

Hydatid disease in the Western Isles

By ISOBEL L. CHISHOLM, M. J. MACVICAR* AND H. WILLIAMS

Department of Microbiology, Raigmore Hospital, Inverness
and * *Department of Agriculture and Fisheries for Scotland,*
Castle Street, Dingwall

(Received 28 May 1982; accepted 5 July 1982)

SUMMARY

An investigation into the incidence of hydatid disease was carried out in the Hebridean islands of Lewis and Skye. The results showed that 20% of the sheep were infected and 10% of the dogs harboured *Echinococcus granulosus*. Sporadic human cases continue to occur but using serological tests we were unable to show evidence of subclinical infection in the population sampled.

INTRODUCTION

Hydatid disease acquired in the United Kingdom probably occurs now only sporadically and in only two areas – the central counties of Wales and the islands of the north and west of Scotland. The disease, caused by the larval form of *Echinococcus granulosus* is a world-wide zoonosis which occurs in communities where man, grazing animals and carnivores – typically sheep and dogs – live in close association. Sporadic cases of hydatid disease have occurred regularly in the Hebridean islands; an analysis of 17 cases treated at Raigmore Hospital, Inverness over the last 16 years shows that all patients were from the islands of Lewis or Skye, or from the immediately contiguous mainland.

In 1974 two young boys from Staffin, Skye were found at routine school medical examination to have enlarged livers. Both were admitted to hospital and hydatid cysts of the liver were successfully removed. One year previously a neighbour of one of the boys had been admitted to a hospital outside this region and was found to be suffering from hydatid disease.

In some parts of the world schemes for the eradication of hydatid disease have been successfully implemented. If such attempts were to be made in Scotland, the first stage would be an adequate local assessment of the prevalence of the condition and of the methods of surveillance. In an attempt to assess the human hazards, and to get more information on the incidence of the disease, we carried out an investigation in Lewis and Skye during the period 1978–1981.

Lewis is the largest of the Hebridean islands with a population of approximately 21000. The population is grouped in small communities or townships with the exception of Stornoway which has a population of 6000. The annual average rainfall is approximately 43 in and the mean summer temperature 54 °F. Much of the land is moorland and suitable only for grazing sheep. The sheep population

numbers about 150 000 and there are approximately 2500 registered owners. The sheep are mainly on the hills in summer, on the common grazings, and are brought back to the crofts in November where they are kept until returned to the hill grazings following lambing in April.

Home slaughtering of sheep for private consumption is believed to be widespread. Killing of homebred sheep at the Stornoway slaughterhouse takes place each year from July to December.

There is an estimated dog population of about 4000 on the island, but no foxes.

Stornoway has a modern hospital of 83 beds which is served by a small, busy laboratory. There is an extensive family practitioner service on the island.

The island of Skye in the Inner Hebrides is also basically a crofting community with a resident population of about 7000 which is greatly augmented by visitors during the summer months. The sheep carried number approximately 150 000, managed under a system similar to that described for Lewis, but the holdings tend to be larger. The only slaughterhouse is open from September to December for the killing of homebred sheep. Home slaughtering for private consumption is believed to be common practice. There are approximately 2000 dogs on the island and foxes are well-established.

Small hospitals are established at Broadford and Portree, and an extensive family practitioner service has been developed. Laboratory services for the island are carried out at Raigmore Hospital, Inverness.

MATERIALS AND METHODS

The incidence of the disease in sheep was established from counts of infected livers and lungs of all sheep slaughtered at Stornoway abattoir during a set time each year – the third week of November. Similar counts had been made by one of us (M.J.M.) for a number of years prior to this survey. Infection in the dog population in Lewis was estimated by post-mortem examination of animals which had been presented to the veterinary practitioners for euthanasia. The whole of the small intestine was removed and sent to the laboratory at Raigmore where it was opened longitudinally and the inner lining scraped and washed through sieves of 2 mm, 425 μ m and 250 μ m mesh. The residue of each sieve was separately washed into a tray with a black base and examined using an illuminated magnifier ($\times 5$). The majority of *E. granulosus* was retained by the 425 μ m mesh and they were identified and counted. The method was that described for the examination of dog faeces (Walters & Clarkson, 1980).

Evidence of human infection was sought for solely by serological methods. Two thousand eight hundred and sixty serum samples from outpatients and inpatients at the Stornoway hospital and 200 serum samples from Skye were available for examination. All sera were stored at -20°C prior to testing.

