Cystic echinococcosis in Italy from the 1950s to present

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Abstract. In Italy the epidemiological pattern of cistic echinococcosis (CE) is incomplete and the information for most regions is out of date, contradictory, and almost exclusively limited to the intermediate hosts. The disease is found most frequently in particular social and economic conditions: widespread use of extensive or semi-extensive sheep farming, illegal slaughtering, and high numbers of sheepdogs and other types of dogs. The highest incidence in sheep is found in Sardinia (70.6-92.8%), Sicily (6.5-36.5%), Basilicata (5-28%), Abruzzo (22%) and Tuscany (47%). It North Italy, it is never higher than 0.5% in slaughtered sheep. No data are available on the biomolecular characterization of the strains of *E. granulosus* in Italy, apart from Sardinia and recently Lazio. G1 (Sheep strain), G7 (Pig strain) G2 (Tasmanian sheep strain) have been identified in Sardinia and G1 and G3 (Buffalo strain) have been recently isolated in Lazio. In Italy, CE has was also found in buffaloes (2.63-9.8%) and horses (<1%). However, further epidemiological surveys and genotyping study are necessary. The small quantity of up to date information on the diffusion of *E. granulosus* in dogs (Abruzzo 4%, Sardinia 6-10% and Sicily 19.3%) highlights the need for modern, fast, sensitive and low risk diagnostic methods which would provide a true picture of the pattern of the infection in this host.

Key words: Echinococcus granulosus, livestock, dogs, epidemiology, Italy.

Cistic echinococcosis (CE) is an important problem for public health and the economy in the many parts of the world where the infection is endemic or hyperendemic. It is of particular importance in the Mediterranean Region (MR), where it is one of the principal parasitic infection in livestock. It is the most important parasite zoonosis in the area and it is of great social importance. In MR, CE is directly linked to ovine strains (G1) although others of 10 Echinococcus granulosus genotypes (G1-G10) identified in the world have been found in the area. The equine strain (G4) has been found in Spain, Italy, Lebanon and Syria, the camel strain (G6) in North Africa and the Middle East and the swine strain (G7) in Spain, the Slovak Republic and Poland (Thompson and McManus, 2002).

CE is a particular problem in the areas where grazing, and in particular pastoralism, is widespread. The close relationship between dog, sheep and man, makes more likely that the life cycle of the parasite can be completed. At the moment, the sheep strain (G1) seems to be the only one which is found in all the MR and it is the main responsible for the disease in humans. Its high prevalence is closely connected to the following factors, which are linked to the social and economic conditions of the population: continued widespread use of traditional techniques when raising small ruminants (extensive or semi-extensive grazing), illegal slaughtering of the animals, and the presence of high number sheepdogs which gravitate to the sheep raising areas.

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Although there are numerous studies on the geographical distribution and the prevalence and incidence of CE in animals, these are fragmented both in geographical area and time, and of little use in defining a sufficiently precise pattern of the epidemiological situation. Only partial and sporadic studies have been made on CE in the countries on the Southern and South Eastern areas of the Mediterranean basin. Also data from EU Mediterranean countries is not complete, being limited to some geographical areas. Although the data has to be treated with caution, it indicates that in certain countries the disease is alarmingly widespread and that the infection has to be considered endemic or highly endemic in most MR countries, with the exception of Malta and the Republic of Cyprus where the infection has almost completely disappeared (Seimenis and Battelli, 2003).

The epidemiological pattern of CE in animals is also incomplete in Italy. The data for numerous regions is often contradictory, as it is often limited to information on the prevalence of the disease in slaughtered animals and is also out of date. Data on the diffusion of the parasitosis in dogs is always limited to a number of areas. There are no systematic epidemiological investigations which document how many, and what strains of E. granulosus are actually present in the country. Recent studies in Sardinia have found G1 (Sheep strain) in sheep, cattle and pigs and G7 (Pig strain) in pigs (Varcasia et al., 2004a). Further research has found G2 (Tasmanian sheep strain) in cattle in Sardinia and G1 and G3 (Buffalo strain) in pigs in Lazio (Busi et al., 2004). CE has been found in horses with G4 (Horse strain), although its presence has not been confirmed by molecular investigation.

