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Interactions of mefloquine with praziquantel in the *Schistosoma mansoni* mouse model and *in vitro*

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Objectives: Mefloquine has interesting antischistosomal properties, hence it might be an attractive partner drug for combination treatment with praziquantel. The aim of this study was to evaluate activities of mefloquine/praziquantel combinations against *Schistosoma mansoni in vitro* and *in vivo*.

Methods: Dose–response relationships were established following exposure of adult *S. mansoni* to mefloquine, praziquantel and fixed dose combinations of mefloquine/praziquantel *in vitro*. *S. mansoni*-infected mice were treated orally with selected doses of single drugs and drug combinations 7 weeks post-infection.

Results: We calculated *in vitro* LC_{50} values of 0.024 and 1.9 μg/mL for praziquantel and mefloquine, respectively. Mefloquine/praziquantel combinations showed synergistic effects, with combination index (CI) values <1 when adult *S. mansoni* were simultaneously incubated with both drugs *in vitro*. Reduced viabilities were also observed when schistosomes were first exposed to mefloquine followed by praziquantel *in vitro*. ED_{50}s of 62 mg/kg and 172 mg/kg were determined for mefloquine and praziquantel against adult *S. mansoni in vivo*, respectively. Combinations of praziquantel (50 or 100 mg/kg) followed the next day by mefloquine (50 or 100 mg/kg) treatment revealed only moderate total worm burden reductions of 47.8%–54.7%. On the other hand, when both drugs (100 mg/kg each) were either given simultaneously or mefloquine was given prior to praziquantel, high total and female worm burden reductions of 86.0%–93.1% were observed. For the later treatment regimen, synergistic effects ($CI<1$) were calculated when mefloquine and praziquantel were combined using a fixed dose ratio based on their ED_{50}s.

Conclusions: Combinations of mefloquine and praziquantel may have clinical utility in the treatment of schistosomiasis.

Keywords: schistosomiasis, combination chemotherapy, activity, combination index, isobolography

Introduction

In the treatment of tuberculosis, cancer or malaria, drugs are often given in combination to increase their therapeutic advantages.[1–3] The clinical effect of a combination of two drugs should either be the sum (additive behaviour) or ideally even exceed (synergy) the individual effect of each drug. On the other hand, for adverse events, antagonism (the effect of the two drugs being less than the effect of each drug) is preferable. In addition, combination chemotherapy is a viable therapeutic strategy to delay the development of drug resistance.[4]

There is scarce information available as to whether antischistosomal drug combinations provide an increased therapeutic efficacy over monotherapy. A few clinical trials have evaluated praziquantel plus oxamniquine combinations and combinations of praziquantel with an artemisinin derivative.[5,6] In the laboratory, combinations of 'something old' (praziquantel) with 'something new', e.g. novel experimental drugs such as Ro 15-5458[7] or nilutamide,[8] were studied. However, the great disadvantage of these combinations (involving novel drug candidates) is the long drug development process (12–15 years), and therefore the associated high costs, in the order of $1 billion.[9]

Another possibility for an antischistosomal drug combination might be a polytherapy with praziquantel and the antimalarial drug mefloquine. Several laboratory studies have demonstrated interesting antischistosomal properties of mefloquine. For example, a single 200 mg/kg oral dose of mefloquine achieved a worm burden reduction of 72% in mice harbouring a chronic *Schistosoma mansoni* infection.[10] In addition, in a randomized, exploratory open-label trial in Côte d'Ivoire in *Schistosoma*

haematobium-infected schoolchildren, a mefloquine/artesunate combination achieved a cure rate of 61% and an egg reduction rate of 95%.[11]

The aim of the present study was to evaluate the effect of mefloquine/praziquantel combinations against *S. mansoni in vitro* and *in vivo*. A preliminary study has already pointed to significant worm burden reductions following treatment with praziquantel plus mefloquine in mice infected with *Schistosoma japonicum*.[12] We determined whether the potency of this drug combination is additive, antagonistic or synergistic.[13] In addition, we analysed whether mefloquine/praziquantel combinations should be given simultaneously or in sequence.

