Published in final edited form as:

Acta Trop. 2010 March ; 113(3): 248–256. doi:10.1016/j.actatropica.2009.11.006.

Widespread co-endemicity of human cystic and alveolar echinococcosis on the eastern Tibetan Plateau, northwest Sichuan/southeast Qinghai, China

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Abstract

Cystic echinococcosis (CE) or hydatid disease is known to be cosmopolitan in its global distribution, while alveolar echinococcosis (AE) is a much rarer though more pathogenic hepatic parasitic disease restricted to the northern hemisphere. Both forms of human echinococcosis are known to occur on the Tibetan Plateau, but the epidemiological characteristics remain poorly understood. In our current study, abdominal ultrasound screening programs for echinococcosis were conducted in thirty-one Tibetan townships in Ganze and Aba Tibetan Autonomous Prefectures of northwest Sichuan Province during 2001–2008. Hospital records (1992–2006) in a major regional treatment centre for echinococcosis in Sichuan Province were also reviewed. Of 10,186 local residents examined by portable ultrasound scan, 645 (6.3%) were diagnosed with echinococcosis: a prevalence of 3.2% for CE, 3.1% for AE and 0.04% for dual infection (both CE and AE). Human cystic and alveolar echinococcosis in pastoral areas was highly co-endemic, in comparison to much lower prevalences in semi-pastoral or farming regions. The high ultrasound prevalence in these co-endemic areas in northwest Sichuan Province was also reflected in the hospital study, and hospital records furthermore indicated another possible highly co-endemic focus in Guoluo Prefecture of Qinghai Province, located at the border of northwest Sichuan. These chronic cestode zoonoses constitute an unparalleled major public health problem for pastoral Tibetan communities, and pose great difficulties for adequate treatment access and effective transmission control in such remote regions.

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Keywords

Cystic echinococcosis; Alveolar echinococcosis; Ultrasound; Prevalence; Tibetan; Sichuan Province; Qinghai Province

1. Introduction

Human echinococcosis refers to infection with the larval (metacestode) stage of zoonotic cestodes (tapeworms) belonging to the genus *Echinococcus*. Four main species were recognized until recently, namely, *Echinococcus granulosus*, *E. multilocularis*, *E. oligarthrus* and *E. vogeli* (Rausch et al., 1972; Kumaratilake and Thompson, 1982). A new (fifth) species of *Echinococcus*, named *E. shiquicus*, has recently been described by our team in wildlife hosts from the eastern Tibetan Plateau, China (Xiao et al., 2005), however its infectivity to humans is unknown (Li et al., 2008). All the classic four recognized *Echinococcus* species of carnivores can infect humans (ie. zoonotic) and may cause three clinical forms of echinococcosis, i.e. cystic echinococcosis (CE) caused by *Echinococcus granulosus*, alveolar echinococcosis (AE) caused by *E. multilocularis*, or polycystic echinococcosis due to *E. vogeli* or *E. oligarthrus*. The distribution of *E. granulosus* is cosmopolitan and is the predominant cause of human echinococcosis worldwide (McManus, 2002). Transmission of *E. oligarthrus* and *E. vogeli* is restricted to Central and South America where sporadic cases may occur, especially due to the latter species (D'Alessandro, 1997). *E. multilocularis* is also a relatively rare parasitic disease in humans and is restricted to the Northern Hemisphere, with primary transmission in wildlife (cycling between foxes and rodents). Human AE cases have however occurred more frequently in foci in Alaska, northern and central Europe, Central Asia, Siberia, China and Japan (Craig, 2003).

In China, human CE has been demonstrated to be widespread in at least twenty-one of its thirty-one provinces, but was more prevalent in the following northwest Provinces or Autonomous regions: Qinghai, Gansu, Sichuan, Ningxia, Xinjiang, Inner Mongolia, Tibet and Yunnan (Shi, 1997; Wen and Yang, 1997; Craig, 2004). From the 1990s active mass screening surveys using portable ultrasound began to reveal very high prevalence rates of human alveolar echinococcosis in several agricultural counties of Gansu and Ningxia provinces (Craig et al., 1992; Yang et al., 2006), and more surprisingly in pastoral Tibetan communities in western Sichuan Province (Li et al., 2005).

With the aim of understanding the epidemiology of human echinococcosis (both CE and AE) in Tibetan communities of the eastern Tibetan Plateau, mass screening programs for echinococcosis were conducted in thirty-one Tibetan townships located in western Sichuan Province. In addition, hospital records of echinococcosis patients post-operatively confirmed were reviewed from one of the main treatment centres in the region.

