

Effect of *Artemisia annua* and *Artemisia afra* tea infusions on schistosomiasis in a large clinical trial

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ABSTRACT

Background and objective: Schistosomiasis (bilharzia), a serious neglected tropical disease affecting millions, has few cost-effective treatments, so two *Artemisia* wormwood species, *A. annua* and *A. afra*, were compared with the current standard praziquantel (PZQ) treatment in an 800 patient clinical trial, August–November of 2015.

Methods: The double blind, randomized, superiority clinical trial had three treatment arms: 400 for PZQ, 200 for *A. annua*, and 200 for *A. afra*. PZQ-treated patients followed manufacturer posology. *Artemisia*-treated patients received 1 l/d of dry leaf/twig tea infusions divided into 3 aliquots daily, for 7 days with 28-day follow-up.

Results: Of 800 enrolled patients having an average of >700 *Schistosoma mansoni* eggs per fecal sample, 780 completed the trial. Within 14 days of treatment, all *Artemisia*-treated patients had no detectable eggs in fecal smears, a result sustained 28 days post treatment. Eggs in fecal smears of PZQ-treated patients were undetectable after D21. More males than females who entered the trial had melena, but both genders responded equally well to treatment; by D28 melena disappeared in all patients. In all arms, eosinophil levels declined by about 27% from D0 to D28. From D0 to D28 hemoglobin increases were greater in PZQ and *A. afra*-treated patients than in *A. annua*-treated patients. Hematocrit increases were greater from D0 to D28 for patients treated with either PZQ or *A. annua* compared to those treated with *A. afra*. Gender comparison showed that *A. afra*-treated males had significantly greater hemoglobin and hematocrit increases by D28 than either PZQ or *A. annua*-treated males. In contrast, PZQ and *A. afra*-treated females had greater hemoglobin and hematocrit increases than *A. annua*-treated females. Both adults and pediatric patients treated with *A. annua* responded better compared to PZQ treatment.

Conclusion: Both *A. annua* and *A. afra* provided faster effective treatment of schistosomiasis and should be considered for implementation on a global scale.

Abbreviations: AM, artemether; ASAQ, artesunate amodiaquine; ART, artemisinin; AS, artesunate; PZQ, praziquantel

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Background

Schistosomiasis, also known as bilharzia or snail fever, is a neglected tropical disease (NTD) caused by *Schistosoma* sp. of parasitic flatworms, and in humans especially includes *S. mansoni*, *S. haematobium*, and *S. japonicum* (WHO 2018). These parasites may infect the urinary tract or the intestines. Signs and symptoms include abdominal pain, diarrhea, bloody stool, or blood in the urine. In those with chronic infections, liver damage, kidney failure, infertility, or bladder cancer may occur. Schistosomiasis affects almost 210 million people worldwide, and an estimated 200,000 people die from it each year. Co-infection of malaria and schistosomiasis in the tropics is common (Keiser and Utzinger 2012; Moriyasu et al., 2018). With increasing migration and travel between infested areas of the world, bilharzia is even becoming a threat to more developed regions like Europe, mainly because doctors are not trained in diagnosing and treating the NTDs that are more common in the developing world (de Laval et al. 2014).

Schistosomiasis disease progression includes the presence of parasite eggs in the feces, eosinophilia, and below-normal hemoglobin and hematocrit levels. There is also often melena (black stools) resulting from hemolysis in the gastrointestinal tract. Although some traditional medicines have been reported active against bilharzia (Sheir et al., 2001; Chen et al., 2008), praziquantel (PZQ) is currently the only cost effective treatment available today. Some reports, however, suggest that PZQ is not always effective (Fallon et al., 1995; Ismail et al., 1996; Melman et al., 2009) and resistance may be emerging (Vale et al., 2017). To our knowledge, there is no other drug in development against schistosomiasis. Recently annual mass drug administration of pediatrics with PZQ + albendazole over a 5 yr period proved unsuccessful in reducing bilharzia prevalence (Sircar et al., 2018).

While PZQ is the preferred treatment, there are trials showing that when combined with an artemisinin (ART), e.g. artesunate (AS) or artemether (AM), the outcome is improved (Pérez del Villar et al. 2012). AS alone or in combination with sulfadoxine-pyrimethamine is not as effective as either PZQ or PZQ + AS. There were also anecdotal reports that *Artemisia annua* has been used to treat bilharzia. For example, in Dagana, Senegal, patients drank 3 cups of a tea infusion over 7 days with results similar to those showing efficacy against malaria. In Egypt, *A. inculca* also was reportedly used in a mouse model against bilharzia (Lotfy et al., 2006). Infected mice were orally gavaged with an ethanol extract of the plant at a dose of 800 mg/kg. Both juvenile and adult worms were reduced by 75% and 50%, respectively.