Materials and methods

Animals and parasites

Female NMRI mice ($n=125$, age=3 weeks, weight ~35 g), obtained from Harlan Laboratories (Horst, The Netherlands), were kept under standard conditions (temperature, ~25°C; humidity, 70%; 12 h light and 12 h dark cycle) with free access to water and rodent diet in accordance with the Swiss national and cantonal regulations on animal welfare. Experiments were approved by the local veterinary agency (permit 2070). *S. mansoni* cercariae (Liberian strain) were collected after exposing *Biomphalaria glabrata* to light for 3 h. Mice were infected subcutaneously with ~80 cercariae.

Drugs

Praziquantel was purchased from Sigma (Buchs, Switzerland), and mefloquine hydrochloride was kindly provided by Mepha Pharma AG (Aesch, Switzerland). For *in vitro* studies, drugs were dissolved in 100% DMSO (Fluka, Buchs, Switzerland) to obtain stock solutions of 10 mg/mL. For *in vivo* studies, drugs were prepared as suspensions in 7% (v/v) Tween 80 and 3% (v/v) ethanol before oral administration to mice (10 mL/kg).

In vitro assay procedures

Preparation of adult S. mansoni *and culture conditions*

Forty-nine-day-old adult schistosomes, removed by picking from the hepatic portal system and mesenteric veins from infected NMRI mice, were washed with PBS (pH 7.4) and kept in RPMI 1640 culture medium [supplemented with 5% inactivated fetal calf serum (iFCS) and 100 U/mL penicillin and 100 μg/mL streptomycin (Invitrogen, Carlsbad, CA, USA)] at 37°C in an atmosphere of 5% CO_2 until use.

Combination chemotherapy studies on adult S. mansoni *in vitro*

In a first step, the lethal concentrations (LC_{100}) that kill all schistosomes within 72 h of *in vitro* drug exposure and the median effect concentrations (LC_{50}) were determined for praziquantel and mefloquine. Drugs were serially diluted in 24-well plates (Costar) in RPMI 1640 culture medium and 2 male and 2 female worms were added to each well. Praziquantel concentrations of 1, 0.5, 0.2, 0.1, 0.05, 0.01 and 0.001 μg/mL and mefloquine concentrations of 10, 9, 6, 5, 2, 1, 0.5, 0.2, 0.1 and 0.01 μg/mL were tested. Each drug concentration was assessed in duplicates and repeated once ($n=12$ worms/drug concentration). For the interaction studies, mefloquine and praziquantel were added simultaneously in a first experiment at a fixed dose ratio based on the calculated LC_{50} values (1.9 μg/mL for mefloquine and 0.024 μg/mL for praziquantel) and 2-fold dilutions were carried out up and down (7.6 and 0.1 μg/mL; 3.8 and 0.05 μg/mL; 1.9 and 0.024 μg/mL; 0.95 and 0.0125 μg/mL; 0.475 and 0.006 μg/mL; and 0.238 and 0.003 μg/mL of mefloquine and praziquantel, respectively). In addition, we studied a 5-fold dilution of the LC_{50} value (combination of 0.03 μg/mL mefloquine and 0.0004 μg/mL praziquantel). Worms were incubated at 37°C and 5% CO_2 for 72 h, their viabilities recorded using a microscope (8–40-fold magnification; Carl Zeiss AG, Germany) and the mean viability of the 12 examined worms calculated as described previously.[14] Worms were classified as dead if no movement was observed for 2 min and worms had a dark colour. In a second experiment, schistosomes were exposed to LC_{50}:LC_{50} (1.9 μg/mL for mefloquine and 0.024 μg/mL for praziquantel) and 0.5 LC_{50}:0.5 LC_{50} (0.95 μg/mL for mefloquine and 0.0125 μg/mL for praziquantel) and drug addition was spaced by the respective half-life of the drugs in mice.[15,16] In more detail, schistosomes were exposed to (i) praziquantel followed by mefloquine 1 h post-incubation and (ii) mefloquine followed by praziquantel 17 h post-exposure. The viabilities of these worms were assessed 72 h after drug incubation. Worms incubated in medium containing the highest solvent concentration used (1% DMSO) served as controls in all experiments.