Antigen. Sheep cysts were obtained from the Stornoway abattoir. Collection, concentration and clearing of sheep hydatid fluid were performed by standard methods (Pozzuoli *et al.* 1972). Scolices were also collected aseptically and cultured by the method described by Smyth (1967). The culture fluid was removed between 14 and 21 days later and processed in the same way as hydatid fluid. Both antigens, once standardized, were found equally effective for all the tests performed. Storage of the antigen was in liquid nitrogen.

Table 1. *Hydatid infection of sheep carcasses, Lewis 1976-1980*

Year	No. of carcasses examined	No. (%) carcasses infected
1976	226	26 (11.5)
1977	216	36 (16)
1978	367	87 (22)
1979	420	87 (21)
1980	493	92 (19)

Table 2. *Echinococcus granulosus infection in dogs*

No. of dog intestines examined	Number and percentage with	
	<i>E. granulosus</i> present	Other worms/cestodes/nematodes present
50	5 (10%)	22 (44%)

Serological Tests. Five tests were employed for the examination of human sera. These were the indirect haemagglutination test (IHAT), latex agglutination test (LAT), countercurrent electrophoresis (CCEP), complement fixation test (CFT) and immunoelectrophoresis (IEOP). Screening was carried out by IHAT, LAT and CCEP and any serum considered positive in any one of these tests was further tested by CFT and IEOP.

The IHAT was based on the method described by Thorburn & Williams (1972) in which sheep red blood cells tanned with pyruvic aldehyde were sensitized with either hydatid cyst fluid or hydatid scolices culture fluid. Sera yielding titres of 64 or greater were tested further. The LAT was a simple slide agglutination test using an antigen commercially available (Immuno Ltd, Sevenoaks, Kent). The CCEP test was carried out on cellulose acetate membrane (Gentilini, Pinon & Niel, 1972; Mansueto *et al.*, 1980) and all sera giving rise to one or more precipitin lines were tested further. The CFT was the method used by Bradstreet (1969) and titres of 4 or greater were considered significant. The IEOP was performed as described by Guisantes *et al.* (1975) and the presence of Capron's arc 5 was considered diagnostic of hydatid disease (Capron, Vernes & Biguet, 1967).

Several serum samples were separated by sucrose gradient centrifugation. The separation was carried out according to the method of Vesikari & Vaehri (1968). Preformed gradients of sucrose (12-38.5%) were made in cellulose nitrate tubes with a gradient former. 0.5 ml serum was layered onto the gradient and centrifugation at 40000 rpm (99000 g) was carried out at 4 °C for approximately 16 hours.

IgE levels were measured by radioimmunoassay (RIA) using reagents commercially available (Pharmacia (GB) Ltd, Hounslow, Middlesex). Known positive human sera from cases of hydatid disease were available as controls.

RESULTS

Table 1 shows results of examination of sheep carcasses at Stornoway abattoir. The figures for 1976 and 1977 were also available and are included. The results indicate that about 20% of sheep in Lewis are infected with the larval form of the parasite.

Table 3. Results of the indirect haemagglutination test (IHAT) in 3060 samples of sera from patients resident in Lewis and Skye

Titre	Lewis, 2860 sera		Skye, 200 sera	
	No.	%	No.	%
1024	1	—	—	—
512	1	—	3	1.5
256	2	—	2	1.0
128	15	0.5	—	—
64	43	1.5	10	5
32	130	4.5	23	11.5
16	505	18.0	49	24.5
8	1205	43.0	74	37.0
< 8	858	30.0	39	19.5

Table 4. Numbers of sera (with titres of ≥ 64 in the IHAT) which react in LAT, CCEP and CFT

	IHAT ≥ 64	LAT	CCEP	CFT	LAT/CCEP	LAT/CFT	CCEP/CFT
Lewis	62	9	28	8	2	3	1
Skye	15	0	2	4	0	0	2

Table 5. Serological results of four cases of hydatid disease

Site of cyst	IHAT*	LAT	CFT*	CCEP	IEOP †	IgM* antibody	IgG* antibody	IgE† antibody
Liver	1024	+	32	ND	ND	8	32	501
Liver	256	+	16	+	+	4	16	32
Lung	1024	+	64	+	+	8	32	ND
Liver	128000	+	128	+	+	32	> 256	212

* Results expressed as a titre.

† Results expressed in units/ml.

Dogs. The intestines of 50 dogs were examined by the methods described. The results are shown in Table 2. The five positive dogs comprise four working collies from widely separated areas of Lewis and one pet spaniel from Stornoway. Their ages ranged from 2 to 10 years.