2. Materials and methods

2.1. Study sites

Parts of populations in a total of thirty-one townships, located in Ganze and Aba Tibetan Autonomous Prefectures of northwest Sichuan were screened by mass ultrasound. Participants in the study were selected on a voluntary basis. The involved townships included twenty-four pastoral townships within Ganze and located in the counties of Shiqu, Seda, Baiyu, Ganzi, Dege or Yajiang, where altitudes ranged from 3700m to 4500m, and the main occupation of local residents was raising yaks and sheep/goats as the primary source of income. In addition, three farming (primary agricultural) townships were selected from Yajiang and Danba Counties (Ganze Prefecture) and Maerkang County (Aba Prefecture), with altitudes varying from 2010m

to 2680m Four semi-pastoral townships were also included and were located in Rangtang County (Aba Prefecture), with altitudes ranging from 3451m to 3600m, where local people subsisted on both agricultural and livestock grazing (Figure 1). Screening programs for abdominal echinococcosis using portable ultrasound (GE, LOGIQ α 100, Wuxi, China) were performed in Spring or early Winter during 2001-2008 in the selected sites. In cooperation with County level Centers for Disease Control (CDC) and local health administrators (cadres), information about the purpose of the screening program was spread to the villagers and townships. Volunteers were self-selected by informed consent and were assured free diagnosis and medical treatment with long-term albendazole drug therapy for echinococcosis if diagnosis was indicated. Recommendation was also provided for possible surgical intervention (cyst/lesion removal or drainage) if appropriate. Persons with other infections or medical conditions were examined and referred to local health clinics for further investigation or treatment.

2.2. Questionnaire

For each self-selected participant a questionnaire was completed using Tibetan registration auxiliaries, designed to obtain information on demographics, animal ownership and potential risk factors for echinococcosis including the source of drinking water, dog ownership, the frequency of dog contact, ownership of fox skin products etc.

2.3. Criteria for diagnosis and classification of echinococcosis

Diagnosis and classification of cystic echinococcosis (CE) was made using portable ultrasound according to the criteria proposed by the World Health Organization Informal Working Group on Echinococcosis for CE (Pawlowski et al., 2001). On the basis of conformational features of cysts, CE lesions (primarily in the liver) were differentiated into 6 types including CL, CE1, CE2, CE3, CE4, and CE5. In this study, CL cysts of a parasitic origin were exclusively counted, and CL cysts of a non-parasitic origin were excluded by comprehensive analysis of other factors, particularly such as the patient age, and partially by further observation during follow-up. In general, the CL type refers to a cystic lesion without clear rim indicating the parasite is at an early stage of development if the cyst is of a parasitic origin, while the presence of CE1 or CE2 is suggestive of active stages of the disease, while CE3 suggests the parasite is at a transitional stage, and CE4 or CE5 implies an involution, necrotic or inactive parasite (Pawlowski et al., 2001).

Diagnosis of AE was dependent on detection of distinctive tumour-like lesions in the liver characterized by a non-homogeneous hyperechoic structure and with poorly defined verge, containing scattered calcifications, and/or a central necrotic cavity with a hyperechoic pseudoliquid structure (Pawlowski et al., 2001; Bresson-Hadni et al., 2006) and further classification was based on the criteria proposed previously (Li et al., 2004) and the PNM system (Kern et al., 2006). Briefly, on the basis of the features and the size of lesions, AE lesions were classified into three types and eight subtypes which indicated different stages of the disease. AE lesions ≤ 5 cm, normally without central necrosis detected, were confirmed as AE1 and differentiated further as, AE1s (single lesion) and AE1m (multiple lesions), which indicated an early stage of the disease. Alveolar lesions measured $>5\text{cm} \leq 10\text{cm}$ were classified as AE2 including three subtypes recorded as AE2s (single lesion), AE2m (multiple lesions) and AE2f (presence of central necrotic fluid, regardless of the number of lesions), suggestive of a developing parasite, while AE lesions measured $>10\text{cm}$ at diameter were confirmed as AE3 indicative of an advanced stage of the disease, which include three subtypes, i.e. AE3s (single lesion), AE3m (multiple lesions) and AE3f (presence of central necrotic fluid, regardless of the number of lesions).

2.4. Serology

Persons with confirmative or suspected CE/AE lesions or with other space-occupying lesions in the liver were asked to give a five ml venous blood sample for detection of *Echinococcus* antibodies using ELISA with recombinant AgB as antigen for CE or AE, and ELISA with recombinant Em18 antigen for AE, as described elsewhere (Sako et al., 2002; Xiao et al., 2003; Mamuti et al., 2004).