A. annua, a generally recognized as safe (GRAS) medicinal herb (Duke, 2001), also known as sweet wormwood, has anthelmintic properties. *Schistosoma* sp. rely on detoxification of heme (Skelly et al., 2014), so it is likely that the same mechanisms and constituents of *Artemisia* that also inhibit crystallization of hemozoin in malaria will be active against this parasite. How efficacious *A. annua* is against the bilharzia helminths in a controlled setting, however, remains an open question, and here we compared in human patients the efficacy of PZQ with two *Artemisia* species.

Methods

Plant material, handling and administration

Leaves and twigs of field-grown *Artemisia annua* L. and *Artemisia afra* Jacq. ex Willd. (Asteraceae) were collected in France (PAR), Senegal (SEN), Burundi (BUR), and Luxembourg (LUX) (Voucher ids are in Table 1). The harvested plant material was sun-dried, leaves and small twigs removed, and then coarsely ground by plant crusher (LUX and PAR) or by hammer mill (SEN and BUR). *A. annua* given to patients was either LUX or BUR; *A. afra* was 80% SEN : 20% PAR.

Phytochemical analysis

ART was measured using gas chromatography mass spectroscopy (GCMS) instead of HPLC to avoid ART false positives (Smith et al., 2010). Extraction and assay of artemisinic compounds, total flavonoids, phenolic acids, and essential oil components were as detailed in Weathers and Towler (2014); essential oil GCMS oven method was from Tzenkova et al. (2010). NIST ion signatures and authentic standards were used to identify compounds (Table 1). Total terpenoids were assayed spectrophotometrically using santonin as standard and a vanillin sulphuric acid method: dried plant extract in 500 µl methanol containing 2% (w/v) vanillin, plus 500 µl H₂SO₄, then incubated 20 min at 60 °C, then chilled and OD₆₀₈ read.

Study design

Based on *S. mansoni* positive stool examinations, 800 patients were selected (Fig. 1) from five sites in Maniema Province, Democratic Republic of Congo (Fig. S1) for testing, August–November of 2015. All of the selected patients verified that they had not taken any medication for treatment of *S. mansoni* infection. Patients were randomly assigned to one of three arms in this superiority trial: PZQ, *A. afra*, or *A. annua* (Fig. 1). All patients were clinically examined before and during treatment, and throughout the follow-up period. The research protocol was registered and approved by the Comité d’Ethique de l’Ecole de Santé Publique de Kinshasa (MIN.RST/SG/180/001/2016) acting for the DRC Ministry of Health. All adult patients provided signed, written consent to participate in the trial; parents signed for minor patients. The study followed guidelines of the Declaration of Helsinki and Tokyo for humans. Normally one would do a Power Analysis to determine minimum sample size. However, that requires some preliminary data to establish reasonable cutoffs. To our knowledge, no reasonable preliminary data were available, so when no previous study exists, the effect size is determined from literature review, logical assertion, and conjecture. If we want a 95 % confidence interval (p -value < 0.05) and allow a margin of error around 7% – 10%, then the ideal sample size would be around 200 (Suresh and Chandrashekar, 2012). Given that *A. annua* is GRAS and *A. afra* is an indigenous plant used for centuries to treat various ailments in Southern Africa, this approach was deemed safe and justified.

Criteria for inclusion of patients

Excluded were children under the age of six, elderly patients (over 60), pregnant women, infants, and patients with concomitant acute or severe chronic diseases. Patients with the hepatosplenic form of the disease were also excluded from the trial.

Clinical and laboratory analyses

Stool examinations for presence of parasite eggs used the Kato–Katz quantitative technique (Santos et al., 2005). Two slides were prepared from each patient on days 0, 3, 7, 14, 21, and 28, each with a clearance time of 24 h. Analyses took place at the Lubile Health Center Laboratory with a second analysis at the Kalima Reference General Hospital Laboratory. Clinical examinations for melena were performed at the Health Centers of each site. Jet-black stools that were also sticky were defined as melena. The endpoint for evaluating drug efficacy was parasitological stool examination and the absence of viable *S. mansoni* eggs in the samples over a 28-day follow-up period. Eosinophil counts, hemoglobin, and hematocrits were measured at intake and day 28 for each patient. Hemoglobin of each patient was measured at two sites: at the reference laboratory and at peripheral health centers using spectrophotometry (540 nm) and Drabkin’s Reagent, with discrepancies settled by using the reference lab value.

