MISCELLANEOUS MAMMALS (*DASYPUS NOVEMCINCTUS, MYOCASTOR COYPUS, PROCYON LOTOR,* AND *BASSARICUS ASTUTUS*) AS HOSTS FOR *SCHISTOSOMA HAEMATOBIUM* (IRAN)

Robert E. Kuntz, Jerry A. Moore, and Tao-cheng Huang

Department of Parasitology, Southwest Foundation for Research and Education, San Antonio, Texas

In a continuing search for animals which may be employed for parasite carcinogenesis and other aspects of schistosomiasis haematobia, four mammals (armadillo, *Dasypus novemcinctus;* nutria, *Myocastor coypus;* raccoon, *Procyon lotor;* and ringtail (*Bassaricus astutus*) were exposed to infection by the Iran strain of *Schistosoma haematobium.* Based upon overall host-parasite relationships, none of these may be recommended as models for *S. haematobium.* Moderate numbers of schistosomes were recovered from the armadillos, nutrias, and ringtails. The raccoon was essentially resistant to infection. None of the animals under observation developed apparent pathology or sustained parasite residence in the urogenital system.

Various parameters have been employed to characterize the mammalian schistosomes biologically as well as morphologically. A significant consideration has been information on definitive host-parasite relationships which elucidate susceptibility patterns, degrees of immunity, and resistance to infection and allied aspects of biology which have contributed substantially to a better understanding of the adult part of the parasite cycle. Furthermore, an evaluation of different definitive host-parasite systems, especially those including representatives from different major taxonomic categories of mammals, have provided indications suggesting which species could be used for experimental schistosomiasis. There is an accumulation of data from numerous laboratories which have used small numbers of different species of animals for different schistosomes. However, the work of Stirewalt et al. (1) was among the first in which attempts were made to establish a comparative evaluation of host susceptibilities. This investigation initiated subsequent researches concerned with a broad spectrum of mammalian hosts, domestic as well as wild, exposed to schistosomes from different geographic areas. It also provided the impetus for studies which would suggest mammals that might be considered as natural reservoirs of infection.

The present paper is one in a series in which many species of mammals have been exposed to *S. haematobium* (2-7) in an attempt to collect much needed information on the least understood of the schistosomes of man and, above all, search for definitive host systems that might allow for continuing research on the basic biology of this schistosome and possibly parasite carcinogenesis (8).

MATERIALS AND METHODS

Armadillos (*Dasypus novemcinctus,* Order Xenthara), raccoons (*Procyon lotor,* Order Carnivora), and ringtails (*Bassaricus astutus,* Order Carnivora) were captured in the vicinity of San Antonio. Nutria (*Myocaster coypus,* Order Rodentia) were provided through the courtesy of Dr. Franklin Sogandares-Bernal, Southern Methodist University, Dallas. Armadillos were initially fed on a mixture of whole eggs blended with canned dog food, then on a mixture of water soaked dry dog food (Purina) and canned dog food. Nutrias were fed dried rodent and dried dog food as well as supplements of lettuce and other green vegetables. Raccoons and ringtails subsisted on canned dog food, water soaked dog food, and occasionally discarded mice. After stabilization to laboratory conditions, hosts were handled and infected by customary procedures practiced in this and other laboratories. Cercariae shed by a number of snails (*Bulinus truncatus*) were pooled to enhance chances for a balanced sex ratio of schistosomes. Mammals were anesthetized (Sernylan) then exposed to infection as counted

numbers of cercariae in drops of water on coverslips were placed on the abdomen which was clipped and cleansed with water prior to exposure.

Hosts were maintained for 3 to 25 weeks post exposure. At time of necropsy hosts were given a lethal dose of Sernylan. Visceral organs were removed and examined individually. Schistosomes were recovered manually as well as by saline perfusion of hepatic portal system and parts of the mesenteric venous circulation. Small samples or crushes of fresh tissue were taken at random to determine the presence of schistosome eggs and to judge the percentage of eggs viable. Eggs per gram (EPG) of tissue were determined by aliquot sampling of organs digested by the KOH technic (2.5% potassium hydroxide 12-24 hrs at 40 C) (9).

RESULTS

Three of 6 armadillos died. Death, however, was not necessarily attributed to schistosome infections, even though moderate numbers of parasites were present at post mortem examination (Table I). Sexual differentiation was not feasible for worms recovered from a host (A-9) examined 3 weeks post infection. Sexes were recognizable, but there was no mating of schistosomes in a host which died at the end of 4 weeks infection. Schistosome returns ranged from 17.8 to 43.4% of the cercariae applied, the former recorded for an armadillo examined 28 weeks post exposure to 500 cercariae. All hosts produced a preponderance of male *S. haematobium*, and all possessed a large proportion of the parasite population in the liver and associated hepatic-portal veins. Moderate numbers of adult schistosomes were found in branches of the mesenteric veins associated with the small and large intestine.

