

The effects of management of infection source of echinococcosis in Linzhi, Tibet autonomous region of China

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Abstract

Background Echinococcosis is highly endemic in western and northern China, Tibet Autonomous Region (TAR) is one of the most serious prevalent areas where the prevalence of echinococcosis in 2016 was 1.66% in humans and dog infection rate was 7.30%. Linzhi is located in southeastern part of TAR where the prevalence of echinococcosis in 2016 was 1.55% in humans and in dog infection rate was 7.28%. Dogs are the main infection source for the transmission of echinococcosis to humans. A control and prevention campaign based on dog management has been conducted in the past three years. This study is to evaluate the effects of dog management on infection rate of dogs.

Methods Data of dog population, registration and de-worming of 7 counties/district in Linzhi from 2017 to 2019 were collected. Domestic dog fecal samples were collected from each endemic town of 7 counties/district in Linzhi in 2019 to determine the infection of dogs using coproantigen ELISA. Data analysis was processed using SPSS statistics to compare dog infection rate between 2016 and 2019 by chi-square test, and maps were mapped using ArcGIS.

Results In Linzhi, domestic dog population decreased from 17407 in 2017, 16512 in 2018, to 12663 in 2019, while the registration rate increased from 75.92% in 2017, 95.48% in 2018, to 98.64% in 2019. Similarly, stray dog population decreased from 14336 in 2017, 13067 in 2018, to 11837 in 2019, while sheltered rate increased from 84.63% in 2017, 92.32% in 2018, to 96.63% in 2019. Dog de-worming frequency increased from 4 times per annum in 2017 to 12 times in 2019, indicating almost every dog was dewormed monthly. A total of 2715 dog fecal samples were collected for coproantigen ELISA assay. The dog infection rate was 2.84% (77/2715) in 2019, which was significantly lower than 7.28% (45/618) in 2016 ($P < 0.05$).

Conclusion Increased dog registration, decreased dog population and increased dog de-worming frequency contributed to significantly decreased dog infection rate in Linzhi, TAR. Control and prevention campaign based on dog management can significantly decrease dog infection with *Echinococcus* spp in echinococcosis endemic areas.

Background

Echinococcosis is a severe zoonotic disease caused by larval stage of the genus *Echinococcus*. Cystic echinococcosis (CE) caused by metacestode of *Echinococcus granulosus* and alveolar echinococcosis (AE) caused by metacestode of *Echinococcus multilocularis* are two forms of echinococcosis. CE has a global distribution while AE is mainly confined to the northern hemisphere [1,2]. *Echinococcus* spp. has a complex life cycle that involves two hosts. The main definitive hosts are dogs, which harbor adult worms in their small intestines. Humans and herbivores, particularly sheep, are intermediate hosts of this parasite. The intermediate hosts become infected by ingesting the eggs released in the feces of definitive hosts. Dogs, as the definitive hosts, are pivotal in the transmission of echinococcosis [1].

Echinococcosis was found to be highly endemic in the pasture areas of northwestern China where the prevalence of echinococcosis in human was 0.28% (estimated prevalence base on case detection rate in sample population), in livestock was 4.68% (10186/217774), and dog infection rate was 4.25% (4750/111832) [3,4]. Echinococcosis, threatening more than 50 million people in China, has been listed as a key parasitic disease and a national control and prevention campaign has launched by Chinese central government since 2007 [5,6]. Control and prevention of echinococcosis relies on dogs de-worming with praziquantel, safe livestock slaughtering conditions, and free patients screening and treatment.

The Tibet Autonomous Region (TAR) of China is located in the Qinghai-Tibet Plateau, which was reported as one of the most serious endemic regions of the world [3,4,7]. The prevalence of echinococcosis remains at a high level despite the fact that there have been control and prevention campaign launched over years. According to epidemiological survey of echinococcosis in TAR in 2016, the prevalence of echinococcosis in humans was 1.66% (estimated prevalence base on case detection rate in sample population), in livestock was 11.84% (249/2103), and dog infection rate was 7.30% (552/7564) [7]. The government of TAR has strengthened control and prevention campaign of echinococcosis with increased financial support and more technical guidance since 2017. More attention has been paid on dog management due to ubiquity of dogs in TAR. Various measures have been taken to strengthen management of infection sources of echinococcosis, including dog registration, reduction of dog population, and the most importantly dog de-worming monthly.