In vivo studies

Monotherapy

Forty-nine days post-infection, groups of 6–11 mice were treated orally with subtherapeutic single oral doses of mefloquine (50 and 100 mg/kg) and praziquantel (50, 100, 150 and 200 mg/kg). Untreated mice served as controls. At 21 days post-treatment, animals were killed by the CO_2 method and dissected. Worms were removed by picking, then sexed and counted as described in previous publications.[17] For the calculation of the ED_{50} values, worm burden reductions obtained in recent experiments with effective doses of mefloquine (200 mg/kg) and praziquantel (400 mg/kg) were included.[10,17]

Effect of treatment schedule

We evaluated whether the administration schedule has an influence on the activity of the drug combination. Six groups of mice were treated with combinations of mefloquine and praziquantel (50 mg/kg mefloquine plus 50 mg/kg praziquantel, 100 mg/kg mefloquine plus 100 mg/kg praziquantel) administered either simultaneously or on subsequent days (mefloquine followed by praziquantel or praziquantel followed by mefloquine). Untreated mice were included as controls. Worms were recovered as described above.

Effect of drug interactions

To determine the combination dose effect, four groups of mice were treated with combinations based on their ED_{50}s (1:2.8 ratio and 2-fold dilutions up and down). The treatment was administered on subsequent days (mefloquine followed by praziquantel), as this regimen has shown the highest activity in our experiments *in vitro* and *in vivo*. Three weeks post-treatment, mice were killed and processed as described above.

Scanning electron microscopy study

We collected adult *S. mansoni* 72 h post-treatment from three mice as described above, which had been treated with (i) mefloquine (60 mg/kg), (ii) praziquantel 170 mg/kg, and (iii) mefloquine (60 mg/kg) followed on the next day by praziquantel (170 mg/kg). Worms were fixed with 2.5% (v/v) glutaraldehyde in PBS (pH 7.4) for several hours. The schistosomes

were then washed twice with double-distilled water, dehydrated in ascending ethanol concentrations and critically point dried (Bomar SPC-900; Tacoma, WA, USA). Finally, worms were sputter coated with 20 nm gold particles and observed using a high-resolution scanning electron microscope (Phillips XL30 ESEM) at an accelerating voltage of 5 kV.

Statistical analysis

LC_{50} and ED_{50} values, combination index (CI), dose reduction index (DRI) and isobologram plots were calculated using the CompuSyn software package (ComboSyn, Paramus, NJ, USA). LC_{50} plots were drawn using XL*fit*® (xlfit5, IDBS, Guildford, UK). We used the Kruskal–Wallis (KW) test to compare the medians of the worm burdens in the monotherapy versus combination chemotherapy treatment groups [version 2.4.5 Statsdirect (Cheshire, UK)]. A difference in median was considered to be significant at a significance level of 5%.

Results

In vitro studies

Determination of LC_{50} values of monotherapy

The dose–response curves of mefloquine and praziquantel are depicted in Figure 1. LC_{50} values of 0.024 and 1.9 μg/mL were calculated for praziquantel and mefloquine, respectively. The corresponding LC_{75} and LC_{95} values are 0.04 and 0.11 μg/mL for praziquantel and 3.4 and 9.2 μg/mL for mefloquine, respectively.

