Pharmacokinetics of praziquantel in *Schistosoma mansoni* and *Schistosoma haematobium* infected school- and preschool-aged children

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Authors no longer present at the Institution where the work was performed: Dr. Isabel Meister (Gunma University, Japan), Dr. Anna Neodo (Imperial College London, UK)
Abstract

There is a growing consensus to include preschool-aged children in preventive chemotherapy programs with praziquantel, aiming to control schistosomiasis. However, pharmacokinetic data, crucial to establish safe and effective dose for this age group, are sparse. The objective of this study was to establish and compare pharmacokinetic parameters of praziquantel in schistosomiasis infected preschool- and school-aged children.

Two pharmacokinetic trials in school- and preschool-aged children infected with *S. mansoni* or *S. haematobium*, were conducted in Côte d’Ivoire. Dried blood spot samples were taken from 492 children at ten time points following a single oral dose of 20, 40 or 60 mg/kg of praziquantel and analysed using liquid chromatography mass spectrometry. Non-compartmental analysis (NCA) was performed to obtain pharmacokinetic parameters of R- and S-praziquantel (R- and SPZQ) and R-trans-4-hydroxy-praziquantel.

No significant differences in pharmacokinetic parameters between species-specific infections were observed. While pharmacokinetic parameters differed significantly between age groups for *S. mansoni*, this trend was not observed with *S. haematobium*.

Neither area under the curve nor the maximal blood concentration presented clear dose proportionality for R and S-PZQ. Logistic regression indicated a relationship between RPZQ AUC and $C_{\text{max}}$ and probability of cure.

Praziquantel is subject to complex metabolic processes, following erratic absorption. While NCA is very informative base for a better understanding of the drug, a more targeted approach in a form of population modelling is needed to quantify the factors influencing metabolic processes and draw conclusions.

Introduction
Schistosomiasis, first described in 1851 by Theodor Bilharz (1), represents a major public health problem in rural tropical and subtropical areas of the world (2, 3). Caused by blood–dwelling flukes of the genus *Schistosoma*, *S. haematobium*, *S. japonicum* and *S. mansoni* being the principle species, it affects over 250 million people (4). Acute schistosomiasis manifests with flu–like symptoms however, if not treated, it can result in severe chronic consequences, e.g. hepatic fibrosis, kidney failure or bladder cancer (5, 6).

Despite years of research, praziquantel (PZQ) remains the only available effective drug against schistosome infections (7, 8). As a drug with relatively good efficacy and tolerability, PZQ has been successfully used in large scale drug administration programs (i.e. preventive chemotherapy) for the last decade (8, 9).

One of the most vulnerable groups, affected by schistosomiasis, are children (10, 11). Preventive chemotherapy programs target school-aged children (SAC), while their younger peers, pre-schoolers (PSAC; < 6 years), are excluded from official treatment programs and treated on individual basis only, creating the so called “treatment gap” (12, 11). Nonetheless, early parasitic infection could exacerbate the clinical impact of schistosomiasis and its subsequent morbidity (13). Additionally, PSAC might have a role in maintaining local transmission of the disease, therefore it is crucial to include them in treatment programs (13).

One of the reasons behind targeting only SAC in preventive chemotherapy programs is the lack of the pharmacokinetic (PK) data, which would guide the establishment of safe and effective dose of PZQ for PSAC (11, 14, 15). Available data on the drug’s absorption, distribution, metabolism and elimination processes (ADME) is mostly derived from studies in healthy adult volunteers, carried out years ago, while basic PK information in the target population is lacking (8, 9, 16). Since physiological and
enzyme differences between adults and children have been described in detail, the
extrapolation of adult dosages to children is uncertain (17–19). Furthermore, the only
commercially available formulation of PZQ is a racemic mixture of both enantiomers,
R- and SPZQ, resulting in a big tablet difficult for small children to swallow, while
the discussion regarding which entity is responsible for antischistosomal activity of
the drug, is ongoing (8, 16). The metabolism of both enantiomers has been studied
and there is some evidence that the main human metabolite, R-trans-4-hydroxy-
praziquantel (R-trans-4-OH-PZQ), mainly originating from enantio-selective
metabolism of RPZQ, might contribute to antischistosomal activity of PZQ (20–22).
We conducted two PK studies using dried blood spot sampling (DBS) with the aim to
elucidate PK parameters of PZQ in PSAC and SAC, infected with S. mansoni or S.
haematobium, each embedded in a single-blind, randomised, placebo controlled dose-
finding study (23, 24). For the first time, concentration over time course of all three
etities, namely RPZQ, SPZQ and the main human metabolite, R-trans-4-OH-PZQ,
were analysed. Non-compartmental analysis (NCA) was conducted in order to derive
PK parameters of the analytes and relationships between drug exposure and efficacy
were explored. Our study contributes to a better understanding of PZQ and possibly to
the development of a paediatric formulation.

