Protein farnesyl transferase inhibitors for the treatment of malaria and African trypanosomiasis

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Protein farnesyl transferase inhibitors (PFTIs) have been developed as oncology therapeutics but recent studies have supported the use of PFTIs for the treatment of eukaryotic pathogens. Data supporting PFTIs for the treatment of African sleeping sickness caused by Trypanosoma brucei sp, and for the therapy of malaria caused by Plasmodium spp is reviewed. Protein prenylation in T brucei and P falciparum has been studied using a variety of techniques, including recombinant and native enzyme assays. Studies have demonstrated farnesylation and geranylgeranylation in these parasites. A variety of PFTIs have shown growth inhibition activity and killing of T brucei and P falciparum, yet not all mammalian PFTIs are active on parasitic PFTs. Protein farnesyl transferase as well as protein geranylgeranyl transferase type II enzymatic activities have been demonstrated in T brucei and P falciparum, but protein geranylgeranyl transferase type I activity may be lacking from these parasites, perhaps explaining the extreme sensitivity of these organisms to PFTIs compared with mammalian cells. Given that PFTIs are relatively non-toxic in short-term administration to humans, PFTIs specific to parasites are not required for therapy. Thus, the challenge in PFTI drug development is not to identify selective antiparasite compounds, but to identify compounds with sufficient potency and pharmacokinetic properties to produce satisfactory drugs for malaria and African sleeping sickness.

Keywords Antiprotozoal drugs, malaria, *Plasmodium*, protein farnesyl transferase, *Trypanosoma*, Trypanosomiasis

Introduction

Malaria and African trypanosomiasis have a significant impact on human life in the tropics and subtropics. Malaria infection resulted in approximately 1,124,000 deaths in 2002, with the majority occurring in children in sub-Saharan Africa [1]. African trypanosomiasis (also known as African sleeping sickness) is re-emerging in a dramatic manner, with an estimated 50,000 deaths in 2002 [2]. Neither disease has a vaccine, nor is one likely to be available for widespread use for many years. Efforts to limit the diseases by controlling the respective arthropod vectors have been thwarted by scarce resources, resistant mosquitoes (in the case of malaria), and the overwhelming scale of the problem. Simple measures such as insecticide impregnated bed-nets have been shown to dramatically decrease rates of malaria transmission and need to be more widely used. However, there are high rates of human infections and good chemotherapeutics will be required to treat new cases of malaria and African trypanosomiasis for the indefinite future. The drug armamentarium for malaria has been severely diminished by the development of widespread resistance to standard drugs. The drugs for African sleeping sickness have always been deficient in terms of safety, efficacy and ease of administration. This review will discuss the efforts to develop a new class of antiprotozoan compounds, protein farnesyl transferase inhibitors, which will hopefully help fill the gap for much needed therapeutics for these tropical diseases.

