THE ANTI-SCHISTOSOMAL ACTIVITY OF *BOSWELLA DAZIELII* IN THE TREATMENT OF SCHISTOSOMIASIS

Rwang Pam Gyang

Abstract

Nine batches of laboratory mice each consisting of five replicates were infected with 200 cercariae of the helminth parasite per mouse, using the paddling technique. Mice in eight batches were subsequently treated with the plant extract at weekly intervals while the 9th batch was kept as infected but untreated control. A single oral dose of 40mg/kg body weight was given. The efficacy of the extract against different ages of development of the disease was measured by mean percentage worm reduction, mean percentage reduction in tissue egg count by a qualitative evaluation of hepatosplenic disease. The parasite was highly susceptible to the extracts administered on days 0, 7, 14, 42 and 49 post-infection but the helminth parasite was not controlled by treatment on days 21, 28 and 35. While mice treated on days 0, 7 and 14 showed cure with normal livers and spleens, those treated on days 42 and 49 showed cure but had hepatosplenic disease comparable to those of mice in untreated control and the mice treated on days 21, 28 and 35 where the extract was ineffective. Cure rate, given by percentage worm reduction, ranged from 96-98%. As a result of this, I am recommending that drugs from organic source should be encouraged since they are effective in the treatment of this disease.

Introduction

After malaria, Schistosomiasis is the second prevalent tropical parasitic disease in the World. As a result, much effort has been put for their control. Although several methods are available for the control of the disease and candidate vaccines have been tested in the past decade, none is commercially available currently (Waine and Mcmanus, 1997). Hence, chemotherapy remains one of the main approaches to control the disease (Wu et al, 1993). At the moment praziquantel (Pzq) is the drug of choice for the treatment of the disease. However, Johansen et al (1996) found severe liver lesions attributable to dead worms after praziquantel treatment of goats infected with *Schistosoma bovis*.

Aside from the minor side effect caused by parziquantel, another problem is the development of resistance to drug. Concerned about this negative development, it has become imperative to seek for new drugs that may be effective, safe and environmentally friendly. Also, praziquantel (the drug of choice) is an inorganic drug whose continued excretion into the environment may be harmful. With the current global concern about the safety of the environment, it would be advantageous to have drugs developed from organic sources, especially those of plant origin. From the foregoing, it therefore, becomes necessary to investigate the efficacy of *Boswellia dazielli*.

Methodology

Procedure Involving Plant Collection of Plant Material

Flic plant *Boswellia dazielli* was chosen following literature searches, which have been previously identified by Audu (1989), the plant was collected from Bauchi area with the aid of local herbalists.

Preparation of Plant Material for Extraction

Only plant part (leaves) specified for the traditional usage was collected and shade dried. The dry part was pounded to powder in a wooden mortar and stored in labeled containers.
Extraction of Crude Plant Extract

Equal quantities of the powdered plant material in separate containers were mixed with distilled water and Methanol solution each at a ratio of 1:5 weigh per volume (W/V) according to the method described by Ibrahim et al (1984), and Kela et al (1989 a, b) each in one litre pyrex conical flask. This was allowed to stand for 48 hours at ambient temperature, and the suspension filtered first through two layers of thin cloth inserted in 2.0mm mesh and then through a 0.32mm mesh gauze. The filtrate was then concentrated to dryness in vacuo at 60°C using a rotary evaporator coupled to Thermo-regulator. Extract obtained were scraped and stored in a refrigerator at 4°C in labeled specimen bottles.

Parasite
Collection of Urine and Stool Specimens

Urine and stool specimens were collected from Miri Primary School pupils, Pathology Department of the General Hospital Bayara and Specialist Hospital Bauchi.

Treatment of Urine and Stool Samples and the Infection of Snails

The snails, the intermediate host (adult Bulimi and Biomphalaria spp) infected with the parasite following method described by Standen (1952); Webbe and James (1971); Frandsen (1975). Infected samples were introduced into the aquarium where the snails are living until they became infected as they continue to interact in the infected environment/aquarium.