Table 1
Partial phytochemical composition of *Artemisia* cultivars used in this clinical trial (mg/g DW).

Phytochemical	<i>A. afra</i> PAR	SEN	1 : 4 Blend	<i>A. annua</i> LUX	BUR
Voucher id	LG0019528 Université de Liège	LG0019529 Université de Liège	Not applicable	MNHN17732 Herbarium Luxembourg	LG0019527 Université de Liège
Total terpenoids ^a and flavonoids ^b					
Total terpenoids	47.92a	31.94a	35.14	63.89x	45.14x
Total flavonoids	3.74a	3.03b	3.18	5.55x	3.84y
Artemisinin compounds					
Artemisinin	nd	0.045	0.036	1.34x	1.70y
Arteannuin B	nd	nd	nd	0.93	nd
Deoxyartemisinin	nd	nd	nd	0.32x	0.39y
Artemisinic acid	nd	nd	nd	0.86	nd
Flavonoids					
Luteolin	0.07a	0.11a	0.11	0.07	nd
Phenolic acids					
Chlorogenic acid	0.45a	2.36b	1.98	1.32x	0.09y
Rosmarinic acid	nd	nd	nd	nd	nd
Coumarins					
Scopoletin	0.10a	0.10a	0.10	0.06x	0.05x
Essential oils					
Camphor	3.26a	0.72b	1.24	0.44x	0.33y
Caryophyllene ^c	nd	nd	nd	nd	nd
Caryophyllene oxide ^c	nd	nd	nd	1.27	nd
β-pinene ^c	nd	nd	nd	nd	nd
1,8 cineole (eucalyptol)	0.47a	0.27b	0.31	0.03	nd
Borneol ^c	0.67a	0.07b	0.19	nd	nd
Spathulenol ^c	0.12	nd	0.02	nd	nd
β-neoclovene ^c	0.51a	0.13b	0.21	nd	nd
Phytol ^c	nd	nd	nd	0.40x	0.68y
Thujone ^c	nd	0.86	0.69	nd	nd

Plant cultivar origins (BUR, Burundi; LUX, Luxembourg; PAR, Paris; SEN, Senegal) had an $n \geq 4$. Significance at $p \leq 0.05$; a,b letters compare *A. afra* PAR and SEN; x,y letters compare *A. annua* LUX and BUR; nd, not detectable. Statistical analysis impossible when 1 of the 2 samples was nd.

^a Expressed as santonin equivalents.

^b Expressed as quercetin equivalents.

^c Expressed as camphor equivalents.

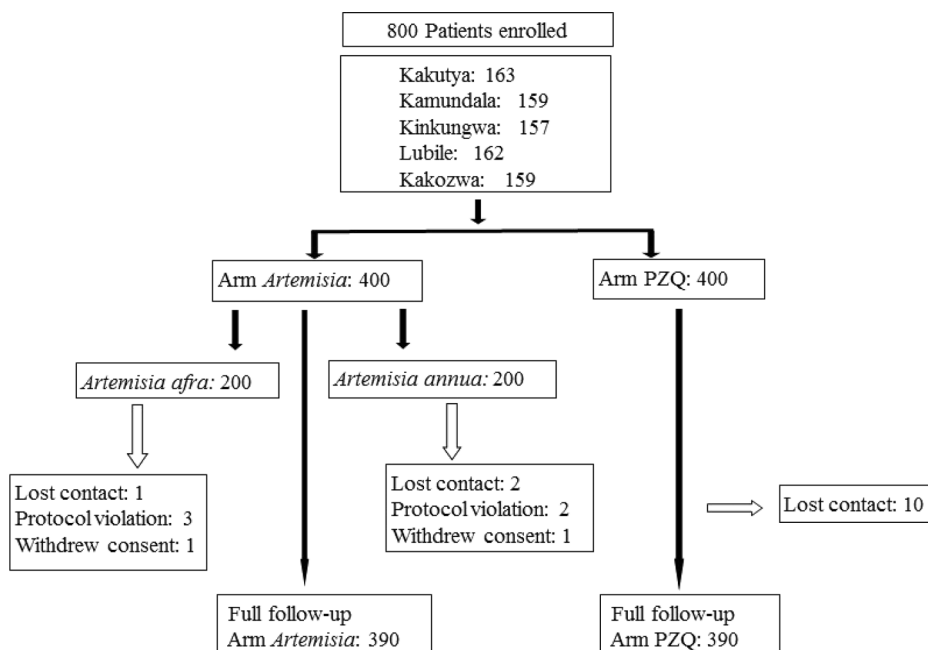


Fig. 1. Study design. PZQ, praziquantel.

