TRITERPENOIDS AS INHIBITORS OF PLASMODIUM LIVER-STAGE DEVELOPMENT

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INTRODUCTION

Malaria is one of the foremost public health problems in Africa. It is endemic in 90 countries, affecting nearly 40% of the global population. The increasing prevalence of drug-resistant *Plasmodium falciparum* strains is one of the greatest challenges in malaria control. In order to overcome drug-resistance, new antimalarial drugs are urgently needed. Most of the available antimalarial agents kill blood stage parasites and only a limited number of drugs act on liver stages. In fact, the study of *Plasmodium* liver stage development (Fig. 1) has been hampered by limitations in the experimental approaches required to quantify hepatocyte infection by the parasite.

Therefore, the development of new drugs targeting *Plasmodium* liver stages represents an important and under-exploited site of intervention [1, 2].

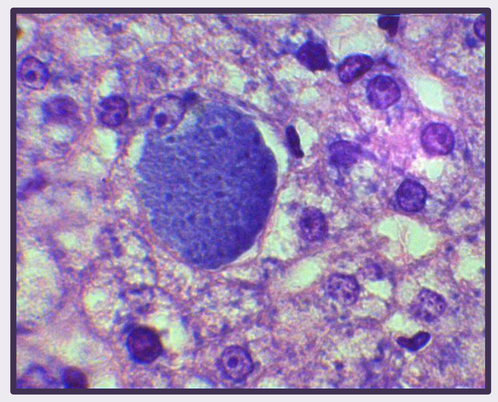

Figure 1. *Plasmodium* spp. liver stage. .

Figure 2. *Momordica balsamina.*

RESULTS AND DISCUSSION

Previously, bioassay-guided fractionation of the methanol extract of the aerial parts of *Momordica balsamina* led to the isolation of several cucurbitane-type triterpenoids. Many of those compounds and acylated derivatives displayed *in vitro* antimalarial activity against blood schizonts of chloroquine-sensitive and -resistant strains of *Plasmodium falciparum* [3-5].

In this study, compounds **1** – **5** (Fig. 3) were evaluated for their *in vitro* activity against liver stages of the rodent malaria parasite *P. berghei*, using a recently described bioluminescence imaging method [1]. This method uses a transgenic *P. berghei* parasite, PbGFP-Luc$_{con}$, expressing the bioluminescent reporter protein luciferase to visualize and quantify parasite development in Huh-7 cells, a human hepatoma cell line (Fig.3 – bars). Compound toxicity was also assessed on the same cell line through the fluorescence measurement of cell confluency (Fig. 3 – line)

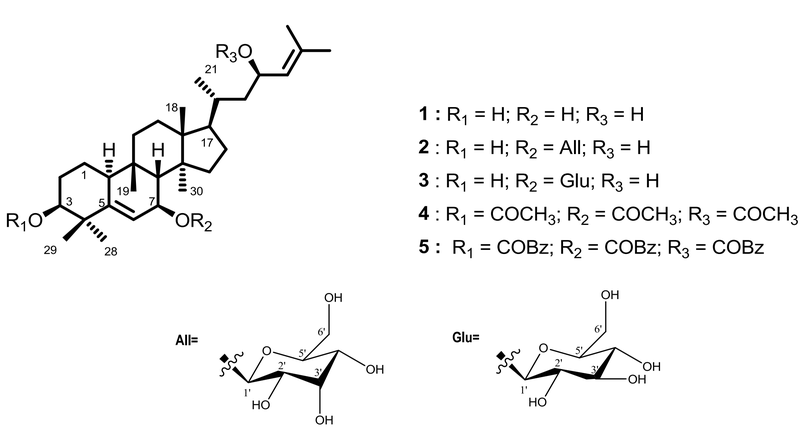

Figure 3. Chemical structures of the isolated compounds (**1 -5**)

Compounds **1** - **4** exhibited activity against *P. berghei* liver stages *in vitro* (Fig. 4). Balsaminol F (**1**) and, in particular, Balsaminoside B (**2**) displayed significant toxicity against Huh-7 cells at 15 µM.

Triacetylbalsaminol F (**5**) showed the most potent inhibitory activity against the liver stages of *Plasmodium*, with no detectable toxicity towards the Huh-7 cells, at the concentrations employed.

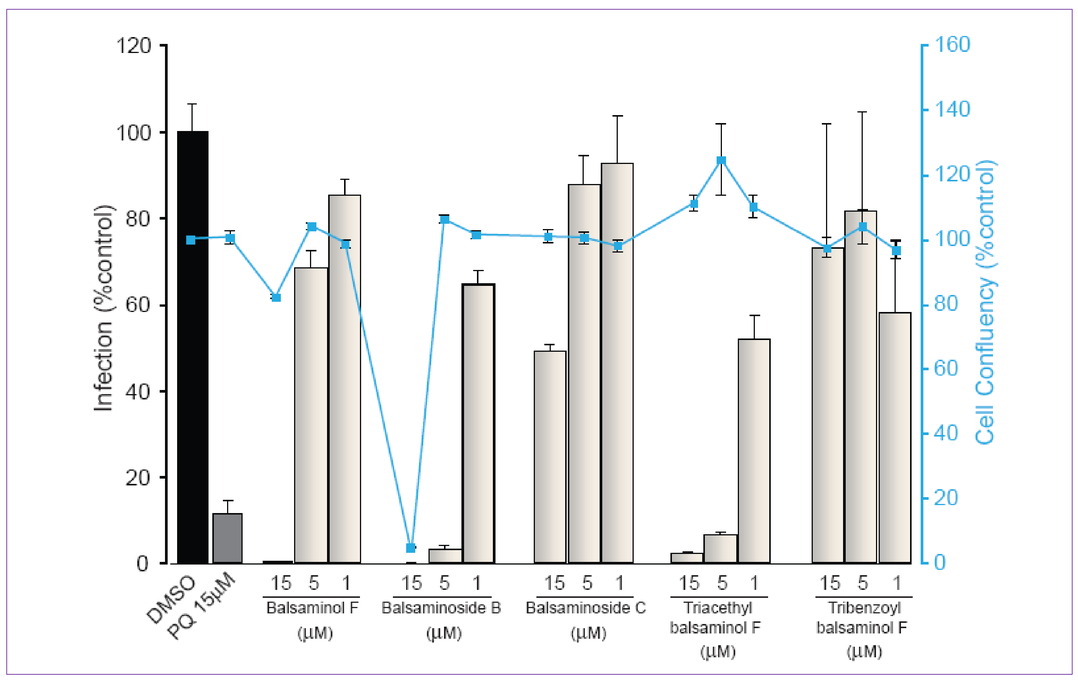

Figure 4. Drug inhibition of liver stage infection, determined by measurement of luciferase activity (bars), and compound toxicity, assessed by fluorescence measurement of cell confluency (line), in *Pb*GFP-Luc$_{con}$-infected Huh-7. PQ- primaquine, used as positive control. DMSO- solvent-treated control. Error bars represent the standard deviations of three independent measurements.

CONCLUSION

Triacetylbalsaminol F (4) displayed higher *in vitro* efficacy than primaquine against *P. berghei* liver forms, warranting further exploitation of its mechanism of action.

REFERENCES

1. Ploemen et al (2009) PloS One 4: e7881; 2. Prudêncio et al (2008) Cell Microbiol. 10: 218–24; 3. Ramalhete et al (2009) Bioorg. Med. Chem. 17: 6942-51; 4. Ramalhete et al (2010) Bioorg. Med. Chem. 18, 5254-60; 5. Ramalhete et al (2011) Bioorg. Med. Chem. 19, 330-8.