2.5. Hospital study

All patients who were treated surgically for echinococcosis in Aba Military Hospital (located in Maerkang, Aba Prefecture, Sichuan Province) (Figure 4) during 1992 to 2006, and post-operatively confirmed by histopathology as echinococcosis, were included in this study (n=1312). Further information about age, gender, domicile, ethnicity, and post-operative diagnosis, etc was collected. This hospital has 200 beds of which 95% are used to admit non-military patients.

Diagnosis of echinococcosis before operation was made by abdominal B-ultrasound examination and/or computed tomography (CT) (Ren et al., 2008). Post-operative confirmation was made by histopathology, as well as by PCR and DNA sequencing of isolates for some cases (Li et al., 2008). Briefly, morphological identification of CE infection was based on the observation of the structure of cystic lesions characterized by unilocular cysts with a thick laminated layer, presence of a germinal layer, and/or brood capsules with protoscoleces. For AE histopathology, the presence of large numbers of vesicles with different sizes and shapes with a thin laminated layer, and concurrence of distinct hyperplasia of fibro-connective tissue and cellular infiltration of eosinophils, lymphocytes and plasma cells, resulted in a diagnosis.

2.6. Statistical analysis

Data were analyzed using Epi Info™ (version 3.5; Centers for Disease Control and Prevention, Atlanta, GA), statistical significance was set at $P < 0.01$.

3. Results

3.1. Mass screening using ultrasound

A total of 10,186 participants originating from Tibetan communities of thirty-one townships in nine counties in Sichuan Province were registered by questionnaire and examined by abdominal ultrasound. Population sample (age ranged from 1-92 years; median 32.8 years) comprised 50.4% (5133) females and 49.6% (5053) males. Persons of Tibetan ethnicity comprised 96.1% of the sampled population. The other participants listed their ethnicity as Han (3.9%). Questionnaire data revealed 46.9% (4781) were herdsman who raised livestock including yaks, sheep and/or goats as the primary source of their income. Other listed occupations included student (19.1%), farmer (12.2%), part-time herdsman (4.5%), public servant (7.4%), preschooler (3.6%), and other (6.3%).

3.1.1. Prevalence of human echinococcosis—Of 10,186 volunteers examined by abdominal ultrasound scanning, 311 (3.1%) were confirmed to have AE infection, 330 (3.2%) to have CE, and 4 (0.04%) individuals to have dual infection with both CE and AE (Table 1, Figure 2 and Figure 3).

Of 315 persons with a confirmative image of AE lesions (including four persons with dual infection), 74 (23.5%) had hepatic lesions of AE1 type (67 AE1s and 7 AE1m), 142 (45.1%) had AE2 lesions in the liver (75 AE2s, 20 AE2m and 47 AE2f), while hepatic lesions characterized by AE3 were detected in 99 (31.4%) individuals (34 AE3s, 3 AE3m and 62 AE3f) (Table 2). In addition, 136 single AE lesions were located in the right hepatic lobe, and 36 were

in the left hepatic lobe. Involvement of both lobes was observed in 79 AE cases. More than one alveolar lesion was detected in the liver in the remaining 64 cases.

Of 334 persons who presented with cystic images indicative of CE cysts (including 4 cases with dual infection), nine (2.7%) were detected to have CL type lesions, 103 (30.8%) were classified to have CE1 lesions, 98 (29.3%) had CE2 cysts, 12 (3.6%) had CE3 lesions, 80 (24.0%) and 17 (5.1%) had CE4 or CE5 type cysts, respectively. Furthermore, multiple CE lesions with mixed type cysts were detected in an additional 15 cases (4.5%), i.e. 5 patients with CE1 and CE2 cysts, 3 with CE1 and CE4 cysts, 5 with CE2 and CE4 cysts, 1 with CE2 and CE5 cysts, and 1 with CE3 and CE5 type cysts (Table 2). In addition to the liver, one or more CE cystic lesions were identified in the peritoneal cavity in 24 patients, 7 cases in the spleen and 1 case in the kidney.

3.1.2. Echinococcosis prevalence at township level—Of 7773 volunteers originating from 24 pastoral townships that were examined by ultrasound, 324 (4.2%) were confirmed to have CE infection, 301 (3.9%) to have hepatic AE infection, and 4 (0.04%) persons presented with dual infection (both CE and AE). The overall prevalence of echinococcosis in pastoral townships ranged from 2.0% (10/492) in Seke Township, Seda County to 21.0% (21/100) in Mengyi Township, Shiqu County. The highest AE prevalence (9.4%, 59/631) was recorded in Yiniu Township of Shiqu County, and the highest CE prevalence (14.0%, 14/100) was recorded in Mengyi also in Shiqu County. No AE cases were detected in Honglong Township, Yajiang County, however CE prevalence was 2.3% (Table 1). Therefore, both human cystic and alveolar echinococcosis was shown to be highly co-endemic in 23 of 24 pastoral townships studied in western Sichuan (Table 1).