Simultaneous drug administration

In Figure 1, the dose–response curve of adult *S. mansoni* exposed simultaneously to praziquantel and mefloquine (LC_{50}:LC_{50}) *in vitro* is shown. Figure 2 illustrates the combination dose effect using an isobologram.

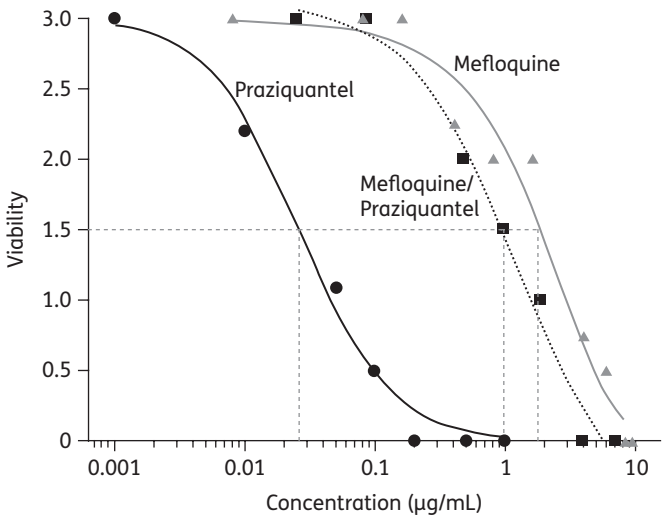

Figure 1. Dose–response curves of mefloquine and praziquantel and combined mefloquine/praziquantel (LC_{50}:LC_{50}) against adult *S. mansoni* *in vitro* using a viability score.[17] The broken line represents the concentration required to achieve a medium effect level (LC_{50}). Data points represent mean values of viabilities of three assay wells (12 schistosomes) for each drug concentration and combination.

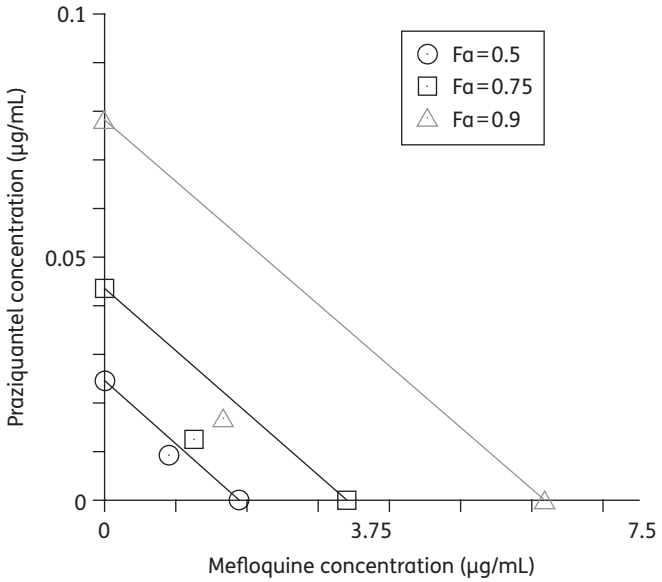

Figure 2. Isobologram showing the synergistic interaction of a mefloquine/praziquantel combination using the LC_{50}:LC_{50} ratio *in vitro* at the LC_{50}, LC_{75} and LC_{90}. Fa, fraction affected.

Praziquantel/mefloquine combinations showed a synergistic effect on adult *S. mansoni*, with CI values <1 (CI 0.87 at the LC_{50} and 0.40 at the LC_{95}). To achieve a 50% reduction of schistosome viability the praziquantel and mefloquine concentrations could be reduced 2.6- and 2.1-fold, respectively.

Spaced drug administration

When praziquantel and mefloquine were added to the *in vitro* cultures with a time lag corresponding to their respective half-lives, only schistosomes exposed first to mefloquine and 17 h later to praziquantel were affected and showed reduced viabilities (a viability of 1 for the combination of 1.9 μg/mL mefloquine and 0.024 μg/mL praziquantel and a viability of 1.5 for the combination of 0.95 μg/mL mefloquine and 0.0125 μg/mL praziquantel) within 72 h. Parasites exposed to praziquantel for 1 h followed by mefloquine revealed only a slight loss of viability (viability of 2.0 for both combinations tested) in comparison with the untreated controls.