Human sera. Table 3 shows the results of screening 3060 human sera from patients resident in Lewis and in Skye using the IHAT. The 77 serum specimens which gave titres of 64 or more were further tested by the LAT, CCEP and the CFT and the results are shown in Table 4.

Sera positive to a titre of 64 in the IHAT test, and that reacted in any of the other tests, were fractionated on sucrose gradients and the fraction tested by haemagglutination. The results showed that all the reactant sera were positive in the IgM fractions only. No antibody was detected in the IgG fraction in contrast to the panel of positive sera used as controls (Table 5) where antibody was always

Table 6. Serological results in two cases of hydatid disease discovered during the survey

Patient	Site of cyst	IHAT*	LAT	CFT*	CCEP	IEOP	IgM*	IgG*	IgE†
1	Liver	1024	+	32	-	+	8	32	> 500
2	Liver	256	+	16	+	+	4	16	100

* Results expressed as a titre.

† Results expressed in units/ml.

detected in the IgG fractions. The radioimmunoassay test for IgE antibody carried out on the sera under investigation showed levels consistently below those from patients with proven hydatid disease (Table 5).

During the course of the survey, two clinical cases of hydatid disease occurred. Both were suspected on clinical grounds and the diagnosis confirmed at operation. The serological results of these two cases are shown in Table 6.

DISCUSSION

With approximately 20% of the sheep in Lewis infected with the larval form and approximately 10% of dogs harbouring the adult worm of *Echinococcus granulosus* it was clear that the parasite is well-established on the island and the potential for human infection must remain high. Over the last decade considerable attempts have been made to inform the local island communities of the importance of making offal unavailable to dogs by burying dead sheep and regular deworming of dogs. However, the fact that nearly 50% of dogs harbour helminths suggests that present methods are not having much success.

Using sera that were available through the hospitals we were unable, in a considerable sample of the population, to show evidence of subclinical infection. We must assume that either the tests were not sufficiently sensitive, that the wrong population group was screened, or that despite the zoonotic potential the risk to the human population is small. There has been some discussion on the most suitable test for epidemiological purposes in hydatid disease. The Casoni skin test has been considered inadequate because of non-specificity (Varela-Diaz *et al.* 1976) and the emphasis has shifted to serological techniques. The LAT, CCEP and IHAT have been used and have been favoured for their good correlation with IEOP and the demonstration of Capron's arc 5 which is the definitive test for the presence of parasite antibody in human sera. Picardo & Guisantes (1981) favoured the IHAT based on a study of 123 selected sera, provided that a minimum titre of 128 was accepted. Under these conditions the results were similar in sensitivity and specificity to the LAT and CCEP tests. We used the IHAT as a screening test, and even when the critical level was dropped to 64, and the test system supplemented by the LAT and IEOP tests we were not able to confirm the specificity in any of the LAT positive sera, and none showed specific antibody in the IgG fraction of gradient separated globulins. During the course of the survey the two cases which occurred were readily detected by the tests that we employed as were a number of other cases from outside the United Kingdom.

An analysis of the age range of cases of hydatid diseases seen at Raigmore Hospital, Inverness over the last 16 years shows that the majority occurred in males over 50 years of age apart from two schoolboys of 6 and 13 years, respectively. It is probable that this group was not well-represented in the group sampled serologically and in these communities this is probably the most difficult group to sample. The two cases that occurred during the survey were males of 50 and 67 years respectively. A review of 77 cases of hydatid disease in England and Wales, however, during the period of 1973-1977 failed to show age or sex preponderance. (Communicable Disease Report, 1978.)

Our failure to detect cases might be due to the fact that such cases occur only in exceptional circumstances. Strain variation in *E. granulosus* is well known and it could be that the endemic strain is not as highly infective for man as are some others (Smyth & Davies, 1974). An epidemiological survey, using the Casoni skin test, carried out on healthy persons in two Welsh communities, showed a substantial difference between those living in an endemic area (19.5% reactors) and those in an area free of the condition (1.7%). In the school population of Staffin, where the two school cases occurred in 1974, 13 out of 65 (20%) were shown to react. It is possible that the reactive sera detailed in Table 4, or a proportion of them, represent past exposure to the parasite which failed to develop.

Hydatid disease can be and has been eradicated from endemic areas such as Iceland (Beard, 1973) and Tasmania (McConnell & Green, 1970). A decision or an attempt at eradication of hydatid disease in the Hebrides could be considered for economic, social or medical reasons. The indications are that considerable organization would be necessary, but that laboratory surveillance of the measures would be practicable.