In contrast, a much lower prevalence of human echinococcosis was recorded in farming or semi-farming townships, i.e. only 5 CE cases (3 CE4 and 2 CE5 advanced type cysts) were detected in 3 farming townships, with a prevalence of 0.3% (5/1738). Furthermore, only 0.1% (1/675) and 1.5% (10/675) of local people in 4 semi-farming townships had CE or AE infection, respectively (Table 1).

3.1.3. Prevalence by gender and age—Of 645 persons with an evidence of abdominal echinococcosis, 394 (CE=212, AE=179, dual infection=3) were female, and 251 (CE=118, AE=132, dual infection=1) were male. In other words, the prevalence of echinococcosis (CE or AE) in females was 7.7% (394/5133) (4.1% for CE, 3.5% for AE and 0.1% for dual infection), and 5.0% (251/5053) for male (2.3% for CE, 2.6% for AE and 0.02% for dual infection). Statistical analysis revealed that the prevalence of echinococcosis in females (7.7%) was significantly higher than that for males (5.0%) ($\chi^2=31.49$, $p<0.01$).

Among the 645 persons with abdominal detectable *Echinococcus* lesions, the average age was 40.9 years. The youngest patient with CE was 3 years and the oldest was 81 years, and the average age of CE cases detected by ultrasound was 38.6 years (n=330). However, age specific prevalence of CE cyst type varied. The average age of subgroup population with CE cysts at an early stage (CL) lesions was 11.3 years (n=9), while average age group with CE cystic lesions at active stages (CE1, CE2), at transitional stages (CE3) or inactive stages (CE4, CE5), was 38.2 years (n=201), 36.3 years (n=12) and 43.5 years (n=97), respectively. The average age of persons diagnosed in the communities with hepatic AE was 43.4 years (age range 6-81 years). The subgroup of cases with early AE1 lesions had an average age of 39.7 years (n=74), while the mean age of the population with AE2 lesions was 44.7 years (n=142), and 44.0 years for the subgroup with advanced AE3 lesions (n=99). In addition, the average age for 4 persons with dual infection was 39.8 years (ranging 11 to 66 years).

For human AE prevalence showed an increase with age, which peaked at the >60 years group (6.5%). Further analysis revealed a significant difference of AE prevalence between age groups ($\chi^2=108.91$, $p<0.01$, 6 degrees of freedom). For abdominal CE prevalence peaked at 5.0% in the > 40 to ≤ 50 years age group, and then decreased. The four persons with dual CE and AE infection were detected across the age cohort (Figure 5).

3.1.4. Prevalence of echinococcosis by occupation—Herdsman had the highest risk for abdominal echinococcosis infection, with a total prevalence of 10.6% (505/4781) (5.5% for CE and 5.0% for AE). Farmers had the lowest prevalence of 0.4% (5/1245) (0.4% for CE). For other occupations, the prevalence was recorded as 2.2% (10/453) for part-time herdsman (0.2% for CE and 2.0% for AE), 1.7% (34/1944) for students (0.9% for CE, 0.8% for AE and 0.05% for dual infection), 1.9% (7/371) for preschooler (1.6% for CE and 0.3% for AE), 3.9% (29/749) for public servants (2.0% for CE and 1.9% for AE), and 8.6% (55/643) for others (3.6% for CE, 4.8% for AE and 0.2% for dual infection). The prevalence for both CE and AE combined proved to be of statistical significance between occupations ($\chi^2=325.28$, $p<0.01$, 6 degrees of freedom).

3.2. Serology

A total of 191 (77.6%) CE sera and 147 (85.0%) AE sera showed positive response to *E. granulosus* recombinant antigen B (rAgB), while 28.9% (71/246) CE sera and 87.3% (151/173) AE sera were recognized by *E. multilocularis* recombinant Em18 (rEm18). Moreover, significant differences of seropositivities with ELISA-rAgB or ELISA-rEm18 were observed in CE/AE cases with echinococcal lesions at different stages. Detailed description of the serology profiles was reported elsewhere (Li et al., 2010).