In order to evaluate the effects of dog management in the past three years in TAR, we collected data of dog registration, dog population, and dog de-worming from Linzhi, one of the seven prefectures in TAR. Dog feces from Linzhi were collected and coproantigen ELISA method was used to determine dog infection with *Echinococcus* spp. And the differences in dog infection between 2016 and 2019 were analyzed. The results of this work may help to understand the effectiveness of control and prevention campaign in Linzhi and benefit the other echinococcosis endemic areas.

Methods

Study area

Linzi is located in the southeastern part of TAR between latitudes 26°52'~30°40' N, and between longitudes 92°09'~98°47' E. It is in the middle and lower reaches of the YarlungZangbo River with an average altitude of 3100m, a total area of approximately 11.7 km², and an overall population of 231,000. Linzi has 1 administrative district and 6 counties, which are Bayi District, Gongbujiangda, Milin, Motou, Bomi, Chayu, and Lang County. Among these 7 counties/district, Bayi, Gongbujiangda, Bomi, and Chayu has were reported as co-endemic areas of both CE and AE while the other 3 counties were reported as CE endemic areas[4,7].

Data collection

Data of dog population, registration and de-worming were collected from annual prevention and control report of Linzi Center for Disease Control and Prevention, TAR.

Measures of dogs management

Dog registration

For the convenience of management, a file has been created for each domestic dog, including the owner's name, sex, age, and date of each de-worming.

Dog de-worming

De-worming monthly for domestic dog with praziquantel, and each de-worming date was recorded on dog registration files. Dogs weighing less than 5kg were given 50mg each time; dogs weighing 5-15kg were given 200mg each time; dogs weighing more than 15kg were given 400mg each time. Dog feces were buried in depth or burned after de-worming.

Reduction of dog population

Various measures have been taken to control dog population, including building shelters to contain stray dogs as many as possible, restricting domestic dogs to 2 individuals per household, leashing domestic dogs, etc. Data on domestic and stray dogs population were collected by local veterinarians.

Dog infection assay

1 village was randomly selected from each endemic town of 7 counties/district of Linzi, and 20 households with dogs were randomly selected from each selected village according to dogs registration files. Only 1 dog fecal sample was collected from each selected household. The collected fecal samples were frozen at -80°C for at least 72h to inactive any potential *Echinococcus* eggs. All dog fecal samples were tested for *Echinococcus* coproantigens by sandwich ELISA (Dog *Echinococcus* coproantigens ELISA kit, Combined, Shenzhen, China).

Statistical analysis

Data analysis was performed using SPSS Statistics version 21.0 (IBM, New York, USA), maps were mapped using ArcGIS version 10.1 (ESRI, Redlands, USA). Dog infections were expressed as percentages, Infection rates in 2016 and 2019 were compared using chi-square test, and the level of statistical significance was set at $P \leq 0.05$.

Results

Dog registration

As in Table 1, registration rate of domestic dog was increased respectively each year from 75.92% (13216/17407) in 2017, 95.48% (15766/16512) in 2018, to 98.64% (12491/12663) in 2019. Almost all the domestic dogs in each counties/district have been registered for management. And domestic dog population decreased from 17407 in 2017, 16512 in 2018, to 12663 in 2019. The results show that the dog management is strengthened gradually.

Table 1 Registration of domestic dogs in Linzi, TAR in 2017-2019

County/ District	2017			2018			2019		
	Domestic dog No.	Registered dog No.	Registration Rate(%)	Domestic dog No.	Registered dog No.	Registration Rate(%)	Domestic dog No.	Registered dog No.	Registration Rate(%)
Bayi	3759	2722	72.41	2820	2679	95.00	2669	2643	99.03
GongbuJiangda	3759	3759	100.00	3310	3310	100.00	2305	2259	98.00
Milin	3287	1446	43.99	2651	2545	96.00	2168	2147	99.03
Motuo	1193	453	37.97	1054	1011	95.92	401	397	99.00
Bomi	2863	2863	100.00	2771	2660	95.99	2661	2608	98.01
Chayu	1912	1339	70.03	2126	1781	83.77	2048	2028	99.02
Lang	634	634	100.00	1780	1780	100.00	411	409	99.51
Total	17407	13216	75.92	16512	15766	95.48	12663	12491	98.64

Dog de-worming frequency

In 2017, annual de-worming frequency was 4 times per annum, and the number increased to 11 in 2018 and 12 in 2019, which indicated that almost all domestic dogs had been de-wormed each month (Table 2).