In vivo studies

ED_{50} calculation for mefloquine and praziquantel

For the ED_{50} calculation we also included results obtained from previous experiments in our laboratories.[10,17] Treatment of *S. mansoni*-infected mice with 50, 100 or 200 mg/kg mefloquine resulted in total worm burden reductions of 44.1, 64.0 (Table 1) and 93.4%,[17] respectively. We calculated an ED_{50} of 62 mg/kg and an ED_{95} of 262 mg/kg.

Worm burden reductions of 13% (50 mg/kg; Table 1) up to 96% (400 mg/kg)[10] were observed following treatment with praziquantel. Praziquantel given at 172 mg/kg and 592 mg/kg is estimated to achieve worm burden reductions of 50% and 95%, respectively.

Table 1. Effect of praziquantel and mefloquine monotherapies administered at single oral doses of 50–100 mg/kg and praziquantel/mefloquine combinations (50/50 and 100/100 mg/kg) following three different treatment schedules to mice harbouring a 49-day-old *S. mansoni* infection

	Drug	Dose	No. of mice investigated	No. of mice cured	Mean number of worms (SD)			Total worm burden reduction (%)	Female worm burden reduction (%)
					total	males	females		
Control	—	—	20	—	24.7 (14.7)	14.1 (7.6)	10.7 (7.6)		
Monotherapy	praziquantel	50	4	0	21.5 (10.5)	10.0 (3.4)	11.5 (7.2)	13.0	0
		100	6	0	21.0 (9.5)	11.5 (5.8)	9.5 (3.8)	15.0	11.2
	mefloquine	50	11	0	13.8 (9.4)	8.0 (11.6)	5.8 (4.2)	44.1*	45.8
		100	9	0	8.9 (3.9)	6.4 (3.4)	2.4 (1.4)	64.0*	77.6*
Combination chemotherapy, simultaneous application	praziquantel/mefloquine	50/50	8	0	18.3 (8.5)	10.6 (5.7)	7.6 (3.1)	25.9	29.0
	praziquantel/mefloquine	100/100	8	1	3.5 (3.9)	2.1 (2.9)	1.4 (1.1)	86.0*	86.9
Combination chemotherapy, mefloquine followed by praziquantel	praziquantel/mefloquine	50/50	8	0	9.5 (6.8)	5.0 (3.6)	4.5 (3.3)	61.5	58.0
	praziquantel/mefloquine	100/100	9	4	1.7 (2.0)	0.9 (1.1)	0.8 (1.0)	93.1**	93.0*
Combination chemotherapy, praziquantel followed by mefloquine	praziquantel/mefloquine	50/50	8	0	12.9 (7.8)	6.6 (4.3)	6.3 (3.7)	47.8	41.1
	praziquantel/mefloquine	100/100	9	0	11.2 (8.8)	8.2 (6.6)	3.0 (3.1)	54.7	72.0

*$P<0.05$.
**$P<0.001$.

Effect of combination treatment regimen on efficacy

Based on the results obtained in our *in vitro* experiments, which showed differences depending on the treatment schedule used (simultaneous or spaced incubation), we were interested whether these findings could also be documented *in vivo*. Mice were divided in three groups. Group 1 was treated with both drugs simultaneously, group 2 was treated with mefloquine followed by praziquantel 24 h later, and group 3 was treated with praziquantel followed by mefloquine 24 h later. We used subtherapeutic doses of 50 and 100 mg/kg for each of the drugs.