Results

Method revalidation

The linearity range of calibration lines was 0.009–2.232 μg/ml for R- and SPZQ and
0.179 to 44.600 μg/ml for R-trans-4-OH-PZQ, with a coefficient of correlation (R²)
above 0.99. Results of the partial revalidation are summarised in Tables S1–S3.

S. mansoni study
2540 DBS were analysed from 229 children (94 PSAC and 135 SAC), 11 (9–11) DBS per participant. Of these, 398 (16%) measurements for RPZQ were under the lowest level of quantification (LLOQ), while 311 (12%) and 439 (17%) were under the LLOQ for SPZQ and R-trans-4-OH-PZQ, respectively. Between 29 (40 mg/kg) and 33 (60 mg/kg) PSAC and 42 (60 mg/kg) and 47 (20 mg/kg) SAC per treatment arm participated in the PK study. Both sexes were uniformly represented in each group and treatment arm (55% females for PSAC, 53% for SAC). Actual doses administered ranged from 17.0–23.3 mg/kg (median of 20.2 mg/kg) for 20 mg/kg, 35.4–42.7 (median of 39.2) mg/kg for 40 mg/kg and 56.3–64.3 (median of 60.0) mg/kg for the 60 mg/kg treatment arm. Egg reduction rates (ERRs, based on geometric mean) were 90.8%, 95.3% and 97.9% for PSAC and 83.9%, 98.3% and 99.1% for SAC (from the lowest to the highest dose of PZQ, respectively). Cure rates (CRs) ranged from 65.5% for 20 mg/kg to 78.1% for 60 mg/kg (74.1% for standard dose of 40 mg/kg), for PSAC. Among SAC, CRs were 28.9%, 68.9% and 82.9%, with increasing dosages, respectively. All the patients’ characteristics are summarised in Table 1.

**S. haematobium study**

Altogether, 2808 DBS were analysed, 11 (9–11) DBS per patient. 402 samples (14%) were under LLOQ for RPZQ, 345 (12%) for SPZQ and 474 (17%) for R-trans-4-OH-PZQ. 39 PSAC received 20 mg/kg, 43 PSAC 40 mg/kg and 41 PSAC 60 mg/kg (45, 47 and 45 SAC, respectively). The number of boys and girls was balanced between the treatment groups. Actual doses were 18.8–22.2 (median of 20.0) mg/kg, 37.5–41.4 (median of 40.0) mg/kg and 58.3–61.4 (median of 60.0) mg/kg. CRs for PSAC were 88.2%, 77.5% and 67.6% for 20, 40 and 60 mg/kg, respectively, with corresponding ERRs of 98.6%, 97.1% and 96.3%. SAC exhibited CRs of 54.6%, 72.5% and 63.4% and ERRs of 97.5%, 98.8% and 97.5% (20, 40 and 60 mg/kg,
respectively). Children’s demographics and parasitological characteristics are summarised in more details in Table 1.

Non-compartmental analysis

PK parameters are summarised in Table 2 for *S. mansoni* and in Table 3 for *S. haematobium*, presented as medians with interquartile range (IQR). Concentration-over-time profiles of R- and SPZQ and the metabolite for *S. mansoni* and *S. haematobium* SAC and PSAC are presented in Figures 1 and 2, respectively.

Overall, a great inter-patient variability was observed. T<sub>1/2</sub> could be estimated in 75 (70–81)% of subjects and was similar for all analytes across different dosages and the two age groups and parasite species, at approximately 3–8 h for R- and SPZQ and 2–5 h for the metabolite. T<sub>max</sub> was 1.5 to 6 h for the metabolite, while the enantiomers attained their highest concentration in blood at 0.5–3.0 h in the four populations of children studied.

In *S. mansoni*-infected SAC, the highest C<sub>max</sub> values were observed at 60 mg/kg for R-<i>trans</i>-4-OH-PZQ (13.57; 9.81–16.43 μg/ml), followed by SPZQ (1.00; 0.62–1.37 μg/ml) and RPZQ (0.29; 0.22–0.53 μg/ml). PSACs revealed considerably higher C<sub>max</sub> compared to SAC for all three analytes as summarised in Tables 2 and 3. In the *S. haematobium* study, in SAC, the highest values for C<sub>max</sub> at 60 mg/kg PZQ were observed for R-<i>trans</i>-4-OH-PZQ (11.49; 9.05–13.78 μg/ml), followed by SPZQ (1.25; 0.78–1.86 μg/ml) and RPZQ (0.44; 0.25–0.81 μg/ml). PSAC revealed similar C<sub>max</sub> and no differences were observed compared to PSAC for any dose or analyte.