The first investigation of CE in slaughtered animals, which covered all areas of Italy, dates back to 1952 (Pellegrini and Cilli, 1955). This confirmed, as had previously been observed, that the parasitosis was more widespread in the islands and South Italy. It was found in 4.08% of cattle in North Italy. 11.45% in Central Italy, 13.27% in Southern Italy and 55.09% in Sardinia. The figures for its presence in sheep were as follows: 15.93% in North Italy; 14.94% in South Italy; 21.74% in Central Italy with a peak of 41.67% in Tuscany; 68.72% in Sardinia; 21.44% in Sicily. It was found in 1.82% of goats in North Italy (6.22% in Piedmont), 8.14% in Central Italy (31.37% in Tuscany), 10.33% in South Italy, 7.55% in Sicily and 12.75% in Sardinia. Swine CE varied from highs of 19.81% in Sardinia, 14.66% in Basilicata, and 13.24% in Umbria, to lows of 3.21% in Sicily and 2.01% in Emilia.

Using annual data collected by the Ministry of Health in 1972-1977, Schiavo et al., (1979) found the following prevalence: sheep 11.6%, goats 5.6%, cattle 1.54%, pigs 1.11%, horses 0.45%. Another investigation which provided "a reasonably approximate pattern, even though underestimated" of the diffusion of CE in Italy was that by Romboli et al. (1980), which used official information and data. From the national data for 1968-1978, they found that CE was present in 8.1-15.2% of cattle, 8.1-15.3% of sheep, 2.7-8.9-% of goats 0.7-1.2-% of and 0.4-0.9% of horses. Regional data for 1972-1978 in the different animal species confirmed that CE was more prevalent in all species in Central and South Italy and the islands than it was in North Italy.

Finally the more recent investigation by Lorenzini and Ruggieri (1987) confirmed that the parasitosis was more widespread in South Italy and the islands and in particular to those areas where animal husbandry was less developed and the animals, mostly sheep, were raised using traditional methods. The study was limited by the method of collecting data (questionnaires sent to the principal abattoirs in each province, and the fact that the data for sheep and goats was aggregated).

The results of national investigations on cattle and buffalo slaughtered by Inalca S.p.A (Ospedaletto Lodigiano), found CE in less than 1% of cattle in North Italy and in 2.63% of buffaloes. In Central Italy, the prevalence in cattle was far higher (10%-12.53%), while in buffaloes it was 3.53%. The figures for South Italy were even higher: cows 22.04%, bulls 11.11%, calves 1.85% (CE was absent in calves in other areas), buffaloes 9.8% (Fattori *et al.*, 2000). This data confirms that the parasitosis is not widespread in North Italy but also that it is not diminishing in the rest of the country.

There is much less epidemiological data for North Italy than there is for the South and islands, and it is generally data collected from slaughterhouses. Data are limited for Valle d'Aosta, Piedmont and Emilia-Romagna. In Valle d'Aosta, CE prevalence

ranged 0.08% to 0.18% in "slaughtered animals" in 1995-2003 (data supplied by Valle d'Aosta Health Authorities). The "2002 Report of the veterinary services of the Piedmont Region Health Services" found CE in 0.011% of slaughtered cattle. It was found in 0.51% of slaughtered sheep and 0.085% of sheep in general. However deeper examination of the reports shows that the prevalence was calculated for, respectively, the number of "sheep and goats" slaughtered and the total number of sheep and goats registered in the animal husbandry survey. Faggioli et al. (2001) found that the presence of CE in legally slaughtered animals in Emilia Romagna between 1996 and 1999 was as follows: pigs 0.95/million, cattle 0.39%, sheep 0.30%, goats 0.39%, horses 0.34%.

There is no information on the diffusion of canine echinococcosis in the northern Italy; the CA-ELISA immunoenzymatic method (Echinotest, Bommelli, Bern, Switzerland) found positive for the infection 9 of 19 (alpine) farm dogs and 27% of 60 in the Val di Susa (Rossi, pers. comm., 2004).

There is more data available for Central and South Italy, but it is still not enough to create a sufficiently clear pattern of the spread, and in particular of the infection trend over time. Let us take the Abruzzo region as an example. The evolution of the parasitosis in livestock, and in particular in sheep and goats, does not appear to be linear because in some periods the region was one of the most affected ones and in others the infection prevalence decreased. In 1981, Manilla (1986) found EC in 50.8% of adult sheep in Teramo. CE was present in an average of 10.6% of "sheep and goats" in the period 1972-1984 (Gargiulo et al., 1987). In a study confined to adult animals in the period 1985-1989, Schiavo *et al.* (1992) found a reduction in the number of positive cattle (3.5%-2.3%) and horses (3.8%-1%) and a great stabilization of the situation in pigs (0.3%-0.6%). In goats there was a fall from 16.3% to 4.5%, but with a peak of 22.5% in 1988. On average, the situation for sheep was stable during the 5 year period. More recent data for the province of Teramo (1985-1994) shows that CE was present in an average of 32.14% of "sheep and goats" and in between 4 and 6% of cattle, horses and pigs (Tieri and Gatti, 1995). Recent surveys ("Research Programs Relevant National Interest" PRIN 2003), found CE in 22% of 2-5 year old sheep in the province of Teramo, with total fertility of 4.6%. The only data available for dogs in Abruzzo showed 4% of E. granulosus positive dogs, of which 2.66% were stray dogs and 50% sheepdogs (Di Ventura et al., 1995).