Drug administration

In the *Artemisia* treatment arms, patients drank 0.33 l of *A. annua* or *A. afra* infusion 3 times daily for 7 days. Infusion was prepared as follows: 5 g dried leaves and twigs of *A. annua* or *A. afra* were added to 1 l of boiling

water, infused for 10 min, and filtered through a sterilized 1 mm mesh. For the PZQ group, patients were treated for three consecutive days with PZQ tablets (60 mg/kg daily), followed by placebo (sugar tablets) for 4 days in order to follow the 7 day treatment schedule for the *Artemisias*. The placebo tablets were identical in appearance and called A (PZQ) and C (placebo).

Table 2
Demographics of study group.

Treatment arm	Gender		Age		
	Male (%)	Female (%)	≤ 5 yr	6 – 15 yr (%)	≥ 16 yr(%)
<i>A. annua</i>	136 (63.6%)	78 (36.4%)	0	115 (53.7%)	99 (46.3%)
<i>A. afra</i>	87 (48.9%)	91 (51.1%)	0	1 (0.6%)	177 (99.4%)
PZQ	255 (65.7%)	133 (34.2%)	0	115 (29.7%)	273 (70.3%)

PZQ, praziquantel.

Patients took all medications and placebo orally, under supervision of one of the investigators. The investigators were blind as to which patients received which treatment; the identity of each group was kept in a sealed envelope. Each time patients returned during their 7 days of treatment they were observed for evaluation of adverse side effects. During treatment and through D28 all patients were advised to avoid the disease transmission areas where there was infested water, and they reported doing so. Patients were followed for 28 days post enrollment into the trial. Treatment followed WHO 2000 General Guidelines for Methodologies on Research and Evaluation of Traditional Medicine (http://apps.who.int/iris/bitstream/handle/10665/66783/WHO_EDM_TRM_2000.1.pdf;jsessionid=58426E99-AFB3D5CEDC7DF3E3686E4D87?sequence=1).

Block randomization (100 blocks, 8 patients per block) was used to allocate the three treatment arms to the patients so that the investigators did not know the nature of the medication that each patient was receiving. The envelope system was used to administer the treatment randomly. The envelope concealed the treatment that the patient would receive. Envelopes were numbered sequentially on the outside and inside was a code on each inner envelope of drug corresponding to the three treatments. The randomization list contained a pre-specified sequential number. Patients were assigned to each of the three arms as they arrived at clinic and were observed to be infected. No special selection or preference was made for assignment unless exclusion criteria were present. In that case, the patient was not assigned to the study. To double-blind the trial, *A. annua* and *A. afra* arms received the infusion plus PZQ placebo from D0–7. The PZQ arm received PZQ tablets the first three days followed by PZQ placebo on D4, 5, 6, and 7; these same patients also received *Artemisia* placebo from D0–7. PZQ placebo consisted of pill-shaped saccharose/glucose candies purchased in a pharmacy. The placebo for *Artemisia* tea infusion involved using the tea at very low dose, so *A. annua* and *A. afra* placebo teas were prepared with 0.2 g/l of plant material and PZQ patients drank placebo infusion of one *Artemisia* species or the other. Technical assistants doing parasite counts and other analyses also were unaware of arm assignments.

Concomitant treatment

Medical treatment was maintained for patients already receiving treatment for another known disease at the beginning of the trial. However, patients receiving other antimicrobials (e.g., oxamniquine, metrifonate, artemisinin derivatives) were not admitted to the trial because those medications could interfere with interpretation of the study's drug response.

Management and statistical analysis of data

Proportions were compared using the chi-square test. Continuous and normally distributed data were compared by analysis of variance (ANOVA), one-way ANOVA test, and Student's *t*Test. Data were analyzed using SPSS for Windows version 1.0.0.903 and R for Windows version 3.4.2. *P*-values < 0.05 were considered statistically significant.