The results are summarized in Table 1. When mefloquine and praziquantel were administered simultaneously at doses of 50 mg/kg each, low total (25.9%) and female (29.0%) worm burden reductions were observed, which were even lower than worm burden reductions observed with mefloquine (50 mg/kg) alone. When both drugs were given at 100 mg/kg simultaneously, high total and female worm burden reductions of 86.0% ($P=0.014$) and 86.9% ($P=0.091$), respectively, were calculated.

High significant worm burden reductions (total worm burden reduction of 93.1% ($P<0.001$) and female worm burden reduction of 93.0% ($P=0.008$) were observed when mice were treated with 100 mg/kg mefloquine followed by 100 mg/kg praziquantel 24 h later. When half of these doses were given, total and female worm burden reductions decreased to 61.5% and 58.0%, respectively.

Combinations of praziquantel (50 or 100 mg/kg) followed the next day by mefloquine (50 or 100 mg/kg) treatment revealed only moderate total worm burden reductions of 47.8%–54.7%, which were similar to the results obtained with mefloquine monotherapy.

Combination dose–effect analysis

We used a constant ratio design based on the ED_{50}s of both drugs (1:2.8) to analyse whether a mefloquine/praziquantel combination reveals additive, antagonistic or synergistic effects. Since the highest worm burden reductions were obtained when praziquantel followed mefloquine administration, this treatment schedule was employed to determine the combination dose effect. Significant total worm burden reductions of 91.8% and 97.8% were observed at the two highest concentrations tested (Table 2). At a dose of 30 mg/kg mefloquine and 85 mg/kg praziquantel, total and female worm burden reductions of 51.6% and 48.8%, respectively, were observed, which were not statistically significant. At the lowest dose tested (15 mg/kg mefloquine, 42.5 mg/kg praziquantel), low total and female worm burden reductions of 19.2% and 19.8%, respectively, were achieved. The ED_{50} dose of the combination was calculated as 101.8 mg/kg (26.8 mg/kg mefloquine and 75.1 mg/kg praziquantel, corresponding to a 2.3-fold dose reduction for each drug). A CI of 0.87 was determined at the median dose–effect level. At the ED_{75}, ED_{90} and ED_{95}, CI values were below 0.8. Hence, at the dose ratio for combined praziquantel and mefloquine (1:2.8), synergistic interactions were observed.

Scanning electron microscopy study

As expected, only very mild tegumental alterations (a handful of blebs, swelling and fusion of the tegumental ridges) were

Table 2. Effect of praziquantel/mefloquine combinations using a constant ratio design based on the ED_{50}s administered to mice harbouring a 49-day-old *S. mansoni* infection

	Dose (mg/kg)	No. of mice investigated	No. of mice cured	Mean number of worms (SD)			Total worm burden reduction (%)	Female worm burden reduction (%)
				total	males	females		
Control	—		—	15.2 (6.3)	8.0 (2.9)	7.2 (3.5)	—	—
Combination chemotherapy, mefloquine followed by praziquantel	15 (mefloquine) and 42.5 (praziquantel)	4	0	12.3 (3.3)	6.7 (1.3)	5.5 (2.1)	19.2	19.8
	30 (mefloquine) and 85 (praziquantel)	4	0	7.3 (1.2)	3.7 (0.6)	3.7 (0.6)	51.6	48.8
	60 (mefloquine) and 170 (praziquantel)	4	2	1.3 (1.5)	0 (0)	1.3 (1.5)	91.8*	82.6*
	120 (mefloquine) and 340 (praziquantel)	4	3	0.3 (0.6)	0.3 (0.6)	0 (0)	97.8*	100*

*$P < 0.05$.

observed on adult schistosomes collected from mice treated with 60 mg/kg mefloquine and 170 mg/kg praziquantel, respectively (Figure 3a–d). Many worms revealed no tegumental damage. On the other hand, the majority of worms had already been expelled from a mouse treated with a combination of mefloquine and praziquantel 72 h post-treatment. Only a single worm could be recovered that showed extensive blebbing on its mid-body (Figure 3e).