Table 2 De-worming frequency of domestic dogs in Linzhi, TAR in 2017-2019

County /District	2017			2018			2019		
	Domestic dog No.	deworming doses	Annual deworming frequency	Domestic dog No.	deworming doses	Annual deworming frequency	Domestic dog No.	deworming doses	Annual deworming frequency
Bayi	3759	14460	4	2820	32698	12	2669	29853	11
GongbuJiangda	3759	13514	4	3310	37296	11	2305	26901	12
Milin	3287	14372	4	2651	30544	12	2168	25740	12
Motuo	1193	4162	3	1054	11389	11	401	4684	12
Bomi	2863	11452	4	2771	30356	11	2661	31474	12
Chayu	1912	7633	4	2126	23806	11	2048	24171	12
Lang	634	1759	3	1780	21286	12	411	4721	11
Total	17407	67352	4	16512	187375	11	12663	147544	12

Stray dog population

So far, 3 shelters have been established for stray dogs in Linzhi, TAR, and stray dog population has gradually decreased from 14336 in 2017, 13067 in 2018, to 11837 in 2019, while sheltered rate of stray dog was increased from 84.63% in 2017, 92.32% in 2018, to 96.63% in 2019. The results indicated that the vast majority of stray dogs have been sheltered by 2019 (Table 3).

Table 3 Stray dog population in Linzhi, TAR in 2017-2019

County/ District	2017			2018			2019		
	Stray dogs No.	Sheltered dog No.	Sheltered rate (%)	Stray dogs No.	Sheltered dog No.	Sheltered rate (%)	Stray dogs No.	Sheltered dog No.	Sheltered rate (%)
Bayi	3016	2684	88.99	2684	2518	93.82	2280	2166	95.00
GongbuJiangda	2914	2475	84.93	2486	2368	95.25	1751	1663	94.97
Milin	2064	1785	86.48	1954	1759	90.02	1780	1719	96.57
Motuo	1795	1468	81.78	1616	1437	88.92	1567	1536	98.02
Bomi	2715	2468	90.90	2684	2538	94.56	2986	2926	97.99
Chayu	986	684	69.37	917	846	92.26	855	825	96.49
Lang	846	568	67.14	726	597	82.23	618	603	97.57
Total	14336	12132	84.63	13067	12063	92.32	11837	11438	96.63

Dog infection

A total of 2715 fecal samples were collected from domestic dogs in all 7 counties/district in Linzhi, TAR. The infection rate with *Echinococcus* spp. among dogs was determined by the coproantigen ELISA. The dog infection rate in Linzhi was 2.84% (77/2715) in 2019, significantly lower than that in 2016, which was 7.28% (45/618) ($P=0.05$). At the county level, the highest dog infection rate was 3.78% (40/1058) in Bayi district, followed by 3.36% (26/774) in Lang county, and the lowest dog infection rate was 0% (0/200) in Chayu county in 2019. There was a significant

difference in dog infection between 2019 and 2016 in 2 county/district (Bayi district and Motuo county) ($P \leq 0.05$), and the other 5 counties showed no significant difference ($P > 0.05$) (Table 4).