The last epidemiological data showed that CE was found in 47% of sheep slaughtered in the province of Arezzo (Bio and Fagiolo, 2004).

These high prevalences in the intermediate hosts confirms that in Central Italy the previous available data were probably underestimated and further investigations are necessary to better understand the diffusion of the infection.

Past research in Apulia found a reduction in E. granulosus in dogs from 12.9% in 1955-1958, to 10.51% in 1962-1963 and 5.73% in 1971-1974 (Puccini et al., 1975). There were more epidemiological investigations into CE in intermediate hosts. Between 1975 and 1982, CE was found in an average of 4.88% of sheep and 3.9% of goats (Puccini and Tassi, 1983). In the period 1989 to 1993, Schiavo and Pansini (1996) found that the presence of CE varied between 3.2% and 0.47% in sheep and 5.88% and 0.35% in goats slaughtered by Health Service 1 of Taranto. Puccini (pers. comm.) found CE present in the following amounts in 4 slaughterhouses in the province of Lecce in the period 1978-1987: cattle 7%, sheep 14%, and goats 12%. In the slaughterhouse of Foggia in 2003, CE was present in the following amounts: cattle 5.74%, adult sheep 5%, horses 0.02% (Puccini, pers. com.).

In 1996-2002 in Basilicata, Quaranta (2003) found CE in 2-3% of cattle, 5-28% of sheep, 4-25% of goats, 0.05-0.5% of pigs and 0.04-0.1% of horses. Surveys in Campania in cattle, sheep, goats, swine and horses slaughtered in 26 abattoirs in the provinces of Avellino and Salerno found an average presence of less than 5%. In certain districts, the presence in sheep and cattle was between 16% and 21% (Cringoli *et al.*, 1998). Capurso *et al.* (1968) found relatively high diffusion in dogs in the city of Naples and that it was present in 1% of dogs in some districts of the province.

Finally, the finding of CE in buffaloes, not only in this region but also in other areas of Italy, highlights the need for further research aimed to establish the diffusion, the epidemiological situation which favours the presence of the parasite in this species and to clarify which specific genotypes are responsible for it.

There has been more research on the parasitosis in Sicily. The first research was carried out by Bertocchi (1951) who found CE in 6-10% of cattle slaughtered in Palermo, Messina and Catania and 2.08% of dogs in the province of Palermo. Panebianco and Sciutteri (1955), found CE in 4.6% of dogs in Messina and slightly less (3.4%) was found in Palermo (Gallo and De Girolamo, 1960). By contrast, Virga and Giannetto (1998) found that the prevalence in sheepdogs treated with arecoline hydrobromide was significantly higher in the provinces of Agrigento (23.2%) and Palermo (16.2%). Samples taken between 1981 and 1985 at the Palermo slaughterhouse found the highest prevalence in sheep was 29.41%, in cattle 2.87%, in horses 2.69% and in pigs 0.32% (Demma et al., 1987). In 1988-1989, Virga (1991) examined the registrations carried out by the veterinary inspectors to comply with the O.M. 21 April 1964, and found the following highest prevalences: cattle 2.29%, sheep/goats 10.90%, pigs 0.82%, and horses 3.1%. Magliarditti and Niutta (1995) found an even serious situation for local adult animals, with CE present in 11.13% of cattle, 43.22% of sheep, 2.85% of goats and 4.71% of pigs. Poglayen et al. (2001) found 49% positive results in the Sicilian Black Pig of the Nebrodi Park (ME) and more recent investigations (1988-2000) found CE present in 15.6% of them and fertility of 1.9% (Scala et al., 2001). Later Poglayen et al. (2003), found CE in 15% of legally slaughtered sheep in 8 provinces, with a maximum of 36.5% in Palermo and a minimum of 6.5% in Agrigento. They also found an average fertility of 31%. This varied from 90% in Agrigento to 13% in the provinces of Enna, Messina and Ragusa. Finally Taenia hydatigena was found in 10.5% of the dogs in the municipal dog pound of the province of Trapani (Nobile et al., 1993) and E. granulosus in 19.3% and T. hydatigena in 17.5% of sheepdogs in the provinces of Agrigento and Palermo (Giannetto et al., 1997). This confirms how easy it is for the dogs to have access to the viscera of slaughtered animals or of those which had a natural death. While there was not enough data to establish a complete pattern of the situation on the island, it was clear that there were risks for man and domestic animals, above all in rural areas where less advanced social conditions and animal husbandry encourages the spread of CE.