Discussion

In several medical fields the search for effective drug combinations has been recognized as an important strategy for a successful treatment outcome and to delay drug resistance.[1–3] To our knowledge, we have performed the first analysis of the pharmacodynamic interactions of mefloquine/praziquantel combinations against *S. mansoni*. *In vitro* and *in vivo* studies were conducted, which allow assessment of drug combinations in much more detail than do clinical studies. We used isobologram and CI analyses, which are popular methods to analyse drug interactions of combination chemotherapy.[13] It is interesting to note that although several studies have analysed the *in vitro* and *in vivo* antischistosomal efficacy of praziquantel combinations, including the effect of combined treatment of mefloquine and praziquantel against *S. japonicum*,[12] in depth modelling of dose–effect relationships, defining additive effects, synergy or antagonism of these combinations have not been carried out to date.

Synergistic interactions were observed in the *S. mansoni* mouse model and *in vitro* when praziquantel was combined with mefloquine. This finding is encouraging since the control of schistosomiasis, a chronic and debilitating disease, relies on a single drug, praziquantel.[6,18] The need to develop alternative treatment options, including drug combinations, has been repeatedly emphasized because the development of a praziquantel-resistant schistosome strain is a threat.[6,19]

It has been suggested to analyse a series of different fixed dose ratios in combination treatment experiments to confirm whether two drugs behave additively or synergistically, since it has been shown that the effect might depend on the ratio of the drugs used.[20] For the evaluation of mefloquine/praziquantel combinations *in vivo* we used the median effect analysis only.[13] *In vitro*, however, a range of different dose ratios (including fixed ratios based on the LC_{50} presented here) were tested and analyses of these data confirmed the synergistic properties of the mefloquine/praziquantel combinations (data not shown).

It is interesting to note that the best results were achieved in the *S. mansoni* mouse model when praziquantel treatment followed mefloquine. On the other hand, only moderate worm burden reductions were achieved when praziquantel was administered prior to mefloquine. These findings were also observed *in vitro*. The effects of praziquantel on schistosomes might play a role in the antagonistic effects observed when praziquantel was administered first. Exposure of schistosomes to praziquantel results in a calcium-dependent contraction of the musculature, an increase in tension and a disrupted tegument of the worms.[21,22] One could speculate that due to the paralysis and damage of worms caused by praziquantel, the uptake of mefloquine by schistosomes might be decreased, resulting in lower

