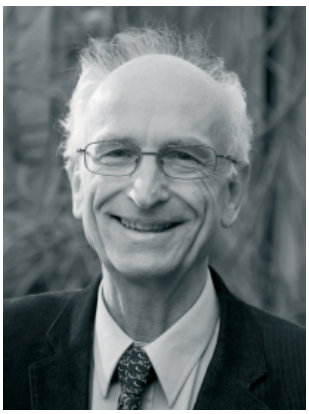

1964 - 1966	MD – Max Planck Institute for Medical Research, Heidelberg (Prof. J.C. Rüegg)
1966 - 1970	PostDoc - Dartmouth Medical School, Hanover NH, USA (Prof. L.H. Noda); Internship and residency at hospitals in Heidelberg, Stuttgart, and Karlsruhe
1970 - 1980	Group Leader in Biophysics - Max Planck Institute for Medical Research, Heidelberg
1975	Habilitation in Biochemistry
1980 - 2007	Professor - Institute of Biochemistry II, University of Heidelberg / BZH; now professor emeritus

Heiner Schirmer

Drugs und transmission blockers against pediatric malaria

Goal

To develop affordable and accessible medicines for malaria

Background

Falciparum malaria is a disease which in its dangerous form mainly affects preschool children, pregnant women and tourists.

Our work focuses on redox milieu-targeting drug combinations against pediatric malaria.

The biochemical networks that maintain the cytosolic redox potential at values below –250 mV are based in many organisms on a dual system, the glutathione system and the thioredoxin system (Buchholz *et al* 2010). We study these systems primarily in the protozoal parasite *Plasmodium falciparum*, its insect vector *Anopheles gambiae* and the human host. Differences in the proteins of the redox networks are exploited for the development of species-specific and stage-specific therapeutic agents. We focus as targets on the disulfide reductases glutathione reductase and thioredoxin reductase, as well as dihydrolipoamide dehydrogenase. The phenothiazine methylene blue (MB), a subversive substrate and inhibitor of disulfide reductases, is currently tested as a partner in antimalarial drug combinations (Bountogo *et al* 2010, Müller *et al* 2009, Schirmer *et al* 2008).

Research Highlights

When reducing MB, the disulfide reductases utilize the flavin cofactor and not the active site cysteine pair (Fig.1) for electron transfer (Buchholz *et al* 2008).

Pyocyanin, a social signal and respiratory pigment from *Pseudomonas aeruginosa* is a natural counterpart of the synthetic drug methylene blue (see legends of Figs 1 and 2; Schirmer *et al* 2008).

Antimalarial MB-combination therapies like MB-artemisinine and MB-amodiaquine are currently studied by Olaf Müller, Peter Meissner and Boubacar Coulibaly in clinical trials at the Centre de Recherche en Santé de Nouna (CRSN) in Burkina Faso. Coulibaly did his thesis work at the BZH and, in 2004, was the first Burkinabé to obtain a PhD in biochemistry. MB is not only active against Plasmodium schizonts but also against Plasmodium gametocytes (Coulibaly *et al* 2009) which means that MB can block transmission of the disease from patient to patient via the mosquito. Thus MB-containing antimalarial drug combinations may become important for malaria elimination programs.

Up to June 2010, the combination of MB and amodiaquine was considered an ethical drug; this drug combination is effective, safe, affordable, ac-

