RESISTANCE TO SCHISTOSOMA BOVIS IN SHEEP INDUCED BY AN EXPERIMENTAL FASCIOLA HEPATICA INFECTION

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ABSTRACT: Sheep infected with Fasciola hepatica for 10 wk acquired a substantial level of resistance to challenge with Schistosoma bovis. The worm burden was reduced by 87.2% (P < 0.01) compared with that of a control group. But when sheep primarily were infected with S. bovis and 6 wk later with F. hepatica, no significant reduction in the S. bovis burden was observed.

Cross-resistance between Schistosoma mansoni and Fasciola hepatica has been demonstrated in mice by Hillyer (1979, 1981) and Christensen et al. (1978, 1980). Calves harboring mature primary infection with Schistosoma bovis also showed significant resistance to challenge with F. hepatica (Sirag et al., 1981). Reciprocal resistance between S. bovis and Fasciola gigantica was detected in Sudanese zebu calves (Yagi et al., 1986). Monrad et al. (1981) found a high level of resistance to F. hepatica in sheep with 2-3 wk-old and 7-8 wk-old S. bovis infections; however, older infections were not protective. Similarly, primary infection of sheep with S. mansoni followed by oral infection with F. hepatica resulted in a reduction of the F. hepatica burden (Haroun and Hillyer, 1988). Immunity to schistosomes using heterologous trematode antigens was reviewed by Hillyer (1984).

This is the first demonstration of cross-resistance induced by a primary infection of F. hepatica in sheep to challenge with S. bovis.

MATERIALS AND METHODS

Fasciola hepatica metacercariae were produced from laboratory-bred Lymnaea truncatula infected with 5-10 miracidia per snail derived from eggs of laboratory-maintained sheep.

The Salamanca strain of S. bovis was maintained in Castellana sheep, and Planorbarius metidiensis was used. Recently-emitted cercariae were used to infect animals.

Three groups of 6 3-mo-old Castellana sheep were used. Figure 1 illustrates the experimental plan. Control animals were infected percutaneously with 400 S. bovis cercariae each for 30 min by submerging the forelimb in a plastic receptacle containing the cercarial suspension of S. bovis. The leg was sheared and washed with water immediately before infection, according to the technique of Van Wyk et al. (1975). Animals in experiment I primarily were infected with 80 F. hepatica metacercariae administered by esophageal probe and challenged after 10 wk with a single exposure to 400 S. bovis cercariae. In experiment II, animals were exposed to 400 S. bovis cercariae and infected 6 wk later with 220 F. hepatica metacercariae.

Necropsies were performed 24 wk after last infection. Schistosoma bovis adults were recovered from the mesenteric and gastric radicles, the infrahepatic branches of the portal vein and the internal iliac veins, using a modification of the perfusion technique developed for use in sheep by McCully and Kruger (1969). The worms remaining in the veins beneath the serosa of the intestine were counted in situ. Fasciola hepatica worms were recovered by dissection of the bile ducts. The degree of resistance was evaluated by comparing the number of worms recovered in infected and control groups.

Statistical analysis of the results was carried out using a 1-way ANOVA.

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FIGURE 1. Experimental plan: fill patterns indicate infection with 400 Schistosoma bovis cercariae (□), infection with 80 Fasciola hepatica metacercariae (■), challenge infection with 400 S. bovis cercariae (□) or challenge infection with 220 F. hepatica metacercariae (■).
Table I. Recovery of Fasciola hepatica and Schistosoma bovis adult worms in experiments I and II and control groups.*

<table>
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<tr>
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<th>Mean worm recovery ± SD</th>
<th>R1 (%)</th>
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<tr>
<td></td>
<td>Animals</td>
<td>F. hepatica</td>
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<tr>
<td>Control</td>
<td>6</td>
<td>—</td>
</tr>
<tr>
<td>Experiment I</td>
<td>6</td>
<td>55.5 ± 23.6</td>
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<tr>
<td>Experiment II</td>
<td>6</td>
<td>192.3 ± 14.9</td>
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* Experiment I, sheep infected with 80 F. hepatica metacercariae and challenged with 400 S. bovis cercariae; experiment II, sheep infected with 400 S. bovis cercariae and challenged with 220 F. hepatica metacercariae; control, sheep infected with 400 S. bovis cercariae.
† R, percentage reduction of S. bovis burden. R = ([number of worms in control − number of worms in experiment] / number of worms in control) × 100.
‡ One-way analysis of variance.

RESULTS

Worm recoveries are indicated in Table I. A substantial level of protection to S. bovis, in terms of worm reduction, developed in sheep infected with a single dose of 80 metacercariae. The mean burden of S. bovis in the group previously infected with F. hepatica was much lower than that in the control group, corresponding to an 87.2% reduction. ANOVA showed that the difference between the means was significant (P < 0.01). The results of S. bovis recovery in experiment II showed that a F. hepatica infection did not have influence on a previously established S. bovis infection, as the difference between the mean burdens of S. bovis in experiment II and the control group was not significant (P > 0.7).

DISCUSSION

There is no evidence that primary infection of sheep with F. hepatica induces resistance to homologous challenge in terms of reduction in the number of worms recovered from this latter infection. This fact has been reported by numerous authors (Boray, 1967; Sinclair, 1971a, 1973; Knight, 1980; Sandeman and Howell, 1981). Attempts to stimulate resistance to F. hepatica by immunization with somatic or metabolic products (Ross, 1967; Sandeman et al., 1980) and homogenates of lymph nodes or spleen (Sinclair, 1971b) consistently were unsuccessful in inducing protection in sheep. However, the findings from experiment I presented here provide evidence of heterologous resistance. These facts suggest that F. hepatica might evade the host’s immune system by eliciting humoral and cellular responses to nonessential epitopes that may prove vital for S. bovis.

Data from experiment II seem to indicate that the protective mechanisms triggered by F. hepatica act on the primary stages of S. bovis but not on established adult worms. We have found no reference to the resistance to S. bovis in sheep induced by F. hepatica infections. Yet, Yagi et al. (1986) found that F. gigantica experimental infections protected cattle against S. bovis challenge. Studies (Monrad et al., 1981; Sirag et al., 1981) in line with the present one showed that a primary infection with S. bovis protected sheep and calves to a challenge with F. hepatica. Sheep infected with S. mansoni and challenged with F. hepatica also presented (Haroun and Hillyer, 1988) a reduction in the F. hepatica burden. Similarly, infection of mice with F. hepatica conferred significant resistance to a heterologous S. mansoni challenge (Christensen et al., 1978). It is not known whether the observed heterologous resistance involves immunological factors, mechanical barriers, or both.

Heterologous trematode antigens have been used (Hillyer, 1984) to induce immunity against S. mansoni. It has been established (Hillyer, 1979) that cross-reactive antigens isolated from F. hepatica protect against S. mansoni and that some of these antigens share common epitopes with other trematodes such as S. bovis.

It has been found (J. Rojas, 1991, pers. comm.) that 125-, 69-, 36-, and 17-kDa F. hepatica antigens were shared by S. bovis. These common antigens may well be responsible for the heterologous resistance reported here. Further studies and the isolation of these antigens will be necessary to elucidate their possible protective role.

ACKNOWLEDGMENTS

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