Results

Primary outcome of the trial was that both *Artemisia* sp. were equally effective with PZQ in eliminating schistosome eggs from patient feces. Secondary trial outcomes were that both *Artemisia* sp. were significantly faster than PZQ in eliminating schistosome eggs from patient feces, and both *Artemisia* sp. had fewer adverse effects on patients.

Demographics of the study population

There were 780 patients who completed the trial with 115 pediatric patients in each of the PZQ and *A. annua* arms; there was 1 pediatric patient in the *A. afra* arm (Table 2). The average age was 26.1 yr in the PZQ group, 18.1 yr in the *A. annua* group, and 35.8 yr in the *A. afra* group. In the PZQ group, 65.7% were male, 34.3% were female; 29.6% were pediatric patients, and 70.4% adults. In the *A. annua* group, 63.6% were male, 36.4% were female; 53.7% were pediatric patients, and 46.3% adults. In the *A. afra* group, 48.9% were male, 51.1% were female; 0.6% were pediatric patients, and 99.4% adults.

Egg elimination from fecal samples

Patients had an average of 739, 748, and 726 eggs in their stool samples on D0 for PZQ, *A. annua*, and *A. afra*-treated patients, respectively. There was 100% elimination of eggs in stool samples in all *A. annua* and *A. afra*-treated patients within 14 days from first treatment. In contrast, PZQ patient egg elimination was not complete until after 21 days (Fig. 2). The egg count dropped by 91.2 and 90.9%, respectively, to an average of about 6 eggs per specimen by D3 for *A. annua* and *A. afra*. PZQ-treated patients at D3, however, only had a 67%

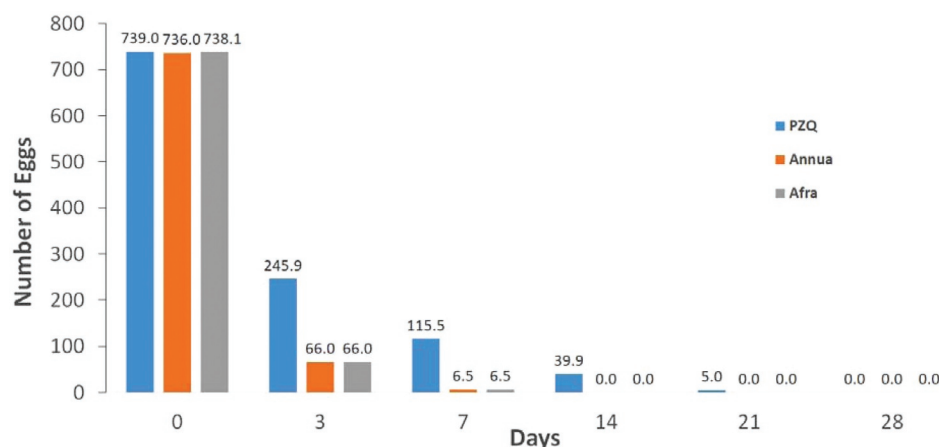


Fig. 2. Eggs disappeared faster in fecal samples and within 14 days for both *A. annua* and *A. afra*-treated patients compared to those treated with PZQ (praziquantel).

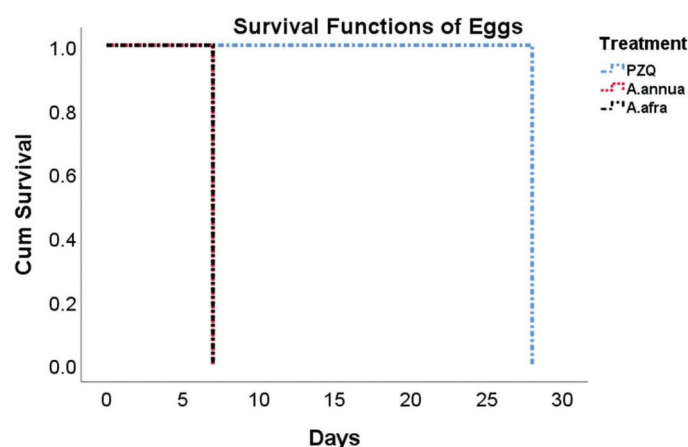


Fig. 3. Log rank analysis of cumulative egg survival after treatment with *A. annua*, *A. afra*, or PZQ (praziquantel).

decrease in fecal eggs. By D7 both *Artemisia* patient groups had < 1% of the original number of eggs while 15% of the initial egg count in PZQ patients still remained. There were no significant differences between *Artemisia* species from D3 through D28; neither were there differences between D3 and D7, and D7 and D14. However, there was a significant difference in egg count between *Artemisia* species and PZQ from D3 through D21 (Fig. 3).