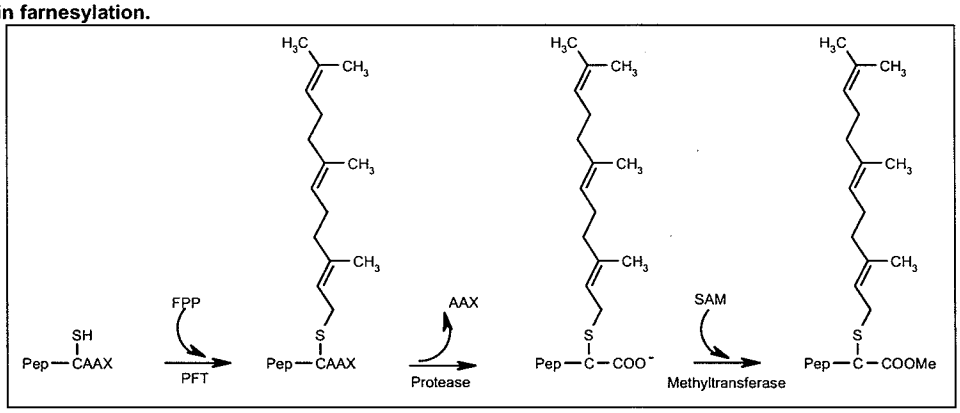
Protein prenylation in eukaryotic cells

Protein prenylation refers to the covalent attachment of isoprenoid groups, specifically 15-carbon farnesyl or 20-carbon geranylgeranyl, to cellular proteins. The structure of the prenyl groups attached to proteins in mammalian cells was determined by Glomset and co-workers in 1990 [3]. The addition of a prenyl moiety to the C-terminus of certain proteins creates a hydrophobic tail that facilitates membrane associations and certain protein-protein interactions. Examples of prenylated proteins include small GTPases such as Ras, Rab, Rac and Rho that play a role in cellular signal transduction, cell-cycle progression and intracellular vesicle trafficking [4]. Other examples of prenylated proteins include the γ-subunits of heterotrimeric G proteins, nuclear lamins, several proteins in the phototransduction cascade, and an antigen of the hepatitis δ virus [5].

Protein prenylation is mediated by three distinct enzymes: protein farnesyl transferase (PFT), protein geranylgeranyl transferase (PGGT)-I and PGGT-II. PFT recognizes the so-called CAAX motif (CAAX denotes a cysteine followed by two amino acids (usually aliphatic), and an amino acid 'X' that is usually one of Ser, Met, Ala or Gln [5]) at the terminus of specific proteins and attaches farnesyl from farnesyl pyrophosphate to the thiol group of the cysteine (Figure 1). PFT is a zinc-dependent enzyme with α- and β-subunits. PGGT-I shares the same α-subunit as PFT, but has a distinct β-subunit. PGGT-I attaches geranylgeranyl to proteins with the CAAX motif in which X = Leu or Phe. For both PFT and PGGT-I, other possible X residues may be tolerated. Following the action of PFT or PGGT-I, a protease cleaves the terminal 'AAX' tripeptide from the newly prenylated protein (Figure 1). A final processing step involves the methylation of the terminal carboxylic acid by the prenyl protein methyltransferase enzyme (Figure 1). A third prenylation enzyme, PGGT-II, attaches two geranylgeranyl groups onto terminal cysteine residues of proteins ending with CC, CCXX or CXC amino acid motifs. To date, these digeranylgeranylated proteins have been found exclusively in the Rab family of low-molecular weight G proteins [5].

Protein prenylation has received significant attention due to the discovery that the Ras oncogene is farnesylated and that

Figure 1. Protein farnesylation.



The first step involves the addition of a farnesyl group to the cysteine located at the C(4)-position from the carboxy-terminus of substrate proteins. Next, the prenyl-protein protease removes the three terminal amino acids. Finally, the exposed farnesylcysteine carboxylic acid is converted to a methyl ester. **CAAX** Cys, two amino acids (usually aliphatic) and an amino acid 'X' that is usually one of Ser, Met, Ala or Gln; **FPP** farnesyl pyrophosphate, **Pep** peptide, **PFT** protein farnesyl transferase, **SAM** S-adenosyl-methionine.

Ras function depends on post-translational farnesylation [6]. Mutations in Ras are associated with 20 to 25% of human tumors, and specifically with 90% of pancreatic carcinomas [7•]. As many as 30 pharmaceutical companies have initiated drug discovery programs to generate PFT inhibitors (PFTIs) for the treatment of cancer. Some of these compounds are well advanced in clinical trials for various malignancies [7•]. Extensive research into the biochemistry, cell biology and inhibitor development of PFT provides an excellent opportunity to explore the potential of PFTIs as agents for treating neglected diseases such as malaria and African sleeping sickness.