Animals
Experimental Mice

Six-weeks old laboratory mice were collected from National Veterinary Research Institute (NVRI), Vom, Plateau State, Nigeria. Mice were maintained on balanced mouse pellets.

Experimental Procedure Infection of the Final Host

When the infected snails after the incubation period, were exposed to strong electric light in small containers, the shedding of cercariae started. These cercariae which has a maximum survival time of 48 hours were then collected and used to infect the mice. The infection was through a very simple method (routinely at Danish Bilhaziasis Laboratory and many other laboratories) called paddling method, whereby the mice were put into water containing cercariae for 30 mins (Christensen et al, 1979).

Administration of Plant Extract

There were eight experimental cages, each contained five mice replicates. One cage/week was selected and the mice therein treated with the extract. The first cage selected for treatment had mice treated on day zero. The rest of the experimental cage were treated on days 7,14,21,28,35,42 and 49. The extract was applied at rate of 40mg/kg, thus, mice were weighed individually to determine the amount of extract to administer. Dose for each mouse was dissolved in pre-determined volume of water (0.3ml). The volume represents what a mouse would readily take after starving it of water for 24 hours. All treated mice were therefore starved of water prior to extract treatment. Control mice were not treated. The last cage was treated on the 49th day and after 56th day when eggs were first observed in the faeces of the mice, recording of data was started.

Effectiveness of the extract was determined on the basis of percentage reduction in worm yield, percentage reduction in tissue egg count in treated mice compared with untreated/infected control. Method for quantifying the parameters are well documented by Christensen et al (1984) for reduction on worm yield and tissue egg count.

Students t-test was applied to determine the significance or otherwise of differences between corresponding mean values of experimental and control groups.

Results

For all the parameters considered, Mice treated on days 0,7,14,42 and 49 were highly susceptible to treatment with the plant extract at the dose of 40 mg/kg. On the other hand those treated on days 21, 28, and 35 were not susceptible. The mean worm recovery and the percentage worm reduction were shown in Table 1. The mice treated on days 0,7,14,42 and 49 had significant decline in mean worm recover compared to the control at P < 0.05 significant level. Those
treated on the other days had comparable mean worm recovery with the control. The percentage cure rate given by the percentage reduction mean worm recovery showed that the extract treatment achieved 96-98% cure based on data for days 0, 7, 14, 42 and 49.

Table 1: Mean Recovery of Adult Worm from Mice Infected with Cercariae and Treated at Weekly Interval

<table>
<thead>
<tr>
<th>Treatment scheduled (days) after infection</th>
<th>Worm burden</th>
<th>Percentage worm recovery</th>
<th>Percentage cure rate over control</th>
<th>t-cal.</th>
<th>Students’ t-test</th>
<th>Remark</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infected/untreated control</td>
<td>Infected</td>
<td>Untreated</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>28</td>
<td>56</td>
<td>1</td>
<td>96</td>
<td>96</td>
<td>P &lt;0.05 Sig.</td>
</tr>
<tr>
<td>7</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>96</td>
<td>96</td>
<td>P &lt;0.05 Sig.</td>
</tr>
<tr>
<td>14</td>
<td>1</td>
<td>1</td>
<td>0.5</td>
<td>98</td>
<td>98</td>
<td>P &lt; 0.05 Sig.</td>
</tr>
<tr>
<td>21</td>
<td>46</td>
<td>48</td>
<td>23</td>
<td>17.86</td>
<td>17.86</td>
<td>ns</td>
</tr>
<tr>
<td>28</td>
<td>48</td>
<td>48</td>
<td>24</td>
<td>14.29</td>
<td>14.29</td>
<td>ns</td>
</tr>
<tr>
<td>35</td>
<td>40</td>
<td>40</td>
<td>20</td>
<td>28.57</td>
<td>28.57</td>
<td>ns</td>
</tr>
<tr>
<td>42</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>96</td>
<td>96</td>
<td>P &lt;0.05 Sig.</td>
</tr>
<tr>
<td>49</td>
<td>1</td>
<td>1</td>
<td>0.5</td>
<td>98</td>
<td>98</td>
<td>P &lt; 0.05 Sig.</td>
</tr>
</tbody>
</table>

Means for each treatment were compared with control at P<0.05 Sig. = Significant.
Ns = Not significant.