Melena

At D0, 28%, 12%, and 12% of the total enrolled number of patients had melena for PZQ, *A. annua*, and *A. afra*-treated patients, respectively. By D28 no patient in any treatment arm had melena (Table 3).

Pediatric benefits

A. annua-treated pediatrics (aged 6–15) fared better with faster elimination of eggs after D3 than PZQ-treated pediatrics. Serum changes in eosinophils, hemoglobin, and hematocrit from D0 to D28 were not significantly different from changes observed in adults. Age comparisons could not be made between the two *Artemisia* species due to an inadequate number of pediatric cases treated with *A. afra*.

Serum responses

Although eosinophil levels were not statistically different among the three trial arms, when compared from D0 to D28, hemoglobin and hematocrit levels differed (Fig. 4). Hemoglobin and hematocrit levels were statistically different among all three arms ($p < 0.05$). Hemoglobin increases from D0 to D28 were 1.69, 1.78, and 1.98 g/dl for *A. annua*, PZQ, and *A. afra*, respectively. Hematocrit levels increased from D0 to D28 and were 5.3, 5.4, and 5.5% for *A. afra*, PZQ, and *A. annua*, respectively. From D0 to D28 hemoglobin increases were greater

in PZQ (95% C.I: (16.9%, 20.7%)) and *A. afra*-treated patients (95% C.I: (17.6%, 22.4%)) than in *A. annua*-treated patients (95% C.I: (15.0%, 17.7%)). Hematocrit increases were greater from D0 to D28 for patients treated with either PZQ (95% C.I: (15.9%, 18.1%)) or *A. annua* (95% C.I: (16.9%, 20.7%)) compared to those treated with *A. afra* (95% C.I: (12.5%, 16.1%)).

Gender responses

Of 403 patients who had melena at trial enrollment, there were about twice as many males (273) than females (130) (Table 3). While there was no significant difference in eosinophils among the three treatment arms, there were some significant differences in hemoglobin and hematocrit values between genders from D0 to D28 in patients treated with the two plant species. *A. afra*-treated males had greater increases in hemoglobin and hematocrit levels than either PZQ or *A. annua*-treated males (Table 3). In contrast, PZQ and *A. afra*-treated females had higher hemoglobin and hematocrit levels than *A. annua*-treated females.

Adverse effects

Vomiting, abdominal pain, and headache were the most common adverse events observed in PZQ-treated patients at 26.5%, 18.5%, and 15.5%, respectively. In the two *Artemisia* arms, no patients suffered any of those or other undesirable effects (Table S1).

Discussion

This trial showed that both *A. annua* and *A. afra* tea infusions were at least 30% faster acting against bilharzia than PZQ, but with equal efficacy at D28 for both genders. There also were fewer adverse effects on patients treated with an *Artemisia* tea infusion than with PZQ. The

Table 3
Gender responses of patients in the trial.

Gender & treatment	Overall	Patients with melena		Eosinophils (%/patient)		Hemoglobin (avg g/dl/patient)		Hematocrit (avg %/patient)	
		D0	D28	D0	D28	D0	D28	D0	D28
Male:	478	273	0	3.85	2.64	10.77	12.71	36.24	41.98
<i>A. afra</i>	87	52	0	4.08	2.67	11.01	13.26 ^a	39.20	45.03 ^a
<i>A. annua</i>	136	71	0	3.85	2.76	10.43	12.35	33.31	39.41
PZQ	255	150	0	3.78	2.57	10.88	12.71	36.80	42.31
Female:	302	130	0	3.36	2.59	11.39	12.97	38.70	43.60
<i>A. afra</i>	91	41	0	3.62	2.51	11.57	13.29 ^a	40.70	45.44 ^a
<i>A. annua</i>	78	20	0	2.86	2.63	11.43	12.72	37.73	42.17
PZQ	133	69	0	3.48	2.62	11.24	12.90 ^a	37.89	43.17 ^a

^a Significance within genders between arms from D0 to D28 ($p < 0.05$). PZQ, praziquantel.