Protein prenylation in *Plasmodium* and trypanosomatid parasites

Protein prenylation occurs in a variety of pathogenic protozoa, including *Trypanosoma brucei* [8], *Trypanosoma cruzi* [9], *Leishmania* species [9], *Plasmodium falciparum* [10•], *Toxoplasma gondii* [11], *Giardia lamblia* [12] and *Entamoeba histolytica* [13]. Our research group has worked extensively on the PFT enzyme of trypanosomatid parasites [9,14•]. PFT enzyme activity was initially detected in cytosolic fractions of *T brucei* using the substrate, yeast Ras1 protein containing the C-terminal sequence Cys-Val-Ile-Met [8]. The *T brucei* PFT enzyme was subsequently purified by affinity methods using the CAAX peptide, Ser-Ser-Cys-Ala-Leu-Met [15•]. As with PFTs of mammalian cells, *T brucei* PFT is a heterodimer. The subunits are larger in the *T brucei* PFT compared with the yeast and mammalian homologs. This is a result of numerous insertions that occur at a distance from the active site as predicted by molecular modeling using the known mammalian PFT structure [9,14•]. Similar insertions are observed in *T cruzi* and *Leishmania* PFTs [9]. The function of these insertions is unknown. The substrate specificity, as it pertains to the CAAX motif, is different in the trypanosomatid PFTs compared with mammalian PFT, with more restricted preferences to peptides with Met or Gln in the X position. Differences in four residues in contact with the substrate in the active site of *T brucei* PFT help account for the differences in specificity and suggest the potential for selective inhibition of the parasite enzyme over the mammalian enzyme [14•].

The native PFT of *P falciparum* was characterized by Chakrabarti *et al* [16•]. It was partially purified from *in vitro* cultured *P falciparum* using $(NH_4)_2SO_4$ precipitation and anion exchange chromatography. As with trypanosomatid PFTs, *P falciparum* PFT acts preferentially on CAAX peptides with Met or Gln in the X position, with X = Met being the best substrate. Radiolabeled farnesol, presumably after conversion to farnesyl pyrophosphate by parasite enzymes, was incorporated into 50- and 22- to 28-kDa proteins, but much of the label in the 22- to 28-kDa proteins was geranylgeranyl and the 50-kDa proteins were found to be exclusively farnesylated [16•]. Incorporation of radiolabeled prenylation precursors was greatest in the trophozoite to schizont and schizont to ring transition states but was almost absent in the ring to trophozoite stage. Inhibition of cell growth by FTI-276 (University of Pittsburgh) or GGTI-298 at 5 µM was accompanied by a general reduction of prenylated proteins [14•]. Interestingly the farnesylated 50-kDa protein incorporation was not preferentially affected, as expected with FTI-276 which should be a PFT-specific inhibitor. This could be due to off-target effects on cell proliferation and secondary reduction in protein prenylation.

The existence of PGGT-I enzyme in *T brucei* and/or *P falciparum* is an area of debate. In early studies, Yokoyama *et al* analyzed cytosolic fractions of *T brucei* for prenylation activity and suggested that the presence of PGGT-I activity was separate from PFT activity [8]. However, the activity was weak and could possibly have resulted from non-specific associations of the [^{3}H]geranylgeranyl to the H-Ras-Cys-Val-Leu-Leu substrate used in the experiments. Subsequently, the genome of *T brucei* has been fully sequenced (and partially annotated), and a definite ortholog to the PGGT-I β-subunit cannot be identified by basic local alignment search tool (BLAST). In contrast, the *T brucei* orthologs to the PFT β-subunit and the PGGT-II β-subunits are readily identifiable by BLAST search of the GeneDB website [17] with yeast and mammalian PFT and PGGT-II sequences. The apparent absence of PGGT-I from *T brucei*

suggests an explanation for the relative toxicity of PFT inhibitors to *T brucei* cells compared with the effects of PFT inhibitors in mammalian cells. It may be that PGGT-I partially fulfills the function of PFT in mammalian cells when they are treated with PFT inhibitors and thus rescues the mammalian cells from the toxic effects of the inhibitors. In contrast, the lack of redundancy in prenylation enzymes of *T brucei* may render these cells exquisitely sensitive to PFT inhibitors. Of note, the *T cruzi* genome has a probable homolog to PGGT-I β-subunit [17]. Data from *T cruzi* cultures indicates that this organism is relatively insensitive to PFT inhibitors, which is consistent with the above idea.

Chakrabarti *et al* demonstrated PGGT-I activity from purified fractions of *P falciparum* lysates [10•]. However, it is not possible to definitely infer the existence of the putative PGGT-I β-subunit gene from the completed *P falciparum* genome. The genome contains three genes that are orthologous to protein prenyltransferase β-subunits. One is very likely the PFT β-subunit [18] based on conserved residues in the putative active site compared with the human PFT β-subunit. The two remaining β-subunit genes (MAL6P1.28 and PFL0695c) each share approximately 20 to 30% sequence identity with PGGT-I and to PGGT-II from other species. These genes have not been cloned or biochemically characterized, and therefore it is not possible to assign specific functions to them.

