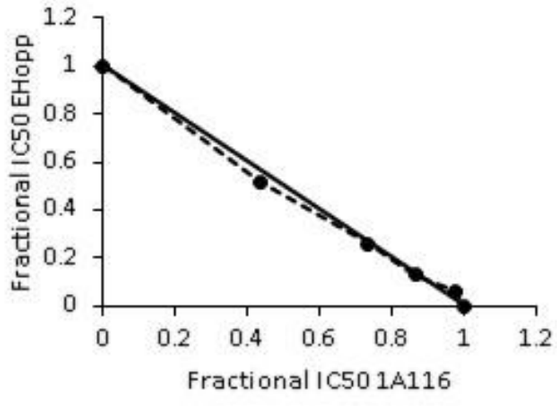
EHop-016 at the dose of 2.5 µM did not reduce RBC invasion efficiency



None of the treated parasites developed into the trophozoite stage



EHop-016 at 1.4 µM caused a significant, time-dependent reduction in parasitaemia



EHop-016 had additive effect with 1A116 (isobologram analysis)

CONCLUSIONS

EHop-016 showed a promising activity that raises attention on this class of molecules as potential antimalarials and deserves further investigation.

ACKNOWLEDGMENTS

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