Area under the curve (AUC) values showed considerable differences between SAC and PSAC infected with *S. mansoni* for the metabolite and RPZQ at all three dosages administered and for SPZQ at 20 and 60 mg/kg. No difference in AUC values between SAC and PSAC infected with urinary schistosomiasis, for any of the
analytes, at any dose, was observed. Logistic regression confirmed this finding—a significant effect of dose on AUC was found for children, infected with *S. mansoni*, but not for children infected with *S. haematobium*.

When comparing the AUC of cured and uncured PSAC using Mann-Whitney analysis, no differences were observed for any of the analytes and both species of the parasite, while cured SAC infected with *S. mansoni* were reaching significantly higher AUCs compared to uncured. *S. haematobium* infected SAC revealed significantly higher AUC values of RPZQ and SPZQ between cured and uncured children, while there was no significant difference in metabolite exposure (data not shown). Logistic regression indicated a positive relationship between RPZQ exposure (for both AUC and Cmax) and probability of cure, in which higher exposure led to higher probability of cure, for all children analysed together, as shown in Figures 3 and 4.

**Discussion**

Schistosomiasis remains a considerable public health problem, despite the years of efforts to control it (2, 3). PZQ is the treatment of choice and has been successfully used for decades (7, 8). While SAC are treated regularly, PSAC have been up to this day either excluded from preventive chemotherapy programs or treated off-label with the WHO recommended dose of 40 mg/kg PZQ, used for adults and SAC (11, 12, 25).

To date, several studies have been conducted describing efficacy and safety of PZQ in young children (13, 26, 27). PK of PZQ has been studied mostly in healthy adults (28–31), while ADME processes of PZQ in children, except for the recent study by Bustinduy *et al.* (16), remain largely unexplored.

We have, for the first time, quantified all three main analytes of PZQ (RPZQ, SPZQ and R-trans-4-OH-PZQ) using an intensive sampling scheme in blood. Our PK studies were embedded in two randomised controlled clinical trials allowing us to...
explore the PK of a large cohort of children treated with 3 different dosages of PZQ in both age groups of children, PSAC and SAC, infected with either \textit{S. mansoni} or \textit{S. haematobium}. The sampling technique used was novel DBS technology, which proved to be an excellent tool. It does not require medical staff or a hospital environment, which is crucial for rural settings. Furthermore, collection of blood is comparably less invasive than venepuncture, which is a benefit for sensitive populations, such as children. Adding up to these advantages are also the transport conditions- DBS do not require a cold chain and can be easily transported from the field to the laboratory (32, 33).

We did not observe pronounced differences in PK parameters between SAC and PSAC and between species-specific infections (Tables 2 and 3). While we observed significantly higher exposure for the enantiomers (R- and SPZQ) in PSAC compared to SAC, infected with \textit{S. mansoni}, the metabolite was present in significantly lower quantities. This finding could indicate slower metabolism and lower clearance in young children compared to their older peers. Interestingly, these differences were not significant in children harbouring urinary schistosomiasis.

The debate about which analyte is driving the antischistosomal activity of PZQ is still ongoing. In a recent PK study in \textit{S. mansoni}-infected children, Bustinduy \textit{et al} proposed SPZQ to be the eutomer, since it exhibited a higher and longer exposure (AUC and half-life) compared to RPZQ (16). They also suggested a correlation between AUC of PZQ, in particular of SPZQ, and CR. In our study, we observed higher AUCs for SPZQ compared to RPZQ as well, however logistic regression analysis points to a positive relationship between AUC and \( C_{\text{max}} \) of RPZQ and probability of cure for both infection species and age groups, indicating there might be a relationship between RPZQ exposure and CR. However, one should bear in mind
that the sample size our assumptions are based on is quite small. As CRs increased
with increasing dosages, we had few uncured children to sufficiently represent the
exposure of this population.

The observed interspecies differences in susceptibility to PZQ enantiomers and their
impact on efficacy of the drug in human, is an important issue. We recently showed
that SPZQ had a higher and significant efficacy against S. haematobium compared to
S. mansoni both in vitro and in vivo. It might therefore contribute to the overall
activity of PZQ on S. haematobium. R-trans-4-OH-PZQ also showed signs of
antischistosomal activity in vitro towards both species of the parasite, more
pronounced in case of S. haematobium (21). Its contribution to the activity of RPZQ
could explain a higher sensitivity of S. haematobium to PZQ within in vitro/in vivo
frame, however in our studies we did not observe any correlation between exposure of
the metabolite and probability of cure.