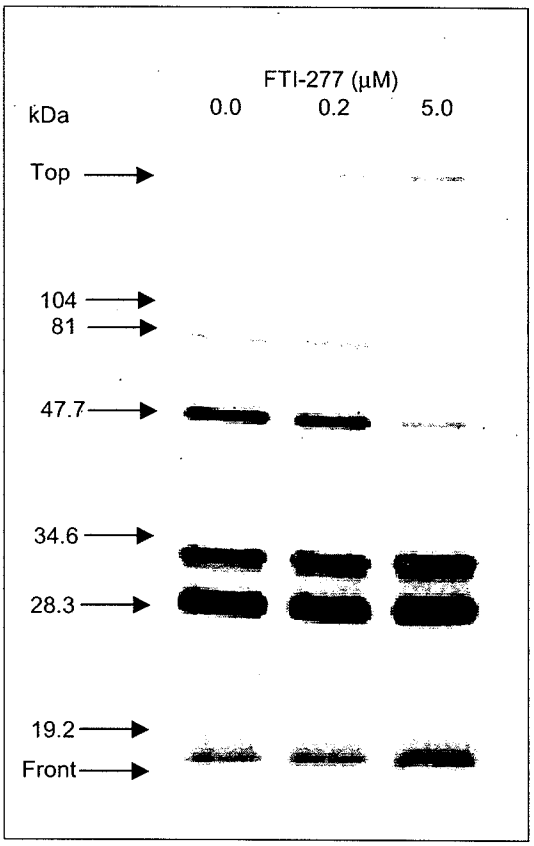
Both *T brucei* and *P falciparum* appear to have PGGT-II enzyme activity. Cells of these species that were grown with [^{3}H]mevalonate or [^{3}H]farnesol in the presence of PFTIs demonstrate intensely radiolabeled proteins, particularly in the 20- to 28-kDa molecular-weight range, despite the blockage of specific farnesylated proteins (Figure 2 shows labeling of *T brucei* cells with intense bands around 28- and 32-kDa). The low MW proteins are consistent with geranylgeranylated proteins, probably Rab proteins that are typically PGGT-II substrates.

Based on a bioinformatic approach, a list of putative prenylated proteins for *T brucei* and *P falciparum* has been compiled. A subset is listed in Table 1. For *T brucei*, a search of the annotated genome was conducted for proteins with a cysteine at the -4 position from the C-termini. The proteins that might be prenylated were predicted from this based on the mammalian PFT crystal data and *in vitro* enzyme data [9,14•,19]. This same method was used to predict prenylated proteins for *P falciparum*; however, due to difficulty in predicting intron/exon junctions, very few credible predictions could be identified. An alternative approach involved BLAST searches against the *P falciparum* annotated proteins [18] using known or suspected prenylated proteins. The genome was then searched for a putative CAAX box upstream or downstream of the annotated stop.

PFTIs

The structures of several PFTIs are shown in Figure 3. Most PFT inhibitors operate as peptidomimetics binding to the enzyme in the CAAX binding pocket. FTI-276 is an obvious mimetic of the CVFM tetrapeptide (Figure 3). Hamilton's research group at Yale University replaced the cysteine with an imidazole to give the inhibitor more drug-like properties [20]. The lone electron pair on the imidazole nitrogen is assumed to coordinate with the zinc atom in the catalytic site of the enzyme. This type of interaction has been shown to occur with other imidazole-containing PFT inhibitors and PFT enzyme [21•]. As seen in Figure 3, most of the compounds contain an imidazole, which acts as the metal coordinating group that binds the catalytic zinc atom. Exceptions are Sch-66336 (Schering-Plough Research Institute; Figure 3) and the benzophenone-containing compound (Figure 3) [22]. The structure of PFT bound to Sch-66336 demonstrates that this compound blocks the opening to the catalytic site [W Windsor, personal communication]. The benzophenone inhibitor is predicted to bind deep in the catalytic pocket of PFT based on docking experiments using the rat PFT enzyme structure [22].