Table 2 shows the tissue egg distribution in different segments of the gastrointestinal tract (GIT) and the associated organs. The percentage reduction in tissue egg count revealed that there was remarkable decline in mean tissue egg for mice treated on days 0, 7, 14, 42, and 49 compared with the control. The total eggs for days 0, 7, 14, 42 and 49 differed significantly (P< 0.5) from the control value. Cure as depicted by percentage reduction in mean tissue egg count was up to 97%.

Tissue egg count was considered for different segment of the GIT and associated organs (Liver, spleen and pancreas). It was observed that the rectums and colons had eggs counts which were higher in the rectums than in the colons. The two segments accounted for 55.70% of the tissue eggs, the rest of the GITs accounted for 34.62%, while the livers, spleens and pancreas accounted for 9.68%.

Visual examination of the liver and spleen of mice treated on days 0, 7 and 14 showed healthy organs comparable to those of healthy mice and histological study showed normal liver cell configuration without granuloma or sign of necrosis. On the other hand both control mice and mice treated on days 21, 28, 35, 42 and 49 showed visible indication of gross hepatosplenogenic disease; sandy patches and hepatosplenormegaly.
Discussion

Doenhoff and Bain (1978) and Sabah et al. (1986) independently investigating the chemotherapy of Schistosomiasis, using mouse models had difficulty in determining exactly the stages at which the migratory phases of the diseases were unaffected by treatment. Doenhoff and Bain (1978) observed that worm at 3, 4 and 5 weeks post infection were not susceptible to drug treatment despite the fact that during this period they had reached the hepatic portal system and so were subject to the same titre of the schistosomicide available against them and found to be effective on worms 6 and 7 weeks after infection. Sabah et al. (1986) also performed a similar experiment, and observed that the drug used was effective against development stages at weeks 1, 2, 5 and 6 but not at weeks 3 and 4 post-infection.

The result obtained in this study also revealed that the early migratory stages and adults were susceptible to the plant extracts treatments, mostly on days 0, 7, 14, 24 and 49 and that intermediary stages i.e. those treated on days 21, 28 and 35 were not. This report agrees entirely with those of Doenhoff and Bain (1978) but differ slightly from those of Sabah et al. (1986). The difference exists in the duration after infections when worms are not responsive to extract. Whereas Sabah et al. (1986) found the worms of weeks 3 and 4 not responsive to treatment, in the present research, worms, were also not responsive 5 weeks after infection. This may have been influenced by over-all duration of pre-patency period. With shorter pre-patency period, the period of non-susceptibility may narrow down.

Conclusion

Apart from the side effects, resistance to the drug of choice, praziquantel an inorganic drug whose continued excretion into the environment may be harmful. With the current global concern about the safety of the environment, it would be advantageous to have drugs developed from organic sources, especially those of plant origin. There have been a number of claims regarding the efficacy of some local indigenous plants in the treatment of Schistosomiasis by traditional healers. One of such plants is the *Boswellia dazielii*. As this plant was therefore assayed and its efficacy established, it is hoped that this will provide the basis for development of an effective, safe and environmentally friendly treatment of Schistosomiasis.
References

Christensen, N.O.; Frandsen, and P. Nansen: l’lie Effects of Some Environmental Conditions, Final Host and Parasite Related Factors on the Penetration of *Schistosoma Monsoni* Cercariae into Mice Z. Parasitenkd 1979, 59, 267-275.


Frandsen, F. Host Parasite Relationship of *Bulinus Forskalii* and *Schistosoma Intercalation* Fischer, 1934, from Cameroon./. Helminthol. 1975, 49.