3.3. Aba hospital study

During 1992-2006, records were identified for 1312 patients (male=719, female=593) with post-operatively confirmed echinococcosis in Aba Military Hospital. The majority 79.6% (n=1044) were confirmed as CE, and 268 as AE infection. Persons of Tibetan ethnicity comprised 97.4% (1278/1312) of these patients, and all the others were Han Chinese. The mean age of 1292 patients (information about age was not available for the other 20 patients) was 42.8 years, ranging from 4 to 81 years. The youngest CE case was 4 years old and the oldest 81 years, with a mean age of 42.9 years (n=1027). While the average age of AE cases was 42.5 years (n=265), with an age range of 8-79 years. The main treatment used for CE was endocystectomy, and for AE cases resection of total/partial alveolar lesion.

Of 1044 patients treated with CE infection, 84.8% (885) had cystic lesions located in the liver, 11.7% (122) in the abdominal cavity or pelvic cavity, and 1.6% (17) in both abdominal cavity and pelvic cavity as well as the liver. Involvement of other organs or tissues was also recorded: spleen CE (1 case), subcutaneous CE (8 cases), lung (4 cases), brain (3 cases), vertebra (2 cases), bone (1 case), and eye (1 case).

Of 268 AE cases treated, alveolar lesions were detected only in the liver in 93.7% (251) of patients. In the remaining 17 AE patients, other organs were also involved, i.e. the brain (5 cases), lung (5 cases), brain and lung (1 case), vertebra (5 cases) and bladder (1 case).

The majority (99.9%) of the 1312 echinococcosis patients treated at Aba Military Hospital lived in Sichuan or Qinghai provinces, and only one CE patient came from the Tibetan Autonomous Region (Changdu County) (Figure 4). For 1044 CE cases, 56.5% (590) originated from 17 counties of Sichuan Province. The remaining 453 (43.4%) CE cases came from 5 counties of Guoluo Prefecture of Qinghai Province (Figure 4). Of 268 AE cases treated at Aba Military Hospital, 150 (56.0%) resided in 13 counties of Sichuan Province, while 118 (44.0%)

AE cases lived in the same 5 counties of Guoluo Prefecture, Qinghai Province, i.e. Banma, Dari, Gande, Jiuzhi, and Maqin counties (Figure 4).

4. Discussion

Human cystic echinococcosis (CE) caused by *E. granulosus*, results in more lost DALYs (disability adjusted life years) globally than that due to onchocerciasis or Chagas disease (Budke et al., 2006), with the greatest echinococcosis burden in Central Asia and China (Craig et al., 2007). The Chinese Ministry of Health carried out a national survey in 2002 for 8 important parasitic diseases, and found the prevalence of human CE was highest (2.5%) in Tibetan communities (Craig et al., 2008). In western China human alveolar echinococcosis, caused by *E. multilocularis*, is also endemic albeit with a more focal distribution than CE and generally considered mainly to be a zoonotic disease in poor Han or Hui upland agricultural communities (Craig et al., 1992; Yang et al., 2006). Human CE and AE cause chronic cystic or vesicular lesions respectively, which may eventually be highly pathogenic and very difficult to treat (WHO, 1996; McManus et al., 2003).

We now demonstrate the occurrence of a major co-endemic focus of human CE/AE in Tibetan pastoral communities present in northwest Sichuan and southeast Qinghai at the eastern edge of the Tibetan Plateau. The average total ultrasound prevalence of human abdominal echinococcosis in 10,186 persons screened in two Tibetan Autonomous Prefectures of northwest Sichuan was 8.1% (4.2% for CE, 3.9% for AE, and 0.05% for dual CE/AE infection). The highest co-endemicity (10.6% total prevalence) at community level occurred in pastoralists (herdsmen occupation), in which approximately half the cases detected were due to the most pathogenic form AE. Human echinococcosis cases were also detected after mass screening in agricultural/semi-farming Tibetan communities but with significantly lower ultrasound prevalence (<0.5% for either disease). This high co-endemicity for CE and AE at pastoral community level in northwest Sichuan, was also verified by examination of hospital records in one important treatment centre in Aba Tibetan Autonomous Prefecture, and also identified CE and AE cases originating from another highly co-endemic pastoral region, i.e. Guoluo Tibetan Autonomous Prefecture in Qinghai Province (Han et al., 2006; Yu et al., 2008), which borders northwest Sichuan.