Figure 2. Inhibition of protein farnesylation by FTI-277 in *T brucei*.



T brucei from the bloodstream was cultured in the presence of [^{3}H]mevalonolactone (which is hydrolyzed in cells to mevalonic acid and subsequently incorporated into farnesylpyrophosphate and geranylgeranyl pyrophosphate). Proteins were resolved by sodium dodecyl sulfate-polyacrylamide gel electrophoresis and the gel subjected to fluorography. The cells were grown in varying concentrations of FTI-277. The arrows mark bands that are thought to represent farnesylated proteins.

(Reproduced with permission from The American Society for Biochemisty and Molecular Biology Inc and Yokoyama K, Trobridge P, Buckner FS, Van Voorhis WC, Stuart KD, Gelb MH: ***Protein farnesyltransferase from Trypanosoma brucei. A heterodimer of 61- and 65-kDa subunits as a new target for antiparasite therapeutics.*** *J Biol Chem (1998)* ***273****(41):26497-26505. © 1998 The American Society for Biochemisty and Molecular Biology Inc.)*

Table 1. Predicted prenylated proteins in *T brucei* and *P falciparum*.

Identification	Protein	CAAX motif
Trypanosoma brucei		
Tb10.70.5440	DnaJ	Cys-Thr-Gln-Gln
Tb10.70.4100	Protein kinase	Cys-Ile-Ile-Met
Tb10.70.0590	GTP-binding protein	Cys-Val-Ile-Met
Tb08.11J15.1190	Protein tyrosine phosphatase	Cys-Ala-Ile-Met
Tb10.389.1800	Vesicle-associated membrane protein	Cys-Thr-Leu-Met
Tb927.2.5160/30J2.30	Chaperone protein DnaJ	Cys-Thr-Gln-Gln
Tb11.01.2530	Kinesin-like protein	Cys-Val-Thr-Met
Tb11.02.1380	TRYPARP actin-like protein	Cys-Lys-Cys-Gln
Plasmodium falciparum		
PFL2250c	rac-βserine/threonine protein kinase	Cys-Ser-Gly-Phe*
PF11_0183	GTP-binding protein	Cys-Cys-Leu-Leu*
PF13_0119	GTP-binding protein	Cys-Ile-Phe-Phe*
PFL1500w	GTP-binding protein	Cys-Asn-Phe-Ile*
PF11_0139	Protein tyrosine phosphatase	Cys-His-Phe-Met
PF11_0197	Protein phosphatase 1 regulatory subunit-like	Cys-Leu-Asn-Lys*
PFI0515w	Synaptobrevin-like protein	Cys-Ser-Ile-Met

*Proteins containing a putative CAAX box upstream or downstream of the annotated stop.

Figure 3. The structures of the CAAX peptide (CVFM) and selected PFTIs.