No eggs were detected in random fresh tissue crushes or in the potassium hydroxide (KOH) organ digests of host A-11 eight weeks post exposure. Eggs were present, however, in the liver, pancreas, and all levels of the small and large intestine of a host (A-6) necropsied at 28 weeks. There was approximately 450 and 300 eggs per gram (EPG) for the liver and pancreas respectively with a maximum of 975 EPG for the posterior part of the large intestine. There was no apparent pathology.

TABLE I. *Worm recoveries from hosts exposed to cercariae of Schistosoma haematobium (Iran)*

Host No./Sex	Cercariae/Snails	Duration of Infection (Weeks)	Worm Recoveries: Principal Organs						Worm Recoveries				Percentage of Recovery
			Lungs	Liver + Hepatic Portal Veins	S. Int.	Pancreas	Spleen	L. Int.	Prs	♂♂	♀♀	Total	
a. Armadillo: *Dasypus novemcinctus* (Linnaeus)													
A 9♀	500/28[a]	3	0	219	0	0	0	0	U	U[b]	U[b]	219	43.8
A 10♀[c]	500/28	4	0	157	0	0	0	0	0	110	47	157	31.4
A 11♀[c]	500/28	8	0	130	4	0	0	14	14	126	0	154	30.8
A 3♂	500/50	12	0	69	48	0	0	92	76	57	0	209	41.8
A 6♀[c]	500/45	28	0	67	10	6	0	6	18	53	0	89	17.8
A 4♂	1000/50	19	0	156	25	0	0	253	200	34	0	434	43.4
b. Nutria: *Myocastor coypus* (Molina)													
N 2♂	1000/77	19	0	155	16	10	2	31	60	94	0	214	21.4
N 1♂	1000/77	20	0	138	20	4	0	12	38	98	0	174	17.4
N 3♀	1000/77	26	0	95	20	2	0	22	34	71	0	139	13.9
c. Raccoon: *Procyon lotor* (Linnaeus)													
R 3♀	2000/140	8	0	18	0	2	0	0	3	14	0	20	1.4
R 1♂	2000/71	10	0	5	0	1	0	1	0	6	0	6	0.3
d. Ringtail: *Bassariscus sastutus* (Lichtenstein)													
Rt 1♂	1000/77	25	0	81	14	2	0	22	12	94	0	118	11.8
Rt 2♂	1000/77	25	0	67	18	0	0	2	56	77	0	189	18.9

[a] 500/28 = 500 cercariae obtained from 28 *Bulinus truncatus*
[b] U = immature worms of undetermined sex
[c] Died

The 3 nutrias exposed to 1000 cercariae each were sacrificed at 19 to 26 weeks. Worm returns ranged from 13.9 to 21.4%. The highest percentage was recorded in the oldest infection. There was a greater prevalence of male schistosomes and the majority of the worms resided in the liver and hepatic-portal vessels. Parasites occurred, however, in both the small and large intestine. All hosts had parasites in the pancreas. No eggs were recorded for fresh tissue crushes of KOH tissue digests, and there was no macroscopic evidence of pathology.

Two young adult *Procyon lotor* exposed to 2000 cercariae each produced a minimum number of schistosomes when examined 8 to 10 weeks later. The majority of *S. haematobium*, some of which were poorly developed, occurred in the liver and hepatic-portal vessels, 3 resided in the pancreas, but none were found in the small intestine. A single small worm resided in veins on the large intestine of 1 raccoon. No eggs or egg remnants were present in fresh tissue crushes taken at random from the principal viscera.

Two adults of another carnivore, *Bassaricus astutus*, were necropsied 25 weeks post exposure to 1000 cercariae each. There was a pronounced dominance of males and the greater proportion of parasite population was perfused from the liver and hepatic-portal veins. Moderate numbers of worms occurred along the intestinal tract. The total return of *S. haematobium* ranged from 11.8 to 18.9%. Tissue crushes were prepared from the viscera of both hosts. Only a few viable eggs were found in the liver. Larger numbers of eggs with a greater proportion of viables were noted in the walls of the small and large intestine.

DISCUSSION

The ability of a broad spectrum of domestic as well as wild animals to accommodate *S. mansoni* in varying degrees is fairly well documented (10-12), and the infection of even wider range of mammals by *S. japonicum* is an accepted fact. The biology of *S. haematobium* in its definitive hosts, however, until recent years has been only poorly understood, and the assumed inability of this species as well as other members of the terminal spine egg complex to parasitize many mammals has been considered as a characteristic of these parasites. A sparsity of definitive host-parasite information of the latter group of mammalian schistosomes has resulted from a lack of exploratory researches at the laboratory level and to the unavailability of these schistosomes to more than a few institutions.

Mammals employed in the present study are somewhat limited in number but represent the first evaluation of 4 different genera as hosts for *S. haematobium* (Iran). The armadillo has had limited use as an experimental host for helminth parasites, but the South American armadillo (*Euphractus sexcinctus*) has been infected with *S. mansoni* (11). As indicated in the present investigation, *Dasypus novemcinctus* is moderately susceptible to infection by *S. haematobium*, but host-parasite relationships were unusual in that no significant pathology developed even after 19 to 28 weeks and no parasites or eggs were associated with the urogenital system. The armadillo apparently can support a comparatively large population of *S. haematobium*, but host-parasite relationships are not conducive to egg production by mature parasites and to the usual pathology resulting from egg deposition in tissues. This host, therefore, would not be a practical candidate for continuing studies in schistosomiasis haematobia.