A total of 645 cases of human cystic (n=330) and alveolar (n=311) echinococcosis, as well as 4 mixed CE/AE cases, were identified by mass ultrasound screening in 8 counties of west Sichuan Province, i.e. counties of Shiqu, Seda, Ganze, Baiyu, Dege, Yajiang, Rangtang and Maerkang. Patient records from Aba Military Hospital in Maerkang (Sichuan) between 1992-2006, for 1312 confirmed echinococcosis cases (n=1044 CE, n=268 AE), identified patient domicile in an additional 5 counties in neighbouring Qinghai Province, i.e. counties of Banma, Dari, Jiuzhi, Gande and Maqin. More detailed analysis of these hospital cases has been reported elsewhere (Ren et al., 2008).

Western China has long been known to be endemic for human cystic echinococcosis (CE) (Craig et al., 1991), however the distribution of human alveolar echinococcosis (AE) the most pathogenic form, appears more focal with apparent hotspots in Gansu, Ningxia, Xinjiang, Qinghai and Sichuan provinces/regions, some of which are also co-endemic for CE (Craig et al., 1992; Yang et al., 2006; Li et al., 2005; Han et al., 2006; Yu et al., 2008; Schantz et al., 2003; Zhou et al., 2000). Human CE and AE are co-endemic in only a few other world regions, notably eastern Turkey, Central Asia and Siberia (Craig, 2003). The current study now identifies a geographic area on the eastern edge of the Tibetan Plateau of approximately 313,200 km² involving at least 11 pastoral counties over 2 adjacent Provinces, with the greatest levels of co-endemicity of human CE and AE so far described worldwide.

In Tibetan pastoral communities, domestic dogs are kept in large numbers to guard property and livestock, and are usually tied during daytime and released at night. As Tibetan Buddhism forbids killing animals including dogs, with exception of food provision, large populations of stray dogs exist especially around temples. During the livestock slaughtering season (October to December), dogs (both owned and stray) are frequently fed with raw offal (including livers and lungs of yaks, sheep or goats) by herdsman. In addition, dogs may also prey on small mammals in around townships and adjacent pastures. A necropsy study of stray dogs in Ganze Prefecture revealed a 29.5% prevalence for *E. granulosus* and 11.5% for *E. multilocularis* (Qiu et al., 1989). A recent diagnostic purgation study in Shiqu County identified 8% of owned dogs infected with *E. granulosus* and 12% with *E. multilocularis* infection (Budke et al., 2005). The Tibetan fox (*Vulpes ferrilata*) appears to be the main sylvatic definitive host of both *E. multilocularis* and *E. shiquicus* in these pastoral areas (Qiu et al., 1995; Xiao et al., 2005), and ownership of fox skins was shown to be a risk factor for human AE (Wang et al., 2006). To date however, there is no evidence that human echinococcosis can be caused by *E. shiquicus* (Li et al., 2008). Yaks as well as sheep and goats appear to be key intermediate hosts for transmission of *E. granulosus* on the Tibetan Plateau (He et al., 2000). In addition, the high altitude grassland is abundant in small mammal communities, and up to 5 species (in the genera *Microtus*, *Cricetulus* and *Ochotona*) have so far been identified as possible key reservoir intermediate hosts of *E. multilocularis* (Giraudoux et al., 2006). The involvement of dogs as well as foxes in transmission of *E. multilocularis*, the diversity of small mammal potential hosts, landscape / habitat ecology of small mammals, together with poor hygiene and other risk behaviors (Li et al., 2005; Wang et al., 2006), seem to be major factors contributing to the high prevalence of both human cystic and alveolar echinococcosis in pastoral areas of the eastern Tibetan Plateau.

Landscape ecology suitable for transmission of *E. multilocularis* can vary over short distances (10 km) (Giraudoux et al., 2003). An interesting observation in the current study was the absence of AE cases detected in one pastoral township (Honglong in Yajiang County), where altitude (4168m), average number of dogs owned (1.2) and livestock ownership, was similar to other pastoral townships with high human AE prevalence (e.g. Yiniu Township in Shiqu County at 4200m and average 2.9 dogs owned per household). Subtle differences in landscape ecology could affect suitable habitat for potential small mammal hosts and their population dynamics which enable transmission of *E. multilocularis* (Giraudoux et al., 2003).