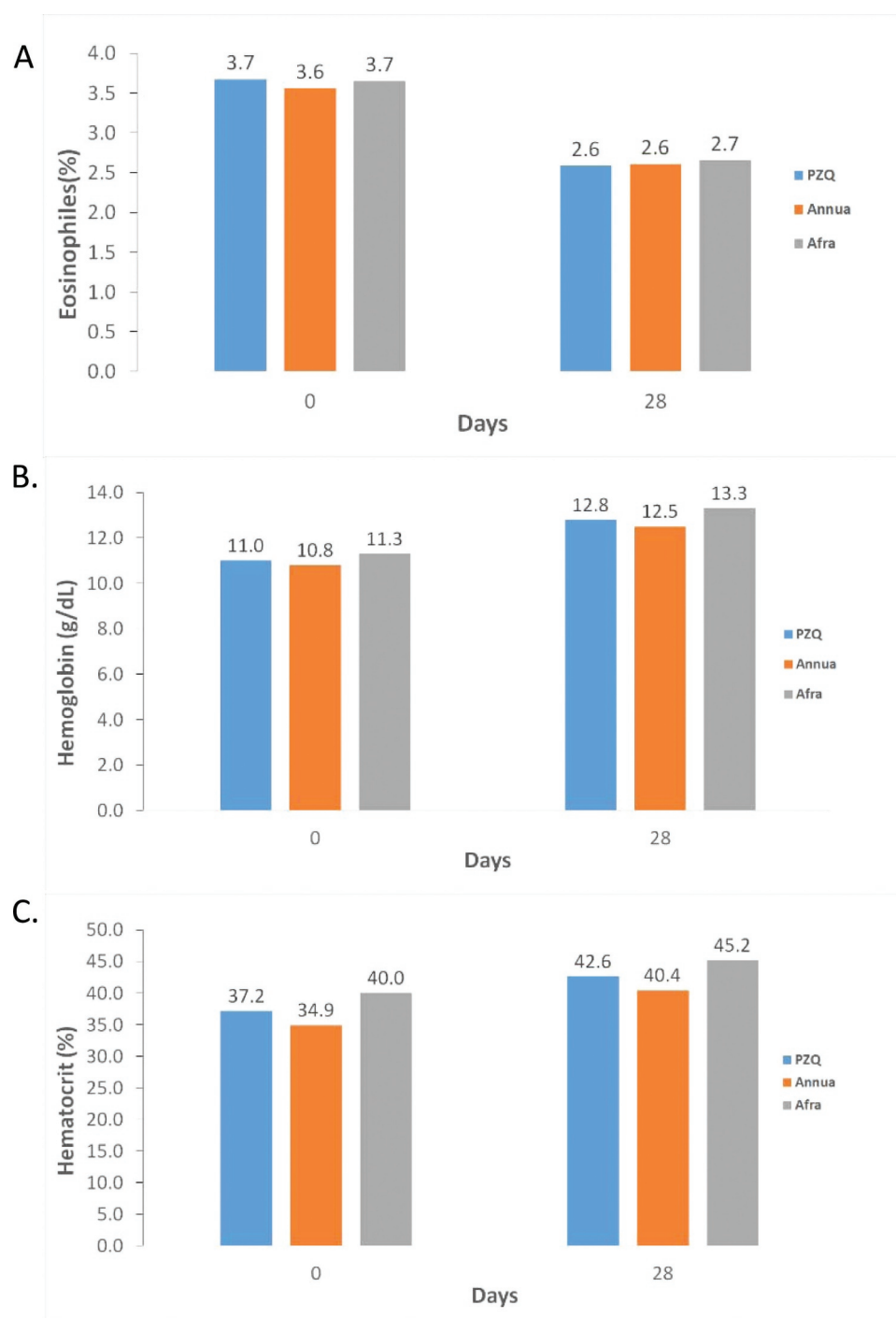


Fig. 4. Serum differences in patients treated with praziquantel (PZQ), *A. annua*, and *A. afra*. A, eosinophil (%); B, hemoglobin (g/dL); C, hematocrit (%).

ART derivatives, AM and AS, have been used in a variety of human trials against bilharzia, and in combination with PZQ. A meta-analysis of those accumulated trial data showed that combining either AM or AS with PZQ was more efficacious than the individual drugs (Pérez del Villar et al. 2012). PZQ was also more effective than an AS sulfadoxine-pyrimethamine combination. Used as a bilharzia prophylaxis, AM and AS each also provided significant protection against schistosomiasis compared to placebo (Pérez del Villar et al. 2012).