The presence of CE in horses in various regions of South and Central Italy and in Sicily means that greater epidemiological awareness is necessary not only for buffaloes, as was said above, but also for this species, and bio-molecular research must be carried out to establish if G4 (horse strain) is present in Italy, given that biological tests and morphometric investigations by Macchioni and Gallo (1967) found that it was present in Sicily.

Unlike in other areas of Italy, CE in Sardinia has always been investigated organically as can be seen from the large quantity of literature on the subject, although due to limitations of space it is impossible to cite these exhaustively. The island has always been an ideal model for the study of the parasitosis. Firstly there are a large number of sheep and goats (about 3,000,000 sheep and 250,000 goats), with an advanced branch of production which has encouraged advances in the whole sector. Secondly the high number of dogs (150,000) (Cannas et al., 1990), the sheep farming methods used (extensive, semi-extensive), illegal slaughtering and the social economic and cultural conditions of the shepherds have resulted in the level of CE in sheep remaining at 87%, in goats at 24%, in cattle at 30% and in pigs at 20%. The fertility rate varies from 40% in sheep to 31% in goats, 25% in cattle and 25% in sheep (Arru et al., 1990; Conchedda et al., 1997). In dogs it is found on average in 11% of stray dogs and 25.42% of sheepdogs with an overall average regional presence of 16.2% (Arru et al., 1990). In addition, despite the fact that three eradication campaigns have been conducted in Sardinia (in 1960, 1978 and 1987) (Arru et al., 1999), recent data showed that the prevalence of CE was still high, being present in 75.6% of sheep in the province of Sassari, with fertile cysts being found in 6.9% of the animals examined and in 9.1% of those infected (Scala et al., 2000a). Research in the province of Cagliari (Scala et al., 2000b) in 1999 found similar results, with CE present in 72.2% of sheep. The situation found in 2000 in Goceano, a district of Central Sardinia, between the provinces of Sassari and Nuoro is still more worrying, with CE present in 92.8% of sheep and fertile cysts in 27.1% (Soro et al. 2002). The preliminary results of a second study being carried out at present on animals legally slaughtered in the provinces of Sassari and Nuoro show the levels of infection (82.6%) and fertility (17.3%) to be significantly higher than those of the first study which were, respectively, 70.6% and 7.3%. This confirms that the parasitosis is closely linked to certain areas where traditional methods of sheep farming are used. After more than a decade of silence on the diffusion of E. granulosus in definitive hosts, research on this was carried out in 2003 as part of the research project PRIN 2003. Faecal sample were taken from 300 dogs and microscopic examination of these found 8.25% Taenia spp. positive. CA-ELISAs performed with the commercial kit (Echinotest, Bommeli CH) found 3% positive while two ELISA which employed monoclonal antibodies (Mabs: EmA9 and EgC3) found 6% and 10% positive respectively (Varcasia et al., 2004b).

It is thus evident that at present, with the exception of the previously cited cases, there is a lack of epidemiological and diagnostic research and molecular characterization studies in Italy, both in dogs and in intermediate hosts. For example, data of cyst fertility have only been collected in the Italian islands and principally in Sardinia. There are few studies on the prevalence of the parasitosis in wild animals (particularly useful when the same strain of E. granulosus is present in both domestic and wild animals). Research carried out by Arru et al. (1986) found CE was present in 1.18% of foxes in Sardinia while Guberti et al. (1983) found CE in 16.9% of wolves. The positive results of immune-enzymatic CA-ELISA tests on faecal samples from 6 wolf packs (Canis lupus) in Piedmont (Val di Susa) and of fertile cysts in sheep carcasses which had been preyed on by wolves in the same area (Rossi, pers. com.) confirm that more studies are needed in this field.

In conclusion, it is clear that there is very little information on the diffusion of canine echinococcosis and also that there is a grave lack of knowledge on the epidemiological situation for CE in intermediate hosts, with the exception of the few examples. This is particularly true in certain areas of Central and South Italy where sheep farming is widespread. Standard data from the Public Health Services is of itself insufficient to determine the existence or absence of situations of possible risk in particular zones, or at the local level, and is statistically of such questionable value that it makes accurate evaluation of the evolution of the parasitosis over time more difficult.

Given that CE is still widespread in certain areas where the relationship between dogs, humans and sheep is very close, it is of great importance for public health to create a rapid and sensitive method for diagnosing the parasitosis in dogs, as they are the sole transmitter of the parasitosis to humans and animals.

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