We would like to thank the following for their co-operation during the survey: Mr David Skinner, MRCVS; Mr D. M. Graham; Mr Philip Hill, Chief MLSO, Lewis Hospital; the staff at the Stornoway abattoir; Mr D. Cox, Mr S. Millard, Mr I. Hastings and Mr C. Ross, Raigmore Hospital. In addition we would like to thank Miss C. M. Mackenzie for help with the typescript.

This work was supported by a research grant from the Scottish Home and Health Department.

REFERENCES

- BEARD, T. C. (1973). The elimination of *Echinococcus* from Iceland. *Bulletin of the World Health Organization* **48**, 653-660.
- BRADSTREET, C. M. P. (1969). A study of two immunological tests in the diagnosis and prognosis of hydatid disease. *Journal of Medical Microbiology* **2**, 410-432.
- CAPRON, A., VERSES, A. & BIGNET, J. (1967). Le diagnostic immunoelectrophoretique de l'hydatidose. In *Le Kyste hydatique du Foie* (ed. J. Coudert and P. Goitard), pp. 27-40. Lyon. SIMEP. (Journées Lyonnaises d'Hydatidologie 1966.)
- COMMUNICABLE DISEASE REPORT (1978). Hydatid disease 1973-1977. weekly edition CDR 78/21.
- GENTILINI, M., PISON, J. M. & NIEL, G. (1972). Immunoelectrodiffusion sur membrane d'acetate de cellulose. *Nouvelle Press Médicale* **1**, 1499-1501.
- GUISANTES, J. A., YARZABAL, L. A., VARELA-DIAZ, V. M., RICARDES, M. I. & COLTORTI, E. A. (1975). Standardization of the immunoelectrophoresis test with whole and purified hydatid cyst fluid antigens for the diagnosis of human hydatidosis. *Revista do Instituto de Medicina Tropical de São Paulo* **17**, 69-74.

- MANSUETO, S., MIGNECO, G., TRIPI, S. & PICONE, D. M. (1980). Simplified counter-immunoelectrophoresis (CIEP) with a commercially produced antigen on cellulose acetate membrane for the diagnosis of hydatidosis. *Transactions of the Royal Society of Tropical Medicine and Hygiene* **74** (2), 260-261.
- MCCONNELL, J. D. & GREEN, R. J. (1979). The control of hydatid disease in Tasmania. *Australian Veterinary Journal* **55**, 140-145.
- PICARDO, N. G. A. & GUI SANTES, J. A. (1981). Comparison of three immunological tests for seroepidemiological purposes in human Echinococcosis. *Parasite Immunology* **3**, 191-199.
- POZZUOLI, R., MUSIANI, P., ARRÙ, E., PIANTELLI, M. & MAZZARELLA, R. (1972). *Echinococcus granulosus*: Isolation and characterization of sheep hydatid fluid antigens. *Experimental Parasitology* **32**, 45-55.
- SMYTH, J. D. (1967). Studies on tapeworm physiology. XI. *In vitro* cultivation of *Echinococcus granulosus* from the protoscolex to the strobilate stage. *Parasitology* **57**, 111-138.
- SMYTH, J. D. & DAVIES, Zena (1974). Occurrence of physiological strains of *Echinococcus granulosus* demonstrated by *in vitro* culture of protoscoleces from sheep and horse hydatid cysts. *International Journal for Parasitology* **4**, 443-445.
- THORBURN, H. & WILLIAMS, H. (1972). A stable haemagglutinating antigen for detecting toxoplasma antibodies. *Journal of Clinical Pathology* **25**, 762-767.
- VARELA-DIAZ, V. M., COLTORTI, M., RICARDES, M. I., PREZIOSO, U., SCHANTZ, P. M. & GARCIA, R. (1976). Evaluation of immunodiagnostic techniques for the detection of human hydatid cyst carriers in field studies. *The American Journal of Tropical Medicine and Hygiene* **25**, 617-622.
- VESIKARI, T. & VAHERI, A. (1968). Rubella: A method for rapid diagnosis of a recent infection by demonstration of the IgM antibodies. *British Medical Journal* **2**, 221-223.
- WALTERS, T. M. H. & CLARKSON, M. J. (1980). The prevalence of *Echinococcus granulosus* in farm dogs in mid Wales. *Veterinary Parasitology* **7**, 185-190.