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FURTHER OBSERVATIONS ON THE RELATIONSHIP BETWEEN SERUM MITOCHONDRIAL ASPARTATE TRANSAMINASE AND PARASITIC INFECTION. UNEXPECTED ABSENCE OF HUMAN SCHISTOSOMIASIS AROUND LAKES GEORGE AND EDWARD (UGANDA)

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ABSTRACT

1) Contrary to prediction, no cases of schistosomiasis could be detected in two lakeside communities.
2) Serum levels of mitochondrial aspartate transaminase (mAAT) in these communities were apparently unrelated to the presence or absence of detectable malaria parasites.
3) By contrast, serum mAAT was significantly raised in 3 'healthy' cases of schistosomiasis who were otherwise comparable with the lakeside dwellers. (Significance was reached notwithstanding the paucity of cases).
4) These observations should be extended especially in respect of the Asian region where different species of Schistosoma are involved in parasitisation, but progress has been blocked by lack of finance.

INTRODUCTION

Ongom and Bradley (1) studied a community living beside the Nile at Panyagoro, West Nile (Uganda) and found an extremely high level of infection with Schistosoma mansoni. They suggested that communities living in similar conditions elsewhere, including specifically the area of the study reported here, would be found to have similar parasite loads. The Hippopotamus population of Lakes George and Edward is infected with parasites of the same genus (2,3) and carrier snails of an appropriate species are present (4). Nevertheless, many Uganda residents believed that these lakes were safe and it therefore seemed worth determining what the prevalence really was, for reasons both of scientific and local public health interest.

The matter was of additional interest because a preliminary study (5) had shown high serum levels of the mitochondrial isoenzyme (mAAT) of aspartate transaminase (EC 2.6.1.1) in hospitalised cases of schistosomiasis, with little or no change in the level of cytosolic isoenzyme. These patients, however, probably had multiple parasitic

infections, including malaria, so that a causal relationship could not be established with confidence. (The enzyme concerned is still commonly referred to as glutamic-oxalacetic transaminase, GOT. Serum isoenzyme studies to date are rather few and most are unreliable because of technical difficulties).

We thus had an opportunity to learn from an experiment of Nature. Beginning with the presumption that the area would be found to be highly malarious, three possible patterns suggested themselves: a) the area would prove to be free of schistosomiasis, in which case any elevated levels observed of serum mAAT must be due to other causes, possibly malaria, or b) the area would prove to be heavily infected, or c) a few isolated cases would be found among a population generally free of the disease, in which case any association of elevated mAAT levels with infection would gain in significance.

GEOGRAPHY, SUBJECTS AND PROCEDURES

The twin lakes, George and Edward, lie at an altitude of about 914 m and are linked by the broad Kazinga channel, in which there is negligible current. The outflow from Lake Edward flows by the Semliki river (over a series of rapids) to Lake Albert and thus connects with the Nile - near Panyagoro.

On the shores of Lake George there is, or was, a fish processing factory and an adjacent village, Kasenyi, whose inhabitants made their living by selling fish to the factory. Village children spend much of their day in the water and the men spend 6 or more hours per day fishing with nets from small, crude dug-out canoes - almost continually in contact with the water. Children whose parents are employed directly by the factory live in a camp provided for employees. Most of these go to school, but nevertheless spend several hours each day in contact with lake water, since they go down to the lake after school is over. At Mweya Lodge, on Lake Edward, life is rather similar, but the people there do not engage in commercial fishing and contact with water is less intensive. Most samples were from Kasenyi and to these were added a few from National Parks rangers stationed at Mweya, and their children, and a few from patients of the hospital at Kilembe Mines.

The area is highly malarious, as our results confirm. It was also one of the areas severely affected by a sleeping sickness epidemic in the early years of the century. The human population was evacuated at that time as part of the control measures and is still restricted to a small number, or varied tribal origins, all recent immigrants.

The survey occupied 3 weeks in mid-1970 and made use of facilities at a temporary biological research station at Kasenyi. Assay of mAAT was carried out there to avoid the consequences of any changes during storage, but otherwise work in the field was confined to collection of faecal samples, blood smears and serum, the latter being stored at -20°C. Enzyme assays were conducted according to Varley (6) or Boyde (7): results are quoted as IU/l at 25°C. Serum total protein was measured by means of a refractometer; blood smears were dried, fixed in ethanol and stained with Leishman's stain; faecal samples were examined for eggs of S. mansoni after concentration using the acid-ether sedimentation technique.