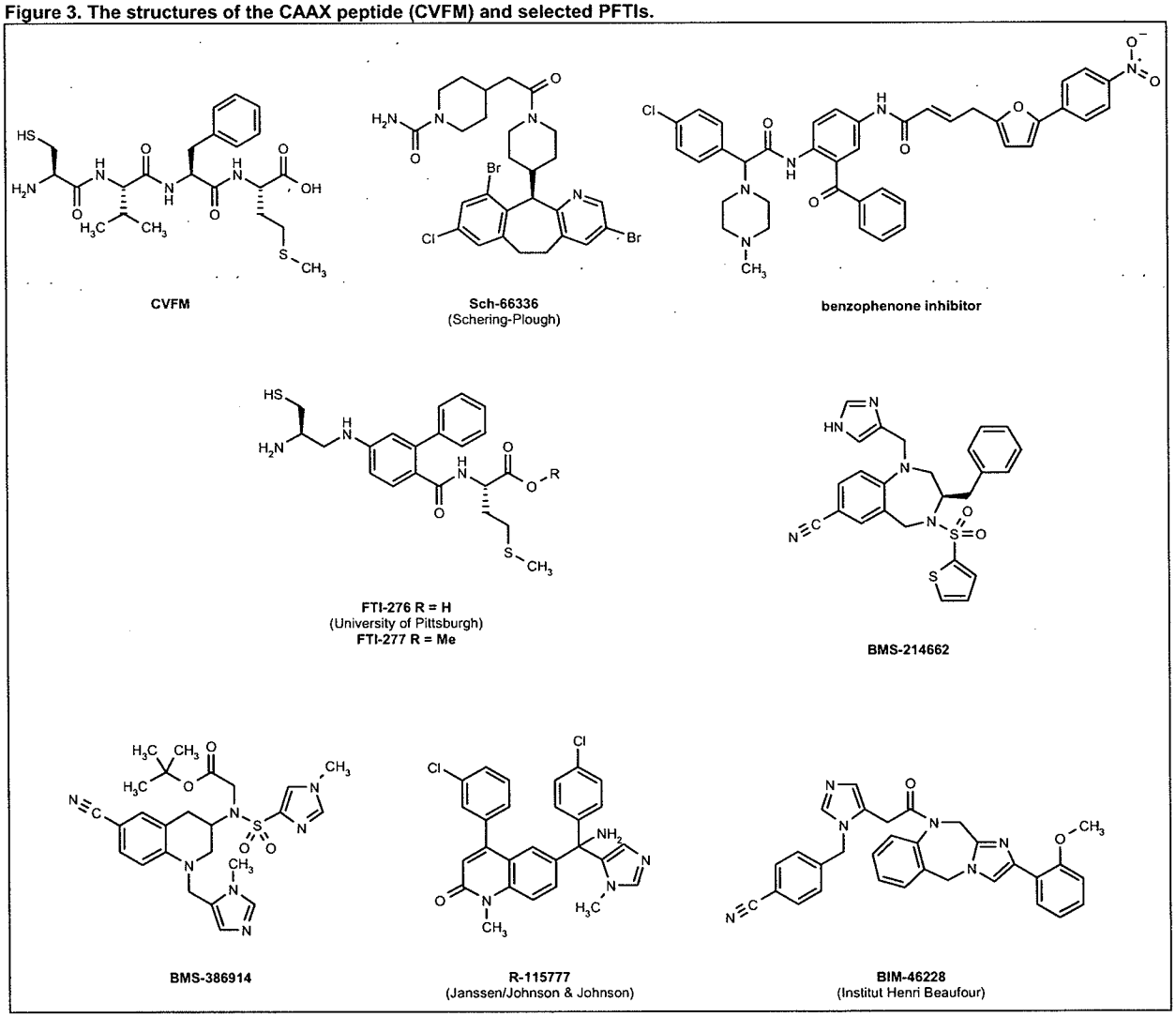


Table 2. Enzyme and cellular activity of PFTIs.

Compound	PFT inhibition (IC_{50} (nM))			Cell growth inhibition (ED_{50} (nM))		
	T brucei	*P falciparum*	Mammalian	*T brucei*	*P falciparum*	Mammalian
BMS-214662	1.90	8.00	1.35[a]	200	200.00	1000
BMS-386914	50.00	0.70	1.20	500	5.00	> 5000
R-115777 (Janssen Pharmaceutica BV/Johnson & Johnson; Figure 3)	3.50	10.00	0.10	2500	3000.00	5000
Sch-66336 (Schering-Plough Research Institute)	No inhibition at 50 nM	> 250	9.00	8500	3000.00	> 10,000
BIM-46228 (Institut Henri Beafour; Figure 3)	30% at 50 nM	7.20	-	> 10,000	> 1000.00	-
FTI-276 (University of Pittsburgh)	1.70	1.00[b]	4.00	> 25,000	> 60,000.00	> 25,000
FTI-277	-	-	-	500	3400.00	> 25,000
JJ-10	1.80	14.50	0.82[c]	1000	> 20,000	> 25,000
JJ-23	-	-	-	25	3200.00	25,000
Benzophenone #4	-	210.00[d]	-	-	-	-