In conclusion, we have described PK processes of PZQ in a pediatric population using
three different dosages and comparing the two most prevalent species of Schistosoma
spp. Based on our findings, the species of schistosome infection (S. mansoni versus S.
haematobium) does not play a role in influencing PK parameters of PZQ. Age seems
to have an effect however no clear pattern has been observed. While SPZQ is present
in higher quantities than RPZQ, it did not show a relationship with CRs, while logistic
regression indicated a positive relationship between RPZQ exposure and cure.

Population pharmacokinetics studies with RPZQ are currently on going to study the
patterns and eventual relationships of patients’ characteristics (e.g. weight, co-
infections) with PK parameters in more detail.

Methods and materials

Chemicals and reagents
R- and SPZQ and R-trans-4-OH-PZQ were kindly provided by Merck KgaA (Darmstadt, Germany). Eleven-fold deuterated PZQ (PZQd11), acquired from Toronto Research Chemicals (Ontario, Canada), was used as an internal standard (IS). Acetonitrile, ammonium formate, ammonium acetate and formic acid of MS grade were purchased from Sigma-Aldrich (Buchs, Switzerland). Ultrapure water was obtained using a Millipore Milli-Q water purification system (Merck Millipore, MA, USA). Human blood was supplied by the local blood donation centre (Basel, Switzerland). PZQ tablets (Cesol™, 600 mg) were donated by Merck KgaA (Darmstadt, Germany).

**LC–MS/MS equipment**

A 6460 Series triple quadrupole liquid chromatography–mass spectrometry, LC-MS/MS, (Agilent Technologies, Basel, Switzerland) was used to perform all the measurements. The LC module consisted of a 1290 series binary pump (G1312B), followed by 1200 Series Micro Vacuum (G1379B) degasser, Agilent 1260 Infinity High Performance autosampler (G1367E), equipped with a 12900 Infinity series Thermostat (G1330B) and electrospray ionization source (G1958–65138). MS/MS analyses were performed in positive ionization mode. Mass Hunter Workstation software B.06.00 (Agilent Technologies, Basel, Switzerland) served to operate the instrument and analyse the data.

**LC–MS/MS method and partial validation**

The LC-MS/MS method was adapted from a recently validated method by Meister et al (34). Briefly, the compounds of interest were primarily separated from remaining matrix by a column trapping system (HALO C–18, 4.6 × 5 mm, Optimize Technologies, OR, USA), to minimize instrument contamination. 10 mM ammonium acetate with 0.015% formic acid in water (mobile phase A) at a flow rate of 0.3
ml/min served as a loading solution. After 1.00 minute of loading, a mixture of 20 mM ammonium acetate and acetonitrile (1:4, mobile phase B) was used to elute the analytes from the trapping to the chiral column (Lux Cellulose–2 (150x4.6 mm, 3 µm, Phenomenex, CA, USA)), with a flow rate ascending to reach 0.4 ml/min at 3 minutes and remaining steady until 9.49 min. In the last minute (9.50–10.50), mobile phase A was used again to re-equilibrate the trapping column at a flow rate of 1.0 ml/min. LC-MS/MS parameters are summarised in Table S4 and chromatograms are depicted in Figures S1–S2.

Partial revalidation was performed to ensure compliance with FDA regulations, since the LC-MS/MS method (34) was transferred to another system.

In brief, selectivity was assured by analysing 18 blank DBS extracts compared to double blank (pure extraction solvent without IS) DBS samples to exclude potential endogenous substances interference. Calibration lines (CL) were plotted as analyte peak area (normalised to IS) vs. concentration and fitted using linear regression (Figure S3). The suitable weighting factor (1/x^2) was chosen to result in the minimal total error. Accuracy and precision were determined using quality control (QC) samples with known analyte concentration, 6 replicates of four concentration levels across the linearity range (LLOQ, low, middle and high concentrations). By comparing measured to nominal concentration (in percentage), accuracy was calculated. Inter- and intra-batch precision was analysed by measuring 3 batches of samples per day on 3 different days. Precision of +/- 15% (+/- 20% at LLOQ) was considered adequate, while the acceptable accuracy ranged from 85–115 % (80–120% at LLOQ). Possible enhancing or suppressive matrix effects were evaluated by comparing the signal of biological matrix (DBS extract) and organic solvent, both spiked with analytes. The stability of all entities has been reported elsewhere (28, 34).
Quality control and standard preparation

The preparation of the solutions was adapted from Meister et al (34). Briefly, stock solutions of the analytes were prepared freshly in acetonitrile to obtain 1.0 mg/ml concentrations for R- and SPZQ, 5.0 mg/ml for R-trans-4-OH-PZQ and 1.25 mg/ml for PZQd11. A working solution of 53.0 μg/ml PZQ and 1071.0 μg/ml trans-4-OH-PZQ was used to prepare fresh CL and QC samples and diluted with acetonitrile to reach a range of 0.2–53.0 for PZQ and 4.0–1071.0 μg/ml for R-trans-4-OH-PZQ. The IS solution was diluted 4:1 (v/v) with water to produce extraction solvent.