Although a human CE prevalence of 0.3% (mean age = 61.6 years) was found after ultrasound screening in farming areas, all cysts were confirmed by ultrasound to be involutive or inactive i.e. CE4 or CE5 types, which may indicate less recent transmission of *E. granulosus* in those non-pastoral areas. Furthermore, no human AE cases were detected by mass screening in such farming areas. By contrast, upland farming/agricultural communities have been identified as important AE foci in some other parts of China (Craig et al., 1992) as well as in Europe (Giraudoux et al., 2003). In mixed farming/pastoral Tibetan areas both CE and AE were detected during screening, but the prevalence was lower (0.2% for CE and 1.5% for AE), in comparison with the higher altitude pastoral regions. Traditionally, females in Tibetan communities are usually responsible for home chores, such as feeding dogs and collecting yak dung for fuel. Women may therefore have more opportunities to be exposed to environments contaminated by *Echinococcus* spp eggs, resulting in the significantly higher prevalence we observed in females. Older age groups of either sex, are at greater risk probably because they have more opportunities for exposure over time.

Portable ultrasound has been applied for community screening for abdominal echinococcosis in China since the early 1990s (Craig et al., 1992; Yang et al., 2006). Cases identified in remote rural areas have benefited from early diagnosis (Bartholomot et al., 2002; Li et al., 2005), which improves chances for better treatment and prognosis. Classification of ultrasound images of

CE based on the criteria proposed by WHO provided information about the stage of this disease as well as choice of treatment procedures (WHO, 2003). In the current study, patients with diagnosis of CE cysts that were classified on ultrasound image as CL(of a parasitic origin), CE1, CE2 or CE3, were considered to be active and growing and therefore recommended for either long-term oral albendazole therapy (6 months) with periodic follow-up, and/or for surgical treatment (endocystectomy or percutaneous approaches). Conversely, for patients with CE4 or CE5 hydatid cysts indicative of an involutive or inactive parasite, no positive therapy was considered necessary, rather a 'wait and watch' approach was adopted (Kern, 2003). For classification of alveolar echinococcosis cases the PNM system proposed by the WHO Informal Working Group for clinical settings (Kern et al., 2006) was applied. In addition, criteria for the classification of ultrasound images of alveolar echinococcosis was proposed by us for application in resource poor community settings (Li et al., 2004). In the current study, 31.4% of AE cases detected were categorized as AE3 (lesions measured >10cm), indicative of an advanced stage with poor prognosis, because of impossibility of radical removal of alveolar lesions and poor efficacy of albendazole on late stage lesions (Pawlowski et al., 2001; Kern et al., 2006). AE cases that exhibited a lesion with a central necrotic cavity was detected in a total of 109 AE patients classified as AE2 or AE3, however, the presence of central necrosis was more frequent in cases with AE3 lesions (62.6%, 62/99) compared to those with AE2 type lesions (33.1%, 47/142) ($\chi^2=20.45, p<0.01$). We also identified 4 cases that exhibited dual hepatic infection with both cystic and alveolar echinococcosis in the current study; an unusual and complicated clinical situation, and though rare, has been previously reported in western China (Wen et al., 1992; Yang et al., 2008).

In conclusion, a large co-endemic focus of human cystic and alveolar echinococcosis has been confirmed on the eastern Tibetan Plateau within Tibetan communities, primarily covering the border region of northwest Sichuan and southeast Qinghai, with an overall combined CE/AE prevalence of 6.3%. Prevalence was significantly higher in pastoral communities (8.1%) compared to that in semi-pastoral (1.6%) or farming communities (0.3%). Further active screening programmes and early surgical and/or medical interventions, particularly for alveolar echinococcosis, are imperative to reduce the public health impact of human echinococcosis in this remote resource-poor region of western China.

Finally, the initiation in 2006 of a national echinococcosis control programme (focused on dog dosing with praziquantel and active mass screening) which has so far covered 114 counties in western China, may succeed in interrupting transmission and reducing the public health impact of echinococcosis, but needs to be permanently implemented in these co-endemic areas of the eastern Tibetan Plateau.

Acknowledgments

The study was supported by Grant Number RO1 TW001565 from the Fogarty International Center of NIH. The content is solely the responsibility of the authors and doesn't necessarily represent the official views of the Fogarty International Center or the National Institutes of Health. The funders had no role in the study design, data collection and analysis, decision to publish, or preparation of the manuscript. This study was also supported in part by Sichuan Provincial Department of Health, China, a PhD split-site studentship (to TL) between the University of Salford, UK and SIPD/ Sichuan CDC, China, the Japan Society for the Promotion of Science (JSPS) to AI (17256002, 21256003) and the Japan-China Medical Exchange Program from JSPS to MN.

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Figure 1.

Study sites of mass screening programs for abdominal echinococcosis by B-ultrasound in Sichuan Province, China A: Localities of study sites. : townships with detection of AE cases; : townships with detection of CE cases; : townships with detection of both CE and AE cases; : townships without echinococcosis cases detected. B: China map with the locality of Beijing (capital), Sichuan Province, Qinghai Province and Tibetan Autonomous Region (TAR).