Activity against *T brucei* PFT was tested with partially purified protein from *T brucei* procyclic forms [26], or using recombinant protein produced in Sf9 cells [14•]. Activity against *P falciparum* PFT was measured using partially purified protein from infected erythrocytes provided by Chakrabarti *et al* [16•], except for the benzophenone data which were taken from the literature [22]. Activity against mammalian PFT were measured using recombinant rat enzyme [26], with the exception of BMS-214662 and FTI-276 where values were taken from the literature as noted and were obtained using human enzyme. Note that all values may not be perfectly comparable because of different assay methods used for the PFT assays. Cell data were collected using *T brucei* bloodstream forms (strain BF427), *P falciparum* strain 3D7, or murine 3T3 fibroblasts. [a]reference [27•], [b]reference [16], [c]reference [28], [d]reference [22].

Piggy-backing for parasite drug development

Numerous PFTIs are being developed for clinical use. We, therefore, screened compounds against parasite PFT enzymes and against cultures of trypanosomatids and *Plasmodium*. Many of these compounds had IC_{50} values in the low nanomolar range against parasite PFTs (Table 2). Some compounds showed selective activity against the mammalian PFT (particularly Sch-66336), which was not surprising since the compounds were optimized for activity against mammalian enzymes [23]. Compounds that are significantly more selective for parasite enzymes have not yet been identified. Interestingly, for compounds with similar ability to block mammalian and parasite PFTs, the cytoxicity against cultures of *T brucei* or *P falciparum* is dramatically greater than the cytotoxicity against mammalian cells (Table 2). There are at least two theoretical reasons for this observation, although these have not been established experimentally. One possible reason may be, as explained above, due to a lack of redundancy in the prenylation enzymes in *T brucei* (and possibly *P falciparum*) which may render parasite cells more sensitive to PFT inhibitors. The second potential explanation may relate to the functional importance of proteins that undergo farnesylation in the cells. This is speculative because little is known about the parasite proteins that undergo farnesylation. The only trypanosomatid protein specifically shown to be farnesylated is the Rho of *T cruzi* (it is not known if the Rho homologs of *T brucei* or *P falciparum* are farnesylated) [24]. Functional Rho and/or other farnesylated proteins may be indispensable for the survival of these protozoa.

FTI-276 and FTI-277 (Figure 3), and JJ-10 and JJ-23 were the first compounds determined to have potent activity against *T brucei* PFT and cells [15,25]. Subsequent analogs have been generated with ED_{50} values as low as 5 nM [25]. Unfortunately, further development of these peptidomimetic compounds has been thwarted by pharmacokinetic problems relating to the methionyl unit. Specifically, the compounds need to be made as methyl esters (ie, FTI-277 and JJ-23) in order to be adequately hydrophobic to penetrate into cells (note the potency of FTI-277 and JJ-23 compared with the free acid forms, FTI-276 and JJ-10). It is assumed that intracellular enzymes metabolize the ester prodrugs to active acid compounds that bind PFT with high affinity. The difficulty has been that animal serum rapidly metabolizes the methyl ester prodrugs, such that the compounds are too quickly converted to the free acid forms in the circulation before being able to enter the cells and act on their target. In order to address this liability, alternative compounds with substitutions of the methionyl unit have been generated, but these have not retained the necessary potency against PFT to result in low ED_{50} values. Additional variants are being investigated.

The Bristol-Myers Squibb series of compounds (eg, BMS-214662 and BMS-386914; Figure 3) have produced the most promising PFTIs and have become the focus of efforts for drug development projects in our research group supported by the Medicines for Malaria Venture and by the Drugs for Neglected Diseases Initiative. BMS-214662 inhibited *T brucei* and *P falciparum* PFTs at single digit nanomolar concentrations and had favorable activity on parasite growth (Table 2). A diverse collection of several hundred PFTIs related to this lead compound were also sampled. Compounds with the tetrahydrobenzodiazepine core (eg, BMS-214662) proved to be less potent than another series of compounds containing a tetrahydroquinoline