Table 4 Dog infection in Linzhi, TAR in 2016 and 2019

County/ District	2016				2019				P-value
	Total dog No.	Dog fecal sample No.	Positive sample No.	Infection rate(%)	Total dog No.	Dog fecal sample No.	Positive sample No.	Infection rate(%)	
Bayi	6775	117	27	23.08	2783	1058	40	3.78	<0.05
GongbuJiangda	6673	81	4	4.94	2393	166	2	1.20	>0.05
Milin	5351	80	1	1.25	2229	192	5	2.60	>0.05
Motuo	2988	80	6	7.50	432	120	2	1.67	<0.05
Bomi	5578	80	1	1.25	2721	205	2	0.98	>0.05
Chayu	2898	100	3	3.00	2078	200	0	0.00	>0.05
Lang	1480	80	3	3.75	426	774	26	3.36	>0.05
Total	31743	618	45	7.28	13062	2715	77	2.84	<0.05

Discussion

The echinococcosis has been included in the list of 17 neglected tropical diseases by World Health Organization (WHO) [8]. China, specifically the Qinghai-Tibet Plateau region, has the highest endemicity of echinococcosis in the world [3,4,7]. The public health burden is large because the diseases are difficult to treat, requiring complex surgical procedures and long-term high dose antihelminthic treatment [9]. In addition the unique social-cultural background and lifestyle in Tibetan communities adds further difficulties to diseases control and prevention. Echinococcosis has been considered as a serious public health issue in China, especially in TAR.

Dog is the main definitive host for *E. granulosus* and a major host for *E. multilocularis* if infected by ingesting small mammalian with metacestodes infection [9-12]. Dogs are considered as the main risk to humans echinococcosis due to their close relationship with human [13-15]. Human become infected by ingesting the Echinococcus eggs released in the feces of dogs. Dogs are an important part of most Tibetan pastoral families and communities, which resulted in large numbers of dogs around, including domestic and stray dogs. Therefore, how to reduce dog population in environment and decrease dog infection are essential for echinococcosis control and prevention.

Domestic dogs in the Qinghai-Tibet Plateau, especially those of herdsmen, have a wide range of activities and can roaming on the grassland freely. The feces of infected domestic dogs are scattered around the herdsmen's living areas, which can easily lead to dog owners' infection. Thus management measures for domestic dogs include leashing domestic dogs to limit their roaming range and reduce their chance of infection, restricting domestic dogs to 2 individuals per household to control dog population and reduce the risk of transmission. And the most importantly, de-worming monthly. All domestic dogs should be registered, wear tags to distinguish them, and record each date of de-worming on the file, so as to ensure the monthly de-worming. Dog feces are buried in depth or burned after de-worming to cut off the transmission route. By 2019, the registration rate of domestic dogs in Linzhi was 98.64% (Table 1), the number of domestic dogs was gradually decreased (Table 1), and the annual deworming frequency reached 12 times (Table 2). These management measures were effective, the infection rate of dogs in Linzhi in 2019 was significantly lower than that in 2016.

Dogs that are not leashed, not wear tags and not registered are considered as stray dogs. Stray dogs usually have a larger range of activities than domestic dogs and are more likely to be infected by ingesting internal organs of infected livestock and small mammalian. In addition, the feces of infected stray dogs are scattered in the environment, so stray dogs are more important for the transmission. The most important management measure for stray dogs is to contain them as much as possible, and de-worming of sheltered stray dogs. Sheltered dog feces are buried in depth or burned after de-worming to reduce environmental contaminant. Table 3 show that by 2019, the vast majority (96.63%) of stray dogs have been contained in shelters and dewormed, there are only a few stray dogs in the environment, which greatly reduces the risk of echinococcosis transmission. Therefore, the infection rate of stray dogs was not detected in this study.

According to National plan for echinococcosis and other key parasitic diseases prevention (2016–2020) [6], all echinococcosis endemic counties were categorized as three classes according to their prevalence in humans and dog infection rate. Class I (prevalence in humans $\geq 1\%$ or dog infection rate $\geq 5\%$) is the most serious level. The epidemiological survey of echinococcosis in TAR in 2016 reported that the prevalence in humans in Linzhi was 1.55%, and the infection rate of dogs was 7.28% (45/618) [7]. As shown in Fig. 1, among 7 counties/district of Linzhi, there were 2 counties were categorized as class I (Red in Fig. 1A) and the other 5 counties were categorized as class II ($0.1\% \leq$ prevalence in humans $< 1\%$ or $1\% \leq$ dog infection rate $< 5\%$) (Orange in Fig. 1A) in 2016. After three years with effort on control and prevention, 2 counties in Linzhi were

down-categorized as class II, a total of 5 counties were categorized as class II (Orange in Fig.1B), and the other 2 counties were down-categorized as class III (0% prevalence in humans or 0% dog infection rate) (Yellow in Fig.1B) in 2019. The dog infection level of Linzhi in 2019 was significantly lower than that in 2016. This change illustrated that dog management can significantly decrease dog infection with *Echinococcus* spp.