CL and QC samples were prepared freshly for every analytical run. For the CL, blank human blood was spiked with the mixture of analytes to reach a final concentration range of 2.232 to 0.009 (LLOQ) μg/ml for R- and SPZQ, and of 44.6 to 0.179 (LLOQ) μg/ml for R-trans-4-OH-PZQ. For the QC, 6 samples of high, medium, low and LLOQ concentrations were prepared similarly and spotted on the DBS cards (903 Protein Saver Snap Apart Cards®, Whatman, UK). Disks of 5 mm in diameter were punched from DBS samples and extracted with 200 μl of extraction solvent.

Study design and ethical considerations

Ethical clearance was obtained by the Ethics Committee of North-western and Central Switzerland (EKNZ 162/2014) and the Ministère de la Santé et de l’Hygiène Publique in Côte d’Ivoire (CNER, 037/MSLS/CNER-dnk). The trial was registered as International Standard Randomised Controlled Trial (ISRCTN15280205). The S. mansoni study was carried out in the Azaguié region of Côte d’Ivoire between November 2014 and February 2015 (23). S. haematobium PK study was implemented in Azaguié, Côte d’Ivoire between November 2015 and January 2016 (24).

94 PSAC (age 2–5 years) and 135 SAC (age 6–15 years) with confirmed S. mansoni infection using duplicate Kato-Katz method were included in the PK study.
PSAC and 137 SAC infected with *S. haematobium* (diagnosed using the urine filtration method) participated in the second PK trial. All children were stratified according to infection intensity and randomised to receive 20, 40 or 60 mg/kg PZQ or placebo (data not shown). Randomisation, masking, field and laboratory procedures have been presented elsewhere (23, 24). PZQ tablets (600 mg Cesol™) were administered according to the calculated dose per kilogram of body weight in half (*S. mansoni*) and quarter (*S. haematobium*) tablet increments. Since the bioavailability of PZQ is known to be influenced by food (8, 35), the treatment was administered after a standardised breakfast. For PSAC, the tablets were crushed and the powder was suspended in a mixture of sugar syrup and water to mask the taste.

**DBS samples collection**

Capillary blood (+/− 0.1 ml) was obtained using a finger pricker (e.g. Accu-check Softclix Pro®; Roche, Switzerland) at 0:00, 0:30, 1:00, 1:30, 2:00, 2:30, 3:00, 6:00, 8:00, 10:00 and 24:00 hours after treatment with PZQ. Four drops of blood at each time point were transferred on the DBS cards, dried for approximately 1 h and stored afterwards in plastic bags with desiccant. The cards were transferred to Basel and kept at −80°C.

**Non-compartmental analysis**

The following PK parameters of RPZQ, SPZQ and *R*-trans-4-OH-PZQ were obtained using the Winnonlin software (version 5.2; Certara, Princeton, NY, USA):

- \( C_{\text{max}} \): maximal blood concentration (µg/ml)
- \( T_{\text{max}} \): time needed to reach \( C_{\text{max}} \) (h)
- \( \text{AUC}_{\text{last}} \): area under the curve between 0 and the last positive concentration (h*µg/ml)
- \( T_{1/2} \): terminal half-life; time in which half of the absorbed drug is eliminated (h)
C\text{max} and T\text{max} are observed parameters, while T_{1/2} was calculated as \( T_{1/2} = \ln 2/\lambda \). Constant of elimination (\( \lambda \)) was determined by the program using non-linear regression of the natural logarithm of concentration values in the elimination phase. Since the absorption of PZQ is erratic, half-life was only estimated for those patients who had a single peak in concentration and where the elimination phase was well estimated by the algorithm. AUC_{last} (AUC) was calculated from 0 to the last quantifiable positive concentration, using linear trapezoidal rule. Volume of distribution and renal clearance were not reported, since bioavailability is needed to estimate them adequately. PK parameters were estimated for each study participant and the median and IQR were calculated for patients of each treatment arm. AUC and C\text{max} values of both R- and SPZQ were compared between cured and uncured patients. CR was defined as the percentage of patients who were positive for infection at the baseline but were not excreting eggs at the follow up. ERR was expressed as the geometric mean egg output after treatment divided by the geometric mean egg output before treatment (36).