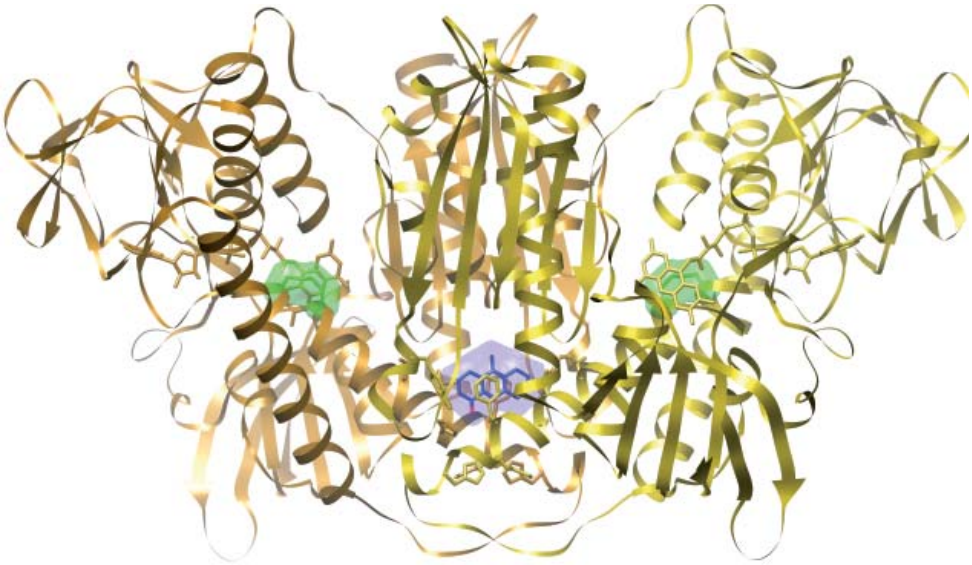

Fig. 1: Cytosolic human glutathione reductase homodimer with bound pyocyanin (PYO). *Pyocyanin (blue) and FAD (yellow) are represented as ball and stick models. Additionally, the surfaces of the catalytic cysteines Cys58/Cys63 and Cys58'/Cys63' (green) and of PYO (blue) are shown. Azure B (monodemethyl MB), the major metabolite of MB, binds to the same site as pyocyanin. The MB structure itself is too large to be accommodated here (Karin Fritz-Wolf, personal communication).*

cessible and available in sufficient dosages. The rumours, however, that cationic MB might interfere with the growth of phospho-tau filaments and thus delay the onset of Alzheimer disease have recently contributed to a shortage of and a price explosion for MB as a cGMP-grade raw material from less than € 150 to € 30000 *(http://www.alzforum.org/new/Schirmer.asp)*. If this situation does not change MB has no future as a drug for malaria as a disease of the poor. As a consequence, we study the cell biochemistry of azure B, the major metabolite of MB in man. Azure B (see legends of Figs. 1 and 2) may indeed be the active form of MB in a number of therapeutic indications, with MB serving as a pro-drug of azure B.

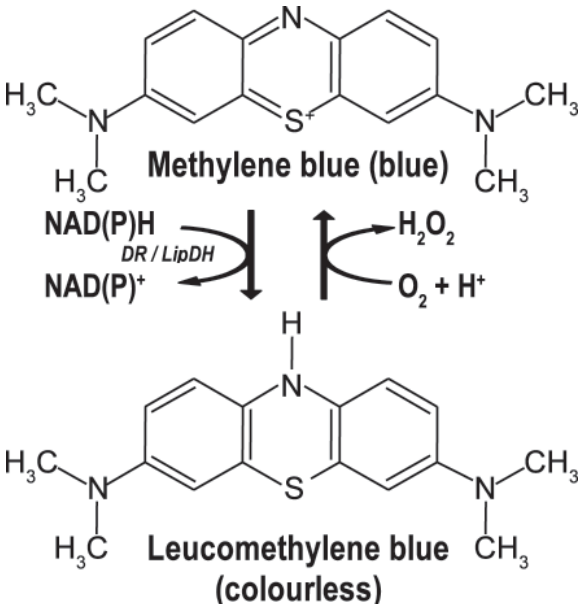

Fig. 2 Methylene blue as an H_2O_2-producing subversive redox-cycler. *The enzyme glutathione reductase and other disulfide reductases of the malaria parasite catalyze the reduction of methylene blue to leucomethylene blue. Leucomethylene blue auto-oxidizes instantaneously regenerating MB and producing parasitocidal H_2O_2. Pyocyanin and azure B can undergo the same redox-cycling as MB.*

Selected Publications 2008 - 2010

Bountogo M, Zoungrana A, Coulibaly B, Klose C, Mansmann U, Mockenhaupt FP, Burhenne J, Mikus G, Walter-Sack I, Schirmer RH, Sié A, Meissner P, Müller O (2010) Efficacy of methylene blue monotherapy in semi-immune adults with uncomplicated falciparum malaria: a controlled trial in Burkina Faso *Trop Med Int Health 15*, 713-717

Buchholz K, Putrianti ED, Rahlfs S, Schirmer RH, Becker K, Matuschewski K (2010) Molecular genetics evidence for the *in vivo* roles of the two major NADPH-dependent disulfide reductases in the malaria parasite. *J Biol Chem 285:* 37388-37395

Buchholz K, Schirmer RH, Eubel JK, Akoachère MB, Dandekar T, Becker K, Gromer S (2008) Interactions of methylene blue with human disulfide reductases and their orthologues from *Plasmodium falciparum*. *Antimicrob Agents Chemother 52*, 183-191

Coulibaly B, Zoungrana A, Mockenhaupt FP, Schirmer RH, Klose C, Mansmann U, Meissner P, Müller O (2009) Strong gametocytocidal effect of methylene blue-based combination therapy against falciparum malaria: a randomised controlled trial. *PloS ONE 4*, e5318

Müller O, Sié A, Meissner P, Schirmer RH, Kouyaté B (2009) Artemisinin resistance on the Thai-Cambodian border. *The Lancet 374,* 1418-1419

Schirmer RH, Adler H, Zappe HA, Gromer S, Becker K, Coulibaly B, Meissner P (2008) Disulfide reductases as drug targets: Methylene blue combination therapies for falciparum malaria in African children. *Flavins and Flavoproteins 16,* 481-486

Awards and Honors

1976	Appointment as a Bicentennial Lecturer in Philadelphia and Boston
2002-2009	Dream Action Award of the Dutch chemical company DSM

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