Rodents, on the other hand, have been examined extensively as hosts for *S. mansoni* and other schistosomes in Africa (12), but only limited information has been forthcoming for *S. haematobium*. Gear *et al.* (13), however, have made comparative evaluations of *S. haematobium* infection in wild rodents adaptable to the laboratory in South Africa. In a search for useable experimental hosts, Kuntz and Malakatis (2, 3) exposed a number of species of rodents to *S. haematobium* in Egypt. A broad range of susceptibilities was revealed. It was concluded that albino mice (*Mus musculus*), hamsters (*Mesocricetus auratus*), spiny back mice (*Acomys cahirinus*), and gerbils (*Gerbillus pyramidum*), were usable hosts, but albino rats (*Rattus norvegicus*), cotton rats (*Sigmodon hispidus*), guinea pigs (*Cavia copaya*), and others were poor hosts. The Nile rat (*Arvicanthis niloticus*) was judged a good host based on return of worms and production of numerous eggs in tissues and passage with excreta. With

this known range of host-parasite relationships, it is no surprise that the rodent *Myocastor coypus*, although a natural host for *Heterobilharzia americana*, is not an acceptable host for *S. haematobium*, especially if there is a concern for pathology and involvement of the urogenital system.

Carnivores are recognized as poor hosts for *S. mansoni* as well as for *S. haematobium*. Cats and dogs were virtually resistant to infection by the latter species (2). It also was demonstrated (3) that 3 wild carnivores (Egyptian weasel, *Mustela nivalis subpalmata*; mongoose, *Herpestes i. ichneumon*, and Egyptian fox, *Vulpes v. aegyptiaca*) would not accept the infection with the Egyptian strain of *S. haematobium*. The raccoon has followed the typical pattern for carnivores exposed to the schistosomes of man, even though it is a common, naturally infected host for *H. americana* in Louisiana (14). The ringtail, on the other hand, has demonstrated a moderate accommodation to *S. haematobium* with deposit of a few eggs in different levels of the intestinal tract at 25 weeks post infection.

In the present investigation, 4 species of animals representing 3 orders of mammals have been subjected to experimental infection by *S. haematobium* (Iran). None can be recommended as hosts for continuing studies in experimental schistosomiasis haematobia since parasite returns, except for the armadillos, were low, and in none of the hosts was the parasite able to sustain residence in the urogenital system. In the absence of appreciable egg deposits, and thus no apparent pathology in any of the organ systems, it must be assumed that these hosts do not provide an acceptable host-parasite situation. These hosts, however, could possibly be considered for investigations directed toward host resistance to *S. haematobium* infection.

REFERENCES

1. M. E. STIREWALT, R. E. KUNTZ and A. S. EVANS, Am. J. Trop. Med. 31: 57-82 (1951).
2. R. E. KUNTZ and G. M. MALAKATIS, Exptl. Parasitol. 4: 1-20 (1955).
3. R. E. KUNTZ and G. M. MALAKATIS, J. Parasitol. 41: 467-475 (1955).
4. R. E. KUNTZ, B. J. MYERS and A. W. CHEEVER, Bull. World Health Organ. 45: 21-25 (1971).
5. R. E. KUNTZ, B. J. MYERS, J. A. MOORE and T. C. HUANG, Exptl. Parasitol. 29: 33-41 (1971).
6. B. J. MYERS, R. E. KUNTZ, T. C. HUANG and J. A. MOORE, Lab. Animal Care 20: 1004-1006 (1970).
7. B. J. MYERS, R. E. KUNTZ, T. C. HUANG and J. A. MOORE, Proc. Helm. Soc. Wash. 37: 189-192 (1970).
8. A. W. CHEEVER, R. E. KUNTZ, B. J. MYERS, J. A. MOORE and T. C. HUANG, Am. J. Trop. Med. and Hyg. 23: 429-448 (1974).
9. A. W. CHEEVER, Bull. World Health Organ. 39: 328-331 (1968).
10. R. E. KUNTZ, Am. J. Trop. Med. and Hyg. 4: 383-413 (1955).
11. A. V. MARTINS, Bull. World Health Organ. 18: 931-944 (1958).
12. G. S. NELSON, C. TEESDALE and R. B. HIGHTON, Ciba Foundation Symposium on Bilharziasis, (Eds. G. E. W. Wolstenholme and M. O'Connor) J. and A. Churchill Ltd., London 1962, pp. 127-149.
13. J. H. S. GEAR, D. H. S. DAVIS and R. J. PITCHFORD, Bull. World Health Organ. 35: 213-221 (1966).
14. E. A. MALEK, L. R. ASH, H. F. LEE and M. D. LITTLE, J. Parasitol. 47: 619-623 (1961).