To compare the PK parameters, statistical analysis was conducted using Prism (version 7.03, GraphPad, CA, USA). Mann-Whitney or Kruskal-Wallis multiple comparison statistical test, depending on the number of groups being compared, were used to study parameters of children in different treatment arms, age groups and parasite species. A P value of <0.05 was considered as statistically significant. A logistic regression was employed to characterize exposure-response relationship for RPZQ, the proposed active enantiomer, using 0.05 level of significance, with the following parameters: AUC, C\text{max}, dose (treated as categorical and as continuous measure) and infection type.
Acknowledgements

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We would like to thank all the participating children and their guardians in the villages of Azaguié region, Côte d’Ivoire. We are grateful to Prof. Dr. Jörg Huwyler for continuous support and Dr. Peter Bonate for the help with the statistical analysis.

Transparency declarations

None to declare.

References


16. Bustinduy AL, Waterhouse D, de Sousa-Figueiredo JC, Roberts SA,


Table 1: Patient characteristics.

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<td>33</td>
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<td>No. of girls</td>
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<td>19.6 (6-168)</td>
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<td>37.5 (6-828)</td>
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Schistosoma mansoni

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<td>No. of children</td>
<td>39</td>
<td>43</td>
<td>41</td>
<td>45</td>
<td>47</td>
<td>45</td>
</tr>
<tr>
<td>No. of girls</td>
<td>23</td>
<td>20</td>
<td>26</td>
<td>25</td>
<td>25</td>
<td>26</td>
</tr>
<tr>
<td>Age (years)*</td>
<td>3.7 (2-5)</td>
<td>3.9 (2-5)</td>
<td>4.0 (2-5)</td>
<td>8.8 (6-15)</td>
<td>9.0 (6-14)</td>
<td>9.2 (6-15)</td>
</tr>
<tr>
<td>Weight (kg)*</td>
<td>14.9 (±2.8)</td>
<td>15.0 (±2.6)</td>
<td>15.2 (±2.1)</td>
<td>24.3 (±5.2)</td>
<td>25.0 (±6.2)</td>
<td>25.3 (±8.0)</td>
</tr>
<tr>
<td>No. of children absent at follow-up</td>
<td>5</td>
<td>3</td>
<td>3</td>
<td>1</td>
<td>7</td>
<td>4</td>
</tr>
<tr>
<td>Geometric mean no. of eggs/ml of urine (min-max)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Before treatment</td>
<td>6.9 (1-63)</td>
<td>6.8 (1-145)</td>
<td>8.0 (0.3-72)</td>
<td>20.4 (1-2317)</td>
<td>16.6 (1-237)</td>
<td>15.7 (1-223)</td>
</tr>
<tr>
<td>After treatment</td>
<td>0.1 (0-7)</td>
<td>0.2 (0-5)</td>
<td>0.3 (0-27)</td>
<td>0.5 (0-14)</td>
<td>0.2 (0-5)</td>
<td>0.4 (0-20)</td>
</tr>
<tr>
<td>Egg reduction rate - %</td>
<td>98.6</td>
<td>97.1</td>
<td>96.3</td>
<td>97.5</td>
<td>98.8</td>
<td>97.5</td>
</tr>
<tr>
<td>No. of children positive for infection</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Before treatment</td>
<td>34</td>
<td>40</td>
<td>37</td>
<td>44</td>
<td>40</td>
<td>41</td>
</tr>
<tr>
<td>After treatment</td>
<td>4</td>
<td>9</td>
<td>12</td>
<td>20</td>
<td>11</td>
<td>15</td>
</tr>
<tr>
<td>Cure rate - %</td>
<td>88.2</td>
<td>77.5</td>
<td>67.6</td>
<td>54.6</td>
<td>72.5</td>
<td>63.4</td>
</tr>
</tbody>
</table>

Schistosoma haematobium

*Mean (min-max)
*Mean ±SD.
*Presented by Coulibaly et al.(23)
Table 2: PK parameters of SAC and PSAC infected with *S. mansoni*