Meanwhile, we noticed that although dog infection rate in Linzhi was significantly decreased, it still remained at a relatively high level. In some counties, dog registration, reduction of dog population and dog de-worming have been carried out better (from the data), the vast majority of dogs have been registered and de-wormed monthly by 2019, however, the dog infection still remained high, which seems to be a contradiction. The coproantigen ELISA method has been used to determine dog infection and may have cross reaction with other cestode infection resulting false positive [2,16], which may be one of the reasons why infection rate remained relatively high. In addition there may be some problems during implementation of dog de-worming because it is difficult to ensure that dogs take praziquantel successfully during the process, which may lead to ineffective de-worming.

Conclusions

This work described the changes of dog registration, dog population and dog de-worming frequency in Linzhi, TAR from 2017 to 2019, and compared dog infection between 2016 and 2019. The results illustrate that control and prevention based on dog management can effectively reduce dog infection in echinococcosis endemic areas.

Abbreviations

AE: alveolar echinococcosis; CE: cystic echinococcosis; ELISA: Enzyme-linked immunosorbent assay; TAR: Tibet Autonomous Region; WHO: World Health Organization.

Declarations

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Authors' contributions

YW, LYW, GQ designed the study; BCM, GQ, HSP collected data and samples; YW, BCM, HSP performed ELISA assays; LYW, GQ analyzed data; YW, LYW drafted the manuscript. All authors read and approved the final manuscript.

Availability of data and materials

All relevant data can be found within this paper.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Consent to publish was secured from the study participants.

Competing interests

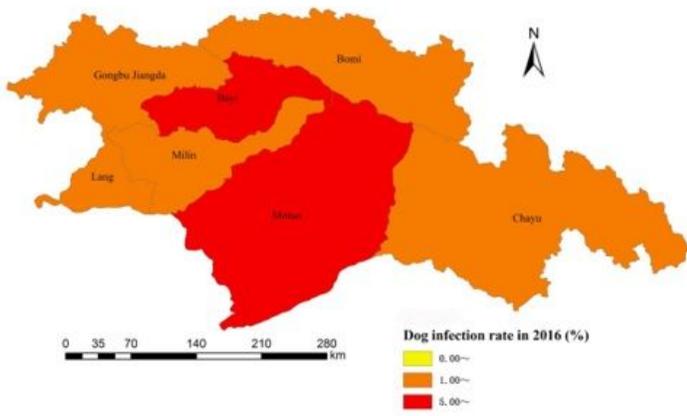
The authors declared no competing interests.

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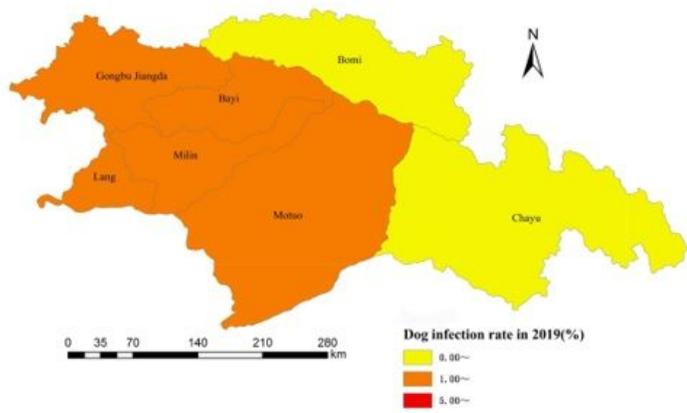
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Figures



A Dog infection distribution by county in Linzhi, TAR in 2016



B Dog infection distribution by county in Linzhi, TAR in 2019

Figure 1

Dog infection distribution by county in Linzhi, TAR