(THQ) core (eg, BMS-386914). Concerns regarding the gastrointestinal toxicity observed in clinical trials with BMS-214662 also demonstrated that molecules with the THQ core are preferable. BMS-386914 and a number of other THQ compounds inhibit *P falciparum* PFT at subnanomolar concentrations. At concentrations that inhibit proliferation, specific inhibition of farnesol incorporation into a farnesylated 50-kDa protein occurs with BMS-386914, but there is no inhibition of incorporation into a geranylgeranylated 22- to 28-kDa protein. This shows that these inhibitors specifically block PFT but not PGGT activity. To produce complete killing *in vitro*, *P falciparum* requires exposure to 30-fold the ED_{50} of BMS-386914 for 72 h. *In vitro* selection of *P falciparum* mutants resistant to THQ inhibitors leads to point mutations in the active site of PFT, further demonstrating that these inhibitors kill *P falciparum* by inhibiting PFT. BMS-386914 demonstrated an ED_{50} value of 5 nM, which is twice as potent as chloroquine *in vitro*. BMS-386914 is not well absorbed when administered orally, but when administered by subcutaneous infusion pump to mice infected with *P berghei*, it was able to suppress parasitemia in five out of five animals and eliminate parasites completely in 60% of the animals [23].

Changing the methyl-imidazole attached to a sulfonamide group to a 2-pyridine dramatically improved oral absorption of the THQ series of compounds. However, the most potent members of this series do not achieve the long serum half-lives that are desirable for antimalarial compounds (which are ideally administered orally once daily). New modifications to the molecule, particularly at the sulfonamide nitrogen where profound changes have little effect on enzyme potency, have led to significant improvements in stability in liver microsome assays and in circulating half-lives of the compounds in rodents. A structural model of the *P falciparum* PFT (based on the rat PFT-BMS-214662 complex) is helping to guide the production of second-generation THQ inhibitors with retained or even improved potency. A new THQ compound synthesized by our research group suppresses parasitemia in rats infected with *P berghei* when dosed orally. This new compound or a similar analog will hopefully be the subject of more intensive preclinical evaluation as the project for PFTIs for malaria moves ahead.

BMS-214662 dramatically suppressed parasitemia in the mouse model of African sleeping sickness [W Van Voorhis, unpublished data]. Because of concerns with the tetrahydrobenzodiazepine group of molecules mentioned above, we have mainly focused our research on THQ-containing compounds for the development of anti-*T brucei* drugs. The most potent THQ inhibitor against cultures of *T brucei* had an ED_{50} value of approximately 60 nM. Efforts to improve potency are guided by a structural model of the *T brucei* enzyme. Since it is potentially desirable to have a parenterally administered drug for treating patients with African sleeping sickness, we have not focused on the issue of oral bioavailability. The immediate task in the *T brucei* project is the identification of compounds with sufficient potency and pharmacokinetic properties to cure infection. Subsequent testing will address the question of central nervous system penetration to cure late-stage African sleeping sickness.

A research group at Ludwig-Maximilians University in Germany has discovered a separate class of PFT inhibitors with antiplasmodial activity [22]. These compounds contain a benzophenone scaffold (Figure 3). *In silico* docking studies indicate that the compounds bind to the active site of PFT without direct association with the catalytic zinc atom. The most potent inhibitor in the class has an IC_{50} value of 64 nM, but has poor solubility in aqueous solution. A less potent compound had better solubility and demonstrated activity in the *P vinckei* model of malaria in mice (ED_{50} = 21 mg/kg when dosed intraperitoneally for 3 days). The compounds appear to be non-toxic at doses of < 100 mg/kg, but they do not have oral activity.

Conclusions

Protein farnesyl transferase inhibitors have potent cytotoxic activity against *P falciparum* and *T brucei* parasites. Despite little specificity of available inhibitors for parasite PFT over mammalian PFT, the inhibitors are much more toxic to parasitic cells than mammalian cells. This is likely due to the absence of back-up enzymes (eg, PGGT-I) in the parasites or due to greater dependence of parasite cells on farnesylated proteins. Hence, the challenge in PFT drug development is not to identify selective antiparasite compounds, but to identify compounds with sufficient potency and pharmacokinetic properties to make satisfactory drugs for malaria and African sleeping sickness. Over the next few years, optimization of these compounds and the opportunity to begin clinical trials are anticipated.

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