<table>
<thead>
<tr>
<th>S. mansoni</th>
<th>R-trans-4-OH-PZQ</th>
<th>RPZQ</th>
<th>SPZQ</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>20 mg/kg</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SAC</td>
<td>T$_{1/2}$ (h)</td>
<td>2.73 (2.24–3.46)</td>
<td>3.12 (2.15–5.97)</td>
</tr>
<tr>
<td></td>
<td>Tmax (h)</td>
<td>2.0 (2.0–2.5)</td>
<td>1.0 (0.5–1.5)</td>
</tr>
<tr>
<td></td>
<td>C$_{max}$ (μg/ml)</td>
<td>6.43 (3.94–9.53)$^{a,b}$</td>
<td>0.07 (0.05–0.14)$^{a,b}$</td>
</tr>
<tr>
<td></td>
<td>AUC$_{max}$ (h*μg/ml)</td>
<td>29.87 (19.60–43.11)$^{a,b}$</td>
<td>0.27 (0.16–0.46)$^{a,b}$</td>
</tr>
<tr>
<td>PSAC</td>
<td>T$_{1/2}$ (h)</td>
<td>2.88 (2.24–3.81)</td>
<td>5.02 (3.96–6.70)</td>
</tr>
<tr>
<td></td>
<td>Tmax (h)</td>
<td>2.50 (1.50–3.00)</td>
<td>1.50 (1.00–2.50)</td>
</tr>
<tr>
<td></td>
<td>C$_{max}$ (μg/ml)</td>
<td>3.24 (2.26–7.34)$^{a,b}$</td>
<td>0.33 (0.15–0.60)$^{a,b}$</td>
</tr>
<tr>
<td></td>
<td>AUC$_{max}$ (h*μg/ml)</td>
<td>16.84 (11.52–33.28)$^{a,b}$</td>
<td>1.35 (0.74–2.86)$^{a,b}$</td>
</tr>
<tr>
<td><strong>40 mg/kg</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SAC</td>
<td>T$_{1/2}$ (h)</td>
<td>3.09 (2.66–3.82)</td>
<td>3.91 (1.86–6.57)</td>
</tr>
<tr>
<td></td>
<td>Tmax (h)</td>
<td>2.5 (2.5–3.0)</td>
<td>1.50 (1.00–2.00)</td>
</tr>
<tr>
<td></td>
<td>C$_{max}$ (μg/ml)</td>
<td>10.32 (8.21–14.17)$^{a,b}$</td>
<td>0.25 (0.15–0.37)$^{a}$</td>
</tr>
<tr>
<td></td>
<td>AUC$_{max}$ (h<em>μg/ml)$^</em>$</td>
<td>56.59 (46.18–81.85)$^{a,b}$</td>
<td>0.78 (0.57–1.08)$^{a,b}$</td>
</tr>
<tr>
<td>PSAC</td>
<td>T$_{1/2}$ (h)</td>
<td>3.42 (2.68–3.80)</td>
<td>5.65 (3.70–7.80)</td>
</tr>
<tr>
<td></td>
<td>Tmax (h)</td>
<td>2.00 (1.50–3.00)</td>
<td>1.00 (0.50–2.00)</td>
</tr>
<tr>
<td></td>
<td>C$_{max}$ (μg/ml)</td>
<td>6.92 (4.08–10.16)$^{a}$</td>
<td>0.49 (0.36–0.93)$^{a}$</td>
</tr>
<tr>
<td></td>
<td>AUC$_{max}$ (h<em>μg/ml)$^</em>$</td>
<td>32.74 (21.76–50.42)$^{a}$</td>
<td>1.71 (1.15–2.73)$^{a}$</td>
</tr>
<tr>
<td><strong>60 mg/kg</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SAC</td>
<td>T$_{1/2}$ (h)</td>
<td>3.19 (2.96–4.40)</td>
<td>4.49 (2.00–7.82)</td>
</tr>
<tr>
<td></td>
<td>Tmax (h)</td>
<td>3.00 (2.50–6.00)</td>
<td>1.50 (1.00–2.00)</td>
</tr>
<tr>
<td></td>
<td>C$_{max}$ (μg/ml)</td>
<td>13.57 (9.81–16.43)$^{a,b}$</td>
<td>0.29 (0.22–0.53)$^{a,b}$</td>
</tr>
<tr>
<td></td>
<td>AUC$_{max}$ (h<em>μg/ml)$^</em>$</td>
<td>89.22 (61.98–145.97)$^{a}$</td>
<td>1.00 (0.83–1.93)$^{a,b}$</td>
</tr>
<tr>
<td>PSAC</td>
<td>T$_{1/2}$ (h)</td>
<td>3.54 (3.00–4.41)</td>
<td>5.95 (4.42–8.75)</td>
</tr>
<tr>
<td></td>
<td>Tmax (h)</td>
<td>2.50 (1.50–3.00)</td>
<td>1.50 (1.00–2.00)</td>
</tr>
<tr>
<td></td>
<td>C$_{max}$ (μg/ml)</td>
<td>8.29 (5.69–11.85)$^{a,b}$</td>
<td>0.69 (0.43–1.25)$^{a,b}$</td>
</tr>
<tr>
<td></td>
<td>AUC$_{max}$ (h<em>μg/ml)$^</em>$</td>
<td>49.94 (28.57–75.12)$^{a,b}$</td>
<td>2.74 (1.71–4.03)$^{a,b}$</td>
</tr>
</tbody>
</table>

*Significant difference between SAC and PSAC of the same dose and analysis. Significant difference between 20 and 60 mg/kg of the same age group and analysis.