There is a trace amount of artemisinin in the *A. annua* used to prepare the placebo tea infusion. One could argue that the *A. annua* placebo tea's small amount of artemisinin (4% of the *A. annua* treatment arm level), may influence the results of the PZQ study; and the *A. afra*-

treated patients and the *A. afra* placebo had ~2.4% and ~0.1% of the *A. annua* treatment arm level, respectively. Compared to studies using only AM or AS, these amounts would be ineffective (Pérez del Villar et al. 2012) and thus for *A. afra* the elimination of schistosome eggs was unlikely from only artemisinin. It is possible, however, that small amounts of artemisinin may work synergistically with other components in the *Artemisia* sp. or even with PZQ to eliminate the parasite.

In one of the PZQ ± AM trials, Hou et al. (2008) determined that all four treatment regimens had equivalent outcomes. The four treatment arms were: A, 60 mg/kg PZQ + 6 mg/kg AM; B, 60 mg/kg PZQ + AM placebo; C, 120 mg/kg PZQ + 6 mg/kg AM; D, 120 mg/kg PZQ + AM placebo. Those results suggested that by using a single initial potent

dose of AM, the amount of PZQ could be substantially reduced. Similar studies are still needed for tea infusions to determine their best posology.

Interestingly, *A. afra* was as efficacious as *A. annua* in the rapid elimination of parasite eggs in patient feces. Furthermore, *A. afra*-treated patients had higher hemoglobin and hematocrit levels than *A. annua*-treated patients. *A. annua* has considerably more ART than *A. afra* and since some hemolysis occurs during ART treatment, hemoglobin decline is characteristic of drugs that contain ART (Kurth et al., 2016). Notably, in female PZQ patients, hematocrit and hemoglobin levels were also significantly higher than in females treated with *A. annua*, an observation not observed for males.

How then does one explain the efficacy of *A. afra* that had at most traces of ART? Studies done with *Plasmodium falciparum* showed that many other phytochemicals in *A. annua* have antimalarial activity, albeit with a higher IC₅₀ than ART (see review by Weathers et al., 2014). Similarly, there is also antischistosomal activity of many phytochemicals. For example, 17 of 20 different phytochemicals in a high throughput screening assay had an IC₅₀ between 1.3 and 37.8 μ M against the *S. mansoni* NAD⁺ catabolizing enzyme, SmNACE (Kuhn et al., 2010). Of those 17, luteolin (IC₅₀ = 8.4 μ M), myricetin (IC₅₀ = 22.0 μ M), and quercetin (IC₅₀ = 3.9 μ M) are usually present in *Artemisia* species. Thus, it is likely that the non-artemisinic phytochemicals in *A. afra* account for the efficacy of this *Artemisia* species. Future studies are needed to determine the efficacy and mechanism of action of other phytochemicals that are anti-schistosomal.

Conclusion

Although all treatment arms yielded similar outcomes 28 days after patient intake, *A. annua* and *A. afra* tea infusions given for 7 days were faster than PZQ at eliminating schistosome eggs from patient feces. *Artemisia*-treated patients also exhibited fewer adverse drug effects than PZQ-treated patients. Although posology requires further development, *A. annua* and *A. afra* tea infusions should be considered as part of the global effort to combat schistosomiasis.

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Conflict of interest

We confirm that there are no known conflicts of interest associated with this publication and there has been no significant financial support for this study that could have influenced its outcome.

Author contributions

J.M.: Designed study, oversaw clinical activities, did PCR analysis, analyzed data, edited manuscript

L.C-V.: Conceived of study, provided *A. afra* PAR and *A. annua* BUR, managed project finances, wrote and edited manuscript

M.I.: Supervisor of the study, in the health centers.

L.C.: Did statistical analyses, analyzed data, edited manuscript

P.Lutgen: Provided *A. annua* LUX, edited manuscript

C.P.: Aided study design, interpreted data

N.M.: Assisted in obtaining national research permits from the

Ministry of Scientific Research

J.B.: Coordinator of the National Malaria Program, helped with authorization and realization of the study

B.M.: Helped establish the ethical protocol of the study

P.Lalukala: Minister of Public Health of the province, helped manage the study

G.M.: Provided *A. afra* SEN, established *Artemisia* sp. herbarium vouchers, edited manuscript

D.M.: Helped interpret biological and molecular data

M.T.: Did phytochemistry of *Artemisia* sp. samples, analyzed data, edited manuscript

P.W.: Analyzed data, wrote and edited manuscript.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at [doi:10.1016/j.phymed.2018.10.014](https://doi.org/10.1016/j.phymed.2018.10.014).

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