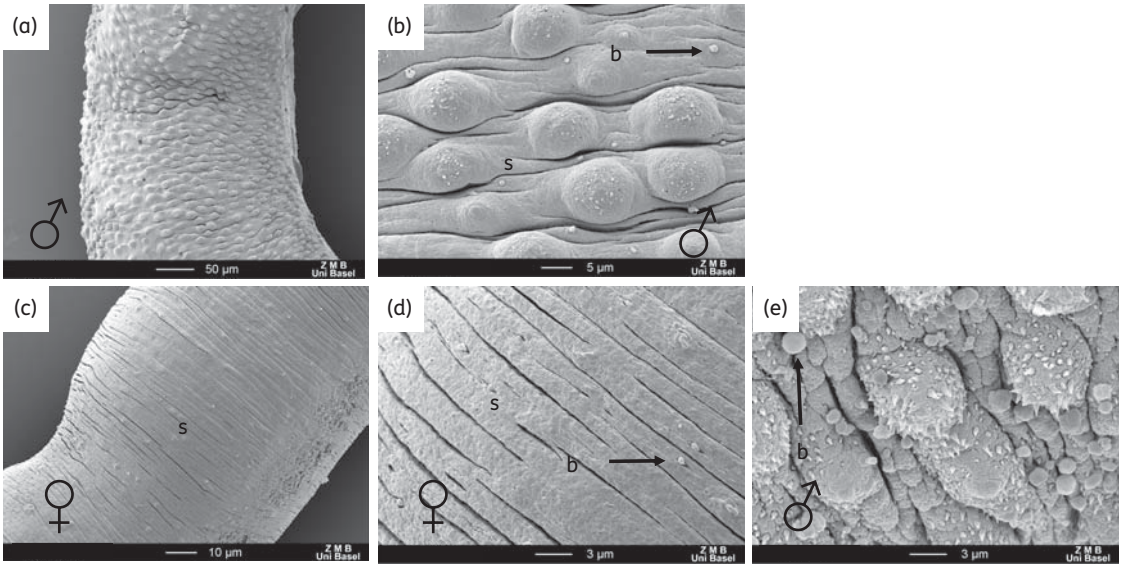

Figure 3. Scanning electron microscopy of adult *S. mansoni* recovered from mice 72 h after the administration of a single 170 mg/kg oral dose of praziquantel (a and b) and 60 mg/kg mefloquine (c and d). (a) Mid-body of a male specimen treated with 170 mg/kg praziquantel. (b) Higher magnification of (a). Blebbing (b) and swelling (s) observed. (c) Mid-body of a female specimen treated with 60 mg/kg mefloquine. (d) Higher magnification of (c). Blebbing (b) and swelling (s) visible. (e) Extensive blebbing observed on a male worm following a combination of 60 mg/kg mefloquine and 170 mg/kg praziquantel administered on subsequent days.

activities of the drug combination. On the other hand, the mechanism of action of mefloquine on schistosomes has not yet been elucidated. However, *in vivo* and scanning electron microscopy studies have shown that adult schistosomes exposed to mefloquine were not affected immediately and only died after 24–72 h.[23] Hence, mefloquine-treated schistosomes might still be able to interact with praziquantel, resulting in increased activities of mefloquine/praziquantel combinations.

Since our *in vitro* and *in vivo* findings are encouraging, exploratory randomized open-label trials have been launched to investigate the effect of mefloquine/praziquantel combinations in schistosome-infected children. We will treat children first with mefloquine or mefloquine/artesunate (3 day regimen, in line with the common malaria treatment schedule), followed by praziquantel on day 4, since this treatment schedule achieved the highest activity *in vitro* and *in vivo*. In addition, the advantage of a spaced application of the drugs (in contrast to simultaneous administration) is that this treatment regimen does not raise regulatory and review challenges that combination products would require.[24] In a first step we will administer the standard doses (mefloquine 25 mg/kg, artesunate/mefloquine 300/250 mg and praziquantel 40 mg/kg); however, since synergistic interactions were observed in the present study *in vivo*, we are hoping in a next step to be able to lower the doses of these drugs to decrease the prevalence of adverse events and also costs. Of note, we have not included artesunate in this *in vitro* and *in vivo* study, as a preliminary experiment in our laboratory has shown similar worm burden reductions (both 86%) of mefloquine/praziquantel/artesunate (all 100 mg/kg administered simultaneously) compared with mefloquine/praziquantel (both 100 mg/kg simultaneously) in mice harbouring adult *S. mansoni*. Our result is in line with numerous experiments, which have documented a greater sensitivity of juvenile schistosomes towards the artemisinins than the adult worm in laboratory animals.[25] However, why increased cure rates have been observed with mefloquine/artesunate over mefloquine in our previous study in *S. haematobium*-infected children cannot be explained at the moment, as only moderate cure rates were observed in the group of children treated with artesunate.[11]

In conclusion, we have demonstrated that a combination of mefloquine/praziquantel reveals synergistic behaviour in the treatment of *S. mansoni*-infected mice and *in vitro*. The effect of mefloquine/praziquantel combinations on *S. haematobium* should be also studied in detail. To assess the clinical utility of this drug combination in the treatment of schistosomiasis, proof-of-concept studies in schistosome-infected children have been launched.

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Transparency declarations

None to declare.

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