---

487 Significant difference between SAC and PSAC of the same dose and analysis. Significant difference between 20 and 60 mg/kg of the same age group and analysis.

488 Significant difference between R and SPZQ of the same dose, age group, and species. Significant difference between the doses of 20 and 40 mg/kg of the same age group and species of the parasite.

489 $^*$T$_{1/2}$ could be estimated in 75 (70–81)% (median (QR)) of the subjects.
Table 3: PK parameters of SAC and PSAC infected with *S. haematobium*

<table>
<thead>
<tr>
<th>S. haematobium</th>
<th>R-trans-4-OH-PZQ</th>
<th>RPZQ</th>
<th>SPZQ</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>SAC</td>
<td>PSAC</td>
<td>SAC</td>
</tr>
<tr>
<td>20 mg/kg</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>T</em>_{max} (h)</td>
<td>3.28 (2.54-4.78)</td>
<td>3.49 (2.79-5.21)</td>
<td>4.41 (3.52-9.36)</td>
</tr>
<tr>
<td><em>C</em>_{max} (μg/ml)</td>
<td>2.50 (1.50-3.00)</td>
<td>2.00 (1.50-3.00)</td>
<td>2.50 (1.50-6.00)</td>
</tr>
<tr>
<td><em>AUC</em>_{max} (h*μg/ml)</td>
<td>4.64 (4.08-5.84)^+</td>
<td>4.92 (3.92-7.73)^+</td>
<td>0.18 (0.09-0.35)^+</td>
</tr>
<tr>
<td>40 mg/kg</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>T</em>_{max} (h)</td>
<td>3.70 (3.18-4.52)</td>
<td>3.71 (2.92-4.71)</td>
<td>4.33 (2.37-8.20)</td>
</tr>
<tr>
<td><em>C</em>_{max} (μg/ml)</td>
<td>3.00 (2.25-6.00)</td>
<td>3.00 (1.75-3.00)</td>
<td>2.00 (1.50-3.00)</td>
</tr>
<tr>
<td><em>AUC</em>_{max} (h*μg/ml)</td>
<td>8.16 (6.44-10.16)^+</td>
<td>8.14 (6.18-11.47)^+</td>
<td>0.32 (0.16-0.62)^+</td>
</tr>
<tr>
<td>60 mg/kg</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>T</em>_{max} (h)</td>
<td>3.86 (3.39-4.55)</td>
<td>3.64 (3.03-4.09)</td>
<td>5.34 (3.30-7.90)</td>
</tr>
<tr>
<td><em>C</em>_{max} (μg/ml)</td>
<td>4.50 (2.88-6.00)</td>
<td>3.00 (2.00-3.00)</td>
<td>2.50 (1.00-3.00)</td>
</tr>
<tr>
<td><em>AUC</em>_{max} (h*μg/ml)</td>
<td>11.49 (9.05-13.78)^+</td>
<td>11.49 (10.65-15.09)^+</td>
<td>0.44 (0.25-0.81)^+</td>
</tr>
</tbody>
</table>

Significant difference between SAC and PSAC: ^+^, ^a^, ^b^, ^c^,
Significant difference between R and SPZQ of same dose, age group and species: ^+^, ^a^, ^b^, ^c^,
Significant difference between the doses of 20 and 40 mg/kg (\(^\ast\)) or 20 and 60 mg/kg (\(^\ast\)) for same age group, analyte and species of the parasite. *T*_{max} could be estimated in 75 (70-81)% (median (IQR)) of the subjects.
Figure 1: Concentration over time profiles (mean with SD as a shaded area) for increasing doses of all analytes in *S. mansoni* infected children.

**SAC S. mansoni**

- A. R-trans-4-OH-PZQ
- B. RPZQ
- C. SPZQ

**PSAC S. mansoni**

- D. R-trans-4-OH-PZQ
- E. RPZQ
- F. SPZQ
Figure 2: Concentration over time profiles (mean with SD as a shaded area) for increasing doses of all analytes in *S. haematobium* infected children.
Figure 3: AUC versus probability of cure for RPZQ and all groups of children

Figure 4: C_{max} versus probability of cure for RPZQ and all groups of children