RESULTS

No schistosome ova were found in stool samples from any of the 60 persons studied at Kasenyi and Mweya (Group 2, adults; Group 3, children). Three cases of active schistosomiasis (Group 1) were discovered at Kilembe Mines Hospital, 30 km away, all at routine pre-employment medical examination of men who had come from other parts of the country to find work, and thus presumably felt themselves to be in a satisfactory state of health. All three showed marked elevation of serum mAAT.

It will be seen from the Table that the mean level of serum mAAT (and Total AAT) of group 1 is considerably higher than in other groups. If 'Student's' t test is applied, the differences are found to be formally significant ($p < 0.001$).

TABLE 1. Summary of Results. Mean ± standard deviation (with Bessel's correction) or number of cases

Group No.	Description	No. of cases	Age (years)	mAAT (grade)[a]	Total AAT (IU/l)	Alkaline phosphatase[b] (IU/l)	Protein (g/l)	Malaria parasites found (number)
1	Schistosomiasis	3	27 ± 13.0	2.67 ± 0.58	23.8 ± 13.7	97.7 ± 8.9	68.3 ± 3.2	1
2	Adults from Kasenyi and Mweya	48	30.7 ± 7.3	1.29 ± 0.62	7.0 ± 3.2	95.5 ± 17.8	68.6 ± 6.3	11
3	Children from Kasenyi and Mweya	12	9.8 ± 2.6	0.92 ± 0.26	6.35 ± 1.3	101 ± 19.8	57.2 ± 9.0	2
4	Malaria-positive Kasenyi and Mweya (Includes all such cases from Groups 2 and 3 but not from group 1)	13	27.0 ± 11.4	1.31 ± 0.63	5.7 ± 1.6	91.8 ± 17.3	65.6 ± 8.6	all (i.e. 13)

[a] For the purposes of this paper, the original literal grading system for mAAT results (ref. 7) is replaced by numerical grades, to allow statistical calculations, as follows: grade 0 (= grade A), 2-4 IU/l; grade 1 (= grade B), 4-6 IU/l; grade 2 (= grade C), 6-9 IU/l; grade 3 (= grade D), 9-14 IU/l.

[b] Alkaline phosphatase assay was omitted, for lack of material, in 3 cases from group 2.

The Total AAT results in groups 2-4 are a little low, probably reflecting loss during storage before assay: this would presumably affect group 1 also. The high Total AAT level of group 1 can be largely accounted for by mAAT.

Of 60 persons studied in the main survey area (Kasenyi and Mweya) 2 showed mAAT of grade 3. One was a woman of 25 years with serum Total AAT of 23.5 IU/l, alkaline phosphatase 150 IU/l and serum protein 60 g/l. Very probably she had active liver disease caused by some agent other than S. mansoni. The mAAT result in the other case is suspect, since the serum Total AAT was only 5.4 IU/l. This was a man of 25, malaria-positive, with serum protein of 68 g/l; alkaline phosphatase was not done.

DISCUSSION

The above results show that schistosomiasis is either absent from the population studied, or at least that the level of infection was too low for us to detect. (Serological diagnostic methods were not available to us at the time). We could conceivably have missed light or scattered infections, but not the level of parasite load seen at Panyagoro. This is perhaps surprising but may be explicable on the basis that there was for many years almost no human population in the area, so that the parasite died out at that time and has not been re-introduced since, or not in sufficient numbers for us to detect. This would imply also that there is no fully effective local non-human host/reservoir for the parasite.

Notwithstanding the small number of cases of schistosomiasis studied, a comparison of serum mAAT levels between this group and the Kasenyi-Mweya people reaches statistical significance, reinforcing the earlier suggestion that there may be a causal relationship.

ACKNOWLEDGMENTS AND NOTES

We thank the Medical Research Council, London, for financial assistance, Mr. A.M. Nanji for technical assistance and the staff of the International Biological Programme Research Station at Kasenyi for hospitality and facilities. At the time of this work JDGD was Senior Medical Officer at Kilembe Mines Ltd. and T.R.C.B. was Professor of Biochemistry at Makerere University, Kampala, Uganda.

The very-greatly-desired extension to larger numbers of cases was prevented first by political-social difficulties and latterly by lack of finance, despite applications to the World Health Foundation (HK), World Health Organisation, and various Research Funds of the University of Hong Kong.

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