Figure 2.
Ultrasound images of different types of hepatic echinococcal lesions in patients detected in northwest Sichuan

Figure 3.

Hepatic ultrasound images of the patient with dual infection (both CE and AE) in northwest Sichuan Province

CE and AE lesions were marked by a circle, respectively

Figure 4.

Geographic distribution of 1312 human cases of echinococcosis post-operatively confirmed during 1992-2006 in Aba Army Hospital. Solid and open bars indicate AE and CE cases, respectively. The length of bars shows approximate proportion of each disease in the county.

* symbol shows the locality of Aba Army Hospital. Full name of each county is shown as follows: SQ=Shiqu, GZ=Ganzi, SD=Seda, BY=Baiyu, XL=Xinlong, LH=Luhuo, DF=Daofu, DB=Danba, RT=Rangtang, AB=Aba, REG=Ruoergai, MEK=Maerkang, HY=Hongyuan, HS=Heishui, LX=Lixian, XJ=Xiaojin, JC=Jinchuan, BM=Banma, JZ=Jiuzhi, GD=Gande, MQ=Maqin, DR=Dari, CD=Changdu.

Figure 5.
Prevalence of echinococcosis by age groups in northwest Sichuan Province

Table 1
Prevalence of human echinococcosis at township levels by ultrasound scanning in northwest Sichuan Province

County	Township	No. examined	CE (n) %	AE (n) %	Dual infection	Total %
Shiqu	Niga	475	4.00(19)	4.00(19)	0	8.00
	Mengsha	356	9.55(34)	7.02(25)	1	16.85
	Yiniu	631	3.33(21)	9.35(59)	0	12.68
	Anizha	381	5.51(21)	8.14(31)	1	13.91
	Xiazha	584	7.02(41)	4.79(28)	0	11.82
	Qiwu	349	6.88(24)	1.72(6)	0	8.60
	Derongma	108	10.19(11)	4.63(5)	0	14.81
	Mengyi	100	14.00(14)	7.00(7)	0	21.00
Ganzi	Hongqi	212	11.32(24)	5.66(12)	1	17.45
	Kalong	549	0.91(5)	3.83(21)	0	4.74
	Chalong	614	2.61(16)	6.35(39)	0	8.96
	Chaza	116	2.59(3)	2.59(3)	0	5.17
Seda	Dade	123	1.63(2)	3.25(4)	0	4.88
	Daze	310	2.58(8)	3.23(10)	1	6.13
	Niduo	229	3.93(9)	1.75(4)	0	5.68
	Seke	492	0.61(3)	1.42(7)	0	2.03
Baiyu	Maqiong	302	3.97(12)	1.66(5)	0	5.63
	Nata	271	1.48(4)	2.58(7)	0	4.06
	Acha	258	3.10(8)	0.78(2)	0	3.88
	Anzi	119	1.68(2)	2.52(3)	0	4.20
Rangtang	Gaduo	274	0(0)	1.82(5)	0	1.82
	Nannuda	145	0(0)	1.38(2)	0	1.38
	Rongnuda	162	0(0)	0.62(1)	0	0.62
	Zhongrangtang	94	1.06(1)	2.13(2)	0	3.19

County	Township	No.examined	CE (n) %	AE (n) %	Dual infection	Total %
Dege	Dagun	269	3.35(9)	0.37(1)	0	3.72
	Yading	198	7.07(14)	1.52(3)	0	8.59
	Axu	117	5.13(6)	0(0)	0	5.13
Yajiang	Honglong	610	2.30(14)	0(0)	0	2.30
	Waduo	584	0(0)	0(0)	0	0.00
Maerkang	Zhuokeji	571	0.88(5)	0(0)	0	0.88
Danba	Badi	583	0(0)	0(0)	0	0
Total		10186	3.24(330)	3.05(311)	4	6.33

Table 2

Ultrasonic classification of echinococcal lesions in 649 cases (including 4 patients with dual infection) in northwest Sichuan Province

Subtype of AE	No.cases	Proportion %	Subtype of CE	No.cases	Proportion %
AE1s	67	21.27	CL	9	2.69
AE1m	7	2.22	CE1	103	30.84
AE2s	75	23.81	CE2	98	29.34
AE2m	20	6.35	CE3	12	3.60
AE2f	47	14.92	CE4	80	23.95
AE3s	34	10.80	CE5	17	5.09
AE3m	3	0.95	Mix	15	4.49
AE3f	62	19.68			
Total	315	100.00	Total	334	100.00